



Effect of Using Bacteria in Different Combinations in Concrete

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Abstract : The objective of this research work is to isolate and identify calcite precipitating bacteria and to check the suitability of these bacteria for use in concrete to improve its strength. Bacteria to be incorporated in concrete should be alkali resistant to endure the high pH of concrete and endospore forming to withstand the mechanical stresses induced in concrete during mixing. They must exhibit high urease activity to precipitate calcium carbonate in the form of calcite. Bacterial strains were isolated from alkaline soil samples of a cement factory and were tested for urease activity, potential to form endospores and precipitation of calcium carbonate. . Experimental work was carried out to assess the influence of bacteria on the compressive strength and tests revealed that bacterial concrete specimens showed enhancement in compressive strength. The efficiency of bacteria toward crack healing was also tested. Substantial increase in strength and complete healing of cracks was observed in concrete specimens cast with *B. megaterium*, *B. licheniformis* and *B.pumilus*. This indicates the suitability of these bacterial strains for use in concrete. The enhancement of strength and healing of cracks can be attributed to the filling of cracks in concrete by calcite which was visualized by scanning electron microscope Water and other salts seep through these cracks, corrosion ,and thus reduces the life of concrete. So there was a need to develop an inherent biomaterial.

Key Words : Bacteria; Concrete; Compressive strength; Crack healing.

1.0 Introduction

Researches on the application of bacteria on concrete has started nearly two decades back. This Chapter briefly discuss about various types of bacteria, mechanism of calcite precipitation, previous studies on the application of bacteria.

1.1 Bacteria

Bacteria are single celled microbes. The cell structure is simpler than that of other organisms as there is no nucleus or membrane bound organelles². Instead their control centre containing the genetic information is contained in a single loop of DNA³. Some bacteria have an extra circle of genetic material called a plasmid. The plasmid often contains genes that are responsible for differences between bacteria . For example it may contain a gene that makes the bacterium resistant to a certain antibiotic.

1.1.1 Bacillus megaterium

Bacillus megaterium is a rod-like, Gram-positive, mainly aerobic spore forming bacterium found in widely diverse habitats. With a cell length of up to 4 μm and a diameter of 1.5 μm , *B. megaterium* is amongst the biggest known bacteria^{4,6}. The cells often occur in pairs and chains, where the cells are joined together by polysaccharides on the cell walls

1.1.2 Bacillus licheniformis

Bacillus licheniformis is cultured in order to obtain protease for use in biological laundry detergent⁵. The bacterium is well adapted to grow in alkaline conditions, so the protease it produces can withstand high pH levels, making it ideal for this use - the other components of detergents create an alkaline pH. The protease has a pH optimum of between 9 and 10 and is added to laundry detergents in order to digest, and hence remove, dirt made of proteins. This allows for much lower temperatures to be used, resulting in lower energy use and a reduced risk of shrinkage of garments or loss of colored dye

1.1.3 Bacillus pumilus

Bacillus pumilus contains one circular chromosome including about 4000 genes and 3600-3900 proteins with varying length in the range of 3.7 to 3.8 Mbp. 41% of the DNA base pairs in B. pumilus are G-C. The cellular structure of B. pumilus is similar to other Bacillus species such as B. subtilis, B^{4,6}. megaterium, and B. cereus, the outer layer of the peptidoglycan cross-links in B. pumilus is covered by teichoic and lipoteichoic acids same as the most other Gram positive bacteria. These acids contain polyglycosyl phosphates with mono- and disaccharides as their monomers that can play a role in adhesion to different surfaces like the host cells⁷. On the other hand, these phosphate groups on the surface of B. pumilus can provide net negative charge on the cell surface that allowing to capture some essential cations such as Ca²⁺ and Mg²⁺ that are necessary for cell life.

2.0 Experimental Programme

2.1 Material

For casting of concrete generally used raw materials are Cement, Fine aggregate (River sand), Coarse aggregate and Water were used. Additionally Bacteria was used to investigate the effect on properties of concrete.

2.1.1 Cement

Ordinary Portland cement were used satisfying all the IS requirements was used in making the concrete. The physical properties of cement were specific gravity 3.15, Specific area (m²/kg) 319.

2.1.2 Fine Aggregate

Sand i.e., fine aggregate obtained locally from nearest river passing through 4.75 IS sieve having fineness modulus -2.60 & confirming to zone-III as per IS: 383-1970. Its specific gravity was 2.74 respective

2.1.3 Coarse aggregate

Blue metal i.e., Coarse aggregate obtained locally. 20 mm Nominal size graded aggregate having fineness modulus - 3.92 & confirming to IS:383-1970 specification. The physical properties of CA were specific gravity 2.72, Water absorption - 0.9%.

2.2 Mix Proportions

The process of selecting suitable ingredients of concrete and determining their relative amounts with the objective of producing a concrete of the required, strength, durability, and workability as economically as possible, is termed the concrete mix design. The mix design was done based on IS 10262-2009

Table 2.1 mix proportions

	Cement	FA	CA	Water
In kg/m ³	437	673	1138	197
Ratio	1	1.54	2.6	0.45

3.0 Results And Discussion

The result of experimental investigation carried out to determine Compressive strength, Split tensile strength, Flexural strength, are discussed here in after.

3.1 Compressive strength

The average compressive strength of concrete cube at 7 days 26.66 N/mm² and which is increased by 47% after 28 days which attains the value of 38.98 N/mm²

Table-3.1 Compressive Strength for Cube

S.NO	Specimen	7 DAYS	28 DAYS
1.	C1	26	38
2	C2	28	39.5
3	C3	26	40
Average		26.66	38.9

3.2 Split tensile strength

The average split tensile strength of concrete cylinder at 7 days 1.74 N/mm² and which is increased by 44% after 28 days which attains the value of 2.51 N/mm²

Table-3.2 Split Tensile Strength for Cylinder

S.NO	Specimen	7 days	28 days
1.	C1	1.7	2.5
2	C2	1.79	2.56
3	C3	1.74	2.45
Average		1.74	2.51

3.4 Flexural strength

The average flexural strength of concrete beam at 28 days which attains the value of 5.8 N/mm²

Table-3.3 Flexural Strength for Beam

S.NO	Specimen	28 DAYS
1.	C1	5.6 N/mm ²
2	C2	6 N/mm ²
3	C3	5.6 N/mm ²
AVERAGE		5.8 N/mm ²

4.0 Quantification of Crack Healing

Crack formation is a commonly observed phenomenon in concrete structures. Although micro crack formation hardly affects structural properties of constructions, increased permeability due to micro crack networking may substantially reduce the durability of concrete structures due to risk of ingress of aggressive substances particularly in moist environments[1]. In order to increase the often observed autogenously crack-healing potential of concrete, specific healing agents can be incorporated in the concrete matrix.

5.0 Micro Structural Analysis of Bacterial Concrete

5.1 Creation of cracks

Crack was created in the concrete specimens by introducing a thin copper plate of 0.3mm thickness in the fresh concrete paste up to a depth of 100mm. The plate will be removed during demoulding after 24h resulting in prism with a narrow groove on the upper surface, with a depth of 10mm and a width of 0.3mm.



fig 5.1 placing copper sheet



fig 5.2 artificial cracking

5.1.1 Crack repair techniques

Crack is to be repaired by the use of CaCO_3 precipitating bacteria. The surface treatment with bacterial calcite deposition will consist of injecting the cells of the three bacterial strains mixed with silica gel into the crack after 28 days of curing of concrete specimens. The three bacterial strains to be used are *Bacillus megaterium*, *Bacillus licheniformis* and *Bacillus pumilus*. Two traditional repair techniques namely epoxy coating and cement grout are to be used as surface treatment of cracks. The results of traditional repair techniques are to be compared with surface treatment by bacterial calcite deposition.



fig 5.3 self healing after 60 days curing control concrete



fig 5.4 self healing after 60 days curing bacterial concrete

5.2 Sem (Scanning Electron Microscope) Analysis

SEM is a high magnification microscope. The calcite precipitation by bacterial isolate in the micro cracks and pores in concrete samples is to be analysed using SEM. SEM photographs are to be obtained using Jeol JSM – 6390 apparatus at an accelerating voltage of 0.5 to 30 kV. Broken pieces of cube samples obtained from compressive strength test are to be collected and dried at 100°C in oven for 3 days. Samples are to be gold coated with a sputter coating prior to examination.

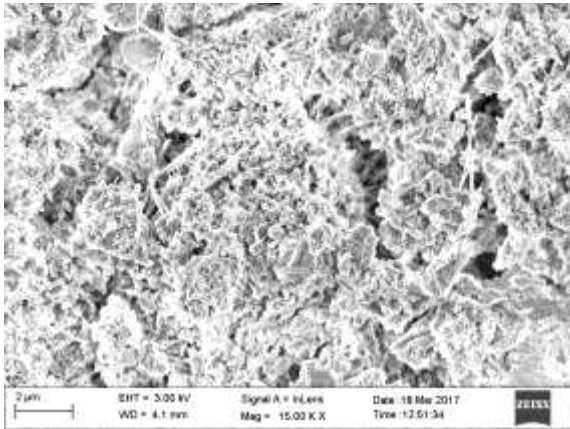
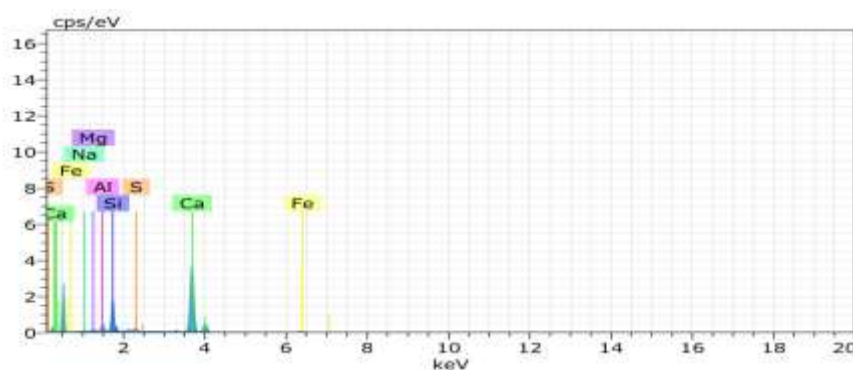


fig 5.5 sem analysis of bacterial concrete

5.3 Edax Analysis

Energy-dispersive X-ray spectroscopy (EDS, EDX, EDXS or XEDS), sometimes called energy dispersive X-ray analysis (EDXA) or energy dispersive X-ray microanalysis (EDXMA), is an analytical technique used for the elemental analysis or chemical characterization of a sample. It relies on an interaction of some source of X-ray excitation and a sample. Its characterization capabilities are due in large part to the fundamental principle that each element has a unique atomic structure allowing a unique set of peaks on its electromagnetic emission spectrum^[2] (which is the main principle of spectroscopy). To stimulate the emission of characteristic X-rays from a specimen, a high-energy beam of charged particles such as electrons or protons (see PIXE), or a beam of X-rays, is focused into the sample being studied. At rest, an atom within the sample contains ground state (or unexcited) electrons in discrete energy levels or electron shells bound to the nucleus^[2]. The incident beam may excite an electron in an inner shell, ejecting it from the shell while creating an electron hole where the electron was. An electron from an outer, higher-energy shell then fills the hole, and the difference in energy between the higher-energy shell and the lower energy shell may be released in the form of an X-ray. The number and energy of the X-rays emitted from a specimen can be measured by an energy-dispersive spectrometer. As the energies of the X-rays are characteristic of the difference in energy between the two shells and of the atomic structure of the emitting element, EDS allows the elemental composition of the specimen to be measured



graph 5.1 EDAX analysis

6.0 Conclusion

The important conclusions drawn from the above study are listed as follows:

1. To improve the properties of cement mortar or concrete the appropriate bacteria should be selected judiciously. For example, *Bacillus pumilus* could not survive in the given environment whereas another *Bacillus* species *Bacillus sphaericus* survived.

2. Crack is to be repaired by the use of CaCO₃ precipitating bacteria. The surface treatment with bacterial calcite deposition will consist of injecting the cells of the three bacterial strains mixed with silica gel into the crack after 28 days of curing of concrete specimens
3. Compressive strength (at 7-day and at 28-day) of mortar cube found to be increasing with the increase of bacteria concentration up to 10⁷ cells/ml.
4. The optimum doses of bacteria found to increase the average compressive strength by 113% (at 7-day) and 77% (at 28-day) over the control specimen. The more increase in strength after 7 day curing may be due to the presence of nutrient medium and it getting depleted as it reaches 28 days and causing death of bacteria
5. The minimum cumulative water absorption is obtained for a cell concentration of 10⁹ cells/ml. Optimum dose of bacterial cell concentration found to increase the cumulative water absorption over the control specimen.
6. The morphology of the bacterial calcite was found out by FESEM. It shows the direct involvement of bacteria in calcite production. We can see rod shaped impressions which is consistent with the dimensions of the bacteria on the calcite crystals. This is matching with the previous study
7. The XRD analysis was conducted for bacterial and control mortar cubes. The XRD result after 28 day curing shows the presence of more calcite peaks in bacterial mortar sample than the control specimen. Presence of more calcite peaks signifies the presence of more calcite in the sample.
8. A layer was observed to be formed over curing water of bacterial specimen after a few days. The XRD analysis of this layer confirmed that this layer is of calcite. This layer was not observed on the curing solution with control specimen. It can be concluded from this information that calcite was produced by bacteria which is responsible for the improved compressive strength of mortar cube.

7.0 References

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