INTERNATIONAL JOURNAL OF AGRICULTURE & BIOLOGY
ISSN Print: 1560–8530; ISSN Online: 1814–9596
19F–195/2020/24–1–68–76
DOI: 10.17957/IJAB/15.1410
http://www.fspublishers.org

Full Length Article

Fine Root Turnover Characteristics of Alfalfa under Drip Irrigation at different Phosphorus Levels

Qianbing Zhang, Junwei Zhao, Shengyi Li, Weihua Lu, Xuzhe Wang and Chunhui Ma*
The College of Animal Science & Technology, Shihezi University, Shihezi, 832003, Xinjiang, China
For correspondence: qbz102@163.com; chunhuima@126.com
Received 11 December 2019; Accepted 31 January 2020; Published 20 April 2020

Abstract

The present study investigated fine roots turnover characteristics of alfalfa under drip irrigation when different phosphorus levels were applied (0, 50, 100, and 150 kg ha^{-1}); including production and mortality were evaluated for two years (2017 and 2018). The results showed that the total hay yield of alfalfa at 100 kg P ha^{-1} was significantly higher than control and 50 kg P ha^{-1} treatments in two years (P < 0.05). There were two peaks of fine root standing crop and production, respectively, which reached the maximum at 100 kg P ha^{-1}. With the increase of soil depth, the fine root standing crop of alfalfa decreased gradually under different P treatments. Fine root production at 100 kg P ha^{-1} was significantly higher than control and 50 kg P ha^{-1} treatments. Fine root mortality at 100 and 150 kg P ha^{-1} treatments were significantly higher than control and 50 kg P ha^{-1}. The turnover rate at 100 kg P ha^{-1} treatment reached the maximum in two years at 0.2134 yr^{-1} and 0.1814 yr^{-1}, respectively. Pearson correlation analysis showed that annual total hay yield of alfalfa and annual production of fine root had significant correlation (P < 0.01). At 100 kg P_2O_5 ha^{-1}, the turnover rate of fine roots in alfalfa was improved, and fine root production of alfalfa significantly affected the above dry matter yield. © 2020 Friends Science Publishers

Key word: Alfalfa; Fine root standing crop; Production; Mortality; Turnover rate

Introduction

Root system, as an important functional organ of plants, provides material basis for plant growth and regeneration, and directly affects plant performance (Jha and Mohapatra 2010). Fine roots generally refer to roots less than 2 mm in diameter (Finér *et al.* 2011; Liu *et al.* 2014). These are the most active part of the underground part of the crop, which absorb nutrients and water. It has a huge surface area, strong physiological activity, and strong response to the availability of soil resources (Bennett *et al.* 2002). Although, the proportion of fine roots in total root system is not high, fine roots can acquire nutrient resources and release organic matter into the soil. Meanwhile, fine roots play an important role in energy and material flow in the biosphere. These can not only transfer carbon into underground carbon pools, but also promote plant uptake of water and nutrients (Pregitzer *et al.* 2002). Moreover, fine roots can return nutrients fixed by photosynthesis to the soil, which can make plants grow sustain ably. Fine root turnover is an important component of material cycle and energy flow in ecosystem (Majdi 2001). Nutrients and organic matter enter soil through fine root turnover are important sources of soil fertility maintenance (Lee and Jose 2003). It plays an important role in restoring and increasing soil fertility, improving plant nutrition and primary productivity. Therefore, the study on fine root turnover of alfalfa is of great significance to clarify the above-ground yield formation of alfalfa.

Alfalfa (*Medicago sativa* L.) is perennial high-quality leguminous forage, known as the "King of Forage" (Yacoubi *et al.* 2011). The growth, death and turnover of fine roots of alfalfa are closely related to the external climatic and soil environmental conditions. Soil nutrient directly affect the physiological activity of fine roots and the distribution of carbohydrates, thus affecting the production and turnover of fine roots of alfalfa (Zhu *et al.* 2013). Thus, had great significance to the growth and development of alfalfa, productivity and the flow of matter and energy in ecosystems (Chang *et al.* 2012).

When the availability of soil resources increased, it could promote the growth and biomass accumulation of fine roots, and the ability to absorb water and nutrients of fine roots would also be enhanced (Zhang *et al.* 2019). The results showed that the improvement of soil water and nutrient availability could stimulate the growth, increase the biomass of fine roots, branching of lateral roots, and then increase account of new roots, meanwhile, it can prolong or shorten the life span of fine roots, and increase the respiration rate (Williamson *et al.* 2001). Phosphorus is a large number of elements needed for alfalfa growth and its

To cite this paper: Zhang Q, J Zhao, S Li, W Lu, X Wang, C Ma (2020). Fine root turnover characteristics of alfalfa under drip irrigation at different phosphorus levels. *Intl J Agric Biol* 24:68–76

application can accelerate cell division, promote root and above ground growth, thereby promoting the transport of photosynthate in plant leaves, and then increase the above ground yield (Singh and Reddy 2014). Other studies found that fine root C/P ratio was positively correlated with phosphorus use efficiency of above ground litter (Okada *et al.* 2017). It has become an important nutrient limiting the growth of plants and soil organisms in terrestrial ecosystems (Vitousek *et al.* 2010). Studies have shown that the application of appropriate phosphorus can improve the extracellular enzymes involved in decomposition, which is conducive to the degradation of cellulose and lignin in fine roots of crops, thus promoting the decomposition of fine roots of crops (Cormier *et al.* 2015). Fertilization can increase soil enzyme activity, which is beneficial to the decomposition of fine roots and soil organic matter (Jiang *et al.* 2014).

Minirhizotron technology is a method of recording and studying plant roots by scanning images. Its greatest advantage is to dynamically monitor the underground parts of the same sample at different times in a fixed point, continuous, periodic and non-destructive manner. This technique has been widely used to study the growth and death dynamics of fine roots and estimate their turnover. It has been successfully applied in alfalfa fine root turnover research (Ren *et al.* 2015).

There are continuous material circulation and energy flow between root system and soil, which fundamentally affect the process of nutrient utilization by crops. At present, the research on fine roots of alfalfa mainly focused on the biomass, spatial distribution and dynamic change in soil. Production and death cycle of alfalfa fine root under phosphorus treatment was relatively few, especially the study on the characteristics of fine root turnover under drip irrigation with different phosphorus application rates are rarely reported, and the relationship between the indexes of fine root of alfalfa under drip irrigation was still unclear. Therefore, this experiment studied the growth and turnover of fine roots of alfalfa under different P treatment and drip irrigation conditions, to understand the growth and death dynamics and spatial distribution characteristics of fine roots of alfalfa, and clarified the dynamic characteristics of fine roots rotation of alfalfa. To provide theoretical basis for the study of nutrient uptake by drip alfalfa under irrigation fine root rotation, the high quality and efficient production of alfalfa, the relationship between phosphorus application and fine root turnover was clarified in Xinjiang oasis region of China.

Materials and Methods

Site description

Field experiments were conducted in 2017 and 2018 at the agricultural demonstration park of the Tianye Group Agricultural Research Institute, Shihezi, Xinjiang, China (44°26'N, 85°95'E). The maximum temperature occurs in July, and the highest precipitation in May in 2017 and 2018 (Fig. 1). The annual number of sunshine hours ranged from 2721~2818 h, and the soil type was grey desert soil. The physical and chemical properties of the 0~20 cm plough layer soil are shown in Table 1.

Experimental design

This experiment adopted single factor randomized block design with phosphorus fertilizer rates: 0, 50, 100 and 150 $kg{\cdot}ha^{-1}$. The phosphorus fertilizer was mono ammonium phosphate (total P_2O_5 52%) with three repetitions. Fertilizer was dripped with water at the branching stage, first cut, sec cut and third cut 3~5 days after cutting.

In this experiment, uninoculated WL354HQ alfalfa seeds (Beijing Zhengdao Ecology Technology Co., Ltd.) were sown on April 19, 2015 with spacing of 20 cm, the sowing depth of 1.5~2.0 cm using 18 $kg{\cdot}ha^{-1}$ seed rate in a plot with area of 5 m x 8 m. 1 m wide walkway was set up between the districts to prevent water infiltration between residential areas. The drip irrigation belt was buried at the shallow 8~10 cm surface soil with a spacing of 60 cm. The working pressure of the drip irrigation belt was 0.1 MPa with the diameter of 12.5 mm, the dripper flow of 1.1 $L{\cdot}h^{-1}$, and the dripper spacing of 20 cm. An internal-embedded drip irrigation belt was used and the distance of the drop head was 20 cm. In addition to water factors, other weeding management was carried out in accordance with the local farmland according to the field growth.

Minirhizotron installation and data acquisition

The growth and death of alfalfa roots were continuously observed in 2017 and 2018 by CI-600 root monitoring system (CID BIO-Science, United States), and the minirhizotron were installed in the experimental field in April 10, 2016. A total of 12 minirhizotrons were buried in each plot. According to Johnson *et al.* (2001) and other methods, minirhizotrons were installed in the center of the experimental plot (Fig. 2). The length of minirhizotrons was 1 m, and the installation angle was 45° with the ground. The exposed part of the soil surface was about 12～15 cm, and the vertical depth was about 60 cm. The matching plastic cap of the minirhizotron was applied to the mouth of the minirhizotron. The exposed part of the minirhizotron was wrapped in two layers with black plastic bags and fastened with rubber band and tape to prevent exposure of the scanner during root measurement, dust and water entering the inner wall of the minirhizotron after the minirhizotron cover slipped or damaged. Then a wooden stick was inserted about 1 m long into the position 10 cm away from the minirhizotron in order to find the location of the minirhizotron intuitively during observation, and prevent the human inadvertent destruction of the minirhizotron body during alfalfa cutting.

Table 2: Fine root crop of alfalfa in different soil layers (cm·cm^{-3})

Soil depth (cm)	Phosphorus treatments	2017 22/5	2017 21/6	2017 21/7	2017 22/8	2017 23/9	2018 23/5	2018 24/6	2018 24/7	2018 23/8	2018 24/9
0～20	0 kg· ha^{-1}	0.1835c	0.2359d	0.1955d	0.2495c	0.2431d	0.1952d	0.2266d	0.202d	0.2414c	0.2191d
	50 kg· ha^{-1}	0.196b	0.2623c	0.2391c	0.2778b	0.2809c	0.2313c	0.2808c	0.2262c	0.2942b	0.2597c
	100 kg· ha^{-1}	0.2411a	0.3992a	0.3318a	0.3413a	0.3327a	0.2770b	0.3356a	0.2938a	0.3335a	0.3251a
	150 kg· ha^{-1}	0.2461a	0.3484b	0.2832b	0.3335a	0.3058b	0.2942a	0.3146b	0.2812b	0.3230a	0.2807b
20～40	0 kg· ha^{-1}	0.0891c	0.1746c	0.1982d	0.1855d	0.1642c	0.0886d	0.1555d	0.1786d	0.1723d	0.1499c
	50 kg· ha^{-1}	0.0933c	0.1928b	0.2064c	0.1999c	0.1688c	0.1351c	0.1865c	0.2022b	0.1894c	0.1645b
	100 kg· ha^{-1}	0.1960a	0.2817a	0.2628a	0.2693a	0.2076a	0.1975a	0.2565a	0.2142a	0.2483a	0.1996a
	150 kg· ha^{-1}	0.1768b	0.1940b	0.2221b	0.2295b	0.1807b	0.1792b	0.2142b	0.1933c	0.2309b	0.1954a
40～60	0 kg· ha^{-1}	0.0678c	0.0905c	0.0884b	0.0944b	0.0604b	0.0712b	0.0924d	0.0857c	0.092c	0.0671b
	50 kg· ha^{-1}	0.0771b	0.100ab	0.0935b	0.1219a	0.0631b	0.0974a	0.1099b	0.1006b	0.1204b	0.0927a
	100 kg· ha^{-1}	0.0807b	0.1061a	0.1122a	0.1262a	0.0866a	0.0973a	0.1313a	0.1626a	0.1297a	0.0954a
	150 kg· ha^{-1}	0.1126a	0.0952bc	0.0926b	0.1267a	0.0800a	0.0920a	0.1007c	0.094b	0.1148b	0.0992a

The different small letter in column is significant ($P < 0.05$). The same

Table 1: Soil physicochemical properties (0–20 cm depth) at the experimental station

Years	Bulk density (g cm^{-3})	Available P (mg kg^{-1})	Organic matter (g kg^{-1})	Alkaline N (mg kg^{-1})	Total P (g kg^{-1})	Available K (mg kg^{-1})
2017	1.47	16.5	25.6	73.1	0.21	330
2018	1.46	15.2	24.1	71.3	0.19	324

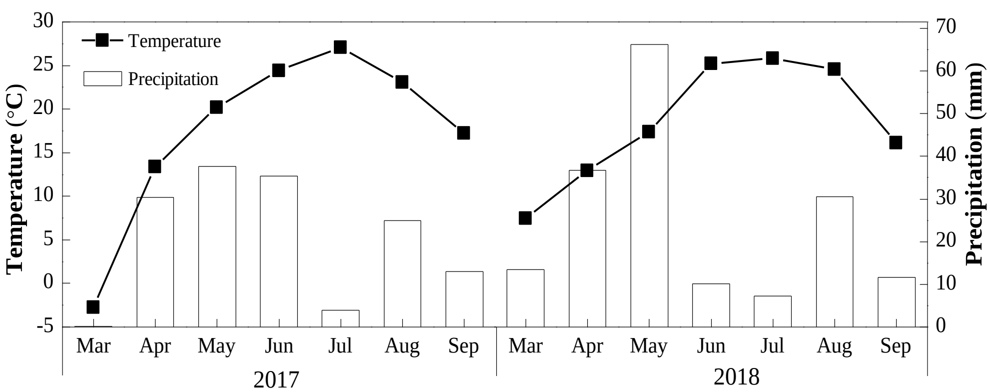

Fig. 1: Precipitation and average temperature at the experimental site during the growing season of alfalfa in 2017 and 2018

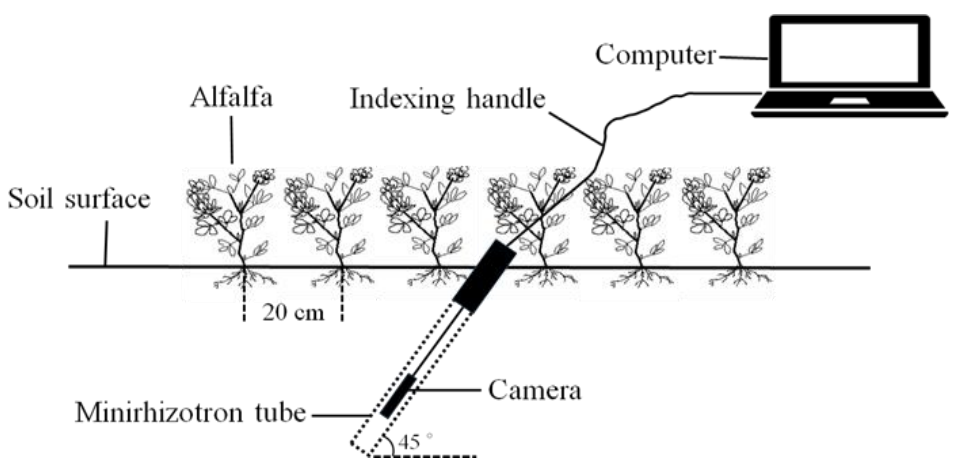

Fig. 2: Minirhizotrons installation

From May to September in 2017 and 2018, CI-600 was used every 30 days to scan and collect root growth images in minirhizotron, totally 10 times. And the stage of crop at the time of measurement was the early flowering stage. The specific image scanning dates were May 22, June 21, July 21, August 22, September 23, in 2017, May 23, June 24, July 24 and August 23, September 24, in 2018, respectively. The collected pictures were brought back to the laboratory for processing with the root image analysis by WinRHIZO TRON MF 2014b. Roots appearing in observation windows were recorded as living roots in white and old roots in brown, while those appearing in observation windows are recorded as dead roots when fine roots completely turn black, cortex shedding or obvious folds and disappearance occur. The vertical depth of the minirhizotron was about 60 cm which divided into 0~20, 20~40 and 40~60 cm from top to bottom. Roots less than 2 mm in the observation window were recorded only.

Measurement index and method

Fine root length density: Taking the root length density as the basic parameter, taking the existing length of the fine root in the whole minirhizotron as a whole, the root length density of the whole root canal is the total standing crop. The root length density of the fine root in different diameter classifications is the standing crop of the fine root in different diameter classifications. And the root length density of the fine root in different soil layers is the standing crop of the fine root in different soil layers. The concrete formulas are as follows:

$$RLD = RL \times \sin\theta / (A \times 4 \times DOF)$$

In the formula, RL (cm) is the root length of the whole minirhizotron, A (cm) is the area of the scanned image (422.3 cm^2), A×4 (cm) is the area of the whole minirhizotron, RLD ($cm.cm^{-3}$) is the root length density production, DOF is the thickness of the soil layer, generally 0.2～0.3 cm. In this study, DOF (cm) is 0.2 cm. Because the minirhizotron is 45° from the ground, it is necessary to multiply the fine root length density by sin 45° to obtain the unit volume root length density at the vertical height. The vertical depth of micro-root canals is about 60 cm, and the standing crop of fine roots in 0~20 cm, 20~40 cm and 40~60 cm soil layers are expressed every 20 cm from top to bottom.

Fine root production and mortality of alfalfa: Fine root production of alfalfa refers to the root length of new roots and the increase of elongation growth of old roots during the last sampling period and the previous sampling period in the same treatment during the growing season. Fine root mortality includes the reduction of original root length caused by the death of original roots and the feeding of root-feeding animals.

Turnover rate: Turnover rate (yr^{-1}) was = Annual fine root production ($cm.cm^{-3}\cdot yr^{-1}$)/maximum annual fine root standing crop ($cm.cm^{-3}$).

Statistical analysis

WPS 2016 was used to collate the data of fine root standing crop, production, mortality. DPS 7.05 (Data Processing System, China) was used to analyze the data. Duncan method was used to analyze the difference significance of the data (P < 0.05). Pearson correlation analysis in SPSS 19.0 (SPSS Inc., Chicago, IL, USA) was used to analyze the relationship between fine root standing crop and each factor. Drawing with Origin 8.0 (OriginLab OriginPro, USA).

Results

Total hay yield of alfalfa

The total hay yield of alfalfa increased first and then decreased with the increase of phosphorus application, and reached the highest at 100 kg P ha^{-1}, which was 20.74 t·ha^{-1} and 19.83 t·ha^{-1} in 2017 and 2018, respectively (Fig. 3). The total hay yield in 2017 was higher than 2018 under P application treatments. The total hay yield of alfalfa at 100 kg P ha^{-1} was significantly higher than 0 kg P ha^{-1} and 50 kg P ha^{-1} treatments (P < 0.05). The total hay yield of alfalfa at 50 kg P ha^{-1} was not significantly different from 150 kg P ha^{-1}.

Total fine root in standing crops

The fine root in standing crop of alfalfa under different phosphorus treatments had two peaks in June and August in both years, respectively (Fig. 4). Fine root standing crop declined from August to September. With the increase of phosphorus application, the fine root standing crop of alfalfa increased first and then decreased, and reached the maximum value at 100 kg P ha^{-1}. The fine root in standing crop of alfalfa at 100 kg P ha^{-1} and 150 kg P ha^{-1} treatments was larger than 50 kg P ha^{-1} treatment. The fine root in standing crop of alfalfa at 100 kg P ha^{-1} treatment was 34.00 to 61.05% higher than 0 kg P ha^{-1} treatment. The fine root in standing crop of alfalfa at 100 kg P ha^{-1} was 17.80 to 41.78% higher than 50 kg P ha^{-1} treatment (P < 0.05).

In all soil layers, there were two peaks of fine root in standing crop of alfalfa during June and August of each year (Table 2). Fine root in standing crop of alfalfa was the lowest in May and mainly concentrated in 0~20 cm under different soil layers. With the increase of soil depth, the fine root standing crop of alfalfa decreased gradually. In 0–20 cm soil layer, the highest fine root standing crop of alfalfa was 100 kg P ha^{-1} than 0 kg P ha^{-1} and 50 kg P ha^{-1} treatments (P < 0.05). Except September 24, 2018, the fine root standing crop of alfalfa at 100 kg P ha^{-1} treatment was significantly higher than other treatments in 20~40 cm soil layer (P < 0.05). In 40~60 cm soil layer, the fine root standing crop of alfalfa applied with 100 kg P ha^{-1} treatment was significantly larger than 0 kg P ha^{-1} treatments (P < 0.05).

Fine root production and mortality

There were two peaks in fine root production of alfalfa during May-June and July–August, of both years respectively (Fig. 5). The fine root production of 100 kg P ha^{-1} treatment was significantly higher than 0 kg P ha^{-1} and 50 kg P ha^{-1} treatment in each period (P < 0.05). Except for

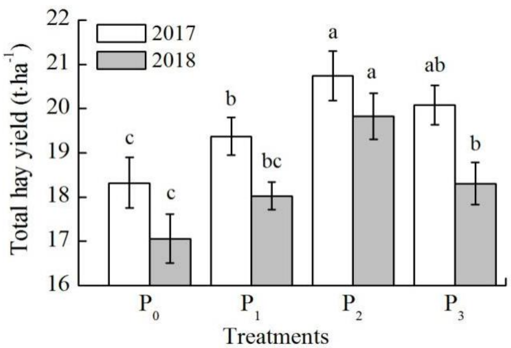

Fig. 3: Total hay yield of alfalfa under drip irrigation in different fertilization treatments in 2017 and 2018

Note: The small letters in the picture indicate that different treatments have significant differences at the 0.05 level

P_0, P_1, P_2, and P_3 mean 0 kg· ha^{-1}, 50 kg· ha^{-1}, 100 kg· ha^{-1} and 150 kg· ha^{-1}, respectively

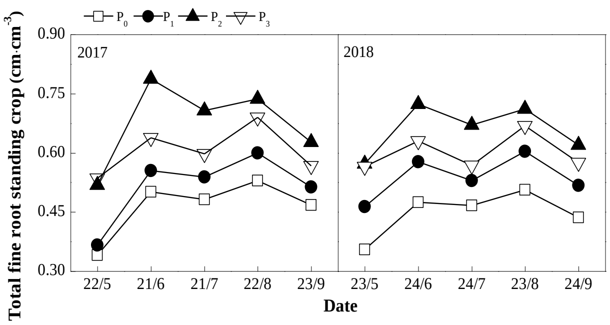

Fig. 4: Total fine root standing crop of alfalfa under different treatments (cm·cm^{-3})

22 August to 23 September in 2017 and 24 June to 24 July in 2018, the fine root production of phosphorus application was significantly higher than 0 kg P ha^{-1} ($P < 0.05$). The fine root production of 100 kg P ha^{-1} treatment was 27.24 to 76.91% higher than 50 kg P ha^{-1} treatment. Fine root mortality reached the highest level in August to September in two years, and the fine root mortality of 100 kg P ha^{-1} reached the maximum, 0.1903 and 0.1640 cm^{-3}, respectively. The mortality of 100 kg P ha^{-1} and 150 kg P ha^{-1} treatments was significantly higher than 0 kg P ha^{-1} and 50 kg P ha^{-1} treatments ($P < 0.05$). Fine root mortality of 100 kg P ha^{-1} treatment reached the lowest level in May to June, 0.0601 and 0.0620 cm^{-3}, respectively.

Fine root turnover rate

The annual production of fine root, annual mortality of fine root, maximum annual standing crop of fine root and turnover rate all reached the maximum under 100 kg P ha^{-1} treatment. The turnover rates were 0.2134 and 0.1814 at 100 kg P ha^{-1} treatment, respectively, which were higher 10.0 and 5.8% than 0 kg P ha^{-1} treatment (Table 3). Turnover rate at 100 kg P ha^{-1} treatment was significantly higher than other treatments ($P < 0.05$), but there was no significant difference between 150 kg P ha^{-1} treatment and 0 kg P ha^{-1} treatment in 2018 ($P > 0.05$). The annual production of fine root, maximum annual standing crop of fine root of 100 kg·P ha^{-1} treatment was significantly larger than 0 kg P ha^{-1} treatment, and the annual fine root production and the maximum annual fine root standing crop at 100 kg P ha^{-1} treatment were significantly larger than other treatments ($P < 0.05$). The annual fine root mortality at 100 kg P ha^{-1} and 150 kg P ha^{-1} treatments was significantly higher than 0 kg P ha^{-1} and 50 kg P ha^{-1} treatments ($P < 0.05$).

Correlation between annual total hay yield and annual standing crop, production, mortality of fine root and turnover rate

Pearson correlation analysis showed (Table 4) that annual total hay yield of alfalfa and annual production of fine root had significant correlation ($P < 0.01$), between annual total hay yield of alfalfa and annual fine root standing crop had significant correlation ($P < 0.05$) and annual production of fine root and the annual fine root standing crop ($P < 0.01$). Fine root mortality and annual fine root standing crop and annual fine root production had significant correlation ($P < 0.05$). There was no significant correlation between turnover rate and other indicators ($P > 0.05$).

Discussion

The growth of forage roots will affect the accumulation of dry matter in the aerial part; fine roots had a great impact on the growth and mortality of alfalfa roots. Fine root production and mortality directly affected the net primary productivity of alfalfa (Norby *et al.* 2004). Studies have shown that when a certain amount of fertilizer is applied to the soil, the availability of nutrients can be increased, the distribution of C to the underground increases, the growth of fine roots and the accumulation of biomass can be promoted, and the ability of fine roots to absorb nutrients and water can be enhanced (Meinen *et al.* 2009). In present study, the total hay yield of alfalfa reached its maximum when the application of phosphorus was 100 kg·ha^{-1}. The main reason was that proper application of phosphorus fertilizer could significantly increase the chlorophyll content of alfalfa leaves, thereby increasing the photosynthesis rate of alfalfa, promote the growth of alfalfa plants, and then increasing the hay yield of alfalfa (Aranjuelo *et al.* 2007). When phosphorus fertilizer was applied, alfalfa had higher fine root production, which indicated that the application of phosphorus fertilizer could increase the absorption surface area, facilitate alfalfa to absorb nutrients in soil, and further promote the growth of above ground in alfalfa plants.

Fertilization under drip irrigation had a significant effect on fine root growth of alfalfa. The fine root standing crop and production of alfalfa under different phosphorus application treatments increased first and then decreased with the increase of fertilizer application. This indicated that

Table 3: Fine root turnover rate of alfalfa in different treatments (yr^{-1})

Years	Phosphorus treatments	Annual production of fine root ($cm \cdot cm^{-3} \cdot yr^{-1}$)	Annual mortality of fine root ($cm \cdot cm^{-3} \cdot yr^{-1}$)	Maximum annual standing crop of fine root ($cm \cdot cm^{-3}$)	Turnover rate (yr^{-1})
2017	0 kg· ha^{-1}	0.1027d	0.0709d	0.5295d	0.1940d
	50 kg· ha^{-1}	0.1245c	0.0879c	0.5996c	0.2076b
	100 kg· ha^{-1}	0.1679a	0.1406a	0.7870a	0.2134a
	150 kg· ha^{-1}	0.1421b	0.1343b	0.6896b	0.2060c
2018	0 kg· ha^{-1}	0.0867d	0.0664c	0.5057d	0.1714c
	50 kg· ha^{-1}	0.1078c	0.0945b	0.6040c	0.1785ab
	100 kg· ha^{-1}	0.1312a	0.1191a	0.7235a	0.1814a
	150 kg· ha^{-1}	0.1173b	0.1148a	0.6687b	0.1755bc

Table 4: The correlation analysis between annual standing crop of fine root and annual production of fine root, annual mortality of fine root, turnover rate

Index	Annual total hay yield	Annual standing crop of fine root	Annual production of fine root	Annual mortality of fine root
Annual standing crop of fine root	0.9860*			
Annual production of fine root	0.9980**	0.9940**		
Annual mortality of fine root	0.9350	0.9780*	0.9560*	
Turnover rate	0.9240	0.8660	0.9120	0.8000

Note: * Significant correlation was found at the 0.05 level (bilateral), ** significant correlation was found at the 0.01 level (bilateral)

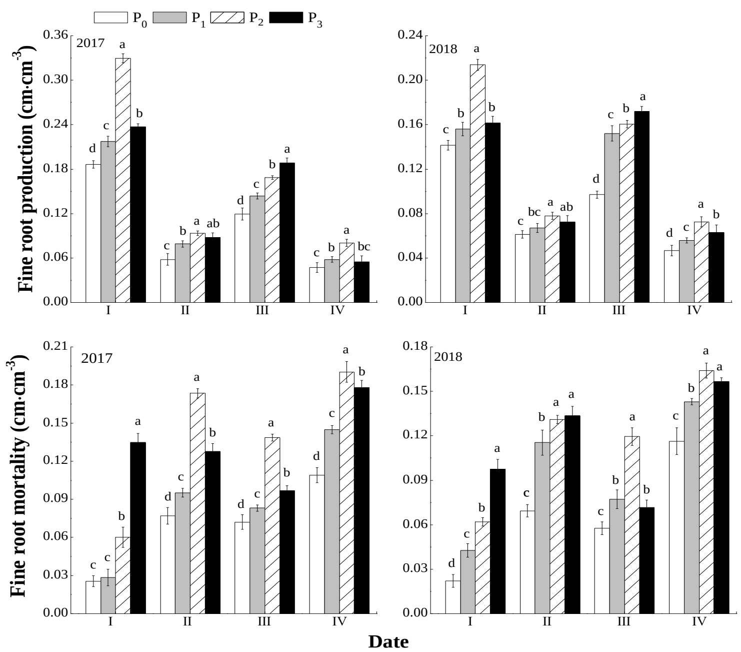

Fig. 5: Fine root production and mortality of alfalfa in different treatments ($cm \cdot cm^{-3}$)

Note: Ⅰ: 22/5 - 21/6, 2017; 23/5 - 24/6, 2018 Ⅱ: 21/6 - 21/7, 2017; 24/6 - 24/7, 2018

III: 21/7 - 22/8, 2017; 24/7 - 23/8, 2018 Ⅳ: 22/8 -23/9, 2017; 23/8 -24/9, 2018

The lowercase letters in the picture indicate that different treatments have significant differences at the 0.05 level

P_0, P_1, P_2, and P_3 mean 0 kg· ha^{-1}, 50 kg· ha^{-1}, 100 kg· ha^{-1} and 150 kg· ha^{-1}, respectively

root growth had a strong fertilizer-tropism. To meet the demand of alfalfa for phosphorus, alfalfa enlarges its root development by expanding the root volume and increasing the root length to promote the absorption of limited nutrients in the soil (Akhtar and Siddiqui 2009). It indicated that proper application of phosphorus fertilizer could effectively promote the growth and accumulation of fine roots, but excessive application of phosphorus fertilizer would restrict root growth, reduce root length and surface area, thus slowing down the growth process of alfalfa.

It was found that the seasonal variation of fine root nutrients of alfalfa was different (Table 1), which reflected the response of plant fine root growth to nutrients. From May to June, temperature gradually increased, nutrient availability and mobility in soil were higher, fine root growth accelerated, plants would allocate more C to fine root, root nutrient uptake increased (Son and Hwang 2003). In mid-summer, the precipitation in Xinjiang was less, and the soil water content is often reduced due to evaporation and strong transpiration of plants will result in the decrease of fine root biomass of alfalfa and accelerates fine root death under drought conditions (Yuan and Chen 2010). Under drought conditions, the amount of water absorbed by fine roots decreased, but the respiration of roots to maintain cell membranes and enzyme activities still consumed a large amount of carbon. Therefore, the amount of fine root death increased in order to reduce energy consumption (Bai and Li 2003).

The death of fine roots is related to the distribution of photosynthates to roots, which is a complex physiological and ecological process. Studies have shown that the more water and nutrients the fine roots absorb, the more carbon they distribute to the fine roots, and the longer their life span (Luo and Zhao, 2019). Once the absorption capacity of fine roots decreases, the distribution of carbon to fine roots decreases immediately, and fine roots grow old and die (Bai and Li 2003). In this study, fine root death of alfalfa changed little from the end of June to August. From August to September, fine root death was the highest in growing season, because with the advent of autumn, the above-ground parts of alfalfa began to wither, photosynthesis of leaves weakened, temperature began to decrease (Brassard *et al.* 2009), fine root began to stop growing gradually, carbohydrates allocated to roots decreased (López *et al.* 2001), and soil temperature continued to decrease. Fine roots died a lot during this period (Jha and Mohapatra 2010). Moreover, alfalfa had the last cuts after September, further aggravating the death of fine roots. The fine root mortality of alfalfa exceeded the growth rate after September. This may be due to the quickening of fine root death at the end of the growing season with the reduced supply of irrigation and growth after the above ground part harvested. With the rapid end of the growing season and the arrival of winter, underground heat conditions will become more uncomfortable.

The availability of soil nutrients after fertilization was affected by many factors, such as rhizosphere, root exudates and microorganisms. The response of fine roots was different in different soil layers. After fertilization, surface nutrient branches infiltrate downward after irrigation, which will increase the nutrient content in deep soil and increase the existing fine roots (Zhou and Wang 2015). Moreover, in present study, the fertilizer was applied with water drip. Drip irrigation can change the conditions of water and temperature in surface soil, which was beneficial to root growth. At the same time, suitable soil temperature and water conditions can also contribute to the decomposition of litter on the ground, thus improving the soil conditions in surface soil, enriching the nutrients in surface soil and promoting the growth and accumulation of fine roots (Amin *et al.* 2014). Therefore, the distribution of fine roots in surface soil is the largest. Suitable application of phosphorus could promote root growth, strengthen rhizosphere process and increase the proportion of fine roots, and promote alfalfa growth and nutrient uptake in early stage (Wang *et al.* 2013). Under suitable environmental conditions, the activities of soil animals and microorganisms increased, which was more conducive to the decomposition of fine roots. Phosphorus application directly affected the metabolic activities of plants, soil animals and microorganisms, the quantity of soil enzyme secretion, the activity of litter decomposition enzymes (Qualls and Richardson 2000), and the increase of enzyme activity can also contribute to the decomposition, transformation of organic matter and the release of nutrient elements in litter. In this study, a small amount of phosphorus application significantly reduced the fine root standing crop and production of alfalfa (Fig. 4), resulting in the whole absorption capacity of root system to decline, and ultimately inhibit the growth of root system.

The root distribution of alfalfa has obvious vertical characteristics. The total fine root standing crop of 0~20 cm and 20~40 cm soil are significantly higher than 40~60 cm soil, mainly due to the differences in spatial distribution of soil resources availability and environmental conditions (Kalliokoski *et al.* 2010). The vertical distribution of fine roots was mainly attributed to the higher temperature in the surface soil and the higher content of available nutrients in the soil is conducive to the growth and absorption of fine roots, and plays a greater role in nutrient acquisition and carbon cycle (Ibrahima *et al.* 2010). Secondly, the bulk density and texture of the surface soil were good, while the lower soil temperature and poor soil are not conducive to the growth of fine roots (Makita *et al.* 2011). Lower nutrient content in deep soil also affects root distribution in deep soil. With the increase of soil depth, soil compactness gradually increases, and permeability was poor, which was harmful to fine root growth (Zhou and Shangguan 2007). Phosphorus application treatments was significantly greater than no phosphorous application, probably because no application of fertilizer would inhibit root growth, reduce root density and lower biomass.

Fine root turnover was the main way to return carbon and nutrients to soil and was determined by fine root life, fine root senescence affects fine root life (Xiong *et al.* 2017). The faster fine root senescence, the shorter fine root life, the faster fine root turnover, the greater carbon consumption, the more nutrients returned to the soil, and the faster nutrient cycle. The highest fine root turnover was achieved at 100 $kg{\cdot}ha^{-1}$ phosphorus application. This relatively high fine root turnover rate indicated that plants had vigorous life activities, and could continuously produce new fine roots to replace old fine roots to absorb water and nutrients, thus increasing the use efficiency of water and nutrients in roots (Cormier *et al.* 2015). This experiment showed that the fine root turnover rate in 2018 was lower than 2017 (Table 3). It may be that the growth of alfalfa for many years affected the secretion behavior of alfalfa roots, and the HPO_4^{2-} secretion significantly reduced. Because of the relative stability of phosphorus in soil, the decrease of HPO_4^{2-} secretion in root system changed the micro-domain cycle balance of phosphorus at the root-soil interface, and in fact reduced the chance of phosphorus reuse by root system. Studies on crop roots show that changes in phosphorus availability can change the morphology and configuration of plant roots, increase the length, density of root hairs and the length and quantity of lateral roots, increase the distribution of available nutrients in higher areas of soil, and thus improve the effective absorption of phosphorus (Jing *et al.* 2010).

Conclusion

There were two peaks of fine root standing crop and fine root production of alfalfa in June and August under different phosphorus application treatments, respectively. Fine root mortality reached the highest from August to September. With the increase of phosphorus application, the fine root standing crop of alfalfa increased first and then decreased, and reached the maximum value at 100 kg P ha^{-1} treatment. Alfalfa fine root standing crop mainly concentrated in 0~20 cm, with the increase of soil depth, alfalfa fine root standing crop decreased gradually. The turnover rates of the two-year at 100 kg P ha^{-1} treatment higher than 0 kg P ha^{-1} treatment were 10 and 5.8%. The annual fine root production and maximum standing crop at 100 kg P ha^{-1} treatment was significantly larger than other P application treatments. When the amount of P_2O_5 was 100 kg·ha^{-1}, the turnover rate of fine roots of alfalfa was improved, and fine root standing crop and fine root production of alfalfa significantly affected the dry matter yield above ground.

Acknowledgements

The research was supported by the National Natural Science Foundation of China (31660693), the Fok Ying Tung Education Foundation (171099), the the China Postdoctoral Science Foundation (2018T111120, 2017M613252), the Youth Innovation Talent Cultivation Program of Shihezi University (CXRC201605) and the China Agriculture Research System (CARS-34).

References

Akhtar M, Z Siddiqui (2009). Effects of phosphate solubilizing microorganism and *Rhizobium* spp. On the growth, nodulation, yield and root-rot disease complex of chickpea under field condition. *Afr J Biotechnol* 8:3479–3488

Amin BAZ, B Chabbert, D Moorhead, I Bertrand (2014). Impact of fine litter chemistry on lignocellulolytic enzyme efficiency during decomposition of maize leaf and root in soil. *Biogeochemistry* 117:169–183

Aranjuelo I, JJ Irigoyen, M Sánchez-Díaz (2007). Effect of elevated temperature and water availability on CO_2 exchange and nitrogen fixation of nodulated alfalfa plants. *Environ Exp Bot* 59:99–108

Bai WM, LH Li (2003). Effect of irrigation methods and quota on root water uptake and biomass of alfalfa in the Wulanbuhe sandy region of China. *Agric Water Manage* 62:139–148

Bennett JN, B Andrew, CE Prescott (2002). Vertical fine root distributions of western redcedar, western hemlock, and salal in old-growth cedar-hemlock forests on northern Vancouver Island. *Can J For Res* 32:1208–1216

Brassard BW, HYH Chen, Y Bergeron (2009). Influence of environmental variability on root dynamics in northern forests. *Crit Rev Plant Sci* 28:179–197

Chang RY, BJ Fu, GH Liu, XL Yao, S Wang (2012). Effects of soil physicochemical properties and stand age on fine root biomass and vertical distribution of plantation forests in the loess plateau of China. *Ecol Res* 27:827–836

Cormier N, RR Twille, KC Ewel, KW Krauss (2015). Fine root productivity varies along nitrogen and phosphorus gradients in high-rainfall mangrove forests of Micronesia. *Hydrobiologia* 750:69–87

Finér L, M Ohashi, K Noguchi, Y Hirano (2011). Fine root production and turnover in forest ecosystems in relation to stand and environmental characteristi-cs. *For Ecol Manage* 262:2008–2023

Ibrahima A, ZEA Mvondo, JC Ntonga (2010). Fine root production and distribution in the tropical rainforests of south-western cameroon: Effectsof soil type and selective logging. *For Biogeosci For* 3:130–136

Jha P, KP Mohapatra (2010). Leaf litter fall, fine root production and turnover in four major tree species of the semi-arid region of India. *Plant Soil* 326:481–491

Jiang XY, LX Cao, RD Zhang, LJ Yan, Y Mao, YW Yang (2014). Effects of nitrogen addition and litter properties on litter decomposition and enzyme activities of individual fungi. *Appl Soil Ecol* 80:108–115

Jing JY, YK Rui, FS Zhang, Z Rengel, JB Shen (2010). Localized application of phosphorus and ammonium improves growth of maize seedlings by stimulating root proliferation and rhizosphere acidification. *Field Crops Res* 119:355–364

Johnson MG, DT Tingey, DL Phillips, MJ Storm (2001). Advancing fine root research with minirhizotrons. *Environ Exp Bot* 45:263–289

Kalliokoski T, T Pennanen, P Nygren, R SieväNen, HS Helmisaari (2010). Belowground interspecific competition in mixed boreal forests: Fine root and ectomycorrhiza characteristics along stand developmental stage and soil fertility gradients. *Plant Soil* 330:73–89

Lee KH, S Jose (2003). Soil respiration, fine root production, and microbial biomass in cottonwood and loblolly pine plantations along a nitrogen fertilization gradient. *For Ecol Manage* 185:263–273

Liu C, WH Xiang, PF Lei, XW Deng, DL Tian, X Fang, CH Peng (2014). Standing fine root mass and production in four Chinese subtropical forests along a succession and species diversity gradient. *Plant Soil* 376:445–459

López B, S Sabaté, CA Gracia (2001). Annual and seasonal changes in fine root biomass of a *Quercus ilex* L. forest. *Plant Soil* 230:125–134

Luo WC, WZ Zhao (2019). Adventitious roots are key to the development of nebkhasin extremely arid regions. *Plant Soil* 442:471–482

Majdi H (2001). Changes in fine root production and longevity in relation to water and nutrient availability in a Norway spruce stand in northern Sweden. *Tree Physiol* 21:1057–1061

Makita N, Y Hirano, T Mizoguchi, Y Kominami, M Dannoura, H Ishii, L Finér, Y Kanazawa (2011). Very fine roots respond to soil depth: Biomass allocation, morphology, and physiology in a broad-leaved temperate forest. *Ecol Res* 26:95–104

Meinen C, D Hertel, C Leuschner (2009). Biomass and morphology of fine roots in temperate broad-leaved forests differing in tree species diversity: is there evidence of below-ground overyielding? *Oecologia* 161:99–111

Norby RJ, J Ledford, CD Reilly, NE Miller, EG O´Neill (2004). Fine-root production dominates response of a deciduous forest to atmospheric CO_2 enrichment. *Proc Nat Acad Sci* 101:9689–9693

Okada KI, SI Aiba, K Kitayama (2017). Influence of temperature and soil nitrogen and phosphorus availabilities on fine-root productivity in tropical rainforests on mount Kinabalu, Borneo. *Ecol Res* 32:145–156

Pregitzer KS, JL Deforest, AJ Burton, MF Allen, RRL Hendrick (2002). Fine root architecture of nine North American trees. *Ecol Monogr* 72:293–309

Qualls RG, CJ Richardson (2000). Phosphorus enrichment affects litter decomposition, immobilization, and soil microbial phosphorus in wetland mesocosms. *Soil Sci Soc Amer J* 64:799–808

Ren AT, WH Lu, JJ Yang, CH Ma (2015). Seasonal change patterns in the production and mortality of fine roots in cotton and alfalfa. *Acta Pratacult Sin* 24:213–219

Singh SK, VR Reddy (2014). Combined effects of phosphorus nutrition and elevated carbon dioxide concentration on chlorophyll fluorescence, photosynthesis, and nutrient efficiency of cotton. *J Plant Nutr Soil Sci* 177:892–902

Son Y, JH Hwang (2003). Fine root biomass, production and turnover in a fertilized *Larix leptolepis*, plantation in central korea. *Ecol Res* 18:339–346

Vitousek PM, S Porder, BZ Houlton, OA Chadwick (2010). Terrestrial phosphorus limitation: mechanisms, implications, and nitrogen-phosphorus interactions. *Ecol Appl* 20:5–15

Wang X, HL Tang, JB Shen (2013). Root responses of maize to spatial heterogenous nitrogen and phosphorus. *J Plant Nutr Fert* 19:1058–1064

Williamson LC, SPCP Ribrioux, AH Fitter, HMO Leyser (2001). Phosphate availability regulates root system architecture in Arabidopsis. *Plant Physiol* 126:875–882

Xiong YM, X Liu, W Guan, BW Liao, YJ Chen, M Li, CR Zhong (2017). Fine root functional group based estimates of fine root production and turnover rate in natural mangrove forests. *Plant Soil* 413:83–95

Yacoubi R, C Job, M Belghazi, W Chaibi, D Job (2011). Toward characterizing seed vigor in alfalfa through proteomic analysis of germination and priming. *J Proteom Res* 10:3891–3903

Yuan ZY, HYH Chen (2010). Fine root biomass, production, turnover rates, and nutrient contents in boreal forest ecosystems in relation to species, climate, fertility, and stand age: literature review and meta-analyses. *Crit Rev Plant Sci* 29:204–221

Zhou ZC, ZP Shangguan (2007). Vertical distribution of fine roots in relation to soil factors in *Ppinus tabulaeformis* Carr. forest of the loess plateau of China. *Plant Soil* 291: 119–129

Zhou ZH, CK Wang (2015). Reviews and syntheses: soil resources and climate jointly drive variations in microbial biomass carbon and nitrogen in China's forest ecosystems. *Biogeosciences* 12: 6751–6760

Zhu FF, M Yoh, FS Gilliam, XK Lu, JM Mo (2013). Nutrient limitation in three lowland tropical forests in southern china receiving high nitrogen deposition: insights from fine root responses to nutrient additions. *PLoS One* 8; Article e82661

Zhang H, H Liu, S Wang, X Guo, X Gong, J Sun (2019). Modelling the soil water dynamics under micro-sprinkling hose irrigation for distorted roots of transplanted cotton. *Intl J Agric Biol* 21:191–200