

# Microbial Concrete and Influence of Microbes on Properties of Concrete

Pratyush

Guest Faculty, Kamla Nehru Institute of Technology Sultanpur, India

**Abstract:** *The concrete is very commonly used for the structures, roads, monuments and modern buildings despite weak in tension having negligible ductility and less resistance to cracking. As the concrete structures most of the times are exposed to variant weather / atmospheric conditions may develop surface deterioration, even invisible cracks may also develop on the surface with passage of time. The microbes in concretes may help mineralization to introduce calcium carbonate precipitation is called microbiologically induced calcite precipitation (MICP). This MICP is highly impermeable in nature and used to remediate the cracks in building materials. The precipitated calcite layer improves the concrete performance, do heal the surface and is corrosion resistance too besides being used as water purifier. In this paper the various species of microbes which affect the behaviour of mortar specimens and performance of concrete in terms of compressive / flexural strength, efficiency, self healing characteristics of specific bacteria when introduced with concrete, called microbial concrete is studied and reviewed. The literature review reveals that when Bacillus sp. CT-5 as microbe is used, the cement mortar compressive strength is increased by 36% [14, 22, 37, 44].*

**Keywords:** Microbial concrete, microbiologically induced calcite precipitation (MICP), properties of concrete

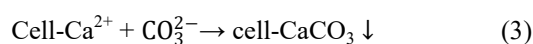
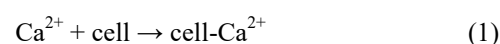
## 1. Introduction

Concrete is most used mansion materials. In spite of its remarkable properties it has drawbacks of cracking. There are distinctive factors causes crack in concrete. Weathering induces multiplied porosity to weaken the structure and the surface appearance is insipid. The exposure to intense weathering picks up porosity, diminishing the mechanical features of the concrete. The permeability of the concrete depends on the porosity. As the more open pores in concrete structures enhances permeability to involve more ingress / penetration of substances like brutal gases and liquids from the surrounding atmosphere, the concrete structures lead to deterioration. In recent days the deterioration on the surface of concrete due to aging and harsh exposure to variant weather and atmosphere is treated through microbial precipitation in concrete called microbial concrete. It is an amalgamation of biology and technology. The microbial concrete utilizes bacteria to enhance the compressive and flexural strength and durability of concrete. The outcome of microbial activities from microorganism is generally calcium carbonate – the precipitates. The microbial activities are natural and free from pollution. The calcium carbonate called calcite fills the pores and voids to make concrete denser. The literature survey says both naturally and in laboratory conditions the bacteria from natural habitats precipitates and the self healing agent remain in hibernated conditions inside the concrete for even up to two hundred years and starts activating the soluble nutrients when water seeps in through cracks to convert to insoluble calcium carbonate.

The calcium carbonate in the form of precipitation seals the cracked surface on the concrete as bone fractures in the human body healed naturally by osteoblast cells. The steel used in RCC or CC gets corroded when it comes in contact with oxygen available in air or water ingresses or seeps in through cracks is consumed during the metabolic biochemical reactions to form calcium carbonate helps in arresting corrosion of steel being eaten away to increase the

life of steel. The abundant bacteria species are diverse in nature and precipitate to form mineral carbonates in natural environments. The pH of surrounding is enhanced by carbon oxide and ammonia converted from urea that is hydrolyzed from bacteria and fungi; and urease a nickel dependent enzyme found in plants. The calcite precipitation is promoted by the bacteria primarily in alkaline pH conditions. It is found that the calcium ions bond to the cell walls when the negative charge or potential of the bacterial cell surface is resulted. The literature also reveals that calcium carbonate crystals' precipitation will occur on bacterial cell walls provided high concentrations of calcium ions are close to the bacteria and carbonate ions in super saturation levels.

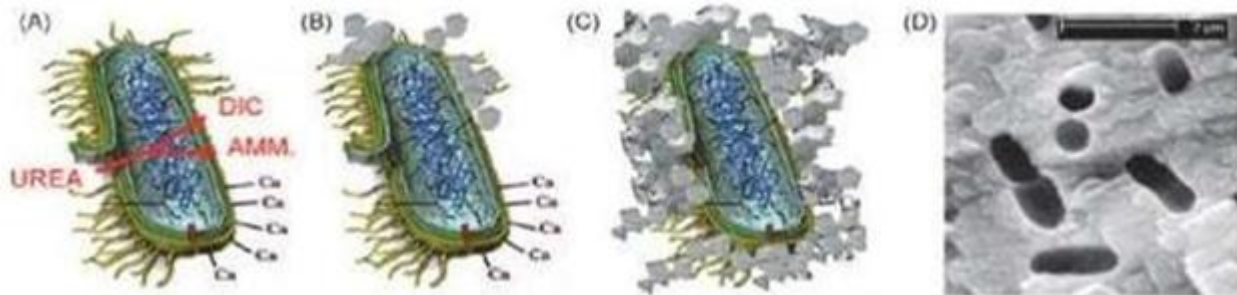
MICC was initially introduced by Ramakrishna, Jonkers, H.M. & Schlangen, E. (2007), Jonkers, H. (2008), and Mayur Shantilal Vekariya, & Prof. Jayeshkumar Pitroda (2012) to remediate cracks and fissures through self healing characteristics of microbial concrete. The tremendous improvement in physical properties of concrete and its behaviour with introduction of specific microorganism in concrete mortar has been attracting the interest of researchers in India. The previous studies *have* shown an improvement of 18% in compressive strength of cement mortar with Bacillus and Pseudomonas aeruginosa [14, 18, 22]. As mentioned earlier some of the bacterial species are used to produce urease acting as catalyst to produce CO<sub>2</sub> and NH<sub>3</sub> which increases pH in the surrounding to precipitate ions Ca<sup>2+</sup> and CO<sub>3</sub><sup>2-</sup> as CaCO<sub>3</sub>. The possible biochemical reactions to precipitate calcium carbonate (CaCO<sub>3</sub>) at the cell surface providing a nucleation site is summarized as below:



Volume 5 Issue 12, December 2016

[www.ijsr.net](http://www.ijsr.net)

Licensed Under Creative Commons Attribution CC BY



**Figure 1:** Precipitation of Calcium Carbonate (Calcite) by bacterial cell

The figure 1 depicts the events occurring during microbially induced carbonate precipitation processes. In the process (A) calcium ions are attracted towards the bacterial cell wall being negatively charged. It is reported that in the microenvironment of the bacteria the dissolved inorganic carbon and ammonium are released when urea is added to the bacteria. The process (B) where the presence of calcium ions results in a phenomenon of local super saturation state leading to heterogeneous precipitation of calcium carbonate seen on bacterial cell wall and after some times the whole cell is seen encapsulated as depicted in fig 1(c). The image (D) is depicted the involvement of bacterial cell being in dead state after restricted nutrient transfer in calcium carbonate precipitation.

## 2. Application, Advantages and Disadvantages

The usage of microbial concrete is becoming popular. The use of microorganisms in concrete provided more life to concrete structures. The self sealing of cracks, natural surface treatment, improvement in physical properties, long lasting of river banks, durable roads and erosion prevention of loose sand with capability to arrest corrosion in steel used in RCC have made microbial concrete significantly more popular in bio civil engineering. The availability of microorganisms is no issues as this is abundantly available in nature and produced under laboratory conditions also. The microbes viz *Bacillus Subtilis*, *Bacillus Thuringiensis* and *Bacillus Sphaericus* used in concrete basin provide excellent purification results to clean the stream of rivers by eliminating smell of water. This technique could effectively be applied for tank walls and floor linings, lake water, sewerage channels and for stagnant water. With this technique the BOD of 1400 could be reduced to 1.4. The microbial concrete being eco-friendly has numerous advantages and disadvantages too, as given below:

### Advantages

- Enhancement in compressive strength and stiffness values
- Remediation/ sealing of cracks in concrete
- Due to bacterial chemical process better resistant towards freeze-thaw attack reduction
- Due to surface treatments reduction in gas permeability
- Arrest in corrosion due to consumption of oxygen ingress through substances
- By sealing the paths of ingress reduction in corrosion of reinforced concrete
- Better life of concrete structures/ surfaces
- Reduction in water and chloride ion permeability
- Resistance to acid at pH greater than 1.5

- Eco friendly, antifungal, natural water purifier
- Restoration material for stone building/ monuments

### Disadvantages

- Cost of bacterial concrete doubled in comparison to conventional concrete
- Poor bacterial growth in any atmosphere and media
- No prescribed IS code in terms of doses of bacteria for optimum performance
- Costly investigation as calcite precipitation layers are complex, requiring skilled staff
- Completion of bacterial activity a great concern
- Limited research in the field of MICP
- Lacks in substantial commercial applications

## 3. Types of Microbial Concrete and Various Microbes

The widely used microbial concrete for healing the cracks, surface treatment and as a water purifier along with microbes and a group of microbes with specific species for a particular operation used in constructional materials for Calcium Carbonate precipitation in addition to crystal, metabolism, nutrients and applications are tabulated in Table 1, 2, 3.

**Table 1:** Types of microbes used in concrete [15, 20, 22, 37, 41, 42, 50, 53, 54]

Sl. No.	Name of the microbes	Types of microbial concrete
1.	i. <i>Sporosarcina pasteurii</i> [20] ii. <i>Bacillus pasteurii</i> [22] iii. <i>Bacillus pasteurii</i> [42] iv. <i>Bacillus sphaericus</i> [50]	Crack healing microbial concrete
2.	i. <i>Bacillus pseudifirmus</i> [41] ii. <i>Bacillus cohnii</i>	Self Healing microbial concrete
3.	i. <i>Bacillus sphaericus</i> ii. <i>E. coli</i> . [42]	Microbial concrete for surface treatment
4.	i. <i>Bacillus cereus</i> [15] ii. <i>Bacillus pasteurii</i> [22] iii. <i>Shewanella</i> [37] iv. <i>Sporosarcina pasteurii</i> [53] v. <i>Bacillus sp. CT-5</i> [54]	Concrete and as cement mortar
5.	<i>Bacillus phaericus</i> , <i>Bacillus Subitilis</i> , <i>Thiobacillus</i> , <i>Bacillus thuringiensis</i>	Microbial concrete as water purifier

**Table 2:** Various construction Materials using MICP [20, 42, 60]

Microorganisms	Metabolism	Nutrients	Application
Bacillus subtilis	Hydrolysis of urea	Nutrient broth, urea, CaCl <sub>2</sub> .2H <sub>2</sub> O, NH <sub>4</sub> Cl, NaHCO <sub>3</sub>	Crack in concrete remediation
Bacillus Cercus	Oxidative deamination of amino acids	Growth media (peptone, extract yeast, KNO <sub>3</sub> , NaCl) + CaCl <sub>2</sub> .2H <sub>2</sub> O, Actical, Natamycine	Biological Mortar
Bacillus subtilis	Hydrolysis of urea	Nutrient broth, urea, CaCl <sub>2</sub> .2H <sub>2</sub> O, NH <sub>4</sub> Cl, NaHCO <sub>3</sub>	Bacterial concrete
Bacillus sphaericus	Hydrolysis of urea	Extract yeast, urea, CaCl <sub>2</sub> .2H <sub>2</sub> O	Crack in concrete remediation
Bacillus subtilis	Oxidative deamination of amino acid	Peptone: 5g/ lit, NaCl: 5g/lit, yeast extract: 3 g/lit	Bacterial concrete

**Table 3:** Microorganisms for Calcite Precipitation [13,15,19,30,60]

Microorganisms	System	Types of Crystal
Photosynthetic organism: Synechococcus GL 24	Meromictic lake	Calcite (CaCO <sub>3</sub> )
Nitrogen cycle Bacillus cerens	Ammonification and nitrate reduction	Calcite (CaCO <sub>3</sub> )
Sulphate Reducing Bacteria: Isolate SRB LV form 6	Anoxic hypersaline lagoon	Dolomite (Ca(Mg)CO <sub>3</sub> )
Nitrogen cycle Bacillus subtilis	Urea degradation in synthetic medium	Calcite (CaCO <sub>3</sub> )
Photosynthetic organism: Chlorella	Lurcene Lake	Calcite (CaCO <sub>3</sub> )
Nitrogen cycle Bacillus subtilis JC3	Ammonification (Ammono acid degradation)	Calcite (CaCO <sub>3</sub> )

#### 4. Influence of Microbes on Various Properties of Concrete

##### A. Compressive strength

With the inclusion of microbial biomass viz Bacillus pasteurii [22, 37], the compressive strength of concrete is enhanced from 7 days to 28 days as shown in Table 4. It can also be seen that average mortar compressive strength with the addition of anaerobic microorganism increases to an extent of 25.29% relative to control in 28 days but starts reducing relatively to 11.06% when cell consumption per ml of water is increased further from 105 to 107 as shown in Table 5.

**Table 4:** Variation of compressive strength with biomass Vs days

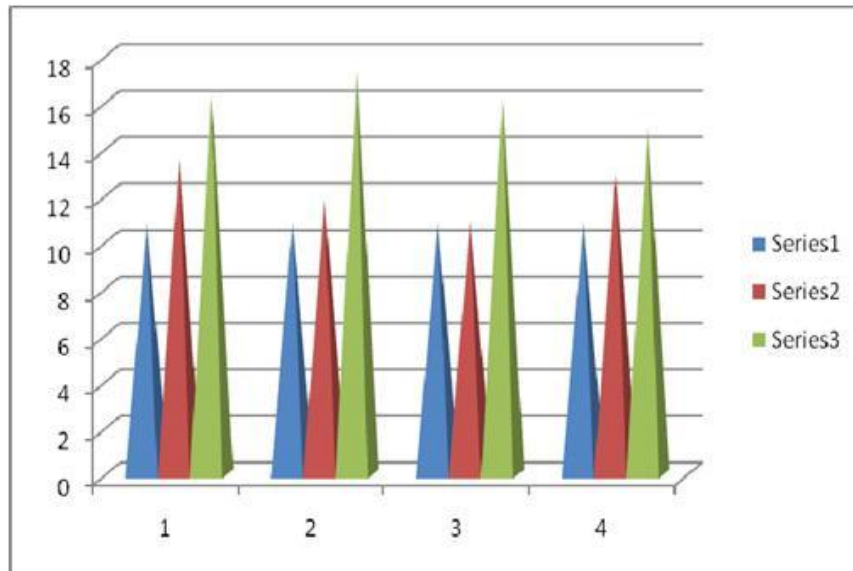
	Bacillus pasteurii, cells/cm <sup>3</sup>	Compressive Strength, MPa		Compressive Strength, MPa	
		After 7-days		After 28-days	
		Average	Standard deviation	Average	Standard deviation
Control	0	47	1.38	55	1.27
Live	3.8 X 10 <sup>3</sup>	55	2.39	62	3.23
	3.8 X 10 <sup>5</sup>	54	1.55	63	1.97
	3.8 X 10 <sup>7</sup>	57	1.27	65	0.87
Killed	3.8 X 10 <sup>3</sup>	53	3.73	56	2.36
	3.8 X 10 <sup>5</sup>	59	3.46	55	2.42
	3.8 X 10 <sup>7</sup>	60	1.81	61	0.92
Live	7.6 X 10 <sup>3</sup>	46	2.08	65	0.81
	7.6 X 10 <sup>5</sup>	50	2.73	55	2.33
	7.6 X 10 <sup>7</sup>	55	1.28	55	0.44
Killed	7.6 X 10 <sup>3</sup>	57	1.47	59	3.19
	7.6 X 10 <sup>5</sup>	61	2.88	56	2.57
	7.6 X 10 <sup>7</sup>	66	1.41	58	0.79

**Table 5:** Effect of the anaerobic microorganism's addition on mortar strength

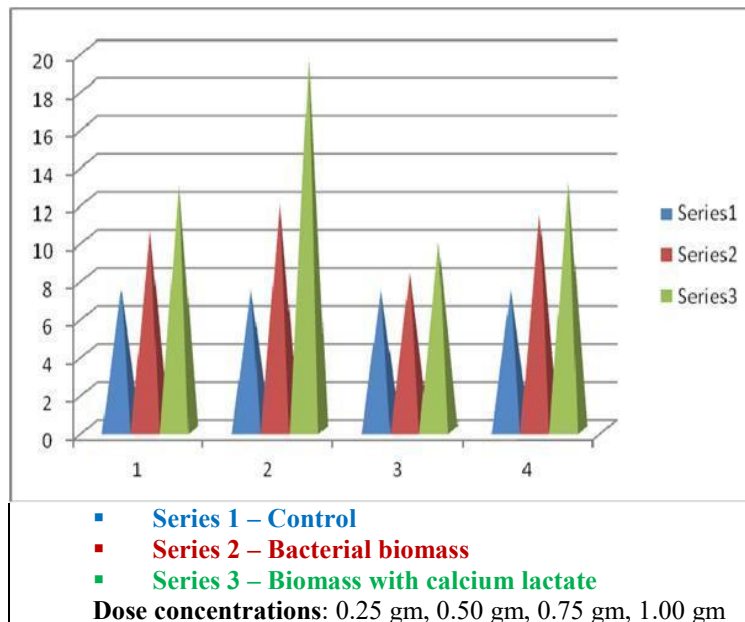
Cell conc./ ml of water	Average Mortar Compressive Strength in MPa					
	7 days		14 days		28 days	
	Strength ±S.D	% Increase relative to control	Strength ±S.D	% Increase relative to control	Strength ±S.D	% Increase relative to control
Nil	12.60±0.47	-	16.00±0.81	-	23.13±0.23	-
10	12.74±0.89	1.11	16.21±0.22	1.31	24.21±0.43	4.66
102	12.87±0.46	2.14	16.44±0.38	2.75	25.00±0.88	8.08
103	12.98±0.81	3.01	16.87±0.64	5.43	25.40±0.84	9.81
104	13.4±0.53	6.34	17.10±0.37	6.87	25.44±0.97	9.98
105	14.70±0.74	16.67	19.50±0.42	21.87	28.98±0.86	25.29
106	13.80±0.58	9.52	17.50±0.81	9.38	26.52±0.27	14.65
107	13.00±0.23	3.17	17.00±0.45	6.25	25.69±0.74	11.06

The comparative compressive strength (Mpa) of mortar cube test specimen for control, bacterial biomass and biomass with calcium lactate at different dose concentrations (0.25, 0.50, 0.75, 1.00 gm) using *Bacillus cohnii* and *Bacillus megaterium* is shown at figures 2 and 3. The compressive strength of mortar cube test specimens with *Bacillus cohnii* as microorganism increases in case of biomass with calcium lactate with a dose concentration of 0.5 gm to approx 17Mpa amongst all four doses of concentration for control, bacterial biomass and biomass with calcium lactate whereas except for bacterial biomass dose of 0.25gm compressive strength of mortar cube decreases for rest of dose concentrations.

Similarly the compressive strength of mortar cube test specimens with *Bacillus megaterium* as microorganism increases in case of biomass with calcium lactate with a dose concentration of 0.5 gm to approx 19Mpa amongst all four doses of concentration for control, bacterial biomass and biomass with calcium lactate whereas except for bacterial biomass dose of 0.50gm compressive strength of mortar cube decreases for rest of dose concentrations. This trend of an increment in compressive strength up to 28 days could be due to the behavior of bacterial cells within the mortar matrix.



**Figure 2:** Comparative compressive strength (Mpa) of mortar using *Bacillus cohnii* at different doses



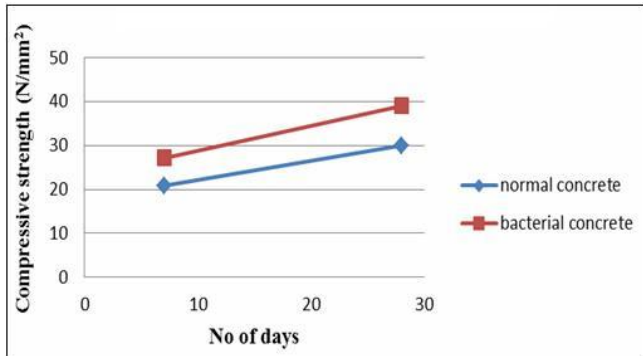
**Figure 3:** Comparative compressive strength (Mpa) of mortar using *Bacillus megaterium* at different doses

**B. Compression and Flexural Strength**

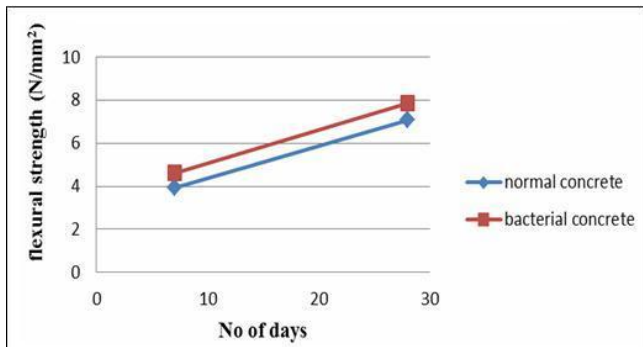
The compressive strength test and flexural strength test comparison on concrete cubes and beams for different number of days for normal and bacterial concrete is shown at figure 4 & 5 and Table 6 where it can be visualized that both compressive and flexural strengths are linearly increasing with number of days of cementation process with

the reason that the precipitation of calcite heals the pores to increase the density of bacterial concrete over the normal concrete by reducing the porosity. The compressive strength and flexural strength values for specimen concrete test cubes and beams of bacterial concrete has an edge over normal concrete throughout the test period.





**Figure 4:** Compressive strength test comparison on concrete cubes



**Figure 5:** Flexural strength test comparison on concrete beams

**Table 6:** Compressive strength and Flexural strength tests

	Normal concrete	Bacterial concrete	Days
Compressive strength on concrete cubes, N/mm <sup>2</sup>	20.84	27.09	7
	29.99	38.98	28
Flexural strength on concrete beams, N/mm <sup>2</sup>	3.92	4.6	7
	7.06	7.85	28

### C. Weight Increase

The weight increase is revealed in specimens while specimens used water-cement ratio of 0.5 and 0.7 during test of influence of w/c ratio and calcium source on a deposited carbonate by De Muynck et al., 2005. However during sportivity test by De Muynck et al., (2008) the water penetration in the specimen was carried out though with the absorption of bacteria and formation of carbonate crystals as precipitation increases the weight of mortar specimens and when water absorption comparison were carried out with untreated samples it was found more pronouncing for w/c 0.7. The shrinkage of concrete with bacterial suspension in different medium is reported to be 13%, 20% and 34% less than that of the control beam when beams with bacterial concentration of  $1 \times 10^6$  cells/ml,  $1 \times 10^7$  cells/ml and  $1 \times 10^8$  cells/ml, are applied at 28 day of investigation [34]. Beams made with bacteria suspended in water, urea-CaCl<sub>2</sub> and phosphate-buffer is reported to have less mean expansions than that of the control beams and as it shows resistance towards alkalinity [34]. Likewise in reference to resistance towards sulphate attack the effect of different concentration of bacteria is also reported to have less mean expansions than that of the control beams [34].

### D. Influence of nickel ion concentration

It was found that the incorporation of nickel ion served the purpose of increasing the urease activity as well as increased calcium carbonate precipitation as urease is nickel ion dependent enzyme. The Effect of nickel ion concentration on calcite precipitation using *Bacillus cohnii* was found to be higher in nickel ion containing culture medium in comparison to control sample. More specifically, to serve the purpose of increased calcite precipitation, nickel ion concentration of 5µm was found appropriate.

### 5. Conclusions

The microbial concrete is becoming popular in Civil Engineering as this technology is proving better than any available conventional technology because of its being eco-friendly and self-healing. It improves the physical properties, increases durability of various building materials, improves compressive strength, reduces permeability & water absorption, arrest the corrosion in steel and very convenient for usage. The *B.cohnii* and *Bacillus megaterium* were found urease positive and able to precipitate calcite within mortar specimen. Nickel as a promoter of urease activity has a profound effect on bacterial growth when incorporated in urea based broth prior to inoculation. In addition to this, a dose of 0.5 gm of the dry bacterial biomass can be used as a standard dose to fulfill the aim of crack filling as response of 0.5gm of dose was found to be best than all other doses used.

### 6. Acknowledgement

The data liberally used from the publications of Ravindranatha, et, al; Varenayam Achal, et. al.; Leena Chaurasia, et.. al.; Mohini P. Samudre, et. al.; Beena Kumari, as at reference [67], [68], [69], [70], [71], [72] is gratefully acknowledged.

### References

- [1] Franklin C, McLean and Baird Hastings A. (1934). A biological method for the estimation of calcium ion concentration. *J. Biol. Chem.* 107: 337.
- [2] IS: 12269 (1987), Indian standard specification for 53 grade ordinary Portland cement, Bureau of Indian standard, New Delhi.
- [3] IS 4301: part 4 (1988), Methods of physical tests for hydraulic cement: determination of consistency of standard cement paste, Bureau of Indian standard, New Delhi.
- [4] IS 4301: part 6 (1988), Methods of physical tests for hydraulic cement: determination of compressive strength of hydraulic cement (other than masonry cement) (with amendment no.1), Bureau of Indian standard, New Delhi.
- [5] Hewlett, P.C., (1990), "Methods of protecting concrete – coatings and linings". In: Dhir RK, Green JW, editors. *Proceedings of international conference: Protection of concrete.* Scotland: Dundee; p. 105–34.
- [6] J. G. Holt, Ed., *Bergey's Manual of Determinative Bacteriology*, 9th ed. Williams & Wilkins, Baltimore, 1994.

- [7] Zhong, L., and Islam, M. R. (1995). "A new microbial process and its impact on fracture remediation." *70<sup>th</sup> Annual Technical Conference and Exhibition of the Society of Petroleum Engineers*, Oct 22-25, Dallas, Texas.
- [8] Nolan, E.; Basheer, P.A.M. & Long, A.E. (1995). "Effects of three durability enhancing products on some physical properties of near surface concrete", *Construction and Building Materials*, Vol. 9, pp. 267–272.
- [9] Ramachandran V.S. (Ed), *Concrete Admixtures Handbook; Properties, Science & Technology*, Noyes Publications, New Jersey, 1995, pp 1007-1009.
- [10] Gollapudi, U. K., Knutson, C. L., Bang, S. S., and Islam, M. R. (1995). "A new method for controlling leaching through permeable channels." *Chemosphere*, 30, 695-705.
- [11] Ciurli S, Marzadori C, Benini S, Deiana S, Gessa C., "Urease from the soil bacterium: *Bacillus pasteurii* immobilization on Capolygalacturonate." *Soil Biol Biochem* 28, 1996, pp. 811-817.
- [12] Claisse, P. A., Elsayad, H. A., and Shaaban I. G. (1997). "Absorption and sorptivity of cover concrete." *Journal of Materials in Civil Engineering*, 9, 105-110.
- [13] Tai C.Y. and Chen F.B., Polymorphism of  $\text{CaCO}_3$  precipitated in a constant composition environment. *AIChE Journal*, 44(8), 1790-1798 (1998)
- [14] Ramakrishnan V., Bang SS and Deo KS. (1998). A novel technique for repairing cracks in high performance concrete using bacteria. Proc. Int. conf. on high performance high strength concrete, Perth, Australia, 597-618.
- [15] Le Metayer-Levrel, G., Castanier, S., Oriol, G., Loubiere, J.F., Perthuisot, J.P. (1999), "Applications of bacterial carbonatogenesis to the protection and regeneration of lime stones in buildings and historic patrimony". *Sediment Geol.*; 126(1-4):25-34.
- [16] Castanier S., Le Metayer-Levrel G., Oriol G., Loubiere J.F., Perthuisot J.P., (1999), "Bacterial carbonatogenesis and application of preservation and restoration of historic property", *Proceedings of microbes and art; the role of microbial communities on the degradation and protection of cultural heritage*, Florence, Italy.
- [17] P. Tiano, L. Biagiotti, G. Mastromei, "Bacterial biomediated calcite precipitation for monumental stones conservation: methods of evaluation", published by *Journal of Microbiological Methods*, Vol.36, Issues 1-2, 1999, pp.139-145
- [18] Stocks-Fischer S., Galinat J and Bang S. (1999). Microbiological precipitation of  $\text{CaCO}_3$ . *Soil Biol. Biochem.* 31: 1563- 1571.
- [19] González-Muñoz M.T., Ben-Chekroun K., Ben-Aboud A., Arias J.M. and Rodríguez-Gallego M., Bacterially induced Mg-Calcite formation: Role of  $\text{Mg}^{2+}$  in development of crystal morphology, *Journal of Sedimentary Research*, 70, 559-564 (2000)
- [20] Bang, S.S. and Ramakrishnan, V. (2001). "Microbiologically-enhanced crack remediation (MECR)." *Proceedings of the International Symposium on Industrial Application of Microbial Genomes*, Daegu, Korea, 3-13.
- [21] Basheer, L., Kropp, J., Cleland, D.J., (2001), "Assessment of the durability of concrete from its permeation properties: a review". *Constr Build Mater.*; 15(2-3):93-103.
- [22] Ramachandran SK., Ramakrishnan V and Bang S.S. (2001). Remediation of concrete using microorganisms. *ACI Mater. J.* 98: 3-9.
- [23] Bachmeier KL, Williams AE, Warmington JR, Bang SS., "Urease activity in microbiologically-induced calcite precipitation." *Journal of Biotechnology* 93, 2002, pp. 171-181.
- [24] J. L. Day, A. V. Ramakrishnan, and S. S. Bang, "Microbiologically Induced Sealant For Concrete Crack Remediation," *16th Engineering Mechanics Conference*, 2003, pp.1-8.
- [25] Rodríguez-Navarro, C., Rodríguez-Gallego, M., Ben Chekroun, K., González-Munoz, M.T., (2003) "Conservation of ornamental stone by myxococcus xanthus-induced carbonate biomineralization". *Appl Environ Microbiol.*; 69(4):2182-93.
- [26] Hammes, F., Seka, A., De Knijf S., Verstraete, W., (2003), "A novel approach to calcium removal from calcium-rich industrial wastewater". *Water Res.*; 37(3):699-704.
- [27] Hamilton, W.A. (2003), "Microbially influenced corrosion as a model system for the study of metal microbe interactions: a unifying electron transfer hypothesis", *Biofouling*, Vol.19, 65-76.
- [28] Nermati, M., Voordouw, G., (2003), "Modification of porous media permeability using calcium - carbonate produced enzymically in situ". *Enzyme Microbe Technol.*; 33(5); 635-42.
- [29] Khan, M. I. (2003). "Isoresponses for strength, permeability and porosity of high performance Mortar." *Building and Environment*, 38, 1051-1056.
- [30] Sanchez-Moral S., Canaveras J.C., Laiz L., Saiz-Jimenez C., Bedoya J., and Luque L. Biomediated precipitation of calcium carbonate metastable phases in hypogean environments: A short review, *Geomicrobiology Journal*, 20(5), 491-500 (2003)
- [31] Hammes, N. Boon, J. De Villiers, W. Verstraete, S. D. Siciliano, and J. De Villiers, "Strain-Specific Ureolytic Microbial Calcium Carbonate Precipitation," published in *Applied and Environmental Microbiology*, vol.69, no.8, 2003, pp. 4901-4909.
- [32] Annie Peter, J., Lakshmanan, N., Devadas Manoharan, P., Rajamane, N.P. & Gopalakrishnan, S. — Flexural Behaviour of RC Beams Using Self Compacting Concrete, published in *The Indian Concrete Journal*, June 2004, PP 66-72
- [33] Horner-Devine MC, Carney KM, Bohannon BJM, "An ecological perspective on bacterial biodiversity". *Proceedings Royal Society Biological Science B* 271, 2004, pp.113-122.
- [34] V. Ramakrishnan, R. K. Panchalan, S. S. Bang, and R. City, "Improvement of Concrete Durability By Bacterial Mineral Precipitation," *International Conference on Fracture*, 2005.
- [35] Muynck W, Dick J, De Graef B, De Windt W, Verstraete W, De Belie N., "Microbial ureolytic calcium carbonate precipitation for remediation of concrete surfaces." *Proceedings of International conference on*

- concrete repair, rehabilitation and retrofitting, South Africa, Cape Town, 2005, pp. 296-297.
- [36] De Belie M., De Graef B., De Muyanck, W. and Verstraete, W., (2005). "Bacteria as protagonists for concrete: Bacterial cleaner and bacterial builder". fib Symposium "Keep Concrete Attractive", Budapest.
- [37] Ghosh P., Mandal S., Chattopadhyay B and Pal S. (2005). Use of microorganism to improve the strength of cement mortar. *Cement and Concrete Research*. 35:1980-1983.
- [38] Dick, J.; De Windt, W.; De Graef, B.; Saveyn, H.; van der Meer, P.; De Belie, N. & Verstraete, W. (2006). "Bio-deposition of a calcium carbonate layer on degraded limestone by *Bacillus* species", *Biodegradation* Vol.17, pp.357-367.
- [39] Moon, H.Y., Shin, D.G. & Choi, D.S. (2007). "Evaluation of the durability of mortar and concrete applied with inorganic coating material and surface treatment system", *Construction and Building Materials*, Vol. 21, pp. 24-33.
- [40] Jonkers, H. (2007). "Self healing concrete: a biological approach. In: van der Zwaag, S. (Ed.), *Self Healing Materials: An alternative Approach to 20 Centuries of Materials Science*", Springer, Netherlands, pp. 195-204.
- [41] Jonkers, H.M. & Schlangen, E. (2007). "Crack repair by concrete-immobilized bacteria". In: Schmetts, A.J.M., van der Zwaag, S. (Eds.), *Proc. of First International Conference on Self Healing Materials*, Noordwijk, The Netherlands.
- [42] De Muyanck, W., Debrouwer, D., De Belie, N., Verstraete, W., (2008) ", Bacterial carbonate precipitation improves the durability of cementitious materials". *Cement and Concrete Research* 38, 1005-1014.
- [43] De Belie, N., De Muynck, W., (2009), "Crack repair in concrete using bio deposition," *Concrete Repair, Rehabilitation and Retrofitting II*, pp.777-782.
- [44] Achal V., Mukherjee A., Basu P. C. and Reddy M.S. (2009a). Lactose mother liquor as an alternative nutrient source for microbial concrete production by *Sporosarcina pasteurii*. *Journal of Industrial Microbiology and Biotechnology* 36:433-438.
- [45] Achal, V., Mukherjee, A., Basu, P. C., and Reddy, M. S. (2009), "Strain improvement of *sporosarcina pasteurii* for enhanced urease and calcite production", *Journal of industrial Microbiology and Biotechnology*, 36, 981-988
- [46] H. M. Jonkers and E. Schlangen, "A two component bacteria-based self-healing concrete," *Concrete Repair, Rehabilitation and Retrofitting II*, 2009, pp. 215-220.
- [47] Jonkers, H.M.; Thijssen, A.; Muyzer, G.; Copuroglu, O. & Schlangen, E. (2010). "Application of bacteria as self-healing agent for the development of sustainable concrete", *Ecol Eng*, Vol.36, pp.230-235.
- [48] K. D. Arunachalam, K. S. Sathyanarayanan, B.S. Darshan, and R. B. Raja, "Studies on the characterization of Bio sealant properties of *Bacillus sphaericus*," *International Journal of Engineering Science and Technology*, vol.2, no.3, 2010, pp. 270-277.
- [49] J.Y. Wang, K. Van Tittelboom, N. De Beliel and W. Verstraete. Potential of Applying Bacteria to Heal Cracks in Concrete published in conference on substantial construction materials and technologies in June 2010
- [50] V. Achal; A. Mukherjee; and M. S. Reddy. 2010. *Microbial Concrete: A way to enhance the Durability of Building Structures*. *J. Materials for Civil Engg.*
- [51] S. Sunil Pratap Reddy, M.V. Seshagiri Raob, P. Aparnac and Ch. Sasikalac. Performance of standard grade bacterial (*bacillus subtilis*) concrete published in *asian journal of civil engineering (building and housing)* vol. 11, no. 1 (2010) pages 43-55
- [52] S. S. Bang, J. J. Lippert, U. Yerra, S. Mulukutla, and V. Ramakrishnan, "Microbial calcite, a bio-based smart nano material in concrete remediation," *International Journal of Smart and Nano Materials*, vol.1, no.1, 2010, pp. 28-39.
- [53] Achal, V., Mukherjee, A. & Reddy, M.S., (2011a). "Effect of calcifying bacteria on permeation properties of concrete structures", *J Ind Microbiol Biotechnol*, Vol. 38, pp. 1229-1234.
- [54] Varenayam Achal; Abhijit Mukherjee; and M. Sudhakara Reddy. "Microbial Concrete: Way to Enhance the Durability of Building Structures "This paper is part of the *Journal of Materials in Civil Engineering*, Vol. 23, No. 6, June 1, page 730-734, 2011.
- [55] Muhammad Isha, Afifudin. H and Mohd Saman H. "Bacillus Subtilis – And Thermus Thermophilis – Derived Bioconcrete in enhancing concrete compressive strength ", *International sustainability and civil engineering journal*, Vol. 1, No.1 (July 2012 ).
- [56] Dhama N., Reddy M and Mukherjee A. (2012). Improvement in strength properties of ash bricks by bacterial calcite. *Ecol Eng* 39: 31-35.
- [57] Chahal N., Siddique R and Rajor A. (2012). Influence of bacteria on the compressive strength, water absorption and rapid chloride permeability of fly ash concrete. *Construction and Building Materials* 28(1): 351- 356.
- [58] Annamalai S., Arunachalam K and Sathyanarayanan K., (2012). Production and characterization of Bio Caulk by *Bacillus pasteurii* and its remediation properties with carbon nano tubes on concrete fractures and fisures. *Materials Research Bulletin* 47(1): 362- 368.
- [59] C. C. Gavimath, B. M. Mali, V. R. Hooli, J. D. Mallpur, A. B. Patil, D. P. Gaddi, C.R. Ternikar and B.E. Ravishankera. Potential application of bacteria to improve the strength of cement concrete published by *International Journal of Advanced Biotechnology and Research* ISSN 0976- 2612, Vol 3, Issue 1, 2012, pp 541-544
- [60] Srinivasa Reddy V, Achyutha Satya K, Seshagiri Rao M V, Azmatunnisa. Biological approach to enhance strength and durability in concrete structures published by *International Journal of Advances in Engineering & Technology*, Sept 2012 Vol. 4, Issue 2, pp. 392-399
- [61] Mayur Shantilal Vekariya, Prof. Jayeshkumar Pitroda Addressing Infrastructure Durability and Sustainability by Self Healing Mechanisms - Recent Advances in Self Healing Concrete and Asphalt Published by Elsevier Ltd. Vol. 54 in 2013 page no, 39-57

- [62] Srinivasa Reddy V, Sreenivasa Rao D, Seshagiri Rao M V, Sasikala Ch. Permeation Properties of Bacterial Concrete published in IOSR Journal of Mechanical and Civil Engineering (IOSR-JMCE) e-ISSN: 2278-1684 Volume 5, Issue 6 (Mar. - Apr. 2013), PP 08-12
- [63] V. Ramakrishnan, Ramesh K. Panchalan & Sookie S. Bang, Improvement of concrete durability by bacterial mineral precipitation, published by Elsevier.
- [64] S.A. Abo-El-Enein , A.H. Ali , Fatma N. Talkhan , H.A. Abdel-Gawwad “Application of microbial biocementation to improve the physicomechanical properties of cement mortar” published in HBRC Journal (2013) 9, 36–40
- [65] Jagadeesha Kumar B G, Prabhakara R, Pushpa H. (2013). Bio mineralisation of calcium carbonate by different bacterial strains and their application in concrete crack remediation. International journal of advances in engineering and technology. 6(1): 202-213.
- [66] Abhijitsinh Parmar, Ankit Patel, Vismay Shah , Sandeep Khorasiya, Dipan Patel. (2013). “Improvement on the concrete cracks by using Bacillus pastuerii.” International Journal for Scientific Research & Development (IJSRD) | Vol. 1, Issue 1, 2013
- [67] Ravindranatha1, N. Kannan, Likhith M. L, “Self-Healing Material Bacterial Concrete”, IJRET: International Journal of Research in Engineering and Technology eISSN: 2319-1163 | pISSN: 2321-7308
- [68] Varenayam Achal, Abhijit Mukherjee, and M. Sudhakara Reddy, “Microbial Concrete: A Way to Enhance Durability of Building Structures”, Thapar University, Patiala, Punjab, India-147 004.
- [69] Mayur Shantila Vekariya, Jayeshkumar Pitroda, “Bacterial Concrete: New Era For Construction Industry”, B.V.M. Engineering College, Vallabh Vidhyanagar- Gujarat-India, (2013)
- [70] Leena Chaurasia, Rajesh K Verma and Vishakha Bisht, “Microbial Carbonate Precipitation by Urease Producing Bacteria in Cementitious Materials”, Environmental Science and Technology Group, CSIR-Central Building Research Institute, Roorkee-247 667, India (2014)
- [71] Mohini P. Samudre, M. N. Mangulkar , S. D. Saptarshi, “A Review of Emerging Way to Enhance theDurability and Strength of ConcreteStructures: Microbial Concrete” , JNEC, Aurangabad, Maharashtra, India (2014)
- [72] Beena Kumari, “Microbial Concrete: A Multi-Purpose Building Material-An Overview”, Thapar University, Patiala, Punjab, India, (2015)