Study on the Effect of Biogrouting in Cochin Marine Sand

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Abstract: Cochin Marine Sand (CMS) are highly permeable, loose soil having low shear strength. Traditional methods of soil improvement include mechanical stabilization, chemical stabilization and stabilization through grouting. Cement and chemical grouting are the most commonly adopted methods. These stabilizations may affect the ground water flow and cause release of carbon dioxide pollutants to both water as well as air. Environmental friendly and economic ways of soil stabilization techniques have to be developed alternatively. Biogrouting aims at improving the properties of sandy soil through precipitation of calcium carbonate ($CaCO_3$) between the grains. Biogrout production depends upon Microbial Induced Calcite Precipitation (MICP) where microbes are used as grouting material. Change in compaction, shear strength and permeability characteristics of Cochin marine sand (CMS) with varying concentrations of Bacillus subtilis bacteria (1 x 10^7, 1 x 10^8 and 1 x 10^9 cells/ml), cementation reagent consisting of Urea and Calcium chloride dihydrate (0.5 M, 1 M and 2 M) and treatment duration (10, 20 and 30 days) were studied and reported. Egg white and yolk was used as organic supplement. Calcium carbonate precipitation and XRD analysis was done to confirm the MICP. Bacterial concentration of 1 x 10^9 cells/ml offered best results at 1 M cementation reagent at 30 days of treatment. The results were retarded at higher concentration of cementation reagent. Improvements are observed with increase in treatment days. Thus the results indicate that biogrouting is effective for improving the properties of sandy soil.

Keywords — Bacillus subtilis, Biogrouting, Cementation reagent, Cochin Marine Sand, MICP, Soil improvement.

I. INTRODUCTION

The soil stabilization aims at improving the properties of soil. In regions were the soil properties are insufficient to satisfy the desired functions, soil stabilization is concerned. Stabilization via grouting is the most commonly used technique in construction industry. Of these, chemical grouting and cement grouting are the prevailing methods. But these stabilization methods are causing threats to environment by polluting ground water flow. Biogrouting is an emerging, ecofriendly technique for improving the properties of sandy soils. Microbes are used as grouting material and chemicals including urea ($CO(NH_2)_2$) and calcium chloride dihydrate ($CaCl_2.2H_2O$) are introduced into the soil. Microbial Induced Calcite Precipitation (MICP) is an effective biogrouting approach which is gaining popularity in geotechnical engineering applications. The MICP utilizes urea hydrolysis to yield calcium carbonate precipitation. The hydrolysis of urea into ammonium and carbonate is catalyzed by urease enzyme in ureolytic bacteria. The presence of ammonium rises the pH and $CaCO_3$ precipitates are formed in presence of calcium chloride dihydrate. The studies have reported that formation of calcite precipitate is accelerated in presence of non-ureolytic bacteria.

Cochin areas are undergoing rapid industrialization and economic growth. The soil along coastal regions are classified as marine soil. Most of the Cochin areas are located along the sea coast. The marine soils consist of sandy, silty and clayey deposits. Marine sand is a type of loose and collapsible sand found in coastal regions. Higher water penetration in the marine sand causes reduction in strength. Pores in between the sand particles need to be bridged to reduce the permeability of soil. Thus necessary steps are to be adopted to improve the shear strength and reduce the permeability of soil.

In this study, the change in compaction, shear strength and permeability characteristics of Cochin Marine Sand (CMS) is studied. The non-ureolytic bacteria bacillus subtilis was used for the study. Optimal count of bacteria and cementation reagent was established by varying bacterial concentration and cementation reagent at 10, 20 and 30 days of treatment duration.

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II. Experimental Study

A. METHODOLOGY

Bacteria, urea and calcium chloride need to be injected into the soil to induce urea hydrolysis. When they are mixed prior to injecting into the soil, reaction will start immediately and calcite precipitate would be formed. Thus the desired action of clogging the pores of soil will not happen. In this work, required concentration of diluted bacterial solution and equal molars of cementation reagent were flushed into the soil specimen sequentially. Thorough mixing was done for proper distribution. Thus the reaction happened effectively with homogeneous distribution of microbes and cementation reagent. Bacterial solution was initially supplied with organic supplement of 1% by weight of soil. Egg white and yolk were provided as organic supplement. Three concentrations of bacteria used for the study were 1 x 10^7, 1 x 10^8 and 1 x 10^9 cells/ml. 0.5 M, 1 M and 2 M were the cementation reagent concentrations selected.

B. MATERIAL COLLECTION

Cochin marine sand, Bacillus subtilis, Nutrient, Urea (NH_2CONH_2) and Calcium Chloride Dihydrate ($CaCl_2.2H_2O$) were used in this study.

COCHIN MARINE SAND (CMS)

Marine sand was collected from Vallarpadam at a depth of 1 m. It was greyish in colour.

Fig 1 CMS

TABLE I
MATERIAL PROPERTIES OF CMS

Properties	Values
Specific gravity	2.6
Percentage of clay size particles (%)	0.98
Percentage of silt size particles (%)	8.81
Percentage of sand size particles (%)	90.02
Percentage of gravel size particles (%)	0.19
Soil Classification (Indian Standard Classification System)	Poorly graded sand (SP)
Optimum moisture content (%)	14.76
Maximum dry density (g/cc)	1.65
Angle of internal friction (˚)	29
Cohesion (kN/m^2)	0
CBR Value	4.14 for 5 mm penetration
Coefficient of permeability (cm/s)	2.94 x 10^{-3}

BACILLUS SUBTILIS

The bacteria Bacillus Subtilis strain JC3 was used in this study. Pure culture of Bacillus Subtilis was collected from the department of Agricultural Microbiology, College of Horticulture, Vellanikkara, Thrissur.

CHEMICALS

Urea (NH_2CONH_2) and Calcium Chloride Dihydrate ($CaCl_2.2H_2O$) were used for the work. They are known as cementation reagent. Sufficient amount of cementation reagent is essential for the urea hydrolysis to take place effectively.

NUTRIENT

Egg white and yolk were used as organic supplement after removing fat (1% by weight of soil). It contributes to the major source of nutrient for the growth of microbes.

C. TESTING PROGRAM

The usefulness of B. subtilis in promoting MICP was confirmed using set of test tubes. A series of Standard Proctor test, Direct shear test, Permeability test, CBR test were conducted on treated CMS. XRD analysis were conducted for sample at the optimal count of bacteria and optimum concentration of cementation reagent.

III. Result and Discussion

The effect of biogrouting on compaction, shear strength and permeability characteristics of treated CMC were studied and the results are reported below.

A. CALCIUM CARBONATE DEPOSITION

The calcite precipitation induced by microbes was confirmed using set of test tube experiments. Equi-molar contents of cementation reagent with concentrations of 0.5 M, 1 M and 2 M was used. The amount of bacterial cell concentration used was constant which was supplied with organic nutrient, egg white and yolk. The various concentrations of cementation reagent and bacillus subtilis solution was thoroughly mixed which contained 50 mL of total solution volume. Visual examination of the test setup was evaluated with increase in treatment duration of 10, 20 and 30 days.

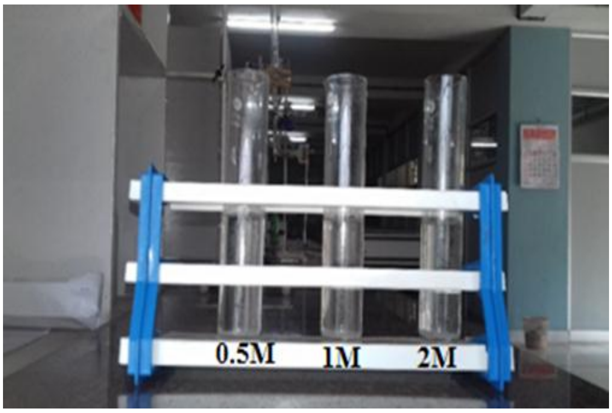

Fig 2 Test tube experiment

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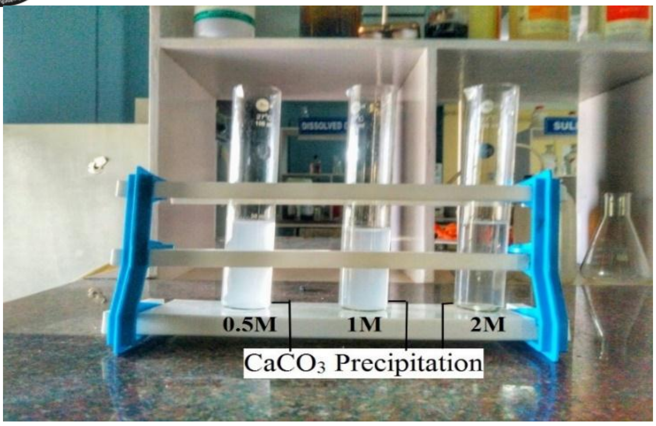

Fig 3 Test tube experiment after 10 days

Greater precipitation of calcite was observed at 1 M cementation reagent. Lesser precipitation was noticed at 2 M urea-$CaCl_2$ concentration.

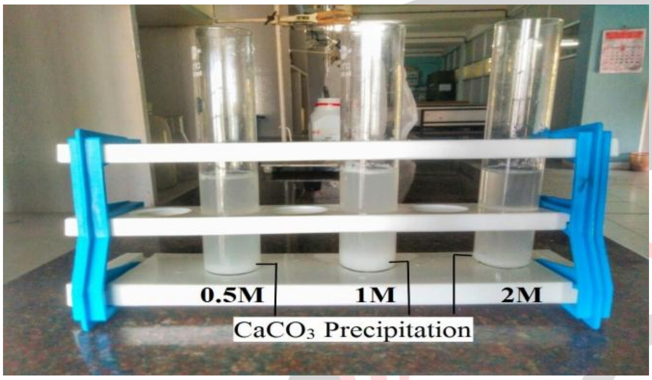

Fig 4 Test tube experiment after 20 days

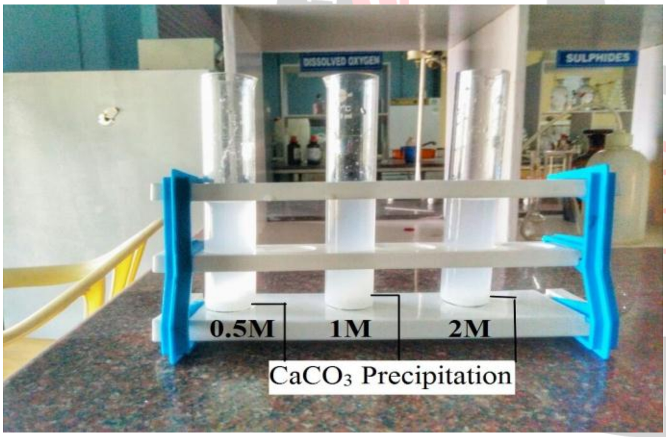

Fig 5 Test tube experiment after 30 days

The nutrient played an important role in the growth of the bacterial culture with increment of treatment days. Thus the role of Bacillus subtilis bacteria in inducing MICP and the inhibitory action of higher concentration of cementation reagent was confirmed. The MICP has increased with increase in treatment duration also.

B. OMC

Proctor test was done to determine the OMC (%) and MDD (g/cc) of treated soil samples. OMC values for various bacterial concentrations are graphically represented in following figures.

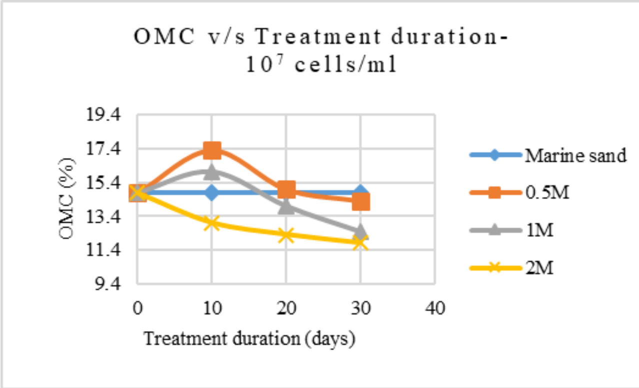

Fig 6 Variation of OMC for 10^7 cells/ml with increase in treatment days for 0.5 M, 1 M and 2 M cementation reagent

OMC has increased initially than control specimen and then reduced with increase in treatment duration. The surface area of calcite is larger compared to the surface area of soil which resulted in the initial increase of optimum moisture content.

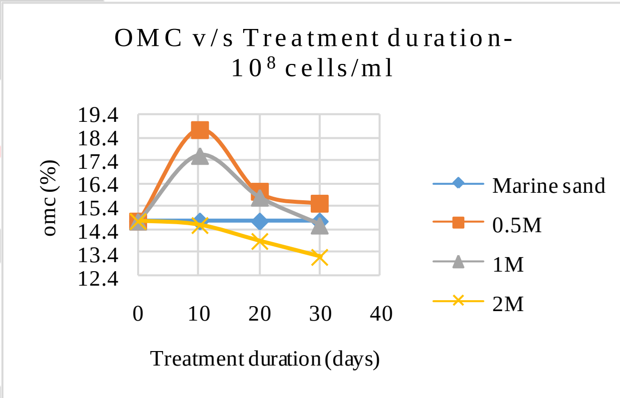

Fig 7 Variation of OMC for 10^8 cells/ml with increase in treatment days for 0.5 M, 1 M and 2 M cementation reagent

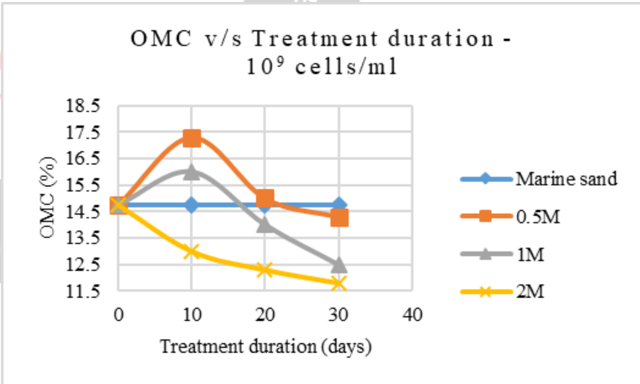

Fig 8 Variation of OMC for 10^9 cells/ml with increase in treatment days for 0.5 M, 1 M and 2 M cementation reagent

Since due to the increased calcite precipitation with time, the voids got filled up and the specimen became denser. Thus the required water content to achieve its maximum compaction has reduced.

C. MDD

MDD represents the amount of soil compaction achieved at the optimal water content. Soil compacted at its MDD will

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be densely packed with least voids. MDD values obtained are graphically represented in following figures.

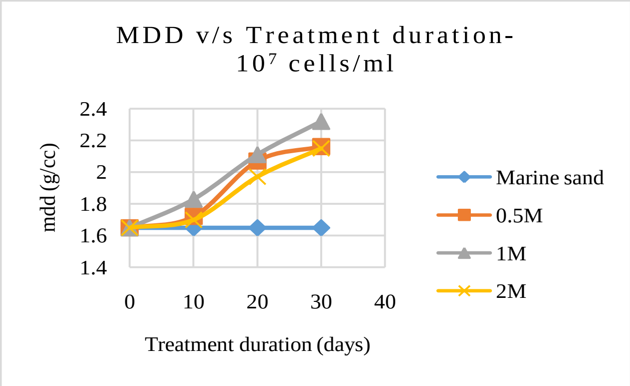

Fig 9 Variation of MDD for 10^7 cells/ml with increase in treatment days for 0.5 M, 1 M and 2 M cementation reagent

Studies have revealed that higher concentration of cementation reagent inhibits urea hydrolysis. The reason is that at higher urea-$CaCl_2$ concentration, the surrounding environment becomes saltier and thus the bacterial life gets affected.

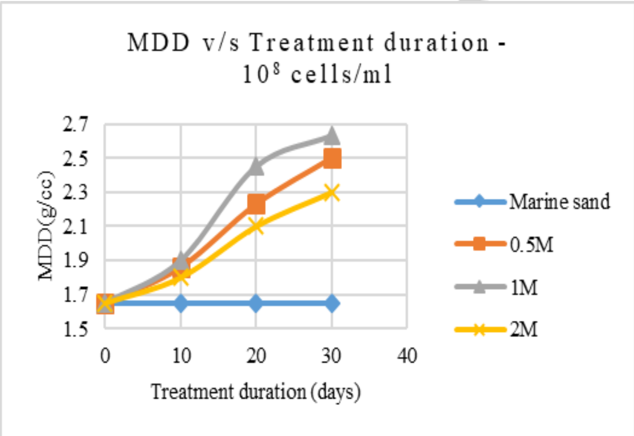

Fig 10 Variation of MDD for 10^8 cells/ml with increase in treatment days for 0.5 M, 1 M and 2 M cementation reagent

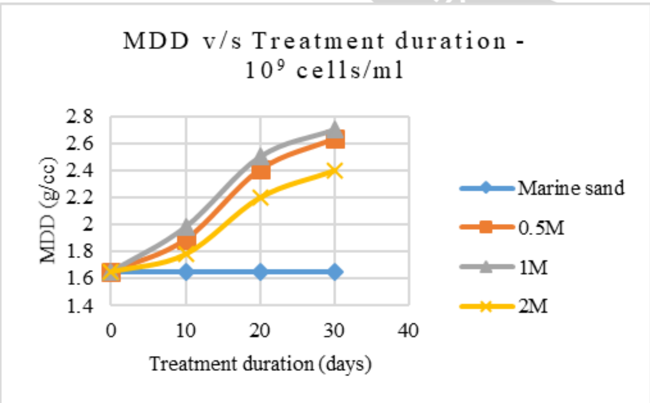

Fig 11 Variation of MDD for 10^9 cells/ml with increase in treatment days for 0.5 M, 1 M and 2 M cementation reagent

Due to calcite precipitation induced by nucleation sites of microbes, air voids present in the soil specimen has reduced and the soil became denser. Dense soil has a close arrangement of soil particles, and this contributed to more inter-particle contact points per unit volume. Thus the dry density increased with increase in concentration of bacterial cells. The maximum value of MDD is observed at 1 M concentration of cementation reagent. The value of MDD at 2 M concentrated cementation reagent on 30 days reduced compared to 1 M on 30 days. With increase in treatment duration, the precipitation remained high for longer periods, resulting in prolonged periods of nucleation. Thus the dry density increased with increase in treatment duration.

D. COEFFICIENT OF PERMEABILITY

A series of permeability tests were carried out in CMS treated with microbes and cementation reagent. The samples are filled in moulds with corresponding OMC and MDD.

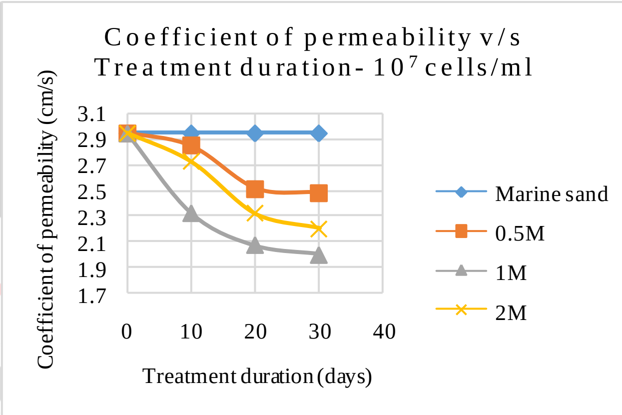

Fig 12 Variation of k value for 10^7 cells/ml with increase in treatment days for 0.5 M, 1 M and 2 M cementation reagent

The test results showed that coefficient of permeability (k) (cm/s) has reduced with increase in bacterial concentration. This is because of the $CaCO_3$ precipitation that accelerated in the presence of non-ureolytic bacteria. The precipitated calcite clogged the pores in the soil and thereby permeability reduced.

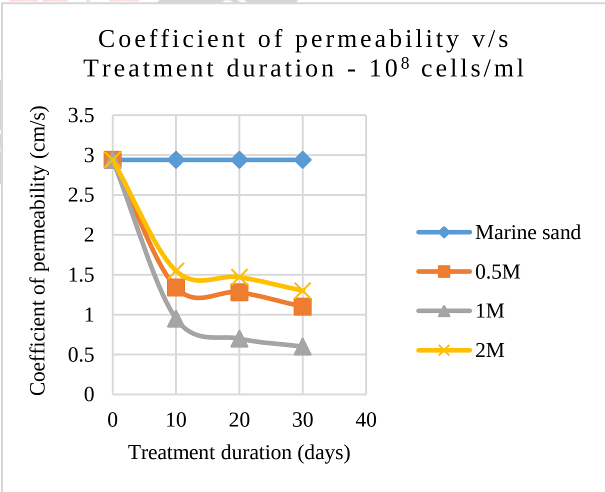

Fig 13 Variation of k value for 10^8 cells/ml with increase in treatment days for 0.5 M, 1 M and 2 M cementation reagent

For 10^7, 10^8 and 10^9 cells/ml bacterial concentration, greater reduction was observed for 1 M cementation reagent and treatment duration of 30 days.

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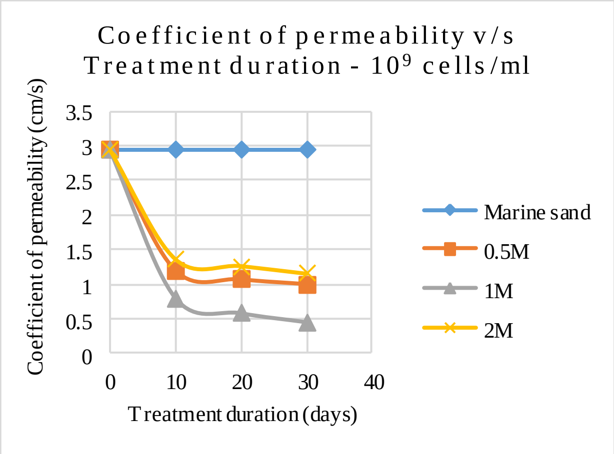

Fig 14 Variation of k value for 10^9 cells/ml with increase in treatment days for 0.5 M, 1 M and 2 M cementation reagent

The permeability of 10^9 cells/ml at 1 M and 30 days of treatment comes in the drainage property range of 10^{-5} to 10^{-4}, which is poor permeable in nature. As the concentration of molarity increased from 0.5 to 1, permeability reduced considerably. But when the molarity increased to 2, the permeability increased. The reason is that higher concentration of urea-$CaCl_2$ inhibited the urease activity and further the reaction slowed down. Also, the role of treatment duration on biogrouting was identified. Since the bacterial cells are growing, improvement was observed with increase in treatment days.

E. ANGLE OF INTERNAL FRICTION AND COHESION

The direct shear test was conducted on treated soil samples and the effects are discussed below. The tests were conducted on soil samples by initiating MICP based on corresponding MDD. The shear strength parameters such as cohesion (c) and angle of internal friction φ (°) are found out. As the soil was cohesion less, the value of c = 0. Angle of internal friction was evaluated after treatment and improvement were noted.

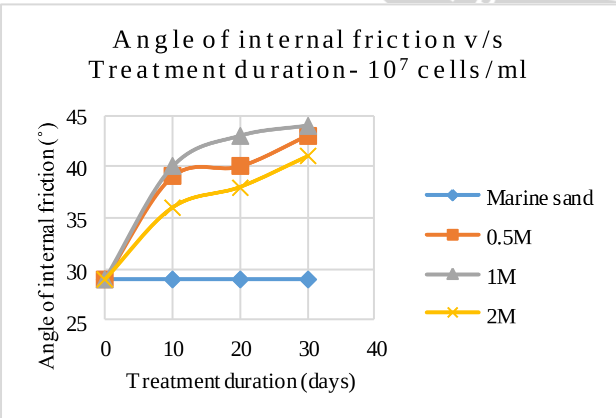

Fig 15 Variation of φ value for 10^7 cells/ml with increase in treatment days for 0.5 M, 1 M and 2 M cementation reagent

It was observed that angle of internal friction has increased with increase in bacterial concentration. The produced calcite filled up the pores and thus the density of soil specimen increased. The φ value increased due to the increment in density of the treated specimen. Thus higher increment was observed for 10^9 cells/ml at 1 M cementation reagent after 30 days.

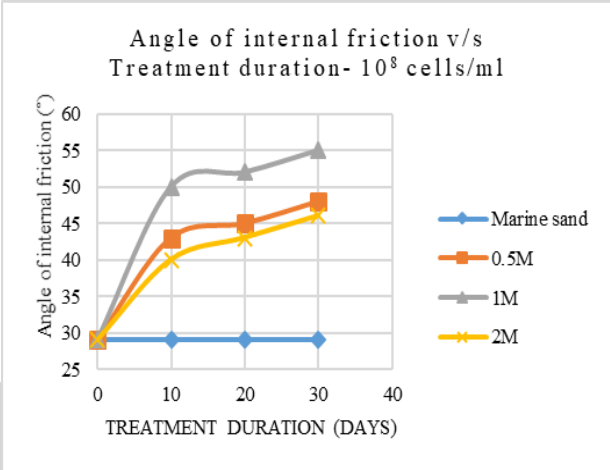

Fig 16 Variation of φ value for 10^8 cells/ml with increase in treatment days for 0.5 M, 1 M and 2 M cementation reagent

The obtained value of angle of internal friction is 69° which is greater than 45°, thus the soil comes under the category of dense sand. The shear strength of soil mass was found to be depend upon the density of soil, grain size distribution, shape and structure of the grain.

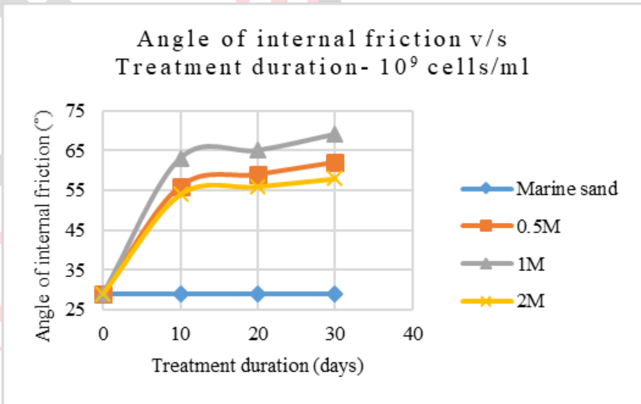

Fig 17 Variation of φ value for 10^9 cells/ml with increase in treatment days for 0.5 M, 1 M and 2 M cementation reagent

Since the metabolic activities of microbes are inhibited at higher concentrations of equi-molar urea and calcium chloride, the value of angle of internal friction reduced considerably. Also, the growing microbial cells accelerated the calcite precipitation with increase in treatment duration.

F. CBR

CBR tests were carried out on specimen treated with bacterial concentrations. CBR test is used for evaluating the subgrade and materials used in sub-base and base course. The test results have been correlated with the thickness of various materials required for the construction of flexible pavements. The graphical representation of CBR values are represented below.

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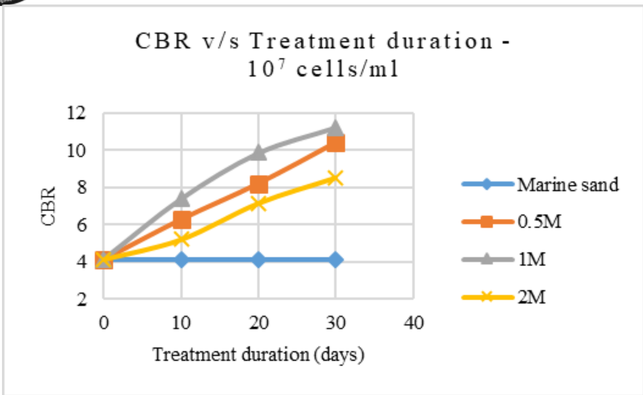

Fig 18 Variation of CBR value for 10^7 cells/ml with increase in treatment days for 0.5 M, 1 M and 2 M cementation reagent

CBR value exhibited increment with increase in concentration of bacterial cells. CBR value showed higher increment for 10^9 cells/ml at 1 M cementation reagent after 30 days. CBR value of 24.08 comes in the range 10 to 30 hence treated soil falls under the category strong soil. The bonding between the particles due to calcite precipitation, cemented the treated specimen and thus CBR value increased. Thus the treated sample can be used for pavement subgrade applications.

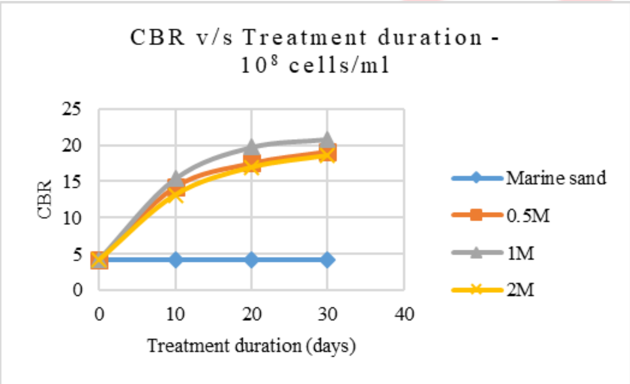

Fig 19 Variation of CBR value for 10^8 cells/ml with increase in treatment days for 0.5 M, 1 M and 2 M cementation reagent

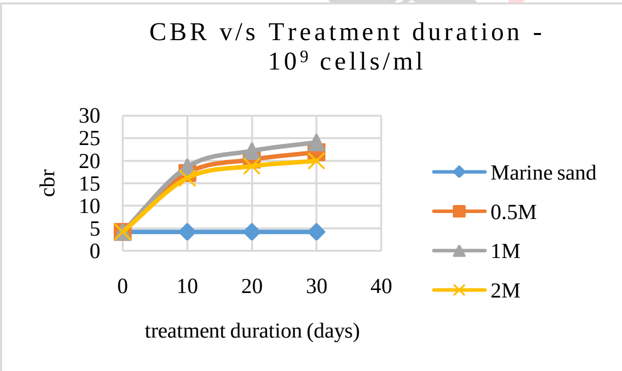

Fig 20 Variation of CBR value for 10^9 cells/ml with increase in treatment days for 0.5 M, 1 M and 2 M cementation reagent

The increase in molarity of cementation reagent up to 1 M showed increase in CBR, but beyond that CBR reduced due to inhibitory action of cementation reagent in microbial activity. The microbes used were growing cells and thus calcite precipitation accelerated over time. Thus the CBR value has increased with increase in treatment duration.

G. XRD ANALYSIS

Bacillus subtilis concentration of 1 x 10^9 cells/ml and 1 M cementation reagent gave the best results. XRD test was conducted on the sample of 1 x 10^9 cells/ml bacterial concentration, 1 M cementation reagent and at 30 days of treatment to identify the microbially precipitated crystals in treated CMS. Figure 21 shows the test result of XRD analysis conducted.

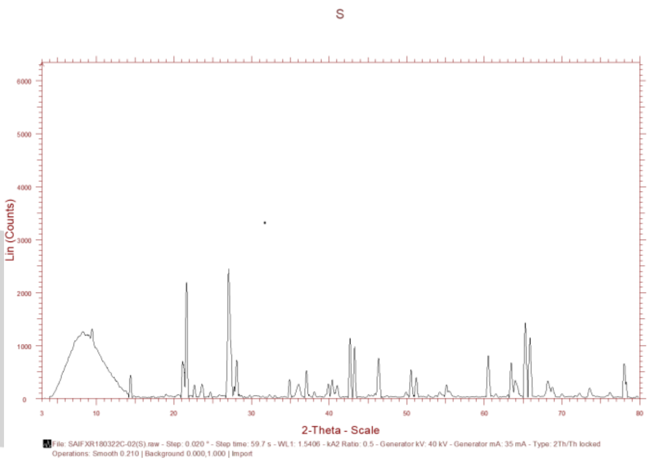

Fig 21 XRD test results on treated CMS

Larger number of peaks are observed in treated CMS sample. Thus greater calcite precipitate was confirmed by XRD analysis. Hence the ability non-ureolytic bacteria to accelerate the $CaCO_3$ precipitation was confirmed.

IV. EFFECT OF BIOGROUTING IN TREATED CMS

From the tests conducted, the usefulness of MICP in improving the shear strength, compressibility and reducing the permeability of sandy soil has been established. Bacillus subtilis concentration of 1 x 10^9 cells/ml and 1 M cementation reagent gave the best results. The calcite precipitation produced by microbial action got filled up in the void spaces and resulted in improved strength of sand samples. The percentage improvement of 10^9 cells/ml with respect to 10^8 and 10^7 cells/ml from control specimen showed only slight variation. Thus the improvement will be slowed down at further higher concentrations. Improvement in soil properties was observed with increase in treatment duration also. 10, 20 and 30 days of treatment period was selected as variables. Similar trend in percentage improvement was observed with increment in treatment duration. Hence considering the percentage improvement and economical point of view, the bacterial cells up to 10^9 cells/ml is only studied in the present work.

Applications of MICP on CMS

- Minimized settlement due to improved strength gained by filling up of voids due to reduction in permeability.
- Failure in road and airfields can be prevented due to increased density of soil mass.

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• Retaining wall failure can be tackled due to increased shear strength value.
• Erosion potential in the coastal areas can be tackled due to reduced permeability characteristics.
• Reducing liquefaction potential of soil due to increased bonding between particles caused by calcite precipitation.
• Can be suggested for pavement applications as the CBR value increased with the treatment.

V. Conclusion

In this paper study on the effect of biogrouting in compaction, shear strength and permeability characteristics of Cochin Marine Sand is presented. From this experimental study, it is revealed that

• The bacterial concentration of 10^9 cells/ml and 1 M concentration of cementation reagent at 30 days of treatment showed highest improvement of shear strength, CBR value and greatest reduction in permeability for CMS.

• OMC of sandy soil has reduced from 14.76% to 12.5% with percentage reduction of 15.13%. MDD of CMS has increased from 1.65 g/cc to 2.7 g/cc with percentage improvement of 63.64%. Formation of calcite bonds has contributed to reduction in water content and increased density of specimen.

• Coefficient of permeability value has reduced from 2.94 x 10^{-3} to 0.45 x 10^{-3}. Percentage reduction in permeability value for treated CMS was 84.69%. This may be due to the particle to particle contact and bio-clogging.

• Angle of internal friction has increased from 29˚ to 69˚ with percentage improvement as 137.93%. Higher improvement in angle of internal friction was attributed due to greater density and porous structure of sand.

• CBR value of CMS has increased from 4.14 to 24.08 with percentage improvement of 481.64%. Stronger bond between particle has contributed to greater improvement in CBR value.

• Thus the influence of non-ureolytic bacteria in accelerating the $CaCO_3$ precipitation was proved. B. subtilis promoted the addition of nucleation sites in the form of bacterial cells. Utilization of nutrient medium has improved the efficiency of MICP.

• Highest increment was observed at 1 M concentration of cementation reagent. Beyond that point properties have reduced. This is due to the inhibitory action of cementation reagent on microbial activity at higher saltier environment.

• Geotechnical properties were found to be improved with increment in treatment duration. Treatment with growing cells of microbes resulted in increased calcite precipitation.

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