Hindawi
Advances in Civil Engineering
Volume 2021, Article ID 6683930, 21 pages
https://doi.org/10.1155/2021/6683930

*Review Article*

# Geotechnical Engineering Properties of Soils Solidified by Microbially Induced $CaCO_3$ Precipitation (MICP)

**Jia Liu**,[1] **Gang Li**,[2] **and Xi'an Li**[1]

[1]*School of Geological Engineering and Geomatics, Chang'an University, Xi'an, Shaanxi 710054, China*
[2]*Shaanxi Key Laboratory of Safety and Durability of Concrete Structures, Xijing University, Xi'an, Shaanxi 710123, China*

Correspondence should be addressed to Gang Li; t_bag945@126.com

Received 23 December 2020; Revised 27 April 2021; Accepted 10 June 2021; Published 22 June 2021

Academic Editor: Ning-Jun Jiang



Microbially induced calcium carbonate precipitation (MICP) uses the metabolic function of microbes to carry out biochemical reactions with other substances in the environment. Through the controlled growth of inorganic minerals, soil particles are cemented and soil pores are filled to solidify the soil and reduce its permeability. Thus, the application of this technology was foreseen in geotechnical engineering and environment (building antiseepage, contaminated soil restoration, slope soil erosion, and sand liquefaction). In this review article, based on current research findings, the urea hydrolysis and the cementation mechanism of MICP are briefly described. The influences of factors such as enzyme activity, cementation solution concentration, pH, temperature, grouting method, and particle size on MICP-treated soil are discussed. The engineering properties of MICP-treated soils are evaluated, for instance, the strength, stiffness, liquefaction resistance, permeability, and durability. The applications of MICP technology in ground improvement, geotechnical seepage control, foundation erosion resistance, and fixation of heavy metals are summarized. Finally, future directions of the development of MICP technology are elucidated to provide a reference and guidance for the promotion of MICP technology in the geotechnical engineering field.

## 1. Introduction

With the rapid socioeconomic development and accelerated urbanization, urban infrastructure construction is facing unprecedented development worldwide. In this process, a series of engineering challenges exist, including improvement of weak ground, treatment of liquefied soil, remediation of contaminated soil, seepage and leakage control on dams, and dust and sand fixation. Conventional methods used to address these challenges have the disadvantages of high construction difficulty, long construction period, high energy consumption, single performance, and secondary pollution [1]. Microbially induced calcium carbonate precipitation (MICP) has emerged in recent years; it uses calcium carbonate ($CaCO_3$) induced by bacteria to cement loose soil particles, thereby improving the mechanical properties of soil. Compared with conventional materials, MICP technology exhibits greater potential for application and environmental sustainability in geotechnical engineering area and is suggested as a feasible alternative by many researchers [2–4].

Since the 1960s, researchers have gradually realized that microbial metabolic activities are directly involved in redox processes in the environment, thereby altering the geological characteristics and affecting the material cycle and migration of the biosphere. In nature, there is a large number of microorganisms that continuously multiply, grow, and migrate in the soil; participate in mineralization reactions; produce secretions; and degrade organic matter [5]. Microbial mineralization is the process by which microorganisms use their metabolic reactions to produce the urease enzyme. This enzyme catalyzes the decomposition of urea in the surrounding environment to generate carbonate ($CO_3^{2-}$), which reacts with calcium ion ($Ca^{2+}$) to produce $CaCO_3$ precipitates. MICP technology was initially applied in the field of seepage and leakage control of porous media materials and was later extended to the repair of cracks in ancient buildings and rock materials. Studies on soil improvement started relatively late [6]. Whiffin [7] was the first to propose the use of MICP technology for cement loose sand particles to improve soil mechanical properties such as the sand strength and stiffness of sand. Mitchell and Santamarina [8] identified the extensive application value and potential of microbially modified rocks and soils. The National Research Council of the United States established microbial geotechnical engineering technology as an important research topic of the 21st century [9]. Currently, MICP technology is mainly used in research on sand and is slowly being extended to other types of soil, such as silty soil, expansive soil, clayey purple soil, and red clay. Existing studies have shown that MICP technology can significantly improve the shear strength, liquefaction resistance, and erosion resistance of soils and substantially reduce the permeability of soils [10].

As a new research branch in geotechnical engineering, microbial environmental geotechnical engineering is an interdisciplinary subject of microbiology, chemistry, and geotechnical engineering that has been developed for over a decade. The biochemical reaction process of MICP is detailed in this review article, and the factors influencing MICP-treated soil are discussed. The relevant findings regarding MICP technology in applications such as ground improvement, rock-soil seepage control, ground erosion resistance, and contaminated soil remediation are summarized. The findings described in this review can provide a reference for the development and application of MICP technology in the geotechnical engineering field.

## 2. Principles of MICP Technology

*2.1. Metabolic Process of MICP.* MICP is a common phenomenon of microbial mineralization in nature [4, 6]. It involves a series of biochemical reactions with other substances in the environment through metabolic functions and absorbing, transforming, removing, and degrading these substances, and it induces the formation of mineral precipitates such as carbonate and sulfate through biological processes, thereby improving the mechanical properties of the soil. The production of $CaCO_3$ precipitates mainly depends on the presence of ${CO_3}^{2-}$ and $Ca^{2+}$ in an alkaline solution environment, which combine to reach a saturation state so that $CaCO_3$ precipitates. Different metabolic types of microorganisms can form different ways of bioinduced mineralization, such as urea hydrolysis, denitrification, ferric reduction, and sulfate reduction [5, 11]. Among them, urea hydrolysis to precipitate carbonate is the most simple, directly and easily controlled mechanism of MICP, which is widely used [12].

The urea hydrolysis is that urease enzyme produced by bacteria catalyzes hydrolysis of urea to ${NH_4}^+$ and ${CO_3}^{2-}$ [13]. The production of ${NH_4}^+$ by ureolysis ultimately results in an increase in pH. The ${CO_3}^{2-}$ content in solution increases with increasing pH. In urease catalysis, urease breaks the covalent bonds of urea, and urease-urea reaction intermediates are formed between the active center of urease and the substrate molecules of urea through short-range noncovalent forces, for instance, hydrogen bonds, ionic bonds, and hydrophobic bonds. The main reactions of ${CO_3}^{2-}$ crystallization induced by urease-producing bacteria are expressed as follows [14, 15]:

$$\begin{aligned}
CO(NH_2)_2 + 2H_2O &\longrightarrow H_2CO_3 + 2NH_3,\\
H_2CO_3 + 2NH_3 &\leftrightarrow 2NH_4^+ + 2OH^-,\\
H_2CO_3 &\longrightarrow H^+ + HCO_3^-,\\
HCO_3^- + H^+ + 2OH^- &\leftrightarrow CO_3^{2-} + 2H_2O,\\
Ca^{2+} + CO_3^{2-} &\leftrightarrow CaCO_3\downarrow.
\end{aligned} \tag{1}$$

Notably, the equilibrium state of ${NH_4}^+$ or $NH_3$ in the solution is very important. The concentration of ${NH_4}^+$ or $NH_3$ is governed by Henry's law for gases dissolution, which makes it special in MICP because the bacteria represent a source of ${NH_4}^+$, and could be manipulated by changing the temperature or pH, by introducing a sink for $NH_3$ or by releasing it out of the liquid by aeration. $NH_3$ that results from urea hydrolysis inside the cell diffuses out of the cell membrane due to the concentration gradient, and the pH of the cytoplasm equilibrates at 8.4 (${NH_4}^+ : NH_3 = 70 : 30$). ${NH_4}^+$ diffuses out of the cell, increasing the membrane potential, which occurs since ${NH_4}^+$ and $NH_3$ equilibrate when the surrounding solution reaches a pH of 9.25 [7, 16]. Lauchnor et al. [17] investigated the ureolysis rate with whole cells to determine the relationship between the ureolysis rate and the concentrations of ${NH_4}^+$. Lee et al. [18] investigated the ${NH_4}^+$ concentration in the MICP process and used meter-scale experiments for the removal of ${NH_4}^+$. Qin and Cabral [19] thought that $NH_3$ exists mainly in the form of ${NH_4}^+$ ions within the temperature (25°C) and pH values (7) by studying the kinetics of urea hydrolysis catalyzed by urease. Krajewska [20] also investigated $NH_3$ and considered that the role of urease in soil is to make urea available to plants by converting it to $NH_3$; if the hydrolysis rate is too rapid, it may cause $NH_3$ volatilization and then impact the environment [21]. Therefore, in the MICP process, the hydrolysis rate and ammonia and ammonium concentration should be of concern to investigators. In application, the method of inhibiting urease activity can be employed to avoid ammonia emission.

Based on the study of urea hydrolysis in solution and solid agar, realizing the metabolic process of MICP has some limitations at the microscale. Microfluidic chip technology can overcome this limitation and can be used for real-time observation of reactions, less reactant consumption, and excellent controllability of environmental factors. Wang et al. [22–24] utilized glass slides and microfluidic chips to carry out real-time in situ microscale experiments (Figure 1(a)) and realized the visualization of $CaCO_3$ precipitation process. By observing the behavior of bacteria and $CaCO_3$ crystals in this process, understanding MICP at the particle scale was improved. Based on microfluidic chip technology, He et al. [25] developed a visualization system for microbial reinforcement (Figure 1(b)) and carried out in situ microscopic research on the mechanism of $CaCO_3$ mineralization induced by microorganisms. The deposition of $CaCO_3$ crystals in the process of microbial reinforcement

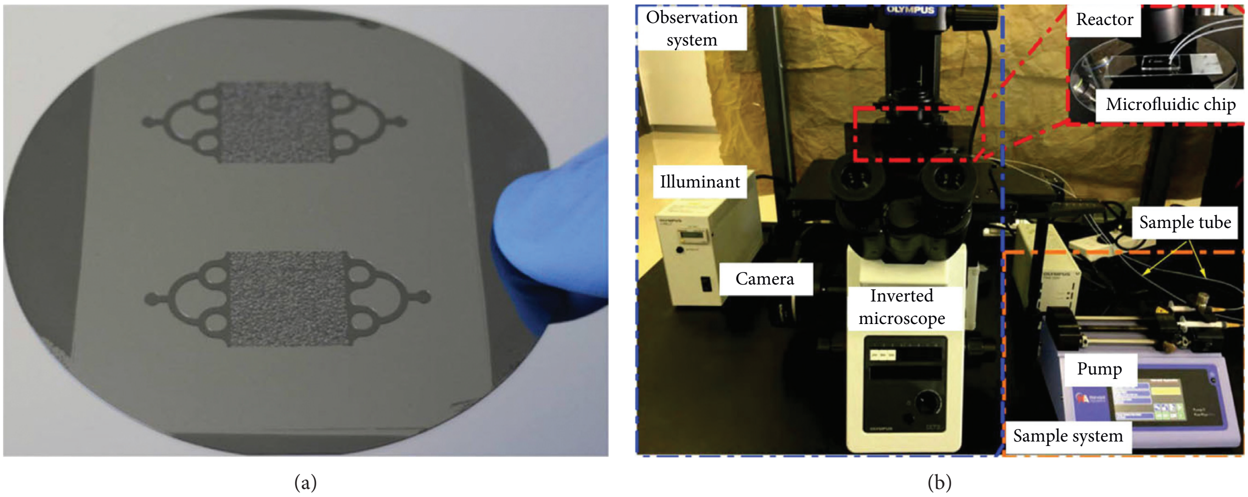


Figure 1: Microfluidic chip technology and its visualization: (a) microfluidic chip [23]; (b) schematic images of the microfluidics system [25].

was observed, and its temporal and spatial distribution, deposition mode, and growth rate were quantified.

*2.2. Cementing Mechanism of MICP.* The microbially induced formation of calcite may exhibit two extreme distribution states in the pores of soil particles. One is the formation of calcite of equal thickness around the soil particles. In this case, the cementation among soil particles is relatively weak, and there are no significant improvements in the soil properties. The other is the formation of calcite only at positions where soil particles are in contact with each other. This type of distribution allows calcites to be employed entirely for cementation among soil particles, which is beneficial to the improvement in soil properties [11]. Cui et al. [26] considered that the effective cementation of calcite crystals can be divided into two modes: (a) calcite-calcite cementation (caused by calcite crystal clusters between adjacent sand particles, Figure 2(a)); (b) particle-contact cementation (caused by calcite crystals precipitated at the particle-particle contacts, Figure 2(b)). Wang et al. [13, 23, 24, 27] used microfluidic chip technology to study the size, quantity, distribution, and morphology of $CaCO_3$ in the MICP process. The results showed that the formation of $CaCO_3$ crystals was distributed in both narrow and open pore throats. The higher the bacterial concentration was, the more the crystals formed in the same volume were. The shape and size of $CaCO_3$ precipitates changed during the MICP process. Irregular-shaped $CaCO_3$ precipitates formed during the initial stage but dissolved when new $CaCO_3$ crystals formed. In addition, with the dissolution of irregular-shaped $CaCO_3$, spherical and rhombohedral $CaCO_3$ crystals formed. He et al. [25] carried out an in situ micromesoscopic study on the mechanism of microbially induced $CaCO_3$ mineralization using a visualization system for microbial cementation. They concluded that the convection and diffusion of solute molecules significantly influence the distribution of $CaCO_3$ crystals and that nonuniform spatial and temporal distributions of $CaCO_3$ are present. The nonuniform temporal distribution was reduced as the reaction proceeded, while the nonuniform spatial distribution was maintained throughout the entire reaction process (0 to 2200 min). There were two patterns of $CaCO_3$ precipitation, i.e., at the pores and at the sand contacts, in microsized pipes, with the $CaCO_3$ at the pores growing uniformly and the $CaCO_3$ at the sand contacts exhibiting axes with different growth rates.

## 3. Analysis of the Influence Factors of Soil Solidification by MICP

Researchers have conducted in-depth and systematic studies on MICP-treated soils. The results showed that the main factors that affect the solidification results of MICP include enzyme activity, cementation solution concentration, pH, temperature, grouting method, and particle size [28].

*3.1. Enzyme Activity.* Urea hydrolysis mainly occurs through urease produced by microorganisms. Urease activity has a great influence on the formation of $CaCO_3$ precipitation. Miftah et al. [29] conducted tube experiments under different enzyme concentrations of 1.25, 2.5, 5, 10, 11, and 15 mL/L to determine the appropriate concentration of enzyme that yields a higher mass of precipitation. The results showed that the mass of precipitation and the precipitation ratio at the enzyme concentration of 10 mL/L, which is equivalent to 4650 U/L, were larger than those at other concentrations, so this concentration was selected for enzyme-induced carbonate precipitation (EICP) solution. Almajed et al. [30] obtained sand specimens with a high strength (1.8 MPa) treated with EICP by adding nonfat powdered milk to the treatment solution. The unconfined compressive strength increased as the concentration of enzyme (from 0.85 g/L to 3 g/L) increased. Martin et al. [31] successfully used a similar method to enhance the UCS of Ottawa 20/30 silica sand, F85 silica sand, Soda-lime glass beads, and local washed quarry sand with 12,600 U/l urease

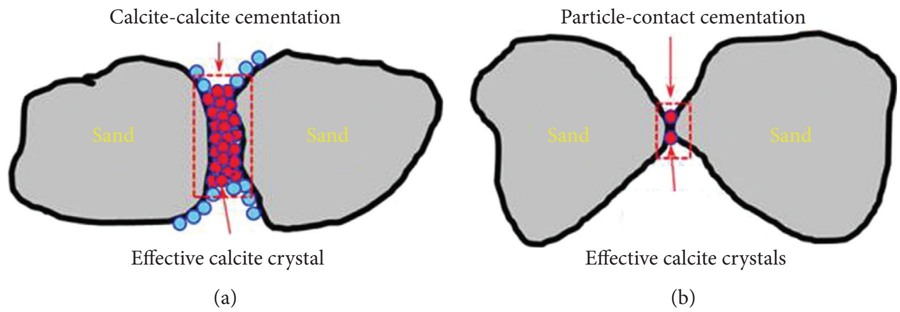


Figure 2: Effective calcite crystals in biocemented sand: (a) calcite-calcite cementation; (b) particle-contact cementation [26].

enzyme. Their experimental results indicated that the strength of Ottawa 20/30 sand was the largest, reaching 1.5 MPa. Konstantinou et al. [32] carried out a cementation test of very coarse sands (mean particle size is 1820 $\mu$m) by controlling the urease activity in bacterial populations used for biological cementation. The results showed that MICP on very coarse-grained materials was successful when urease activities less than 10 mmol/L/h were used with bacterial populations with optical densities equal to or greater than 2. Baziar et al. [33] utilized soybean enzyme to induce carbonate precipitation to stabilize loose soil-inducing fugitive dust and conducted wind tunnel tests to examine their erosion performances. The results indicated that soybean enzyme was easy to prepare and could help the control of dust emission caused by wind erosion. Chen et al. [34] investigated the effects of soybean urease-induced carbonate precipitation on the water retention ability of the ISO standard sand; the sands treated with urease (urease activity was obtained as 5.4 mM/min) and 1 mol/L urea-calcium chloride solutions had greater water retention ability. Instead of bacteria, Yang et al. [35] applied urease directly to fine sand with a premixing method. The results indicated that saturated specimens (urease activity was 5.77 mM urea/min) obtained better moisture stability than unsaturated specimens (urease activity was 4.55 mM urea/min). Some scholars [36, 37] have also used urease extracted from soybean to induce $CaCO_3$ deposition to solidify sand. In addition, urease can induce the formation of $CaCO_3$ crystals, which is effective in sand fixing. However, the application cost of using purified urease is expensive [36]. Some scholars extracted urease from plants for research. Because the cultivation conditions favorable for bacterial growth could not guarantee stable urease production [7, 32], most scholars discussed the different MICP processes with a focus on controlling the bacterial concentration [38–43].

*3.2. Cementation Solution Concentration.* Currently, the cementation solutions used in MICP tests are mostly a mixture of urea and $Ca^{2+}$ solutions. Different concentrations and compositions of cementation solution significantly affect the $CaCO_3$ yield, mineralization efficiency, crystal structure, spatial distribution, and mechanical properties. Rebata-Landa [44] treated samples under the condition of circulating nutrient solution for 64 days using a 60 mL plastic syringe, showing that the $CaCO_3$ content in samples gradually increased with increasing cementation solution concentration and reaction time. Nemati et al. [45] showed that the mineralization efficiency of $CaCO_3$ in solution exceeded 80% when the $CaCl_2$ concentration was less than 15 g/L, while the $CaCO_3$ production was essentially zero when the concentration exceeded 20 g/L; that the mineralization efficiency of $CaCO_3$ could reach 99% when the concentration ratio of $CaCl_2$ and urea was 2.5 or 3.0; and that too high or too low concentration ratio decreased the mineralization efficiency. Qabany et al. [46] carried out tests on the MICP cementation of quartz sand using *S. pasteurii* and concluded that within the concentration range of 0.25 to 1.00 mol/L, the higher the concentration of cementation solution was, the larger the size and the more nonuniform the distribution of the generated calcite crystals (Figure 3). Qabany and Soga [47] treated sand samples using 0.1, 0.25, 0.5, and 1.0 mol/L solutions of urea and $CaCl_2$ mixtures and found that the use of high concentrations rapidly reduced the permeability at an early stage. In addition, some scholars explored the $CaCO_3$ deposition amount, sample strength, permeability, and optimal conditions for $CaCO_3$ deposition for different concentrations of cementing solution (Table 1). The deposition of $CaCO_3$ increases as the concentration of cementation solution increases, but there are differences in the optimal cementation schemes, such as [41, 48, 50] (Figure 4). According to the research results of [49, 51], the maximum deposition of $CaCO_3$ occurs in cement solutions with high $Ca^{2+}$ concentration. Therefore, the cementation solution significantly influences the bacterial activity, $CaCO_3$ precipitation, and soil cementation in MICP (Table 1). In applications, appropriate cementation solutions and concentration ratios should be selected according to the specific reinforcement requirements to meet the needs of actual engineering projects.

*3.3. pH.* Most urease-producing bacteria commonly used in MICP are heterotrophic facultative aerobic bacteria, which are suitable for growth in slightly alkaline environments. Therefore, the pH value has an important influence on growth [28]. Henze and Randall [52] revealed that *S. pasteurii* survived in a solution of pH 11.2, which allows MICP to create durable building materials. Stocks-Fischer et al. [14] examined the effect of pH on the activity of urease extracted from *B. pasteurii* and found that the urease activity gradually increased with increasing pH and peaked at pH of

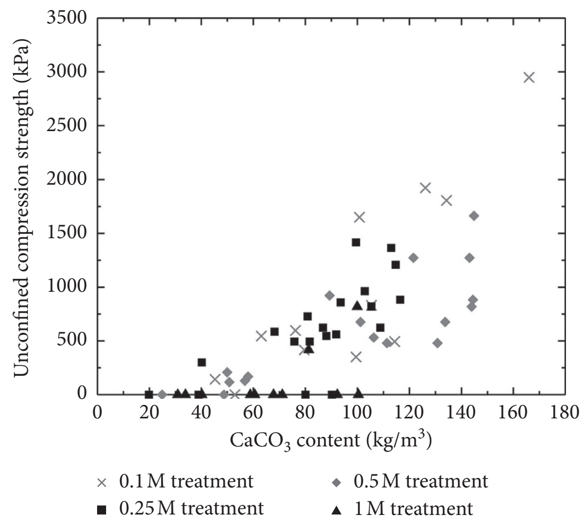


Figure 3: Unconfined compression strength plotted against $CaCO_3$ for different treatments [46].

Table 1: Previous studies on different cementation solution concentrations.

| Reference | Bacteria | Cementation solution concentration | Conclusion |
|---|---|---|---|
| [41] | *B. megaterium* | 0.25, 0.5, 1.0 mol/L urea-calcium chloride | The shear strength of the specimens treated with 0.25 mol/L cementation reagent improved by 26–57%, and the hydraulic conductivity reduction ranged from 16 to 73% |
| [48] | *S. pasteurii* | 0.25, 0.5, 1.0 mol/L urea; 0.25, 0.5, 1.0, 2.0 mol/L calcium chloride | The UCS of sand column treated with 1 mol/L urea and $CaCl_2$ solution is the largest |
| [49] | *S. pasteurii* | 2.5, 25, 250 mM $Ca^{2+}$; 333, 666 mM urea | Increasing urea and $Ca^{2+}$ concentrations increase the amount of carbonate precipitated. The $CaCO_3$ precipitated depend more on the $Ca^{2+}$ concentration than the amount of urea |
| [50] | *S. pasteurii* | 0.5, 1.0, 1.5 mol/L urea and calcium chloride, concentration ratio: 1 : 1, 1 : 2, and 2 : 1 | The $CaCO_3$ content and peak strength of the samples increase with the increasing of the cementation solution concentration, with the maximum being at concentration ratio 2 : 1 |
| [51] | *S. pasteurii* | 0.25, 0.75, 1, 2 mol/L urea and calcium chloride | An increase in urea and calcium concentrations increased the $CaCO_3$ precipitation. The greatest amount of $CaCO_3$ was achieved at the condition of 1 mol/L urea and 2 mol/L calcium chloride |

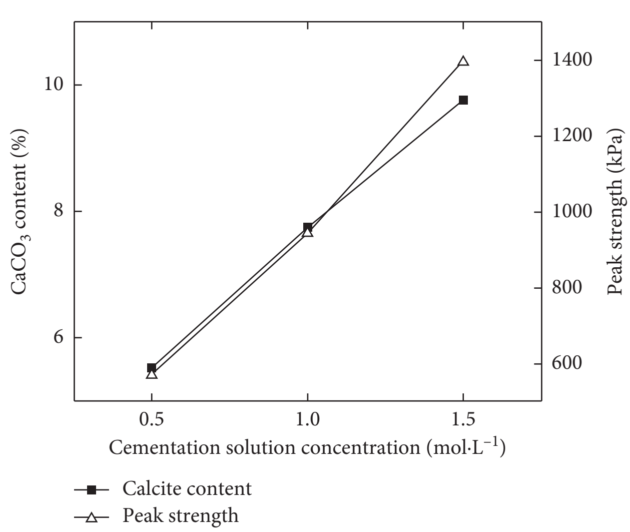


Figure 4: Relationship between $CaCO_3$ content and peak strength with different cementation solution concentrations [50].

7.5–8.0. Whiffin [7] inoculated bacteria grown under different pH conditions into a standard test solution with a pH of 7 and urea concentration of 25 mM/L and then measured the urea decomposition rate after incubation at 25°C for 5 h. The unit urea decomposition rate of the bacteria was the highest between pH of 7 and 8, which is slightly lower than the optimum growth pH of *S. pasteurii*. pH values have an important role not only in the growth of bacteria but also in metabolism, $CaCO_3$ deposition, and soil property improvement. Different pH values will change the concentrations of $NH_3$, $NH_4^+$, $CO_3^{2-}$, and $HCO_3^-$ in the solution, thus changing the formation rate and yield of $CaCO_3$ [53]. Li et al. [54] investigated the precipitation kinetics and crystal morphology of MICP at initial pH values of 6.0, 6.5, 7.0, and 8.0 and concluded that high pH favored $CaCO_3$ precipitation (Figure 5). The $CaCO_3$ precipitates were mainly calcite crystals, which gradually changed from prism to pyramid-like or irregular polyhedral shapes with increasing deposition time. Cheng et al. [55] carried out biocemented sand

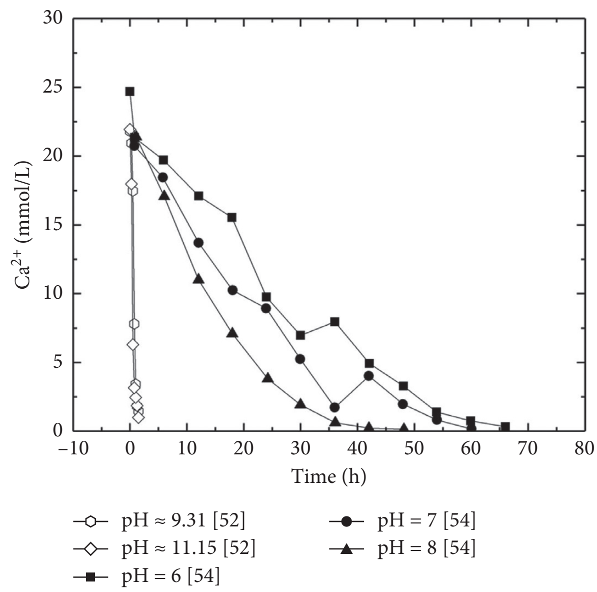


Figure 5: Changes in $Ca^{2+}$ concentration at different pH values.

column tests using *B. sphaericus* under initial neutral, acidic, and alkaline pH conditions. They found that the $CaCO_3$ content in the samples increased with increasing pH, whereas the cemented sand column had the highest strength under neutral conditions. Keykha et al. [56] employed *S. pasteurii* for the biocementation of silty clay soil at pH values of 5, 6, 8, and 9 and showed that both the $CaCO_3$ content and the UCS of the sample increased with increasing pH. Ferris et al. [57] found that hydroxide ions improve the pH around the cells and that the appropriate pH for the MICP reaction ranges from 6.5 to 9.3. Urease still maintains good activity at pH of 9.0, and high pH can promote the conversion of bicarbonate ions to carbonate ions and the precipitation of $CaCO_3$ [58]. In summary, pH has an important influence on the growth and metabolism of urease-producing bacteria as well as on $CaCO_3$ precipitation and soil property improvement. High pH is conducive to the precipitation of $CaCO_3$ and improvement in the solidified soil strength. In actual engineering projects, a higher pH should be selected for solidification by MICP while considering the specific engineering needs. The impact of pH on the growth of bacteria, bacterial metabolic activity, urease activity, $CaCO_3$ morphology, $CaCO_3$ precipitation, and strength is summarized in Table 2.

*3.4. Temperature.* Changes in the ambient temperature affect the growth, reproduction, and functional metabolism of bacteria, thereby altering $CaCO_3$ production, precipitation rate, crystal type, crystal morphology, particle size, and cementation pattern of $CaCO_3$ between soil particles, which impacts the effectiveness of MICP-treated soil (Figure 6). Ferris et al. [57] investigated the effect of urease activity on the urea hydrolysis reaction in a groundwater environment of 10–20°C and found that urease activity depends on the temperature to a certain extent. A study by Nemati and Voordouw [61] showed that as the temperature increased from 20°C to 50°C, the urease activity and $CaCO_3$ formation rate increased, which further affected the crystal morphology of the $CaCO_3$ precipitates (Figure 7). Cheng et al. [55] found that the amount of precipitated $CaCO_3$ at 50°C was more than three times that at 25°C, while the strength dropped by 60% as the temperature decreased from 50°C to 25°C. The main reason for these results is that when the temperature is high, the generated $CaCO_3$ particles are relatively small and cover the surface of the sand particles, whereas when the temperature is low, less precipitation of $CaCO_3$ occurs. However, the particles are relatively large, which can effectively bind the sand particles and thus increase the strength. Gillman et al. [62] pointed out that as the ambient temperature increased from 5°C to 20°C, the rate of urea decomposition by urease-producing bacteria increased nearly 18-fold. Kralj et al. [63] experimentally discovered that a change in the temperature of the inorganic salt solution influenced the precipitation rate of $CaCO_3$ without affecting its crystal type. By comparing the effect of temperature on the improvement in the strength and water absorption of limestone with *S. psychrophila* and *B. sphaericus*, Muynck et al. [60] found that the bacteria more effectively reduced the water absorption of limestone at low temperature (37°C) and achieved the best improvement in the strength of samples at moderate temperatures (20°C and 28°C), while both bacteria exhibited poor performance in improving the strength and impermeability at low temperature (10°C). Keykha et al. [56] applied an equal amount of *S. pasteurii* solution to cement silty sand columns at pH of 9 and temperatures of 30, 40, and 50°C and found that the cemented sand column at 40°C had the highest UCS. Bang et al. [64] investigated the wind erosion resistance of *S. pasteurii*-cemented samples at 20, 35, and 45°C and concluded that the wind erosion resistance of the samples gradually increased as the curing temperature increased. Peng et al. [65] found that the lower the soil temperature was, the lower the strength and the higher the permeability of *S. pasteurii*-treated soil were. For example, the UCS of the sand column at 10°C was one-third of that at 25°C, while the permeability at 10°C was three orders of magnitude higher than that at 25°C (Figure 8). In summary, the appropriate temperature for the MICP process is similar to the optimum temperature for the growth and metabolism of urease-producing microorganisms. At low temperatures, the crystal particles of $CaCO_3$ are large, $CaCO_3$ is uniformly distributed between the particles, and overall strength of the cemented samples is high. In contrast, at high temperatures, the crystal particles of $CaCO_3$ are small and the strength of the cemented samples is low, but the $CaCO_3$ yield is high and the erosion resistance is enhanced.

*3.5. Grouting Methods.* The homogeneity of MICP-treated soil is currently one of the important factors restricting the development of MICP technology. Grouting methods determine the $CaCO_3$ content and distribution homogeneity of the treated soil and further affect the strength and permeability of the soil. Common grouting methods include injection, soaking, and spraying (Table 3). Whiffin et al. [66]

Table 2: Impact of pH on some factors.

| Factors | pH | Conclusion | Reference |
|---|---|---|---|
| Growth of bacteria | pH = 7, 8, 9 | The bacterial concentration is highest at pH of 9 | [59] |
| Bacterial metabolic activity | pH = 9.31, 11.15, 11.95, 12.24, 12.48 | The pH is kept above 12; the urease-producing bacteria are inactivated | [52] |
| Urease activity | pH = 6, 7, 8, 9, 10 | The enzyme activity increased at fast rate, peaking at pH of 8.0 and then decreasing slowly at higher pHs | [14] |
| | pH = 7, 8, 9 | The highest urease activity occurred at pH = 9 | [59] |
| $CaCO_3$ morphology | pH = 6.0, 6.5, 7.0, 8.0 | High pH promoted $CaCO_3$ precipitation, and precipitates were mainly calcite crystals, gradually changing from prism to pyramid-like or irregular polyhedral shapes | [54] |
| $CaCO_3$ precipitation | pH = 3.5, 7, 9.5 | The maximum $CaCO_3$ deposition at pH = 9.5 | [55] |
| | pH = 9.31, 11.15, 11.95, 12.24, 12.48 | The initial pH decreased to below 11.15, and the $Ca^{2+}$ concentration begins to decrease, hence $CaCO_3$ precipitation | [52] |
| | pH = 5, 6, 8, 9 | The increase in pH increased the $CaCO_3$ precipitation | [56] |
| Strength | pH = 5, 6, 8, 9 | The compressive strength of silty clay soil samples increased steadily as pH increased from 5 to 9 | [56] |

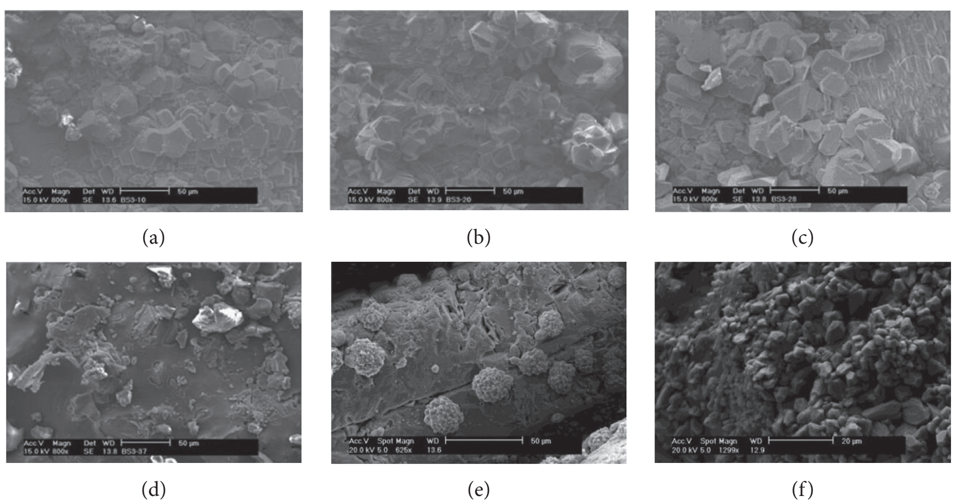


Figure 6: $CaCO_3$ crystals formed at different temperatures, (a)–(d) [60], (e)–(f) [55]: (a) 10°C; (b) 20°C; (c) 28°C; (d) 37°C; (e) 25°C; (f) 50°C.

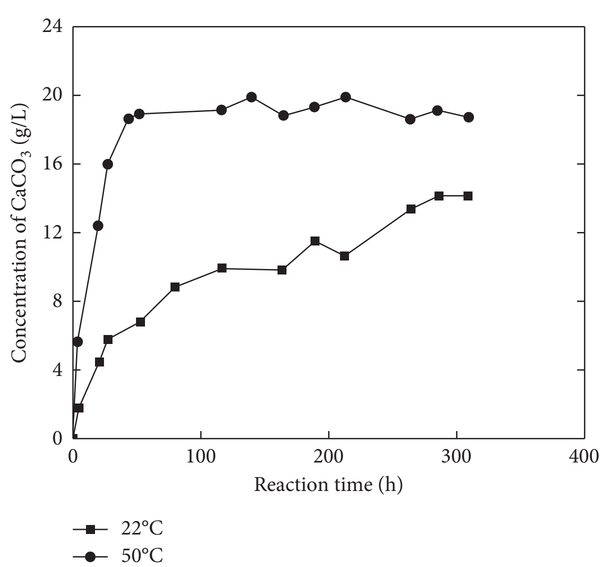


Figure 7: Effect of temperature on enzymatic $CaCO_3$ production [61].

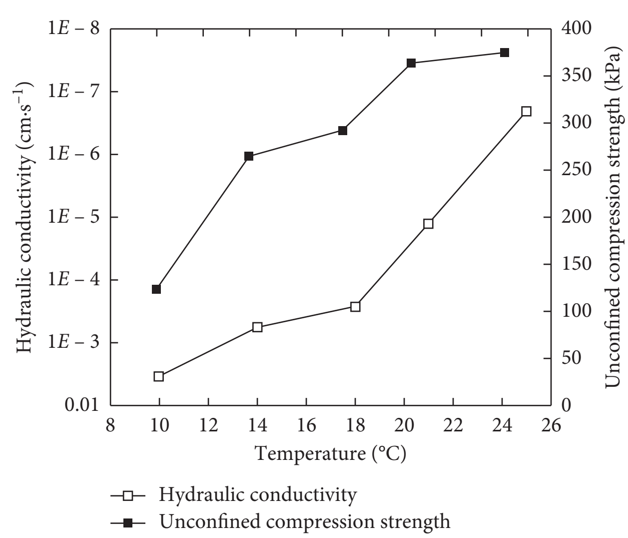


Figure 8: Unconfined compression strength and hydraulic conductivity of treated samples at different temperatures [65].

Table 3: Advantages and disadvantages of grouting methods.

| Method | Procedure | Advantage/disadvantage | Reference |
|---|---|---|---|
| Two-phase grouting method | Placement: bacterial injection, $CaCl_2$ injection | Avoiding clogging | [66] |
| Parallel injection method | The bacteria and the cementation fluid were injected at the same time | Most calcite precipitating close to the inlet area | [67] |
| Repeated staged injection method | Bacterial injection, static period for 2 h, cementation fluid injection | Reducing porosity | [67] |
| Soaking method | Bacterial injection, soaking in the cementation fluid | The pores becoming smaller or even blocked, the cementation fluid being difficult to penetrate into | [68] |
| Electro-biogrouting method | Imposing electric field with a certain voltage gradient | Promoting the diffusion of biomass in soil pore, more uniform | [69] |
| Spraying method | Spraying bacteria solution evenly on the surface of sand practices | Improving the average hardness and compressive strength | [70] |
| Unsaturated percolation method | Using peristaltic pump to control the infiltration rate (the saturation is about 75%) | Having better curing effect, with the average compression strength being 19.7 MPa | [71] |

proposed a 2-phase grouting method, in which a bacterial suspension is injected into the soil and allowed to stand for a period of time. After the bacteria adhere to the surface of the soil particles, the cementation solution is injected to start solidification. This grouting method can prevent the clogging of the injection nozzle to a certain extent and improve the homogeneity of the $CaCO_3$ that is generated inside the soil. Compared to the mixed injection method, this method can reduce the porosity of the solidified soil by an additional 20% using the same cementation solution and bacterial suspension [67]. Liang et al. [68] noted that the solidification result using the step-by-step grouting method on sand was superior to that using the soaking method. When the mixture of 0.5 mol urea and 0.5 mol calcium chloride was poured at 50 mL/h, the average UCS with the former method increased by 15.58% compared to that with the soaking method. Cui et al. [72] introduced 0.05 mol/L $CaCl_2$ solution (referring to a mixed bacterial suspension) into a pure bacterial suspension for artificial intervention in the bacterial distribution and comparatively analyzed the effects of the injection of pure/mixed bacterial suspension, mixed bacterial suspension solution, and traditional pure bacterial suspension on the dynamic characteristics of biocemented sand. The test results showed that the injection of a pure/mixed bacterial suspension could effectively increase the uniformity of the distribution of $CaCO_3$ crystals in the biocemented soil, thereby obtaining biocemented soil with a higher $CaCO_3$ content, larger dynamic elastic modulus, and greater energy dissipation capacity. Xu et al. [69] used the electro-biogrouting method (EBM) to solidify silt and concluded that compared with the traditional gravity grouting method and the low-pressure grouting method, the EBM promoted the diffusion of microorganisms in soil pores by applying an electric field with a certain voltage gradient in the silt, resulting in more uniform dispersion of $CaCO_3$. Zhan and Qian [70] treated sand with biocement using a spraying method; after seven spraying applications, the average hardness and compressive strength of the samples reached 31.5 GPa and 0.67 MPa, respectively. Andres et al. [71] compared the solidification results using the percolation method and reverse injection method to treat unsaturated soil samples and found that the percolation method achieved a better solidification result, with an average UCS of 19.7 MPa. In summary, the distributed grouting method, multiconcentration grouting method, and electroosmotic grouting method are more valuable than the single grouting method in terms of improving the uniformity of the distribution of $CaCO_3$ and thereby increasing the overall strength of the solidified soil.

*3.6. Particle Size.* The soil particle size is an important factor affecting the MICP solidification results (Table 4), mainly because it affects the retention, adsorption, and transport of microorganisms between sand particles. Generally, bacterial cells have a size of approximately 0.5–3.0 $\mu$m; in particular, the sizes of *Sporosarcina* and *Bacillus* species are mostly 1–5 $\mu$m. Too small soil particles hinder the flow of bacteria and cementation solution in soil, meaning that $CaCO_3$ cannot be formed or is unevenly distributed, thereby resulting in poor solidification [8]. In addition, only the $CaCO_3$ deposited at the contact points between the soil particles can effectively improve the mechanical properties of the soil; excessively large particles reduce the number of contact points between soil particles, causing $CaCO_3$ to be mostly distributed on the surface of coarse particles, which causes poor solidification. Regarding the size compatibility between soil particles and microorganisms, Li et al. [75] proposed a formula for judging the suitability of soil characterized by the permeability coefficient for MICP technology and verified the formula through tests on the mechanical properties of nine types of soil. Cui et al. [73] investigated the effect of particle size on biocementation and concluded that the interparticle pores of sand of smaller particle size are more easily densely filled by $CaCO_3$ crystals, obtaining a larger proportion of effective $CaCO_3$ crystals and thus a biocemented sample that is "structurally" stronger and has a higher UCS. Rebata-Landa [44] tested cemented soil columns composed of 11 types of soil including kaolin, silt, fine sand, coarse sand, and gravel, from which the particle size ranges for effective cementation were obtained, and proposed a formula for calculating the $CaCO_3$ content

Table 4: Effect of soil particle size on consolidation.

| Material | Particle size | Conclusion | Reference |
|---|---|---|---|
| Sand | A: 1.25~2.5 mm<br>B: 0.5~1.25 mm<br>C: 0.04~0.5 mm | Group C has the highest unconfined compression; the intergranular pores are easily filled by $CaCO_3$ crystals | [73] |
| Sand | Mikawa $D_{50} = 600\,\mu$m<br>Toyoura $D_{50} = 200\,\mu$m | The UCS of Mikawa sand is higher than that of Toyoura sand | [74] |
| Sand | Ottawa silica<br>$D_{50} = 0.46$ mm<br>Mississippi<br>$D_{50} = 0.33$ mm | The solidification strength of Ottawa silica sand is higher than that of Mississippi sand | [42] |

suitable for two particle size ranges. Amarakoon and Kawasaki [74] compared the UCS of cemented silica sands with two particle size ranges (Mikawa sand $D_{50} = 600\,\mu$m and Toyoura sand $D_{50} = 200\,\mu$m) and found that the Mikawa sand, with a larger mean particle size, exhibited a better solidification result, with a UCS of 3 MPa, while the Toyoura sand, with a smaller mean particle size, showed a fair solidification result, with a UCS of approximately 1 MPa. Similar to that of Amarakoon and Kawasaki, a study by Zhao et al. [42] concluded that the solidification strength of Ottawa silica sand with $D_{50} = 0.46$ mm was more than two times that of Mississippi sand with $D_{50} = 0.33$ mm. In summary, particle size is the main factor affecting the MICP solidification results. Soil cementation and strength can be improved by enhancing the particle size gradation of soil. Solidification by MICP should consider the size of the soil particles. However, in practical engineering, the grades of soil particles vary, and different consolidation processes (concentration of the bacterial suspension, cementation solution concentration, grouting methods, etc.) could be used to consolidate the soil.

## 4. Engineering Properties of Soil Solidified by MICP

*4.1. Strength.* UCS and shear strength are important indicators for characterizing soil strength. By comparing the strengths of intermediate compressible and highly compressible clays before and after solidification by MICP, Animesh and Ramkrishnan [76] found that the UCS values of the two types of soils increased 1.5 times and 2.9 times due to solidification and determined the optimum mixing ratio of the bacterial suspension and cementation solution. Van Paassen et al. [77] noted that $CaCO_3$ crystals produced by MICP form bridges between sand particles, thereby increasing the strength and stiffness of the sand, and evaluated the feasibility of ground improvement by MICP through the analysis of the effects of the substrate solubility, $CaCO_3$ yield, reaction rate, and side-products on MICP. Gowthaman et al. [78] found that the UCS of soil reached 420 kPa after 10 days of treatment of indigenous microorganisms with pure chemicals. In comparison, after treatment of the indigenous microorganisms with inexpensive low-grade chemicals, the UCS of the soil was significantly improved, reaching 820 kPa, and the treatment cost was reduced by 96%. Rong and Qian [79] presented the feasibility of the solidification of loose sandstone particles by MICP. The cemented bio-sandstone had a satisfactory compressive strength at certain ages, and as the magnesium carbonate content increased, the compressive strength and porosity both increased. Zhao et al. [42] compared the mechanical properties of soil treated with *S. pasteurii* and urease and found that the UCS of the biotreated soil was approximately five times that of the urease-treated soil, indicating that bacteria are more effective for soil solidification. Liu et al. [80] investigated the effect of ultrasound on MICP-treated sand and showed that after treatment by optimal ultrasonic irradiation, the production of $CaCO_3$ in aqueous solution and in sand columns increased by 28.5% and 35.6%, respectively, and the UCS of the treated sand samples reached 1.25 MPa, which was 91.6% higher than that of the control group.

Although the MICP treatment method can significantly improve the strength of the soil, it can also cause significant brittle failure of the treated soil, which is a problem that can be effectively addressed using the fiber reinforcement method. Choi et al. [81] found that the use of polyvinyl alcohol (PVA) fiber reinforcement can increase the UCS and splitting tensile strength of sand by 138% and 186%, respectively, while decreasing the permeability by 126% and reducing the brittleness (expressed as the ratio of UCS to splitting tensile strength) to half that of plain sand. Fiber reinforcement can increase the precipitation rate and yield of microbially induced $CaCO_3$, and the cementation effect of $CaCO_3$ has a facilitating role in fiber reinforcement. Hence, fiber reinforcement technology and MICP technology can be combined to achieve complementary advantages (Figure 9). Fang et al. [82] pointed out that the addition of fibers not only reduced the permeability of MICP-treated coral sand but also improved the dry density and uniaxial tensile strength as well as the ductility, failure strain, and tensile strength. The fiber content had a greater effect on the mechanical properties of the treated coral sand than the fiber length, with an optimum added fiber content of 0.2% and optimum fiber lengths of 9 mm and 12 mm. Xie et al. [83] pointed out that the incorporation of fibers into bio-cemented sand could greatly improve the UCS and residual strength of the soil samples and significantly improve the toughness of the soil samples upon failure (Figure 10). When a sample is damaged, the fibers act as "bridging" (Figure 11) that crosses the fracture and can bear a certain tensile stress, thus effectively inhibiting the further development of the fracture and delaying the total failure of the sample. Zheng

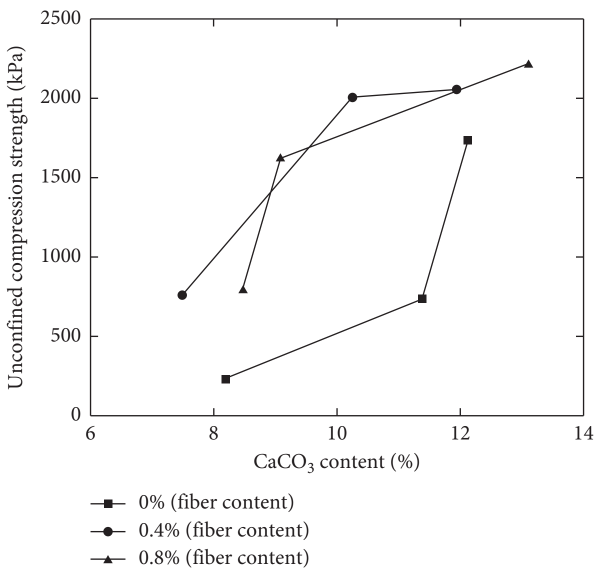


Figure 9: Unconfined compression strength plotted against $CaCO_3$ content for different fiber contents [81].

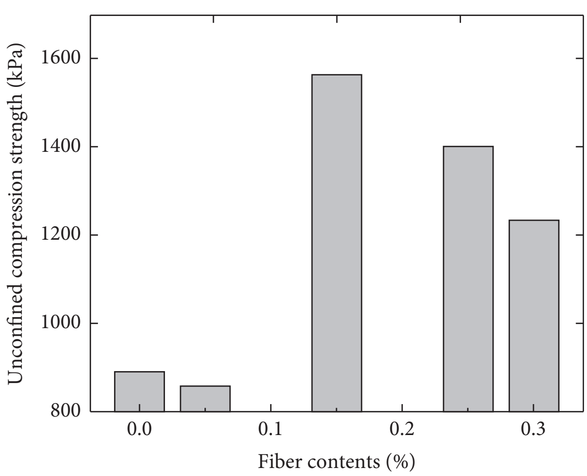


Figure 10: Unconfined compression strength of samples with different fiber contents [83].

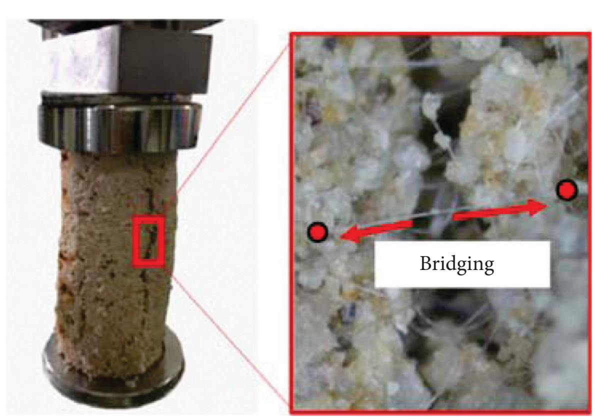


Figure 11: "Bridging" effect of fiber impedes the further development of crack [83].

et al. [84] pointed out that, in the MICP process, $CaCO_3$ crystals can be effectively deposited onto the surface of the fibers, improving their surface roughness, and that a mixture of $CaCO_3$ and sand can provide anchorage to the fibers, thereby improving the shear strength and strain-softening properties of the biocemented sand to a certain extent. In summary, MICP technology can significantly improve the strength of soil, and a combination of MICP and fiber reinforcement can effectively solve the brittle failure problem of the solidified soil while improving the soil strength. In actual engineering projects, suitable solidification methods can be selected in response to engineering needs.

*4.2. Stiffness.* Stiffness is an important indicator for characterizing the ability of soil to resist elastic deformation and can be measured by the shear wave velocity obtained from shear modulus or bender element tests. DeJong et al. [11, 85, 86] used bender elements to monitor the variation in the shear wave velocity of microbially induced cemented sand and obtained the relationship between the shear wave velocity and the duration of injection and treatment. The shear wave velocity reached a maximum of 540 m/s at 1700 min, and the shear stiffness was significantly improved. In addition, grouting and treatment tests were carried out on a shallow foundation model, and static load test results showed that the bearing capacity and stiffness of the foundation were significantly improved, as demonstrated by a nearly fivefold decrease in the settlement under the same load. Van Paassen et al. [87, 88] carried out in situ grouting and improvement of a 100 $m^3$ sand foundation. After the completion of treatment, shear wave velocities of different regions were detected. The shear wave velocities in different parts of the sand foundation increased significantly with an increase in cementation time, with an average shear wave velocity of 300 m/s, indicating that stiffness of the sand foundation increased effectively. In addition, it was suggested that the increase in stiffness should be quantified as a function of the volume of the injected solution and the distance from the injection points. Liu et al. [89] concluded that the role of $CaCO_3$ in the MICP process is to strengthen the connection between adjacent soil particles and increase the stiffness of the soil so that it can bear higher stress under the same strain. In summary, MICP technology can significantly improve the soil stiffness and is effective in improving soils that are mainly subjected to compressive stress.

*4.3. Liquefaction Resistance.* Saturated loose sand is prone to liquefaction under seismic or wave loads, causing severe damage to buildings or transportation infrastructure. Therefore, it is necessary to reinforce soil against liquefaction. As a new foundation treatment method, MICP reinforcement technology could effectively improve the antiliquefaction performance of liquefied sand foundation (Figure 12). After MICP treatment, the compactness of sand is increased, and the cement could improve the cementation between particles, which can improve the strength of soil, reduce the internal pore water pressure, inhibit the occurrence of large strain of sand, and protect against the

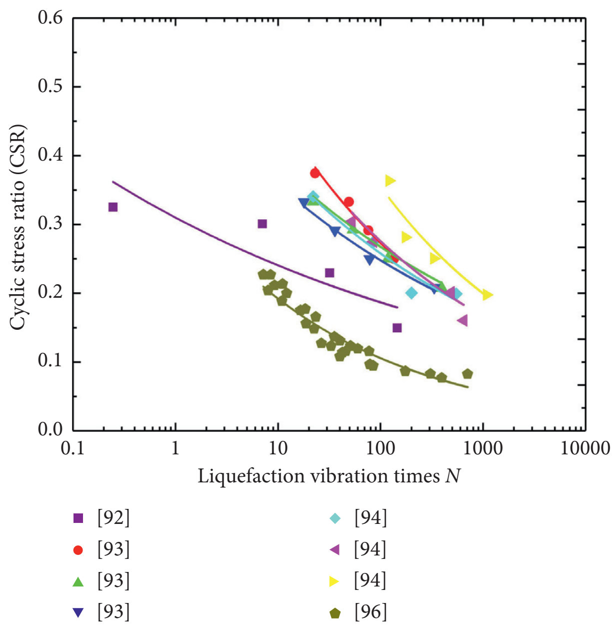


Figure 12: Relationship between CSR and $N$.

liquefaction effect. Liu et al. [90] divided the development of the pore pressure in MICP-cemented calcareous sand into four stages: the initial stage, stable development stage, rapid development stage, and complete liquefaction stage. The dynamic shear stress ratio and deformation resistance of the cemented calcareous sand were markedly improved, indicating that MICP cementation can significantly improve the liquefaction resistance of calcareous sand. Zhang et al. [91] carried out shake table tests on MICP-treated calcareous sandy foundations in the South China Sea. Compared to those of untreated foundations, the excess pore water pressure and ground settlement of the MICP-treated calcareous sandy foundation decreased, indicating that the liquefaction resistance of the treated calcareous sandy foundation was considerably improved. Zamani and Montoya [92] investigated the liquefaction characteristics of MICP-treated fine silty sand and found that the liquefaction resistance of fine silty sand was significantly improved after MICP treatment, the efficiency of which depended on the fines content (dictating the relative density) and the fabric governing the structure, indicating that MICP is a potential method for silty sand improvement. Xiao et al. [93] argued that the cyclic shear stress and confining pressure are important factors affecting the liquefaction of calcareous sand; an increase in the confining pressure causes a decrease in the liquefaction resistance of plain sand and MICP-treated sand samples, and an increase in cyclic shear stress causes a decrease in the cyclic strength. The number of liquefaction cycles increased with an increase in $CaCO_3$ content, indicating that MICP treatment can significantly improve the liquefaction resistance of calcareous sand. Han et al. [94] conducted dynamic triaxial tests of MICP-treated sand and concluded that solidified sands with different strengths can meet different engineering requirements, the solidification time can be shortened to 1 to 2 days by reducing the use of bacterial suspensions and nutrients, and MICP is effective in improving the liquefaction resistance of sand. Han and Cheng [95] characterized the effect of different calcium salts on the MICP solidification efficiency using ammonium and found that calcium acetate was the most effective in improving the mechanical properties of liquefiable sand and that the hydraulic permeability of the solidified sample was significantly reduced. Cheng et al. [96] explored the liquefaction resistance of solidified sand using dynamic triaxial tests and found that when the amount of $CaCO_3$ that precipitated in the solidified sample was higher, the axial deformation of the sand sample was smaller, and the increase in the liquefaction resistance was more pronounced. The use of soil desaturation induced by microorganisms to alleviate soil liquefaction is also a method for preventing foundation liquefaction. The desaturation effect of soil is realized by the gas produced by the microbial denitrification process, which can effectively reduce the saturation of sand.

*4.4. Permeability.* The calcite formed by microbial mineralization fills the pores of soil particles, reducing the porosity and permeability of the soil (Figure 13). Lai et al. [97] tested the permeability of two kinds of sand using MICP technology at low pH; the results showed that the sample with a large decrease in permeability contained more $CaCO_3$, while the sample with a small decrease in permeability contained less $CaCO_3$. Therefore, the content of $CaCO_3$ was the key factor that affected permeability. Chu et al. [98] used MICP technology to form a cementation layer of a certain thickness on the sand surface. When an average of 2.1 kg of calcium (Ca) per $m^2$ of sand surface was precipitated, the permeability coefficient of the sand ranged from $10^{-4}$ m/s to $10^{-7}$ m/s. Qabany and Soga [47] investigated the permeability of British standard grade $D$ silica sand with different dry densities and cementation concentrations. The results demonstrated that both loose samples and dense samples showed a reduction in permeability with an increase in the amount of $CaCO_3$ precipitation. Shen et al. [99] used *Bacillus megaterium* to solidify sandy clayey purple soil. The results showed that, with the formation of $CaCO_3$, its permeability initially decreased with an increase in $CaCO_3$ content and then decreased by two orders of magnitude. Jawad and Zheng [100] pointed out that MICP technology has greater strength improvement and better solidification efficiency for dry sand and a better reduction in permeability for saturated sand. From the above-mentioned analysis, it can be concluded that, with $CaCO_3$ precipitation, the soil permeability decreased significantly, and the permeability decreased rapidly as the $CaCO_3$ content increased. Therefore, MICP technology has great prospects for the anti-seepage of dams, embankments, and reservoirs. Notably, MICP has been utilized to improve soil properties for different purposes [101], but these properties are not independent. The permeability, strength, shear wave, and uniformity of precipitation are correlated. Dawoud et al. [102, 103] examined the relationship between hydraulic conductivity, $s$-wave velocity, and calcite content with different treatment solutions, confining pressure and back

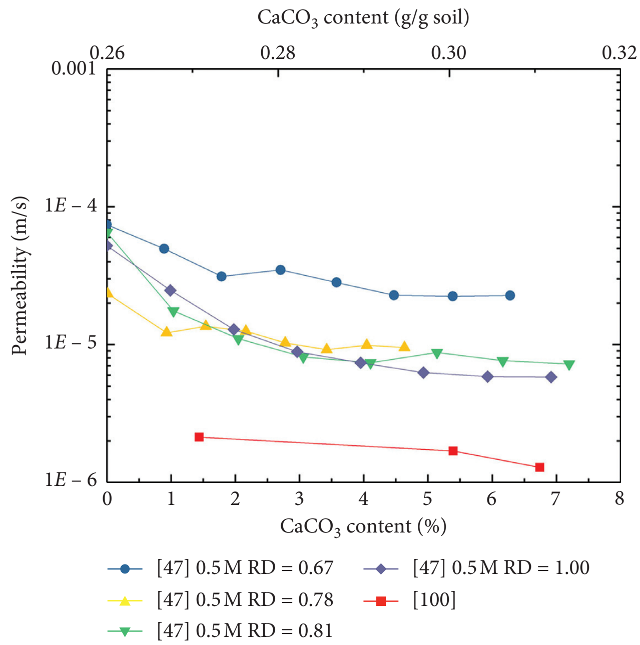


FIGURE 13: Relationship between permeability and $CaCO_3$ content.

pressure. The results indicated that the *s*-wave velocity could be considered a direct indicator of the calcite content. All samples had similar characteristic, and the hydraulic conductivity at early stages of treatment decreased slightly and then decreased quickly. The lack of uniform precipitates caused earlier clogging of the pores and a dramatic decrease in hydraulic conductivity. The researchers suggested that less uniformity was attributed to the common distribution of the injected bacteria and examined the effect of surfactants on the transport and distribution of the bacteria and on MICP efficiency and distribution. The results indicated that less than 2% of the bacterial cells were retained in sand columns when surfactant was applied, and had a better distribution of calcite precipitation.

*4.5. Durability.* Durability is an important indicator for evaluating the long-term performance of MICP-treated sand under environmental parameters (such as wet-dry cycles, freeze-thaw cycles, acid rain, wind erosion, etc.). Cheng et al. [104] investigated the mechanical properties of MICP-cemented biobricks; showed that the compressive strength of biobricks prepared under partially saturated conditions was 9 MPa, which is twice that under fully saturated conditions; and evaluated their water absorption, resistance to salt attack, and fire resistance. The results showed that biobricks are suitable for use as a building material and could also be a green alternative to traditional fired clay or cement bricks. Liu et al. [105] employed MICP to treat tiles to form an erosion-resistant layer and found that MICP could significantly improve the water resistance of ceramics, with higher concentrations of bacteria and cementation reagents enabling the sediment layer to provide better protection. The concentration threshold reduced the positive effect of the bacterial concentration on the water resistance of the surface of the sample, and the protection layer provided satisfactory durability and effectively alleviated the weathering of the ancient clay roofing tiles. Chae et al. [106] found that, with a wind speed of 15 m/s, the amount of wind erosion of medium sand or fine sand after solidification using 0.5 M MICP solution was relatively small, so appropriate injection methods should be chosen to reduce soil erosion, considering the soil properties. According to the wind tunnel test results, Nikseresht et al. [107] argued that an increase in penetration resistance decreased the soil loss of soil MICP solidified with vinasse and molasse to approximately one-third of that of the blank sample, indicating that the soil treated by molasse and vinasse had a high resistance to wind erosion. Huang et al. [108] pointed out that due to the poor cementation between particles within the MICP-grouted shale residual soil, when the frost heave force generated by the pore water during the freeze-thaw cycle is greater than the MICP cementation strength, the cementation fails, which causes cracking of the sample, and the shedding area increases with an increase in water content and the number of freeze-thaw cycles. Gomez et al. [109] applied a bacterial suspension and cementation solution to cement and stabilize the surface of loose tailings sand, eventually forming a 2.5 cm thick hard cementation layer. The test results showed that the effective improvement depth reached 28 cm, significantly increasing the erosion resistance of the loose tailings sand. Jiang et al. [110] conducted a series of seepage erosion tests on mixed soil samples (sand : kaolin = 5 : 1) after biocementation and solidification. The results showed that the critical hydraulic gradient and shear stress of the solidified soil were significantly improved, while the amount of internal erosion under the same hydraulic conditions was significantly reduced. Cheng et al. [111] continuously injected artificial acid rain (pH = 3.5) into cemented sand columns. He found that mass was continuously lost and that the strength was reduced by up to 40%, indicating that biocemented sand has a low resistance to acid rain erosion, mainly because the $H^+$ in acid rain reacts with the $CaCO_3$ between the sand particles, destroying the cementation of $CaCO_3$. Liu et al. [112] conducted a series of experiments on the long-term engineering performance of MICP-treated sandy soil under wet-dry cycles, freeze-thaw cycles, and acid rain conditions. The results showed that the durability of MICP-treated soil was weak under these conditions. The drop in UCS was nearly 80% after one wet-dry cycle, 58% after 15 freeze-thaw cycles, and 83% after 15 days of immersion in acid rain solution. In conclusion, the resistance to wind and water erosion of MICP-treated sand is significantly improved; freeze-thaw and wet-dry erosion are weak; and the resistance to acid rain erosion is poor because of the dissolution of $CaCO_3$ in acid.

## 5. Engineering Applications of Soil Solidification Using MICP

*5.1. Ground Improvement.* MICP technology can significantly improve the strength, stiffness, and liquefaction resistance of soil and is suitable for ground improvements such as improving the permeability of sand. Van Paassen et al.

[87, 88, 113] carried out a large-scale in situ sand foundation grouting and improvement experiment (Figure 14). First, a 1 $m^3$ sand pile experiment was conducted. With a single-point injection method, the bacterial suspension and cementation solution were sequentially injected from the center of the cubic sand pile at a constant flow rate. After 40 days of continuous treatment, the average content of $CaCO_3$ precipitates in the sand body was 100 kg/$m^3$, and the highest uniaxial compressive strength of the sand sample was 9 MPa. On this basis, a 100 $m^3$ large-scale grouting experiment was conducted. Three injection wells and three pumping wells were arranged, and treatment was carried out continuously for 16 days using grout of the same composition to obtain a 40 $m^3$ clearly visible cemented sand body with a wedge shape. The average amount of $CaCO_3$ precipitation was 110 kg/$m^3$, the uniaxial compressive strength of the cored samples ranged from 0.7 to 12.4 MPa, and the average shear wave velocity of the sand foundation was 300 m/s, indicating that the strength of the foundation after MICP treatment was significantly improved. Liu et al. [114] carried out an in situ experimental study of the MICP treatment of calcareous foundations on an artificially reclaimed island. The results showed that after three to four MICP treatments, a gradual increase in ground surface strength was detected. After nine MICP treatments, the surface strength was greater than 10 MPa and reached 20 MPa; the treated depth of the foundation reached 70 cm; and the UCS reached 821 kPa. Montoya et al. [115] conducted MICP treatment tests on the liquefiable sand free field using a centrifuge shake table and found that, after treatment, the pore pressures at different depths of the site decreased to varying degrees with different seismic intensities and that the postshaking surface settlement of the MICP-treated site was significantly smaller than that of untreated loose sand but that the surface acceleration was enhanced to some extent compared to that of untreated loose sand. Darby et al. [116] conducted 80 g centrifuge shake table tests on three different sets of treatment models with $CaCO_3$ contents of 0.8%, 1.4%, and 2.2%; the results showed that, after MICP treatment, the cone penetration resistance of the sand increased from 2 MPa to 5, 10, and 18 MPa and that the shear wave velocity increased from 140 m/s to 200, 325, and 660 m/s. As the level of cementation increased, the liquefaction resistance increased, no further liquefaction occurred, and the mechanical properties of the model sample gradually changed from those of soil to those of rock. He et al. [117] conducted shake table tests using microbial denitrification to produce gas bubbles and found that, at an acceleration of 0.5 m/$s^2$, the untreated saturated loose sand was completely liquefied, with an excess pore pressure ratio near 1; the surface settled markedly; and the volumetric change reached 5%. After different levels of microbial treatment, the saturation of the sand decreased within the range 95%–80%. For the model with 80% saturation and at an acceleration of 0.5 m/$s^2$, the excess water pressure ratio was only 0.1, and almost no settlement occurred at the surface.

Figure 14: Field test of MICP foundation reinforcement [113].

*5.2. Foundation Seepage Control.* The application of microorganisms in the field of soil antiseepage mainly uses the mineralization of microorganisms and microbial membranes. Compounds produced by microbial mineralization have the effect of filling and cementing in materials. Extracellular polymeric substances (EPS) secreted by microorganisms can be attached to the surface and interior of porous materials to form microbial membranes, which reduces the permeability of porous materials. Chu et al. [98] used microbial technology to construct a reservoir using 2.1 kg of $CaCl_2$ per $m^2$ of the sand surface, and after MICP surface treatment, the permeability of the sand could be reduced from an order of $10^{-4}$ m/s to an order of $10^{-7}$ m/s. Subsequently, samples were taken from the pool bottom for a four-point flexural test, the flexural strengths were found to be 90 to 256 kPa, and the UCSs of the sidewalls and pool bottom were found to be 215 to 932 kPa. Subsequently, samples were taken from the base of the pond for a four-point bending test; their flexural strengths were found to fall into the range of 90 to 256 kPa; and the UCSs of the samples taken from the walls and the base of the pond bottom were found to fall between 215 and 932 kPa; that is, different parts of the pond had strengths of varying degrees (Figure 15(a)). Liu et al. [119] reinforced the levee model by spraying microbial cells and nutrient salts and performed flume tests on the treated model. After multiple days of scouring, no erosion damage occurred in the overall model, except for a small amount of fine sand on both sides of the model sample, which was produced by the water flow. The permeability coefficient of the surface soil decreased from $4 \times 10^{-4}$ m/s to $7.2 \times 10^{-7}$ m/s, and the maximum UCS was 9 MPa. Tan et al. [118] carried out field tests of MICP seepage control in three sections of the clay banks of the Dawa Reservoir by utilizing the mineralization of microorganisms and monitored both internal water heads with the pressure measuring pipe inside the dam and the leakage amount at the leakage site. The results showed that this technology could rapidly reduce the permeability coefficient of the clay bank by two orders of magnitude (Figure 15(b)). Gao et al. [120] proposed a


(a) (b)

Figure 15: Permeability reduction effect: (a) a low-permeability layer surrounding the boundary of the pond [98]; (b) filled pores in the dam [118].

construction process for seepage control channels by MICP treatment: the channel site to be constructed was treated using the injection pipe process, followed by excavation, and then the channel surface was treated using spray and soaking techniques (with treatment liquid consisting of ureolytic bacteria, 0.5 mol/L calcium chloride, and 0.5 mol/L urea). The test results showed that the construction process can effectively reduce the permeability coefficient of the sand surface and thus meet the engineering requirements. Cheng et al. [121] applied $CaCO_3$ precipitation techniques combined with the reaction of sodium alginate with $Ca^{2+}$ ions to form a gel-like calcium alginate for sand seepage control treatment. The test results showed that the permeability of the treated sand decreased from $5.0 \times 10^{-4}$ m/s to $2.2 \times 10^{-9}$ m/s and that the seepage control efficiency increased by one to two orders of magnitude compared to that treated with pure MICP technology. Blauw et al. [122] were among the earliest to apply biofilm technology to seal leakages in clay-core embankments along the Danube River, Austria, using in situ microbial growth for 23 days of nutrient injection. After 10–14 weeks, the discharge of the dam per unit time significantly decreased from 17.33 $m^3$ per day before repair to 2.35 $m^3$ per day after repair. The leakage at the grout inlet was tested again after 5 months and was found to be only 10% to 20% of that prior to treatment, indicating the feasibility of biofilm technology in the restoration of Earth structures. Therefore, microbial technology can achieve soil seepage prevention. For different engineering problems, different types of microbial reaction processes and construction techniques can be adopted. However, the natural environment of an actual project is relatively complex, and its durability needs further study.

*5.3. Erosion Resistance of Foundations.* MICP-treated soil has good resistance to hydraulic and wind erosion. Bang et al. [64] conducted six biocement surface spray treatments of a sandy embankment model. The flume scour test of the hydraulic model showed that the treated model experienced continuous erosion from the overtopping water with different flow rates for 30 days without collapse. The anatomical test of the model revealed that $CaCO_3$ aggregated within 3 cm of the surface and formed a crust-like layer. In addition, biocement can be utilized to control piping erosion within the soil. Jiang et al. [110] showed that, after the sand-kaolin mixture was treated with biocement, the critical hydraulic gradient increased significantly, and the mass of solid particles carried away by water erosion was greatly reduced. To prevent and control the erosion damage to embankments caused by overtopping flow, Liu et al. [119] improved the mechanical properties of embankment surface sand by spraying microbial cells and nutrient salts into the embankment surface layer so that the gelation of $CaCO_3$ could rapidly precipitate in the sand pores. The results showed that the maximum UCS could be 9 MPa and that the permeability coefficient was reduced from $4 \times 10^{-4}$ m/s to $7.2 \times 10^{-7}$ m/s. MICP technology can effectively improve the erosion resistance of the surface of the embankment model to prevent and control damage caused by overtopping flow (Figure 16). Naeimi and Chu [123] performed wind tunnel tests to verify the ability of microorganisms to treat sand surfaces for dust suppression and wind erosion resistance and estimated the amount of biological dust suppressants needed, which was less than the amount of conventional dust suppressant currently used for road and airport field applications. To study the feasibility, stability, and vegetation recoverability of the field stabilization of surface sand, Gomez et al. [109] conducted field tests of MICP-treated surfaces at a Canadian mine site. The test area consisted of four treated plots of different concentrations of MICP reaction solution, where each plot had an area of 2.4 m × 4.9 m and a design treatment depth of 0.3 m. After surface treatment, dynamic penetration tests showed that the ground penetration resistance significantly increased, with the crust thickness ranging from 0.64 to 2.5 cm. The $CaCO_3$ content on the surface of the crust was approximately 2.1%, and the formed $CaCO_3$ decreased with an increase in depth and was only 0.5% at a depth of approximately 10 cm. The results from the standard penetration test showed no significant degradation of the ground strength on day 64. Zhan et al. [124] used the enzymatic action of *Paenibacillus mucilaginosus* to absorb and transform $CO_2$ to produce carbonate ions, which then react with calcium ions present in the environment to mineralize and form a calcite cementation layer with certain mechanical

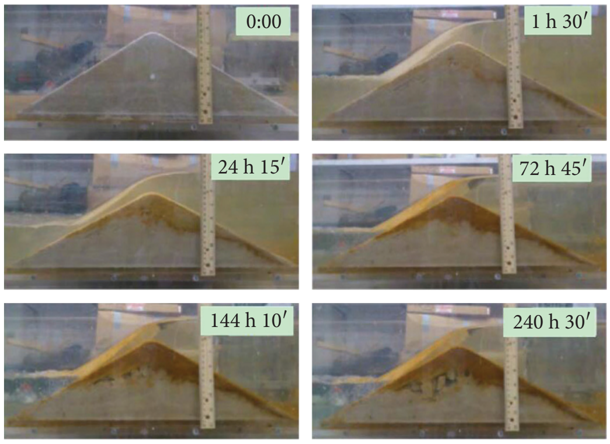


Figure 16: Flume test of treated model by spray method [119].

properties for dust suppression, and carried out a 900 $m^2$ field application using this technique. After biotreatment, the treated field area had an average solidification thickness of 13.2 mm and Shore hardness of 24.6. In a wind erosion test with a wind speed of 12 m/s, the mass loss of the treated soil was 30 g/($m^2$·h), representing a decrease from 2600 g/($m^2$·h) of the original untreated soil. In the rainfall erosion test, the mass loss of the biotreated soil was approximately 60 g/($m^2$·h), showing a decrease in the mass loss of 750 g/($m^2$·h) of the original untreated soil, and the residual Shore hardness remained more than 90% of that prior to rainfall erosion. In addition, the hard structure of the calcite surface layer formed by microbial mineralization can also improve soil moisture retention, which is beneficial to soil water retention and plant growth. Li et al. [125] combined straw checkerboard barrier (SCB) technology and MICP technology to study desertification prevention and control, for which 16 key factors were summarized and their parameters and effects were analyzed in detail. The results showed that MICP technology can effectively compensate for the deficiencies of SCB technology, and a combination of the two technologies could achieve satisfactory results in mitigating desertification and accelerating the process of sand fixation, vegetation recovery, and ecological restoration.

*5.4. Fixation of Heavy Metals.* Microbial mineralization converts ionic heavy metals into solid-phase minerals and precipitates heavy metal ions, resulting in a decrease in their bioavailability. The metabolites (sulfur ions and phosphate ions) of some microorganisms undergo precipitation reactions with metal ions, converting toxic and harmful metal elements to nontoxic or low-toxicity metal precipitates. Fujita et al. [126] conducted tests on the remediation of $^{90}Sr$ contamination at a site in the state of Washington in the Unites States, testing the addition of urea and molasses to promote the growth of native ureolytic microbes and the solidification and precipitation of heavy metals in the field and circulating the treatment using injection piping several meters apart during treatment and pumping. The test results showed that MICP technology can be employed to treat $^{90}Sr$ contaminants in the field. Xu et al. [127] also carried out a field experimental study on the removal of heavy metal ions by MICP. Gram-positive bacteria were utilized as the strain for mineralization to treat an abandoned iron ore mine, with a field remediation depth of 20 cm and an area of 1000 $m^2$. The exchangeable concentrations of As, Pb, Cd, Zn, and Cu in the contaminated soil were 14.01, 4.95, 0.64, 33.46, and 12.95 mg/kg before treatments. The parameters for field MICP spray treatment included an ambient temperature of 30°C, pH of 5.5, urea dosage of 12.65 kg, and bacterial dosage of 160 L. After remediation, the exchangeable concentrations of the abovementioned heavy metals were reduced to 2.37, 1.25, 0.311, 16.67, and 3.42 mg/kg, respectively. Tests have shown that treatment of heavy metal-contaminated soil by spraying with salt mineralization microbes is effective, with the highest heavy metal removal rate being 83%, thereby significantly lowering the risk that heavy metal ions will be absorbed by crops. Cheng et al. [128] selected soil *Bacillus* bacteria as carbonate mineralization bacteria and used their substrate-induced enzymatic decomposition to produce $CO_3^{2-}$ and thus mineralize and solidify the available heavy metals in the soil, such as the precipitation of $Cd^{2+}$ to a stable state of carbonate, obtained in an available heavy metal removal rate that exceeds 50%. Ganesh et al. [129] used iron-reducing bacteria (*Shewanella alga*) and sulfate-reducing bacteria (*Desulfovibrio desulfuricans*) to reduce hexavalent uranium to tetravalent uranium and then precipitated tetravalent uranium to form uraninite ($UO_2$ (s)), which was then removed from aqueous solution. Macaskie et al. [130] showed that the Gram-negative bacterium *Citrobacter* secretes large amounts of hydrogen phosphate ions via phosphatases to form minerals with heavy metals on the bacterial surface. Sondi et al. [131] successfully precipitated heavy metal ions from $SrCl_2$ and $BaCl_2$ solutions using urease to obtain $SrCO_3$ and $BaCO_3$ and investigated the effect of urease on the crystal growth process and final crystal shape during the precipitation process. Uniform nanoscale spherical particles were formed at the initial stage of the reaction, and the spherical particles were transformed into rod-like clusters of alkaline minerals in the subsequent stage of the reaction. Deng et al. [132] isolated a strain of *Penicillium chrysogenum* from soil contaminated by smelter slag and applied it to the remediation of contaminated soil, leaching out 62.8% of the Cd, 55.5% of the Cu, 53.9% of the Zn, and 14.4% of the Pb. Ren et al. [133] explored the leaching efficiency of *Aspergillus niger* for heavy metal-contaminated soils in Shenyang Smeltery and discovered that the leaching efficiency was the highest with a soil-solution ratio of 5%, reaching 25.2%, 98.3%, 2%, and 15.7% for Cu, Cd, Pb, and Zn, respectively. Banerjee et al. [134] isolated a strain of *Pseudomonas brenneri* from coal mine wastewater, which was able to remove 96.3% of 60 mg·$L^{-1}$ Cr (VI) within six days under optimized conditions (pH = 6.0, temperature: 30°C). Zheng et al. [135] verified that, in addition to effectively adsorbing Cr (VI), *Bacillus subtilis* has the ability to reduce Cr(VI) to Cr(III) under aerobic conditions, indicating that the *nfrA* gene is directly involved in the synthesis of reductase. In summary, microbial remediation technology, which is based on biomineralization, remediates heavy metal-contaminated soil through the solidification or leaching of heavy metal ions. This technology is simple and easy to operate, does not cause secondary pollution, and can effectively reduce the impact of metal ions on the environment, showing favorable application prospects.

## 6. Conclusion and Prospects

As a new research field in geotechnical engineering, microbial geotechnology is an interdisciplinary subject involving microbiology, chemistry, and geotechnical engineering. In this review article, the hydrolysis reaction and cementation mechanism of MICP were briefly described; the influences of factors such as the bacterial concentration, cementation solution concentration, pH, temperature, grouting method, and particle size on MICP-treated soil were discussed; engineering properties such as the strength, stiffness, liquefaction resistance, permeability, and durability of MICP-treated soil were evaluated; and the applications of MICP technology in the areas of soil reinforcement, geotechnical seepage control, sand liquefaction control, fixation of heavy metals, wind erosion control, and sand fixation were summarized. Hence, this review article provides a reference for the development of MICP technology in the field of geotechnical engineering. The main conclusions are as follows:

(1) MICP is a common microbial mineralization phenomenon in nature. Through metabolic reactions, MICP absorbs, transforms, removes, and degrades substances in the environment. Researchers utilized different metabolic processes to attain MICP, which include urea hydrolysis, denitrification, ferric reduction, and sulfate reduction.

(2) The solidification efficiency of MICP is affected by factors such as the bacterial concentration, cementation solution concentration, pH, temperature, grouting method, and particle size. The selection of a reasonable optimization scheme is especially important for improving the mechanical properties of solidified soil.

(3) MICP technology can significantly improve the strength, stiffness, liquefaction resistance, and durability of soil and reduce the permeability of the soil, which has notable advantages for embankment and dam seepage control projects.

(4) MICP technology is applicable to soil improvement, geotechnical seepage control, sand liquefaction control, contaminated soil remediation, wind erosion prevention, and sand fixation. This method is expected to be implemented in $CO_2$ sequestration, desert greening, marine land reclamation, concrete repair, and ancient building restoration.

(5) Currently, most of the MICP research has been conducted under laboratory conditions, while field-scale geotechnical engineering projects have been rarely conducted. The application of this technology in geotechnical engineering faces certain practical challenges, such as technology optimization, operability, cost, equipment, and environmental factors.

Based on current research results, we believe that future MICP research should focus on the following aspects:

(1) For different types of geotechnical materials and performance improvement requirements, bacteria with high enzyme-producing ability, high environmental adaptability, and high efficiency in solidification should be cultivated, taking into account the related costs

(2) The engineering properties of the rock and soil should be improved by stimulating the production, reproduction, and metabolism of native microorganisms in the soil, which will in turn promote the homogeneity of MICP-treated soil by optimizing the grouting method

(3) By means of meso/microscopic test methods, the meso/microscopic structural changes in the solidified soil caused by microbial cementation filling, ion transport, and fiber bending and interweaving should be investigated to reveal the underlying synergistic solidification mechanisms of MICP and fiber reinforcement

(4) Based on studies of the erosion resistance of MICP-treated soils with single factors (wind erosion, water erosion, salt erosion, freeze-thaw erosion, and ultraviolet erosion), the durability of MICP-treated soils considering combinations of various factors should be evaluated

## Data Availability

The data used to support the findings of this study are available from the corresponding author upon request.

## Conflicts of Interest

The authors declare that there are no conflicts of interest regarding the publication of this paper.

## Acknowledgments

This study was funded by the National Natural Science Foundation of China (41877225); Fundamental Research Funds for the Central Universities, CHD (300102261719); Natural Science Basic Research Program of Shaanxi Province (2021JM-535); and Special Fund for Scientific Research by Xijing University (XJ18T01).

## References

[1] H. L. Liu and M. H. Zhao, "Review of ground improvement technical and its application in China," *China Civil Engineering Journal*, vol. 49, no. 1, pp. 96–115, 2016.

[2] S. Safavizadeh, B. M. Montoya, and M. A. Gabr, "Microbial induced calcium carbonate precipitation in coal ash," *Géotechnique*, vol. 69, no. 8, pp. 727–740, 2019.

[3] M. Umar, K. A. Kassim, and K. T. Ping Chiet, "Biological process of soil improvement in civil engineering: a review," *Journal of Rock Mechanics and Geotechnical Engineering*, vol. 8, no. 5, pp. 767–774, 2016.

[4] S. G. Choi, I. Chang, M. Lee, J. H. Lee, J. T. Han, and T. H. Kwon, "Review on geotechnical engineering properties of sands treated by microbially induced calcium carbonate precipitation (MICP) and biopolymers," *Construction and Building Materials*, vol. 246, Article ID 118415, 2020.

[5] H. L. Liu, P. Xiao, Y. Xiao, and J. Chu, "State-of-the-art review of biogeotechnology and its engineering applications," *Journal of Civil and Environmental Engineering*, vol. 41, no. 1, pp. 1–14, 2019.

[6] C. X. Qian, A. H. Wang, and X. Wang, "Advances of soil improvement with bio-grouting," *Rock and Soil Mechanics*, vol. 36, no. 6, pp. 1537–1548, 2015.

[7] V. S. Whiffin, "Microbial $CaCO_3$ precipitation for the production of biocement," Ph. D thesis, Murdoch University, Perth, Australia, 2004.

[8] J. K. Mitchell and J. C. Santamarina, "Biological considerations in geotechnical engineering," *Journal of Geotechnical and Geoenvironmental Engineering*, vol. 131, no. 10, pp. 1222–1233, 2005.

[9] J. T. DeJong, K. Soga, E. Kavazanjian et al., "Biogeochemical processes and geotechnical applications: progress, opportunities and challenges," *Géotechnique*, vol. 63, no. 4, pp. 287–301, 2013.

[10] J. He, J. Chu, H. L. Liu, Y. F. Gao, and B. Li, "Research advances in biogeotechnologies," *Chinese Journal of Geotechnical Engineering*, vol. 38, no. 4, pp. 643–653, 2016.

[11] J. T. DeJong, B. M. Mortensen, B. C. Martinez, and D. C. Nelson, "Bio-mediated soil improvement," *Ecological Engineering*, vol. 36, no. 2, pp. 197–210, 2010.

[12] N. K. Dhami, M. S. Reddy, and A. Mukherjee, "Biomineralization of calcium carbonates and their engineered applications: a review," *Frontiers in Microbiology*, vol. 4, p. 314, 2013.

[13] Y. Z. Wang, K. Soga, J. T. Dejong, and A. J. Kabla, "Effects of bacterial density on growth rate and characteristics of microbial-induced $CaCO_3$ precipitates: a particle-scale experimental study," *ASCE Journal of Geotechnical and Geoenvironmental Engineering*, vol. 147, no. 6, 2021.

[14] S. Stocks-Fischer, J. K. Galinat, and S. S. Bang, "Microbiological precipitation of $CaCO_3$," *Soil Biology and Biochemistry*, vol. 31, no. 11, pp. 1563–1571, 1999.

[15] W. De Muynck, N. De Belie, and W. Verstraete, "Microbial carbonate precipitation in construction materials: a review," *Ecological Engineering*, vol. 36, no. 2, pp. 118–136, 2010.

[16] O. Dawoud, "The applicability of microbially induced calcite precipitation (MICP) for soil treatment," Ph. D thesis, University of Cambridge, Cambridge, UK, 2015.

[17] E. G. Lauchnor, D. M. Topp, A. E. Parker, and R. Gerlach, "Whole cell kinetics of ureolysis by *Sporosarcina pasteurii*," *Journal of Applied Microbiology*, vol. 118, pp. 1321–1332, 2015.

[18] M. Lee, M. G. Gomez, A. C. M. S. Pablo et al., "Investigating ammonium by-product removal for ureolytic bio-cementation using meter-scale experiments," *Scientific Reports*, vol. 9, Article ID 18313, 2019.

[19] Y. J. Qin and J. M. S. Cabral, "Kinetic studies of the orease-catalyzed hydrolysis of orea in a buffer-free system," *Applied Biochemistry and Biotechnology*, vol. 49, pp. 217–240, 1994.

[20] B. Krajewska, "Ureases I. Functional, catalytic and kinetic properties: a review," *Journal of Molecular Catalysis B: Enzymatic*, vol. 59, pp. 9–21, 2009.

[21] B. Krajewska, "Ureases. II. Properties and their customizing by enzyme immobilizations: a review," *Journal of Molecular Catalysis B: Enzymatic*, vol. 59, pp. 22–40, 2009.

[22] Y. Z. Wang, K. Soga, and N. J. Jiang, "Microbial induced carbonate precipitation (MICP): the case for microscale perspective," in *Proceedings of the 19th International Conference on Soil Mechanics and Geotechnical Engineering*, Korea Geotechnical Society, Seoul, Korea, September 2017.

[23] Y. Z. Wang, K. Soga, J. T. Dejong, and A. J. Kabla, "A microfluidic chip and its use in characterising the particle-scale behaviour of microbial-induced calcium carbonate precipitation (MICP)," *Géotechnique*, vol. 69, no. 12, pp. 1086–1094, 2019.

[24] Y. Z. Wang, K. Soga, J. T. Dejong, and A. J. Kabla, "Microscale visualization of microbial-induced calcium carbonate precipitation processes," *Journal of Geotechnical and Geoenvironmental Engineering*, vol. 145, no. 9, 2019.

[25] X. He, G. L. Ma, Y. Wang et al., "Visualization investigation of bio-cementation process based on microfluidics," *Chinese Journal of Geotechnical Engineering*, vol. 42, no. 6, pp. 1005–1012, 2020.

[26] M. J. Cui, J. J. Zheng, R. J. Zhang, H. J. Lai, and J. Zhang, "Influence of cementation level on the strength behavior of bio-cemented sand," *Acta Geotechnica*, vol. 12, pp. 971–986, 2017.

[27] Y. Z. Wang, C. Konstantinou, K. Soga, J. T. Dejong, G. Biscontin, and A. J. Kabla, "Enhancing strength of MICP-treated sandy soils: from micro to macro scale," 2006, http://arxiv.org/abs/2006.15760.

[28] L. Y. Yin, C. S. Tang, Y. H. Xie, C. Lv, N. J. Jiang, and B. Shi, "Factors affecting improvement in engineering properties of geomaterials by microbial-induced calcite precipitation," *Rock and Soil Mechanics*, vol. 40, no. 7, pp. 2525–2546, 2019.

[29] A. Miftah, H. K. Tirkolaei, and H. Bilsel, "Biocementation of calcareous beach sand using enzymatic calcium carbonate precipitation," *Crystals*, vol. 10, no. 10, pp. 1–15, 2020.

[30] A. Almajed, H. K. Tirkolaei, E. Kavazanjian, and N. Hamdan, "Enzyme induced biocementated sand with high strength at low carbonate content," *Scientific Reports*, vol. 9, p. 1135, 2019.

[31] K. Martin, H. K. Tirkolaei, and E. Kavazanjian, "Enhancing the strength of granular material with a modified enzyme-induced carbonate precipitation (EICP) treatment solution," *Construction and Building Materials*, vol. 271, Article ID 121529, 2021.

[32] C. Konstantinou, Y. Z. Wang, G. Biscontin, and K. Soga, "The role of bacterial urease activity on the uniformity of carbonate precipitation profles of bio-treated coarse sand specimens," *Scientific Reports*, vol. 11, p. 6161, 2021.

[33] M. H. Baziar, M. Sanaie, and O. E. Amirabadi, "Mitigation of dust emissions of silty sand induced by wind erosion using natural soybean biomaterial," *International Journal of Civil Engineering*, vol. 19, pp. 595–606, 2021.

[34] Y. B. Chen, Y. F. Gao, C. W. W. Ng, and H. W. Guo, "Bio-improved hydraulic properties of sand treated by soybean urease induced carbonate precipitation and its application part 1: water retention ability," *Transportation Geotechnics*, vol. 27, Article ID 100489, 2021.

[35] B. Yang, H. Li, H. Z. Li, N. L. Ge, G. B. Ma, and H. J. Zhang, "Experimental investigation on the mechanical and hydraulic properties of urease stabilized fine sand for fully

permeable pavement," *International Journal of Transportation Science and Technology*, 2020.

[36] Y. F. Gao, H. Meng, J. He, Y. S. Qi, and L. Hang, "Field trial on use of soybean crude extract for carbonate precipitation and wind erosion control of sandy soil," *Journal of Central South University*, vol. 27, pp. 3320–3333, 2020.

[37] M. Wu, Y. F. Gao, J. He, and Y. Liu, "Laboratory study on use of soybean urease-induced calcium carbonate precipitation with xanthan gum for stabilization of desert sand against wind erosion," *Chinese Journal of Geotechnical Engineering*, vol. 42, no. 10, pp. 1914–1921, 2020.

[38] H. Y. Xu, J. J. Lian, and Y. Yan, "Experimental study of MICP solidified sand under the coupling of multiple test factors," *Journal of Tianjin University*, vol. 53, no. 5, pp. 517–526, 2020.

[39] L. Cheng, C. X. Qian, and R. X. Wang, "Study on the mechanism of calcium carbonate formation induced by carbonate-mineralization microbe," *Acta Chimica Sinica*, vol. 65, no. 19, pp. 2133–2138, 2007.

[40] A. S. Thawadi and R. Cord-Ruwisch, "Calcium carbonate crystals formation by ureolytic bacteria isolated from australian soil and sludge," *Journal of Advanced Science & Engineering Research*, vol. 2, no. 1, pp. 12–26, 2012.

[41] W. S. Ng, M. L. Lee, C. K. Tan, and S. L. Hii, "Factors affecting improvement in engineering properties of residual soil through microbial-induced calcite precipitation," *Journal of Geotechnical and Geoenvironmental Engineering*, vol. 140, no. 5, 2014.

[42] Q. Zhao, L. Li, C. Li, M. D. Li, F. Amini, and H. Z. Zhang, "Factors affecting improvement of engineering properties of MICP-treated soil catalyzed by bacteria and urease," *Journal of Materials in Civil Engineering*, vol. 26, no. 12, 2014.

[43] M. O. Cuthbert, M. S. Riley, S. Handley-Sidhu et al., "Controls on the rate of ureolysis and the morphology of carbonate precipitated by *S. Pasteurii* biofilms and limits due to bacterial encapsulation," *Ecological Engineering*, vol. 41, pp. 32–40, 2012.

[44] V. Rebata-Landa, *Microbial Activity in Sediments: Effects on Soil Behavior*, Georgia Institute of Technology, Atlanta, GA, USA, 2007.

[45] M. Nemati, E. A. Greene, and G. Voordouw, "Permeability profile modification using bacterially formed calcium carbonate: comparison with enzymic option," *Process Biochemistry*, vol. 40, no. 2, pp. 925–933, 2005.

[46] A. A. Qabany, K. Soga, and C. Santamarina, "Factors affecting efficiency of microbially induced calcite precipitation," *Journal of Geotechnical and Geoenvironmental Engineering*, vol. 138, no. 8, pp. 992–1001, 2012.

[47] A. A. Qabany and K. Soga, "Effect of chemical treatment used in MICP on engineering properties of cemented soils," *Géotechnique*, vol. 63, no. 4, pp. 331–339, 2013.

[48] Z. M. Liu, Y. C. Sun, Q. P. Feng, and J. Peng, "Study on influence of excess urea in MICP cementation solution," *Journal of Disaster Prevention and Mitigation Engineering*, vol. 40, no. 4, pp. 574–580, 2020.

[49] G. D. O. Okwadha and J. Lin, "Optimum conditions for microbial carbonate precipitation," *Chemosphere*, vol. 81, pp. 1143–1148, 2010.

[50] C. C. Wu, J. J. Zheng, H. J. Lai, M. J. Cui, and Y. Song, "Experimental study of the strength enhancing mechanics of bio-cemented sand and its influential factors," *Journal of Civil & Environmental Engineering*, vol. 42, no. 1, pp. 31–38, 2020.

[51] H. A. Keykha, B. B. K. Huat, A. Asadi, and S. Kawasaki, "Microbial induced calcite precipitation by *Sporosarcina pasteurii* and *Sporosarcina aquimarina*," *Environmental Geotechnics*, vol. 6, no. 8, pp. 562–566, 2019.

[52] J. Henze and D. G. Randall, "Microbial induced calcium carbonate precipitation at elevated pH values (>11) using *Sporosarcina pasteurii*," *Journal of Environmental Chemical Engineering*, vol. 6, pp. 5008–5013, 2018.

[53] F. Hammes and W. Verstraete, "Key roles of pH and calcium metabolism in microbial carbonate precipitation," *Reviews in Environmental Science and Biotechnology*, vol. 1, no. 1, pp. 3–7, 2002.

[54] W. Li, W. S. Chen, P. P. Zhou, L. Cao, and L. J. Yu, "Influence of initial pH on the precipitation and crystal morphology of calcium carbonate induced by microbial carbonic anhydrase," *Colloids and Surfaces B: Biointerfaces*, vol. 102, pp. 281–287, 2013.

[55] L. Cheng, M. A. Shahin, R. Cord-Ruwisch, M. Addis, T. Hartanto, and C. Elms, "Soil stabilisation by microbial-induced calcite precipitation (MICP): investigation into some physical and environmental aspects," *International Congress on Environmental Geotechnics, Melbourne*, pp. 440–448, 2014.

[56] H. A. Keykha, A. Asadi, and M. Zareian, "Environmental factors affecting the compressive strength of microbiologically induced calcite precipitation-treated soil," *Geomicrobiology*, vol. 34, no. 10, pp. 1–6, 2017.

[57] F. G. Ferris, V. Phoenix, Y. Fujita, and R. W. Smith, "Kinetics of calcite precipitation induced by ureolytic bacteria at 10 to 20°C in artificial groundwater," *Geochimica Et Cosmochimica Acta*, vol. 68, no. 8, pp. 1701–1710, 2004.

[58] W. S. Ng, M. L. Lee, and S. L. Hil, "An overview of the factors affecting microbial-induced calcite precipitation and its potential application in soil improvement," *World Academy of Science*, vol. 62, pp. 723–729, 2012.

[59] X. J. Liu, J. Y. Fan, J. Yu, and X. Gao, "Solidification of loess using microbial induced carbonate precipitation," *Journal of Mountain Science*, vol. 18, no. 1, pp. 265–274, 2021.

[60] W. D. Muynck, K. Verbeken, and N. D. Belie, "Influence of temperature on the effectiveness of a biogenic carbonate surface treatment for limestone conservation," *Applied Microbiology and Biotechnology*, vol. 97, no. 3, pp. 1335–1347, 2013.

[61] M. Nemati and G. Voordouw, "Modification of porous media permeability, using calcium carbonate produced enzymatically in situ," *Enzyme and Microbial Technology*, vol. 33, no. 5, pp. 635–642, 2003.

[62] E. Gillman, M. A. Morgan, and M. Sherwood, "Urease activity in Irish soils at 6°C," *Biology and Environment: Proceedings of the Royal Irish Academy*, vol. 95B, no. 1, pp. 19–26, 1995.

[63] D. Kralj, L. Brecevic, and A. E. Nielsen, "Vaterite growth and dissolution in aqueous solution II. Kinetics of dissolution," *Journal of Crystal Growth*, vol. 143, no. 3, pp. 269–276, 1994.

[64] S. C. Bang, S. H. Min, and S. S. Bang, "Application of microbiologically induced soil stabilization technique for dust suppression," *International Journal of Geo-Engineering*, vol. 3, no. 2, pp. 27–37, 2011.

[65] J. Peng, X. He, Z. M. Liu, Q. P. Feng, and J. He, "Experimental research on influence of low temperature on MICP-treated soil," *Chinese Journal of Geotechnical Engineering*, vol. 38, no. 10, pp. 1769–1774, 2015.

[66] V. S. Whiffin, L. A. Van Paassen, and P. Marien, "Microbial carbonate precipitation as a soil improvement technique," *Geomicrobiology Journal*, vol. 24, no. 5, pp. 417–423, 2007.
[67] D. J. Tobler, E. Maclachlan, and V. R. Phoenix, "Microbially mediated plugging of porous media and the impact of differing injection strategies," *Ecological Engineering*, vol. 42, no. 5, pp. 270–278, 2012.
[68] S. H. Liang, J. Dai, X. Li, J. G. Niu, Y. D. Huang, and Y. M. Yin, "Study of effects of different curing methods on the strength of bio-cemented sand soil," *Industrial Construction*, vol. 47, no. 2, pp. 82–86, 2017.
[69] C. Y. Xu, H. B. Wang, and C. T. Bai, "Experimental study of improving carbonate deposition distribution in silt by EBM grouting," *Industrial Construction*, vol. 47, no. 4, pp. 80–85, 2017.
[70] Q. Zhan and C. X. Qian, "Stabilization of sand particles by biocement based on $CO_2$ capture and utilization: process, mechanical properties and microstructure," *Construction and Building Materials*, vol. 133, pp. 73–80, 2017.
[71] Q. Andres, S. Zhang, and X. H. Cheng, "Study of the MICP injection in unsaturated sandy soils," *Industrial Construction*, vol. 45, no. 7, pp. 28–30, 2015.
[72] M. J. Cui, J. J. Zheng, and H. J. Lai, "Effect of method of biological injection on dynamic behavior for bio-cemented sand," *Rock and Soil Mechanics*, vol. 38, no. 11, pp. 3173–3178, 2017.
[73] M. J. Cui, J. J. Zheng, and H. J. Lai, "Experimental study of effect of particle size on strength of bio-cemented sand," *Rock and Soil Mechanics*, vol. 37, no. S2, pp. 397–402, 2016.
[74] G. G. N. N. Amarakoon and S. Kawasaki, "Factors affecting the improvement of sand properties treated with microbially-induced calcite precipitation," in *Congress Geo-Chicago: Sustainability, Energy and the Geoenvironment*, Geo-Chicago, Chicago, IL, USA, August 2016.
[75] X. Li, S. J. Wang, B. H. He, and T. Y. Shen, "Permeability condition of soil suitable for MICP method," *Rock and Soil Mechanics*, vol. 40, no. 8, pp. 2956–2964, 2019.
[76] S. Animesh and R. Ramkrishnan, "Study on effect of microbial induced calcite precipitates on strength of fine grained soils," *Perspectives in Science*, vol. 8, pp. 198–202, 2016.
[77] L. A. Van Paassen, C. M. Daza, M. Staal, D. Y. Sorokin, W. Van der Zon, and M. C. M. Van Loosdrecht, "Potential soil reinforcement by biological denitrification," *Ecological Engineering*, vol. 36, pp. 168–175, 2010.
[78] S. Gowthaman, S. Mitsuyama, K. Nakashima, M. Komatsu, and S. Kawasaki, "Biogeotechnical approach for slope soil stabilization using locally isolated bacteria and inexpensive low-grade chemicals: a feasibility study on Hokkaido expressway soil, Japan," *Soils and Foundations*, vol. 59, pp. 484–499, 2019.
[79] H. Rong and C. X. Qian, "Cementation of loose sand particles based on bio-cement," *Journal of Wuhan University of Technology*, vol. 29, no. 4, pp. 1208–1212, 2014.
[80] Z. M. Liu, J. Peng, J. Li, and E. R. Song, "Experimental study on improving effect of microorganism solidifying sand by ultrasonic," *Journal of Zhejiang University*, vol. 54, no. 7, pp. 1–7, 2020.
[81] S. G. Choi, K. J. Wang, and J. Chu, "Properties of biocemented, fiber reinforced sand," *Construction and Building Materials*, vol. 120, pp. 623–629, 2016.
[82] X. W. Fang, Y. Yang, Z. Chen, H. L. Liu, Y. Xiao, and C. N. Shen, "Influence of fiber content and length on engineering properties of MICP-treated coral sand," *Geomicrobiology Journal*, vol. 37, no. 6, pp. 582–594, 2020.
[83] Y. H. Xie, C. S. Tang, L. Y. Yin, C. Lv, N. J. Jiang, and B. Shi, "Mechanical behavior of microbial-induced calcite precipitation (MICP)-treated soil with fiber reinforcement," *Chinese Journal of Geotechnical Engineering*, vol. 41, no. 4, pp. 675–682, 2019.
[84] J. J. Zheng, Y. Song, H. J. Lai, M. J. Cui, and C. C. Wu, "Experimental study on the shear behavior of fiber-reinforced bio-cemented sand," *Journal of Civil and Environmental Engineering*, vol. 41, no. 1, pp. 15–21, 2019.
[85] J. T. DeJong, M. B. Fritzges, and K. Nusslein, "Microbially induced cementation to control sand response to undrained shear," *Journal of Geotechnical and Geoenvironmental Engineering*, vol. 132, no. 11, pp. 1381–1392, 2006.
[86] J. T. DeJong, K. Soga, S. A. Banwart et al., "Soil engineering in vivo: harnessing natural biogeochemical systems for sustainable, multi-functional engineering solutions," *Journal of The Royal Society Interface*, vol. 8, no. 54, pp. 1–15, 2011.
[87] L. A. Van Paassen, M. P. Harkes, G. A. Van Zwieten, W. H. Van Der Zon, W. R. L. Van Der Star, and M. C. M. Van Loosdrecht, "Scale up of biogrout: a biological ground reinforcement method," in *Proceedings of the 17th International Conference on Soil Mechanics and Geotechnical Engineering*, pp. 2328–2333, Lansdale IOS Press, Alexandria, Egypt, October 2009.
[88] L. A. Van Paassen, R. Ghose, T. J. M. Van Der Linden, W. R. L. Van Der Star, and M. C. M. Loosdrecht, "Quantifying biomediated ground improvement by ureolysis: large-scale bio-grout experiment," *Journal of Geotechnical and Geoenvironmental Engineering*, vol. 136, no. 12, pp. 1721–1728, 2010.
[89] P. Liu, G. H. Shao, and R. P. Huang, "Mechanical Properties and constitutive model of MICP-cemented sand," *Journal of Southeast University (Natural Science Edition)*, vol. 49, no. 4, pp. 720–726, 2019.
[90] H. L. Liu, P. Xiao, Y. Xiao, J. P. Wang, Y. M. Chen, and J. Chu, "Dynamic behaviors of MICP-treated calcareous sand in cyclic tests," *Chinese Journal of Geotechnical Engineering*, vol. 40, no. 1, pp. 38–45, 2018.
[91] X. L. Zhang, Y. M. Chen, Z. Zhang et al., "Performance evaluation of liquefaction resistance of a MICP-treated calcareous sandy foundation using shake table tests," *Chinese Journal of Geotechnical Engineering*, vol. 42, no. 6, pp. 1023–1031, 2020.
[92] A. Zamani and B. M. Montoya, "Undrained cyclic response of silty sands improved by microbial induced calcium carbonate precipitation," *Soil Dynamics and Earthquake Engineering*, vol. 120, pp. 436–448, 2019.
[93] P. Xiao, H. L. Liu, Y. Xiao, A. W. Stuedlein, and T. M. Evans, "Liquefaction resistance of bio-cemented calcareous sand," *Soil Dynamics and Earthquake Engineering*, vol. 107, pp. 9–19, 2018.
[94] Z. G. Han, X. H. Cheng, and Q. Ma, "An experimental study on dynamic response for MICP strengthening liquefiable sands," *Earthquake Engineering and Engineering Vibration*, vol. 15, no. 4, pp. 673–679, 2016.
[95] Z. G. Han and X. H. Cheng, "Nutritive salt's impact on microorganism strengthening liquefiable sandy soil," *Industrial Construction*, vol. 45, no. 7, pp. 19–22, 2015.
[96] X. H. Cheng, Q. Ma, Z. Yang, Z. C. Zhang, and M. Li, "Dynamic response of liquefiable sand foundation improved by bio-grouting," *Chinese Journal of Geotechnical Engineering*, vol. 35, no. 8, pp. 1486–1495, 2013.

[97] Y. M. Lai, J. Yu, S. Y. Liu, J. F. Liu, R. K. Wang, and B. Dong, "Experimental study to improve the mechanical properties of iron tailings sand by using MICP at low pH," *Construction and Building Materials*, vol. 273, Article ID 121729, 2021.

[98] J. Chu, V. Ivanov, V. Stabnikov, and B. Li, "Microbial method for construction of an aquaculture pond in sand," *Géotechnique*, vol. 63, no. 10, pp. 871–875, 2013.

[99] T. Y. Shen, S. J. Wang, L. Xue, X. Li, and B. H. He, "An experimental study of sandy clayey purple soil enhanced through microbial-induced calcite precipitation," *Rock and Soil Mechanics*, vol. 40, no. 8, pp. 3115–3124, 2019.

[100] F. Jawad and J. J. Zheng, "Improving poorly graded fine sand with microbial induced calcite precipitation," *British Journal of Applied Science & Technology*, vol. 17, no. 2, pp. 1–9, 2016.

[101] N. J. Jiang, C. S. Tang, T. Hata, B. Courcelles, O. Dawoud, and D. N. Singh, "Bio-mediated soil improvement: the way forward," *Soil Use and Management*, vol. 36, no. 2, pp. 185–188, 2020.

[102] O. Dawoud, C. Y. Chen, and K. Soga, "Microbial induced calcite precipitation for geotechnical and environmental applications," in *Proceedings of GeoShanghai International Congress: New Frontiers in Geotechnical Engineering*, pp. 11–18, Geo-Shanghai 2014, Shanghai, China, May 2014.

[103] O. Dawoud, C. Y. Chen, and K. Soga, "Microbial induced calcite precipitation (MICP) using surfactants," in *Proceedings of the GeoCongress: Geo-Characterization and Modeling for Sustainabliliy*, pp. 1635–1643, Geo-Congress 2014, Atlanta, Georgia, February 2014.

[104] L. Cheng, T. Kobayashi, and M. A. Shahin, "Microbially induced calcite precipitation for production of "bio-bricks" treated at partial saturation condition," *Construction and Building Materials*, vol. 231, Article ID 117095, 2020.

[105] S. Y. Liu, R. K. Wang, J. Yu, X. Q. Peng, Y. Y. Cai, and B. X. Tu, "Effectiveness of the anti-erosion of an MICP coating on the surfaces of ancient clay roof tiles," *Construction and Building Materials*, vol. 243, Article ID 118202, 2020.

[106] S. H. Chae, H. Chung, and K. Nam, "Evaluation of microbially induced calcite precipitation (MICP) methods on different soil types for wind erosion control," *Environmental Engineering Science*, vol. 26, no. 1, 2021.

[107] F. Nikseresht, A. Landi, G. Sayyad, G. Ghezelbash, and R. Schulin, "Sugarecane molasse and vinasse added as microbial growth substrates increase calcium carbonate content, surface stability and resistance against wind erosion of desert soils," *Journal of Environmental Management*, vol. 268, Article ID 110639, 2020.

[108] M. Huang, J. X. Zhang, G. X. Jin et al., "Magnetic resonance image experiments on the damage feature of microbial induced calcite precipitated residual soil during freezing-thawing cycles," *Chinese Journal of Rock Mechanics and Engineering*, vol. 37, no. 12, pp. 2846–2855, 2018.

[109] M. G. Gomez, B. C. Martinez, J. T. DeJong et al., "Field-scale bio-cementation tests to improve sands," *Proceedings of the Institution of Civil Engineers-Ground Improvement*, vol. 168, no. 3, pp. 206–216, 2015.

[110] N. J. Jiang, K. Soga, and O. Dawoud, "Experimental study of mitigation of soil internal erosion by microbially induced calcite precipitation," in *Proceedings of the Geo-Congress: Geo-Characterization and Modeling for Sustainability*, pp. 1586–1595, ASCE, Atlanta, GA, USA, February 2014.

[111] L. Cheng, R. Cord-Ruwisch, and M. A. Shahin, "Cementation of sand soil by microbially induced calcite precipitation at various degrees of saturation," *Canadian Geotechnical Journal*, vol. 50, no. 1, pp. 81–90, 2013.

[112] S. H. Liu, K. J. Wen, C. Armwood et al., "Enhancement of MICP-treated sandy soils against environmental deterioration," *Journal of Materials in Civil Engineering*, vol. 31, no. 12, 2019.

[113] L. A. Van Paassen, *Bio-Mediated Ground Improvement: From Laboratory Experiment to Pilot Applications*, Geo-Frontiers Congress, Dallas, TX, USA, 2011.

[114] H. L. Liu, G. L. Ma, Y. Xiao, X. M. Ding, and X. W. Fang, "In situ experimental research on calcareous foundation stabilization using MICP technique on the reclaimed coral reef islands," *Chinese Ground Improvement*, vol. 1, no. 1, pp. 26–31, 2019.

[115] B. M. Montoya, J. T. DeJong, and R. W. Boulanger, "Dynamic response of liquefiable sand improved by microbial-induced calcite precipitation," *Géotechnique*, vol. 63, no. 4, pp. 302–312, 2013.

[116] K. M. Darby, G. L. Hernandez, J. T. DeJong, R. W. Boulanger, M. G. Gomez, and D. W. Wilson, "Centrifuge model testing of liquefaction mitigation via microbially induced calcite precipitation," *Journal of Geotechnical and Geoenvironmental Engineering*, vol. 145, no. 10, 2019.

[117] J. He, J. Chu, and V. Ivanov, "Mitigation of liquefaction of saturated sand using biogas," *Géotechnique*, vol. 63, no. 4, pp. 267–275, 2013.

[118] Y. F. Tan, Z. J. Guo, H. J. Chen, and Z. Chen, "Study on application of microbial tracing consolidation technology in the seepage prevention of earth bank," *Journal of Hohai University (Natural Science)*, vol. 46, no. 6, pp. 521–526, 2018.

[119] L. Liu, Y. Shen, H. L. Liu, and J. Chu, "Application of biocement in erosion control of levees," *Rock and Soil Mechanics*, vol. 37, no. 12, pp. 3410–3416, 2016.

[120] Y. F. Gao, X. Y. Tang, J. Chu, and J. He, "Microbially induced calcite precipitation for seepage control in sandy soil," *Geomicrobiology Journal*, vol. 10, pp. 1–10, 2019.

[121] L. Cheng, Y. Yang, and J. Chu, "In-situ microbially induced $Ca^{2+}$ alginate polymeric sealant for seepage control in porous materials," *Microbial Biotechnology*, vol. 12, no. 2, pp. 324–333, 2019.

[122] M. Blauw, J. W. M. Lambert, and M. N. Latil, "Biosealing: a method for in situ sealing of leakages," in *Proceedings of the International Symposium on Ground Improvement Technologies & Case Histories*, 2011.

[123] M. Naeimi and J. Chu, "Comparison of conventional and bio-treated methods as dust suppressants," *Environmental Science and Pollution Research*, vol. 24, no. 29, pp. 23341–23350, 2017.

[124] Q. W. Zhan, C. X. Qian, and H. H. Yi, "Microbial-induced mineralization and cementation of fugitive dust and engineering application," *Construction and Building Materials*, vol. 121, pp. 437–444, 2016.

[125] S. H. Li, C. Li, D. Yao, and S. Wang, "Feasibility of microbially induced carbonate precipitation and straw checkerboard barriers on desertification control and ecological restoration," *Ecological Engineering*, vol. 152, Article ID 105883, 2020.

[126] Y. Fujita, J. L. Taylor, L. M. Wendt, D. W. Reed, and R. W. Smith, "Evaluating the potential of native ureolytic microbes to remediate a 90Sr contaminated environment," *Environmental Science and Technology*, vol. 44, no. 19, pp. 7652–7658, 2010.

[127] Y. B. Xu, C. X. Qian, and Z. W. Lu, "Remediation of heavy metal contaminated soils by bacteria biomineralization,"

*Chinese Journal of Environmental Engineering*, vol. 7, no. 7, pp. 2763–2768, 2013.

[128] L. Cheng, C. X. Qian, R. X. Wang, and J. Y. Wang, "Bioremediation process of $Cd^{2+}$ removal soil by bacteria a biomineralization," *Journal of the Chinese Ceramic Society*, vol. 38, no. S1, pp. 215–221, 2008.

[129] R. Ganesh, K. G. Robinson, G. D. Reed, and G. S. Sayler, "Reduction of hexavalent uranium from organic complexes by sulfate-and iron-reducing bacteria," *Applied and Environmental Microbiology*, vol. 63, no. 11, pp. 4385–4391, 1997.

[130] L. E. Macaskie, A. C. R. Dean, A. K. Cheethan, R. J. B. Jakeman, and A. J. Skarnulis, "Cadmium accumulation by a Citrobacter sp.: the chemical nature of the accumulated metal precipitate and its location on the bacterial cells," *Journal of Medical Microbiology*, vol. 133, no. 3, pp. 539–544, 1987.

[131] I. Sondi and E. Matidjevic, "Homogeneous precipitation by enzyme-catalyzed reactions-strontium and barium carbonates," *Chemistry of Materials*, vol. 15, no. 6, pp. 1322–1326, 2003.

[132] X. H. Deng, L. Y. Chai, Z. H. Yang, Y. Shi, H. X. Tong, and Z. X. Wang, "Preliminary bioleaching of heavy metals from contaminated soil employing indigenous *Penicillium chrysogenum* strain," *Journal of Central South University*, vol. 19, no. 7, pp. 1973–1979, 2012.

[133] W. X. Ren, P. J. Li, and X. J. Li, "Removal of heavy metals from contaminated soils using organic acids produced by *Aspergillus niger*," *China Environmental Science*, vol. 28, no. 8, pp. 736–741, 2008.

[134] S. Banerjee, B. Kamila, S. Barman, S. R. Joshi, T. Mandal, and G. Halder, "Interlining Cr (VI) remediation mechanism by a novel bacterium *Pseudomonas brenneri* isolated from coalmine wastewater," *Journal of Environmental Management*, vol. 233, pp. 271–282, 2019.

[135] Z. Zheng, Y. Li, X. Zhang et al., "A *Bacillus subtilis* strain can reduce hexavalent chromium to trivalent and an nfrA gene is involved," *International Biodeterioration & Biodegradation*, vol. 97, pp. 90–96, 2015.