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Experimental Study on Bacterial Concrete using Ureolytic Micro-organism

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ABSTRACT

Concrete structures usually show some self-healing capacity, i.e. the ability to heal or seal freshly formed micro-cracks. This property is mainly due to the presence of non-hydrated excess cement particles in the materials matrix, which undergo delayed or secondary hydration upon reaction with ingress water. In this research project we develop a new type of self-healing concrete in which bacteria mediate the production of minerals which rapidly seal freshly formed cracks, a process that concomitantly decreases concrete permeability, and thus better protects embedded steel reinforcement from corrosion. Nowadays , a novel technique is based on the application of bio-mineralization of bacteria in concrete . In present Microbiologically Enhanced Crack Remediation (MECR) technique is used for remediation which involves a selective microbial plugging process, in which microbial metabolic activities promote calcium carbonates precipitation , thus reducing the pore concentration and increase in compressive strength and durability. In this investigation, the results of a study carried out to investigate the ability of Ureolytic bacteria to enhance the compressive strength and split tensile strength, of concrete. The urease producing aerobic alkalophilic bacteria Bacillus subtlis was used in the present study. Ureolytic bacteria used in the present studies were isolated from various sources like cowshed, poultry farm, milk, soil and pigeon dung. Four different cell concentration (0, 10 3 ,10 5 , 10 7cells/ml) of bacteria were used in making the concrete mixes. Tests were performed on standard size of cubes, and cylinders as per Indian standards for compressive strength, and split tensile strength at 7 days, and 28 days.

Keyword : *Bacterial concrete, Ureolytic bacteria, MECR*

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1. INTRODUCTION

Concrete is an absolutely essential component of construction materials used in infrastructure and most buildings. Despite its versatility in construction, it is known to have several limitations. A lot of research has been carried out around the globe to improve properties of concrete. Recently, it has been found that microbial mineral precipitation resulting from metabolic activities of favourable microorganisms in concrete can improve the overall behaviour of concrete. In this technique ureolytic bacteria (microorganism) are used hence the concrete is called Bacterial or Self healing or bio or Microbial concrete. The “Microbial concrete” can be prepared by adding spore forming bacteria in the concrete that are able to continuously precipitate calcite, this process of production of calcite precipitation is called Microbiologically Induced Calcite Precipitation (MICP).

Microbial calcite precipitation is mainly due to ureolytic activity and carbonate bio mineralization of bacteria. Under suitable conditions, most bacteria are capable of inducing carbonate precipitation. In addition, carbonate particles can also be produced by ion exchange through the cell membrane. The basic principle for this process is that the microbial urease hydrolyzes urea to produce ammonia and carbon dioxide and the ammonia released in surrounding subsequently increases pH, leading to accumulation of insoluble calcium carbonate. Bacterial $CaCO_3$ formation through urea hydrolysis is known as Bacterial Calcite Precipitation (BCP). Bacterial Cultures improves the strength of cement sand mortar and crack repair on surfaces of concrete structures. Bacterial Calcite Precipitation (BCP) is highly desirable because it is pollution free and natural. There are several applications of BCP; the most important is strength development. Bio concrete has many advantages over the ordinary concrete; it needs a much shorter time; it is suitable for in-situ process; raw material of bio cement are produced at a lower temperature and can be used as eco-construction material as it consumes less energy and less CO2 . Bacteria to be incorporated in concrete should be alkali resistant to endure the high pH of concrete.

The "Bacterial Concrete" can be made by embedding bacteria in the concrete that are able to constantly precipitate calcite. Several bacteria have the ability to precipitate calcium carbonate. These bacteria can be found in soil, sand, natural minerals. The selection of the bacteria is depend on the survive capability of bacteria in the alkaline environment. Most of the microorganisms die in an environment with pH value of 10 or above. The use of microbial concrete in Civil Engineering has become increasingly popular. From enhancement in durability of cementious materials to improvement in sand properties, from repair of limestone monuments, sealing of concrete cracks to highly durable bricks, microbial concrete has been successful in one and all.

This technology also offers the advantage of being novel and eco- friendly. This special kind of concrete has multiple usages.With recent encouraging reports on compressive strength enhancements achieved in conventional concrete through Microbiologically Induced Calcium Carbonate Precipitation (MICCP). The present study was aimed at isolation and characterization of more efficient urease producing bacterial strains for improving the strength of cement concrete.

1.1 MICCP Process

MICCP also known as Microbiologically Induced Calcium Carbonate Precipitation is done by a general type of bacteria called the urease positive bacteria. These are common type of bacteria found on lime mines and are cause of lime deposition in lime mines. The bacteria produces an enzyme called urease positive enzyme, which breaks down urea present into ammonia and carbonate ion. This carbonate ion inturn combines with the calcium salt to produce impermeable calcite layer.

1.2 History Of Microbiology

Bacteria (singular: bacterium) are relatively simple, single-celled (unicellular) organisms. Their genetic material is not enclosed in a special nuclear membrane, bacterial cells are called prokaryotes. Typically a few micrometers in length, bacteria have a wide range of shapes, ranging from spheres to rods and spirals. Bacteria are ubiquitous in every habitat on Earth, growing in soil, acidic hot springs, radioactive waste, water, and deep in the Earth's crust, as well as in organic matter and the live bodies of plants and animals. There are typically 40 million bacterial cells in a gram of soil and a million bacterial cells in a millilitre of fresh water; in all, there are approximately five nonillion (5 x 10 3) bacteria on Earth, forming much of the world's biomass. Bacteria are vital in recycling nutrients, with many steps in nutrient cycles depending on these organisms, such as the fixation of nitrogen from the atmosphere and putrefaction. However, most bacteria have not been characterized, and only about half of the phyla of bacteria have species that can be grown in the laboratory.The study of bacteria is known as bacteriology, a branch of microbiology.

2. INVESTIGATION

The main objective of the present experimental investigations is to obtain specific experimental data, which helps to understand the Bacterial concrete and its characteristics. In the present experimental investigation, studies have been carried out on the material which is used for this investigation and also investigate behaviour of fresh and hardened properties of ordinary grade concrete and standard grade concrete with and without addition of Bacteria. The hardened properties like compressive strength of concrete, compressive strength and split tensile strength of concrete are determined by conducting suitable laboratory tests on concrete in hardened state.

2.1 Methodology

2.2 Material used and their properties

The ordinary concrete used in the test program consisted of cementing materials, mineral aggregates and corrosion inhibitor with the following specifications:

1. Ordinary Portland Cement (53 Grade)
2. Graded fine aggregates.
3. Graded coarse aggregates.
4. Water.
5. Ureolytic bacteria

1. CEMENT

Portland cement of 53 grade available in local market was used in the investigation. The cement used was tested for various properties as per IS: 4031-1988 and found to be confirming to various specifications of IS:12269-1987 having specific gravity of 3.15.

2. FINE AGGREGATE

Natural river sand well graded passing through 4.75mm sieve having fineness modulus 2.71 and specific gravity of 2.64 was used.

3. COARSE AGGREGATE

Locally available coarse aggregate with equal proportion of size 20 mm conforming to IS: 383-1970 having fineness modulus 6.613, specific gravity of 2.8 and water absorption 1% were used as coarse aggregate.

4. WATER

Ordinary drinking water available in the construction laboratory was used for casting all specimens of this investigation.

5.BACTERIA

A Ureolytic bacterial solution required for casting cubes and cylinder provided by microbiologist as per required proportion in terms of 10^3, 10^5 , 10^7 (cells/ml).

2.3 Mix Design of M20

Specified minimum strength = 20 N/mm^2

Cement:-Ultratech 53 grade OPC (Refer Table No. 5 of IS: 456-2000)

Workability:
i) Compacting factor = 0.9 ii) Degree of quality control Good

Test Data For Material supplied:

a) CEMENT:-
i) Specific gravity = 3.15
ii) Avg. comp. strength 7 days = 46.5 more than 33.0 OK
28 days = 55.0 more than 43.0 OK

b) COARSE AGGREGATE:-
20mm Graded Crushed stone aggregate
Specific gravity = 2.8
Water absorption = 1.%
Free (surface) moisture = 0

c) FINE AGGREGATE (Coarse sand):-
Type Natural (BODELI)
Specific gravity = 2.6
Water absorption = 0.5%
Free (surface) moisture = 0

Water cement ratio= max 0.60 as per IS: 456-2000 page 20

Table: 1 Material required for 1 metre cubic concrete

Mix proportion 1: 1.58: 3.16: 0.50	
Cement	383kg
Sand	606.8kg
Coarse aggregate	1213.6kg
Water	191.6litre

2.4 Manufacturig Process of Concrete.

Production of quality concrete requires greater care at every stage of manufacture of concrete. If precise care is not taken, and good rules are not observed, the resultant BIO concrete is going to be of bad quality. With the same material if intense care is taken to exercise control at every stage, it will result in good bacterial concrete. The various stages of manufacture of bacterial concrete are:

(a) Batching (b) Mixing (c) Transporting (d) Placing (e) Curing

Batching:
The measurement of materials for making bacterial concrete is known as batching. There are two methods of batching,
1. Volume batching 2. Weight batching

(i) Volume batching**:** Volume batching is not a good method for proportioning the material. The Volume of moist material in a loose condition weights much less than the same volume of dry compacted material therefore volume batching are not used for general purpose.
(ii) Weight Batching**:** Batching by weight is more accurate and leads to more uniform proportioning and quality of concrete. For important concrete structure weight batching system should be adopted. Different types of weigh batchers are available, the particular type to be used, depends upon the nature of the job.

Mixing
The aim of mixing of concrete is to produce a homogenous and uniform bacterial concrete. This means that the different constituted material of concrete be uniformly distributed throughout the concrete mass. There are two methods adopted for mixing concrete:
(I) Hand mixing (II) Machine mixing

Hand Mixing**:** Hand mixing is practiced for small scale unimportant concrete works. As the mixing cannot be thorough and efficient, it is desirable to add 25 percent more cement to cater for the inferior concrete produced by this method.
Machine Mixing**:** The mixing process is carried out in electrically operated mixer. The materials are laid in uniform layers, one on the other in the order – coarse aggregate, fine aggregate and cement. Dry mixing is done to obtain a uniform color. Required amount of bacterial water is added. The workability tests are carried out immediately.

Curing of concrete:
Concrete hardens due to chemical reactions between Portland cement and water. Concrete derives its strength by the hydration of cement particles. The hydration of cement is not a momentary action but a process continuing for long time. The rate of hydration is fast to start with, but continues over a very long time at a decreasing rate. The quantity of the product of hydration and consequently the amount of gel formed depends upon the extent of hydration. After 24 hours of casting the specimens is to be de moulded and is transferred to the curing tank, wherein they will allow to cure for 7 and28 days before testing.

3. TESTS

WORKABILITY TEST
Slump test is used to determine the workability of fresh concrete. Slump test as per IS: 1199 – 1959 is followed. The apparatus used for doing slump test are Slump cone and Tamping rod.

Procedure to determine workability of fresh concrete by slump test.
i) The internal surface of the mould is thoroughly cleaned and applied with a light coat of oil.
ii) The mould is placed on a smooth, horizontal, rigid and 415on-absorbent surface.
iii) The mould is then filled in four layers with freshly mixed concrete, each approximately to one-fourth of the height of the mould.
iv) Each layer is tamped 25 times by the rounded end of the tamping rod (strokes are distributed evenly over the cross section).
v) After the top layer is rodded, the concrete is struck off the level with a trowel.
vi) The mould is removed from the concrete immediately by raising it slowly in the vertical direction.
vii) The difference in level between the height of the mould and that of the highest point of the subsided concrete is measured.

viii) This difference in height in mm is the slump of the concrete.

Reporting of Results

The slump measured should be recorded in mm of subsidence of the specimen during the test. Any slump specimen, which collapses or shears off laterally, gives incorrect result and if this occurs, the test should be repeated with another sample. If, in the repeat test also, the specimen shears, the slump should be measured and the fact that the specimen sheared, should be recorded.

COMPRESSIVE STRENGTH TEST

Out of many test applied to the concrete, this is the utmost important which gives an idea about all the characteristics of concrete. By this single test one judge that whether Concreting has been done properly or not. For cube test two types of specimens either cubes of 15 cm X 15 cm X 15 cm or 10cm X 10 cm x 10 cm depending upon the size of aggregate are used. For most of the works cubical moulds of size 15 cm x 15cm x 15 cm are commonly used.

This concrete is poured in the mould and tempered properly so as not to have any voids. After 24 hours these moulds are removed and test specimens are put in water for curing. The top surface of these specimen should be made even and smooth. This is done by putting cement paste and spreading smoothly on whole area of specimen. These specimens are tested by compression testing machine after 7 days curing or 28 days curing. Load should be applied gradually at the rate of 140 kg/cm2 per minute till the Specimens fails. Load at the failure divided by area of specimen gives the compressive strength of concrete.

APPARATUS

Compression testing machine

PREPARATION OF CUBE SPECIMENS

The proportion and material for making these test specimens are from the same concrete used in the field.

MIXING

Mix the concrete either by hand or in a laboratory batch mixer

HAND MIXING

(i)Mix the cement and fine aggregate on a water tight none-absorbent platform until the mixture is thoroughly blended and is of uniform color

(ii)Add the coarse aggregate and mix with cement and fine aggregate until the coarse aggregate is uniformly distributed throughout the batch

(iii)Add water and mix it until the concrete appears to be homogeneous and of the desired consistency

SAMPLING

(i) Clean the mounds and apply oil

(ii) Fill the concrete in the molds in layers approximately 5cm thick

(iii) Compact each layer with not less than 35strokes per layer using a tamping rod (steel bar 16mm diameter and 60cm long, bullet pointed at lower end)

(iv) Level the top surface and smoothen it with a trowel

CURING

The test specimens are stored in moist air for 24hours and after this period the specimens are marked and removed from the molds and kept submerged in clear fresh water until taken out prior to test.

PRECAUTIONS

The water for curing should be tested every 7days and the temperature of water must be at 27+-2oC.

PROCEDURE

(I) Remove the specimen from water after specified curing time and wipe out excess water from the surface.

(II) Take the dimension of the specimen to the nearest 0.2m

(III) Clean the bearing surface of the testing machine

(IV) Place the specimen in the machine in such a manner that the load shall be applied to the opposite sides of the cube cast.

(V) Align the specimen centrally on the base plate of the machine.

(VI) Rotate the movable portion gently by hand so that it touches the top surface of the specimen.

(VII) Apply the load gradually without shock and continuously at the rate of 140kg/cm2/minute till the specimen fails

(VIII) Record the maximum load and note any unusual features in the type of failure.

Compressive strength = (Load in N/ Area in mm$^{2)}$=……………N/mm^2

SPLIT TENSILE STRENGTH TEST

This test method is used for the determination of splitting tensile strength of cylindrical concrete specimen.Splitting tensile strength is helpful for the following purposes;
Splitting tensile strength is generally greater than the direct tensile strength and lower than the flexural strength (modulus of rupture).Splitting tensile strength is used in the design of structural light weight concrete members to evaluate the shear resistance provided by concrete and to determine the development length of the reinforcement.

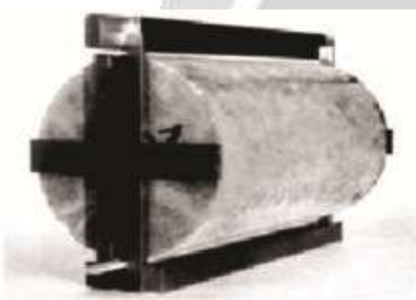
$$T = \pi l d$$

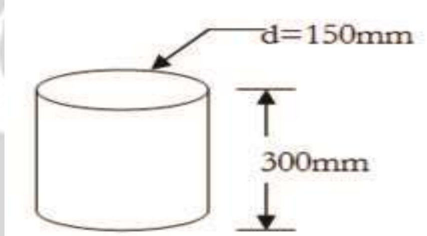
Where, T = Splitting tensile strength (to be reported in 0.05 MPa multiples)
P = Applied load
l = length of the specimen (mm)
d = Diameter of the specimen (mm)

Apparatus

1.Testing Machine
2.Supplementary Bearing Bar Or Plates (If the diameter or the largest dimension of the upper bearing face or the lower bearing block is less than the length of the cylinder to be tested, a supplementary bearing bar or plate of machined steel shall be used. The bar or plate shall be manner that the load will be applied over the specimen.)
3.Bearing Strips (Two bearing strips of nominal 1 /8 in [3.2 mm] thick plywood, free of imperfections, approximately 1 in. [25 mm] wide, and of a length equal to, or slightly longer than, that of the specimen shall be provided for each specimen. The bearing strips shall be placed between the specimen and both the upper and lower bearing blocks of the testing machine or between the specimen and supplemental bars or plates, when used.

Size Of The Specimen





Standard Specimen

The specimen is a cylinder of 150mm diameter and 300mm height. Determine the diameter to the nearest 0.25mm by averaging the three diameters. Determine the length to the nearest 2mm by averaging at least two lengths.

Size Of Bearing Strips

According to ASTM specifications, the bearing strips should be 3.2mm thick and 25mm wide. There is no restriction on their length.

Procedure:

This test method consists of applying a diametrical force along the length of a cylindrical concrete at a rate that is within a prescribed range until failure. This loading induces tensile stresses on the plane containing the applied load and relatively high compressive stresses in the area immediately around the applied load. Although we are applying a compressive load but due to Poisson’s effect, tension is produced and the specimen fails in tension. Tensile failure occurs rather than compressive failure because the areas of load application are in a state of triaxialcompression, thereby allowing them to withstand much higher compressive stresses than would be indicated by a uniaxial compressive strength test result. Thin, plywood bearing strips are used to distribute the load applied along the length of the cylinder.The maximum load sustained by the specimen is divided by appropriate geometrical factors to obtain the splitting tensile strength.

Calculations:-Calculate the splitting tensile strength of the specimen as

$$T = \pi l d$$

4 . EXPERIMENTAL RESULT

Table 2 . Test of cement materials

Test	Value	Value specified by IS 12269 : 1987
Initial setting time	35 min	30min minimum
Final setting time	185 min	600min maximum
Fineness test	10gm retained on sieve	10gm retained on sieve

Table 3 :-Strength of concrete at 7 and 28 days.

Properties	Normal		10^3 (Cells/ml)		10^5 (Cells/ml)		10^7(Cells/ml)	
	7 day N/mm^2	28 day N/mm^2	7 day N/mm^2	28 day N/mm^2	7 day N/mm^2	28 day N/ mm^2	7 day N/mm^2	28 day N/mm^2
Compressive	13.7	25.6	15.5	26.3	12.4	27.4	14.3	29.8
Split	1.41	2.82	1.68	3.1	2	3.3	2.8	3.5

Table 4. comparison of normal concrete and bacterial concrete

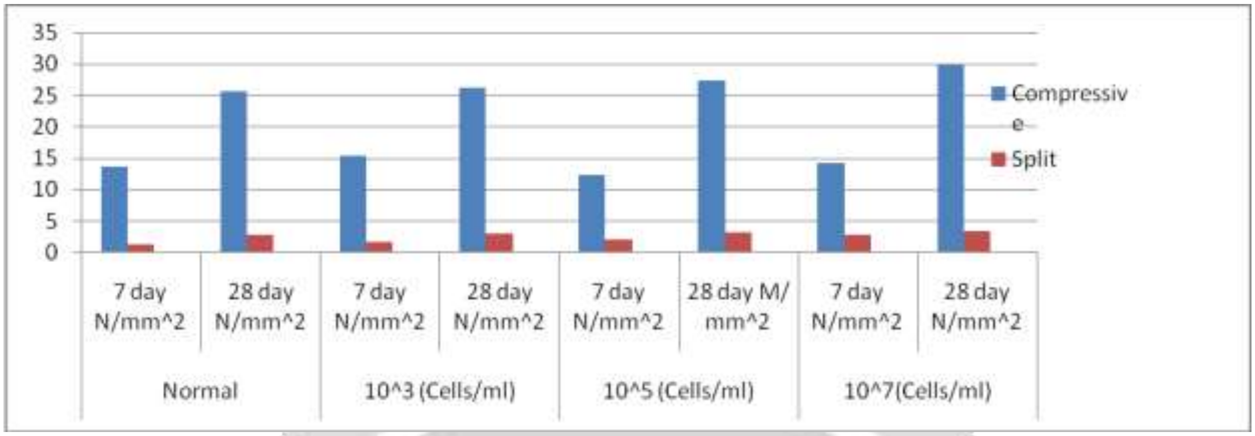


Table 5 Physical properties of coarse aggregate

Test	Aggregate	As per IS 383
Bulk density	1630 Kg/M^3	-
Sp. Gravity	2.8	2.5-3
Fineness modulus	7.4	-
Water absorption	1%	

Table 6:-Sieve analysis of fine aggregate

Sr. No.	IS Sieve Size	Weight Retained (gm)	Cummulative Weight Retained (gm)	Cummulative % Weight Retained	Cummulative % passing	Grading Limits IS 383-1970 Zone II
1	10 mm	0	0	0	100	100
2	4.75mm	6	6	0.6	99.4	90-100
3	2.36mm	45	51	5.1	94.9	75-100
4	1.18mm	190	241	24.1	75.9	55-90
5	600 micron	360	601	60.1	39.9	35-59
6	300 micron	310	911	91.1	8.9	8-30
7	150 micron	75	986	98.6	1.4	0-10
8	< 150 micron	14				
9	**Total**	**1000**		**279.6**		

Fine Modulus = Cummulative % Weight Retained /100

= 279.6/100 = 2.796

Table 7:-Sieve analysis of coarse aggregate (20mm)

Sieve size (mm)	Weight retain (gms)	Cumulative weight retain (gms)	Cumulative percentage weight retain(%)	Cumulative percentage weight passing(%)
20	700	700	0.7	99.3
12.5	7360	8060	80	20
10	1340	9400	94.4	5.6
4.75	610	10000	100	0
2.36	0	-	100	0
1.18	0	-	100	0
Receiver	0	-	100	0
Total	10000	-	-	-

Table 8:-Physical properties of fine aggregate

Test	Fine Aggregate
Bulk density	1831 Kg/M^3
Sp. Gravity	2.6
Fineness modulus	2.7
Water absorption	0.5%

5 . CONCLUSIONS:

Based on experimental investigation carried out, following conclusion is drawn:-

1. Bacillus Subtilus can be produced from the lab which is proved to be safe and cost economic.

2.The use of this biological repair technique is highly desirable because the mineral precipitation induced has a result of microbial activities is pollution free and natural, however further experiments have to be done to examine the durability of this crack technique.

3.From the above it can be concluded that the Bacillus Subtillus can be easily cultured and safely used in improving the performance characteristics of concrete.

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