Made available through Montana State University's ScholarWorks

Effect of Fines Content on Calcium Carbonate Precipitation and Thermal Properties of Biocemented Sand

Pinar Gunyol, Mohammad Khosravi, A. J. Phillips, Kathryn Plymesser, Albert E. Parker

This material may be downloaded for personal use only. Any other use requires prior permission of the American Society of Civil Engineers. This material may be found at https://doi.org/10.1061/JGGEFK.GTENG-11925

Accessibility Disclaimer:
For a more accessible version of this document, please submit an accessibility request form through the Montana State University Library website.

Effect of Fines Content on Calcium Carbonate Precipitation and Thermal Properties of Bio-cemented Sand

by

Pinar Gunyol, S. M. ASCE
Department of Civil Engineering
Montana State University, Bozeman, MT 59717

Mohammad Khosravi, Ph.D., M. ASCE
Department of Civil Engineering
Montana State University, Bozeman, MT 59717
(Corresponding author: email: mkhosravi@montana.edu, phone: 7202890497)

Adrienne Phillips, Ph.D.
Department of Civil Engineering
Montana State University, Bozeman, MT 59717
(email: adrienne.phillips@montana.edu)

Kathryn Plymesser, Ph.D., P.E.
Department of Civil Engineering
Montana State University, Bozeman, MT 59717
(email: kathryn.plymesser@montana.edu)

Albert Parker, Ph.D.
Center for Biofilm Engineering
Department of Mathematical Sciences
Montana State University, Bozeman, MT 59717
(email: parker@math.montana.edu)

Submitted

May 2023

ABSTRACT

In this study, the impacts to soil thermal properties during and after bio-cementation via Microbially-Induced Calcite Precipitation (MICP) method on silty silica sand specimens with varying fines content (0%, 5%, and 15%) were investigated. Firstly, calcium conversion was measured after each pulse, then the MICP treated specimens were tested for cementation uniformity. The evolution of thermal conductivity of silty soils with the MICP treatment was assessed using a TR-3 thermocouple probe. The results show that thermal conductivity of silty sands increased by 17% for specimens treated to 9.7% $CaCO_3$. The improvement in thermal conductivity was attributed to the formation of calcium carbonate bridges binding the soil grains together. The results suggested that the thermal conductivity of silty soil depends on water content, the number of treatment pulses, and the treatment uniformity through the soil specimen. Presence of fines content in the soil was found to play an important role in the distribution and uniformity of biocementation through the soil specimen. However, no statistically significant difference in the thermal conductivity values of MICP treated specimens with different fines content was observed ($p > 0.05$). The average calcium carbonate content ranged between 10.7% and 7.2% for the soils with 0% and 15% fines content, respectively. The findings of this research could be used to improve the efficiency of geothermal boreholes and other energy geo-structures using MICP by improving thermal conductivity of dry and partially saturated soil.

Introduction

In recent years, there has been an increasing interest in the use of biological technologies in geotechnical engineering to improve thermo-hydro-mechanical properties of geomaterials (Mitchell et al. 2013). Certain microbes can contribute to altering their surrounding chemical environments to induce the precipitation of calcium carbonate, a form of bio-cement (Van Paassen

2009; DeJong et al. 2010; Phillips et al. 2013). One mechanism that has been researched extensively is the microbially or plant produced enzyme urease, which promotes the hydrolysis of urea to alter water content and saturation conditions. In the presence of calcium, this change in saturation induces the precipitation of calcium carbonate (Eqn 1) (Phillips et al. 2016; Hommel et al. 2020; Mitchell et al. 2013; Phillips et al. 2013; Ebigbo et al. 2012).

$$(NH_2)_2CO + 2H_2O + Ca^{2+} \rightarrow 2NH_4^+ + CaCO_3 \quad \text{(Eqn. 1)}$$

In this process, the precipitation of CaCO3, commonly in the stable mineral form of calcite, can bind together the soil particles that make up porous media. Bio-cemented sand treated by MICP has been reported to provide a reduction in soil settlement (DeJong, Fritzges, and Nüsslein 2006; Van Paassen 2009), an increase in soil shear strength (Ismail et al. 2002; DeJong, Fritzges, and Nüsslein 2006; Chou et al. 2011), and an increase in soil stiffness (Montoya and DeJong 2015; Feng and Montoya 2016; Lin et al. 2016).

MICP technique has been recently proposed as an alternative for improving the thermal properties of soil (Venuleo et al. 2016; Martinez et al. 2019; Xiao et al. 2021; Wang et al. 2020). Heat transfer in soil takes place primarily through conduction, and flow of heat is largely controlled by the contacts between the solid particles. Precipitation of calcite results in an increase in the solids content and therefore, an increase in the average dry density of the soil through the process of binding particles together at their contacts by calcite bridges (Whiffin, van Paassen, and Harkes 2007). Calcite crystals formed around soil particles increase the number of particle contacts and enhance heat transfer paths through solid particles resulting in a higher thermal conductivity of the treated soil (Venuleo et al. 2016). MICP treatment of sand could lead to a significant increase in soil thermal conductivity by up to 250% (Venuleo et al. 2016) particularly in dry and partially saturated soils where the calcite crystals replace the air. The same increase in calcium carbonate led

47 to 330% increase in thermal conductivity at the degree of saturation, S_r = 0%, while the increase in
48 thermal conductivity was 37% at S_r = 100% (Martinez et al. 2020). Previous studies on the effect
49 of bio-cementation on soil thermal behavior have mainly focused on thermal properties of medium
50 sand (e.g., median size of 0.2 mm). The presence of fine-grained soil (e.g., silt) in the soil matrix
51 adds complexity to the applicability of MICP techniques due to smaller pore sizes and larger
52 specific surface areas (Zamani and Montoya, 2017). Thus, the thermal properties of bio-cemented
53 soil as a function of the amount of $CaCO_3$ formed and fines content require further investigation.
54 The effect of grain size on thermal conductivity is primarily linked to number of physical
55 contacts among soil particles (Beziat et al. 1992). The increase in thermal conductivity with
56 increasing fines content is expected to continue until the sand-silt granular mixture reach its
57 maximum dry density. Zhang et al. (2015) mixed sands with different median particle size, Silica
58 12/20 (with median particle size, d_{50}=1.03 mm), Ottawa 20/30 (with d_{50} = 0.78 mm) and ASTM
59 graded (d_{50} = 0.36 mm) and observed that the thermal conductivity of sand improved with fines
60 content. The results suggested that thermal conductivity of the mixed sand initially increases up to
61 a critical fines content (F_{cr}) due to more efficient interparticle contacts and contacts per unit volume.
62 For the coarse sand (Silica 12/20 with median particle size, d_{50}=1.03 mm), the critical fines content
63 was 70% and a 30% increase in thermal conductivity was observed. Although presence of fines
64 could improve thermal properties of soil, previous studies have indicated that homogeneous
65 distribution of bacterial cells in fine-grained soils or soils with fines content could be harder to
66 achieve due to the reduction in pore spaces and the increase in filtration effect (Rebata-Landa and
67 Santamarina 2006; Zamani 2017). Based on Zamani (2017), cementation distribution in silty
68 Nevada sand with 15% silt is less uniform compared to the clean Nevada sand (d_{50} = 0.15mm).

This paper summarizes the results of a series of column experiments on the effect of fines content on bio-cementation process and thermal properties of bio-cemented silty sand. The ureolytic bacterium *Sporosarcina pasteurii* was used as a source of urease enzyme and calcium and urea containing solutions were injected into sand-filled columns to promote MICP. Thermal conductivity was monitored to evaluate the alteration in the soil matrix as mineral forms in time and its effect on thermal properties of the sand. Following, an assessment of treatment homogeneity (i.e., uniform distribution of $CaCO_3$ through the treated soil matrix) was conducted. The results of the experiments and analyses provide insights into the thermal behavior of bio-cemented silty sands.

Materials and Methods

The laboratory testing program characterized soil samples of Ottawa (F-65) Sand (US Silica, Ottawa, Illinois) mixed with 0%, 5%, and 15% silt (Sil-Co-Sil 250 obtained from U.S. Silica in 2020) (by weight). Figure 2 presents the grain size distributions and SEM images for both Ottawa sand and Silica silt. Ottawa sand is a uniform white silica sand classified as SP (USCS ASTM 2487) with rounded grains, a quartz content of 99.7%, and minimal fines (US Silica 2016). Properties of the uniform sand included the following: maximum void ratio, e_{max} = 0.83; minimum void ratio, e_{min} = 0.51; specific gravity of solids, G_s = 2.65; median particle size, d_{50} = 0.20 mm. Sil-Co-Sil 250 is a highly granular, non-plastic silt with a quartz content of over %99.5. The material is low in moisture, chemically inert, and acid resistant. Properties of the silt material included the following: G_s = 2.75; plasticity index, PI=0 (Price, 2018, U.S. Silica). Table 1 summarizes the e_{max}, e_{min}, and G_s of silty sand with silt content of 0, 10, and 15%. e_{max} and e_{min} were determined following ASTM D4253 (ASTM 2002a) and ASTM D4254 (ASTM 2002b), while G_s was determined following ASTM D792.

All soil specimens were prepared in columns with an inner diameter of 50-mm and a height of 120 mm. The soil samples were constructed layer by layer; each layer was compacted to a target relative density of D_r = 50%, up to the top. The amount of soil required for each lift was determined based on the desired density. Compaction was performed in eight lifts with moist soil (a moisture content of 7%) distributed evenly for each layer. To prevent excessive compaction of lower layers, Ladd's (1978) compaction method was used. A portable, battery-operated TEMPOS Thermal Properties Analyzer and a TR-3 thermal probe were used to measure thermal conductivity of soil samples. Figure 1 displays a schematic of the system setup and the PolyVinyl Chloride (PVC) column with the thermal probe. The TR-3 sensor is a 100-mm long, 2.4-mm diameter needle that was inserted into the specimen through the cap and kept inside the column throughout the experiment (Figure 1). The design allowed for evaluation of thermal conductivity over time as mineralization takes place in the sand matrix, without causing disturbance in the soil. A minimum of 10 mm of soil material parallel to the TR-3 sensor probe in all directions was required to avoid errors in measuring thermal properties of the soil. The inside surface of the PVC column was covered with low-friction Teflon tape (Low-Friction FEP Tape, McMaster-Carr) to minimize the attachment of the sand to the column. Two non-corrosive wire mesh filters were placed at the bottom and top of the sand specimen to prevent intrusion of the sand material into the injection or effluent tubing.

Thermal experiments were conducted in three stages. After the soil sample was prepared in the column, the TR-3 sensor was inserted into the untreated moist soil and the thermal conductivity of the soil was measured (Stage 1). The samples were then saturated with deionized water and the thermal conductivity of the saturated soil was recorded (Stage 2). The saturated columns were subjected to MICP treatments. During the treatment phase, the thermal conductivity of the

specimens was recorded in 2-hour intervals between each cementation treatment and 16-hours after biological treatment, while for the saturation stage, the thermal conductivity at the end of each were measured (Stage 3). A control column was treated only with deionized water, without bacteria, urea, or calcium sources.

Microbial Growth

Prior to injections of treatments, a starter culture was prepared by aliquoting 1 mL of *Sporosarcina pasteurii* (ATCC 11859) from thawed frozen stock to 100 mL of filter sterilized Brain-Heart Infusion broth (BHI, Becton Dickinson) amended with 2% urea (20 g/L urea, Fisher Scientific). The cultures were incubated overnight at 30°C on a 150-rpm shaker. Following incubation, 1 mL of this culture was added to 100 mL of growth promoting media for 24 hours for use (injection into the column) the next day. This starter was then used to inoculate a bacterial solution for the treatment process.

Media

The process of MICP is done using an alternating set of fluids introduced into a medium. The bacterial growth solution, BHI + Urea, was used to propagate bacterial cultures for treatment (Table 2). The calcium-providing solutions, CMM+ (Table 2), share the same base set of ingredients, but include calcium chloride.

Injection Strategy

The columns were designed such that pulsed injections with resting batch reaction periods between injections of enzymes and calcium containing solutions were repeatable (i.e., flow rate and hydraulic residence time) between different specimen types. The compacted specimens were disinfected by injecting 1.5 pore volumes of 70% ethanol and 10% bleach. The specimens were then rinsed and saturated by injecting two pore volumes of deionized water prior to MICP treatment

phase. Biological and cementation solutions (1.5 pore volumes) were injected using an alternating method (Gunyol et al. 2022). The alternating method involved flipping the soil specimens and injecting the reagents from the top and then the bottom of the column (while ports remained fixed). This method of alternating injection between the column ends by flipping the specimens was shown to be more effective at achieving a uniform CaCO3 distribution along the specimen height compared to other injection methods such as bottom-to-top injection, for the thermal conductivity testing (Gunyol et al. 2022). The volume of injection fluids per pulse were 100 mL for 0% fines specimen, 95 mL for 5% fines specimen, and 85 mL for 15% fines specimen, which was based on the calculated pore volumes, as summarized in Table 3. The injection rate was kept constant at 4 mL/min throughout the experiments. An injection rate of 4 mL/min was selected for the testing because higher rates caused localized precipitation and weak cementation due to insufficient time for the bacteria to hydrolyze urea, while slower rates allowed more uniform cementation along the column by providing time for greater urea hydrolysis efficiency (Gunyol et al. 2022). The system was not monitored for the pressure increase. A summary of the soil specimens tested in this study and fines content (0%, 5%, 15%, denoted throughout as F00, F05 and F15) for each specimen is presented in Table 3. Specimens denoted with P32 (32 pulses of injections) were subjected to 24 pulses of CMM+ and 8 pulses of biological treatments. Specimens denoted with P16 (16 pulses of injections) received 12 pulses of CMM+ and 4 pulses of biological treatments. Table 4 provides the details of the injection program for the treatment solutions. Following the MICP treatments, the specimens were dried in a 40 ºC oven for further analysis.

Ca^{2+} concentration determination: *Effluent fluid sampling*

The column design allowed for assessment of the reaction progression (for example, collection of effluent samples of the fluids to assess the urea hydrolysis and subsequent calcium precipitation).

160 A syringe pump (KD Scientific) was used to inject solutions into the column(s). A sample of
161 effluent was collected from the fluids that were in the soil specimens after each batch period. The
162 effluent samples were analyzed for calcium concentration via colorimetric calcium assay and urea
163 concentration via a modified colorimetric Jung assay (Jung et al., 1975; Phillips, 2013).

164 ***$CaCO_3$ determination:*** *Acid washing cemented specimen*

165 $CaCO_3$ content was determined using a gravimetric acid washing method. The specimens were
166 divided into 5 sections along the length. Each section was crushed to powder using a blender. Then
167 1 g sample was added to 10 % (V/V) HNO_3 to allow for $CaCO_3$ to be dissolved. The mass of $CaCO_3$
168 was calculated by measuring the difference between oven-dried mass of the soil sample before and
169 after the acid washing process.

170 ***Microstructure Analysis***

171 Samples from MICP-treated columns with 0%, 5% and 15% fines content, and the control were
172 saved for further analysis through microscopy imaging. Scanning Electron Microscopy (SEM) and
173 Energy Dispersive Spectroscopy (EDS) were conducted using JEOL JSM-6100 and Zeiss SUPRA
174 55VP field emission systems (ICAL, Bozeman, Montana), respectively. Voltages used for SEM
175 and EDS imaging were 1.00 KV and 15 KV, respectively. The Energy-Dispersive x-ray
176 Spectrometer (EDS) was used to generate characteristic x-rays to examine individual elements
177 encountered. To ensure accurate imaging of the soil samples without any disruption or distortion
178 caused by charging, all samples were coated with a thin layer of Iridium using a sputter coater. The
179 sputter coater process involved bombarding the surface of the samples with high-energy ions, which
180 deposited a thin layer of Iridium on the surface. The samples were coated for 90 seconds at a current
181 of 20 mA.

Analysis of Variance (ANOVA)

To determine the effect of: 1) fines content on thermal conductivity, k_t, 2) the effect of Ca^{2+} conversion on the change in thermal conductivity Δk_t, and 3) how the effect of Ca^{2+} conversion on Δk_t depends on fines content, an analysis of variance (ANOVA) was performed. The p-value obtained by ANOVA was compared to a significance level of 0.05 to test the null hypothesis, which stated that the thermal conductivities were all equal under all conditions. A significance level of α=0.05 indicates a 5% risk of incorrectly concluding a difference exists (rejecting the null hypothesis) when there is no actual difference.

Results

The results of a series of column experiments combined with thermal testing are presented to illustrate the effects of fines content on the distribution of bio-cementation and thermal properties of bio-cemented sandy silt. The results include calcium carbonate precipitation profile and the evaluation of thermal conductivity of MICP treated sand specimens (Ottawa F-65) with different fines content (Sil-Co-Sil 250) (0%, 5%, 15%, denoted throughout as F00, F05 and F15) and different pulse treatments (16 or 32 pulses, denoted as P16 or P32). For example, results for 0% fines content and 16 pulses of treatment are denoted by F00-P16 and results for 15% fines content and 32 pulses of treatment is denoted by F15-P32. An ANOVA was also conducted to investigate whether fines content and Ca^{2+} conversion resulted in a statistically significant difference in thermal conductivity.

Effect of fines content on calcium carbonate distribution and precipitation

Figure 3 shows the cumulative Ca^{2+} (g) conversion calculated based on the concentration of calcium data from the effluent samples collected during each pulse compared to the concentration in the injected fluids (influent). The results show that the calculated Ca^{2+} conversion was the highest for

205 the specimen with 0% fines (~ 22.6 g) and lowest for the specimen with 15% fines (~ 16.3 g). The
206 results from effluent sampling method and post-treatment acid digest method were consistent. The
207 results from acid digestion method indicated that the amount of total $CaCO_3$% present within the
208 specimen was the lowest in the specimen with 15% fines and the highest in the specimen with 0 %
209 fines. The specimen with 0%, 10%, and 15% fines had an average $CaCO_3$ of 10.7%, 9.23%, and
210 7.16%, respectively across the five layers in the specimens that were tested. Lower precipitation in
211 the sample with fines content was expected as the samples with a higher silt content had lower
212 measured pore volume, and thus received lower amount of injection fluids (Zamani, 2017).
213 Measured pore volumes of specimens with 0%, 5%, 15% fines were 65 mL, 62 mL, 55 mL,
214 respectively.

215 The $CaCO_3$ content distributions of specimens with 0%, 5% and 15% silt content for
216 specimens receiving 32 and 16 pulses are presented in Figures 4a and 4b, respectively. For all
217 specimens, the amount of precipitated $CaCO_3$ gradually decreased as the distance from the top and
218 bottom injection ports increased. For the specimens that received 32 pulses, the average amount of
219 $CaCO_3$ present in one gram samples ranged from 14.7%-11.6% for the section near the bottom port,
220 8.3%-3.23% for the mid-section, and 10.7%-8.7% for the top injection port. The observed bio-
221 cementation distribution along the soil height could be attributed to the filtration of bacterial cells
222 through the soil particles, as well as encapsulation of bacteria, resulting in a higher amount of
223 CaCO3 precipitation at the top and bottom layers (Whiffin, van Paassen, and Harkes 2007).
224 Filtration of bacteria through silty soils could result in a nonuniform distribution of bacteria through
225 the soil sample with a higher concentration close to the injection port (Zamani and Montoya, 2018).
226 The $CaCO_3$ precipitation pattern is primarily linked to the delivery and efficiency of urease activity
227 of the injected bacteria (Van Paassen 2009; Barkouki et al. 2011). Bacterial cells can act as

nucleation sites of mineral formation during MICP (Stocks-Fischer et al. 1999; Kirkland et al. 2021). Bacterial cells tend to adhere to particle-particle contacts causing a significant amount of calcium carbonate to precipitate at these locations (DeJong et al. 2010). As the bacterial cells are traveling through the pore spaces, they are filtered through the soil particles with generally a reduction of microbe concentration along the injection path (Feng and Montoya 2016; Ginn et al. 2002).

The effect of fines content on uniformity of bio-cementation throughout the specimen is evident in Figure 4, which shows that the reduction in cementation in the mid-height of the column becomes more pronounced as the fines content increases. The presence of fines content reduces pore volume, resulting in decreased soil permeability and potentially increased filtration of bacterial cells (Zamani and Montoya 2018). The reduction in soil permeability could result in slower transport of injected fluids through the specimen. Slower flow allow more bacteria to attach to particle surfaces (Foppen and Schijven 2006; Harkes et al. 2010; Cheng et al. 2013; Gunyol et al. 2022). Therefore, a higher concentration of bacteria around the injection ports can hydrolyze a greater amount of urea, resulting in localized precipitation of $CaCO_3$ in samples with fines. The experimental results suggest that uniformity in cementation distribution improved by increasing the number of pulses from 16 to 32. The improvement in uniformity was more pronounced for the specimen with 15% fines content. By increasing the number of treatments from 16 to 32 pulses, the $CaCO_3$% (g/g) at the mid-height of specimen F15 increased by 5.5 times in the F15-32P specimen compared to F15-16P, while this increase was only 2.9 times in specimen F00-32P compared to F00-16P. The higher increase of $CaCO_3$% in the specimen with 15% fines might be due to the movement of loose calcium carbonate crystals and cemented silt particles. Moreover, the

precipitation around the silt particles could form silt aggregates that might detach from the sand grains (Zamani and Montoya 2018).

Effect of calcium carbonate precipitation and fines content on thermal conductivity

Figure 5 shows the variation of thermal conductivity (k_t) throughout three different phases: 1) initial condition (untreated soil sample with 7% water content), 2) after saturation and before treatment, and 3) MICP treatment phase. During the treatment phase, the thermal conductivity of the specimens was recorded in 2-hour intervals between each cementation treatment and 16-hours after biological treatment, while for the saturation stage, the thermal conductivity at the end of each was measured. The specimens remained saturated throughout the treatment stage.

The specimens were initially compacted with a water content of 7%. After compaction, three untreated control soil specimens with fines content of 0%, 5%, and 15% were placed in an oven at 40° C and their thermal conductivities were measured. The dried untreated control soil specimens (0% water content) had thermal conductivity values of 0.22, 0.25, and 0.26 W/mK for fines content of 0%, 5%, and 15%, respectively. Figure 5 also includes the thermal conductivity of an untreated soil saturated with deionized water with k_t = 2.48 W/mK as a control sample. This value was similar to the thermal properties of other pore filling fluids (cementation solution with k_t = 2.44 W/mK and biological solution without *S. pasteurii* with k_t = 2.52 W/mK). Therefore, the results were only presented for the sample saturated with water.

As shown in Figure 5 and Table 5, the initial values of k_t for the untreated columns with 7% water content and 0%, 5%, and 15% fines were 1.81, 2.08, and 2.13 W/mK, respectively. The results showed a rapid increase in thermal conductivity when the samples were saturated and the air (k_{air}~0.02 W/mK) in the pore space in the soil matrix is replaced with water (k_{water}~0.3 W/mK). Under the saturated and untreated conditions, thermal conductivity increased with increasing fines

content. In the untreated saturated columns with $Ca^{2+} = 0$, it was observed that F15 had the highest mean thermal conductivity with a k_t value of 2.75 W/mK, followed by F05 with a value of 2.65 W/mK, and F00 with 2.55 (p-value = 0.120). The p-value was greater than the significance level, suggesting that there was no statistically significant difference between the thermal conductivity values of untreated columns with the range of fines content studied here.

The thermal conductivity of all specimens increased with the number of pulses compared to the control saturated specimen, as shown in Figure 5. The increase in thermal conductivity was attributed to the reduction in porosity caused by $CaCO_3$ precipitation, which plays a governing role (Yun and Santamarina 2008). The binding of $CaCO_3$ crystals to the grains increases the cross-sectional area of contact points between the particles, resulting in a reduction in intergranular thermal resistivity. SEM imaging (shown in Figure 6b and 6d for 16 and 32 pulses of MICP treatment, respectively) revealed that the grains were bonded together by $CaCO_3$ bridges that could act as conductive paths ($k_{calcite}$ ~5 W/mK), allowing heat to transmit through the inter-grain region.

Bio-cementation was observed to form around the silt particles as well as between the larger sand particles in soil columns with fines content, as revealed by scanning electron microscopy (SEM) (Figure 6b and 6d) and energy-dispersive X-ray spectroscopy (EDS) images (Figure 7). This is due to the bacteria's ability to attach to both sand and silt particles and induce the precipitation of calcium carbonate. The degree and uniformity of cementation were found to depend on several factors, including soil properties (Figure 6a vs Figure 6b and Figure 6c vs Figure 6d) and the number of pulses of treatment as shown in Figure 6. Furthermore, the cemented silt particles occupy the spaces between the cemented sand grains, thereby improving the heat transfer between the particles.

Figure 8 shows the evolution of thermal conductivity with calculated cumulative Ca^{2+} (g) conversion. The Ca^{2+} conversion was determined by subtracting the measured effluent Ca^{2+}

concentration from the initial Ca^{2+} injected (CMM+, cementation solution). The increase in Ca^{2+} conversion implies a higher degree of $CaCO_3$ precipitation, which results in an increase in the solid mass of the soil matrix. The increase in thermal conductivity under saturated conditions is calculated as $\Delta k_n = (k_{treated_n} - k_{untreated})$, where n is the number of pulses, $k_{untreated}$ is the thermal conductivity of the saturated untreated specimen, and $k_{treated_n}$ represents the thermal conductivity of saturated treated specimen after n pulses. A correlation between the calcium conversion and the increase in thermal conductivity was observed for all specimens with varying fines content. The change in thermal conductivity (Δk_t) increased by 17 W/mK for every 1000g Ca^{2+} converted (p-value < 0.0001), indicating that there was a statistically significant difference between the thermal conductivity values of columns with Ca^{2+} conversion.

Effect of Fines Content on Thermal Properties of Bio-cemented Soil

The effect of fines content on thermal properties of bio-cemented soil was investigated using: 1) the change in the mean thermal conductivity with the number of pulses (Figure 5), 2) the rates of increase in thermal conductivity with the number of pulses (Figure 5), and 3) the change in mean Δk_t values with Ca^{2+} conversion (Figure 8). As shown in Figure 5 and summarized in Table 5, F15-P32 resulted in the highest mean thermal conductivity value of 2.88 W/mK, followed by F05-P32 with a value of 2.79 W/mK and F00-P32 with 2.68 (p-value = 0.118). The rates of increase in thermal conductivity (k_t) of the columns were also not statistically significant, with values of 0.016 for F15, 0.019 for F05, and 0.017 for F00 (p-value of 0.259). The mean Δk_t values were calculated as 0.150 W/mK for F00, 0.141 W/mK for F05, and 0.138 W/mK for F15 (p-value ≥ 0.814). In all three cases, the p-value was greater than the significance level, indicating that there was not enough evidence to suggest that the effect of bio-cementation on the increase of thermal conductivity (Δk_t) was dependent on the range of fines content studied here.

Although the results failed to show a statistically significant effect on the thermal conductivity of treated columns with different fines content, the SEM images of the specimens F15P16 (Figure 6b) after 16 pulses and F15P32 (Figure 6d) after 32 pulses of MICP treatment showed that the presence of fine particles can help fill gaps between coarser grains, increasing the number of mineral bridges between grains and the number of paths for heat transfer. This process reduces pore size and improves cementation, as the silt particles are cemented to each other and the sand particles. Additionally, the precipitation of calcium carbonate increases soil density and improves soil thermal properties. Most importantly, the cemented silt particles can transfer heat between sand grains without being displaced, which enhances heat flow through the system. For the untreated saturated sample, the difference between the thermal conductivity of samples F00 and F15 was 0.25 W/mK. For the bio-treated sample, the difference between the two (F00-32P and F15-P32) was only 0.14 W/mK, suggesting that the effect of fines content on thermal conductivity was reduced by increasing the number of pulses which could be attributed to the reduction in porosity due to $CaCO_3$ precipitation.

Discussion

Previous studies investigating the effect of microbial-induced calcium carbonate precipitation (MICP) on the thermal properties of soils have focused exclusively on sand, with contributions from the authors listed in Table 6. These studies have examined the impact of MICP bonding between soil particles (Venuleo et al. 2016; Wang et al. 2020), degree of saturation (Martinez et al. 2019; Xiao et al. 2021), and gradation and porosity (Xiao et al. 2021) on thermal properties of MICP-treated soils. However, the influence of fines content on cementation uniformity and thermal conductivity of MICP-treated sand specimens are not fully understood. As discussed in the previous sections, fines content could influence the precipitation of calcium carbonate and uniformity of

342 MICP treated soil, the results. Nevertheless, our results show that there is no statistically significant
343 difference in thermal conductivity values among untreated columns with varying fines content.

344 Table 6 summarizes the details of previous studies including soil properties (D_r and d_{50}),
345 thermal testing method, specimen dimension, and the method used to calculate the average $CaCO_3$
346 content in the specimen. Figure 9 plots the changes in the thermal conductivity ($\Delta k = k_{treated} -$
347 $k_{untreated}$) obtained in this study versus average $CaCO_3$% in and compares them with those obtained
348 by Martinez et al. (2019) (Figure 9a), Xiao et al. (2021), and Wang et al. (2020) (Figure 9b). $k_{treated}$
349 and $k_{untreated}$ are thermal conductivities of saturated treated and saturated untreated specimens. The
350 average $CaCO_3$% presented for this study was calculated based on the amount of $CaCO_3$% present
351 in the soil along the thermal probe length. $CaCO_3$ content was defined as the ratio of the mass of
352 $CaCO_3$ to the mass of oven-dried MICP-treated soil (Figure 9a) (similar to Martinez et al. (2019)),
353 while Xiao et al. (2021) and Wang et al. (2020) defined the $CaCO_3$ content as the ratio of the mass
354 of $CaCO_3$ to the mass of MICP-treated soil after HCl dissolution (Figure 9b).

355 The results indicate that there is a positive correlation between the increase in thermal
356 conductivity (Δk) and the amount of $CaCO_3$ precipitation in the soil matrix. The maximum Δk after
357 bio-cementation was 0.44 W/mK, representing a 17% increase in thermal properties, for 9.7%
358 $CaCO_3$ in this study. The results of this study are consistent with those obtained by Martinez et al.
359 (2019) for silica sand with a mean particle size of 0.26 mm, where an increase of 0.44 W/mK was
360 observed for 6.72% increase in $CaCO_3$ content. Direct comparison of the results with those obtained
361 by Xiao et al. (2021) and Wang et al. (2020) is challenging because different testing methods were
362 used. Xiao et al. (2021) utilized a hot disk thermal analyzer through a single-sided heat-transfer
363 process, while Wang et al. (2020) used a dual probe heat pulse (DPHP) sensor. In all cases,

however, an increase in $CaCO_3$ content results in a significant increase in Δk, indicating that MICP treatment could effectively enhance the thermal conductivity of sand with fines content.

Conclusion

A series of column experiments was conducted to investigate the effect of fines content on bio-cementation process and thermal properties of bio-cemented silty sand. Five soil columns were subjected to MICP treatment with alternating injection method, resulting in a similar cementation pattern within the columns for all specimens with higher concentration at the injection port and lower concentration at the middle of the column. Measurements from effluent sampling and post-treatment acid digest methods showed less mineral with increasing fines content. The average calcium carbonate content ranged between 7.2% for F15-16P with 15% fines content and 10.7% for F00-32P with 0% fine. This was expected as the samples with higher silt content have lower pore volume and received lower amount of injection fluids. For all specimens, the amount of precipitated $CaCO_3$ gradually decreased as the distance from the top and bottom injection ports increased. This was attributed to the filtration of bacterial cells through the soil particles, resulting in a higher cementation near the injection points. The effect of filtration became more prominent as the fines content in the specimens increased.

Measurements of thermal conductivity were obtained prior to and during MICP treatment. Thermal conductivity increased with the number of pulses for all specimens compared to the control specimen. Under saturated and untreated conditions, a 15% increase in fines content resulted in an approximately 11% increase in thermal conductivity. This increase in thermal conductivity is expected to continue until the sand-silt granular mixture reaches its maximum dry mass density. The difference in thermal conductivity between samples F00 and F15 was reduced by 1.8 times after MICP treatment, compared to their initial saturated state. In the untreated dry and initial

conditions, the specimen with 15% fines (F15) had over 20% higher conductivity than the specimen without fines (F0), likely due to improved particle packing. However, when saturated and treated with MICP, F15 had only a 4% higher conductivity than F0, suggesting that the effect of fines content on thermal conductivity can be mitigated by increasing the number of pulses. This observed behavior was attributed to the reduction in porosity resulting from $CaCO_3$ precipitation. It appears that the increased filtration effect resulting from reduced pore size in the presence of fine particles was found to have a negligible impact on the effect of bio-cementation on the thermal conductivity of the soil.

The results presented herein suggest that MICP treatment can be a viable option to increase the thermal conductivity of soils within the range of fines content studied here (less than 15%), which can enhance the efficiency of geothermal systems.

Data availability statement

Some or all data, models or code generated or used during the study are available upon reasonable request.

Acknowledgement

The authors appreciate the assistance of the staff of the center for biofilm engineering (CBE) at MSU.

References

Barkouki, T. H. , B. C. Martinez, B. M. Mortensen, T. S. Weathers, J. D. De Jong, T. R. Ginn, N. F. Spycher, R. W. Smith, and Y. Fujita. 2011. 'Forward and Inverse Bio-Geochemical Modeling of Microbially Induced Calcite Precipitation in Half-Meter Column Experiments', 90.

Beziat, A., Dardaine, M., Mouche, E. 1992. Measurements of the thermal conductivity of clay-sand and clay-graphite mixtures used as engineered barriers for high-level radioactive waste disposal. Applied clay science, 6(4), 245-263.

411 Cheng, L., R. Cord-Ruwisch, and M.A. Shahin. 2013. 'Cementation of sand soil by microbially
412 induced calcite precipitation at various degrees of saturation', *Canadian Geotechnical Journal*,
413 50: 81-90.
414 Chou, Chiung-Wen, Eric Seagren, A. Asce, Ahmet Aydilek, M. Asce, and Michael Lai. 2011.
415 'Biocalcification of Sand through Ureolysis', *Journal of Geotechnical and Geoenvironmental*
416 *Engineering*, 137.
417 DeJong, Jason T., Michael B. Fritzges, and Klaus Nüsslein. 2006. 'Microbially Induced
418 Cementation to Control Sand Response to Undrained Shear', *Journal of Geotechnical and*
419 *Geoenvironmental Engineering*, 132: 1381-92.
420 DeJong, Jason T., Brina M. Mortensen, Brian C. Martinez, and Douglas C. Nelson. 2010. 'Bio-
421 mediated soil improvement', *Ecological Engineering*, 36: 197-210.
422 Ebigbo, A., A. Phillips, R. Gerlach, R. Helmig, A. B. Cunningham, H. Class, and L. H. Spangler.
423 2012. 'Darcy-scale modeling of microbially induced carbonate mineral precipitation in sand
424 columns', *Water Resources Research*, 48.
425 Feng, K. , and B. M. Montoya. 2016. 'Influence of Confinement and Cementation Level on the
426 Behavior of Microbial-Induced Calcite Precipitated Sands under Monotonic Drained Loading',
427 *Journal of Geotechnical and Geoenvironmental Engineering*, 142: 04015057.
428 Foppen, J. W., and J. F. Schijven. 2006. 'Evaluation of data from the literature on the transport and
429 survival of Escherichia coli and thermotolerant coliforms in aquifers under saturated
430 conditions', *Water Res*, 40: 401-26.
431 Ginn, Timothy R., Brian D. Wood, Kirk E. Nelson, Timothy D. Scheibe, Ellyn M. Murphy, and T.
432 Prabhakar Clement. 2002. 'Processes in microbial transport in the natural subsurface', *Advances*
433 *in Water Resources*, 25: 1017-42.

Gunyol, P., M. Khosravi, A. Phillips, and K. Plymesser. 2022. 'Thermal Properties of Bio-Cemented Sand.' in, *Geo-Congress 2022*.

Harkes, Marien P, Leon A Van Paassen, Jacco L Booster, Victoria S Whiffin, and Mark CM van Loosdrecht. 2010. 'Fixation and distribution of bacterial activity in sand to induce carbonate precipitation for ground reinforcement', *Ecological Engineering*, 36: 112-17.

Hommel, Johannes, Arda Akyel, Zachary Frieling, Adrienne Phillips, Robin Gerlach, Alfred Cunningham, and Holger Class. 2020. 'A Numerical Model for Enzymatically Induced Calcium Carbonate Precipitation', *Applied Sciences*, 10: 4538.

Ismail, Mostafa, H. A. Joer, Mark Randolph, and A. Meritt. 2002. 'Cementation of porous materials using calcite', *Géotechnique*, 52: 313-24.

Jung, D., Biggs, H., Erikson, J., Ledyard, P.U. 1975. New colorimetric reaction for end-point, continuous-flow, and kinetic measurement of urea. Clinical Chemistry, 21(8), 1136-1140.

Kirkland, Catherine M., Arda Akyel, Randy Hiebert, Jay McCloskey, Jim Kirksey, Alfred B. Cunningham, Robin Gerlach, Lee Spangler, and Adrienne J. Phillips. 2021. 'Ureolysis-induced calcium carbonate precipitation (UICP) in the presence of CO2-affected brine: A field demonstration', *International Journal of Greenhouse Gas Control*, 109: 103391.

Lin, H., M.T. Suleiman, D. G. Brown, and E. Kavazanjian. 2016. 'Mechanical Behavior of Sands Treated by Microbially Induced Carbonate Precipitation', *Journal of Geotechnical and Geoenvironmental Engineering*, 142: 04015066.

Martinez, A., L. Huang, and M. G. Gomez. 2019. 'Thermal conductivity of MICP-treated sands at varying degrees of saturation', *Géotechnique Letters*, 9: 15-21.

455 Martinez, Alejandro, Lin Huang, and Michael Gomez. 2020. 'Enhancement of the thermal
456 conductivity of sands via microbially-induced calcite precipitation', *E3S Web of Conferences*,
457 205: 09011.
458 Mitchell, Andrew C., Adrienne Phillips, Logan Schultz, Stacy Parks, Lee Spangler, Alfred B.
459 Cunningham, and Robin Gerlach. 2013. 'Microbial CaCO3 mineral formation and stability in
460 an experimentally simulated high pressure saline aquifer with supercritical CO2', *International*
461 *Journal of Greenhouse Gas Control*, 15: 86-96.
462 Montoya, B. M., and J. T. DeJong. 2015. 'Stress-Strain Behavior of Sands Cemented by
463 Microbially Induced Calcite Precipitation', *Journal of Geotechnical and Geoenvironmental*
464 *Engineering*, 141: 04015019.
465 Phillips, A.J. 2013. Biofilm-induced calcium carbonate precipitation application in the subsurface,
466 Bozeman, Montana : Montana State University. Bozeman, Montana.
467 Phillips, Adrienne J, Alfred B Cunningham, Robin Gerlach, Randy Hiebert, Chiachi Hwang,
468 Bartholomeus P Lomans, Joseph Westrich, Cesar Mantilla, Jim Kirksey, and Richard Esposito.
469 2016. 'Fracture sealing with microbially-induced calcium carbonate precipitation: A field
470 study', *Environmental science & technology*, 50: 4111-17.
471 Phillips, Adrienne J., Robin Gerlach, Ellen Lauchnor, Andrew C. Mitchell, Alfred B. Cunningham,
472 and Lee Spangler. 2013. 'Engineered applications of ureolytic biomineralization: a review',
473 *Biofouling*, 29: 715-33.
474 Rebata-Landa, Verónica, and J. Santamarina. 2006. 'Mechanical limits to microbial activity in deep
475 sediments', *Geochemistry Geophysics Geosystems - GEOCHEM GEOPHYS GEOSYST*, 71.
476 Stocks-Fischer, S., Galinat, J.K., Bang, S.S. 1999. Microbiological precipitation of CaCO3. Soil
477 Biology and Biochemistry, 31(11), 1563-1571.

Van Paassen, Leon Andreas. 2009. 'Biogrout, ground improvement by microbial induced carbonate precipitation'.

Venuleo, Sara, Lyesse Laloui, Dimitrios Terzis, Tomasz Hueckel, and Mounir Hassan. 2016. 'Microbially induced calcite precipitation effect on soil thermal conductivity', *Géotechnique Letters*, 6: 39-44.

Wang, Zhaoyu, Nan Zhang, Jinhua Ding, Qi Li, and Junhao Xu. 2020. 'Thermal conductivity of sands treated with microbially induced calcite precipitation (MICP) and model prediction', *International Journal of Heat and Mass Transfer*, 147: 118899.

Whiffin, Victoria S., Leon A. van Paassen, and Marien P. Harkes. 2007. 'Microbial Carbonate Precipitation as a Soil Improvement Technique', *Geomicrobiology Journal*, 24: 417-23.

Xiao, Yang, Yifan Tang, Guoliang Ma, John S McCartney, and Jian Chu. 2021. 'Thermal conductivity of biocemented graded sands', *Journal of Geotechnical and Geoenvironmental Engineering*, 147: 04021106.

Yun, Tae, and J. Santamarina. 2008. 'Fundamental study of thermal conduction in dry soils', *Granular Matter*, 10: 197-207.

Zamani, A., and B. M. Montoya. 2018. 'Undrained Monotonic Shear Response of MICP-Treated Silty Sands', *Journal of Geotechnical and Geoenvironmental Engineering*, 144: 04018029.

Zamani, Atefeh. 2017. "Liquefaction Mitigation of Silty Sands with Microbial Induced Calcium Carbonate Precipitation." In.

Zhang, Nan, Xinbao Yu, Asheesh Pradhan, and Anand J. Puppala. 2015. 'Effects of Particle Size and Fines Content on Thermal Conductivity of Quartz Sands', *Transportation Research Record*, 2510: 36-43.