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ORIGINAL PAPER

Effect of microbial-induced calcite precipitation towards strength and permeability of peat

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Abstract

Peat is known as problematic ground with low bearing capacity and extensively high compressibility. Bio-cementation or commonly known as microbial-induced calcite precipitation (MICP) has been recently introduced as a ground improvement alternative for peat under waterlogged condition. Using isolated bacteria strains P19 and P21 from tropical peat, it is found that unconfined compression strength (UCS) increases with bacteria concentration at a reducing rate. A maximum unconfined compressive strength of 82.05 kPa was measured with bacteria strain P21 at 10^8 CFU/mL. For the range of cementation reagent varying from 0.1 to 4.0 mol/kg, the largest strength improvement occurred at 1 mol/kg and 2 mol/kg using indigenous bacteria and bacteria strain P21, respectively, for peat with sand content of 25%. At 4.0 mol/kg, the cementation reagent has detrimental effect to MICP resulting in significant reduction in strength. Due to MICP, the UCS of peat increases with sand content. Calcium carbonate precipitation results in a reduction of permeability and an increment of strength of peat–sand mixture under a submerged condition up to 28 days.

Keywords Unconfined compressive strength · Calcium carbonate precipitation · Urea · Calcium chloride

Introduction

Tropical lowland peatlands covered approximately 23 million ha in Southeast Asia with the most extensive coverage situated in the coastal zone of Southeast Asia, especially in the countries such as Indonesia and Malaysia (Melling 2016; Mutalib 1992). However, peatland construction is usually unfavourable with its poor bearing capacity, low permeability, high compressibility with high creep rates and often difficult accessibility (Hashim and Islam 2008; Kolay et al. 2011). Peat stabilisation can be done through deep mixing and surface stabilisation along with chemicals additives or binders (Islam and Hashim 2009, 2008). Calcium-based binders including Portland cement or lime were commonly used for such efforts promoting cementation with strong ground improvement (Kazemian et al. 2011a; Pourakbar and Huat 2017; Souliman and Zapata 2011). However, the use of Portland cement as the binders is detrimental to the environment, and its manufacturing was known to contribute 7% of man-made CO_2 emissions globally due to carbonate decomposition (Gartner 2004; Matthews et al. 2009; Pourakbar and Huat 2017).

An alternative sustainable method known as microbial-induced carbonate precipitation (MICP) that utilises natural biological process for ground improvement was introduced and widely studied (El Mountassir et al. 2018; Mujah et al. 2017). Microbial-induced carbonate precipitation works by bio-precipitation of carbonate crystal on soil materials bridging soil particles and sealing void leading to improvement of engineering (DeJong et al. 2006). MICP is one of the biomineralisation processes that can produce minerals via a number of different metabolic activities including photosynthesis, denitrification, ammonification, sulphate reduction, methane oxidation and with urea hydrolysis by ureolytic bacteria being one of the most popular and easily controlled reactions used to precipitate minerals in a short period of

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time (Dhami et al. 2012; Stocks-Fischer et al. 1999; Zhu and Dittrich 2016). MICP based on hydrolysis of urea by ureolytic organism is followed with the introduction of calcium (Ca^{2+}) ions into the environment leading to calcite ($CaCO_3$) precipitation (Achal et al. 2009; Stocks-Fischer et al. 1999). The reaction of MICP through urea hydrolysis is as followed:

$$CO(NH_2)_2 + 3H_2O \rightarrow 2NH_4^+ + HCO_3^- + OH^- \quad (1)$$

$$HCO_3^- + H_2O + OH^- \leftrightarrow CO_3^{2-} + 2H_2O \quad (2)$$

$$Ca^2 + CO_3^{2-} \rightarrow CaCO_{3\,(s)} \quad (3)$$

Calcite precipitation through MICP of treated soil has shown improvement of strength and reduction of permeability (Cheng et al. 2013; Chu et al. 2012; DeJong et al. 2006, 2010; Montoya and DeJong 2015; Whiffin et al. 2007; Zhao et al. 2014). MICP as emerging soil improvement method has been widely studied for granular inorganic soil (Al Qabany and Soga 2013; Feng and Montoya 2015; Hamdan et al. 2017; Lee et al. 2013; Sharma and Ramkrishnan 2016; Smith et al. 2017). MICP treatment was commonly practiced through flushing of cementation reagents with constant flow rate or surface percolation and soaking of soil specimens (Cheng and Cord-Ruwisch 2014; Harkes et al. 2010; Khodadadi et al. 2017). MICP for soil improvement was intensively studied in inorganic soil especially sand (Al Qabany and Soga 2013; Al Qabany et al. 2011; Harkes et al. 2010; Whiffin et al. 2007; Zhao et al. 2014). Recently, the application was extended to residual soil consisted of about 40% of sand particles and 60% of fine grained particle (Lee et al. 2013; Ng et al. 2012) and marine clay (Ivanov et al. 2015). Sandy organic silt was studied showing strength improvement with injection of *S. pasteurii* for initiation of MICP.

A study done by Canakci et al. (2015) on MICP to stabilise organic soil (sandy organic silt) obtained from the Sakarya region of Turkey using *S. pasteurii* NCIMB 8221 showed $CaCO_3$ increased by about 20% in the treated samples resulting in improvement of compressibility and shear strength of the organic soil. Sato et al. (2016) found that it is possible to solidify peaty soil in Hokkaido, Japan, using indigenous urease activity and addition of urease from sward beans (*Canavalia gladiate*). Another study treatment of peaty soil (loss of ignition of 65.815%) with fibre incorporated with bio-cementation using isolated native bacteria showed higher fibre content gained higher strength (Chen et al. 2021). Gowthaman et al. (2021a) has proposed the use of scallop powder addition with *Sporosarcina* sp. (SIID 33,506) to improve the unconfined compressive strength of clayey amorphous peat with organic content of 39% from Tomikawa, Hokkaido, Japan. Although various effort was made to study MICP on organic soil, there is still a lack of study on the effect of bio-cementation for tropical peat stabilisation. The objective of this study is to evaluate the effect of MICP towards stabilisation of peat in term of strength gain and permeability changes under different isolated indigenous bacteria concentration, cementation reagent dosage and sand content.

Methodology

Materials

Soil samples

Tropical peat was collected at Curtin University Malaysia, Miri, Sarawak (Fig. 1). Visual observation shows that the peat sample was dark brown in colour. Collected peat was classified and characterised based on ASTM D4427 (ASTM 2018) method. The collected peat had natural moisture content between 670 and 800% and organic content between 94 and 96%. The properties of the collected peat including the moisture content, fibre content, organic content, ash content, pH, type of peat and specific gravity are summarised in Table 1. River sand was obtained locally and washed by rinsing with tape water for several times to minimise the presence of impurities and oven dried. Figure 2 shows the particle size distribution curve of river sand.

Bacterial culture

MICP based on urea hydrolysis was used for this study. Previous study makes use of exogenous bacteria as urease source. Ureolytic non-pathogenic bacteria such as *S. pasteurii* and *B. megaterium* were commonly introduced to the target soil to induce urea hydrolysis facilitating bio-cementation mechanism (Achal et al. 2009; Ng et al. 2012). The previous study had also shown the presence of ureolytic microbial sources in tropical peat (Blonska 2010; Phang et al. 2018). Hence, urease bacteria isolated from tropical peat were prioritise. Indigenous sources of ureolytic bacteria isolated from tropical peat (isolated from the same area of peatland used for current study), bacteria strain P19 (GenBank MH639002) which was identified as *Enteractinococcus* sp. and bacteria strain P21 (Genbank MH639001) identified as *Staphylococcus* sp. were obtained from the Faculty of Engineering and Science, Curtin University, Malaysia, and used for the study.

A seed culture was produced by transferring a small amount of the selected bacterial culture into 100 ml of Tryptic Soy Broth (TSB) (Merck, USA) supplemented with sterile urea (2%) (Sigma, USA) and incubated in orbital shaker up to 48 h at 28 °C and 120 rpm. The cell culture

Fig. 1 Site of peat obtained for this study (image obtained from Google Earth)

was harvested by centrifugation for 10 min at 5000 g at 4 °C. The harvested cells were then washed twice in sodium phosphate buffer 0.1 M of pH 7 to remove metabolic waste products from bacterial growth that may cause interference with experimental study. Bacterial cells were resuspended with saline solution at required concentration of 10^5, 10^6, 10^7 and 10^8 CFU/mL. Indigenous urealytic sources were also observed in this study without addition of bacteria.

Specimen preparation

Reconstituted peat specimens were used for the experiment. Coarse fibre roots were removed, and wet peat slurry was passed through 2-mm sieve and collected. The collected peat slurry was then mixed and stored in a single container to homogenise it. In average, the moisture content of the peat slurry was 400%. Sand was frequently used as filler for study related to calcium-based stabilisation effort (Nikookar et al. 2012; Rahgozar and Saberian 2016; Saberian and Rahgozar 2016; Sing et al. 2008; Venuja et al. 2017; Wong et al. 2013). For this study, peat slurry was mixed with 25%, 50% and 75% of dry sand to weight of wet peat slurry. Acidic pH of peaty soil was usually adjusted with $NaHCO_3$ to provide an environment favouring MICP (Chen et al. 2021; Sato et al. 2016). Hence, high concentration of cementation reagent was used since no pre-adjustment of pH was done for this study. Cementation reagent was prepared by adding equal molars of urea and calcium chloride in the range of 0.1–4.0 mol/kg of wet peat slurry (Table 2). XRF analysis of industrial grade urea and calcium chloride is shown in Table 3. To produce each stabilised peat admixture, peat–sand mixture was mixed with different dosages of cementation reagents and 100 mL of bacteria culture for every kg of peat slurry followed by homogenising for 5 min with kitchen mixer to ensure uniform distribution. For control, 100 mL of distilled water for every kg of peat slurry was added to maintain the water content in the peat–sand mixture. Table 4 shows the test plan for this study. Set 1 studied the effect of bacteria concentration

Table 1 Basic properties of natural peat used in this study

Basic soil property	
Natural moisture content (%)	670–800
Fibre content (%)	50–60
Organic content (%)	94–96
Ash content (%)	4
pH	3.9–4.9
Von post-designation	H3–H5
Specific gravity	1.11–1.23

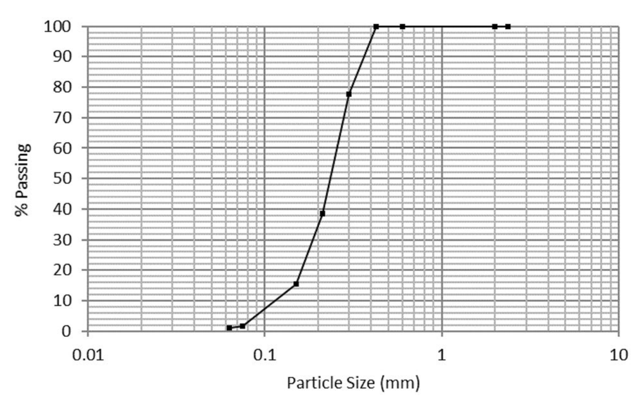


Fig. 2 Particle size distribution curve of river sand

for different types of bacteria to MICP. Two isolated bacteria from peat at different concentrations (four combinations) was used in this study. Considering three replicates, in total, 24 samples had been prepared and tested. Set 2 investigated the effect of cementation reagents to MICP with five combinations (15 samples). Set 3 explored the effect of sand content and curing period to unconfined compression strength, in total 12 combinations. With three replicates for each combination, in total, 36 samples had been prepared and tested. Set 4 studied the effect of sand content and curing period to permeability of MICP treated peat. Nine combinations (27 samples) had been considered in set 4.

The moulds used for the unconfined compression test were PVC tubes with 50-mm internal diameter and 250 mm long. For falling head tests, the moulds were cylindrical PVC tubes of 63-mm internal diameter and 300 mm long. The peat slurry was placed carefully into the PVC tube minimising air trapping. MICP treatment was commonly performed through flushing or injection technique and surface percolation method (Mujah et al. 2017). Those techniques ensure continuous feeding of oxygen and flow of cementation reagent with or without bacteria agent. Peat has low permeability or hydraulic conductivity, where most peat area is in swampy, waterlogged and anoxic condition (Chason and Siegel 1986; Landva and Pheeney 1980). Previous study has suggested the possibility of MICP in peat with premixing and wet curing method (Phang et al. Submitted-b). Hence, for this study. wet curing for soft soil stabilisation simulating saturated field condition was performed (EuroSoilStab 2001; Hebib and Farrell 2003). In general, the tubes containing mixed samples were placed vertically submerged in water with surcharge load of 9 kPa. The samples were cured at room temperature for required period before subjecting to testing.

Table 2 Chemical composition of materials for the stabilised peat

	Sand (%)	Calcium chloride (%)	Urea (%)
SiO_2	89.28	-	-
CaO	1.21	47.44	0.15
K_2O	0.96	-	0.02
Fe_2O_3	1.33	-	-
P_2O_5	1.48	0.39	0.35
MgO	0.25	-	-
Al_2O_3	4.26	-	0.01
SO_3	-	-	0.07
Cl	0.88	52.14	0.11
TiO_2	0.28	-	-
SrO	0.0083	0.02	-
CuO	0.0071	0.007	0.01
Rb_2O	0.0033	-	-
ZnO	0.0049	-	-
ZrO_2	0.05	-	-
V_2O_5	-	-	0.01

Geotechnical tests

Unconfined compression test and falling head permeability tests were performed to quantify the mechanical properties of the test specimens. MICP study previously shown that MICP occurring in soil may lead to bio-cementation of soil that leads to improve of unconfined compressive strength of soil and reduction in permeability. The effectiveness of bio-cementation to peat strength was evaluated by measuring unconfined compressive strength (UCS) according to the procedure described in ASTM D2166 (ASTM 2016). After curing, the samples were extruded and trimmed carefully with minimum disturbances to form specimens with a diameter-to-height ratio of 1:2 (53 mm × 106 mm). The specimens were then tested using universal testing machine (Lloyd Instruments) with the loading rate of 2.0 mm/min. The unconfined compressive strength was recorded as the peak stress of the soil stress–strain curve or was identified as peak stress corresponding to vertical strain reaches 20% as described by a previous study (Sing et al. 2008). All tests were done in triplicates, and results were shown in mean value. Selected samples were dried and proceed to calcium carbonate quantification. Permeability changes due to MICP were evaluated through hydraulic conductivity measurements of peat with conventional falling head apparatus (ELE International, UK) according to ASTM 4511 method (ASTM 2011). The time taken for a measured quantity of water to flow through the specimen was recorded, and the coefficient of permeability (m/s) was calculated using a standard formula.

Determination of calcium carbonate content

The amount of $CaCO_3$ precipitation in the samples was determined by acid washing technique (Keykha et al. 2017; Mortensen et al. 2011). Samples were dried in the oven with its mass measured before and after rinsing with HCl (5 M).

Table 3 Dosage concentration of cementation reagent consisting of calcium chloride and urea

mol/kg	$CaCl_2$(%)	Urea(%)	Total (%)
0.1	**1.11**	**0.6**	**1.71**
0.2	**2.22**	**1.2**	**3.42**
1	**11.1**	**6.01**	**17.1**
2	**22.2**	**12.01**	**34.21**
4	**44.39**	**24.02**	**68.42**

Table 4 Experimental design for peat stabilisation study for geotechnical testing

Set test	Test	Sand (%)	Bacteria	Cementation reagent dosage (mol/kg)	Curing duration (days)
1	UCS	25	P19 (10^5,10^6,10^7,10^8CFU/mL), P21 (10^5,10^6,10^7,10^8 CFU/mL), indigenous	2	28
2	UCS	25	Selected from set test 1, indigenous	0.1, 0.2, 1, 2, 4	28
3	UCS	25, 50, 75	Selected from set test 1	Selected from set test 2	3, 7, 14, 28
4	Permeability	25, 50, 75	Selected from set test 1	Selected from set test 2	3, 7, 28

Generally, the samples were rinsed multiple times on filter paper to allow HCl (Merck, Germany) to dissolve the carbonate salt while passing through the filter. The difference between the measured mass before and after rinsing was taken as the mass of $CaCO_3$, and the results were expressed at percentage of precipitated $CaCO_3$ over the dry mass of specimens. pH of wet peat after curing was also measured based on ASTM D4972. All tests were done in triplicates, and results were shown in mean value.

Scanning electron microscopy (SEM) and X-ray diffraction analysis

For specimen with the highest strength, a portion of peat fabric was cut and preserved for SEM imaging and XRD analysis. The fabric of the peat surface with calcium carbonate precipitation was observed by scanning electron microscopy. The collected dry samples were mounted directly into the SEM stubs and sputter-coated with a gold/palladium mixture. X-ray diffractometer (XRD) was done to identify crystal phase of precipitated $CaCO_3$ shown in SEM analysis. Samples were analysed using XRD and crystalline mineral phases search done using Crystallography Open Database (COD) (Rev. 198,327) (http://www.crystallography.net/cod/) as described by Gražulis et al. (2011).

Results and discussion

Unconfined compressive behaviour

To investigate the influence of MICP on stabilised peat, experimental results of unconfined compression tests on

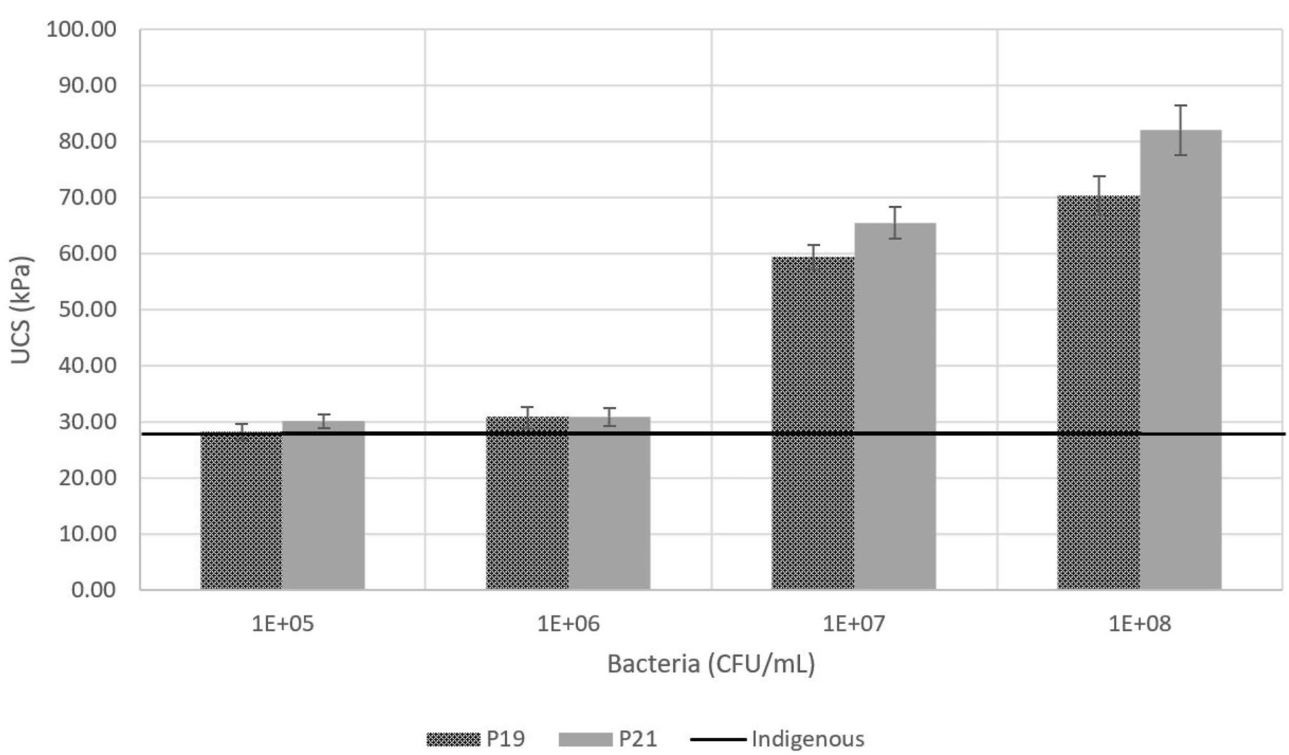


Fig. 3 Effect of bacteria type and concentration towards unconfined compressive strength for peat mixed with 25% sand

the test specimens are focused on the effects of bacteria types and concentration, cementation reagent dosage, different amount of sand and curing time. The results of such effects on the unconfined compressive strength of the test specimens are discussed.

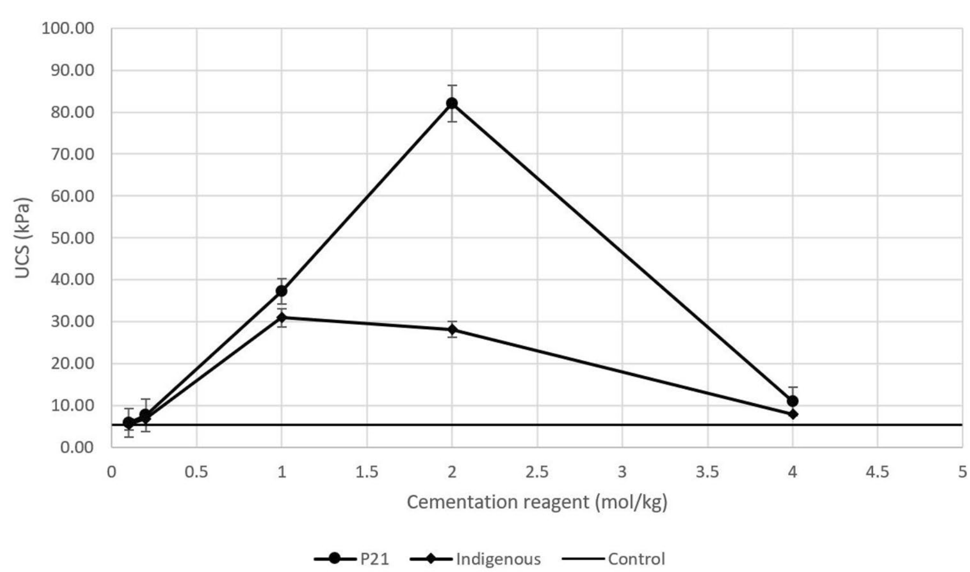
Effect of bacteria type and concentration

Figure 3 shows the experimental results of the effect of different types of bacteria including indigenous bacteria presence in peat and concentration (CFU/mL) of the added ureolytic bacteria strains on the UCS of the stabilised peat. Each test specimen was prepared with 25% sand and cementation reagent dosage of 2 mol/kg cured at room temperature submerged in water for 28 days. Bacteria strain P19 and P21 previously isolated from the same tropical peat were used for the study. Both bacteria strains have shown high urease activity (> 400 U/mL) in aqueous solution and found to precipitate calcite (Phang et al. Submitted-a). Bacteria strain P19 and P21 were added at different concentrations along with cementation reagent, whereas for indigenous bacteria, only cementation reagent was added to the peat sand specimens before curing. At 10^5 and 10^6 CFU/mL, specimens treated with bacteria strain P19 showed 28.36 kPa and 30.91 kPa, whereas for bacteria strain P21, UCS were 30.16 kPa and 30.91 kPa. The improvement of UCS for both bacteria strains at 10^5 and 10^6 CFU/mL were low as compared to indigenous bacteria with 28.11 kPa. UCS increment was more obvious at 10^7 CFU/mL for both bacteria strains suggesting the threshold of bacteria concentration needed for MICP treatment for current peat sand mix. The UCS was about double of that for indigenous bacteria. However, UCS only increases by 20 to 30% when the concentration for both bacteria strains is increased from 10^7 to 10^8 CFU/mL. This may imply that not all the bacteria contribute to bio-cementation towards strength improvement. The highest UCS was observed with bacteria strain P21 at concentration of 10^8 CFU/mL at 82.05 kPa and bacteria strain P19 at 70.36 kPa. Hence, the results suggested that bacteria strain P21 has better strength improvement with MICP than bacteria strain P19 in 25% sand mixed with peat. Bacteria addition (10^7 and 10^8 CFU/mL) showed higher UCS gain as opposed to the use of solely indigenous bacteria. The mount of ureolytic bacteria introduced may affect urea hydrolysis rate which affect calcium carbonate precipitation and ultimately towards performance of MICP. Apart from urease activity governed by ureolytic bacteria, the bacteria cells also contribute to nucleation side for stable calcium carbonate formation that leads to bio-cementation (Ferris et al. 1996; Phillips et al. 2013).

Effect of cementation dosage

For this part of the study, the total cementation components which contained urea and calcium chloride are added to peat slurry in a range of 0.1–4 mol/kg towards wet weight of peat slurry to identify its effect towards MICP with 25% sand as filler after 28 days curing. Figure 4 shows the experimental results of the effect of concentration dosage with added bacteria strain P21 (10^8 CFU/mL) and without bacteria addition (indigenous) on the UCS of the stabilised peat after 28 days curing. Overall, treated samples showed higher strength compared to control sample (peat with 25% sand) without treatment with 5.3 kPa. The highest increment was observed at 2 mol/kg for sample treated with bacteria strain P21 while 1 mol/kg for indigenous bacteria. At 2 mol/kg, the

Fig. 4 Effect of cementation reagent dosage towards unconfined compressive strength



UCS is similar to that at 1 mol/kg indicating the cementation reagent dosage of 1 mol/kg is adequate for the indigenous bacteria and additional calcium ions could not be utilised by indigenous bacteria for bio-cementation. The UCS observed for sample treated with bacteria strain P21 and solely with indigenous bacteria at 0.1 and 0.2 mol/kg were rather low with slight improvement as compared to control (peat with 25% sand only). Specimen treated with bacteria strain P21 showed increasing strength gain with increasing dosage of 0.1, 0.2, 1.0 and 2 mol/kg up to 82.05 kPa while a drop-in strength at 4 mol/kg to 10.97 kPa. The trend for indigenous bacteria showed strength increment at 0.1, 0.2 and 1 mol/kg up to 30.91 kPa, slight reduction of strength at 2 mol/kg and continued with a significant reduction in strength at 4 mol/kg down to 7.93 kPa. Such phenomena may suggest that excessive amount of cementation components has detrimental effect at strength gain. High concentration of cementation reagents may inhibit the activity of bacteria. Besides, the cementation reagent contains calcium chloride which is highly soluble and may contribute to ion exchange in soil. Ion exchange of soil may contribute to hardening of soil without any cementation occurrence (Gray 1970; Moayedi et al. 2013). Calcium chloride may improve the strength of peat to an extent, but excessive concentration of $CaCl_2$ may cause strength reduction (Kazemian et al. 2011b). Peat has a significant high cation exchange capacity (CEC) due to the presence of humic substances including humic acid and fulvic acid (Chen and Wang 2006; Kazemian et al. 2011a). Literatures suggest that humic substances may react with calcium ions (Ca^{2+}) and inhibit or retard calcium-based stabilisation of peat which may explain lower strength gain at 0.1 and 0.2 mol/kg (Chen and Wang 2006; Huat et al. 2014; Jawad et al. 2014). However, humic substances in peat and its natural nitrification process may benefit the removal of ammonia produced during MICP which is a consent for environmentally friendly MICP process (Lee et al. 2019; Pal et al. 2010; Terry et al. 2018). High ammonia released from MICP may pose a threat to human health and other organisms in the environment (Lee et al. 2019). Other successful efforts to remove ammonia produced from MICP was done through struvite precipitation (Gowthaman et al. 2021b).

Effect of sand filler and curing duration

The influence of sand content with MICP on stabilisation peat was studied, and the UCS results of stabilised peat for 3, 7, 14 and 28 days of curing time are shown in Fig. 5. Based of the above study, bacteria strain P21 at 10^8 CFU/mL with cementation dosage of 2 mol/kg of peat slurry was used for treated specimens for this part of the study. It can be observed from Fig. 5 that the unconfined compressive strength of MICP treated test specimens increased while increasing the duration of curing in water and the amount of sand percentage. When the 75% of sand was applied, the unconfined compressive strength of test specimens increased progressively from 5.15 to 28.92, 55.32 and 94.85 kPa at the respective curing time in water of 3, 7, 14 and 28 days. The highest UCS was observed at 75% followed by 50% and 25% sand at 94.85, 87.56 and 82.38 kPa, respectively. UCS for the treated samples were 15.6, 13.3 and 9.2 times larger than those untreated for sand content of 25, 50 and 75%, respectively. It is evident from the findings that the magnitude of strength gains of treated specimens compared to untreated specimens of different amount of sand content and duration of curing in water suggested bio-cementation effect of MICP which improved the UCS of test specimens. Treatment of sands via MICP has resulted in increases in UCS of greater than three orders of magnitude and up to four orders of magnitude (Al Qabany and Soga 2013; Al Qabany et al. 2011; Terzis and Laloui 2018; van Paassen et al. 2010). Calcium carbonate precipitation in the treated specimens was quantified and presented in Fig. 6. The trends showed increasing $CaCO_3$ content with increasing curing durations and sand content. The highest amount was recorded for treated peat with 75% sand at 0.13 g/g $CaCO_3$ precipitated. From the results, increasing amount of sand was shown to increase precipitated $CaCO_3$ though cementation reagent and bacteria concentration are lower due to decreasing peat content. As urea hydrolysis progresses, CO_3^{2-} would be produced and bind with Ca^{2+} as calcium carbonate bridging particles for peat sand mixture. In this process, ammonium would be produced as waste, and the available of ammonium may lead to sorption of humic substances displacing Ca^{2+} from peat particles increasing the conversion towards $CaCO_3$ (Slavek et al. 1982; Tipping 2002). These may suggest high late strength of those treated at 28 days along with higher $CaCO_3$ precipitation. The UCS of the peat sand mixture increases linearly with curing period, indicating the continuation of the calcium carbonate formation up to 28 days under submerged condition. This is possible in view of the peat is waterlogged, and the bacteria strain P21 was isolated from peat.

Permeability

Previous study of MICP has shown bio-clogging effect that reduced permeability of the treated samples with precipitation of calcium carbonate at soil pore space. Permeability of the treated and untreated peat sand mixture was assessed through saturated hydraulic conductivity (K) corrected to standard temperature of 20 °C of samples cured at duration of 7, 14 and 28 days. As expected, Fig. 7 shows that in overall, the hydraulic conductivity of treated samples was reduced with increasing curing periods and was increased with sand content. The lowest value was observed for treated peat with 25% sand with 3.21×10^{-7} m/s as

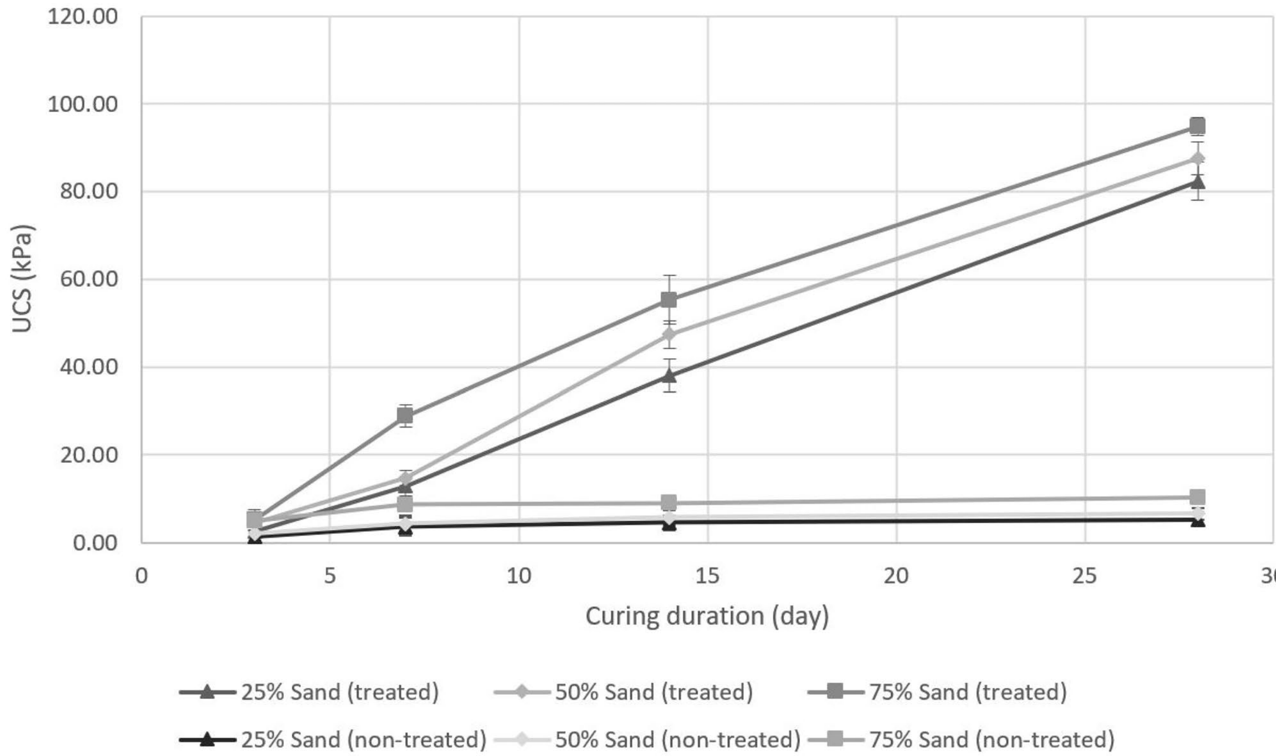


Fig. 5 Effect of amount of sand as fillers and curing duration

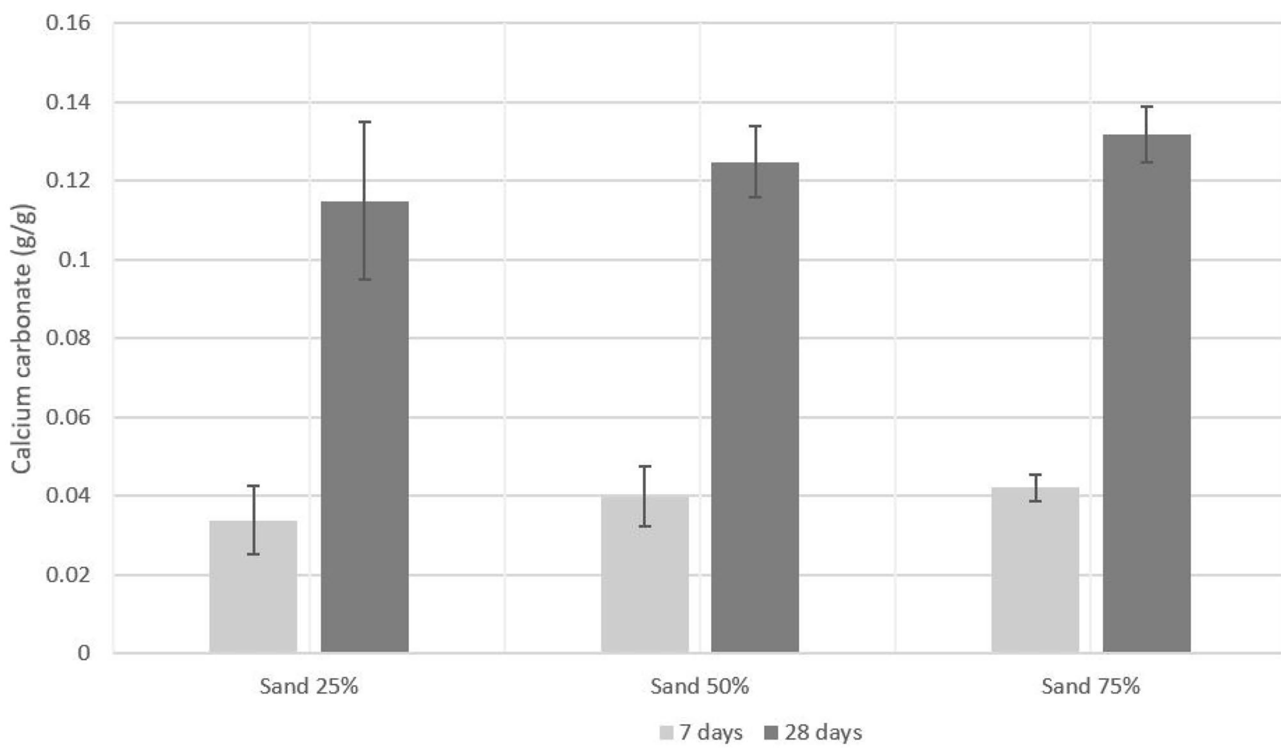


Fig. 6 Calcium carbonate precipitation of treated peat with 25%, 50% and 75% sand after 7 and 28 days of curing period

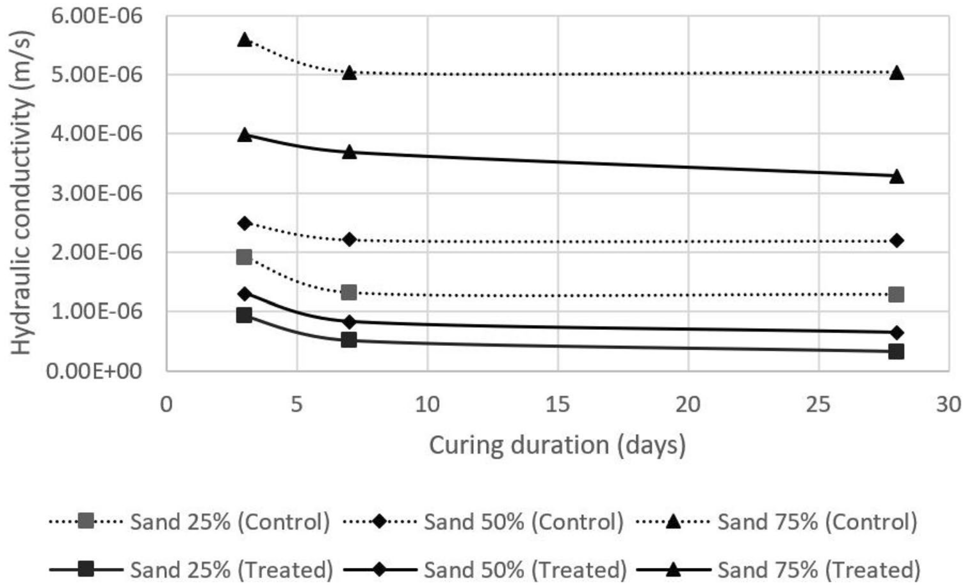


Fig. 7 Effect of MICP on hydraulic conductivity of peat at sand mixture of 25%, 50% and 75% up to 28 days curing period

compared to control with 1.28×10^{-6} m/s. Hydraulic conductivity after 28 days curing for 50% sand (treated), 75% sand (treated), 50% sand (control) and 75% sand (control) was 3.21×10^{-7} m/s, 6.47×10^{-7} m/s, 2.19×10^{-6} m/s and 5.04×10^{-6} m/s, respectively. As shown in Fig. 8, permeability reduction for 25%, 50% and 75% sand was in a range of 51.31–74.93%, 48.01–70.47% and 28.66–34.78%, respectively. It is found that permeability reduction up to 28 days for both 25% sand and 50% sand is similar, while 75% sand indicates a smaller reduction. The MICP treated sand columns were reported to achieve as much as 90–100% reduction in permeability from initial values (Bang et al. 2001; Gollapudi et al. 1995; Tobler et al. 2011). van Paassen (2009) reported biotreated soils with approximate 60% reduction in the initial permeability with 100 kg/m^3 $CaCO_3$ precipitation, while Ivanov et al. (2010) have observed permeability reduction at a range 50–99% for MICP treated soil.

Scanning electron microscopy and X-ray diffraction analysis

X-ray diffraction analysis was performed on selected samples in order to study the polymorph of calcium carbonate formed in the stabilised peat specimens. Figure 9 shows XRD analysis of crystal phase in the specimens for the representative samples of MICP treated and untreated peat with sand crystal. It is shown that calcium carbonate in

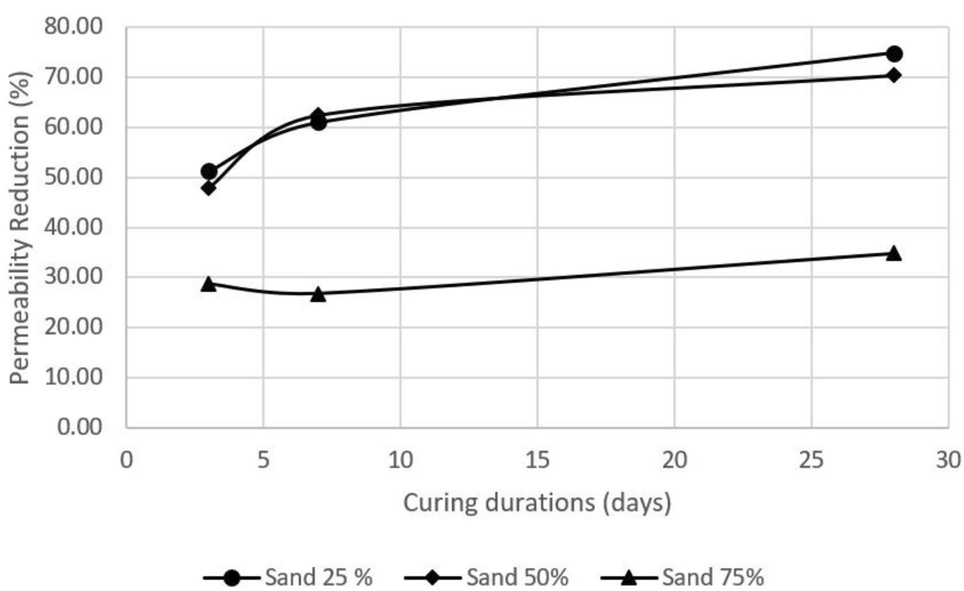


Fig. 8 Reduction in permeability due to MICP towards peat at sand mixture of 25%, 50% and 75% up to 28 days curing period

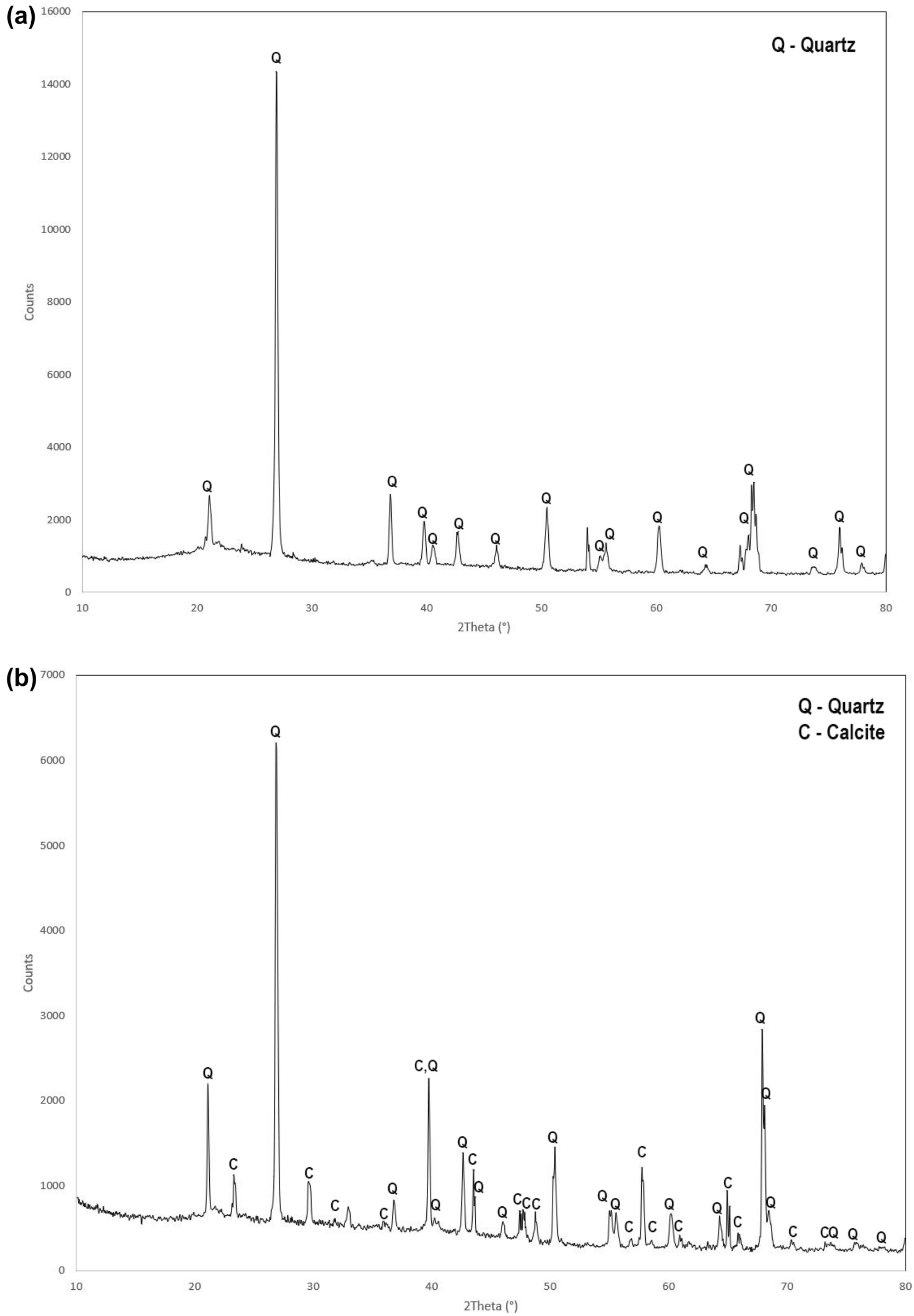


Fig. 9 X-ray diffraction analysis of **a** control peat specimens (25% sand; 28 days curing) and **b** MICP treated peat (25% sand; 28 days curing)

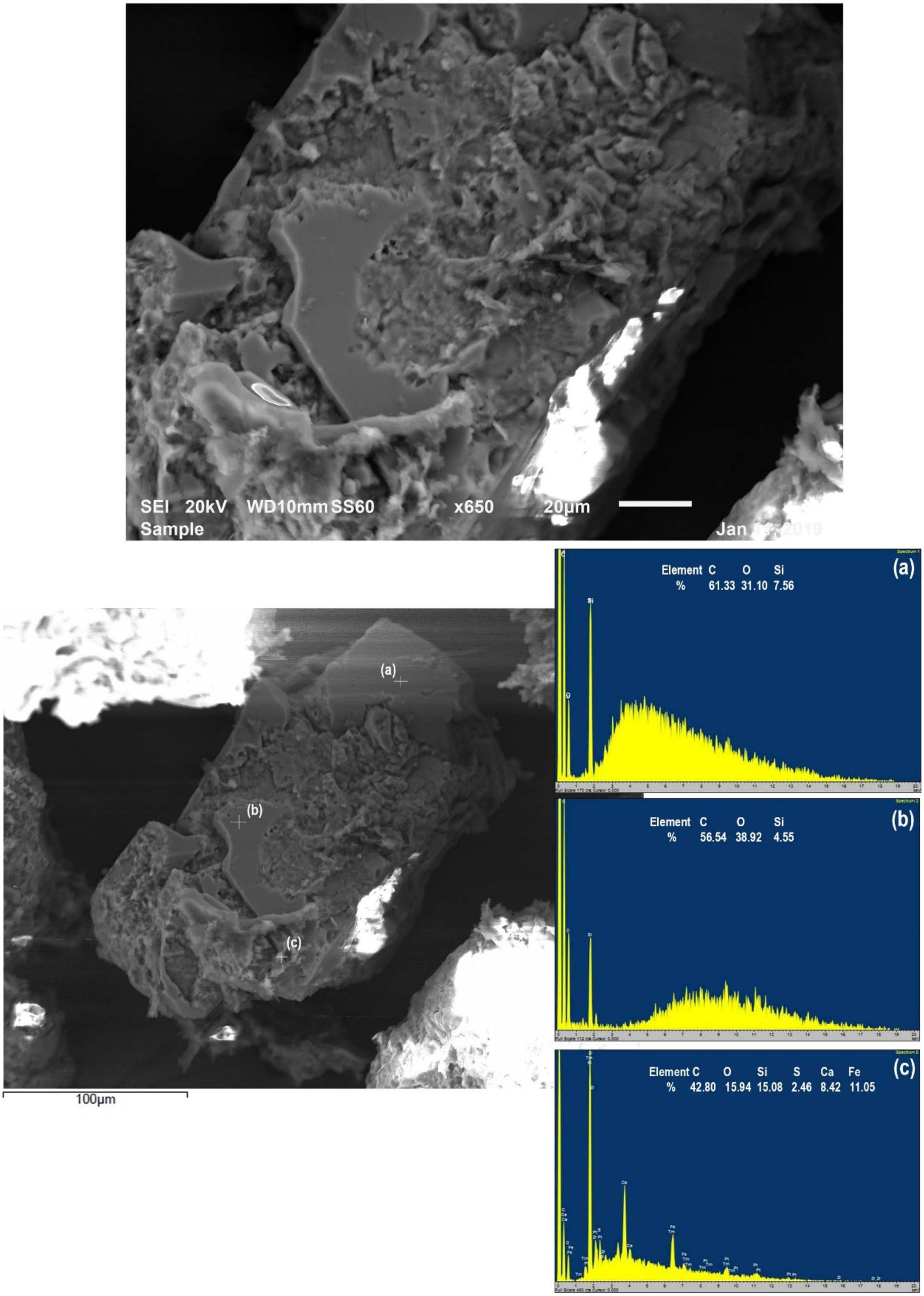


Fig. 10 SEM and EDS analysis at locations (**a**), (**b**) and (**c**) of MICP treated peat (20% sand; 28 days curing)

calcite polymorph was seen the presence along with sand as quartz as compared to untreated samples which were observed with only quartz crystal. SEM analysis coupled with X-ray spectroscopy was commonly used to visualise bio-cementation or bio-precipitation on inorganic soil due to MICP (Burbank et al. 2012; DeJong et al. 2006). Mineralisation due to biological effort may lead to different polymorphs of $CaCO_3$ such as calcite, aragonite, vaterite, monohydrocalcite ($CaCO_3 \cdot H_2O$), hexahydrocalcite or ikaite ($CaCO_3 \cdot 6H_2O$) and less favourable amorphous calcium carbonate (Anbu et al. 2016). Calcite and vaterite are the common precipitation with calcite deemed as the primary and thermodynamically stable product of $CaCO_3$ in many MICPs (Anbu et al. 2016; Ganendra et al. 2014; Spanos and Koutsoukos 1998; Stocks-Fischer et al. 1999). Figure 10 shows the micro surface of MICP treated representative peat sample. EDX spectra in Fig. 10a and b showed the presence of sand particle along with peat surface, while EDX spectra in Fig. 10c showed the presence of calcium along with silica suggesting calcium carbonate precipitation on sand and peat fabric. This suggested that bio-cementation occurred bridging sand and peat fabric which results in strength improvement. This in turn leads to bio-clogging which may explain the reduction of permeability.

Conclusion

MICP of peat, normally waterlogged, has been investigated using bacteria strains P19 and P21 isolated from tropical peat. At both concentrations of 10^5 and 10^6 CFU/mL, UCS for peat mixed with 25% sand using strain P19 and P21 is similar to that using indigenous bacteria. The UCS is double at bacteria concentration of 10^7 CFU/mL as compared to UCS contributed by bacteria concentration of 10^5 CFU/mL and 10^6 CFU/mL. Meanwhile, about 30% increment in UCS when bacteria concentration was increased from 10^7 to 10^8 CFU/mL implies not all the added bacteria contributed to bio-cementation. A maximum UCS of 82.05 kPa was measured with bacteria strain P21 at concentration of 10^8 CFU/mL for peat with 25% sand and 2 mol/kg cementation reagent dosage. The cementation reagent dosage of 4 mol/kg has a detrimental effect to MICP and resulted in significant reduction in strength similar to untreated peat. It is found that both UCS and calcium carbonate precipitation for treated peat were seen to be increased with increasing sand content. UCS and calcium carbonate precipitation increase with curing period indicating bio-cementation may occur under submerged condition up to 28 days. As expected, the permeability of the treated test specimen of stabilised peat is reduced with MICP treatment and increased with sand content. The reduction in permeability up to 28 days for both 25% sand and 50% sand is similar which ranged approximately from 50 to 70%, while 75% sand indicates a smaller reduction around 30%. XRD and SEM–EDS showed that the presence of calcite in MICP treated specimens bridging peat and sand particles improving UCS and reducing permeability due to clogging. The positive findings of this research work prove the possibility of MICP of peat with filler effect of sand and variation of bacteria and cementation reagent dosage under a submerged condition.

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Declarations

Conflict of interest The authors declare no competing interests.

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