



Effects of tree size and organ age on variations in carbon, nitrogen, and phosphorus stoichiometry in *Pinus koraiensis*

YanJun Wang^{1,2,3} · Guangze Jin^{1,2,3} · Zhili Liu^{1,2,3}

Received: 16 September 2023 / Accepted: 17 October 2023
© The Author(s) 2024

Abstract Carbon (C), nitrogen (N), and phosphorus (P) are of fundamental importance for growth and nutrient dynamics within plant organs and deserve more attention at regional to global scales. However, our knowledge of how these nutrients vary with tree size, organ age, or root order at the individual level remains limited. We determined C, N, and P contents and their stoichiometric ratios (i.e., nutrient traits) in needles, branches, and fine roots at different organ ages (0–3-year-old needles and branches) and root orders (1st–4th order roots) from 64 *Pinus koraiensis* of varying size (Diameter at breast height ranged from 0.3 to 100 cm) in northeast China. Soil factors were also measured. The results show that nutrient traits were regulated by tree size, organ

age, or root order rather than soil factors. At a whole-plant level, nutrient traits decreased in needles and fine roots but increased in branches with tree size. At the organ level, age or root order had a negative effect on C, N, and P and a positive effect on stoichiometric ratios. Our results demonstrate that nutrient variations are closely related to organ-specific functions and ecophysiological processes at an individual level. It is suggested that the nutrient acquisition strategy by younger trees and organ fractions with higher nutrient content is for survival. Conversely, nutrient storage strategy in older trees and organ fractions are mainly for steady growth. Our results clarified the nutrient utilization strategies during tree and organ ontogeny and suggest that tree size and organ age or root order should be simultaneously considered to understand the complexities of nutrient variations.

Project funding: The work was financially supported by the National Key R&D Program of China (2022YFD2201100), Natural Science Foundation of Heilongjiang Province of China (TD2023C006), and the Fundamental Research Funds for the Central Universities (2572022DS13).

The online version is available at <http://www.springerlink.com>.

Corresponding editor: Yu Lei.

Supplementary Information The online version contains supplementary material available at <https://doi.org/10.1007/s11676-024-01705-x>.

✉ Zhili Liu
liuzl2093@126.com

¹ Center for Ecological Research, Northeast Forestry University, Harbin 150040, People's Republic of China

² Key Laboratory of Sustainable Forest Ecosystem Management-Ministry of Education, Northeast Forestry University, Harbin 150040, People's Republic of China

³ Northeast Asia Biodiversity Research Center, Northeast Forestry University, Harbin 150040, People's Republic of China

Keywords Tree size · Organ age (or root order) · Carbon (C) · Nitrogen (N) · Phosphorus (P) · *Pinus koraiensis*

Introduction

Carbon (C), nitrogen (N), and phosphorus (P) are crucial elements that play significant roles at individual to community levels in plant growth, biochemical functioning, and various ecophysiological processes, including photosynthesis, energy flow, and nutrient cycling (Sternner and Elser 2002; Wright et al. 2004). The ratios of C:N, C:P and N:P are indicators of plant nutrient use efficiency, and relative nutrient limitations in terrestrial ecosystems are frequently used as input parameters in some ecological models (Ågren 2004; Zhang et al. 2019). Therefore, the study of nutrient traits (i.e., C, N, P, C:N, C:P and N:P ratios) can aid in the development of a more mechanistic understanding of

nutrient utilization, plant ontogenetic strategies, and ecosystem structure and function (Ågren 2008; Tang et al. 2018).

As ontogeny (i.e., whole-tree development) progresses, large trees due to their height advantage are able to capture most of the light resources in forests, resulting in significant light limitations for smaller understory trees (Martínez-Vilalta et al. 2007). This likely implies a considerable role of tree size for nutrient utilization strategies. Previous studies have confirmed that size is a significant regulator of nutritional traits, but only based on leaf nutrient studies, such as in evergreen (He and Yan 2018) and deciduous species (Seddall and Reich 2013), where leaf N content increased with tree size. No directional shift has been reported between leaf P contents and height at an individual level (He and Yan 2018). Leaf C, N and C:N ratios varied significantly with diameter at breast height (DBH) in two dominant rainforest tree species (Martin and Thomas 2013). Apparently, tree size modulates the underlying strategies for nutrient utilization (Anderson-Teixeira et al. 2016; Kuusk et al. 2018a), but there was no consistent pattern for the effect of tree size. In addition, the roles of different organs were not considered in the studies mentioned. There was no rounded system of nutrient variation at the whole-plant level, and our study aims to fill this information gap.

To date, several studies have shown that plant nutrients are allocated to satisfy the demands of specific organ functions (Minden and Kleyer 2014; He et al. 2015; Xiong et al. 2021). For instance, leaves are photosynthetic organs and play a key role in capturing light and fixing C (Wright et al. 2004). Branches provide mechanical support, hydraulic pathways for transporting resources (Fortunel et al. 2014). Roots are underground organs used to absorb nutrients and water from the environment (McCormack et al. 2015). Therefore, studies at the individual level, which include the major plant organs rather than for a single organ, can provide a better foundation for understanding nutrient allocation strategies and the cooperative mechanism between organs.

More importantly, organ age (needles, branches and fine roots) has been neglected in these studies, despite a potential functional divergence within organs on nutrient variations (Ågren 2008; Kuusk et al. 2018a). Surprisingly, a comprehensive consideration of nutrient variation among the major organs of 49 tree species by Li et al. (2010) found that nutrient traits significantly decreased with increasing twig and root order across all species. Thus, additional studies are needed to determine nutrient variations within organs at an individual level, and to examine underlying allocation strategies. Ecologists, according to specific physiological functions and nutrient utilization strategies of roots, divide fine roots into two broad groups: absorptive roots (mainly 1st–3rd) and transport roots (mainly 4th and 5th) (Pregitzer 2002; McCormack et al. 2015). Whether nutrient-related functional divergence exists in needles and branches within

organ age classes is not clear, suggesting that it is necessary to consider nutrient variation patterns across organ ages and root orders. This research will aid understanding of the inherent trade-off mechanisms of nutrients within organs.

Nutrient traits in different plant organs significantly vary with environmental factors, especially for soil directly in contact with root systems (Li et al. 2018). Zhao et al. (2016) explored nutrient variations in major plant organs across several Chinese forest ecosystems and found that soil N and P contents have a significant influence on these nutrients in leaves and fine roots. However, Liu et al. (2013) indicated that soil nutrient levels were not the main driver for leaf N and P content in a shrub biome transect. However, these studies were conducted at regional scales; it is necessary to determine whether soil nutrient content affects nutrient variations for a given tree species at a local scale.

Pinus koraiensis Siebold & Zucc. is the most important component in mixed broadleaved-Korean pine forests of northeastern China. Determining nutrient differences in needle, branch and fine root throughout tree development, organ age and root order is a crucial pathway for understanding plant nutrient use strategies. In this study, nutrient traits were measured in needles, branches and fine roots with different organ ages or root orders of variable sized *P. koraiensis*. Soil properties were also determined to relate the importance of tree size, organ age and root order on nutrient traits. Specifically, the questions were: (1) Are tree size, organ age or root order the main drivers of plant nutrient traits rather than soil factors? (2) At a whole plant level, are the variations in plant nutrients tightly related to organ-specific functions? and (3) Is there a common nutrient strategy throughout the tree and organ development process?

Materials and methods

Field work was carried out in a 9-ha old-growth (about 300 years old) mixed broadleaved-Korean pine forest permanent dynamic monitoring plot in Liangshui National Nature Reserve (47°10'50" N, 128°53'20" E), Heilongjiang Province, northeastern China. This area has a typical temperate climate, with mean annual temperature and precipitation of 0.3 °C and 676 mm, respectively. More than 60% precipitation falls between May and August, and the area is snow-covered from December to April (Liu et al. 2019). *P. koraiensis* is the principal tree species with average DBH 30 cm. The main companion tree species are *Acer pictum* subsp. *Mono* (Maxim.) H. Ohashi, *Betula costata*, and *Acer ukurunduense*.

To methodically unveil the importance of tree size, organ age or root order, and soil factors on plant nutrient variations, 64 healthy *P. koraiensis* individuals of various sizes (0.3–100 cm DBH; Table 1) were selected. Two sample

Table 1 Statistical information for diameter at breast height (DBH), tree height and soil factors

Factor	Minimum	Maximum	Mean (SD)	CV (%)
DBH (cm)	0.3	100	41.4 (30.7)	74.1
Height (m)	0.5	32.9	18.7 (9.2)	49.1
Soil water content (g g ⁻¹)	0.3	1.3	0.7 (0.2)	37.6
Soil C content (mg g ⁻¹)	42.8	270.8	119.2 (52.0)	43.6
Soil N content (mg g ⁻¹)	3.2	13	6.9 (2.3)	33.8
Soil P content (mg g ⁻¹)	0.3	1.8	0.9 (0.4)	42.3

SD Standard deviation, CV Coefficient of variation

seedlings DBH < 1.0 cm (Vernier calipers were used to measure the DBH with an accuracy of 0.01 mm), 30 sample trees $1.0 \leq \text{DBH} \leq 30$ cm, (the most similar sized trees had a DBH variation of about 1 cm), and 32 sample trees $30 < \text{DBH} \leq 100$ cm (the most similar sized trees had a DBH variation of about 2 cm) were selected (Liu et al. 2020).

Sampling was carried out in mid-October 2018, until growth of new needles and branches finished. An upper south side branch of the one-third canopy, which carried all needle age classes was randomly selected and removed. Needle and branch age classes were determined by the existence of polycyclic shoots, and the degree of darkness and hardness of needles and branches (Eimil-Fraga et al. 2015). Needles and branches from the current year were designated as being 0-year old, those from the previous year were 1-year old, and so on. Needles and branches were collected, based on their age, packaged, and transported to the laboratory.

For sampling fine roots, soil and other impurities surrounding the root system were removed, and an area near each tree base was excavated to expose main lateral roots. To confirm the specific tree, root branches were traced back to the trunk, and then removed from the main lateral roots, packaged and transported to the laboratory. Subsequently, deionized water was used to wash away residual soil and tweezers used to pick out any impurities. Fine roots < 2 mm in diameter were categorized using the classification of Pregitzer (2002). First order roots were identified as the most distal roots, second order roots from the intersection of two first order roots, up to fourth order roots. Roots of the same order were packaged together for chemical analysis.

Three soil subsamples were collected from the base of each tree with an angle of 120° between any two sampling directions. Soil collection was at the base of each tree, and ranged from 0 to 10 cm using a 5 cm diameter soil corer (Yang et al. 2019). The three subsamples were mixed together, impurities removed (Pregitzer 2002), sealed and taken to the laboratory for air drying.

Needles, branches, fine roots, and air-dried soil were oven dried at 65 °C until reaching a consistent mass (precision of 0.0001 g) prior to chemical analysis. The samples were crushed and passed through a 0.149 mm mesh and placed in envelopes.

C contents in plants and soil were measured by a Multi CN/3000 analyzer (Analytic Jena AG, Germany). N and P contents were determined by an AQ400 automatic discontinuous chemical analyzer (SEAL Analytical, Mequon, USA) after concentrated H₂SO₄ digestion. The determination of soil water content (g g⁻¹) was conducted using a drying oven (Wang et al. 2016). C, N and P levels were quantified in mg g⁻¹, and stoichiometric ratios determined based on mass.

A one-way ANOVA was conducted, with Duncan's post hoc multiple comparisons ($p < 0.05$), to contrast variations in nutrient traits across different organs. The mean \pm standard error (SE) presents the results. Differences in nutrient traits among organs was evaluated by the coefficients of variation (CV). The higher stability was expressed as lower CV values, and vice versa.

Generalized linear models (GLMs) quantified the relative importance of tree size (DBH or height), organ age or root order, and soil factors for nutrient traits among organs. In order to confirm the best descriptive parameter for size, the lower Akaike information criterion (AIC) values of the linear regressions between DBH or height against each nutrient trait were selected (Table S1). Burnham and Anderson (2004) demonstrated that, when the differences of AIC values of both models were smaller than 2.0, indicates models with comparatively equal degree of support and are indistinguishable from one another. These principles guided the selection of the descriptive parameter of size for each organ. To avoid losing precision when estimating regression coefficients, the variance inflation factor (VIF) was calculated for each pair of bivariate independent variables to assess the degree of collinearity among the parameters. The variance inflation factors (VIFs) for the independent variables (Tables S2, S3, and S4) were less than 10. This suggests that there was no significant multicollinearity among the independent variables, allowing us to retain all of them for further analysis (Chatterjee and Price 1991).

The scatterplots show the relationship in nutrient traits against tree size, organ age or root order among different organs by a standardized major axis (SMA) estimation method (Warton et al. 2006). A likelihood ratio test was employed to assess the variability of the regression slope (Warton and Weber 2002). If there was no significant difference between slopes, a common slope was employed.

In order to avoid the influence of DBH or height on organ age or root order, the 64 *P. koraiensis* trees were divided into three DBH groups: group I (0–10 cm), group II (11–50 cm), and group III (51–100 cm). Similarly, they were also divided into three height groups: group I (0–10 m),

group II (11–20 m), and group III (> 20 m) (Liu et al. 2020). All statistical analysis used R-4.0.3 (R Core Team 2020), and the data were \log_{10} -transformed for normalization before analysis.

Results

The nutrient traits differed considerably among organs for the 64 *P. koraiensis* trees. C content in branches was markedly higher than in needles or fine roots, and needle N and P contents were considerably greater than in branches or fine roots ($p < 0.05$; Table 2). The ratios of C:N and C:P followed the same pattern: branch > fine root > needle. N:P ratios were not different in needles and branches, but markedly higher

than in fine roots ($p < 0.05$; Table 2). Additionally, needles had the smallest CV (%) of nutrient traits among organs, and C content the smallest among nutrient traits (Table 2). Nutrient traits among organs were principally influenced by size, organ ages or root orders and not by soil factors ($p < 0.05$; Table 3). Therefore, soil properties were not considered as important factors in this investigation.

Excluding C content and C:N ratios, needle nutrient traits were significantly related to DBH in different organ age groups ($p < 0.05$; Fig. 1; Table S5). With increasing DBH, needle N increased in 1-year-old needles and decreased in 3-year-old needles ($p < 0.05$; Fig. 1; Table S5). DBH had a negative influence on P for most needle age groups and a positive influence on C:P and N:P ratios. A common slope of 0.002 was used for N:P to

Table 2 Carbon (C), nitrogen (N), and phosphorus (P) content and their ratios in needles, branches, and fine roots of *Pinus koraiensis*

Nutrient	Needle		Branch		Fine root	
	Mean \pm SE	CV (%)	Mean \pm SE	CV (%)	Mean \pm SE	CV (%)
C (mg g ⁻¹)	498.5 \pm 0.92B	2.9	536.3 \pm 1.02A	3.0	476.8 \pm 0.94C	3.06
N (mg g ⁻¹)	16.6 \pm 0.16A	15.4	9.2 \pm 0.23B	37.9	9.0 \pm 0.23B	39.4
P (mg g ⁻¹)	2.1 \pm 0.03A	18.9	1.2 \pm 0.02C	29.4	1.4 \pm 0.03B	34.0
C:N	30.9 \pm 0.35C	17.9	75.0 \pm 3.58A	75.4	61.7 \pm 1.59B	39.8
C:P	252.1 \pm 3.18C	19.6	518.7 \pm 13.29A	39.9	375.7 \pm 8.18B	33.7
N:P	8.2 \pm 0.09A	16.7	8.6 \pm 0.28A	50.5	6.4 \pm 0.09B	21.9

Different letters in the same row indicate significant differences between organs ($p < 0.05$)
SE Standard error; CV Coefficient of variation

Table 3 Influence of size (DBH or height), organ age, root order, and soil factors on needle, branch, and fine root nutrient traits using a GLM

Organ	Nutrient	Tree size (DBH or tree height)	Organ age (or root order)	Soil water content	Soil C content	Soil N content	Soil P content	Intercept
Needle	C	-0.048	0.003	-0.097	0.024	0.030	0.083	-0.008
	N	0.074	-0.622***	0.038	-0.201*	0.163*	-0.033	1.512***
	P	-0.370***	-0.567***	-0.090	0.133	-0.077	0.172	1.379***
	C:N	-0.046	0.617***	-0.061	0.217**	-0.204**	0.100	-1.501***
	C:P	0.301***	0.564***	0.069	-0.069	0.005	-0.076	-1.372***
	N:P	0.438***	0.043	0.132	-0.298**	0.216*	-0.197*	-0.105
Branch	C	0.114	-0.113*	0.115	-0.082	0.222*	-0.071	0.275
	N	0.492***	-0.092	0.029	0.032	0.008	-0.039	0.223
	P	0.043	-0.444***	-0.048	0.035	0.122	-0.116	1.081***
	C:N	-0.413***	0.066	0.055	-0.003	-0.090	0.119	-0.162
	C:P	-0.116	0.378***	0.021	-0.008	-0.197*	0.229*	-0.920***
	N:P	0.2923***	0.210***	0.064	0.013	-0.190	0.162	-0.511***
Fine root	C	0.290***	0.165**	-0.068	0.220*	-0.206*	-0.049	-0.397**
	N	-0.075*	-0.798***	0.046	-0.131*	0.140**	0.051	1.919***
	P	-0.191***	-0.720***	-0.008	0.053	-0.023	0.179**	1.732***
	C:N	0.051	0.787***	-0.040	0.148**	-0.163**	-0.052	-1.894***
	C:P	0.197***	0.725***	-0.025	-0.002	-0.030	-0.082	-1.744***
	N:P	0.178**	-0.339***	0.110	-0.316**	0.232*	-0.089	0.815***

* $p < 0.05$, ** $p < 0.01$, *** $p < 0.001$

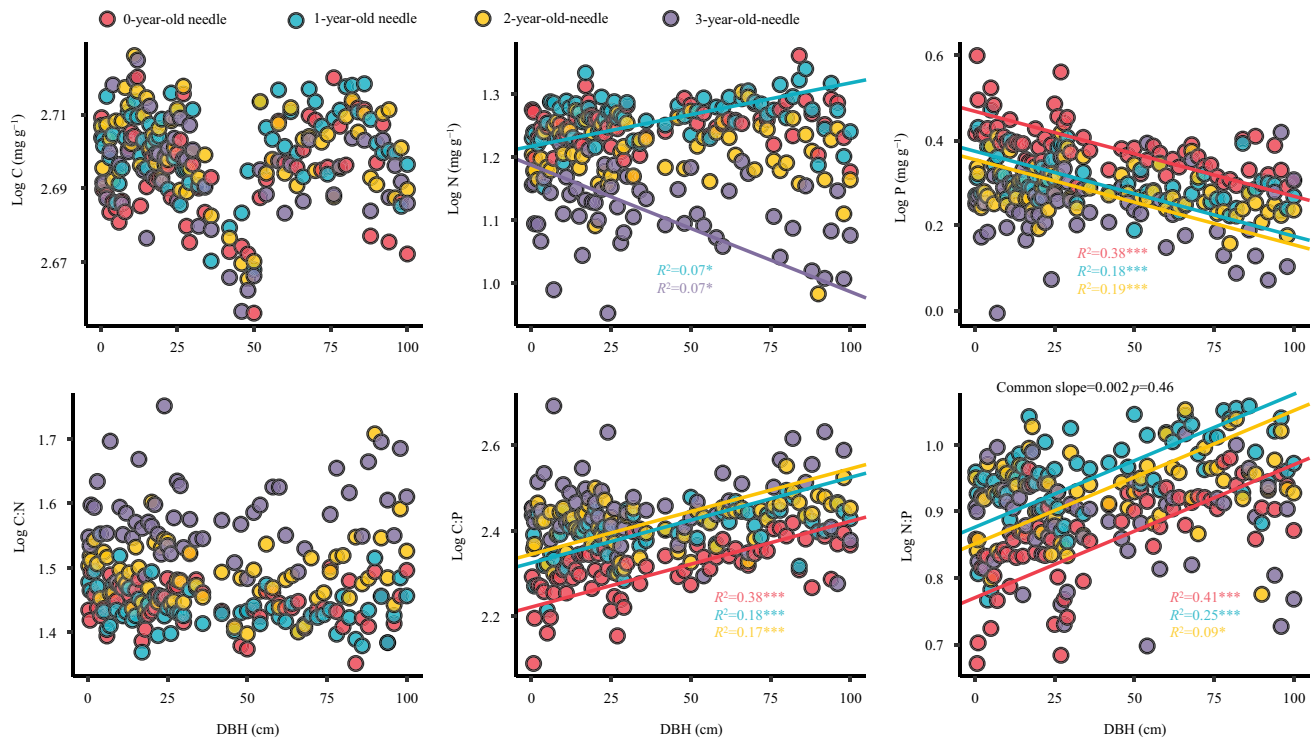


Fig. 1 Variation in nutrient traits in different needle age and DBH groups for *Pinus koraiensis*. Different colored data points represent 0-year-old, 1-year-old, 2-year-old and 3-year-old needles, respec-

tively. The trend line and regression information are marked in the figure as significant. * $p < 0.05$, ** $p < 0.01$, *** $p < 0.001$

DBH. In addition, the newest needles were more closely related to DBH than other needle age groups (Fig. 1).

For branches, C, N contents and N:P ratios increased with height for most branch age groups, and C:N ratios decreased significantly. There were no significant trends for P and C:P ratios ($p < 0.05$; Fig. 2; Table S5).

Root nutrient traits were positively correlated with height in most individuals ($p < 0.05$; Fig. 3; Table S5). The common slopes of 0.001, -0.009 and 0.009 were used for the relationships of C, P and C:P ratios against height, respectively ($p < 0.05$; Fig. 3; Table S5).

Nutrient traits were significantly correlated with organ age or root order and consistent among organs across different size groups ($p < 0.05$; Figs. 4–6; Table S6). In general, all nutrient traits increased with organ age or root order except for C. Common slopes of 0.064 were used for the relationships of needle C:N ratios and age ($p < 0.05$; Fig. 4; Table S6), common slopes of -0.128 and 0.128 for the relationship of branch P content and age, and branch C:P ratios, respectively ($p < 0.05$; Fig. 5; Table S6); common slopes of -0.131 , 0.133 and -0.086 were employed for the relationships of P, and C:P and N:P ratios against root order, respectively ($p < 0.05$; Fig. 6; Table S6). The effects of organ age or root order for fine root nutrient

traits were greater than for needles and branches in all size groups ($p < 0.05$; Figs. 4–6; Table S6).

Discussion

In this study, there were significant variations in nutrient traits among organs of 64 *P. koraiensis* trees in northeastern China (Table 2). Branches contained the most C content, needles the most N and P. Stoichiometric ratios shared a general pattern in most samples, branch > fine root > needle (Table 2). These findings agree with previous reports at an individual level (Zhang et al. 2020), a community level (Yan et al. 2016), and even at global scales (Zhang et al. 2018b), suggesting organ nutrient variations are strongly regulated by organ-specific functions.

Needles are photosynthetic organs with higher N and P requirements to produce a range of enzymes necessary for photosynthesis and to meet the needs of tree development (Sternier and Elser 2002; Zhang et al. 2018a). Branches are structural support organs with higher carbon demands to construct structural tissues, to support new shoot development and for nutrient transport (Xiong et al. 2021). Fine roots, as the below-ground analogue of leaves, connect with

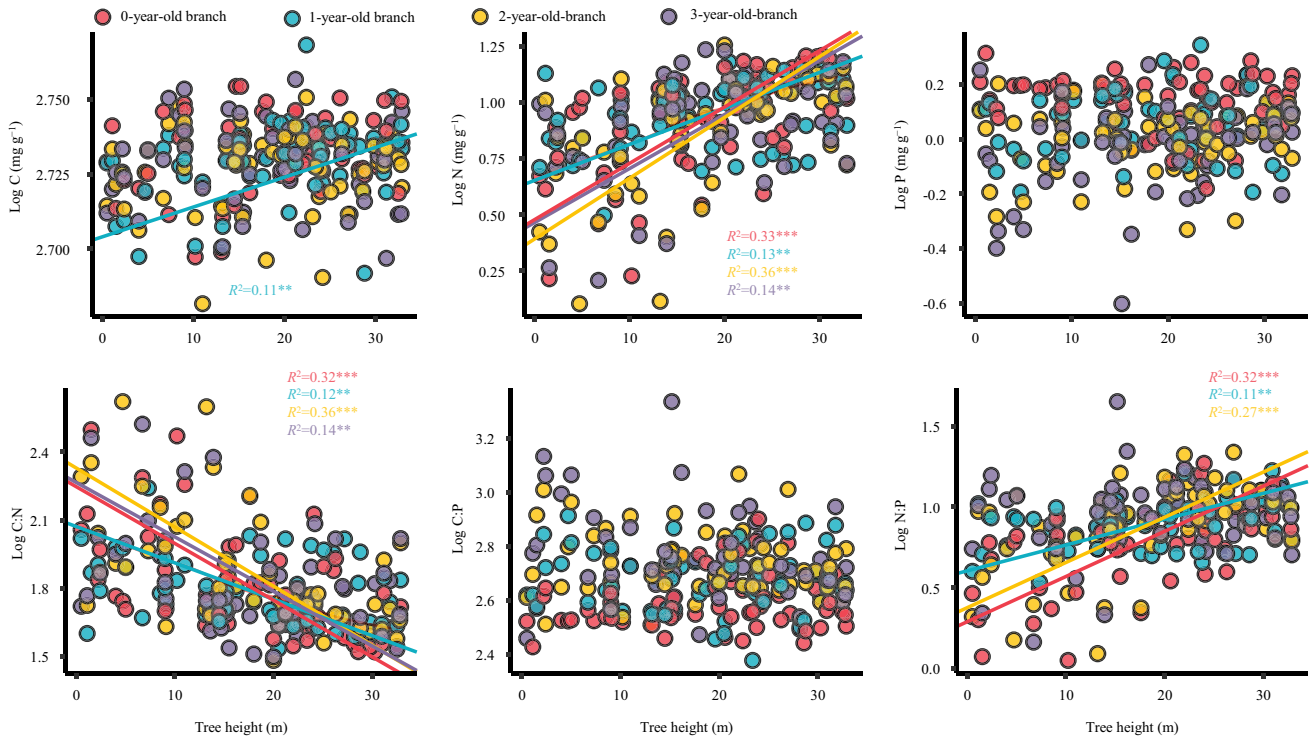


Fig. 2 Variation in branch nutrient traits across branch ages and heights for *Pinus koraiensis*. Different colored data points for nutrient traits represent 0-year-old, 1-year-old, 2-year-old and 3-year-old

branches, respectively. The trend line and regression information are marked in the figure as significant. * $p < 0.05$, ** $p < 0.01$, *** $p < 0.001$

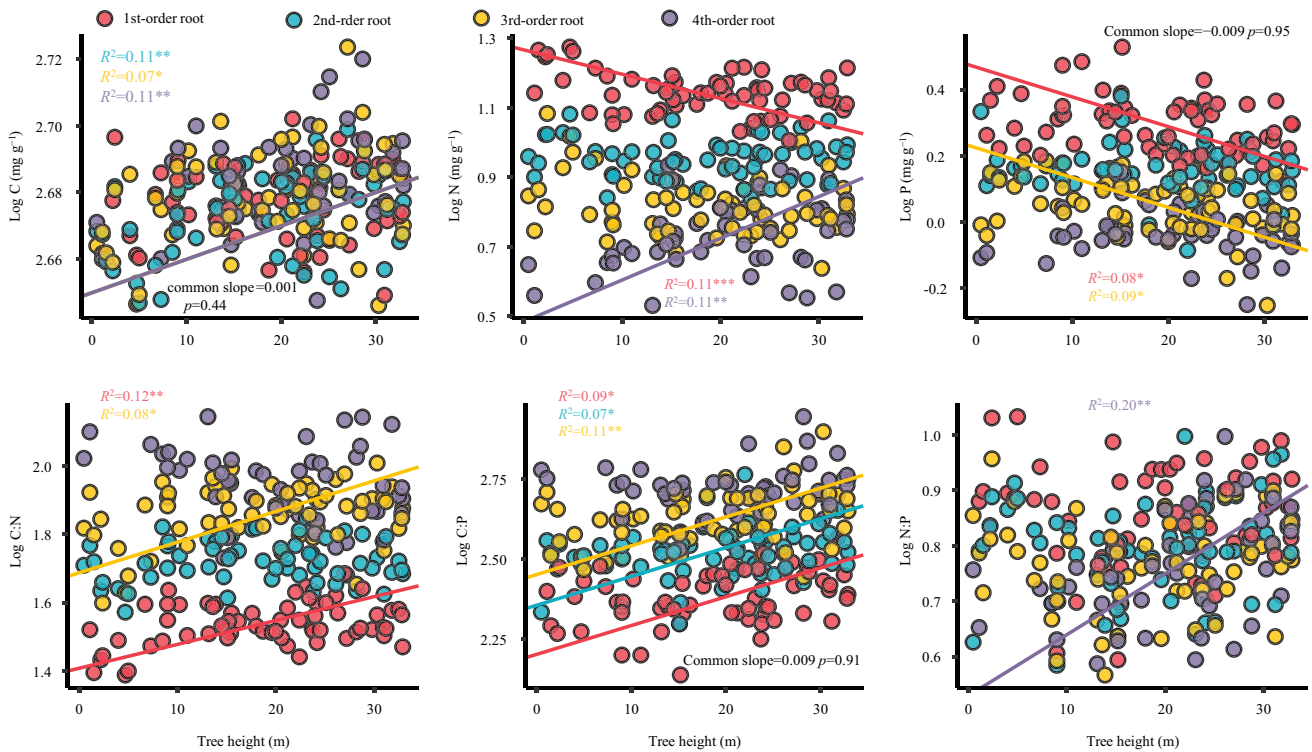


Fig. 3 Variation in root nutrient traits in different root orders and tree heights for *Pinus koraiensis*. Different colored data points for nutrient traits represent 1st-order, 2nd-order, 3rd-order and 4th-order roots,

respectively. The trend line and regression information are marked in the figure as significant. * $p < 0.05$, ** $p < 0.01$, *** $p < 0.001$

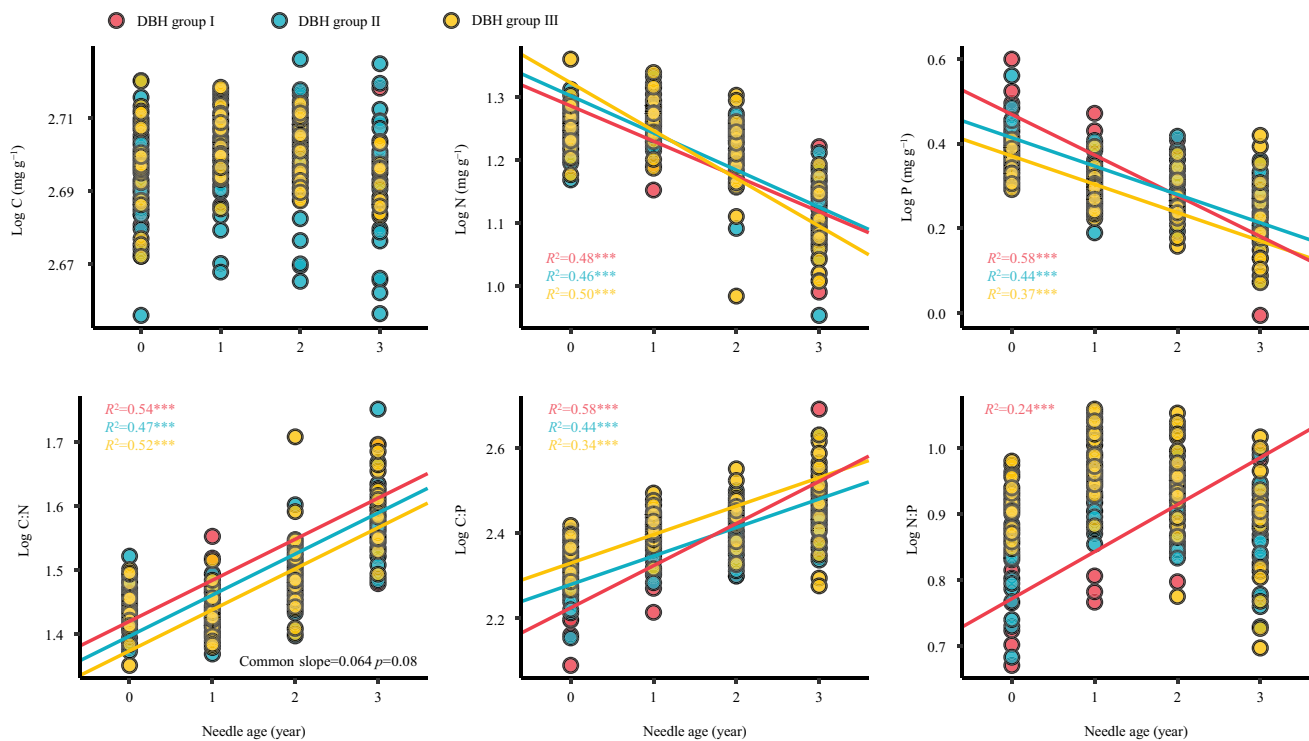


Fig. 4 Variation in needle nutrient traits for different size (DBH) and needle age groups for *Pinus koraiensis*. Different colored data points for nutrient traits represent three groups based on DBH. DBH group I: DBH ≤ 10 cm; DBH group II: $10 < \text{DBH} \leq 50$ cm; DBH group III:

$50 < \text{DBH} \leq 100$ cm. The trend line and regression information are marked in the figure as significant. * $p < 0.05$, ** $p < 0.01$, *** $p < 0.001$

soil directly, require sufficient N and P to support complicated physiological metabolic activities and transport the additional N and P to needles and branches (Yuan et al. 2011).

Additionally, Han et al. (2011) suggested that highly required nutrients should be shown as low variability (or high stability). In this study, C has as relatively lower variability (CVs) (Table 2), emphasizing that C is the most stable element which maintains the construction of the basic framework within plants. Needles are then the relatively stable organs with lower variability (CVs) (Table 2). It is likely that plants transfer sufficient nutrients from other organs to sustain the stability of needle nutrients for photosynthesis (Zhao et al. 2016). Our study revealed that nutrient variations among different plant organs also depends on cooperation between organs. It is also a consequence of plant evolution (Zhang et al. 2019). Our results contribute to the knowledge on nutrient variations and the underlying mechanisms at individual levels compared to using a single plant organ.

Previous studies found that soil factors had a negative or positive affect on nutrient traits among organs (Zhao et al. 2016; Xiong et al. 2021), with some suggesting that smaller trees were more sensitive to environmental factors at regional to global scales (Liu et al. 2010; Li et al. 2018).

In this study, nutrient variations among organs were influenced by tree size, organ age or root order, rather than soil factors (Table 3). The contrasting results are possibly due to the different study scales, i.e., the nutrient traits may be more influenced by soil factors at large scales caused by the significant variation of soil properties, while nutrient traits are more influenced by biological characteristics at the individual level, such as tree size, organ age or root order.

Tree size is an important characteristic governing nutrient utilization and acquisition at the whole plant level (Ågren 2008; Martin and Thomas 2013; Li et al. 2018). To date, little research has focused on nutrient variations at the whole plant level. This study comprehensively considered nutrient variation among major plant organs as trees grew and found that nutrient traits converged in needles and fine roots and tended decrease with size. Nutrient traits in branches tended to increase with size (Figs. 1–3).

Higher nutrient levels in needles and roots related to higher metabolic activity in smaller trees, reflecting greater requirement for protein synthesis to sustain rapid growth to reach the upper canopy as quickly as possible. In turn, increasing light availability intensifies the demand for nutrients supplied by roots during the sapling-to-mature transition (Cavaleri et al. 2010; Zhang et al. 2017). An intense transport of nutrients between needles and roots is through

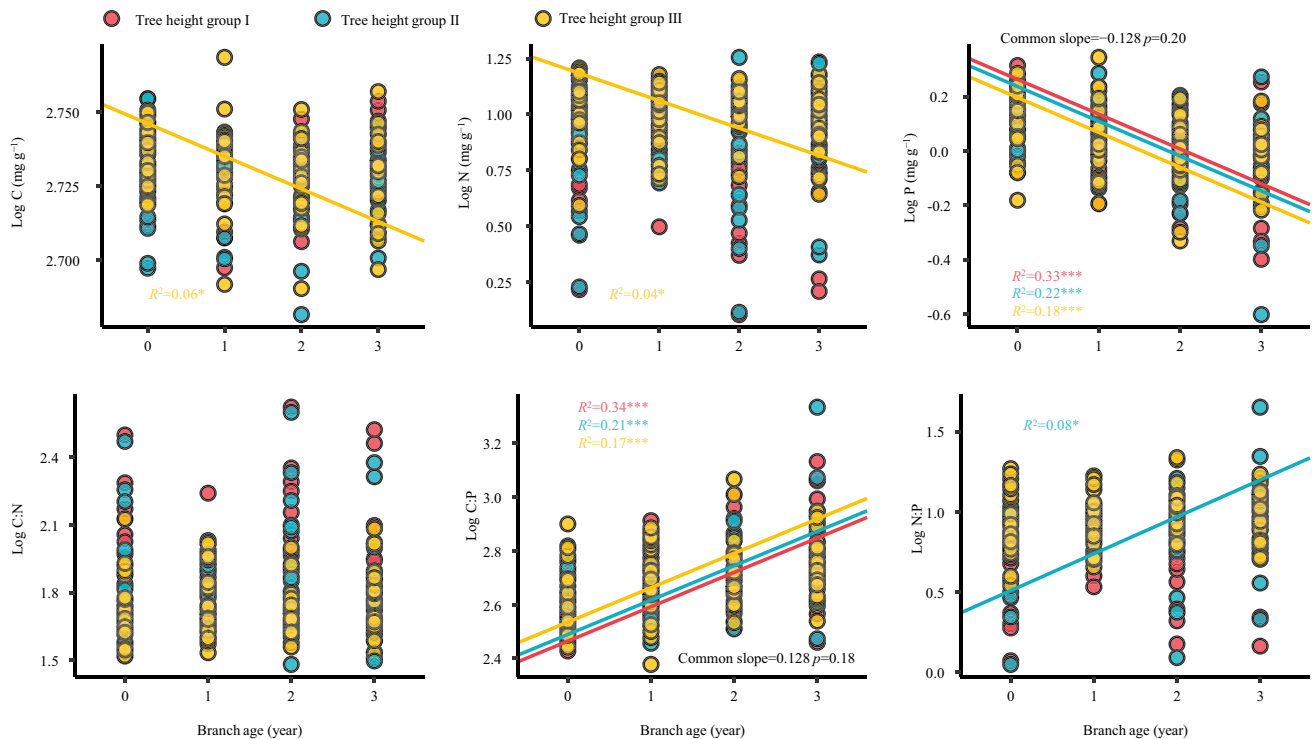


Fig. 5 Variation in branch nutrient traits for different size (tree height) and branch age groups for *Pinus koraiensis*. Different colored data points for nutrient traits represent three groups based on tree height. Tree height group I: Tree height ≤ 10 m; Tree height group II:

$10 < \text{Tree height} \leq 20$ m; Tree height group III: Tree height > 20 m. The trend line and regression information are marked in the figure as significant. * $p < 0.05$, ** $p < 0.01$, *** $p < 0.001$

branches, which represent mechanical support and nutrient transport tissues of plants. The increased need for support and nutrient transport in branches as a tree grows drives greater nutrient requirement (Yan et al. 2016). In summary, as a tree grows, organs function as cooperative systems dominate the nutrient variation among organs and is closely related to nutrient allocation and utilization to ensure survival.

From the perspective of tree ontogeny processes, small trees with higher relative nutrient levels to support intense metabolic activity, the process is referred to as the ‘nutrient acquisition strategy’. Conversely, as trees mature and volume increases, a significant amount of nutrients accumulate, and large trees become an internal nutrient pool to better meet environmental challenges. This is referred to as the ‘nutrient storage strategy’. The modification of this strategy is a progressive process that occurs throughout the life of the tree, rather than occurring abruptly.

From the perspective of internal nutrient variation, organ age or root order as the biological characteristic driving nutrient variation has been largely ignored by previous research. Organ age or root order generally has a negative effect on N and P and a positive effect on their stoichiometric ratios among the major organs (Figs. 4–6). These results are consistent with those of Li et al. (2010). Younger organ

fractions are the main site for complex, intense physiological activity, which drives the demand for higher N and P and enhances N and P utilization efficiency to facilitate rapid protein or enzyme synthesis (Li et al. 2010; McCormack et al. 2015). These nutrient differences are reflected as larger chloroplasts and more mesophyll cells in younger needles (Kuusk et al. 2018b); more repetitive production of growth units for support structure in younger branches (Kay et al. 2005; Fortunel et al. 2014); and more cortical cells which absorb nutrients and support metabolic activity in lower-order roots (Pregitzer 2002; McCormack et al. 2015). Eventually, all older or higher order organ fractions become more structurally robust, owing to the accumulation and increased lignification of cell walls and fibrous tissue (Niinemets and Lukjanova 2003; Kuusk et al. 2018a). Increasing demand for nutrients and water transport efficiency results in the thickening and multiplication of conduits in older branches (Kay et al. 2005; Fortunel et al. 2014). Xylem proportions and diameters increase, leading to expanded transport capacity in higher-order roots (Fortunel et al. 2014). This finding confirmed previous studies and there is a distinguishable functional divergence within organs: nutrient content is produced or absorbed in younger organ fractions, and stored or transport in older organ fractions (McCormack et al. 2015).

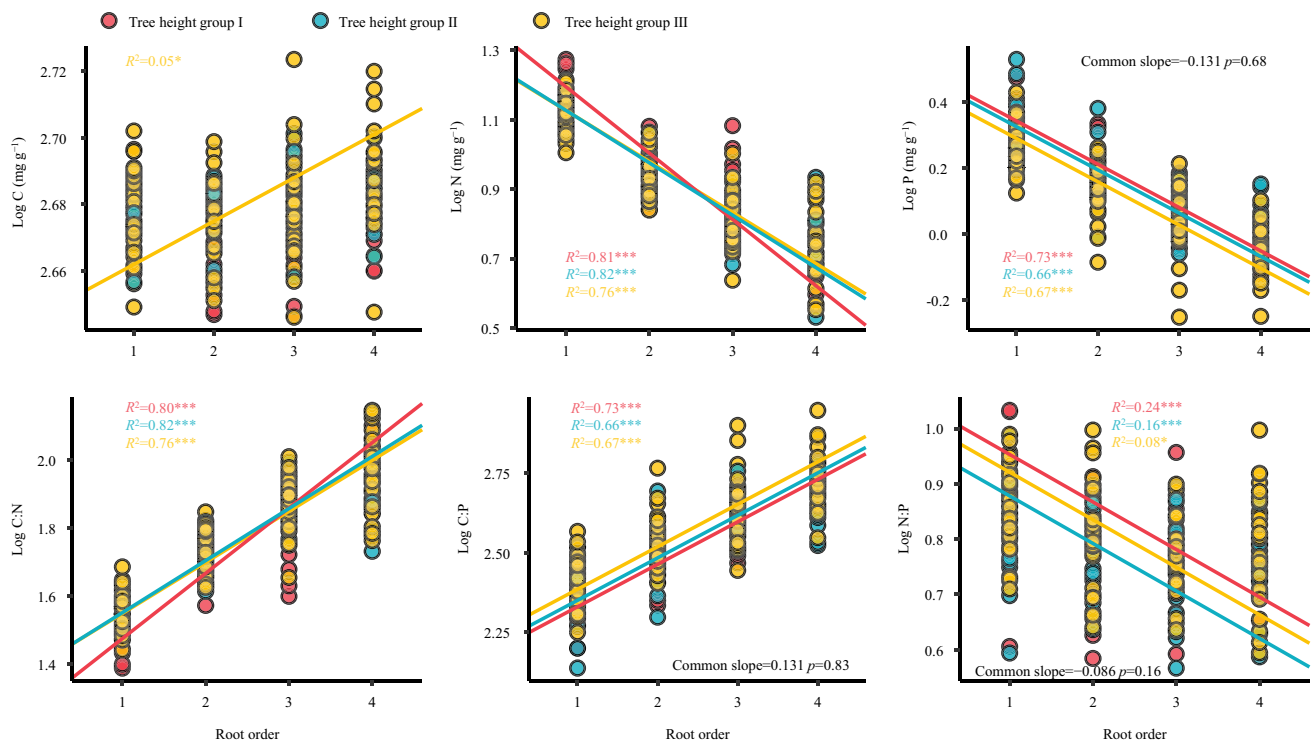


Fig. 6 Variation in root nutrient traits for different size (tree height) and root orders for *Pinus koraiensis*. Different colored data points for nutrient traits represent three groups based on tree height. Height group I: Tree height ≤ 10 m; height group II: $10 < \text{Tree height} \leq 20$

m; height group III: Tree height > 20 m. The trend line and regression information are marked in the figure as significant. * $p < 0.05$, ** $p < 0.01$, *** $p < 0.001$

This is a key pathway to meet nutrient demands resulting from the formation of new tissues and is regarded as a mechanism for nutrient conservation and utilization within a species for growth and survival. Similar to the widely acknowledged absorptive roots and transport roots with different physiological functions (McCormack et al. 2015), needles and branches of different ages also may be split into different functional groups: photosynthetic needles and nutrient transport needles and elongated branches and nutrient transport branches. We suspect that they are based on secondary growth, which is also the most directly indication of nutrient use strategy change and amplifies functional divergence (McCormack et al. 2015). The findings in this study demonstrate and refine the existence of an underlying trade-off strategy between nutrient investment in transport and nutrient acquisition within organs that seeks to optimize plant growth to adapt to changing environmental conditions. Thus, future studies should take sufficient consideration to nutrient variation within organs. In addition, our results need to be anatomically confirmed and perhaps refined.

Conclusion

This study extensively considered nutrient traits among different organs (i.e., needles, branches, and fine roots)

of *P. koraiensis* in a natural forest. Our results indicate that nutrient variation in organs are regulated by tree size, organ age or root order rather than by soil properties. Nutrient traits decreased in needles and fine roots and increased in branches with tree size. At the organ level, C, N, and P contents decreased and stoichiometric ratios increased markedly with organ age and root order. These findings confirms that plant growth depends on the functional coordination among organs. We also proposed a nutrient acquisition strategy in younger trees for survival. Conversely, nutrient storage strategy in older trees is mainly for steadily growth. Our findings advance knowledge about plant nutrient variations in different plant organs at an individual level, and highlight modification of the nutrient strategy. This will allow identification of underlying tree growth mechanisms and clarify internal nutrient strategies under rapid global change.

Acknowledgements We would like to thank Dr. Joseph Elliot at the University of Kansas for his assistance with the English language and grammatical editing of the manuscript.

Funding The work was financially supported by the National Key R&D Program of China (2022YFD2201100), Natural Science Foundation of Heilongjiang Province of China (TD2023C006), and the Fundamental Research Funds for the Central Universities (2572022DS13).

Open Access This article is licensed under a Creative Commons Attribution 4.0 International License, which permits use, sharing, adaptation, distribution and reproduction in any medium or format, as long as you give appropriate credit to the original author(s) and the source, provide a link to the Creative Commons licence, and indicate if changes were made. The images or other third party material in this article are included in the article's Creative Commons licence, unless indicated otherwise in a credit line to the material. If material is not included in the article's Creative Commons licence and your intended use is not permitted by statutory regulation or exceeds the permitted use, you will need to obtain permission directly from the copyright holder. To view a copy of this licence, visit <http://creativecommons.org/licenses/by/4.0/>.

References

- Ågren GI (2004) The C:N:P stoichiometry of autotrophs-theory and observations. *Ecol Lett* 7(3):185–191. <https://doi.org/10.1111/j.1461-0248.2004.00567.x>
- Ågren GI (2008) Stoichiometry and nutrition of plant growth in natural communities. *Annu Rev Ecol Evol Syst* 39(1):153–170. <https://doi.org/10.1146/annurev.ecolsys.39.110707.173515>
- Anderson-Teixeira KJ, Mcgarvey JC, Muller-Landau HC, Park JY, Gonzalez-Akre EB, Herrmann V, Bennett AC, So CV, Bourg NA, Thompson JR (2016) Size-related scaling of tree form and function in a mixed-age forest. *Funct Ecol* 29(12):1587–1602. <https://doi.org/10.1111/1365-2435.12470>
- Burnham KP, Anderson DR (2004) Basic use of the information-theoretic approach. Model selection and multimodel inference. Springer, New York, pp 98–148
- Cavaleri MA, Oberbauer SF, Clark DB, Clark DA, Ryan MG (2010) Height is more important than light in determining leaf morphology in a tropical forest. *Ecology* 91(6):1730–1739. <https://doi.org/10.1890/09-1326.1>
- Chatterjee S, Price B (1991) Regression diagnostics. John Wiley, New York
- Eimil-Fraga C, Sánchez-Rodríguez F, Álvarez-Rodríguez E, Rodríguez-Soalleiro R (2015) Relationships between needle traits, needle age and site and stand parameters in *Pinus pinaster*. *Trees* 29(4):1103–1113. <https://doi.org/10.1007/s00468-015-1190-7>
- Fortunel C, Ruelle J, Beauchene J, Fine PV, Baraloto C (2014) Wood specific gravity and anatomy of branches and roots in 113 Amazonian rainforest tree species across environmental gradients. *New Phytol* 202(1):79–94. <https://doi.org/10.1111/nph.12632>
- Han WX, Fang JY, Reich PB, Woodward FI, Wang ZH (2011) Biogeography and variability of eleven mineral elements in plant leaves across gradients of climate, soil and plant functional type in China. *Ecol Lett* 14(8):788–796. <https://doi.org/10.1111/j.1461-0248.2011.01641.x>
- He D, Yan ER (2018) Size-dependent variations in individual traits and trait scaling relationships within a shade-tolerant evergreen tree species. *Am J Bot* 105(7):1–10. <https://doi.org/10.1002/ajb2.1132>
- He MZ, Zhang K, Tan HJ, Hu R, Su JQ, Wang J, Huang L, Zhang YF, Li XR (2015) Nutrient levels within leaves, stems, and roots of the xeric species *Reaumuria soongorica* in relation to geographical, climatic, and soil conditions. *Ecol Evol* 5(7):1494–1503. <https://doi.org/10.1002/ece3.1441>
- Kay AD, Ashton IW, Gorokhova E, Kerkhoff AJ, Liess A, Litchman E (2005) Toward a stoichiometric framework for evolutionary biology. *Oikos* 109(1):6–17. <https://doi.org/10.1111/j.0030-1299.2005.14048.x>
- Kuusk V, Niinemets Ü, Valladares F (2018a) A major trade-off between structural and photosynthetic investments operative across plant and needle ages in three Mediterranean pines. *Tree Physiol* 38:543–557. <https://doi.org/10.1093/treephys/tpx139>
- Kuusk V, Niinemets Ü, Valladares F, Tjoelker M (2018b) Structural controls on photosynthetic capacity through juvenile-to-adult transition and needle ageing in Mediterranean pines. *Funct Ecol* 32:1479–1491. <https://doi.org/10.1111/1365-2435.13087>
- Li A, Guo DL, Wang ZQ, Liu HY (2010) Nitrogen and phosphorus allocation in leaves, twigs, and fine roots across 49 temperate, subtropical and tropical tree species: a hierarchical pattern. *Funct Ecol* 24(1):224–232. <https://doi.org/10.1111/j.1365-2435.2009.01603.x>
- Li Y, Tian DS, Yang H, Niu SL (2018) Size-dependent nutrient limitation of tree growth from subtropical to cold temperate forests. *Funct Ecol* 32(1):95–105. <https://doi.org/10.1111/1365-2435.12975>
- Liu C, Wang X, Wu X, Dai S, He JS, Yin WL (2013) Relative effects of phylogeny, biological characters and environments on leaf traits in shrub biomes across central Inner Mongolia. *China J Plant Ecol* 6(3):220–231. <https://doi.org/10.1093/jpe/rt028>
- Liu FD, Yang WJ, Wang ZS, Xu Z, Liu H, Zhang M, Liu YH, An SQ, Sun SC (2010) Plant size effects on the relationships among specific leaf area, leaf nutrient content, and photosynthetic capacity in tropical woody species. *Acta Oecol* 36(2):149–159. <https://doi.org/10.1016/j.actao.2009.11.004>
- Liu ZL, Jiang F, Li FR, Jin GZ (2019) Coordination of intra and inter-species leaf traits according to leaf phenology and plant age for three temperate broadleaf species with different shade tolerances. *For Ecol Manage* 434:63–75. <https://doi.org/10.1016/j.foreco.2018.12.008>
- Liu ZL, Hikosaka K, Li FR, Jin GZ, Ostertag R (2020) Variations in leaf economics spectrum traits for an evergreen coniferous species: tree size dominates over environment factors. *Funct Ecol* 34(2):458–467. <https://doi.org/10.1111/1365-2435.13498>
- Martin AR, Thomas SC (2013) Size-dependent changes in leaf and wood chemical traits in two Caribbean rainforest trees. *Tree Physiol* 33(12):1338–1353. <https://doi.org/10.1093/treephys/tpt085>
- Martínez-Vilalta J, Vanderklein D, Mencuccini M (2007) Tree height and age-related decline in growth in Scots pine (*Pinus sylvestris* L.). *Oecologia* 150(4):529–544. <https://doi.org/10.1007/s00442-006-0552-7>
- McCormack ML, Dickie IA, Eissenstat DM, Fahey TJ, Fernandez CW, Guo DL, Helmisaari HS, Hobbie EA, Iversen CM, Jackson RB, Leppalammi-Kujansuu J, Norby RJ, Phillips RP, Pregitzer KS, Pritchard SG, Rewald B, Zadworny M (2015) Redefining fine roots improves understanding of below-ground contributions to terrestrial biosphere processes. *New Phytol* 207(3):505–518. <https://doi.org/10.1111/nph.13363>
- Minden V, Kleyer M (2014) Internal and external regulation of plant organ stoichiometry. *Plant Biol* 16(5):897–907. <https://doi.org/10.1111/plb.12155>
- Niinemets Ü, Lukjanova A (2003) Needle longevity, shoot growth and branching frequency in relation to site fertility and within-canopy light conditions in *Pinus sylvestris*. *Ann for Sci* 60:195–208. <https://doi.org/10.1051/forest:2003012>
- Pregitzer KS (2002) Fine roots of trees—a new perspective. *New Phytol* 154(2):267–273
- R Core Team (2020) R: a language and environment for statistical computing. R foundation for statistical computing, Vienna, Austria
- Sendall KM, Reich PB (2013) Variation in leaf and twig CO₂ flux as a function of plant size: a comparison of seedlings, saplings and trees. *Tree Physiol* 33(7):713–729. <https://doi.org/10.1093/treephys/tpt048>

- Sterner RW, Elser JJ (2002) Ecological stoichiometry the biology of elements from molecules to the biosphere. Princeton University, Press Princeton
- Tang ZY, Xu WT, Zhou GY, Bai YF, Li JX, Tang XL, Chen DM, Liu Q, Ma WH, Xiong GM, He HL, He NP, Guo YP, Guo Q, Zhu JL, Han WX, Hu HF, Fang JY, Xie ZQ (2018) Patterns of plant carbon, nitrogen, and phosphorus concentration in relation to productivity in China's terrestrial ecosystems. *Proc Natl Acad Sci U S A* 115(16):4033–4038. <https://doi.org/10.1073/pnas.1700295114>
- Wang AJ, Liu BY, Wang ZQ, Liu G (2016) Monitoring and predicting the soil water content in the deeper soil profile of Loess Plateau. *China Int Soil Water Conse* 4(1):6–11. <https://doi.org/10.1016/j.iswcr.2016.02.001>
- Warton DI, Weber NC (2002) Common slope tests for bivariate errors-in-variables models. *Biom J* 44(2):161–174. [https://doi.org/10.1002/1521-4036\(200203\)44:2%3c161::AID-BIMJ161%3e3.0.CO;2-N](https://doi.org/10.1002/1521-4036(200203)44:2%3c161::AID-BIMJ161%3e3.0.CO;2-N)
- Warton DI, Wright IJ, Falster DS, Westoby M (2006) Bivariate line-fitting methods for allometry. *Biol Rev* 81(2):259–291. <https://doi.org/10.1017/S1464793106007007>
- Wright IJ, Reich PB, Westoby M, Ackerly DD, Baruch Z, Bongers F, Cavender-Bares J, Chapin T, Cornelissen JH, Diemer M, Flexas J, Garnier E, Groom PK, Gulias J, Hikosaka K, Lamont BB, Lee T, Lee W, Lusk C, Midgley JJ, Navas ML, Niinemets U, Oleksyn J, Osada N, Poorter H, Poot P, Prior L, Pyankov VI, Roumet C, Thomas SC, Tjoelker MG, Veneklaas EJ, Villar R (2004) The worldwide leaf economics spectrum. *Nature* 428(6985):821–827. <https://doi.org/10.1038/nature02403>
- Xiong JL, Dong LW, Lu JL, Hu WG, Gong HY, Xie SB, Zhao DM, Zhang YH, Wang XT, Deng Y, Ran JZ, Niklas KJ, Degen A, Deng JM (2021) Variation in plant carbon, nitrogen and phosphorus contents across the drylands of China. *Funct Ecol* 36(1):174–186. <https://doi.org/10.1111/1365-2435.13937>
- Yan BG, Ji ZH, Fan B, Wang XM, He GX, Shi LT, Liu GC (2016) Plants adapted to nutrient limitation allocate less biomass into stems in an arid-hot grassland. *New Phytol* 211(4):1232–1240. <https://doi.org/10.1111/nph.13970>
- Yang DX, Song L, Jin GZ (2019) The soil C:N:P stoichiometry is more sensitive than the leaf C:N:P stoichiometry to nitrogen addition: a four-year nitrogen addition experiment in a *Pinus koraiensis* plantation. *Plant Soil* 442(5):183–198. <https://doi.org/10.1007/s11104-019-04165-z>
- Yuan ZY, Chen HY, Reich PB (2011) Global-scale latitudinal patterns of plant fine-root nitrogen and phosphorus. *Nat Commun* 2:344. <https://doi.org/10.1038/ncomms1346>
- Zhang H, Yang XQ, Wang JY, Wang GG, Yu MK, Wu TG (2017) Leaf N and P stoichiometry in relation to leaf shape and plant size for *Quercus acutissima* provenances across China. *Sci Rep* 7:46133. <https://doi.org/10.1038/srep46133>
- Zhang JH, Zhao N, Liu CC, Yang H, Li ML, Yu GR, Wilcox K, Yu Q, He NP (2018) C:N:P stoichiometry in China's forests: from organs to ecosystems. *Funct Ecol* 32(1):50–60. <https://doi.org/10.1111/1365-2435.12979>
- Zhang JH, He NP, Liu CC, Xu L, Chen Z, Li Y, Wang RM, Yu GR, Sun W, Xiao CW, Chen HYH, Reich PB (2019) Variation and evolution of C:N ratio among different organs enable plants to adapt to N-limited environments. *Global Change Biol* 26:2534–2543. <https://doi.org/10.1111/gcb.14973>
- Zhang JH, He NP, Liu CC, Xu L, Yu Q, Yu GR (2018) Allocation strategies for nitrogen and phosphorus in forest plants. *Oikos* 127(10):1506–1514. <https://doi.org/10.1111/oik.05517>
- Zhang J, Wang YZ, Cai CT (2020) Multielemental stoichiometry in plant organs: a case study with the Alpine *Herb Gentiana rigescens* across Southwest China. *Front in Plant Sci* 11:441. <https://doi.org/10.3389/fpls.2020.00441>
- Zhao N, Yu GR, He NP, Wang QF, Guo DL, Zhang XY, Wang RL, Xu ZW, Jiao CC, Li NN, Jia YL (2016) Coordinated pattern of multi-element variability in leaves and roots across Chinese forest biomes. *Global Ecol Biogeogr* 25(3):359–367. <https://doi.org/10.1111/geb.12427>

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