

# Role of Bacterial Biopolymers in Modifying the Structural Integrity of Limestone Blocks an Experimental Study

Martin Sohil\*

Department of Mechanical Engineering, University of Applied Sciences Muenster, Germany

## Abstract

This study investigates the role of bacterial biopolymers in altering the structural integrity of limestone blocks. By introducing specific biopolymers produced by *Bacillus* and *Pseudomonas* strains, the experiment aimed to assess changes in the mechanical and physical properties of limestone, including compressive strength, porosity, and water absorption. The results indicated that bacterial biopolymers significantly enhanced the durability and structural integrity of the limestone blocks. This research highlights the potential of biopolymers in the preservation and restoration of stone materials, offering a sustainable approach to enhancing the longevity of limestone structures.

**Keywords:** Bacterial biopolymers; Limestone blocks; Structural integrity; Compressive strength; Porosity; Water absorption; Stone preservation

## Introduction

Limestone, widely used in construction and historical monuments, is vulnerable to environmental degradation, leading to structural weakening over time. Traditional methods for reinforcing limestone often involve synthetic chemicals that may pose environmental risks [1]. Recently, bacterial biopolymers have emerged as a potential eco-friendly alternative for improving the durability and strength of stone materials. This study focuses on understanding how biopolymers produced by specific bacterial strains, such as *Bacillus* and *Pseudomonas*, can influence the physical and mechanical properties of limestone [2]. By exploring the interaction between bacterial biopolymers and limestone, this research aims to provide insights into sustainable methods for stone preservation and restoration.

## Methodology

### Preparation of bacterial cultures

**Bacterial Strains:** The study utilized two bacterial strains known for their biopolymer production: *Bacillus subtilis* and *Pseudomonas aeruginosa*. These strains were selected based on their ability to produce extracellular biopolymers that can interact with calcium carbonate, the primary component of limestone [3,4]. Bacteria were cultured in a nutrient-rich medium at 30°C under aerobic conditions. The growth medium contained essential nutrients to stimulate biopolymer production. After 48 hours of incubation, the bacterial cultures reached their peak biopolymer production, as confirmed by optical density measurements and biopolymer extraction.

### Biopolymer extraction and preparation

**Extraction Process:** The bacterial cultures were centrifuged at 10,000 rpm for 15 minutes to separate the cells from the supernatant containing the biopolymers. The supernatant was then subjected to precipitation using cold ethanol (1:3 ratio) [5]. The precipitated biopolymers were collected by centrifugation and dried at 40°C to obtain a powder form. The extracted biopolymers were dissolved in deionized water to prepare 1% (w/v) biopolymer solutions. These solutions were then filtered to remove any impurities before application to the limestone blocks.

### Preparation of limestone blocks

**Limestone Samples:** Experimental limestone blocks, each measuring 5 cm x 5 cm x 5 cm, were prepared from a single block of high-purity limestone to ensure uniformity in composition and structure [6,7]. The limestone blocks were thoroughly cleaned with deionized water and air-dried to remove any surface contaminants that could interfere with the biopolymer application.

### Application of bacterial biopolymers

**Treatment Process:** The limestone blocks were divided into three groups: (1) untreated control, (2) treated with *Bacillus* biopolymer solution, and (3) treated with *Pseudomonas* biopolymer solution. Each block was immersed in the respective biopolymer solution for 24 hours to ensure deep penetration of the biopolymer into the stone matrix. After treatment, the blocks were air-dried for 48 hours at room temperature [8]. Following this, the blocks were cured in a humidity-controlled chamber (60% relative humidity) for seven days to allow the biopolymers to fully interact with the limestone.

## Results and Discussion

### Compressive strength

**Results:** Limestone blocks treated with bacterial biopolymers showed a significant increase in compressive strength compared to untreated controls. Specifically, biopolymers produced by *Bacillus* strains resulted in a 20% increase, while *Pseudomonas* biopolymers achieved a 15% increase. The enhancement in compressive strength can be attributed to the biopolymers filling micro-cracks and pores within the limestone, effectively bonding the stone matrix [9]. This suggests that bacterial biopolymers can serve as a natural reinforcement agent, improving the load-bearing capacity of limestone without altering its appearance.

\*Corresponding author: Martin Sohil, Department of Mechanical Engineering, University of Applied Sciences Muenster, Germany, E-mail: Smartin07@gmail.com

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### Porosity and water absorption

Results: The introduction of bacterial biopolymers significantly reduced the porosity and water absorption of the limestone blocks. The porosity of blocks treated with *Bacillus* biopolymers decreased by 25%, while *Pseudomonas* biopolymers led to a 22% reduction. Correspondingly, water absorption rates dropped by 30% and 27% for *Bacillus* and *Pseudomonas*-treated blocks, respectively. The reduction in porosity and water absorption is indicative of the biopolymers' ability to penetrate and seal the pores within the limestone [10]. This decreased permeability likely contributes to the improved resistance of the limestone blocks to environmental factors such as moisture and freeze-thaw cycles, enhancing their durability and lifespan.

### Surface morphology

Results: Scanning electron microscopy (SEM) revealed that bacterial biopolymers formed a thin, uniform layer on the surface of the limestone blocks, filling microvoids and creating a smoother surface texture. The surface coating provided by the biopolymers not only contributes to the mechanical strength but also protects the limestone from weathering. This layer acts as a barrier against pollutants and moisture, further preserving the stone's integrity. However, long-term studies are required to assess the durability of this coating under various environmental conditions.

### Conclusion

The experimental study demonstrates that bacterial biopolymers have a significant positive impact on the structural integrity of limestone blocks. The enhanced compressive strength, reduced porosity, and lower water absorption rates suggest that biopolymers produced by *Bacillus* and *Pseudomonas* strains can be effectively used for the preservation and restoration of limestone structures. These findings open up new possibilities for utilizing biopolymers in sustainable construction practices, particularly in the conservation of historic monuments. Future research should focus on the long-term performance of these biopolymer treatments in real-world environmental conditions and their potential application to other types of stone materials.

### Acknowledgement

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### Conflict of Interest

None

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