

# Experimental Study on Bacterial Cement Composites

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**Abstract:-** Concrete in most structures is designed to crack in order to let embedded steel reinforcement take over tensile stresses. Crack formation is also a typical phenomenon related to durability. Percolation of cracks may lead to leakage problems or ingress of deleterious materials, causing deterioration of the concrete matrix or corrosion of embedded steel reinforcement. Durability can be enhanced by preventing further ingress of water and other substances. In recent years a bacteria-based self-healing concrete is being developed in order to extend the service life. A two component healing agent is added to the concrete mixture. The agent consists of bacteria and an organic mineral precursor compound. Self healing agents such as epoxy resins, bacteria, fiber are used to heal cracks in concrete. Among these, bacteria used in concrete are effective. When the bacteria is mixed with concrete the calcium carbonate precipitates forms and these precipitates filling the cracks and makes the crack free concrete. The state of art results in all projects show that material designed as self healing agents.

**Keywords:** Bacterial concrete, Calcium carbonate, Epoxy resins, Self healing agent.

## 1. INTRODUCTION

Concrete in most structures is designed to crack in order to let embedded steel reinforcement take over tensile stresses. Crack formation is also a typical phenomenon related to durability. Percolated cracks may lead to leakage problems or ingress of harmful materials, which can cause deterioration of the concrete matrix or reinforcement corrosion. Durability can be enhanced by preventing further ingress of water and other substances. Self-healing is characterized by regaining performance after a defect occurs. Damage targeted in bacteria-based self-healing concrete particularly relates to increased durability and leakage prevention and extending service life of concrete structures. Jonkers [1] introduced a two-component healing agent to be added to the concrete mixture, consisting of bacteria and a mineral precursor compound. Upon cracking the system is activated by ingress water. Bacteria convert the mineral precursor compound into the mineral calcium

carbonate, better known as limestone. Precipitation of the limestone on the crack surface enables sealing and plugging of the cracks, making the matrix less accessible to water and other deleterious materials. Advancement in concrete technology is in its strength improvement and its enhancement in durability, using pollution-free and natural methods.

Last few years several investigations are made in bacterial concrete. H.M.Jonkers gives an overview of durability of bacterial concrete. His paper deals with the self healing repair mechanism. He concludes that the bacterial concrete heals sub millimeters crack size of 0.15mm. This concrete is very effective in wet environment and controls the corrosion of steel reinforcement. Nithin kumar et al studied about the fracture of bacterial concrete. He considered the fracture parameters such as stress Intensity Factor(K), Fracture Energy(Gf), crack mouth opening displacement and brittleness number(S). His analytical and experimental investigation concludes

that the fracture in bacterial concrete is less compared to conventional due to high stress intensity factor, low deflection and less fracture energy. Jagadeesh Kumar et.al compared three species of bacillus such as *Bacillus flexus*, *Bacillus Sphaericus* and *Bacillus pasteurii*. From this he concluded that the bacillus flexus is an best option in MICP(Microbially Induced Calcite Percipitation).

The main goal of the present research therefore was to develop a type of sustainable self-healing concrete using a sustainable self-healing agent. It was reported that the effect of bio-deposition improves the durability of cement mortar/concrete specimens. It was also observed that deposition of  $\text{CaCO}_3$  crystals decreased the water absorption of the sample depending on the inherent porosity of the specimen leading to a decrease in the carbonation rate by about 25–30% [3]. Another aspect of concrete is its liability to cracking, a phenomenon that hampers the material's structural integrity and durability. The effects of durability problems reflect so much on the money spent for maintenance and repair of concrete structures [2]. Self-healing concrete is a product that will biologically produce limestone to heal cracks that appear on the surface of concrete structures. Specially selected types of the bacteria genus *Bacillus*, along with a calcium-based nutrient known as calcium lactate, and nitrogen and phosphorus, are added to the ingredients of the concrete when it is being mixed. These self-healing agents can lie dormant within the concrete for up to 200 years [4].

Use of bacteria in concrete remediation is an unorthodox concept in current concrete research. It is however, a new approach to an old idea that a microbial mineral deposit constantly occurs in natural environment. The long term goal is to understand the significance of micro-organisms in concrete structures [6]. Therefore, bacterially induced calcium carbonate precipitation has been proposed as an alternative and environmental friendly crack repair technique [5]. Durability problems such as crack formation are typically tackled by manual inspection and repair, i.e. by impregnation of cracks with cement or epoxy-based or other synthetic fillers. An integrated healing agent will save manual inspection and repair and moreover increases the structure's durability. Addition of such an agent to the concrete mixture would save money and environment .

## 2. METHODOLOGY

The methodology for producing a self-healing concrete involves the following steps.

- i. Selection and cultivation of bacteria.
- ii. Preparation of test specimens.
- iii. Characterization studies
  - X-ray diffraction
  - Scanning Electron Microscopy (SEM)
  - Thermo-Gravimetric Analysis (TGA)
  - Compressive Strength and Tensile Strength Testing
  - Ultrasonic Pulse Velocity

### 2.1 Selection of Bacterial Species

Spore forming alkali-resistant bacteria can be isolated from its source. Bacterial strains such as *Bacillus pasteurii*, *Escherichia coli*, *Bacillus sphaericus*, *Bacillus subtilis*, *Bacillus cereus* etc., are commonly used for research works. Initially these bacteria are obtained from the source and first cultured in a solid media and then transferred to nutrient broth (liquid media) which is sterile and kept shaking in an incubator.

### 2.2 Measurement of Bacterial Cells

Concentration of bacterial cells is measured by Haemocytometer and optical density could be found by spectrophotometer analysis before adding bacteria to cement composites. Gram staining method was used to determine the morphology of the bacterial strains and the bacterial cultures are tested for urealytic activity and also calcium carbonate precipitation [7]. Before addition to cement cement mixture for test specimen preparation, bacteria should be cleaned from culture residues by repeated centrifugation and resuspension of obtained cell pellet in a clean tap water. Ureolytic bacteria such as *B. sphaericus* could precipitate  $\text{CaCO}_3$  by conversion of urea into ammonium and carbonate. *subtilis* (CMBS). It is found that there is 28% improvement in compressive strength of CMBS incorporated concrete compared to control concrete with optimum concentration [8]. Matrix capillary water of young

concrete is typically characterized by pH values between 11 and 13. Bacteria added to the concrete mixture thus do not only have to resist mechanical stresses due to mixing but should also be able to withstand a high alkalinity for prolonged periods. Most promising bacterial agents for incorporation in the concrete matrix therefore appear to be alkaliphilic (alkali-resistant) spore-forming bacteria. As the concrete matrix is toxic due to ingress oxygen (diffusion through matrix capillaries) incorporated bacteria also need to be oxygen tolerant.

Such aerobic alkaliphilic spore-forming bacteria occur within the genus *Bacillus*, and several representatives of these were therefore selected to test their applicability as healing agent in concrete [9]. The starting point of the research is to find bacteria capable of surviving in an extreme alkaline environment. Cement and water have a pH value of up to 13 when mixed together, usually a hostile environment for life most organisms die in an environment with a pH value of 10 or above. The search concentrated on microbes that thrive in alkaline environments which can be found in natural environments. Samples of endolithic bacteria (bacteria that can live inside stones) will be collected along with bacteria found in sediments in the lakes. Strains of the bacteria genus *Bacillus* will be found to thrive in this high-alkaline environment.

### 2.3 Preparation of Test Specimens

Bacterial concrete casted by using ordinary Portland cement mixed with bacterial concentration 106 cells/ml of water. Conventional concrete samples are also casted in parallel. The specimens are cured under tap water at room temperature and tested at 7, and 28 days .

### 2.4 Characterization Studies

The formation of calcite by means of bio-mineralization can be analysed by using various characterization techniques or methods. These techniques are specialized or involve all modes of microbial analysis like imaging, diffraction and spectroscopy, including light, X-rays, neutron or electron as primary radiation. To conduct the above studies, samples should be collected from the tested

mortar or concrete specimens in the form of powders or broken pieces .

## 3. HEALING AGENT

### 3.1 Self-healing

Current material design in engineering follows the concept of damage prevention. An alternative design principle is that of self-healing materials, according to the concept of damage management as introduced by Vanderzwaag [2]. Damage formation does not necessarily cause problems, if it is subsequently healed in an autonomous process. Self healing materials have to serve some roles and meet several properties. Damages should be sensed, followed by transportation of healing agent to the damage site, triggering repair of the damage. In the ideal case self-healing materials are cheap and have properties equal or superior to currently used materials, with the ability to heal defects of any size, multiple times, completely and autonomously.

In case of concrete durability performance is mainly considered for damage to be healed, in order to reduce costs of repair and maintenance. An overview of characteristics for selfhealing concrete is given by Jonkers [10]. Target for self-healing concrete is to reduce matrix permeability by sealing or blocking cracks. Healing agent is incorporated in the concrete matrix and acts without human intervention. Preference lies in agents working as a catalyst, enabling multiple healing events. To make the material technically and economically competitive, healing agent should be cheap in relation to the low price of concrete, remain potentially active for long periods of time and be concrete compatible to not negatively affect material characteristics.

### 3.2 Microbial healing

Concrete already has a built-in healing mechanism due to on-going chemical, physical and mechanical processes. Most significant is precipitation of calcium carbonate [11]. Average limit for which healing can still occur is a crack width of 0.2 mm. Carbonation reaction lies at the base of the calcium carbonate production, where diffused carbon dioxide reacts with

the hydration product calcium hydroxide as can be seen in Eq. (1).



The principle of microbial healing also lies in the precipitation of calcium carbonate [4]. Ingress water activates dormant bacteria. Dense layers of calcium carbonate are produced by bacterial conversion of an incorporated mineral precursor compound. In case of calcium lactate the reaction is as given in Eq. (2), where bacteria only act as a catalyst.



From the metabolic conversion of calcium lactate carbon dioxide is produced, which further reacts with the calcium hydroxide from the concrete matrix according to the chemical reaction in Eq. (1), producing additional calcium carbonate.

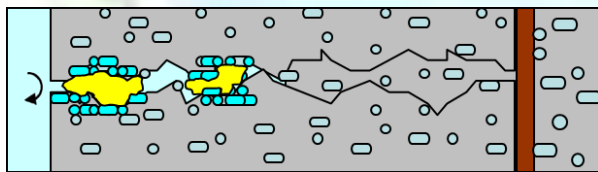


Fig 1: Scenario of crack-healing by concrete-immobilized bacteria

### 3.3 Direct addition

Healing agent mainly consists of bacteria and a mineral precursor compound. First important consideration was to choose concrete compatible bacteria. Bacteria should survive and remain active in the highly alkaline environment. Since concrete structures are designed to last at least 50 to 100 years, bacteria should remain viable for a long period of time. Therefore a specific group of alkaliphilic spore-forming bacteria was selected. The thick cell walled spores are produced by bacteria when living conditions become less favourable.

The combination of suitable bacteria and calcium lactate as mineral precursor compound calcium lactate indeed resulted in production of calcium carbonate precipitates in concrete cracks. The observed mineral production in time however appeared limited when calcium lactate and bacterial spores were directly added in unprotected form to the concrete mixture,

probably due to full integration of the precursor compound in the matrix limiting its access to bacteria [12]. Increased potential for long-term viability and activity may be reached when integrated bacterial spores are immobilized or protected and the precursor compound is kept accessible for bacterial conversion. Calcium lactate [15] however, appeared to be the most suitable compound as its application as main healing agent ingredient resulted in even enhanced concrete compressive strength values. Tests on concrete compatibility showed no significant influence on flexural and compressive strength characteristics for concentrations of added bacteria up to 109 cm.

## 4. EFFECT OF HEALING AGENT ADDITIONS ON SPECIMEN STRENGTH

As incorporation of healing agents to concrete may have unwanted negative effects on material properties, development of compressive strength of control specimen without additions as well as specimen with bacteria or organic compound additions was investigated. Incorporation of a high number of bacteria appears to have a mildly negative effect on compressive strength development as bacterial test specimen appeared almost 10% weaker than control specimen. Effect of organic compound incorporation on development of strength appeared however strongly dependent on compound identity.

## 5. BACTERIAL SPORE FORMATION

Addition of manganese to the growth medium stimulated the formation of bacterial spores substantially. Light microscopic analysis of growing cultures. Spores could be easily visualized by ESEM analysis due to their thick cell walls and their diameter appeared to be typically in the size range of 0.8–1  $\mu\text{m}$ .

## 6. RESULTS AND DISCUSSIONS

One major problem associated with crack formation is that the process results in a drastic increase in material permeability increasing the risk of matrix and embedded reinforcement degradation by ingress water and other aggressive chemicals. Active bacterially mediated mineral precipitation could

result in crack-plugging and concomitant decrease in material permeability. As bacteria function as catalyst, a suitable mineral precursor compound needs additionally to be incorporated in the material matrix to provide a truly autonomous repair mechanism. However, the maximal allowable amount of mineral precursor compound introduced to the concrete mixture is likely limited as larger quantities may negatively affect other concrete properties such as setting time and (final) strength. Self-healing concrete should be able to heal or seal, by filler material formation, freshly formed cracks to inhibit ingress of water and other chemicals which could cause preliminary degradation[14] of the material matrix or embedded reinforcement. In this study we investigated the bio-mineral production capacity of cement stone specimen in which bacteria were incorporated as healing agent. The integrated bacteria applied in this study are affiliated to alkali-resistant spore-forming species of the genus *Bacillus*. In conclusion we can state that alkali-resistant spore-forming bacteria related to the genus *Bacillus* represent promising candidates for application as self-healing agent in concrete and probably other cement-based materials. It is found that cement stone incorporated bacterial spores are able to convert concomitantly incorporated calcium lactate to calcium carbonate-based minerals upon activation by crack ingress water [13]. Although concrete with a high self-healing (crack healing) potential is wanted, the addition of healing agents such as bacteria and/or (organic) chemical compounds to the paste may result in unwanted decrease of strength properties.

## 7. CONSIDERATIONS

In order to consider practical application several characteristics have to be determined. Viability and functionality of incorporated bacteria is enhanced until several months after concrete casting. For practice long-term self-healing capacity is needed, ideally for the duration of the service life of the concrete structure. Also multiple healing events should be possible. Cost efficiency is also important. Concrete is a relatively cheap construction material, and adding a self-healing material to the concrete mixture has to be economically feasible. E.g. the return on investment price could come from savings on otherwise needed repair and maintenance costs. In order to minimize the price of the healing agent, its

production should be straightforward with large output and little loss, minimizing the use of complex procedures, heating and cooling.

## 8. CONCLUSION

From the above discussion the bacteria such as *Bacillus Pasteuri*, *Bacillus megaterium*, *Bacillus subtilis* are having some disadvantages and also *Pseudomonas aeruginosa* are undoubtedly pathogen and cannot be directly applied in building structures like houses and offices because of health concerns. Finally we conclude that the *Bacillus Sphaericus* and *Escherichia Coli* have some advantageous than above bacteria.

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