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Eco-friendly method of biocementation for soil improvement and environmental remediation in the context of Viet Nam: a state-of-the-art review

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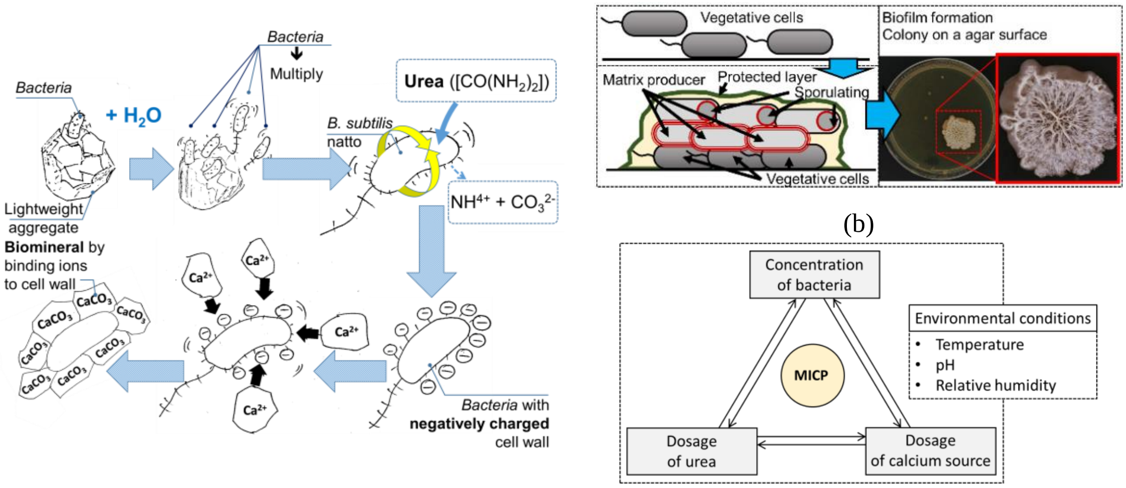
Abstract. Scientists have been using microorganisms to improve soil strength and durability through a process called biocementation. This technique involves inducing mineral precipitation to form a cement-like material that enhances soil properties. Biocementation has been successful in various applications, including soil stabilization, erosion control, and groundwater remediation. Researchers are optimizing nutrient concentrations and ratios to create a favorable environment for microbial activity and promote efficient mineral formation. Different microorganisms have varying abilities to induce mineralization, and cycle treatments have shown promise in stimulating biomineralization processes. Biocementation is a sustainable and eco-friendly technique that can stabilize and immobilize contaminants in soil and groundwater, preventing the spread of pollutants. Despite these challenges, biocementation holds great potential for innovative soil improvement and environmental remediation. Recent studies, experiments, testing devices, and results from research groups in the world and Viet Nam in recent years were reviewed to gain insight into this promising approach. Ongoing research aims to develop cost-efficient and sustainable methods for large-scale production and application of biocementing agents. Further research is needed to uncover the intricate mechanisms and identify optimal strategies for applications and environmental conditions.

Keywords: biocementation, microorganisms, sustainable, stabilization.

Classification numbers: 2.7.2, 3.4.3, 3.7.2

1. INTRODUCTION

This comprehensive paper overviews current and potential future biomineralization technologies, explicitly focusing on microbial induced calcium carbonate precipitation (MICP). MICP is a biochemical process facilitated by urease enzyme bacteria that generate carbonate ions (CO_3^{2-}) and calcium ions (Ca^{2+}) within a nutrient solution. Various factors influence precipitation reactions, including the type of bacteria with urease activity, cell concentration, pH, temperature, reaction period, and nutrient source. In recent years, there has been growing interest in the study of MICP as a sustainable and cost-effective alternative method for concrete crack repair [1-10]. *Bacillus* strains such as *Bacillus cohnii*, *Bacillus pasteurii*, *Bacillus pseudofirmus*, and *Bacillus subtilis*, etc. have been extensively studied and utilized in MICP applications [11-13]. Optimal nutrient sources, such as urea and calcium concentrations, should be carefully considered to achieve desired outcomes while minimizing costs. The resulting calcium carbonate products from MICP exhibit different morphological forms, including calcite, aragonite, and vaterite, renowned for their exceptional stiffness and durability in harsh conditions.



(a) (b) (c)

Figure 1. (a) Mechanism of biomineralization by MICP. Adapted with modification from [7].
(b) Mechanism of bacterial biofilms formation. Adapted with modification from [14].
(c) The relation between three main components for MICP.

Based on the concept and research findings, MICP was explored for self-healing concrete and soil improvement and stabilization purposes [15 - 21]. MICP is a promising way to repair cracks and make concrete structures more durable. By using microorganisms that can create calcium carbonate, damaged areas can be healed, restoring the structure of the concrete. MICP can also help improve and strengthen the soil. By introducing bacteria that can create calcium carbonate, soil erosion can be reduced, and the soil's load-bearing capacity can be increased. This method is eco-friendly and sustainable. However, this approach has some limitations, such as the need for many injection pipes to ensure consistent performance in uneven soil and the potential for clogging near the nozzle hole. Despite these limitations, MICP holds promise as a bio-inspired way to improve and stabilize soil sustainably. Biomineralization technologies are divided into aboveground construction materials, near-ground-surface stabilization, and subsurface applications. These technologies have specific requirements for implementation. Biomineralization helps create innovative building materials and remediate existing structures [22, 23]. It can also be used for soil and dust stabilization, offering alternatives to traditional methods. Biomineralization can quickly fill cracks in self-healing concrete while building materials like bricks require rapid formation of robust bonds. Although biomineralization technologies may be more expensive, they are reliable and easy to implement. Ureolytic biocementation is a promising greener alternative with lower carbon emissions [24 - 26]. Ureolysis-induced calcium carbonate precipitation is the most widely employed approach for large-scale applications.

Figure 1 offers a detailed illustration of the biomineralization mechanisms involved in MICP, as discovered in a study conducted with *Bacillus subtilis*. The figure outlines each stage of the biomineralization process, from urea hydrolysis to forming $CaCO_3$ crystals, providing a valuable visual representation of this intricate process. Additionally, the figure highlights the stages of bacterial biofilm formation by *Bacillus subtilis*, showcasing the attachment, colonization, growth, and maturation phases. This depiction offers a better understanding of these phenomena and how they are explicitly observed in *Bacillus subtilis*. *Bacillus subtilis* and other bacteria can create biofilms, complex communities of bacteria embedded in a self-produced matrix. The bacteria attach to a surface initially, forming a monolayer, then multiplying and secrete, which helps form microcolonies. As the biofilm matures, mushroom-like structures can develop, providing structural stability and protection. The matrix also facilitates communication between the bacterial cells within the biofilm. Not only does precipitate $CaCO_3$ help increase the density of separated particles, but bacterial biofilm also plays a role in cementation [27 - 29].

As shown on Figure 1c, concentration of bacteria plays a significant role in the biomineralization process, with higher concentrations requiring increased doses of urea and Ca^{2+}. However, adding a bacterial nutrient such as yeast extract or glucose can enhance precipitation even at low initial bacterial concentrations by stimulating bacterial growth [8] and increasing bacterial concentration. The urease activity test results indicate that the presence of this nutrient can impact the activation and strength of bacterial activity, potentially causing a delay in urea decomposition when bacteria are present at low concentrations. Enhancing biomineralization's capacity and effectiveness through MICP is beneficial for improving biocementation. This process, in turn, can facilitate the upscaling of bacterial treatment for larger quantities of sand or soil. However, it is essential to consider the cost implications associated with increasing the effectiveness of MICP. As the effectiveness increases, the costs may also rise. Therefore, optimizing the bacterial concentration and nutrient sources is crucial to balance efficacy and cost-effectiveness. Two valuable suggestions to address this concern are using wasted or recycled nutrient sources and removing unnecessary organic nutrient sources, focusing primarily on urea and calcium sources. By using wasted or recycled nutrient sources, we can minimize costs by repurposing materials that would otherwise go to waste. Additionally, eliminating organic nutrient sources and focusing on essential nutrients can simplify the process and potentially reduce expenses associated with unnecessary additives. Various types, such as lactose, glucose, corn starch, tapioca, and soybean meal, could be used to grow and biomineralize the bacteria. The optimum concentration of this component is from 2 to 20 g/L in the bacterial solution, with the range of the bacterial spore from 4.28×10^8 to 8.05×10^9 spores/mL. As mentioned before, to maintain the properties of concrete, the concentration of organic carbon sources should be controlled carefully. There needs to be more than a small amount of this component for the biomineralization quickly. The hash condition could slow down or delay both the spore-cell transform and the mineral-forming process. Unlike harsh conditions in self-healing concrete, MICP for soil/sand stabilization through biomineralization can be obtained with less challenge. However, because of the sizeable porous volume among the separated materials (sand grains, soil particles), a large amount of precipitated products is necessary to fill out and create a high cementation level.

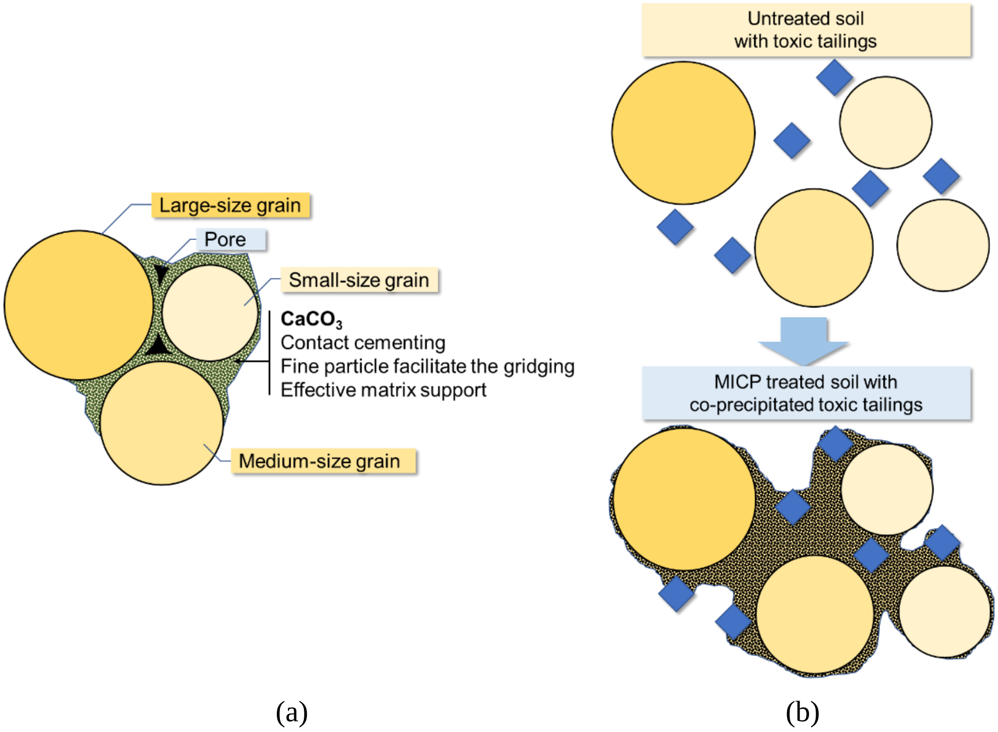


Figure 2. (a) Biocementation of separated grains through MICP. (b) Biocementation process: Coprecipitation of toxic metals from the solution, simultaneously enhancing sorption capacity through increased surface area.

Remediating mine tailings is a promising application of biomineralization (Figure 2), considering the inherent risks associated with their collapse and the release of toxic metals such as Cu, Pb, Zn, Cd, Cr, Se, and As, which pose hazards to human and environmental health [30-32]. The leaching of heavy metals into surrounding water bodies through precipitation events amplifies these concerns. Practical strategies for reducing metal leaching rates involve immobilizing or shielding the metals from water exposure. Traditional methods such as phytoremediation, thermal treatments, excavation, electro-reclamation, and capping have been explored. However, the biomineralization of heavy metal-loaded soils or mine tailings has emerged as a viable treatment option due to the stability exhibited by the resulting MICP products in various geological settings. Studies have demonstrated the significant potential of MICP as a sustainable and efficient approach for remediating water contaminated with heavy metals. MICP remediation has proven effective even at elevated metal concentrations surpassing estimated toxicity thresholds, as long as a sufficient proportion of the initial microbial population survives to initiate urea hydrolysis and induce calcium carbonate precipitation. In a study conducted in 2013, the remediation of Cr(VI) from chromium slag was investigated using biocementation with the ureolytic bacterium *Bacillus sp.* CS8 [33]. Bacterial cells were used to prepare a consolidated structure resembling bricks from chromium slags, and a sequential extraction was performed to analyze the distribution of Cr(VI). The results showed a significant decrease in Cr(VI) mobility in the exchangeable fraction of the slag, with a subsequent increase in Cr(VI) concentration in the carbonated fraction after bioremediation. After bioremediation, the bricks developed high compressive strength and low permeability, and column tests revealed a remarkable decrease in Cr(VI) concentration in the leachate. Scanning electron microscopy-energy-dispersive spectroscopy (SEM-EDS) analysis confirmed the MICP process in the bioremediation of the Cr slag, with the incorporation of Cr(VI) into the calcite surface forming a solid complex that hinders its release into the environment.

In the context of environmental remediation, biocementation of hazardous waste has been explored to reduce the mobility of contaminants. However, there need to be more comprehensive studies evaluating its efficacy. A study assessed the physicochemical factors influencing the stabilization of hazardous products through *in-situ* MICP [34]. *Pararhodobacter sp.* was used to investigate the Pb-contaminated kiln slag (KS) and leach plant residue (LPR) collected from Kabwe, Zambia. Biocemented KS and KS/LPR exhibited leachate Pb concentrations below the detection limit, resistance to slaking, and maximum unconfined compressive strengths of 8 MPa for KS and 4 MPa for KS/LPR. These findings highlight the efficacy of biocementation in stabilizing hazardous waste and suggest its potential for environmental remediation applications. In another study, MICP was employed for the removal of ammonium by-products from the effluent of a biocementation system through struvite precipitation [35]. Urea hydrolysis by the supplied microorganisms resulted in calcium carbonate formation, cemented the soil particles, and improved the matrix strength. However, the production of ammonium during the process posed a challenge for field-scale applications. A two-stage study was conducted to address this matter. The first stage focused on optimizing the rinsing conditions for ammonium removal from the soil. In contrast, the second stage evaluated the influence of pH conditions, molar ratio, and calcium ions on struvite precipitation. The study demonstrated that the struvite precipitation technique could remove approximately 90 % of ammonium from the effluent, with a molar ratio of NH^{4+}:Mg^{2+}:PO_4^{3-} of 1:1.2:1, providing favorable conditions for effective removal.

2. BIOCEMENTATION METHOD AND EXPERIMENTAL STUDIES

2.1. Biocementation method

2.1.1. Biochemical processes involved in biocementation

According to Journals and Books from Nature Communications, Elsevier publisher, Springer publisher, and MDPI publishers, biocementation and biologging are natural soil stabilizing and strengthening processes when bacteria-induced calcite precipitation in soil [36-43]. Biocementation refers to how living organisms, such as bacteria, algae, and fungi, are utilized to precipitate minerals and create cement-like materials. The biochemical processes involved in biocementation vary depending on the specific organism and the environmental conditions. However, some standard biochemical processes occur during biocementation: Microbial Metabolism, Carbonate Precipitation, Extracellular Polymeric Substances (EPS), pH Regulation, Biofilm Formation. Bacteria play a crucial role in biocementation. Certain species of bacteria, such as *Sporosarcina pasteurii*, produce an enzyme called urease. Urease catalyzes the hydrolysis of urea, a compound found in many organic materials, to produce carbonate ions (CO_3^{2-}) and ammonium (NH^{4+}). When carbonate ions come into contact with calcium ions (Ca^{2+}), calcium carbonate is formed. This mineral is crucial for biocementation. During the process, the produced carbonate ions react with calcium ions, resulting in the precipitation of calcium carbonate. The precipitation process involves the creation of calcium carbonate crystals, their growth, and the subsequent aggregation of crystals to form a solid cementitious matrix. Bacteria involved in biocementation often produce EPS, which act as a glue-like substance that helps bind the mineral precipitates together. This contributes to the strength and stability of the resulting biocement. Bacteria also regulate the pH of their surrounding environment during biocementation [44-46], which is crucial for controlling the biocementation rate and preventing the formed minerals from dissolving. Biofilms, which are structured communities of cells embedded in a matrix of EPS, provide a favorable microenvironment for biocementation. Biofilm formation enhances the effectiveness and efficiency of biocementation processes.

2.1.2. Factors influencing on biocementation process

Microorganisms are used for biocementation reactions, but their effectiveness depends on many factors. The type of microorganism used, environmental conditions like temperature and nutrient availability, and substrate composition are all important. Time is also a factor, as is the proper mixing and distribution of microorganisms and metabolites. Inhibition factors like chemicals or heavy metals can hinder the process. By understanding and controlling these factors, biocementation processes can be optimized for desired strength and durability.

Firstly, selecting a suitable microorganism is crucial. Different species or strains have different abilities and preferences for mineral precipitation. Ureolytic bacteria like *Sporosarcina pasteurii* are often used due to their ability to produce enzymes like urease that help with mineral precipitation. The choice of microorganism should be based on its compatibility with the target substrate and desired outcomes. Temperature, pH, moisture levels, and nutrient availability also play a significant role in the effectiveness of biocementation. Understanding the specific requirements and tolerances of the selected microorganisms is essential for creating suitable environmental conditions. The composition of the substrate being treated is another critical factor, as it affects the potential for biocementation. Time, proper mixing, and distribution of microorganisms are also essential considerations for uniform and effective biocementation. However, inhibition factors like chemicals or heavy metals can impede microbial activity and mineral precipitation. Researchers and engineers can optimize biocementation processes for various applications by considering and controlling these factors. Ongoing research and advancements in biocementation techniques continue to improve our understanding and ability to harness the potential of microorganisms for sustainable geotechnical and construction applications.

2.2. Outline of research problematic

As described in Figure 3, there are a variety of applications for biomineralization technologies. Some are already being used today, while others are still developing. Regardless, the potential for these technologies is vast and exciting. From improving medical treatments to creating new building materials, biomineralization can change our world in many positive ways. In building and construction materials, there is a growing interest in using biomineralization to create innovative materials that can remediate existing structures and provide self-healing properties. The stabilization applications section also explores the potential of microorganisms and enzymes to stabilize and immobilize soil, dust, and toxic mine tailings, providing a more sustainable alternative to traditional methods. Finally, the engineering applications section showcases the latest developments in biomineralization technologies for more profound subsurface applications, such as leaky well sealing, which have already reached commercial sector applications. Applying MICP using bacteria has shown promise in improving soil strength, reducing permeability, and enhancing load-bearing capacity. While biocementation research and development are globally widespread, Viet Nam has actively pursued studies and applications in this field. Vietnamese researchers have investigated local microbial strains and their efficacy in biocementation processes, considering the unique environmental conditions and challenges specific to the region. This local focus contributes to advancing biocementation knowledge and its application in Viet Nam.

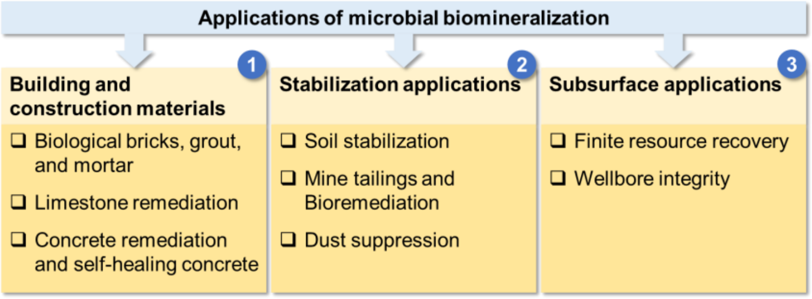


Figure 3. Technologically advanced applications of microbial biomineralization: (1) Building and construction materials, (2) Ground stabilization applications, and (3) Subsurface applications.

Table 1. Current research problematic in the field of biocementation [47 - 52].

Research Problematic	Content	Situation	On going
Limited understanding of microbial mechanisms and interactions	-Lack of comprehensive knowledge about the specific metabolic pathways and enzymes involved in biocementation. -Insufficient understanding of microbial ecology and community dynamics during biocementation. -Limited knowledge about the effects of different microbial species and strains on biocementation efficiency and effectiveness.	Progress has been made in identifying and characterizing key microbial species and their metabolic pathways in biocementation.	Further research is still needed to fully understand the complexity of microbial interactions, community dynamics, and their response to varying environmental conditions.
Lack of standardization and optimization	-Inconsistencies in experimental setups and methodologies make comparing and replicating research findings challenging. -Limited standardized protocols for evaluating and quantifying the performance and durability of biocemented materials. -Insufficient optimization of parameters such as microbial concentrations, nutrient availability, environmental conditions, and substrate composition for achieving optimal biocementation outcomes.	Efforts have been made to develop standardized protocols and experimental procedures for evaluating biocementation processes.	More work is needed to establish widely accepted standards and optimize parameters for consistent and reliable biocementation outcomes.

Limited field-scale application and scalability	-Few studies have focused on successfully translating laboratory-scale biocementation to field-scale applications. -Challenges in upscaling biocementation processes while maintaining effectiveness and cost-efficiency. -Limited understanding of the long-term performance, durability, and stability of biocemented materials under real-world conditions.	Some successful field-scale applications of biocementation techniques have been reported, demonstrating the potential for practical implementation.	Challenges remain regarding upscaling from laboratory to field conditions, ensuring long-term performance and durability, and achieving cost-effective large-scale implementation.
Environmental considerations and ecological impact	-Potential risks associated with the introduction of non-native microorganisms into natural ecosystems. -Limited understanding of the ecological consequences of microbial activities and mineral precipitation in different environments. -Lack of comprehensive assessment of the environmental sustainability and ecological impact of biocementation techniques.	There is increased awareness of the need to assess and minimize potential ecological risks associated with the use of non-native microorganisms.	Further research is required to better understand the long-term ecological consequences and develop sustainable biocementation practices.
Economic feasibility and commercial viability	-Limited cost-effectiveness and high production costs of microbial cultures and nutrient sources for large-scale biocementation. -Challenges in integrating biocementation techniques into existing construction practices and standards. -Limited understanding of the economic viability and long-term benefits of implementing biocementation in various industries.	Advances have been made in exploring cost-effective materials and optimizing biocementation processes to improve economic feasibility.	More work is needed to fully evaluate the long-term economic benefits and establish the commercial viability of biocementation techniques.

Table 1 presents issues that require attention in the field of biocementation. The research problem can be succinctly summarized as a need for more standardization in experimental protocols, inconsistent results and data interpretation, and a limited understanding of the underlying mechanisms governing biocementation. These challenges pose significant obstacles to the development of biocementation technologies. Therefore, further research in this field is necessary to overcome these challenges and to advance biocementation technologies.

2.3. Experimental studies

In lab experiments for biocementation, researchers can closely control conditions and work on a small (from 10 cm in height to 100 m^3) volume of sand samples. Nevertheless, what works in the lab may only work on a small scale.

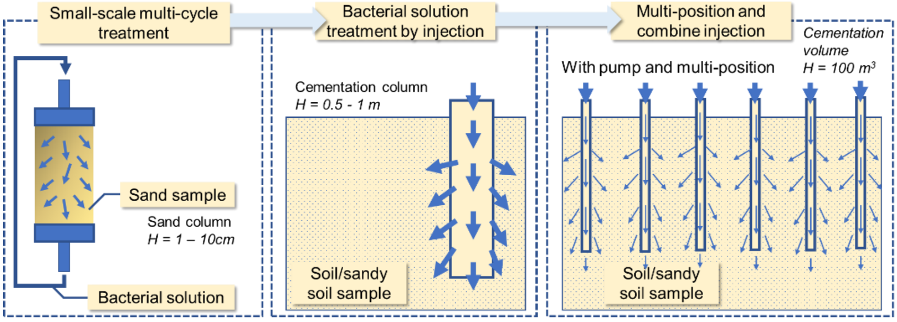


Figure 4. Evolution of biocementation treatment method from lab-scale to large scale experiments.

There are many factors to consider, such as the type and amount of microorganisms, nutrient availability, temperature, pH levels, and the characteristics of the material being treated. Scaling up the process may take longer and cost more. It is essential to consider the time microorganisms take to make minerals and achieve the correct strength. Developing cost-effective ways to produce and use microorganisms and nutrients is also essential. Large-scale projects need to be compatible with existing construction techniques and materials (Figure 4). The design should also consider changes to the material from biocementation. The environmental impact should be considered, and any risks from using microorganisms and nutrients should be evaluated. The material should be stable and sustainable for the long term.

2.3.1. Laboratory-scale studies on biocementation using different microorganisms

Laboratory-scale studies have been conducted to assess the biocementation potential of various microorganisms. These investigations aim to evaluate different species in terms of their ability to precipitate minerals and enhance the strength and durability of cementitious materials. Among the microorganisms studied, *Sporosarcina pasteurii* has been commonly used due to its urease production, which generates carbonate ions for calcium carbonate precipitation. *Bacillus spp.* have also been explored, as they produce urease and exopolysaccharides, contributing to biocementation.

Similarly, *Sporosarcina ureae* has shown promise in improving the mechanical properties of cement-based materials. *Pseudomonas aeruginosa*, known for its biofilm formation, has been studied, highlighting enhanced strength and reduced permeability. Additionally, with their organic acid secretion and enzyme production, fungi have demonstrated potential in biocementation, showing improved compressive strength and reduced water absorption. These laboratory-scale studies provide valuable insights into the biocementation capabilities of microorganisms, aiding in process optimization and potential applications in construction and environmental engineering. Scientists from Hokkaido University [32] studied how to strengthen the soil on slopes in a cold area of Japan. They found a new type of bacteria that can help make the soil stronger by causing a chemical reaction that creates a cement-like substance. The researchers tested how well this technique worked on different types of soil and found that it could be a good way to improve the stability of slopes. They also discovered that the bacteria worked best in colder temperatures and when the soil had more fine particles. The study showed that this technique could be useful for stabilizing slopes, but there are some limitations to consider. Testing was also conducted on sand samples to assess cementation under slope conditions for rain and wind-simulation experiments (Figure 5). The findings indicated a positive impact from using MICP, particularly when accelerated through nano calcite as nuclei to enhance $CaCO_3$ crystallization.

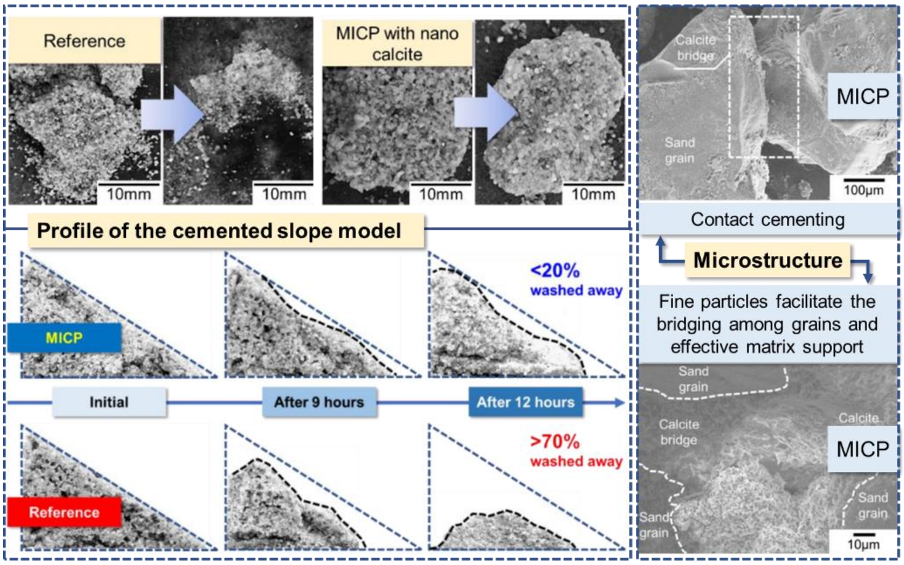


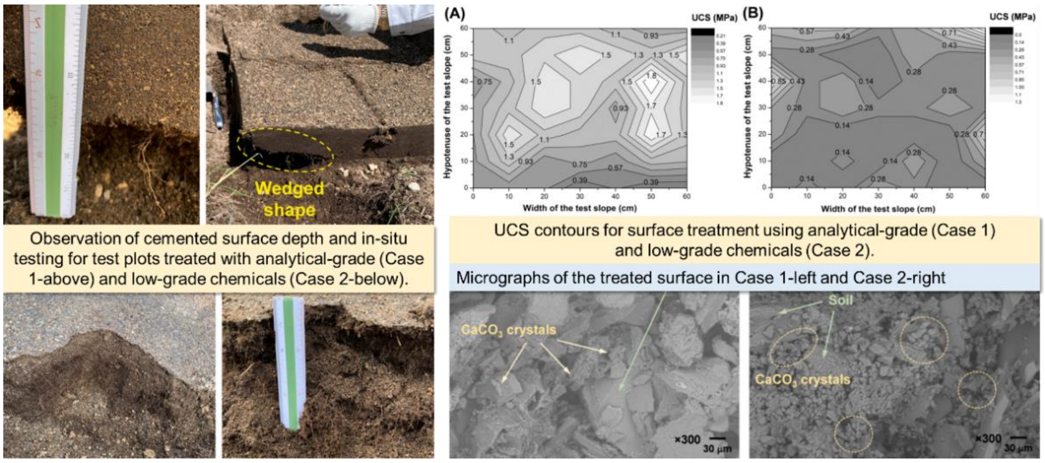
Figure 5. Cementation and outlooking of sand samples after erosion test, microstructure analysis of the sand grains treated by MICP with nano calcite. Adapted with modification from [53].

2.3.2. Field-scale experiment of biocementation for soil improvement and environmental remediation

Biocementation is a helpful technique for improving soil and solving environmental problems. Microorganisms are injected into the soil with a nutrient solution that produces calcium carbonate, which can help strengthen the soil, reduce permeability, and improve load-bearing capacity. Biocementation has been successfully used in civil engineering projects, stabilizing slopes, preventing erosion, and reinforcing foundations. It has also been used for ground improvement, densifying weak or loose soils by binding soil particles. Biocementation can immobilize pollutants and help restore degraded lands by improving soil quality and facilitating vegetation establishment [24, 54 - 56]. It can also protect against erosion and wave impact in sediment or coastal structures. Biocementation is a sustainable solution being explored and improved for practical implementation.

Based on previous studies, in 2023, the research group from Hokkaido University [20], conducted field experimentation of biocementation using low-cost cementation media to preserve slope surfaces. Research showed that using low-grade chemicals for *in-situ* stabilization of slopes through microbial-induced carbonate precipitation is effective. In this project, *Lysinibacillus xylanilyticus*, previously isolated from Hokkaido, Japan Two, was used with low-cost nutrients.

Researchers conducted test plots to study the effectiveness of low-grade chemicals versus analytical-grade media in stabilizing slope surfaces. After 20 days, it was found that using low-grade chemicals, such as snow-melting agents, fertilizer urea, and beer yeast, significantly improved the treated slope surfaces. The cost was also reduced by 97 % compared to using analytical-grade media. This study highlights the potential and cost-effectiveness of using low-grade chemicals in this method. The researchers also discovered that a combination of low-grade chemicals formed a strong layer up to 10 cm deep, with a strength of 1.02 MPa (Figure 6). It is important to note that the purity of these chemicals is crucial, and pre-treating beer yeast can enhance the results. Overall, low-grade chemicals are a viable option for this type of work, and reducing costs is a crucial factor in bridging the gap between lab-scale and real-scale application of the method known as MICP.


Observation of cemented surface depth and in-situ testing for test plots treated with analytical-grade (Case 1-above) and low-grade chemicals (Case 2-below).

UCS contours for surface treatment using analytical-grade (Case 1) and low-grade chemicals (Case 2).

Micrographs of the treated surface in Case 1-left and Case 2-right

Figure 6. Cemented soil surface, compressive strength and microstructure. Adapted with modification from [20].

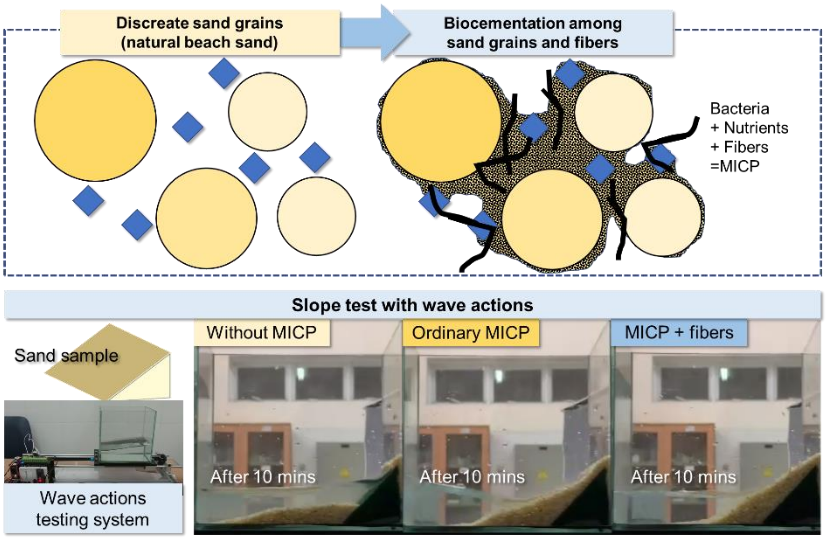


Figure 7. Mechanism of anti erosion effects on sand beach by biocementation through MICP and improved MICP with wave actions test.

MICP is a popular eco-friendly technology in geotechnical and environmental engineering. Also, island microbial technology is a promising way to use MICP to strengthen and protect islands and coastal regions. It produces calcium carbonate, like calcareous sand, and can reinforce islands, as reported in a publication for soil-sand stabilization of the Hokkaido region

[32]. The testing done on sand samples for erosion effect under wave actions testing system showed that using MICP helps maintain the shape and structure of the sand. This effect is even more pronounced when combined with fibers, as shown in Figure 7. These findings suggest that using MICP and fibers could be a good option for preventing erosion in coastal areas. The optimal pH for the mineralization is around 9. Generally, island microbial technology can improve the strength, stiffness, bearing capacity, and erosion resistance of calcareous sand on islands. However, *in-situ* tests are needed to confirm bacterial urease activity's efficiency and calcium carbonate deposition rate in calcareous sand on islands. A reliable numerical model is also crucial for understanding the biochemical processes of MICP and soil reinforcement on islands.

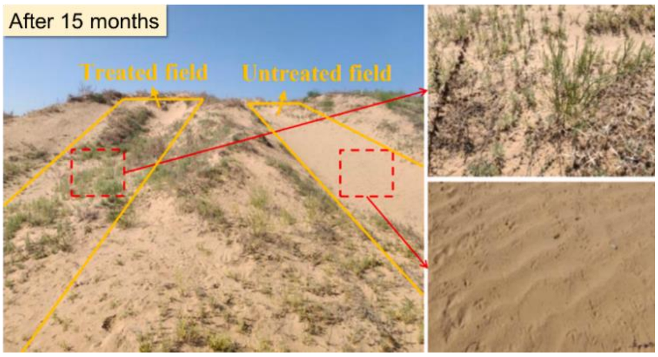


Figure 8. Comparison of the untreated and SICP treated field in Ningxia, China after a 15-month period. Adapted with modification from [57].

Biocementation has emerged as a promising approach for soil stabilization and wind erosion control in geotechnical and construction applications. This process involves using natural biological processes, specifically microbially- or enzyme-induced carbonate precipitation (EICP), to produce cementitious materials known as bio-cement. One cost-effective solution for carbonate precipitation and biocementation is using soybean urease, resulting in a technique known as Soybean-Induced Carbonate Precipitation (SICP) [57, 58]. The calcium carbonate produced through SICP can effectively cement soil particles, significantly improving soil strength (Figure 8). Various strategies have been explored to optimize the biocementation process, including using inexpensive waste materials for bio-cement treatment and bacterial cultivation.

Additionally, modifications to the treatment materials and procedures, such as using lysed cells, low pH conditions, and the salting-out technique, have been investigated. Field studies have demonstrated that biocementation can effectively enhance soil surface strength and resistance to wind erosion. Moreover, the bio-cemented soil ground, even with low-concentration treatments, has shown the ability to support the germination and growth of local plants. These findings highlight the potential of biocementation as a sustainable and environmentally-friendly approach for soil stabilization and erosion control, with potential applications in diverse geotechnical and construction contexts.

2.3.3. Comparative analysis of different biocementation techniques

When selecting a biocementation technique, careful consideration must be given to the advantages and disadvantages associated with each option. Among the available methods are MICP and EICP, which involves the use of enzymes such as urease to catalyze the hydrolysis of urea and induce the precipitation of calcium carbonate in soil. EICP, similar to MICP, offers benefits regarding environmental sustainability and cost-effectiveness. It can be applied to various soil types and has potential applications in geotechnical engineering, construction, and environmental remediation. However, EICP also presents challenges related to enzyme availability, stability, and optimization of reaction conditions. The effectiveness may vary depending on the specific enzymes used and their compatibility with the soil environment.

Compared to MICP, EICP does not rely on the presence and activity of live microorganisms, simplifying the application process and providing greater control over the precipitation process. Nonetheless, further research and development are needed to fully understand and optimize the effectiveness of EICP under different soil conditions and engineering applications. Each approach has its benefits and drawbacks, requiring a clear understanding before deciding. For instance, MICP is cost-effective and compatible with a wide range of soils but exhibits a slow reaction rate and uneven distribution. EICP offers speed and precision but may be costly and challenging to obtain. Biomineralization methods involving algae and fungi should be evaluated, considering their advantages and disadvantages. While these methods may prove beneficial in certain circumstances, achieving consistent control and performance may be challenging. Deposition techniques offer greater flexibility but require careful regulation and may be slower to execute. Ultimately, the choice of the optimal approach will depend on the specific project requirements. It is encouraging to see ongoing scientific exploration aimed at enhancing biocementation methods.

3. REVIEW OF BIOCEMENTATION AND EXPERIMENTAL STUDIES IN VIET NAM

3.1. Analytical techniques for evaluating the effectiveness of biocementation

Analytical techniques play a pivotal role in assessing the efficacy of biocementation processes. These techniques enable researchers to examine and quantify various aspects of the biocementation phenomenon, such as the degree of mineralization, changes in the microstructure, composition analysis, and mechanical properties of the treated materials. Commonly employed analytical methods include scanning electron microscopy (SEM), X-ray diffraction (XRD), energy-dispersive X-ray spectroscopy (EDS), Fourier-transform infrared spectroscopy (FTIR), and various mechanical testing approaches. By utilizing these analytical techniques, researchers can gain valuable insights into the effectiveness and performance of biocementation, facilitating further advancements and optimization of this sustainable approach for various applications in soil improvement, environmental remediation, and construction materials. Table 2 summarizes some commonly used analytical techniques [48, 59, 60] for assessing the effectiveness of biocementation.

Preparing standardized soft samples in MICP sand or soil testing for unconfined compressive strength (UCS) tests is often difficult, costly, and time-consuming. Luckily, the needle penetration test (NPT) was developed in Japan [61] as a more straightforward way to estimate the UCS of soft rocks indirectly. The needle penetrometer is a small, portable, non-destructive testing device that measures load and penetration depth to calculate the needle penetration index (NPI). The NPT is user-friendly and appropriate for assessing the mechanical durability of sand samples that underwent MICP. As MICP-treated sand samples may have improved cohesion and strength due to the formation of calcium carbonate bonds, the NPT can provide essential insights into their mechanical properties. By measuring the resistance encountered when a needle is inserted into the sand sample, the NPT can assess the firmness and compactness of the material [62]. It can help determine the degree of cementation achieved through MICP and evaluate the effectiveness of the treatment. The compressive strength test remains a crucial and valuable method for assessing the effectiveness of MICP on soil stabilization and biocementation. It quantitatively measures the material's ability to withstand applied loads and is widely used in engineering applications. However, the NPT test offers additional advantages in certain scenarios. It provides a qualitative assessment of the soil's firmness and compactness, allowing for a quick and relatively simple evaluation of the effects of MICP. The NPT can be particularly useful in field applications where on-site testing is needed or when conducting preliminary assessments. By combining the compressive strength test and the NPT, researchers can understand the mechanical properties and performance of the MICP-treated soil. The UCS estimated that NPT could be a valuable tool for evaluating the improvement of slopes [32].

Table 2. Analytical techniques for assessing the effectiveness of biocementation.

Techniques	Properties and Performance
X-Ray Diffraction (XRD)	XRD is a valuable tool for determining what minerals are in biocemented materials. It does this by using X-rays to see how much certain minerals, like calcium carbonate, are present. This technique helps to check if biocementation was successful and gives information about the quality and properties of the material.
Scanning Electron Microscopy (SEM)	SEM allows for the examination of biocemented materials at a microstructural level. It provides detailed information about precipitated minerals' morphology, distribution, and interfacial characteristics. SEM can also be used to observe the bonding between microorganisms, minerals, and the substrate, providing insights into the effectiveness of the biocementation process.
Energy-Dispersive X-ray Spectroscopy (EDS)	EDS is often coupled with SEM to determine the elemental composition of biocemented materials. It identifies and maps the distribution of different elements, including calcium, carbon, and other trace elements. EDS analysis helps understand the spatial distribution of minerals and their correlation with the microorganisms involved in the biocementation process.
Fourier Transform Infrared Spectroscopy (FTIR)	FTIR spectroscopy analyzes the functional groups and chemical bonds present in biocemented materials. It can detect changes in chemical composition, such as the presence of carbonates, organic matter, or other compounds. FTIR analysis aids in assessing the formation of mineral phases and the overall quality of the biocemented material.
Permeability Testing	Permeability testing measures the flow of fluids through biocemented materials and provides insights into their durability and resistance to fluid transport. It helps assess the reduction in permeability achieved through biocementation and the effectiveness of the treatment in preventing the migration of contaminants or water.
pH and Chemical Analysis	Monitoring pH changes and conducting chemical analysis of pore water or leachates can provide information about the interaction between microorganisms, minerals, and the surrounding environment. It helps assess the impact of biocementation on water chemistry and the potential leaching of by-products.
Mechanical Testing	It is necessary to do mechanical tests to see how strong, stiff, and long-lasting biocemented materials are. These tests, like unconfined compressive strength (UCS), needle penetration test (NPT), and shear tests, help us see how much better the materials are after biocementation. By comparing biocemented materials to untreated ones or other cementation methods, we can measure how well biocementation works.

The NPT measures the resistance of soil to penetration by a needle or probe, indicating its strength and stability. The changes in soil strength can be assessed and compared by conducting

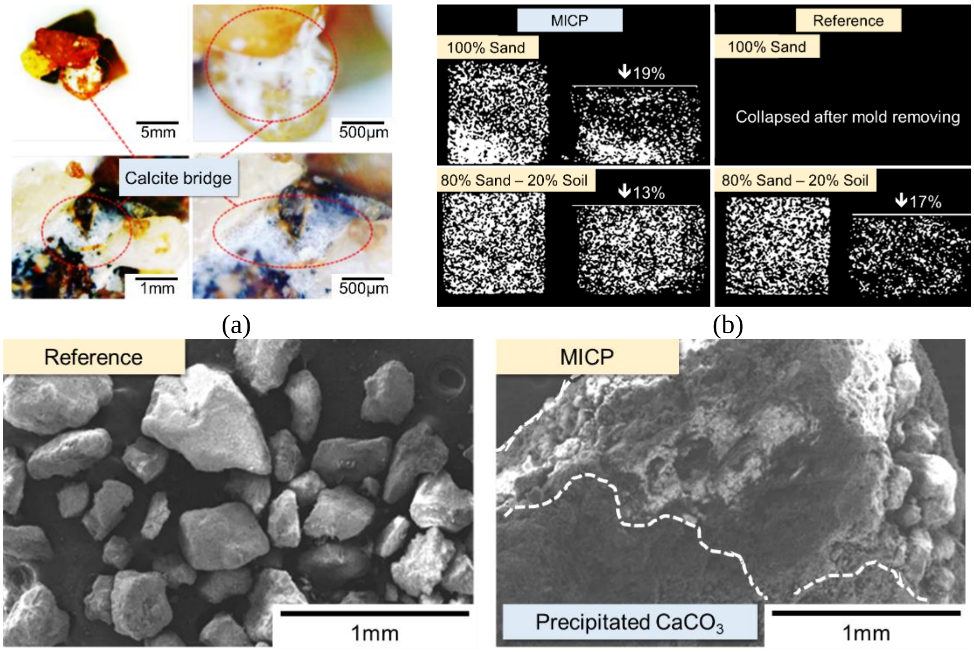
NPT before and after applying biocementation techniques. An increase in UCS values after the treatment indicates improved slope stability and resistance to deformation. This information is crucial for evaluating the effectiveness of biocementation methods in slope stabilization projects and making informed decisions regarding the suitability of the treated slopes for long-term use.

The mentioned analytical techniques, used individually or in combination, allow researchers and engineers to evaluate the effectiveness of biocementation processes, assess the quality of biocemented materials, and optimize the process parameters for desired outcomes. The characterization of unconventional materials used in biocementation can present challenges due to their atypical composition, which may require different tests and methods than conventional materials. The standardized methods and apparatus typically used to characterize conventional materials may need to be revised to evaluate these unconventional materials' specific properties. In such cases, alternative approaches such as chemical analysis or customized testing protocols may be necessary. Additionally, the lack of standardized methods tailored to these materials further complicates their characterization. Addressing these limitations requires developing specialized techniques and protocols that can accurately assess the unique properties and composition of biocemented materials.

3.2. Case studies biocementation in Viet Nam

It has been found that the self-healing concrete field has achieved positive results and advancements in recent years in Viet Nam [3, 14, 63, 64]. Laboratory studies, such as those conducted in 2019 and beyond [65, 66], have explored the possibility of using *Bacillus subtilis* (HU58) bacteria for soil cementation. Although the experiments primarily employed small soil-like specimens and unconventional approaches, deviating from standardized testing methods in geotechnical engineering, significant findings were obtained, warranting further research in subsequent phases. Bacteria were directly mixed into the soil at a concentration of less than 5 %, displaying metabolic activity for calcium carbonate precipitation. Material characterization through SEM, XRD, and FTIR analysis confirmed the presence of a mixture of calcite and aragonite as the precipitated $CaCO_3$. These mineral phases were crucial in cementing sand grains and filling the microstructure components. However, future studies should incorporate Raman spectroscopy with FTIR to discern potential polymorphs. The results also demonstrated the benefits of incorporating precipitated $CaCO_3$ in the sand matrix. The specimens' water absorption and deformation rate affirmed the prevention of water penetration and improved stability of the sandy samples.

Biocementation proved effective in maintaining the shape and integrity of the samples during the compression test. The impact of MICP was particularly evident in the case of the 100 % sand sample without bacteria, which wholly collapsed upon mold removal and was consequently unsuitable for compression testing (Figure 9b). Notably, the micrographs in Figure 9c clearly depict the consolidation and cementation of individual sand grains into a cohesive and densely compacted solid through the MICP process. The experimental findings demonstrated the significant potential of *Bacillus subtilis*, a sporulating microorganism, in MICP. This strain naturally forms spores, particularly under challenging environmental conditions or nutrient-limited environments, allowing for prolonged viability. Consequently, adding supplementary chemicals or nutrients, as required, presents a viable option for restarting or accelerating the MICP process. The optimization of bacterial concentration and nutrient selection emerges as a practical approach. Exploring alternative nutrient sources and streamlining the process can effectively maximize the advantages of MICP for large-scale applications while ensuring cost-effectiveness.



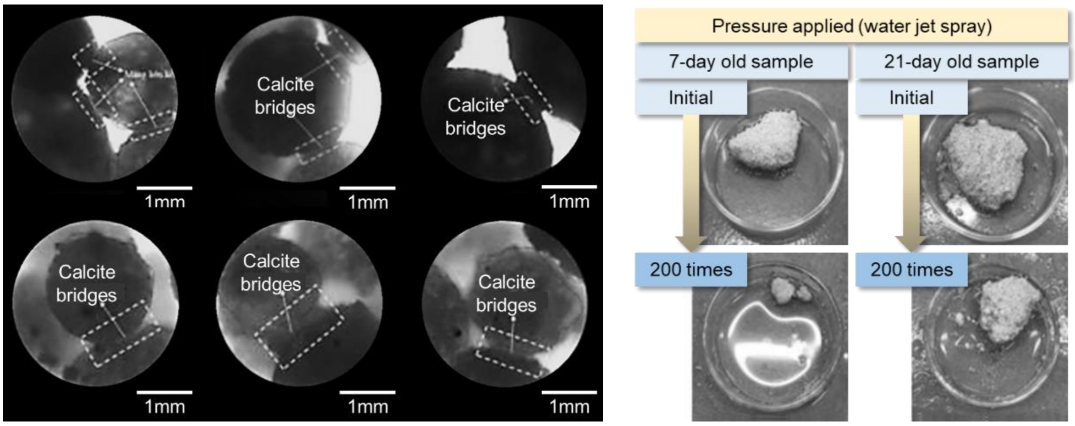
(a) (b)

(c)

Figure 9. (a) The sand grains are bonded by the biocementing process. (b) Vertical deformation after compression of the bacterial-treated cubes and the control group. Adapted with modification from [66, 67]. (c) SEM image of microstructure of the discrete control sand sample and the sand sample after cementing by bacterial biomineralization. Adapted with modification from [67].

A study investigated the mechanism and process of MICP by utilizing a constructive model of soil biocementation using expanded glass granules [68]. In this study, *Sporosarcina pasteurii*, a ureolytic bacterium, and a mixture of well-packaged expanded glass granules were employed as a soil model. Microscopic observations revealed the presence of bonding bridges between adjacent glass granules placed side by side on a petri dish and in a cylindrical mold. The impact of curing time on the bonding materials was a decisive factor for specimens cured for 7 and 21 days. Utilizing the soil particle packing model with glass granules facilitated the investigation of bacterial cementing ability. These findings provide valuable insights for next studies on soil biocementation involving different strains of bacteria.

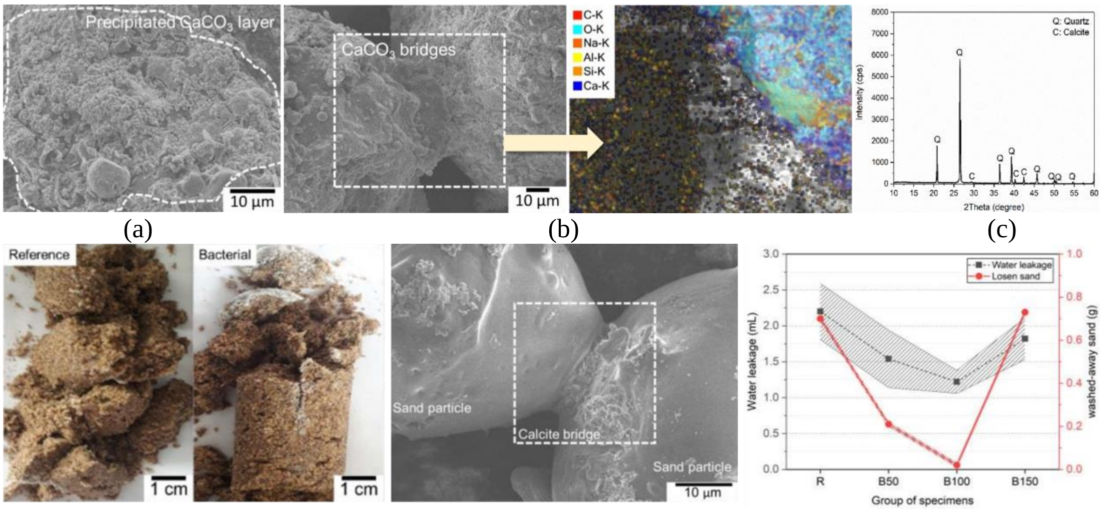
Based on the 21-day specimens (Figure 10b), the deformation experienced was minimal. The shape and bond between the glass granules remained intact. However, after conducting 200 times of spraying on 7-day-old samples, it was discovered that the bulk size could not be sustained under prolonged pressure. On the other hand, the 21-day sample maintained its bulk state despite some damage. This result, combined with microstructure analysis (Figure 10a), indicates that the binding ability of generated calcite by bacteria in the case of glass granules is highlighted at 21 days. While a more extended curing period of 21 days yields the best results, it acknowledges that this duration may be impractical in specific applications due to time constraints. However, a combination of various processes, such as natural carbonation and different phases of MICP, along with other chemical reactions, could potentially reduce the required curing time in mineral-rich environments. This concept opens up possibilities for optimizing the MICP process by leveraging multiple factors and mechanisms to enhance the efficiency of biocementation. Further research and experimentation could lead to innovative approaches for achieving robust and timely cementation using MICP techniques.



(a) (b)

Figure 10. (a) Microscopic observation of bonding bridges between glass granules after 7-day and 21-day curing periods. (b) Dimensional variation results of 7-day and 21-day samples after 200 instances of applied pressure (water jet spray). Adapted with modification from [68].

In 2021, a study explored the solidification and cementation of separated sand grains using ureolytic bacteria, specifically *Sporosarcina pasteurii* [69]. Ureolytic bacteria are known to release urease enzymes, which catalyze the hydrolysis of urea to produce ammonium and carbonate ions. In a calcium-rich environment, these carbonate ions react with calcium ions to form calcium carbonate precipitation. The study's objective was to investigate the potential of ureolytic bacteria to act as an adhesive mortar to solidify and cement individual sand grains. The study's findings aimed to contribute to the understanding and developing of innovative techniques for sand consolidation and bonding using biologically induced calcium carbonate precipitation. Based on that promising results, in 2022, a LAB-scale study [70] utilized *Sporosarcina pasteurii* for ureolytic MICP to reinforce soil sand. Preliminary findings highlighted the importance of increased $CaCO_3$ crystal formation over the curing period, as it played a critical role in establishing strong bonding between soil-sand particles. Additionally, positive results from water permeability tests and materials analysis using SEM and XRD confirmed the biocementation of sand specimens by bacterial action. Furthermore, experimental outcomes regarding the supplementation of oxidizing agents demonstrated a beneficial effect on enhancing MICP capacity. Despite numerous challenges, biomineralization or biocementation emerges as a promising technique for geotechnical engineering and building material applications, fostering sustainable development. Notably, the employment of *Sporosarcina pasteurii* and oxidizing agent biocementation exhibited a superior response compared to previous techniques. In a similar study [71], researchers found a way to use biological techniques to create bio-cement to strengthen loose sandy soil. They grew microorganisms in a lab to create the necessary chemical reactions, forming calcium carbonate. They then used a circulating pump to introduce the solution to sand columns, which improved their strength by up to 5 MPa. By analyzing the microstructure, the researchers could understand how the process worked and gain insights into its practical applications.



(a) (b) (c)

(d) (e) (f)

Figure 11. (a) SEM image of precipitated $CaCO_3$ layer on sand grains. (b) SEM image of $CaCO_3$ bridges between sand grains. (c) XRD pattern of the precipitated $CaCO_3$ [70]. (d) Sand samples with and without MICP. (e) Calcite bridges in sand column (f) Water leakage through the sand column. Adapted with modification from [72].

Figure 11 illustrates the results of the analysis conducted on the biocemented samples. The scanning electron microscopy (SEM) images reveal the presence of a well-formed $CaCO_3$ layer on the surface of the sand particles, indicating successful precipitation. Moreover, the formation of $CaCO_3$ bridges between the sand particles can be observed, signifying the consolidation of the material. The elemental distribution within the $CaCO_3$ bridges is mapped using EDS, providing insights into the distribution of calcium and carbon throughout the bridging structures. Additionally, XRD pattern confirms the crystal structure of the precipitated $CaCO_3$, validating its composition. Overall, this figure visually demonstrates the formation of the $CaCO_3$ layer, bridges, and provides valuable information about their elemental composition and crystalline structure. Including oxidizing agents provided favorable conditions that sustained the high MICP capacity of *Sporosarcina pasteurii*. The results obtained through sand column experiments indicated that strong intergranular bridging played a significant role in erosion resistance. The cementation and stabilization of separated and unstable particles within the soil-sand system were markedly improved. Overall, the findings presented in this study signify the promising potential of MICP as a biocementation technique to stabilize and enhance surface soil structure.

In 2023, a research group [73] worked in biocementation with a technique similar to MICP, EICP, to produce bio-cement. The researchers extracted an enzyme called Urease from soybean seeds, which exhibited high urease activity in bio-cement production. The study investigated the urease activity under different pH conditions and measured electrical conductivity in the solution resulting from urea hydrolysis. To enhance the strength of sandy soil, the researchers employed percolation treatment, which involved solidifying the soil at the particle contacts. The compressive strength of the treated sand was measured, with the maximum and minimum values recorded as 1004.2 kPa and 359.9 kPa, respectively. The precipitation of calcium carbonate through EICP significantly reduced the void volume and permeability of the soil. The initial permeability was measured at 10 cm/s. Analysis techniques such as XRD and SEM were used to confirm the presence of carbonate precipitation. In addition to the technical aspects, the study assessed the cost and environmental benefits of reducing CO_2 emissions compared to conventional cement (Portland cement).

4. CHALLENGES AND FUTURE DIRECTIONS

4.1. Limitations and challenges in the implementation of biocementation techniques

Table 3. Limitations and challenges of biocementation.

Limitations and challenges	Content
Reaction Kinetics	Biocementation processes, such as microbial-induced carbonate precipitation, often exhibit slow reaction kinetics. The precipitation of minerals takes time, which can be a constraint in time-sensitive projects or applications requiring rapid soil stabilization or remediation. Accelerating the reaction kinetics while maintaining the desired effect is an area of ongoing research.
Environmental Sensitivity	Biocementation is sensitive to environmental factors such as temperature, pH, nutrient availability, and moisture content. Suboptimal environmental conditions can hinder the growth and activity of microorganisms, impacting the effectiveness of the process. Understanding and optimizing these environmental parameters are crucial for successful biocementation.
Variability in Microbial Performance	Different microbial strains exhibit varying performances in biocementation processes. Selecting suitable microbial species or strains compatible with the specific project conditions and target materials is essential. Additionally, maintaining the viability and activity of microorganisms during transportation and application can be challenging.
Uniform Distribution of Microorganisms and Precipitated Minerals	Achieving a uniform distribution of microorganisms and the resulting precipitated minerals throughout the treated material is critical for consistent and reliable biocementation. Uneven distribution can lead to variations in material properties, compromising the overall effectiveness of the treatment. Developing techniques to ensure uniform distribution is an area of active research.
Scale-Up and Cost	Scaling up biocementation techniques from laboratory or pilot-scale to field-scale applications can be difficult. It's essential to consider the cost-effectiveness of implementing large-scale production and application of microbial cultures, nutrient solutions, and monitoring equipment. Evaluating economically viable solutions before adopting biocementation in various industries and sectors is crucial.
Long-Term Durability	Assessing the long-term durability and performance of biocemented materials is crucial for their acceptance and successful implementation. Factors such as material aging, potential degradation of microbial activity, and the ability of the cemented material to withstand environmental stresses over time need to be considered. Long-term monitoring and evaluation of biocemented materials are essential to ensure their effectiveness.
Integration with Existing Construction Practices	Incorporating biocementation techniques into existing construction practices and regulations can be challenging. Collaboration between researchers, engineers, and regulatory bodies is necessary to establish guidelines, standards, and acceptance criteria for biocementation applications. Education and awareness programs are also vital to promote the understanding and acceptance of biocementation among industry professionals and stakeholders.

Implementing biocementation techniques in practical applications faces several limitations and challenges that must be addressed for successful adoption. These challenges include the cost implications associated with specialized bacteria, nutrient sources, and monitoring equipment, requiring cost-effective solutions and resource optimization. Scaling up biocementation from the laboratory to field applications requires careful planning to ensure uniform distribution of bacteria and nutrients, consistent environmental conditions, and logistical management. The long-term durability and performance of biocemented materials, including resistance to environmental factors and mechanical stresses, must be thoroughly evaluated. Regulatory and approval processes, ecological considerations, and sustainability necessitate collaboration between researchers, industry stakeholders, and regulatory bodies to establish guidelines, standards, and protocols for safe and effective implementation. Table 3 lists some critical considerations of biocementation techniques [47 - 49].

Addressing these limitations and challenges requires ongoing research, technological advancements, and collaborative efforts among researchers, engineers, industry practitioners, and regulatory authorities. Overcoming these obstacles will contribute to successfully implementing and adopting biocementation techniques in various fields, including construction, environmental remediation, and infrastructure development.

4.2. Environmental impacts and sustainability of biocementation

When considering methods for soil stabilization and environmental cleanup, keeping in mind their impact on the environment and sustainability is necessary. Selecting microorganisms carefully to ensure they are native and will not harm the environment is one effective way to minimize ecological risks. Additionally, finding sustainable alternatives to the chemicals and nutrients used is crucial. Utilizing fewer resources and less energy to create and transport materials can be helpful, as can discovering ways to recycle water. Long-term testing is crucial to assess the compatibility of these methods with the ecosystem and to evaluate their environmental impact over extended periods. Monitoring and analyzing the ecological changes, such as biodiversity, soil fertility, and water quality, is crucial to understand any potential long-term effects and ecological interactions resulting from implementing these methods. This information is essential for ensuring the sustainability and responsible use of biocementation techniques, and it can guide decision-making and regulatory frameworks to mitigate any adverse effects on the ecosystem. The sustainability of biocementation lies in its potential to provide environmentally friendly and long-lasting solutions for various applications. Biocementation offers several advantages that contribute to its sustainability. Firstly, it utilizes natural biological processes and does not rely on synthetic chemicals, reducing the environmental impact. Additionally, using microorganisms or enzymes in the process can be more energy-efficient than conventional cement production methods. Biocementation also has the potential to utilize waste or by-products as nutrient sources, promoting circular economy principles and reducing waste generation. Furthermore, biocementation can enhance the durability and strength of soils or construction materials, leading to a longer lifespan and reduced maintenance needs. However, challenges remain, such as optimizing process efficiency, scalability, and compatibility with different soil and environmental conditions. Continued research and development in biocementation techniques are essential to maximize its sustainability potential and ensure its practical implementation in various fields. Combining biocementation with green building and waste reduction can make soil improvement more sustainable. Continuously researching and collaborating with stakeholders to ensure these practices are sustainable and environmentally beneficial in the long run is vital.

5. CONCLUSIONS

In conclusion, biomineralization emerges as a natural process with considerable promise for engineering applications. This paper has provided an in-depth exploration of microbial-induced mineral precipitation and applications. Specifically, the focus has been on developing and ongoing research of biomineralization applications. Our review has underscored the influential parameters governing biomineralization, with significant attention given to the advancement of novel construction materials, soil stabilization techniques to mitigate public health hazards, and subsurface applications targeting the enhancement of wellbore integrity. Notably, calcium carbonate is the most frequently employed mineral, primarily activated through microbial urea hydrolysis. Construction and soil stabilization advancements have shown considerable progress, some of which have already achieved commercialization.

Moreover, several other technologies are on the precipice of full-scale implementation, paving the way for subsequent commercial opportunities. Nevertheless, it is imperative to recognize that additional research and development initiatives are indispensable for fully unleashing the extensive capabilities of biomineralization in engineering. These endeavors will undoubtedly pave the way for uncovering novel applications in construction, environmental science, biotechnology, and medicine. Ultimately, such advancements will contribute significantly to developing environmentally sustainable methodologies and economically viable solutions. Biological manufacturing methods, such as engineered mineral precipitation, are promising to reduce the energy-intensive processes associated with cement manufacturing and promote resource efficiency and climate conservation. Mainly, microbially induced calcium carbonate precipitation demonstrates the potential to diminish the carbon footprint of building and construction materials significantly. However, future work is required to expand its application scope beyond the realms currently dominated by cement. Biomineralization finds value in diverse areas. There are instances where biomineralization offers unique advantages that do not directly compete with traditional cement, such as the coprecipitation of specific groundwater contaminants like strontium and the restoration of good integrity in ultrafine leaks. The progress of engineered microbial mineral formation from ideas to real-world technology highlights the significance of biomineralization in solving engineering problems. Improving biomineralization methods and their utilization for finding sustainable solutions in different industries is crucial.

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