Experimental study of the effect of bacillus megaterium bacteria on cement concrete

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Abstract. The presence of voids in cement concrete may lead to a reduction in its performance when exposed to high shrinkage and settlement. This study focused on utilization of bacteria for improving the performance by reducing the voids in the concrete. Bacillus family bacteria were found to be the great healers for the concrete. Bacillus megaterium bacteria of bacillus family with a concentration of $10^8$ CFU have been utilized in the current study. A total of 48 specimens were cast and tested for their mechanical strength and water absorption after 7 and 28 days of curing. The test results indicate that the compressive, split tensile and flexural strengths increased to 12.91%, 10.28% and 9.02% respectively after 28 days of curing as compared to standard M30 grade concrete mix. The water absorption value of bacterial concrete was also found to be less as compared to the standard concrete mix. This is because of the filling of the cracks in concrete due to calcite precipitation produced by the Bacillus megaterium bacteria. Therefore, the Bacillus megaterium bacteria of bacillus family can be effectively utilized to improve the mechanical strength by reducing the voids.

Keywords: Bacillus megaterium, compressive strength, splitting tensile strength, flexural strength, water absorption.

1. Introduction

Concrete is the most useful material in the construction industry to build the road pavement, commercial buildings, residential building and bridges. Development of minor cracks into major cracks in the concrete structures gets exaggerated due to the less tensile strength and reduced durability. This is due to the presence of voids and increased permeability of cement concrete that reduces the durability and strength of the concrete [1]. There are a number of research works that aimed at reducing the utilization of cement by replacing it with some waste materials [2]–[6]. Many research works have been carried out that aims at reducing the voids and permeability in the concrete [7]–[10]. Different methodologies have been adopted for this purpose such as using epoxy, siloxane, acrylics and bacillus family bacteria[11]–[13].Epoxy treatment on cement concrete showed an improvement in the durability and mechanical properties[14]. Ferdous et al. have found that epoxy when used in resins form tends to improve the splitting tensile strength by 2.25 times greater than the standard concrete and shows enhancement in durability against saline solution, air, water and hygrothermal environments [14]. Various research works reveals that the presence of bacteria such as sporosarcina pasteurii, ureolytic and bacillus licheniforms tends to seal the cracks in the concrete [13],
N. Chahal et al. investigated the influence of Sporosarcina pasteurii bacteria with a concentration of 10^5 cells/ml of bacteria on the water absorption and strength in compression of concrete and found that the former got increased by 22% and later got reduced four-times [19]. Santosh A. Kadapure et al. concluded that the optimum concentration of bacillus sphaericus bacterial solution out of 10^3, 10^5 and 10^7 cells/ml along with 10% to 20% replacement of cement with fly ash is 10^5 cells/ml [20]. It has been observed from the previous research works that the mechanical properties of concrete have been significantly enhanced with the presence of bacteria and reduces permeability and water absorption [21] [22]. R. Siddque et al. has found that the mechanical properties of the concrete in terms of its compressive strength have been enhanced due to the presence of the bacteria. The water absorption, porosity and the permeability of the concrete also got reduced due to the presence of the bacteria [23]. N. Balam et al. focused on the tests regarding water permeability and rapid chloride, strength in compression and water absorption with 10^6 cells/ml bacterial solution of S. pasteurii stain and have found that chloride permeability, water absorption decreased by 10% and 20% respectively and strength in compression by 20%[24]. R. Siddque et al. found that the replacement of cement with silica fume in proportions of 5%, 10% and 15% at 10^5 cells/ml bacterial solution shows improvement in the mechanical properties at 28 days of testing and a reduction in chloride permeability, water absorption and porosity due to the presence of bacteria in concrete[25]. The problem of strength and durability due to the presence of voids and increased permeability is also a concern in case of geopolymer concrete[26]–[28]. Various research works have also been reported for the utilization of bacteria and superplasticizer to improve the strength and workability of geopolymer concrete [29]–[32]. Therefore the bacteria have now become one of the prime constituent of the concrete construction for restoring and maintaining the strength and durability properties.

In the current work investigations have been done to study the effect of bacillus megaterium bacteria, which belongs to bacillus family, on the mechanical properties as well as on water absorption of concrete. Bacillus megaterium bacteria have been obtained in powder form from IMTECH (MTCC) Chandigarh and are used with a concentration 10^8 cells/ml.

2. Experimental Program

2.1. Materials

Ordinary Portland cement of 43 grade was used and tested for various properties as per IS: 8112-2013[33]. Soundness and specific gravity of cement were determined as 2mm 3.15 respectively. Initial & final setting times of cement were determined to be 55 minutes and 470 minutes respectively. Natural river sand was used as fine aggregate which lies in Zone-3. Moisture content and specific gravity of fine aggregate were 2.68 and 1.4% respectively. Locally available coarse aggregates with maximum nominal size of 20mm and 10mm were taken in this study. Specific gravity of coarse aggregate of 20mm and 10mm are 2.72 and 2.70 respectively. Bacillus megaterium bacteria of Bacillus family were used. The bacteria have been obtained in powder form from IMTECH (MTCC) Chandigarh. Bacillus megaterium bacteria provide calcite precipitation in the void of concrete specimens during and after healing times that helps in making the structure more compact and durable. PCE based admixture ‘GLENIUM’ superplasticizer was used to enhance the workability of the mix.

2.2. Bacterial Solution

The bacteria have been obtained in powder form and freeze state form from IMTECH MTCC Chandigarh. The powder form of bacteria has been converted into the liquid solution for the purpose of mixing it in concrete. At initial stage, a conical flask having 0.3gm beef extract, 0.5gm peptone and 0.5gm sodium chloride was used to culture nutrient broth media. Meanwhile the pH of media cultured is kept in the range of 7 to 7.4. The bacillus megaterium bacterium feeds on broth media solution cultured. The bacteria in the powder form, using inoculation on Petri dishes, were applied in stripes on agar plate to produce liquid solution of bacteria as shown in figure 1. The dishes were then subjected to incubator at 37°C after 24 hours. To prevent environmental adulteration and further decay of
bacillus megaterium bacteria, autoclave nutrient broth media in a conical flask was used and all the equipments were cleaned and disinfected very carefully. The inoculated liquid solution, kept in the conical flask, was then shifted to the orbital shaker and rotated at a rate of 150 rpm for 24 hours in order to develop the group of bacteria. The bacteria so prepared have been utilized for the concrete mix preparation.

2.3. Mix Preparation

Concrete mix of grade M30 has been prepared by using OPC 43 grade cement, locally available course aggregates of 20 mm (60%) and 10 mm (40%) maximum nominal sizes, natural sand of zone-III, bacillus megaterium bacteria in liquid form having density $10^8$ cell/ml and Polycarboxylic ether based superplasticizer are prepared using of cement, fine aggregate and coarse aggregate. Fine aggregate and coarse aggregate were first mixed in pan mixer with small quantity water for around two minutes in order to minimize dust due to mixing of aggregate in pan mixer. After that the required quantity of cement has been added and the mixing is continued for next two minutes. Then the remaining water along with the bacterial solution were added in pan mixer and mixed for three minutes. The mix is then casted into the desired moulds.

2.4. Casting of Samples

The casting of samples has been done in accordance with IS-1199-1959[34]. After proper mixing, the concrete has been poured into the moulds in three layers. Each layer has been tamped 25 times. The moulds were then compacted to remove the air voids using vibrating table. After compaction these moulds are kept intact for 24 hours. The de-moulding of samples was done after 24 hours and all the specimens were then cured in water till the time of testing. A total of 24 cube specimens of size 150mm x 150mm x 150 mm, 12 cylinder of height 200mm and diameter 100mm and 12 beams of size 100mm x 100mm x 500mm were cast for standard and bacterial concrete. The samples so cast have been tested for the strength in tension, compression and flexural and also for water absorption at desired age of curing. Standard precautions were taken during the preparation of samples and the moulds were tightened properly so that there was no chance of slurry leakage.

2.5. Testing

The cured samples were tested for strength in compression as per IS 516: 1959[35]. The testing was done while the surface of the concrete sample was in wet condition using digital compression testing machine of 200 tonnes capacity. The load was applied at the rate of 2kN/mm2/min and the loads at failure were recorded for all the samples. Cylindrical samples of diameter 150mm and height 300 mm were cast for the test of split tensile strength which was carried out using the same digital compression testing machine according to IS 5816: 1999[36] at 7 and 28 days. The loads at which the test beams failed were recorded and then used to determine the flexural strength. The representative strength, in all the testing, has been considered by taking the average of the strength of three samples measured at 7 and 28 days of curing. To find out the absorption capacity of concrete specimen under
environment condition, water absorption test was conducted in accordance with[37]. The specimens were cast and cured in the same manner as that for compressive strength test. After 7 and 28 days of curing, the cured specimens were taken out from water tank and were dried in oven at 105oC for a period of 24 hours. After drying, the weights (W1) of the specimens were recorded using a digital weighing machine. The specimens were then again kept in curing pond for next 24 hours for the purpose of absorbing water. After 24 hours of water absorption the specimens were taken out and their weights (W2) were recorded again. The water absorption capacities of the samples were calculated using equation (1). The representative water absorption values correspond to the average of the water absorption values of the three specimens.

\[
\text{Water Absorption} = \frac{(W_2-W_1)}{W_1} \times 100
\]  

(1)

3. Result and discussion

A total of 36 samples were cast for the compressive, split tensile and flexural strength test. Additionally 12 samples were cast for water absorption test. The testing has been carried out as per the Indian standard codes i.e. IS: 516-1959[35] and IS 5816: 1999[36]. The testing has been done for bacterial culture mix concrete as per Indian standard testing methods. In this experimental investigation, the effect of bacillus megaterium bacteria on the strength and water absorption properties of concrete samples has been studied. The testing for all the specimens was performed at 7 and 28 days of curing.

3.1. Effect on compression strength

Strength in compression of all the 12 cubes of sides 150 mm was determined using a digital compression testing machine of capacity 2000kN as per the standard procedure given in IS:516-1959[35]. The load is applied at a rate of 140kg/cm2/min and the strength in compression is calculated by dividing the load at failure by the surface area of the tested cube. Figure 2 shows the average of the strength in compression of three specimens at 7 and 28 days of testing, both for the standard concrete and the bacterial concrete.

![Fig. 2 Compressive strength of standard and bacterial concrete](image)

It is evident from Figure 2 that the compressive strength of bacillus megaterium bacterial concrete, having a concentration of $10^8$cells/ml, has found to be improved as compared to the standard concrete at 7 and 28 days of testing. The strength in compression for the bacterial concrete was obtained as 40.59MPa against the 36.23 MPa strength of standard concrete at 7 days of testing. This 12.03%
enhancement in compressive strength of bacteria containing concrete is due to the formation of colonies by the bacteria and the calcite precipitation in the concrete that might have filled the voids thereby improving the density and the strength of concrete[19]. At 28 days of testing the strength of the standard and bacterial concrete was obtained as 52.35 MPa and 59.11 MPa. As compared to the percentage increase in strength at 7 days of bacterial concrete with the standard concrete, the 28 days strength has also shown almost similar increase in strength. This might be due to the fact that most of the colonies and the calcite precipitation by the bacteria has been done in the initial days[20].

3.2. Effect on splitting tensile strength

12 cylinders of 200mm height and 100mm diameter were cast and cured in water for 7 and 28 days and tested for splitting tensile strength using compression testing machine in accordance with IS 5816: 1999[36] and IS: 516-1959[35]. The test results of standard and bacterial concrete have been shown in Figure 3.

The splitting tensile strength test results, as shown in Figure 3, revealed that the strength of bacillus megaterium bacterial concrete is increased as compared to standard concrete both at 7 and 28 days. The improvement in splitting tensile strength is less as compared to the strength in compression. The maximum enhancement in splitting tensile strength at 7 and 28 days of testing due to the incorporation of bacteria in the concrete has been obtained as 9.88% and 10.28% respectively. The increase in splitting tensile strength of bacterial concrete might be due to the deposition of layers of calcite on the surface of concrete samples and this calcite precipitation by bacteria have the ability to fill the pores in the concrete [20].

3.3. Effect on flexural strength

Total of 12 beams of 100mm x100mm x500mm size of standard and bacterial concrete have been cast and left in water tank for curing at 7 and 28 days. Beam samples are taken out from curing tank and tested for flexural strength by applying load using three point load method and recorded the load at which the test beams failed. This load is then used to calculate the flexural strength in accordance with IS:516-1959[35]. The test results of standard and bacillus megaterium bacterial concrete have been shown in Figure 4.
Figure 4 indicate that the flexural strength of bacillus megaterium bacterial concrete has shown almost similar trend of enhancement in strength as that for the compressive and splitting tensile strength at 7 and 28 days of testing. The maximum increase in flexural strength of bacillus megaterium bacterial concrete as compared to standard concrete have been observed as 8.52% and 9.02% at 7 and 28 days respectively. This increase in flexural strength of bacterial concrete is also due to the same reason as that for the increase in splitting tensile strength i.e. the deposition of layer of calcite on the surface of samples and the calcite precipitation by bacteria filling the pores in the concrete [20].

### 3.4. Effect on water absorption

Water absorption values for all the 12 specimens of size 150 mm x 150 mm x 150 mm were calculated using the formula as given in IS: 1124-1974[37]. All the cubes were weighed using digital weighing machine after heat curing in oven for 24 hours and after water curing for 24 hours immediately after oven curing. Results of water absorption test are shown in Figure 5.

Figure 5 reveals that the bacillus megaterium bacteria incorporated concrete tends to decrease the value of water absorption as compared to standard concrete. The water absorption of bacillus megaterium bacterial concrete has reduced by 5.25% and 7.35% at 7 and 28 days respectively. “The deposition of a layer of calcite crystals near the cells and the surface of bacterial concrete resulted in less water absorption compared to conventional concrete”[20]. Test results revealed that water
absorption of controlled concrete has been significantly reduced by employing bacteria in the matrix of concrete[21].

4. Conclusion

In this paper investigations have been carried out to study the effect of bacillus megaterium bacterial solution on the mechanical properties and water absorption of cement concrete. Based on the laboratory investigation, following conclusion can be drawn from this investigation:

1. Bacillus megaterium bacteria were found capable of filling the pores in the concrete[1]. Strength in compression of the concrete gets increased by 12.03% and 12.91% at 7 and 28 days respectively as compared to standard cement concrete.

2. The deposition of layers of calcite on the surface of concrete specimens improves the splitting tensile strength and the flexural strength[20]. The maximum increase in splitting tensile strength at 7 and 28 days of testing, due to the incorporation of bacteria in the concrete, has been obtained as 9.88% and 10.28% respectively and the maximum increase in flexural strength of bacillus megaterium bacterial concrete as compared to standard concrete have been observed as 8.52% and 9.02% at 7 and 28 days respectively.

3. The water absorption of bacillus megaterium bacterial concrete has reduced due to the filling of the voids by bacteria[21]. The water absorption in this study has reduced by 5.25% and 7.35% as compared to standard concrete at 7 and 28 days respectively.

4. Therefore the bacillus megaterium bacteria can be effectively utilized to improve the strength and durability properties of the concrete.

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