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Full Length Article

Effect of Biogas Residue on Saline Soil Microbial Community Structure based on High-Throughput 16S rRNA Metagenomics Analyses

Chun-Fang Shi[1,2], Cun-Zhu Liang[1*], Xiao-Yun Leng[2], Yue-Ming Wang[3] and Zhi-Yong Wang[4]
[1]*Department of Ecology, School of Ecology and Environment, Inner Mongolia University, Hohhot 010021, China*
[2]*Department of Biotechnology, School of Life Science and Technology, Inner Mongolia University of Science and Technology, Bao Tou 014010, China*
[3]*Department of Telecommunication, School of Information Engineering, Inner Mongolia University of Science and Technology, Bao Tou 014010, China*
[4]*Baotou City Soil and Fertilizer Workstation, Bao Tou 014010, China*
[*]For correspondence: bilcz@imu.edu.cn

Abstract

Soil salinization is a relatively common form of soil degradation in semiarid and arid regions of northern China. This phenomenon threatens the development of agriculture as well as the ecosystem. The purpose of this study was to investigate the improvement effect of biogas residue on saline soils by comparing the microbial community structure of saline soil, farmland soil and saline soil improved with biogas residue in the Tumochuan Plain of Inner Mongolia, China. The microbial community structures were examined using high-throughput 16S rRNA metagenomic analysis. Due to the improvement of biogas residue, the pH of saline soil was decreased from 8.27 to 7.96, the total salt content was decreased from 2.89 to 2.08, and the water content was decreased from 6.53 to 4.33%. The application of biogas residue increased the abundance of *Acidobacteria* at the phylum level and GP4 and GP6 at the order level in the saline soil. The microbial community structure and composition of saline soils amended with biogas residue were similar to those of farmland soil. The biogas residue had a positive effect on the microbial community of the saline soil and contributed to its improvement.

Keywords: Biogas residue; High-throughput sequencing; Microbial community structure; Saline soil

Introduction

The amount of land impacted by salinization has reached over 1000 million hectares (Mha) globally (Amini *et al.*, 2016). Soil salinization has become an important environmental problem that directly affects the sustainable development of agriculture. With the increasing trend of land resources cultivation, research has focused on the development and utilization of saline-alkali land (Singh, 2016; Peng *et al.*, 2017). In China, there are many salinization phenomena that occur in farmland soil, such as that of the Tumochuan Plain. The Tumochuan Plain is an important agricultural irrigation area and grain-producing region in Inner Mongolia, China. The agricultural irrigation in this area relies on water diversion, mainly from the Yellow River (Liu *et al.*, 2017). Due to the specific hydrogeological conditions on the plain and the long-term use of irrational flood irrigation, secondary salinization affects a large area of the soil in this region. The salinization of cultivated land has increased from one-third to one-half of the area, resulting in soil compaction, decreased fertility, reduced crop yield, land abandonment and many other issues. Land degradation, primarily due to soil salinization, restricts the sustainable economic development of the Tumochuan Plain (Jing *et al.*, 2016).

Salinized soils can be improved in a variety of ways. In recent years, there has been increasing research into the use of various types of industrial wastes to improve saline soil, including coal-fired flue gas desulfurization waste (Fan *et al.*, 2016), agricultural waste straw (Zhao *et al.*, 2016; Huo *et al.*, 2017), and excess sludge from sewage treatment plants (López-Valdez *et al.*, 2010; Fernández-Luqueño *et al.*, 2013). Biogas residue is an anaerobic fermentation residue of organic matter. As large amounts of organic matter, humic acid and nutrients needed for crop growth are contained in biogas residue, it is often used as a fertilizer (Abubaker *et al.*, 2012, 2013), though its utility for improving salinized soil has not been sufficiently investigated. Soil microbes are the main decomposers and transformers of organic matter in terrestrial ecosystems, participating in soil carbon and nitrogen cycles and other biogeochemical processes as well as the soil mineralization process (Dai *et al.*, 2017). By changing the physical and chemical properties of soil and its biological characteristics,

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soil microbes play an important role in the decomposition of organic matter and in soil nutrient conversion and supply (Luo *et al.*, 2017). Indeed, soil microbes are of great significance in the nutrient cycling of ecosystems, as they promote energy and material circulation and maintain normal ecosystem functioning (Singh and Gupta, 2018). The microbial species composition and community structure of soil largely determine the biological activity of the soil, and soil microbes can be sensitive to changes in the climate as well as soil environmental conditions. Therefore, soil microbes are increasingly studied as one of the most sensitive indicators of soil quality change, and more attention is being devoted to soil quality evaluation (Lagomarsino *et al.*, 2009; Dai *et al.*, 2016).

In this study, the composition and structure of the microbial communities in farmland soil, saline soil and amended saline soil were compared. We hypothesized that soil amended with biogas residue would show a shift in the microbial community. The objectives were to determine the effects of biogas residue amendment on the microbial community composition and structure of salinized soil and to provide a theoretical basis for the reconstruction of secondary saline-alkaline soil in the Tumochuan Plain in Inner Mongolia, China.

Materials and Methods

Study Site

The study was conducted in Tuoketuo County (40°29.262′N, 111°33.52′E, 1040 m above sea level (ASL)) in Hohhot, Inner Mongolia, China. The mean annual precipitation is approximately 410 mm, and the mean annual temperature is approximately 6.5°C (1956–2013).

Soil Sampling

A total of 9 samples were assessed in this study. Three treatments were designed: CK, saline soil; CL, biogas-improved saline soil (addition of 20 kg/m^2 biogas residue to saline soil); and GD, farmland soil. There were three replicates for each treatment. Each treatment plot was 15 m^2 (3 m × 5 m), and all treatments were cultivated with millet. The soil was treated in March 2016, and soil samples were obtained in early September 2016. In each plot, 10 soil samples were obtained at a depth of 5–25 cm in the plough layer during the harvest season. After removing impurities, the 10 soil samples were mixed and used as a pooled soil sample. The soil samples were sealed in polyethylene bags and stored at −80°C until analysis. The tested soil type was chestnut soil (CST).

DNA Extraction and Polymerase Chain Reaction (PCR) Amplification

Microbial DNA was extracted from 0.5 g of each soil sample using a Power Max Soil DNA Isolation Kit (MO Bio Laboratories, Solana Beach, CA) according to the manufacturer's protocols. The V3-V4 regions of the 16S rRNA gene were amplified using the primers 338 F 5′-ACTCCTACGGGAGGCAGCA-3′ and 806 R 5′-GGACTACHVGGGTWTCTAAT-3′. PCR amplification was carried out based on previously described methods (Peng *et al.*, 2015). The resulting products were sequenced using the Illumina MiSeq platform at Shanghai Majorbio Bio-Pharm Technology Co., Ltd. (Shanghai, China).

Statistical Analysis

After filtering, stitching, and removing chimaeras, the data were clustered into operational taxonomic units (OTUs) for species classification, and OTU similarity was set to 97%. The OTUs were clustered using Usearch (version 7.0). The sequences were aligned against the Silva database (SSU111 version: http://www.arb-silva.de/). The alpha diversity, including Chao, ACE, Sobs, Simpson diversity, Shannon diversity, Berger-Parker, Simpsoneven and Shannoneven diversity indices, as well as the observed species and phylogenetic diversity, were subjected to statistical analysis using the MOTHUR package (version v.1.30.1). Principal coordinate analysis (PCoA), which was used to measure dissimilarity at phylogenetic distances based on UniFrac analysis, was performed with QIIME (version v.1.7.0). Heatmap figures were obtained using the Vegan packages (version v.2.0.10) in the R program.

Results

Chemical Properties of the Soil

The physical and chemical properties of the soil samples are presented in Table 1. The pH, water content and total salt content of the saline soil were the highest among the three soil types, which were 8.27, 6.53%, 2.89 g/kg, respectively. The organic matter, alkaline N, available P and available K of the saline soil were the lowest of the three soil samples, which were 4.2, 28.9, 8, 128 mg/g, respectively. Compared with the saline soil, the pH, water content and total salt of the improved saline soil were reduced, whereas the organic matter, alkaline N, available P and available K were increased. These values were close to those of the farmland soil.

Composition of the Bacterial Communities

After the denoising, normalization, and clustering at the 5% cut-off level, a total of 3129 OTUs were obtained for all samples. These OTUs belong to 34 phyla, 81 classes, 174 orders, 320 families, 565 genera, and 1093 species. The dominant phyla were *Proteobacteria* (25.92%), *Acidobacteria* (21.27%), *Actinobacteria* (13.15%), and *Chloroflexi* (14.25%), followed by the *Bacteroidetes* (9.31%) and *Gemmatimonadetes* (6.81%) (Fig. 1).

Table 1: Physical and chemical properties of the soil samples

Soil samples*	pH	Soil water content (%)	Total salt (g/kg)	Organicmatter (mg/g)	Alkaline N (mg/g)	Available P (mg/g)	Available K (mg/g)
CK	8.27	6.53	2.89	4.2	28.9	8	128
GD	7.89	4.66	1.83	12.7	38.5	27.3	524.6
CL	7.96	4.33	2.08	6.49	38.5	13.4	396.7

*CK: saline soil; GD: farmland soil; and CL: improved saline soil

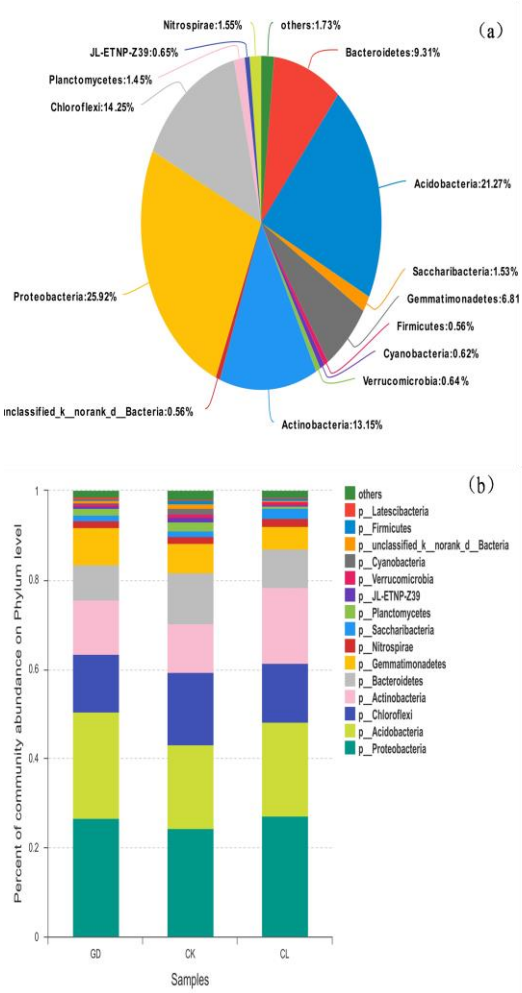

Fig. 1: (a). Relative abundances of soil bacterial phyla from the total soil samples; and (b). Different soil samples (CK: saline soil; GD: farmland soil; and CL: improved saline soil)

The structures of the bacterial communities differed among the studied soil groups. The *Proteobacteria* and *Acidobacteria* phyla were the most dominant in all soil groups, followed by *Chloroflexi* and *Actinobacteria* in the farmlandsoil and biogas-improved saline soils and *Chloroflexi* and *Bacteroidetes* in the saline soil (Fig. 1).

Alpha Diversity of the Bacterial Communities

The alpha diversity can reflect the abundance and diversity of microbial communities, which includes a series of indices based on statistical analyses. The Sobs, Chao, ACE, jack, and bootstrap indices reflect the community richness; the Simpsoneven, Shannoneven, Heip, and Smith-Wilson indices reflect the community evenness; and the Shannon, Simpson, Berger-Parker, and coverage indices reflect community diversity.

Analysis α-diversity showed that the richness and evenness indices of the saline soil were higher than those of the farmland soil and biogas-improved saline soil (Table 2). Shannon diversity indices showed the same trend. Berger-Parker and Simpson indices for the saline soil were lower than those of the farmland soil and biogas-improved saline soil. Exposure to the biogas

residue altered the diversity of the soil bacterial community in the biogas-improved saline soil; however, the results of multiple comparisons showed that diversity indices was not significantly different among the three types of soils.

Comparison of Bacterial Communities among the Three Soil Types

PCoA can be used to evaluate the similarity or variability of sample community compositions, and the species composition of two samples is closer when their distance in a PCoA map is closer. As shown in Fig. 2, the effect of the main factor Ⅰ was 64.83%, and the effect of the main factor II was 13.13%. The microbial community composition of the improved soil was similar to that of the farmland soil but was significantly different from that of the saline soil.

A heatmap can reflect the diversity and similarity of the microbial community compositions of different samples at different levels of classification using colour gradients. The species in the samples were clustered at the genus level, and heatmap was constructed according to the abundances of the sequences contained in the different OTUs in the samples after clustering (Fig. 3). Analysis of the differences in bacterial community structures showed that the abundance of soil microbes in the saline soil was slightly lower than that in the farmland and improved soils. Compared with their abundances in the saline soil, the abundances of Subgroup 6, *Sphingomonas*, RB41, *Gemmatimonadaceae*, *Nitrosomonadaceae*, JG30-KF-CM45, *Acidimicrobiales*, *Actinobacteria*, *Saccharibacteria*, *Steroidobacter*, *Streptomyces*, *Arthrobacter*, *Rhodospirillaceae*, *Chitinophagaceae*, *Blastococcus*, *Gemmatimonadaceae*, *Rubrobacter*, JG34-KF-361, and *Gaiella* were increased in the improved soil. The abundances of *Anaerolineaceae*, OM1-clade, S085, S0134-terrestrial-group, *Nitrospira*, Subgroup 7, GR-WP33-30, TK10, AKYG1722, PAUC26f, JL-ETNP-Z39, *Pontibacter*, unclassified-norank-d-Bacteria, and norank-o-BD2-11-terrestrial-group were reduced. In addition the soil microbes present in biogas residue-improved soil were similar to those in the farmland soil. The bacterial community structures of the farmland and improved soils clustered together, indicating that these two types of soils have high similarity. This result was in agreement with the results of PCoA analysis. These findings show that the addition of biogas residue changes the structure of the soil microbial community, such that it is more similar to that of farmland soil.

The heatmap in Fig. 3 depicts the relative abundance of each bacterial genus in each sample. The lower horizontal axis provides the sample names, the right vertical axis specifies the bacterial genera, the left vertical axis indicates the species cluster tree, and the upper horizontal axis provides the sample cluster tree.

Table 2: The alpha diversity indices of the bacterial communities in the three different soil types

Soil samples* Alpha diversity index		CK	GD	CL
Community richness	ACE	482±18.99	437±26.1	442±13.65
	Chao	481±14.23	444±26.02	449±21.56
	Sobs	453±15.89	412±24.52	418±13.58
Community diversity	Shannon	4.72±0.23	4.50±0.08	4.61±0.21
	Berger-Parker	0.09±0.04	0.11±0.03	0.11±0.05
	Simpson	0.02±0.01	0.03±0.01	0.03±0.01
Community evenness	Simpsoneven	0.12±0.05	0.086±0.03	0.1±0.04
	Shannoneven	0.77±0.04	0.748±0.02	0.76±0.03

*CK: saline soil; GD: farmland soil; and CL: improved saline soil

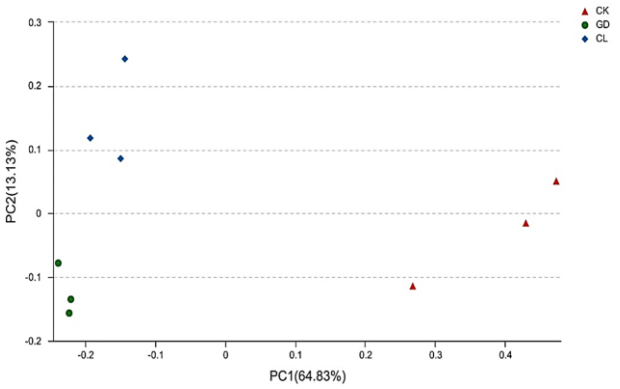

Fig. 2: The results of the PCoA analysis based on OTU abundance (97% similarity clusters)

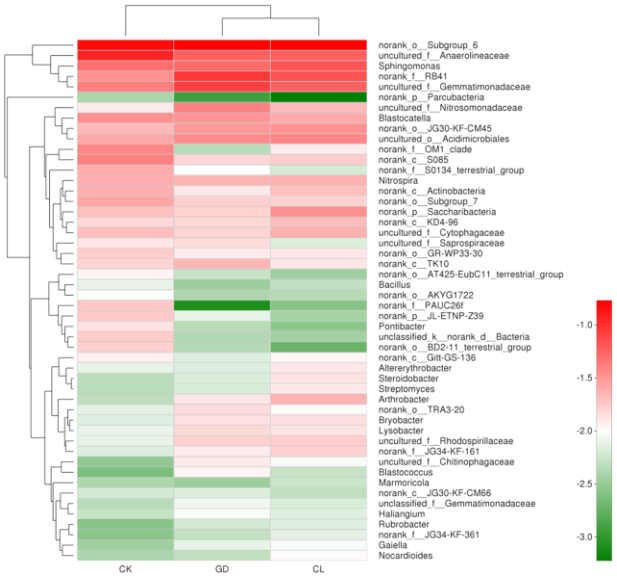

Fig. 3: The results of the clustering analysis as a heatmap of the three soil types

Discussion

Diversity indices provide important information about the rarity or commonness of species in a community. Analysing these indices is an effective method for examining the

microbial community structure of different soils because they describe both the community diversity and richness (Harcha *et al.*, 1997; Staddon *et al.*, 1997). In general, a higher the diversity index indicates higher microbial community diversity. The results of this study found that among the three kinds of soils assessed, most of the microbial diversity indices evaluated were highest in the saline soil and lowest in the farmland soil. There are several possible reasons for these findings, including the long-term and extensive use of pesticides or fertilizers in the farmland soil. Another possibility is the presence of continuous cropping factors that limit the growth of certain soil bacteria (Ekundayo, 2003) while stimulating the growth of others, thus promoting their dominance species and decreasing diversity.

Indicators of soil quality include chemical, physical and biological factors. Most of the physical and chemical indicators of soil health respond slowly compared with biological indicators, such as microbial biomass, C, N, biodiversity, soil enzymes, and soil respiration (Cardoso *et al.*, 2013). Soil microbial communities often change more rapidly with management and environmental alterations (Kennedy and Stubbs, 2006). Thus, analysing bacterial community structure is useful for evaluating soil health (Avidano *et al.*, 2005). The results of the PCoA and heatmap analyses in this study showed that the microbial community structure and composition of saline soils amended with biogas residue were similar to those of farmland soil, which indicates that the quality of saline soil was improved by the biogas residue.

The results of the present research showed that the dominant phyla in the saline soil were *Proteobacteria, Acidobacteria*, *Chloroflexi* and *Bacteroidetes.* This is slightly different from the results of other researchers, who found that the majority of taxa in saline soil were *Proteobacteria*, *Bacteroidetes* and *Firmicutes* (Mesbah *et al.*, 2007; Hollister *et al.*, 2010). This result indicates that the main groups of saline soil microbes differby region, salinization level, and climatic conditions.

The main bacterial groups in the three kinds of soil samples studied were *Proteobacteria, Acidobacteria, Actinobacteria, Chloroflexi, Bacteroidetes* and *Gemmatimonadetes*, among which *Proteobacteria* and *Acidobacteria* were the common dominant groups. In contrast, the relative contents of *Proteobacteria* and *Acidobacteria* differed among the soil types, with the relative contents of these phyla being lower in the saline soil than in the farmland and biogas-improved soils.

As the dominant orders in *Proteobacteria*, the relative contents of *Rhizobiales*, *Sphingomonadales* and *Nitrosomonadales* in the biogas-improved soil were higher than those in the saline soil. The contents of *Rhizobiales* in the saline, biogas-improved and farmland soils were 3.38, 4.88 and 3.54%, those of *Sphingomonadales* were 4.74, 7.1 and 5.63%, and those of *Nitrosomonadales* were 0.79, 1.48 and 2.94%. The bacteria of the *Rhizobiales* order (collectively, rhizobia) have the ability to form symbiotic relationships with plants, to form nodules and participate in nitrogen fixation,to increase soil fertility, and to promote plant growth (Díaz *et al.*, 2011). *Sphingomonas* bacteria can promote seed germination and plant growth, maintain the natural nitrogen balance, and remove toxic soil substances (Bending *et al.*, 2003; Tsavkelova *et al.*, 2007). Members of *Nitrosomonas*are ammonia-oxidizing bacteria that play a major role in the oxidation of nitrogen and participate in global nitrogen cycling (Kowalchuk and Stephen, 2001; Chang *et al.*, 2017). These findings suggest that biogas facilitates an increase in the abundance of beneficial microbial taxa in saline soil. In agreement with this finding, Lu and coauthors (Lu *et al.*, 2015) reported a significant increase in beneficial microbial taxa abundance (*Acidobacteria, Proteobacteria*) following BPC-PS amendment in salt-stressed soil.

The *Acidobacteria* comprise a new class of bacteria recently characterized on the basis of molecular ecology research. These microbes are distributed in various ecosystems, especially in soil. Numerous studies have demonstrated that *Acidobacteria* play a key role in soil material circulation and construction of the ecological environment (Janssen, 2006; Liu *et al.*, 2016). *Acidobacteria* are generally considered to be oligotrophs and versatile heterotrophs, exhibiting slow metabolic rates under low-nutrient conditions (Ward *et al.*, 2009). However, this was not the case in our study as our results showed that *Acidobacteria* to have the second highest abundance at the phylum level in saline soil. A previous study also detected *Acidobacteria* in an alkaline environment (Xiong *et al.*, 2012). *Acidobacteria* and *Proteobacteria* are often intimately associated with each other in the environment and may influence each other's position in the community (Janssen *et al*., 2002; Jones *et al*., 2009). Thus, the ratio between *Proteobacteria* and *Acidobacteria* (P/A) may provide insight into the general nutrient status of soils. Low P/A ratios have been hypothesized to be indicative of oligotrophic soils, whereas high ratios are observed under copiotrophic conditions (Smit *et al.*, 2001). However, our results did not confirm this hypothesis. Instead, we found that the P/A ratio was in the order saline soil (1.293) >improved soil (1.274) >farmland soil (1.113). Ellis *et al.* (2003) concluded that the *Acidobacteria* may be indicators of healthy soil. Our present results also support this conclusion. At the phylum level, the contents of *Acidobacteria* in the three soil types were saline soil (18.8%) <improved soil (21.2%) <farmland soil (23.9%); at the order level, GP4 and GP6 were the most abundant subgroups in all three soil types. The abundance of GP6 in the three types of soil was improved soil (11.02%) >farmland soil (10.87%) >saline soil (8.45%), and that of GP4 was farmland soil (8.73%) >improved soil (6.34%) >saline soil (4.04%). These results indicate that the abundances of both GP6 and GP4 were lowest in the CK soil.

Conclusion

In conclusion, the results of this study show that biogas residue has positive effects on the microbial community structure of saline soil. Biogas residue tends to shape the bacterial community structure of saline soils, such that, they become similar to that of farmland soil. The application of biogas residue in the present study increased the abundance of *Acidobacteria* at the phylum level and GP4 and GP6 at the order level in saline soil. Both the microbial community structure and the abundance of *Acidobacteria* can be useful indicators of soil health because of the sensitivity of soil microbes to soil quality. The results of this study indicate that biogas residue is beneficial for amending saline soil.

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References

Abubaker, J., H. Cederlund, V. Arthurson and M. Pell, 2013. Bacterial community structure and microbial activity in different soils amended with biogas residues and cattle slurry. *Appl. Soil Ecol.,* 72: 171–180

Abubaker, J., K. Risberg and M. Pell, 2012. Biogas residues as fertilisers – effects on wheat growth and soil microbial activities. *Appl. Energ.,* 99: 126–134

Amini, S., H. Ghadiri, C. Chen and P. Marschner, 2016. Salt-affected soils, reclamation, carbon dynamics, and biochar: a review. *J. Soil Sediment,* 16: 939–953

Avidano, L., E. Gamalero, G.P. Cossa and E. Carraro, 2005. Characterization of soil health in an italian polluted site by using microorganisms as bioindicators. *Appl. Soil Ecol.,* 30: 21–33

Bending, G.D., S.D. Lincoln, S.R. Sørensen, J.A. Morgan, J. Aamand and A. Walker, 2003. In-field spatial variability in the degradation of the phenyl-urea herbicide isoproturon is the result of interactions between degradative *Sphingomonas* spp. and soil pH. *Appl. Environ. Microbiol.,* 69: 827–834

Cardoso, E.J.B.N., R.L.F. Vasconcellos, D. Bini, M.Y.H. Miyauchi, C.A. Santos and M.A. Nogueira, 2013. Soil health: looking for suitable indicators. what should be considered to assess the effects of use and management on soil health? *Sci. Agric.,* 70: 219–303

Chang, Y., J. Fan, J. Su, H. Ming, W. Zhao, Y. Shi, F. Ji, L. Guo, S. Zan, B. Li, H. Guo and D. Guan, 2017. Spatial abundance, diversity, and activity of ammonia-oxidizing bacteria in coastal sediments of the liaohe estuary. *Curr. Microbiol.,* 74: 632–640

Dai, J., H. Wu, Z. Chang, G. Zeng, L. Jie, S. Guo, X. Li, L. Huang, L. Lu and Y. Yuan, 2016. Responses of soil microbial biomass and bacterial community structure to closed-off management (an ecological natural restoration measures): a case study of dongting lake wetland, middle china. *J. Biosci. Bioeng.,* 122: 345–350

Dai, X., H. Wang and X. Fu, 2017. Soil microbial community composition and its role in carbon mineralization in long-term fertilization paddy soils. *Sci. Total Environ.,* 580: 556–563

Díaz, R., C. Vargaslagunas, M.A. Villalobos, H. Peralta, Y. Mora, S. Encarnación, L. Girard and J. Mora, 2011. Argc orthologs from rhizobiales show diverse profiles of transcriptional efficiency and functionality in sinorhizobium meliloti. *J. Bacteriol.,* 193: 460–472

Ekundayo, E.O., 2003. Effect of common pesticides used in the niger delta basin of southern nigeria on soil microbial populations. *Environ. Monit. Assess.,* 89: 35

Ellis, R.J., P. Morgan, A.J. Weightman and J.C. Fry, 2003. Cultivation-dependent and -independent approaches for determining bacterial diversity in heavy-metal-contaminated soil. *Appl. Environ. Microbiol.,* 69: 3223–3230

Fan, Y., T. Ge, Y. Zheng, H. Li and F. Cheng, 2016. Use of mixed solid waste as a soil amendment for saline-sodic soil remediation and oat seedling growth improvement. *Environ. Sci. Pollut. Res. Int.,* 23: 21407–21415

Fernández-Luqueño, F., E. Vázquez-Núñez, C. Martínez-Suárez, G. Salomón-Hernández, C. Valenzuela-Encinas, O.F. Hernández, J.M. Ceballos-Ramírez and L. Dendooven, 2013. Bacterial community composition of a saline-alkaline soil amended or not with wastewater sludge and contaminated with polycyclic aromatic hydrocarbons (pahs). *Afr. J. Microbiol. Res.,* 7: 3605

Harcha, B.D., R.L. Corrella, W. Meechb, C.A. Kirkbyc and C.E. Pankhurstc, 1997. Using the gini coefficient with biolog substrate utilisation data to provide an alternative quantitative measure for comparing bacterial soil communities. *J. Microbiol. Meth.,* 30: 91–101

Hollister, E.B., A.S. Engledow, A.J. Hammett, T.L. Provin, H.H. Wilkinson and T.J. Gentry, 2010. Shifts in microbial community structure along an ecological gradient of hypersaline soils and sediments. *ISME J.,* 4: 829–838

Huo, L., H. Pang, Y. Zhao, J. Wang, C. Lu and Y. Li, 2017. Buried straw layer plus plastic mulching improves soil organic carbon fractions in an arid saline soil from northwest china. *Soil Till. Res.,* 165: 286–293

Janssen, P.H., 2006. Identifying the dominant soil bacterial taxa in libraries of 16s rrna and 16s rrna genes. *Appl. Environ. Microbiol.,* 72: 1719–1728

Janssen, P.H., P.S. Yates, B.E. Grinton, P.M. Taylor and M. Sait, 2002. Improved culturability of soil bacteria and isolation in pure culture of novel members of the divisions *acidobacteria*, *actinobacteria*, *proteobacteria*, and *verrucomicrobia*. *Appl. Environ. Microbiol.,* 68: 2391–2396

Jing, Y., Y. Li, Y. Yao, D. Tuo, Y. Lin, C. Di, N. Sha and H. Luo, 2016. Enzyme activities of saline-alkali soil and its relationship with soil microbial biomass and physicochemical factor. *J. Agric. Sci. Technol.,* 18: 128–138

Jones, R.T., M.S. Robeson, C.L. Lauber, M. Hamady, R. Knight and N. Fierer, 2009. A comprehensive survey of soil acidobacterial diversity using pyrosequencing and clone library analyses. *ISME J.,* 3: 442–453

Kennedy, A.C. and T.L. Stubbs, 2006. Soil microbial communities as indicators of soil health. *Ann. Arid Zone.,* 45: 287–308

Kowalchuk, G.A. and J.R. Stephen, 2001. Ammonia-oxidizing bacteria: a model for molecular microbial ecology. *Annu. Rev. Microbiol.,* 55: 485–529

Lagomarsino, A., M.C. Moscatelli, A.D. Tizio, R. Mancinelli, S. Grego and S. Marinari, 2009. Soil biochemical indicators as a tool to assess the short-term impact of agricultural management on changes in organic c in a mediterranean environment. *Ecol. Indica,* 9: 518–527

Liu, J., Y. Sui, Z. Yu, Q. Yao, S. Yu, H. Chu, J. Jin, X. Liu and G. Wang, 2016. Diversity and distribution patterns of acidobacterial communities in the black soil zone of northeast china. *Soil Biol. Biochem.,* 95: 212–222

Liu, X.B., S.G. Dong, B.W. Liu, H.B. Feng, L.I. Zheng-Kui and L.W. Liu, 2017. Hydrogeoehemical characteristics and genesis of groundwater in the tumochuan plain of inner mongolia. *Acta Geosci. Sin.,* 38: 919–929

López-Valdez, F., F. Fernández-Luqueño, M.L. Luna-Guido, R. Marsch, V. Olalde-Portugal and L. Dendooven, 2010. Microorganisms in sewage sludge added to an extreme alkaline saline soil affect carbon and nitrogen dynamics. *Appl. Soil Ecol.,* 45: 225–231

Lu, H., M.S. Lashari, X. Liu, H. Ji, L. Li, J. Zheng, G.W. Kibue, S. Joseph and G. Pan, 2015. Changes in soil microbial community structure and enzyme activity with amendment of biochar-manure compost and pyroligneous solution in a saline soil from central china. *Eur. J. Soil Biol.*, 70: 67–76

Luo, S., S. Wang, L. Tian, S. Li, X. Li, Y. Shen and C. Tian, 2017. Long-term biochar application influences soil microbial community and its potential roles in semiarid farmland. *Appl. Soil Ecol.*, 117: 10–15

Mesbah, N.M., S.H. Abou-El-Ela and J. Wiegel, 2007. Novel and unexpected prokaryotic diversity in water and sediments of the alkaline, hypersaline lakes of the wadi an natrun, egypt. *Microbial. Ecol.*, 54: 598–617

Peng, M., H. Jia and Q. Wang, 2017. The effect of land use on bacterial communities in saline-alkali soil. *Curr. Microbiol.*, 74: 325–333

Peng, M., X. Zi and Q. Wang, 2015. Bacterial community diversity of oil-contaminated soils assessed by high throughput sequencing of 16S rRNA Genes. *Int. J. Environ. Res. Pub. Hlth.*, 12: 12002–12015

Singh, J.S. and V.K. Gupta, 2018. Soil microbial biomass: a key soil driver in management of ecosystem functioning. *Sci. Total Environ.*, 634: 497–500

Singh, K., 2016. Microbial and enzyme activities of saline and sodic soils. *Land Degrad. Dev.*, 27: 706–718

Smit, E., P. Leeflang, S. Gommans, J.V.D. Broek, S.V. Mil, and K. Wernars, 2001. Diversity and seasonal fluctuations of the dominant members of the bacterial soil community in a wheat field as determined by cultivation and molecular methods. *Appl. Environ. Microbiol.*, 67: 2284–2291

Staddon, W.J., L.C. Duchesne and J.T. Trevors, 1997. Microbial diversity and community structure of postdisturbance forest soils as determined by sole-carbon-source utilization patterns. *Microbial. Ecol.*, 34: 125–130

Tsavkelova, E.A., T.A. Cherdyntseva, S.Y. Klimova, A.I. Shestakov, S.G. Botina and A.I. Netrusov, 2007. Orchid-associated bacteria produce indole-3-acetic acid, promote seed germination, and increase their microbial yield in response to exogenous auxin. *Arch. Microbiol.*, 188: 655–664

Ward, N.L., J.F. Challacombe, P.H. Janssen, B. Henrissat, P.M. Coutinho, M. Wu, G. Xie, D.H. Haft, M. Sait, J. Badger, R. Barabote, B. Bradley, T.S. Brettin, L.M. Brinkac, D. Bruce, T. Creasy, S.C. Daughertyet, T.M. Davidsen, R.T. DeBoy, J.C. Detter, R.J. Dodson, A.S. Durkin, A. Ganapathy, M. Gwinn-Giglio, C.S. Han, H. Khouri, H. Kiss, S.P. Kothari, R. Madupu, K.E. Nelson, W.C. Nelson, I. Paulsen, K. Penn, Q. Ren, M.J. Rosovitz, J.D. Selengut, S. Shrivastava, S.A. Sullivan, R. Tapia, L.S. Thompson, K.L. Watkins, Q. Yang, C. Yu, N. Zafar, L. Zhou and C.R. Kuske, 2009. Three genomes from the phylum *acidobacteria* provide insight into the lifestyles of these microorganisms in soils. *Appl. Environ. Microbiol.*, 75: 2046–2056

Xiong, J., Y. Liu, X. Lin, H. Zhang, J. Zeng, J. Hou, Y. Yang, T. Yao, R. Knight and H. Chu, 2012. Geographic distance and ph drive bacterial distribution in alkaline lake sediments across tibetan plateau. *Environ. Microbiol.*, 14: 2457–2466

Zhao, Y., Y. Li, J. Wang, H. Pang and Y. Li, 2016. Buried straw layer plus plastic mulching reduces soil salinity and increases sunflower yield in saline soils. *Soil Till. Res.*, 155: 363–370

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