



Bio-Based Self-Healing Concrete for Sustainable and Durable Concrete Infrastructure

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Abstract

In this study, bio-self-healing concrete was manufactured using a natural phenomenon called microbial-induced calcium carbonate precipitation (MICP). The *bacillus cereus* bacteria isolated from Qatari soil was used for this purpose. These bacteria have endured the harsh weather of high temperatures, humidity, and alkaline soil conditions. Hence, are a potential candidate for long-term self-healing in concrete structures that are subjected to the climate of the Middle Eastern region. The bacteria were encapsulated in sodium alginate beads then the beads were added to the cement-sand mortar. The nutrients for bacteria such as urea, calcium nitrate, yeast extract, and calcium chloride were mixed in mortar as dry constituents. After curing for 28 days, cracks were artificially induced in the prismatic samples, which were reinforced with steel rebars at the tensile side. Samples were placed in water to instigate self-healing. It was observed that the bacteria healed the cracks up to 0.70 mm. It is concluded that the used bacteria are viable in the alkaline concrete matrix and capable of producing calcium carbonate.

Keywords: Bio Self-Healing Concrete; MICP; *Bacillus Cereus*; Autonomous Crack Filling; Infrastructure Durability

1 Introduction

Reinforced concrete (RC) infrastructure in the Middle Eastern Region suffers cracking under the harsh climate of high temperature, humidity, and chloride environment. Cracks are generated in concrete under mechanical, as well as, environmental loading. These cracks allow the ingress of deleterious agents into concrete, which in turn cause the corrosion of reinforcing steel rebars. Once corrosion is initiated the durability of the RC structure is drastically reduced (Sohail et al., 2018, 2022a). Several remedies are available to enhance the lifespan of RC structures, namely strengthening with carbon fibers using repair mortar, or injecting the epoxy/slurry into the cracks (Sohail et al., 2021; Al Nuaimi et al., 2021). However, these solutions do not protect against cracking and subsequent corrosion as the surface area of exposed concrete is so large that it is physically impossible to cover.

Autonomous self-healing using bacterial strains has emerged as a potential candidate to overcome the cracking issues of concrete under service (Tziviloglou et al., 2016; Van Tittelboom et al., 2010). The healing is done by the metabolism of bacteria who consume nutrients and produce calcium carbonates in the cracked portion. Calcium carbonate is inherently a binding material in itself and bonds with the cement matrix and hence concrete gets healed. This calcite precipitation is called microbial-induced calcium carbonate precipitation (MICP) (Wiktor & Jonkers, 2011; Wen et al., 2019; Sohail et al., 2022bb).

Several naturally occurring bacterial strains are capable of producing MICP under a suitable environment. Most commonly used are *Sporocarsinapastuerii* and most of the *bacillus* genes. The pathway for producing MICP is the urea decomposition by the urease enzymes produced by bacteria (De Belie & Wang, 2016; Tittelboom et al., 2016). The living cells produced enzymes through metabolism, which then work as a catalyst in the hydrolysis of urea into ammonia and carbonate, the pH of the surroundings is then increased. Ammonia (NH₄⁺) and CO₃²⁻ (carbonic acid) are formed after upon further hydrolysis of urease. The bacterial walls are charged negatively, they form a bond with the cations of Ca²⁺, and reach with CO₃ ions and calcium carbonates (Anbu et al., 2016).

Among the MICP-forming bacteria, not all can produce calcites under a highly alkaline environment which is encountered in concrete. That is why they are needed to be encapsulated in the polymeric beads and then incorporated in to the concrete matrix. The beads used in literature are of different materials, sizes, and shapes. A few examples of encapsulating materials are ethylene oxide and propylene oxide (Wang et al., 2014b), silica sol-gel (Yang et al., 2011), lightweight clay particles (Tziviloglou et al., 2016), hydrogels (Wang et al., 2014a), engineered aggregates (EA) Pan et al., 2023), glass granules (Zhang et al., 2021), and expanded perlite (Alazhari et al., 2018). Sodium alginates, carrageenan, and chitosan are biopolymers that are used as carriers of healing agents.

In this study, a locally available *bacillus cereus* strain that was isolated locally from Qatari soil was studied for calcite precipitation in concrete. These strains have the advantages that they are weathered and evolved under a harsh climate of high temperature, humidity, salty environment, and high pH soils. Hence, it is expected that these strains will be viable in high-pH of concrete and could produce calcite precipitation in cracks. These strains were encapsulated in sodium alginate

beads. These strains are never been used in concrete, especially for application in the Middle Eastern Region. Bio self-healing concrete made *bacillus cereus* isolated from Qatari soils can arrest the degradation problem in structures subjected to Middle Eastern climates.

2 Experimental Detail

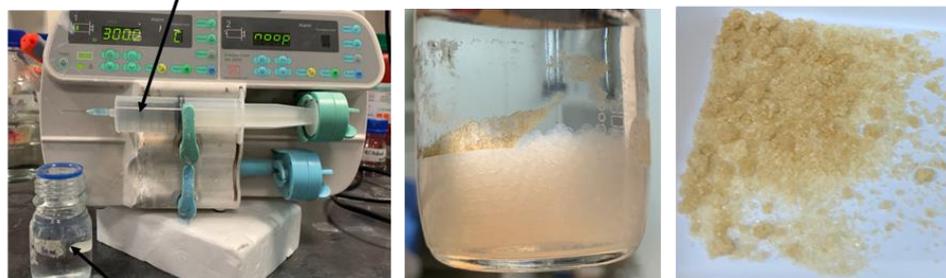
2.1 Bacterial Strains and Sodium Alginate Beads

The *bacillus cereus* bacteria were selected because already showed calcite precipitation in previous works where soil stabilization was performed with MICP. The *bacillus cereus* strains performed MICP at 40°C in the field in previous works (Oualha et al., 2020; Bibi et al., 2018). These bacteria were preserved in 30% glycerol at -80° in the Qatari Mineral Precipitating Strains Bank. They were then revived as fresh living cells by striking them on a medium of Luria-Bertani (LB).

The growth media, Luria Bertani, and Urea Medium were used to grow the strains before adding in the beads. The LB was comprised of NaCl (5 g/l), tryptone (10 g/l), and yeast extract (5 g/l) whereas UM contained NaCl (5 g/l), Peptone (2 g/l), glucose (1 g/l), KH₂PO₄ (2 g/l), and of Urea (20 g/l). After that, the strains were recovered and inoculated in liquid media so that they could be placed in sodium alginates.

The beading procedure is presented in Fig.1. The sodium alginate of 1 % by weight was mixed in 1 litre of autoclaved water. A magnetic stirrer was placed in the bottle to mix the alginates homogeneously once the spores were formed in the urea media. The mixture of bacterial spores inside the media was added to the sodium alginates. They were stirred for 15 minutes. Then the mixture was introduced into a 2% calcium chloride solution, drip by drip, as shown in Fig.1a. Fig.1b shows the bead in the CaCl₂ solution, and Fig.1c shows the dried bead.

Sodium Alginate with bacteria spores



a) 2% (w/v) Calcium chloride

b) Sodium alginate beads with immobilized bacteria

c) Dried beads

Fig.1: Procedure for encapsulation of bacteria in the sodium alginate beads, a) sodium alginate was dripped in to 2% calcium chloride solution, b) fresh beads, c) dried beads

Nutrients for the bacterial strains were mixed into the mortar. The nutrients such as urea, yeast and calcium nitrate were introduced as dry ingredients in the cement-sand mortar. Several researchers used the selected nutrients for bacteria. The selected amount was sufficient for bacteria to get food and grow.

2.2 Mortar for the Self-Healing Process

Table 1 shows the mortar mixture used in this study. A ratio of 1:2.5 of cement/sand was selected, whereas, water/cement ratio was 0.5. Three mixes were prepared, the control mix was cement, sand, and water. The second mix was comprised of urea, yeast, and calcium nitrate while keeping the other ingredient the same as the control mix. One mix was prepared by adding sodium alginate

beads containing *bacillus* species from local soil. The nutrients were mixed in a mortar in a dry state. The beads were dried before adding in mortar. The urea and yeast were 27 g and 4 g per cement weight, respectively, whereas, calcium nitrate was 33g by weight of cement.

Table 1: Concrete mix proportion for self-healing through bacterial strains

| Number of mixes | Mortar Mix type | Cement in grams | Sand in grams | Water-cement ratio | SP | Sodium Alginate Beads (g) | Urea in grams | Yeast Extract (g) | Calcium Nitrates (g) |
|-----------------|--|-----------------|---------------|--------------------|-----|---------------------------|---------------|-------------------|----------------------|
| | | | | | (g) | | | | |
| 1 | Control mix | 666 | 1665 | 333 | 10 | - | - | - | - |
| 2 | Control + nutrients (CN) | 666 | 1998 | 400 | 10 | - | 27 | 6 | 33 |
| 3 | Control + nutrients + <i>B. cereus</i> | 666 | 1665 | 333 | 10 | 16 | 27 | 6 | 33 |

2.3 Mortar Samples for Generating Cracks

The compressive strength was evaluated by casting and testing 50 x 50 x 50 mm cube samples following ASTM C109/C109M-02. (2002). Beads and Nutrients may affect the strength adversely, which is why compressive strength is essential to measure. To generate the cracks in the mortar matrix, prism samples of 40 x 40 x 160 mm were cast having steel rebar at the tensile side. The steel rebar was 4 mm in diameter and 15 mm in length. Bars were placed in their location with the help of fixing wires. Cube and prism samples were cured for 28 days by immersion in distilled water. Three cube samples were tested for each mix. Whereas prism samples were loaded using four-point bending using a manual hydraulic machine as shown in Fig. 2. The load was applied manually upto 5 MPa until the 3 to 5 cracks appeared in the mortar matrix. Formed cracks were between 50 to 1000 µm. Cracks appeared in the shear zone as no shear reinforcement was provided. All the prism samples were placed underwater for 3 days to increase the urease activities of the bacteria and observe if, there is, any mineral precipitation.

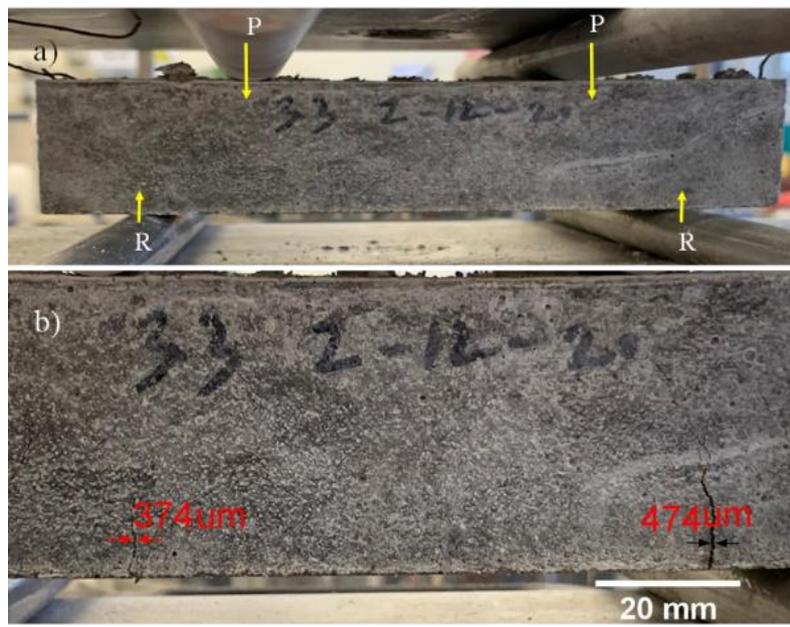


Fig. 2: a) prism samples under four-point loading for cracks formation. Crack generation in prism samples.
b) Cracks up to 474 µm in the samples with *bacillus cereus*

2.4 SEM and XRD on the Cracked Surface

Broken samples from the cracked portion were taken for SEM and XRD analysis. White precipitation was also analysed under XRD.

3 Results And Discussion

After the cracks were generated in the prisms, they were immersed under double distilled water this would instigate the transfer from spores to living cells of bacteria. The samples of mortar without beads (control), with only nutrients, and samples with only beads were also placed underwater after cracking. The samples having spores of *bacillus* are shown in Fig. 3.

Fig. 4 shows the sample of the beads containing bacillus cereus after immersion in water for 14 days. It can be observed that the formed cracks are filled with white precipitates of calcium carbonates. The cracks were filled where they passed through the beads. Cracks from 170 to 683 μm were filled with local strains.

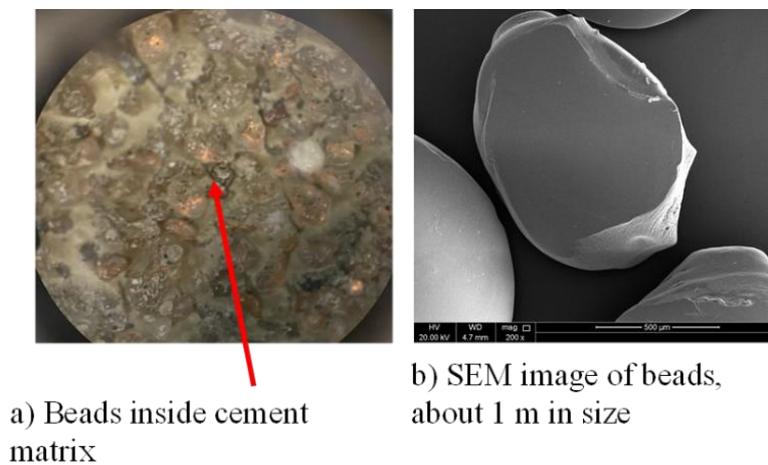


Fig. 3: a) Beads survival in mortar matrix under the microscope, b) SEM image of a bead before adding in mortar

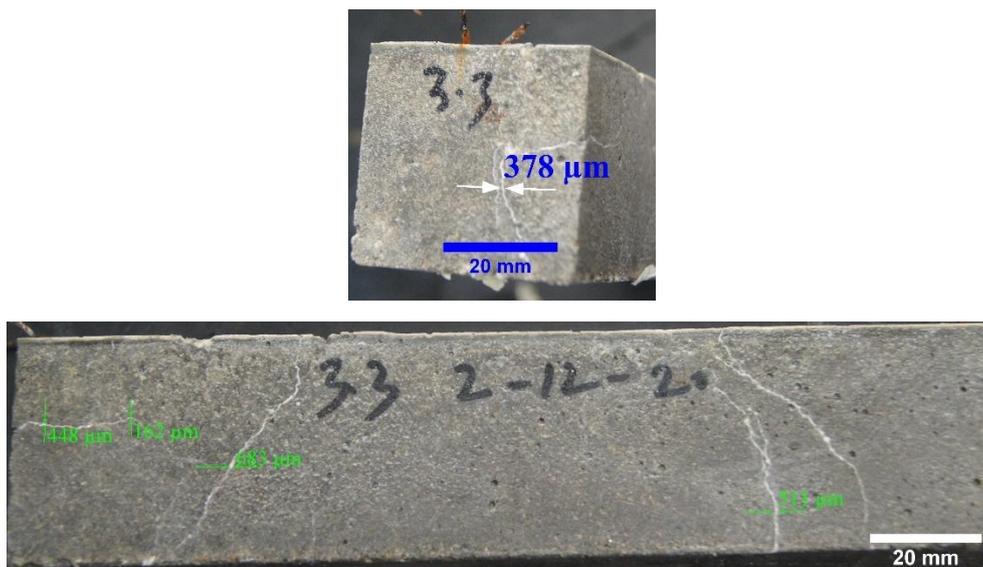


Fig. 4: The prism samples showing the self-healing of the cracks with local bacterial strains. The cracks of up to 680 microns were filled with calcium carbonates

Fig. 5 shows the SEM and EDX analysis of bacterial surroundings. It can be seen that bacteria were viable in the cement matrix. The presence of magnesium and phosphates in EDX analysis confirms the bacteria in concrete (**Fig. 5b**). **Fig. 5c** shows the hexagonal crystalline structure of calcium carbonates. **Fig. 5d** shows the EDX analysis of the white precipitates of calcium carbonates.

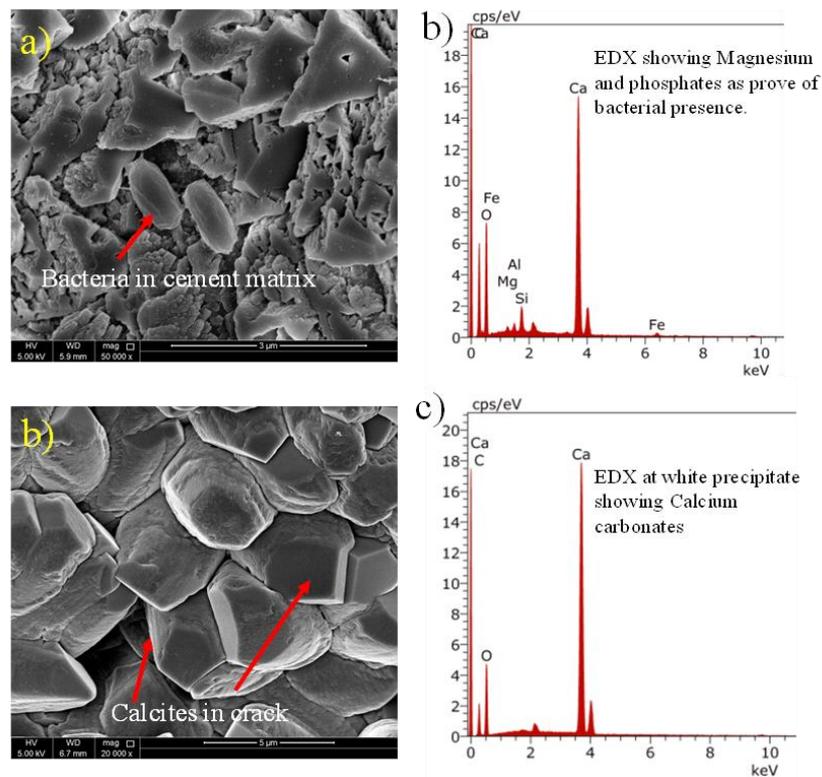


Fig. 5: a) SEM of bacterial survival in a cement matrix, b) EDX of bacterial surroundings, c) Calcium carbonate mineral (crystalline shape), d) EDX of calcium carbonate mineral

The XRD analysis of white precipitates filling the cracks was performed. **Fig. 6** shows the XRD spectra of the material filling the cracks for *bacillus*. The materials were similar in mineralogy, that is, calcium carbonate.

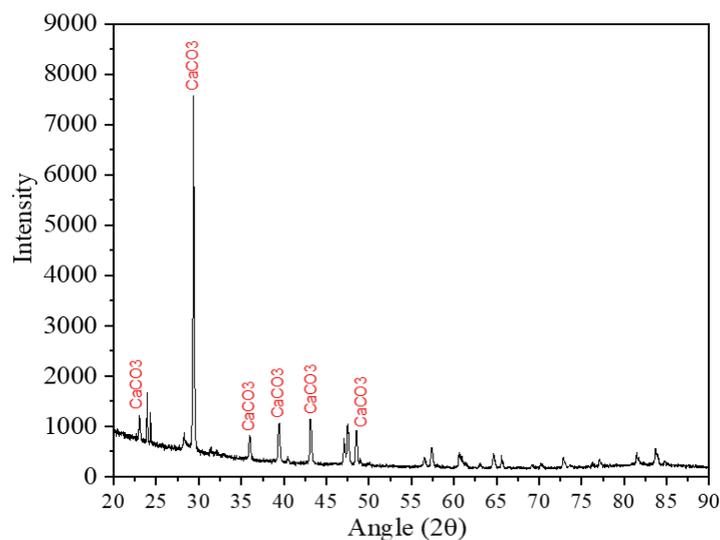


Fig. 6: XRD spectra of crack filling material in samples with local strains of bacillus cereus

4 Conclusion

The bio self-healing concrete is the need of the hour to arrest the degradation and enhance the infrastructure durability in the Middle Eastern Region. In this study, local bacterial strains of *Bacillus cereus* were embedded in sodium alginate beads and then these beads were incorporated into the cement mortar. Then cracks were generated in the prism samples and placed in the water.

It was observed that locally isolated *Bacillus cereus* from Qatari soil could survive in the cement matrix and also could perform healing of cracks. Crack from as small as 70 μm to 683 μm were healed with a white precipitate formed by bacteria in the cracks. Hence, the proposed bio-self-healing concrete could enhance the durability of RC structures subjected to harsh climatic conditions.

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