



Effects of bacterial remediation on compressive strength, water absorption, and chloride permeability of lightweight aggregate concrete



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HIGHLIGHTS

- The properties of LWAC containing Bacteria *S. pasteurii* in mix water, in LWA and simultaneously in both LWA and mix water was presented.
- Calcite producing bacteria improved the strength properties of LWAC.
- Production of calcite on the LWAC were proven through SEM images.

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ABSTRACT

Structural lightweight aggregate concrete (LWAC) offers such advantages as reduced dead load and decreased slab and beam size of concrete structures. To this may be added the economic advantages of artificial lightweight aggregates (LWA). However, LWAC basically suffers from higher porosity and water absorption compared to normal-weight concrete. Due to the negative side-effects of certain chemical techniques, biological methods have been proposed as an environmental friendly strategy for reducing concrete porosity and diminishing water absorption. In this regard, calcium carbonate precipitation induced by micro-organisms has found wide applications in construction technology for its effect on improved quality of building materials. This paper presents the results of an experimental investigation carried out to evaluate the influence of *Sporosarcina pasteurii* at cell concentrations of 10^6 cells ml^{-1} on water absorption, water permeability, compressive strength, and rapid chloride permeability (RCP) of LWAC. For the purposes of this study, Leca aggregates were left to soak in a solution of urea- CaCl_2 containing bacteria for 6 days to investigate biological improvement of aggregate quality. Next, four types of LWAC were made under the three treatments of bacterially-treated aggregates, bacteria inoculated in the concrete mix water, and both techniques employed simultaneously and with no bacteria used in either the aggregate or the concrete mix solution as the control. The results revealed an average reduction of about 10% in water absorption, 20% increase in compressive strength, and 20% reduction in chloride penetration in the experimental specimens relative to the same properties in the control ones. Furthermore, scanning electron microscopy (SEM) analysis revealed denser and lower porosity of LWAC specimens with bacteria in their concrete mix water and aggregates as compared to those with bacteria used only in their concrete mix water.

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1. Introduction

The primary purpose of using lightweight aggregate concrete (LWAC) is to reduce the dead load of a concrete structures, which

thereby leads to reduced size of the load bearing elements. Structural LWAC mixtures can be designed to achieve strengths, mechanical properties, and durability similar to those of conventional normal weight concrete [1–3]. Compared to normal weight concrete, however, the aggregates in LWAC are characterized by a higher water absorption, which needs to be reduced. Many research efforts have been devoted to the reduction of water absorption of LWAC along with their enhanced durability and compressive strength [4]. Among these, a number of studies have been devoted over the past decades to the bacterially-induced and mediated mineralization of normal weight concrete.

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There are different strategies to decrease water permeability of LWAC leading to increase in durability characteristics of concrete. However, most of these strategies such as utilizing epoxy cover, acrylic resins, and silicon based polymers involve the use of materials which are non-compatible with concrete and their output production are hazardous to environment [5,6]. Many researchers use biomineralization as a natural process and environmental friendly solution to improve the durability and mechanical properties of concrete [5]. Wasim et al. by defining bio-concrete as a viable solution for controlling crack propagation reported that use of various bacteria incorporation techniques is an environmental solution way to crack healing [6].

The microbially induced calcium carbonate precipitation process is an effective and eco-friendly technology that can be applied to solve various environmental problems [7–10]. Van Paassen et al. [11] reported that microbial induced carbonate precipitation may not be completely environmental friendly, because ammonium and nitrate are formed during the ureolysis-driven process, which can be toxic and hazardous to human health and soil microorganisms at high concentrations. Ganendra et al. [12] recently found that it was advantageous over ureolysis-driven processes because the calcium formate did not release the ammonia to the air or produce nitric acid when applied to building materials, resulting in decreased risk of pollution and bio-deterioration of the materials [10]. Therefore, using bacteria in concrete and cement base material not only does not damage the environment but also is an eco-friendly strategy compared to chemical solution.

Some studies have investigated the applicability of bacteria that help calcium carbonate to precipitate in concrete in an attempt to enhance the compressive strength of mortar or concrete [6,13–18].

Ghosh et al. [19] studied the potential of *Shewanella* and *E. coli* in enhancing the compressive strength of mortar specimens and observed the greatest improvement equal to 25% after 28 days in specimens treated with *Shewanella* at a cell concentration of 10^5 cells ml^{-1} but no noticeable increase in those treated with *E. coli* (the low urease producing bacteria). They concluded that the type of micro-organism might play the primary role in improving concrete strength [19].

Achal et al. [20] treated mortar cubes with *Sporosarcina pasteurii* and observed an improvement of 17% in the compressive strength of their specimens. They argued that microbial calcite might have precipitated on cell surfaces and, eventually, within the mortar pores to plug them, ultimately leading to the cessation of oxygen and nutrient flow into the cells. Consequently, the cells have subsequently either died or turned into endospores to act as organic fiber enhancing the compressive strength of the mortar cubes [20]. Ramakrishnan et al. reported increased concrete resistance toward alkali, freeze-thaw attack, and drying shrinkage in addition to a reduction in permeability upon application of bacterial cells [21]. Achal et al. reported decreased water permeability in bio-remediated cement mortar cubes treated with *Sporosarcina pasteurii* [22].

Nosouhian et al. used two bacterial strains in a concrete mix water to examine the effects of simultaneous use of the bacteria in a harsh sulfate environment. They found that not only did the 28-day compressive strength of the experimental concrete containing the strains increase by about 20% relative to that of the control, but that its chloride penetration also reduced [23].

Drawing upon the numerous studies carried out on bio-deposition demonstrating its effects on modified and/or reduced concrete porosity [24–27]. The present experimental study was designed to investigate the influence of using calcium-carbonate-producing bacteria on a special type of lightweight aggregate (LWA), named Leca. Additionally, the proposed method of bacterial remediation is compared with another method in which bacteria are used in the mix water of the concrete. The properties of LWAC

are finally evaluated using such indicators as water absorption, compressive strength, and rapid chloride permeability (RCP). Scanning electron microscopy (SEM) is also employed to visualize the effects of bacterial incorporation.

2. Experimental program

2.1. Bacteria and their growth conditions

Sporosarcina pasteurii (*S. pasteurii*) PTCC 1645 (DSM 33, ATCC 11859, CCM 2056, NCIB 8841, NCTC 4822) was used in the current study as the calcium carbonate precipitating agent. The bacterium reportedly has the ability to precipitate calcium carbonate given a calcium source and urea via biological cementation [25,27–29]. Nutrient broth-urea medium (8 g nutrient broth, 2% urea, and 25 mM CaCl_2) was used as the culture medium.

The culture was incubated at 37 °C in a shaker incubator operating at 150 rpm for 48 h. Afterwards, the bacterial cells were harvested by centrifuging at 6000 rpm for 10 min. The 48 h-old grown cells were finally washed twice in the saline solution.

2.2. Gram staining

Gram staining is a method of differentiating bacterial species into two large groups (Gram-positive and Gram-negative) based on the chemical and physical properties of their cell walls. Although previous experiments have shown *Sporosarcina pasteurii* used in the current study to be gram-positive and that its cell walls are capable of precipitating carbonate calcite [15,30–34], the Gram staining test was conducted to ensure no gram-negative or other micro-organisms were present in the culture. For this purpose, bacteria were picked up from a previously prepared solid culture and placed on a clean glass slide using a sterile loop. The slide was then heat fixed by passing it several times through a flame; care was taken to maintain the temperature within the required limit since too much heat would lead to the production of staining artifacts and disrupt the normal morphology of the bacterial cells. This was followed by the staining procedure in 4 steps. Step 1 involved flooding the slide with crystal violet ($\text{C}_{25}\text{H}_{30}\text{Cl}$) in 60 s followed by washing with tap water. In step 2, the slide was exposed to Gram's iodine for 90 s to bind it to crystal violet and trap it in the cell; the slide was washed with tap water at the end. As the third step the cells were carefully decolorized with 95% ethanol until the thinnest parts of the smear became colorless before washing its water. In the fourth step, the slide was flooded with safranin, pink color (10% Fuchsine) for 60 s and washed with water. Finally, the slide was allowed to air dry. An Olympus BH2 microscope was used to prepare the relevant micrographs shown in Fig. 1.

2.3. Mix design and concrete construction

Concrete mixes were designed as per ACI 211.2 [35] to obtain a 28-day compressive strength of 25 MPa. For this purpose, fine and coarse lightweight aggregates of Leca were used in addition to Portland cement and a super plasticizer. A detailed description of the concrete mixes is given in Table 1.

2.4. Casting procedure and specimen details

All the specimens were prepared using identical weights and proportions of ingredients; the only difference, however, being the presence or absence of bacteria in the water used in the concrete mix water and using bacterial enhanced aggregates in mixture. Two preparations of the coarse aggregates were made. In

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