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REPORT

INTRODUCTION and PURPOSE

In the last decade, anomalous drought episodes occurred in different regions worldwide with consequent negative effects on human health and ecosystems functionality (Fouillet et al., 2006; Anderegg et al., 2012). Taking into account the predictions of further increases in temperature, associated with longer and more frequent drought episodes, it is fundamental to develop tools to predict the potential effects of rising temperatures on organisms and ecosystems. Extreme drought events accompanied by heat waves represent a threat to productivity, competitive ability, and survival of trees in several biomes and in different vegetation types (McDowell et al., 2008; Allen et al., 2010). Forest decline and mortality imply rapid and large-scale ecological impacts, as well as severe social and economic

consequences, especially in already threatened Mediterranean-climate areas. The identification of species that can survive and grow well despite periodic droughts is critical to the future success of forest protection and management.

Tree resistance to water stress is conferred by different physiological and morpho-anatomical traits. The drought-induced canopy die-back, tree decline, and subsequent mortality are thought to be mainly triggered by 'hydraulic failure' i.e. extensive xylem embolism formation which fully compromise root-to-leaf water transport (Nardini et al., 2013). The maintenance of functional xylem conduits is necessary to sustain gas exchange at leaf level as well as photosynthetic activity, and thus for plant survival under moderate or severe water deficit conditions.

The first objective of the present COST STSM was to quantify the vulnerability against drought induced embolism (cavitation resistance) of 12 Mediterranean woody species (trees and shrubs) in order to identify the most resistant species to survive under prolonged drought conditions. The second objective was to test the reliability of wood density as a key trait to be used as an index of species-specific vulnerability to extreme drought episodes.

MATERIALS AND METHODS

Vulnerability curves and measurements of wood density were performed on four-years-old potted plants of 12 different species (Table 1). The selected species are evergreen/deciduous trees or sub-shrubs native to the Mediterranean region (Pignatti, 2002).

Vulnerability curves (VCs) were elaborated using the Cavitron technique (Cochard et al. 2005, Fig. 1). The method is based on a modified centrifuge and allows elaboration of VCs of stem segments through repeated measurements of the hydraulic conductance (K) of excised xylem segments (generally twigs) while exposed to negative pressures.

Potted plants were abundantly watered and left overnight covered with a black plastic bag in order to allow full hydration and refilling of embolized conduits. The main stem was cut under water and re-cut several times using shears and razor blades, perfused at high pressure (1.2 bar for 30 minutes at both ends) with filtered distilled water, girdled at both ends, and inserted into the rotor of the centrifuge (length = 28 cm). The centrifugal force was

applied to generate negative pressure in the stem (ψ) and to force water through the sample. The pressure was progressively lowered and the variation of sample hydraulic conductance measured (K). The percent loss of hydraulic conductivity was calculated as PLC = (1-K/Kmax) × 100.

Species	**Family**
Arbutus unedo L.	Ericaceae
Cotinus coggygria Scop.	Anacardiaceae
Cistus salviifolius L.	Cistaceae
Emerus major Mill.	Fabaceae
Ligustrum vulgare L.	Oleaceae
Phillyrea angustifolia L.	Oleaceae
Pistacia lentiscus L.	Anacardiaceae
Prunus mahaleb L.	Rosaceae
Paliurus spina-christi Mill.	Rhamnaceae
Prunus spinosa L.	Rosaceae
Spartium junceum L.	Fabaceae
Salvia officinalis L.	Lamiaceae

Table 1- List of the 12 Mediterranean studied species, and relative families.

Wood density of all stem segments used for hydraulic measurements were determined using a water displacement method (Hughes, 2005). A 2-3 cm-long segment was cut from the proximal end of each sample and left overnight immersed in water. After bark removal, the sample fresh volume (V) was measured by water displacement, the dry mass (DM) was determined (24h at 70°C), and wood density calculated as ρW = DM /V.

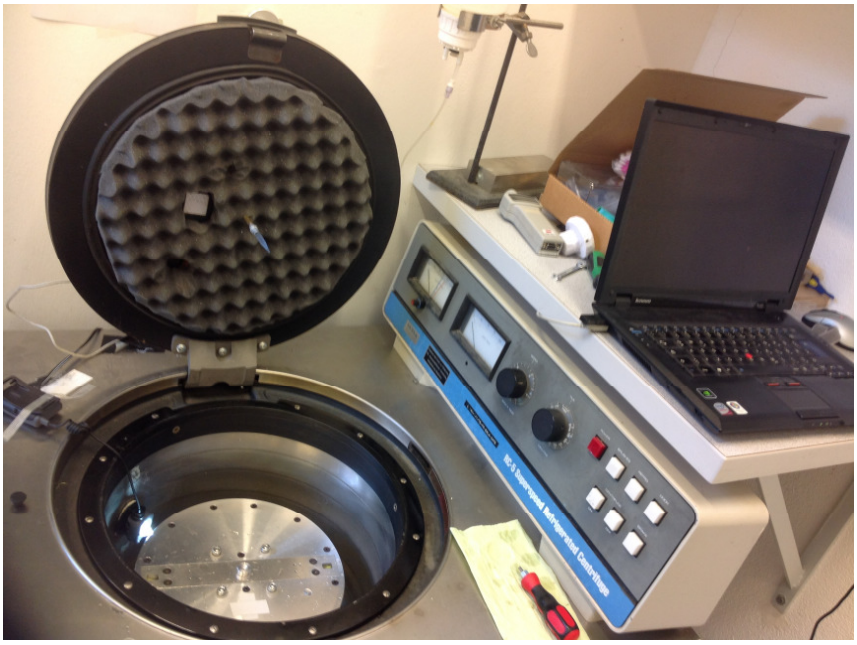

Figure 1- Cavitron with big rotor (diameter = 28 cm)

MAIN RESULTS CONNECTED TO THE ACTION AIMS

At least four hydraulic experiments per plant species were performed including approximately 18 PLC and relative Ψ measurements ranging between 0 and -13 MPa. All curves belonging to the same species were plotted together. The relationship between Ψ and PLC was elaborated to extrapolate the reference values ψ_{12}, ψ_{50}, and ψ_{88}, i.e. the xylem water potential inducing 12, 50, and 88% PLC, respectively. These physiological parameters reflect species' resistance against drought induced cavitation, and the more negative they are, the lower is the species´ vulnerability against drought induced xylem dysfunction (Fig. 2).

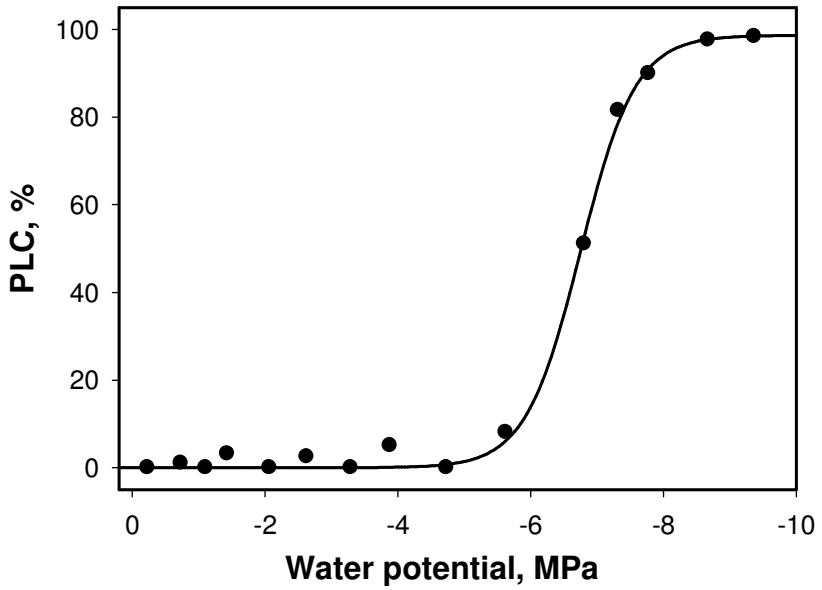

Figure 2- The vulnerability curve (VC) with a typical sigmoidal shape.

It was not possible to successfully extrapolate the ψ_{12}, ψ_{50}, and ψ_{88} for some studied species due to their extremely low stem hydraulic conductivity and resins production. Overall, the observed ψ_{50} values ranged between -4.94 MPa and -11.43 MPa suggesting a generally high resistance against drought induced embolism of the selected species. A different evidence of drought tolerance was observed among species co-occurring in the same habitat: the hydraulic safety seems to be lower in deciduous plants (for example *P. spinosa*) and higher for evergreen plants (for example *P. angustifolia*). In contrast with other studies (Hacke et al., 2001; Jacobsen et al., 2007), no statistically significant correlation was found between Ψ_{50} and wood density (Fig. 3).

The results of the present COST STSM open insights into possible future effect of the global warming on Mediterranean and semi-Mediterranean vegetation communities. The collected data will help to set up a database on species-specific resistance against drought induced embolism of Mediterranean vegetation improving our ability to predict which tree species are most exposed to drought induced mortality risk. Indeed, the loss of water transport capacity is known to impact stomatal aperture, and hence gas exchange rates and photosynthesis. As a consequence, the carbohydrate reserves of plants are depleted, embolism refilling/repair is impaired, and the plant is exposed to desiccation and death (McDowell et

al., 2008; Anderegg et al., 2012; Nardini et al., 2013). Hence, data regarding species-specific hydraulic safety margins are urgent, because they represent valuable indicators of species´ future distributions, including their probability of local extinction.

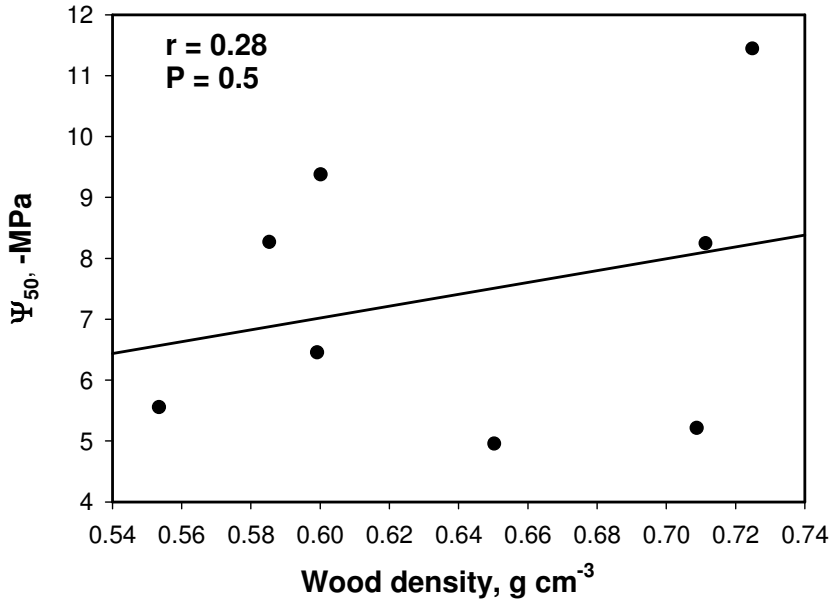

Figure 3- Correlation between wood density and xylem water potential inducing 50% loss of conductivity (Ψ_{50}). Correlation coefficient and P-value are also reported.

Future efforts have to be addressed to the study of the vulnerability to cavitation of a wider number of Mediterranean shrub and tree species. On the basis of our results, the Cavitron technique seems to be unsuitable for assessing vulnerability to cavitation in several angiosperm species, probably due to their anatomical and physiological features. In this light, other techniques should be used for the future screening, i.e. the bench dehydration technique (Sperry et al., 1988). In our case, we are planning to perform VCs of the 12 studied species with the dehydration technique in spring 2015, before a paper will be written. The study of other physiological and morpho-anatomical key traits correlated to the cavitation resistance of species is fundamental in order to identify easily measurable parameters to be used as index of species-specific vulnerability to extreme drought episodes.

OTHER COMMENTS

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