

Article

Fine-Root Soil Stoichiometry of *Picea schrenkiana* Fisch. et Mey. and Its Correlation with Soil Environmental Factors under Different Nitrogen Input Levels in the Tianshan Mountains, Xinjiang

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Abstract: Nitrogen enters the soil surface along with the deposition and destroys the element balance of an ecosystem, which has an important impact on underground ecological processes. As active interfaces with the environment, fine roots play a key role in the processes of underground ecosystems and nutrient cycles. Nitrogen in deposition is mainly in two forms, namely organic nitrogen and inorganic nitrogen, which may have different responses to the ecological balance of fine roots and the soil environment; however, in *Picea schrenkiana* Fisch. et Mey., as a dominant species in the Tianshan Mountains of Xinjiang, it is not clear how different proportions of nitrogen deposition affect the element balance and interactions between fine roots and soil. In this study, from May 2018 to October 2020, five groups of in situ control experiments with different proportions of exogenous nitrogen addition (different ratios of ON–IN, CK = 0:0, N1 = 10:0, N2 = 7:3, N3 = 5:5, N4 = 3:7, and N5 = 0:10, were mixed and then used with equal total amounts of 10 kg·N·ha⁻¹·a⁻¹) were conducted on *Picea schrenkiana*. The results showed that inorganic nitrogen had a stronger effect on the carbon, nitrogen, and phosphorus contents of fine roots under different proportions of exogenous nitrogen addition, indicating that the fine roots of *Picea schrenkiana* had a greater response to inorganic nitrogen sources. In a mixed organic–inorganic nitrogen source with the same proportion of organic and inorganic nitrogen, the reaction between fine-root nitrogen (TN = 7.6 g·kg⁻¹–10.8 g·kg⁻¹) and soil phosphorus (TP = 0.99 g·kg⁻¹–1.93 g·kg⁻¹) was stronger, indicating that the *Picea schrenkiana* ecosystem may be a nitrogen-limited forest ecosystem. In addition, different proportions of nitrogen source inputs have an indirect impact on the fine-root stoichiometry and biomass of different root sequences through the impact on soil environmental factors and stoichiometry. Therefore, our research provides insights into the impact of increases in nitrogen on the nutrient cycling of mountain forests in arid areas and provides small-scale support for a research database of forest ecosystem responses to nitrogen deposition.

Keywords: nitrogen deposition; ecological stoichiometry; fine roots; forest ecosystem; arid area

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1. Introduction

Nitrogen deposition associated with fossil fuel burning remains a significant environmental challenge; however, there is currently limited understanding of the influence of organic and inorganic N on fine-root nutrient uptake and balance [1]. It has been estimated that the global nitrogen deposition rate will increase by two–three times by the end of the 21st century [1]. As an important part of terrestrial ecosystems, forest surface soil is the primary interface connecting with the atmosphere, which is significantly affected by nitrogen deposition. An increasing number of studies have proven that an increase in nitrogen deposition not only affects forest aboveground ecosystems but also has an

important impact on underground ecosystems [1,2]. Nitrogen cycling is one of the core processes of elemental cycling in underground ecosystems. Studying the impact of nitrogen deposition on elemental cycling in underground ecosystems has a positive role in regulating the process of soil material cycling and improving its utilization efficiency [2].

Roots are the main way for plants to obtain water and nutrients, while fine roots (diameter of ≤ 2 mm), as active interfaces with the environment, play a key role in underground ecosystem processes and nutrient cycling [3]. Fine roots are the main component of carbon fixation in soil. Due to the fast turnover speed, the carbon and nutrient content returned to soil through fine roots is equal to or higher than that of litter [4]. Nitrogen deposition may have different effects on fine-root morphology, physiological characteristics, and soil environments. Among them, the balance and interaction of elements between fine roots and soil can reflect changes in the ecological processes of underground ecosystems. The results obtained by studying the effects of exogenous nitrogen inputs on fine roots and soil are highly variable. For example, when other nutrients are not simultaneously restricted in a subtropical evergreen broad-leaved forest limited by nitrogen, N treatment increased the concentration of fine-root C and N but had no significant effect on the concentration of P [5]; however, in tropical forests without nitrogen limitation, the addition of N significantly reduced the carbon contents of fine roots [6]. In addition, nitrogen application can significantly reduce soil pH values, thus leading to changes in the availability of soil N, reducing the soil C/N ratio, and improving the total nitrogen as well as N/P ratio of fine roots. Some studies have also shown that nitrogen application did not change the P content in fine roots [7], which may be related to different tree species in the study area. The time of the nitrogen application test also affected the final results. For example, a one-year nitrogen deposition test of Chinese fir seedlings found that the C content, C/N ratio, and C/P ratio of fine roots were significantly lower [8], while a two-year test found that nitrogen application significantly reduced P contents in soil but had no significant impact on C and N contents. The results of the different effects of nitrogen deposition on fine-root element circulation are closely related to the region, vegetation, climate, soil, nitrogen background value, nitrogen application test mode, and other factors of the research object, and are uncertain.

Scholars have usually used a single nitrogen source in simulated nitrogen deposition experiments. In fact, nitrogen in nitrogen deposition mainly includes organic nitrogen components and inorganic nitrogen components, and different plants have a great preference for nitrogen components' absorption [9]. Many plant species have been proven to be able to absorb complete organic nitrogen from soil, especially in nitrogen-limited ecosystems. For example, *Cymbidium hybridum* [10] and eucalyptus [11], growing in subtropical rainforests, can completely absorb organic nitrogen; however, there is also evidence that plants use almost the same amount of organic nitrogen and inorganic nitrogen or prefer to absorb inorganic nitrogen. For example, some subtropical plants have a strong preference for inorganic nitrogen, and organic nitrogen may become an important nitrogen source only when soil inorganic nitrogen is insufficient [12]. The influence of compound nitrogen sources on the underground element cycle is also unclear. Therefore, the effects of increased nitrogen deposition on forest fine roots and soil can be more comprehensively understood through organic nitrogen and inorganic nitrogen addition experiments.

The mountain forests of the Tianshan Mountains in Xinjiang are widely populated with *Picea schrenkiana* Fisch. et Mey. forests. As a dominant species, *Picea schrenkiana* plays an irreplaceable ecological function in soil conservation, water conservation, biodiversity protection, carbon fixation, and oxygen release [13]. The root system of *Picea schrenkiana* is shallow, and the depth of the root system is generally not more than 60 cm after adulthood, which is mainly distributed in the soil mineral layer [14]. At present, most studies on the response of the natural forest of *Picea schrenkiana* to exogenous nitrogen addition cover the effects of nitrogen addition on forest productivity, litter decomposition, soil nutrients, etc., but research on the interaction of nitrogen sources with underground ecological processes, such as, mainly, fine roots, is relatively insufficient, and the processes as well as mechanisms of the impact of nitrogen addition on fine roots are still unclear.

In this study, the effect of exogenous nitrogen addition on the fine roots and soil ecochemometric characteristics of *Picea schrenkiana* in the Tianshan Mountains of Xinjiang was studied by a field simulation control experiment with compound nitrogen addition. This study aims to answer the following questions: (1) What is the effect of different proportions of nitrogen source addition on the fine-root and soil ecological stoichiometry of *Picea schrenkiana* forests? (2) Do the different proportions of nitrogen source addition change the interaction between fine-root soil ecological stoichiometry and environmental factors in *Picea schrenkiana* forests?

2. Materials and Methods

2.1. Overview of the Study Area

The study area is located in the *Picea schrenkiana* forest area (43°47' N, 87°18' E) near Juhuatai, Gangou Township, which is in the middle mountain belt at the northern foot of the Tianshan Mountains, Xinjiang Uygur Autonomous Region, approximately 56 km away from Urumqi City, with an altitude of 2000–2400 m. The climate of the study area is a temperate continental climate, with large temperature differences in different seasons. The annual average temperature is approximately 0–4 °C. The annual precipitation is low, and the seasonal difference is large. The annual precipitation is approximately 176 mm. The soil type is weakly acidic gray-brown forest soil. The main forest type of the Tianshan Mountains is boreal temperate coniferous forest, and *Piceas chrenkiana* is the dominant species on the shady slopes of the middle- and high-altitude mountains in the study area.

2.2. Experimental Design

In this experiment, urea (CO(NH₂)₂) and glycine were selected as organic nitrogen sources, and ammonium nitrate (NH₄NO₃) was selected as the inorganic nitrogen source to simulate nitrogen deposition. A fixed sample plot with the same altitude, slope, aspect, and growth trend of trees was set in the study area. Eighteen quadrats were set in the sample plot, with an area of 3 m × 3 m, and a 5 m buffer zone was set between each quadrat. Six gradients were designed for the two nitrogen source interaction experiments. A total of one control and five nitrogen treatments were set, including applying nitrogen fertilizer with organic nitrogen and inorganic nitrogen according to three gradients, low, medium and high, and setting three organic nitrogen/inorganic nitrogen ratio gradients according to the local nitrogen deposition level [15] (10 kg N ha⁻¹ a⁻¹) (Table 1). Three replicates were set for each treatment group; that is, three quadrats were randomly selected for the same treatment in the same forest. From 2018, nitrogen was applied at the end of the month (growing season) from May to October each year. The nitrogen fertilizer required by each sample was dissolved in 500 mL of distilled water, a hand-held pressurized spray was used to evenly spray the sample plot, and the same amount of distilled water was sprayed in the control sample plot.

Table 1. Nitrogen application gradient in the cross-test.

Gradient	Organic N/kg·hm ⁻¹ ·a ⁻¹	Inorganic N/kg·hm ⁻² ·a ⁻¹
N0	0	0
N1	10	0
N2	7	3
N3	5	5
N4	3	7
N5	0	10

2.3. Field Survey and Sampling

Fine-root samples and soil samples were collected in September 2021. Fine-root sample collection: Within 1–1.5 m of the trunk base of the sample tree in each sample plot, 3 samples were collected with an area of 20 cm at a soil depth of 0–30 cm along the direction of the lateral root connected with the main root 20 cm × 20 cm soil block (0–10 cm, 10–20 cm, and 20–30 cm). The complete root system was selected, bagged, marked, and transported to the laboratory. After the fine roots were brought back to the laboratory, the surface attachments were cleaned. Tweezers were used to classify roots according to the size of the root order. The root system was classified according to the methods of Pregitzer [16] and others. The roots located at the most distal end of the root system and without branches are called first-order fine roots; first-order fine roots grow on second-order fine roots, and second-order fine roots grow on third-order fine roots until the whole complete root system is divided (Figure 1). All fine-root samples were placed into envelopes and baked in an oven (65 °C) for approximately 8 h to achieve a constant weight before weighing. All of the root samples were crushed, passed through a 2 mm sieve, and then placed into bags for testing.

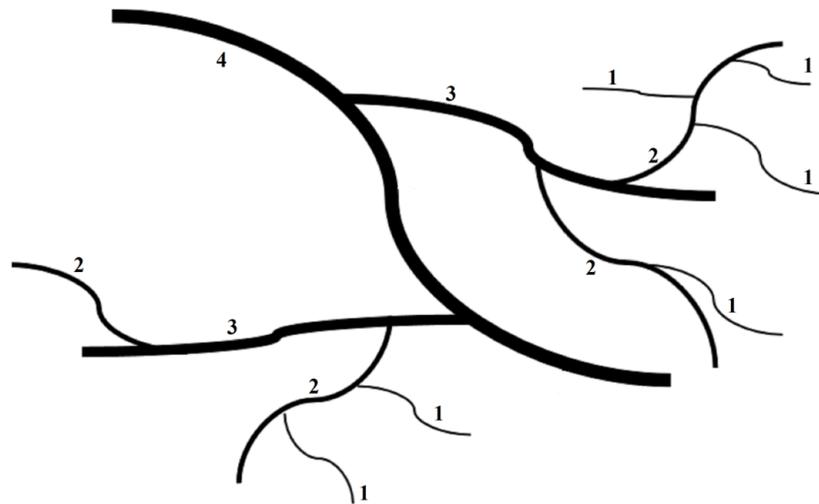


Figure 1. Schematic diagram of a root sequence [16]. 1 is the first-order fine roots; 2 is the second-order fine roots; 3 is the third-order fine roots; and 4 is the fourth-order fine roots.

Soil samples were collected according to random and uniform sampling standards. Litter layers were removed from the sample plots and excavated in each sample plot (with a plane of 20 cm × 20 cm, 60 cm high), three soil columns were collected, and samples were collected every 10 cm in the layers (i.e., 0–10 cm, 10–20 cm, and 20–30 cm). After the samples were collected, the soil samples were screened and stored after natural air drying for the determination of physical and chemical properties.

2.4. Determination of the Ecological Stoichiometry Characteristics of Fine Roots

Organic carbon was determined by the potassium dichromate external heating method; the semimicro Kjeldahl method was used for total nitrogen, the fresh soil samples were extracted from a 2 M KCl solution and filtered, and the contents of NH_4^+ and NO_3^- were then analyzed with a flow injection autoanalyzer; and $\text{HClO}_4\text{-H}_2\text{SO}_4$ molybdenum antimony anti-colorimetry was used for total phosphorus. All of the soil layers and root sequence fine roots were placed in a 65 °C oven for drying [17].

2.5. Determination of Soil Environmental Factors and Ecological Stoichiometry Characteristics

Determination of soil physical and chemical indices: soil moisture content was determined by the drying method; pH values were measured by a pH meter; organic carbon was determined by the potassium dichromate external heating method; the semimicro Kjeldahl method was used for total nitrogen; and $\text{HClO}_4\text{-H}_2\text{SO}_4$ molybdenum antimony anti-colorimetry was used for total phosphorus [17].

2.6. Mathematical Statistical Methods

SPSS23 and Origin2018 were used for statistical analyses of fine-root values of different nitrogen additions, different soil layers, and root orders by the LSD method. Single-factor analysis of variance was used to compare the intragroup differences among different data groups. CANOCO5 software was used for redundancy analysis (RDA), and R was used to establish a partial least squares path model (PLS-PM).

3. Results

3.1. Fine-Root Biomass and Ecological Stoichiometry

A single-factor ANOVA diagram of the ecological stoichiometry and biomass characteristics of fine roots of *Picea schrenkiana* in different soil layers under different nitrogen source input levels (Figure 2) showed that the biomass, organic carbon, and nitrogen-related stoichiometry of fine roots of *Picea schrenkiana* were most sensitive to inorganic nitrogen, and the content of total phosphorus was relatively stable under the application of exogenous nitrogen. Compared with different nitrogen source treatments, the biomass and ecological stoichiometry of fine roots in each soil layer changed significantly under the N3 (organic nitrogen:inorganic nitrogen = 1:1) treatment. The C/N and C/P ratios of fine roots increased with the deepening of the soil layers, and the most significant changes were found in the 0–10 cm soil layer. The N/P ratio of fine roots had no significant differences among different soil layers under other N treatments, except the N4 treatment; it first decreased and then increased with an increase in the proportion of inorganic nitrogen in the mixed organic–inorganic nitrogen source.

The single-factor analysis of the variance in fine-root ecological stoichiometry and biomass characteristics of *Picea schrenkiana* in different root orders under different nitrogen source input levels (Figure 3) showed that the fine-root biomass and ecological stoichiometry were basically consistent with the soil layer. A single nitrogen source had a strong impact on the fine-root biomass and ecological stoichiometry at all levels. Although the fine-root total phosphorus had a strong response to organic nitrogen, the rest had a greater response to inorganic nitrogen. Compared with different nitrogen source treatments, there was no significant difference in total nitrogen and total phosphorus contents under the treatment of the mixed organic–inorganic nitrogen source; the carbon/nitrogen, carbon/phosphorus, and nitrogen/phosphorus ratios decreased first.

3.2. Soil Environmental Factors and Ecological Stoichiometry

The single-factor ANOVA of soil physical and chemical factors as well as ecological stoichiometry under different soil layers at different nitrogen source input levels in the study area showed that the response of soil and fine roots to external-source nitrogen was consistent (Figure 4). Compared with the mixed organic–inorganic nitrogen source, except for total phosphorus, a single nitrogen source had more significant effects on soil and had a more significant response to single inorganic nitrogen addition. Compared with different treatments of the mixed organic–inorganic nitrogen source, the effect of the mixed organic–inorganic nitrogen source on the stoichiometric ratio was more obvious, showing a “U” trend, and was lower than the control group as a whole. The changing range of the carbon/phosphorus and nitrogen/phosphorus ratios was larger than that of the carbon/nitrogen ratio, which also verified that total phosphorus was more sensitive to the response of the mixed organic–inorganic nitrogen source.

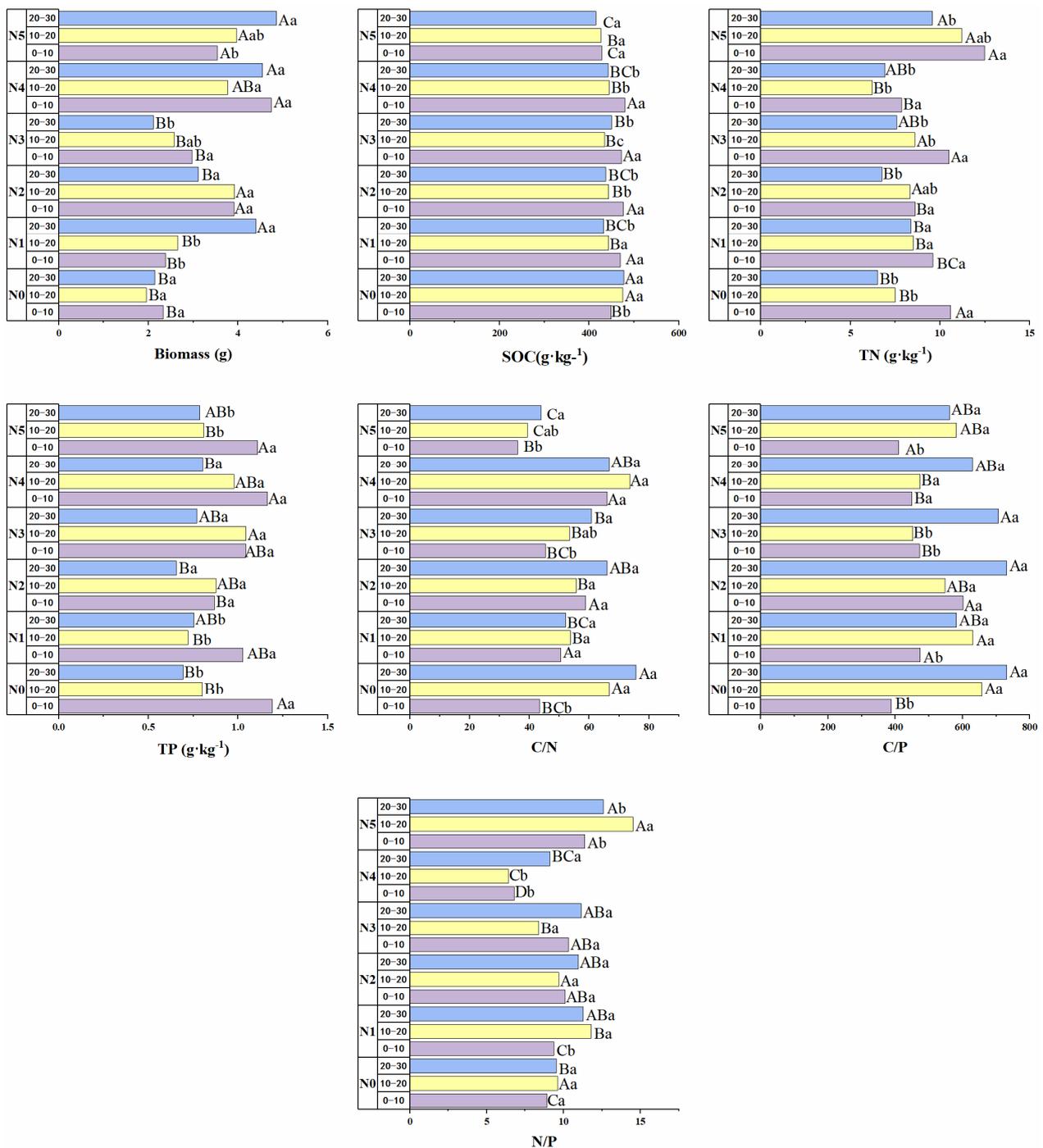


Figure 2. One-way ANOVA of soil ecological stoichiometry and biomass characteristics of *Picea schrenkiana* in different soil layers under different nitrogen source input levels in the Tianshan Mountains, Xinjiang. Different capital letters indicate significant differences in the ecological stoichiometry of elements in the same soil layer under different nitrogen sources ($p < 0.05$); different lowercase letters indicate significant differences in the ecological stoichiometry of different soil layer elements under the same nitrogen source addition ($p < 0.05$).

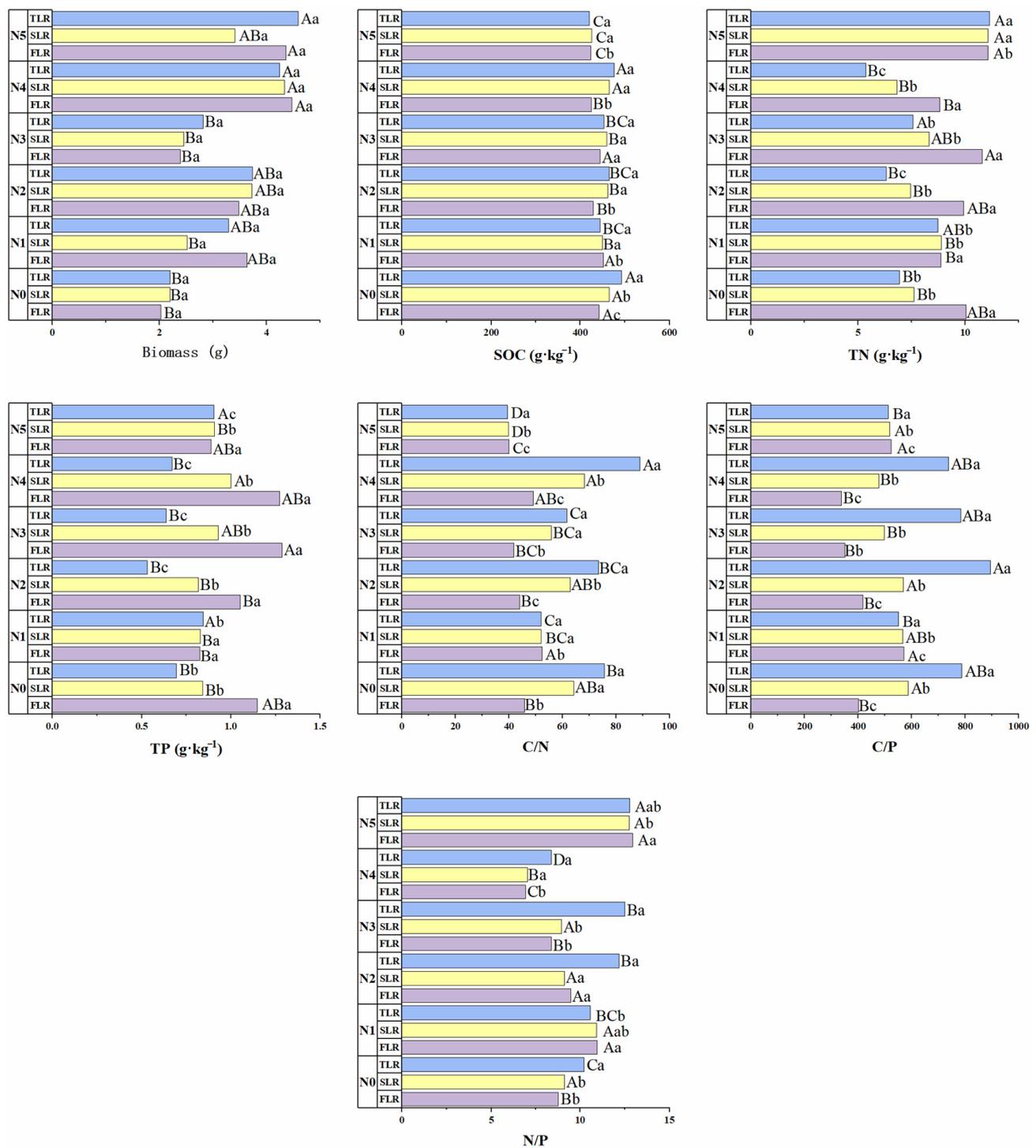


Figure 3. One-way ANOVA for soil ecological stoichiometry and biomass characteristics of *Picea schrenkiana* with different root sequences at the level of different nitrogen source inputs in the Tianshan Mountains, Xinjiang. FLR is first-order fine roots; SLR is second-order fine roots; and TLR is third-order fine roots. Different capital letters indicate that there is a significant difference in the ecological stoichiometry of elements in the same root order under different nitrogen sources ($p < 0.05$); different lowercase letters indicate that there is a significant difference in the ecological stoichiometry of elements in different root orders under the same nitrogen source addition ($p < 0.05$).

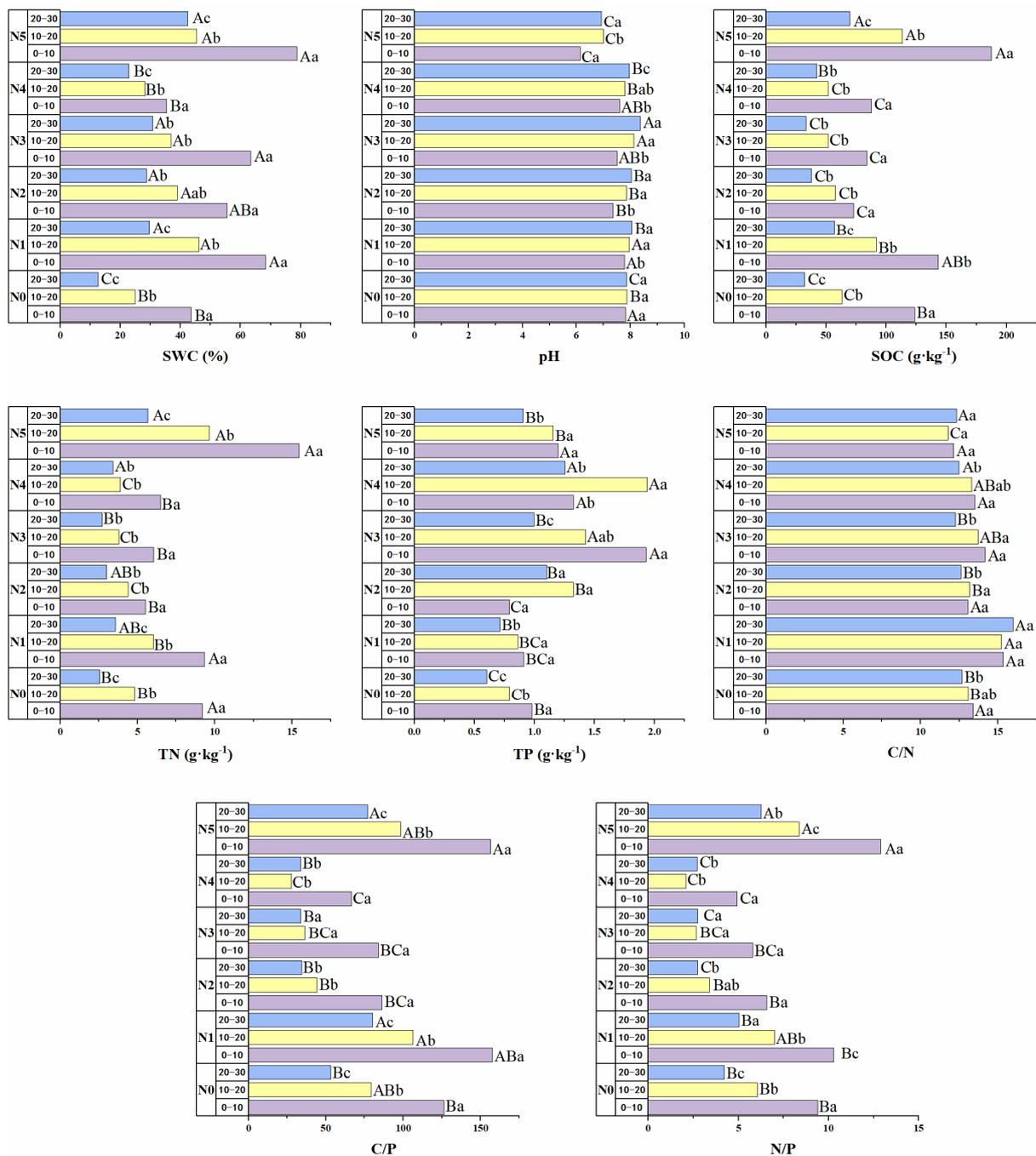


Figure 4. One-way ANOVA of soil ecological stoichiometry and biomass characteristics of *Picea schrenkiana* in different soil layers under different nitrogen source input levels in the Tianshan Mountains, Xinjiang. FLR is first-order fine roots; SLR is second-order fine roots; and TLR is third-order fine roots. Different capital letters represent significant differences in the ecological stoichiometry of elements in the same root order under different nitrogen sources ($p < 0.05$); different lower case letters represent significant differences in the ecological stoichiometry of elements in different root orders under the same nitrogen sources ($p < 0.05$).

3.3. Fine-Root Soil Ecological Stoichiometry

The RDA results of organic nitrogen, inorganic nitrogen, and soil indicators showed (Figure 5a,b) that the fine-root biomass and total nitrogen had the strongest ecological stoichiometry correlation with soil, in which the interpretation amount of the inorganic nitrogen ordination axis was greater than that of organic nitrogen and the mixed organic–inorganic nitrogen source; the cumulative characteristic values of the relationship between the primary root ecological stoichiometry and environmental factors of Axis I and Axis II reached 78.7% and 97.3%, respectively (Table 2). In Figure 5c, the cumulative characteristic value of the relationship between the first-level root ecological stoichiometry and environmental factors of Axis I and Axis II reached 97.8%, and there was no significant bias in the fine-root soil ecological stoichiometry correlation under the mixed organic–inorganic nitrogen source deposition compared with the application of a single nitrogen source.

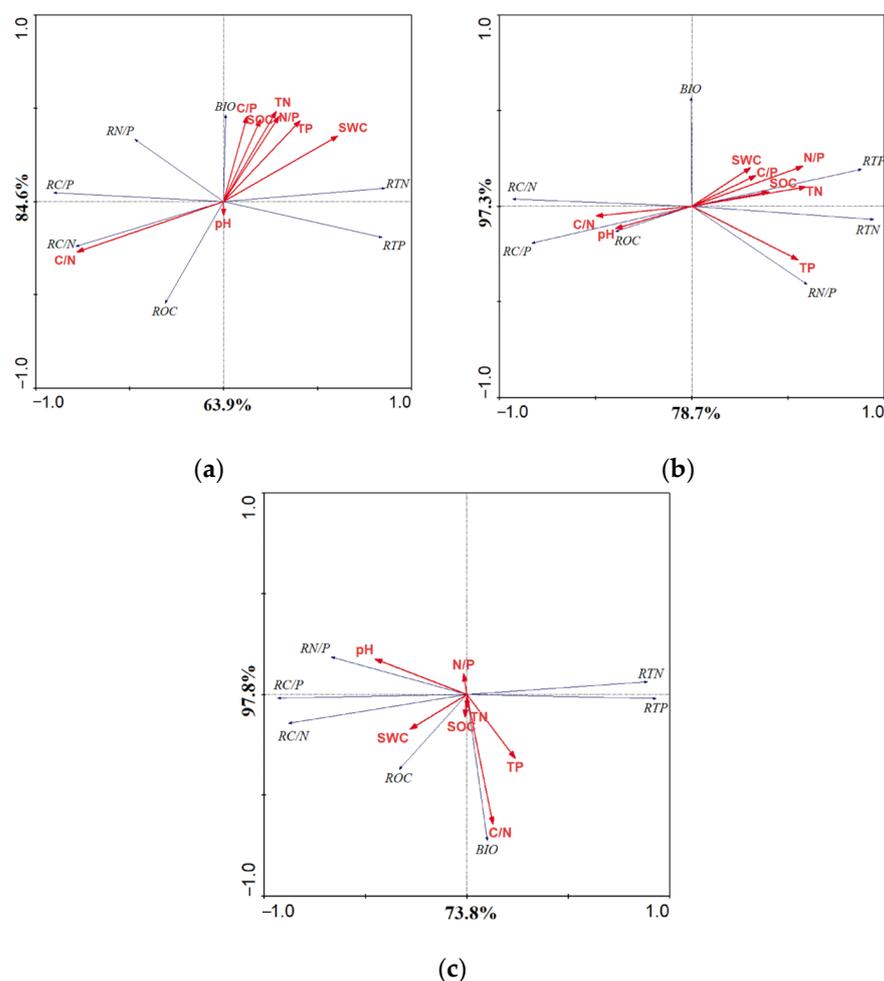


Figure 5. RDA of the fine-root–soil stoichiometry of *Picea schrenkiana* at different root sequences under the mixed organic–inorganic nitrogen source input levels in the Tianshan Mountains, Xinjiang. Figure 5 (a) shows the stoichiometric RDA of fine-root soil of *Picea schrenkiana* at the level of organic nitrogen inputs; (b) shows the stoichiometric RDA of fine-root soil of *Picea schrenkiana* at the level of inorganic nitrogen inputs; and (c) shows the stoichiometric RDA of fine-root soil of *Picea schrenkiana* at the level of the mixed organic–inorganic nitrogen source inputs. BIO is fine-root biomass, ROC is fine-root organic carbon, RTN is fine-root total nitrogen, RTP is fine-root total phosphorus, RC/N is the fine-root carbon/nitrogen ratio, RC/P is the fine-root carbon/phosphorus ratio, RN/P is the fine-root nitrogen/phosphorus ratio, SWC is soil water content, SOC is soil organic carbon, STN is soil total nitrogen, STP is soil total phosphorus, SC/N is the soil carbon/nitrogen ratio, SC/P is the soil carbon/phosphorus ratio, and SN/P is the soil nitrogen/phosphorus ratio.

Table 2. Redundancy analysis of eigenvalues for the fine-root–soil stoichiometry of *Picea schrenkiana* at different root sequences under the level of the mixed organic–inorganic nitrogen source.

Cumulative Percentage Variance	Axis I	Axis II	Axis III	Axis IV
Organic nitrogen relation	63.90	84.60	99.30	100.00
Inorganic nitrogen relation	78.70	97.30	99.80	100.00
Mixed organic–inorganic nitrogen relation	73.80	97.80	99.10	100.00

The partial least squares structural variance model (PLS-PM) showed that the input of external nitrogen sources indirectly affected the stoichiometry and biomass of fine roots of different root orders through the impact on soil environmental factors and stoichiometry (Figure 6). Under the single nitrogen source inputs the soil promoted fine roots, while under the mixed organic–inorganic nitrogen source inputs the environment mostly inhibited fine roots; however, under different nitrogen sources fine roots with different root orders had a positive effect. From the model, it was found that the effect of the soil ecological stoichiometry on the ecological stoichiometry of low-order fine roots was greater than that of high-order fine roots.

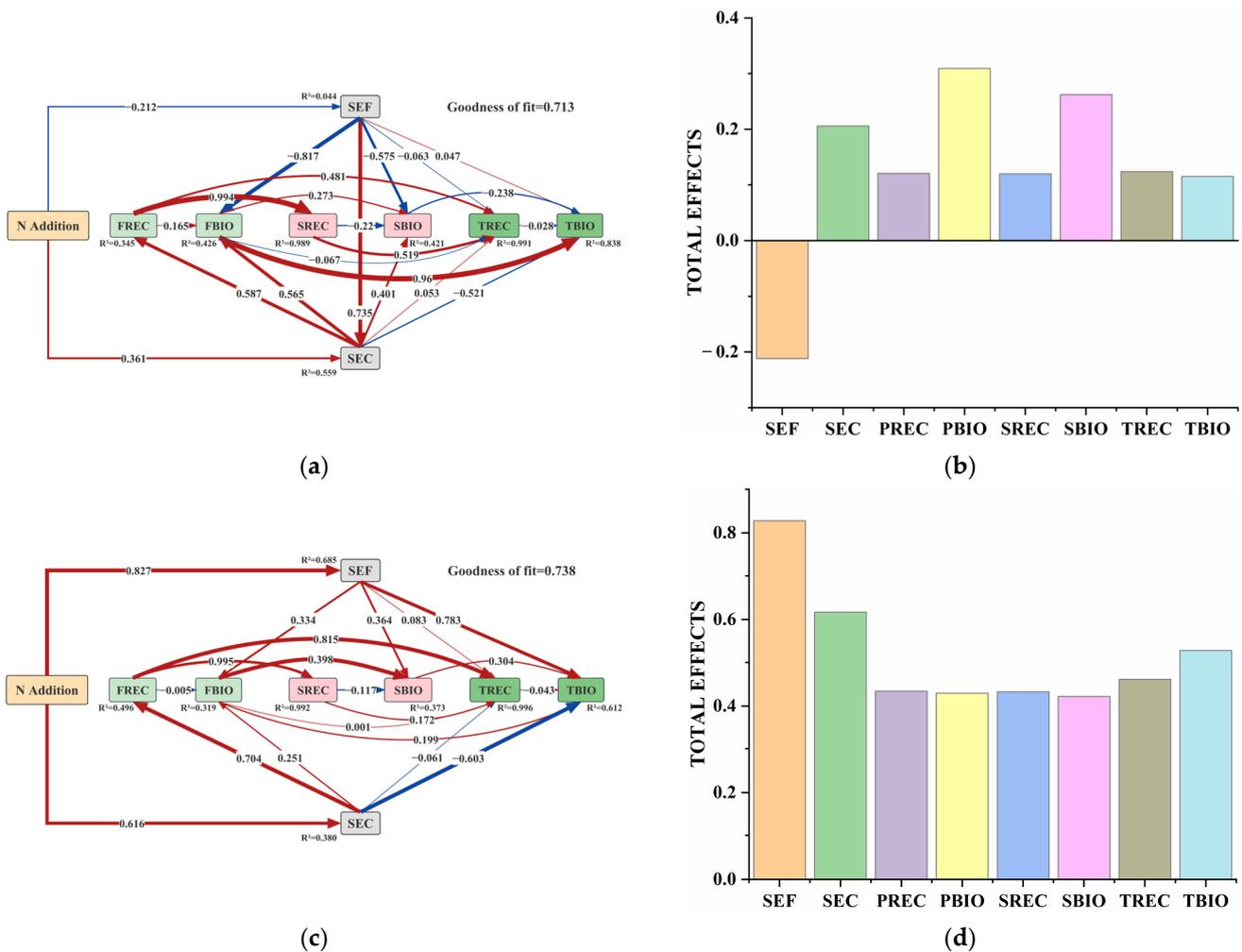


Figure 6. Cont.

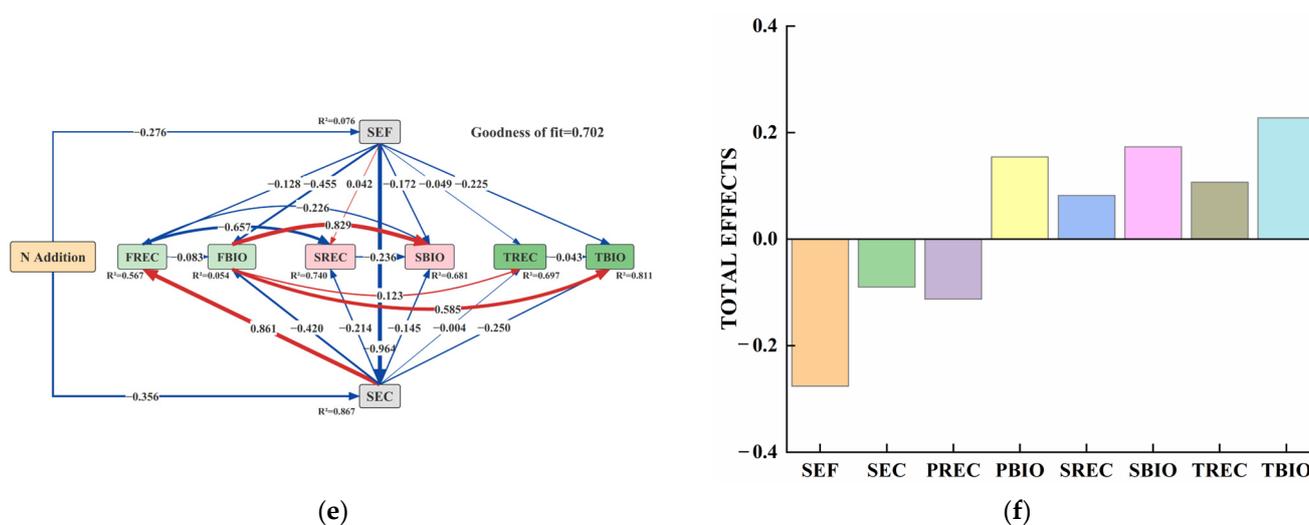


Figure 6. PLS model of the fine-root–soil stoichiometry of *Picea schrenkiana* with different root sequences at the level of the mixed organic–inorganic nitrogen source inputs in the Tianshan Mountains, Xinjiang. (a) shows the stoichiometric PLS model of fine-root soil of *Picea schrenkiana* at the organic nitrogen input level; (b) is the overall impact histogram of the fine-root soil stoichiometric PLS model of *Picea schrenkiana* at different nitrogen input levels; (c) shows the stoichiometric PLS model of the fine-root soil of *Picea schrenkiana* at inorganic nitrogen input levels; (d) is the overall impact histogram of the fine-root soil stoichiometric PLS model of *Picea schrenkiana* at different nitrogen input levels; (e) shows the stoichiometric PLS model of fine-root soil of *Picea schrenkiana* at the mixed organic–inorganic nitrogen source level; and (f) shows the overall impact histogram of the fine-root soil stoichiometric PLS model of *Picea schrenkiana* under different nitrogen input levels. The red arrow represents a positive correlation, and the blue arrow represents a negative correlation. N addition denotes the nitrogen source, SEC is the soil ecological stoichiometry, SEF is the soil environmental factor, FRTN is the first-order fine-root ecological stoichiometry, FBIO is the first-order fine-root biomass, SRTN is the second-order fine-root ecological stoichiometry, SBIO is the second-order fine-root biomass, TRTN is the third-order fine-root ecological stoichiometry, and TBIO is the third-order fine-root biomass.

4. Discussion

4.1. Ecological Stoichiometry Effects of Different Proportions of Nitrogen Inputs on the Fine Roots of *Picea Schrenkiana*

Ecological stoichiometry provides an effective way to explore the mutual coupling of carbon, nitrogen, and phosphorus between plants and soils, as well as the relationship between plant growth and nutrient supply. Ecological stoichiometry of the root system is the most direct adaptive strategy for plants to change soil environments. Organic carbon is the most important element that constitutes the dry matter of a plant, and in previous studies on temperate forests nitrogen addition generally increased the soil organic carbon content, contrary to the conclusions obtained in this study [18]. In this study, it was found that different nitrogen addition levels inhibited the organic carbon content of fine roots, and that inorganic nitrogen had a stronger effect. These results can be attributed to the excessive inputs of exogenous nitrogen. High nitrogen levels are not necessarily conducive to material accumulation but will lead to the excessive growth of fine roots, which will lead to unbalanced nutrient absorption, thus affecting the accumulation of organic carbon in fine roots. At the same time, 75% of the organic carbon in roots is used for fine-root respiration [19], while the addition of nitrate to inorganic nitrogen improves fine-root respiration and consumes a large amount of nonstructural carbohydrates (NSCs) [20], leading to a greater reduction in fine-root organic carbon. Many studies have shown that nitrogen deposition generally increases the nitrogen content in the fine roots of plants [6,21], indicating that temperate forests are mostly nitrogen-restricted forests. This study further

supports this conclusion. Exogenous nitrogen inputs directly increase the availability of soil nitrogen; therefore, fine roots can absorb nitrogen from the soil, which is absorbed by root cells to increase the total nitrogen content of fine roots [22]. Our data showed that the total nitrogen of fine roots was significantly higher under inorganic nitrogen treatment; the response of fine roots was the largest when organic nitrogen was used to replace 25%–30% of inorganic nitrogen at the same nitrogen level under the addition of the mixed organic–inorganic nitrogen source. This may be because, when inorganic nitrogen is added, it acidifies the soil more significantly than organic nitrogen does, reduces the release of nitrous oxide in the soil, further enriches the nitrogen sources that can be absorbed by fine roots, and increases the total nitrogen in fine roots [23]. Other studies have found that nitrogen addition not only affects the absorption of nitrogen by fine roots but also may increase the absorption of phosphorus [24]; however, some studies have also shown that nitrogen deposition may not significantly affect the phosphorus content in plants' fine roots [25]. In this study, we did not find that the addition of nitrogen to *Picea schrenkiana* had a promoting effect on phosphorus, and the fine-root total phosphorus had a relatively stable level under the different proportions of nitrogen source addition. Different proportions of nitrogen source inputs will lead to unbalanced nitrogen and phosphorus deposition, which may aggravate the limitation of phosphorus in an ecosystem. As a place with an active plant physiological metabolism, roots need to maximize the stability of phosphorus content under the condition of exogenous nitrogen addition to ensure the normal physiological activities of fine roots [26].

The carbon/nitrogen ratio of the plant body is an important indicator reflecting the physiological metabolism status of the plant body. It represents the ability of fine roots to absorb nitrogen and assimilate carbon, indicating the level of nitrogen utilization efficiency and carbon fixation efficiency of fine roots. The addition of exogenous nitrogen reduced the C/N in the fine roots of *Picea schrenkiana*, which is consistent with the meta-analysis results of Li et al. [27]. This may be because exogenous nitrogen inputs promote nutrient transport in fine roots by increasing nitrogen use efficiency and reducing carbon assimilation capacity [28]. The fine-root C/P ratio plays an important role in reflecting the effect of plant roots on soil carbon accumulation as well as nutrient cycling and can be used to evaluate the phosphorus utilization efficiency of plants. In this study, it was found that the C/P ratio of fine roots decreased significantly with the addition of inorganic nitrogen, which may be because the addition of inorganic nitrogen significantly reduced the organic carbon content of fine roots and did not significantly increase the growth rate of fine roots, while the total phosphorus in fine roots remained stable. The ratio of nitrogen to phosphorus can be used as a sensitive index of plant responses to nitrogen deposition. At present, most research results indicate that a low nitrogen/phosphorus ratio ($N/P < 14.5$) indicates that plants are more constrained by nitrogen. In this study, different proportions of nitrogen inputs increased the nitrogen-to-phosphorus ratio in fine roots, and the biochemical reaction mechanisms in plant cells responded to exogenous nitrogen inputs. The total phosphorus ensures the rapid synthesis of proteins or enzymes and maintains stability, but an increase in the total nitrogen in fine roots increases the nitrogen/phosphorus ratio in fine roots [29]; however, the N/P ratio of fine roots of *Picea schrenkiana* was less than 14.5 under different nitrogen sources, which further proved that the *Picea schrenkiana* forest was a nitrogen-limited forest.

In this study, the influence of the ecological stoichiometry of fine roots of *Picea schrenkiana* increased gradually with an increase in the inorganic nitrogen ratio in the nitrogen sources. The preference of fine roots of *Picea schrenkiana* for inorganic nitrogen was much higher than that for organic nitrogen, which was consistent with the research results of Sun [30]. First, organic nitrogen provides a slower and more stable nitrogen supply. Nitrate in organic nitrogen requires higher energy consumption than ammonium ions in inorganic nitrogen. To absorb nitrogen in soil faster when inorganic nitrogen is sufficient, plants will preferentially absorb inorganic nitrogen, resulting in a stronger response of fine roots to inorganic nitrogen. Second, *Picea schrenkiana* grows in acidic forest soil with a

special nitrogen conservation mechanism. Research by Zhang et al. [31] showed that nitrate in organic nitrogen can be effectively fixed in organic nitrogen pools in acidic soil; therefore, fine roots have a strong response to inorganic nitrogen inputs. In addition, microorganisms in rhizosphere soil may compete with plants' fine roots for organic nitrogen, making a large part of organic nitrogen unable to be absorbed and utilized by plants [30].

4.2. Effect of Different Proportions of Nitrogen Source Inputs on the Soil Ecological Stoichiometry of *Picea Schrenkiana*

Exogenous nitrogen entering forest ecosystems will directly affect the exchange of energy flow and material flow in soil. The carbon and nitrogen cycle processes of terrestrial ecosystems are closely related, and nitrogen inputs have a great impact on soil organic carbon. Previous researchers conducted eight consecutive years of simulated nitrogen deposition tests in the NITREX experimental base, which showed that exogenous nitrogen inputs can significantly promote the production and leaching processes of soil soluble organic carbon [32]. In this study, single nitrogen sources of organic nitrogen and inorganic nitrogen promoted soil organic carbon. This is because the single nitrogen addition reduced soil pH, inhibited soil microbial activity, reduced the soil carbon mineralization rate, and regulated the excitation effect to reduce soil organic carbon loss, thus promoting organic carbon accumulation. The content of soil organic carbon decreased under the application of the mixed organic–inorganic nitrogen source, which may be because, when applying the mixed organic–inorganic nitrogen source, compared with nitrate in inorganic nitrogen, microorganisms preferentially absorb ammonium nitrogen (organic nitrogen) because its energy cost is lower, which means that, in the same test cycle, soil microorganisms have a selection effect and smaller organic nitrogen inputs enhance soil microbial activity, leading to a decrease in soil organic carbon content [33]. Previous studies have generally shown that exogenous nitrogen inputs will change the net nitrogen mineralization rate of soil, increase the release of nitrogen elements, and significantly increase the total nitrogen content in soil [34]. Our data show that different proportions of nitrogen source inputs can significantly increase the soil total nitrogen content, and that the increase in soil total nitrogen is more significant under inorganic nitrogen addition. The addition of exogenous nitrogen will promote the nitrification and ammonification of soil nitrogen as well as improve the biological availability of nitrogen. The input amount of soil nitrogen exceeds the amount of nitrogen lost due to N_2O emissions, NO_3^- leaching, and other ways, which will lead to the accumulation of nitrogen [35]. Nitrate in inorganic nitrogen promotes the absorption of nitrogen by soil, accelerates the mineralization rate of soil organic matter, the nitrification and denitrification of ecosystems, and increases soil nitrogen content [36]. Phosphorus is also one of the main limiting elements in temperate forest ecosystems and has a single source in the soil. This study shows that different proportions of exogenous nitrogen addition increase the total phosphorus content in the soil, and Keeler et al. [37] also obtained consistent research results. Organic nitrogen can improve the total phosphorus content of soil because the deposition of organic nitrogen stimulates the activity of soil microorganisms, thereby improving the activity of related enzymes and accelerating the decomposition of litter and organic matter, thus promoting the transformation of phosphorus in the soil. The above process leads to an increase in soil phosphorus content [38]. The input of inorganic nitrogen will cause the iron–aluminum-bound phosphorus (NaOH Pi and NaOH Po), which accounts for 40% of the total phosphorus content of forest soil, to be hydrolyzed and released into the soil, supplementing the absorbed and utilized phosphorus content of the soil [39], thus increasing the total phosphorus content in the soil.

The C/N ratio of soil can indicate the nitrogen mineralization ability of soil, and its size can indicate whether nitrogen will be fixed or mineralized during the decomposition of organic matter. A higher C/N ratio is conducive to soil nitrogen fixation, and soil nitrogen mineralization will increase when the soil C/N ratio is low. In this study, different proportions of nitrogen source addition reduced the soil carbon/nitrogen ratio. It may be that, with an increase in the nitrogen deposition concentration, the nitrogen enriched in

the soil increases, which reduces the soil carbon/nitrogen ratio and further strengthens the soil nitrogen mineralization rate. The soil C/P ratio is a characterization parameter of the availability of phosphorus. The smaller the C/P ratio is, the higher the availability of phosphorus in soil. Due to the relatively large amount of precipitation in the study area, the topsoil is easily leached, leading to carbon loss, resulting in a relative lack of carbon availability and low soil carbon as well as phosphorus. Therefore, the phosphorus availability in the study area is high. The ratio of nitrogen to phosphorus in soil can usually indicate that the ecosystem is limited by nitrogen or phosphorus. The general view is that a low nitrogen and phosphorus ratio can indicate that plants are limited by nitrogen, while a high nitrogen and phosphorus ratio can indicate that plants are limited by phosphorus. In this study, the addition of inorganic nitrogen increased the ratio of soil nitrogen to phosphorus, which may be because inorganic nitrogen played a significant role in promoting soil total nitrogen and total phosphorus; the increase in total phosphorus was higher than that of total nitrogen, leading to a decrease in the ratio of soil nitrogen to phosphorus, which further indicates that the study area may be a nitrogen-restricted forest.

The soil of *Picea schrenkiana* forests is more sensitive to ecological stoichiometry changes when the inorganic nitrogen source accounts for a large proportion. The addition of inorganic nitrogen inhibited the decomposition of litter [40], and the turnover rate decreased, resulting in the accumulation of ecological stoichiometric elements in the soil. Additionally, the growth of plants in low-temperature environments mainly depends on organic nitrogen. Under relatively warm conditions, inorganic nitrogen is used as the main nitrogen nutrient [41]. In the sampling season, the temperature in the study area was high, and plants were likely to have used inorganic nitrogen.

4.3. Ecological Stoichiometry Correlation and Influencing Factors of Fine-Root Soil of *Picea Schrenkiana* with Different Proportions of Nitrogen Inputs

Ecstoichiometry exchange between vegetation and soil constitutes an important part of the biochemical cycle of terrestrial ecosystems. Therefore, there is an inevitable relationship between the ecological stoichiometry of soil and fine roots. After redundant analysis of fine-root and soil ecological stoichiometry in this study, it was found that the correlation degree of fine-root soil ecological stoichiometry was different under different proportions of nitrogen source addition, in which fine-root total nitrogen had the strongest correlation with soil ecological stoichiometry; the interpretation amount of the inorganic nitrogen ordination axis was greater than that of organic nitrogen and the mixed organic–inorganic nitrogen source. Exogenous nitrogen inputs can increase the fixation of nitrate nitrogen so that more nitrogen is stored in the surface soil, and the root layer of *Picea schrenkiana* is also mainly located in the surface layer; therefore, it is conducive to the absorption of nitrogen by the fine roots of *Picea schrenkiana* and reduces its leaching amount [42]. The residue produced after the death of fine roots is one of the important sources of soil total nitrogen increase; therefore, total nitrogen has the strongest correlation with soil ecological stoichiometry. Because the changes in nitrogen and phosphorus in plant tissues are synergetic, fine-root total phosphorus and soil total nitrogen also show a significant correlation under the addition of inorganic nitrogen. Generally, the absorption of organic nitrogen by roots will be reduced only when the nitrogen demand of *Picea schrenkiana* is met; however, there may have been organic nitrogen saturation in the study area. Therefore, after applying inorganic nitrogen, the absorption capacity of inorganic nitrogen was enhanced instead of being inhibited, resulting in a situation in which *Picea schrenkiana* could use organic nitrogen but the overall absorption of inorganic nitrogen was more dominant, forming a more complete mechanism for the absorption and assimilation of inorganic nitrogen [43]. Carbon creates a parallel sink for nitrogen; after nitrogen application the carbon distribution pattern changes, and more carbon elements are used to maintain the growth of aboveground parts, resulting in the acceleration of forest growth, but the distribution proportion of underground organic carbon is relatively lower, resulting in a reduction in fine root organic carbon content. Forest soil ecosystems are more complex, especially in forest

ecosystems, and the biochemical properties of the interface between plant roots and soil are unique. The change in soil ecostochiometry is affected not only by nitrogen addition but also by the quantity and capacity of plant root exudates, the decomposition and turnover rate of litter, mycorrhizal quantity and interaction intensity, and the quantity as well as structure of microbial communities. The fine-root soil ecostochiometric relationship is a complex process of interactions in forest ecosystems.

The PLS model analysis of fine-root soil in this study found that, due to the change in fine-root order, there were differences in the changing amplitude of carbon and nitrogen among different root orders, and their correlation with environmental factors also changed to some extent. The effect of soil ecostochiometry on the ecostochiometry of low-order fine roots was greater in higher-order roots and was a step-by-step response. There are large differences in the ways in which fine roots of different root orders input carbon and nitrogen into the soil, leading to the difference in the correlation between the soil carbon, nitrogen, and phosphorus ecological stoichiometry and fine-root orders in the *Picea schrenkiana* forest. The first-order and second-order fine roots mainly secrete nutrients through the root system, while the third-order fine roots transport nutrients to the soil through apoptotic cells [44]. The change in the interpretation of soil nitrogen by first-order and second-order fine roots may be closely related to the morphology and function of different root orders of fine roots. As the root tip of the plant, first-order fine roots reach a higher level of nutrient absorption when oxygen-free elements are added; therefore, they are less sensitive to nutrient addition than the fine roots of the rest of the root order. Although bipolar roots are absorbing roots, they need more nutrients. As transport roots, second-order fine roots have a relatively poor absorption capacity, relatively lower nitrogen content, and stronger lignification; therefore, fine roots have a stepwise response to the ecological stoichiometry in soil.

5. Conclusions

In this study, the response of the fine-root soil ecological stoichiometry of *Picea schrenkiana* to atmospheric nitrogen deposition was measured through two years of in situ control experiments with different nitrogen sources, and the relationship between fine-root soil ecological stoichiometry and conditions of nitrogen addition was analyzed. The results showed that the soil ecological stoichiometry of *Picea schrenkiana* had different responses to different proportions of nitrogen source addition and had a stronger response to inorganic nitrogen. The response of fine-root ecostochiometry and its ratio to exogenous nitrogen inputs at different concentrations was different in different soil layers and root orders, but there was no significant difference in the plastic response of low-order and high-order fine-root chemometric traits to exogenous nitrogen addition. Different proportions of nitrogen source input have a positive impact on the fine-root stoichiometry and biomass of different root orders through an indirect impact on soil environmental factors and stoichiometry. Taken together, these results contribute to a better understanding of the mechanisms by which N addition affects fine-root soil element cycling in temperate forests. Our results emphasize that studies based on a single N source may underestimate the important effects of N deposition on temperate forest growth, and that it is necessary to consider the importance of different components of N deposition as well as the degree of preference for N component uptake by different plant species.

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