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Soil Microbial Community Responds to Elevation Gradient in an Arid Montane Ecosystem in Northwest China

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Abstract: This study sought to clarify the composition of soil fungal and bacterial communities along an elevation gradient in an arid montane ecosystem as well as the influence of environmental factors (soil properties, climate, topography, and plant diversity) upon soil microbial community structures. Four vegetation types—montane desert steppe (mean elevation: 1761 m), montane shrub (mean elevation: 2077 m), subalpine coniferous forest (mean elevation: 2485 m), and subalpine shrub (mean elevation: 2903 m)—were sampled on the western slope of the Helan Mountains. The 16SrRNA gene and ITS1 were performed by single-molecule real-time (SMRT) sequencing with the PacBio sequencing platform. The Chao1 and Shannon–Wiener diversity of soil fungi and bacteria were more diverse in the soil of the lower elevation gradient compared to that of the upper one. Differences in abundance among phyla were found via One-way ANOVA(analysis of variance), yet the dominant soil fungal phyla (Ascomycota, Basidiomycota, and Mortierellomycota) and bacterial phyla (Proteobacteria, Acidobacteria, and Bacteroidetes) were the same across the elevation gradient. Pearson correlations and redundancy analysis (RDA) indicated that plant diversity (Shannon–Wiener diversity [*H*] and Margalef richness [*D*]), solar radiation, mean annual temperature, soil organic matter, soil moisture content, slope, mean annual precipitation, and elevation all significantly influenced the community composition of different soil fungal and bacterial phyla. Although plant diversity significantly affects fungal and bacterial diversity, the results imply that the influence of plant functional diversity on soil microbial community variation should not be ignored.

Keywords: Helan Mountains; high-throughput sequencing; microbial community; vegetation type

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1. Introduction

Microorganisms are fundamental biological components of soil ecosystems, and their community composition, diversity, and functioning can significantly influence various ecological and biochemical processes, such as plant community assembly, the flux of nutrients, and carbon storage [1]. Bacteria are undoubtedly the most abundant, diverse group of soil microorganisms, whose role in soil formation, litter decomposition, and nutrient cycling is paramount [2]. Fungi, another large group of soil microorganisms, figure prominently in multiple ecosystem processes, such as decomposing litter [3], forming mutualisms with plants [4–6], and shaping the plant community via pathogenic activity [7]. Initially, it was thought that soil microorganisms were globally distributed at random [8,9], due to their small size and strong dispersal ability [1]. However, mounting empirical evidence is showing that the composition and diversity of soil microbial communities could instead exhibit a regular spatial distribution [10,11].

The spatial distribution and abundance of soil microbial communities are jointly driven by a variety of factors [12,13]. Elevation gradient is recognized for exerting one of the most

important effects on soil microbial community structure and diversity. Along an elevation gradient, predictable shifts in environmental factors occur, such as temperature, humidity, and light, whose rate of change is 1000 times faster than along the latitude gradient [14,15]. Recent studies of elevation effects on soil microbial diversity have uncovered neutral, single peak, or negative patterns [16]. For example, Singh et al. [17] found that soil microbial diversity on Jeju Island, Korea, was U-shaped on its sunny slope but unimodal (hump-shaped) on its shady slope. In a subtropical forest, Wu et al. [18] reported the diversity of its soil microbial communities gradually decreased as elevation increased. Later work by Li et al. [19] revealed that soil microbial community diversity in forests and meadows below and above the treeline, respectively, differed in their altitudinal patterns, perhaps because of distinct soil factors shaping microbial communities at the treeline. Collectively, these findings suggest ecosystems with different zonal and azonal soils, temperature and precipitation regimes, and vegetation, could strongly influence both the composition and structure of the soil microbial community.

Lying within a montane ecotone of the monsoon and non-monsoon regions of China, the flora and vegetation of the Helan Mountains are distinguished by transitional features, which make it a highly diverse but fragile ecosystem [20]. The Helan Mountains are also the boundary between the desert and temperate steppe ecosystems in Northwest China. The vegetation of the Helan Mountains, as distributed upward along its elevation gradient, shows a remarkable spatial pattern: desert steppe, montane shrub, subalpine coniferous forest, and subalpine shrub. Many researchers have investigated the plant diversity [21] and soil nutrient cycling [22] of the Helan Mountains, but our knowledge of the spatial patterns of its soil microbial community remains limited [23,24]. The Helan Mountains are divided into an eastern steeper slope and a western slope. This distinctive topography generates stark differences in climate, soil, and plant species. To date, only our team [25], as well as Ma et al. [24], have studied the distribution and drivers of soil microbial community on the eastern slope, by using high-throughput sequencing and phospholipid fatty acid (PLFA), respectively. However, the soil microbial community patterns and drivers along the elevation gradient on the western slope remain unclear. In this study, we used high-throughput sequencing technology to investigate the composition and diversity of soil microbial communities along an elevation gradient on the western slope of the Helan Mountains. Our aim was to explore the shift in patterns of soil microbial communities along that elevation gradient and to elucidate the environmental factors driving the shifts in this understudied arid montane ecosystem.

2. Materials and Methods

2.1. Study Area

The Helan Mountains (38°27′–39°30′ N, 105°41′–106°41′ E) stretch and divide the Inner Mongolia Plateau and the Ningxia Plain in a North-South orientation. Annual precipitation varies from 200 mm at the foot of the mountains to 500 mm at their peak elevation (3556 m a.s.l.), and the mean annual evaporation can reach 2000 mm. Four vegetation types are distinguishable along the elevation gradient: desert steppe in the foothills, montane shrub in low mountainous areas, and subalpine coniferous forest and subalpine shrub at higher elevations. Their soil types are gray calcium soil, skeleton soil, gray cinnamon soil, and subalpine shrub soil, respectively [26].

2.2. Plots and Collecting of Samples

Three transect lines (located along three ranges, named 'Beisi', 'Hallau', and 'Nansi') were established along the elevation gradient, from north to south, on the western slope of the Helan Mountains (Figure 1). In each transect line, we selected a sampling site in each of the four vegetation types (i.e., desert steppe, montane shrub, subalpine coniferous forest, and subalpine shrub). With a hierarchical sampling design, tree plots, shrub subplots, and herb quadrats were established for plant diversity estimation and soil sampling. At each site, three replicate tree sampling plots (20 m × 20 m) more than 100 m apart were

set up at the same elevation. Due to the lower elevation of the Beisi range, the subalpine shrub is absent. A total of 33 sampling tree plots were established. In each plot, three shrub sampling subplots (5 m × 5 m) were randomly positioned, with a single herb sampling quadrat (1 m × 1 m) nested in their center. Tree identity, diameter at breast height (DBH), tree height, and crown size of all live trees above 1.3 m in height were recorded in every tree plot. For shrubs, their respective identity, height, and crown size in each subplot were recorded. In the quadrat, the identity, height, and ground cover of herb plants were recorded. Five soil cores (4-cm diameter, 20-cm depth) were collected from the 0–20 cm layer in each tree sampling plot and mixed as one soil sample. These soil samples were divided into two parts: one was kept in ice and then delivered to labs (State Key Laboratory Breeding Base of Land Degradation and Ecological Restoration of Northwest China, Ningxia University), frozen at −80 °C for high-throughput sequencing of the soil microbial community, and the other part was air dried and sieved through a 2-mm mesh to measure soil physical and chemical properties.

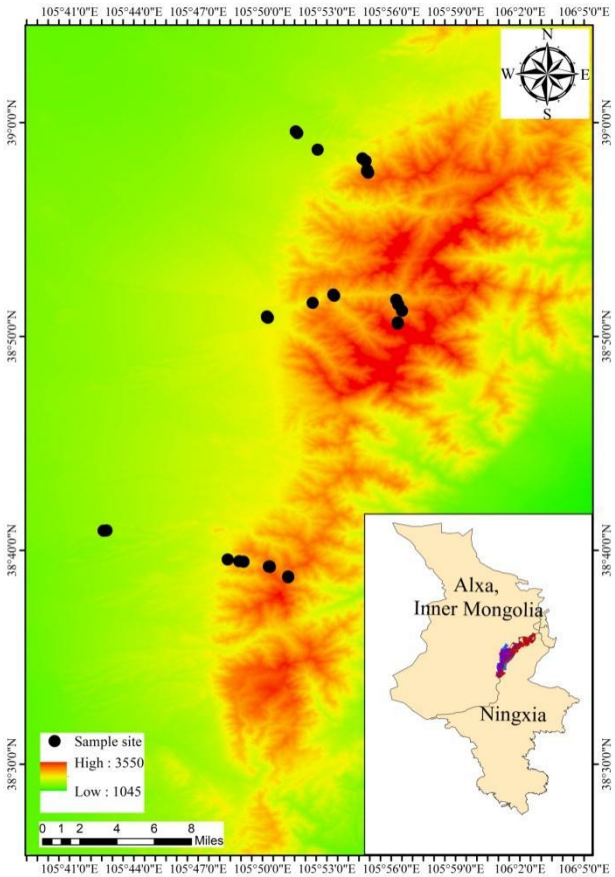

Figure 1. Map of the study area in the Helan Mountains of China, showing the locations of sampling sites along the three transects spanning an elevation gradient.

2.3. Soil Physical and Chemical Properties

Stone and plant roots were carefully separated and removed from the soil samples. The oven-drying method was used to measure soil moisture content (MC) by drying the soil samples at 105 °C for 24 h. Soil pH was measured in a 1:2.5 volume soil:water suspension by using a compound electrode (Leici Instrument, Shanghai, China) [14]. Soil organic carbon (SOC) was quantified by the potassium dichromate titration method; soil total nitrogen (TN) was quantified by the semimicro-Kjeldahl method; soil total phosphorus (TP) was quantified by the molybdenum antimony colorimetric method. All procedures for the determination of SOC, TN, and TP were based on the methods described in Gregorich and Carter [27].

2.4. Plant Diversity

The Margalef richness (D) and Shannon–Wiener diversity (H) were calculated for each plot, subplot, and quadrat [28].

$$D = S - 1/\ln N \quad (1)$$

where S is the number of species and N is the abundance.

$$H = -\sum_{i}^{S} P_i \ln P_i \quad (2)$$

where P_i is the relative abundance of the species i; $P_i = N_i/N$, where N_i is the density of the species i, S is the number of species, and N is the total number of individuals.

2.5. Climate and Topography

The latitude, longitude, and elevation of each plot were recorded with a portable GPS(global positioning system) unit (GPSMAP 62sc, Garmin, Shanghai, China). Climate data, obtained from a global meteorological database (http://www.worldclim.org, accessed on 1 July 2021), consisted of the annual mean temperature (MAT), average annual precipitation (MAP), solar radiation (SPRAD), and vapor pressure (VAPR). Slope (SLOP) and aspect (ASP) were obtained from a digital elevation model (DEM), which is based on the coordinates of the tree plots.

2.6. High-Throughput Sequencing of Fungi and Bacteria

High-throughput sequencing of soil microbial community was performed externally by the BMK Biotechnology Co., China. Soil DNA extraction was carried out by using a soil DNA extraction kit (Omega Bio-tek, Norcross, GA, USA), and the purity and concentration of the extracted total DNA were examined by 1% agarose gel electrophoresis. Sequence amplification of the fungal ITS1 region was conducted with the primer sequences ITS1F (5′-CTTGGTCATTTAGAGGAAGTAA-3′) and ITS2R (5′-GCTGCGTTCTTCATCGATGC-3′) [29]. For the PCR amplifications of the V3–V4 region of the bacterial 16SrRNA gene, the primer sequences used were 338F (5′-ACTCCTACGGGAGGCAGCAG-3′) and 806R (5′-GGACTACHVGGGTWTCTAAT-3′) [30]. All PCR amplifications were run by using high-fidelity DNA polymerase on an ABI GeneAmp® 9700 PCR instrument (PerkinElmer, ABI, Waltham, MA, USA). Each PCR reaction consisted of 2 μL of 10 × FastPfu buffer, 2 μL of dNTPs (2.5 mmol·L^{-1}), 0.8 μL of each forward/reverse primer (5 μM), and 0.2 μL of rTaq polymerase. For the DNA template PCR amplifications, the conditions were as follows: initial denaturation at 98 °C for 2 min, 25–30 cycles at 98 °C for 15 s, annealing at 55 °C for 30 s, followed by an extension at 72 °C for 5 min [31]. The target fragments were enzymatically cleaved, and the PCR products were recovered by using the AxyPrepDNA gel recovery kit (AXYGEN, Corning, Wujiang, Jiangsu China) then purified and quantified to form a library for sequencing. This was implemented by using the single-molecule real-time sequencing (SMRT Cell) method based on the PacBio sequencing platform. All sample data were divided based on PCR primer sequences and barcode sequences. The sequencing data were analyzed by using the Quantitative Insights Into Microbial Ecology (QIIME 1.9.1) toolkit. Then, the FLASH (v1.2.11) was used to remove low-quality or ambiguous reads to make high-quality reads. According to an identity of 97%, the sequences were clustered into taxonomic operating units (OTUs) in UPARSE (v7.0.1090). Representative sequences of these OTUs with a 97% homology were taxonomically annotated by referring to the UNITE (v8.0) fungal ITS database and the SILVA (v132) bacterial database.

2.7. Data Analysis

Alpha diversity indices of OTUs (i.e., Shannon–Wiener diversity [H], Margalef richness [D], and Chao1) and plant diversity (D and H) were computed in PAST4.09 [32]. After environmental variables, soil fungi, and bacteria diversity and soil sample OTUs passed the equal variance and normality test, One-way ANOVAs (analysis of variance) were performed in PAST4.09, and Tukey's post hoc pairwise was conducted, with the

significance level set at $p < 0.05$. Person's correlation was conducted to measure the relationship between soil microbial composition and environmental factors. Non-metric multidimensional scaling analysis (NMDS analysis) was applied to the OTUs of soil fungal and bacterial communities at different sites and elevations based on the binary_jaccard algorithm. Redundancy analysis (RDA) was used to explore the relationships between soil microbial diversity and climate (annual mean temperature [MAT], average annual precipitation [MAP], solar radiation [SPRAD]), topography (elevation [ALT], slope [SLOP], aspect [ASP]), soil properties (soil moisture content [MC], soil organic carbon [SOC], nitrogen [TN], pH, phosphorus [TP]) and plant diversity(*D* and *H*). Monte Carlo permutation tests with data replacement were run to determine whether the effects of environmental factors were statistically significant. The NMDS of soil microbial communities, community stacking plots, and heatmaps of Person's correlation at the phylum level were plotted using the "vegan" package of R 3.6.3 [33].

3. Results

3.1. Soil, Climate, and Plant Diversity along the Elevation Gradient

The distribution of soil physicochemical properties, climate, topography, and plant diversity at different elevations on the western slope of the Helan Mountains are shown in Table 1. Soil pH, TN, and TP remained similar, whereas SOC tended to increase and then decrease with rising elevation, peaking in the subalpine coniferous forest at 2485 m. Moreover, the MC at high elevations (2485 and 2903 m) was significantly higher than that at low and middle elevations (1761 and 2077 m). Moving up the elevation gradient, both MAP and SPRAD increased while MAT declined. Plant diversity *H* and *D* were greatest in the montane shrub vegetation type (at 2077 m) and lowest in the subalpine coniferous forest (at 2485 m).

Table 1. Soil physiochemical properties and environmental factors along the elevation gradient.

Factors	Elevation (m a.s.l.)			
	1761 m	2077 m	2485 m	2903 m
pH	7.86 ± 0.32	7.86 ± 0.43	7.74 ± 0.37	7.66 ± 0.47
MC (%)	8.55 ± 3.32 b	8.76 ± 2.98 b	22.38 ± 10.16 a	18.90 ± 2.97 a
TN ($g \cdot kg^{-1}$)	0.34 ± 0.14	0.58 ± 0.84	0.29 ± 0.16	0.38 ± 0.17
TP ($g \cdot kg^{-1}$)	0.80 ± 1.11	0.74 ± 0.53	0.84 ± 0.65	0.22 ± 0.22
SOC ($g \cdot kg^{-1}$)	30.75 ± 27.61 b	51.00 ± 25.77 b	126.50 ± 38.42 a	47.83 ± 13.50 b
ALT (m)	1761.97 ± 173.43 d	2077.74 ± 127.18 c	2485.27 ± 78.73 b	2903.99 ± 78.24 a
MAP (mm)	211.33 ± 13.91 d	240.22 ± 14.22 c	265.00 ± 9.73 b	287.50.11 ± 4.50 a
SRAD ($kJ \cdot m^{-2} \cdot day^{-1}$)	16522.64 ± 115.98 d	16394.59 ± 107.57 c	16263.94 ± 88.82 b	16092.92 ± 4.33 a
MAT (°C)	6.19 ± 1.16 a	3.63 ± 1.05 b	1.88 ± 0.63 c	0.55 ± 0.72 d
ASP (°)	297.90 ± 41.18 ab	234.92 ± 126.65 ab	204.67 ± 137.18 b	334.69 ± 9.17 a
SLOP (°)	3.77 ± 1.86 c	22.11 ± 4.70 b	26.81 ± 3.08 a	24.60 ± 2.02 ab
H	1.54 ± 0.28 b	2.31 ± 0.25 a	0.44 ± 0.09 d	1.11 ± 0.37 c
D	1.41 ± 0.49 b	3.17 ± 0.67 a	0.26 ± 0.02 c	0.99 ± 0.44 b

Note: MC, soil moisture content; TN, total nitrogen; TP, total phosphorus; SOC, soil organic carbon; ALT, elevation; MAP: average annual precipitation; SRAD, sun radiation; MAT, average annual temperature; ASP, aspect; SLOP, slope; *H*, Shannon–Wiener diversity; *D*, Margalef richness. Values are the mean ± SD; different lowercase letters a, b, c and d within the same row indicate significant differences ($p < 0.05$).

3.2. Soil Microbial Diversity and Composition along the Elevation Gradient

3.2.1. Fungal and Bacterial Diversity

Both the Chao1 and Shannon–Weiner diversity of soil fungi or bacteria were highest under montane shrub vegetation at 2077 m elevation. For fungi, the lowest Chao1 and Shannon–Weiner values were found in the desert steppe at 1761 m. For bacteria, the Shannon–Weiner diversity was lowest in the subalpine coniferous forest at 2485 m, while the Chao1 value was lowest in desert steppe at 1761 m (Table 2).

Table 2. Diversity of soil fungi and bacteria along the elevation gradient. Values are the mean ± SD; different lowercase letters a and b within the same row indicate significant differences ($p < 0.05$).

Elevation (m)	Fungi		Bacteria	
	Chao1	Shannon	Chao1	Shannon
1761	161.82 ± 51.61 b	4.45 ± 1.21 b	1276.54 ± 192.76 b	8.77 ± 0.28 b
2077	227.84 ± 63.34 a	5.53 ± 0.37 a	1659.75 ± 166.15 a	9.10 ± 0.20 a
2485	174.43 ± 33.06 ab	4.45 ± 0.75 b	1307.11 ± 150.58 b	8.67 ± 0.32 b
2903	165.22 ± 43.06 b	4.60 ± 0.90 ab	1345.40 ± 213.88 b	8.82 ± 0.20 ab

The stress value was less than 2 for the NMDS of soil fungi and bacteria along the elevation gradient, indicating the reliability of our NMDS analysis. The distribution of fungal and bacterial communities at different elevations revealed substantial intra-group similarity and pronounced inter-group variability, implying different compositions between soil fungal and bacterial communities at different elevations. Specifically, both the soil fungi and bacteria at 1761 m were more discrete than at 2077 m, 2485 m, and 2903 m, which revealed the more composition variability of soil microbial community appeared in montane desert steppe (Figure 2).

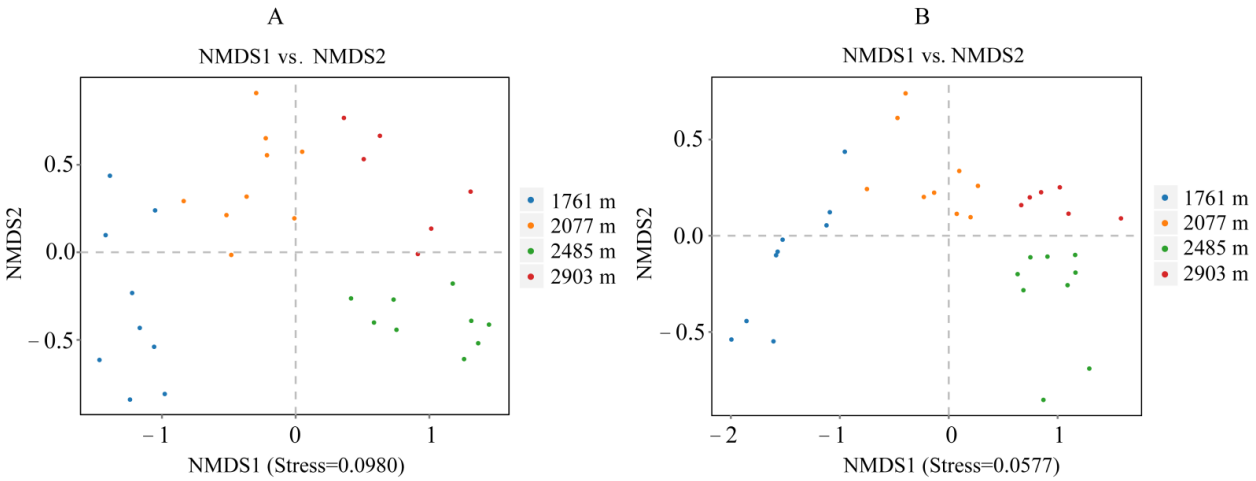

Figure 2. Non-metric multidimensional scaling analysis of soil fungal (**A**) and bacterial (**B**) communities at different elevations.

3.2.2. Fungal and Bacterial Community Composition

The fungal community members with a relative abundance >1% belonged to five phyla: Ascomycota, Basidiomycota, Mortierellomycota, Glomeromycota, and Chytridiomycota. The Ascomycota, Basidiomycota, and Mortierellomycota have abundances exceeding 10%, together accounting for more than 95% of the total abundance of OTUs (Figure 3A). Hence, these were the dominant fungal phyla in the soil, irrespective of elevation. Bacteria belonged to ten phyla—Verrucomicrobia, Gemmatimonadetes, Planctomycetes, Actinobacteria, Chloroflexi, Rokubacteria, Nitrospirota, Bacteroidetes, Acidobacteria, and Proteobacteria. Among them, Proteobacteria, Acidobacteria, and Bacteroidetes constituted 70% of the total abundance, making them the dominant bacteria present in soil along the elevation gradient (Figure 3B). The ANOVA results show that Ascomycota decreased significantly with elevation ($F = 68.4348$, $p < 0.001$) increasing, being lowest at 2485 m though significantly higher at 2903 m (Figure 4A). By contrast, the abundance of Basidiomycota increased significantly with higher elevation ($F = 81.1623$, $p < 0.001$). Glomeromycota was lower than other phyla, and it was significantly reduced at 2485 m in the subalpine coniferous forest ($F = 20.2637$, $p < 0.001$). Although the abundance of Mortierellomycota was considerable (3.1%–19.9%), its high intra-group variation did not result in significant differences among the four elevations (Figure 4A).

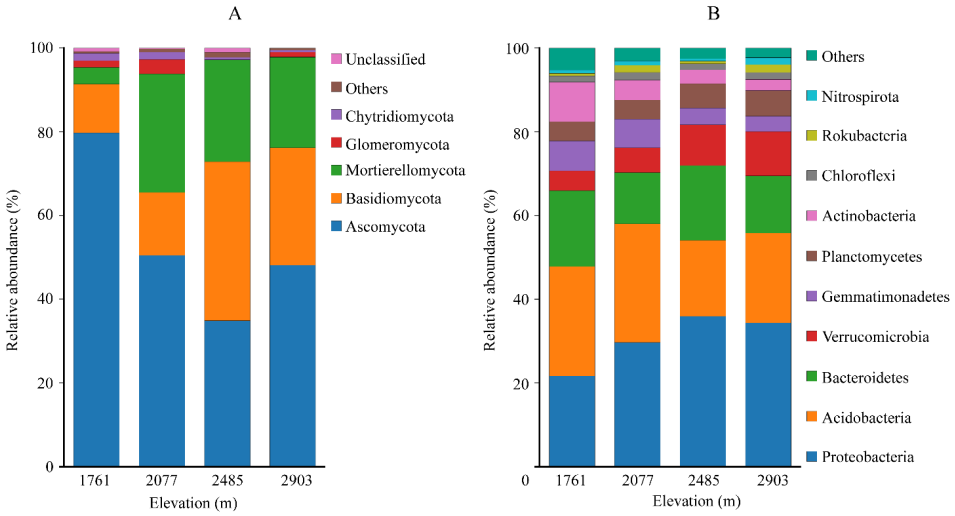

Figure 3. The relative abundance of fungal (**A**) and bacterial (**B**) phyla present in soil at different elevations corresponding to four vegetation types.

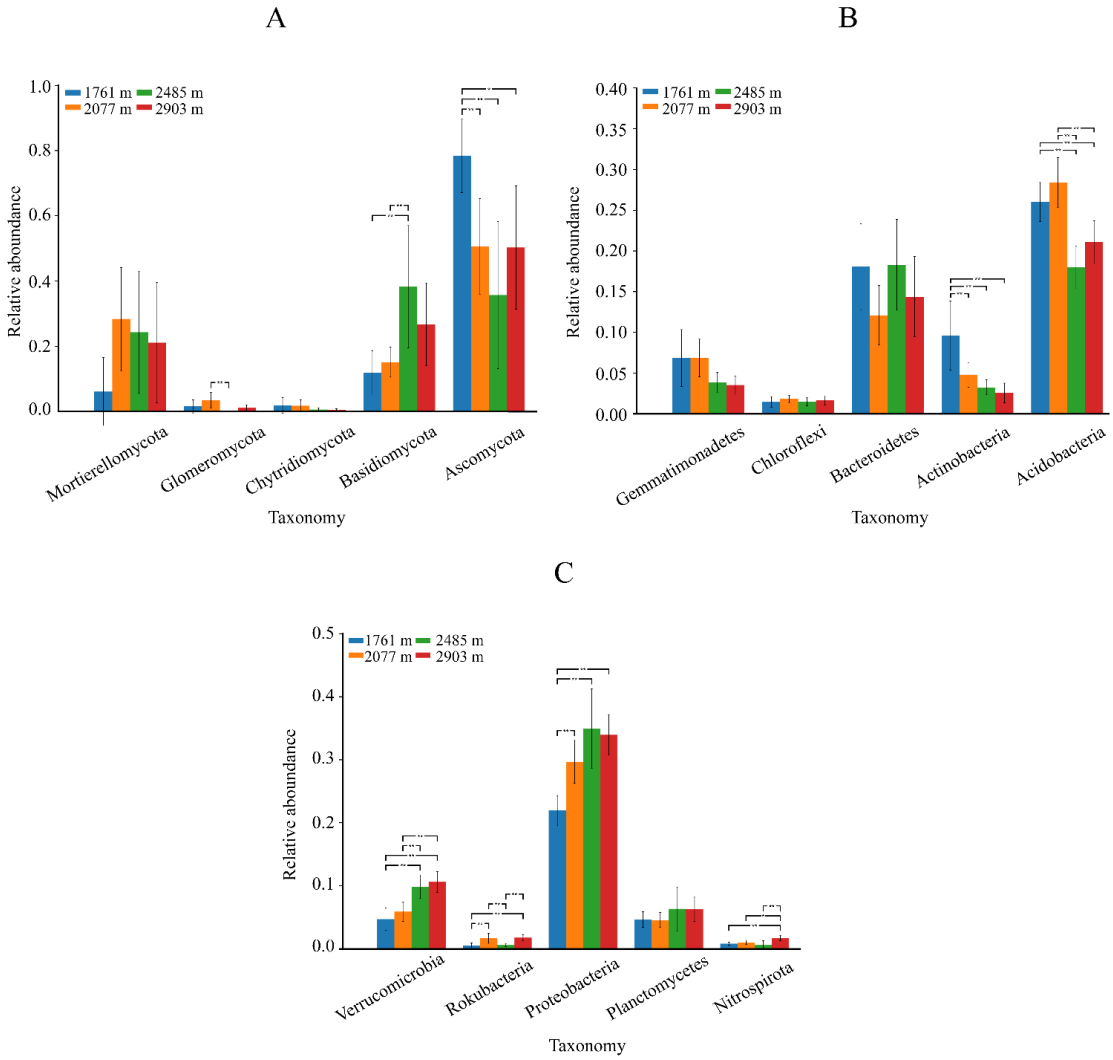

Figure 4. Comparison of the relative abundance of major phyla of fungi (**A**) and bacteria (**B**,**C**) in the soil communities at different elevations. Asterisks indicate significance level. Signif. Codes: $p < 0.05$ (*), $p < 0.01$ (**), $p > 0.05$ (blank).

Among the bacterial communities, Proteobacteria significantly increased ($F = 22.4559$, $p < 0.001$) in relative abundance moving up the elevation gradient (Figure 4B), whereas Acidobacteria were significantly more abundant ($F = 12.3158$, $p < 0.001$) at lower (1761 and 2077 m) than higher elevations (2485 and 2903 m). The relative abundance of Bacteroidetes did not significantly change along the elevation gradient (Figure 4C).

3.3. Soil Microbial Community and Environmental Factors

3.3.1. Relationships between Soil Microbial Composition and Environmental Factors

According to the heatmaps and RDA results (Figures 5 and 6), the dominant soil fungi phyla clustered into three groups based on their relationship to environmental factors. The first group comprised the Mortierellomycota and Basidiomycota, whose abundances significantly increased with an increasing ALT, MAP, MC, SOC, and SLOP. In contrast, both Ascomycota and Chytridiomycota were significantly more abundant at higher temperatures and in arid areas at low elevations. Chytridiomycota may be considered a separate group, being significantly more abundant where plant diversity was higher. Soil TP, TN, and pH did not seem to significantly affect any of the dominant fungi.

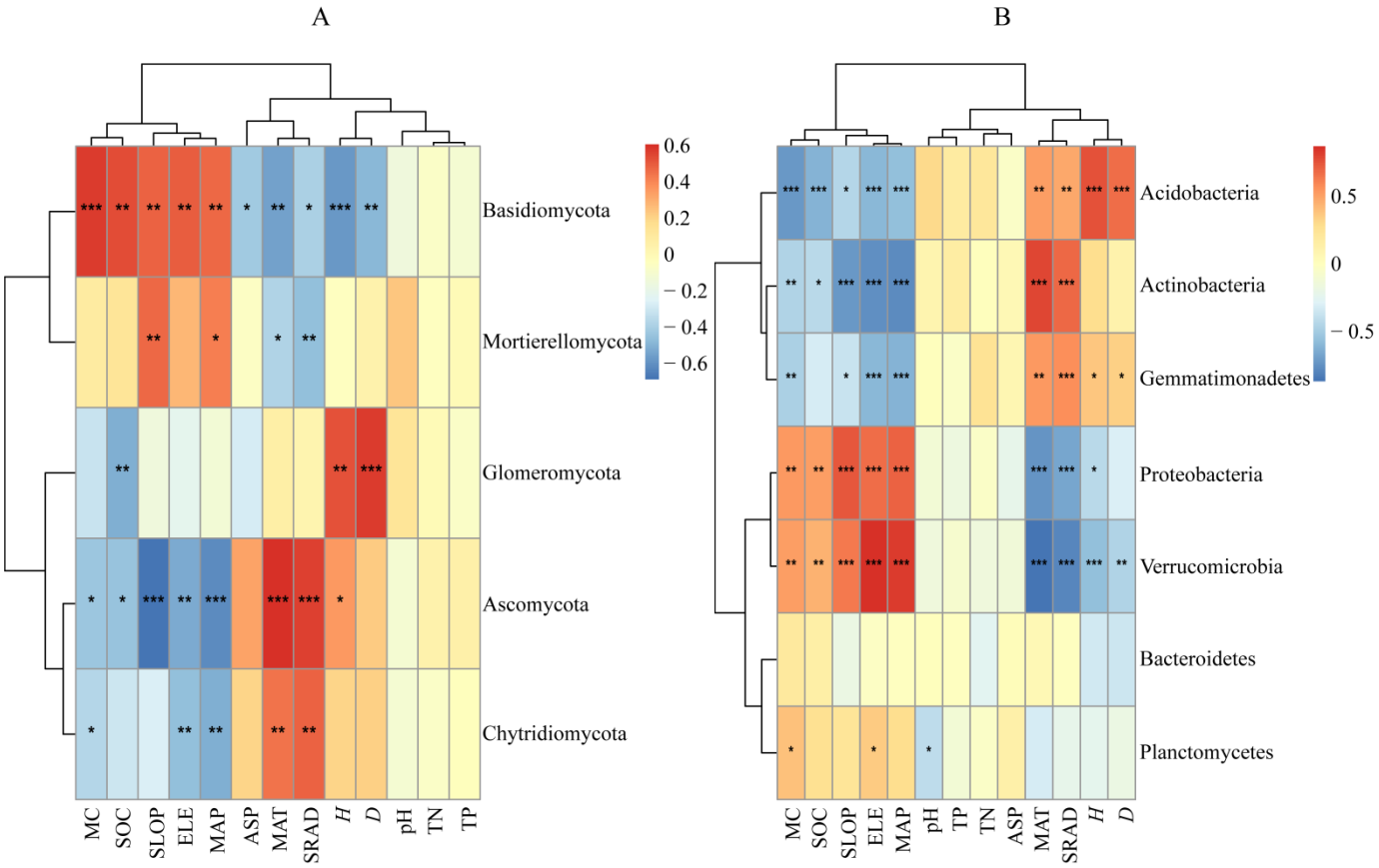

Figure 5. Heatmaps of soil fungi (**A**) and bacteria (**B**) vis-à-vis the environmental factors. Red represents positive correlations and blue represents negative correlations. MC, soil moisture content; TN, total nitrogen; TP, total phosphorus; SOC, soil organic carbon; ALT, elevation; MAP: average annual precipitation; SRAD, sun radiation; MAT, average annual temperature; ASP, aspect; SLOP, slope; *H*, Shannon–Wiener diversity; *D*, Margalef richness. Asterisks indicate significance level. Signif. Codes: $p < 0.05$ (*), $p < 0.01$ (**), $p < 0.001$ (***), $p > 0.05$ (blank).

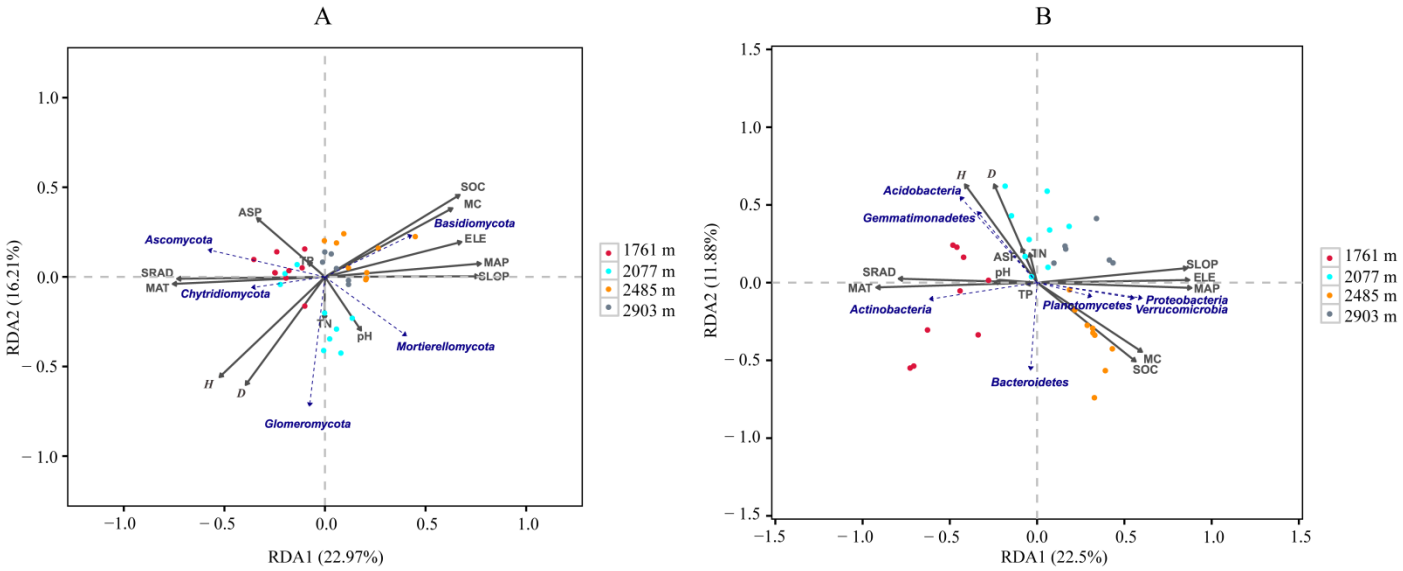

Figure 6. RDA of the soil microbial community at phylum level across the four elevations. The solid close arrows are the environmental factors; the dash close blue arrows represent the dominant phylum of soil fungi (**A**) and bacteria (**B**) that were included in the models. The direction of the arrow indicates a positive or negative correlation between the environmental factors and the ordination axes. The angle of the arrow reflects the strength of correlation between the environmental factors, with small angles indicating strong correlations. MC, soil moisture content; TN, total nitrogen; TP, total phosphorus; SOC, soil organic carbon; ALT, elevation; MAP: average annual precipitation; SRAD, sun radiation; MAT, average annual temperature; ASP, aspect; SLOP, slope; *H*, Shannon–Wiener diversity; *D*, Margalef richness.

The relationships between the dominant bacteria phyla and their environment identified three bacterial groups. The first consisted of the Actinobacteria, Acidobacteria, and Gemmatimonadetes, all of which were more abundant at low elevations, wherein Acidobacteria and Gemmatimonadetes were more abundant in the montane shrub community that also harbored the greatest plant diversity. Proteobacteria, Planctomycetes, and Verrucomicrobia clustered in a different group associated with higher elevations. The Bacteroidetes were not significantly affected by any of the examined environmental factors.

3.3.2. Relationships between Soil Microbial Diversity and Environmental Factors

According to Pearson correlations, only pH had a significant positive correlation with fungal Chao1, but pH negligibly influenced the diversity of Bacteria. SOC had a significant negative relationship with bacterial Shannon–Wiener diversity. Plant diversity *H* and *D* showed significant positive correlations with bacterial diversity, indicating that plant richness could influence the composition of the below-ground microbial community (Table 3).

Table 3. Pearson correlation coefficients between fungal or bacterial diversity and 13 environmental factors.

Environment	Fungi		Bacteria	
	Chao1	Shannon	Chao1	Shannon
pH	0.450 **	0.148	0.034	0.046
MC	−0.062	−0.198	−0.120	−0.185
TN	0.296	0.189	−0.018	0.100
TP	0.272	0.060	0.103	0.153

Table 3. *Cont.*

Environment	Fungi		Bacteria	
	Chao1	**Shannon**	**Chao1**	**Shannon**
SOC	−0.155	−0.219	−0.246	−0.403 *
ALT	−0.029	−0.161	−0.016	−0.073
D	0.306	0.286	0.461 **	0.461 **
H	0.356 *	0.324	0.583 **	0.517 **
MAT	−0.066	0.115	−0.113	0.007
MAP	0.060	−0.117	0.079	−0.005
SRAD	−0.108	0.118	−0.130	−0.068
ASP	−0.125	0.033	−0.087	0.186
SLOP	0.159	0.080	0.215	0.018

Note: MC, soil moisture content; TN, total nitrogen; TP, total phosphorus; SOC, soil organic carbon; ALT, elevation; MAP: average annual precipitation; SRAD, sun radiation; MAT, average annual temperature; ASP, aspect; SLOP, slope; *H*, Shannon–Wiener diversity; *D*, Margalef richness. Asterisks indicate significance level. Signif. Codes: $p < 0.05$ (*), $p < 0.01$ (**), $p > 0.05$ (blank).

4. Discussion

The variation of diversity and abundance in soil microbial communities depends on environmental factors. Ping et al. [34] found that the overall composition of microbial communities may differ greatly along the elevation gradient in Changbai Mountains, China, while their dominant groups remain mostly similar in terms of relative abundance. Our results support this view. On the western slope of the Helan Mountains, the dominant phyla of soil fungal communities along the elevation gradient were the same, consisting of Ascomycota, Basidiomycota, and Mortierellomycota, and likewise, for bacterial communities, the dominant phyla were Proteobacteria, Acidobacteria, and Bacteroidetes. Further, the dominant phyla of fungi and bacteria matched those found on the eastern slope of the Helan Mountains [25], implying the stability of these phyla through the mountain slope. These results suggest that elevation is not a factor determining which phyla become dominant in the soil microbial community of the Helan Mountains (Figures 5 and 6; Table 3). Although our study's sampling sites and plant species differed from those investigated elsewhere, similar dominant phyla of the soil microbial community were found relative to the same plant functional groups as inferred from the vegetation type [35]. For example, Dassen et al. [36] also found that the functional identity of plants had a strong influence on soil microbial community composition.

Many studies have demonstrated a relationship between plant diversity and soil microbial diversity, but there is yet no consensus on this pattern, nor a consistent understanding of the factors driving it [37,38]. In the Changbai Mountains, China, plant diversity declines with rising elevation but the diversity of eukaryotic microbes does not change markedly [39]. Johnson et al. [40] found that soil microbial diversity and plant diversity were positively associated. We also found a positive relationship between plant and microbial diversity; for instance, soil fungal and bacterial richness and diversity were greatest in the montane shrub vegetation type, whose plant diversity was also greatest (Table 2). A significant positive relationship between plant diversity and soil microbial (especially bacterial) diversity was confirmed by Pearson's correlation coefficient test (Table 3).

However, our results reveal that desert steppe featured a remarkably higher plant species richness than either subalpine coniferous forests or subalpine shrubs (Table 1), yet their soil microbial diversity remained similar (Table 2). This is in contrast with studies that have shown that similar plant functional groups (grasses, legumes, small herbs, tall herbs) have a similar soil microbial composition [36]. Some studies have presented evidence that soil microbial diversity and composition are affected more by plant functional diversity (e.g., community-weighted means of trait values (CWM) or the sum of pairwise functional distances between species weighted by their relative abundances) than plant species diversity (the number of different species) [41,42]. As opposed to plant species diversity, plant functional diversity represents the functional trait distance, or difference in terms of one or

more functionally traits (e.g., leaf size, seed size, canopy height, etc.) between plants [43]. Our inconsistent conclusions about how plant diversity is related to soil microbial diversity may be interpreted from the perspective of plant diversity. Accordingly, further studies are needed to verify the impact of plant functional traits or diversity on soil fungal or bacterial diversity in the Helan Mountains.

In addition, some environmental factors are recognized as being significant drivers of soil microbial communities. Soil type [44], climate [45], and topography [13] are usually the most influential in determining the composition of soil microbial communities in montane ecosystems. Consistent with other findings [46,47], soil fungal and bacterial community structures in our study were mainly influenced by moisture and temperature (these are mainly determined by local climate types and elevation gradient). However, unlike some other studies [48–50], as well as our study on the eastern slope of the Helan Mountains [25], the influence of TN or pH on the composition and diversity of the soil microbial community was negligible on the western slope of the Helan Mountains. Compared to the steep eastern slope, the western slope rises slowly with a gradual rise in elevation, which likely explains why the soil TN and pH changed negligibly across the elevation gradient (Table 1). Therefore, microbial community assemblies do not depend on TN and pH.

5. Conclusions

We investigated the composition and structure patterns of soil fungal and bacterial communities along an elevation gradient on the western slope of the Helan Mountains. We found that soil microbial community diversity was significantly greater at lower elevations than that of the upper ones. However, the dominant phyla were consistent across all elevations sampled. The dominant soil fungal phyla were Cysticercales, Streptomycetes, and Peridiomycetes, and the dominant bacterial phyla were Aspergillus, Acidomycetes, and Synechococcus. RDA and Pearson correlation coefficients revealed the environmental factors (plant diversity, solar radiation, mean annual temperature, soil organic matter, soil moisture content, slope, mean annual precipitation, and elevation) significantly contributed to shaping the community composition of different phyla. Especially, plant diversity significantly influenced soil bacterial diversity. However, one should note that the presence and abundance of different phyla of fungi and bacteria are unable to illustrate their activity in nature (for example, mineralizing, catabolic, enzymatic, respiratory) in our study. Further research is needed to investigate the function of soil microbes and their relationship to the environment.

Our study strengthens the current understanding of the patterns of soil fungal and bacterial communities along an elevation gradient on the western slope of the Helan Mountains. It also paves a way forward to better predict the response of microbial communities to changing environmental conditions in mountainous regions.

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