Scientia Horticulturae 145 (2012) 76–83

Contents lists available at SciVerse ScienceDirect

Scientia Horticulturae

journal homepage: www.elsevier.com/locate/scihorti

Differential responses of stomatal morphology to partial root-zone drying and deficit irrigation in potato leaves under varied nitrogen rates

Fei Yan[a,b,c,d], Yanqi Sun[c,e], Fengbin Song[a,b,*], Fulai Liu[c,**]

[a] Northeast Institute of Geography and Agroecology, Chinese Academy of Sciences, Changchun 130012, China
[b] Graduate University of Chinese Academy of Sciences, Beijing 100049, China
[c] University of Copenhagen, Faculty of Science, Department of Agriculture and Ecology, Højbakkegaard Allé 13, DK-2630 Taastrup, Denmark
[d] College of Plant Science, Jilin University, Changchun 130062, China
[e] College of Water Resources and Architectural Engineering, Northwest A&F University, Yangling, Shaanxi 712100, China

ARTICLE INFO

Article history:
Received 7 June 2012
Received in revised form 26 July 2012
Accepted 26 July 2012

Keywords:
ABA
Carbon isotope discrimination
Partial root-zone drying
Nitrogen
Stomatal aperture
Stomatal density

ABSTRACT

The effects of partial root-zone drying (PRD) as compared with deficit irrigation (DI) on stomatal morphology of potato (*Solanum tuberosum* L.) under varied nitrogen (N) rates were investigated. The plants were grown in split-root pots under three N rates, viz., 70 (N1), 125 (N2), and 200 (N3) mg N kg^{-1} soil, respectively. For each N rate, PRD and DI plants received the same amount of water, which allowed re-filling one half of the PRD pot to 100% water holding capacity. Across the three N rates, guard cell size was larger in DI than in PRD, whereas stomatal pore aperture area (*SA*) was similar between the two irrigation treatments. Stomatal density (*SD*) was affected by both N rate and irrigation treatment and was lower in PRD than in DI under N2 and N3, whereas the reverse was the case under N1. Plant leaf area increased with increasing N rate, but was unaffected by the irrigation treatment. *SD* positively correlated with leaf N concentration and xylem sap ABA concentration for the DI plants, but not for the PRD plants. Nonetheless, negative linear relationships of *SD* to the mean soil water content in the pots and the carbon isotope discrimination in the leaves were found across all treatments. Regression analyses showed that it was *SA* rather than *SD* positively correlated with the stomatal conductance and the transpiration rate per unit leaf area in the DI; however such relationships were not evident in the PRD. In conclusion, compared to DI, PRD led to a more conservative control in plant water use via modulating stomatal morphology; the smaller stomata combined with a lower *SD* in the plants had efficiently reduced plant water use under high N rate, which maintained a better soil water moisture condition in the PRD pots.

1. Introduction

Worldwide shortage of freshwater resource has stimulated research into development of water-saving agricultural practices aiming at producing ‘more crop per drop’ (Davies et al., 2011). In the last two decades, biological water-saving irrigation strategies such as deficit irrigation (DI) and partial root-zone drying (PRD) irrigation that exploit the plant drought adaptation mechanisms have been developed and have shown great potential to enhance crop water use efficiency (WUE) (Bacon, 2004; Davies and Hartung, 2004). The physiological basis for improving WUE under both PRD and DI involves utilizing the ABA-based root-to-shoot signaling system decreasing stomatal conductance (g_s) thereby curtailing transpiration rate during moderate soil drying (Stoll et al., 2000; Liu et al., 2006a,b; Dodd, 2009). Accumulated evidence indicates that, by alternately drying and wetting part of the root system, PRD induces much stronger ABA signal than does DI under a similar soil water deficit, which may result in a fine-tune stomatal control over plant water use and thus an increase in WUE (Dodd et al., 2008; Liu et al., 2009).

Stomatal conductance (g_s) is determined predominantly by the aperture of the stomatal pores (*SA*) as well as the density of stomata (*SD*) on the leaf surface. It can be computed by physically based formulas that take both *SA* and *SD* into account (Brown and Escombe, 1900; Parlange and Waggoner, 1970; Franks and Farquhar, 2001). Therefore, manipulation of both *SA* and *SD* may modify g_s thereby bringing about an increase of WUE (Wang et al., 2007). To date, research into the mechanisms by which water-saving irrigation strategies such as PRD and DI improve plant WUE has mainly been focused on the significance of the xylem-borne ABA signaling in regulating *SA* and thus g_s in a short-time scale (Stoll et al., 2000). However, little attention has been paid to the plasticity in *SD* in

* Corresponding author at: Northeast Institute of Geography and Agroecology, Chinese Academy of Sciences, Changchun 130012, China.
** Corresponding author. Tel.: +45 3533 3392; fax: +45 3533 3478.
E-mail addresses: songfb@neigae.ac.cn (F. Song), fl@life.ku.dk (F. Liu).

0304-4238/$ – see front matter © 2012 Elsevier B.V. All rights reserved.
http://dx.doi.org/10.1016/j.scienta.2012.07.026

response to the irrigation treatments and its significance in controlling plant water use in a long-term perspective (Shimada et al., 2011).

Even though both *SA* and *SD* can be modified by environmental cues, it is widely accepted that *SA* is more dynamic while *SD* is more static in response to changing environments (Franks and Farquhar, 2007). Thus, a temporal (minutes to hours) change of g_s for a leaf under changing environments will be due mainly to a change of *SA*; whereas plants exposed to varied environments for a longer period (e.g., weeks to months), alterations in both *SA* and *SD* may account for the change of g_s. Early evidence has shown that crop grown under deficit irrigation may modify both *SA* and *SD* to optimize g_s thereby improving WUE (Zhang et al., 2006). However, until now the underlying mechanisms of modifying *SD* in response to soil water deficits are not fully understood.

In the present study, potato plants (*Solanum tuberosum* L.) were grown in split-root pots under three N rates and were subjected to PRD and DI during tuber initiation and tuber bulking stages. Stomatal morphology in the abaxial leaf surface, transpiration rate per unit leaf area, g_s, N concentration and Δ^{13}C in the leaf biomass, ABA concentration in the xylem sap were determined. The objective was to investigate the relative significance of changes in *SA* and *SD* in controlling g_s under PRD and DI regimes with varied N rates. The possible mechanisms modifying *SD* across different water and N environments are discussed.

2. Materials and methods

2.1. Plant material and growth condition

The experiment was conducted from March to May, 2011 in a climate controlled greenhouse at the experimental farm of the Faculty of Life Sciences, University of Copenhagen, Taastrup, Denmark. Potato tubers (*S. tuberosum* L. cv. Folva) were planted in split-root pots (25.2-cm diameter and 40-cm tall) on 11th March. The pots were evenly separated into two compartments with plastic sheets such that water exchange between the two compartments was prevented. A section of the plastic sheet (width (5 cm) × height (10 cm)) was removed to allow a seed tuber to be planted in the top-center of the pots (see Fig. 1 – the split-pot system in Wang et al. (2010b)). The pots were filled with 20.2 kg of naturally dried soil with a bulk density of 1.14 g cm^{-3}. The soil was classified as sandy loam, having a pH of 6.7, total C 12.9 g kg^{-1}, total N 1.4 g kg^{-1}, NH_4^+-N 0.7 mg kg^{-1}, NO_3^--N 19.1 mg kg^{-1}. The soil was sieved passing through a 2 mm mesh and had a volumetric soil water content (vol.%) of 30.0% and 5.0% at water holding capacity and permanent wilting point, respectively. Before planting, the seed potatoes were exposed to 12–14 °C with constant dim overhead light (a PAR of 100–150 μmol s^{-2} s^{-1}) for sprouting. During planting only one sprout was retained. The roots from this sprout were evenly distributed between the two separated compartments. The average soil water contents in the pots were monitored by a time domain reflectometer (TDR, TRASE, Soil Moisture Equipment Corp., CA, USA) with probes (35 cm in length) installed in the middle of each soil compartment. The climate conditions in the greenhouse were set at: 16/14 ± 2 °C day/night air temperature, 15 h photoperiod and >500 μmol m^{-2} s^{-1} photosynthetic photon flux density (PPFD) supplied by sunlight plus metal-halide lamps.

2.2. N and irrigation treatments

Three N rates, i.e., 70, 125, and 200 mg N kg^{-1} soil, denoted as N1, N2 and N3, respectively, were included in the experiment. The N fertilizer supplied as NH_4NO_3 was mixed thoroughly with the soil before filling the pots. In addition, P and K were also applied

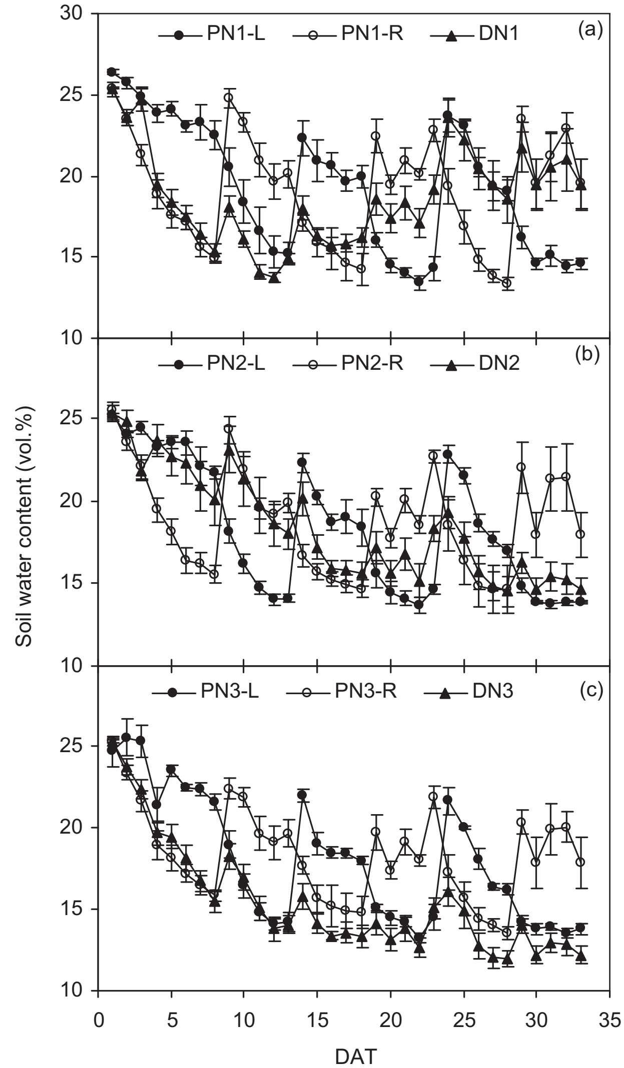

Fig. 1. Daily average soil water content (vol.%) in the pots of potato plants under PRD and DI irrigation in combination with three N rates (N1, a; N2, b; N3, c). PN1, PN2, PN3, DN1, DN2, and DN3 denote the combinations of PRD and DI with the three N-fertilization rates, respectively; PRD-L and PRD-R indicate the left and right sides of the PRD pots. Error bars indicates the standard error of the means (S.E.) (n = 4).

as KH_2PO_4 (380 mg kg^{-1} soil) and K_2SO_4 (130 mg kg^{-1} soil) into the soil to meet the nutrients requirement for plant growth. From tuber initiation to tuber bulking stages (30–64 days after planting), the plants were exposed to PRD and DI treatments. In PRD, one soil compartment was watered daily at 18:00 h to an average soil water content of 28% while the other was allowed to dry until the average soil water content in the pots reached ca. 12–15%, then the irrigation was shifted to the dry compartment; in DI, the same amount of water used for the PRD plants was irrigated evenly into the whole pot. Here, we did not include a fully irrigated control since the main purpose of the current study was to exploit the possible mechanisms by which the PRD treatment over-performs DI in terms of improving WUE as reported in earlier studies (Liu et al., 2006a,b).

The experimental set-up was a complete factorial design comprising six treatments and each treatment had four replicates. The irrigation water was tap water with negligible concentration of nutrients. The irrigation treatments lasted for 5 weeks, during which each soil compartment of the PRD pots had experienced 3 drying/wetting cycles. The changes of daily average soil water content in the soil compartments/pots during the treatment period are shown in Fig. 1. Since different N rates significantly affected