Postprint: Analysis of Rhizosphere and Non-rhizosphere Bacterial Characteristics of Young ‘Zhongshanshan 118’ in the Northern Anhui Huaihe Plain

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Abstract

To investigate the effects of the ecological landscape greening tree species ‘Zhongshanshan 118’ on the soil ecosystem of the Huai River Plain in northern Anhui, this study analyzed the differences in rhizosphere and non-rhizosphere soil nutrients, microbial communities, and their influencing factors. The results showed: (1) The rhizosphere of 4-year-old ‘Zhongshanshan 118’ exhibited weak enrichment capacity for macronutrients, with hydrolyzable nitrogen and available phosphorus showing deficits; total nitrogen was comparable to that of the non-rhizosphere, while there was low-level enrichment of total phosphorus, total potassium, and available potassium; the rhizosphere showed strong enrichment capacity for the micronutrients iron, copper, and manganese, with rhizosphere available manganese content being significantly higher than that of the non-rhizosphere; both rhizosphere and non-rhizosphere soils of ‘Zhongshanshan 118’ were alkaline, with rhizosphere pH being slightly lower than non-rhizosphere pH, and rhizosphere electrical conductivity being greater than that of the non-rhizosphere; the rhizosphere showed a certain degree of enrichment of available sodium, available magnesium, and available calcium, as well as some enrichment of organic carbon and organic matter. (2) The ANOSIM inter-group community structure difference significance test yielded an R value of 0.224 (P=0.022), with the R value being significantly greater than 0, indicating significant differences between the rhizosphere and non-rhizosphere bacterial communities of ‘Zhongshanshan 118’. There were 5,283 OTUs shared between the rhizosphere and non-rhizosphere of ‘Zhongshanshan 118’, with 1,307 being rhizosphere-specific and 1,265 being non-rhizosphere-specific. Rhizosphere bacterial diversity was slightly lower than that of the non-rhizosphere. The dominant bacterial phyla in both were Firmicutes, Proteobacteria, Acidobacteria, Bacteroidetes, Chloroflexi, Myxococcota, Actinobacteria, and Crenarchaeota, among which only Acidobacteria showed significant positive correlation with available phosphorus. Among the relatively abundant phyla, the contents of Firmicutes and Bacteroidetes in the rhizosphere (17.94% and 5.21%) were higher than those in the non-rhizosphere (1.84% and 3.90%), and neither showed significant correlation with environmental factors. The abundances of Clostridia, Bacteroidia, and Bacilli in the rhizosphere were 14.05%, 1.32%, and 2.06% higher than those in the non-rhizosphere, respectively. (3) The UPGMA clustering tree showed that the rhizosphere sample R4 of 'Zhongshanshan 118' clustered independently, non-rhizosphere samples NR4 and NR5 clustered together, and NR1, NR2, and NR3 clustered with rhizosphere samples R1, R2, R3, and R5. The species diversity and abundance of R4 were significantly lower than those of other samples and were unevenly distributed. In conclusion, although the rhizosphere effect of young 'Zhongshanshan 118' was weak, it was the main factor responsible for the enrichment of Firmicutes and Bacteroidetes in the rhizosphere; during cultivation, micronutrients should be supplemented abundantly and macronutrients should be supplemented appropriately.

Full Text

Rhizosphere and Non-Rhizosphere Bacterial Characteristics Analysis of Young Taxodium 'Zhongshanshan 118' in the Huaihe Plain of Northern Anhui

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Abstract

To investigate the effects of the ecological landscaping tree species Taxodium 'Zhongshanshan 118' on the soil ecosystem of the Huaihe Plain in northern Anhui Province, this study analyzed differences in soil nutrients, microbial communities, and their influencing factors between rhizosphere and non-rhizosphere soils. The results showed: (1) The 4-year-old T. 'Zhongshanshan 118' exhibited weak enrichment capacity for macronutrients in the rhizosphere, with deficiencies in hydrolyzable nitrogen and available phosphorus. Total nitrogen remained comparable to non-rhizosphere soil, while slight enrichment occurred for total phosphorus, total potassium, and available potassium. The rhizosphere demonstrated strong enrichment ability for micronutrients iron, copper, and manganese, with available manganese content significantly higher than in non-rhizosphere soil. Both rhizosphere and non-rhizosphere soils were alkaline, with rhizosphere pH slightly lower and electrical conductivity higher than non-rhizosphere soils. The rhizosphere also showed moderate enrichment of available sodium, magnesium, calcium, organic carbon, and organic matter. (2) Anosim analysis of community structure differences between groups yielded an R value of 0.224 (P=0.022), significantly greater than 0, indicating significant differences between rhizosphere and non-rhizosphere bacterial communities. The two environments shared 5,283 OTUs, with 1,307 OTUs unique to the rhizosphere and 1,265 unique to the non-rhizosphere. Bacterial diversity was slightly lower in the rhizosphere. Dominant phyla in both environments included Firmicutes, Proteobacteria, Acidobacteriota, Bacteroidetes, Chloroflexi, Myxococcota, Actinobacteria, and Crenarchaeota. Only Acidobacteriota showed significant positive correlation with available phosphorus. Among abundant phyla, Firmicutes and Bacteroidetes were more abundant in the rhizosphere (17.94% and 5.21%) than in non-rhizosphere soil (1.84% and 3.90%), though neither showed significant correlation with environmental factors. The classes Clostridia, Bacteroidia, and Bacilli were 14.05%, 1.32%, and 2.06% more abundant in the rhizosphere, respectively. (3) UPGMA clustering showed rhizosphere sample R4 formed a separate branch, non-rhizosphere samples NR4 and NR5 clustered together, and NR1, NR2, and NR3 clustered with rhizosphere samples R1, R2, R3, and R5. Sample R4 exhibited significantly lower species diversity and abundance with uneven distribution. In conclusion, despite the weak rhizosphere effect of young T. 'Zhongshanshan 118', it was the primary factor enriching Firmicutes and Bacteroidetes in the rhizosphere. Cultivation practices should emphasize micronutrient supplementation with moderate macronutrient application.

Keywords: Taxodium 'Zhongshanshan 118', rhizosphere, non-rhizosphere, bacteria, soil nutrients, high-throughput sequencing

Introduction

The Huaihe Plain of northern Anhui Province lies at the northern edge of the subtropical zone, characterized by flat terrain and extensive cultivated land. Landscaping plants and crops in this region are vulnerable to extreme weather events such as freezing and chilling damage during climatically unstable years, causing substantial economic losses. Consequently, planting tall trees and shelterbelts is essential for maintaining ecosystem stability. Fast-growing trees currently used for urban-rural greening and agroforestry networks in this area are limited primarily to poplar, plane trees, and sweetgum, resulting in unstable and incomplete forest structures. Taxodium 'Zhongshanshan 118' represents a collective term for superior hybrid clones derived from crosses among baldcypress (T. distichum), pondcypress (T. ascendens), and Montezuma cypress (T. mucronatum). These varieties exhibit rapid growth, with annual diameter and height increments reaching 1.5–2 cm and 1.5–2 m, respectively, ultimately developing into straight, tower-shaped trees 30–50 m tall. Their foliage remains dark green during the growing season, turning orange-yellow in autumn. With minimal pest and disease issues, resistance to category-12 typhoons, and lifespans reaching 3,000 years, Taxodium 'Zhongshanshan 118' represents an excellent ecological landscaping species. The rapid growth characteristics of Taxodium are closely related to efficient utilization of soil moisture and nutrients, where plant rhizosphere and non-rhizosphere microorganisms significantly promote decomposition and transformation of soil nutrients.

The rhizosphere is defined as the microzone within 14 mm of plant roots, with areas beyond this range considered non-rhizosphere soil. As a unique interface between plant roots and soil, the rhizosphere hosts the most active exchange of materials and energy among plants, soil, and microorganisms, playing a crucial role in ecosystem nutrient cycling. Soil bacteria constitute a major component of soil ecosystems, accounting for 70–90% of total soil microbial biomass. Plant growth-promoting rhizobacteria (PGPR) can decompose and transform soil nutrients into plant-available forms through metabolic activities, promoting plant growth. Some bacteria secrete enzymes and hormones that enhance plant adaptability to drought, salinity, and pathogen attack, providing protection for healthy plant development. Thus, rhizosphere microorganisms are intimately linked to plant growth and health. Simultaneously, plants can alter rhizosphere nutrient content and physicochemical properties through root activities, subsequently changing rhizosphere microbial community composition and creating differences between rhizosphere and non-rhizosphere microbial communities and diversity.

Taxodium 'Zhongshanshan 118' [(T. distichum × T. mucronatum) × T. mucronatum] is a superior cultivar obtained by backcrossing 'Zhongshanshan 302' with its male parent Montezuma cypress, certified as an improved variety by the National Forestry and Grassland Administration (Approval No.: National R-SC-TD-001-2006). This cultivar offers rapid growth, high ornamental value, and tolerance to salinity and waterlogging, and has been widely applied in urban landscaping, agroforestry networks, and riparian ecosystems. Current research on 'Zhongshanshan 118' has primarily focused on introduction and application, breeding and cultivation, hybrid breeding, heterosis utilization and mechanisms, waterlogging stress tolerance, and wood properties, with few studies examining its interactions with soil ecosystems. This study investigated 4-year-old 'Zhongshanshan 118' rhizosphere and non-rhizosphere soils using 16S rRNA high-throughput sequencing to reveal soil bacterial community structure, species composition, and differences, as well as relationships with soil physicochemical factors, thereby clarifying the effects of 'Zhongshanshan 118' on soil quality and microorganisms and providing fundamental data for exploring relationships among its rapid growth characteristics, soil properties, and microorganisms.

Materials and Methods

1.1 Sample Collection

The sampling site was located at Suzhou Botanical Garden in Yongqiao District, Suzhou City, Anhui Province (116°09 –118°10 E, 33°18 –34°38 N), situated in northern Anhui in the heart of the Huaihe Plain. The 'Zhongshanshan 118' plantation covered 100 m × 200 m with flat terrain, uniform site conditions, and yellow loam soil. The experimental 1-year-old cuttings of 'Zhongshanshan 118' were purchased from Jiangsu Jingjiang and were 4-year-old seedlings at the time of sampling. Plantation management included manual weeding, timely irrigation, and fertilization with urea. In late June 2020, a 50 m × 50 m plot was randomly selected away from garden paths. Using a diagonal method, a central sampling point was established with four additional points at equal distances along the diagonal, resulting in five sampling points for soil collection. At each point, complete root systems were excavated. After removing large soil clumps, loosely adhering soil shaken from roots was collected as non-rhizosphere samples (NR1–NR5), while soil remaining tightly attached to roots was collected with sterile brushes as rhizosphere samples (R1–R5).

1.2.1 Soil Physicochemical Property Determination

Soil pH, electrical conductivity, organic matter, organic carbon, total nitrogen, total phosphorus, total potassium, hydrolyzable nitrogen, available phosphorus, available potassium, available copper, zinc, iron, manganese, available sodium, calcium, and magnesium were measured using glass electrode method, electrode method, dichromate oxidation-external heating method, sulfuric acid-accelerator digestion-Kjeldahl method, NaOH fusion-molybdenum antimony spectrophotometry, atomic absorption spectrophotometry, alkali diffusion method, sodium bicarbonate extraction-molybdenum antimony colorimetry, ammonium acetate extraction-atomic absorption method, DTPA-TEA extraction, ICP-AES, and ammonium acetate extraction-ICP-AES, respectively. Each indicator was measured in triplicate.

1.2.2 Genomic DNA Extraction, PCR Amplification, and Sequence Analysis

Genomic DNA was extracted using the CTAB method, and its purity and concentration were assessed. Bacterial 16S V4 region was amplified using Phusion® High-Fidelity PCR Master Mix with GC Buffer and primers 515F (5 -GTGCCAGCMGCCGCGG-3) and 806R (5 -GGACTACHVGGGTWTCTAAT-3). Target products were recovered, libraries constructed, and sequenced after quality control. Raw Illumina NovaSeq paired-end reads were assembled and quality-filtered using FLASH version 1.2.7 and Qiime version 1.9.1 to obtain Clean Tags. Chimeras were filtered using vsearch version 1.3.0 to obtain Effective Tags for subsequent analysis. Effective Tags with 97% similarity were clustered using Uparse version 7.0.1001, and OTU sequences were taxonomically annotated against the SILVA138 SSUrRNA database using the Mothur method.

1.2.3 Data Analysis

Independent samples t-tests were performed on rhizosphere and non-rhizosphere soil physicochemical factors using R software. Data are presented as mean ±

standard error (SEM), with $P < 0.05$ indicating statistical significance. Nutrient enrichment was assessed using the enrichment ratio E = [(rhizosphere content - non-rhizosphere content)/non-rhizosphere content] × 100%. Diversity indices were calculated using Qiime version 1.9.1, and differences between rhizosphere and non-rhizosphere were tested using rank-sum tests in R. Anosim analysis was performed using the vegan package in R to test significant differences between groups, where $R > 0$ indicates greater between-group than within-group differences and P values indicate significance. Unifrac distances were calculated using Qiime to construct UPGMA clustering trees. Spearman correlation coefficients between environmental factors and microbial abundance were analyzed using the psych package in R and visualized with the pheatmap package.

Results

2.1 Analysis of Rhizosphere and Non-Rhizosphere Soil Physicochemical Properties

As shown in Table 1 , the 'Zhongshanshan 118' plantation soil in Suzhou Botanical Garden was alkaline, with rhizosphere pH slightly lower than non-rhizosphere and electrical conductivity higher in rhizosphere soils. Independent t-tests revealed that only available manganese was significantly higher in rhizosphere than non-rhizosphere ($P < 0.05$), while other physicochemical factors showed no significant differences ($P > 0.05$). Mean values and enrichment ratios indicated that 'Zhongshanshan 118' rhizosphere enriched available iron (44.21%), available copper (27.38%), available manganese (26.59%), available sodium (23.92%), organic carbon (8.22%), organic matter (8.15%), available magnesium (7.73%), total phosphorus (6.52%), available calcium (4.20%), available potassium (3.73%), and total potassium (3.09%), with particularly high enrichment of available iron, manganese, and copper. Total nitrogen remained comparable between rhizosphere and non-rhizosphere, while hydrolyzable nitrogen, available zinc, and available phosphorus showed deficits of 2.14%, 14.95%, and 34.43%, respectively.

2.2 Analysis of Rhizosphere and Non-Rhizosphere Soil Microbial Diversity

Sequencing depth indices ranged from 0.980 to 0.997, Q20 from 98.79% to 99.44%, GC content from 52.77% to 57.43%, and Effective Tags percentage from 70.44% to 81.23%, indicating high sequencing depth and quality. Overall analysis showed that rhizosphere soil microbial species numbers, Chao1, ACE, Simpson, Shannon, and PD whole tree indices averaged 240, 3,828.033, 3,733.681, 0.984, 9.157, and 186.018, respectively—all slightly lower than non-rhizosphere values of 3,743, 4,096.895, 4,151.882, 0.997, 10.019, and 213.703. Diversity index differences between rhizosphere and non-rhizosphere were not significant (Table 2). For individual samples, rhizosphere sample R4 exhibited markedly lower species diversity and abundance with highly uneven distribution compared to other samples (Table 2, Figure 1 [Figure 1: see original paper]).

2.3 Soil Microbial Community Structure Analysis

Anosim analysis yielded an R value of 0.224 ($P = 0.022$), significantly greater than 0, indicating significant differences between 'Zhongshanshan 118' rhizosphere and non-rhizosphere microbial communities. The two environments shared 5,283 OTUs, with 1,307 OTUs (19.97% of total rhizosphere OTUs) unique to rhizosphere and 1,265 OTUs (19.45% of total non-rhizosphere OTUs) unique to non-rhizosphere. OTU annotation revealed microorganisms from both bacterial and archaeal domains, belonging to 89 phyla, 182 classes, 371 orders, 489 families, 735 genera, and 313 identifiable species. UPGMA clustering showed rhizosphere sample R4 formed a separate branch, non-rhizosphere samples NR4 and NR5 clustered together, and NR1, NR2, and NR3 clustered with rhizosphere samples R1, R2, R3, and R5 (Figure 1 [Figure 1: see original paper]).

2.4 Soil Microbial Community Composition

As shown in Figure 2 [Figure 2: see original paper]A, rhizosphere and non-rhizosphere soils shared consistent dominant bacterial groups but in different proportions. Dominant phyla included Firmicutes, Proteobacteria, Acidobacteriota, Bacteroidetes, Chloroflexi, Myxococcota, Actinobacteria, and Crenarchaeota, accounting for 17.94%, 25.61%, 7.29%, 5.21%, 3.03%, 3.14%, 3.63%, and 1.19% in rhizosphere, and 1.84%, 30.98%, 9.81%, 3.90%, 3.12%, 3.71%, 5.55%, and 2.08% in non-rhizosphere, respectively. Firmicutes and Bacteroidetes were 16.19% and 1.31% more abundant in rhizosphere than non-rhizosphere. In sample R4, Firmicutes dominated at 76.77% of total bacterial phyla (Figure 1 [Figure 1: see original paper]).

Dominant classes included Clostridia, Gammaproteobacteria, Alphaproteobacteria, Bacteroidia, Bacilli, Chlamydiae, Vicinamibacteria, and Holophagae, representing 15.08%, 17.14%, 8.47%, 5.19%, 2.82%, 4.46%, 2.44%, and 0.83% in rhizosphere, and 1.03%, 19.88%, 11.09%, 3.87%, 0.76%, 6.79%, 2.98%, and 2.18% in non-rhizosphere, respectively (Figure 2 [Figure 2: see original paper]B). Clostridia, Bacteroidia, and Bacilli were 14.05%, 1.32%, and 2.06% more abundant in rhizosphere. In sample R4, Clostridia accounted for 66.73% of total bacterial classes.

2.5 Correlation Analysis Between Microorganisms and Environmental Factors

Spearman analysis revealed that organic carbon, organic matter, and total nitrogen were extremely significantly positively correlated with Myxococcota. Available phosphorus was significantly positively correlated with Acidobacteriota and Entotheonellaeota. Chlamydiae was significantly negatively correlated with electrical conductivity, available potassium, and available magnesium. Available zinc was significantly negatively correlated with SAR324 clade (Figure 3 [Figure 3: see original paper]A). Alphaproteobacteria was significantly positively correlated with pH and dry/wet ratio but significantly negatively correlated with electrical conductivity, total phosphorus, total potassium, hydrolyzable nitrogen, available potassium, available manganese, available copper, available sodium, available calcium, and available magnesium. Total nitrogen was significantly negatively correlated with Bacilli. Available zinc was significantly negatively correlated with Bacteroidia, Alphaproteobacteria, and Gammaproteobacteria. Total nitrogen was significantly positively correlated with Polyangia and Myxococcia. Total potassium was significantly negatively correlated with Vicinamibacteria. Hydrolyzable nitrogen was significantly negatively correlated with Anaerolineae. Available phosphorus was significantly positively correlated with Thermoleophilia. Total phosphorus was significantly negatively correlated with Longimicrobia. Total potassium and available magnesium were significantly negatively correlated with KD4.96 (Figure 3 [Figure 3: see original paper]B).

Discussion

3.1 Analysis of Rhizosphere and Non-Rhizosphere Soil Physicochemical Property Differences

Soil physicochemical properties reflect soil quality, which is crucial for plant growth and development. Long-term cultivation of specific plants reciprocally affects soil quality and ecosystems. Young ‘Zhongshanshan 118’ showed deficiencies in hydrolyzable nitrogen and available phosphorus in the rhizosphere, comparable total nitrogen to non-rhizosphere soil, and slight enrichment of total phosphorus, total potassium, and available potassium, indicating weak enrichment capacity for macronutrients during the vigorous growth stage. Available manganese was significantly higher in rhizosphere soil. As a directly plant-available form, manganese is closely associated with photosynthesis, enzyme metabolism, and carbohydrate accumulation. Additionally, ‘Zhongshanshan 118’ showed high-level enrichment of available iron and copper. Iron plays important roles in photosynthesis and nutrient accumulation, while copper stabilizes chlorophyll function and participates in plant respiratory metabolism. These findings indicate that young ‘Zhongshanshan 118’ exhibits strong positive rhizosphere effects on micronutrients iron, manganese, and copper to meet growth demands under conventional cultivation. Enrichment of available sodium, magnesium, and calcium was relatively low, with rhizosphere electrical conductivity slightly higher than non-rhizosphere. Studies have shown that ‘Zhongshanshan 118’ can accumulate small amounts of Na^+ while selectively absorbing and transporting Ca^{2+}, Mg^{2+}, and K^+ to maintain balance with Na^+, thereby enhancing salt tolerance. Organic carbon and organic matter indicate soil fertility, and their enrichment in the rhizosphere suggests higher potential fertility than non-rhizosphere soil.

Only available manganese differed significantly between young ‘Zhongshanshan 118’ rhizosphere and non-rhizosphere soils, indicating limited influence of young root systems on soil physicochemical properties and weak rhizosphere effects.

Tong et al. (2019) analyzed rhizosphere and non-rhizosphere soil nutrient characteristics of different-aged *Choerospondias axillaris*, finding that 3-year-old trees only enriched total nitrogen with significant differences, while 15- and 30-year-old trees enriched total nitrogen, total phosphorus, hydrolyzable nitrogen, available phosphorus, total potassium, and available potassium, demonstrating weak rhizosphere effects in young trees. Song et al. (2014) found minimal differences in soil physicochemical factors between rhizosphere and non-rhizosphere of young *Amorpha fruticosa* on the Tibetan Plateau, with soil fertility improvement becoming apparent with increasing planting years. During the peak growing season, young 'Zhongshanshan 118' showed low enrichment of macronutrients nitrogen, phosphorus, and potassium but high enrichment of micronutrients iron, manganese, and copper, along with moderate enrichment of Ca^{2+}, Mg^{2+}, and Na^{+}, providing valuable guidance for fertilization practices.

3.2 Analysis of Rhizosphere and Non-Rhizosphere Soil Bacterial Differences

Rhizosphere and non-rhizosphere bacterial communities of 'Zhongshanshan 118' differed significantly, with slightly lower diversity and 1,307 unique OTUs in the rhizosphere. This trend aligns with studies on moso bamboo and other plants, suggesting that plant roots actively filter and inhibit certain microorganisms, resulting in lower rhizosphere microbial diversity. Other studies indicate that environmental factors such as drought, low temperature, high temperature, and high humidity increase root exudates in plants like black locust, *Caragana jubata*, and *Rhododendron moulmainense*, enhancing rhizosphere bacterial selectivity and ultimately reducing rhizosphere microbial diversity. In June 2020, Suzhou experienced average rainfall of 45.6 mm, 12 cloudy days, 10 rainy days, 4 sunny days, and 4 overcast days, with average temperatures of 30.2°C (day) and 20.5°C (night). High temperature and humidity promoted soil bacterial proliferation, while root exudates selectively inhibited some microbial growth, creating negative bacterial effects in the rhizosphere.

Among abundant bacterial groups, Firmicutes and Bacteroidetes were more abundant in 'Zhongshanshan 118' rhizosphere without significant correlation with environmental factors, suggesting plant species was the main enrichment factor. Firmicutes primarily includes Bacilli and Clostridia, which can degrade insoluble compounds, remediate crude oil, fix atmospheric nitrogen, control plant diseases and pests, and treat industrial wastewater. Clostridia are generally obligate anaerobes distributed in animal intestines, high-temperature compost, and biogas systems, possessing strong degradation capabilities and metabolic activity for industrial enzyme production. Bacteroidetes contain more plant growth-promoting bacteria and dominate in solid-state anaerobic digestion of corn straw. The enriched Firmicutes and Bacteroidetes in 'Zhongshanshan 118' rhizosphere may promote rapid nutrient accumulation and conversion into plant-available forms. Spearman analysis showed that Acidobacteriota, abundant in both rhizosphere and non-rhizosphere, was only significantly positively correlated with available phosphorus, which was low in rhizosphere, consistent with its lower abundance there. In Danish wastewater treatment plants, Acidobacteriota potentially participates in nitrogen and phosphorus removal and iron reduction. Organic carbon, organic matter, and total nitrogen were extremely significantly positively correlated with Myxococcota, warranting further research on environmental factor regulation of microbial abundance for cultivation applications.

As a fast-growing species, ‘Zhongshanshan 118’ typically enters its rapid growth phase at 5–6 years. This study of 4-year-old plantation rhizosphere and non-rhizosphere soils provides baseline data on bacterial structure, types, distribution, and influencing factors for young trees. Future research should examine microbial characteristics across different stand ages to support sustainable plantation management and investigate rhizosphere and non-rhizosphere microbial features under waterlogged versus normal conditions to comprehensively explore waterlogging tolerance mechanisms.

References

BHATTACHARYYA P, JHA D, 2012. Plant growth-promoting rhizobacteria (pgpr): emergence in agriculture[J]. World J Microb Biot, 28(4): 1327-1350.

CAO HQ, HE MC, SHU YS, et al., 2019. Effects of canine-derived compound probiotics on cecal microflora in splenic qi asthenia canines[J]. Chin J Anim Nutr, 31(8): 3810-3820.

CHEN ME, ZHAO C, HAN SN, et al., 2021. High-throughput sequencing reveals the microbial community structure in Taorong-type Daqu[J]. Food Sci, 42(8): 106-113.

CHENG YL, YANG Y, WANG ZY, et al., 2015. Development and characterization of EST-SSR Markers in Taxodium ‘zhongshansa’[J]. Plant Mol Biol Rep, 33(6): 1804-1814.

DING XJ, JING RY, HUANG YL, et al., 2017. Bacterial structure and diversity of rhizosphere and bulk soil of Robinia pseudoacacia forests in Yellow River Delta[J]. Acta Pedol Sin, 54(5): 1293-1302.

DONG H, LV YC, REN LW, et al., 2021. Microbial community structure of activated sludge in neomycin sulfate waste water[J]. Chem Bioeng, 38(8): 25-31.

Edgar RC, 2013. UPARSE: highly accurate OTU sequences from microbial amplicon reads[J]. Nat Methods, 10(102): 996-998.

GE E, XU SH, XU Y, 2019. Review on influencing factors of rhizosphere microbiome assemblage[J]. Acta Agric Zhejiangensis, 31(12): 2120-2130.

GE YL, SUN T, 2020. Soil microbial community structure and diversity of potato in rhizosphere and non-rhizosphere soil[J]. Ecol Environ, 29(1): 141-148.

GUO H, TANG WP. 2020. Enzyme Activity and microbial community diversity in rhizosphere and non-rhizosphere soil of Larix principis-rupprechtii[J]. Ecol Environ, 29(11): 2163-2170.

KRISTENSEN JM, SINGLETON C, CLEGG L A, et al., 2021. High diversity and functional potential of undescribed "Acidobacteriota" in Danish wastewater treatment plants[J]. Front Microbiol, (12): 643950.

LI X, JIAO Y, DAI G, et al., 2016. Soil bacterial community diversity under different degrees of saline alkaline in the Hetao Area of Inner Mongolia[J]. China Environ Sci, 36(1): 249-260.

LI Y, HE XM, YANG XD, et al., 2018. The microbial community diversity of the rhizosphere and bulk soils of Lycium ruthenicum in different habitats[J]. Acta Ecol Sin, 38(17): 5983-5995.

Li Y, Zeng C, Xiao FP, et al., 2019. Bacterial structure and diversity of rhizosphere and bulk soil of Phyllostachys edulis forest[J]. South For Sci, 47(5): 20-23.

LI YY, XU TT, AI Z, et al., 2023. Diversity and predictive functional of Caragana jubata bacterial community in rhizosphere and non-rhizosphere soil at different elevations[J]. Environ Sci, 1-12.

LIU ZX, ZHU TH, ZHANG J, 2005. Research advances in root exudates and rhizosphere microorganisms of forest trees[J]. World For Res, 18(6): 25-31.

LV R, CHANG F, ZHANG XC, et al., 2022. Responses of soil bacterial and fungal community structure and diversity to microbial bran organic manure in the Loess Plateau[J]. Environ Ecol, 4(2): 40-49.

LV YH, ZHAO Y, ZHANG YP, 2020. Research on the distribution characteristics of soil bacteria communities under the influence of two main sand-fixing plants in Minqin desert area of Gansu province[J]. Ecol Environ, 29(4): 717-724.

MA HY, LIN SM, XU YC, et al., 2008. Salt (NaCl) stress on growth, ion uptake and transport of (Taxodium distichum×T. mucronatum) ×T. mucronatum 'Zhongshansha' breeding clones[J]. J Zhejiang For Coll, 25(3): 319-323.

MA L, YANG HM, ZHONG H, et al., 2011. Introduction research progress of Ascendens Mucronatum and its application in Kunming[J]. Forest Inv Plan, 36(1): 19-25.

MA Y, LI LZ, ZHANG DG, et al., 2019. Distribution characteristics of nutrients and microbial biomass in rhizosphere and non-rhizosphere soils of dominant plants in degraded alpinr meadow[J]. Acta agrestia sin, 27(4): 797-804.

PENG JG, GONG JY, FANG YH, 2022. Diversity of soil microbial communities in rhizosphere and non-rhizosphere of Rhododendron moulmainense[J]. Sci silvae sin, 58(2): 89-99.

PENG K, DONG Z, DI YM, et al., 2021. Contrasting analysis of microbial community composition in the water and sediments of the north canal based on 16s rrna high-throughput sequencing[J]. Environ Sci, 42(11): 5424-5432.

QIU J, HOU YL, XU LL, et al., 2019. High throughput sequencing analysis of rhizosphere soil bacteria diversity in different mulberry varieties[J]. J South Agric, 50(3): 585-592.

SONG X, ZHANG LJ, Dai WA, et al., 2014. Effects of Amorpha fruticosa plangting on soil nutrient characteristics at rhizosphere and non-rhizosphere in Tibetan Plateau[J]. Pratac Sci, 31(7): 1226-1232.

SONG ZQ, WANG L, LIU XH, et al., 2015. Diversities of firmicutes in four hot springs in yunnan and tibet[J]. Biotechnol, 25(5): 481-486,436.

TONG LH, GAO J, JIN YS, 2019. Dynamic changes of microorganisms in the rhizosphere of strawberry during different growth stages[J]. J Beijing Agric Coll, 34(4): 10-15.

TONG Q, CHEN MT, LONG JQ, et al., 2019. Research on soil nutrient characteristics at rhizosphere and nonrhizosphere for different age groups of Choerospondias axillaris[J]. J Cent South Univ For Technol, 39(12): 108-113.

WANG ZY, CHENG YL, YIN YL, et al., 2016. Genetic linkage map construction and QTL mapping of seedling height, basal diameter and crown width of Taxodium 'Zhongshanshan 302'×T. mucronatum[J]. Springerplus, 5(1): 936.

WANG ZY, XU JH, LI HG, et al., 2015. Rooting capabilities for Taxodium 'Zhongshanshan' 302, 118, and 405[J]. J Zhejiang A & F Univ, 32(4): 648-654.

WOLF M, 2004. Phylogeny of Firmicutes with special reference to mycoplasma (Mollicutes) as inferred from phosphoglycerate kinase amino acid sequence data[J]. Int J Syst Evol Microbiol, 54(3): 871-875.

WU WX, HUANG XQ, ZHANG L, et al., 2020. Crucifer clubroot disease changes the microbial community structure of rhizospheresoil[J]. Acta Ecol Sin, 40(5): 1532-1541.

XU LX, HE YJ, 2019. Comparison of Bacterial diversity between rhizosphere and non-rhizosphere soil of Maize based on 16S rDNA high-throughput sequencing[J]. J Shanxi Agric Sci, 47(7): 1212-1216.

XUE HY, GUO WW, QU XL, et al., 2021. Structure and diversity of soil nematode community in soil under Tsuga dumosa forest in galongla mountain[J]. Acta Pedol Sin, 2021, 58(1): 246-258.

YANG Y, LIU BR, 2015. Distribution of soil nutrient and microbial biomass in rhizosphere versus non-rhizosphere area of different plant species in desertified steppe[J]. Acta Ecol Sin, 35(22): 7562-7570.

YIN YL, YU CG, HUA JF, 2019. Breeding utilization and related research progress of Taxodium ‘Zhongshanshan’[J]. J Plant Resour Environ, 28(4): 99-106.

YIN YL, YU CG, HUA JF, et al., 2014. A trial on the silviculture of Taxodium hybrid ‘Zhonshanshan118’ planted in the hydro-fluctuation belt of the Three Gorges Reservoir within the Wanzhou district area of Chongqing City[J]. J For Eng, 28(2): 110-114.

YU CG, LI Y, XIE YF, et al., 2016. Effects of NaCl stress on growth and absorption, transportation and distribution of ions in zhongshanshan seedlings[J]. Plant Physiol J, 52(9): 1379-1388.

ZHANG CM, LI YF, DONG BC, et al., 2021. Effect of alkali treatment on microbial community structure in solid-state anaerobic digestion of corn straw[J]. Food Ferment Ind, 48(4): 1-8.

ZHANG JP, HUANG T, HUANG XQ, et al., 2022. Experimental basis of soil science[M]. Beijing: Science Press: 17-128.

Note: Figure translations are in progress. See original paper for figures.