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Effect of Microbially Induced CaCo3 Precipitation (MICP) Technique on Geotechnical Properties of Sand and Clay

Priyanka Prasad[1*], Bhavini Desai[2], Dr. Nitin Joshi[3]

[1]M.E.Scholar, Applied Mechanic and Structural Engineering Department, The Maharaja Sayajirao University, Baroda, Vadodara, Gujarat, India
[2]Temporary Assistant Professor, Applied Mechanic and Structural Engineering Department, The Maharaja Sayajirao University, Baroda, Vadodara, Gujarat, India
[3]Associate Professor, Applied Mechanic and Structural Engineering Department, The Maharaja Sayajirao University, Baroda, Vadodara, Gujarat, India

ABSTRACT

This study investigates the application of Microbially Induced Calcite Precipitation (MICP) on sand and black cotton soil, utilizing bacteria and varying cementation reagent concentrations. The primary objective is to enhance the geotechnical properties of these soils, which are commonly used in construction and engineering projects. MICP, a biogeochemical process facilitated by urease-producing bacteria, promotes the formation of calcite, thereby improving soil stability and strength. Experimental setups involved treating samples of sand and black cotton soil with different concentrations of bacterial culture and cementation solutions. The results demonstrate significant improvements in soil properties, including increased cohesion and reduced permeability. Variations in bacterial and cementation reagent concentrations were analyzed to determine the optimal combination for maximum calcite precipitation and soil enhancement. This study highlights the potential of MICP as a sustainable and effective technique for soil stabilization, providing valuable insights into the application of bio-cementation in diverse soil types.

***IndexTerms*:Microbially induced calcite precipitation, Cementation reagent, Bacillus subtilis, Sand, Black cotton.**

INTRODUCTION

Various challenges which are faced during and after the construction of a structure are associated with soil due to its poor bearing capacity, settlement, drainage, erosion, liquefaction, etc. The conventional ground improvement methods involve enhancing the physical, chemical, mechanical and mineral composition of the soil. These techniques involve addition of cement, lime, chemical, and supplementary cementitious material such as fly ash, rice husk ash, and blast furnace slag for soil stabilization. Cement, lime, and chemicals can lead to air pollution by emitting carbon dioxide during the manufacturing phase or usage and permanent contamination of soil and groundwater, Microbially induced calcite precipitation (MICP) is an alternative approach to ground improvement which strengthens the soil and reduces seepage by bio plugging and cementation induced by calcite precipitation. Bacteria are the dominant microorganisms in soils. They are found even at large depths in the earth's lithosphere, but in smaller numbers. There are from 10^9 to 10^{12} organisms in a kilogram of soil near the ground surface. Some bacteria can make spores to endure adverse environmental changes. Bacteria vary in shape and may be nearly round, rodlike, or spiral.

Stabilization of soil which alters the one or more properties on it to desire the requirements. In this process, Ureolytic and non-Ureolytic bacteria are used to produce urease enzymes to hydrolyze the urea into ammonium and carbonic acid. Calcium carbonate cement is precipitated due to calcium ions with the help of bacterial cells. This precipitation coats the soil particle form soil cement matrix, thus increases desired mechanical properties include strength and stiffness of soil matrix.

Aims & Objectives

- The primary aim of the MICP technique is to enhance soil properties by inducing the formation of calcite. The precipitation of calcite within the soil matrix results in, Improved soil strength, Reduced permeability, Enhanced durability Making it ideal for construction and infrastructure projects.
- To identify suitable microbial strains capable of promoting calcite precipitation in the targeted soil.

- To optimize the MICP process parameters, such as microbial concentration, nutrient supply and environmental conditions, to achieve the desired soil improvements.
- To assess the effectiveness of the MICP technique in enhancing soil properties through laboratory experiments.
- To evaluate the long-term stability and durability of MICP.
- To explore potential applications of MICP in different geotechnical and environmental projects.

MATERIALSANDMETHOD

Black cotton soil

Black cotton was used due to its swelling, shrinkage and low permeability. Black cotton soil used in this project was sourced from Karjan a city and Municipality in Vadodara District. Comprehensive laboratory test such as hydrometer analysis, liquid limit, plastic limit, shrinkage limit, free swell index and specific gravity was conducted to determine physical properties of black cotton soil according to the method specified in the relevant Indian standard code of practice.

Table 1 Index properties of Black Cotton Soil

Description of test	Standard code applicable	Results
Liquid limit %	IS 2720- Part-5	40.04
Plastic limit %		22.49
Plasticity index		25.20
Shrinkage limit %	IS 2720-Part-6	15.41
Free swell index %	IS 2720-Part-40	80
Specific gravity	IS 2720-Part-3	2.61
Maximum dry density, kg/m^3	IS 2720-Part-7	1520
Optimum moisture content %		19.2

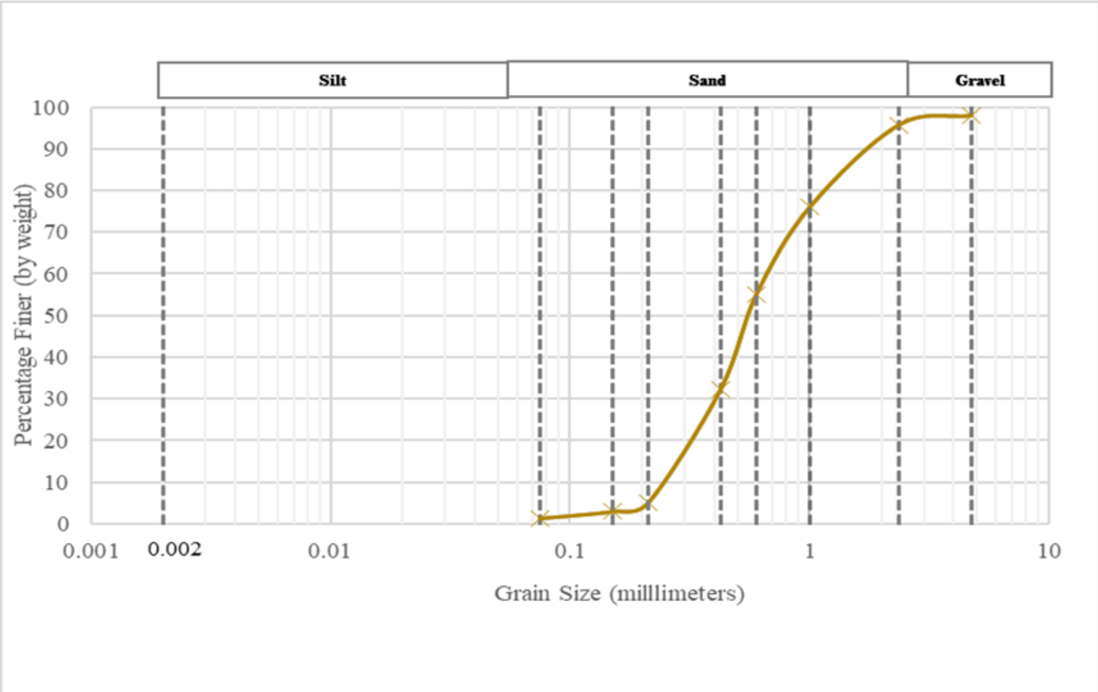

Fig -1: Particle size distribution curve for black cotton soil

Sand

Sandy soil was preferred for this experiment due to its granular nature, lack of cohesion and high permeability. Sand used in the project was sourced from the Orsang River flowing through Bhadarpur village in Sankheda taluka in district Vadodara in Gujarat state. Before being utilized, the sand underwent a natural drying process to eliminate excess moisture test. Laboratory test such as sieve analysis and relative density test was performed to determine the properties of sand.

Table 2 Index properties of Sand

Description of test	Standard code applicable	Results
D10 (mm)	IS 2720-4 [22]	0.251
D30 (mm)		0.408
D60 (mm)		0.694
Coefficient of Uniformity, Cu		2.77
Coefficient of Curvature, Cc		0.95
IS Soil Classification		SP
Maximum Density (kg/m3)	IS 2720 -14 [24]	1700
Minimum Density (kg/m3)		1450

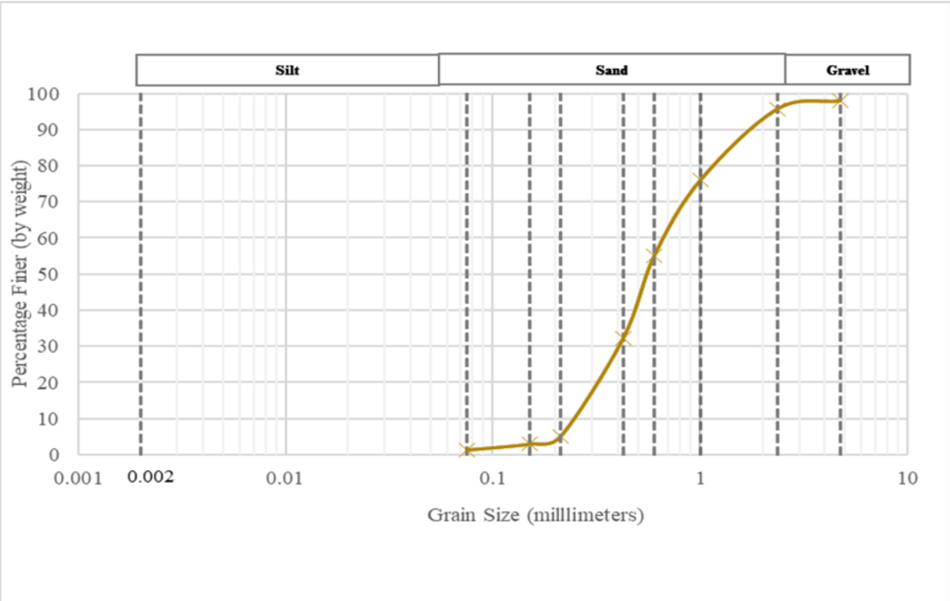

Fig -2: Particle size distribution curve for Sand

Bacteria

The present study used Bacillus subtilis by Green life biotech laboratory which was locally available in the market with 1x 10^9 cfu/ml, it is also known as B. subtilis. Bacillus subtilis is a gram-positive road-shaped bacterium known for its robustness and versatility making it valuable in various applications. It is a spore forming bacterium producing endospores that are highly resistant to environmental stress. It is nonpathogenic and widely used. Thus, it can thrive in diverse environment.

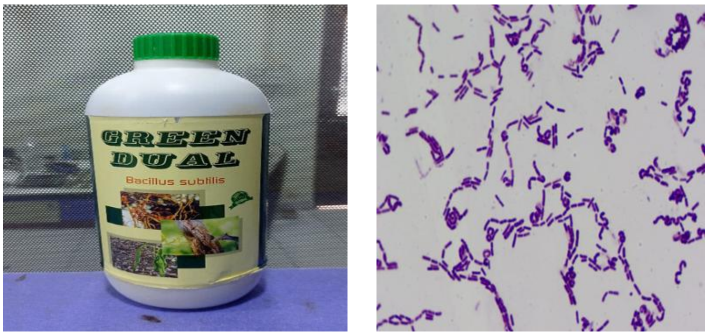

Fig -3: Bacillus subtilis and microscopy images

Cementation reagent

The cementation reagent used were urea, calcium chloride and nutrient broth. The chemical considered for making cementation reagent were based on literature review and under the guidance of Flourish biotech laboratory, Vadodara. Amount of chemical required for preparing 100 ml solution are mentioned in the below table.

Table -3: Cementation reagent

Chemical	Amount	Amount	Amount	Amount
	0.25M	**0.50M**	**0.75M**	**1M**
	gm	gm	gm	gm
Urea	1.60	3.20	4.80	6.40
Cacl2	2.675	5.350	8.025	10.700
Nutrient broth	0.3	0.3	0.3	0.3

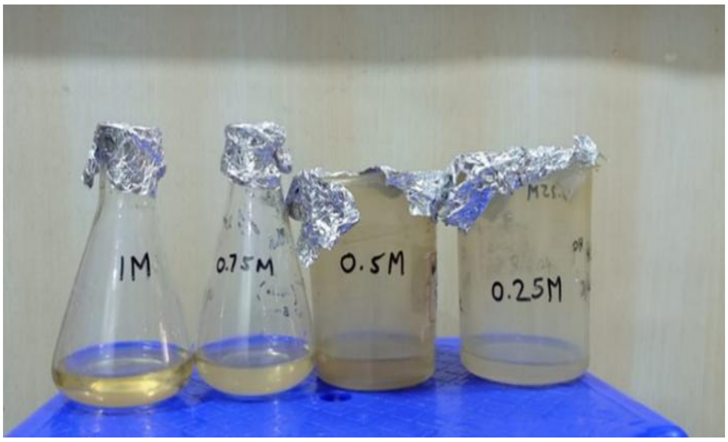

Fig -4: Cementation reagent

METHODS

Sample preparation

The sand and black cotton soil was mixed manually with different proportions of bacillus subtilis and cementation reagent as mentioned in the table below, an initial trial was run on 100 gm of soil sample of sand and black cotton to know the effectiveness of the bacteria and the cementation reagent and to asses viable results. Based on the results further soil sample were prepared to carry out experimental test, the sample was cured for 14 and 28 days, after whichvarious geotechnical test were conducted on the prepared sample.

Table -4: Mix designation

Mix designation	Mix
2:98	0.25M cementation reagent + 2% bacillus subtills + 98% soil
4:96	0.50M cementation reagent + 4% bacillus subtills + 96% soil
6:94	0.75M cementation reagent + 6% bacillus subtills + 94% soil
8:92	1.00M cementation reagent + 8% bacillus subtills + 92% soil

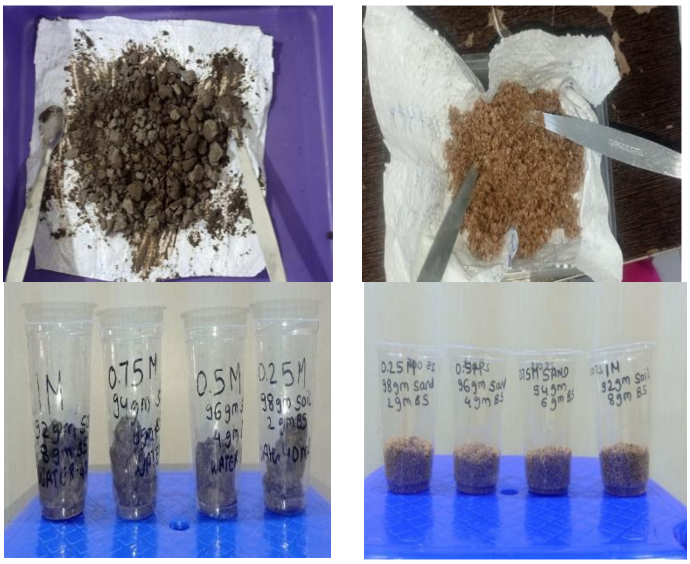

Fig -5: Sample preparation

EXPERIMENTAL DETAILS AND TREATMENT PROCEDURE

Standard proctor test (IS:2720 Part 7-1980)

Standard proctor test was performed on the black cotton soil in a proctor mold of 1000 cc with an internal diameter of 100mm and height of 127.3 mm. The soil was compacted in 3 layers giving 25 blows per layer with the 2.6 kg rammer. The need of the test was to know the maximum dry density and optimum moisture content of soil, so that to identify the moisture level at which soil can reach its high density, ensuring stability and strength with minimal water usage.

Fig -6: Standard proctor test

Triaxial test (IS:2720 Part 11-1993)

PVC pipe were used to cast the sample of size 38mm dia and 76mm height, black cotton soil was compacted in the mold with MDD 1.52gm/cc and OMC 19.2%. Three numbers of samples were prepared for each concentration of bacteria for 14 and 28 days, cementation reagent was applied at an interval of 7 days for each sample for 14 days and 28 days

Fig -7: Triaxial test

Consolidation test (IS:2720 Part 15-1986)

Consolidation test was performed on an undisturbed bacteria treated black cotton soil sample, by extracting it with the help of sampler having height 20 mm and dia 60 mm, the sample was placed in the consolidation apparatus and incremental loads were applied while measuring deformation over time. The test was performed to identify the key parameters like coefficient of consolidation and compression index, which could help in predict future settlements under similar conditions.

Fig -8: Consolidation test

Direct shear test (IS:2720 Part-13-1986)

Samples for direct shear tests were prepared in four different trays measuring 28.4 cm in length, 23.4 cm in width, and

8 cm in height at 0.25M, 0.5M, 0.75M and 1M concentration. The depth of sample was set at 4 cm. the amount of sand used was determined based on achieving a density of 1.7 gm/cc and the volume of the tray. Initially, bacteria cementation reagent and soil were mixed. Then, the cementation reagent was added at an interval of 7 days once after mixing for 14 days and three times after mixing for 28 days Subsequently, the test was performed

Fig -9: Direct shear test

California bearing ratio test (IS:2720 Part-16-1987)

The CBR test sample was prepared in a mold with a diameter of 150 mm and a height of 175mm, including a detachable collar of 60 mm, as per IS 2720 part 16 for both sand and black cotton clay. Cementation reagent was applied at an interval of 7 days on both samples, and after 28 days, the test was performed on the bacteria – treated sample.

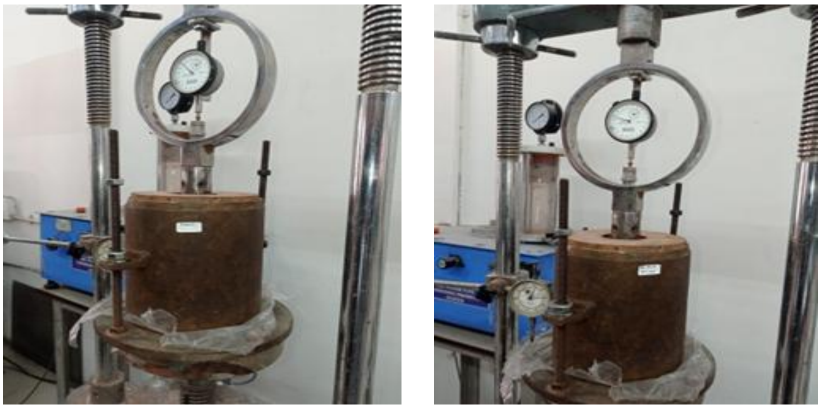

Fig -10: California bearing ratio test

Permeability test (IS:2720 Part -17-1986)

Permeability test sample was prepared as an undisturbed sample inmold of dia 5 cm, and height 10 cm, with density of 1.7 gm/cc and 10% of OMC for sand and 1.52 gm/cc MDD and 19.2 OMC for black cotton soil at four different concentrations. Cementation reagent was applied at an interval of 7 days, and after completion of 28 days constant head test for sand and falling head test for clay was performed on the treated sample.

Fig -11: Permeability test

Determination of calcium carbonate test (IS:2720 Part-23-1976)
5 gm of treated soil was allowed to stir, mix and settle for half an hour in100 ml of 1N HCL solution in a conical flask. Then 25 ml solution was transferred in other flask by filtering it with the help of filter paper 6 to 8 drops of bromothymol blue was dropped in the flask and stirred. Then the 25 ml filtered solution was titrated against 1N NaoH solution till the color changes to blue, and the burette reading was noted. Thus, the percentage of Caco3 present in soil was computed by the amount of 1N NaoH consumed by 5 gm of soil.

Fig -12: Determination of caco3

RESULTS AND DISCUSSIONS

The results obtained are mentioned in below table and graphs.
Triaxial test (IS:2720 Part 11-1993)

Table -5: Triaxial Test Results Black cotton soil

Sample	Cohesion, kPa		Angle of internal friction, Deg.	
Natural state	131.41		3.24	
	14 Days	**28 Days**	**14 Days**	**28 Days**
0.25M	137.29	139.25	3.17	5.04
0.5M	149.06	167.69	6.12	8.15
0.75M	138.27	140.24	3.98	5.30
1M	154.95	167.69	5.65	6.97

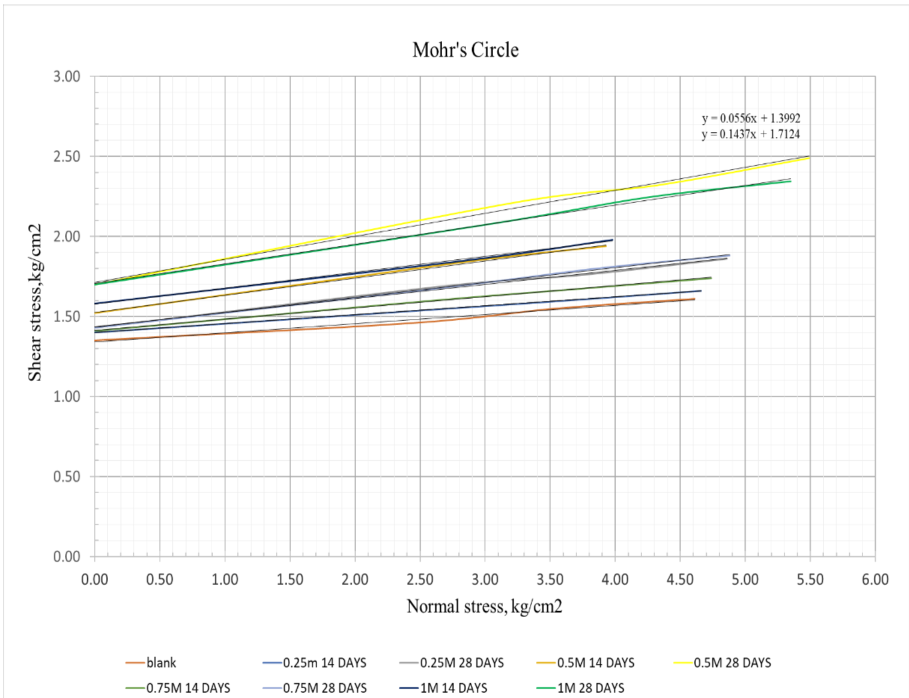

Fig -13: Failure envelop of natural black cotton soil and MICP treated soil samples at different concentration for 14 and 28 days

Direct shear test (IS:2720 Part-13-1986)

Table -6: Direct shear Results Sand

Sample	**Angle of internal friction, Deg.**	
Natural state	36.66	
	14 Days	**28 Days**
0.25M	37.5	37.91
0.5M	39.82	41.03
0.75M	37.88	39.12
1M	37.07	38.32

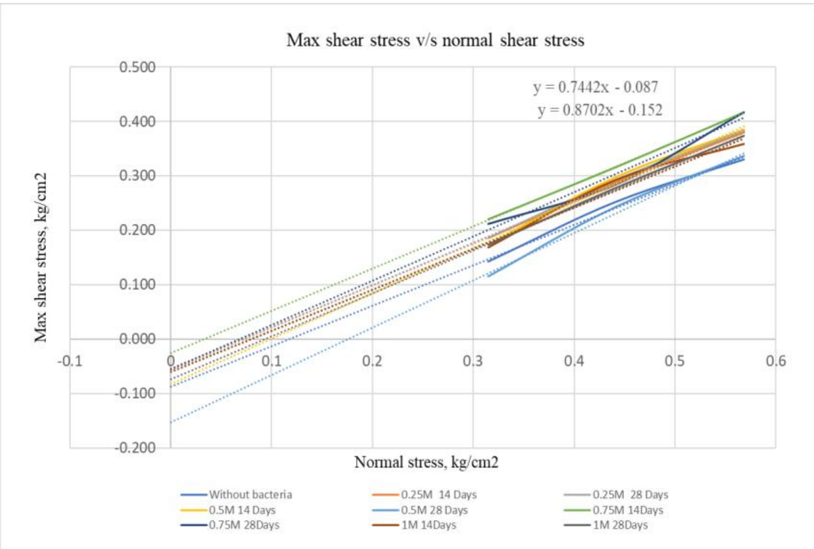

Fig -14: Max. shear stress v/s normal stress graph for natural sand andMICP treated sand samples at different concentration for 14 and 28 days

Permeability test (IS:2720 Part -17-1986)

Table -7: Permeability test results for 28 Days

Sample	0.5M Black cotton	0.5M Sand
	m/sec	m/sec
Natural state	0.05414	0.061
0.25M	0.04162	0.059
0.5M	0.03258	0.054
0.75M	0.03407	0.053
1M	0.03703	0.055

Consolidation test (IS:2720 Part 15-1986)

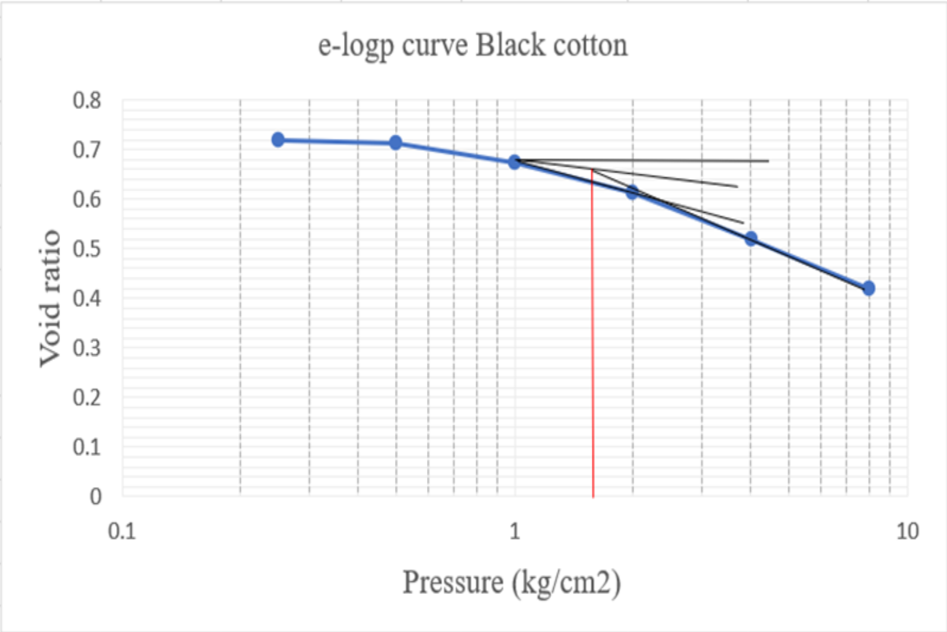

Fig -15: e-logp curve black cotton soil natural state

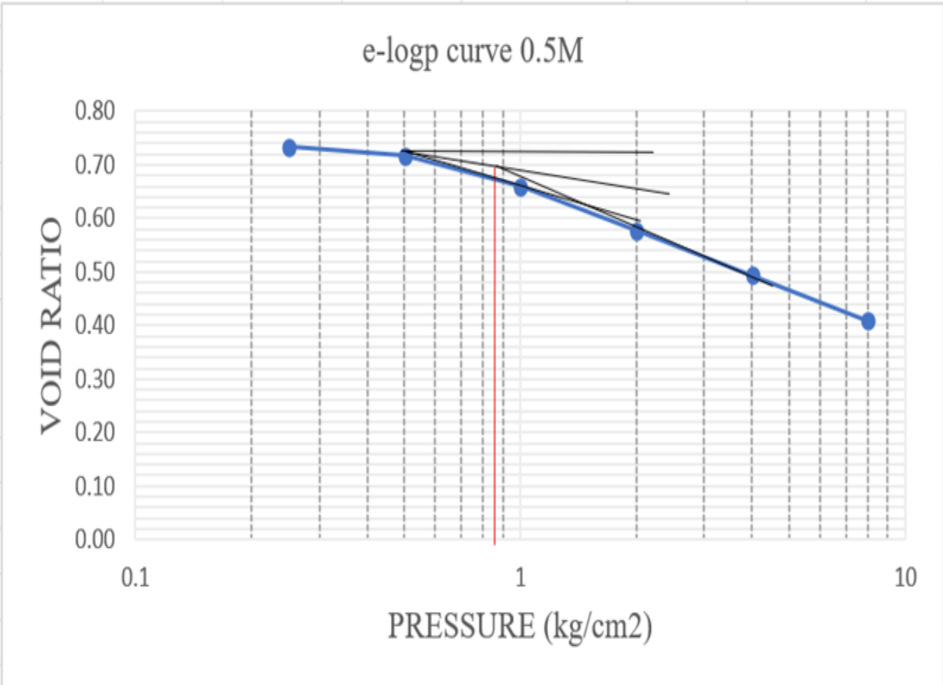

Fig -16: e-logp curve black cotton soil 0.5M

Table -8: Consolidation test results

Consolidation Test Black cotton		
Parameters	**Natural state**	**0.5M 28Days**
Preconsolidation pressure (Pc), kPa	156.906	78.4532
Compression index (Cc)	0.33	0.280
Coefficient oof compressibility (av)	0.060	0.081
Coefficient of volume change (mv)	0.036	0.049

California bearing ratio test (IS:2720 Part-16-1987)

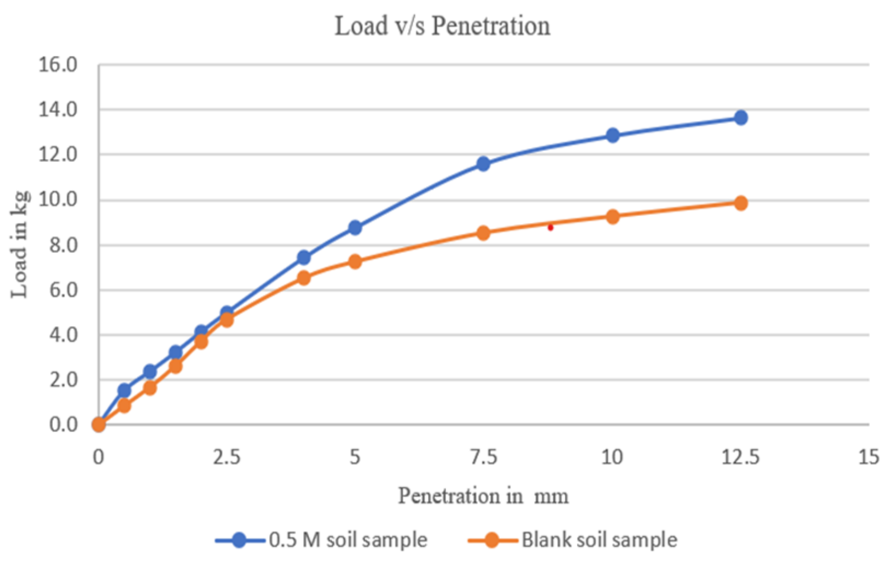

Fig -17: Load v/s Penetration graph black cotton soil

Table -9: C.B.R test results black cotton soil

Black cotton soil		
CBR	**Natural state**	**0.5 M 28 Days**
CBR at 2.5 mm penetration in %	6.721	7.157
CBR at 5 mmpenetration in %	6.925	8.379

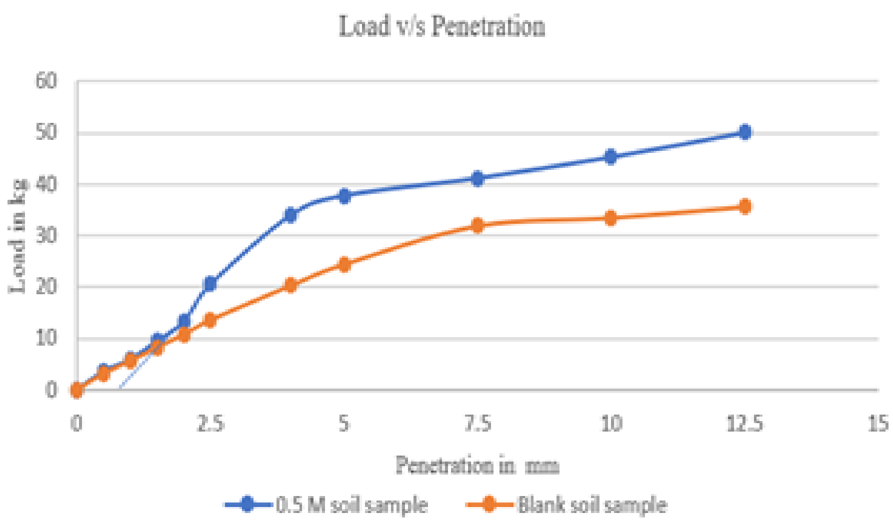

Fig -18: Load v/s Penetration graph sand

Table -10: C.B.R test results sand

Sand		
CBR	**Natural state**	**0.5 M 28 Days**
CBR at 2.5 mm penetration in %	6.721	7.157
CBR at 5 mmpenetration in %	6.925	8.379

Determination of calcium carbonate test (IS:2720 Part-23-1976)

By measuring the concentration before and after treatment, effectiveness of microbial process and its extent to improve soil properties was determined.

Table -11: Caco3 results in % black cotton soil

Determination of Caco3 in % Black cotton soil						
Initial trial	**Days**	**Black cotton**	**0.25 M**	**0.5 M**	**0.75 M**	**1 M**
	14	0	13.4	17.5	12	10.5
Triaxial test	14	0	12.6	15.5	11.2	10.3
	28	0	12.8	16.2	12.4	11
Consolidation test	14	0	12.4	16.4	11.7	9.2
	28	0	13.1	16.8	12.2	10.3
Permeability test	14	0	12.2	18.1	13.2	10.7
	28	0	12.7	18.6	14.2	10.9

Table -12: Caco3 results in % sand

Determination of Caco3 in % Sand						
Initial trial	**Days**	**Sand**	**0.25 M**	**0.5 M**	**0.75 M**	**1 M**
	14	0	5.5	11.5	7.5	6.5
Direct shear test	14	0	4.2	10.4	8.5	6
	28	0	4.9	11.5	9.7	6.3
Permeability test	14	0	5.2	10.7	7.2	6.3
	28	0	5.8	11.2	7.7	6.5

CONCLUSIONS

The bacterial treatment on black cotton soil and sand has significantly altered their engineering properties, indicating potential applications in geotechnical engineering. The consolidation test results showed that bacterial treatment reduces the preconsolidation pressure and compression index while increasing the compressibility and volume change coefficients. The permeability test demonstrates a reduction in permeability. The California Bearing Ratio (CBR) test indicates improved soil strength, and the direct shear test shows increased shear strength over time. The triaxial test results reveal higher cohesion and angle of internal friction with increased bacterial concentration and curing time, particularly at 0.5M concentration. The determination of calcium carbonate indicates that 0.5M concentration is most effective for $CaCO_3$ formation, while higher concentrations may inhibit it. Overall, bacterial treatment improves soil stability and strength, offering promising applications in construction projects.

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