agronomy

MDPI

Article

Differential Responses of Bacterial Communities in Rhizosphere and Bulk Soils of Cotton to Long-Term Amelioration Practices Based on Freezing Saline Water Irrigation and Plastic Mulching in a Coastal Saline Soil

Xiaogai Wang [1,†], Luming Wang [2,†], Zhenhua Yu [3], Yinping Tian [2], Yu Xu [2,4], Lianfu Wu [2], He Wang [2], Kai Guo [2,*] and Xinzhen Wang [2,*]

[1] School of Life Science and Engineering, Handan University, Handan 056005, China; wangxiaogaizc@126.com
[2] Key Laboratory of Agricultural Water Resources, Hebei Key Laboratory of Soil Ecology, Center for Agricultural Resources Research, Institute of Genetics and Developmental Biology, Chinese Academy of Sciences, Shijiazhuang 050022, China; wangloo0917@126.com (L.W.); tianyinping@sjziam.ac.cn (Y.T.); njauxuyu@163.com (Y.X.); wulianfu1234@163.com (L.W.); wangheucas@outlook.com (H.W.)
[3] State Key Laboratory of Black Soils Conservation and Utilization, Northeast Institute of Geography and Agroecology, Chinese Academy of Sciences, Harbin 150081, China; yuzhenhua@iga.ac.cn
[4] Hebei Provincial Laboratory of Water Environmental Science, Hebei Provincial Academy of Ecological and Environmental Sciences, Shijiazhuang 050021, China
* Correspondence: guokai@sjziam.ac.cn (K.G.); xzwang@sjziam.ac.cn (X.W.); Tel./Fax: +86-031-185-817732 (K.G.); +86-031-185-811816 (X.W.)
† These authors contributed equally to this work.

Abstract: Soil amelioration in coastal saline areas plays an important role in alleviating land resource shortages, improving regional ecological environments, ensuring food security, and promoting economic development. Plastic mulching (M) and the combination of freezing saline water irrigation and plastic mulching (WIM) are successful amelioration practices that dramatically reduce the salinity of surface soil and facilitate plant growth in coastal saline soil. However, the bacterial responses that are closely related to these amelioration practices in coastal saline soil remain poorly understood. In this study, bacterial richness and diversity, community composition, and potential ecological functions in the rhizosphere and bulk soils of cotton in M and WIM treatments, along with a control treatment, were investigated using high-throughput sequencing in a coastal saline field. The results showed that both the M and WIM treatments increased bacterial richness and alpha diversity, which were in general significantly higher in bulk soil than in rhizosphere soil. Non-metric multidimensional scaling and the Bray–Curtis dissimilarity analysis revealed that the bacterial community in rhizosphere soil was assembled far from those in the control and bulk soils and behaved more specifically in rhizosphere soil than in bulk soil. The relative abundances of most of the dominant phyla showed opposite trends of variation in bulk and rhizosphere soils compared to those in control soil in both M and WIM treatments; in particular, the specific bacterial groups of Proteobacteria and Actinobacteria decreased in bulk soil but significantly increased in rhizosphere soil. Functional groups of chemoheterotrophy, aerobic chemoheterotrophy, and nitrate reduction were predominant in rhizosphere rather than bulk soil, according to the Functional Annotation of Prokaryotic Taxa. These findings improve the understanding of the mechanism of bacterial responses to amelioration practices M and WIM in coastal saline soils and provide valuable information for the development of amelioration techniques based on agricultural practices and soil microbiome to enhance plants' adaptability to saline soil in the future.

Keywords: salinity; coastal saline soil; amelioration practice; soil microorganism; rhizosphere; bacterial community

Citation: Wang, X.; Wang, L.; Yu, Z.; Tian, Y.; Xu, Y.; Wu, L.; Wang, H.; Guo, K.; Wang, X. Differential Responses of Bacterial Communities in Rhizosphere and Bulk Soils of Cotton to Long-Term Amelioration Practices Based on Freezing Saline Water Irrigation and Plastic Mulching in a Coastal Saline Soil. *Agronomy* **2024**, *14*, 103. https://doi.org/10.3390/agronomy14010103

Academic Editor: Wanting Ling

Received: 16 November 2023
Revised: 25 December 2023
Accepted: 29 December 2023
Published: 31 December 2023

1. Introduction

Soil salinity is a primary ecological matter which threatens environmental resources, food demand, and human health in many countries and regions of the Earth and causes the degradation of soil structure and a reduction in crop yield worldwide [1–8]. Approximately 1.1×10^9 hectares of soil suffers with salinity stress; moreover, the area of saline soil is increasing by 1.5×10^6 hectares per year owing to human-induced factors such as excessive irrigation water, irrigation with saline water, and poor agricultural management, and some other natural factors such as low precipitation, high temperature and enhanced surface evaporation [2,3,6,7,9–11]. Owing to the high salinity and poor structure of salinized soil, soil humus is easily lost, thereby resulting in a decrease in the content of soil organic matter, fertility, and crop yield [12]. Exploiting saline soils as a potential land resource for agricultural development to alleviate the shortage of land resources may improve regional ecological environment, ensure food security, and promote economic development [2,3,10,13]. Several measures, including physical, chemical, biological and mechanical methods, have been employed to remediate salinized soil, among which salt leaching with fresh water is the most effective and universally used approach [3,14,15].

Coastal saline soil is a typical saline soil with high salinity and low nutrient content, which limits the growth and development of plants [10]. Amelioration practices implementing biochar [13,16,17], vermicompost, humic acid fertilizer [4], phytoremediation [15,18], rainfall leaching, ditch and pipe drainage systems, plastic mulching, and freezing saline water irrigation [18–20] have been developed for the remediation of coastal saline soil. Plastic mulching (M) improves saline soil by reducing evaporative soil water and preventing accumulation of soluble salts to the surface soil with water flow [18,21]; this process has remarkably improved microbial abundance, soil respiration, and the rate of plant seed germination [20,22]. Irrigation using freezing saline water makes full use of local saline water resources to save freshwater resources, significantly reduces soil salinity, and promotes plant growth, consequently becoming a typical case of using saline water resources to desalinize coastal saline soil in the semi-humid continental climatic zone [14,19,20,22]. In the coastal saline soil in North China, the combination of freezing saline water irrigation in cold winter and plastic mulching in spring (WIM), which fully considers the seasonal climate characteristics and laws of water, salt movement, and plant growth, has been a successful amelioration practice, dramatically reducing the salinity of surface soil and facilitating plant growth in field experiments [14,19,20]. This technology holds soil moisture and inhibits salt return in spring, thus providing suitable conditions for sowing and for the emergence of spring crops [14,19,20].

Soil microbial communities have a key ecological function in maintaining and regulating the function of various ecosystems by participating in the cycling process of most soil elements in the soil ecosystem and storing material and energy for plant productivity [12,23–25]. The activities of soil microbial communities have a close association with the decomposition and transformation of organic and inorganic substances, and with plant growth, development, and stress tolerance in eco-agriculture [1,18,26–29]. Their activities are mainly reflected by their richness, diversity, structure, and function, which respond sensitively to environmental changes, such as changes in the physicochemical properties of soil, climate change, human activities, and host plant species, developmental stage, litter and root exudates [30–32]. Consequently, these properties of soil microbiota are often used as bioindicators of soil quality [24,25,32,33]. Moreover, an immense number of microorganisms live in the plant rhizosphere, which is the pivotal interface for tight interplay between microorganisms and plants [34–37]. Soil microorganisms related to plant roots are very important for plant growth, health, productivity, stress resistance and biological control of plant diseases and insect pests, and they are considered to be the second genome of plants [7,34,36,38–40]. Recently, rhizosphere microbes have attracted extensive attention, and a considerable number of studies have broadened our knowledge of soil microbial responses to botanical and environmental changes [36,37,39–43].

With the rapid development of high-throughput sequencing (HTS) and bioinformatics analysis in recent years, a powerful tool has been provided for research on microbiome [44,45]. HTS is a culture-independent molecular technique that is characterized by precision, rapidity and an informative nature [46]. Amplification sequencing is the most widely used method for quickly revealing the composition of microbiota [47]. The development of high-throughput sequencing has given us a better understanding of the responses of soil microbiome to different crop planting patterns [48], fertilizer applications [49], tillage practices [50] and crop types [51] in agroecosystems. In addition, these factors provided us with an in-depth understanding of the coastal saline soil microorganisms under different conditions. Studies showed that Proteobacteria Actinobacteria, Bacteroidetes and Firmicutes were the most predominant taxa in the rhizosphere of saline soil, and the salinity is the dominant factor influencing the changes in microbial community structure [52,53]. Furthermore, the plant root recruited specific root-derived bacteria (RDB), such as Bacillus, Pseudomonas, and Rhizobacteria, into the rhizosphere to enhance the plant's resistance to salt stress [7,54,55]. In our previous work, we revealed the responses of the different root zone microbiota of different plant types under long-term phytoremediation in the coastal saline soil, and found that marine-associated taxa have a high relative abundance in the coastal saline soil and decrease in amelioration soils as a result of high-throughput sequencing [33,51]. However, we still lack a deep understanding of differential responses of bacterial communities in rhizosphere and bulk soils to different amelioration practices in coastal saline soil. Therefore, this study investigated the responses of bacterial richness and diversity, community composition, and potential functions in rhizosphere and bulk soils under long-term amelioration practices in a coastal saline field; examined the effects of different amelioration practices on bacterial communities; and analyzed the different responses of bacterial communities in rhizosphere and bulk soils to amelioration practices.

2. Materials and Methods

2.1. Site Description and Treatments

This study was conducted in a coastal saline field in Haixing County, Hebei Province, China (117°33′49″ E and 38°10′02″ N). Details of the climate, soil, and water characteristics in this region and the experimental design of the long-term amelioration practices initiated in 2008 have been previously reported [20,22,33,56]. In this study, three experimental treatments with three replicates were selected, including control (wasteland without amelioration), plastic mulching (M, with 0.07 mm polyethylene plastic film in spring, Runtian, Jiangsu, China), and the combination of freezing saline water irrigation and plastic mulching (WIM, with a salinity of 9.59 $g \cdot L^{-1}$ saline water irrigation in winter and 0.07 mm plastic film in spring) [20,22,33,45]. Briefly, saline water was irrigated in winter when the temperature was less than −10.3 °C to form a 180 mm ice layer, and plastic mulching was applied in the following spring after the ice melted and the meltwater infiltrated the soil [20,22,33]. Subsequently, cotton (*Gossypium hirsutum* Linn.) seeding was conducted in May.

2.2. Soil Sample Collection

The rhizosphere and bulk soil samples of cotton in M and WIM treatments (0–20 cm; named M_Bulk, M_Rhizosphere, WIM_Bulk, and WIM_Rhizosphere, respectively), along with soil samples from the control treatment (0–20 cm; named Control), with three replicates per sample, were collected in August 2017 (at the flowering stage of cotton growth). Bulk soil was collected from plots without visible roots between two cotton plants (60 cm). For each sample, soil from five randomly selected plots was sieved through 2 mm meshes, fully mixed, and kept at −80 °C for DNA extraction and 4 °C or air dried for physicochemical analysis. Rhizosphere soil samples were collected using a previously reported method with some modifications [35,57]. After loose soil was removed from the surface of cotton roots, the roots were placed in a tube with 30 mL phosphate-buffered solution (pH 7.0) and shaken at 180 rpm for 30 min in a shaker. Then, the turbid solution was centrifuged

at 10,000× *g* for 2 min after removal of the roots, and the precipitate was collected as a rhizosphere soil sample and stored at −80 °C for DNA extraction.

2.3. Physicochemical Properties of Soil

Soil pH and electrical conductivity (EC) were measured using a pH meter (FE28, Mettler-Toledo, Zurich, Switzerland) and a conductivity meter (DDS-307A, LEI-CI, Shanghai, China), respectively, with the filtrate of 5 g air-dried soil and 25 mL deionized water without CO_2 mixed at 200 rpm for 30 min in a shaker. Moisture content (MC) was measured using the drying–weighing method by drying soil samples at 105 °C. Total carbon (TC) and total nitrogen (TN) contents were directly measured using a CHNS elemental analyzer (Vario MAX, Elementar, Langenselbold, Germany). Soil nitrate nitrogen (NO_3^--N) and ammonia nitrogen (NH_4^+-N) contents were measured using an ultraviolet spectrophotometer (UV-6100S, Shanghai Metash Instruments Co., Ltd., Shanghai, China) with the dual-wavelength method [58] and indophenol blue colorimetry method (A_{625} nm), respectively, in the filtrate of 5 g fresh soil and 50 mL of 2 M KCl mixed at 200 rpm for 1 h in a shaker. The available potassium (AK) was measured in an extract of air-dried soil with 1 M ammonium acetate solution at a ratio of 1:5 (*w*/*v*) using a flame photometer (FP640, INASA Instrument, Shanghai, China). The available phosphorus (AP) was measured in an extract of air-dried soil with 0.5 M sodium bicarbonate solution at a ratio of 1:5 (*w*/*v*) using the molybdenum blue method [59]. Soil organic carbon (SOC) was measured using the dichromate oxidation method [60].

2.4. Soil DNA Extraction and Amplicon Sequencing

Total genomic DNA was extracted from the soil using a FastDNA Spin Kit (MP Biomedicals, Santa Ana, CA, USA) according to the manufacturer's instructions and saved at −80 °C for further analysis. DNA concentration and purity were measured using a NanoDrop One spectrophotometer (Thermo Fisher Scientific, Madison, WI, USA). The V4 region of bacterial 16S rRNA gene was amplified with the primers 515F (5′-GTGYCAGCMGCCGCGGTAA-3′) and 806R (5′-GACTACNVGGGTWTCTAAT-3′) [61,62]. A high-throughput sequencing library was constructed using a two-step polymerase chain reaction (PCR) amplification. The first-step PCR amplification was carried out in a 25 μL mixture containing 12.5 μL 2 × premix Ex TaqTM (Takara Biotech, Dalian, China), 0.5 μL (10 μM) each of forward and reverse primers, 1 μL DNA template (20–30 ng/μL) and 10.5 μL double-distilled H_2O. The PCR amplification conditions were as follows: initial denaturation at 95 °C for 3 min, followed by 28 cycles of denaturation at 95 °C for 30 s, annealing at 50 °C for 1 min, and extension at 72 °C for 1 min, and a final extension at 72 °C for 10 min. PCR products were detected via 1.5% agarose gel electrophoresis and purified using AMPure XP beans (Beckman Coulter Inc., Brea, CA, USA) to remove the redundant bases and primer dimers. Then, the second-step PCR amplification was performed with the same conditions as mentioned above, except that the Illumina sequencing connector and barcode sequence were added to the mixture and amplified for eight cycles. After detection via gel electrophoresis and purification using AMPure XP beans (Beckman Coulter Inc., Brea, CA, USA), the PCR products were sent for high-throughput sequencing using an Illumina MiSeq platform (Genewiz, Nanjing, China). The sequencing data obtained in this study were deposited in the Sequence Read Archive (SRA) under the BioProject accession number PRJNA970964.

2.5. Bioinformatic Analysis

Paired-end reads were joined after the adapter and primer sequences were removed from the raw reads of high-throughput sequencing using Cutadapt v. 1.18 [63,64]. Low-quality sequences (expected errors per base > 0.001, containing N, and length < 150 bp) were excluded. The UCHIME algorithm (uchime3_denovo) was used to remove the chimeric sequences [65]. High-quality sequences were clustered into operational taxonomic units (OTUs) at a similarity level of 97% using VSEARCH v. 2.21.1 and denoised using the

UNOISE algorithm v. 3. The OTUs of bacteria were classified based on the Silva database v. 132 using the RDP Classifier [66], whereas the OTUs that were unclassified and allocated as chloroplasts or mitochondria were removed. For statistical analysis, the OTUs table was further subsampled, with 11,000 sequences for each sample. Functional prediction of soil bacterial community was performed using Functional Annotation of Prokaryotic Taxa (FAPROTAX) [67].

2.6. Statistical Analysis

Statistical analysis of physicochemical properties of soil, observed OTUs, bacterial alpha diversity indexes, including Chao1, ACE, Shannon, Simpson, and Fisher, and the relative abundance of dominant bacterial taxa was performed in R v. 3.4.3 with the "dplyr" package based on the Kruskal–Wallis rank sum test [33,68]. The non-metric multidimensional scaling (NMDS) analysis was performed using R v. 3.4.3 with the "vegan" package based on the Bray–Curtis distance to visualize the structure of microbial communities. Heatmaps showing the Bray–Curtis dissimilarity and functional profiles of bacterial communities in rhizosphere and bulk soils under different amelioration practices were conducted using R v. 3.4.3 with the "pheatmap" package. The Bray–Curtis dissimilarity is an algorithm method of weighted parameters based on species abundance and presence or absence. Venn diagram showing the distribution of bacterial OTUs under different amelioration practices was generated using R v. 3.4.3 with the "gplots" package.

3. Results

3.1. Physicochemical Properties of Soil

The physicochemical properties of bulk soil under amelioration practices M and WIM, and those of the control soil, are summarized in Table 1. The highest EC (3.45 ± 1.47 mS·m^{-1}) was recorded in the control soil, followed by those of M_Bulk soil (1.02 ± 0.60 mS·m^{-1}) and WIM_Bulk soil (0.94 ± 0.50 mS·m^{-1}). M and WIM treatments significantly decreased salt concentration, as represented by the EC value of bulk soil compared to that of the control; however, no significant difference in salt concentration was observed between M and WIM treatments. TN contents in all soils were similar; however, TC contents in M_Bulk and WIM_Bulk soils were slightly but non-significantly higher than that in the control soil, which led to a higher soil C:N ratio in the M and WIM treatments than that in the control treatment. The highest NH_4^+-N and NO_3^--N contents were recorded in the control soil, followed by those of M_Bulk and WIM_Bulk soils. However, AP and AK contents increased in M_Bulk soil but decreased in WIM_Bulk soil compared to those in the control soil.

Table 1. Physicochemical properties of soil in different amelioration treatments (mean ± standard deviation).

Properties	Control	M	WIM
pH	8.07 ± 0.08 b	8.41 ± 0.24 b	8.87 ± 0.10 a
EC (mS·m^{-1})	3.45 ± 1.47 a	1.02 ± 0.60 b	0.94 ± 0.50 b
TC (%)	1.42 ± 0.05 a	1.50 ± 0.04 a	1.46 ± 0.06 a
TN (%)	0.08 ± 0.01 a	0.08 ± 0.00 a	0.08 ± 0.00 a
SOC (%)	0.70 ± 0.11 a	0.74 ± 0.06 a	0.68 ± 0.08 a
MC (%)	25.08 ± 0.98 a	25.17 ± 1.98 a	23.32 ± 0.65 a
NH_4^+-N (mg·kg^{-1})	1.15 ± 0.36 a	0.74 ± 0.16 ab	0.47 ± 0.16 b
NO_3^--N (mg·kg^{-1})	85.56 ± 50.86 a	58.24 ± 43.96 a	23.06 ± 11.34 a
AK (mg·kg^{-1})	132 ± 26 a	144 ± 10 a	119 ± 10 a
AP (mg·kg^{-1})	39.16 ± 11.87 a	48.66 ± 25.08 a	23.14 ± 13.30 a
C:N	17.25 ± 2.63 a	18.05 ± 1.31 a	18.84 ± 1.07 a

M—plastic mulching; WIM—the combination of freezing saline water irrigation and plastic mulching; EC—electrical conductivity; TC—total carbon; TN—total nitrogen; SOC—soil organic carbon; MC—moisture content; NH_4^+-N—ammonia nitrogen; NO_3^--N—nitrate nitrogen; AK—available potassium; AP—available phosphorus. Values followed by different letters in the same row are significantly different according to Duncan's test ($p < 0.05$).

3.2. Bacterial Richness and Diversity in Soil

A total of 242,253 high-quality 16S rRNA gene sequences were obtained from all 15 soil samples after quality filtering, ranging from 11,734 to 20,872 reads per sample (mean = 16,163). Both M and WIM treatments increased bacterial richness and alpha diversity in soil compared to the control treatment, which were generally significantly higher in bulk soil than in rhizosphere soil (Figure 1). Respectively, 2453, 2355, 2167, 2121 and 1991 observed OTUs were identified in WIM_Bulk, M_Bulk, M_Rhizosphere, WIM_Rhizosphere and control soils (Figure 1). The number of observed OTUs was higher in both M and WIM treatments than that in the control treatment and was significantly higher in bulk soil than that in rhizosphere soil. Additionally, the highest bacterial Chao1, ACE, Shannon, and Fisher diversity indexes were recorded in WIM_Bulk soil, followed by those of M_Bulk, M_Rhizosphere, WIM_Rhizosphere and control soils (Figure 1). The bacterial alpha diversity was significantly higher in WIM_Bulk soil than in WIM_Rhizosphere soil, as was evident from the bacterial Chao1, ACE, Shannon and Fisher diversity indexes, and was significantly higher in M_Bulk soil than in M_Rhizosphere soil according to the bacterial Shannon, Simpson and Fisher diversity indexes (Figure 1).

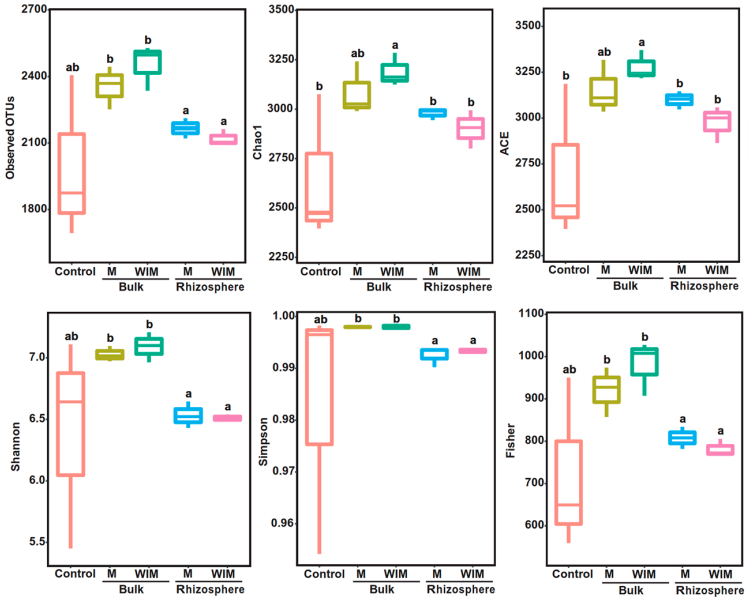

Figure 1. Observed OTUs and alpha diversity indexes of soil bacterial communities. Different letters on the top of the square indicate significant differences among treatments (detected via Kruskal–Wallis rank sum test, $p < 0.05$). M, plastic mulching; WIM, the combination of freezing saline water irrigation and plastic mulching.

3.3. Composition of Bacterial Community in Soil

At the phylum level, Proteobacteria, Bacteroideta, Gemmatimonadota, Acidobacteria, Planctomycetota, Chloroflexi, Actinobacteriota, Verrucomicrobiota, Myxococcota, Methylomirabilota, Firmicutes, Nitrospirota, Cyanobacteria and Entotheonellaeota were the most predominant taxa accounted for under different amelioration conditions (Figure 2; Table S1). Changes in the relative abundances of certain dominant bacterial taxa showed obvious opposite trends of variation in rhizosphere and bulk soils compared to those in control soil in both M and WIM treatments (Figure 2; Table S1). M and WIM treatments increased the relative abundances of Gemmatimonadota, Acidobacteria, Planctomycetota, Chloroflexi, Myxococcota and Nitrospirota in the bulk soil but decreased those in the rhizosphere soil, whereas these treatments decreased the relative abundances of Proteobacteria, Actinobacteriota and Firmicutes in the bulk soil (except for Firmicutes in WIM_Bulk soil) but significantly increased those in the rhizosphere soil, indicating the cotton had recruited such specific bacterial groups from the bulk soil to the rhizosphere soil (Figure 2;

Table S1). Furthermore, the relative abundances of Acidobacteria, Planctomycetota, and Nitrospirota were significantly increased in bulk soil, whereas the relative abundances of Gemmatimonadota and Myxococcota significantly decreased in rhizosphere soil in M and WIM treatments (Figure 2; Table S1).

A total of 4291 bacterial OTUs were identified based on a 97% similarity level from 15 soil samples, of which 3479, 3440, 3174, 3090 and 3009 OTUs were from the M_Bulk, WIM_Bulk, M_Rhizosphere, WIM_Rhizosphere and control soil samples, respectively (Figure 3). A large proportion of OTUs were shared across M and WIM treatments, with 3076 OTUs in bulk soil and 2670 OTUs in rhizosphere soil, accounting for 80.04% and 74.29% of the total OTUs shared in M and WIM treatments, respectively (Figures 3 and 4). Meanwhile, 403 OTUs and 364 OTUs accounting for 10.49% and 9.47% of the total OTUs in bulk soil were unique in M and WIM treatments, respectively, and 504 OTUs and 420 OTUs accounting for 14.02% and 11.69% of the total OTUs in rhizosphere soil were unique in M and WIM treatments, respectively (Figures 3 and 4). In addition, 2503 OTUs in M treatment and 2497 OTUs in WIM treatment accounted for 60.31% and 61.91% of the total OTUs shared by bulk and rhizosphere soils, respectively (Figures 3 and 4). Meanwhile, 976 OTUs and 671 OTUs accounting for 23.52% and 16.17% of the total OTUs in M treatment were unique in bulk and rhizosphere soils, respectively, and 943 OTUs and 593 OTUs accounting for 23.38% and 14.70% of the total OTUs in WIM treatment were unique in the bulk and rhizosphere soils, respectively (Figures 3 and 4).

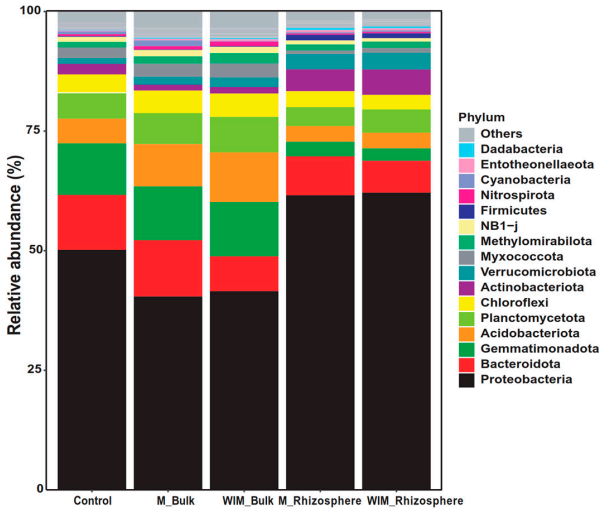

Figure 2. Stacked column showing the dominant phyla bacteria in soil under different treatments. M—plastic mulching; WIM—the combination of freezing saline water irrigation and plastic mulching.

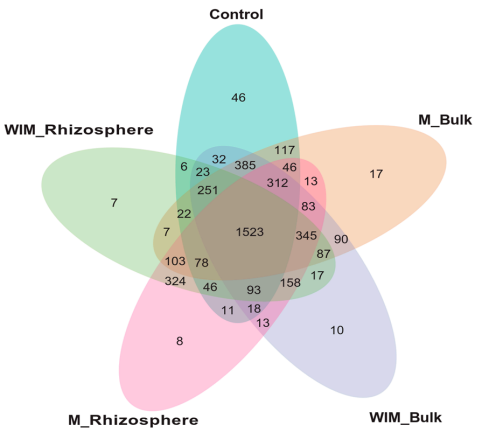

Figure 3. Venn diagram showing the distribution of bacterial OTUs in different treatments. M—plastic mulching; WIM—the combination of freezing saline water irrigation and plastic mulching.

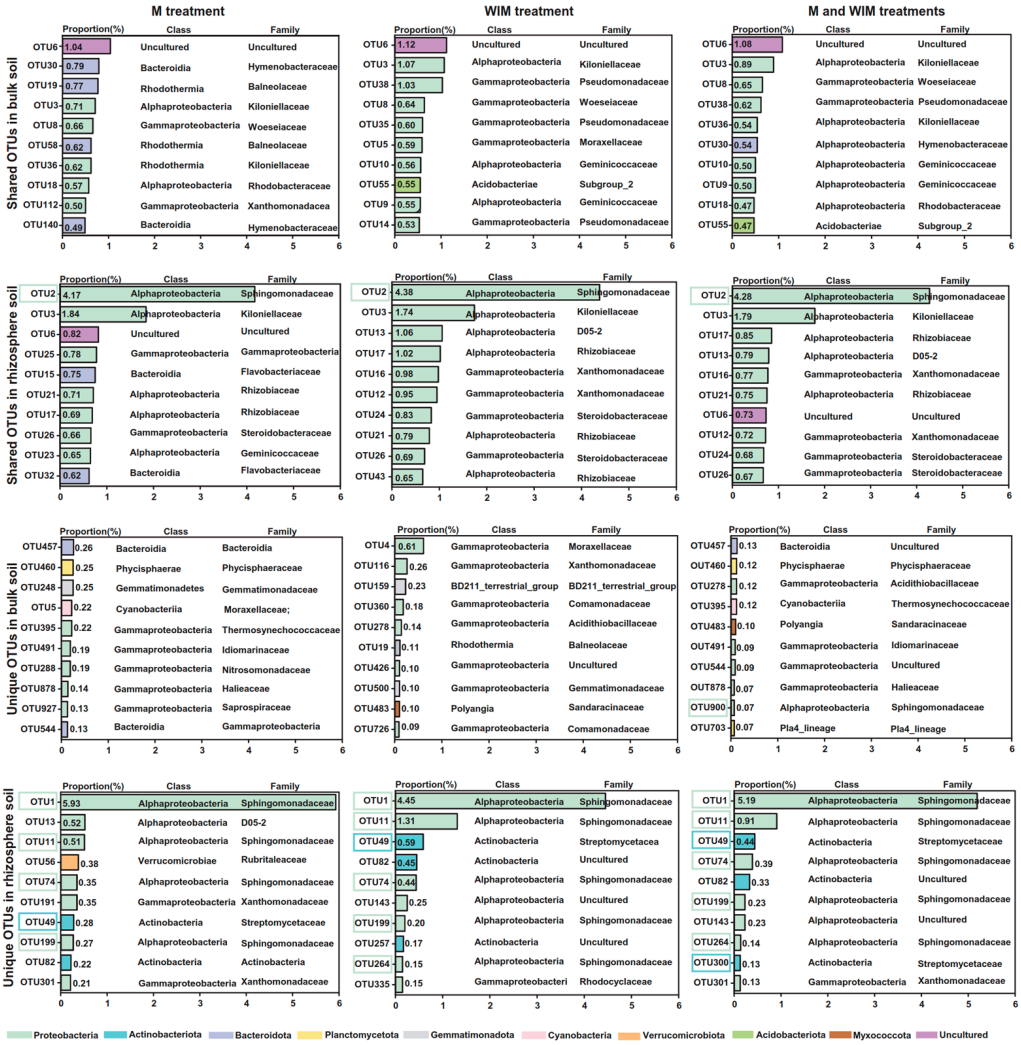

Figure 4. The OTUs of bacterial taxa that were shared and unique in bulk and rhizosphere soils of cotton in M and WIM treatments.

The distribution of shared and unique OTUs in the top 10 proportions of bacterial taxa in bulk and rhizosphere soils was analyzed (Figure 4). Among the shared bacterial taxa OTUs in bulk and rhizosphere soils, either individually or together in M and WIM treatments, the total proportion of the 10 top predominant OTUs of Proteobacteria in rhizosphere soil was greatly increased compared to that in bulk soil (Figure 4). The predominant OTUs of bacterial taxa of the 10 top proportions, including OUT49, OUT82, OUT257, and OTU300, were identified as Actinobacteria at the class level or Streptomycetaceae at the family level of phylum Actinobacteriota, which were unique to rhizosphere soil (Figure 4). In addition, the other predominant bacterial taxa OTUs of the 10 top proportions were identified as Sphingomonadaceae of the phylum Proteobacteria, of which OUT1, OUT11, OUT74, OUT199, and OTU264 were unique to rhizosphere soil, except for OTU2 that was found in bulk and rhizosphere soils, and OTU900 was unique to bulk soil (Figure 4).

Non-metric multidimensional scaling and the Bray–Curtis dissimilarity analysis revealed that both M and WIM treatments changed the assemblage of the microbial community compared to that of the control (Figures 5 and 6). The bacterial community in rhizosphere soil was assembled far from that in the control and bulk soils and behaved more specifically in rhizosphere soil than in bulk soil, and the difference between WIM and the control treatments was slightly higher than that between M and the control treatments (Figures 5 and 6). Two-dimensional plots assessed by NMDS ordination based on the Bray–Curtis distance showed that the bacterial communities in the rhizosphere soils in M

and WIM treatments clustered closely and were separate from those in bulk and control soils, whereas the bacterial communities in bulk and control soils were not clearly separated along the first axis (Figure 5). According to the Bray–Curtis dissimilarity of microbial communities in soil, higher dissimilarities in WIM_Bulk (0.597) and in WIM_Rhizosphere (0.781) were observed than those in M_Bulk (0.574) and in M_Rhizosphere (0.758) compared to that of the control, respectively, indicating that bacterial communities were influenced more by WIM treatment than by M treatment (Figure 6). In addition, the Bray–Curtis dissimilarity of soil microbial communities between rhizosphere and control soils was higher than that between bulk and control soils (Figure 6). Furthermore, the Bray–Curtis dissimilarity of microbial communities in bulk soils (M_Bulk and WIM_Bulk, 0.473) was higher than that in rhizosphere soils (M_Rhizosphere and WIM_Rhizosphere, 0.355), indicating that bacterial communities in bulk soil were influenced more by different amelioration practices than those in rhizosphere soil (Figure 6).

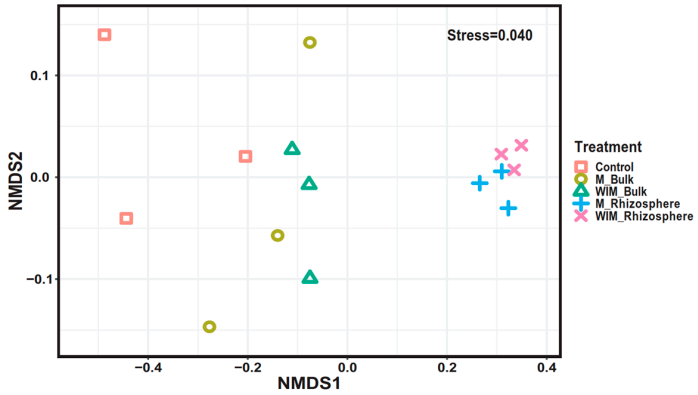

Figure 5. Non-metric multidimensional scaling (NMDS) ordination of bacterial communities in soil based on the Bray–Curtis distance. M—plastic mulching; WIM—the combination of freezing saline water irrigation and plastic mulching.

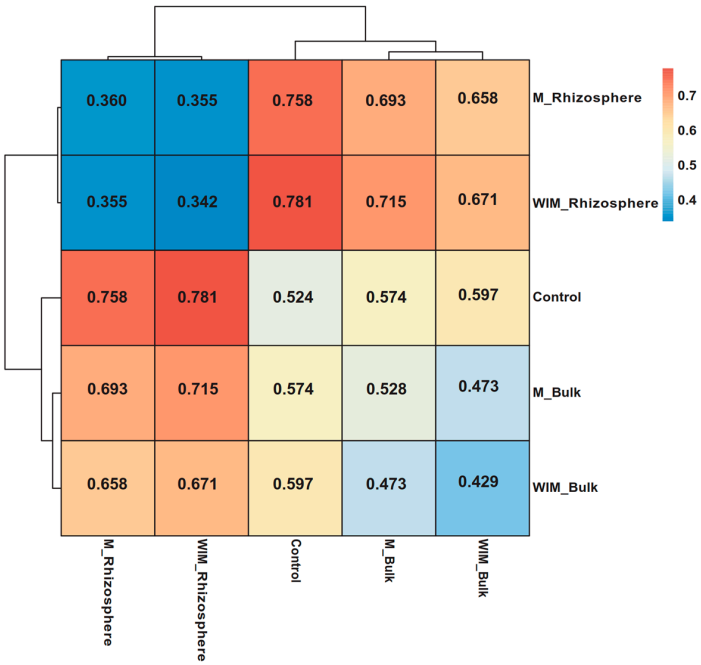

Figure 6. Heatmap showing the dissimilarity of bacterial communities between different treatments. The red color indicates high values and the blue color indicates low values. The higher the difference in value or color, the greater the difference in microbial community structure between treatments. M—plastic mulching; WIM—the combination of freezing saline water irrigation and plastic mulching.

3.4. Functional Prediction of Bacterial Community in Soil

The ecological functions of bacterial communities in rhizosphere and bulk soils of cotton under the amelioration practices M and WIM, as well as in control soil, were predicted using the FAPROTAX database. A total of 92 categories were identified, and the top 14 categories with high relative abundance (>1%) are shown in a heatmap (Figure 7). The functional groups of hydrocarbon degradation and aromatic compound degradation were dominant in the control soil (Figure 7; Table S2). The functional groups, including fermentation, nitrate reduction, aerobic chemoheterotrophy, chemoheterotrophy, chitinolysis, and cellulolysis were significantly elevated in the rhizosphere soil after both M and WIM treatments, whereas the functional groups, including human pathogens pneumonia, human pathogens all, human associated, and animal parasites or symbionts were significantly increased in the bulk soil after both M and WIM treatments (Figure 7; Table S2). Moreover, the photoautotrophy and phototrophy groups decreased in the rhizosphere soil compared to the control soil and bulk soil after both M and WIM treatments (Figure 7; Table S2).

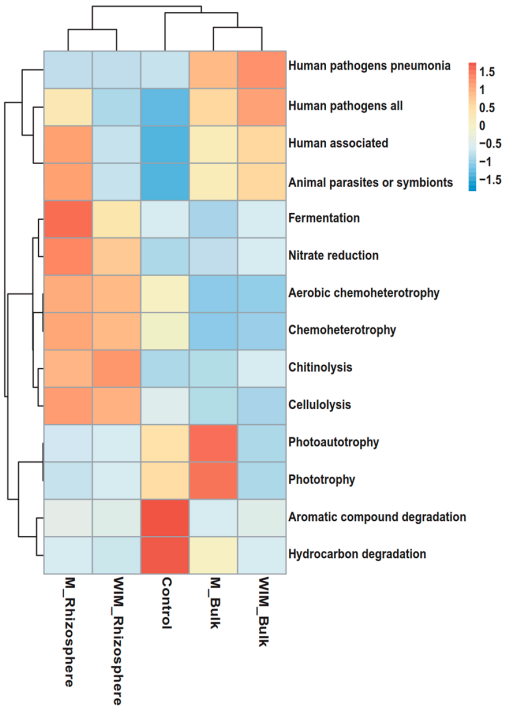

Figure 7. FAPROTAX clustering heatmap. M—plastic mulching; WIM—the combination of freezing saline water irrigation and plastic mulching.

4. Discussion

4.1. Effects of Different Amelioration Practices on the Bacterial Community in Coastal Saline Soils

Amelioration practices M and WIM are successful amelioration techniques that dramatically desalinize coastal saline soil, thereby alleviating the abiotic stress of salt on cotton seedling emergence and enabling a certain amount of production, with a higher level of desalinization at seedling stage and productivity under WIM practice than under M practice [14,19,20,22,33]. However, a high salt concentration inhibited cotton germination and productivity in the control treatment without amelioration practices M and WIM [14,20,22,33]. The salt content or EC of soil is an important environmental factor affecting plant growth and the soil microbial community [7,25,69]. Microenvironmental changes from plant litter and root exudates are important factors determining microbial richness, diversity, structure, and function [32]. Therefore, changes in microbial communities between amelioration treatments and control, as well as between WIM and M treatments in the present study, may have resulted mainly from the growth of cotton vegetation and reduced soil salinity, along with varying degrees of reduction under different amelioration

practices. Consistently, our previous study indicated that a decrease in salt concentration led to an environment suitable for diverse microorganisms, and the exogenous carbon inputs from plant growth resulted in changes in soil microbial communities in amelioration treatments [33,51]; greater changes in soil microbial communities were observed as a result of the WIM treatment compared with M [33].

4.2. Responses of Cotton Rhizosphere and Bulk Soil Bacterial Communities to Amelioration Practices in Coastal Saline Soils

The assembly of the rhizosphere microbial community is a dynamic and complicated process that is affected by various environmental factors, including the physicochemical properties (such as pH, salinity, and moisture) of soil, biological interactions (such as promotion, symbiosis, and competition), and the rates of birth–death, speciation–extinction, and migration of species [38,70,71]. Rhizosphere microorganisms are more affected by plants than bulk microorganisms, with the rhizosphere microbial community becoming increasingly plant-specific with the growth of plants, whereas the initial rhizosphere microbial community is similar to that in bulk soil [1,31,35,37,57,72]. Chen et al. [71] hypothesized that microbes in the rhizosphere of the plant *Cinnamomum migao* were mainly recruited via colonization from bulk soil microbial reservoir and were then filtered by the surplus carbon released by plant roots, which indicated that the rhizosphere microbiome with low diversity was more conserved than the bulk soil microbiome. Consistent with these findings, in the present study, Proteobacteria and Actinobacteriota were dramatically recruited from bulk soil to rhizosphere soil of cotton. Furthermore, the uniquely predominant bacterial groups of Actinobacteria and Streptomycetaceae belonging to Actinobacteriota in rhizosphere soil rather than in bulk soil might be filtered by carbon inputs from cotton roots (Figure 4), as Actinobacteria can effectively regulate the decomposition and synthesis of organic matter in soil and affect the carbon content in soil [73]. Therefore, the bacterial community behaved more specifically and was more conserved in rhizosphere soil than in bulk soil in this study, with lower richness and alpha diversity as well as lower distribution distance in rhizosphere soil than those in bulk soil (Figures 1, 5 and 6). These results may be largely attributed to the primary role of plants in the selective or filtering effect on microbial communities through root exudates, which play major roles in determining plant–microbe interactions in the rhizosphere [31,42,71,72,74]. Similar to our findings, bacterial diversity decreased in previous studies, even though rhizospheres recruited unique bacterial species for colonization [40,69]. A possible reason for this result is that the sequencing depth was finite, and as result, certain species with low abundance were missing. Another possible reason is that some unnecessary sequences, such as low quality, unclassified, chimeric sequences may have been discarded during the assemblage of the rhizosphere community based on the current sequencing depth, which has been confirmed by other studies [37,40,75,76].

4.3. Specific Bacterial Groups under Amelioration Practices in Coastal Saline Soils

Despite the high biodiversity of soils, Proteobacteria, Actinobacteria, and Firmicutes are the dominant bacterial phyla in microbial communities in the rhizosphere and endosphere of plants [31,76]. Proteobacteria and Actinobacteria are typical halophilic or halotolerant microbes that are widespread in the root microbiome and specific to root niches or plant vegetation, and have beneficial ecological significance in hypersaline regions [31,72,77,78]. Actinobacteria, which can produce up to 45% bioactive microbial metabolites, most of them halophilic groups, have received much attention in studies on microorganisms in saline soil [72,79]. In the present study, those unique Streptomycetaceae of Actinobacteriota and uniquely enriched Sphingomonadaceae of Proteobacteria in the rhizosphere soil were discovered to be beneficial plant growth-promoting bacteria (PGPB) that promote plant health (Figure 4), especially when the plants are exposed to abiotic or biotic stressors [31,40,76,80–82], which might improve the resistance of cotton to salt stress.

Many microorganisms have substantial beneficial effects on their plant host, improving their acquisition of nutrients and their resistance against abiotic stresses such as heat,

drought, and salinity [31,57]. Planctomycetes, Acidobacteria and Nitrspirota are oligotrophic organisms that prefer nutrient-poor environments and are capable of degrading recalcitrant carbon, whereas Proteobacteria prefer nutrient-rich environments [76,77,83]. In this study, the abundance of Proteobacteria significantly increased in rhizosphere soil but decreased in bulk soil, with opposite trends in the abundances of Planctomycetes, Acidobacteria and Nitrspirota (Figure 2), indicating a higher nutrient distribution in rhizosphere soil compared to that in bulk soil. According to the Functional Annotation of Prokaryotic Taxa, the functional groups of chemoheterotrophy and aerobic chemoheterotrophy greatly increased in rhizosphere soil but decreased in bulk soil, whereas the functional groups of photoautotrophy and phototrophy greatly decreased in rhizosphere soil (Figure 7). Similarly, we previously found that the functional groups of chemoheterotrophy and aerobic chemoheterotrophy decreased in ameliorated soils, which were considered bulk soils in this study [33]. Notably, this finding corresponded to the increased uniquely predominant bacterial groups of Actinobacteria at the class level and Streptomycetaceae at the family level belonging to Actinobacteriota in rhizosphere soil as compared to those in bulk soil, which was reported to be positively correlated with chemoheterotrophy and aerobic chemoheterotrophy involved in the carbon cycle [84]. Organisms in the families Streptosporangiaceae and Sphingomonadaceae can metabolize various carbon compounds, liberating utilizable carbon sources for other microorganisms [76,80,85]. In addition, functional groups of nitrate reduction greatly increased in rhizosphere soil but decreased in bulk soil in the present study, possibly owing to the presence of organisms of the Sphingomonadaceae family with an interesting blend of metabolic attributes and respiratory NO_3^- reduction [76,80]. Therefore, we speculated that the exogenous organic compound inputs resulting from cotton root metabolites recruited abundant and active heterotrophic microorganisms involved in nutrient cycles in the rhizosphere to support the adaptation of cotton to an ameliorated salt environment.

5. Conclusions

In this study, the responses of bacterial richness and diversity, community composition, and potential ecological functions in the rhizosphere and bulk soils of cotton to successful amelioration practices M and WIM in a coastal saline field were assessed. Compared to the control treatment, both the M and WIM treatments increased bacterial richness and alpha diversity and showed similar community compositions according to the relative abundances of the predominant bacterial taxa and the proportion of shared bacterial OTUs. In addition, bacterial communities in the rhizosphere and bulk soils of cotton responded differently to amelioration practices, with the bacterial community in rhizosphere soil assembled far from those in the control and bulk soils. The richness and alpha diversity of bacterial communities were in general significantly higher in bulk soil than in rhizosphere soil. The relative abundances of halophilic or halotolerant Proteobacteria and Actinobacteria decreased in bulk soil but significantly increased in rhizosphere soil. The functional groups of chemoheterotrophy, aerobic chemoheterotrophy, and nitrate reduction greatly increased in rhizosphere soil but decreased in bulk soil. These findings contributed to our understanding of the microbial community assembly process under coastal saline soil amelioration practices and provided valuable information for adapting management practices to facilitate microbe-based amelioration techniques in agricultural ecosystems.

Supplementary Materials: The following supporting information can be downloaded at: https://www.mdpi.com/article/10.3390/agronomy14010103/s1, Table S1: Relative abundances of the 15 top dominant bacterial taxa in different soil samples; Table S2: Comparisons of functional community profiles of bacteria in different treatments using FAPROTAX.

Author Contributions: X.W. (Xiaogai Wang) and L.W. (Luming Wang): experiment design, field sampling, data curation, formal analysis, writing—original draft; Z.Y. and Y.X.: writing—review and editing; Y.T.: writing—review and editing, project administration; K.G., L.W. (Lianfu Wu) and H.W.: field sampling, writing—review and editing; X.W. (Xinzhen Wang): data curation, formal analysis,

writing—review and editing, funding acquisition. All authors have read and agreed to the published version of the manuscript.

Funding: This work was supported by the National Key Research and Development Program of China (2022YFD190010204), the Science and Technology Project of Hebei Education Department (ZC2021012), the National Natural Science Foundation of China (41807058), the Natural Science Foundation of Hebei Province (C2021503002), and the National Key Research and Development Program of China (2021YFF1000403, 2018YFD0800306).

Data Availability Statement: The datasets presented in this study are available in the SRA database under the BioProject accession number PRJNA970964: https://www.ncbi.nlm.nih.gov/sra/PRJNA970964 (accessed on 10 May 2023).

Acknowledgments: We thank the staff from the research team led by Xiaojing Liu for their assistance in soil sampling, and Ruibo Sun for his assistance with the data analysis and writing ideas.

Conflicts of Interest: The authors declare that this research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

References

1. Qin, Y.; Druzhinina, I.S.; Pan, X.; Yuan, Z. Microbially mediated plant salt tolerance and microbiome-based solutions for saline agriculture. *Biotechnol. Adv.* **2016**, *34*, 1245–1259. [CrossRef] [PubMed]
2. Hossain, M.S. Present scenario of global salt affected soils, its management and importance of salinity research. *Int. J. Biol. Sci.* **2019**, *1*, 1–3.
3. Li, H.; Zhao, Q.; Huang, H. Current states and challenges of salt-affected soil remediation by cyanobacteria. *Sci. Total Environ.* **2019**, *669*, 258–272. [CrossRef] [PubMed]
4. Liu, F. Maize (*Zea mays*) growth and nutrient uptake following integrated improvement of vermicompost and humic acid fertilizer on coastal saline soil. *Appl. Soil Ecol.* **2019**, *142*, 147–154. [CrossRef]
5. Soldan, R.; Mapelli, F.; Crotti, E.; Schnell, S.; Daffonchio, D.; Marasco, R.; Fusi, M.; Borin, S.; Cardinale, M. Bacterial endophytes of mangrove propagules elicit early establishment of the natural host and promote growth of cereal crops under salt stress. *Microbiol. Res.* **2019**, *223–225*, 33–43. [CrossRef] [PubMed]
6. Etesami, H.; Glick, B.R. Halotolerant plant growth–promoting bacteria: Prospects for alleviating salinity stress in plants. *Environ. Exp. Bot.* **2020**, *178*, 104124. [CrossRef]
7. Li, H.; La, S.; Zhang, X.; Gao, L.; Tian, Y. Salt-induced recruitment of specific root-associated bacterial consortium capable of enhancing plant adaptability to salt stress. *ISME J.* **2021**, *15*, 2865–2882. [CrossRef]
8. Choudhary, M.; Jat, H.S.; Mukhopadhyay, R.; Kakraliya, M.; Poonia, T.; Phogat, A.; Dixit, B.; Kumar, R.; Arora, S.; Yadav, R.K.; et al. Functional diversity and behavioral changes of microbial communities under salt affected soils. *Appl. Soil Ecol.* **2023**, *190*, 105017. [CrossRef]
9. Li, Z.; Liu, X.; Zhang, X.; Li, W. Infiltration of melting saline ice water in soil columns: Consequences on soil moisture and salt content. Agric. *Water Manag.* **2008**, *95*, 498–502. [CrossRef]
10. Bharti, P.; Singh, B.; Bauddh, K.; Dey, R.K.; Korstad, J. Efficiency of bioenergy plant in phytoremediation of saline and sodic soil. In *Phytoremediation Potential of Bioenergy Plants*; Bauddh, K., Singh, B., Korstad, J., Eds.; Springer: Singapore, 2017; pp. 353–369.
11. Etesami, H.; Alikhani, H.A. Halotolerant plant growth-promoting fungi and bacteria as an alternative strategy for improving nutrient availability to salinity-stressed crop plants. In *Saline Soil-Based Agriculture by Halotolerant Microorganisms*; Kumar, M., Etesami, H., Kumar, V., Eds.; Springer: Singapore, 2019; pp. 103–146.
12. Zhang, W.; Wang, C.; Rui, X.; Wang, L. Effects of salinity on the soil microbial community and soil fertility. *J. Integr. Agric.* **2019**, *18*, 1360–1368. [CrossRef]
13. He, K.; He, G.; Wang, C.; Zhang, H.; Xu, Y.; Wang, S.; Kong, Y.; Zhou, G.; Hu, R. Biochar amendment ameliorates soil properties and promotes miscanthus growth in a coastal saline-alkali soil. *Appl. Soil Ecol.* **2020**, *155*, 103674. [CrossRef]
14. Zhang, X.; Guo, K.; Xie, Z.; Feng, X.; Liu, X. Effect of frozen saline water irrigation in winter on soil salt and water dynamics, germination and yield of cotton in coastal soils. *Chin. J. Eco-Agric.* **2012**, *20*, 1310–1314. [CrossRef]
15. Jing, C.; Xu, Z.; Zou, P.; Tang, Q.; Li, Y.; You, X.; Zhang, C. Coastal halophytes alter properties and microbial community structure of the saline soils in the yellow river delta, china. *Appl. Soil Ecol.* **2019**, *134*, 1–7. [CrossRef]
16. You, X.; Yin, S.; Suo, F.; Xu, Z.; Liu, L. Biochar and fertilizer improved the growth and quality of the ice plant (*Mesembryanthemum crystallinum* L.) shoots in a coastal soil of yellow river delta, China. *Sci. Total Environ.* **2021**, *775*, 144893. [CrossRef] [PubMed]
17. Cui, L.; Liu, Y.; Yan, J.; Hina, K.; Hussain, Q.; Qiu, T.; Zhu, J. Revitalizing coastal saline-alkali soil with biochar application for improved crop growth. *Ecol. Eng.* **2022**, *179*, 106594. [CrossRef]
18. Long, X.; Liu, L.; Shao, T.; Shao, H.; Liu, Z. Developing and sustainably utilize the coastal mudflat areas in China. *Sci. Total Environ.* **2016**, *569–570*, 1077–1086. [CrossRef] [PubMed]
19. Guo, K.; Liu, X. Dynamics of meltwater quality and quantity during saline ice melting and its effects on the infiltration and desalinization of coastal saline soils. *Agric. Water Manag.* **2014**, *139*, 1–6. [CrossRef]

20. Li, X.; Guo, K.; Feng, X.; Liu, H.; Liu, X. Soil respiration response to long-term freezing saline water irrigation with plastic mulching in coastal saline plain. *Sustainability* **2017**, *9*, 621. [CrossRef]
21. Sadegh-Zadeh, F.; Seh-Bardan, B.J.; Samsuri, A.W.; Mohammadi, A.; Chorom, M.; Yazdani, G.A. Saline soil reclamation by means of layered mulch. *Arid Land Res. Manag.* **2009**, *23*, 127–136. [CrossRef]
22. Guo, K.; Liu, X. Infiltration of meltwater from frozen saline water located on the soil can result in reclamation of a coastal saline soil. *Irrig. Sci.* **2015**, *33*, 441–452. [CrossRef]
23. Aislabie, J.; Deslippe, J.R. Soil microbes and their contribution to soil services. In *Ecosystem Services in New Zealand–Conditions and Trends*; Dymond, J.R., Ed.; Manaaki Whenua Press: Lincoln, New Zealand, 2013; pp. 143–161.
24. Epelde, L.; Burges, A.; Mijangos, I.; Garbisu, C. Microbial properties and attributes of ecological relevance for soil quality monitoring during a chemical stabilization field study. *Appl. Soil Ecol.* **2014**, *75*, 1–12. [CrossRef]
25. Ma, J.; Ibekwe, A.M.; Yang, C.; Crowley, D.E. Bacterial diversity and composition in major fresh produce growing soils affected by physiochemical properties and geographic locations. *Sci. Total Environ.* **2016**, *563–564*, 199–209. [CrossRef] [PubMed]
26. Ruppel, S.; Franken, P.; Witzel, K. Properties of the halophyte microbiome and their implications for plant salt tolerance. *Funct. Plant Biol.* **2013**, *40*, 940–951. [CrossRef] [PubMed]
27. van der Heijden, M.G.A.; Wagg, C. Soil microbial diversity and agro-ecosystem functioning. *Plant Soil* **2013**, *363*, 1–5. [CrossRef]
28. Hartman, K.; Van Der Heijden, M.G.A.; Wittwer, R.A.; Banerjee, S.; Walser, J.C.; Schlaeppi, K. Cropping practices manipulate abundance patterns of root and soil microbiome members paving the way to smart farming. *Microbiome* **2018**, *6*, 14.
29. Somenahally, A.C.; McLawrence, J.; Chaganti, V.N.; Ganjegunte, G.K.; Obayomi, O.; Brady, J.A. Response of soil microbial Communities, inorganic and organic soil carbon pools in arid saline soils to alternative land use practices. *Ecol. Indic.* **2023**, *150*, 110227. [CrossRef]
30. Bastida, F.; Moreno, J.L.; Hernández, T.; García, C. Microbiological degradation index of soils in a semiarid climate. *Soil Biol. Biochem.* **2006**, *38*, 3463–3473. [CrossRef]
31. Rodriguez, P.A.; Rothballer, M.; Chowdhury, S.P.; Nussbaumer, T.; Gutjahr, C.; Falter-Braun, P. Systems biology of plant-microbiome interactions. *Mol. Plant* **2019**, *12*, 804–821. [CrossRef]
32. Liu, F.; Mo, X.; Kong, W.; Song, Y. Soil bacterial diversity, structure, and function of suaeda salsa in rhizosphere and non-rhizosphere soils in various habitats in the yellow river delta, china. *Sci. Total Environ.* **2020**, *740*, 140144. [CrossRef]
33. Sun, R.; Wang, X.; Tian, Y.; Guo, K.; Feng, X.; Sun, H.; Liu, X.; Liu, B. Long-term amelioration practices reshape the soil microbiome in a coastal saline soil and alter the richness and vertical distribution differently among bacterial, archaeal, and fungal communities. *Front. Microbiol.* **2022**, *12*, 768203. [CrossRef]
34. Berg, G. Plant-microbe interactions promoting plant growth and health: Perspectives for controlled use of microorganisms in agriculture. *Appl. Microbiol. Biot.* **2009**, *84*, 11–18. [CrossRef] [PubMed]
35. Chen, L.; Brookes, P.C.; Xu, J.; Zhang, J.; Zhang, C.; Zhou, X.; Luo, Y. Structural and functional differentiation of the root-associated bacterial microbiomes of perennial ryegrass. *Soil Biol. Biochem.* **2016**, *98*, 1–10. [CrossRef]
36. Mahmud, K.; Missaoui, A.; Lee, K.; Ghimire, B.; Presley, H.W.; Makaju, S. Rhizosphere microbiome manipulation for sustainable crop production. *Curr. Plant Biol.* **2021**, *27*, 100210. [CrossRef]
37. Ling, N.; Wang, T.T.; Kuzyakov, Y. Rhizosphere bacteriome structure and functions. *Nat. Commun.* **2022**, *13*, 836. [CrossRef] [PubMed]
38. Grover, M.; Ali, S.Z.; Sandhya, V.; Rasul, A.; Venkateswarlu, B. Role of microorganisms in adaptation of agriculture crops to abiotic stresses. *World J. Microb. Biot.* **2010**, *27*, 1231–1240. [CrossRef]
39. Berendsen, R.L.; Pieterse, C.M.J.; Bakker, P.A.H.M. The rhizosphere microbiome and plant health. *Trends Plant Sci.* **2012**, *17*, 478–486. [CrossRef] [PubMed]
40. Shi, Y.; Pan, Y.; Xiang, L.; Zhu, Z.; Fu, W.; Hao, G.; Geng, Z.; Chen, S.; Li, Y.; Han, D. Assembly of rhizosphere microbial communities in *Artemisia annua*: Recruitment of plant growth promoting microorganisms and inter-kingdom interactions between bacteria and fungi. *Plant Soil* **2021**, *470*, 127–139. [CrossRef]
41. Berg, G.; Smalla, K. Plant species and soil type cooperatively shape the structure and function of microbial communities in the rhizosphere. *FEMS Microbiol. Ecol.* **2009**, *68*, 1–13. [CrossRef]
42. Nie, M.; Zhang, X.; Wang, J.; Jiang, L.; Yang, J.; Quan, Z.; Cui, X.; Fang, C.; Li, B. Rhizosphere effects on soil bacterial abundance and diversity in the yellow river deltaic ecosystem as influenced by petroleum contamination and soil salinization. *Soil Biol. Biochem.* **2009**, *41*, 2535–2542. [CrossRef]
43. Lu, T.; Ke, M.; Lavoie, M.; Jin, Y.; Fan, X.; Zhang, Z.; Fu, Z.; Sun, L.; Gillings, M.; Peñuelas, J. Rhizosphere microorganisms can influence the timing of plant flowering. *Microbiome* **2018**, *6*, 1–12. [CrossRef]
44. Jiang, X.; Li, X.; Yang, L.; Liu, C.; Wang, Q.; Chi, W.; Zhu, H. How microbes shape their communities? A microbial community model based on functional genes. *Genom. Proteom. Bioinf.* **2019**, *17*, 91–105. [CrossRef] [PubMed]
45. Ning, K.; Tong, Y. The fast track for microbiome research. *Genom. Proteom. Bioinf.* **2019**, *17*, 1–3. [CrossRef] [PubMed]
46. Di Bella, J.M.; Bao, Y.; Gloor, G.B.; Burton, J.P.; Reid, G. High throughput sequencing methods and analysis for microbiome research. *J. Microbiol. Meth.* **2013**, *95*, 401–414. [CrossRef] [PubMed]
47. Caporaso, J.G.; Kuczynski, J.; Stombaugh, J.; Bittinger, K.; Bushman, F.D.; Costello, E.K.; Fierer, N.; Peña, A.G.; Goodrich, J.K.; Gordon, J.I.; et al. QIIME allows analysis of high-throughput community sequencing data. *Nat. Methods* **2010**, *7*, 335–336. [CrossRef] [PubMed]

48. Zhou, Y.; Yang, Z.; Liu, J.; Li, X.; Wang, X.; Dai, C.; Zhang, T.; Carrión, V.J.; Wei, Z.; Cao, F.; et al. Crop rotation and native microbiome inoculation restore soil capacity to suppress a root disease. *Nat. Commun.* **2023**, *14*, 8126. [CrossRef]
49. Wang, F.; Chen, S.; Wang, Y.; Zhang, Y.; Hu, C.; Liu, B. Long-term nitrogen fertilization elevates the activity and abundance of nitrifying and denitrifying microbial communities in an upland soil: Implications for nitrogen loss from intensive agricultural systems. *Front. Microbiol.* **2018**, *9*, 2424. [CrossRef]
50. Sun, R.; Li, W.; Dong, W.; Tian, Y.; Hu, C.; Liu, B. Tillage changes vertical distribution of soil bacterial and fungal communities. *Front. Microbiol.* **2018**, *9*, 699. [CrossRef]
51. Wang, X.; Sun, R.; Tian, Y.; Guo, K.; Sun, H.; Liu, X.; Chu, H.; Liu, B. Long-term phytoremediation of coastal saline soil reveals plant species-specific patterns of microbial community recruitment. *mSystems* **2020**, *5*, e00741-19. [CrossRef]
52. Ren, H.; Zhang, F.; Zhu, X.; Lamlom, S.F.; Zhao, K.; Zhang, B.; Wang, J. Manipulating rhizosphere microorganisms to improve crop yield in saline-alkali soil: A study on soybean growth and development. *Front. Microbiol.* **2023**, *14*, 1233351. [CrossRef]
53. Ahmed, V.; Verma, M.K.; Gupta, S.; Mandhan, V.; Chauhan, N.S. Metagenomic profiling of soil microbes to mine salt stress tolerance genes. *Front. Microbiol.* **2018**, *9*, 159. [CrossRef]
54. Khan, M.A.; Asaf, S.; Khan, A.L.; Jan, R.; Kang, S.-M.; Kim, K.-M.; Lee, I.-J. Rhizobacteria AK1 remediates the toxic effects of salinity stress via regulation of endogenous phytohormones and gene expression in soybean. *Biochem. J.* **2019**, *476*, 2393–2409. [CrossRef] [PubMed]
55. Abulfaraj, A.A.; Jalal, R.S. Use of plant growth-promoting bacteria to enhance salinity stress in soybean (*Glycine max* L.) plants. *Saudi J. Biol. Sci.* **2021**, *28*, 3823–3834. [CrossRef] [PubMed]
56. Guo, K.; Zhang, X.; Li, X.; Liu, X. Effect of freezing saline water irrigation in winter on the reclamation of coastal saline soil. *Resour. Sci.* **2010**, *32*, 431–435.
57. Edwards, J.; Johnson, C.; Santos-Medellín, C.; Lurie, E.; Podishetty, N.K.; Bhatnagar, S.; Eisen, J.A.; Sundaresan, V. Structure, variation, and assembly of the root-associated microbiomes of rice. *Proc. Natl. Acad. Sci. USA* **2015**, *112*, 1414592112. [CrossRef] [PubMed]
58. Armstrong, F.A.J. Determination of nitrate in water ultraviolet spectrophotometry. *Anal. Chem.* **1963**, *35*, 1292–1294. [CrossRef]
59. Crouch, S.R.; Malmstadt, H.V. A mechanistic investigation of molybdenum blue method for determination of phosphate. *Anal. Chem.* **1967**, *39*, 1084–1089. [CrossRef]
60. Shaw, K. Determination of organic carbon in soil and plant material. *Eur. J. Soil Sci.* **1959**, *10*, 316–326. [CrossRef]
61. Walters, W.; Hyde, E.R.; Berg-Lyons, D.; Ackermann, G.; Humphrey, G.; Parada, A. Improved bacterial 16s rRNS gene (V4 and V4-5) and fungal internal transcribed spacer marker gene primers for microbial community surveys. *mSystems* **2016**, *1*, e00009-15. [CrossRef]
62. Sun, R.; Ding, J.; Li, H.; Wang, X.; Li, W.; Li, K.; Ye, X.; Sun, S. Mitigating nitrate leaching in cropland by enhancing microbial nitrate transformation through the addition of liquid biogas slurry. *Agric. Ecosyst. Environ.* **2023**, *345*, 108324. [CrossRef]
63. Martin, M. Cutadapt removes adapter sequences from high-throughput sequencing reads. *EMBnet J.* **2011**, *17*, 10–12. [CrossRef]
64. Sun, R.; Chen, Y.; Han, W.; Dong, W.; Zhang, Y.; Hu, C.; Liu, B.; Wang, F. Different contribution of species sorting and exogenous species immigration from manure to soil fungal diversity and community assemblage under long-term fertilization. *Soil Biol. Biochem.* **2020**, *151*, 108049. [CrossRef]
65. Edgar, R.C.; Haas, B.J.; Clemente, J.C.; Quince, C.; Knight, R. UCHIME improves sensitivity and speed of chimera detection. *Bioinformatics* **2011**, *27*, 2194–2200. [CrossRef]
66. Glöckner, F.O.; Yilmaz, P.; Quast, C.; Gerken, J.; Beccati, A.; Ciuprina, A.; Bruns, G.; Yarza, P.; Peplies, J.; Westram, R.; et al. 25 years of serving the community with ribosomal RNA gene reference databases and tools. *J. Biotechnol.* **2017**, *261*, 169–176. [CrossRef] [PubMed]
67. Louca, S.; Parfrey, L.W.; Doebeli, M. Decoupling function and taxonomy in the global ocean microbiome. *Science* **2016**, *353*, 1272–1277. [CrossRef] [PubMed]
68. Corder, G.W.; Foreman, D.I. Comparing more than two unrelated samples: The Kruskal–Wallis H-Test. In *Nonparametric Statistics for Non-Statisticians*; Hoboken, N.J., Ed.; John Wiley and Sons, Inc.: Hoboken, NJ, USA, 2009; pp. 99–121.
69. Zheng, Y.; Xu, Z.; Liu, H.; Liu, Y.; Zhou, Y.; Meng, C.; Ma, S.; Xie, Z.; Li, Y.; Zhang, C. Patterns in the microbial community of salt-tolerant plants and the functional genes associated with salt stress alleviation. *Microbiol. Spectr.* **2021**, *9*, e0076721. [CrossRef] [PubMed]
70. Dini-Andreote, F.; Stegen, J.C.; van Elsas, J.D.; Salles, J.F. Disentangling mechanisms that mediate the balance between stochastic and deterministic processes in microbial succession. *Proc. Natl. Acad. Sci. USA* **2015**, *112*, 1414261112. [CrossRef] [PubMed]
71. Chen, J.; Huang, X.; Sun, Q.; Liu, J. Bulk soil microbial reservoir or plant recruitment dominates rhizosphere microbial community assembly: Evidence from the rare, endangered Lauraceae species Cinmaomum migao. *Ecol. Indic.* **2023**, *148*, 110071. [CrossRef]
72. Yadav, A.N.; Kaur, T.; Kour, D.; Rana, K.L.; Yadav, N.; Rastegari, A.A.; Kumar, M.; Paul, D.; Sachan, S.G.; Saxena, A.K. Saline microbiome: Biodiversity, ecological significance, and potential role in amelioration of salt stress. In *New and Future Developments in Microbial Biotechnology and Bioengineering*; Elsevier: Amsterdam, The Netherlands, 2020; pp. 283–309.
73. Lozano, Y.M.; Hortal, S.; Armas, C.; Pugnaire, F.I. Interactions among soil, plants, and microorganisms drive secondary succession in a dry environment. *Soil Biol. Biochem.* **2014**, *78*, 298–306. [CrossRef]
74. Bais, H.P.; Weir, T.L.; Perry, L.G.; Gilroy, S.; Vivanco, J.M. The role of root exudates in rhizosphere interactions with plants and other organisms. *Annu. Rev. Plant Biol.* **2006**, *57*, 233–266. [CrossRef]

75. Matthews, A.; Pierce, S.; Hipperson, H.; Raymond, B. Rhizobacterial community assembly patterns vary between crop species. *Front. Microbiol.* **2019**, *10*, 581. [CrossRef]
76. Luo, T.; Min, T.; Ru, S.; Li, J. Response of cotton root growth and rhizosphere soil bacterial communities to the application of acid compost tea in calcareous soil. *Appl. Soil Ecol.* **2022**, *177*, 104523. [CrossRef]
77. Canfora, L.; Bacci, G.; Pinzari, F.; Lo Papa, G.; Dazzi, C.; Benedetti, A. Salinity and bacterial diversity: To what extent does the concentration of salt affect the bacterial community in a saline soil? *PLoS ONE* **2014**, *9*, e106662. [CrossRef] [PubMed]
78. Santos, S.S.; Rask, K.A.; Vestergård, M.; Johansen, J.L.; Priemé, A.; Frøslev, T.G.; Martín González, A.M.; He, H.; Ekelund, F. Specialized microbiomes facilitate natural rhizosphere microbiome interactions counteracting high salinity stress in plants. *Environ. Exp. Bot.* **2021**, *186*, 104430. [CrossRef]
79. Maithani, D.; Sharma, A.; Gangola, S.; Chaudhary, P.; Bhatt, P. Insights into applications and strategies for discovery of microbial bioactive metabolites. *Microbiol. Res.* **2022**, *261*, 127053. [CrossRef] [PubMed]
80. Anderson, C.R.; Condron, L.M.; Clough, T.J.; Fiers, M.; Stewart, A.; Hill, R.A.; Sherlock, R.R. Biochar induced soil microbial community change: Implications for biogeochemical cycling of carbon, nitrogen and phosphorus. *Pedobiologia* **2011**, *54*, 309–320. [CrossRef]
81. Viaene, T.; Langendries, S.; Beirinckx, S.; Maes, M.; Goormachtig, S. *Streptomyces* as a plant's best friend? *FEMS Microbiol. Ecol.* **2016**, *92*, 119. [CrossRef] [PubMed]
82. Nozar, R.M.; Ramos, L.M.; da Luz, L.A.; Almeida, R.N.; Lucas, A.M.; Cassel, E.; de Oliveira, S.D.; Astarita, L.V.; Santarém, E.R. Halotolerant *Streptomyces* spp. induce salt tolerance in maize through systemic induction of the antioxidant system and accumulation of proline. *Rhizosphere* **2022**, *24*, 100623. [CrossRef]
83. Yang, W.; Cai, A.; Wang, J.; Luo, Y.; Cheng, X.; An, S. Exotic spartina alterniflora Loisel. invasion significantly shifts soil bacterial communities with the successional gradient of saltmarsh in eastern China. *Plant Soil* **2020**, *449*, 97–115. [CrossRef]
84. Wu, Z.; Lyu, H.; Liang, W.; Jing, X.; Wang, Y.; Ma, X. Microbial community in indoor dusts from university dormitories: Characteristics, potential pathogens and influence factors. *Atmos. Pollut. Res.* **2021**, *12*, 321–333. [CrossRef]
85. Schlatter, D.C.; DavelosBaines, A.L.; Xiao, K.; Kinkel, L.L. Resource use of soilborne streptomyces varies with location, phylogeny, and nitrogen amendment. *Microb. Ecol.* **2013**, *66*, 961–971. [CrossRef]