



Effects of long-term nitrogen addition and precipitation reduction on the fine root dynamics and morphology in a temperate forest

Ning Dong¹ · Jun Zhou² · Guoyong Yan¹ · Guancheng Liu¹ · Yajuan Xing^{1,2} · Qinggui Wang^{1,2}

Received: 17 August 2021 / Revised: 4 January 2022 / Accepted: 15 March 2022 / Published online: 29 March 2022
© The Author(s), under exclusive licence to Springer-Verlag GmbH Germany, part of Springer Nature 2022

Abstract

Fine roots (< 2 mm in diameter) are the main organ for obtaining resources from the belowground part of forest and also act as a hub linking the ecological processes of plant and soil. However, in the context of global climate change, it remains unclear how fine root dynamics and morphology in temperate forests respond to increased nitrogen deposition and reduced precipitation in growing season on a year-round time scale. In this study, the minirhizotrons were used to observe the response of fine root dynamics (production, mortality, turnover and life span) and morphology (diameter, single root surface area, single root length) to long-term nitrogen addition (N, 50 kg N ha⁻¹ yr⁻¹), precipitation reduction (W, -200 mm yr⁻¹) and their interactive treatments (NW) in a broad-leaved Korean pine forest in Changbai Mountains over a two-year period. The results showed that N significantly increased the average diameter of fine roots. Compared to the control treatment, all treatments significantly reduced the average monthly number and surface area of live root. However, the morphological traits of the individual root in each treatment differed between the growing and non-growing seasons. All three treatments (N, W, NW) reduced annual production of fine root over the two observation periods, while the effect on annual mortality varied between years. N, W and the interaction of NW did not significantly change the annual turnover of fine root in the first observation period but increased significantly in the second. The median life span of fine root born in both non-growing seasons was significantly lower than that of fine root born in the growing season. Our results show that changes in the growth strategy of fine root depend on the complex interrelationship between their own morphology, soil layer and seasonal climatic conditions.

Keywords Growing season and non-growing season · Fine root production and turnover · Minirhizotron technique · Temperate forest

Abbreviations

N	Nitrogen addition	FRP	Fine root average production
W	Precipitation reduction	FRM	Fine root average mortality
NW	The interaction of nitrogen addition and precipitation reduction	FRT	Turnover rate of fine root
TC	Total carbon	FRP ^M	Fine root average monthly production
TN	Total nitrogen	FRM ^M	Fine root average monthly mortality

Communicated by Judy Simon.

✉ Yajuan Xing
xingyajuan@163.com

✉ Qinggui Wang
qgwang1970@163.com

¹ School of Life Sciences, Qufu Normal University, 57 Jingxuan West Road, Qufu 273165, China

² Department of Agricultural Resource and Environment, Heilongjiang University, 74 Xuefu Road, Harbin 150080, China

Introduction

The root system of trees plays an important role in anchoring and transporting water and nutrients. Coarse roots form the main framework of the root system, while fine roots play a more active role in the cycling of water, nutrients and carbon (Rossi et al. 2016; McCormack et al. 2017). Fine roots play an indispensable role in biogeochemical cycling and supporting plant functions (Hendricks et al. 1993). Although their biomass accounts for a small proportion of the total forest biomass, the production and turnover of fine root account for about 33% of the global terrestrial net primary

productivity (Jackson et al. 1997). Fine root turnover is a regulatory factor of soil carbon and nitrogen cycle, and its contribution to soil organic matter pool is equal to or greater than that of aboveground litter (Nadelhoffer et al. 1992; West et al. 2004).

Fine roots are sensitive to the change of external environment, and their functional characters are highly plastic to the changes of soil nutrients and water (Finér et al. 2011; Eisenstat et al. 2013). With the development of industry, some human activities lead to the acceleration of atmospheric nitrogen deposition rate, which may lead to the increase of soil nitrogen efficiency (Galloway et al. 1995; Galloway et al. 2004; Lu et al. 2011; Deng et al. 2018). At the same time, the variation of precipitation pattern will also change the soil water availability. These two environmental factors are the key affecting element of fine root dynamic (Yuan and Chen 2012). In the context of global climate change, they may change at the same time and interact each other (Li et al. 2021). Therefore, studying how fine root dynamics respond to increased nitrogen deposition and altered precipitation is important for understanding and predicting carbon and nutrient cycling in forest ecosystems.

As an important mainstay of terrestrial ecosystems, forest ecosystems play an important role in mitigating global warming and maintaining the global carbon cycle (Bonan et al. 2008). However, our research on how the fine roots of forest tree respond to climate change is still incomplete, and the findings of simulated nitrogen deposition and precipitation reduction experiments carried out in different regions and tree species are inconsistent (Comas et al. 2013; Li et al. 2015). Some studies have found that nitrogen addition and precipitation reduction increased fine root production and turnover (Santantonio. 1982; Yuan and Chen. 2010; Wang et al. 2012), while other studies and meta-analyses have come to the opposite conclusion (Li et al. 2015; Li et al. 2017). The situation that fine root dynamics were not significantly changed by increasing nitrogen and reducing water was also happened (Ostertag 2001; Finér et al. 2011). In addition, the heterogeneity of soil resource in different soil layers results in different dynamics and distribution patterns of fine root vertically and horizontally, which can also lead to different effects of nitrogen addition and precipitation reduction on fine root dynamics in the surface and deeper layers (Yan et al. 2017; Lozanova et al. 2019). The reason for these contradictory conclusions may be that, on the one hand, climate types, processing time and different measurement methods may draw different conclusions. For example, the sequential coring method in subtropical forests found that medium nitrogen addition significantly increased the fine root turnover rate of surface fine root (Wang et al. 2019). On the contrary, the minirhizotrons showed the opposite result. In temperate forests, it was found that the fine root production and turnover rate estimated by the ingrowth

core method were much lower than those by the sequential soil cores method. On the other hand, the effect of nitrogen addition and drought on fine root dynamics depends to some extent on the state of the soil resource prior to nitrogen application and its exposure to drought stress (Idol et al. 2000; Kou et al. 2018). As the hidden other half of terrestrial ecology, these difficult to quantify influences increase the difficulty of observing the dynamics of fine root growth and to some extent hinder the development of root ecology.

The interaction of nitrogen addition and precipitation reduction may have a more complex effect on fine root than these two environmental factors acting alone (Dewar. 1993; Wang et al. 2017). In northern temperate forests, fine roots stressed by soils with elevated nitrogen content may be more susceptible to drought. That is because the nitrogen addition leads to a decrease in the longevity and standing crops of fine roots in the forests of the region, which makes the fine root water uptake less efficient (Smithwick et al. 2013). In addition, the interaction of N addition and water stress may also affect the changes in root distribution and dynamics in different soil layers (Joslin et al. 2000; Lima et al. 2010), but the strength or direction of these interactions remains to be studied (McMurtrie et al. 2012; Abramoff and Finzi 2015). In previous studies, the treatment time of simulated nitrogen increase and water decrease was relatively short (Pregitzer et al. 1993; Wang et al. 2012), and few studies focused on the effect of long-term nitrogen and water interaction on fine root dynamics. In addition, most of the research on fine root dynamics are currently focused on the growing season, while less research have been done on fine root growth processes during the non-growing season when the soil is frozen (Fitzhugh et al. 2001). But the response of fine root dynamics and morphology to changes in environmental factors is likely to differ between the non-growing and growing seasons (Yin et al. 2017; Wang et al. 2019). Changes in fine root dynamics and morphology during the non-growing season may also have an impact on the growth process of fine root during the growing season (Pakkala et al. 2015; Song et al. 2017), especially in temperate forests where the temperature is lower and the soil freezing degree is deeper in the non-growing season.

In the past 30 years, the total nitrogen deposition rate increased significantly and the precipitation decreased in Northeast China (Liu et al. 2013). In order to study the effects of long-term nitrogen addition and precipitation reduction and their interaction on fine root dynamics in Changbai Mountains northeast China, a long-term experiment of increasing nitrogen and reducing precipitation was conducted in a broad-leaved Korean pine forest. The minirhizotron technology was selected to observe the dynamic of fine root in situ for a long time, which has incomparable advantages in the study of seasonal dynamics of fine root (Johnson et al. 2001). We hypothesized that (1) long-term

nitrogen addition and precipitation reduction will reduce the production of fine root, but the turnover rate will be accelerated, and the life span of fine root will be reduced. (2) The effect of long-term interaction of nitrogen addition and precipitation reduction on fine root dynamics is different from that of nitrogen addition and precipitation reduction alone. (3) The response of fine root dynamics and morphology to different treatments differs between the growing and non-growing seasons.

Materials and methods

Field site and experimental design

This study was conducted in Changbai Mountains Natural Reserve, Jilin province, Northeast China (42°24' N, 128°06' E). The average altitude of this area is 738 m a.s.l., the annual average temperature is 3.69 °C, the average temperature in growing season is 15 °C, and in non-growing season is -0.6 °C. The highest monthly average temperature is 21.5 °C, which often occurred in August. The lowest monthly average temperature is -17.3 °C, which often occurred in January. The area has a temperate monsoon climate, with an average annual precipitation of about 790 mm. Most of the precipitation is mainly concentrated from May to October, and the non-growing season precipitation is mainly in the form of snowfall. The soil of the study site is classified as Eutric Cambisol (FAO classification) with 31.54% sand, 42.18% silt, 26.28% clay, and 25.42% organic matter in the topsoil in the 0–20-cm soil layer. The study site is an original broadleaved red pine forest > 300 years old, and the dominant trees in the study site are *Fraxinus mandshurica*, *Pinus koraiensis* and *Tilia amurensis*. The tree density of the study site is approximately 435 trees per hectare. The main dominant shrubs species in the study site are *Corylus mandshurica*, *Lonicera japonica* and *Philadelphus schrenkii*. The dominant herbaceous species include *Anemone cathayensis*, *Adonis amurensis* and *Filipendula palmate* (Yan et al. 2020).

In 2009, a long-term field experiment of throughfall transfer and artificial nitrogen addition was established in a broad-leaved Korean pine forest in Changbai Mountains. Six 50 m × 50 m standard plots were randomly established in the area with similar site conditions. There was a buffer zone of more than 20 m between each quadrat to avoid mutual interference. Three quadrats were treated with precipitation reduction, and the other three quadrats were not. The precipitation transfer facility was composed of polycarbonate V-shaped transparent plate with high transparency (95%). In order to ensure the normal flow of air, the light transmitting plate was fixed on an aluminum frame about 1 m away from the soil surface. In the growing season, the polycarbonate V-shaped transparent plate was used to intercept

about 30% of the natural rainfall which was equivalent to a drought year's precipitation rate (Ying et al. 2006). In the non-growing season, the polycarbonate board was removed to allow snowfall to fall on the forest surface naturally. Previous studies have shown that the annual average level of atmospheric nitrogen deposition in Changbai Mountains area is 23 kg N ha⁻¹ yr⁻¹, and it is expected that the nitrogen deposition rate in this area will double that in 2050 (Lü and Tian 2007). Therefore, the amount of nitrogen applied in this study was 50 kg N ha⁻¹ yr⁻¹ was approximately twice the average annual atmospheric nitrogen deposition in the Changbai Mountain area, and ammonium nitrate was selected as the additional nitrogen source to simulate nitrogen deposition. To investigate the effect of nitrogen deposition increase on the fine roots, each of the six 50 m × 50 m standard plots was divided into two 25 m × 50 m subplots using a PVC sheet. The PVC sheet was inserted 50 cm deep into the ground in order to avoid nutrient mobilization between adjacent subplots. One subplot of each plot was treated with nitrogen addition while the other was not. Starting in May 2009, weighed ammonium nitrate (NH₄NO₃) was mixed with 40 L of deionized water and sprayed using a backpack sprayer to uniformly apply it to each nitrogen-enhanced treatment subplot at the beginning of each month during the growing season (May–October). The control and precipitation reduction areas were simultaneously sprayed with an equal volume of deionized water without nitrogen addition (40L) to avoid differences in water application. To sum up, four treatments were set up in this study, which were the control, nitrogen addition (N, + 50 kg N ha⁻¹ yr⁻¹), precipitation reduction (W, -30%, about 200 mm yr⁻¹) and the interaction of nitrogen addition and precipitation reduction (NW) and each treatment was repeated three times (Fig. S1).

Minirhizotron installation and image collection

In July 2015, five points were randomly selected for each sample plot to install minirhizotrons (external diameter 7 cm, inner diameter 6.4 cm, length of 100 cm). A total of 60 minirhizotrons were installed, and minirhizotrons had an angle of 30 to the ground, the belowground length of minirhizotrons was 84 cm and the vertical observation depth of root system was 42 cm. In order to minimize the heat exchange, black tape and yellow tape were used successively to wrap the bare part of each minirhizotron. The orifice of the minirhizotron was covered with a black rubber cover to prevent rainwater and other litter from entering the canal. After installation, the minirhizotron was stabilized more than 12-month delay period to stabilize the density of the surrounding fine root to stabilize the observation environment (Fig. S1).

From October 2017 to October 2019, the minirhizotron images were collected once a month. The first observation

period was from November 2017 to October 2018, and the second was from November 2018 to October 2019. The image of root growth was collected at the same position, and the size of the image was 19.60×21.56 cm. WinRHIZO Tron MF 2012 (Regent instrument Inc., Quebec, Canada) software was used to analyze and process the collected images to obtain the original data of fine root indicators for calculation. Because there were many herbaceous roots in the 0–10-cm soil layer of the sample plot, we could not accurately identify the roots of tree and herbaceous plant. In order to ensure the accuracy of the observation results, we did not include them in range of image analysis. Therefore, the range of soil layers we analyzed was 10–40 cm (one soil layer per 10 cm).

Root image analysis

During the interval between the two images, the white non-lignified fine roots were marked as “new” in their properties. If the “new fine roots” in the previous observation period remained white or brown on the subsequent images, they were still living roots, and this type of root was marked as “alive.” When the color of root changed or the epidermis folded or turned from brown to black and new roots were no longer produced in the subsequent image analysis, we defined it as dead root and label it as “dead.” The root that disappeared in the interval between two image acquisitions was also divided into dead root and marked as “gone.”

In this study, the production of fine root was calculated by the change of living fine root length per unit observation window area, and the death of fine root was calculated by the change of dead fine root length per unit measurement window area. The fine root production of each sampling interval was determined by adding the length of all new fine roots and the elongation growth of all living roots. The fine root mortality of each sampling interval was determined by adding the length of all dead roots and the length of disappeared fine roots (Xiong et al. 2018; Wang et al. 2019). It was assumed that the daily production and mortality of the fine root were constant within the sampling interval between the consecutive image acquisition dates and then estimated the monthly production and mortality in the two observation dates (Yan et al. 2017).

The estimation method of fine root turnover rate was as follows: Turnover 1 = the ratio of annual growth yield of fine root to observed average live root length; Turnover 2 = the ratio of annual fine root length death to observed average live root length; Turnover 3 = the average of the above two indicators (Wang et al. 2019).

The life span of fine root was estimated from the first appearance to the first disappearance of fine root. The data recorded by each root include date of birth, date of death and soil depth. Roots born between November of

the previous year and April of the next year were defined as non-growing season birth, and roots born between May and October of the next year were defined as growing season birth.

Soil parameters

Soil samples were collected in September 2018 and September 2019, respectively. Three soil samples were collected randomly from each sub-sample plot separately using a soil auger after removal of plant debris and mixed as replicates. All samples were air-dried and sieved through a 2-mm soil sieve, and roots, stones and other debris were removed. Soil moisture content was analyzed by weight, i.e., by weighing 10 g of fresh soil in an oven at 105 °C for 24 h until a constant weight was reached. The soil total carbon (TC) and nitrogen (TN) were measured using an automated TC/TN analyzer (multi N/C 3100, Analytik Jena AG, Germany). The soil pH value was measured in 1:2.5 (soil/water) suspension using a pH meter (SX7150, China).

Statistical analysis

Kolmogorov Smirnov was used to test whether the data were normally distributed, and Levene's was used to test whether the data satisfy the homogeneity of variance. One-way ANOVA was used to analyze the effects of different treatments on fine root production, death, turnover, average diameter, monthly average living root surface area, monthly average living root number, single root surface area and single root length in different soil layers, and LSD method was used for multiple comparisons. The survival curve and median root life of each treatment were estimated by K-M analysis, and the median life was estimated by 50% survival rate. This method is a nonparametric estimation method, also known as cumulative survival rate, and does not need to make any assumptions about the distribution of the estimated data (Guo et al. 2008; Repo et al. 2014). As some of the fine roots are still alive at the end of the observation, previous studies have shown that the life span of fine roots is non-normally distributed (Hendrick and Pregitzer 1993; Lópe et al. 2001). The use of median lifetimes in survival analyses is therefore more representative. Log-rank test was used to test the difference of survival curve of different treatments, birth dates and soil layers (Guo et al. 2008). Cox proportional hazard regression analysis was used to simultaneously determine the effects of fine root diameter, length and surface area, birth season and soil layer on fine root longevity of each treatment. All data were analyzed by SPSS 19.0 (SPSS, IBM, USA).

Results

Climatic conditions and soil physicochemical properties

The meteorological data for this study were obtained from the greenhouse data sharing platform (<http://data.sheshiyuan.yi.com/>). The average temperature of the first observation period was 3.75 °C and that of the second was 4.14 °C (Fig. S2), which were both higher than the long-term annual average temperature (3.27 °C). The total precipitation of the first observation period was 730.8 mm, and that of the second was 504 mm. Compared with the long-term average annual precipitation (714.6 mm), the first observation period was more abundant, while the second was less. The maximum snow cover thickness (41 cm) and snow cover days (137 d) in the first non-growing season were higher than those in the second non-growing season (7 cm) and (78 d) (Yan 2020). Both the W and NW significantly reduced soil water content ($p < 0.01$) (Table S2), with all treatments having a lower soil water content in the second observation period than that in the first. The N significantly reduced the soil pH and increased the TN content significantly ($p < 0.05$).

Fine root morphological traits

The N significantly increased the average diameter of fine root ($P < 0.01$), the W significantly increased the average diameter of fine root in 10–20-cm soil layer in the second observation period ($P < 0.01$), while the interaction of NW

significantly decreased the average diameter of fine root in 30–40-cm soil layer (Fig. 1). In addition, the N, W and the interaction of NW all significantly reduced the average live root surface area and number of living root in each soil layer ($P < 0.01$) (Fig. 1).

The response of individual root morphology to different treatments was various between the growing and non-growing seasons (Fig. 2). For example, the N did not significantly change the individual fine root length in the two non-growing seasons ($p > 0.05$), while it increased significantly in the two growing seasons ($p < 0.01$) (Fig. 2). Except for the first non-growing season, the W increased individual root length and surface area significantly ($p < 0.01$) (Fig. 2). Overall, the interaction of NW did not significantly alter the morphology of individual fine root ($p > 0.05$).

Fine root dynamics

In the first observation period, the N, W and the interaction of NW all significantly reduced the annual fine root average production and mortality (Hereinafter referred to as FRP and FRM, $p < 0.01$). Compared with the treatment of N and W alone, the interaction of NW significantly increased FRM and FRP ($P < 0.05$) (Table 1), but this difference was not significant in the second observation period. Compared to the control, the N, W and the interaction of NW did not significantly change the annual turnover rate of fine root (Hereinafter referred to as FRT, Table 1). In the second observation period, only the N reduced both FRM and FPM significantly ($P < 0.01$), and all three treatments (N, W, NW) significantly increased FRT (Table 1).

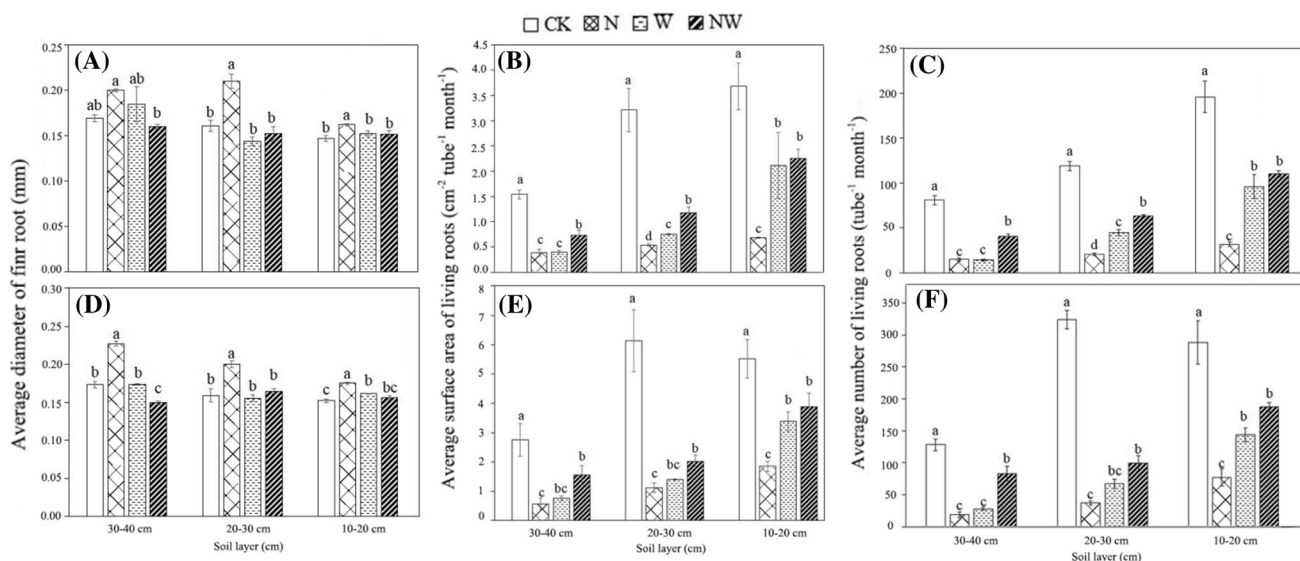


Fig. 1 A, B and C are the effects of N addition, precipitation reduction and their interaction on the average diameter of fine roots, average surface area and average number of living roots in different soil

layers during the first observation period. D, E, F are for the second observation period, as above. ($n = 3$, $P < 0.05$). Different letters (lowercase letters) represent statistical significances among treatments

Fig. 2 Effects of N addition, precipitation reduction and their interaction on the average diameter of fine roots, individual surface area and length of living roots in growing season and non-growing season ($n = 3, P < 0.05$). Different letters (lowercase letters) represent statistical significances among treatments

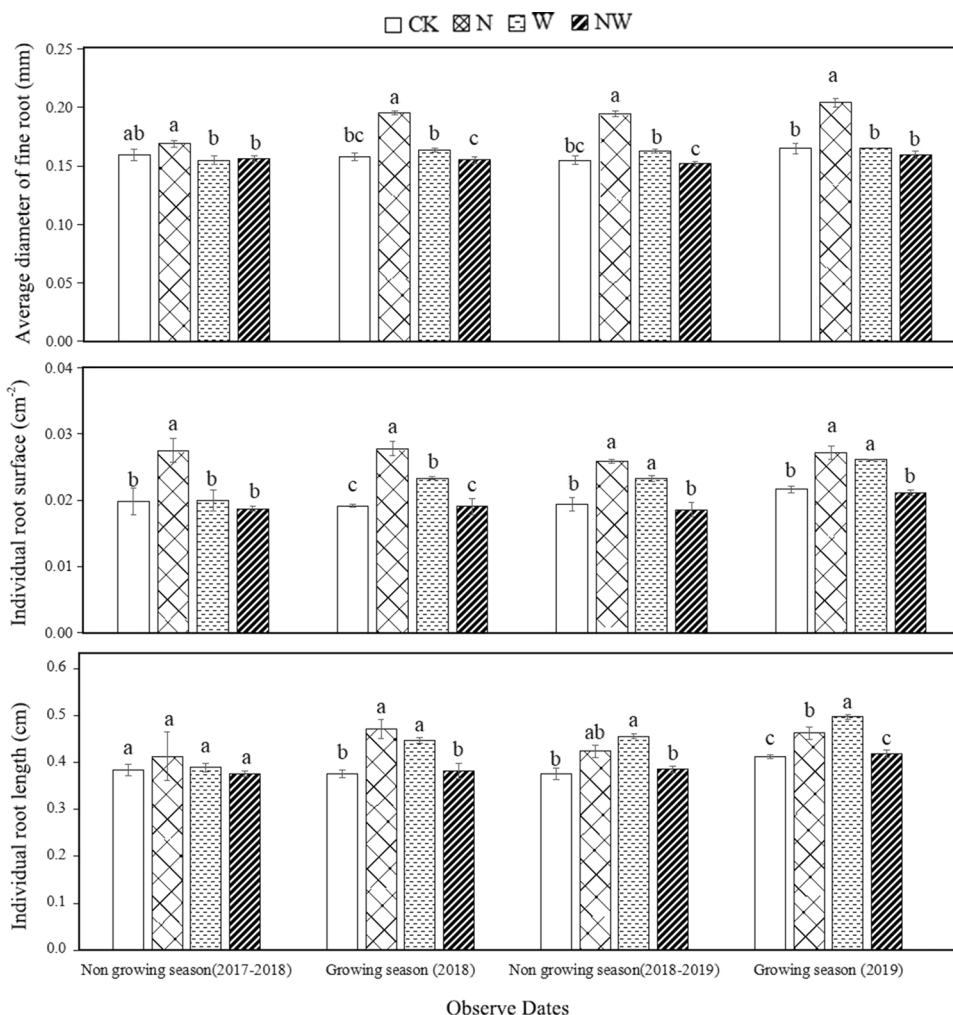


Table 1 Effects of different treatments on fine root dynamics during 2017/11–2019/10

Year	Treatment	FRT (a ⁻¹)	FRP (mm cm ⁻² a ⁻¹)	FRM (mm cm ⁻² a ⁻¹)
2017/11–2018/10	CK	1.41 ± 0.09 (ab)	1.75 ± 0.14 (a)	0.67 ± 0.02 (a)
	N	1.35 ± 0.03 (ab)	0.29 ± 0.03 (d)	0.16 ± 0.01 (d)
	W	1.25 ± 0.05 (b)	0.84 ± 0.10 (c)	0.32 ± 0.04 (c)
	NW	1.53 ± 0.06 (a)	1.36 ± 0.03 (b)	0.51 ± 0.06 (b)
2018/11–2019/10	CK	0.87 ± 0.02 (c)	1.07 ± 0.08 (a)	0.75 ± 0.11 (a)
	N	1.31 ± 0.04 (a)	0.65 ± 0.05 (b)	0.38 ± 0.01 (b)
	W	1.07 ± 0.07 (b)	0.67 ± 0.11 (b)	0.72 ± 0.04 (a)
	NW	1.14 ± 0.06 (b)	0.83 ± 0.10 (b)	0.84 ± 0.01 (a)

Each value is the mean (SE) of three replications, and statistical analysis unit is each plot. Different letters represent statistical significances among different treatments for the same year

The fine root average monthly production and mortality (hereinafter referred to FRP_m and FRM_m) showed obvious seasonal variation pattern, and their variation trends were multi-peak distribution (Figs. 3 and 4). The FRP_m and FRM_m showed a trend of increasing, then decreasing and then increasing in all four treatments, which were higher in the growing season than in the non-growing season. The

maximum values occurred in the growing season and the minimum values in the non-growing season. In the second growing season, the peak value of the N treatment occurred the latest.

The effects of N, W and the interaction of NW on fine root dynamics differed between the growing and non-growing seasons (Figs. 3 and 4). For FRP_m, this difference

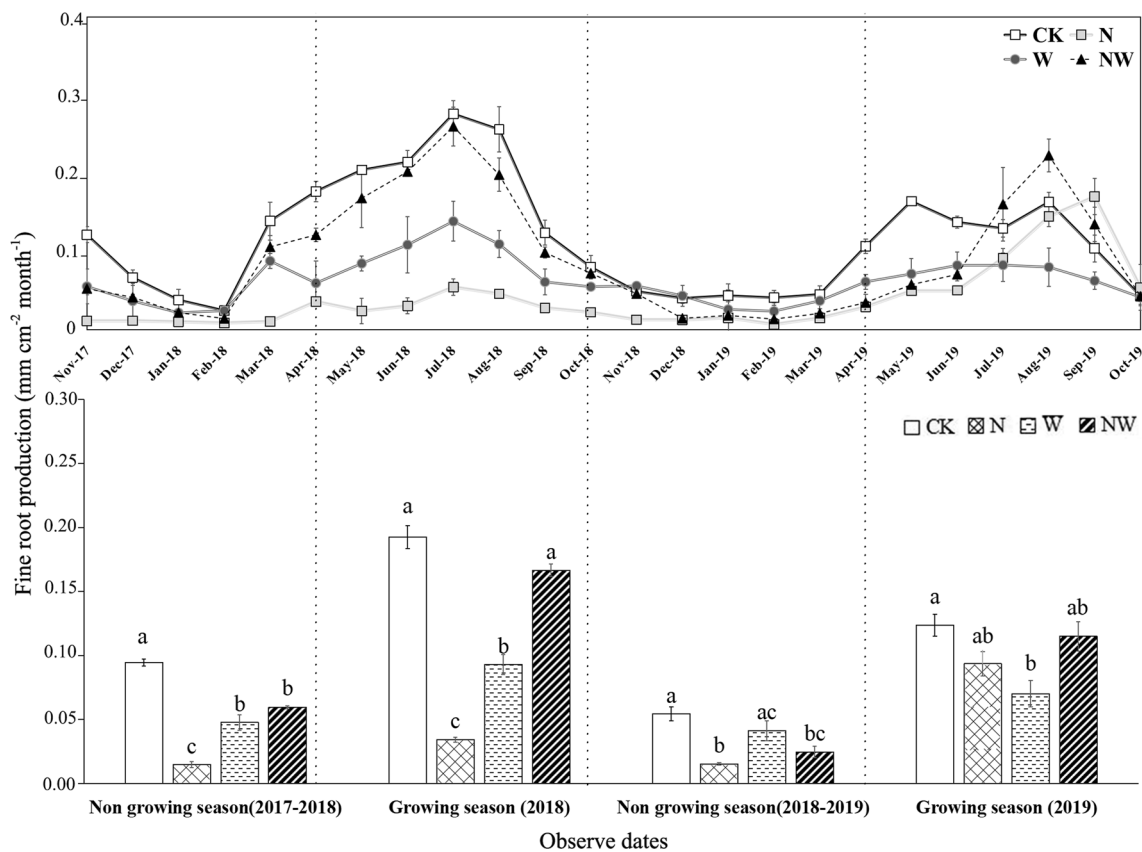


Fig. 3 Effects of different treatments on the seasonal dynamics of monthly fine root production ($n=3$, $P<0.05$). Different letters (lowercase letters) represent statistical significances among treatments

occurred mainly in the second observation period. Compared with the control, the FRP_m of N and the interaction of NW decreased significantly in the non-growing season, but there was no significant difference in the growing season, while the trend of W was opposite (Fig. 3). For FRM_m , this difference mainly occurred in the first observation period. The FRM_m of N and W had no significant change in the non-growing season, but decreased significantly in the growing season (Fig. 4).

Overall, FRP_m in all treatments was lower in the second growing season than in the same period of the first observation period. The FRP_m decreased by 25.62% (control), 20.74% (W) and 38.62% (NW), respectively; however, the N increased by 122.10%. FRM_m in all treatments was higher in the second non-growing season than in the same period of the first observation period, which increased by 12.17% (control), 128.90% (N), 122.18% (W) and 65.93% (NW), respectively.

The median life span of fine root differed significantly among treatments at the same birth time based on the log-rank test ($p<0.01$) (Table 2, Fig. 5). Compared with the control, the N, W and the interaction of NW all significantly reduced the median life span of fine root born at each period

(Table 2). For all treatments, the median life span of fine root born in the first growing season was the longest, while that of fine root born in the second observation season was the shortest (Table 2, Fig. 5). For the same treatment, the median life span of fine root born in the growing season was greater than that of fine root born in the non-growing season, and the median life span of fine root born in the first observation period was greater than that of fine root born in the second observation period (Table 2, Fig. S4).

Cox proportional hazards regression analysis showed that root length, soil layer and birth period all had a significant effect on fine root life span when controlling for other factors, but the effect of average fine root diameter was not significant (Table 3). One-centimeter increase root length increased life span by 1.1% according to the hazard ratios for each year (e.g., $100(e^{-0.177}-1)=16.2\%$). The risk of fine root mortality increased by 40.4% (e.g., $[1.404-1]\times 100\%$) and 33.5% (e.g., $[1.335-1]\times 100\%$) with the increasing soil layer, respectively. This meant that the median lifespan of fine roots surviving in the 10–20-cm soil layer was greater than that of the bottom layer (Table 3, Fig. S3). The N, W and the interaction of NW all

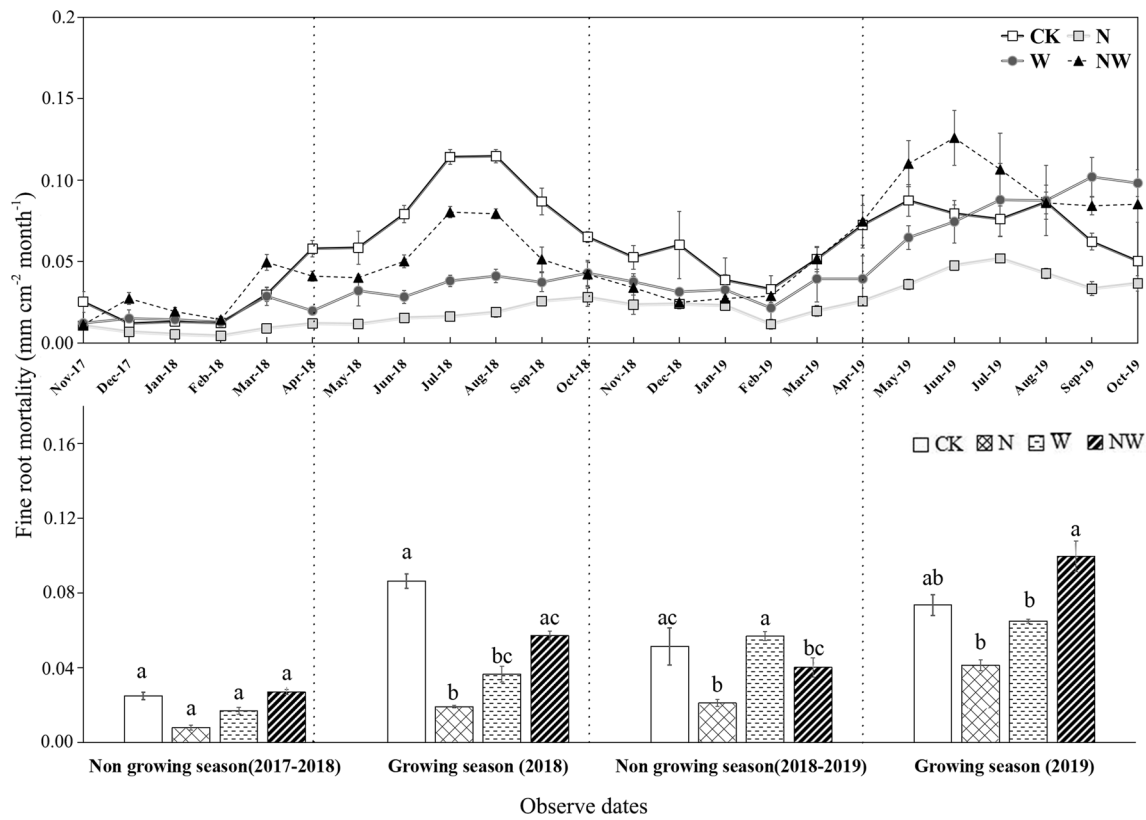


Fig. 4 Effects of different treatments on the seasonal dynamics of monthly fine root mortality ($n=3$, $P<0.05$). Different letters represent statistical significances among treatments

Table 2 Effects of different treatments on the mean life span (MeaL) and median life span (MedL) \pm SE (d) of fine roots

Time of birth	CK		N		W		NW	
	MeaL(d)	MedL (d)	MeaL(d)	MedL (d)	MeaL(d)	MedL (d)	MeaL(d)	MedL (d)
NGS 1	393 \pm 3	326 \pm 9(a)	271 \pm 9	198 \pm 6(b)	359 \pm 7	293 \pm 12(c)	334 \pm 6	268 \pm 5(d)
GS 1	386 \pm 2	489 \pm 0(a)	309 \pm 4	308 \pm 6(b)	340 \pm 3	260 \pm 4(c)	344 \pm 2	369 \pm 4(d)
NGS 2	269 \pm 3	ND(a)	209 \pm 4	233 \pm 3(b)	234 \pm 3	264 \pm 4(c)	209 \pm 4	208 \pm 4(d)
GS 2	140 \pm 1	ND(a)	119 \pm 2	118 \pm 5(c)	135 \pm 2	155 \pm 2(b)	125 \pm 1	ND(b)
NGS	400 \pm 3	373 \pm 8(a)	266 \pm 6	215 \pm 4(b)	349 \pm 5	273 \pm 8(c)	314 \pm 5	254 \pm 5(d)
GS	368 \pm 2	462 \pm 6(a)	286 \pm 4	271 \pm 6(d)	315 \pm 3	336 \pm 5(c)	330 \pm 2	360 \pm 4(b)
OP1	426 \pm 2	462 \pm 7(a)	318 \pm 5	274 \pm 5(c)	371 \pm 4	337 \pm 5(b)	379 \pm 3	345 \pm 3(b)
OP2	262 \pm 2	265 \pm 0(a)	194 \pm 3	203 \pm 4(c)	210 \pm 3	209 \pm 6(b)	193 \pm 3	171 \pm 6(c)

Median root life span with different letters is significantly different among treatments at $P<0.05$ (log-rank test by product-limit (Kaplan–Meier) survival analysis). ND was not available or unable to be calculated because 50% survival was not reached. NGS 1 represents the first non-growing season. GS 1 represents the first growing season. NGS 2 represents the second non-growing season. GS 2 represents the second growing season. OP1 represents the first observation period. OP2 represents the second observation period

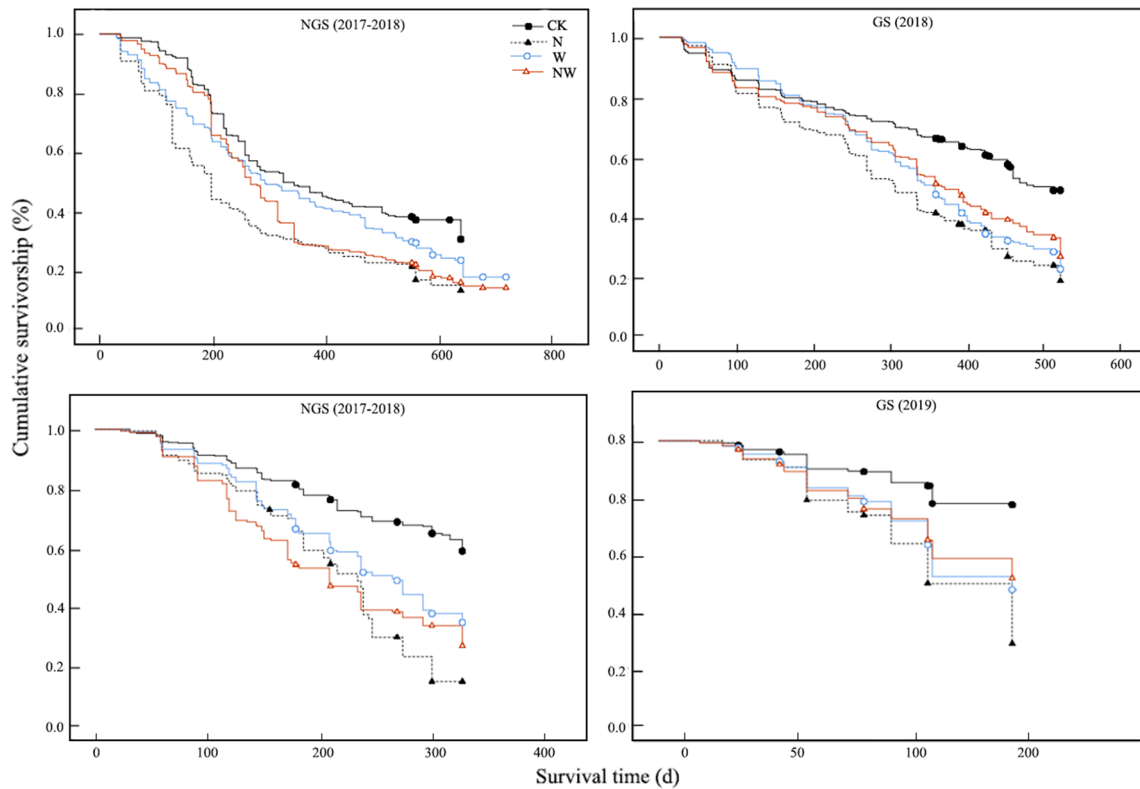


Fig. 5 Survivorship curves of fine roots born in different observation period. NGS (2017–2018) represents the first non-growing season. GS (2018) represents the first growing season. NGS (2018–2019)

represents the second non-growing season. GS (2019) represents the second growing season

Table 3 Results of Cox proportional hazards regression analyses of root life span for all roots appearing from 2017/11 to 2019/10

Effect factors	B	SE	Wald	df	P value	Hazard ratio
Length (cm)	- 0.177	0.032	29.677	1	0.000	0.838
Diameter (mm)	0.209	0.130	2.600	1	0.107	1.233
Depth (cm) (Ref* = 10–20-cm soil layer)	-	-	389.686	2	0.000	-
20–30-cm soil layer	0.340	0.021	265.300	1	0.000	1.404
30–40-cm soil layer	0.289	0.018	266.288	1	0.000	1.335
Time of birth (Ref* = Non-growing season 2017- 2018)	-	-	1023.356	3	0.000	-
Growing season (2018)	- 0.276	0.019	213.446	1	0.000	0.759
Non-growing season (2018–2019)	0.182	0.028	42.478	1	0.000	1.200
Growing season (2019)	0.633	0.033	376.186	1	0.000	1.883
Treatment (Ref* = CK)	-	-	1207.255	3	0.000	-
N	0.708	0.026	715.799	1	0.000	2.031
W	0.550	0.021	697.332	1	0.000	1.734
NW	0.510	0.020	668.884	1	0.000	1.665

*The reference soil layer, time of birth or treatment to which life span of roots of deeper soil layer, of roots born in other periods, of other treatments was compared

significantly increased the risk of fine root mortality, with the lowest survival rate of fine root under the N (Table 3). Thus, in terms of the degree of change in mortality risk, the N had the strongest effect on the median life span of fine roots, while the soil layer had the least.

Discussion

Effects of N, W and the interaction of NW on fine root morphology

Root distribution patterns in the soil can be influenced by a combination of both biotic and abiotic factors within the soil, and the heterogeneity of the soil resource can also lead to variation in the vertical and horizontal distribution of the root system (Schenk and Jackson 2002; Gerland et al. 2002). Previous studies have shown that the number, distribution pattern, production and mortality of fine root decrease with the increase of soil depth, which is consistent with our results (Wang et al. 2019; Li et al. 2020, Wang et al. 2020a). The morphological characteristics of fine root in response to the N and W and the interaction of NW may be influenced both by changes in the physical and chemical properties of the soil and by their own growth strategies. On the one hand, soil bulk density and nutrient content may decrease with the increase of soil layer, which is not conducive to the growth of fine root (Clark et al. 2003; Li et al. 2020). The root system balance the demand of resource transport and absorption by building larger diameter root, which leads to the decrease of the average number of living root and the surface area of monthly living root (Eissenstat and Yanai 1997).

For W, drought stress leads to a reduction in soil water content, which may result in the root system having no sufficient resources to build more fine roots (Idol et al. 2000; Eldhuset et al. 2013; Zang et al. 2014). The increase of drought stress in the second observation period and the decrease of snow cover thickness in non-growing season will lead to the deepening of soil freezing. In this period, the existing living fine roots had a larger diameter to resist the influence of external adverse environmental factors, so as to reduce the mortality (Guo et al. 2008; McCormack and Guo 2014). Under drought stress, the interaction of NW may enhance uptake capability by selecting for a reduction in the average diameter of bottom fine root. It was also found that in the two observation periods, the average number of living root and surface area of fine root in the bottom layer were higher than those in the W treatment (some soil layers reached a significant level). This indicated that the drought resistance of the fine root in the bottom layer might be enhanced to a certain extent by increasing nitrogen on a year-round time scale.

In different periods, the high plasticity of individual fine root morphology enables it to change its morphological characteristics when soil nutrients and water changes, so as to drive the change of root survival strategy (McCormack and Iversen 2019). The aboveground part basically stopped growing in the non-growing season and entered the dormancy state. At this time, there was little demand for root foraging nutrients. The N makes fine roots only need to increase the average diameter or the individual surface area to meet the nutrient requirements of plant. In the growing season, the physiological activities of plant are more vigorous (Persson 1985; Wang et al. 2016; Wang et al. 2020a, b). At the same time, nitrogen addition may increase the physiological activity of fine root (Clark et al. 2003; Burton et al. 2012; Tu et al. 2015; Li et al. 2020), and they need to increase single root length to maintain the balance between absorption and transport function. For W, drought stress causes fine roots to increase their individual root length and surface area in order to expand their range and area of contact with the soil and thereby explore and exploit new soil resources (Robinson et al. 2003; Zhou et al. 2019). Long-term W treatments may make the soil more permeable so that the fine roots have less resistance to developing new soil spaces (Robinson et al. 2003; Barber and Silberbush 2015). So the fine roots that are living in the soil bottom layer may not need to increase in diameter to enhance the ability to resist soil resistance.

Overall, the minirhizotrons do not distinguish the fine roots of different tree species and what our study presents is the combined results of the changes in the fine root morphology of several tree species and their interactions with several environmental factors, the internal differences of which we cannot know any more.

Effects of N, W and the interaction of NW on fine root dynamics

Production and turnover of fine root

Fine root dynamics control the potential uptake and cycling of nutrients and water in forest ecosystems (Hendricks et al. 1993; Matamala et al. 2003). Our results indicate that nitrogen addition reduces fine root production, which is similar to the results of previous studies (Wang et al. 2019; Yuan and Chen 2010; Peng et al. 2017). This response may be beneficial for root growth, as the cost of putting large amounts of carbon into fine root production may outweigh its benefits under conditions of high nutrient utilization. Optimal allocation theory also suggests that an increase in soil nitrogen content leads to a decrease in carbon input to the root system by the plant and a consequent decrease in fine root production (Eissenstat and Yanai 1997; Yan et al. 2017).

Burton et al. (2012) considered that the turnover rate of fine root with the increase of nitrogen availability might be an instantaneous response to the increase of soil nitrogen availability (Mei et al. 2007). As plants adapt to an environment of increased nitrogen effectiveness, the rate of fine root turnover may occur accordingly to accommodate changes in overall survival strategies. This study also showed that nitrogen addition significantly increased fine root turnover while significantly decreasing fine root production, a situation that could lead to a steady decrease in fine root biomass, so this state may not persist. The significant increase in fine root turnover after nitrogen addition may be due to the fact that, on the one hand, the N treatment may significantly increase the fine root nitrogen content and thus promote fine root respiration, and if the input of photosynthetic products to the root system does not meet its respiratory consumption, the fine root life will be shortened and turnover will be accelerated (Gough et al. 2004; Pregitzer et al. 2000).

Drought as a multifaceted environmental stress may have a negative impact on fine root production (Brunner et al. 2015). Precipitation reduction may inhibit the physiological processes of fine root and making them less productive, while some short-term precipitation reduction experiments have found that drought increases fine root production (Gaul et al. 2008a, b). As of 2019, precipitation reduction treatments within this sample plot have been carried out for 10 years. The response of fine root dynamics to long-term water-reducing treatments may differ from short-term (Vicca et al. 2012; Brunner et al. 2015). We also found that FRP was higher in the interaction of NW than that in the N and W alone (not significant in the second year). The reason for this may be that the nitrogen addition may alleviate the degree of drought in the root system to some extent (Zhang et al. 2020). Drought-induced reductions in net photosynthesis and productivity of plants result in a lower proportion of photosynthetic products being allocated to the root system (Hagedorn et al. 2016). In addition, the reduced mobility of nutrients in the soil due to lower soil water content limits the availability of nutrients to plants (Dijkstra et al. 2015; Raven et al. 2018). Plants under drought stress may optimize their root carbon allocation strategies to maintain nutrient uptake. A recent $^{13}\text{CO}_2$ pulse-labeling experiment has shown that, under drought conditions, plants invest more in carbon allocation to rhizodeposition or to mycorrhizal symbiosis and less in root biomass. However, under the interaction of N addition and water reduction, plants allocated more C to the root system and less to the soil to counteract the drought effect (Wang et al. 2021). During the first observation period, N addition significantly reduced soil pH (Table S2), which may have led to an increase Al^{3+} concentration in soil and thus inhibited root production (Joslin et al. 2000; Smithwick et al. 2013). However, the water reduction treatment may have reduced this negative effect by decreasing

the contact area of fine roots with the soil. This may be the reason for the higher FRP of the NW treatment compared to the N addition or water reduction treatment alone. However, in the second observation period, the reduction in total precipitation led to a significant increase in the degree of drought stress, at which time the interaction of NW provided limited relief from the adverse environmental factors, so the differences in FRP between the three treatments (N, W and NW) were not significant. This is consistent with our hypotheses 1 and 2.

The precipitation reduction did not significantly change the annual average turnover of fine root in the first observation period but significantly increased it in the second year, which may be related to changes in snow thickness during the two non-growing seasons. In the first non-growing season the thick winter snowpack provides good protection for the fine roots against cold conditions and the spring snowmelt also provides them with water and nutrients to withstand the drought of the growing season (Groffman et al. 2001). However, the second non-growing season has a thin snowpack and limited nutrient and water availability from spring snowmelt. In such conditions, the increased freezing of the soil encourages root mortality and causes more damage to the fine roots (Henry and Biochemistry 2007). In addition, the isolated precipitation after the growing season will accelerate the lignification of fine root and reduce their absorption capacity (Anna and Tuomo 2010). Fine roots may choose to enhance water uptake efficiency by increasing old root mortality and new root emergence, which will allow fine roots to optimize water transport and uptake efficiency and faster turnover rates (Eissenstat et al. 2000; Chenlemuge et al. 2013). During the same period, for the NW treatment, an increase in respiration rate and a decrease in uptake capacity of the fine root occurred simultaneously, which led the fine roots to choose to increase the turnover rate to ensure the efficiency of nitrogen uptake.

Effect of different treatments on the life span of fine root

The belowground process of temperate forest has obvious seasonal variation due to the influence of climate factors (Hendrick et al. 1996; Pregitzer et al. 2000). Our study found that the FRPm and FRMm of all treatments tended to decrease at the end of the growing season and increase at the end of the non-growing season, even reaching a stage peak. This may be related to rising soil temperatures in spring and lower soil temperatures later in the growing season.

Compared to the control, the N, W and the interaction of NW all significantly reduced the median life span of fine root in all observation periods (Fig. 5) which is consistent with the trend in fine root turnover rate in the second observation period. For all treatments, the life span of fine root born in the growing season was significantly longer than that born

in the non-growing season, which was different from some previous studies (Jones et al. 2003; Gaul et al. 2008b). The life span of fine root is influenced by a number of environmental factors at the same time, and their life span may be the result of competition between organs for photosynthetic products (Yanai and Eissenstat 2002). Some studies suggest that the carbon source for fine roots born before the spring leaf spread is mainly carbohydrates stored in the previous year, and that once these sources are depleted the fine root die (Marshall and Waring 1985; Pregitzer 2003). Fine roots born earlier than leaves during the non-growing season may compete with aboveground parts for photosynthetic products and thus cause changes in internal source-sink relationships (Pregitzer 2003). Insufficient carbohydrates allocated to the belowground portion of fine root will result in a forced reduction in the longevity of fine root born during the non-growing season. During the growing season, the aboveground foliage growth is almost complete. Even if the fine roots are more physiologically active during this period, a large amount of photosynthetic products can be transported to the ground in time. In addition, higher soil temperature and precipitation during the growing season are also more suitable for fine roots to survive and their life span will be higher than that of fine root born during the non-growing season.

Previous studies of fine root anatomy in this sample site have shown that increasing nitrogen, reducing water and their interaction all significantly reduced fine root cortex thickness in some of the dominant species during the growing season and increased it significantly in the non-growing season (Zhang et al. 2020; Yan 2020). We hypothesize that fine roots will choose to reduce the thickness of the cortex during the growing season to optimally uptake and transport capacity, so that those with thicker cortex (and lower uptake capacity) may be eliminated at this stage, resulting in fine roots born out of season barely lasting more than a year. These results are consistent with our hypothesis 3, that is, the dynamic and morphological responses of fine root to different treatments are various in growing and non-growing season.

Factors influencing the longevity of fine root

Cox regression analysis showed that when only a single factor was considered, root length, season of birth, soil layer and nitrogen addition, precipitation reduction and their interaction all had a highly significant effect on fine root longevity, while mean root diameter had no significant effect on fine root longevity. Firstly, our study was on mixed coniferous forest, which resulted in a mixed pattern of variation in fine root longevity across multiple species (Beyer et al. 2013; Zhang et al. 2020). That may have resulted in average

diameter not having a significant effect on fine root longevity. In some pure plantations, the diameter of the fine root may be a key determinant of fine root longevity (Guo et al. 2008; Gu et al. 2011; Gu 2017). Secondly, the turnover of fine root did not decrease with increasing diameter either. Indeed some studies suggest that diameter may not be the only factor determining fine root longevity and turnover, and the specific root length and root tissue density seem to be more convincing in explaining fine root dynamics (Ryser 1996; Weemstra et al. 2020).

Cox regression analysis showed that when only root length was considered, the longer the fine roots, the higher the survival rate, perhaps because longer fine roots are able to explore new soil resources to a greater extent, and the benefits of maintaining fine roots outweigh the maintenance costs, so they live longer. Cox regression with the dynamics of fine root production and mortality both indicated that the survival rate of fine root in the second non-growing season was extremely significantly reduced. This was associated with a reduction in snow thickness (Gaul et al. 2008b; Sutinen et al. 2014), and increased fine root mortality during the non-growing season may also have been the main reason for the reduced fine root production in some of the treatments during the second observation period and the delayed onset of peak FRP in the nitrogen addition treatments during the growing season.

We found that the fine root mortality increased with the deepening of soil layer, which was consistent with the trend of turnover of fine root in different soil layers (Table S1; Fig. S3). This may be because the nitrogen availability of deep soil in the sample plot cannot meet the needs of plant, and the bottom fine roots obtain nutrients through rapid turnover. The nitrogen addition may increase the content of available nitrogen in deep soil, thus breaking the balance and increasing the energy consumption of fine root respiration. The content of available nitrogen in surface soil is generally higher than that in deep soil, and the fine roots in this soil layer will get more carbon allocation (Pearcy et al. 1987). The bottom soil is located at the end of the root system, it takes a long time to get carbon, and it is more likely to die if the carbon supplement is not timely (King et al. 2002).

Conclusions

The nitrogen addition and precipitation reduction and their interaction significantly reduced the monthly average number and surface area of living root in each soil layer, but the effect on the morphology of individual root was different from that of the whole. This result suggests that when fine roots are exposed to environmental stress, fine roots

may drive changes in overall survival strategies by altering the morphology of individual root. Both long-term nitrogen addition and precipitation reduction and their interaction reduced the annual production of fine root. The different treatments did not significantly alter the average annual turnover of fine root in the first observation period. Compared with the control, different treatments did not significantly change the turnover of fine root in the first observation period, but significantly increased it in the second observation period, which may decrease the standing stock of fine root. These results may also be due to changes in fine root vertical distribution pattern, growth strategy and ecosystem carbon cycle caused by changes in environmental factors. At present, the response mechanism of fine root dynamics of different dominant tree species to the nitrogen addition, precipitation reduction and their interaction is still unclear, and further research is needed.

Supplementary Information The online version contains supplementary material available at <https://doi.org/10.1007/s10342-022-01445-9>.

Acknowledgements We gratefully acknowledge associate professor Guanhua Dai from Changbai Mountain Forest Ecosystem research Station, Institute of Applied Ecology, Chinese Academy of Sciences, for his advice about field experiment design and suggestions on an earlier draft of this manuscript.

Author's contributions QW and YX designed the study, got grants from the foundation, supervised data collection and edited the manuscript. QW, ND, YX, JZ, GY and GL contributed to the whole manuscript preparation and design and wrote the main manuscript text. QW, ND, YX, JZ, GY and GL prepared all figures, ND, YX, GY, JZ, GL and QW prepared field experiments, prepared tables and collected literatures. All authors reviewed the manuscript.

Funding This research was supported by grants from the National Natural Science Foundation of China (41575137, 41773075).

Availability of data and material Data are available from the corresponding author on reasonable request.

Declarations

Conflicts of interest The authors declare that they have no conflict of interest.

References

- Abramoff RZ, Finzi AC (2015) Are above-and below-ground phenology in sync? *New Phytol* 205(3):1054–1061. <https://doi.org/10.1111/nph.13111>
- Barber SA, Silberbush M (2015) Plant root morphology and nutrient uptake, roots, nutrient and water influx, and plant. *Growth* 49:65–87. <https://doi.org/10.2134/asaspecpub49>
- Beyer F, Hertel D, Leuschner C (2013) Fine root morphological and functional traits in *Fagus sylvatica* and *Fraxinus*

- excelsior saplings as dependent on species, root order and competition. *Plant Soil* 373(1):143–156. <https://doi.org/10.1007/s11104-013-1752-7>
- Bonan GB (2008) Forests and climate change: forcings, feedbacks, and the climate benefits of forests. *Science* 320(5882):1444–1449. <https://doi.org/10.1126/science.1155121>
- Brunner I, Herzog C, Dawes MA, Arend M, Sperisen C (2015) How tree roots respond to drought. *Front Plant Sci* 6:547. <https://doi.org/10.3389/fpls.2015.00547>
- Burton AJ, Jarvey JC, Jarvi MP, Zak DR, Pregitzer KS (2012) Chronic N deposition alters root respiration-tissue N relationship in northern hardwood forests. *Glob Change Biol* 18(1):258–266. <https://doi.org/10.1111/j.1365-2486.2011.02527.x>
- Chenlemuge T, Hertel D, Dulamsuren C, Khishigiargal M, Leuschner C, Hauck M (2013) Extremely low fine root biomass in *Larix sibirica* forests at the southern drought limit of the boreal forest. *Flora-Morphol Distrib, Funct Ecol Plants* 208(8–9):488–496
- Clark LJ, Whalley WR, Barraclough PB (2003) How do roots penetrate strong soil?. In: *Roots: the dynamic interface between plants and the earth*. Springer, Dordrecht, pp. 93–104. <https://doi.org/10.1023/A:1026140122848>
- Comas L, Becker S, Cruz VM, Byrne PF, Dierig DA (2013) Root traits contributing to plant productivity under drought. *Front Plant Sci* 4:442. <https://doi.org/10.3389/fpls.2013.00442>
- Deng L, Peng C, Zhu G, Chen L, Liu Y, Shangguan Z (2018) Positive responses of belowground C dynamics to nitrogen enrichment in China. *Sci Total Environ* 616:1035–1044. <https://doi.org/10.1016/j.scitotenv.2017.10.215>
- Dewar RC (1993) A root-shoot partitioning model based on carbon-nitrogen-water interactions and Munch phloem flow. *Funct Ecol*. <https://doi.org/10.2307/2390216>
- Dijkstra FA, He M, Johansen MP, Harrison JJ, Keitel C (2015) Plant and microbial uptake of nitrogen and phosphorus affected by drought using 15N and 32P tracers. *Soil Biol Biochem* 82:135–142. <https://doi.org/10.1016/j.soilbio.2014.12.021>
- Eissenstat DM, McCormack ML, Du Q (2013) Global change and root lifespan. *The Hidden Half, Fourth Edition*. CRC Press, In *Plant Roots*, pp 399–412
- Eissenstat DM (2000) Root dynamics and global change: an ecosystem perspective. *New Phytol* 147:33–42
- Eissenstat DM, Yanai RD (1997) The ecology of root lifespan. *Adv Ecol Res* 27:1–60. [https://doi.org/10.1016/S0065-2504\(08\)60005-7](https://doi.org/10.1016/S0065-2504(08)60005-7)
- Eldhuset TD, Nagy NE, Volařík D, Børja I, Gebauer R, Yakovlev IA, Krokene P (2013) Drought affects tracheid structure, dehydrin expression, and above-and belowground growth in 5-year-old Norway spruce. *Plant Soil* 366(1):305–320. <https://doi.org/10.1007/s11104-012-1432-z>
- Finér L, Ohashi M, Noguchi K, Hirano Y (2011) Fine root production and turnover in forest ecosystems in relation to stand and environmental characteristics. *Ecol Manag* 262(11):2008–2023. <https://doi.org/10.1016/j.foreco.2011.08.042>
- Fitzhugh RD, Driscoll CT, Groffman PM, Tierney GL, Fahey TJ, Hardy JP (2001) Effects of soil freezing disturbance on soil solution nitrogen, phosphorus, and carbon chemistry in a northern hardwood ecosystem. *Biogeochemistry* 56(2):215–238. <https://doi.org/10.1023/A:1013076609950>
- Galloway JN, Dentener FJ, Capone DG, Boyer EW, Howarth RW, Seitzinger SP, Asner GP, Cleveland CC, Green PA, Holland EA, Karl DM, Vorosmarty CJ (2004) Nitrogen cycles: past, present, and future. *Biogeochemistry* 70(2):153–226. <https://doi.org/10.1007/s10533-004-0370-0>
- Galloway JN, Schlesinger WH, Levy H, Michaels A, Schnoor JL (1995) Nitrogen fixation: anthropogenic enhancement-environmental response. *Global Biogeochem Cycles* 9(2):235–252. <https://doi.org/10.1029/95GB00158>

- Gaul D, Hertel D, Leuschner C (2008a) Effects of experimental soil frost on the fine-root system of mature Norway spruce. *J Plant Nutr Soil Sci* 171(5):690–698. <https://doi.org/10.1002/jpln.200700284>
- Gaul D, Hertel D, Borken W, Matzner E, Leuschner C (2008b) Effects of experimental drought on the fine root system of mature Norway spruce. *For Ecol Manage* 256(5):1151–1159. <https://doi.org/10.1016/j.foreco.2008.06.016>
- Gough CM, Seiler JR, Maier CA (2004) Short-term effects of fertilization on loblolly pine (*Pinus taeda* L.) physiology. *Plant Cell Environ* 27(7):876–886. <https://doi.org/10.1111/j.1365-3040.2004.01193.x>
- Groffman PM, Driscoll CT, Fahey TJ, Hardy JP, Fitzhugh RD, Tierney GL (2001) Colder soils in a warmer world: a snow manipulation study in a northern hardwood forest ecosystem. *Biogeochemistry* 56(2):135–150. <https://doi.org/10.1023/a:1013039830323>
- Gu J, Yu S, Sun Y, Wang Z, Guo D (2011) Influence of root structure on root survivorship: an analysis of 18 tree species using a minirhizotron method. *Ecol Res* 26(4):755–762. <https://doi.org/10.1007/s11284-011-0833-4>
- Gu J, Wang Y, Fahey TJ, Wang Z (2017) Effects of root diameter, branch order, soil depth and season of birth on fine root life span in five temperate tree species. *Eur J Forest Res* 136(4):727–738. <https://doi.org/10.1007/s11284-016-1385-4>
- Guo D, Mitchell RJ, Withington JM, Fan PP, Hendricks JJ (2008) Endogenous and exogenous controls of root life span, mortality and nitrogen flux in a longleaf pine forest: root branch order predominates. *J Ecol* 96(4):737–745. <https://doi.org/10.1111/j.1365-2745.2008.01385.x>
- Gerland U, Moroz JD, Hwa T (2002) Physical constraints and functional characteristics of transcription factor–DNA interaction. *Proc Natl Acad Sci* 99(19):12015–12020. <https://doi.org/10.1073/pnas.192693599>
- Hagedorn F, Joseph J, Peter M, Luster J, Pritsch K, Geppert U, Kerner R, Molinier V, Egli S, Schaub M, Liu JF (2016) Recovery of trees from drought depends on belowground sink control. *Nature Plants* 2(8):1–5. <https://doi.org/10.1038/nplants.2016.111>
- Hendrick RL, Pregitzer KS (1996) Temporal and depth-related patterns of fine root dynamics in northern hardwood forests. *J Ecol*. <https://doi.org/10.2307/2261352>
- Hendrick RL, Pregitzer KS (1993) Patterns of fine root mortality in two sugar maple forests. *Nature* 361(6407):59–61. <https://doi.org/10.1038/361059a0>
- Hendricks JJ, Nadelhoffer KJ, Aber JD (1993) Assessing the role of fine roots in carbon and nutrient cycling. *Trends Ecol Evol* 8(5):174–178. [https://doi.org/10.1016/0169-5347\(93\)90143-D](https://doi.org/10.1016/0169-5347(93)90143-D)
- Henry HA (2007) Soil freeze–thaw cycle experiments: trends, methodological weaknesses and suggested improvements. *Soil Biol Biochem* 39(5):977–986. <https://doi.org/10.1016/j.soilbio.2006.11.017>
- Idol TW, Pope PE, Ponder F Jr (2000) Fine root dynamics across a chronosequence of upland temperate deciduous forests. *Ecol Manage* 127(1–3):153–167. [https://doi.org/10.1016/S0378-1127\(99\)00127-9](https://doi.org/10.1016/S0378-1127(99)00127-9)
- Jackson R, Mooney HA, Schulze ED (1997) A global budget for fine root biomass, surface area, and nutrient contents. *Proc Natl Acad Sci* 94(14):7362–7366. <https://doi.org/10.1073/pnas.94.14.7362>
- Jones RH, Mitchell RJ, Stevens GN, Pecot SD (2003) Controls of fine root dynamics across a gradient of gap sizes in a pine woodland. *Oecologia* 134(1):132–143. <https://doi.org/10.1007/s00442-002-1098-y>
- Joslin JD, Wolfe MH, Hanson PJ (2000) Effects of altered water regimes on forest root systems. *New Phytol* 147(1):117–129. <https://doi.org/10.1046/j.1469-8137.2000.00692.x>
- Johnson MG, Tingey DT, Phillips DL, Storm MJ (2001) Advancing fine root research with minirhizotrons. *Environ Exp Bot* 45(3):263–289. [https://doi.org/10.1016/S0098-8472\(01\)00077-6](https://doi.org/10.1016/S0098-8472(01)00077-6)
- Kou L, Jiang L, Fu X, Dai X, Wang H, Li S (2018) Nitrogen deposition increases root production and turnover but slows root decomposition in *Pinus elliptica* plantations. *New Phytol* 218(4):1450–1461. <https://doi.org/10.1111/nph.15066>
- King JS, Albaugh TJ, Allen HL, Buford M, Strain BR, Dougherty P (2002) Below-ground carbon input to soil is controlled by nutrient availability and fine root dynamics in loblolly pine. *New Phytol* 154(2):389–398. <https://doi.org/10.1046/j.1469-8137.2002.00393.x>
- Li FL, McCormack ML, Liu X, Hu H, Bao WK (2020) Vertical fine-root distributions in five subalpine forest types shifts with soil properties across environmental gradients. *Plant Soil* 456(1):129–143. <https://doi.org/10.1007/s11104-020-04706-x>
- Li W, Jin C, Guan D, Wang Q, Wang A, Yuan F, Wu J (2015) The effects of simulated nitrogen deposition on plant root traits: a meta-analysis. *Soil Biol Biochem* 82:112–118. <https://doi.org/10.1016/j.soilbio.2015.01.001>
- Li X, Zhang C, Zhang B, Wu D, Zhu D, Zhang W, Ye Q, Yan J, Fu J (2021) Nitrogen deposition and increased precipitation interact to affect fine root production and biomass in a temperate forest: implications for carbon cycling. *Sci Total Environ* 765:144497. <https://doi.org/10.1016/j.scitotenv.2020.144497>
- Lintunen A, Kalliokoski T (2010) The effect of tree architecture on conduit diameter and frequency from small distal roots to branch tips in *Betula pendula*, *Picea abies* and *Pinus sylvestris*. *Tree Physiol* 30(11):1433–1447. <https://doi.org/10.1093/treephys/tpq085>
- Lima TTS, Miranda IS, Vasconcelos SS (2010) Effects of water and nutrient availability on fine root growth in eastern Amazonian forest regrowth. *Brazil New Phytol* 187(3):622–630. <https://doi.org/10.1111/j.1469-8137.2010.03299.x>
- Liu X, Zhang Y, Han W, Tang A, Shen J, Cui Z, Vitousek P, Erisman JW, Goulding K, Christie P, Fangmeier A, Zhang F (2013) Enhanced nitrogen deposition over China. *Nature* 494(7438):459–462. <https://doi.org/10.1038/nature11917>
- López B, Sabaté S, Gracia CA (2001) Fine-root longevity of *Quercus ilex*. *New Phytol* 151(2):437–441. <https://doi.org/10.1046/j.0028-646x.2001.00189.x>
- Lozanova L, Zhiyanski M, Vangelova E, Doncheva S, Marinov MP, Lazarova S (2019) Dynamics and vertical distribution of roots in European beech forests and Douglas fir plantations in Bulgaria. *Forests* 10(12):1123. <https://doi.org/10.3390/f10121123>
- Lü C, Tian H (2007) Spatial and temporal patterns of nitrogen deposition in China: synthesis of observational data. *J Geophys Res Atmos*. <https://doi.org/10.1029/2006JD007990>
- Lu M, Yang Y, Luo Y, Fang C, Zhou X, Chen J, Li B (2011) Responses of ecosystem nitrogen cycle to nitrogen addition: a meta-analysis. *New Phytol* 189(4):1040–1050. <https://doi.org/10.1111/j.1469-8137.2010.03563.x>
- Li N, Yang Z, Sun Y, Xu H, He Y, Yan T, Ma J (2017) The relationships between root morphology, N absorption and utilization and grain yield in rice with different N use efficiencies. *Sci Agric Sinica* 50(14):2683–2695. <https://doi.org/10.3864/j.issn.0578-1752.2017.14.005>
- Mei L, Gu J, Zhang Z, Wang Z (2010) Responses of fine root mass, length, production and turnover to soil nitrogen fertilization in *Larix gmelinii* and *Fraxinus mandshurica* forests in Northeastern China. *J Res* 15(3):194–201. <https://doi.org/10.1007/s10310-009-0176-y>
- Marshall JD, Waring RH (1985) Predicting fine root production and turnover by monitoring root starch and soil temperature. *Can J for Res* 15(5):791–800. <https://doi.org/10.1139/x85-129>

- Matamala R, Gonzalez-Meler MA, Jastrow JD, Norby RJ, Schlesinger WH (2003) Impacts of fine root turnover on forest NPP and soil C sequestration potential. *Science* 302(5649):1385–1387. <https://doi.org/10.1126/science.1089543>
- McCormack ML, Guo D (2014) Impacts of environmental factors on fine root lifespan. *Front Plant Sci* 5:205. <https://doi.org/10.3389/fpls.2014.00205>
- McCormack ML, Iversen CM (2019) Physical and functional constraints on viable belowground acquisition strategies. *Front Plant Sci* 10:1215. <https://doi.org/10.3389/fpls.2019.01215>
- McCormack ML, Guo D, Iversen CM, Chen W, Eissenstat DM, Fernandez CW, Li L, Ma C, Ma Z, Poorter H, Reich PB (2017) Building a better foundation: improving root-trait measurements to understand and model plant and ecosystem processes. *New Phytol* 215(1):27–37. <https://doi.org/10.1111/nph.14459>
- McMurtrie RE, Iversen CM, Dewar RC, Medlyn BE, Näsholm T, Pepper DA, Norby RJ (2012) Plant root distributions and nitrogen uptake predicted by a hypothesis of optimal root foraging. *Ecol Evol* 2(6):1235–1250. <https://doi.org/10.1002/ece3.266>
- Mei L, Han Y, Yu S, Shi J, Wang Z (2007) Impact factors on fine root seasonal dynamics in *Fraxinus mandshurica* plantations. *Front Forest China* 2(3):298–304. <https://doi.org/10.1007/s11461-007-0048-6>
- Nadelhoffer KJ, Raich JW (1992) Fine root production estimates and belowground carbon allocation in forest ecosystems. *Ecology* 73(4):1139–1147. <https://doi.org/10.2307/1940664>
- Ostertag R (2001) Effects of nitrogen and phosphorus availability on fine-root dynamics in Hawaiian montane forests. *Ecology* 82(2):485–499. [https://doi.org/10.1890/0012-9658\(2001\)082\[0485:EONPA\]2.0.CO;2](https://doi.org/10.1890/0012-9658(2001)082[0485:EONPA]2.0.CO;2)
- Pakkala TA, Köliö A, Lahdensivu J, Pentti M (2015). The effect of climate change on freeze-thaw cycles in Nordic climate. In: Durability of reinforced concrete from composition to protection. Springer, Cham, pp.145–154. https://doi.org/10.1007/978-3-319-09921-7_13
- Pearcy RW, Björkman O, Caldwell MM, Keeley JE, Monson RK, Strain BR (1987) Carbon gain by plants in natural environments. *Bioscience* 37(1):21–29. <https://doi.org/10.2307/1310174>
- Peng Y, Guo D, Yang Y (2017) Global patterns of root dynamics under nitrogen enrichment. *Global Ecol Biogeogr* 26(1):102–114. <https://doi.org/10.1111/geb.12508>
- Persson H (1985) The dynamics of fine roots of forest trees. *Acta Phytogeogr Suecica* 16:215–224
- Pregitzer KS, Hendrick RL, Fogel R (1993) The demography of fine roots in response to patches of water and nitrogen. *New Phytol* 125(3):575–580. <https://doi.org/10.1111/j.1469-8137.1993.tb03905.x>
- Pregitzer KS, King JS, Burton AJ, Brown SE (2000) Responses of tree fine roots to temperature. *New Phytol* 147(1):105–115. <https://doi.org/10.1046/j.1469-8137.2000.00689.x>
- Pregitzer KS (2003) Woody plants, carbon allocation and fine roots. *New Phytol*. <https://doi.org/10.1046/j.1469-8137.2003.00766.x>
- Raven JA, Lambers H, Smith SE, Westoby M (2018) Costs of acquiring phosphorus by vascular land plants: patterns and implications for plant coexistence. *New Phytol* 217(4):1420–1427. <https://doi.org/10.1111/nph.14967>
- Repo T, Sirkkiä S, Heinonen J, Lavigné A, Roitto M, Koljonen E, Sutinen S, Finér L (2014) Effects of frozen soil on growth and longevity of fine roots of Norway spruce. *Forest Ecol Manag* 313:112–122. <https://doi.org/10.1016/j.foreco.2013.11.002>
- Robinson D, Hodge A, Fitter A (2003). Constraints on the form and function of root systems. In *Root ecology*. Springer, Berlin, Heidelberg, pp. 1–31. https://doi.org/10.1007/978-3-662-09784-7_1
- Rossi S, Couture É, Plante X, Morin H (2016) Fine roots and ectomycorrhizal colonization in black spruce subjected to reductions in soil moisture. *Botany* 94(1):23–30. <https://doi.org/10.1139/cjb-2015-0093>
- Ryser P (1996) The importance of tissue density for growth and life span of leaves and roots: a comparison of five ecologically contrasting grasses. *Funct Ecol*. <https://doi.org/10.2307/2390506>
- Santantonio D (1982) Production and turnover of fine roots of mature Douglas-fir in relation to site. *Mol Biotechnol*. <https://doi.org/10.1007/s12033-014-9829-y>
- Schenk HJ, Jackson RB (2002) Rooting depths, lateral root spreads and below-ground/above-ground allometries of plants in water-limited ecosystems. *J Ecol*. <https://doi.org/10.2307/3072232>
- Sutinen S, Roitto M, Lehto T, Repo T (2014) Simulated snowmelt and infiltration into frozen soil affected root growth, needle structure and physiology of Scots pine saplings. *Boreal Environ Res* 19(5):281–295
- Smithwick EA, Eissenstat DM, Lovett GM, Bowden RD, Rustad LE, Driscoll CT (2013) Root stress and nitrogen deposition: consequences and research priorities. *New Phytol* 197(3):712–719. <https://doi.org/10.1111/nph.12081>
- Song Y, Zou Y, Wang G, Yu X (2017) Altered soil carbon and nitrogen cycles due to the freeze-thaw effect: a meta-analysis. *Soil Biol Biochem* 109:35–49. <https://doi.org/10.1016/j.soilbio.2017.01.020>
- Tu LH, Peng Y, Chen G, Hu HL, Xiao YL, Hu TX, Liu L, Tang Y (2015) Direct and indirect effects of nitrogen additions on fine root decomposition in a subtropical bamboo forest. *Plant Soil* 389(1):273–288. <https://doi.org/10.1007/s11104-014-2353-9>
- Vicca S, Luysaert S, Peñuelas J, Campioli M, Chapin FS III, Ciais P, Heinemeyer A, Höglberg P, Kutsch WL, Law BE, Malhi Y, Janssens IA (2012) Fertile forests produce biomass more efficiently. *Ecol Lett* 15(6):520–526
- Wang C, Geng Z, Chen Z, Li J, Guo W, Zhao TH, Cao Y, Shen S, Jin D, Li MH (2017) Six-year nitrogen–water interaction shifts the frequency distribution and size inequality of the first-order roots of *Fraxinus mandshurica* in a mixed mature *Pinus koraiensis* forest. *Front Plant Sci* 8:1691. <https://doi.org/10.3389/fpls.2017.01691>
- Wang N, Wang C, Quan X (2020a) Variations in fine root dynamics and turnover rates in five forest types in northeastern China. *J Forest Res* 31(3):871–884. <https://doi.org/10.1007/s11676-019-01065-x>
- Wang C, Han S, Zhou Y, Yan C, Cheng X, Zheng X, Li MH (2012) Responses of fine roots and soil N availability to short-term nitrogen fertilization in a broad-leaved Korean pine mixed forest in northeastern China. *PLoS ONE* 7(3):e31042. <https://doi.org/10.1371/journal.pone.0031042>
- Wang R, Cavagnaro TR, Jiang Y, Keitel C, Dijkstra FA (2021) Carbon allocation to the rhizosphere is affected by drought and nitrogen addition. *J Ecol* 109(10):3699–3709. <https://doi.org/10.1111/1365-2745.13746>
- Wang W, Mo Q, Han X, Hui D, Shen W (2019) Fine root dynamics responses to nitrogen addition depend on root order, soil layer, and experimental duration in a subtropical forest. *Biol Fertil Soils* 55(7):723–736. <https://doi.org/10.1007/s00374-019-01386-3>
- Wang Y, Li Z, Wang Z, Gu J (2020b) Functional trait plasticity but not coordination differs in absorptive and transport fine roots in response to soil depth. *Forests* 11(1):42. <https://doi.org/10.3390/f11010042>
- Weemstra M, Kiorapostolou N, van Ruijven J, Mommer L, de Vries J, Sterck F (2020) The role of fine-root mass, specific root length and life span in tree performance: a whole-tree exploration. *Funct Ecol* 34(3):575–585. <https://doi.org/10.1111/1365-2435.13520>
- Wang C, Han S, Zhou Y, Zhang J, Zheng X, Dai G, Li MH (2016) Fine root growth and contribution to soil carbon in a mixed mature *Pinus koraiensis* forest. *Plant Soil* 400(1–2):275–284. <https://doi.org/10.1007/s11104-015-2724-x>

- West JB, Espeleta JF, Donovan LA (2004) Fine root production and turnover across a complex edaphic gradient of a *Pinus palustris*–*Aristida stricta* savanna ecosystem. *For Ecol Manag* 189(1–3):397–406. <https://doi.org/10.1016/j.foreco.2003.09.009>
- Xiong D, Yang Z, Chen G, Liu X, Lin W, Huang J, Bowles FP, Lin C, Xie J, Li Y, Yang Y (2018) Interactive effects of warming and nitrogen addition on fine root dynamics of a young subtropical plantation. *Soil Biol Biochem* 123:180–189. <https://doi.org/10.1016/j.soilbio.2018.05.009>
- Yan G, Chen F, Zhang X, Wang J, Han S, Xing Y, Wang Q (2017) Spatial and temporal effects of nitrogen addition on root morphology and growth in a boreal forest. *Geoderma* 303:178–187. <https://doi.org/10.1016/j.geoderma.2017.05.030>
- Yan G, Han S, Zhou M, Sun W, Huang B, Wang H, Xing Y, Wang Q (2020) Variations in the natural ^{13}C and ^{15}N abundance of plants and soils under long-term N addition and precipitation reduction: interpretation of C and N dynamics. *Forest Ecosyst* 7(1):1–13. <https://doi.org/10.1186/s40663-020-00257-w>
- Yanai R, Eissenstat D (2002) Root life span, efficiency, and turnover. *Plant Roots*. <https://doi.org/10.1201/9780203909423.ch13>
- Yin C, Xiao Q, Sun Y, Liu Q, Pang X (2017) *Picea asperata* pioneer and fibrous roots have different physiological mechanisms in response to soil freeze-thaw in spring. *Biol Plant* 61(4):709–716. <https://doi.org/10.1007/s10535-017-0728-5>
- Ying W, Ming-kui CAO, Bo T, Ke-rang LI (2006) The characteristics of spatio-temporal patterns in precipitation in China under the background of global climate change. *Geogr Res* 25(6):1031–1040. <https://doi.org/10.11821/yj2006060010>
- Yuan ZY, Chen HY (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(4):204–221. <https://doi.org/10.1080/07352689.2010.483579>
- Yuan ZY, Chen HY (2012) A global analysis of fine root production as affected by soil nitrogen and phosphorus. *Proc Royal Soc b: Biol Sci* 279(1743):3796–3802. <https://doi.org/10.1098/rspb.2012.0955>
- Zhang X, Xing Y, Wang Q, Yan G, Wang M, Liu G, Wang H, Huang B, Zhang J (2020) Effects of long-term nitrogen addition and decreased precipitation on the fine root morphology and anatomy of the main tree species in a temperate forest. *Forest Ecol Manag* 455:117664. <https://doi.org/10.1016/j.foreco.2019.117664>
- Zhou M, Yan G, Xing Y, Chen F, Zhang X, Wang J, Zhang J, Dai G, Zheng X, Sun W, Wang Q, Liu T (2019) Nitrogen deposition and decreased precipitation does not change total nitrogen uptake in a temperate forest. *Sci Total Environ* 651:32–41. <https://doi.org/10.1016/j.scitotenv.2018.09.166>
- Zang U, Goisser M, Häberle KH, Matyssek R, Matzner E, Borken W (2014) Effects of drought stress on photosynthesis, rhizosphere respiration, and fine-root characteristics of beech saplings: a rhizotron field study. *J Plant Nutr Soil Sci* 177(2):168–177. <https://doi.org/10.1002/jpln.201300196>

Publisher's Note Springer Nature remains neutral with regard to jurisdictional claims in published maps and institutional affiliations.