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# Regulatory effect of the amendment with vermicompost and coconut-shell biochar on microbial ecology in coastal saline soil

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**Abstract:** Soil salinization is a major challenge in soil management, and remediating saline soils is crucial for sustainable soil resource development. Although vermicompost and biochar are frequently employed as soil amendments, their micro-ecological remediation mechanism for acid saline soils needs further verification. This study aimed to investigate whether vermicompost co-applied with coconut-shell biochar exerts a synergistic positive impact on coastal saline soil micro-ecology. A pot experiment was conducted to examine the micro-ecological regulation effect of vermicompost co-applied with coconut-shell biochar on coastal saline soil. The study revealed that applying the amendment containing vermicompost and coconut-shell biochar significantly improved the quality of coastal saline soils by reducing soil salinity, increasing the soil pH and organic matter (OM), and enhancing nutrient availability and enzyme activity. The soil salinity changed from moderate to mild, and total water-soluble salts (TS) significantly decreased by 72.8%. The soil pH raised significantly by 6.5%. The contents of OM, alkali-hydrolyzed nitrogen (AN), available phosphorus (AP), and available potassium (AK) increased by 20.8%, 50.4%, 80.9%, and 41.6%, respectively. Moreover, the three enzyme activities of urease (UE), acid phosphatase (ACP), and catalase (CAT) increased by 835%, 17.1%, and 130%, respectively. Additionally, vermicompost co-applied with coconut-shell biochar significantly impacted bacterial diversity and community composition. Notably, vermicompost co-applied with coconut-shell biochar boosted the growth of key salt-tolerant bacterial groups. Specifically, the relative abundance of Acidobacteriota, Gemmatimonadetes, Actinobacteria, and Chloroflexi increased by 167%, 888%, 86.7%, and 123%, respectively. Redundancy analysis showed that vermicompost co-applied with coconut-shell biochar could reduce TS by increasing pH, available nutrients, bacterial diversity, and enzyme activities in coastal soils. To sum up, vermicompost co-applied with coconut-shell biochar played a crucial role in positively influencing the soil micro-ecological environment. It effectively reduced soil salinity and would hold great potential for improving saline soil conditions.

**Keywords:** vermicompost; coconut-shell biochar; coastal saline soil; micro-ecology; soil biochemical properties; bacterial community

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## 1. Introduction

The escalating annual salinity rate of 10% has turned soil salinization into a significant challenge [1]. Saline soils are extensive in coverage and broadly distributed in China, especially in coastal areas, where the land salinization is very serious and

has become a crucial problem of soil environment [2]. High salinity levels can impair the soil aggregate structure, thereby reducing aeration, permeability, and water-holding capacity, while also disturbing nutrient cycling in saline soils [3–5]. In saline soils, excessive salinity, degradation of soil structure, and nutrient deficiencies are three primary limitations to plant growth [6]. Reducing soil salinity, enhancing soil nutrient content, and promoting soil microbial community diversity are crucial strategies for improving saline soil. Currently, the primary measures for improving saline soils encompass engineering (e.g., irrigation salt washing), chemical (e.g., desulfurization gypsum), and biological measures (e.g., halophytes), among others [7–9]. Nevertheless, these measures are not without limitations. For instance, the high costs and energy consumption associated with hydraulic engineering impede its widespread use in salinity management. Additionally, chemical amendments such as gypsum often fail to deliver positive effects [10] and may lead to secondary contamination, thereby exacerbating ecological and environmental risks [11]. Likewise, phytoremediation necessitates a prolonged period and specific conditions to attain satisfactory restoration outcomes [12]. Hence, adopting suitable measures to enhance the micro-ecological environment of saline soil is vital for promoting crop production and maintaining sustainable ecosystem function.

Vermicompost serves as a superior organic fertilizer and soil conditioner, characterized by its rich nutrient content, high porosity, aeration, and water-holding capacity [13]. It can improve soil physicochemical characteristics, including soil aeration, porosity, bulk density, pH, electrical conductivity (EC), organic carbon content, and bacterial community diversity, resulting in increased soil fertility [13–15]. Biochar is a solid material that is rich in carbon, highly porous, and alkaline in nature, which can not only increase the content of soil organic carbon but also enhance the soil's capacity to retain nutrients and water [16–18]. The porous structure and high adsorption capacity of biochar create an optimal environment for soil microorganisms, which in turn promotes the growth and activity of beneficial microbes. Moreover, it can slow down the liberation of fertilizer nutrients within the soil, thereby enhancing the efficiency with which nutrients are utilized [19,20]. Vermicompost and biochar have been extensively used as soil conditioners to boost soil carbon sequestration, upgrade soil quality, and improve nutrient use efficiency. However, past research has predominantly concentrated on grain crops, with limited attention given to the micro-ecological environment of coastal saline soils.

In recent years, the combined use of vermicompost and biochar has gained growing attention for its potential to improve soil fertility and enhance plant growth in agricultural ecosystems [21–23]. Vermicompost co-applied with biochar has the capacity to enhance soil stability and boost crop productivity while simultaneously mitigating the adverse impacts of agricultural activities on ammonia and nitrous oxide emissions [22]. In our prior research, we established the volume ratios of vermicompost at 2, 2.5, and 3, respectively, and those of coconut-shell biochar at 1, 1.5, and 2, respectively. These two materials were subsequently paired and mixed and then added into saline soil with the volume ratio of 20 parts. The findings demonstrated that the optimal volume ratio of vermicompost to coconut shell biochar was 3:1.5. However, whether vermicompost and coconut-shell biochar have a positive cooperative effect on reducing soil salinity and improving the micro-ecological

environment of acidic coastal saline soil remains unknown. Hence, the purpose of this study was to explore whether the combined application of vermicompost and coconut-shell biochar exerts a positive cooperative effect on the micro-ecological environment of coastal saline soils. The results of this study would be anticipated to offer technical guidance and a theoretical foundation for the reclamation of coastal saline soils.

## 2. Materials and methods

### 2.1. Experiment design

The pot experiment was performed in a greenhouse. The tested soil was collected from the 0–20 cm arable layer in Nanxi Village, Wenchang City, Hainan Province (N 19°59'24.5", E 110°37'16.6"), China. The tested soil amendment was the best combination of vermicompost and coconut-shell biochar developed by our research team. The tested crop was cherry tomato (*Lycopersicon esculentum* Mill.). Two groups were established: one involved the complete absence of fertilizers (CK), and the other used only soil amendment with vermicompost and coconut-shell biochar (T), and the volume ratio of vermicompost, coconut-shell biochar, and saline soil was set at 3:1.5:20. Each experiment group was designed to include six replicates. Other management practices were identical in the two groups. The experiment lasted 120 days, with data collected at the end of the study. Soil organic matter (OM), pH, and nutrient levels (nitrogen, phosphorus, and potassium) were analyzed before and after the experiment. Following the conclusion of the study, soil enzyme activities and bacterial communities were assessed. The fundamental chemical characteristics of the tested saline soil, vermicompost, and coconut-shell biochar were presented in **Table 1**.

**Table 1.** Basic properties of vermicompost, coconut-shell biochar, and saline soil before treatments: organic matter (OM), total nitrogen (TN), total phosphorus (TP), total potassium (TK), and total water-soluble salts (TS).

Indicators	Vermicompost	Coconut-shell biochar	Saline soil
pH	7.13	9.48	6.24
OM/g·kg <sup>-1</sup>	236	543	11.7
TN/g·kg <sup>-1</sup>	9.74	2.81	0.68
TP/g·kg <sup>-1</sup>	45.2	1.50	1.68
TK/g·kg <sup>-1</sup>	7.43	15.3	0.29
TS/%	-	-	3.79

### 2.2. Soil sampling

Soil samples were collected 120 days after planting cherry tomatoes. The plants were pulled out after 120 days of planting cherry tomatoes. Every two pots of soil with the same treatment are thoroughly mixed into one soil sample. Six soil samples in total were collected for further examination. Each sample was split into two portions. The first portion was air-dried, passed through a 2-mm sieve to remove large particles and plant debris, and then used for analyzing soil biochemical characteristics. The second portion was preserved at –80 °C for forthcoming microbial community analysis.

### 2.3. Analysis of soil chemical properties and enzyme activity

Soil pH, organic matter (OM), alkali-hydrolyzed nitrogen (AN), available phosphorus (AP), available potassium (AK), and total water-soluble salts (TS) were quantified using the standard methods outlined by Lu [24]. The soil pH was measured using a pH electrode (Leici, Shanghai, China) at a soil-to-water ratio of 1 to 2.5. The OM content was determined by the potassium dichromate volumetric method. The contents of AN, AP, and AK were analyzed using a diffusion method, Olsen method, and ammonium acetate extraction flame photometry, respectively. The TS content was measured using electrical conductivity. Three kinds of soil enzyme activities of urease (UE), acid phosphatase (ACP), and catalase (CAT) were determined by kit (Suzhou Keming Biological Technology Co., LTD., [www.cominbio.com](http://www.cominbio.com)).

### 2.4. Soil bacterial community analysis

Total soil DNA extraction was performed with a Power Soil DNA Isolation Kit (MO BIO Laboratories, Carlsbad, CA, United States) following the manufacturer's instructions. Bacterial profiling was conducted using 16S rRNA gene sequencing, targeting the amplification of the V3-V4 region of the bacterial 16S gene. Amplification was performed using the primers 319F (5'-ACTCCTACGGGAGGCAGCA-3') and 806R (5'-GGACTACHVGGGTWTCTAAT-3'), and the resulting amplicons were subsequently combined with adapter and barcode sequences. The PCR was implemented following the conditions reported by Wu et al. [25]. The sequencing libraries were prepared and sequenced by Biomarker Biotechnology Co., Ltd. (Beijing, China), employing the Illumina NovaSeq 6000 sequencing system (Illumina, Santiago, CA, United States). The final compelling reads were obtained after chimeric sequences were identified and removed using the UCHIME v4.2 software ([www.drive5.com](http://www.drive5.com)). Analysis of the sequencing data was performed using the Quantitative Insights into Microbial Ecology (QIIME 1.8.0) toolkit. The optimized sequences were then clustered into operational taxonomic units (OTUs) at the 97% sequence similarity level using Usearch v10 software. A naive bayes classifier was employed to annotate the feature sequences, using the SILVA database as the reference, in order to obtain the species classification information for each feature. Using mothur version 1.30 on the BMKCloud ([www.biocloud.net](http://www.biocloud.net)), the alpha diversity indices, including Chao1, abundance-based coverage estimator (ACE), Shannon, and Simpson, were determined. The taxa with differential abundance were identified with the linear discriminant analysis (LDA) effect size (LEfSe) method. Significance was assigned to LDA scores exceeding the preset threshold of 4.0. Additionally, redundancy analysis (RDA) and Pearson correlation analysis were conducted to investigate the relationships between soil environmental factors and bacterial communities.

### 2.5. Statistical analysis

Replicate data were presented as the mean  $\pm$  standard deviation (SD). Calculations and comparisons of treatment means for each experiment were performed using the Statistical Product and Service Solutions (SPSS) v.17.0 software package (IBM Corp., Armonk, NY, United States). To assess significant differences among

means, Duncan's tests and one-way analysis of variance (ANOVA) were employed, using a significance threshold of  $p \leq 0.05$ .

### 3. Results

#### 3.1. Variations in soil biochemical properties

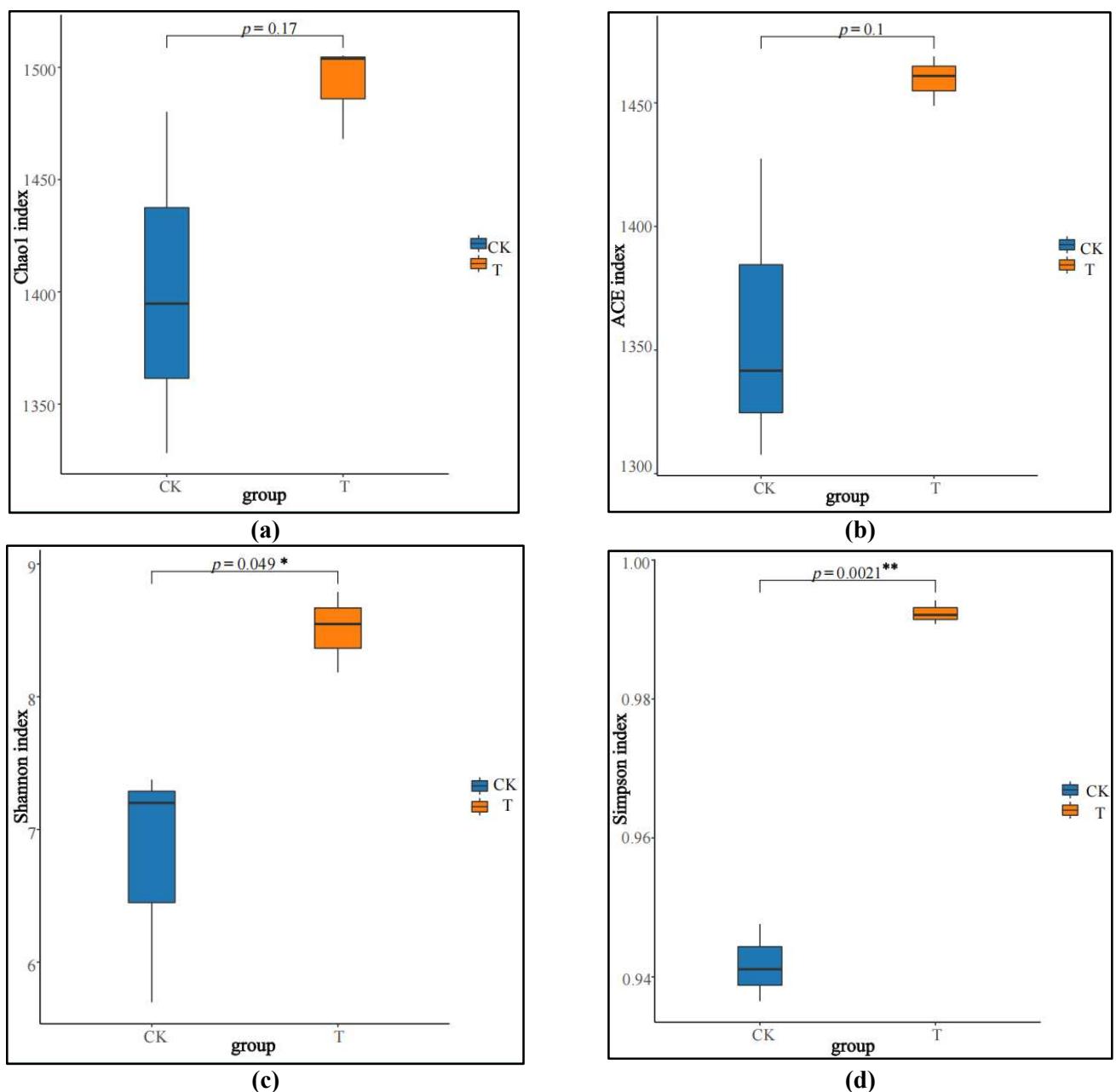
The biochemical properties of saline soil were notably influenced by soil amendment with vermicompost and coconut-shell biochar (Table 2). The soil pH value and nutrient contents were substantially elevated after adding amendments. The soil pH value increased significantly by 6.5% ( $P < 0.05$ ). The contents of OM, AN, AP, and AK increased by 20.8%, 50.4% ( $P < 0.05$ ), 80.9% ( $P < 0.05$ ), and 41.6% ( $P < 0.05$ ), respectively. However, soil TS changed from moderate to mild, significantly decreasing by 72.8% ( $P < 0.05$ ). Moreover, the three enzyme activities of UE, ACP, and CAT increased by 835% ( $P < 0.05$ ), 17.1%, and 130% ( $P < 0.05$ ) in T treatment, respectively.

**Table 2.** Effects of soil amendment on soil chemical properties, nutrient content, and enzyme activity. Lowercase letters indicate significant differences between treatments ( $P < 0.05$ ). Values shown are the mean  $\pm$  standard error ( $n = 3$ ). CK: saline soil. T: saline soil + soil amendment (the volume ratio was 3:1.5 of vermicompost to coconut-shell biochar). total water-soluble salts (TS), organic matter (OM), alkali-hydrolyzable nitrogen (AN), available phosphorus (AP), available potassium (AK), urease (UE), acid phosphatase (ACP), and catalase (CAT).

Treatment	pH	TS /mg·kg <sup>-1</sup>	OM /g·kg <sup>-1</sup>	AN /mg·kg <sup>-1</sup>	AP /mg·kg <sup>-1</sup>	AK /mg·kg <sup>-1</sup>	UE /μg·d <sup>-1</sup> ·g <sup>-1</sup>	ACP /μmol·d <sup>-1</sup> ·g <sup>-1</sup>	CAT /μmol·d <sup>-1</sup> ·g <sup>-1</sup>
CK	6.14 $\pm$ 0.03 b	3.81 $\pm$ 0.30 a	10.1 $\pm$ 1.44 a	50.2 $\pm$ 2.25 b	109.3 $\pm$ 2.51 b	211.5 $\pm$ 4.41 b	76.02 $\pm$ 8.70 b	11.48 $\pm$ 1.44 a	5.82 $\pm$ 0.78 b
T	6.54 $\pm$ 0.07 a	1.18 $\pm$ 0.12 b	12.2 $\pm$ 1.01 a	74.7 $\pm$ 6.60 a	181.6 $\pm$ 15.6 a	299.5 $\pm$ 1.31 a	710.9 $\pm$ 20.7 a	13.44 $\pm$ 1.01 a	13.4 $\pm$ 0.89 a

#### 3.2. Variations in soil bacterial communities

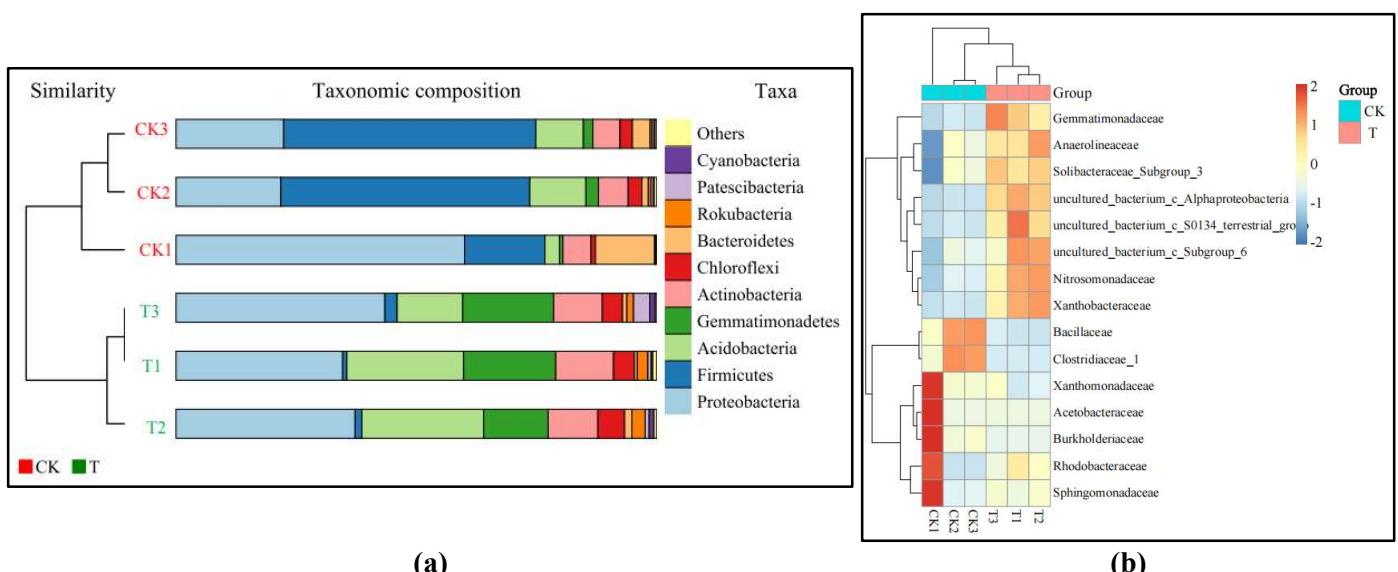
Soil amendments significantly affected the bacterial richness and diversity in saline soil (Figure 1). The richness indices Chao1 (Figure 1a) and ACE (Figure 1b) increased by 8.1% and 7.2% ( $p > 0.05$ ), respectively. The diversity indices Shannon (Figure 1c) and Simpson (Figure 1d), ranging from 6.76 to 8.68 and from 0.94 to 0.99, were significantly increased by 28.4% ( $p < 0.05$ ) and 5.4% ( $p < 0.05$ ) in T treatment compared with CK, respectively.



**Figure 1.** Boxplot of alpha diversity index of soil bacterial communities: **(a)** Chao1 index; **(b)** ACE index; **(c)** Shannon index; **(d)** Simpson index. CK: saline soil. T: saline soil + soil amendment (the volume ratio was 3:1.5 of vermicompost to coconut-shell biochar).

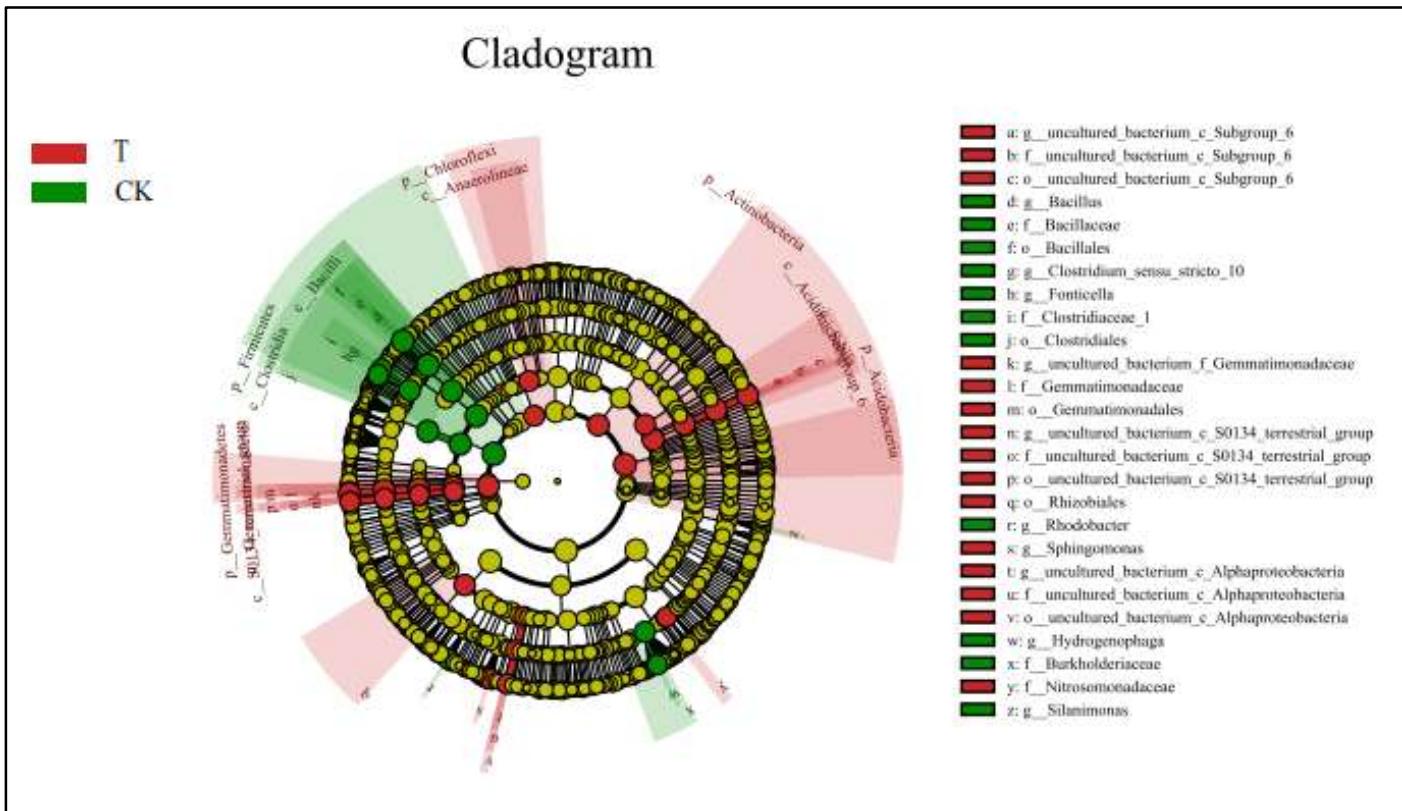
The soil bacterial communities showed a similar composition in both treatments of T and CK. However, the relative abundance of some bacteria was significantly different between two treatments (Figure 2). At the phylum classification level, species with a relative abundance above 1% were categorized as predominant bacteria. The main predominant bacteria were Proteobacteria, Firmicutes, Acidobacteria, Gemmatimonadetes, Actinobacteria, Chloroflexi, Bacteroidetes, etc. in CK. Firmicutes had the greatest relative abundance (39.9%), followed by Proteobacteria (35.2%). However, that of Proteobacteria was the highest (37.9%), followed by Acidobacteria (21.7%) in T treatment (Figure 2a). Relative to CK, the relative

abundance of Firmicutes and Bacteroidetes in T treatment was decreased by 96.3% ( $p < 0.05$ ) and 83.6% ( $p > 0.05$ ), respectively. However, that of Acidobacteria, Gemmatimonadetes, Actinobacteria, and Chloroflexi was significantly boosted by 167% ( $p < 0.05$ ), 888% ( $p < 0.05$ ), 86.7% ( $p < 0.05$ ), and 123% ( $p < 0.05$ ) in T treatment, respectively. UPGMA clustering analysis showed that bacterial community composition clustered into two different branches in the two treatments (**Figure 2a**), which implies that the application of soil amendments in saline soils led to significant changes in soil bacterial community structure. At the family level, species with a relative abundance greater than 1% were also chosen to determine dominant bacteria (**Figure 2b**). In comparison to CK, T exhibited a substantial decrease in the relative abundance of Bacillaceae (97.0%,  $p < 0.05$ ) and Clostridiaceae\_1 (82.6%,  $p < 0.05$ ). However, that of Gemmatimonadaceae, Nitrosomonadaceae, Xanthobacteraceae, and Solibacteraceae\_Subgroup\_3 was significantly increased by 757% ( $p < 0.05$ ), 401% ( $p < 0.05$ ), 283% ( $p > 0.05$ ), and 86.7% ( $p > 0.05$ ) in T treatment, respectively.



**Figure 2.** Soil bacterial community composition with relative abundance > 1%: **(a)** Binary-jaccard UPGMA clustering analysis of bacterial community at phylum level; **(b)** Heatmap of similarities between the bacterial communities based on Euclidean distance indices at family level. CK: saline soil. T: saline soil + soil amendment (the volume ratio was 3:1.5 of vermicompost to coconut-shell biochar).

LEfSe analysis further revealed species with statistical differences between the two treatments of T and CK (**Figure 3**). 14 biomarker taxa were identified in CK, which mainly included one phylum (Firmicutes), two classes (Bacilli and Clostridia), two orders (Bacillales and Clostridiales), three families (Bacillaceae, Clostridiaceae\_1, and Burkholderiaceae), and six genera (such as *Bacillus*, *Fonticella*, etc.). And 24 biomarker taxa were identified in T treatment, which mainly included four phyla (Gemmatimonas, Acidobacteria, Actinobacteria, and Chloroflexi), five classes (such as Gemmatimonadetes, Acidimicrobii, etc.), five orders (such as Gemmatimonadeles, Rhizobiales, etc.), five families (such as Gemmatimonadaceae, Nitrosomonadaceae, etc.), and five genera (such as *Sphingomonas*, etc.).

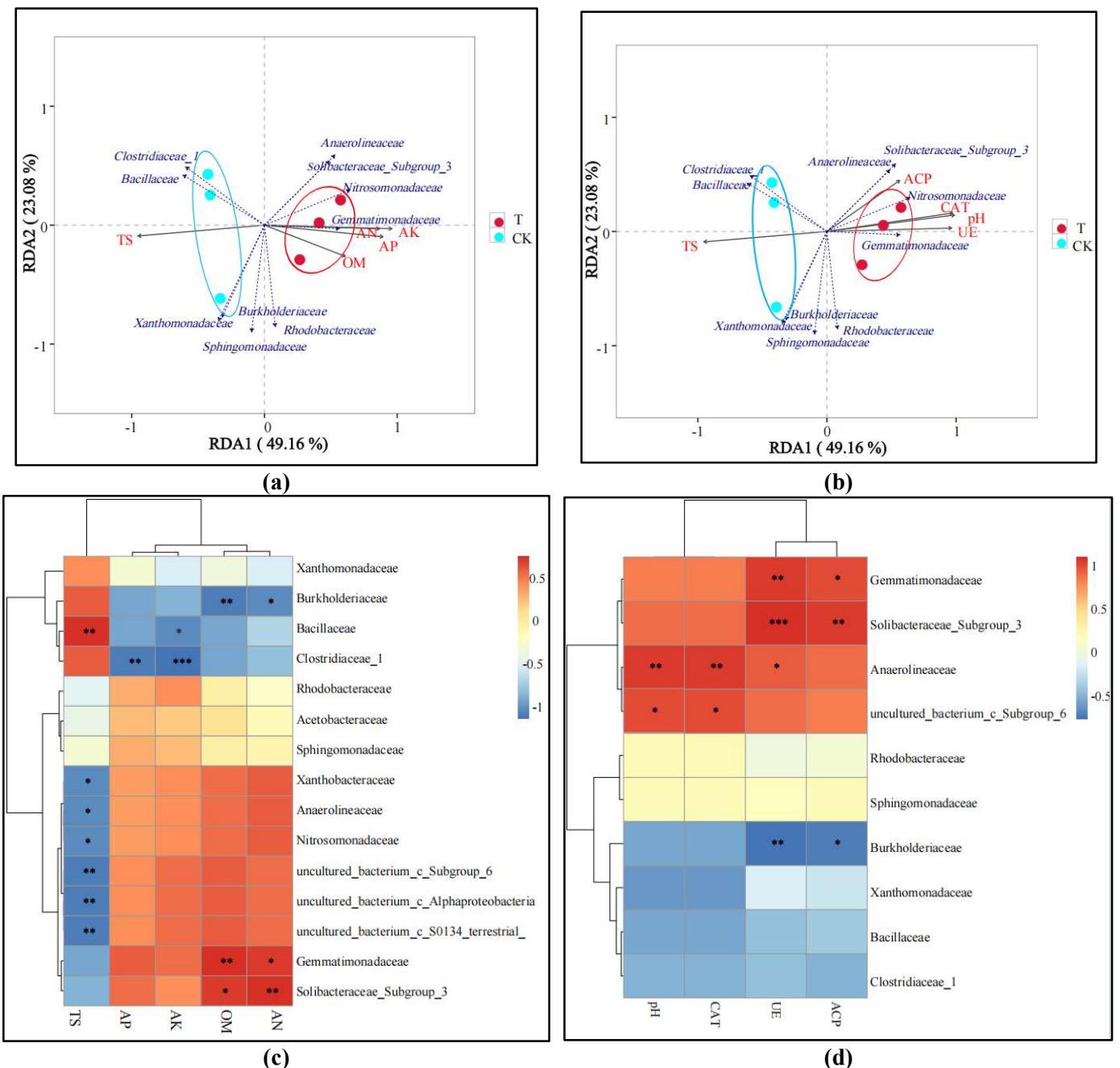


**Figure 3.** Cladogram of line discriminant analysis effect size (LEfSe) analyses based on the composition of bacteria communities. The line discriminant analysis (LDA) scores exceeding the preset threshold of 4.0 were deemed significant. The innermost circle corresponds to the phylum taxonomic level, with the outer circles representing the taxonomic levels of class, order, family, and genus in turn. Node size reflects abundance, significantly discriminant taxon nodes are colored, and branch areas are shaded based on the highest-ranked group for that taxon. If the taxon was not significantly differentially represented among the sample groups, the corresponding node was highlighted in yellow. CK: saline soil. T: saline soil + soil amendment (the volume ratio was 3:1.5 of vermicompost to coconut-shell biochar).

### 3.3. Correlations between soil biochemical properties and bacterial communities

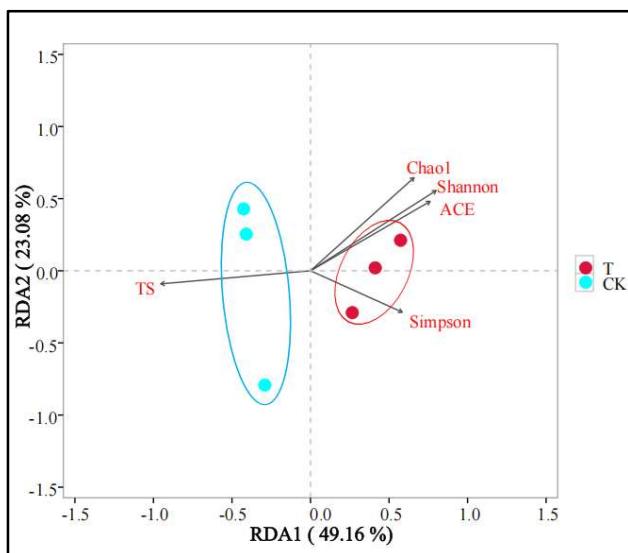
Redundancy analysis (RDA) was employed to explore the relationships between soil biochemical properties and bacterial communities (**Figure 4a,b**). The first two axes of the RDA captured 72.2% of the total variation in soil bacterial community structure. The respective relative abundance of dominant bacterial families, such as Gemmatimonadaceae, Solibacteraceae\_Subgroup\_3, and Nitrosomonadaceae showed positive correlations with soil available nutrients (AN, AP, and AK), OM, pH, and enzyme activities (UE, ACP, and CAT), but negative correlations with soil TS (**Figure 4a,b**). Pearson correlation analysis was utilized to further examine the relationship between microbial community structure and soil biochemical properties (**Figure 4c,d**). The results revealed significant correlations between dominant soil bacterial families and soil biochemical properties. Specifically, Gemmatimonadaceae and Solibacteraceae\_Subgroup\_3 showed positive correlations with soil OM, AN, UE, and ACP ( $P < 0.05$ ), while Nitrosomonadaceae showed a negative correlation with soil TS ( $P < 0.05$ ). The results also showed that soil TS exhibited negative correlations with soil OM, AN, AP, AK, pH, UE, ACP, and CAT (**Figure 4a,b**). Similarly, it was also

negatively correlated with soil bacterial abundance indices (ACE and Chao1) and diversity indices (Shannon and Simpson) (**Figure 5**).



**Figure 4.** Co-relationships between soil bacteria communities and soil environmental factors. Redundancy analysis (RDA) of (a) soil nutrients and (b) soil enzyme activities; Pearson correlation heat map for (c) soil nutrients and (d) soil enzyme activities. Reds signify positive correlations, while blues denote negative correlations.

\*  $P \leq 0.05$ ; \*\*  $P \leq 0.01$ ; \*\*\*  $P \leq 0.001$ . CK: saline soil. T: saline soil + soil amendment (the volume ratio was 3:1.5 of vermicompost to coconut-shell biochar).



**Figure 5.** Redundancy analysis (RDA) of bacterial diversity index and soil salinity. CK: saline soil.

T: saline soil + soil amendment (the volume ratio was 3:1.5 of vermicompost to coconut-shell biochar).

#### 4. Discussion

##### 4.1. Effect of vermicompost co-applied coconut-shell biochar on biochemical properties in saline soil

Acidic soil has the disadvantages of reducing the availability of soil nutrients, destroying soil structure, and not being conducive to the soil microbial activity and the growth and development of crops. In this study, the coastal saline soil belongs to acid soil, whose pH value was significantly improved after adding soil amendments with vermicompost and coconut-shell biochar (Table 2). The primary reason is that the high pH (9.48) and alkaline functional groups (such as  $-\text{OH}$  and  $-\text{COOH}-$ ) of coconut-shell biochar neutralize acid ions in the coastal saline soil [23,26]. It might also be because the mineralization and humification of OM in vermicompost result in the production of a large number of “ $-\text{OH}$ ” and other functional groups, which in turn consume  $\text{H}^+$  ions [27]. Vermicompost co-applied with coconut-shell biochar can help enhance soil nutrient retention and create a nutrient-rich environment that benefits soil microorganisms [28,29], which promotes the OM decomposition. Additionally, combining biochar with vermicompost can enhance the decomposability of vermicompost and result in sustained increases in OM [30]. Vermicompost and coconut-shell biochar, which contain high levels of N, P, and K, directly contributed to the increased nutrient content of coastal saline soil [31]. Previous studies had shown that vermicompost can significantly elevate soil nitrogen, phosphorus, and potassium levels [32–34].

Higher salinity levels in saline soils result in decreased soil nutrient accessibility and reduced soil enzyme activity, thereby inevitably restricting agricultural production. All biochemical reactions in the soil are accomplished with the participation of soil enzymes, and the level of soil enzyme activity can reflect soil transformation and fertility. Our prior research had demonstrated that incorporating vermicompost alone into saline soil at a volume ratio of 3:20 enhanced the activities

of soil UE, ACP, and CAT by 15.4%, 21.4%, and 63.4%, respectively [33]. Conversely, the addition of coconut-shell biochar alone to saline soil at a volume ratio of 1:20–4:20 significantly suppresses the activities of soil UE and ACP. Notably, the CAT activity increased within the range of 21.3%–71.1%, with optimal treatment effects observed at a volume ratio of 3:20 [34]. In this study, we found that the combined application of the two materials had a better effect, and the addition of vermicompost and coconut-shell biochar in a volume ratio of 3:1.5 to 20 parts of coastal saline soil could significantly increase the activities of soil UE, ACP, and CAT by 835%, 17.1%, and 130%, respectively.

Furthermore, this study also found that the combined application of vermicompost and coconut-shell biochar was found to be more effective in reducing salinity in saline soil than either material alone [33,34]. Coconut-shell biochar, characterized by its numerous pores and robust sorption capacity, contains ionic hydroxyl and carboxyl functional groups [35]. These features result in a surface with a high density of negative charges, which promotes the adsorption of cations and reduces TS in soils. Vermicompost, with its strong sorption ability [13], adsorbs salts and amino acids resulting from the microbial decomposition of OM, which helps to reduce soil TS. The co-application of vermicompost and coconut-shell biochar enhances the precipitation of inorganic salts, leading to greater reductions in TS values compared to the individual use of vermicompost or coconut-shell biochar [33,34,36,37].

#### 4.2. Effect of vermicompost co-applied coconut-shell biochar on bacterial community in saline soil

As we predicted, the vermicompost co-applied with coconut-shell biochar can increase the diversity of the bacterial community (**Figure 1**). Vermicompost has been shown in some studies to increase the richness and diversity of bacterial communities and promote the enrichment of beneficial bacteria, which is conducive to superior plant growth [32]. Adding vermicompost and coconut-shell biochar to saline soil can help reduce saline pressure and provide nutrients that support microbial reproduction [20], thus positively influencing microbial diversity [38,39]. This could be due to the direct effect of increased soil nutrients on microbial diversity after applying microbe-rich vermicompost [40], and the increase in microbial diversity can also notably optimize soil environment [41]. This might be due to applying microbe-rich vermicompost directly increasing soil nutrients, which in turn drives the increase of microbial diversity [40]. That is, microbial diversity and soil nutrients exhibited a positive feedback mechanism, with both being enhanced synergistically after adding microbe-rich vermicompost [42].

Microbial community is crucial in maintaining soil microecological stability [43,44]. In this study, the application of soil amendment with vermicompost and coconut-shell biochar increased the relative abundance of most dominant bacteria, such as Proteobacteria (Nitrosomonadaceae and Xanthobacteraceae), Acidobacteria (Solibacteraceae), Gemmatimonadetes (Gemmatimonadaceae), Actinobacteria, Chloroflexi, etc., but decreased the relative abundance of Firmicutes (Bacillaceae and Clostridiaceae\_1) and Bacteroidetes (**Figure 2**). Several studies have indicated that

the abundance of Firmicutes and Bacteroidetes tends to be higher under high-salt conditions [45]. Similarly, these two phyla have been shown to possess tolerance mechanisms and robust structural adaptations in response to elevated salinity stress [46]. Proteobacteria, Gemmatimonadetes, Chloroflexi, and Actinobacteria have been found to display remarkable adaptability to high-salinity environments, a trait that frequently results in their higher abundance in saline soils [46–48]. Vermicompost co-applied with coconut-shell biochar significantly increased their relative abundance in coastal saline soil, and this increase in abundance subsequently boosted their metabolic activities and interactions with plants, effectively alleviating the saline stress associated with the use of vermicompost and coconut-shell biochar. The significant differences in microbial community compositions between the treatment involving co-application of vermicompost and coconut-shell biochar and the treatment with sole application of chemical fertilizer might be attributed to the high salinity characteristic of coastal saline soils [49,50]. A high level of salinity has been documented to inhibit microbial growth and decrease microbial diversity. In particular, the relative abundances of Actinobacteria and Acidobacteria under high salinity were markedly lower than those under low salinity [45], and only those microorganisms with salt tolerance (Firmicutes, Proteobacteria, Chloroflexi, etc. were capable of surviving in high-salinity soils [51]. In addition, Proteobacteria are capable of accumulating soil nutrients and enhancing both plant growth and stress resistance [52,53]. Acidobacteria are capable of breaking down animal and plant residues and contributing to the metabolism of single-carbon compounds in nutrient-poor soils [54]. Chloroflexi are able to assimilate and take up a wide range of organic acids derived from both biotic and abiotic sources in the environment [55]. Gemmatimonadetes are capable of preventing and controlling certain plant diseases [56]. Actinobacteria serve as a crucial source of antibiotics, biocides, and antifungal agents in agricultural practices, and they also have the ability to enhance crop growth [57]. This study demonstrated that the relative abundance of Acidobacteria, Gemmatimonadetes, Actinobacteria, and Chloroflexi was significantly increased by 167%, 888%, 86.7%, and 123%, respectively, after co-application of vermicompost and coconut-shell biochar in coastal saline soils. These results further indicated that vermicompost co-applied with coconut-shell biochar improved the microecological environment of coastal saline soil.

#### 4.3. Influential factors in salinity reduction of coastal soils

In this investigation, RDA analysis revealed that dominant bacterial families in coastal saline soils, including Gemmatimonadaceae, Nitrosomonadaceae, and Solibacteraceae\_Subgroup\_3, exhibited positive correlations with soil available nutrients (AN, AP, and AK), OM, pH, and enzyme activities (UE, ACP, and CAT) and negative correlations with soil TS. However, soil TS displayed negative correlations with soil OM, AN, AP, AK, pH, UE, ACP, and CAT (**Figure 4a,b**). Gemmatimonadaceae and Nitrosomonadaceae are tightly linked to the capacity for carbon (C) and nitrogen (N) cycling in soils [58,59], indicating that the reduction of soil salinity after vermicompost co-applied coconut-shell biochar to coastal saline soil is achieved by enhancing soil C and N cycling capacity. This enhancement is realized

by increasing the contents of OM and available nutrients and enhancing the enzyme activities of coastal saline soil. The findings further indicated that soil salinity was inversely related to soil bacterial richness and diversity (**Figure 5**). This suggests that vermicompost co-applied with coconut-shell biochar can also reduce soil salinity by increasing the richness and diversity of bacterial communities in coastal soils.

## 5. Conclusion

Vermicompost co-applied with coconut-shell biochar significantly alleviated soil saline stress and improved the micro-ecological environment of coastal soils by enhancing soil enzyme activity and nutrient availability, mainly through regulation of bacterial richness and diversity. Therefore, to address agricultural requirements for improving the well-being of coastal saline soils, organic amendments such as vermicompost and biochar should be applied in a rational, economical, and efficient manner.

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