Fine Root Distribution in Planted Robinia pseudoacacia Mixed Forests on Saline-Alkali Soils of the Yellow River Delta (Postprint)

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Abstract

To investigate the spatial distribution patterns of fine roots in artificial Robinia pseudoacacia mixed forests and pure forests on saline-alkali soils of the Yellow River Delta, Fraxinus velutina-R. pseudoacacia mixed forest, Ailanthus altissima-R. pseudoacacia mixed forest, and R. pseudoacacia pure forest were selected. Using the soil core method for sampling, the vertical distribution of tree fine roots in different stands on saline-alkali soils was examined in terms of fine root biomass density, surface area density, volume density, and root length density. The vertical distribution patterns of fine roots of different tree species were analyzed based on biomass, and differences in fine root distribution among plantations and soil influencing factors were investigated. The results showed that Fraxinus velutina-R. pseudoacacia mixed forest exhibited significantly higher fine root biomass, surface area, volume, and root length than both Ailanthus altissima-R. pseudoacacia mixed forest and R. pseudoacacia pure forest. In Fraxinus velutina-R. pseudoacacia mixed forest, 95.77% of fine root biomass was distributed in the 0–60 cm soil layer; in Ailanthus altissima-R. pseudoacacia mixed forest, 85.37% was distributed in the 0–40 cm layer; whereas fine roots in R. pseudoacacia pure forest were relatively evenly distributed, with 66.38% of biomass in the 0–40 cm layer. Fraxinus velutina displayed the highest fine root biomass, significantly exceeding other tree species. Fine root surface area density, volume density, and root length density in Fraxinus velutina-R. pseudoacacia mixed forest were significantly higher than in R. pseudoacacia pure forest; those in Ailanthus altissima-R. pseudoacacia mixed forest were higher than in R. pseudoacacia pure forest, but the difference was not significant. The total number of fine root tips in Fraxinus velutina-R. pseudoacacia mixed forest and Ailanthus altissima-R. pseudoacacia mixed forest was 2.34 and 1.23 times that of R. pseudoacacia pure forest, respectively, while the total number of forks was 6.15 and 1.66 times that of R. pseudoacacia pure forest, respectively.

Fine root biomass in all three forest types showed significant positive correlations with soil available phosphorus and available potassium contents; fine root biomass in Fraxinus velutina-R. pseudoacacia mixed forest showed extremely significant positive correlations with alkali-hydrolyzable nitrogen and organic matter contents. Appropriate mixed planting patterns enhanced plantation fine root biomass, improved plant capacity to absorb soil nutrients, and increased plantation adaptability to saline-alkali site conditions.

Full Text

Introduction

Roots are the organs that provide water and nutrients for normal plant growth, and their morphology and distribution directly reflect how trees adapt to site conditions, playing a decisive role in plant growth [?]. Root systems, especially fine roots (diameter $<$ 3 mm), play important roles in plant function and energy flow and material cycling in terrestrial ecosystems [?]. Although fine root biomass accounts for a relatively small proportion of total root biomass, it possesses a huge absorption surface area and serves as the primary organ for water and nutrient uptake. Differences in fine root biomass have a decisive impact on plant growth and development [?]. Fine roots exhibit relatively greater changes compared to roots of other diameter classes due to continuous growth and mortality, and they serve as important indicators of environmental change. The absorption of soil nutrients and water by fine roots and the quantity and quality of root exudates can all reflect plant health status [?]. Fine root biomass reliably reflects the capacity of root systems to absorb soil nutrients [?], making research on plant root systems highly significant. Roots are the underground portion of plants and are not easily studied. Although numerous studies on root systems have been reported in recent years [?], research on mixed forest root systems has gradually increased, suggesting that mixing significantly affects the fine roots of mixed species, with different outcomes for different species combinations [?]. For example, Qian et al. [?] found that fine root biomass of Korean pine (*Pinus koraiensis*) differed between pure and mixed forests, being higher in pure stands than in mixed stands with Manchurian walnut (*Juglans mandshurica*), but showing no significant difference compared to mixed stands with Manchurian ash (*Fraxinus mandschurica*), with mixing increasing the proportion of Korean pine fine roots in the surface soil layer (0–10 cm). Shi et al. [?] found that root biomass in mixed alder (*Alnus cremastogyne*) and cypress (*Cupressus funebris*) plantations was 1.5 times that of cypress pure stands. Zhai et al. [?] reported that in mixed stands of sand-located poplar (*Populus canadensis*) and black locust (*Robinia pseudoacacia*), black locust fine root biomass was greater than in pure stands, with poplar promoting black locust fine root growth and increasing the proportion of black locust fine root biomass in the mixed stand. Lin [?] found that fine root biomass and root length in black locust pure stands were lower than in mixed stands with tree-of-heaven (*Ailanthus altissima*) and velvet ash (*Fraxinus velutina*), with mixing causing surface aggregation of black locust fine roots. However, understanding of root distribution characteristics in different types of mixed plantations in saline-alkali soils remains limited.

The ecological environment of the Yellow River Delta is fragile with weak resistance to disturbance and poor adaptability to environmental changes, leading to severe ecosystem degradation [?]. To improve this situation, large-scale afforestation was conducted in the 1980s, primarily using salt-tolerant species such as black locust, velvet ash, and tree-of-heaven (*Ailanthus altissima*). These plantations have played important roles in soil and water conservation. Current research on plantations in this region has focused mainly on soil water-salt dynamics [?], biological communities [?], soil nutrients [?], and soil biochar changes [?]. Few studies have investigated fine root distribution in plantations, with only Du et al. [?] examining root distribution and fine root growth in mixed black locust and velvet ash plantations in saline-alkali soils, but lacking comparisons among different stand types. This study compares tree root conditions among different mixed stands and black locust pure stands from the perspective of fine root distribution, exploring spatial distribution differences in fine root biomass among different stands and planting patterns in the Yellow River Delta and the relationship between fine root biomass and soil characteristics, to provide theoretical basis and support for vegetation restoration and appropriate species selection in saline soils of the Yellow River Delta.

1 Study Site Description

The study site was located at the Yellow River Delta Production Base of the Jinan Military Region in Gudao Town, Hekou District, Dongying City, Shandong Province, with geographical coordinates of 118°39 -119°8 E, 37°47 -37°84 N, covering an area of approximately 2,366.7 hm^2. Gudao Forest Farm lies within the eastern beach area of the Yellow River North Dam, near the sea on the east and north sides. The region has a warm temperate semi-humid monsoon climate, with an average annual temperature of 12.8 °C, average annual ground temperature of 15.0 °C, average annual frost-free period of 234 days, frozen soil period of 44 days, average annual sunshine of 2,728.5 h, average annual relative humidity of 65%, average annual precipitation of 690.5 mm with uneven distribution (69% in summer and only 2% in winter), and effective accumulated temperature of approximately 4,300 °C [?]. The surface soil type in the experimental forest is salinized fluvo-aquic soil with the following basic physicochemical properties: pH 8.8 (soil:water ratio 1:5), available nitrogen content 75.4 mg · kg ^{1}, available phosphorus content 4.0 mg · kg ^{1}, available potassium content 92.6 mg · kg ^{1}, and total water-soluble salt content 0.08% [?].

The experimental plantations were 31-year-old artificial mixed forests: velvet ash and black locust mixed forest (FR), and tree-of-heaven and black locust mixed forest (AR). Both mixed forests were established in row patterns, with one row of velvet ash (or tree-of-heaven) alternating with one row of black locust. Black locust pure stands (RR) and black locust in mixed stands exhibited severe dieback and obvious missing trees, while dieback was not apparent in tree-of-heaven and velvet ash. Understory vegetation included cogongrass (*Imperata cylindrica*), silvergrass (*Triarrherca saccharifora*), and morning glory (*Pharbitis nil*). Tree growth status for each stand type is shown in Table 1 .

2.1 Fine Root Sampling

In November 2016, four 20 m × 20 m standard plots were established in each of the velvet ash and black locust mixed forest (FR), tree-of-heaven and black locust mixed forest (AR), and black locust pure forest (RR). Two adjacent standard trees were selected in each plot. Fine root sampling employed the soil column method [**?**]. Eight standard trees and four soil columns were sampled from each stand type, totaling 24 standard trees and 12 soil columns.

Soil column sampling was conducted between two standard trees, with column dimensions of 50 cm × 50 cm in length and width and 100 cm in depth. The vertical profile was divided into five layers at 20 cm intervals: 0-20 cm, 20–40 cm, 40–60 cm, 60–80 cm, and 80–100 cm. Each layer was broken up and sieved, and roots were selected and identified based on color characteristics. Root diameter was measured with vernier calipers and classified, with fine roots defined as < 3 mm and coarse roots > 3 mm [**?**]. Root length and surface area of roots < 3 mm were analyzed using a root scanning system. Finally, all roots were oven-dried at 80 °C to constant weight to determine dry weight. Additionally, soil samples were collected from the 12 standard plots of the three plantation types using the ring knife method, with one sample taken from each soil layer in each plot to determine soil water content.

Soil from the soil columns was mixed and subsampled, then air-dried and passed through a 0.20 mm sieve for determination of soil physicochemical properties. Soil pH was determined by potentiometry (soil:water ratio 1:2.5), electrical conductivity by conductometry (soil:water ratio 1:5), available nitrogen by alkali-hydrolysis diffusion method, available phosphorus by extraction-molybdenum antimony colorimetry, available potassium by flame photometry, and organic matter content by potassium dichromate oxidation-external heating method [**?**].

2.2 Data Processing

Fine root biomass density, root length density, surface area density, and volume density were calculated using the following formulas:

Where: DW is fine root biomass density ($g \cdot m^{3}$); Wd is fine root dry weight (g); DL is root length density ($m \cdot m^{3}$); Lr is root length (m); DA is fine root surface area density ($m^2 \cdot m^{3}$); Ar is fine root surface area (m^2); DV is fine root volume density ($cm^3 \cdot m^{3}$); Vr is fine root volume (cm^3); Vs is soil volume (m^3), $Vs = l^2 \times h$, where l is the layer width (0.5 m) and h is the layer thickness (0.2 m).

Excel 2013 was used to calculate means, totals, densities, and percentages of root biomass, surface area, length, and volume. SPSS 22.0 software was used for correlation analysis among treatments, with ANOVA performed for inter-treatment data. Duncan' s method was used to test for significant differences, with significance level at $P < 0.05$ and highly significant level at $P < 0.01$.

3.1 Fine Root Biomass Distribution Characteristics

Root biomass density is an important indicator of underground plant growth and the most direct expression of root system development [?]. As shown in Figure 1 [Figure 1: see original paper]A, fine roots of the velvet ash and black locust mixed forest were mainly distributed in the 0-60 cm soil layer, accounting for 95.77% of the total; the 0-20 cm layer contained the most fine roots (54.44% of the total), while the 80-100 cm layer had the smallest biomass (0.907% of the total). Fine roots of the tree-of-heaven and black locust mixed forest were mainly distributed in the 0-40 cm layer (85.37% of the total), with the 0-20 cm layer containing the most (46.85% of the total). Fine root biomass of the black locust pure stand was mainly distributed in the 0-40 cm layer (66.38% of the total), with the 0-20 cm layer containing 33.95% of the total, while the 40-60 cm layer had the lowest biomass density (5.4% of the total).

Fine root biomass density in all three stand types was highest in the 0-20 cm layer, with the velvet ash and black locust mixed forest showing significantly higher fine root biomass in this layer than the tree-of-heaven and black locust mixed forest and black locust pure stand ($P < 0.05$). The velvet ash and black locust mixed forest also had significantly higher fine root biomass density in the 40-60 cm layer than the other two stands ($P < 0.05$). The minimum fine root biomass layer was 80-100 cm for the velvet ash and black locust mixed forest, and 40-60 cm for both the tree-of-heaven and black locust mixed forest and the black locust pure stand. Root biomass density of the black locust pure stand was relatively uniformly distributed throughout the 0-100 cm soil profile.

As shown in Figure 1B, velvet ash had the highest fine root biomass, significantly higher than all other tree species except black locust in pure stands ($P < 0.05$). Black locust fine root biomass in both mixed stands and tree-of-heaven fine root biomass in the mixed stand were significantly lower than in the black locust pure stand ($P < 0.05$). Velvet ash fine root biomass in the mixed stand was mainly distributed in the 0-60 cm layer (96.72% of the total), with 89.85% in the 0-20 cm layer, showing strong surface aggregation. Black locust fine root biomass in the velvet ash mixed stand was mainly distributed in the 0-20 cm and 40-60 cm layers (86.16% of the total). Fine root biomass of both tree-of-heaven and black locust in the mixed stand was mainly distributed in the 0-40 cm layer. Black locust fine root biomass in the pure stand was mainly distributed in the 0-40 cm and 60-80 cm layers. Black locust fine root biomass in the tree-of-heaven mixed stand was highest in the 20-40 cm layer, while tree-of-heaven and other species in the three plantation types had maximum fine root biomass in the 0-20 cm layer.

3.2 Fine Root Surface Area, Length, and Volume Density Distribution Characteristics

Fine root surface area density of the velvet ash and black locust mixed forest was 3.03 and 1.67 times that of the tree-of-heaven and black locust mixed forest and black locust pure stand, respectively, with significant differences ($P < 0.05$). In the 0-20 cm layer, fine root surface area density of the velvet ash and black locust mixed forest was 16.85 $m^2 \cdot m^{3}$, significantly higher than the other two stands and also significantly higher than other layers within the same stand. Fine root surface area in the 0–20 cm layer of both mixed stands was significantly higher than in other layers ($P < 0.05$), while the black locust pure stand had maximum fine root surface area in the 20–40 cm layer (Figure 2 [Figure 2: see original paper]A).

Average fine root length densities of the velvet ash and black locust mixed forest, tree-of-heaven and black locust mixed forest, and black locust pure stand were 1,533.05 $m \cdot m^{3}$, 1,204.22 $m \cdot m^{3}$, and 1,186.59 $m \cdot m^{3}$, respectively. Fine root length density of the velvet ash and black locust mixed forest was significantly higher than the other two stands ($P < 0.05$), with no significant difference between the tree-of-heaven mixed stand and pure stand ($P > 0.05$). In the 0–20 cm layer, fine root length of the velvet ash mixed stand accounted for 67.48% of the total, with a length density of 5,172.48 $m \cdot m^{3}$, significantly higher than the other stands ($P < 0.05$) and 3.18 and 3.33 times that of the tree-of-heaven mixed stand and pure stand, respectively. The tree-of-heaven mixed stand and pure stand had maximum fine root length densities in the 40–60 cm and 20–40 cm layers, respectively, at 1,877.84 and 2,455.14 $m \cdot m^{3}$ (Figure 2B).

Average fine root volume densities of the velvet ash and black locust mixed forest, tree-of-heaven and black locust mixed forest, and black locust pure stand were 725.41 $cm^3 \cdot m^{3}$, 532.06 $cm^3 \cdot m^{3}$, and 437.37 $cm^3 \cdot m^{3}$, respectively. Fine root volume density of the velvet ash mixed stand was significantly higher than the other two stands ($P < 0.05$), while the tree-of-heaven mixed stand was significantly higher than the pure stand ($P > 0.05$). In the 0–20 cm layer, fine root volume densities of the two mixed stands were 2,481.72 $cm^3 \cdot m^{3}$ and 2,112.44 $cm^3 \cdot m^{3}$, respectively, significantly higher than the pure stand ($P < 0.05$), with no significant difference between the two mixed stands ($P > 0.05$). The black locust pure stand had maximum fine root volume density in the 20–40 cm layer at 880.17 $cm^3 \cdot m^{3}$ (Figure 2C).

3.3 Fine Root Branching Structure

Total fine root tip numbers of the velvet ash and black locust mixed forest, tree-of-heaven and black locust mixed forest, and black locust pure stand were 91,276, 47,831, and 38,969, respectively, with total fork numbers of 48,460, 13,117, and 7,882, respectively (Figure 3 [Figure 3: see original paper]). Fine root tip and fork numbers of both mixed stands were significantly higher than the pure stand ($P < 0.05$), with tip numbers being 2.34 and 1.23 times that of the pure stand,

and fork numbers being 6.15 and 1.66 times that of the pure stand. Fine root tips of the mixed stands were mainly distributed in the 0–40 cm layer, while those of the pure stand were mainly in the 0–40 cm and 60–80 cm layers. Fine root forks of the velvet ash mixed stand were mainly distributed in the 0–60 cm layer, those of the tree-of-heaven mixed stand in the 0–40 cm layer, and those of the pure stand in the 0–40 cm and 60–80 cm layers.

3.4 Correlations Between Fine Root Biomass Density and Soil Properties

As shown in Table 2 , soil electrical conductivity, pH, alkali-hydrolyzable nitrogen, available phosphorus, available potassium, and organic matter content in all three plantation types showed increasing trends with soil depth, while soil water content showed the opposite trend. The three plantation types showed certain differences in soil water content, alkali-hydrolyzable nitrogen, available phosphorus, available potassium, and organic matter content in the 0–40 cm layer, but no significant differences in the 60–100 cm layer. The two black locust mixed stands had lower electrical conductivity, alkali-hydrolyzable nitrogen, available phosphorus, and organic matter content in the 0–20 cm layer than the pure stand. Specifically, the velvet ash mixed stand had electrical conductivity, available phosphorus, and organic matter contents of 93.95%, 82.61%, and 64.29% of those in the pure stand, respectively, with significant differences. The tree-of-heaven mixed stand had electrical conductivity and available phosphorus contents of 87.38% and 57.45% of those in the pure stand, respectively, with significant differences. The pure stand had significantly lower soil water content in the 0–20 cm layer than both mixed stands.

Fine root biomass density of the three plantation types showed certain correlations with soil physicochemical properties. In the velvet ash mixed stand, fine root biomass density was significantly negatively correlated with soil water content, significantly positively correlated with available phosphorus and available potassium contents, and highly significantly positively correlated with alkali-hydrolyzable nitrogen and organic matter contents. In the tree-of-heaven mixed stand, fine root biomass density was significantly negatively correlated with soil electrical conductivity and water content, and significantly positively correlated with soil pH, water content, available phosphorus, available potassium, and organic matter contents. In the black locust pure stand, fine root biomass density was significantly negatively correlated with soil electrical conductivity and significantly positively correlated with available phosphorus and available potassium contents.

4 Discussion and Conclusion

This study demonstrated that fine roots of black locust plantations in the saline soils of the Yellow River Delta exhibited a certain degree of surface aggregation, gradually decreasing with soil depth. For example, fine root biomass of black locust in pure stands was mainly distributed in the 0–40 cm layer, differing from results reported by Wang et al. [?] and Wan et al. [?] (20–40 cm). The main reason for this discrepancy may be differences in site conditions leading to substantial variations in the vertical distribution of black locust roots. More than 60% of black locust fine root biomass was concentrated in the surface and near-surface soil layers, likely related to higher salt content in deeper soils of the Yellow River Delta. The extensive distribution of tree roots in surface soil layers can mitigate the harmful effects of soil salinization on plant roots and improve plant survival and growth rates [?]. Surface aggregation of fine roots of velvet ash and tree-of-heaven in mixed stands was greater than in black locust pure stands, possibly because black locust is a deep-rooted species. Under coastal saline-alkali conditions in the Yellow River Delta, deep-rooted species are more susceptible to salt-alkali stress than shallow-rooted species, reducing their productivity, which explains why black locust growth was inferior to that of velvet ash and tree-of-heaven [?].

This study indicates that mixing in Yellow River Delta plantations enhanced surface aggregation of black locust roots, consistent with previous findings [?]. In the 0–60 cm layer, the proportion of black locust fine root biomass increased from 71.69% in pure stands to 87.91% and 90.34% in mixed stands. This likely relates to black locust' s strategy for acquiring soil resources [?]. In pure stands, black locust fine roots adopted a resource-acquisition strategy with relatively uniform biomass distribution, increasing utilization of deeper soil to minimize competition for soil nutrients and water and achieve optimal resource allocation. In mixed stands, to compete with other species (tree-of-heaven, ash) for soil nutrients and water, black locust adopted a rapid resource-acquisition strategy, with extensive distribution of fine roots in shallow soil layers to fully absorb nutrients in surface soil [?].

Soil water content and fine root biomass were lowest in black locust pure stands of the Yellow River Delta. Previous studies have found that fine root distribution in soil is significantly correlated with and primarily controlled by soil water content, which is basically consistent with this study [?]. This may be because black locust is a water-consuming species. The sampling time in this study was November, when precipitation is low and ground evaporation is strong in the Yellow River Delta, resulting in low soil water content. Lower soil water content is not conducive to root growth (especially fine roots), or may cause slow growth or even death of fine roots. Therefore, fine root biomass was lower in pure stands and higher in mixed stands [?].

The purpose of tree mixing is to promote tree growth to achieve better ecological and economic benefits. Zhang et al. [?] reported that fine root biomass and total root biomass in mixed stands of Manchurian ash (*Fraxinus mandshurica*) and Dahurian larch (*Larix gmelinii*) were higher than in pure stands, particularly significantly increasing the proportion of fine roots to total roots, which benefits plant growth. This study differs, showing that under the same site conditions, planting density, and spacing in the Yellow River Delta, black locust mixed with velvet ash and tree-of-heaven resulted in significantly lower black locust fine root biomass in mixed stands than in pure stands, indicating that mixing was not conducive to black locust root growth. This may be related to differences in tree species and site conditions, and also suggests that only appropriate mixing can promote root growth and improve stand productivity [**?**].

Further research should integrate analysis of fine root morphological characteristics under different silvicultural modes with soil physicochemical properties in different layers, and investigate the mechanisms underlying fine root biomass distribution patterns and differences from perspectives of plant adaptation strategies to salt-alkali stress and interspecific/intraspecific competition. Additionally, seasonal dynamic monitoring of fine roots in the Yellow River Delta is needed to elucidate seasonal dynamics and functions of root systems in different stands, thereby better guiding management during plantation establishment and community management and the selection of tree species and planting patterns.

The study of fine root distribution in mixed black locust plantations in saline soils of the Yellow River Delta indicates that tree fine roots were mainly distributed in the 0–40 cm soil layer, with mixed stands, especially the velvet ash and black locust mixed stand, showing higher fine root biomass, surface area, volume, and length than black locust pure stands. Mixed stands had higher total fine root tip and fork numbers than pure stands, with the velvet ash and black locust mixed stand having the highest values. Fine root growth in plantations was affected by soil physicochemical properties, with higher contents of alkali-hydrolyzable nitrogen, available phosphorus, available potassium, and organic matter promoting fine root growth, while higher soil electrical conductivity inhibited fine root growth.

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