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APPLICATION OF BACTERIA IN CEMENT CONCRETE: AN OVERVIEW

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This paper reviews distinct bacteria used in Abstractcement concrete and how they can be used as healing agents, this paper has provides a brief overview of the concrete properties varying with adding bacteria, in concrete, nano-cracks are naturally present this leads to concrete deterioration due to the entry of deleterious substances into concrete, consequence as a failure, this concrete needs to be rehabilitated, Self-healing methods are adopted to overcome these circumstances, Calcite precipitation in concrete outcomes from the incorporation of bacteria along with their nutrient as a calcium source. Bio-mineralization methods deliver beneficial outcomes in concrete sealing of micro cracks, it is possible to seal the newly formed micro-cracks in concrete by continuous hydration mechanism. The various ureolytic bacteria include Bacillus Pasteurii, and nonureolytic bacteria include bacillus cohnii. Bacillus Substilis which can produce calcite join with urea and calcium source to seal newly formed microcracks for the enhancement of the concrete pore structure, for better outcomes bacterial doses were optimized. The paper demonstrates that the technique of encapsulation will lead to better results than the technique of direct application and also demonstrates that the use of bacteria can enhance concrete's strength and performance

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1. INTRODUCTION

Concrete is a brittle composite cementitious material that easily fractures under tensile loading. For this reason, reinforcement is installed to carry the tensile cross-sectional forces after cracking. From this point of view, reinforced concrete is always designed to allow the occurrence of cracks. Cracks such as not regarded as a failure of reinforced concrete as long as the prevailing crack width criterion is not exceeded. However, they provide preferential accesses for aggressive agents such as chlorides, sulfates, and carbonate. These aggressive agents can not only induce corrosion of reinforcement steel but degrade the concrete. Thus service life of a reinforced concrete structure is shortened besides, cracks cause leakage in a concrete structure such as water reservoirs, roof, and water pipes and negatively affects their functionality. For solving this problem as a novel idea, selfhealing of cracks has attracted much attention worldwide in Recent years, and some reviews on self-healing have been published mainly focusing on healing agents and methods to evaluate the efficiency of self-healing. [12]

Cracking can be repaired using materials such as silicates or mortar however these repair techniques are time-Consuming costly therefore the focus has recently shifted to the use of smart materials for damage prevention and decay minimization of the concrete structure an interesting alternative to the repair and rehabilitation of deteriorated concrete elements and structures is the use of self-healing concept. An innovative and promising approach is to implement an automatic repair i.e show called self-healing approach. To date many self-healing approaches including Autogenous, adhesive-based, mineral admixture based and bacteria-based methods have been developed among these technologies microbial induced self-healing of concrete cracks has become a popular area of research recently this technologies principally based on the application of minerals (CaCO₃) producing bacteria, which is a common and environmental friendly microbial community in the natural environment first introduced ureolytic bacteria as healing agents for concrete cracks which promote the enzymatic hydrolysis of urea to CO₂ and ammonia some researches also applied ureolytic bacteria eg. Sparcina pasteurii, Bacillus sphericus to the durability improvement and surface remediation of concrete or limestone. Healing of concrete crack was clearly improved by this method nevertheless, ureolytic bacteria were mainly externally applied on the surface of concrete structures with cracks this process cannot be recognized as authentic self-healing. Moreover, two ammonium ions concurrently generated for each carbonate ions during the formation of CaCO₃ which increase the nitrogen loading and have negative effects on the environment and human health. Recently the feasibility to use nonureolytic bacteria such as Bacillus Cohnii, bacillus substilis as a healing agent was proposed by Joners et.al. the bacteria incorporated in concrete matrix proliferate after activation by ingress water and oxygen via cracks and metabolize organic compounds (calcium lactate) instead of urea as the electron donor to produce CaCO₃. To protect the incorporated bacteria from crushing during mixing and form the high Alkanity, immobilization and encapsulation are prerequisites to maintain high efficiency and mineral forming capacity over periods of time. [21]

Bacteria concrete can repair itself due to calcite formation makes a concrete denser. So permeability and porosity of concrete reduced and property of concrete enhanced further

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research continuous must optimized doses of bacteria and nutrition.

compressive strength with the addition of different bacteria with variant quantity are given below in table 1.1

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Addition of bacteria not only repair the cracks it improves the overall behaviour of concrete and enhances the

Table 1.1 Mechanical Properties of concrete using different Bacteria [24]

S.NO.	Bacteria Used	Best Results	Bacterial concentration
1	Bacillus sp. CT- 5	Compressive strength 40% more than control concrete	5×10 ⁷ cells/mm ³
2	Bacillus megaterium	The maximum rate of strength development was 24% achieved in highest grade 50Mpa	30×10 ⁵ CFU/ml
3	Bacillus substilis	12% increment in C.S while using lightweight aggregate	2.8×10 ⁸ cells/ml
4	Bacillus aerius	11.8% increment in C.S as compare to control concrete	10 ⁵ cells/ml
5	Sporosarcina pasteurii	35% increment in C.S as compare to control	10 ⁵ cells/ml
6	AKKR5	10% increment in C.S as compare to control concrete	10 ⁵ cells/ml
7	Shewanella species	25% increment in C.S as compare to control mortar	100000 cells/ml

Note CS = compressive strength

1.1 SELF HEALING BASED ON BACTERIA

The idea of bacteria based self- healing is to utilize bacteria to promote precipitation of CaCO₃ in cracks in , suggested to use bacteria to induced the precipitation of CaCO3 to repair cracks the precipitation of CaCO₃ can be caused by various metabolic pathways such as the hydrolysis of urea and the oxidation of organic acid compare to other pathways for generating carbonate by hydrolysis of urea has several advantage for instance it can easily be controlled and it has the partial to produce high amount of carbonate within short time catalyzed by means of urease, urea is degraded to carbonate and ammonium and consequently increases PH value, as well as on carbonate concentration in bacterial environment. One mole of urea is hydrolyzed intracellularly to one mole of ammonia and one mole of carbonate which spontaneously hydrolyze to one mole of ammonia and carbonic acid these product subsequent reach the equilibrium in water to form bicarbonate and two mole of ammonium and hydroxide ions

$$CO (NH_2)_2 + H_2O \rightarrow NH_2COOH + NH_3$$
 (1)

$$NH_2COOH + H_2O \rightarrow NH_3 + H_2CO_3$$
 (2)

$$H_2CO_3 \to HCO_3^- + H^+$$
 (3)

$$2NH_3 + 2H_2O \rightarrow 2NH_4^+ + 2OH^-$$
 (4)

$$HCO_{3}^{-} + H^{+} + 2NH^{+}_{4} + 2OH^{-} \rightarrow CO_{3}^{2-} + 2NH^{+}_{4} + 2H_{2}O$$
 (5)

$$Ca^{2+} + cell \rightarrow cell - Ca^{2+}$$
 (6)

Cell
$$-Ca^{2+} + CO_3^{2-} \rightarrow cell - CaCO_3$$
 (7)

In the presence of calcium ions, calcium carbonate is precipitated once a certain supersaturation level is reached. Because of the negative charge of the cell wall, calcium ions are attracted. As a result, the crystals precipitated on the bacterial cell besides precipitation cells take place in the bulk liquid phase.

(non-urea olytic bacteria) these following reactions are given below.

$$CaC_6H_{10}O_6 + 6O_2 \rightarrow CaCO_3 + 5CO_2 + 5H_2O$$
 (8)

$$CO_2 + Ca (OH)_2 \rightarrow CaCO_3 + H_2O$$
 (9)

Another metabolic pathway to produce $CaCO_3$ is the oxidation of organic acids compared to the hydrolysis of urea, which produces excessive ammonium, the oxidation of organic acids has less environmental impact. Moreover, during the precipitation calcium carbonate through this metabolic way, CO_2 is produced as well. The produced CO_2 can also react with portlandite $(Ca(OH)_2)$, which is quantitatively an important hydration product of portland cement, to form more calcium carbonate.[12]



Volume: 07 Issue: 10 | Oct 2020 www.irjet.net p-ISSN: 2395-0072

However, to use bacteria to heal cracks in concrete, some technical problems have to be solved the bacteria should protect not only against the alkaline environment in concrete but also against the decreasing space in the matrix when hydration of cement proceeds when the bacteria spores, which can be viable up to 50 years, was directly added in the concrete, their lifetime dramatically decreased to only a few months, this caused by the hydration of cement grains as the cement grains. As the cement grains hydrate, most pores become smaller than bacterium spores with a size of 1 μm . which causes the cell to collapse.

Immobilization of bacteria in porous clay, aggregate before the mixing of concrete can prolong the lifetime of bacteria enormously self-healing can trigger later when cracks intersect theses clay particles. Contemporarily researches According to existing research two method are available to applying healing agent in concrete first one direct application of bacteria and second encapsulation method, in case of direct application of bacteria use bacteria as spore or liquid culture medium with optimum concentration aid in cement/concrete this bacterial concentration enhance the property of concrete and provide the healing capacity, but in this method some limitation healing capacity decrease day by day due to decrease the pore size, survival capacity of bacteria reduce, in concrete many stresses came on bacteria and concrete structure dense during aging so that we need a encapsulation method, in this method we provide a protection to bacteria, for this method we immobilized bacteria in capsule-like hydrogel, expanded perlite particle encapsulated bacterial spore that method enhance the property of concrete and healing efficiency after cracking rupture capsule release healing agent and heal the crack due to some chemical reaction. Maximum crack width 0.799 mm heal in the encapsulation method. This method increases the durability of concrete and makes a dense structure reduce porosity and healing capacity to reduce a maintenance cost to avoid the corrosion causes. [24]

The addition of bacteria and their nutrition in the concrete matrix. after some time crack appears and bacteria get oxygen and water in the cracks so bacteria activated it produces calcite that helpful to seal the cracks and then goes into the dormant state carry the work long time bacteria also reacts with calcium lactate and produce calcite and improve strength. Calcium lactate as nutrition of bacteria, If the crack depth was more it difficult to repair because proper oxygen and nutrition not received to bacteria, indirect application of bacteria maximum depth 27.2mm was healed [24]

2. RESEARCH FINDINGS

Recently many research going on bacteria-based self-healing various papers demonstrate the addition of bacteria improves the cement concrete property some research paper summaries below.

Santosh K. Ramchandran et.al. (2001) studied Bacteria Bacillus Pasteurii and Pseudomonas Aeruginous, Glass distilled water, and buffer used for media preparation. B.Pasteuri was suspended in two solutions (saline and phosphate buffer) to prepare samples with various concentrations and to cure them with urea and calcium chloride solution. Results show that suspended in saline have a low strength as compared to phosphate buffer. The combination of Bacteria increases the compressive strength of 21.27% and 18% at 7 and 28 days respectively with concentration 3.8×10⁷ cell /cm³. Live bacteria another set of killed bacteria at the same concentration and compressive strength increase 27% and 10% at 7 and 28 days. With a single B. with killed cell, compressive strength increases by 40% and 5.4% at 7 and 28 days. Crack filled with 5.2×107cell/cm3 has been found to induce the maximum compressive strength. Using stiffness test made crack and filled with a mixture of B. Pasteurii and sand cured with urea and calcium chloride solution with cell concentration 3.8×109cell/cm3, Crack filled with bacteria, and sand higher strength value found when compared to those treated only sand. Crack depth 25.40mm repair B.pasteurii has average strength increased by 61% of control. Crack filled with concentration 5.2×10⁷cell /cm³ have been found to induce maximum compressive strength, SEM XRD test results show that calcite precipitation in the sample that enhance mortar properties and durability and provide healing power. [1]

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Willem Demuynck et.al. (2006) Investigated bacterial treatment on the surface of mortar/concrete is better than conventional treatment, use sparcina pasteurii as a form of pure culture and ureolytic mixed culture from sewage sludge biomass, prepared cube (w/c 0.5) immersed in a 1-day old stock culture of S. pasteurii for 24 hours after that should be immersed in nutrient solution and prepare another cube specimen with mixed ureolytic sludge (0.5-1 mm thick) and immersed in a nutrient solution. The author did sorptivity, gas permeability, oxygen flow rate tests that determine contact angle and colour measurement. SEM, XRD test analysis revealed Caco₃ precipitation, reduction of permeability due to bacterial treatment in sample occurred to same as treatment with penetration sealant, contact angle values with different treatment decreasing order silanes/siloxanes>polyurethane coating >acrylates >bacterial treatment >untreated, treated with B. sphericus, urea, calcium acetate, nutrient broth resulted increasing the contact angle compared to untreated samples. Pure culture treated samples have low water absorption. All the test results revealed bacterial treatment gives a promising result than others. [2]

Henk M. Jonkers et.al. (2008) Investigated the ability to heal micro-crack using bio-mineralization, Used bacteria bacillus cohnii, Bacillus halodurans, and bacillus pseudofirmus (6×10^8 cm⁻³) a form of spore, and make a cement stone 4cm cube with incorporated bacteria, determine their influence on compressive and tensile strength with and without bacteria, test their compressive strength after 3,7, 28days curing,



Volume: 07 Issue: 10 | Oct 2020 www.irjet.net

splitting tensile test were perform on cement stone cylinder added organic amount 0.5% of the weight of cement use OPC with 0.4 and 0.5 water-cement ratios and add calcium lactate (0.5% of the weight of cement) B.cohnii (1×10^8) spore/cm³ as an organic compound to the paste mixture can result in unwanted strength loss incorporation of a high number of bacterial spore in paste $(6\times10^8\text{cm}^{-3})$ resulted in about 10% decrease in compressive strength, XRD SEM tests results show that calcite precipitation in a sample, 10% loss in compressive strength when added B. pseudofirmus. Extend mineral production decrease the permeability, the bacterial based two-component system seems promising more sustainable high strength. [3]

Henk M.Jonker et.al. (2008) investigated when using two species bacillus pseudofirmus and B.cohniI in the cement stone both bacteria used as a spore at vegetative cell pellets. This study describes the process of making spore formation and vegetative cells, use manganese as a catalyst for the formation of spore. The author prepares a 4cm cube of cement with water-cement ratio 0.4 and 0.5 and replaces part of the water to the spore and determine the viability of the spore in the cement. The most probable number technique is used to find out the viability of spore, for determining pore size distribution in cement cube, determined by mercury intrusion Porosimetry (MIP). For compressive strength, cube(0.4w/c) prepared with incorporated bacteria B. Pseudofirmus (6×108) spore/cm3, an organic compound added to the cement is 1% of weight cement and also prepared cement stone cube with incorporated B. Cohnii (1×108 spore/cm³) and Calcium lactate (0.5% weight of cement) produce a crack and cured with water. SEM test results analyzed and visualize the spore size 0.8 to $1~\mu m$ experiment shows several viable cell decrease with increase the specimen age. Due to the precipitation of calcium carbonate that reduces the permeability of the cube, bacterial spore added to cement remains viable till 4 months due to continuous decreases pore size diameter as per cement stone setting with time. [4]

Severine Anne et.al. (2009) studied applying a biogenic treatment on the old building plaster (10µm) used bacteria (B.Cereus), suitable for limestone. Prepared a culture and nutrient media and spread on the plaster surface $(1 l/m^2)$. After 7 days biomineralization optimum and observed SEM, GIXD, XRD test results show that calcite, Halite, Sylite crystal on the treated sample, prepare different sample like untreated (S1), treated sample (S2t), treated two times (S2tt), treated at the center (S3t), treated at boarder (S3tb), compare their SEM test results and found more denseness in a treated sample and, treatment reduced the water absorption. S2tt have found more calcite coating, one of the demerits arise in method salt deposition in one case Vaterite was found (aging 53days) but was not found again, in other sample, GIXD spectra obtain a higher concentration of halite and Sylvite on the edges than the center. So we said biogenic treatment enhance durability and reduce water absorption. [5]

Jianyun Wang et.al. (2011) studied the use of silica gel and polyurethane as a carrier to bacteria bacteria immobilized in silica gel and polyurethane comparing both with a different parameter like permeability, compressive strength healing efficiency, experimental results show that immobilize Bacteria in silica gel (25%by mass) and polyurethane (11%by mass) due to caco₃ precipitation, crack mortar specimen healed. when immobilizing bacteria in polyurethane had higher strength regain (60%) and low permeability (10^{-10} to 10^{-11} m/sec.) compare with specimen healed by immobilizing bacteria in silica gel have strength regain only 5% and permeability 10^{-7} to 10^{-9} that results indicate that polyurethane has more potential as bacteria carrier for self-healing of concrete. [6]

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Amirreza et.al. (2013) investigated Proteus mirabilis and Proteus Vulgaris both ureolytic bacteria used in harden and fresh concrete, prepare mixed culture (MC) in liquid media first apply culture on hardening concrete, and also add in fresh concrete. prepared concrete specimen with bacteria and without bacteria tested their strength, density at 1,7,14, 21, 28 days, broken concrete specimen treated with bacteria for 30 days then tested again their strength density, ultrasonic pulse velocity test (UPV) when treated with MC forms precipitation of caco₃ on cracks, macro crack partially and micro-cracks fully healed and deeper crack evaluated by UPV, hence 85% improvement is observed, those cracks treated with microorganism 10% improvement in compressive strength, it was found that MC treatment suitable in broken concrete but fresh concrete does not give the promising results due to high PH. [7]

Varenyam Achal et.al.(2013) studied used a Bacillus sp. Bacteria to enhance the mortar compressive strength and durability, bacillus sp. lead to more than 50% reduction in permeability and porosity, 27.2mm depth of artificial crack healed by using bacteria and sand mixture, Bacteria bind sand particles due to precipitation of caco₃, when aid bacteria at 5×10⁷ cells/ml concentration give the optimum result in compressive strength, RCPT(rapid chloride permeability test), porosity test, this bacteria culture on nutrient agar at PH8, prepare the mortar cube with water/cement ratio 0.47 and incorporated the bacterial(aq). While comparing RCPT test results value opted for the bacterial specimen has very low (975.33) and the control specimen has moderate (3177) so bacteria-based mortar gives a promising result. XRD, SEM test results shows that caco₃ precipitation on the bacterial specimen and Bacillus sp. Lead to a more than 50% reduction in the porosity in a mortar and increase the compressive strength as 40% of control, bacteria enhance the mortar strength and durability. [8]

J.Y. Wang et.al. (2014) Investigated that bacteria spore should be encapsulated in a hydrogel capsule because this hydrogel capsule has a water retention capacity that helps to bacteria for healing and calcium carbonate precipitation in the crack. hydrogel like a polymer chain for making a hydrogel using UV rays and high temperature then bacteria and there nutrition like urea and calcium incorporated in



Volume: 07 Issue: 10 | Oct 2020 www.irjet.net p-ISSN: 2395-0072

hydrogel and check various properties like water absorption capacity, permeability, and viability of bacteria. Prepared mortar specimen, B.spharicus spore was successfully encapsulated in hydrogel so the viability of bacteria, not decreases. When the specimen cured fully submersed in water condition crack healing efficiency was better. Submersion of water not required that specimen crack width (0to0.5mm) healed successfully. Due to mechanical stress and denseness of concrete, bacterial spores are compressed and the functionality of bacteria is reduced so we used the encapsulation method for better outcomes. The average water permeability was decreased. This method gives promising results and hydrogel as a good carrier of bacteria.

Y. Zhang et.al. (2014) investigated microbial mortar treated with three different calcium source (Cacl₂, Ca(Ch₃Coo)₂, Ca(No₃)₂), when treated with Ca(Ch₃coo)₂ were found twice strength than treated with Cacl2, Ca(No3)2 as a calcium source. The result of the mercury intrusion porosimetry test shows that pore size distribution is more uniform when samples treated with Ca(ch₃coo)₂ as calcium sources, SEM, XRD test results revealed calcium carbonate cemented in microbial mortar. Calcium Acetate (Ca(Ch₃coo)₂) is a more suitable calcium source than other sources, microbial mortar consists of Bacteria Sporosarcina Pasteurii, sand, nutrient solution (calcium source and urea with equal molar concentration). Prepare a microbial mortar specimen and water absorption, UCS, BTS test followed. Treated with Ca(Ch₃oo)₂ was the larger dry density and low water absorption ratio, due to the calcite distribution of the microbial mortar sample was more uniform, a compressive strength of microbial mortar was enhance treated with calcium acetate, average BTS (Brazilian splitting tensile strength) of the acetate sample was higher than other two samples it was 2.4 and 3.0 times that of chloride and nitrate samples respectively, Most suitable source for microbial treatment is calcium acetate. [10]

Rafat Siddique et.al. (2015) investigated the influence of cement baghouse filter dust (CBFD), prepared a control and bacterial concrete and partial replacement of ordinary Portland cement (0%, 10%, 20%, 30%), determine their compressive strength, water absorption, porosity, chloride permeability, and sorptivity after 28 and 56 days of curing. Isolate ureolytic bacteria (10⁵ cells/ml) from marble sludge, the prepared concrete cube with proportion (1:1.45:2.98) with 0.5 W/c ratio, incorporated bacteria cell in the concrete matrix, prepare cube for compressive strength, and SEM XRD test followed, after 28 and 56 days compressive strength consist of CBFD (0%, 10%, 20%, 30%) and bacterial concrete (0% CBFD) have 10% increment in strength, Maximum reduction in porosity after 28 days observed in bacterial concrete was 2.80% an, Porosity of normal and bacterial concrete increases as increase of CBFD in the matrix. After 28 days sample with (30%, 20%, 10%) CBFD was observed to have chloride permeability was moderate range similar trend at 56 days. Sorptivity of control concrete was 1×10⁻⁷mm/s at

28 days while a concrete mixture containing 10%,20%,30% CBFD showed an increasing trend, at 28 days concrete mixture containing 10% CBFD compressive strength equal to the target strength of 28N/mm² required for designing. [11]

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Haoliang Huang et.al. (2015) studied almost all types of selfhealing methods and mechanisms. (self-healing in cementitious material, autogenous self-healing, self-healing based on mineral admixture, self-healing based on bacteria, self-healing based ion adhesive agent (epoxy are encapsulated pre-embedded in concrete) are reviewed. Literature shows that all mechanism of self-healing is effective depends upon some particular condition, we cannot say that a particular method of self-healing is the best, this depends on particular condition and situation. The author discussed additional cost for realizing self-healing in a concrete structure. While we talk about Autogenous self-healing due to further hydration of unhydrated cement, there is the main drawback in method was when mixing water to cement reaction started and mineral admixture reacted rapidly so further use not possible, in bacteria-based self-healing provide nutrient to bacteria is costly so further research going on using a sugarbased nutrient for bacteria, all the bacteria-based healing provide promising result but all have some limitation. [12]

Mianluo et.al. (2015) investigated bacteria that can heal the crack with a substrate in the concrete matrix. Bacteria heal 0.1 to 0.3mm crack width repair rate was 85% and 0.3 to 0.5 mm 50 to 60% in 20 days. The difficulty to repair a 0.8mm corresponding repairing rate was lower than 30%. prepare the bacterial concentration 10^9 cells /ml and cement specimen prepared by OPC with mixing proportion (30g substrate and 10ml bacterial liquid, 1-liter cement paste)made a cylinder and prism of cement make artificial crack 0.1 to 1 mm cured for 21days best result opted in wet curing. XRD and EDS, SEM test results confirm caco $_3$ crystal on the sample when a crack more than 60days crack healing ratio very small due to survivable death, fewer substrata, and cement paste porosity decrease. [13]

Ehsan Mostavi et.al. (2015) investigated the use sodium silicate as a healing agent encapsulated in double-walled polyurethane /urea-formaldehyde (PU/UF), double-walled microcapsule were incorporated into self-healing concrete beam micro-crack were created middle of the beam, and the healing process of concrete was monitored by the portable ultrasonic non-destructive digital indicating tester (PUNDIT). It was found that the healing rate in a concrete beam in consist of a 5% microcapsule(weight of cement) was higher than 2.5% microcapsule, optimum preparation procedure for capsule consist of PH 3.1 and agitation rate of 1000 rpm and curing temp 57° C. All test results revealed sodium silicate has better potential as a carrier of bacteria. [14]

V. Wiktor et.al. (2015) investigated the field performance of bacteria based repair system. Use parking garage floor there some cracks due to freeze and thaw attack prepare two type solution first one consist of (sodium silicate (alkaline buffer), sodium glunate (carbon source for bacteria) and alkaliphilic



Volume: 07 Issue: 10 | Oct 2020 www.irjet.net p-ISSN: 2395-0072

bacteria and Second solution consist (calcium nitrate, alkaliphilic bacteria), applied the solution to cracks location and treated. Compare treated and untreated cracks, find their porosity, permeability, and efficiency of repairing. determine resistance to freeze and thaw attack, the author finds promising results to treated cracks with bacteria, leakage is prevented, and lower mass loss $(1.9\pm0.3\text{kg/m}^2)$ untreated $(3.6\pm1.3\text{ kg/m}^2)$ that revealed bacteria have good resistance to freeze and thaw attack. Parking garage used a Portland slag cement slag content has lower carbonation compared OPC this lead also to aragonite formation, bacteria precipitate caco₃ that pour cracks and enhance the property of concrete. [15]

E. Tziviloglou et.al. (2016) Investigated bacteria-based selfhealing concrete increases liquid tightness in cracks, use spores derived from alkaliphilic bacteria and incorporated in the lightweight aggregate (LWA expanded clay particle), bacterial spores(108CFU/liters) with calcium lactate (200g/L), yeast extract (4g/L), Prepare OPC mortar specimen consist of (CEM1, water, sand (0.125/1mm), sand (1/4mm), LWA(1/4 mm) all kg/m³),three different proportion of mortar mix design as follow one REF (463, 231.5, 855, 825, 0 all kg/m³) another mix CTRL consist(463, 231.1, 855,0 ,257(LWA)all kg/m³) another mix B consist (463,231.1,855, 0, 280 (weight include impregnated healing agent)). Prepare prisms for determining compressive, flexural strength on 3,7,28 days. Damage is induced by 3 points bending test largest crack was 350µm now cracked sample stay in three different healing environments compare control and bacterial mortar specimen. crack water permeability test(REF 71% and 80%, mix B 69% and 91%, 31% and 82% for CTRL at 28 and 56 days respectively),oxygen consumption measurement test ,XRD,ESEM test. in compressive strength, as expected incorporated healing agent increase the air content delay the hardening of cement approximately one day, consequences the early age (3days) flexural and compressive strength of mix B specimen was 54% and 63% respectively lowered than control. Healing rate in wetdry condition higher than the submerged condition. [16]

Chandani Kumari et.al. (2016) studied that bacteria can enhance the property of mortar author use Bacillus Cohnii nonureolytic bacteria their optimum growth at PH10 (6.05×10⁷ CFU/ml)use Portland slag cement while aid the bacterial concentration initial and final setting time of cement increased marginally, soundness test results for control 2.0 mm and when added bacterial concentration 10⁷,10⁶,10⁵ CFU/ml have 2.0, 2.5, 3mm respectively, prepared mortar cube with 1:6 proportion to add the bacteria with different concentration like 107,106,105 CFU/ml that improve the compressive strength as 49.18%, 32.78%, 26.3% at 28 days respectively cured with a concentration of 0.2 Mol per liter calcium chloride in water, while Cured with normal tap water of bacterial concentration 10⁵ CFU/ml have 2.53% improve compressive strength. Cured with urea-cacl2 solution increased by 11.03%. For drying shrinkage test prepare prism, result was opted control 0.38% and 0.08%, 0.05%, 0.05% at bacterial cell concentration $10^5,\ 10^6,\ 10^7$ CFU/ml respectively. For performing XRD, the SEM test prepares a sample passes 100 μm IS sieve, results showed calcite precipitation in the sample. All tests revealed that bacteria enhance mortar property and durability. [17]

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R. Alghmghamri et.al. (2016) investigated the vacuum impregnation technique used for impregnating self-healing agents (polymer-based sodium-silicate) into a lightweight aggregate (4-8mm). After impregnation sodium-silicate coated with PVA. Prepared concrete specimen, use PUNDIT-PL200 to determine the crack depth, microstructure analysis of the healed specimen, and compare it to the control specimen. Sodium silicate led to improve strength and reduce water absorption nearly half, XRD SEM FT-IR results show that healing due to sodium-silicate produces more C-S-H gel to heal the crack. Flexural strength recovery and water absorption examined, LWA showed 80% recovery of precracking strength, three-point bending test to induced crack with 0.3 mm width after first-round cure the specimen check their properties. Using impregnated LWA showed a 50% reduction in the sorptivity test index compared with the control cracked specimen. These results show that the healing agent improves the strength and durability of concrete, and LWA has potential as a carrier to the healing agent. [18]

Mohamed Al Azhari et.al. (2017) studied expanded perlite as a carrier of bacteria, Use calcium acetate or calcium lactate as substrate, Replaced 20% fine aggregate to the expanded perlite consist nutrient and calcium acetate (precursor). The study shows a minimal number of spore and nutrient (yeast extract) and calcium acetate required for optimum selfhealing process in mortar cube. Use Bacillus Paseudofirmus Bacteria (spore) in a mortar, Embedded the bacteria precursor nutrient in the expanded perlite (EP) have water absorption 146% when the EP contain spore (8×109cell/ml) (CPS), EP contains nutrient (CPN) and prepare mortar specimen w/c 0.5, and was replaced by CPN only(M:100), M:90 to M:50 a different combination CPN and CPS value were added in ratio 9:1,4:1, 7:3, 3:2,1:1, Visualize crack after healing and before, no healing obtain in (M: 100, M:90, M:50, M:80), In M: 60 and M: 70 there is maximum ratio b/w them so the healing is maximum and lack healing M80 and M90 due to a low number of spore, Paper revealed the proportion of available spore to calcium acetate and yeast extract is an important factor to generate efficient self-healing in the moist and humid environment required, for faster healing wet and dry cyclic environment prefer. [19]

Mohammed AL-Anshari et.al. (2017) investigated what is an effect on the encapsulation method due to the modification of calcium nitrate. This modification is targeted to reduce the adverse effect on compressive and flexural strength of cement mortar/concrete. Various mortar specimen prepared with different proportions of microcapsule (0.50%, 0.75%, 1%, 1.5% by the weight of cement) determine their



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compressive, flexural strength, and elastic modulus. The results show that modification enhances the mortar mechanical properties, calcium nitrate used as a healing agent and encapsulated in microcapsule as a two-phase (aqueous and continuous) for preparation of self-healing micro capsule used these steps (composition, emulsification, and polymerization) SEM test result of microcapsule shows that optimum healing efficiency achieved incorporating microcapsule with average diameter 58.7µm. after 28 days the compressive and flexural strength decreases but does not $cross\,10\% and\,17\%\,respectively.\,The\,average\,strength\,of\,the$ microcapsule does not differ to control mixed strength, prepared the load-displacement curve using the LVDT machine. Microcapsule decreases mortar stiffness, ANOVA analysis shows that modified microcapsule does not significantly adverse effect in mixed strength. 0.50% micro capsule gives a promising result. According to the study order to achieve both acceptable strength and elastic modulus, it recommended using a 0.75% microcapsule. [20]

Jiayuang Zhang et.al. (2017) studied that B.cohni immobilized in expanded perlite (EP) clay particle and expanded clay (EC) particle and check their healing rate in concrete specimen B.Cohni immobilized in EP particle that heals the crack up to 0.79mm and EC particle 0.45mm, in concrete matrix bacterial spore concentration is 5.2×108cell/cm³ and calcium lactate 9.6 kg/m³, Compare these sample result to control and checked healing rate at 7, 14, 28 days respectively. XRD, FSEM test results shows the formation of calcite in a concrete specimen. EP, EC particle coated with a geopolymer. All the test results revealed that expanded perlite has a great potential as bacteria carrier higher healing rate in 28 days. [21]

Kunamimei Vijay et.al. (2018) investigated when increasing the percentage of calcium lactate in a concrete matrix (0.5, 1, 1.5, 2, 2.5 % of the weight of cement) and bacillus substilis spore powder (0.5%weight of cement 2million CFU/gram) was added to both spore powder and culture form. Cultured bacillus substilies with a concentration of 1×10^5 cell/ml were mixed with concrete prepare a cube were used for studies. A maximum 12% increase in compressive strength has opted with the addition of 0.5% calcium lactate in concrete. SEM, EDX test results examined, and show the formation of ettringite in pores and calcite make a concrete dense. Use the response surface method for optimizing experimental data. The addition of higher concentration calcium lactate increases precipitation of caco3 that decreases the compressive strength as slide percent. Use the electric resistivity method to determine cracked surface healed fully or partially and compare the crack and uncracked surface of their electrical resistivity graph. Optimum result in concrete matrix obtain 0.5 % calcium lactate (weight of cement) and 0.5% bacterial spore (weight of cement). Conclude that add calcium lactate should in lower for better compressive strength. [22]

Jing Xu et.al. (2018) studied the use a ceremsite material for immobilization of bacteria (sporsarcina pasteurii), bacteria

cultured and make a spore (10°cells/ml) for the alkali erosion treatment, Ceramsite particle was first treated by NaOH solution after the treatment solution was prepared at a water-cement ratio 10, incorporated bacteria into Ceremsite particle and check the viability of spore. Porous Ceremsite particle loaded with nutrient (group N), spore only (group S), both spore and nutrient (SN), spore together with or without peptone and beef extract. For incorporation of bacteria into Ceremsite particle, immersed 196g Ceremsite in a 150ml solution consists of spore and nutrient for two hours. Ceremsite without loading is the control group (group c), in mortar matrix (cement, sand, water, Ceremsite, beef extract, urea, calcium nitrate, basalt fiber, water-reducing admixture with varied quantity) prepared mortar cube found their compressive strength, regain ratio, water absorption, SEM test results show that group SN had a much better healing effect than the other group, regain ratio was increased 20% compared to control group, water absorption decreased 30%, due to caco₃ precipitation the percentage of crack healing was 10% (N or S group), crack up to 273µm can be healed and crack closer ratio was 86% in 28days in SN group. Heat treatment instead of number socking could increase the porosity of Ceremsite that increase the immobilization capacity, the optimal heating temperature was 750 degree Celsius, and the maximum crack width healed by 0.3mm, these experiment revealed Ceremsite particle have to potential to work as the carrier to bacteria in healing. [23]

3. CONCLUSIONS

This review paper revealed that ureolytic bacteria use urea to produce calcite as a healing agent like bacillus pasteurii and non-ureolytic bacteria use a calcium source to produce calcite like bacillus cohnii this paper has appraised various types of bacteria that can be used for healing purpose, bacteria have a beneficial effect on compressive strength and durability of mortar and concrete cube. The benefit of this concrete decrease water penetration and permeability (50%).

These following conclusion can be drawn are

- [1] MICP treatment improves the compressive strength, tensile strength, durability, and enhance the property of cement concrete.
- [2] For promising results should use calcium lactated (0.50%) and bacteria with low concentration.
- [3] When we increase the substrate in the concrete matrix give the negative results should be minimum.
- [4] The optimum CFU for bacteria is 10⁷ that gives promising results in strength and healing efficiency.
- [5] The encapsulation method best carriers of bacteria are hydrogel, Polyurathane, sodium silicates, Ceremsite, EP, LWA they give promising results in healing.
- [6] When cracks more than 60 days crack healing ratio is very less due to survival death, less substrate, and cement porosity.
- [7] Mixed culture treatment suitable in broken concrete but in fresh concrete not give the promising results

Volume: 07 Issue: 10 | Oct 2020 www.irjet.net p-ISSN: 2395-0072

This type of concrete environment friendly but the initial budget of this concrete greater than normal concrete this is the main drawback of this concrete.

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