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# Above- and below-ground growth of cotton in response to drip irrigation under mulch film

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### Abstract

Long-term use of drip irrigation technology may lead to root degradation and affect shoot growth and yield. A field experiment was conducted to investigate above- and belowground growth responses to drip irrigation under mulch film (DI) in comparison to flood irrigation under mulch film (FI) in cotton. The monolith method was used to harvest roots at seven timepoints in the growth periods, and the root length, and shoot and root dry weight were measured. The total root length per plant in the 0–10 cm soil layer was higher under DI, whereas in the 30–60 cm soil layer roots were longer under FI. From 65 to 96 days after sowing (DAS), the rate of increase in root length was lower under DI than FI. Total root length decreased after 125 DAS under DI, and was mainly centered in the 0–40 cm soil layer and at distances of 30–70 cm from drip-lines. The shoot:root ratio at 125 DAS was higher under DI than FI, but at 160 DAS the shoot:root ratio abruptly declined under DI. The decline in root length under DI during advanced growth stages may be attributable to the higher root density in shallow soil layers and the increase in the shoot:root ratio. These results suggest that, it are important to increase yield of cotton plants under DI early development of a deep root system and initial control of shoot growth by regulation of water and fertilizer supply.

**Keywords:** Cotton, Drip irrigation, Flood irrigation, Mulch film, Root. **Abbreviations:** DI\_drip irrigation under mulch film; FI\_flood irrigation under mulch film; DAS\_days after sowing.

#### Introduction

Water shortage is a common problem in agricultural production worldwide and is one of the most important ecological factors limiting crop productivity (Ali et al., 2009). Therefore, water-saving measures, such as drip irrigation (Bhattarai et al., 2008), are increasingly utilized in agricultural production. In China, arid and semi-arid regions account for 52.5% of the country's total land area (Zhang and Xu, 2011), of which Xinjiang is an irrigated arid area. Drip irrigation was first applied for agricultural production in Xinjiang in the early 1990s, and DI technology was developed from practical experiences in the field (Hu and Li, 2003).

The DI technique is beneficial for water and fertilizer utilization because of the drip irrigation strategy and the warming effect of mulch film. In cotton fields, for example, irrigation and nitrogen applications were reduced by about one-third and one-fifth, respectively, under DI compared with flood irrigation (Zhang et al., 2002). However, long-term use of drip irrigation can cause negative impacts on root systems of crops (Carmi et al., 1992; Carmi et al., 1993; Bhattarai et al., 2008), such as reduction in taproot length and density of root growth in shallow soil layers (Klepper, 1991; Wei et al., 2002). The root system of plants cannot respond rapidly to environmental change. However, the rhizosphere environment is variable and the soil moisture, temperature, pH and other environmental factors often vary at different growth stages or in different soil microenvironments (Afshari et al., 2011), thus nutrient or water stress on root growth might occur even in

high-input conditions. For example, the mass flow of nitrate may be affected greatly by short-term water shortage and under such conditions the root system may not be able to respond effectively to a variety of potential stresses (Zhang et al., 2009). Because of root intgrating shoot growth (Gregory and Eastham, 1996), degradation of the root system is likely to impact on normal shoot growth and crop yield. Consequently, studies on root system dynamics are important to help to achieve balanced supply of water and fertilizer.

Many studies have investigated plant shoot structure and function (Gregory, 2006). Studies on cotton root systems are greatly outnumbered by those on aboveground parts because the soil restricts root observation and the study of plant roots is difficult under field conditions (Lynch, 1995). Few studies have investigated root development under DI conditions.

The main objective of the present study was to explore the spatial dynamics of root and aboveground growth in response to DI in a field trial.

# Results

#### Changes in root length under DI and FI

On the basis of total root length per plant four stages in the cotton growth period were distinguished an initial period of gradual growth, followed by a phase of rapid growth, a period of relative stability, and finally a decline phase (Fig 3). Cotton

Table 1. Timing of water and nitrogen fertilizer applications in the drip irrigation (DI) and flood irrigation (FI) treatments after

sowing of cotton seeds on 25 April.

Treatment		Total	Date of application									
			12 Jun	19 Jun	26 Jun	3 Jul	10 Jul	17 Jul	24 Jul	1 Aug	8 Aug	16 Aug
Water	DI	4000	260	300	420	500	500	500	500	400	340	280
$(m^3 hm^{-2})$	FI	6000		1200			1800		1800		1200	
Nitrogen	DI	280		10	20	30	40	40	40	40	30	30
$(kg N hm^{-2})$	FI	200		48					100		52	

root systems grew significantly faster under FI than DI in the period of rapid growth. Consequently total root length peaked about 15 d earlier under FI than that under DI. Compared with the FI treatment, cotton root length decreased after 100 DAS under DI, particularly from 125 DAS to 160 DAS, whereas the total root length under FI did not change over the same period. The length of cotton roots in the different soil layers before 82 DAS showed no significant difference between the two treatments (Fig 4). The root length in the 0-60 cm soil depth at 82 DAS was significantly higher under FI than that under DI, especially in the 30-60 cm soil layer, which indicated this period was critical for root growth under FI. The root length in the 0-10 cm soil layer was significantly higher under DI than that under FI after 82 DAS, whereas in the 30-60 cm soil layer root length was significantly higher in the FI treatment. From 120 DAS to 160 DAS, the root length decreased in the 0–10 cm soil layer under both DI and FI, and decreased significantly in the 20-40 cm layer under DI, whereas under FI almost no change in the 20-40 cm layer was observed over the same period. With regard to the change in cotton root length at different soil depths under DI and FI from 125 to 160 DAS, three soil layers were distinguished on the basis of root length variation at different soil depths: surface (0-10 cm), middle (10-40 cm) and lower (40-60 cm) layers. Roots in the surface layer decreased in length under both DI and FI, although the degree of decrease differed between the treatments; root length in the FI treatment decreased by 7.7% from 125 to 160 DAS, whereas the percentage decrease was higher (13.8%) under DI, which may be because of the higher root density in the surface soil layer under DI than that under FI (Fig 4). During the same period, changes in root length in the middle soil layer differed from those in the surface layer; root length under FI increased (5.6%), but significantly decreased by 22% under DI. In the lower soil layer, the root length increased by 10.16% and 36.01% under DI and FI, respectively. The region of decline in root length was in the 0-40 cm layer under DI, and at distances of 20-70 cm from drip-lines, but the most obvious changes were at distances of 30 cm and 50 cm from drip-lines, and between adjacent mulch films. The root length density was higher in the surface soil layer (0-10 cm) under DI than that under FI (Fig 5). The root density was higher in the lower soil layer (40-60 cm) under FI than that under DI. From 125 to 160 DAS, the root density decreased in the 20-30 cm soil layer under DI and at distances of 30-70 cm from drip-lines, which further indicated the cotton roots of plants grown under DI declined in the middle soil layer. In the same period, the root density increased under FI in this area.

# Responses of shoot and root growth to DI and FI

No difference in the biomass of reproductive organs under DI and FI was observed before110 DAS (Fig 6), whereas the biomass of vegetative organs under DI was higher than that under FI at the same timepoints. At 125 DAS, vegetative and reproductive organ biomass under DI was significantly higher than that under FI, whereas root biomass showed the opposite trend. This result indicated that the smaller root system of cotton plants grown under DI was required to support the water and nutrient demands of a larger shoot system and consequently, the shoot:root ratio under DI was significantly higher than FI at 125 DAS (Fig 6D). The vegetative organ:root ratio under DI was significantly higher than under FI before 120 DAS, whereas the opposite trend was observed after 120 DAS (Fig 7). The reproductive organ:root ratio before 110 DAS showed no significant difference between the two treatments. However, the reproductive organ:root ratio under DI was significantly higher than that under FI after 110 DAS. Thus, under DI, cotton plants mainly produced vegetative growth at early stages, and subsequently mainly produced reproductive growth. In addition, the small root system of cotton plants at late stages of growth under DI is mainly responsible for meeting the high water and fertilizer requirements of reproductive organs. The shoot dry weight of plants grown under DI abruptly decreased from 125 to 160 DAS. Field observations indicated the main reason for the decrease was the shedding of a large number of leaves, squares and bolls. The shoot:root ratio under DI also decreased to the same level as that of the FI treatment. In the same period, shoot dry weight and the shoot:root ratio both increased under FI.

# Discussion

#### Environmental effects on cotton root growth

In the present study, cotton root length abruptly declined from 125 to 160 DAS under DI, whereas no change occurred under FI (Fig 3). The decline in the DI treatment might be attributable to the differences in environmental conditions. Cotton root development under DI showed the following characteristics: the root distribution is concentrated in shallow soil layers (0-20 cm), and fewer roots occur at lower soil depths; in addition, development of numerous fine roots with the change in environment after cessation of water and fertilizer supply (at 125 DAS) was implied, because root dry matter was significantly lower under DI than that under FI, but almost no change in root length occurred. Under DI root growth and development would be more susceptible to declines in soil moisture, nutrients and temperature because of the shallow distribution of the root system (Zhang and Cai, 2005). In addition, because more squares and bolls develop under DI (data not shown), greater quantities of nutrients and carbohydrates will be transported to the reproductive organs during advanced growth stages, whereas carbohydrate supply to the roots will be reduced. Together with increased root degradation by rhizosphere insects and parasites (Wiesler and Horst, 1993; Eissenstat et al., 2000), the decline in root biomass would more rapid because of greater fine-root formation under DI. The decline in cotton root length during advanced growth stages under DI also might partly reflect local accumulation of salt. The soil at the experimental site has a naturally high salt



Fig 1. Plant spacing and irrigation system used in the experiment. The cotton image was obtained from the College of Resources and Environment, China Agricultural University.



**Fig 2.** Schematic illustration of the root excavation method. A monolith method was used to harvest roots at 40, 65, 82, 96, 110, 125, and 160 DAS. Soil cubes with 10 cm sides  $(1000 \text{ cm}^3)$  were dug individually in a soil volume of 70 cm × 20 cm and to a depth of 60 cm surrounding four plants. The total number of monoliths excavated for the four plants was 84.

content (EC 2.38 mS cm<sup>-1</sup>, soil:water 1:1). Soil salinity migration shows the following characteristics under DI: in the horizontal direction, salinity migrates to the region between adjacent mulch films and accumulates in the upper soil layer (0-10 cm); in the vertical direction, the salt content of the tillage layer (0-30 cm) decreases, because it is maintained in a moist state, therefore soil salt migrates and accumulates below the tillage layer (Zhou et al., 2006; Malash et al., 2008). Most cotton roots were distributed in the surface soil layer under DI and more fine roots were indicated to form under DI than FI (because at 125 DAS, the root dry matter was significantly lower under DI than that under FI, but almost no change in root length occurred). Thus, the roots located between adjacent mulch films might be more susceptible to death as a result of salt accumulation, but this hypothesis requires further study. Previous studies have found that root growth shows seasonal variation. For example, Gregory et al. (1997) studied root growth of several cereal crops over 6 years in northern Syria. It was shown that, under the same soil conditions, root mass differed between seasons; seasonal rainfall had a significant effect on root growth, and the highest root biomass coincided with the season of highest rainfall, whereas the lowest root mass corresponded with the season of least rainfall. A study at Breda in northern Svria also found significant differences in barley root dry matter in different growing seasons (Gregory et al., 1994).

# Root system development under DI

Providing an adequate supply of water and fertilizer for shoot development while simultaneously ensuring normal growth of the root system is difficult under DI. Therefore, careful regulation of water and fertilizer supply is important to achieve a high yield while stimulating development of an architecturally balanced root system. The cotton root system under FI was healthier and better able to meet the nutrient and water needs of the shoot system throughout the plant's life cycle than in plants grown under DI. The nutrient and water demands of shoots increased rapidly with square and boll formation in reproductive growth stage, and if shoots are unable to meet the energy demands of the roots, carbohydrate transport to the root system is reduced. Therefore, it is very important for cotton plants to build a well-developed root system before the plants enter the transition from vegetative to reproductive growth. The maximum root length approximately coincided with the onset of flowering under FI, whereas total root length peaked after the start of flowering under DI. Although the nutrient and water requirements of cotton plants will be lower during advanced growth stages, it is still necessary to maintain relatively high root activity for normal boll opening and to achieve a high yield. Considerably more roots were distributed at deep soil layers under FI than DI, which is important for resistance to stressful conditions such as drought or low temperature, and will enable the plant to utilize nutrients and water in deep soil layers. To achieve development of a healthy root system, cultivation of cotton under DI should focus on the following two aspects. First, a large and deep root system should be developed as quickly as possible and shoot growth should be controlled before the plants enter the reproductive growth stage. As mentioned above, excessive shoot growth is the most important reason for decline of cotton root systems under DI. Therefore, carbohydrate allocation during early cotton growth and development should be regulated to promote root growth. Second, with regard to root architecture, it is beneficial to stimulate root development in deeper soil layers to maintain the vitality of the root system during advanced growth stages, and so that the nutrient and water needs of the shoots can be met. In our experiment, the root length declined in the 0-10 cm soil layer under FI from 125 to 160 DAS, but the roots in the 10-60 cm soil depth continued to grow, and thus the root system was able to compensate for the decline in root density in shallow soil layers. Under DI, root length declined in the 0-40 cm soil layer, but increased in the 40-60 cm layer. Therefore, roots in the deeper



**Fig 3.** Total root length per cotton plant throughout the growth period under FI and DI. The growth period was divided into four stages based on the changes in total root length. The arrow indicates the onset of flowering. Error bars represent the standard error of the mean (n = 3).



**Fig 4.** Total root length at different soil depths on different days after sowing (DAS) of cotton plants grown under FI and DI.

soil layers will maintain higher vitality at advanced growth stages. Thus, development of a deep root system not only prevents reliance on absorption of nutrients and water from shallow soil layers, but at advanced growth stages the plant is able to obtain water and nutrients from deeper soil layers to increase productivity.

#### Materials and methods

### Study site

The field experiment was conducted at the Korla Experimental Station of the Xinjiang Academy of Agricultural Sciences, China, in the 2009 cropping season. The site has an arid climate typical of the area with 56 mm average annual rainfall , 2497 mm annual evaporation , 2878 h annual sunshine hours ,  $4252^{\circ}$ C of  $\geq 10^{\circ}$ C accumulated temperature , and 205 frost-free days per annum. The soil type at the study site is a sandy loam soil. Physicochemical properties of the soil were analyzed before sowing. The chemical properties of the 0–30 cm soil



**Fig 5.** Projection of root length density distribution in the soil profile at different growth stages under DI and FI. The wireframe diagrams were constructed with Surfer 7.0 (Golden Software, 2002). From 125 to 160 DAS, the root density decreased in the 20–30 cm soil layer under DI and at distances of 30–70 cm from drip-lines, in the same period, the root density increased under FI in the same areas.

layer were:  $NO_3^{-}-N$  36.54 mg kg<sup>-1</sup>,  $NH_4^{+}-N$  6.53 mg kg<sup>-1</sup>, pH (H<sub>2</sub>O) 8.0, soil density 1.33 g cm<sup>-3</sup>, Olsen-P 2.88 mg kg<sup>-1</sup>, NH<sub>4</sub>OAc-extracted K 187.5 mg kg<sup>-1</sup>, and organic matter 7.65 g kg<sup>-1</sup>. The soil had a high salt content with electrical conductivity (EC; water:soil 1:1) of 2.38 mS cm<sup>-1</sup>. Thus the soil is severely saline according to the classification standards proposed by Wang (1993).



Fig 6. Changes in cotton shoot (vegetative and reproductive organs) and root biomass and the shoot:root ratio on different days after sowing (DAS) under DI and FI.



Fig 7. Changes in cotton nutrition organ:root ratio and reproductive organ:root ratio on different days after sowing (DAS) under DI and FI.

# Experimental design and plant material

The experiment included two treatments i.e. drip irrigation under mulch film (DI) and flood irrigation under mulch film (FI). A randomized block design with three replicates was used. In total there were six plots each  $10 \text{ m} \times 12 \text{ m}$  in area were used. Seeds of cotton (Gossypium hirsutum L.) cv. XLZ9 were obtained from the Xinjiang Academy of Agricultural Sciences, China, and were sown on 25 April, 2009, at a density of 220,000 plants ha<sup>-1</sup>. Two rows of plants (with 20 cm spacing between rows) were sown on either side of the irrigation drip-line, with 50 cm spacing between the first row and the drip-line, and spacing of 10 cm between plants within a row (Figure 1). The volume of water supplied by DI and FI during the experiment was 4000 m<sup>3</sup> hm<sup>-2</sup> and 6000 m<sup>3</sup> hm<sup>-1</sup> respectively Which are irrigation rates commonly applied in the local area. Water was supplied on 10 dates at weekly intervals for DI beginning from 12 June, and on four dates at 2-3 weekly intervals for FI (Table 1). Irrigation was last applied on 16 August and 8 August under DI and FI, respectively.Urea was applied as fertilizer at rates of 350 kg N hm<sup>-2</sup> and 400 kg N hm<sup>-2</sup>, of which 20% and 50% was applied before sowing as base fertilizer under DI and FI, respectively, and the remainder

was applied with irrigation (Table 1). In addition,  $150 \text{ kg P}_2\text{O}_5 \text{ hm}^{-2}$  (superphosphate) and  $150 \text{ kg K hm}^{-2}$  (potassium chloride) were applied before sowing as base fertilizer.

# Root and shoot harvest

Four uniform cotton plants were selected, and aboveground parts were excised at ground level and divided into stems, leaves and reproductive organs. All samples were dried at  $105^{\circ}$ C for 30 min, then oven-dried at 70°C for 3 days and weighed. A monolith method (Böhm, 1979) was used to harvest roots at 40, 65, 82, 96, 110, 125, and 160 days after sowing (DAS). Soil cubes with 10 cm sides (1000 cm<sup>3</sup>) were dug individually in a soil volume of 70 cm × 20 cm and to a depth of 60 cm surrounding four plants (Fig. 2). The total number of monoliths excavated for the four plants was 84. Each soil block was placed in a separate plastic bag and labeled with the spatial coordinates. Roots were sieved with a stainless steel mesh (1 mm diameter) and rinsed with water to remove the soil from the roots.

### Root length measurement and dry weight analysis

Roots collected from each soil block were scanned with a digital scanner (Epson V700, Djakarta, Indonesia) at 200 dpi with grayscale pixels and saved as TIF image files. For scanning, the root sample was placed in a glass rectangular dish (200 mm  $\times$  150 mm) containing a layer of water 4–5 mm deep to untangle the roots and minimize root overlap. When necessary, the roots of one soil block were separated into subsamples until they could be placed in the dish. The images were analyzed using DELTA-T SCAN version 1.0 software (Delta-T Devices, Burwell, UK).

The total root length per plant (m plant<sup>-1</sup>) was calculated by dividing the total root length for the 84 soil blocks by four (the number of plants sampled). Root length density was presented as wireframe diagrams constructed with Surfer 7.0 (Golden Software, 2002).

After scanning, the roots were oven-dried at 70°C for 3 days and weighed.

#### Conclusion

The growth and development of cotton roots is strongly affected by environmental conditions. In response to DI the highest root density of cotton plants was in the upper soil layers (0–20 cm), and fewer roots developed at deeper soil layers. Higher shoot and lower root dry matter were observed at advanced growth stages under DI than under FI. Root length showed a significant decline at advanced growth stages under DI, which would have a negative impact on aboveground growth and development. Therefore, to increase yield of cotton plants under DI, early development of a deep root system and initial control of shoot growth by regulation of water and fertilizer supply are important.

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