

Jointly soil properties affect N and P uptakes and utilizations in *Pinus tabulaeformis* Carr. and *Quercus liaotungensis* Koidz. subjected to growing media with decomposed litter

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Abstract

Monocultured pine plantation is suffering ecological degradation that is highly associated with low regeneration. Decomposed litter is an important soil amendment for enhancing regeneration through promoting nitrogen (N) and phosphorus (P) uptakes and utilizations. It is necessary to detect key soil attributes that contributed to this positive effect for regenerations in pine plantations. In this study, in-situ soils and litter were collected from local Chinese pine (*Pinus tabulaeformis* Carr.) plantations (objective) and secondary forests dominated by Liaodong oak (*Quercus liaotungensis* Koidz.) (control). Soils were used for culturing one-year-old Chinese pine and Liaodong oak seedlings with a prolonged photoperiod in a greenhouse. Litter was composted with effective microorganisms and mixed to soils at ratios of 0% (control), 25%, and 50% (v/v). Compared to the control, the 25% ratio decreased shoot height and root-collar diameter, and the 50% ratio decreased the comprehensive seedling quality. Decomposed litter addition reduced shoot biomass and P content in pine seedlings and utilizations for N and P in both species. Multivariate linear regression indicated that high pH in growing media impaired root P content and biomass increments in shoot and root parts, and high organic matter content inhibited N content and concentration in shoots. Overall, the addition of decomposed litter resulted in overdoses of nutrient supply for both species. Our results contradict the argument that N and P released from decomposed litter are both beneficial for regenerations in plantations, neither did in secondary forests.

Keywords: Chinese pine; Liaodong oak; nutrient utilization; potted seedling culture; regeneration; secondary forests; silviculture

Introduction

Land-use conversion causes intensive changes of landforms since the Industrial Revolution. Artificial intervention is a strong force that converts natural primary forests (old growth with no intervention ever) to secondary forests (SFs) (natural recovery) or planted forests (artificial planting) (Liu *et al.*, 2018). As of 2020, tree plantations and planted forests together accounted for 7% of global forest area (World Resources Institute, 2023). Plantation forests established with monocultural species can cause ecosystem defects, such as impeding

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regional forest recovery (Zhu *et al.*, 2023), degrading biodiversity (Cardoso *et al.*, 2023; He *et al.*, 2023), and decreasing soil pools (Ding *et al.*, 2021; Liu *et al.*, 2023). Low rate of regeneration is a large challenge for sustainable management of plantation forests (Yang *et al.*, 2023). Either low germination or poor growth of juvenile seedling may cause failure of regeneration in monocultured tree plantations (Wang *et al.*, 2023) by high stand density (Ali *et al.*, 2019), low sunlight transmittance (Wang *et al.*, 2023), improper meteorological condition (Guo *et al.*, 2019), low plant diversity (Gomez-Aparicio *et al.*, 2009), and soil quality (Ali *et al.*, 2019). Restoration is a practical operation to promote regeneration and increase species diversity in tree plantations (Lengyel *et al.*, 2020).

Stand soil availability is a key factor that can matter for understory richness (Su *et al.*, 2022). Nitrogen (N) is an important component of forest soils and plays a fatal role in ecosystem health (Sardar *et al.*, 2023). Mineralized N is heavily needed if a high production is expected by a forest plantation (Yang *et al.*, 2015). Most pieces of phosphorus (P) are bound to soil components that generate an obstacle for P uptake (Zhou *et al.*, 2022). The function of triphosphadenine relies on P supply to capture and convert sunlight energy to essential substance needed by plants. Low N and P availabilities are vital factors that limit plantation quality, which is usually coped with by exogenous N and P additions (Yang *et al.*, 2015; Zhu *et al.*, 2016). Industrial development stimulated atmospheric N emission and deposition onto forests that may have broken the balance of N-P availabilities. In subtropical forests, N deposition was found to decrease soil labile P fractions but increase intermediate P fractions, which together strengthened local P limitation (Chen *et al.*, 2018). In tropical forests, N input was reported to prolong the period of litter decomposition with carbon, N, and P all decreased in local soils (Zhu *et al.*, 2016). In temperate forests, exogeneous N input reduced inorganic P availability (Yang *et al.*, 2015). Frequent uses of chemical fertilizers experience the risk of ground water contamination (Machado *et al.*, 2022). The N-P balance should be maintained by synchronized N and P availabilities both at high levels in plantation forests.

Litter decomposition is a major access through which bound N and P are released and moved to soils (Peh *et al.*, 2012). Under a natural condition, litter decomposition is a complicated process which mainly determined by interactions with temperature (Liu *et al.*, 2017), pH value, moisture, fauna activity (Fernandez *et al.*, 2022), degradative enzyme activity in soils (Wang *et al.*, 2013), and forest type (Barlow *et al.*, 2007). Decomposed litter can also be used as a part of growing substrates for artificially cultured seedlings. The conversion of forested land use had a strong effect to modify N and P releases from litter decomposition (Zhang *et al.*, 2018). In Brazilian Amazon, litter was decomposed faster in *Eucalyptus urophylla* plantations than in primary forests (dominated by *Bertholettia excelsa* and *Vismia* spp.) (Barlow *et al.*, 2007). In contrast, soil N and P contents were higher in secondary forests than in local plantations subjected to temperate (Chen *et al.*, 2017) and subtropical climates (Zhu *et al.*, 2021). It is still disputed whether regenerations need the full release of mineral N and P in litter remain controversial (Ni *et al.*, 2021).

Lignocellulose in litter is decomposed at a low rate which is controlled by bacterial and fungal activities (Wang *et al.*, 2020). It is impossible to upregulate activities of decomposers *in situ*, but both decomposition and mineralization can be promoted through composting (Liu *et al.*, 2021). This can be explained by efficient degradation of lignocellulose in composted litter with high ligninolytic enzymes (Martinez-Garcia *et al.*, 2021). Therefore, amending soils with compost is a practical approach to promote nutrient cycling and availability in planted forests (Boafo *et al.*, 2020). Composted litter is a high-quality organic amendment that retains abundant N and P reserves (Turp *et al.*, 2021). Amendment using composted manure has been proven to promote quality of plantation forests (Mohale *et al.*, 2021). Composted litter, however, has attracted rare attention to its usage as an amendment for planted forest trees.

Chinese pine (*Pinus tabulaeformis* Carr.) plantation accounts for a major proportion of planted pine forests in North China (Cao and Chen, 2017; Deng *et al.*, 2018). Monocultured Chinese pine plantations

occupy a large area of forested lands in montane regions of eastern Liaoning of Northeast China (Wang *et al.*, 2022d; Yin *et al.*, 2021). Many of these plantations were established on lands converted from secondary broad-leaf forests dominated by Liaodong oak (*Quercus liaotungensis* Koidz.) (Wang *et al.*, 2012). Currently, many monocultured Chinese pine plantations were reported to suffer ecological degradation symptomized as high mortality and low timber productivity (Li *et al.*, 2020b). In these plantations soils were reported to experience severely low P availability which may have modified fungal community structure and impaired the success of restoration (Wang *et al.*, 2022a). People start to appeal to convert monocultured Chinese pine plantations to mixed forests with more broad leaf species introduced to overcome all abovementioned drawbacks and strengthen biodiversity (Duan *et al.*, 2022b; Yin *et al.*, 2021). Secondary forests can induce natural regeneration to an acceptable extent for newly grown pine saplings (Ge *et al.*, 2021). However, the regeneration in plantations usually fails to meet desired expectations. It is practical to cope with this issue using ecological instrument and soil amended with composted litter can be an excellent candidate as a solver.

In this study, typical Chinese pine plantations were chosen as the objective forest type with adjacent secondary Liaodong oak forests as a baseline for comparison. Our objective is to test decomposed litter amendment effect on nutrient uptake and utilization in pine and oak seedlings. We also have a purpose to unravel the mechanism of multiple soil attributes together accounting for growth and biomass outcomes through N and P utilizations.

Materials and Methods

Study area and stand description

Chinese pine (*P. tabuliformis* Carr.) plantations and Liaodong oak (*Q. liaotungensis* Koidz.) forests were chosen in a forest farm located at Caohekou town (40°53' N, 120°54' E), Benxi, Liaoning, China. Forests were subjected to a temperate continental monsoon climate with an average temperature of 6.5 °C (-33–32 °C) and an annual precipitation of 926.3 mm. Local frost-free period was averaged to be 127 days throughout the year when air humidity ranged in 50-60%. Local Chinese pine plantations had averaged stand density of 260 ± 51 (mean ± standard deviation) stems per hectare with averaged tree height of 13.44 ± 2.09 m, diameter at breast height of 23.07 ± 0.21 cm, and lowest alive branch height of 6.89 ± 0.40 cm. Subsequent growth was measured on the basis of these parameters as annual increment of growth. Plantations were mostly converted from secondary forests dominated by Liaodong oak trees with an average stand age of 35 ± 3 years, which. Local secondary forests harbored other tree species of *Acer pictum* subsp. mono (Maxim.) H. Ohashi, *Fraxinus rhynchophylla* Hance., *Juglans mandshurica* Maxim., *Phellodendron amurense* Rupr., *P. koraiensis*, and *Quercus mongolica* Fisch. Ex Ledeb, and shrub species of *A. triflorum* Kom., *A. mandshuricum* Maxim., and *Syringa reticulata* subsp.

Litter compost and soil preparation

In April, 2022, litter was collected in 20 m × 30 m plots of Chinese pine and Liaodong oak stands (Figure 1). Five plots were set in stands for litter collection. Adjacent two plots were placed in a distance of at least 500 m. Space in a plot was divided to 24 grids and each had an area of 25 m² (5 m × 5 m). Half of grids were assigned for litter collection and the other adjacent half were used as buffers. Grids and buffers were arranged alternatively in a plot to prevent intervention of any collecting grid. A 1 m²-area (1 m × 1 m) square was set in the centre of a collecting grid and a total of 24 squares were placed in a plot. Litter was collected from the surface of forest floor at depth of 5 cm aboveground. Collected litter pieces were piled in two stacks which were distinguished by two forest types and maintained in a warehouse to dry in air without any wind ventilation.

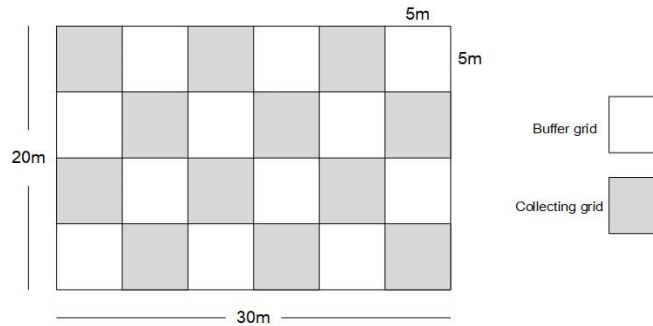


Figure 1. Layout of field sampling of litter and soils in forest stands

Soils were also collected from grids of litter collection. Before soil collection, all left pieces of litter at forest floor were removed from a collecting grid. Soils were collected in bulk of a volume of 0.2 m^3 ($1 \text{ m} \times 1 \text{ m} \times 0.2 \text{ m}$, length \times width \times height) at a depth of 20–40 cm below the ground. We did not collect soils from the surface because chemical component contents in soils at this depth were found to be higher than those in deeper soils due to the “surface aggregation” phenomenon in local Chinese pine plantations (Wang *et al.*, 2022c). Collected soils were screened through a 1 cm sieve to remove stones and dead wood chips. Thereafter, soils were screened through a 2 mm sieve to reserve fine and even particles. Residues of plant roots and dead bug bodies were manually removed. Soils were stacked on plastic sheets and arranged in two piles which were also distinguished by forest types. Soils were dried in air on the same day with litter and maintained until compost was finished.

Two weeks later, litter piles were smashed to 2 cm-chips by a grinder and prepared for composting. Commercial effective microorganisms (EM) powder (Gelyin Ecol. S&T Inc., Zhangjiagang, Jiangsu, China) was used as the fermentation catalyst at an application rate of 100 g per 20-L litter chips. Eight 60-L vessels (top diameter \times bottom diameter \times height, 39 cm \times 35 cm \times 53 cm) (Qinglong Plastic Production Ltd., Changzhou, Jiangsu, China) were used for composting with alternatively additions of litter chips and EM powder. Four vessels were used for composting litter from plantations and the other four for litter from secondary forests. An aperture in diameter of 2 cm was made at the centre of bottom at a height of 7 cm aboveground to leak fermentation secretion. Tops of vessels were covered by plastic sheets with a hole left in the centre for penetrating an erected temperature meter hanged overhead. Vessels were placed in a greenhouse roofed by diaphanous plastic sheets where air temperature ranged between 32 °C and 56 °C (night/day). To keep greenhouse at warm temperature at night, firewood was continuously burnt since sunset. Aerobic composting persisted for three weeks when litter chips composted at an inner temperature of 72 ± 3 °C. Decomposed litter chips shrank in volume with surface height in vessels continuously decline until to about half of that at the commencement.

Seedling culture and experiment layout

A total of 240 one-year-old Chinese pine and Liaodong oak seedlings were cultured as plant materials tested in this study. In late April, 2022, seedlings with an uniform size per species were chosen for transplant. Some oak seedlings had acorns attached to lateral roots, which were excised off to eliminate impacts on nutrient uptake through remobilization ruled in response to soil fertility (Gao *et al.*, 2021). Five initial Chinese pine seedlings grew with height of 4.52 ± 0.29 cm and root-collar diameter (RCD) of 2.13 ± 0.19 mm, which for initial Liaodong oak seedlings were measured to be 9.69 ± 1.03 cm and 2.41 ± 0.39 mm, respectively. Seedlings were transplanted to 1.83-L plant-growing pots (top diameter \times bottom diameter \times height, 14.7 cm \times 11.5 cm \times 13.5 cm), that were filled with three types of growing media of forest soils amended by decomposed litter applied at rates of 0 (control), 25%, and 50% (v/v). A total of 240 pots were used for an arrangement of number resulting from combined two forest types (plantation vs secondary forest), three decomposed litter ratios, and

two tested seedling species (Chinese pine vs Liaodong oak) with 20 replicates for every combined arrangement. Five germinant plantlets were transplanted to one pot and thinned to leave only one of them after one-week maintenance to screen for the one with a strong stem and an activated growing performance. Thinned pots were placed in a 0.5 m × 0.5 m spacing on seedling benches at a height of 7 cm aboveground in the same greenhouse where litter chips were composted. Sunshade net was used to roof greenhouse to control exposure to sunlight irradiation.

Potted seedlings were placed in a split-plot arrangement with forest type (plantation vs secondary forest) as the main plot and decomposed litter ratio as the sub-plot (control, 25%, and 50%). Initial chemical properties in growing media are shown in Table 1 for forest soils and decomposed litter with methods disclosed in a following paragraph. Two seedling species were arranged in a block of 40 pots (20 pots for a species), which were randomly placed to accounting for the random coefficient in the split-plot design. Totally, six blocks of potted seedlings were placed in a greenhouse where positions of every block or every pot per block were rearranged every week to eliminate the edge effect. Chinese pine seedlings have a slow growing rate as to ~8 cm in height needs a growing season at the cost of intensive use of chemical fertilizers (Shi *et al.*, 2019). Only in four year can Chinese pine seedlings grow to a height of about 50 cm (Li *et al.*, 2023). Oak seedlings also have a low growing rate with episodic growing patterns (Birge *et al.*, 2006). Therefore, supplemental lighting using light-emitting diode (LED) illumination was suggested to be used to accelerate growth of juvenile pine (Guo *et al.*, 2022; Li *et al.*, 2020a) and oak seedlings (Gao *et al.*, 2021). We employed LED lighting in this study with an expectation to promote seedling growth and exhibited more probably responses to combined factors. LED lamps were used for continuous lighting with a spectrum comprising 69.4% red light, 30.2% green light, and 0.4% blue light according to Duan *et al.* (2023). Seedlings started to be exposed to supplemental lighting one week after transplant. LED lamps were hanged 1.7 m aboveground and photoperiod photon flux density (PPFD) was measured to be ~70 $\mu\text{mol}^{-1} \text{m}^{-2} \text{s}^{-1}$ 45 cm beneath the lighting diodes. Supplemental lighting was used to prolong daily photoperiod up to 18 h during which inner PPFD in greenhouse was measured to be higher than 70 $\mu\text{mol}^{-1} \text{m}^{-2} \text{s}^{-1}$ 1.25 m aboveground. Local sunlight provided irradiation with sufficient PPFD in a daily time of 12 h from 07:00 am to 19:00 pm, which was extended by supplemental lighting to 01:00 am the next morning. This photoperiod regime has been used in previous studies for supplemental plant culture (An *et al.*, 2018; Duan *et al.*, 2023; Gao *et al.*, 2021).

Table 1. Chemical properties in ratios of mixed soils and decomposed litter of *Pinus tabulaeformis* Carr. plantations and secondary forests dominated by *Quercus liaotungensis* Koidz.

Forest type	Soil	OM ¹ (%)	pH	TN ² (g/kg)	TP ³ (g/kg)	NH ₄ -N ⁴ (mg/kg)	NO ₃ -N ⁵ (mg/kg)	AP ⁶ (mg/kg)
Plantation	Control ⁷	10.11 ⁸	5.74	3.42	1.12	28.32	93.15	3.92
	25%	8.70	5.56	2.73	0.77	18.64	134.15	34.35
	50%	9.66	5.78	3.08	0.96	15.17	127.38	60.18
	Litter	55.85	6.78	14.57	3.06	27.78	127.21	1250.61
Secondary forest	Control	7.23	5.53	2.61	0.69	17.88	130.17	8.55
	25%	11.42	5.78	3.74	1.22	24.45	100.46	30.12
	50%	11.39	5.84	3.65	1.28	31.22	39.14	41.14
	Litter	54.91	6.99	13.37	2.74	27.16	126.47	1249.66

¹ OM, organic matter; ² TN, total nitrogen content; ³ TP, total phosphorus content; ⁴ NH₄-N, ammonium nitrogen (N) content; ⁵ NO₃-N, nitrate N content; ⁶ AP, available phosphorus (P) content; ⁷ ratios of decomposed litter to forest soils in 0% (control), 25%, and 50%; ⁸ all values are averaged from five replicated samples.

Seedlings were watered every 2-3 days according to daily temperature. Inner temperature was controlled to be lower than 40 °C in daytime by rolling up plastic covers of side walls and employing sunshade nets. Potted seedlings were watered at a daily rate of 180 ml per pot. This amount was estimated through a simulation of

natural rainfall to local regenerated seedlings in plantations. As it was described above, annual precipitation was equal to a rate of 98.96 mm per stem per day, which was further equal to a pot's amount considering the proportional decline of volume (1.83 L) for a pot relative to 1 stere of rainfall.

Sampling and chemical analysis

The experiment was ended up in mid-September, when apical buds were formed and seedlings were about to fall in deep hardening dormancy (Bergervoet *et al.*, 1999). Ten pots of seedlings were randomly selected from a combined treatment per block. Height and RCD were measured for each chosen seedling, which was subsequently divided to shoot and root parts. Shoot part included leaves (needles), twig (especially for oak) and stem. Both shoot and root parts were dried in an oven for 72 h, cooled down to room temperature, and measured for dry mass biomass. A 0.2 g sample was digested in 5 mL mixture of 98% (v/v) sulfuric acid (H₂SO₄) and hydrogen peroxide (H₂O₂) and used for determining total N concentration using the classic Kjeldahl method (An *et al.*, 2018). Total P concentration was determined using another 0.2 g sample digested in mixed H₂SO₄ and perchloric acid and analysed using Inductively Coupled Plasma Optical Emission spectroscopy (ICP-OES) (Vista MPX, Varian Medical System Inc., Palo Alto, U.S.) (Xu *et al.*, 2019).

Substrates of sampled seedlings were also collected to air dried, pass 2 mm sieve, and used for chemical analysis. Chemical properties in soils, composed litter, and post-experiment substrates were determined according to previous studies (Duan *et al.*, 2023; Wei *et al.*, 2017). Total N and P concentrations (both in plants and for growing media) were determined using a 1.0 g dried sample and employed the same methods as described above. Mineral N (identified as ammonium N [NH₄⁺-N] and nitrate N [NO₃⁻-N]) was determined by extracting from 2 mM KCl solution and quantified using the flow injection instrument (Lachat Instrument, Hach Inc., Loveland, CO, USA). Available P content was determined using a 5 g sample extracted with 50mL distilled water and analysed through ICP-OES as described above. Organic matter was determined using the titration method with dichromate oxidation and ferrous ammonium sulfate. The pH value was measured by a meter (3020 pH indicator, Jenway Inc., Dunmow, U.K.).

Parameter calculation and statistical analysis

Nutrient utilization index was calculated for crimitive N (NUI) and P elements (PUI) (Wang *et al.*, 2017; Zhao *et al.*, 2019):

$$\text{NUI} = \frac{\text{Mass}_{\text{Shoot}}}{\text{TN}_{\text{Shoot}}} \quad (1)$$

$$\text{PUI} = \frac{\text{Mass}_{\text{Shoot}}}{\text{TP}_{\text{Shoot}}} \quad (2)$$

where, Mass_{Shoot} is the total mass in shoot part; TN_{Shoot} and TP_{Shoot} are total N and P concentrations (%) in shoot parts of seedlings, respectively. Dickson quality index (DQI) was employed to assess comprehensive seedling quality (Li *et al.*, 2018):

$$\text{DQI} = \frac{\text{Mass}_{\text{Whole}}}{\left(\frac{\text{Height}}{\text{RCD}} + \frac{\text{Mass}_{\text{Shoot}}}{\text{Mass}_{\text{Root}}}\right)} \quad (3)$$

where, Mass_{Whole} and Mass_{Root} are biomass for whole plant and root, respectively. Nutrient content was calculated as the product of concentration and biomass. Nutrient states of seedlings subjected to varied decomposed litter ratios in growing media were characterized with vector analysis (An *et al.*, 2018). With the control (no decomposed litter) as the reference, the 25% and 50% ratios were diagnosed for their relative N

and P states estimated by synthesizing relative changes of biomass, nutrient concentration (x-axis), and nutrient content (y-axis). Relative nutritional changes were diagnosed not only in main effects of decomposed litter ratio, but also with combined effects of forest type and seedling species. Interpretations of vectors were adapted from Salifu and Timmer (2003) (Salifu and Timmer, 2003).

Data were tested to follow normal distributions with homogeneous variances; hence no transformation was needed. Data were analysed using a split-plot modelling design with forest type (plantation vs secondary forest) as the main plot and ratio of decomposed litter (control, 25%, and 50%) as the sub-plot. Two tested seedling species (pine vs oak) were nested to sub-plots whose random placement was taken as the random factor. Ten replicates were embedded to each combined treatment and one randomly sampled seedling accounted for a replicate per treatment. Three-way analysis of variation (ANOVA) was used to detect combined effects of forest type, decomposed litter ratio, seedling species, and their interactions on seedling parameters. When significant interactive effects were detected, results were compared by one-way ANOVA with combined effects incorporated to factors-synthesized treatments. Tukey test was employed for comparison and the critical value of 0.05 was used to characterize the probability of significance. Multivariate linear regression was used to detect combined contributions from growing media characters to seedling parameters to detect factors in growing media that shaped seedling growth and nutrient uptake.

Results

Seedling growth and comprehensive quality assessment

Both decomposed litter ratio and seedling species variation had main effects on seedling height and RCD (Table 2), whose results are shown in Figure 2.

Table 2. Analysis of variation (ANOVA) on growth in seedling height, root-collar diameter, biomass accumulation in *Pinus tabuliformis* Carr. and *Quercus liaotungensis* Koidz. seedlings according to a split-plot model with forest type as the main plot and decomposed litter ratio as the sub-plot nested by seedling species

Source of variation	df ¹	Seedling height		RCD ¹		Shoot biomass		Root biomass	
		F value	p value	F value	p value	F value	p value	F value	p value
Forest type (F)	1	0.02	0.8766	0.52	0.4743	0.52	0.4738	1.74	0.1904
Decomposed litter ratio (D)	2	5.26 ²	0.0066	7.27	0.0011	38.32	<0.0001	9.84	0.0001
Seedling species (S)	1	510.78	<0.0001	23.09	<0.0001	514.34	<0.0001	187.27	<0.0001
F × D	2	0.25	0.7822	1.42	0.2460	2.99	0.0543	1.8	0.1707
D × S	2	0.23	0.7931	0.56	0.5746	8.66	0.0003	2.48	0.0887
F × D × S	3	0.24	0.8652	0.72	0.5424	0.34	0.796	2.85	0.0408

¹ df, degree of freedom; RCD, root-collar diameter; ² values in bold font indicate significant effects on the variable.

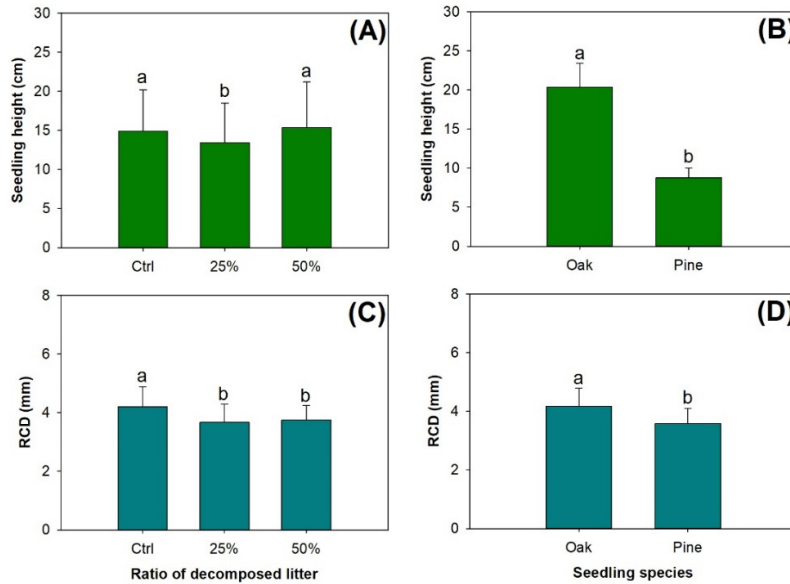


Figure 2. Growth in seedling height (A, B) and root-collar diameter (RCD) (C, D) in *Quercus liaotungensis* Koidz. (Oak) and *Pinus tabuliformis* Carr. (Pine) seedlings subjected to different ratios of decomposed litter addition at 0% (Ctrl), 25%, and 50%. Columns mark means with error bars presenting standard errors. Different letters above bars indicate significant differences identified by Tukey test at 0.05 level.

Combined factors of forest type, decomposed litter ratio and seedling species had interactive effects on root biomass (Table 2). Combined treatments of these three factors were also found to have a significant effect on root biomass according to one-way ANOVA (Sum of squares [SS]: 28.96; *F* value: 20.53; *p* value < 0.0001). These combined treatments were also identified to have significant effect on shoot biomass (SS: 34.00; *F* value: 55.99; *p* value < 0.0001).

Shoot and root biomass results in response to all combined treatments are shown in Figure 3. Compared to pine seedlings, oak seedlings had overall lower shoot biomass (mean \pm standard error; pine, 1.59 ± 0.32 g; oak, 0.62 ± 0.16 g), and shoot biomass was not significantly different among oak seedlings subjected to combined litter addition and forest type (Figure 3A). Compared to the control, decomposed litter addition decreased shoot biomass either in a ratio of 25% or in 50% (control, 1.37 ± 0.56 g; 25%, 0.98 ± 0.38 g; 50%, 0.96 ± 0.42 g).

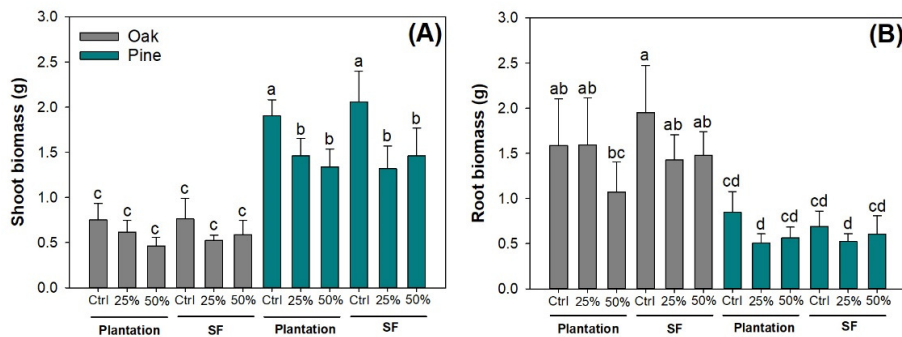


Figure 3. Biomass increments in shoot (A) and root parts (B) in *Q. liaotungensis* Koidz. (Oak) and *P. tabuliformis* Carr. (Pine) seedlings subjected to different ratios of decomposed litter addition at 0% (Ctrl), 25%, and 50% in two forest types of pine plantation vs secondary oak forests (SF). Columns mark means with error bars presenting standard errors. Different letters above bars indicate significant differences identified by Tukey test at 0.05 level.

Root biomass reversed to be greater in oak seedlings than in pine seedlings (oak, 1.52 ± 0.43 g; pine, 0.62 ± 0.17 g). Controlled oak seedlings with litter addition from secondary forests also had greater root biomass than pine seedlings. Again, root biomass was greater in controlled seedlings (1.27 ± 0.56 g) compared to those receiving decomposed litter in ratios of 25% (1.01 ± 0.50 g) and 50% (0.93 ± 0.38 g). Root to shoot ratio (RS) was only responsive to seedling species variation (*F* value: 370.28; *p* value < 0.0001). Oak seedlings had a higher RS (2.58 ± 0.02) than pine seedlings (0.41 ± 0.02). Decomposed litter ratio had a main effect on DQI (*F* value: 4.79; *p* value: 0.0102), whose combination with forest type also had an interactive effect on DQI (*F* value: 5.44; *p* value: 0.0056). Decomposed litter from plantations at the ratio of 50% lowered DQI compared to the control (Figure 4). DQI was not statistically difference among litter ratios from secondary forests. Compared to no-litter control, decomposed litter addition at 50% ratio reduced DQI by 21.71% (0.48 ± 0.16 and 0.38 ± 0.12 , respectively), but the 25% ratio did not cause any significant difference (0.42 ± 0.10).

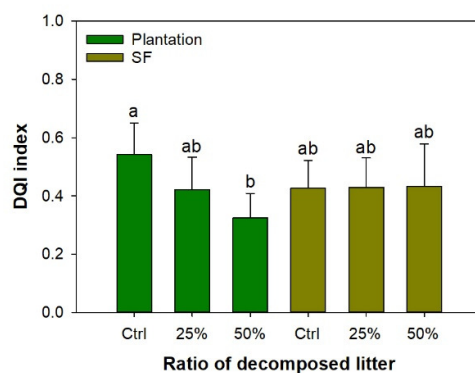


Figure 4. Comprehensive quality assessment through Dickson quality index (DQI) in *Q. liaotungensis* Koidz. (Oak) and *P. tabuliformis* Carr. (Pine) seedlings subjected to different ratios of decomposed litter addition at 0% (Ctrl), 25%, and 50% from two forest types of pine plantation vs secondary oak forests (SF). Columns mark means with error bars presenting standard errors. Different letters above bars indicate significant differences identified by Tukey test at 0.05 level.

N and P concentrations

Factors of forest type, decomposed litter ratio, and seedling species had significant combined effects on shoot N concentration (Table 3). Pine seedlings had generally higher shoot N concentration compared to oak seedlings (pine, 13.89 ± 2.44 mg g⁻¹; oak, 7.12 ± 1.43 mg g⁻¹). Results about N and P concentrations in shoots and roots of pine seedlings and oak seedlings are shown in Figure 5A, B. Combined effects of forest type, decomposed litter ratio, and seedling species on shoot P concentration are shown in Figure 5 C, D.

Table 3. ANOVA on nitrogen (N) and phosphorus (P) concentrations and contents in shoots and roots in *P. tabuliformis* Carr. and *Q. liaotungensis* Koidz. seedlings according to a split-plot model with forest type as the main plot and decomposed litter ratio as the sub-plot nested by seedling species.

Source of variation	Shoot N concentration		Root N concentration		Shoot P concentration		Root P concentration	
	<i>F</i> value	<i>p</i> value	<i>F</i> value	<i>p</i> value	<i>F</i> value	<i>p</i> value	<i>F</i> value	<i>p</i> value
Forest type (F)	71.87	<0.0001 ¹	0.30	0.5839	0.40	0.5305	0.21	0.6472
Decomposed litter ratio (D)	19.98	<0.0001	4.12	0.0189	16.51	<0.0001	0.21	0.8140
Seedling species (S)	475.95	<0.0001	7.42	0.0075	230.97	<0.0001	11.77	0.0009
F × D	0.34	0.7153	2.08	0.1299	2.09	0.1285	3.78	0.0259
D × S	8.91	0.0003	3.08	0.0499	0.68	0.5073	0.45	0.6371
F × D × S	2.77	0.0453	1.20	0.3125	2.57	0.0578	4.85	0.0033
	Shoot N content		Root N content		Shoot P content		Root P content	

	<i>F</i> value	<i>p</i> value	<i>F</i> value	<i>p</i> value	<i>F</i> value	<i>p</i> value	<i>F</i> value	<i>p</i> value
Forest type (F)	19.02	<0.0001	0.01	0.9342	0.30	0.5880	0.66	0.4186
Decomposed litter ratio (D)	3.43	0.0359	6.01	0.0033	6.88	0.0015	8.20	0.0005
Seedling species (S)	672.44	<0.0001	69.52	<0.0001	724.98	<0.0001	194.89	<0.0001
F × D	2.12	0.1251	2.40	0.0955	0.08	0.9230	2.08	0.1302
D × S	2.63	0.0768	4.39	0.0146	3.52	0.0330	2.69	0.0721
F × D × S	3.22	0.0256	1.86	0.1409	0.86	0.4640	2.66	0.0521

¹ Values in bold font indicate significant effects on the variable.

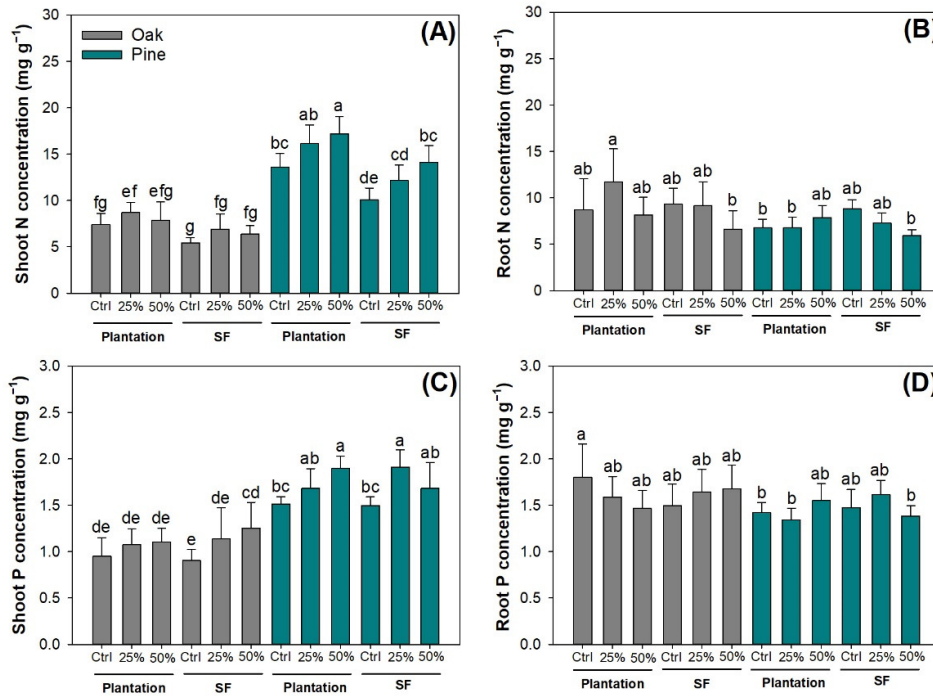


Figure 5. Nitrogen (N) and phosphorus (P) concentrations (A, B) and contents (C, D) in shoot (left) and root parts (right) in *Q. liaotungensis* Koidz. (Oak) and *P. tabuliformis* Carr. (Pine) seedlings subjected to different ratios of decomposed litter addition at 0% (Ctrl), 25%, and 50% in two forest types of pine plantation vs secondary oak forests (SF). Columns mark means with error bars presenting standard errors. Different letters above bars indicate significant differences identified by Tukey test at 0.05 level.

Relative N and P Nutritional States and Utilizations

Nutritional vector diagnosis indicated that decomposed litter addition at ratios of 25% and 50% both resulted in relative states of excessive nutritional supplies for N and P elements (Figure 6 A, B). No matter source of litter was derived from either plantations or secondary forests, decomposed litter addition still resulted in excessive nutritional states in reference to the control (Figure 6 C, D). Again, both oak and pine seedlings showed relatively excess of N and P supplies in litter treatments at two ratios relative to the control (Figure 6 E, F).

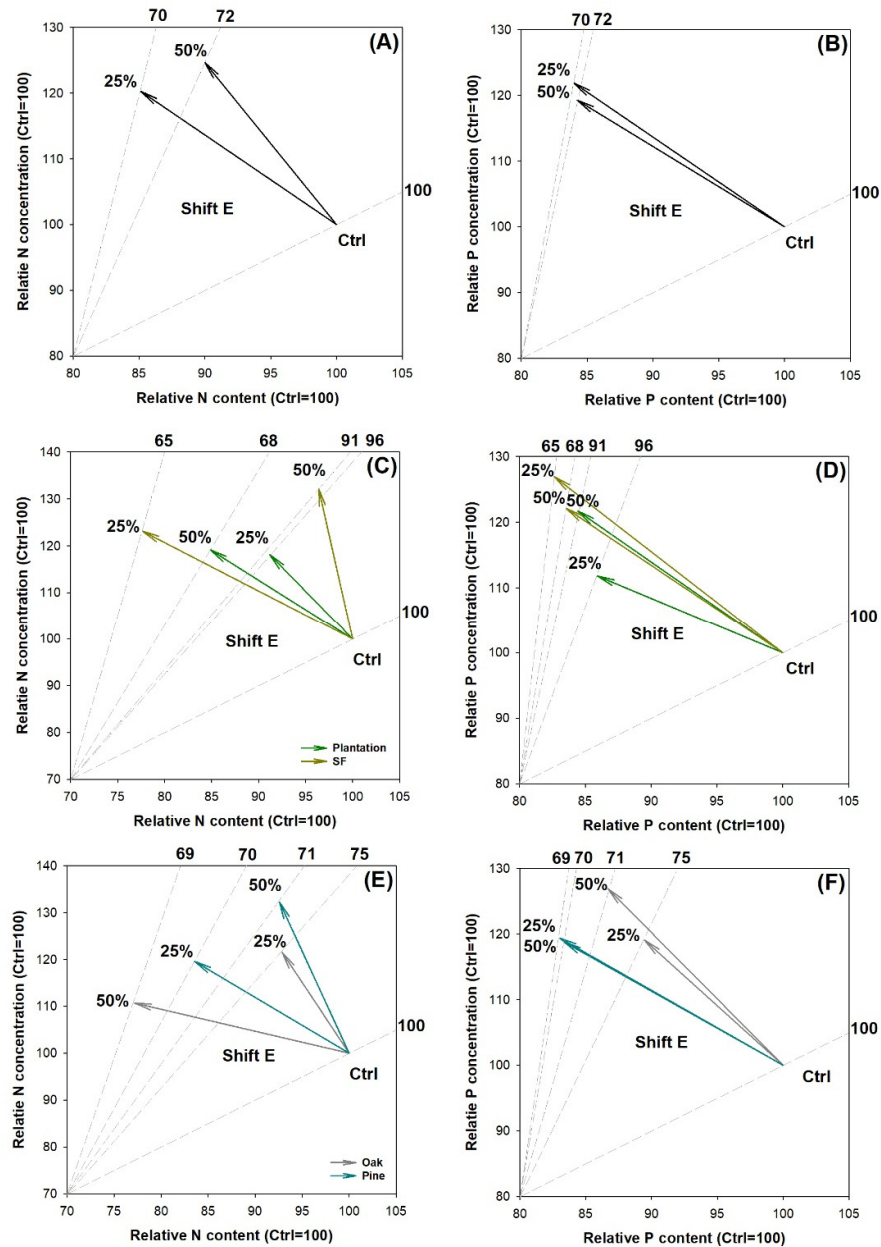


Figure 6. Vector nomographs of N (A, C, E) and P (B, D, F) nutritional states of *Q. liaotungensis* Koidz. (Oak) and *P. tabuliformis* Carr. (Pine) seedlings subjected to different ratios of decomposed litter addition at 0% (Ctrl), 25%, and 50% in two forest types of pine plantation vs secondary oak forests (SF). No litter control is taken as the reference from which relative changes in N and P contents are lined in vectors along the x-axis and in N and P concentrations are lined in vectors along the y-axis. Biomass of reference is taken as a baseline of 100 and values of biomass in litter-addition treatments were valuated proportionally relative to the baseline. Shift E is interpreted as a result of excess nutrition supply with possible diagnostic of toxic accumulation due to overdose of nutrient loading according to Salifu and Timmer (2003).

Decomposed litter addition had combined effects on NUI with source of forest type (F value: 3.32; p value: 0.0399) and seedling species (F value: 3.79; p value: 0.0256). Compared to the control, decomposed litter addition decreased NUI and PUI for two sources in plantations and secondary forests (Figure 7 A, C). Controlled seedlings receiving no decomposed litter in secondary forests showed higher NUI than those in plantations. Again, seedlings with decomposed litter addition showed lower NUI in both oak and pine seedlings, and controlled pine seedlings had higher NUI and PUI than controlled oak seedlings (Figure 7 B, D).

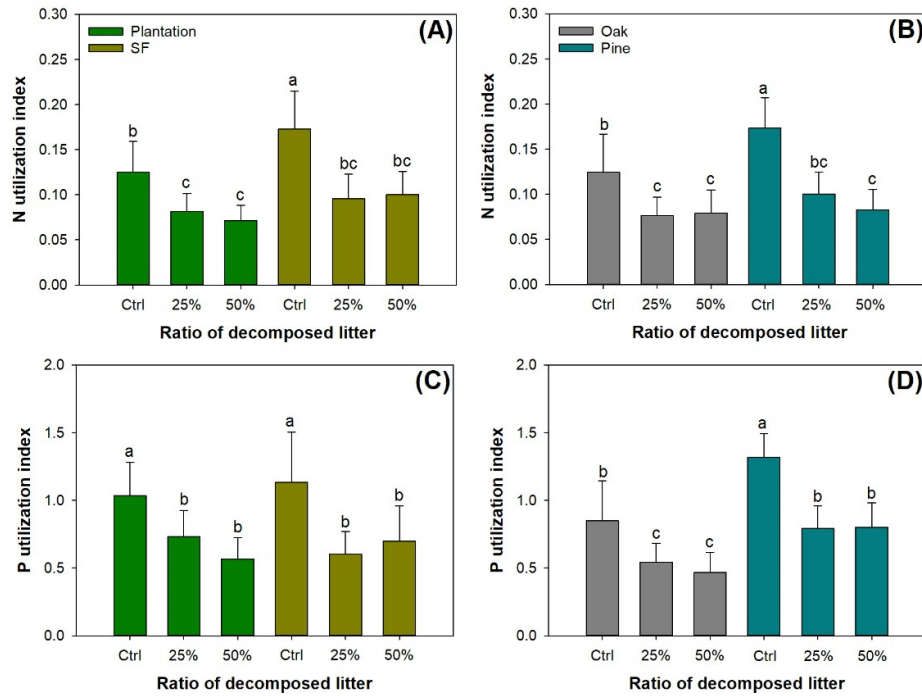


Figure 7. Nutrient utilization indices for N (NUI) (A, B) and P (PUI) (C, D) in shoots of *Q. liaotungensis* Koidz. (Oak) and *P. tabuliformis* Carr. (Pine) seedlings subjected to different ratios of decomposed litter addition at 0% (Ctrl), 25%, and 50% in two forest types of pine plantation vs secondary oak forests (SF). Columns mark means with error bars presenting standard errors. Different letters above bars indicate significant differences identified by Tukey test at 0.05 level.

Chemical properties in growing media

Decomposed litter ratio had combined effects with forest type or seedling species on most chemical property parameters in growing media (Table 4). Combined effects of forest type and litter addition ratio on substrate properties are shown in Figure 8. Combined effects of seedling species and litter ratio on substrate properties are shown in Figure 9.

Table 4. ANOVA on physical and chemical properties in substrates for planting *P. tabuliformis* Carr. and *Q. liaotungensis* Koidz. seedlings according to a split-plot model with forest type as the main plot and decomposed litter ratio as the sub-plot nested by seedling species

Source of variation	pH		OM ¹		Total N ² content		Total P ³ content	
	F value	p value	F value	p value	F value	p value	F value	p value
Forest type (F)	53.28	<0.0001 ⁴	188.97	<0.0001	108.16	<0.0001	692.96	<0.0001
Decomposed litter ratio (D)	463.18	<0.0001	80.67	<0.0001	157.90	<0.0001	186.56	<0.0001
Seedling species (S)	0.31	0.5798	0.66	0.4172	0.94	0.3339	0.07	0.7947
F × D	120.33	<0.0001	0.96	0.3846	4.78	0.0103	31.80	<0.0001
D × S	23.63	<0.0001	0.26	0.7722	0.07	0.9337	0.46	0.6337
F × D × S	1.03	0.3812	1.05	0.3755	0.39	0.7585	1.29	0.2826
	Ammonium N content		Nitrate N content		Available P content			
	F value	p value	F value	p value	F value	p value		
Forest type (F)	15.58	0.0001	115.88	<0.0001	64.73	<0.0001		
Decomposed litter ratio (D)	4.14	0.0185	28.57	<0.0001	427.74	<0.0001		
Seedling species (S)	0.05	0.8283	1.66	0.2006	0.38	0.5409		
F × D	0.68	0.5075	42.36	<0.0001	2.91	0.0586		
D × S	2.23	0.1130	12.69	<0.0001	0.65	0.5218		
F × D × S	2.40	0.0717	1.72	0.1664	1.17	0.3241		

¹ OM, organic matter; ² N, nitrogen; ³ P, phosphorus; ⁴ Values in bold font indicate significant effects on the variable.

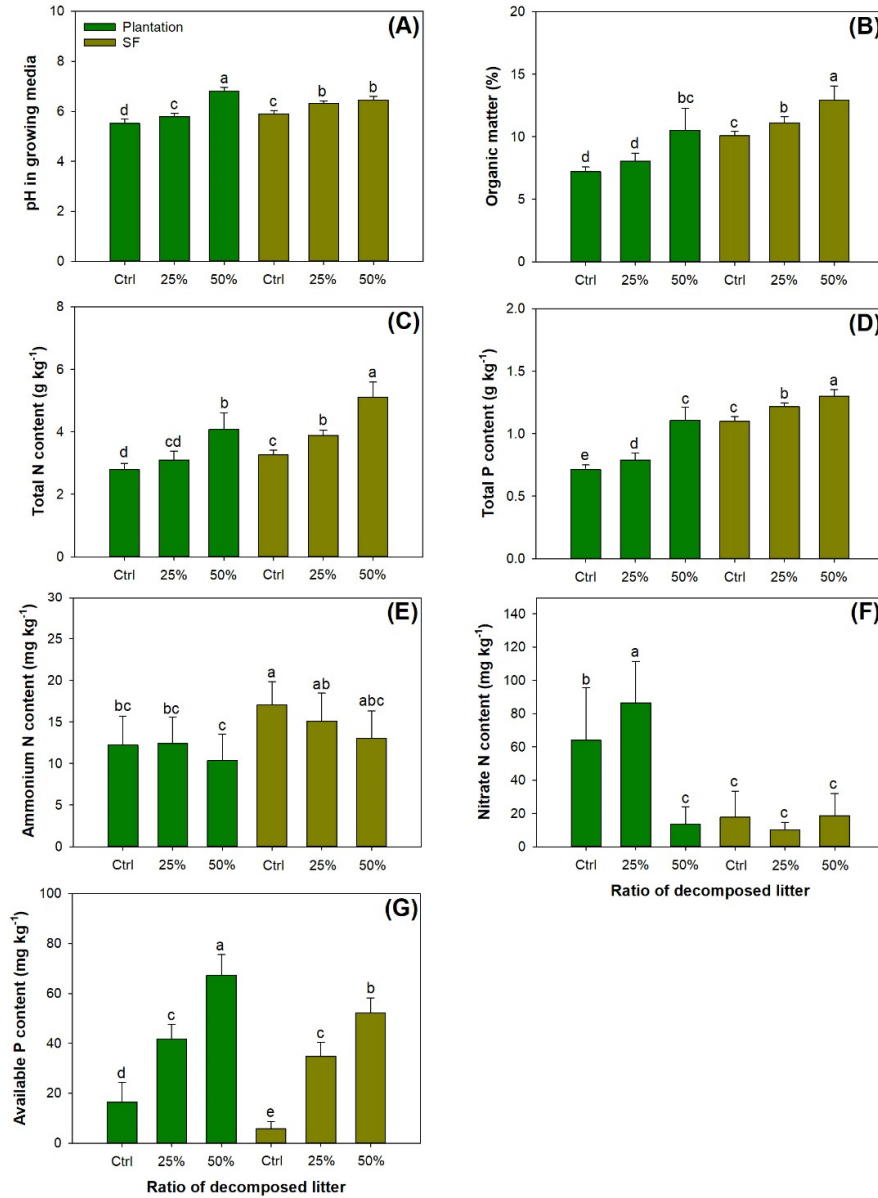


Figure 8. Soil property parameters in growing media for *Q. liaotungensis* Koidz. (Oak) and *P. tabuliformis* Carr. (Pine) seedlings subjected to different ratios of decomposed litter addition at 0% (Ctrl), 25%, and 50% in two forest types of pine plantation vs secondary oak forests (SF). Columns mark means with error bars presenting standard errors. Different letters above bars indicate significant differences identified by Tukey test at 0.05 level.

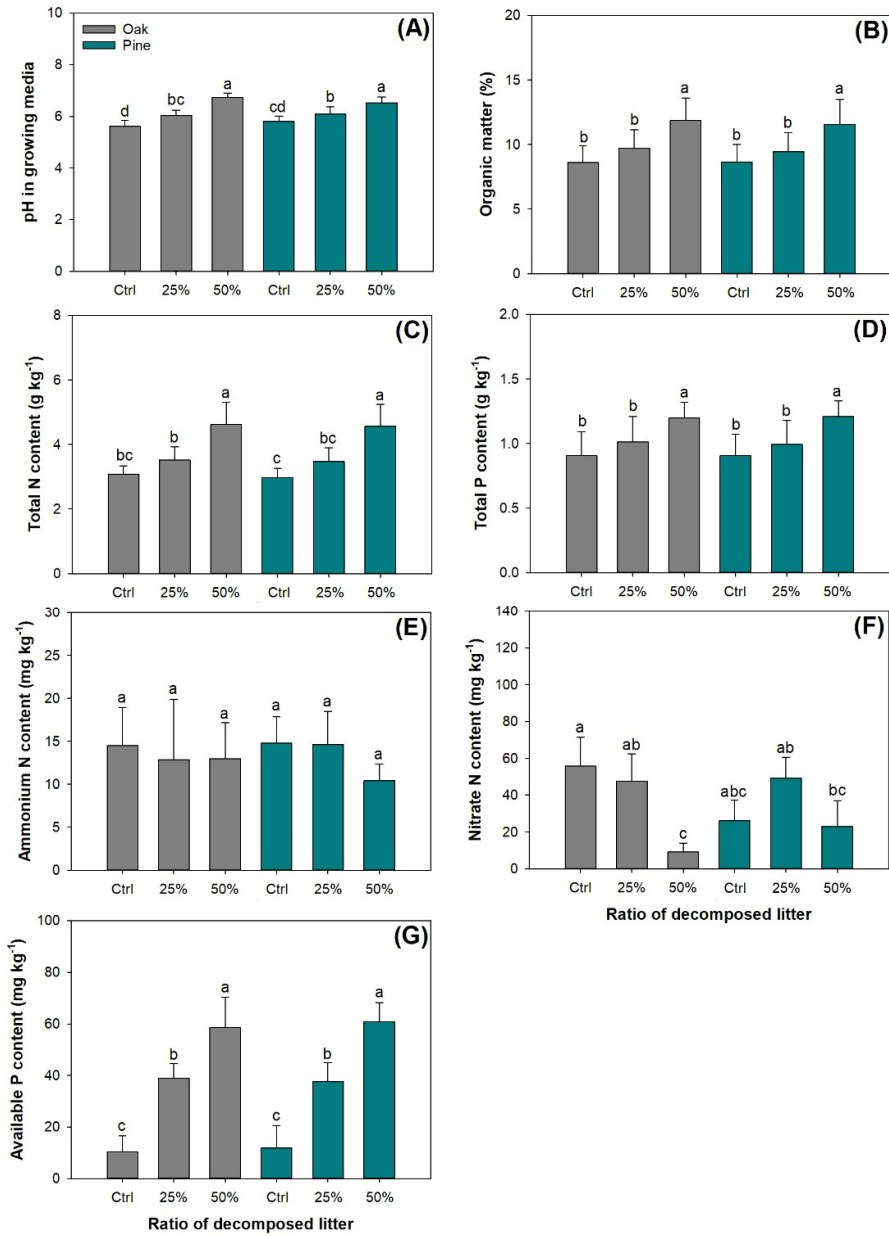


Figure 9. Soil property parameters in growing media for *Q. liaotungensis* Koidz. (Oak) and *P. tabuliformis* Carr. (Pine) seedlings subjected to different ratios of decomposed litter addition at 0% (Ctrl), 25%, and 50%. Columns mark means with error bars presenting standard errors. Different letters above bars indicate significant differences identified by Tukey test at 0.05 level.

Driving forces of growing media for seedling parameters

No chemical property in growing media contributed to seedling height, while pH generated negative contributions to RCD and biomass in shoot and root parts (Table 5). In addition, nitrate N content also generated a jointly negative contribution to shoot biomass, which showed a stronger driving force than pH.

Organic matter generated negative contributions to N content and concentration in seedling shoots with a jointly positive contribution of ammonium N content to shoot N concentration (Table 5). In addition,

ammonium N content also had a positive contribution of nitrate N content to root N concentration with a jointly positive contribution. Nitrate N content in growing media had a positive contribution to root N concentration with a jointly negative contribution from available P content.

Available P content in growing media had a tiny positive contribution to shoot P concentration (Table 5). Again, pH had a negative contribution to root P content.

Table 5. Multivariate linear regression of plant variables against chemical properties in growing substrates using combined forest soils and decomposed litters for *P. tabuliformis* Carr. and *Q. liaotungensis* Koidz. seedlings

Independent	Height			RCD ¹			ShootBio ²			RootBio ³		
	PE ⁴	SE ⁵	F ⁶	PE	SE	F	PE	SE	F	PE	SE	F
Intercept	14.57	0.59	605.84*** ⁷	5.94	0.91	43.03***	4.91	0.99	24.79***	3.01	0.71	18.03***
pH				-0.34	0.15	5.25*	-0.59	0.15	14.88**	-0.32	0.12	7.52**
OM ⁸												
TN ⁹												
TP ¹⁰												
AN ¹¹												
NN ¹²							-5.58×10 ⁻³	1.94×10 ⁻³	8.25**			
AP ¹³												
	ShootNc ¹⁴			RootNc ¹⁵			ShootNt ¹⁶			RootNt ¹⁷		
	PE	SE	F	PE	SE	F	PE	SE	F	PE	SE	F
Intercept	13.12	1.66	62.81***	5.29	0.85	38.67***	20.97	4.07	26.50***	9.97	1.58	39.74***
pH												
OM	-0.54	0.17	9.61**				-0.80	0.40	4.01*			
TN												
TP												
AN	0.08	0.02	19.22***	0.15	0.06	7.33**						
NN				0.02	0.01	6.06*				0.05	0.02	5.86*
AP										-0.07	0.03	5.00*
	ShootPc ¹⁸			RootPc ¹⁹			ShootPt ²⁰			RootPt ²¹		
	PE	SE	F	PE	SE	F	PE	SE	F	PE	SE	F
Intercept	1.22	0.07	301.53***	1.54	0.02	428.62***	1.65	0.10	272.29***	4.94	1.19	17.30***
pH										-0.53	0.19	7.61**
OM												
TN												
TP												
AN												
NN												
AP	4.55×10 ⁻³	1.65×10 ⁻³	7.55**									

¹ RCD, root-collar diameter; ² ShootBio, shoot biomass; ³ RootBio, root biomass; ⁴ PE, parameter estimate; ⁵ SE, standard error; ⁶ F, F value for evaluating PE; ⁷ number of asterisk indicates level of significance: *, $p < 0.05$; **, $p < 0.01$; ***, $p < 0.0001$; ⁸ OM, organic matter; ⁹ TN, total nitrogen (N) content; ¹⁰ TP, total phosphorus (P) content; ¹¹ AN, ammonium N content; ¹² NN, nitrate N content; ¹³ AP, available P content; ¹⁴ ShootNc, shoot N concentration; ¹⁵ RootNc, root N concentration; ¹⁶ ShootNt, shoot N content; ¹⁷ RootNt, root N content; ¹⁸ ShootPc, shoot P concentration; ¹⁹ RootPc, root P concentration; ²⁰ ShootPt, shoot P content; ²¹ RootPt, root P content.

Discussion

Growth and biomass in regenerated seedlings

It is generally considered that a regional condition for local seedling growth should be accompanied with natural exposure to sunlight spectrum and its photoperiod. Chinese pine and Liaodong oak belong to slowly growing species compared other local trees, such as larch and ash (Wei *et al.*, 2012; Wei *et al.*, 2013b; Yin *et al.*, 2023). The low growing rate will limit performances of our pine and oak seedlings in response to properties in manipulated growing substrates which may not be as apparent as expected in a short term as well. Therefore, we employed LED lighting to supplement light and extend photoperiod which can promoted seedling growth

and relevantly physiological activities (Li *et al.*, 2017; Li *et al.*, 2018; Wei *et al.*, 2013a). The spectrum we used in this study looked with a red-light tinted colour. A similar spectrum with red-light component of about 70% was demonstrated to promote growth in *Aronia melanocarpa* (Duan *et al.*, 2023), *Quercus variabilis* Blume (Gao *et al.*, 2021), *Capsicum frutescens* L. var. *fasciculatum* (Sturt.) Baily, and *Capsicum frutescens* L. var. *longum* Bailey (Wang *et al.*, 2022b). Therefore, our illumination environment was an explicitly designed arrangement that can mostly implement the experiment following our design.

In this study, soils were amended by decomposed litter and the decomposition was enforced as a replacement of compost. Restored seedling height matters for the height of regeneration layer and thereafter the competition for light resources and success of regeneration (Kuehne *et al.*, 2020; Magnoux *et al.*, 2018). Root collar growth was usually taken as a strong predictor of the ability of regenerations to establish in stands (Searle *et al.*, 2022; Wei *et al.*, 2021). The addition of decomposed litter to growing media at a ratio of 25% reduced both height and RCD in seedling compared to the control. These suggest that decomposed litter impaired shoot growth. This inhibitory effect on shoot morphology concurred with that found on shoot biomass, especially in pine seedlings. Biomass in pine seedlings was more retained in shoot parts compared to oak seedlings. No allelopathic effect was speculated to account for these inhibitions. For example, it was found that decomposed litter of *Gaultheria shallon* Pursh had showed no significant effect on growth in *Tsuga heterophylla* (Raf.) Sarg. and *Thuja plicata* Donn ex D, Don (Mallik and Prescott, 2001). The allelopathic effect of leaf litter on seedling growth was found on herbaceous plants. An instance indicated that leaf litter of *Lantana camara* inhibited the growth of *Raphanus sativus*, *Lactuca sativa*, *Bidens pilosa*, *Bidens bipinnata* and *Urena lobata* (Wang *et al.*, 2015). The inhibition effect of decomposed litter addition on shoot biomass of pine seedlings was found for both sources of litter collected from stands of plantations and secondary forests, and controlled pine seedlings did not show varied changes in shoot biomass between two sources of forest types. All these results together confirm that decomposed litter addition cannot benefit shoot growth in seedlings with activated shoot elongation.

Seedling quality assessment is a commonly used parameter that can be used to predict future performance of transplant seedlings. Dickson quality index was frequently employed as an assessment parameter for seedling culture at the late stage before transplant to screen the comprehensive quality of shaped seedlings (Li *et al.*, 2017). DQI was identified to synchronize with transplant seedling survival and growth (Parra and Maciel, 2018). Thus, this parameter was frequently used for assessing quality of artificially cultured seedlings (Li *et al.*, 2018; Wei *et al.*, 2020), but morphology and biomass in naturally regenerated seedlings can also be used to predict quality for further performance in secondary forests (Khurana and Singh, 2001; Tsakalidimi *et al.*, 2013). In our study, the 50% ratio of decomposed litter derived from Chinese pine plantations resulted in lower DQI compared to the control. Therefore, nutrient releases from decomposed litter did not benefit growth and biomass accumulation in Chinese pine and Liaodong oak seedlings at the regeneration layer. However, litter also functions to maintain soil moisture and mediate soil microbial communities, which are further determinative by regional meteorological factors (Martinez-Garcia *et al.*, 2021; Vos *et al.*, 2021). Rainfall may dilute high nutrient pool in soils and the change of temperature will modify the rate of microbial activities (Sherman *et al.*, 2012). Future work is encouraged to conduct investigations in natural forests to validate findings in this study.

Nutritional response to decomposed litter addition

Our vector analysis revealed that addition of decomposed litter resulted in excessive supplies of N and P. This negative effect remains to be the same case when being interacted with forest type or seedling species. These resulted from dual responses of increases in nutritional concentration in seedling shoots, but decreased biomass accumulation as described in preceding part. Regarding the decreased biomass in response to abundant nutrient supply, stand soils may have already had chemical properties that can well fulfil the demand for nutrient uptake by two seedling species; hence additional nutrient delivery through decomposed litter will

result in an excessive state. This highly meets the theoretical model of biomass responses to nutrient supply at an application stage from optimum to overdose (Salifu and Timmer, 2003; Xu *et al.*, 2019). Compared to other plantations and secondary forests in other regions of Northeast China, ammonium N content in soils (17.88–28.32 mg kg⁻¹) was comparable with that in Heilongjiang (14–20 mg kg⁻¹) (Zhou *et al.*, 2021) and other parts of Liaoning (3.5–16.7 mg kg⁻¹) (Yang *et al.*, 2014). Soil nitrate content in our study (93.15–130.17 mg kg⁻¹) was much higher than that in other studies (14–77 mg kg⁻¹) (Yang *et al.*, 2014; Zhou *et al.*, 2021). Hence, it was the abundant content of nitrate N in soils that fulfilled the demand for N. Soil available P content in our initial soils ranged in 3.92–8.55 mg kg⁻¹, which was comparable with that in another second forest in Liaoning (1.8–5.1 mg kg⁻¹) (Diao *et al.*, 2020; Zhang *et al.*, 2022). The explanation of P effect on seedlings was nearly the same of that of N.

Both N and P utilizations were found to be lower in litter addition treatments than in the control, which also demonstrated the negative effect of decomposed litter addition in accordance with above mentioned findings. Although the estimate of DQI involves parameters about seedling growth and biomass, it was illustrated that the change of this variable synchronized with changes in others reflecting responses of photosynthetic product and nutrient assimilation (Duan *et al.*, 2023; Li *et al.*, 2018). Regarding no difference of DQI was found between ratios of decomposed litter addition, it was litter addition that reduced nutrient utilization by seedlings no matter how much was mixed to soils.

Compared to oak seedlings, pine seedlings showed a smaller shoot stem size but greater shoot biomass accumulation. This was because pine seedlings were evergreen and preserved leaves accounted for the major body of shoot biomass. Oak is deciduous species with a nature to allocate more biomass to roots for N and P absorptions. This is the reason why pine seedlings showed higher NUI and PUI values compared to oak seedlings. Addition of litter, however, diminished the species-specific variation in NUI, but failed to reduce the high PUI in pine seedlings even with litter addition.

Seedlings in the control also showed significantly different utilizations in soils between forest types or seedling species. NUI was higher in soils from secondary forests than in plantations. Higher shoot N concentration in Chinese pine plantations and unchanged shoot biomass increment together resulted in a lower NUI in plantations. Newly grown seedlings in secondary forests can utilize N at a lower rate to produce shoot biomass which equals to that in plantations. These findings concur with those in previous studies (Cao and Chen, 2017; Wang *et al.*, 2022c), which demonstrated that temperate secondary forests had a higher N utilization efficiency in oak and pine seedlings compared to that in pine plantation.

Driving forces of chemical properties in growing media

No chemical properties in growth media accounted for seedling height growth, which may be due to the use of supplemental lighting as an acceleration of shoot elongation. Substrate pH value impaired RCD and biomass increments in shoot and root parts, which reflected the preference of acid soils by pine and oak seedlings in our study. Low root P content resulted from high pH which also indicated that the limit of P uptake by roots accounted for impairment of diameter and biomass increments in high pH soils.

Ammonium N content in growing media had no effects on growth and biomass increments, suggesting pine and oak seedlings did not utilize mineral N in this form. However, ammonium N content undertook the major contribution to N uptake, regarding findings that N concentration was increased in growing media with high ammonium N content. However, nitrate N content had an effect to induce biomass allocated from shoot to root; hence it promoted root N content and meanwhile reduced shoot biomass. Overall, soil ammonium N can benefit N uptake by oak and pine seedlings, but nitrate N induced more biomass allocated to roots for N uptake (Wei *et al.*, 2017; Zhou *et al.*, 2022).

Organic matter content in growing media showed negative contributions to shoot N content and concentration, which can be understood as an inhibition of N allocation upwards to shoot part. Regarding that soil OM showed no effect on root N uptake, it is meaningless to speculate that OM regulated N availability

through releasing rate (Duan *et al.*, 2022a). Three possible explanations may be involved. Firstly, decomposed litter contained a high content of OM, whose input impaired N uptake and allocation. Secondly, large amount of N was retained in OM without a sufficient rate of release which controlled N allocated to shoots. Finally, diversity of microbial communities in growing media was modified by high OM that inhibited N uptake (Duan *et al.*, 2022a). More work is needed to draw more confirmative conclusions.

Limits of this study

Our study has four limits that cannot be overcome by current experiment and findings. Firstly, our results should be validated by *in-situ* experiments and more investigations are encouraged to validate findings in this study in natural forests and plantations. Secondly, the two seedling species tested in this study both had slowly growing rates, which should be tested in a longer time of observation. Thirdly, macro-elements of N and P were tested in this study to examine their functions and cycles in litter-soil-plant, but many micro-elements are also released by decomposed litter and needed by regenerations. Future work should consider nutritional utilizations in a vaster spectrum of element species and detect a more comprehensive conclusion. Finally, we conducted this study in a greenhouse, where the impact of density was erased by an even arrangement of planting spacing. In real forests, however, understory regeneration highly depends on population density (Ali *et al.*, 2019; Wei *et al.*, 2019a; Wei *et al.*, 2021; Yang *et al.*, 2023), which should be considered in future experimental designs and arranged in new investigation layouts.

Conclusions

Our study was conducted in a controlled environment in a greenhouse where *P. tabuliformis* Carr. and *Q. liaotungensis* Koidz. Seedlings were cultured in growing media created by mixing *in-situ* soils and decomposed litter addition at ratios of 0%, 25%, and 50%. Generally, our results showed that decomposed litter addition would result in overdoses of N and P inputs and impaired N and P utilizations by both seedling species. The 50% ratio of litter addition should be avoided because it heavily decreased comprehensive seedling quality to compete at regeneration layer by enhancing facets against seedling growth and nutrient uptake, such as high pH value and high OM content. The high pH generated a negative effect on utilization, which resulted from inhibitions on biomass accumulation and P uptake. Overall, soils in Chinese pine plantations and Liaodong oak dominated secondary forests can well fulfil nutritional demands by Chinese pine and Liaodong oak seedlings. The full releases of N and P nutrients from decomposed litter may not be beneficial for regenerations of these two species in natural forests. Future work is suggested to identify our findings in fields of activated forests.

Authors' Contributions

Conceptualization: SG and RF; Data curation: SG and DR; Formal analysis: SG, DR and LT; Funding acquisition: SG; Investigation: DR and LT; Methodology: SG and RF; Project administration: SG; Resources: RF; Software: SG and DR; Supervision: SG; Validation: LT and RF; Visualization: DR and LT; Writing - original draft: SG; Writing - review and editing: RF. All authors read and approved the final manuscript.

Ethical approval (for researches involving animals or humans)

Not applicable.

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Conflict of Interests

The authors declare that there are no conflicts of interest related to this article.

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