Assessments of Soil Properties by Using Bacterial Culture.

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ABSTRACT

In recent years high rapid development of infrastructures in metro cities of useful land and compelled the engineers to improve the properties of soil to be the load transferred by the infrastructure, ex: Buildings,bridges, roadways etc. The soil improvement is continuously increasing using different methods to improve the mechanical properties of different type of soil, such as black cotton, red alluvial, murum and sand. The methods of treating soil with chemical and cement grout are used widely in geotechnical projects. The chemical and cement utilized alter the subsurface pH level and hinders groundwater flow. To overcome their effect, more sustainable method is the need of the hour. Hence, an attempt has been made to use of microorganisms, nutrients, and biological processes naturally present in subsurface soils to improve the engineering properties of soil in sustainable way. The calcite precipitation was achieved using the microorganism BacillusPasteurii(NCIB8841 or NCIM2477), an aerobic bacterium pervasive in natural soil deposits. The permeabilityand shear strength were conducted in this project these experiments demonstrate that indigenous bacteria can induce significant quantities of calcite precipitation that calcite precipitation can result in measurable changes to geotechnical soil properties. The variable percentage amount of bacteria was flushed through from top to bottom on selected soils and observes for different durations. The shear strengths of microbial soils is increases while permeability is reduces. The primary purpose of microbial soils is to improve its stability, increasing its bearing capacity and reduce settlements and lateral deformations. Bio-clogging &bio-cementation of soils could be used in geotechnical engineering to improve the mechanical properties of soil,these methods can replace the more energy demanding mechanical compaction methods or the expensive and environmentally unfriendly chemical grouting methods. However, to adopt the microbial method effectively, an integration of engineering, microbiological, ecological studies and design consideration are required. The studies on microstructure, full scale model study and long term assessment will be required for the further strengthening the concept of microbial soil.

KEYWORDS: biological processes, boiclogging & biocementation , stabilization, improvement in soil properties

INTRODUCTION

In many regions around the world the mechanical properties of soils are insufficientfor the desired land use in various purposes. Roads and railway tracks undergo settlement and require continuous maintenance. Dikes and slopes can become unstable and slopes, coasts and rivers aresubject to erosion of soils. Stabilization of soil (ground improvement) can be desirable for these applications. Before and during construction, soil stabilization is often applied at or from the surface by using constructive approaches like compaction, installing nails, sheets or piles, or mixing the soil with lime or cement. When stabilization of a soil mass is required deeper in the underground these surface techniques are insufficient and strengthening techniques, like deep mixing, cement or chemical grouting or ground freezing are being used.

Traditional ground improvement methods have several limitations. The action radius is limited to the proximity of the mixing equipment. High pressures are often required to inject the grouts due to their high viscosity or fast hardening time. Freezing is only a temporal solution during construction work. Next to that most of these methods are expensive, require heavy machinery, disturbing urban infrastructure and involve chemicals with significant environmental impact. Finally, these methods significantly reduce the permeability and increases shear strength of soil.

Microbial Induced Calcite Precipitation (MICP)

The natural lithification of sediments, digenesis, occurs as a result of physical, chemical and biological processes. After deposition, sediments are compacted as they are buried beneath successive layers of sediment and cemented by minerals that precipitate from groundwater. In general, these natural transitions from soil to rock are slow processes, but in some cases natural digenesis can be relatively quick. In sandy soils of which the mineral composition mainly comprises calcium carbonate (calcarenites), biochemically induced dissolution and precipitation of calcium carbonate, have led to significant cementation of unconsolidated sand. Water uptake or putrefaction of tree roots is considered to play a major role in this process.

When supplied with suitable substrates, micro-organisms can catalyze chemical reactions in the subsurface resulting in precipitation (or dissolution) of inorganic minerals, which change the mechanical soil properties. This study focuses on microbial induced precipitation of calcium carbonate.

Most of the inorganic carbon on the earth surface is present as layers of limestone of which a significant portion is of biogenic origin. Many organisms can induce the precipitation of calcium carbonate, but not all can be used for ground improvement.

Precipitation of calcium carbonate occurs when a solution is oversaturated, the amount of calcium and carbonate ions in solution exceeds the solubility product, i.e. the solution gets oversaturated.

Microbial induced calcite precipitation (MICP) is very economical method. So, we can use this method for the small project also. Potential geotechnical applications of this method are as follows:

A. To prevent piping and to enhance stability of earth dams and dikes.
B. To construct the reservoirs and ponds.
C. Seepage control.
D. To control erosion of the banks or coastal area.
E. To increase slope and excavation stability.
F. To reduce the liquefaction potential of soil to enhance the stability of dams.

G. To increase the bearing capacity of foundations and to decrease soil expansion potential.
H. To fix leakages of ground water in underground constructions.

MATERIALS AND METHODS

MATERIALS

Microorganism-An isolated bacterial culture of Bacillus pasteurii NCIM2477 was used in this study. The source of these bacteria was NCIMB Ltd which was a microbiology and chemical industry.Soils-Black cotton soil, Red alluvial soil, murum and sandThe soil sample passing through 4.75mm IS sieve and sample collected in indapur local area.Chemicals- Agar, Peptone, NaCl, Yeast extract, beef extract,urea.

METHODS

Preparing of urea nutrient agar

This media used for the cultivation of Bacillus pantothenticus. Table (1) and Table (2) showthe Solid contents and liquid contents of this media respectively.

Method of Preparation

Components shown in table 1 to distilled water and bring the volume to 1L. Mix well. Gently heat and bring to boiling. Autoclave for 15mins at 15psi pressure -121oC. Cool to 50-55oC.Aseptically add 50mL of sterile urea solution table 2. Mix well. Immediately after that, urea nutrient agar plate medium was poured into approximately 20 culture plates under laminar flow. The laminar flow hood provided filtered air in order to reduce the risk ofcontaminating the culture growth media prior to introduction of the B.pasteurii cultures.Once the poured plates were solidified (after approximately 15 minutes under the laminar flow hood).

Table 1 shows the details of solid contents of the media

Composition	Quantity
Agar	15gm
Peptone	5gm
Nacl	5gm
Beef extract	2gm

Table 2 shown the details of liquid contents of the media

Composition	Quantity
Urea solution	50ml

* urea solution (Add urea to distilled water and bring the volume to 100mL. Mix well. Filter sterilize.)

Making Bacteria Culture

One loopful of pure colony was taken from the stock bacteria and streaked onto each plate, then all plates were then inverted and incubated for 48-72 hours at 30°C in a incubator, once colony growth of B. pasteurii had occurred.

The preparing for calcium carbonate precipitation agar

For culturing the bacilluspasteurii and test its ability for calcium carbonate precipitation, a special agar was created and known as a precipitation agar, which contain agar for made the medium as a semi solid, urea medium and CaCl2.2H2O. The following components table 3 were mixed to create the urea medium solution.The mixed thoroughly in 500 mL of distilled water until they dissolved. The pH of the resulting urea medium solution was

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adjusted to 6.0 using with 5 N HCl prior to autoclaving.

Distilled water was then added to reach the final required volume (1 L) and autoclaving for 15mins at15psi pressure -121oC, the pH of the urea medium was measured to be 7.0 and stirring the solution to aerate it. A 20 ml volume of calcium chloride solution (18.5 g $CaCl_2$/100 mL distilled water) was then added to the aerated urea (1000 mL). Then the resulting solution was poured into culture plates under a laminar flow hood.

Table 3 shows the details of the precipitation agar

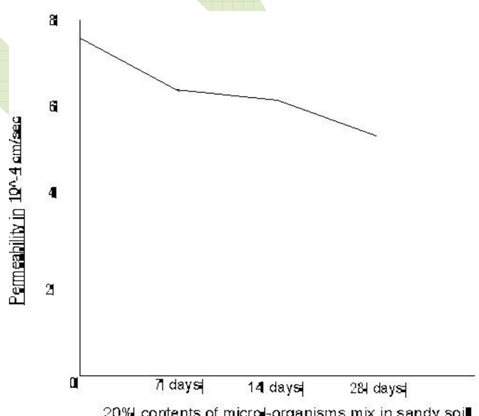
Constituents	Quantity
Agar	7gm
Nutrient broth powder	3gm
Urea	20gm
$(NH_2(CO)NH_2$	10gm
NH_4Cl	2.12gm
$NaHCO_3$	2.12gm
Distilled Water	1 lit
calcium chloride solution/ $CaCl_2.2H_2O$	18.5 g $CaCl_2$/100 mL distilled water

RESULTS AND DISCUSSION

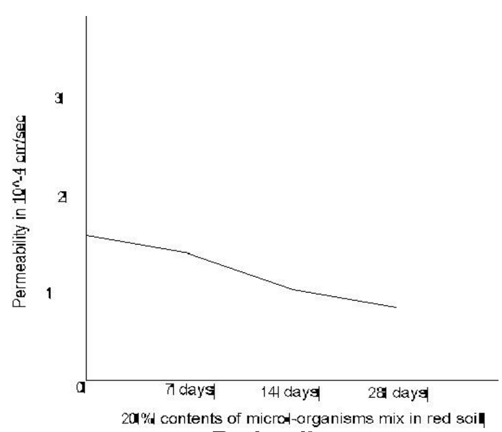
As discussed earlier permeability test, unconfined compression test were conducted in the laboratory. From these laboratory tests D10 size, optimum dose and condition period of microorganisms was determine for the maximum shear stress and minimum permeability, The detail results of permeability tests and direct shear stress tests conducted in laboratory are elaborated in this part.

Permeability test results

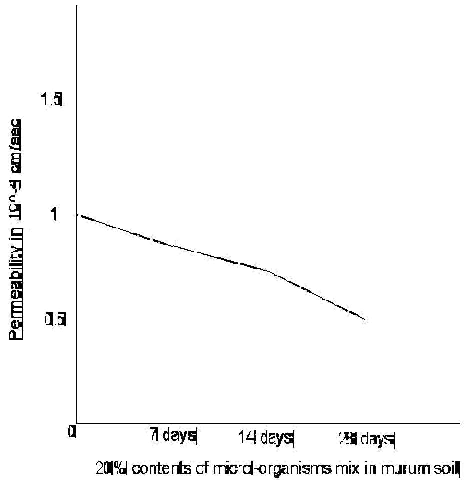
Figure shows the variation of coefficient of permeability for soils of different D10 size. The bacterial solution content is placed on x-axis and coefficient of permeability (K) is on y-axis. The plot also content variation of coefficient of permeability with respect to condition period of 3, 7 and 28 days. The coefficient of permeability observes to be decrease with content of bacterial solution and condition period.



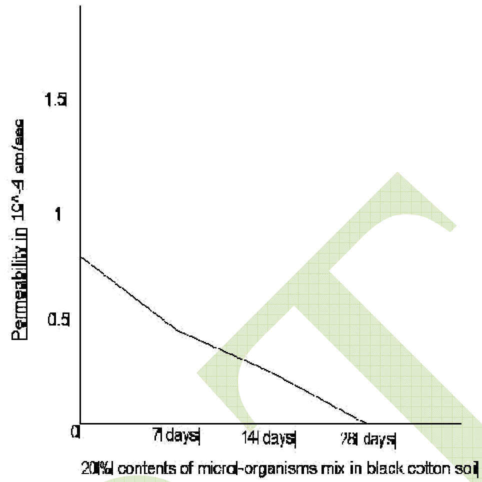
Sandy soil



Red soil



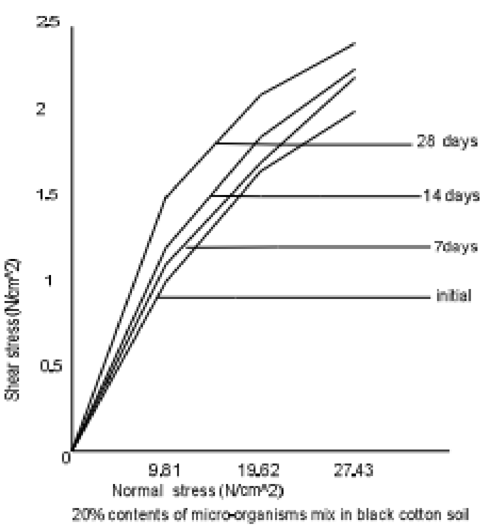
Murum soil

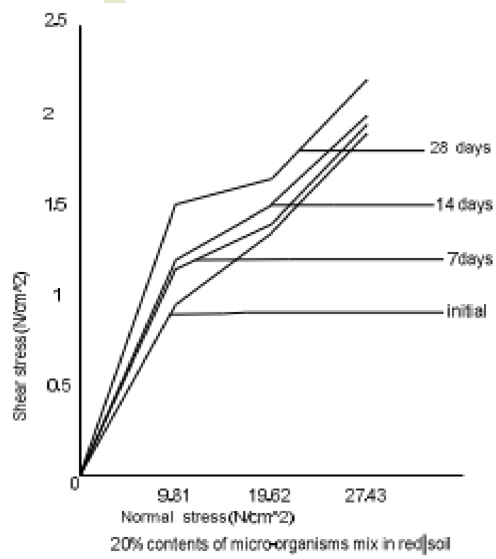


Black cotton soil

RESULTS OF DIRECT SHEAR TEST

The bacterial solution content of normal stress (N/cm^2) is placed on x-axis and shear stress (N/cm^2) is on y-axis. The plot also variation of shear stress with respect to condition period of 7, 14 and 28 days. The coefficient of shear stress is increase with content of normal stress and condition period.





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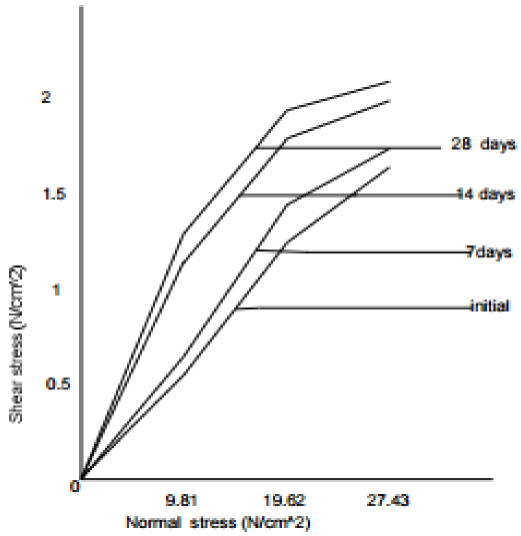
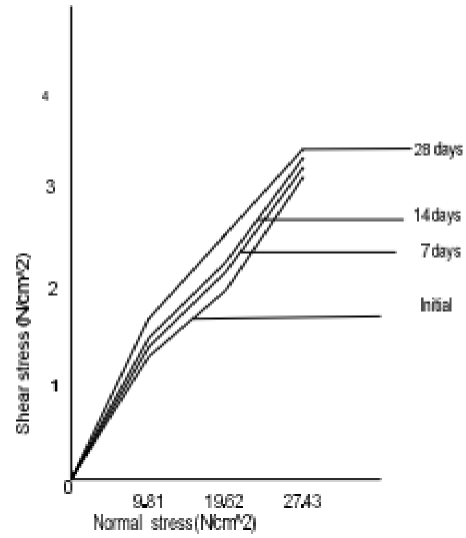


20% contents of micro-organisms mix in sandy soil



20% contents of micro-organisms mix in murum soil

CONCLUSION

The number of experiments has been performed that demonstrate that indigenous bacteria can be used to induce calcite precipitation and substantially increase shear strength and permeability of sands. Testing consisted of a permeability experiment and direct shear tests. The experiment demonstrated that indigenous bacteria could induce sufficient calcite precipitation to significantly modify soil-engineering properties

We believe that the use of indigenous bacteria to precipitate calcite is an important tool for making biomodification of soils more economically feasible. With more experimentation, we expect that uniform distribution of high calcite-precipitation levels can be achieved and implemented in engineering and construction practices.Culturing the Bacillus pasteurii on the precipitation agar shows clearly calcium carbonate forming and the shape of calcite.Forming calcite in the region where bacteria cultured on the agar indicates the role of bacteria in calcite formation and decreasing crystals when increase the distance from culturing region promote that. Urea and agar used as a feed for bacteria.Filling the pores and voids by calcite and covering the particles with cementation process would decrease the porosity and permeability of soil. The permeability of sand and soils reduces with the addition of microorganism in theSand and soils. This may be due to filling of the pores and voids by calcite and coveringthe particles with cementation process.

FUTURE SCOPE

The more development in genetically modified microorganisms will increase in calcium carbonate precipitation and more resistance against temperature, environment changes. Microorganisms selected for genetical modification are selected with respect to region. The present work is more enthusiastic and encouraging. Further study is required to work on microbial induced calcite precipitation with various types of bacteria. The special attention should be given for the practical use. There will be scope in varying depths of microorganism reinforced sand, soils and using no of treatment at the time of preparation of sample. The growth of bond due to microbial treatment need to understand from the point of sustainability and its life.

Applications

The microbial cementation could be used for the following civil and environmental engineering applications:

• Enhancing stability for retaining walls, embankments and dams; Reinforcing or stabilizing soil to facilitate the stability of tunnels or underground constructions.
• Increasing the bearing capacity of piled or non-piled foundations; reducing the liquefaction potential of soil.
• Binding of the dust particles on exposed surfaces to reduce dust levels; increasing the resistance to petroleum borehole degradation during drilling and extraction.
• Increasing the resistance of offshore structures to erosion of sediment within or beneath gravity foundations and pipelines.
• In situ ground improvement.

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