



Influence of saline drip-irrigation on fine root and sap-flow densities of two mature olive varieties

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ABSTRACT

Salt stress is known to influence water use and carbon allocation in trees; however, information about the effects of salt exposure on water uptake and below-ground carbon investment is scant, especially for adult trees. Consequently, this study examined these variables in two mature olive varieties (*Olea europaea* L.) that differ in their NaCl tolerance: Barnea (tolerant) and Proline (sensitive). Trees were irrigated using water with electrical conductivities of 1.2, 4.2 (both varieties) and 7.5 dS m⁻¹ (Barnea only) for 11 years. At each treatment level, we measured soil properties, root morphology, root biomass:necromass ratio, root xylem sap osmolality, and root sap-flow as well as leaf conductance and morphology. Both varieties exhibited reduced fine root biomass under salinity which was only partially compensated for by higher specific root areas under moderate salinity. Proline variety exhibited a smaller fine root system under moderate salinity than Barnea trees, likely causing the lower sap-flow density in coarse roots of Proline compared to Barnea. The higher biomass:necromass ratio of the Barnea root system under moderate salinity is indicative of lower root turnover rates and thus a more efficient carbon use than in Proline trees. Besides differences in ion exclusion capacities, the ability of the fine root system to resist the deleterious effects of salinity seems to affect the salt resistance of mature olive varieties by influencing water uptake and carbon allocation.

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1. Introduction

Water scarcity is a world-wide problem of increasing severity, limiting agricultural development especially in arid and semi-arid areas (Chartzoulakis, 2005). In order to overcome this shortage, the use of low quality-water (as saline ground water) is of increasing importance in Mediterranean climates. However, the use of saline irrigation water as well as freshwater irrigation in combination with high rates of evaporation can affect soil quality, plant growth and ultimately yield (Beltran, 1999).

Soil salinity causes several challenges for plants including water scarcity (osmotic effect) and accumulation of Na⁺ and Cl⁻ ions to potentially toxic levels (ion-specific effect). Both of these factors can cause low stomatal and mesophyll conductances which

were thought to be the main limitations of photosynthesis in olive (Loreto et al., 2003). Furthermore, photosynthesis and growth are largely determined by plants ability to take up water (Nardini et al., 2006). Thus it is essential to understand factors influencing water uptake and carbon allocation under salinity.

Physiological mechanisms that plants use to cope with excess salt include salt exclusion (Walker et al., 1984), excretion and compartmentation (Drennan and Pammenter, 1982; Verslues et al., 2006), as well as active osmotic adjustment (Tattini et al., 2006). Reduced water uptake by Mediterranean sclerophylls is regarded advantageous by reducing Na⁺ transport to above-ground organs (Cimato et al., 2010 and references therein). Shoot growth of olive seedlings was found to be more sensitive to salinity than is root growth, resulting in higher root:shoot ratios (Perica et al., 2008; Cimato et al., 2010 and references therein). However, no studies have examined this among adult trees although ontogenetic differences in salt responses are likely (e.g., Marin et al., 1995). Changes in fine root biomass as well as in root conductivities (Rewald et al., 2011a) and root morphology can have large effects on water uptake (Gregory, 2006), while changes in root turnover rates will have an impact on the overall carbon budget of trees (Bloomfield et al., 1996). Reduced water uptake capacities by means of reduced

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fine root biomass/surface area might be especially unfavourable in arid ecosystems possessing high temperature and irradiance levels throughout the year. These reduced capacities can even have carryover effects during periods of stress relief (i.e. after rainfall or additional irrigation for salt leaching; Cimato et al., 2010).

The cultivation of olive (*Olea europaea* L.) for fruit and oil production is continuously being extended across Mediterranean countries. Although this expansion is facilitated by the general drought and salt-tolerance of this species, olive varieties differ in their sensitivity to salt (Gucci et al., 1997; Wiesman, 2009). Olive varieties with an improved salt-tolerance have been bred and selected for sustainable agriculture under high salt conditions. For example, mature trees of Barnea variety growing in the Negev desert are known to sustain stem growth and yield under saline irrigation as high as 7.5 dS m^{-1} (Weissbein et al., 2008; Rewald et al., 2011a). However, the mechanisms underlying salt tolerance in adult trees *in situ* remain largely unknown. Previous research on olive salt-tolerance has focused on rather short-term above-ground organ and tissue damage of seedlings growing under controlled conditions, i.e. avoiding interactive stresses like high radiation and temperature which olive plants face under natural conditions (Cimato et al., 2010). Thus, beside the documented difference in ion exclusion capacities, information on other traits that account for rootstocks' salt-tolerance and subsequently sustained above-ground growth and yield remain scarce (e.g., Ben-Ahmed et al., 2006; Tabatabaei, 2006).

In the current paper, we present the results of a long-term study on two differently salt-tolerant varieties of mature olive trees subjected to salinity in the Negev desert. The study examined whether salinity influenced (i) fine root density and morphology and subsequent root water uptake rates, (ii) fine root biomass:necromass ratio with implications for carbon allocation, and (iii) whether these traits differ between olive varieties. We hypothesized that salt resistance in mature olive trees growing under high evaporative demand is related to the ability to sustain sufficient-sized fine root systems for ample water uptake at reasonable carbon costs.

2. Materials and methods

2.1. Study site and varieties

The study was conducted between November 2008 and January 2009 in the Ramat Negev Experimental Station located in the Negev Desert, Israel ($31^{\circ}05'00''\text{N}$, $34^{\circ}41'03''\text{E}$; 305 m a.s.l.). The average annual precipitation in this area is 70–90 mm (November–April). Daily temperatures varied between 35°C (July–August) and 5.5°C (January). The soil type of this region is Typic Torrifluent derived from loess, with a clay content of 6–8% ($\text{pH}_{\text{H}_2\text{O}}$: 6.6–9.0). Soil moisture levels (at 15 cm distance to the drip line and soil depth of 0–20 cm) ranged between 25 and 28 vol.% equal field capacity (Oron et al., 1999). Soil moisture was similar in all plots during the measurement period.

Rooted cuttings of olive (*Olea europaea* L.) varieties Barnea and Proline were planted in 1997. Barnea is an olive variety originating from native olive trees grown in the western Negev Desert, Israel (Wiesman, 2009). Barnea trees are large and high yielding. Proline is an olive genotype member of the Israeli olive germplasm collection from unknown origin (Wiesman, 2009). The trees are average in size and producing high-quality olives. Previous studies on stem increment (Rewald et al., 2011a) and fruit yield measurements (Weissbein et al., 2008) determined that Barnea variety possessed an higher 'horticultural tolerance' (see Cimato et al., 2010) to salinity than Proline trees.

The experimental orchard was divided into two plots. The first plot contained five individuals of both varieties per treatment, planted in rows 4 m apart with 6 m between rows. Since

established, plants were either irrigated with freshwater (Control; electrical conductivity, EC_{W} 1.2 dS m^{-1}), or moderately saline water (NaCl, EC_{W} 4.2 dS m^{-1} , moderate salinity). The second plot with the same soil characteristics and spatial arrangements (approx. 150 m apart), contained five Barnea trees that were irrigated with severe saline water (NaCl, EC_{W} 7.5 dS m^{-1}). During the first 3 years after planting, plants received additional irrigation following any rainfall event to prevent excessive salinization of the root zone. Most olive seedlings developed well for the first 2 years and gradually started to be productive from the third year, according to the variety (Weissbein et al., 2008). However, Proline seedlings could not be established under severe saline irrigation.

The saline water originated from local wells and was adjusted by mixing with freshwater or NaCl. The different mixtures of water used for irrigation did not differ in any parameter of relevance apart from NaCl content. The olive trees were drip-irrigated (50 cm distance between emitters); 656 mm of water were supplied per year (see Wiesman et al., 2004 for details). Usually twice a year (March/November), supplements of 100 mm water were added in order to leach salt excesses from the rhizosphere; however, during or directly before the experimental period neither soil leaching nor precipitation events occurred.

2.2. Specific leaf area and leaf conductance

One hundred full-grown leaves (50 sun- and 50 shade-leaves) per variety and treatment were harvested of five tree individuals in early January 2009, scanned with the computer program WinFolia 2005b (Régent, Québec, Canada) and weighed after drying (70°C , 48 h) to determine the specific leaf area (SLA; $\text{cm}^2 \text{ g d. wt.}^{-1}$). Leaf conductance ($\text{mmol m}^{-2} \text{ s}^{-1}$) was measured one sunny day in December 2008 between 9:30 and 13:00 h (EET) on 25 leaves (five leaves per tree) per variety and treatment (Leaf Porometer; Decagon, Pullman, USA). During the measurement period, air temperature (in 6 m height) was $13.2 \pm 0.1^{\circ}\text{C}$, VPD was $0.27 \pm 0.01 \text{ kPa}$ and global radiation was $452 \pm 30 \text{ W m}^{-2}$ (mean \pm SE). Measurements were alternated among treatment groups to avoid systematic temperature effects (T_{leaf} : $14.8\text{--}20.9^{\circ}\text{C}$).

2.3. Root xylem sap osmolality

In January 2009, ten root-branches per variety and treatment (i.e. two roots per tree individual) were carefully collected in the field at distances of 30–50 cm from the drip line; each sample procedure was performed between 11:00 and 13:00 h (EET). Between 60 and 120 min following excavation, xylem sap was extracted from 15 to 25 cm-long root-branches by using a pressure chamber (Arimad 3000; mrc, Holon, Israel; Goodger et al., 2005). About 3–5 mm of the cortex was removed from the apical end of the root segment to prevent xylem sap contamination. After increasing the pressure and discarding the initial drop of sap, the pressure was increased further to collect xylem sap. Sap samples (10 μl) were absorbed with discs of filter paper and the osmolality of these discs was determined immediately with an osmometer (Vapro 5520; Wescor, Logan, USA).

2.4. Root biomass and necromass

In December 2008, 12 soil cores (auger inner diameter $d_i = 5 \text{ cm}$, down to a depth of 50 cm in 10 cm sections) per cultivar and treatment were taken according to a predetermined sampling scheme (Fig. 1). We sampled roots down to 50 cm deep between two tree individuals per variety and treatment; the sampling scheme was based on results from a preliminary study on the horizontal and ver-

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