



Effect of reclamation of abandoned salinized farmland on soil bacterial communities in arid northwest China

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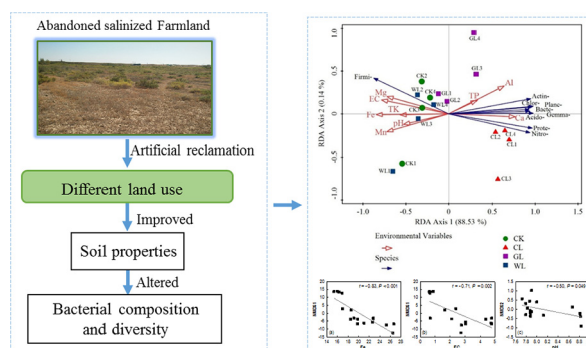
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HIGHLIGHTS

- The reclamation of abandoned salinized farmland has led to a significant decrease in soil pH and EC.
- Bacterial community composition and diversity have been greatly affected by abandoned farmland reclamation.
- Bacterial diversity indices (i.e. ACE, Chao and Shannon) dramatically increase after the reclamation, with the highest in CL.
- Soil Fe concentration, followed by EC and pH, are the major drivers of bacterial communities.

GRAPHICAL ABSTRACT



ARTICLE INFO

Article history:

Received 23 November 2017

Received in revised form 20 February 2018

Accepted 21 February 2018

Available online xxxx

Editor: Frederic Coulon

Keywords:

Abandoned salinized farmland

Reclamation

Soil bacterial community

Composition and diversity

454 pyrosequencing

ABSTRACT

Understanding the impact of reclamation of abandoned salinized farmland on soil bacterial community is of great importance for maintaining soil health and sustainability in arid regions. In this study, we used field sampling and 454 pyrosequencing methods to investigate the effects of 5-year reclamation treatments on soil properties, bacterial community composition and diversity. The four reclamation treatments are: abandoned salinized farmland (CK), cropland (CL), grassland (GL) and woodland (WL). We have found soil properties are significantly altered by abandoned salinized farmland reclamation. In particular, the lowest soil pH and electrical conductivity (EC) values are observed in CL ($P < 0.05$). The dominant phyla are Firmicutes, Proteobacteria, Chloroflexi, Actinobacteria and Acidobacteria in all treatments. At the genus levels, the relative abundance of *Bacillus*, *Lactococcus*, *Streptococcus* and *Enterococcus* in CK, GL and WL is significantly higher than in CL. Bacterial diversity indices (i.e. ACE, Chao and Shannon) dramatically increase after the reclamation, with the highest in CL. Similar patterns of bacterial communities have been observed in CK, GL and WL soils, but significantly different from CL. Regression analyses indicate that the relative abundance of these phyla are significantly correlated with soil Fe, pH and EC. Results from non-metric multidimensional scaling (NMDS) and redundancy analysis (RDA) indicate that soil Fe content, EC and pH are the most important factors in shaping soil bacterial communities. Overall, results indicate that abandoned salinized farmland reclaimed for CL significantly decrease soil pH and EC, and increase soil bacterial community diversity. Soil Fe concentration, EC and pH are the dominant environmental factors affecting soil bacterial community composition. The important role of Fe concentration in shaping bacterial community composition is a new discovery among the similar studies.

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1. Introduction

Soil salinization has become a serious environmental problem worldwide, it causes major reductions in arable land, crop productivity and quality (Shahbaz and Ashraf, 2013). China has <9% of the world's arable land to feed over 22% of its global population (Chen, 2007). Under the pressure of a rapid population growth, more and more abandoned salinized farmlands will be reclaimed to arable land. Van den Berg and Kellner (2005) found that a process of vegetation recovery through natural succession is time consuming in practice, and has little effect on soil salinity control, and Yang et al. (2016) reported that artificial reclamation is more effective than natural recovery of an abandoned salinized farmland. However, how to evaluate the recovery effects. Generally speaking, the criteria for evaluating the reclamation of abandoned salinized farmland have largely involved assessments of soil quality, physicochemical properties, vegetation coverage and economic benefits (Yang et al., 2016; Hahn and Quideau, 2013). However, the effect of abandoned salinized farmland reclamation on soil microbial ecology remains poorly understood.

Microbes represent the largest proportion (80–90%) of the Earth's biodiversity. They play an essential role in ecosystem processes (Prosser, 2012), including soil structure maintenance, organic matter decomposition, inorganic compound transformation and nitrogen fixation (Sengupta and Dick, 2015). Bacteria are the most abundant and diverse group of microbes in the soil, and major drivers of biogeochemical cycles and participate in maintaining agricultural sustainability (Gans et al., 2005). Soil bacterial communities are known to be particularly sensitive to changes in environmental factors, such as soil physical and chemical properties (Fierer et al., 2007), vegetation types (Wu et al., 2008), latitude (Zhang et al., 2015) and land use (Yergeau et al., 2006). Therefore, changes in soil bacterial composition and diversity are often used to reflect soil nutrient and environmental quality changes.

The Manasi River Basin in Xinjiang is the fourth largest irrigated agricultural area in China. The amount of cultivated land in the basin increased from 156 km² in 1949 to 6221 km² in 2010. It is important to protect the quality of agricultural soil in this basin. Excess flood irrigation in the 1970s and 1980s caused the water table to rise near the soil surface in the region, elevating the salt content of the shallow groundwater. Evaporation led to salt accumulation at the soil surface, resulting in extensive area salinized. More than a quarter of the farmland in the Manasi River Basin had been abandoned due to its declining productivity. This contributed to serious ecological problems, e.g., wind erosion, reduction of biological diversity, and desertification (Fan et al., 2008; Pimentel et al., 1995; Zhao et al., 2005). The introduction of a farming system that combines drip irrigation with plastic film mulch since 2000 has made it possible to reclaim some of the abandoned salinized farmland in this area. Artificial reclamation of abandoned salinized farmland has a potential to alter soil properties in our previous study (Yang et al., 2016). However, it remains unclear what effect abandoned salinized farmland reclamation may have on the recovery of soil bacterial community.

Manasi River Basin in northwest China provides an excellent opportunity to study the effect of abandoned salinized farmland reclamation on the soil bacterial community. The present study aims to investigate the effect of abandoned salinized farmland reclamation on soil bacterial community using 454 pyrosequencing. The objectives are to (1) assess soil property changes after the abandoned salinized farmland reclamation, (2) determine the composition and diversity of bacterial community between different reclamation treatments, (3) explore possible soil properties leading to the changes in soil bacterial community.

2. Material and methods

2.1. Site descriptions and sampling

A long-term field experiment was established in 2011 at Manasi River Basin (44°37'N, 86°08'E) in Xinjiang Province, which is an

important grain-producing area in northwest China (Fig. 1). The average annual precipitation and evaporation are 153 mm and 2005 mm, respectively. The average annual frost-free period is 169 days. Soil is classified as grey desert soil based on the local classification (Gong et al., 1988) and as Yermosol following the FAO-UNESCO system. There is no significant spatial variation in soil salinity distribution in the reclamation area according to a soil survey done by the local Soil and Fertilizer Station in 2000 (F.H Zhang et al., 2017). Vegetation on abandoned salinized farmlands is sparse, mainly consisting of *Suaeda glauca* Bunge, *Kalidium foliatum* and *Salsola collina* Pall. Grazing is prohibited in the area.

The experiment consists of three treatments: (i) cropland (CL); (ii) grassland (GL); (iii) woodland (WL). In addition, 5 ha of abandoned salinized farmland with natural succession was used as control (CK). A summary of each reclamation treatment is given in Table 1.

Soil samples were taken in the middle of July 2016, four replicates from the four plots. Three soil cores (5 cm diameter, 0–20 cm) were randomly collected using a soil auger from each individual plot and combined to form one composite soil sample per plot. Soil samples were put into sterile plastic bags, packed on ice upon collection and immediately transported to the laboratory. After plant residues and visible stones were removed, soil samples were passed through a 2-mm-mesh sieve. They were then separated into two subsamples, one portion was air-dried for the determination of the general properties, and the remainder was stored in a –80 °C refrigerator for molecular analysis.

2.2. Soil sample processing

A set of chemical indices of soil sample were measured, namely soil water content, pH, electrical conductivity (EC), total P, total K and metal elements (Ca, Mg, Al, Fe and Mn). Soil pH and EC were measured in a soil–water suspension (1:5 and 1:2.5 soil–water ratio, respectively). Soil total P and metal elements concentrations were measured after placing a soil sample (0.25 g) into a mixture of HNO₃–HF–HClO₄ (4:4:2, v/v) in a tetrafluoroethylene digestion tank at 180 °C for 12 h. The concentrations of total P, total K, Ca, Mg, Al, Fe and Mn in the extracts were measured with ICP-AES (Perkin Elmer 2100DV).

2.3. DNA extraction and PCR amplification

Each soil microbial DNA was extracted from 0.5 g subsamples using a Power Soil DNA Isolation Kit (Mo Bio Laboratories, Carlsbad, CA, USA) according to the manufacturer's instructions. The extracted soil DNA was dissolved in a 50 ml TE buffer, quantified by spectrophotometer and stored at 20 °C until use. The V4–V5 hypervariable regions of bacterial 16S rRNAs were utilized to amplify a 392 base pair fragment of the 16S rRNA gene for 454 pyrosequencing using the primer sets: 515F and 907R (515F 5'-GTGCCAGCMGCCGCGG-3', 907R 5'-CCGTCGAATT CMTTTR AGTTT-3'). This gene region is the most appropriate for accurate phylogenetic reconstruction of bacteria (Biddle et al., 2008). The V4–V5 regions of each sample were amplified using PCR (27 cycles of denaturation at 95 °C for 30 s, annealing at 55 °C for 30 s, and extension at 72 °C for 45 s; with a final extension at 72 °C for 10 min). The PCR reactions were performed in triplicate in a 20-μL mixture including 4 μL of 5 × Fast Pfu buffer, 2 μL of 2.5 mM dNTPs, 0.8 μL of each primer (5 μM), 0.4 μL of Fast Pfu polymerase, 10 ng of template DNA. PCR products were pooled together and purified using the Agarose Gel DNA purification kit (TaKaRa), and then a sequencing was conducted using the Roche 454 FLX Titanium Sequencer (Haas et al., 2011) at Shanghai Majorbio Bio-Pharm Technology Co., Ltd. (Shanghai, China).

2.4. Pyrosequencing data processing and analysis

Raw sequence data were passed through quality-filters to reduce the error rate using the Quantitative Insights into Microbial Ecology (QIIME) software (Caporaso et al., 2010). In brief, reads containing ambiguous

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