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The composition of soil fungal communities is more dependent on biocrust type than on shrub cover in the Mu Us Desert

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ABSTRACT

Desertification-control policies have been applied in the Mu Us Desert since the 1950s. The landscape there is characterized by patches of shrub plants and well-developed lichen and moss crusts, some covered by shrub canopies and some in interspace soils. Little is known about how shrub cover and biocrusts shape soil fungal community structure in this ecosystem. Using high-throughput amplicon sequencing, the effects of biocrust types and shrub cover on soil fungal communities were analyzed. The results showed that biocrust types were more important than shrub cover in affecting soil properties and shaping soil fungal communities. Among all the measured soil properties, significant effects of shrub cover on soil pH and available P were observed. Biocrust types had significant effects on soil total organic carbon, C:N, and C:P ratios. Fungal taxa relating to plant pathogens and formation of lichens, (e.g., the Eurotiomycetes and Dothideomycetes and the of genera *Endocarpon* and *Knufia*) were dominant across biocrust types and shrub cover. Furthermore, although relative abundances of dominant fungal taxa were statistically similar among microhabitats, abundances of lichenized and pathogenic fungi differed significantly among biocrust types, with the former showing higher abundances in lichen crusts, and the latter exhibiting higher abundances in moss crosts. Soil total nitrogen and C:N were correlated with fungal community structure. Our results highligh the dominant role of biocrust types over shrub cover in shaping soil fungal communities in the Mu Us Desert. With the succession from lichen to moss crusts, increasing N limitation (soil TOC:TN ratio) may drive higher abundances of pathogenic fungi in lichen crusts and fewer lichenized fungi in moss crusts.

1. Introduction

Biological soil crusts (biocrusts), are formed by the combination of surface soil with various organisms, including eukaryotic microalgae, cyanobacteria, heterotrophic bacteria, fungi, lichen, and moss. It is documented that more than 70% of the global dry land surface is covered by biocrusts (Belnap et al., 2016; Weber et al., 2016; Rodriguez-Caballero et al., 2018). The biocrust development theory is known as the general successional sequence, which suggests that the biocrust is formed in a chronological order, with different photosynthetic organisms emerging on the soil surface (Weber et al., 2016). The well-known pioneer colonizers are the filamentous and mobile cyanobacteria (e.g., *Microcoleus* spp). Later, species like *Nostoc* and *Scytonema* that bind to the soil become dominant, forming cyanobacterial crusts.

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When lichen species (e.g., *Collema* spp. and *Toninia* spp.) and bryophytes successionally colonize, lichen and moss crusts are formed (Belnap and Eldridge, 2003; Büdel et al., 2016; Weber et al., 2016). The biocrusts have essential ecological benefits, for example binding soil particles, enhancing soil stability, increasing soil aggregation, and decreasing soil erosion (Bowker et al., 2006; Büdel et al., 2016; Lange and Belnap, 2016; Rosentreter et al., 2016).

Microbial communities are shaped simultaneously by stochastic and deterministic processes resulting from dispersal events on a large spatial scale (e.g., from several to thousands of kilometers) and by environmental filtering on a microhabitat scale, e.g., at the mm scale or smaller (HilleRisLambers et al., 2012; Xu et al., 2021a). UV radiation, temperature variability and soil water availability are the key factors shaping dryland soil microbial communities (Charley and West, 1977; Xie and Steinberger, 2001; Lüneberg et al., 2019). In drylands, the heterogenous landscapes shaped by microhabitats such as shrub cover and biocrust types affect these factors through the ecological roles of shrub canopies, plant roots, lichens, and mosses. Therefore, these microhabitats are crucial for the survival of microorganisms. For example, a microhabitat with the presence of shrub cover (shrub(+)) may have lower ultraviolet (UV) radiation and temperature variability and higher water availability and nutrient contents than that with the absence of shrub cover (shrub (−)). With different thicknesses and photosynthetic organisms, microhabitats created by different biocrust types may also show differences in stress, nutrient conditions, and biotic interactions. With minimal differences in climatic factors, habitat heterogeneity is documented to promote the beta diversity of plants, insects, and microorganisms at small geographic scales (Amarasekare, 2003). Previous studies also suggest that biomass and compositions of microbial communities are strongly related to microhabitat types (Bates and Garcia-Pichel, 2009; Steven et al., 2014). Moreover, a prior study demonstrated that microbial community compositions were significantly different regarding shrub(−) vs. shrub(+) (Mueller et al., 2015). Further, photosynthetic organisms, bacterial and fungal communities are also reported to be distinct regarding different types of biocrusts (Albright et al., 2019; Xu et al., 2020). However, our understanding of how soil fungal communities is shaped in desert biocrusts is still not complete. While some studies have examined the simultaneous effects of shrub cover and biocrust types on microbial communities, the majority of these investigations were conducted in North America or Europe (Belnap and Eldridge, 2003; Mueller et al., 2015; Büdel et al., 2016; Albright et al., 2019). The corresponding East Asian literatures are exceedingly rare, so it is still not clear whether similar or distinct patterns would be observed in East Asia, e.g., deserts in China. In addition, we do not know if the patterns are driven by similar abiotic factors.

Before the 1950s, the Mu Us Desert was characterized by an arid climate, strong wind erosion, and severe sandstorms resulting from moving sand dunes. Those stressful conditions were challenging for species survival, and brought significant harm to local agricultural and pastoral production. To control the desertification, the Chinese government initiated a series of policies, including the erection of straw checkerboards and planting xerophytic shrub species. Currently, the heterogenous landscape there is characterized by patches of shrub plants and well-developed lichen and moss crusts occupying the soil surface. We collected samples considering two types of microhabitats. The biocrust types (lichen vs. moss crusts), and the shrub cover (shrub(-) vs. shrub(+)). We regarded soil properties as quantitative indicators of microhabitats. We then compared the differences in soil properties, alpha diversity (species richness at the sample scale), beta diversity (differences in community composition among samples), and the relative abundances of distinct fungal taxa and guilds among these habitats. Specifically, we asked: (1) Do microhabitats including shrub cover and biocrust types change soil physiochemical properties, fungal diversity, and community composition? (2) Which type of microhabitat shows the strongest influence on soil properties and fungal communities? (3) What are the key environmental factors governing the differences in fungal community composition among microhabitats?

2. Materials and methods

2.1. Study site and soil sampling

The Mu Us Desert, located in Yulin, Shaanxi, China, was formed during the early Tertiary period approximately 360 million years ago. Its formation was a result of continental drift, as the course of the Yellow River shifted southward, depositing sediment in the area. To control desertification, the Chinese government has applied a series of policies, including erecting straw checkerboards, planting xerophytic shrub species, limiting livestock grazing, and establishing artificial sand-fixing forests. Currently, this desert is greening, and heterogeneous landscapes have been formed, with patches of shrub plants and well-developed lichen and moss crusts covering the soil surface.

The sampling sites were located at Heilonggou (110.1 °E, 38.7 °N), Mu Us Desert, Shaanxi, China (Fig. S1). The mean annual temperature here is about 7.8 °C, and the mean annual precipitation is 397 mm (Fick and Hijmans, 2017), mostly occurring from July to September. Soils there are typically the loessal type. Biocrust samples were collected in early September 2021. According to the two types of microhabitats considered, lichen and moss crusts were collected, each in both the shrub(−) and shrub(+) categories (Fig. S1). The biocrusts that can be naturally removed from the soil surface were collected. The biocrust samples were collected in five randomly selected sand dunes, with a distance between any adjacent dunes of more than 100 m to achieve randomization. For each sand dune, a transect with a 2 m width and a 30 m length was pre-established in the interdune area. Soils from different microhabitats were randomly collected along the transect. The samples were collected by spades, which were sterilized by ethanol burning once before sampling. In total, five subsamples were collected in each sampling line and were pooled into one composite sample for each biocrust type, shrub(−) or shrub(+). In total, twenty samples were obtained, including ten samples for lichen crusts and ten samples for moss crusts, each including five samples representing shrub(−) or shrub(+). The samples were separated into two parts: one part was stored at 4 °C for measuring soil properties, and the other part was stored at −20 °C for DNA extraction.

2.2. Measuring soil properties

Soil pH was tested in a soil water suspension (1:5, w/v); soil total organic carbon (C) was quantified by the $K_2Cr_2O_7$–H_2SO_4 oxidation method (Nelson et al., 1996), total nitrogen (N) by the Kjeldahl procedure (Bremner, 1965), total phosphorus (P) by the molybdenum blue method (Murphy and Riley, 1962), and available P by Olsen's method (Olsen, 1954). The soil C:N and C:P ratios were calculated dividing the total organic carbon by total nitrogen and total phosphorus, respectively, to be the proxies of N and P limitation.

2.3. DNA extraction and amplicon sequencing

Soil total DNA was extracted from a 0.5 g sample using the PowerSoil® DNA Isolation kit (Qiagen, Hilden, Germany). The fungal ITS rRNA gene was amplified using primer set gITS7 (5′-GTGARTCATCGARTCTTTG-3′)/ITS4 [5′-TCCTCCGCTTATTGATATGC-3′]) (Ihrmark et al., 2012). The ITS4 primer was linked with a unique 12 bp barcode for each sample. The amplification was performed using a 25 μL mixture containing 1 μL forward and reverse primer, respectively, 1 μL template DNA (10 ng μL^{-1}), 9.5 μL ddH_2O, and 12.5 μL of MasterMix containing Taq DNA Polymerase, PCR Buffer, Mg^{2+}, and dNTPs (Tsingke, China). The amplification was conducted using the following program: denaturation at 94 °C for 5 min, and 35 cycles of 94 °C for 30 s, 56 °C for 30 s, and 68 °C for 45 s, and a final extension at 72 °C for 10 min. To exclude potential contamination, negative controls were performed. Duplicate

PCR reactions were performed per sample and pooled for purification using 1% (w/v) agarose gel electrophoresis in 1.0 × TAE buffer, and the band of about 250 bp was excised and purified using the AxyPrep DNA Gel Extraction Kit (AP-GX-250, Axygen, USA). Quality of the PCR product was assessed by A260/280 and A260/230 ratios using a NanoDrop ND-2000 Spectrophotometer (NanoDrop Technologies Inc., Wilmington, DE). Then, libraries were constructed with equimolar mixed amplicons using a TruSeq DNA kit and sequenced using an Illumina Novaseq platform.

2.4. Bioinformatic analyses

Paired-end reads were merged using the FLASH software (Magoč and Salzberg, 2011). Low-quality sequences were removed, which referred to the sequences with a length <200 bp, more than 2 ambiguous base 'N', and an average base quality score <30. Chimera checking was conducted by Usearch 8 in the *de novo* mode (Edgar et al., 2011). The remaining sequences were processed using the Qiime2 pipeline (Bolyen et al., 2019). The sequences were denoised using the dada2 algorithm, and in total, 795 amplicon sequence variants were obtained. Singletons with total ASVs of less than or equal to 5 reads were removed. All samples were re-sampled to 11326 sequences per sample, referring to the lowest sequence reads of all samples for downstream analyses. The UNITE database (version 8.2) (Nilsson et al., 2019) was used to assign taxonomic information of the ITS sequences. According to the taxonomic information, functional groups of the fungal community were predicted using the FUNGuild database (version 1.1) (Nguyen et al., 2016).

2.5. Statistical analyses

All statistical analyses were performed in R [Version 4.2.2 (R-Core-Team, 2023),]. Fungal richness and taxonomic and phylogenetic diversity were respectively represented by Chao1, Shannon, and Faith Phylogenetic Diversity (Faith PD) indices, which were calculated using the "microeco" package (Liu et al., 2020). To depict differences in community composition of the fungal community in any pair of microhabitats, nonmetric multidimensional scaling (NMDS) analysis and permutational multivariate analysis of variance (PERMANOVA) based on Bray-Curtis dissimilarity were complementarily performed, respectively, using "*metaMDS*" and "*adonis2*" functions of the "*vegan*" package (Oksanen et al., 2023).

Differences in soil properties, fungal alpha diversity, relative abundances of fungal classes, genera, and functional groups among microhabitats were tested by non-parametric Kruskal-Wallis analyses. Relationships between soil properties and fungal diversity indices and relative abundances of dominant fungal taxa were explored using Spearman's correlation with the "psych" package (Revelle, 2022). To disentangle the relationships between fungal beta-diversity and soil properties, partial mantel tests based on Spearman's correlation were performed using the "vegan" package.

3. Results

3.1. Differences in soil physiochemical properties among microhabitats

Whether considering lichen or moss crust individually, soil pH and

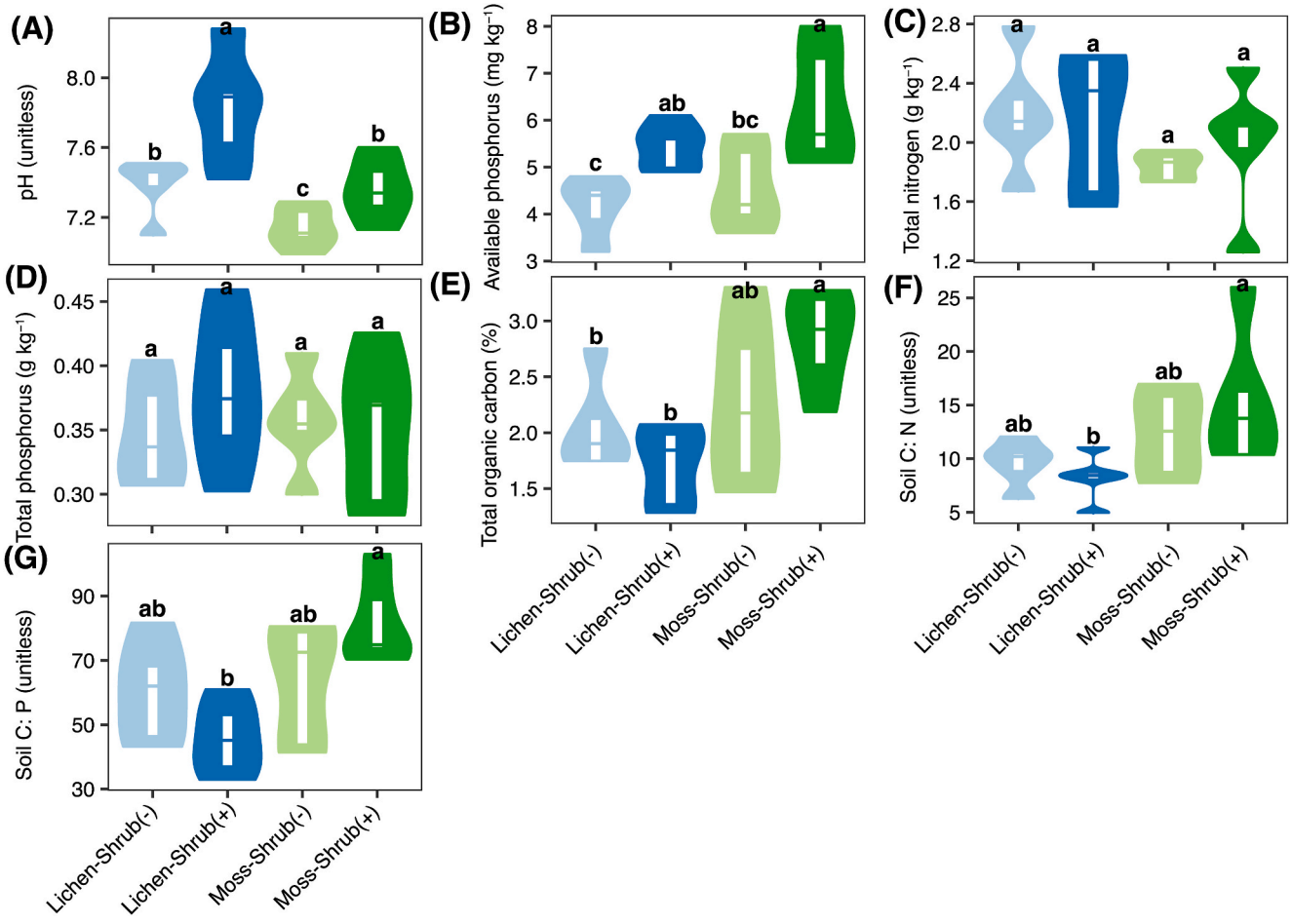

Fig. 1. Soil physiochemical properties among biocrust types (lichen vs. moss crusts) and shrub cover ("-" vs. "+"). Different letters indicate non-parametric Kruskal-Wallis statistical significance at $P < 0.05$.

available P content were significantly higher in microhabitats shrub(+) than shrub(−) (Fig. 1A and B), while the other soil properties were not significantly different between shrub(−) and shrub(+) (Fig. 1C–G). Similarly, whether considering shrub cover individually, soil pH of the moss crust was significantly higher than that of the lichen crust (Fig. 1A), while soil total N, total P, and available P contents were similar among biocrust types (Fig. 1B–D). Moreover, considering shrub (+) individually, soil total organic C, C:N, and C:P were significantly higher in moss than in lichen crusts, but these properties were not significantly different between biocrust types when considering neither shrub(−) nor shrub(+) individually (Fig. 1E–G).

3.2. Soil fungal alpha diversity among microhabitats

Compared to soil properties, less significant differences in fungal alpha diversity among microhabitats were observed. Only fungal Shannon diversity was significantly lower in shrub(−) than shrub(+), considering lichen crust individually, whereas no obvious differences in other fungal alpha diversity indices were observed among microhabitats (Fig. 2).

3.3. Fungal community composition among microhabitats

The NMDS plots showed that beta diversity was more obvious among biocrust types than shrub cover (Fig. 3). Points representing moss crusts were more closely clustered than those of lichen crusts and were distributed mainly at the right side of the NMDS1 axis, with a narrow distribution on the NMDS2 axis. In contrast, those points of lichen crusts were distributed widely in the plots, with wide ranges of distribution on both axes (Fig. 3A). Moreover, the PERMANOVA results suggested that the fungal community composition was not significantly different ($P < 0.1$) between shrub(−) and shrub(+) when considering the lichen crust individually, and was significantly different ($P < 0.05$) in the pairwise comparisons of lichen-shrub(+) vs. moss-shrub(−), moss-shrub(−) vs. lichen-shrub(−), and moss-shrub(+) vs. lichen-shrub(−) (Fig. 3B).

Eurotiomycetes, accounting for more than 75% of the total reads in all samples, was the most dominant fungal class in our study site, followed by Dothideomycetes (0.5%) and Sordariomycetes (0.15%, Table 1 and Fig. S2A). Similarly, *Endocarpon* (20%) and *Knufia* (7%) were the representative fungal genera (Table 2 and Fig. S2B). Considering any of the top ten fungal classes and genera in relative abundances, no significant difference between shrub(−) and shrub(+) was observed (Tables 1–2, Fig. S2), while relative abundances of *Knufia* and *Coniochaeta,* were significantly lower in lichen than in moss crusts.

For fungal functional groups, the results showed that lichenized and pathogenetic fungi were more dominant than saprotrophic fungi (Fig. 4). No significant difference in fungal functional groups was observed between shrub(−) and shrub(+) (Fig. 4A–C), while the relative abundance of lichenized fungi in lichen-shrub(−) was significantly higher than that in moss-shrub(−) and moss-shrub(+) (Fig. 4A). The relative abundance of the pathogenic fungi was significantly higher in moss-shrub(−) than in lichen-shrub(−) and lichen-shrub(+)(Fig. 4B).

3.4. Key factors driving patterns of fungal community composition among microhabitats

Compared to the relative abundances of fungal taxa or functional groups, less numbers of soil properties were significantly correlated with fungal alpha or beta diversity indices (Fig. S3). Only soil total N had marginal ($P < 0.1$) and significant ($P < 0.05$) negative correlations, respectively, with fungal Chao1 and Faith PD indices, while fungal beta diversity was not significantly correlated with any measured soil properties. Further, relative abundances of the *Coniochaeta*, *Knufia*, *Neophaeococcomyces,* and pathogenic fungi had significantly (or marginally) negative and positive correlations, respectively, with soil total N and C:N. Similarly, abundances of lichenized fungi had significantly (or

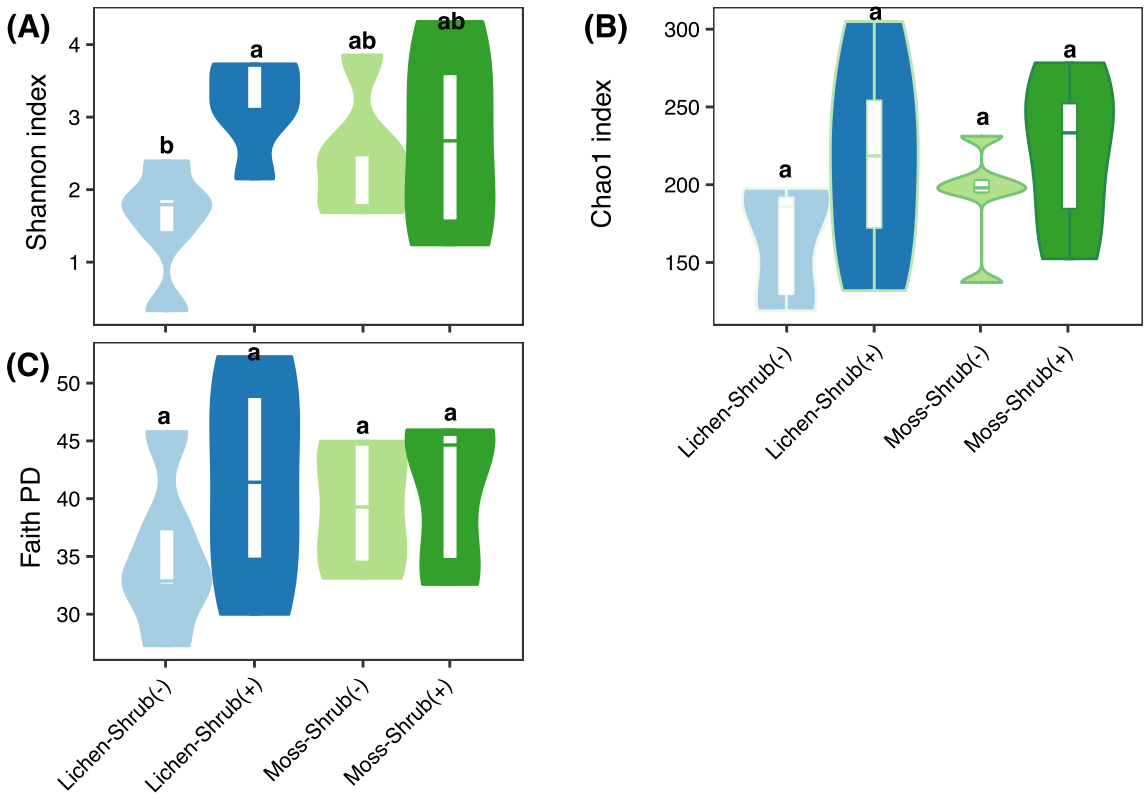

Fig. 2. Fungal alpha diversity among biocrust types (lichen vs. moss crusts) and shrub cover ("-" vs. "+"). Different letters indicate non-parametric Kruskal-Wallis statistical significance at $P < 0.05$.

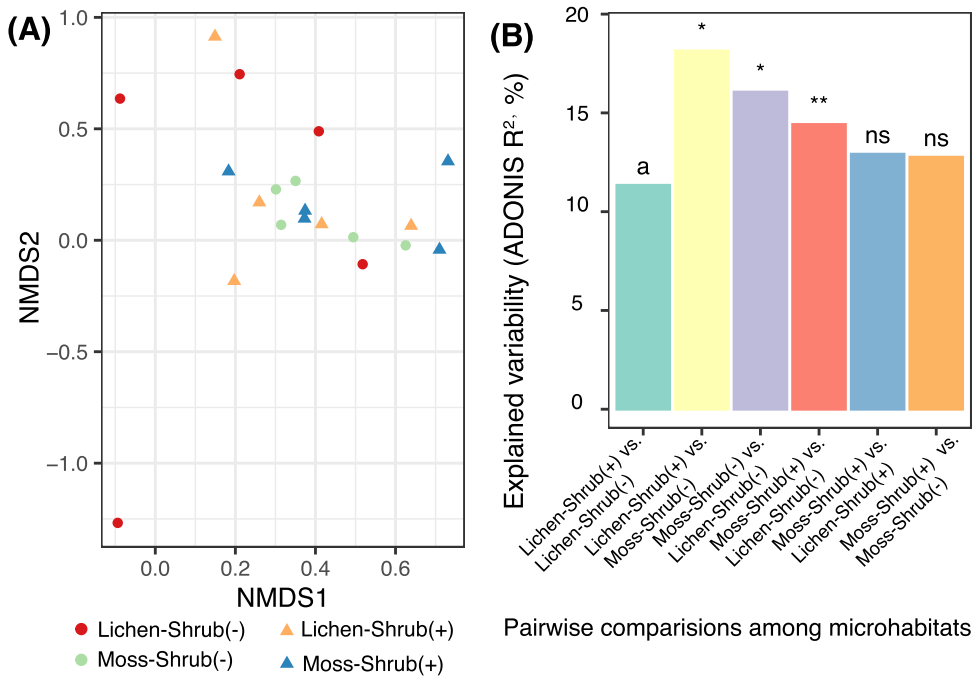

Fig. 3. Variations of fungal community compositions among biocrust types (lichen vs. moss crusts) and shrub cover ("-" vs. "+"). (A) Non-metric multidimensional scaling (NMDS) analysis and (B) Permutational multivariate analysis of variance (PERMANOVA) based on Bray-Curtis dissimilarity matrix. Circles and triangles respectively represent shrub(+) and shrub(−). Significance level, a: $P < 0.1$, *: $P < 0.05$, **: $P < 0.01$, ***: $P < 0.001$, ns: not significant.

Table 1
Relative abundances (%) of fungal classes among biocrust types (lichen vs. moss crusts) and shrub cover ("-" vs. "+").

Classes	Lichen-Shrub(−)	Lichen-Shrub(+)	Moss-Shrub(−)	Moss-Shrub(+)
Leotiomycetes	0.005 ± 0.008a	0.005 ± 0.008a	0.005 ± 0.005a	0.011 ± 0.012a
Lecanoromycetes	0.014 ± 0.019a	0.004 ± 0.005a	0.019 ± 0.017a	0.011 ± 0.015a
Tremellomycetes	0.014 ± 0.013a	0.016 ± 0.015a	0.019 ± 0.019a	0.014 ± 0.008a
Pezizomycetes	0.011 ± 0.010a	0.048 ± 0.047a	0.002 ± 0.004a	0.014 ± 0.010a
Agaricomycetes	0.007 ± 0.007b	0.028 ± 0.029 ab	0.041 ± 0.028a	0.025 ± 0.022 ab
Mucoromycetes	0.055 ± 0.029a	0.042 ± 0.037a	0.049 ± 0.016a	0.058 ± 0.030a
Orbiliomycetes	0.062 ± 0.109a	0.039 ± 0.072a	0.275 ± 0.587a	0.019 ± 0.012a
Sordariomycetes	0.152 ± 0.055a	0.196 ± 0.104a	1.759 ± 3.514a	0.166 ± 0.072a
Dothideomycetes	0.560 ± 0.209a	1.633 ± 1.547a	1.718 ± 1.025a	1.104 ± 0.613a
Eurotiomycetes	91.102 ± 12.2a	75.97 ± 25.01a	93.251 ± 5.536a	87.048 ± 8.835a
Other Fungi	8.019 ± 11.836a	22.018 ± 24.711a	2.861 ± 1.744a	11.531 ± 8.785a

Only the top ten classes in relative abundances ("mean ± sd") are shown. Different lowercase letters indicate significant difference at $P < 0.05$.

Table 2
Relative abundances (%) of fungal genera among biocrust types (lichen vs. moss crusts) and shrub cover ("-" vs. "+").

Genera	Lichen-Shrub(−)	Lichen- Shrub(+)	Moss-Shrub(−)	Moss-Shrub(+)
Ochroconis	0.064 ± 0.034a	0.279 ± 0.334a	0.366 ± 0.5a	0.177 ± 0.158a
Aspergillus	0.302 ± 0.124a	0.275 ± 0.063a	0.371 ± 0.079a	0.302 ± 0.112a
Neophaeococcomyces	0.025 ± 0.017BCE	0.016 ± 0.017c	0.563 ± 0.329a	0.835 ± 1.154 ab
Coniochaeta	0.002 ± 0.004b	0.004 ± 0.008b	1.595 ± 3.501a	0.028 ± 0.033a
Bradymyces	0.049 ± 0.077a	0.117 ± 0.117a	1.466 ± 2.328a	0.191 ± 0.234a
Veronaea	0.055 ± 0.025a	0.058 ± 0.031a	1.695 ± 3.569a	0.085 ± 0.04a
Placidium	39.788 ± 37.769a	17.657 ± 39.166a	0.12 ± 0.054a	0.113 ± 0.048a
Endocarpon	43.23 ± 35.179a	30.554 ± 26.53a	20.652 ± 25.585a	22.123 ± 32.804a
Knufia	7.318 ± 7.184c	27.081 ± 21.261BCE	67.711 ± 25.707a	58.326 ± 37.028a
Other Fungi	9.168 ± 12.112a	23.959 ± 25.041a	5.462 ± 2.586a	17.821 ± 13.553a

Only the top ten genera in relative abundances ("mean ± sd") are shown. Different lowercase letters indicate significant difference at $P < 0.05$.

marginally) positive and negative correlations, respectively, with soil total N and C:N (Fig. 5 and Fig. S3). According to the numbers of soil properties that were significantly correlated with relative abundances of fungal taxa or diversity indices, soil total N and C:N tended to be the key factors driving patterns of fungal community composition among microhabitats (Fig. S3). A summary of how these factors shape soil fungal communities among microhabitats is shown in Fig. 6.

4. Discussion

4.1. Biocrust type is more important than shrub cover in shaping soil fungal communities

In desert ecosystems, microorganisms colonize specific microhabitats to adapt to the stressful conditions of arid environments. They adapt to diverse stressors such as water and nutrient limitations and UV radiation by surviving in various locations, including shrub(−) and shrub(+), on the surface, and inside and underneath different biocrusts (Fierer et al., 2012; Sajjad et al., 2022). Therefore, theoretically, different soil

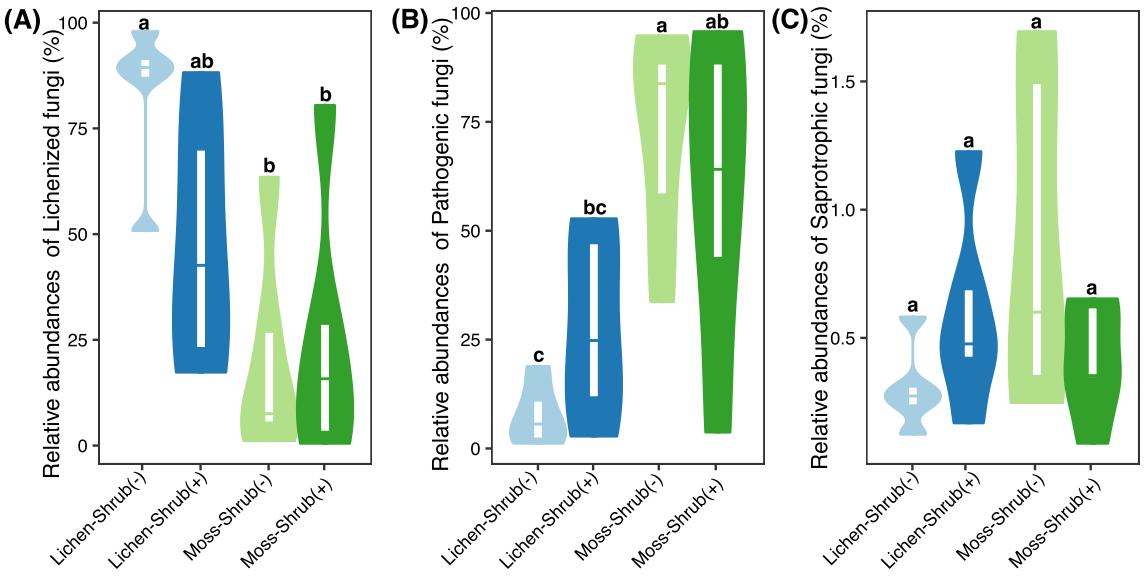

Fig. 4. Relative abundances of fungal guilds among biocrust types (lichen vs. moss crusts) and shrub cover ("-" vs. "+"). Different letters indicate non-parametric Kruskal-Wallis statistical significance at $P < 0.05$.

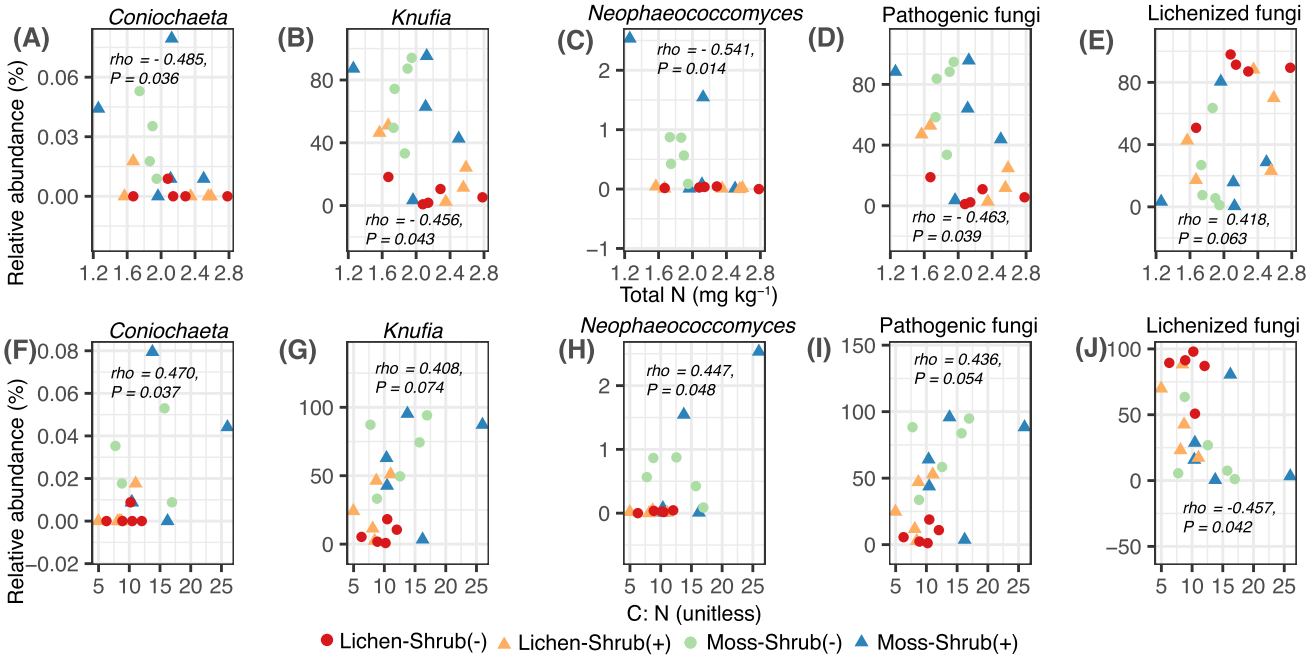

Fig. 5. Scatter plots showing correlations between soil fungal communities and soil total N (A–E), and between soil fungal communities and soil C:N (F–J). Spearman's *rho* and *P* values are indicated.

fungal communities should be observed in microhabitats characterized by different biocrust types and shrub cover. However, we only observed lower fungal Shannon diversity in lichen-shrub(−) than in lichen-shrub (+), while Chao1 and Faith PD were similar among microhabitats. This resembles a previous North American study (Mueller et al., 2015) where lower phylogenetic diversity and similar richness of soil fungal communities in shrub(+) than shrub(−) had been observed, even where the biocrust is not as well developed as in this study. In addition, similar to previous studies conducted in the Gurbantunggut desert (Zhang et al., 2018; Xu et al., 2021b), fungal diversity was not significantly different between lichen and moss crusts.

At the community level, our results generally showed that fungal beta diversity was influenced by the combined effects of biocrust types and shrub cover, with a stronger effects of biocrust types being observed (Fig. 3). Mueller et al. (2015) found that the beta diversity of a fungal community was significantly different between shrub(−) and shrub(+). Complementarily, our results further suggested that when considering lichen crust individually, fungal community composition was distinct between shrub(−) and shrub(+); when considering shrub(−) individually, fungal community composition was different between lichen and moss crusts. Also, when considering neither shrub(+) nor moss crust individually, fungal communities were not statistically different among microhabitats (Fig. 3B). Compared with shrub(+), fungal communities inhabiting shrub(−) may suffer from lower water and nutrient availability, wider temperature ranges, and higher UV radiation; similarly, higher survival stress should also exist in lichen than in moss crusts because of lichens' lower thickness and water-retaining availability. Thus, we inferred that, in arid ecosystems, soil fungal community was

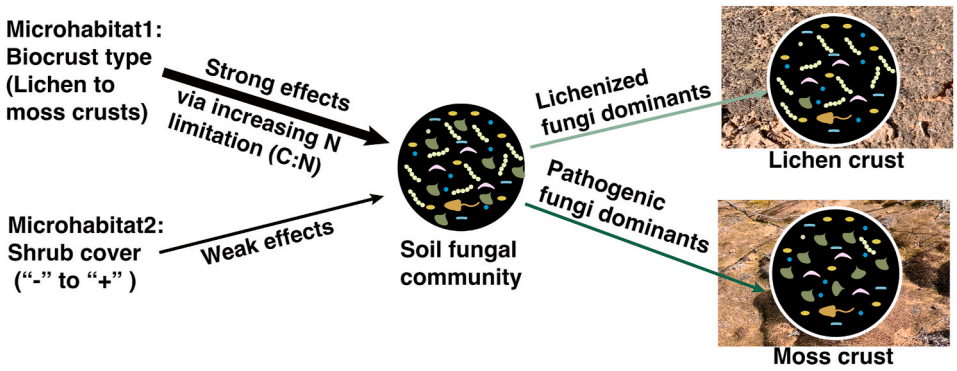

Fig. 6. Conceptual framework illustrating the effects of microhabitat factors on soil fungal community. The biocrust types (lichen vs. moss crusts) exert stronger effects than shrub cover (as indicated by "-" vs. "+") on soil fungal communities. The succession of biocrusts from lichen to moss crusts corresponds with an increasing nitrogen limitation (indicated by the soil C:N ratio), resulting in a shift of fungal community from dominance by lichenized fungi to dominance by pathogenic fungi.

more likely to be influenced by additional factors in more stressful microhabitats.

At specific taxa or guilds level, our results further suggested the stronger effects of biocrust types on relative abundances of specific fungal groups: lichenized fungi were more abundant in lichen than in moss crusts (Fig. 4A). This makes sense for a variety of reasons. Firstly, the dominant fungal taxa observed in this study were in line with those found in global biocrusts, e.g., Eurotiomycetes, Dothideomycetes, and *Endocarpon* (Bates et al., 2012; Egidi et al., 2019; Liu et al., 2021). Secondly, most taxa affiliated with Eurotiomycetes or Dothideomycetes, e.g., *Endocarpon* and *Placidium*, are lichenized fungi (Esslinger, 2021) that form symbioses with photobionts such as cyanobacteria or eukaryotic microalgae. Lichenized fungi can absorb water and nutrients and protect photobionts by filaments, and photobionts can provide organic compounds (e.g., extracellular saccharides) to lichenized fungi (Lange and Belnap, 2016; Rosentreter et al., 2016). Moreover, the more abundant *Knufia* and *Coniochaeta* in moss than in lichen crusts could be explained because these species were typically plant parasites or pathogens (Damm et al., 2010; Tedersoo et al., 2014; Irinyi et al., 2015). In microhabitats created by moss crusts, the presence of large amounts of mosses (as they are plants) may foster these species from these two genera and increase their abundances. Moreover, similar to Mueller et al. (2015), we did not observe relative abundances of any fungal taxa to be significantly different between shrub(−) and shrub(+), further suggesting the more essential role of biocrust type than the presence or absence of shrub cover in shaping soil fungal communities.

4.2. Soil total nitrogen and C:N are the key determinants of fungal community composition patterns among microhabitats

Nitrogen is the most important factor limiting primary production worldwide (Vitousek et al., 2002). Our results showed that soil total N and C:N were the key factors driving patterns of soil fungal community composition among microhabitats. These results aligned with those of prior studies that were conducted on large spatial scales (Hu et al., 2019; Chen et al., 2021) and at a single site in arid or semi-arid environments (Nie et al., 2018). In correspondence with our results (Fig. S3), Negative correlations of fungal diversity and soil nitrogen have been reported in deserts ecosystems (Wang et al., 2018; Zhang et al., 2022). This suggests that high levels of nitrogen in soil may inhibit the diversity of fungal communities. The mechanisms driving this negative correlation are likely complex, but could probably be the direct and indirect effects of nitrogen on the relative abundances of certain fungal groups e.g., the lichenized fungi. This is supported by our data where the abundances of lichenized fungi were positively and negatively correlated with soil total N and C:N, respectively (Fig. 5). This could potentially be explained by the fact that the potential nitrogen fixation rate is higher in lichen than in moss crusts (Xu et al., 2021b). The higher potential nitrogen fixation rate may lead to higher nitrogen availability and less nitrogen limitation (e.g., lower values of the C:N) in lichen than in moss crusts, which may promote the synthesis of lichen biomass. Moreover, with the development of biocrust, the synthesis of a large amount of moss biomass may compete for soil nitrogen resources with lichen species, overconsuming nitrogen nutrients in soil and hence leading to the higher nitrogen limitation in moss than in lichen crusts.

This study simultaneously explored the effects of shrub cover and biocrust types in structuring soil fungal communities. We observed that shrub cover and biocrust types were all effective, but biocrust types were more important in altering soil properties and fungal community composition. Compared to lichen crust, the higher N limitation in moss crust promoted pathogenic fungi and inhibited lichenized fungi in their relative abundance (Fig. 6). These results have significant implications for desertification control in the Mu Us Desert. Firstly, the greater importance of biocrust types compared to shrub cover suggests that conservation and restoration efforts should prioritize biocrusts rather than solely focusing on increasing shrub cover. Additionally, it is of great importance to take into account the dynamics of biocrust succession in desertification management strategies, considering their considerable influence on soil fungal communities. However, these findings are based solely on our observation in the Mu Us desert; whether the same or similar results would be found in global drylands needs to be further verified by future large-scale studies.

Data availability

Data will be made available on request.

CRediT authorship contribution statement

Lin Xu: Writing – review & editing, Writing – original draft, Visualization, Validation, Software, Project administration, Methodology, Investigation, Funding acquisition, Formal analysis, Data curation, Conceptualization. **Chaonan Li:** Software, Methodology, Data curation. **Wenjun Xiong:** Investigation. **YongPing Kou:** Writing – review & editing, Validation, Resources, Investigation. **Ping Zou:** Investigation. **Bingjie Jiao:** Investigation. **Minjie Yao:** Writing – review & editing, Methodology, Investigation, Conceptualization. **Junming Wang:** Writing – review & editing, Supervision. **Bingchang Zhang:** Investigation. **Xiangzhen Li:** Writing – review & editing, Validation, Software, Methodology, Conceptualization.

Declaration of competing interest

The authors declare that they have no known competing financial

interests or personal relationships that could have appeared to influence the work reported in this paper.

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Appendix A. Supplementary data

Supplementary data to this article can be found online at https://doi.org/10.1016/j.funeco.2024.101352.

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