

Division - Soil Processes and Properties | Commission - Soil Biology

Variations of soil bacterial microbial community and functional structure under different land-uses

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ABSTRACT: To reveal the differences between land-use patterns and the changes in soil properties, the changes in soil bacterial microbial communities and functions driven by land-use patterns were studied by 16SrRNA Gene Fragments and its high-throughput sequencing, relying on the wetland, farmland and forestland soils in the Hongxing National Nature Reserve of Heilongjiang Province. The study area was slightly acidic soil, and the water content, soil organic carbon content, total soil nitrogen and phosphorus contents of wetland soil were higher than farmland and forestland. The suitable C/N of wetland and farmland soil could promote the decomposition of organic matter by microorganisms. The OTUs sequence, Shannon diversity index, ACE index and Chao1 index were significantly higher in wetland than in farmland and forestland. Proteobacteria, Acidobacteria, Bacteroidetes and Actinobacteria were the dominant bacteria in the tested soil samples. The abundance of Proteobacteria in wetland soil samples was significantly higher than in farmland and forestland. The abundance of Acidobacteria in farmland soil samples was significantly higher than in wetland and forestland. According to FAPROTAX algorithm and BugBase phenotype analysis, land-uses can change the abundance of soil functional microorganisms. The dominant functional genes are mainly Chemoheterotrophy and Aerobic chemoheterotrophy bacteria. Nitrification and Aerobic ammonia oxidation bacteria involved in nitrogen cycle in farmland soil are significantly different from wetland and forestland. Land-use mode leads to significant differences between Mobile elements and Gram negative bacteria. It is mainly related to the changes in soil physical and chemical properties such as soil pH, bulk density, soil organic carbon and total nitrogen contents.

Keywords: land-uses, soil properties, bacterial community, functional structure, high throughput sequencing.

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INTRODUCTION

Wetland soil, an important environmental factor of wetland ecosystems, is highly significant for maintaining the balance and succession of wetland ecosystems (Pei et al., 2009). A change in the original function of wetland soil will promote changes in the types of wetland vegetation, soil biology and wetland biodiversity, and ultimately lead to a change in the function of wetland ecosystems (Angeloni et al., 2006). Changes in land-use patterns have led to alterations in vegetation types. In recent years, researchers in China and throughout the world have focused on changes in different land-use patterns, such as the transformation of the natural soil of forestland and grassland into farmland soil (Davidson and Ackerman, 1993; Mclauchlan, 2006; Zhao and Zhang, 2010), or the conversion of farmland into natural vegetation (Knops and Tilman, 2000; Laganier and Angers, 2020). Physical and chemical properties of the soil and its microbial environment have also changed (Wang et al., 2019a; He et al., 2020).

As one of the most important components in the wetland ecosystem, microorganisms play critical roles in the nutrient cycle of wetland soil media and soil composition and ecological restoration (Deng et al., 2019). Microorganisms are very sensitive to changes in the external soil environment, which is reflected through changes in their community composition and diversity (Vogel and Conedera, 2020). Microbial community composition and diversity are considered to be important factors to evaluate the changes in soil quality. The abundance of important Proteobacteria, Acidobacteria and Actinobacteria in the soil change in different land-uses (Zhou et al., 2017). These bacterial microorganisms are key functional groups of the nitrogen and carbon cycle (Li et al., 2021). Current research shows that the changes in soil microbial diversity, community composition and function in alpine wetlands are related to land-use and soil properties (Duan et al., 2020; Lin et al., 2021).

This study utilized three different land-use of wetland, farmland and forestland in the Hongxing National Nature Reserve (Heilongjiang Province, China) as the research object to analyze the differences in soil physical and chemical properties and nutrient contents. Changes in the richness and diversity of soil bacterial communities and the composition characteristics of related bacterial microbial functional groups were evaluated to clarify the mechanism of soil properties and microbial communities and functions under the conditions of wetland land-use changes in cold regions and further explore the geochemical cycle process of wetland ecosystems. These results can provide a scientific theoretical basis to monitor and restore wetland soil degradation in cold regions.

MATERIALS AND METHODS

Study area

The study area was located in the Hongxing National Nature Reserve, which is located northeast of Heilongjiang Province, China (128° 21' 40" ~ 128° 53' 30" E and 48° 41' 20" ~ 49° 11' 00" N). The wetland area is 111,995.3 hectares. The reserve belongs to the northern temperate continental monsoon climate. Annual average temperature is -0.7 °C; annual average precipitation is 500-610 mm; and the average relative humidity is 71.1 %.

Hongxing National Nature Reserve has high biodiversity, with typical water, swamp, meadow wetland, shrub and forest vegetation. There are 1,929 species present in the area, including 885 species of plants and 197 species of bryophytes in 49 families. There are 38 species of ferns that belong to 11 families and 650 species of seed plants that belong to 88 families (Jin et al., 2016). The soils in the sampled municipalities of Sucre are classified as Alfisols, Mollisols and Vertisols (IGAC, 2016).

The wetland park is divided into seven types of wetlands that include rivers, flood plains, herbs, mosses, swamp meadows, shrub swamps and forest swamps. Among

them, the herb and swamp meadow wetlands constitute the main wetland landscape of the wetland park, which is surrounded by farms. The vegetation of the soil sample collection site is as follows: (1) wetlands with the primary predominant plants *Deyeuxia angustifolia*, *Carex lehmanii*, *Carex meyeriana*, *Carex appendiculata*, *Caltha palustris*, *Cyamus flavicomus*, and *Pedialaris langiflora*; (2) forestland: the primary forest with mixed coniferous and broad-leaved forests and broad-leaved forests as the main plant types; and (3) farmland: the main planting crop is soybean (*Glycine max*), which is continuously planted once a year. At the end of each April, agricultural machinery tillage is conducted once, and the soybean is sown. Farm fertilization and field management are performed according to the local practices. The crop is harvested at the end of September each year, and the land is idle from the end of October to the middle of next April.

Sample collection

This experiment was conducted from August 20 to September 3, 2021, in the Key Laboratory of Wetland Ecology and Environmental Research in the cold regions of Harbin University (Harbin, China) and the Hongxing National Nature Reserve in Heilongjiang Province, China. According to the characteristics of land-use in the nature reserve, three 100 blocks of meadow wetlands, forests and the surrounding soybean farms were selected in a 100 × 100 m quadrat. Five sampling points were established according to the “S” type. A 5 cm soil shovel was used to collect soil samples that were 0.00 ~ 0.20 m deep at each sampling point. The mixed soil samples were combined into one repetition, and each sample was collected three times. In each quadrat, after the fresh soil sample was passed through a 2 mm sieve, the root residues were removed and placed in a sampling box. Part of the soil samples were brought back to the laboratory for storage as soon as possible at -20 °C for analysis of the bacterial microbial community structure. Another part was stored at 4 °C to determine the physical and chemical properties of the soil.

Determination of soil physical and chemical properties

Soil pH was measured in all soil samples. Soil moisture content was measured by drying. The ring knife method was used to determine the soil bulk density. The soil conductivity was measured by extraction. The methods of concentrated H₂SO₄ digestion and Kjeldahl were used to determine the total nitrogen (N) content of the soil samples. Content of alkali hydrolyzable N in the soil samples was determined using the alkali hydrolyzable diffusion method. Total soil phosphorus (P) was determined by HClO₄ and H₂SO₄ digestion molybdenum antimony anti colorimetry (Bao, 2005). Available P in the soil samples was determined by NaHCO₃ extraction molybdenum antimony anti colorimetry. Dichromate external heating method was used to determine soil organic matter content. Soil organic carbon was determined by the concentrated sulfuric acid potassium dichromate external heating method (Lu, 2000).

DNA extraction and high-throughput assay

Genomic DNA of the soil microorganisms was extracted with an Omega E.Z.N.A DNA Kit (Omega Bio-tek, Norcross, GA, USA). The extracted genomic DNA was detected by 1 agarose gel electrophoresis. Polymerase Chain Reaction (PCR) was performed on a Geneamp 9700 PCR system (Applied Biosystems, Thermo Fisher Scientific, Waltham, MA, USA). The universal primers 515f (5'-gtgccagcmg-cg-3') and 907r (5'-ccgtcaattcmtragtt-3') were used to amplify the V3-V4 region of the bacterial 16S rRNA gene. The PCR products were quantified using a QuantiFluor® - ST fluorometer (Promega, Madison, WI, USA), and the samples were adjusted as needed for sequencing. Finally, they were sent to Shanghai Meiji Biotechnology Co., Ltd. (Shanghai, China) for high-throughput sequencing using an Illumina HiSeq 2500 PE250 platform (San Diego, CA, USA).

Statistical analysis

Community diversity parameters (Shannon and Simpson indices) and community richness parameters (ACE and Chao indices) were used to conduct alpha diversity analyses with mothur software. The R software package was used to analyze and describe the calculation β visually. Diversity was examined by one-way analysis of variance (ANOVA) and the least significant difference. Data were statistically analyzed using Microsoft Excel 2007 (Redmond, WA, USA) and SPSS 22.0 (IBM, Inc., Armonk, NY, USA). Microbial functions of the soil bacteria were predicted by FAPROTAX and FUN Guide (Nguyen et al., 2016; Liang et al., 2020).

BugBase was used to annotate the functions of bacteria, and the operational taxonomic unit (OTU) table that clustered by sequences with 97.0 % consistency was used as the input file. The out table was standardized by the predicted number of 16S copies, and the microbial phenotype was then predicted using the preprocessed database. The threshold was automatically selected by the BugBase tools. It can be divided into the following seven categories: Gram positive, Gram negative, Biofilm forming, Contains mobile elements, Aerobic, Anaerobic and Facultative anaerobic (Ward et al., 2017; Du et al., 2020).

RESULTS

Effects of land-use on soil physical and chemical properties

The pH of soil samples was neutral (Table 1). Soil bulk density (BD) index of the farmland was the highest (1.08 Mg m^{-3}), while those of forestland were the lowest (0.83 Mg m^{-3}). Soil water, soil organic carbon (SOC), total N (TN) and total P (TP) of the wetland were significantly higher than those of the forestland and farmland ($p < 0.05$), and the contents of SOC of the wetland were 22.34 and 29.67 % higher than those of the forestland and farmland, respectively. The ratio of SOC to soil total N (TN) was the highest in forestland soil, and the ratio of SOC to the soil TP was the highest in wetland soils.

Microbial diversity of soil bacteria

The Wayne diagram was drawn based on the overlap of microbial species in the soil samples tested, which was used to analyze the common and unique soil microbial species among the different soil samples. As shown in figure 1, a total of 4,565 bacterial microbial operational taxonomic unit (OTU) sequences were detected in the soil tested. A total of 354 of these were unique to wetland soil bacteria, which accounted for 7.75 % of the total number of OTU sequences. The number of unique OTU sequences of bacteria

Table 1. Physical and chemical properties in the soil tested

Soil properties	Wetland	Farmland	Forestland
pH	6.88 ± 0.02 b	6.66 ± 0.03 c	6.92 ± 0.03 a
Bulk density (Mg m^{-3})	0.89 ± 0.07 b	1.08 ± 0.03 a	0.83 ± 0.06 b
Moisture (%)	72.36 ± 0.43 a	36.95 ± 0.29 b	38.11 ± 0.54 b
Organic carbon (g kg^{-1})	39.12 ± 0.17 a	27.51 ± 1.06 c	30.38 ± 2.23 b
Total nitrogen (g kg^{-1})	4.85 ± 0.31 a	3.42 ± 0.07 b	3.28 ± 0.33 b
Total phosphorus (g kg^{-1})	1.64 ± 0.05 a	1.33 ± 0.06 c	1.52 ± 0.02 b
Rate of soil organic carbon to soil total nitrogen	8.06 ± 0.38 b	8.04 ± 0.47 b	9.26 ± 1.02 a
Rate of soil organic carbon to soil total phosphorus	23.85 ± 0.98 a	20.68 ± 0.16 b	19.98 ± 0.04 b

Values presented as means \pm SD ($n = 3$). SD, standard deviation. * $p < 0.05$ (t-test).

in the farmland soil was 156, which accounted for 3.41 % of the total OTU sequences. The number of unique OTU sequences of the soil bacteria in forestland was 132, which accounted for 2.89 % of the total. The number of OTU sequences of the wetland bacteria was higher than those of the forestland and farmland soil.

Microbial alpha diversity

The alpha diversity index reflects the number and relative abundance of species in the community, and it is the result of competition or symbiosis among species in the community using the same habitat. By observing the information on diversity abundance of bacterial and microbial species, the changes in alpha diversity index obtained by the statistical t-test are shown in table 2. Shannon and Simpson indices and the Ace and Chao indices of the diversity index of each soil sample differed. After different land-use patterns, OTUs number, ACE index and Chao1 index showed similar patterns of changed. The OTUs and Shannon, Ace diversity and Chao1 diversity indices of the wetland soil samples were significantly higher than those of the farmlands and forestland ($p < 0.05$). The Simpson diversity index of the forestland and farmland soil samples was significantly higher than that of the wetland soil ($p < 0.05$), and there was no significant difference between them.

Variation of microbial community abundance

As shown in figure 2, the composition of dominant bacterial species in different soil samples was similar, but the levels of abundance of bacterial microbial communities in the soil samples of land-use patterns differed significantly from those of the phyla, classes, orders and families. At the taxonomic level of bacterial community in the soil samples tested,

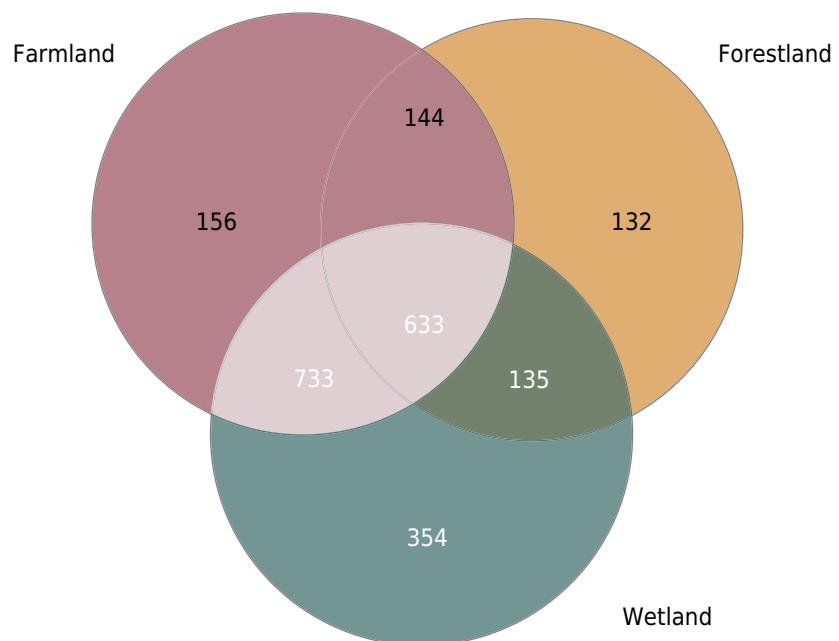


Figure 1. Venn diagram of soil microorganism based on the level of OTUs.

Table 2. Alpha diversity index of the soil bacteria in the studied area under different land-uses

Land-use	OTUs number	Shannon index	Simpson index	ACE index	Chao1 index
Wetland	1855±108 a	6.12±0.02 a	0.004±0.000 b	1319.56±23.63 a	80.77±1.34 a
Farmland	1044±43 c	5.57±0.20 b	0.014±0.003 a	1280.81±18.19 c	60.66±1.90 b
Forestland	1666±91 b	5.49±0.09 b	0.013±0.001 a	1302.98±37.91 b	61.87±1.82 b

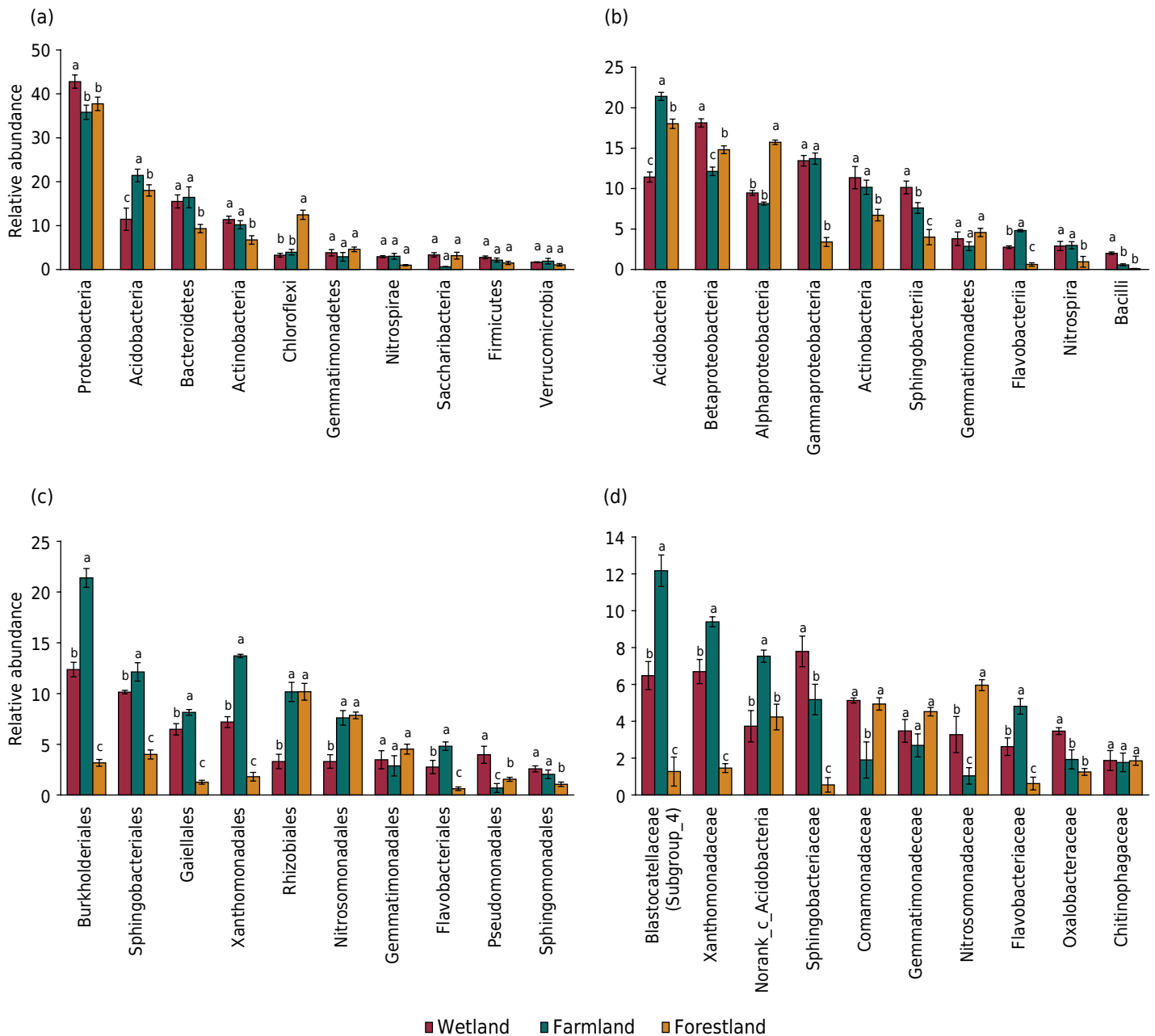


Figure 2. Relative abundance of soil bacterial communities under different land-uses at the levels of phylum, class, orders and families. (a) microbial community abundance at the phyla level; (b): microbial community abundance at the class level; (c): microbial community abundance at the orders level; (d): microbial community abundance at the families level.

Proteus, Acidobacteria, Bacteroidetes and Actinobacteria were the dominant bacteria. The abundance of Proteus in wetland soil samples was significantly higher than those in the farmland and forestland ($p < 0.05$). The abundance of Acidobacteria in the farmland soil samples was significantly higher than those in the wetland and forestland ($p < 0.05$). The abundance of Bacteroidetes and Actinobacteria in the wetland and farmland soil samples was significantly higher than in the forestland ($p < 0.05$). Acidobacteria, Proteus, Actinomycetes and Sphingobacterium were the dominant bacteria at the class level.

Correlation analysis of environmental factors

An RDA analysis revealed the correlation between the level of bacterial OTUs, community composition and varied environmental factors in the different soil samples. The analysis indicated significant differences in environmental factors among the soil samples. The interpretation of RDA1 axis, RDA2 axis and the first two axes of OTUs were 63.49, 11.92

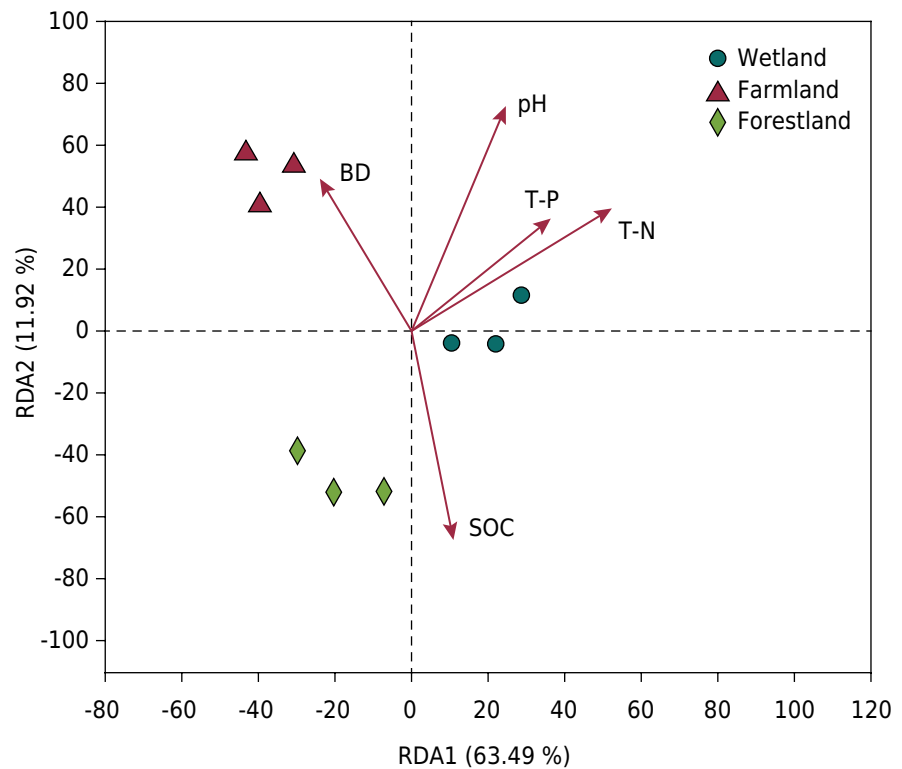


Figure 3. The RDA analysis of the soil bacterial community at the OTU level. pH: soil pH values; BD: Soil bulk density; SOC: soil organic carbon; T-N: soil total nitrogen; T-P: soil total phosphorus.

and 61.05 %, respectively. Among them, the soil samples on the first principal component axis are obviously clustered. The longer rays of pH, TN and SOC indicate that they strongly correlate with the bacterial community composition of soil samples. Wetland pH, TP and TN positively correlated with the wetland soil. There was a positive correlation between the SOC and woodland and between the BD and farmland.

Changes in the microbial functions of soil bacteria

The FAPROTAX algorithm selected the top 10 functional groups in the soil bacterial community according to their sequence ranking, and statistically analyzed the functional bacterial abundance. As shown in figure 4, land-use patterns can change the abundance of soil functional microorganisms. The dominant functional genes of bacterial community in the tested soil samples were Chemoheterotrophy and Aerobic chemoheterotrophy. The abundance of Chemoheterotrophy and Aerobic chemoheterotrophy in the wetland soils was 6.35 and 15.58 % higher than that in the farmland and forestland, respectively, but there was no significant difference among the groups. The abundance of nitrification-reducing bacteria in the forestland soil samples differed significantly from those in the wetland and farmland ($p < 0.05$). The abundance of Aerobic ammonia oxidation bacteria in the forestland soil samples was 76.44 and 91.27 % higher than those in the wetland and farmland, respectively ($p < 0.01$). There was no significant difference in the functional microbial abundance among the other soil samples.

BugBase phenotype prediction

BugBase was used to predict the abundances of seven bacterial phenotypes based on their different land-use modes (Table 5). At the level of microbial community, the abundance of aerobic bacteria in the forestland and farmland soil samples was slightly higher than that in the wetland. The change in bacterial abundance was primarily driven by Proteobacteria and Acidobacteria and finally caused by differences in the abundances of Actinobacteria, Bacteroidetes, Chloroflexi, Verrucomicrobia, Firmicutes and Planctomycetes.

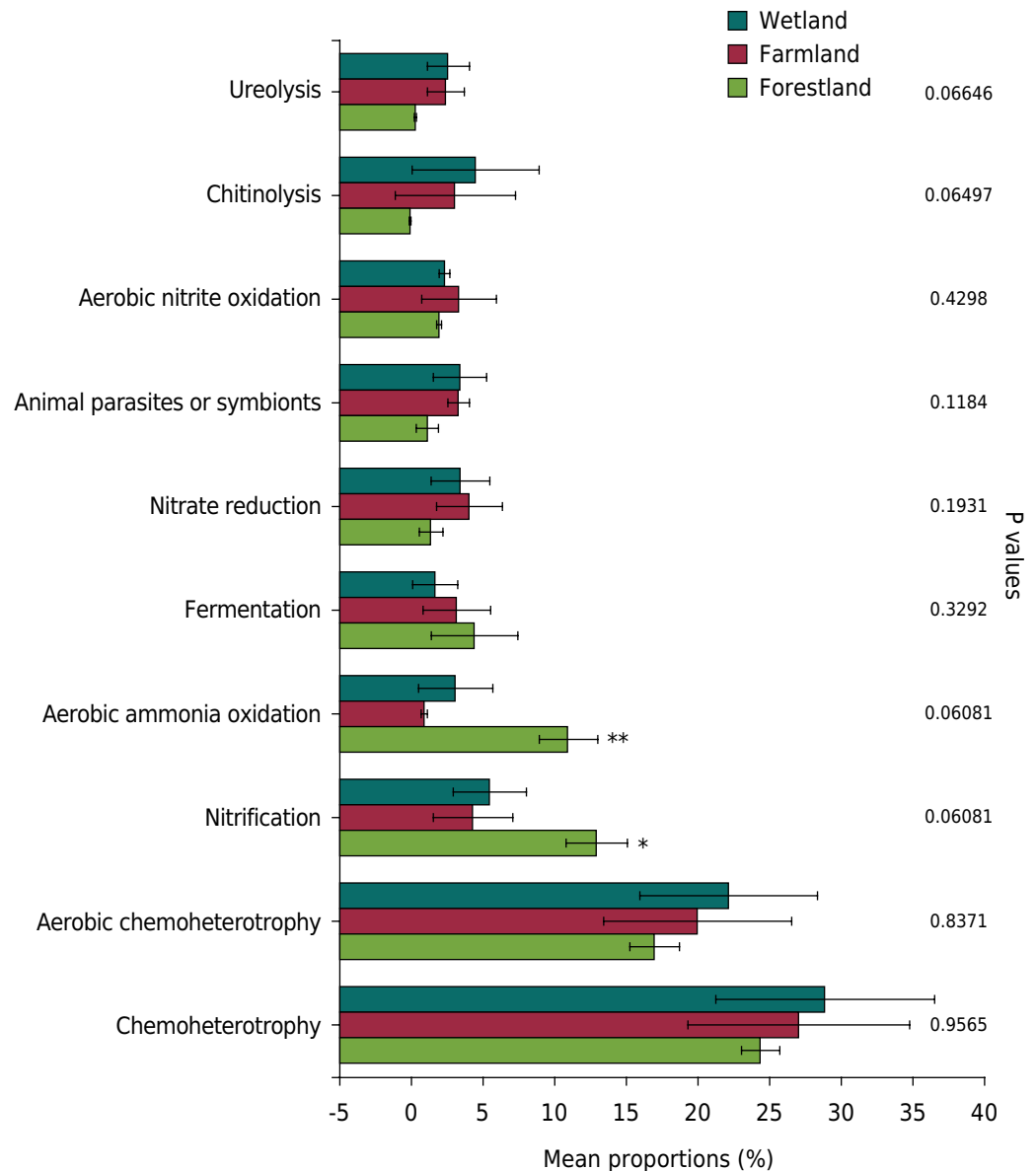


Figure 4. Abundance of soil bacterial community predicted by functional genes composition. * : $p < 0.05$; ** : $p < 0.01$; and *** : $p < 0.001$.

The abundance of anaerobic bacteria in the forestland and farmland soil samples was slightly higher than that in the wetland, but there was no significant difference among the groups. The bacterial abundance was primarily driven by Acidobacteria, which was ultimately attributed to the changes in Bacteroidetes and Proteobacteria. The abundance of facultatively anaerobic bacteria in the wetlands was higher than in the farmland and forestland, but there was no significant difference among the groups. The change in bacterial abundance was primarily driven by Proteobacteria and Actinobacteria. The bacterial abundance of phyla that contain mobile elements in the wetland soil samples was significantly higher than those in the farmland and forestland ($p < 0.05$). The species composition of bacteria primarily included members of Proteobacteria and Actinobacteria, and the change in bacterial abundance was primarily driven by Proteobacteria and Bacteroidetes. The bacterial abundance of those that form biofilms in the farmland and forestland was slightly higher than that in the wetland, but there was no significant difference among the groups. The changes of bacterial abundance were primarily caused by Proteobacteria, Acidobacteria, Actinobacteria, Chloroflexi and Nitrospirae. The abundance of Gram-negative bacteria in the forestland samples was higher than those in the wetland and farmland, which significantly correlated with them ($p < 0.01$). The

variation in bacterial abundance was primarily caused by Proteobacteria, Acidobacteria, Bacteroidetes and Nitrospirae. There was no difference in the abundance of Gram-positive bacteria among the soil samples from wetland, farmland and forestland. The changes in bacterial abundance were primarily caused by Actinomycetes, Chloroflexi and Firmicutes.

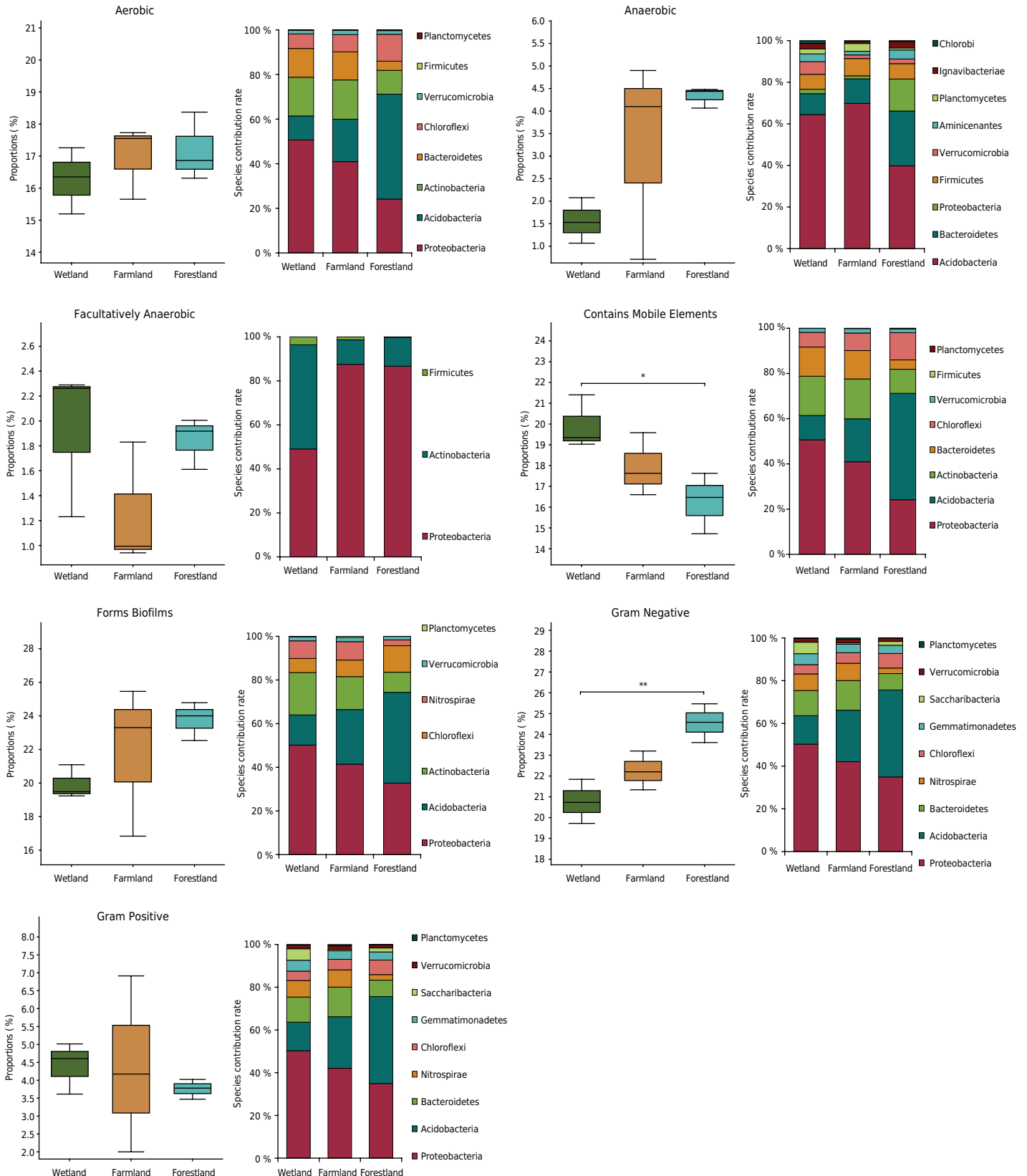


Figure 5. Prediction of bacterial BugBase phenotype in land-uses.

DISCUSSION

The process of soil nutrient cycling and transformation in wetlands in cold regions is easily affected by land-use patterns and surface vegetation types (Li et al., 2009; Zhang et al., 2019). Relevant studies show that the basic nutrient indices, such as the soil pH, TN and TP will change according to land-use (Zheng et al., 2010; Zhu et al., 2019). The indicators described above are important components of soil nutrients and can directly reflect soil fertility, quality and ecosystem productivity (Bruland and Richardson, 2006; Zhao et al., 2008; Zapata-Rios et al., 2012; Zhang et al., 2020). In this study, the organic carbon content of wetland soil was higher than that of farmland and forestland. Because the wetland in cold regions has been in the form of a swamp for a long time, the rich wetland vegetation on the soil surface and the perennial low ambient temperature can promote the process of soil humic acidification and improve the accumulation of organic matter in the wetland soil (Wang et al., 2019a).

Soil organic carbon in the forestland was lower than that in the wetland, which could be owing to the frequent enhancement of aerobic microbial activities because of the exchange between soil and external gas, which improves the rate of organic matter mineralization (Liu et al., 2013). The abundance of herbaceous plants on the surface results in a biomass accumulation of litter on the surface of wetland, which aids in the input of organic carbon to the soil (Wang et al., 2017). Owing to the frequent human disturbance of farmland soil, reasonable agricultural cultivation measures reduce the input of organic fertilizer and straw, which reduces the source of SOC (Xiao et al., 2003). Simultaneously, the relative lack of N will lead to the strengthening of soil carbon mineralization, and the N content of farmland soil also accelerates the mineralization of SOC (Yang et al., 2008). Therefore, the content of organic carbon is significantly lower than that of natural wetland ecological type soil. The soil C/N ratio can be used to measure the ability of soil N to become mineralized. An appropriate soil C/N ratio is conducive to the decomposition of organic matter by soil microorganisms and the release of soil organic N. If the C/N ratio increases, the decomposition of organic N by soil microorganisms will be limited, and the competition for inorganic N will increase (Zhang et al., 2004). Thus, this limits the normal nutrient absorption and growth of plants.

In this study, the higher carbon storage in the cold region of the study area results in a consistently low C/N. The results showed that the soil C/N ratio was significantly related to environmental and climatic factors, such as the annual average temperature, precipitation and days of sunshine; soil physical factors, such as the pH value, BD and TN; and geographical factors (Wang et al., 2011). Temperature can affect the process of soil microbial decomposition of organic matter and also lead to the different distribution of vegetation types. The C/N ratio of alpine wetlands is higher than that of alpine meadows and grasslands (Liu et al., 2012). The reason is that there are differences in the substrate "carbon pool" available to microorganisms in different types of vegetation (Bai et al., 2011).

As the most active part of wetland ecosystems, soil microorganisms can respond more sensitively to changes in land-use and can be used to evaluate the quality and health level of wetland soil. The soil microbial community structure was significantly affected by land-use patterns (Fu et al., 2019). In this study, the land-use pattern significantly changed the number of soil bacterial OTUs and the Shannon, Ace and Chao1 index and significantly changed the abundance of bacterial community in wetland soil.

The analysis of the composition of soil microbial community from the land-use mode shows that the dominant species of soil microbial community in wetland park nature reserves were Proteobacteria, Acidobacteria, Bacteroides and Actinobacteria, which can account for more than 80% of the total bacterial community in the soil samples tested. This indicates that these four phyla of bacteria are strongly adaptable environmentally, and the results are consistent with those of a previous study (Zhang et al., 2016; Zeng et al., 2017).

Amoeba are primarily involved in the decomposition of SOM and occupy the primary position in the bacterial community (Li et al., 2016; Xu et al., 2021). The abundance of amoeba in wetland soils was significantly higher than those in farmland and forestland. The abundance of Proteobacteria positively correlated with the soil carbon content and positively regulated with the soil organic matter content (Bardhan et al., 2012). However, its abundance negatively correlated with that of the soil BD, which is consistent with the changes in abundance of Proteobacteria in the soil and soil physical and chemical properties in this study (Ren et al., 2018).

From the class level, the changes in abundance of *Proteus* in wetland soil were primarily caused by Betaproteobacteria and Gammaproteobacteria. *Proteus* is considered to be an oligotrophic bacterium, which is related to the storage of SOC and content of TN (Zhang et al., 2015). Acidobacteria are both acidophilic and oligotrophic. Their distribution is regulated by environmental factors, such as pH, organic carbon, TN, C/N, P, nitrate N, ammonium N, soil humidity, soil temperature and soil respiration (Feng et al., 2016; Wang et al., 2016). They have important ecological functions, such as degrading plant residue polymers and participating in iron cycles, the light absorption of Fenna-Matthews-Olson (FMO) proteins under plant photosynthesis, and in the metabolism of single carbon compounds (Radajewski et al., 2002; Müh et al., 2007; Blöthe et al., 2008; Kalyuzhnaya et al., 2008; Lu et al., 2010; Pankratov et al., 2011).

Changes in the soil Acidobacteria were studied in different types of vegetation in Germany, and the abundance of soil Acidobacteria in meadows was found to be significantly higher than that in forestland soils, indicating that there were significant differences in the community structure of Acidobacteria in different places in the same ecological environment (Naether et al., 2012). Acidobacteria, as the second largest group of soil bacteria in northern China, is much less abundant in forestland soil (Zhang et al., 2014; Liu et al., 2015). The abundance of Acidobacteria also decreased significantly after the Amazon virgin forest in South America was transformed into farmland (Navarrete et al., 2013). High-throughput sequencing in samples from the study area showed that the change of abundance of farmland soil was significantly higher than that of the forestland and wetland at the level of Acidobacteria and class, which could be caused by environmental factors (Wu et al., 2007). Bacteroidetes species are Gram negative obligate anaerobic bacilli, which include *Bacteroides*, *Flavobacterium* and *Sphingobacterium*. This study shows that the relative abundance of *Bacteroides* in wetlands and farmland is higher than that in forestland under land-use, which is primarily caused by the difference in relative abundance between *Sphingobacterium* and *Flavobacterium*. Studied the soil microbial community structure during the transformation of Napahai Wetland from swamp to meadow and reclaimed wetland, it showed that *Bacteroides* was the secondary predominant flora in the soil, and the relative abundance of *Bacteroides* gradually increased after the soil degradation and reclamation of swamp and meadow wetlands (Chen et al., 2015; Ligi et al., 2014). The change of relative abundance was primarily affected by the alternation of dry and wet seasons, and the contents of SOC, soil TN, C/N, soil TP and available P were significantly affected (Zheng et al., 2021). The use of high-throughput sequencing technology to study the bacterial community structure in constructed river wetland sediments indicated that the difference in abundance of *Bacteroides* and other dominant flora in the wetland sediments was not only related to plant types and soil physical and chemical properties but was also affected by the reverse identified flora in the bacterial community (Zhang and Dai, 2019).

As an important microbial resource, Actinomycetes have a complex secondary metabolic system, which can produce many secondary metabolites with novel chemical structure and significant biological activity (Yu et al., 2016). Actinomycetes can decompose organic carbon under extreme environments in soils. It is dominant in poor and barren soil conditions and can decompose more difficult organic carbon by infiltrating its mycelia into enormous plant tissues (Xiao, 2014). In this study, the abundance of Actinomycetes

in wetland and farmland soil was higher than that in forestland. A correlation analysis of the environmental factors showed that the composition of soil microbial communities in wetland and farmland positively correlated with soil pH, TN content, TP content and BD. These environmental factors also restricted the relative abundance of Actinomycetes. The relative abundance of Actinomycetes in the reclamation wetland of Sanjiang Plain was 74.4 % higher than that in the marsh wetland soil. The difference in relative abundance of Actinomycetes significantly negatively correlated with the contents of soil water, soil carbon and N and significantly positively correlated with the soil pH, which is consistent with the results of this study (Xu, 2017).

The results showed that the chemoheterotrophic and aerobic chemoheterotrophic bacteria were primarily involved in the soil carbon cycle under different land-use patterns, which was consistent with the results of previous research (Siles and Margesin, 2016). The abundance of nitrification and aerobic ammonia oxidation functional genes involved in the soil N cycle in forestland was significantly higher than those in the wetland and farmland (Wu et al., 2021).

It was reported that the abundance of nitrification and aerobic ammonia oxidation functional genes decreased with the increase in BD, which was consistent with the change of BD of different land-use types in this study, which may contribute to the reduction of conductivity, and the gas exchange capacity between the soil and atmosphere is strong, which can accelerate the decomposition of organic matter by aerobic and nitrifying bacteria and microorganisms, thus, driving the process of soil N metabolism (Chen et al., 2020). An analysis of the BugBase phenotypes showed that there was a high relative abundance of aerobic bacteria with Proteobacteria and Acidobacteria as the predominant bacteria in farmland soil, which could be related to the frequent agricultural measures (Bronick and Lal, 2005). The dispersion of soil particles could affect the availability of oxygen. The farmland soil primarily contains Acidobacteria, which grows best in anaerobic and microaerobic environments, and the acidic soil is suitable for the growth of some subpopulations of Acidobacteria (Wang et al., 2016). Changes in the relative abundance of Bacteroidetes and Proteobacteria lead to the difference in anaerobic bacteria among different land-use modes. The relative abundance of facultative anaerobic bacteria driven by Proteobacteria and Actinomycetes in the wetland was higher than in farmland and forestland. Most species of Proteobacteria are facultative or obligate anaerobic and heterotrophic and can rely on the energy of photosynthesis for self-sufficiency (Zhao et al., 2012). The abundance of facultative anaerobic bacteria is substantially affected by the rich organic carbon and TN content of wetland soil (Velmourougane et al., 2017).

This study identified a positive correlation between mobile elements and antibiotic resistance genes in soil. As the main source of antibiotics, Actinomycetes cause the abundance of mobile elements in wetland soil to be significantly higher than those in farmland and forestland. Bacteria that form biofilms can degrade refractory compounds, enhance the effects of bioremediation, contribute to plant growth and root and stem restoration and protect moisture in the soil (Ligi et al., 2014). The functional groups in the biofilm-forming bacteria in the farmland and forestland were relatively abundant, indicating that their bacterial functional groups are promising for the restoration of ecological sites (Guo, 2021). Simultaneously, facultative anaerobic and biofilm-forming bacteria negatively correlated with the soil pH values. High soil pH values can lead to changes in the structure of soil particles, thus, forcing differences in microbial phenotypes. The relative abundance of Gram negative bacteria in the forestland was significantly higher than those in the farmland and wetland ($p < 0.01$). Gram negative bacteria primarily obtain their carbon nutrients from plant residues (Wong et al., 2009). The abundance of litter in forests provided a rich source of carbon for Gram negative bacteria. The results of this study show that the change in land-use leads to the heterogeneity of the living environment of microorganisms. Physical and chemical properties of the soil and

the composition, functional groups and phenotypic differences of soil microorganisms changed. Soil microorganisms can balance the distribution of resources in terms of their growth, survival and reproduction, produce selective traits or functional responses, and then regulate the phenotypic changes of microorganisms to enhance the adaptability of environment (Zhao et al., 2021).



CONCLUSION

Different land-use patterns in the studied cold region wetland led to differences in soil physical and chemical properties, the classification of dominant microbial communities and the relative abundance of functional genes. The analysis was primarily driven by land-use patterns in the changes of soil properties and biological microenvironment. Contents of SOC, TN, and TP and the Shannon diversity, Ace and Chao1 index of a wetland park nature reserve in the cold region were higher than those of the farmland and forestland. In the classification of soil bacterial community in land-use mode, Proteobacteria, Acidobacteria, Bacteroidetes and Actinomycetes were the predominant bacteria. Land-use can change the abundance of soil functional microorganisms. The dominant functional genes were primarily for Chemoheterotrophy and Aerobic chemoheterotrophy bacteria. The nitrifying and aerobic ammonia-oxidizing bacteria involved in the N cycle in farmland soil differed significantly from those in the wetland and forestland, which are primarily related to the change in soil physical and chemical properties. The BugBase phenotype analysis predicted differences in the relative abundance of seven phenotypic functional bacteria and their primary microbial communities. The soil pH, bulk density, and the contents of organic carbon and TN driven by different land-use modes affect the structure and function of microbial community.

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