Potentiality of Yeast Strain On Cement Concrete specimen

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Abstract

The objective of the present study is investigated the potential application of isolated soil yeast *C. Tropicalis* to improve the properties of cement concrete specimen. Parameters for growth conditions were studied. Using injection method, calcium carbonate was precipitated on the surface of concrete specimen which resists water absorption to enhance the durability of concrete. Urease producing *C.tropicalis* was identified by urease test and CHROMagar medium. Crystals were confirmed by microscopic and SEM images.

Keyword: Concrete, *C.tropicalis,* calcium carbonate, urease, water absorption

INTRODUCTION

Ureolytic microorganism makes a vital contribution to the accumulation of carbonate in several natural habitats which is formed by autotrophic pathways can deplete CO2 from the environment, and able to induce CaCO3 precipitation in vitro. The role of microorganisms in a wide range of geological processes such as rock dissolution, rock formation, the transformation of soils and sediments, genesis and the degradation of minerals and fossil-fuel are examined by geo-microbiology which is closely related to microbial ecology and biogeochemistry. Microbes can encourage the dissolution of poorly soluble minerals to soluble compounds such as CaCO3, ion and manganese hydroxides. They can also selectively concentrating or diluting them.

Microorganisms can accumulate inorganic materials through various processes as intracellular deposition, adsorption and cellular fixation, and extra-cellular precipitation of

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insoluble compounds. Bacteria were not only at the centre of the precipitated carbonate crystals but were also evenly distributed between the inner core and the outer surface of the precipitated crystals, as well as being present on the outer surface. Microbes can act as concentrating agent on the study of bio-mineralization (Bharathi and Meyyappan, 2014).

Unicellular fungi are called yeasts. Over the years evidence accumulated that soil yeasts exert a positive effect on soil structure, nutrient recycling and even plant growth. The role of some soil yeasts in soil aggregate formation has been known since 1970s. Some yeast especially affects soil structures as they are able to produce extracellular polymeric compounds that bind soil particles together (Botha, 2011).

The mechanism of urease enzyme activity creates a local microenvironment that allows optimized extra cellular precipitation of minerals. Very low microbes can withstand extreme alkaline environment. Ureolytic microorganisms were selected on the basis of urease producing ability which utilizes urea to produce ammonia in the reaction medium. That medium producing pink colour under the influence of phenol red indicator in general urease test broth was used. The calcium carbonate precipitation was complex mechanism which is a function of cell concentration and pH of the medium including ionic strength. During the mechanism, urea gets degradation which converts carbonic acid and ammonia. Immediately the carbonic acid gets hydrolyse into
ammonia and bicarbonate.

$$NH_2CONH_2+H_2O \rightarrow NH_2COOH+NH_3$$
$$NH_2COOH+H_2O \rightarrow NH_3+H_2CO_3$$

The bicarbonates which are immediately react with calcium ions present in the medium converted to calcium carbonate (Bharathi, 2014).

Concrete which forms major component in the construction industry. And it is cheap, easily available and convenient to cast. Various factors are involved to damage the life of concrete as acid pollution, water absorption, stress and heat. Increased water absorption can decreases the durability of cement concrete.
A novel technique is adopted in re-mediating cracks and fissures in concrete by utilizing Microbiologically Induced Calcite or Calcium Carbonate Precipitation is a technique called bio-mineralization which is highly desirable because the Calcite precipitation induced as a result of microbial activities is pollution free and natural. Research leading to microbial

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Calcium Carbonate precipitation and its ability to heal cracks of construction materials has led to many applications like crack remediation of concrete, sand consolidation, restoration of historical monuments and other such applications. So it can be defined as "The process can occur inside or outside the microbial cell or even some distance away within the concrete. Often bacterial activities simply trigger a change in solution chemistry that leads to over saturation and mineral precipitation. Use of these Bio mineralogy concepts in concrete leads to potential invention of new material called ―Bacterial Concrete" (Mayur et al.,2013)

We are used isolated yeast strain *C.tropicalis* instead of bacterial strain. Previous research has shown that the bacterial strains are able to precipitate calcium carbonate on their cell constituents and in their micro-environment by conversion of urea in to ammonium and carbonate. The yeast strain degradation of urea locally increases the pH and promotes the microbial deposition of calcium carbonate in a calcium rich environment.

MATERIALS AND METHODS

Candida tropicalis

From agricultural soil, sample was collected, using sterile water serially diluted and plated on sabouraud dextrose agar, *Candida tropicalis* colonies are cream colour with mycelial border. They are smooth, glabrous and yeast like in appearance.

Microscopic morphology shows spherical budding yeast like

No capsules present on Indian ink preparation.

Germ tube test shows negative, Hydrolysis of Urea shows positive, Growth on Cyclohexidine medium shows positive, Growth at 37^0 C shows positive

On CHROM agar medium shows bluish purple color.

The following materials were used for concrete making.

Portland cement, sand and aggregates are used by weight in the ratio of 1:2:2 to prepare cement concrete specimen

Urease test

The media composition for urease test are; urea (20g/l), Na_2HPO_4 (9.5g/l), KH_2PO_4 (9.1g/l), SD broth (0.1g/l) and 0.01g phenol. pH was made to 7. This test detects the ability of organism to produce urease enzyme. This enzyme converts urea to ammonia and CO_2, which convert the environment alikaline and turns pink colour referred as urease positive.

Calcite medium/L

Urea 20g, NaHCO3 2.21g, NH4Cl 10g, CaCl2.2H2O 25g, SD broth 3g

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Role of factors for calcite production

Biomass concentration was determined spectrometrically red at 600nm. Maximum obtained 0.165 Optical densities of 48hrs.

Parameters as pH and temperature of Calcium carbonate precipitating medium conditions were optimized at pH 8 and 40°C respectively

Estimation of Calcium ions

Using the standard graph(R2=0.994), our test sample value was generated by carrying titration with EDTA. This was alkalized by using ammonia buffer. End point was obtained by using EBT indicator, which turns steel blue color from reddish pink colour.

Cementation solution

Equimolar mixture of Urea and Calcium chloride in 1L preparation was used after the inoculation of culture applied on the surface of cement concrete specimen.

Water absorption

Prepared specimen was weighed initially and allowed immersed into water overnight. The overnight saturated specimen was weighed accurately. Then dried oven with 30mnts and weighed.

$$\% \text{ water absorption} = \frac{W_{Saturation} - W_{ovendried}}{W_{ovendried}} \times 100$$

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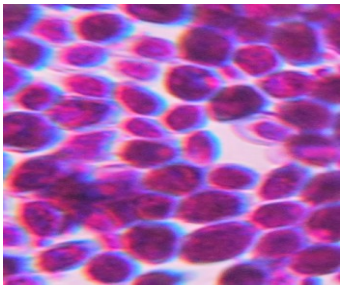
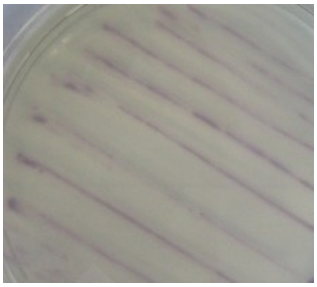
RESULTS

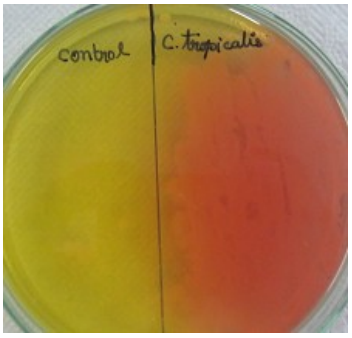
Isolation of *C.tropicalis* from soil

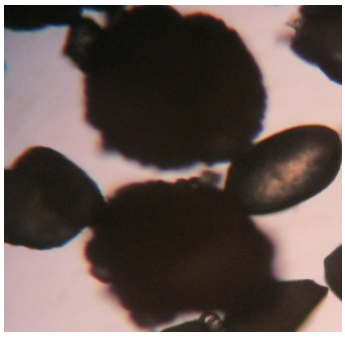
A.  B.  C. 

A. Isolated *C.tropicalis* shows creamy mycelial border on SD broth

B. Gram staining shows sphere shape

C. On CHROMagar shows bluish purple

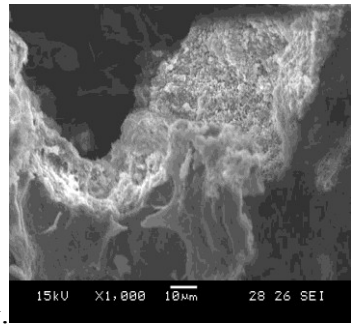
D. 

E.  F. 

D. On Urea agar medium shows pink color

E. Microscopic view of crystals

F. After 48hrs filtered Calcium carbonate

G. 

H.  I. 

G. SEM image for precipitated Calcium carbonate

H. Calcium carbonate precipitation on the concrete specimen

I. Compared with control specimen white precipitates occurred on the test specimen

From the experiment, the calculated absorption for test specimen by percentage was 5.5% and the control specimen without precipitation was observed 10.32% of

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absorption.

Control

Saturated Weight – 89.6g

Dried weight – 81.22g

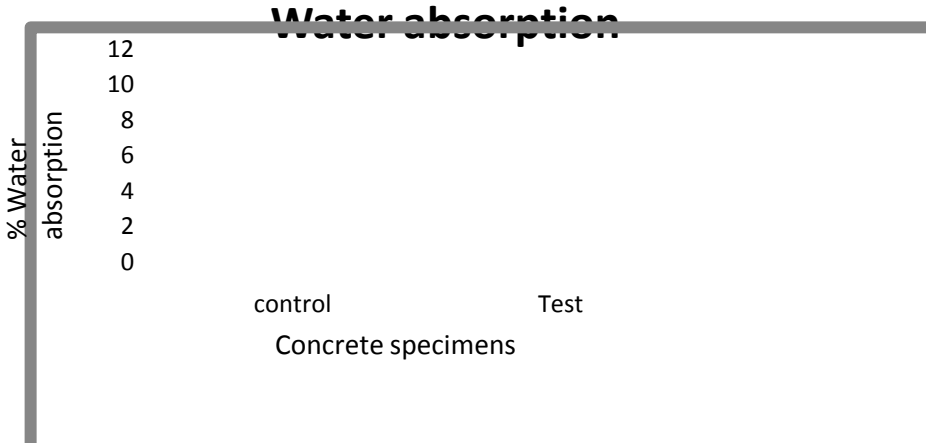
% Absorption – 10.32

Test

Saturated weight – 84.2g

Dried weight – 79.8g

% Absorption – 5.5



CONCLUSION

We conclude that the isolated soil yeast *C.tropicalis* can able to precipitate calcium carbonate on the surface of specimen. May also be able to seal cracks by bio-mineral formation after being revived by water and growth nutrients entering freshly formed cracks, hence the application of yeast will improve the resistance of water absorption and enhance the durability of cement concrete possibly and sufficiently.

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