学　位　論　文　内　容　の　要　旨

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学　位　論　文　題　名

Exploration and utilization of airborne bacteria for soil improvement using microbial induced carbonate precipitation
(MICP による地盤改良のための空中浮遊菌の探索と利用)

With increasing awareness of energy and environmental crisis, novel technology as one of the countermeasures to the global crisis has been attracting more and more attention in a wide range of research fields. In the realm of geotechnical engineering, the past two decades have witnessed a rapid growth of Microbial Induced Carbonate Precipitation (MICP) technology, which achieves cementation of particles by carbonate from urea hydrolysis catalyzed by microbial urease precipitates at the presence of calcium ions. Despite the tremendous development, there are still some challenges that hinder the progress of upgrading this technology. On the one hand, the issue of cost reduction has received considerable critical attention since cost efficiency is a key determinant of further development of a novel technique. More and more research attempts now are focused on using waste materials as cementation reagents instead of raw materials for treatment. On the other hand, in order to cope with various complicated conditions in scaled-up field applications, the employed bacteria should be improved in terms of survivability and adaptability. To date, many studies have been devoted to addressing these issues to further improve MICP as an eco-friendly technique for the next generation. This work focuses on addressing the challenges faced by bacteria used for biocementation. This dissertation work aims to explore the potential of airborne bacteria as candidates for MICP applications. There are two primary objectives of this study: i) to investigate the occurrence of ureolytic bacteria in air samples collected from different sampling sites and characterize the selected isolates after screening; ii) to further improve the effectiveness of biocementation through applying multiple strains that were selected from air isolates.

Chapter 1 makes a brief introduction of the background, scope, novelty, and importance of this dissertation work.

In Chapter 2, a comprehensive review was performed on current challenges faced by the MICP technique and research trends in bioaerosols. In the end, as an important motivation for this research work, the potential of airborne bacteria as candidates for MICP applications was discussed.

Chapter 3 presents the results from the first stage of this investigation. Airborne bacteria sampled using an air sampler were collected from three main sites with distinct climatic features in Japan, Sapporo, Tsukuba, and Okinawa. Sampling sites with higher vegetation density tend to have a higher population of airborne bacteria. Among tested colonies, 10-20 % of ureolytic bacteria were identified using two culture media. After three rounds of screening, 30 isolates stood out from about 1500 isolates with good performance in precipitating carbonate and were then identified in gene analysis. Most isolates were found belonging to two orders, *Micrococcales* and *Bacillales*. They are from two

of three major phyla that include most culturable bacteria, and many bacteria generalists.

In Chapter 4, a series of characterization tests were conducted on selected strains, including temperature dependence (15-35 ℃) of bacterial growth and bacterial urease activity, pH dependence (pH 4-9), and tolerance to various cementation media. The results revealed that many of these isolates are unique and resilient in many aspects. For instance, one strain showed a cold tolerant feature, and the other strain exhibited relatively high urease activity at acidic conditions. Among characterized strains, two strains with excellent performance were then evaluated in sand column solidification tests, revealing a high efficiency of carbonate precipitation in a two-week treatment compared to many existing studies.

In Chapter 5, at the third stage of this study, the mixing of two strains was evaluated in precipitation tests in order to further improve the effectiveness of cementation towards complicated conditions. Selected strains were mixed in different ratios before applying to cementation media under different conditions. The findings suggest that mixed strains can cope with less favorable conditions for cementation more flexibly than mono strains do, showing a stable and high efficiency of carbonate precipitation in varying situations. In a broad sense, it also indicates that mixed strains might have more advantages than mono strains in the improvement of soils with complex properties.

In Chapter 6, based on the findings on the influence of humic acid (HA) on bacteria and the cementation process, HA was proposed as an amendment to the improvement of biocementation effectiveness. As a bio-stimulant, HA of a small addition to the culture media can significantly increase the bacteria population in a short period, at the same time, increasing the urease activity. With remarkable cation exchange capacity, it can serve as nucleation sites for calcium carbonate precipitation and support the soil matrix as a skeleton. Besides, its buffering capacity can significantly inhibit the emission of ammonia gas during the treatment process. Ideally, this topic would be a fruitful area for further investigation.

Chapter 7 summarizes the major findings of this study and gives application guidelines based on the outcomes. As the first study to explore the potential of airborne bacteria for biocementation applications, the experimental work presented here provides some new insight into the improvement of cementation efficiency through the selection and utilization of various bacteria. Due to time constraints, this work cannot provide a comprehensive understanding of how the proposed method can enhance the cementation efficiency of natural soils. Therefore, further investigations into several topics were recommended. Hopefully, it can contribute to the development of MICP as a real sustainable technology.