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Improving Poorly Graded Fine Sand with Microbial **Induced Calcite Precipitation**

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Authors' contributions

This work was carried out in collaboration between both authors. Both authors read and approved the final manuscript.

Article Information

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Original Research Article

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ABSTRACT

Because of the rapid growth of cities in the present days, it has become the establishment of infrastructure in the lands previously was considered inappropriate an order cannot be avoided. Therefore, to improve the engineering properties of these lands, various techniques have been applied, such as vibroflotation, dynamic compaction, and composite foundations. Recently, an innovative and sustainable technique called Microbial Induced Calcite Precipitation (MICP) has emerged for soil improvement. This study aims to explore the effectiveness of the MICP technique for improving the engineering properties of the poor graded fine sandy soil. The influence of factors such as grain size distribution and initial water content of untreated sand on the effectiveness of the MICP technique was investigated.

A set of laboratory tests were conducted, including optical density (OD600), calcium carbonate content, unconfined compressive strength, and soil permeability. The results indicate that MICP is effective for this type of sand. The results also demonstrate that use of MICP is more effective for sand with initial water content of 0 (i.e., dry) with respect to increasing the strength, while the MICP is slightly better for sand with initial water content of 100 (i.e., saturated) for the purpose of decreasing the permeability.

Keywords: Microbial-induced calcite precipitation (MICP); soil improvement; degree of saturation; permeability; UCS test; Sporosarcina pasteurii.

1. INTRODUCTION

The idea of simulating nature to improve loose soil is encouraging and motivating to all researchers. Recently, a technique called Microbial Induced Calcite Precipitation (MICP) has emerged as an innovative, cost effective, and sustainable method for soil improvement.

MICP is commonly carried out by injecting, flushing or percolating chemical reagents (e.g., urea and calcium chloride (CaCl₂) and ureolytic bacteria (e.g., *Sporosarcina pasteurii*) to the location where soil treatment is required. Ureolytic bacteria catalyze the hydrolysis of urea to produce ammonium and carbonate ions, which interact with calcium ions (e.g., calcium chloride and calcium acetate) to form calcite that precipitates throughout the soil matrix [1,2].

$$(NH_2)_2CO + 3H_2O \rightarrow 2NH_4^+ + HCO_3^- + OH^-$$

 $CaCl_2 + HCO_3^- + OH^- \rightarrow CaCO_3 \downarrow + H_2O + 2CI^-$

During the last two decades, many researches were conducted using the MICP method for the objective of improving the mechanical and engineering properties of granular soil, where it gave promising results with regard to cohesion, internal friction angle, stiffness, strength and permeability [3,4].

Previous studies targeted diverse engineering properties of soil. Dennis and Turner [5], Nemati et al. [6], DeJong et al. [7] and Heinze et al. [8] investigated the reduction of soil hydraulic conductivity, whereas Whiffin et al. [9], van Paassen [10], and Rong et al. [11] studied the improvement in soil stiffness.

A number of factors must be considered to enable the use and control of the MICP process in field applications, including the concentrations of bacteria solution, the concentrations of the chemical solutions, in addition to methods to introduce the bacteria and these chemical solutions to the soil. For example, clogging at locations adjacent to the chemical solutions injection points needs to be prevented, especially at low injection rates; this is considered a major problem in terms of utilizing the process in different setups.

This study investigated the suitability of the MICP technique to improve the engineering properties

of poorly graded (one particle size only) fine sand. The study also investigated the effects of the initial water content, curing conditions, OD600, and calcium carbonate content on the MICP efficiency.

2. MATERIALS AND METHODS

2.1 Preparation of Soil Specimens

Pure silica sand grinded to particle size of 0.1 mm was selected for the current study (Fig. 1). The poorly graded sand column was prepared by packing the dry sand (with a unit weight of 16 kN/m³, porosity of 37.67%, and pore void volume of about 33 ml) into a polyvinyl chloride (PVC) column of 80 mm high and 37 mm inner diameter. The coefficient of permeability of the untreated silica sand was approximately 1.9 x 10⁻⁵ m/s.

2.2 Bacterial Culture and Cementation Solution

The ureolytic bacterium used in the current study was *Sporosarcina pasteurii* (ATCC 11859). The ATCC 11859 was cultivated under sterile aerobic batch conditions in a yeast extract medium (20 g/l yeast extract, 10 g/l ammonium sulfate, 0.13 M Tris buffer, pH = 9). The optical density OD600 of the bacterial culture varied between 1.8 and 1.9. As described in [1,10,12,13,14] the biocementation was conducted using highly concentrated cementation solution consisting of equal moles of anhydrous calcium chloride (1 M, 111 g/l) and urea (1 M, 60 g/l).

2.3 MICP Procedure

Microbially induced carbonate precipitation for soil treatment was conducted using gravity-induced downward precipitation at a flow rate of 0.150 l/h. Initially, the sand columns were divided into two groups. The first group was dry sand columns, and the second group was saturated sand columns. The two groups were flushed with 33 ml bacterial culture, followed by 3 hours of retention time and then repeated flushes with a 33 ml cementation solution. The MICP reaction time was 24 h with highly concentrated cementation solution. The flushing with 33 ml cementation solution was repeated every 24 h. The whole tests were performed at a temperature of $30\% \pm 2\%$. After 7 days of

reaction, the soil specimens were put in the oven at a temperature of 60°C. After 7 days, half of the soil samples were flushed with 1 L of tap water and then submerged in a basin containing tap water for 24 h, after the curing stopped.

2.4 CaCO₃ Content

To determine the precipitated CaCO₃ in the soil specimens, specimens were crushed, oven-dried and recorded the weights (M_{dry}). The dry soil was soaked in HCl solution (1 M) to dissolve precipitated CaCO₃, then washed with water and drained, and finally oven-dried. The weights were recorded again (M_{residual}). The difference between the weights before and after this process is considered to be the weight of the CaCO₃ precipitated in the specimen [15].

M_{calcite}= M_{dry} - M_{residual}

2.5 Permeability, Unconfined Compressive Strength and Calcium Carbonate Content

The permeability tests were conducted using the falling head method according to ASTM D5856 – 15 before and after the MICP treatment. These tests determined the reduction in permeability and the relation with the amount of forming CaCO₃ crystals (i.e., gram/gram calcite per sand).

Also, the unconfined compression test (UCT) was conducted at a constant loading rate of 1.5%/min in accordance with ASTM D2166/D2166M-13 (ASTM 2013). These tests were conducted to establish the relationship between the strength of the soil samples and its CaCO₃ content and crystal formation. Before the UCT test, half of the soil samples were washed with 1 L of tap water and then submerged in basin contain tap water for 24 h, followed by an air dried process at 30℃ for 24 h.

2.6 Microscopy Investigation

The scanning electron microscope (SEM) helps to study the calcite bonds and their distribution within treated sand. Additionally, the SEM provides a good comprehension of the bonding between the calcite crystals and the sand particles. An FEI Quanta 200 ESEM/VPSEM scanning electron microscope was used to conduct this test. The samples were coated with gold and carbon using a BAL-TEC / SCD 050 sputter coater. This test was conducted in the

Central Laboratory at Huazhong University of Science and Technology.

3. RESULTS AND DISCUSSION

3.1 Effect of Using Poorly Graded Fine Sand

For the purpose of examining the effectiveness of the MICP technique to improve the engineering properties of poorly graded fine sand (only the sand remaining on the sieve of 0.1 mm) and to verify the feasibility of using this technique for this type of soil. The experiment was conducted using crunched silica sand. After conducting sieve analysis for crunched silica sand, the sand particles remained on the 0.1 mm sieve were collected and used to prepare the sand column specimens.

After preparing the samples by packing the sand into a PVC pipe and bacteria and cementation solution for seven days, and after the completion of curing stage, different tests were conducted. Figs. 2 - 4 show the results of those tests. The test results show that the MICP technique effectively improved the engineering properties of the poorly graded fine sands. The OD600 test for the effluent showed that the soil was able to retain a high concentration of bacteria cells, approximately 88.33% of initial concentration of bacteria cells, which is consistent with the content of precipitated calcium carbonate.

The test for the calcium carbonate content showed that the ratio of the weight of calcium carbonate to that of the sand in the samples was approximately 0.29 on average. The high concentration of precipitated calcium carbonate showed significant effect on the test results of unconfined compressive strength and permeability. This agree with previous report by El-Nahhal et al. [16]. The unconfined compressive strength reached up to 3000 kPa, while the reduction of permeability reached up to 90%. All these results showed high efficiency of the MICP method to improve the engineering properties of the poorly graded fine sands.

3.2 Initial Degree of Saturation S

Figs. 2 and 3 show a comparison of measured strength between saturated and dry treated soils. It can be seen that the initial water content has a significant impact on the effectiveness of biocementation. For similar bacteria concentration

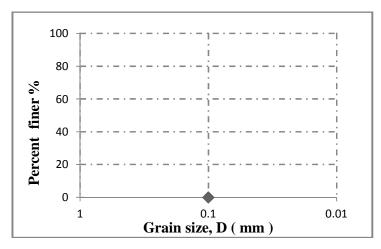


Fig. 1. Particle size distribution of the silica sand

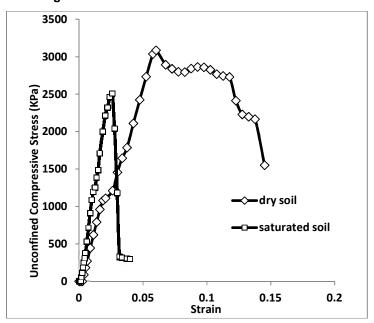


Fig. 2. Stress-strain curves of treated sand with different initial water content

(OD600 = 1.8) and similar retention time, the dry sand samples (i.e., S=0) had greater unconfined compressive strength than those of the saturated sand samples (i.e., S=100). This occurred because the sand particles in the dry sand columns had the ability to absorb more bacteria cells than those in the saturated sand columns. This explanation was based on the values of OD600 (= 0.210 and 0.528 for dry and saturated sand columns, respectively) of the first effluent.

The same phenomenon was illustrated when testing soil samples in a state of saturation (i.e., unconfined compressive strength was conducted

on the soil samples flushed with water as a last step of curing and before the UCT test), as shown previously by Heinze et al. [8]. However, the unconfined compressive strength for samples in dry test was much higher than that in saturated test. This might be because of the additional cementation effect produced by the calcium salt. The additional strength due to calcium salt may disappear after the samples become wet again, and thus this effect needs to be further studied in the future.

Fig. 2 also shows that the dry soil samples had higher failure strain than saturated soil samples. The value of the strain corresponding to the

highest unconfined compressive strength was 0.060 and 0.026 for dry and saturated sand columns, respectively, which means that the strain of dry soil samples was three times that of saturated soil samples.

Even when the comparison was made between the strain in both cases (dry and saturated sand samples) at the same value of unconfined compressive strength, it is found that the strain for dry soil condition was always greater. For example, for unconfined compressive strength of 2400 kPa, the strain was 0.047 and 0.023 for dry and saturated sand columns, respectively.

3.3 Coefficient of Permeability

The coefficient of permeability of the cured specimens (still contained in molds) was determined using the falling-head permeability test. Yasuhara et al. [17] reported that more

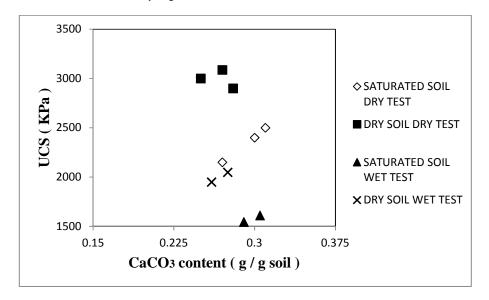


Fig. 3. Relationship between strength and calcite content for dry and saturated samples

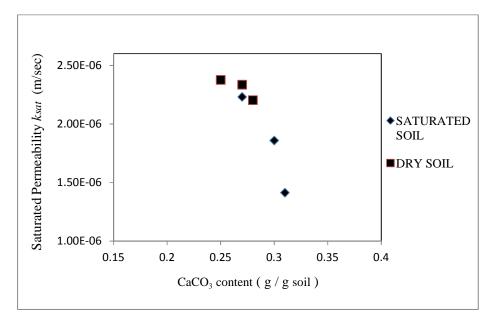


Fig. 4. Relationship between permeability and calcite content for dry and saturated samples

treatment cycles using high concentration solution of urea and CaCl2 produced greater reduction in coefficient of permeability of sand. Al Qabany and Soga [18] reported that, for four injection cycles, high concentration cementation solution (i.e., range from 0.1 to 1.0 M) produced a quicker and greater reduction in coefficient of permeability. The formation of calcium carbonate precipitation near the particle-particle contacts reduces the pore throats and restricts water flow. Ferris et al. [19] noticed a reduction of 15% to 20%in permeability, while Whiffin et al. [9] noticed a reduction from 22% to 75% in permeability. However, it is imaginable that the permeability could be reduced further with more treatment.

In addition, Fig. 4 shows a comparison of measured permeability between saturated and dry treated soils. It can be seen that the initial water content has a significant impact on the effectiveness of bio-clogging. In general, saturated sand samples showed higher calcium carbonate content than the dry soil samples, which is reflected on the permeability values, where the decrease in the permeability of saturated sand samples was slightly greater than that of dry samples.

After all, in both cases (saturated and dry samples) the use of MICP technique has significant effect on permeability, where the average decreases in the permeability of the dry

and saturated sand samples were approximately 87% and 90%, respectively, after seven treatment cycles.

3.4 Optical Density and Cell Concentration of Bacteria

The OD600 value is an indicator and measure of bacterial content and soil matrix's ability to retain bacteria. Fig. 5 shows the comparison of OD600 for the effluent No. 1, 4, and 7 (i.e., the effluent of first, fourth, and seventh day) for both dry and saturated sand samples, which have been treated with a solution of bacteria having initial OD600 of 1.8. This Figure illustrates that OD600 for the first effluent was 0.528 and 0.210 for saturated and dry sand samples, respectively, and this means that the dry soil was able to retain more bacteria cells. In spite of that, the initial OD600 was 1.8 for both types of samples, but the dry sand was able to retain 1.59, while the saturated soil retained just 1.27. On the other hand, the OD600 of the effluent of the fourth and seventh day was very low in general, despite that the dry sand showed higher values than saturated sand, which is because the dry sand retained more bacteria at the first day and as a result was able to lose more bacteria in the following days.

Lastly, Fig. 6 shows the initial bacteria cell concentration before treatment the sand (bacteria solution) and bacteria cell concentration

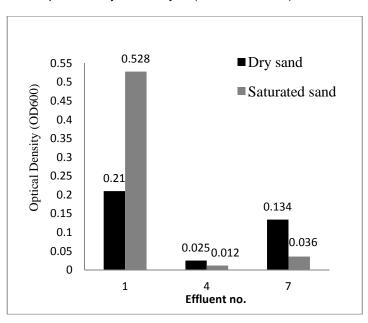


Fig. 5. Relationship between OD600 and effluent number for dry and saturated samples

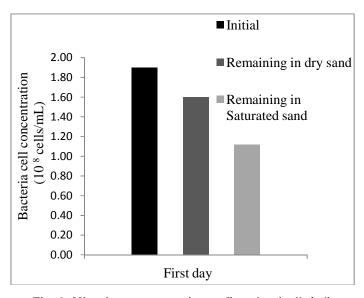


Fig. 6. Microbe concentration at first day (cells/ml)

retain inside the dry and saturated sand after first chemical solution treatment. Following Ramachandran et al. [20], bacteria cell concentration can be calculated as follows:

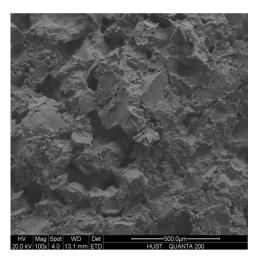
$$Y = 8.59 \times 10^7 \times (OD600)^{1.3627}$$
 (1)

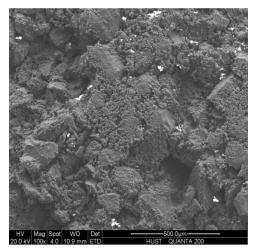
Where, Y is the bacteria cell concentration (= 1.9×10^8 , 1.6×10^8 , 1.12×10^8 cells/ml) of initial bacteria solution, retained bacteria in the dry and saturated sand, respectively.

3.5 Scanning Electronic Microscopy (SEM)

To investigate the impact of particle size and initial degree of saturation on the crystal

formation of treated samples, SEM analysis was conducted and the results are shown in Fig. 7. The CaCO3 measurements and SEM images indicate that the amount of CaCO₃ precipitated in saturated samples was slightly higher than that in dry samples. It was also indicated that the crystals produced in saturate samples were relatively small in size (about 5-10 µm in diameter) and almost fully covered the surface of the sand grains (see Fig. 7, right column, where the crystals could not provide strength development like the dry case. On the other hand, the samples treated at dry condition produced smaller amount of crystals but larger size (15-20 um, see Fig. 5, left column), which can close the gap between the adjacent sand grains and thus promote strength development.





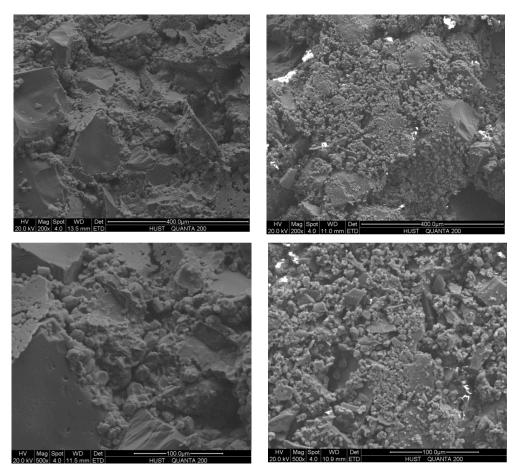


Fig. 7. SEM of CaCO₃ crystals formed at different initial degree of saturation of dry sand (left) and saturated sand (right)

4. CONCLUSIONS

Microbial induced calcite precipitation (MICP) is a very complex process, particularly when it occurs between sand particles for improving mechanical and engineering properties of soil. Many factors may affect this process. This study investigated several factors, including particle size of sand, initial degree of saturation, reaction time, and curing conditions.

The results presented in this study revealed that using the MICP technique can improve the engineering properties of poorly graded fine sand. However, the efficiency of MICP in improving the soil properties varied significantly depending on the conditions. The dry sand samples (i.e., initially dry sand before adding the bacteria solution) were found to yield greater unconfined compressive strength relative to the saturated sand samples (i.e., initially saturated sand before adding the bacteria solution). Moreover, the dry sand samples yielded higher failure strain

relative to the saturated sand samples. The results also showed that the decrease of permeability of dry sand was relatively smaller than that of saturated sand. Also, the curing condition had significant impact on the compressive strength, where the treated sand samples that soaked in water during the last step of curing showed greater decrease of compressive strength, as compared to the treated sand samples with curing that does not include soaking in water.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

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