





Ecohydrological relations in a wild cherry tree plantation for timber production

PhD Thesis

Antonio J. Molina Herrera

to be eligible for the Doctor degree

Supervised by:

Dr. Carme Biel Loscos

Dr. Pilar Llorens García

Tutor:

Dr. Jordi Martínez Vilalta

Doctorate in Terrestrial Ecology

Universidad Autònoma de Barcelona, November 2015







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Resum

Els diferents treballs presentats en aquesta tesi han permès aprofundir en diversos aspectes relacionats amb el maneig de plantacions de cirerer per a fusta de qualitat en un context mediterrani, així com en uns altres més purament relacionats amb processos hidrològics, fisiològics o ecològics i amb la metodologia utilitzada per la mesura d'aquests.

En el primer capítol es van testar els efectes que poden tenir sobre la producció de fusta el tipus de sòl (argilenc amb escàs percentatge d'elements gruixuts, enfront de sorrenc amb elevat percentatge d'elements gruixuts), el reg per degoteig (reg enfront de no reg) i el llaurat del sòl (llaurat mecànic enfront de vegetació espontània). Els resultats van mostrar que tant el tipus de sòl com el reg van tenir efectes significatius, mentre que el llaurat de sòl no va suposar increments notables en el increment en el volum de fusta. L'extrapolació dels resultats a tot el torn de plantació va permetre establir que, si la tendència observada es manté, el torn de plantació pot ser massa elevat per al sòl de tipus sorrenc, mentre que s'aconseguiria una reducció del voltant de deu anys al aplicar el reg, independentment del tipus de sòl. D'altra banda, la valoració econòmica i ecològica dels resultats del llaurat del sòl va permetre establir una forta reducció dels costos associats i una alta diversitat de la vegetació herbàcia acompanyant, amb els beneficis associats que aquesta última podria portar amb si.

En el següent capítol es va estudiar la partició de la pluja amb un disseny experimental d'alta resolució espacial i temporal (que va permetre estudiar els processos a diferents escales), i amb un seguiment de l'estructura forestal i el clima com a possibles factors explicatius. Mentre que certes pluges d'escassa magnitud van provocar elevades intercepcions de pluja, els resultats a escala global (acumulat per a tot el període d'estudi) van indicar una baixa capacitat d'intercepció de la plantació, amb el que això pot suposar en matèria de reg. Des del punt de vista metodològic, es va posar de manifest que deu pluviòmetres per mesurar la trascolació són suficients per recollir la variabilitat espacial d'aquesta variable a l'escala global.

En el tercer i quart capítol així com en l'annex, es va avaluar la transpiració i la seva relació amb variables ambientals (evapotranspiraciò i contingut hídric del perfil de sòl) i estructurals de la vegetació, d'una banda, i diversos aspectes metodològics relacionats amb l'us de sensors de flux de saba per estimar la transpiració, per una altra. Com a resultats més importants, es va posar de manifest el control estomàtic de la transpiració per a rangs concrets de contingut hídric del sòl, així com de la utilitat de les podes per millorar l'eficiència en l'ús de l'aigua a escala de plantació (capítol 3). En relació als aspectes metodològics, es va confirmar que alguns mètodes àmpliament utilitzats en altres espècies per estimar l'àrea conductora no van ser vàlids per a l'espècie estudiada. D'altra banda, es van establir els perfils radials i azimutals de la densitat de flux de saba (capítol 4) i es va comprovar que no considerar la variació que l'àrea conductora té al llarg del període vegetatiu pot suposar importants errors en l'estimació de la transpiració (annex).

Finalment, en el capítol cinc, gràcies a l'us d'isòtops estables de l'aigua, es va arribar a la conclusió que, encara que les arrels fines dels arbres es van presentar al llarg del tot el perfil de sòl (0-100 cm), l'extracció principal d'aigua en l'època vegetativa es va produir fonamentalment de les capes superficials, unes vegades a 25 cm de profunditat i unes

altres a menor profunditat. Aquest patró d'extracció d'aigua, juntament amb el similar observat per a la vegetació herbàcia acompanyant, va semblar indicar una competència per l'ús de l'aigua entre els arbres i les herbàcies. D'altra banda, els resultats en el sòl de pitjor qualitat agronòmica (el de tipus sorrenc indicat en el primer paràgraf d'aquest resum), amb taxes transpiratives molt baixes en les condicions de no reg, van indicar que la tècnica isotòpica podria ser no adequada en aquestes condicions, on la mescla de les aigües del xilema i floema invalidaria l'assumpció inicial de la relació directa entre l'aigua de xilema i l'aigua del sòl.

Resumen

Los diferentes trabajos presentados en esta tesis han permitido profundizar en diversos aspectos relacionados con el manejo de plantaciones de cerezo para madera de calidad en un contexto mediterráneo, así como en otros más puramente relacionados con procesos hidrológicos, fisiológicos ó ecológicos y con la metodología utilizada para el monitoreo de éstos.

En el primer capítulo se testaron los efectos que pueden tener sobre la producción de madera el tipo de suelo (arcilloso con escaso porcentaje de elementos gruesos frente a arenoso con porcentaje elevado de elementos gruesos), el riego por goteo (riego frente a no riego) y el laboreo del suelo (laboreo mecánico frente a vegetación espontánea). Los resultados mostraron que tanto el tipo de suelo como el riego tuvieron efectos significativos, mientras que el laboreo no supuso incrementos notables en el volumen maderable. La extrapolación de los resultados a todo el turno de plantación permitió establecer que, si la tendencia observada se mantiene, el turno de plantación puede ser demasiado elevado para el suelo de tipo arenoso con porcentaje elevado de elementos gruesos, mientras que se lograría una reducción de alrededor de diez años al aplicar el riego, independientemente del tipo de suelo. Por otra parte, la valoración económica y ecológica de los resultados del laboreo del suelo permitió establecer una fuerte reducción de los costes asociados y una alta diversidad de la vegetación herbácea acompañante, con los beneficios asociados que esta última puede traer consigo.

En el segundo capítulo se estudió la partición de la lluvia con un diseño experimental de alta resolución espacial y temporal, además de un seguimiento de la estructura forestal y las variables meteorológicas como posibles factores explicativos de la variabilidad espacio-temporal. Mientras que ciertas lluvias de escasa magnitud provocaron elevadas pérdidas por interceptación, los resultados para todo el periodo de estudio indicaron una baja capacidad de interceptación de la plantación, con lo que ello puede implicar en materia de riego. Desde el punto de vista metodológico, se puso de manifiesto que diez pluviómetros son suficientes para recoger la variabilidad espacial de la trascolación a escala de parcela.

En el tercer y cuarto capítulos, así como en el anejo, se evaluó la transpiración y su relación con las variables ambientales (evapotranspiración y contenido hídrico del perfil de suelo) y estructurales de la vegetación. Así mismo, se analizaron diversos aspectos metodológicos relacionados con el empleo de sensores de flujo de savia para estimar la transpiración. Como resultados más destacables se puso de manifiesto el control estomático de la transpiración para rangos concretos de contenido hídrico del suelo, así como de la utilidad de las podas para mejorar la eficiencia en el uso del agua a escala de plantación (capítulo 3). En lo que se refiere a los aspectos metodológicos, se confirmó que algunos métodos ampliamente utilizados en otras especies para estimar el área conductora, no fueron válidos para la especie estudiada. Por otro lado, se establecieron los perfiles radiales y azimutales de la densidad de flujo de savia (capítulo 4) y se comprobó que no considerar la variación a lo largo del período vegetativo del área conductora puede suponer importantes errores en la estimación de la transpiración (anejo).

Resumen

Por último, en el capítulo cinco, gracias al empleo de isótopos estables del agua, se llegó a la conclusión de que, aunque las raíces finas de los árboles se localizaron a lo largo del todo el perfil de suelo (0-100 cm), la extracción de agua durante el período vegetativo se produjo principalmente de las capas más superficiales, unas veces a 25 cm de profundidad y otras a menor profundidad. Este patrón de extracción de agua, junto con un patrón similar observado para la vegetación herbácea acompañante, pareció indicar una competencia por el uso del agua entre los árboles y las herbáceas. Finalmente, las tasas transpirativas tan bajas obtenidas en condiciones de no riego en los árboles situados en el suelo de peor calidad agronómica (el de tipo arenoso indicado en el primer párrafo de este resumen), indicaron que la técnica isotópica podría ser no adecuada en dichas condiciones, ya que una posible mezcla de las aguas del xilema y floema, en estas condiciones de estrés severo, invalidaría la asunción inicial del método de una relación directa entre el agua del xilema y el agua del suelo.

Summary

All different works presented in this thesis have allowed a further study of some of the main aspects related to the management of wild cherry tree plantations for timber production under Mediterranean climatic conditions. Moreover, key hydrological, physiological and ecological processes have been monitored and the methodology used for that purpose tested.

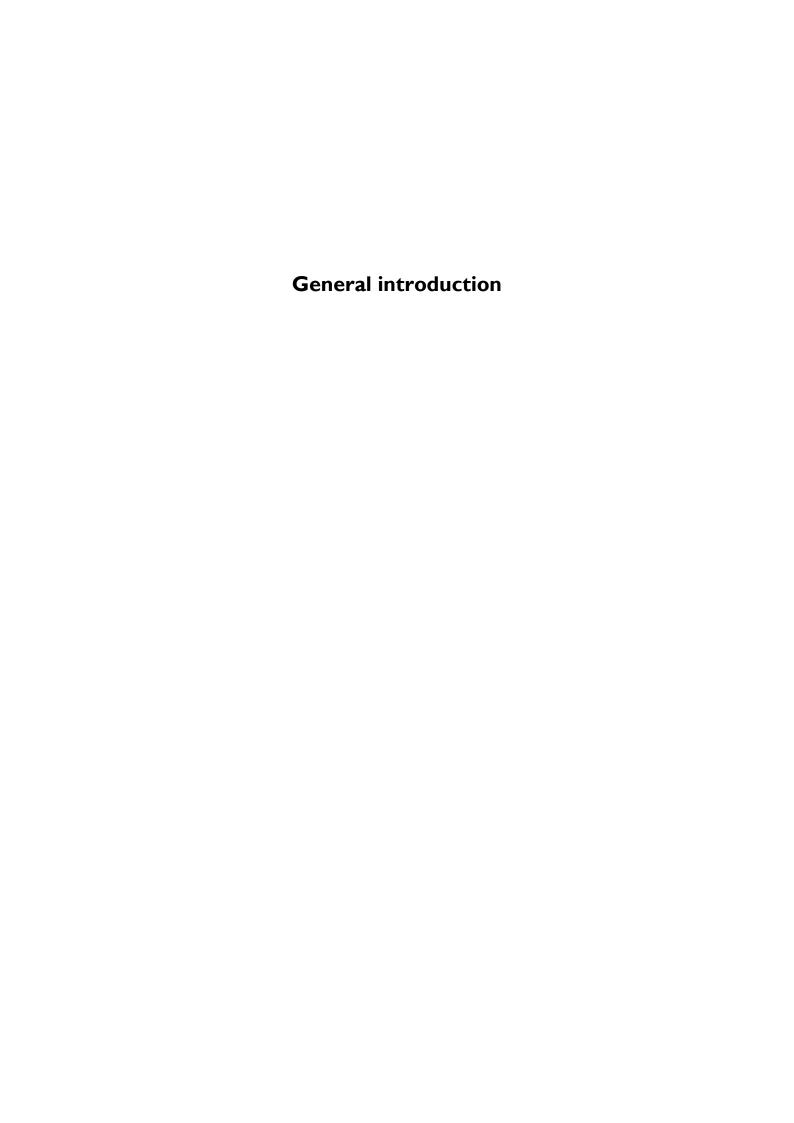
Within the first chapter, the potential effects of soil type (clay-loam texture with negligible presence of gravels versus sandy-loam texture with high proportion of gravels), irrigation (drip irrigation versus rainfed conditions) and soil tillage (soil tillage versus spontaneous herbaceous plants leaved to grow) on timber volume were tested. Results showed that both soil type and irrigation had significant effects on the timber volume increase, whereas soil tillage did not. Extrapolation of these results to the future allowed setting that, if the observed tendency is maintained, the rotation length will be too high for the sandy soil, whereas a reduction of ten years in the rotation length will be obtained if irrigation is applied, regardless the soil type. On the other hand, the economic and ecological assessment of soil tillage results allowed establishing a strong reduction of the associated costs and high herbaceous vegetation diversity, with the potential related benefits the latter could imply.

In the following chapter rainfall partitioning was studied by using an experimental design of high spatial and temporal resolution, together with the monitoring of forest structure and climate variables as potential driving factors. While certain rainfall events of low magnitude caused high interception losses, results at global scale (the accumulated value for the whole study period) indicated a low rainfall interception capacity for the plantation. From a methodological point of view, it was showed that ten tipping buckets were sufficient to collect the spatial variability of throughfall at the global scale.

Within chapters three and four and in the annex, stand transpiration and its dependency with environmental (evapotranspiration and soil water content) and forest structural variables were assessed in one hand, and in the other, certain methodological aspects related to the use of sap flow sensors were evaluated. As for the most relevant results, the linear relationship between transpiration and soil water content, and the ability of branch pruning to improve the water use efficiency at stand scale, are highlighted (chapter 3). With regards to methodological aspects, it was confirmed that some methods widely used in other species to estimate the sapwood area were not suitable for the specie under study. In addition, radial and azimuthal profiles of the sap flux density were defined (chapter 4), and it was confirmed that the non consideration of the variation of the sapwood area along the vegetative period may cause relevant errors in the transpiration estimates (annex).

Finally, in chapter five, thanks to the use of stable water isotopes, it could be concluded that, even though fine tree roots were observed all through the soil profile (0-100 cm), the root water extraction during the vegetative period was mainly obtained from soil superficial layers, sometimes from 25 cm depth, and some other times from more superficial depths. This water uptake pattern for wild cherry tree, together with that similar one observed in the spontaneous vegetation, is likely to be an indication of water use competence between trees and herbaceous plants. On the

other hand, results from the lowest agronomic quality soil (that of sandy texture indicated in the first paragraph of this summary), with very low tree transpiration rates under rainfed conditions, seemed to indicate that water isotopes technique can be inadequate for such conditions, where the mixture of xylem and phloem waters may invalidate the assumption of a direct linkage between xylem and soil waters.



General introduction

General introduction

In the last decades there has been a growing interest in both noble wood plantations and bush plantations for food and medicine uses since they can become alternative options to the traditional Mediterranean crops. In 2011, the Ecophysiology group of the Institut de Recerca i Tecnologia Agroalimentaries (IRTA) led a national project with other partners involved (another group of IRTA and other groups from IDAEA-CSIC, UPC and UdL), named "Ecohydrological relations in agroforestry interfaces, what is the role in the regulation of water and carbon?". The project was funded by the Ministry of Economy and Competitiveness in the years 2011-2013 (AGL 2010-21012), together with the FPI scholarship grant for which the author of this document was the beneficiary. The project was primarily aimed at evaluating different hydrological and physiological variables related to different management strategies in the abovementioned plantations under Mediterranean climatic conditions. Within this context, this thesis has focused on one of the studied plantations, i.e. the wild cherry tree plantation located in the facilities of IRTA Torre Marimon in Caldes de Montbui (Barcelona).

Ecophisiology of wild cherry tree

Wild cherry (*Prunus avium* L.) is a tree belonging to the Rosacea family which may reach heights of 20-30 m. This tree has normally a straight trunk thanks to its pronounced apical dominance, which is maintained until maturity is reached (Cisneros, 2004). This aspect gives him a high interest for wood production purposes, in addition to its highly valued properties in the world of carpentry. This hierarchy of the apical bud also occurs in secondary branches, so that the tree structure consists of number of repetitions or fractals on their different floors. Its flowering usually starts after at the end of winter cold and its root system usually consists of multiple roots which normally decrease with soil depth.

Its origin comes from Caucasian and Balkan regions, and its natural presence in Europe is widely spread (natural distribution of the species is showed in Figure I), increased by its significant expansion as crops for the production of fruits by the Roman Empire. In our country, it is mainly concentrated in areas of continental climate and also transition zones with oceanic climate, with an average annual temperature ranging

between 4-12 °C, average minimum temperature of the coldest month ranging between 6.4 to 0 °C, average annual rainfall ranging from 750 to 1550 mm and from 110 to 375 mm in the summer period (Guardiola, 2014). In contrast, its natural presence in Mediterranean regions is incidental, found only in areas close to rivers and shaded places (Cisneros, 2004; Cisneros et al., 2012).

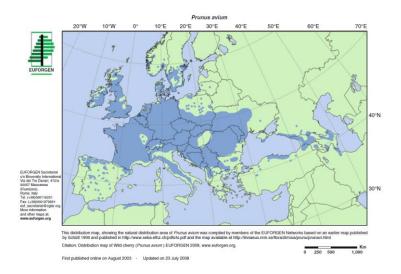


Figure 1. Natural range of Prunus avium L. (source EUFORGEN 2009 www.euforgen.org)

It is very sensitive to soil waterlogging, as its root system easily tends to become rotten (Cisneros et al., 2012). For this reason, wild cherry trees prefer permeable soils with good aeration. Also, it grows under optimal conditions in soils with high capacity for water retention, but may live in areas with limited soil water retention capacity if additional water during the vegetative period is supplied (Montero et al., 2003).

Noble timber plantations in a Mediterranean context

The Mediterranean basin consists of a great variety of landscapes (forests, shrublands, agricultural areas, urban areas...) of different sizes and uses (Forman, 1995; Forman and Godron, 1986), since it is the result from the interaction between man and nature during thousands of years. Nowadays, is the most vulnerable region of Europe, given its fragility against climate change (Schröter et al., 2005) and the dramatic change in land use promoted by the abandonment of agriculture. The later was encouraged by the expectations of better life in the city by rural people, since rural areas were suffering a lack of economic stimulus from the European Common Agricultural Policy (Shucksmith et al., 2005). In Spain, for instance, agricultural land decreased by 2.3

million hectares between 1985 and 2009 (data from MARM, 2009), bringing an increase of naturally regenerated forests.

Land-use change may have important implications on hydrology and other environmental aspects such as erosion and fire risks, diversity loss, etc. In our country, while these implications have been widely studied in forested areas since they are the main source of freshwater downstream (e.g. Gallart et al., 2011, López-Moreno et al., 2008; Pausas, 2004), in marginal agricultural areas of low productivity these environmental implications have not been carefully evaluated (Anglès, 2009; Lasanta-Martínez et al., 2005). As a consequence of the Reforestation Program for Agricultural Land, derived from the transposition of the EU Directive "EEC Regulation 2080/92", which main objectives were to avoid erosion and to increase CO₂ capture, since 1993 700.000 ha of plantations have been set up in Spain in zones proceeding from abandoned crops. The majority of them in the regions of Castilla-León, Andalucía, Castilla-La Mancha, Extremadura, Galicia and País Vasco (MAPA, 2006).

Thanks to the economic incentives derived from these political programs, forest and/or agroforestry plantations with noble or high quality timber species, have become more relevant in Europe, being France, Italy and Spain where the highest increments have been registered for the last thirty years (Aletà and Vilanova, 2006; Aletà and Vilanova, 2014; Ducci et al., 2013). In our country, even though a lack of information exists due to the fact that these species are not exhaustively accounted for in the agricultural inventories, walnut and wild cherry tree plantations are currently the ones occupying the greatest area. It can be said that walnut tree plantations for forestry purposes occupy an area of approximately 5000 hectares, while wild cherry plantations occupy around 3000 hectares (Aletà and Vilanova, 2014).

The price of noble wood has being maintained stable in the last years with a maximum of 1000 €/m³ (Loewe et al., 2013). This means that plantations for the obtaining of high quality timber may imply additional incomes for farmers and therefore an alternative ways to diversify their production and the related risks. In addition, the ecological functions of these plantations dealing with CO₂ capture and biodiversity are already documented (Ducci et al., 2013). However, these may not be reached in areas with water restriction like the Mediterranean (Gracia et al., 2010; Jackson et al., 2009), since species within noble plantations usually present high water requirements, such as

wild cherry tree. In this respect, the increase in greenhouse gases in the last decades is expected to enhance air temperature to at least I °C in the Mediterranean regions of our country (IPCC, 2014). With regards to rainfall, the amount is not expected to change but its distribution along the year is expected to. Therefore, this may involve the combination of higher evapotranspiration and reduced water availability, thus negatively affecting plant growth and development during the vegetative period.

Under this complex context in the Mediterranean basin, it seems necessary a multidisciplinary approach for addressing any matters dealing with either vegetation, water or with the proper management of both to improve the resilience of the natural systems. In this regard, Ecohydrology (Rodriguez-Iturbe, 2000), a new discipline which focus on plant-water-atmosphere continuum as the central working topic, has seen its popularity increasing in the last ten years. That is the reason for setting both the title and the structure (next section) of this thesis.

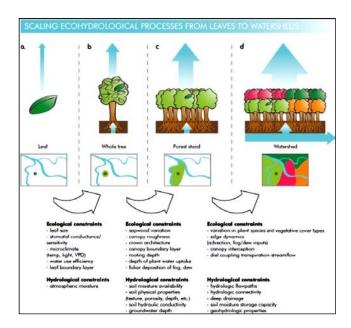


Figure 2. Conceptual model for scaling ecohydrological processes from leaves to whole trees to forest stands to watersheds, with emphasis on the ecological and hydrological controls on water fluxes that must be understood and quantified when transferring ecohydrologic information across scales. Taken from Asbjornsen et al. (2011).

This multidisciplinary approach is shown in Figure 2, where the variables and parameters that should be taken into account for a proper scale change are indicated, from leaf to watershed (Asbjornsen et al., 2011). Noble plantations are also suitable to be studied under this multidisciplinary approach. Apart from the necessary studies which focus on plant breeding programs to select the best quality material (e.g. Nocceti et al., 2010), there is also the need for others assessing the plantation role in

the local water cycle and also how these plantations may be physiologically affected when alternative agronomic practices are considered. This acquires special importance in the Mediterranean regions of our country, in which very limiting conditions may exist for these plantations, for which complementary information of this kind (such as that presented in Guàrdia, 2013) is of crucial relevance for their correct development. Evidence can be found from several wild cherry tree plantations that suffer from deficient development due to either inadequate selection of the location, inadequate management practices or both (Vilanova, personal communication).

General structure of the thesis and objectives

The general objective of this thesis was to study certain aspects that were scientifically little documented at the time about wild cherry tree plantations growing in Mediterranean environments and therefore, to increase the available information to a more efficient and sustainable management of this type of plantions. That is this reason why this thesis covers different fields of study, from agronomy to forest hydrology, through ecology or ecophysiology:

In **chapter I** the roles played by three relevant agronomic factors affecting the wood production, i.e. the type of soil, the tillage regime and the irrigation regime, were evaluated. Also the experimental context of the thesis is presented, in which the soil characteristics of the experimental plot and a detailed description of the observed spontaneous herbaceous species in the non-tillaged part within the plantation are given.

Apart from the results observed in relation to tillage and irrigation, this chapter provides key information to understand the following chapters, that is, the experimental plot presented two contrasting areas with respect to soil properties, and therefore different soil performances for a correct development and growth of the studied species. One part was characterized by sandy loam texture, high proportion of gravels and high spatial variability on its physico-chemical properties, while the other part showed more homogeneous properties, clay loam texture and negligible presence of gravels. In this respect, although most measurements were taken for the entire experimental plot, each chapter emphasizes the most relevant results and/or methodological aspects: in chapter 4, chapter 5 and annex, the entire experimental and its related variability was considered plot, while chapters 2 and 3 only considered the soil type in which trees were growing under the best environmental conditions.

A detailed study of the rainfall partitioning is presented in *chapter 2*, where throughfall, stemflow and interception loss were monitored together with climate and forest structure variables. To this end, a subplot was established in the area of the plantation where the trees presented the largest sizes, and consequently where the highest effects of forest structure on the rainfall partitioning were expected.

The general objective of **chapter 3** was to determine the stand transpiration by sap flow sensors, and to analyze how the dynamics on soil water content and evaporative atmospheric conditions were affecting through simple modelling. On the other hand, since pruning is a common practice in this type of plantations for obtaining trunks free of branches, how this practice affected tree transpiration was studied and subsequently the water use productivity at plot scale.

Chapter 4 represents a complementary study to chapter 3. In this chapter were assessed the possible methodological errors arising from the use of sap flow sensors in noble wood plantations, and therefore how they may affect the scaling-up from measuring point (heat pulse velocity) to stand (depth of water). Since in chapter 3 the sensors were placed at 1.3 m height on the eastern sides and with two defined measurement points for each tree, an additional measurement effort was made here in order to assess whether this approach was good enough to give robust estimates of transpiration: sap flow was measured in four locations in several trees (north, south, east and west) and with an increased density of measurement points within the sapwood area. Moreover, several methodologies to estimate the sapwood area were also tested, since its precise calculation is a key aspect for the estimation of tree transpiration.

Originally, **Chapter 5** was foreseen to evaluate the effect of soil type, soil tillage and drip irrigation in the root water uptake pattern of trees within the plantation, through the use of stable water isotopes. The initial idea was to apply a mixed model that would elucidate the soil depths from which water was taken by roots, through iterative comparisons between the signal (i.e. isotopes composition) of xylem water and those from soil waters. However, after a deep analysis of the isotopes data, different errors in the experimental design and some methodological constrains arose, and thus making unfeasible to test some of the initial hypotheses. Therefore, this chapter is intended to serve as a guide for other works aiming to obtain the pattern of root water extraction in similar environmental conditions.

Finally, given the high sapwood area growth observed in the studied species during the vegetative period, different values for that area may be considered when using it as a scalar to estimate transpiration with sap flow sensors. For this reason, a small comparative study regarding this aspect is presented in the **annex**.

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CHAPTER I

The role of soil characteristics, soil tillage and drip irrigation in the timber production of a wild cherry orchard under Mediterranean conditions

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Chapter I

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Abstract

Over the last decade high-quality timber plantations have increased in Europe because of the constant high market price of timber and economical incentives from the EU. These latter are mainly due to timber plantations' role in CO2 capture. Noble wood plantations have also been established in Mediterranean areas, but many of them suffer from low growth rates due to deficient plantation management and/or non-optimal environmental conditions. Furthermore, little information exists about soil and water management in these plantations and how different soil characteristics may affect management results. In this study, a trial was established in a pure wild cherry plantation under Mediterranean conditions. The trial evaluated the effects that soil type (low soil quality versus good performance for woody crops), soil management (soil tillage versus no tillage), irrigation regime (drip irrigation versus no irrigation) and their interactions may have on wood production. Soil water content and the spontaneous vegetation that appeared in the alleys of the no-tillage treatments were also measured.

The results showed that sandy-clay-loam soil with a water-holding capacity of 101.5± 5.2 mm had 65% more wood volume increase during the study period than sandy-loam soil with a water-holding capacity of 37.9±8.0 mm. Conventional tillage or zero tillage with the presence of spontaneous vegetation did not differ significantly in wood volume increment, regardless of the type of soil. Although soil water content was significantly increased by tillage in sandy-loam soil, this effect was not enough to increase tree wood volume. On the other hand, the application of drip irrigation did increase wood production by up to 50%. Therefore, 10 years less on the plantation's rotation length can be anticipated when applying irrigation: from 40 to 30 years (sandy-loam soil) and from 56 to 46 years (sandy-loam soil).

In conclusion, deep soil characterization of the site is essential before deciding whether to develop a plantation of this type in areas under soil water content limitations caused by deficient soil structure and texture. In addition, our results show important savings can be made by reducing soil tillage, as less tillage leads to greater ground cover and biodiversity. Further investigations are required to examine how long-lasting the effects are and what other benefits can be expected when this type of plantation is managed in a more sustainable way.

Keywords: Agroforestry systems, wood production, *Prunus avium* L., soil water content, ground cover, spontaneous vegetation

Introduction and objectives

Wild cherry tree (*Prunus avium* L.) is present in most mixed-forest ecosystems with a temperate climate in Eurasia. Poland and Germany are the European countries where its natural area is greater, with about 40,000 ha in each (Ducci et al., 2013).

Wild cherry timber is one of the most highly valued noble woods in Europe and prices can reach up to 1,000 €/m³ (Loewe et al., 2013; Martinsson, 2001). Nowadays, though wild cherry plantations have steadily increased over the last 25 years in Europe, self-production is still far away from satisfying the furniture industry's demand (Ducci et al., 2013).

The increase in the cultivated area of noble wood has been promoted by reforestation programmes (e.g. EEC Regulation 2080/92), not only because of its economic value, but also due to its roles in improving biodiversity and CO2 capture and in the diversification of land uses (Cambria and Pierangeli, 2012).

Wild cherry plantations have also been established in Mediterranean areas, though to a lesser extent. Here irrigation is frequently used to confront summer drought (Ducci et al., 2013). This is especially necessary in soils with low water-retention capacity, as is the case in several places in Spain where plantations have been established (Vilanova, personal communication). Nowadays, many of these plantations suffer low growth rates due to deficient plantation management and/or non-optimal environmental conditions (Aletà and Vilanova, 2014).

Intensive management is normally required for noble wood plantations in Mediterranean regions. Silvicultural guidelines and scientific papers about managing this type of plantation focus on two main aspects: on the one hand, how timber production is affected by pruning or thinning (Cisneros et al., 2006; Kupka, 2007; Springmann et al., 2011) and, on the other, how the mixing of wild cherry trees with other species, in mixed plantations (Kerr, 2004; Loewe et al., 2013) or in agroforestry systems (Campbell et al., 1994; Chifflot et al., 2006; Dupraz et al., 1995), affects tree growth. In contrast, little attention has been paid to evaluating the effects of soil and water management on the timber production of pure plantations (Ripoll Morales et al., 2013) or to evaluating the effects of different management under distinct soil types.

Environment-friendly soil management is increasing in Europe due to EU incentives (e.g. Sustainable Land Management practices, UNEP-UNDP-UNCCD, 2008) and a more concerned society, especially in areas subjected to high erosion risk and water constraints such as the Mediterranean. Zero tillage or no tillage (NT), like the most extreme cases of reduced soil tillage (T), may contribute to reducing / compensating for the negative effects of conventional soil tillage on soil properties, such as high water loss because of direct soil evaporation, destruction of soil structure, nutrient losses, soil compaction, high erosion rates and increased surface runoff (Jemai et al., 2013; Palese et al., 2014; Soane et al., 2012). Moreover, crop yield in tree plantations is not generally less under NT than under T. Although a reduction in crop production under NT has sometimes been described (Martínez-Mena et al., 2013), in most cases the effect is not significant or is even positive. For example, Gómez et al. (1999), Hernández et al. (2005), Palese et al. (2014) and Soriano et al. (2014) found that production did not differ significantly under NT or T in Mediterranean olive orchards; Montanaro et al. (2012) found higher fruit yield under NT management in peach orchards; Raimundo (2003) showed that nut production was not significantly different between NT and T; and Martins et al. (2010) found that chestnut yield was lower under T than in the different NT systems tested. Thus, NT does not clearly promote reduction in the yield of fruit and nut trees in Mediterranean areas. These findings are a sound basis for the hypothesis that wood production will behave in a similar way.

One NT method is to allow spontaneous vegetation to remain, as against NT with commercial seeded cover crops. The first method is cheaper, may be stable over time due to self-reseeding (Bond and Grundy, 2001) and may enhance the biodiversity of an agro-ecosystem, since ruderal vegetation is an integral component of agro-ecosystems and plays an important role in diversifying the land (Marshall et al., 2003).

To the authors' knowledge, this is the first study of noble timber plantations that compares yield under different soil types, soil managements and irrigation regimes. To this end, a trial was established in a pure wild cherry plantation under Mediterranean conditions. It aimed to evaluate the effects that soil type (bad performance versus good performance for woody crops), soil management (soil tillage versus no tillage), irrigation regime (drip irrigation versus non-irrigation) and their interactions may have

on wood production. Soil water content and the spontaneous vegetation that appeared in the alleys of the no-tillage treatments were also complementary measured.

Materials and methods

Situation and environmental characteristics of the study site

The study was carried out from 2011 to 2013 at the IRTA-Torre Marimon experimental facilities (Caldes de Montbui, NE Spain, at 159m a.s.l).

The experimental plantation was located on an alluvial terrace with carbonated alluvial deposits as parent materials (IGME, 1976). Before tree planting, two soil samples revealed two different scenarios of water availability for plants growing there: in the eastern part, a sandy matrix with high gravel and stone content was found, while in the western part a silt matrix with some gravel was observed.

The climate is Mediterranean with mean annual values (1991-2010) for temperature, evapotranspiration and rainfall of 14.4 ± 0.2 °C, 846.8 ± 23.3 mm and 599.4 ± 33.4 mm, respectively.

Cherry orchard and experimental design

The trees in the experimental wild cherry plantation (clone Salamanca 4) were planted for timber production in 2008. Tree density was 625 trees ha⁻¹ with spacing of 4 m between trees and rows (16 m² per tree). Rows followed a north-south orientation. The mean values of height and diameter at breast height at the beginning of the experiment, in December 2010, were 4.7±0.1 m and 5.7±0.1 cm, respectively. In line with timber production practice for obtaining trunks free of branches, tree pruning in September 2011 and June 2013 removed approximately one third of the total biomass. Total dry biomass removed ranged from 0.8 to 10.9 kg tree⁻¹ as a consequence of tree vigour differences.

The experiment used a split-plot design with three replications arranged in a complete block design. The main plot factor was soil management (T, soil tillage or NT, no tillage) and the subplot factor was drip irrigation (I, irrigated or NI, non-irrigated). The subplots were separated from each other by buffer tree rows and each subplot contained four sample trees.

Irrigated treatments were drip-irrigated from May to September with 4 emitters (16 l h^{-1} tree⁻¹) located 25 and 50 cm from the trees on their north and south sides. Daily doses were calculated at the beginning of each week as a function of the weekly sums of reference evapotranspiration (ET₀) (Kc´ \pm T₀, Kc′ values from 0.26 to 0.6 depending on the month) and rainfall (R) during the previous week (I= Kc´ \pm T₀-R), and applied from Monday to Friday. There was no irrigation when ET₀ was lower than R.

Table I. Means (and standard deviations) of the main soil characteristics of the studied blocks. FC: field capacity, WP: wilting point. FC and WP were estimated according to Saxton et al. (1986).

			Volume of	FC - WP	Organic matter	
	Depth		stones and		_	
	(cm)	Texture	gravels (cm³/cm³ soil)	(% of soil volume)	(% of dry weight)	рΗ
		Sandy-				7.92
	0-25	loam	0.53 (0.09)	11.43 - 4.70	1.19 (0.10)	(0.05)
		Sandy-				
	25-50	loam	0.47 (0.12)	9.23 - 3.14	0.51 (0.09)	8.3 (0.12)
Block		Sandy-				
1	50-100	loam	0.47 (0.12)	8.80 -2.44	0.24 (0.06)	8.6 (0.06)
		Sandy-				7.88
	0-25	loam	0.46 (0.14)	18.79 - 9.05	1.56 (0.05)	(0.05)
		Sandy-				8.02
	25-50	loam	0.33 (0.19)	13.83 - 8.51	0.92 (0.17)	(0.07)
Block		Sandy-				
2	50-100	loam	0.33 (0.19)	14.15 - 6.10	0.58 (0.03)	8.3 (0.1)
						7.95
	0-25	Loam	0.24 (0.05)	26.22 - 13.82	1.45 (0.12)	(0.03)
						8.05
	25-50	Loam	0.04 (0.01)	24.21 - 12.67	0.81 (0.09)	(0.03)
Block						8.17
3	50-100	Loam	0.04 (0.01)	21.18 - 10.77	0.34 (0.10)	(0.02)

Total irrigation was 125, 214 and 300 mm in 2011, 2012 and 2013, respectively. The low irrigation amount in 2011 was due to an irrigation system malfunction on successive days in early summer.

Soil was tilled with a mouldboard ploughing to a depth of 30 cm every 3-4 months. In the no-tillage treatments, spontaneous vegetation was mowed every two months along the irrigation lines, and twice a year on the rest of the land surface. Plant residues were left on the ground as mulch.

Finally, several meteorological variables (rainfall, air temperature and humidity, wind velocity and direction and solar radiation) were monitored at an automatic weather station located in a clearing 50 m from the cherry plantation. Penman-Monteith FAO reference evapotranspiration (Allen et al., 2006) was calculated by using these data.

Soil characteristics

To characterize the soil variability within the experimental plantation in greater detail, three soil samples were taken during the study period:

- (i) A morphological characterization of soil profile in two open pits on the edges of the plantation (Table S1).
- (ii) Three composite samples in each subplot (depths of 0-0.25 m, 0.25-0.50 m and 0.50-1.00 m), from two points at distances I and 2 m from the trunk of an inside tree. The soil samples were dried and sifted through a 2-mm sieve to calculate the following characteristics (analytical methods specified): soil texture (densitometry), soil pH (soil: water ratio= 1:2.5, w/vol), soil salinity (electrical conductivity of soil extract 1:5 w/vol), calcium carbonate (after HCl treatment, the CO₂ emitted is manometrically measured) and organic carbon (acid-dichromate digestion) (USDA-NRCS, 2004).
- (iii) A specific sampling to determine gravel and stone content. Soil volumes from 0.0080 to 0.0156 m³ (0.20*0.20*0.20 m and 0.25*0.25*0.25 m) were extracted from two sampling locations per block. Samples were sieved through a 2-mm sieve to retain particles bigger than 2 mm. These pits were also used to measure soil bulk density (compliant cavity method, USDA-NRSC, 2004).

Soil water-holding capacity in the subplots was calculated by the Saxton et al. model (1986). This used the soil texture and soil depth calculated from (ii) and the volumetric

stone content calculated from (iii), with the latter considered as an empty soil volume (stone water-holding capacity = 0 mm).

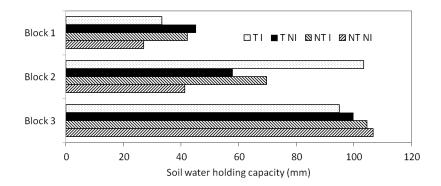


Figure I. Soil water holding capacity (mm) of each block and treatment: T I: Tillage irrigation; T NI: Tillage No irrigation; NT I: No Tillage Irrigation and NT NI: No tillage No irrigation.

The main characteristics of the soil profiles observed were (see Table I for detailed information according to soil depth): i) the gravel and sand content was higher in block I than in block 3; block 2 was the transitional stage between them; ii) the organic matter content was very low in the topsoil of all blocks; and iii) no soil nutrient deficiencies were observed. The high range of soil texture and gravel content resulted in sharp differences in soil water-holding capacity between blocks I and 3 (Figure I). For its part, block 2 showed soil characteristics from the other two blocks and thus higher variability in the soil water-holding capacity of its subplots (Figures I and 2).

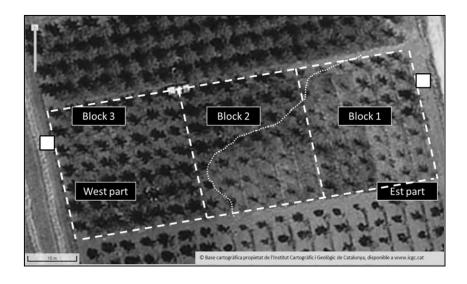


Figure 2. The cherry orchard study plot located at Torre Marimon (Caldes de Montbui, Barcelona, NE Spain) with the situation of the two pits opened (white square). The dotted line indicates the boundary between the two types of soil observed (see text for details), ICGC (2014).

Evaluating the wood volume increment in cherry trees

Wood production was expressed as the volume increment of saleable wood from the cherry trees (WVI, m³) between January 2011 and December 2013. As trees in 2013 still showed good apical dominance, we considered that the entire volume increment during the study period was saleable wood. To this end, diameter at breast height (DBH, cm) and total height (H, m) at the beginning and at the end of the study period were used as follows, by considering tree form as a conical frustum:

WVI =
$$I/3 \cdot \Pi \cdot H \cdot [R^2+r^2+(R \cdot r)]$$

where R and r are the circumference radius at the tree base and at the maximum tree height (H), respectively. R and r were estimated as a function of DBH according to a linear regression of diameter versus height ($y = -1.44 \times + 11.86$, $R^2 = 0.98$, N = 10.818 trees), calculated with data from 6 different wild cherry clones for timber where diameter was measured in 50-cm height increments along the tree trunks (Vilanova et al., unpublished data).

Volume increment of saleable wood was analysed by analyses of variance and least-square mean separation methods, following the above-mentioned split-plot experimental design. The statistical analyses were performed twice: taking into account the three blocks and ruling out block 2, as it had characteristics that were a mix of the other two blocks, thus reducing variability in the data for this heterogonous block. Both the main plot factor (soil management) and the subplot factor (irrigation) had two levels, with block considered a fixed factor. All analyses were computed by the GLM procedure of Statistical Analysis Systems (SAS, 1999).

Complementary measurements: soil water content under tree cover and spontaneous ruderal vegetation

Volumetric soil water content (cm³ cm³) was measured at 0-30 cm soil depth from May 2012 to September 2013. Twelve 30 cm-long TDR probes (details of the probes in Martínez Fernández and Ceballos, 2001), one in each subplot, were placed vertically 12.5 cm north of one of the four sampled trees (equidistant from the first drip emitter and the tree trunk in the irrigated treatments). Every two weeks, readings were taken with a TDR measuring device (Time Domain Reflectometry, TEKTRONIX, 1502 C) and were later converted to soil water-content values, following Topp et al. (1980). T-student tests compared NT and T treatments in each block studied. Previously,

homogeneity of variances was tested (Levene test). All analyses were computed using Statistical Analysis Systems (SAS, 1999).

The spontaneous ruderal vegetation in the NT plots was monitored during the three seasons (2011-2013). Two permanent 0.25 m² randomly assigned quadrats were placed in each of the three blocks considered. Each quadrat was sampled to obtain the species composition, following Bolòs et al. (1993), and the total surface covered by vegetation through visual identification (%).

The plant community was analysed in terms of floristic and functional structures. Floristic structure was summarised in terms of biodiversity. Functional structure was characterised by Raunkiaer life forms and the flowering season.

In addition, in 2012 we sampled on 9 different days from April to mid-June to obtain the specific aboveground biomass (g m^{-2}) and the surface covered by each species. The biomass data were used to calculate the Shannon-Wiener diversity index (H') as follows:

 $H' = \sum p_i \cdot log_2 \cdot p_i$

where p_i is the proportion of the biomass of i species in the total sample of S species.

Biodiversity and the proportions of total cover were subjected to pooled analyses of variance for measurements over time (Gomez and Gomez, 1984), with two main factors considered: block and time of observation. Homogeneity of variances was tested: square root transformation was chosen to homogenize the variance in biodiversity, while the arcsine of the proportion was chosen for total ground cover. When the main effects or the "block x time of observation" interaction were significant (P<0.05), least square means were computed and compared (Tukey-Kramer method). The total aboveground biomass (g m⁻², dry weight; only for 2012) data were transformed (natural logarithm) prior to a single-factor (block) analysis of variance. The GLM procedure of Statistical Analysis Systems (SAS, 1999) was used for the analyses.

Results

Meteorological conditions during the experiment

Annual rainfall (2011, 2012 and 2013) was 773, 462 and 640 mm, while reference evapotranspiration was 1,026, 1,025 and 854 mm, respectively. Figure 3 shows the

contrasted monthly distributions of rainfall and potential evapotranspiration, together with the average monthly values for air temperature. All the growing seasons were characterized by rainfall deficits and high water evaporation demands (up to 3.7 kPa for DPV).

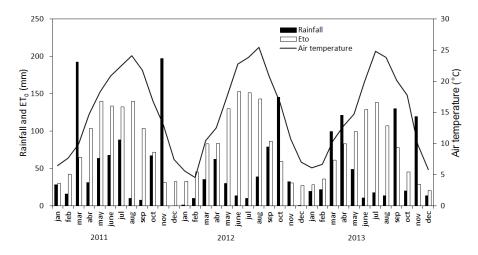


Figure 3. Monthly cumulated rainfall and potential evapotranspiration (ET0, mm) and mean monthly air temperature during the 3 years of experiment (2011, 2012 and 2013).

Wood volume increment in the wild cherry trees (WVI)

Results from the analysis showed that WVI was mainly affected by irrigation, with volumes up to 50% higher under the irrigation regime than under no irrigation, which highlights the importance of irrigation during stress periods under Mediterranean climate conditions, as was expected (Table 2). In contrast, soil management has no significant effect on WVI, although, in the analysis that took into account the three blocks, the block x soil management interaction was highly significant (P=0.0014).

Table 2. Analysis of variance of the effects of block, soil tillage, and irrigation on the wood volume increase (m3).

Considering three blocks				Considering two blocks				
Factor	d.f.	MS	F value	Р	d.f.	MS	F value	Р
Block (B)	2	0.000645	9.98	0.091	I	0.001202	186.28	0.046
Soil tillage (T)	ı	0.000383	5.92	0.135	I	9.1569E-05	14.19	0.165
B x T (Error a)	2	0.00006466	7.75	0.001	I	6.451E-06	1.65	0.210
Irrigation (Ir)	I	0.000909	109.02	<.0001	I	0.000294	75.04	<.0001
T x Ir	I	0.0000003	0.04	0.850	I	1.606E-06	0.41	0.527
Residual	40				26			

The relative importance of the two factors involved in the interaction can be measured by observing the ANOVA performed with only two blocks, where the block effect was significant while the soil management effect was not. The least-square means of the levels of the effects considered in the ANOVAs (Table 3) revealed that the block effect was more important than the soil management treatment effect in explaining the variance in WVI. There was a great difference in the means comparing block I and block 3, achieving intermediate values in block 2. On excluding block 2 from the analysis, significant differences between block I and 3 were found in the WVI means, whereas the mean values obtained in tillage and in no-tillage treatments were not significantly different.

Table 3. Least square means of the levels of the significant (P<0.05) sources of variance (see Table 2) of the analysis on the wood volume increase (m^3). Least square means of each factor with different letter were significantly different at P<0.05 (Tukey-Kramer test).

Considering three blocks				Considering two blocks			
Factor	Levels	LS means		Factor	Levels	LS means	
<u>Ir</u>	no-irrigated	0.0094	A	lr	no-irrigated	0.0097 A	
	irrigated	0.0181	В		irrigated	0.0158 B	
BxT	BI - no tillage	0.0054	Α	В	ВІ	0.0066 A	
	BI - tillage	0.0079	AB		В3	0.0189 B	
	B2 - no tillage	0.0106	В				
	B2 - tillage	0.0207	D				
	B3 - no tillage	0.0168	С				
	B3 - tillage	0.0210	D				

Soil water content under tree cover

Figure 4 shows the means of soil water content averaged through the blocks during the study period and grouped by irrigation conditions. The variability of the time series (standard deviations) indicated that soil type clearly affected soil water content, especially in the irrigated treatments in the 2012 and 2013 growing seasons (Figure 4.b). In these irrigated treatments, variation in mean soil water content during the study period was low (from 0.4 to 0.3 cm³ cm⁻³). In contrast, soil water content in the NI treatments had higher ranges, reaching very low values (close to 0.1 cm³ cm⁻³) in summer periods and recovering to maximum values of 0.37 cm³ cm⁻³ in the 2013 winter.

Furthermore, NT and T treatments behaved in different ways, depending on the block. No significant differences were found in blocks 2 or 3 in the t-student tests (p=0.55 and p=0.67, respectively), while soil tillage involved higher soil water content in the T treatments of block I (p=0.042).

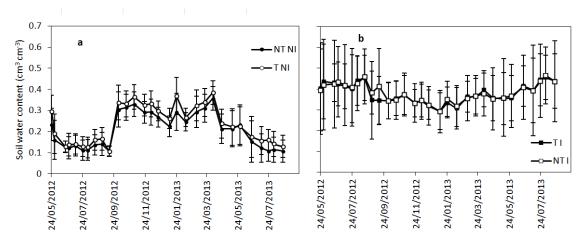


Figure 4. Means and standard deviations of soil water content in the 0-30 horizon (cm³cm⁻³) for the treatments grouped by the irrigation subplot effect: a: NT NI: No tillage No Irrigation and T NI: Soil Tillage No Irrigation.

Spontaneous ruderal vegetation in the NT plots: biodiversity, cover percentage and biomass

During the study period, a total of 45 species from 17 botanical families were observed in the permanent quadrats located in NT plots (Table S2). The number of taxa and botanical families increased over the three years.

Therophytes were the predominant life form (81.4%, or 93% if 5 taxa that are not exclusively therophytes are included). The predominant morphotype was dicotyledonous (86.7%). The specific composition of the plant community made blooming possible throughout the year, but it was particularly important from April to September, when the highest percentages of species at that phenological stage were found (>50%) (Table S2).

As all the sources of variation that affect biodiversity and total ground cover were significant at p<0.05 level except block, ground cover did not differ much between the three blocks in the plantation. The overall mean values were 6.83 for biodiversity and 53.78% for total ground cover. The block x time interaction was clearly significant (P< 0.0005 in both variables); the evolution of the total ground cover least square means over time showed a similar trend for the three blocks, although the curves were interlaced (Figure 5).

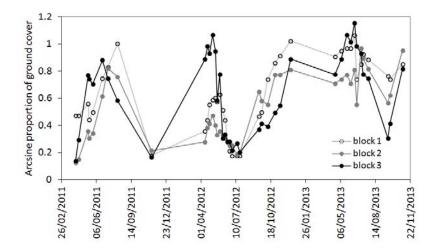


Figure 5. Least-square means of the total ground cover (arcsine transformed) over time in the no tillage plots.

For the aboveground biomass of the spontaneous vegetation (AB) and its ecological diversity, calculated by the Shannon-Wiener diversity index, H', between 18 April and 13 June 2012, the results showed that there were significant differences between blocks (analyses not shown). Block 3 had the highest AB and at the same time the lowest H'. Thus, there was also a gradient of ecological diversity in the study plantation. The mean values of the H' were around 2, which dramatically decreased at the end of the spring period.

Discussion and conclusions

This study evaluated the effect of two contrasted soil types (one with low soil quality for woody crops and one of good agronomical quality), two irrigation regimes (drip irrigation versus no irrigation) and two kinds of soil management (soil tillage versus no tillage with the presence of spontaneous vegetation) on the timber yield of a wild cherry plantation during three years and under Mediterranean climate conditions.

Soil type led clearly to tree growth differences, as expected. The trees growing in sandy-clay-loam soil and the soil water-holding capacity (SWHC) of 101.5± 5.2 mm (block 3) showed 65% greater wood volume increase during the study period than the trees growing in sandy-loam soils, with a high presence of gravels and thus lower SWHC (37.9±8.0 mm) (block 1) (Table 2, right). However, when also considering data from block 2 in the ANOVA analysis (Table 2, left), the block effect was not significant. This was because soil in block 2 showed characteristics intermediate between the other two blocks and thus greater variability (SWHC of 68.1±26.3 mm). In

consequence, in the ANOVA analysis with the three blocks, the block effect was smaller and the error term was higher, resulting in lower sensitivity to finding significant differences between the two contrasted soils in the timber production of our experimental plantation.

Conventional tillage (3-4 times per year to a depth of 30 cm) in our young wild cherry plantation did not lead to any significant differences in wood volume increment from years 4 to 6 of the plantation, when compared with zero tillage with the presence of spontaneous vegetation. Water availability is the main limiting factor when planting trees in association with herbaceous vegetation in dry climates such as the Mediterranean (Baldy et al., 1993; Miller and Pallardy, 2001). Observations in other Mediterranean tree plantations with zero tillage showed that fruit or nut yield did not increase when conventional tillage changed to natural vegetation ground cover (Gómez et al., 1999; Hernández et al., 2005; Martins et al., 2010; Palese et al., 2014, Soriano et al., 2014). For its part, agroforestry systems with wild cherry trees in Mediterranean areas have shown contradictory results in relation to tree growth. Dupraz et al. (1995) observed a big decrease in trees' water use due to the presence of perennial herbaceous crops with larger root systems. In contrast, Chifflot et al. (2006) studied the effect that the presence of intercrop or spontaneous vegetation had on tree diameter growth as compared to weeded control by herbicide. These authors found greater diameter growth in the intercrop system, followed by control and by spontaneous vegetation.

In our case, the complementary measurements of soil water content to a depth of 30 cm and of spontaneous vegetation could help to interpret our results. The effect of soil tillage on soil water content varied according to soil type. Soil water content in block I increased through soil tillage by I4%, while no significant increases were observed in either block 2 or block 3. The results obtained in blocks 2 and 3 corroborate previous studies carried out under Mediterranean conditions, where no tillage had similar effects or was even more positive than soil tillage for both soil water storage and soil water dynamics (Celano et al., 2011; Martins et al., 2010; Palese et al., 2014). In this respect, the significant increase in soil water content induced by soil tillage in block I should have been counterbalanced by other effects promoted by no-tillage management, as the wood volume of wild cherry trees was not significantly affected by

soil tillage in block I. Thus, the non-significant differences in spontaneous vegetation ground cover (visually estimated) between the blocks, or the lower aboveground biomass (calculated by weighing dry matter) found in block I than in the other two blocks, seem to indicate that the competition between trees and ruderal vegetation for resources such as water, light and nitrogen was very similar in all blocks or even lower in block I. Therefore, we hypothesized that other negative factors caused by soil tillage on soil structure were of greater weight in block I, although they are not sufficient to lead to timber differences.

The use of drip irrigation increased wood production in all types of soil studied, with up to 50% higher production under irrigation. This underlines the importance of avoiding soil water deficits if this species is to develop correctly in Mediterranean climate conditions, as expected (Juhász et al., 2013). This is especially important when assessing the rotation length of the plantation, i.e. the time required for obtaining optimum wood of high quality (diameter of 40 cm at breast height), and thus the economic benefits from the plantation. In this respect, by using diameter growth curves proposed for this species (Cisneros, 2004), and assuming that the observed differences are maintained during the whole lifespan of the plantation, a reduction of 10 years in rotation length would be expected when applying irrigation. In block 3, this would mean a reduction from 40 to 30 years (non-irrigated versus irrigated trees), whereas at the opposite site, block 1, the reduction would be from 56 to 46 years.

As soil tillage could be avoided and assuming that our results are maintained from year 3 to year 8 (6 years of soil tillage: during the first and second years soil tillage is required, since competition between young trees and spontaneous vegetation is likely to appear), we estimated the cost of dealing with soil tillage. Considering the normal prices in the region for mouldboard (100€ per hour; 3 hours required for the total plantation, 4 times per year) and for mowing the spontaneous vegetation (10€ per hour; 4 hours required for the total plantation, 4 times per year), we calculated that an 87% cost reduction could be achieved. This would be greater if the non-significant differences in wood growth are maintained in the future.

In conclusion, our results indicated that timber production with wild cherry trees is greatly affected by the water available in soil. Therefore, the deep soil of the site has to be characterised before a decision can be taken on whether a plantation of this type

could be developed in areas under soil water-content limitations, such as block 1. Furthermore, drip irrigation gave higher timber production in all the soils studied, which could reduce rotation length by 10 years. Thus, this aspect should be carefully evaluated when the economic aspects of a plantation of this type are being assessed. However, in the short term, wood yield was not affected by soil tillage, as observed in other tree plantations, which suggests that less intense soil management would be possible in these plantations. This would also occasion environmental benefits such as the increased biodiversity of plant ground communities. Further studies of this type of plantation are required, to examine other questions, such as how long effects last (i.e., is the lack of effect of soil tillage on wood volume maintained during the whole lifespan of the plantation?), the different effects of soil tillage on soil water content depending on the soil type, and other benefits that could be expected if and when land is managed in a more sustainable way.

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Rainfall partitioning in a young wild cherry tree plantation: has the interception a significant role in the water balance?

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Chapter 2

Rainfall partitioning in a young wild cherry tree plantation: has the interception a significant role in the water balance?

Abstract

During two years, we investigated the spatial and seasonal variations of the rainfall partitioned into throughfall and stemflow in a young wild cherry tree plantation under Mediterranean climatic conditions, in relation to canopy and bulk rainfall characteristics. To this end, throughfall was measured by 58 tipping buckets randomly distributed in a plot within 8 trees, and stemflow was monitored in all the trees. Canopy cover was periodically measured with sky-oriented photographs in the throughfall devices and by considering a mean inclination angle from the 83 rainfall events studied. Observed values of throughfall and stemflow for the whole study period were 96 and 1.1 % of bulk rainfall, thus interception loss only accounted for a 3 % of bulk rainfall. At event scale, throughfall ranged from 70 to 114 % while stemflow did from 0 to 3 %. No significant effect of the canopy cover increase from one growing period to another was observed in throughfall, although this water flux was reduced by 4 %. Dripping points which spatially concentrate throughfall were observed in the devices with higher canopy cover. This result was the responsible for rightly skewing the throughfall distributions and as a possible explanation to the observed positive correlations between throughfall and canopy cover. Finally, from the calculations to obtain the optimum numbers of tipping buckets for measuring throughfall, we conclude that 10 or 48 tipping buckets were suitable for measuring this water flux at the whole study period or event scales, respectively.

Keywords: throughfall, stemflow, timber, hydrology, noble wood

Introduction and objectives

Gross rainfall in vegetation ecosystems is partitioned into throughfall, stemflow or interception loss. Throughfall and stemflow are the water cycle components which naturally provide water supply for plant use and growth, while interception loss represents the part of gross rainfall which is captured and after evaporated from the vegetation surfaces, and thus does not contribute to soil recharge.

Rainfall partitioning is influenced by both climatic factors and structural properties of vegetation (Crockford and Richardson, 2000; Levia and Frost, 2006). In some Mediterranean areas, characterized by high atmospheric demand, annual interception loss can represent up to of 30 % of gross rainfall (Llorens and Domingo, 2007). These interception rates are found in natural forest or poorly-managed forest plantations (Shachnovich et al., 2008; Molina and Del Campo, 2012). However, in sparse forest or in forest/plantations intensely managed, throughfall and stemflow play a more important role in the local water cycle (Molina and Del Campo, 2012; Teklehaimanot et al., 1991). In this sense, several studies have observed stand interception loss rates lower or close to 10% of gross rainfall (Gash et al., 1995; Jackson, 2000; Schnabel and Mateos, 2001; Samba et al., 2001).

High spatial variability of throughfall is expected in clumped forest, characterized by high tree spacing, especially during wind-driven rainfall events (David et al., 2006). Reduced tree density increases the ventilation in forest ecosystems and leads to higher evaporation rate during rainfall (Teklehaimanot et al., 1991), enhances the proportion of throughfall which directly reaches the soil surface without touching vegetation surfaces (Teklehaimanot et al., 1991; Molina and Del Campo, 2012), and also reduces the canopy storage capacity of vegetation to intercept water (Pypker et al., 2011). On the other hand, although most frequently reported for tropical rain forests (e.g., Lloyd and Marques, 1988, Zimmermann et al., 2009), the concentration of throughfall in certain points (i.e. drip throughfall points) and the subsequent reduction in others, may be favored by canopy structures or leaf morphologies such as inclined branches or leaves. Therefore, rainfall partitioning mechanisms in forest/plantation ecosystems are complex and local variability of these water fluxes is expected to be linked to canopy variability (Pypker et al., 2011).

Plantations for producing wood of high quality have increased in Europe over the last decades because of a constant high market price of high-quality timber and economical

incentives from the EU to promote the reforestation of rural abandoned areas (Ducci et al., 2013). To our knowledge, no specific crop factors are available for modulating irrigation (Allen et al., 2006) in this type of plantations, so they are normally taken from fruit tree plantations (ex. Ripoll Morales et al., 2013). Therefore, rainfall partitioning studies are essential to a correct estimation of crop factors in plantations of noble wood.

The general objective of this work was to evaluate the magnitude of the rainfall partitioned into throughfall, stemflow and interception loss in a young wild cherry plantation (*Prunus avium* L.) characterized by low tree density and under Mediterranean climate conditions. Since the study species is deciduous, different rainfall partitioning patterns are expected between the leafed and leafless periods not only depending on forest structure, but also on differences in rainfall characteristics between seasons. Therefore, the effects of canopy and rainfall characteristics on the rainfall partitioning were also assessed. Finally, in order to obtain a robust estimation of mean throughfall in this type of plantation, the optimum number of tipping buckets was calculated.

Materials and methods

Study site and experimental plot

The present study was conducted on the IRTA facilities of the Torre Marimon site (Caldes de Montbui, Spain) at an elevation of 170 above sea level (41° 36 '47 '' N, 2°10'11 ''E). This area has a Mediterranean climate with a hot summer and a moderate winter. The rainfall distribution is similar among the seasons (1915-2102): 27, 21, 33 and 19 % of rainfall in spring, summer, autumn and winter, respectively. Mean annual rainfall, reference evapotranspiration and mean annual temperature are 622 ± 163 mm, 846.8 ± 23.3 and 14.4 ± 0.2 ° C.

The study plot was located within an experimental wild cherry tree plantation planted in 2006 and managed to timber production. Tree density is 625 trees ha⁻¹ with 4 m of spacing between trees and rows (16 m² per tree). Mean tree height in December 2011 was 4.7±0.1 m and mean diameter at breast height was 5.7±0.1 cm; about 3 m of tree height was free of branches at this time period as consequence of two previous prunnings. Soils in the plantation are of two types, sandy-loam soils with about 60 % of gravels and loam soils with negligible presence of gravels. This difference in soil

characteristics highly affected tree growth rate, and the trees growing in the former were rather limited (Molina et al., 2016). For the rainfall partitioning experiment presented here, an experimental plot of rectangular shape $(8x16 = 128 \text{ m}^2)$ was established in the soil type with no limitations for tree growth.

According to timber production practices, trees in the experimental plantation were pruned at September 2011 and June 2013, removing from 2 to 4 main branches from the lowest part of the crowns. However, as the objectives in this study did not include assessing the direct effect of pruning on rainfall partitioning, the June 2013 pruning was not carried out in the experimental plot.

Bulk rainfall and meteorological data

Bulk rainfall was measured in an open area close to the experimental plot, by means of two tipping buckets (Rain-O-Matic, 0.2 mm per tip and orifice of 200 cm², Pronamic, Silkeborg, Denmark; ECRN-50, I mm per tip and orifice of 50 cm², Decagon Devices, Pullman, USA) placed at I m height. In a tower located 2 m above the plantation canopy and at about 5 m from the experimental plot, measurements of net radiation (Q-7 Net Radiometer, Campbell Scientific, Logan, USA), air relative humidity and temperature (RH-T sensor, Decagon Devices, Pullman, USA) and wind direction and velocity (Davis cup anemometer, Decagon Devices, Pullman, USA) were carried out. All data were recorded every 10 min intervals (tipping buckets: EMCR-50, Decagon Devices, Pullman, USA; measurements at tower: CR1000, Campbell Scientific, Logan, USA).

The bulk rainfall measurements were regularly calibrated during the study period. The linear regression comparing the bulk rainfall measured by the two tipping buckets resulted in slope and intercept which were not statistically different than I and 0, respectively (R² = 0.99), and the mean relative error equal of 0.07 %. Also, the calibrated data was systematically verified by comparing with the data from an official meteorological station at ca. 500 m from our experimental plot (Servei Meteorològic de Catalunya, UTM X430803 and UTM XY4607309).

For rainfall partitioning analyses, it is necessary to establish the time among rainfall showers, i.e. the inter-event time, to define a rainfall event. In our case, rainfall showers with time exceeding 8 h between them were considered as different events.

This assumption seems to depart from reality in many events during our study period, with less time necessary to dry the canopy. However, as we did not carry out complementary measurements to assess the inter-event time directly (Llorens et al., 2014), we finally decided to consider this to be consistent with the majority of rainfall interception studies (e.g. Klaassen et al., 1996; Limousin et al., 2003). Therefore, 83 rainfall events > I mm occurred during the study period (see below). The averages for rainfall size, rainfall intensity, maximum intensity during 30 minutes and rainfall duration were estimated from the two rainfall tipping buckets.

Throughfall and stemflow

Throughfall and stemflow were measured from March 2012 to March 2014. Throughfall was measured with 58 automatic throughfall collectors randomly placed in the same fixed position at 30 cm height (Figure I). The collectors were the same 0.2 mm tipping bucket devices than the one used for measuring bulk rainfall (Rain-O-Matic, 0.2 mm per tip and orifice of 200 cm², Pronamic, Silkeborg, Denmark). The data was recorded every 10 min (Em5b data logger, Decagon Devices, Pullman, USA). To randomly locate the throughfall collectors, the experimental plot of 128 m² was divided into 128 subplots of I m side length, then 58 subplots were randomly selected and the collectors were placed at the centre (Figure 2).



Figure 1. Photographs of the experimental plot during installation in February 2012. Note that manual stemflow collectors were changed to 10 L containers 2 weeks later.

Stemflow was measured in the 8 trees within the experimental plot with polyurethane collars sealed with silicone rubber following Llorens et al. (1997). Two stemflow rings were connected to 0.2 mm tipping buckets and recorded every 10 minutes (Rain-O-Matic, 0.2 mm per tip and orifice of 200 cm², Pronamic, Silkeborg, Denmark); the other six stemflow rings were connected to 10 L containers. The 10-minutes stemflow values for these 6 trees were obtained from the weekly weighted 10 L containers

according to the average temporal distribution given by the two tipping buckets (following Valente et al., 1997). The equivalent stemflow depth for each tree (mm) was obtained by considering the tree density of 625 tree ha⁻¹.

To ensure the quality of data, several actions were carried out during the study period: cleaning of all the tipping buckets I-2 days before the rainfall events occurred. Calibration of the tipping buckets was manually assessed every six months. Finally, weed was regularly cut with a brushcutter to keep them with a height of 5 cm, and the cuttings were left as mulching to prevent the splash into the throughfall tipping buckets.

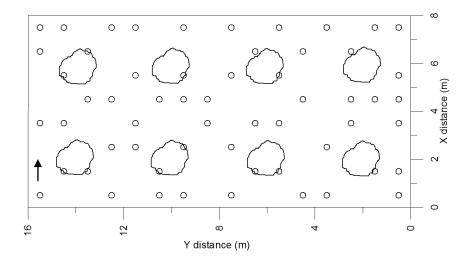


Figure 2. Monitoring design of the experimental plot showing the 58 tipping buckets for measuring throughfall and the 8 trees in which stemflow was measured.

Optimum number of tipping buckets for measuring throughfall

Two methodologies were used to calculate the optimum numbers of tipping buckets for measuring throughfall: the Monte Carlo routine (MC) and by prefixing an error in % of throughfall mean (PE). MC sampling was performed following Rodrigo and Avila (2001); throughfall data from the 58 collectors were 250 times randomly ordered, and for each random grouping (2-58 collectors), the mean and other statistics such as the coefficient of variation (CV) and the 95 % confidence interval were calculated. In Figure 3 are shown 2 examples of calculation.

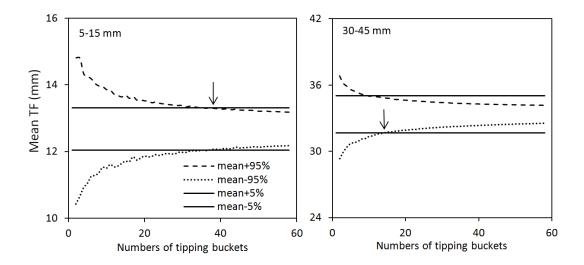


Figure 3. Two examples showing the throughfall averages \pm 5% (from the 58 tipping buckets) and the mean for each grouping (2-58) \pm 95 % confidence intervals. For the 5-15 mm class, the MC method gives 38 tipping buckets while 18 for the 30-45 mm class.

PE was carried out following the approach of Kimmins et al. (1973). The optimum number of tipping buckets (n) was calculated as:

$$n = (Z_c^2 CV^2)/c^2$$

where Z_c is the critical value of the 95 % confidence level (Z_c = 2, Holwerda et al., 2006), CV is the coefficient of variation and c is the prefixed error (5% of the mean).

All the data used was checked for normality by the Kolmogorov-smirnoff test modified by Lilliefors and by observing the residual plots.

Canopy cover as a function of rainfall inclination angle

Canopy cover was estimated every 20 to 30 days above the 58 throughfall collectors during early morning hours by means of 35mm lens photographs. A sky-orientated photograph was taken from the centre of each device with a Canon EOS 400D digital camera mounted on a tripod at 50 cm height (20 cm from the throughfall collector) and horizontally levelled. For each rainfall event, a rainfall inclination angle was calculated following Herwitz and Slye (1995). A mean rainfall inclination angle of 7.89° from the vertical (range from 0 to 27.09°) was obtained from all the rainfall events and the central part of each photograph, corresponding to this rainfall inclination angle, was used to obtain the canopy cover. To this end, the methodology described in Llorens and Gallart (2000) was applied by using the Greenpix software for image treatment

(Casadesus et al., 2007). A total of 1856 photographs (58 locations x 32 photos at each location) were processed following the described methodology.

Biometric measurements

Tree diameter at breast height (DBH), canopy cover in two fixed positions 50 cm apart from each tree (north and south aspects) and height were regularly measured during the study period in the 8 trees within the experimental plot. Furthermore, in the leafed period of 2013, the number and angle of branches were also measured (Table 1).

Table 1. Tree characteristics in the experimental plot. Data is from August 2013. Green and brown covers were estimated with the Greenpix software for image treatment (Casadesus et al., 2007).

Tree	Height (m)	DBH (m)	Green cover (%)	Brown cover (%)	Number of main branches	Mean angle branches (°)
1	6.7	11.2	57.0	2.8	15	67.5
2	6.5	10.8	61.7	4.5	19	58.3
3	6.4	11.1	62.4	3.2	13	60.0
4	5.9	10.7	68.0	3.1	8	32.0
5	6.2	10.7	67.0	4.7	15	57.8
6	6.4	11.1	77.4	2.8	20	65.5
7	6.4	10.5	63.7	3.6	17	63.3
8	6.5	11.1	76.0	2.9	13	66.7

Results

Canopy cover

Canopy cover (CC) followed the common pattern described for deciduous forests, with two contrasted leafless and leafed periods along the year and the transition periods between them (Figure 4). The comparison between the leafed periods indicated that canopy cover doubled in both magnitude and variability in the second year of study, increasing from 18.3±9.1 % to 40.8±23.1 %. This increase in CC was particularly important in locations with values between 20 and 40% in 2012 (inlet in Figure 4), while the locations with the lowest (around 0 %) and the highest CC values gave very similar values between 2012 and 2013. In contrast, no significant differences were found when compared the leafless periods, thus indicating that the increase in the woody parts of canopy did have a negligible effect on the overall CC increase from

one year to the other. Therefore, we considered the two leafless periods as a single leafless period for the next rainfall partitioning analyses.

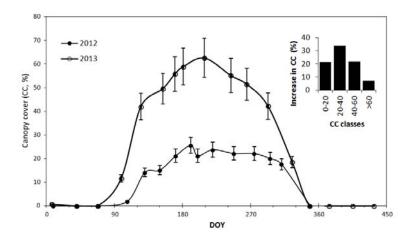


Figure 4. Means and standard deviations of canopy cover during the study period (n= 58 for each sample day). Inlet figure: Mean canopy cover increase between 2012 and 2013 for classes of 20% length. DOY = day of the year.

Rainfall characteristics

Over the 24-month study period, the observed 83 rainfall events yielded a total rainfall of 1090 mm, or 754.6 mm and 335.4 mm in the leafed and leafless periods, respectively. Most of the events (53%) were of small size (1-5 mm) and accounted for 11 % of the total rainfall. 4 events larger than 60 mm were recorded, and they accounted for 30 % of the total gross rainfall (Figure 5). The maximum rainfall intensity during 30 minutes and the duration of the rainfall events varied greatly, from 0.8 to 80.8 mm h⁻¹ and from 0.2 to 56.5 h, respectively. 73 % of the rainfall events had average wind speed from 0.4 to 10 m s⁻¹ (41 and 33 % from 0.4 to 5 and from 5 to 10 m s⁻¹, respectively), while 5 % of rainfall events had average wind speed higher than 20 m s⁻¹. Most of the winds during rainfall were easterly winds (32 % NE and 58 % SE).

On the other hand, the distributions of rainfall size and event frequency were very similar between the 2012 and 2013 leafed periods. In contrast, the leafless period was characterized by few events in the intermediate classes and two events of high size which accounted for the 15.2 % of the total rainfall (Figure 5).

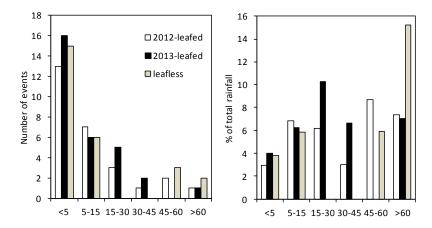


Figure 5. Rainfall event frequency and rainfall size distribution classified by phenophases, the 2 leafed periods in 2012 and 2013 and the whole leafless period.

Throughfall: Whole study period scale

Cumulative throughfall for the 58 collectors was divided by corresponding bulk rainfall and multiplied by 100 to give relative throughfall for all the study period. Relative throughfall was normally distributed but positively skewed, with mean \pm standard deviation values of 96±7 % and ranging from 86 to 120 % (Table 2). Most of the throughfall data felt in the range from 86 to 100 % (79 % of the total), and 20% of the collectors showed cumulative throughfall higher than 100 %.

Table 2. Statistics for the relative throughfall for all the study period. Throughfall values are expressed as percentage of bulk rainfall (BR). SD is the standard deviation value and CV is the coefficient of variation. The skewness in the throughfall distribution was tested by the Fisher coefficient test and the Kolmogorov-Smirnov test was used to test the normality.

Number of observations (N)	58
BR (mm)	1090
Mean throughfall (%)	96
Median throughfall (%)	94
Maximum throughfall (%)	120
Minimum throughfall (%)	86
SD throughfall (%)	7
CV (%)	8
Skewness (Fisher coef.)	1.01
p-value (Kolmogorov-Smirnov test)	0.365

On the other hand, the relative throughfall was calculated for each studied year in order to study the spatio-temporal persistency during the study period. The relative throughfall distribution was not constant between the study years and it seemed to

follow a random pattern (Figure 6). In addition, most of the collectors gave differences lower than 10 % (n=46), 9 between 10 and 20 % and 3 between 20 and 32 %.

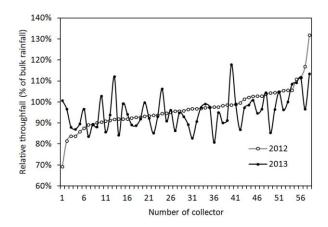


Figure 6. Relative throughfall for the 58 collectors in 2012 and 2013. Data from 2012 was ranked while 2013 data was not.

Throughfall: Event scale

A first examination of the throughfall data for the 83 sampled events revealed a high correlation between the mean throughfall and the bulk rainfall (y=0.96x-0.023, $R^2=0.99$, n=83). Spatial variability, expressed by the coefficient of variation between collectors (CV), decreased exponentially with event size, with CV from 40 % to 18% for events < 20 mm and an almost constant CV of 10 % for events with size of 20 mm on (data not shown).

The shapes of the throughfall frequency distributions showed more heterogeneous results. Throughfall was not normally distributed in 35 % of the rainfall events. The Fisher skewness coefficient had a wide range (from -2.3 to 2.2) and showed two different patterns depending on event size: events larger than 40 mm were always right skewed, whereas the skewness in smaller events was more random (Figure 7; left). Furthermore, 23% of the events had throughfall higher than bulk rainfall, but any of these events was larger than 40 mm. This indicates that the locations that rightly skewed the throughfall distributions did not produce a significant effect on the mean throughfall. In this sense, the octile skew as a measure of skewness insensitive to extreme values (Zimmerman et al., 2009), also showed that most of the events that required a transformation due to skewed distributions (values between -0.2 and 0.2) were smaller than 40 mm (Figure 7, right).

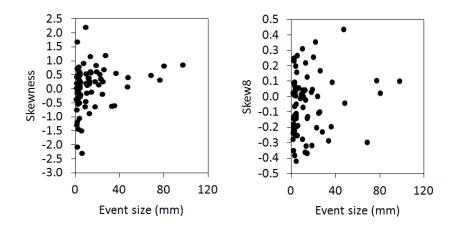


Figure 7. Skewness and octile skew (skew8) as a function of event size

Stemflow: Whole study period and event scales

Stemflow (mm) was divided by corresponding bulk rainfall and multiplied by 100 to give relative stemflow. The relative stemflow for the whole studied ranged from 0.9 to 1.2 % and showed a normal distribution but negatively skewed (Table 3). This result was explained by one tree (tree 7 in Table I) which gave stemflow values systematically lower than the others.

Table 3. Statistics for the cumulative values of stemflow. All stemflow values are expressed as percentage of bulk rainfall. SD is the standard deviation value and CV is the coefficient of variation. The skewness in the throughfall distribution was tested by the Fisher coefficient test and the Kolmogorov-Smirnov test was used to test the normality.

Number of trees sampled (N)	8
Bulk rainfall (mm)	1090
Mean stemflow (%)	1.1
Median stemflow (%)	1.1
Maximum stemflow (%)	1.2
Minimum stemflow (%)	0.9
stemflow SD(%)	0.1
Stemflow CV (%)	9.6
Skewness (Fisher coef.)	-1.9
p-value (Kolmogorov-Smirnov test)	0.498

Regarding the event scale, stemflow also followed a linear relationship with bulk rainfall (y= 0.0132x, R^2 = 0.76, n=83). A exponential decrease of the CV with event size was also observed, but in this case the rainfall size which pooled the stemflow data was 10 mm (30.4±17.8 and 21.3±11.7 for 0-10 mm and >10 mm events, respectively). On the

other hand, the range of stemflow changed with event size: from 0 to 3 % for events lower than 25 mm, and from 0.4 to 1.5 % for events higher than 25 mm.

Relationships between throughfall and stemflow with canopy characteristics

Relative throughfall was firstly compared between the 2012 and 2013 leafed periods and secondly between the whole leafless period and the leafed one. Throughfall in the leafed period of 2013 was lower than that in 2012 (92 versus 96 %) but the t-student analyses indicated not significantly differences, despite the high difference on canopy cover between the two studied years. On the other hand, non significant differences were neither found when comparing the whole leafless period versus the leafed one.

Mean throughfall did not correlate with mean canopy cover when all the 83 events were considered (analysis not shown). However, when the 2013 leafed period was only considered, as the one with higher canopy cover, and the rainfall events were classified by size, positive linear regressions between throughfall and canopy cover appeared for several events larger than 5 mm (Figure 8). Note that, though not significant, the trend for events smaller than 5 mm was of decreasing throughfall as canopy cover increased.

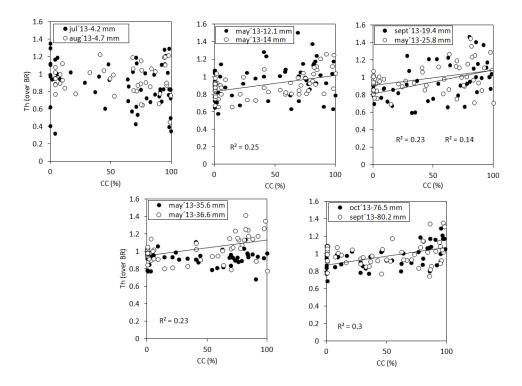


Figure 8. Examples of the relationship between throughfall (over bulk rainfall, BR) and canopy cover (%), with 2 events per class (see Figure 5 for event classes). All the events selected were from the 2013leafed period. R² is only shown when significant.

On the other hand, t-students analyses indicated that relative stemflow was not significantly different between the 2012 and 2013 leafed periods (0.5 \pm 0.1% versus 0.9 \pm 0.1%), while significant differences were observed between the whole leafed and leafless periods (0.7 \pm 0.1% versus 1.5 \pm 0.2%).

Optimum number of tipping buckets for measuring throughfall

Since time frequency of throughfall measurements can vary according to the objectives of the study, we analyzed the optimum number of tipping buckets to measure throughfall for the whole study period and event scales. In the first case, the 58 relative throughfall data (as % of bulk rainfall) were considered for the both tested methods, i.e., the Monte Carlo method, MC, and the pre-setting error method, PE. For the event scale, we first split the throughfall data into two groups: events < 30mm and events > 30 mm. For the events < 30mm, 10 events normally distributed were randomly selected, while all events normally distributed were selected for events > 30 mm (7 events).

The results for the whole study period scale were very similar between the methods tested, with 9 and 10 tipping buckets for MC and PE, respectively (Figure 9). However, the results at event scale were more contrasted. Optimum numbers calculated with PE behaved as expected from the exponential relationship between throughfall variability (expressed by CV) and event size, with a reduction of magnitude and variability with event size. In contrast, MC gave similar results for both events groups, and also no clear solutions were found in a high proportion of the events selected (9 out of 17 events) (see Figure 3 for more details about the calculation procedure).

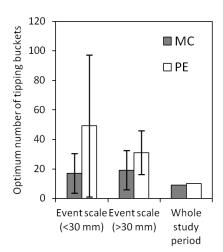


Figure 9. Optimum numbers of tipping buckets for measuring throughfall at event scale (means and standard deviations) and at the whole study period scale (means). MC and PE were the two methods tested (see text for details).

Discussion

We conducted a study in order to primarily assess how rainfall was partitioned into throughfall, stemflow and interception in a wild cherry tree plantation under Mediterranean conditions. Most of tree plantation types need irrigation for a proper development in Mediterranean areas, but the net rainfall (=throughfall + stemflow) which is considered for the irrigation calculation is normally only available for common cultivars such as olive (Gómez el at., 2001), apple (De Miranda and Battler, 1986) or vineyard (Brecciaroli et al., 2012). In our study, throughfall for the whole study period showed a mean value of $96 \pm 7 \%$ of bulk rainfall, and a range from 70 to 114 % for the event scale. For its part, stemflow showed a mean value of 1.1 \pm 0.1 % and it ranged from 0 to 3 %. Therefore, interception loss for the whole study period accounted for 3 % of bulk rainfall. This interception loss value is much lower than the one expected for this type of plantation, when compared either to deciduous forests or fruit tree plantations growing under similar climatic conditions, or plantations of evergreen trees with similar forest structure (Brecciaroli et al., 2012; Deguchi et al., 2006; De Miranda and Battler; 1986, Fares et al., 2008; Gómez el at., 2001; Jackson, 2000; Levia and Frost, 2006; Llorens and Domingo 2007; Schnabel and Mateos, 2001; Muzylo et al., 2012; Obreza and Pitts, 2002). In contrast, stemflow is in the range presented in review works (Llorens and Domingo 2007; Levia and Frost, 2003).

Rainfall partitioning under vegetated ecosystems is affected by climate and canopy structural characteristics (Crockford and Richardson, 2000). In conditions of high evaporative demand, interception loss can account for until values of 30 % of gross rainfall (Llorens and Domingo, 2007) and rainfall events of low magnitude are usually the ones with the highest rainfall interception rates (Llorens et al., 1997). Our rainfall event distributions (frequency and size) were very similar between the two studied leafed periods and characteristics of Mediterranean climate, with several events of low rainfall size, several with intermediate size and a few events of high size but which greatly contributed to total (Figure 5). On the other hand, canopy cover significantly increased from 18 to 41 % from 2012 to 2013 growing seasons. While stemflow was significantly affected by canopy cover when comparing between the leafed and leafless periods (0.7±0.1 % versus 1.5±0.2 %, respectively), the throughfall was not in any of the comparisons, and thus the throughfall reduction from 96 to 92 % (from 2012 to 2013 growing period) was not affected by the canopy cover increase. In addition,

canopy cover in 2013 was inversely related to throughfall (Figure 8), contrary to general expected trend of throughfall decrease with canopy cover increase (e.g., Molina and Del Campo, 2012; Zimmerman et al., 2009). However, other authors have observed the inconsistent pattern of higher throughfall amounts near stems than at crown edges (Keim et al., 2005; Ford and Deans, 1978) or negligible effects of canopy structure in the spatial variations of throughfall (Shinohara et al., 2010). In a recent study, Fan et al. (2015) found a strong positive power correlation between relative throughfall and canopy cover in a subtropical pine plantation and they attributed this fact to the greater effect of meteorological variables than that of canopy structure on the spatial variability of throughfall. Also these authors found the lowest throughfall values in the midway between tree rows. The authors supported their explanation on the fact that the highest throughfall occurred in the windward size of tree trunks, i.e. higher throughfall in the east side of trunk respect to the west side (tree row), in line with other throughfall results at tree level (e.g. David et al., 2006). In this respect, we further studied the effects of wind velocity on the statistics of the throughfall distributions for events with size higher than 20 mm as they showed similar CV unaffected by event size: no clear correlations were observed in any of the dispersion plots between wind velocity and the rainfall distribution statistics (example in Figure 10). Fan et al. (2015) placed the throughfall devices in a systematically way respect to tree trunks (5 at east side of trunks, 5 at west side of trunks and 5 in the midway between tree rows), while our 58 tipping buckets were randomly placed in the 128 m² experimental plot, most of them at least at 2 m from tree trunks (Figure 2). In addition, although we did not prune the sample trees during the study period, they had already their crowns affected by two previous pruning interventions. Therefore, the explanation of Fan et al. (2015) may not be satisfactory for our results.

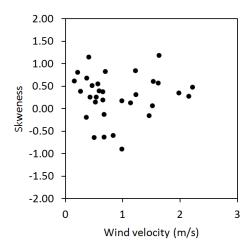


Figure 10. Skewness versus wind velocity (m/s) for event size >20 mm (n=32)

On the other hand, a high numbers of throughfall concentration points, i.e. dripping points, were observed as indicated by the skewness coefficients, especially in the events of size higher than 40 mm (Figure 7). This fact produced that throughfall at the whole period scale was also rightly skewed. In addition, 35 % of the events were not normally distributed. Such skewed throughfall frequency distributions are usually observed in tropical forest with dripping canopy structures, e.g. leaf or branches with inclined angles, which promote the concentration of throughfall (Lloyd and Marques, 1988; Zimmermam et al., 2009); consequently, other points at soil surface should be drier (Howerda et al., 2006; Zimmermam et al., 2009). Thus, dripping and drying are expected to be counterbalanced in vegetated ecosystems. However, this compensation seems to be explained by a systematic arrangement of the throughfall devices in the case of low tree density or isolated trees, or by a random one whether the canopy cover complexity can be assumed to show a homogeneous spatial distribution. Therefore, our experimental design seemed to be optimum to obtain a general value for rainfall partitioning fluxes at stand scale but not to relate them with neither canopy structure or wind velocity and direction. In addition, we hypothesize that wild cherry trees show particular leaf characteristics of the studied species which lead to concentrate throughfall and thus to retain low rainfall amount (low leaf storage capacity): inclined angle and high leaf water repellency.

Taking into account our high spatial resolution for measuring throughfall of 0.45 tipping buckets per m², we also calculated using two different methods the number optimum of tipping buckets to measure throughfall for two different time scales, since the exact number of gauges necessary to accurately estimate throughfall volume varies according

to the objectives and required precision of the specific study (Levia and Frost, 2006). For the whole study period scale, both methods gave similar results, and 10 tipping buckets were found to be suitable to give a robust estimation of the mean throughfall. For the event scale, while with the PE method (Kimmins, 10973) an increase in the numbers of tipping buckets was observed when CV increased (means from 30 to 48 to events>30 mm and events<30 mm, respectively), it was not the case for the MC method (Rodrigo and Avila, 2001), with about 20 tipping buckets regardless the event size (Figure 8). The result observed with the MC method is close to the one recommended in other studies (n=30, in Levia and Frost, 2006) and higher than 8 tipping buckets, as the one recommended for weekly measuring throughfall in a bamboo forest for different rainfall sizes (Shinohara et al., 2010). In contrast, our values contrasted to the one presented by Holwerda et al. (2006) of 100 devices in tropical rain forest. In this case, the authors recommended using roving strategies to reduce throughfall variability, as other authors have already pointed out (e.g., Asdak et al., 1998; Ritter and Regalado, 2010). Therefore, we can conclude that 48 tipping buckets can be considered as suitable for measuring throughfall at event scale in the studied plantation.

Conclusions

Interception loss has been showed to only represent a small part of rainfall in our young wild cherry plantation oriented to timber production. Therefore, this water flux can be considered negligible for noble plantations of similar growing in semiarid conditions, and consequently specific factors for accounting this water loss to atmosphere in the irrigation calculations can be omitted. High proportion of dripping points have observed in places with high canopy cover, and contrary to the expected trend, throughfall and canopy cover have been positively correlated. We hypothesize that our randomly distributed experimental design and particular leaf characteristics of the studied species may explained this result, so more studies about canopy characteristics (such as leaf storage capacity and leaf water repellency) of the studied species and with other type of throughfall devices arrangement could clarify these questions. Finally, 10 or 48 tipping buckets has been showed to be sufficient to estimate the mean throughfall for the global and the event scale, respectively.

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CHAPTER 3

Transpiration response to evaporative demand, soil water deficit and branch pruning of a wild cherry tree plantation under Mediterranean climate

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Chapter 3

Transpiration response to evaporative demand, soil water deficit and branch pruning of a wild cherry tree plantation under Mediterranean climate

Abstract

The two main aims of this study were a) to determine the stand transpiration of a wild cherry plantation for timber production (clone Salamanca 4, Sa-4) under Mediterranean climate conditions and b) to assess the effects of environment (evaporative demand and soil water deficit) and tree pruning on stand transpiration. To this end, transpiration was measured during the 2012 and 2013 growing seasons by heat pulse sap flow sensors together with climate, soil and forest structure variables; and branches were pruned in June 2013, with about a third of total crown biomass removed. Also, two soil water availability treatment plots were established in the study: in one plot, trees received drip irrigation (I), whereas in the other plot trees grew under rainfed conditions (NI). Stand transpiration dynamics differed between the plots and revealed physiological behaviour that is thought to be closely related to stomatal closure differences. Total stand transpiration accounted for 127 and 60 mm or 9.6 and 4.5% of ET for I and NI plots, respectively. On the other hand, branch pruning reduced by approximately 30% the stand transpiration of both plots and had a positive effect on water productivity (basal area over stand transpiration, cm² litres⁻¹) for both plots (from 2012 to 2013): from 2.1 to 2.4 and from 2.9 to 3.7 cm² litres⁻¹ for I and NI, respectively. Finally, stand transpiration was simply modelled as a function of reference evapotranspiration and soil water content. The modelling scheme followed highlighted the differences in physiological behaviour between the trees receiving drip irrigation and those growing under rainfed conditions.

Keywords: Agroforestry systems, *Prunus avium* L., timber, noble hardwood, sap flux density, pruning

Introduction and objectives

Agriculture in Mediterranean areas suffers natural water scarcity, a condition that will increase in the future together with the greater unpredictability of extreme climatic events that may affect crop production. Diversification of land use is a practice that may allow farmers to adapt better economically to these new conditions, given that plant species differ in their environmental responses. In Europe, forest plantations for wood production have been widely promoted in recent decades, mainly to confront desertification and rural abandonment (e.g. EU Regulation 2080/92), especially of species with rapid growth rates in short rotation plantations. Forest plantations of noble hardwoods for high-quality timber have also grown, in response to the imbalance between exports and imports (Ducci et al., 2013), though to a lesser extent because of their greater rotation lengths and, in consequence, longer periods for obtaining economical benefits.

In the last two decades, wild cherry (*Prunus avium* L.) has been a species widely used in reforestation programmes using noble hardwood (Ducci et al., 2013; Montero et al., 2003). Recognition of wild cherry's role in improving biodiversity and CO₂ capture is increasing (Loewe et al., 2013). In consequence, studies aiming to select and improve wild cherry trees to give them the best growth rates, forms and wood properties have increased in response to growers' demands (Curnel et al., 2003; Diaz et al., 2007; Ducci et al., 2013; Martinsson, 2001; Nocetti et al., 2010). Moreover, special attention has been paid to the effect that mixing wild cherry trees with other species has on tree diameter growth, either in mixed plantations with forest species (Kerr, 2004; Loewe et al., 2013) or in agroforestry systems for tree crops (Campbell et al., 1994; Chifflot et al., 2006; Dupraz et al., 1995).

As a consequence of these reforestation programmes, wild cherry plantations have also been established in Mediterranean areas, although these areas are outside the natural range of this species and irrigation is normally required to face summer drought (Ducci et al., 2013). Most of these irrigated plantations are nowadays managed in line with selvicultural guidelines mainly concerned with the effects of pruning and thinning interventions on diameter growth (e.g. Cisneros et al., 2006). Little attention has been paid to alternative management practices, such as zero tillage, irrigation deficit or zero irrigation (Molina et al., 2016; Ripoll Morales et al., 2013). Therefore,

information about transpiration or water use of wild cherry plantations and its relationships with environmental conditions is rather limited (Cabibel and Isbérie, 1997; Lambs et al., 2008), even though this variable is known to be essential to proper irrigation and thus to the evaluation of new management strategies, especially in areas with limiting conditions, such as the Mediterranean (Fereres and Soriano, 2007). In this sense, branch pruning is a common practice in this type of plantation to obtain stems free of branches and so increase timber value (Loewe et al., 2013). However, the effect that this practice may have on water use is unknown, as is the extent to which water productivity or the water transpired per obtained yield might be affected.

The present study specifically addresses stand transpiration in an experimental plantation that used the clone (Salamanca 4, Sa-4) most planted in Mediterranean areas of Spain. Thus, its main objectives are a) to determine the stand transpiration of wild cherry trees planted in a Mediterranean area, b) to assess the effects of environmental conditions (evaporative demand and soil water deficit) and tree pruning on stand transpiration, and c) to model stand transpiration as a function of reference evapotranspiration and soil water content.

Materials and methods

Experimental site

The experimental site is located near Barcelona, at IRTA's experimental station in Caldes de Montbui ($41^{\circ}36'47''$ N, $2^{\circ}10'11''$ E) at a height of 170 m asl. The climate is Mediterranean with a mean annual (1991-2010) temperature of $14.4 \pm 0.2^{\circ}$ C, reference evapotranspiration of 846.8 ± 23.3 mm and rainfall of 599.4 ± 33.4 mm, obtained from a weather station on the site (Servei Meteorològic de Catalunya, UTM X430803, UTM XY4607309).

The wild cherry tree plantation was established in 2006 at a tree density of 625 trees ha⁻¹ and with spacing between the trees and between the rows of 4 m (16 m² tree⁻¹). In December 2010, the average tree height was 4.7±0.1 m and the mean diameter at breast height was 5.7±0.1 cm. In line with timber-oriented practices to obtain trunks free of branches, trees are pruned every two to three years during the growing season, with approximately one third of the total crown volume removed from the

lowest part of crowns. In June 2013, average total dry biomass removed in the pruning was 5.7 ± 2.8 kg tree⁻¹.

The plantation was established on an alluvial terrace with two clearly different zones separated by a transition zone with mixed materials from both. One zone was characterized by its sandy-loam texture and 80% gravel content; and the other one, with sandy-clay-loam texture and much less gravel (about 10%). The study focused on trees planted in the latter zone, with higher available soil water (112 mm to a soil depth of 100 cm) and thus with better tree growth rates (Molina et al., 2016).

Two treatment plots were established in the study, according to the soil water availability for the trees. In one plot, trees were submitted to drip irrigation (I), while in the other plot trees grew under rainfed conditions (NI). Drip irrigation was used during the growing season with 4 emitters ($16 \text{ I h}^{-1} \text{ tree}^{-1}$) located 25 and 50 cm to the north and south of the trees. Daily doses (applied from Monday to Friday) were calculated at the beginning of each week as a function of the product of the weekly sums of reference evapotranspiration (ET_0) multiplied by a crop coefficient (Kc), which had values from 0.43 to 0.6 depending on the month, minus the rainfall (R) of the previous week ($\text{I= Kc'} \cdot \text{ET}_0\text{-R}$). Irrigation was not applied when ET_0 was lower than R.

Sap flow measurements

The stand transpiration in the I and NI plots was calculated from sap flow measurements by the heat ratio method (HRM) (Burgess et al., 2001) in 4 and 3 trees, respectively, in 2012. In 2013, two more trees were measured in each treatment (6 and 5 trees, respectively). The biometric characteristics of the measured trees are given in Table I.

Table 1. Tree biometric characteristics (maximum values in growing seasons) of the trees selected for sap flow measurements, in the NI and I plots. DBH: diameter at breast height, H: height and As: sapwood area (see text for details of its definition).

			2012			2013	
Treatme	Sample	DBH	H	As	DBH	H	As
nt	tree	(cm)	(m)	(cm ²)	(cm)	(m)	(cm^2)
	I	9.5	7.1	67.4	11.3	7.6	93.8
I	2	9.6	6.6	68.8	11.1	7.2	90.7
1	3	10.2	6.7	77.2	11.7	7.4	100.2
1	4	8.8	5.8	58.3	10.4	6.6	80. I
NI	5	7.8	6.4	46.3	8.8	7.1	58.3
NI	6	9.3	6.9	64.8	10.5	7.1	81.6
NI	7	9	5.9	60.8	9.9	6.3	72.9
I	8	8.6	7.1	55.8	11.7	7.7	100.2
1	9	8.2	6.3	51.0	10.1	7.1	75.8
NI	10	8.1	6.4	49.8	9.9	7.1	72.9
NI	П	8	6.7	48.6	10.2	7.3	77.2

Sap flow sensors (ICT International, Australia) were programmed to measure every 100 seconds and to average the data every 30 minutes. One sensor was installed at each sample tree, 1.3 m high and on the east side of the trunk. The velocity of the heat pulse emitted by the heater needle was measured by thermocouples placed 1.25 and 2.75 cm from the cambium, and 5 cm up and down from the heater (Burgess et al., 2001). Thermal diffusivity and wood moisture fraction values were calculated from several tree ring cores, following Kravka et al. (1999). To correct heat pulse velocities when necessary, the alignment of the probes was checked yearly by testing the difference between the measurements and the baseline, corresponding to zero sap flow measured during the first leafless week in December. A possible underestimation caused by the probe-induced effects of wounding (Barret et al., 1995) was assessed from wound width, following Swanson and Whitfield (1981) and using the results for plum trees (*Prunus domestica*) from Fernandez et al. (2006), since there is no wound width data available for wild cherry.

As azimuthal and radial variations of sap flux density may lead to major biases in tree transpiration calculations (e.g. Kume et al., 2012), two independent experiments were carried out during 2014 (Molina et al., 2015). Briefly, the azimuthal variation was analysed by measuring sap flux density with 4 HRM sensors located at the four compass points of 6 trees. The radial variation was tested by measuring sap flux

density with Compensation Heat Pulse Method (CHPM) sensors (Green et al., 2003) placed 1.0, 1.9 and 3.0 cm from the cambium of 14 trees (east side). On the one hand, the azimuthal variation results showed that the ratio between the sap flux density on the east side and the average from the four sides ranged from 0.73 to 1.46, with no clear systematic pattern observed between trees. Thus, in this study no further correction was made to the readings taken on the east sides. On the other hand, we assumed that HRM sensors with measurements at two sapwood depths were sufficient to characterize the radial profile of sap flux density in our small trees (inner measurement covering a mean 72.4% of total sapwood in 2013), as direct comparisons between HRM and CHPM measurements in particular trees were not possible during the experiment.

Sapwood increment during the growing season was also taken into account because the wild cherry trees were young and they can be considered a fast-growing species at this age (Guan et al., 2012). Stem diameter of the sample trees was measured monthly during the growing seasons. It was assumed that sapwood depth grew proportionally to tree diameter in the growing season, as in other young fast-growing trees (Guan et al., 2012). To estimate the sapwood area from stem diameters, thickness of sapwood and bark was visually identified in several tree cores during the experiment (Nadezhdina et al., 2002). Sapwood area (As, cm²) was calculated monthly as a function of diameter at breast height (As = $0.93 \times DBH^{1.9028}$, R²=0.98, N=20). Moreover, based on our field observations, and following Beauchamp et al. (2013), as the sensors remained in a fixed location during the growing season, trees grew around the sensors. For each month, the sapwood area was divided into two concentric bands (outer and inner band), delimited by the mid-point between the thermocouple location (Hatton et al., 1990; Bleby et al., 2004). The sapwood increment during the month was assigned to the outer thermocouples, i.e. the cross-section area of the outer band was equal to the increment band due to growth of trees around the sensors during the month plus the previous outer band. Sap flux for each tree (litres tree-1) was thus calculated by multiplying the outer band by the sap flux density measured in the outer thermocouples; and the inner band, by the inner sap flux density measured in the inner thermocouples; and then adding both. Finally, stand transpiration (mm day⁻¹) was calculated from the tree sap flux means for each treatment plot and the tree density of the plantation (625 tree ha⁻¹).

Canopy cover (CC) in the growing seasons was calculated about every twenty days in the early morning by means of sky-oriented photographs taken with an 18 mm-lens Canon EOS 400D digital camera mounted on a tripod 50 cm high and horizontally levelled. Two sky-oriented photographs were taken of each tree at two fixed positions (north and south) 50 cm apart from the tree trunk. Moreover, in a 128 m² rectangular plot, 58 sky-oriented photographs per day were also taken at random positions on the same sampling days. This information was used to scale canopy cover data from tree to stand scale by using a linear regression ($CC_{stand\ scale} = CC_{tree\ scale} \cdot 0.38$, $R^2 = 0.95$). This relationship was used to rescale all the tree scale photographs and CC at stand scale was linearly interpolated between measured dates to obtain daily values. The images were analysed with standard software based on HUE colour data (Casadesus et al., 2007).

In 2013, the Leaf Area Index (LAI) was also calculated in the sample trees, using two leaf cylinder collectors (each with 0.096 m^2 surface) placed at a height of I m and 50 cm north and south of the tree trunk. Leaves were collected every 15 days and weighed after drying at 105° C. Specific leaf area (SLA) was calculated from a sampling of 10 leaves of 12 trees during the June 2013 pruning (15.06 \pm 4.02 m^2 kg $^{-1}$), using the Winfolia software (Regent Instruments Inc, Quebec, Canada). The LAI per tree (m^2 m^{-1}) was calculated as the accumulated dry mass, converted into surface, during the whole year from the two positions divided by the collecting area.

Meteorology and soil water content

Meteorological conditions were measured at 2 m height at a standard weather station located in an open area 50 m away from the plantation. Rainfall (ECRN-100, Decagon Devices, Pullman, USA), air temperature and humidity (RH-T sensor, Decagon Devices, Pullman, USA), wind velocity and wind direction (Davis cup anemometer, Decagon Devices, Pullman, USA) and solar radiation (PYR Solar radiation sensor, Decagon Devices, Pullman, USA) were measured every 60 seconds and averaged every 30 minutes (Em-50, Decagon Devices, Pullman, USA). The data collected were systematically verified by comparison with data from an official meteorological station (Servei Meteorològic de Catalunya, UTM X430803, UTM Y4607309) located about 500 m from the plantation.

Soil water content was measured under the crown projection of sample trees. At the mid-point between the southern drip emitter and the tree trunk, and at the same position for the non-irrigated trees (NI), 3 10 cm-long probes (10-HS, Decagon Devices, Pullman, USA) were inserted vertically in the trees, with their centres at depths of 25, 50 and 100 cm. 60-second measurements were averaged and stored every 30 minutes (Em-5, Decagon Devices, Pullman, USA). To avoid bad contact between the sensors and the soil matrix, gravel was removed before installation. Sensor readings were systematically corrected by taking into account the volumetric percentage of gravel estimated for each point measurement (Molina et al., 2016).

In 2013, two soil pits were excavated to a depth of 2 m ($Im \times Im \times 2$ m) to study root distribution in the plantation. 85% of the total fine roots (<2mm of diameter) were found in the first 75 cm of soil profile. Thus, only the probes placed at 25 and 50 cm depth were used to calculate the relative extractable water by roots (REW), following Granier et al. (2000):

$$REW = (W - W_m) / (W_{FC} - W_m)$$
 (1)

where W is the average soil water content from the probes, W_m is the minimum soil water content during the study period and W_{FC} is the soil water content at field capacity. REW values higher than I were made equal to I (Granier et al., 2000).

Analysis of the response of stand transpiration to atmospheric evaporative demand, stand structure and soil water content

The evaporative demand of the atmosphere was expressed as Penman-Monteith FAO reference evapotranspiration (ET $_0$), following Allen et al. (2006).

Stand transpiration, for the irrigated and non-irrigated plots, was first divided by their canopy cover fraction values at stand level to obtain normalized values (T_{norm1} and T_{norm2}).

The normalized stand transpiration not affected by soil water deficit during the growing season, i.e. the transpiration of the irrigated plot (Tnorm_I), was calculated, following Ewers et al. (2008), by:

$$Tnorm_{l} = a \cdot (1 - exp^{-b \cdot ET0})$$
 (2)

where a and b are fitting parameters. The fit of the parameters was done for each growing season separately, after it was seen that stand transpiration was much higher at the beginning of the 2013 growing season.

The upper envelope of the relationship between Tnorm₁ and ET₀ was used to predict stand transpiration as a function of ET₀ under optimal conditions, representing the maximum stand transpiration (Tnorm_{max}) without environmental limitations:

$$Tnorm_{max} = a_{max} \cdot (I - exp^{-bmax} ETO)$$
 (3)

This upper boundary was calculated by the quantile regression technique, a satisfactory methodology used for modelling transpiration in other tree species and conditions (e.g. Poyatos et al., 2005; Petzold et al., 2011). We fitted parameters a_{max} and b_{max} to the upper 95% quantile to calculate the maximum stand transpiration in the 2012 and 2013 growing seasons.

The effect on stand transpiration of soil water content depletion was assessed as described by Granier et al. (2000):

$$f(REW) = Tnorm_{NI} / Tnorm_{max} = [p_1 + p_2 \cdot REW - [(p_1 + p_2 \cdot REW)^2 - 2.8 \cdot p_1 \cdot p_2 \cdot REW]^{1/2}] \cdot 0.714$$
(4)

where p_1 and p_2 are fitting parameters.

To fit the model, data from the 2012 growing season were selected to avoid any interference promoted by the branch pruning carried out in June in the stand transpiration. In addition, days with average vapour pressure deficit higher than 0.6 Kpa were selected in order to reduce probable errors in measurements of sap flow and vapour pressure deficit in such conditions of low evaporative demand (Ewers and Oren, 2000).

The stand transpiration models for the trees growing under rainfed conditions (NI) were thus obtained as:

$$T = Tnorm_{max} \cdot f (REW) \cdot CC f (sapwood growth)$$
 (5)

where CC is canopy cover fraction values at stand level, f (sapwood growth) is the ratio that accounts for the difference between the conducting sapwood areas of plots, i.e. the sapwood

for the trees growing under rainfed conditions over the sapwood for the drip-irrigated trees at the beginning of each growing season.

Quantile regression analyses were done in R (v.3.2.2, R Development Core Team, Vienna, Austria) and the parameters in the non-linear regressions were calculated in SPP (v.15.0, SPSS Incl., Chicago, USA).

Results

Environmental conditions, canopy cover dynamics and tree transpiration

Both growing seasons were characterized by dry summers (June-August) with low rainfall inputs (63 and 43 mm in 2012 and 2013, respectively, or 14 and 7% of annual rainfall) and high values for reference evapotranspiration (with mean daily values of 5 and 4 mm day⁻¹ in 2012 and 2013, respectively) (Fig. 1a). The REW dynamics in the NI plot showed strong soil water depletions in both growing seasons, although these were lower in the 2013 growing season because of a higher rainfall recharge in early spring (171 mm and 91 mm of rainfall in April and May, respectively). In contrast, REW values in the I plot were always higher than 0.4 except for a short period at the beginning of 2012 (Fig. 1b), which is the critical REW threshold at which tree transpiration is expected to decrease (Granier et al., 2000).

Canopy cover development started in mid-April in both years and reached its maximum values in May and June (Fig. 1c). In the 2012 growing season, canopy cover was maintained until October, while the pruning in June 2013 reduced the tree crowns by 56 and 63% for I and NI plots, respectively. The differences in canopy cover between plots remained almost constant during the entire study period (Fig. 1c). In addition, LAI calculated under tree crowns by the collection of fallen leaves during the 2013 growing season gave values of 2.82 and 1.42 m² m⁻² for I and NI plots, respectively.

Maximum daily stand transpiration was 8.5 and 5.4 litres tree "day" for I and NI plots in 2012; and 12.8 and 6.7 litres tree "day" in 2013 (Fig. Id). Total stand transpiration during the study period accounted for 127 and 60 mm or 9.6 and 4.5% of ET_0 for I and NI plots, respectively. Furthermore, pruning at the end of June 2013 caused major decreases in stand transpiration. The comparison between the 15-day periods before and after pruning, both with very similar climate conditions (means of ET_0 , 4.10 and

4.11 mm day⁻¹), showed a reduction of approximately 30% in both plots: from 10.26 to 6.95 litres day⁻¹ and from 5.04 to 3.57 litres day⁻¹ for I and NI, respectively.

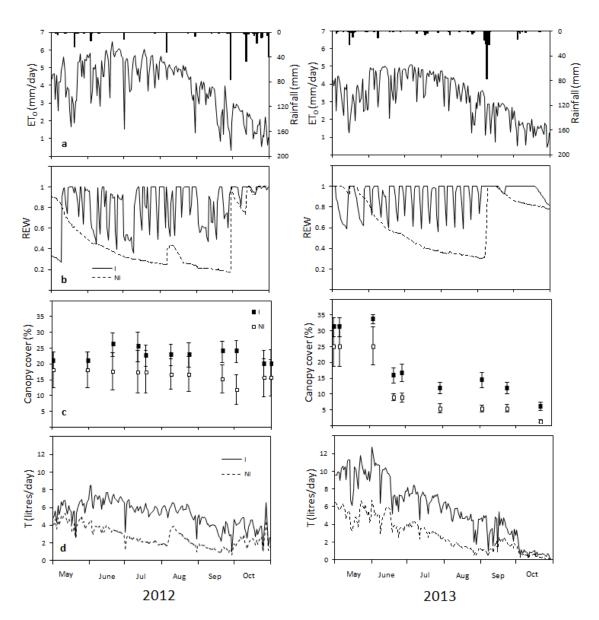


Figure 1. Time series of (a) reference evapotranspiration (ET₀) and rainfall, (b) relative extractable soil water (REW), (c) canopy cover at stand scale and (d) tree transpiration (T) in the 2012 and 2013 growing seasons. It drip irrigated plot, NI: non-irrigated plot. The canopy cover values are means and standard deviations for each sampled day. Mean values are shown for tree transpiration and REW.

Modelling of stand transpiration

Daily values of the normalized stand transpiration in the irrigated plot (Tnorm₁) were clearly exponentially related to reference evapotranspiration in both growing seasons. In consequence, the fitted models were able to predict stand transpiration (Figure 2, Table 2). In 2012, the plateau of maximum values on Tnorm₁ was found to be close to 2, while it was close to 2.7 in 2013.

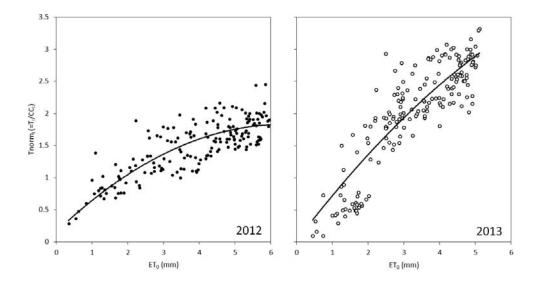


Figure 2. Normalized stand transpiration of the irrigated plot in relation to reference evapotranspiration (Table 2 gives the models' fitting parameters).

In order to obtain a single model for the irrigated plot as a function of ET_0 , we further applied a correction factor to account for the sapwood difference between the two growing seasons, as a possible explanation of the stand transpiration difference found. Thus, the ratio of the sapwood in early spring 2013 over the sapwood in early spring 2012 (f_{As}) was applied to the 2012 model when predicting 2013 data. As this new model gave similar stand transpiration predictions to the original one, it allowed us to use the same set of parameters for the entire study period (Table 2).

Table 2. Parameters of the stand transpiration models under irrigation conditions (I plot) as a function of reference evapotranspiration and of the observed and modelled stand transpiration for the 2012 and 2013 growing seasons, and for the entire study period. Parameters of the single model accounted for the sapwood difference between the two growing seasons through applying a correction factor to 2013 data (f_{As}; see text). Confidence intervals of the parameters are shown in brackets.

				Total stand transpiration (mm)	
Model	a 2.1	b 0.36	f_{As}	Observed	Modelled
2012	(1.91,2.27) 6.60	(0.28,0.42) 0.12	-	60.64	60.71
2013	(3.73,9.47)	(0.05,0.18) 0.36	-	66.38	65.70
Single	(1.91,2.27)	(0.28,0.42)	1.4	127.02	122.07

The next step in the modelling of the stand transpiration of wild cherry trees growing under rainfed conditions was to obtain the maximum stand transpiration values (Equation 3), i.e. the stand transpiration without environmental limitations derived from I plot data. Figure 3 shows the normalized stand transpiration of the irrigated and rainfed plots expressed over the maximum stand transpiration, in order to point out their different patterns from the expected maximum. It can be noted from the Figure that the trees growing under irrigated conditions transpired under optimum conditions at the beginning of both growing seasons. In 2012, the transpiration of trees growing under irrigated conditions steadily decreased from mid-June to the end of the growing season, while in 2013 these decreasing patterns were more intermittent, with periods of maximum transpiration between transpiration decreases. These results from the irrigated plot seem to indicate that, although soil water content was not a limiting factor, our wild cherry trees showed steady stomata closure due to physiological adjustments in the transpiration and photosynthesis processes, but these differed between the 2012 and 2103 growing seasons, probably due to the 2013 pruning.

On the other hand, the stand transpiration dynamics of the trees growing under rainfed conditions showed the clear effect of soil water content depletion and how these dynamics changed with respect to maximum stand transpiration. At the beginning of 2012, trees growing under rainfed conditions transpired at a ratio very close to that of the maximum time series, while this was not the case at the beginning of 2013 (Figure 3).

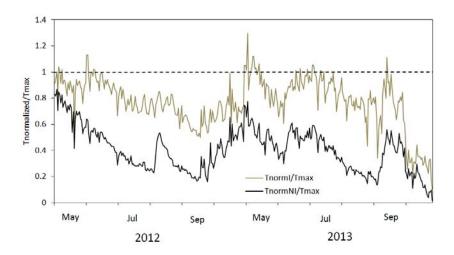


Figure 3. Normalized stand transpiration for the I and NI plots (dividing by canopy cover fractions) over the maximum stand transpiration (Tmax). The maximum stand transpiration was calculated from I data by using Equation 3.

To account for the effect of soil water depletion, we fitted the parameters of Equation 4 in the relationship between REW and the ratio of actual stand transpiration to the maximum transpiration (Figure 4). Thus, f(REW) was derived (p_1 =1.38 (1.19-1.58), p_2 =1.23 (1.17-1.28), R^2 =0.89; 95% confidence intervals). Figure 4 shows that stand transpiration of the NI plot was linearly related with REW for the entire REW range, although different positive slopes were found, since REW of about 0.6 was the threshold for data pooling: changes in REW from 0.2 to 0.6 affected stand transpiration in a stronger way than REW from 0.6 to 1.

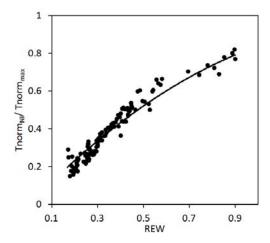


Figure 4. Ratio of normalized stand transpiration for the trees growing under rainfed conditions (NI) over the maximum stand transpiration (calculated with the 95% quantile technique) in relation to extractable water (REW); data for 2012. The line indicates the polynomial model fitted to data (Equation 4); see text for details about fitted parameters. Note that the ratio never reached I, as indicated in Figure 3, i.e. trees growing under rainfed conditions did transpire with environmental limitations during the whole study period.

Once f(REW) was obtained, we modelled the stand transpiration for the rainfed plot (NI) as a function of the dynamics of maximum stand transpiration, soil water content and canopy cover. In addition, a correction factor which accounted for the sapwood differences between the NI and I plots was applied (Equation 5).

Figure 5 shows the time series of observed *versus* modelled data. In 2012, the stand transpiration was well predicted for most of the growing season (33.0 and 31.1 mm for modelled and observed data, respectively), except for October, for which there was an overestimate of 39% (7.2 and 4.3 mm for modelled and observed data, respectively). In 2013, there was a general overestimate of 33%, with the maximum differences between the modelled and observed values at the end of the growing season. In addition, a model overestimate of less importance occurred at the beginning of the

growing season (20.0 mm and 14.01 mm for modelled and observed data, respectively). This means that stand transpiration was much better predicted after the branch pruning (in July and August), with total accumulated values of 9.33 and 10.0 mm for the modelled and the observed data, respectively.

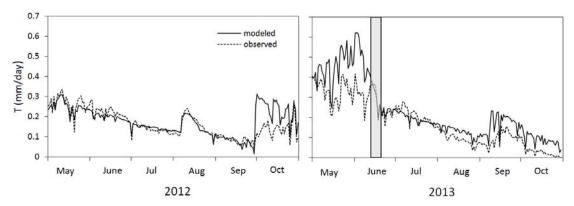


Figure 5. Time series of observed and modelled (Equation 5) stand transpiration of the NI plot in the 2012 and 2013 growing seasons. The 2013 tree pruning period is indicated by the filled box.

Discussion

Wild cherry stand transpiration under irrigation and rainfed conditions

Uncertainties about the scaling up from sap flux density measurements to stand transpiration have been evaluated elsewhere (e.g. Hatton et al., 1990; Kume et al., 2012). For the species studied here, Cabibel and Isberie (1997) presented the first results about azimuthal variation in wild cherry tree sap flux density. They also explained the specific variation caused by drip irrigation. Since we studied previously (Molina et al., 2015) the radial and azimuthal variation of sap flux density in this plantation, the biases in several methods for calculating sapwood area and the effect of drip irrigation on them, we will not go further into this question here.

Our wild cherry trees (6 years old in 2011), growing under drip irrigation (I) and under rainfed conditions (NI), showed maximum water consumption in the early periods of the 2012 and 2013 growing seasons, 8.5 to 12.8 litres day⁻¹ and 5.4 to 6.7 litres day⁻¹, respectively. These values are in the low range when compared with other wild cherry plantations of similar age growing in Mediterranean areas, although transpiration in other plantations was highly heterogeneous as these were affected by either high intertree or intra-tree variability or both. Cabibel and Isberie (1997) compared tree transpiration during one summer month in trees growing under irrigation versus

rainfed conditions and obtained big differences: 101 litres day⁻¹ versus 15 litres day⁻¹, respectively. These authors also observed preferential water flow pathways in the soil-plant system, in contrast with the results found in the study plantation (Molina et al., 2015). Dupraz et al. (1995) found, in trees growing under rainfed conditions for two summer months, mean tree water consumption of 15 litres day⁻¹. Chifflot (2003) observed mean tree water consumption of about 5 litres day⁻¹ in wild cherry trees growing under irrigation conditions. Lambs et al. (2008) found for two summer months in wild cherry trees under rainfed conditions a mean tree water consumption of 9.5 litres day⁻¹.

The effect of pruning on stand transpiration

Branch pruning in noble wood plantations is a common practice that aims to get a maximum of free tree trunks and so increase timber value (Kupka, 2007). In our study, branches were pruned at the beginning of the summer period, when trees are expected to recover better due to better wound occlusion (Springmann et al., 2011).

It has been suggested that, in the short term, a certain level of branch pruning promotes stem diameter growth through physiological responses (Pinkard and Beadle, 2000). While most of the literature on the effects of branch pruning focuses on Eucalyptus plantations (e.g. Muñoz et al., 2008; Pinkard and Beadle, 2000; Pinkard et al., 2004), for which it has been suggested that an optimum of branch pruning of 40 to 50% of the total crown increases diameter growth (Pinkard and Beadle, 2000), the results in wild cherry tree plantations are the opposite, with negligible or even negative effects of pruning on diameter growth (Kupka, 2007; Springmann et al., 2011). In our study, the branch pruning carried out in June 2013 greatly reduced canopy cover (decreases of 56 to 63% for I and for NI plots, respectively), but the basal area increment (BAI) in the 2013 growing season was very similar to the BAI in the 2012 growing season, with differences of about 6% for the two plots (data not shown). Thus, the monthly tree diameter measurements within the growing periods showed that our wild cherry trees grew mainly from April to June, with 91 and 97% of the total cumulative basal area growth achieved in this period (Figure 6). Branch pruning also led to a reduction of about 30% in stand transpiration. Thus, the higher transpiration expected in the entire 2013 growing season, due to both higher canopy cover and sapwood areas, was not achieved (Figure 1). Therefore, when BAI is expressed over

tree transpiration, as a proxy for water productivity at stand scale (WP, Fereres and Soriano, 2007), the ratio was very similar between the growing periods, since BAI and stand transpiration were similar in the plots studied (data not shown). Therefore, we hypothesize that branch pruning at the end of June seems to be optimal, since WP did not change between the growing seasons as and it is recognized as a valuable tool for managing plantations in water-limited regions such as the Mediterranean (Fereres and Soriano, 2007).

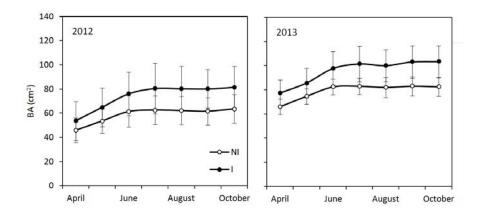


Figure 6. Means and standard deviations of basal area (BA, cm²) during the 2012 and 2013 growing seasons.

Stand transpiration response to environmental conditions and modelling

Reference evapotranspiration (ET₀) calculated by the Penman-Monteith FAO equation was chosen to express the evaporative demand of the atmosphere in this study, since its integrative approach to modelling the evapotranspiration of a reference crop with optimum soil water conditions has been satisfactorily used elsewhere (Allen et al., 2006); and the data are normally available from official climate station networks on a daily basis. The stand transpiration of our wild cherry growing under drip irrigation conditions showed logarithmic relationships with ET₀ (Figure 2), thus indicating the well-known stomatal closure to high vapour pressure deficits (Granier et al., 2000; Petzold et al., 2011; Poyatos et al., 2005). A greater stand transpiration response to ET₀ was observed in 2013 as a consequence of a higher stand transpiration rate during most of the 2013 period (Figure 3). Thus, a single set-up of model parameters for the entire study period was not possible. However, after applying a simple correcting factor dealing with the different water conducting areas between 2012 and 2013, the

results from the modelling were acceptable and comparable to others (Table 2) (Ewers et al., 2008; Lagergren and Lindroth, 2002; Petzold et al., 2011; Poyatos et al., 2005).

The next step in modelling was to obtain the maximum stand transpiration from data of the trees growing under irrigation conditions (Figure 3) and the soil water depletion function from data of the trees growing under rainfed conditions (Figure 4). The approach followed in this study allows highlighting the physiological differences between wild cherry trees growing under irrigation conditions versus rainfed ones, as both data were used in the modelling. The greater differences between the modelled and observed data for the trees growing under rainfed conditions were found at the end of the growing seasons and also, but to a lesser extent, in the early spring of 2013 before branch pruning (Figure 5). In October 2013, the differences between modelled and observed data were the highest yet, with the former about 6 times higher. These biases may arise from the uncertainty about sap flow measurements under low vapour pressure conditions (Ewers and Oren, 2000; Lagergren and Lindroth, 2002; Oren and Pataki, 2001) or from errors related to the canopy cover measurements under low leaf density conditions. These uncertainties probably explained the results on model fitting during the other periods with similar evaporative demand conditions, although they were probably not the only factors responsible, as well as the model response to the soil water content increase. The tree transpiration response to the big changes in soil water content in these periods, caused by high-intensity showers, was of low intensity in both treatments (for instance, in October 2012 when REW increased from 0.2 to I), while the model dramatically indicated intense stand transpiration responses, especially in October 2012. Our f(REW) indicated a strong limitation of stand transpiration when REW< 0.6, while other studies observed lower REW values for data pooling: 0.25 for pine and spruce (Lagergren and Lindroth, 2002), 0.3 for poplar (Petzold et al., 2011) and 0.4 as a mean value for various species (Granier et al., 2000). This seems to indicate that wild cherry trees are more sensitive than other species to soil water content depletion, since optimum transpiration was found for a smaller REW range (0.6-1). Therefore, the intense stand transpiration responses predicted by the model in these rainy periods of low evaporative demand also contrasted with the negligible basal area growth data during these periods (Figure 6), which indicates a high degree of stomatal closure at this time of year. However, the model overestimate at the beginning of 2013 was unexpected and could not be explained by the difference in

the water conducting area between the I and NI plots, since a correction factor to account for this was included in the modelling. Thus, this result seems to indicate contrasting physiological behaviour between our experimental plots during this period (Figure 3). Further physiological measurements may help to interpret these differences.

Conclusions

The stand transpiration of wild cherry trees growing under irrigation conditions was about twice that of the trees growing under rainfed conditions. The branch pruning carried out in the second year of the study (by the end of June) led to tree water use reductions of about 30%, but basal area increment was very similar in the two years of study, which resulted in very similar water productivity. This result indicates the role of branch pruning on controlling water use when managing this type of plantation under water scarcity conditions, especially when most of the year's basal area increment was achieved before the branch pruning.

Wild cherry has been shown to be more sensitive to soil drought in the 0-50 cm soil layers than many other species, as indicated by the function f(REW) showed in the present study. The logarithmic relationship between the stand transpiration of our irrigated trees and ET₀ allowed us to develop a simple model to account for the effects of soil water content and canopy cover evolution on stand transpiration, and for the physiological differences between trees growing under irrigation and rainfed conditions. However, further physiological measurements are required to explain the considerable differences between modelled and observed values under rainfed conditions.

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Effect of irrigation on sap flux density variability and water use estimate in cherry (*Prunus avium*) for timber production: azimuthal profile, radial profile and sapwood estimation

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Chapter 4

Effect of irrigation on sap flux density variability and water use estimate in cherry (*Prunus avium*) for timber production: azimuthal profile, radial profile and sapwood estimation

Abstract

Information on tree water use in plantations for high quality wood is scarce, thus studies are needed to properly estimate the irrigation demand of these plantations. Plant water use estimation with sap flow sensors has been used extensively. However, biases in tree sap flow estimate can arise from variations on radial and azimuthal profiles of sap flux density and also from the sapwood area considered for the upscaling from sap flux density to sap flux. This work aimed to 1) study the spatial variations of sap flux density in cherry trees in a timber orientated plantation, 2) compare several methods to estimate sapwood depth in cherry trees and 3) to evaluate the effect of drip irrigation on these factors. The results showed that most of the studied trees had decreasing radial sap flux density profiles with depth as expected. However, the three irrigated trees of bigger sizes still showed high sap flux densities in their inner tissues, at contrast with the rest of the trees and especially with the non irrigated ones of similar size with values close to 20 % of the sap flux density measured at I cm depth from cambium. On the other hand, the different methods tested to estimate sapwood depth gave significantly different results and only the two methods of visual identification in wood cores based on color change and measurements of sap flux densities along the xylem radius may be considered suitable for scaling purposes. Moreover, azimuthal variation pattern was found to be random in all the studied trees, and the ranking between the aspects (north, south, east and west) was not affected by either drip irrigation or sun exposition, and thus measuring sap flux density in any particular aspect has been shown to be suitable to estimate the overall tree sap flux. We conclude that more studies are necessary to properly assess the radial profile of sap flux density, especially when considering the high sap flux density in the inner tissues of the three bigger irrigated trees as compared to the other trees, and also how this pattern seemed to indicate sapwood depths values very contrasted to the ones estimated from color change in wood cores.

Keywords: wood, heartwood, cherry tree, heat pulse, tree circumference

Introduction and objectives

Plantations of Angiosperm trees for high quality timber production (commonly named hardwood) have increased in recent years in Europe as a consequence of two main causes, a permanent demanding market that is not entirely satisfied by the own production, and the EU regulations promoting their establishment due to their important environmental role in CO2 capture (Cambria et al., 2012). The later is especially noticeable when considering that timber from the tree species normally used, such as walnut or cherry, take between 30 and 50 years to get its maximum market value (Cisneros, 2004).

The plantations for high quality timber production are normally developed in areas subject to depopulation, where the normal cultivars are not economically viable. Management of these plantations consisted of several operations such as pruning, thinning, soil tillage, fertilization and irrigation (Cambria and Pierangeli, 2012), and as in any other economical activity, the related cost should be carefully evaluated. Irrigation demand of these plantations is frequently estimated using the FAO procedures with crop factors of fruit plantations, because specific crop factors for timber plantations are still scarce. However, the orchard management of timber plantations is normally quite different than that of fruit plantations (leading for instance to different tree architecture and tree density), and this could lead to important biases in the evapotranspiration estimate.

Sap flow sensors based on heat pulse methods have been widely demonstrated as a valuable tool for measuring water use by trees (Burgess et al., 2001; Green et al., 2003). There are, however, several factors to take into account when scaling up to the whole tree, such as the spatial variations of sap flux density within the tree, i.e., the radial and the azimuthal variations (Chang et al., 2014; Kume et al., 2012; Nadezhdina et al., 2002), and a correct estimation of the sapwood depth (Čermák and Nadezhdina, 1998). Furthermore, orchard management techniques such as branch punning and localized irrigation could contribute to add variability (Cabibel and Isberie, 1997; Lu et al., 2000).

Radial variation in sap flux density has been a topic widely studied in several tree species under different conditions (e.g., Nadezhdina et al., 2002; Gebauer et al., 2008; Cohen et al., 2008). The general finding is a decreasing of the sap flux density from the

outer to the inner sapwood, though this profile may show different shapes (Alvarado-Barrientos et al., 2013; Gebauer et al., 2008; Kubota et al., 2005) and it could vary with species, tree age and environmental conditions (Čermák and Nadezhdina, 1998). Azimuthal variation of sap flux density, for its part, is understudied as compared to radial variation (Kume et al., 2012), though it may be of higher magnitude (e.g., Shinohara et al., 2013; Lu et al., 2000). In the other hand, the observed results are more contrasting (e.g., Cabibel and Isberie, 1997; Kume et al., 2012; Tsuruta et al., 2010; Shinohara et al., 2013) and consequently general conclusions about tree circumference profile of sap flux density are difficult to be drawn.

Sapwood is the outer part of the xylem conducting sap, which contains living parenchyma cells (Čermák and Nadezhdina, 1998). It can be estimated by several methods, but they normally show contrasted results even for the same species (Čermák and Nadezhdina, 1998; Nadezhdina et al., 2002). As example, Poyatos et al. (2007) found that the sapwood area estimated from radial patterns of sap flux density was 1.5–2 times larger than sapwood area estimates made in the field based on visual inspection of wood cores.

For three years we have been monitoring water use by cherry trees in a timber orientated plantation in a Mediterranean area, under rainfed and irrigated conditions. To this end, sap flow sensors, based on heat pulse with two radial measurements within the sapwood were installed at the east aspect of the studied trees. Sapwood was determined by visual identification of sapwood and heartwood in tree cores.

The aim of this work was to evaluate the accuracy of the approach followed during these three years of experiment (2011-2013) to estimate tree water use with a deeper study in 2014 of the spatial variations of sap flux density (is representative enough measuring sap flux density in two points and only in one aspect of trees?) and, on the other hand, to compare several methods to estimate sapwood depth in cherry trees (was sapwood depth well estimated?). Finally, the effect of drip irrigation on these aspects was also evaluated.

Materials and methods

Study site

The study was carried out at the Torre Marimon site (Caldes de Montbui, Spain) in the IRTA facilities (41° 36 '47 '' N, 2°10'11 'E, 170 m.a.s.l). Climate is Mediterranean, with mean annual (1999-2012) rainfall and potential evapotranspiration of 599.4 \pm 33.4 and 846.8 \pm 23.3 mm, respectively. Soils are basic and have two contrasting characteristics, sandy-loam soils with about 60 % of gravels and loam soils with negligible presence of gravels; these differences are due to an alluvium trend brought by a close river did not affect to the same extent the study area.

Measurements were conducted during the growing season of 2014 in an 8 year-old cherry tree (*Prunus avium* L.) plantation orientated to timber production with tree spacing of 4x4 m (625 trees ha⁻¹). Trees are pruned every two years at the middle of the growing season. Pruning follows the common practices, where the lower part of crowns is removed, and it approximately accounted for one third of total aboveground biomass.

Meteorological conditions during the experiments were measured in an open area next to the plantation. Mean daily values of potential evapotranspiration and maximum temperature during the summer months (July and August) were 4.6 ± 1.3 mm and 29 ± 2.4 °C respectively, while total rainfall accounted only for 45.4 mm.

Experiment 1: Radial variation in sap flux density and sapwood area estimation methods

Radial variation of sap flux density was studied by mean of two sensors based on the compensation heat pulse method (CHPM, Green et al., 2003) with 4 radial measurements at 0.5, 1.2, 2.1 and 3.2 cm from bark, and the bark of 2 mm depth was not previously removed to install the sensors. The CHPM sensors were programmed to measure each 30 minutes, and were charged with a battery of 80 mAh and 12 V connected to a CR-1000 logger (Campbell Scientific, USA). Each sensor consisted of three needles 40 mm in length and 1.8 mm in diameter. The needle placed in the centre is the heater that emits the heat pulse during I second. Then, the temperature increase is systematically measured during 8 minutes in the other two needles at 0.5 cm and I cm upstream and downstream respectively. Heat pulse velocity is estimated from the time taken to obtain the same temperature upstream and downstream.

The measurements were simultaneously conducted during seven days at 1.3 m height and at the east aspect of the trunk in two close trees with similar diameter but different irrigation treatment (drip irrigated versus non-irrigated trees). Seven days after the two CHPM sensors were moved to other two trees and this was repeated seven times, resulting in a total of 14 trees sampled and a sampling period of 49 days (Table I). Irrigation treatment consisted of four emitters per tree (16 I h⁻¹ tree⁻¹) located at 25 and 50 cm at north and south sides from trunk. Daily doses were estimated at the beginning of each week as the 60 % of the ET₀ for the previous week, and irrigation was not applied when ET₀ was lower than rainfall. Total irrigation per tree ranged from 8 to 125 I day⁻¹ when applied.

Table 1. Diameter at breast height (DBH) and sapwood depth, estimated from colour change in wood cores (VDm, see text for details), of the studied trees in the radial experiment. Drip irrigated (I) and non-irrigated trees (NI).

			Sapwood	
Irrigation	Tree n°	DBH (cm)	depth(cm)	Measurement period (DOY)
NI	I	7.0	2.9	
1	2	7.7	2.8	198-204
NI	3	7.7	2.7	
1	4	8.6	3.3	205-211
NI	5	7.7	3.4	
1	6	9.4	3.5	191-197
NI	7	9.3	3.0	
1	8	11.4	4.7	184-190
NI	9	9.4	3.2	
1	10	11.9	3.8	212-218
NI	11	10.6	3.4	
1	12	13.3	4.4	219-225
NI	13	12.1	3.6	
I	14	12.8	4.3	226-232

Heat pulse velocity was corrected for effects of probe-induced wounding (Barret et al., 1995), following the numerical approximation proposed by Swanson and Whitfield (1981) as a function of wound width. Since no wound width is available from literature for the study specie, we adopted the results of Barret et al. (1995) confirmed by Fernandez et al. (2006) in plum trees (*Prunus domestica* L.) of 1.8+2×0.3 mm. Sap flux density was estimated from corrected heat pulse velocity following Barrett et al. (1995).

Once the seven days-period of CHPM measurements finalized in each couple of trees, we proceeded to core the measured trees. Seven different methods were used to determine the limit between sapwood and heartwood: (I) methyl orange or (2) lugol staining, (3) visual differentiation based on colour change (VD_m) , (4) dye injection (DI_m) , (5) radial variation of wood density (WD_m) , (6) wood water content (WC_m) and (7) radial profile of sap flux density.

Methyl orange and lugol methods were prior tested in several cores taken in neighbour trees, as no colour change was observed in any of the samples these methods were not further considered.

For the other methods we extracted one core from each tree with a Pressler increment borer (Suunto Finland) at 35 cm below the sensor position. The cores collected were immediately taken to laboratory and kept in a refrigerator. Visual identification of sapwood and heartwood radiuses were measured (VD_m) and cores were sectioned into small cylinder sections of 8-10 mm length, and volumetric fraction of water and basic density of wood (WD_m and WC_m) were estimated following Nadezhdina et al. (2002). The hole done by this first coring was then used to inject 0.1 % acid fuschin dye (following Umebayashi et al., 2007) and 2-3 hours after we extracted a second core at 15 cm under the sensor position (at 20 cm distance from the first core) to estimate the coloured part as sapwood (DI_m). Moreover, 4 extra trees were cored in the last week of the experiment to determine sapwood through VD_m and DI_m, though the coring for DI_m was taken at 5 cm from the first hole, instead of 20 cm. Sapwood depth was also estimated from the radial profile of sap flux density (7) taking into account the point where the sap flux density approached to zero by fitting linear regressions to data (e.g., Nadezhdina et al., 2002, Cohen et al., 2008).

Finally, sapwood depth was expressed as percent of xylem radius to compare between the methods (SW_r). The t-student test for paired data was used for comparing the methods.

Experiment 2: Azimuthal variation in sap flux density

Azimuthal variation in sap flux density was studied by mean of HRM sensors (Burgess et al., 2001) with two radial measurements at 1.25 (outer measurement) and 2.75 (inner measurement) cm from bark. The HRM sensors were programmed to measure

every 60 minutes, and were charged with 12-W solar panels connected to their internal batteries (ICT International, Australia). Each sensor consisted of three needles 35 mm in length and 1.3 mm in diameter. The needle placed in the centre is the heater that emits the heat pulse during 1 or 2 seconds, and the temperature increase is then systematically measured during 100 seconds in the other two needles at 0.5 cm upstream and downstream from the heater. Each couple of measures (inner and outer) is used to estimate the heat pulse velocity by considering a thermal diffusivity average value of 0.002 cm² s⁻¹ estimated from the cores taken in the first experiment for sapwood determinations, following Burgess et al. (2001).

The measurements were continuously carried out from May to August 2014 in 6 trees within 2 different DBH classes (Table 2). Four sensors were inserted in each tree approximately 1.3 m above the ground at north, east, south and west aspects (following Shinohara et al., 2013 and Lu et al., 2000).

Table 2. Diameter at breast height (DBH) and sapwood depth, estimated from the relationship with diameter obtained with the VDm (see text for details), of the studied trees in the azimuthal experiment. I: Irrigation period, NI: water stress period (see Figure 1 for details).

Tree	DBH	Sapwood	
n°	(cm)	depth (cm)	Measurement period (DOY)
15	8.1	3.5	96-190 (I),191-234 (NI)
16	8.7	3.7	96-190 (I),191-234 (NI)
17	9.0	3.8	96-190 (I),191-234 (NI)
18	10.1	4.2	96-190 (I),191-234 (NI)
19	10.3	4.3	96-190 (I),191-234 (NI)
20	11.2	4.6	96-190 (I),191-234 (NI)
-			

The sensors were placed at slightly different heights to avoid interferences among the readings. All trees were drip irrigated from May to June following the same irrigation scheme than in experiment I. However, in order to study whether the ranking

between the aspects were maintained in each tree due to induced soil water stress on azimuthal variation, drip irrigation was stopped from July on (Figure 1).

The heat pulse velocity was corrected for the effects induced by probe misalignment and wounding. Probe misalignment correction was done by comparing the baselines corresponding to zero sap flow during the first leafless week of measurement to the baselines from all the measurements. Wounding correction was applied in the same way than in the radial experiment (1.3+2x0.3 mm) but following the mathematical approximation of Burgess et al. (2001).

Sap flux density was estimated following the same methodology than that in the radial experiment (Barrett et al., 1995). Finally, tree sap flux was calculated from the sap flux density estimated for the inner and outer measurements assuming the sapwood area divided into two concentric bands delimited by the mid-point between them (Bleby et al., 2004). The calculations were made for the entire tree from each of the four aspects, and also for the average sap flux density.

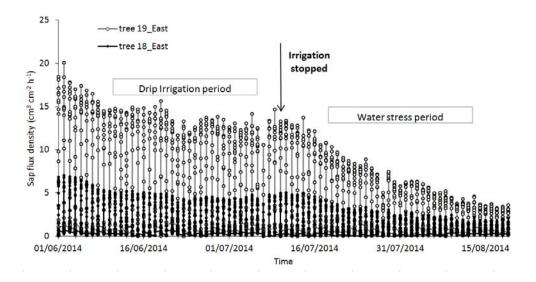


Figure 1. Mean sap flux density (I data every hour) of two sampled trees in the azimuthal experiment (see Table 2 for details).

Results

Radial variation of sap flux density

As expected, mean sap flux density was higher in all the irrigated trees (Figure 2) except for one couple of trees where the radial profile was very similar between the irrigated tree and the non irrigated one (trees 5 and 6, data not shown) (similar to the radial profile from tree 9). The differences in sap flux density were not constant along the radial profile, and differed among trees. The highest mean difference among irrigated and non irrigated trees was found in the most inner measurement at 3 cm from cambium.

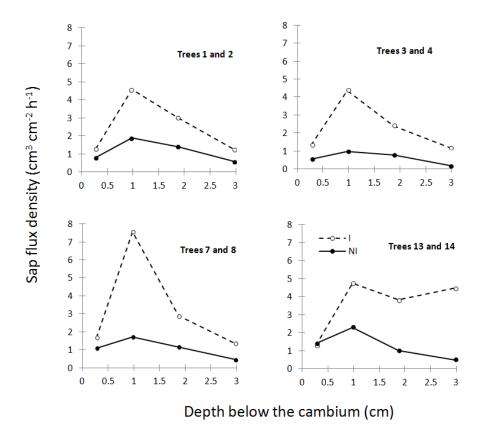


Figure 2. Examples of radial patterns of sap flux density in 4 irrigated trees (I) and 4 non-irrigated trees (NI). Data shown per tree is the average of all measurements during 7 seven days and standard deviations are not shown to improve the understanding. Tree characteristics are shown in Table I. Note that all the studied trees were not shown because they behaved in a similar manner that the ones presented

The results of expressing sap flux density as relative to the expected maximum (ratio over the measurement at I cm from cambium) are presented in Figure 3. The radial profile pattern was very similar for all the non irrigated trees, with maximum sap flux densities from cambium to I cm depth (from 0.8 to I.I cm h⁻¹ at the outer

measurement) and a decrease from this distance on (except for one tree with a value of 0.45 in the outer measurement) until an average reduction of about 24 % at a distance of 3 cm from cambium, regardless the difference on sapwood depth between the trees (from 2.7 to 3.6 cm). In contrast, radial profile of the irrigated trees was less consistent in the outer part (average normalized values from 0.5 to 1), and also sap flux densities were lower. On the other hand, in the other point measurements, the radial profile in the non-irrigated trees (trees 1, 3, 5, 7, 9, 11 and 13) was very similar to those of the irrigated trees with smaller size (tress 2, 4, 6 and 8) but quite different from those of the biggest irrigated trees (trees 10, 12 and 14), still presenting high sap flux density in their inner xylem tissues (Figure 3). In this sense, it is important to remark that the sapwood depth range was very similar between the non-irrigated and the irrigated trees, from 2.9 to 3.6 and from 2.8 to 4.3 cm respectively (see Table 1).

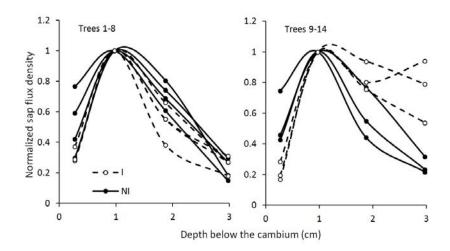


Figure 3. Normalized sap flux density (over the measurement at 1.2 cm from bark) in the irrigated (I) and the non-irrigated trees (NI). Tree characteristics are shown in Table 1.

The unexpected, high difference found in the outermost measurements between the irrigated and the non irrigated trees (mean normalized sap flux density of 0.5 versus 0.9) made us reconsider the quality of the raw data measured with the CHPM sensors. In Figure 4 we show two examples of heat pulse velocity from the four radial measurements taken in two trees during one day: the inconsistencies in the measurements were especially important in the outermost thermocouples (0.5 cm from cambium) as indicated by high fluctuations in the heat pulse velocity values, while in the other measurements no sudden fluctuations were observed during the day. These fluctuations appeared in all the tested trees, so we decided to discard the outermost measurements for the rest of the analysis. However, we decided to keep

these data in Figures 2 and 3 to show the effect of the presence of bark and cambium in the outermost measurements in cherry trees.

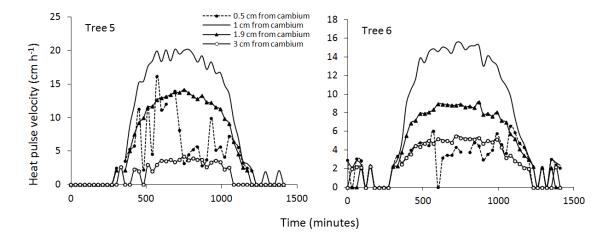


Figure 4. Raw data (heat pulse velocity) from two CHPM sap flow sensors during I day of measurements in two trees (one measurement each 30 minutes) at four depths from cambium.

Comparison of methods to determine sapwood

Basic density of wood and volumetric water content were not significantly different between the irrigated and the non irrigated trees, ranging from 388.5 to 513.9 kg m⁻³ and from 171.9 to 302.7 kg m⁻³ respectively. Radial variation of basic density of wood and volumetric water content was different depending on tree, but a systematic decrease with depth was found for volumetric water content in the irrigated trees (Figure 5). However, this pattern was not consistent enough to distinguish between sapwood and heartwood, and consequently this method was considered not useful to determine the limit between these two tissues in cherry trees.

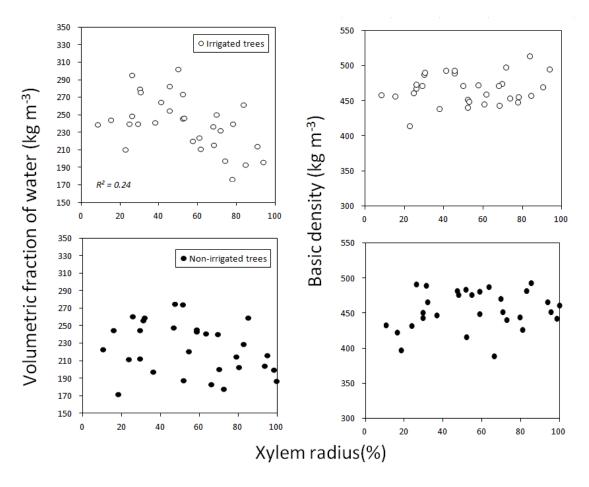


Figure 5. Radial profiles of volumetric water content and basic density (kg m⁻³) in the irrigated (open symbols) and the non-irrigated trees (closed symbols). Only R² values from significant linear regressions were presented.

The results for the rest of methods are presented in Table 3. For the VD_m, three regions were clearly observed in all cores, with the darkest part close to tree pith, identified as heartwood, and turning lighter when moving outer. In the case of radial variation of sap flux density, linear regressions (R² values higher than 0.9) were fitted to all data excluding the outermost readings (as explained in Figure 4), except for trees 10, 12 and 14 where the inner sap flux density remained higher than 60 % of the maximum expected, as indicated in Figure 3. On the other hand, with the DI_m we found a discontinuous pattern for all the trees, with coloured bands (the expected conducting tissue) and non coloured ones alternating along the xylem radius. In order to estimate the sapwood depth, we assumed that the limit between sapwood and heartwood was located in the most inner part of the last coloured band found.

Table 3. Sapwood depth relative to xylem radius determined from: visual differentiation based on colour change (VDm), dye injection (Dlm) and radial profile of sap flux density (SFr). Note that for trees 11, 12 and 14 linear regressions were not fitted to the sap flow data. Different letters indicate significant mean differences in the paired t-student tests used to compare among methods. SD is standard deviation.

Tree	VDm	Dlm	SFr	
I	0.7	0.3	1.0	
2	0.7	0.5	1.2	
3	0.7	0.6	0.9	
4	8.0	0.4	8.0	
5	0.6	0.6	8.0	
6	0.7	0.6	0.9	
7	0.7	0.6	0.6	
8	8.0	0.5	8.0	
9	0.7	0.4	8.0	
10	0.9	0.5	0.7	
П	0.7	0.5	-	
12	0.7	0.5	-	
13	0.6	0.5	0.7	
14	0.6	0.5	-	
Mean	0.7a	0.5b	0.8c	
SD	0.1	0.1	0.2	

Paired t-student tests used to compare the methods in Table 3 showed that SW_r was significantly different between the methods, and estimation from DI_m resulted in the lowest values. This type of results was also found when including the four complementary measurements carried out during the last week of the experiment in 4 extra trees sampled at a closer distance between the cores (5 instead of 15 cm).

The effect of irrigation on heartwood formation

The influence of irrigation on heartwood formation was assessed by comparing the relationships between tree diameter and heartwood diameter for irrigated and non irrigated trees, obtained with the VD_m method (Figure 6). Heartwood diameter was very similar in both treatments for diameter lower than 10 cm (non significant differences at p-level < 0.001). However, for bigger trees, irrigated individuals had lower heartwood diameters than those of the non irrigated ones, as indicated by a lower value of the a parameter in the quadratic functions fitted to all the data (0.14 versus 0.53).

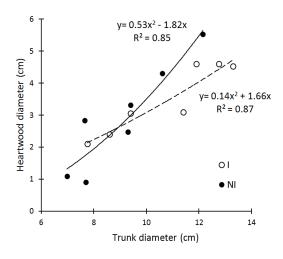


Figure 6. Relationship between trunk diameter (cm) and heartwood diameter (cm) for the irrigated and the non-irrigated trees. Quadratic functions (y=a x²+b x) were fitted and forced through the origin.

Azimuthal variation of sap flux density

Diurnal courses of sap flux densities followed a bell-shaped pattern in all the studied aspects during both periods, with and without irrigation. The coefficient of variation (CV) of the 24 hourly sap flux densities from the 4 aspects of each tree (from the mean of the measurements at two sapwood depths), was higher in the period without irrigation and ranged from 11 to 75 and from 27 to 81 % per tree respectively.

The azimuthal variation of sap flux density for each tree, i.e. the ranking between the aspects for each tree, was expressed as the relative differences over the mean as follow: $RD = ([(SF_i - SF_{mean})/SF_{mean}]/N) \cdot 100$, where SF_i was the daily average from one particular aspect, SF_{mean} the daily average from the 4 aspects measured, and N the number of days studied.

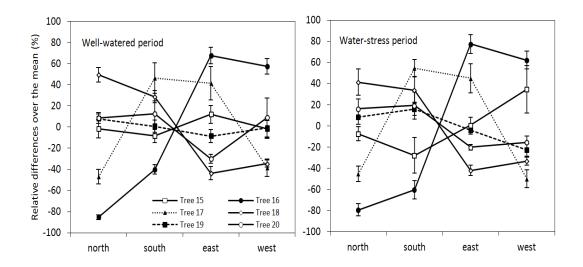


Figure 7. Relative differences over the mean (average and standard deviation values, %) for each study tree and for the both studied periods, the irrigation and non irrigation ones.

During the irrigation period, RD was different depending on the sample tree. For instance, tree 16 showed the lowest sap flux density at the north side (-85 \pm 2 %), while it was the highest at this aspect for tree 18 (50 \pm 7 %). In contrast, tree 19 showed very similar values in all the studied aspects (around 0 %) (Figure 7, left).

The soil water deficit induced by stopping irrigation produced a mean general decrease of 92 % in tree sap flux. The variability between the aspects was higher than in the irrigation period, with a mean increase of the CV values of 23±21 and 35±32 % for the outer and inner measurements, respectively. In contrast, the ranking between the aspects was quite constant for each tree between the two periods (except for tree 15), as indicated by very similar average and standard deviation values for the RD between the irrigation and the without irrigation periods for each tree (Figure 7).

Cumulative tree sap flux calculations were made for each of the four aspects considering sap flux density in each of them as representative of all the sapwood area. As a way to determine which aspect was most representative of the overall cumulative tree sap flux, these calculations were expressed as a percentage of the mean of the four aspects. Average± standard deviation values were 88.3±46.9, 106.4±34.4, 108.7±42.7 and 96.6±38.1 % for north, south, east and west aspect respectively.

Discussion

In this work we evaluated the effects of irrigation on radial and azimuthal variation of sap flux density in 8 years old timber-orientated cherry tree plantation in a

Mediterranean area of Spain. Moreover, several methods for estimating the conducting sapwood area have been tested in order to evaluate its likely effect on the scaling up from sap flux density to tree sap flux.

Radial variation of sap flux density

Several studies have reported a decrease of sap flux density as a result of declines in soil water content, naturally induced through a contrasted dry season or by comparing different irrigation practices (Lu et al., 2000; Philipps et al., 1996). In this study we simultaneously measured drip irrigated and non-irrigated ones, during the dry season (from July to September). We found that irrigated trees showed higher sap flux densities than the non-irrigated ones, but differences were not constant along the xylem radius, with mean differences of 2.5, 2.6 and 5.4 times higher for distances of 1.2, 2.1 and 3.2 cm from bark, respectively. These differences were consequence of a different decrease of sap flux density with sapwood depth related to tree size, with similar patterns for the non irrigated trees and the smaller irrigated ones (7 trees and 4 trees respectively) and a contrasted pattern for the bigger irrigated ones (3 trees) (Figure 3).

The decreasing of sap flux density with sapwood depth is frequently found in literature (e.g., Ford et al., 2004; Cohen et al., 2008) and functions such as the Weibull or the Gaussian ones are frequently used for describing the flow shape (Gebauer et al., 2008; Kubota et al., 2005). In this sense, Nadezhdina et al. (2002) found a peaked distribution on the radial sap flux density pattern (with the maximum at 80 % of total radius) of a tree species very similar to the one studied here, i.e., Prunus serotina, but 19 years old. In our study, the outer thermocouples, placed at 0.5 cm from cambium, were discarded because of their inconsistent behaviors (Figure 4), and thus we could not conclude whether the radial sap flux pattern of P. avium was characterized by a monotonous decrease of sap flux with depth or by a peaked distribution, as the two most common (Cohen et al., 2012). However, we believed that showing the effect of not removing the non homogenous tissue (bark and cambium) has on the quality of the readings was important due to this fact is not usually mentioned in the related literature, especially for species with small bark thickness. This inconsistent behavior was the result of bark and cambium interferences in the measurements (Steve Green, personal communication); first, heat pulse emitted by the heater is likely affected by this sharp change on water conductivity and then heat pulse is not propagated correctly (Steve Green, personal communication), and second, the measurement radius of thermocouples should probably be higher than 3 mm as in other heat pulse methods (taking into account that our non conducting tissue was approximately of 2 mm depth) and thus readings would be accordingly affected. Moreover, the high and persistent difference found between the irrigated and the non irrigated trees seems to indicate that despite the difference on depth of non conducting tissue was about 0.3 mm, this small difference could lead to important effects of the abovementioned explaining aspects.

In addition, when only considering data from of all our non irrigated trees together with the 4 smaller irrigated ones, the radial profile showed similar decreases on the normalized data and consequently a consistent difference of about 2.5 times along the radial profile in the absolute data. However, the other 3 irrigated trees (the biggest trees in the experiment) showed lower radial variations and less pronounced decreases with depth, and even high sap flux densities in the inner tissues (tree 14 in Figure 2). This result is in contrast with the results observed in maritime pines of different sizes, with steeper declines in the trees with the biggest sizes (Delzon et al., 2004). These trees had a small difference of about I cm in the sapwood depth as compared to the non irrigated ones with bigger size (see next section), thus tree size was not the only responsible of these differences (Čermák and Nadezhdina, 1998) but also the irrigation applied during 4 growing seasons (during the experiment presented here and the 3 previous years). It is widely accepted that leaves are connected to new xylem and though xylem can be viable to a large depth in the xylem, sap movement will not necessary occur due to the increasing resistance to lateral flow with xylem depth (Cohen et al., 2012). We found significant differences on leaf area index between irrigated and not irrigated trees of bigger size (data not shown), and also high differences in sap flux density (Figure 2). This supports that, though xylem is viable for both type of trees, irrigation seems to induce changes in the xylem which is actively conducting water in order to maintain the ratio of cross sectional area of actively conducting xylem to leaf area constant (Cohen et al., 2012).

Sapwood determination and heartwood formation

Our results have shown that several methods, which have been used for delimiting sapwood depth in other species (methyl orange, lugol, and radial variation of xylem density and xylem water content), were not suitable for delimiting the sapwood depth in Prunus avium and thus these methods are not recommended for scaling purposes for this species. In this sense, Čermák and Nadezhdina (1998) found that the radial variation of xylem water content was useful to delimit the sapwood depth for some species but not for others. In contrast, the rest of the methods showed clear differentiation between sapwood and heartwood tissues but significantly different values for the sapwood depths. Visual identification carried out in ring cores is related with the different chemical processes taking place in the two tissues, and our results are in agreement with those from Nadezhdina et al. (2002), who found three different colored parts in ring cores of Prunus serotina and a sapwood depth very similar to ours (75 % of the xylem radius versus 80 %). However, while these authors found a clear relationship between these three different colored parts and the sap flux density measured along the xylem radius, normally considered as a robust proxy for sapwood depth estimation (Cohen et al., 2008; Poyatos et al., 2007), this was not the case in the present study. Though we found a significant difference between these two methods (13 % in average), this bias may be considered as negligible for scaling purposes (from point measurement to plantation transpiration), because of the low value of sap flux density at deeper depths (and consequently the sap flux at these depths) in the trees where the estimation of sapwood depth was accomplished with the sap flow measurements (11 out of 14 trees). In contrast, the high sap flux densities at the inner depths of the other 3 bigger irrigated trees did not match with the sapwood depth values found in the other two suitable methods, and how it was discussed before it remains an open question for further studies, especially when considering that this could introduce important errors when scaling from measurement to tree transpiration.

In addition, a particular result was found for the dye injection method, with a systematic lower value (average of 25 %) of the xylem radius in all the tested trees, as compared with those from either the radial profile of sap flux density or the visual identification. This may be related with the discontinuous pattern of the water conducting areas showed by the dye injection method. This pattern is more related to

ring porous species with water conducting regions and non conducting regions within the sapwood (Tsuruta et al. 2010), and thus was unexpected due to the semi-diffuse porous wood of *Prunus avium* (García-Esteban et al., 2003). In this sense, the results of Boumghar (2012) when using dye to differentiate between sapwood and heartwood showed that all the sapwood was coloured in the studied ring porous species so this discontinuous pattern was not found. We thus hypothesize that an artifact could be introduced with the dye injection method and thus complementary anatomical studies are needed in this species.

On the other hand, heartwood formation was affected by irrigation for tree diameters bigger than 10 cm (Figure 6), pointing out again the hypothesis that the inner tissues of the bigger trees in our experiment, subjected to cumulate water stress periods underwent anatomical changes as the result of faster growth and bigger diameter. This result seems to be an open research question to be resolved for the wood industry, because the presence of heartwood in high quality woods is nowadays increasing their market values (Vilanova, personal communication).

Azimuthal variation of sap flux density

Sap flux density was measured in four different aspects of six trees (24 sensors) during two periods, the first characterized by irrigation and the second one by no irrigation and probably a certain degree of water stress given by summer conditions (high potential evapotranspiration, low rain).

The decrease in tree sap flux from one period to the other was as expected for the species, with a general reduction of 92 % in tree sap flux compared to a reduction of 85 % when comparing between irrigated trees and non irrigated trees observed during one experiment carried out in summer under Mediterranean climate (Cabibel and Isbérie, 1997). This indicates the high sensibility of this species to dry periods characterized by high soil water deficit and water evaporative demand, as other authors have already pointed out (e.g. Juhász et al., 2013; Cabibel and Isbérie, 1997). On the other hand, the sap flux density variability within each tree (CV of the 4 aspects) was higher during the water deficit period, with a mean increase of about 30 % respect to the irrigation period. In contrast, the ranking between the aspects for each tree was quite constant between the periods, pointing to a relevant weight in the azimuthal pattern of the individual structural conditions in each tree.

Azimuthal variability in sap flux density can be larger than the radial variability (e.g., Shinohara et al., 2013). However, this variability has been found less predictable, because it can be explained by differences on tree exposure to sun (Granier, 1987), soil water content around the tree (Cabibel and Isbérie, 1997), anatomical xylem structure (Tateishi et al., 2008) or crown architecture (Lu et al., 2000). In this sense, we expected the azimuthal variability to be highly controlled by the soil water content around trees with a virtually negligible effect of ET, as suggested by the results of Cabibel and Isbérie (1997) in a drip irrigation experiment (with 2 emitters of 4 L h⁻¹, I m east and I m west from trunk) in cherry trees. However, we found no connection between the soil water content around the tree (emitters placed north and south sides from trunk, 0.8 m² each wet bulb) and the ranking in the sap flux density of the different aspects during the irrigation period. We thus postulate that most of the primary roots were affected by drip irrigation, and consequently no clear connections or preferential fluxes between roots and leafs from the same aspect could appear. In addition, the way the trees were pruned led to not systematic differences on tree architecture and hence on azimuthal variability of anatomical xylem structure, which could also affect the azimuthal pattern of sap flox density in the studied trees. Therefore, when we compared the overall tree sap flux estimated considering the sap flux density from each aspect and the one considering the mean sap flux density, all of them performed similarly for all the studied trees, with average tree sap flux from each aspect not being significantly different than tree sap flux estimated from the average sap flux density.

Conclusions

As presented in this work, the maximum sap flux density in our cherry trees of 8 years old is located at 1 cm from cambium or closer, because the values in inner tissues were of lower magnitude in all the studied trees and consequently decreasing profiles with depth were observed. Our sensors also measured sap flux density at 0.5 mm from cambium, but the readings were finally discarded due to interferences promoted by the presence of bark and cambium on the readings. However, we did not find any advice regarding this fact in either the CHPM manual or articles using this method, so we decided to show the effect that this thin non conducting tissue had on the readings. In contrast, in the HRM method, as the one we used in the azimuthal experiment and in the previous measurements from 2011 to 2013, the outermost measurements (at

1.25 cm from cambium) are not affected by bark+cambum depth lower than 5 mm, which is properly described in the corresponding manual. Therefore, further studies are necessary to assess the sap flux density close to the cambium in order to a better assessment of the radial profile shape in cherry trees for timber production.

The different methods to estimate sapwood gave contrasted results and a number of them are considered as not suitable. The differences found between visual identification and radial profile methods can be considered negligible for scaling purposes due to lower values of sap flux density at these depths.

The effect of irrigation on spatial variations of sap flux density affected to different extents. All the irrigated trees gave higher sap flux densities along the xylem radius and the radial profiles of normalized values were similar between irrigated and non irrigated trees. However, more measurements are needed in the inner depths of irrigated trees of bigger sizes as shown in our results. On the other hand, the azimuthal variation was found to be unpredictable and we consequently conclude that any aspect can be selected to measure sap flow in young cherry trees drip irrigated and pruned to timber production.

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CHAPTER 5

Water isotopes measurements in a noble plantation with two contrasted soil types under Mediterranean conditions. Lessons learned and implications for assessing water uptake by trees

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Chapter 5

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Abstract

In the present study, we took advantage from having two soil types, within our wild cherry experimental plantation, with very contrasting soil water holding capacities and therefore expected differences in tree water use. During three years, water isotopic compositions of rainfall, plant xylem (wild cherry tree plus the most representative spontaneous herbaceous plants) and soil were monitored. We first aimed at testing the hypothesis that the soil water isotopic composition in depth, as well as its variability, was different between soil types. Second, we tested if we could decipher at which soil depth trees and ground cover vegetation extract water, aiming at observing possible water use competence or differences between their water use strategies. The soil of lower quality showed higher variability in both water isotopes, as well as a greater effect of evaporation in the uppermost layers; the latter resulting on more enriched soil water, sometimes until a depth of 50 cm. The direct inference of water uptake by plants indicated that trees and the spontaneous herbaceous plants were taking water from the superficial layers; most of the time from depths lower than 25 cm. Finally, thanks to transpiration data, we observed how the trees with the lowest transpiration rates presented much enriched values for the water isotopes (as indicated by the deuterium excess) with respect to water samples from soil and other trees. Therefore, we concluded that water isotopes technique may not provide successful results for assessing water uptake by trees under soil water stress conditions.

Keywords: Agroforestry systems, wood production, Prunus avium L., stable isotopes,

water use

Introduction and objetives

Differences in the physical properties of the stable isotopes of hydrogen and oxygen in water result in fractionation during physicochemical reactions, thus providing important information about physical and biological processes. A particular use of water stable isotopes in plant was promoted when it was discovered that hydrogen and oxygen isotope were not fractionated during root water uptake (Zimermam et al., 1968).

Over the last decades, several studies have measured water stable isotopes in xylem and soil to a better understanding of water utilization by plants and therefore to assessing the possible different water uptakes strategies of coexisting species (Ehleringer and Dawson, 1992; Querejeta et al., 2007; White et al., 1985; Wang et al., 2010). For a particular moment, a gradient in the water isotopic composition can be observed in soil profiles because of evaporation in the uppermost surface layers, remaining the upper soil horizons enriched in heavy isotopes (Allison and Hughes, 1983, Zimmermann et al., 1967). The change of water isotopic composition with soil depth allows elucidating the depth at which roots uptake water through comparing the isotopic composition of xylem water with that of soil water from different depths. To this end, mixing models are usually used, since they can estimate the proportional contributions of soil water sources through an iterative process (Phillips and Gregg, 2003; Link, 2014; Wang et al., 2010).

Apart from the errors associated to plant and soil sampling and to water extraction (Barnard et al., 2006; Dawson and Ehleringer, 1993; Martín-Gómez et al., 2015; Walker et al., 2001), some considerations should be taken into account when using water stable isotopes to elucidate tree water uptake. In some cases, there is not a gradient of the water isotopic composition in the soil profile, because there is no evaporation from uppermost surface layers (Burgess et al. 2000), in others there is an hydraulic redistribution of soil water by trees (Dawson, 1993), and may also occur that the migration of water vapour from uppermost surface layers offset the enrichment caused by evaporation (Barnes and Allison, 1988). On the other hand, the enriched water in the phloem may be mixed with xylem water, especially in young non suberized stems (Dawson and Ehleringer, 1993; Walker et al., 2001). In this context, Brandes et al. (2007) observed more enriched water in tree xylem than in the

uppermost soil surface layers; the authors indicated that a possible explanation could be the low tree transpiration rate during winter, which could promote the mixing of phloem and xylem waters.

The use of water stable isotopes in agronomy has been less profuse than in natural forest ecosystems. In agricultural systems, root water uptake has been traditionally studied by excavating and the subsequent direct measurement of the root proportions in the different depths. However, this is a destructive technique and does not always give the true water uptake (Dawson and Pate, 1996; Green and Clothier, 1999). In this respect, water stable isotopes provide valuable information in systems where trees and grasses are growing in association, such is the case of some agricultural systems, because they allow assessing the belowground competence in a lesser destructive and time-consuming way (Fernández et al., 2008; Link, 2014).

In the present study, we took advantage from having two soil types, within our wild cherry experimental plantation, with very contrasting soil water holding capacities (Molina et al., 2016) and therefore expected differences in tree water use. One soil was characteristic of Mediterranean areas where agriculture was not developed due to low soil quality, while the other was representative of soils used for agriculture under the same climatic conditions. Our first aim was to test the hypothesis that the soil water isotopic composition in depth, as well as its variability, was different between soil types during the three years of our study. The second objective was to test if we could decipher at which soil depth trees and spontaneous herbaceous plants extract water, aiming at observing possible water use competence or differences between their water use strategies. Finally, we hypothesized that trees with the lowest transpiration rates may present enrichment of the heavy isotopes in their xylem water.

Materials and methods

Study area and experimental design

The study was carried out at the IRTA-Torre Marimon experimental facilities (Caldes de Montbui, NE Spain, at 159m a.s.l). The climate is Mediterranean with mean annual values (1991-2010) for temperature, evapotranspiration and rainfall of 14.4 ± 0.2 °C, 846.8 ± 23.3 mm and 599.4 ± 33.4 mm, respectively. The experimental plantation was located on an alluvial terrace with two soils of contrasted characteristics: one soil with

sandy-loam texture and high presence of gravels (about 50 %) and the other with loam texture and negligible presence of gravels. From this point on, the former will be referred as forest soil and the later as agriculture soil. See Molina et al. (2016) for a detailed description of both soils.

The trees in the experimental wild cherry plantation (clone Salamanca 4) were planted for timber production in 2008. Tree density was 625 trees ha⁻¹ with spacing of 4 m between trees and rows (16 m² per tree). Rows followed a north-south orientation. Trees are pruned every one to two years for obtaining trunks free of branches, in line with timber production practices. In this study, the water samples from soils and trees were obtained from plots under rainfed conditions and where the ground vegetation was left to spontaneously appear.

Water sampling: rainfall, groundwater, soil and plant

Water sampling for the stable isotopes determinations was carried out during 3 years (2011 to 2013) under rainfed conditions. All the sample types were collected following the standard procedures described by the International Atomic Energy Agency (IAEA). Rainfall samples were collected approximately every two months with a funnel connected to a 1 L bottle with a 5 cm layer of paraffin oil on top to avoid evaporation. Rainwater samples were filtered to remove paraffin oil and residues. Groundwater was sampled in a deposit used for irrigation, which housed water from several wells located very close to the experimental plantation. After field sampling, all the samples were immediately sealed in glass vials (air-tight tubes, Duran GL-18, Duran Group GmbH, Mainz, Germany).

Plant and soil samples were collected twice a year, at early and late summer, and at least 5 days from the last rainfall event. Rainfall, evaporative demand as well as soil water content for the period prior to the sampling campaigns is presented in Table I. Soil water content was calculated from FDR sensors (10-HS sensors, Decagon Devices, USA) placed at 25, 50 and 100 cm depth in several places within the experimental plot. Molina et al (2016) included a detailed description of environmental variables during the study period.

Table 1. Environmental conditions during the period prior (7 days) to the sampling campaigns. Cumulated reference evapotranspiration (ET_0), cumulated weekly rainfall and soil water storage in the 100 cm soil profile.

Sampling date	,	Rainfall	Soil water storage (mm)		
		depth (mm)	Agriculture	Forest	
21-07-2011	35.2	28.7	186	79	
15-09-2011	31.2	0	183	95	
5-07-2012	39.3	10.6	131	65	
26-09-2012	18.3	0.8	115	59	
15-07-2013	35.2	3.1	150	73	
30-09-2013	19.1	0	159	96	

Four trees were sampled in the forest soil and 2 trees in the agriculture soil, according to the higher soil variability in the forest plot (Molina et al. 2016), and thus a higher expected variability of the water isotopes in the xylem tissue. The samples were taken from the south-facing tree crown. Sunlit suberized twigs of I year age were harvested (Dawson and Ehleringer, 1993) near midday, bark and phloem were removed, and the xylem was immediately sealed in glass vials (air-tight tubes, Duran GL-18, Duran Group GmbH, Mainz, Germany). Plant samples from ruderal vegetation were taken in 2012 and 2013 campaigns for the two most representative species within the plantation, i.e. *Conyza bonariensis* and *Lactuca serriola*. In this case, samples were collected from the root crown, as the best proxy for source water in herbaceous species (Barnard et al., 2006; Sánchez-Bragado et al., 2015). No ground cover vegetation samples were collected when the studied species were not entirely green.

Soil cores were extracted with an automatic auger at I and 2 m from the base of each sampled tree, in the east-west direction. Soil samples were taken from the cores at 25, 50 and 100 cm depths for all the sampling dates. Soil samples at I cm depth were taken also on 26th September 2012 to further explore the expected differences between soil types at the uppermost soil layers. Samples were rapidly sealed in glass vials following the same procedure than that for the other samples.

Finally, all the samples collected in the field were placed on dry ice and kept frozen in our laboratory until water extraction and isotopic analysis processing.

Water extraction and isotopic analysis

Water extraction and isotopic analysis were carried out at the Department of Crop and Forest Sciences, Universitat de Lleida (Spain). The extraction of water from the soil and xylem samples was performed by cryogenic vacuum distillation (Dawson and Ehleringer, 1993). The extraction system consisted of 10 sample tubes connected with Ultra-TorrTM fittings (Swagelok Company, Solon, OH, USA) to 10 U-shaped collection tubes specifically designed for this system. The sample tubes were submerged in mineral oil at a constant temperature (110–120°C) to evaporate water and the U-tubes were cooled with liquid nitrogen to condense the water vapour. The extraction system was connected to a vacuum pump (model RV3; Edwards, Bolton, UK) to guarantee the flow of water vapour from the sample tubes to the collection tubes and to prevent contamination with atmospheric water vapour. The entire system maintained constant vacuum pressures of c. 10⁻² mbar.

The stable isotope composition of oxygen and hydrogen in water samples was determined by Cavity Ring-Down Spectroscopy (CRDS), using a Picarro L2120-*i* coupled to a high-precision vaporizer A0211 (Picarro Inc. Sunnyvale, CA, USA) at the Serveis Científico-Tècnics of the Universitat de Lleida (Lleida, Spain). Overall uncertainty (determined as the standard error of repeated analyses of a reference sample not included in the calibration, N=20) was 0.05% and 0.17%, for oxygen and hydrogen, respectively. All isotopic ratios were expressed relative to an international standard (VSMOW, Vienna Standard Mean Ocean Water) in per mil notation (%):

$$\delta^{18}$$
O or δ H = (R_{sample}/R_{standard} -I) x 1000

where R_{sample} and $R_{standard}$ are the heavy to light isotopic ratios of the sample and the standard, respectively.

The potential presence of organic contaminants was checked using the post-processing software Picarro ChemCorrect 1.2.0. In 2012, a new Micro-Combustion ModuleTM for the removal of contaminants was integrated in-line between the Picarro's Vaporizer and the L2120-i water isotope analyser. Therefore, the samples for 2012 and 2013 were analyzed using this module, which removes the contaminants through oxidation in a way that up to ca. 0.5% of organic contaminants only pure water arrives to the analyser. In all cases, we applied the post-processing correction described by

(Martín-Gómez et al., 2015), which has shown to be as effective as the module in correcting the effect of organic contaminants.

Complementary measurements: root density in soil profile and tree transpiration

In each soil type, one squared soil pit of about Im^3 (1.25 m depth) was excavated in June 2013. One of the faces of the pit was divided into volumes of 0.05 m³ (0.25 x 0.20 x I m³), all tree roots of each volume were collected and classified by diameter (<=2 mm or >2 mm) and their fresh and dry weights were obtained. A subsample of fine roots was used to calculate the ratio of dry weight over length, i.e. the specific length (cm/g), in order to estimate the length density for each soil layer (cm cm⁻³).

At the same time, tree transpiration was measured with sap flow sensors (HRM sensors, ICT International, Australia) based on heat pulse method (Burgess et al., 2001). Sensors were placed at 1.3 m height in several samples trees within the plantation, including those sampled for xylem water (as described before). The velocity of the heat pulse emitted by the heater needle was measured with thermocouples placed at 1.25 and 2.75 cm depth from cambium (outer and inner couples of measurements, respectively), and at 5 cm distance up and down from the heater (Burgess et al., 2001). Thermal diffusivity and wood moisture fraction values were estimated from several tree ring cores following Kravka et al. (1999). Correction of the heat pulse velocities was done for probe alignment and the probe-induced effects of wounding (Barret et al., 1995). Once corrected sap flux densities were estimated, tree transpiration for each tree (litres tree⁻¹) was calculated by multiplying the outer band by the sap flux density measured in the outer thermocouples and the inner band by the inner sap flux density measured in the inner thermocouples, and then summing up both.

Results and discussion

Isotopic composition of water samples

 δ^{18} O in rainfall ranged from -11.67 to -0.88 ‰, while δH ranged from -81.24 to -4.31 ‰ (Table 2). The local meteoric water line (LMWL), which reflects the evaporation in the course of precipitation in our study site, was similar to the one observed in Barcelona city, at 42 km northwest of the study area (http://www-naweb.iaea.org/napc/ih/IHS_resources_gnip.html) and also to the global meteoric

water line (Craig, 1961). However, the LMWL showed a lower value for the deuterium excess (d= $\delta H - 8 \delta^{18}O$), i.e. the intercept of the line (7.2 ‰ for LMWL versus 13.4 ‰ and 10 ‰ for the water line in Barcelona and the global one, respectively), indicating that the rainfall in our study site was more evaporated or enriched with heavy isotopes (Figure 1).

Table 2. Isotopic compositions of rainfall, soil water, groundwater and xylem water.

Туре	Sample amount	δ ¹⁸ O δH (‰)							
		Max.	Min.	Mean	SD.	Max.	Min.	Mean	SD.
Rainfall	24	-0.88	-11.67	-5.14	2.71	-4.31	-81.24	-32.57	21.26
Soil water	178	7.53	-7.53	-4.10	2.50	-4.11	-62.06	-37.37	10.05
Groundwater	7	-4.97	-5.43	-5.18	0.18	-31.47	-36.93	-33.38	2.34
Xylem water	54	3.18	-4.89	-2.87	1.52	-3.41	-48.35	-34.14	9.14

Groundwater samples showed very isotopic stable conditions, with $\delta^{18}O$ ranging from -5.43 to -4.97 ‰ and SD = 0.18 ‰, and δ H ranging from -36.93 to -31.47 ‰ and SD = 2.34 ‰ (mean deuterium excess of 8.05 \pm 1.28 ‰), and all their values falling in the LMWL (Fig. 1). In contrast, water isotopes of plant and soil showed more variability. Whereas there were several samples falling close to the LMWL, also high proportion of them fell to the right side of the LMWL, reflecting higher evaporation than that in rainfall water. The wide ranges observed in plant and soil waters will be described in detail in the next sections.

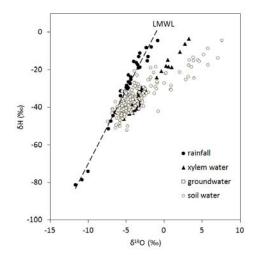


Figure I. Water isotopes ($\delta^{18}O$ and δH) of rainfall, xylem water, groundwater and soil water. The LMWL represents the local meteoric water line ($\delta H = 7.74 \ \delta^{18}O + 7.21$).

Isotopic variability within the soil profiles

Water isotopic composition in both soil types showed, as expected, isotopic enrichment in the shallow soil layers, given the role of summer evaporative demand in the study site, characterized by Mediterranean climatic conditions.

For all the sampling dates and for both soil types, soil evaporation influenced the soil water isotopic signal until a soil depth between 25 and 50 cm, as showed the small δH and $\delta^{18}O$ variations between 50 and 100 cm depth in all soil profiles (Figure 2). Isotopic enrichment was the highest at 1 cm depth (Figure 3). These values in the upper part of the tested soils are in the range observed in other studies carried out in semiarid or arid conditions (Asbjornsen et al., 2007; Chimner and Cooper, 2004; Moreno-Gutierrez et al., 2012). Marked differences in the isotopic stable composition between the studied soils appeared at 1 cm depth, as indicated by the statistically significant differences found and also by the high differences observed in the deuterium excess, that ranges from -51.92 to -8.46 ‰ and from -15.23 to -4.11 ‰ for the forest and agriculture soil, respectively.

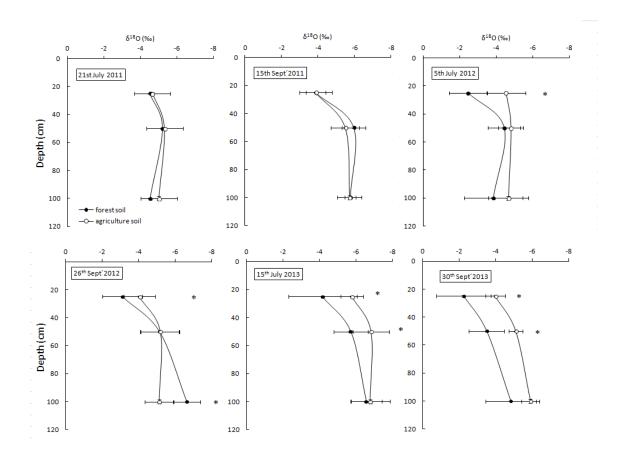


Figure 2. δ^{18} O means and standard deviations of the sampled soil profiles. Note that an additional sampling was carried out at I cm depth on 26th September 2012. * indicates significant differences at p-level <0.05.

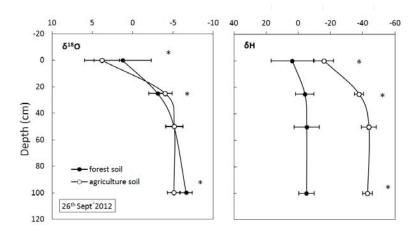


Figure 3. $\delta^{18}O$ and δH means and standard deviations of the sampled soil profiles for the sampling date on 26th September 2012, in which an additional sampling was carried out at 1 cm depth . * indicates significant differences at p-level <0.05.

Significant differences in isotopic composition between soil types were also observed for 4 of the 6 sampling dates at 25 cm depth, 2 dates at 50 cm depth and only one date

at 100 cm depth (Figure 2). Moreover, variability in both δ^2H and $\delta^{18}O$ was always higher in the forest soil, regardless the soil depth (Figure 4).

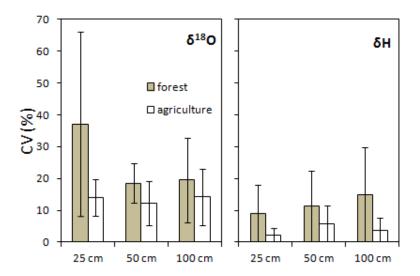


Figure 4. Coefficients of variation (CV) for δ^{18} O and δ H for the studied soils at 25, 50 and 100 cm depth.

The results at I cm depth (Figure 3) showed a wide range in $\delta^{18}O$ and δH in the forest soil, in contrast to the one observed in the agriculture soil. Besides, comparing forest soil isotopic composition at I and 25 cm depth we can observe that for some samples (17% of the total) there was no enrichment in the shallowest part of the profile. This result is in agreement with the work of Barnes and Allison (1983) and Allison et al. (1984) that considering δH variation, described soil profiles divided in two parts, a narrow upper one in which water movement is mainly promoted by vapour diffusion, resulting in very negative δH values, and a lower one with an expected declining of the deuterium enrichment. Therefore, our results seem to indicate that this physical phenomenon is more likely to appear in soils in which evaporation is expected to play a more important role, such as the forest soil.

On the other hand, some results found could be relevant for the assessment of water uptake by plants. First, the higher variability found in the forest soil should be carefully considered, because this variability can complicate the linkages between xylem and soil waters. And second, although similar results have been showed in several studies (e.g. Barnes and Allison, 1988; Burgess et al., 2000; Goldsmith et al., 2012; Querejeta et al., 2007), the homogeneity observed in the δH or $\delta^{18}O$ in depth (from 50 to 100 cm)

should be carefully evaluated, because difficult to assess the possible contribution to xylem water from soil water at these depths.

Water isotopic composition of xylem of trees and spontaneous herbaceous plants and its relation to soil profiles

Figure 5 shows the δ^{18} O values for the trees and spontaneous herbaceous plants xylem samples and the soil samples taken at 25 cm depth. In most of the sampling dates, the wild cherry trees and the ground vegetation had δ^{18} O more positive or similar values than those in soil at 25 cm depth, thus indicating a likely root water uptake from the upper soil layers (Figure 2). The high differences observed between trees and soil δ^{18} O in the first sampling date (21 July 2011 in Figure 5), when the trees were 6 years old, seemed to indicate that trees relied on very superficial water at this young stage. These results are in agreement with those observed by Dupraz et al. (1995) in agroforestry system with wild cherry trees of 4 years old growing under Mediterranean climate conditions. These authors pointed out that trees were extracting water from more superficial soil layers than the herbaceous intercrops Moreover, herbaceous intercrops clearly affected trees transpiration and reduce up to six times tree growth when compared to trees growing without herbaceous intercrops. Regarding our experiment, in some cases the ground vegetation showed more enriched δ^{18} O values than the wild cherry trees, especially *Coniza bonarienses*, but in other cases they presented similar values, which indicated a likely competence for soil water from these upper layers.

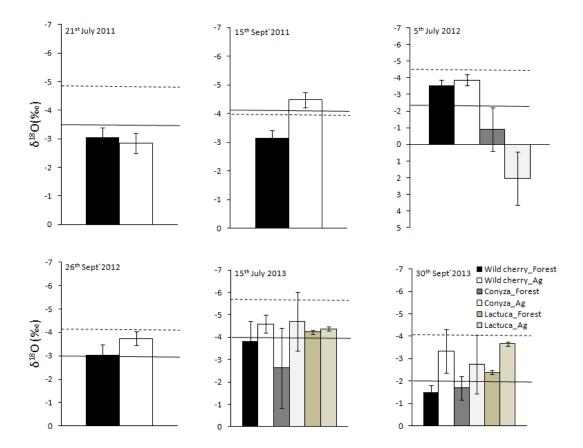


Figure 5. Means and standard deviations of $\delta^{18}O$ values in plant samples for each sampling date and for each soil type (Ag: agriculture soil). Continuous and discontinuous lines indicate the mean $\delta^{18}O$ of soil water at 25 cm depth for the forest and the agriculture soil, respectively.

The direct inference of water uptake by the studied plants by comparing the isotopic signature of the xylem water with soil water at 25 cm depth has been shown to be not suitable in some of the sampling dates. From the plantation management view, this difficult the assessment of the water use competence between the planted trees and the ground vegetation. We may indicate that a possible water use competence have been occurred, although we have not information on the magnitude and temporal extends. For solving these questions a shallow soil sampling (e.g. taking samples every 5cm from 0 to 25 cm) would have been necessary.

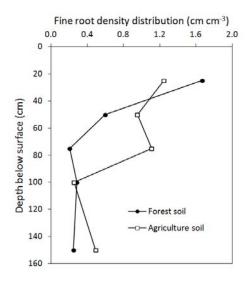


Figure 6. Fine root densities (cm cm⁻³) in the two types of soils studied.

The two soil pits, carried out in 2013, indicated different root fine density distribution along the soil profile between the soil types (Figure 6). Agriculture soil showed homogeneous root distribution from 0 to 80 cm depth (with about 80 % of the total fine roots in these depths) and a decrease in fine root proportion from 80 cm depth on. In contrast, a high concentration of fine roots was observed in the first 30 cm in the forest soil (about 60 %). These results indicated different root developments between the studied soils, although not linked to the ones observed in the water sampling carried out in 15th July 2013 (the closest on time). In both cases, trees relied on surface soil water at this time period, when soil water was a limiting or an almost limiting factor in the forest and the agriculture soil, respectively: relative extractable water during this period was below and close to 0.4 for the forest and the agriculture soil, respectively (Molina et al., submitted); since 0.4 is considered as the threshold for water transpiration for several tree species (Granier et al., 2000). The isotopic data from late summer also indicated that trees relied on surface soil water in this period, in contrast to other results observed in system with trees and herbaceous crops growing together (Link, 2014). In this sense, Link (2014) showed that walnut and poplar trees growing in an agroforestry system mainly relied on water from 10 to 20 cm in early season, while these water uptakes shifted in the late season for both tree species, when trees were mainly taking the water from the soil range depth of 40-70 cm.

Several trees with low or even zero transpiration rates showed high enrichment (as indicated by low deuterium excess (Figure 7). This enrichment was only found in trees

growing in the forest soil, in which the soil water availability along the profile was about two times lower than in the agriculture soil (Table I). These results indicate therefore the limitation of the method for trees growing under high water stress, with high degree of stomatal closure, since one of the most important assumptions when using water isotopes for tracing trees water uptakes was not reached (i.e. the water replacement in tree crown through tree transpiration). This aspect was already pointed out by Dawson and Ehleringer (1993), when considered that the mixture of enriched phloem water and xylem water was probable in young non suberized stems. In addition, Brandes et al. (2007) indicated that a possible explanation of their observed xylem enrichment may be related to the low tree transpiration rate during winter. Therefore, since most of our xylem samples of trees growing in the forest soil were enriched, we can consider probable the effect of the described phenomenon in this plot.

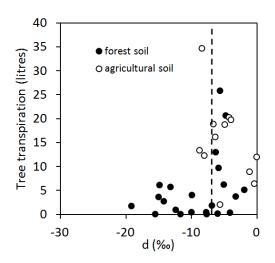


Figure 7. Relationship between Deuterium excess (d= δ H – 8 δ ¹⁸O, ‰) of tree xylem water and cumulated tree transpiration (during the 7 days before sampling campaigns) for each type of soil studied. Discontinuous line indicates the highest d of soil samples at 25 cm soil depth observed along the study period.

Conclusions

In the present study, we took advantage from having two soil types within our experimental wild cherry tree plantation. First, the isotopic composition along the profile between the soil types was compared. Evaporation from the uppermost layers affected to a greater extent in the forest soil, and also higher spatial variability of both water isotopes was observed. The later should be carefully evaluated when water uptake from water isotopes technique is intended to be used in similar soil conditions. On the other hand, the direct inference through direct comparison between waters from plant xylem and from soil samples taken at different depths indicated that trees and spontaneous herbaceous plants were both extracting water from the upper soils

layers. Therefore, water use competence is expected. On the other hand, the complementary information on transpiration allowed us to asses that most of trees with low transpiration rates had their xylem samples enriched in the heavy isotopes. Thus, we hypothesized that the mixture of xylem and phloem waters, leaded by high degree of stomatal closure, represents a clear limitation for the use of the water isotopes technique under high water stress conditions.

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ANNEX

How much does the sapwood area increase along the growth season affect the determination of sap flow in wild cherry trees?

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Annex

How much does the sapwood area increase along the growth season affect the determination of sap flow in wild cherry trees?

Abstract

In sap flux density methods the up scaling from tree level measurements to stand transpiration involves different errors. Most of the studies using these methods consider a unique value for the sapwood area during the whole season. However, for fast-growing tree species, as sapwood normally changes during the growing season, an important bias could appear in the sap flow estimate if the sapwood increment is not considered. This short communication compares the sap flow estimation when considering a monthly value for sapwood area with the sap flow estimations considering 3 values, one obtained at the beginning of growing period, another one at the end and the last one as the average from both, respectively. The average biases (expressed as difference with the estimate with a monthly value for sapwood area) were $49.6 \pm 15.2 \, \text{l}$ tree⁻¹, $38.3 \pm 3.6 \, \text{l}$ tree⁻¹ and $6.3 \pm 2.6 \, \text{l}$ tree⁻¹, respectively. The biases were expressed as a function of the annual diameter increment and R^2 ranged from 76 to 83%. Our findings indicate that ignoring sapwood variation along the growth season in fast-growing species could cause important errors in the sap flow estimate, similar in magnitude to others errors widely studied.

Keywords: sapwood area, heat pulse, sap flow, wood, transpiration, water cycle

Introduction and objectives

Water consumption by trees is a key question for agriculture and forestry, especially in Mediterranean and semiarid areas where water supply is commonly applied to maintain high levels of productivity (Savé et al., 2012). Accurate estimates of transpiration can be done by several different methods (Rana and Katerji, 2000; Wullschleger et al., 1998), including the thermal sap flow methods, in which are included those based on sap flux density measurements (Smith and Allen, 1996).

In sap flow sensors, up-scaling from point measurements to plot scale involves different errors related to, among others, sensor characteristics (James et al., 2002; Nadezdhina et al., 2002), site-dependent factors and their variability (Hatton et al., 1995; Tsuruta et al., 2010), the biometrics used and the sample size (Hatton et al., 1995; Kume et al., 2010). Furthermore, in some cases sap flow is not suitable for estimating transpiration because sap flow underestimate transpiration under conditions of low sap flow (Alarcon et al., 2000). To up-scale to tree level, apart from certain parameters such as wood density or thermal diffusivity, it is necessary to consider a specific sapwood area to convert from sap flux density into sap flow. Most forest studies using sap flow sensors consider a unique value for this scalar, normally estimated as a function of the diameter measured at the beginning, the middle or the end of the study (e.g. Forrester et al., 2010; Poyatos et al., 2007); moreover, in some studies it is not specified when the diameter is measured (Kume et al., 2010).

Fast-growing tree species are becoming more important due to their feasibility in agroforestry systems and forest plantations in terms of wood production (Forrester et al., 2010; Guan et al., 2012). Typical examples of genus used are *Acacia, Pinus, Populus, Eucaliptus, Juglans, Prunus, Paulownia* or *Grevillea*. In these species, even if sapwood area normally changes, sometimes rapidly, during the growing season, only some studies take into account a changing value for the sapwood area (e.g. Guan et al., 2012; Zalesni et al., 2006). In many others a unique value is used and consequently the sap flow is biased and should be corrected (e.g. Forrester et al., 2010; Meiresonne et al., 1999).

This paper examines the bias in the sap flow estimate when considering 3 fixed values for sapwood area (from the beginning of the growing period, "initial", from the end of the growing period, "final", and the average from both, "middle") versus the sap flow

estimate considering a sapwood area that monthly changes. The bias for the entire growing season is presented as a function of the annual diameter increment.

Materials and methods

Study site and overview of the experimental setup

The study was carried out at the Torre Marimon site (Caldes de Montbui, Barcelona, Spain) within the IRTA facilities (41°36 ′47 ′′ N, 2°10′11 ′′E, 170 m.a.s.l), and in the context of an experimental project focused on studying the effects of soil performance, soil tillage and irrigation on growth and tree water relations in a wild cherry tree (*Prunus avium* L.) plantation orientated to timber production. Tree density is 625 trees ha⁻¹ and the spacing between trees and rows are both 4 m. The historical mean \pm standard error values (1999-2012) of rainfall and potential evapotranspiration for the site are 599.4 \pm 33.4 and 846.8 \pm 23.3mm respectively. Soils are basics, have a depth range from 0.5 to 2 m and from sandy-loam to clay-loam textures.

Heat pulse velocity measurements

Heat pulse velocity (cm h⁻¹) was measured by sap flow sensors (HRM, ICT International, Australia) based on the Heat Ratio Method (Burgess et al. 2001). The measurements were recorded every 100 s and averaged over 30 minutes. Each sensor consisted of a heater located 5 mm apart from two pairs of thermocouples located downstream and upstream. Sap flux density is estimated at 12.5 mm and 27.5 mm from the cambium. The sap flow sensors were installed in 12 trees (one sensor per tree), one in each experimental plot, at 1.3 m height and at east aspect (tree characteristics in Table 1).

Table 1. Characteristics of the sampled trees during the 2012 growing season. Height was measured at the end of the growing period and diameter at 1.3 m (DBH), sapwood area and green relative cover are expressed by season-wide mean and standard error values. Green cover was estimated from 2 sky-orientated photos (18 mm lens) regularly taken at 50 cm south and north from each tree.

Irrigation	Soil Tillage	Tree	Height(m)	DBH (cm)	Sapwood area(cm²)	Green cover (%)
Yes	No	I	4.7	6.7 (±0.1)	30.8 (±0.8)	23.7 (±3.4)
Yes	No	2	6.5	8.5 (±0.1)	51.2 (±1.5)	45.2 (±5.3)
Yes	No	3	6.7	9.3 (±0.1)	61.2 (±2.0)	34.2 (±4.1)
No	No	4	4.3	5.5 (±0.03)	19.8 (±0.3)	12.0 (±3.2)
No	No	5	4.9	6.4 (±0.05)	28.3 (±0.5)	18.4 (±3.2)
No	No	6	6.0	7.6 (±0.1)	39.9 (±0.9)	20.8 (±2.3)
Yes	Yes	7	5.1	6.9 (±0.1)	33.3 (±1.1)	31.9 (±4.0)
Yes	Yes	8	6.9	9.9 (±0.2)	69.3 (±2.6)	44.4 (±5.3)
Yes	Yes	9	6.8	9.1 (±0.2)	59.3 (±2.5)	36.3 (±4.5)
No	Yes	10	5.0	6.2 (±0.1)	26.5 (±0.5)	17.1 (±3.4)
No	Yes	11	6.2	8.7 (±0.1)	53.0 (±1.2)	24.7 (±3.1)
No	Yes	12	6.3	9.I (±0.I)	57.9 (±1.4)	39.3 (±4.7)

From heat pulse velocity to sap flux density

First, heat pulse velocity was converted into sap flux density by considering the wood thermal diffusivity and the wood-depending factors of the studied specie, following Burgess et al. (2001). Second, corrections to sap flux density were done as follow. Non parallelism among needles was accomplished by comparing baselines from the last week of measurements, in which negligible sap flow occurred, to baselines from all the measurements. Possible underestimation caused by the probe-induced effects of wounding was assessed from wound width, following Swanson and Whitfield (1981) and using the results for plum trees (*Prunus domestica*) from Fernandez et al. (2006), since there is no wound width data available for wild cherry.

From sap flux density to sap flow: accounting for the sapwood variation along the growing season

In our trees heartwood growth occurs in the dormancy period as in other species in northern temperate climates (Beauchamp et al., 2013), so we reasonably assumed that sapwood depth increase along the growing season corresponded to that observed in diameter (Guan et al., 2012). Stem diameter at sensor height was monthly measured during the study period at the middle of each month. To estimate the sapwood depth from stem diameter, thickness of bark, sapwood and heartwood were measured in

several cores (5 mm increment borer). The differentiation between sapwood and heartwood depths was done based on color change following Nadezdhina et al. (2002).

During the growing season, the radial profile of sap flux density changes, with the maximum value extending outwards as new xylem is formed (Beauchamp et al., 2013). For each month, the sapwood area was divided into two concentric bands (outer and inner band), delimited by the mid-point between the thermocouple location (hereinafter "monthly" estimate; n= 8 for each tree; 8 months of study). The sapwood increment during the month was assigned to the outer thermocouples, i.e. the cross-section area of the outer band was equal to the increment band due to growth of trees around the sensors during the month plus the previous outer band. Sap flux for each tree (litres tree-1) was thus calculated by multiplying the outer band by the sap flux density measured in the outer thermocouples; and the inner band, by the inner sap flux density measured in the inner thermocouples; and then adding both.

The total sap flow calculated with the monthly estimate was compared to three different estimates considering I unique value for sapwood area: a value measured at the beginning of the experiment ("initial"), a value measured at the end of the experiment ("final") and the average from the initial and final values ("middle"). The calculations were done as before, that is, considering the cross sectional sapwood area divided into two concentric bands delimited by the mid-point between the measurements.

The bias was expressed as a function of the diameter increment. The functions were selected based on the explained variance (R²) and the differences between the observed and predicted values, expressed by their Root Mean Square Errors (RMSE) and Mean Absolute Errors (MAE).

All the sap flow calculations were carried out with the Sap Flow Tool Software (version I.3, ICT International, Australia).

Results and discussion

The evolution of the sapwood area during the vegetative period of 2012 is given in Figure I. The highest growth of sapwood occurred during May (6.8 cm² month⁻¹); two weeks after all the leaves were formed, followed at a distance by June growth (1.4 cm² month⁻¹). From July to November, an almost constant growth of ca. 0.8 cm² month⁻¹

was maintained. Figure I also shows the high variability of the sapwood area among the studied trees (mean standard error = 4.7 cm²). The observed, progressive increase in the standard error from May to December (3.5 to 5.3 cm²) was the result of the cumulative effect of both natural soil heterogeneity and the factors tested, all of which affected tree growth.

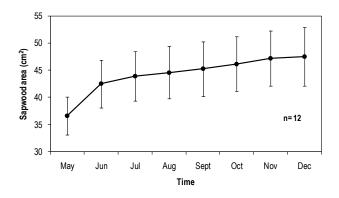


Figure I. Means and standard errors of sapwood area (cm2) during the study period of 2012.

Figure 2 shows an example of the accumulated monthly sap flow estimates for the tree with the highest diameter increment during the study period (tree n° 8 in Table I). The "initial" estimate gave the same sap flow in May than the monthly estimate, because the sapwood area considered was the same. However, as consequence of both a high sap flux density and a marked sapwood area increment in the summer months, the sap flow was largely underestimated (-13.8 % to -19.2 % from June to August). The "middle" estimate gave a high overestimation (15.3 %) in the month of maximum sap flow (May) due to sapwood area considered was much higher than the monthly one. However, from June on there was an underestimation (from -0.5 % to -12.0 %). This temporal pattern produced a compensation of errors in the total sap flow estimated using the middle value, as will be shown for all the studied trees (Figure 2). The "final" estimate overestimated sap flow (from 30.7 % to 12.8 %) mainly during the months of maximum flow (from May to June), but in the remaining period gave similar estimate because the sapwood area slightly changed during this period.

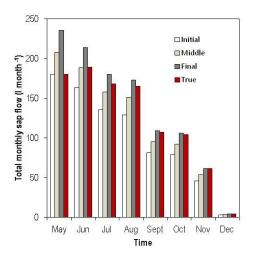


Figure 2. Total monthly sap flow (I month-1) for the tree with the highest diameter increment during the study period (tree n°8 in table 1) calculated using the four estimations of sap flow tested.

Figure 3 compares, for each studied tree, the sap flow estimates using a unique sapwood value, the initial value (Figure 3A), the average value from initial and final ones (Figure 3B) and the final value (Figure 3C), and their corresponding biases respect to the estimate using a monthly measured sapwood area. In general, the higher the sapwood area increment during the study period, the higher the bias. As expected, the better estimate was obtained when the average from the initial and final sapwood areas was used. The mean \pm standard absolute error values of the bias were 49.6 \pm 15.2 I tree⁻¹, 6.3 \pm 2.6 I tree⁻¹ and 38.3 \pm 3.6 I tree⁻¹ (or 11.9, 1.5 and 9.2 % of the mean total sap flow) for "initial", "middle" and "final" estimates, respectively. In the three cases, the bias in total sap flow was regressed against the diameter increment; all the regressions were statistically significant and R² ranged from 0.76 to 0.83. The "middle" estimate gave the best fit, with RMSE and MAE of 4.1 and 3.2 I tree⁻¹, respectively. The "final" estimate gave an overestimation of 11.2 % for the biggest tree.

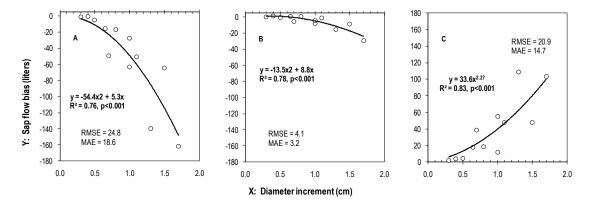


Figure 3. Bias (expressed as difference) between the estimation of total sap flow considering a monthly value of sapwood area and using the initial value (A), the middle value (B) and the final value (C). Regressions in A and B were forced through the origin.

Studies regarding the effect that temporal variation on sapwood area may exert in sap flow are scarce. Although this may be a small source of error for large and old trees (Guan et al., 2012), it could be of high magnitude for young trees of fast-growing species, and this is the contribution of this study. The extrapolation from tree-scale sap flow measurements to stand-scale involves many different sources of error (e.g. Cérmak and Nadezhdina, 1998; Clearwater et al., 1999; Nadezhdina et al., 2002). Radial and azimuthal variations of sap flux density have been two of the topics more deeply studied, due to important biases in the sap flow are expected if they are not considered. In our study, the results indicated that if no variation of sapwood is considered in the sap flow estimate, maximum values of biases of -16.5 % ("initial" estimate) and 11.7 % ("final" estimate) were observed. Our results also shows that considering average sapwood area value from the initial and the final sapwood areas, the error ranged from -2.9 to 3.8 % of total sap flow. However, this small error could change in magnitude and sign if sap flow pattern and/or sapwood growth were different during the study period; hence, the distribution of monthly biases would change and the offset in the total sap flow estimate for the entire growing season as well. Furthermore, none of the calculations using a single sapwood area were able to detect changes in sap flow related with sapwood increase, producing sap flow patterns far from the calculated using the monthly sapwood value (Figure 2).

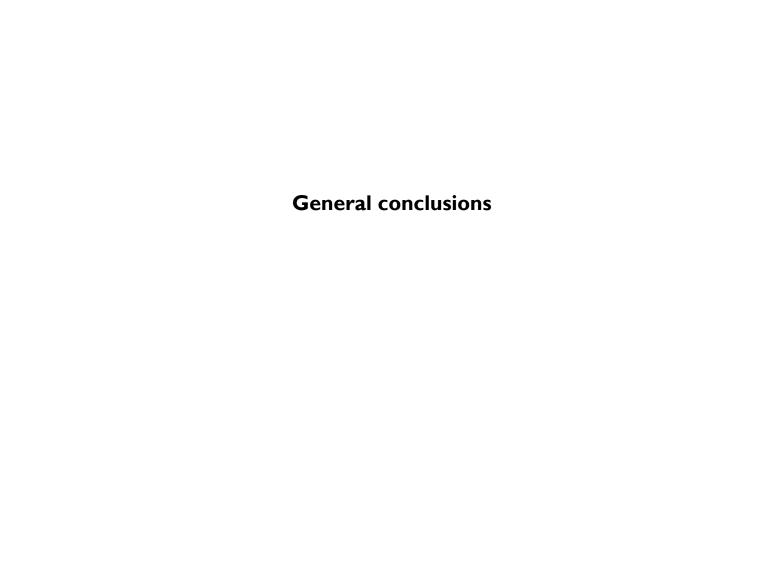
Our findings indicate that ignoring sapwood variation along the growth season in fast-growing specie could cause important error in the sap flow estimate, especially if the evolution of sap flow along the season is of interest. In this sense, the errors observed in this study are in the range of others errors widely described, such as those depending on the scalar or the numbers of trees used when extrapolating from point measurement to stand transpiration (Hatton et al., 1995; Kume et al., 2010).

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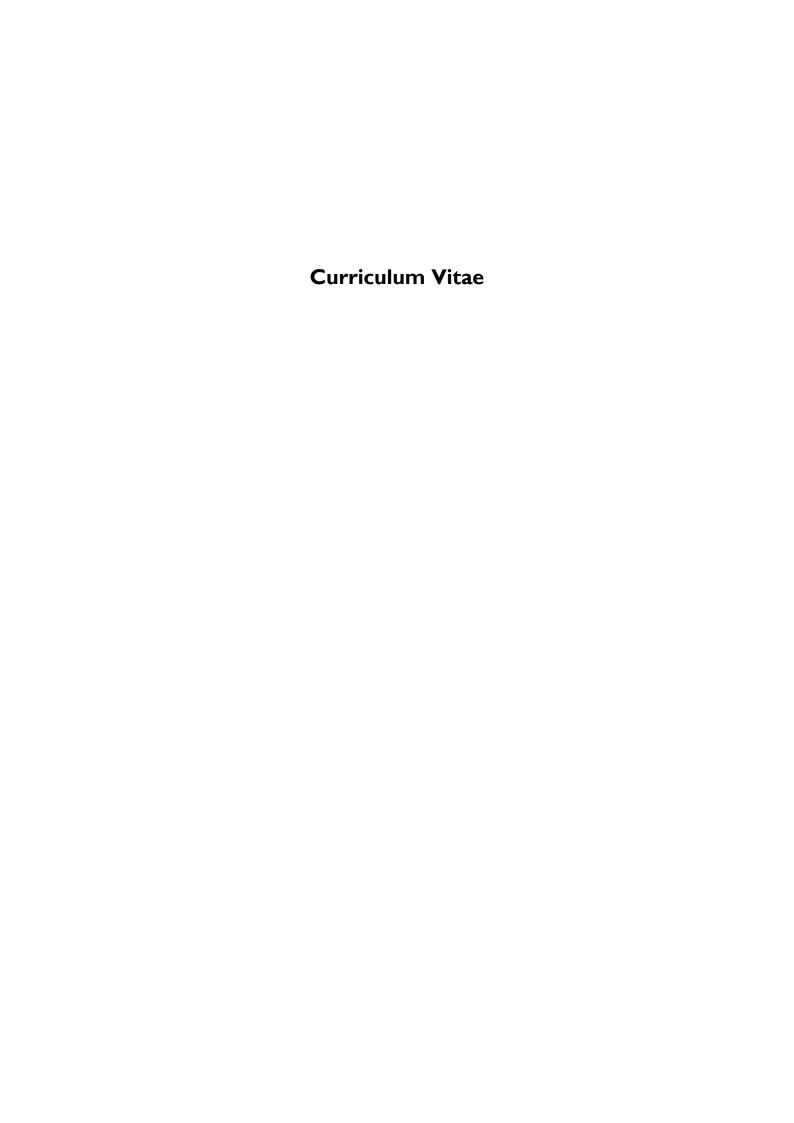


General conclusions

General conclusions

- 1. A good characterization of soil properties prior to the establishment of wild cherry tree plantations is essential, especially of those that are related to the water availability within the soil profile. In soils of low performance, such as the one presented in this thesis, with properties more common of forested areas in Spain, noble plantations can have rotation lengths of about 50 years. Therefore, even though funding for land reforestation are still available, the abovementioned aspect should be carefully valued prior to plantation establishment.
- A reduction of about 10 years in the rotation length can be reached in a soil
 with good performance for agriculture (e.g. in this study with about 100 mm of
 water retention capacity until 100 cm depth) whether irrigation during the
 vegetative period is applied.
- 3. The threshold value of relative extractable water (REW) in which the transpiration of the studied species behaved differently was 0.6 for the 0-50 cm soil profile. A linear response in transpiration was observed in the REW range from 0 to 0.6. REW values higher than 0.6 showed optimal tree transpiration.
- 4. Soil tillage did not have a significant effect on the wood volume increment observed during the three years of study, bringing therefore economical and ecological (as indicated by biodiversity indexes) benefits along. Although the water stable isotopes results suggested that wild cherry tree and the spontaneous vegetation relied on the soil water from the uppermost soil layers in the non-tillaged plots, the non-significant effect on growth seemed to indicate different root water uptake strategies.
- 5. The results on rainfall partitioning in the subplot, where the trees showed optimal growth rates, indicated a low interception loss, in comparison with other forest plantations of similar age and growing at similar conditions. In this sense, although canopy cover was double in the second year of the study, this did not significantly affect throughfall, and as a consequence rainfall interception loss.
- 6. Ten tipping buckets were sufficient to collect the throughfall variability of the studied plantation, and therefore to obtain a mean value for this water flux.

- 7. Branch pruning, carried out at the end of June, reduced tree transpiration approximately in a 30 %. The diameter growth was not affected due to the fact that most of the annual diameter growth has already been reached. This fact suggests that pruning may be considered as an important tool for controlling water use, without affecting growth, in this type of plantation.
- 8. Some methodological aspects to be highlighted:
 - a. Sap flow sensors can be successfully used to estimate the stand transpiration of wild cherry tree plantations.
 - b. Several validated methods to estimate sapwood area are not suitable for wild cherry.
 - c. Important biases can be produced in the transpiration estimates whether the sapwood area increase during vegetative period is not considered. An average value for sapwood area from vegetative period has been shown to be suitable for a proper calculation.
 - d. Water isotopes technique may not be suitable for a sucessful estimation of water uptakes by trees growing under soil water stress conditions.



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Academic Background

- Nov'11- Oct'15. Under the Doctorate programme of "Terrestrial Ecology". Autonomous University of Barcelona (UAB).Barcelona, Spain. Ecohydrological relations in a wild cherry plantation for timber production. PhD thesis supervisors: Pilar Llorens and Carme Biel.
- Dic´10. Master Degree in Hydraulic Engineering and Environment. Polytechnic University of Valencia (UPV). Valencia, Spain.
- Dic'08. Degree in Forest Engineer. University of Cordoba. Cordoba, Spain.
 - Sept'07-Jul'08. Seneca scholarship. Polytechnic University of Valencia (UPV). Valencia, Spain.
 - Sept'05-Jun'06. Socrates-Erasmus scholarship. Gent. Belgium.

Professional Experience

- Nov'11-Nov'15. PhD student in the Institute of Research and Technology Food and Agriculture (IRTA). Caldes de Montbui, Spain. Research scholarship (FPI grant) funded by the Spanish Ministry of Economy and Competitiveness.
- May'11-Oct'11. High qualified technician for research and professional activities in the Spanish Council of Scientific Research (IDAEA-CSIC). Supervised by: Pilar Llorens. Barcelona, Spain.
- o Abr'10-Dic'10. Internship in Vaersa A.S. Valencia, Spain.
- Mar'09-Mar'10. Assistant scholar- UPV. Hydraulic Engineering and Environment Department. Supervised by: Antonio D. Del Campo García. Project: The determination of hydrological and recovery forest canopy factors of the Mediterranean forest and its social perception. Valencia, Spain.
- Sept'08-Oct'08. Scholarship from UPV. Hydraulic Engineering and Environmental Department. Supervised by: Antonio D. Del Campo García. Project: The determination of hydrological and recovery forest canopy factors of the Mediterranean forest and its social perception. Valencia, Spain.
- May'08-Jul'08. Field technician. Institute of Agronomy Research of Valencia (IVIA), Department of Sustainable Development. Valencia, Spain. Supervised by Fernando Pomares.

Supervised Master thesis

- Jan'15-Jul'15. Master Degree Thesis of Giuseppe Carta. University of Barcelona.
- Jan'15-Jul'15. Master Degree Thesis of Elizabet Aguado. University of Barcelona.

- Feb '09-Jun '09. Graduate Degree Thesis (Forestry) of Manuel Franco Torres. UPV.
- Mar'10- Jun'10. Graduate Degree Thesis (Forestry) of Mariela Alarcón Polo. UPV.

Stays in Nationally and Internationally recognized centres

- Oct'14-Dec'14. Stay at the Plant and Food Research Centre. Palmerston North. New Zealand. Supervised by: Brent Clothier. Main objective: to get familiar with SPASMO modelling and to develop a short experiment in an apple plantation.
- 17-21 Feb´14. International Doctoral Course: Stable Isotopes in Forest Ecosystem Research. INRA.
 Nancy. France.
- Sept'13-Dec'13. Stay at the University of Western Australia. Perth. Australia. Supervised by: Pauline Grierson. Main objective: integrative approach to the use of isotopes in ecosystems (from lab to field).
- 20-23 May 09. Training school "Forest and water stress in a changing environment: from cell to ecosystems". Orvieto. Italy.

Publications: SCI Journals

- Antonio Jaime Molina, Ramón Josa, María Teresa Mas, Antoni M.C. Verdu, Pilar Llorens, Xavier Aranda, Robert Savé and Carme Biel., 2016. The role of soil characteristics, soil tillage and drip irrigation in the timber production of a wild cherry orchard under Mediterranean conditions. European Journal of Agronomy, 72: 20-27.
- Antonio Jaime Molina, Xavier Aranda, Giuseppe Carta, Pilar Llorens, Rafael Romero, Robert Savé, Carme Biel. Effect of irrigation on sap flow variability and transpiration estimate in Prunus avium for timber production: azimuthal profile, radial profile and sapwood estimation. Agricultural Water Management.
- Inma Funes, Xavier Aranda, Carmen Biel, Joaquim Carbó, Francesc Camps, Antonio J Molina, Felicidad de Herralde, Beatriz Grau, Robert Savé, Future climate change impacts on apple flowering date in a Mediterranean subbasin Agricultural Water Management. In press in Agricultural Water Management.
- María González-Sanchís, Antonio D. Del Campo, Antonio J. Molina, Tarcísio J.G. Fernandes, 2015.
 Modeling adaptive forest management of a semi-arid MediterraneanAleppo pine plantation. Ecological Modelling, 308:34-44. DOI http://dx.doi.org/10.1016/j.ecolmodel.2015.04.002
- A. Garcia-Prats, Del Campo Antonio, Tarcísio Fernandes J.G, Antonio Molina J., 2015. Development of a Keetch and Byram—Based drought index sensitive to forest management in Mediterranean conditions. Agricultural and Forest Meteorology, 205:40-50. DOI http://dx.doi.org/10.1016/j.agrformet.2015.02.009
- Molina, A.J., Latron, J., Rubio, C.M., Gallart, F., Llorens, P., 2014. Spatio-temporal variability of soil water content on the local scale in a Mediterranean mountain area (Vallcebre, North Eastern Spain). How different spatio-temporal scales reflect mean soil water content. Journal of Hydrology, 514:182-192.
- Del Campo, A., Fernandes, T., Molina, A.J., 2014. 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. DOI 10.1007/s10342-014-0805-7

- Molina, A.J., Del Campo, A., 2012. The effects of experimental thinning on throughfall and stemflow: A contribution towards hydrology-based silviculture in Aleppo pine plantations. Forest Ecology and Management, 269:206-213.
- Molina, A., Del Campo, A., 2011. Leaf area index estimation in a pine plantation with LAI-2000 under direct sunlight conditions: relationship with inventory and hydrologic variables. Forest Systems, 20:108-121.

Chapter books/Non SCI journals

- Molina, A.J., Llorens, P, Aranda, X., Savé, R., Biel, C. 2015. Hidrología y crecimiento de una plantación de cerezo para madera de calidad en el mediterráneo: papel del tipo de suelo, el riego y el laboreo. Cuadernos de la Sociedad Española de Ciencias Forestales, 41: 23-34.
- Garcia-Prats, A., Del Campo-García, A., Molina, A.J.2015. Evaluación de la recarga producida mediante manejo de una masa de Pinus halepensis con técnicas de silvicultura hidrológica. Cuadernos de la Sociedad Española de Ciencias Forestales, 41: 23-34.
- Manrique Alba, A., Del Campo-Garcia, A.D., Fabra-Crespo, M., Molina-Herrera, A.J. 2013. El LiDAR como herramienta para la aplicación de silvicultura hidrológica adaptativa. In: Sociedad Española de Ciencias forestales (Ed.) 6º Congreso Forestal Español, Vitoria 10-14 Jun. Ref. 6CFE01-280.
- Garcia Prats, A., Del Campo Garcia, A., Sanchis González, M., Molina Herrera, A. 2013. Calibración del modelo biome-bgc en condiciones de clima mediterráneo. In: Sociedad Española de Ciencias forestales (Ed.) 6º Congreso Forestal Español, Vitoria 10-14 Jun. Ref. 6CFE01-282.
- González-Sanchis, M., Del Campo, A., Molina, A.J., Fernandes, T.F, 2014. Adaptive water-oriented forest management using BIOME-BGC in Mediterranean Aleppo pine plantations. XXIV IUFRO World Congress 2014, United States, 5-11 October.
- Del Campo, A., Gonzalez-Sanchis, M. Fernandes, T. Molina, A.J., 2014. Bringing water quantification into the management of semiarid forests: a need for implementing adaptive silviculture and watershed services programs XXIV IUFRO World Congress 2014, United States, 5-11 October.
- Ruiz-Pérez, G., Medici, C., Pasquato, M., Gonzalez-Sanchis, M. Molina, A., Fernandes, T., del Campo, A.,
 Francés, F., García Prats, A. 2014. Mediterranean vegetation-water interactions: a model comparison at different scales. XXIV IUFRO World Congress 2014, United States, 5-11 October.
- Llorens, P., García-Estríngana, P., Molina, A.J., Latron, J., Domingo, F., Gallart, F. La interceptación en especies representativas de la montaña mediterránea. Monitorización y modelización de la partición de lluvia por pino silvestre y roble pubescente en las cuencas de investigación de Vallcebre, en Estudios de Interceptación en España. Capítulo de libro editado por Belmonte Serrato y Romero Díaz. IEA. Fundación Instituto Euromediterráneo del Aqua.
- Molina, A. Master Degree thesis. Aproximación al ciclo hidrológico de una masa de *Pinus Halepensis* con diferentes grados de cobertura vegetal. UPV. Spain.

- Mar'15. Member of the Scientific Committee of the third meeting of the Forest Hydrology working group of the Spanish Society of Forest Sciences (Valencia, Spain), and responsible for the presentation:
 - Hydrology and Growth in a wild cherry tree plantation for timber: the role of soil type, irrigation and soil tillage.
- Abr'14. Posters presentation in the EGU General Assembly (Vienna, Austria):
 - Influence of canopy traits on spatio-temporal variability of throughfall in Mediterranean Downy oak and Scots pine stands by Llorens, P., García-Estríngana, P., Latron, J., Molina, A.J., Gallart,
 - Seasonality on the rainfall partitioning of a fast-growing tree plantation under Mediterranean conditions by Molina A.J., Llorens, P., Aranda, X., Savé, R., Biel, C.
- o Abr'13. Posters presentation in the EGU General Assembly (Vienna, Austria):
 - Seasonal and spatial variability of rainfall redistribution under Scots pines and downy oaks in Mediterranean conditions by García-Estríngana, P., Llorens, P., Latron, J., Molina, A.J., Gallart, F.
 - The effects of rainfall partitioning and evapotranspiration on the temporal and spatial variation of soil water content in a Mediterranean agroforestry system" de Biel, C., Molina, A.J., Aranda, X., Llorens, P., Savé, R.
- Sept'12. Oral presentation in the XI Symposium Spain-Portugal for Water Relations in plants, Seville (Spain):
 - Influence of the soil management on the Ecohydrology relationships in an agroforestry plantation:
 Results of the first year.
- Abr'12. Posters presentation in the EGU General Assembly (Vienna, Austria):
 - The effects of rainfall partitioning and evapotranspiration on the temporal and spatial variation of soil water content in a Mediterranean agroforestry system" de Biel, C., Molina, A.J., Aranda, X., Llorens, P., Savé, R.
 - Effects of rainfall partitioning in the seasonal and spatial variability of soil water content in a Mediterranean downy oak forest" de García-Estríngana, P., Latron, J., Molina, A.J., Llorens, P.
- Sept'08. Oral presentation in the second meeting of the Forest Hydrology working group of the Spanish Society of Forest Sciences, Madrid (Spain):
 - Objectives and preliminary results of one hydrology study.