

Bio-cementation for Slope Soil Stabilization against Surface Erosion: A Bench-scale Preliminary Investigation

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ABSTRACT: Surface instability of residual soil slope due to erosion is one of the widespread issues all over the world. Bio-cementation is a novel soil improvement method, wherein urease-producing bacteria or urease enzyme can be used to bind sand particles through calcite formation, enhancing the engineering properties of soil. This research work aims to assess the feasibility of bio-cementation technique as a potential alternative for surface stabilization of the slopes. A residual slope soil (Hokkaido, Japan) was instrumented in bench-scale slope for solidification test at the laboratory. Native ureolytic bacteria was enriched under optimum environmental conditions and injected to percolate through slope surface, followed by number of injections of cementation solution. The results indicated that relatively a stiff layer/ crust (6–9 MPa) with the thickness ranging from 3–4 cm is found along the slope surface. However, the UCS values decreased with the depth of the slope. The hydraulic conductivity of the slope soil reduced from 1.9×10^{-2} cm/s to 1.85×10^{-3} cm/s (by 90%) due to the cementation, which could inhibit the excess infiltration of rainwater and enhance the surface runoff.

1. INTRODUCTION

Studies by several investigators have demonstrated that residual soil slope failures can be attributed to several factors such as climatic conditions, geological features, soil characteristics, topography, vegetation level and/ or combination of the above (Rahardjo et al., 2005; Morbidelli et al., 2018; Zhang et al., 2018). Residual soils are generally unsaturated, and most of the failures in residual soil slopes occurs during or soon after the heavy rainfall (Rahardjo et al., 2005). Rainfall results infiltration and surface runoff in a slope structure. Infiltration of rainwater into the slope is governed by rainfall intensity and soil infiltrability (Oh and Vanapalli, 2010). The rate of infiltration is relatively high in the early stages of rainfall and decreases with the time; and once the surface zone reached its saturated state, the ponding or runoff starts occurring over the slope surface.

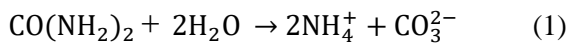
Typically, partially saturated/ unsaturated residual soils experience high matric suction due to negative pore water pressure effect, that contributes to high shear strength in soil matrix compared to that in fully saturated state. The infiltration of excess rainwater into the slope may increase

the water content, and this in turn results in a decrease in the matric suction and effective stress. As a result, additional shear strength afforded by the matric suction would be reduced enough to trigger the shallow slip failures and surficial sliding parallel to the slope surface (Oh and Vanapalli, 2010; Morbidelli et al., 2018).

Surface runoff erosion is another serious issue often reported by researchers. Surficial erosion is a process that sheet flow generated during rainfall scours the slope surface (Qing-quan et al., 2001). Soil loss on the slope occurs when the scouring capability of the surface runoff exceeds the erosion resisting capacity of the soil body. The erosion process can be considered in three steps. Firstly, as explained earlier, ponding results runoff over the surface under gravitational force and thus generates the surface sheet flow. Then, when the scouring ability exceeds the resisting capacity, the scouring of soil particles is induced. Finally, the scoured soil particles are transported downslope by overland flow. Those materials would be deposited in drainage systems and other ground surfaces that could obstruct or damage passengers and other properties (Omar et al., 2018).

Up to the date, a number of methods have been extensively studied and implemented to enhance the surficial stability of residual soil slopes, and some of them are as follows: vegetation cover, exotic grass implications, geosynthetic applications, mechanical compaction and chemical based stabilizations (e.g. cement, ashes, stabilizing additives and geopolymers) (Chirico et al., 2013; Daraei et al., 2018; Kumar and Das 2018). Although the chemical grouting is frequently utilized for erosion control and slope stability (Arya et al., 2017; Daraei et al., 2018), they could result many negative impacts including environmental (air, land and water) pollutions and influx of dangerous substances to the geo-environment. Also, due to deliberation on healthy future, current intentions of most of the researchers target on modifying the soil by eco-friendly and sustainable improvement techniques (Dilrukshi et al., 2016; Gowthaman et al., 2018). Therefore, in this study, recently developed bio-cementation technique (DeJong et al., 2010; Whiffin et al., 2014) is investigated as a potential candidate for slope soil stabilization against surface erosion.

Bio-cementation is a novel grouting technique, introduced recently in geotechnical engineering practice to minimize the environmental problems. The concept of bio-grouting is based on the ability of urease producing bacteria to precipitate calcium carbonate in the presence of urea and calcium sources (van Paassen et al., 2010; Whiffin et al., 2014). The technique involves two major biochemical reactions: (i) hydrolysis of urea by urease enzyme produced by the bacteria (Eq. (1)), (ii) precipitation of calcium carbonate in the presence of Ca^{2+} ions (Eq. (2)) (Cheng and Cord-Ruwisch, 2014; Amarakoon and Kawasaki, 2018). The formed calcium carbonate minerals act as cementing agents that bind the soil particles together, enhancing mechanical and geotechnical properties of soil matrix (DeJong et al., 2010).



Numerous studies have reported that settlement of bio-cemented sand has been significantly reduced (DeJong et al., 2010; van Paassen et al., 2010); unconfined compressive strength, shear strength and stiffness have been increased (Cheng and Cord-Ruwisch, 2014; Whiffin et al., 2014; Lin et al., 2016); permeability and erodibility have been decreased (Jiang and Soga, 2017; Omar et al., 2018). Moreover, impact of key environmental factors such as bacteria type, cell concentration, pH, degree of temperature, resources concentrations on the efficacy of bio-cementation have been extensively investigated, and deeper understanding regarding various aspects have been provided by researchers (Martinez et al., 2013; Danjo and

Kawasaki 2016; Cheng et al., 2017; Gowthaman et al., 2019a).

Most of the current exploitations of bio-cementation technology have been addressed in elementary-scale (sample ranges few centimeters), requiring upscaling of the system developed at the elementary scale for the practical and field implementations. This paper aims to address the bio-cementation technique at the bench-scale, demonstrating the feasibility of surface percolation method for treating unsaturated residual soil slope. Bench-scale experiments ($10^1\sim 10^2$ centimeters) are crucial because they are the intermediate step between elementary-scale and large-scale experiments. Although most of the optimization of biological, biochemical and geotechnical factors are performed in elementary scale experiments, bench-scale studies are still essential to enable optimization of the treatment formulation, injection procedures and treatment methods. Further, during the development stage, bench-scale studies would be more economically benefitable compared to the large-scale attempts which lead to high material and treatment costs (Gowthaman et al., 2019b). This work also aims to study the feasibility of applying segregated cell pellets of the bacteria for bio-cementation.

2. MATERIALS

2.1. Soil used

The soil used in the current study was well graded (SW based on Unified soil classification system/ USCS) sandy soil obtained from natural slope, Onuma, Hokkaido, Japan. The sandy soil is with 50% less than 0.25 mm in grain size.

2.2. Bacteria and culture medium

The urease active bacteria used in this current work were *Lysinibacillus xylanilyticus* isolated from the native soil. The isolation process was the same that explained clearly in the previous work (Gowthaman et al., 2019a). The bacteria were cultivated in a sterile aerobic batch growth medium consisting 20 g/L yeast extract, 17.5 g/L tris buffer and 10 g/L ammonium sulfate (NH₄-YE medium) with a pH value of 7.0~7.2. After 48 hours of incubation at 25°C, the optical density (OD₆₀₀) of the harvested culture medium was adjusted between 4.5 and 5. The urease activity of the bacteria was approximately 0.8~1 U/mL/OD₆₀₀ (where 1U = 1μmol urea hydrolyzed per minute).

2.3. Cementation solution

The cementation solution used in the present study contained 1 mol/L anhydrous calcium chloride (CaCl₂, 111 g/L), 1 mol/L urea (CO(NH₂)₂, 60 g/L) and nutrient broth (6 g/L). All the solutions were prepared using deionized distilled water.

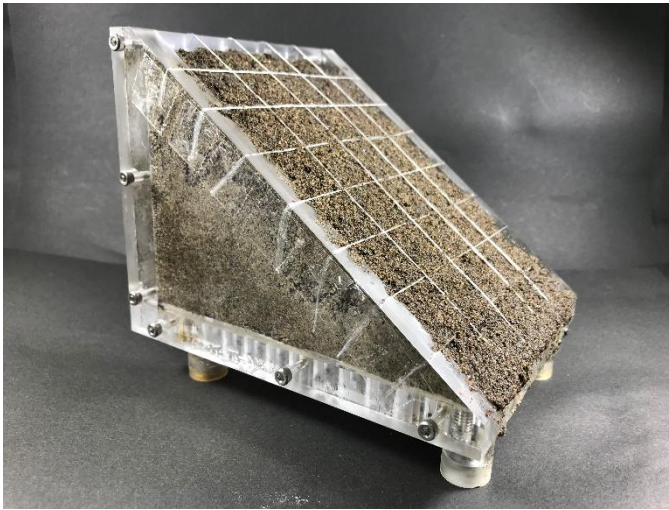
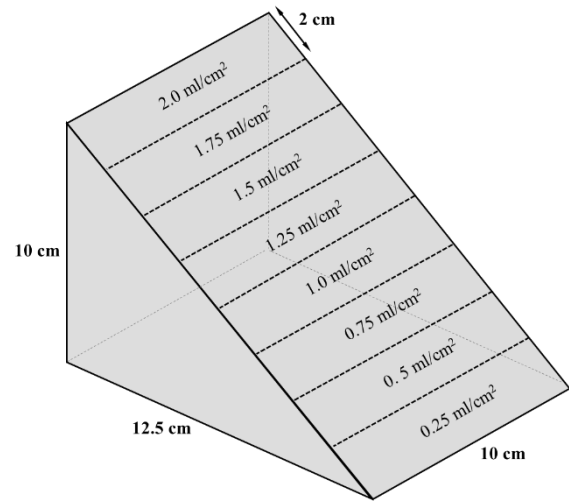


Fig. 1. Experimental setup used to treat the residual slope soil and the quantity of chemicals injected in every slot during every injection cycle



3. METHODS

3.1. Laboratory set up

Laboratory setup comprised of an assembled transparent slope mold that can be dismantled from all vertical sides (screw-fixed). The triangular face dimensions were 12.5 cm and 10 cm (horizontal and vertical respectively), and the width space of the mound was 10 cm. The gradient of the slope was designed to the gradient of standard cut slope (1:1.2) in Japan based on the road earthwork guidelines, Japan. The bottom of the slope contained a large number of small holes to facilitate the drain out of the solutions during treatment. The filter sheet was placed at the bottom of the slope to prevent the fine soil particles from being flushed out during treatment. The natural soil (neither oven dried nor chemically treated) was used at its native chemical and physical conditions in the experiment. 1500 g of soil was packed into the mold in five consecutive layers (layer by layer) ensuring that each layer was compacted evenly using blowing rod to achieve at least 90% of the maximum dry density, thereby to maintain consistency of experiments. The experimental set up is shown in Fig. 1.

3.2. Treatment process

A two-phase injection scheme was adopted for the treatment as recommended by many previous studies (Whiffin et al., 2014; Cheng et al., 2017; Gowthaman et al., 2019a). The bacteria culture was injected in the first phase. The injection of cementation solution was performed in the second phase. Two hours of time space was given between the two phases, that was to immobilize the bacteria cells with the soil particles. The cementation solution was injected every 24 hours during the 14 days treatment period, whereas bacteria culture was injected only two times (day 1 and day 7).

A grid-based injection method was performed. The slope surface was divided into eight slots along the longer side,

and each slot was further divided horizontally into five grids (Fig. 1). The injection volumes of solutions were approximately calculated based on the pore volume of the soil below each grid. All the solutions were simply applied to the surface of each grid and allowed to percolate naturally under its own gravitational and capillary forces. The injection was started from the bottom slope (bottom grids) and completed at top slope (top grids). Both the bacteria culture and cementation solution were injected in same rate of around 10 mL/min. The effluent collected through the bottom slope was used to study the internal bio-chemical conditions, so that the pH and Ca^{2+} concentration in the effluent of each injection were measured.

3.3. Segregation of cell pellets

Simultaneously, another identical slope was treated using segregated cell pellets in the place of bacteria culture. The segregation process was the same explained by Fujita et al. (2017). The bacteria culture medium was centrifuged at 10°C, 8000 rpm for 5 minutes using centrifuge tubes. The supernatant solution was then removed from the tubes, and cell pellets were washed few times by distilled water. Subsequently, the cell pellets were resuspended in distilled water to the same previous optical density (OD_{600}). The segregation process is conceptionally illustrated in Fig. 2.

3.4. Unconfined compressive strength test

After 14 days of treatment, the slope was flushed with distilled water to wash all the excess soluble salts prior to the UCS estimation. UCS of the specimen was estimated using needle penetrometer (Maruto Corporation, Tokyo, Japan). The needle penetrometer testing is an ISRM (International Society for Rock Mechanics) recommended method for indirect determination of UCS of soft, weak to very weak rocks and cemented soil specimens (Ulusay 2014).

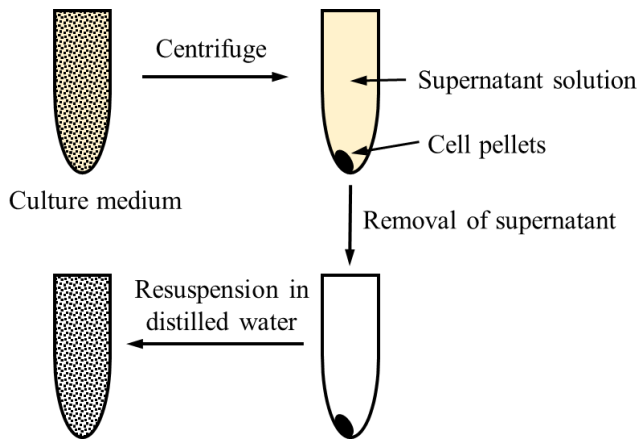


Fig. 2. Segregation process of cell pellets.



Fig. 3. Profile of the bio-cemented slope before flushing the salts and uncemented soil particles.

The correlation that was developed by conducting needle penetration test and standard UCS test in accordance with ISRM guidelines of rock characterization (on 114 natural soft rock samples and 50 improved soils with cement), was used to estimate the UCS at different points in solidified slope profile and discussed in this paper.

3.5. Permeability test

The permeability tests were carried out using falling head method using a DIK 4000 system (Daiki Rika Kogyo Co., Ltd., Saitama, Japan). The samples (5 cm in diameter and 5 cm in height) were solidified under the conditions that of slopes were treated, and further detail on testing methodology of permeability can be found in Wilson et al. (2019). The permeability of soil samples was measured before and after the treatment and compared.

4. RESULTS AND DISCUSSION

4.1. Profiling bio-cemented slope

In this study, the feasibility of bio-cementation on a residual slope soil was demonstrated using bench-scale model solidification test. Fig. 3 presents the bio-cemented profile of slope after the treatment by 14 injections of cementation solution and 2 injection of bacteria culture solution. It can be seen that the entire slope was successfully cemented by the surface percolation method used in this study, suggesting high implementation potential of this technique.

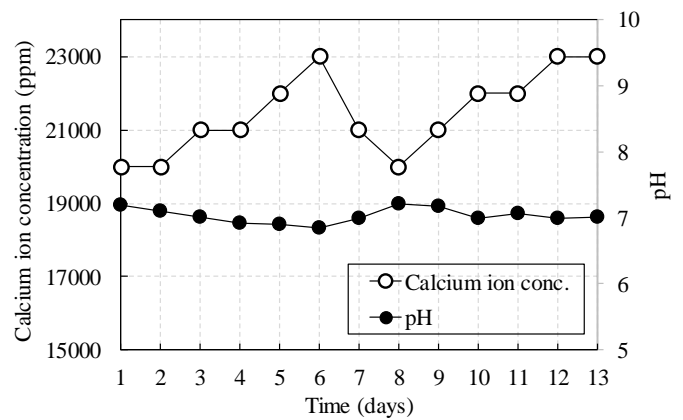


Fig. 4. pH and Ca^{2+} concentration measured at the effluent of every injection.

Basically, well graded soil has relatively low permeability compared to the uniformly graded soils due to the low void spaces. Thus, the risk of local clogging in natural soils is high during the treatment. In order to avoid the local clogging in slope, relatively a higher injection rate (10~20 mL/min) of cementation solutions was used herein as recommended by Cheng and Cord-Ruwisch (2014). The injection rate varied because of the decrease in permeability of slope soil (with the time) due to the formation of calcium carbonate minerals in pore spaces. The measured pH and Ca^{2+} concentration in the effluent are presented in Fig. 4.

The estimated UCS (using needle penetrometer) at different locations of the cemented slope is presented in Fig. 5(a) with respect to its solidified profile. The UCS values estimated at the surface of the slope is generally higher compared to that at the middle and at the bottom. The UCS in the cemented slope specimen varied between 1 and 9.1 MPa. Relatively closer UCS values (0.7~12.4 MPa) were reported by van Paassen et al. (2010) in their large-scale experiment, where corresponding CaCO_3 contents were reported to be between 12.6 and 27.3%. The highest UCS in the cemented slope was experienced along the bottom corner of the downslope. A similar observation is reported by van Paassen et al. (2009) in cubic meter bio-grout experiment. However, an uncemented zone was experienced in the slope. The zone directly below the top slope was not solidified (Fig. 5(a)), which was remained as same as untreated soil. This indicates that the catalyzing bacteria were not effectively supplied to the specific zone during the treatment. It can be understood that most of the bacteria cells were filtered at the surface zone of the slope, and some of them were washed out from the slope by surface sheet flow during the treatment.

The amount of CaCO_3 formed at a specific location governs the strength and the stiffness at that location. The prime factor that determines the carbonate content profile is distribution of bacteria in the soil matrix (Martinez et

al., 2013). Also, the attachment of bacteria with the soil particles depends on many factors such as grain size distribution, particle roughness and texture, mineralogy, and nature of the bacteria themselves (Mortensen et al., 2011; Gowthaman et al., 2019a). Thus, it was hard to assess the spatial distribution of bacteria and their activity with the time in the slope. However, the performance of the bacteria was indirectly monitored by measuring the chemical conditions of effluent (Fig. 4). Based on the results, it can be seen that the concentration of Ca^{2+} increases while pH decreases with the time, suggesting gradual denaturation of the bacterial urease. Typically, the bacteria would consistently perform for 6-8 days, and afterwards, it might have lysed little by little and become encapsulated in the crystals, resulting in a decrease in performance with time as detected. Therefore, bacterial culture was injected once again, the process called bio-dosing (Martinez 2012) on 7th day in order to maintain the activity consistently. The bio-dosing could produce more carbonate ions, ammonia and hence hydroxide ions from ammonia hydration, resulting increase in pH again. At the same time, more calcium ions were precipitated with produced carbonate ions, leading effluent with low calcium ion concentrations (Fig. 4). And once again, a similar tendency in effluent measurements was observed during the remaining treatment days.

4.2. Feasibility of using segregated cell pellets

Another identical slope was treated using segregated cell pellets instead of regular bacteria culture. This attempt was undertaken to study the distribution of urease enzyme

in the bacteria culture, thereby to enhance the efficiency in bio-cementation. For that, the regular culture was centrifuged, and bacteria cells were resuspended in the distilled water.

The solidified profile and estimated UCS of the solidified slope are presented in Fig. 5(b). The UCS values estimated at the surface are generally higher than that at middle or bottom, similar to the tendency that reported in the slope treated using regular bacteria culture. The UCS values ranged between 0.8 and 6.5 MPa. The highest UCS around 6.3 MPa was experienced at the corner of bottom slope like reported in the previous slope.

The comparison of the UCS values of the slopes treated using regular culture and segregated cell pellets shows that relatively higher CaCO_3 has been precipitated in the slope treated using regular culture, resulted slightly higher UCS. Also, the large uncemented zone (Fig. 5(b)) evidenced that a low carbonate content is precipitated while using segregated cell pellets. The above results indicate that urease enzyme of the bacteria used in this study would be somewhat secreted into the extracellular medium, that contributed to the increase of UCS while using regular culture. However, there was no significant difference observed between the measured urease activities of segregated cell pellets and whole-cell culture medium. Also, a very low urease activity was observed in culture supernatant. As the reason is unclear, further research should demonstrate which of the factor is responsible to the variation.

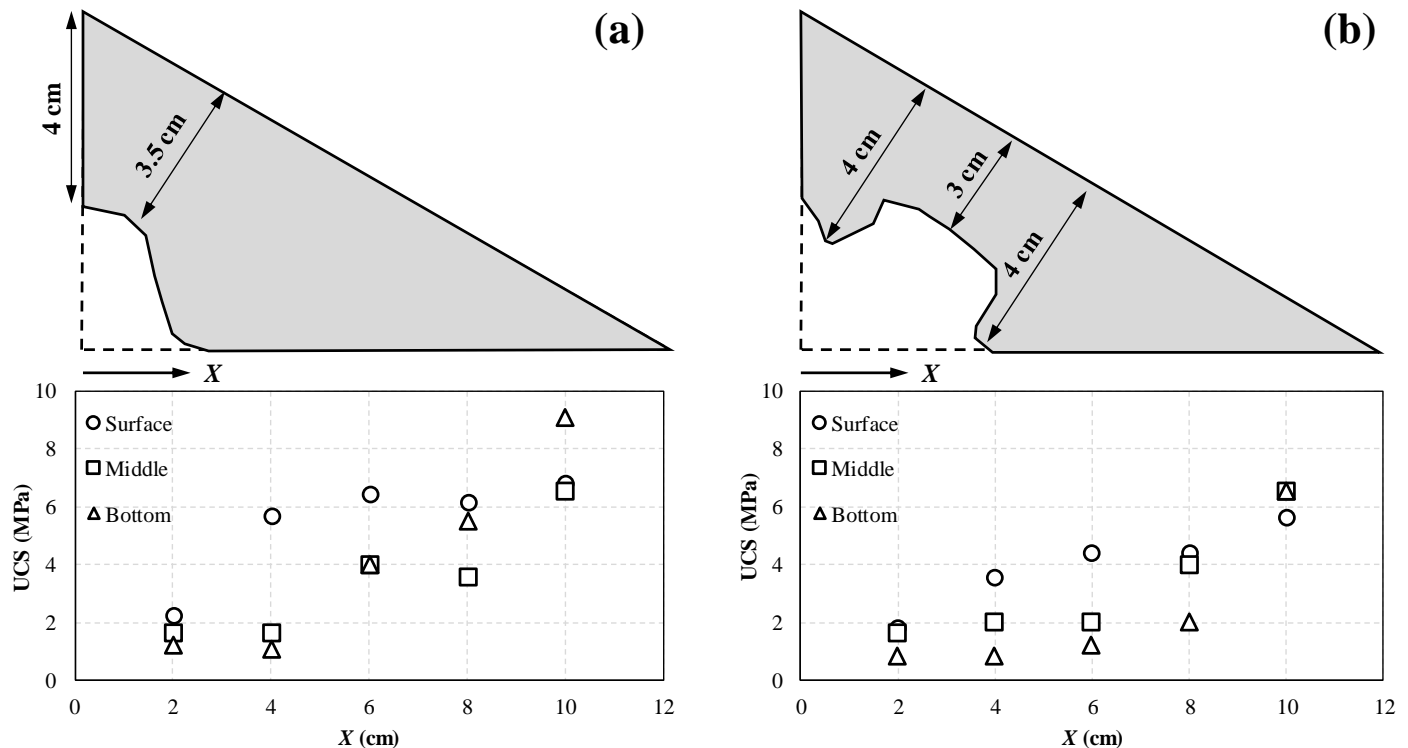


Fig. 5. Sectional profiles and UCS estimated at different locations of bio-cemented slopes treated using (a) culture medium and (b) segregated cell pellets.

In fact, secretion of urease enzyme is bacteria dependent. Some bacteria including *Pararhodobacter* sp. would not secrete the urease enzyme into the extracellular medium, which is firmly localized in or on the cell membrane (Fujita et al. 2017).

4.3. Permeability of cemented specimens

Permeability was measured separately for the specimens treated using regular bacteria culture and segregated cell pellets by falling head method. Also, the permeability of the untreated soil was measured to investigate the reduction caused by the calcium carbonate precipitation. Fig. 6 presents the permeability of the specimens before and after the treatment. The results indicate that the permeability was significantly reduced (approximately by 90%) with the surface percolation technique irrespective of using regular culture or segregated cell pellets. However, a little more reduction was obtained in the specimen treated by using regular culture due to higher deposition of calcium carbonate content as discussed in the previous section. Generally, the permeability of the specimen linearly decreases with increase in precipitated calcium carbonate content (Martinez et al., 2013). The results indicate that this surface percolation treatment technique was attributed to reduce the permeability in slope, which would help to control the infiltration of rainwater into the slope and enhance the surface sheet flow, and thus preserves the slope from failures. However, the reduction in infiltration of storm water might possibly result the water buildup on the bio-cemented stabilized slope surface, leading to additional overburden and sheet flow with high energy. Therefore, additional consideration regarding drainage is highly required. Facilitating the horizontal drains in slope is one of the feasible method to lower the excess surface flow, and that can be easily implied in certain intervals (along the long edge) by using perforated metal or slotted plastic drainages.

4.4. Heterogeneity in bio-cemented slope

Wide range of heterogeneity regarding UCS was observed in the cemented slope specimen, suggesting the calcium carbonate has been precipitated inhomogeneously. The loss of homogeneity was primarily caused by localized precipitation near inlet and by an asymmetric growth of mineral deposits along the transversal direction, as reported by Shahin et al. (2018). As the result of local clogging, development of preferential flow paths could often occur (van Paassen et al., 2009), leading more transport of bacteria and reactants along that way. The zones exposed to the preferential flow would be rich in precipitation, and at the same time, the stagnant zones in the areas downstream of the clog where supply of bacteria and nutrients can become limited, resulting less or no cementation. However, it is very hard to assess the preferential flow path developments in

experimentations. There are many factors probably associated with the above local clogging, and some of them are discussed briefly in this section.

The distribution of microbes is the most important factor determining the spatial distribution of calcium carbonate (Martinez et al., 2013). As discussed before, many factors including grain size distribution, morphology and mineralogy governs the distribution and attachment of the microbes. It is suspected that more bacteria cells might have possibly filtered at the surface level of soil slope, leading less cementation at the bottom.

The concentration of supplied reagents also plays an important role in homogeneous precipitation. During the injection, the soil closest to the injection port was exposed to the reactants significantly than the bottom of the soil profile (Whiffin et al., 2014), leading lower resources availability at deep zones. Therefore, the reactants should be supplied at the adequate concentrations to utilize uniformly throughout the soil matrix. However, Cheng et al. (2017) have reported that calcite precipitation rate and nutrient concentrations should be kept to its minimum to achieve homogeneous treatment. In this study, an adequate concentration of resources was supplied, and 40% of the reactants are not even utilized in the soil during the treatment (measured at the effluent).

The injection rate also another factor required to be considered for homogeneity in cementation (Cheng and Cord-Ruwisch, 2014). The balance between rate of supply and conversion is required to be maintained. The faster injection rates would transport the reactants quickly into the slope soil allowing less time for reaction along the path. In contrast, lower flow rates will leave more reactants in the fluid, resulting congestions and variations in cementation along the profile (Whiffin et al., 2014). Relatively a moderate to faster injection rate was adopted in this study to avoid local clogging in natural well-graded slope soil. The less observed surficial sheet flow during the surface injection suggested that no clogging has occurred during the treatment, leading to cementation all over the slope soil.

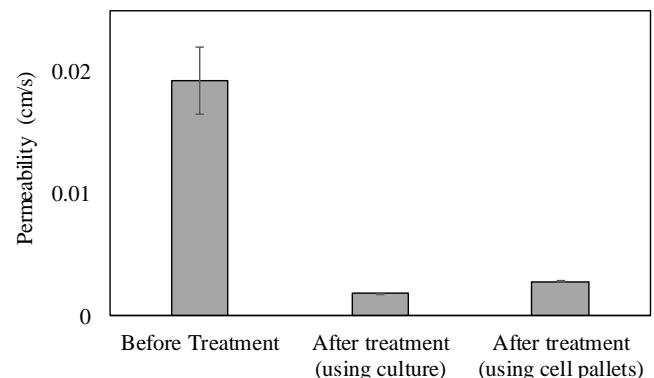


Fig. 6. Permeability of the specimens before and after treatments.

4.5. Implications for field implementations

The results obtained in this bench-scale study provide further understanding, as research move towards up-scaling to field implementation. In the large-scale applications, measuring and monitoring the urea hydrolysis rate, pH, and other chemical conditions at target treatment zones is crucial. Urea hydrolysis rate can be easily managed/ controlled by augmenting bacteria or enrichment in soil. Bio-augmentation of indigenous bacteria would be more sustainable than introducing exogenous bacteria to the native soil. Although bio-stimulation have been reported as more sustainable solution, augmentation of indigenous bacteria also would be sustainable enough and more feasible to control. It is also worth to note that discharging of ammonium by-products to the soil during bio-cementation implementations causes many harmful effects, and thus the subsequent removal of such harmful products must be considered.

5. CONCLUSIONS

A bench-scale experiments have shown the technical feasibility of bio-cementation as slope surface stabilization technique. The study also has demonstrated that using regular culture medium (*Lysinibacillus xylanilyticus*) provides a better cementation compared to that of using segregated cell pellets medium. The UCS values estimated in solidified slope varied between 1 and 9.1 MPa. Relatively higher UCS values were experienced along the slope surface compared to that at depths, suggesting the feasibility of forming a stable crust layer of 3.5~4 cm at the surface. However, large range of heterogeneity was evidenced in the cemented slope. Particularly, certain zone remained as untreated soil, whereas bottom slope corner exhibited the highest UCS. Therefore, further research should demonstrate which of the discussed mechanisms are responsible for the observed heterogeneity in deposition of carbonate. Moreover, the permeability test results suggested that the permeability of the untreated soil has reduced by 90% by the calcium carbonate precipitation, which could inhibit the excess infiltration of rainwater into the slope structure and enhance the surface runoff.

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