Efficiency of Bacteria-Based Self-Healing Mechanism in Concrete

Sally Shabban[1,a], Nancy Hammad[2,b], Amr ElNemr[3,c*]
and Ibrahim G. Shaaban[4,d]

[1,2,3]German University in Cairo, New Cairo City, Main Entrance El-Tagamoa El-Khames,
Cairo, Egypt

[4]Conference coordinator
[4]School of Computing and Engineering University of West London, St Mary's Road, London W5 5RF

[a]Sally.shaaban@student.guc.edu.eg, [b]nancy.saad@guc.edu.eg, [c,*]amr.elnemr@guc.edu.eg,
[d]ibrahim.shaaban@uwl.ac.uk

Keywords: Bacterial Concrete; Bacillus Subtilis; Calcium Carbonate precipitation; Self-healing

Abstract. Concrete is most widely used as an essential building material in the construction industry all over the globe. Concrete deteriorates over time, and cracks eventually form on its surface for many reasons, such as environmental surroundings and extra. This deterioration and cracks might lead to the ingress of water and chemicals that susceptible steel bars or reinforcements to corrosion. Since this deterioration is inevitable, maintenance and repair are also necessary. This process requires skilled labor and is cost-effective. Thus, researchers suggested alternative techniques to enhance concrete's mechanical properties and search for treatments to be applied to concrete's surface for healing and sealing the cracks by producing calcium carbonate precipitation. Therefore, self-healing concrete was introduced; this method is significant as it's proven environmentally friendly. This research aims to investigate the use of liquid bacteria incorporated in concrete mix and assess whether there would be improvements in the mechanical properties of the bacterial concrete compared to the conventional mix and an autogenous self-healing mix. Two different concentrations of an alkaliphile bacterium called Bacillus Subtilis were incorporated into the concrete mixes to test their ability to repair cracks by producing calcium carbonate and sealing them. This experiment showed a remarkable increase in bacterial concrete's compressive and tensile strengths. A visible partial crack sealing was also observed in specimens containing different concentrations of Bacillus Subtilis. Results also indicate that optimum results were obtained when the bacterial solution of concentration 10^8 cells/ml was incorporated, especially at early ages.

1. Introduction

Concrete dominates the construction sector globally due to its low cost, durability, enhanced compressive strength, thermal mass, and versatility [1-2]. Concrete cracking is a common phenomenon due to its relatively low tensile strength. The durability of concrete is affected by these cracks since they provide an easy path for transporting harmful substances and pave the way for water and oxygen along with other chemicals. Such chemicals affect the reinforcement by corrosion. The causes of concrete cracking are various, and each cause results in cracks that differ in depth, shape, probability of occurrence, and effect on the structure's safety and performance. As a result, it is crucial to control the crack width and to heal the cracks as soon as possible. Self-healing or automatic repair is an effective process of sealing harmful cracks without intervening with any human effort or capital investment [1, 7].

The self-healing mechanism is classified into autogenous and autonomous self-healing. The autogenous self-healing includes only the conventional concrete ingredients, which promotes crack repair due to their specific chemical composition and under certain environmental conditions. The ongoing hydration of clinker minerals or carbonation of calcium hydroxide ($Ca\ (OH)_2$) causes cracks to heal after water exposure. However, it is only limited to small cracks and is only effective when water is available. Thus, this process could be difficult to control [4, 7].

Moreover, the autonomous self-healing process uses external components added to the conventional concrete mix, such as microcapsules or bacteria, to produce CaCO3 and seal the cracks eventually. One promising self-healing technique is inducing bacterial calcite precipitation in concrete mixtures. Biomineralization (Bacteria mineralization) is achieved by decomposing urea and calcium. This decomposition produces calcium carbonate ($CaCO_3$), filling the cracks within concrete. Microbial-induced calcite precipitation (MICP) is a process associated with biological mineralization that can heal cracks up to four times more effectively than autogenous mechanism [8-12]

2. Experimental Program

2.1. Materials used

2.1.1. Bacteria

Calcium carbonate precipitating bacteria from Bacillus species called Bacillus Subtilis was used in this study. Such bacteria can form a layer of calcium carbonate and survive in highly alkaline conditions, making it favorable for the high alkalinity nature of concrete. Bacteria is added as a liquid suspension to facilitate the metabolic process upon cell breakage by any additional load or microcracks [13-15]. In this study, the bacterial sample of Bacillus subtilis was obtained from the faculty of agriculture at Ain Shams University and then was taken and maintained in agar on a Petri plate (1% tryptone, 0.5% yeast extract, 1% NaCl, and 1.5% agar) at 4°C. All glassware and instruments used were previously sterilized to avoid contamination by other species. After that, to grow the bacteria, a culture media of Tryptic soybean broth was used as a nutritious medium to support the growth of the bacteria. This inoculated media was then kept for incubating at 37°C temperature. The concentration of bacteria was determined by finding the optical density using a spectrophotometer. The determinacy of the subtilis absorbance was performed by a qualitative procedure involving the changes in OD_{600} (optical density measurement at 600 nm) of spore suspensions during germination. The solution of the bacteria was monitored every 24 hours to obtain the OD_{600}. After 48 hours, the bacteria culture showed 0.65 optical density that reached 1.1 OD_{600}. At elevated pH (10.5), the growth profile of bacillus subtilis was the fastest indicating the best alkali-resistant behavior. Sub-culturing of pure bacterial strain into young exponential cells is performed to convert the optical density into CFU/ mL. OD and CFU/mL are measured, and repeated measurements are done till getting 0.1 OD and the corresponding CFU/mL. A linearized standard curve was plotted for conversion. The bacterial concentration was kept at 10^8 cells/ml and the other at 10^5 cells/ml. The characteristics of pure culture for Bacillus subtilis are given in Table 1

Table 1: Characteristics of Bacillus subtilis

Characteristics of Bacillus subtilis	Value
Growth medium	3
Incubation time	24 hours
Subculture	30 days
Gram stain	Positive
shape	Rod
Oxygen demand	Facultative

2.1.2. Ordinary Portland cement (OPC)

OPC of grade 42.5, according to ES 4756-1 / 2013 [16], was used in this research, with the following physical properties illustrated in Table 2.

Table 2: Physical properties of the used OPC

Characteristics of the used OPC	Value
Fineness	4066 cm^2/gm
Consistency	27.7%
Initial setting time	60 minutes
Final setting time	145 minutes
Compressive strength	MPa
3 days	22
7 days	37
28 days	42.5
Specific gravity	3.15

2.1.3. Aggregate

Conventional crushed limestone aggregate with a 20 mm maximum nominal and 10 mm minimum size was used, and natural sand of 4.75 mm maximum size was added. The specific gravity of coarse and fine aggregate were 2.71 and 2.65, respectively.

2.1.4. Admixture

The superplasticizer (Sikament NN) is a highly effective dual-action liquid that produces free-flowing concrete. It is a substantial water-reducing agent to promote high early and ultimate strengths. Moreover, Sikament NN is compatible with all types of Portland cement and has a recommended dosage of 0.8 – 3 % by weight of cement. This superplasticizer complies with ASTM C494 Type F and has a specific gravity of 1.05.

2.2. Preparation of test specimens

Concrete mixtures (M-0, M-1, and M-2) were prepared with different concentrations of Bacillus subtilis. The cell concentration was determined from the generated bacterial growth curve by observing optical density at 600 nm. The control mix (M-0) was cast without the addition of microbes. The second (M-1) and the third mix (M-2) had different concentrations of 10^5 and 10^8 cells/ml, respectively. The water-cement ratio was 0.35 for all mixes. The quantity of superplasticizers is maintained in the three mixes. Mix proportions are provided in Table 3.

For each mix, twelve cubes (150 mm) and six cylinders (150 mm (D) x 300 mm (H)) mm were cast to evaluate the effect of bacteria on the compressive strength after 7, 28, 56, and 90 days of curing, and the splitting strength after 28 days. For healing monitoring, visualized crack images were provided. Specimens were carefully loaded up to 85% of their ultimate strength (cracking formation) and then unloaded. The pre-cracked samples were re-immersed in water for curing to evaluate the regain of strength. The evaluation methodology is proposed based on the literature [17-20]. An average of three values was considered for each age.

Table 3: Concrete mix proportions

Mix	Cement (Kg/m^3)	Water (Kg/m^3)	Fine Aggregate (Kg/m^3)	Coarse Aggregate (Kg/m^3)	Admixture (L/m^3))	Bacteria Concentration (cells/mL)
M-0	400	140	750	1160	4	-
M-1	400	140	750	1160	4	10^5
M-2	400	140	750	1160	4	10^8

3. Results and Discussion

3.1. Compressive strength

The efficiency of self-healing was mechanically investigated by identifying the regain in the compressive strength of specimens subjected to predefined preloading equals 80 % of the maximum compressive strength (*fc*) at 28 days. This preloading aims to produce internal micro-cracks, then the pre-cracked specimens were immersed and cured in water for 28, 56, and 90 days. The cracking level at 80% of each mixture capacity is illustrated in Figure 1, corresponding to the control mix after 7, 28, 56, and 90 days. The percentage of strength regain for M-2 (concentration 10^8 cells/ml) appeared to be much greater than the lower concentration M-1 (concentration 10^5 cells/ml); however, they have almost the same regain percentages (40%) when tested after 90 days. These findings align with the regain percentages reported by Nguyen and Khushnood et al. [11, 21], who studied the effect of Bacillus subtilis on compressive strength at different ages.

It was revealed that the strength regains decreases with the increase of the pre-cracking age for the two different bacteria concentrations. This behavior may be attributed to two possible phenomena. The pore structure of concrete becomes denser at later age which negatively influences the activity of bacterial spores in the composite. The second reason is the reduced autogenous healing effect due to the almost completion of the hydration process.

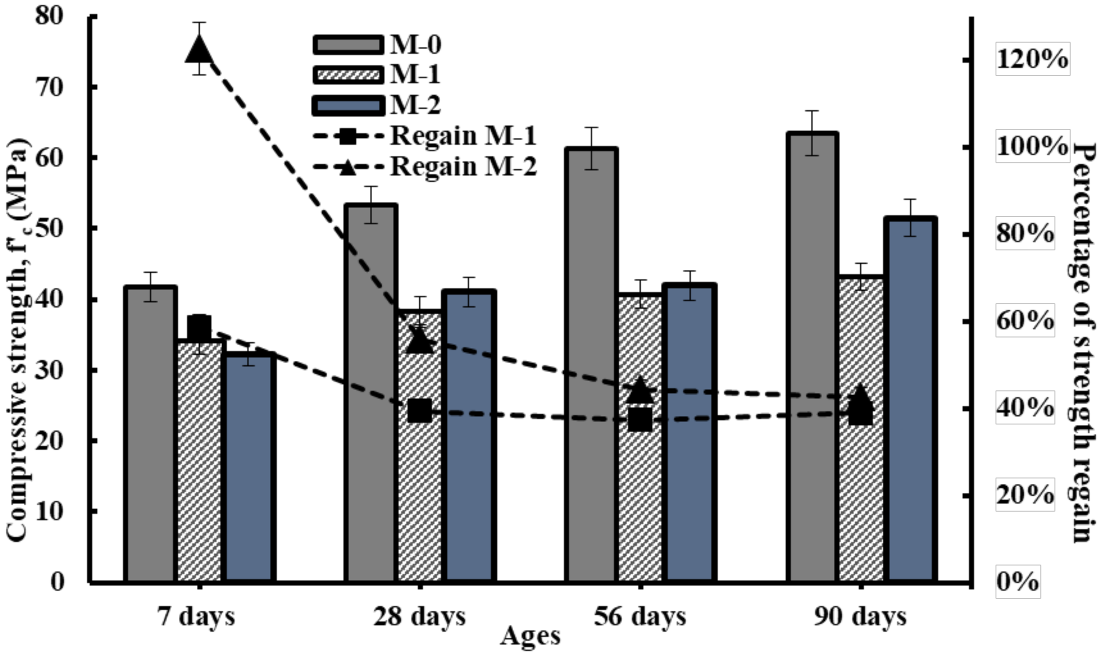


Figure 1: Compressive strength of the control mix (M-0) and the compressive strength regain for specimens pre-cracked (M-1 & M-2) at 7 and 28 days

3.2. Splitting tensile strength

The splitting tensile strength of the control mix and the evolution of the tensile strength of the pre-cracked specimens after 28 days are illustrated in Figure 2. It appears that the incorporation of bacteria led to an increase in the tensile strength as the regained strength, especially of M-2, gave a higher value than the reference tensile strength (M-0) with a percentage of regain of 132%. The percentage of regain for the lower bacteria concentration was reported to be 75%. The regain percentages are compatible with the literature results conducted by Pal et al. [22] and Khushnood et al. [23]. The increase in pore size modification results from microbiologically induced calcium carbonate precipitation. These results are consistent with those of the compressive strength.

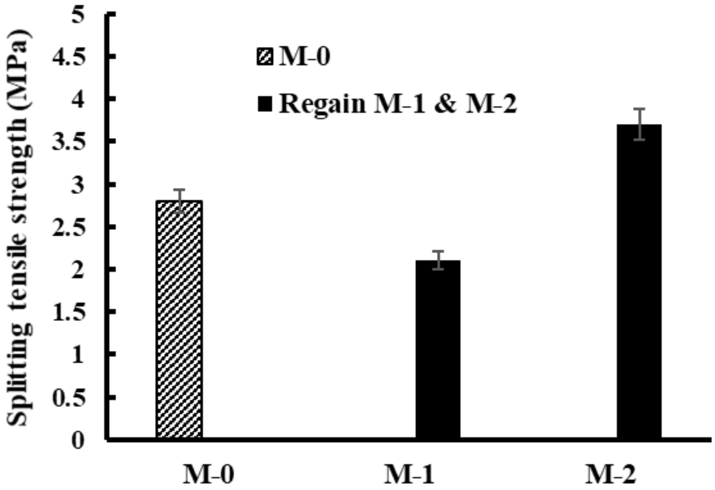


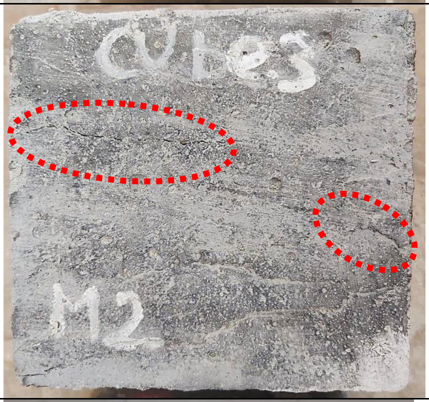
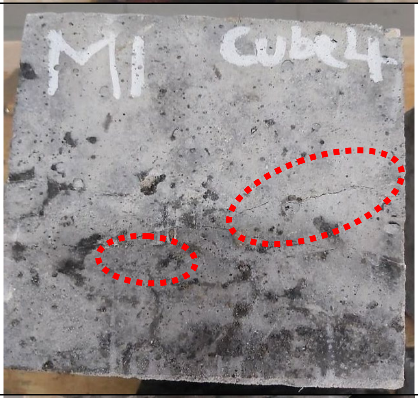
Figure 2: Splitting tensile strength of the control mix (M-0) and the tensile strength regained for specimens pre-cracked (M-1 & M-2) at 28 days

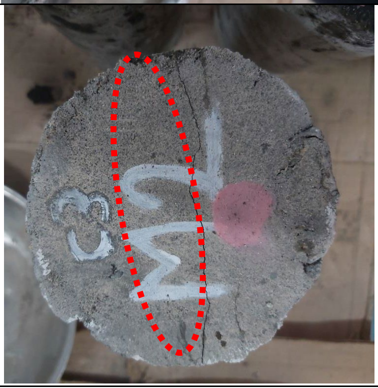
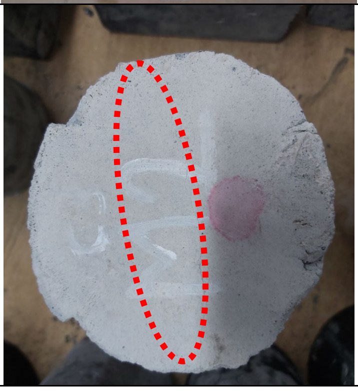
3.3. Visualizing crack healing

The efficiency of crack healing of pre-cracked specimens, including bacteria, is shown in Table 4 after immersion in tap water for 28 days from the predefined cracking times (7 and 28 days). Concrete specimens appeared to have significant crack-healing due to the precipitation products. Autogenous and autonomous bacterial self-healing are the leading causes of this healing. The primary role of bacteria is to precipitate calcium carbonate that fills in the crack and promotes crack healing. This action is performed by producing urease that catalyzes urea ($CO(NH_2)_2$) into ammonium (NH_4) and carbonate (CO_3^{2-}). The Ca^{2+} ions react with CO_3^{2-} prime to precipitation of $CaCO_3$ at the cell wall of the bacteria which acts as a nucleation site, as illustrated in the following equations [7, 24]. Locations of $CaCO_3$ crystals depend on the saturation of calcite and the concentration of Ca^{2+}, CO_3^{2-} in the solution [25].

$$CO(NH_2)_2 + H_2O \rightarrow NH_2\,COOH + NH_3$$
$$NH_2COOH + H_2O \rightarrow NH_3 + H_2CO_3$$
$$H_2CO_3 \leftrightarrow HCO^{3-} + H^+$$
$$2NH_3 + 2H_2O \leftrightarrow 2NH^{4-} + 2OH^-$$
$$HCO^{3-} + H^+ + 2NH_4^+ + 2OH^- \leftrightarrow CO_3^{2-} + 2NH_4+ + 2H_2O$$
$$Ca^{2+} + Cell \rightarrow Cell\text{-}Ca^{2+}$$
$$Cell\text{-}Ca^{2+} + CO_3^{2-} \rightarrow Cell\text{-}CaCO_3\downarrow$$

Table 4: Images showing the evolution of cracks after 28 days of water immersion

	Before healing	After healing
M-1 (pre-cracked at 7 days)		
M-2 (pre-cracked at 7 days)		
M-1 (pre-cracked at 28 days)		
M-2 (pre-cracked at 28 days)		

M-1 (pre-cracked at 28 days)		
M-2 (pre-cracked at 28 days)		

Conclusions

The study includes the use of bacterial self-healing, which is considered an autonomic technique. The experimental program consists of three mixes, one as a reference and another with two different concentrations of 10^5 and 10^8 cells/ml. Twelve cubes and six cylinders for each mix were cast to evaluate the cubes and cylinders' compressive and split tensile strengths. An increase in the compressive strength of the pre-cracked specimens is realized for both bacteria concentrations after 28 days of water curing. The splitting tensile strength of the pre-cracked specimens improved by 75% and 132% for the bacteria concentration of 10^5 and 10^8 cells/ml respectively. Visualized crack healing is provided due to bacterial activity in precipitating calcium carbonate. The concentration of bacteria and aging are the main parameters influencing self-healing properties. It is recommended to furtherly studied the influence of other cement types with bacteria admixture and their healing action. Moreover, the influence of bacteria on the transport properties of concrete is recommended

References

[1] A. Danish, M. Mosaberpanah, & M. Usama Salim. Past and present techniques of self-healing in cementitious materials: A critical review on the efficiency of implemented treatments. Journal of Materials Research and Technology. 9(3) (2020) 6883-6899. https: //doi.org/10.1016/j.jmrt.2020.04.053.

[2] Z. Li, C. Leung, & Y. Xi. Structural renovation in concrete. (2014). CRCPress.

[3] P.K. Jogi, & T.V.S. Vara, Lakshmi. Self healing concrete based on different bacteria: A review. Materials Today: Proceedings. 43 (2021) 1246-1252.

[4] M. Wu, B. Johannesson, & M. Geiker. A review: Self-healing in cementitious materials and engineered cementitious composite as a self-healing material, Construction and Building Materials. 28 (2012) 571-583.

[5] M. Nodehi, T. Ozbakkaloglu, & A. Gholampour. A systematic review of bacteria-based self-healing concrete: Biomineralization, mechanical, and durability properties. Journal of Building Engineering. 49 (2022) 104038. https://doi:10.1016/j.jobe.2022.104038

[6] Qureshi, T. & A. Al-Tabbaa. Self-Healing Concrete and Cementitious Materials. Advanced Functional Materials, (2020) doi: 10.5772/intechopen.92349

[7] T. Talaiekhozan, A. Keyvanfar, A. Shafaghat, R. Andalib, M. Z. Abd Majid, M. A. Fulazzaky, R. M. Zin, C. T. Lee, M. W, Hussin, N. Hamzah, N. F. Marwar, & H. I. Haidar, H. I. A Review of self –healing concrete research development. Journal of environmental treatment techniques. 2(1) (2014) 1-11.

[8] K. Van Tittelboom, N. De Belie, & W. De Muynck, W. Verstraete. Use of bacteria to repair cracks in concrete. Cement and Concrete Research. 40 (2010) 157–166.

[9] J. Zhang, Y. Liu, T. Feng, M. Zhou, L. Zhao, A. Zhou, & Z. Li. Immobilizing bacteria in expanded perlite for the crack self-healing in concrete, Construction and Building Materials. 148 (2017) 610-617 https://doi.org/10.1016/j.conbuildmat.2017.05.021.

[10] Y. C. Erşan, F. B. Da Silva, N. Boon, W. Verstraete, & N. De Belie. Screening of bacteria and concrete compatible protection materials, Construction and Building Materials. 88 (2015) 196–203.

[11] T. H. Nguyen, E. Ghorbel, H. Fares, & A. Cousture. Bacterial self-healing of concrete and durability assessment. Cement and Concrete Composites. 104 (2019) 103340. https://doi.org/10.1016/j.cemconcomp.2019.103340

[12] N. Hosseini Balam, D. Mostofinejad, & M. Eftekhar. Effects of bacterial remediation on compressive strength, water absorption, and chloride permeability of lightweight aggregate concrete, Construction and Building Materials. 145 (2017) 107–116.

[13] N. Schwantes-Cezario, L. P. Medeiros, A. G. De Oliveira, G. Nakazato, R. Katsuko Takayama Kobayashi, & B. M. Toralles. Bioprecipitation of calcium carbonate induced by Bacillus subtilis isolated in Brazil. Int. Biodeterior. Biodegrad. 123 (2017) 200–205.

[14] R. Pei, J. Liu, Wang, S. & M. Yang. Use of bacterial cell walls to improve the mechanical performance of concrete, Cement and Concrete Composites. 39 (2013) 122-130 https://doi.org/10.1016/j.cemconcomp.2013.03.024.

[15] F. Nosouhian, F., Mostofinejad, D., Hasheminejad, H. Concrete durability improvement in a sulfate environment using bacteria, Journal of Materials in Civil Engineering. 28(1) (2015) 04015064 DOI: 10.1061/(ASCE)MT.1943-5533.0001337.

[16] ES 4756-1. (2013). Cement Part :(1) Composition, Specifications and Conformity Criteria for Common Cements.

[17] R. Siddique, V. Nanda, E. Kadri, M.I. Khan, M. Singh, A. Rajor, Influence of bacteria on compressive strength and permeation properties of concrete made with cement baghouse filter dust, Constr. Build. Mater. 106 (2016) 461–469.

[18] N. Chahal, R. Siddique, A. Rajor, Influence of bacteria on the compressive strength, water absorption and rapid chloride permeability of concrete incorporating silica fume, Constr. Build. Mater. 37 (2012) 645–651.

[19] R. Siddique, K. Singh, M. Singh, V. Corinaldesi, A. Rajor, Properties of bacterial rice husk ash concrete, Constr. Build. Mater. 121 (2016) 112–119.

[20] W. Khaliq, MB Ehsan, Crack healing in concrete using various bio influenced selfhealing techniques, Constr. Build. Mater. 102 (2016) 349–357.

[21] R. A. Khushnood, S. Ud din, N. Shaheen, S. Ahmad, F. Zarrar, F. Bio-inspired self-healing cementitious mortar using Bacillus subtilis immobilized on nano-/micro-additives. Journal of Intelligent Material Systems and Structures. 30(1) (2019) 3-15. doi: 10.1177/1045389X18806401

[22] S. Pal, I. Mohanty, & I. Panda. Self-healing conventional concrete using bacteria.Recent Developments in sustainable Infrastructurs. 75 (2021) 441-451.

[23] R. A. Khushnood, Z. A. Qureshi, N. Shaheen, & S. Ali. Bio-mineralized self-healing recycled aggregate concrete for sustainable infrastructure. Science of the Total Environment. 702 (2020) 135007. https://doi.org/10.1016/j.scitotenv.2019.1350070048-969

[24] Luhar, S., & Gourav, S. A. (2015). Review paper on self healing concrete. J. Civ. Eng. Res., 5, 53.

[25] Edvardsen, C. (1999). Water permeability and autogenous healing of cracks in concrete. ACI Materials Journal-American Concrete Institute, 96(4), 448-454