

Article

Ecological Stoichiometry of N and P across a Chronosequence of Chinese Fir Plantation Forests

Juan Cao ^{1,2}, Wende Yan ^{1,3,*}, Taimoor Hassan Farooq ^{1,4,*} , Xiaoyong Chen ^{3,5,*} , Jun Wang ^{1,3}, Chenglin Yuan ^{1,3} , Yaqin Qi ⁶ and Khalid Ali Khan ^{7,8}

- ¹ College of Life Science and Technology, Central South University of Forestry and Technology, Changsha 410004, China; caojuan0527@126.com (J.C.); jwang0829@vip.163.com (J.W.); yuanchenglin0424@gmail.com (C.Y.)
 - ² Institute of Agricultural Resources and Regional Planning, Chinese Academy of Agricultural Sciences, Beijing 100081, China
 - ³ National Engineering Laboratory for Applied Technology of Forestry and Ecology in Southern China, Central South University of Forestry and Technology, Changsha 410004, China
 - ⁴ Bangor College China, A Joint Unit of Bangor University and Central South University of Forestry and Technology, Changsha 410004, China
 - ⁵ College of Arts and Sciences, Governors State University, University Park, IL 60484, USA
 - ⁶ State Key Laboratory of Remote Sensing Science, Aerospace Information Research Institute, Chinese Academy of Sciences (AIRCAS), Beijing 100101, China; qiyq@aircas.ac.cn
 - ⁷ Unit of Bee Research and Honey Production, Research Center for Advanced Materials Science (RCAMS), King Khalid University, P.O. Box 9004, Abha 61413, Saudi Arabia
 - ⁸ Applied College, King Khalid University, Abha 61413, Saudi Arabia
- * Correspondence: csfuywd@hotmail.com (W.Y.); t.farooq@bangor.ac.uk (T.H.F.); xchen@govst.edu (X.C.)

Abstract: Ecological stoichiometry is crucial in understanding nutrient dynamics and its impact on plant growth and development at various ecological scales. Among the different nutrients, nitrogen (N) and phosphorus (P) have been widely recognized as key elements regulating substance transport, energy utilization, and ecosystem conversion. The N:P ratio in plants serves as a sensitive indicator of ecological processes, reflecting the availability and balance of these nutrients. Therefore, studying the ecological stoichiometry of N and P is essential for accurately assessing soil fertility and site productivity, particularly in forest ecosystems with low-fertility soils. In this study conducted in Huitong, Hunan province, southern China, the contents of N and P, as well as the N:P ratios, were investigated in plant-soil systems across four different aged stands of Chinese fir forests (3-, 8-, 18-, and 26-year-old stands). The results revealed varying concentrations of N and P in soils and foliage across the different plantations. Soil N concentrations increased by approximately 4%, 30%, and 22% in 8-, 18-, and 26-year-old plantations compared to the 3-year-old plantation. Soil P concentration was significantly higher in 8-, 18-, and 26-year-old plantations compared to the 3-year-old plantation. The average soil N:P ratio followed the order of 3-year-old plantation > 18-year-old plantation > 26-year-old plantation > 8-year-old plantation. Regarding foliage, both N and P contents exhibited a similar pattern across the different aged leaves, with current-year-old leaves having higher concentrations than 1-year-old, 2-year-old, and 3-year-old leaves in all four Chinese fir plantations. The study further established relationships between soil and foliage nutrient ratios. Soil N:P ratio was positively correlated with soil N content but negatively associated with soil P content. The foliage N:P ratio also showed a significant negative correlation between leaf N and foliage P content. These findings suggest that soil nutrient conditions improved with the aging of Chinese fir plantations, mainly due to increased inputs of above- and below-ground litter. Overall, this study provides valuable insights into the ecological stoichiometry of N and P in Chinese fir plantations, offering a scientific basis for sustainable forest management practices in southern China.

Keywords: *Cunninghamia lanceolata*; plantations; age sequence; macronutrients; substance transport; stoichiometry; sustainable management



Citation: Cao, J.; Yan, W.; Farooq, T.H.; Chen, X.; Wang, J.; Yuan, C.; Qi, Y.; Khan, K.A. Ecological Stoichiometry of N and P across a Chronosequence of Chinese Fir Plantation Forests. *Forests* **2023**, *14*, 1685. <https://doi.org/10.3390/f14081685>

Academic Editors: Timothy A. Martin and Choonsig Kim

Received: 22 May 2023

Revised: 28 July 2023

Accepted: 11 August 2023

Published: 21 August 2023



Copyright: © 2023 by the authors. Licensee MDPI, Basel, Switzerland. This article is an open access article distributed under the terms and conditions of the Creative Commons Attribution (CC BY) license (<https://creativecommons.org/licenses/by/4.0/>).

1. Introduction

Nitrogen (N) and phosphorus (P) are vital elements that often limit plant growth and development [1,2]. N plays a crucial role in amino and nucleic acid composition and actively participates in plants' essential processes, such as photosynthesis, protein synthesis, and enzyme biosynthesis [3]. P is a critical component of biological membranes and adenosine triphosphate (ATP), essential for substance transport, energy utilization, and conversion [4]. At the forest stand level, N and P significantly influence primary production, succession, ecological processes, and ecosystem services in forest ecosystems [5]. Therefore, the distribution patterns of N and P in plant biomass and their concentrations in different plant organs play a crucial role in various metabolic processes across different levels of the biological hierarchy [6]. Recent research has highlighted the importance of considering the stoichiometric relationship between N and P in plant organs and soils as a diagnostic indicator for assessing the nutrient status of an ecosystem [7]. Furthermore, on a global scale, the productivity of temperate and boreal forests is often limited by N, while tropical and subtropical evergreen forests commonly face P limitations [8].

Numerous studies have demonstrated the relationship between the N:P ratio in green foliage and soil nutrient availability, highlighting the internal linkages between nutrient demand and supply [1,5,6,9–12]. In response to low soil P availability, plants typically reduce foliage P concentration [13–15]. The biological characteristics of plant species contribute to the variations in nutrient requirements throughout different growth stages [16]. It is essential to consider that multiple factors, such as geographical regions, forest species, and vegetation, influence nutrient concentration, storage, and stoichiometry in plants and soils. N deposition has been observed to increase the N:P ratio in terrestrial plant ecosystems [17]. Additionally, soil N and P concentrations play a role in determining plant N and P concentrations to some extent. The composition of N and P in plants and their ratios result from complex interactions between plant species and their surrounding environment [10,18,19]. However, few studies have examined the patterns of N and P stoichiometry across a chronosequence of one plantation development.

Chinese fir (*Cunninghamia lanceolata*) is a resilient evergreen coniferous tree belonging to the cypress family Cupressaceae. It is indigenous to China, Vietnam (northern regions) and partially found in Laos and Cambodia. This species holds great significance as a timber resource in South China, playing a vital role in the country's forestry development and ecological stability [20]. Notably, Chinese fir exhibits robust growth characteristics and offers excellent raw materials. It thrives in adverse conditions, including poor and compacted clay soils, displaying exceptional drought tolerance. Except for extremely poorly drained soils, it adapts well to urban environments. Despite resembling conifer species in colder climates, it demonstrates high heat tolerance and can be cultivated at sea level in regions such as Hong Kong. Propagation of Chinese fir is commonly achieved through cuttings.

Forest structure, composition, and age group are vital for ecosystem nutrient cycling. The arrangement of vegetation influences nutrient uptake and retention, impacting soil fertility and ecosystem productivity. Diverse forest compositions contribute to a broader range of nutrient sources, enhancing nutrient availability and supporting sustainable nutrient cycling processes [21–24]. The primary objective of this project was to examine the variations in N and P stoichiometry in leaves and soil across Chinese fir forests of different ages. The specific goals were as follows: (1) to quantify patterns of leaf N and P concentration and N:P ratio in four distinct age groups of Chinese fir forests; (2) to analyze the changes in N-P stoichiometry as Chinese fir stands age; and (3) to establish relationships between N and P concentrations, N:P ratios in plant leaves, and corresponding soil measurements. Based on our hypothesis, we anticipate that the flexibility of N and P stoichiometry will exhibit contrasting trends as leaf age, root size, and stem growth increase. Typically, foliage N and P concentrations decline with longer growing seasons, while the N:P ratio increases in subtropical and tropical regions due to higher average temperatures.

By exploring the ecological stoichiometric flexibility, we aim to gain insights into material cycling and energy flow within the plantation forest ecosystem.

In this study, we only took N and P; the N:P ratio in plants is prioritized over the N:K or P:K ratio due to the crucial roles of N and P in plant growth and development. While potassium (K) is an essential nutrient involved in various physiological processes, it is typically more abundant in soils than N and P. Potassium deficiencies are relatively uncommon in most agricultural soils, emphasizing the greater significance of the N:P ratio for ensuring optimal plant growth [25]. Understanding the roles of N and P in plant physiology and their stoichiometric relationships is essential for comprehending nutrient limitations, ecological processes, and ecosystem productivity in diverse forest ecosystems. Incorporating these aspects helps evaluate plant ecosystems' nutrient state and management more effectively.

2. Materials and Methods

2.1. Overview of the Study Site

The study was conducted at Huitong Forest Ecology Station, a nationally recognized field observation station in Southern China. Situated approximately 580 km southwest of Changsha, Hunan Province, the station (109°45' E, 26°50' N) is known for its typical moist subtropical climate (Figure 1) [26]. The region experiences an average annual air temperature of 16.8 °C, an average annual precipitation of 1260 mm, and a mean annual relative humidity of 80%. The elevation ranges from 270 to 400 m, characterized by a relatively open topography with gentle slopes and some hills. The soil in the area is mountain yellow earth derived from discolored shale and sandstone, with a pH value ranging from 5 to 6. Specific soil characteristics, including bulk density and moisture content, are presented in Table 1. Soil bulk density was calculated by using the core method of the Nanjing Institute of Soil Science, and moisture content was calculated based on wet and dry weight [27].

$$\text{Bulk Density (g/cm}^3\text{)} = \text{Dry Soil Mass (g)}/\text{Soil Volume (cm}^3\text{)}$$

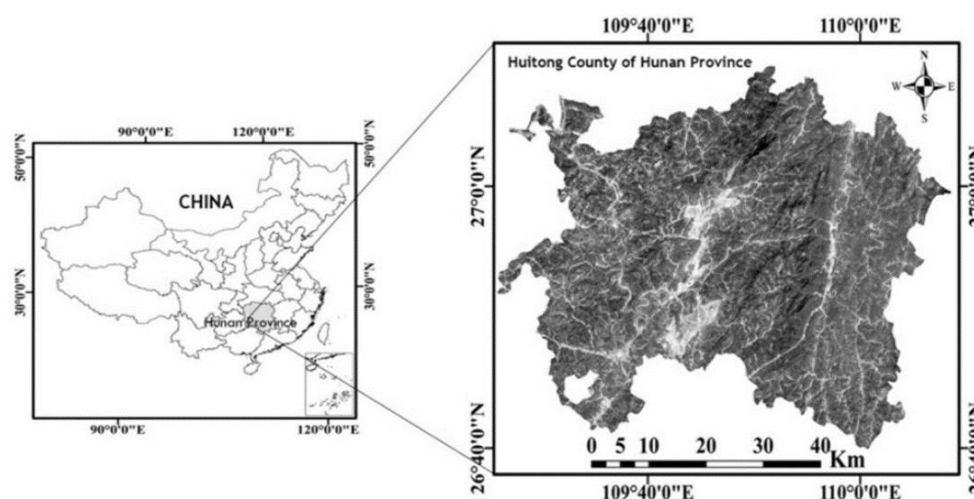


Figure 1. The map of the study area.

2.2. Experimental Design

At the research station, four Chinese fir plantations of different ages were chosen, including 3-, 8-, 18-, and 26-year-old stands. Following the initial planting, all seedlings underwent a three-year tending period to facilitate natural growth. For the study, three experimental plots measuring 20 m × 20 m each were carefully selected to represent each of the four aged Chinese fir stands, taking into account consistent soil type and site conditions. In each plot, various parameters, such as diameter at breast height (DBH), total

height, and canopy density, were measured for all trees. Detailed information regarding the four different-aged stands and the characteristics of the experimental plots can be found in Table 2.

Table 1. Soil bulk density and moisture content of the 3-, 8-, 18- and 26-year-old Chinese fir plantation stands. The average of the three experimental stands was used for each depth.

Stand Age ↓ Soil Depth (cm) →	Soil Bulk Density (g/cm ³)			Soil Moisture Content (%)		
	0–20	20–40	40–60	0–20	20–40	40–60
3	1.23	1.46	1.61	27.41	29.61	29.20
8	1.31	1.17	1.20	27.74	22.29	20.67
18	1.06	1.26	1.39	36.67	33.41	30.36
26	1.24	1.19	1.38	35.05	33.46	26.09
Mean	1.22	1.26	1.39	30.47	29.69	26.58

Table 2. Stand properties in the 3-, 8-, 18- and 26-year-old Chinese fir plantation stands.

Stand Age (Year)	Slope Aspect	Slope Gradient (°)	Canopy Density	Average DBH (cm)	Average Height (m)	Density (tree/hm ⁻²)
3	N	20	0.3	3.7	2.8	2500
8	N	28	0.7	6.8	5.6	2440
18	N	25	0.9	13.8	14.2	1825
26	N	30	0.8	17.1	16.0	1417

2.3. Foliage and Soil Sampling

Sampling was conducted using the mean sample method in forest measurement. From each plot, five standard sample trees that were healthy and mature were selected. Fresh leaf samples were collected based on leaf age. Firstly, fresh branches facing south were chosen from the lateral branches of the sample trees' canopies. Then, the leaves from the lateral branches were categorized into four groups: current year (less than 1-year-old), 1-year-old (between 1 and 2 years old), 2-years-old (between 2 and 3 years old), and 3-years-old (older than 3 years). Notably, in the 3-year-old stands, only the previous three leaf categories were collected. Subsequently, the samples from the same block of the five sample trees were thoroughly mixed, placed into plastic bags, and transported to the laboratory for chemical analysis.

Soil sampling was conducted in the selected plots where foliage was sampled. Three soil samples in each plot (12 for each age group stand) were randomly collected using a metal corer in each plot. The samples were obtained from three different soil depths: 0–20 cm, 20–40 cm, and 40–60 cm. The samples were carefully stratified and mixed at each sampling location to ensure thorough representation. Subsequently, the samples were transported back to the laboratory for further analysis.

The foliage and soil samples were transported to the laboratory for subsequent analyses. Debris and roots were meticulously removed by hand. The leaf samples were dried in an oven at 80 °C until a constant weight was achieved. The soil samples were air-dried at room temperature. Samples were crushed and sieved through 2 mm and 0.15 mm mesh sizes for further analysis [27].

The litterfall biomass and understory vegetation were notably higher in the older forest stands. We specifically chose sampling points and trees where the presence of shrubs around the tree canopy projection area was significantly lower than in the stands. This approach aimed to minimize the direct influence of the shrub layer. However, it is important to note that indirect effects may still be possible as field conditions exist [28].

2.4. Chemical Analysis

Foliage and soil N concentration was measured using the semi-micro Kjeldahl method. Foliage and soil P concentration was estimated using the colorimetric method (molybde-

num blue colorimetric method) on an optical Spectrophotometer after alkaline-digesting procedures before measurement.

2.5. Statistical Analysis

The statistical analyses were conducted using SPSS software (Version 18.0). N and P concentrations, as well as N:P ratios in foliage and soil across the four different aged stands of Chinese fir forests, were subjected to statistical testing. A one-way analysis of variance (ANOVA) was performed to assess the statistical differences between the various growth stages. Additionally, Pearson's correlation analyses were conducted to determine significant relationships between P and N concentrations in foliage and soil. In all statistical analyses, a significance level of $\alpha = 0.05$ was used. The figures were created using PAST 4.03 and Origin software 2020.

3. Results

The soil N concentration showed an average increase of approximately 4%, 30%, and 22% in the 8-, 18-, and 26-year-old stands, respectively, compared to the 3-year-old stand (Table 3). The soil N concentration was significantly higher in the 18- and 26-year-old stands compared to the 3- and 8-year-old stands ($p < 0.05$). However, there was no significant difference in soil N concentrations between the 18-year-old and 26-year-old stands or between the 3-year-old and 8-year-old stands. Significant variations in soil N concentrations among the different-aged plantations were observed in the deeper soil layers, whereas no significant differences were observed in N concentrations within the topsoil layer (0–20 cm) across all four stands ($p > 0.05$). Moreover, the N concentrations in the soil exhibited a decreasing trend with increasing soil depth in all four studied plantations (Table 3).

Table 3. Soil nitrogen (N) and phosphorus (P) concentrations at different soil depths in the 3-, 8-, 18- and 26-year-old Chinese fir plantation stands. Values are the mean $\pm$ SD of three replicates. Different letters indicate significant differences at the same soil depth among different aged plantations ($p < 0.05$).

Soil Depth	Stands			
	3-Year-Old	8-Year-Old	18-Year-Old	26-Year-Old
Soil N concentration ($\text{mg}\cdot\text{g}^{-1}$)				
0–20 cm	1.74 ± 0.23^a	1.75 ± 0.24^a	2.18 ± 0.33^a	1.89 ± 0.18^a
20–40 cm	1.21 ± 0.19^a	1.33 ± 0.16^{ab}	1.66 ± 0.29^b	1.54 ± 0.20^{ab}
40–60 cm	1.02 ± 0.19^a	1.05 ± 0.18^a	1.31 ± 0.15^{ab}	1.40 ± 0.16^b
Mean	1.32 ± 0.37^a	1.38 ± 0.36^a	1.72 ± 0.44^b	1.61 ± 0.27^b
Soil P concentration ($\text{mg}\cdot\text{g}^{-1}$)				
0–20 cm	0.23 ± 0.01^a	0.38 ± 0.03^b	0.39 ± 0.01^b	0.38 ± 0.03^b
20–40 cm	0.21 ± 0.01^a	0.36 ± 0.02^b	0.37 ± 0.04^b	0.35 ± 0.04^b
40–60 cm	0.21 ± 0.01^a	0.33 ± 0.01^b	0.33 ± 0.04^b	0.33 ± 0.04^b
Mean	0.21 ± 0.01^a	0.35 ± 0.03^b	0.36 ± 0.03^b	0.35 ± 0.04^b

The soil P concentration was significantly higher in the 8-, 18-, and 26-year-old stands compared to the 3-year-old stand ($p < 0.05$) (Table 3). This difference in soil P concentration was observed across all soil profile layers in the studied stands. However, there was no significant difference in soil P concentrations among the 8-, 18-, and 26-year-old stands. Overall, soil P concentration exhibited a decreasing trend with increasing soil depth in all examined plantations.

The average soil N:P ratio followed the order of 3-year-old > 18-year-old > 26-year-old plantation > 8-year-old stands (Table 4). There was no significant difference in the N:P ratio between the 18-year-old and 26-year-old stands ($p > 0.05$). Still, both of these stands had significantly lower N:P ratios than the 3-year-old stand and significantly higher N:P ratios than the 8-year-old stand ($p < 0.05$). Across the soil profile, the N:P ratio exhibited a decreasing trend with increasing soil depth in all studied plantations.

Table 4. Soil nitrogen/phosphorus (N:P) ratio of different aged foliage in the 3-, 8-, 18- and 26-year-old Chinese fir plantation stands ($\text{mg}\cdot\text{g}^{-1}$). Values are the mean $\pm$ SD of three replicates. Different letters indicate significant differences at the same soil depth among different aged plantations ($p < 0.05$).

Soil Depth	3-Year-Old	8-Year-Old	18-Year-Old	26-Year-Old
0–20 cm	7.70 ± 1.01^a	4.64 ± 0.40^b	5.62 ± 0.78^b	5.02 ± 0.19^b
20–40 cm	5.87 ± 1.00^a	3.74 ± 0.57^b	4.50 ± 0.53^b	4.45 ± 0.47^b
40–60 cm	4.87 ± 0.97^a	3.22 ± 0.43^b	3.94 ± 0.54^{ab}	4.25 ± 0.39^{ab}
Mean	6.14 ± 1.51^a	3.87 ± 0.74^b	4.69 ± 0.92^c	4.57 ± 0.47^c

In general, both the N and P foliage contents, as well as the N:P ratio, displayed a consistent pattern across different foliage ages in all four Chinese fir plantations (Figure 2). The current-year-old foliage exhibited significantly higher N and P concentrations compared to all other aged leaves in the studied stands ($p = 0.021$), except for foliage N in the 18-year-old stand, where it was almost equal to 1-year-old foliage. However, no significant differences were observed in foliage N and P concentrations among the 1-, 2-, and 3-year-old leaves in the four-aged stands ($p > 0.05$). On average, the foliage N concentration was $10.45 \text{ mg}\cdot\text{g}^{-1}$, $11.04 \text{ mg}\cdot\text{g}^{-1}$, $11.75 \text{ mg}\cdot\text{g}^{-1}$, and $11.71 \text{ mg}\cdot\text{g}^{-1}$ in the 3-, 8-, 18-, and 26-year-old stands, respectively. Correspondingly, the foliage P concentration and N:P ratio were $1.24 \text{ mg}\cdot\text{g}^{-1}$, $1.18 \text{ mg}\cdot\text{g}^{-1}$, $1.33 \text{ mg}\cdot\text{g}^{-1}$, and $1.30 \text{ mg}\cdot\text{g}^{-1}$, and 10.62, 10.32, 9.85, and 11.20 in the respective four-aged plantations (Figure 2).

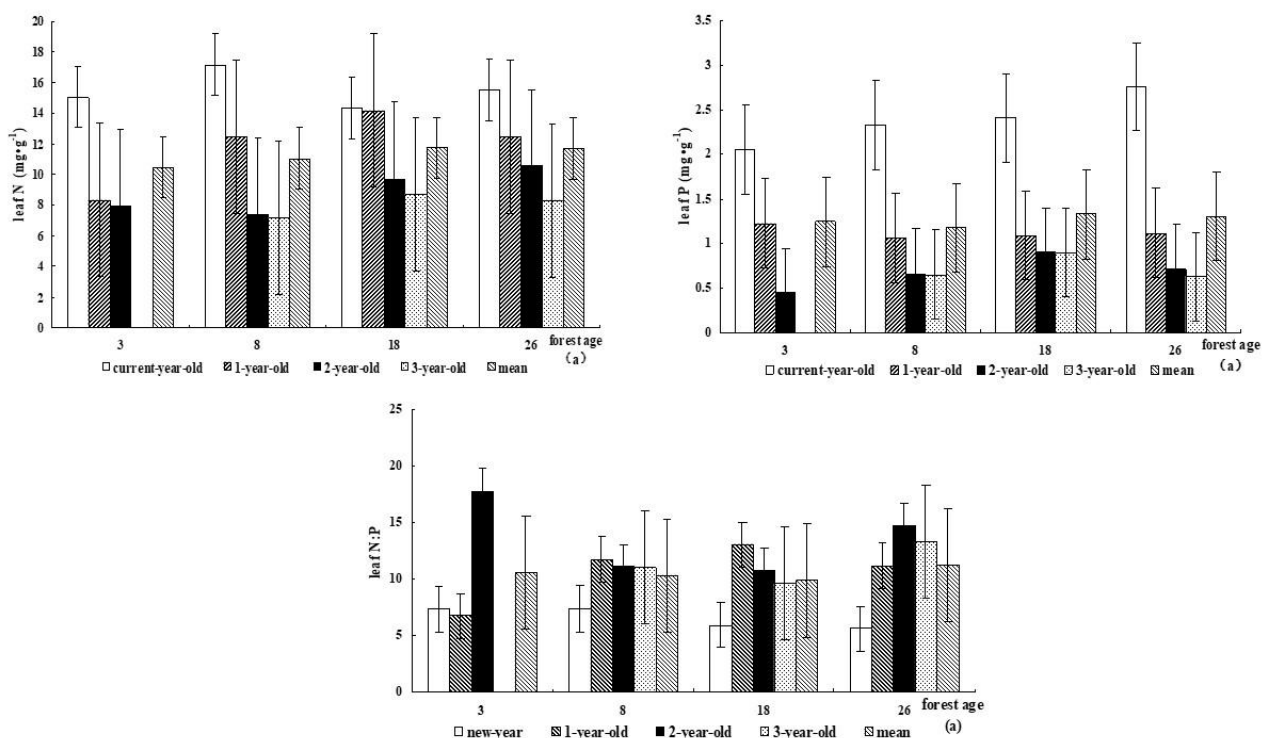


Figure 2. Foliage nitrogen (N), phosphorus (P) concentration, and N:P ratio in different aged in the 3-, 8-, 18- and 26-year-old Chinese fir plantation stands ($\text{mg}\cdot\text{g}^{-1}$). Values are the mean $\pm$ SD of three replicates. Leaves of the lateral branches were divided into 4 parts: current year (less than 1 year), 1-year-old (less than two years but more than one year), 2-years-old (less than 3 years but more than two years), 3-years-old (more than 3 years).

The soil N:P ratio exhibited a significant positive relationship with soil N content but a negative relationship with soil P content ($p = 0.01$). The foliage N:P ratio was negatively correlated with foliage N and leaf P content. Furthermore, a significant relationship was observed between foliage N and leaf P content in the studied stands ($p = 0.01$). However,

no significant correlation was found between soil N content and foliage N content, as well as between soil P content and foliage P content ($p > 0.05$) (Table 5).

Table 5. The correlation analysis of nitrogen (N) and phosphorus (P) contents and N:P ratios in the soil and plant foliage across all different-aged Chinese fir plantation stands.

	Soil N:P	Leaf N:P	Soil N	Leaf N	Soil P	Leaf P
Soil N:P	1	−0.128	0.513 **	0.074	−0.438 **	0.140
Leaf N:P		1	0.014	−0.466 **	0.184	−0.802 **
Soil N			1	−0.228	0.528 **	−0.168
Leaf N				1	−0.293	0.834 **
Soil P					1	−0.319
Leaf P						1

** Correlation is significant at the 0.01 level (2-tailed).

There were no significant correlations between soil bulk density and soil moisture content with any of the N and P parameters in the 3-, 8-, and 26-year-old stands. However, in the 18-year-old stand, a significant negative correlation was observed between soil bulk density and soil N content, while a significant positive correlation was found between soil moisture content and soil N content (Figure 3). Canopy density showed a significant positive correlation with soil P content and a significant negative correlation with foliar P content (Figure 4).

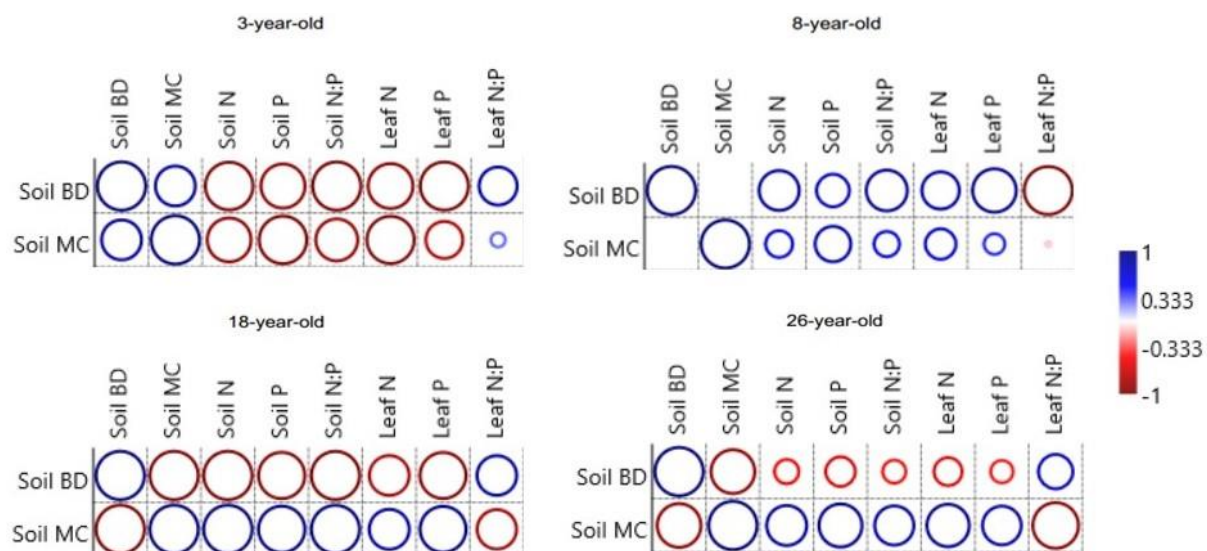


Figure 3. The correlation analysis of soil bulk density and soil moisture content with soil and foliage N and P parameters across all the soil depths in different-aged Chinese fir plantation stands. BD is bulk density and MC and moisture content.

Furthermore, the relationships among all the parameters used in the study, including stand characteristics, soil bulk density, soil moisture content, and soil and foliar N and P parameters at different soil depths (0–20 cm, 20–40 cm, and 40–60 cm), are depicted in Figure 5 for the different-aged Chinese fir stands.

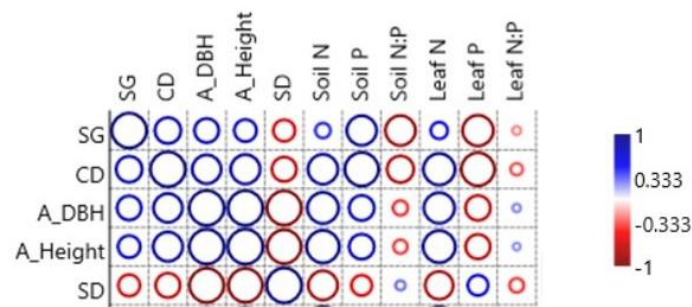


Figure 4. The correlation analysis between stand characteristics and N and P parameters across all different-aged Chinese fir plantation stands and soil depths. SG is slope gradient, CD is canopy density, A_DBH is average DBH, A_Height is average height, and SD is stand density.

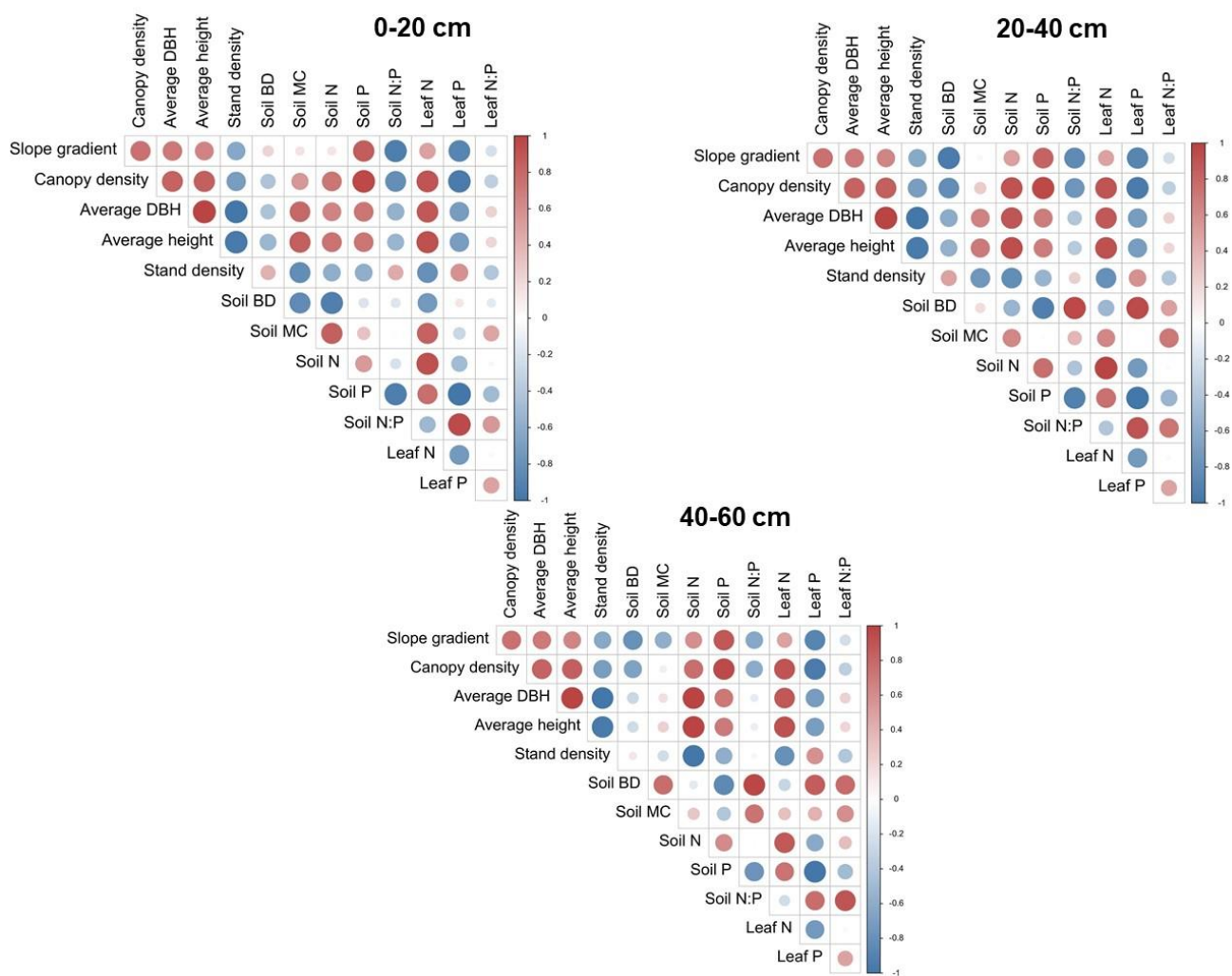


Figure 5. The correlation analysis among all the parameters used in the study (stand characteristics, soil bulk density, soil moisture content, and soil and foliar N and P parameters) in soil depths (0–20, 20–40, and 40–60 cm) across different-aged Chinese fir stands. DBH is the diameter at breast height; BD is bulk density, and MC is moisture content.

4. Discussion

This study observed an inverted pyramid distribution for N concentration and a cylinder distribution for P concentration in the soil profile. The contrasting distribution patterns of these two elements can be attributed to their different sources. N is typically made available in bioavailable forms through plant decomposition. In contrast, the primary

source of P in ecosystems is the weathering of rocks. As mentioned in the results, N concentrations increased from the 3-year-old stands to the 18-year-old stands, followed by a decrease. The peak N concentration was observed in the 18-year-old stands during the middle-aged forest stage. This trend aligns with the findings reported by Zhang et al. (2012) [29], further supporting the consistency of forest stage patterns.

In contrast, Sheng et al. [30] conducted a study on fir forest soils in different developmental stages in China and observed a decrease in N concentrations from young forest stages to middle-aged forest, followed by an increase. These variations may be attributed to differences in site characteristics such as climate, soil conditions, and sampling time. Regarding P concentration, the trend in forest stages aligned with that of N concentration. In the study area, P concentration ranged from 0.21 to 0.39 mg·g⁻¹, which is lower than the national average P content (0.56 mg·g⁻¹) reported by Han et al. [9] and the global average (2.8 mg·g⁻¹) reported by [31]. This suggests that soil P deficiency could be a limiting factor impacting the growth of Chinese fir forests. At the soil profile scale, P concentrations were highest in the surface soil layer (0–20 cm) and decreased vertically, which is consistent with the findings of [32], who proposed that water carries mobile P through the hydrologic continuum from leaf litter leachate to stream flow.

Soil N:P ratio can serve as a diagnostic indicator for assessing N nutrient limitation and saturation, indicating the availability of plant nutrients during growth. However, it is important to note that plants can also acquire N and P from sources such as old leaves and the atmosphere, which means that the soil N:P ratio may not fully reflect nutrient limitation. In our study, the average soil N:P ratios were 6.14, 3.87, 4.69, and 4.57 in the four aged stands. Cleveland and Liptzin [33] estimated the global soil N:P ratio in the surface soil (0–10 cm) to be 13.1, while Tian et al. [34] reported a number-weighted average soil N:P ratio of 5 for China. The soil N:P ratio in the 3-year-old stands was significantly higher than the other stands ($p < 0.05$) and exceeded the average soil N:P ratio in China. However, in the remaining stands, the soil N:P ratios were relatively lower compared to the findings of [34].

The significant difference between the 3-year-old stand and the others primarily resulted from the lower P concentrations in the 3-year-old stand compared to the other three ($p < 0.05$). Several factors have been proposed to explain this discrepancy. On the one hand, the soil N:P ratio is strongly influenced by stand characteristics. For example, in our study, the forest stands were relatively small, which resulted in insufficient shade canopy. Additionally, the combination of high temperature and precipitation in the area led to a high rate of P leaching, as observed by [34]. On the other hand, the young age of the forest created favorable environmental conditions, particularly concerning sufficient light availability. This, in turn, promoted the growth of heliophytes and weeds, contributing to a diverse range of plant species.

Previous research has indicated that foliar N:P ratios can vary depending on soil N and P availability, providing valuable insights into nutrient limitation and plant productivity in specific ecosystems and scales [32]. In this study, the average leaf N content in different growth stages of Chinese fir was found to be 11.29 ± 3.36 mg·g⁻¹, which was significantly lower than the average of 20.2 ± 8.4 mg·g⁻¹ across all 753 species in China [9]. Similarly, the average leaf P content in the four growth stages was 1.26 ± 0.75 mg·g⁻¹, which was lower than the average value reported in China (1.46 ± 0.99 mg·g⁻¹) [9].

In the study area, the foliage N:P ratio ranged from 5.63 to 17.74, with a mean value of 10.49 ± 3.44 , representing a variation of 32.79%. Notably, this average was lower than the arithmetic mean P of China's flora (16.3) and global vegetation (13.8). Several factors contribute to this lower mean foliage N:P ratio observed in this study. Firstly, the lower N levels, influenced by the overall N and P deficiency in China, affected the N:P ratio of the leaves. Additionally, the abundance of precipitation in the region made the soil available N more prone to leaching. Secondly, although N deposition is more severe in subtropical regions, the impact was minimal in the study area due to its location west of Hunan province, with fewer populations. Lastly, it is possible that soil degradation

resulting from rotation plantations, as supported by extensive research on Chinese fir plantations, may have contributed to the observed phenomenon.

Based on the critical N:P ratios of 14 and 16 for N and P limitation, respectively, as Koerselman and Meuleman [35] defined, the study area was primarily restricted by N. The leaf N:P ratio in the four stands decreased from the 3-year-old to the 18-year-old stands, followed by an increase. Therefore, in the middle-aged forest (18-year-old stands), which exhibited the lowest value, N was the limiting factor, while P deficiency was observed across all stands. This finding aligns with the research by Zheng et al. [36], who also reported N as a limiting factor in the growth of fast-growing Chinese fir plantations in subtropical regions with severe N deposition. The correlation analysis demonstrated a highly significant negative association between leaf N and leaf P content and leaf N:P ratio, indicating that changes in leaf N and P primarily influenced the leaf N:P ratio. Considering the earlier analysis, soil N emerged as the primary limiting factor compared to P in our study, which contradicts the findings of Aerts and Chapin [8], who suggested that forest communities in subtropical areas were mainly influenced by soil P for growth.

The initial results revealed significant correlations ($p < 0.05$) among the following traits: soil N, P, and soil N:P ratio, as well as foliage N, P, and N:P ratio. However, no significant relationship was found between the chemical traits of soil and foliage. Although there were similarities in the spatiotemporal dynamics of N and P in both soil and foliage, plant N and P absorption exhibited selectivity [36]. The lower foliage N:P ratio and N being the primary limiting factor suggested that the availability of N was relatively lower compared to P.

5. Conclusions

As stand age increased, no consistent pattern was observed in the soil and foliage N, P, and N:P ratios. The study indicated that N was the primary limiting factor in both the fast-growing phase and other stages of Chinese fir growth. Additionally, P was identified as another limiting factor due to its scarcity. Despite the severity of N deposition in subtropical regions, its impact was minimal in the study area, which was located in remote areas, resulting in insufficient N supply to meet the plant's needs. In line with the previous analysis, the availability of N was relatively lower compared to P. Interestingly, our findings showed no significant differences in foliage N, P, and N:P ratios among the four different-aged stands, suggesting a plant adaptation strategy to the environment. These results can play a crucial role in guiding targeted element supplementation and nutrient management in forest management practices.

Author Contributions: Conceptualization, J.C., T.H.F. and X.C.; methodology, J.C., J.W. and C.Y.; writing—original draft preparation, J.C., T.H.F. and Y.Q.; writing—review and editing, K.A.K., X.C. and W.Y.; funding, W.Y.; Supervision, W.Y. All authors have read and agreed to the published version of the manuscript.

Funding: This was a collaborative study sponsored by different projects, including (1) Joint Funds of the National Natural Science Foundation of China (U21A20187), (2) Forestry Industry Research Special Funds for Public Welfare projects (201404316), (3) Science Foundation of Hunan Province (No. [2013]7), and (4) Hunan Key Advantage Discipline Funds, One Hundred Plan Project of Hunan Province (0842).

Data Availability Statement: Data will be provided at formal request.

Acknowledgments: We thank the Department of Forestry, Huitong County, for granting us access to the research sites. Furthermore, we acknowledge the assistance of Fei Cui, Tiaoxiao Luo, Piaoyun Deng, and Song Yang in the field and laboratory. The authors extend their appreciation to the Deanship of Scientific Research at King Khalid University Saudi Arabia for funding this work through Large Groups Project under grant number RGP2/356/44. Authors also acknowledge the support of the Research Center for Advanced Materials Science (RCAMS) at King Khalid University Abha Saudi Arabia.

Conflicts of Interest: The authors declare that the research was conducted without commercial or financial relationship construed as a potential conflict of interest.

References

- Wang, W.; Peng, Y.; Chen, Y.; Lei, S.; Wang, X.; Farooq, T.H.; Liang, X.; Zhang, C.; Yan, W.; Chen, X. Ecological Stoichiometry and Stock Distribution of C, N, and P in Three Forest Types in a Karst Region of China. *Plants* **2023**, *12*, 2503. [\[CrossRef\]](#) [\[PubMed\]](#)
- Hu, Q.; Sheng, M.; Bai, Y.; Jie, Y.; Xiao, H. Response of C, N, and P stoichiometry characteristics of *Broussonetia papyrifera* to altitude gradients and soil nutrients in the karst rocky ecosystem, SW China. *Plant Soil* **2022**, *475*, 123–136. [\[CrossRef\]](#)
- Ding, P.; Noor, H.; Shah, A.A.; Yan, Z.; Sun, P.; Zhang, L.; Li, L.; Jun, X.; Sun, M.; Elansary, H.O.; et al. Nutrient Cycling and Nitrogen Management Impact of Sowing Method and Soil Water Consumption on Yield Nitrogen Utilization in Dryland Wheat (*Triticum aestivum* L.). *Agronomy* **2023**, *13*, 1528. [\[CrossRef\]](#)
- Sun, Y.; Wang, C.; Chen, X.; Liu, S.; Lu, X.; Chen, H.Y.H.; Ruan, H. Phosphorus additions imbalance terrestrial ecosystem C:N:P stoichiometry. *Glob. Chang. Biol.* **2022**, *28*, 7353–7365. [\[CrossRef\]](#)
- Farooq, T.H.; Li, Z.; Yan, W.; Shakoor, A.; Kumar, U.; Shabbir, R.; Peng, Y.; Gayathiri, E.; Alotaibi, S.S.; Wróbel, J.; et al. Variations in litterfall dynamics, C: N: P stoichiometry and associated nutrient return in pure and mixed stands of camphor tree and masson pine forests. *Front. Environ. Sci.* **2022**, *10*, 903039. [\[CrossRef\]](#)
- Yan, W.; Farooq, T.H.; Chen, Y.; Wang, W.; Shabbir, R.; Kumar, U.; Riaz, M.U.; Alotaibi, S.S.; Peng, Y.; Chen, X. Soil Nitrogen Transformation Process Influenced by Litterfall Manipulation in Two Subtropical Forest Types. *Front. Plant Sci.* **2022**, *13*, 923410. [\[CrossRef\]](#)
- Zhang, W.; Liu, W.; Xu, M.; Deng, J.; Han, X.; Yang, G.; Fen, Y.; Ren, G. Response of forest growth to C:N:P stoichiometry in plants and soils during *Robinia pseudoacacia* afforestation on the Loess Plateau, China. *Geoderma* **2019**, *337*, 280–289. [\[CrossRef\]](#)
- Aerts, R.; Chapin, F.S., III. The mineral nutrition of wild plants revisited: A re-evaluation of processes and patterns. In *Advances in Ecological Research*; Academic Press: Cambridge, MA, USA, 1999; Volume 30, pp. 1–67.
- Han, W.; Fang, J.; Guo, D.; Zhang, Y. Leaf nitrogen and phosphorus stoichiometry across 753 terrestrial plant species in China. *N. Phytol.* **2005**, *168*, 377–385. [\[CrossRef\]](#)
- Zhang, J.; Zhao, N.; Liu, C.; Yang, H.; Li, M.; Yu, C.; Wilcox, K.; Yu, Q.; He, N. C:N:P stoichiometry in China's forests: From organs to ecosystems. *Funct. Ecol.* **2018**, *32*, 50–60. [\[CrossRef\]](#)
- Wu, Y.; Jiang, J.; Chen, B.; Hu, Y. Nutrient resorption efficiency of three tree species in Beijing plain afforestation and its C:N:P stoichiometry. *Ann. For. Res.* **2020**, *63*, 91–102. [\[CrossRef\]](#)
- Fan, H.; Wu, J.; Liu, W.; Yuan, Y.; Hu, L.; Cai, Q. Linkages of plant and soil C:N:P stoichiometry and their relationships to forest growth in subtropical plantations. *Plant Soil* **2015**, *392*, 127–138. [\[CrossRef\]](#)
- Rashid, M.H.U.; Guo, H.; Zheng, S.; Li, L.; Ma, X.; Farooq, T.H.; Nawaz, M.F.; Gautam, N.P.; Wu, P. Effects of low phosphorus availability on root cambial activity, biomass production and root morphological pattern in two clones of Chinese fir. *Forestry* **2022**, *96*, 76–86. [\[CrossRef\]](#)
- Zou, X.; Liu, Q.; Huang, Z.; Chen, S.; Wu, P.; Ma, X.; Cai, L. Allocation of Phosphorus Fractions in Chinese Fir in Response to Low Phosphorus Availability Using ^{32}P Tracer. *Forests* **2022**, *13*, 1769. [\[CrossRef\]](#)
- Fyllas, N.M.; Patino, S.; Baker, T.R.; Bielefeld Nardoto, G.; Martinelli, L.A.; Quesada, C.A.; Paiva, R.; Schwarz, M.; Horna, V.; Mercado, L.M.; et al. Basin-wide variations in foliar properties of Amazonian forest: Physiology, soils and climate. *Biogeosciences* **2009**, *6*, 2677–2708. [\[CrossRef\]](#)
- Pang, Y.; Tian, J.; Zhao, X.; Chao, Z.; Wang, Y.; Zhang, X.; Wang, D. The linkages of plant, litter and soil C:N:P stoichiometry and nutrient stock in different secondary mixed forest types in the Qinling Mountains, China. *PeerJ* **2020**, *8*, e9274. [\[CrossRef\]](#)
- Zhao, M.; Xiang, W.; Tian, D.; Deng, X.; Huang, Z.; Zhou, X.; Peng, C. Effects of increased nitrogen deposition and rotation length on long-term productivity of *Cunninghamia lanceolata* plantation in southern China. *PLoS ONE* **2013**, *8*, e55376. [\[CrossRef\]](#)
- Chen, Y.H.; Han, W.X.; Tang, L.Y.; Tang, Z.; Fang, J. Leaf nitrogen and phosphorus concentrations of woody plants differ in responses to climate, soil and plant growth form. *Ecography* **2013**, *36*, 178–184. [\[CrossRef\]](#)
- Elser, J.J.; Bracken, M.E.; Cleland, E.E.; Gruner, D.S.; Harpole, W.S.; Hillebrand, H.; Ngai, J.T.; Seabloom, E.W.; Shurin, J.B.; Smith, J.E. Global analysis of nitrogen and phosphorus limitation of primary producers in freshwater, marine and terrestrial ecosystems. *Ecol. Lett.* **2007**, *10*, 1135–1142. [\[CrossRef\]](#)
- Wu, X.; Cao, Y.; Jiang, Y.; Chen, M.; Zhang, H.; Wu, P.; Ma, X. Dynamics of Non-Structural Carbohydrates Release in Chinese Fir Topsoil and Canopy Litter at Different Altitudes. *Plants* **2023**, *12*, 729. [\[CrossRef\]](#) [\[PubMed\]](#)
- Farooq, T.H.; Chen, X.; Shakoor, A.; Rashid, M.H.U.; Kumar, U.; Alhomrani, M.; Alamri, A.S.; Ravindran, B.; Yan, W. Unraveling the Importance of Forest Structure and Composition Driving Soil Microbial and Enzymatic Responses in the Subtropical Forest Soils. *Forests* **2022**, *13*, 1535. [\[CrossRef\]](#)
- Ma, X.; Heal, K.V.; Liu, A.; Jarvis, P.G. Nutrient cycling and distribution in different-aged plantations of Chinese fir in southern China. *For. Ecol. Manag.* **2007**, *243*, 61–74. [\[CrossRef\]](#)
- Wang, Z.; Yan, W.; Peng, Y.; Wan, M.; Farooq, T.H.; Fan, W.; Lei, J.; Yuan, C.; Wang, W.; Qi, Y.; et al. Biomass Production and Carbon Stocks in Poplar-Crop Agroforestry Chronosequence in Subtropical Central China. *Plants* **2023**, *12*, 2451. [\[CrossRef\]](#) [\[PubMed\]](#)
- Zhou, L.; Shalom, A.-D.D.; Wu, P.; He, Z.; Liu, C.; Ma, X. Biomass production, nutrient cycling and distribution in age-sequence Chinese fir (*Cunninghamia lanceolata*) plantations in subtropical China. *J. For. Res.* **2016**, *27*, 357–368. [\[CrossRef\]](#)
- Marschner, H. (Ed.) *Marschner's Mineral Nutrition of Higher Plants*; Academic Press: Cambridge, MA, USA, 2011.

26. Liu, Z.; Bai, Y.; Jiang, C.; Wang, S.; Meng, J. Species association of the dominant tree species in an old-growth forest and their implications for silvicultural practices in western Hunan Province, China. *Austrian J. For. Sci.* **2019**, *136*, 3.
27. Farooq, T.H.; Ma, X.; Rashid, M.H.U.; Wu, W.; Xu, J.; Tarin, M.W.K.; He, Z.; Wu, P. Impact of stand density on soil quality in Chinese fir (*Cunninghamia lanceolata*) monoculture. *Appl. Ecol. Environ. Res.* **2019**, *17*, 3553–3566. [[CrossRef](#)]
28. Farooq, T.H.; Chen, X.; Shakoob, A.; Li, Y.; Wang, J.; Rashid, M.H.U.; Kumar, U.; Yan, W. Unraveling the Influence of Land-Use Change on $\delta^{13}C$, $\delta^{15}N$, and Soil Nutritional Status in Coniferous, Broadleaved, and Mixed Forests in Southern China: A Field Investigation. *Plants* **2021**, *10*, 1499. [[CrossRef](#)]
29. Zhang, Z.; Zhang, J.; Slik, J.F.; Cao, K. Leaf element concentrations of terrestrial plants across China are influenced by taxonomy and the environment. *Global Ecol. Biogeogr.* **2012**, *21*, 809–818. [[CrossRef](#)]
30. Sheng, W.T.; Yang, C.D.; Fan, S.H. Variation of Soil Properties of Chinese Fir plantation. *For. Res.* **2003**, *16*, 377–385.
31. Ren, S.J.; Yu, G.R.; Tao, B.; Wang, S.Q. Leaf nitrogen and phosphorus stoichiometry across 654 terrestrial plant species in NSTEC. *Environ. Sci.* **2007**, *28*, 2665–2673.
32. Reed, S.C.; Townsend, A.R.; Davidson, E.A.; Cleveland, C.C. Stoichiometric patterns in foliar nutrient resorption across multiple scales. *N. Phytol.* **2012**, *196*, 173–180. [[CrossRef](#)]
33. Cleveland, C.C.; Liptzin, D. C: N: P stoichiometry in soil: Is there a “Redfield ratio” for the microbial biomass? *Biogeochemistry* **2007**, *85*, 235–252. [[CrossRef](#)]
34. Tian, H.; Chen, G.; Zhang, C.; Melillo, J.M.; Hall, C.A. Pattern and variation of C: N: P ratios in China’s soils: A synthesis of observational data. *Biogeochemistry* **2012**, *98*, 139–151. [[CrossRef](#)]
35. Koerselman, W.; Meuleman, A.F. The vegetation N: P ratio: A new tool to detect the nature of nutrient limitation. *J. Appl. Ecol.* **1996**, *33*, 1441–1450. [[CrossRef](#)]
36. Zheng, L.J.; Huang, Z.Q.; He, Z.M.; Wang, X.Y.; Liu, Z.M.; Liu, R.Q.; Xiao, H.Y. Influence of forest and foliar ages on the composition of stable carbon and nitrogen isotope of *Cunninghamia lanceolata* in subtropical China. *Sci. Silvae Sin.* **2015**, *51*, 22–28.

Disclaimer/Publisher’s Note: The statements, opinions and data contained in all publications are solely those of the individual author(s) and contributor(s) and not of MDPI and/or the editor(s). MDPI and/or the editor(s) disclaim responsibility for any injury to people or property resulting from any ideas, methods, instructions or products referred to in the content.