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ARTICLE

Rhizosphere Microbial Community Responses of *Panax notoginseng* to Contrasting Cultivation Modes

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ABSTRACT: Different cultivation systems of *Panax notoginseng*, including wild growth, understory cultivation, and field cultivation, may create distinct rhizosphere environments. Rhizosphere soil properties and microbial communities are closely linked to the sustainability of *P. notoginseng* cultivation, yet comparative evidence across these cultivation systems remains limited. In this study, rhizosphere soils from two-year-old *P. notoginseng* grown under semi-wild, understory, and conventional field cultivation modes in Yunnan Province, China, were analyzed for soil physicochemical properties and microbial community composition using amplicon sequencing. A total of 6842 bacterial ASVs and 1014 fungal ASVs were obtained after quality control and denoising. Soil properties differed among cultivation modes, with semi-wild cultivation generally associated with higher SOC (soil organic carbon), TN (total nitrogen), and alkali-hydrolyzable nitrogen (AN), whereas the pH of it was lower than the other cultivation modes. Beta diversity analyses based on PCoA analysis and PermANOVA analysis revealed significant differences in both bacterial and fungal community composition, with fungal communities showing larger separation than bacterial communities. Semi-wild cultivation was associated with higher relative abundances of Actinobacteria and Agaricomycetes, whereas field cultivation showed a greater proportion of Proteobacteria and Sordariomycetes. Correlation analysis indicated that soil organic carbon, pH, and inorganic nitrogen were the edaphic factors most consistently associated with microbial community differentiation. These results suggest that cultivation mode was closely associated with rhizosphere soil conditions and microbial community composition in *P. notoginseng*, and that semi-wild cultivation may provide a more favorable rhizosphere environment for ecological stability.

KEYWORDS: *Panax notoginseng*; cultivation mode; rhizosphere soil; microbial community; sustainable cultivation

1 Introduction

Panax notoginseng (Burk.) F. H. Chen is a perennial medicinal herb in the family Araliaceae and one of the most important medicinal plants cultivated in southwestern China [1,2]. Its roots and rhizomes contain a range of bioactive compounds, including saponins, flavonoids, and polysaccharides, and are widely used in pharmaceutical and health-related products [3,4]. With increasing market demand, the sustainable production of *P. notoginseng* has become an important agricultural and ecological issue. Conventional production of *P. notoginseng* mainly relies on intensive field cultivation. However, this species is highly sensitive to soil conditions, and prolonged cultivation under field management often leads to soil degradation, nutrient imbalance, acidification, and increasing incidence of soil-borne diseases [5,6]. These changes are

commonly associated with continuous-cropping obstacles and can substantially reduce plant performance and medicinal quality [7,8]. In recent years, growing attention has been paid to the role of rhizosphere microecology in these problems, particularly the interactions among soil physicochemical properties, microbial community structure, and plant health [9].

The rhizosphere is a highly dynamic interface where root activity, soil resources, and microbial processes interact. Soil physicochemical properties regulate nutrient availability, pH conditions, and microbial habitat suitability, whereas rhizosphere microorganisms contribute to organic matter turnover, nutrient cycling, and pathogen suppression [10–12]. As a result, the ecological status of the rhizosphere depends on tight coupling between soil conditions and microbial community assembly. Management practices that alter light conditions, organic matter input, nutrient supply, and disturbance intensity are therefore expected to influence rhizosphere microecology.

Rhizosphere microbial communities serve as pivotal functional regulators of *P. notoginseng* growth and soil health [13]. The diverse bacterial and fungal taxa inhabiting the healthy rhizosphere of *P. notoginseng* actively drive soil nutrient mineralization, optimize root morphological development, and alleviate autotoxic stress caused by root exudates [10,14]. Additionally, a well-balanced rhizosphere microbiome can effectively constrain the proliferation of soil-borne pathogens such as Fusarium and Phytophthora, the primary causal agents of root rot disease that severely restricts *P. notoginseng* yield and quality. In contrast, unreasonable cultivation practices disrupt microbial community assembly, reduce the abundance and function of beneficial microbial populations, and break rhizosphere ecological homeostasis. Such microbial dysbiosis exacerbates continuous cropping obstacles, suppresses plant growth, and impairs the accumulation of medicinal active substances in *P. notoginseng*. Notably, different cultivation modes reshape rhizosphere microenvironments by altering habitat stability and resource availability, which further restructures microbial diversity and functional profiles, ultimately leading to distinct growth performance and health status of *P. notoginseng* across cultivation systems [15].

During the entire growth and nurturing process of *P. notoginseng*, rhizosphere microorganisms continuously participate in and regulate plant physiological metabolism, nutrient acquisition, and stress adaptation throughout different developmental stages. Specific beneficial rhizosphere bacteria, including Lysobacter, Sphingomonas, and Arthrobacter, improve soil nitrogen and phosphorus cycling efficiency, promote root cell elongation and lateral root formation, and thereby enhance nutrient uptake capacity and above-ground and below-ground biomass accumulation of *P. notoginseng* [11]. Symbiotic arbuscular mycorrhizal fungi further colonize *P. notoginseng* roots, expand the soil absorption range of root systems, and activate jasmonic acid signaling pathways, which not only accelerate vegetative growth but also substantially promote the biosynthesis and accumulation of total saponins, the core medicinal components of *P. notoginseng*, in roots [16]. The enrichment and stable colonization of probiotic consortia can fundamentally restructure the disordered rhizosphere microecology of diseased *P. notoginseng*, significantly elevate the relative abundance of beneficial indigenous microbes, and markedly reduce the proliferation of potential soil-borne pathogens. Such microbial community reconstruction reverses the microecological imbalance of the rhizosphere, restore soil microbial ecological stability, and ultimately suppress root rot occurrence and reduce plant mortality in *P. notoginseng* [17]. Microbial community succession closely matches the unique growth rhythm of *P. notoginseng*, and the sustained colonization of functional beneficial microbes maintains rhizosphere microecological stability, guarantees continuous nutrient supply during long-term field growth, and ultimately determines the growth vigor, survival rate, and medicinal quality of *P. notoginseng*.

To reduce the negative effects associated with conventional field cultivation, alternative cultivation systems such as understory cultivation and semi-wild cultivation have increasingly been proposed [18].

Understory cultivation takes advantage of forest canopy buffering and natural litter input to provide a relatively moderated habitat, whereas semi-wild cultivation aims to mimic the native habitat of *P. notoginseng* by maintaining a low-input and low-disturbance environment [19]. These systems may reduce reliance on chemical inputs and improve ecological stability of the rhizosphere. Nevertheless, most previous studies have focused either on soil deterioration under continuous field cultivation or on the ecological effects of a single alternative mode. Direct comparisons among semi-wild, understory, and field cultivation systems remain scarce.

In particular, it is still unclear how these cultivation modes differ in rhizosphere soil properties, bacterial and fungal community composition, and soil–microbe associations. The main edaphic variables associated with microbial differentiation among cultivation modes also remain insufficiently resolved. Therefore, the present study compared rhizosphere soils of *P. notoginseng* under semi-wild, understory, and field cultivation systems in Yunnan Province, China. The objectives were to: (i) characterize differences in rhizosphere soil physicochemical properties among cultivation modes; (ii) compare bacterial and fungal diversity and community composition across cultivation systems; and (iii) identify the main soil factors associated with microbial community differentiation. We hypothesized that cultivation mode would primarily influence rhizosphere microecology through shifts in soil environmental conditions rather than through large changes in local-scale microbial diversity.

2 Materials and Methods

2.1 Study Sites and Sample Collection

In this study, three cultivation patterns, namely semi-wild cultivation (SW), understory cultivation (U), and conventional field cultivation (F), were investigated. Rhizosphere soil samples were collected from two-year-old Panax notoginseng in the autumn of 2023, with three biological replicates obtained at each sampling site. All sampling sites were located in Wenshan Prefecture, Yunnan Province, including Zhelaxiang Township and Ameng Town of Yanshan County, and the Huge Zone of Shupi Yi Township in Qiubei County. The SW site was situated in Zhelaxiang Township, Yanshan County (1654.8 m altitude; 23.4°N, 104.4°E). The U site was located in Ameng Town, Yanshan County (1524.7 m altitude; 23.4°N, 104.3°E). The F site was established in the Huge Zone of Shupi Yi Township, Qiubei County (1587.8 m altitude; 23.5°N, 104.6°E). The two-year-old Panax notoginseng plants and soil samples used in the three experimental plots corresponding to the three cultivation modes in this study were uniformly cultivated and provided by the Wenshan Sanqi Research Institute. All three sampling sites belonged to the karst landform area of southeastern Yunnan and featured a low-latitude plateau subtropical monsoon climate. The annual average temperature ranged from 15.2°C to 17.2°C. Such climatic conditions ensured sufficient water supply for crop growth, while the karst landform facilitated smooth drainage and effectively avoided soil waterlogging. Overall, all sampling sites met the growth requirements of *P. notoginseng*, which was suitable for conducting adaptive cultivation experiments and exploring the distribution of rhizosphere microbial communities.

This study further analyzed the differences in rhizosphere microbial communities of Panax notoginseng among the three cultivation patterns with 3 replicates. For each biological replicate, 5 healthy two-year-old plants with similar growth status were randomly selected within the corresponding cultivation site. After the plants were carefully excavated, loosely attached bulk soil was gently shaken off, and the soil tightly adhering to the root surface was collected as rhizosphere soil. Each biological replicate consisted of rhizosphere soil collected from multiple individual plants and thoroughly mixed to obtain one composite sample, thereby reducing the influence of plant-level variation. Rhizosphere soil tightly adhering to the root surface of two-year-old *P. notoginseng* was collected, and impurities were removed to obtain qualified

soil samples. All samples were immediately sealed in sterile plastic bags, transported to the laboratory with dry ice, and subsequently divided into two portions for subsequent experiments. One portion was stored at −80°C for high-throughput DNA sequencing, while the other portion was air-dried and sieved through a 2-mm mesh for the determination and analysis of soil physicochemical properties [20].

2.2 Soil Physicochemical Analyses

Soil physicochemical properties in all rhizosphere samples were analyzed following the methods described in Soil Agrochemical Analysis [21]. The measured variables included pH, soil organic carbon (SOC), total carbon (TC), total nitrogen (TN), C/N ratio, P_2O_5, alkali-hydrolyzable nitrogen (AN), ammonium nitrogen (NH^{4+}-N), nitrate nitrogen (NO^{3-}-N), available phosphorus (AP), and readily oxidizable carbon (ROC), dry/wet ratio (D/W), and ROC/SOC ratio (R/S). These variables were used to characterize nutrient status and edaphic differences among cultivation modes. Briefly, soil pH was determined using a soil-to-water ratio of 1:2.5 with a calibrated pH meter. SOC was measured using the potassium dichromate oxidation method. TC and TN were determined using an elemental analyzer, and the C/N ratio was calculated based on TC and TN contents. AN was determined using the alkaline hydrolysis diffusion method. NH^{4+}-N and NO^{3-}-N were extracted using KCl solution and then measured colorimetrically. AP was determined using the molybdenum-antimony colorimetric method after extraction. ROC was measured using the potassium permanganate oxidation method.

2.3 DNA Extraction, PCR Amplification, and Amplicon Sequencing

Microbial DNA was extracted from rhizosphere soil using the E.Z.N.A.® Soil DNA Kit (Omega Bio-Tek, Norcross, GA, USA) according to the manufacturer's instructions. DNA purity was examined using a NanoDrop 2000 spectrophotometer (Thermo Fisher Scientific, Waltham, MA, USA) based on the A260/A280 and A260/A230 ratios, and DNA integrity was checked by 1% agarose gel electrophoresis. For bacterial community analysis, the V3-V4 region of the 16S rRNA gene was amplified using primers 338F and 806R [22]. For fungal community analysis, the ITS region was amplified using primers ITS1F and ITS2R [23]. PCR amplification was performed under the following conditions: initial denaturation at 95°C for 2 min, followed by 27 cycles of 95°C for 30 s, 55°C for 30 s, and 72°C for 60 s, with a final extension at 72°C for 5 min. PCR products were recovered from 2% agarose gels and purified using the AxyPrep DNA Gel Extraction Kit (Axygen Biosciences, Union City, CA, USA). Purified libraries were quantified using the Qubit dsDNA HS Assay Kit (Thermo Fisher Scientific), and samples with concentrations of approximately 50–100 ng/μL were pooled in equimolar amounts. Amplicon sequencing was performed on the Illumina MiSeq PE300 platform (Shanghai Biozeron Co., Ltd., Shanghai, China). All samples were sequenced in a single run to reduce potential batch effects. Negative controls containing molecular-grade water instead of soil were included throughout DNA extraction, PCR amplification, and sequencing to monitor contamination. Reads were imported into QIIME2 for downstream analyses [24]. Amplicon sequence variants (ASVs) were inferred using the DADA2 denoise-paired pipeline with truncation lengths of 220 bp for both forward and reverse reads. Singletons and chimeras were removed during denoising [25]. Taxonomic assignment was performed using the classify-sklearn algorithm in QIIME2, with the SILVA database for bacteria and the UNITE database for fungi [23,26].

2.4 Statistical Analyses

To reduce the influence of unequal sequencing depth, all samples were rarefied to the minimum sequencing depth before diversity analysis using the rarefaction procedure implemented in the vegan

package [27]. Alpha diversity was assessed using the ACE richness index and Shannon diversity index. The ACE index was used to estimate microbial richness, whereas the Shannon index was used to evaluate both richness and evenness. Differences in alpha diversity among cultivation modes were tested using one-way analysis of variance followed by Duncan's multiple range test. Beta diversity was calculated based on Bray-Curtis dissimilarity and visualized by principal coordinates analysis (PCoA). Differences in microbial community composition were evaluated using PermANOVA analysis. Indicator taxa associated with different cultivation modes were identified using indicator species analysis [28]. A correlation heatmap was generated using the pheatmap package in R to visualize correlations [29].

3 Results

3.1 Rhizosphere Soil Physicochemical Properties under Different Cultivation Modes

Rhizosphere soil properties varied among the three cultivation modes, indicating that different management regimes were associated with distinct edaphic environments. In our results, semi-wild cultivation has relatively higher SOC and a lower pH than the other cultivation modes (Table 1). The understory system generally exhibited intermediate characteristics between semi-wild and field cultivation. These patterns suggest that cultivation mode may influence rhizosphere soil conditions through differences in nutrient input, litter accumulation, and disturbance intensity.

Table 1: Soil properties in the three cultivation modes.

	pH	SOC (g/kg)	TC (g/kg)	TN (g/kg)	CN	P_2O_5 (g/kg)	AN (mg/kg)	NH_4^+-N (mg/kg)	NO_3^--N (mg/kg)	Dry/Wet	AP (mg/kg)	ROC (mg/kg)	R/S
SW	4.98	60.22	46.25	3.13	19.26	0.061	408.31	52.84	37.48	0.84	167.34	6.13	0.10
U	4.78	35.90	23.15	1.38	26.07	0.056	220.80	53.69	48.61	0.82	19.63	4.42	0.12
F	5.22	23.60	14.23	1.07	21.96	0.074	136.40	13.77	18.58	0.81	16.50	3.74	0.15

3.2 Alpha Diversity of Rhizosphere Bacterial and Fungal Communities

Alpha diversity analysis showed that cultivation mode had limited effects on local-scale microbial richness and diversity in the rhizosphere of *P. notoginseng* (Fig. 1). Neither the ACE index nor the Shannon index differed significantly among semi-wild, understory, and field cultivation systems for either bacterial or fungal communities ($p > 0.05$). Although no significant differences were detected, several numerical trends were observed. For bacteria, both the ACE richness and the Shannon index were the lowest under understory cultivation. For fungi, ACE richness was highest under field cultivation and lowest under semi-wild cultivation, while Shannon diversity was relatively similar between semi-wild and field cultivation and lower under understory cultivation. These findings suggest that contrasting cultivation modes did not substantially change the overall richness or evenness of rhizosphere microbial communities. Therefore, the microbial response to cultivation mode may not be adequately reflected by alpha diversity.

3.3 Beta Diversity of Rhizosphere Microbial Communities

In contrast to alpha diversity, beta diversity analyses revealed clear differences in microbial community composition among cultivation modes (Fig. 2). PCoA analysis based on Bray-Curtis distance and PermANOVA results showed that both bacterial and fungal communities were separated according to cultivation mode ($p < 0.05$), indicating that semi-wild, understory, and field cultivation supported distinct rhizosphere microbial assemblages. For bacterial communities, samples from field and semi-wild cultivation were relatively independent on the PCoA2 axis, whereas understory samples distributed between the other two groups. For fungal communities, the separation among cultivation modes was more pronounced.

Semi-wild samples were clearly distinguished from the other two groups on the PCoA1 axis ($p < 0.05$). Overall, these results suggest that cultivation mode was closely associated with microbial community composition, and that fungal communities were more responsive than bacterial communities to differences among cultivation systems. This suggests that the ecological effect of cultivation mode lies mainly in restructuring microbial community composition rather than changing microbial diversity.

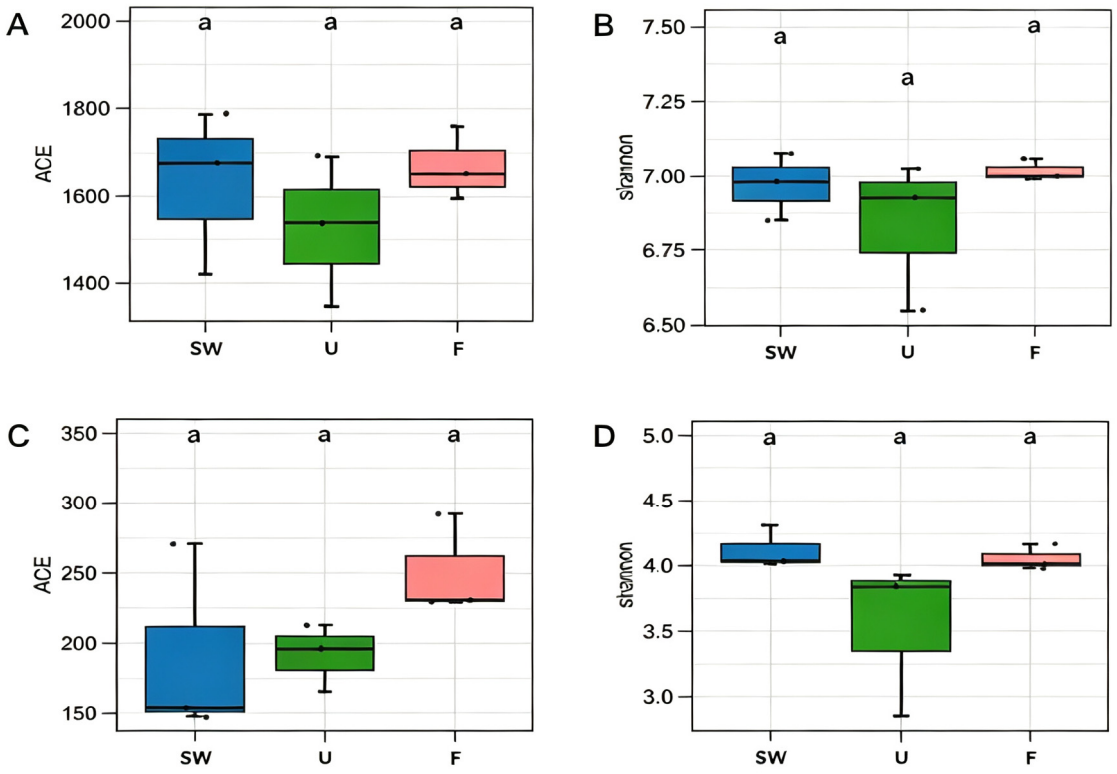

Figure 1: ACE and Shannon indices of bacteria (**A**,**B**) and fungi (**C**,**D**) under different cultivation methods. Different letters indicate significant differences between groups based on Duncan's multiple range test ($p < 0.05$).

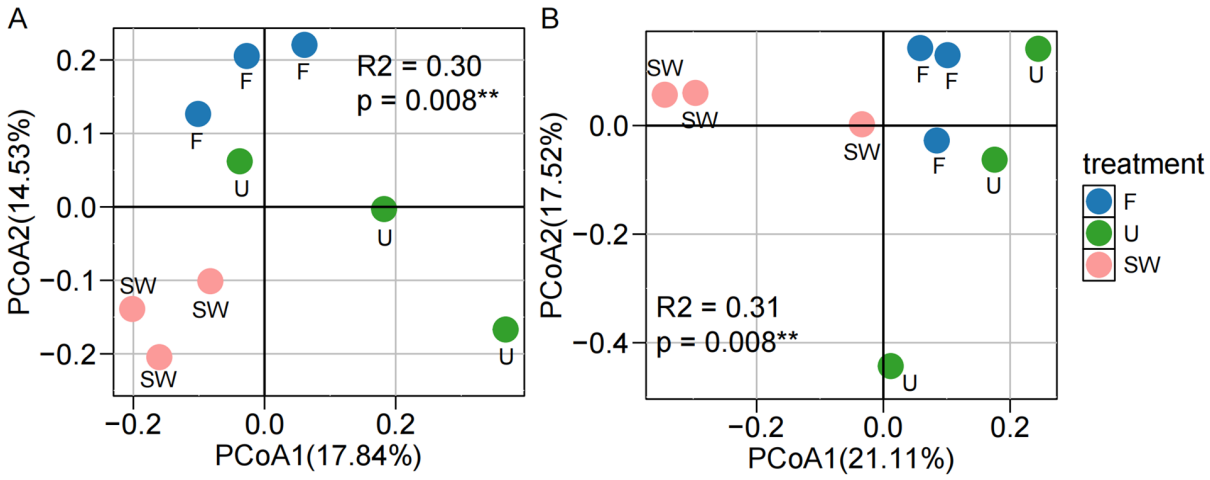

Figure 2: β-diversity of bacteria (**A**) and fungi (**B**) based on PCoA analysis and PermANOVA (**: $p < 0.01$).

3.4 Taxonomic Composition of Rhizosphere Microbial Communities

The class-level composition of bacterial and fungal communities further demonstrated cultivation mode-dependent microbial differentiation (Fig. 3). Anaerolineae, Gammaproteobacteria, and Alphaproteobacteria accounted for large proportions of the bacterial community. However, the relative abundance of specific bacterial classes varied among cultivation modes. In understory cultivation, Anaerolineae and Alphaproteobacteria showed relatively higher abundance than in the other treatments, while Thermoanaerobaculia and Desulfuromonadia were more abundant in semi-wild cultivation. Fungal communities showed more distinct compositional differences among cultivation modes. Semi-wild cultivation was characterized by the enrichment of Dothideomycetes and the depletion of Sordariomycetes compared to understory and field cultivation.

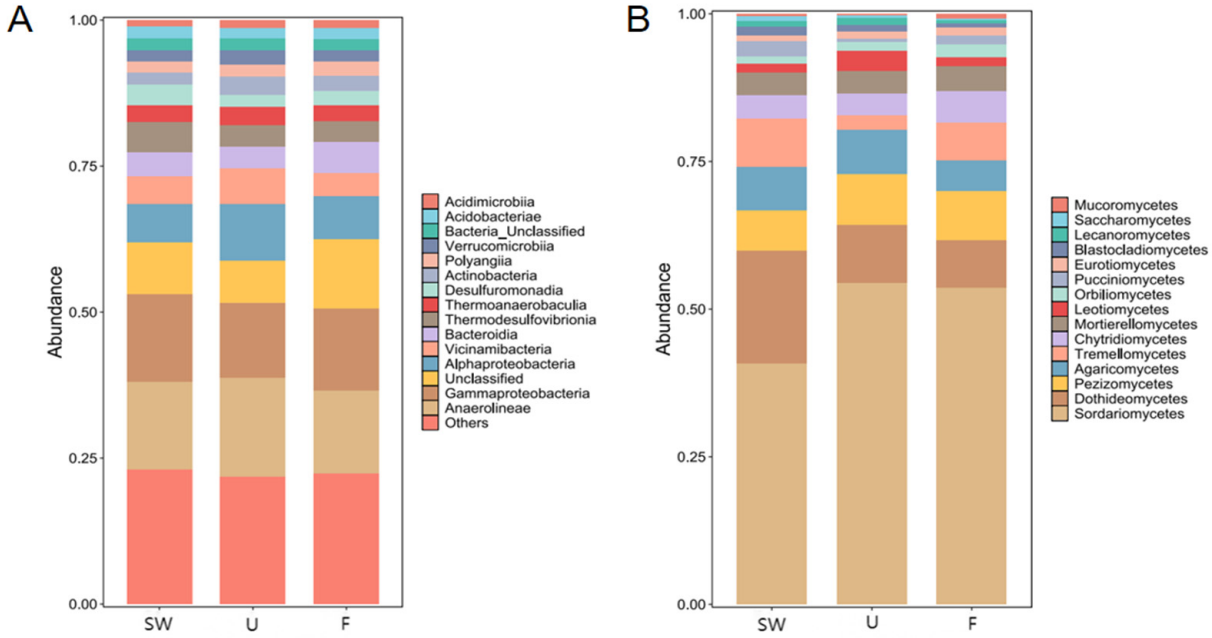

Figure 3: Community composition of bacterial communities (**A**) and fungal communities (**B**).

Indicator species analysis further confirmed that cultivation mode selected for specific microbial taxa (Fig. 4). A relatively large number of cultivation mode-specific bacterial indicator ASVs were identified. There were 9 indicators in the semi-wild cultivation belonging to Gammaproteobacteria, most indicators of U belonged to Anaerolineae (8), while most indicators in the F belonged to Gammaproteobacteria (6), and Alphaproteobacteria (5). However, fewer fungal indicator ASVs were detected, most of which belonged to Dothideomycetes. Besides, there were indicators belonging to Fusarium in the Sordariomycetes. This result suggests that bacterial communities exhibited stronger fine-scale taxonomic differentiation among cultivation modes, while fungal differences were mainly reflected in shifts in the class level. Together, these results indicate that cultivation mode selectively enriched different bacterial and fungal groups, even though overall alpha diversity did not differ significantly. This suggests that the ecological effect of cultivation mode lies mainly in restructuring microbial community composition rather than changing total microbial diversity.

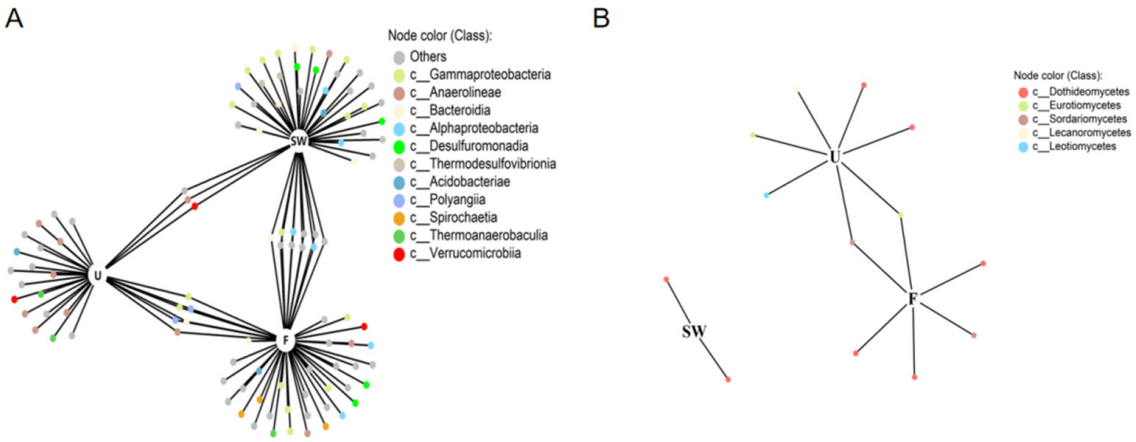

Figure 4: Differential ASVs of bacteria (**A**) and fungi (**B**) in different planting patterns, with colors representing different classes. SW: Semi-wild cultivation; U: Understory cultivation; F: Conventional field cultivation.

3.5 Associations between Soil Properties and Microbial Communities

Spearman correlation analysis revealed strong associations between soil physicochemical variables and microbial communities. Among the measured edaphic factors, including pH, SOC, ROC, TN, NO_3^--N, NH_4^+-N, and P_2O_5 were related to bacterial and fungal community variation in the Panax notoginseng rhizosphere, with different microbial taxa exhibiting significant functional differentiation in their responses to environmental factors (Fig. 5).

For bacterial communities, several taxa associated with beneficial biocontrol functions, including Actinobacteria, Bacilli, Clostridia, and Thermomicrobia, were positively correlated with soil carbon, nitrogen, and phosphorus nutrient indices, but negatively correlated with pH, suggesting that the high organic matter and acidic conditions of semi-wild cultivation may favor these taxa. In contrast, some copiotrophic bacterial groups, such as Methylomirabilia and Desulfobacteridia, were positively related to inorganic nitrogen and available nitrogen, suggesting an association with the high nitrogen input and soil acidification conditions typical of conventional field cultivation. Additionally, Acidobacteriia and Bacteroidia exhibited specific response trends to pH and nutrients, respectively, reflecting an overall fine-tuned adaptation strategy of bacterial communities to habitat changes.

For fungal communities, the response patterns directly corresponded to the ecological differentiation between pathogenic and beneficial taxa. Sordariomycetes and Dothideomycetes, which contain major soil-borne pathogens of *P. notoginseng*, were positively correlated with available phosphorus and rhizosphere effects. This pattern suggests that conventional field cultivation may favor fungal groups commonly associated with disease risk. By contrast, beneficial symbiotic or saprotrophic fungi, such as Glomeromycetes, Archaeorhizomycetes, and Agaricomycetes, showed positive associations with soil organic carbon and negative associations with inorganic nitrogen, indicating a preference for the low-nitrogen and high organic matter conditions of semi-wild cultivation. Meanwhile, Elaphomycetes and Entomophthoromycetes were highly positively related to inorganic nitrogen and negatively correlated with pH, acting as indicator fungi for the high-nitrogen and acidified environment of conventional field cultivation.

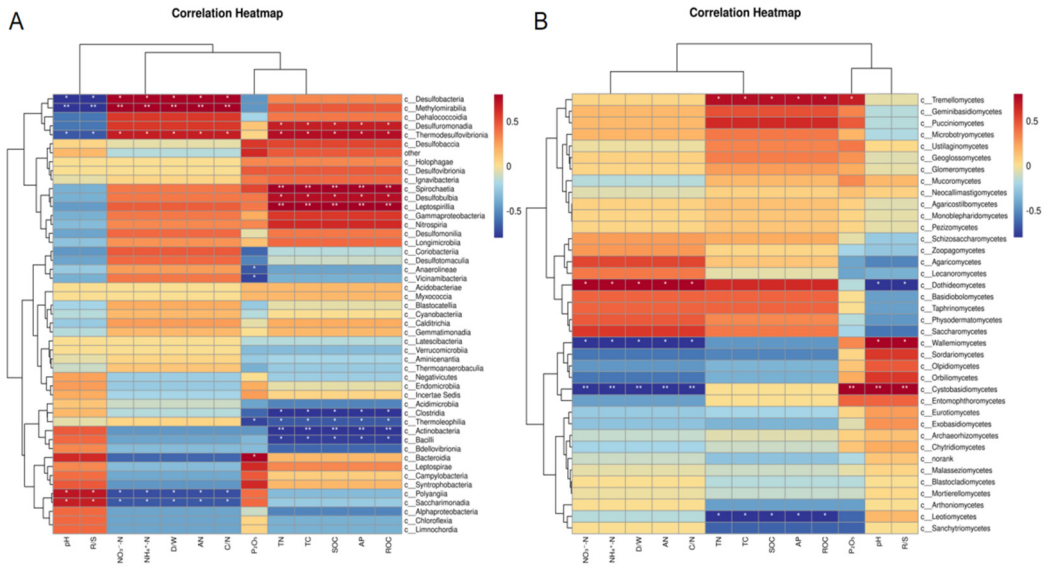

Figure 5: Spearman's correlation between Class-level bacteria (**A**) and fungi (**B**) with environmental factors (Significance levels after FDR correction: p: <0.05*, <0.01**).

3.6 Analysis of Microbial Function Prediction

The three cultivation modes of Panax notoginseng significantly reshaped the metabolic functions of rhizosphere bacteria and the ecological functional group structure of fungi (Fig. 6). The functional annotation of bacteria in each mode identified carbohydrate metabolism, amino acid metabolism, and energy metabolism as the core functions, but there were significant functional differentiations among the modes: field cultivation bacteria had higher abundances in pathways related to rapid cell proliferation, such as replication and repair, translation, and nucleotide metabolism, showing characteristics of high growth and metabolism in eutrophic environments; semi-wild cultivation bacteria dominated in pathways related to stress resistance and interspecies interactions, such as environmental adaptation, signal transduction, and membrane transport, being more adapted to natural complex habitats; understory cultivation bacteria exhibited intermediate functional characteristics, showing a transitional pattern. The ecological functional groups of fungi showed higher abundances of beneficial symbiotic groups such as saprophytic fungi and endophytes in the understory and semi-wild modes, with stronger abilities in organic matter decomposition and plant symbiotic interactions, resulting in a more stable and healthy rhizosphere microecology; whereas field cultivation had lower abundances of symbiotic fungi and relatively enriched pathogenic fungi, leading to a weaker balance in the microbial community and a higher risk of microecological imbalance.

Overall, this indicates that semi-wild and understory cultivation are more conducive to forming a healthy rhizosphere microorganism system with diverse functions and harmonious interactions, while field cultivation microorganisms tend to focus on rapid proliferation, with poorer ecological stability, which is an important microbiological basis for the easy occurrence of soil-borne diseases and continuous cropping obstacles.

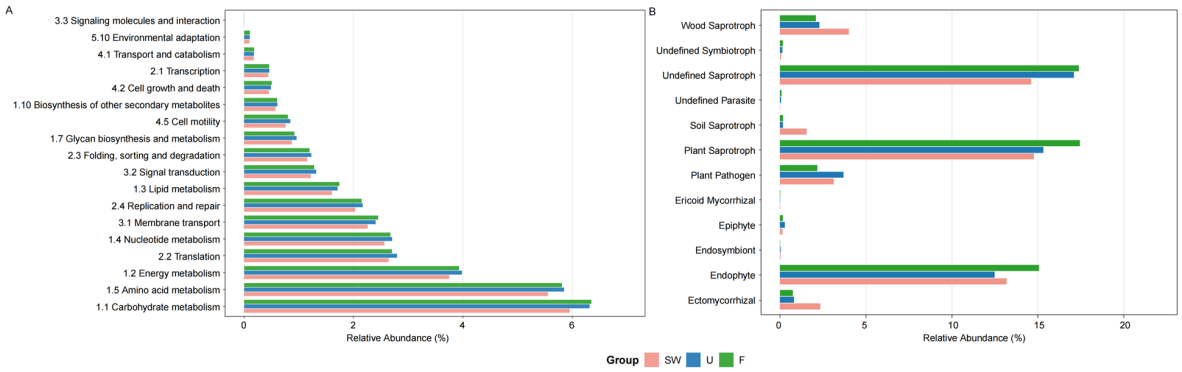

Figure 6: Relative abundance of functions in rhizosphere bacteria (**A**) and fungi (**B**) under three cultivation patterns.

4 Discussion

This study provides a comparative evaluation of rhizosphere soil properties and microbial communities under three cultivation systems of *Panax notoginseng*. Our results showed that cultivation mode was more strongly associated with microbial community composition than with microbial diversity. Specifically, ACE and Shannon indices did not differ significantly among cultivation modes, whereas beta-diversity analysis revealed differences in bacterial and fungal community composition. This pattern indicates that the microbial response to cultivation mode was mainly reflected by community turnover rather than by diversity. Therefore, the ecological influence of cultivation mode should be interpreted together with community composition, dominant taxa, indicator taxa, and soil environmental variables. Such a result is consistent with the view that soil microbial richness can remain relatively stable under environmental variation, whereas microbial composition is often more sensitive to changes in resource availability, pH, nutrient status, and habitat conditions [30].

The distinct edaphic environments observed among cultivation modes may partially explain microbial differentiation. Semi-wild systems typically involve minimal soil disturbance and continuous organic matter input from litter, which promotes carbon accumulation and supports a more buffered soil environment. In contrast, conventional field cultivation often relies on frequent fertilization and disturbance, leading to nutrient loss. Such changes in soil chemical conditions are known to influence microbial habitat suitability, resource competition, and physiological stress, thereby shaping community assembly processes [31]. The strong associations between SOC, pH, and inorganic nitrogen with microbial taxa further support the central role of soil environmental filtering. Carbon availability is a key determinant of microbial metabolism and growth strategies, with higher SOC generally favoring taxa involved in organic matter turnover and anaerobic or facultative processe [32]. The enrichment of Desulfuromonadia and Thermodesulfovibrionia under semi-wild conditions is consistent with this interpretation, as these groups are often associated with carbon-rich and reduced environments. In contrast, the higher abundance of copiotrophic or nitrogen-associated taxa under field cultivation may indicate that nutrient enrichment may favor fast-growing microbial groups adapted to high resource availability.

Semi-wild cultivation was characterized by a higher relative abundance of Dothideomycetes and certain bacterial groups associated with carbon-rich environments. Although Dothideomycetes include both saprotrophic and pathogenic members, their enrichment under semi-wild conditions, together with lower abundance of Sordariomycetes, may reflect a shift toward a more balanced fungal community with reduced dominance of pathogenic taxa. This pattern is consistent with the hypothesis that low-disturbance systems support more stable and functionally diverse microbial assemblages, potentially enhancing ecological

resilience [33]. An additional notable result is that fungal communities exhibited stronger separation among cultivation modes than bacterial communities. This suggests that fungi may be more sensitive to changes in cultivation conditions, particularly those related to organic matter input and plant-derived substrates. Fungi play a central role in decomposing complex organic compounds and are closely linked to plant health through both mutualistic and pathogenic interaction [34]. Therefore, shifts in fungal community structure may have disproportionate impacts on rhizosphere functioning and plant performance. The understory cultivation system generally displayed intermediate characteristics in both soil properties and microbial community structure. This transitional pattern suggests that understory systems may partially mitigate the negative effects of field cultivation while not fully reproducing the ecological conditions of semi-wild habitats. The higher within-group variability observed in understory samples further indicates that this system may be more heterogeneous, possibly due to variation in canopy structure, litter input, and microenvironmental conditions.

It should be noted that the present study was observational rather than experimentally controlled. Therefore, these results should be interpreted as associations rather than direct evidence that cultivation mode alone caused the observed microbial shifts. Although all sampling sites were located within Wenshan Prefecture and shared broadly similar regional climatic conditions, geographic distance and site-specific factors may still confound cultivation-mode effects. This limitation should be considered when interpreting the relationship between cultivation system and rhizosphere microbial structure. Seasonal variation, interannual climate differences, and long-term microbial succession under continuous cropping were not assessed. Future studies using paired-site designs, larger spatial replication, and controlled field experiments are needed to more clearly separate cultivation-mode effects from geographic effects. Moreover, functional prediction amplicon data is inherently uncertain and should be treated only as functional potential.

Despite these limitations, this study provides useful preliminary evidence that contrasting cultivation modes are associated with differences in rhizosphere soil properties and microbial community composition in *P. notoginseng*. The results support the hypothesis that cultivation mode influences rhizosphere microecology primarily through modification of soil environmental conditions rather than through large shifts in microbial diversity [35,36]. The coupling between soil properties and microbial community composition highlights the importance of managing soil carbon inputs, nutrient balance, and pH stability in *P. notoginseng* cultivation [9]. From an ecological perspective, semi-wild cultivation appears to provide a rhizosphere environment more similar to natural systems, characterized by higher carbon availability, reduced nitrogen accumulation, and a microbial community less dominated by potential pathogenic taxa. These findings have implications for sustainable cultivation strategies of medicinal plants. By promoting soil conditions that favor balanced microbial communities and suppress disease-associated taxa, low-disturbance and carbon-enriched systems may contribute to improved plant health and potentially higher quality of medicinal compounds. However, further studies integrating functional gene profiling, pathogen quantification, and plant metabolite analysis are needed to establish direct links between microbial community shifts and medicinal quality outcomes.

5 Conclusions

Cultivation mode was closely associated with rhizosphere soil properties and microbial community composition in *Panax notoginseng*. Although bacterial and fungal alpha diversity did not differ significantly among semi-wild, understory, and field cultivation systems, both community composition and dominant taxa varied substantially across cultivation modes. Semi-wild cultivation was associated with higher relative abundance of Actinobacteria and Agaricomycetes. While field cultivation, was associated with greater proportions of Proteobacteria and Sordariomycetes and with soil conditions characterized by higher

inorganic nitrogen and lower pH. Across all samples, SOC, pH, and inorganic nitrogen emerged as the soil variables most strongly associated with microbial community differentiation. These findings indicate that contrasting cultivation strategies can influence rhizosphere microecology mainly through their effects on soil environmental conditions. From a soil ecological perspective, semi-wild cultivation appears to provide a rhizosphere environment more favorable to microbial community balance and less dominated by pathogen-associated fungal groups. This study provides a basis for further evaluation of ecologically oriented cultivation systems for sustainable *P. notoginseng* production.

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Availability of Data and Materials: Data available on request from the authors. The data that support the findings of this study are available from the corresponding author, Linlin Dong, upon reasonable request.

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