

# Analyzing the use of molasses as an alternate and optimal culture medium in MICP process of sandy soils

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## ABSTRACT

The use of different soil improvement methods has always been accompanied by an assessment of the affect on strength parameters, costs and environmental impacts. Since the new and eco-friendly methods are associated with a high initial cost, optimizing these methods in order to commercialize them is the priority of research projects. The use of biologic methods for soil improvement, despite its high environmental compatibility, has not been welcomed from the economic point of view in most parts of the world and is still being considered as an academic and not an executive method. Soil improvement, using calcium carbonate sedimentation is one of the most environmental friendly biological methods. One of the most influential bacterial suspension parameters for calcium carbonate treatment is its culture medium, usually Nutrient Broth or Yeast Extract. One of the ways to reduce the cost of biodiversity in the soil, is an alternative culture medium. In this research, the use of sugar beet molasses, which is a waste of sugar and sugar factories, has been investigated as a suitable culture medium for biological improvement along with other culture media. It can reduce the cost of producing a suitable culture medium by up to 500 times. The success of soil regeneration after bacterial cultivation has been also evaluated in the present research.

## KEYWORDS

BIOLOGIC METHODS; SOIL IMPROVEMENT; BACTERIAL SUSPENSION; MOLASSES; SUITABLE CULTURE MEDIUM

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## 1. Introduction

Microbial geotechnology is a new branch of geotechnical engineering, Ivanov and Chu [1] which uses microbiological methods to address geological materials used in engineering. The process of confining inorganic microbial substances between soil particles in the porous medium is very common in the research of Stocks-Fischer, Galinat, and Bang [2]. Hydrolysis of urea is one of the first processes associated with the carbonate precipitation due to the microbial processes which has been investigated in the late nineteenth century Ehrlich [3]. One of the most important factors affecting the activity and biology of microorganisms is their culture medium. The medium is solid, liquid or semi-solid, designed to support the growth of microorganisms or cells [4]. As a bacterial culture medium and one of the most common sources of carbon, yeast extract has been used in many researchs, e.g. Rong, Qian, and Li [5], L. Cheng and Cord-Ruwisch [6] and Burbank, Weaver, Green, Williams and Crawford [7]. Van Paassen et al. [8] improved about 100 cubic meters of soil using yeast extracts. Ammonium and yeast extracts have also been used as a medium Zhao et al. [9] and Feng and Montoya [10]. Carbon-based material, as an alternative to yeast extracts, has been employed as bacterial culture media in the research of Whiffin [11]. Sugar beet molasses as one of the carbon sources, taken from the sugar factories output, is very popular because of its low price [12]. Molasses is also used as a microorganism nutrition source in the bioremediation process Boopathy, Manning, and Kulpa [13] and Fujita et al. [14].

The innovations of this research are summarized as follows:

1. Biological improvement performance of soil with microscopic view based on analysis of the results of the FTIR, XRD, SEM, and EDS tests;
2. Evaluating of strength and geotechnical parameters of improved soil;
3. Possibility of replacing sugar beet molasses (as a carbon source) with chemicals as a medium

## 2. Methodology

The materials used in this study were Firoozkooch sand, *Spoorsarcina Pasteurii* bacterial strain and four culture media including Nutrient Broth, Yeast Extract, Potato Extract and Molasse.

In the present study, the improvement process is based on the adding a suspension of bacteria to the soil, followed by injection of a cementation solution (containing one mole of calcium chloride and urea in

one liter of sterilized and distilled water). This equivalent to the amount of suspension of the bacteria entering the soil in the previous step. The process of entering bacteria into the soil is done in two methods for soil improvement as follows:

- Two-step injection (marked by S)
- Bacteria flocculation mixing (marked by F)

The tests performed include three groups:

1. First group: Classical soil mechanics tests including Uniaxial Compressive Strength (UCS), Direct Shear (DS) and Permeability Test (PER).
2. Second group: physical and chemical tests including X-ray diffraction (XRD), Energy-Dispersive X-Ray Spectroscopy (EDS), Fourier-Transform Infrared Spectroscopy (FTIR) and Wet Chemical Analyses (WCA).
3. Third group: Imaging with Scanning Electron Microscope (SEM)

## 3. Results and Discussion

The use of microbial improvement methods created adhesion in sandy soils that doesn't have inherently adhesion between soils particle. This mentioned by Choi, Chu, Brown, Wang and Wen [15], Salifu, MacLachlan, Iyer, Knapp and Tarantino [16] and Mirmohammad sadeghi, Modarresnia, and Shafiei [17].

The improvement degree of geotechnical parameters after biological improvement depends on various parameters including grain size distribution, temperature, bacterial entry and environmental conditions such as ,the presence or absence of of soil contaminants. Accordingly, the uniaxial strength of the improved specimens by the bacteria, grown in the potato extract medium, does not reach even 5% of ones improved by the bacteria grown in the Yeast Extract medium. On the other hand, samples made from bacteria grown in low cost carbon production source of molasses culture medium have a resistance equivalent to 65% of the resistance of samples made from Yeast Extract medium.

- Permeability test: method F with nutrient broth has the lowest permeability with permeation of  $1.35 \times 10^{-3}$  mm/sec in comparison of the molasse samples which has the permeation of  $2 \times 10^{-3}$  mm/sec.
- XRD analyses: The peaks derived from the interpretation of XRD data taken from Nutrient Broth and Yeast Extract media show 93% and 97.5% precipitation of calcium carbonate and

the residual sodium chloride crystallites respectively. In addition, calcium carbonate precipitation is also observed in the interpretation of precipitation data from sugar beet molasse.

- FT-IR analyses: Estimation and interpretation of precipitant created by the molasses medium, show an absorption bond of  $3143\text{ cm}^{-1}$  and  $3047\text{ cm}^{-1}$  indicating, the tensile vibration of the C-H bond in the aromatic ring. This bond also includes an absorption bond of  $2810\text{ cm}^{-1}$ . The low-intensity vibration in the  $2512\text{ cm}^{-1}$  denotes the carbonate group. There are two narrow absorption bonds at  $712$  and  $874\text{ cm}^{-1}$  in the spectrum, which is a clear indication of the calcium carbonate group presence (the two peaks are related to C-O and C = O bonds). According to mentioned interpretations, calcium carbonate compounds are clearly observable at the FTIR test peaks. Particularly, in the case of precipitant caused by the molasse medium, the presence of calcium carbonate crystallites is confirmed.

#### 4. Conclusions

In summary, using the results of this study, we can hope for better, faster and more effective improvement by mixing bacterial fluccation with soil. Also, despite obtaining better results in geotechnical tests from Nutrient Broth and Yeast Extract media, but due to the much lower economic value of molasses, especially in Iran, this carbon source can reduce the cost of bacterial production to some extent and commercialize the method. Justify microbial improvement and operate on an industrial scale.

#### 5. References

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