BR03 - Study on Restoration of Bauxite Residue by Salt-Alkali Tolerant Bacteria

Zhongkai Liu[1], Zeshuang Kang[2], Kun yan[3], Wanchao Liu[4] and Yangyang Wang[5]
1. R&D Process Manager
2. Associate Director
3. Assistant Director
Zhengzhou Non-ferrous Metals Research Institute of CHALCO, Zhengzhou, China
4. Technical Director
Chinalco Environmental Protection and Energy Conservation Group, Beijing, China
5. Associate Professor
Henan University, Kaifeng, China
Corresponding author: lzk1219@qq.com

Abstract

Two salt-alkali tolerant bacteria (ZH-1 and ZH-22) were successfully isolated from bauxite residue disposal site. Both strains were identified by 16S rRNA genes as *Bacillus sp.* The restoration effect of salt-alkali tolerant bacteria on bauxite residue was studied to provide technical support for ecological restoration of bauxite residue disposal site. Bauxite residue was treated with the culture medium of ZH-1 and ZH-22, and the pH value of bauxite residue decreased from 11.57 to about 9 within 45 days and remained basically stable. The addition of ZH-1 and ZH-22 strains increased the organic matter and microbial biomass carbon content in bauxite residue as well as increased the catalase and dehydrogenase activities. The stability of bauxite residue aggregates was significantly improved after the treatment within 30 days, and the improvement was more obvious for the treatment of ZH-22. The analysis of bacterial community showed that the relative abundance of ZH-1 and ZH-22 in bauxite residue increased from 5.06 % and 2.71 % to 23.5 2% and 12.03 %, respectively, during the 30 days incubation. After that, additional carbon and energy source were supplied, and the relative abundance of ZH-1 and ZH-22 further increased, and with ZH-22 more significant. This result indicates that additional supply of carbon source can induce the resurrection of the salt-alkali tolerant bacteria.

Keywords：Bauxite residue, salt-alkali tolerant bacteria, microbial biomass carbon, aggregates.

1. Introduction

The reduction and comprehensive utilization of bauxite residue in alumina industry has become a worldwide problem, so far there is no feasible method for large-scale utilization in the world. A large amount of bauxite residue is disposed by storage, its pollutant migration risk is large, the natural weathering process is slow, and the ecological reconstruction of the yard is difficult. The environmental safety problem of bauxite residue storage is seriously threatening the sustainable development of alumina industry [1]. At the same time, the vegetation reconstruction of bauxite residue disposal site is the most promising method ecological disposal of bauxite residue [2]. However, due to the strong salinization and special physical structure of bauxite residue, it is difficult to plant common plants [3-5]. Therefore, reducing the pH value of bauxite residue and improving the physical structure of bauxite residue are the premise of vegetation reconstruction. At home and abroad, a series of research and practice work have been carried out for bauxite residue soil improvement, such as using foreign soil cover [6], adding modifier [7-9], leaching neutralization [11-13], etc. Although these methods have certain effect, but their efficiencies are not high, there are the problems of secondary pollution or high economic cost.

Microorganisms play an important role in bauxite residue improvement. Soil microorganism is the key driving force of nutrient transformation and circulation of organic matter in soil ecosystem [14]. Microorganism is closely related to many biochemical processes in soil and is sensitive index to characterize soil quality [15]. It can be used as an index of soil ecosystem stability and has environmental remediation function. The metabolic acidogenesis of microorganisms can neutralize the alkalinity of bauxite residue and promote the improvement of lateritic soil, which has great potential in realizing large-scale ecological restoration of bauxite residue [16-21]. Screening suitable strains and providing suitable growth environment to improve microbial activity are the research focus of current microbial methods [22].

Two saline-alkali tolerant bacterial strains (ZH-1 and ZH-22) with high organic acid yield have been screened from bauxite residue yard. Both strains belong to Bacillus (Bacillus sp.). ZH-1 and ZH-22 have strong metabolic acid production ability in alkaline environment, and the organic acids produced (such as citric acid, butyric acid and tartaric acid) are the key factors to reduce the alkalinity of bauxite residue and improve the structure of aggregates. This paper will study the effect of ZH-1 and ZH-22 saline-alkali tolerant bacteria strains on bauxite residue, analyze the effect of their metabolites on bauxite residue pH value, aggregates and their stability, characteristic enzyme activity and microbial carbon, study the changes of community structure during bauxite residue restoration, explore the survival state of saline-alkali tolerant bacteria in bauxite residue, and provide technical support for microbial remediation application in bauxite residue disposal site.

2. Experiment

2.1 Materials and Experimental Methods

The bauxite residue samples were taken from the bauxite residue disposal site of Henan Branch of CHALCO. Table 1 showed the main chemical composition of the bauxite residue. The main chemical composition of the bauxite residue is Al_2O_3, Fe_2O_3, SiO_2, CaO and Na_2O, among which Na_2O is 6.55%.

Table 1. The main chemical composition of the bauxite residue.

Element	Al_2O_3	SiO_2	Fe_2O_3	Na_2O	CaO	MgO	K_2O	TiO_2
Content (%)	25.48	20.58	11.77	6.55	13.97	1.54	2.07	4.14

The collected bauxite residue samples were put into sample bags and stored in 4 °C refrigerator for standby. The liquid medium was prepared according to yeast extract 3.0 g/L, glucose 5.0 g/L, sodium chloride 50.0 g/L. the strain ZH-1 and ZH-22 cultured to logarithmic phase were inoculated into the optimal medium, and then mixed with high-purity water and added into 2 kg bauxite residue, and cultured in biochemical incubator at 30 °C for 4 d, 7 d, 14 d, 21 d, 30 d and 45 d, and then added carbon source at 30 d After the last sampling, part of the bauxite residue was stored in the refrigerator at -20 °C for standby, and the other part was dried in the oven at 50 °C.

2.2 Analysis

2.2.1 Determination of pH Value of Bauxite Residue

The dried samples of raw bauxite residue, 4 d, 7 d, 14 d, 21 d, 30 d and 45 d were taken. Determine the pH of bauxite residue solution according to the ratio of liquid to solid 5:1, weigh 5.0 ± 0.1 g bauxite residue, put it into a 50 ml beaker, add 25 ml high pure water, stir it violently with glass rod for 5 min, then stand for 30–60 min, measure the pH of the supernatant with a calibrated pH meter, and calculate the average value of five parallel tests.

2.2.2 Bauxite Residue Aggregate and its Stability Determination

The original bauxite residue and the naturally dried bauxite residue treated with ZH-1 and zh-22 bacteria for 30 d and 45 d were analyzed for aggregate change. The mechanical stability of aggregates was determined by shavenov dry sieving method [23]. The method proposed by Yoder [24] was used for the determination of water stable aggregates. The content w of each mechanical or water stable aggregate is calculated in formula (1).

$$\mathrm{W}(\%) = \frac{\text{Mass of each mechanical or water stable aggregate(g)}}{\text{Total sample mass(g)}} \times 100 \quad (1)$$

Referring to the method introduced by Chen Shan [25], the stability of bauxite residue aggregate is measured by aggregate failure rate (PAD), and the calculation is shown in formula (2).

$$\mathrm{PAD}(\%) = \frac{W_1 - \mathrm{W}_2}{W_1} \times 100 \quad (2)$$

Where:
W_1: The proportion of > 0.25mm aggregates in mechanically stabilized aggregates.
W_2: The proportion of < 0.25 mm aggregates in water stable aggregates.

2.2.3 Determination of Total Microbial Biomass Carbon

According to the method introduced by Lin Xiangui [26] and Wu Jinshui [27], the content of total microbial biomass carbon in bauxite residue samples was determined.

2.2.4 SEM Analysis

The original bauxite residue, the bauxite residue treated with ZH-1 bacteria for 30 days, and the bauxite residue treated with ZH-22 bacteria for 30 days were scanned by electron microscope to observe the changes of micro morphology of bauxite residue before and after improvement.

2.2.5 Microbial Community Analysis

DNA was extracted from bauxite residue treated with ZH-1 and ZH-22 for 7 d, 14 d, 30 d and 30 d with carbon source by using Fast DNA SPIN Kit for Soil and verified by electrophoresis. The extracted DNA samples were sequenced by high-throughput sequencing to obtain the quantitative changes and survival status of microbial communities in bauxite residue treated by two saline alkali resistant bacteria strains.

3. Results and Discussion

3.1 PH Change During Bauxite Residue Improvement

ZH-1 and ZH-22 bacteria were used to improve the bauxite residue. The bauxite residue samples were dried at the time of 4 d, 7 d, 14 d, 21 d, 30 d and 45 d to determine the pH value of bauxite residue. The results are shown in Figure 1. It can be seen from Figure 1 that the pH of bauxite residue treated with ZH-1 and ZH-22 decreased from 11.57 to 9.28 and 9.26 on the fourth day, to 9.17 and 9.18 on the seventh day, to 8.98 and 9.18 on the 11th day, and then slowly increased, reaching 9.07 on the 14th day and 9.25 and 9.31 on the 28th day respectively. According to the analysis, the pH of bauxite residue decreased significantly after ZH-1 and ZH-22 treatment, and the rising speed of pH was slow, which could be maintained at pH = 9.

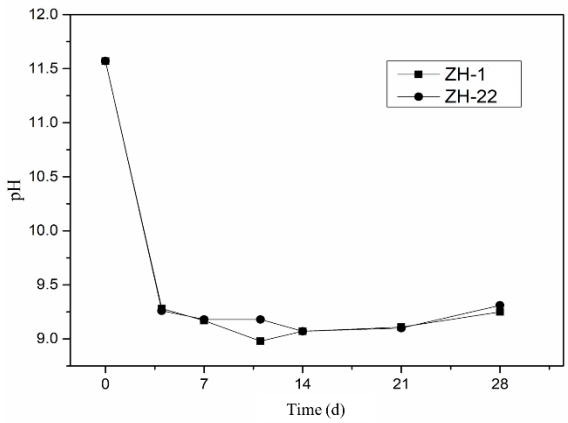

Figure1. Effect of saline-alkali tolerant bacterial strains treatment on pH value of bauxite residue.

3.2 Change of Aggregate in Bauxite Residue Improvement Process

The change process of bauxite residue aggregate in the process of improving bauxite residue by ZH-1 and ZH-22 is shown in Figure 2. Using dry screening method, the original bauxite residue particles are relatively fine, mainly distributed in 0.25–0.5 mm and < 0.25 mm, accounting for 49.44 % and 30.9 % respectively. After the improvement of ZH-1 bacteria, the proportion of aggregates larger than 2 mm was about 50 %, and the proportion of small aggregates decreased significantly. The proportion of 0.25–0.5 mm and < 0.25mm decreased to 9.02 % and 1.45 % respectively. On the other hand, ZH-22 has a more obvious effect on bauxite residue improvement, with the aggregates of > 2mm increasing to 67.01 %, and the proportion of fine aggregates is further reduced. Using wet screening method, it can be seen from Figure 2 that the proportion of large particle aggregates in bauxite residue after microbial improvement increases significantly. The amount of aggregates larger than 2 mm increased from 0.36 % to 14.09 % and 20.72 %, while that of aggregates less than 0.25 mm decreased significantly. There is no obvious regularity in the change of other particle size aggregates.

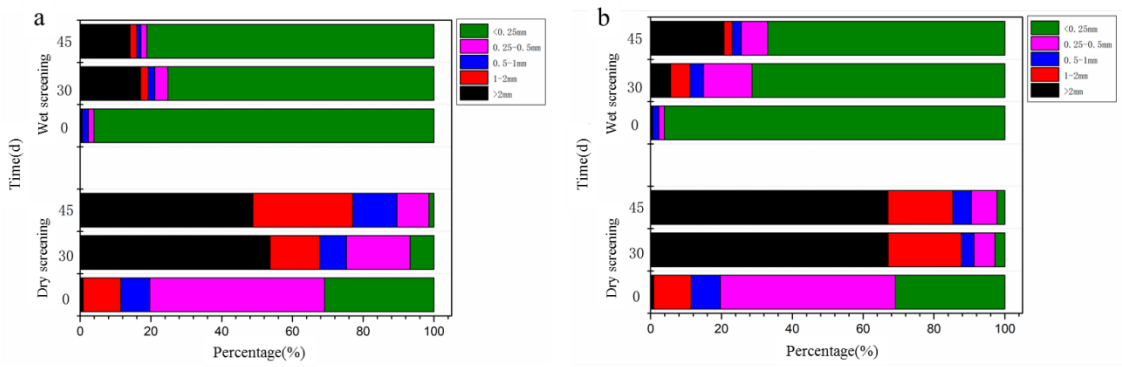

Figure 2. Change of particle size of bauxite residue aggregate treated by saline-alkali tolerant bacterial strains (a: ZH-1; b: ZH-22).

The stability of bauxite residue aggregates treated by the strain was analyzed by the aggregate destruction rate, as shown in Figure 3. After treated with ZH-1 for 30–45d, the aggregate failure rate of bauxite residue decreased from 90 % to 70–80 %, and decreased to 60–70 % after treated with ZH-22 for 30–45 d. After treatment with ZH-1 and ZH-22, the stability of bauxite residue aggregates was significantly enhanced, and the improvement effect of ZH -22 was more obvious.

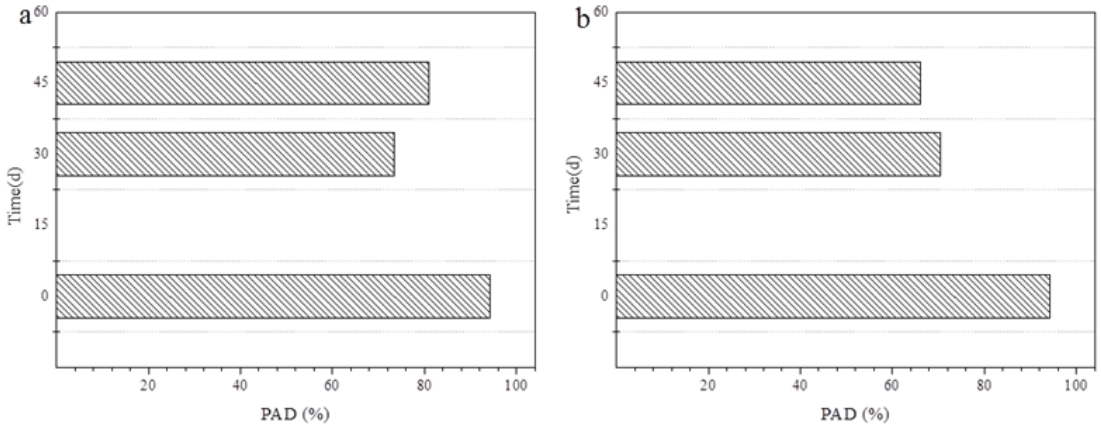

Figure 3. Change of the destruction rate of bauxite residue aggregate treated by saline-alkali tolerant bacterial strains (a: ZH-1; b: ZH-22).

The original bauxite residue, the bauxite residue treated with ZH-1 and the bauxite residue treated with ZH-22 were analyzed by scanning electron microscope, and the results are shown in Figure 4. It can be seen from Figure 4 that the original bauxite residue contains many small flake structure particles with small particle size, irregular shape, unsmooth edge, many edges and corners, and loose structure. After ZH-1 treatment, the bauxite residue surface has larger flake structure, regular shape, flat edge, dense particles accumulation, compact structure, and obvious increase in particle size. After treatment with ZH-22, the structure of bauxite residue has similar changes, and the number of large particles is more than that of the bauxite residue treated with ZH-1 for 30 days. This shows that ZH-1 and ZH-22 are beneficial to the development of bauxite residue physical structure, and the influence of ZH-22 on the physical structure of bauxite residue is more obvious.

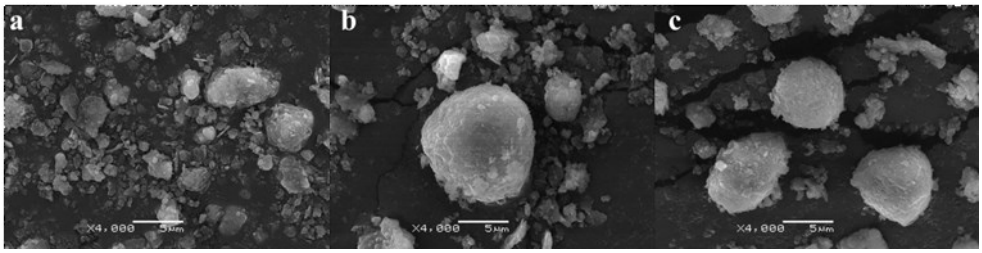

Figure 4. Microstructure changes of bauxite residue before and after improvement of saline-alkali tolerant bacterial strains, (a: unmodified bauxite residue; b: ZH-1 improved bauxite residue; c: ZH-22 improved bauxite residue).

3.3 Changes of Characteristic Enzyme Activity and Microbial Biomass Carbon before and after Bauxite Residue Improvement

3.3.1 Analysis of Organic Matter and Catalase

The effects of ZH-1 and ZH-22 on organic matter content and catalase activity of bauxite residue are shown in Figure 5 and Figure 6. It can be seen from the figure that the organic matter content and catalase activity of bauxite residue increased after ZH-1 and ZH-22 treatment, and the catalase activity increased with the increase of organic matter content.

After treatment with ZH-1 and ZH-22, the content of organic matter in bauxite residue increased the most in 4 days, and the activity of catalase also increased the most; The organic matter content of the bauxite residue treated with ZH-1 reached the maximum value at 7 days, while that of the bauxite residue treated with ZH-22 reached the maximum value at 11 days. After that, the content

of organic matter decreased, remained basically stable and far higher than the original level. It may be that the delay period of microbial flora in the bauxite residue after ZH-22 treatment was longer than that in the bauxite residue after ZH-22 treatment; at the same time, the catalase activity decreased and kept stable and higher than the original level, basically consistent with the change of organic matter content. This shows a good correlation between the two.

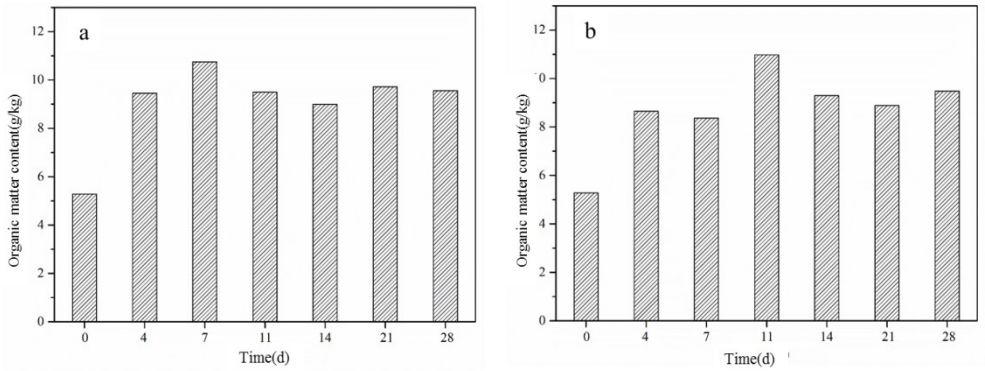

Figure 5. Change of organic matter content in bauxite residue improved by saline-alkali tolerant bacterial strains (a: ZH-1; b: ZH-22).

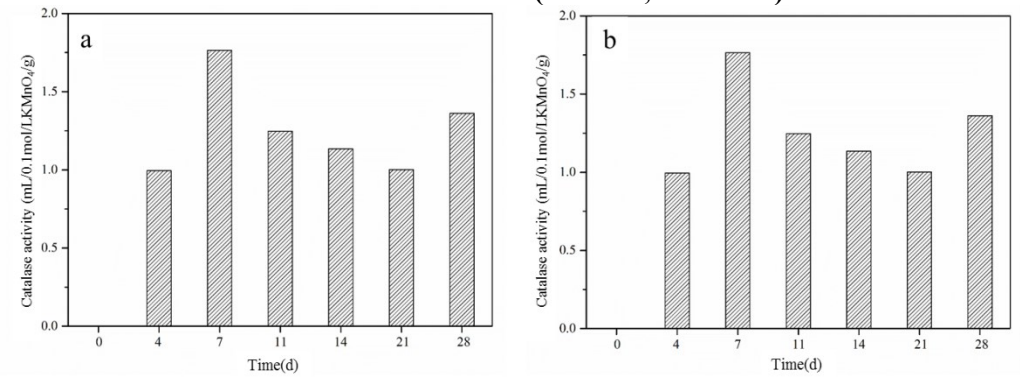

Figure 6. Changes of catalase activity in bauxite residue improved by saline-alkali tolerant bacterial strains (a: ZH-1; b: ZH-22).

3.3.2 Determination of Microbial Biomass Carbon and Dehydrogenase

The effects of ZH-1 and ZH-22 on total microbial biomass carbon content and dehydrogenase activity of bauxite residue are shown in Figure 7 and Figure 8. It can be seen from the figure that after treatment with ZH-1 and ZH-22, the content of microbial biomass carbon (MBC) in the bauxite residue treated with ZH-1 and ZH-22 increased significantly, and kept a steady growth, which was much higher than the initial level; the dehydrogenase activity also had similar changes.

The increase of microbial biomass carbon content in bauxite residue treated with ZH-1 and ZH-22 was the largest at 4 days, and the increase of ZH-1 was greater. It may be that the microbial communities in the bauxite residue treated with ZH-1 and ZH-22 were in logarithmic phase and proliferated in a large number, and the microbial communities in the bauxite residue after ZH-1 treatment increased more rapidly, and the dehydrogenase activity also had similar changes; after that, the microbial biomass carbon in the bauxite residue treated by ZH-22 remained stable The results showed that the microbial community entered the stabilizer and grew stably, and the dehydrogenase activity also had similar changes; at 7 days, the content of microbial biomass carbon in the bauxite residue treated with ZH-1 decreased, which may be due to some reason that restricted the growth of microbial community to some extent, and the activity of dehydrogenase also had similar changes; after treatment with ZH-1, the content of microbial biomass carbon in

bauxite residue decreased after 28 days The results showed that the microbial biomass carbon content of the bauxite residue after ZH-22 treatment continued to increase steadily, which may be that the microbial community in the bauxite residue after ZH-1 treatment reached the decline period, while the microbial community in the bauxite residue after ZH-22 treatment did not reach the decline period because of the long delay period; the dehydrogenase activity in the bauxite residue treated with ZH-1 and ZH-22 increased or decreased within 21 days, and basically remained higher than the initial level Within 21 days, the activity of dehydrogenase in the bauxite residue treated with ZH-1 was close to the initial level, and the activity of dehydrogenase in the bauxite residue treated with ZH-22 was much higher than the initial level. After that, the activity of dehydrogenase in the bauxite residue treated with ZH-1 decreased to far lower than the initial level. The activity of dehydrogenase in the bauxite residue of ZH-22 treatment continued to increase, which was basically consistent with the change of microbial biomass carbon content. This shows the correlation between them.

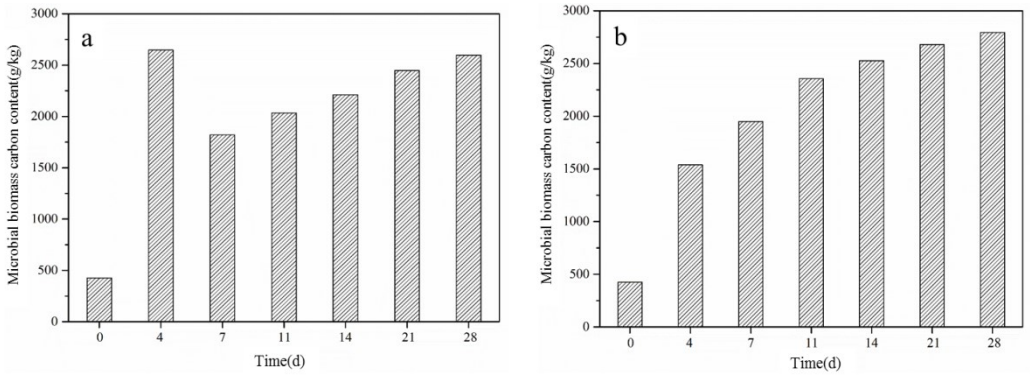

Figure 7. Changes of microbial biomass carbon content in bauxite residue improved by saline-alkali tolerant bacterial strains (a: ZH-1; b: ZH-22).

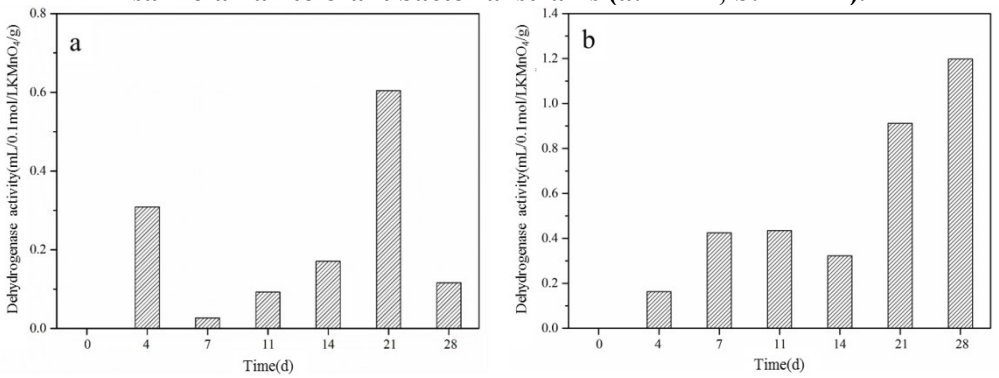

Figure 8. Changes of dehydrogenase activity in bauxite residue modified by saline-alkali tolerant bacterial strains (a: ZH-1; b: ZH-22).

3.4 Analysis of Microbial Community Change

3.4.1 Changes of Dominant Families and Genera in Bauxite Residue

Through high-throughput sequencing, we obtained the change rule and survival state of ZH-1 and ZH-22 in the process of bauxite residue improvement. At the family level, it can be seen from Figure 9(a) that, within 45 days after ZH-1 treatment, with the increase of treatment time, *Bacillaceae* gradually became the dominant family, and the abundance of *Comamonadaceae* and *Peptococcaceae* increased more, and the last three together accounted for more than 70 % and became the main family. However, the initial dominant family, *Enterobacteriaceae*, decreased sharply until it could not be detected. It can be seen from Figure 9(b) that with the increase of

treatment time within 45 days after ZH-22 treatment, the abundance of *Bacillaceae*, *Clostridiaceae-2*, *Peptococcaceae* and *caulobacteraceae* in bauxite residue after ZH-22 treatment has changed, but they still maintain the advantages, accounting for more than 70% in total. The abundance of *Clostridiaceae-2* increased with the decrease of *Bacillaceae*, while the abundance of *Peptococcaceae* and *Caulobacteraceae* changed little.

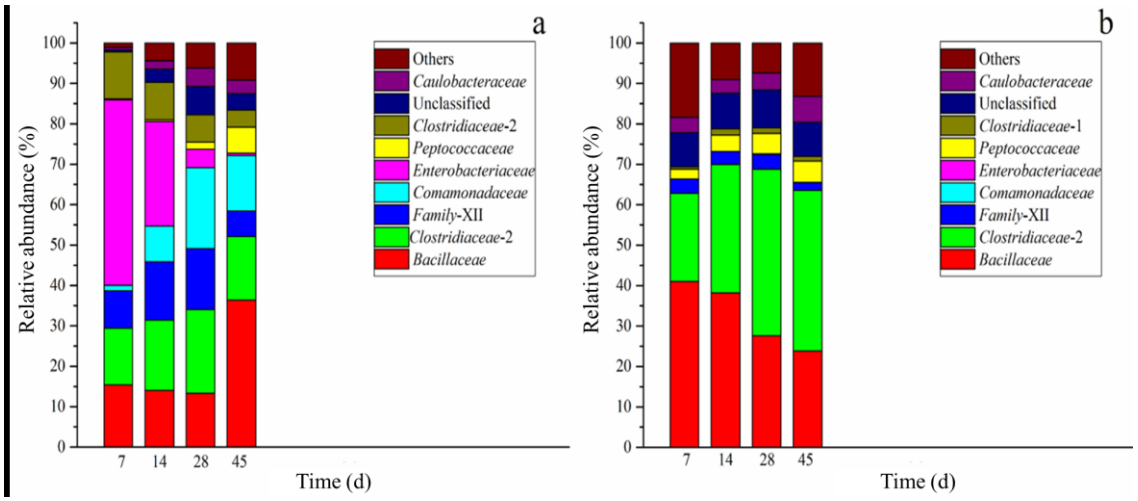

Figure 9. Community composition and relative abundance at the level of microflora in bauxite residue treated by saline-alkali tolerant bacterial strains (a: ZH-1; b: ZH-22).

At the genus level, as shown in Figure 10(a), with the increase of treatment time within 45 days after ZH-1 treatment, *Bacillus* gradually became the dominant genus, and the abundance of *Anaerobacillus*, *Comamonas* and *Brevundimonas* also increased more, the abundance of *Exiguobacterium* did not change much, and the abundance of *Clostridium-sensu-stricto-3* decreased more, but it was still the dominant genus The total proportion is more than 50 %. It can be seen from Figure 10(b) that with the increase of treatment time, *Bacillus* gradually becomes the dominant genus in the bauxite residue after ZH-22 treatment, and the abundance of *Alkaliphilus* increases more, *Exiguobacterium*, *Desulfotomalum* and *Ambiguous_taxa*, *Anaerobacillus* and *Brevundimonas* did not change much, accounting for more than 50 % of the total.

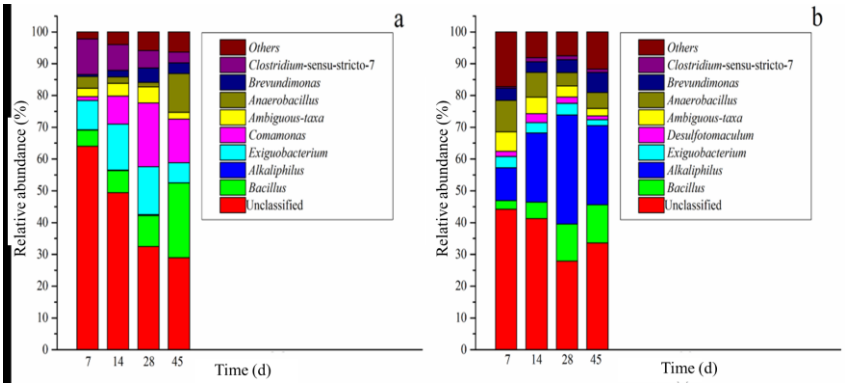

Figure 10. Community composition and relative abundance of microorganisms in bauxite residue treated by saline-alkali tolerant bacterial strains (a: ZH-1; b: ZH-22).

According to the identification results, ZH-1 and ZH-22 belong to *Bacillus sp*. combined with the analysis results at the genus level, ZH-1 and ZH-22 survive well in bauxite residue and increase in quantity. The method of inducing and reviving ZH-1 and ZH-22 in bauxite residue is effective after 30 days. The amount of ZH-1 increased continuously within 4 weeks after adding to bauxite residue, which reached nearly 2 times of the first week at the fourth week, and reached 2.4 times

of that without adding carbon source after the fourth week. The addition of carbon source led to the revival effect obviously; the amount of ZH-22 continued to increase within 4 weeks after adding to bauxite residue, reaching 4.3 times of the first week at the fourth week, and then increased after two weeks Source induced resurrection has certain effect. When added to bauxite residue for one week, ZH-1 accounted for 5.06 %, while ZH-22 only accounted for 2.71 %. Then, the growth rate of ZH-22 was significantly higher than that of ZH-1 and exceeded the proportion of ZH-1 at the fourth week. After adding carbon source, ZH-1 increased significantly, and the proportion of ZH-22 was about twice that of ZH-22. Therefore, the adaptation time of ZH-22 is longer than that of ZH-1. It is speculated that the adaptation time of ZH-1 is 1–2 weeks, while that of ZH-22 is 2–3 weeks, and the growth rate of ZH-22 is faster than that of ZH-1; both of them can be induced to revive by adding carbon source, but the effect of ZH-1 induced resurrection is more obvious.

3.4.2 Species Diversity of Microbial Community in Bauxite Residue

Shannon index is often used to evaluate species richness and diversity of environmental communities. It can be seen from Figure 11 that Shannon index increased gradually in 28 days of ZH-1 treatment, indicating that species richness and diversity of microbial communities in bauxite residue increased. The Shannon index increased slightly at 45 days, and the species richness and diversity of microbial community in bauxite residue had reached a certain balance. The Shannon index did not change much in the 28 days of ZH-22 treatment, which may be due to the longer adaptation period of ZH-22 in bauxite residue, and the species richness and diversity of microbial community in bauxite residue did not change significantly. The Shannon index increased at 45 days, which may be due to the rapid proliferation of ZH-22 in bauxite residue after the adaptation period, which promoted the species richness and diversity of microbial community in bauxite residue. In general, the species richness and diversity of microbial communities in the bauxite residue treated with ZH-22 were higher than those in the bauxite residue treated with ZH-1.

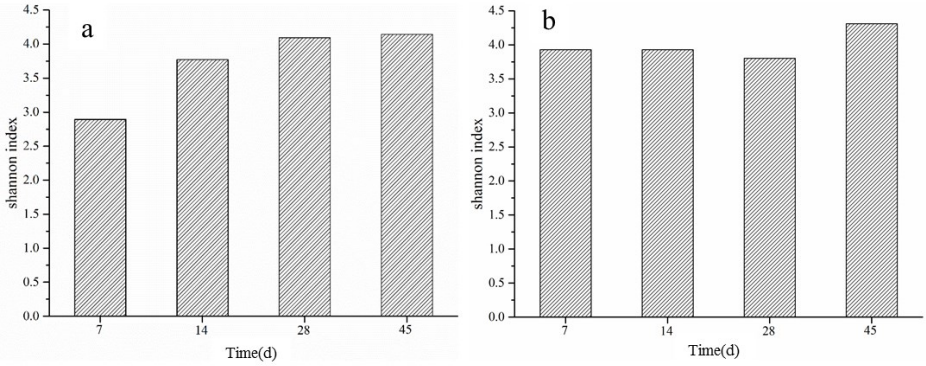

Figure 11. Changes of Shannon index in bauxite residue treated by saline-alkali tolerant bacterial strains (a: ZH-1; b: ZH-22).

3.4.3 RT-PCR Analysis

RT-PCR analysis is shown in Figure 12. It can be seen from Figure 12(a) that the number of ZH-1 gradually increases during the restoration process. The concentration of *Bacillus sp.*. in the original bauxite residue is 3.22×10^5 copies/g soil, and after adding ZH-1, the maximum concentration of *Bacillus sp.* can reach 1.29×10^{10} copies/g soil (45d), but the quantity does not change much from 28 days to 45 days. Therefore, ZH-1 can survive well in the bauxite residue environment, and can reproduce normally, becoming the dominant flora in bauxite residue. After 28 days, additional carbon sources were added to induce the added ZH-1, but the community density did not increase significantly. It may be that the number of ZH-1 in bauxite residue

basically reached saturation, and the possibility of further increase was relatively small. However, the maximum community density of ZH-22 (Figure 12(b)) was only 9.86×10^9 copies/g soil (45d), which was far lower than that of ZH-1 in bauxite residue. Therefore, the adaptability of ZH-22 in bauxite residue was weaker than that of ZH-1. However, ZH-22 could also grow and reproduce normally in bauxite residue, and the community density increased further after 28 days induced by additional carbon source, from 8.74×10^9 copies/g soil (28d) to 9.86×10^9 copies/g The results showed that ZH-22 could maintain a good activity in bauxite residue, and could recover quickly and grow and propagate under suitable environment.

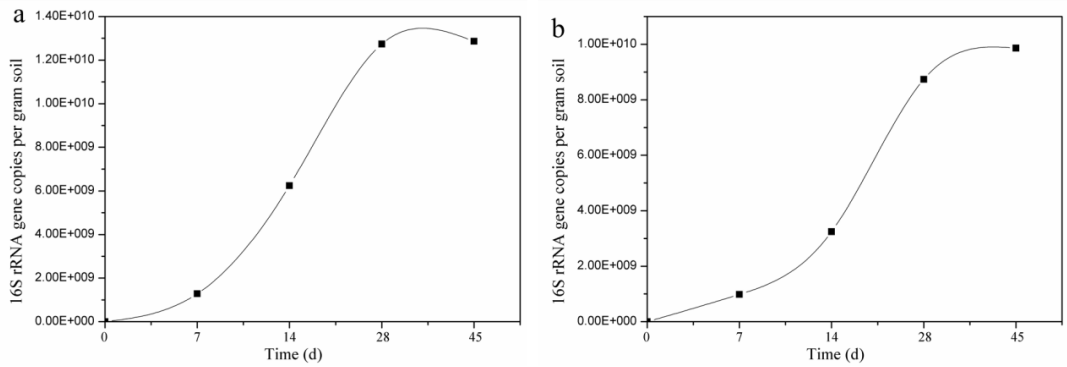

Figure 12. Variation of the number of saline-alkali tolerant bacterial strains in the process of bauxite residue improvement (a: ZH-1; b: ZH-22).

To sum up, the differences of microbial communities in bauxite residue treated by saline alkali resistant bacteria strains ZH-1 and ZH-22 at different times were small, and they would not cause great changes in the microbial communities in bauxite residue. Moreover, ZH-1 and ZH-22 could rapidly proliferate in bauxite residue environment, and could maintain biological activity for a long time, and could be applied to bauxite residue improvement.

4. Conclusions

After treatment with ZH-1 and ZH-22, the pH of bauxite residue decreased significantly, and the pH of bauxite residue decreased to about 9 within 45 days, and remained basically stable; after treatment of ZH-1 and ZH-22, the large aggregates of bauxite residue increased, the stability of aggregates was significantly enhanced, and the physical structure was improved to some extent, and the improvement effect of ZH-22 was more obvious.

After treatment with ZH-1 and ZH-22, the organic matter and catalase activity in bauxite residue increased greatly, and their changes were basically the same; microbial biomass carbon and dehydrogenase activity also increased greatly, and the changes of them were basically the same. After treatment with ZH-1 and ZH-22, the contents of organic matter and microbial biomass carbon and the activity of oxidoreductase in bauxite residue increased.

The relative abundance of ZH-1 and ZH-22 in bauxite residue increased from 5.06 % and 2.71 % to 23.52 % and 12.03 % respectively during the 30-day experimental confirmation period. The carbon source was added to induce the reactivation at 30 days, and the relative abundance increased based on 30 days after half a month.

5. References

1. Shengguo Xue, Yubing Li, and Ying Guo, Environmental impact of bauxite residue: a comprehensive review, *Journal of University of Chinese Academy of Sciences,* Vol. 34, No. 4(2017), 401-412.
2. M. Gräfe, and C, Klauber, Bauxite residue issues: IV. Old obstacles and new pathways for in situ, residue bioremediation, *Hydrometallurgy*, Vol. 108, No. 1/2(2011), 46-49.
3. G. Power, M. Grafe, and C. Klauber, Bauxite residue issues: I. current management，disposal and storage practices, *Hydrometallurgy,* Vol. 108, No. 1/2(2011), 33-45.
4. Feng Zhu, et al, Natural plant colonizationimproves the physical condition of bauxite residue over time, *Environmental Science and Pollution Research*, Vol. 23, No. 22(2016), 1-9.
5. B.E.H. Jones, R.J. Haynes, Bauxite processing residue: acritical review of its formation，properties, storage, and revegetation, *Critical Reviews in Environmental Science and Technology,* Vol. 41, No. 3(2011), 271-315.
6. J.B. Wehr, I. Fulton, N.W. Menzies, Revegetation strategies for bauxite refinery residue: a case study of Alcan Gove in Northern Territory, Australia, *Environmental Management,* Vol. 37, No. 3(2006), 297-306.
7. R. Courtney, T. Harrington, K.A. Byrne, Indicators of soil formation in restored bauxite residues, *Ecological Engineering,* Vol. 58, No. 13(2013), 63-68.
8. J. Eastham, T. Morald, P. Aylmore, Effective Nutrient Sources for Plant Growth on Bauxite Residue, *Water, Air, and Soil Pollution,* Vol. 176, No. 1/4(2006), 5-19.
9. Y. Ma, C. Si, C. Lin, Capping hazardous red mud using acidicsoil with an embedded layer of zeolite for plant growth, *Environmental Technology,* Vol. 35, No. 18(2006), 2314-2321.
10. N. W. Menzies, I.M. Fulton, and W.J. Morrell, Seawater Neutralization of Alkaline Bauxite Residue and Implications for Revegetation, *Journal of Environmental Quality*, Vol. 33, No. 5(2004), 1877.
11. S.J. Couperthwaite, et al, Minimization of bauxite residue neutralization products usingnanofiltered seawater, I*ndustrial & Engineering Chemistry Research,* Vol. 53, No. 10(2014), 3787-3794.
12. Wentao Liang, et al, Effect of strong acids on red mud structural and fluoride adsorption properties, *Journal of Colloid and Interface Science,* Vol. 423, No. 3(2014), 158-165.
13. M. Johnston, et al, Alkalinity conversion of bauxite refinery residues by neutralization, *Journal of Hazardous Materials,* Vol. 182, No. 1/3(2010), 710-715.
14. Ji Zhou, Ting Lei, Status and Prospect of influencing factors and research methods of soil microbial diversity, *Biodiversity*, Vol. 15, No. 3(2007), 306-311.
15. Bo Sun, et al, Soil quality and sustainable environment -- Ⅲ. Biological indicators for soil quality evaluation, *Soils,* Vol. 5(1997), 225-234.
16. R. Courtney, J.A. Harris, M. Pawlett, Microbial Community Composition in a Rehabilitated Bauxite Residue Disposal Area: A Case Study for Improving Microbial Community Composition, *Restoration Ecology,* Vol. 22, No. 6(2015), 798-805.
17. Pankaj Krishna, A.G. Babu, M.S. Reddy, Bacterial diversity of extremely alkaline bauxite residue site of alumina industrial plant using culturable bacteria and residue 16S rRNA gene clones, *Extremophiles,* Vol. 18, No. 4(2014), 665-676.
18. Achim Schmalenberger, et al, Bacterial communities established in bauxite residues with different restoration histories. *Environmental ence & Technology*, Vol. 47, No. 13(2014), 7110-7119.
19. T.C. Santini, et al, In situ neutralisation of uncarbonated bauxite residue mud by cross layer leaching with carbonated bauxite residue mud, *Journal of Hazardous Materials,* Vol. 194, No. 5 (2011), 119-127.
20. M.K. Hamdy, F.S. Williams, Bacterial amelioration of bauxite residue waste of industrial alumina plants, *Journal of Industrial Microbiology & Biotechnology,* Vol. 27, No. 4 (2001),

228-233.
21. Pankaj Krishna, A. Arora, M.S. Reddy, An alkaliphilic and xylanolytic strain of actinomycetes Kocuria sp.RM1 isolated from extremely alkaline bauxite residue sites, *World Journal of Microbiology and Biotechnology,* Vol. 24, No. 12 (2008), 3079-3085.
22. Natasha C. Banning, et al, Development of Microbial Diversity and Functional Potential in Bauxite Residue Sand under Rehabilitation, *Restoration Ecology,* Vol. 19, No. 101 (2011), 78-87.
23. W.C. Dai, et al, Effect of biomass ash application of soil aggregates and organic carbon distributions of citrus orchard soils, *Journal of Soil and Water Conservation,* 2016, 30(2): 260-265. Vol. 30, No. 2 (2016), 260-265.
24. Robert E Yoder, A direct method of aggregate analysis of soils and a study of the physical nature of erosion losses, *Joual of the American Society of Agronomy,* Vol. 28, No. 5 (1936), 337-351.
25. Shan, Chen, et al, Impact of land use patterns on stability of soil aggregates in red soil region of South Chinal, *Journal of Soil and Water Conservation,* Vol. 26, No. 5 (2012), 211-216.
26. Xiangui Lin, *Principles and Methods of Soil Microbiology Research.* Beijing: Higher Education Press, 2010.
27. Jinshui Wu, *Method for Determining Soil Microbial Biomass and Its Application.* Beiing: China Meteorological Press, 2006.