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Fine-root chemical traits rather than morphological traits of Chinese fir (*Cunninghamia lanceolata* (Lamb.) Hook.) plantations vary along an altitudinal gradient in Eastern China

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Abstract

Aim of study: To explore the different patterns of fine-root traits by elucidating changes in their chemistries and morphologies in Chinese fir plantations along an altitudinal gradient.

Area of study: National Mazongling Nature Reserve (Anhui Province).

Materials and methods: Soil and fine roots (≤ 2 mm) samples were extracted from three soil layers (0-10 cm, 10-20 cm, and 20-30 cm) at four altitudes (750 m, 850 m, 1000 m, and 1150 m), after which their nutrient concentrations and morphological traits, respectively, were quantified. We employed mixed model ANOVA to test the effects of altitude, soil layer, and their interactions on the characteristics of soil and fine roots. The relationships between the functional traits of fine roots and climate, soil and stand structures were evaluated by the standard major axis regression and the structural equation model.

Main results: The chemical traits of fine roots were higher at medium altitudes (which decreased significantly with the soil layer). In contrast the morphological traits of fine roots did not change significantly. In chemical traits, both altitude, organic matter components, and soil total phosphorus (TP) exerted dominant effects on fine-root N, and both altitude and soil TP exerted dominant effects on fine-root P. However, in morphological traits, we found that altitude and soil C:N were crucial impact factors.

Research highlights: Fine roots might preferentially adjust their chemical traits rather than morphological traits to facilitate higher root efficiencies in response to variable environmental conditions.

Additional key words: elevational gradient; planted forests; fine-root nutrient concentrations; fine-root morphology; soil nutrients

Abbreviations used: BD (bulk density); LES (leaf economics spectrum); MAP (mean annual precipitation); MAT (mean annual air temperature); OMC (organic matter component); RTD (root tissue density); SEM (structural equation modeling); SMA (standard major axis); SOC (soil organic carbon); SRA (specific root area); SRL (specific root length); SWC (soil water content); TN (total nitrogen); TP (total phosphorus).

Authors' contributions: Conceived and designed the experiments: X Xu and W Fan. Performed the experiments: W Fan, P Deng, A Li, and S Zhang. Analyzed the data: W Fan, J Wang, and H Wang. Drafting of the manuscript: W Fan. Critical revision of manuscript for important intellectual content: X Xu.

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Introduction

Fine roots comprise the primary belowground tissues of plants, which acquire water and nutrients from the soil, and serve as the most active components of plant matter and energy transport (Bardgett *et al.,* 2014; McCormack *et al.,* 2015). Plants adopt various allocation strategies to regulate the traits of fine roots in contrasting environments, with the aim of minimizing the resource investments required to ensure the absorption of water

and nutrients. However, current knowledge relating to fine-root plasticity remains insufficient in contrast to that of leaves (Reich, 2014). Previous studies observed the plastic response of leaves to the availability of resources (Kramer-Walter *et al.,* 2016) and subsequently developed a global leaf economics spectrum (LES) (Tjoelker *et al.,* 2005; Chen *et al.,* 2013; Weemstra *et al.,* 2016). Analogous to leaves, researchers are also beginning to develop a root economics spectrum (RES). The elucidation of fine-root plasticity remains lacking in most ecosystems, as little is known about the relationships between the functional traits of fine roots and environmental factors, particularly for mature trees (Valverde-Barrantes *et al.,* 2007; Hertel *et al.,* 2013). This, a comprehensive understanding of the functional traits of fine roots is critical toward gaining insights into belowground resource acquisition strategies, and how plants adapt to variable environmental conditions.

Fine roots exhibit broad variations in their chemical traits. Previous studies have suggested that the chemical attributes of fine roots can be modified by changing environments (Eissenstat *et al.,* 2000; Hodge, 2004; Bardgett *et al.,* 2014; Liu *et al.,* 2015; Kramer-Walter *et al.,* 2016). Holdaway *et al.* (2011) found that the chemical characteristics of fine roots decreased with lower soil fertility. In a control experiment, it was observed that +N or +P treatments had positive impacts on fine-root N concentrations in contrast to an unfertilized control (Liu *et al.,* 2015). To the best of our knowledge, the chemical traits of roots are intimately correlated with their nutrient uptake. From the cost-benefit perspective, adjustments in the root nutrient uptake rate can avoid alterations of root morphologies, which involves greater resource expenditures (Mou *et al.,* 2013).

The order in which fine-root chemical and morphological traits respond to complex and changeable soil environments remains unclear. According to the cost-benefit perception, the chemical traits of fine roots may initially respond to soil nutrient conditions rather than the morphological traits (Mou *et al.,* 2013). Several investigations observed that this order might depend on the specific ion requirements of plants (Hodge, 2004). Typically, it is more important to enhance the sequestration of immobile ions (*e.g.,* phosphate) via fine-root morphological plasticity, as uptake is limited by root diffusion in the soil but not by uptake at the root surface. In contrast, mobile ions (*e.g.,* NO_3^-) diffuse to the root surface more easily; thus, the improvement of ion uptake at the root surface will greatly increase the capture of NO_3^-.

Several studies revealed that the morphological traits of fine roots did not respond to environmental changes (Leuschner *et al.,* 2004; James *et al.,* 2008; Liu *et al.,* 2015). For example, the morphological traits specific root length (SRL) and root tissue density (RTD) of fine roots did not differ significantly between the clay and sandy soils of spruce (*Picea abies* (L.) Karst.) and beech (*Fagus sylvatica* L.) forests (Weemstra *et al.,* 2017). The morphologies of fine roots in some species likely possess a high phylogenetic conservatism (Kong *et al.,* 2014); thus, they have a very limited capacity to respond to environmental changes. However, according to the resource economics theory (Grime, 1977), the morphological traits of fine roots can reflect the soil nutrient status of plant habitats. The fine-roots of plants in poor soils can survive longer with a higher RTD, as well as a lower SRL and specific root area (SRA) (Pérez-Ramos *et al.,* 2012).

Studies into the variable responses of fine-root traits to changing soil nutrients have been focused on manipulated plots (*e.g.,* fertilization). Uncertainties still remain concerning the responses of fine-root traits to naturally occurring heterogeneous supplies of nutrients in the soil. Altitudinal gradients are considered to be a valuable system for testing the responses of plants to environmental changes (Sundqvist *et al.,* 2013). This is because abiotic factors such as soil fertility, temperature, and precipitation often change along altitudinal gradients (Weemstra *et al.,* 2020). In terms of soil fertility, the general pattern is that concentrations and the supply rates of available mineral nutrients for plants in the soil show a decline with increasing altitude (Sveinbjörnsson *et al.,* 1995; Johnson *et al.,* 2000). However, there are some exceptional cases where soil fertility is not reduced with increasing altitude (Frangi *et al.,* 2005; Averill & Finzi, 2011). For instance, a variety of nutrients such as N, K, Mg, and Ca were generally lowest at mid-altitudes along an altitudinal gradient in New Mexico, USA (Gosz, 1980). Simultaneously, the heterogeneity of soil nutrients also exists between different soil layers, as nutrient concentrations are generally higher in the topsoil due to litter inputs, and their subsequent decomposition under natural conditions (Leuschner *et al.,* 2001; Hodge, 2004).

Mazongling Nature Reserve has rich vegetation with good conservation status and is located in the Tianma National Nature Reserve, in Anhui Province, China. Chinese fir (*Cunninghamia lanceolata* (Lamb.) Hook.) is one of the dominant species in the Mazongling Nature Reserve and occupies a prominent position in China's plantation resources with high quality, fast growth, and high yield. The increased altitude has altered the environmental conditions in this area, including the soil characteristics. Fine roots are very sensitive components of belowground root systems, where variations in their functional traits can reflect different strategies of trees for the uptake of nutrients. In the present study, Chinese fir plantations (under non-manipulated management) were employed as research subjects. We explored the variations between several chemical and morphological traits of Chinese fir fine-roots along an altitudinal gradient, and their relationships with soil characteristics.

We hypothesized that (1) the N and P concentrations of the fine roots would decrease with increasing altitude, and (2) the morphological traits of fine roots would be constant along an altitudinal gradient.

Material and methods

Description of study area

The study area is located in the Mazongling Nature Reserve (extending from 31°10′ to 31°20′ N, 115°31′ to 115°50′ E) of Southwest Jinzhai County, in Anhui Province, China, which was established in 1982. The topography of this region is characterized by mountainous and valleyed landscapes with altitudes ranging from 600 m to 1671 m, and the main slopes ranging from 30° to 40°. The soil type of this region is Cambisols (FAO, 2014) with a loamy texture, which was derived from granite. This area has a subtropical humid monsoon climate. The mean annual precipitation is 1510 mm, with the most rainfall occurring in July (227 mm) and the least in October (30 mm). The annual mean temperature ranges from 13 °C to 15 °C, and the mean annual frost-free period is 210 days. The Mazongling Nature Reserve includes original secondary and plantation forests, with a forest coverage of 95.9%. The dominant tree species in this area include *C. lanceolata, Pinus taiwanensis* Hayata, *Quercus* variabilis Blume, *Carya cathayensis* Sarg., and *Quercus glandulifera* Bl. var. *brevipetiolata* Nakai.

Experimental design

Chinese fir plantations were selected in this study. They were established by seedling afforestation on the logging sites of the natural secondary forest dominated by *P. taiwanensis* and *Quercus* spp. in the early 1980s. An intense thinning was carried out in the mid-1990s due to their high density, and thereafter no management measures were applied to these plantations. In view of the availability of sample sites in the region, four altitude classes (~750 m, 850 m, 1000 m, and 1150 m) (determined by GPS), with similar stand structure, were sampled in July 2018. Each of the four classes was repeated in triplicate (Fig. S1 [suppl]). There were 12 sampling sites in total. The distance between any two sample sites was greater than 100 m, such that their spatial autocorrelation was minimized. For each site, a plot of 20 m × 20 m was randomly set up, which were at least 100 m from the stand edges. The characteristics for the four altitudes are presented in Table 1.

Table 1. Characteristics of study sites (average values) along the altitudinal gradient of *Cunninghamia lanceolata*.

Characteristics	Altitude (m)			
	750	850	1000	1150
MAT (°C)	13.21	12.22	12.13	11.80
MAP (mm)	1484	1496	1574	1596
Density (trees/ha)	1051	2052	1537	2104
Average DBH (cm)	19.62	17.43	17.30	17.75
Average tree height (m)	13.23	12.35	12.01	12.30
Basal area (m^2/ha)	31.98	48.89	36.49	53.46

MAT, mean annual air temperature (data from Zhang *et al.,* 2020). MAP, mean annual precipitation (obtained from the WorldClim-Global Climate Data; Fick & Hijmans, 2017). DBH, diameter at breast height

Fine root and soil sampling

The fine roots were collected via the soil coring method (Vogt & Persson, 1991) and used for measuring the chemical and morphological traits. For each plot, a total of nine random soil cores (5.5 cm diameter) was collected in mid-July 2018. After clearing the litter layer, the soil cores were sampled from the soil surface to a depth of 30 cm using a root auger, after which each core was segmented into three layers (10 cm per soil layer). Thus, nine soil cores were stratified and mixed into three composite fine-root samples per plot. Finally, 36 fine-root samples (12 plots × 3 layers) were obtained for laboratory analysis. The fine-root samples were transported from the field to the laboratory in an icebox. In the laboratory, the core samples were soaked in low temperature deionized water for 24 h, then poured into trays, and gently rubbed to isolate the live fine roots from any impurities and soil. After measuring the morphological traits of the fine roots, they were dried at 65°C to determine their dry mass. Finally, they were pulverized using a grinding machine and sieved through a 0.25-mm sieve for chemical analysis.

Following the fine root sampling, five replicated soil sample subplots were randomly selected in each plot. Following the removal of the litter layers, soil samples were extracted from three soil layers (10 cm per layer) at each sampling subplot using a stainless-steel soil drill (3 cm inner diameter). For each plot, the soil samples from five sampling subplots were stratified and fully homogenized to form three composite soil samples. Finally, 36 soil samples (4 altitudes × 3 replicate plots × 3 layers) were collected. All of the samples were sieved through a 2-mm mesh to separate out large stones, roots, and other impurities.

Fine root and soil physicochemical measurements

All fine roots were scanned using a Microtek digital scanner (MRS-600A3LED). From these images, the WinRhizo root analysis system was used to quantify the length, surface area, and volume (approximated as cylindrical roots) of the fine roots in each sample. Further, the fine-root morphological traits were calculated using the following equations:

Root tissue density (mg/cm^3): RTD = dry fine-root mass/fine-root volume (1)

Specific root area (cm^2/g): SRA = fine-root surface area/dry fine-root mass (2)

Specific root length (m/g): SRL = fine-root length/dry fine-root mass (3)

The N concentrations of the fine roots were determined via an elemental analyzer (EA3000, Vector, Italy), whereas the P concentrations were measured using a flow injection auto-analyzer (FIAStar 5000, FOSS, Sweden).

The soil bulk density (BD) was calculated by weighing oven-dried samples of a known volume by using a cutting ring (inner volume 100 cm^3). The soil water content (SWC) was measured by oven-drying ~20 g of fresh soil at 105°C for 48 h. The soil pH was determined in a 1:2.5 (w/v) soil:water suspension using a pH meter. The remaining soil samples were ground into a powder and sieved through a 0.25-mm sieve after air drying and used for chemical analysis. The soil organic carbon (SOC) and total N (TN) were measured using an elemental analyzer (EA3000, Vector, Italy), whereas the soil total P (TP) was calculated via a flow injection auto-analyzer (FIAStar 5000, FOSS, Sweden). Further, the K, Ca, and Mg concentrations were measured using an atomic absorption spectrophotometer (TAS-990, Bio-Equip, Beijing, China). All physicochemical measurements were made at the Laboratory of Silviculture & Forest Ecology at Anhui Agricultural University in Heifei, China.

Statistical analyses

For this study, data analyses were implemented using R software (v 3.5.1), and the data were checked for normality via the Shapiro-Wilk test prior to analysis. Mixed model ANOVA was employed to test the altitude, soil layer effects, and their interactions on soil characteristics and fine-root traits, and plotted as random terms. The levels of statistical significance were set at $\alpha = 0.05$. Pearson correlation tests were used to examine the associations between fine-root traits, and the analysis was performed using the 'corrplot' package. To determine the relationships between the fine-root traits (SRL, SRA, RTD, fine-root N, and P concentrations) and soil characteristics, the standard major axis (SMA) regression was analyzed by using the 'SMATR' package. The fine-root traits were not correlated with the structural factors of the stands and mean annual air temperature (MAT) via SMA regression, except for the mean annual precipitation (MAP) (Figs. S2 & S3 [suppl]).

We developed structural equation modeling (SEM) to evaluate the direct and indirect effects of environmental factors on the fine-root traits (chemistry and morphology) along an altitudinal gradient. Only the stand structures, climate, and soil variables that were significantly correlated ($p < 0.05$) with the fine-root traits were included in SEM. Considering the strong correlation between the SOC and soil TN, we performed principal component analysis (PCA) using the predictor significantly associated with the fine-root N to create a new index (organic matter component [OMC]). The goodness of fit of the SEM was evaluated using the Akaike information criterion (AIC), Bayesian information criterion (BIC), Fisher' C statistic and the whole-model *p* value (Lefcheck, 2016). The SEM was conducted using the R packages piecewiseSEM (Lefcheck, 2016) and nlme (R Core Team, 2021).

Results

Variations in soil characteristics along an altitudinal gradient

Soil characteristics differed between the four altitudes of the sampling sites. The SWC, SOC, and TN initially decreased and then increased along the altitudinal gradient, with the minimums appearing at 850 m (Fig. 1). The SWC varied significantly between different altitudes ($p < 0.05$) (Table 2). The BD, pH, TP, K, Ca, and Mg were higher at medium altitudes and differed considerably between altitudes ($p < 0.05$) except for BD and K (Table 2, Fig. 1). The SWC, SOC, TN, and TP concentrations decreased significantly across the three soil layers ($p < 0.001$) (Table 2, Fig. 1). As for the soil stoichiometric ratios, the soil C:N tended to be higher at 850 m and showed no distinct trend across the entire 0-30 cm soil sample (Fig. 1). The soil C:P and N:P values were marginally significantly lower at 850 m (Fig. 1) but differed significantly between soil layers ($p < 0.001$) (Table 2).

Relationships among fine-root traits

The correlation analysis of fine-root traits (summed over all the fine root data) revealed that the chemical traits of fine roots were not correlated with their morphological traits ($p > 0.05$) (Table 3), while exhibited a significantly positive correlation between N and P ($p < 0.001$) (Table 3). In terms of the fine-root morphological traits, the

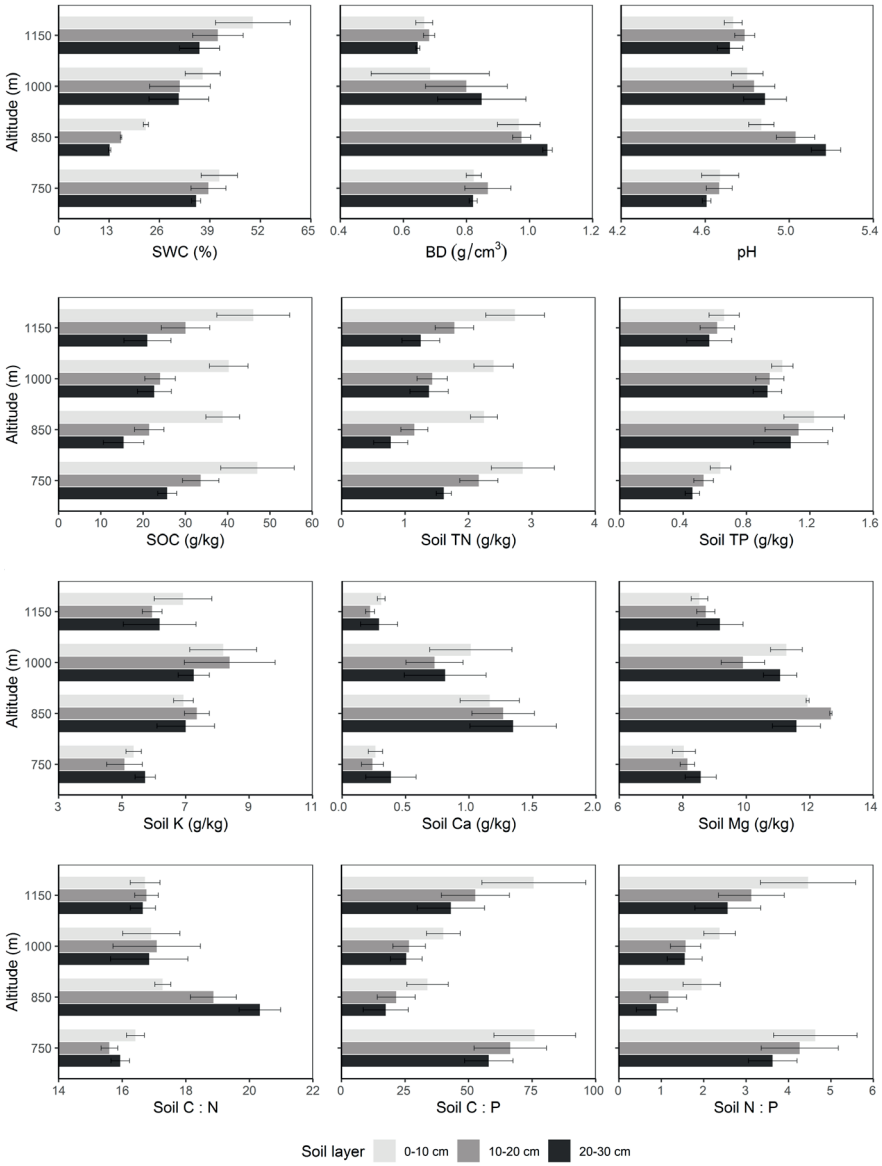

Figure 1. Effects of altitude on soil characteristics. Values are means ± SE (n=3) for individual sample layers. SWC, soil water content. BD, bulk density. SOC, soil organic carbon. TN, total nitrogen. TP, total phosphorus

RTD was negatively correlated to both SRL and SRA ($p < 0.001$), which were positively correlated to each other ($p < 0.001$) (Table 3).

Effects of altitudinal gradient on chemical traits of fine roots Effects of altitudinal gradient on chemical traits of fine roots

As shown in Table 2, there were marginal differences ($p = 0.051$) in N concentration of the fine roots between four altitudes, and there were significant differences ($p = 0.007$) in P concentration. Both N and P concentrations of fine roots were higher at medium altitudes than at low and high altitudes (Fig. 2). The fine-root N and P concentrations decreased significantly from 0-10 cm to 10-20 cm to 20-30 cm (Table 2, Fig. 2). SMA regression analysis showed that the fine-root N was significantly positively correlated with the SOC and soil TN ($p < 0.05$), whereas the fine-root P was significantly positively related to the SOC, TP, Ca, and Mg (Fig. 3). SEM analysis illustrated that fine-root N was dominantly and directly affected by altitude, OMC and soil TP, with a standardized direct effect of -0.32, 0.60, and 0.43, respectively (Fig. 4). SEM analysis also illustrated that fine-root P was dominantly and directly affected by altitude and soil TP, with a standardized direct effect of -0.34 and 0.38, respectively (Fig. 4). In addition, altitude indirectly affected chemical traits of fine roots by regulating MAP, SWC, OMC, and soil TP (Fig. 4). The model explained 53% and 31%, respectively, of the variance in the fine-root N and P (Fig. 4).

Table 2. Effects (p values) of altitude and soil layer (0-10 cm, 10-20 cm, and 20-30 cm) on fine-root traits and soil characteristics.

Characteristics	Altitude	Soil layer	Altitude × Soil layer
Df	3, 8	2, 16	6, 16
Fine-root			
N	0.051	**< 0.001**	0.942
P	**0.007**	**< 0.001**	**0.011**
SRL	0.430	0.063	0.820
SRA	0.398	0.618	0.762
RTD	0.214	0.094	0.260
Soil			
SWC	**0.034**	**< 0.001**	0.593
BD	0.084	0.094	0.192
pH	**0.022**	**0.008**	**< 0.001**
SOC	0.534	**< 0.001**	0.582
TN	0.316	**< 0.001**	0.282
TP	**0.032**	**< 0.001**	0.704
K	0.118	0.674	0.472
Ca	**0.025**	0.301	0.333
Mg	**< 0.001**	0.760	0.226
C:N	0.059	0.128	**0.008**
C:P	0.083	**< 0.001**	0.174
N:P	0.055	**< 0.001**	0.074

Df, degree of freedom. SRL, specific root length. SRA, specific root area. RTD, root tissue density. SWC, soil water content. BD, bulk density. SOC, soil organic carbon. TN, total nitrogen. TP, total phosphorus. Bold values denote statistical significance, $p < 0.05$.

Effects of altitudinal gradients on morphological traits of fine roots

Mixed model ANOVA analysis showed that there was no significant difference ($p > 0.05$) in the morphological traits (SRL, SRA, and RTD) of fine roots along an altitude gradient, and there was also no significant difference ($p > 0.05$) between three soil layers (Table 2, Fig. 2). SMA regression and SEM analysis collectively confirmed the important role of soil C:N in regulating SRL, SRA, and RTD (Figs. 4 & 5). And SEM analysis also illustrated that morphological traits was directed affected by altitude (Fig. 4). In addition, altitude indirectly affected morphological traits of fine roots by regulating MAP, SWC, and soil C:N (Fig. 4). The combination of environmental factors explained 33%, 27% and 30% of the variances in the SRL, SRA, and RTD.

Discussion

Variations in the chemical traits of fine roots across altitudinal gradients depend on soil nutrients

The study results illustrated the predominant role of soil nutrients (OMC and soil TP) in regulating chemical traits of fine roots (Fig. 4). Many studies also suppose that

Table 3. Pearson correlation coefficients between fine-root traits.

	Fine-root N	Fine-root P	SRL	SRA	RTD
Fine-root N	1				
Fine-root P	0.667***	1			
SRL	0.040	-0.195	1		
SRA	-0.232	-0.260	0.699***	1	
RTD	0.069	0.041	-0.579***	-0.549***	1

***, $p < 0.001$. SRL, specific root length. SRA, specific root area. RTD, root tissue density.

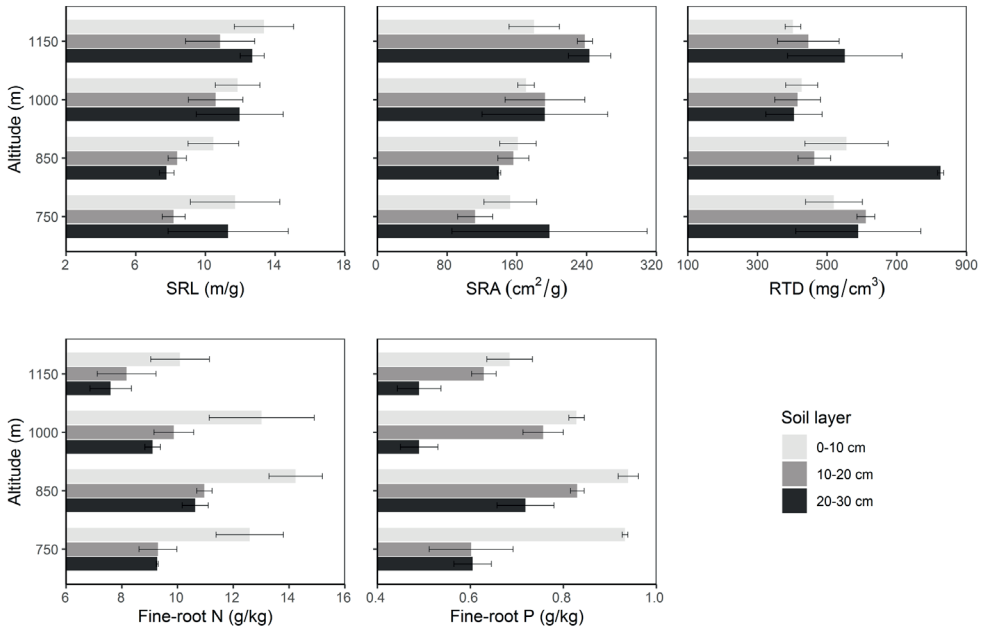

Figure 2. Effects of altitude on fine-root traits. Values are means ± SE (n = 3) for individual sample layers. SRL, specific root length. SRA, specific root area. RTD, root tissue density.

the chemical traits of fine roots respond to changes in soil nutrient conditions (Holdaway *et al.,* 2011; Kramer-Walter *et al.,* 2016; Kramer-Walter & Laughlin, 2017). For fine-root N, the OMC and soil TP served as direct predictors (Fig. 4). Higher OMC and soil TP could increase the N concentrations of fine roots by promoting fine roots absorption. Consistent with this result, in Norway spruce forests, fine-root N heavily increased from low N area (northern Europe) to high N area (central Europe) (Hodge, 2004; Borken *et al.,* 2007). For fine-root P, it had a significant positive relationship with the soil TP (Figs. 3 & 5), which aligned with the fine-root N.

Changes in the fine-root chemical traits of different soil layers accorded with this pattern in our study. Soil nutrients were observed to decrease with deeper soil layers (Pregitzer *et al.,* 1998), with our results confirming this view. For example, the SOC, TN, and TP among the three soil layers were as follows: 0-10 cm > 10-20 cm > 20-30 cm and had significant differences (Fig. 1, Table 2). The fine-root N and P were significantly reduced from 0-10 cm to 10-20 cm to 20-30 cm (Fig. 2, Table 2). This was consistent with the findings for European beech (Terzaghi *et al.,* 2013), which showed that the fine-root N concentration decreased significantly with the soil layer during the growing season.

Besides the detected importance of soil nutrients, the role of altitude in controlling chemical traits of fine roots should not be neglected. Our results demonstrated that altitude directly affected fine-root N and P, and indirectly affected them by regulating MAP, SWC, OMC, and soil TP (Fig. 4). Consistent with this result, we found that fine-root N showed marginal differences (p = 0.051) along an altitudinal gradient, and fine-root P showed significant differences (p = 0.007) (Table 2).

These results agreed with our first hypothesis that fine-root chemical traits varied between altitudes, which were associated with soil nutrients. Previous studies have suggested that the chemical traits of tree fine roots are related to soil nutrients (Holdaway *et al.,* 2011; Kramer-Walter *et al.,* 2016). Kramer-Walter & Laughlin (2017) found that fine-root nutrient concentrations responded with the highest plasticity to the availability of soil nutrients, where under high nutrient treatments, the nutrient concentrations of fine roots were always higher. The rapid response of fine-root chemical traits can increase root efficiencies (Mou *et al.,* 2013).

Morphological traits of fine roots do not differ along an altitudinal gradient

The study results illustrated that soil C:N played a dominant role in controlling the morphological traits of fine roots (Fig. 4). Soil C:N reflects the availability of soil nutrients. According to the resource economics theory (Grime, 1977), fine-root morphologies can shift with soil nutrient availability (Pérez-Ramos *et al.,* 2012). ANOVA results showed that altitudinal gradients and soil depth did not affect soil C:N (Table 3), so the morphological traits of fine roots did not change significantly.

The study results revealed that the altitude was another important predictor, and it also can regulate MAP, SWC, and soil C:N to indirectly affect morphological traits of fine roots (Fig. 4). A previous study reported that soil fertility and precipitation are major factors that influence the morphological traits of fine roots (Zhang *et al.,* 2019). But in our study, no differences were found in SRL, SRA, and

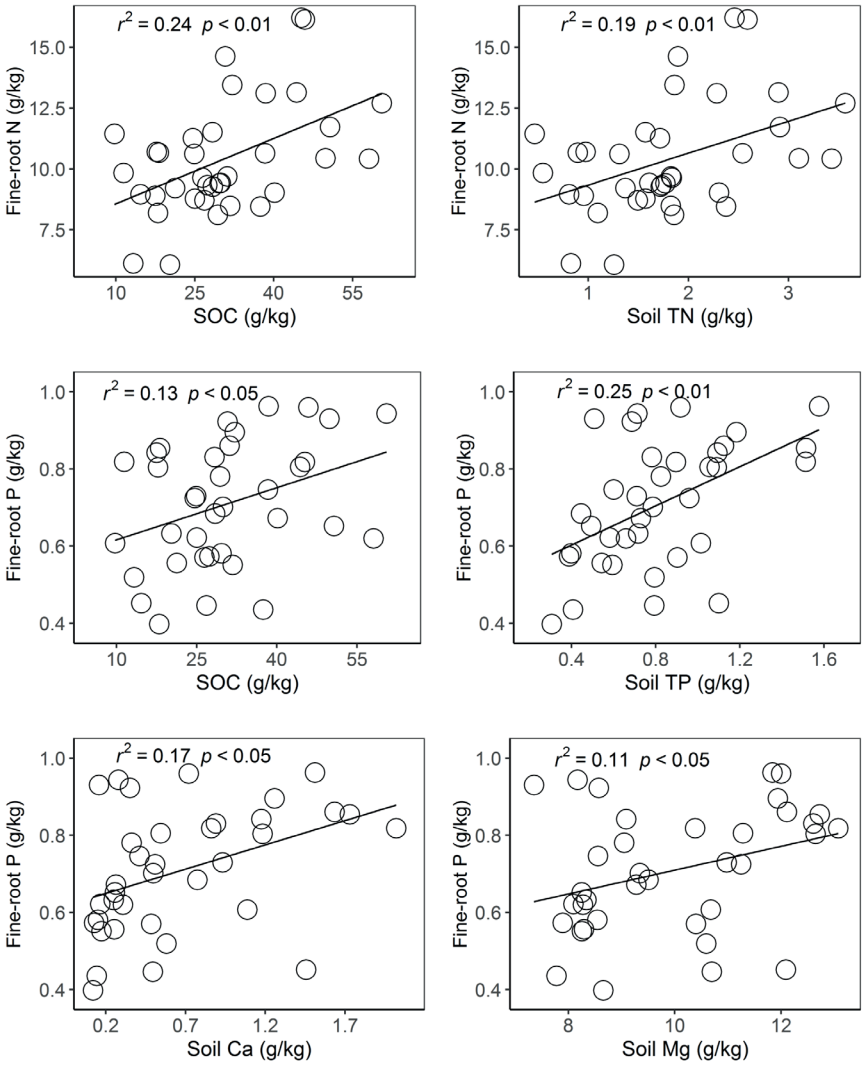

Figure 3. Bivariate relationships between fine-root chemical traits and soil characteristics. Solid lines indicate significant standard major axis (SMA) regression lines. Coefficients of determination *(r^2)* are shown for the SMA regressions using the soil characteristic data. SOC, soil organic carbon. TN, total nitrogen. TP, total phosphorus.

RTD along an altitudinal gradient (Table 3). There are probably some reasons for this. On the one hand, studies have found that conifers typically exhibit a conservative life strategy (Coomes *et al.,* 2005), they may exhibit lower phenotypic plasticity (Hodge, 2004; Grassein *et al.,* 2010). The target species under study for our research (*C. lanceolata*) is a common conifer species in the Southern China. On the other hand, differences in the plasticity of fine-root morphologies may occur in < 0.5 mm roots (Ostonen *et al.,* 2007). For example, the finest roots (<0.03 mm) generated by beech may be relatively longer in rich soils to facilitate the acquisition of resources (Weemstra *et al.,* 2017). Our results indicated that the plasticity of fine-root morphologies was more closely associated with the finest roots, which are most active. In our study, we included all fine-roots ≤ 2 mm, which may have weakened the case for the plasticity of fine-root morphologies. These results indicated that the fine-root morphological traits of our sampling sites did not differ, which agreed with our second hypothesis.

No correlations between the chemical and morphological traits of fine roots

Higher altitudes led to changes in the environments where plants grow, including soil nutrients. Since fine roots are in intimate contact with the soil environment, they are the most active components of plants in terms of the absorption of nutrients. In our study, the chemical and morphological traits of fine roots exhibited various adjustment strategies to the changes in soil nutrients along the altitudinal gradient. Our results aligned with previous studies, which did not find relationships between fine-root

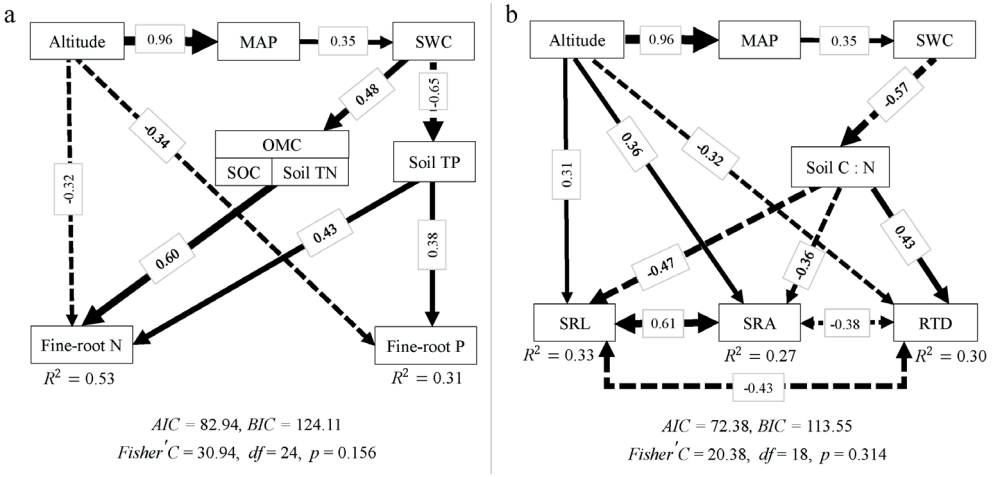

Figure 4. Effects of soil and climate factors on fine-root chemical (a) and morphological (b) traits. Solid and dotted arrows represent significant positive and negative relationships, respectively ($p < 0.05$). Arrow width is proportional to the strength of the relationship. Numbers adjacent to arrows denote standardized path coefficients. The proportion of variance explained *(R^2)* appears alongside every response variable in the model. The organic matter component (OMC) is the first component from a PCA conducted with soil organic carbon (SOC) and total nitrogen (TN).

chemical and morphological traits. Chen *et al.* (2013) found that fine-root N concentrations were not correlated with morphology (*e.g.,* diameter, SRL, length, and RTD). In a greenhouse experiment involving different species of plants under spatially or temporally heterogeneous nutrients regimes, Mou *et al.* (2013) found that correlation coefficients for the morphological vs physiological plasticity of fine roots were essentially zero under pulsed nutrient treatments. We speculated that the main reason was the genetic constraints on the plasticity of fine-root morphological traits (Kramer-Walter & Laughlin, 2017).

Several researchers have proposed a similar economics spectrum for root traits according to LES at a global scale (Chen *et al.,* 2013). Specifically, "fast-root" species should produce thin roots with short life spans and high SRL and N concentrations, while "slow-root" species possess the opposite traits (Reich, 2014). However, there is little support for this notion of a root economics spectrum. Valverde-Barrantes *et al.* (2015) explored the correlations between the chemical and morphological traits of fine roots for ~34 arbuscular mycorrhizal tree species in a temperate zone and tested to what extent they were like the LES. As a result, they found that the specific leaf area was correlated to the leaf N concentration, whereas the SRL was not correlated with the root N concentration (*i.e.,* root traits did not show the same trade-offs as leaf traits). As roots function in a far more complex environment than do leaves, it is difficult to generalize the RES from LES (Valverde-Barrantes *et al.,* 2015). Moreover, the roots of different orders or sizes may perform different functions (*e.g.,* nutrient and water uptake, transportation, or storage); however, this is not the case with leaves (Pregitzer *et al.,* 1998). In addition, mycorrhizal interactions may also affect changes in the functional traits of fine roots (Weemstra *et al.,* 2016).

In conclusion, the chemical traits of fine roots, rather than their morphological traits, exhibit plasticity in response to variations in soil nutrients induced by altitudinal gradients in Chinese fir plantations. The stability of fine-root morphologies suggests that they are restricted by the plant's own heredity. Furthermore, significant changes in the chemical traits of fine roots may be caused by the uptakes of fine-roots in response to different soil nutrient conditions.

Elucidating the relationships between fine-root traits is not parallel to leaf economic spectrum. As the environments within which roots operate are complex, their functions will differ due to their dimensional hierarchy. The notion that leaf traits are strongly affected by the availability of light is based on abundant tree leaf data on the global scale. However, there exists far less available data regarding root traits than those of leaves in the plant traits TRY database (Kattge *et al.,* 2012). Consequently, much more root data needs to be collected and analyzed (according to different root functions, tree species and so on) to refine the root economics spectrum.

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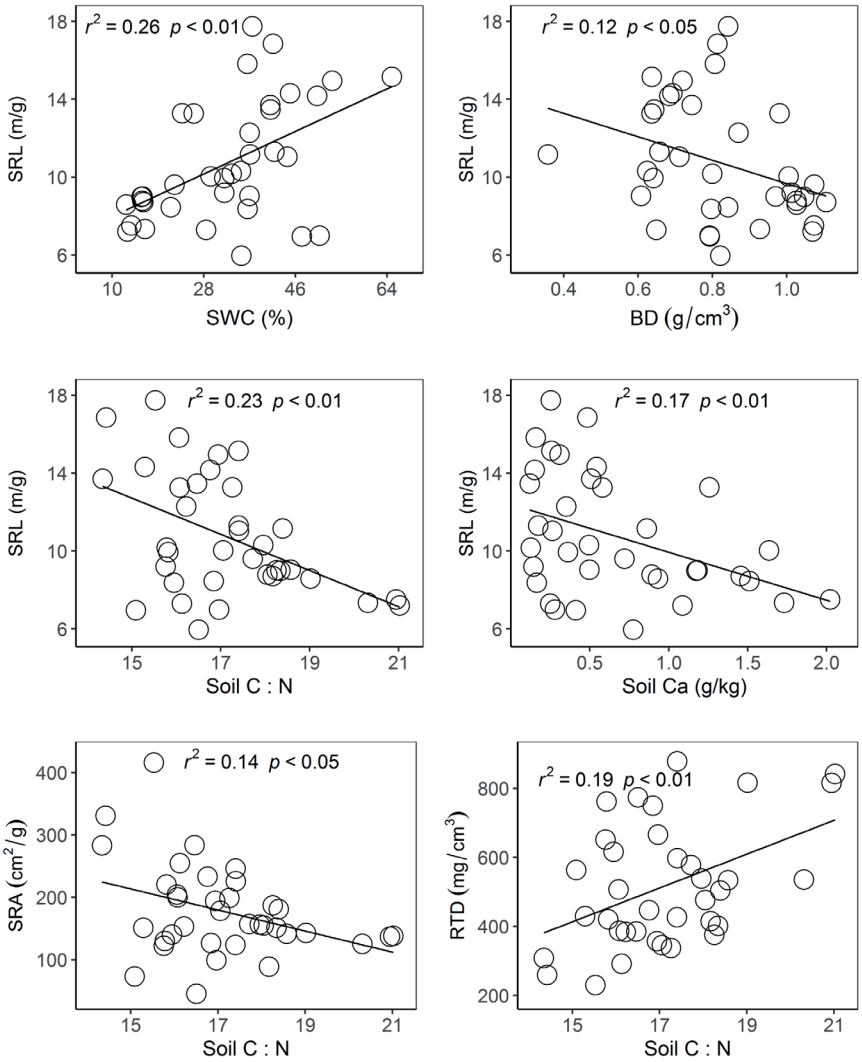

Figure 5. Bivariate relationships between fine-root morphological traits and soil characteristics. Solid lines indicate significant standard major axis (SMA) regression lines. Coefficients of determination *(r^2)* are shown for the SMA regressions using the soil characteristic data. SRL, specific root length. SRA, specific root area. RTD, root tissue density. SWC, soil water content. BD, bulk density.

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