Chapter 7
Experimental Study on Self-Healing of Micro-Cracks in Concrete with Combination of Environmentally Friendly Bacteria

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7.1 Introduction

Concrete is the most extensively used material in building construction globally. However, concrete is weak in tension, ductility, and crack resistance. In addition, cracks can form in concrete structures due to external loads, drying shrinkage, and freeze–thaw action [1, 2]. These cracks provide a pathway for the deterioration of the structures by generating external moisture and corrosion in the reinforcement materials which reduces the life of concrete [3, 4]. Hence, it is important to repair cracks in the structures effectively. The self-healing concrete (SHC) mechanism resembles how the human body naturally repairs wounds over time. The ability of concrete to self-heal cracks would increase the structure's durability and sustainability, extending its service life. Study on autonomous SHC was done in 1994 [5, 6]. SHC is a concrete composite that may automatically cure minor cracks without the need for external diagnosis or human intervention [7]. Cracks in concrete can be filled by materials such as cement or resins. Cement grout is an effective method to fill cracks but it is unable to penetrate fine cracks which are less than a millimeter wide [8]. Therefore, various studies have been carried out globally to overcome this problem leading to the discovery that microbial mineral precipitation acts to heal concrete cracks [9]. Biocementation (bacteria-based method) has been implemented to heal concrete cracks. Biocementation is an environmental friendly and economic process in which bacteria produce the calcium carbonate precipitate at the cracked

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A. Bahrami (ed.), *Sustainable Structures and Buildings*,
https://doi.org/10.1007/978-3-031-46688-5_7

zone [8]. The carbonate precipitate is used in many areas such as the treatment of soil, heavy metal remediation, cementing of sandy soil, etc [10, 11]. Recently, calcium precipitate has been used in many engineering applications and is referred to as the microbial-induced calcite precipitation (MICP) technique. This method is used in concrete cracks to improve concrete durability [12–14].

Compared to conventional concrete, sustainable concrete uses much less energy during manufacture and generates significantly less carbon dioxide. Currently, Portland cement is an extensively utilized material in the manufacture of concrete. Some geographic locations are quickly running out of limestone resources for cement production. Large urban regions are running out of materials to use as aggregates in concrete production. Sustainability necessitates that those in the construction sector consider the whole life cycle of a structure, including construction, maintenance, demolition, and recycling [15–17].

Different types of bacteria (*Bacillus subtilis, Bacillus cereus, Bacillus sphaericus,* etc.) are used for mineral precipitation. This *Bacillus* group has the ability to function in a concrete environment due to the high alkalinity of concrete and the alkaliphilic strains [8]. Through the synthesis of the urease enzyme, ureolytic bacteria precipitate calcium carbonate. The hydrolysis of urea into CO_2 and ammonia, which is catalyzed by this enzyme, raises the pH and increases the concentration of carbonate in the bacterial environment. Sand columns have been used to precipitate calcium carbonate using biodeposition technology [18, 19] and for remediation of cracks in concrete [20]. The MICP technique is a new approach to the remediation of cracks and to protect the serviceability of concrete [1]. *Bacillus subtilis* has been mixed in concrete and was studied for the compressive strength and self-healing properties of concrete. The results revealed that the compressive strength of concrete was noticeably increased and attained the self-healing property due to the continuous precipitation of calcite [21]. Calcite ($CaCO_3$) may be determined by four important variables, including pH, calcium concentration [22], dissolved inorganic carbon concentration, and the existence of nucleation sites [22]. Salmasi and Mostofinejad [23] studied the effects of bacteria on enhancing the strength of cement and mortar using varied concentrations of anaerobic bacteria (*Shewanella* species) and observed strength improvements in their specimens.

A study was conducted to find the effects of different microbes, properties, and efficiency, in addition to an overview of microbial concrete uses [11]. Two types of bacteria (*Sporosarcina pasteurii* and *Bacillus subtilis*) were employed in the study to observe the influence of bacteria on concrete permeability, corrosion, and electrical resistivity in two different mix designs. The results showed decreased water absorption and chloride penetrability and that *Sporosarcina pasteurii* enhances the compressive strength and electrical resistivity in concrete [23]. Researchers concentrated on different ureolyic bacteria similar to *Sporosarcina pasteurii*, *Bacillus sphaericus,* and *Bacillus megaterium*, which are alkali-resistant and have the spore-forming capability to help them live up without nutrients [24–26]. The researchers investigated the impact of calcium acetate, calcium nitrate, calcium chloride, and calcium oxide on MICP by *Bacillus alkalinitrilicus* and *Bacillus subtilis* [27–29]. With and without the inclusion of external calcium sources, the optimum amount to improve the compressive strength was examined utilizing *Bacillus subtilis* [30]. The earlier research clearly demonstrates that bacteria utilize the ureolysis, denitrification, and metabolic conversion of the organic chemical used in microbial concrete as per the MICP processes. The current study proposed to investigate the effect of bacteria (*Bacillus subtilis* and *Bacillus cereus*) on its properties, and self-healing of cracks in concrete compared to conventional concrete.

In the subsequent years, several researchers began to work on this topic. Research on SHC was begun in 2006 [31, 32]. After conducting experimental investigations, the *Bacillus* genus was selected as an ideal SHC therapeutic agent [32].

7.2 Materials

7.2.1 Cement, Aggregates, and Water

Ordinary Portland cement 43 grade was utilized in the study as per Indian standard specifications (IS 8112:1989). The physical properties of cement are presented in Table 7.1. The fine aggregates up to a maximum size of 4.75 mm and coarse aggregates passed through 20 mm and retained on 12.5 mm were used in this study. The specific gravity values of fine and coarse aggregates were 2.66 and 2.74, respectively. Distilled water was utilized for mixing and curing of specimens as well as the preparation of nutrient broth solutions for bacterial species, and casting and curing of microbial samples. The standard M25 grade concrete mixes were prepared.

7.2.2 Selection of Bacteria

By using biomineralization, organisms like *Bacillus* enabled the formation of microbial concrete. In this study, two species were chosen, *Bacillus subtilis* and *Bacillus cereus* obtained from Manidharma Biotech Private Limited, Chennai. These species were gram-positive rod-shape structures and tolerate extreme environmental conditions. It could survive in high alkali conditions in concrete since the formation of $CaCO_3$ signified the presence of bacteria. The culture was kept alive on nutrient agar slants and subcultured on the sterilized medium every 20 days.

Table 7.1 Physical properties of cement

Properties	Values
1. Specific gravity	3.15
2. Consistency (%)	33
3. Initial setting time (min)	87
4. Final setting time (min)	575

7.2.3 Preparation of Bacterial Solution

Bacillus subtilis and *Bacillus cereus* were developed in the Lysogeny broth (LB) medium. LB was nutrition-rich and was primarily used for the growth of bacteria. These species were inoculated in the LB medium for multiplying the growth of bacteria. Then, 12.5 g of LB medium was added to a 250 ml conical flask containing distilled water. LB medium consisted of 10 g of tryptone, 5 g of yeast extract, and 10 g of sodium chloride in 1 L medium. Thereafter, the conical flask was cotton plugged, covered with paper, and a rubber band made this airtight. The solution was sterilized in an autoclave for 10–20 min approximately. The sterilization temperature was maintained at 120 °C for 20 min. The solution was free from contaminants and pure orange in color. The bacteria were added to the media with the help of an inoculation loop. The solution was kept undisturbed for 24 h in the laminar flow chamber. The bacterial solution contained turbidity that indicated the growth of bacteria which was different from the control. After 20 days, it was subcultured in the sterilized medium. The preparation of bacterial solution is displayed in Fig. 7.1.

7.2.4 Preparation of Bacterial Concrete

7.2.4.1 Mix Design

Table 7.2 lists the material requirements for both conventional and bacterial concrete. Four concrete mixtures were chosen for research on the self-healing characteristics of concrete. The proportions of cement, sand, coarse aggregate, and water used in each example were identical. Blend 1 was a representation of nominal concrete with no bacterial broth solution added. In the case of Blend 2, 250 ml of *Bacillus cereus* was added to Blend 1. Similarly, 250 ml of *Bacillus subtilis* was added to Blend 1 to form Blend 3. Finally, 250 ml of a bacterial solution containing *Bacillus subtilis* and *Bacillus cereus* was added to Blend 1, resulting in Blend 4.

Fig. 7.1 Preparation of bacterial solution: (**a**) Subcultured *Bacillus subtilis* bacterial solution, (**b**) Subcultured *Bacillus cereus* bacterial solution

Table 7.2 Mix design of nominal and bacterial concrete

Materials	Conventional concrete (Blend 1)	*Bacillus cereus* with conventional concrete (Blend 2)	*Bacillus subtilis* with conventional concrete (Blend 3)	Combined bacterial with conventional concrete (Blend 4)
Cement (kg)	345	345	345	345
Sand (kg)	750	750	750	750
Coarse aggregate (kg)	1158	1158	1158	1158
Water (m^3)	0.186	0.186	0.186	0.186
Bacterial broth added	Nil	*Bacillus cereus*	*Bacillus subtilis*	*Bacillus subtilis* + *Bacillus cereus*
Bacterial broth solution (ml)	Nil	250	250	250

7.2.4.2 Mixing

Mixing of concrete was done employing a motorized electrical mixer. The required quantities of cement, fine aggregate, and coarse aggregate were weighed and put in uniform layers. The dry mixing was done to get a homogeneous mixture. Then, the estimated quantity of bacterial solution along with water was added and mixed for up to 5 min to obtain the homogeneity. This fresh concrete was used to verify the workability of concrete immediately before casting.

7.2.4.3 Casting

Iron molds of different sizes were used to cast the concrete specimens for the compressive strength test, splitting tensile strength test, and flexural strength test. Initially, the mold was checked for joint movement and lubricated with oil. The prepared fresh concrete was poured into the cube, cylinder, and prism of size 150 mm × 150 mm × 150 mm, 150 mm × 300 mm, and 100 mm × 100 mm × 500 mm, respectively, and compacted manually in three layers. The prepared specimens are depicted in Fig. 7.2.

7.3 Experimental Investigation

In order to determine the slump value, a number of experiments were conducted (ASTM-C143, 2000) on the compressive strength (ASTM-C873, 2000 and ASTM-C943, 2000), splitting tensile strength (ASTM-C496/C496M, 2000), and flexural strength (ASTM-D790, 2000) of different mixes (i.e., conventional and

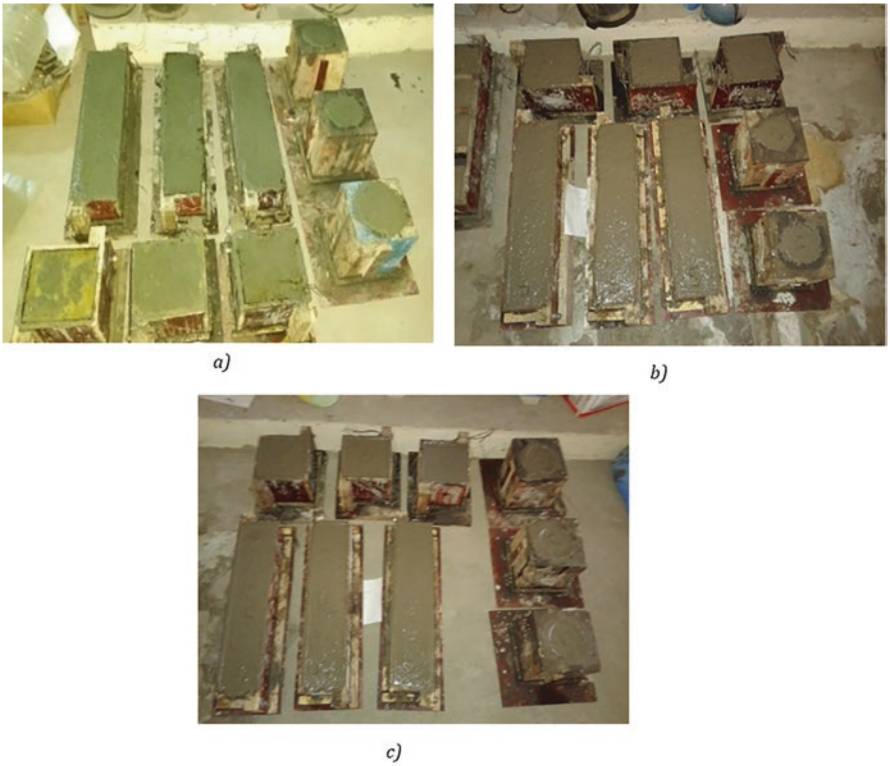

Fig. 7.2 Casting of concrete specimens: (**a**) *Bacillus subtilis* concrete specimens, (**b**) *Bacillus cereus* concrete specimens, (**c**) *Bacillus subtilis* and *Bacillus cereus* concrete specimens

bacteria concrete) with various curing periods of 7, 14, and 28 days [33–37]. Figure 7.3 illustrates the experimental test setups. Three samples for each test were evaluated to ensure the accuracy and repeatability of the results.

7.4 Results and Discussion

7.4.1 Slump Value

The slump cone test was utilized to determine the workability of the concrete mix. True slump, as opposed to shear and collapsible slump, was the desired shape of the slump. The lower the value of the slump, the more workable the mix. Low-slump concrete is stiff, dry, and difficult to work with. Likewise, if there is a significant slump, it can be due to high water content which eventually affects the strength and workability of concrete. According to IS 456:2000, the slump ranges between 50 mm and 100 mm for ordinary reinforced concrete beams and slabs. However, the

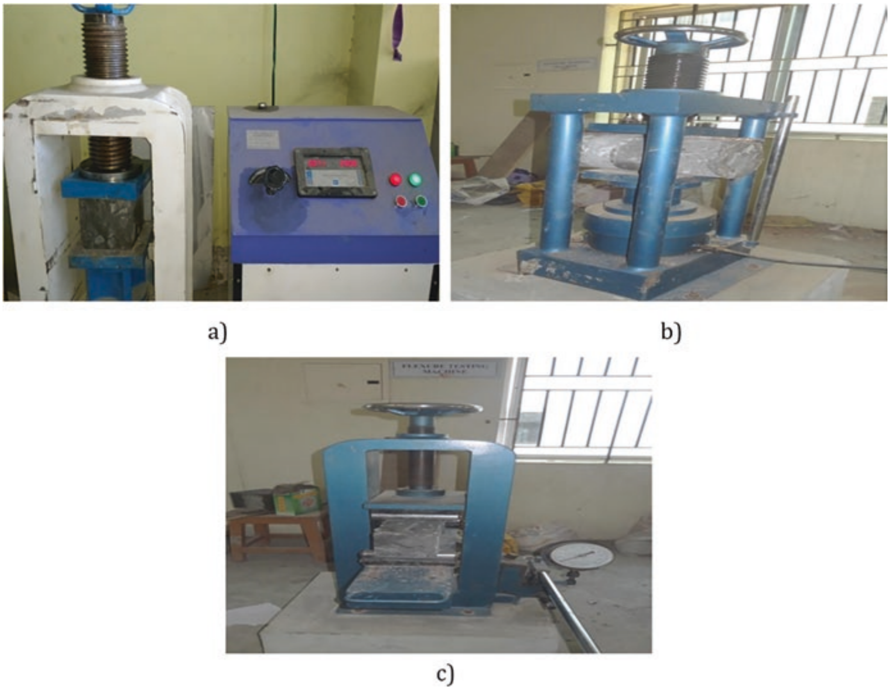

Fig. 7.3 Experimental test setups: (**a**) Compressive strength test, (**b**) Splitting tensile strength test, (**c**) Flexural strength test

Table 7.3 Slump value of different concrete mixes

Concrete mix	Slump value (mm)
Blend 1	90
Blend 2	85
Blend 3	80
Blend 4	70

selected four concrete mixes, Blends 1, 2, 3, and 4, the slump values of 90 mm, 85 mm, 80 mm, and 70 mm, respectively (Table 7.3). According to ASTM-C143, the slump value of concrete used in the original works of beams and slabs is 50–100 mm. Figure 7.4 shows measuring of the slump value.

A 250 ml addition of *Bacillus subtilis* to the nominal concrete mix reduced the slump by 11.11%. However, adding 250 ml of *Bacillus cereus* to the nominal mix reduced the slump value by 5.55%. *Bacillus subtilis* bacterial broth produced a better concrete slump than *Bacillus cereus*. Furthermore, adding 250 ml of bacterial broth, *Bacillus subtilis* and *Bacillus cereus*, to the nominal mix substantially reduced the slump value by 22.22%. Based on the results, the combined action of the selected bacteria created an efficient slump value that can be employed quickly and economically. In the instance of Blend 4, the consistency of fresh concrete, before it set,

Fig. 7.4 Measuring true slump value

was good. Furthermore, the amount of water required was smaller than in the other three scenarios, which could result in a sustainable and cost-effective outcome. The lower the slump, the harder and less workable the concrete would be, but the hardness should not be much less as it would be difficult to construct. Blend 4 had greater strength and durability than other cases, as it is commonly known that the higher the slump, the lower the strength and durability.

7.4.2 Compressive Strength

The compressive strength of concrete is an important parameter to evaluate the concrete performance. The individual and combined effects of both bacteria on the compressive strength of conventional concrete are demonstrated in Fig. 7.5. It is observed that the effect of *Bacillus subtilis* exhibited better results than *Bacillus cereus*, and increased the compressive strength of conventional concrete considerably from 25 MPa to 33 MPa (i.e., 29.8%), whereas *Bacillus cereus* provided a 4% increase in the compressive strength after 28 days of curing.

The combined effect of both bacteria on conventional concrete gave a better compressive strength of 29.7 MPa (i.e., 17.9%) when compared to *Bacillus cereus* blended concrete after 28 days of curing. This obtained value of the compressive strength was slightly lower (i.e., 9.17% decrease) than the compressive strength of Blend 3. From this study, it is evident that *Bacillus subtilis* alone produced better strength than the combination of *Bacillus subtilis* and *Bacillus cereus*. *Bacillus subtilis* remarkably increased the strength of concrete and hastened the healing of micro-cracks in the samples. Figure 7.5 indicates that the strength at 7 days for Blend 3 specimen and other cases varied noticeably, and a comparable difference

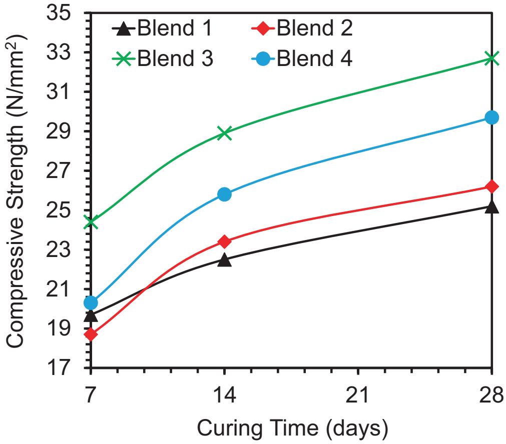

Fig. 7.5 Results of compressive strength test

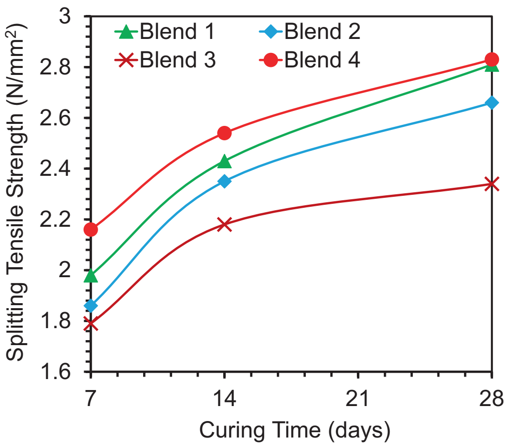

Fig. 7.6 Results of splitting tensile strength test

was witnessed at 28 days as well. The compressive strength is an important factor for bearing greater loads over time; by adding 250 ml *Bacillus subtilis* solution to the standard concrete specimen, the structure's sustainability is improved. In this sense, the use of concrete and the inclusion of reinforcement can be greatly reduced while maintaining the same strength and durability.

7.4.3 Splitting Tensile Strength

Regardless of grade, the concrete's tensile behavior is always poor. However, concrete has some tensile strength that is only substantial when compared to the compressive strength. The current study tested the tensile behavior of concrete for four chosen blend types (Fig. 7.6). The nominal concrete mix (Blend 1) exhibited better splitting tensile strength when compared to individual bacterial broth (Blends 2 and 3). For 28 days, the obtained splitting tensile strengths of Blends 1, 2, 3, and 4 were 2.81 MPa, 2.66 MPa, 2.34 MPa, and 2.83 MPa, respectively. In fact, the addition of *Bacillus subtilis* and *Bacillus cereus* individually reduced the splitting tensile strength of concrete. The overall impact was greater splitting tensile strength. However, the introduction of *Bacillus subtilis* and *Bacillus cereus* eliminated the major difference between Blends 1 and 4. The gap between the maximum splitting tensile strength, Blend 4, and the other three cases appeared to be less by value but not by percentage. The percentage change between Blends 4 and 1 was relatively small (i.e., 0.71%). The percentage variation for Blend 4 and Blend 2 was 6.39%. Similarly, the variation for Blend 3 was significant at 20.94%, which is sustainable. In terms of the concrete strength, *Bacillus subtilis* developed more effective compressive strength while producing lower splitting tensile strength out of all cases.

7.4.4 Flexural Strength

The flexural strength of concrete was determined by doing the flexural strength tests for four different mixtures. The obtained flexural strengths followed the same trend as the splitting tensile strengths (Fig. 7.7). Out of four blends, the bacterial combination of *Bacillus subtilis* and *Bacillus cereus* (Blend 4) demonstrated a good flexural strength value of 4.37 MPa for 28 days. It is seen that Blend 1 had 4.29 MPa flexural strength after 28 days. The increment of the flexural strengths between Blends 1 and 4 was 1.86%, which was not large. However, the flexural strength for 7 days had moderate differences of 3.26 MPa and 3.53 MPa. The individual inclusion of bacterial broth worsened the flexural nature of concrete, as it is evident in Blends 2 and 3 with the strengths of 4.17 MPa and 3.98 MPa.

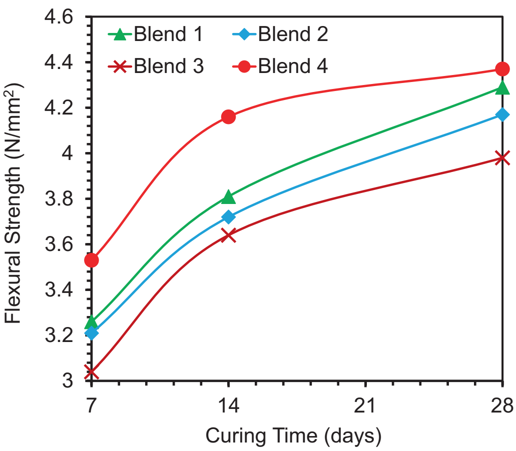

Fig. 7.7 Results of flexural strength test

7.4.5 Self-Healing Characteristics of Combined Bacterial Concrete

Concrete that is capable of self-healing or self-repairing cracks is known as self-healing or self-repair concrete. It not only closes cracks but also partially or completely restores the structure's mechanical properties. Concrete frequently develops surface cracks due to its low tensile strength in comparison to other construction materials. Because they allow the movement of liquids and gases that may contain toxic compounds, these cracks degrade the durability of concrete. If microcracks spread to steel reinforcements, not only concrete but also reinforcements will be vulnerable.

However, SHC is still in its early stages of development, with the current study focusing on healing of tensile cracks in concrete. In this study, the cracks formed in Blend 4, after the compressive strength testing, were observed for its self-healing nature by curing the cracked surface for 7 days. It was seen that the combination of two bacteria, *Bacillus subtilis* and *Bacillus cereus*, acted as a self-healing agent to arrest micro-cracks. When water was provided to bacterial concrete, it infiltrated through the cracks, creating the conditions for bacteria present in concrete to precipitate calcite. Calcite precipitation resulted in filling concrete cracks. Figure 7.8 displays the effect of calcite precipitation filling before and after healing. The figure illustrates a microcrack after performing a compressive strength test on Blend 4. This microcrack signified the failure of the concrete block after it reached its maximum strength. The addition of *Bacillus subtilis* and *Bacillus cereus* automatically cured the microcracks. This study revealed that not only were microcracks healed, but the slump value, splitting tensile strength, and flexural strength of concrete also improved. Thermal stresses and unexpected loads are known to cause the formation of microcracks in structures. However, for such cases, the Blend 4 scenario was the best and most sustainable combination.

Fig. 7.8 Combined bacterial concrete before and after healing

7.5 Conclusions

The objective of this study was to examine healing of concrete microcracks with the addition of bacterial broth to the standard mix concrete. Four concrete mixes, i.e., nominal concrete, *Bacillus cereus* with concrete, *Bacillus subtilis* with concrete, and combined bacteria with concrete were used. The total amount of bacteria added to the nominal mix from Blends 2–4 was 250 ml. The slump cone test, compressive strength test, splitting tensile test, and flexural strength test were performed to determine the mechanical properties of the mixes. The slump values of 90 mm, 85 mm, 80 mm, and 70 mm were obtained. A good slump always results in a lower value, indicating the workability of concrete. However, out of all types, Blend 4 provided an efficient slump value with a 22.22% variance from the nominal mix. In the compressive strength test, out of all selected blends, Blend 3 produced substantial compressive strength with a 29.7% increase above the normal mix. Remarkably, Blend 4 yielded lower compressive strength than Blend 3. Blend 4 exhibited an excellent tensile nature in the splitting tensile test, however, the difference between Blends 4 and 1 was minor. The combined effect of bacteria had little effect on the splitting tensile strength of the mixture. Similar trend was resulted from the flexural strength test. Blend 4 coupled with *Bacillus subtilis* and *Bacillus cereus* healed the microcracks in concrete. When water was added to bacterial concrete, it infiltrated through the crevices, allowing bacteria in concrete to precipitate calcite. Calcite precipitation caused concrete cracks to fill. The chosen SHC blends healed and reduced the need for external intervention to locate and fix internal damage (e.g., cracks), which is a major benefit to the construction industry. In addition, these blends reduce reinforcement corrosion and concrete deterioration while decreasing costs and enhancing the durability. It has the potential to survive for decades or centuries. This is the most significant advantage of SHC. This guarantees that you will not have to replace the concrete surface during its lifetime. SHC requires less upkeep. Cracks in conventional concrete must be filled and sealed. SHC also improved the compressive strength of concrete which is a great advantage to industries while constructing. SHC has the potential to contribute to the infrastructure crisis. This approach can lower the environmental costs of frequent concrete upkeep, demonstrating its usefulness as a sustainable resource. This has social, economic, and environmental benefits, because eliminating or lowering the need for maintenance and/or improving longevity minimizes disruption, as well as costs and material use. With a similar process obtained in this study, the current work can be extended to assess the mechanical properties of concrete for bigger units such as beams, columns, and slabs subjected to a variety of loads.

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