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Effect of Irrigation Regimes on Water Status and Photosynthetic Parameters of Peach-Almond Hybrid (GF677) Seedlings and Cuttings

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Abstract. Almond is one of the drought resistant species suitable for growing in the dry lands of Iran. To evaluate the response of seedlings and cuttings of peach-almond hybrid (GF677) to different irrigation regimes, an experiment was conducted under greenhouse conditions in 2007. The stratified kernels and uniform rooted cuttings were planted in 20-L pots. After determination of field capacity (FC) of pot soils, irrigation treatments were imposed by daily irrigation to FC level or irrigation to FC at intervals 2, 4, or 8 days for 96 days. Stem water potential (SWP), relative water content (RWC), stomatal conductance, transpiration rate, photosynthetic rate, sub-stomatal CO_2, and water use efficiency were determined at 32 day intervals during the experimental period. Results showed that SWP at the three measurement times was lower in seedlings under 8 day irrigation intervals (-2.14 MPa) than in the cuttings (-1.95 MPa). The reduction of RWC throughout the experimental period was higher for seedlings (7.1%) than cuttings (5.1%). Therefore, cuttings of GF677, through better control of stomata, decreased transpiration, increased water use efficiency and appear to be more drought-resistant than their seedling counterparts.

***Additional key words*:** drought, photosynthesis, rootstock, water potential, water stress

Introduction

The use of bitter almond seedlings as rootstocks has been a traditional method for almond orchard establishment in Iran (Rom and Carlson, 1987). Many growers believe that these rootstocks have the most tolerance to drought and soil pathogens. The origin of many of them is, however, unknown and this causes problems with severe orchard heterogeneity (Felipe et al., 1997).

The GF677, a peach-almond rootstock, has been widely used in the Mediterranean area since the 1980s to reduce heterozygosity (De Salvador, 2004). This rootstock has only recently been introduced to Iranian growers. If this rootstock performs well in the arid and semiarid climate of Iran, it can replace the bitter almond seedling rootstock.

The acceptable productivity of scions on bitter almond seedling rootstock, from a grower's view, is due to relatively poor growth in dry farming systems. However, when these species are grown under better conditions, differences in vigor, yield, precocity, etc., were observed (Felipe, 1989). Gomes-Laranjo et al. (2006) stated that unfavorable environmental conditions decrease the kernel yield in dry soils. Practices such as irrigation (Felipe, 1989) and utilization of resistant rootstocks (Connell et al., 2002; Godini and Palasciano, 1997) can enhance the yield. Decrease in water potential of almond due to drought has been reported by Torrecillas et al. (1989) and Castel and Fereres (1982). Drought also causes decreased tree growth, high defoliation rate, kernel weight reduction, discoloration of pericarp and decrease in stomatal conductance and carbon assimilation (Isaakidis et al., 2004).

Torrecillas et al. (1989) observed that water deficiency caused a decline of tree canopy development and, with this decreased photosynthetic efficiency, a reduction in yield. Gomes -Laranjo et al. (2006) observed an increase in dry weight due to irrigation treatment compared to drought treatments and suggested that the decrease in dry weight associated with drought was due to a decline in photosynthetic rate. Shackel et al. (1998) have shown that the physiological response of almond to water stress may be correlated to the water status of plants as measured by stem water potential at midday.

Although drought resistance of vegetative GF677 rootstock has been reported by several researchers such as Alarcon et al. (2002) and De Salvador (2004), there has not been any report on the comparison of drought resistance between cuttings and seedlings of GF677. The difficulty in rooting this hybrid rootstock has stimulated nurserymen to produce seedlings of GF677. In the present study, the responses of GF677 cuttings to water stress with regard to photosynthetic activity, stem water potential (SWP), and relative water content (RWC) were evaluated and compared with those of seedlings obtained from GF677 trees after self pollination.

Materials and Methods

Seeds of self-pollinated trees of GF677 were collected in October 2006 from the Estahban Fig Research Station in Fars Province, Iran. After separating seeds from fleshy parts of the fruits, they were treated with benomyl (5 $g \cdot L^{-1}$) and stored in

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a refrigerator at 5℃ for 4 months. The hardy endocarps of seeds were removed and the kernels were stratified for 4 weeks at 4℃. The hardwood cuttings, 20-25 cm long and at least 1 cm in diameter, were prepared from 1-year-old shoots of mature GF677 trees in December 2006 and were treated with indole-3-butyric acid (3,000 ppm) and benomyl (2 $g \cdot L^{-1}$) and planted in the sand. The stratified kernels and uniform rooted cuttings were planted in 20-L pots filled with a mixture of sand, leaf mould, and loamy soil (1:1:1, v/v/v). The pots were then transferred to a greenhouse, with average day and night temperatures of 36 and 16℃, respectively. One hundred days after sowing of kernels and 60 days after sticking the cuttings, the uniform pots with uniform seedlings and cuttings were divided to four groups of eight pots per group. The first group was irrigated every day to field capacity (FC) level and served as a control. The second, third, and fourth groups were subjected to the three irrigation intervals of 2, 4, or 8 days, respectively, and then irrigated to the FC level. The experiment was carried out for 96 days after starting the irrigation treatments.

SWP was measured three times at 32 day intervals during 96 day experiment using a pressure bomb (PMS Instr. Co., Corvallis, OR, USA). Measurements were made on two fully expanded leaves from each of the three plants representing each treatment. The leaves were enclosed in a cellophane bag covered with aluminum foil for at least 1 h before SWP was determined to enable the water potential in the xylem of the leaf to come to equilibrium with the potential in the xylem of the stem at the point of attachment of the petiole (Garnier and Berger, 1985).

RWC was determined as (FW- DW)/(TW- DW) × 100, where FW is the fresh weight, DW is the dry weight after oven-drying the leaves at 80℃ for 24 h, and TW is the turgid weight after re-hydrating the leaves at 4℃.

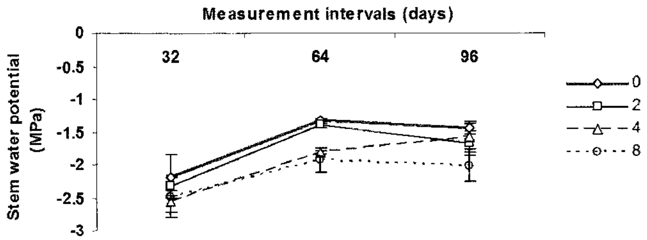
Stomatal conductance (g_s), transpiration rate (E), photosynthesis rate (A), and substomatal CO_2 (C_i) were monitored with a portable photosynthesis system LCA-3 (ADC BioScientific Ltd., Great Amwell, UK) at 32 day intervals. The measurements were performed on six, young, fully expanded, sun-exposed leaves per treatment (two leaves per plant, and three plants per treatment) between 9:00 and 11:00 a.m. Water use efficiency (WUE) was calculated as A/E relationship.

Experimental units were arranged in a completely randomized design, with four treatments and eight pots per treatment. The measurements were done on two leaves per plant and three plants (three pots) per treatment. Other plants were held intact for final biochemical analysis. Analysis of variance was performed using the MSTAT-C software package and significant difference among the mean values was compared by least significant difference test at $P = 0.05$.

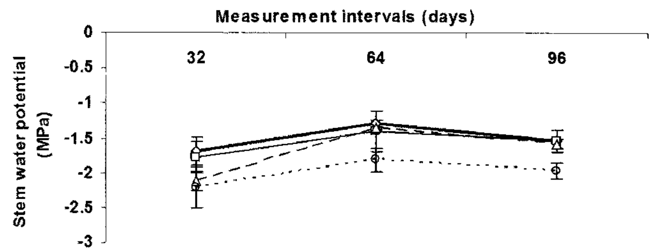
Results

SWP decreased with increasing irrigation intervals in seedlings of GF677 (Fig. 1A). After 32, 64, and 96 days of irrigation, percentage reduction of SWP compared with control were about 17.0, 22.5, and 10.4 under 4 days interval, respectively, and 13.8, 43.3, and 39.6 under 8 days interval, respectively. There was no significant difference in SWP under 0 and 2 day irrigation intervals. In GF677 cuttings, SWP was also reduced by increasing irrigation intervals (Fig. 1B). In cuttings after 32, 64, and 96 days of irrigation, reduction percentage of SWP compared with control were about 26.0, 5.4, and 3.3 under 4 days interval, respectively, and 30.2, 40.3, and 28.1, under 8 days interval, respectively. Under 8 days irrigation intervals, the average mean of SWP of seedlings (-2.14 MPa) was lower than that of GF677 cuttings (-1.95 MPa) during this study. Both types of rootstock had minimum SWP after 32 days of drought period, but after 64 days SWP increased and then decreased significantly thereafter after 96 days.

In both seedlings and cuttings of GF677, RWC values of irrigated plants under 8 day intervals were lower than shorter irrigation intervals; the differences were significant in seedlings at $P = 0.05$ (Figs. 2A and 2B). Sixty four days after the experiment was started, the differences between RWC values of irrigated cuttings under 0 and 2 day irrigation intervals were significantly greater than that of 4 and 8 day irrigation intervals. Ninety six days after treatments started, the differences in RWC values of irrigated cuttings under 8 day intervals and other treatments were significant at $P = 0.05$ (Fig. 2B). In-



(A)



(B)

Fig. 1. Effect of various irrigation regimes on SWP of GF677 seedlings (A) and cuttings (B). Vertical bars are standard deviation of the means.

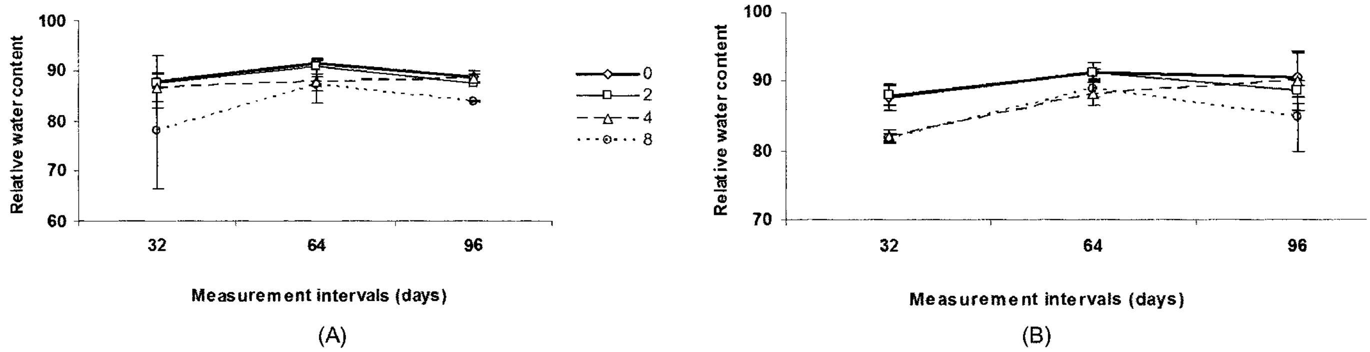


Fig. 2. Effect of various irrigation regimes on RWC of GF677 seedlings (A) and cuttings (B). Vertical bars are standard deviation of the means.

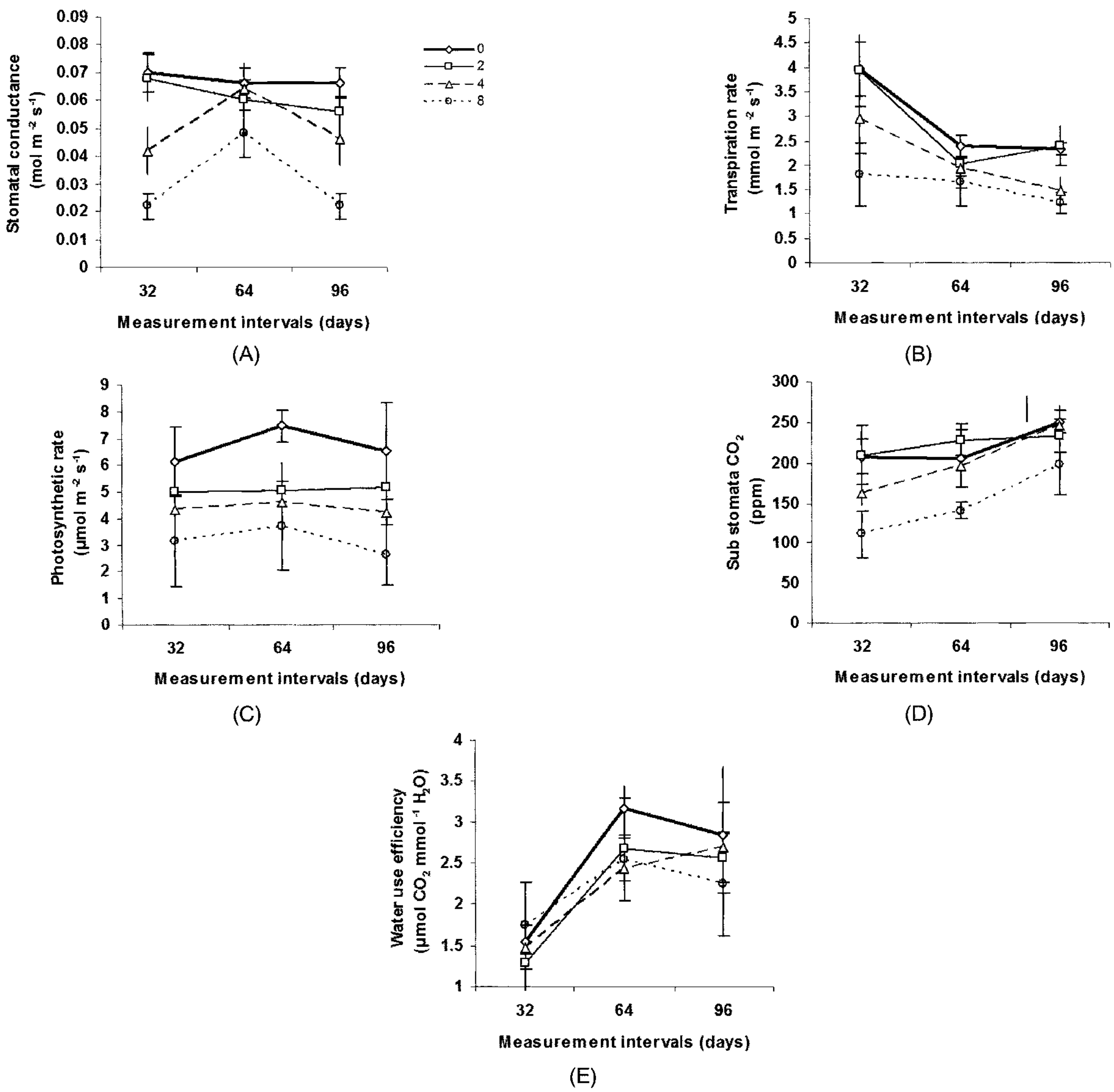


Fig. 3. Effect of various irrigation regimes on stomatal conductance (A), transpiration rate (B), photosynthetic rate (C), sub stomatal CO_2 (D) and WUE (E) of GF677 seedlings. Vertical bars are standard deviation of the means.

creasing irrigation intervals from 0 to 8 days reduced the mean RWC values, averaged over the entire experiment, in seedlings and cuttings by 7.1 and 5.1%, respectively. RWC values of both rootstocks at the end of experiment were significantly lower than at mid season.

Figs. 3 and 4 show the effects of irrigation regimes on

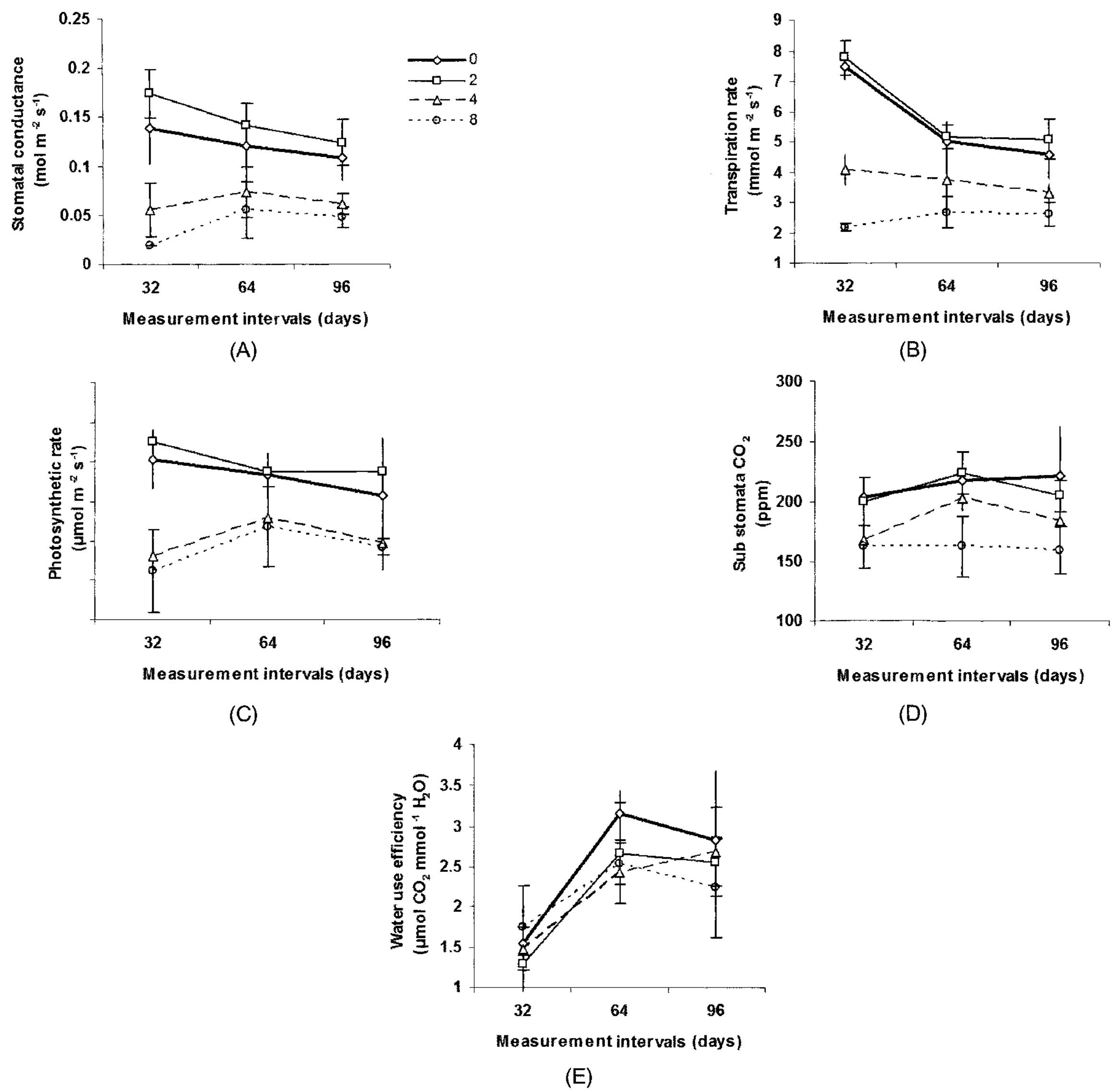


Fig. 4. Effect of various irrigation regimes on stomatal conductance (A), transpiration rate (B), photosynthetic rate (C), sub stomatal CO_2 (D), and WUE (E) of GF677 cuttings. Vertical bars are standard deviation of the means.

stomatal conductance (g_s), transpiration rate (E), photosynthesis rate (A), substomatal CO_2 (C_i), and WUE. In cuttings, g_s was decreased by longer irrigation intervals. By increasing irrigation intervals from 0 to 8 days in seedlings and cuttings g_s decreased to 53.7 and 66.7%, respectively (Figs. 3A and 4A). A decrease in transpiration rate (E) paralleled the decrease in g_s with increasing irrigation intervals. In seedlings and cuttings, E values under 8 day irrigation intervals were always lower than other treatments (Figs. 3B and 4B). In seedlings, 32 days after the experiment started, the difference in E values between 8 day irrigation intervals and the other treatments were significant; however, on the second and third measurements, no significant differences in E values were observed (Fig. 3B). In cuttings on the first and third measurements, there were significant differences between 0 and 2 day and 4 and 8 day irrigation intervals (Fig. 4B). In both seedlings and cuttings, photosynthetic rates (A) were decreased by increasing irrigation intervals (Figs. 3C and 4C). Compared to the control, seedling photosynthetic rates (A) on day 32, 64, and 96 of the experiment decreased by 29.2, 39.2, and 35.7%, and 48.1, 49.9, and 59.4% under 4 and 8 day irrigation intervals, respectively. The equivalent E values under 4 and 8 day irrigation intervals for cuttings were 48.2, 22.8 and 29.2%, and 55.9, 27.8 and 31.1%, respectively. The photosynthetic rate measurements for the cuttings under 2 day irrigation intervals were higher than that for the control. Substomatal CO_2 (C_i) concentrations of both plant types tended to decline with increasing irrigation intervals (Figs. 3D and 4D). Thirty two days after the experiment started, only C_i values of seedlings irrigated under 2 day intervals were not significantly different from the control (0 day interval). After 64 and 96 days, a significantly lower C_i value was observed under 8 day irrigation interval compared to the control; but for plants subjected to 2 and 4 day intervals, values were not significantly different from the control (Fig. 3D). The C_i values for cuttings under 8 days irrigation intervals were significantly lower compared to other treatments, especially after 64 and 96 days (Fig. 4D). In seedlings, WUE values did not differ among treatments, but mean values seemed to decrease with increasing irrigation intervals (Fig. 3E). The results of this study showed that WUE in cuttings increased with increasing irrigation intervals and these values were highly significant under the 8 day irrigation interval, compared to the control (Fig. 4E).

Discussion

Some species avoid drought and do not experience low water potential in their tissues (Tripathy et al., 2000). However, in drought tolerant plants, metabolic processes continue even though tissue water potential decreases (Nilsen and Orcutt, 1996). SWP of GF677 seedlings and cuttings decreased by increasing irrigation intervals, which is similar to the finding of Sircelj et al. (2005) working with apple cultivars. A significant decrease of SWP while the plants had relatively good health suggests that there was a tolerance mechanism in the two types of rootstocks studied. These results are in agreement with the findings of Torrecillas et al. (1989). Under natural conditions, the minimum RWC and stem water potential is observed during maximum evaporative demand due to temporal imbalance between water absorption in the root and water losses by transpiration. Under soil water deficit conditions, this imbalance becomes more severe and causes minimum RWC and SWP of drought treatment relative to the controls (Romero and Botia, 2006). As was reported by Castel and Fereres (1982), a sharp decrease in water potential was observed at the beginning of the drought period. They suggest that this decrease is due to a shallow root system. Apparently, water potential of trees with a shallow root system is affected earlier in the drought cycle than trees with a deep root system.

An obvious decrease of SWP at the end of the season, in spite of cooler weather, may be due to leaf senescence (Lakso et al., 1984), as leaf senescence induces an increase in resistance to water flow in the xylem (Ruiz-Sanchez et al., 1993). Also a lower amount of SWP at the end of the experiment might demonstrate increasing degree of drought stress imposed on the plants (Sircelj et al., 2005) and its effect on osmolyte accumulation (Proebsting et al., 1989; Ranjbar et al., 2002).

The lower SWP of seedlings associated with increasing irrigation intervals displayed a decrease in water flow to leaves that, according to Fernandez et al. (1997), is due to reduced internal plant conductance. Subrahamnyam et al. (2006) showed that inadequate irrigation caused a greater decrease in water potential of sensitive cultivars compared to tolerant ones.

Water stress, as measured by stomatal conductance (g_s), decreased in drought treatments. This agrees with the findings of Rouhi et al. (2007), Romero and Botia (2006), and Torrecillas et al. (1996) for almonds. This response suggests vigorous stomatal regulation that helps to reduce water losses under low water availability conditions. Gradual decrease of A and g_s with increasing drought stress is a special response of adaptive almond species under water stress conditions (Romero et al., 2004). Greater decline in gs of the cuttings can justify higher SWP observed. Transpiration was higher in cuttings than in seedlings due to larger leaf area (data not shown). The decreases in transpiration (E) and stomatal conductance (g_s) with increasing irrigation intervals were higher in cuttings than in seedlings. According to Natali et al. (1996), a significant decrease in transpiration helps to maintaining acceptable plant water potential. This hypothesis also justifies higher RWC in cuttings than in seedlings. However, stomatal closing reduces photosynthesis. Rouhi et al. (2007) observed a decrease in A and gs with C_i in bitter almond. In our study, C_i decreased as g_s decreased indicating that stomatal conductance is a major factor that can limit assimilation. Torrecillas et al. (1999) observed an increase in C_i after soil moisture declined and suggested that C_i enhancement is due to stomata limitation and a reduction in carbon metabolism. We can anticipate that for the GF677 rootstock under our water stress period, photosynthetic system will not be impaired. Increase in WUE values under severe water stress (8 days intervals) was in agreement with Romero and Botia (2006), Natali et al. (1996), and Cheng et al. (1996). Although these high WUE values are associated with a decrease in CO_2 assimilation, it seems that the stomata behave optimally to maximize the amount of carbon gain per unit of water used by partially closing and consequently reducing transpiration (Hsiao, 1990). While not significant in our study, reduction of WUE in seedlings with increasing irrigation intervals can be a result of decreasing A and also steady transpiration (Romero and Botia, 2006). A decrease in stomatal conductance in seedlings was not significant except for the 8 day treatment and they all had relatively similar transpiration rates.

In conclusion, GF677 cuttings had greater ability to withstand drought conditions than self pollinated GF677 seedlings. Cuttings had higher SWP values and also higher RWC as a result of decreasing stomata conductance and transpiration rate under low soil water availability, relating to higher WUE of the cuttings compared to the seedlings under drought conditions.

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