

INFLUENCE OF INJECTION PRESSURE ON THE HYDRAULIC CONDUCTIVITY OF COMPACTED BIO-CEMENTED LATERITIC SOIL

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ABSTRACT

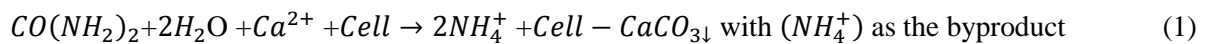
*Despite the use of several soil improvement methods over the years, the sustainability aspect of these methods have recently been queried. The constant search by researchers in the related field for a sustainable method led to the discovery of a method known as microbial induced calcite precipitation. The aim of this study is to evaluate the influence of injection pressure on the hydraulic conductivity of compacted bio-cemented lateritic soil for use in waste containment applications. The soil was treated at the optimum moisture content of British Standard light compactive effort with 1/3rd the pore volume as the bacterial suspension density of 1.80×10^9 cells/ml and cured for a minimum of 12 hours before compaction. The compacted soil samples were then saturated with a cementitious mixture with 20 g of Urea, 10 g of NH_4Cl , 3 g of Nutrient broth, 2.8 g of CaCl_2 and 2.12 g of NaHCO_3 per litre of de-ionized water to aid the microbial induced calcite precipitation process in three circles with 2/3rd pore volume each under varied pressure between 0 to 1 bar and cured for 24 hours. The cured specimens were saturated in water for another 24 hours before being subjected to hydraulic conductivity test. Results obtained showed an initial reduction in hydraulic conductivity from 3.3×10^{-8} m/s under gravity to 3.46×10^{-9} m/s at injection pressure of 0.5 bar and thereafter steadily increased to 8.3×10^{-9} m/s at a peak pressure of 1.0 bar. The least hydraulic conductivity value of 3.46×10^{-9} m/s was recorded at 0.5 bar injection pressure, which was regarded as the optimal injection pressure for the treatment with 1.80×10^9 /ml *S. pasteurii* suspension density/ml. This result did not meet the maximum regulatory value of 1.0×10^{-9} m/s for use in waste containment facilities. It was recommended that similar studies using nano biotechnology be evaluated at higher compactive effort as further studies.*

Keywords: Hydraulic conductivity, *Sporosarcina pasteurii*, Bio-cemented, Lateritic soil, Injection pressure, Pore volume.

1 INTRODUCTION

Soil is a natural material that display different engineering properties due to the various geologic processes it has undergone during its formation, while some have deficiencies in their properties for engineering use, others meet specification for use as engineering materials, for over a century now, engineers have established different procedures for the improvement of soils for engineering use, most of this methods apart from the use of sodium silicate are associated with environmental related problems which has become a source of concern for engineers, one method of soil improvement technique that have received an increased attention of researchers in the related field is the microbially induced calcite precipitation (MICP), it is emerging, novel and innovative which has also been adjudged to be a possible alternative to the conventional

soil improvement methods [1]. MICP is a marriage of research ideas between the microbiologist and the civil engineer, it has been reported to be a promising method of soil improvement that exploits the metabolic paths of bacteria to form calcite precipitation in the soil matrix, resulting in an increase in soil strength, stiffness and decreased hydraulic conductivity [2]. Therefore, the discovery of MICP processes of improving the engineering properties of soil together results from the following studies in the last decade should not be seen to have only disproved the long term misconception of the conventional geotechnical engineering that that activities within the soil community is strictly abiotic, i.e. free from microbial activities [3, 4, 5, 6, 7, 8]. In MICP, calcite (CaCO_3) is the needed by product which is responsible for enhancement of soil properties. The following are some of the mechanisms used in MICP: sulfate reduction, urea hydrolysis, Denitrification (nitrate reduction), photosynthesis, ammonification or methane oxidation [9, 10, 11, 12, 13, 14]. Though there are several biotic procedures that results to MICP, the urea hydrolysis has been the most preferred method in the current subject of study in recent years, because it is direct and can yield up to 90% biochemical efficiency of calcite precipitate in less than 24 hours [3, 15, 16, 17, 18]. This study focuses on MICP via urea hydrolysis pathway. MICP occurs whenever a microbial metabolism results in altering the immediate aqueous phase resulting in the formation of calcite [19]. MICP is currently an evolving area of study has and still receives the attention of researchers as a viable method of soil improvement [18, 20, 21]. However, it is more effective in granular/porous soils. In MICP processes, microorganism hydrolysis urea into ammonia and carbonates, and in the presence of excess calcium source, calcium carbonate is formed as a precipitate as shown in equation 1.



During MICP process, there are two major reactions that may occur individually or concurrently: (1) Bio-cementation in which ions of carbonate are produced, then in the existence of extra dissolved calcium ions, calcite crystals are precipitated. (2) Bio-clogging process on the other hand is targeted at decreasing the hydraulic conductivity of soils and porous rocks entirely due to microbial activity, bio-clogging can then be used to seal a leaking construction pit, landfill etc. [22] reported that the hydraulic conductivity of MICP treated soil specimens was meaningfully decreased when the strength of treated specimens surpasses a certain value, this suggest that bio-cementation and bio-clogging could occur simultaneously. The hydraulic conductivity is one of the vital factors that is used in the evaluation of engineering facilities for dams and waste containment applications in chemical transport in the subsurface [23]. The term laterite is derived from the Latin word- later, denoting brick. The term 'Laterite' appeared first in academic publications more than a century ago, it was first used by [24]. to mean a building material in the mountainous area of Malabar, India [25]. [26] tried to re-define laterite, by suggesting a new description and classification based on rock chemistry; fundamentally on the silica- sesquioxide ratio $\{\text{SiO}_2/(\text{Al}_2\text{O}_3 + \text{Fe}_2\text{O}_3)\}$ in comparison to the chemical composition of the underlying parent rock material. According to [27], a silica-sesquioxide ratio value {i.e., $\text{SiO}_2/(\text{Al}_2\text{O}_3 + \text{Fe}_2\text{O}_3)$ } in the range of 1.33 and 2.0 is lateritic, while value higher than 2.0 is indicative of non-lateritic, typically weathered soils. This soil has featured in a lot of engineering projects, especially in the tropics. Several studies have reported a reduction in hydraulic conductivity of soils using MICP treatments. [28] established a reduction in hydraulic conductivity values from 1×10^{-7} m/s for natural soil to 2.6×10^{-8} m/s for the treated soil. Also [29] reported a decrease in the values of hydraulic conductivity to be as much as 4 orders of magnitude which was dependent on the effective size of the soil specimen (D_{10}), average calcite precipitation content and the initial hydraulic conductivity. Studies by [30] showed a reduction from 5.04×10^{-5} m/s for untreated sand to 1.41×10^{-5} m/s after four cycles of MICP treatment with cementation reagent. Among the aforementioned processes that leads to calcite formation, from studies, it has demonstrated that ureolysis is the most established method being used. [31] have established a reduction in hydraulic conductivity values of lateritic soil by 83%, while [32] have reported a reduction of 99.84% in sand. This study was aimed at evaluating the effect of injection pressure on the hydraulic conductivity of compacted bio-cemented lateritic soil at *S. pasteurii* suspension density 1.80×10^8 cells/ml treated at the optimum moisture content, for use as a liner material in waste containment application.

2 MATERIALS AND METHODS

2.1 Materials

Soil: Lateritic soil with pore throat that allowed the free mobility of the microorganisms when in compacted form, was used in the study. It was obtained from Abagana (Latitude 6° 10' 15" N and Longitude 6° 58' 10" E), in Njikoka Local Government Area, Anambra State, Nigeria as shown in Figure 1 below. The method of disturbed sampling was adopted during the collection of the soil samples at depths between 0.3 m and 3.0m.



Figure 1 Map of Abagana

Bacteria used: The species of bacteria used in this study is *S. pasteurii*, it is a rod-shaped, Gram-positive, urease positive, spore and biofilm forming bacteria, which was cultured from the lateritic soil sample in the Department of Microbiology, Modibbo Adama University, Adamawa State.

2.2 Methods

Bacteria culture/growth medium: The method used in the identification of the bacteria the biochemical method (conventional procedure) as defined in [33]. Bacteria was cultivated in an Ammonium-Yeast Extract media described in [34, 35].

Bacteria cell density: The bacteria suspension density that was used in this study is 1.80×10^9 cells/ml. The pore volume, which is the numerical differences between the degree of saturation when the compacted soil sample is completely saturated and the degree of saturation of compacted soil specimen at optimum moisture content, was computed to determine maximum volume of bacteria suspension and cementation reagent to be used in the study. The use of 2/3 pore volume for cementation reagent with 1/3 pore volume for micro-organism is to ensure that after treatment, the soil does not exceed the needed pore volume for saturation. This was done using the standard Proctor or the British Standard Light compactive effort after repeated trials [36]. The maximum volume of micro-organisms mixed with the soil was 1/3rd of the pore volume as recommended by [37].

2.2.1 Characterization of calcite precipitates through test tube experiments

3ml of *S. pasteurii* for suspension densities of 0, 1.50×10^8 , 6.0×10^8 , 1.20×10^9 , 1.80×10^9 and 2.40×10^9 cells/ml, were mixed with 7ml of the cementitious reagent in three cycles at six hours interval, this was allowed to remain in the laboratory for 24 hours at a temperature range of $24 \pm 2^\circ\text{C}$,

the precipitate resulting from their reactions was filtered using a filter paper, the solids retained on the filter paper was oven dried at a temperature of 105°C and was kept for characterization using XRD test.

2.2.2 X-Ray diffraction analysis and Oxide composition

The X-Ray diffraction analysis and Oxide composition of the Lateritic soil was carried out at the National Geosciences Research Laboratory, Kaduna.

Index properties: The determination of all index properties of the soil were in accordance with procedures as outlined in [38].

Compaction characteristics: The pulverized and air-dried soil sample passing through BS No.10 sieve (2.00 mm aperture) was mixed at the optimum moisture content, with 1/3rd pore volume being the microorganisms for all suspension densities while the remaining being tap water. This was mixed thoroughly with 3000g of the soil sample and cured in polyethene bag at a temperature of 24 ±2°C for at least 12 hours, for the bacteria to be well disseminated within and around the soil before compaction.

Injection method: Cementitious mixture containing 20g of Urea, 10g of NH₄Cl, 3g of Nutrient broth, 2.8g of CaCl₂ and 2.12 g of NaHCO₃ per litre of de-ionized water was injected into the compacted soil at varied pressures ranging between 0 and 1 bar, with the aid of a compressor and the treatment set up until it is saturated as shown in Figure 2. This is the first injection cycle, the second cycle was repeated after six hours, while the third cycle was repeated after twelve hours from the second cycle, this mixture was allowed to cure for 24 hours after the last cycle before a saturation for a period of another 24 hours before commencing the hydraulic conductivity test.

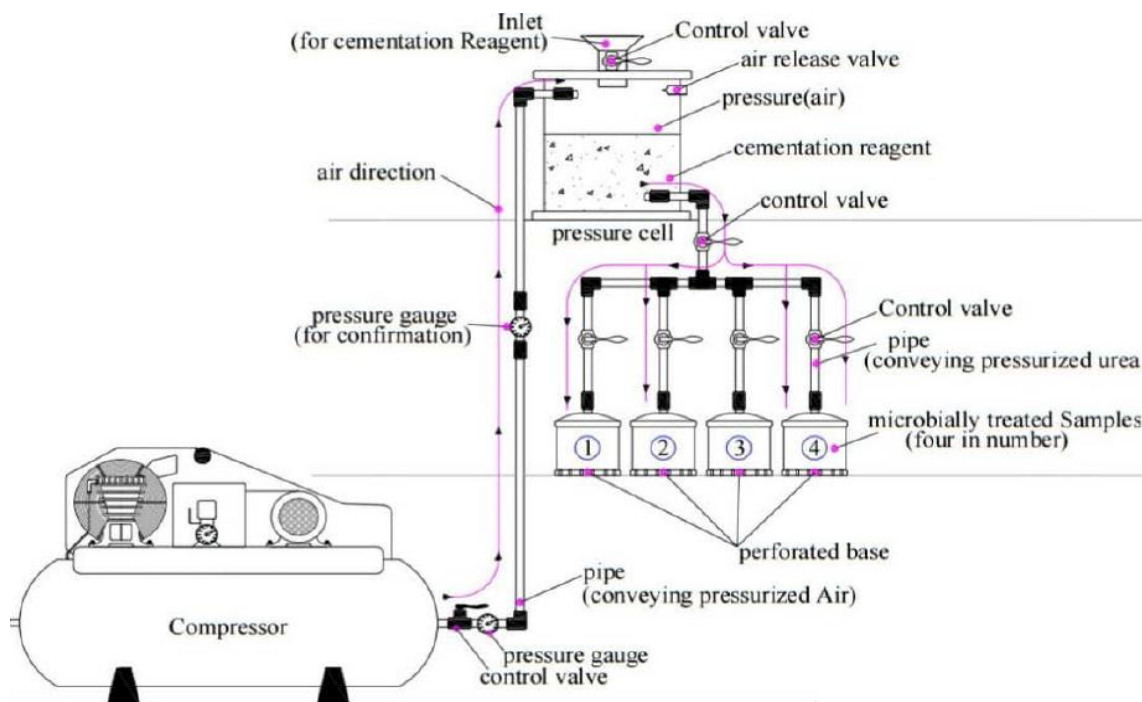


Figure 2. Schematic diagram of injection process

Hydraulic conductivity test: The saturated treated specimens were connected to the rigid wall permeability cells using the falling head condition as suggested by [39] and also in accordance with the procedures outlined in [38, 40].

2.3 Microanalysis

The Scanning Electron Microscope (SEM) for specimens used in this study was carried out in accordance to procedures given in [36].

Bio-film formation

Generally, genotypic and phenotypic methods (i.e., Congo Red Agar, CRA and Microtiter Plate, MTP, respectively) are used to confirm the capability of an organism to form biofilm. [41]. In accordance to procedure outlined in [42], the CRA medium was made using mixtures of 37g/l BHI broth, 50g/l sucrose, 10g/l agar, and 0.8g/l Congo red. Congo red stain was prepared as a concentrated aqueous solution and then autoclaved at 121°C for 15min alone from other medium components, and was added after the temperature of the agar had dropped to about 55°C. The plates were inoculated with the organism and kept in an incubator at a temperature of 37°C for 24 hours. After 24 to 48 hours, the plates were checked for any possible change in the colour of the colonies. An organism that can produce biofilm will indicate black colonies, while non-producing organism developed red colonies (i.e., no change in colour of the CRA).

3 RESULTS AND DISCUSSION

Index properties: Table 1 presents the summarized results of the index properties of the natural soil, the particle passing BS sieve No. 200 is 36.0%, the liquid and plastic limits were found to be 44.0 and 21.6%, respectively; while the plasticity index was 22.5%. Based on the results, the soil is classified as A-4(3) using the [43] classification system and SC using the ASTM Standard [44]. The optimum moisture content and the maximum dry density are 15.3% and 1.83 Mg/m³ respectively. According to Table 2, the soil recorded silica-sesquioxide ratio {SiO₂/(Al₂O₃ + Fe₂O₃)} of 1.64. According to [27], a soil is said to be lateritic, if it has a silica-sesquioxide ratio value between 1.33 and 2.0, while any value higher than 2.0 is indicative of non-lateritic soil. Microstructural analysis using X-ray Diffraction on the precipitates derived through the interaction of *S. pasteurii* and the cementitious reagent confirmed calcite as the dominant composition of the precipitate with few impurities of Quartz and Actinolite which was reported by [45] to be among the most cations frequently associated with biogenic minerals (Figure 3). The XRD spectra presented in this study agrees with that reported by [46].

Table 1. Physical properties of the natural soil

Property	
Natural moisture content (%)	11.3
Percentage Passing No. 200 sieve (wet sieve)	36.0
Liquid Limit (%)	44
Plastic Limit (%)	21.55
Plasticity Index (%)	22.45
CEC(meq/100 g)	5.50
Specific Gravity	2.62
AASHTO classification	A – 4(3)
USCS	SC
Colour	Reddish brown
Dominant Clay Mineral	Kaolinite

Table 2. Oxide composition of the natural soil

Oxide	SiO ₂	Al ₂ O ₃	CaO	TiO ₂	V ₂ O ₅	Cr ₂ O ₃	Fe ₂ O ₃	MnO	CuO	ZrO ₂	LOI	Total
Concentration (%)	56.5	19.00	0.33	2.89	0.061	0.051	15.41	0.075	0.056	0.290	4.54	99.20

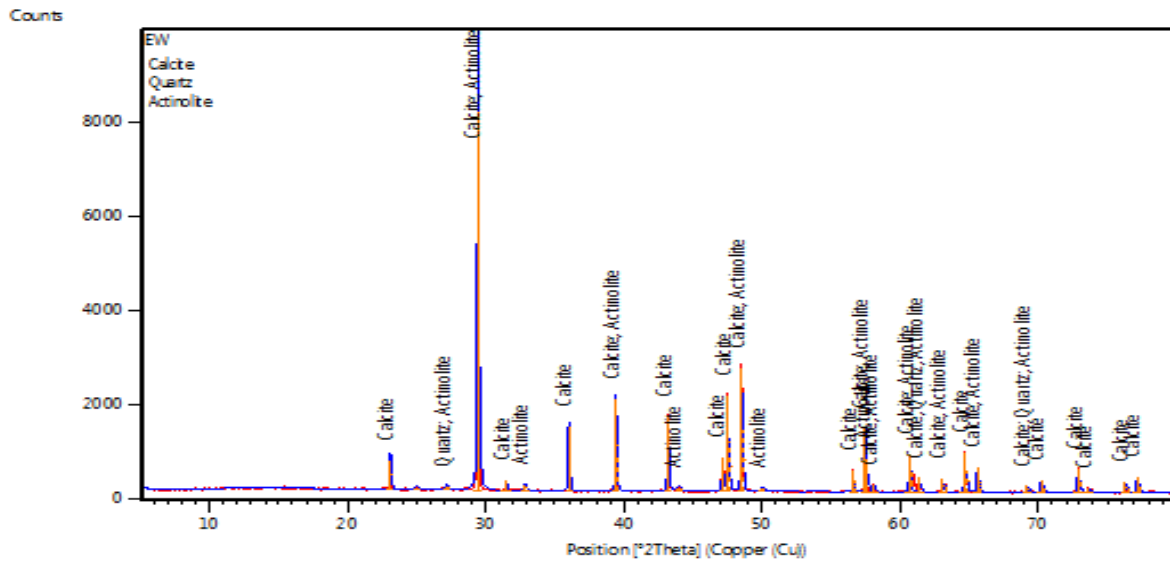


Figure 3. X-ray Diffraction of precipitate obtained test tube experiment

Figure 3, presents the XRD results of the precipitate resulting from the reactions between *Sporosarcina pasteurii* and the cementation reagent, it could be observed that the dominant composition of the precipitate is calcite with few impurities of Quartz and Actinolite which was reported by [45] to be among the most cations frequently associated with biogenic minerals. Therefore, the reaction that produced this precipitate is expected to produce calcite under normal condition, when the Lateritic soil containing the microorganism is treated with the cementation reagent, which will result in enhancing the strength characteristics of the soil as reported in some studies e.g. [18, 20, 21, 47].

Particle size distribution: Figure 4 shows the results of the particle size distribution curve for the natural soil. It was observed that the sand content was 64.6%, silt content was 13.4% while the clay content was 22% respectively.

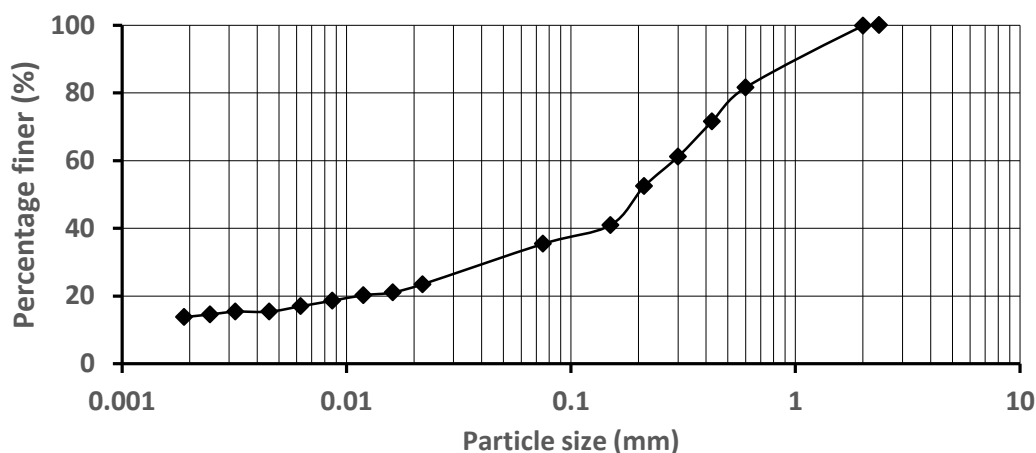


Figure 4. Particle size distribution curve of the natural soil

Figure 5 shows the changes in void ratio and porosity with injection pressure, it was observed that there were reductions in both the void ratio and porosity with increasing injection pressure with the lowest values of 0.445 and 0.308 recorded at injection pressure of 0.5 bar. However, at higher pressures, the values of void ratio and porosity increased to 0.459 and 0.315 recorded at 1.0 bar. For a long time now, there exist a long-standing difficulty for geotechnical researchers and engineers to correctly and reliably predict the hydraulic conductivity of soils, because studies have reported that

laboratory measurement of hydraulic conductivity depends on several parameters: void ratio, porosity, degree of saturation, clay content, pore geometry, pore throat etc. [48]. [49] have reported a linear relation existing between hydraulic conductivity and the void ratio of clay. It should be recognized that the porosity governs the efficiency of MICP treated specimens by means of regulating the replacement of the pore content of soil grains by calcite. It is therefore inferred that the lowest hydraulic conductivity values recorded at 0.5 bar injection pressure could probably be due to the formation of calcite which may have filled the pore spaces through bio-clogging resulting into lower values of void ratio, porosity and hydraulic conductivity.

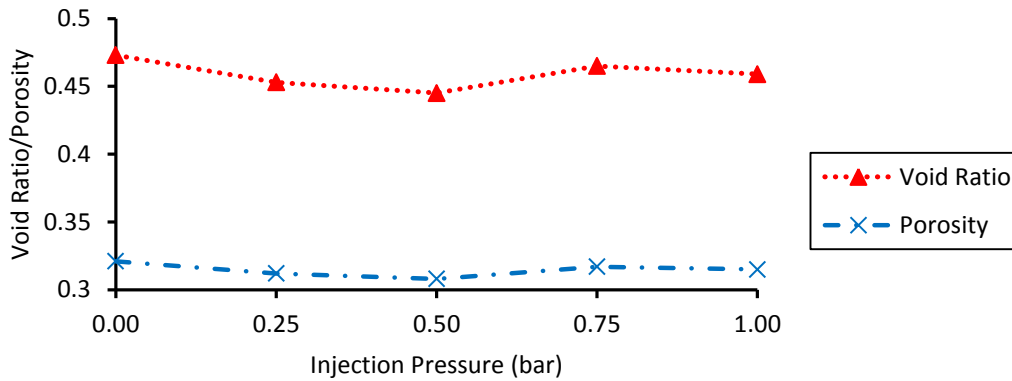


Figure 5. Variation of void ratio and porosity with injection pressure

Figure 6 shows the variation of bulk density and maximum dry density of the treated soil with varying injection pressures. It was observed that the bulk and dry densities increased with increase in injection pressure from 2.05 and 1.78 Mg/m³ under gravity to maximum values of 2.11 and 1.81 Mg/m³ at 0.5 bar injection pressures, respectively. However, bulk and dry densities decreased to lowest values of 2.09 and 1.79 Mg/m³ at 0.75 bar, respectively. It was also observed from Figure 5 that, as the injection pressure increases, both the bulk and dry densities increased to maximum values of 2.11 and 1.81 Mg/m³ at injection pressure of 0.5 bars before decreasing. The injection pressure at which these densities were recorded corresponds to that which recorded the lowest hydraulic conductivity, it was therefore illustrated that hydraulic conductivity of the treated soil is inversely connected to its densities. The observed trend of results could be due to the calcite precipitate may have resulted into plugging the pores within the soil through bio clogging.

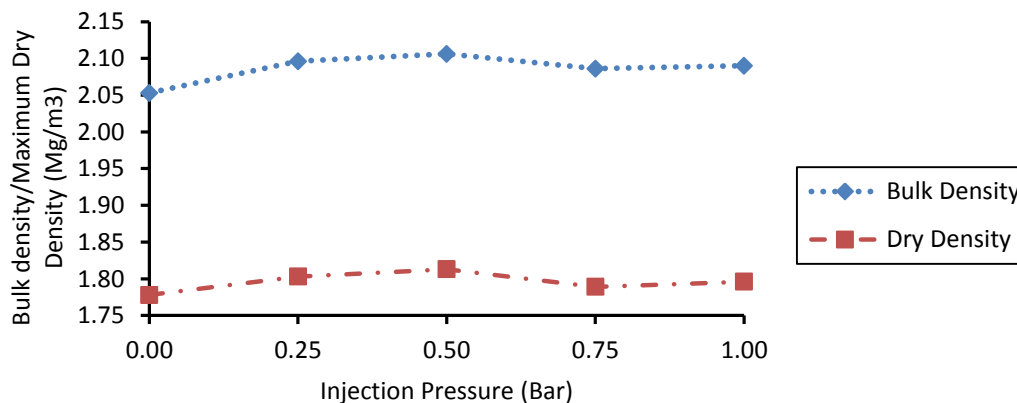


Figure 6. Variation of bulk and maximum dry density with injection pressure

The variation of degree of saturation with injection pressure is presented in Figure 7. It was observed that there was a drastic increase in the degree of saturation from 85.53% under gravity to 94.07 and 95.05% at 0.25 and 0.5 bar injection pressures. However, with higher pressures the degree of saturation decreased to 93.59 and 93.55% at 0.75 and 1.0 bar injection pressures, respectively. The observed decrease in degree of saturation with higher injection pressures could be attributed to possible calcite crystals deposits that may have been formed which eventually filled the pore voids,

resulting in a decreased hydraulic conductivity at higher degree of saturation. In addition, the observed decrease in degree of saturation could also have been due to bio clogging by the presence of calcite crystallization in the soil pore spaces at higher degree of saturation. However, the observed trends are consistent with the results reported by other researchers, [17].

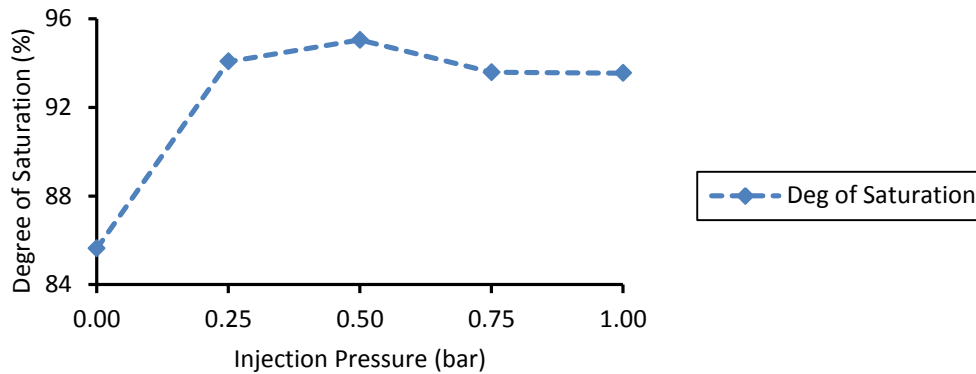


Figure 7. Variation of degree of saturation with injection pressure

Hydraulic conductivity: The change of hydraulic conductivity values with injection pressure is presented in Figure 8. The injection pressure was varied as follows: 0, 0.25, 0.50, 0.75 and 1.0 bar pressures. It was observed that as the injection pressure was increased from gravity to 0.50 bar, there was a steady decrease in the hydraulic conductivity of the treated soil by an order of magnitude, from 3.3×10^{-8} m/s under gravity to 3.46×10^{-9} m/s at 0.50 bar. Further increase in the injection pressure from 0.75 to 1.0 bar, resulted in an increased values of the hydraulic conductivity of the treated soil specimen from 3.46×10^{-9} m/s at 0.50 bar to 8.3×10^{-9} m/s at 1.0 bar. The increase in hydraulic conductivity with higher injection pressures may be attributed to of the pores created by the dispersion created in the compacted soil particle by the injection pressure or possibly micro cracks may have developed within the soil, thereby creating a preferential flow path through which some of the microorganisms may have been flushed out, making their participation in the MICP process difficult. The following are the reduction in hydraulic conductivity when compared to the untreated soil: 86.50%; 89.52%; 85.36 and 74.85% for 0.25, 0.50, 0.75 and 1.0 bar injection pressures respectively. From the results presented, the lowest hydraulic conductivity value of 3.46×10^{-9} m/s was recorded at an injection pressure of 0.5 bar which is considered as the optimum injection pressure. Also during the injection process, it was observed that the time taken by the cementation reagent to saturate the compacted soil decreased with increasing injection pressure as well as injection cycles, this led to reduction in the efficiency of flow for the cementation reagent. The decrease in time taken for saturation maybe due to fine particles of calcite precipitate that may have occupied the pore spaces within the soil, which have resulted in decreasing the hydraulic conductivity values through bio-clogging. Although the lowest hydraulic conductivity of 3.46×10^{-9} m/s recorded in this study was lower than those reported in the literature. This did not meet the maximum recommended hydraulic conductivity value of 1.0×10^{-9} m/s for use in waste containment applications. This reported result agrees with those reported by [32, 50], which reported that bio-clogging is an effective and useful way of decreasing the hydraulic conductivity in soil.

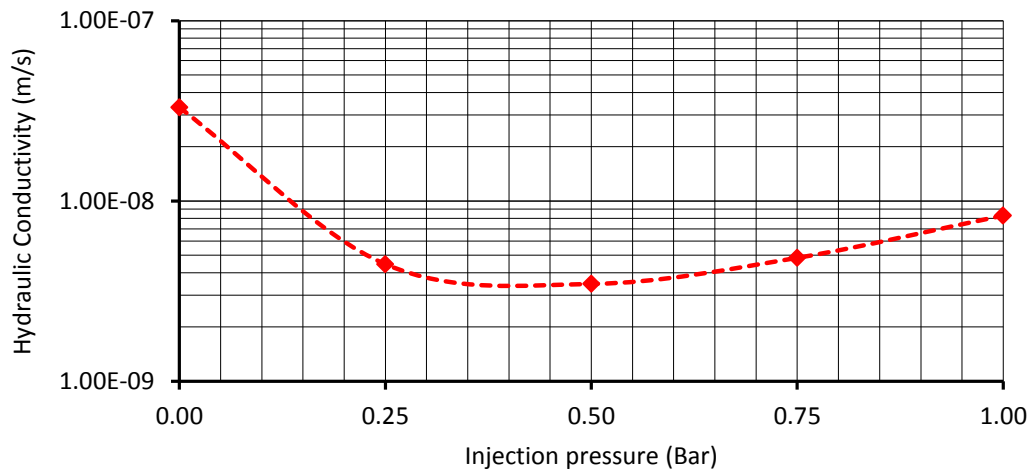


Figure 8. Variation of hydraulic conductivity with injection pressure

Microanalysis

The micrographs of the SEM test carried out on the natural and as well as the treated soils are shown in plates 1A and 1B, respectively. An evaluation of the micrographs of the natural soil it could be observed that there are pores/micro cracks existing as indicated by the arrows in plate 1a which were observed to have been sealed in the treated soil as a result of the calcites formed which may have resulted in the preferential flow paths thereby resulting in decreased hydraulic conductivity in the treated soil through the process called bio-clogging see Plate 1b. The qualitative judgement from the morphology of the micrographs on plates 1a and b are in agreement with the results of study by [51].

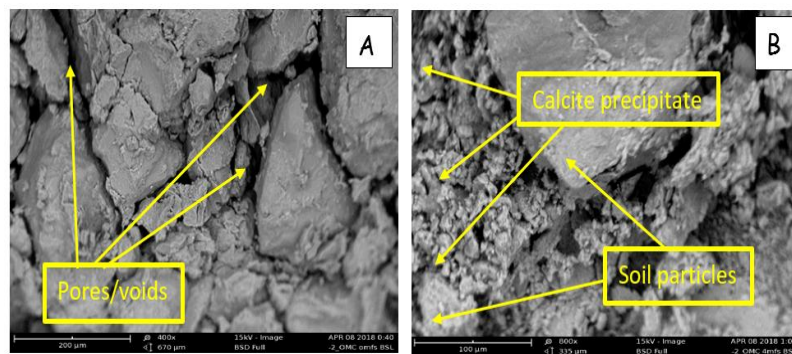


Plate 1. Micrographs of lateritic soil at x800 magnification for; (A) natural lateritic soil (B) Lateritic soil treated with 1.80×10^9 *S. pasteurii* suspension density/ml

Phenotypic formation of bio-film

Interaction between some species of micro-organism with soil and other microbial species could result in the formation of bio-mass and bio-film that reduce hydraulic conductivity in soils. Plate 2 shows the phenotypic expression test on the organism to form bio-film. The black metallic coloration indicates the ability of the organism to form bio-film. Plate 2A shows the reaction from the pure culture of the organism, while 2B shows additional phenotypic expression by other microbial strains apart from bio-film formation, similar results on bio-film forming organisms were reported by [52]. In addition to calcite precipitate that may have occurred during the MICP processes, the formation of bio-film due to the presence of this organism with other species may have played a part in the decreased hydraulic conductivity values recorded in the study.

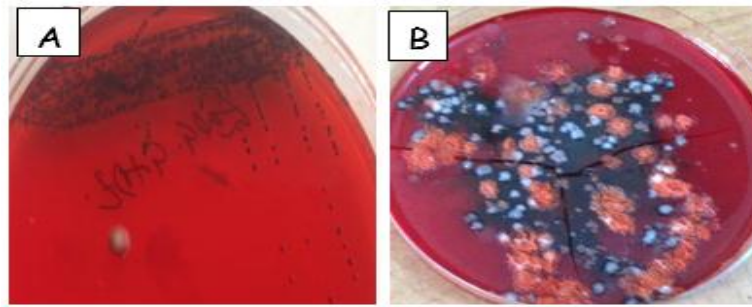


Plate 2. Phenotypic bio-film expression: A. From pure bacterial culture, B. From soil

4 CONCLUSIONS

From the studies and results presented the following conclusions can be made: This study has demonstrated the feasibility of bio clogging through calcite precipitate in addition to biofilm resulting in decreasing the hydraulic conductivity values of the treated soil, this has also provided evidence of the linkage between microbial activities and hydraulic conductivity decrease. At higher degree of saturation calcite crystals were formed at arbitrary locations within the soil, which led to the occupation of calcite crystallization in the soil pore space, resulting in a decreased hydraulic through bio clogging. The optimum injection pressure for the study is 0.5 bar since it recorded the lowest hydraulic conductivity values of 3.46×10^{-9} m/s among the injection pressure variables. This did not meet the maximum regulatory recommended hydraulic conductivity value of 1.0×10^{-9} m/s for use in waste containment applications.

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