Effect of bacterial calcium carbonate precipitation on compressibility and shear strength of organic soil

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Received 22 October 2014; received in revised form 21 April 2015; accepted 22 May 2015
Available online 14 October 2015

Abstract

In the past few years, the use of bacterial calcium carbonate precipitation has become popular as a ground-improvement technique for sandy soil. However, this technique has not been applied to organic soil. This study focused on bacterial calcium carbonate precipitation and its effect on the compressibility and strength of organic soil. A special injection system was prepared for inducing a bacterial solution into several samples. The bacterial solution was supplied to the samples by gravity for 4 days in specific molds designed for this work. Calcite precipitation was observed by monitoring the changes in the pH value and by measuring the amount of calcium carbonate in the organic soil. The changes in compressibility and strength were measured before and after the bacterial treatment. The test results showed that the pH values in the treatment medium reached the ideal values that are appropriate for calcite precipitation. It was found that the amount of precipitated calcium carbonate in organic soil increased by about 20% in the treated samples compared to that in the untreated samples. Moreover, the test results indicated that the bacterial treatment influenced the compressibility and shear strength of the organic soil. The results were supported by an energy-dispersive x-ray (EDX) analysis.

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Keywords: Sporosarcina pasteurii; organic soil; Bacterial calcium carbonate precipitation; Compressibility; Strength

1. Introduction

Organic soils, which are found in many places around the world, are a mixture of finely divided particles of organic matter. In some instances, the soil may contain visible fragments of partly decayed vegetable matter and shells. The amount of organic matter in a soil significantly affects its geotechnical properties, including specific gravity, water content, liquid limit, plastic limit, density, hydraulic conductivity, compressibility, and strength. Constructing structures on an organic ground involves the risk of bearing capacity failure and excessive settlement. In order to prevent these risks, the geotechnical properties of organic soil are improved by ordinary improvement techniques, such as deep mixing with cement or lime, vertical drains, sand columns, and dry jet mixing (Hampton and Edil, 1998; Edil, 2003; Çelik and Canakci, 2011; Jelisic and Leppanen, 2003; Koda et al., 1989; Furstenberg et al., 1983; Yang et al., 1998).

In the past few years, the use of bacterial calcium carbonate precipitation (BCCP) has become popular as a ground-
improvement technique. It is presented as a new and environmentally friendly method (Dejong et al., 2006). This new method has an advantage over conventional chemical treatments, which can be toxic and environmentally harmful and have a limited injection distance (Karol, 2003). The method is also cost-effective in comparison to chemical treatments (Ivanov and Chu, 2008). Many studies in the literature have reported that the microbial-induced calcite precipitation technique is very effective in increasing the shear strength and in decreasing the permeability of sandy and gravelly soil (Van Paassen et al., 2010b; Van Wijngaarden et al., 2010; Van der Star et al., 2011). Scaled-up studies were also performed on sandy soil and successful results were obtained (Van Paassen et al., 2010b).

The main role of bacteria in the calcite precipitation process has been to generate an alkaline environment through different physiological actions (Douglas and Beveridge, 1998; Dejong et al., 2010; Benini et al., 1999; Ciurli et al., 1999; Karatas et al., 2008; Van Paassen et al., 2010a; Hamdan et al., 2011b; Warthmann et al., 2000; Roden et al., 2002; Weaver et al., 2011). *Bacillus pasteurii*, a highly urease active bacteria, plays an important role in CaCO₃ precipitation (Bang et al., 2001; Dejong et al., 2006; Dejong et al., 2010). It should be noted that *B. pasteurii* has been reclassified as *Sporosarcina pasteurii* (Mitchell and Ferris, 2006). As can be seen in Table 1, it exhibits a high urease production. Hence, it has been used for microbial cementation in many studies (Bachmeier et al., 2002; Sarda et al., 2009).

The main nutrient solution that is commonly used to provide the necessary nutrition for the bacteria, as well as the chemical compounds that are needed for soil cementation, contain NaHCO₃, NH₄Cl, CaCl₂, urea, and a nutrient broth (mixture of peptone, yeast extract, beef extract, and NaCl). Under favorable environmental conditions, *S. pasteurii* uses urea as an energy source, producing ammonia (NH₃) and carbon dioxide (CO₂), which tend to increase the pH in the proximal environment. The net increase in pH causes the calcium ions to react with the carbonic acid and hydroxide ions to form calcium carbonate bonds (Dejong et al., 2010). The chemical reaction is given in Eq. (1). The calcite bonds tend to form at the particle-to-particle contacts in the soil matrix, which has an overall cementing effect between soil particles.

\[
\text{Ca}^{2+} + \text{HCO}_3^- + \text{OH}^- \rightarrow \text{CaCO}_3 + \text{H}_2\text{O} \tag{1}
\]

Bacterial calcium carbonate precipitation or cementation has been applied to a variety of civil engineering applications, such as to repair cracks in rock and concrete, to improve the bearing capacity, to reduce permeability, to increase dilative tendencies, and to increase the strain stiffness in sand (Gollapudi et al., 1995; Ramachandran et al., 2001; Bang et al., 2001; Ramakrishnan, 2007; Jonkers et al., 2009; Lo Bianco and Madonia, 2007; Dejong et al., 2006; Whiffin et al., 2007; Ivanov and Chu, 2008; Al Qabany, 2011; Rusu et al., 2011; Chou et al., 2011; Mortensen and Dejong (2011); Al Qabany and Soga, 2013; Martinez et al., 2013; DeJong, 2013).

Gollapudi et al. (1995) used microbial mineral plugging to reduce the porosity of rock fractures. A series of column tests was conducted in order to evaluate the effectiveness of the proposed microbial technique. The flow rate into the fractures via the column was measured during the tests, and the absence of the flow signified the completion of plugging. Their results showed that microbial plugging was highly related to bacterial concentration, pH, the flow rate in the fractures, and the presence of contaminants.

Dejong et al. (2006) demonstrated that microbially cemented specimens exhibited increases in the strength of sandy soil. Their results showed that the behavior of the bacteria-ically cemented specimens was similar to that of gypsum-cemented soil.

Whiffin et al. (2007) examined the effect of microbial carbonate precipitation on the permeability and shear strength of sandy soil. In their study, a 5-meter sand column was used to simulate field conditions. The results from triaxial tests conducted on specimens from the treated sand column showed that the soil porosity, strength, and stiffness were all significantly affected by the calcium carbonate content.

Chou et al. (2011) conducted a laboratory study to evaluate the effect of growing, dead, and resting cells on the geomorphological properties of microbially cemented sand. They performed direct shear and California Bearing Ratio (CBR) tests on sand specimens and found that the bacterial cells effectively improved the geomorphological properties of the sand. An analysis of the sand from CBR specimens treated with growing cells demonstrated that the microbial processes contributed to the clogging of the porous medium.

Martinez et al. (2013) worked on optimizing MICP treatment by varying the procedural parameters, including the flow rates, the flow direction, and the formulations of the biological and chemical amendments. They monitored the physical, chemical, and biological properties essential to the performance of MICP, including shear wave velocity, permeability, calcium carbonate content, aqueous calcium, aqueous ammonium, aqueous urea, and bacterial density. Their experiments showed that the shear wave velocity of treated soil increased from 140 m/s to an average of 600 m/s.

Al Qabany and Soga (2013) conducted unconfined compressive strength and permeability tests on sand samples treated with 0·1, 0·25, 0·5, and 1 M urea–calcium chloride solutions. The MICP treatment increased the strength of the treated samples. The magnitude of this increase depended on the concentration used in the treatment; the use of a low-urea calcium-chloride solution resulted in stronger samples. The use of a high-urea calcium-chloride-concentration solution resulted in a rapid drop in permeability at the early stage of the calcite precipitation, whereas the use of a low-chemical-concentration solution was found to result in a more gradual and uniform decrease in permeability.
In all previous studies, the BCCP technique was used in an attempt to improve the geotechnical properties of granular soil and successful results were obtained. The information in the literature on the application of BCCP to organic soil and its effect on compressibility and strength is very limited. This study focuses on the investigation of the feasibility of calcite precipitation and its effect on the strength and compressibility of organic soil. _S. pasteurii_, a type of non-pathogenic organism found naturally in soil, was used throughout the study.

2. Materials and methods

2.1. Materials

An isolated bacterial culture of _S. pasteurii_ NCIMB 8221 was used in this study. The organic soil was obtained from the Sakarya region of Turkey. This soil is classified as OH “sandy organic silt” according to the Unified Soil Classification System (USCS), and as peat according to Wüst et al. (2003). Chemical and physical analyses were carried out on organic soil and the results are presented in Table 2. In all the tests, organic soil samples that passed through sieve No. 2 mm and remained in sieve No. 100 (0.15 mm) were used. This range was chosen in order to provide enough space for the bacteria to have a cell diameter usually in the range of 0.5–3 μm (Madigan and Martinko, 2003). Moreover, the permeability of the organic soil was measured and found to be 5.2 × 10⁻³ cm/s. The soil was placed in a firing oven at 440°C for 4 h, and the organic content was estimated according to the ASTM D 2974 standard. According to the ASTM D 2974 standard, the ash content of the soil was 40%. A wet sieve analysis of the ash showed that the soil contained 15% silt and clay, 25% sand, and 60% organic materials. The liquid limit of the organic soil was estimated by fall cone tests and found to be 125%.

2.2. Methods

2.2.1. Preparing of urea nutrient agar, bacteria culturing, and counting

Urea nutrient agar was generally used to cultivate all the Bacillus spp. that had the ability to hydrolyze the urea. Table 3 shows the solid and liquid contents of the media. To prepare this media, the solid components were added to distilled water to bring the volume to 1 L and mixed well. The mixture was then gently heated, and brought to a boiling temperature. Later, it was autoclaved for 15 min at 15 psi pressure at 121°C. After autoclaving, it was cooled to 50–55°C. A total of 50 ml of sterile urea solution was added to the mixture and mixed well. The urea nutrient agar medium was then immediately poured into approximately 20 culture plates under a laminar flow hood. The lamenar flow hood provided filtered air to reduce the risk of contaminating the culture growth media prior to the introduction of the _S. pasteurii_ cultures.

One loopful of pure colony was taken from the stock bacteria and streaked onto each culture plate. All the plates were then inverted and incubated for 48–72 h at 30°C in an incubator. After incubation, the colony growth of _S. pasteurii_ had occurred for each of the single resulting colonies transferred from the plate to a 250 ml culture flask containing 200 ml urea nutrient broth. The culture flask was then wrapped in cheesecloth to filter the atmospheric biocontaminants, while providing oxygen to the bacteria, and incubated for 48 hours at 30°C (Lo Bianco and Madonia, 2007). After incubation, 40-ml batches of the bacteria culturing solution were transferred from the culture flask to a series of 50-ml culture tubes. Each culture tube was centrifuged at 3000 rpm for 20 min to separate the suspension from the supernatant. The supernatant was removed by pouring it into a separate flask, and the remaining bacterial pellet was then used for the bacterial treatment process that was applied to each soil specimen.

The instrument DensiCHEK Plus 21255-P1ML1 was used to measure the optical density of the microorganism suspension. The instrument provides values in McFarland units, proportional to the microorganism concentrations. The device was indicated for use with polystyrene and glass test tubes and the reading range was 0.0–4.0 McFarland (approx. cell density of (1.5 × 10⁸–12 × 10⁸ CFU/ml)) at a wavelength of 600 nm. It generated a McFarland value using basic colourimetry, which is a method of measurement that relates the amount of color in a transparent medium (liquid) to the amount of a particular substance in the liquid. In general, the concentration of the substance being measured is proportional to the intensity of the color of the solution. The darker the color is, the higher the concentration.

Table 3
Details of solid and liquid contents of the media.

<table>
<thead>
<tr>
<th>Composition</th>
<th>Quantity</th>
</tr>
</thead>
<tbody>
<tr>
<td>Agar</td>
<td>15 g</td>
</tr>
<tr>
<td>Peptone</td>
<td>5 g</td>
</tr>
<tr>
<td>NaCl</td>
<td>5 g</td>
</tr>
<tr>
<td>Yeast extract</td>
<td>2 g</td>
</tr>
<tr>
<td>Beef extract</td>
<td>1 g</td>
</tr>
<tr>
<td>Urea solution*</td>
<td>50 mL</td>
</tr>
</tbody>
</table>

*Urea solution (add 20 g of urea to distilled water and bring the volume to 100 mL. Mix well. Filter and sterilize).

Table 2
Engineering properties of the organic soil used in the study.

<table>
<thead>
<tr>
<th>Properties of organic soil</th>
<th>Content (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Organic content (%)</td>
<td>60</td>
</tr>
<tr>
<td>pH</td>
<td>6.5</td>
</tr>
<tr>
<td>Natural water content (%)</td>
<td>256</td>
</tr>
<tr>
<td>Liquid limit (%)</td>
<td>125</td>
</tr>
<tr>
<td>Plasticity index (%)</td>
<td>NP</td>
</tr>
<tr>
<td>Specific gravity (g/cm³)</td>
<td>1.97</td>
</tr>
<tr>
<td>CaCO³</td>
<td>0</td>
</tr>
<tr>
<td>NO₃–N (ppm)</td>
<td>50–150</td>
</tr>
<tr>
<td>NH₄–N (ppm)</td>
<td>40–140</td>
</tr>
<tr>
<td>K (ppm)</td>
<td>0–20</td>
</tr>
<tr>
<td>P (ppm)</td>
<td>0.2–0.7</td>
</tr>
</tbody>
</table>
2.2.2. Bacterial treatment solution preparation
To prepare the bacterial treatment solution, urea medium was first created. Nutrient broth (3 g), urea (20 g), NH4Cl (10 g), NaHCO3 (2.12 g), and 500 ml of distilled water, as shown in Table 4, were mixed to create the urea medium solution. Each of the solid ingredients was mixed thoroughly in 500 mL of distilled water until it had dissolved, and the pH of the resulting urea medium solution was adjusted to 6.0 with the use of 5 N HCl prior to autoclaving. Distilled water was then added to reach the final required volume (1 L). After autoclaving, the pH of the urea medium was measured to be 7.0. The pH value of the resulting 1 L of solution was adjusted by aerating the solution to increase the initial pH value from 7.0 to 8.0. In this way, the solution became suitable for biological activity for bacteria. This 1 L of aerated solution was divided into two approximate portions of 800 ml and 200 ml. Spun S. pasteurii cells were added to the 800-ml portion of the aerated urea medium and the flask was gently agitated to re-suspend the cells. A 20-ml volume of calcium chloride solution (18.5 g CaCl2/100 ml distilled water) was then added to the aerated urea (200 ml), which lowered the pH of the solution slightly. The re-suspended bacterial solution was then added to the urea–calcium chloride solution. Five hundred milli litre of the combined solution consisting of urea, calcium chloride, and S. pasteurii cells was injected quickly into the specimens by gravity at a flow rate of approximately 20 ml/min and a hydraulic head of 1 m to prevent the calcium carbonate from precipitating in the pore fluid due to the simultaneous introduction of bacteria and calcium. This initial “biological treatment” was then allowed to set for a minimum of 12 hours to allow the microbes to attach to the particles of the soil samples. After 6 hours, 500-ml nutrient treatments of urea and CaCl2 were injected through the specimens at the same flow rate. This nutrient treatment process was then periodically repeated (every 6 hours) for 4 days.

2.2.3. Sample preparation
One control sample (untreated 1) and three bio-treated samples (bio-treated 1, bio-treated 2, and bio-treated 3) were prepared for consolidation tests in cylindrical molds having a diameter of 75 mm and a height of 20 mm, as shown in Fig. 1. In addition, one control sample (untreated 2) and three bio-treated samples (bio-treated 4, bio-treated 5, and bio-treated 6) were prepared for direct shear tests in box molds having dimensions of 60 × 60 × 20 mm3, as shown in Fig. 2. All the soil samples were placed into the oven for 30 min. at 80 °C to destroy any micro-organisms that may affect the calcium carbonate precipitation process. After that, the organic soil was filled into the molds in a loose state having a dry density equal to 0.6 and 0.69 g/cm3 for the consolidation tests and the direct shear tests, respectively. Each end of the samples was fitted with a filter paper, as shown in Fig. 1. The samples were initially treated by Bacterial media (S. pasteurii, urea, and CaCl2) and then allowed to set for a minimum of 12 hours to allow the microbes to attach to the particles of the soil samples. Then, every 6 hours, nutrient treatments consisting of urea and CaCl2 were injected through the specimens at the same flow rate of approximately 20 ml/min at a hydraulic head of 1 m. This nutrient treatment process was then periodically repeated for 4 days. During the treatment periods, the samples were incubated at 28 °C in a room prepared for this study. After each treatment period, the samples were cured at a room temperature of 25 °C for 5 days, and then consolidation and direct shear tests were performed on the samples. Throughout the treatment periods, the changes in the pH values were monitored at different time intervals. Also, at the end of all the
tests, the amounts of calcium carbonate in the samples were determined by a calcimeter instrument.

2.2.4. PH measurement
Changes in the pH values were monitored to determine the presence of biological activity (8.3–9.3) in the soil samples. The effluents of the samples were measured using a pH meter as a reassurance of calcite precipitating in the soil samples.

2.2.5. Strength and compressibility tests
The effect of BCCP on the compressibility and shear strength was determined by performing direct shear tests and one-dimensional consolidation tests. The consolidation tests were done according to ASTM D2435 for determining the magnitude and the rate of consolidation of the soil when it was restrained laterally and drained axially while being subjected to incrementally applied controlled-stress loading (11, 22, 45, 90, 180, and 360 kPa) and then unloading (180, 90, 45, 5, and 0 kPa). After setting the soil sample in the consolidation cell and applying each controlled-stress loading, the dial readings were taken for a period of 24 h at a range of times (0, 0.25, 0.5, 1, 2, 4, 8, 16, 30, 60, 120, 240, 480, 960, and 1440 min).

Direct shear tests were carried out according to ASTM D3080. This test method covers the determination of the consolidated drained (CD) shear strength of one specimen of a soil material under direct shear boundary conditions. CD means that shear was not started until after the settlement resulting from the applied normal load had stopped, and the shearing force was applied so slowly that no pore pressure developed in the sample. The specimen was deformed at a constant rate of displacement (0.02 mm/min) to make sure that the excess pore water pressure dissipated during the shearing process (Morgenstern and Tchalenko, 1967; Dyvik et al., 1987). Direct shear tests were performed under various normal stresses (13.6, 20.4, and 27.2 kPa).

2.2.6. Calcium carbonate content measurement
In order to measure the amount of precipitated calcium carbonate, a calcimeter was used (Calcimeter Bernard). For this purpose, 1 g of a soil sample was treated with hydrochloric acid (HCL) in an enclosed reactor vessel. The basic principle for determining the amount of CaCO3(%) is based on the volumetric analysis of the carbon dioxide CO2 gas, which evolves during the reaction between the acid solution (HCl-4N) and the carbonate fraction of the soil specimen, as follows:

\[
\text{CaCO}_3 + 2\text{HCl} \rightarrow \text{CaCl}_2 + \text{H}_2\text{O} + \text{CO}_2 \uparrow
\]  

(2)

The resulting pressure generated in the closed reactor is directly proportional to the carbonate content of the specimen. This pressure was measured with a bourdon tube pressure gauge, which was pre-calibrated with reagent grade CaCO3 as

\[
\text{CaCO}_3(\%) = \frac{[(1000 \times m_2 \times (V_1 - V_3)) / (m_1 \times (V_2 - V_3))] \times [(100 + W)/100]} {2HCl2 + CaCl2 + H2O + CO2} \]

(3)

3. Results and discussion

3.1. pH values in treated organic soils

When the organic soil samples were treated with the bacterial solution (bacteria, urea, and CaCl2), the pH values were monitored to determine the presence of biological activity within the soil samples. Fig. 3 depicts the changes in the pH values over time. As can be seen in the figure, the pH values reached around 9.3 for organic soil samples after 12 h. According to Hammes and Verstraete (2002), the pH value was a key parameter in the CaCO3 precipitation. A local rise in pH often causes the bacteria to serve as nucleation sites for crystallization. The decomposition of urea provides a high pH environment (Dejong et al., 2006). In the literature, the ideal range in pH values for the bacteria to precipitate calcite was reported to be between 8.3 and 9.3 (Stocks et al., 1999; Dejong et al. 2010). In the present study, the experimental results showed that the BCCP technique was applicable to organic soil.

3.2. Effect of BCCP on consolidation

Consolidation tests were undertaken using a conventional odometer device. The study focused on the primary consolidation behavior of untreated and treated organic soil samples. Four samples (untreated 1, bio-treated 1, bio-treated 2, and bio-treated 3) were prepared in a loose state having a dry density of 0.6 g/cm3. The loose state was selected to show the effect of calcium carbonate precipitation on the compressibility behavior. The void ratios versus the log pressure curves of the samples are shown in Fig. 4. It is clear from these curves that the variations in void ratios with respect to entire stress levels are similar among the three bio-treated organic soil samples. This indicates that the treatment process used in this study is
repeatable and consistent. Fig. 4 also shows the apparent difference in compressibility behavior between the treated and untreated samples. For instance, the change in void ratio for the treated samples was less than that for the untreated sample. Fig. 4 shows that the BCCP-treated samples have two consolidation yield stresses. On the other hand, the control sample has unique consolidation yield stress. It is believed that the BCCP-treated sample reaches its first consolidation yield point after breakage of the bond between the calcite crystals and the organic soil particles beyond 11 kPa. After this stress level, the solid calcite particles that filled the voids in the organic soil continue to take more stress with very little deformation until 45 kPa that seems to be the second consolidation yield stress. The untreated soil reaches its consolidation yield stress at nearly 11 kPa.

The coefficient of compressibility ($m_v$) is generally used to calculate the vertical settlement of a soil layer having an initial thickness that is subjected to an increase in vertical stress. Fig. 5 shows the variation in $m_v$ under different levels of consolidation stress. The figure indicates that the $m_v$ values were higher for the untreated sample than for the treated samples. This indicates that the compressibility of the treated samples decreased when the voids in the organic soil filled with solid calcite particles that also caused cementation between the organic soil particles. After high stress levels, the coefficient of compressibility values for the untreated and treated soil samples became the same.

The compression index ($C_c$) and compression ratio $C/c/(1 + e_0)$ of the treated and untreated samples were calculated. Table 5 shows the change in these values between stress levels of 11 kPa and 45 kPa. It is seen from the table that, at this range in stress levels, the compression index and the compression ratio of the treated samples were less than those of the untreated sample. On the other hand, beyond this range in stress levels, $C_c$ and $C/c/(1 + e_0)$ were similar for both the treated and untreated samples. The reduction in $C_c$ and $C/c/(1 + e_0)$ indicates that BCCP improves the compressibility behavior of the organic soil.

Fig. 6 and Table 6 show the square root of time vs the deformation graph and time for reaching the end of primary consolidation ($t_{90}$) for the treated and untreated samples for different stress levels. It can be noted from the graph that the end of the primary consolidation of the treated samples is longer than that of the untreated one. This increase can be attributed to the clogging effect of the BCCP process (Whiffin et al., 2007). The clogging of the pore spaces with the calcite particles reduced the dissipation of the water from the soil. This was supported by the permeability values of the treated and untreated samples derived from the oedometer tests. The permeability of the untreated sample was $5.2 \times 10^{-3}$ cm/s and that of the treated samples was $4.5 \times 10^{-4}$ cm/s. Fig. 7 shows the changes in the coefficient of consolidation ($c_v$) with pressure. The effect of the calcite precipitation is clearly seen between the treated and untreated soil samples. For the same stress levels, the $c_v$ values for the treated samples are always less than those for the untreated one. This indicates that the BCCP process reduces the primary consolidation, but that the time for the completion of the primary consolidation increases.

![Fig. 4. Void ratio versus stress of treated and untreated organic soil samples.](image)

![Fig. 5. Coefficient of compressibility of treated and untreated organic soil samples.](image)

**Table 5**

<table>
<thead>
<tr>
<th>Sample</th>
<th>$C_c$</th>
<th>$C/c/(1 + e_0)$</th>
</tr>
</thead>
<tbody>
<tr>
<td>Untreated 1</td>
<td>0.6863</td>
<td>0.1966</td>
</tr>
<tr>
<td>Bio-treated 1</td>
<td>0.4482</td>
<td>0.1284</td>
</tr>
<tr>
<td>Bio-treated 2</td>
<td>0.5047</td>
<td>0.1446</td>
</tr>
<tr>
<td>Bio-treated 3</td>
<td>0.4567</td>
<td>0.1308</td>
</tr>
</tbody>
</table>

![Fig. 6. Square root of time versus deformation for treated and untreated samples.](image)
3.3. Effect of BCCP on shear strength

The shear strength of a soil is defined as the ultimate or the maximum shear stress that a soil can sustain. Cohesion and the angle of internal friction are the two inherent shear strength parameters of a soil. In order to compare these inherent properties of the treated and untreated soils, four samples (untreated 2, bio-treated 4, bio-treated 5, and bio-treated 6) were tested in the direct shear apparatus. Fig. 8 shows the shear stress versus horizontal displacement under different normal stress levels. It can be seen from the figure that the shear stress levels of the treated samples are higher than those of the untreated soil samples. Although the soil samples were prepared in their loose state, it was observed that the treated soil sample behaves like dense granular soils under low normal stress, as shown in Fig. 9. This may be attributed to the cementation of the soil particles by the BCCP process. Under high normal stress, the calcite bonds between the soil particles were broken; therefore, peak shear stress was not observed. The same behavior was also seen in the triaxial tests performed on the bio-cemented sand samples under high back pressure (Ayse, 2010).

Cohesion and the angle of internal friction were estimated at a 2-mm deformation where peak shear stress was observed in the treated soil sample, and 6-mm horizontal displacement was the maximum horizontal displacement attained in the tests. It can be seen from Table 7 that the cohesion values of the untreated sample are 8.4 kPa and 5.8 kPa at horizontal displacements of 2 mm and 6 mm, respectively. These values are consistent with the literature (Edil and Wang, 2000; Huat et al., 2009). The internal friction angles of the untreated soil sample are 30° and 43° for horizontal displacements of 2 mm and 6 mm, respectively. These values are within the range reported by Mesri and Ajlouni (2007) and Landva and La Rochelle (1983). The wide range in internal friction angles of organic soil is attributed to the fiber content and its orientation (Mesri and Ajlouni, 2007). The test results showed that BCCP treatment changed the shear strength parameters of the organic soil. It is seen from Table 7 that the cohesion of the treated soil increased at a low displacement, but decreased at a high

Table 6
Values of square root of \( t_{90} \) under stress levels (11–180 kPa).

<table>
<thead>
<tr>
<th>Pressure (kPa)</th>
<th>Untreated sample</th>
<th>Treated sample</th>
</tr>
</thead>
<tbody>
<tr>
<td>11</td>
<td>2.1</td>
<td>3.2</td>
</tr>
<tr>
<td>22</td>
<td>1.9</td>
<td>2.5</td>
</tr>
<tr>
<td>45</td>
<td>1.8</td>
<td>2.1</td>
</tr>
<tr>
<td>90</td>
<td>1.5</td>
<td>1.9</td>
</tr>
<tr>
<td>180</td>
<td>1.35</td>
<td>1.9</td>
</tr>
</tbody>
</table>

Table 7
Shear strength parameters before and after BCCP at different horizontal displacements.

<table>
<thead>
<tr>
<th>Horizontal displacement (mm)</th>
<th>Cohesion (kPa)</th>
<th>Angle of internal friction (degree)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Untreated</td>
<td>Treated</td>
<td>Untreated</td>
</tr>
<tr>
<td>2</td>
<td>8.4</td>
<td>30</td>
</tr>
<tr>
<td>6</td>
<td>5.8</td>
<td>43</td>
</tr>
</tbody>
</table>
horizontal displacement. The increase in cohesion at a low displacement can be attributed to cementation between the organic soil particles. While low cohesion values at a high horizontal displacement can be explained by the breakage of the cementation bonds. This relative increase can be explained by additional friction between the solid calcite particles precipitated in the organic soil. The change in shear strength seems to be limited. This may be improved with further work.

Although calcite was precipitated in the organic soil, its strength was very different than that of the sand reported by Van Paassen et al. (2009). The cementation with calcium carbonate in sandy soil binds the solid sand particles. However, the same calcium carbonate binds soft organic soil particles. When stress is applied to the sand sample, failure occurs either through strong calcite or sand particles. Therefore, high compressive strength is obtained in MICP-treated sandy soil (Van Paassen et al., 2009). On the other hand, failure occurs through weak organic particles or bonds between strong calcite and weak organic soil particles. Therefore, low strength is obtained in treated organic soil. Hence, it can be concluded that the failure of organic particles governs the strength of bio-cemented organic soil. For this reason, the strength obtained from organic soil is less than that obtained from sandy soil with same amount of calcite. This is also true for concrete. The compressive strength is controlled by both the strength of the aggregate and the strength of the paste, and it depends on which of these two fails first. Studies reported in the literature have shown that concrete containing organic content has lower strength. This is attributed to the failure of the weak points of the concrete matrix that are organic particles (Basri, et al., 1999; Alengaram et al., 2008; Mannan and Ganapathy, 2004).

3.4. Amount of CaCO₃ in organic soil

At the end of consolidation and the direct shear tests, the samples were used to measure the amount of precipitated CaCO₃ in the organic soil. Fig. 10 shows the changes in the amount of CaCO₃ content in all the treated organic soil samples. It can be seen from this figure that the amount of CaCO₃ increased by around 20% in the organic soil. This demonstrates that CaCO₃ precipitation occurs in organic soil. However, the amount of CaCO₃ was comparatively less in the organic soil than in the sandy soil (Sidik et al., 2013). This difference in calcite precipitation in organic soil and sandy soil can be attributed to soluble organic ligands and other organic matter that are well-known inhibitors of CaCO₃ precipitation and crystal growth (Lebron and Suarez, 1998). Researchers have related this inhibition mechanism to a number of factors. According to Lin and Singer (2005a), when the organic molecules are absorbed onto a mineral surface, they can either induce dissolution or impair crystal growth, depending on the saturation conditions. Other studies have proposed that the organic matter content prevents CaCO₃ precipitation by coating the existing CaCO₃ crystal surfaces, thus blocking their nucleation sites and preventing homogeneous crystal growth (Inskeep and Bloom, 1986a; Lebron and Suarez, 1996, 1998; Lin and Singer, 2005, 2006).

SEM images of the organic soil before and after treatment are shown in Fig. 11(a–b). The images revealed CaCO₃ crystals on the surface and pores of the organic soil particles. Figs. 12 and 13 show the EDX spectrum before and after
Fig. 12. EDX analysis of organic soil before BCCP treatment.

Fig. 13. EDX analysis of organic soil after BCCP treatment.
treatment of the organic soil. Fig. 12 reveals that the soil sample before treatment consisted mainly of Si, K, Al, Fe, and Mg that are the main minerals in any soil. An EDX spectrum taken after treatment clearly shows that the main component in the soil is Ca compared to the other minerals, as shown Fig. 13. This indicates that CaCO₃ particles formed in the organic soil.

4. Conclusions

The laboratory-scale experimental results in this study proved that bacterial calcite precipitation is feasible for organic soil. The test results showed that the shear strength and the compressibility behavior of organic soil can be improved by this new technique. The preliminary attempt made in this work revealed that the BCCP method increased the shear strength and reduced the compressibility of the organic soil. The findings of the study should encourage geotechnical engineers to use the BCCP technique as a new and alternative improvement method for organic soil. However, the use of this technique with this soil remains to be quantified in future studies.

Acknowledgments

This work was supported by the Scientific Research Projects Governing Unit of the University of Gaziantep. Project no. MF.12.09. This study was also supported by the Iraqi Ministry of Higher Education and Scientific Research (Grant no. 01276).

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