

Online ISSN

3007-3197

Print ISSN

3007-3189

<http://amresearchreview.com/index.php/Journal/about>

Enhancing Concrete Performance: An Experimental Study on the Influence of Escherichia coli Bacteria on Split Tensile Strength

Abu Zar*

Department of Civil Engineering, Aror University of Art, Architecture, Design and Heritage Sukkur, Sindh, Pakistan. Email: abuzar.faculty@aror.edu.pk

Abdul Manan

Department of Civil Engineering, Aror University of Art, Architecture, Design and Heritage Sukkur, Sindh, Pakistan.

Sadaquat Hussain

Department of Civil Engineering, Aror University of Art, Architecture, Design and Heritage Sukkur, Sindh, Pakistan.

Salman Ali

Department of Construction Materials and Technologies, Budapest University of Technology and Economics

Fahad Ali Sheikh

Department of Civil Engineering, Mehran University of Engineering and Technology, Jamshoro, Sindh, Pakistan

Abstract

Concrete plays an essential part in the construction industry universally, but its tendency to crack and its inadequate tensile strength often leads to the durability issues with time. The strength of concrete directly affects the overall life of a structure, and one of the most common issues affecting its strength is the formation of cracks. Cracks can form due to several factors, including lower tensile strength, a high water-cement ratio, shrinkage, or excessive settlement of soil. Conventional repair methods widely used, tend to be expensive, time-consuming, and often require frequent maintenance. This study reports the outcomes of an experimental analysis carried out to evaluate the influence of self-healing agent such as Escherichia coli (E. Coli) bacteria, on split tensile strength properties of concrete. Cylindrical samples of 100mm x 200mm size were prepared using M30 grade mix design with two groups of 4% and 5% bacterial solution along with 5% calcium lactate as a nutrient. After standard curing for 7 and 14 days, half of the samples were tested directly for split tensile strength, while the remaining bacterial concrete samples were cracked using a Universal Testing Machine (UTM) and then cured again to see their healing and strength regaining. The results showed that the concrete samples with 4% bacterial solution exceeded the strength of normal concrete by 14.41% and 18.63% after 7 and 14 days of curing respectively. On the other hand, the 5% bacterial concrete sample showed improved strength compared to normal concrete. After cracks were introduced, the bacterial concrete samples began to regain strength over time as they continued to cure.

Online ISSN

3007-3197

Print ISSN

3007-3189

<http://amresearchreview.com/index.php/Journal/about>

The sample with 4% bacterial solution regained about 30% of its strength after 7 days and increased to 35.4% after 14 days. Similarly, the sample with 5% bacterial solution recovered 27% of the strength at 7 days and 36.4% after 14 days. While the initial healing wasn't significant, the results clearly showed that the longer the concrete was allowed to cure, the better was the strength regain. These findings support the potential of bacterial concrete as an innovative, low-maintenance and environment friendly solution that can enhance the durability and lifespan of concrete infrastructure.

KEYWORDS: Self-Healing Concrete, Bacterial Concrete, Escherichia Coli (E. coli), Crack Healing In Concrete, Split Tensile Strength, Calcium Carbonate Precipitation

INTRODUCTION

Concrete has been regarded as one of the most popular construction materials that has been used everywhere in the world and can be deemed as indestructible due to their longer service life as compared to the most constructional products [1-4]. Concrete is a composite material, which is formed by the proper mixing of various materials including cement, fine and coarse aggregates, water, and chemical admixtures if necessary. When concrete is mixed with ordinary Portland cement, the blend forms a fluid slurry that can be poured and shaped as needed [5-7]. Concrete, a brittle material being strong in compression but weak in tension, meaning that it will sustain high compressive loads but will fail under the tensile loads. To overcome this problem, reinforced cement concrete (RCC) is used, where steel bars are added to the concrete structures that increases its capacity to bear tensile stresses. Globally, around 10 km³ of Portland cement concrete is produced annually, much of which goes into constructing RCC structures [8]. The quality of concrete is critical to the stability of RCC structures, the concrete mix design must be prepared with precision. One of the biggest challenges in RCC structures is cracking, which, if left untreated, allow water and salts to seep in, leading to corrosion of steel reinforcement and, finally, structural failure. Conventional methods to treat cracks such as cement mortar, epoxy injections, or latex treatments are expensive, time-consuming, harmful and often not durable. In order to address this problem, this study presents a self-repairing concrete concept designed to increase the sustainability of concrete and lessen the need for maintenance. This groundbreaking material has the capability to autonomously mend cracks without requiring any external assistance. By integrating microbial bacteria, specifically Escherichia coli (E. coli), along with a nutrient source of calcium lactate, the concrete acquires the ability to biologically self-heal. This kind of concrete is commonly recognised as "Bacterial Concrete." The idea of bacterial concrete was introduced by V. Ramakrishnan (2001), and it can serve as an effective solution for this issue [9]. The self-healing properties of concrete that utilize bacteria rely on a chemical reaction where the Bacillus genus of bacteria continually forms calcium carbonate (CaCO₃), commonly known as calcite. This process is scientifically acknowledged as Microbiologically Induced Calcite Precipitation (MICP), which is a natural and environmentally friendly occurrence that does not lead to pollution. MICP falls under the larger scientific category of biomineralization, which concerns the creation of minerals by living organisms. In the context of self-repairing or bacterial concrete, the bacteria remain dormant until cracks develop; once fissures appear in the concrete, the bacteria become active [10]. When a crack has been formed and water has penetrated, the bacteria become active and the healing process starts. The active bacteria, when exposed to moisture, form a thick and very

Online ISSN

3007-3197

Print ISSN

3007-3189

<http://amresearchreview.com/index.php/Journal/about>

impermeable coating of calcium carbonate which virtually blocks the cracks and makes the concrete reliable again [10]. The chemical process involved in the formation of healing agent calcium carbonate, is the result of the reaction of CO₂ with calcium hydroxide available in the concrete.

These reactions occur both through microbial metabolic activity and autogenous healing, contributing to effective crack closure. This procedure produces effective autonomous fracture healing based on microorganisms. Meera C.M et al. (2016) discovered that when *Bacillus subtilis* JC3 solution with a concentration of 10⁵ cells/ml was added to concrete mixture, it improved the strength and durability of the concrete by 42% increase in compressive strength and a 63% increase in split tensile strength over conventional concrete. The bacteria also decrease the porosity and enhance the resistance of the concrete to acid and chloride attacks by precipitating calcium carbonate [11]. Pachaivannan et al. (2020) noted that when *Bacillus subtilis* was mixed in concrete mixture, the performance of the concrete mix was observed to have improved noticeably, as opposed to the conventional concrete which experienced a significant increase of up to 19.5% in compressive strength, 17.8% in tensile strength, and 14.2% in flexural strength [12]. These additions were mainly because of the development of calcium carbonate that assisted in filling the pores and decreasing the absorption and permeability of water in concrete. Additionally, the concrete became more flexible and durable, with a ductility factor of 4.92 compared to 4.02 in conventional concrete, and a 26.2% reduction in stiffness [12]. The principal objective of this research was to investigate the effects of inclusion of bacteria on the strength characteristics concrete, specially before and after healing of cracks. The aim of this research was to explore the potential of *E. coli* as a biological agent for promoting self-repair in concrete and enhancing its mechanical performance and durability.

EXPERIMENTAL STUDY

Materials and Properties

The bio-concrete mix consists of OPC Grade 33 (CEM I) cement, natural sand as fine aggregate, coarse aggregate derived from crushed stone or gravel, sourced from the Nooriabad region of Pakistan and normal tap water. To check the quality of the building materials, various tests were performed on aggregates and cement in the laboratory based on ASTM. The fineness modulus of the aggregates was calculated in accordance with ASTM C136. The loose and compacted unit weights were measured following ASTM C29, while the specific gravity and water absorption were evaluated based on ASTM C127 and ASTM C128-15, respectively. The properties of the building materials are summarised in *Table 1* and images taking during test are shown in *Fig. 1*.

Online ISSN

3007-3197

Print ISSN

3007-3189

<http://amresearchreview.com/index.php/Journal/about>

Table 1: Building Materials Properties tested in laboratory

Properties	Fine Aggregate	Coarse Aggregate	Cement
Fineness Modulus	2.846	4.3	
Loose State Unit Weight	1662.75 kg/m ³	1349.07 kg/m ³	
Compacted State Unit Weight	1781.81 kg/m ³	1440.56 kg/m ³	
Specific Gravity	-	2.69	
Water Absorption	-	1.54%	
Consistency			0.33
Initial Setting Time			45 minutes
Final Setting Time			400 minutes



Fig. 1 (a) Specific Gravity Test (b) Water Absorption Test (c) Fineness Modulus Test of coarse and fine aggregate

In this study, *Escherichia coli* (*E. coli*) was employed as the biological agent for self-healing purposes, targeting a final concentration of 10^5 cells/mL. The bacterial culture was grown to an appropriate density, measured at an optical density of 600 nm (OD_{600}), and subsequently diluted in a nutrient-rich feed broth. To prepare the working solution, 0.1 mL of *E. coli* culture at OD_{600} was aseptically introduced into 1 liter of the feed broth shown in **Fig. 2**. The composition of the broth was standardized and consisted of the following constituents: Tryptone – 0.3 g/100 mL, Yeast extract – 0.5 g/100 mL. The bacterial culture was initially incubated under controlled conditions for 48 hours to reach a nominal concentration suitable for downstream applications. After incubation, the culture was diluted according to a predetermined protocol to achieve the desired cell concentration in the final solution. All steps, including media preparation, culture incubation, and dilution, were conducted in accordance with standard operating procedures to ensure accuracy, sterility, and reproducibility of the bacterial solution used for self-healing experiments.

Online ISSN

3007-3197

Print ISSN

3007-3189

<http://amresearchreview.com/index.php/Journal/about>



Fig. 2 (a) E. coli Incubation sample (b) E. Coli Bacterial Solution

Sample preparation and casting

Standard cylindrical samples of 100 mm by 200 mm size were used for specimen preparation in **Fig. 3**. A total of 30 samples were prepared using M30 grade concrete with 0.57 Water-cement ratio. Out of these, 6 samples were prepared using conventional concrete, while the remaining 24 samples were bacterial concrete, incorporating 4% and 5% bacterial concentrations. The mix proportion of 1:2.06:3.03 was adopted based on ASTM standards. Samples named with NC consist of no bacteria and BC consist of bacterial solution. The material quantities were determined using the density formula.

The material quantities were first calculated for a single specimen, and subsequently, only the bacterial solution and calcium lactate (5% of cement weight) were varied for bacterial concrete mixes. Since fine and coarse aggregates are porous, they tend to absorb water. Therefore, a water absorption factor of 1.54% was added to the total water content to compensate for the absorption of both types of aggregates. The water-cement ratio remained constant at 0.57 for all mixes. The bacterial concentration (4% and 5%) was calculated based on the water content, while calcium lactate (5%) was calculated as a percentage of the cement content were summarized in **Table 2**. The concrete mixing was carried out using a mechanical mixer. After a few minutes of initial mixing, the calculated quantities of calcium lactate and bacterial solution was added to the mix. Slump test was performed once the final mixture was ready, to assess workability. The slump test value recorded was 170 mm for conventional concrete and 160 mm for bacterial concrete, representing good workability for both types. After the slump test, the concrete was put into the cylindrical samples in three layers, each layer was compacted using 25 blows of a tamping rod. The surface of the specimens was then finished using trowel and samples were set aside for initial setting. One day later, the samples were taken out of moulds and put in curing tank for curing.

Table 2: Calculated Material Quantity

Mix	Samples	Bacteria %	Cement (kg)	Water (kg)	Fine Agg: (kg)	Coarse Agg: (kg)	Bacterial Sol: (L)	Calcium Lactate (gm)
Mix 1	Normal Concrete	0	0.742	0.422	1.529	2.248	0	0
Mix 2	Bacterial Concrete	4	0.742	0.422	1.529	2.248	18.7	37
Mix 3	Concrete	5	0.742	0.422	1.529	2.248	23.34	37

Mix 1: Conventional concrete with no bacterial solution

Mix 2: Bacterial Concrete with 4% bacterial solution

Mix 3: Bacterial Concrete with 5% bacterial solution



Fig. 3 (a) Casted samples immediately after moulding. (b) Samples prepared and ready for the curing process.

RESULTS AND DISCUSSION

This chapter presents and discusses the results of tests performed on concrete samples after specific curing periods. The aim was to study the effect of *E. coli* bacteria on the strength of the concrete specimens and to compare these results with those of conventional concrete. The samples were put under UTM to check for the split tensile strength of prepared samples.

Split Tensile Strength Test

Tensile strength is the ability of concrete to resist tensile loads, it is known that concrete is not a ductile material, it is very weak in tension. The tensile strength of concrete is considered as 10% of its compressive strength. Samples were tested for strength at 7 and 14 days of curing period. **Fig. 4.** Shows Concrete cylinder placed in the Universal Testing Machine (UTM) during testing, clearly showing the cracking and failure of the specimen. The following formula is used to calculate the tensile strength.

Online ISSN

3007-3197

Print ISSN

3007-3189

<http://amresearchreview.com/index.php/Journal/about>



Fig. 4 Split tensile strength of cylinders in laboratory with UTM



Fig. 5 Cracks filled through the precipitation of calcium carbonate.

Cracks were induced by applying 90% of the load that caused failure in the previously tested bacterial concrete samples at the respective curing times, using the Universal Testing Machine (UTM). Since the crack size could not be precisely controlled, the resulting cracks varied in size. **Fig. 5.** Depicts White calcium carbonate deposits (highlighted in red circle) indicating cracks filling through bacterial activity. **Fig. 6** and **Fig. 7** depict the outcome of the split tensile strength test of normal and bacterial concrete conducted after 7 and 14 days curing, respectively. The concrete containing 4% bacterial solution had more strength compared to normal concrete and 5 % solution concrete. On the other hand, **Fig. 8** and **Fig. 9** show the strength gain of crack induced bacterial concrete samples after 7 and 14 days of curing, respectively.

Online ISSN

3007-3197

Print ISSN

3007-3189

<http://amresearchreview.com/index.php/Journal/about>

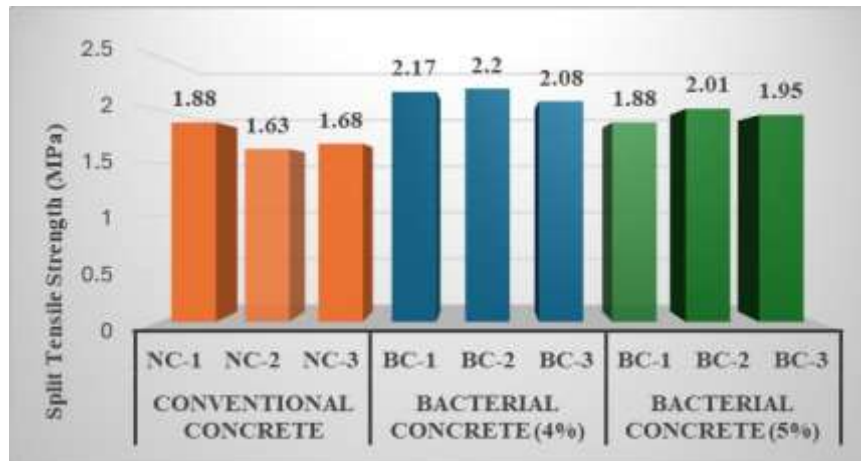


Fig. 6 Split Tensile Strength after 7 days of Curing– Conducted Before Inducing Cracks

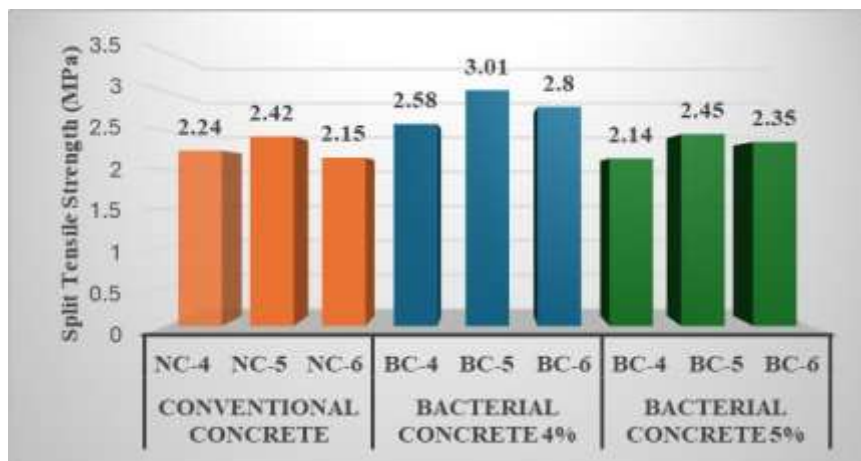


Fig. 7 Split Tensile Strength Test after 14 days of Curing – Conducted before Inducing Cracks

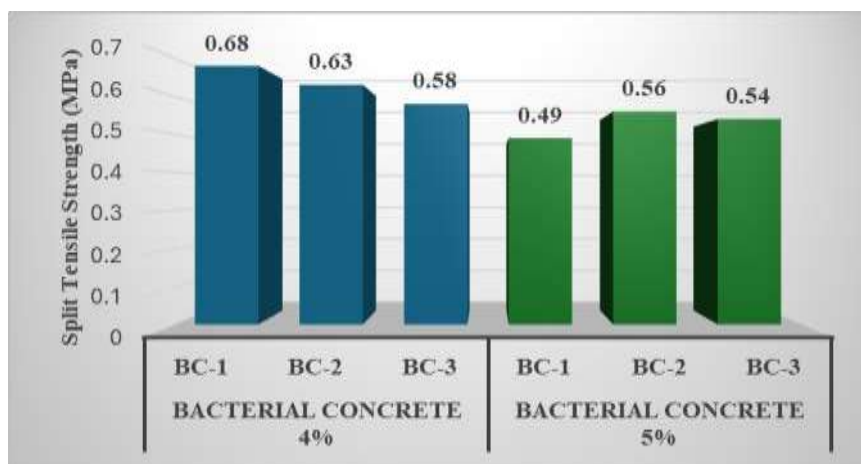


Fig. 8: Split Tensile Strength Test after 7 days of Curing – Conducted After Inducing Cracks

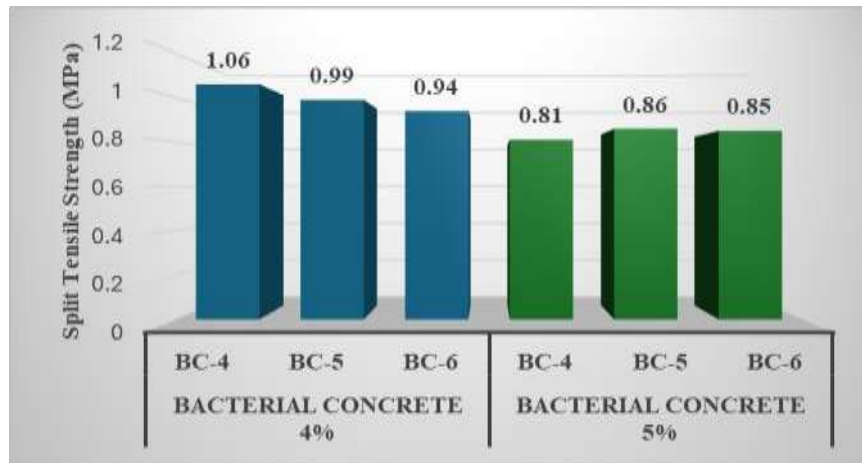


Fig. 9: Split Tensile Strength Test after 14 days of Curing– Conducted After Inducing Cracks

CONCLUSIONS AND RECOMMENDATIONS

The main objective of the study was to investigate the effect of *Escherichia coli* (E. coli) bacteria as a biological healing agent on the split tensile strength of concrete and to compare its performance with that of conventional concrete. In the experimental setup, 5% of the mixing water was replaced with a bacterial solution, and Calcium Lactate was incorporated as a nutrient source at 5% of the cement content. The concrete samples were subjected to split tensile strength tests after curing periods of 7 and 14 days. The results are graphically illustrated in **Fig. 6** and **Fig. 7**.

The results showed that:

After 7 days of curing:

- Normal concrete had an average tensile strength of 1.73 MPa.
- Bacterial concrete with 4% solution achieved 2.15 MPa (+19.53% increase).
- Bacterial concrete with 5% solution achieved 1.95 MPa (+11.11% increase).

After 14 days of curing:

- Normal concrete reached 2.27 MPa.
- Bacterial concrete with 4% solution reached 2.79 MPa (+18.63% increase).
- Bacterial concrete with 5% solution reached 2.31 MPa (+1.7% increase).

These findings show that 4% bacterial solution worked better in improving tensile strength than 5%. The induced cracks were then re-cured, and the bacterial concrete samples were tested to determine their healing performance. In 7 days, the sample containing 4% bacterial solution regained strength of 0.63 MPa whereas the 5% solution regained strength of 0.53 MPa. The strength of the 4% solution was also increased to 0.944 MPa with a 14-day curing period and 0.84 MPa of the 5% solution. These findings are clear indications that the bacterial concrete not only worked effectively in enhancing tensile strength but also displayed self-healing properties. The bacterial mix of 4% was always more effective than the 5% one, indicating that it offers a more optimal ratio of healing effect and material structure in the long term. Moreover, the visual examination proved the appearance of calcium carbonate in the cracks which proved the self-healing process.

Online ISSN

3007-3197

Print ISSN

3007-3189

<http://amresearchreview.com/index.php/Journal/about>

The difficulty of creating observable cracks in the cylindric specimens was one of the challenges. The Universal Testing Machine (UTM) present at the concerned laboratory was not the best one to do this. RCC beam samples would be more effective in the future work, since they crack more naturally when stressed, and more realistic to how structures behave in real-life conditions. To gain a real insight into the potential performance of bacterial concrete in practice, future research should also focus on long-term durability, conduct in-depth microstructural research with the help of such instruments as SEM and EDS, and examine the performance of the concrete in the real environmental conditions. The following steps are significant to demonstrate that bacterial self-healing concrete can be viable, sustainable, and applicable in the construction industry in everyday life.

References

- [1] Bheel, N., Nadeem, G., Almaliki, A.H., Al-Sakkaf, Y.K., Dodo, Y.A. and Benjeddou, O., 2024. Effect of low carbon marble dust powder, silica fume, and rice husk ash as tertiary cementitious material on the mechanical properties and embodied carbon of concrete. *Sustainable Chemistry and Pharmacy*, 41, p.101734.
- [2] Muneer Ahmed, Suliman Khan, Naraindas Bheel, Paul O. Awoyera, Olaolu George Fadugba. Developing high-performance low-carbon concrete using ground coal bottom ash and coconut coir fibre. *Results in Engineering*, Volume 27, 2025, 106607, ISSN 2590-1230.
- [3] Muneer Ahmed, Afsheen, Dileep Kumar, Zuhairuddin, Muhammad Zulkifal, Systematic Literature Review: Mechanical and Sustainability Properties of Jute Fiber Reinforced Concrete. (2026). *Annual Methodological Archive Research Review*, 4(4), 141-149.
- [4] Dileep Kumar, Afsheen, Dr. Gul Muhammad, Nizakat Ali, Muneer Ahmed, Influence of Steel Slag as a Cementitious Material on the fresh and Mechanical Characteristics of Concrete. (2026). *Annual Methodological Archive Research Review*, 4(4), 194-203.
- [5] Waqar, A., Bheel, N., Kirgiz, M.S., Khan, M.B. and Mansoor, M.S., 2025. Agro-Industrial Supplementary Cementitious Materials for Realizing Sustainable Development Goals of the UN and Net-Zero CO₂ Emission. In *Handbook of Innovative Adhesive Technology* (pp. 215-243). Jenny Stanford Publishing.
- [6] Bheel, N., Baikerikar, A.V. and Mohammed, B.S., 2025. Using optimization techniques on mechanical characteristics and sustainability assessment of rubberized concrete blended with PVA fiber through response surface methodology. *International Journal of Concrete Structures and Materials*, 19(1), p.8.
- [7] Bheel, N., Kumar, D., Ismail, F.I., Salilew, W.M., Alwetaishi, M., Waheeb, S.A. and Alkhatabi, L., 2024. Combined effect of jute fiber and corn cob ash on sustainability assessment and mechanical properties of roller compacted concrete using RSM modelling. *Scientific Reports*, 14(1), p.29724.
- [8] Pacheco-Torgal, F. and Labrincha, J.A., 2013. Biotech cementitious materials: Some aspects of an innovative approach for concrete with enhanced durability. *Construction and Building Materials*, 40, pp.1136-1141.

Online ISSN

3007-3197

Print ISSN

3007-3189

<http://amresearchreview.com/index.php/Journal/about>

- [9] Ramakrishnan, Venkataswamy & Ramesh, K. & Bang, S.. (2001). Bacterial concrete. Proceedings of SPIE - The International Society for Optical Engineering. 4234. 10.1117/12.424404.
- [10] Sengupta, Soumita. (2016). A REVIEW ON BACTERIAL CONCRETE.
- [11] Meera, C. M., & Subha, V. (2016). Strength and durability assessment of bacteria based self-healing concrete. IOSR Journal of Mechanical and civil Engineering, 50, 1-7.
- [12] Pachaivannan, P., Hariharasudhan, C., Mohanasundram, M., & Bhavani, M. A. (2020). Experimental analysis of self healing properties of bacterial concrete. Materials Today: Proceedings, 33, 3148-3154.