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## **Evaluating the Application of Microbial Induced Calcite Precipitation Technique to Stabilize Expansive Soils**

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# Evaluating the Application of Microbial Induced Calcite Precipitation Technique to Stabilize Expansive Soils

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## Abstract

Expansive soils, also known as swell-shrink soils have been a problem for civil infrastructures including roads and foundations from ancient times. The use of chemical additives such as cement and lime to stabilize expansive soils is a common practice among geotechnical engineers, especially for lightly loaded structures. However, several occurrences of subgrade failures have been observed after stabilizing with chemical additives. Hence, engineers are in search of sustainable stabilization alternatives. Microbial Induced Calcite Precipitation (MICP) is gaining attention as an environmentally friendly soil improvement technique. Several researchers have successfully tested its feasibility in mitigating liquefaction-induced problems in sandy soils. In this research, the authors are evaluating its effectiveness in stabilizing expansive soils. For this purpose two natural expansive soils with high and low plasticity properties were subjected to MICP treatments. The soil samples were first augmented with bacterium *Sporosarcina Pasteurii* and then treated with Calcium Chloride and Urea. Variables such as microbial concentrations and curing times were studied in this research. Geotechnical testing including Atterberg limits and unconfined compression strength were performed to evaluate the efficacy of MICP treatments. Preliminary results indicate that there is a reduction in plasticity and swelling characteristics of the soils and increase in the unconfined compression strength.

## Introduction and Background

The highly plastic expansive soils cause swelling and shrinking (volume change) with changes in moisture content. Due to these volumetric changes structures built on expansive soils tend to undergo moderate to severe cracking problems (Mitchell, 1986; Nelson and Miller, 1992). In particular, lightly loaded structures such as one or two story residential and industrial structures and pavements have experienced severe damage (Petry & Little, 2002) often associated with substantive repair and mitigation costs. The use of chemical additives such as cement and lime to stabilize these problematic soils is a common practice among geotechnical engineers, especially for lightly loaded structures. However, several occurrences of subgrade failure have been observed after stabilization with chemical additives which indicates a technology gap of sustainable stabilization of expansive soils. Soil stabilization via Microbial Induced Calcite Precipitation (MICP) is one of the several applications of bio-remediated processes that could fill this gap. This technique employs microbes as a primary contributor for soil stabilization. Successful implementation of MICP will have its application in a wide variety of civil engineering fields such as, stability for retaining walls, embankments and dams; controlling soil erosion; stabilizing cohesionless soils; increasing bearing capacity of shallow and deep foundations; and reducing liquefaction potential of soils (Kucharski et al., 2005; Ivanov and Chu, 2008, Kavazanjian and Karatas, 2008, Montoya et al., 2013).

Microbes are often responsible for the chemical cementation of soil in nature due to the precipitation of cementing materials into the voids of soils and rocks (Ivanov and Chu, 2008). Microbes can precipitate cementing materials such as calcium, magnesium, iron, manganese, and aluminum, which are crystallized to form carbonates, silicates, phosphates, sulfides and hydroxides (DeJong et al., 2006). The prime role of microbes in precipitation of minerals is their ability to create an alkaline environment through various physiological activities (Douglas and Beveridge, 1998). Calcium carbonate (calcite) precipitation is observed to be a general mineral precipitation process in the microbial world under the ambient environment (Bang et al., 2001).

Calcite mineralization can occur as a by-product of microbial metabolic activity such as photosynthesis, urea hydrolysis, sulfate reduction and iron reduction. During these different metabolic processes, the alkalinity or pH of the system increases, favoring the calcite precipitation (Knorre and Krumbein, 2000). It is believed that bacteria are dominant soil inhabitants. There are  $10^6$ - $10^{12}$  bacterial cells in a gram of soil (Torsvik et al., 1990). *Sporosarcina pasteurii* (previously known as *Bacillus pasteurii*) species of *Bacillus* group, a common alkalophilic soil bacterium has high urease enzyme activity (Dejong et al., 2006). *S. pasteurii* use urea as an energy source which hydrolyzes Urea ( $\text{CO}(\text{NH}_2)_2$ ) into ammonia ( $\text{NH}_3$ ) and carbonic acid ( $\text{H}_2\text{CO}_3$ ).  $\text{NH}_3$  and  $\text{H}_2\text{CO}_3$  equilibrate in water to form bicarbonate ( $\text{HCO}_3^-$ ), ammonium ( $\text{NH}_4^+$ ) and hydroxide ( $\text{OH}^-$ ) ions. It is during this stage the pH of system increases and shifts the  $\text{HCO}_3^-$  equilibrium to form carbonate ion ( $\text{CO}_3^{2-}$ ). The  $\text{CO}_3^{2-}$  produced will precipitate calcite ( $\text{CaCO}_3$ ) in the presence of  $\text{Ca}^{2+}$  (Dejong et al., 2006). The calcite precipitation is influenced mainly by four factors: calcium ion concentration, dissolved inorganic carbon (DIC) concentration, pH and availability of nucleation sites (Hammes and Verstraete, 2002). This precipitation between particles helps in reducing the permeability, compressibility and increasing soil strength (DeJong et al., 2010).

In this research, two expansive soils were treated using MICP technique to study the efficiency of this technique in stabilizing expansive soils. Two methods of application were investigated using unconfined compressive strength and one-dimensional swell test as performance indicators. Variables such as microbial concentrations and number of treatment cycles were also evaluated. This research is an initial step in understanding the applicability of MICP to expansive soils.

### **Application Methods**

MICP can be achieved in two ways: a) *Bio-stimulation*- This method involves the modification of the environmental condition by stimulating the indigenous bacteria present in the soil, which is typically achieved by introducing nutrients into the soil. b) *Bio-augmentation*- This method involves the introduction of the required microbes along with nutrients needed to stimulate the microbes into the soil. In this research, two approaches to bio-augmentation method were studied; Application Method-1 (AM-1) and Application Method-2 (AM-2).

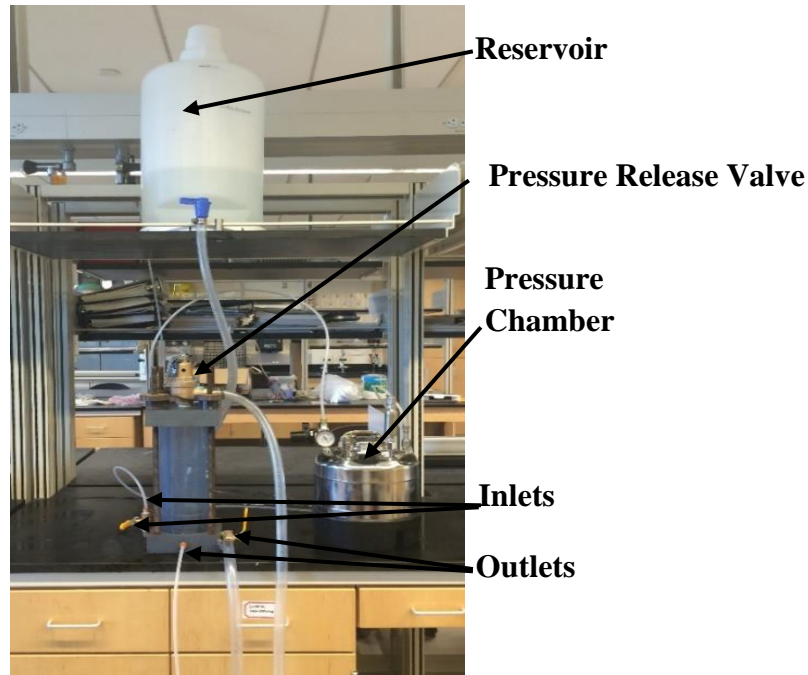
In AM-1, the microbes were mixed in the soil sample along with substrates and the mixed sample was used for further testing. This approach is similar to conventional expansive soil treatment methods using lime or cement. The mixed sample was then compacted at the maximum dry unit weight (MDUW) and optimum moisture content (OMC). The compacted sample was then cured for seven days before being tested for unconfined compressive strength (UCS) and 1-Dimensional (1-D) swell tests.

In AM-2, soil samples were prepared as in the case of the AM-1 method. However, in this case, the prepared samples were placed in a specially designed nutrient delivery system instead of being cured at constant temperature and humidity. Using this system, substrate solutions were passed through the soil samples, and the effluent was collected. For each microbial concentration soils, samples were subjected to one, three and seven pore volumes of effluent. One pore volume (PV) here represents the volume of voids present in the soil sample compacted at MDUW and OMC. A collection of effluent is termed as 'treatment cycle' in this research. After collecting respective pore volumes, samples were then tested for UCS and 1-D swell tests.

### **Nutrient Delivery System**

In order to provide nutrients to the bacteria mixed into the soil, substrate solutions consisting of Urea and  $\text{CaCl}_2$ , need to be passed through the soil sample. As the permeability of expansive soils is very low ( $< 10^{-6}$  cm/s) gravity feeding was not a practical option in view of the time needed to complete each treatment cycle. Hence, for this purpose a nutrient solution delivery system was developed as shown in Figure 1. In this system, the soil sample is housed in a chamber made of schedule 40 PVC tube of 9.4 cm diameter. This chamber is fastened between two PVC plates with dimensions, 15.2 cm x 15.2 cm. This chamber can hold pressures up to 138 kPa. This chamber has two inlets and two outlets as shown in Figure 1. One inlet is connected to the reservoir containing substrate solution while the other inlet is connected to a pressure-regulated container. The reservoir was used to fill the chamber with substrate solution while the pressure-regulated container which also contains the substrate solutions was used to drive the substrate into the soil sample. Similarly, one outlet is used to drain the chamber while the other outlet is used to collect the effluent through the soil sample.

In this system, the soil sample is surrounded by substrate solution which gets pushed through the holes present in the top cap. This arrangement allows the use of single pressure chamber to both apply confinement as well as inlet pressure. The soil sample having dimensions of 7.6 cm (diameter) x 15.2 cm (height) is placed between the top cap and the base pedestal and is wrapped around by latex membrane in order to protect the sample from surficial erosion. The top cap and base pedestal are facilitated with grooves to hold O-rings. The O-rings hold the membrane in place and also prevent water from entering inside the sample. The top cap and bottom pedestal have holes in them through which substrate solution passes through and gets in and out of the soil sample.



**Figure 1 Nutrient delivery system used in this research**

### **Materials Used in this Research**

#### **Soil Characteristics**

Two types of soils along state highway US-95 between Mileposts 16.0 to 18.0 near Marsing, Idaho were selected for this research. The plasticity characteristics of these soils ranged from low to high Plasticity Index (PI). These soils were designated as S1 (low to medium PI) and S2 (high PI). According to the Unified Soil Classification System, both of these soils were classified as high compressible clays identified with the notation CH. Characterization tests such as gradation, Atterberg limits, and compaction characteristics test were conducted on both control soils as per American Standard Testing Methods (ASTM) ASTM D6913, ASTM D4318, and ASTM D698, respectively. In addition to these tests, engineering tests such as Unconfined Compressive Strength (UCS) and 1-D Swell were also performed on the control soils as per ASTM D2166 and ASTM D4546 respectively. These results are presented in Table 1.

**Table 1 Engineering properties of natural soil samples**

Soil Notation		S1	S2
Atterberg Limits	Liquid limit	54	115
	Plastic limit	39	53
	Plasticity index	15	62
Maximum Dry Density (kN/m <sup>3</sup> )		13.6	12.0
Optimum Moisture Content (%)		30	34
% finer than 0.075 mm		70	74
Unified Soil Classification System		CH	CH
Unconfined Compressive Strength (kPa)	Saturated	24.5	28.6
	Unsaturated	58.8	239.5
1-D Swell Strain (%)		2.83	8.85

### **Microbial Characteristics**

The bacterial strain used in this research was ureolytic bacteria, *Sporosarcina pasteurii* (formerly known as *Bacillus pasteurii*). The growth media used to grow the microorganisms was primarily Laurel Broth (LB). The microbial concentration for the AM-1 method was maintained at 10<sup>8</sup> microbial colonies per gram of soil. In the case of the AM-2 method, two microbial concentrations were studied, 10<sup>8</sup> and 10<sup>10</sup> microbial colonies per gram. Commercially available urea and calcium chloride were used in this research as substrates. The concentration of urea and calcium chloride was 333 mM and 250 mM respectively. The concentration of substrate was established from the previous researches conducted on sand through MICP technique.

In order to maintain the consistency of microbial concentration throughout the research, colony formation unit (CFU) method was adopted to determine the concentration of microbes in a given solution. For this purpose, *S. Pasteurii* was cultured in Laurel Broth (LB), incubated for 48 hours at room temperature. After 48 hours of inoculation, the optical density (OD) of the cultured microbes was measured. OD is the method of measuring the concentration of microbes in a sample by measuring the turbidity of the sample at certain wavelength usually 600 nm (Madigan et al., 2012). These cultured microbes were then serially diluted in various ratios such as 1:200, 1:40000, 1:8000000. After serial dilution, 100 µL of the diluted media was taken and then plated in an LB plate. LB plate was prepared by mixing 10 g of LB and 6 g of agar in 400 ml of distilled water. The media after autoclaving was poured into the petri dish. The media solidifies after few hours due to the presence of agar. After 48 hours of plating, the number of colonies was counted. The CFU/ml for each serial dilution is given as per Equation (1).

$$CFU/ml = \frac{\text{Number of colonies per ml plated}}{\text{Total dilution factor}} \quad (1)$$

### **MICP Evaluation**

In order to evaluate the effectiveness of MICP in stabilizing expansive soils, UCS and 1-D swell tests were chosen as performance indicators. For AM-1 treated samples, these tests were conducted after seven days of curing while for AM-2 soil samples the tests were performed after 1 PV, 3 PV and 7 PV of treatments. The treated samples were of same dimensions as UCS tests hence this test was performed on the treated samples at the end of the testing period with any sample alteration. In the case of 1-D swell tests, the samples were trimmed to a diameter of 6.35 cm and thickness of 2.54 cm with the help of the oedometer ring. Samples in the oedometer ring were oven dried in order to let the samples swell from a very dry state. A similar procedure was performed on control soils as well. As explained earlier, 1-D swell tests were performed according to the ASTM-D4546, method A where the samples are allowed to swell to a maximum value before bringing them back to their initial volume. In this paper, only the swell strain data is discussed and not the swell pressure data. The results of these tests are discussed in the following sections.

### Application Method-1

Soil samples treated using AM-1 protocol were tested for UCS and 1-D swell test after seven days of curing. The UCS values for seven days cured samples are shown in Figure 2(a) for both the soil samples. It can be observed from this figure that the UCS value increased from 58.8 kPa to 88.0 kPa for S1 soil sample with an increase of 49.5%, while UCS value decreased by 39.4% for an S2 soil sample. The reduction in case of S2 soil could be due to the high plasticity nature of this soil and inadequate substrate present in the sample. As microbes require moisture to survive, seven days curing may have made microbes dormant and inactive. As soil samples S2 have high fines content (74%, passing through sieve no#200), this may have made the mobility of microbes less possible. Pore size distribution and the proportion of pore filled with water plays an important role in the contact between microbes and soil particles (Chenu and Stotzky, 2002).

The 1-D swell strain data for seven days cured samples are shown in Figure 2(b). It can be observed that the swell strain values decreased by 11% from 2.83% to 2.52% for S1 soil and by 44.1% from 8.85 % to 4.95% for S2 soil. From Figure 2(b), it is evident that bio-augmentation was effective for S1 sample with low plasticity in reducing 1-D swell percentage and increase in strength. Reduction in swelling was also observed for S2. However, the strength did not increase in case of S2 soil samples. One of the reasons for the reduction in swelling may be due to the cationic exchange in the clay particles due to the presence of calcium chloride in the substrate solution.

### Application Method-2

Soil samples treated using AM-2 protocol were tested for UCS and 1-D swell test after collecting one pore volume (1 PV), three pore volumes (3 PV) and seven pore volumes (7 PV) of substrate effluent. The results obtained from both tests are discussed here. In this application method, two different microbial concentrations, M1 and M2 were studied. M1 and M2 represent  $10^8$  and  $10^{10}$  microbial colonies per gram of dry soil, respectively. The UCS test results for both soil types and for both microbial concentrations are presented in Figure 3. It can be observed from this Figure that for S1 soil treated with M1 concentration, the UCS value gradually increased from 25.8 kPa to 54.2 kPa i.e. by 121% of untreated soil strength after 7 PV. However, the treatment did not have a similar effect on the strength of S2 soil. There was a slight increase in UCS value from 28.6 kPa to 32.2 kPa after 7 PV treatment which is an increase in UCS value by 12.6%. UCS values also increased when soils were treated with M2 concentration. The UCS was observed to be 32.8 kPa for S1 soil samples after 7 PV. The increase in the percentage of UCS for S1 after 7 PV was observed to be 34.2 % while little or no change in UCS value was observed in the case of S2.

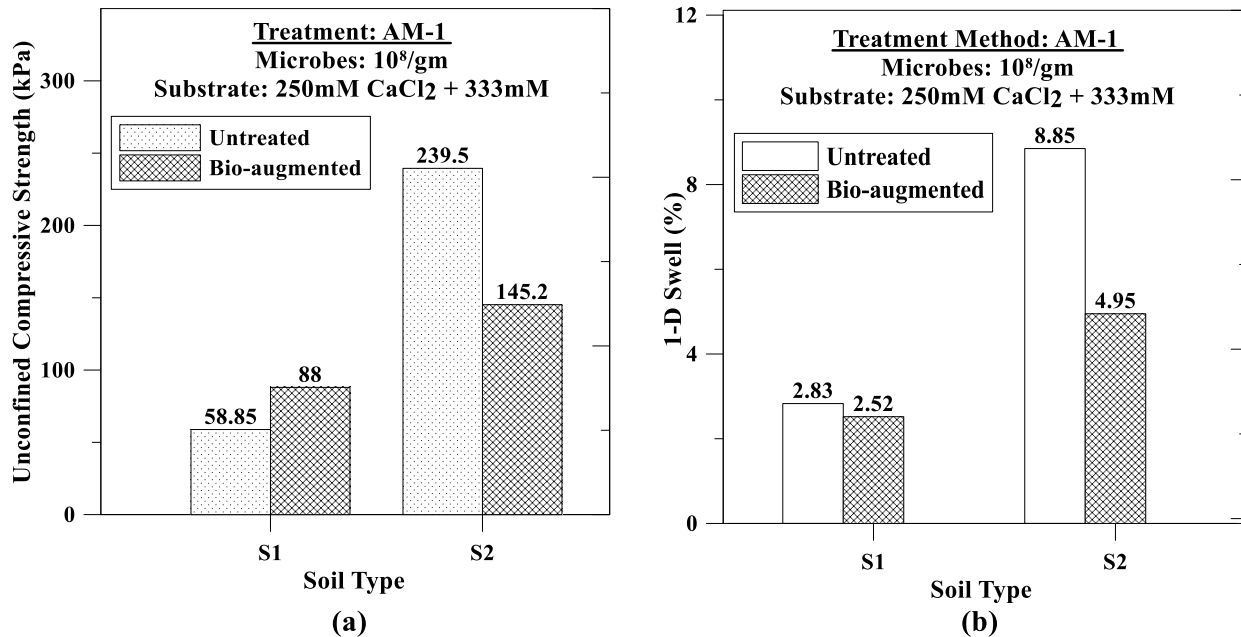
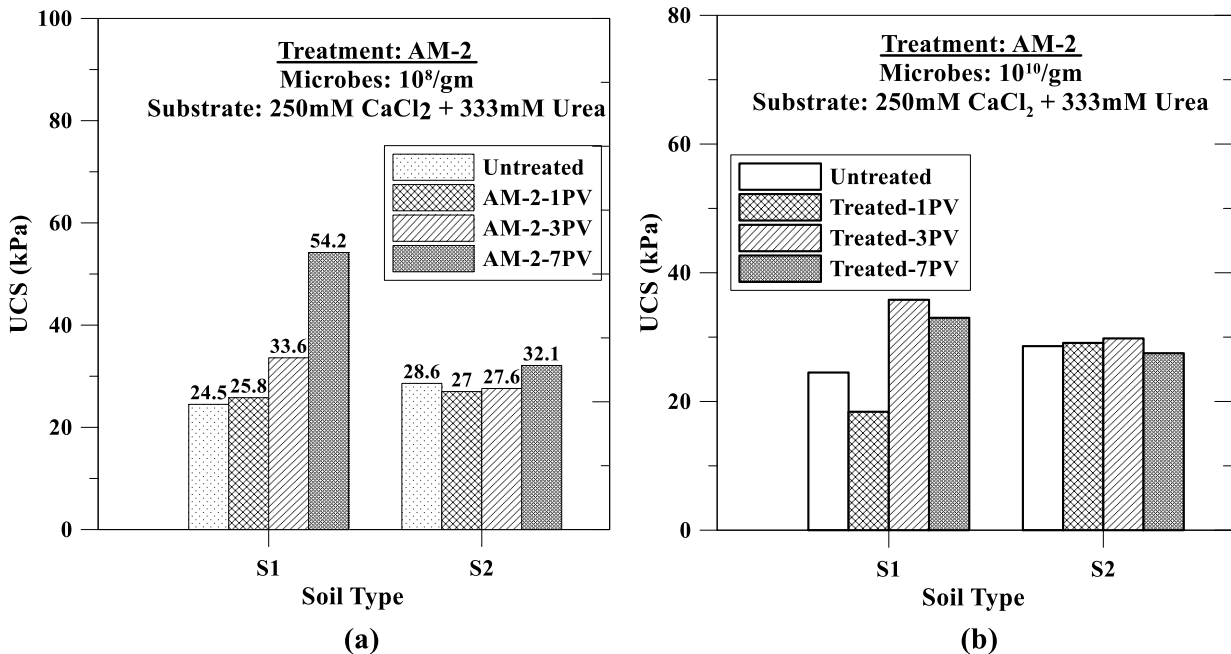


Figure 2 Test data for AM-1 treated soil samples (a) UCS test (b) 1-D Swell test

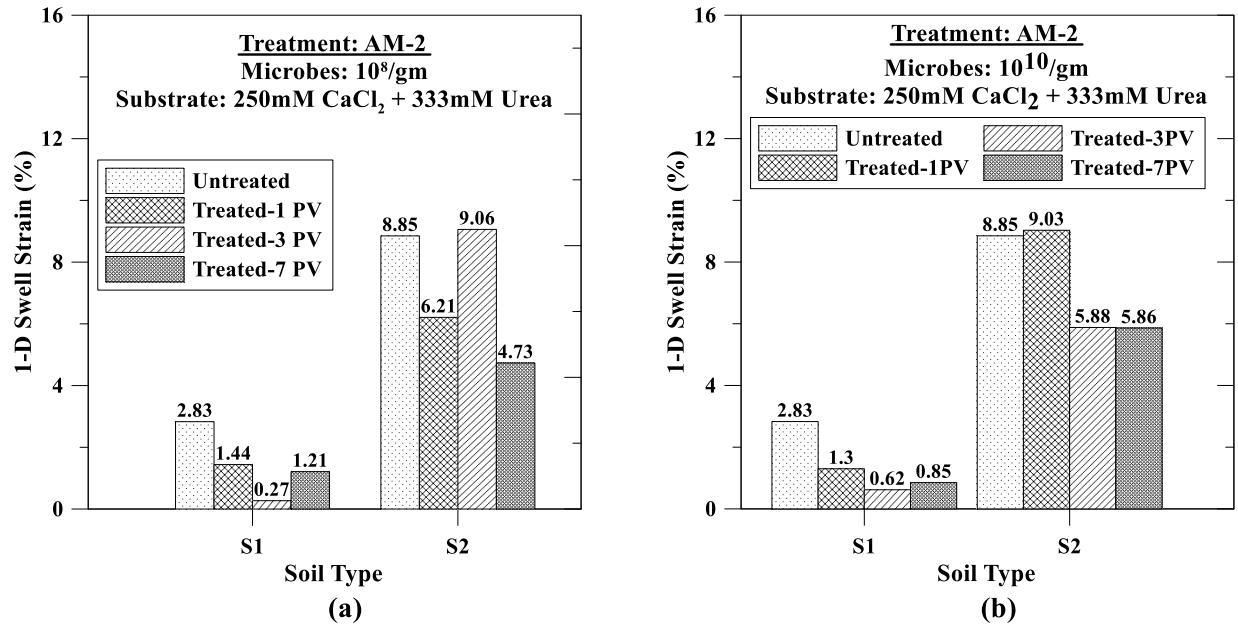
In the case of AM-2, it is evident from Figure 3 (a) that with the increase in a number of pore volumes, the strength also increased. That is microbes in the soil require enough retention period to produce urease enzyme required to hydrolyze urea. Retention period helps bacteria to dwell into the liquid media (Burbank et al., 2013). It is also observed that the MICP technique whether bio-augmented or bio-stimulated is favored in low plasticity index soil such as S1. As in both the cases, the UCS value increased by 49% and 121 % respectively as compared to the untreated S1. However very little or no increase in UCS value was noticed in the case of S2 soil, and this may be due to the absence of microbial activity.

From Figure 3 it can also be observed that increase in microbial concentration did not increase the UCS value. Ramachandran et al. (2001) concluded that higher concentration of bacteria had no improvement in strength. They suggested that slower rates of calcite formation were more prominent in imparting higher strength than higher rates. Comparison between the  $10^8/\text{gm}$  and  $10^{10}/\text{gm}$  microbial concentration for each pore volume shows that the increase in microbial concentration did not increase the strength of these samples. The factors that influence the precipitation of calcite are mainly the concentrations of  $\text{Ca}^{2+}$  and  $\text{CO}_3^{2-}$ , pH of the system and the nucleation site. Bacterial cell surface acts as a nucleation site for the precipitation of the calcite. The solubility product ( $K_{sp}$ ) of calcite is very low ( $3.3 \times 10^{-9} \text{ mol.L}^{-1}$  at  $25^\circ\text{C}$ ), and for precipitation of calcite supersaturation of  $\text{Ca}^{2+}$  and  $\text{CO}_3^{2-}$  must exist. Since calcite has very low  $K_{sp}$ , supersaturation can be achieved by simply mixing  $\text{Ca}^{2+}$  and  $\text{CO}_3^{2-}$  together in moderate concentrations. However, when the reaction takes place rapidly, the crystals formed are very small and powder like with little or no cementation strength (Whiffin, 2004). In order to have large crystal precipitation over an extended period of time with higher cementation strength, the supersaturating product concentration should remain low. The supersaturation of  $\text{CO}_3^{2-}$  is also influenced by the pH of the system. pH can be regulated by the dissociation of urea into  $\text{NH}_4^+$ .  $\text{CO}_3^{2-}$  concentration remains very low below pH 8. Thus, the size of crystal can be increased or decreased by decreasing or increasing the pH of the system (Whiffin, 2004). The presence of higher microbes at the beginning of the might have contributed to higher rates of calcite formation thereby hindering strength development in the case of M2 concentration.



**Figure 3 UCS test results of AM-2 treated soil samples for (a) M1 concentration and (b) M2 concentration.**

The 1-D swell test results of both samples with a microbial concentration of  $10^8/\text{gm}$  are presented in Figure 4 (a) for all three pore volumes. It was observed that the swell strain reduced in the case of S1 samples after all three treatment cycles. Reduction in swell strain was also observed for S2 after 7 PV. Similar results were obtained when both soils were treated with a microbial concentration of  $10^{10}/\text{gm}$  after 7 PV of treatments which is shown in Figure 4 (b).



**Figure 4. 1-D Swell Strain data for AM-2 treated soil samples (a) M1 concentration and (b) M2 concentration.**

1-D swell strain reduced for S1 soil samples after all three pore volumes. It was also observed that after seven pore volumes, the swell reduction was possible i.e. higher the treatment cycles (or retention period) lower the swell strain. Reduction in swelling was also observed for S2 soil. However, the reduced values were still considered problematic swelling strains. One of the reasons for the reduction in swelling for S2 soil may be due to the cationic exchange in the clay particles due to the presence of calcium chloride as substrate. As there was no increase in strength, it was assumed that microbial activity was minimal in this soil.

### Summary

Two expansive soils with varying plasticity characteristics were tested to evaluate the effectiveness of MICP in stabilizing expansive soils. These soils were subjected to two methods of MICP treatments, and their performance was measured by monitoring swelling potential and unconfined compressive strength with various treatments. Variables such as soil type, bacterial population during augmentation, along with the number of treatment cycles were studied in this research. This research is the initial step to understand the applicability of MICP in expansive soils. The data presented in this research supports the applicability of MICP in expansive soils particularly in the case of low plasticity soils such as S1. However, changes in geotechnical properties of high plastic soils such as S2 soil samples' after MICP treatment needs further testing to understand the feasibility of MICP technique in high plastic soils.



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