

ARID ZONE JOURNAL OF ENGINEERING, TECHNOLOGY & ENVIRONMENT

AZOJETE June 2021. Vol. 17(2):185-196 Published by the Faculty of Engineering, University of Maiduguri, Maiduguri, Nigeria. Print ISSN: 1596-2490, Electronic ISSN: 2545-5818 www.azojete.com.ng



#### **ORIGINAL RESEARCH ARTICLE**

## APPLICATION OF MICROBIAL INDUCED CALCITE PRECIPITATION (MICP) TECHNOLOGY IN GEOTECHNICAL AND GEO-ENVIRONMENTAL ENGINEERING: A REVIEW

I. S. Muhammad\*, B. B. Kurna and B. S. U. Ibn Abubakar

Department of Civil and Water Resources Engineering, University of Maiduguri, Maiduguri, Borno State, Nigeria \*Corresponding author's email address: <u>ibrahimcivil73@gmail.com</u>

#### ARTICLE INFORMATION

ABSTRACT

Submitted 2 Nov., 2020 Revised 24 Januray 2021 Accepted 27 Januray 2021

Keywords:
Soil improvement
Remediation
Microorganism
Urease
Calcite
Cementation

One of the most recent areas of research in geotechnical and geo-environmental engineering is bio-mineralization, a natural process in living organisms, also known as Microbial Induced Calcite Precipitation. It is the formation of calcium carbonate from a supersaturated solution due to the presence of microbial cells and biochemical activities. In the process, microbes secrete metabolic products that reacts with ion in the medium to precipitate minerals. Through this process, soil improvement/remediation have been investigated and proven reliable with minute carbon print as compared to conventional binders. This paper presents an overview on the wide application of Microbial Induced Calcite Precipitation in the areas of geotechnical and geo-environmental engineering with the mechanisms and factors influencing its performance explained. The work has also considered both laboratory and field scale researches conducted in these areas. The key contribution of this work is the compilation of different approaches in soil stabilization and remediation, via urease producing microbes, in single source. It also outlined different treatment dosages based on environmental conditions suitable to soil types

© 2021 Faculty of Engineering, University of Maiduguri, Nigeria. All rights reserved.

## I.0 Introduction

The use of soil as engineering material for variety of constructions dates back to ancient time where different types of soil are used for different purposes. In some ancient civilizations clay was used as construction materials for architectural works. However, with the advancement of civilization and discovery of natural pozzolanic materials such as volcanic ash, good improvement in strength was attained (Dan Babor *et al.*, 2009). After the industrial revolution, ordinary Portland cement became most accepted and widely used material due to its high strength, durability, workability not only for building construction but also for soil amelioration (Maclaren and White, 2003).

Due to heterogeneity in soil mass within short span, vertically and horizontally, it is very common to encounter unsuitable soil during site investigation for civil engineering works. In such situation, an engineer is left with the options of either bypassing the bad soil using piles, replacing the soil with appropriate material, redesign the structure to suit the condition or improve the soil by altering its engineering properties (Venkatramaiah, 2006). Soil improvement is primarily categorized into mechanical and chemical methods. Mechanical improvement includes processes such as compaction, consolidation, surcharging and others with a view to increase the strength of the soil. The chemical improvement on the other hand employs the use of additives or binders such as cement, lime, natural and artificial pozzolanas to treat a soil either by direct mixing or injection of grout (Sherwood, 1993). Over the years, several geotechnical works have been successfully implemented using

Portland cement worldwide. However, despite its benefits and wide application, the use of cement has posed a serious environmental concerns.

The major concern lies in the amount of carbon dioxide  $(CO_2)$  emission as a result of cement production which immensely increases the concentration of greenhouse gases in the atmosphere. Carbon dioxide which is the major reason for the greenhouse effect causing global warming, is a byproduct of a chemical conversion process used in the production of clinker, a component of cement, in which limestone  $(CaCO_3)$  is converted to lime (Zulfequar, 2017). The simplified stoichiometric relationship is given in equation (1) below:

$$CaCO_3 + Heat \rightarrow CaO + CO_2 \tag{1}$$

The calcination of calcium carbonate during the production of cement liberates about 0.55 tons of  $CO_2$  per ton of Portland cement which is further emitted into atmosphere. In addition, the huge amount of fossil fuel that is burnt to power the cement plants also emit  $CO_2$  at a rate of 0.4 tons per ton of cement produced (Ilhan *et al*; 2016). In a note shell, the total emission of  $CO_2$  into atmosphere in the production of I ton of Portland cement accumulates to 0.95 tons, and there are more than 2,500 cement power plants worldwide. According to "global carbon budget 2017"by Corinne *et al*; 2018, cement production and burning of fossil fuel accounts for 9.9±0.5 giga-tons of  $CO_2$  emission annually, which is about 10% of the global  $CO_2$  emission.

Other environmental concerns associated with the use of cement include rise in soil pH up to 12-13 due to alkaline hydroxide ion release as a byproduct of hydration, which in turn affects biological organisms by undermining their essential natural activities (Taylor, 1997). The irreversible hydration reaction from cement, produced impervious top soil which affects vegetation growth and increases surface runoff in urban settings adding to flooding (Hansen, 2002 and Rao et *al.*, 2007).

This serious environmental concern from cement production and its use have prompted researchers over the years toward exploring eco-friendly and sustainable alternatives for cement in various civil engineering endeavors. In respect to this, different methods have been explored including the use of geosynthetics which have high tensile strength, flexibility and impervious characteristics and are been applied in soil reinforcement, separation, drainage among others. However, their use revealed high dependent on the material rather than the soil thereby limiting its various geotechnical applications (Ilhan *et al.*, 2016).

The current advancement in technology and deep knowledge in the field of biology and microbiology has made accessible new investigation pathways in geotechnical research and innovation. One of the most promising biological techniques that emerged in the 21<sup>st</sup> century which was first suggested by Mitchell and Santamarina (2005) and deeply explored by De Jong *et al.* (2006), is the Microbial Induced Calcite Precipitation (MICP). Its efficiency in tackling number of geotechnical and geo-environmental problems have been successfully investigated and established in both laboratory and field scale with minute carbon footprint compared to cement (Amin *et al.*, 2017).

#### 2.0 Microbial Induced Calcite Precipitation Mechanism

Microbial Induced Calcite Precipitation (MICP) technique is considered eco-friendly ground improvement and soil reinforcement technique (Atticus *et al.*, 2012). According to Bosak (2011), MICP is the formation of calcium carbonate from supersaturated solution following the presence of microbial cells and bio-chemical activities. The technique is generally based on urease-producing bacteria known as *Sporosarcina pasteurii* (formerly *Bacillus pasteurii*) and their like. It is a soil, non-pathogenic and endospore producing bacteria with an optimum pH for growth of 9.0 that can tolerate extreme conditions (Weil *et al.*, 2012 and Periasamy *et al.*, 2016).

The process involves introducing aerobically cultivated bacteria with highly active urease enzymes into soil. The urease enzyme hydrolyzes urea into one mole of carbonate and two moles of ammonia per mole of urea (Malcolm *et al.*, 2012) as presented in the reactions below:

$$CO(NH_2)_2 + 2 H_2O \rightarrow 2 NH_4^+ + CO_3^{2^-}$$
 (2)

In the presence of  $Ca^{2+}$  derived from calcium source, mostly  $CaCl_2$ ,  $Ca^{2+}$  reacts with carbonate ions ( $CO_3^{2^-}$ ) to form one mole of calcium carbonate ( $CaCO_3$ ) crystals throughout the soil matrix, see equation below:

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

The introduction of bacterial suspension solution plus the injection of calcifying solution carrying the urea and calcium into the soil mass are essential for completing the process. The urea provides energy for the growth of microbes and adequate carbonate ions while calcium solution supplies calcium ions for rapid generation of calcium carbonate precipitation within the matrix (Fatima-Zahra and Benoit, 2018). This occur due to the reaction of ammonia with water producing hydroxyl ion (OH<sup>-</sup>) resulting in pH rise in the media which further facilitate precipitation of calcium ions into CaCO<sub>3</sub> (Mizanur and Neera, 2017). This bio-induced calcite fills pore spaces in the soil and bind soil grains together forming concrete material (Malcolm *et al.*, 2012). Through this process, geo-environmental and geotechnical problems such as remediation, liquefaction of soils, slope stability, erosion control, carbon-dioxide sequestration, healing of rock fractures were achieved.



Figure 1: Ureolysis-driven calcite precipitation (Source De Muyeernck et al., 2010)

## 2.1 Factors Controlling MICP Performance

The performance of bio-calcification in MICP technology depends on the amount and spatial distribution of CaCO<sub>3</sub> precipitation and other environmental factors. These include the following:

# 2.1.1 Bacteria Type and Concentration

Bacteria type and their cell concentration which serves as nucleation sites for CaCO<sub>3</sub> precipitation are necessary for this process. The most common in MICP is genera bacillus and Sporosacina which catalyst urea hydrolysis (Ng et al., 2012). Bacteria cell concentration is directly proportional to urea hydrolysis. Harkes et al. (2010) reported an increase in urease activity and hydrolysis rate with increasing bacterial concentrations, which also corroborates with an investigation by Okwadha and Li (2010).

# 2.1.2 Nutrient

Bacteria nutrients are the source of energy for metabolic activities and essential for calcite production. For effective MICP, nutrients have to be provided for their effective growth and activities. The most common are N, P, K, Mg, Ca and Fe which are supplied during culture and soil treatment stages (Mitchell and Santamarina, 2005; Saranya et al., 2017).

## 2.1.3 pH

pH level also dictates the urease activity, MICP begins at pH 8 and escalate to 9 where urease activity is high (Dupraz et al., 2009). A high pH, is very important for ammonia production by urea hydrolysis. Aerobic bacteria release CO<sub>2</sub> through cell respiration, which is paralleled by an increase in pH due to ammonia production (Ng et al., 2012). At low pH carbonates tends to dissolve instead of precipitating in the media (Cheng et al., 2014).

#### 2.1.4 Soil-microbe Geometric Compatibility

Another important factor is the geometric compatibility between soil and microbes. Soil microbes normally travel within the soil through the pore throat between soil particles, either by self-propelled movement or by passive diffusion. Their sizes ranges from 0.5 to 3.0 µm which means significant amount of silt and clay can hinder their distribution and movement (Ng et al., 2012; Saranya et al., 2017). It is therefore important to take into account the type of soil and its pore throat when selecting bacteria for MICP treatment.

#### 2.1.5 Temperature

Temperature similarly plays a vital role in the catalysis of urea by urease at a range between 27 to 30° C (Mitchell and Ferris, 2005; Okwadha and Li 2010). The urea hydrolysis is stable at 35° C and increase in temperature beyond this does not promote the enzyme activity (Dhami et al., 2014).

# 2.1.6 Concentration of Reactants

The concentration of reactants, urea and Ca<sup>2+</sup>, are well vital for calcite precipitation. Due to the negative charge surface of the microbial cell, they scavenge cation, Ca<sup>2+</sup> for example, which consequently make them ideal nucleation sites for heterogeneous crystal formation. They attract metal cations  $(Ca^{2+})$  and subsequently counter ions  $(CO_3^{2-})$  to precipitate as calcium carbonate on their surface (Philips et al., 2013). However, Okwadha and Li (2010), Corresponding author's e-mail address: ibrahimcivil73@gmail.com

reported that high concentration of calcium chloride and urea above 0.5M decrease the efficiency of calcite precipitation. De Muynck et al. (2010) and Murtala and KhairulAnuar (2017) observed that the best concentrations of urea and calcium chloride for calcite precipitation are 0.5 and 0.25 M respectively. However, the latter is the most widely implemented and reported concentration limits yielding huge and efficient calcite precipitation.

# 2.1.7 Methods of Introducing Bacterial cells and Cementitious Solution

The method of introducing bacteria and cementitious solution into soil plays an important role in the accomplishment of MICP process. Methods such as spraying, mixing, injection or their combination have been used in different scenarios and conditions. In spraying method, which is usually associated but not limited to non-cohesive soils, the bacteria solution is sprayed over the soil at a given rate and followed by spraying of cementitious solution. Prior to this, the soils are saturated with water to allow the microbes adhere to soil particles (Zhaoyu et al., 2018). Mixing method involves mixing an air-dried soil sample with bacterial solution thoroughly and introducing it to desired mould. This is followed by introducing the cementitious solution by injection or spraying (lawad and Zheng, 2016). Injection method usually comes in different manner. This includes mixing bacteria cells and cementitious solution together before injecting into the soil, injection of bacteria solution first followed by cementitious solution and staged injection with or without retention period between phases (Saranya et al., 2017).

# 2.2 Applications of MICP

Ng et al. (2012) investigated the potential of MICP in the improvement of shear strength and permeability of sandy silt. The soil was compacted in a 50mm diameter and 150mm length fabricated mould. Bacillus Megaterium (5  $\times$  10<sup>7</sup>cfu/ml) was mixed with the air dried soil sample prior to the compaction. The cementation reagents used are urea and calcium chloride at a concentration of 0.2M which was injected into the compacted soil at an interval of 6hrs for 48hrs duration at flow rate of 1.7x10<sup>15</sup>m/s. The result revealed a significant improvement in both shear strength and the permeability. The improvements were 164% shear strength increment and 74% reduction in permeability compared to the natural soil.

Dejong et al. (2014) successfully implemented a field scale study on sand covering 3m×3m plot targeting a treatment depth of 15cm. The study uses five different injection/extraction wells across the area and the sand treatment was carried out in two phases. In phase one, Sporosarcina pasteurii suspension was added into a urea-rich solution and injected via the wells. Non continuous injection and extraction of the solution was conducted for up to 50hr for uniform distribution of the solution. In phase two, cementation solution was injected at high flow rate in 1hr followed by 2hrs rest period until uniformity is attained.

Surficial application of MICP was also investigated by Gomez et al. (2015), on a field scale in loose sand deposits at mine site, Saskatchewan, Canada. In the experiment, bacterial culture and nutrient amendment was sprayed on a four (4) 2.4m × 4.9m plots named TPI, TP2, TP3 and TP4 where TP1 was the control. Across the plots TP2 to TP4, different urea and calcium chloride concentrations were applied to observe the cementation efficiency. Sporosarcina pasteurii (ATCC 1376) with different dosages of nutrient broth, urea and CaCl<sub>2</sub> Corresponding author's e-mail address: ibrahimcivil73@gmail.com

were applied at a flow rate of 19L/min on the plot surfaces using two tanks of volume 5,678L (each plot received 568L) each with totes, pumps and spray nozzle attached to the setup. Treatment was conducted in twenty consecutive days with four-day circle of five identical days. After the 20-day period of treatment, TP4 formulation gave the optimum result of 28cm depth soil improvement and 2.5 cm thick cemented crust having good resistance to erosion.

In their study, Firas and Zheng (2016) conducted laboratory experiment on the Unconfined Compressive Strength (UCS) and permeability of grinded silica sand classified as poorly graded sand. The sample was prepared by packing it into polyvinyl chloride (PVC) column of 80mm high and 37mm inner diameter. *Sporosarcina Pesteurii* cultivated under sterile aerobic batch condition in a yeast extract was used in the study. Samples of sand columns were divided into two groups of water saturated and unsaturated. Both groups were then flushed with 33ml of bacterial solution followed by 33ml of cementitious solution after a retention time of 3hrs using gravity induced downward precipitation at flow rate of 0.150l/h. Flushing with cementitious solution was repeated after every 24hrs for 7 days. Unconfined compressive strength of up to 3000 KN/m<sup>2</sup> and 90% drop in permeability rate were observed. The study also revealed that the dry sample of this particular soil had greater compressive strength compared to the water saturated one while reverse case was observed in permeability reduction.

The permeability of Silica sand columns was examined in laboratory using ureolitic bacterium culminating at reduction in permeability value as high as 97% after 24 hrs. The culture medium was prepared using tryptone 15 g/l; soy peptone 5 g/l; NaCl 5 g/l and urea 20 g/l. To the whole medium, which is 1 L, 15 g agar was added. The medium was then autoclaved at 121 °C for 20 min. The culture plates were incubated overnight at 32 °C, and the colonies were transferred into a 50-ml shaking flask with growth solution and shaken for 15 h. Four samples of silica sands were prepared in stainless steel cylinders with a length and inner diameter of 7.5cm and 3.5cm respectively. The sand was washed with HCI solution and rinsed with pure water to get rid of interference of impurities before use. Permeability tests were conducted at 3 different flow rates (0.56, 0.97, and 1.73 ml/min, corresponding to pumping rates of 1, 0.5, and 2 rpm) using pure water to obtain an average conductivity before MICP. Then, 20 ml of bacterial stock solution (approximately 4.44×10<sup>10</sup> cells) was inoculated into each column with a pumping rate of 0.97 ml/min of pure water, and the effluent was collected for cell counting. About 10 pore-volumes later, the MICP solution was then pumped into the columns at a speed of 0.97 ml/min until the test was stopped 72 h later. Hydraulic tests were conducted every 12 h during the MICP test. Calcite crystals filling pores and coated on sand surfaces was shown to be the reason for the reduction subsequent to Scanning Electron Microscopy (SEM) analysis conducted before and after the treatment (Yefie et al., 2017).

Murtala and Khairul-Anuar (2017) investigated the potential of MICP in the improvement of strength and reduction of permeability of tropical residual soil. An isolate of urease active strain of *klebsiella pneumoniae* from the soil used in the study was used to precipitates calcite into the soil using five cementation reagents concentrations. Bacterial concentration of  $1.5x 10^4$ cfu/ml and  $2.9x 10^6$ cfu/ml were obtained and used for the treatment using injection method. Results revealed increase in the strength of the soil after 48hrs of treatment from

80% to 110% as bacterial concentration increases. The same trend was recorded with increase in reagent concentration up to 0.5M after which the strength declined. There were also a decline in permeability by 50% and 65% after 48hrs from  $1.5 \times 10^4$  cfu/ml to 2.9 x  $10^6$  cfu/ml bacterial concentration respectively. The study revealed 0.5M as the optimum reagent concentration for *klebsiella pneumoniae* for effective improvement of residual soil.

Zhaoyu et al. (2018) reinforced sands for mitigating hazard caused by wind erosion. Series of laboratory experiment was performed to evaluate wind resistance of MICP treated sand. The test sand was air-dried and transferred into a tray of mass (m) with dimension of 288mm × 225mm × 25mm. The thickness and mass of the tray were measured. Five (5) percolation holes of diameter of 8mm were designed and fabricated at the bottom of the tray for drainage. Six-layer gauze was placed inside the tray above the holes to prevent sand and water out of the tray. Pre-determined mass of the dry sand was placed in one layer at the bottom of the tray and compacted using a steel rod. The initial dimension of the sand sample was measured, so the total volume of the sample (V) will be known.

Bacillus cereus (Sporosarcina pasteurii, No. ATCC11859) was used in the study with trypsin, soybean peptone, NaCl, urea, and distilled water as main components of the culture medium whereas the cementation solution was urea-CaCl<sub>2</sub> solution with a concentration of Imol/L. All solutions were introduced into the samples by surface spraying method at a rate of 15ml/min. wind erosion test was conducted in a large area in the laboratory(5m×3m) for uniformity of air flow. At the end of 28days curing period, well fixed samples (to avoid movement) were subjected to test using a duct fan with a rated speed of 2800rpm and an air volume of 3000m<sup>3</sup>/h for a period of 90 minutes. The total mass of the sand specimen and the tray (M<sub>i</sub>) were measured at 5min interval in 30min and at 30min interval between 30min and 90min. The amount of wind erosion ( $\Delta$ M) was then calculated by  $\Delta$ M=M<sub>28</sub>-M<sub>i</sub>; the wind erosion rate (a) was calculated by a= (M<sub>28</sub>-M<sub>i</sub>)/(M<sub>28</sub>-m). cylindrical samples of 30mm diameter and 60mm height were also produced for Unconfined Compressive Strength (UCS) test using the same treatment.

Final results revealed that the erosion rates of sample 2,3 and 4 were considerably reduced by 63.97%, 94.75%, and 97.76%, respectively, as compared to the control sample. The Scanning Electron Microscopy (SEM) results indicated bonding effect from the  $CaCO_3$ crystals as a result of the MICP treatment which gives the top layer of the specimen wind erosion resistance. In addition, the result of UCS value of 4 MPa attained has exceeded the minimum requirement of 1 MPa for wind resistance. The technique provides alternative method for mitigating and preventing desertification.

A surficial soil stabilization against water-induced erosion was explored using MICP technique by Xiangrong et al. (2018). The ASTM C-778 Ottawa sand possessed a particle diameter corresponding to 10% finer ( $D_{10}$ ) of 0.26 mm, a particle diameter corresponding to 50% finer ( $D_{50}$ ) of 0.33 mm, a coefficient of uniformity ( $C_u$ ) of 1.35, and a coefficient of curvature ( $C_c$ ) of 0.9. The sand was classified as poorly graded based on the Unified Soil Classification System.

The bacteria, Sporosarcina pasteurii (ATCC 11859), were cultivated in an ATCC specified medium and were contained in an environmental shaker operated at 170 rpm and 33°C. The harvested bacteria and growth medium were then centrifuged and the supernatant was

decanted. The remaining concentrated bacteria solution was stored at 4°C prior to use. The study considered a polymer-modified MICP process aimed at providing moist microenvironment thereby retarding the excess migration of cementitious media. Polyvinyl alcohol (PVA) was used to prepare the PVA-modified cementation solution by carefully adding the granular PVA powder into urea–CaCl2 deionized water solutions under high-speed stirring at 60°C. The percentage weight of the mixed PVA powder in the solution was approximately 7.5% weight per weight. Samples for different cementation concentration media were prepared using different plastic container sizes for both strength and erodability tests. This was accomplished by pouring slowly the Ottawa sand into containers having cementation and bacteria solutions and stirred with glass rod. Samples were kept for 10 days after which they were removed from containers, immersed in water for three days and air-dried prior to tests. The bench-scale surficial stabilization of Ottawa sand showed a uniform soil crust in the surficial region and reduces the erodability of the sand. The shear stress of the treated sand was 500 times higher than the natural one as demonstrated by the erosion function apparatus test.

Bio-sequestration of heavy metals was investigated using MICP. Heavy metals resistant bacteria were used to bio-mineralized lead, iron, zinc and cadmium. Out of the twenty-two bacterial strains isolated from calcareous soil samples, ten most efficient were selected subsequent to urease-calcite production and tolerance to heavy metals toxicity screening. Urease broth medium containing (g/l): urea, 20; NaHCO<sub>3</sub>, 2.12; NH<sub>4</sub>Cl, 10, nutrient broth, 3; CaCl<sub>2</sub>. 2H<sub>2</sub>O, 25, pH 7 was inoculated with overnight grown seed culture and incubated under shaking condition at 37°C for 72 h. Ammonia released from urea hydrolysis was measured as an indicator for urease activity. The metal toxicity experiments were carried out using nutrient broth media supplemented with different concentrations (0-10 mM) of heavy metal salts (zinc chloride, cadmium sulphate, lead nitrate, and iron sulphate) inoculated with 2% overnight grown culture (6.5 Log CFU/ ml). Each metal concentration was tested in and incubated at 35°C for 48 h. The growth of bacteria was measured to examine the feasibility of cells in the presence of these metals. Results indicated that strains WD-2, WD-5, WD-9, BA-3 and BA-7 out of the ten were the highest urease producers and also revealed significant heavy metals resistance efficiency. The urease activity and calcite production in order of efficacy were WD-9 (6.50 U/ml), WD-5 (4.14 U/ml), BA-3 (3.36 U/ml), BA-7(2.20 U/ml) and WD-2 (1.98 U/ml) and WD-9 (10.0 mg/ml), followed by BA-3 (9.32 mg/ml), WD-5 (8.22 mg/ml), BA-7 (7.40 mg/ml), and WD-2 (5.11 mg/ml) respectively. The study proved the efficacy of MICP in sequestering heavy metals with a removal strength of 97.20 % for Pb<sup>+2</sup> as highest and 60.66% for Cd<sup>+2</sup> as lowest after 48h of incubation (Eman, 2019)

Rajabi *et al.* (2019) treated sand and silt for storm control using MICP in laboratory. Samples subjected to different MICP treatments indicated significant positive relationship between urease activity and the amount of calcite precipitation. Wind velocity test revealed that particles of untreated sand and silt began to move at a velocity of 8 and 10 km/h respectively while all biologically treated samples resisted until the wind speed reached 97km/h. This has resulted in soil erosion of 215 and 354 kg m<sup>-2</sup> h<sup>-1</sup> for sand and silt (control) respectively and only 2.5 kg m<sup>-2</sup> h<sup>-1</sup> for MICP treated samples. The results indicated that the application of MICP on soil surface can be an effective alternative for the wind erosion control especially at high velocity.

Corresponding author's e-mail address: ibrahimcivil73@gmail.com

Nasrin *et al.* (2020), explored the removal efficiency of some selected heavy metals via biomineralization using urease-producing bacteria isolated from Iranian mine calcareous soil. In the study, rhizosphere soil samples were collected from 0-20 cm depth across five different contaminated sites in Zanjan province. Serial dilution method was used in the isolation of bacteria with relative resistance to heavy metals. 01 ml of each dilution was spread on nutrient agar plate suplimented with 2 mMZn, 0.5 mM Pb and 0.5 mM Cd and incubated at  $28 \pm 2^{\circ}$  C for 7 days. The study further realized t 76 strains with relative resistance to heavy metals. Out of these 76 isolates, 12 isolates with high color intensity quantitatively were considered. The native isolates tagged A323 and A122 among the isolates appeared to represent the upper and lower urease activity having a value of 1.65 U/ml and 0.85 U/ml respectively. In addition, *Sporosarcina pasteurii* PTCC 1645 (DSM33) with urease activity of 11.08 U/ml was procured and used for comparison with the native microbes in the biomineralization process.

Furthermore, heavy metal precipitation experiment indicated that *Sporosarcina pasteurii* had highest Pb carbonate of 73g/l followed by bacterial isolate C113 and A323 having 26.73g/l and 26.0g/l respectively. *Sporosarcina pasteurii* also had the highest carbonate precipitation in Zn and Cd while among the isolates M212 had the highest in Zn and A323 in Cd. However, the general biomineralization process showed that *Sporosarcina pasteurii* removed 98.71% of Pb, 97.15% of Cd, and 94.83% of Zn. A323 removed 96.25%, 71.3%, and 63.91% of Pb, Cd, and Zn, respectively, and C113 removed 95.93% of Pb, 73.45% of Cd, and 73.81% of Zn. The final results proved that the bacteria are capable of converting soluble metals into insoluble carbonate thereby immobilizing and encapsulating them.

Based on this review, it is suggested that *Sporosarcina pasteurii* is the best candidate for metal fixation, stabilization and remediation in soils following its high efficacy of carbonate precipitation, encapsulation and solidification.

#### 3.0 Conclusion

The application of Microbial Induced Calcite Precipitation technique in soil improvement have increased over the years and has been proven applicable in different fields. A wide variety of microorganisms can be used in the production of urease for bio-calcification. However, microbes such as *Sporosarcina pasteurii* and *B. megaterium* have the greatest potential. The technique is eco-friendly and can be used in areas such as soil remediation, liquefaction resistance of soils, concrete cracks healing, sealing of leakage in hydrocarbon wells and  $CO_2$  sequestration. However, according to researchers like De Jong et al., 2014 and Gomez et al., 2015, despite this virtue the technique requires further investigation to overcome its limitations prior to its commercialization. Some of these limitations include slow microbial process, production of ammonia which can be toxic and risky to public health as well as economic implication in the acquisition of nutrients for field application.

#### References

Amin, M., Zomorodian, SMA. and Okelly, BC. 2017. Reducing the hydraulic erosion of sand using microbialinduced carbonate precipitation. Proceedings of the Institution of Civil Engineers: Ground Improvement 170(2): 112-122.

Atticus, D., Liang C., Mohamed, A. and Shahinand, RC. 2012. Calcium Carbonate Induced Precipitation for Soil Improvement by Urea Hydrolysing Bacteria. Advances in Civil, Environmental, and Materials Research (ACEM' 12) Seoul, Korea, August 26-30

Bosak, T. 2011. Calcite precipitation, microbially induced. In: Reitner, J. and Thiel, V. (eds) Encyclopedia of earth sciences series. Springer, Netherlands, pp 223–227

Cheng, L., Shahin, MA. and Cord-Ruwisch, R. 2014. Soil Stabilization by Microbial-Induced Calcite Precipitation (MICP): Investigation into Some Physical and Environmental Aspects. 7<sup>th</sup> Congress on Environmental Geotechnics, 10 -14 November, Melbourne, Australia, pp 1105 - 1112.

Corinne, LQ., Robbie, MA., Pierre, F., Stephen, S., Julia, P., Andrew, CM. and Jan, IK. 2018. Global Carbon Budget 2017. Earth System Science Data, 10: 405–448

Dan, B., Diana, P. and Loredana, J. 2009. Environmental Impact of Concrete. Universitatea Technica, Gheorghe Asachi, din lasiTomul LV (LIX), fasc. 4

De Jong, JT., Martinez, BC., Ginn, TR., Hunt, C., Major, D. and Tanyu, B. 2014. Development of a Scaled Repeated Five-Spot Treatment Model for Examining Microbial Induced Calcite Precipitation Feasibility in Field Applications. Geotechnical Testing Journal 37(3): 424-435.

De Jong, JT., Fritzges, MB. and Nusslein, K. 2006. Microbial induced cementation to control sand response to undrained shear. Journal of Geotechnical and Geoenvironmental Engineering, 132: 1381–1392.

De Muynck, W., De Belie, N. and Verstraete, W. 2010. Microbial carbonate precipitation in construction materials: a review. Ecological Engineering, 36:118–136.

Dhami, NK., Reddy, MS. and Mukherjee, A. 2014. Synergistic role of bacterial urease and carbonic anhydrase in carbonate mineralization. Applied Biochemistry and Biotechnology, 172: 2552–2561.

Dupraz, S., Menez, B., Gouze, P., Leprovost, R., Benezeth, P., Pokrovsky, OS. and Guyot, F. 2009. Experimental approach of  $CO_2$  biomineralization in deep saline aquifers. Chemical Geology, 265(1-2):54-62.

Eman, ZG. 2019. Biosequestration of heavy metals by microbially induced calcite precipitation of ureolytic bacteria. Romanian Biotechnological Letters, 24(1): 147-153.

Fatima, H. and Benoit, C. 2018. Major Application of MICP Sand Treatment at Multi-scale Level: A Review. Geo Edmonton, Polytechnique Montreal, Montreal, Quebec, Canada

Firas, J. and Jun-Jie, Z. 2016. Improving Poorly Graded Fine Sand with Microbial Induced Calcite Precipitation. British Journal of Applied Science and Technology 17(2): 1-9.

Gomez, MG., Martinez, BC., Dejong, JT., Hunt, CE., Devlaming, LA., Major, DW. and Dworatzek, SM. 2015. Field-scale bio-cementation tests to improve sands. Proceedings of the Institution of Civil Engineers: Ground Improvement, 168(3): 206-216.

Hansen, PJ. 2002. Effect of high pH on the growth and survival of marine phytoplankton: Implications for species succession. Aquatic Microbiology and Ecology, 28: 279–288.

Harkes, MP., Van-Paassen, LA., Booster, JL., Whiffin, VS. and Va- Loosdrecht, MCM. 2010. Fixation and distribution of bacterial activity in sand to induce carbonate precipitation for ground reinforcement. Ecological Engineering, 36:112–117.

Ilhan, C., Im, J. and Cho, GC. 2016. Introduction of microbial biopolymers in soil treatment for future environmentally-friendly and sustainable geotechnical engineering. Sustainability, 8(3): 251-257.

Malcolm, BB., Thomas, JW., Barbara, CW. and Ronald, LC. 2012. Urease activity of ureolytic bacteria isolated from six soils in which calcite was precipitated by indigenous bacteria. Geomicrobiology Journal, 29: 389–395.

Maclaren, DC. and White, MA. 2003. Cement: Its Chemistry and Properties. Journal of Chemical Education, 80(6): 623.

Mizanur, R. and Reena, NH. 2017. Unconfined compressive strength of microbial induced calcite precipitation (MICP) treated soils. Proceedings of the 19th International Conference on Soil Mechanics and Geotechnical Engineering, Seoul, pp. 489-492.

Mitchell, JK. and Santamarina, JC. 2005. Biological considerations in Geotechnical Engineering. Journal of Geotechnical and Geoenvironmental Engineering, 131(10): 1222–1233.

Mitchell, AC. and Ferris, FG. 2005. The coprecipitation of Sr into calcite precipitates induced by bacterial ureolysis in artificial groundwater: temperature and kinetics dependence. Geochim Gosmochim Acta, 69:4199–4210.

Murtala, U. and Khairul-Anuar, K. 2017. Effect of Cementation Reagents Concentrations on Microbial Calcite Precipitation in Residual Soil. Malaysian Journal of Civil Engineering, 29 (Special Issue 1):79-90.

Nasrin, J., Abdolreza, A., Hossein, AA., Hadi, AR. and Farhad, R. 2020. Removal of heavy metals zinc, lead, and cadmium by biomineralization of Urease-producing bacteria isolated from Iranian mine calcareous soils. Journal of Soil Science and Plant Nutrition, 20: 206–219

Ng, SW., Lee, ML. and Hii, SL. 2012. An overview of the factors affecting microbial-induced calcite precipitation and its potential application in soil improvement. World Academy of Science, Engineering and Technology, 62:723–729.

Okwadha, GDO., and Li, J. 2010. Optimum conditions for microbial carbonate precipitation. Chemosphere, 81:1143–1148.

Periasamy, A., Chang-Ho, K., Yu-Jin, S. and Jae-Seong, S. 2016. Formations of calcium carbonate minerals by bacteria and its multiple applications. SpringerPlus, 5: 250.

Phillips, AJ., Gerlach, R., Lauchnor, E., Mitchell, AC., Cunningham, AB. and Spangler, L. 2013. Engineered applications of ureolitic biomineralization: a review. Biofouling, 29: 715–733.

Rajabi, AS., Kiani, F., Khavazi, K., Rouhipour, H. and Khormali, F. 2019. An environmentally friendly soil improvement technology for sand and dust storms control. Environmental Health Engineering and Management Journal, 6(1): 63-71.

Rao, A., Jha, K.N. and Misra, S. 2007. Use of aggregates from recycled construction and demolition waste in concrete. Resource Conservation and Recycling, 50: 71–81.

Corresponding author's e-mail address: ibrahimcivil73@gmail.com

Saranya, K., Sangeetha, H. and Swetha, PK. 2017. Review on microbial induced calcite precipitation. International Journal of Advanced Research in Civil, Structural, Environmental and Infrastructure Engineering and Developing, 3 (1): 364-367.

Sherwood, PT. 1993. Soil Stabilization with Cement and Lime. HMSO: London, UK.

Taylor, HFW. 1997. Cement Chemistry. Thomas Telford Publishing: London, UK.

Venkatramaiah, C. 2006. Geotechnical Engineering. New Age International Publishers. Third edition, New Delhi, pp 697.

Weil, MH., De Jong, JT., Martinez, BC. and Mortesen, BM. 2012. Seismic and resistivity measurement for Real-Time Monitoring of microbially induced calcite precipitation in sand. Geotechnical Testing Journal, 35(2): 330-341.

Xiangrong, W., Junliang,-Tao, AMASCE., Ruotian, BSMASCE., Tri-Tran, SMASCE.

and Stacey, TKMASCE. 2018. Surficial soil stabilization against water-induced erosion using polymer-modified microbially induced carbonate precipitation. Journal of Materials in Civil Engineering, 30(10): 04018267.

Yefei, T., Xinghua, X., Shiqiang, W. and Tao, W. 2017. Microbially induced  $CaCO_3$  precipitation: hydraulic response and micro-scale mechanism in porous media. Science Asia, 43:1-7.

Zhaoyu, W., Nan, Z., Jinhua, D., Chen, L. and Yong, J. 2018. Experimental study on wind erosion resistance and strength of sands treated with microbial-induced calcium carbonate precipitation. Advances in Materials Science and Engineering, 2018: 1-11.

Zulfequar, AK. 2017. Causes and consequences of greenhouse effect and its catastrophic problems for earth. International Journal of Sustainability Management and Information Technologies, 3(4): 34-39