

RESEARCH ARTICLE

Alternate partial root-zone irrigation with high irrigation frequency improves root growth and reduces unproductive water loss by apple trees in arid north-west China

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Abstract Alternate partial root-zone irrigation (APRI) can improve water use efficiency in arid areas. However, the effectiveness and outcomes of different frequencies of APRI on water uptake capacity and physiological water use have not been reported. A two-year field experiment was conducted with two irrigation amounts (400 and 500 mm) and three irrigation methods (conventional irrigation, APRI with high and low frequencies). Root length density, stomatal conductance, photosynthetic rate, transpiration rate, leaf water use efficiency, midday stem and leaf water potentials were measured. The results show that in comparison with conventional irrigation, APRI with high frequency significantly increased root length density and decreased water potentials and stomatal conductance. No differences in the above indicators between the two APRI frequencies were detected. A significantly positive relationship between stomatal conductance and root length density was found under APRI. Overall, alternate partial root-zone irrigation with high frequency has a great potential to promote root growth, expand water uptake capacity and reduce unproductive water loss in the arid apple production area.

Keywords alternate partial root-zone irrigation, apple tree, leaf water use efficiency, root length density, stomatal conductance, water potential

1 Introduction

China has the largest apple production area in the world and accounts for 50% of global production capacity. Annual apple production increased from 800 kt in 1970 to 38 Mt in 2012, and is expected to be 51 Mt by 2020. Arid

north-west China is an important high quality apple production area because of ample sunlight and heat, a large difference between day and night temperatures and a dry climate^[1]. However, water resource availability limits the expansion of production in this area, as does the risk of climate change effects. Therefore it is urgent to enhance irrigation water use efficiency by improving irrigation methods and reducing irrigation amounts^[2].

Under conventional irrigation with large irrigation volumes and long irrigation intervals, there is a alternation of excess water supply and water deficit stress, which results in increasing stomatal conductance and luxury transpiration for a short time after irrigation, followed by reduced water potential and photosynthesis before the next irrigation^[3]. Previous studies indicated that promoting root growth, reducing luxury transpiration and regulating stomatal movement are important approaches to improve water use efficiency of fruit trees^[4,5]. Stomata are the channels for flux exchange between trees and the atmosphere, and adjusting stomatal opening and closing regulates water loss through transpiration^[6].

Stem and leaf water potentials are important indicators to characterize water status of fruit trees, as these determine water movement^[7,8]. When water content of a tree is reduced, the decreased water potential and the increased tension of xylem result in the cavitation and embolization of xylem, which may reduce water transport capacity and ultimately decrease stomatal conductance^[9]. Furthermore, the reduction of stem and leaf water potentials results in the decrease in leaf stomatal conductance under conventional irrigation^[10–12].

However, the variation of stomatal conductance is not only affected by water potential, but is also related to the ion concentration in root signals^[13,14]. When water deficit stress occurs, the root system can send chemical signals to reduce leaf stomatal conductance and luxury transpiration^[15]. Meanwhile, moderate water deficit stress may promote root growth and improve water uptake

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capacity^[16]. According to the above- and below-ground balance theory^[17], fruit trees can transfer more photosynthates into roots under moderate water deficit stress, while more photosynthates will be transported to leaves and fruits under adequate water supply^[18]. Moreover, a significant linear correlation has been found between stomatal conductance and root length density under conventional irrigation^[19], which better reflects the relationship between above-ground and below-ground water status.

Alternate partial root-zone irrigation (APRI) is an innovative irrigation method, which can reduce stomatal conductance and improve water use efficiency^[14,20]. However, the effectiveness and outcomes of different frequencies of APRI on root water uptake capacity and physiological water use have not been reported and need investigation. Therefore, the objectives of this study were (1) to evaluate the impact of APRI with different irrigation frequencies on root length density, stomatal conductance, photosynthetic rate, transpiration rate, leaf water use efficiency, and midday stem and leaf water potentials in apple trees in arid north-west China, and (2) to analyze the relationship between stomatal conductance and root length density to understand the mechanism of efficient water use under this system.

2 Materials and methods

2.1 Experimental site and meteorological parameters

Field experiments were conducted during 2013 and 2014 in an apple orchard at the Shiyanghe Experimental Station, China Agricultural University (37°52' N, 102°50' E, 1581 m) located in a typical continental temperate climate

zone, with annual mean temperature of 8°C, annual accumulated temperature (> 0°C) of more than 3550°C, annual mean sunshine duration of more than 3000 h, annual mean precipitation of 164 mm, annual mean pan evaporation of about 2000 mm, free frost days of 150 d and mean groundwater depth of > 25 m^[21,22]. The soil in the orchard is classified as a Siltic-Orthic Anthrosol, a light sandy loam. The mean bulk density is 1.46 g·cm⁻³, and the mean volumetric water content at field capacity is 0.30 cm³·cm⁻³. A Meteorological Monitoring System (Jauntering, New Taipei City, Taiwan) located in the center of the experimental site automatically recorded net radiation at 4.0 m and precipitation with time intervals of 15 min during the entire 2013 and 2014 growth seasons (Fig. 1).

2.2 Experimental design and field management

Field experiments were conducted in a field of about 0.5 hm² (68 m × 66 m) with 170 trees. Experimental apple trees (*Malus domestica* Borkh. cv Golden Delicious) were planted in 1981 at a row spacing of 6 m and plant spacing of 4 m. Eighteen trees of similar size with no disease or pests were selected to reduce tree damage and workload (Table 1). There were six treatments, including two irrigation amounts and three irrigation methods, and each treatment had three replicates. The two irrigation amounts were 400 and 500 mm, and three irrigation methods were (1) conventional irrigation, irrigated 4 times over the whole growth season (CI), (2) APRI with high irrigation frequency (PRI_H), irrigated 8 times for two subplots alternately, and (3) APRI with low irrigation frequency (PRI_L), irrigated 4 times for two subplots alternately. Irrigation was applied by border irrigation through

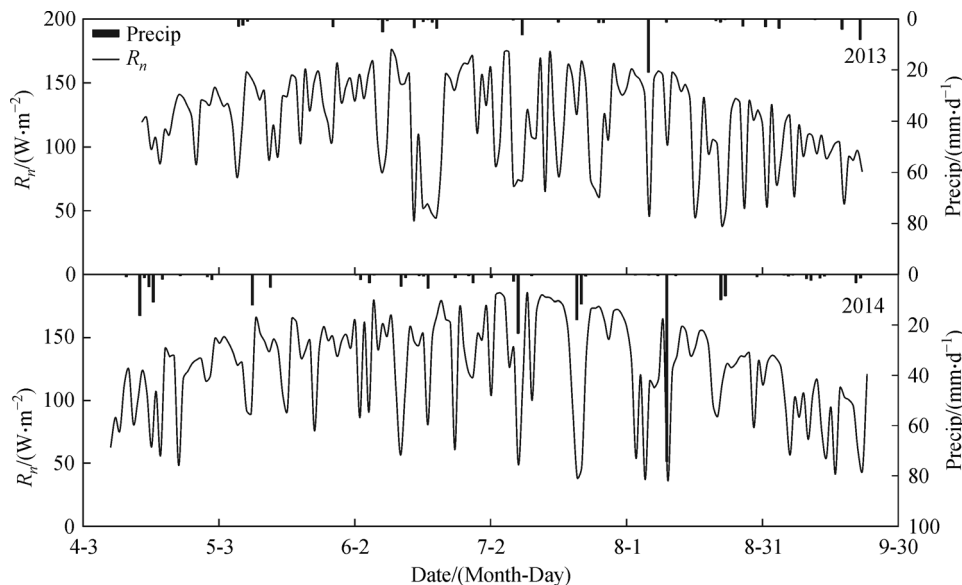


Fig. 1 Seasonal variations of net radiation (R_n) and precipitation (Precip) over the entire 2013 and 2014 growth seasons

Table 1 Trunk diameter, bark depth, radii of sapwood and heartwood of apple trees used for field experiment

Irrigation amount/mm	Irrigation method	Trunk diameter/mm	Bark depth/mm	Radius of sapwood/mm	Radius of heartwood/mm	
400	CI	269	6.0	128	76	
		251	6.3	111	45	
		263	7.0	142	48	
	PRI _H	267	5.9	126	70	
		279	7.1	135	75	
		245	5.8	123	61	
	PRI _L	252	6.0	120	68	
		287	7.9	129	58	
		239	6.0	119	62	
		247	5.0	118	67	
500	CI	275	8.0	141	65	
		259	5.3	135	55	
		266	6.2	126	54	
	PRI _H	258	6.0	125	45	
		281	7.3	131	55	
		263	5.6	125	44	
	PRI _L	279	7.5	121	54	
		249	6.4	105	48	
		Average	263	6.4	126	58
		Maximum	287	8.0	141	76
Minimum	245	5.0	105	44		

Note: CI, conventional irrigation; PRI_H, alternate partial root-zone irrigation with high irrigation frequency; PRI_L, alternate partial root-zone irrigation with low irrigation frequency.

irrigation pipelines from the well in the apple orchard and the irrigation amount was monitored by water meter linked to the pipelines. Irrigation times, quota and date are shown in Table 2.

To avoid the exchange of water and nutrients between neighboring subplots, impermeable film (1.5 m deep, 3.0 m wide and 3.0 m long) was installed around the trees with a ridge (0.3 m high) formed at ground level on 4

Table 2 Irrigation times, quota and date for different treatments

Year	Amount/mm	Method	Detail		
			No. of times	Quota/mm	Date
2013	400	CI	4	100.0	26 April, 31 May, 27 June, 1 August
		PRI _H	8	50.0	26 April, 13 May, 31 May, 19 June, 27 June, 14 July, 1 August, 17 August
		PRI _L	4	100.0	26 April, 31 May, 27 June, 1 August
	500	CI	4	125.0	26 April, 31 May, 27 June, 1 August
		PRI _H	8	62.5	26 April, 13 May, 31 May, 19 June, 27 June, 14 July, 1 August, 17 August
		PRI _L	4	125.0	26 April, 31 May, 27 June, 1 August
2014	400	CI	4	100.0	25 April, 5 June, 13 July, 7 August
		PRI _H	8	50.0	25 April, 24 May, 5 June, 26 June, 13 July, 29 July, 7 August, 31 August
		PRI _L	4	100.0	25 April, 5 June, 13 July, 7 August
	500	CI	4	125.0	25 April, 5 June, 13 July, 7 August
		PRI _H	8	62.5	25 April, 24 May, 5 June, 26 June, 13 July, 29 July, 7 August, 31 August
		PRI _L	4	125.0	25 April, 5 June, 13 July, 7 August

Note: CI, conventional irrigation; PRI_H, alternate partial root-zone irrigation with high irrigation frequency; PRI_L, alternate partial root-zone irrigation with low irrigation frequency.

April, 2013 (Fig. 2). In addition, to avoid the movement of water and nutrients between the wet and dry areas, the APRI plots were also divided into two equal subplots by impermeable film (0.5 m deep) with a ridge (0.3 m high) formed at ground level (Fig. 2). The orchard was fertilized annually with $440 \text{ kg}\cdot\text{hm}^{-2}$ N, $200 \text{ kg}\cdot\text{hm}^{-2}$ P_2O_5 and $150 \text{ kg}\cdot\text{hm}^{-2}$ K_2O as urea (N 46%), diammonium phosphate (P_2O_5 45%, N 17%) and potassium magnesium sulfate (K_2O 24%), respectively. In addition, weeds were controlled manually or with herbicides, and pests with pesticides as needed.

2.3 Measurements

2.3.1 Root length density

The roots samples were taken using a root auger ($10 \text{ cm} \times 10 \text{ cm}$). Samples were taken to 1.5 m deep at 0.1 m intervals. Over the whole growth season, one row of 14 sites from east to west of the subplot were taken in each growth stage (Fig. 2). After sieving the soil sample, the roots of $< 2 \text{ mm}$ in diameter were collected and washed with tap water. The clean roots were arranged regularly on transparent plexiglass plates and scanned to give Tagged Image File Format images (Epson Perfection V700 Photo, Seiko Epson Corp., Nagano, Japan) of 300 dpi for further analysis. The image files were analyzed by WinRHIZO image analysis software (Regent Instrument Inc., Quebec, QC, Canada) to obtain root length density. After roots were removed, the soil was used to backfill sampling holes. In addition, to avoid any sampling effect on tree growth, different replicate trees were sampled in 2013 and 2014.

2.3.2 Stomatal conductance, photosynthetic rate, transpiration rate and leaf water use efficiency

Stomatal conductance, and photosynthetic and transpiration rates were measured using an LI-6400 portable photosynthesis system (LI-COR Biosciences, Lincoln, NE, USA) with a standard chamber. The measurements were taken on four mature leaves of similar size and age from both south and north sides of each treatment at 12:00 on typical sunny days. During the whole growth season, four measurements were taken at the same time as the soil sampling. The data logger of the LI-6400 recorded stomatal conductance, and photosynthetic and transpiration rates immediately after measurement. In addition, leaf water use efficiency was calculated as:

$$\text{WUE}_{\text{leaf}} = \frac{P_n}{T_r} \quad (1)$$

where WUE_{leaf} is leaf water use efficiency ($\text{mmol CO}_2 \cdot \text{mol}^{-1} \text{ H}_2\text{O}$), P_n is photosynthetic rate ($\mu\text{mol CO}_2 \cdot \text{m}^{-2} \cdot \text{s}^{-1}$), and T_r is transpiration rate ($\text{mmol H}_2\text{O} \cdot \text{m}^{-2} \cdot \text{s}^{-1}$).

2.3.3 Midday stem and leaf water potentials

Three hours before the measurement, two mature leaves of similar size and age from both south and north sides near the tree stem were enclosed in opaque plastic bags covered with aluminum foil, and water potentials of the leaves were measured immediately at 12:00 using a pressure chamber (SKPM 1400, Skye, UK). The midday water potentials of these leaves are considered to be equivalent to midday

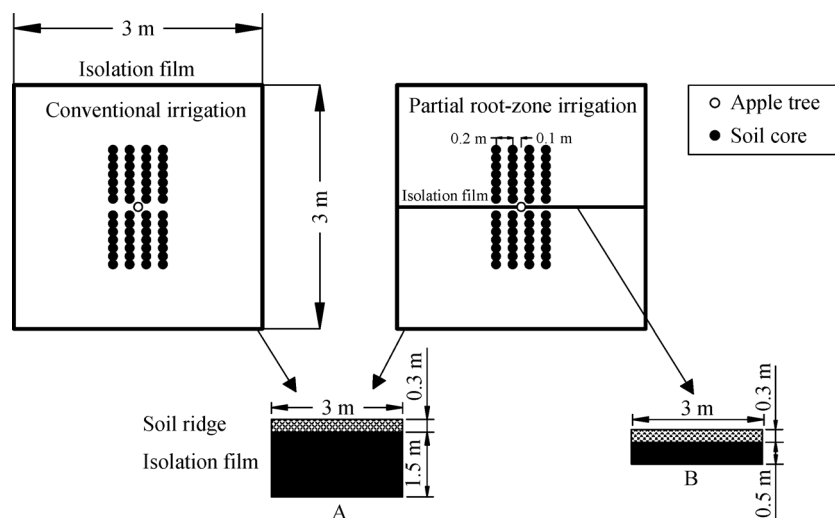


Fig. 2 Description of subplot isolation by impermeable film and ridges, and the positions of soil cores. To avoid the exchange of water and nutrients between neighboring subplots, impermeable film (1.5 m deep, 3.0 m wide and 3.0 m long) was installed around the trees with a ridge (0.3 m high) formed at ground level (A). In addition, to avoid the movement of water and nutrients between the wet and dry areas, the APRI plots were divided into two equal subplots by impermeable film (0.5 m deep) with a ridge (0.3 m high) formed at ground level (B).

stem water potentials^[23]. Midday leaf water potential was also measured for two mature leaves of similar size and age from both south and north sides immediately at 12:00. During the whole growth season, both stem and leaf water potentials were measured at time intervals of 5–7 d.

2.4 Statistics analysis

The general linear model procedure of the SAS software (SAS Institute, Cary, NC, USA) was used to perform analysis of variance with two- and three-way interactions for root length density, stomatal conductance, photosynthetic rate, transpiration rate, leaf water use efficiency, and midday stem and leaf water potentials with the main effects of year, irrigation amount and irrigation method. In addition, treatment means were compared by the least significant differences at $P < 0.05$.

3 Results and discussion

3.1 Root length density and stomatal conductance

Different irrigation methods and amounts had significant effects on root length density (Table 3), but no significant interactions were found for experimental year, irrigation method and amount. Therefore, root length density was further analyzed across experimental years, irrigation methods and irrigation amounts.

Root length density was significantly higher under PRI_H than CI (Fig. 3a). Previous studies indicated that slight water deficit could promote root growth^[14,24]. Our results confirmed that frequent alternate drying and wetting under APRI stimulates root growth in apple trees (Fig. 3a). However, there was no significant difference in root length density between PRI_H and PRI_L (Fig. 3a). Slightly lower root length density under PRI_L can be explained by reduced water uptake capacity of part of the root system in the dry area under PRI_L , which suffers water deficit stress for long time before the next watering interval. It further results in suberization of the root tip surface and the reduction in secondary roots until root death^[20]. In addition, significantly higher root length density with 500-mm irrigation compared to 400-mm irrigation (Fig. 3a) demonstrates that more photosynthates are transferred into the root system with 500-mm irrigation^[25,26]. In addition, there was no significant difference

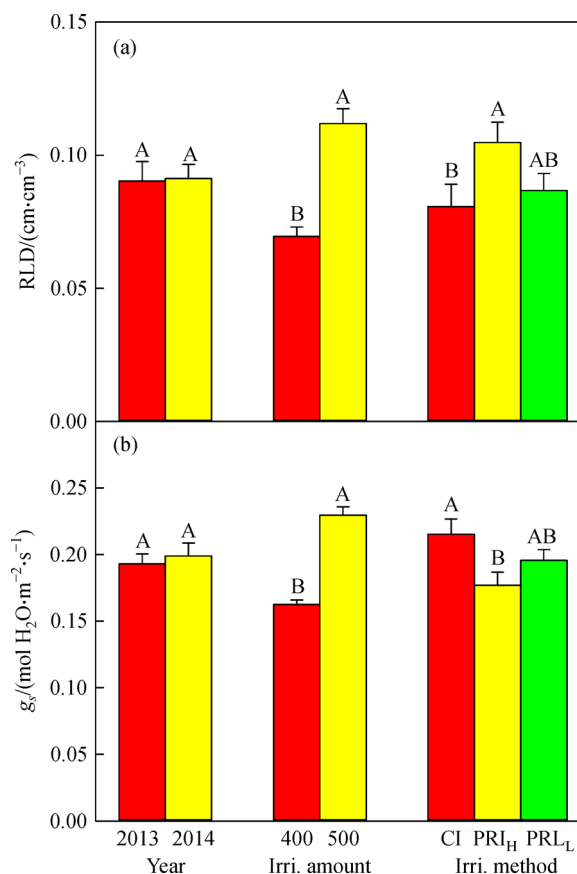


Fig. 3 Root length density (RLD) (a) and stomatal conductance (g_s) (b) during the whole growth season as affected by year, irrigation amount and irrigation method. Bars labeled with different letters are significantly different ($P < 0.05$) and with the same letter are not significantly different ($P \geq 0.05$). Error bars indicate standard error of means. CI, conventional irrigation; PRI_H , alternate partial root-zone irrigation with high irrigation frequency; PRI_L , alternate partial root-zone irrigation with low irrigation frequency.

in root length density between the two years, which was probably due to lack of significant difference in root-zone soil water content^[27].

There were significant effects of different irrigation methods and amounts on stomatal conductance (Table 3), while no significant interactive impact was found for experimental year, irrigation method and irrigation amount. Compared with CI, PRI_H significantly reduced stomatal conductance (Table 3; Fig. 3b), which indicates that more abscisic acid can be produced under PRI_H to

Table 3 Probabilities (P) of different treatments

Source	DF	RLD	g_s	P_n	T_r	WUE _{leaf}	Ψ_{stem}	Ψ_{leaf}
IA	1	< 0.001	< 0.001	< 0.001	< 0.001	0.984	< 0.001	< 0.001
IM	2	< 0.05	< 0.001	< 0.05	< 0.01	0.488	< 0.001	< 0.001

Note: DF, degree of freedom; RLD, root length density; g_s , stomatal conductance; P_n , photosynthetic rate; T_r , transpiration rate; WUE_{leaf}, leaf water use efficiency; Ψ_{stem} , midday stem water potential; Ψ_{leaf} , midday leaf water potential; IA, irrigation amount; IM, irrigation method. P values were shown ($P < 0.05$, significant; $P < 0.01$, strongly significant; $P \geq 0.05$, not significant). The effects of Year and all interactions were non-significant.

decrease stomatal conductance, when the roots are under slight water deficit stress^[28,29]. Also, the moderate reduction in stomatal conductance under PRI_H can decrease luxury transpiration^[27,30,31]. However, APRI with different irrigation frequencies had no significant impact on stomatal conductance (Fig. 3b). Furthermore, compared with 400-mm irrigation, 500-mm irrigation significantly enhanced stomatal conductance (Fig. 3b) because adequate water supply can generate turgor, which is beneficial for stomatal opening^[31,32].

The stomatal conductance under different irrigation methods showed upward trends with the increase in root length density (Fig. 4). The significant linear relationship between stomatal conductance and root length density under APRI and CI indicates that root length density is an important factor for root water uptake capacity^[19]. Moreover, coordinating the relationship between stomatal conductance and root length density with different irrigation methods should first ensure the required transpiration during the normal photosynthetic processes, and then reduce unproductive water consumption of apple trees caused by excessive stomatal conductance^[33].

3.2 Photosynthetic and transpiration rates, and leaf water use efficiency

Since no significant interactions were found for experimental year, irrigation method and irrigation amount for photosynthetic and transpiration rates, and leaf water use efficiency, the data were further analyzed across experimental years, irrigation methods and irrigation amounts, respectively (Table 3). There was no significant effect of

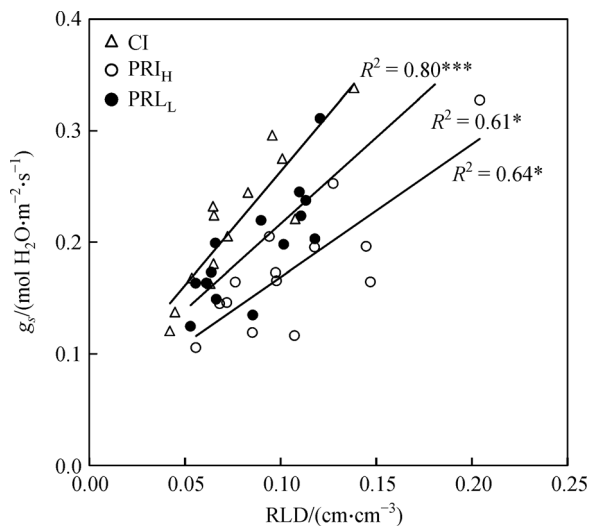


Fig. 4 Relationship between stomatal conductance (g_s) and root length density (RLD) during the entire 2013 and 2014 growth seasons. CI, conventional irrigation; PRI_H, alternate partial root-zone irrigation with high irrigation frequency; PRL_L, alternate partial root-zone irrigation with low irrigation frequency; * $P < 0.05$, ** $P < 0.01$, and *** $P < 0.001$.

the different irrigation methods on photosynthetic and transpiration rates, and leaf water use efficiency (Fig. 5), possibly because APRI does not significantly reduce carbon dioxide assimilation^[20], but limits luxury transpiration without impact on apple tree physiological

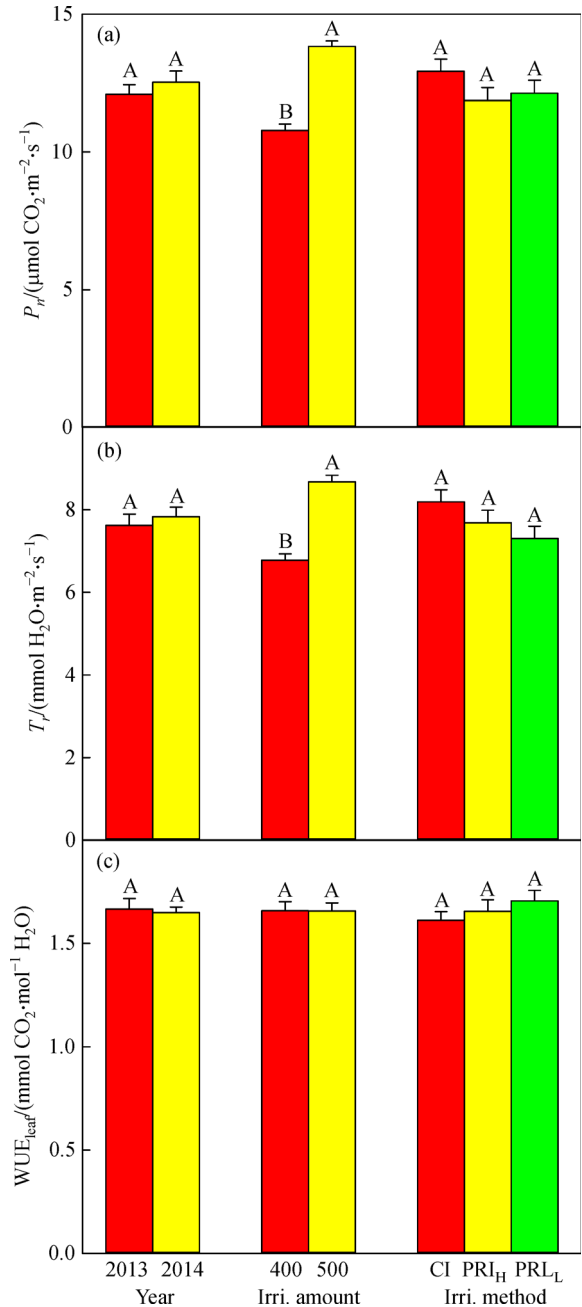


Fig. 5 Photosynthetic rate (P_n) (a), transpiration rate (T_r) (b) and leaf water use efficiency (WUE_{leaf}) (c) during the whole growth season as affected by year, irrigation amount and irrigation method. Bars labeled with different letters are significantly different ($P < 0.05$) and with the same letter are not significantly different ($P \geq 0.05$). Error bars indicate standard error of means. CI, conventional irrigation; PRI_H, alternate partial root-zone irrigation with high irrigation frequency; and PRL_L, alternate partial root-zone irrigation with low irrigation frequency.

processes^[27]. In addition, the slight reduction of photosynthetic and transpiration rates and the slight increase in leaf water use efficiency under APRI indicates that frequently alternating wetting and drying simulates roots, which would enhance the concentration of abscisic acid, and reduce unproductive water loss with the decrease in the vegetative growth^[14,33]. Compared with 400-mm irrigation, 500-mm irrigation significantly improved photosynthetic and transpiration rates (Fig. 5). This indicates that apple trees under 500-mm irrigation have sufficient water storage and suitable water status, which is advantageous for promoting leaf photosynthesis and transpiration, while apple trees under 400-mm irrigation are in water deficit stress, which will change cell structure and chloroplast hydration^[3,34], and reduce enzymatic activity to further decrease leaf photosynthesis and transpiration^[35]. However, irrigation amount had no significant impact on leaf water use efficiency. This indicates that water percolation may have occurred with 500-mm irrigation due to the lower water holding capacity of the sandy soil in the orchard^[36].

3.3 Midday stem and leaf water potentials

No significant interactions were found for experimental year, irrigation method and amount for midday stem and leaf water potentials (Table 3). In comparison with CI, PRI_H significantly reduced the midday stem and leaf water potentials from 0.05 to 0.10 MPa (Table 3; Fig. 6), which was lower than previously reported values^[12,37]. The primary reason for this could be the different irrigation methods and amounts used in those studies. For example, deficit irrigation could result in severe water deficit stress substantially reducing stem and leaf water potentials^[37]; while APRI could cause slight water deficit stress with only a slight reduction of stem and leaf water potentials (Fig. 6). In addition, the greater reduction of irrigation amount under APRI would also result in water deficit stress substantially decreasing water potential^[12]. In this study, the reduction of water potential was slight with APRI maintaining the same irrigation amount as CI (Fig. 3b). This further indicates that the trees under APRI did not suffer physiological water shortage (Fig. 6). In addition, no significant difference was found for midday stem and leaf water potentials between APRI at different irrigation frequencies (Fig. 6). In comparison with 400-mm irrigation, 500-mm irrigation significantly increased the midday stem and leaf water potentials (Fig. 6) due to abundant water storage and suitable water status of the trees under 500-mm irrigation^[34].

In summary, the promotion of root growth and the reduction of unproductive water loss are important approaches for saving water in arid areas^[12]. However, conventional irrigation always tends to increase luxury transpiration without obvious improvement in root water uptake capacity. In contrast, APRI with high irrigation

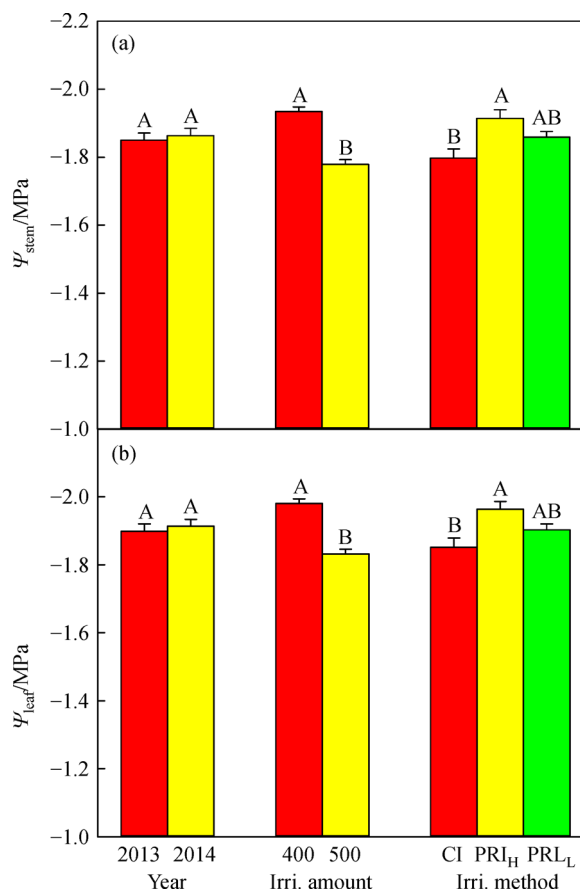


Fig. 6 Midday stem and leaf water potentials [Ψ_{stem} (a) and Ψ_{leaf} (b)] during the whole growth season as affected by year, irrigation amount and irrigation method. Bars labeled with different letters are significantly different ($P < 0.05$), and with the same letter are not significantly different ($P \geq 0.05$). Error bars indicate standard error of means. CI, conventional irrigation; PRI_H, alternate partial root-zone irrigation with high irrigation frequency; and PRI_L, alternate partial root-zone irrigation with low irrigation frequency.

frequency reduces tree luxury transpiration and enhances water use efficiency^[27,31]. In addition, the slight water deficit stress under APRI with high irrigation frequency would further promote root growth^[14,33], expand water uptake capacity^[24] and reduce unproductive water consumption^[30].

4 Conclusions

Our results demonstrate that, compared with conventional irrigation of apple trees, APRI with high irrigation frequency significantly increased root length density, reduced midday stem and leaf water potentials and leaf stomatal conductance, while there was no significant impact of APRI irrigation frequency on the above parameters. However, a significantly positive relationship between stomatal conductance and root length density was found for APRI between the different irrigation frequen-

cies. Therefore, the mechanism of efficient water use in apple trees in arid areas could be that APRI with high irrigation frequency promotes root growth, enhances water uptake capacity and decreases unproductive water loss by apple trees. However, more research is required for a better understanding of efficient water management in the arid apple production area.

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Compliance with ethics guidelines Shaoqing Du, Ling Tong, Shaozhong Kang, Fusheng Li, Taisheng Du, Sien Li, and Risheng Ding declare that they have no conflicts of interest or financial conflicts to disclose.

This article does not contain any studies with human or animal subjects performed by any of the authors.

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