

Innovations

Reevaluation of Alkaliphilic Bacteria as remedy to settlement – structural crack, time saver and cost-efficient.

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Abstract

This study investigated the self-healing concrete (concrete that heal crack itself) and different type of bacillus subtilis bacteria was used during in situ concrete production. The bio mineralization of bacteria in concrete, a biological process commonly seen in few forms of microorganisms. The strength and durability can be increased by using these microorganisms as binders and fillers in concrete. Epoxy treatment is currently used for the repair works which is harmful to the environment and health as toxic fumes and gases evolved may cause serious skin and breathing issues. Hence the use of biological techniques should be focused. The use of biological techniques in concrete lead to the invention of a new building material i.e. bio-concrete. Conventional concrete structures repairs usually involve applying a bonded concrete mortar to the damaged surface. Sometimes, metal pins used to key mortar into the existing structure to prevent been fall away. Underground or great height structures repairs can be particularly expensive and time consuming because of difficulties in gaining access to the structure to mends. Natural processes such as earthquakes, weathering, land subsidence, faults, and human activities generate fissures and fractures historical stone monuments and concrete structures. Structure service life may reduce as fissures and fractures are detrimental In the case of buildings of historic importance and 20 monuments, these cracks tend to disfigure as well ruin the structure. Use of bacterial concrete for remediating these structures will reduce the crack width and increase the strength of the structure.

1. Introduction

Surface opening cracks are a common kind of defects in concrete structures, they permit water penetration or other harmful agents that result in durability loss earlier than expected, so cracks formed renovating and defects becomes indispensable and unavoidable [1-3]. Presently, maintenance and concrete structures repair mostly rely on consistent inspection programmes, which are costly, besides they also depend on a amalgamation of nondestructive testing (NDT) and human sensitivity [5-7]. If there are severe damage, the structural component is changed entirely while repairs are attempted for less extensive damage. Vast amounts of money are spent each year on inspection and repair as direct and indirect costs, the latter often being much higher than the former [8, 9]. For instance, in the USA, the annual economic impact associated with maintaining, repairing, or replacing deteriorating structures is estimated at \$18–21 billion [10]. The American Society of Civil Engineers estimated that \$2.2 trillion are needed for five years, starting from 2012, for repair and retrofit; a cost of \$2 trillion has been predicted for Asia's infrastructure for the

same period [11-13]. Europe spends more than half of its annual construction budget on repair works, while in the UK, repair and maintenance costs account for over 45% of the total expenditure on construction [14, 15]. Moreover, repair works have a significant adverse environmental impact particularly in cases where partial or complete replacement of structures is required [16]. It is known that the production of one tonne of Portland cement (PC), as often being the main constituent on concrete, releases about 0.85–1.1 tonnes of CO₂ [17, 18]. Approximately 3.6 x 10⁹ tonnes of cement were produced worldwide in 2015 (USGSMC, 2016). The CO₂ emissions associated with the production of cement are very significant, and are estimated at 7% of the global anthropogenic CO₂ emissions. Therefore, developing innovative technologies to overcome these challenges has become an urgent necessity [3, 19]. Over the past few decades, the notion that concrete can be designed with a sufficient healing capability and heal its cracks without any external aid has been inspiring field of work for many research groups around the world. Broadly, self-healing processes within cement based materials can be divided into two categories: autogenic and autonomic [2, 20].

2. Cracks

According to American Concrete Institute, crack is an incomplete or complete separation of either masonry or concrete into two or more components produced by breaking or fracturing. Cracking is reviewed as an inherent attribute of reinforced concrete structures also can be created by structure loading itself or other mechanisms, e.g. freeze-thaw cycles, thermal effect and drying shrinkage [1, 6, 21]. Cracks function like openings that permit water penetration and ingress of some truculent chemicals into concrete as well related to durability of a reinforced concrete structure [22]. Cracks commence internally at any stage of structure life where there is disfigurement and deterioration, conventional monitoring of structural integrity totality through regular inspections and repairs needed, this may entail corrosion monitoring, surface repairs, surface washing, admixtures and sealant applications. Last decade, building industry take remarkable interest in engineering concrete as a smart material to assuage issues like excessive routine maintenance, immoderate production and costs. Alleviation of micro scale deterioration is the strategy to achieve this through an autonomous technique that percept and repairs cracks in a targeted style [23-25].

2.1. Causes of cracks

In spite of much research the cracks in concrete are inexorable, it can be the outcome of one or integration of factors like restraint (internal or external) to shortening, subgrade settlement, drying shrinkage, applied load and thermal contraction [4]. Cracks that occur prior to hardening usually are the effect of settlement within the concrete mass, or surface shrinkage (plastic-shrinkage cracks) result from water loss while the concrete is still plastic while cracks that happen after hardening are usually drying shrinkage, subgrade settlement or thermal contraction outcome. While drying, hardened concrete will shrivel about 1/16 in. in 10 ft of length. Cracks can also generated by corrosion of reinforcing steel, alkali- aggregate reactivity, saturated concrete freezing and thawing or sulfate attack. Nevertheless, cracks from these sources may not materialize for years [8, 15].

2.2 Major cracks types

2.2.1 Settlement Cracking

Settlement cracking results from excessive slumps (overly wet concrete), inadequate consolidation (vibration), or inadequacy of adequate cover over embedded items. Settlement cracks may evolve over embedded items, like steel reinforcing or as the concrete settles or relents at adjacent to forms or hardened concrete [7].

2.2.2 Plastic-shrinkage cracks

They are relatively short cracks that might happens before final ending on days when temperature is high, humidity is low or wind and most common in slabs. Surface moisture evaporates quickly than it can be substituted by rising bleed water, makes the surface concrete to shrivel more than the interior concrete [10]. Stresses more than concrete's tensile strength develop as the shrinkage of the surface concrete restrains by interior concrete resulting in surface

cracks. Plastic-shrinkage cracks are of different lengths spaced from a low centimeters (inches) up to 3 m (10 ft) diver and often reach to mid-depth of a slab [5, 6].

2.2.3 Structural cracks

In residential foundations structural cracks usually an outcome of horizontal loading or settlement. Most not all structural cracks generating from applied loads are nearly horizontal means parallel to the floor and happens 16” to 48” from the top of the wall. Heavy equipment next to the foundation or Hydrostatic pressure can result to it [7].

2.2.4 Diagonal cracks

Due to stress build-up at corner, they start from windows corner and other openings and it can also be called reentrant cracks with diagonal reinforcement at openings corner it keeps cracks narrow [13].

2.3 Problems associated with structure with crack

Major problem associated with cracking is public perception, structural dweller believes that once cracks sighted in their floors or walls failure has occur. For wall case, if is unstructured cracks, is not too wide (the acceptable crack is from 1/16” to 1/4”) and water not leaking, it should be considered acceptable [20].

3. Some of the methods been used to repair Concrete structure in the past

3.1 Surface Spalling Repair of Underwater Concrete Structures

Underwater structural elements cover can spall off due to adventitious damages. The damaged concrete cover must be substituted and repaired to avert reinforcement corrosion in the future. Faintly deteriorated areas will turn to more grievous and dangerous damages in little time, predominantly in splash zones. Underwater structure damaged area should be emptied from wobbly concrete and marine growth and loose concrete prior to rehabilitation processes commencement. Afterwards, based on the destruction quantity, spalled region boundary should be saw-cut to a depth of 1.2-2 cm. In swash zones, application of cementitious mortar to the damages area and in case of compact damage region use water tolerant epoxy mortar while large repaired region, use formwork to hold rehabilitation repairing materials at its position[23 - 25].

3.2 Injection of cementitious grout or resin can be employed to repair cracks and or voids

The techniques for injection technique are; Preparation of concrete surface along crack length, fixing of inspection nipples at specific intervals along crack length, Sealing of crack surface along the entire length of the crack, Eliminate contamination by applying fresh water and be convinced that injection path is open. Inject cement grout or epoxy resin through nipples into the crack at one end of the crack [4, 21].

3.3 Epoxy resin

Suitable for crack width of 0.1 mm whereas cement grout is appropriate for crack with width greater than few millimeters.

4. Purpose of self-curing agent

4.1 Investigation of different healing mechanisms

Mortar and concrete are the most universally used building materials all across the globe as they are inexpensive, easily accessible and convenient to cast. Yet crack in these materials is a recurrent phenomenon during its life time due to many reasons, if unattended promptly, outcome is long-term structural deterioration with high danger level and maintenance cost. Presently, epoxy mortar, epoxy, resins and other synthetic mixtures are used for cracks repairs. Bacteria from diverse natural habitats have frequently been delineated to precipitate calcium carbonate both in unprocessed and laboratory conditions [11, 14]. Implementation of bacteria based mineralization notion has causes potential invention of a modern biomaterial that can fill the cracks, openings in mortar and concrete materials. Concrete has an autogenous curing ability as unhydrated cement available in the matrix after water meets

the unhydrated cement hydration happens and CaCO₃ crystals formed as dissolved CO₂ reacts with Ca²⁺, these mechanisms may only cure small cracks [3, 6].

4.2 Bacteria as self-curing agent

Bacteria are relatively simple, single celled organisms. These are classified based on three categories, namely, based on shape, gram stain and oxygen demand. Very large group of microorganisms encompass one of the three domains of living organisms. They are prokaryotic, unicellular and free- living in soil or water or parasites of plants or animals [1, 10]. That is any of a large group of one- celled organisms that require a cell nucleus, replicate by fission or by forming spores and in some occasion trigger disease, mostly abundant life forms on earth are found in all livings and in all of the earth’s environments they normally live outside other organisms while they form most of the kingdom of prokaryotes with one group called archaebacteria [5, 8]. Some bacteria are advantageous to humans like those that stay in the stomach and support digestion while some are dangerous like those that causes disease. Bacillus is the only group of bacteria's that are able to survive this high alkaline environment. Finding a suitable food source for the bacteria that could survive in the concrete took a long time and many different nutrients were tried until it was discovered that calcium lactate was a carbon source that provides biomass. If it starts to dissolve during the mixing process, calcium lactate does not interfere with the setting time of the concrete. Some of the bacteria which come under Bacillus genus are: Bacteria were grown at 37°C in *Luria-Bertani*broth (LB) or in minimal medium M63 supplemented with a carbon source at a concentration of 2 g/L [9].

5. Bacteria as concept

Bacteria naturally occur in nature in various forms. They are present not only on the surface but also beneath the surface of the earth. The various bacteria that can be used in concrete are presented in Figure 1.

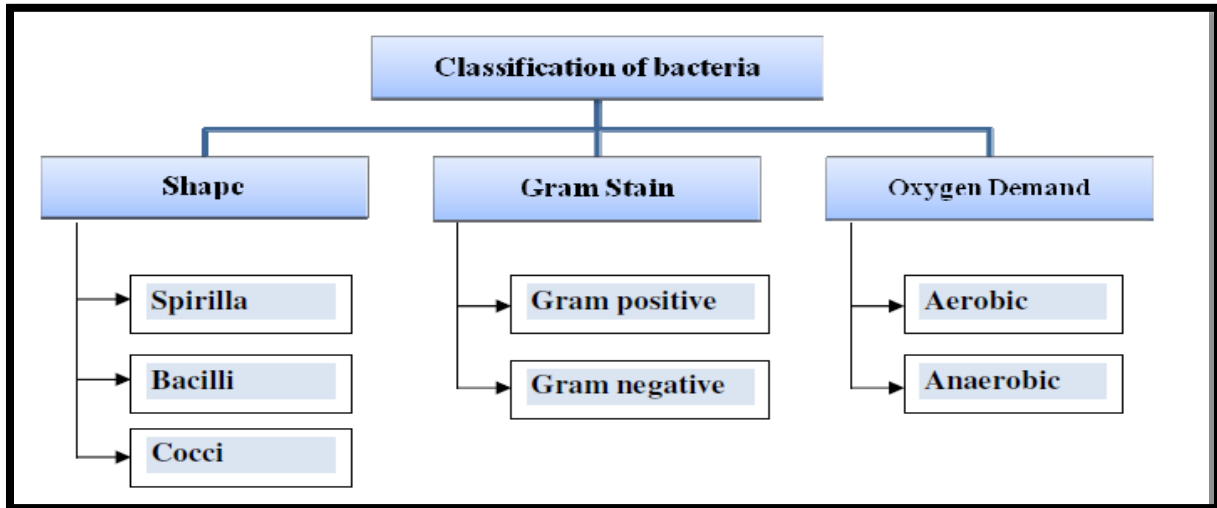


Figure.1: Categorization of bacteria kinds.

5.1 Anaerobic Bacteria

If anaerobic bacteria like closely related specie of shewanella are added to concrete, the compressive strength increases from 25-30%.

Table 1: Bio-chemical physiognomies of Bacillus Subtilis

Physiognomies	Bacillus Subtilis
Size, shape & grain stain	Long rods, 2-3µm in length & 0.60-0.80µm in width gram +ve.
Colour morphology	Irregular, dry, white opaque colonies
Sucrose	Acid & gas.
H ₂ S & indole creation	Absent
Dextrose	No acid & gas
Fermentation lactose	No acid & gas
Methyl red & Vogesproskauer tests	Absent
Nitrate lessen & Citrate usage	
Lipid & starch hydrolysis	Absent
Catalase & Gelatin deeds	Present
	Present

Table 2: Diverse type of curing based on their mechanism and composition

Usage	Kinds of bacteria
For surface curing	B. megaterium, B. pasteurii, Halomonasrurihalina and DeleyaHalophila
For crack curing	B. sphaericus
B. sphaericus	Thiobacillus, Baccillusubitilis and B. sphaericus
<i>Biodeposition curing</i>	<i>Composition of orthodox technique nutrient remedy</i>
<ul style="list-style-type: none"> • Bacillus sphaericus • Ureolytic blend cultures 	Urea NBP calcium chloride; Urea NBP; Urea calcium acetate and Urea NBP calcium acetate.
	Urea NBP calcium chloride; Urea NBP; Urea calcium acetate and Urea NBP calcium acetate.



Figure 2: a) Bacillus subtilis, and b) Bacillus Sphaericus.

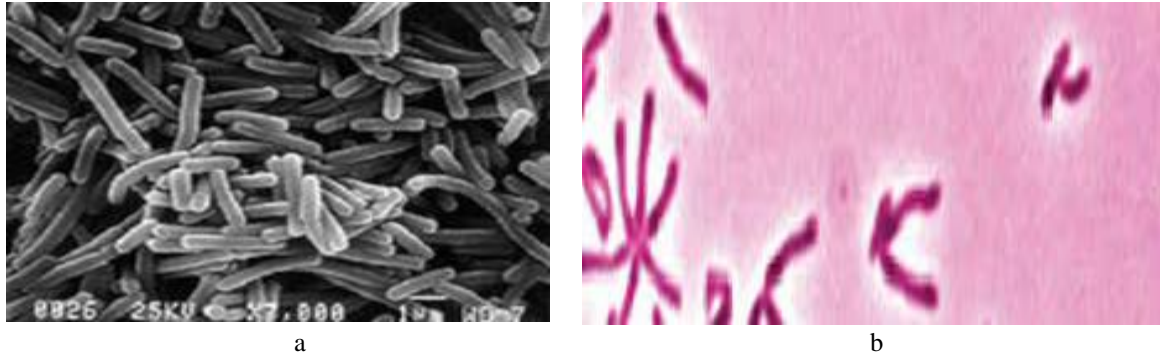


Figure 3: a) *Bacillus pasteurii*, and b) *Bacillus Pseudofirmus*.

5.2 Aerobic Bacteria

The various types of aerobic bacteria that can be used in concrete are, *Bacillus pasteurii*; *Bacillus sphaericus*; *Escherichia coli*; *Bacillus subtilis* (Table 1); *Bacillus cohnii*; *Bacillus pseudofirmus*; *Bacillus halodurans* and *Bacillus massiliensis*.

Bacillus Sphaericus is strictly aerobic gram positive rod shaped bacterium. It is an insecticide against certain strains of diseased mosquitoes. *Bacillus Sphaericus* are pore forming bacterium, dormant for several years and would be able to withstand extreme temperature.

6. Bacteria concrete

They can be produce by embedding bacteria in the concrete that are capable of constantly precipitate calcite.

6.1 Viability of bacteria in concrete

Growth of bacteria in concrete is a most questionable factor because of concrete s high alkalinity which is a restricting aspect for the survival of the bacteria. Only specific alkaliphilic bacteria can survive in such hostile environment of concrete. Therefore, it is necessary to immobilize the bacterial cells and to protect them from the high pH in concrete [9]. Polyurethane (PU) has been widely for immobilization of nutrients and bacterial cells even silica gel was used to protect the bacteria against the high pH in concrete. For effective crack healing, both bacteria and nutrients incorporated into concrete should not disturb the integrity of cement sand matrix and also should not negatively affect other important fresh and hardened properties of concrete. Only spore forming gram positive strain bacteria can survive in high pH environment of concrete sustaining various stresses [11, 17].

6.2 Bacteria Remediate Cracks Process

When the concrete is mixed with bacteria (*Bacillus subtilis*), the bacteria go into a dormant state, a lot like seeds. All the bacteria need is exposure to the air to activate their functions. Any cracks that should occur provide the necessary exposure. When the cracks form, bacteria very close proximity to the crack, starts precipitating calcite crystals. When a concrete structure is damaged and water starts to seep through the cracks that appear in the concrete, the spores of the bacteria germinate on contact with the water and nutrients. Having been activated, the bacteria start to feed on the calcium lactate nutrient. Such spores have extremely thick cell walls that enable them to remain intact for up to 200 years while waiting for a better environment to germinate. As the bacteria feeds oxygen is consumed and the soluble calcium lactate is converted to insoluble limestone [2, 8]. The limestone solidifies on the cracked surface, thereby sealing it up. Oxygen is an essential element in the process of corrosion of steel and when the bacterial activity has consumed it all it increases the durability of steel reinforced concrete constructions. Tests all show that bacteria embedded concrete has lower water and chloride permeability and higher strength regain than the surface application of bacteria. The last, but certainly not least, key component of the self-healing concrete formula is the bacteria themselves. The most promising bacteria to use for self-healing purposes are alkaliphilic (alkali resistant) spore-forming bacteria. The bacteria, from the genus *Bacillus*, *subtilis* it is of great concern to the

construction industry whether or not these bacteria are “smart” enough to know when their task is complete because of safety concerns. *Bacillus Subtilus* which is a soil is harmless to humans as it is non-pathogenic microorganism.

6.3 Chemistry of the Process

Microorganisms (cell surface charge is negative) draw cations including Ca^{2+} from the environment to deposit on the cell surface. The following equations summarize the role of bacterial cell as a nucleation site.



The bacteria can thus act as a nucleation site which facilitates in the precipitation of calcite which can eventually plug the pores and cracks in the concrete. This microbiologically induced calcium carbonate precipitation (MICCP) comprises of a series of complex biochemical reactions. As part of metabolism, *B.Subtilus* produces urease, which catalyzes urea to produce CO_2 and ammonia, resulting in an increase of pH in the surroundings where ions Ca^{2+} and CO_3^{2-} precipitate as CaCO_3 [10]. These create calcium carbonate crystals that further expand and grow as the bacteria devour the calcium lactate food. The crystals expand until the entire gap is filled. In any place where standard concrete is currently being used, there is potential for the use of bacterial self-healing concrete instead. The advantage of having self-healing properties is that the perpetual and expected cracking that occurs in every concrete structure due to its brittle nature can be controlled, reduced, and repaired without a human work crew. Bacterial self-healing concrete also prevents the exposure of the internal reinforcements. This form of self-healing concrete was created to continuously heal any damage done on or in the concrete structure. It was made to extend the life span of a concrete structure of any size, shape, or project and to add extra protection to the steel reinforcements from the elements. With this process, money can be saved, structures will last far longer, and the concrete industry as a whole will be turning out a far more sustainable product, effectively reducing its CO_2 contribution

6.4 Ability of the Bacterial Concrete to Repair the Cracks

Attention will be given on closure of cracks (blocking the path for ingress of water and ions) and how it regains mechanical properties (Figure 4). Cracks in concrete samples subjected to diverse loading situations will be scrutinized before and after the curing. Also the micro-organisms such as fungi, bacteria, yeasts, cyano bacteria, mosses, algae, lichens etc. are accountable to metabolism operation that produce microbial displacement of a protective CaCO_3 layer. Additionally, this procedure results in re-establishment of the bond in pieces of mineral building materials and fortify against further stone material decay. To demonstrate the positive outcomes of microbial CaCO_3 precipitation, concrete porosity expansion leads to increase in capillary water uptake, enlargement in gas permeability along with freeze-thaw deteriorate, outrageous carbonation rate and high chloride migration. Cracks can be healed by using calcium carbonate precipitating micro-organisms. These organisms are embedded in the concrete matrix after immobilization on diatomaceous earth, and will start the precipitation of CaCO_3 once a crack occurs. Through this process the cube’s crack will be coated with a layer of calcium carbonate, but in the meantime the crack faces bond together. This test is carried out by the initiation of small cracks on the cube and exposing them to the atmosphere for a period of 2 – 3 weeks.



Figure 4: Cube with initiated crack b) Healing of crack due to *B. Subtilis* by M.I.C.P

6.5: Benefits and shortcomings of self-curing concrete

Some of the merits and difficulties arising from self-healing concrete are displayed in Table 3.

Table 3: Disadvantages and advantages of Self- Healing concrete with examples

Advantages	Structure Service life expanded as inner iron as well steel structure protected.	Material manufacture pollution and energy decrease especially CO ₂ from iron and steel manufacturer
Disadvantages	Bacteria Lifetime might be reduce than concrete own	Compressive strength reduces as 20% of volume were filled with clay pellets of healing agent
Examples	Crack concrete Tensile strength of increase	Overall strength increase at least 18.35%

7. Concept of bacillus subtilis as best curing bacteria

Bacillus Subtilis is a Gram – Positive bacterium rod - shaped and catalase positive .it was originally named vibrio subtilis by Christian Gottfried Ehrenberg and renamed bacillus subtilis by Ferdinand Colm in 1872. Bacillus subtilis are typically rod shaped and are about 4 to 10 micrometer long and 0.25 to 1 micrometer in diameter with a cell volume of about 4.6 FL at stationary phase. As with other members of the genus bacillus, it can form an endospore to survive extreme environmental condition of temperature and desiccation. Bacillus subtilis is a pervasive bacterium commonly retrieved from water, soil, air, and putrefy plant residue. The bacterium develops an endospore that permits it to endure maximal conditions of heat and environment desiccation. Bacillus subtilis produces assortment of proteases and disparate enzymes that accredit it to demean different natural substrates and subscribe to nutrient cycling. Nevertheless, under most state the organism is not biologically energetic but exists in the spore form (Alexander, 1977). Bacillus subtilis is reviewed a benign organism as it does not have traits that cause disease, it does not examined pathogenic or toxigenic to plants, humans or animals also the probable danger associated with use of bacterium in fermentation application in facilities is low.

7.1 Reproduction of bacillus subtilis

Bacillus subtilis can divide symmetrically to make two daughter cells (Binary Fission), or symmetrically producing a single endospore that can remain viable for decades and is resistant to unfavorable environmental condition such as drought, salinity, extreme pH, radiation. It is found in soil, water, air decomposing plant matter.

7.2 Uses of bacillus subtilis

Bacillus Subtilis is widely used laboratory studies, but more for genetic research as oppose to health research. It is utilized in the creation of various antibiotics such as Diffcidin, Oxydiffcidin, Bacilli and Bacitracin which is helpful in treating bacterial skin infections and preventing infection in minor cuts and burns. Also, as an important source of industrial enzymes and polymers, as well as a Probiotic. Strain CF-3 is aerobic, motile and can grow under a concentration of NaCl from 2% to 7%. It utilizes citrate, starch and liquefied gelatin, and produces acid from glucose. It is tested positive for Voges–Proskauer reaction, methyl red and contact reaction (Table 4). The results showed that strain CF-3 had the same properties with *Bacillus subtilis* and made a foundation for the identification by searching for sequence homology among published reference sequences with the BLAST tool.

Table 4: Physiological and biochemical physiognomies of bacillus subtilis

Parameters	Bacillus subtilis
2, 5 and 7% NaCl, amyolysis, contact reaction & V-P test	Growing
10% & Facultative anaerobic	Not growing
Glucose oxidation, and citrate utilization	Growing

8. Making of self-healing concrete

The bacteria, *Bacillus subtilis*, was selected since it has capability to lie inactive for hundreds of years without absorbing nutrients and yet can survive in highly alkaline environments of pH 13. For the nutrient, Calcium lactate was selected because it is originated from carbon that supplies bacteria with biomass. Its ingestion produces an alkaline environment, favoring the activities of calcium precipitation, without hindrance to concrete setting [5], [17] – [19]. *Bacillus subtilis* and the Calcium lactate are curbed in a capsule made of ethyl Cellulose, the capsule is mixed into cement in its adjustable form and turn brittle when concrete hardens. When cracks appear in the cement, the capsule cracks and then the bacteria are vulnerable to water and air, produce germination. Through oxygen absorption, bacillus transforms soluble calcium lactate to insoluble limestone, which occupies the cracks (Figure 5).

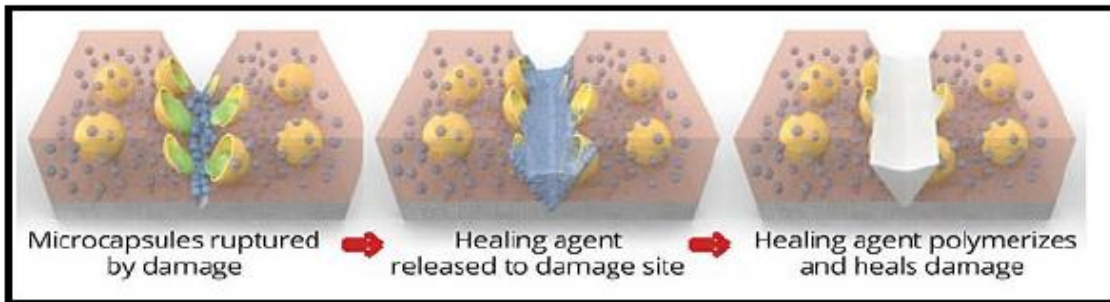


Figure 5: Making of Self – healing concrete

Conclusion

This study has justified need for using bacteria such as bacteria subtilis as crack-curing agent. Surface opening cracks that are common kind of defects in concrete structures though permission of water penetration or other harmful agents. This later resulted in durability loss earlier than expected, so cracks formed renovating and defects becomes indispensable and unavoidable. To cater for this unpleasant occurrence bacteria as self-curing agent is needed. Bacteria are relatively simple, single celled organisms, occur naturally in nature in various forms and also present not only on the surface but also beneath. *Bacillus Subtilis* a Gram – Positive bacterium rod - shaped and catalase positive that was originally named vibrio subtilis can divide symmetrically to make two daughter cells (Binary Fission) that can remain viable for decades.

The bacteria, *Bacillus subtilis* has capability to lie inactive for hundreds of years without absorbing nutrients and yet can survive in highly alkaline environments of pH 13. While, calcium lactate also supplies bacteria with biomass that utilizes citrate, starch and liquefied gelatin, and produces acid from glucose.

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