

## Effects of Soil Warming, Precipitation Exclusion, and Their Interaction on Fine Root Production of Chinese Fir Seedlings (Postprint)

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### Abstract

To reveal the belowground response and adaptation of Chinese fir (*Cunninghamia lanceolata*), the most important plantation tree species in China, to global warming and altered precipitation patterns, a two-factor experiment of soil warming and precipitation exclusion was conducted on Chinese fir seedlings at Chenda State-owned Forest Farm in Sanming City, Fujian Province. The experiment included four treatments: control (CK), soil warming at 5°C (W), 50% precipitation exclusion (P), and combined soil warming and precipitation exclusion (WP). The minirhizotron method was used to investigate the effects of soil warming, precipitation exclusion, and their interaction on fine root production of Chinese fir seedlings (characterized by fine root birth number) during the one-year experimental period. Two-way ANOVA revealed that soil warming and precipitation exclusion had no significant effect on total fine root birth number, but their interaction was highly significant. Compared with CK, total fine root birth number in the W treatment increased significantly, while that in the WP treatment was significantly lower than in both the W and P treatments. Repeated measures ANOVA of soil warming, precipitation exclusion, and season revealed that soil warming  $\times$  season and precipitation exclusion  $\times$  season both had significant effects on fine root birth number. Compared with CK, fine root birth number in the W treatment increased significantly in spring, that in the P treatment increased significantly in autumn, while that in the WP treatment decreased significantly in summer and winter. Three-way ANOVA of soil warming, precipitation exclusion, and diameter class indicated a significant effect of soil warming  $\times$  precipitation exclusion  $\times$  diameter class. Fine root birth number in the 0-1 mm diameter class in the W treatment was significantly higher than in CK, but that in the WP treatment was significantly lower than in both the W and P treatments. Three-way ANOVA of soil warming, precipitation exclusion,

and soil layer showed no significant interaction among soil warming, precipitation exclusion, and soil layer. Only in the 20-40 cm soil layer was fine root birth number in the P treatment significantly higher than in CK. The results indicate that soil warming and precipitation exclusion have significant interactive effects on fine root production of Chinese fir seedlings, and such interactions also vary with different seasons and diameter classes.

## Full Text

## Preamble

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**Title:** Effects of Soil Warming and Precipitation Exclusion and Their Interaction on Fine Root Production of Chinese Fir (*Cunninghamia lanceolata*) Seedlings

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## Abstract

Global warming is expected to profoundly affect precipitation patterns. Root production plays a key role in ecosystem carbon, nutrient, and water cycling; however, the response of root production to soil warming and precipitation exclusion is not well understood. This study aimed to explore the belowground responses and adaptability of Chinese fir (*Cunninghamia lanceolata*), the most important timber species in southern China, to global warming and precipitation

changes. A factorial experiment of soil warming (ambient, +5°C) and precipitation exclusion (ambient, 50%) was conducted at the Chenda State-owned Forest Farm in Sanming, Fujian Province. We measured changes in fine root production (fine root birth) after one year of soil warming and precipitation exclusion using the minirhizotron method.

Two-way analysis of variance (ANOVA) showed that soil warming and precipitation exclusion had no significant effect on the total number of fine roots produced, whereas their interaction had a marked effect. Compared to the control plots, the total number of fine roots produced increased significantly in the warmed soil plots, but decreased significantly in the warmed soil plus precipitation exclusion plots compared to the warmed plots and the precipitation exclusion plots, respectively. Repeated-measures ANOVA including soil warming, precipitation exclusion, and season showed that the interaction of soil warming and season, and the interaction of precipitation exclusion and season had significant effects on the number of fine roots produced. Compared to the control plots, the number of fine roots produced increased significantly in the warmed soil plots in the spring, and in the precipitation exclusion plots in the autumn, but decreased significantly in the warmed soil plus precipitation exclusion plots in the summer and winter.

A three-way ANOVA including soil warming, precipitation exclusion, and diameter class showed that the interaction of soil warming, precipitation exclusion, and diameter class had a significant effect on the number of fine roots produced, which was significantly higher in the soil warming treatment than in the control at 0–1 mm. However, the number of fine roots produced was significantly lower in the soil warming plus precipitation exclusion treatment than in the precipitation exclusion treatment and soil warming treatment, respectively. These results indicate that the effect of the interaction of soil warming and precipitation exclusion on the number of fine roots produced mainly occurred at 0–1 mm. The three-way ANOVA including soil warming, precipitation exclusion, and soil layer showed that the three-way interaction had no effect on the number of fine roots produced. Only the precipitation exclusion treatment resulted in a significantly higher number of fine roots than the control in the soil layer of 20–40 cm. This suggests that the interaction of soil warming and precipitation exclusion on the number of fine roots produced had similar effects at different soil layers.

It is concluded that the interaction of soil warming and precipitation exclusion influenced root production through changing the fine root seasonal distribution and diameter class allocation, which may play important roles in the growth of Chinese fir.

**Keywords:** soil warming; precipitation exclusion; fine root production; fine root birth

## 1. Study Area Overview

The study was conducted at the Chenda observation site of the Sanming Forest Ecosystem and Global Change Research Station, located in the Jinsiwan Forest Park Chenda State-owned Forest Farm in Sanming City, Fujian Province (26°19 N, 117°36 E). The region has a mid-subtropical monsoon climate with an elevation of 300 m, mean annual temperature of 19.1°C, mean annual precipitation of 1749 mm, and mean annual evaporation of 1585 mm.

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## 2. Experimental Design

The experiment employed a completely randomized design with two factors: soil warming and precipitation exclusion. Four treatments were established: (1) CK (control, no warming and no precipitation exclusion); (2) W (soil warming of +5°C, no precipitation exclusion); (3) P (precipitation exclusion of 50%, no warming); and (4) WP (soil warming of +5°C plus precipitation exclusion of 50%). Each plot measured 2 m × 2 m, with 2 m × 2 m buffer zones around each plot to prevent interference between adjacent plots.

Soil was collected from a nearby Chinese fir forest. After removing stones and other debris, the soil was mixed by layer and backfilled using a compaction method to approximate the original bulk density. The soil layers were 0–10 cm, 10–20 cm, and 20–70 cm. All plots were equipped with identical heating cables installed at 10–20 cm and 20–70 cm depths in a grid pattern to ensure uniform warming. The cables were spaced 20 cm apart, with seedlings positioned between cable lines.

In March 2013, ten half-sib Chinese fir seedlings were uniformly planted in each plot. The mean seedling height was (25.7 ± 2.52) cm and mean basal diameter was (3.35 ± 0.48) cm. Two minirhizotubes were installed at a 45° angle directly beneath two seedlings per plot to monitor fine root growth and mortality dynamics. The tubes were 90 cm long with a diameter of 5 cm.

For precipitation exclusion, transparent panels were installed 5 cm above the ground surface, covering 50% of each plot area. Panels were arranged at 0.05 m intervals across the plot. Power was supplied to begin soil warming in January 2014. Total precipitation during the experimental period was 1994.2 mm.

shows the soil bulk density before and after backfilling at different depths.

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## 3. Root Image Collection and Data Acquisition

A Bartz minirhizotron imaging system was used for continuous observation of fine roots over a one-year period from January 2014 to January 2015. Images were collected in the first and last ten days of each month. After collection,

images were processed in the laboratory using Rootfly image analysis software to obtain data on fine root length, number of fine roots per image, and other parameters. Live roots were identified as white or brown roots, while dead roots were defined as those with cortex shedding or wrinkled epidermis. Fine root production was quantified using the number of fine root births per image.

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## 4. Data Processing and Analysis

The observation period was divided into seasons based on subtropical climate conditions: spring (March, April, May), summer (June, July, August), autumn (September, October, November), and winter (December, January, February). Two-way ANOVA was used to test the effects of soil warming and precipitation exclusion on the annual number of fine root births per tube. Repeated-measures ANOVA was employed to examine the effects of soil warming, precipitation exclusion, and season on fine root births. Three-way ANOVA was used to test the effects of soil warming, precipitation exclusion, and diameter class or soil layer on fine root births. One-way ANOVA and LSD tests were used for pairwise comparisons between treatments. All statistical analyses were performed using SPSS 19.0 software with a significance level of  $P = 0.05$ .

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## 2. Results

### 2.1 Soil Temperature and Moisture

Soil warming had a highly significant effect on soil temperature but no effect on soil moisture, while precipitation exclusion had a highly significant effect on soil moisture but no effect on temperature. The interaction between warming and precipitation exclusion had no significant effect on either temperature or moisture.

During the one-year experimental period, the mean soil temperature and moisture content (0-10 cm) were: - CK:  $(21.56 \pm 6.74)^{\circ}\text{C}$ ,  $(23.33 \pm 3.23)\%$  - W:  $(26.77 \pm 6.75)^{\circ}\text{C}$ ,  $(18.96 \pm 3.54)\%$  - P:  $(22.35 \pm 6.10)^{\circ}\text{C}$ ,  $(20.02 \pm 4.09)\%$  - WP:  $(26.93 \pm 6.01)^{\circ}\text{C}$ ,  $(16.05 \pm 3.85)\%$

Soil temperature in W and WP treatments was significantly higher than CK ( $P < 0.05$ ), while there was no significant difference between P and CK ( $P > 0.05$ ). Soil moisture in WP treatment was significantly lower than W ( $P < 0.05$ ).

[Figure 1: see original paper] shows the annual changes in soil temperature and moisture under different treatments.

presents the two-way ANOVA results for the effects of soil warming, precipitation exclusion, and their interaction on soil temperature, moisture, and total fine root production.

## 2.2 Total Fine Root Production

Soil warming and precipitation exclusion alone had no significant effect on total fine root production, but their interaction was highly significant ( $P < 0.01$ ). There was no significant difference in total fine root production between CK and P treatments ( $P > 0.05$ ). Under non-excluded precipitation conditions, W treatment significantly increased total fine root production ( $P < 0.05$ ). However, under precipitation exclusion conditions, WP treatment significantly decreased total fine root production compared to W and P treatments ( $P < 0.05$ ).

[Figure 2: see original paper] shows the total number of fine roots produced per tube over one year under different treatments.

## 2.3 Seasonal Dynamics of Fine Root Production

The interaction between soil warming and season, and between precipitation exclusion and season, had highly significant effects on fine root production ( $P < 0.01$ ), while the three-way interaction among soil warming, precipitation exclusion, and season had no significant effect ( $P > 0.05$ ). In spring, W treatment had significantly higher fine root production than CK ( $P < 0.05$ ), while other treatments showed no significant differences. In autumn, P treatment had significantly higher production than other treatments ( $P < 0.05$ ). In summer and winter, WP treatment had significantly lower production than CK ( $P < 0.05$ ), while other treatments showed no significant differences. Season had a significant effect on fine root production across all treatments ( $P < 0.05$ ), with summer production being significantly higher than other seasons in CK, W, and P treatments ( $P < 0.05$ ).

shows the P-values from repeated-measures ANOVA examining the effects of soil warming, precipitation exclusion, and season on fine root production.

## 2.4 Diameter Class Allocation and Vertical Distribution of Fine Root Production

The interaction among soil warming, precipitation exclusion, and diameter class had a highly significant effect on fine root production ( $P < 0.01$ ). The interaction between soil warming and diameter class was significant ( $P < 0.05$ ), while the interaction between precipitation exclusion and diameter class was not significant ( $P > 0.05$ ). For 0-1 mm diameter roots, W treatment had significantly higher production than CK ( $P < 0.05$ ), while WP treatment had significantly lower production than both W and P treatments ( $P < 0.05$ ). There were no significant differences among treatments for 1-2 mm diameter roots. The 0-1 mm diameter class accounted for the vast majority of total fine root production: 94% in CK, 96% in W, 95% in P, and 97% in WP.

The interactions between soil warming and soil layer, precipitation exclusion and soil layer, and the three-way interaction among all factors had no significant effects on fine root production ( $P > 0.05$ ). Only precipitation exclusion

treatment showed significantly higher fine root production in the 20–40 cm layer compared to CK ( $P < 0.05$ ), while other treatments showed no significant differences between layers.

[Figure 3: see original paper] shows the number of fine roots produced per tube across different seasons, diameter classes, and soil layers.

and present the P-values from three-way ANOVA examining the effects of soil warming, precipitation exclusion, and diameter class or soil layer on fine root production.

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### 3. Discussion

#### 3.1 Total Fine Root Production

This study found that soil warming significantly increased total fine root production. This is consistent with many previous studies [16–19]. The primary mechanism is that soil warming promotes soil nitrogen mineralization [21], increasing nitrogen availability and thus benefiting fine root production. Precipitation exclusion alone tended to increase fine root production, though not significantly. This may be because the exclusion period was only one year, which may be too short to show significant effects. Additionally, the study region has abundant precipitation, so the drought intensity may not have been severe enough to significantly affect production. However, according to optimal allocation theory, when water becomes a limiting factor for plant growth, plants allocate more carbohydrates and nutrients to fine root growth relative to aboveground parts [22]. Related studies have shown that reduced precipitation promotes fine root production. For example, Gaul et al. [23] found increased fine root production during a 14-week precipitation exclusion experiment in Norway spruce.

This study also showed that WP treatment significantly reduced total fine root production compared to W and P treatments. This occurs because the combination of warming and precipitation exclusion exacerbates soil drought, thereby reducing fine root production [24]. The combined stress may also affect photosynthesis, reducing carbon supply for root growth.

#### 3.2 Seasonal Dynamics of Fine Root Production

The study found that fine root production in Chinese fir seedlings peaked in summer and spring, consistent with previous research [25, 26]. Hendrick et al. [25] used minirhizotron technology to study northern hardwood forests, while Cheng et al. [26] studied naturally regenerated young forests in Sanming, Fujian, both finding peak fine root production in late spring and early summer. This seasonal pattern may be attributed to several factors: in early spring, rapid soil temperature recovery, increased precipitation, higher soil moisture, and carbohydrate reserves from the previous year all favor extensive root growth [27]. In summer,

trees fix relatively more photosynthates, allocating substantial amounts belowground to meet the carbon demands of fine root growth [28]. The decline in autumn production is associated with reduced precipitation, lower temperatures, and decreased carbohydrate allocation to roots [25].

In this study, W treatment significantly increased spring fine root production, likely due to higher soil temperatures, enhanced nutrient mineralization, and adequate spring soil moisture. P treatment significantly increased autumn production, possibly because reduced soil moisture triggered increased fine root production to acquire water. WP treatment significantly decreased summer and winter production, primarily due to excessively high soil temperatures and low moisture in summer, and extremely low soil moisture in winter that reduced photosynthesis and carbon allocation belowground while increasing root respiration.

### 3.3 Diameter Class Allocation and Vertical Distribution

The results show that the interaction between soil warming, precipitation exclusion, and diameter class significantly affected fine root production, indicating that the interaction effects mainly occurred in the 0–1 mm diameter class. Fine roots of 0–1 mm are the primary organs for water and nutrient absorption and are most sensitive to changes in soil environmental factors and photosynthate availability. In contrast, 1–2 mm roots serve mainly transport functions, are more lignified, have lower maintenance costs, and are less sensitive to environmental changes. The 0–1 mm diameter class contributed the most to fine root production, accounting for 94–97% of total production across treatments, consistent with previous studies [26, 29, 30].

The study also found no significant interaction effects involving soil layer, indicating that the effects of soil warming, precipitation exclusion, and their interaction on fine root production were similar across soil layers. However, P treatment showed significantly higher production in the 20–40 cm layer than in the 0–20 cm layer, likely because surface soil dries faster due to direct exposure to air and limited water replenishment under precipitation exclusion, causing roots to migrate deeper to access moisture.

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## 4. Conclusion

This study demonstrates that while soil warming and precipitation exclusion alone did not significantly affect fine root production, their interaction had highly significant effects. W treatment significantly increased total fine root production, while WP treatment significantly decreased it compared to W and P treatments. Season significantly affected fine root production, with the interaction altering the seasonal distribution of production. The interaction effects primarily occurred in the 0–1 mm diameter class, with no significant depth-dependent effects. These findings indicate that fine root production responses

to the interaction of soil warming and precipitation exclusion operate mainly through changes in seasonal timing and diameter class allocation, which may have important implications for Chinese fir seedling growth under global change scenarios.

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