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An Experimental Investigation on Properties of High Strength Bacterial Concrete (Bacillus Subtilis)

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Abstract - *Concrete is a very vital component among construction materials which is widely been in use in infrastructure. Despite of its vital usage for construction purposes, it still has several limitations. It is estimated that production of cement alone contributes to about 7% of global anthropogenic CO_2 emissions which is responsible for green house effect resulting in global warming. A novel eco friendly technique of self healing is amongst one of the approach to investigate the crack healing mechanism in enhancing properties of concrete. Microbiologically Induced Calcite Precipitation (MICP), is a process wherein a highly impermeable layer of calcite precipitate formed over the surface of an already existing concrete layer, due to microbial activities of the bacteria which seals the cracks in the concrete structure.*

This paper focuses on the basic process involved in formation of bacterial concrete and outlines the experimental studies carried out for investigation in the enhancement of the strength parameters of bacterial concrete. The microstructure analysis of bacterial concrete was done using SEM which revealed distinct calcite crystals formed in concrete and thus indicated that the bacteria served as the nucleation sites for the mineralization process.

***Key Words*: Bio-calcification, Calcium Carbonate, bacterial concrete, calcite precipitation, self-healing, sustainable, MICP**

1. INTRODUCTION

Concrete is the most basic component which is widely used in public infrastructure/buildings and its serviceability is often difficult, yet a lot of service is required frequently. It has a little resistance to cracking, weak in tension and has limited ductility. Also, failures like corrosion can result in structural failures with possibly serious long term operational consequences. Taking into account, the consistent research works done across the world; various steps have been taken timely to overcome such shortcomings in cement concrete.

Recent research has shown that biotechnology can actually be a helpful tool to repair cracks in and pre-existing concrete structures by using specific species of bacteria in concrete. This new type of concrete, that is prepared to repair itself, shows a potentially huge enhancement in public infrastructure's service-life, thereby considerably reducing the maintenance costs and lowering CO_2 emissions.

1.1 The Self Healing Process

It is essential to comprehend the bacteria types that will survive in the concrete, how they work to enhance the life-span of the structure, what sort of catalyst will initiate the process of chemical reaction in the bacteria and what would happen to those particular species of specialized bacteria when they are exposed to the catalyst, and how they work together for not only healing the cracks, but also strengthening the overall structure into which they are incorporated.

When the bacteria are given exposure to the elements such as "oxygen" and "food," they go through a chemical process that causes them to interwine, thus filling up the crack formed, and strengthening the concrete structure. In this way, it helps to extend the structure lifespan and fixing the damage caused. The process of healing a crack can take a couple of days.

1.2 Chemistry of the Process

STEP 1	$CO(NH_2)_2+H_2O \rightarrow NH_2COOH+ NH_3$	Urea gets hydrolyzed
STEP 2	$NH_2COOH + H_2O \rightarrow NH_3 + H_2CO_3$	Ammonia released
STEP 3	$H_2CO_3 \rightarrow 2 H^+ + 2CO_3^{2-}$	Bicarbonate formed
STEP 4	$NH_3 + H_2O \rightarrow NH_4^- + OH^-$	Ammonium and hydroxide ion giving rise to pH.
STEP 5	$Ca^{2+} + CO_3^{2-} \rightarrow CaCO_3$	Precipitate formed

Table -1: Steps involved in MICP Process

Bacterial concrete is a new approach to an old thought that a microbial mineral precipitate constantly forms in a natural environment and causes healing. This concrete utilizes calcite precipitation by suitable species of bacteria; hence it is called Microbial or Bacterial concrete. Preparation of such type of concrete can be done by adding spore forming bacteria in the concrete that are able to continuously do precipitation of calcite, this process of formation of layer of calcite precipitate is known as Microbiologically Induced Calcite Precipitation (MICP).

In MICP process, Microorganisms draw cations including which includes calcium (Ca^{2+}) from the environment to deposit on the cell surface. The bacteria can thus act as a nucleation site which helps in the calcite precipitation, hence, joining the pores of the concrete. The MICP process contains of a series of complex biochemical reactions. As a metabolism function, Bacteria produce urease, which helps to catalyze urea to produce CO_2 and NH_4,that results in pH increase in the surroundings where Ca^{2+} and CO_3^{2-} ions precipitate. These create crystals of $CaCO_3$ that further keeps on expanding in process as the bacteria consume up the food i.e. calcium lactate and this process continues till the entire gap is filled. [6]

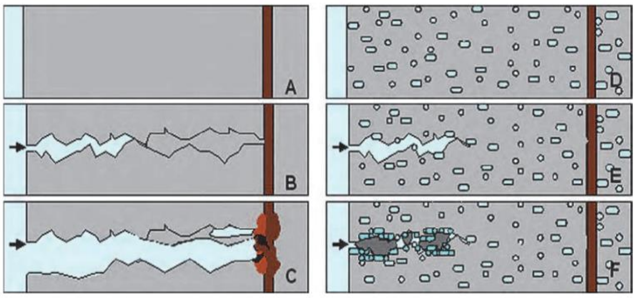

Figure -1: Ingress of moisture restricted by Calcite precipitation

It is found that precipitation of calcite (see Figure 1) which was a result of metabolic activities by bacterial cultures in concrete enhances the properties of concrete like the compressive strength and durability of concrete resulting in lesser corrosion, thus, improvement in the overall strength of concrete structures.

1.3 Types of Bacteria

There are different species of Bacteria which are utilized for making bacterial concrete and enhance the strength and durability of concrete structures. According to the literature review, following are different species of the Bacteria which were used in the earlier investigations in the concrete:

APPILCATIONS	TYPES OF BACTERIA
As crack healer	B. pasteurii
	Deleya halophila
	Halomonas eurihalina
	Myxococcus xanthus
	B. megaterium
For surface treatment	B. sphaericus
As water purifier	B. subtilis
	B. sphaericus
	Thiobacillus

Figure -2: Various Bacteria Species

2. EXPERIMENTAL PROCEDURE

The main aim of the present experimental program is to obtain particular experimental data, which helps to comprehend the properties of High Strength Bacterial concrete and also the enhancement in the Strength characteristics. This experimental program is categorized into:

2.1. Culture and Growth of Bacteria (Bacillus Subtilis)

Bacteria culture was supplied in a freeze-dry condition by Microbial Type Culture Collection, (MTCC- an institute of IMTECH), Chandigarh.

2.1.1. Colonial characteristics

Colonies of strain **MTCC 121** were round in shape, convex, smooth and appeared to be translucent on a nutrient agar plate. The colony size reached up to a diameter of 2-3 mm within 24 hours of incubation period in aerobic conditions at 37°C temperature.

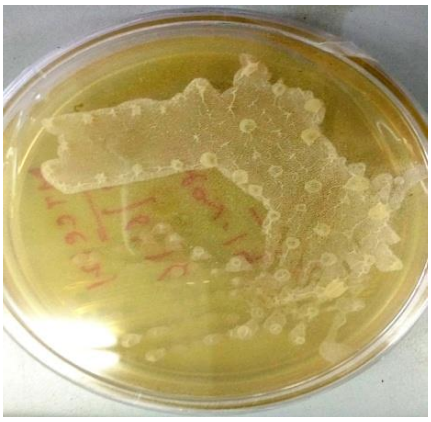

Figure -3: Morphology of colony of strain MTCC 121 on nutrient agar plate.

2.1.2. Analysis from morphological test

Individual cells of strain MTCC 121 came out as Gram positive and long rod shaped.

Figure -4: Phase contrast microphotograph of strain

The mediums prepared for bacteria (MTCC 121) culture were:

- Medium 1-**Nutrient Broth medium** (Peptone, NaCl, yeast extract),
- Medium 2-**Urea medium** (Nutrient Broth, Urea, $NaHCO_3$, $CaCl_2$, NH_4Cl)

2.2. Casting of cubes

Materials Used- Portland Pozzolona Cement, Coarse aggregates (10 mm and 20 mm in equal proportion), Fine aggregates (zone III), Potable Water, and Chemical Admixture (Auramix 200).

2.2.1. Compressive Strength of Mortar Cubes

A set of three cubes (70.6 mm), each of mortar (1:3 cement sand ratio), with different concentrations (nil, 10^4, 10^5, 10^6 and 10^7 cell concentration/ml) of bacteria and different media (nutrient broth medium and urea medium) were casted using vibrating machine. The compressive strength results at 7 and 28 days are obtained. (Table 1 and Table 2).

Cell concentration/ml of mixed water in Nutrient Broth medium	Cement Mortar's Compressive Strength in MPa	
	Average Compressive Strength (7 Days)	Average Compressive Strength (28 Days)
Nil (Control)	19.5	27
10^4	20	28
10^5	22	32.2
10^6	25.4	36
10^7	21.2	30.5

Table -2: Compressive Strength of Bacterial Cement Mortar in Nutrient broth medium

Cell concentration/ml of mixing water in Urea medium	Cement Mortar's Compressive Strength in MPa	
	Average Compressive Strength (7 Days)	Average Compressive Strength (28 Days)
Nil (Control)	19.5	27
10^4	21.5	30
10^5	26.6	33.5
10^6	25	31.6
10^7	22	28.2

Table -3: Compressive Strength of Bacterial Cement Mortar in Urea medium

The optimized value were obtained as 10^6 Cell concentration/ml of mixed water in Nutrient Broth medium and 10^5 Cell concentration/ml of mixing water in Urea Based medium.

2.2.2. Compressive Strength of Concrete Cubes

MATERIAL	CODE PROVISION	PROPERTIES
Mix Propertion (M50)	IS 10262-2009	1 : 1.32 : 2.84 (w/c = 0.35)

Table -4: Mix Design

The concrete mix design (Table 4) was carried out and the optimized amounts of bacteria with both media were mixed with it. Conventional concrete were casted as well in parallel. The compression testing of cubes (150 mm) was done at 7 and 28 days, confirming to IS 516 and results are obtained (see Figure 5).

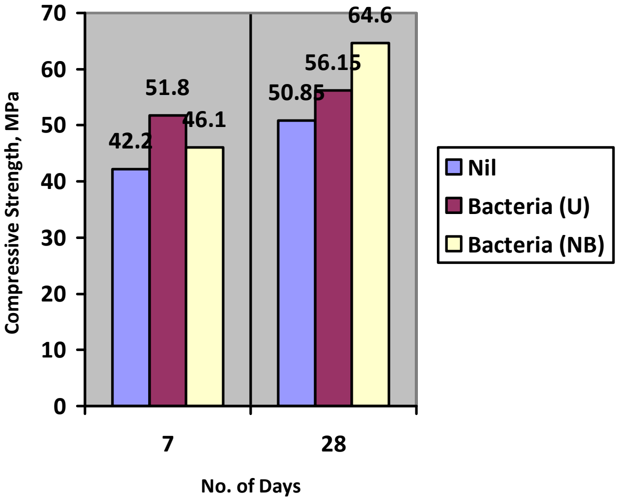

Figure -5: Compressive Strength at 7 and 28 days

2.2.3. Split Tensile Test

Cylinders of size 100x200 mm were also casted in a similar and tensile strength was obtained (see Figure 6).

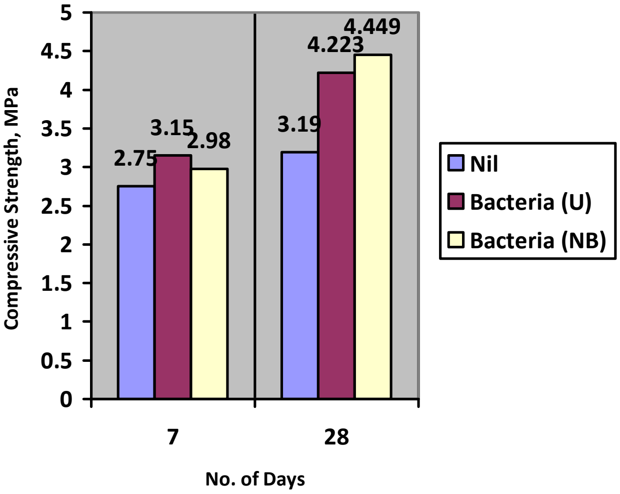

Figure -6: Split Tensile Strength at 7 and 28 days

2.3. Scanning Electron Microscopy

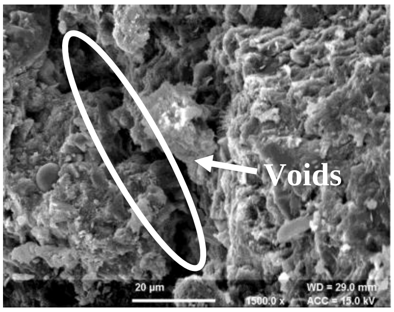

Figure -7 (a): Conventional Concrete (no bacteria)

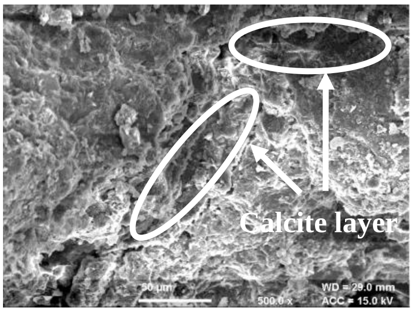

Figure 7 (b): Bacterial Concrete (NB Medium)

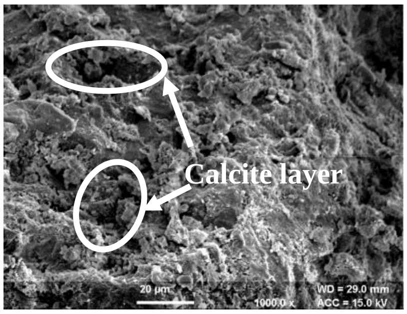

Figure -7 (c): Bacterial Concrete (Urea Medium)

3. CONCLUSIONS

Following are the results and conclusions drawn:

- At an optimum cell concentration of bacteria, the strength achieved is maximum stage.
- Compressive strength of concrete at 7 days increases about 22.7% for urea medium and 9.2% for NB medium.
- Compressive strength of concrete at 28 days increases about 10.4% for urea medium and 27% for NB medium.
- Tensile strength of concrete at 7 days increases about 14.54% for urea medium and 8.36% for NB medium.

- Tensile strength of concrete at 28 days increases about 32% for urea medium and 39.4% for NB medium.
- Bacterial concrete with Nutrient Broth (NB) medium achieved greater strength as compared to Urea medium at 28 days due to better adaptability for NB medium.
- SEM analysis shows the formation of calcite layer in bacterial concrete.

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BIOGRAPHIES

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