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Enhancing the Strength Parameters of Dispersive Soil with Microbes and Jute Fibres as Sustainable Alternative

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ABSTRACT

Aim of this study was assessing the characteristics of dispersive soil based on percentage of dispersion and degree of dispersion and to improve the strength of soil using microbes. This research has utilized the Microbial Induced Calcium Carbonate process (MICP) in conjunction with jute fibre for the improvement the erosive resistance in dispersive soil. Calcite formation occurred as a consequence of microbial biomass in voids of dispersive soil. Calcium carbonate was synthesized in the gaps of the soil matrix during the microbiological process. *Bacillus sphaericus* bacteria were used in this experiment, along with a 1 cm length of jute raw fibre of 1 cm long and a cell concentration of 6.4E+08 CFU mL⁻¹. The findings of the Unconfined compressive strength (UCS) test showed following of MICP treatment with and without jute fibre augmentation, UCS values causing the 11 and 13 times. Crumb test findings showed no colloidal solution was generated after microbial treatment, which was used for confirmation of the degree of dispersiveness reduction. Addition of jute fibres indicating better precipitation values of more than 4% due to the internal bonding strength. Ground renovation through microbial cementation yielded promising benefits, suggesting sustainability.

Keywords: *Bacillus sphaericus*, dispersive soil, Jute fiber, microbial induced calcite precipitation (MICP), unconfined compressive strength.

INTRODUCTION

Dispersive soil is sensitive to water erosion. It poses a challenge to the construction of waterretaining structures such as embankments, dams, and canals. Wet soil loses its strength and is eroded by water, causing problems with stability during excavation and construction (Moravej et al., 2018). Many earthen dams, hydraulic structures, and roadway embankments face danger through dispersive soil erosion. Dispersion-induced erosion which can cause piping processes in earth dams, account to about 37% of all earth dam failures globally (Goodarzi and Salimi, 2015). Dispersive soil particles erode in running water as a result of deflocculation, which occurs when interparticle repulsion force surpasses the attraction force, causing suspension of soil particles. The concentration of sodium ions inside the soil structure determines the dispersiveness of the soil primarily. Sodium cations (Na⁺) predominate in dispersive clays, whereas conventional clays have more calcium, potassium, and magnesium cations in the pore water, and hence have a higher percentage of exchangeable sodium cations. Sodium clinging to montmorillonite, rather than the Na+ in the pore water, appears to be the source of dispersiveness. Minerals and dissolved salts in the water play a significant role in erosion in dispersive soil. The presence of colloidal materials in dispersive soil, ground improvement like mechanical methods unsuitable (Suriya et al., 2020). Cement is predominant material used in soil stabilization, leads to emission of Carbon dioxide

(Sharma et al., 2021). Literature study shows the dispersive process influenced by gradation, soil mineralogical composition and availability of water (Stocks-Fischer et al., 1999; Ng et al., 2012; Pakbaz et al., 2012; Imran et al., 2020).

Intergranular attraction and specific gravity of soil particles determine the erodibility of the soil. The effect of cation type, electrolyte concentration, and pH are the main elements that have influence on dispersion process. Clay particles in a dispersive soil may scatter and remain suspended in water once exposed to water. As a result, use of several experiments for the identification of the dispersive soil and estimation of its dispersive potential using physical and chemical parameters was made in the investigation. Increasing the need for identification of dispersive clay soils using methods such as the double hydrometer, ESP, SAR, Pinhole test, and material composition, tested in laboratory.

Microbial calcite precipitation is a viable approach for stabilizing dispersive soils through biomineralization (Pakbaz et al., 2021); Sajadi et al., 2021). Microbially produced calcite in the soil matrix causes additional bonding strength, bringing down erodibility potential (Comadran-Casas et al., 2021). The aim of this present study is to stabilize dispersive soil using Bacillus sphaericus, for a decrease in the degree of dispersiveness. Jute fibres were added for enhancement of calcite production and bonding between the soil grains. Microbes present in the soil took sufficient nutrient and precipitating calcite in the voids, due to metabolic activity. This process is called microbial induced calcite precipitation (MICP) (Suriya and Sangeetha, 2021). Curing period, bacterial cell density, precipitating agent concentration, and temperature are the variables that have been investigated for the strength procurement during the study (Dejong et al., 2006; Cheng et al., 2012; He et al., 2016). Decrease in pH during microbial activity and the stability of exchangeable sodium ions are considered as key contributors to the drop observed in erosion potential of the soil samples. Ultimately, this process leads to the sustainable soil stabilization. Tests were performed before and after biological stabilization for the measurement of the level of dispersion in the soil samples.

MATERIAL AND METHODS

Soil samples used in the investigation were collected on May 2021 from Chengalpattu district

which has a latitude and longitude of 12° 41' 38.1588" N and 79° 58' 32.3832" E, Tamil Nadu, India. Laboratory study was conducted for the soil for the analysis of the index properties. Confirmation tests were also conducted for getting knowledge of the degree of dispersion of the soil. These includes double hydrometer test, pin hole test, Cation exchange capacity (CEC), Exchangeable sodium percentage (ESP), Sodium Adsorption Ratio (SAR) and X-Ray diffraction methods. *Bacillus sphaericus* was purchased from MTCC, with a collection account number 7542, used for the current study.

Characteristics of dispersive soil

The specific gravity of the soil was found as 2.61 using the density bottle method. Soil samples were identified for predominantly silt and clay particles of 63% and coarse-grained particles of 37%. Figure 1 is the grain size distribution curve, and Table 1 lists the properties of the soil and its corresponding values. Double hydrometer test was carried out with and without chemicals additives considering the presence of a higher percentage of fine grained. The results of a double hydrometer test graph are shown in Figure 2. Table 2 shows the classification of soil dispersiveness, and degree of dispersion for a study of the current soil was assessed as 83.2% (Maharaj et al., 2013).

Pinhole test

It was performed in accordance with BS 1377: Part 5:1990 clause 6.2, as illustrated in fig 3. The test is a direct, qualitative evaluation of clay soil dispersibility and, as a result, colloidal erodibility. A cylindrical soil specimen with 25 mm length and 35 mm diameter was punctured with a 1 mm diameter hole (Djokovic et al., 2018). Flow rate and outlet run-off turbidity of distilled water were measured at head levels of 50, 180, and 380 mm. Flow velocities of roughly 30 to 150 cm s⁻¹ were achieved with 50, 180, and 380mm heads at hydraulic slopes of around 2 to 15. The test was carried out with its natural water content of soil. For the current study, the flow rate at natural content was finally optimized as 1.3 ml s⁻¹ under 50mm head . Hole size was increased to 1.8 mm from 1mm diameter. The discharge cloudiness at the end of the test was observed to be dark. Turbidity of the outlet runoff water was used in the determination of the specimen's erosion resistance.



Fig. 1. Grain size characteristics curve of soil



Fig. 2. Double hydrometer curves for soil

Table	e 1.	Properties	of di	ispersiv	ve soil	

Description parameters	Values
Gravel (%)	1
Sand (%)	36
Silt (%)	29
Clay (%)	34
Gs	2.61
Classification	CI
Dry density (kN/m ³)	15.5
OMC (%)	22.5
Liquid limit $W_L(\%)$	56.33
Plastic limit W _P (%)	33.02
Shrinkage limit Ws (%)	9.9
Plasticity index lp (%)	23.31
Free swell index FSI (%)	26.1

Table 3 shows the results of the pinhole test carried out according to the ASTM classification system. Figure 4 shows the modus operandi for the verification of pinhole test results through the dissecting of soil samples in addition to characterizing the discharge quality.

Cation exchange capacity

A 50 gm oven-dried soil sample was placed in a conical flask with 100 ml of 1N of NH_4OAc . It was then shaken for 60 min before being kept overnight. The contents were transferred to a buchner funnel using a filter paper. The soil was leached with 400 ml of NH_4OAc , 80 to 100 ml at a time. It took 1 hr for the leaching rate to reach



Fig. 3. Pin hole apparatus

its maximum. The leachate was kept for the determination of the amount of sodium that could be exchanged. The soil was then leached with 450 ml of a 10% potassium chloride (KCL) solution with a pH of 2.5, with the effluent collected in a flask. The liquid was placed in a volumetric flask and increased to 500ml. A distillation flask was filled with 25 to 50 ml of the aforesaid extract, a few drops of phenolphthalein indicator, and 40% sodium hydroxide (NaOH) until the contents became alkaline. The distillate was collected in 4% boric acid after being distilled with ammonia. Sulphuric acid (H_2SO_4) was used for the titration of absorbed ammonia (Shainberg et al., 1980). Cation exchange capacity (CEC) value was found as 50 meq/100g using (Eq. 1).

$$CEC \frac{meq}{100} g = \frac{N \times R \times final \text{ volume of leachate } \times 100}{volume of Leachatetaken \times \text{wt. of soil}} (1)$$

where: N – normality of H_2SO_4 , R – ml of H_2SO_4 required for titration.

Exchangeable sodium percentage

The NH₄OAc extract was transferred to a 250ml beaker and evaporated to dryness on a water bath for exchangeable sodium determination during CEC measurement (Eq. 3) (Saidi, 2012). The residue was digested on a water bath for 30 min after the beaker was covered with watch glass. 10 ml 6N HCl (Hydrochloric acid)

S.No.	Percent Dispersion	Degree of dispersion
1	<30	Non-dispersive
2	30 to 50	Intermediate
3	>50	Dispersive
4	83.2	Dispersive soil

Table 3. Pinhole test parameters

S.No	Test Parameter	Values Head (mm)
1	Head (mm)	50
2	Final Flow through Specimen (ml s ⁻¹)	1.3
3	Cloudiness at the end of flow	Dark
4	Hole size (mm)	1.8
5	Dispersive Classification	D1

was added, and the contents were well mixed by stirring. The contents were filtered using Whatman No. 42 filter paper after the addition of 15ml of distilled water. The filtrate was collected in a 100ml volumetric flask and cleansed before being made up to 100 ml. The amount of sodium in the filtrate was calculated using a flame photometer. Exchangeable sodium Percentage (ESP) was found as 14 using (Eqn. 2) with value was greater than the 10, this was confirmed as Dispersive soil.



Fig. 4. Pin hole soil samples before and after the test

$$Na \frac{meq}{100}g = \frac{10 \text{ x R x final volume of leachate x 100}}{Weight of thesoil X 23 X 1000}$$
(2)

where: R – galvanometer reading

$$ESP = \frac{Exchangeable \text{ sodium } x \text{ 100}}{\text{cation exchange capacity}}$$
(3)

Salt seepage occurs in soils with ESP values greater than 10. Table 4 shows the criteria for classification of dispersive clays using ESP data. Table 4 shows the ESP determined for the tested soil sample, and also the classification.

Sodium adsorption ratio

Measurement of soluble sodium related to soluble divalent cations like calcium and magnesium in a soil water solution is called as Sodium Adsorption Ratio (SAR). It can be used for the determination of the exchangeable sodium percentage of soil that has been equilibrated with a specific water solution. The SAR (sodium absorption ratio) of the pore water is a metric used in the estimation of the involvement of sodium in dispersion when free salts are present (Robbins, 1984).

$$SAR = \frac{Na}{\sqrt{0.5 (Ca + Mg)}}$$
(4)

The amount of available sodium in soil was determined by transferring 5 g air dried soil to a 250 ml conical flask and adding 100 ml 1N ammonium acetate. After 30 min of shaking, the contents were filtered through Whatman No. 40. The amount of sodium in the filtrate was calculated using a flame photometer. Details of categorization are provided in Table 5. SAR value noted was 13.6 using Eqn. 2.

 Table 4. Dispersive percentage based on ESP

S.No.	ESP	Degree of dispersion	
1	<7	Non-dispersive	
2	7 to 10	Intermediate	
3	>10	Dispersive soil	
4	14 - Current study	Dispersive soil	

X-Ray diffraction analysis

It is ideally suited for identification of the clay minerals. The samples were analyzed using the X-ray diffraction method and Bragg's law. X-rays were coated the soil sample for the assessment of the mineral parameter in relation to wavelength. The data was collected using an X-ray diffractometer (X'pert series III) and analyzed using high score plus 3.0 software. The predominant minerals identified were 1. Quartz, 2. Montmorillonite, 3. Chlorite, 4. Carbonate, 5. Vermiculate, 6. Quartz and 7. Chlorite. The XRD images observed are shown in Figure 5.

Isolation of microbial strains

The *Bacillus sphaericus* MTCC bacterial strain purchased was used in morphological studies performed. Details are provided in Table 6. The present organism satisfies the temperature, pH, Bacterial concentration and well driven reactions with added nutrients during the process of MICP. It has the ability to withstand at different temperature and higher pH for its survival. Table 7 shows the strains isolated from the glass vials



Fig. 5. X-Ray diffraction pattern for soil

 Table 5. Dispersive percentage based on SAR

S.No.	SAR	Degree of dispersion
1	<6	Non-dispersive
2	6 to 10	Intermediate
3	>10	Dispersive soil
4	13.6 - Current study	Dispersive soil

and cultivated in basic nutritional media. It has the ability to create urease at the end of metabolic activity and to survive in an alkaline environment for long periods of time. In addition, the cells were diluted on sterile normal saline before being cultivated in calcium carbonate precipitation media (Consoli et al., (2009); Teng et al., (2020)). For 3 to 5 d, the incubation was done aerobically at 37°C. The isolated cultures were kept at 4°C until their used (Fig 6a).

The isolated colonies were subjected to Gram Staining for primary identification (Fig 6b). The cultures were smeared onto a clean grease free slide, then air dried and heat fixed. Then crystal violet was added and allowed to stand for 1 min and then washed with tap water. Gram's iodine solution was added and allowed to stand for 1 min and then washed with tap water. The smear was then subjected to decolorization using 90% ethanol and then washed using tap water. Finally, safranin was added and allowed to stand for 45s and then washed with tap water. Following this, the slide was examined under a microscope for determination of the morphology, which was confirmed as gram positive.

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S.No.	Properties	Bacillus sphaericus	
1	Genus	Bacillus	
2	Form	Round	
3	Gram reaction	+ ve	
4	Size	3.0 μm × 0.5 μm	
5	Species name	Sphaericus	
6	Collection Acc. No	7542	
7	Growth condition	Aerobic	
8	Sub-culturing period	60 d	

Note: *µm micron meter

Table 7. Inoculation medium details

S.No.	Growth medium additives	Microbe taken for the study <i>Bacillus</i> <i>sphaericus</i>
1	Growth medium Name	Tryptic soy agar
2	Additives 1	Trypticase soy broth 30.0 g
3	Additives 2	Agar 15.0 g
4	Additives 3	Distilled water 1.0 L

Experimental design

The soil sample was prepared in a PVC cylindrical mould of length and width ratio 2 adopted, with a length of 100 mm and a diameter of 50 mm. Jute fibres were sterilized for getting rinsed in denoised water before mixing with soil (Fig 6c). Jute fibres are naturally stiff, have a high tensile strength, and are biodegradable. They are readily available in the local markets. Jute fibres



Fig. 6 a) Isolation of bacterial strain in petri plates, b) calcium carbonate precipitation of bacterial strains, c) Jute fiber

of 0.75% in total soil weight were completely mixed with the soil sample. Optimization of jute fibres length, 1 cm ensured the increase in calcium carbonate precipitation through urease-producing microorganisms (Munshi and Chattoo, 2008). Optimization of jute fiber length was observed based on the literature, which reveals less than 15 mm has maximum efficiency (Imran et al., 2020). Cylindrical mould was prepared by closing the top and bottom and providing inlet and outlet values. Details are shown in Figure 7. The soil was mixed with a bacterial solution and Cementation solution (urea) at optimized concentration of 6.4E+08 CFU mL⁻¹ (Soon et al., 2014). 30 numbers of soil samples were prepared and tested for the current study. In that, 15 samples were prepared with jute fibres and other 15 samples were prepared without jute fibres for calculating the rate of production of calcite during the microbial metabolism. There are 3 numbers of replicates for each curing period (1, 3, 7, 14, and 28 d) respectively. The bacterial solution was prepared and injected under pressure into the soil sample in 50 mL increments. Cementation solution (urea) was also added twice a day at 12-hr interval to the sample. After a curing period of 1, 3, 7, 14, and 28 d, a strength test was performed for the quantification of calcite was observed for both with and without



Fig. 7. Experimental set up

jute fibre, in average of three numbers of replicates. Bacterial development between soil grains was triggered by the jute fibre in the soil (Shao et al., 2014; Lei et al., 2020; Zhao et al., 2020). As a result, it was employed as an environmentally safe method of soil stabilisation to improve microbial precipitation.

RESULTS AND DISCUSSIONS

Raw soil was subjected to an unconfined compression test (UCC), which revealed an unconfined compressive strength (UCS) of 110 kPa. The UCS values of soil samples tested for bacterial solution and cementation solution increases with increase in curing period. Flow of bacterial solution was managed by gravity, and bacteria were fixed in a soil matrix under controlled room temperature conditions. Soil sample were examined for UCC for various curing days. The results are provided in Table 8. Tabulated values show enhancement of microbes enhanced with jute fibre treated soil samples with higher strength. On an average, increase of 1.5 times UCS values was seen, than the samples with microbial solution alone. Over the curing period of 1, 3, 7, 14, 28 days, UCS values of soil treated with microbes with jute fibres were 180, 230, 450, 630, 1520 kPa respectively.

From day one, all the ureased treated samples showed incremental rise in Unconfined Compression values (q_u). There was no discernible change in the soil samples after a 28d curing period. The results of the analysis using a scanning electron microscope are displayed (Fig. 8a and b). *Bacillus sphaericus* demonstrates a rise in UCS values 5 times after 28 d for the microbially treated

 Table 8. Results of unconfined compression values (kPa)

Curing period	Microbe (Bacillus sphaericus)		
Curing period	without jute fiber	with jute fiber	
Day 1	120	180	
Day 3	170	230	
Day 7	240	450	
Day 14	320	630	
Day 28	1250	1520	

sandy soils (Umar et al., 2016). A comparison graph for bacterially treated soil samples with and without jute fibers drawn with data after a 28d curing period is shown in Figure 9. Use of gravimetric analysis with HCL for the determination of calcite secreted throughout the curing phase in the present study (Hejazi et al., 2012). Details are shown in Figure 10. The figures for calcite production of MICP treated samples were 2, 3, 4, 5, 6 percentage for the curing period of 1, 3, 7, 14, 28 days. whereas the figures for soil samples of MICP with jute fibres were 3, 5, 7, 9, 10 percentage for the same curing period. From the results, it was observed that calcite precipitation increased with increase in curing period. Pin hole test was repeated for MICP with Jute fiber soil sample, hole diameter decreased to 1.3 mm and the effluent was cleared for the same optimized condition. In addition, a Crumb test was performed for MICP augmented jute fibre stabilized soil for the determination of the degree of dispersiveness (Chen et al., 2021). Dirt of a small quantity was placed in the water overnight and observation was made. Figure 11 shows no colloidal clouds developed following MICP treatment as a result of reduced soil pH.



Fig. 8. a) SEM image of without jute fiber, b) SEM image of with jute fiber



Fig. 9. UCS of soil sample with and without jute fiber at 28^{th} day



Fig. 10. Calcite quantification for 1, 3, 7, 14, 28 curing day



Fig. 11. Crumb test before and after treatment

CONCLUSIONS

For the sustainable ground improvement elements of this work, dispersed soil was treated using a microbial driven calcium carbonate process and coupled with jute fibre utilizing Bacillus sphaericus. With traditional approaches, stabilisation of dispersive soil would be more difficult than regular soil. Jute fibres are important in the development of calcite in a lump sum, when treated with microbes. The inclusion of jute fibre has a substantial impact on the development of calcite when compared to standard MICP treatment. The UCS value of an untreated dispersive soil was 110 kPa, however after the addition of Bacillus sphaericus, the UCS values were 120, 170, 240, 320, and 1250 kPa at 1, 3, 7, 14, and 28 d of curing period respectively. The experimental findings of microbial treatment with jute fibres revealed that 1520 kPa for 28 d curing period was sufficient. Compared to untreated soil, the unconfined compressive strength of MICP treatment was 11 times higher, and MICP with a 13 times improvement in jute fibres. SEM analysis, has been proved considerable increase in calcite precipitation for MICP with jute fibre samples. Gravimetric analysis was used for the determination of the quantity of calcite precipitated during the microbiological process. The proportion of calcite measured during MICP treatment was determined as 6%. The addition of jute fibre to the soil, together with a microbial treatment, resulted in a 10% calcite precipitation. Addition of jute fibres at the end of the 28d period resulted in a 4% increase in calcite.

Cation Exchange Capacity (CEC) of MICP treated soil was 18 meq/100g and CEC for MICP with jute fibre treated soil was found as 14. It was observed that there was a 36% and 28% reduction, after the MICP and MICP with jute fibres treatment respectively. These values indicate that predominant minerals, responsible for the dispersiveness were modified in the MICP treatment. CEC is also directly influencing the pH of the soil, when CEC decreases, pH value of the soil also decreases. Exchangeable sodium percentage (ESP) of MICP treated and jute fibre bio treated soil was 6. ESP result reveals that bio treated soil reduces the excessive amount of ESP, which improves the bonding between the soil grains. Due to the metabolism of microbes, calcite precipitate (calcium source) in the soil voids which makes the strong cementation bonding between the surface of the soil. Sodium absorption ratio result was improved through the calcium ions after bio treatment with and without jute fibres and it was found as 4.5. Pin hole test was also repeated for the MICP treated and MICP with jute treated soil specimen at same optimized condition of 1.3 ml·s⁻¹ under 50 mm head. The size of the puncture does not show any notable increase after the experimentation. Pin hole test results reveals that decrease in the degree of dispersiveness after the MICP treatment and MICP with jute fibre treatment. The ESP and SAR tests for MICP soil were repeated, and the results given in table 5 and 6, indicating range was in non-dispersive nature. The crumb test confirmed a reduction in the degree of soil dispersiveness. Calcite precipitation in the soil matrix also showed microbial abundance as higher in jute-treated soil samples. Variation in the length of jute fibre and the concentration of bacteria would be made in future experiments.

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Pa Suriya (Research Scholar) conducted all the experiments and wrote the manuscript. Dr. S.P. Sangeetha (Professor) guided in conducting the experiments and helped in revised the manuscript.

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