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To cite this article: M Umar et al 2019 IOP Conf. Ser.: Mater. Sci. Eng. 527 012005

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# Performance evaluation of lime and microbial cementation in residual soil improvement

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Abstract: Microbially Induced Calcite Precipitation (MICP) is a technique that utilizes the concept of microbial involvements in calcium carbonate precipitation within the soil matrix structure. This leads to the cementation of the soil particles and consequently improving the strength and stiffness of the soil. In this study microbial carbonate precipitations were induced in tropical residual soil via urea hydrolysis. An isolate of urease active strain of Klebsiella pneumoniae UM123 was used to precipitates calcite into the soil with the aim of improving the engineering properties of the soil. Bacteria concentrations of  $2.9 \times 10^6$  cfu/ml and 0.5 M cementation reagents concentrations were used to evaluate the strength and hydraulic conductivity of the soil. Treatment durations of 24, 36, 48 and 60 hours were used in the study. The results obtained indicated a general increase in the strength and reduction of hydraulic conductivity of the treated soil with the increase in treatment durations up to 48 hours. It was also revealed that the higher the amount of calcite precipitated the more the strength improvement and reduction of hydraulic conductivity. Appropriate percentage of lime that satisfied the initial consumption and fixation capacity of the soil sample was found to be 6%. Though, combination of MICP with lime does not significantly improve the strength at early stage of the treatment, it has substantially reduced the hydraulic conductivity of the treated residual soil particularly at the early curing period when lime alone increases the hydraulic conductivity.

Keywords: Microbial cementation, lime, residual soil, strength improvement

#### **1. Introduction**

Microbial activities leading to biomineralization processes are generally believed to be active in every environment on earth surface [1-3]. Hence, the interaction of microbes and formations of carbonate minerals at near-surface of the earth are predominantly high when compared with the subsurface. This is because the near surface of the earth is richer in minerals and the pore fluids that serves as nucleation sites [4]. Soil microorganisms and other aqueous media are found to be largely responsible for the precipitations of calcium carbonate either in natural or experimental settings. Therefore, microbial involvement in biomineralization processes has been considered as the most important factor in the formation of carbonate sediments and soil carbonate deposits [5-7].

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The natural formations of some variety of minerals such as silicates, phosphates, and carbonates have been recognized to be influenced by microorganisms [8]. Microorganisms are generally believed to influence precipitation of carbonate minerals by locally modifying the geochemical conditions and serving as potential nucleation sites [9]. Therefore, microorganisms primarily promote the precipitations of calcium carbonates through their metabolic processes in both natural environment and experimental setup. The ability of microbes to precipitates calcium carbonates minerals provides an opportunity to geotechnical engineers to explore into the possibility of utilizing the concept in soil improvement process; with the aim of finding alternative to traditional methods. This is because the traditional methods usually require high amounts of energy, costs, have limitations with regards to treatment range and sometimes require materials which have considerable impact on the environment [10, 11].

Hence, Microbially Induced Calcite Precipitation (MICP) method is a relatively new, sustainable and environmentally friendly technique in soil improvement processes. The technique utilizes the microbial activities that are native to the soil to alter and improve the ground condition [10]. Soil improvement using MICP can meet the green construction requirement as this treatment causes minimal disturbance to soil environment [12]. Majority of the studies conducted on bio-mediated ground improvement were based on urea hydrolysis. This is because urea hydrolysis is regarded as the most easily controlled carbonate generating reactions; and has the capability of generating high concentrations of carbonate within a short period of time. Micro-organisms containing the enzyme urease are mainly used in this technique.

Urea hydrolysis has been among the first processes linked with microbial carbonate precipitation in the late nineteenth century [1]. Hence, in the late  $20^{th}$  century the process attracted numerous industrial applications that include bioremediation [13-15], treatment of wastewater [16] and refurbishment of calcareous stone materials [17]. Similarly, studies on the improvement of strength of concrete [18], plugging of rock pores for enhanced oil recovery [19] and soil improvement have been successfully conducted. Urea hydrolysis generally follows a series of chemical reactions that leads to the formation of ammonia ions (NH<sub>4</sub><sup>+</sup>) and carbonate ions (CO<sub>3</sub><sup>2-</sup>). The carbonate ions then react with the dissolve calcium from the supplied calcium chloride to form calcium carbonate crystals. The chemical reactions are presented in Equations 1 to 3.

$$\begin{aligned} H_2 N - CO - NH_2 + 2H_2 O &\to 2NH_4^+ + CO_3^{2-} \\ CaCl_2 &\to Ca^{2+} + Cl^{2-} \end{aligned} \tag{1}$$

$$Ca^{2+} + CO_3^{2-} \to CaCO_3 \downarrow \tag{3}$$

However, once the calcium carbonate is precipitated it binds the soil particles together and will only dissolve very slowly, either when continuously flushed by acidic groundwater or as a result of acidifying processes in the pores. Hence, when sufficient calcium carbonate is precipitated, durable soil improvement can be achieved [20], but this may be costly. Meanwhile, lime stabilization precipitates calcium silicate hydrate gel which is stronger than calcite; that coat and bind the soil particles together to provide long term strength. Therefore, since microbial cementation provides an immediate strength improvement with uncertainties in its long term durability; combining the two techniques is expected to provide both immediate and long term strength improvement through the precipitation of calcite and calcium silicate hydrate. Hence, this study evaluates the combined effects of bacteria and lime on the strength and hydraulic conductivity of MICP treated residual soil.

#### 2. Materials and method

#### 2.1 Bacteria isolation, cultivation and cementation reagents preparation

The urea hydrolyzing bacterium was isolated from the soil sample used in the study and was identified and named as *Klebsiella pneumoniae strain UM123* and tested for urea hydrolysis. The isolated bacteria were inoculated into Stuart's urea broth containing phenol red which indicated a pH change

associated with ammonium production from urea hydrolysis reaction. The colour changed from yellow-orange to pink was an indication that urea hydrolysis occurred. The strain was then cultivated in American Type Culture Collection (ATCC) specific yeast extract–based medium containing 20g yeast extract, 10g ammonium sulphate in 1 Litre 0.3 M Tris buffer solution at pH 9.0. After 24 hours incubation at 30 °C, the culture was then harvested and stored at 4°C prior to use. The bacterium was grown to its late exponential growth phase and concentrations of  $2.9 \times 10^6$  cfu/ml at optical density (OD<sub>600</sub>) was obtained and used in the study. The cementation reagents consist of 3g nutrient broth and 0.5 M concentrations of CaCl<sub>2</sub> and urea.

## 2.2 Calcite contents determination

The calcite contents of the MICP treated samples were determined using gravimetric analysis of acidified samples. 10g of the powder sample was used after oven drying at 105°C for 24 hours. Hydrochloric acid 2 M was added to the prepared powdered sample and carbon dioxide was liberated due to the reaction between calcite and hydrochloric acid. The residue was collected and oven dried again and the loss in weight was used to estimate the percentage of calcite contents in the specimen. The methods was based on the assumption that the increment of carbonate content in the soil after the MICP treatment was purely caused by the formation of calcium carbonate. The method was adopted from a study by [21].

# 2.3 Initial consumption of lime

Initial consumption of lime test was conducted to determine the appropriate quantity of lime that is required to be added to the soil to achieve a pH value of 12.40 at 25°C. This pH is required to facilitate reaction between the lime and the clay components in the soil to be stabilized.

# 2.4 Lime fixation capacity

For efficient lime stabilization, the lime that would be added to the soil must first satisfy an affinity of the soil for lime. This affinity is referred to as lime fixation. The lime fixation capacity corresponds with the lime percentage where further addition of lime does not bring about further changes in the plastic limit of the soil. The test was conducted by adding varying percentage of lime to the soil and determining the plastic limit of the soil until no further increase in the plastic limits were observed.

# 2.5 Lime stabilization

Based on the results of the initial consumption of lime and lime fixation capacity tests; 6% lime by dry weight of the soil was added into the soil specimens and properly mixed with water to the optimum moisture content of the soil. The soil-lime mixture was then compacted into a 50mm diameter and 100mm height prefabricated steel mould to a maximum dry density of 1.390 Mg/m<sup>3</sup>. The compacted soil-lime mixtures were then extruded and cured for 3, 7, 14, 21 and 28 days before the Unconfined Compressive Strength tests. However, some specimens were tested for the strength improvement immediately after the lime treatment before curing.

# 2.6 Lime and microbial cementation experiments

Lime and microbial cementation experiments were conducted in two stages; the first stage involved mixing the residual soil specimens with a liquid medium containing the microorganism (*Klebsiella pneumoniae strain UM123*) at a concentration of  $2.9 \times 10^6$  cfu/ml. The cementation reagents (urea, calcium chloride and nutrient broth) were then sprayed to the soil-bacteria mixture at 6 hours interval for 48 hours to the optimum moisture content of the soil (31.2%); while curing at atmospheric temperatures. The second stage involves mixing the soil-bacteria-reagents mixture with 6% lime and compacted into a 50mm diameter and 100mm height prefabricated steel mould to a maximum dry density of 1.390 Mg/m<sup>3</sup>. The compacted soil-bacteria-lime mixtures were then extruded and cured at the same conditions with the lime stabilized residual soil before the Unconfined Compressive Strength tests. On the other hand, similar procedures of lime and microbial cementation experiments were also

adopted on the soil specimens prepared for hydraulic conductivity determination. However, the prepared specimens were compacted in a 100mm diameter and 110mm height prefabricated steel mould for hydraulic conductivity tests.

# 3. Experimental results and discussion

# 3.1 Residual soil

The tropical residual soil used in the study was collected from a site at Universiti Teknologi Malaysia UTM, Johor Campus. Preliminary laboratory data for classification purpose that comprised of index and engineering properties of the soil were obtained. The soil sample was classified as Gravelly clay of high plasticity (CHG) based on British Soil Classification System (BSCS). Table 1 presents the Index and engineering properties of the soil.

Properties	Description
Gravel (%)	32
Sand (%)	10
Silt (%)	32
Clay (%)	26
Moisture content (%)	36
Liquid limit (%)	71
Plastic limit (%)	47
Plasticity Index (%)	24
Specific gravity	2.62
MDD $(Mg/m^3)$	1.390
OMC (%)	31.2
Classification(BSCS)	CHG
UCS (kPa)	30.4
Hydraulic conductivity (m/s)	2.35E-06

**Table 1.** Index and Engineering Properties of the Soil Sample.

#### 3.2 Initial consumption of lime

The initial consumption of lime test was conducted on the residual soil sample with the aim of establishing the optimum lime content required to be added to the soil sample to attain a pH of 12.4. Figure 1 presents the percentage of lime required to satisfy the initial and flocculation reactions of the residual soil used in the study as obtained from the initial consumption of lime test. It was found that to attain a pH of 12.4; lime content of 6% is required.



Figure 1. Initial Consumption of Lime (ICL) of the residual soil sample.

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When the lime was mixed with the soil; the water present in the soil causes the dissolution of lime thereby inducing a strongly alkaline medium. As such the divalent calcium cations of lime are dissolved. In the first moments of the lime treatment, calcium ions fixes to the surface of the clay mineral and replaces most of the available exchangeable cations of the clay minerals [22]. The alkaline environment is responsible for the slow dissociation of silica and alumina from the clay mineral structure, which then reacts with the lime resulting in the formation of cementitious calcium silicate hydrate and calcium aluminate hydrate gels that bond the soil particles together [23-25].

# 3.3 Lime fixation capacity

Lime fixation capacity of a soil refers to the affinity of the soil for lime and plastic limit of a soil is used to indicate the amount of lime fixation in clayey soils. The lime fixation capacity corresponds with the lime percentage where further addition of lime does not bring about further changes in the plastic limit of the soil. The test was conducted by adding varying percentage of lime to the soil and determining the plasticity properties of the soil until no further increase in the plastic limits were observed. Hence, as reported in many studies lime has the ability to change the plasticity of a soil after treatment. As such, detailed study of the changes in the soil plasticity properties attributed to lime amendment will assist in understanding the plasticity behaviours of the lime-stabilized soils.

Figure 2 presents the relationship between plastic limits of the residual soil with the percentage lime contents. The plastic limits increases with increase in the lime content up to 6% lime, beyond which the increase in the plastic limit was relatively constant; except for 28 days curing which the plastic limit continue to increase with the increase in lime content. Hence, the continuous increase of plastic limit at all lime contents at 28 days curing may be as a result of some physicochemical reactions between the soil and lime since the phenomenon only takes place at prolonged curing period. The plastic limit of a soil can be described as the measure of cohesion of the soil particles against cracking when the soil is remoulded. The shear resistance and cohesion between the soil particles should be low enough to allow the individual particles slides over one another easily.



Figure 2. Variation of plastic limits with lime content.

On the other hand, the interparticle shear resistance should be enough to hold the soil mass together in remoulded state. Hence, the plastic limit is a measure of the water content of soil when it approaches a certain shear resistance. With the addition of lime, the thickness of the diffuse double layer decreases, which increases the charge concentration and thereby the viscosity of the pore fluid. As a result, the interparticle shear resistance increases, leading to a sharp increase in the plastic limit. Moreover, the lime-induced flocculation enhances the interparticle resistance against movement,

leading to an increased plastic limit. Therefore, the plastic limit does not change much when the lime content is increased beyond 6%.

#### 3.4 Effect of treatment durations on the MICP process

Effect of treatment duration on the MICP process was evaluated using 24, 36, 48 and 60 hours treatment periods. Figure 3 presents the correlation between unconfined compressive strength, calcite content and treatment duration. The unconfined compressive strength of the residual soil increases with increase in treatment duration up to 48 hours. The unconfined compressive strength attained its highest value of 63.7 kPa at 48 hours treatment duration. However, at longer treatment duration beyond 48 hours the strength of the treated soil did not further improve. It was therefore observed that beyond 48 hours treatment duration, the strength generally declined. Hence, the declined in strength after prolonged treatment duration of more than 48 hours may be attributed to the decays in urease activity with time, due to cell lysis, porosity reduction due to calcite precipitations and wash out of urease from the soil.



Figure 3. Correlation between UCS, calcite content and treatment durations.

Similarly, it was also observed that the calcite content continue to increase with the increase in treatment durations up to 48 hours; beyond which it decreases. This may be due to the decline in bacterial activity or factors such as decay of the urease activity with time, wash out of urease from the soil due to continuous injection of the reagents. These findings are in good agreement with the study conducted by [26] who suggested that longer treatment duration of more than three days could have adverse effect on the microbial activity and strength development.

#### 3.5 Strength properties of the residual soil treated with lime and microbial cementation

Figure 4 presents the relationship between curing time in days and strength improvement of the MICP treated, lime stabilized residual soil and combination of lime and microbial cementation treatment. The strength improvement of the microbial cementation was peaked at 7 days. Conversely, the strength of lime stabilized residual soil generally improved with increase in curing time up to 28 days. The pozzolanic reaction that takes place between lime and clay particles of the soil was solely responsible for the cementation that substantially improve the long term performance of the stabilized soil [27, 28].

Hence, in order to evaluate the combined effects of MICP and lime stabilization, appropriate percentage of lime (6%) was added to the MICP treated residual soil and tested for strength immediately without curing. Therefore, an immediate strength improvement of 22% relative to lime

stabilized residual soil was obtained. After 3 days of curing the strength of the soil-bacteria-lime mixture improved by only 12%. On the other hand, the strength of the combined MICP and lime stabilization was slightly lower than the strength of the lime stabilized residual soil after 7 days of curing. Thereafter, the strength of the combined MICP and lime stabilization continues to decrease with increase in curing time. This clearly indicates that MICP treatment can only complement the strength improvement of lime stabilization at very early stage.



**Figure 4.** Correlation between curing time and strength improvement of the MICP treated, lime stabilized residual soil and combination of lime and MICP treatment.

The declined in strength of the combined MICP and lime stabilization after prolong curing period may be attributed to the inability of the microorganisms to survive under very high alkaline environment; as the pH of the soil rises to 12.4 when 6% lime was added. Under normal condition, MICP treated residual soil continues to gained strength when cured for up to two weeks after treatment. This is because the microbial activity continues to utilize the remaining nutrients and reagents to precipitates additional calcites until completely exhausted. But with the addition of lime the environment becomes too alkaline for ureolytic bacteria's survival and activities.

3.6 Hydraulic conductivity properties of the residual soil treated with lime and microbial cementation Hydraulic conductivity of the residual soil was also evaluated using lime stabilization, MICP treatment and combination of the two processes. Treated specimens were cured for 1, 3, 7, 14, 21 and 28 days before testing. Figure 5 presents the correlation between hydraulic conductivity of the MICP treated, lime stabilized and combination of MICP and lime stabilized residual soil with curing time. It was also observed that the hydraulic conductivity of the MICP treated residual soil decreases with increase in the curing time up to 7 days; thereafter the hydraulic conductivity remains constant. However, for the lime stabilized residual soil, the hydraulic conductivity initially increases at the early curing period up to 7 days. Hence, after 7 days of curing the hydraulic conductivity started to decrease with increase in curing period up to 28 days. Similarly, short term increase in permeability was reported by Townsend and Klym [29] after treating heavy clay with lime. Hence, it was also revealed that the possibility of increased hydraulic conductivity can as well be reverse in the long term when the calcium aluminates hydrate and calcium silicate hydrate gels are formed; as they subsequently crystallise and fill in the pore spaces of the treated soil [30]. This theory was reinforced by this study and the research of [31] who reported that the permeability of a lime-fly ash-aggregate canal liner showed initial increases in permeability which gradually decreased to produce flow rates comparable with the natural clay.



Figure 5. Correlation between curing time and hydraulic conductivity of the MICP treated, lime stabilized residual soil and combination of lime and MICP treatment.

On the other hand, for the combination of MICP with lime stabilization, the hydraulic conductivity decreases by 26%, 40% and 61% when cured for 1, 3 and 7 days respectively. Hence, the hydraulic conductivity remains constant for the remaining curing period. The immediate decrease in the hydraulic conductivity is attributed to the calcite precipitation via urea hydrolysis that filled in the soil voids and decreased the permeability of the soil. Though, combination of MICP with lime does not significantly improve the strength at early stage of the treatment, it has substantially reduced the hydraulic conductivity of the treated residual soil particularly at the early curing period when lime alone increases the hydraulic conductivity.

#### 4. Conclusions

The longer the treatment duration the higher the amount of calcite precipitated and the more the strength improvement up to 48 hours treatment duration. However, the bacterial activity declined after 48 hours treatment period.

Appropriate percentage of lime that satisfied the initial consumption and fixation capacity of the soil sample was found to be 6%.

The combined lime and microbial cementation produced an immediate strength improvement of 22% relative to lime stabilized residual soil. However, the strength of the treated soil decreased with continues increase in curing time.

Though, combination of MICP with lime does not significantly improve the strength at early stage of the treatment, it has substantially reduced the hydraulic conductivity of the treated residual soil particularly at the early curing period when lime alone increases the hydraulic conductivity.

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## Acknowledgement

This research was financially sponsored by the Ministry of Education Malaysia through Fundamental Research Grant Scheme (FRGS); Grant No. 4F628.