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SOIL BIOLOGY

# Effects of Alpine Wetland Degradation on Soil Microbial Structure and Diversity on the Qinghai Tibet Plateau

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**Abstract**—The characteristics of microbial structure in different soil degradation stages caused by the drought processes from wetland to meadow on the Qinghai Tibet Plateau were analyzed. The composition and diversity of soil bacteria and fungi were also analyzed by using high-throughput sequencing technology. The results showed that Proteobacteria was the highest abundance among bacteria, and Ascomycota was the highest among fungi. The degradation transition from alpine wetland to alpine meadow had insignificantly affected on the dominant bacteria, but had significantly affected on Gracilibacteriae and Ignavibacteriae bacterial phyla ($P < 0.05$), which characterized low abundance. The relative abundance of the dominant, fungal phyla Mortierellomycota ($P < 0.05$) significantly increased in soils along the degradation gradient. There was no significant difference between soil bacteria and fungi for Alpha diversity in different soil degradation stages. Beta diversity was found to be significant difference in soil bacterial structure for alpine swamp wetland and alpine meadow. Soil pH, water content, total organic carbon (TOC), total nitrogen (TN) decreased significantly ($P < 0.05$) with the degradation stages. RDA analysis showed that TN and TOC achieved the highest effect on the bacteria number expressed as operational taxonomic units and their Shannon index, moreover soil water content significantly affected on fungi number and Shannon index. TN and bacterial number had a significant positive correlation ($P < 0.05$). The relative abundance of Gracilibacteriae, Ignavibacteriae and Elusimicrobiae, which are beneficial to soil C and N contents and that Gemmatimonadetes are beneficial to N fixation, decreased in the drought processes of alpine wetland. This result could increase the relative abundance of the fungi phylum of Mortierellomycota, and might decrease the soil microbial diversity.

**Keywords:** alpine swamp wetland, high-throughput sequencing, soil microbial diversity

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## INTRODUCTION

The degradation of alpine swamp wetland on the Qinghai-Tibet Plateau becomes increasingly serious problem [10]. This degradation may cause the changes of soil nutrient status and microbial structure which play a vital role in the ecological functions of alpine wetland [33]. A lot of research has been conducted on the effects of alpine swamp wetland degradation on soil microbial structure and soil nutrients [2, 14, 17, 18, 36]. Soil microorganisms are the key of all ecological functions in terrestrial ecosystems, therefore, the study on microbial diversity in soils of different geographic localizations will contribute to the preservation of their ecological balance [24, 31]. The soil types and nutrients will be changed due to the degradation of alpine swamp wetland, so as to affect the microbial diversity of alpine wetland [5, 27]. Soil bacteria and fungi are the two important components of soil microbial communities, but the soil bacteria have more diversity and larger quantity than soil fungi [22]. It also noted that the area of alpine wetland was shrinking, and its degradation succession was occurred, therefore the bacterial diversity generally decreased. Thus, the degradation of alpine wetland will lead to a decrease in soil bacterial diversity [27]. It had also been reported that with the succession of alpine wetland to alpine meadow, the diversity of soil bacteria increased gradually [15]. At present, there is great uncertainty about the researches on soil bacterial diversity in different stages of alpine wetland degradation. Some researches already showed that soil fungi compared to bacteria were more sensitive to the changes of soil nutrients and other environmental factors [29], and there are only a few studies on the diversity of soil fungi in different degradation stages of alpine swamp wetlands. Studying the effects of alpine wetland degradation on soil bacterial and fungal diversities will be a great scientific significance. In this paper, non-degradation, light

degradation and severe degradation in alpine swamp wetlands are selected to explore the difference of soil microbial community structure, diversity and edaphic factors in the different degradation stages. The 16S rDNA of soil bacteria and internal transcribed spacer (ITS) gene of fungi were sequenced by high-throughput sequencing. The following two scientific questions will be addressed: (1) What microbial community structure in alpine swamp wetlands and how their changes in degradation succession sequence would be happened. (2) What is the relationship between soil properties and soil microbial community in different degradation successions. With the above two questions, we try to reveal the responses of soil bacterial and fungal diversity to alpine wetland degradation, and to provide a scientific theoretical support for the protection and restoration in alpine wetlands.

## MATERIALS AND METHODOLOGY

### *Research Areas*

The sample site is located at Dawu Town, Maqin County, Guoluo Tibetan Autonomous Prefecture, Qinghai Province, west China (34°27′48″ N, 100°12′49″ E, with an average altitude of 3730 m). Meteorological characteristics in the study areas are displayed with continental cold humid climate, mean annual temperature of –3.9°C, and mean annual precipitation of 528.8 mm. The resources of alpine wetland are rich and grazing time is in winter season. Plant communities are mainly composed of Cyperaceae, Gramineae and other families. Because of the aggravation of the drought in global warming, the number of the freeze-thaw mound in alpine swamp wetland increase, but their sizes decrease conspicuously. Then the degradation successions of drought processes from wetland to meadow will be occurred. The research site is 4 $hm^2$. The composition of dominant species, coverage of dominant species, average height of vegetation, amount of freeze-thaw mounds and their sizes in alpine swamp wetland are detailed in the literature [13].

### *Sample Plot Selection and Experimental Treatment*

The alpine swamp wetland stage one (Non-degradation, ND, Fig. S1a), stage two (Light degradation, LD, Fig. S1b) and stage three (Severe degradation, SD, Fig. S1c) were selected based on the three stages of the drought processes in alpine swamp wetland which was dominated by *Kobresia tibetica*. Vegetation coverage and amount of freeze-thaw mounds were surveyed. Radially sampling lines, three in total, were randomly selected starting from alpine wetland center as shown in Fig. S1d. Six sampling points were chosen by an interval of 25 m with two points at each wetland type on one line (Fig. S1d).

### *Sample Analyses*

**Soil sampling.** According to five-point sampling method, five quadrats of 10 × 10 cm were selected in 6 sampling spots in different degradation stages of alpine swamp wetland in August 2019. Sampling time is the vigorous growth period of herbage. After alcohol disinfection of sampling tools, soil layer at 0–30 cm was taken and mixed, and other impurities were removed. A certain amount of soil was sampled into a sterile tube. Each sample was 10 g and put quickly into a liquid nitrogen tank for frozen storage (eighteen samples). In laboratory, it was sealed and transported with dry ice. The remaining soil samples delivered to the laboratory for soil nutrients determination.

**Analysis of soil physical and chemical properties.** Soil moisture was measured by TDR350 (soil moisture, temperature and conductivity tester) produced by Spectrum Company (U.S.). The contents of total nitrogen (TN), total phosphorus (TP) and available phosphorus (AP), ammonium nitrogen ($NH_4^+$-N), nitrate nitrogen ($NO_3^-$-N) were determined by AA3 Continuous Flow Analyzer from SEAL company (Germany), and organic carbon content (TOC) was determined by external oxidation heating method of potassium dichromate – concentrated sulfuric acid solution. Soil pH was determined by pHS-3C pH meter (soil : water = 1 : 2.5).

**Analysis of microbial diversity and community structure.** Beijing Ovison Gene Technology Co., Ltd. was used to determine the gene sequence of 16S rDNA gene V3+V4 region of soil bacteria (the general primer sequences are 336F: 5'-GTACTCCTACGGGAGGCAGCA-3' and 806R: 5'-GTGGACTACHVGGGTWTCTAAT-3') and ITS1 rDNA gene ITS1 and ITS2 regions of soil fungi (the general primer sequences are ITS1F: 5'-CTTGGTCATTTAGAGGAAGTAA-3' and ITS2R: 5'-TGCGTTCTTCATCGATGC-3') by high-throughput sequencing. The assay steps were from genomic DNA extraction, genomic DNA quality inspection, PCR amplification, PCR products electrophoresis detection, PCR products purification, Miseq library construction, Miseq library quality inspection and then to Illumina Miseq platform sequencing.

The original sequence data was firstly conducted, and then the optimal sequence was obtained by sequence splicing, filtering and chimerism removal. The OTU clustering and annotation were conducted. Uclust [21] was used to analyze the biological information of OTUs at 97% similarity level, then alpha diversity and beta diversity were analyzed based on the clustering results; Silva [19] (Release128/132 http: www.arb-silva.de) database was used to annotated the OTUs of bacteria, and Unite [1] (Release 7.2 http://unite.ut.ee/index.php) fungi database was used to annotated the OTUs of fungi. Based on the annotation results, the classification information of each classifi-

cation was obtained, and the correlation analysis of sample composition and community difference of results among samples were conducted.

Alpha diversity refers to the number of species in a population and the number and distribution of each species. For estimating species abundance and diversity of microbial communities, alpha diversity analysis will include the following statistical analysis indexes:

(1) Chao1: species richness index. Used to estimate the number of OTUs in the community, and the calculation formula is as follows:

$$\text{Chaol} = \text{Obs} + \text{n1}(\text{n1}-1)/2(\text{n2}+1). \quad (1)$$

Where Obs is the observed amount of OTUs, n1 is the amount of OTUs with one sequence, and n2 is the amount of OTUs with two sequences.

(2) Goods Coverage: observed depth. The calculation formula is as follows:

$$C = 1 - \text{n1}/N. \quad (2)$$

Where n1 is the amount of OTUs with one sequence, and N is the total amount of OTUs of the sample. The index represents the coverage rate of each sample library. The higher value is, the higher the probability of the detected sequence in the sample will be, and then the lower the probability of the sequence not detected will be. Whether the sequence results represent the real conditions of microorganisms of the sample is reflected by the index.

(3) Observed Species: the actually amount of observed OTUs with the increase of sequencing depth.

(4) Shannon index (H): used to estimate the microbial diversity. The calculation formula is as follows:

$$H = -\sum \text{Pi}\ (\ln \text{Pi}). \quad (3)$$

Where Pi is the proportion of individuals belonging to its kind in the sample, Pi = Ni/N. Ni is the amount of individuals of species i, and N is the total amount of individuals of all species. The more evenly distribution of the individuals is, the larger H will be. And the higher the Shannon value is, the higher the community diversity will be.

(5) Simpson index (1-D): used to estimate the microbial diversity, and the result is delivered as 1-D value. The formula is as follows:

$$1\text{-}D = 1 - \sum(\text{Ni}/N)^2. \quad (4)$$

Simpson index gives attention to both richness and uniformity, in which D value reflects the probability that two individuals are randomly selected from the same sample and the same class. The D value is the total number of individuals found in a specific species (Ni) divided by the total number of individuals found (N).

The value of D is between 0 and 1, where 0 means infinite diversity and 1 means no diversity. Therefore, the greater the 1-D value, the higher the community diversity.

Beta diversity is to compare the difference in communities from another angle. The differences of soil microbial community composition are focused on by Beta diversity analysis. Non metric multidimensional scaling (NMDS) [20] is a data analysis method that simplifies the research objects (samples or variables) in multidimensional space to low-dimensional space to be positioned, analyzed and classified. The original relationship in objects can be remained.

### *Data Analysis and Mapping*

The analysis platform of this research was Qiime platform, and the data calculation and mapping of the structure composition of soil microbial community and Beta diversity (NMDS) were completed in own script R of Qiime. One-way ANOVA and Duncan were conducted by SPSS Statistics 20.0 to analyze the diversity and abundance of soil microbial communities and the differences of soil physical and chemical properties in different degradation stages of alpine swamp wetland. The differences of bacterial and fungal phylum between non-degradation and severe degradation were analyzed by Metastats significant difference analysis. The correlations between soil physical and chemical indexes, OTU amount and Shannon diversity were tested by Pearson correlation coefficient method. The Detrended Correspondence analysis (DCA) was conducted to the number of OTU and Shannon diversity matrix of soil samples by Canoco for Windows 4.5. Canonical correspondence analysis (CCA) or Redundancy analysis (RDA) were selected according to the gradient length of the first axis in DCA results. In the analysis, if the size of the first axis of Lengths of gradient is greater than 4.0, CCA should be selected. If it is from 3.0 to 4.0, both RDA and CCA can be selected. If it is less than 3.0, the result of RDA is better than CCA, and then to analyze the effects of soil physical and chemical properties on the amount of OTU and Shannon diversity of soil microbial community. Cano Draw for Windows was used for mapping.

## RESULTS AND ANALYSIS

### *Composition of Soil Microbial Community*

**Composition of bacterial community.** A total of 44 identified species were detected in the 18 soil samples at the bacterial categorical level, as shown in Fig. S2, the horizontal distribution groups of bacteria in different degradation stages of alpine swamp wetland were mainly focused on Proteobacteria, Acidobacteria, Actinobacteria, Chloroflexi, Nitrospirae and Gemmatimonadetes. Among them, Proteobacteria obtained the highest relative abundance value in soil bacteria, and the relative abundance values of the other front bacterial phyla from high to low were Acidobacteria, Actinobacteria and Chloroflexi (relative abundance >7%). In the three soil degradation stages, the average relative abundance of Proteobacteria was 38.7% (ND),

33.7% (LD), 31.2% (SD), respectively. Acidobacteria was 21.3% (ND), 27.7% (LD), 28.0% (SD), Actinobacteria was 13.8% (ND), 13.8% (LD), 15.4% (SD), and that of Chloroflexi was 8.4% (ND) and 7.3% (LD), 7.9% (SD), respectively. In the dominant soil bacterial phyla, the abundance of Proteobacteria decreased, and Acidobacteria increased. Actinobacteria slightly first increased and then decreased, and the relative abundance of Chloroflexi decreased first and then increased. After the significant difference analysis among Meatstats groups, the effect of alpine swamp wetland degradation on the relative abundance of dominant bacterial phyla was not significant. The alpine wetland was developed into alpine meadow, therefore the relative abundance of Gracilibacteria, Ignavibacteriae, TM6_Dependentiae, Nitrospinae and Elusimicrobia and other phyla decreased significantly by 0.06, 0.21, 0.02, 0.01, and 0.10%, respectively ($P < 0.05$), while the relative abundance of Deinococcus-Thermus, Gemmatimonadetes and Firmicutes obviously increased by 0.02, 2.84 and 1.25%, respectively ($P < 0.05$) (Table S1).

A total of 371 groups were detected at the categorical level of bacterial genus. 51 groups, whose average relative abundance were more than 0.1%, had been identified in three degradation stages. They accounted for 31.4% of the total relative abundance, and the genera with more than 64.1% of the total relative abundance had not been identified (Fig. S3).

**Composition of soil fungal community.** According to the horizontal distribution histogram of Mycota (Fig. S4), a total of 18 identified fungal groups were obtained. Five phyla, Ascomycota, Mortierellomycota, Basidiomycota, Rozellomycota and Glomerycota, were identified with having an average relative abundance more than 1%. In the dominant fungal phyla, the relative abundance of Ascomycota was the highest, which is more than 45.0% among ND, LD and SD degradation stages. Comparing with non-degradation, the relative abundance of Ascomycota in light degradation decreased by 2.56%, and that in severe degradation increased by 1.95%. Followed by Mortierellomycota, whose the average relative abundance was 13.4, 19.2 and 31.3% in ND, LD and SD degradation stages, respectively. The average relative abundance of Basidiomycota was 15.2, 6.9 and 10.1% in ND, LD and SD, respectively, and meanwhile there existed unclassified fungi. The relative abundance of Ascomycota and Basidiomycota decreased first and then increased, and the relative abundance of Mortierellomycota increased. According to the significant difference analysis among Meatstats groups, the relative abundance of dominant fungal Mortierellomycota ($P < 0.05$) significantly increased with the degradation of alpine swamp wetland. At the same time, the relative abundance of some fungi with low abundance such as Rozellomycota, Chytridiomycota and Kickxellomycota, was obviously reduced by 5.66, 0.67 and 0.23%, respectively ($P < 0.05$) (Table S2). It can be inferred from the horizontal column chart of eubacterium (Fig. S5) that a total of 357 fungal groups have been obtained, of which only 31 had been identified with an average relative abundance of more than 0.1%. *Mortierella* had a relative abundance of more than 10.0% in ND, LD and SD soil fungi, which belongs to Mortierellomycota phyla. The relative abundance of *Mortierella* significantly increased by the degradation of alpine swamp wetland by 17.9% ($P < 0.05$). At the level of fungi phyla, there existed more than 42.0% of the total relative abundance was not identified.

### *Analysis of Soil Microbial Diversity*

It can be inferred from Table S3 that the sequencing coverage rates of bacteria and fungi in the three degradation stages were all greater than 0.96 and 0.99, respectively, indicating that the probability of the detected bacterial and fungal sequences in the sequenced soil samples is low, and the real existence of bacteria and fungi in the soil can be reflected by the sequencing results. The results showed that the Chao1 and Shannon diversity indexes of soil bacteria were consistent with the amount of observed OTU, which showed that the Chao1 and Shannon index decreased with the degradation of alpine swamp wetland. Among which, Chao1 and Shannon indices corresponding to ND were the largest and were 2758.49 and 9.21, respectively. The SD was the lowest, which were 2561.98 and 8.94, respectively. According to the results of soil fungal abundance and diversity, the average Chao1 index, the average amount of observed OTUs, the average Shannon index and the average Simpson index of soil fungi showed a trend of increasing firstly and then decreasing. The Chao1 index (642.13), the amount of observed OTUs (485.00), Shannon index (4.96) and Simpson index (0.87) of LD were all the largest. The results of variance analysis showed that the Alpha diversities of soil bacteria and fungi had no significant difference in different degradation stages.

According to the results of NMDS analysis (Fig. S6) at the bacterial level, the distance among ND, LD and SD was large, and there was intersection between ND and LD, but the overlap is too narrow, which indicates that the degradation of alpine wetland into alpine meadow had changed the bacterial community structure of underground soil. At the fungal level, ND, LD and SD groups intersected in pairs, indicating that the alpine wetland had developed into an alpine meadow caused by drought, and there was no difference in the fungal community structure of underground soil.

Note: if the soils in the same group are in a circle, it means that the difference between groups is not obvious, and the non-intersection of circles between groups means that there is a certain difference between the groups. a) NMDS analysis of bacterial OTU level, b) NMDS analysis of fungal OTU level.

### *Soil Physical and Chemical Properties*

The soil physical and chemical properties of 0–30 cm in ND, LD and SD were conspicuously different. With the degradation of alpine swamp wetland, the soil moisture (M), TOC and TN of which showed a downward trend, while soil pH, TP, available nitrogen and AP all increased firstly and then decreased (Table S4). The results of one-way ANOVA showed that comparing with ND, soil pH of SD decreased significantly by 0.37% ($P < 0.05$), and M of LD and SD decreased by 4.68 and 7.45%, respectively ($P < 0.05$). The TOC and TN of LD and SD were lower than those of ND. The TOC decreased by 19.80 and 58.05 g $kg^{-1}$, respectively ($P < 0.05$), and TN decreased by 2.35 and 4.14 g $kg^{-1}$ ($P < 0.05$). The TP and AP showed as: LD > ND > SD, and the $NH_4^+$-N and $NO_3^-$-N showed as: LD > SD > ND. Generally, alpine wetland degrading into alpine meadow had a huge effect on soil physical and chemical indexes especially the obvious decrease of soil pH, M, TOC, TN and TP ($P < 0.05$).

### *Relationship between Soil Microbial Diversity and Physical and Chemical Properties*

According to OTUs amount of bacteria and fungi and Shannon diversity index matrix, the value of gradient length on the first axis in DCA results was 0.128, and RDA was selected in this paper. Figure S7 shows the RDA sequencing results of the amount of bacterial OTUs, Shannon diversity index and soil physical and chemical properties of 0–30 cm. 33.21 and 33.48% of the total species variables were explained by the first and second order axes respectively. The interpretation rate of each soil physical and chemical properties in Simple Effects showed that TN and TOC accounted for 22.5 and 18.0% of the total physical and chemical indexes respectively, which had the highest impact on the amount of bacterial OTUs and Shannon index. pH, M, TOC, TN, TP, $NH_4^+$-N, $NO_3^-$-N, AP and the first order axis were positively correlated. Pearson correlation coefficient method showed that total nitrogen had the greatest impact on the amount of OTUs ($R = 0.523$, $P < 0.05$) and Shannon diversity ($R = 0.419$, $P > 0.05$), indicating that bacterial number and diversity were sensitive to TN (Fig. S9). Figure S8 shows the RDA ordination results of the amount of fungal OTUs and Shannon diversity index and soil physical and chemical properties of 0–30 cm. 41.39 and 45.51% of the total species variables were explained by the first and second order axes respectively. The M and pH were the most important properties according to simple effects, accounting for 8.6 and 6.0%, respectively. M, pH and TN were negatively correlated with the amount of OTUs and Shannon diversity. The other five properties were positively correlated with the amount of OTUs and Shannon diversity. The impact of physical and chemical indicators on fungal community shown in Pearson results was not significant, indicating that the sensitivity of the amount and diversity of fungi to soil physical and chemical properties was low. At the meanwhile, Pearson correlation analysis showed that there was a significant positive correlation between the amount of OTUs and Shannon diversity ($P < 0.01$) (Fig. S9).

## DISCUSSION

Bacteria obtains the overwhelming proportion in the total amount of microorganisms in Xiaopohu wetland, Qinghai Lake basin [30]. Similarly, the number of soil bacteria in Dawutan alpine swamp wetland and the different degradation stages in Maqin County of Guoluo Prefecture was greater than fungi. The composition of soil bacterial phyla includes Proteobacteria, Acidobacteria, Actinomycetes and Chloroflexi. And there is little difference of the horizontal composition of soil dominant bacteria among light degradation, severe degradation and non-degradation, which indicating that the degradation successions of alpine swamp wetland to alpine meadow, and the soil bacterial specificity of different degradation degrees decrease. The relative abundance of Proteobacteria and Acidobacteria was the highest in non-degradation, light degradation and severe degradation. This is consistent with the research results of Lu [15]. Among the dominant bacteria, Proteobacteria is widely distributed in different environments, and therefore it is considered to be related to carbon utilization. Acidobacteria has a great correlation with soil nutrition, and the relative abundance of Acidobacteria in poor soil is high [4, 26]. In this paper, we found that although there was no significant difference between Proteus and Acidobacteria in the degradation process of alpine swamp wetland, the abundance of Proteobacteria and Acidobacteria continued to decline, indicating that the degradation of alpine swamp wetland might lead to the decline of carbon utilization efficiency of wetland and the soil would become barren. During the degradation succession from alpine wetland into alpine meadow, the abundance decreased, and the bacterial flora with relatively large abundance were Gracilibecteria, Ignavibacteriae and Elusimicrobia. Among them, carbon and nitrogen metabolism balance of wetland vegetation growth could be promoted by Gracilibecteria [7], and nutrient release might be directly or indirectly promoted by Ignavibacteriae phyla through degradation of organic matter or formation of anoxic environment [6]. Elusimicrobia phyla is closely related to nitrogen fixing bacteria [39]. Gemmatimonadetes obtain significantly increasing and relatively larger abundance, which has strong denitrification function, which will reduce the content of nitrogen in soil [8]. For soil fungal communities in alpine swamp wetland, Ascomycota, Mortierellomycota, Basidiomycota are the main dominant fungi. The degradation of alpine swamp wetlands will significantly increase the relative abundance of dominant fungi Mortierello-

mycota. Some studies have shown that Mortierellomycota can dissolve the phosphorus which is hard to use in soil, and is inferred that it has more growth advantages in low phosphorus soil [16]. Under anaerobic reduction condition, the alpine swamp wetland has high soil carbon, nitrogen, phosphorus and other nutrients because of the accumulation of organic nutrients, providing abundant nutrition for vegetation and microorganisms. With the degradation of wetland into meadow, the soil moisture content decreased and the soil aeration greatly improved, which is more suitable for the reproduction and growth of aerobic microorganisms such as Acidobacteria and Gemmatimonadetes [3, 15, 35]. At the meanwhile, content of soil water is the key factor affecting the soil carbon and nitrogen cycle through affecting soil microbial activities, especially carbon and nitrogen cycle, and the soil organic carbon and total nitrogen will be influenced, which resulting in the decline of the abundance for Graciliberia, Ignavibacteriae, Elusimicrobia and other bacteria involved in the balance of carbon and nitrogen metabolism.

Diversity index is an effective method to analyze the structure of soil microbial community. If the diversity index is higher, the abundance and diversity of microbial community will be higher [12], the relationships between soil microorganism and their physical and chemical environments will be more complex, and the grassland ecosystem will be more stable [37]. Soil microbial diversity is often affected by the interaction of vegetation and soil factors. The analysis of the correlation between the amount and diversity of microorganisms and environmental factors promotes to understand the interaction, inter-impact and formation process of various organisms in the ecosystem. It was found by Lu that the soil water content, soil organic matter and total nitrogen decreased in the process of wetland degradation, and the total phosphorus was ranked as: swamp meadow = wetland > meadow. The diversity of soil bacteria and fungi increased with the degradation of wetland [15]. The results of the research compared with that showed that the change of wetland degradation nutrients was similar, but the change trend of microbial diversity was different. The details of the results are that the soil water content, soil organic matter and total nitrogen decreased significantly, and the total phosphorus was fanked as: light degradation > non-degradation > severe degradation. Although the change of the diversity of bacteria and fungi in degraded wetland soil was not obvious, the quantitative change trend was that the diversity of soil bacteria decreased gradually, and the diversity of fungi increased firstly and then decreased. The results of RDA analysis showed that the total nitrogen content and organic carbon content had the highest impact on the amount and diversity of soil bacteria. The change of bacterial diversity may be due to the requirement for soil from the survival and reproduction of microorganisms to provide nutrients, and the period with high soil nutrient content is the vigorous growth period of vegetation community. Besides, most of the soil nutrients are absorbed by plants, thus the nutrients for microbial growth are less [25]. The amount and diversity of fungi were more negatively affected by soil moisture. When the wetland degenerated into swamp meadow, soil moisture decreased significantly. Therefore, the number of soil fungi increased in swamp meadow, due to the decrease of soil moisture and the increase of soil aeration. Pearson correlation analysis showed that except for the significant correlation between the amount of soil bacteria and the total nitrogen content, the correlation between the amount of soil bacteria and other soil physical and chemical properties was not conspicuous, so as to the difference between Shannon index of soil bacteria and soil physical and chemical properties. There was no obvious correlation between the amount of soil fungi and Shannon index and soil physical and chemical characteristics. The degraded grassland of Leymus chinensis grassland in Hulunbeier was studied by Shang et al. [23] and the results showed that available nitrogen, available potassium, organic matter and total potassium were the main driving factors to bacterial community. In addition, it was found by Zhao et al. [38]. that Shannon index of soil fungi had no obvious correlation with soil physical and chemical properties and plant community functional properties. The results of Yu et al. [34] showed that the bacterial community diversity was more affected by soil environmental factors, and pH and water content had conspicuous effects on bacterial and fungal community diversity of saline alkali soil in Ancient lake wetland. The difference between the results of this paper and previous studies may be the differences among regional climate, soil nutrients, vegetation community structure, and the amount and type of root exudates. For instance, the studies have shown that soil microbial diversity is affected by aboveground vegetation through influencing soil organic carbon, organic nitrogen, soil moisture, temperature, aeration and pH. If the plant community diversity is richer, the composition of litter and root exudates will be more abundant, and the soil microbial diversity will be higher [28]. However, the degradation of alpine wetland will result in conspicuous changes in the composition of plant community and the proportion of different plant species [11, 32], which also affecting the structure and diversity of soil microbial community.

To sum up, the aridity of alpine swamp wetland has different impacts on soil nutrients, soil microbial community structure and diversity, which resulting in the obvious decrease of soil total organic carbon, total nitrogen and total phosphorus. In the soil bacterial, the Gracilibecteria phyla involved in carbon and nitrogen metabolism, the Ignavibacteriae family promotes nutrient release, the Elusimicrobia phyla is closely related to nitrogen fixing bacteria are relatively abundant, and the relative abundance of Gemmati-

monadetes significantly increased which is not conducive to nitrogen fixation and obtains strong denitrification function. The relative abundance of Mortierellomycota significantly increased in soil fungal community, which was beneficial for dissolving the phosphorus that is hard to be absorbed in soil. The alpine swamp wetland developed into alpine meadow caused by drought, and the diversity of bacteria and fungi decreased, which indicating that the drought of alpine swamp wetland was not conducive to the stability of soil ecosystem, and would affect the vital ecological role of alpine swamp wetland in the process of geochemical cycle and climate regulation. The results in our previous research showed that the process of alpine swamp wetland degradation to alpine meadow would lead to obvious decline in plant community productivity and ecosystem carbon sink function [9]. Although this process is not beneficial to the suppression of climate change, an obvious transformation of swamp wetland ecosystem function from ecological function to production function can be realized, which is conducive to the development of local animal husbandry.

## CONCLUTIONS

For last several decades, the alpine swamp wetland has been degraded and shrunk due to land use and climate changes, it is necessary to protect these alpine swamp wetlands and utilize the degraded alpine wetland properly. It is suggested that surface soil moisture, fertilizers, such as nitrogen, phosphorus, organic matters and other nutrients, should be supplemented to maintain the ecological balance of the degraded alpine swamp wetlands and to improve their productivity. At the meanwhile, there are more species of bacteria and fungi in alpine wetland that have not been identified at the family, genus and species level. It is necessary that high-throughput sequencing and gene chip technology should be adopted to analyze the differences of soil microbial functional genes in alpine wetland and to provide scientific suggestions for the protection of alpine wetland and the governance of degraded wetland as well.

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## CONFLICT OF 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.

## SUPPLEMENTARY INFORMATION

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