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Xinzhuan Zhang

Shangrao Normal University

Zhongjun Hu

Shangrao Normal University

Huahua Pan

Shangrao Normal University

Yijun Bai

Shangrao Normal University

Ying Hu

Research Center for Eco-Environmental Sciences, Chinese Academy of Sciences

Shulan Jin (✉ jsl809@163.com)

Shangrao Normal University

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Effects of rare earth elements on bacteria in rhizosphere, root, phyllosphere and leaf of soil-rice ecosystem

Xinzhuan Zhang¹, Zhongjun Hu¹, Huahua Pan¹, Yijun Bai¹, Ying Hu², Shulan Jin¹✉

1. Shangrao Normal University, Shangrao, 334000, China.

2. Research Center for Eco-Environmental Sciences, Chinese Academy of Sciences, Beijing, 100085, China

✉ e-mail: jsl809@163.com

The effects of rare earth mining on rice biomass, rare earth element(REE) content and bacterial community structure was studied by pot experiment. The research showed that the REE content in rice roots, shoots and grains was significantly positive correlated with that in soil, and the dry weight of rice roots, shoots and grains was highly correlated with soil physical and chemical properties, nutrient elements and REE contents; The exploitation of rare earth minerals inhibited a-diversity of endophytic fungi in rhizosphere, root, phyllosphere and leaf of rice, significantly reduced the abundance index, OTU number, Chao, Ace index and also significantly reduced the diversity index-Shannon index and Inv-Simpson index, also reduced uniformity index: Pielou's evenness index, Simpson's evenness index, etc, which caused β -diversity of bacteria to be quite different. The exploitation of rare earth minerals reduces the diversity of bacteria, but forms dominant bacteria, such as *Burkholderia*, *Bacillus*, *Buttiauxella*, *Acinetobacter*, *Bradyrhizobium*, *Candida koribacter*, which can degrade the pollutants formed by exploitation of rare earth minerals, alleviate the compound pollution of rare earth and ammonia nitrogen, and also has the function of fixing nitrogen and resisting rare earth stress; The content of soil available phosphorus in non mining area is low, forming the dominant bacteria of *Pantoea*, which has the function of improving soil phosphorus availability. Rare earth elements and physical and chemical properties of soil affect the community structure of bacteria in rhizosphere and phyllosphere of rice, promote the parallel movement of some bacteria in rhizosphere, root, phyllosphere and leaf of rice, promote the construction of community structure of bacteria in rhizosphere and phyllosphere of rice, give full play to the growth promoting function of Endophytes, and promote the growth of rice. The results showed that the rice biomass in mining area was guaranteed due to the growth promoting effect of endophytic bacteria, However, the mining of mineral resources causes the compound pollution of rare earth and ammonia nitrogen, which makes REE content of rice in mining area significantly higher than that in non mining area.

China is a world power in the production, export and consumption of rare earth resources. China has about 50 percent of the world's rare earth resources and provides about 90 percent of the global supply of rare earth according to reports¹. The southern China, which is centered on Ganzhou City, Jiangxi Province, is an important distribution area of ion-type rare earth resources in the world, and its reserves account for about 70% of the world². The southern China is not only rich in ion rare earth resources, but also an important rice production area in the world. Illegal and disorderly exploitation of rare earth resources caused large amount of rare earth elements to enter the soil and water environment, which affects the growth of plants, animals and

microorganisms in the environment, thus changes the ecological environment and affects human health³⁻⁴ through the food chain. Rice is the main grain in southern China, and the yield and quality of rice are closely related to the ecological environment of farmland. Microorganism is the most sensitive indicator of farmland ecological environment. The microorganism is affected by climate factors, soil physical and chemical properties, plant species/genotypes, and also affected by heavy metal pollution⁵⁻⁶

With the extensive use of rare earth resources in many industries, the world's demand for rare earth resources increased from 136,000 t in 2017 to 151,000 t in 2020⁷. The increasing demand promotes the development and processing of rare earth. The impact of rare earth development and smelting on the environment has attracted much attention. It is reported that the REE content in soil of rare earth mining area in South China is 396-2314mg/kg⁸, which is much higher than the average REE content in soil of China. The concentration of rare earth in water is 1000 times⁹ higher than that of ordinary fresh water. Rare earth elements in ecosystems not only affect plants and animals, but also affect the growth, development, reproduction, migration and colonization of microorganisms¹⁰⁻¹¹. Rare earth elements affect the growth indexes of plants, including the biomass of aboveground and underground parts. It affects the index of plant resistance system, such as plant enzyme activity, etc. It also affect the REE content in plants¹²⁻¹³. The effect of rare earth elements on microorganisms is significant. The appropriate concentration of rare earth elements can promote the growth and reproduction of bacteria and the increase of metabolites. High concentration of rare earth can inhibit microorganism¹⁴. Chu Haiyan *et al.*¹⁵ found that La³⁺ has strong toxic effect on bacteria, actinomycetes, fungi and other microorganisms in the red soil microflora. He Qizhuang *et al.*¹⁶ found that rare earth elements had strong inhibitory effect on *Escherichia coli*, *Staphylococcus aureus* and *Candida albicans*. The research of Wang Yousheng *et al.*¹⁷ showed that the changes of soil REE content and physical and chemical properties significantly affected the diversity and abundance of soil bacteria. The research of Wang Jianhong¹⁸ showed that the microbial biomass in the areas with serious accumulation of rare earth was inhibited, the bacterial diversity in the non rare earth mining area was higher than that in the mining area, and the species were relatively rich. The REE content is the main factor affecting the microbial structure. Rare earth elements are very important for *methoxybacteria* in extremely acidic environment.¹⁹⁻²¹

After rare earth elements enter the soil, on one hand, they directly affect the microorganisms in the soil-plant ecosystem, on the other hand, they indirectly affect the microorganisms by influencing the physical and chemical properties of the soil and the plant biochemical system, while the microorganisms react on the soil and plants, and affect the migration and transformation of rare earth elements in soil and plant. Microorganisms in soil are oxidized, nitrated, ammoniated, nitrogen fixation and vulcanization, which promote the decomposition of soil organic matter and nutrient transformation. Endophytes have significant effects on plants, e.g. endophytic bacteria have nitrogen fixation effect on rice. The common endophytic azotobacters are *Pseudomonas*, *Spirulina*, *Enterobacteriaceae* and *Bacillus* to reduce the free N₂ in the atmosphere into the available NH₄⁺by nitrogenase²²⁻²³.The endogenous bacteria can secrete indoleacetic acid, which can promote the cell elongation, cell division and root growth of plants, assist host rice to improve the absorption efficiency of N, P and K, improve the production of gibberellin and salicylic acid in rice, and inhibit the production of jasmonate and abscisic acid to promote rice growth²⁴⁻²⁶. Endophytic bacteria can help to resist sheath blight, and the effect of disease resistance is better

than that of chemical fungicides. The mechanism of rice endophytes assisting host rice disease resistance mainly includes secretion of antibiotics, hyperparasitism, niche competition and induction of rice self-resistance²⁷⁻²⁸. To resist abiotic stress, endophytic bacteria can help heavy metal contaminated plants to carry out bioremediation, e.g. Zhou et al.²⁹ found that endophytic bacteria *Stenotrophomonas maltophilia* R5-5 significantly reduced the cadmium content in rice plants by down regulating the gene expression of cadmium absorption and transport proteins in rice and changing the community structure of endophytic bacteria. Endophytic bacteria can increase the biomass yield of host plants under different heavy metal stress, which indicates that Endophytic bacteria can effectively protect plants from the toxicity of many heavy metals (Cd, Cu, Cr, Mn, Ni, Pb and Zn).³⁰

At present, the research on the effect of endophytes on rice is mainly carried out in non polluted habitats, but the research on heavy metal polluted habitats is relatively lacking, and the research on rare earth polluted habitats has not been reported. In this study, the soil of typical REE mining area in Ganzhou City of Jiangxi Province was taken as the research object, and the soil of non mining area was taken as the control check (CK). The effects of rare earth elements on bacteria in rhizosphere, root, phyllosphere and leaf of soil rice ecosystem were discussed, which has scientific research value and practical significance.

Results

Effects of mineral exploitation on soil physical and chemical properties, REE content, rice biomass and REE content.

It can be seen from table 1 that the contents of rare earth elements, ammonia nitrogen, nitrate nitrogen, total P and available P in soil of H and M at both tillering stage and maturity stage of rice are significantly higher than those of L, and the pH value is lower than that of L. The results show that both H and M are soil samples in the mining area, and the mining of rare earth minerals can significantly improve the REE content in soil and affect the physical and chemical properties of soil.

Figure 1 Growth of rice at tillering stage

It can be seen from Fig. 1 that the tillering shoots of rice cultivated in soil M are the most at tillering stage, and the rice is dense and growing well; The rice cultivated in H soil has more tillers and leaves, which is more dense and grows better; The tillering shoots of rice cultivated in L soil were less and the growth vigor was general. It can be seen from Fig. 2 that the weight of root, shoot and grain of rice in soil M is the largest, with an average of 38.9g, 93.8g and 123.8g respectively, followed by H, with an average of 33.7g, 84.7g and 103.7g respectively, and then L, with an average of 22.8g, 52.1g and 63.7g respectively. The weight of root, shoot and grain of M

was 1.15, 1.11 and 1.19 times of H, and 1.71, 1.80 and 1.94 times of L, respectively.

Figure 2 Dry weight of grain, shoot and root of rice

Different letters indicate significant differences between potted rice in different soils

Figure 3 Content of rare earth elements in shoots and roots of rice at tillering stage and grains, shoots and roots at maturity stage

Different letters indicate significant differences between pot experiment rice in different soil (lowercase letters indicate significant differences in the same stage, capital letters indicate significant differences between the two stages), $P < 0.05$

It can be seen from Fig. 3 that the REE contents in different parts of rice at tillering stage and maturity stage are significantly positively correlated with the REE contents in soil. The REE contents in roots, shoots and grains of rice cultured in soil of H were the highest, followed by M, and L was the lowest. At tillering stage, the REE content in rice roots, shoots of H were 2.28 and 1.89 times that of M, 4.53 and 2.98 times that of L, respectively. At maturity stage, the REE content in rice roots, shoots and grains of H were 1.32, 1.91 and 1.17 times that of M, and 2.76, 2.83 and 1.33 times of L, respectively.

Effects of soil REE content and physicochemical properties on microbial diversity in rhizosphere, root, phyllosphere and leaf of rice.

Figure 4 Dilution curve of bacterial 16S gene sequence

Effect of soil rare earth elements on bacteria α -diversity, including abundance index: OTU number, Chao, Ace; Diversity index: Shannon Weiner index, Simpson's diversity index; Evenness index: Pielou's evenness index, etc. GN (H1, M1, L1), R (H1, M1, L1), YJ (H, M, L) and YN (H, M, L) in Fig. 4 respectively represent the bacteria in the root, rhizosphere, phyllosphere and leaf of rice at tillering stage, GN (H2, M2, L2) and R (H2, M2, L2) respectively represent the bacteria in the root and rhizosphere of rice at maturity stage, the same below. As can be seen from the Fig.4 that the number of OTU is affected by soil rare earth elements. The number of OTU in rhizosphere, root, phyllosphere and leaf of rice cultivated in soil of H and M of rare earth mining area is mostly lower than that of rice cultivated in soil L of non rare earth mining area. At tillering stage, the difference of OTU number among three different soil cultures was the largest. The OTU number of L sample on the same band number was the largest, followed by H, and the smallest was M. when the number of sequencing bands was 61,000, the OTU number of L sample was 2.23 and 2.34 times that of H and M, respectively. The OTU number of rhizosphere bacteria in L sample was the largest, followed by H and M. when the number of sequencing bands was 27,000, the OTU number of rhizosphere bacteria in L sample was 1.24 and 1.81 times that of H and M respectively. When the number of sequencing bands was 39,000, the OTU number of rhizosphere bacteria in L sample was 1.47 and 2.07 times that of H and M respectively.

As can be seen from the Fig.5 that the Shannon index of rhizosphere, root, phyllosphere and leaf of rice cultivated by H and M in mining area was lower than that of L in non mining area,

especially the Shannon index of rhizosphere and endophyllum of rice cultivated by H and M in tillering stage was significantly lower than that of L, which were 0.55 and 0.61 times that of L, respectively. The Simpson's evenness index of rhizosphere, root, phyllosphere and leaf in tillering stage and maturity stage of rice was affected by rare earth mining. The Simpson's evenness index of rhizosphere bacteria in H and M groups was 0.62 and 0.44 times higher than that of L in tillering stage, The Simpson's evenness index of M group was 0.44 times that of L and H. The Chao1 index of rhizosphere, root, phyllosphere bacteria in M group was lower than that in L group at tillering stage and maturity stage, and the significant difference was that the bacteria in rhizosphere and root at tillering stage were 0.53, 0.46, 0.52 and 0.48 times that in L group, respectively. The richness of most samples in group H and M was lower than that in group L. the richness of rhizosphere and endophytic bacteria in group H and M was significantly lower than that in group L at tillering stage and maturity stage, which were 0.54, 0.42, 0.72, 0.43, 0.74, 0.52, 0.65 and 0.48 times that in group L, respectively. The inv-Simpson of rhizosphere bacteria in group H and M was significantly lower than that in group L (0.34, 0.18, 0.44 and 0.18 times of that in group L), while inv-Simpson of rhizosphere bacteria in group M was significantly lower than that in group L (0.32 and 0.20 times of that in group L). Mining reduced the Pielou's evenness index of bacteria, and the most important factor was the leaf bacteria at tillering stage. The index of H and M groups was only 0.56 and 0.67 times of L, respectively.

Figure 5 α -diversity of bacteria in root, rhizosphere, leaf and phyllosphere of rice at tillering and maturity stages

Notes: Different letters indicate the significant difference among bacteria of different soil pot-experiment rice (small letters indicate the significant difference in the same stage, capital letters indicate the significant difference between the two stages), $P < 0.05$

Figure 6 β -diversity of bacterial community structure of pot-experiment rice in different soils based on NMDS analysis

A and B are the results of NMDS analysis at tillering stage and maturity stage respectively in Figure 6. It can be seen from Figure 6 that the bacteria in different parts of rice cultivated in the same soil have a greater degree of dispersion, that is, the bacteria in rhizosphere, root, phyllosphere and leaf are dispersed in both tillering stage and maturity stage; The bacteria in the

same part of rice cultivated in soils with different REE contents were also dispersed, especially the difference between H and M in mining area and l in non mining area was larger. In Fig. 6, the stress values of A and B are 0.087 and 0.058, respectively, which are less than 0.1, indicating that the difference of NMDS Bray Curtis is significant. The effect of rare earth mining on β -diversity of bacteria is obvious.

Effects of soil REE content and physicochemical properties on bacterial community structure in rhizosphere, root, phyllosphere and leaf of rice

Figure 7 Bar chart of the phylum level

It can be seen from Fig.7 that there are Proteobacteria, Firmicutes, Actinobacteria, acidobacteria, etc. in rhizosphere, root, phyllosphere and leaf of rice at tillering stage and in rhizosphere, root of rice at maturity stage. Exploitation of rare earth mining has significant effect on Proteobacteria of rice at tillering stage. The relative abundance of this phyla in group H and M is much higher than that in L, which is 4.33 and 5.77 times that in L, respectively; The relative abundance of Firmicutes in rhizosphere of H and M groups was significantly higher than that of L at tillering stage, and the number of bacteria in leaves was significantly lower than that of L. the former was 3.47 and 3.95 times of L, and the latter was 0.37 and 0.11 times that of L, respectively; The relative abundance of H and M was 0.61 and 0.14 times higher than that of L, respectively; The relative abundance of H and m was 4.40 and 1.44 times higher than that of L, respectively. The relative abundance of Actinobacteria in rhizosphere of group H and M was 0.29 and 0.50 times that of group L, respectively. The relative abundance of Actinobacteria in rhizosphere and root of group H and M was 1.81 and 2.15 times that of group L, and 1.88 and 4.62 times that of

group L, respectively.

Figure 8 Heatmap of genus level

As can be seen from Fig.8 that the exploitation of rare earth significantly increased the relative abundance of *Ralstonia* and *Burkholderia* in rice roots at tillering stage, and decreased the relative abundance of *Pantoea*, *Acidovorax*, *Azospira*, *Buttiauxella* and *Kosakonia* in roots. The relative abundance of *Ralstonia* in roots of H and M groups was 63.42 and 7.5 times that of L, and the relative abundance of *Pantoea* in roots of L group was 3.29 and 8474 times that of H and M group, respectively. The relative abundances of *Burkholderia* in M group were 9.23 and 87.71 times higher than those in L group, respectively. The relative abundances of *Bacillus*, *Clostridium sensu stricto*, *Azospira*, *Sideroxydans*, *Cellulomonas* and *Candida solibacter* in rice rhizosphere increased, and the relative abundances of *Bacillus* and *Pantoea* in rice phyllosphere also increased, while the relative abundances of *Exiguobacterium*, *Buttiauxella*, *Acinetobacter* and *Staphylococcus* decreased. The relative abundance of *Bacillus* in group M was 8.01 and 8.28 times of that in group L, respectively. The relative abundance of *Exiguobacterium* and *Buttiauxella* in group L was 44.41, 194.48, 100.83 and 2.11 times of that in group H and M, respectively. The relative abundance of *Acinetobacter* and *Comamonas* in leaves increased, and the relative abundance of *Pantoea*, *Exiguobacterium*, *Weissella* and *Staphylococcus* in leaves of group L decreased. The relative abundances of *Pantoea*, *Exiguobacterium*, *Weissella* and *Staphylococcus* in leaves of group L were 4.72, 55.03, 61.81, 2.88, 45.93, 454.58, 33.00 and 31.43 times of those in leaves of group H and M, respectively. In addition, *Herbaspirillum* in roots of group H was significantly higher than that in roots of group L at tillering stage, which was 7.08 times of that in roots of group L, *Bacillus* in leaves of group H was significantly higher than that in leaves of group L,

which was 3.61 times of that in leaves of group L, and *Buttiauxella* in group M was significantly higher than that of group L, which was 15.14 times that of group L.

As can be seen from Fig. 8 that the relative abundances of *Bradyrhizobium*, *Burkholderia* and *Anaeromyxobacter* in rice roots at maturity stage significantly increased due to the exploitation of rare earth, while the relative abundances of *Georgfuchsia*, *Pelomonas*, *Methylocystis*, *Streptomyces* and *Methylosinus* decreased significantly. The relative abundances of *Bradyrhizobium* in H and M groups were 3017 and 11.89 times that of L, respectively. The relative abundance of *Methylosinus* was 5.24, 11.66, 6.20 and 6.34 times that of H and M, respectively. The relative abundance of *Bradyrhizobium*, *Burkholderia*, *Geobacter*, *Denitisoma* and *Candidatus Koribacter* in rhizosphere increased, while that of *Gaiella* decreased. The relative abundance of *Geobacter*, *Candidatus koribacter* and *Candidatus solibacter* in rhizosphere of H and M was 1.73, 2.68, 3.57 and 64.18 times that of L, respectively, The relative abundance of *gaiella* in rhizosphere of group L was 601.5 and 601.5 times that of H and M, respectively.

Environmental factors affecting bacterial community of rhizosphere, root, phyllosphere and leaf in soil

Figure 9 Canonical Correspondence Analysis (CCA) and variation partition analysis (VPA) of rice bacterial community

The bacterial community structure and construction process were affected by environmental

factors. The relationship between bacterial community and environmental factors was analyzed by CCA. The results are shown in Fig. 9. Due to the differences in REE content and physicochemical properties of soil samples, each group of samples is obviously separated. Fig. 9A shows the environmental factors affecting the three groups of samples in H, M and L at the tillering stage of rice. It can be seen from the figure that the main environmental factors are ammonia N, nitrate N, organic matter and available K.

Fig. 9b shows the environmental factors affecting the three groups of samples H, M and L at the maturity stage of rice. It can be seen from the figure that the main environmental factors are the contents of rare earth elements in shoots, grains, ammonia nitrogen, organic matter and total nitrogen. According to Table 1, we can see that Shannon, Inv_Simpson, Richness, Pielou evenness of rice bacteria in rhizosphere, root, phyllosphere and leaf at tillering stage were significantly negatively correlated, highly negatively correlated or negatively correlated with REE content, total N, ammonia N, nitrate N, total P and available P in soil, rice roots, shoots, and highly positively correlated with pH. Simpson evenness was significantly negatively correlated with REE contents in rice roots, shoots, negatively correlated with REE contents in soil, ammonia and nitrogen; and positively correlated with available K, organic matter and pH. Chao. value was highly positively correlated with REE contents in roots, shoots of rice, and positively correlated with available K, organic matter and pH. As can be seen from Table 2 that the REE content in soil, rice roots, shoots, grains and soil physical and chemical properties are negatively correlated with the diversity, richness and evenness of bacteria in rhizosphere, root, phyllosphere and leaf at maturity period of rice, and significantly or highly negatively correlated with REE contents, ammonia N, nitrate N and pH in soil. As can be seen from Table 3 that the dry weight of rice roots, shoots, and grains are positively correlated with REE content, total N, ammonia N, nitrate N, total P, available P, available K, and organic matter in soil, rice roots, shoots, and negatively correlated with pH.

Table 1 Pearson correlation of rare earth element content, soil physical and chemical properties and α -diversity of rice bacteria at tillering stage

	Shanno n	Inv_ Simpson	Observed_ richness	Pielou_ evenness	Simpson_ evenness	chao.value
Soil REE content	-1.000*	-0.981	-0.812	-0.991	-0.751	0.521
Root REE content	-0.806	-0.893	-0.28	-0.865	-.998*	0.936
Shoot REE content	-0.815	-0.899	-0.294	-0.872	-.997*	0.931
Total N	-0.327	-0.166	-0.834	-0.224	0.35	-0.613
ammonia N	-0.987	-0.946	-0.882	-0.964	-0.657	0.404
nitrate nitrogen	-0.794	-0.683	-1.000**	-0.724	-0.225	-0.075
Total P	-0.559	-0.415	-0.948	-0.467	0.097	-0.388
available P	-0.47	-0.318	-0.91	-0.373	0.2	-0.482
available K	0.261	0.417	-0.38	0.363	0.816	-0.951
organic matter	0.272	0.427	-0.369	0.374	0.823	-0.955
pH	0.949	0.883	0.945	0.909	0.53	-0.254

** At 0.01 level (two tailed), the correlation was significant, *. At 0.05 level (two tailed), the correlation was significant, n = 3

Table 2 Pearson correlation of rare earth element content, soil physical and chemical properties and α -diversity of rice bacteria at maturity stage

	Shannon	Inv_ Simpson	Observed_ richness	Pielou_ evenness	Simpson_ evenness	chao.value
Soil REE content	-0.914	-0.962	-0.957	-0.892	-0.914	-0.981
Root REE content	-0.754	-0.839	-0.83	-0.72	-0.755	-0.88
Shoot REE content	-0.441	-0.562	-0.549	-0.394	-0.442	-0.628
Grain REE content	-0.589	-0.696	-0.685	-0.546	-0.589	-0.752
Total N	-0.695	-0.588	-0.6	-0.731	-0.695	-0.52
ammonia N	-0.905	-0.956	-0.951	-0.882	-0.905	-0.976
nitrate nitrogen	-.999*	-0.983	-0.986	-1.000**	-.999*	-0.964
Total P	-0.786	-0.692	-0.703	-0.816	-0.785	-0.63
available P	-0.797	-0.705	-0.715	-0.827	-0.796	-0.644
available K	-0.704	-0.598	-0.61	-0.739	-0.703	-0.53
organic matter	-0.777	-0.682	-0.693	-0.809	-0.777	-0.62
pH	0.994	1.000*	1.000**	0.987	0.994	0.994
Root weight	-1.000**	-0.991	-0.993	-.998*	-1.000**	-0.976
Leaf weight	-0.993	-1.000*	-1.000**	-0.986	-0.993	-0.994
Grain weight	-1.000**	-0.989	-0.991	-.999*	-1.000**	-0.973

**. At 0.01 level (two tailed), the correlation was significant, *. At 0.05 level (two tailed), the correlation was significant, n = 3

Table 3 Pearson correlation of rare earth element content, soil physical and chemical properties and dry weight of root, shoot and grain of rice

	nitrate nitrogen	Total P	availa ble P	avail able K	organic matter	pH	Total N	amm onia N	Soil REE content	Root REE content	Shoot REE content	Grain REE content
Root weight	.999*	0.783	0.794	0.701	0.775	-0.99 4	0.693	0.907	0.915	0.757	0.444	0.592
Leaf weight	0.987	0.708	0.721	0.616	0.699	-1.00 0**	0.607	0.948	0.955	0.826	0.542	0.679
Grain weight	.999*	0.791	0.802	0.71	0.783	-0.99 3	0.702	0.901	0.91	0.748	0.433	0.581

**. At 0.01 level (two tailed), the correlation was significant, *. At 0.05 level (two tailed), the correlation was significant, n = 3

4. Discussion

Previous studies have shown that lower concentrations of rare earth can promote the growth of crops, while higher concentrations can inhibit or even harm the growth of crops^{12-13,32}. In this study, the shoots of rice tillers cultivated in soil of H and M in rare earth mining area were significantly more than those in non mining area, especially rice of M with medium rare earth content grew the most densely. The dry weight of rice root, shoot and grain of soil H and M was

significantly heavier than that of soil L. M was the heaviest, followed by H, and L was the lightest. The results are basically consistent with previous studies. Due to extraction of ionic rare earth by $(\text{NH}_4)_2\text{SO}_4$ leaching, ammonia N in mining area seriously exceeded the standard, pH value decreased, which caused dry weight of root, shoot, grain of cultivated rice in mining area to have significant negative correlation with nitrate nitrogen, and negative correlation with total nitrogen, ammonia nitrogen, and significant positive correlation with pH. A large number of studies have shown that there is a significant positive correlation between the REE content in crops and that in soil²⁻⁵. In this study, the content of rare earth elements in soil is $\text{H} > \text{M} > \text{L}$, and the content of rare earth elements in rice root, shoot and grain is $\text{H} > \text{M} > \text{L}$, which is basically consistent with the previous research results.

Chao *et al.*²¹ considered that the REE content is the main factor affecting the microbial structure in rare earth mining area. Jiang Zhaowei *et al.*³³; Zhou *et al.*³⁴ thought that high concentration of rare earth elements had inhibitory effect on bacteria. Yang Jun *et al.*³⁵ showed that when the concentration of rare earth ions reached a certain value, it had antibacterial effect, and its antibacterial effect was selective. Some studies have shown that exogenous rare earth element Yttrium destroyed the structure of soil microbial community and significantly reduced OTUs, Ace, Chao1 and Shannon index³⁶. Chen Xi *et al.*³⁷ found that rare earth mining seriously damaged the soil quality of the mining area, caused low diversity and evenness of microbial community. In this study, the OTU number of rice bacteria in rhizosphere, root and phyllosphere cultured in soil H and M of rare earth mining area was lower than that in soil L of non rare earth mining area at tillering stage and maturity stage. In addition, the number of OTUs of bacteria in leaf of rice cultured in soil M was significantly lower than that in soil L. The richness and Chao indexes of bacteria in rhizosphere, root and phyllosphere of rice cultured in soil H and M were significantly lower than those in soil L at tillering stage and maturity stage. The Shannon index of H and M was significantly lower than that of L. The Pielou's evenness index and Simpson's evenness index of H and M were lower than that of L. It shows that the exploitation of rare earth reduces α -diversity of rice bacteria, which is consistent with the previous research conclusions of many scholars. Mineral exploitation affects the REE content in soil, soil physical and chemical properties and rice growth, caused β -diversity of bacteria to be quite different.

Many studies have shown that soil heavy metals, physical and chemical properties, nutrient elements are important factors affecting microbial community structure³⁹⁻⁴⁰. Wei Zhiwen⁴¹ found that rare earth pollution affected the abundance of bacteria and fungi and changed their community structure. Wang Yousheng⁴² found that the contents of bacteria, fungi and *Actinomyces* decreased significantly, and the abundance and diversity of soil bacteria changed significantly. However, the dominant species of soil bacteria was Firmicutes (phylum), which was related to *Proteus*, *Actinomyces*, *Bacteroidetes* and *Acidobacteria*. Firmicutes (phylum) are affected by rare earth elements^{42,3,5}. In this study, the abundance of Firmicum (phylum) in rice rhizosphere of H and M at tillering stage was significantly higher than that of L, and the abundance of Firmicum (phylum) in rice rhizosphere of H was significantly higher than that of M and L; The abundance of L was significantly higher than that of H and M; The abundance of Firmicutes in rice rhizosphere of L was significantly lower than that of H and M. It can be seen that Firmicutes (phylum) in soil-plant system are sensitive to rare earth elements, but the mechanism is very complex. Actinobacteria and Acidobacteria are greatly affected by soil physical and chemical properties. The former is directly proportional to pH value, while the latter is

inversely proportional to pH value^{3,5}. The pH value of rare earth mining area was reduced by mineral exploitation, and the abundance of Actinobacteria in rice rhizosphere of L was significantly higher than that in H and M, and the abundance of acidobacteria in rice rhizosphere of L was significantly lower than that in H and M.

The exploitation of rare earth minerals significantly reduced the diversity and evenness of soil-plant system microorganisms, and enhanced the resistance and growth of some bacteria, resulting in the formation of some dominant bacterial genera in rare earth mining area. For example, due to mineral exploitation, a large amount of acid mineral water was formed, and the abundance of *Candida solibacter* and *Candida koribacter* in rice rhizosphere of M soil was higher⁴³. Many dominant bacteria can promote the growth of rice, increase the yield of rice, and make the dry weight of rice root, shoots and grain be highly or extremely significantly negatively correlated to α -diversity of bacteria. Some scholars believe that the biological effect of rare earth ions may be similar to that of Ca ions because the radius of rare earth ions is close to that of Ca ions, Rare earth ions may occupy or replace Ca ions in organisms⁴⁴⁻⁴⁵. Some researchers have explored the mechanism of the interaction between rare earth ions and microorganisms at the protein level. They believe that rare earth ions can affect the production of differential proteins by microorganisms, thus controlling the growth, reproduction and metabolism of microorganisms⁴⁶⁻⁴⁷. The differential proteins mainly include transporters, stress-related proteins, metabolism related proteins, etc. Chai Ruijuan *et al.*⁴⁸ found that the effect of rare earth ions on promoting microbial reproduction was inhibited by chlorpromazine, indicating that the mechanism of La and Ce promoting bacterial growth may be related to calmodulin.

Soil environment, plants and microorganisms interact with each other, soil and plants affect bacteria, and bacteria also react on plants and soil⁴⁹. Bacteria in soil-plant system can be divided into beneficial bacteria, neutral bacteria and harmful bacteria. Harmful bacteria harm plant growth, and beneficial bacteria can play an important role in promoting plant growth, nutrient absorption and stress response⁵⁰⁻⁵³. Due to mineral exploitation, the REE content and ammonia nitrogen in soil increased significantly, the vegetation was destroyed, and the content of organic matter was low, which affected the ecological environment⁵⁴. At tillering stage, *Ralstonia* abundance in root, rhizosphere, leaf of rice in H soil was high, accounting for 17.8%, 2.18% and 8.12% respectively. *Ralstonia* is a plant pathogenic bacteria, affecting the growth of rice⁵⁵, which may be one of the reasons why the dry weight of root, shoot and grain of group H is lower than that of group M. The abundance of *Herbaspirillum*, *Azospirillum* and *Bacillus* in rice root, the abundance of *Azospirillum* in rice root and rhizosphere, The abundance of *Bacillus* in rice root, rhizosphere, leaf and phyllosphere of H is relatively high. Nitrogen fixation of *Herbaspirillum* and *Azospirillum* can promote plant growth, nutrient absorption and photosynthetic efficiency⁵⁶⁻⁵⁸. *Bacillus* has strong growth promoting effect on rice, potential antibacterial activity and strong antagonistic activity. It can produce iron carrier in iron limited medium and produce four kinds of secondary metabolites, causing damage to cell membrane of *Magnaporthe grisea*⁵⁹⁻⁶⁰. These beneficial bacteria may be an important reason why rice biomass in H group is significantly higher than that in L group.

Because of mineral exploitation, soil rare earth elements, total nitrogen, ammonia nitrogen and nitrate nitrogen in M were significantly higher than those in non mining areas, pH decreased, soil acidity increased, forming a compound pollution of rare earth and ammonia nitrogen, causing bacterial species to be monotonous. At tillering stage, *Burkholderia* abundance in rice root and rhizosphere of M was very high, and *Acinetobacter*, *Buttiauxella* and *Comamonas* abundance in

rice leaf was very high. *Burkholderia* is a microorganism with the ability to degrade ammonia nitrogen efficiently. It can transform ammonia nitrogen into substances needed by itself or beneficial to the environment. It has the functions of biological control, promoting plant growth, decomposing toxic substances and bioremediation. It is a potential genus for remediation of high ammonia nitrogen pollution in rare earth mine wasteland in South Jiangxi Province⁶¹. *Acinetobacter* can improve the resistance of plants to Cu, the ability to remove Ni and the ability to separate rare earth elements⁶²⁻⁶⁴. *Buttiauxella* could significantly increase the biomass and chlorophyll content of shoots and roots of plants treated with Cd⁶⁵ and *Comamonas* also had a good ability to degrade a variety of environmental pollutants⁶⁶. These beneficial bacteria may be an important reason for the higher biomass of rice in M than that in H and L.

In the maturity stage, *Burkholderia*, *Bradyrhizobium*, *Ideonella* and *Pleomorphomonas* were found in rice rhizosphere of H. the abundance of *Geobacter* and *Denitisoma* was higher. The abundance of *Bradyrhizobium* in rice roots of M was especially high in maturity stage, and the abundance of *Geobacter* and *Candida_koribacter* in rice rhizosphere was high. *Bradyrhizobium Rhizobium* not only colonizes in legumes, but also in rice. The symbiosis between *Bradyrhizobium Rhizobium* and rice can achieve biological nitrogen fixation, which plays an important role in ensuring food security and ecological construction⁶⁷. These fungi may be another important reason why rice yield of H and M is significantly higher than that of L.

The contents of available P and ammonia nitrogen in L soil were significantly lower, and the relative abundances of *Pantoea* and *Acidovorax* in rice roots were higher at tillering stage. *Acidovorax* is the pathogen of brown streaked disease of rice⁶⁸, which may be one of the reasons why rice yield of L is lower than that of H and M. *Pantoea* can promote the growth of alfalfa, promote the growth of wheat root and shoot, improve the availability of phosphorus in soil, and reduce the application of fertilizer⁶⁹. The relative abundance of *Exiguobacterium* in rice phyllosphere of L is high. Some studies have shown that it has broad-spectrum antibacterial effect on Gram-positive and gram-negative foodborne pathogenic bacteria. It has antagonistic effect on a variety of plant pathogenic fungi. It shows the ability of nitrogen fixation, phosphate solubilization and iron carrier generation⁷⁰⁻⁷¹. The relative abundance of *Weissella* in leaves is high, which is beneficial bacteria⁷². The relative abundances of *Bradyrhizobium*, *Ideonella*, *Methylocystis*, *Streptomyces* and *Georgfuchsia* were higher in rice roots at maturity stage. *Methylocystis* can inhibit the growth of *Staphylococcus albicans* and *Bacillus subtilis* with spores, produce single cell protein, and have an important impact on the global C cycle⁷³. *Streptomyces* has the potential of biocontrol against plant pathogenic fungi⁷⁴. *Georgfuchsia* is an aromatic compound degrading bacterium in aquifers⁷⁵. These beneficial bacteria may be an important reason for the healthy growth of L rice under the condition of low soil fertility.

At tillering stage, *Ralstonia* was relatively abundant in rice root of H soil and moved vertically to leaf, resulting in higher relative abundance of *Ralstonia* in leaf; The relative abundance of *Ideonella* and *Bacillus* in rice rhizosphere was high, and moved horizontally to the root and leaf of rice, resulting in higher relative abundance in root and leaf; The relative abundance of *Burkholderia* and *Buttiauxella* in the rice rhizosphere of M was high, and moved horizontally to the root and leaf of rice, resulting in higher relative abundance of *Burkholderia* and *Buttiauxella* in the root and leaf; The relative abundance of *Exiguobacterium* in rice phyllosphere of L was high, and it moved horizontally into leaf, so that the relative abundance of *Exiguobacterium* in the leaf was also high. The relative abundance of *Azospira*, *Anaeromyces*

Obacter and *Georgfuchsia* in the rice rhizosphere of H was higher in the maturity stage, and moved horizontally to the root, which made the relative abundance of *Azospira* higher in the root. The horizontal movement of these bacteria makes them play an active role in the process of rice growth and become an important reason for the healthy growth of rice⁷⁶.

Conclusion

1) The REE contents in rice roots, shoots and grains were significantly positively correlated with those in soil, causing the REE contents in all parts of rice in mining area to be significantly higher than those in non mining area. The dry weight of rice roots, shoots and grains was highly correlated with soil physical and chemical properties, nutrient elements and rare earth elements.

2) The exploitation of rare earth minerals inhibited α -diversity of bacteria in rhizosphere, root, phyllosphere and leaf of rice, significantly reduced the abundance index of OTU number, Chao1 and Ace index, decreased the diversity index of Shannon and inv-Simpson index, reduced the evenness index: Pielou's event index, Simpson's evenness index, etc., which caused β -diversity of bacterial to be different; Rare earth elements enhance the resistance and growth of some bacteria, and form some dominant bacteria genera in the rare earth mining area. They play an important role in promoting plant growth, nutrient absorption and stress response, which has become an important reason for the dry weight of rice cultivated in the soil of H and M in the mining area is significantly higher than that of L in non mining area.

3) Rare earth minerals reduce the diversity of bacteria, but form the dominant bacteria, such as *Burkholderia*, *Bacillus*, *Buttiauxella*, *Acinetobacter*, *Bradyrhizobium*, *Candida koribacter*, which can degrade the pollutants formed by rare earth mining, alleviate the compound pollution of rare earth and ammonia nitrogen, and have the functions of nitrogen fixation and resistance to rare earth stress; The content of soil available phosphorus in non mining area is low, forming the dominant bacteria of *Pantoea*, which has the function of improving soil phosphorus availability.

4) Soil rare earth elements and soil physical and chemical properties affect the community structure of bacteria in rhizosphere and phyllosphere of rice, promote the parallel movement of some bacteria in rhizosphere, root, phyllosphere and leaf of rice, promote the construction of community structure of bacteria in rhizosphere and phyllosphere of rice, give full play to the growth promoting function of Endophytes, and promote the growth of rice.

Materials and methods

Soil collection and material preparation

The sampling point of rare earth mining area are 1500 meters away from REE mining area (latitude and longitude is 115°42'6"E, 24°52'9"N) in Shipai village, Wenfeng Township, Xunwu County, southern Jiangxi Province; 1500 meters away from REE mining area (latitude and longitude is 114°59'36"E, 25°23'57"N) in Longshe village, Jiading town, Xinfeng County, southern Jiangxi Province and the sampling point of non REE mining area is located in Choukou village, Maojialing Township, Xinzhou District, Shangrao City of Jiangxi Province (longitude and latitude is 117°57'46"E, 28°25'1"N). 50 kg of 20 cm topsoil were collected at three sampling points respectively. Air dry the soil sample naturally, remove the grass roots, litter, stones and other debris, and sift through 100 mesh for standby. The tested soil was loaded into a 26 cm high

and 22 cm diameter rigid PVC bucket with 5 kg of soil, 5.4 g of urea (containing 46.6% N), 0.6 g of potassium chloride (containing 62.9% K₂O) and 1.00 g of superphosphate (containing 14.0% P₂O₅). The soil was mixed with N, P and K fertilizers, and rice was planted after a week of submergence.

Rice pot experiment and sample collection

Rice variety Jiangzao 361 was purchased from Jiangxi Keyuan Seed Industry Co., Ltd. The whole growth period of this variety is 110.2 days. After participating in the regional test of rice in Jiangxi Province from 2013 to 2014, it is widely planted in Jiangxi Province. The seeds were disinfected with 10% H₂O₂ solution for 15 min, then washed with water for many times.

After germination, the seeds were sown in the soil without rare earth pollution for seedling raising. Four weeks later, the rice seedlings were transplanted into PVC buckets with culture soil prepared. Two seedlings were transplanted into each barrel. The pot experiment was carried out in the greenhouse. The daily illumination, duration of illumination, temperature and humidity of the greenhouse were adjusted to be close to the outdoor. According to the growth characteristics of Jiangzao 361 rice variety, the pot-experiment rice was managed in combination with the experience and methods of rice planting management in Jiangxi Province. According to the REE content in soil, the samples were labeled as H, M and L. The soil with highest REE content was H, the lowest was L, and the one between them was M. Because three replicates were made for each sample, the pot-experiment rice was labeled with H1, H2, H3, M1, M2, M3, L1, L2, L3 respectively.

Rhizosphere soil, rice roots, shoots were collected at tillering stage, and rhizosphere soil and rice roots were collected at maturity stage, which was used for molecular experiments.

After maturity of rice, the roots, shoots and millet of rice were harvested respectively, washed with deionized water, then placed in the ventilated room and dried naturally until constant weight and weighed. Husk the millet and collect the unpolished rice. The unpolished rice, shoot and root were ground into powder and stored at room temperature for analysis.

Chemical analysis

The soil physical and chemical properties and nutrient elements are tested by the relevant detection methods in the Agricultural Industry Standard of the People's Republic of China (2006). Soil pH determination: NY/T 1121.2-2006, determination of total P in soil: alkali soluble molybdenum antimony anti spectrophotometry HJ632-2011, determination of soil available phosphorus: sodium bicarbonate extraction molybdenum antimony anti spectrophotometry HJ704-2014, determination of soil ammonia nitrogen, nitrite nitrogen and nitrate nitrogen: potassium chloride solution extraction spectrophotometry HJ634-2012, determination of soil available K and slow acting potassium content: NY/t889-2004, Determination of soil organic matter: NY/T 1121.6-2006. The test results can be seen in Table 4.

Add 0.25 g of soil sample in a 100 ml digestion tube (three replicates for each sample), add 5 ml aqua regia (HNO₃:HCl = 3:1), balance overnight at room temperature, and then heat to 120 °C for 12 h, then warm to 140 °C until the soil color turns white, and the digestion procedure refers to reference 3. After digestion, the sample was cooled in a fume hood and diluted to 50ml with ultrapure water. After filtration with 0.45μm membrane, the contents of 15 rare earth elements (Y, La, Ce, Pr, Nd, Sm, Eu, Gd, Tb, Dy, Ho, Er, Tm, Yb, Lu) were determined by ICPMS-2030 (Shimadzu Corporation, Shimadzu Institute, Kyoto, Japan). The quality was monitored by the standard reference substance GBW07405 of the national standard substance research center. The

test results are shown in Table 1.

Add 0.2g of crushed unpolished rice, 0.1g of shoot, and 0.1g of root to a 50-ml polyethylene centrifuge tube (three replicates for each sample) respectively, soaked in 3 ml of superior pure nitric acid overnight, and digested in a microwave digestion oven (Mars, Matthew Inc., USA), The whole process of digestion was controlled by GBW07603 (GSV-2), a national first-class reference material (Institute of Geophysical and Geochemical Exploration, Ministry of Geology and Mineral Resources of China). The digestion solution was diluted to 50ml with ultrapure water, filtered with 0.45μm membrane. The contents of 15 rare earth elements were determined by ICPMS-2030.

Table 4 Soil physical and chemical properties, nutrient elements and REE content (mg/kg)

Sample Name	pH	Total N	Ammonia N	Nitrate N	Total P	Available P	Available K	organic matter (%)	REE content
H1	5.28	1351.96	308.37	29.7	338.9	13.59	70.82	2.81	916.56
L1	6.58	1425.36	198.08	24.98	114.49	4.03	77.32	4.58	140.13
M1	4.97	2624.05	311.43	34.27	1141.24	87.1	81.5	5.67	824.05
H2	5.52	1425.31	124.37	29.33	180.6	9.9	83.34	4.11	805.64
L2	6.63	1551.15	38.55	25.33	112.93	1.44	85.49	4.05	110.33
M2	5.2	2839.02	113.75	31.65	1220.12	106.47	111.49	5.34	735.09

Note: H1, M1 and L1 are rice and soil samples in tillering stage, H2, M2 and L2 are rice and soil samples in maturity stage, the same below.

Pretreatment and DNA extraction of soil and plant samples

The loose and massive soil of rice root samples were removed, and then immersed in 15 ml PBS buffer containing Silwet L-77 (pH 7.0, 0.02% Silwet L-77) and shook in shaking table for 10 min. Then the suspension obtained was poured into a sterile centrifuge tube and centrifuged at 6000 g for 5 min. The precipitates were taken as rhizosphere soil samples and stored at -80 °C for subsequent DNA extraction. After shaking, the roots were washed in 75% ethanol for 10 min, 2.5% sodium hypochlorite for 10 min, and finally washed in sterile water for 5 times to remove the residual microorganisms on the root surface (scanning electron microscope was used to check whether it was clean). The cleaned roots were defined as root samples^{77,78}. The root samples were ground in PBS with sterile mortar and pestle, washed into the centrifuge tube and left for 30 minutes. The supernatant was centrifuged at 6000 g for 5 min, and the cell particles obtained (endophyte samples) were stored at - 80 °C for subsequent DNA extraction.

The leaves were placed in a sterile conical flask (250 ml volume) with 100 ml PBS (pH 7.0, 0.02% Silwet L-77) and stirred violently for 30 min. the leaves were then ultrasonically washed in water bath for 10 min. The supernatant (phylobacteria sample) was concentrated at 0.22 μM nitrocellulose membrane filter. Before extraction of DNA, the membrane was stored at -80°C. Then the leaves were washed in 75% ethanol for 3 min, 2.5% sodium hypochlorite for 5 min, and washed with sterile water for 5 times. Then the leaves were ground in PBS with sterile mortar and pestle, washed into the centrifuge tube and left for 30 minutes. The supernatant was transferred to a new centrifuge tube and centrifuged at 6000 g for 5 min. The extracted cell microspheres (endophyte samples) were stored at -80°C before DNA extraction.

The soil samples of rice rhizosphere were pretreated with propidium monoazide (PMA) to remove the DNA interference of dead microorganisms⁷⁹. Then, the DNA of bacteria sample in

rhizosphere soil, root, phyllosphere and leaf was extracted by FastDNA™SPIN Kit for Soil(MP Biomedicals LLC, USA). The DNA concentration and purity were determined by nanodrop 2000, and then stored at -20°C in refrigerator for subsequent 16S rRNA gene analysis.

Amplification and sequencing of 16S rRNA gene

The V4 region of 16S rRNA was amplified by bacterial specific primers 515F(5'-GTGCCAGCMGCCGCGGTAA-3') and 806R (5'-GGACTACHVGGGTWTCTAAT-3'). Phusion high fidelity enzyme (high fidelity PCR master with GC buffer) was used in PCR. The process was as follows: pre-denaturation for 5 min in 95°C, denaturation for 30s in 94°C , annealing for 35s in 55 °C, extension for 30s in 72 °C . there were 30 cycles above, then extension for 8 min in 72 °C. The gel Life Technology (USA) was used for recovery and purification. After being qualified by gel electrophoresis, the accurate double stranded DNA concentration was quantified by Qubit3.0 (Life Technology, USA).

The NEB kit was used to construct the library, The standard database building process of DNA Library Prep Kit for Illumina. NEBNext Ultra (NEB#e7370S/L) was adopted. The length distribution and concentration of the constructed library was detected by agilent2100 and qPCR, respectively. The qualified library was sequenced by Illumina hiseq2500 (PE250 reads). The sequencing samples included soil and plant samples (rhizosphere soil samples, phyllospheric bacteria samples, endophytic bacteria samples in leaves and endophytic bacteria samples in roots) at tillering stage and maturity stage. The data obtained after sequencing were used for quality control.

The 16S rRNA gene sequences obtained by high-throughput sequencing were pre-treated and quality controlled by QIIME2 software. Quality control adopted the parameters recommended by Bokulich *et al.*³¹ According to the sequence similarity, they were classified into multiple OTUs. Those gene sequences with similarity level of 0.03 (97% sequence similarity, approximately equal to species level) were selected for subsequent analysis, and the OTU representative sequence was annotated. The reference sequence from Silva database (db128 version) is used, the command of mothur software (classify. SEQS) is used for annotation, and RDP algorithm is used for annotation algorithm.

Statistical analysis

QIIME2 software was used to analyze the α -diversity (richness index, Simpson evenness index, Shannon index, etc.) and β -Diversity (based on UniFrac and Bray Curtis equidistance matrix) of different samples was analyzed by R software vegan package. Nonparametric multivariate statistical algorithms such as PERMANOVA.ANOSIM in vegan package of R software were used to analyze the effects of soil basic physical and chemical properties, nutrient factors and REE content on rice. Pearson test, canonical correspondence analysis (CCA) and variation partitioning analysis (VPA) based on CCA were used to analyze the contribution of environmental variables to bacterial community. Origin9.0 is used to draw Heatmap, Software of Metasee and Qiime are used for data visualization^{6,21}.

Statement on guidelines as experimental research and field studies on rice

Experimental research and field studies on rice (cultivated rice and wild rice), including the collection of rice comply with relevant institutional, national, and international guidelines and legislation in the methods section. The measurement process of microorganism and rare earth content in different parts of rice will not affect the local soil microorganism and ecological environment, etc. During the process of experiment, aseptic sampling was carried out to avoid

contamination. The experimental group and control group have been tested at the same time, and the whole experimental process of the research is safe, which is based on references as below.

- 1.The methods of soil collection and material preparation are mainly from reference 2-5.
- 2.The methods of potted rice and sample collection are mainly from reference 3-5.
- 3.The soil physical and chemical properties and nutrient elements were detected by the relevant detection methods in the Agricultural Industry Standard of the People's Republic of China (2006).
- 4.The determination methods of rare earth elements in soil and rice are mainly from reference 2-5.
- 5.The pretreatment and DNA extraction of soil and plant samples are mainly from reference 77-79.
- 6.Amplification and sequencing of 16S r RNA gene is mainly from references 31 and 35.
7. Statistical analysis is mainly from references 6 and 21.

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Author contributions:

X.Z.Z. performed all the experiments with the help of S.L.J., Z.J.H., H.H.P., Y.J.B. and Y.H., X.Z.Z. wrote the manuscript. S.L.J. and Z.J. H. made the data processing and analysis. X.Z.Z.. prepared Figs. 1–6. S.L.J. prepared Figs. 7–9. All the authors reviewed the manuscript and approved the version to be published.

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The authors declare no competing interests.

Additional information

Correspondence and requests for materials should be addressed to S.J.