

Article

Effect of an Organic Fertilizer of *Ganoderma lucidum* Residue on the Physical and Chemical Properties and Microbial Communities of Saline Alkaline Soil

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Abstract: Saline-alkali land is the main reserve soil resource. The amount of arable farmland soil can be increased through improvement and utilization of saline-alkali land. Bio-organic fertilizer is an effective saline-alkali soil conditioner, but there are few studies on the improvement of saline-alkali soil with traditional Chinese medicine residues (TCMRs). In this study, an organic fertilizer made from *Ganoderma lucidum* residue (GLR) was mixed with saline-alkali soil at different proportions; physicochemical properties, enzyme activity, and microbial community characteristics of the soil were investigated. The results showed that the soil pH, as well as bulk density and electrical conductivity were significantly reduced, while the soil moisture content and porosity were significantly increased after the GLR organic fertilizer incorporation. Soil invertase and soil amylase activities significantly increased, as well as the diversity and richness of the soil microbial community structure. The abundance of the dominant phyla, Bacteroidota, Actinobacteriota, Chloroflexi, Firmicutes, and Basidiomycota, increased, while the abundance of the dominant Proteobacteria and Ascomycota phyla decreased. The best improvement effect is obtained when the application ratio of GLR organic fertilizer is 25%. The findings showed that TCMRs have positive application prospects in saline-alkali soil improvement.

Keywords: saline-alkali soil; high-throughput sequencing; soil physical and chemical properties; soil enzyme activity; bacterial community



Citation: Deng, P.-B.; Guo, L.-P.; Yang, H.-T.; Leng, X.-Y.; Wang, Y.-M.; Bi, J.; Shi, C.-F. Effect of an Organic Fertilizer of *Ganoderma lucidum* Residue on the Physical and Chemical Properties and Microbial Communities of Saline Alkaline Soil. *Water* **2023**, *15*, 962. <https://doi.org/10.3390/w15050962>

Academic Editor: Helvi Heinonen-Tanski

Received: 30 January 2023
Revised: 23 February 2023
Accepted: 27 February 2023
Published: 2 March 2023



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1. Introduction

Improvements of saline-alkali soil are aimed at increasing the area of arable land and increasing crop yield [1]. However, the existing methods of saline-alkali land treatment have the problem of “treating the symptoms but not its cause”. This will lead to the alternation of “desalting” and “returning salt”, which will cause much interference with the farmland soil ecology (microbial community) [2]. Advancement of science and technology has aided our understanding of saline-alkali soil remediation. At the same time, biological soil improvement methods, such as the use of organic fertilizers, have received widespread attention [3]. Bio-organic fertilizers (microbial organic fertilizers) have the advantages of traditional organic fertilizers and are enriched with added bacterial agents. As such, these soil amendments contain a large amount of organic matter together with beneficial soil microorganisms. Traditional Chinese medicine residues (TCMRs) are rich in cellulose, polysaccharide, and soil nutrients, such as organic carbon, nitrogen, and phosphorus [4,5]. They can be used as raw material for producing bio-organic fertilizer. The bacteria and toxic substances contained in fresh residue can be completely eliminated by high-temperature aerobic fermentation. In this way, the harmless treatment of TCMRs can be realized [6].

Studies have shown that biosolids play a positive role in improving soil properties, promoting the healthy growth of plants, and increasing the diversity of beneficial microorganisms [7,8]. Some studies have shown that TCMRs have beneficial effects on saline-alkali soil improvement. Through the application of Mailuoning TCMRs in combination with inorganic fertilizer and biological bacterial fertilizer, the soil enzyme activity was improved, the formation of granular structure in raw soil was promoted, and the raw soil was rapidly matured. The effect is better than that of chicken manure [9]. Bai et al. [10] applied Chinese medicinal residues, such as *Sophora japonica*, to the arid and abandoned saline-alkali soil in Ningxia, and found that the numbers of bacteria, fungi, and actinomycetes in the saline-alkali soil after the residue application were significantly higher than those in the control. The activity of alkaline phosphatase and catalase was significantly promoted. Bai et al. [11] conducted a black soil fertilizer application experiment and found that the application of organic fertilizer increased the microbial biomass carbon/nitrogen and the number of fungi and bacteria. Li et al. [12] studied the relationship between bio-organic fertilizers and nutrients in saline-alkali soil and rhizosphere microorganisms. The results showed that the content of available phosphorus and potassium in the saline-alkali soil after the fertilization treatment increased to varying degrees, and the microbial biomass of the plant roots increased. Ma et al. [13] found that organic fertilizer from TCMRs improved the soil fertility, especially in terms of organic matter, nitrogen, and phosphorus. The application rates (0.2–0.6%) were beneficial for improving the physiological parameters of maize seedlings. However, there is very limited research on the improvement of saline-alkali soil by organic fertilizers derived from GLR.

This study researched the use of GLR to (i) explore the effect of the organic fertilizer of GLR on the physical and chemical properties, enzyme activity and microbial community structure of saline-alkali soil, (ii) provide a theoretical basis for the improvement of saline-alkali soil with organic fertilizers from TCMRs, and (iii) open up a new way for the resource utilization of TCMRs. The results of this experiment can provide reference for the development and utilization of saline-alkali soil in arid and semi-arid areas, and the high-value utilization of organic solid wastes, such as TCMRs.

2. Materials and Methods

2.1. Experimental Materials

GLR: GLR was provided by the Baotou Traditional Chinese Medicine Co., Ltd. (Baotou, China).

GLR organic fertilizer: After the GLR was naturally dried in the shade and pulverized, the carbon-nitrogen ratio was 35/1, and the water content was adjusted to 65%. The nano-compost covering film was naturally fermented at room temperature for 42 days. The physical and chemical parameters of compost products were determined in the Environmental Biotechnology Laboratory of Inner Mongolia University of Science and Technology. Total humus (HS) was 37.56%, organic matter content was 79.84%, pH was 4.79, moisture content was 46.12%, and electrical conductance was 1.25 $\mu\text{S}/\text{cm}$.

Saline-alkali soil: Soil samples were collected from Dabusutai Village in the town of Haya Hutong, Jiuyuan District, Baotou City, Inner Mongolia (109°26′22.22″ E–109°26′24.78″ E, 40°31′57.33″ N–40°31′58.3″ N). Soil samples from the 0–20 cm layer of treated sample plots were dried in the shade and passed through a 3 mm sieve for analysis.

2.2. Experimental Method

2.2.1. Experimental Design

The experiment of improving saline-alkali soil with GLR organic fertilizer was carried out by indoor pot planting. A total of 6 treatments were set up by adding a GLR organic fertilizer in the following proportions: 0% (FSF), 8.3% (FSE), 10% (FSD), 12.5% (FSC), 16.7% (FSB), and 25% (FSA). Each treatment was repeated 5 times for a total of 30 pots. The plastic flowerpots were 22 cm in diameter and 28 cm in height.

2.2.2. Determination of Soil Physical and Chemical Properties

Soil physical and chemical properties were determined according to Lu (2000) [14]. The main measured indicators were soil bulk density, soil particle density, soil porosity, moisture content, pH, electrical conductance, organic matter content, effective phosphorus content, ammonium nitrogen, hydrolyzed nitrogen. The $\text{pH}_{(1:5)}$ value was determined using a calibrated pH meter (PHSJ-4F, Leici, Shanghai, China). Electrical conductance was determined using a conductivity meter (DDSJ-318T, Leici, Shanghai, China).

2.2.3. Determination of Soil Enzyme Activity

The 3,5-dinitrosalicylic acid colorimetric method was used to determine invertase activity and amylase activity [15]. The sodium phenate-sodium hypochlorite titration colorimetric method was used to determine urease activity [16]. Catalase activity was determined by potassium permanganate colorimetric titration [17]. Acid phosphomonoesterase activity was determined by the disodium phenylphosphate colorimetric method [18].

2.2.4. Determination of Soil Microbial Community Structure

Microbial community analysis was performed by the Shanghai Meiji Biomedical Technology Co., Ltd. (Shanghai, China). Sequencing of the V4–V5 region of the 16S rRNA gene of bacteria and ITS1 + ITS2 region of fungi was done on an Illumina MiSeq platform. Corresponding regions of primers, 16SV4 + V5 region primers (515F and 806R), were used to identify the bacterial diversity, and ITS1 + ITS2 region primers (ITS5-1737F and ITS2-2043R) were used to identify fungal diversity.

2.2.5. Data Analysis

Qiime1.9.1 sequencing was used to analyze microbial-community-related indicators. The experimental data were processed and analyzed with Excel and SPSS Statistics 23.0 (IBM, Armonk, New York, NY, USA). The parameters between different treatments were analyzed using a one-way analysis of variance (ANOVA) with Tukey's post hoc test at the level of $p < 0.05$. The relationships between the main phylum of the fungal and bacterial communities and environmental factors were analyzed by redundancy analysis (RDA).

3. Results and Discussion

3.1. Effects of the GLR Organic Fertilizer on the Physicochemical Properties and Enzyme Activity of Saline-Alkali Soil

3.1.1. Changes in Physicochemical Properties of Saline-Alkali Soil

Soil bulk density, porosity, and particle density are important indicators of soil physical properties. Their values can better reflect soil texture, permeability, water-holding capacity, and erosion resistance [10]. Table 1A shows that the addition of the GLR organic fertilizer significantly increased the water content and porosity of the saline-alkali soil and decreased the soil bulk density and soil particle density. Among them, the water content and porosity of the FSA treatment were higher than those of the other treatments, which were significantly higher than those of the FSF treatment. The soil particle density and bulk density were significantly lower than those of the FSF treatment.

The pH value can reflect the pH value of the soil, and the electrical conductance usually reflects the soluble salt content of the soil. Organic matter is also the main source of soil nutrients [19]. Table 1B shows that the addition of the GLR organic fertilizer significantly increased the contents of available phosphorus, alkali-hydrolysable nitrogen, organic matter, and ammonium nitrogen in the saline-alkali soil and reduced the pH value and electrical conductance of the saline-alkali soil. Among them, the effect of FSA and FSB treatment is the most significant.

Table 1. Physical (A) and chemical (B) properties of saline-alkali soil improved by adding a GLR organic fertilizer in different proportions. For each row, the means \pm SD with different letters were significantly different between treatments ($p < 0.05$, using the HSD test).

(A)	Sample					
	FSA (25%)	FSB (16.7%)	FSC (12.5%)	FSD (10%)	FSE (8%)	FSF (0%)
ρ_b (g/cm ³)	0.449 \pm 0.006 d	0.579 \pm 0.005 cd	0.582 \pm 0.012 cd	0.736 \pm 0.017 bc	0.891 \pm 0.013 b	1.434 \pm 0.154 a
W (%)	25.164 \pm 1.834 a	20.772 \pm 0.401 ab	18.140 \pm 0.397 bc	17.011 \pm 0.571 c	11.667 \pm 0.457 d	6.464 \pm 0.057 e
ρ_s (g/cm ³)	1.813 \pm 0.779 c	2.022 \pm 0.082 bc	2.085 \pm 0.060 b	2.452 \pm 0.054 a	2.539 \pm 0.042 a	2.562 \pm 0.054 a
f	0.751 \pm 0.014 a	0.713 \pm 0.011 ab	0.720 \pm 0.011 ab	0.699 \pm 0.007 ab	0.648 \pm 0.007 b	0.440 \pm 0.058 c
(B)	FSA (25%)	FSB (16.7%)	FSC (12.5%)	FSD (10%)	FSE (8%)	FSF (0%)
P (mg/kg)	36.86 \pm 0.04 a	28.78 \pm 0.04 b	21.51 \pm 0.03 c	19.78 \pm 0.03 d	18.38 \pm 0.03 e	15.15 \pm 0.05 f
AHN (g/kg)	11.70 \pm 0.62 a	7.90 \pm 0.23 b	7.466 \pm 0.84 b	6.5 \pm 0.84 bc	4.9 \pm 0.70 c	1.63 \pm 0.47 d
C (g/kg)	43.24 \pm 0.41 a	39.26 \pm 0.37 b	28.43 \pm 0.47 c	26.68 \pm 0.60 d	13.79 \pm 1.76 e	5.42 \pm 0.37 f
AN (mg/kg)	10.66 \pm 0.05 a	10.08 \pm 0.08 a	8.82 \pm 0.31 b	6.99 \pm 0.27 c	6.93 \pm 0.38 c	4.86 \pm 0.08 d
pH	7.51 \pm 0.02 f	7.78 \pm 0.02 e	7.86 \pm 0.03 d	8.11 \pm 0.02 c	8.23 \pm 0.01 b	8.98 \pm 0.02 a
EC (mS/cm)	6.55 \pm 0.02 d	7.37 \pm 0.07 c	7.38 \pm 0.01 c	7.5 \pm 0.03 bc	7.52 \pm 0.02 b	7.68 \pm 0.05 a

Note: ρ_b —bulk density; ρ_s —soil particle density; W—moisture content; f —porosity; P—available phosphorous; AHN—alkali-hydrolysable nitrogen; C—organic matter; AN—ammonium nitrogen; EC—electrical conductance. FSA, FSB, FSC, FSD, FSE, and FSF represent saline-alkali soil samples with GLR organic fertilizer addition ratio of 25%, 16.7%, 12.5%, 10%, 8.3%, and 0%, respectively.

After application of the GLR organic fertilizer on arid saline-alkali land, the physical and chemical properties of the soil significantly improved. Compared with the FSF treatment, the bulk density, electrical conductivity, and pH of the saline-alkali soil treated with the GLR organic fertilizer were significantly reduced, the soil moisture content and porosity significantly increased. The FSA treatment showed the best improvement effect. The reason may be that after the addition of the GLR organic fertilizer, the structure of the soil improved. Soil porosity, moisture, and permeability increased, and salt leaching was promoted, which is beneficial for the reduction of soil salinity. On the other hand, the reason may be that the organic acid produced after the decomposition of GLR organic fertilizer can reduce the soil pH, buffer the saline-alkali soil, and finally improve soil structure and reduce soil salinity.

3.1.2. Changes of Enzyme Activities in Saline-Alkali Soil

Soil enzymes are sensitive parameters for evaluating soil remediation effects. The enzyme activity is related to soil nutrients, soil types, vegetation characteristics, soil microorganisms, soil moisture, temperature, bulk density, and the properties of the enzymes themselves [20]. As shown in Table 2, the addition of the GLR organic fertilizer significantly increased the activity of invertase, amylase, urease, catalase, and acid phosphomonoesterase. The soil enzyme activity in the FSA and FAB treatments was the highest.

Table 2. Enzyme activity of saline-alkali soil improved by addition of different amounts of GLR organic fertilizer. For each row, the means \pm SD with different letters were significantly different between treatments ($p < 0.05$, using the HSD test).

Sample	FSA (25%)	FSB (16.7%)	FSC (12.5%)	FSD (10%)	FSE (8%)	FSF (0%)
INV (mg/(g·24 h))	8.71 \pm 0.11 a	6.69 \pm 0.21 b	3.56 \pm 0.15 c	3.33 \pm 0.11 c	2.62 \pm 0.16 d	0.51 \pm 0.13 e
AMS (mg/(g·24 h))	7.44 \pm 0.43 a	7.38 \pm 0.47 a	2.09 \pm 0.58 b	1.61 \pm 0.51 bc	1.39 \pm 0.54 bc	0.4 \pm 0.05 c
URE (mg/(g·24 h))	26.32 \pm 0.02 bc	26.69 \pm 0.24 ab	26.63 \pm 0.04 ab	26.69 \pm 0.05 ab	26.82 \pm 0.07 a	26.23 \pm 0.01 c
CAT (mg/(g·24 h))	35.05 \pm 1.79 a	35.86 \pm 2.22 a	34.48 \pm 0.18 a	32.95 \pm 1.45 a	25.61 \pm 1.35 b	18.21 \pm 2.15 c
ACP (mg/(g·24 h))	32.84 \pm 0.03 a	32.74 \pm 0.06 a	32.75 \pm 0.07 a	32.75 \pm 0.07 a	32.62 \pm 0.05 a	32.13 \pm 0.12 b

Note: INV—invertase activity; URE—urease activity; CAT—catalase activity; ACP—acid phosphomonoesterase activity; AMS—amylase activity.

3.1.3. Correlation between the Amount of the GLR Organic Fertilizer and Soil Physicochemical Properties and Enzyme Activity

Soil organic matter is a comprehensive indicator reflecting soil fertility and quality, and is an especially important source of N and P elements [21]. As further indicated by Figure 1A, the addition of the GLR organic fertilizer was significantly related to soil organic matter. Available phosphorus, ammonium nitrogen, and alkali-hydrolysable nitrogen were all significantly positively correlated, indicating that the organic fertilizer of GLR significantly increased the content of soil nutrient elements, and the effect of the FSA treatment (addition amount of 25%) was the most obvious. This was consistent with the findings of other researchers. For example, Zhang et al. [22] found that the application of biochar and organic fertilizer could effectively improve the physical structure of the saline-alkali soil, and the higher the amount of fertilizer applied, the more it is conducive to the storage of carbon and nitrogen. Compound fertilizers and bio-organic fertilizers could significantly increase the content of available phosphorus, available potassium, and alkali-hydrolyzed nitrogen, reduce the total salt content, and increase the soil fertility of typical saline-alkali soils [23].

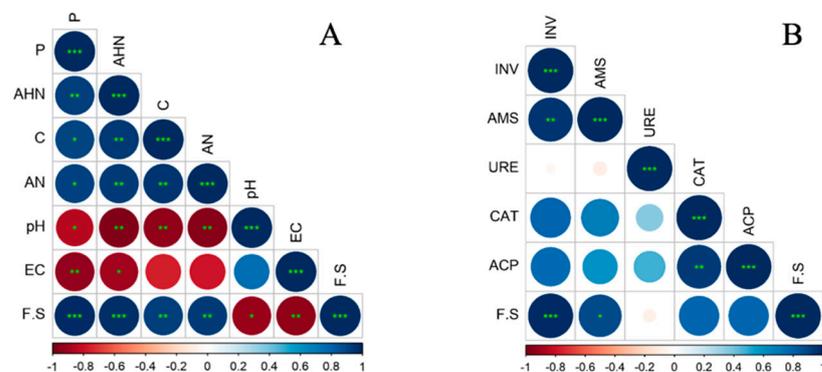


Figure 1. Heatmap of the correlation between the application amount of GLR organic fertilizer and soil physicochemical properties and enzyme activity. Note: (A) correlation between the application amount of GLR organic fertilizer and soil physicochemical properties; (B) correlation between the application amount of GLR organic fertilizer and enzyme activity; P—available phosphorous; AHN—alkali-hydrolysable nitrogen; C—organic matter; AN—ammonium nitrogen; EC—electrical conductance; F. S—addition of GLR organic fertilizer; INV—invertase activity; URE—urease activity; CAT—catalase activity; ACP—acid phosphomonoesterase activity; AMS—amylase activity; Pearson analysis * ($p < 0.05$), ** ($p < 0.01$), *** ($p < 0.001$). Positive correlation is displayed in blue, and negative correlation is displayed in red. The color intensity and the size of the circle are proportional to the correlation coefficient.

Soil enzyme activity can reflect the strength and intensity of various biochemical processes in soil. Relevant studies have shown that soil enzyme activity can represent important indicators such as soil fertility, quality, and microbial activity, etc. [24]. In this study, most of the enzyme activity in saline-alkali soils supplemented with the GLR organic fertilizer was found to be significantly higher than that in the control group (FSF) (Figure 1B). Studies have shown that soil organic matter is the storage place of enzyme sources. N, P, K, and other elements in soil are substrates and products of enzyme action [25]. The results showed that GLR organic fertilizer increases the organic matter content of saline-alkali soil and promotes soil enzyme activity.

3.2. Effects of the GLR Organic Fertilizer on the Microbial Community Structure in Saline-Alkali Soil

A total of 771,397 16S rRNA sequences were obtained from the six soil samples by pyrosequencing of bacterial 16S rRNA genes. Cluster analysis was performed based on a similarity of 97%, and 3986 operational taxonomic units (OTUs) were obtained. A total of

1,363,553 valid sequences were obtained from the six soil samples by pyrosequencing of fungal ITS genes, and 1133 OTUs were obtained by a cluster analysis based on a similarity of 97%. Figure 2 shows that the rarefaction curves of soil bacterial and fungal alpha diversity in the different treatments tended to be flat, indicating that sequencing had become saturated [26].

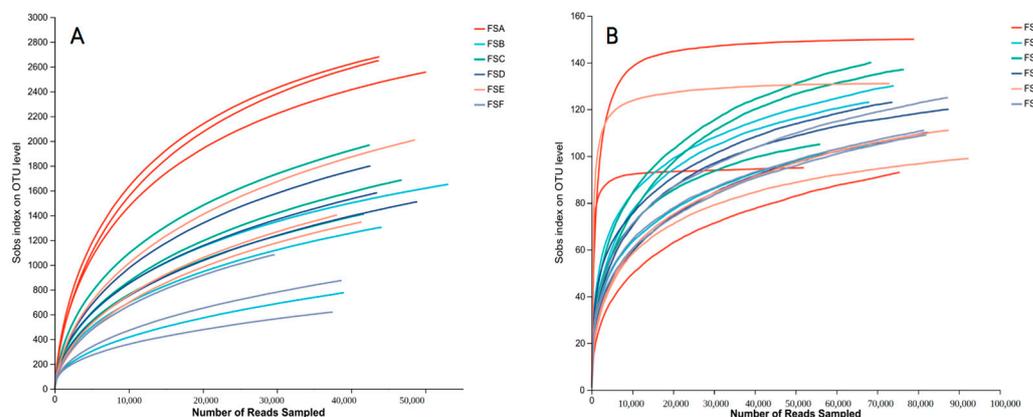


Figure 2. Bacterial and fungal rarefaction curves of different soil treatments. Note: (A) rarefaction curve of bacterial community; (B) rarefaction curve of fungal community. FSA, FSB, FSC, FSD, FSE, and FSF represent saline-alkali soil samples with GLR organic fertilizer addition ratio of 25%, 16.7%, 12.5%, 10%, 8.3%, and 0%, respectively.

3.2.1. Variation of Microbial Community Structure Diversity Index in Saline-Alkali Soil

Alpha diversity reflects the microbial richness and diversity of soil bacteria and fungi (97% similarity). The data in Table 3 show that the organic fertilizer of GLR significantly improved the richness and diversity of the bacterial community structure in the saline-alkali soil but had no significant effect on the diversity of the fungal community structure. The Sobs, Chao1, and Shannon indices of the soil bacterial community in the FSA treatment were significantly higher than those of the other treatment groups.

Table 3. Alpha diversity index of the microbial community in saline-alkali soil improved by different treatments. For each row, the means ± SD with different letters were significantly different between treatments ($p < 0.05$, using the HSD test).

		FSA (25%)	FSB (16.7%)	FSC (12.5%)	FSD (10%)	FSE (8%)	FSF (0%)
Bacteria	Sobs	2628.33 ± 36.84 a	1242.67 ± 254.61 bc	1686.67 ± 161.09 b	1629.67 ± 86.77 b	1585.67 ± 212.28 b	857.00 ± 133.87 c
	Shannon	6.00 ± 0.12 a	4.50 ± 0.28 bc	4.85 ± 0.13 b	4.97 ± 0.12 b	4.48 ± 0.22 bc	4.2 ± 0.05 c
	Simpson	0.01 ± 0.003 b	0.04 ± 0.082 a	0.03 ± 0.003 ab	0.02 ± 0.006 ab	0.05 ± 0.009 a	0.04 ± 0.006 a
	Chao1	3073.44 ± 43.17 a	1849.98 ± 266.71 bc	2429.51 ± 196.88 ab	2358.54 ± 57.18 b	2287.85 ± 247.25 b	1366.89 ± 185.38 bc
Fungi	Sobs	112.67 ± 18.676 a	119.67 ± 7.129 a	127.00 ± 11.200 a	117.67 ± 3.929 a	113.67 ± 9.333 a	117.67 ± 3.772 a
	Shannon	2.53 ± 0.66 a	1.99 ± 0.178 a	1.89 ± 0.045 a	1.81 ± 0.114 a	2.15 ± 0.637 a	1.74 ± 0.066 a
	Simpson	0.18 ± 0.094 a	0.23 ± 0.037 a	0.25 ± 0.008 a	0.26 ± 0.018 a	0.25 ± 0.081 a	0.28 ± 0.012 a
	Chao1	122.68 ± 15.733 a	141.59 ± 5.880 a	140.62 ± 13.984 a	134.65 ± 1.368 a	120.94 ± 7.839 a	139.47 ± 5.474 a

3.2.2. Variation of Dominant Bacteria in Microbial Communities in Saline-Alkali Soils Dominant Phyla of Soil Microbial Communities

Figure 3 shows the composition of microbial communities with species abundance higher than 5% at the phylum level in the improved saline-alkali soil with different treatments. The dominant phyla under the FSF treatment were only Proteobacteria. The dominant phyla under the FSB treatment were Proteobacteria and Bacteroidota. The dominant bacterial phyla under the FSC, FSD, and FSE treatments were Proteobacteria, Bacteroidota, and Firmicutes. Proteobacteria, Actinobacteriota, Firmicutes, and Bacteroidota were the dominant phyla under the FSA treatment. The above results indicated that the application of the GLR organic fertilizer increased the number of dominant bacterial phyla in the saline-alkali soil, increased the abundance of the dominant bacterial phyla Bacteroidota,

Firmicutes, Actinobacteriota, and Chloroflexi, and decreased the abundance of the dominant bacterial phylum Proteobacteria. The dominant phylum in the fungal community of the six soil treatments was Ascomycota. The addition of the GLR organic fertilizer had no significant effect on the number and abundance of the dominant phylum in the fungal community in the saline-alkali soil.

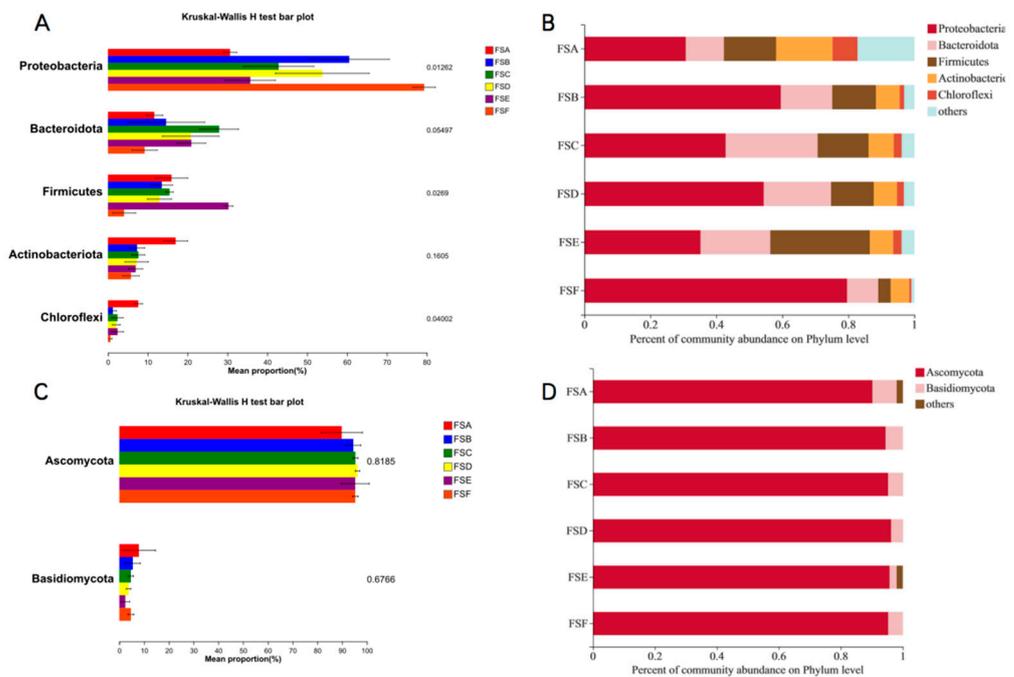


Figure 3. Relative abundance at the phylum level of microbial communities in soils with different treatments. Note: (A) differences in relative abundance of bacterial communities; (B) relative abundance of bacterial communities; (C) differences in relative abundance of fungal communities; (D) relative abundance of fungal communities.

Figure 4 shows the correlation analysis between the application amount of the GLR organic fertilizer and the relative abundance of the dominant phyla. The results showed that with the increase in the added amount of the GLR organic fertilizer, the types of bacteria in each treatment gradually increased, and the FSA treatment had the highest number of species. From an abundance perspective, the added GLR organic fertilizer treatment reduced the soil salinity, increased the abundance of Bacteroidota, Actinobacteriota, Chloroflexi, and Firmicutes, and decreased only the Proteobacteria.

Figure 5 shows the correlation analysis between the relative abundance of the dominant phyla and the soil physical and chemical properties. Some studies have found that Proteobacteria have a good salt tolerance and belong to the group of moderately halophilic bacteria [27]). In this study, Proteobacteria were found to be positively correlated with soil conductivity and pH, and negatively correlated with the addition of the GLR organic fertilizer (Figure 5A). This result shows that Proteobacteria have the characteristics of halophilic bacteria, which is consistent with the findings of Canfora et al. [28]. They also found that the dominant bacteria in saline-alkali soil also contain Proteobacteria, and the abundance of Proteobacteria is positively correlated with soil salinity. The abundance of Firmicutes was negatively correlated with soil salinity, and the abundance of Bacteroidota in saline-alkali soil increases after autumn irrigation [29]. Bacteroidota is a tolerant bacteria group [11]. In this study, Bacteroidota is the dominant bacteria species in saline-alkali soil. Chloroflexi have a strong dependence on soil organic matter content [28]. Actinobacteriota are aerobic microorganisms suitable for growth in environments with high oxygen content. Actinobacteriota and Firmicutes have a relatively high tricalcium phosphate solubilizing

activity [31]. In this experiment, after adding the GLR organic fertilizer to improve saline-alkali soil, the content of organic matter and available phosphorus in the saline-alkali soil significantly increased, the soil structure improved, the soil porosity increased, and the soil particle density decreased (Table 1). Thus, the abundance of Chloroflexi, which are highly dependent on organic matter, and the abundance of Firmicutes with tricalcium phosphate solubilizing activity will increase.

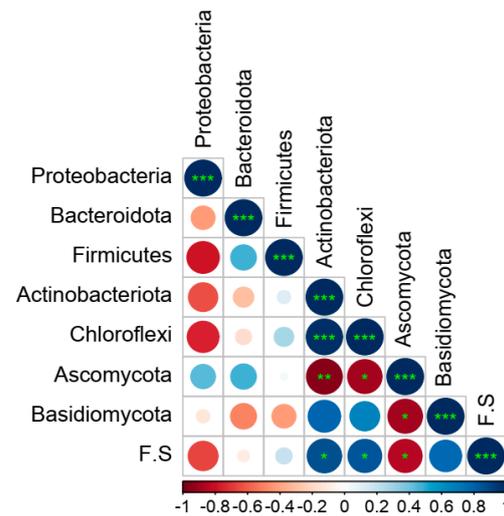


Figure 4. Heatmap of the correlation analysis between the application amount of GLR organic fertilizer and the dominant phyla of bacteria and fungi. Note: F.S—addition amount of GLR organic fertilizer; Pearson analysis * ($p < 0.05$), ** ($p < 0.01$), *** ($p < 0.001$). Positive correlation is displayed in blue, and negative correlation is displayed in red. The color intensity and the size of the circle are proportional to the correlation coefficient.

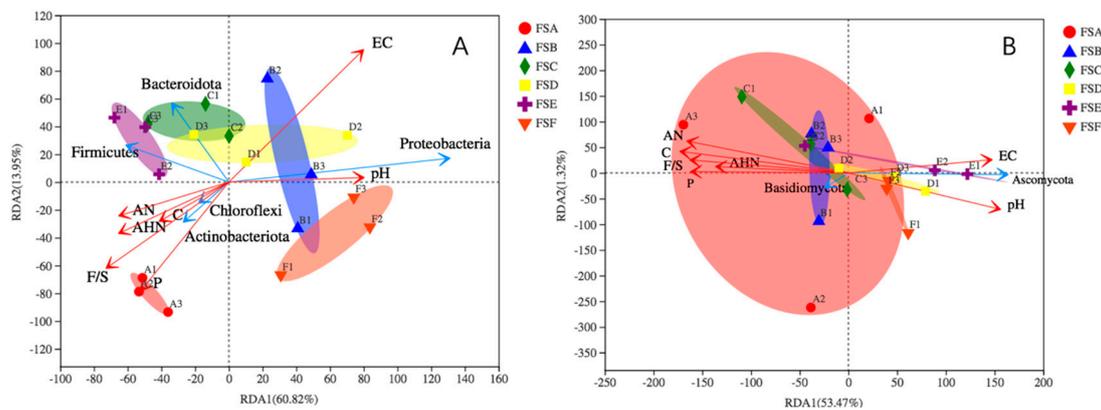


Figure 5. Correlation analysis between dominant microbial communities and environmental factors under different treatments. Note: (A) correlation between dominant bacterial phyla and environmental factors; (B) correlation between dominant fungal phyla and environmental factors; P—available phosphorus; AHN—alkali-hydrolysable nitrogen; C—organic matter; AN—ammonium nitrogen; EC—electrical conductance; F/S—addition amount of GLR organic fertilizer. The blue arrow shows species vectors. The red arrow indicates environmental factors. The length of the arrow of environmental factors represents the degree of influence of environmental factors on species data. The angle between the arrows represents positive and negative correlation (acute angle—positive correlation; obtuse angle—negative correlation; right angle—no correlation).

As the increase of soil porosity improves the permeability of soil, it increases the abundance of aerobic Actinobacteria. Dai et al. [32] found that Firmicutes in the saline soil of

Yinbei were positively correlated with conductivity, and the abundance of Actinobacteriota and Bacteroidota was negatively correlated with soil pH.

The most abundant fungi were Ascomycota and Basidiomycota. Ascomycota play an important role in the terrestrial cycle of ecosystems and decompose organic matter in the nutrient cycle. They also form a symbiotic relationship with other organisms, such as plants [33]. Basidiomycota are the main driving force for restoring soil aggregate stability [34]. In this experiment, the addition of GLR organic fertilizer increased the abundance of Basidiomycota.

Dominant Genera of Soil Microbial Communities

Figure 6 shows the composition of bacterial and fungal communities with a genus-level abundance higher than 5% in soils under different treatments. As shown in Figure 6A, genera having sums of bacterial species abundance $\geq 10\%$ were considered dominant genera. The dominant genera in the soil bacterial community in the FSA treatment were *Bacillus* and *Halomonas*. The dominant genera in the FSE, FSD, FSC, and FSB treatments were all *Halomonas*, and the dominant genus in the FSF treatment was *Stenotrophomonas*. The organic fertilizer of GLR significantly increased the abundance of the dominant genera *Bacillus*, *Salinimicrobium*, *Salegentibacter*, and *Alkalibacterium* and decreased the relative abundance of *Pseudomonas* and *Stenotrophomonas* in the saline-alkali soil. As shown in Figure 6D, the dominant genera of the soil fungal communities in the six treatments were *Aspergillus* and *Talaromyces*. Addition of the GLR organic fertilizer increased the abundance of *Thermomyces* and *Alternaria* (except for unclassified species).

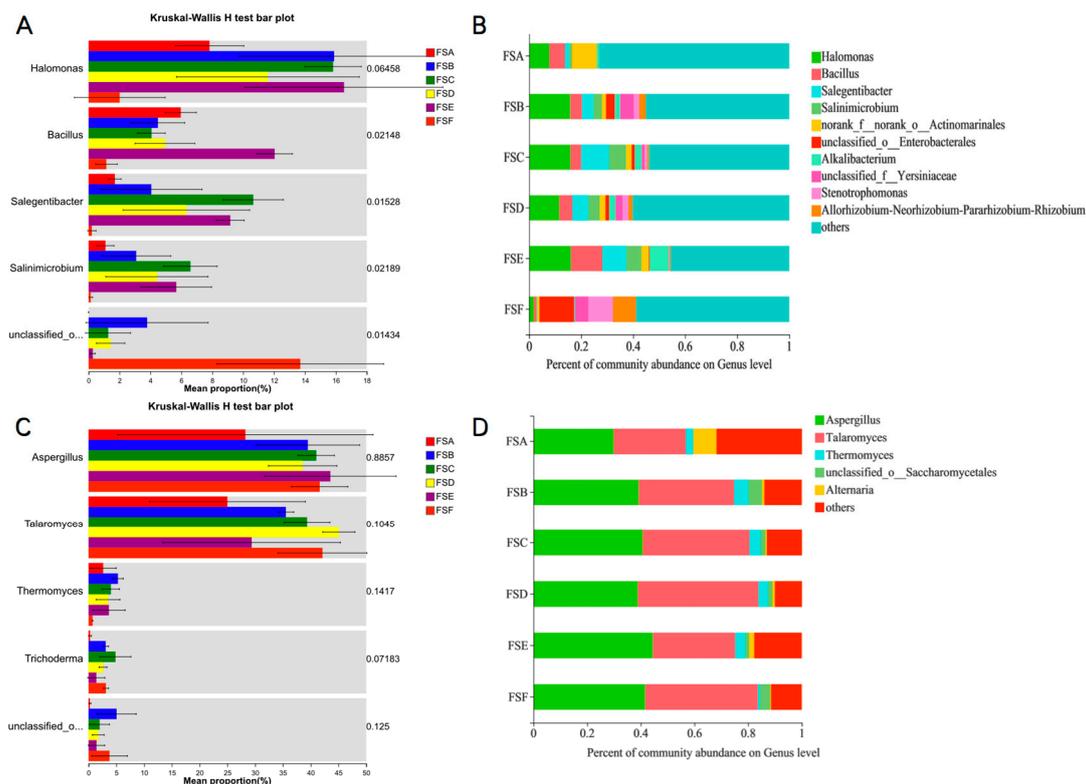


Figure 6. Relative abundance of microbial communities at the genus level in different treatments. Note: (A) differences in relative abundance of bacterial communities; (B) relative abundance of bacterial community; (C) differences in relative abundance of fungal communities; (D) relative abundance of fungal community.

From the bacterial genus level, after adding GLR organic fertilizer, the number of dominant genera species is only 1–2, and there is no obvious change. From the perspective of the abundance of the dominant genera, after the addition of GLR organic fertilizer,

the abundance of *Salegentibacter* and *Pseudomonas* significantly decreased. Some studies found that *Salegentibacter* and *Pseudomonas* were moderately halophilic bacteria [35,36]. Peng et al. [37] explored the effect of land use on the saline-alkali bacterial community. They found that *Bacillus* had a strong salt tolerance and participated in the nitrogen cycle. This microorganism can be used for bioremediation of saline-alkali soil. Shi found that *Bacillus* and *Stenotrophomonas* were the main bacterial species in Anda saline-alkali soil [38]. Thus, the studies above indicate that *Salegentibacter* and *Pseudomonas* may be used as indicator species for improving the salinity level of the studied areas. *Bacillus* can be used as an indicator species for the restoration of saline-alkali land. In the fungal community, there was no difference in the species and abundance of the dominant genera. The dominant genera were *Aspergillus* and *Talaromyces* with higher abundance values in the treated soils. *Aspergillus* can secrete acids, such as formic acid, which can convert insoluble phosphate into soluble phosphate and reduce soil pH value [39]. *Trichoderma* is a promising strain that can induce plant resistance under salt stress conditions and reduce the impact of salt damage on plants. Some studies have found that *Trichoderma harzianum* has a strong salt tolerance [40]. In conclusion, the addition of GLR organic fertilizer improved the soil quality and increased the abundance of salt-alkali tolerant bacteria in the short term. It may be that some elements in GLR organic fertilizer are beneficial to promote the increase of salt-alkali-tolerant bacteria. At the same time, it also promoted the abundance of fungi with the characteristics of acid production, alkali reduction, and increased plant resistance.

4. Conclusions

The results showed that GLR organic fertilizer could improve saline-alkali soil by reducing pH, conductivity, bulk density, and density of saline-alkali soil, increasing porosity and organic matter, improving soil enzyme activity, and regulating soil microbial community structure. Among them, the effect of GLR organic fertilizer application ratio of 25% (FSA) is the most significant. Overall, GLR can be used for saline-alkali soil improvement. The findings can provide a theoretical basis for improving saline-alkali soils, rational utilization of TCMRs, and promoting sustainable development of modern agriculture.

Author Contributions: Conceptualization, C.-F.S.; methodology, H.-T.Y.; formal analysis, J.B.; investigation, X.-Y.L.; data curation, Y.-M.W.; writing—original draft preparation, H.-T.Y. and L.-P.G.; writing—review and editing, P.-B.D. and C.-F.S.; funding acquisition, C.-F.S. All authors have read and agreed to the published version of the manuscript.

Funding: This work was supported by the Science and Technology Plan Project of Inner Mongolia (Grant No. 2020GG0071).

Data Availability Statement: The data supporting the findings of this study are available from the corresponding author upon reasonable request.

Conflicts of Interest: The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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