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The Effect of Self-Healing Microorganism-Encapsulating Concrete on Enhancing Concrete Compressive Strength

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Investigating microbially induced calcite precipitation (MICP) of concrete in bacillus and the durability of crack-filled repair structures were the goals of this work. The purpose of this study was to investigate the self-healing effect of concrete in bacillus and the strength of crack-filled repair structures. The characteristics of calcium carbonate particles and the performance of those penetrating cement were observed using optical microscopy. The cement block pressure test was used to study the factors affecting the healing rate of concrete materials mixed with microorganisms. The results showed that the microorganisms had good microscopic morphology. Microbial mixed soil had good compression resistance, and the ability to play a repairing role in mixed soil composite materials was that organisms could be attached to the concrete tightly. The main fracture behavior of the mixed soil was a small-hole rupture, while no macroscopic damage or large-pore ruptures were observed in the mixed soil matrix. *Sporosarcina pasteurii* exhibited better potential than Bacillus subtilis and could act as a self-healing agent in the concrete. The test results proved that *S. pasteurii* produced a colloidal adhesive to fill and repair cracks. The study designed concrete of different densities to create cubes having different compressive strengths, water permeability, and water absorption to further observe the ability of Bacillus to fill the cracks and prevent water penetration. The results showed a 60 % increase in the compressive strength of the coarse aggregate experimental sample and a 36 % decrease in the compressive strength of the fine aggregate experimental sample, relative to the same properties in the control sterile sample. Samples indicating the use of bacteria in the aggregate were denser and less porous. It was proven that the use of microorganisms could achieve self-healing ability in concrete materials, fill up pores, and establish functional effects.

Keywords: microbially induced calcite precipitation, concrete, self-healing, compressive strength.

1. INTRODUCTION

The infrastructure is aging all over the world, as we can see that the tensile strength of the concrete is reduced after many years, and the cracks and pores are enlarged because of hydration and shrinkage. Erosive substances intrude into concrete; steel bars rust; and temperature, water vapor, and air hazards cause the durability of engineering materials to decline. The cracks in the building body need to be artificially reinforced, and the maintenance cost is high. A considerable amount of money is invested in maintenance and reinforcement, wherein cement and petrochemical building materials are used the most. Concrete is a strong and relatively inexpensive building material that is widely used [1 – 3], with advantages including high compressive strength, usability, versatility, compatibility with steel bars, and fire resistance. The disadvantage is that concrete is prone to cracks after being exposed to wind, sun, and rain for a long time, and the tiny cracks caused by high temperature and climate change gradually expand, which reduces the durability of concrete and shortens the service life of engineering structures. Cement needs to go through high-temperature sintering at 1450 °C, which releases a large amount of CO_2 [4]. In addition to the long-term heat energy consumption, the emitted carbon dioxide accounts for 7 % of global emissions. Cement is a highly polluting material that is extremely damaging to the planet as factories continue to develop cement. The main cause of the shortened concrete life cycle is the atmospheric moisture content and temperature of the surrounding environmental conditions, which lead to the cracking of the concrete structure [5]. Over the years, the research and improvement of cement products and maintenance methods have continued, but the progress has been slow. In recent years, the problem of cracks has been solved by adding microorganisms to induce crack healing, and a mechanism of the self-healing of concrete cracks has been developed, because calcium carbonate is one of the materials that is compatible with concrete components [6]. The bacteria of the genus *Sporosarcina pasteurii* that exist in nature [7] have urea-decomposing enzymes, which can remove chemical substances from wastewater [8], adsorb heavy metals [9 – 11], produce biocrystalline minerals [12 – 16], and have the traits of easy cultivation.

Hames [17] showed that the urease in microorganisms utilizes the pH, dissolved inorganic carbon concentration, calcium concentration, and available nucleation sites to hydrolyze urea to form ammonia and carbamic acid, further decomposing the carbonate ions (CO_3^-), forming an alkaline environment, and the nucleation sites on the surface of microorganisms adsorb calcium ions to form stable and continuous calcium carbonate crystals [18]. This MICP

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method uses the negative charge on the surface of microorganisms to adsorb divalent cations, and Ca^{2+} has stronger ion selectivity than Mg^{2+} [19, 20]. Jonkers [21] proposed the use of bacteria-induced mineral precipitation (MICP) technology to self-heal concrete cracks without human intervention, repair cracks, and fill micropores [22 – 27]. Aerobic microorganisms were shown to significantly increase the compressive strength of cement mortars [28, 29]. MICP is composed of a series of complex biochemical reactions. Bacterial urease catalyzes urea to produce CO_2 and ammonia, resulting in an increase in the pH value of the surrounding environment. The nucleation site on the cell surface adsorbs calcium ions and carbonates in the form of $CaCO_3$. The step-wise decomposition of urea to produce calcium carbonate is shown in the following equations [30 – 33]:

$$\mathrm{CO(NH_2)_2 + H_2O \rightarrow NH_2COOH + NH_3}; \tag{1}$$

$$\mathrm{NH_2COOH + H_2O \rightarrow NH_3 + H_2CO_3}; \tag{2}$$

$$\mathrm{H_2CO_3 \leftrightarrow HCO_3^- + H^+}; \tag{3}$$

$$\mathrm{2\,NH_3 + 2\,H_2O \leftrightarrow 2\,NH_4^+ + 2OH^-}; \tag{4}$$

$$\mathrm{HCO_3^- + H^+ + 2\,OH^- \leftrightarrow CO_3^{2-} + 2\,H_2O}; \tag{5}$$

$$\mathrm{Ca^{2+} + Ba \rightarrow Ba - Ca^{2+}}\ ; \tag{6}$$

$$\mathrm{Ba - Ca^{2+} + CO_3^{2-} \rightarrow Ba - CaCO_3}. \tag{7}$$

The common pH value of the concrete mixture is close to 12, and the metabolism of some alkaliphilic microorganisms promotes the precipitation of calcium carbonate in the form of calcite to seal cracks [34], which is suitable for the generation of $CaCO_3$ [1, 9].

This paper discusses the impact of the existence of microorganisms on the durability of concrete, the effect of different calcium sources on coarse-aggregate and fine-aggregate concrete, and studies the compressive strength and water absorption of cement mortar.

2. EXPERIMENTAL PROCEDURE

2.1. Raw materials

As coarse and fine aggregates, Portland Cement of a Taiwanese brand and Yilan County sandstone were used as the freshwater standard aggregates.

Fine aggregate. The properties of natural river sand are listed in Table 1. Coarse aggregate. The coarse aggregate considered in this study is listed in Table 2.

Table 1. Properties of fine aggregate

Particles size, mm	Water absorption, %	Specific gravity	Ratio, %
2.36	0.54	2.719	41.08
1.18	0.57	2.629	40.71
0.60	0.61	2.671	11.79
0.30	0.65	2.487	4.82
0.15	0.68	2.438	0.89
0.075	0.69	2.456	0.71

2.2. Microbial material

S. pasteurii BCRC11596 (bought from Taiwan Culture Center BCRC) was cultured in a sterile setting, and the number of bacteria was determined using a UV800 spectrometer (in the Microbiology Laboratory of China Medical University, Taiwan).

Table 2. Properties of coarse aggregate

Particles size, mm	Water absorption, %	Specific gravity	Ratio, %
19.0	0.37	2.757	19.90
12.7	0.43	2.714	19.60
9.50	0.43	2.571	15.80
4.75	0.45	2.525	15.20
2.36	0.54	2.719	11.30
1.18	0.57	2.629	11.00
0.60	0.61	2.671	3.50
0.30	0.65	2.487	1.50
0.15	0.68	2.438	1.10
0.075	0.69	2.456	1.10

Strain culture preparation. Culture media, sterilized at 121 °C for 30 min with 18.5 g of beef in 1000 mL of distilled water, 5 g of protein, and pH 9.5 – 11.0. Following the inoculation of the microorganisms, the culture medium was shaken for 24 h at a temperature of 30 °C and a rotational speed of 170 rpm. The bacteria were allowed to settle to the bottom after centrifugation at 8000 rpm and then removed and resuspended in distilled water. More than 10^8 germs per milliliter were present in the bacterial suspension.

Induction of calcium carbonate formation. After culturing the suspension of 10^8 bacteria at room temperature for 24 h, 18.5 g of beef was added per liter along with 4 g of urea, and mixed well. Afterward, calcium chloride or calcium lactate was added to create a calcium carbonate coating (approximately 2.5 mL of bacillus liquid was added to 50 mL of the medium).

2.3. Construction of concrete test samples

Stones (coarse aggregate), sand (fine aggregate), water, and Portland cement are the major components of concrete. Water absorption for coarse aggregate samples made with a 10-cube mold is 0.54; specific gravity is 2.60; and W/C is 0.62. Water absorption for fine aggregate samples was made with a 5-cube mold is 0.62; specific gravity is 2.57; and W/C is 0.45. 4.8 % of the bacterial culture was added while the cement and aggregate were blended well. No bacteria were added to the control samples. All samples were cured after demoulding, either in water or at room temperature. Use a 200-ton automatic compression testing machine to carry out concrete pressure tests and water absorption tests on the seventh, fourteenth, and twenty-eighth days. The concrete's composition is displayed in Table 1.

2.4. Test method

2.4.1. Compressive strength test

S. pasteurii was cultured in NBU media in order to conduct the compressive strength test of cement mortar.The coarse aggregate samples were divided into 4 groups: bacterial sample (Ba-in water; Ba-in air) and control (in water; in the air) is shown in Fig. 1. The fine aggregate samples were divided into 6 groups: bacterial sample (Ba-in water; Ba-in the air; Ba-Soak in water the next day) and control (in water; in air; Soak in water the next day). Concrete pressure tests were carried out with a 200-ton automatic compression testing machine.

a b

Fig. 1. Cube aggregate: a – in water; b – in the air

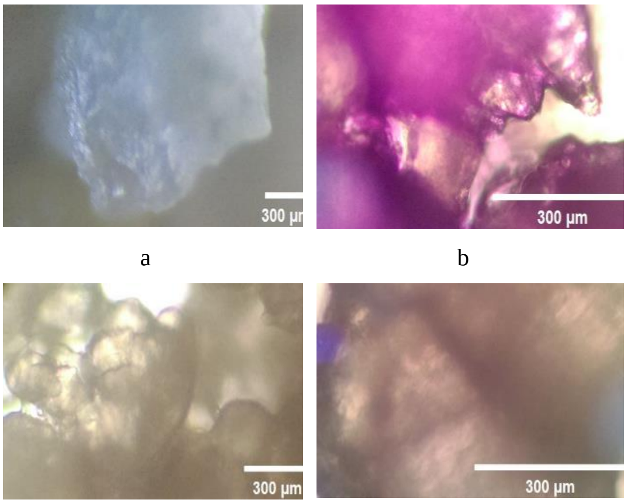
2.4.2. Water absorption test

For an absorption test to determine permeability, five centimeter cement cubes were dried at 105 °C for 24 h and weighed, and the equilibrium between two readings taken at 24-h intervals had to be less than 0.14 %. Following this, the test sample was submerged in water for 24 h. The test sample's weight was measured after removing the sample from the water and patting the surface dry with a towel. The water absorption percentage was equivalent to the penetration ratio.

3. RESULTS

3.1. Compression test

Initially, many crystal shapes were produced by diverse calcium sources [35 – 38]. The best calcium salt to use was determined as shown in Fig. 2.

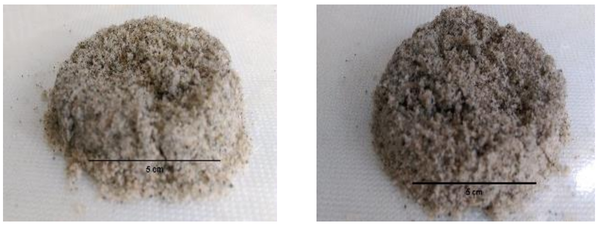


a b

c d

Fig. 2. Calcium carbonate crystallization using different calcium salts: a – $CaCl_2$ 100# crystal; b – $CaCl_2$ 400# crystal; c – $C_6H_{10}CaO_6$ 100# crystal; d – $C_6H_{10}CaO_6$ 400# crystal

The calcium lactate with fine aggregate #200 generated a relatively fine calcium carbonate powder while calcium chloride produced a relatively coarse powder. The picture of the sample in Fig. 3 shows that the calcium lactate group was added, and the density of calcium carbonate formed was relatively high. The results of the compressive strength testing on the samples of the coarse aggregate after 7, 14, and 28 days of curing are shown in Fig. 4. *S. pasteurii*-made coarse aggregates had the highest compressive strength.



a b

Fig. 3. Bacteria with various calcium salts and fine aggregate 100# added: a – $CaCl_2$ thicker density; b – $C_6H_{10}CaO_6$ finer density

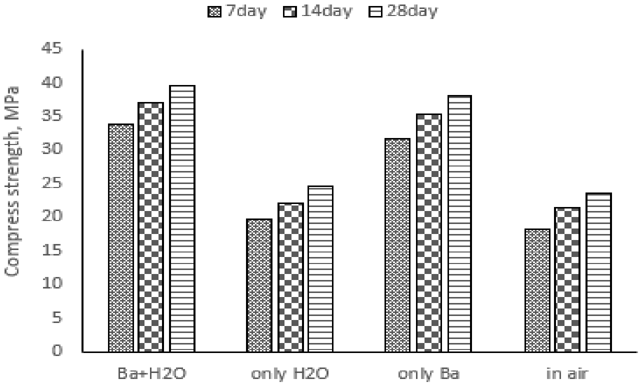


Fig.4. Compressive strength of coarse aggregate, W/C = 0.62. Ba-in water: bacteria cube in water; Ba-in air: bacteria cube in the air; in water: cube in water; in the air: cube in air

Both the hydration of the bacterial culture in the air (38.1 MPa) and the hydration of the aseptic concrete (23.6 MPa) in the air were compared to the hydration of the bacteria test sample in water for 28 days (39.5 MPa). The results demonstrated resistance to compression in medium-strength concrete (MSC). In comparison to the bacterial fine aggregate, the compressive strength of the no-bacterial fine aggregate increased by 36 % after 28 days, illustrative of the high-strength concrete's (HSC) in Fig. 5 compressive opposition.

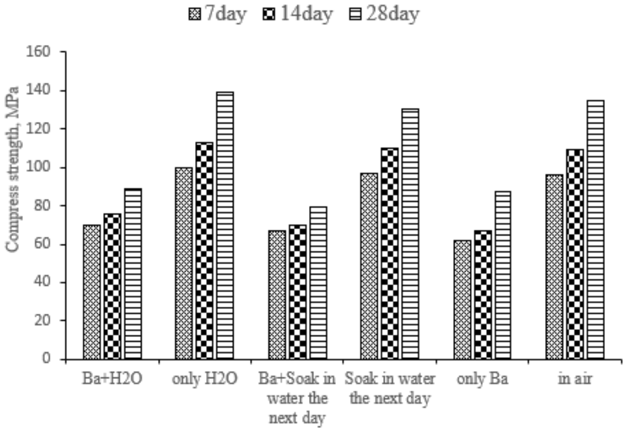
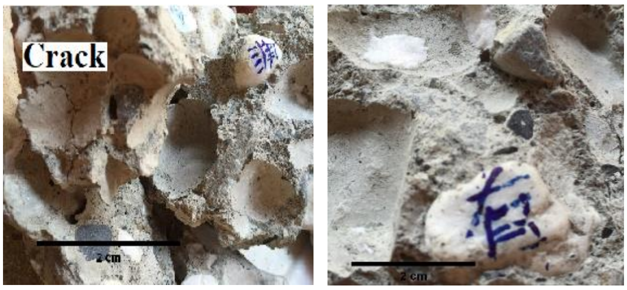


Fig. 5. Compressive strength of fine aggregate, W/C = 0.45. Ba-in water: bacteria cube in water; Ba-in the air: bacteria cube in air; Ba-Soak in water the next day; in water: cube in water; in air: cube in the air; Soak in water the next day

Due to the small pores between the fine aggregate particles, which make it impossible for bacteria to fill the pores, no-bacterial fine aggregate had a 36 % higher compressive strength than the bacterial fine aggregate. The calcium carbonate that bacteria make to fill in large spaces in the coarse aggregate concrete may boost compressive strength. Under a microscope, it may be possible to discern an increase in the compressive strength of the coarse aggregates due to vaterite precipitation caused by *S. pasteurii* [39]. The aseptic cement mortar had significant cracks when the coarse aggregate burst apart, as shown in Fig. 6.



a b

Fig. 6. Microorganisms' effects on pressure cracking: a – absence of bacteria; b – presence of bacteria and the absence of crack

The behavior of the microorganisms filling pores in the matrix of the cement mortar might be responsible for the increase in the compressive strength. The initial stage of concrete curing offered microbial cells good nutrition because the cement mortar was still porous, but because it was a new habitat, it was not favorable for growth. Moreover, when the curing period increased, the cells matured more slowly as they were less active, the pH of the cement was still high, and the curing duration was extended. Vaterite is built up on the cell surface and in the concrete as a result of the interaction of the developing cells with the various cement ions. This caused a large number of matrix pores to clog, which in turn caused the cells to cease obtaining nutrition and oxygen and either died or turned into endospores, which acted as organic fibers and increased the matrix's compressive strength. This explained the behavior of the concrete cubes made with microbial cells that demonstrated increased compressive strength after 28 days [29]. It was thus concluded that the consolidation of internal pores with microbially produced calcium carbonate precipitation was mostly responsible for the cementitious concrete cube's improved compressive strength as shown in Fig. 5. The compression resistance of cubic samples submerged in water was larger than that of samples in air. The hydration reaction is an exothermic reaction, which will explain the porosity of the slurry continues to diminish as a result of the water's ability to absorb a lot of heat, that will increase compressive strength and volume stability.

3.2. Water absorption test

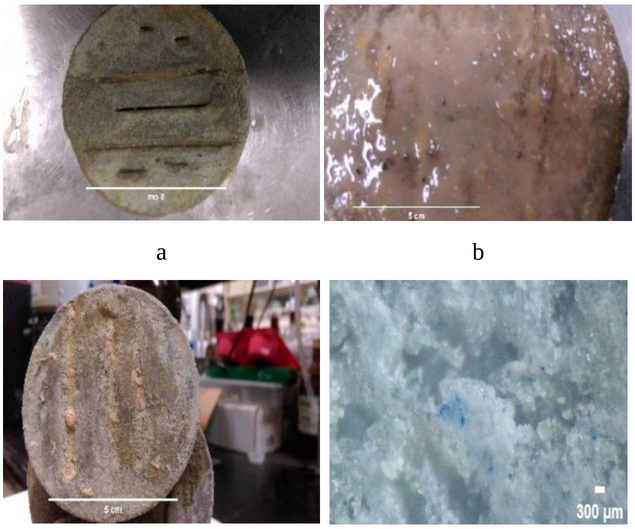
The water absorption of the cement blocks containing 4.8 % bacteria was 1.4 %, compared with 15 % for the control group, indicating that *S. pasteurii* had used calcium lactate to produce finer calcium carbonate to repair the pores and reduce the leakage of cement [40].

3.3. Crack patching

As shown in Fig. 5, *S. pasteurii* [41] filled the concrete gaps. Hydration Mechanism In the cracks of C-S-H cement and C_2S and C_3S limestone, bacterial spores gather to form colloids, fill the gaps, and prevent water infiltration and crack expansion. When these colloids precipitate and dry, they transform into vaterite calcium carbonate. Pasteurella provides calcium carbonate colloids to concrete for repair. Once exposed to water, the bacterial spores that had hibernated in the cracks of the concrete revived and began to repair themselves.

3.4. Microbial sealant effect

We fabricate artificial gaps to efficiently repair concrete cracks with a width of less than 0.1 to 0.2 cm. To fix the damaged structural fractures, a microbial plugging procedure was used. As a microbial sealant, it utilized microbial metabolic processes to create calcium carbonate precipitation [34, 42 – 43]. Wet colloidal bacteria mucilaginum filled and repaired the cracks that ranged from 0.1 to 2.36 mm in size, as shown in Fig. 7.



a b

c d

Fig. 7. Diagram illustrating the calcium carbonate precipitation process for filling the slit: a – artificial slit; b – slit hydration (viscose) after 28 days; c – filling and repairing slit; d – precipitation of calcium carbonate crystals under a 40# microscope

4. CONCLUSIONS

The main objective of this study was to evaluate the efficacy of microbial concrete, which required the direct addition of *S. pasteurii* to the concrete to feed on calcium lactate and convert it to insoluble calcium carbonate, which then filled the fractures as the concrete solidified [44]. A potential remedy for concrete was the bacterium *S. pasteurii*, which is found in concrete mixtures and has the ability to heal itself. The presence of the bacterial cultures did not have any detrimental influence on the compressive strength because they were found to be better than conventional concrete with respect to the parameters of the current study. The development of microbial concrete will pave the way for an inexpensive alternative to concrete sealants, materials that are environmentally friendly, and lengthen the lifespan of buildings. Our results indicated that *S. pasteurii* utilized calcium lactate rather than calcium chloride and exhibited higher activity under various settings [45]. The key conclusions are listed below:

1. The calcium chloride-induced rhombohedral shape was characteristic of the most stable form of calcite [35 – 38], calcium lactate-induced vaterite [39].
2. The compressive strength of the coarse aggregate was 23.6 – 39.5 MPa. Calcium lactate was better than calcium chloride for filling cracks because of its high price and relatively low water absorption.
3. Because the river sand and stone powder in concrete have small pores and fine aggregates have strong hardness, the compressive strength of bacterial fine aggregates, which ranges from 79 to 89 MPa, did not rise as anticipated. The coarse aggregates had large diameters and pores, Greater amounts of bacteria and adhesives filled the pores and crevices of coarse aggregates to increase compressive strength. The compressive strength of no-bacterial fine aggregates increased by 36 % compared with bacterial fine aggregates. Which is newfound. This means that the calcium carbonate formed by the bacteria can fill the small gaps between the aggregate.
4. The cement block containing 4.8 % bacteria absorbed less water (1.4 %) than the control (15 %). This means that the calcium carbonate formed by the *S. pasteurii* can fill the small gaps between the aggregate.
5. The hydration reaction of cement will release heat, water can absorb a lot of heat, increase the rate of hydration reaction, accelerate the hardening rate of cement than in air and increase the compressive strength.
6. When evaluating the development of microbiological concrete, take into account cost-effectiveness as well as the choice of premium concrete sealers that offer materials that are durable for buildings and safe for the environment.
7. A bacterium called *S. pasteurii* was found in the soil. Calcium ions in water can be adsorbed by bacterial surfaces. This can then be converted to calcium carbonate in the presence of carbonates to cover the spaces between rocks and gravel in the soil and form an impermeable barrier. If used properly, it can channel carbon dioxide from the atmosphere into the water, reducing land sinking, reducing the greenhouse effect and slowing global warming.

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