



# Formation of water-impermeable crust on sand surface using biocement

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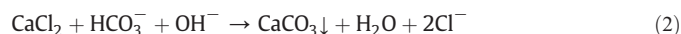
## ABSTRACT

This paper examines the feasibility of using calcium-based biocement to form an impermeable crust on top of a sand layer. The biocement used was a mixture of calcium salt, urea, and bacterial suspension, which hydrolyzed urea with production of carbonate and an increase of the pH level. Applying 0.6 g of Ca per cm<sup>2</sup> of sand surface, the permeability of the biocemented sand can be reduced from 10<sup>−4</sup> m/s to 1.6·10<sup>−7</sup> m/s (or 14 mm/day) due to formation of the crust on sand surface. The rupture modulus (maximum bending stress) of the crust was 35.9 MPa, which is comparable with that of limestone. The formation of a water-impermeable and high strength crust layer on sand surface could be useful for the construction of aquaculture ponds in sand, stabilization of the sand dunes, dust fixation in the desert areas, and sealing of the channels and reservoirs in sandy soil.

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## 1. Introduction

Biocementation is an innovative technology based mainly on application of urease-producing microorganisms together with urea and calcium ions in a permeable soil [1–4]. Hydrolysis of urea by enzyme urease causes calcium carbonate precipitation and formation of a cemented product according to the following equations [5]:



It has been shown that urease-producing bacteria or enzyme urease can be used to bind sand particles through calcite formation, a process known as biocementation [4,6–8].

There are many potential applications of biocementation in civil engineering such as enhancing stability of slopes and dams, reducing the liquefaction potential of soil, road construction, prevention of soil erosion [1–5], and reparation of the cracks in concrete [7,9–11]. The formation of aquaculture ponds or reservoirs in sandy soil is a new application of biocementation, which has not been studied. These ponds could be used for outdoor commercial aquaculture, such as fish, shrimp and mollusk production [12], for large-scale cultivation of algae [13], for biofuel production in desert coastal area, or as water collecting reservoirs.

It is known that excessive seepage from aquaculture pond or reservoir is a major problem in areas with highly permeable soils.

Seepage from stable ponds causes 45% to 87% total water losses [14,15] and the seepage rate (or water permeability) could rise up to 182 mm/day [16]. Seepage from the aquaculture pond causes not only the loss of water but also leakage of nutrients needed for aquaculture [17–19]. It may also cause pollution of groundwater with nutrients, organic aquacultural wastes, and pathogens from aquaculture pond. For example, when the total average of 1021.2 kg/ha potassium was applied to the newly constructed shrimp ponds, the estimated loss of potassium due to seepage was 101.2 kg/ha [20]. Therefore, cutting off seepage and decreasing permeability of soil are an important design consideration for the applications listed above. The feasibility of using calcium-based biocement to seal or construct the water pond in sand is discussed in this paper.

## 2. Materials and methods

### 2.1. Urease-producing bacteria

Halotolerant and alkalophilic strain of urease-producing bacteria *Bacillus sp.* VS1 has been isolated from sand of tropical beach. It was spore-forming, Gram-positive bacteria with rod-shaped cells. The nearly full-length 16S rRNA gene was amplified conventionally by Polymerase Chain Reaction (PCR) with the primers 27F, 530F, 926F, 519R, 907R and 1492R. Purified PCR products were sequenced using the ABI PRISM 3730xl DNA sequencer and the ABI PRISM BigDye Terminator Cycle according to the manual of the manufacturer (Life Technologies Corporation, California, US). The partial nucleotide sequences were assembled to produce the full-length nucleotide sequence of 16S rRNA gene deposited in NCBI GenBank under accession number [JF896459](https://www.ncbi.nlm.nih.gov/nuclot/JF896459). To identify microbial strain, the full-length nucleotide sequence of its 16S rRNA gene was compared with

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