WATER RELATIONS AND PHOTOSYNTHESIS AS CRITERIA FOR ADEQUATE IRRIGATION MANAGEMENT IN 'TAHITI' LIME TREES

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ABSTRACT: Irrigation scheduling based on soil moisture status is one of the most useful methods because of its practicality and low cost. The effects of available soil water depletion on evapotranspiration (*ETc*), transpiration (*E*), leaf water potential at predawn (Ψ_P) and midday (Ψ_M), stomatal conductance (*gs*) and net CO_2 assimilation (*A*) in lime 'Tahiti' trees (*Citrus latifolia*) were evaluated to improve irrigation schedule and minimize water use without causing water stress. The trees were spaced 7 × 4 m and drip-irrigated by four drippers with the available soil water content (*AWC*) depleted by suspension of irrigation (40 days). Leaf water potential was measured on a pressure chamber (Ψ_P and Ψ_M) and leaf gas exchange was measured by infrared gas analyzer (*E*, *gs* and *A*). Evapotranspiration was determined with the aid of weighing lysimeter. Water soil content and potential (Ψ_S) were monitored with TDR probes and tensiometers, respectively, installed at 0.3, 0.6 and 0.9 m depths. Meteorological variables were monitored with an automatic weather station in the experimental area. The threshold AWC level for the onset of *ETc* decline was 43%, and 60% for *gs, A, E* and Ψ_P. Also, Ψ_P was more sensitive to *AWC* than Ψ_M, and is therefore a better tool for irrigation. When *AWC* was around 60%, values of Ψ_P and Ψs were -0.62 MPa and -48.8 kPa, respectively.
Key words: evapotranspiration, citrus, lysimeter, gas exchange, tensiometer

RELAÇÕES HÍDRICAS E FOTOSSÍNTESE COMO CRITÉRIOS PARA MANEJO ADEQUADO DA IRRIGAÇÃO EM PLANTAS DE LIMEIRA 'TAHITI'

RESUMO: Programar práticas de irrigação com base na umidade do solo é um dos métodos mais usuais devido sua praticidade e baixo custo. O efeito do esgotamento da água disponível do solo sobre a evapotranspiração (*ETc*), transpiração (*E*), potencial de água na folha ao amanhecer (Ψ_P) e ao meio-dia (Ψ_M), condutância estomática (*gs*) e assimilação líquida de CO_2 (*A*) em plantas de limeira 'Tahiti' (*Citrus latifolia*) foi avaliado para melhorar o manejo da irrigação, minimizando água e evitando o estresse hídrico. As plantas foram espaçadas de 7 × 4 m e irrigadas por quatro gotejadores com a diminuição da água disponível no solo (*AD*) provocada pela suspensão da irrigação (40 dias). Para as medidas do potencial da água na folha foi utilizado uma câmara de pressão (Ψ_P e Ψ_M) e para as medidas de trocas gasosas um analisador de gases por infravermelho (*A, E* e *gs*). Foi utilizado um lisímetro de pesagem para a determinação da *ETc*. A umidade e potencial de água no solo (Ψ_S) foram monitoradas por sondas de TDR e tensiômetros, respectivamente, instalados a 0,3, 0,6 e 0,9 m de profundidade. As variáveis meteorológicas foram monitorados por uma estação agrometeorológica automática na área. O limite de *AD* na qual a *ETc* começou a diminuir foi de 43%, ao passo que para *gs*, *A*, *E* e Ψ_P foi de 60%. Ainda, o Ψ_P foi mais sensível a *AD* do que as medidas de Ψ_M podendo ser recomendado como ferramenta para manejo de irrigação. Quando *AD* estava próximo de 60%, os valores de Ψ_P e Ψ_S eram -0,62 MPa e -48,8 kPa, respectivamente.
Palavras-chave: evapotrasnpiração, citros, lisímetro, trocas gasosas, tensiômetro

INTRODUCTION

Increasing world water scarcity and irrigation costs demand developing irrigation methods that minimize water use (Jones, 2004). Localized systems such as microsprinkler and drip irrigation play a major role in reducing the amount of water applied to agricultural crops (Folegatti et al., 2004). However, the maximum water use efficiency depends on adequate irrigation scheduling.

Irrigation scheduling based on soil moisture status is one of the most useful methods because of its practicality and low cost. The principle is that soil moisture can be decreased to a threshold in which water can no longer be transported quickly enough to the roots to respond to transpirational demand, triggering crop water stress (Allen et al., 1998).

A well-known mechanism to prevent plant dehydration under environmental constraint is the stomatal closure (Hall et al., 1975; Syvertsen, 1982; Savé et al., 1995). This response affects both transpiration and photosynthesis of citrus species in different degrees, depending on water stress level (Vu & Yelenosky, 1988; Medina et al., 1998; 1999).

The threshold level of available soil water (*SWA*) that can be depleted is usually determinated by comparing the long-term relationship between yield or tree development and soil moisture levels for a particular location. Moisture levels can be more accurately assessed using plant physiological characteristics such as stomatal conductance, net CO_2 assimilation, leaf water potential and fruit or trunk growth (Davies & Albrigo, 1994). However, few experimental studies use these approaches under field conditions and focus on irrigation scheduling. The purpose of this work was establishing an adequate threshold for available soil water using physiological characteristics to minimize water use in 'Tahiti' lime trees.

MATERIAL AND METHODS

Experimental area

The experiment was carried for 40 days at the end of the winter season, 2004, in a 1.0-ha plot planted in 2001 with 'Tahiti' acid lime trees (*Citrus latifolia* Tanaka), grafted on 'Swingle' [*Poncirus trifoliata* (L.) Raf. × *Citrus paradisi* Macf.] citrumelo rootstock. Trees were spaced 7 × 4 m and drip-irrigated by four, pressure-compensated drippers each, flow rate 4 L h^{-1}, emission uniformity 95%. The irrigation was automatically controlled by a head unit programmer and electro-hydraulic valves. The fraction of wetted area in relation to tree canopy cover at a 0.3 m depth under 3 h of irrigation was around 22%.

The orchard was located in Piracicaba, São Paulo State, Brazil (22º41'58''S, 47º38'42''W; elevation 511 m). Average annual temperature in the area is 21.4ºC and annual rainfall is 1257 mm. Along the study period, the rainfall was 8.5 mm, and average air temperature, relative humidity and reference evapotranspiration were 22.2°C, 61% and 3.9 mm, respectively. The soil is a Rhodic Kandiudalf, clay texture, 5% average slope. Available water capacity was 125 mm m^{-1}, and the bulk density was 1300 kg m^{-3} determined over soil samples collected every 0.2 m down to 1.0 m deep.

Orchard floor was kept cleaned during the experimental period. Ordinary pest control practices were performed and the fertilization was done as recommend by Raij et al. (1992).

Lysimetric and plant measurements

A weighing lysimeter (4 m × 1.3 m depth) (Campeche, 2002) containing one tree, was located near the center of the experimental area. The lysimeter was repacked with soil during installation to equalize bulk densities to the surrounding field. Weight variations were transmitted through three electronic cells to a data logger recording and storing the load cell output every at night time (24h00) when calm conditions prevailed (average wind <1.5 m s^{-1}). Combined calibration and voltage reading errors elicited 0.82 kg accuracy. Daily crop evapotranspiration (*ETc*) was calculated from changes in lysimeter weight and converted to mm d^{-1}, based on lysimeter surface area (12.56 m^2). The lysimeter was irrigated and managed like all plants in the area.

In the experimental plot, six trees with similar structure and development were chosen and divided into two groups (treatments): irrigated (*X*), where daily irrigation based on evapotranspiration was applied during the whole experiment; and non-irrigated (*Y*) where irrigation was suspended. Plant growth measurements – tree height, trunk circumference, and canopy diameter – were taken on each treatment (Table 1). Canopy volume was calculated using the procedure of Hutchinson (1977).

Estimation of available water and soil water potential

Measurements of soil volumetric water content in *X* and *Y* trees were made with a TDR cable tester (model 1502 C Tektronix, Beaverton, OR, USA) every three days.

Table 1 - Characteristics of lysimeter-grown tree, irrigated and non-irrigated 'Tahiti' lime trees before starting the experimental period.

Plant	Height	Canopy diameter	Canopy volume	Trunk circumference
	------------------- m -------------------		m^3	m
Irrigated (X)[1]	3.25 ± 0.23	3.59 ± 0.06	10.47 ± 0.62	0.35 ± 0.02
Non-irrigated (Y)[1]	3.32 ± 0.13	3.63 ± 0.08	10.90 ± 0.71	0.34 ± 0.03
Lysimeter	3.05	3.60	9.88	0.32

[1]Mean ± SE (n = 3).

Three-wire TDR probes were installed 0.3, 0.6 and 0.9 m deep in the soil, and at 1.0 m laterally from trunk of trees, under wetted dripper area. The dielectric constant (*Ka*) of the soil was converted to soil volumetric water content by a calibration obtained for this soil (Tommaselli & Bacchi, 2001). Soil water potential (Ψs) was monitored with tensiometers set 0.6 m deep (n = 3). Daily variations in available soil water in lysimeter-grown were monitored by water potential sensors (Watermark® model 253-L; Campbell Scientific, Logan, UT, USA) connected to the lysimeter data logger, at the same depth of TDR sensors. The soil water potentials were converted to soil volumetric water content by the Soil Water Retention Curve fitting software (Van Genuchten, 1980).

The available soil water content (*SWA*) was calculated with the aid of the equation:

$$SWA = \left[\frac{(\theta a - \theta pwp)}{(\theta fc - \theta pwp)} \right] \times 100$$

where: *SWA*= available soil water content, %, θ_{fc} = soil volumetric water content at field capacity, $m^3\ m^{-3}$ (10 kPa), θ_{pwp} = soil volumetric water content at the permanent wilting point, $m^3\ m^{-3}$ (1500 kPa), θ_a = average soil volumetric water content (between 0.3, 0.6 and 0.9 m depth), $m^3\ m^{-3}$

Leaf water potential measurements

Leaf water potential was measured every three days at predawn (Ψ_P; 06h00-06h30) and at noon (Ψ_M; 12h00-12h30) with a Scholander-type pressure chamber (model 3005; Soil Moisture Equipment Corporation, Santa Barbara, CA, USA). At each measurement time, two stems (four to eight leaves) per tree were sampled by excision in the northward portion of the canopy, at about 1.5 m of the soil surface.

Leaf gas exchange measurements

Two, fully-expanded, similar leaves at external canopy positions per tree per treatment, were sampled for gas exchange measurements. Net CO_2 assimilation (*A*), transpiration (*E*), and stomatal conductance (*gs*) were determined every three days at 09h30-10h30 with a portable photosynthesis system (IRGA, model LI-6400, Li-Cor, Lincoln, NE, USA) on the same days that soil and leaf water potentials were determined. Incident photosynthetic photon flux density (*PPFD*) was fixed based on environmental conditions just prior to the beginning of measurements (Figure 2A), using an artificial quartz halide light source (LI-6400-02 LED light source, Li-Cor) controlled with a quantum sensor inside the leaf cuvette. Air CO_2 concentration was fixed at 350 ±10 μmol mol^{-1}. The actual Water Use Efficiency (*WUE*) was calculated as *A/E* (Machado et al., 1999).

Determining the available soil water and its threshold level

The relationship between (*ETc*) and the reference evapotranspiration (*ETo*) was determined before the beginning of the experiment. Both *ETo* and meteorological variables were obtained from an automatic weather station (model CR21x; Campbell Scientific, Logan, UT, USA) equipped with sensors for air temperature, relative humidity, global and net radiation, wind speed and Penman-Monteith algorithm (Allen et al., 1998), located 70 m away from lysimeter. Irrigation water was applied daily from day one to 15 of the trial lysimeter, to compensate *ETc* losses of the previous day. An average crop coefficient (*ETc/ETo*) was obtained and *ETc* was thus estimated as 0.89 × *ETo* during the deficit irrigation period. Plants of treatment *X* were irrigated based on *ETo* and a crop coefficient of 1.0 (one) to ensure that no water limitation would occur.

Irrigation in non-irrigated (*Y*) and lysimeter-grown trees was suspended on the 16th day of the trial. Lysimeter's evapotranspiration was then considered (recorded) measured evapotranspiration (ET_R) and compared to the estimated *ETc* (0.89 x *ETo*). When the relationship ET_R/ETc was lower than 1.0 (one), the threshold level of available soil water (*SWA*) based on evapotranspiration was determined. This procedure was also adopted to establish the threshold level of *SWA* based on *A, E, gs,* Ψ_P, and Ψ_M.

RESULTS AND DISCUSSION

Evapotranspiration

Estimated tree evapotranspiration (*ETc*) and measured evapotranspiration (ET_R) during the drying period (Figure 1A) tended to increase from day one of the experimental period, with the exception of two pronounced troughs (at days 13 and 33) caused by decreases in both global and net radiation, as well as air vapor pressure deficit (Figures 2A and B). The time-course of the relative evapotranspiration (ET_R/ETc) was almost constant along the first 25 days after the beginning of the drying period, varying around one when the available soil water content (*SWA*) was higher than 43%, and then declining sharply (Figure 1B). This result was close to a *SWA* of 46% reported by Allen et al. (1998) under similar environmental conditions and citrus tree development. However, it was lower than that threshold level recommended by Marler & Davies (1990) to 'Hamilin' trees (from 65 to 70%) in Florida. When *SWA* was around 43%, values of leaf and soil water potential were Ψ_P=-0.79 MPa, Ψs=-82.1 kPa (0.6 m). That suggests some level of water stress when compared with the results of Vu & Yelenosky (1988) and Machado et al. (2002). Pires (1992) suggested irrigating citrus when Ψs is between -50 and -70 kPa. Medina & Machado (1998) did not observe

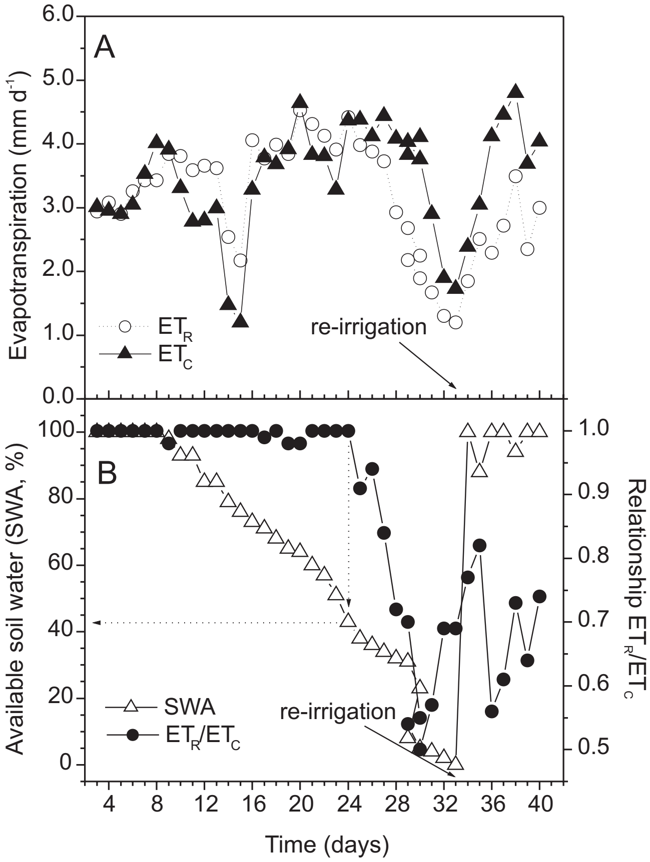

Figure 1 - Estimated crop evapotranspiration (*ETc*) and measured evapotranspiration (ET_R) throughout the experiment (A). Relative evapotranspiration (ET_R/ETc) and available soil water (*SWA*) (B). The arrow indicates when the lysimeter-grown tree was re-irrigated, 34 days after the beginning of the experimental period.

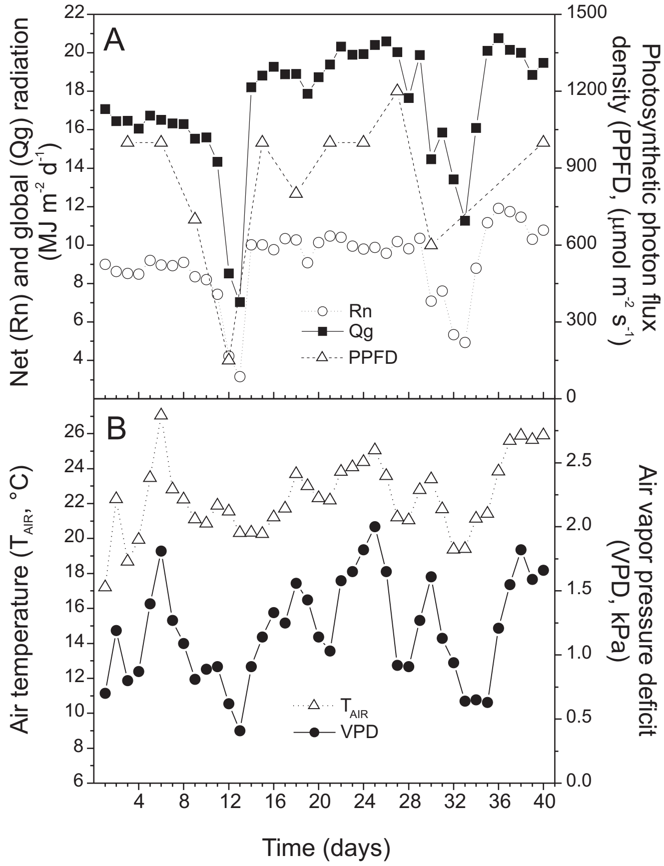

Figure 2 - Net and global radiation, photosynthetic photon flux density (A), air temperature and air vapor pressure deficit (B) throughout the experiment.

changes in *A*, *E*, *WUE*, leaf water potential and leaf relative water content in 'Valencia' oranges when the substrate water potential was higher than -40 kPa. Shalhevet & Levy (1990) suggested that Ψ_P should be maintained higher than -0.72 MPa to avoid water stress, but there may be large variability between plant species.

Thirty-four days after suspension of irrigation, the lysimeter-grown tree showed general leaf chlorosis with severe stress (Ψ_P=-2.70 MPa), and thus irrigation was re-established. Even after re-irrigation, ET_R/ETc remained lower than 1.0 (one) until the end of the experiment period. Ginestar & Castel (1996) reported that two weeks of irrigation were necessary for total rehydration of 'Clementine' mandarin trees subjected to severe water stress (Ψ_P =- 4.0 MPa) under field conditions. The involvement of stomata is among the factors causing non-recovery of ET_R. There was only partial recovery of stomatal opening in stressed 'Valencia' orange trees after re-irrigation (Medina et al., 1999). This recovery delay may be associated to accumulation of abscisic acid in the leaves during drying cycle (Davies & Zhang, 1991; Liu et al., 2003) or to damages to biochemical and/or photochemical processes of photosynthesis, which reduce stomatal conductance via increased intercellular CO_2 concentration (Wong et al., 1979).

Leaf gas exchange

Mean values of *A*, *gs* and *E* in irrigated plants ranged from 4 to 7 µmol m^{-2} s^{-1}, 0.05 to 0.11 mol m^{-2} s^{-1}, and 1.2 to 2.60 mmol m^{-2} s^{-1}, respectively (Figure 3). These values are lower than those reported under optimum environmental conditions (Syvertsen & Lloyd, 1994), especially for *A* which normally varies between 12 and 14.5 µmol m^{-2} s^{-1} (Medina, 2003). Such a difference was probably caused by fluctuations of environmental elements, such as *PPFD*, air temperature, and relative humidity to levels below those for optimum physiological, along the experimental period. Seasonal effects on leaf gas exchange had already been reported by Machado et al. (2002) who recorded higher photosynthetic rates on citrus trees during summer than in winter.

There were significant differences in *gs*, *A* and *E* between irrigated and non-irrigated trees only when *SWA* was below 60% (Figures 3B, D and F). In such condi-

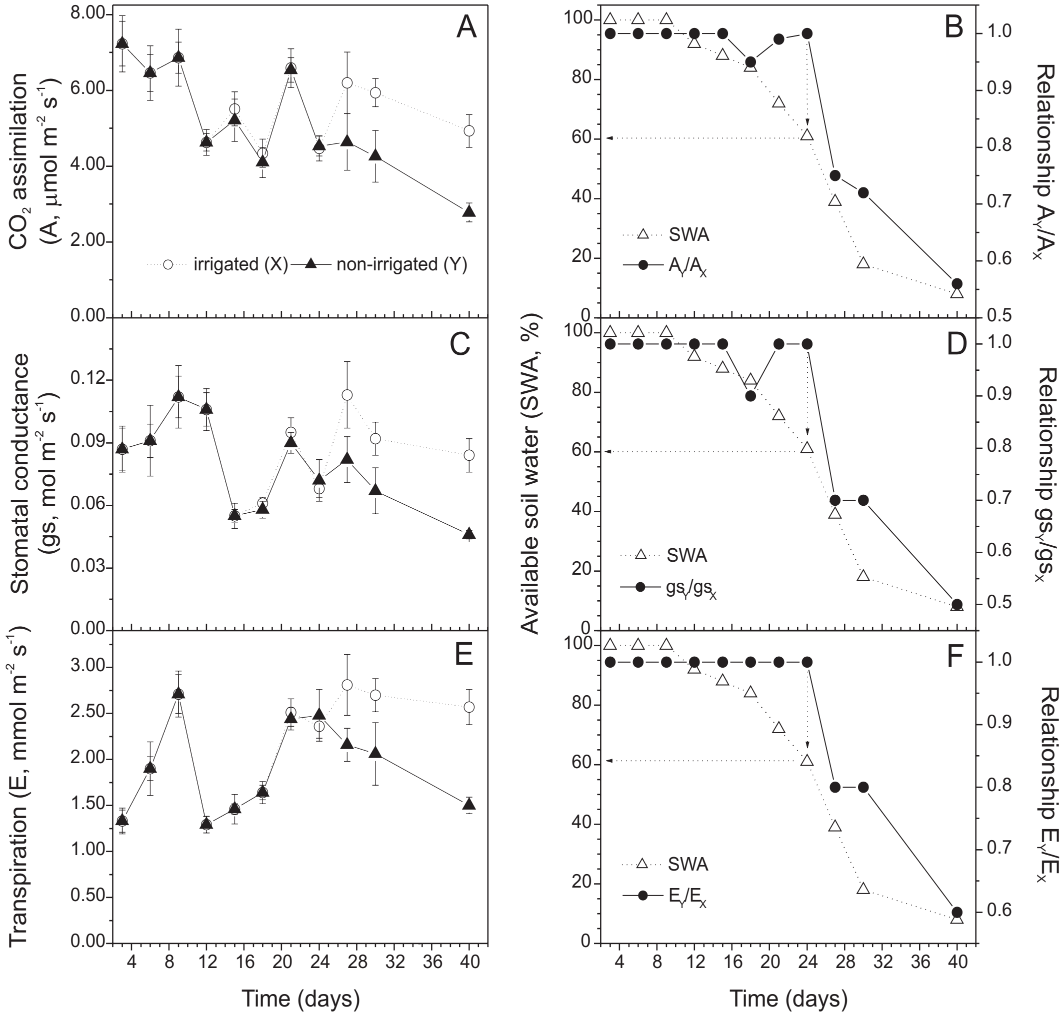

Figure 3 - Time-course of CO_2 assimilation (A) stomatal conductance (C), and transpiration (E) in irrigated (*X*) and non-irrigated 'Tahiti' lime trees (*Y*). Relationships between physiological variables and soil water content (*SWA*) throughout the experiment: CO_2 assimilation (B); stomatal conductance (D); transpiration (F). Each point represents the mean value ± SE (n = 6).

tions, the mean values of leaf and soil water potential were -0.62 and -48.8 kPa (0.6 m), respectively.

The leaf gas exchange measurements were more sensitive to *SWA* than evapotranspiration measured in the lysimeter. The reduction on stomatal conductance did not decrease the evapotranspiration possibly because leaf gas exchange measurements were taken in leaves located externally to the canopy and completely exposed to solar radiation, an unfavorable environmental condition in comparison to conditions inside tree canopy. Shaded leaves have higher leaf water potential and, as a consequence, higher stomatal aperture and transpiration (Syvertsen et al., 1981). Therefore, non-exposed leaves could have contributed to the maintenance of optimum evapotranspiration rates up to 43% *SWA*.

Water use efficiency (*WUE*) during the water withholding period as well as the relationship WUE_Y/WUE_X and *SWA* are shown in Figure 4. Since there were no differences between irrigated and non-irrigated trees (Figure 4A), it can be speculated that 'Tahiti' lime trees use the stomatal control mechanism to prevent plant dehydration and preserve plant water status. Brakke & Allen Jr (1995) did not find significant changes in *WUE* of citrus when comparing measurements taken at *SWA* ranging on 37 to 56%. The determination of a threshold level of *SWA* based on the relationship WUE_Y/WUE_X was not possible since it did not vary ($WUE_Y/WUE_X \approx 1.0$) along the experimental period (Figure 4B).

Leaf water potential

After 24 days of water deficit, Ψ_P and Ψ_M were reduced with Ψ_P showing significant decrease and reaching -0.9 MPa (Figure 5A). Although non-irrigated plants had shown different Ψ_P and Ψ_M in comparison to irrigated plants, the latter exhibited low mean throughout the experimental period (-0.57 MPa). The Ψ_P values were lower than the values recorded for the same irrigated trees during rainy summer (-0.35 MPa) even for the well-irrigated plants. Small wetted soil area (22% in this study) may lead to partial root wetting and reduction of Ψ_P as con-

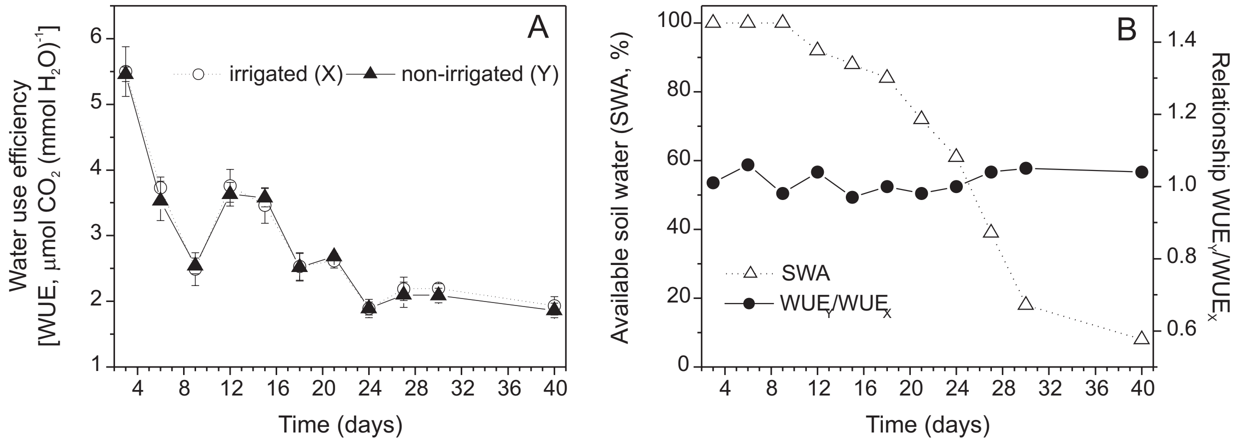

Figure 4 - Water use efficiency (*WUE*) (A), relationship between *WUE* in irrigated (*X*) and non-irrigated (*Y*) 'Tahiti' lime trees and soil water content (*SWA*) throughout the experiment (B). Each point represents the mean value ± SE (n = 6).

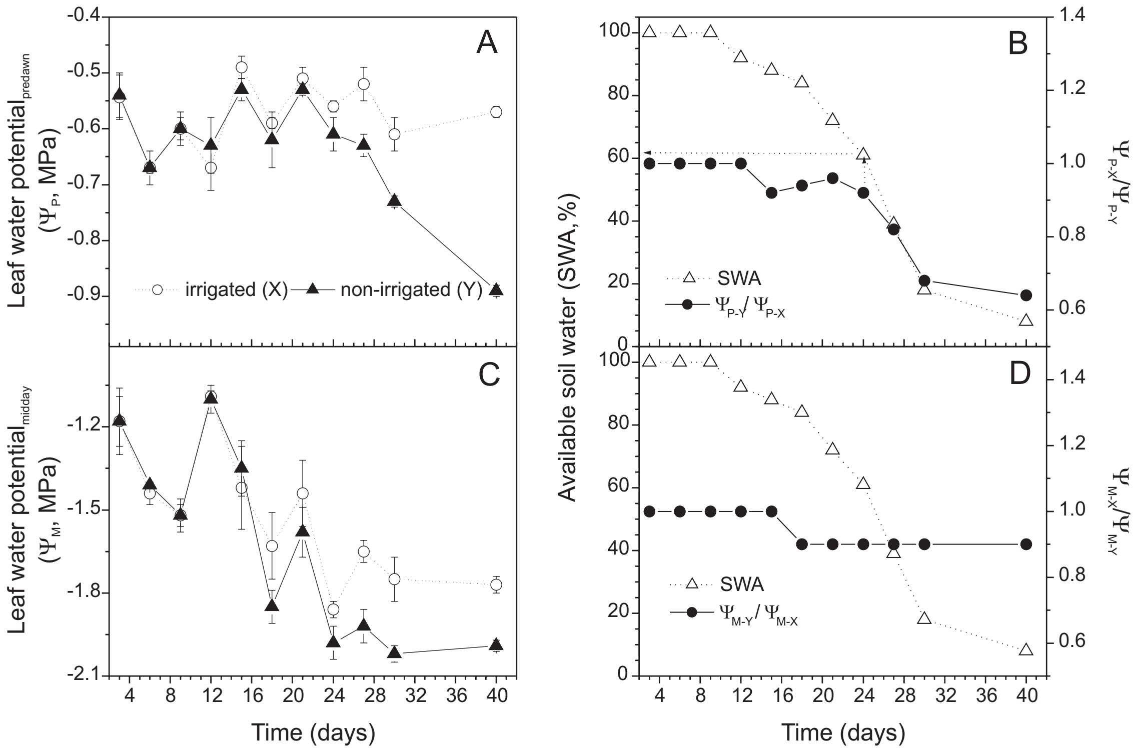

Figure 5 - Leaf water potential of 'Tahiti' lime measured at predawn (A) and noon (C) in irrigated (*X*) and non-irrigated plots (*Y*) throughout the experiment. The relationship between relative leaf water potential (*X/Y*) measured at predawn (B) and noon (D) and soil water content (*SWA*). Each point represents the mean value ± SE (n = 6).

sequence. Bernardo (1995) and Vermeiren & Jobling (1997) recommend the wetting of 50% of the potential zone root in perennial crops. Results of some recent, long-term experiments evaluating yield and citrus tree development under different levels of wetted area, have agreed that the wetting of around 50% of tree soil surface area is the ideal procedure (Bielorai, 1982; Smajstrla & Koo, 1984; Castel, 1994; Souza et al., 2003). However, Gowing et al. (1990) reported reduction of transpiration and leaf expansion in apple trees that had 50% of wetted roots when compared to well-irrigated ones. This is an important consideration to be made since increasing wetted area results in high irrigation costs by increasing the number of emitters, tube diameter, and water pump power. The soil temperature dropped to 7.5ºC during the experimental period, so it can be hypothesized that the low soil temperature could have affected the root hydraulic conductivity and root hormone content and, consequently, the hydration of shoot tissues (Elfving et al., 1972; Veselova et al., 2003). However, Machado et al. (2002) did not found environmental influence on leaf water potential at predawn on potted citrus plants during either January, March or July, despite differences in leaf gas exchange being recorded. During the experimental period, large variations in Ψ_M were observed on both treatments, and probably reflect weather conditions, as observed in other studies (Scholander et al., 1965; Southwick & Davenport, 1987; Domingo et al., 1996; Jones, 2004).

The relative leaf water potential at predawn (Ψ_{P-X}/Ψ_{P-Y}) decreased sharply when *SWA* was lower than 60% (Figure 5B). However, the relative leaf water potential at noon (Ψ_{M-X}/Ψ_{M-Y}) did not present steady drop as predawn measurements (Figure 5D), with a little drop to 0.9 when *SWA* was close to 80%, but remaining constant throughout the drying period. Some studies suggest that predawn values (maximum) are better indicators of irrigation needs than noon values (Shalhevet & Levy, 1990; Ginestar & Castel, 1996; Urribarrí et al., 1996; Domingo et al., 1996; González-Altozano & Castel, 2000). In fact, plant tissues are hydrated to a maximum right before sunrise when there is no water restriction, whereas measurements taken at noon reflect transpiring tissues, use more nitrogen gas and are more susceptible to fluctuations of environmental conditions, especially incident radiation, air temperature, and vapor deficit pressure.

Although the *SWA* was almost depleted by the end of drying period, the lowest value of leaf water potential was around -0.89 MPa, and higher than that obtained for the lysimeter-grown tree (Ψ_P=-2.70 MPa). This difference was probably caused by the fact that the root systems of field-grown trees were larger than that of the lysimeter-grown three, allowing better water uptake by the increased, exploited soil volume. Allied to the maintenance of *WUE* (there were no differences between irrigated and non-irrigated plants, Figure 4A) this phenomenon indicates the great ability of field-grown trees in avoiding the negative effects of water deficit.

For irrigation scheduling purpose, *SWA* of 60% is the threshold level to avoid negative physiological effects. Such *SWA* threshold level is correlated with leaf and soil water potentials of -0.62 MPa and -48.8 kPa (at 0.6 m depth), respectively.

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