



# Experimental Study of Bacterial Concrete Using Pseudomonas Bacteria for Sustainable Construction

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## Abstract:

This study explores the potential of bacterial concrete as a sustainable construction material, specifically utilizing Pseudomonas bacteria for enhancing the mechanical properties and durability of concrete. Bacterial concrete incorporates microorganisms that promote calcite precipitation, a process that can effectively heal cracks and improve the overall longevity of concrete structures.

In this research, concrete samples were prepared with varying concentrations of Pseudomonas bacteria and subjected to a series of experimental tests to evaluate their impact on compressive strength, flexural strength, and crack healing efficiency. The results indicated that the incorporation of Pseudomonas bacteria significantly enhanced the compressive and flexural strengths of the concrete compared to control samples without bacteria. Furthermore, the healing efficiency of cracks was assessed through visual inspection and mechanical testing, demonstrating the ability of bacterial activity to regenerate structural integrity.

The findings suggest that bacterial concrete has the potential to address common issues related to concrete deterioration, such as cracking and reduced durability, thus contributing to more sustainable construction practices. This study highlights the feasibility of integrating microbial technology into concrete applications, providing a novel approach to enhancing the lifespan of structures while reducing maintenance costs and environmental impact. Future research will focus on optimizing bacterial strains and concentrations for even greater effectiveness in various environmental conditions, paving the way for innovative solutions in sustainable construction.

**Keywords:** concrete; concrete surface treatment; bacteria; curing; mechanical properties; eco-friendly.

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## 1. Introduction

Concrete is one of the most widely used construction materials globally due to its strength, durability, and versatility. However, conventional concrete is prone to various forms of deterioration, such as cracking, corrosion, and environmental degradation, which can significantly reduce its lifespan and structural integrity. These challenges necessitate innovative approaches to enhance the durability and sustainability of concrete structures.

One promising solution lies in the integration of biotechnology into concrete through the use of bacteria, particularly Pseudomonas species. Bacterial concrete leverages the natural processes of microorganisms to promote calcite precipitation, a mechanism that can effectively heal cracks and improve the material's overall performance. This biological self-healing property not only enhances the durability of concrete but also reduces maintenance costs and extends the service life of structures.



The use of Pseudomonas bacteria in concrete is particularly advantageous due to their robust metabolic pathways, which enable them to thrive in alkaline environments and produce calcium carbonate as a byproduct of their metabolic activity. This calcium carbonate precipitate can fill cracks and voids within the concrete matrix, restoring its mechanical integrity and preventing the ingress of harmful agents.

This study aims to experimentally investigate the effects of incorporating Pseudomonas bacteria into concrete on its mechanical properties and crack healing capabilities. A series of concrete samples will be prepared with varying concentrations of bacteria and subjected to rigorous testing to evaluate compressive strength, flexural strength, and crack healing efficiency.

By understanding the impact of bacterial activity on concrete performance, this research seeks to contribute to the growing body of knowledge on sustainable construction materials. The findings of this study will not only provide insights into the practical applications of bacterial concrete but also highlight its potential to address some of the most pressing challenges in modern construction, such as sustainability, durability, and long-term maintenance. Ultimately, this research aspires to pave the way for innovative and eco-friendly solutions that enhance the resilience of infrastructure in a rapidly changing environment.

## 2. Materials and Methods

### 2.1. Cement, Sand, Aggregate, and Water

Ordinary Portland cement 43 grade was used in this study, and it was tested in compliance with Indian Standard Specifications code IS 4031-1988. Additionally, natural sand that met Indian Standard Specifications IS 383-1970 was used in its construction. For fine and coarse aggregates, the maximum diameters allowed were 4.75 mm and 20 mm, respectively. In this study project, M 30 grade concrete was utilised, along with concrete ingredients that were derived from it.

### 2.2. Pseudomonas Bacteria

This experiment was conducted using a strain of Bacillus pseudomonas bacteria that was

obtained from the Department of Biotechnology at GLA University in Mathura.

### 2.3. Culturing Process of Bacteria

To reach the late exponential phase, an isolated strain of Bacillus pseudomonas bacteria was cultured for one day at 37 °C on a mechanical shaker after being injected in nutrient broth media. In this study, an isolated culture of bacteria from GLA University's Department of Biotechnology in Mathura was employed. It was continuously cultured on nutrient agar slants. The colony grew in an uneven white pattern on the nutrient agar. In a 100 mL tapered flask, a single colony of the culture is immunised into a 25 mL nutritive solution and left to proliferate. This setup calls for the apparatus to be spun at 125 revolutions per minute and the temperature to be kept at 30 degrees Celsius.

The inclusion of peptone (gramme per litre), sodium chloride (gramme per litre), and yeast extract (gramme per litre) all benefited in the growth of the bacteria culture. The growth of pseudomonas bacteria is seen in Figure 1. Two quantities of Pseudomonas bacteria—106 and 107 cf/mL—were investigated for this study. The cell concentration in the sample was determined using the following equation: Formula (1) [7]

$$y = 8.59 \times 10^7 x^{1.36}$$

where x = OD reading and y = the number of bacterial cells present in one milliliter of solution.

### Gram Staining

Spreading a fresh bacterial culture onto an empty microscope slide, letting it dry naturally, and then fixing it with a small bit of heat is the process of Gram-staining. The slide was immersed in gentian violet solution for a duration of one minute. The slide was then gently washed with treated water for two to three seconds before being immersed in an iodine solution for one minute. After cleaning the slide once again, 95 percent ethanol was applied and left on it until no more stain was seen dripping off the surface. The slide was then cleaned again and resuspended in safranin for ten seconds. It was then cleaned and allowed to air dry on a paper towel. Blue is the colour of gram-positive cells.



Figure 1. Growth of Pseudomonas Bacteria.

#### 2.4. Compressive Strength

Cubes of 100 mm by 100 mm by 100 mm were used for the compression strength test, and the casting process followed Indian Standard Specifications code IS 516 1959. All of the concrete specimens have to be cast at room temperature. Following 28, 56, 90, and 180 days of curing, the compressive strength of the microbial concrete samples was ascertained using IS code 516-1959.

#### 2.5. Split Tensile Strength

The concrete test's split tensile strength measured 100 mm by 200 mm. The casting of each specimen was completed in compliance with Indian Standard Specifications code IS 516 1959. Following the bacterial concrete's compressive and tensile tests, the shattered pieces of concrete were examined. Concrete cubes' fractured pattern indicated that they shatter unevenly, whereas concrete cylinders' fractured pattern indicated that tiny fractures are formed in the concrete cylinder close to the notch.

#### 2.6. Acid Attack and Sulphate Attack

For a certain amount of time, the concrete cube was submerged in a solution made of sulfuric acid and magnesium sulphate. Two distinct containers were filled with a mixture of 5% sulfuric acid and 5% magnesium sulphate in 1000 millilitres of tap water. There

were changes made to the 5% sulfuric acid and 5% magnesium sulphate solution every 7, 21, 28, and 56 days. After the samples deteriorated, the weight loss of the samples was largely investigated. Additionally, some research has demonstrated that changes in compressive strength might be a sign of corrosion. Concrete samples were exposed to seven-day attack cycles by Durning and Hicks, who used  $H_2SO_4$  solutions containing 1% and 5%  $H_2SO_4$ , respectively. To guarantee accuracy, the samples were cleaned and weighed following each cycle. They investigated the impact of silica fume deposition on resistance to exposure to  $H_2SO_4$ . This study found that the specimens' resistance rose in tandem with the amount of silica fume in the cement, which ranged from 0% to 30% by weight of the cement. The number of compounding periods of the samples in the 1 percent  $H_2SO_4$  solution was observed to sharply rise with a 30 percent replacement.

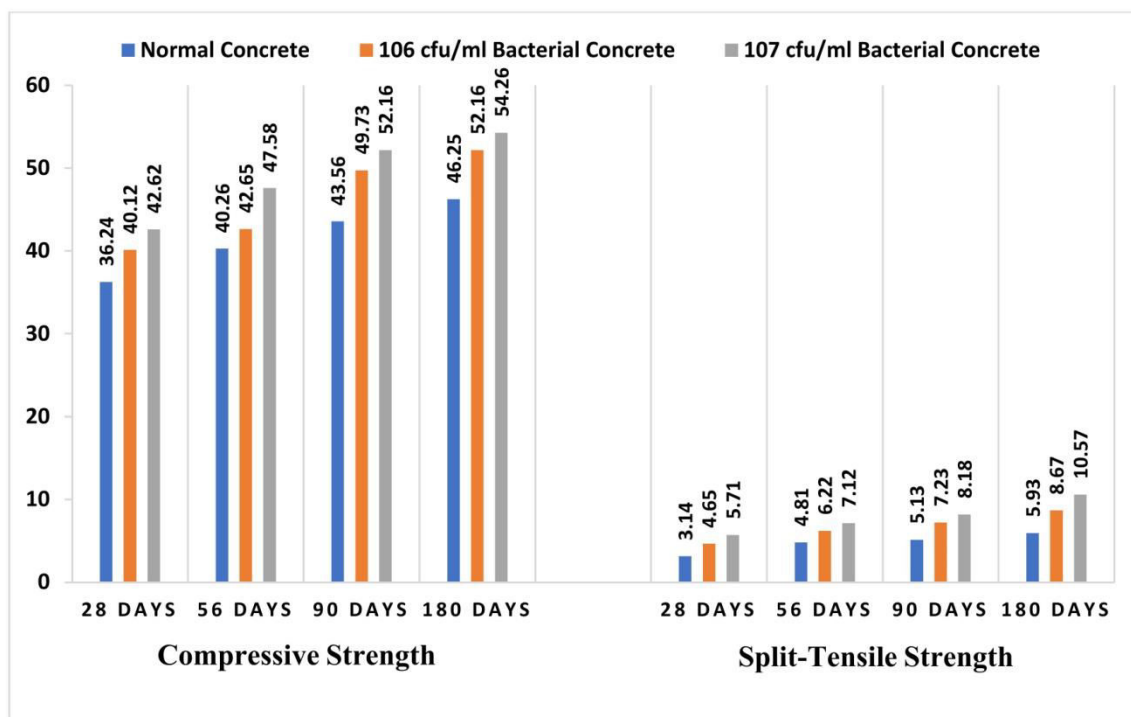
### 3. Discussion of Test Results

#### 3.1. Effect of Bacterial Solution on Compressive Strength and Split Tensile Strength

Figure 2 shows that bacterial concrete outperforms regular concrete in terms of split tensile strength and compressive strength. When it comes to precipitating calcium carbonate in concrete, bacterial cultures work

incredibly well. The results show that there was a maximum 16 percent improvement in compressive strength and a maximum 12 percent increase in split tensile strength when compared to ordinary concrete. The compressive strength of concrete made entirely of calcium lactate will be less than

that of concrete mixed with microorganisms because calcium lactate does not change after curing. It's also crucial to remember that bacteria convert calcium lactate to calcium carbonate ( $\text{CaCO}_3$ ), which is how calcium carbonate is created.



**Figure 2. Compressive and tensile strength.**

### 3.2. Effect of $\text{H}_2\text{SO}_4$ and $\text{MgSO}_4$ Attack on Pseudomonas Bacterial Concrete

The precipitation of  $\text{CaCO}_3$  inside the gaps of the concrete caused by the presence of microorganisms in the concrete increases the strength of the material while decreasing its permeability.  $\text{CaCO}_3$ , which is produced during the process of biomineralization, helps to increase strength, water permeability, reduce water absorption, sulphate resistance, and acid attack resistance. The use of microbes in concrete has several advantages: it is less harmful to the environment, increases strength, seals cracks in the concrete without the need for outside assistance, resists freeze-thaw cycles, has a lower permeability, has good durability, lessens the deterioration of reinforced concrete, and lowers repair and maintenance costs. Because bio concrete has a high acid durability index and a low permeability coefficient, it is less prone to erosion and needs fewer expensive repairs. In the industrial sector, these characteristics

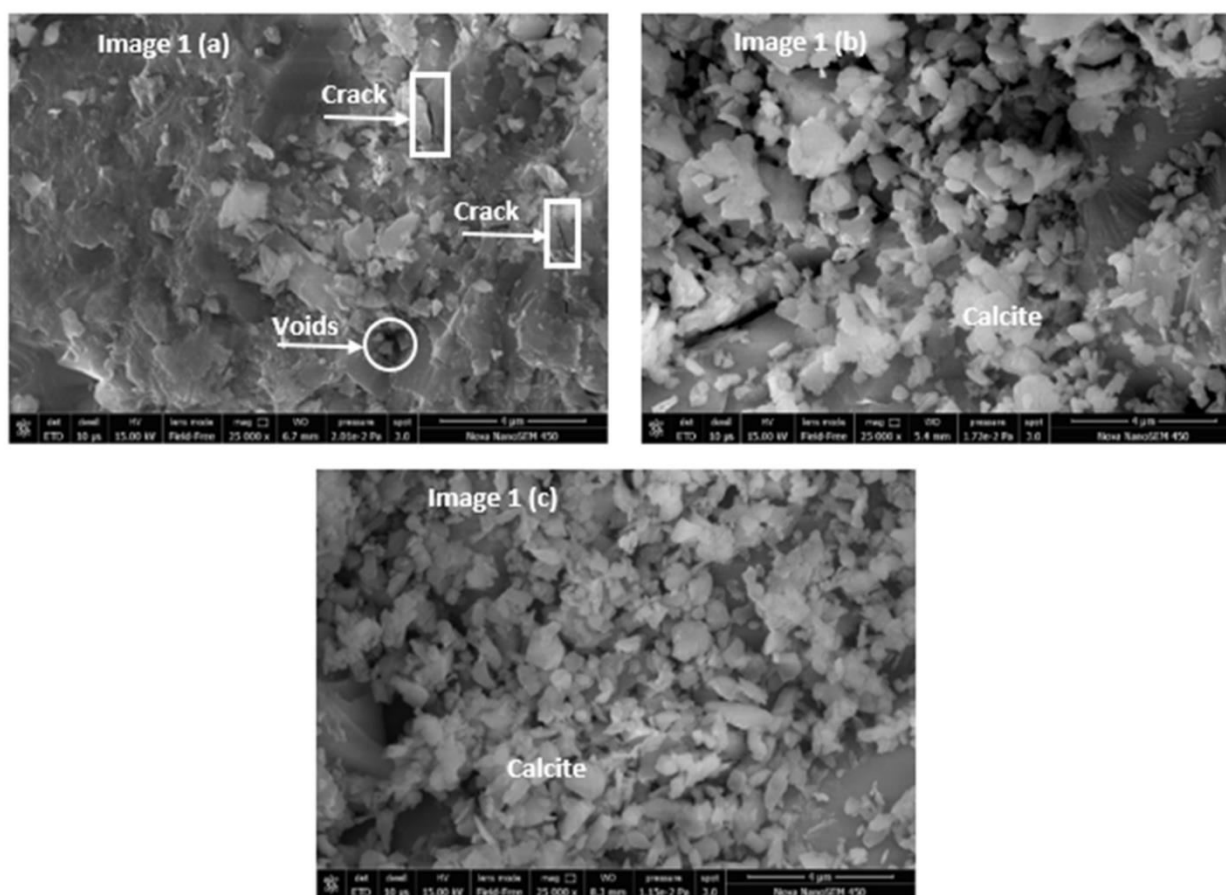
provide microbiological concrete an advantage. The samples' appearance following their exposure to  $\text{MgSO}_4$  and  $\text{H}_2\text{SO}_4$  solutions for 28, 56, and 90 days. Concrete is rapidly and extensively deteriorated by  $\text{H}_2\text{SO}_4$  acid. The specimens showed clear spalling and more severe damage to the corners and edges due to the stress concentration caused by the ettringite crystal formation. This was explained by the concentration of tension caused by the ettringite crystallisation.

For instance, the microorganisms caused the original face of the samples to be deteriorated in addition to the obvious damage around the corners and edges of the bacterial concrete specimens. Both the weight loss and the compressive strength loss were calculated at different ages. It was demonstrated that the mechanical characteristics and mass loss % are lower with the presence of bacteria than when the microorganisms are absent.

### 3.3. Scanning Electronic Microscopy (SEM)

The microstructure of regular concrete is evident in the SEM analysis picture for control concrete, as depicted in Figures 3 and 4 Image 2a. The picture was obtained at a magnification of 25,000 and a sub-acceleration voltage of 15,000 V. Using a scanning electron microscope, an image of Pseudomonas microbiological concrete was captured at a magnification of 25,000 and a sub acceleration voltage of 15,000 V. Figures 9 and 10 depict the microstructural examination

of the microbial concrete, respectively, with b and c. The images from the scanning electron microscopy analysis show that the Pseudomonas microbiological concrete contains calcite precipitation. Each and every specimen had pores, CSH, and CH. The pores of microbiological concrete were found to contain calcite, which is aptly illustrates how calcite growth results in a decrease in porosity, an increase in strength, and the healing of fractures.



**Figure 3. SEM image of conventional concrete and bacterial concrete for 28 days. Image 1 (a) conventional concrete sample after 28 days, Image 1 (b) 10<sup>6</sup> cf/mL Pseudomonas Bacterial concrete after 28 days, and Image 1 (c) 10<sup>7</sup> cf/mL Pseudomonas Bacterial concrete after 28 days.**

According to the concrete's scanning electron microscope measurements, the concrete was denser at 180 days than it was at 28 days. Furthermore, bacterial concrete was discovered to be much denser than regular concrete. After 28 days, the concrete in Image 1a of Figure 9 had significant volume microcracks and gaps, however Image 1b of Figure 9 showed that the 10<sup>6</sup> cfu/mL concrete's fractures and gaps were substantially filled. Comparably, it can be

observed in Figure 9 Images 1b,c that the 10<sup>7</sup> cfu/mL concrete had more filled gaps and fractures than the 10<sup>6</sup> cfu/mL concrete. Furthermore, as Figure 4 illustrates, Pseudomonas Bacterial concrete thickens over 180 days compared to 28 days. Additionally, Images 1b,c of Figure 10 demonstrate that the holes and fissures in 10<sup>7</sup> cfu/mL concrete are filled more thoroughly than in 10<sup>6</sup> cfu/mL concrete.

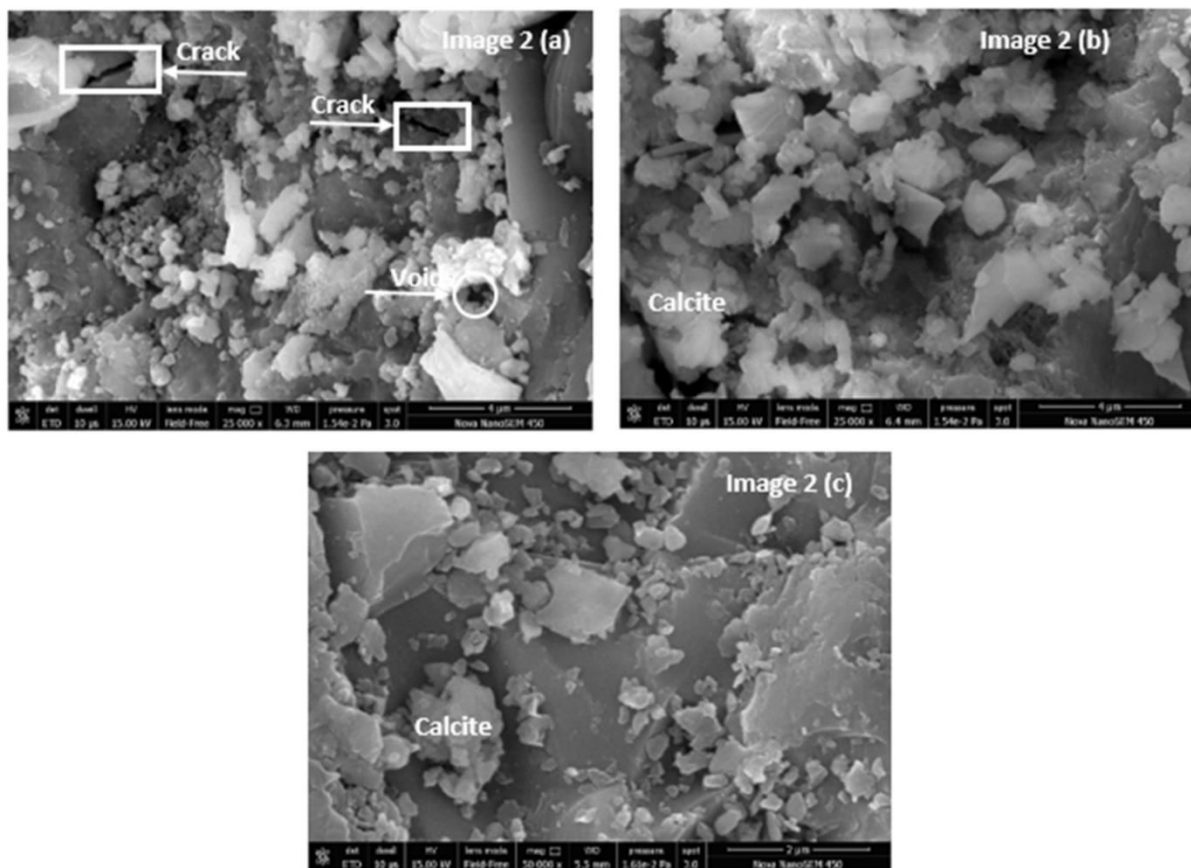


Figure 4. SEM image of conventional concrete and bacterial concrete for 180 days.

#### Image

2(a) conventional concrete sample after 180 days, Image 2 (b)  $10^6$  cf/mL Pseudomonas Bacterial concrete after 180 days, and Image 2 (c)  $10^7$  cf/mL Pseudomonas Bacterial concrete after 180 days.

#### 4. Conclusions

This study successfully demonstrated the viability of using Pseudomonas bacteria in the development of bacterial concrete, highlighting its potential as a sustainable construction material. The experimental investigations provided several key insights into the performance enhancements offered by the incorporation of these microorganisms: Enhanced Mechanical Properties: The results indicated a significant improvement in both compressive and flexural strength in concrete samples containing Pseudomonas bacteria compared to conventional concrete. This enhancement is attributed to the calcite precipitation facilitated by bacterial activity, which contributes to the overall structural integrity of the concrete.

Effective Crack Healing: The ability of Pseudomonas bacteria to induce self-healing in concrete was evident through the assessment of crack healing efficiency. Visual inspections and mechanical tests confirmed

that bacterial activity effectively filled cracks and voids, restoring the material's load-bearing capacity and extending its lifespan.

Sustainability Benefits: The findings underscore the potential of bacterial concrete to address common issues associated with conventional concrete, such as cracking and deterioration, thus reducing the need for frequent repairs and maintenance. By promoting longevity and reducing lifecycle costs, this innovative approach aligns with contemporary goals for sustainable construction practices.

Future Applications: The study opens avenues for further research into optimizing bacterial strains, concentrations, and concrete mix designs for even greater effectiveness. Investigating the performance of bacterial concrete under various environmental conditions will provide valuable insights into its applicability in diverse construction scenarios.

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