

**UNIVERSITAT POLITÈCNICA DE VALÈNCIA**

DEPARTAMENTO DE INGENIERÍA HIDRÁULICA Y MEDIO  
AMBIENTE

PROGRAMA DE DOCTORADO DE INGENIERÍA DEL AGUA Y  
MEDIOAMBIENTAL



PhD Thesis

**Water-oriented management in forest plantations: combining  
hydrology, dendrochronology and ecophysiology**

By

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Supervisor

Antonio D. Del Campo

Valencia, October 2014

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TESIS DOCTORAL

**Manejo orientado al agua en plantaciones forestales: combinando  
hidrología, dendrocronología y ecofisiología**

Presentada por *Tarcísio José Gualberto Fernandes*  
para optar al grado de doctor por la Universidad  
Politécnica de Valencia

Dirigida por:

Dr. Antonio D. Del Campo

Valencia, Octubre 2014

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## **NOTE TO READERS - PhD Thesis structure**

This thesis is structured as follows:

Previously to the presentation of the chapters, the reader will find the outline of the thesis and their objectives, including a diagram depicting the structure. Following, abstracts of the thesis written in English, Spanish, Valencian and Portuguese. **Chapter 1** an introduction setting the basis for the research, defining the basic science review, experimental research area, general material and methods and identifying the main questions addressed in the thesis are presented. Chapters 2, 3 and 4 are three papers published or sent to publication in different international journals. **Chapter 2** – *Coupling daily transpiration modelling with forest management in a semiarid pine plantation* (iForest Biogeosciences and Forestry – Accepted for publication); **Chapter 3** – *Hydrology-oriented (adaptive) silviculture in a semiarid pine plantation: how much can be modified the water cycle through forest management?* (European Journal of Forest Research – Published doi: 10.1007/s10342-014-0805-7) and **Chapter 4** – *Growth, tree water-use and water-use efficiency in response to proactive adaptive silviculture in a stagnated Mediterranean pine forest* (Manuscript). Finally in **Chapter 5**, a general discussion, a

summary of the key findings of the thesis and their implications, limitations, some recommendations and future research are presented.

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## **OUTLINE OF THIS THESIS AND THEIR OBJECTIVES**

Information on the water-use by forest is undoubtedly important and necessary (Ferraz et al. 2013), especially in water scarcity areas that are suffering the main negative impacts of climate change (Giorgi and Lionello 2008). The expected increase in the importance of water as a limiting factor for the future of many Mediterranean forests supports its inclusion as a priority in forest management. However, instead of just determining how much water is used by a forest, which is usually already well studied (see chapter one), it is also important to evaluate this water-use as a response of forest management practices. This need generates a pressing question: What forest management strategies can be established to promote a better water-use, keeping forest resilience traits to face climate change? In the literature consulted, thinning as a forest management practice is recognized as an alternative to promote improvements in the hydrologic balance, maintaining forest resilience. In order to contest this open question this thesis proposes three integrated studies (Fig. 1) performed in an experimental area of Aleppo pine subject to thinning (see Materials and Methods for a detailed description of experimental area used in this thesis).

Transpiration is recognized as a key factor by forest managers to calculate hydrological balance. Forest transpiration can be modelled using different approaches. Nevertheless, many of them require homogeneous forest coverage, or an individual calibration/validation process for each cover stand. Artificial Neural Network (ANN) may be a method capable of overcoming this problem by including forest cover heterogeneity, produced by forest management practices, as an input variable. Thus, the first study, presented on chapter two of this thesis, was modelling an ANN to estimate daily transpiration independently of cover heterogeneity. Then the ANN modelled was used for gap-filling when needed and those results were used in the following studies (chapter three and four).

The secondly study, presented in chapter three, addressed the question of how the water balance and the tree-growth changed as a consequence of thinning. To this end, the influence of thinning intensity and its effect on the short-term (thinned in 2008) and mid-term (thinned in 1998) on the water-balance components and growth (Basal Area Increment, BAI) were investigated. Two main aspects are dealt with in this chapter, firstly the influence of thinning intensity and secondly the effect of the elapsed time since thinning on the changes in the

distribution of water fluxes. These distributions allowed the calculation of water balance in each plot studied. Furthermore, the use of dendrochronology as a tool in hydrologic studies, specifically in the sap flow velocity correlation, showed an interesting approach for a better understanding of tree transpiration after thinning.

The third study, presented in the chapter four, has three interesting points concerning thinning, water-use and water-use efficiency. First, the relationships between growth and climate were studied at mid-term in order to identify if thinning can improve forest resilience to face climate changes. Second, growth, Oxygen isotope ( $\delta^{18}\text{O}$ ) and intrinsic water-use efficiency (WUEi) at short and mid-term were studied in order to explore the influence of thinning in these physiological attributes. Third, the relationships between water-use and intrinsic water-use efficiency was explored to identify how these factors were affected by thinning at short-term.

In the final chapter the main results are discussed, showing a summary of the key findings and their implications and limitations, and some recommendations to future research are advanced.

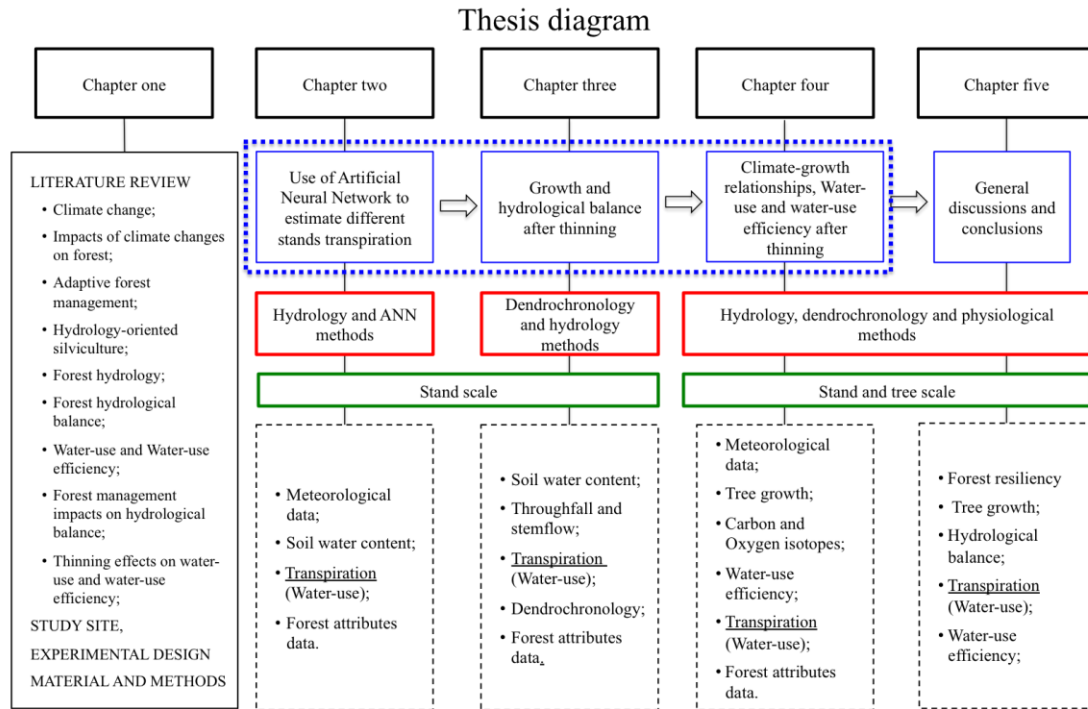


Fig. 1 – PhD thesis diagram. Blue box represents the main aspects of each chapter; Red boxes the methodology used; Green boxes the scale studied and areas within black dotted line represent the main data used, except chapter five where key words found in discussion and conclusions are presented. Underlined word represents the transversality of this thesis.

## **NOVEL ASPECTS OF THIS THESIS**

This dissertation has certain aspects that can be considered as novel within the context of hydrology-oriented silviculture studies in Mediterranean areas, they are:

- 1) In this thesis we report the use of Artificial Neural Networks to estimate stand transpiration coupled with forest management (Chapter two). To our knowledge our paper is the first to do so.
- 2) This work encompasses a wide array of inter-related variables affecting the hydrological balance; i.e. soil water content, water-use, intrinsic water-use efficiency and growth as a function of thinning intensity and the effects of the elapsed time since thinning. Few if any publications deal with this subject in Aleppo pine in Mediterranean areas. (Chapters three and four);
- 3) Here we have analysed thinning responses of Aleppo pine to climate change observed in the last fifteen years in Mediterranean areas by means of a dendroclimatology approach. There are but a handful of similar observations on the earliest manifestations of climate change in the Mediterranean forests (Chapter four);

4) In this thesis I have evaluated together water-use and water-use efficiency after thinning in Aleppo pine, while also assessing the effects of biotic and abiotic factors. This combined approach has shown to be a powerful research tool. I am not aware of any other study that contrasts WU measured, by sap flow sensors, with WUE<sub>i</sub> values obtained by isotope analysis, subject to different thinning intensities (Chapter four);



## **FUNDING**

The research leading to this thesis was conducted in the Department of Water and Environmental Engineering at the Universitat Politècnica de València, in collaboration with the Federal University of Acre, with scholarship support of Mundus 17 coordinated by Porto University, and CAPES (Coordination for the Improvement of Higher Education Personnel) Ministry of Education in Brazil, with indirect support (wife scholarship).

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Thank you!

## **ABSTRACT**

Assessment of forest water-use (WU) is undoubtedly important and necessary, especially in water scarcity areas that are already suffering the main negative impacts of climate change. However, instead of just determining how much water is used by a forest, it is also important to evaluate how forest-WU responds to forest management practices such as thinning, a widely recognized alternative to promote improvements in the hydrologic balance while maintaining or improving forest resilience. Thus, this thesis proposes three integrated studies performed in an area of Aleppo pine subject to experimental thinning in Eastern Spain. The first study was modelling an artificial neural network (ANN) to estimate daily WU independently of forest heterogeneity provided by thinning. Stand WU was accurately estimated using climate data, soil water content and forest cover (correlation coefficient, R: 0.95; Nash-Sutcliffe coefficient, E: 0.90 and root-mean-square error, RMSE: 0.078mm/day). Then the ANN modelled was used for gap-filling when needed and those results were used in the following studies. The secondly study addressed the question of how tree-growth, WU and water balance changed as a consequence of thinning. To this end, the influence of thinning intensity and its effect at short-term (thinned in 2008) and at mid-term (thinned in

1998) on the water-balance components and tree-growth were investigated. The high-intensity thinning treatment showed significant increases in mean annual tree-growth from 4.1 to 17.3 cm<sup>2</sup> yr<sup>-1</sup>, a rate which was maintained in the mid-term. Mean daily WU ranged from 5 (control) to 18 (high intensity thinning) l tree<sup>-1</sup>. However, when expressed on stand basis, daily WU ranged from 0.18 (medium intensity thinning) to 0.30 mm (control plot), meaning that in spite of the higher WU rates in the remaining trees, stand WU was reduced with thinning. Large differences were found in the water balance components between thinning plots and control. These differences might have significant implications to maintain forest resilience, and improve forest management practices. The third study, brings forth two interesting points and their responses to thinning, WU and intrinsic water-use efficiency (WUEi). First, the relationships between growth and climate were studied at mid-term in order to identify if thinning can improve forest resilience. Second, the relationships between WU and WUEi was explored to identify how these factors were affected by thinning at short-term. A substantial limitation of tree-growth imposed by climatic conditions was observed, although thinning changed the tree-growth-precipitation relationships. Significant differences in WUEi were found

after thinning at mid-term, however no significant difference was observed at short-term. Despite this, in general  $WUE_i$  decreased when precipitation increased, with different slopes for each thinning intensity. Different patterns of the relationship between  $WU$  and  $WUE_i$  were found, being positive for thinned plots and negative for control plot at short-term. Finally this thesis suggest that thinning in Aleppo pine plantations is effective in changing the relationships between  $WU$  and  $WUE_i$ , furthermore, this thesis introduces a novel contribution by looking at the inter-related effects on growth,  $WU$ ,  $WUE_i$  and water balance in Mediterranean forest subject to thinning.

## RESUMEN

La evaluación del uso del agua (UA) de los bosques es sin duda, importante y necesaria, especialmente en áreas con manifiesta escasez de agua y que ya están sufriendo con los principales impactos negativos del cambio climático global. No basta sin embargo sólo determinar la cantidad de agua utilizada por un bosque, también es importante evaluar cómo los bosques responden a prácticas de manejo forestal, tales como el clareo, que es ampliamente reconocida como una alternativa para promover mejoras en el equilibrio hidrológico, manteniendo y o mejorando a la vez la resiliencia de los bosques. En esta tesis se proponen tres estudios integrados realizados en una zona de pino carrasco (*Pinus halepensis* Mil.) en el este de España, sometida a clareos experimentales. El primer estudio fue la modelación de una red neuronal artificial (RNA) para estimar el UA diario, independientemente de la heterogeneidad del bosque como consecuencia del clareo. La UA se estimó con precisión a partir de datos climáticos, del contenido de agua del suelo y de la cubierta forestal (coeficiente de correlación,  $R=0,95$ ; coeficiente de Nash-Sutcliffe,  $E= 0,90$  y el Error de la raíz cuadrada de la media,  $RMSE= 0,078\text{mm/día}$ ). A continuación, la RNA modelada se utilizó para rellenar datos faltantes, cuando fuera necesario, y estos

(datos completos) se utilizaron en los siguientes estudios. El segundo estudio abordó la cuestión de cómo el crecimiento de los árboles, el UA y el balance hídrico cambian como consecuencia del clareo. Con este fin, fueron investigados la influencia de la intensidad del clareo y su efecto a corto-plazo (clareado en 2008) y a mediano-plazo (clareado en 1998) en el crecimiento de los árboles y en los diferentes componentes del balance hídrico. Los árboles del clareo de alta intensidad mostraron aumentos significativos en el promedio de la tasa de crecimiento anual de 4,1 a 17,3 cm<sup>2</sup> año<sup>-1</sup>, una tasa que se mantuvo en el mediano plazo. El promedio diario de UA por árbol osciló entre 5 (parcela control) a 18 litros (clareo de alta intensidad). Sin embargo, cuando se expresa a nivel de parcela el UA diario osciló entre 0,18 (clareo de intensidad media) a 0,30 mm (parcela control), lo que significa que a pesar de las mayores tasas de UA en los árboles remanentes, el UA a nivel de parcela se redujo con el clareo. Se encontraron asimismo grandes diferencias en los diferentes componentes del balance hídrico entre las parcelas clareadas y el control. Estas diferencias pueden tener importantes implicaciones para mantener la resiliencia de los bosques mediterráneos y mejorar las prácticas de manejo forestal en áreas que pueden sufrir con la escasez de agua. El tercer estudio, ilustra dos aspectos interesantes con respecto al



UA y la eficiencia intrínseca del uso del agua (EUAi) así como sus respuestas al clareo. En primer lugar, se estudiaron las relaciones entre el crecimiento de los árboles y el clima con el fin de identificar si el clareo puede mejorar la capacidad de recuperación de los bosques. En segundo lugar, las relaciones entre UA y EUAi fue explorada para determinar cómo estos dos factores se vieron afectados por el clareo a corto plazo. Se observó una limitación sustancial del crecimiento de los árboles impuesta por las condiciones climáticas en la área estudiada, además fue posible observar que el clareo cambió las correlaciones de la precipitación con el crecimiento de los árboles. Se observaron diferencias significativas en la EUAi después del clareo a medio-plazo, sin embargo no se observó diferencia significativa en el corto-plazo. A pesar de esto, en general la EUAi se reduce cuando la precipitación aumenta mostrando diferentes pendientes en esta relación para cada intensidad de clareo. Se encontraron diferentes patrones de la relación entre UA y EUAi, siendo positivo en las parcelas clareadas y negativo en la parcela control a corto-plazo. Por último, los resultados expresados en esta tesis sugieren que el clareo en las plantaciones de pino carrasco es eficaz en cambiar las relaciones entre UA y EUAi. En esta tesis se introduce una novedosa

contribución relativa a los efectos interrelacionados del crecimiento, UA, EUAi y del balance de agua en el bosque mediterráneo sujeto al clareo.

## RESUM

L'avaluació de l'ús de l'aigua (UA) dels boscos és sens dubte, important i necessària, especialment en àrees amb manifesta escassetat d'aigua i que ja estan patint els principals impactes negatius del canvi climàtic global. No es suficient determinar la quantitat d'aigua utilitzada per un bosc, també és important avaluar com els boscos responen a pràctiques de maneig forestal, com ara l'aclarida, que és àmpliament reconeguda com una alternativa per a promoure millores en l'equilibri hidrològic, mantenint i millorant al mateix temps la resiliència dels boscos. En esta tesi es proposen tres estudis integrats realitzats en una zona de pi blanc (*Pinus halepensis* Mil.) sotmesa a aclarides experimentals a l'est d'Espanya,. El primer estudi es basaria en la modelació d'una xarxa neuronal artificial (RNA) per a estimar l'UA diari, independentment de l'heterogeneïtat del bosc com a conseqüència de l'aclarida. La UA es va estimar amb precisió a partir de dades climàtiques, del contingut d'aigua del sòl i de la coberta forestal (coeficient de correlació,  $R=0,95$ ; coeficient de Nash-Sutcliffe,  $E= 0,90$  i l'Error de l'arrel quadrada de la mitjana,  $RMSE= 0,078\text{mm/día}$ ). A continuació, la RNA modelada es va utilitzar per a omplir la manca de dades, quan fóra necessari, utilitzant aquestes dades completes en els següents estudis. El segon estudi va

abordar la qüestió de com el creixement dels arbres, l'UA i el balanç hídric canvien com a conseqüència de l'aclarida. Amb aquesta finalitat, van ser investigats la influència de la intensitat de l'aclarida i el seu efecte a curt termini (clarejat en 2008) i a mig termini (clarejat en 1998) en el creixement dels arbres i en els diferents components del balanç hídric. Els arbres de l'aclarida d'alta intensitat van mostrar augments significatius en la mitjana de la taxa de creixement anual de 4,1 a 17,3  $\text{cm}^2 \text{ año}^{-1}$ , una taxa que es va mantindre en el mig termini. La mitjana diària d'UA per arbre va oscil·lar entre 5 (parcel·la control) a 18 litres (aclarida d'alta intensitat) . No obstant això, quan s'expressa a nivell de parcel·la l'UA diari va oscil·lar entre 0,18 (aclarida d'intensitat mitjana) a 0,30 mm (parcel·la control), la qual cosa significa que a pesar de les majors taxes d'UA en els arbres romanents, l'UA a nivell de parcel·la es va reduir amb l'aclarida. Es van trobar així mateix grans diferències en els diferents components del balanç hídric entre les parcel·les clarejades i el control. Estes diferències poden tindre importants implicacions per a mantindre la resiliència dels boscos mediterranis i millorar les pràctiques de maneig forestal en àrees que poden patir amb l'escassetat d'aigua. El tercer estudi, il·lustra dos aspectes interessants respecte a l'UA i l'eficiència intrínseca de l'ús de l'aigua (EUAi) així com les seues

respostes a l'aclarida. En primer lloc, es van estudiar les relacions entre el creixement dels arbres i el clima, a fi d'identificar si l'aclarida pot millorar la capacitat de recuperació dels boscos. En segon lloc, les relacions entre UA i EUAi va ser explorada per a determinar com estos dos factors es van veure afectats per l'aclarida a curt termini. Es va observar una limitació substancial del creixement dels arbres imposada per les condicions climàtiques en l'àrea estudiada, a més va ser possible observar que l'aclarida va canviar les correlacions de la precipitació amb el creixement dels arbres. Es van observar diferències significatives en l'EUAi després de l'aclarida a mig termini, no obstant això no es va observar cap diferència significativa en el curt termini. A pesar d'açò, en general l'EUAi es redueix quan la precipitació augmenta, mostrant diferents pendents en esta relació per a cada intensitat d'aclarida. Es van trobar diferents patrons de relació entre UA i EUAi, sent positiu en les parcel·les clarejades i negatiu en la parcel·la controlada a curt termini. Finalment, els resultats expressats en esta tesi suggereixen que l'aclarida en les plantacions de pi blanc és eficaç per a canviar les relacions entre UA i EUAi. En esta tesi s'introdueix una nova contribució relativa als efectes interrelacionats del creixement, UA, EUAi i del balanç d'aigua en el bosc mediterrani subjecte a l'aclarida.

## RESUMO

A avaliação do uso de água (UA) pelas florestas é, sem dúvida, importante e necessária, especialmente em áreas que apresentam escassez de água e que já estão sofrendo os impactos negativos das mudanças climáticas globais. No entanto, ao invés de apenas determinar o quanto de água é usado por uma floresta também é importante avaliar como a floresta responde às práticas de manejo florestal, como por exemplo o desbaste que é reconhecido como uma alternativa viável para promover a melhoria do balanço hidrológico mantendo a resiliência da floresta. Neste sentido, esta tese propõe três estudos científicos integrados que foram realizados em uma área desbastada experimentalmente de *Pinus halepensis* Mil. localizada ao leste da Espanha. O primeiro estudo consiste na modelagem de uma rede neural artificial (RNA) para estimar o uso de água (transpiração) diária independentemente da heterogeneidade da floresta produzida pelo desbaste. O uso de água pela floresta remanescente após o desbaste foi estimado com precisão, utilizando dados climáticos, umidade do solo e a cobertura florestal em porcentagem, apresentando coeficiente de correlação, R: 0,95; coeficiente de Nash-Sutcliffe, E: 0,90 e erro quadrático médio, RMSE: 0,078 milímetros/dia. Em seguida, a RNA

modelada foi utilizada para o preenchimento de dados faltantes e estes resultados foram utilizados nos seguintes estudos. O Segundo estudo aborda a questão de como o crescimento das árvores, o uso de água e o balanço hídrico na floresta é alterado em consequência do desbaste. Para este fim, foram investigadas a influência da intensidade de desbaste e seu efeito no curto prazo (parcelas desbastadas em 2008) e no médio prazo (parcela desbastada em 1998) sobre o crescimento das árvores remanescentes e nos diferentes componentes do balanço hídrico. As árvores remanescentes mostraram aumentos significativos na taxa média de crescimento anual de 4,1 a 17,3 cm<sup>2</sup>/ano. Taxa essa que se manteve a médio prazo. A média diária de uso de água por árvores oscilou entre 5 (parcela controle) a 18 litros (desbaste de alta intensidade). No entanto, quando se expressa a nível de parcela o uso de água diário variou entre 0,18 (desbaste de media intensidade) a 0,30 mm (parcela controle), o que significa dizer que apesar das maiores taxas de uso de água ocorrer nas árvores remanescentes, o uso de água a nível de parcela reduziu com o desbaste. Ademais, grandes diferenças foram encontradas nos diferentes componentes do balanço hídrico entre parcelas desbastadas e a parcela controle. Estas diferenças têm significantes implicações para manter a resiliência florestal, bem como, melhorar as práticas de manejo voltadas

à água em áreas que podem sofrer com a escassez de água. **O terceiro** estudo ilustra dois aspectos interessantes relacionados com o uso de água e o uso eficiente de água bem como as suas respostas ao desbaste. Em primeiro lugar, foi estudado as relações entre o crescimento das árvores remanescentes e o clima, a médio prazo, com o fim de identificar se o desbaste pode melhorar a resiliência das florestas frente às mudanças climáticas. Segundo, as relações entre uso de água e uso eficiente de água foram analisadas com o objetivo de identificar como estes fatores foram afetados, a curto prazo, pelo desbaste. Neste terceiro estudo, identificou-se uma limitação substancial no crescimento das árvores impostas pelas condições climáticas, apesar do desbaste mudar a relação existente entre crescimento e precipitação. Diferenças significativas no uso eficiente de água foram observadas após o desbaste a médio prazo, entretanto nenhuma diferença significativa foi observada a curto prazo. Apesar disso, em geral o uso eficiente de água diminui quando há maior precipitação, com diferenças para cada intensidade de desbaste. A curto prazo foram encontradas diferentes padrões na relação entre uso de água e uso eficiente de água, sendo positivo para as parcelas desbastadas e negativo para a parcela controle. Finalmente, esta tese sugere que o desbaste em plantações de *Pinus halepensis* Mill. é eficaz para mudar as



relações existentes entre uso de água e uso eficiente de água, além disso, esta tese apresenta uma nova contribuição para os efeitos inter-relacionados no crescimento, uso de água, uso eficiente de água e balanço hídrico em florestas mediterrâneas submetidas ao desbaste.

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## **CHAPTER ONE**

**Basic science review, experimental research area and general material and methods**

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## **Basic Review**

### **Climate Change**

The Framework Convention on Climate Change (UNFCCC), in its Article 1, defines climate change as: *“a change of climate which is attributed directly or indirectly to human activity that alters the composition of the global atmosphere and which is in addition to natural climate variability observed over comparable time periods”*.

The scientific consensus about climate change is clearly expressed in the reports of the Intergovernmental Panel on Climate Change (IPCC). Created in 1988 by the World Meteorological Organization and the United Nations Environmental Programme. The IPCC's main purpose is to evaluate the state of climate science as a basis for informed policy action, primarily on the basis of peer-reviewed and published scientific literature (Oreskes, 2004).

The Fifth Assessment Report of the Intergovernmental Panel on Climate Change, published in 2013, is the latest update of the activities of the IPCC, compiling and synthesizing the studies by thousands of scientists around the world about global warming (see: IPCC, (2014) in: <http://www.climatechange2013.org>). These are their main findings:

- Global warming is unequivocal. Combining land and ocean temperature, data shows an increase of about 0.89°C over the period 1901–2012 and about 0.72°C over the period 1951–2012;
- The major cause of global warming is, with very high degree of certainty<sup>1</sup>, the emission of gases by human activities. Evidences indicating the human origin of the problem have become more robust since the previous IPCC, (2007) report;
- Ocean warming dominates the total heating rate, serving as a buffer to atmosphere warming. However this warming decreases the ability to absorb carbon dioxide – a major greenhouse gas – and which can accelerate atmospheric effects when it reaches saturation;
- Anthropogenic forcings<sup>2</sup> have made a substantial contribution to upper-ocean warming (above 700m) observed since the 1970s. This anthropogenic ocean warming plus glacier mass loss has contributed to the global sea level rise observed (1.7 mm year<sup>-1</sup> between

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<sup>1</sup> The degree of certainty in key findings of the Fifth Assessment Report of the Intergovernmental Panel on Climate Change is based on the author teams' evaluations of underlying scientific understanding and is expressed as a qualitative level of confidence (from very low to very high) and, when possible, probabilistically with a quantified likelihood (from exceptionally unlikely to virtually certain).

<sup>2</sup> Forcing due to human, rather than natural, factors. Such factors include increased greenhouse gas concentrations associated with fossil fuel burning, sulphate aerosols produced as an industrial by-product, human-induced changes in land surface properties among other things.

1901 and 2010. However between 1993 and 2010, the rate has reached 3.2 mm year<sup>-1</sup>);

Changes of average precipitation in a much warmer world will not be uniform, with some regions experiencing increases, and others with decreases or not much change at all. The high latitudes are likely to experience greater amounts of precipitation due to the additional water-carrying capacity of the warmer troposphere. Many mid-latitude arid and semi-arid regions will likely experience less precipitation.

All of these possible effects combined can generate new causes, tending to amplify the consequences of climate change. Moreover, an intensification of the water cycle, may determine a change in many components of the hydrological cycle such as: precipitation patterns, intensity and extremes; atmospheric water vapour; evapotranspiration; soil moisture and runoff (Zollo et al., 2012), especially in areas with a dry and semi-arid Mediterranean climate (Vargas-Amelin and Pindado, 2013).

## **Climate change in Mediterranean Areas**

Over the last decade, a number of studies have assessed how climate change may affect the Mediterranean region (Christensen et al., 2007; Parry et al., 2007; Giorgi and Lionello, 2008; Estrela et al., 2012). Scientists are confident that observed global warming will be accompanied by significant changes in Mediterranean climate, although considerable uncertainty remains. This is primarily because of the acknowledged weaknesses of global climate models (GCMs) in assessing regional climate changes at an appropriate scale. Nevertheless long-term climatic series have demonstrated a significant increase in heat waves and drought impacts over the last several decades (Andreu et al., 2007)

Annual mean temperatures in Mediterranean areas will rise more than the global average and the warming is likely to be larger in summer (Christensen et al. 2007), as a consequence these areas are subject to a slow but steady desertification process, particularly in the south and the east of the Mediterranean (Matteucci et al., 2011). In the Mediterranean area many models, including different scenarios of climate change, foresee summers characterized by an increase in frequency, of extreme daily events of precipitation despite a general



decrease in average precipitation (Giorgi and Lionello, 2008; Estrela et al., 2012; Parry et al., 2007). Thus this tendency can lead to longer dry periods, increasing the risks of droughts, interrupted by extreme intense precipitation events, enhancing the risks of floods and erosion.

Spain is considered one of the most vulnerable countries to climate change within the European Union, due to its geographic and socio-economic characteristics (Vargas-Amelin and Pindado, 2013), with a territory of 506 000 km<sup>2</sup>, has a clear imbalance of water availability between the wetter north, and dryer central and south-eastern areas. The mean annual precipitation is approximately 670 mm/year, varying from 2200 mm in the north of the country to 120 mm in the southeast (Estrela et al., 2012). Fig. 1 shows the mean precipitation of peninsular Spain for the period 1940–2010. A detailed study about the impacts of climate change on water resources in Spain can be found in Estrela et al. (2012). These authors report that the impact of climate change on hydrological design and water resources management could be one of the most important challenges faced by hydrologists and water managers.

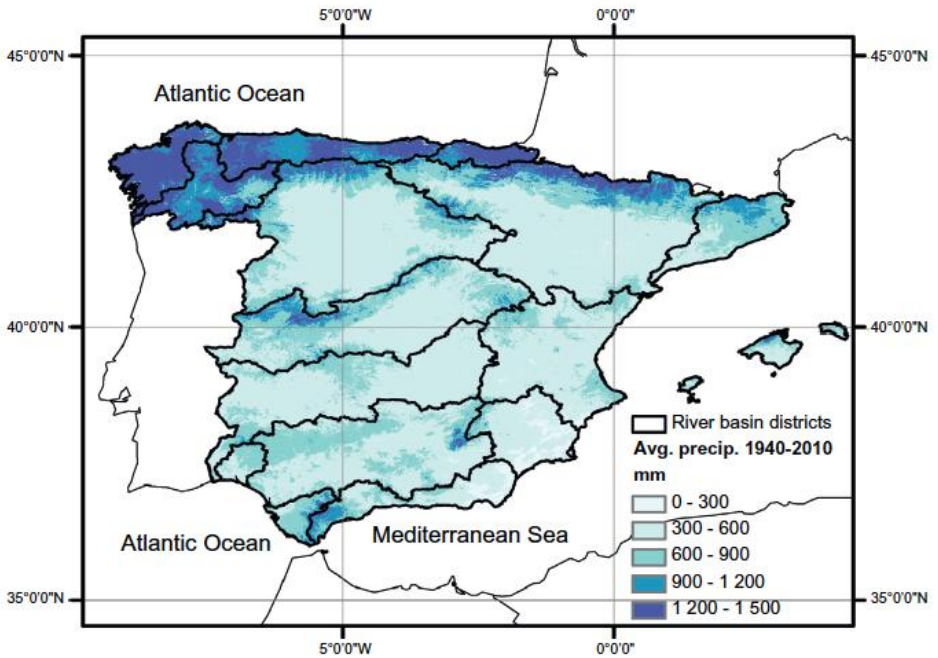


Fig. 1 - Map of average precipitation for the period 1940–2010. River Basin Districts are shown inside of the boundaries of Spain. Source: (Vargas-Amelin and Pindado, 2013).

Considering that climate change is irreversible<sup>3</sup>, and that areas under water scarcity, such as the Mediterranean Spain, can experiment negative effects, as mentioned above, special attention should be given to forests, especially due to the role they play in the hydrological cycle.

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<sup>3</sup> A change is said to be irreversible if the recovery timescale from this state due to natural processes is significantly longer than the time it takes for the system to reach this perturbed state. Such behaviour may arise because the timescales for perturbations and recovery processes are different, or because climate change may persist due to the long residence time of a CO<sub>2</sub> perturbation in the atmosphere.

## **Impacts of climate change on forests**

The role of forest dynamics in the global climatic system is likely to be long-lasting, complex, and difficult to predict. Because of their longevity and because adaptive measures, such as replacement of species, are harder to implement than in agricultural systems, forests may be particularly vulnerable to climatic change (Houghton, 1996). There are many possible impacts of climate change on forests, but these will differ according to location, past history, vegetation type, management activities and a range of other factors (Innes et al., 2009). However, many studies report that forests are particularly sensitive to climate change, because the long life-span of trees does not allow for rapid adaptation to environmental changes. Chmura et al., (2011) consider that warming may be favourable to forest growth at high elevation sites where the growing season is currently limited by low temperatures or snow cover, but would enhance the effects of drought in areas that are currently moisture-limited, as many areas of Mediterranean Spain. Hence, the amount of water available to trees will depend on the amount and timing of precipitation, as well as the amounts of surface runoff, deep drainage, and evaporation.

Across Europe climate change is expected to have quite different impacts on forest ecosystems varying with e.g. latitude, altitude, local ecological conditions and forest management practices (Jacobsen et al., 2013). The two most outstanding recent efforts in this theme in Europe are developed in ForeStClim (“Transnational Forestry Management Strategies in Response to Regional Climate Change Impacts”, an EU-funded environmental project addressing forests and climate change, ended in 2012; see: [www.forestclim.eu](http://www.forestclim.eu)), and MOdels for adapTIVE forest Management, MOTIVE (that represents the state of the art in science-based information on adaptation of European forests to climate change, completed in 2013; see: <http://motive-project.net/>). Both projects are motivated by increasing evidences of climate change impacts, and the need to identify adequate forest management practices to face these impacts. In Spain, the main efforts are reported in the National Climate Change Adaptation Plan (PNACC<sup>4</sup> in Spanish). Within the PNACC, extensive impacts and vulnerability assessments have already

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<sup>4</sup> The PNACC is the reference framework for the coordination of the Public Administrations efforts in dealing with the assessment of impacts, vulnerability, and adaptation options to address the impacts of climate change in a series of sectors and natural resources acknowledged as potentially affected.

been carried out in Spanish forests, especially the reports by (Gracia et al., 2005; Hierro et al., 2011).

Studies of climate change impacts on forests are dependent on model assumptions (Loehle, 2000), and have been the target of much criticism. However there is a consensus that the effects of climate change on forests can reduce growth and survival, predispose forests to disturbance by wildfire, insects, and diseases; and eventually change forest structure and composition at the landscape scale (Chmura et al., 2011). Despite this, Vayreda et al. (2012) affirm that warming had almost no effect on growth change in the drier areas of Peninsular Spain, where tree species are, presumably, more adapted to long dry periods. This is in disagreement with the study of Lindner, (2000) which report that small changes in climate may lead to relatively strong shifts on growth. Moreover many studies carried out in different Mediterranean areas have documented episodes of forest decline<sup>5</sup> and mortality induced by water stress and/or high temperatures (Sánchez-Salguero, 2012 and references therein). A large number of episodes of forest mortality

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<sup>5</sup> Manion and Lachance, (1992) described forest decline as an episodic event characterized by premature, progressive loss of tree and stand vigor and health over a given period without obvious evidence of a single clearly identifiable causal factor such as physical disturbance or attack by an aggressive disease or insect.

associated with drought and heat stress have been detected worldwide in recent decades, suggesting that some of the world's forested ecosystems may be already responding to adverse changes in climate (Martínez-Vilalta et al., 2012). Carnicer et al., (2011) reported increased crown defoliation associated with recent droughts, as well as widespread increases in standing tree mortality between 1989–1996 and 1997–2007 in Iberian forests of Spain. These authors show that Iberian forests are experiencing long-term chronic effects due to severe climate change-related droughts, and that these effects are progressively more pronounced in more xeric localities. Vayreda et al., (2012) in a study about how climate changes interact with stand structure and Management in Spanish forests, affirm that the patterns of mortality (both mortality occurrence and mortality rate) also showed a significant relationship with water availability. Moreover they assert that water availability is a good indicator of favourable conditions for growth. Furthermore, warmer temperatures will increase Mediterranean evapotranspiration, especially in summer and hasten seasonal depletion of soil moisture. Thus, dry summers in Mediterranean Spain may become even drier, which is consistent with recent observations (Sarris et al., 2007; Sarris et al., 2011; Linares et al., 2011). Hence, the most significant challenge facing

Mediterranean forests is an increase in the frequency, duration, and intensity of droughts, as discussed above.

Genetic and silvicultural approaches to increase adaptive capacities and to decrease climate-related vulnerabilities of forests should be based on ecophysiological knowledge. Effective approaches to climate adaptation will likely include assisted migration of species and populations, and density management (Chmura et al., 2011). However Sjölund and Jump, (2013) warn that adaptive strategies should be carefully considered to improve forest resilience<sup>6</sup>. Furthermore the adaptive capacity in the forest sector is limited in the Mediterranean region where large forest areas are only extensively managed or unmanaged (Lindner et al., 2010). At present, only limited practical guidelines are available to managers about how to address climate change, and the following uncertainties about the specific conditions that will prevail at local and regional levels, are huge (Crow, 2012).

In the light of MOTIVE project, Jacobsen et al., (2013) propose two approaches to forest managers for treating uncertainties facing the expected climate change impacts: 1) reactive approach and 2) proactive

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<sup>6</sup> Resilience is the capacity of an ecosystem to maintain or regain normal development following disturbance (Adams et al., 1994).

approach. The reactive manager can be described as a decision maker who awaits and observes the actual outcomes and impacts of climate change as it develops, and adjusts management gradually in response to observed effects only. The proactive management approach is in the literature also referred to as the fully adaptive management approach. As opposed to the former, the decision maker applying this approach does not only observe the development of the current climate and the state of the forest ecosystems; the manager also assesses and foresees likely developments and impacts of climate change using different sources of information and observations.

As related above, and in the available literature, the negative impacts of climate change in Mediterranean forest are unquestionable. In what follows we deal only with the proactive approach i.e. adaptive forest management, judging this as a better way to adapt Mediterranean areas to climate change (Fitzgerald et al., 2013).

### **Adaptive Forest Management**

Adaptive management of natural resources was developed in the 1970's by C.S. Holling and co-workers at the International Institute for Applied Systems Analysis (see: Holling, 1978) and it is steadily gaining



wider acceptance by forest managers. According to Nyberg, (1998), *adaptive management is a systematic process for continually improving management policies and practices by learning from the outcomes of operational programs. Its most effective form— “active” adaptive management—employs management programs that are designed to experimentally compare selected policies or practices, by evaluating alternative hypotheses about the system being managed.* The idea of Adaptive Management was applied to forest resources, creating the concept of Adaptive Forest Management (AFM). In the literature there are many definitions of AFM, such as those of Taylor et al., (1997); Walter, (1997); Bormann et al., (1999); MacDonald et al., (1999); Stankey et al., (2003); Bunnell et al. (2007); Innes et al., (2009); Williams, (2011); von Detten, (2011); Yousefpour et al. (2013), among others. In the Dictionary of Forestry (Helms, 1998) AFM is defined as *"a dynamic approach to forest management in which the effects of treatments and decisions are continually monitored and used, along with research results, to modify management on a continuing basis to ensure that objectives are being met."*

AFM has become especially popular and has gained new attention in current climate change debates (von Detten 2011).

Nevertheless, an important component of any adaptive forest management, to face the new perspective of climate change, should anticipate potential problems and devise strategies for avoiding, minimizing or overcoming them. However the forest ecosystems are complex and dynamic, and effective strategies should be continuously modified. Yousefpour et al. (2013) report that the implementation of adaptive forest management in response to climate change requires that forest managers have access to accurate information regarding the direction and magnitude of climate change, and an accurate assessment of how the system will respond to the climate drivers. Innes et al., (2009) consider that adaptive forest management is a tool that could enable managers to adjust the structure and the consequent functioning of the forest ecosystem to resist harmful impacts of climate change, and to utilize the opportunities created by those changes. Moreover, despite the intuitive appeal of adaptive management as a strategy to face climate change, it has not been translated easily into the practice of forest planning (Hoogstra-Klein and Burger, 2013), thus it is necessary to modify traditional forest management strategies (Lindner, 2000).

In this sense, it would be possible to consider an effective form of adaptive forest management combining monitoring, research and the

means to change traditional silvicultural practices to reach novel management proposals. Although these may be feasible and comprehensible in principle, the long time frames required to gather information from experiments may not match the shorter time frames required for decision-making. In addition, when results do become available from long-term experiments on topics such as for example water-use, water-use efficiency and growth, they may no longer be relevant due to continuous climate changes, especially in Mediterranean areas. Among forest adaptive management options, when it comes to Mediterranean areas, it is important to highlight the adaptive management that especially takes into account water resources, in this work called hydrology-oriented silviculture (HOS), this adaptive strategy is considered: *“a set of forest management techniques that uses scientific information to formulate strategies in order to optimize the forest water balance, to face the new challenges of global climate change”*.

### **Hydrology-oriented silviculture**

Hydrology-oriented silviculture (HOS) is a novel concept, despite the fact that the main idea is not recent. In the literature consulted, HOS is usually referred to as water-saving, however this concept is more

general and can be applied also for agriculture, while HOS is restricted to forestry.

It is clear that forests can provide a range of economic, social and environmental benefits. However, in many situations dense forests as those from afforestation can also decrease surface water generation and groundwater recharge (see: (Webb et al., (2012) and references therein). This is of concern if forest expansion occurs where water resources are already under pressure (van Dijk and Keenan, 2007). Hence, special attention should be given to the forest–water relationships in the Mediterranean, where large areas of forest are unmanaged and agricultural and pasture areas that were abandoned and eventually turned into “forested” areas, which could be consuming more water. For example, Willaarts (2012), in a study about water-use in Spanish forests reported that forests consume larger proportion of annual rainfall in Spain than agriculture, and this highlights the crucial role they play in the general water balance. This author affirms that Spanish forests evapotranspire 39% of the annual incoming rainfall, although during droughts this ratio can rise up to 42% and that these results provide evidence of the importance of placing forest and land management at the core of hydrological planning, especially in the context of climate

change. Studying the effects of forest management on the water cycle should, thus, become a priority for policy makers because adaptive forest management policies in Mediterranean ecosystems should have a strong hydrological foundation (Planinsek et al., 2010; Molina and del Campo, 2012; Del Campo et al., 2014).

According to Robledo and Forner, (2005) the relationship between forest ecosystems and the water sector is two-ways. On the one hand, given that forest play an important role as water cycle regulators, while deforestation and degradation of forests might increase the vulnerability of the water sector. Thus, silvicultural treatments should also be adapted to hydrological resources. Traditional forest management models usually design optimal stand densities to maximize timber production through thinning practices. Hence the former need to be re-evaluated from a “water-saving” point of view. In this sense, it is important to understand the components of forest hydrology and how silvicultural practices can affect them. Following this introduction we present a brief review of forest hydrology, its components, and a description of the impact of forest management in forest hydrology, with emphasis on thinning.

## **Forest Hydrology**

The International Symposium on Forest Hydrology in 1965 was the first forum where researchers from experimental watersheds from all over the world came together, exchanged viewpoints, and presented significant results on forest-soil-water relationships and forest watershed behaviour (McGuire and Likens, 2011). The concept of forest hydrology has since then been widely used and has become a forestry discipline with greater emphasis in those regions where water resources are suffering fluctuations due to climate change, such as in Mediterranean areas.

From the point of view of the hydrologists, *Forest hydrology* is that specialized part of hydrological sciences that emphasizes the influence of trees and forests and their management on the regimen, quality and quantity of water (DeWalle, 2003). However, from the point of view of foresters, *Forest hydrology* addresses the hydrologic processes within forested areas and the output of water resources from them (Barten et al., 2008). Combining these two points of view, we can say that *Forest hydrology is a science that allows analysis of forest and water connections to make predictions about forests and water that can*

*address current and anticipated future issues, including climate change and forest management practices.*

In the last decade, considerable progress has been made in forest hydrological research all over the world and new opportunities are emerging to apply the results to solving forest management issues related to water resources. Forest scientists and managers know that forest management strategies should lead to preservation of hydrological flows, mitigation of extreme hydrological events, retention of soils and sediments, support productivity and biodiversity, as well as maintenance and purification of water supply (Creed et al., 2011; Landsberg and Sands, 2011). Despite these effects, both (forest scientists and forest managers) have focused on one topic that is particularly relevant for forest hydrology; that is, the comparative advantages and disadvantages of forest cover in optimizing water balance while preserving or even improving forest resilience. As reported in many studies (Landsberg and Gower, 1997; Farley et al., 2005; Chang, 2006; Almeida et al., 2007; Buytaert et al., 2007; Pichler et al., 2011; Willaarts, 2012; Chen et al., 2014), tree canopies reduce water availability, through interception and transpiration from the foliage. Hence, there is no question that forest

removal, even if it is only partial, changes the forest hydrological balance (Calder, 2007).

### **Forest hydrological balance**

The main components of forest hydrological balance are: Interception, Throughfall, Stemflow, Runoff, Drainage, Soil evaporation and Transpiration. These components and their interplay are illustrated in Fig. 2 and summarized in the equations 1 and 2. In this topic some components will be considered in detail, while others components, which were not directly dealt with in this thesis, will be only mentioned.

$$Ppt = I + E + Tr + S + R + D \quad (1)$$

$$I = Ppt - Th - St \quad (2)$$

where:

$Ppt$  is precipitation (rain, snow, or fog);  $I$  is Intercepted water that is evaporated from canopy;  $E$  is soil evaporation;  $Tr$  is transpiration by trees and understory vegetation;  $S$  is soil water storage;  $R$  is runoff;  $D$  is drainage below root zone,  $Th$  is Throughfall and  $St$  is Stemflow.



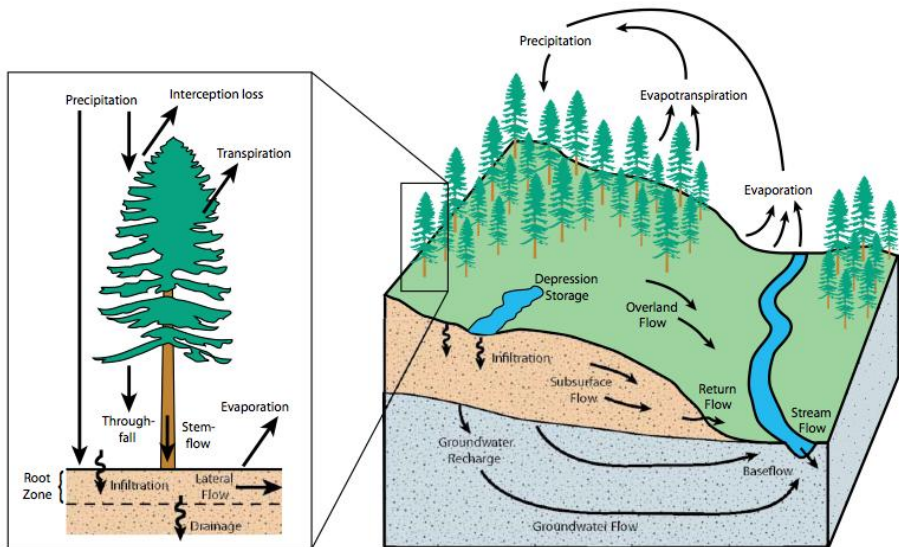


Fig. 2 - Diagrammatic representation for the main components of the forest hydrologic balance (see: Equation 1 and 2) (Source: Winkler et al., 2010)

## Precipitation

Precipitation is the input in the forest hydrological balance, and in any region it is usually described in terms of the annual mean expressed in millimetres of rain or meters of snow. However, precipitation is enormously variable, both spatially and temporally. The seasonal distribution of precipitation can influence the composition, structure, and function of terrestrial ecosystems (Landsberg and Gower, 1997). Another important input in the system, usually not reported, is the

fog (Bruijnzeel et al., 2006), being an important source of water in many coastal regions of the world, which are characterized by arid, semi-arid or Mediterranean types of climate (Dawson and Vidiella 1998). Fog is simply a cloud of minute water droplets that exists at ground level, developing when the air at ground level is cooled enough to cause saturation. When fog is intercepted by vegetation it is considered as additional water input (Gordon et al., 1994).

Vegetation affects precipitation patterns by mediating moisture, energy and trace-gas fluxes between the surface and atmosphere (Bonan, 2008; Matteucci et al., 2011;.Sheil and Murdiyarso, 2009) claimed that the replacement of the natural forest cover by other land cover might lead to an important reduction in the mean continental precipitation. However, in the last decades scientists have debated whether vegetation increases precipitation. Despite being a well established fact that plants put moisture back in the air through their leaves by a process known as evapotranspiration, but the quantity and geographical reach of precipitation generated by large forests remains unclear (Spracklen et al., 2012).

## **Interception, Throughfall and Stemflow**

Interception is that part of the rain that falls on the vegetation and evaporates without reaching the ground (Klaassen et al., 1998). Forest interception is an important event in the hydrologic cycle because of its effects on rainfall deposition, soil moisture distribution, snow accumulation and snowmelt, wind movement, heat dissipation (with impact/reduction on transpiration rate), and impact energy of raindrops on soil erosion (Chang, 2006). It is of particular importance in semi-arid and arid regions where soil moisture is a limiting factor influencing plant productivity, and where runoff and groundwater recharge are essential for meeting agricultural, industrial, and residential water demands (Carlyle-Moses 2004).

In forests the interception of precipitation by the canopy and its loss through evaporation is typically ca 20% of incident precipitation (Wang et al., 2007). Despite this, there are many studies with a wider range of values. For example Dunkerley and Booth, (1999) found 1.1-5.8% for shrubs in an arid region of Australia. Carlyle-Moses et al., (2010) found 9 to 24% in five tropical plantation species. (Waring and Schlesinger, 1985) report values between 10–40%. Llorens et al., (1997)

in NE Spain found variations between 15% of incoming precipitation for low intensity, long rain events with high air humidity, and up to 49% in intermediate duration and intensity rain events and under low ambient humidity. Pérez et al., (2014) studying interception losses in Southern Spain, found that *Pinus pinea* and *Cistus ladanifer* intercepted 26.86% and 16.36% of the total precipitation, respectively. Putuhena and Cordery, (2000) found in a mature eucalypt forest interception in the range 19–25%, and in *Pinus radiata* stand (13-16 year old) about 38%. Oyarzún et al., (1985) found 18% in *Pinus radiata* with 80% of forest cover and about 9% in pasture-forest with cover between 37 to 65%. Mazza et al., (2011) found average interception losses of 23% and 40% in thinned and unthinned areas of *Pinus pinea* respectively. In coniferous plantations this range usually is 55-80%, while broadleaves is 40-64% (Nisbet, 2005), in *Pinus halepensis* Mill. Maestre et al., (2003) report that the interception value usually is up to 50% of the annual rainfall amount, and that rain events smaller than 10 mm are completely or almost completely intercepted. However, Yaseef et al., (2010) found 11% interception in an Aleppo pine plantation, while Del Campo et al. (2014) in an experimental thinning in Aleppo pine, found values between 12.4 and 39.6%, the latter value corresponding to the control plot.

All these variations reported above depend strongly on the timing and intensity of rainfall, the vegetation structure and the meteorological conditions controlling evaporation during and after rainfall (Muzylo et al., 2009). Therefore, to calculate accurately the interception losses from a forest requires knowledge of the canopy storage capacity (Landsberg and Gower, 1997), which is defined as the maximum possible water storage after quick drainage has stopped (Klaassen et al. 1998). However forest interception assessment, has traditionally been based on the difference between the total of precipitation within in an adjacent open area and the sum of throughfall and stemflow in the forest (Putuhena and Cordery 2000; Huber and Iroumé, 2001; Dunkerley, 2010).

Throughfall is considered the amount of water that cannot be intercepted by tree canopy and that reaches the understory, while stemflow is considered the amount of water that is intercepted by stems and branches and flows down the tree trunk into the soil (Davie, 2008). In forest hydrology the sum of both is commonly called net precipitation (Muzylo et al., 2009; Winkler et al., 2010). Throughfall and stemflow, by modifying the spatial distribution of the water reaching the soil, have significant hydrological impacts, especially in areas with important

periods of water scarcity (Llorens et al., 2011). For dense canopies throughfall may be very low until the canopy saturates. In most cases some water will reach the ground after the start of a rain event and before the canopy is completely saturated because of drip and (possibly) stemflow. The amount will depend on tree architecture and precipitation intensity (Landsberg and Sands, 2011). Stemflow acts like a “funnel” collecting water from a large area of canopy but delivering it to the soil in a much smaller area: the surface of the trunk at the base of a tree (Davie, 2008), although reaching lower fractions of incoming rainfall, is very significant because of its effects on soil water recharge, surface flow, erosion and especially in the heterogeneity of spatial distribution of soil water (Levia Jr. and Frost, 2003).

Forest management practices, which alter canopy structure, can alter interception, throughfall and stemflow values. A large number of studies have identified the influence of thinning on precipitation redistribution, usually expressed as the statistically significant differences in these flows between thinned and unthinned areas. For example Molina and Del Campo, (2012) found that throughfall increased significantly after thinning to 83.8%, 67.7% and 61.3% of the bulk rainfall for high, moderate and low intensity treatments, respectively.

Despite this, these authors reported that the absence of significant differences found between low intensity thinning and the control plot indicates that a minor silvicultural intervention (in their case, a reduction of 26.2% in basal area) is not enough to increase the water reaching the soil surface via throughfall. Mazza et al., (2011) found an increase of 11.7% of net precipitation (throughfall+stemflow) in the thinned area compared to control area. However, stemflow measurements were very low, comprising only 0.20% of the total precipitation in the thinned area and 0.27% in the control. Slodicak et al., (2011) in their study of thinning in Scots pine, found that thinned increasing throughfall in 2–8% compared to control plot and this increase persisted for six years after thinning, being significantly different for the first four years.

In summary, many studies report different values of the effects of thinning on forest interception, thus a better understanding of these effects is needed, especially considering that the amount of water that reaches the soil (throughfall+stemflow) is one of the main component of hydrology-oriented silviculture in the Mediterranean areas.

## **Soil water content**

Soil infiltration rate and water storage capacity are important hydrological parameters reflecting the soil and water conservation function of forests (Bens et al., 2007). The amount of water to a given depth of the soil volume at any specific time, depends on the balance between water input by net precipitation and losses through runoff, drainage, soil evaporation, transpiration by trees and the understory vegetation integrated over time (Landsberg and Sands, 2011). Despite this, changes in soil water content depends on the initial water content and the rate of infiltration relative to the rate of drainage out of the root zone, which in turn depends on soil hydraulic properties, the depth of the soil, and root penetration (Landsberg and Gower, 1997). Thus, it is clear that the infiltration rate and storage capacity of the soil on the forest floor are of paramount importance (Keller, 1992).

Forests with soils of good structure and which are rich in organic matter have relatively high water-retention capacity. However, the influence of the forest ceases as soon as the soil is saturated<sup>7</sup> (Fig. 3).

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<sup>7</sup> Soil is saturated when it has reached its maximum water content; if any more is added, it will either drain downward or a surface flux occurs.



In addition to this, dynamic soil water movement also can be altered by tree density. David et al., (2007) suggest that the groundwater lifted by the roots can be released into the upper soil layers before it is reabsorbed by shallow roots and finally transpired by trees. On the other hand, the reverse sap flow phenomena also can occur, i.e. the movement of water from tree to the soil (Buckley et al., 2011). Thus, in a moist soil the rate of water absorption is controlled primarily by two factors 1) the rate of transpiration and 2) the efficiency of root systems as absorbing surfaces. When the soil dries the availability of water begins to be limited by decreasing water potential and soil hydraulic conductivity. Soil aeration, soil temperature, and the concentration and composition of the soil solution also may sometimes limit absorption of water (Pallardy, 2008a).

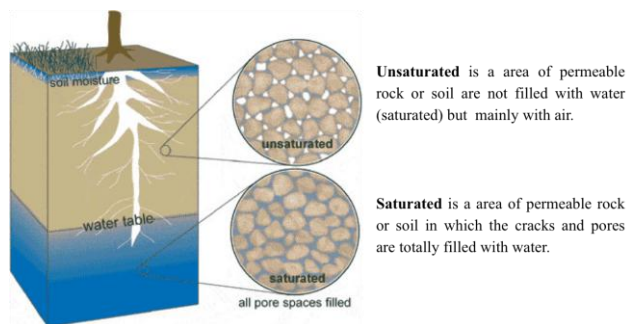


Fig. 3 - Definition an example of unsaturated and saturated soil. Adapted from: (<http://quizlet.com/4706106/water-saturation-and-weathering-flash-cards/>)

In the last decade several review papers summarize a large number of studies on the effects of various forest management practices on forest floor and soil (Swank et al., 1989; Johnson, 1992; Vesterdal et al., 1995; Thibodeau et al., 2000; Johnson and Curtis, 2001; Johnson et al., 2002; Bens et al. 2007; Hoover, 2011; Dighton et al., 2012; Persson, 2012; Chen et al., 2014). For example, the increase of soil water availability after thinning has been related to reduced soil water stress, as a consequence of a reduction of both canopy interception and forest transpiration (Swank et al., 1989; Bréda et al., 1995). Chen et al. (2014) in their study about effects of thinning intensities on soil infiltration and water storage capacity in a Chinese Pine-Oak mixed forest, report that some soil properties changed drastically after thinning, while pH showed no change under different thinning intensities. These authors, also reports that the soil infiltration rate and water storage capacity firstly increased with the increase of stand density but then decreased after arriving at a certain degree. Thus, a better understanding of thinning effects on forest soil is helpful to provide more strategies for hydrology-oriented silviculture in Mediterranean areas.

## **Evaporation**

Micrometeorologists and hydrologists often combine evaporation and transpiration into one measurement, largely because the methods used to determine evapotranspiration (ET) do not distinguish between the two fluxes, yet the two processes are quite different (Bond et al., 2008). A number of methodologies have been developed to measure ET or its components (i.e., transpiration and evaporation) across a spectrum of spatial scales (Lin et al., 2010; Wang and Dickinson, 2012).

Jasechko et al. (2013) posit that the biological pump of water into the atmosphere during photosynthetic gas exchange (that is, transpiration) dominates water losses from the continents, representing 80% to 90% of terrestrial evapotranspiration. This is especially so because plant roots are able to tap into groundwater and deeper soil-water reservoirs, transpiration effectively moves deep sources of water into the atmosphere, whereas evaporation is only effective for water at or near the surface, which explains the very high proportion of transpiration to the overall evapotranspiration flux. However, ET is closely linked with vegetation characteristics and environmental constraints (Allen et al., 1998). In arid and semiarid regions, evapotranspiration is often nearly

equal to precipitation, while in humid areas, it is limited by available energy (Zhang et al., 2001). In forest ecosystems, evapotranspiration can represent over 60% of the precipitation (Table 1) (Schlesinger and Jasechko, 2014), thus ET (i.e., transpiration and evaporation) can be considered as a key parameter in hydrological forest studies. Furthermore, evaporation can be divided into two major components, canopy evaporation (after rain) and soil evaporation. The rate of evaporation of water from wet canopies depends mainly on the atmospheric vapour pressure deficit, and on wind speed, and is not controlled by physiological tree processes, while soil evaporation initially proceeds at a high rate approximating that of open water and is determined by the energy available to vaporise water at the soil surface (Landsberg and Sands, 2011). These two components of evaporation are difficult to measure separately in forest ecosystems (Schlesinger and Jasechko, 2014) and are generally calculated as the difference between the total input (rain) and other easily measured components.

**Table 1 - Some studies of water balance. Ppt is precipitation. Number between brackets refers to total number of studies.**

<b>Ecoregion</b>	Precipitation (mm)	Transpiration (% of Ppt)	Evaporation (% of Ppt)	Runoff and Recharge/Drainage (% of Ppt)
Agricultural	998.6 (5)	44.0 (3)	55.7 (3)	
Boreal Forest	578.7 (6)	45.2 (6)	22.4 (5)	12.0 (1)
Desert	252.9 (16)	41.2 (10)	52.5 (11)	
Mediterranean Shrubland	456.3 (4)	43.0 (4)	48.0 (4)	7.0 (2)
Steppe	189.7 (3)	44.7 (3)	48.3 (3)	5.0 (2)
Temperate Deciduous Forests	798.7 (10)	48.8 (9)	26.0 (8)	30.0 (2)
Temperate Forest	1258.8 (13)	36.3 (13)	27.5 (13)	53.0 (6)
Temperate Grassland	478.4 (9)	49.9 (7)	59.8 (6)	0.0 (1)
Tropical Grassland	1543.8 (5)	56.4 (5)	37.4 (5)	
Tropical Rainforest	2207.4 (8)	47.3 (8)	20.5 (8)	31.8 (4)
Tundra	310.0 (1)	34.0 (1)	8.0 (1)	58.0 (1)
<b>Average</b>	<b>824.8 (80)</b>	<b>44.6 (69)</b>	<b>36.9 (67)</b>	<b>24.6 (19)</b>

Adapted from: Schlesinger and Jasechko (2014)

The amount of water evaporated from forests ecosystems depends on the amount of moisture available in the soil, which in turn is largely a function of soil texture, depth, and, of course, climate (Anderson et al., 1976). Thus, changes in the amount and (or) type of vegetation caused by fires, insects, disease, harvesting, and silvicultural practices, can alter evaporation from the forest (i.e., evaporation or sublimation of intercepted precipitation and evaporation from the soil surface) (Bonan 2008). Hence, thinning, by removing only part of the forest canopy can alter evaporation values. Hibbert, (1983) report that, if

water is not a limiting factor, the relationship between thinning and decreases in evaporation is generally considered as linear. On the other hand, canopy openings by thinning also might allow for increased evaporation from the soil and consequently an increased in transpiration rate from understory vegetation (Knoche, 2005). Thus, understanding how changes in forest cover affect this hydrological component is critical to comprehend the effects of silvicultural practices on forest water balance.

## **Transpiration**

As observed in Table 1, there are wide ranges of transpiration values among different ecosystems. Plant transpiration accounts for 60–80% of evapotranspiration on land, thus, there is no doubt that terrestrial vegetation is a dominant force in the global water cycle. Furthermore, transpiration is a major determinant of local microclimate and rainfall (Schlesinger and Jasechko, 2014). Plant transpiration refers to water losses from the soil to the atmosphere after uptake by the roots (Asbjornsen et al., 2011) and it is a dominant factor in plant-water relationships (Pallardy, 2008a). The ability of roots to supply water for plant transpiration depends on the hydraulic conductance of the root

system (determined by fine-root conductivity and total fine-root surface area), the distribution of roots within the soil profile along with the ability to produce new roots dynamically as soil water is used and replenished, and soil water availability throughout the rooting zone (Bond et al., 2008).

The first quantitative measurements of water loss (transpiration) from plants appear to have been made by Stephen Hales, prior to 1727. He measured water loss by weighing potted grapevines, apple and lemon trees, and various herbaceous plants. Several studies were made during the second half of the nineteenth century, of which the best known are those by von Höhnel, published in 1881 and 1884 (Pallardy, 2008b).

Models of transpiration have been developed for many species with a wide variation of physiological detail (see: Dekker et al., 2000 for a basic review). According to Bond et al. (2008) the Penman–Monteith equation (Monteith, 1965) is widely accepted as the definitive mechanistic description of relationships between vegetation properties and environmental drivers that influence transpiration, and is arguably one of the most important contributions of its time to vegetation science. However, the difficulty involved in accurate estimation of the conductance terms in the Penman–Monteith equation is often

overlooked, even though estimates of transpiration for forests are sensitive to these terms due to the strong coupling between stomatal conductance and transpiration (Bond et al., 2008). In this sense a more robust approach is needed (Asbjornsen et al., 2011).

Transpiration rates for whole plants or individual branches can be determined by techniques that measure the rate at which sap ascends by the xylem. All of these methods use heat as a tracer for sap movement (sap flow), but they are fundamentally different in their operating principles (Smith and Allen, 1996). Three sap flow techniques have been widely used to measure tree transpiration in forests: heat pulse velocity, tissue heat balance, and radial flowmeters (Wang and Dickinson, 2012). (Granier, 1985; 1987) was a pioneer in the use the heat dissipation procedure, which is based on measuring the heat transferred by the flow of sap between two probes inserted into the xylem, one of which is heated. In the last years a heat ratio method proposed by Burgess et al., (2001) was developed providing increased accuracy at low and reverse flow rates. Čermák et al., (2004) presented in chronological order the main methods developed for sap flow measurements. However the most significant developments in measure tree transpiration rate within the last 25 years have come about as a result of technological advances in sap



flow sensors. These sensors can measure water movement in the xylem accurately and over extended periods (Lagergren et al., 2008; Asbjornsen et al., 2011). More recently Dynamax Inc. (<http://www.dynamax.com/SapFlow.htm>) offers stem heat balance sensor models for different stem sizes and has recently introduced a new micro-sensor to facilitate sap flow measurements in thin stems with diameters ranging between 2.1 and 5.0 mm (Langensiepen et al., 2014). Then, the best way to determine the transpiration rate of a stand is to measure the sap flow of every tree in a plot large enough to be unaffected by edge effects. This ideal is rarely achieved because of cost and logistic considerations, and so forest hydrologists must rely on scaling up estimates of water use obtained on a limited number of representative trees (Wullschleger et al., 1998; Čermák et al., 2004). However, Davis et al., (2012) presented a detailed step by step to self-made sap flow sensors, which can reduce significantly the cost (98% of total).

In summary the measurement of stem sap flow is a widely used indirect approach for estimating tree transpiration (Sun et al., 2012), and the use of sap flow sensors to quantify water use of individual trees (Fig. 4b) is relatively common (Zeppel et al., 2008), allowing extrapolation to the overall forest stand after a simple scaling-up exercise (Aranda et al.,

2012) (Fig 4c), where a common practice is to multiply the individual value of transpiration by total sapwood area (Buckley et al., 2011; Buckley et al., 2012), by number of trees (Schiller and Cohen, 1998; Schiller, 2011; Ungar et al., 2013) or by frequency of tree diameter (Del Campo et al. 2014).

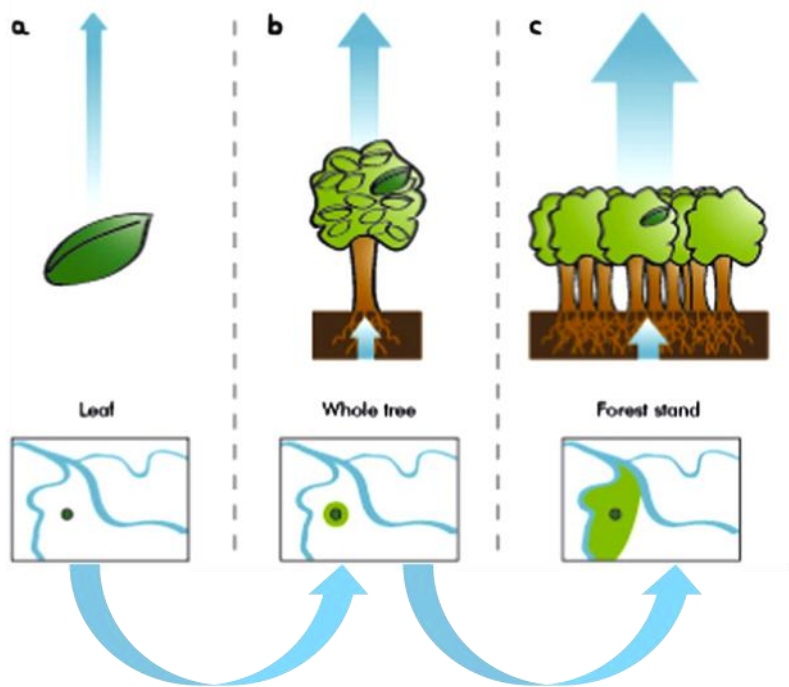


Fig. 4 - conceptual model for scaling up tree transpiration from leaves (a) to whole trees (b) to forest stands (c). Adapted from Asbjornsen et al. (2011)

Forest management practices, such as thinning, affect the amount of water transpired (Winkler et al. 2010). Despite this, these changes have two contrasting points of view. Firstly, usually, after thinning there are increases in tree transpiration and secondly due to the reduced number of remaining tree, usually, there is a decrease in total forest transpiration. However, a very low thinning intervention might result in non-significant changes in total transpiration due to the increase in individual tree transpiration. In this sense, a better knowledge of transpiration at different thinning intensities is needed.

### **Runoff and Drainage**

As observed in Table 1 there are fewer studies on runoff and drainage in forest ecosystems. Runoff refers to the portion of precipitation running over the land surface or through the soil profile to nearby stream channels. When precipitation intensity is greater than soil-infiltration rate, or precipitation amount exceeds soil-infiltration or percolation capacity, runoff happens. Infiltrated water can become surface runoff again as it flows laterally and downslope or routes to nearby stream channels as subsurface runoff. It is difficult to separate surface runoff and subsurface runoff in hydrological analysis (Chang,

2006). Drainage is the amount of water leaving the systems (Fig. 2). In the case of a forest stand, drainage out of the system means drainage out of the rooting zone of the trees and is difficult to measure. In most water balance models, drainage is calculated by allowing the soil profile to saturate with water and then calculating as runoff or drainage, the excess precipitation that occurs after saturation. (Landsberg and Gower, 1997).

For a catchment<sup>8</sup>, the runoff and drainage terms determine the catchment water yield<sup>9</sup> (Landsberg and Gower, 1997). The first review of forest catchment water yield experiments throughout the world can be found in Hibbert, (1965), where this author, after careful study of the thirty-nine forest treatment studies, made the following generalizations: 1) Reduction of forest cover increases water yield. 2) Establishment of forest cover on sparsely vegetated land decreases water yield. 3) Response to treatment is highly variable and, for the most part, unpredictable. Bosch and Hewlett, (1982) in a review of effects of vegetation changes on water yield, report that coniferous and eucalypt

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<sup>8</sup> A catchment is defined as the drainage area that contributes water to a particular point along a channel network (or a depression), based on its surface topography (Wagener et al., 2007).

<sup>9</sup> Klein et al. (2013) consider as water yield (WY), the difference between precipitation (P) and evapotranspiration (ET) for a particular area ( $WY = P - ET$ ).

cover types cause ~40 mm change in annual water yield per 10% change in forest cover. Deciduous hardwoods are associated with a 25-mm change in yield per 10% change in cover, while 10% changes in brush or grass lands seem to result in ~10mm change in annual yield. On other hand, studies have shown that re-establishment of forest cover (forestation) results in a decrease in water yield, though responses to treatment are highly variable and unpredictable (Hubbart, 2009). Despite this, Wang et al., (2011) report that catchment studies are integrated systems, and impacts on water yield should be considered collectively rather than focusing on specific processes such as forest interception, stemflow, and infiltration. For an extensive review of paired forest catchment studies see Webb et al., (2012) and references therein.

### **Water-use and Water-use Efficiency**

Forest scientists have long sought to develop reliable techniques for estimating whole plant water-use in trees (Wullschleger et al., 1998), and this topic has attracted considerable attention from the public over the last centuries (Andréassian, 2004). However, water use by forest tree species goes beyond the specific differentiation in terms of sensitivity to environmental factors (Aranda et al., 2012). Thus, understanding the

water use in trees is complex because it requires a good knowledge of the hydrological forest balance (Dvorak, 2012).

As reported above, forest water use or water loss encompasses two separate processes: Firstly, water is taken up by tree roots from the soil and evaporated through the stomata on the surface of leaves. This is called transpiration and is a physiologic process responding to soil water status and atmospheric factors. The second process is the interception of water by the leaves surfaces, branches and trunks during precipitation, and its subsequent evaporation. In other words total forest water use (WU) is the sum of transpiration and evaporation of intercepted water (Cannell, 1999; Almeida et al., 2007). Despite this, many authors (e.g. Wullschleger et al., 1998; Simpson, 2000; Schiller et al., 2002; Schiller et al., 2003; Gyenge et al., 2009; Schiller, 2011; Aranda et al., 2012) do not consider the water losses by interception as WU. Especially when these authors scale-up individual tree transpiration rates to obtain stand-level water use. These two different concepts of WU usually promote erroneous comparison between forests water-use. Moreover, Hubbard et al., (2010), consider WU only the water lost by trees to the atmosphere when stomata open to acquire CO<sub>2</sub> for photosynthesis. Therefore, this concept by itself does not consider the water lost during night-time or

through cuticular transpiration. Aranda et al. (2012) report that it can be a mere consequence of the recharge of internal water reservoirs on the first night hours, but also a consequence of incomplete stomatal closure at night. Although theoretically it should not be referred to as “use”, it should be considered as a pathway of water loss. In that sense, this concept of water-use definitively should not be used to define water use, although it may be used to determine water-use efficiency.

Water-use efficiency (WUE) in turn, is one of the key parameters of ecohydrology<sup>10</sup> and ecosystem management (Ito and Inatomi, 2011). However, their definition depends upon the particular context in which it is being discussed, including where the water is in relation to the plant (i.e. inside the plant or in its environment), the time scale over which efficiency is measured (e.g. instantaneous exchange of water vapour for carbon dioxide gas versus biomass accumulation or yield) and the precise measure of efficiency in relation to carbon gain (i.e. carbon dioxide influx, biomass accumulation or economic yield) (Bacon,

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<sup>10</sup> Ecohydrology has been recognized as a field that explicitly studies the interactions between ecological and hydrological processes. Thus, ecohydrological studies typically focus on understanding the linkages, interactions and feedbacks between hydrologic flows and ecosystem processes, as well as how these interconnections are manifested and exert distinct controls across multiple scales (Asbjornsen et al. 2011).

2004). Albaugh et al., (2013) report that WUE in a forest depends on the objective of a particular study, that can be expressed as stem volume production per unit volume of water transpired or in the case of bioenergy, expressed as biomass production (i.e., all components that will be used for bioenergy) per unit water transpired, or divided by estimates of annual evapotranspiration rates (Almeida et al., 2007). In most of the literature (Jones, 1992; Dye, 2000; Bacon, 2004; Hubbard, 2010;), WUE is discussed either in terms of an instantaneous measurement of the efficiency of carbon gain per water loss; or as an integral of such an efficiency over time, commonly expressed as the ratio of water use to biomass accumulation, or harvestable yield.

In the latter half of the twentieth century, the discovery that the carbon-isotope fractionation capability of the photosynthetic process, and that the resultant ratio of stable carbon isotopes within the plant tissues could be used to assess both an instantaneous or integrated measure of plant WUE, moved the discussion of WUE firmly within the realms of modern plant science (Bacon, 2004), and has been used in a wide range of plant species to assess intrinsic water use efficiency -  $WUE_i$  (Cumbie et al., 2011). The term intrinsic was introduced to compare photosynthetic properties independent of (or with a common)



evaporative demand (Osmond et al., 1980) and has been widely related to long-term trends in the internal regulation of carbon uptake and water loss in plants (Linares et al., 2011). A more detailed description of WUEi by stable isotopes in trees is presented below and for a better understanding of the physiological theory see: (Farquhar and Richards, 1984; Farquhar et al., 1989; Scheidegger et al., 2000; McCarrol and Loader 2004; Seibt et al., 2008; Werner et al. 2012). However, before entering into the topic of intrinsic water use efficiency (WUEi) by stable isotopes in trees, it is important to get a clear understanding of the stable isotope notation, as follows.

### **Stable Isotope notation**

The ratio (R) of the rare-to-common (or heavy-to-light) stable isotope in any material contains valuable information about both processes and sources. In addition, because of the very small absolute abundances of each isotope in any material, by convention the stable isotope composition is expressed as the difference in isotope abundances

in a sample relative to an international standard, usually VSMOW<sup>11</sup> and/or VPBM<sup>12</sup>.

Relative abundances are then used to discuss particular issues of interest, and this is done more easily than with absolute isotope abundance or ratios, which only vary in the third decimal place. This has led to the use of the widely accepted ( $\delta$ ) notation, where the isotope ratio of our unknown sample (SA) is expressed relative to an internationally accepted standard as:

$$\delta^x E = \left( \frac{R_{sample}}{R_{standard}} - 1 \right) \times 1000 \quad (3)$$

where E is the element of interest (e.g. Carbon or Oxygen); x is the atomic mass of heaviest isotope of element E in the ratio (R);  $\delta$  value is multiplied by 1000 to allow the expression of small differences in units that are convenient to use, parts per thousand (ppt), or the commonly used “per mil” notation represented by the symbol, ‰.

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<sup>11</sup> The standard mean ocean water (SMOW), is no longer available, so the International Atomic Energy Agency or IAEA (<http://www.iaea.org/>) makes (mixes) an equivalent water sample in Vienna of a similar isotope value now known as VSMOW.

<sup>12</sup> The carbon isotope standard, the fossil belemnite from the PeeDee geological formation is no longer available and instead the IAEA “builds” an equivalent carbon standard in Vienna of a similar isotope value (VPDB) though for carbon isotope analyses it is still referred to as PDB.

## **Carbon and Oxygen stable isotope and their application in WUE studies**

The major attraction of stable isotope techniques in ecophysiological studies lies on their ability to integrate plant processes over time. This is particularly interesting if we aim to determine tree responses to environmental changes (Ferrio et al. 2005). During the synthesis of plant biomass, various ecophysiological and climatological processes affect its stable isotope ratios of carbon and oxygen, and it can be successfully used to inferences about water-use efficiency in forest (McCarroll and Loader, 2004; Ferrio et al., 2005; Matteo et al., 2010; Kruse et al., 2012)

During photosynthesis, C<sub>3</sub> plants take preferentially <sup>12</sup>C instead of <sup>13</sup>C. The magnitude of this event is affected by several physiological processes, and is expressed in terms of carbon isotope discrimination ( $\Delta^{13}C_{plant}$ ), as defined by Farquhar et al. (1989):

$$\Delta^{13}C_{plant} = \frac{\delta^{13}C_{air} - \delta^{13}C_{plant}}{\left(1 + \frac{\delta^{13}C_{plant}}{1000}\right)} \quad (4)$$

where:  $\delta^{13}C_{air}$  and  $\delta^{13}C_{plant}$  refer to air and plant composition, respectively.

Variations in  $\Delta^{13}\text{C}_{\text{plant}}$  are frequently used to analyse temporal or spatial trends in plant carbon–water relations (Seibt et al. 2008). These authors report that the most often used version of the Farquhar et al., (1982) model—the linear form—relates  $\Delta^{13}\text{C}_{\text{plant}}$  to  $c_i/c_a$ , which is the ratio of intercellular ( $c_i$ ) to atmospheric ( $c_a$ )  $\text{CO}_2$  mole fractions:

$$\Delta^{13}\text{C}_{\text{plant}} = a + (b - a) \frac{c_i}{c_a} \quad (5)$$

Where:  $a$  is the fractionation during  $\text{CO}_2$  diffusion through the stomata (4.4‰; O’Leary 1981), and  $b$  is the fractionation associated with reactions by Rubisco and PEP carboxylase (27‰; Farquhar and Richards 1984).

The  $c_i/c_a$  ratio reflects the balance between assimilation rate ( $A$ ) and stomatal conductance ( $g_s$ ) according to Fick’s laws:  $A = g_s(c_a - c_i)$ . Probably,  $g_s$  is one the most relevant and complex plant physiological processes, acting during plant evolution as a crossroad in the interplay between carbon fixation and water loss (Aranda et al. 2012). However, the  $g_s$  value is related to  $\text{WUE}_i$  by a constant factor equal to 1.6 ‰, hence linking the leaf gas exchange of carbon and water (Seibt et al. 2008). Thus it is possible to express discrimination ( $\Delta^{13}\text{C}_{\text{plant}}$ ) in terms

of net assimilation ( $A$ ) and conductance of water vapour ( $g_s$ ), which is termed the intrinsic water-use efficiency ( $WUE_i$ ) (Loader et al., 2011).

$$\frac{A}{g_s} = WUE_i = \frac{(c_a - c_i)}{1.6} \quad (6)$$

Thus,  $WUE_i$  derived from plant isotope data is often applied as indicator of long-term trends in the internal regulation of carbon uptake and water loss in plants (Linares et al., 2011). In other words  $WUE_i$  is a measure of the efficiency of carbon gain per water loss, expressed in  $\mu\text{molCO}_2/\text{molH}_2\text{O}$ . Thus, Carbon stable isotopes have provided key insights into biogeochemical interactions between plants, soils, and the atmosphere (Dawson et al., 2002).

Since both stomatal conductance ( $g_s$ ) and photosynthetic rates ( $A$ ) have an influence on  $c_i/c_a$ , it is often difficult to determine the extent to which inter-specific differences in plant  $\delta^{13}\text{C}$ , consequently  $WUE_i$ , are determined by differences in  $g_s$ ,  $A$ , or both (Scheidegger et al. 2000; Keitel et al., 2006; Seibt et al. 2008; Barnard et al., 2012). To separate the independent effects, Scheidegger et al. (2000) propose measuring both  $\delta^{13}\text{C}$  and  $\delta^{18}\text{O}$  creating the dual isotope conceptual model (Fig. 5). Whereas  $\delta^{13}\text{C}$  reflects  $c_i/c_a$ ,  $\delta^{18}\text{O}$  generally varies with ambient humidity, which in turn reflects changes in water use (Dawson et al., 2002). In

particular, the  $\delta^{18}\text{O}$  signature of plant matter largely reflects abiotic (and non-physiological) effects on water source enrichment and evaporative demand (Kruse et al., 2012). In this sense, the oxygen stable isotope composition of plant tissue ( $\delta^{18}\text{O}$ ) can help separate the independent effects of  $A$  and  $g_s$  on  $\delta^{13}\text{C}$ , because  $\delta^{18}\text{O}$  shares dependence on  $g_s$  with  $\delta^{13}\text{C}$ , but it is thought to be independent of the variation in  $A$  (Scheidegger et al. 2000; Grams et al., 2007; Moreno-Gutiérrez et al., 2011; Roden and Farquhar, 2012).

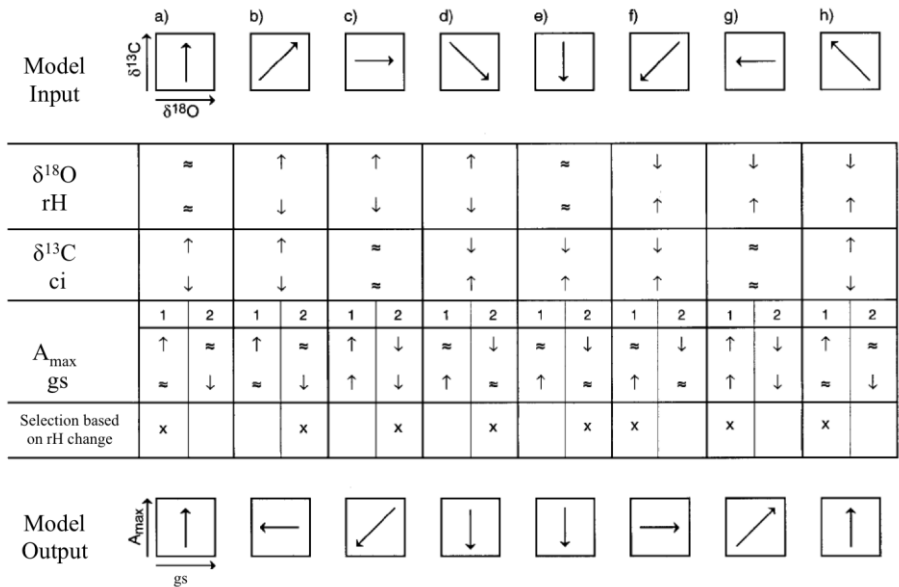


Fig. 5 - The conceptual dual isotope model: scheme of the eight scenarios from a) to h) based on all likely  $\delta^{13}\text{C}$ -  $\delta^{18}\text{O}$  combinations (model input). The changes are shown by the arrows. Relative humidity (rH) is derived from  $\delta^{18}\text{O}$ , partial pressure of  $\text{CO}_2$  in the leaf intercellular spaces (ci) from  $\delta^{13}\text{C}$  (up - or downward arrows represent increasing or decreasing values,  $\approx$  indicates insignificant changes). For each scenario there are two possible cases, denoted as 1 and 2, with corresponding changes in average maximum net photosynthesis ( $A_{\text{max}}$ ) and stomatal conductance (gs). The model output is determined by the selection based on the rH change. Adapted from Scheidegger et al. (2000).

*"A short explanation about dual isotope model"*

Fig. 5 shows all combinations of  $\delta^{13}\text{C}$  and  $\delta^{18}\text{O}$  values in eight likely scenarios (a to h) where the outputs of the model (i.e. stomatal conductance (gs) and photosynthetic rates ( $A_{\text{max}}$ ) are selected on the

basis of changes in relative humidity (rH). These authors (i.e. Scheidegger et al. 2000) show different environmental conditions that cause higher ( $\uparrow$ ), lower ( $\downarrow$ ) or similar ( $\approx$ )  $\delta^{13}\text{C}$  and  $\delta^{18}\text{O}$  values. For example, we refer to scenario f) where both  $\delta^{13}\text{C}$  and  $\delta^{18}\text{O}$  values decrease. We know from the observed decrease in  $\delta^{18}\text{O}$  that rH must have increased, in the same way a decrease in  $\delta^{13}\text{C}$  promotes an increasing in  $c_i$ . This increase is explained in two possible cases (1)  $A_{\text{max}} \approx$  and  $g_s \uparrow$  or (2)  $A_{\text{max}} \downarrow$  and  $g_s \approx$ . However “the most likely case” is (1) because the isotope patterns are related to a strong stomatal reaction, whereas  $A_{\text{max}}$  remains relatively unaffected. This variation, in turn, drives changes in transpiration rates (water use) and  $g_s$ . On the other hand, in the scenario e) where  $\delta^{18}\text{O}$  show insignificant changes ( $\approx$ ), and  $\delta^{13}\text{C}$  values decrease ( $\downarrow$ ). We could say that rH also show insignificant changes. However, the decrease in  $\delta^{13}\text{C}$ , in this scenario, indicates an increase in  $c_i$  that can be explained by (1)  $\approx A_{\text{max}}$  and  $\uparrow g_s$  or (2)  $\downarrow A_{\text{max}}$  and  $\approx g_s$ . In this case “the most likely case” is (2). In summary, stable isotope analysis is a powerful tool for assessing plant carbon and water relations and their impact on biogeochemical processes at different scales (Werner et al., 2012; Barnard et al. 2012; Roden and Siegwolf,



2012). The powerful approach using simultaneous assessment of  $\delta^{13}\text{C}$  and  $\delta^{18}\text{O}$  (dual-isotope model) can be used successfully in the analysis of WU and WUE in hydrology-oriented silviculture. However, care is needed to avoid various pitfall of over-interpretation (see Roden and Siegwolf, 2012).

### **Forest Management practices and their impact on hydrological balance**

The rapid climate changes observed have modified the fundamental rationale for managing forests toward resilience and sustainability. Resilient forests, are those that can accommodate a changing climate or changes in natural or anthropogenic disturbance while maintaining their diversity, health, and productivity (Crow, 2012). While sustainable forests are those that provide not only timber for present and future generations, but also maintain their environmental values and social services. Thus, forest management in a changing environment is a crucial research task, which needs the development of new methods and research techniques (Cescatti and Piutti, 1998). Furthermore, these changes increase our need to understand the interactions between forests and water (Calder et al., 2007). Despite this,

water-saving has not been considered a target in many afforestation programs, which are typically more centred in either increasing productivity or optimizing forests as carbon sinks. The expected increase in the importance of water as a limiting factor for the future of many forests gives support to its inclusion as a priority in the planning of new forest systems. However, gaps in knowledge may prevent an effective inclusion of water issues as a management priority in many cases (Aranda et al., 2012). A large number of small field-scale experimental studies using a paired catchment approach have been conducted to better understand forest hydrologic processes, their interactions with the environment, and their eco-hydrologic impacts (Amatya et al., 2011). Additionally, forest managers are still working to fully understand the effects of forest management practices and their modifications on hydrological balance.

As reported above, commercial plantations or natural forest, normally: (i) increase interception loss due to more extensive canopy cover and leaf area density; (ii) increase transpiration loss due to increased biomass and total leaf area; (iii) increase disturbance of the soil structure, infiltration and moisture holding capacity (Smakhtin, 2001). Inversely, forest clear-cut, or converting forest to vegetation of smaller

sizes and/or lower densities, is expected to cause an increase in water availability (Chang, 2006), i.e. decrease interception and decrease stand transpiration. However, forest management effects on water vary with the size of the area treated the type of treatment, forest type, soils, climate, and time after treatment.

In summary, forest management practices affect the pathways of water within the forest system, which in turn affect watershed outputs. So, understanding how management practices affect the water balance of forests is crucial for managing water fluxes in forests and for making an efficient use of water (Forrester et al., 2012). It is clear that, many factors have the potential to affect forest-water relationships. Several of these factors are amenable to management control and there are possibility to design and manage forest plantations intentionally for a better water-use (Vanclay, 2009). Hence, an understanding of the processes controlling water-use and water-use efficiency in forests is important for management decisions (Dye, 2000), and constitute a key to hydrology-oriented silviculture.

From the point of view of forest managers, the most important issues associated with forest hydrology are the question of silvicultural practices and clear-cutting and the effects of water balance of forests on

the growth of trees (Landsberg and Gower, 1997). Silvicultural practices have the potential to alter canopy conductance, leaf area, sapwood area and the water potential gradients that exist between soil and canopy and as a result, change both the amount of water transpired and the amount of carbon assimilated during photosynthesis (Hubbard et al. 2010). Thinning, which partially removes trees from a forest in order to redistribute tree growth or to leave fewer and consequently more valuable stems, is an important and common silvicultural practice (Loustau et al., 2005; Weiskittel et al., 2011), and represent a proactive forest management method to maintain productivity (Magruder et al., 2013). It decreases stand density and leaf area, improves tree productivity, reduces wildfire risk and maintains the forest's health and increases in many cases the use of resources such as water. Thinning also can involve increased availability of light, water and nutrients, increased uptake of those resources by individual trees and an increase in resource-use efficiencies (Forrester, 2013), and has been extensively used to enhance tree water status and growth by means of increasing soil water availability, which is achieved as a consequence of reduced stand transpiration and canopy interception of precipitation (Aussenac and Granier, 1988; Bréda et al., 1995; Moreno-Gutiérrez et al., 2011).

Thinning may also increase the efficiency with which trees and stands use water. Higher resource acquisition or availabilities have been associated with higher WUE of trees and stands (Stape et al., 2004), which may be reinforced following thinning. Thinning effects on WUE have received little attention, but this information provides an important basis for linking silvicultural interventions to the management of water resources (White et al., 2009). Furthermore, thinning has an impact not only in the interrelationships among residual trees but also on the surrounding microclimate conditions (Aranda et al., 2012).

Despite of general positive effects of thinning on tree water relationships, it has been also observed in some cases, an abrupt change in the evaporative demand around remaining trees (Aranda et al., 2012). However, this increased demand can be offset by a concomitant decrease in transpiration of the stand. As a result there would be little or no changes in the forest hydrological balance after thinning. Thus, a reliable study is needed to answer if it is possible that thinning actually results in increases of water-yield, and more specifically what is the best thinning intensity to increase water yield while maintaining or improving forest resilience in a scenario of constant climate change?

## **Experimental research area**

Here is presented a general description of the experimental site, the tree species (*Pinus halepensis* Mill.) as well as materials and methods used in the completion of research for this thesis. Specific methods and more detailed descriptions can be found in the corresponding Material and Methods sections corresponding to each scientific paper in the Chapters 2, 3 and 4.

### **The species**

*Pinus halepensis*, also named Aleppo pine after the largest city of Aleppo-Syria, is a pine native to the Mediterranean area. In Spain, this species is known as “Pino carrasco”. This species is generally found at low altitudes, from 350 to 970 m.a.s.l., but it can grow at altitudes of 1400 m in the south of Spain, and up to 1700 m in Morocco, Algeria and Tunisia (Farjon, 2005). Aleppo pine can be found over its entire distribution range at rainfall from 200mm to more 1500 mm, i.e. ranging from lower-arid to the humid bioclimate. It does, however, occur most abundantly in the semi-arid to sub-humid zones between 350m and 700m. (Quézel, 2000). It is considered one of the most important forest species

in the Mediterranean Basin (Maestre et al., 2003) and tends to be dominant in forest stands where it is present (Alberdi et al., 2013). This species has been over the past decades the most widely used for afforestation and reforestation schemes in large areas of Mediterranean Spain. Especially because of low-technical requirements for nursery production, high resistance to adverse climatic and soil conditions, and because it was also considered a pioneer species, favouring the establishment of late successional ones (Maestre and Cortina, 2004). Moreover, its drought tolerance has led to its use in high-density plantations for controlling soil erosion in degraded lands and recently for converting cereal crops lands and old fields into forests via the European Farmland Afforestation Programme promoted by the Community Agrarian Policy (CAP) (Navarro et al., 2013).

According to the National Forest Inventory of Spain the species occupies 1,500,000 hectares (MARM, 2012), (Fig. 6), being 23% in the Valencia community, 19% in Catalonia and 14% in Castilla-La Mancha (Serrada et al., 2008). The resistance of Aleppo pine to environmental stresses is related to its ability to avoid lethal damage (embolism) resulting from drought, high temperature and insolation, which are the

most common stress factors in the area of its natural distribution (Schiller, 2000). Thus, this is considered key species to study water-use relationships in Mediterranean areas when taking into consideration climate change.

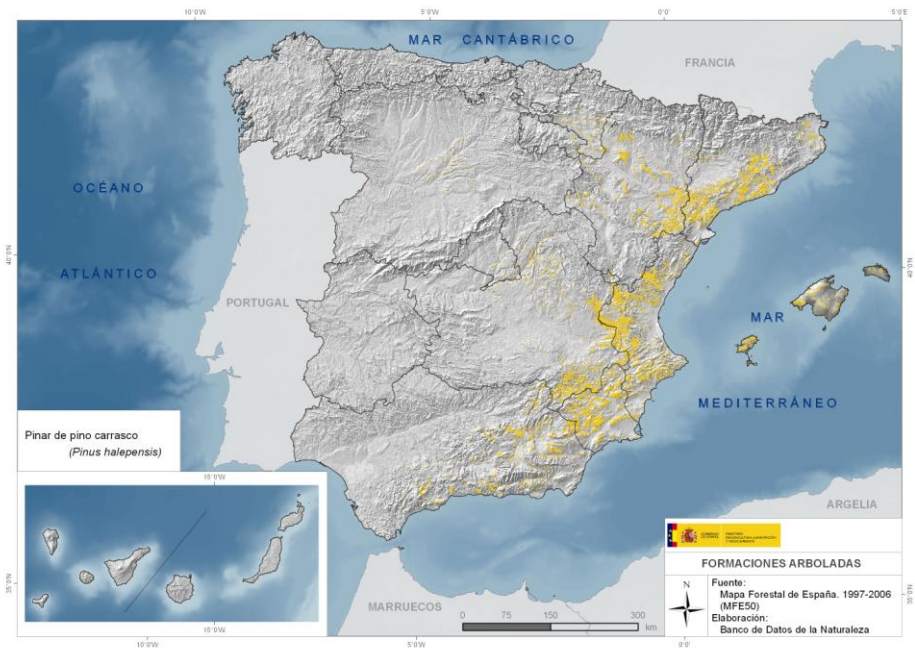


Fig. 6 - Distribution of *Pinus halepensis* Mill. in Spain (Source: [http://magrama.gob.es/es/biodiversidad/servicios/banco-datos-naturaleza/informacion-disponible/formacion\\_arboladas.aspx](http://magrama.gob.es/es/biodiversidad/servicios/banco-datos-naturaleza/informacion-disponible/formacion_arboladas.aspx))

## The site

This thesis was carried out in a Public Forest located in Ayora's county in the southwest of Valencia province, Spain (39° 05' N, 1° 12' W)



at 950 m.a.s.l. denominated as “La Hunde forest”, where the species *Pinus halepensis* Mill. is dominant (Fig. 7). The *P. halepensis* plantations were established during the late 1940s with high densities (ca. 1500 trees ha<sup>-1</sup>), and no forest management has been carried out mainly due to the perceived role of the forest in soil protection.

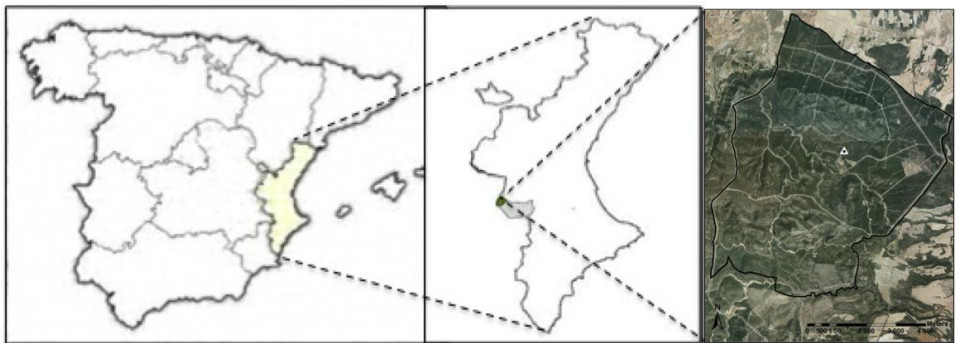


Fig. 7 - Localization map of the experimental area - Comunidad Valenciana, Spain and Ayora-Cofrentes Valley - La Hunde public forest

The climate is the main factor that shapes the physical environment in Mediterranean areas. Yearly rainfall varies from 100 to more than 2500 mm while the average temperature ranges from 5 to 20 °C. The winter temperature occasionally falls below 0 °C at sea level, and snow and below-zero temperatures are common at high altitudes. The existence of an intense summer drought period is the prevalent feature of Mediterranean climates at a global scale (Moussouris and Regato, 1999). According De Martonne aridity index (De Martonne,

1926), the climate in the experimental area is classified as Mediterranean semiarid or semiarid following (Thorntwaite, 1948), that is characterized by mild winters and hot and dry summers, with precipitation concentrated in autumn, winter and early spring (Fig. 8). Total rainfall varies strongly from year to year and torrential downpours or dry winds can occur.

The average (1960-2011) annual precipitation in the experimental area is 479 mm, typically with high intra and inter-annual variability (Fig. 8). Mean annual temperature (1960-2011) is 14.2 °C (Fig. 9). Mean annual potential evapotranspiration is 749 mm (Thorntwaite) and reference evapotranspiration is 1200 mm (Hargreaves) (Pérez-Cueva, 1994). The experimental area is characterized by an important annual variation in temperatures, with hot summers and cold winters (Fig 9). Mean monthly maximum air temperature ranges between 11.2°C (December and January) and 31.5°C (July and August), while mean monthly minimum temperature ranges between 2.6°C (from December and January) and 15.8°C (July and August).

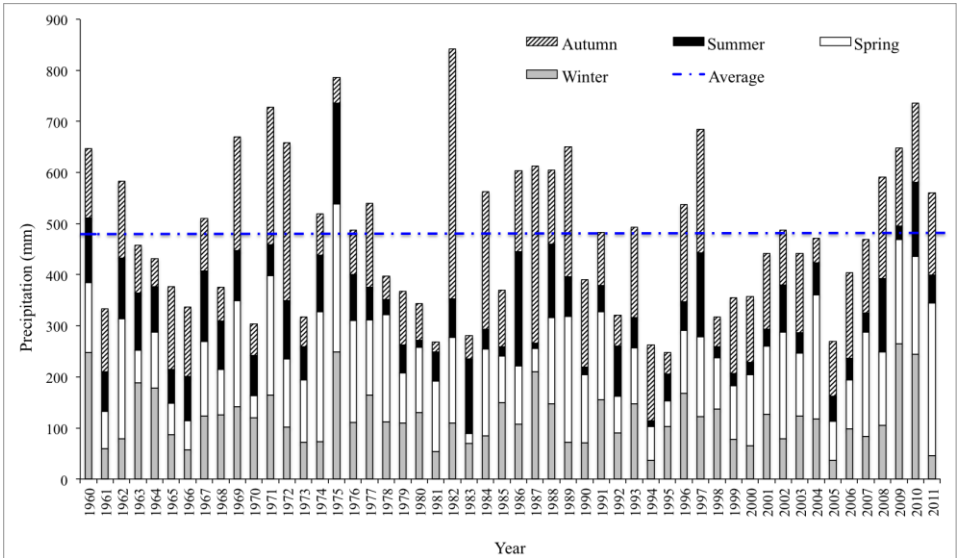


Fig. 8 - Yearly precipitation (mm) in the experimental area per season

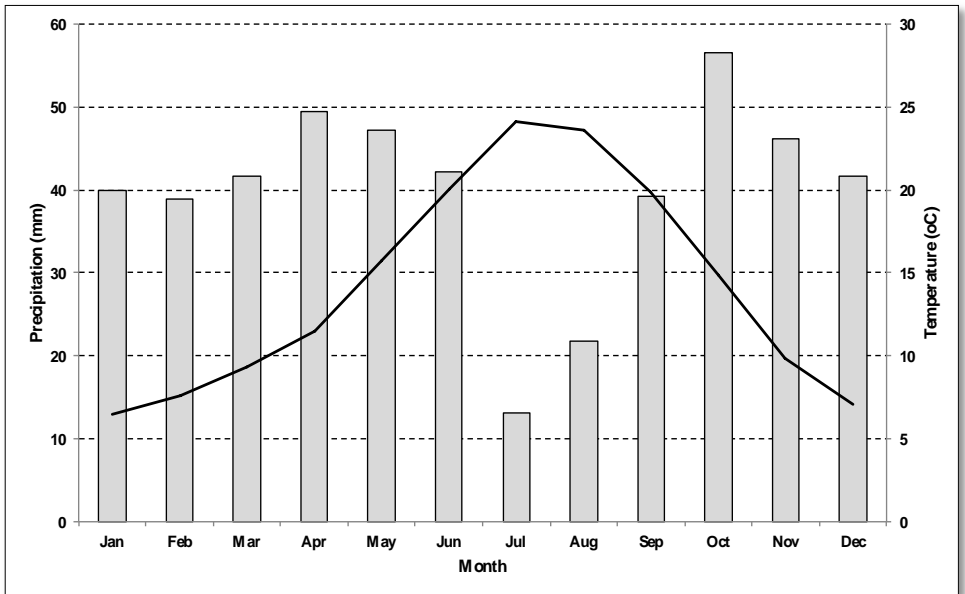


Fig. 9 - Monthly average (1960-2011) precipitation and mean temperature in the research experimental area.

## **Experimental thinning treatments**

Thinning is a silvicultural treatment made to reduce stand density of trees primarily to improve growth, enhance forest health, or to prevent potential mortality. Thinning is usually defined in terms of the method being applied, which determines which trees are to be removed. For example, considering the tree's crown classification, we would have:

*Thinning from Above:* The removal of trees from the dominant and co-dominant crown classes in order to favour the best trees of those below.

*Thinning from Below:* The removal of trees from the lower crown classes to favour those in the upper crown classes.

In this experimental study *thinning from below* was considered as the best choice. For this purpose three blocks or repetitions were randomized, 0.36 ha each (Fig. 10). Each block was further divided into four plots (30x30m, or effectively 28x28m when taking into account the border effects). At the beginning of 2008 three plots corresponding to thinning treatments at different intensities (Low, Moderate and High intensities) and another one as control were established to study the effect at short term (Fig. 10). All plots had less than 5 % of slope (Fig 11), so surface runoff processes can be considered negligible. In 2009,

forest inventories were carried out in all the plots in order to assess different forest variables (Table 1). An additional plot was thinned leaving approximately 10% of the trees in late 1998. It was considered in this study to assess the mid-term effects of thinning.

Table 2 - Forest structure variables in each plot studied. DBH is average diameter at breast height, BA is basal area.

<b>Treatment</b>	<b>Cover (%)</b>	<b>Density (trees ha<sup>-1</sup>)</b>	<b>DBH (cm)</b>	<b>Mean height (m)</b>	<b>BA (m<sup>2</sup> ha<sup>-1</sup>)</b>
Control (C)	84	1489	17.8 ± 5.1	11.5	40.1
Low intensity (L)	68	744	21.2 ± 4.1	12.2	27.2
Medium intensity (M)	50	478	21.7 ± 4.0	11.3	18.2
High intensity (H)	22	178	20.4 ± 1.6	12.2	9.4
High intensity-1998 (H <sub>98</sub> )	41	155	25.2 ± 5.0	12.6	13.6

The soils in these plots are alkaline with pH between 7 and 8.2, relatively shallow (50-60 cm) and have sandy-silty loam texture. Field capacity in each plot was calculated as the average of SWC's readings in three dates (28-29/March/09; 12/Oct/10; 23/March/11) selected because rainfall in the previous two days was higher than 30 mm (see: soil water content point on the chapter three for a detailed description of field capacity).

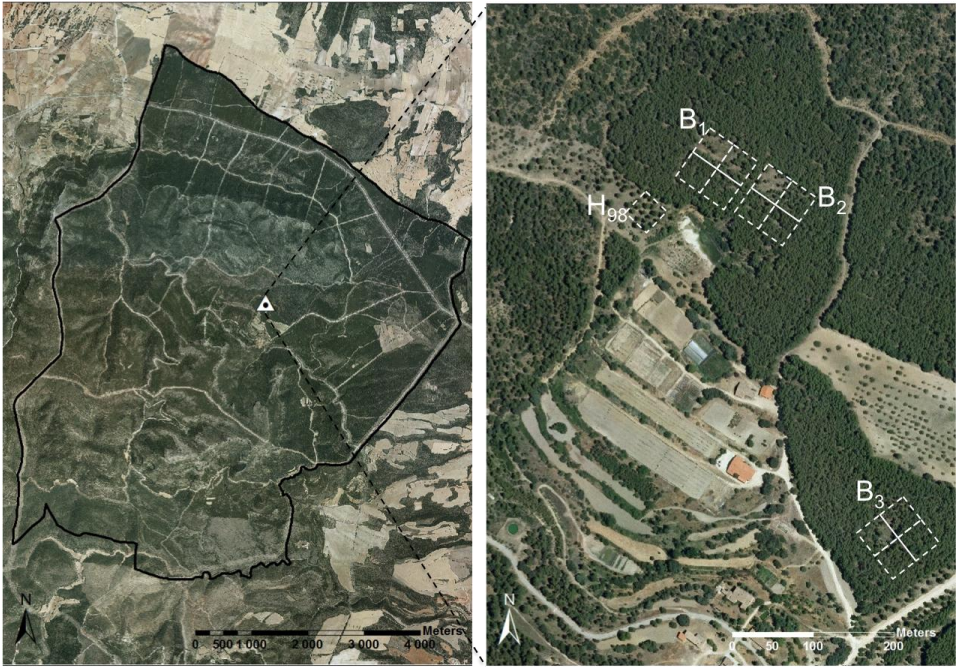


Fig. 10 - La Hude public forest. The triangle in shows the location of the experimental area. H<sub>98</sub> is the experimental plot thinned in 1998; B<sub>1</sub>, B<sub>2</sub> and B<sub>3</sub> are the blocks with four plots each which were thinned in 2008 at High, Medium and Low intensity and the fourth is the control (unthinned) plot.



Fig. 11 - a – thinning practice and b –biomass removal after thinning

## **General Material and Methods**

### **Forest Hydrological balance determinations**

Since this is a complex study that involves many measurements of forest hydrology components, all determinations were not started and ended at the same time due to field testing, calibration and improvement procedures. Therefore, precipitation, temperature, relative humidity and soil water content measurements began in March 2009 and ended in May 2011, transpiration measurements started in June 2009 and ended in May 2011. Precipitation partitioning, i.e throughfall and stemflow began in March 2009 and ended in February 2010 and September 2010 respectively. Thus, interception was assessed from March 2009 to February 2010. Additionally, values of historical precipitation and mean temperature from the period 1960-2008 were obtained in the “Ayora - La Hunde” and “Enguera - Las arenas” meteorological stations closest to the study site.

### **Precipitation and precipitation partitioning**

Precipitation ( $P_{pt}$ ) partitioning into throughfall ( $Th$ ), stemflow ( $St$ ), and interception loss ( $I$ ) after it reaches the canopy represents the

initial interaction between water cycle and forest (Sun et al. 2013). *Ppt* was measured with a standard rain collector (rain collector II, Davis instruments, USA) using a recording rain gauge with 0.2 mm per tip, on an open field 150 m from the experimental plots, without vegetation influence (Fig. 12a), the data were stored every 5 min with a data-logger (Onset, USA). *Th* was measured at one-week intervals by using four throughfall gutters collectors in each plot for all blocks, draining to 25 litres deposits. These collectors were made of PVC and their dimensions are 400 cm x 13.17 cm wide, set at 50 cm above the soil and sloping at 5° towards a 25-L plastic container, providing a total collecting area per treatment of 6.3 m<sup>2</sup> (Fig. 12b). For *St* measurement trees were equipped with a polyvinyl tube fixed and sealed around the trunk at about 1.3 m height, with a draining hole connected to a 10 l deposit (Fig. 12c). Four trees per treatment were selected with diameter ranges according to the DBH distribution found in each plot (Table 2). They were measured weekly (on trees belonging to block I) To scale up stemflow from sample trees we multiplied the stemflow average of each plot by the tree density in the same plot. Finally to calculate Interception loss (I) it was used the equation 2 (see: Forest hydrological balance section).



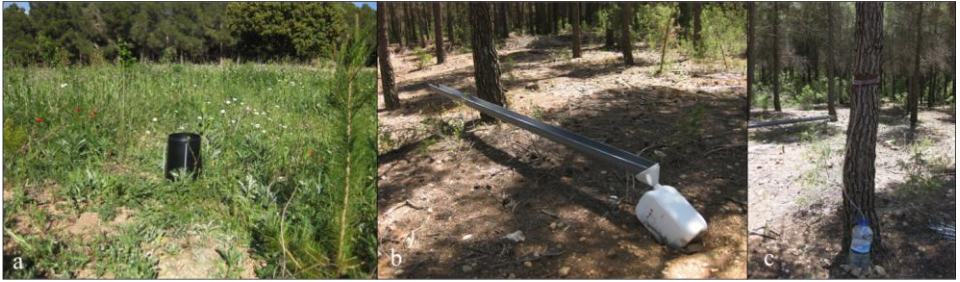


Fig. 12 - a – Precipitation gauge in an open field 150 m from the edges of experimental plots; b – Gutter collectors of throughfall and 25 litres deposit; c – polyvinyl tube collar fixed around the trunk to collect stemflow.

### **Transpiration (water-use)**

Throughout this thesis transpiration is also defined as water-use (WU). Tree transpiration was measured by means of sap flow sensors (HRM-30, ICT International Pty Ltd., Armidale/Australia - Burgess et al. 2001) (Fig. 13) in four trees per plot (Block I) chosen according to the frequency distribution of diameters. To convert the heat pulse velocity to sap flow velocity, raw values were corrected for probe misalignment (Burgess et al., 2001), differences in thermal diffusivity and wounding, by examining samples under light microscope (Barret et al., 1995). Baseline correction in the obtained series was performed according to Buckley et al. (2012). The accumulated daily values of sap flow ( $l\ day^{-1}$ ) (water transpired by the entire tree) were estimated considering a radial

distribution of the sap flux velocity in the sapwood of each selected tree following Delzon et al., (2004). The sapwood area per tree was measured after extracting samples with an increment core (5 mm) and measuring to the nearest 0.01 mm with a measuring table (LINTAB 6.0, Frank Rinn, Heidelberg, Germany) coupled with the TSAP-Win software package (Rinn, 2011). Total transpiration by the pine forest ( $Tr$ ,  $\text{mm day}^{-1}$ ) per treatment, was calculated by multiplying the daily transpiration ( $L \text{ day}^{-1} \text{ tree}^{-1}$ ) by the frequency of diameter class ( $f$ ) and the stand density (trees  $\text{ha}^{-1}$ ), using Equation:

$$Tr(\text{mm day}^{-1}) = \frac{\sum(\text{Tr}_n * f_n) * N^o}{10000} \quad (7)$$

Where:  $Tr_n$  is the daily transpiration of each treatment;  $f_n$  is the frequency of each tree class measured;  $N^o$  is the number of trees by stand (Table. 2) and division by 10,000 is the transformation of litres  $\text{ha}^{-1}$  into mm (Schiller, 2011).

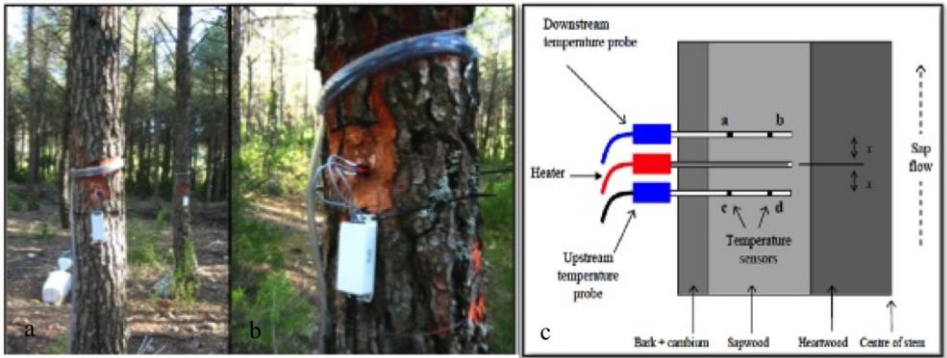


Fig. 13 - a and b – Transpiration sensor and c – Functional schematic diagram of sap flow sensor (Source: HRM-30, ICT International Pty Ltd manual)

To describe the relationships between water-use and water-use efficiency in the chapter four, the water-use (tree transpiration data) values starting in April to March of the following year, was considered as "physiological year". This approach is consistent with growth dynamic of Aleppo pine trees (De Luis et al., 2007; 2011) .

### Soil water content

Soil water content (SWC,  $\text{cm}^3 \text{cm}^{-3}$ ) was measured every 20 minutes by means of capacitance sensors, and averaged to a daily value, (Echo, Decagon Devices Inc., Pullman, WA). In each treatment plot (block I), 3 sensors were placed in the soil under the crown-projected area at 30 cm depth under 2 randomly selected trees out of the 4 sampled

for transpiration and other three sensors away from the direct influence of trees. The daily values obtained for each sensor were individually calibrated, averaged and converted into a single daily value per treatment. Soil depth in all plots was in the 50-60 cm range, below which fragmented rocks were very abundant.

### **Others components of Hydrological balance**

Bulk precipitation, transpiration, throughfall and stemflow were measured in the forest plots. The total amount of water reaching the soil was then considered to be the sum of throughfall and stemflow. In order to estimate infiltration into the soil, we assume that surface runoff is negligible and that water penetrates into the soil recharging the available soil water content (SWC) until SWC is equal or higher than field capacity ( $SWC/FC = 1$ ); i.e the soil reaches saturation. This term was divided into stand transpiration ( $Tr$ ) and the residue was considered upper soil horizon evaporation and the interception and transpiration of understory. Once the soil saturation is attained deep infiltration begins and proceeds at a rate equal to  $P_n$ .

## **Dendrochronology and Dendroclimatology**

Two 5mm increment cores were taken from each tree (north and south), to integrate the potential variability in tree growth around the stem and to help identify false or missing tree-rings; they were taken at 1.30 m in each tree after the completion of the transpiration measurements in the same trees (Fig 13 and 14a). Additionally thirty-two trees four trees per plot in each block were cored to avoid the possible under-estimation of the climate-growth correlations (Mérian et al., 2013), except in one of the Medium thinning intensity plots which was badly damaged due to strong winds in 2009. Each core was air dried and mounted on a wooden support (Fig 14b,c) then it was sanded and cleaned until xylem cells were clearly identified under the stereomicroscope (Fig 14d,e). All cores were visually cross-dated and measured to the nearest 0.01mm with a measuring table (LINTAB 6.0, Frank Rinn, Heidelberg, Germany) coupled with the TSAP-Win software package (Rinn, 2011) (Fig 14e). Some cores were also verified with scan ImageJ software (Rasband, 2011) to enhance contrast. Cross-dating of the tree-ring width (TRW) series was evaluated using the COFECHA programme (Holmes, 1983) and with the tutorial by Grissino-Mayer, (2001). Additionally the

dplR R-package (Bunn, 2008) was used to better visualize possible errors. The cores that presented missing rings were discarded in the following analyses. The average series length was 50.2 ( $\sigma=2.51$ ) years, autocorrelation at 1-year averages was 0.76 ( $\sigma=0.09$ ), and Gini coefficient, which describes annual changes in the inequality of size and size increment was 0.42 ( $\sigma= 0.08$ ), where 0 indicates perfect equality (the size or growth of all individuals is the same) and 1 indicates perfect inequality. The average series correlation to the master chronology is 0.79 ( $\sigma=0.07$ ), excluding the cores from High intensity thinning made in 1998, which were analysed separately obtaining a correlation of 0.80 ( $\sigma=0.09$ ). To compare the real impact of thinning on the growth in the chapter two, Basal Area Increment (BAI) in  $\text{cm}^2 \text{yr}^{-1}$  was calculated for each tree. BAI was preferred to ring width in this chapter because it provides a more accurate quantification of wood production due to the continually increasing diameter of a growing tree (Biondi and Qeadan, 2008; Martín-Benito et al., 2010; Michelot et al., 2012; Primicia et al., 2013; Wang et al., 2012).

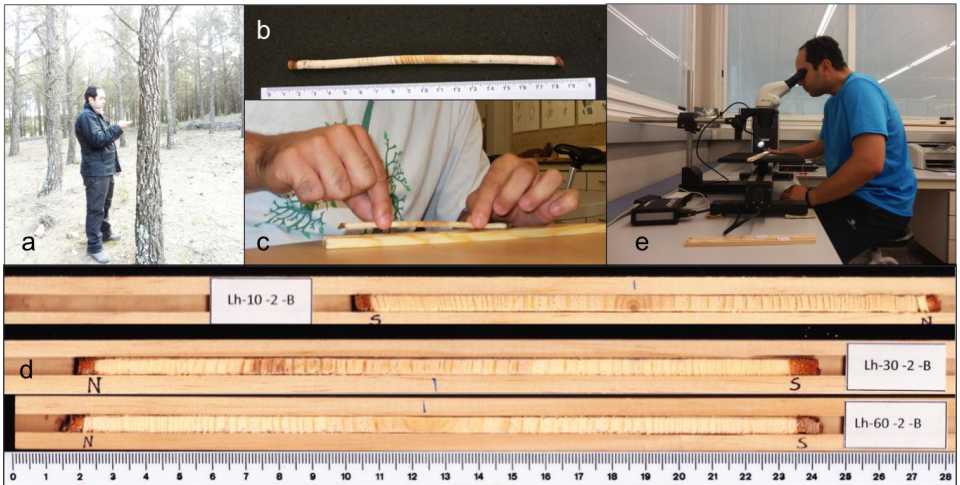


Fig. 14 - Dendrochronology procedures a – core extracting in the experimental area; b – Air-dry core before mounting; c – mounting on a wooden support; d – final result after sanding and cleaning; e – measurement in LINTAB 6.0 coupled with TSAP-WIN software

To perform the dendroclimatic analysis, the tree ring width (TRW) series were detrended using DetrendeR – R package (Campelo et al., 2012) and dplR – R package (Bunn, 2008), to reduce the noise caused by systematic non-climatic change due to tree age (Cook and Briffa, 1990), using a cubic smoothing spline function a wavelength fixed at 67% (proximally  $2/3$  – Cook et al., 1990) of the length of the series, with a 50% frequency response. In some cores a negative exponential method was utilized. Each measured series was standardized dividing observed values by predicted values to obtain dimensionless a TRW index series (TRWi) which was averaged using a robust bi-weight

mean. Additionally the temporal autocorrelation was removed from each series using an autoregressive model (Cook and Briffa, 1990) obtaining the standard and residual chronology. To determine the length of the residual chronology for which climatological responses would be tested, we used a running (20-year) mean of the expressed population signal (EPS) statistic (Wigley et al., 1984) to provide an indication of chronology reliability.

### **Carbon and Oxygen isotopes analyses and intrinsic water-use efficiency**

Carbon and Oxygen data are presented as  $\delta^{13}\text{C}$  and  $\delta^{18}\text{O}$  respectively relative to the international VPDB standard expressed in parts per thousand (‰).  $\delta^{13}\text{C}$  is inversely and linearly correlated with  $C_i/C_a$ , the ratio of intercellular to atmospheric  $\text{CO}_2$  concentrations in leaves. This ratio reflects the relative magnitudes of net photosynthesis rate ( $A$ ) and stomatal conductance ( $g_s$ ), and thus  $\delta^{13}\text{C}$  is a good indicator of plant intrinsic water use efficiency ( $\text{WUE}_i$ ), which is given by the ratio  $A/g_s$  (Farquhar et al., 1989). On the other hand,  $\delta^{18}\text{O}$  of plant organic material is related to  $e_a/e_i$  which is the ratio of atmospheric to



leaf intercellular water-vapour pressure, and thus is strongly affected by changes in  $g_s$  (Scheidegger et al., 2000; Barbour, 2007). Since  $\delta^{18}\text{O}$  is related to  $g_s$ , but is unaffected by  $A$ , it can help separate the independent effects of  $A$  and  $g_s$  on  $WUE_i$  (Scheidegger et al. 2000; Barbour 2007; Moreno-Gutiérrez et al., 2011). For this propose, analyses of  $\delta^{18}\text{O}$  and  $\delta^{13}\text{C}$  were performed on subsamples of rings from three trees per plot in the transpiration sensors block.

Samples from the previously measured and dated cores were taken for isotopic analyses. The sanded core surfaces were thoroughly cleaned with ethyl alcohol p.a. and small sections, including both early and latewood, were taken with a surgical blade under the stereomicroscope (Fig. 15a,b). These samples were ground in an agate mortar (Fig. 15c) and weighed to the nearest 0.001g in Ag capsules for  $\delta^{18}\text{O}$  analysis and Sn capsules for  $\delta^{13}\text{C}$  analysis (Fig. 15d,e).

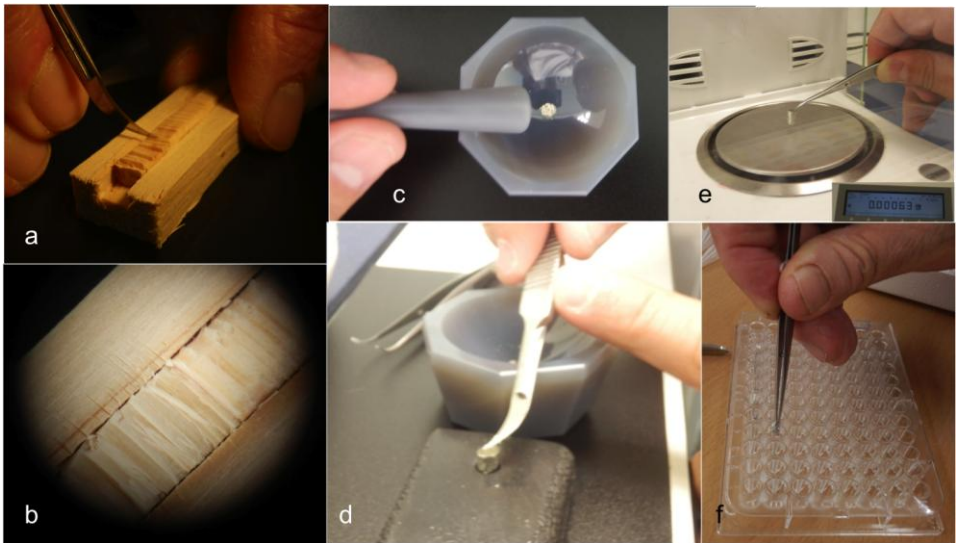


Fig. 12 - Isotopic analysis procedures. a – wood material extraction by surgical blade; b – stereomicroscope view; c – grinding in an agate mortar; d – putting sample in the capsules; e – weighing of wood sample f – placing capsule in a tray before sending it for analysis.

The stable carbon and oxygen isotopic signatures of the wood samples were measured online using an isotope ratio mass spectrometer (Finnigan MAT, delta S, delta XL plus, delta XP) via a Conflo II interface for the combustion/pyrolysis of organic material at the Paul Scherrer Institut, Ecosystem Fluxes Research Group, Villigen, Switzerland.

Stable isotopes in samples from the rings for the period 1995 to 2010 for Control and High-98 were analysed in order to explore the

effect of thinning for a longer period of time and for the period 2004 to 2010 for the more recent thinning treatments.

### **General statistical analysis**

Specific statistical analyses were carried out for each set of data throughout this thesis. Before the analyses, data were tested for normality and homogeneity of variances by Kolmogorov-Smirnoff and Levene tests, respectively. The t-student test or Tukey's post hoc test or Kruskal-Wallis test were the procedures used for mean comparisons among treatments in each of the variables considered: tree-growth (BAI and TRW); tree-growth index (BAI<sub>i</sub> and TRW<sub>i</sub>); sap flow velocity; sap flow; soil water content; tree water use and stand transpiration; throughfall; stemflow; stable isotopes ( $\delta^{18}\text{O}$  and  $\delta^{13}\text{C}$ ) and intrinsic water use efficiency. A more detailed description of each statistical analysis carried out can be found in each chapter presented below.

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## **CHAPTER TWO**

Coupling daily transpiration modelling with forest management in a  
semiarid pine plantation

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## **Coupling daily transpiration modelling with forest management in a semiarid pine plantation**

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## **Abstract**

Estimating forest transpiration is of great importance for Adaptive Forest management (AFM) in the scope of predicted climate change. AFM in the Mediterranean usually generates a mosaic of different canopy cover within the same forest. A large diversity of models and methods to estimate forest transpiration have been developed, but most of them require a homogeneous forest cover, or an individual calibration/validation process per each cover stand. Hence, a model capable of reproducing accurately the transpiration of the whole canopy-cover mosaic is necessary. In this paper, the use of an Artificial Neural Network (ANN) is proposed as a flexible tool capable of estimating forest transpiration using the forest cover as an input variable. To that end, two years of sap flow, soil water content and other environmental variables were experimentally collected under 5 Aleppo pine stands of different canopy cover. Then, the ANN was modelled. Stand transpiration was accurately estimated using climate data, soil water content and forest cover through the ANN approach (correlation coefficient, R: 0.95; Nash-Sutcliffe coefficient, E: 0.90 and root-mean-square error, RMSE: 0.078mm/day). Finally, when the input value for

soil water content was not available, it was computed using the process-based model Gotilwa+. Then, this computed soil water content was used as input in the modelled ANN. This combination predicted the forest transpiration with values of R: 0.90, E: 0.63, and RMSE: 0.068 mm/day. Artificial Neural Network proved to be a useful and flexible model to predict the transpiration dynamics of an Aleppo pine stand regardless of the heterogeneity of forest cover produced by adaptive forest management.

**Keywords:** Adaptive forest management; Artificial neural network (ANN); Forest water-use; *Pinus halepensis* Mill.

## 1. Introduction

Transpiration is one of the most important components in the terrestrial water cycle, representing 80-90% of terrestrial evapotranspiration (Jasechko et al. 2013). In Mediterranean ecosystems it may account, on an annual basis, for about three-quarters of the overall forest evapotranspiration (Lawrence et al. 2007). Thus, its role in the water fluxes and within the soil-vegetation-atmosphere system is crucial to understand forest-water use (Asbjornsen et al. 2011) and to implement proactive adaptive measures in forests in the scope of global change (Fitzgerald et al. 2013, Ungar et al. 2013). This is particularly important in Mediterranean ecosystems, which have been identified as one of the areas that is most vulnerable to global climate change (IPCC 2007, Lindner et al. 2010, Vargas-Amelin and Pindado 2013).

Adaptive forest management (AFM) in Mediterranean ecosystems aims to link the ecophysiology and management of dryland forest via construction of an ecosystem-level water balance (Ungar et al, 2013). In the Mediterranean Spain, *Pinus halepensis* Mill. (Aleppo pine) reforestations occupy over 5x10<sup>5</sup> ha; where AFM may improve stand resilience and the balance between green and blue water budgets (Biro



& Gracia 2011, Molina & Del Campo 2012). Thus, prediction of forest transpiration under different forest management scenarios becomes a key element to design appropriate AFM.

Modelling transpiration the factors that influence it is quite complex, as nonlinear and complex interactions do exist (Asbjornsen et al. 2011). The fundamental controls are the available water in the soil, the ability of the plant to transfer water from the soil to its leaves and the ability of the atmosphere to absorb the transpired water (Davie 2008). Forest transpiration is estimated using different approaches. Drekker et al. (2000) reported that from all different types of process orientated forest transpiration models, four different perspectives are found: the cooling of leaves, the assimilation of CO<sub>2</sub>, the energy balance and the water balance, all showing a wide variation of physiological detail. The most widely used are those based on the energy balance, which are mostly derived from the Penman-Monteith equation. These models are commonly implemented in the Process Based Models (PBM). Nevertheless, these methods usually require a homogeneous forest cover, or an individual calibration/validation process for each cover stand. Since application of AFM usually leads to a mosaic of different forest cover

and structures within the same forest, a method capable of including this heterogeneity would be necessary.

Artificial Neural Network (ANN) may be a method capable of overcome this problem by including forest cover heterogeneity as an input variable. An ANN is a flexible mathematical structure, which is capable of identifying complex nonlinear relationships between input and output data sets. The ANN models have been found useful and efficient, particularly in problems for which the characteristics of the processes are difficult to describe using physical equations. These models are well suited to situations where the relationship between the input variable and the output is not explicit. ANN's map the implicit relationship between inputs and outputs through training by field observations (Shirgure 2013). Furthermore, ANN's usually obtain regression coefficients around 0.90, performing more accurately than other approaches such as statistical or PBM.

In recent years, ANN's have intensively been applied in forest and agriculture hydrology, e.g. estimating evapotranspiration (Kumar et al. 2002, Kumar et al. 2011, Adeloye et al. 2012, Huo et al. 2012), trunk sap flow (Liu et al. 2009) and transpiration (Zee 2001a, Zee 2001b, Vrugt et al. 2002, Garcia-Santos 2007, Meijun et al. 2007, Li et al. 2009).

In forest management ANN's have also commonly been applied to estimate tree volume (Gorgens et al. 2009, Silva et al. 2009, Diamantopoulou and Milios 2010, Özçelik et al. 2010, Yu & Jia-yin 2012, Özçelik et al. 2014), growth modelling (Castro et al. 2013), tree height (Binoti et al. 2013a), or to describe diameter distribution (Leite et al. 2011; Binoti et al. 2013b). However, none of these applications include forest management as an input variable, at least in the estimation of transpiration. This implementation would allow: i) extended capabilities and use of ANN-based modelling, ii) better determining and quantifying the influence of forest management practices on transpiration, iii) better adapting forests to new climate conditions by increasing their resilience.

Nevertheless, despite the above-mentioned ANN's advantages, its application sometimes becomes limited when some of the required input data are not easily available to forest managers. Under these circumstances a combination of ANN and a PBM, can be a useful alternative. Although their sophistication allow them, in principle, to reproduce the complex dynamics of forest ecosystems in detail, it makes also difficult their use and evaluation (Van Oijen et al. 2005). Hence,

PBM could be used in some cases to feed the ANN's with the estimate of those required input data not readily available.

The main objective of this work is to develop a reliable model for estimating transpiration in forest stands considering explicitly the influence of forest management as an input variable. This general objective derives into the following specific objectives: i) to explore the relationships between measured forest transpiration and explicative environmental variables in order to define the most reliable empirical model (Linear Models and ANN-based approaches); ii) to incorporate forest cover as a forest management-derived variable into the selected modelling approach and to evaluate its predictive ability; iii) to study the model performance when soil moisture needs to be derived from other modelling approaches (Gotilwa+).

## **2. Material and Methods**

Sap flow, soil water content and other environmental variables were registered during 2 years in 5 Aleppo pine stands, with different canopy cover. Then, with the aim to analyse the performance of Multiple Linear Regression (MLR) and General Linear Model (GLM) in transpiration estimation, both models were developed and validated

using the field data. Subsequently, the ANN was modelled and used to estimate the stand transpiration using climate data, soil water content and forest cover as input variables. Finally, when the input value soil water content is not available, it was computed using the process-based model Gotilwa+. Then, this computed soil water content was used as an input value in the modelled ANN.

## **2.1 Experimental site and empirical data obtaining**

The experimental site and design have been thoroughly described in Molina and Del Campo (2012). Briefly, the study area is a public forest of 4 682 ha located in the Ayora valley region (39° 5′ N; 1° 13′ W, 943 m a.s.l.), in the Southwest of Valencia province (Spain). The climate is Mediterranean with an average total annual rainfall of 478 mm and a mean annual temperature of 13.7 °C (1960-2010). Soils are Calcisols derived from Triassic limestone (gravel and boulders) with high percentage of carbonates (26-38%, pH 7.7-8.2) and of sandy-silty loam texture. Twenty two percent of the area is occupied by *Pinus halepensis* Mill. plantations ranging between 50-60 years old and with high tree density due mainly to low forest management.

In 2008, an experimental thinning was performed in a plantation patch in 30x30 m plots, reducing the forest cover from 84% (C - control plot) to 22% (H - high intensity plot), 50% (M - medium intensity plot) and 68% (L - low intensity plot). In addition, a nearby area thinned in 1998 (H98) with 41% cover was considered in this study as the temporal evolution of the high intensity treatment (showing no significant differences in number of tree per ha -  $p < 0.05$ ). This fact introduces more complexity in the study as different tree densities (in addition to different covers) are considered (Table 1). Hence, five plots are considered in this work.

Table 1 - Characteristics of control and thinned plots. DBH: diameter at breast height; H<sub>98</sub>: the plot thinned in 1998 (rest thinned in 2008, except the control plot). Adapted from Molina and Del Campo (2012)

Thinning Treatment	Forest Cover (%)	Density (trees ha <sup>-1</sup> )	DBH (cm)	Height (m)
Control (C)	84	1489	17.8 ± 5.1	11.5
Low intensity (L)	68	744	21.2 ± 4.1	12.2
Medium intensity (M)	50	478	21.7 ± 4.0	11.3
High intensity (H)	22	178	20.4 ± 1.6	12.2
High intensity-1998 (H <sub>98</sub> )	41	155	25.2 ± 5.0	12.6

For the present study, several hydrological (transpiration and soil moisture) and environmental variables: air temperature, T, °C; solar radiation, Sr, MJ m<sup>-2</sup> day<sup>-1</sup>; rainfall, P<sub>pt</sub>, mm; relative humidity, RH, %;

and wind speed,  $W_s$ ,  $m\ s^{-1}$ , were collected from June 2009 to March 2011 (Table 2). Measurements of Ppt, T, and RH were carried out by a single sensor (HR/T sensor, Decagon Devices, Pullman, USA) placed at 1 m above the ground and close to the treatment plots. Values were recorded and averaged every 30 minutes. Data were afterwards used to obtain values for mean, maximum and minimum daily temperature and vapour pressure deficit (VPD) (Allen et al. 1998). The daily values for solar radiation, and wind speed were obtained from the Almansa weather station, located near the study site. Transpiration was measured by means of sap flow sensors (HRM-30, ICT International Pty Ltd., Armidale/Australia) (Burgess et al. 2001) in four trees per plot according to the frequency distribution of diameters. To convert the heat pulse velocity to sap flow velocity, raw values were corrected for probe misalignment (Burgess et al. 2001), differences in thermal diffusivity and wounding, by examining samples under light microscopy (Barret et al. 1995). Baseline correction in the obtained series was performed according to Buckley et al. (2012). The accumulated daily values of sap flow ( $l\ day^{-1}$ ) (water transpired by the entire tree) were estimated considering a radial distribution of the sap flux velocity in the sapwood of each selected tree (Delzon et al. 2004). The sapwood area per tree was

measured after extracting samples with an increment core (5 mm) and measuring to the nearest 0.01 mm with a measuring table (LINTAB 6.0, Frank Rinn, Heidelberg, Germany) coupled with the TSAP-Win software package (Rinn 2011). Total transpiration by the pine forest ( $Tr$ , mm day<sup>-1</sup>) per treatment, was calculated by multiplying the daily transpiration (L day<sup>-1</sup> tree<sup>-1</sup>) by the frequency of diameter class ( $f$ ) and the stand density (trees ha<sup>-1</sup>), using Equation (1):

$$Tr(\text{mm day}^{-1}) = \frac{\sum(Tr_n * f_n) * N^o}{10000} \quad (1)$$

Where:  $Tr_n$  is the daily transpiration of each treatment;  $f_n$  is the frequency of each tree class measured;  $N^o$  is the number of trees by stand (Table 1) and division by 10 000 is the transformation of liters/ha into mm.

Soil water content (SWC, cm<sup>3</sup> cm<sup>-3</sup>) was measured every 20 minutes by means of capacitance sensors, and averaged to a daily value, (Echo, Decagon Devices Inc., Pullman, WA). In each treatment, 3 sensors were placed under the crown-projected area at 30 cm depth under 2 randomly selected trees out of the 4 sampled for transpiration. The sensor daily values were individually calibrated, averaged and converted



into one daily value per treatment. Table 2 shows a summary of the different data available for the present study.

The medium intensity treatment presented numerous data gaps in most variables (including transpiration and soil moisture); for this reason it was considered as the better choice to test the model performance when fed with PBM derived variables (objective iii). Hence this treatment was discarded for model building, evaluation and validation either with linear or ANN techniques.

Table 2 - Mean  $\pm$  standard deviation values of the different registered data in each treatment (years 2009-2011). N: number of suitable days for analysis; Tr: Transpiration ( $\text{mm day}^{-1}$ ); SWC: average daily soil water content ( $\text{cm}^3 \text{ cm}^{-3}$ ); Ppt: precipitation (mm);  $T_{\text{max}}$ ,  $T_{\text{min}}$  and  $T$  are maximum, minimum and mean temperature respectively ( $^{\circ}\text{C}$ ); RH: relative humidity; VPD: vapour pressure deficit (kPa); Ws: wind speed ( $\text{m s}^{-1}$ ); Sr: solar radiation transformed in equivalent evaporation\* ( $\text{mm day}^{-1}$ )

Variable	Thinning intensity	2009	N	2010	N	2011	N	
Tr	High	0.44 $\pm$ 0.26	187	0.34 $\pm$ 0.20	309	0.14 $\pm$ 0.10	90	
	High-1998	0.29 $\pm$ 0.15		0.22 $\pm$ 0.13	128	-		
	Medium	-	-	0.19 $\pm$ 0.12	99	0.13 $\pm$ 0.09		
	Low	0.31 $\pm$ 0.18	187	0.38 $\pm$ 0.26	309	0.19 $\pm$ 0.14		
	Control	0.32 $\pm$ 0.26		0.47 $\pm$ 0.36		0.25 $\pm$ 0.19		
SWC	High	0.19 $\pm$ 0.06		0.26 $\pm$ 0.06		0.28 $\pm$ 0.02		
	High-1998	0.11 $\pm$ 0.04		0.19 $\pm$ 0.03		128		0.20 $\pm$ 0.03
	Low	0.13 $\pm$ 0.07		0.19 $\pm$ 0.06		309		0.19 $\pm$ 0.04
	Control	0.15 $\pm$ 0.07	0.22 $\pm$ 0.06	0.22 $\pm$ 0.03				
$T_{\text{max}}$	-	23.2 $\pm$ 9.42	20.1 $\pm$ 8.94	11.0 $\pm$ 4.33				
$T_{\text{min}}$	-	11.2 $\pm$ 6.08	8.5 $\pm$ 5.91	2.3 $\pm$ 3.43				
$T$	-	16.9 $\pm$ 7.52	13.9 $\pm$ 7.16	6.1 $\pm$ 3.22				
RH	-	0.6 $\pm$ 0.20	0.7 $\pm$ 0.15	0.8 $\pm$ 0.12				
VPD	-	1.0 $\pm$ 0.88	0.7 $\pm$ 0.59	0.3 $\pm$ 0.17				
Ppt	-	1.5 $\pm$ 4.32	1.8 $\pm$ 4.67	1.9 $\pm$ 5.06				
Ws	-	2.3 $\pm$ 1.32	2.3 $\pm$ 1.30	2.4 $\pm$ 1.53				
Sr	-	7.7 $\pm$ 3.81	7.2 $\pm$ 3.35	4.8 $\pm$ 2.11				

\* Solar radiation expressed in  $\text{MJ m}^{-2} \text{ day}^{-1}$  was converted to equivalent evaporation in  $\text{mm day}^{-1}$  by using a conversion factor equal to the inverse of the latent heat of vaporization ( $1/L = 0.408$ ) (Allen et al., 1998).

## **2.2 Transpiration modelling**

### **2.2.1 Multiple Linear Regression (MLR) and General Linear Models (GLM)**

Despite the fact that neural networks are widely considered as a powerful tool in the identification of highly non-linear systems, as it is the case of the target variable in this research, a more traditional approach based on multiple linear regression (MLR) has been also carried out, as it might provide an interesting modelling benchmark using appropriate final performance index (see section 2.4). Not only the classical, well known MLR approach was used, but also a linear regression implemented through a simplified neural network with only two layers and linear nodes, which will be referred in the following as general linear model (GLM) (Özesmi et al. 2006). These two models (MLR and GLM) represent adequate benchmarks to evaluate the relative merits and performance of neural network approach (Özesmi et al. 2006, Liu et al. 2009). Both methods have individually been applied to each of the treatments with the different forest cover, and also to the totality of data pooled together, but including forest cover as an input variable. In all cases, the target variable to be predicted was daily transpiration.

Concerning input variables, a correlation analysis and a stepwise method was performed to assess variable selection process, which is common to both mentioned approaches (MLR and GLM).

### **2.2.2. Transpiration based on Artificial Neural Network**

Following Gorgens et al. (2009), artificial neural network (ANN) modelling comprises three stages: pre-processing (including variable selection, data division and data scaling and normalization), processing (ANN architecture and network training process) and post-processing or model evaluation. In each of the mentioned stages, the following criteria were used:

#### ***Stage 1: Data pre-processing***

An adequate selection of relevant inputs for the ANN strongly conditions the final performance of the model, in particular when there are known interrelationships between predictor variables. According to previous researches (Liu et al. 2009), the most relevant variables that determine transpiration rates are known physical variables, i.e., VPD, Sr, RH, T, Ws, LAI. It is clear that each of these variables is an active variable influencing in some extension the final transpiration rates, although the physical mechanisms involving groups and subgroups of

variables acting simultaneously are complex non-linear processes. This fact makes it difficult the choice of optimal predictor variables. For this purpose, a sensitivity analysis was conducted, in order to avoid input variables combinations that might yield to an ill-conditioned system (Smith 2006). Different groups of input variables were tested in order to find the best combinations for improved transpiration rates predictions.

A first issue when using ANN modelling strategies is the division of data into training and test sets, as it can significantly affect the selection of optimal ANN structure and the evaluation of its forecasting performance (Zhang et al. 1998). Sometimes a third sample usually named the cross-validation set is also used to avoid the overtraining problem or to determine the stopping point of the training process. Although there is not a clear guidance for selecting the size of such subgroups, we have applied a commonly used criterion, using a training set size of 70% of the data, and reserving the rest for test and cross-validation sets (15% and 15% respectively).

Obviously, the use of representative subsets is of significant importance for the performance of the neural network and its final generalization capacity. To account for it, and for each of the treatments, transpiration data was previously classified into four classes, with class

limits set at  $(\mu-\sigma)$ ,  $\mu$  and  $(\mu+\sigma)$  where  $\mu$  is average value and  $\sigma$  is the standard deviation. For each of the classes, the data were then classified considering the VPD range and the different seasons separately. Finally, a *t-test* statistic to check for sample mean differences was applied over the training, test and cross validation sets, finding no significant statistical differences between sample means ( $p<0.05$ ).

The variables under consideration in this research span different ranges. In order to ensure that all of them receive equal attention during the ANN training process, it is important to scale them. To do so, the original data were transformed to the interval [0;1], according to equation 2, with the exception of two variables that are mentioned below.

$$x_{\text{norm}} = 0.5 * \left( \frac{x_0 - \bar{x}}{x_{\text{max}} - x_{\text{min}}} \right) + 0.5 \quad (2)$$

Where  $x_{\text{norm}}$ = normalized value;  $x_0$ = original value;  $\bar{x}$  = average value of original series  $x_{\text{max}}$  = maximum value;  $x_{\text{min}}$ = minimum value.

In the case of forest cover, no transformation is needed, as original values already lie in the interval [0;1]. The second exception is the VPD variable. In neural network modelling, the probability distribution of the input data does not have to be strictly known (Burke & Ignizio 1992), although in some cases a skew reduction in the original

data can be desirable, if it is very significant. That is the case of the variable VPD in our research, which was transformed with an exponential function (eq. 3).

$$x_{\text{norm}} = 1 - e^{-\bar{x} * x_0} \quad (3)$$

Obviously the direct output of the neural network (transpiration) is a scaled or normalized value, which needs to be reconverted to its original range before proceeding to the model performance evaluation procedure (Taylor & Smith 2006).

### ***Stage 2: ANN architecture and training algorithm***

A feed forward artificial neural network (FFNN) is proposed in this research as a mathematical model to predict transpiration rates. A FFNN consists of a set of sensory units that constitute the input layer, one or more hidden layers of computational nodes and an output layer of computational nodes producing the output of the network (Haykin 1994). Information passes only in one direction, from the input nodes to the ones in succeeding layers until it reaches the output layer. Strength of connexions between nodes of successive layers is represented by the weight values, acting as parameters of the neural network model. The output layer has as many nodes as target variables. In our case, it will

include only one node, corresponding to the only target variable, i.e., daily transpiration. Concerning the number of hidden layers, there is actually no consolidated theory to tell how many hidden layers are needed, although it has been proved that only one layer of hidden units with non-linear nodes is sufficient to approximate any function to an arbitrary level of precision (Hornik et al. 1989). According to it, a topology of three layers with a single output node in the third layer is proposed. The number of nodes in the input layer, “ $i$ ”, is the number of predictor variables used. A problem in FFNN building arises concerning the specification of the number of nodes required in the hidden layer, being clear that the more complex the mapping between variables is, the larger the number of hidden nodes. In our case, a range of neural networks were tested varying the number of hidden nodes from  $i/2$  to  $2i$ , in order to select the optimal dimension of the network, appropriate to the complexity of the problem. In a similar way, a family of activation functions in the hidden and output layer were tested (logistic, hyperbolic tangent, and exponential for the hidden layer; exponential and identity function in the output node) (Vrugt et al. 2002, Meijun et al. 2007, Liu et al. 2009).



Concerning the training algorithm, it can be addressed as an unconstrained nonlinear minimization problem in which weight values are iteratively modified to minimize the overall mean squared error between the desired and actual output values of scaled transpiration. There are a variety of powerful algorithms in the literature, but none of them can guarantee in practice the global optimal solution for a general nonlinear optimization problem, with efficiency and computational performance depending largely on the structure and characteristics of the unknown error function. In most practical applications, the faster convergence and more efficient methods are the second order methods like Broyden, Fletcher, Goldfarb and Shanno (BFGS), Levenberg-Marquardt and conjugate gradient methods (Zhang et al. 1998). In this research, a powerful training algorithm is required, as many networks are to be tested with different topologies and activation functions. Satisfactory results were obtained employing only the BFGS algorithm, due to its faster training speeds and better approximation and generalization ability. (Vojislav 2001, Igel & Hüsken 2003, Nawi et al. 2006, Jing-Hua et al. 2010). In general, network configuration involving weights with very large values are not desirable. For this reason, a regularization weight decay procedure (Bishop 1995) was incorporated

in the training process. This method essentially consists in adding a term to the error function penalizing solutions with large weight values.

### ***Stage 3: Model evaluation***

The prediction accuracy of observed transpiration values can be measured using a variety of metrics, assessing thus the performance of the different models tested. Usual statistics have been computed, including R, E and RMSE (see section 2.4), no matter the type of model under examination.

## **2.3 Process Based Model**

In the present study, the PBM GOTILWA+ was applied to estimate SWC that might not be readily available to forest managers. GOTILWA+ is an improved version of GOTILWA described in Gracia et al. (1999) and Kramer (2001). It can estimate accurately the water flux of Mediterranean forest ecosystems (different single-tree species stands: coniferous or broad leaved, evergreen or deciduous) under changing environmental conditions, due either to climate or to forest management. For this reason, it was used here with the purpose of estimating both transpiration (directly) and soil water content (to feed the ANN). The model requires a number of inputs to simulate forest growth parameters

at a daily time scale. The inputs describe the forest structure and physiology, soil and climate conditions. The forest structure was considered heterogeneous, not in terms of forest cover but diametrical distribution, which was defined according to the initial tree inventory. Leaf photosynthesis and stomata conductance parameters for *Pinus halepensis* were included in the GOTILWA+ model, based on measured data from experimental plots in Collserola (Catalonia) (<http://www.creaf.uab.es/gotilwa+/SPParameters.htm>). Soil parameters were obtained from previous works (Del Campo et al. 2008). Daily climatic data were obtained as previously described in this section. The GOTILWA+ performance was analysed comparing measured and estimated transpiration and soil moisture values, respectively, considering the parameters of model evaluation (see point 2.4).

## **2.4 Model evaluation**

For inter-comparisons between the measured and simulated transpiration using the ANN model, multiple-linear regression (MLR), general linear models (GLM) and GOTILWA+, three parameters were used to evaluate the model, i.e. Coefficient of correlation (R); Nash-Sutcliffe Efficiency (E) that compares the performance of the model to a

model that only uses the mean of the observed data. A value of 1 indicates a perfect model, while zero indicates a performance no better than simply using the mean, and negative values indicate even worse performance. (Bennett et al. 2013); and root mean squared error (RMSE) - that is an effective measure of deviation of model predictions from the observed data (Whitley et al. 2012).

$$R = \frac{\sum_{i=1}^n (Y_{obs_i} - \bar{Y}_{obs})(Y_{sim_i} - \bar{Y}_{sim})}{\sqrt{\sum_{i=1}^n (Y_{obs_i} - \bar{Y}_{obs})^2} \sqrt{\sum_{i=1}^n (Y_{sim_i} - \bar{Y}_{sim})^2}} \quad (4)$$

$$E = 1 - \left( \frac{\sum_{i=1}^n (Y_{sim_i} - Y_{obs_i})^2}{\sum_{i=1}^n (Y_{obs_i} - \bar{Y}_{obs})^2} \right) \quad (5)$$

$$RMSE = \sqrt{\frac{\sum_{i=1}^n (Y_{obs_i} - Y_{sim_i})^2}{n}} \quad (6)$$

Where: n represent the number of data considered,  $Y_{obs_i}$  represents the daily transpiration observed by heat pulse method for the  $i^{th}$  value;  $Y_{sim_i}$  is the simulated transpiration for the  $i^{th}$  value,  $\bar{Y}_{obs}$  and  $\bar{Y}_{sim}$  represent the average values of corresponding variable. In addition, a graphical analysis was used to compare and verify presence of heteroscedasticity.

### **3. Results**

#### **3.1. Linear analysis**

Empirical linear correlations between input variables and transpiration are quantitative indicators of the degree of statistical linear dependence among them. Such values are shown in Table 3. It should be noted that correlations differ significantly among the different treatments (plots) considered in this research. Values tend to be generally higher for the case of intensive thinning, while the lowest ones correspond to the case of control treatment. It can be observed that expected high values in case of strongly physically interrelated variables (i.e., VPD and transpiration), can be as low as 0.29 for the control treatment, indicating the extent to which the type of treatment can in practice affect actual relationships between variables and quantitative patterns of the transpiration data. Other variables, such as wind speed, precipitation and soil water content, present negative and modest correlations for individual treatments and also for the grouped data. From these results, it follows that some variables do not contribute significantly to explain transpiration (e.g.  $W_s$ , which is no further considered), whereas for other

variables, co-linearity makes advisable to consider only those with higher correlations (the case of RH with Ppt).

Table 3 - Linear correlation matrix between all input variables and daily transpiration (mm) per treatment.  $T_{\max}$ ,  $T_{\min}$  and  $T$  are maximum, minimum and mean temperature respectively ( $^{\circ}\text{C}$ ); RH: relative humidity; VPD: vapour pressure deficit (kPa); Ppt: precipitation (mm); Ws: wind speed ( $\text{m s}^{-1}$ ); Sr: solar radiation transformed in equivalent evaporation ( $\text{mm day}^{-1}$ ); SWC: average daily soil water content ( $\text{cm}^3 \text{cm}^{-3}$ ).

Treatment	$T_{\max}$	$T_{\min}$	T	RH	VPD	Ppt	Ws	Sr	SWC
High intensity	0.86	0.75	0.84	-0.76	0.82	-0.32	-0.17	0.85	-0.35
High intensity-1998	0.82	0.69	0.79	-0.75	0.73	-0.47	-0.27	0.76	-0.31
Low intensity	0.58	0.45	0.55	-0.53	0.46	-0.28	-0.16	0.64	0.03
Control	0.42	0.30	0.38	-0.40	0.29	-0.25	-0.14	0.53	0.22
All grouped	0.59	0.47	0.56	-0.53	0.49	-0.29	-0.16	0.63	0.02

Linear based models (MLR and GLM) were developed for transpiration estimation, both for each individual treatment and for the grouped treatments. In total, 10 different models were formulated, varying essentially the family of input variables considered. For each of the models, R, RMSE and E values have been computed for evaluation purposes (Table 4). A first observation that can be derived from such values is the similar performance of both approaches. This was an expected result, as both models comprise essentially a linear regression between inputs and output, although formally applied in a different

manner. On the other hand, it can be observed that the type of treatment is affecting significantly the prediction ability of the models, with E coefficient ranging from 0.63 to 0.84 (Table 4). Fig. 1 shows estimated vs. observed values of transpiration by MLR. Although results can be reasonably acceptable, it is noticeable the fact that for larger values of transpiration, a systematic underestimation error occurs, no matter the treatment under consideration (Fig. 1a-d). Such limitation of the linear models was investigated, and several attempts were made to improve results.

Table 4 - Results of Multiple Linear Regression (MLR) and General Linear Method (GLM) for each treatment. R: correlation coefficient; RMSE: root mean squared error; E: Nash-Sutcliffe efficiency.  $T_{max}$ ,  $T_{min}$  and T are maximum, minimum and mean temperature respectively; RH: relative humidity; VPD: vapour pressure deficit; Sr: solar radiation; SWC: soil water content; Cover: Forest cover.

Model	Treatment	R	RMSE	E	Inputs
MLR	High intensity	0.91	0.093	0.84	$T_{max}$ , T, Sr, RH, SWC,
	High intensity-1998	0.87	0.071	0.76	$T_{max}$ , $T_{min}$ , Sr, RH, VPD, SWC
	Low intensity	0.81	0.135	0.66	$T_{max}$ , $T_{min}$ , T, Sr, RH, VPD, SWC
	Control	0.79	0.194	0.63	$T_{max}$ , T, Sr, RH, VPD, SWC
	All grouped	0.77	0.161	0.59	$T_{max}$ , T, Sr, RH, VPD, SWC, Cover
GLM	High intensity	0.91	0.093	0.84	$T_{max}$ , $T_{min}$ , T, Sr, RH, VPD, SWC
	High intensity-1998	0.87	0.071	0.76	$T_{max}$ , $T_{min}$ , T, Sr, RH, VPD, SWC
	Low intensity	0.81	0.135	0.66	$T_{max}$ , $T_{min}$ , T, Sr, RH, VPD, SWC
	Control	0.79	0.193	0.63	$T_{max}$ , $T_{min}$ , T, Sr, RH, VPD, SWC
	All grouped	0.75	0.166	0.56	$T_{max}$ , $T_{min}$ , T, Sr, RH, VPD, SWC, Cover

The inclusion of an input variable related to forest management, i.e. forest cover, did not improve the model performance in either case (GLM nor MLR – All grouped, Table 4), but it decreased both the Nash-Sutcliffe ( $E < 0.6$ ) and the R coefficients, and it worsened the predictive ability for the higher values of transpiration (Fig. 1e,f). To all data grouped, the stepwise procedure of MLR showed better results than GLM, although it identified different inputs according to the treatment (e.g.  $T_{\min}$  was used as input only in High intensity-1998 and Low intensity thinning treatment) Table 4.



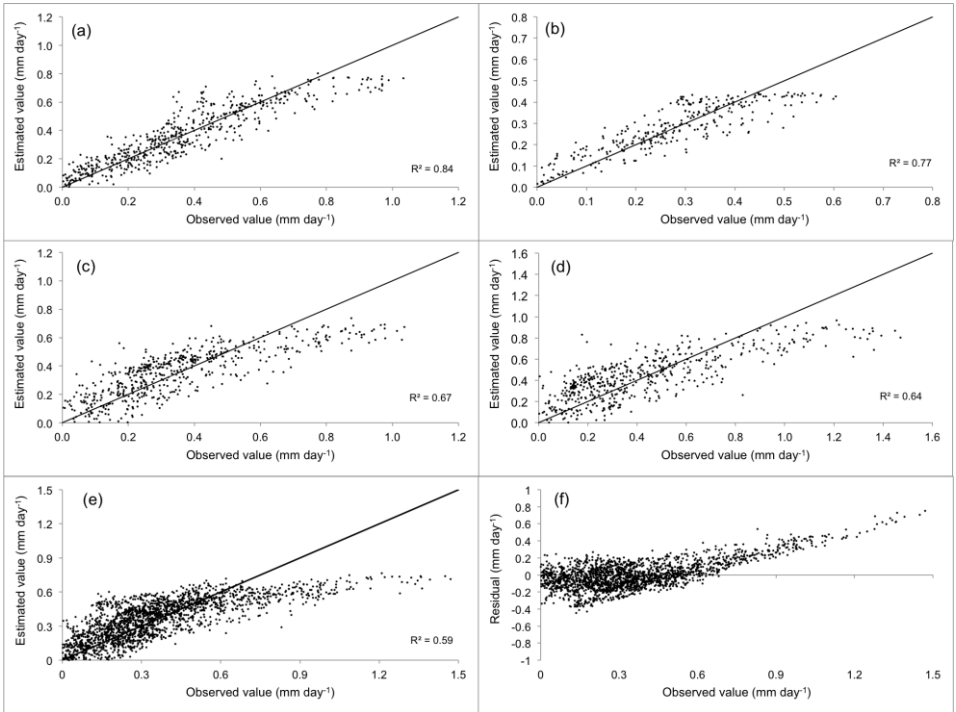


Fig. 1 - Observed vs. estimated values of stand transpiration by Multiple Linear Regression. a – High intensity; b – High intensity 1998; c – Low intensity; d – Control; e – All treatments grouped. f – Residual values for all treatments grouped

### 3.2. Transpiration based on ANN

An initial sensitivity analysis has been carried out to identify importance of the different predictor variables involved in transpiration estimation. The error function showed the greatest increase when SWC (soil moisture) was not taken into account, and thus, it can be considered a key variable in the input family. The two subsequent variables that

most affected the prediction error were  $S_r$  and  $T_{\max}$ . All the neural networks tested included at least those three variables, and then, following a stepwise procedure, increasing number of inputs were added to the network tested, incorporating other possible predictor variables. Best results are obtained when the input family consists of variables  $T_{\max}$ ,  $T_{\min}$ ,  $T_{\text{mean}}$ ,  $S_r$ , RH, VPD, SWC and Cover, as indicated in Table 5. The number of hidden nodes ( $n_h$ ) was found to be optimal for the network 8-9-1, and thus,  $n_h=9$  was selected. Fig. 2 shows the values of E and RMSE resulting after trained networks with different size of the hidden layer. Concerning the activation function, best results were obtained using the hyperbolic tangent function in the hidden nodes and exponential function in the single node of the output layer.

Table 5 - Inter-comparison between measured and simulated transpiration for each treatment, using the selected Artificial Neural Network (8-9-1). Last row represents the ANN general performance. R: correlation coefficient; RMSE: root mean squared error; E: is Nash-Sutcliffe Efficiency.  $T_{max}$ ,  $T_{min}$  and T are maximum, minimum and mean temperature respectively; RH: relative humidity; VPD: vapour pressure deficit; Sr: solar radiation; SWC: soil water content; Cover: Forest cover.

Treatment	R	RMSE	E	Inputs
High intensity	0.96	0.065	0.92	Tmax, Tmin, T, Sr, RH, VPD, SWC, Cover
High intensity-1998	0.91	0.060	0.82	
Low intensity	0.94	0.081	0.88	
Control	0.96	0.092	0.91	
ANN	0.95	0.077	0.90	

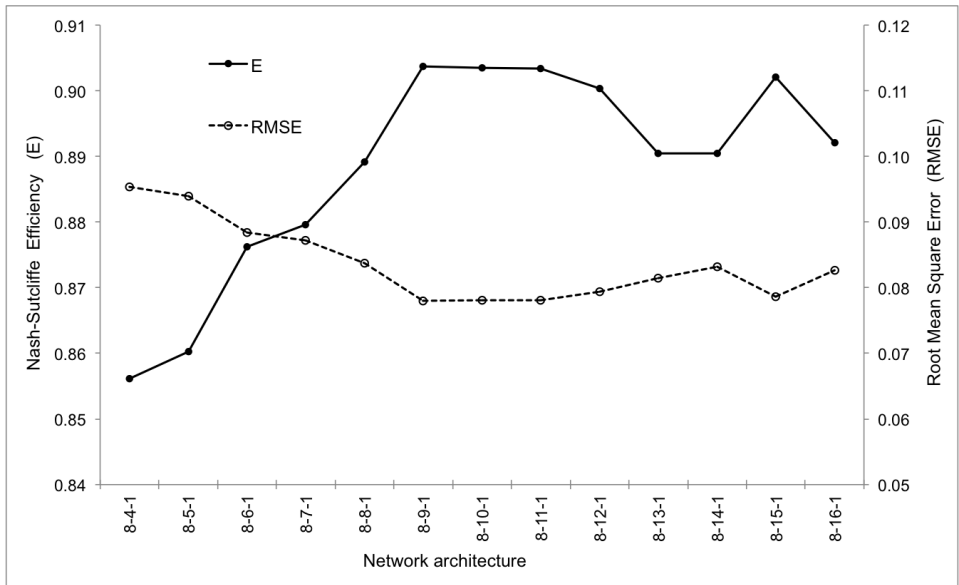


Fig. 2 - Evolution of Nash-Sutcliffe efficiency (E) and root mean square error (RMSE) in each architecture of artificial neural network tested

Fig. 3 shows the transpiration estimated values versus the observed ones, in  $\text{mm day}^{-1}$ . Results are satisfactory, with E values ranging from 0.82 to 0.92. It should be noted here that quality of results does not differ significantly when considering the network with pooled data in Fig. 3e (all treatments grouped) or when networks are used separately for each treatment (specific cover) in Fig. 3a-d. Table 6 shows the average error obtained after transpiration estimation in different seasons. It should be noted that significant differences in model performance can be identified depending on the season considered, with worst results corresponding to autumn (Fig. 4). Additionally, it is clear that estimation errors are larger for higher values of transpiration, as can be derived from the residuals analysis (Fig. 3f). A more detailed discussion of these results will be presented in next section.

Table 6 - Average of observed (Obs.) and estimated (Est.) transpiration ( $\text{mm day}^{-1}$ ) using Artificial Neural Network per season and per treatment. Difference represents the average error per season and per treatment.

Season	N	High intensity		High intensity-98		Low intensity		Control		Difference ( $\text{mm day}^{-1}$ )
		Obs.	Est.	Obs.	Est.	Obs.	Est.	Obs.	Est.	
Summer-2009	284	0.166	0.156	0.097	0.101	0.082	0.077	0.077	0.072	0.016
Autumn-2009	352	0.089	0.084	0.069	0.057	0.086	0.080	0.095	0.085	0.034
Winter-2009-10	180	0.029	0.022	0.021	0.020	0.031	0.026	0.034	0.031	0.015
Spring-2010	321	0.087	0.086	0.060	0.069	0.120	0.115	0.156	0.158	-0.005
Summer-2010	300	0.164	0.169	0.028	0.027	0.174	0.168	0.202	0.209	-0.005
Autumn-2010	273	0.094	0.100			0.084	0.101	0.102	0.106	-0.027
Winter-2010-11	270	0.037	0.045			0.052	0.060	0.066	0.071	-0.021
Spring-2011	93	0.051	0.057			0.067	0.070	0.091	0.093	-0.011
Difference ( $\text{mm day}^{-1}$ )		-0.0020		0.0015		-0.0010		-0.0022		-0.0037

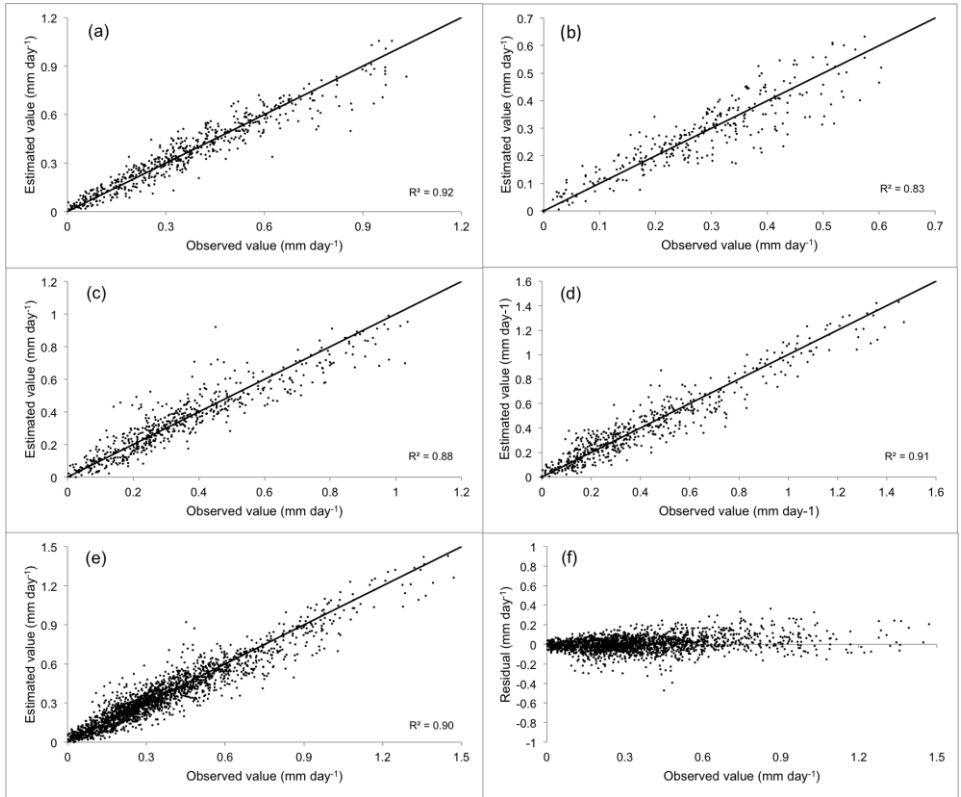


Fig. 3 - Observed vs. estimated values of stand transpiration by ANN. a – High intensity; b – High intensity 1998; c – Low intensity; d – Control; e – All treatments grouped. f – Residual values for all treatments grouped

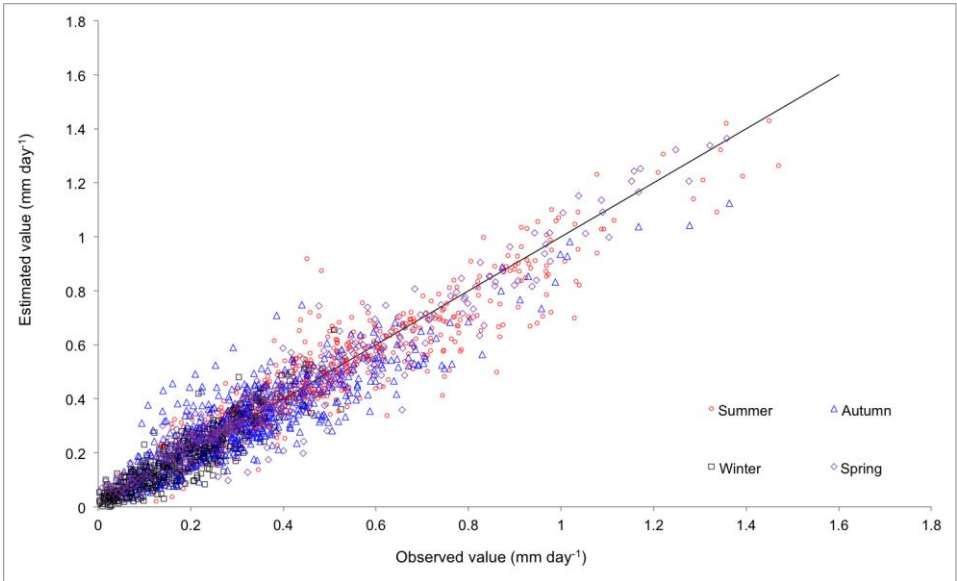


Fig. 4 - Observed vs. estimated values of stand transpiration by ANN with indication of season: winter (black square), spring (purple rhombus), summer (red circle) and autumn (blue triangle)

### 3.3. Use of the PBM Gotilwa+

The use of PBM was considered here to estimate input variables not readily available such as SWC. However, Gotilwa+ can also predict transpiration directly from the parameters introduced in the model. Gotilwa+ predicted SWC acceptably well (E from 0.47 to 0.68), although the estimated values for transpiration revealed a poor predictive ability of the model, except for the high intensity thinning treatment (E=0.57) (Table 7). Comparing ANN and GOTILWA+ performance in terms of

transpiration, ANN predicted the stand transpiration more accurately than GOTILWA+ (Table 7). The performance of this ANN-GOTILWA+ combination was validated comparing measured and estimated transpiration values of control, high, low and high 1998 thinning intensities (Table 7). Despite that the combination performed worse than ANN, the predicted transpiration values were still accurate (E ranging from 0.52 to 0.77), justifying its further use to estimate stand transpiration when SWC data is not available. Hence these results support our hypothesis of using the PBM to feed the ANN as a useful tool to estimate the stand transpiration when some of the ANN input data are not readily available. In this sense, ANN-GOTILWA+ was used to predict transpiration values of the medium thinning intensity treatment, where SWC data were not available. The results were validated using the available recorded data of transpiration, showing that ANN-GOTILWA+ predicted accurately the transpiration, significantly increasing the E value from -6.99 (Gotilwa+) to 0.63 (ANN-GOTILWA+), (Fig. 5 and Table 7).



Table 7 - Inter-comparison between observed and estimated Soil Water Content (SWC) using Process-based Model (GOTILWA+). Observed and estimated transpiration (Tr) using GOTILWA+, Artificial Neural Network (ANN) selected and the combination ANN-GOTILWA+(SWC) is also compared. R: coefficient of correlation; RMSE: Root Mean Squared Error; E: Nash-Sutcliffe Efficiency.

Treatment	Parameters	SWC: Gotilwa+	Tr: Gotilwa+	Tr: ANN	Tr: combination ANN+GOTILWA+
High	R	0.8	0.8	0.96	0.95
	RMSE	0.002	0.152	0.065	0.112
	E	0.63	0.57	0.92	0.77
High - 98	R	0.70	0.73	0.91	0.91
	RMSE	0.037	0.130	0.060	0.082
	E	0.47	0.19	0.82	0.68
Low	R	0.86	0.68	0.94	0.9
	RMSE	0.036	0.427	0.081	0.124
	E	0.68	-2.38	0.88	0.71
Control	R	0.84	0.79	0.96	0.85
	RMSE	0.039	0.463	0.092	0.220
	E	0.64	-1.11	0.91	0.52
Medium	R	-	0.87	-	0.90
	RMSE	-	0.318	-	0.068
	E	-	-6.99	-	0.63

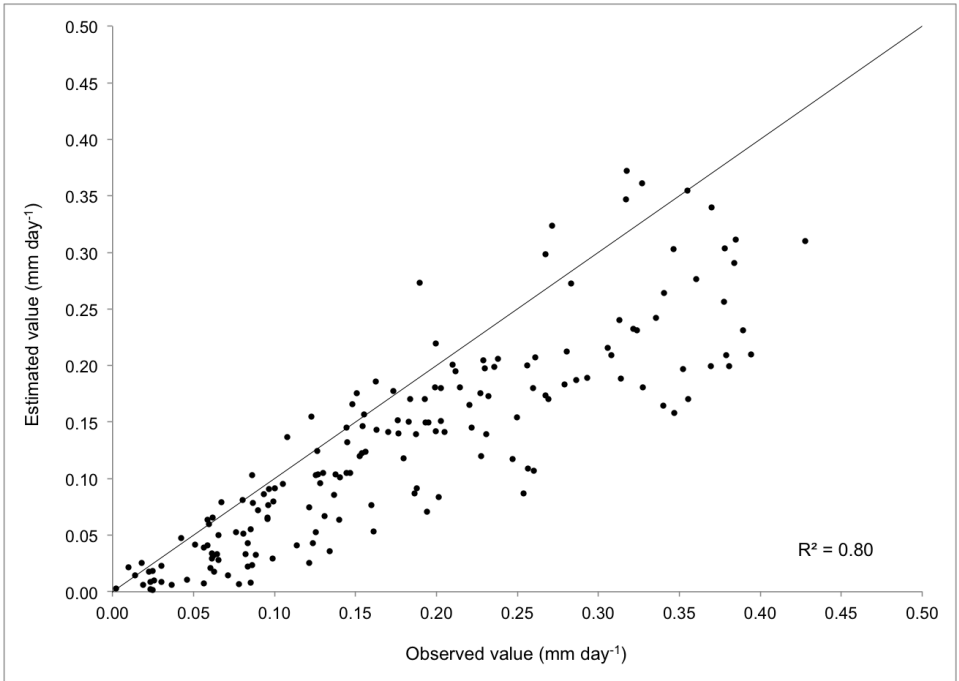


Fig. 5 - Observed vs. estimated values of stand transpiration in the medium intensity thinning treatment when the soil water content (SWC) was generated by Gotilwa+

#### 4. Discussion

The influence of AFM on different transpiration modelling approaches in a semiarid pine plantation has been studied in this work. The linear approach identified the inputs SWC, T, Sr, RH and VPD as those that have influence in forest transpiration, as stated by Asbjornsen et al. (2011). It yielded acceptable performance when considering isolated treatments. Other works (Lagergren and Lindroth 2002, Liu et al.

2009, Ungar et al. 2013) reported similar R values when predicting transpiration with linear models. However, it was observed here that, as forest cover increases, the model performance decreases. It is probably due to the stand competition as a primary determinant of transpiration over the environmental variables. The inclusion of forest cover in the grouped linear model worsened its performance. The results from this analysis indicate that, even under identical site and climatic conditions, input variables for modelling stand transpiration may differ according to cover, thus reducing considerably their practical use in operational forest management. The common outcome in these cases is the development of specific models in response to threshold behaviour in environmental drivers (Lagergren and Lindroth 2002, Mackay et al. 2012, Ungar et al. 2013), such as soil moisture or forest cover as in our case. This fact would complicate the practical applicability of the models beyond the specific conditions where they were developed.

The artificial neural network modelling approach showed better performance than any of the two linear models applied for transpiration estimation, MLR and GLM. In particular, neural network modelling showed an improved capability to predict higher values of transpiration, giving E values well over 0.82. Nevertheless, quality of predictions is not

the same for all seasons. In particular, the performance of the model for autumn is not as good as it is for the rest of the year. In the cases when larger prediction errors occurred, they can be partially explained by the variability of weather conditions along the day. However, this daily variability is not considered in the model, as only a single daily value is used for each variable. In any case, the relative average error in transpiration prediction remains quite low, even in autumn ( $< 11\%$ ).

Previous studies have also reported evidences on the relationship between forest transpiration and season, finding different performance of models depending on the season (Farrington et al. 1994, Borghetti et al. 1998, Schiler and Cohen 1998, Cohen et al. 2008, Whitley et al. 2009, Chirino et al. 2011). Such differential behaviour of transpiration is common in geographical regions with marked seasonal variations in the interrelationships between soil water availability, air temperature, vapour pressure deficit and rain distribution.

The structure of the neural network proposed here in is a typical FFNN of three layers fully connected, with standard configuration. Having only one node in the output layer, and once the input variables were selected, the most relevant decision that determines the dimension or the geometry of the network is the number of hidden nodes ( $n_h$ ) in the

hidden or intermediate layer. Optimal network geometry is highly problem dependent (Maier & Dandy 2000). We applied a trial and error procedure, training a number of different networks with different hidden layer size. Best performance was obtained for  $n_h=9$ , which can be considered a “medium size” network performing adequately and adapted to the problem characteristics, and achieving a balance between having sufficient free weights to capture the essential relationships between variables, and not too many to produce over-fitting of the training data and affect the network ability to generalise (Kumar et al. 2011). Concerning the activation functions representing the individual nodes internal operation, several options were tested, following previous authors. The choice of hyperbolic tangent activation function in the hidden nodes has been applied for similar problems in the past, like transpiration and trunk sap flow (Meijun et al. 2007, Liu et al. 2009), with satisfactory results. The most extended option for the output layer is that of linear activation functions (Vrugt et al. 2002, Garcia Santos 2007), as it avoids directly the limitation of the range of possible output values. In our case, though, the tests carried out with this option yielded a systematic sub-estimation of the higher values of transpiration. To overcome this problem, the exponential activation function in the output

node has been used in this research, producing in fact better transpiration estimates, and reducing the observed tendency to sub-estimate the higher values of transpiration.

The overall good performance showed by the selected neural network which includes all treatments, offers particularly interesting benefits in practice. It has the flexibility to estimate forest transpiration under different silvicultural treatments, using as inputs not only the usual meteorological variables and soil water content, but also forest cover. In other words, it can be successfully applied to estimate forest transpiration in a wide range of conditions, and can effectively incorporate diverse scenarios including climate change projections. As such, it constitutes a useful modelling tool to assess recommendations concerning thinning intensities in a managing context.

GOTILWA+ reproduced accurately the soil water content while transpiration was systematically underestimated. Some studies compare simulated and measured forest stand transpiration using GOTILWA+ (Kramer et al. 2002, Morales et al. 2005), but none of them using direct sap flow measurement in Aleppo pine stands. However, both of them registered an underestimation of forest transpiration. Kramer et al. (2002) applied GOTILWA+ and reported a general systematic discrepancy

between observed and estimated transpiration, and an underestimation at high radiation levels and at low temperature. In the same way, Morales et al. (2005) applied GOTILWA+ to estimate the actual evapotranspiration at different vegetation types and found a bias of 40-50 % where actual evapotranspiration was generally underestimated. Despite this underestimation, the use of GOTILWA+ is still justified, at least to estimate the daily soil water content required by the ANN as an input data, when it is not available.

## **5. Conclusions**

The results obtained in this research for transpiration prediction based on artificial neural networks techniques are promising, revealing an interesting potential for future practical benefits in water-oriented forestry and applied hydrology.

It has been successfully evaluated the ability of ANN computational schemes to predict actual measured transpiration rates from climatic data, soil water content and forest attributes. According to the reported results, ANN presents an improved capability for transpiration prediction as compared to that of linear models. This result

is consistent with the inherently non-linear relationships associated with the complex physical mechanisms underlying transpiration process.

This method to estimate the transpiration can alleviate the need for numerous time-consuming plant transpiration experiments using the heat pulse method, where only a few new points of measurement will be needed to adjust and verify the neural network model performance presented. The introduction of prediction methods based on advanced mathematical tools like the one explored herein can be of particular interest. Its use can effectively reduce the number of experiments to be held, without significantly affecting the overall knowledge of expected transpiration rates, for a given area and a given forest treatment. The fact that silvicultural treatment, represented as forest cover variation, is actually an input to the neural network extends the practical benefits of the method. In this sense, forest transpiration can be estimated under a wide range of conditions, including in the analysis, for instance, diverse managing scenarios or even climate change projections. From this perspective, the present approach represents a useful model strategy to assess recommendations for forest management taking into consideration forest water use.



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### **CHAPTER THREE**

Hydrology-oriented (adaptive) silviculture in a semiarid pine plantation:  
how much can be modified the water cycle through forest management?

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**Hydrology-oriented (adaptive) silviculture in a semiarid pine plantation: how much can be modified the water cycle through forest management?**

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## **Abstract**

Hydrology-oriented silviculture might adapt Mediterranean forests to climatic changes, although its implementation demands a better understanding and quantification on the water fluxes. The influence of thinning intensity (high, medium, low and a control) and its effect on the mid-term (thinned plots in 1998 and 2008) on the water cycle (transpiration, soil water and interception) and growth (Basal Area Increment, BAI) were investigated in 55-year-old Aleppo pine trees. Thinning enhanced a lower dependence of growth on climate fluctuations. The high intensity treatment showed significant increases in the mean annual BAI (from 4.1 to 17.3 cm<sup>2</sup>) that was maintained in the mid-term. Thinning intensity progressively increased the sap flow velocity ( $v_s$ ) in all cases with respect the control. In the mid-term, an increased functionality of the inner sapwood was also observed. Mean daily tree water use ranged from 5 (control) to 18 (high intensity) l tree<sup>-1</sup>. However, when expressed on an area basis, daily transpiration ranged from 0.18 (medium) to 0.30 mm (control), meaning that, in spite of the higher transpiration rates in the remaining trees, stand transpiration was reduced with thinning. Deep infiltration of water was also enhanced with

thinning (about 30% of rainfall) and did not compete with transpiration, as both presented opposite seasonal patterns. The changes in the stand water relationships after ten years were well explained by the forest cover metric. The blue to green water ratio changed from 0.15 in the control to 0.72 in the high intensity treatment, with the remaining treatments in the 0.34-0.48 range.

Key words: Blue-green water; Dendrochronology; Forest hydrology; *Pinus halepensis*; Transpiration

## 1. Introduction

Pine reforestation of barren lands has been called as an appropriate technique for soil and water conservation in the Mediterranean region, in which millions of hectares have been reforested, especially in Spain and Turkey (FAO 2010). Most of the plantations were established with one or several pine species (*Pinus halepensis*, *P. pinaster*, *P. pinea*) due to their excellent performance under harsh climatic and edaphic conditions. The improvement in site conditions (microclimate and soil properties) brought by the pine forests is expected to trigger late-successional species to spontaneously establish and to stimulate the ecosystem towards a more mature stage. However, this has not been the case in many semiarid areas, where additional management is needed to promote stand dynamics (Sánchez-Salguero et al. 2010). Thinning, the introduction of re-sprouting shrubs or the integration of small-scale spatial heterogeneity into the stand management strategy might be adequate techniques to improve stand resilience and overcome decline or stand stagnation (Maestre and Cortina 2004; Molina and del Campo 2012).



Under a general scope of precipitation decrease and evapotranspiration increase due to climate change in the Mediterranean, proactive adaptive management is becoming a basic strategy to either maintain or to gradually adapt current forest ecosystems (Birost and Gracia 2011; Fitzgerald et al. 2013; Lindner et al. 2010). Artificial plantations are a special case of forests with low resilience to environmental shifts, and they could be really favoured from this adaptive silviculture (Ungar et al. 2013). However, this type of silviculture is underdeveloped in many aspects as compared to that traditionally oriented to timber production. In this sense, guidelines dealing on how to maintain site productivity, enhance soil water content or promote tree and stand resilience (most adapted species, proper density, etc.) are needed for specific regions or ecosystem types.

The long-term effect of forest management on the water cycle has been a topic deeply studied in the field of forest hydrology by comparing the water flux in the catchment outlet (Webb et al. 2012). These types of studies are very useful to know the global impacts of forest management at catchment scale. However, land uses in Mediterranean catchments are very heterogeneous and the forest management should consider it accordingly. In this sense, there is still a

need for an improved understanding of how the different elements of the water cycle are affected and what is the management margin that foresters have to manipulate these elements (del Campo 2013). That is to say, it is needed to develop guidelines for a more efficient implementation of hydrology-oriented silviculture, which pursues the quantification and the manipulation of water cycle components in forests according to management objectives (Molina and del Campo 2012). The terms blue and green water (Falkenmark 2003) are usually referred to in this context (Birot and Gracia 2011).

Several studies conducted in the Mediterranean region have focused on the short-term effects of thinning on tree-water use and have demonstrated an increment in throughfall, an improvement of conditions for tree growth and a reduction of the evapotranspiration (Aussenac 2000; Ganatsios et al. 2010; Molina and del Campo 2012). Previous studies have addressed the water balance in Aleppo pine plantations in the eastern Mediterranean (Schiller and Cohen 1998; Ungar et al. 2013). However, understanding how and how much the water fluxes are mutually affected as a function of the intensity of the treatments, or how and how much their mid-term effect is offset by densification and

expansion of the canopy and root systems (Aussenac and Granier 1988; Andréassian 2004; Delzon and Loustau 2005) remains unclear.

The aim of this paper is to study how the water balance and the tree growth change as a consequence of thinning. Previous results on the effect of thinning intensity on the throughfall and stemflow were reported in Molina and del Campo (2012). Two main aspects are dealt with in this work, firstly the influence of thinning intensity and secondly the effect of the elapsed time since thinning. The experimental hypothesis is, in the first case, that clearing more trees increases the ratio of blue to green water (decreases the evapotranspiration's components), whereas in the second case, this ratio decreases with the time elapsed since thinning. To address these questions five experimental plots were established, three of them were thinned in 2008 at different intensities, the fourth was thinned in 1998 and the fifth was a control. Specific questions to be addressed are: i) what is the effect of the thinning intensity and the effect of the elapsed time since thinning on tree transpiration, soil water and growth? ii) How are affected the throughfall and stemflow by the time elapsed since the thinning intervention? iii) How does the global water balance of the forest change with both the thinning intensity and the elapsed time since thinning?

## **2. Material and methods**

### **2.1. Study site and experimental trial**

The study was carried out in a planted pine forest located in the southwest region of Valencia province in Spain (39° 05' 30" N, 1° 12' 30" W) at 950 m a.s.l. The average annual rainfall is 465.7 mm and typically shows high intra- and inter-annual variability. The mean annual temperature is 13.7 °C, the mean annual potential evapotranspiration is 749 mm (Thornthwaite), and the reference evapotranspiration is 1200 mm (Hargreaves). The soils display a basic pH of 7.6, are relatively shallow (50-60 cm) and have a sandy-silty loam texture.

The *P. halepensis* plantations were established in the area during the late 1940s with high densities (approximately 1500 trees ha<sup>-1</sup>), and no forest management has been carried out due to the role of the forest in soil protection (corroborated by the personnel at the nearby forest nursery). In late 1998, a 100 meters-wide strip thinning was performed in order to break the continuity of the forest canopy, as a typical fire prevention practice. This high intensity thinning (H98) left a final stand density of 155 trees ha<sup>-1</sup> and cover of 41% in 2009. In this area the understory vegetation is removed every two to three years (last

removal in 2008). In February 2008, a new experimental thinning including several intensities was carried out in the vicinity of the H98 treatment (80 m apart, same stand). In this case, each thinning treatment was performed in an experimental plot of 30×30 m. One plot was not thinned (control, C) and the other plots were thinned at three different intensities: high (H), medium (M) and low (L). Thinning removed the less developed trees and was performed to achieve a relatively homogeneous tree distribution (based on forest cover) in the plots. Thinning was conducted and supervised by the Forest Service of Valencia; timber and debris were removed and piled outside the plots. All plots were on a slope of less than 5 %. Forest structure characterization in all plots was accomplished in March 2009 as indicated in Molina and del Campo (2012) and presented in Table 1.

Table 1 - Means of the forest-structure variables in the control and thinned (2008 and 1998) plots

Thinning Treatment	Cover (%)	Basal area* (m <sup>2</sup> ha <sup>-1</sup> )	Density (trees ha <sup>-1</sup> )	DBH <sup>2</sup> (cm)	LAI (m <sup>2</sup> m <sup>-2</sup> )	Mean height** (m)	Heartwood area** (cm <sup>2</sup> )	Sapwood area** (cm <sup>2</sup> )	Crown projected area** (m <sup>2</sup> )	Crown volume** (m <sup>3</sup> )
Control (C)	84	40.1	1489	16.9	2.6	11.5	0.58	230.9		
Low intensity (L)	68	27.2	744	17.7	1.7	12.2	0.32	256.3		
Medium intensity (M)	50	18.2	478	17.5	1.7	11.3	0.25	250.8		
High intensity (H)	22	9.4	178	20.4	0.5	12.2	3.9	332.0	15.6	53.4
High intensity-1998 (H98)	41	13.6	155	25.2	0.9	12.6	8.8	504.7	25.8	93.8

H98 is the plot thinned in 1998; the rest thinned in 2008, except the control plot. DBH, diameter at breast height without bark, is estimated from two wood cores (south and north) at a height of 1.3 m. Mean bark thickness is 2.8 cm. Sample size is all the trees in each plot (\*) or 12 trees per plot (\*\*). Adapted from Molina and del Campo (2012).

## **2.2. Tree selection and characterisation**

A total of 20 trees (four per plot) were selected to study growth and transpiration by considering a diameter distribution of 3 classes in each plot (<20.5, 20.5-26.5, >26.5 cm). Two trees were selected from the lowest and the highest diameter class respectively, and the other two were selected from the middle class. This sample, although modestly sized, falls within the range considered in tree water-relations studies (Granier 1987; Klein et al. 2013; Martínez-Vilalta et al. 2002). Trees were selected from both the 2008 thinning (C, L, M and H) and the 1998 thinning (H98). The effect of forest management intensity was studied in the 2008 plots, whereas the influence of time elapsed since thinning was studied in both high intensity plots (1998 and 2008) assuming a chronosequence (Major 1951), i.e., regional climate, parent material (soil origin), topography and biota were considered as constant between both plots, whereas time was considered as variable. In this case, although both plots were the same age, we assumed both are two different temporal stages of the same population. In these treatments, characterization of trees was additionally performed, as these should have different architecture (Table 1): total height and begin of crown

height were measured with an optical hypsometer (Suunto, Finland), crown diameter was calculated by averaging the measurements of two orthogonal crown diameter projections, crown volume was estimated after summing up the two volumes (the lower third as a cylinder and the other two thirds as a pyramid), and bark thickness was estimated by averaging two measurements from the north and south sites of the trunks.

### **2.3. Tree growth**

Tree growth was studied using dendrochronological procedures. Two cores (north and south) were extracted from each selected tree at the end of the study (5-mm increment core). To avoid under-estimation (Mérián et al. 2013), between four and eight additional trees per plot were cored in the same way. All cores were visually cross-dated and measured to the nearest 0.01 mm (LINTAB 6.0, coupled with the TSAP-Win software package). Cross-dating of the tree-ring series was evaluated using COFECHA software (Holmes 1983). Basal Area Increment (BAI) was selected as an indicator of growth because it is closely related to the sapwood area and was calculated both per year and per tree. The BAI series were standardised by dividing the raw BAI by the expected values (dimensionless index) using a double de-trending



method: first, the series were adjusted by a negative exponential curve or a linear regression, second, a cubic smoothing spline function was used with a wavelength fixed at 66% of the length of the series and a 50% frequency cut-off; third, autoregressive modelling was carried out on each series to remove temporal autocorrelations (Cook and Briffa, 1990). The indexed residual series were subsequently averaged using a bi-weighted robust mean to obtain the residual chronologies using the dplR R-package (Bunn 2008; R Core Team 2013). The result of this procedure was a Basal Area Increment Index (BAIi).

#### **2.4. Transpiration and tree-water use determinations**

The study of the water cycle spans the period from March 27<sup>th</sup> 2009 to May 31<sup>st</sup> 2011, when most of the measurements were available, giving a total of 796 days, or 25 months, or 2.08 years. However, other time spells are included as seen in next points. Sap flow velocity was measured through the HRM method (Burgess et al. 2001; Hernandez-Santana et al. 2011; Williams et al. 2004) in all sample trees and programmed to average every hour. One sap flow sensor (HRM sensor, ICT International, Australia) was installed in each selected tree on the north side of the trunk at a 1.3-m height. A heater emits the heat pulse,

and the temperature increase is subsequently measured in two needles containing two thermocouples each, located 27.5 mm and 12.5 mm from their bases. Each pair of measurements (inner and outer) is then used to estimate the heat pulse velocity at the both depths and converted to sap flow velocity,  $v_s$  (Burgess et al. 2001). The system was powered by a 12-V battery connected to a solar panel and a data-logger (Smart Logger, ICT).

Sapwood area was obtained by subtracting heartwood area from the inner-bark area (Giuggiola et al. 2013) from the cores extracted for the growth analysis. The sapwood area was divided into four different sections to assign different  $v_s$  values and consequently to estimate daily values of sap flow ( $\text{l day}^{-1}$ ) (Hatton et al. 1990; Delzon et al. 2004) as the sum of their multiplications: 1) the  $v_s$  from the outer thermocouple was assigned to the sapwood area from the cambium to middle point located between the outer and inner thermocouples (i.e., 20-mm depth); 2) the  $v_s$  from the inner thermocouple was assigned to the sapwood area from the middle point to inner depth of the sensor (27.5-mm depth from the cambium); and 3,4) the remaining area from the inner depth to the beginning of the heartwood or to the pith (if heartwood was not present) was divided into two halves and then, the  $v_s$  value from the inner

thermocouple was multiplied by 0.75 and 0.25 respectively. Sap flow was up-scaled by the number of trees (density) to get the stand transpiration (mm) taking into account the diameter frequency distribution of trees in the different treatments.

Data were quality controlled for any possible spikes and gaps. In some cases, sap flow data were lost for more than 15-day spells (several months in the case of the M treatment), because of datalogger/sensors malfunction, battery failure and rodents activity. In these cases, an Artificial Neural Network (ANN) model was used to estimate transpiration (mm) using cover forest, soil moisture and meteorological data (coefficient of correlation: 0.95; Nash-Sutcliffe coefficient: 0.90).

## **2.5. Soil water content and climate variables**

The soil water content (SWC,  $\text{m}^3 \text{m}^{-3}$ ) was continuously measured for the whole period in all treatments every 20 minutes by means of FDR sensors (EC-TM, Decagon Devices Inc., Pullman, WA) connected to several EM50 (Decagon) data-loggers. In each treatment, between 6 and 9 sensors were placed at a 30 cm depth considering either tree influence or not (i.e., with/without crown influence: sensors

increased from 6 (C) to 9 (H) in order to account for this cover variability). Field calibrations were carried out by determining the gravimetric water content in 4 sampling dates (saturation, field capacity, between field capacity and wilting point and wilting point) to obtain the full range of soil water content in the study site. After the data readings were corrected, the current volumetric water content was divided by soil water content at field capacity (SWC/FC), in order to have a relative variable to be used in the comparisons among the treatments. Field capacity in each treatment was calculated from the average of SWC's readings in three dates (28-29/March/09; 12/Oct/10; 23/March/11) in which the rainfall depth was higher than 30 mm in the previous two days. These dates were selected in cool days (see next point). Data gaps in SWC for a whole treatment were minimal due to the use of several sensors and data-loggers, which always allowed for reliable data. However, in the medium treatment the number of data gaps was higher and estimations were performed from linear or polynomial functions fitted with the neighbouring sensors ( $r^2 > 0.80$ ).

The rainfall (Gr) was continuously measured by means of a tipping-bucket rain gauge with 0.2-mm resolution (7852 Davis, USA) located in an open area at 400 m apart from the experimental plots.

Measurements of air temperature and relative humidity were collected by a single sensor (RH/T sensor, Decagon Devices, Pullman, USA) placed at a 1 m height close to the rainfall gauge. The data were subsequently used to obtain values for the vapour pressure deficit.

## **2.6. Throughfall and stemflow on a tree basis (H and H98 treatments)**

A previous study (Molina and del Campo 2012) addressed the influence of thinning intensity on throughfall and stemflow in this site. The influence of time elapsed since thinning on the rainfall partitioning was studied here on a tree basis, i.e., under the crowns of the H and H98 trees. The throughfall was measured under the crown-projected area of each sample tree by a set of 18 collectors ( $\emptyset$ : 12 cm) systematically placed on six radial axes ( $60^\circ$ ) with respect to the trunk (72 collectors per treatment), at a 50 cm height and separated 50 cm from each other. The data collection was conducted between November 2009 and February 2010, the main rainy season in the area, by gauging the collectors 1 to 3 days after. Evaporation during rainfall was expected to be negligible and very similar among isolated trees of different structure (Pereira et al. 2009). Therefore, the collectors' measurements (throughfall) were

assumed to be representative of the water saturation capacity of crown. The stemflow was initially measured in the same season, although the low amount of stemflow and its irregularity made it advisable to extend the measuring until June 2010. This variable was measured in all sample trees by fixing plastic collars (cut lengthwise) to the trunks at a 1.3-m height. Water from the collars was subsequently collected in 10-l tightly closed containers at intervals of 6-12 days.

## **2.7. Data analysis**

The total set of 796 days was divided into four day-types according to daily precipitation and daily mean temperature. First, days were grouped into dry or wet spells (D, W). A dry spell was considered to begin when none of the previous 14 consecutive days registered a daily precipitation higher than 5 mm (day 14<sup>th</sup> would belong to wet and day 15<sup>th</sup> would belong to dry; that is to say, wet spell is set to a minimum to 14 days long, whereas dry spells do not have minimum limit). This resulted in 18 and 17 wet and dry spells respectively. Secondly, in any of these periods, each single day was classified as cool or warm (C, W) if its mean temperature was respectively lower or higher than 13.2°C, the

overall mean of the data set. The DC, DW, WC and WW codes are used for each day-type.

Differences in BAI<sub>i</sub>,  $v_s$ , sap flow and SWC/FC among treatments were analysed with ANOVA (Steel and Torrie 1989) with treatment and tree considered as fixed factors. When ANOVA indicated significant differences between treatments, the Tukey post-hoc test was selected for the comparison of multiple means. In every case, the data were examined to ensure normality using the Kolmogorov–Smirnov test and the homogeneity of variance using the Levene test. When these assumptions were violated, the variables were transformed with power functions to achieve homoscedasticity or, alternatively, a nonparametric Kruskal–Wallis test was used (with the Tamhane’s T2 test used to compare multiple means). Relationships between different variables (growth and transpiration) were investigated through Pearson correlations and linear regression models. Here, the residuals were examined for normality and independence (Steel and Torrie 1989). A significance level of  $p < 0.05$  was used for all analyses. Data were analysed with SPSS© 16.0. All statistical proofs were performed considering only empirical data, i.e. data estimated to fill in gaps were used exclusively for water balances. In the case of  $v_s$ , sap flow and

SWC/FC the analyses were also carried out for the four different day-types.

### **3. Results**

#### **3.1 Climatic conditions during the study period**

From April 2009 to May 2011 the total precipitation was 1545 mm, 511 mm higher than expected (1961-2007). Despite this observation, drought spells occurred throughout the study period: only 26 mm of rain were registered in the 17 dry spells registered, accounting for 210 days out of the 796 days. The longest dry spell lasted 48 days (summer 2009). From December 2009 to April 2010, precipitation was much higher than expected. Regarding temperature, 412 days were classified as cool days ( $T < 13.2$  °C) and 383 days as warm ( $T > 13.2$  °C) (Fig. 1). Mean vapour pressure deficit (VPD) for each category was  $0.37 \pm 0.17$ ,  $1.56 \pm 0.73$ ,  $0.26 \pm 0.17$ ,  $0.96 \pm 0.57$  kPa for DC, DW, WC and WW respectively. Frequency of each day-type in the whole study period is presented in Table 2.



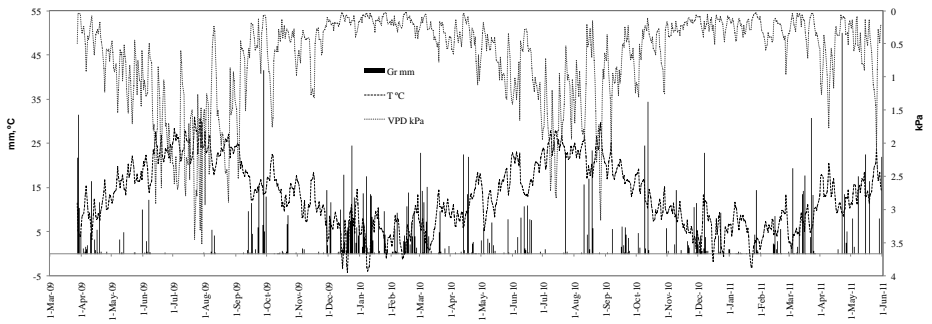


Fig. 1 - Meteorological conditions during the study period expressed as accumulated daily values for precipitation and mean daily values for the other variables: Gr is daily rainfall, T is the mean daily temperature of the air, and VPD is the mean daily vapour pressure deficit of the air

Table 2 - Mean values of daily  $v_s$  ( $\text{cm h}^{-1}$ ), accumulated daily sap flow ( $1 \text{ day}^{-1}$ ) and daily soil water content relative to field capacity (SWC/FC,  $\text{cm}^3 \text{ cm}^{-3}$ ) as regards the four day-types considered in this study (DC, dry-cool; DW, dry-warm; WC, wet-cool; WW, wet-warm) and the sapwood depth (the case of  $v_s$ )

<b>Variable and factor</b>	<b>C</b>	<b>L</b>	<b>M</b>	<b>H</b>	<b>H98</b>
<b><math>v_s</math> (<math>\text{cm h}^{-1}</math>)</b>					
Outer	1.18a	1.26b	1.45c	3.46d	1.79*
Inner	0.70a	0.88b	0.94b	2.66c	1.64*
DC (8.7%)	0.71a	0.78ab	0.93b	1.98c	1.43*
DW (17.7%)	0.94a	1.24b	2.29c	5.59d	2.58*
WC (43.1%)	0.64a	0.66a	1.03b	1.54c	0.87*
WW (30.4%)	1.42a	1.62b	2.62c	4.00d	2.07*
<b>Sap flow(1 day-1)</b>					
DC	3.57a	3.82a	4.54a	12.00b	12.9
DW	4.58a	6.51b	11.33c	33.80d	22.90*
WC	2.72a	2.70a	5.08b	7.94c	8.03
WW	6.89a	8.00a	12.93b	24.08c	18.94*
<b>SWC/FC</b>					
DC	0.66a	0.61a	0.78b	0.81b	0.70*
DW	0.49a	0.44a	0.62b	0.67b	0.59*
WC	0.84a	0.86a	0.83a	0.96b	0.89*
WW	0.67a	0.69a	0.75ab	0.82b	0.71*

Different letters in a same row indicate significant differences at  $p\text{-value} < 0.05$  in that variable for that level of the factor. In the H98 column, \* indicates significant differences with the H treatment at  $p\text{-value} < 0.05$

### 3.2. Tree Growth

The BAI and BAI index (BAIi) series showed similar patterns until thinning both for the intensity (C, L, M, H) and the elapsed time (H, H98) treatments (Figs. 2 and 3) with significant correlations between any

pair of treatments in all cases (Pearson correlation  $p$ -value $<0.001$ , 1960-1997 series). This similar pattern changed sharply in response to the thinning treatments carried out either in 1998 (case of elapsed time) or in 2008 (case of thinning intensity) and no significant correlation (neither for BAI and BAIi) was detected from this time onwards. In the intensity treatments, the BAI changed from an annual mean of  $4.09\pm 1.36$  and  $3.61\pm 1.19$   $\text{cm}^2$  (1960-2007) in H and C treatments respectively to an annual mean of  $17.29\pm 5.96$  and  $3.47\pm 0.14$   $\text{cm}^2$  (2008-2010) respectively. L and M thinning treatments presented intermediate values ( $5.18\pm 0.39$  and  $7.23\pm 2.07$   $\text{cm}^2$  in the 2008-2010 spell respectively) (Fig. 2). In the case of H98, the BAI increased from an annual mean of  $3.52\pm 1.03$   $\text{cm}^2$  in 1960-1997 to  $18.31\pm 8.92$   $\text{cm}^2$  in 1998-2010 (Fig. 2).

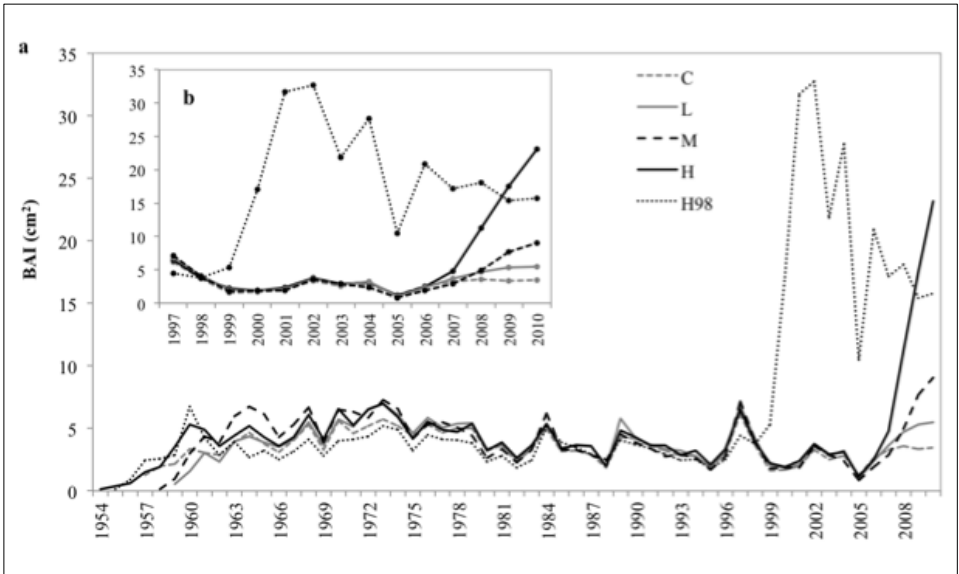


Fig. 2 - Mean Basal Area Increment (BAI) values in each treatment along the entire growth period analysed (1960-2010, a) and a close-up (b) showing differences in growth after thinning in 1998 (H98) and in 2008 (remaining treatments). C – control, L – low intensity; M – medium intensity; H – high intensity 2008; H98 – high intensity 1998

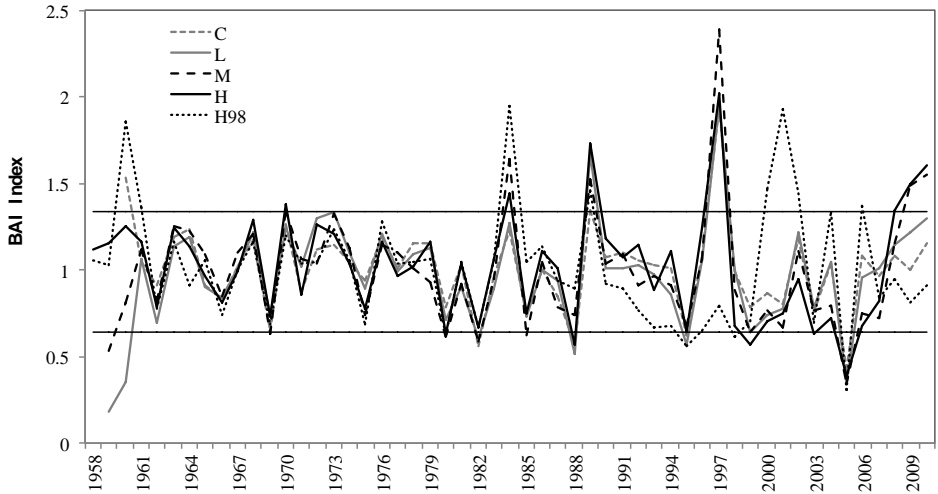


Fig. 3 - Mean Basal Area Increment index (BAIi) in each treatment along the entire growth period analysed (1960-2010). Horizontal lines delimitate the 10-90% range. C – control, L – low intensity; M – medium intensity; H – high intensity 2008; H98 – high intensity 1998

Parametric tests indicated significant differences in the BAIi after thinning treatments, either for the intensity or the elapsed time factors ( $p$ -value $<0.001$ ). In the former case, C and L presented BAIi values significantly lower than those of M and H treatments, whereas in the latter case, BAIi in H98 was significantly lower in the period 1998-2010 than that for H in the period 2008-2010 ( $1.03\pm 0.45$  and  $1.48\pm 0.13$  respectively). Another remarkable result was that, independently of the treatments, the BAIi chronology (Fig. 3) reflected a first stage with low amplitude oscillations until the early 1980s and a second stage from this

date onwards in which the amplitude of the oscillations was higher: most of the values (89%) falling outside of the central interval (10%-90%) belong to years after 1980 (57% of the years). This observation was especially relevant in the driest years (e.g., 1988, 1995 and 2005), which induced a sharp decrease in growth.

### **3.3. Tree-water use and stand transpiration**

The water use data showed a variable behaviour depending on the different factors being considered in this study: the variable considered (i.e.,  $v_s$  [*mean, outer, inner*], sap flow or stand transpiration), type of analysis (thinning intensity or elapsed time), or the day-type (spells) (Table 2; Fig. 4). Thinning intensity affected significantly  $v_s$  both on its outer and inner measurements, progressively increasing with the intensity of the treatment from C to H (Table 2). Moreover  $v_s$  (mean) was also significantly affected by thinning intensity according to the day-type considered, with warm days (either from wet or dry spells) associated to higher differences among treatments (Table 2). The maximum differences among treatments were found when  $v_s$  (mean) was high, where the Tukey test yielded four single-treatment groups. Sap flow differences among treatments were also significant, with higher

differentiation in the warm spells too. Tukey tests always isolated the H treatment from the others, whereas C and L were frequently joined in a same group; M treatment showed an intermediate pattern between the L and H treatments in most of the cases (Table 2). In several trees from the H treatment, daily sap flow in June and July 2009 (dry warm spell) surpassed the amount of 50 l.

Regarding the elapsed time from thinning, the  $v_s$  was significantly different between H and H98 in all analyses, with the latter showing values about one half of H, except in the inner sapwood  $v_s$ , where differences were lower (Table 2). In this comparison, the sap flow ( $l\ day^{-1}\ tree^{-1}$ ) presented significant differences between both treatments only in the warm spells and the magnitude of these differences was lower (about one third), thus indicating relatively less changes in total tree-water consumption after ten years of thinning intervention. The weighted (from each day-type frequency) daily average of sap flow during the study period was 5.22, 5.08, 8.52, 17.77 and 14.39 litres of water transpired per tree for treatments C, L, M, H and H98 respectively.

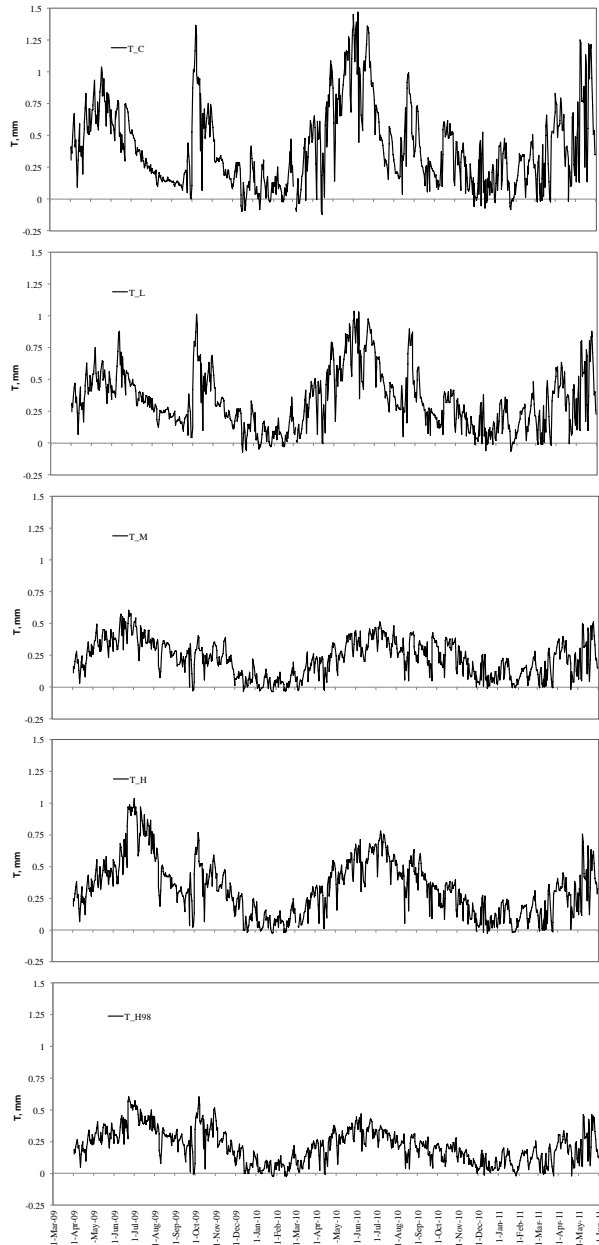


Fig. 4 - Daily mean accumulated values of transpiration (mm) in each treatment along the entire study period (March 2009-May 2011). C – control, L – low intensity; M – medium intensity; H – high intensity 2008; H98 – high intensity 1998



Considering the frequency distribution of diameters in the different treatments as well as the tree density (Table 1), the previous sap flow values correspond to 0.297, 0.256, 0.180, 0.271 and 0.170 mm transpired per day or 108.6, 93.4, 65.7, 98.9 and 61.9 mm transpired per year for treatments C, L, M, H and H98 respectively.

The tree growth (BAI) presented many significant correlations with  $v_s$  and the sap flow (Table 3). These correlations were stronger ( $p$ -value $<0.001$ ) when considering transpiration variables from the previous year, i.e., the 2010 BAI with the 2009 transpiration variables. Also, the association was higher for warmer periods, with Pearson's coefficient reaching values higher than 0.8. The BAI<sub>i</sub> showed few significant correlations with the transpiration variables and these were weaker than in the case of BAI ( $p$ -value $<0.05$ ). Based on those correlations, simple linear regressions between  $v_s$  and BAI were developed. The models fitted were  $v_{s \text{ inner } 2009} = 0.071 * \text{BAI}_{2010} + 0.5961$  ( $p < 0.001$ ,  $r^2 = 0.70$ , d.f.: 14; MAE=0.44; MAPE=52.9%) and  $v_{s \text{ outer } 2009} = 0.098 * \text{BAI}_{2010} + 0.9689$  ( $p < 0.001$ ,  $r^2 = 0.65$ , d.f.: 14; MAE=0.68; MAPE=35.3%).

### 3.4. Soil water content

According to the calibration, the sensors output increased the estimated volumetric water content in a general manner. Soil water content at field capacity varied among treatments in spite of the fact of their vicinity:  $0.306 \pm 0.026$ ,  $0.264 \pm 0.016$ , and  $0.252 \pm 0.026$ ,  $0.316 \pm 0.023$  and  $0.201 \pm 0.025 \text{ m}^3 \text{ m}^{-3}$  for C, L, M, H and H98 respectively, thus showing a different water holding capacity. In this sense, comparisons among treatments were done based on the relative variable water content to field capacity ratio (SWC/FC) (Fig. 5).

Table 3 - Pearson correlation coefficients (only high significant cases with p-value<0.001 are shown) between tree growth (Basal Area Increment, BAI) and different transpiration variables (sap flow velocity  $v_s$  and sap flow), as regards the study year (2009 and 2010) and the day-type (DC, dry-cool; DW, dry-warm; WC, wet-cool; WW, wet-warm). N=20.

	BAI 09	BAI10
Sap flow_09	0.736	0.763
Sap flow_09_DC	0.765	0.78
Sap flow_09_DW	0.785	0.82
Sap flow_09_WC	0.727	0.747
Sap flow_09_WW	0.751	0.761
Sap flow_10_DW	0.725	0.736
Sap flow_10_WW	0.679	0.684
$v_{s\_inner\_09}$	0.752	0.835
$v_{s\_inner\_10}$		0.74
$v_{s\_outer\_09}$		0.807
$v_{s\_inner\_09\_DC}$	0.733	0.81
$v_{s\_inner\_09\_DW}$	0.764	0.849
$v_{s\_inner\_09\_WC}$		0.76
$v_{s\_inner\_09\_WW}$	0.727	0.804
$v_{s\_outer\_09\_DC}$		0.81
$v_{s\_outer\_09\_DW}$		0.817
$v_{s\_outer\_09\_WC}$		0.745
$v_{s\_outer\_09\_WW}$		0.781
$v_{s\_inner\_10\_DC}$		0.677
$v_{s\_inner\_10\_DW}$	0.709	0.785
$v_{s\_inner\_10\_WW}$		0.704
$v_{s\_outer\_10\_DW}$		0.719

Significant differences were obtained for either the thinning intensity analysis or the elapsed time analysis in the four day-types

considered (Table 2). In the first case, the treatment H showed always more relative water content than C and L treatments (range 67-96% and 44-86% for H and C, L respectively), whereas M had an intermediate behaviour, showing significant differences with H in the wet-cool spells (lower relative water content) and with C, L in the dry spells (higher relative water content). Regarding the elapsed time analysis, differences between H and H98 were significant and maintained in the four day-types considered, showing the latter lower values.

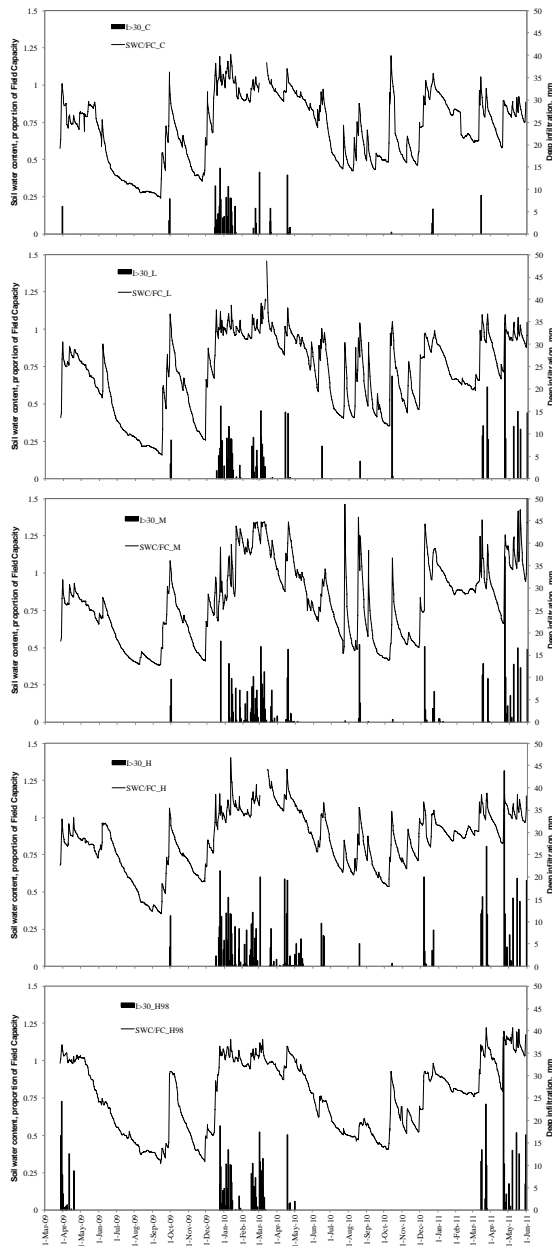


Fig. 5 - Daily mean values of soil water content, expressed as its proportion over field capacity (SWC/FC), and deep infiltration ( $I_{>30\text{cm}}$ ) in each treatment along the entire study period (March 2009-May 2011). C – control, L – low intensity; M – medium intensity; H – high intensity 2008; H98 – high intensity 1998

### 3.5. Throughfall and stemflow in the elapsed time analysis

Throughfall in the thinning intensity analysis was presented in Molina and del Campo (2012). Regarding the elapsed time analysis, a total rainfall of 146.8 mm was collected during the rainy spells when throughfall was measured; spells ranged from 2.4 to 56.4 mm. The throughfall under the crown-projected area of the sampled trees was always higher for treatment H (ranging from 55.2 to 77.0% of the gross rainfall) than for H98 (from 39.3 to 67.0%) (Fig. 6). A significant difference ( $p$ -value $<0.001$ ; d.f.:142) was found between the total accumulated values of the throughfall in H ( $60.3\pm 6.8\%$  of the gross rainfall) and H98 ( $51.4\pm 9.7\%$  of the gross rainfall), accounting for a mean general difference of 8.9%. Linear regressions between the throughfall and the rainfall collected in each period were highly significant for both treatments (Throughfall= $0.64 * \text{Gross\_Rainfall} - 0.75$ ;  $p < 0.001$ ,  $r^2 = 0.97$ , d.f.: 28; MAE=1.17; MAPE=12.9%, and Throughfall= $0.58 * \text{Gross\_Rainfall} - 1.57$ ;  $p < 0.001$ ,  $r^2 = 0.94$ , d.f.: 27; MAE=1.79; MAPE=36.7%, for H and H98, respectively). The comparison between the models indicated that the intercepts were found to be different at  $p$ -value $<0.001$ , whereas the slopes were not.

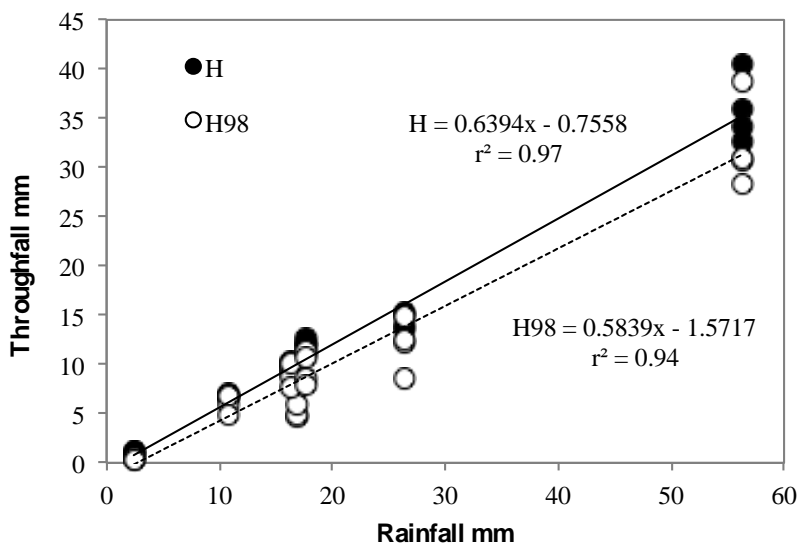


Fig. 6 - Relationship between throughfall (mm) and rainfall (mm) in the high intensity treatments (H and H98). N= 28 and 27 for H and H98 respectively. Regressions models were statistically significant at p-value<0.001. Statistical comparisons between the models indicated that intercepts were different at p= 0.001.

The stemflow showed a similar trend for both treatments, with no significant differences in the accumulated values for the entire study period, averaging  $24.03 \pm 1.24$  l tree<sup>-1</sup> in H and  $17.73 \pm 9.45$  l tree<sup>-1</sup> in H98 (Fig. 1 - Complementary materials). A high variation in stemflow was noted among the different periods as well as high intra-variation in each treatment, an observation that was much more pronounced in treatment H98. The linear regressions showed a poor relationship between the stemflow and the rainfall in both treatments (not shown).

### 3.6. Water balance for the whole study period

An attempt of integration of previous results in a global water balance is presented in Table 4, which shows this balance for all the treatments on a stand basis (as mm and as % of rainfall). Stand interception ( $I_t$ ) was subtracted to gross rainfall (mm) to compute total throughfall (stemflow was considered negligible). Interception was estimated on a daily basis based on the exponential model of throughfall reported in Molina and del Campo (2012). In the case of H98, the model was modified in order to decrease throughfall 0.82 mm under the area covered by the trees (41%), as seen in this work (0.82 is the difference in the intercepts of the linear equations presented in 3.5). The remaining precipitation (net precipitation,  $P_n$ ) was considered to be infiltration ( $I$ ).  $P_n$  fallen when SWC/FC was higher or equal to 1 was considered as deep infiltration (Fig. 5), which is a proxy for deep percolation. Summing up the values per treatment, it yielded 207.6, 395.1, 455.5, 647.0 and 505.9 mm for C, L, M, H and H98 treatments respectively in the whole period analysed (796 days). These amounts were irregularly distributed among the four day-types: no deep infiltration was registered in the dry-warm spells, negligible amounts in the dry-cool, between 14% (H) and 8.6%



(C) of the total was infiltrated in the wet-warm periods and between 86% (H) and 91% (C and L) was infiltrated in the wet-cool periods.

Table 4 Itemisation of the water cycle at the stand scale for the whole study spell (March 2009 to May 2011) for each treatment (C: control, L: low intensity; M: medium intensity; H: high intensity 2008; H98: high intensity 1998). Gr: gross rainfall; It: interception loss; Thr: throughfall; T: stand transpiration; I>30cm: deep infiltration; E: evaporation from soil, litterfall and grass/scrub transpiration. ET total: summing up of the evapotranspiration terms; B/G: blue (deep infiltration) to green (total evapotranspiration) ratio. See text for details.

	<b>Gr</b>	<b>It</b>	<b>Thr</b>	<b>T</b>	<b>I&gt;30cm</b>	<b>E</b>	<b>ET total</b>	<b>B/G</b>
<b>mm</b>								
<b>C</b>	1545	611.8	933.2	319.8	207.0	406.3	1338.0	0.155
<b>L</b>	1545	517.6	1027.4	264.2	395.5	367.7	1149.5	0.344
<b>M</b>	1545	400.2	1144.8	180.8	455.8	508.3	1089.2	0.418
<b>H</b>	1545	191.6	1353.4	261.1	647.4	445.0	897.6	0.721
<b>H98</b>	1545	419.5	1125.5	168.4	499.5	457.6	1045.5	0.478
<b>%</b>								
<b>C</b>	100	39.6	60.4	20.7	13.4	26.3	86.6	0.155
<b>L</b>	100	33.5	66.5	17.1	25.6	23.8	74.4	0.344
<b>M</b>	100	25.9	74.1	11.7	29.5	32.9	70.5	0.418
<b>H</b>	100	12.4	87.6	16.9	41.9	28.8	58.1	0.721
<b>H98</b>	100	27.1	72.8	10.9	32.3	29.6	67.7	0.478

Subtracting deep infiltration ( $I_{>30\text{cm}}$ ) to  $I$  corresponds to the  $P_n$  that fell when soil water content was below field capacity. This term was divided into stand transpiration ( $T$ ) and a residue that includes the upper soil horizon evaporation and the interception and transpiration of understory. The total evapotranspiration (interception plus stand transpiration plus the residual component) ranged from 86.6% of gross rainfall in the control treatment to 58.1% in the high intensity treatment H, with the remaining treatments concentrated around the value of 70% (H98, M and L, from lower to higher evaporation). Concordantly, blue ( $I_{>30\text{cm}}$ ) to green (total evapotranspiration) water ratios ranged between 0.15 (C) to 0.72 (H). Time elapsed since thinning decreased this ratio to 0.49, which is close to that of the medium intensity thinning, with similar forest cover.

#### **4. Discussion**

Proactive adaptive silviculture is a key element for coping with the impacts of global change in the Mediterranean forests due to their higher vulnerability (Lindner et al. 2010). This brings into focus the need to develop, implement and improve adaptation measures. In the present

study, the effect of thinning interventions was studied by focusing on the change in the distribution of water fluxes and tree growth in a planted pine forest in a semiarid climate.

#### **4.1. Tree growth**

The dendrochronological approach applied in this work facilitated the study of the stand growth during most of its lifespan. Growth was highly limited in the pre-thinning stages of the stand, with basal area showing increments around 3.5-4 cm<sup>2</sup> year<sup>-1</sup>. The BA increments for this species in the eastern region of Spain are within 8-11 cm<sup>2</sup> year<sup>-1</sup> for a DBH of 20-25 cm (Condés and Sterba 2008), although wider ranges (2-15 cm<sup>2</sup> year<sup>-1</sup>) and higher maximum values (22.2 cm<sup>2</sup> year<sup>-1</sup> in the wettest area of the species in Spain) have also been reported (Linares et al. 2011; Sánchez-Salguero et al. 2010). In this study, the interventions improved the basal area increment in all cases, although in the low intensity thinning the value is still low as regards the reported ranges. On the contrary, the high intensity thinning treatments revealed the improved growth capacity of the species when competence is suppressed. Delzon and Loustau (2005) showed that growth in a *P. pinaster* stand with 150 trees ha<sup>-1</sup> began to decline at approximately 40

years. In this work, the inter-annually averaged BAI in H98 of approximately  $18 \text{ cm}^2 \text{ tree}^{-1} \text{ year}^{-1}$  from thinning on, indicates the potential and lasting response associated with thinning, even for the mature state of the studied trees (55 years old).

Tree ring chronologies reflect the complex interaction of climatic and environmental conditions at sites where samples are taken (Fritts 1976). In our case, the BAIi chronology reflected a two-stage pattern of oscillations, with a break point around the early 1980s that was independent of the treatment. Water availability is one of the main factors that control growth in this species, with a significant relationship between tree growth and rainfall commonly observed (De Luis et al. 2009; Raventós et al. 2001; Sanchez-Salguero et al. 2010). In fact, in such semiarid well-drained sites as the experimental plots, a maximum response of the tree rings to precipitation is expected (Meko et al. 1995), and could be the driving factor for the higher amplitude of oscillations in the second stage of our chronology (from the early 1980s onwards). In this work, two observations occurred concurrently: first, the tree density became excessive in the early 1980s, necessitating at least one thinning intervention according to Montero et al (2001). Second, these years correspond to a generalised warming and drying trend in the

Mediterranean climate, conditions that are reported to affect the growth of this species (Linares et al. 2011; Sarris et al. 2007, 2011). An increasing number of missing tree rings from the late 1980s has been reported in this species in a nearby area (Raventós et al. 2001). Long, intense and continuous droughts can have a spatial structuring effect on the growth index, especially if successive vegetative seasons record strong precipitation deficits (Planchon et al. 2008), as observed in 1994-1995 and 2004-2005 in this study. Fig 2 shows that the amplitude of oscillations is higher and the BAI<sub>i</sub> value is below 1 in C; L, M and H in the last years before the thinning, but it changed to 1.08, 1.22, 1.39 and 1.48 respectively for 2008-2010. By the same, during the 1999-2007 spell, the H98 showed a trend towards values higher than 1, averaging 1.12. Taken together, these observations mean that a lack of forest management together with rainfall variability is highly reflected in the BAI index chronology, which shows a higher and stronger dependence of growth on climate fluctuations.

#### **4.2. Tree water use and stand transpiration**

The tree water use results explain how the water fluxes have changed in the short and mid term after thinning. The availability of such

limited resources such as water, radiation or nutrients has increased for the remaining trees, and as a consequence, tree water use has increased after thinning, which is a known fact (Medhurst et al. 2002; Morikawa et al. 1986). The results of tree water use have shown that differences in transpiration among the treatments varied depending on the considered variable, i.e., sap flow velocity ( $v_s$ ), sap flow or stand transpiration.

First,  $v_s$  always increased with the intensity of the intervention, independently of the spells or the depth of the thermocouple (either inner or outer). This result might be due to an increase in the hydraulic conductivity of the sapwood soon after thinning (Medhurst et al. 2002), which might be also positively correlated with the increased water availability (White et al. 1998). The outer and inner  $v_s$  data allow additional insight into the intrinsic differences between treatments in the sapwood functionality. We observed changes in the radial variation of  $v_s$  with treatment and hence in the permeability of the sapwood: the mean differences between the outer and inner records went from 40% in the C treatment to approximately 28% in the H, and only 8% in H98. The detailed work of Cohen et al. (2008) does not explain this fact completely. If we add a hypothetical third point to these series (zero velocity at the pith depth) and fit the proper functions, the C series fits

better to a logarithmic pattern, whereas H98 fits better to a linear one. In studying the question of why trees growing on better sites showed greater sapwood permeability and conductance, Shelburne and Hedden (1996) obtained logarithmic and linear fits for poor and good sites, respectively. This changing pattern in the hydraulic conductivity of the sapwood could be explained by differences in permeability due to a higher functionality of tracheids. Better sites allow for additional functional tissue in the inner sapwood, which thus enhances the tree's ability to obtain adequate nutrients and water. In our case, the H98 trees had been growing in better conditions (lower competence) for a longer period of time, and hence, their conductive anatomy was more adapted (the outer and inner thermocouples were located in rings formed after the 1998 thinning, i.e., under low stand density conditions), whereas the sapwood permeability drops off much more rapidly in the inner sapwood of trees growing in high stand density (higher competence) like the C treatment, because many tracheids have become non-functional. In contrast, the H trees, showed intermediate pattern because, on the one hand, they are growing on an appropriate density since 2008 and show higher permeability along the sapwood, but, on the other hand, they have

had little time to adapt to the new stand conditions, and the rings before 2008 were formed under worse conditions due to excessive competence.

Second, the values of sap flow found in this work for 55-year-old *P. halepensis* trees ranged from 5 to 18 l day<sup>-1</sup> which are consistent, in the case of the H treatment (basal area 9.4 m<sup>2</sup> ha<sup>-1</sup>), with those found in the same species and basal area by Schiller and Cohen (1998) under more arid Mediterranean conditions (maximum and average of 49 and 17 l day<sup>-1</sup>, respectively). A noticeable finding is that the larger H98 trees (diameter, crown volume, height, etc.) used less water than the smaller H trees. This observation is not common in the literature, where the opposite pattern is usually reported (O'Grady et al. 1999). However, this difference is relatively low despite the higher differences in  $v_s$  between both treatments. The reason for these observations again underscores the relative importance of the inner and outer sapwood in the total tree-water use for both treatments. The H is nearly exempted from water deficits year-round, thus exhibiting a type of water-spending behaviour indicative of the sudden release of water and nutrient resources. This observation is reflected in notably wide new rings (outer sapwood growth), high sap velocities and different response to vapour pressure deficit (Fig. 2 - Complementary materials). In H98, a higher number and



better functionality of tracheid layers in the inner sapwood allows for sufficient water supply to its dense and expanded crown, thus minimising the differences with H, where the impact of previous branches and tissues is lower. However, this pattern is likely to change in the short term because the cool spells indicate no differences between them.

However, the results for stand transpiration give a different ranking for the treatments to that obtained on a tree basis. Effectively, when the number of trees is taken into account, the C treatment is the most water-spending stand in spite of its lower growth. Low growth and high green water losses are the reasons to implement hydrology-oriented (proactive adaptive) silviculture. Transpiration values presented here are in the range of those reported for the species by Ungar et al. (2013), although the long-term average in our case is lower. This could be due to the cumulative effect of wet days in our case that led to a relative lower importance of days owning high evaporative demand (high VPD, see Table 2 and Fig. 1). Nevertheless, our data are subjected to errors. In addition to the errors associated with the heat pulse method for the  $v_s$  estimation (Hatton et al. 1990), computation of the sap flow from  $v_s$  may lead to important overestimations that have been widely reported and discussed in tree water use studies (Delzon et al. 2004; Cohen et al.

2008). We established a pattern of variation of  $v_s$  with the area of the sapwood annuli sampled by each thermocouple (Hatton et al., 1990). In contrast, the results from Cohen et al. (2008) indicate that the pattern of  $v_s$  in this species in Israel decreases considerably, with negligible values beyond a 4-cm depth. This reasoning would make our values of sap flow to decrease. By contrast, applying the correction factor from Delzon et al. (2004) for maritime pines (a fast-growing species in a more humid climate), our overall sap flow values would increase. Thus, it can be argued that our criteria can yield acceptable values, particularly if we consider that the years of study were particularly wet (the correction factor from the latter reference may be more appropriate).

Although the H trees transpired more water than H98, there was more water in the soil in the former than in the latter. The interception loss is a direct cause for this difference because net precipitation in H was approximately 10% higher. The elapsed time from thinning changed the tree storage capacity, as indicated by a difference of 0.82 mm between the intercepts (Rutter, 1963). The throughfall (49 - 62% of rainfall) and stemflow (0.25 - 0.5% of rainfall) values fall within the range reported in other studies conducted on isolated trees in the Mediterranean area (Belmonte Serrato 1997; Pereira et al. 2009).

Another important difference between H98 and H (and extensively to the remaining treatments) is that found in the hydraulic properties of the soil, as field capacity (and absolute soil water content) was lower in H98. This could be due either to spatial variations in the sites (corroborated by the BAI chronology, Fig. 2, which indicated lower growth of H98 trees) or to the forest treatments themselves. If we assume the chronosequence hypothesis of this work (which establishes that both soils are similar), then it is needed to address why a lower water holding capacity have appeared in H98 after the thinning intervention. A possible explanation could derive from the higher density of root systems in the H treatment, which contribute (death or alive) substantially to soil organic matter (Persson 2012); their cavities can readily fill with water during and after major precipitation events, thus affecting soil hydraulic properties (Devitt and Smith 2002). This reasoning would mean that thinning from high to low densities might affect the soil water holding capacity in the mid term.

#### **4.3. Water balance**

Thinning has affected all the water cycle components considered in this study. The water balance indicates that the first main

difference among treatments is found in the interception loss (C intercepted 27% more rainfall than H), as previously reported (Molina and del Campo 2012). Thinning also diminished the stand transpiration in all cases, although this reduction was lower than that found for rainfall interception (differences <10% of rainfall). The important point here is that, in spite of the higher water use rates maintained in the remaining trees, the stand transpiration component is reduced even in the low intensity thinning treatment. Our results also indicate a little competence between transpiration and the deep infiltration terms for the throughfall water, as they present opposite seasonal patterns. This would mean that the differences in throughfall in wet-cool spells are relocated into deep infiltration water if the soil is wet enough; all the treatments presented important differences in this term against the control (>12%). The term for soil/understory evaporation owns higher uncertainty due to its residual nature. This component yielded low differences among treatments (<10% of rainfall), which would mean that, in spite of clearing vegetation, the evaporation is not severely increased. The ratio of the evaporation term to rainfall deduced in this work (0.24-0.33) is slightly lower to that reported by Ungar et al. (2013) in a drier climate (0.34-0.42). Thus, higher soil or understory evaporation in the more

intensive treatments (M, H and H98) is compensated with reduced transpiration due to a lower tree number per unit area. Regarding the temporal evolution, water balance between H and H98 indicates that the latter evolves towards the values found for the medium intensity treatment, with similar cover but higher number of trees. Ten years after thinning primary and secondary growth have affected the entire tree architecture, including the crown densification and a higher complexity in canopy structure. However, our results would indicate that these changes could be easily integrated into the forest cover metric, at least with regards their effects on the tree-water relationships. In fact, regressing the blue to green water ratio on the forest cover (proportion) the model yields a good fit ( $r^2 > 0.96$ ;  $p < 0.01$ ; Fig. 3 - Complementary materials).

## **5. Conclusion**

Concluding, we can confirm the two experimental hypotheses tested in this work and consider the results meaningful for a hydrology-oriented silviculture for these types of plantations. Methods from dendrochronology and hydrology have proven useful in deepening our understanding of the effects of thinning in this semiarid forest. The lack

of forest management led to growth stagnation and to a higher and stronger dependence on climate fluctuations (rainfall variability), as was reflected by the BAI. Management of these forests for maximising blue water budgets is effective both on the short and the mid-term according to the usual forestry timeframes. Ten years after thinning, tree growth and transpiration remained enhanced with higher relative inner sapwood permeability, providing evidence of the maintenance of a more effective sapwood area. However, there are important limitations in our results. First, due to the atypical wet years studied, this balance is expected to change drastically in drier years (Schiller and Cohen 1998; Ungar et al. 2013). Secondly, the residual evaporation term should be experimentally addressed in order to have a better estimation of errors associated with the global water balance. Thirdly, the vegetation structure of this site is quite simple, with little importance of the understory (even in the 1998 thinning, where scrub weeding is accomplished regularly) meaning that translating these results to other forest plantations need caution. This study has also identified some questions and relationships between the measured variables (e.g. transpiration and BAI, soil properties in H98) that require further evaluation and should be the subject of successive studies in drier years. In any case it is evident from our view the

importance of these water-centred studies in the field of forest management.

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**Complementary materials**

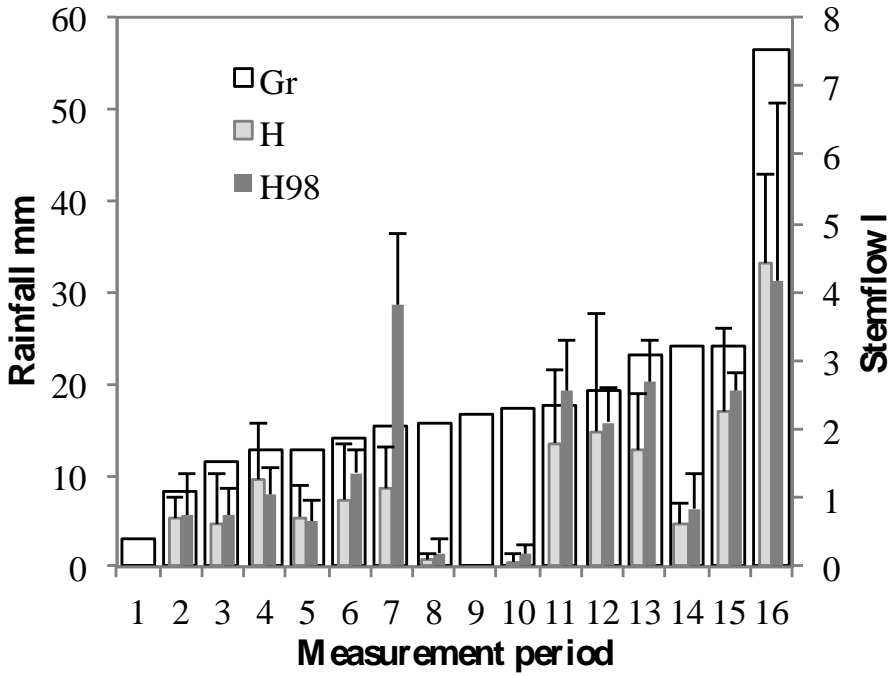


Fig. 1 - Mean and standard deviation values of tree stemflow (l) and rainfall (mm) in the different spells studied for this variable

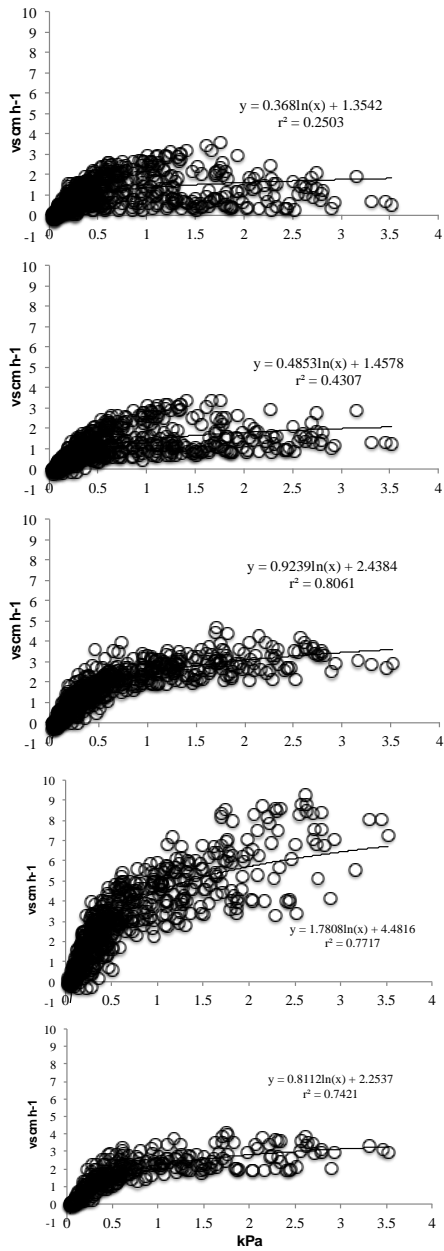


Fig. 2 - Mean daily values of sap flow velocity ( $\text{cm h}^{-1}$ ) as a function of the mean daily values of VPD (KPa) for each treatment

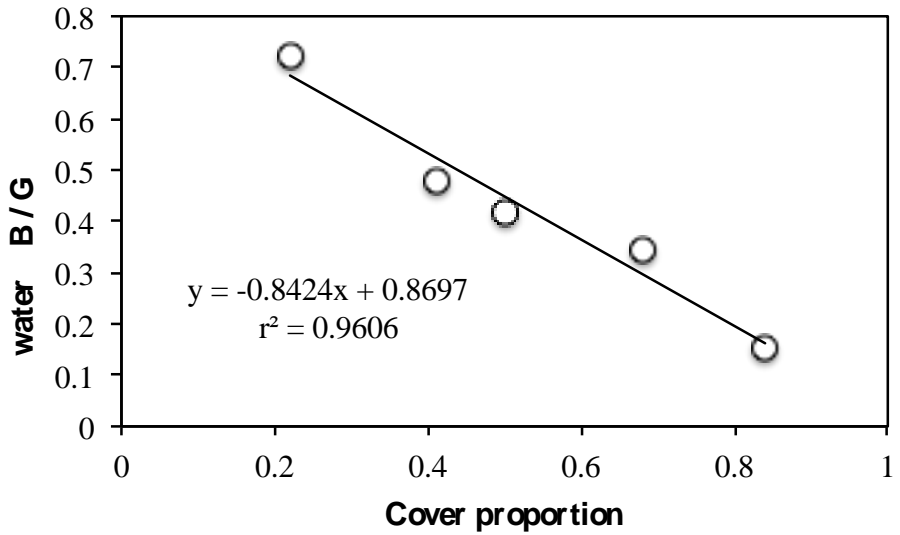


Fig. 3 - Relationship between the blue to green water ratio (B/G) and forest cover (proportion). N= 5. The model was statistically significant at  $p\text{-value} < 0.01$ .



## **CHAPTER FOUR**

Growth, tree water-use and water-use efficiency in response to proactive adaptive silviculture in a stagnated Mediterranean pine forest

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**Growth, tree water-use and water-use efficiency in response to proactive adaptive silviculture in a stagnated Mediterranean pine forest**

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## **Abstract**

In water-scarcity prone regions, adaptive management has been proposed to focus on the forest and water relationships, i.e. on the implementation of a hydrology-oriented silviculture that should consider both the stand reducing total transpiration and competition and the tree levels (improving growth, water-use (WU) and intrinsic water-use efficiency, WUE<sub>i</sub>). The main goal of this work is the evaluation of thinning in a semiarid forest at mid- and short-term on tree growth and physiological processes that condition WU and WUE<sub>i</sub> promoting enhanced forest resilience to face climate change. To this end a stagnated Aleppo pine plantation was experimentally thinned in 1998 at high intensity (H98) and in 2008 including three different intensities, High (H); Medium (M) and Low (L) and a control (C). A substantial limitation of tree-growth imposed by climatic conditions was observed, although thinning changed the tree-growth-precipitation relationships. Significant differences in WUE<sub>i</sub> were found after thinning between H98 and control (mid-term), with higher values in the latter, up to 114.9  $\mu\text{molCO}_2/\text{molH}_2\text{O}$ . However, no significant difference was observed in the thinning made in 2008 (short-term). Despite this, in general WUE<sub>i</sub> decreased when precipitation increased, with different slopes for each thinning intensity. Different

patterns of the relationship between WU and WUE<sub>i</sub> were found, being positive for thinned plots (H, M and L) and negative for C and H98. On the contrary, no relationship was found between  $\delta^{18}\text{O}$  and WU. In the preceding analysis the dual isotope ( $\delta^{13}\text{C}$  and  $\delta^{18}\text{O}$ ) conceptual model held only partially, pointing to the need for more detailed studies, before making a straightforward application of this model to assess changes in water-use. Our results suggest that thinning in Aleppo pine plantations is effective in changing the relationships between WU and WUE<sub>i</sub>, and that these in turn are not dependent of thinning intensities. This paper introduces a novel contribution relating WU and WUE<sub>i</sub> in Mediterranean forest subject to thinning.

**Keywords:** Dendrochronology; Forest management; Hydrology-oriented silviculture; *Pinus halepensis* Mill.; Stable isotopes; Transpiration

## **1. Introduction**

Climate change scenarios predict large increases in temperature and decreases in precipitation in the Mediterranean region (Christensen et al. 2007; Parry et al. 2007). Thus, water resource availability in the Mediterranean will be seriously jeopardized in the foreseeable future (García-Ruiz et al. 2011). Several studies (Parry et al. 2007, Giorgi and Lionello 2008, Estrela et al. 2012) point to the warming of winters and to the increase of both the length of the dry season and the frequency of extreme events in Mediterranean areas. These changes may substantially impact Mediterranean forests; therefore, forest managers need adaptive strategies that aid in achieving the efficiency and effectiveness of forest management under changing water resource availability. Moreover, Sjölund and Jump (2013) warn that adaptive strategies should also be carefully considered in order to improve forest resilience. However, the adaptive capacity in the forest sector is limited in the Mediterranean region where large forest areas are only extensively managed or remain unmanaged (Lindner 2000; Lindner et al. 2010, Torras et al. 2012). Hence, there is a need to develop reliable guidelines for adaptive

management according to the eco-regional and social context of the forest being managed (Fitzgerald et al. 2013).

In water-scarcity prone regions, adaptive management has been proposed to focus on the forest and water relationships, i.e. on the implementation of a hydrology-oriented silviculture (Del Campo et al. 2014). This silviculture should be implemented by considering both the stand structure and the tree levels. In the former, the focus is more related to the hydrologic performance of the physical structure of the forest (density, LAI, canopy storage etc.), whereas in the latter, the focus is on the water-related physiological performance of the tree species under consideration. Thinning may improve water-use and water-use efficiency and has been proposed as an option in hydrology-oriented silviculture to reduce the use of water as consequence of the reduction of total transpiration and competition among trees (Del Campo et al. 2014). Additionally, thinning results in changes in the forest microclimate, which lead to major changes in the ecophysiological behavior of the tree, with respect to photosynthesis and transpiration phenomena (Aussenac 2000) and consequently the amount of carbon taken up by the tree. Thus, thinning approaches to increase adaptive capacities and to decrease

climate-related vulnerabilities of forests should be based on ecophysiological knowledge (Chmura et al. 2011).

Water-use (WU) is traditionally known as the amount of water transpired by trees that can be measured by sap flow techniques (Burgess et al. 2001; Albaugh et al. 2013). Water-use efficiency WUE in turn, is a measure of the amount of water used per carbon gain (Brienen et al. 2011) and depends upon the particular context in which it is being discussed, including where the water is in relation to the plant, the time scale and the precise measure of efficiency in relation to carbon gain (Bacon, 2004). In most of the literature, WUE is discussed either in terms of an instantaneous measurement of the efficiency of carbon gain per water loss, or as an integral of such an efficiency over time, commonly expressed as the ratio of water use to biomass accumulation, or harvestable yield (Dye 2000; Hubbard et al. 2010).

Plant carbon stable isotope composition ( $\delta^{13}\text{C}$ ) provides a time-integrated proxy of plant intrinsic water-use efficiency (WUE<sub>i</sub>) during the growing season (Farquhar et al. 1989; Warren et al. 2001; Dawson et al. 2002; Klein et al. 2005; Matteo et al. 2010, Andreu-Hayles et al. 2011; Klein et al. 2013). The term intrinsic was introduced to compare photosynthetic properties independent of (or with a common)



evaporative demand (Osmond et al. 1980) and has been widely related to long-term trends in the internal regulation of carbon uptake and water loss in plants (Linares et al. 2011), (see: WUEi theory in Material and Methods). The relationship between  $\delta^{13}\text{C}$  and WUEi exists because the isotope discrimination of plants is linearly linked to the  $C_i/C_a$  ratio, where  $C_i$  is the partial pressure of  $\text{CO}_2$  in the leaf intercellular spaces and  $C_a$  that of the ambient air (Farquhar et al. 1982; Sheidegger et al. 2000). However, increases in  $\delta^{13}\text{C}$ , interpreted as a reduction of  $C_i$ , can be the result of either i) reduced stomatal conductance ( $g_s$ ) (at a constant photosynthetic capacity- $A$ ) or ii) increase in  $A$  (at a constant  $g_s$ ). In addition,  $\delta^{18}\text{O}$  in plants is mainly determined by the isotopic composition of the soil water, the leaf water enrichment due the transpiration (water-use), and biochemical fraction during incorporation (Sheidegger et al. 2000). The idea is that  $\delta^{18}\text{O}$ , as a proxy for evaporative flux, will be modified by stomatal conductance ( $g_s$ ) but not by carbon assimilation ( $A$ ). In other words, while  $\delta^{13}\text{C}$  does not provide any indication of whether changes in WUEi are due to changes in photosynthesis or transpiration, if tree-ring  $\delta^{18}\text{O}$  increases with  $\delta^{13}\text{C}$  this would indicate that changes in WUEi were caused by a reductions in stomatal conductance rather than increases in photosynthesis (Silva and Horwath 2013). This approach (the

dual-isotope model-Sheidegger et al. 2000) could be used successfully in the analysis of WU and WUE in hydrology-oriented silviculture. Many studies (Warren et al. 2001; McCarroll and Loader 2004; Loader et al. 2011; Keenan et al. 2013; Battipaglia et al. 2013; Silva and Horwath 2013 and references therein) have used the carbon isotopes to quantify WUE<sub>i</sub> in trees. In recent years the number of studies that relate the WUE<sub>i</sub> and forest management have increased (Martin-Benito 2010; Brooks and Mitchell 2011; Forrester et al. 2012; Kruse et al. 2012), but fewer studies have been carried out in Aleppo pine plantations under different thinning intensities (Moreno-Gutiérrez et al. 2012). In trees where WU has been assessed by the sap-flow method, the dual isotope approach would help to unravel the effects of climate on forest growth and water use efficiency after thinning. Thus, studies on the WU and WUE<sub>i</sub> of Aleppo pine plantations are helpful when attempting to understand and quantify how much water the plants need and to determine how future climate-warming-induced hydrological changes will impact the *Pinus halepensis* Mill. (Aleppo pine) forest, especially in Mediterranean areas due to this species remarkable ability to withstand drought stress (Moreno-Gutiérrez et al. 2012), and because it is one of the most ecologically and economically important species of the

Mediterranean region (Ne'eman and Trabaud 2000). Hence, the study of water use (WU), water-use efficiency (WUE) and growth as they respond to thinning in *P. halepensis*, is justified in order to achieve a better understanding of the interactions between the water cycle and Aleppo pine forests in a Mediterranean context where natural and human-driven changes in land use are taking place under a global change framework.

The main objective of this work is the evaluation of thinning in a semiarid forest as a practice hydrology-oriented silviculture on a tree scale, i.e., on the plant physiological processes that condition tree-water use and promote a better resistance/adaptation to climate change. This paper addresses the following questions: i) how do the thinning treatments affect the tree growth and water-related physiological processes? ii) What is the influence of time elapsed since thinning and thinning intensity on these tree traits? iii) How do the changes on these traits respond to climate irregularity? To address these objectives, dendrochronological approach and isotopes analyses ( $\delta^{13}\text{C}$  and  $\delta^{18}\text{O}$ ) were used to evaluate growth and WUE<sub>i</sub> (A and g<sub>s</sub>) in one plot thinned in 1998 at high intensity, three plots thinned in 2008 at high, medium and low intensities, and in a control plot. This was applied to the same trees

where water-use was estimated by sap flow method (Del Campo et al. 2014). Additionally we address the question of whether the dual-isotope model (Sheidegger et al. 2000) can be applied to test the agreement between measured water-use and predicted responses by using tree-ring  $\delta^{13}\text{C}$  and  $\delta^{18}\text{O}$ .

## **2. Material and Methods**

In order to study the effects of thinning on water-use, tree transpiration and meteorological variables were registered during 2 years in 5 Aleppo pine stands. To analyse the influence of thinning on growth and intrinsic water-use efficiency at short and mid-term, a dendrochronological approach combined with stable isotopes analyses was carried out. Finally relationships between growth, environmental data (Temperature and Precipitation) and stable isotopes ( $\delta^{13}\text{C}$  and  $\delta^{18}\text{O}$ ) in tree-rings were studied.

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

The experimental site, “La Hunde”, is located in the Ayora valley region (39° 5′ N; 1° 13′ W, 943 m a.s.l.), in the Southwest of Valencia province (Spain). The climate is Mediterranean with an average

total annual precipitation of 477 mm and a mean annual temperature of 14.1 °C (1960-2010). The soils are relatively shallow (50-60cm), display a basic pH of 7.6 and are of sandy-silty loam texture. Precipitation is concentrated from October to May representing 75.7% of total rainfall; the dry season usually occurs in July and August contributing only with 7.3%. When comparing the averages for the period 1960-2010 with those for 1995-2010, a slight decrease in total precipitation and an increase in average temperature (Fig. 1) can be observed. Twenty two percent of the area is occupied by *Pinus halepensis* Mil. plantations ranging between 50-60 years old and with high tree density (ca 1500 trees/ha) due mainly to low forest management.

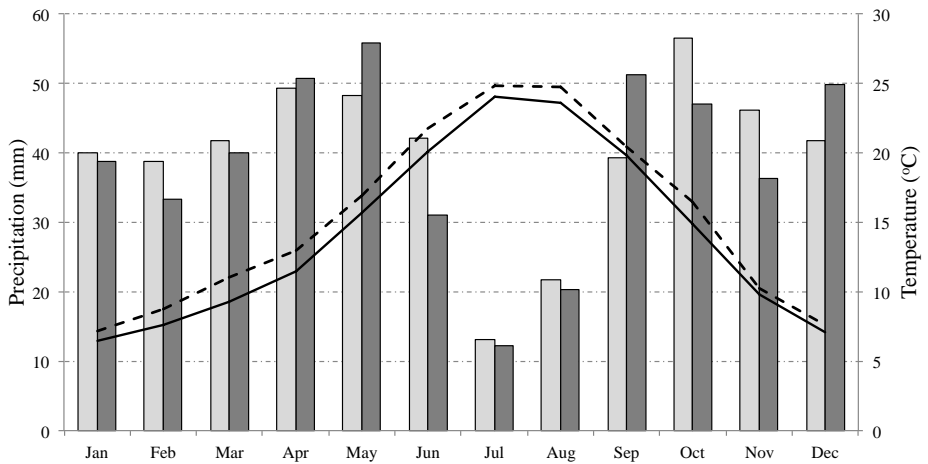


Fig. 1 - Climatic diagram. Grey and dark grey bars indicate total monthly precipitation (mm) in the period 1960 to 2010 and 1995 to 2010 respectively. Continuous line and dashed line represent monthly mean temperature ( $^{\circ}\text{C}$ ) in the period 1960 to 2010 and 1995 to 2010 respectively.

The experimental set-up is the same described in Del Campo et al. (2014), where an area was heavily thinned in 1998 leaving approximately 10% of the trees (H98). In this area a plot was established and sampled to assess the mid-term effects of thinning. Adjacent to this area another experimental area was set consisting of a randomized block design with three blocks, 0.36 ha each. Each block was further divided into four plots (30x30m), three of them corresponding to thinning treatments performed in 2008 at different intensities (High-H, Medium-M and Low-L) and a control plot (C), common to both experimental areas (Table 1).

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

<b>Treatment</b>	<b>Cover (%)</b>	<b>Density (trees ha<sup>-1</sup>)</b>	<b>DBH (cm)</b>	<b>Mean height (m)</b>	<b>BA (m<sup>2</sup> ha<sup>-1</sup>)</b>
Control (C)	84	1489	17.8 ± 5.1	11.5	40.1
Low intensity (L)	68	744	21.2 ± 4.1	12.2	27.2
Medium intensity (M)	50	478	21.7 ± 4.0	11.3	18.2
High intensity (H)	22	178	20.4 ± 1.6	12.2	9.4
High intensity-1998 (H <sub>98</sub> )	41	155	25.2 ± 5.0	12.6	13.6

## 2.2. Measurement of data

Water use and intrinsic water use efficiency were measured in one representative block, with three treatments (H, M and L) plus control (C) plot and the block thinned in 1998 (H98) from April 2009 to May 2011. For dendrochronological studies trees from all blocks were sampled by coring in Jun 2011 and isotopes analyses were carried out in April 2012.

### 2.2.1. Dendrochronological approach

Two 5mm increment cores were taken from each tree (north and south), to integrate the potential variability in tree growth around the stem and to help identify false or missing tree-rings; they were taken at 1.30 m in each tree after the completion of the transpiration

measurements in the same trees. Additionally thirty-two trees, four trees per plot in each block were cored to avoid the possible under-estimation of the climate-growth correlations (Mérian et al., 2013), except for Medium thinning intensity because one of the plots was badly damaged due to strongly winds in 2009.

Each core was mounted on a wooden support and sanded until wood cells were clearly identified under the stereomicroscope. All cores were visually cross-dated and measured to the nearest 0.01mm with a measuring table (LINTAB 6.0, Frank Rinn, Heidelberg, Germany) coupled with the TSAP-Win software package (Rinn 2011). Some cores were also verified with scan ImageJ software (Rasband 2011) to enhance contrast. Cross-dating of the tree-ring width (TRW) series was evaluated using the COFECHA programme (Holmes, 1983) and with the tutorial by Grissino-Mayer (2001), additionally the dplR R-package (Bunn 2008) was used to better visualize possible errors. The cores that presented missing rings were discarded in the following analyses. The average series length was 50.2 ( $\sigma=2.51$ ) years, autocorrelation at 1-year averages was 0.76 ( $\sigma=0,09$ ), and Gini coefficient, which describes annual changes in the inequality of size and size increment was 0.42 ( $\sigma= 0.08$ ), where 0



indicates perfect equality (the size or growth of all individuals is the same) and 1 indicates perfect inequality. The average series correlation to the master chronology is 0.79 ( $\sigma=0.07$ ), excluding the High intensity thinning made in 1998, which were analysed separate, obtaining a correlation of 0.80 ( $\sigma=0.09$ ).

To perform the dendroclimatic analysis, the TRW series were detrended using DetrendeR – R package (Campelo et al. 2012) and dplR – R package (Bunn 2008), to reduce the noise caused by systematic non-climatic change due to tree age (Cook and Briffa 1990), using a cubic smoothing spline function a wavelength fixed at 67% (proximally  $2/3$  – Cook et al. 1990) of the length of the series, with a 50% frequency response. In some cores a negative exponential method was utilized. Each measured series was standardized dividing observed values by predicted values to obtain dimensionless TRW index series (TRWi). TRWi was averaged using a robust bi-weight mean. Additionally the temporal autocorrelation was removed from each series using an autoregressive model (Cook and Briffa 1990) obtaining the standard and residual chronology. To determine the length of the residual chronology for which climatological responses would be tested, we used a running

(20-year) mean of the expressed population signal (EPS) statistic (Wigley et al. 1984) to provide an indication of chronology reliability.

### **2.2.2. Carbon and Oxygen isotopes analysis**

Carbon and Oxygen data are presented as  $\delta^{13}\text{C}$  and  $\delta^{18}\text{O}$  respectively relative to the international VPDB standard:  $\delta[\text{‰}] = [(R_{\text{sample}}/R_{\text{standard}})-1] * 1,000$ ; where:  $R_{\text{sample}}$  and  $R_{\text{standard}}$  are the ratios of  $^{13}\text{C}/^{12}\text{C}$  or  $^{18}\text{O}/^{16}\text{O}$  in the sample and standard expressed in parts per thousand (‰).

$\delta^{13}\text{C}$  is inversely and linearly correlated with  $\text{C}_i/\text{C}_a$ , the ratio of intercellular to atmospheric  $\text{CO}_2$  concentrations in leaves. This ratio reflects the relative magnitudes of net photosynthesis rate ( $A$ ) and stomatal conductance ( $g_s$ ), and thus  $\delta^{13}\text{C}$  is a good indicator of plant intrinsic water use efficiency ( $\text{WUE}_i$ ), which is given by the ratio  $A/g_s$  - Eq. 6 (Farquhar et al. 1989). On the other hand  $\delta^{18}\text{O}$  of plant organic material is related to  $e_a/e_i$  the ratio of atmospheric to leaf intercellular water vapour pressure, and thus is strongly affected by changes in  $g_s$  (Scheidegger et al. 2000; Barbour 2007). Since  $\delta^{18}\text{O}$  is related to  $g_s$ , but is unaffected by  $A$ , it can help separate the independent effects of  $A$  and

gs on WUEi (Scheidegger et al. 2000; Barbour 2007; Moreno-Gutiérrez et al. 2011). For this propose, analyses of  $\delta^{18}\text{O}$  and  $\delta^{13}\text{C}$  were performed on subsamples of rings from three trees per plot in the transpiration sensors block (see: 2.1 section). Stable isotopes in the rings for the period 1995 to 2010 for Control and High-98 were analysed in order to explore the effect of thinning for a longer period of time and for the period 2004 to 2010 for the more recent thinning treatments. Samples from the previously measured and dated cores were taken for isotopic analyses. The sanded core surfaces were thoroughly cleaned with ethyl alcohol p.a. and small sections, including both early and latewood, were taken with a surgical blade under the stereomicroscope. These samples were ground in an agate mortar and weighed to the nearest 0.001g in Ag capsules for  $\delta^{18}\text{O}$  analysis and Sn capsules for  $\delta^{13}\text{C}$  analysis.

The stable carbon and oxygen isotopic signatures of the wood samples were measured online using an isotope ratio mass spectrometer (Finnigan MAT, delta S, delta XL plus, delta XP) via a Conflo II interface for the combustion/pyrolysis of organic material at the Paul Scherrer Institut, Ecosystem Fluxes Research Group, Villigen, Switzerland.

### 2.2.3. Water-use efficiency

The isotopic discrimination ( $\Delta$  - Farquhar and Richards, 1984) between carbon from atmospheric  $\text{CO}_2$  and plant carbon from  $\text{C}_3$  plants, which occurs as a result of preferential use of  $^{12}\text{C}$  over  $^{13}\text{C}$  during photosynthesis, is defined in Eq. 1.

$$\Delta = (\delta^{13}\text{C}_{\text{atm}} - \delta^{13}\text{C}_{\text{wood}})/(1 + \delta^{13}\text{C}_{\text{wood}}/1000) \quad \text{Eq. 1}$$

where  $\delta^{13}\text{C}_{\text{atm}}$  and  $\delta^{13}\text{C}_{\text{wood}}$  are the isotopic ratios of carbon ( $^{13}\text{C}/^{12}\text{C}$ ) in atmospheric  $\text{CO}_2$  and plant material, respectively.

$\Delta$  is linearly related to  $\text{C}_i/\text{C}_a$ , which is the ratio of intercellular ( $\text{C}_i$ ) to atmospheric ( $\text{C}_a$ )  $\text{CO}_2$  mole fractions, by Eq. 2 (Farquhar et al. 1982):

$$\Delta = a + (b - a)\text{C}_i/\text{C}_a \quad \text{Eq. 2}$$

where  $a$  is the fractionation during  $\text{CO}_2$  diffusion through the stomata (4.4‰; O'Leary 1981), and  $b$  is the fractionation associated with reactions by Rubisco and PEP carboxylase (27‰; Farquhar and Richards 1984).  $\text{C}_a$  and  $\delta^{13}\text{C}_{\text{atm}}$  were obtained for each year using the relations found by Feng (1999) by means of equations 3 and 4 respectively.

$$\text{C}_a = 277.78 + 1.35 \times e^{0.01572 \times (\text{year} - 1740)} \quad \text{Eq. 3}$$

$$\delta^{13}\text{C}_{\text{atm}} = -6.429 - 0.0060 \times e^{0.0217 \times (\text{year} - 1740)} \quad \text{Eq. 4}$$

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

$$C_i = \delta^{13}C_{\text{atm}} - \delta^{13}C_{\text{wood}} - a/b - a \quad \text{Eq. 5}$$

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

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

#### 2.2.4. Water use

The heat ratio method (HRM) allows the measurement of sap flow in woody parts of tree trunks and the derivation of transpiration estimates for trees and stand (Burgess et al. 2001). Advantages of the sap flow technique are that measurements are stable and reliable at short temporal scales, and can be used in any terrain and this makes the technique especially suited for physiological studies at plant level (Asbjornsen et al. 2011; Aranda et al. 2012; Sun et al. 2012). Thus, the sap flow can be considered a valuable method to estimate the tree water-use (Zeppel et al., 2008). Total transpiration by the Aleppo pine forest (Tr, mm day<sup>-1</sup>) per each treatment, was calculated by multiplying the daily transpiration (L day<sup>-1</sup>tree<sup>-1</sup>) by the frequency of diameter class (f)

and stocking density ( $N^{\circ}$  trees  $ha^{-1}$ ). When data series presented gaps, they were completed by means of Artificial Neural Network.

To describe the relationships between water-use and water-use efficiency, we considered as "physiological year" the water-use values starting in April to March of the following year. This approach is consistent with growth dynamic of Aleppo pine trees (De Luis et al. 2007, 2011).

### **2.2.5. Meteorological variables**

Measurements of precipitation (mm) and air temperature ( $^{\circ}C$ ) were carried out by a single sensor (HR/T sensor, Decagon Devices, Pullman, USA) placed at 1 m height and close to all treatments for the period 2008-2011, values were recorded every 10 min and averaged every 30 min. The value of average precipitation and temperature from the period 1960-2008 was obtained in the Ayora and Almansa meteorological stations nearest to the study site. Additionally the data was grouped seasonally following the same approach than for the atmospheric circulation patterns as described by Pasho et al. (2011): Spring (April-May), Summer (June-August), Autumn (September-

November) and winter (December-March) (Fig. 1-Complementary materials).

To analyse the influence of drought on tree growth since the responses of forest thinning, we used the Standardized Precipitation Index (SPI) developed by McKee et al. (1993) and calculated using R-package SPEI index (Beguería and Vicente-Serrano 2013). The SPI was calculated by adjusting the precipitation series to a given Pearson III distribution probability that was considered more robust due to its three parameters (Pasho et al. 2011). Positive SPI values indicate greater than median precipitation and negative values indicate lower than median precipitation. For the purpose of this study we consider that a drought event starts when SPI value reaches -1.0 and ends when SPI becomes positive again.

### **2.3. - Data treatment and analysis**

All statistical procedures, except crossdating check were performed using R 3.0.2 GUI 1.62 (R Core Team, 2013). Pre-treatment (1995 to 1998 or 2004 to 2007) and post-treatment (1999 to 2010 or 2008 to 2010) differences for TRW,  $\delta^{13}\text{C}$ ,  $\delta^{18}\text{O}$ ,  $\Delta^{13}\text{C}$  and WUE<sub>i</sub> were analysed. Before the analyses, data were tested for variance normality

and homogeneity by Shapiro-Wilk test and Levene tests, respectively. The t-student test and Kruskal-Wallis test were the procedures used for mean comparisons between the treatments in each of the variables considered, using the R-package *Agricolae* (Mendiburu 2014). A Linear Model (LM) was used to test the difference in the slopes among treatments after thinning. To verify the impact of thinning on WUE<sub>i</sub> and  $\delta^{18}\text{O}$  the data were divided into dry years and wet years, we consider a year as dry when the sum of SPI- 3 month values was less than zero and wet when greater than zero.

### **2.3.1. Dendroclimatic analysis**

To verify the influence of thinning on the dendroclimatic responses, Control and High 98 residual chronologies were used. The relationship between each residual chronology (TRW<sub>i</sub>) of each plot and climatic data (Temperature, Precipitation, SPI and Seasons) for the common period (1960-2010) was determined by bootstrapped response functions (Guiot 1991) and to investigate the stability of growth-climate relationships it was used the moving response by *bootRes* R-package (Zang and Biondi 2013). The coefficients of the response functions are obtained via principal-component regression, which produces reliable



regression coefficients in case of multi-collinearity (Fritts 1976).

Correlation analysis was made using Pearson's product moment correlation. Significance of correlation and response function coefficients were tested at 0.05 level using 1000 bootstrapped estimates, obtained at random with replacement from the initial data set (Biondi and Waikul 2004; Wang et al. 2012; Zang and Biondi 2013). The temporal window used for calculating growth-climate relations extends from the previous April to October of the growth year for the common period (Magruder et al. 2013), which was considered adequate to explain the tree-ring width variations (Beck et al. 2013).

### **3. Results**

#### **3.1. Influence of thinning at short and mid-term**

Tree growth, expressed as TRW, showed a general variability in the initial growth of the stand (ca 15 years), likely related to genetics and environmental conditions, followed by a progressive decrease and stagnation for all trees, probably related to increase in competition between trees (Fig. 2a). In the following years TRW variations, were very small but still showing a clear response to climatic conditions, especially in dry years (e.g. 1987-1988; 1994-1995 and 2004-2005). Immediately after thinning remaining trees showed significant increase in TRW, when compared with control trees (Fig. 2a and 2b). Table 2 summarizes the differences for mean TRW before and after thinning. There were significant differences ( $p < 0.05$ ) between C and H98 after thinning (mid-term analysis). However in the short-term analysis no significant differences ( $p < 0.05$ ) were found between C and L and between L and M thinning treatments. TRW in H is significantly different ( $p < 0.05$ ) after thinning from the other treatments.

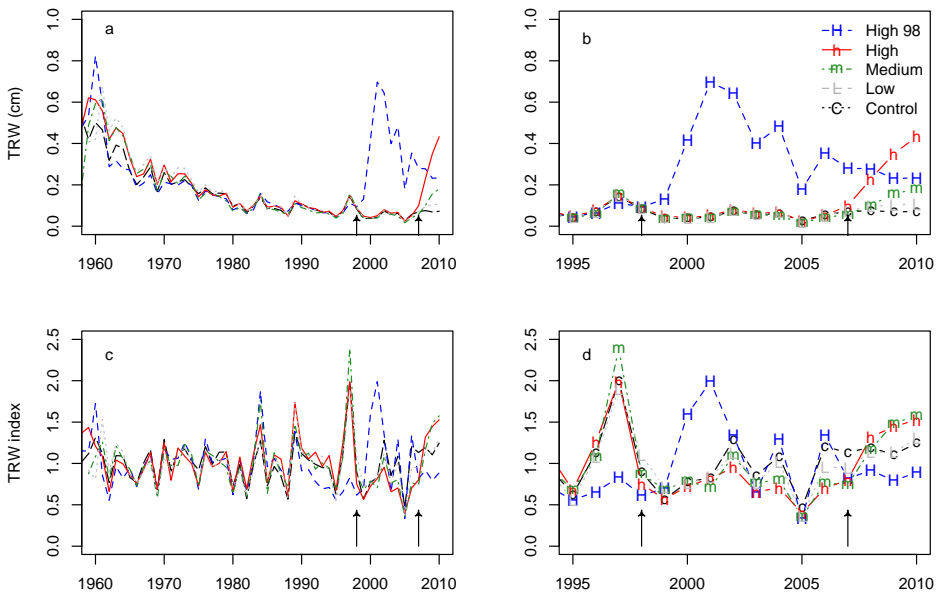


Fig. 2 - Average Tree Ring Width (TRW) in cm for all treatments; time span 1960 to 2010 (a) and 1995 to 2010 (b). c and d are the residual chronologies (TRW index) for all treatments; time span 1960 to 2010 and 1995 to 2010 respectively. Arrows indicate the thinning times.

Expressed population signals (EPS) of all TRW residual chronology were above the critical value of 0.85 (Wigley et al. 1984) and the mean of the correlations between series from each tree over all trees, also indicate a good agreement ( $>0.80$ ). In all chronologies we observed index reductions in years characterized as dry years. On the other hand, the periods characterized by substantial increase in radial growth were observed in wet years and after thinning at different scales (Fig. 2). Despite the observed graphical differences after thinning (Fig. 2b) no

significant differences ( $p < 0.05$ ) were found in the TRW index before and after thinning (data not shown).

Table 2 - Mean values of Tree Ring Width (TRW) in cm; intrinsic Water Use Efficiency (WUEi) in  $\mu\text{molCO}_2/\text{molH}_2\text{O}$  and Oxygen isotopic signature  $\delta^{18}\text{O}$  in ‰. Mean values followed by the same letter did not differ by the Kruskal-Wallis test ( $p < 0.05$ ) at short-term, or t-test ( $p < 0.05$ ) at mid-term.

Time span	Thinning	Plots (intensity)	TRW*	WUEi	$\delta^{18}\text{O}$
Mid-term (1995-2010)	Before ('95-'97)	Control	0.09 (a)	98.5 (a)	28.7 (a)
		High-98	0.06 (a)	97.4 (a)	28.9 (a)
	After ('98-'10)	Control	0.06 (b)	98.8 (a)	28.3 (b)
		High-98	0.27 (a)	95.6 (b)	29.5 (a)
Short-term (2004-2010)	Before ('04-'07)	Control	0.05 (a)	100.4 (a)	28.2 (a)
		High	0.07 (a)	101.8 (a)	28.3 (a)
		Medium	0.06 (a)	96.4 (a)	28.1 (a)
		Low	0.05 (a)	100.5 (a)	28.2 (a)
	After ('08-'10)	Control	0.07 (c)	94.4(a)	28.2 (b)
		High	0.42 (a)	97.6 (a)	29.3(a)
		Medium	0.19 (b)	94.1 (a)	29.4 (a)
		Low	0.12 (bc)	92.1 (a)	28.5 (ab)

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

WUE<sub>i</sub> ranged from 80.6 to 114.9  $\mu\text{molCO}_2/\text{molH}_2\text{O}$ . Significant differences ( $p < 0.05$ ) were found after thinning between H98 and C (mid-term analysis) (Table 2), with higher values in the control trees. However in the year 2003 a change of pattern was observed between both plots (Fig. 3a). No significant difference was observed in the short-term analysis (Table 2), although in the single year comparison (Fig. 3), the H plot showed a marginal significant difference ( $p < 0.1$ ) and higher WUE<sub>i</sub> than C immediately after thinning (2008) (Fig. 3b), probably due to the increase in growth-rate. However, in the year 2010 the C plot showed slightly higher values of WUE<sub>i</sub> in all treatments (Fig. 3).

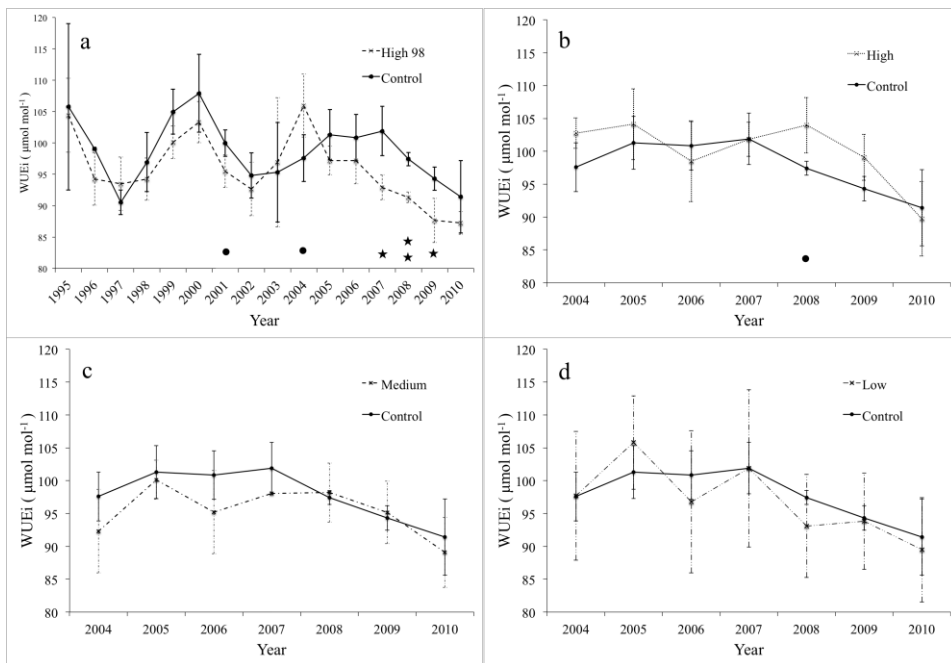


Fig. 3 - Intrinsic Water Use efficiency (WUEi) by single years. a – Control and high intensity thinned in 1998, b – Control and high intensity thinned in 2008, c – Control and Medium intensity thinning and d – Control and Low intensity thinning. Data shown are average values  $\pm$  SD. Stars near the axis indicate significant difference ( $p < 0.05$ ), double star ( $p < 0.01$ ), and dot ( $p < 0.1$ ) in the year.

Values for Oxygen isotopic signature ( $\delta^{18}\text{O}$  in ‰) ranged from 26.4 to 32.1‰. Before thinning values were very similar among all individual trees (average =  $28.5 \pm 0.84$ ). Mid-term analysis (1998-2010) showed significant difference ( $p < 0.05$ ) between the H98 and C after thinning. Over their common period after thinning the  $\delta^{18}\text{O}$  (‰) values of the H98 trees (Fig. 4a) was on average 1.8 ‰ higher than that of the Control trees from 1998 to 2004. In the period 2005-2009 this difference

was on average only 0.3 ‰, becoming higher again in 2010. In summary,  $\delta^{18}\text{O}$  (‰) increase related to thinning was more consistent year to year, with significant differences noted for the first years and these differences became more significant in dry years, as in the period 1999-2001 and 2003-2004 (Fig. 4a). In the short-term analysis (2004-2010), H and M  $\delta^{18}\text{O}$  were significantly different from C after thinning (2008-2010) and non-significant differences were found between C and L (Table 2). Despite this,  $\delta^{18}\text{O}$  (‰) showed an increase after thinning, relative to the C plot in all treatments (Fig. 4b-d).

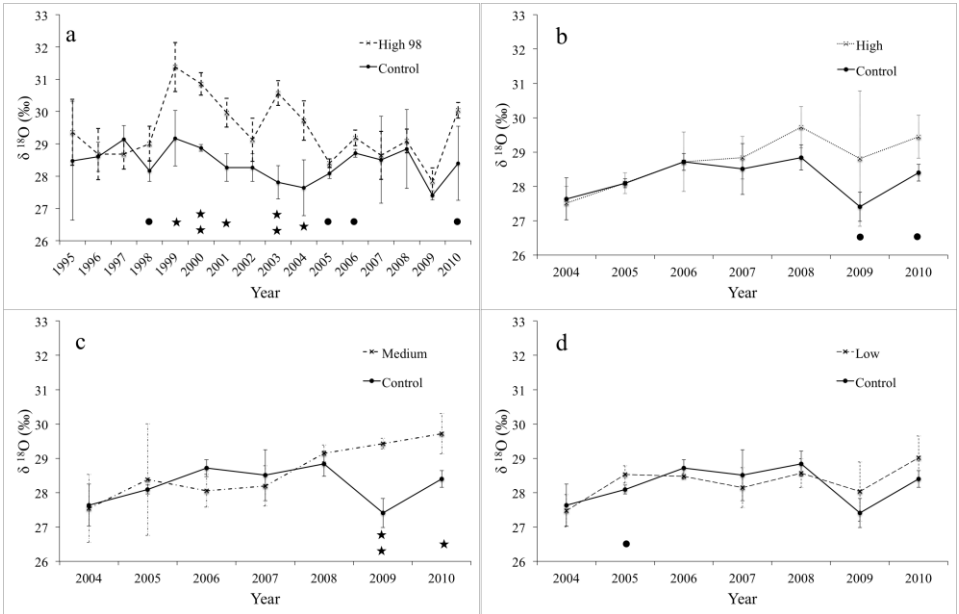


Fig. 4 - Tree-ring Oxygen isotopic signature ( $\delta^{18}\text{O}$  in ‰). a – Control and High intensity thinning in 1998, b – Control and High intensity thinning in 2008, c – Control and Medium intensity thinning and d – Control and Low intensity thinning. Data shown are average values  $\pm$  SD. Stars near the axis indicate significant difference ( $p < 0.05$ ), double star ( $p < 0.01$ ), and dot ( $p < 0.1$ ) in the year.

### 3.2. Relationships with climate after thinning

Analysing the SPI at 1 and 3 monthly time scales, differences were observed between the years 2009 and 2010, noting that 2009 presented a short period of drought ( $\text{SPI} < 0$ ) (Fig. 5). However for 12-month time scale, SPI was positive in both years. In the years 1994-1995



and 2004-2005 the SPI values were negative along the year, in all time scales. Thus, as result a substantial limitation of tree growth imposed by climatic conditions was observed (Fig. 2a-b). Similar SPI patterns were observed in the period 1998-2000, however no negative impact on the growth of H98 trees was observed (Fig. 2b), despite the general low water availability.

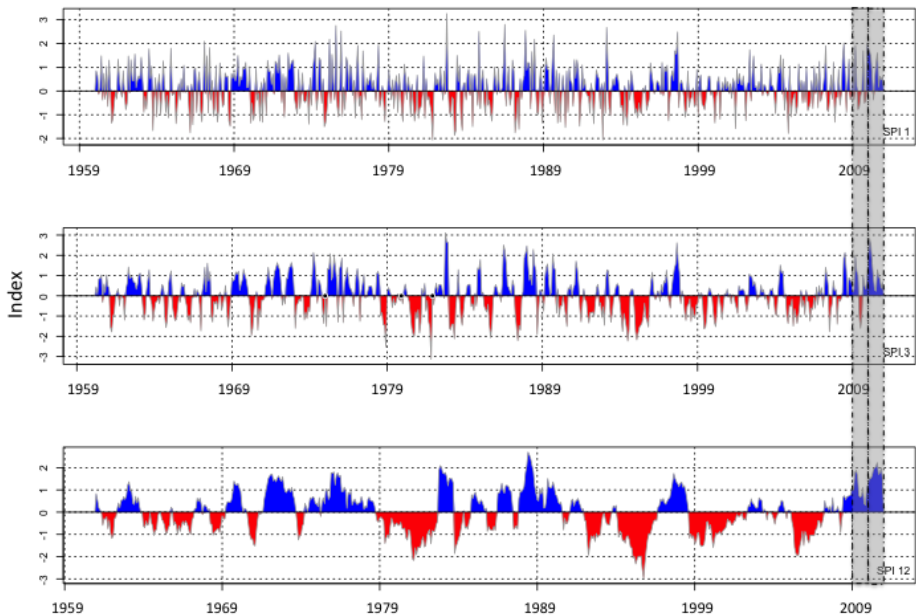


Fig. 5 - Evolution of the drought index (SPI) at different time scales (1, 3, 12 months respectively) in the study area for the period 1961 to 2010. Grey-shaded area refers to years 2009 and 2010 corresponding to the period of measurement of water use (tree transpiration) in this study.

Growth, WUE<sub>i</sub> and  $\delta^{18}\text{O}$  were also compared between H98 and

C trees in dry and wet years, indicating a significant effect of thinning: TRW showed significant differences ( $p < 0.01$ ) in dry and wet years after thinning; significant differences were also obtained for  $\delta^{18}\text{O}$  (‰) after thinning, with  $p < 0.01$  in dry years and  $p < 0.05$  in wet years. However, after thinning there were significant differences in WUEi ( $p < 0.01$ ) in dry years and non-significant in wet years (Table 3).

Table 3 - Mean values of Tree Ring Width (TRW) in cm; Carbon isotopic discrimination ( $\Delta^{13}\text{C}$ ) in ‰; intrinsic Water Use Efficiency (WUEi) in  $\mu\text{molCO}_2/\text{molH}_2\text{O}$  and Oxygen isotopic signature ( $\delta^{18}\text{O}$ ) in ‰. Mean values between treatments followed by the same letter did not differ by the t-test. \* and \*\* indicate significant differences at  $p$ -value  $< 0.05$  and  $< 0.01$  respectively.

Time	Year type	Plots	TRW	WUEi	$\delta^{18}\text{O}$
Before thinning	Dry	Control	0.06 (a)	101.4 (a)	28.3 (a)
		H98	0.06 (a)	99.3 (a)	29.2 (a)
	Wet	Control	0.12 (a)	94.8 (a)	28.9 (a)
		H98	0.07 (a)	93.8 (a)	28.7 (a)
After thinning	Dry	Control	0.04 (b)	100.7 (a)	28.3 (b)
		H98	0.33 (a)**	96.7 (b)**	29.2 (a)**
	Wet	Control	0.06 (b)	97.3 (a)	28.2 (b)
		H98	0.37 (a)**	94.2 (a)	29.5 (a)*

The static bootstrapped correlation function between tree-ring residual chronologies (TRWi) and climate data showed different patterns between control and thinned plots, represented by H98 in this analysis (Fig. 2 – Complementary materials). The negative effect of temperature on the growth index during the previous September was significant ( $p < 0.05$ ) in the C plot while the precipitation signal prevailed over temperature in the current year. We detected high and positive correlation with September precipitation of current year and growth in the C plot, whereas for the H98 plot a high positive and significant ( $p < 0.05$ ) was detected in May of current year (Fig. 2 - Complementary materials). Considering the seasons, the highest correlation coefficients with TRWi for control plot were found for summer precipitation (June to August;  $r = 0.50$ ;  $p < 0.05$ ) and negative and significant correlation for summer mean air temperatures ( $r = -0.44$ ;  $p < 0.05$ ) of current year. In autumn (September to November) precipitation and temperature did not have any significant growth effect on the C, while for the H98 plot, autumn temperature was correlated with growth ( $r = 0.37$ ,  $p < 0.05$ ) (data not shown). When considering the bootstrapped moving response function analysis, care should be taken because growth-climate relationships are not necessarily stable over time (Herrero et al. 2013),

especially after thinning (Magruder et al., 2013). In our study, when this dynamics was considered the bootstrapped moving response function showed different pattern after thinning (1998), (Fig. 6). In this figure it can be observed that in the seasons when thinning has importance, the H98 plot showed less consistent patterns between precipitation of current year and growth (Fig. 6b) than in the C plot (Fig. 6a). In this case, the control showed higher positive correlations of the precipitation from July to September of current year with growth. i.e. a higher dependence on the precipitation during the growing season. Another important difference in this correlation analysis could be detected in June of current year, where the reduction of precipitation during the last 15 years (Fig. 1) induced contrasting responses between C and H98 plots (Fig. 6).

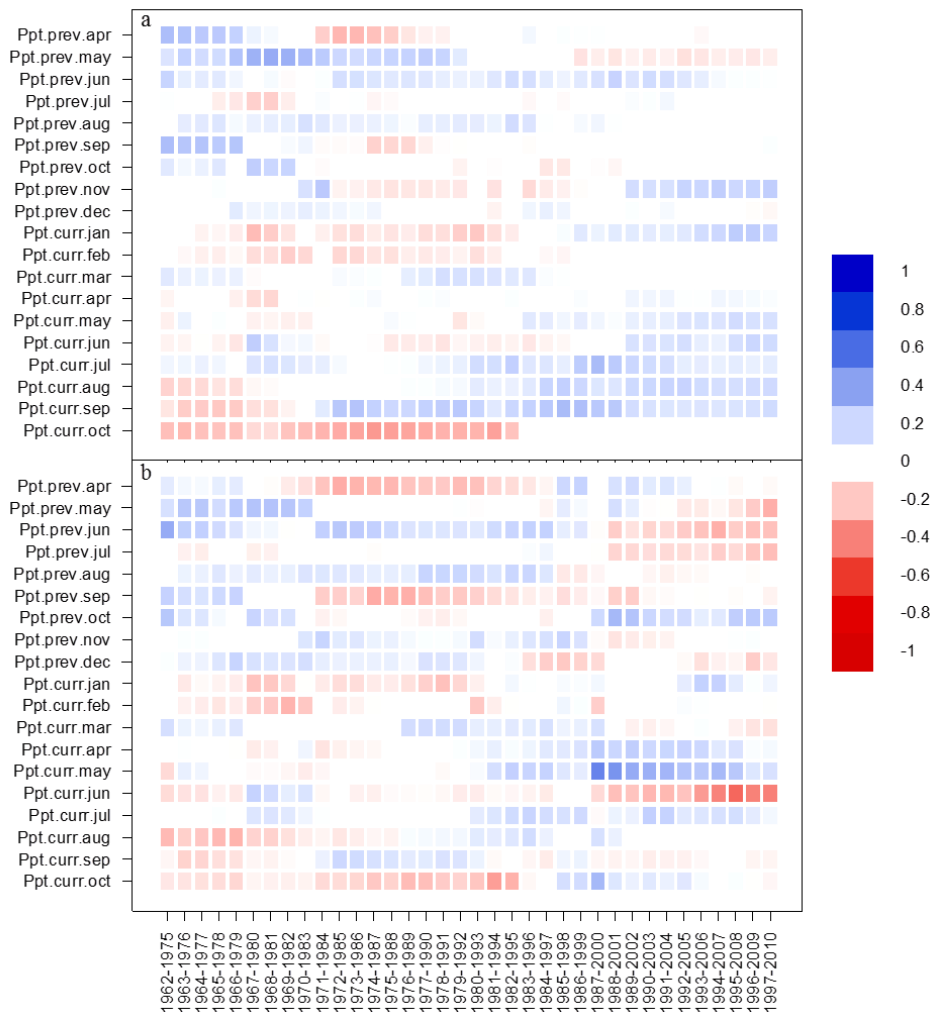


Fig. 6 - Moving response functions of precipitation (Ppt) from April of previous (prev.) year to October of current (curr.) year for a residual tree-ring chronology (TRWi). The window size is fourteen years, and the windows have been offset by one year. a - is Control plot and b - is Thinned plot (H<sub>98</sub>). For a better interpretation of colours in this figure, see the web version of this article.

Independently of the influence of thinning WUE<sub>i</sub> decreased when water supply (precipitation) increased (Fig. 7a;  $r^2=0.48$ ,  $p<0.01$  – all data together), reflecting the general effect of water availability on biomass gain and the water carbon balance; in other words, WUE<sub>i</sub> increased in dry years. However, the response of WUE<sub>i</sub> to precipitation changed with thinning: results showed no significant difference in the slopes of this response ( $F=0.27$ ,  $p\text{-value}=0.85$ ) before thinning (Fig. 7b) and significant differences in the slopes ( $F=11.03$ ;  $p\text{-value}=0.021$ ) after thinning (Fig. 7c).

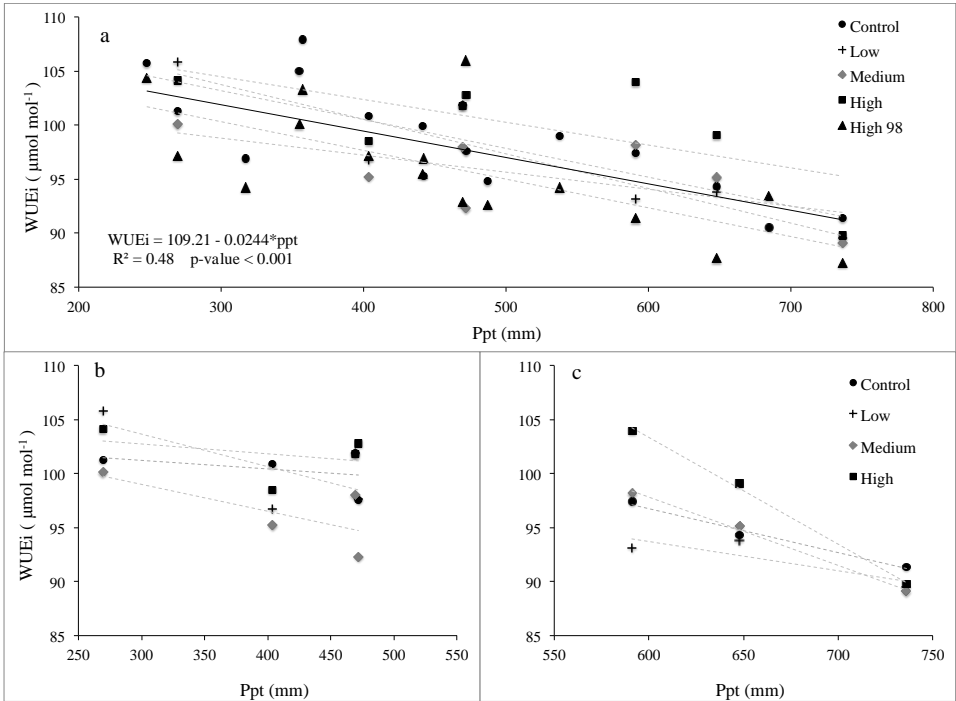


Fig. 7 a – Relationships between mean intrinsic water use efficiency (WUE<sub>i</sub> -  $\mu\text{molCO}_2/\text{molH}_2\text{O}$ ) and total precipitation (Ppt - mm) for all treatments and years together. Black line and equation refers to all treatments together. b – only short-term before thinning (2004-2007) and c – only short-term after thinning (2008-2010)

### 3.3 Water use and its relationships with WUE<sub>i</sub> and $\delta^{18}\text{O}$

Water use, measured as mean transpiration per tree (litres), was highest in the trees in the H plot followed by H98>M>L>C. However when the number of trees per plot was taken into consideration, transpiration at stand level showed the highest water use (litres/ha) in

plot C, but no differences between M and L were found. Considering the seasons we observed that the summer is the period of highest water use in all treatments, followed by spring, autumn and winter (Fig. 3 – Complementary materials). However the pattern is different for each treatment, i.e. in the summer-2009, characterized as a dry-summer in this study (ppt=26.6 mm), the H intensity thinning plot used more water than the other treatments, but in the summer 2010, characterized as a wet-summer (ppt=144.8 mm), the plot C used more water than all other treatments (Fig. 4 – Complementary materials). In other words, when water is a limiting factor in summer, a lower number of trees (less density) allows for a higher tree-water use. A slight difference was observed between the M and H98 treatments when comparing both summer periods.

Among treatments, WU in the years 2009-2010 varied from 475.5 to 8601.6 l/tree while WUE<sub>i</sub> varied from 80.6 to 101.2  $\mu\text{molCO}_2/\text{mmolH}_2\text{O}$ . Different patterns of the relationship between WU and WUE<sub>i</sub> were found in each treatment (Fig. 5 – Complementary materials), despite of non-significant linear regressions (p-value>0.1). At short-term there are positive relationships between WU and WUE<sub>i</sub> for thinned plots together (H, M and L) despite the low coefficient of



correlation (Fig. 8a). However, for C and H98 in the period of analyses (2009-2010) a negative non-significant relationship was observed (Fig. 8b-c). On the contrary, no relationship was found between  $\delta^{18}\text{O}$  and WU (Fig. 8d-f).

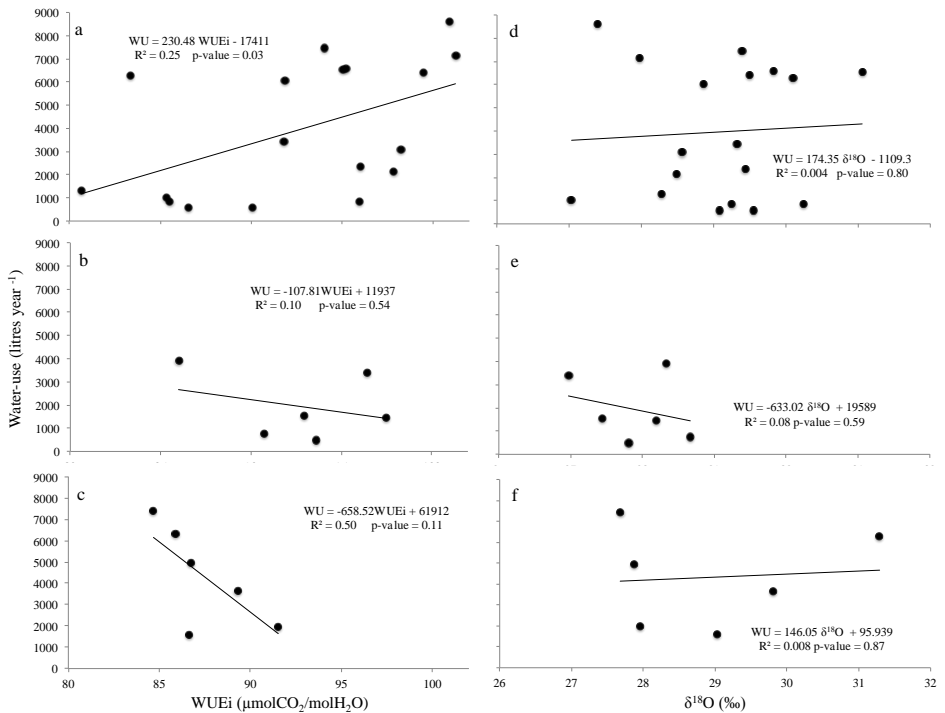


Fig. 8 a, b and c – Relationships between WU (litres year<sup>-1</sup>) and WUEi (μmolCO<sub>2</sub>/molH<sub>2</sub>O) for thinned data (H, M and L together), Control and High 98 respectively. d, e and f – Relationships between WU (litres year<sup>-1</sup>) and δ<sup>18</sup>O (‰) for thinned data (H, M and L together), Control and High 98 respectively

## **4. Discussion**

Hydrology-oriented silviculture by thinning might help adapt Mediterranean forest to climate changes. However achieving this goal demands a better understanding and quantification of water use and water use efficiency. Here, the effects of thinning at short and mid-term were studied by focusing on the water use and water use efficiency and their relationships in a planted pine forest in a semiarid climate.

### **4.1. Interpreting trends in growth, $\delta^{18}\text{O}$ and WUEi**

After thinning there are significant differences in growth among experimental treatments, while we found no significant differences before, suggesting that the thinning treatments were able to induce differences in tree growth rates. Both High and Medium thinning treatments were able to induce a high growth rate that did not occur under the Low intensity thinning treatment. However, when compared within the same thinning intensity H98 vs H, but at different times (1998 and 2008 respectively), the treatment thinned earlier showed a higher initial growth rate than the treatment thinned in 2008, showing that the delay in the decision to thinning could hampers initial growth-rates. Also,

H98 showed lower WUE<sub>i</sub> and higher  $\delta^{18}\text{O}$  than the control plot, suggesting that thinning changes important physiological attributes of remaining trees at mid-term, due a reduction of competition for water resources. These results are in agreement of with those found by Brooks and Mitchell (2011) where significant  $\delta^{18}\text{O}$  increases were associated with the time since thinning, being most significant those differences with control values soon after thinning. However, Martín-Benito et al. (2010) suggest that increases in  $\delta^{18}\text{O}$  might be due to the hotter and drier environment induced in the stand after thinning, and that this increase might not be related to changes in stomata conductance (gs). At short term significant differences between control and thinned plots were observed in  $\delta^{18}\text{O}$ , but no significant differences among thinned plots (H, M and L) were found. Similar results were found by Moreno-Gutiérrez et al. (2011) who studied the effects of two thinning intensities (High and Medium) in Aleppo pine under Mediterranean conditions. Regarding WUE<sub>i</sub> at short-term, we believe that the lack of differences is due to the fact that the years after thinning were very similar in terms of overall water availability, despite the different seasonal distribution (Fig. 1 - Complementary materials). However, in Fig. 7 we can observe that

WUE<sub>i</sub> values respond differently to each thinning intensity in relation to total precipitation. These significant differences among the slopes of WUE<sub>i</sub> vs precipitation suggest that WUE<sub>i</sub> responds to water availability only when thinning was most intense (Fig. 7c). Gyenge and Fernández, (2014) report that thinning can increase the absolute amount of water reaching the soil due to the decrease in precipitation interception. It is interesting to note that in our case a similar response was observed using net precipitation values taken from Del Campo et al. (2014) instead of total precipitation, i.e. the different slopes between WUE<sub>i</sub> and precipitation in each thinning intensity can not be attributed to an increase in water availability due to canopy reduction, but rather to a decrease in competition among trees. Indeed, high intensity thinning (H) showed higher WUE<sub>i</sub> values immediately after thinning compared with C trees (Fig. 3b), indicating an increase in photosynthetic capacity (A), since the water-use also increased in H trees. On the contrary, M and L showed lower WUE<sub>i</sub> values than C, probably due to higher values of stomatal conductance. Undeniably, reduced A, which would also produce similar results on WUE<sub>i</sub>, are highly unlikely in these thinning plots due to the growth rates observed after thinning.

In the selected dry years WUE<sub>i</sub> and  $\delta^{18}\text{O}$  were clearly affected by thinning, indicating that forest management induced physiological changes in remaining trees. These effects are enhanced specially in dry years. Our results are consistent with those of Moreno-Gutiérrez et al. (2012), who in two sharply contrasting stands of *Pinus halepensis* found strong correlation between WUE<sub>i</sub>,  $\delta^{18}\text{O}$  and growth with annual precipitation, thus indicating that tree performance in semiarid environments is largely determined by inter-annual changes in water availability. Furthermore, Wang et al. (2012) report that a prolonged drought can become the dominant climatic factor responsible for increased WUE<sub>i</sub> because it reduces C<sub>i</sub> through a reduction of stomatal conductance. In addition, Ponce Campos et al. (2013) report an intrinsic sensitivity of some plant communities to water availability, and a shared capacity to tolerate low annual precipitation as well as their response to high annual precipitation. These authors suggest that during the drier years, high-productivity sites become water limited to a greater extent, resulting in higher WUE<sub>i</sub> values similar to those encountered in less productive, more arid ecosystems. Our results are similar, when

considering that thinned plot (H98) became more productive after thinning than unthinned (C) plot due to increases in resources availability.

#### **4.2. The relationships between climate and growth after thinning**

Some noteworthy differences suggest that trees in C are more sensitive to water shortages than trees in H98, thus indicating that trees in C plot need to rely more heavily on current year precipitation than those in H98. De Luis et al. (2007, 2011) found that the dynamics of cambial activity in Aleppo pine is characterized by two major growth phases, one in Spring (March-May) and another in Autumn (September-November) interrupted in the Summer period (June-August). However, Moreno-Gutiérrez et al. (2012), indicate that tree-ring growth in Aleppo pine is also influenced by weather conditions during the summer months. In our study, tree growth in the C plot was constrained by water deficit and high temperature stresses during the summer, as suggested by the negative relationship with June-August temperature (previous and current year), and by a positive relationship with precipitation of June-August of current year (Fig. 2 – complementary materials). This result is similar to that found by Andreu et al. (2007) in their study about how climate increases regional tree-growth variability in Iberian pine forests,

and Vicente-Serrano et al. (2010) that showed a progressive increase in the effect of temperature on tree growth during the second half of the 20th century. In contrast, the thinned plot (H98) showed a positive correlation with previous summer temperature and a significant and positive relationship only with May precipitation of current year. Furthermore, this result can be observed also in the Fig. 6 where higher amount of precipitation in June-September fosters growth in the C plot, with its relationship becoming significantly more positive when the competition among trees increased. However, these relationships were lower in H98 as a consequence of less competition, emphasizing the importance of water supply, when the vessels are formed (González and Eckstein 2003). These effects are interesting considering the changes observed in precipitation along the last 15 years (1995-2010) in our study site (Fig. 1), when an increase in May precipitation (15 %) in relation to historic data (1960-2010) was observed. This result confirms our hypothesis that thinning can increase the resilience of trees to climate variations, thus thinning should be considered as an effective means to achieve hydrology-adaptive silviculture. However, when thinning is delayed, as in our experimental plots (H vs H98), the results are less effective due to stagnation during the previous decade (Fig. 2).

### 4.3. Coupling water use with $\delta^{13}\text{C}$ and $\delta^{18}\text{O}$

The impact of drought on tree growth in our study site has been underlined by the high  $\delta^{13}\text{C}$  values observed in those years when dry conditions prevailed along the year (e.g. 1995 and 2005). This is in agreement with other studies in Aleppo pine in Mediterranean areas (Ferrio and Voltas 2005; Sarris et al. 2013). High  $\delta^{13}\text{C}$  values, in turn, can reflect a reduction in stomatal conductance as a consequence of a drought (Eilmann et al. 2010). Both, growth expressed either as BAI or TRW and water-use (WU) in the remaining trees, were strongly higher in H plot than in C plot. However WUEi was not significantly different despite H showing higher WUEi than C. This result might suggest parallel increments of similar magnitude in both A and  $g_s$  values after thinning. Furthermore, there was a positive correlation between growth and water-use across all individuals from the thinned plots (H, M and L together,  $r=0.75$   $p<0.001$ ). Thus, we speculate that a tight stomatal control of tree transpiration and carbon assimilation rate in *P. halepensis* occurs independently of thinning intensity.



$\delta^{13}\text{C}$  and  $\delta^{18}\text{O}$  ratios can be useful to characterize the diversity of water use strategies present in severely water-limited ecosystems (Moreno-Gutiérrez et al. 2011). These authors report that Aleppo pine shows a more “conservative” water use strategy (low  $g_s$ , high  $\text{WUE}_i$ ) during peak growing season when  $A$  increases and that this species is also capable of tapping deeper soil water sources during this period. Hence, considering that thinning can promote a better root distribution in the soil volume and increase water availability, an increase in the stomatal conductance is expected. This increase, in turn promotes an increase in  $\text{WU}$ , but not in  $\text{WUE}_i$  due to increases in the rate of growth, as observed in our study for H, M and L intensities. Since changes in  $\text{WUE}_i$  might be attributed to alterations in  $A$  (Photosynthetic rate) or  $g_s$  (stomata conductance) or a combination of both, the dual-isotope model (Scheidegger et al. 2000) was tested. This approach is attractive in hydrology-oriented silviculture, when considering that changes in  $g_s$  indicate also changes in  $\text{WU}$ . Barnard et al. (2012) report that, if the source of water for trees is the same, then inter-tree variation in  $\delta^{18}\text{O}$  is due to tree physiological processes. Furthermore, a growing number of studies have reported that when plants have the same water source, as in

our case, variation in  $\delta^{18}\text{O}$  is strongly correlated with relative humidity (RH) and evaporative demand, which influences  $g_s$  values. Thus, because RH varied temporally but not spatially among plots in our study, we used the conceptual framework proposed by Scheidegger et al. (2000) to compare measured differences in  $\delta^{13}\text{C}$  and  $\delta^{18}\text{O}$  to the theoretical predictions in  $g_s$  and A, based on the two isotopes measured. Thus, the dual isotope model was tested in each treatment for the years 2009 to 2010 (WU measurement period) to verify if that model can be used to interpret changes in WU observed in those years. We found however that this approach holds only (changes in  $g_s$  equal changes in WU) for H98 and M treatments in our case (Table 4). McDowell et al. (2003) report that, if thinning does result in increased soil water availability, then crown-scale stomatal conductance ( $g_s$ ) should increase because foliage balances atmospheric demand for water vapour with the supply from the soil-plant hydraulic system via  $g_s$ . This increase in turn also increases WU, as observed in our study of H, M and L trees, indicating that competition could be a relevant factor for explaining  $\delta^{13}\text{C}$  and  $\delta^{18}\text{O}$  variations, and their relationships with WU. Furthermore, increases in the positive correlation between  $\delta^{13}\text{C}$  and  $\delta^{18}\text{O}$  were observed after thinning

(data not shown), that can be interpreted as a result of changes in  $g_s$  (Barnard et al., 2012).

Table 4 – Results of dual-isotope model.  $A_{max}$  is average maximum net photosynthesis and  $g_s$  is stomatal conductance. up - or downward arrows represent increasing or decreasing values,  $\approx$  indicates insignificant changes.

Net ppt (mm)*	Treatment	2009→2010		Sheidegger et al. (2000)			This paper's data		
		$\delta^{13}C$	$\delta^{18}O$	Scenario	Theoretical WU (gs)	* $A_{max}$	WU (gs) observed	* $A_{max}$	Scenario
933	Control	↓	↑	d	$\approx$	↓	↑	$\approx$	f
1027	Low	↓	↑	d	$\approx$	↓	↑	$\approx$	f
1145	Medium	↓	$\approx$	e	$\approx$	$\approx$	$\approx$	↓	e
1353	High	↓	↑	d	$\approx$	↓	↓	↓	c
1125	High 98	$\approx$	↑	c	↓	↓	↓	↓	c

\*  $A_{max}$  in both is based on each scenario obtained

Fig. 9 presents a 3D plot with  $\delta^{13}C$ , WU and  $\delta^{18}O$  values in x, y and z-axes. Although we consider 2009 a year with a “dry summer” due to the short period of drought observed (Fig. 2), higher values of WU in

2009 were observed, probably due to high precipitation in September (115.3 mm) compared with 2010 (30.6 mm). This precipitation increases the water availability for autumn that as reported above, is one of the two major growth phases for Aleppo pine (De Luis et al., 2007, 2011), consequently an increase in the tree transpiration (WU) could be expected. However, in 2010 the amount of precipitation in the autumn was mainly in October and November (80% of total), months when the temperature becomes limiting for transpiration (data not shown). These patterns (higher values of WU in 2009 vs 2010) were observed only in H and H98, while no changes could be seen in C, L and M. In other words, H and H98 showed a reduction in WU values despite of the increases in  $\delta^{18}\text{O}$  observed (Table 4). It indicates that the use of  $\delta^{18}\text{O}$  in thinning studies, not necessarily explains observed changes in water-use.

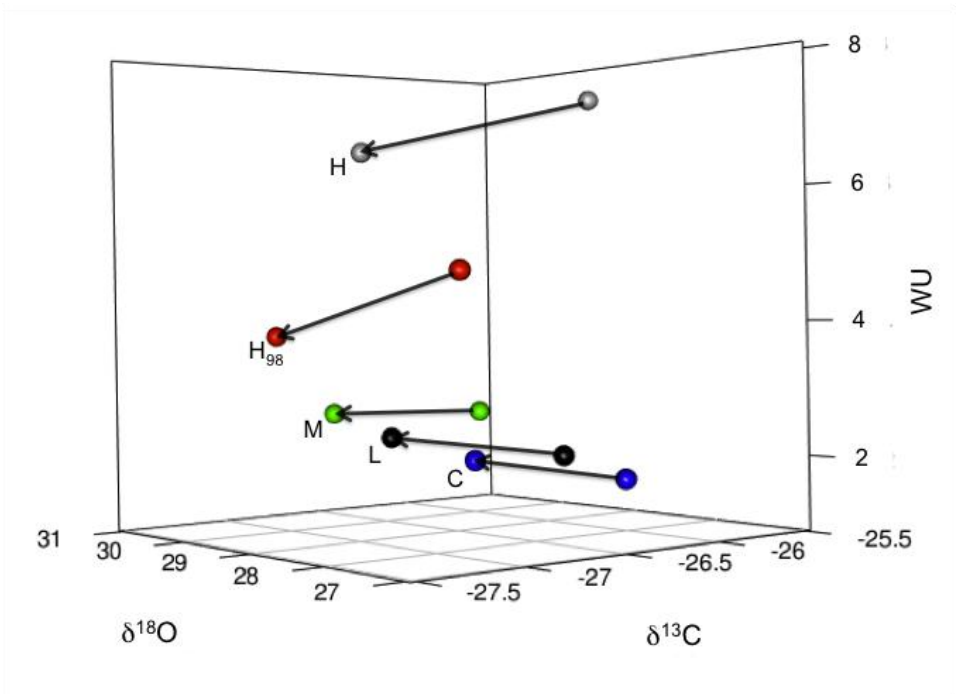


Fig. 9 - A tree-dimensional plot with  $\delta^{18}\text{O}$ ,  $\delta^{13}\text{C}$  in ‰ and Water use in  $\text{m}^3$ . Arrows direction indicates 2009 to 2010 year

## 5. Conclusions

While some of the effects of thinning have been pointed out in other studies, this paper introduces a novel contribution relating water-use (WU) as measured by sap flow sensors, and intrinsic water-use efficiency (WUEi) derived from stable isotope analyses in the same trees in a Mediterranean Aleppo pine plantation subjected to different thinning intensities. Thinning was capable of inducing differences in tree growth

rates. High and Medium intensity thinning were able to induce a high tree-growth that did not occur under Low intensity treatment. The high intensity treatment thinned earlier (H98) showed a higher initial growth rate than the similar treatment thinned in 2008 (H), showing that the delay in the decision to thinning hampers initial growth-rates. In the dendroclimatic analysis, thinning changed the tree-growth-precipitation relationships, showing an increase in positive correlation in spring for H98, while for the control trees this correlation was lower. These results are interesting considering that in the last fifteen years an increase in spring precipitation has been observed in our study site, bringing forth a significant benefit to improve forest resilience to face climate changes. Measurements of stable isotopes in tree-rings can afford powerful information about the physiological and environmental factors that control WU and WUEi. In our study, WUEi at short-term showed non-significant differences between thinned and control plots. Thus, we can conclude that the changes observed in water-use can be explained by changes in stomatal conductance ( $g_s$ ) due to reduced competition after thinning, concomitantly with changes in photosynthesis rate (A). This conclusion can be confirmed by the high growth rates observed after thinning. However at mid-term there are significant differences between

treatments, proving thus that thinning alters the relationships between these two important physiological variables ( $A$  and  $g_s$ ). These relationships have led to the dual isotope ( $\delta^{13}\text{C}$  and  $\delta^{18}\text{O}$ ) conceptual model (Sheidegger et al. 2000) which seems like a useful approach that can be applied to identify changes in  $g_s$  indicating also changes in water use. These findings are attractive in hydrology-oriented silviculture in Mediterranean areas, when considering that changes in  $g_s$  indicate also changes in WU. However our results point to the need of more detailed studies considering the dual model, before making assumptions about changes in water-use in Mediterranean forests, thus the dual-isotope conceptual model must be used with great care in the interpretation of thinning studies, in addition to those reported in Barnard et al. (2012).

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## Complementary materials

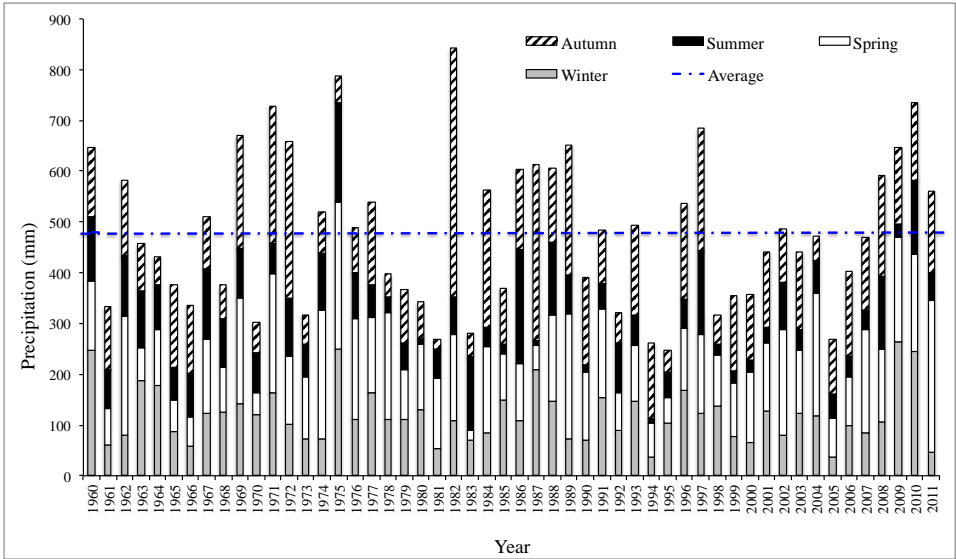


Fig. 1 - Seasonal values of total precipitation during the period 1960 at 2011, following the same approach than for the atmospheric circulation patterns as describe Pasho et al. (2011). Spring (April-May), Summer (June-August), Autumn (September-November) and winter (December-March)

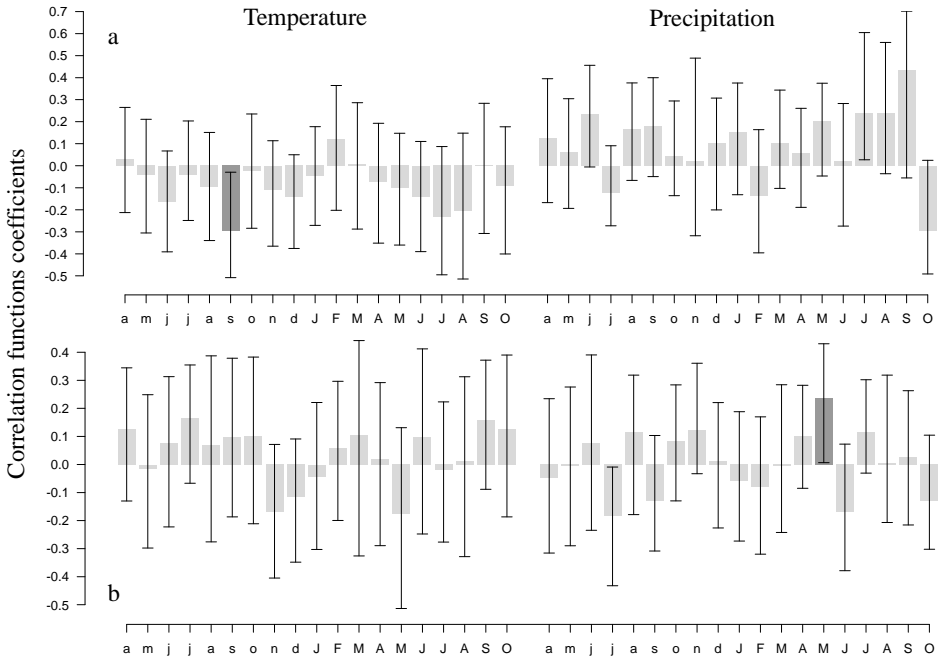


Fig. 2 - Static bootstrapped correlation function for temperature and precipitation from April of previous year to October of current year. a - Control plot, b - Thinned plot (H98). The darker bars indicate a coefficient significant ( $p < 0.05$ ), the lines represent the 95%-confidence interval, and lowercase letters represent the month of previous year (April to December) and uppercase letters month of current year (January to October)

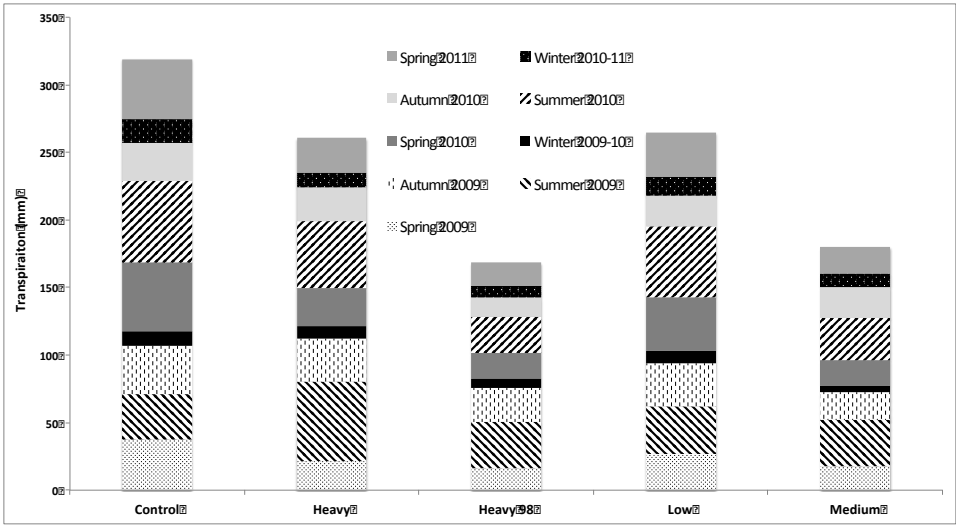


Fig. 3 – Total transpiration (WU) in mm for each treatment per season during the period of study (Adapted from Del Campo et al. 2014). Spring is represented only with April and May months and winter with December at March as describe in Pasho et al., 2011 (see section 2.2.3)

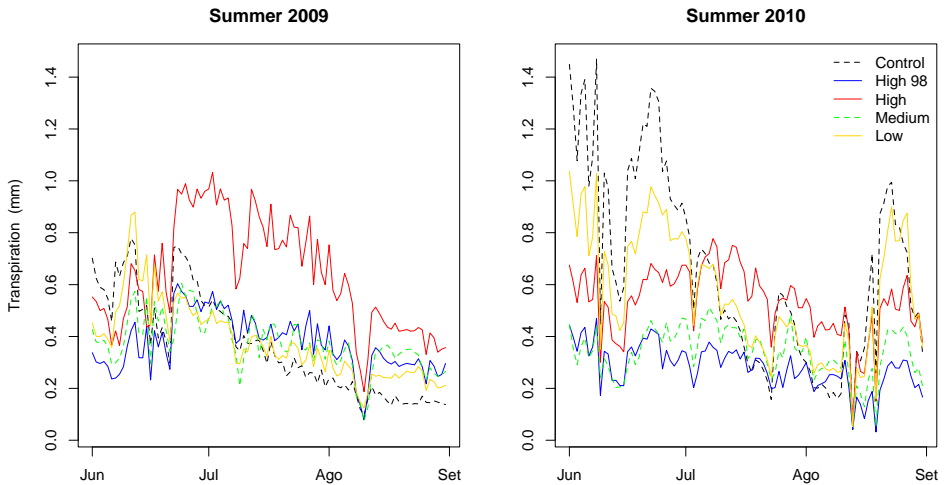


Fig. 4 - Daily water use in mm in the summer period

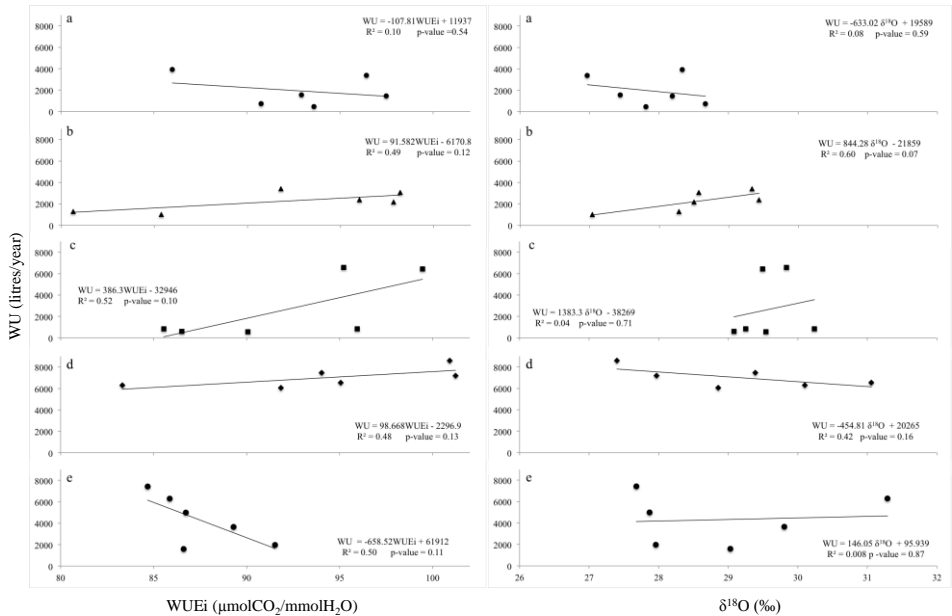


Fig. 5 - right-Relationships between WU (litres/year) and WUEi (μmolCO<sub>2</sub>/mmolH<sub>2</sub>O) and left-Relationships between WU (litres/1000) and δ<sup>18</sup>O (‰) for Control, Low, Medium, High and High 98 plots respectively

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## **CHAPTER FIVE**

General discussion; key findings of this thesis and their implications;  
limitations; some recommendations and future research

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## **General discussion, key findings, limitations and further work**

### **1 – General discussion**

Hydrology-oriented silviculture by thinning might help adapt Mediterranean forest to climate changes. However achieving this goal demands a better understanding and quantification of hydrological balance components. Furthermore, intrinsic water-use efficiency should be considered as an important measurement that might help understand ecophysiological changes in response to thinning. Hence, in this thesis, the effects of hydrology-oriented silviculture on hydrological balance components, on tree growth and intrinsic water-use efficiency were studied in a planted Aleppo pine forest in a semi-arid Mediterranean area subject to different thinning intensities. These effects were studied both at short and mid-term.

The core of this thesis consists of three scientific papers, as reported in "Note to readers - PhD Thesis structure", however a more detailed general discussion beyond the discussion sections of each article, is presented below.

## **1.1 – Use of ANN to estimate forest transpiration with different canopy coverage**

In forest hydrology studies sensor failures are unavoidable and create gaps in the measurements series. A common problem in the data acquisition is power breaks, in particular when the power system is based on solar panels (Papale 2012), as those used here for transpiration measurement. The questions are: 1) Are transpiration data gaps a problem in forest hydrology studies? 2) Should we fill these gaps in the measured data and 3) Which are the methods available to fill missing data? In the consulted literature, these answers are usually unavailable, showing either incomplete transpiration data or filled-in series without information about the method used for gap-filling, with few exceptions e.g. (García-Santos 2012). It is clear that the answers to questions 1 and 2 are affirmative. The presence of gaps is a problem and it is necessary to fill them. In relation to question 3, all the gap-filling methods make use of measured data to reconstruct missing periods. In transpiration data, it is common to use linear regressions using meteorological information, such as vapour pressure deficit (that integrate temperature and humidity), or when gaps are a small temporal window, it is possible to use average

data before and after the gap, or filled-in using valid values on previous or subsequent days at the same time. However, García-Santos (2007) proposes the use of Artificial Neural Network as the best way for gap-filling in transpiration studies. The Artificial Neural Networks (ANNs) are purely empirical, nonlinear regression models with a medium level of implementation difficulties (Papale 2012). This author reports that ANNs, as with all the purely empirical models, can only map and extract information present in the data set used in the parameterization; for this reason the data set must be accurate and as wide and homogenous as possible, to represent the different forest conditions. However, managed forests experience rapid changes in their conditions that drastically modify the water-fluxes and their relationships with meteorological drivers. These changes represent a potential problem to fill gaps through traditional models because even with identical meteorological conditions, there are possibilities of varying responses in water-fluxes.

As presented and discussed in Chapter two of this thesis, ANN proved to be a flexible tool to estimate transpiration considering forest management interventions, with root-mean-square error (RMSE) equal to 0.078 mm/day and correlation coefficient (R) of 0.95. These results are

in agreement to those found in Meijun et al. (2007), Liu et al. (2009) and Li et al. (2009) in poplar, *Pyrus pyrifolia* and *Prunus avium* respectively. Adeloye et al. (2012) and Huo et al. (2012) also found similar results when estimating reference crop evapotranspiration (RMSE = 0.08–0.11 mm/day) using ANN. However, Vrugt et al. (2002) found  $R=0.89$  for transpiration rates of an Austrian Pine stand, furthermore these authors reported that ANN showed high sensitivity to daily values of soil water content. This sensitivity to soil water content also were found in our ANN; however in our case this sensitivity might be better explained by soil water content differences among treatments (Chapter three – Table 2, Fig 5), in addition to daily variations. This is in agreement with Aranda et al. (2012) who report that the sensitivity to soil water availability might explain the different strategies in the sap flow at daily and seasonal temporal scales. In our study we found that ANN predicts accurately the observed data with a subtle dispersion especially in autumn (Chapter two - Fig. 4). In this case they can be partially explained by the variability of weather conditions along the day. However, this daily variability is not considered in the model in our case, where a single mean daily value was used. Despite this, as shown in chapter four (Fig. 1 – Complementary materials), transpiration occurs mainly in summer and spring, thus this

autumn variability does not limit the overall ANN performance. Furthermore the possibility of inserting forest cover as an input in the model extends its applicability in other hydrology-oriented silviculture studies, with some care as indicated below (see Further research and limitations below).

## **1.2 – The hydrological balance after thinning**

In cases where forests are dominated by few tree species, such as many Mediterranean and temperate forests, water relations at the tree level can be more easily scaled up to the stand level, denoting its effect on the local hydrological balance (Klein et al. 2013). As reported in chapter one, forest management practices could trigger positive or negative impacts on the water cycle depending on the context. Furthermore, for a given climatic condition, there are different options for developing sustainable forest management strategies in order to control or minimize the potential hydrological impacts of tree plantations (Aranda et al. 2012). Thus in chapter three of this thesis, the hydrological balance in the study area after thinning was presented and discussed giving a clear idea that thinning might be a good alternative for

implementing hydrology-oriented silviculture. However selecting the best thinning intensity, at present is premature.

The main results showed that canopy interception represents 12.4 to 39.6% of total precipitation (Chapter three – Table 4), showing that thinning increased the total amount of water that reaches the soil; consequently differences in soil water content values were observed (Chapter three – Fig. 5). Hence, different transpiration responses to soil water deficits among treatments were evident when dry conditions occurred (Chapter three – Table 2), although the overall response of sap flow velocity to increasing vapour pressure deficit showed that above a threshold, sap flow velocity remains constant with increasing evaporative demand for all plots (Chapter three – Fig. 2 of electronic supplementary materials). Despite this, it is expected that at long-term, the positive effects of thinning on soil water availability may be offset by an increased water demand of remaining trees by enlarged canopy coverage, as has been observed in the High 98 thinned plot (Figure 1 - below). Similar effects are also reported in other thinning studies (Swank et al. 1989; Bréda et al. 1995; Mazza et al. 2011, Slodicak et al., 2011).

Stand transpiration showed values between 11.7 and 20.7% of total precipitation (Chapter three – Table 4) or 15 to 34.3% of net-precipitation; the higher values representing the control plot, and the lower values corresponding to high intensity thinning. In other words, a decrease in the stand transpiration after thinning was observed. Despite this, increases in transpiration values were observed at tree level, as expected. This increase was mainly due to less competition among trees and the concomitant higher soil water availability. In contrast, Klein et al. (2013) found transpiration to be 50.8 to 95% of total precipitation. Schiller (2011) in Yatir forest found values between 47.3 and 64.3%, while Ungar et al. (2013) report values between 49.1 and 59.4% for the same forest. However, these studies were carried out in areas with significantly lower precipitation than those found in this thesis, i.e. when water is a limiting factor percentage transpiration values tend to be higher. Nonetheless, average transpiration values are in agreement, ca 0.18-0.30 mm/tree/day for similar tree density stands. Using this comparison we can be confident that our measurements of transpiration are representative and thus observed transpiration rates (water-use) is a reasonable measure on which to base our relationships with stable isotopes in tree rings as reported in chapter four of this thesis.

Evaporation loss from soil, litterfall and grass/scrub transpiration was obtained indirectly and varied between 26.3 and 32.9%. These results are in agreement with those of Ungar et al. (2013) that found, in Aleppo pine undergrowth, on average 26.6%. Furthermore, these authors in order to develop a water-balance approach and to estimate maximum sustainable tree-density for lower-rainfall regimes, used values above 39% of evaporation loss and grass/scrub transpiration. Klein et al. (2013) also found similar values (25.7 %) in an Aleppo pine plantations with 1222 tree/ha, corresponding to an intermediate stand between control and low thinning in our study site (Chapter three – Table 1).

The difference between precipitation (P) and evapotranspiration (ET) for a particular area is defined as water yield ( $WY = P - ET$ ) (Klein et al. 2013), considered in chapter three as deep infiltration (see: Water balance for the whole study period in chapter three). Thus, in the study period of this thesis (March 2009 – May 2011) WY were 207; 395.5; 455.8; 647.4 and 499.5mm that represent 13.4%; 25.6%; 29.5%; 41.9% and 32.3% of total precipitation for Control, Low, Medium, High and High98 respectively (Chapter three – Table 4 and Fig. 1 below). These results confirm that reducing forest density increased water yield as



expected, being in agreement with those found in Klein et al. (2013) which considering a "typical" stand density of 300 trees ha<sup>-1</sup>, found 28.6% of water yield. In contrast Yaseef et al. (2010) studying annual values of the hydrological balance components for the Yatir forest (Aleppo pine plantations with 300 trees/ha) in the period 2003-2007, found 7% on average. Schiller, (2011) report 6% for the same forest in the period 2003-2006. Nonetheless, it is clear that the main explanation for this large difference is due to total precipitation observed in our study area, in contrast with that of the latter reported studies.

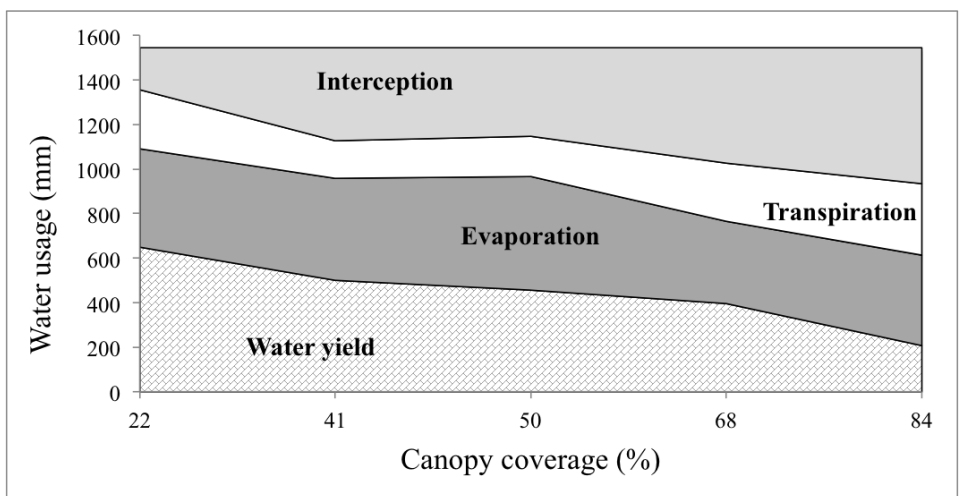


Fig. 1 Schematic representation of changes in the partitioning between hydrological components in response to thinning for a *Pinus halepensis* Mill. forest. 22, 41, 50, 68 and 84% of canopy coverage, refer to High, High98, Medium, Low and Control plot respectively.

In summary, these large differences in the water balance components between thinning plots and control (Fig 1 above) could have significant implications at larger-scale hydrological balances, with further implications for water yield, water availability to maintain forest resilience, and appropriate land use management.

### **1.3 – Relationship between tree-growth and climate after thinning**

In this thesis significant differences in growth among experimental treatments were found, while we found no significant differences before thinning (Chapter four – Table 2). The observation that growth increased after thinning is not unique; in fact, foresters throughout the world rely on thinning-induced growth response for increasing stem-wood yield (McDowell et al. 2003). In the chapter three of this thesis, thinning showed improved growth (Basal area increment - BAI) in all plots, although in the low-intensity thinning, the value is still low. On the contrary, the high intensity thinning treatments revealed the potential growth capacity. Growth by tree-ring chronologies reflects the complex interaction of climatic and environmental conditions (Fritts 1976). However, changes in the environmental conditions that trees experience does not only originate from climatic changes, but also from

changes in forest stand structure due to forest management practices (Churakova (Sidorova) et al., 2014), such as thinning. Thus, in chapter four, the relationships between climate and tree-growth were studied to identify thinning responses to observed climate variation (annual and seasonal). Vicente-Serrano et al. (2010) report that, in forests located in the most arid areas, the positive trend observed in the potential evapotranspiration rates, increased water stress which had a negative effect on forest growth. Thus, changes in precipitation may not be the main factor that drives changes in forests growth; the interplay with temperature may also have a significant role. It is well known that increased temperature usually has a positive effect on the activity and growth of trees, nevertheless the effect can be negative if there is no corresponding increase in precipitation, thus water stress may be enhanced. In the last fifteen years this pattern was observed in the study area, i.e. increase in temperature and decrease in precipitation (Chapter four – Fig. 1). In chapter four of this thesis (Fig 6 and Fig 2 - Complementary materials), tree growth responses analysis to temperature and precipitation were presented. Contrasting the thinned plot (H98) with the control plot, different correlation values were observed. In other words, thinning was able to change the relationship between tree-growth

and climate (temperature and/or precipitation). Furthermore, the prolonged drought observed in the periods 1999-2001 and 2004-2005 (Chapter four – Fig. 5) clearly showed a negative effect on tree-growth (Chapter three – Fig. 2). But despite the general impacts of drought, their effects are not homogeneous between thinned (only H98 in this case) and control plots (Chapter four – Fig. 2b). Such a result points out to particularities of the hydrological stress that should be confirmed and developed at a local-scale in hydrology-oriented studies. Hence, the relationships between climate and growth found in this thesis corroborate the idea that thinning is a useful tool to maintain or improve forest resilience to face the observed climate change. However, we are not yet able to make an accurate inference about what would be the best thinning intensity to maintain forest resilience in response to climate change observed or expected in modelled scenarios.

#### **1.4 – Water-use and water-use efficiency after thinning**

Water-use and water-use efficiency are crucial to further the understanding of hydrological forest processes. Their importance increases when new forest responses occur due to silvicultural practices, such as thinning. As reported along this thesis, thinning practices may

represent a proactive forest management method (Magruder et al. 2013) that involves increases in the availability of light, water and nutrients. As a consequence individual trees could make a more efficient use of these resources (Forrester 2013). However, Gravatt et al. (1997), suggest that improvements in tree-water-relations may not always occur after thinning or may be ephemeral depending upon thinning intensity. In this thesis (Chapters three and four), the evidence of thinned plots differences in growth,  $\delta^{18}\text{O}$ , WU and WUEi were detected in different ways. These results indicated that the ecophysiological mechanisms of adaption to thinning among the studied plots differed. Despite of the high number of studies that report growth,  $\delta^{18}\text{O}$ , WU and WUEi in Aleppo pine plantations in Mediterranean areas (see: previous chapters) no studies have been found on the impact of thinning in these attributes taken together, that would provide data for comparison.

Intrinsic WUE, i.e. the ratio between photosynthesis and stomatal conductance, is readily calculated from leaf gas exchange data. Yet under field conditions, large fluctuations in these fluxes mean that point measurements can only provide transient values. On the other hand, tree-ring stable carbon isotope composition  $\delta^{13}\text{C}$ , provides a time-

integrated indicator of WUE<sub>i</sub>, reflecting the ratio of intercellular to atmospheric CO<sub>2</sub> concentrations during the period in which the carbon was fixed (Klein et al. 2013). Thus, dendrochronology studies and stable isotopes analysis might be successfully used to infer changes on photosynthesis and stomatal conductance after thinning practices, as presented in chapter four of this thesis. *Pinus halepensis* Mill. (Aleppo pine) is considered an isohydric<sup>13</sup> species, due to its capacity to avoid drought by reducing stomatal conductance to a minimum (Klein et al. 2011, Moreno-Gutiérrez et al. 2012; Klein et al. 2013; Battipaglia et al. 2014), which in turn affects carbon uptake, with indirect impact on plant metabolism. Sarris et al. (2013) report that Mediterranean pines, reduce stomatal conductance in dry years, which in turn increases  $\delta^{13}\text{C}$  and decreases  $\delta^{18}\text{O}$  values in tree-rings compared to more humid years. On the contrary, Martin-Benito et al. (2010) suggest that increases in  $\delta^{18}\text{O}$  might be due to environmental changes after thinning and that this increase might not be related to changes in stomatal conductance. However, in this thesis, despite the droughts observed in years where SPI

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<sup>13</sup> Iso-/aniso-hydric water use strategy is a conceptual model describing species that moderate internal water potential in dry soil and atmosphere by stomatal closure (isohydric) versus those that maintain open stomata as far as possible under dry soil and air at the cost of more negative internal water potential (anisohydric) (Kjellgren, 2010).

values were lower than zero (Chapter four – Fig. 5), H98 plot immediately after thinning showed high basal area increment (Chapter three – Fig. 2) and  $\delta^{18}\text{O}$  (Chapter four – Fig. 4a), reflecting that drought effects, in this case, did not impact negatively plant metabolism compared with control plot. Furthermore, increases in WU, which can be related to stomatal conductance ( $g_s$ ) after thinning was observed for all thinned plots at short-term. However, no significant differences in  $\text{WUE}_i$  between Control and thinned plots were observed. Thus, we can conclude that changes in WU caused by changes in  $g_s$  due to thinning, should scale positively with concomitant changes in photosynthesis rate (A) ( $\text{WUE}=\text{A}/g_s$ ). This is similar to the results reported by Hu et al. (2010) who found that differences in A should reflect differences in transpiration, and therefore differences in  $g_s$ .

These findings have several implications, as for example the confirmation that Aleppo pine in the study area coped well with drought years, and that quantifying the separate sensitivities of A and  $g_s$  to environmental influences after thinning, can greatly enhance our ability to identify major limitations to photosynthesis and growth. Furthermore, this knowledge can greatly improve the usefulness of the analysis of tree

ring as a tool to guide forest management decisions to face climate changes by means of improvement of forest resilience. Thus, an important result of this thesis was achieving a better understanding of which hydrology-oriented silviculture as an adaptive management could produce a best and feasible resilience level under water scarcity scenarios. However, to understand the dynamic interactions between water-use, water-use efficiency and forest management practices more detailed studies are needed.



## **2 - Key findings and conclusions of this thesis**

Hydrology-oriented silviculture pursues the aim of establishing more ecological, stable and site-adapted forests. Thus, its main goal is to promote forest resilience and at the same time optimize the forest hydrological balance. This target can be reached by thinning, which can increase water availability for remaining trees and hence water yield. Furthermore it can promote higher water use efficiency. These issues are the main considerations of this thesis.

A few important advances have been made throughout this thesis, they are:

- The sap flow methodology to measure tree water uptake, has made possible the study of the effects of thinning on transpiration at both levels, tree and forest stand with a high temporal resolution. However the presence of data gaps frequently produces inaccurate estimates of the total water transpired by trees. In order to circumvent this problem, an artificial neural network was developed and successfully applied in this thesis.
- The results of tree water-use have shown that differences in transpiration among treatments varied depending on the

considered up-scaling stages, i.e. sap flow velocity, tree transpiration or stand transpiration. For example, when the number of trees is taken into account, the unmanaged control plot is the most water-spending stand, however the remaining trees in High intensity thinning showed higher transpiration rates than control trees;

- The transpiration percentage of total precipitation between Low and High and between Medium and High 98 plots showed subtle differences (17.1% - 16.9% and 11.7 – 10.9% respectively), however the differences in growth rate were strongly significant;
- In this thesis, it was found that differences either in forest density or cover play an important role in the main hydrologic balance components; thus, thinning is expected to affect not only interception, but also transpiration and evaporation from soil. Subtle differences between control and Low thinned plot in all water balance components, reinforce the idea that a slight intervention does not impact water balance at short-term. Hence, the water balance obtained on chapter three corroborated the idea that thinning is a useful tool to improve water balance in water

scarcity-prone areas to face expected climate changes;

- Thinning was also capable of inducing differences in tree-growth, where High and Medium intensity thinning were able to induce higher tree-growth rates. The remaining trees in High intensity treatment thinned earlier (H98) showed a higher initial growth rate than the similar treatment thinned in 2008 (H), showing that the delay in the decision to thinning hampers the initial boost in growth-rates;
- Dendroclimatic analysis showed that thinning changed the tree-growth-precipitation relationships, showing an increase in positive correlation in spring (season in which an increase in precipitation was observed in the last 15 years) for H98, while for the control trees this correlation was lower, bringing forth a significant benefit to improve forest resilience to face climate changes;
- The combination of traditional hydrology field-studies together with dendrochronology and stable isotopes approaches provides a useful tool to understand water-use (WU) and intrinsic water-use efficiency (WUEi) changes after thinning;

- The impact of thinning on WU and WUEi showed an inherent relationship with precipitation. Despite this, different relationships (slopes) between WUEi and net precipitation for each thinning intensity were found, which might be attributed to a decrease in competition among trees, rather than to an absolute increase in water availability. Hence, thinning should be considered as a useful tool to mitigate the possible impacts of total precipitation decrease;
- In conclusion, the general results obtained of this thesis highlight the relevance of the underlining hypothesis that unmanaged forests are more water consuming than managed forests. Therefore, future forest policies in Mediterranean arid regions should pay more attention to the effect of thinning on forest water balance, especially in light of future climate changes.

### 3 - Further research and limitations

Since the core of this thesis consists of three interconnected scientific papers (Fig.1 – Outline of this thesis and their objectives), specific limitations encountered (*indicated in italic and underlined*) and future research lines are condensed here.

- This thesis contributes to improve the comprehension of water-oriented management in this study area. Nonetheless, forest responses to management practices are dependent on many factors, as soil properties, climate, species, among others. Thus, it is necessary to take great care when *making extrapolations with these results outside of this experimental area;*
- *Transpiration data collected in this thesis refer only to two years, both considered as "wet years" (total precipitation 35.6% and 54.3% higher than historical data 1960-2010).* Thus, it is necessary to collect more field information and re-train artificial neural network (ANN) in order to check if the ANN can be successfully used also in dry years;
- *The absence of drought-years after thinning did not provide the opportunity to make inferences on tree responses to drought at*

different stand densities. Maybe the re-installation of all sensors and other field equipment might be recommended;

- In this study the transpiration values are comparable to data in other published studies. However, it seems advisable to give more consideration to: sap flow sensor calibration, base-line identification, total sapwood area assessment, active sapwood area and azimuthal differences in sap-velocity. In other words the plant transpiration routine by sap flow sensors need more detailed studies, also more sensors per tree and per plot;
- Total water-use, i.e. stand transpiration, in thinned plots was lower than in the control plot. These measurements do not represent a change after thinning in the traditional sense, because tree transpiration was not measured prior to thinning, but the magnitude of change from 2009 to 2010 in thinned plots compared to control plot, suggests a physiological response to thinning. In this sense it is important to consider in new studies the possibility of measuring all plots before thinning in order to identify the real physiological change on each tree studied.

- The residual evaporation term (evaporation from soil, litterfall and grass/scrub transpiration – Chapter three – Table 4), that represent in average more than 28% of water balance, should be experimentally addressed in order to have a better estimate of errors associated with the overall water balance;
- *Deep water movements was not measured* in this thesis, however the installation of piezometers might be recommended to address its movements and improve information of soil water flux;
- A more detailed analysis on changes in soil properties produced by thinning could be usefully to explain observed soil water content differences among plots in our study. Furthermore the assumption that residual roots after thinning (roots of cut trees) contribute to soil water availability differences is a open question in thinning studies;
- *The elevated cost of stable isotope analyses* represents a limitations to explore in more detail the nuances of the relationships between WU by sap flow sensors and WUE<sub>i</sub> by

dendrochronological approach in thinning studies, face to face the Scheidegger dual isotopic model.



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