

## Development of Microbial Geotechnology in Singapore

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**ABSTRACT:** Both nature processes and laboratory studies have shown that microorganisms can be used to improve the engineering properties of soil. As such, it is possible to develop methods that utilize the microbial process to treat soil in a way similar to that of cement. When more knowledge is accumulated through research findings and technology development, a new branch of geotechnical engineering – the Microbial Geotechnology can be established. The Microbial Geotechnology can have the following three applications: (a) biocementation to increase the strength of soil, (b) bioclogging to reduce the permeability of soil, and (c) biogas to increase the liquefaction resistance of sandy soil. Some types of microorganisms or bioprocesses that may contribute to the biocementation, bioclogging or biogas effects are identified and discussed. Some experimental data are presented to show that the permeability of sand can be reduced by four orders of magnitude and the strength of sand can be increased to a substantial value after the soil has been treated using bacteria. However, the whole study still stays at the laboratory stage and much more efforts are required to turn this scientific idea into viable technologies.

## INTRODUCTION

Chemical grouting is a common technique used in geotechnical engineering to improve the mechanical properties of soil. In adopting this method, chemical grout is either injected into the voids of soil or mixed mechanically with soil to increase the strength or reduce the permeability of soil. However, chemical grouting is generally expensive and sometimes toxic for environment.

An alternative approach is to use microorganisms to treat soil through a biocementation or bioclogging process (Mitchell and Santamarina, 2005; Ivanov and Chu, 2008; DeJong et al., 2010). Biocementation is a process to bind the soil particles together through agents or slime generated by microbial activities in soil so that the mechanical properties of soil can be improved. Bioclogging is a process to fill the voids, channels or fissions in soil or rock by microbial products to reduce the permeability of soil. Biogas is a process to generate tiny gas bubbles in otherwise saturated soil to increase the liquefaction resistance of sand or to reduce permeability. Due to the small size of bacterial cells and the low viscosity,

bacteria have much better permeating ability than cement and thus are more ideal than cement in terms of construction process. Other major advantages of biocementation, bioclogging, or biogas are that the cost involved can be much reduced and the environmental impact can be much lower (Ivanov and Chu, 2008). However, microbial processes are more complicated than chemical grouting because both microbial and chemical components are involved and its geotechnical application can be soil specific or site environment specific.

The use of microbial technology into geotechnical engineering may lead to the development of a new discipline which has been called Microbial Geotechnology by Ivanov and Chu (2008). The body of knowledge accumulated within this discipline can provide a guide to future research and applications of microbial technologies in geotechnical engineering. The microorganisms that can be used for biocementation, bioclogging or biogas can be called biocement.

There are the following mechanisms that contribute to biocementation, bioclogging or biogas production in soil or rock: 1) filling of the voids and channels in soil with inorganic compounds settled due to activity of microorganisms (Gollapudi et al., 1995; Ramachandran et al., 2001; DeJong et al., 2006; Whiffin et al., 2007; De Muynck et al., 2010); 2) filling in the pores and channels with microbial biomass and polysaccharides (Gerbersdorf et al., 2008; Gonzalez-Munoz, 2010); 3) binding or cementing soil particles with inorganic compounds, which is mediated by microorganisms (DeJong et al., 2006; Whiffin et al., 2007; De Muynck et al., 2010; van Paassen et al., 2010); 4) binding the soil particles with microbial biomass and polysaccharides; 5) forming salt bridges between soil particles and colloids mediated by microorganisms; 6) generating gas bubbles in soil through a microbial process to reduce the liquefaction potential of sandy soil.

Potential geotechnical applications of microbial soil treatment include: 1) to form grout curtains to reduce the migration of pollutants after accidental spill or leakage of toxic pollutant into permeable soil; 2) to prevent piping of earth dams and dikes; 3) to construct reservoirs and ponds in sandy soil; 4) to control seepage or erosion of riverbanks or coastal dikes; 5) to enhance the stability of soil slope; 6) to reduce the liquefaction potential of soil; 7) to increase the bearing capacity of foundations; 8) to fix leakages of groundwater in underground constructions; 9) to control settlement or soil deformation; and 10) to decrease soil expansion potential; 11) to seal the drippings in the tunnels; 12) to fix cracks on rocks; 13) to immobilize sand surface in deserts; 14) to fix soil surface to prevent aerosolization of soil surface pollutants.

## RESULTS

### Biocement selections

A method for the screening of the suitable physiological groups of prokaryotes has been suggested (Ivanov and Chu, 2008). Physiological state of microbial cells

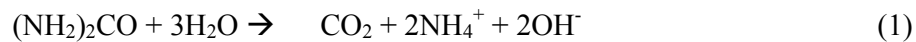
used in biocementation could be monitored using flow cytometry or confocal laser scanning microscopy (Ivanov et al., 2009). Biocements based on the processes shown in Table 1 have been developed and used in laboratory studies.

**Table 1. A set of biological methods used for biocementation, bioclogging, and production of biogas in soil**

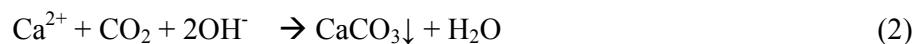
	Bacteria			
	UPB	IRB, UPB, and SRB	AOB or OTB	NRB
<b>Bioprocess</b>	Urea hydrolysis	Simultaneous or sequential ferric reduction, sulfate reduction, urea hydrolysis	Nitrification or oligotrophy	Nitrate reduction
<b>Inorganic components of biocement</b>	Calcium salts and urea	Iron ore/iron salts/magnesium salts and urea	Ammonium salts/organic substance	Nitrate and organic substance
<b>Product</b>	Calcium carbonate	Ferric/ferrous/magnesium hydroxide, carbonate or sulfide	Polysaccharides	Nitrogen gas

The following biocements have been tested to assess their effect of biocementation and bioclogging on sandy soil: 1) Biocements containing calcium chloride, urea, and urease-producing bacteria; and 2) iron-based biocement which was ferrous/ferric-containing solution produced by iron-reducing bacteria from iron ore and organic waste, with an addition of urea and urease-producing bacteria to increase pH.

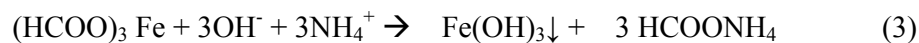
The major chemical reactions related to the common biocements are as follows:



which is performed by urease-producing bacteria, and

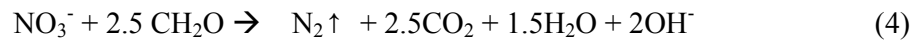


The major chemical reactions in iron-based biocement could be formation and precipitation of ferric/ferrous hydroxides or carbonates during activity of urease-producing bacteria, for example

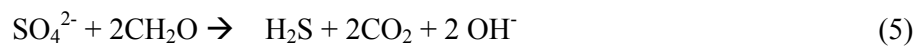


Organic and inorganic salts of ferrous or ferric iron for this reaction can be produced from iron ore and organic electron donor using pure or enriched cultures of iron-reducing bacteria. One source for selection of these bacteria could be anaerobic sludge of municipal wastewater treatment plants (Ivanov et al., 2005; 2009).

Production of hydroxide ions can be performed also by denitrifying bacteria using organic substances as electron donor:

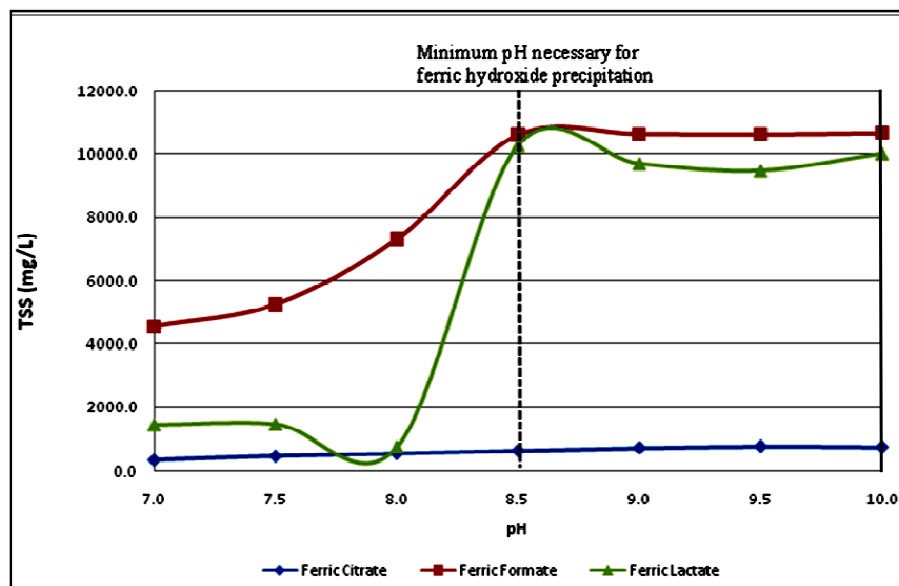


or sulphate-reducing bacteria:



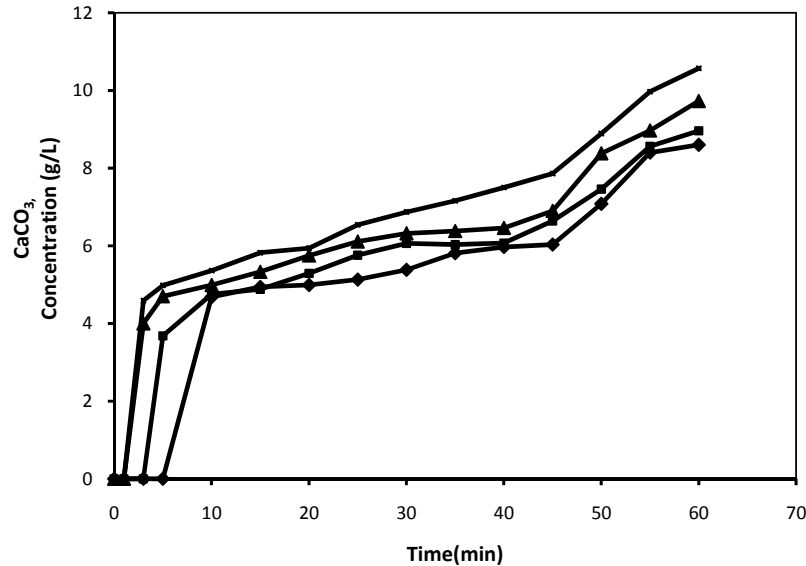
Typical pH that can be created by these processes is from 8.3 to 9.5.

Salts of ferrous/ferric iron with organic acids can be used for biocementation too. Cheap source of organic acids, especially formic and acetic acids, can be reject water of municipal wastewater treatment plants (Ivanov et al., 2005; 2009). Lactic acid can be produced by fermentation of organic carbohydrate wastes. The salts of ferric formate and lactate are suitable for the precipitation at pH above 8.5 (Fig. 1). It means that ferric salts of organic acids produced by fermenting bacteria could be suitable as the components of biocement mixture. Value of pH *in vitro* or *in situ* can be controlled by activity of UPB, NRB, or SRB increasing pH.



**Fig.1. Precipitation of ferric insoluble compounds (total suspended solids, TSS) as affected by pH.**

Depending on the quantity of urease-producing bacteria or activity of urease the rate of precipitation could be faster. For example, calcium-based grout can form precipitate after 10 minutes. This precipitation can continue for several hours as shown in Fig. 2.

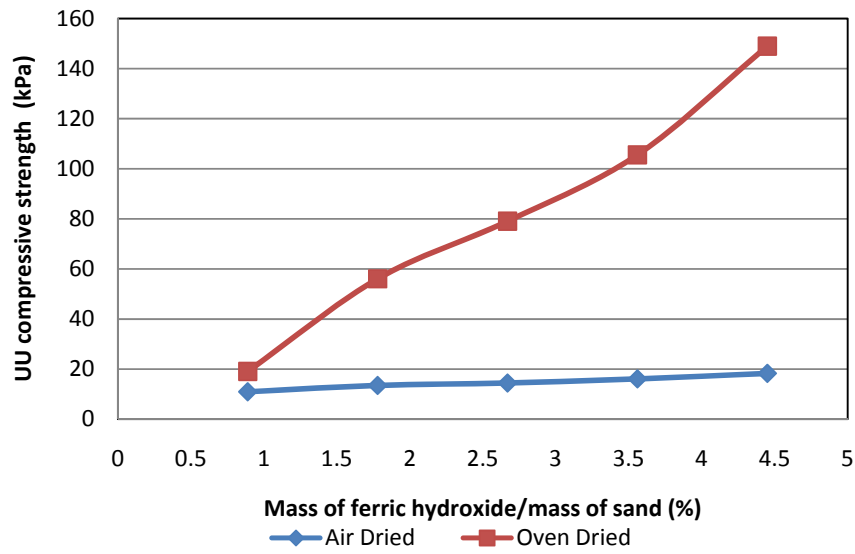


**Fig. 2. Precipitation of CaCO<sub>3</sub> (as Total Suspended Solids, TSS) from the mixture of CaCl<sub>2</sub> and urea with different concentrations of pure urease (pH was changed from 8.5 to 9.1 in 60 min)**

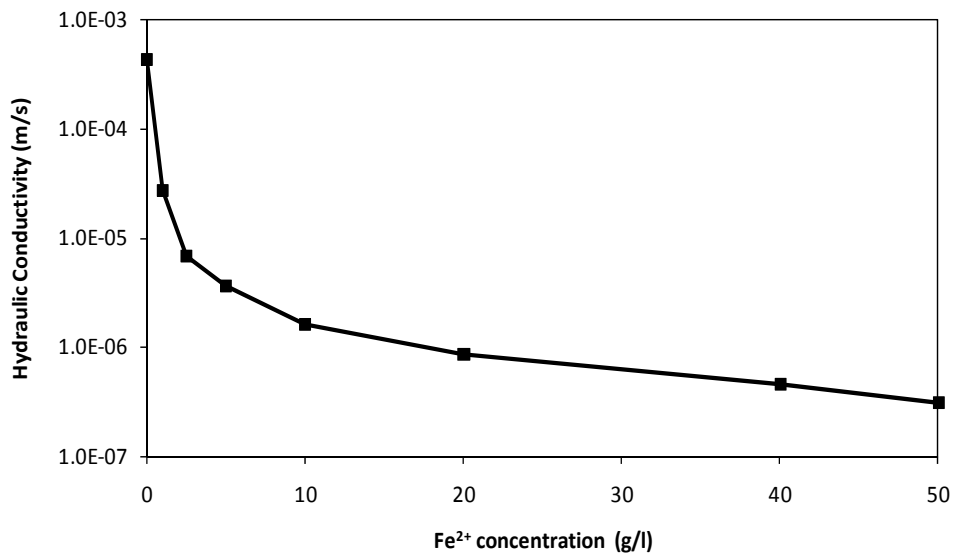
### Sand treated with biocement

The mixtures of UPB suspension, ferrous solution produced by IRB from iron ore, and urea in stoichiometric ratio of urea/Fe as 3:1 was used for several treatments of 200 g of pure sand in a plastic column for 12 h with the drainage of the mixture from the column at the end of each treatment. Constant head permeability tests in a triaxial cell and unconsolidated undrained (UU) triaxial tests were carried out to measure the change in the soil properties of the treated soil. The UU compressive strength of a series of tests on sand treated using Fe compound precipitation are shown in Fig. 3. For air dried samples (25°C), the improvement is insignificant. However, for oven dried samples (60°C), the UU strength increased from 19 to 56 kPa just for two treatments and to 149 kPa after 5 treatments. The permeability of clean sand treated using the Fe compound precipitation method can reduce from an order of 10<sup>-3</sup> m/s to 10<sup>-7</sup> m/s as shown in Fig. 4.

Clean sand treated by calcium salt can stand as a cylinder at dry state and acquire an unconfined compressive strength (UCS) of 1600 kPa as shown in Fig. 5. At wet state, that is, after treatment using liquid form biocement, the maximum UCS attained is 800 kPa (Fig. 5). The permeability of sand has also reduced.



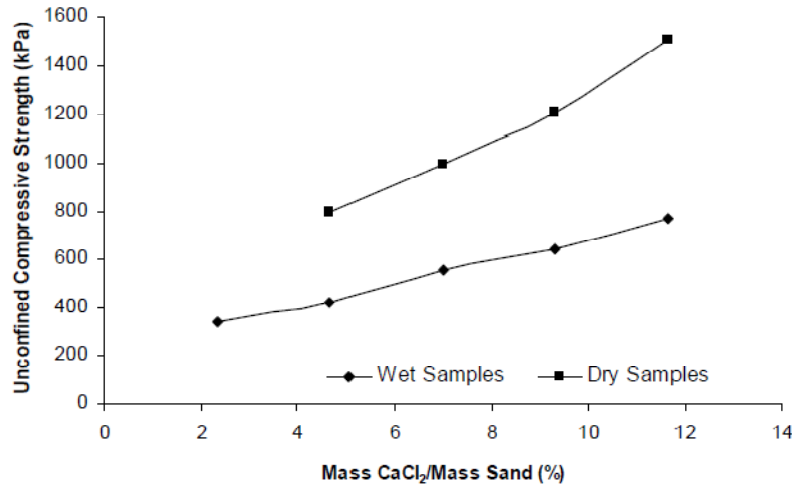
**Fig. 3. The UU compressive strength of both oven dried and air dried sand samples treated using iron-based grout. Points show the values in the sequence from 1 to 5 treatments of the sample.**



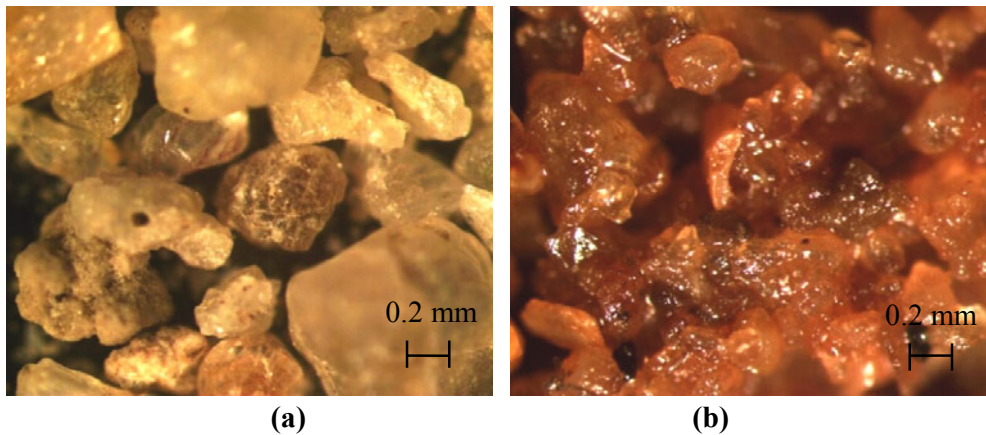
**Fig. 4. Permeability of clean sand treated by biocement.**

## MICROSTRUCTURES

The light micrograph of untreated and biotreated sand samples are shown in Fig. 6. A comparison of the two pictures shows that the sand particles are bound together after it is treated by the iron-based biocement. It can also be seen that the pores are filled by minerals or slimes produced by the microbial process. This may explain partially why biotreatment can increase the shear strength and reduce the permeability of soil.



**Fig. 5.** The unconfined compressive strength (UCS) results of both the wet and air dried sand samples biotreated using calcium-based biocement.



**Fig. 6.** Light microscope photos of (a) Untreated sand and (b) Sand treated with the iron-based biocement

## CONCLUSIONS

It is proposed in this paper to establish a new branch of geotechnical engineering – the Microbial Geotechnology. Two potential applications of Microbial Geotechnology have been discussed: (a) biocementation to increase the strength of soil and (b) bioclogging to reduce the permeability of soil. Some types of microorganisms or bioprocesses that may contribute to the biocementation, and bioclogging effects are identified and discussed. The experimental data presented show that the permeability of sand can be reduced by four orders of magnitude and the strength of sand can be increased to a substantial value after the soil has been treated using bacteria. The present experiments show that the iron-based biocement may be more beneficial to be used for bioclogging purpose. Further study is required to identify potential application of iron-based biocement for biocementation.

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