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EFFECT OF CRUSHED SAND AND BACILLUS SUBTILIS ON THE CANTABRO LOSS OF BACTERIAL CONCRETE

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ABSTRACT

Bacterial concrete has emerged as a remedial measure for healing cracks in structures such as bridges, RCC buildings, RCC pipes, canal linings and pavements. Crack formation is an extremely common occurrence in concrete structures, and allows water and different chemicals to enter the concrete through cracks, diminishing its strength. In addition, it has consequences on the reinforcement once it comes into contact with water, CO_2 and other chemicals. The repair of cracks within concrete requires regular maintenance and special kinds of treatment, which can be very expensive. In bacterial concrete, particular types of microorganism can be extremely useful for refurbishing cracks in existing concrete structures. In this research, an experimental investigation was made to prevent cracks in concrete using *Bacillus subtilis* bacteria and calcium lactate. *Bacillus subtilis* bacteria with calcite lactate were used at varying percentages of 5%, 10% and 15% cement weight for M40 grade concrete. The fine aggregate used in all the mixes was crushed rock sand. A Cantabro loss test was conducted for all the mixes after 3, 7, 14 and 28 days of curing. An empirical relation between flexural and compressive strength is proposed in the form of $f_t = 0.66\sqrt{f_{ck}}$ for river sand mixes and $f_t = 0.89 f_{ck}^{\ 0.46}$ for crushed rock sand. An empirical relation is also proposed between Cantabro loss and flexural strength for bacterial concrete.

Keywords: *Bacillus subtilis*; Bacterial concrete; Cantabro loss; Compressive strength; Flexural strength; Split tensile strength

1. INTRODUCTION

In any building construction, cement concrete is a primary substance in the modern era of infrastructural projects across the globe. Like this material is lying face down to fracture in arrears towards in elastic and less resistant to straining demands the practice of rebar in it. Since it bonds with steel bars, concrete becomes more effective in resisting tension than without any reinforcement, and the tensile strength of concrete is relatively lower than compressive strength. Any cracks that have formed expose the reinforcement and thus affect its structural integrity, leading to corrosion. However, it does crack and endure genuine wear and tear throughout the years of its predictable term of service (Wang et al., 2012) but is not versatile and cannot handle high levels of strain. Ordinary concrete can tolerate from near zero to 1% strain before giving out. Concrete that is able to heal on its own normally seeks to repair these flaws, thus increasing the service lifetime of any given concrete structure. There exists a material which is a type of

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Self-healing concrete in growth which will solve several of the issues usually related with ordinary concrete (Ramadhansyah et al., 2011). Self-healing concrete consists of a mixture of microorganisms (*Bacillus subtilis*) fused into the concrete, and calcium lactate and nutrient broth food to support these microorganisms once they become active (Li & Herbert, 2012). The microorganisms, feeding on the food supply provided, heal the cracks. This paper will make an in-depth case for the method that extent part behind microorganism that helps to heal the concrete, and describes the various parts that extent part enclosed within the process and the way they work severally and put together. The paper deals with sensible applications of self-healing concrete, as well as its real-world integration in structures. The abrasion resistance, i.e. surface abrasion loss and cantabro loss at various ages of concrete from 3 days to 90 days, is influenced by the flyash content and presence of M-Sand (Rao et al., 2016). To measure the abrasion resistance on pervious concrete, a new method is proposed in combination with other three methods (the loaded wheel test, surface abrasion test, and Cantabro test). They concluded that the Cantabro test shows promising results (Dong et al., 2010).

1.1. The Biological Self-healing Process

It is vital to hustle what forms of bacteria can live in concrete, however they effort to rise the robustness of structures, what the chemical agents are with the purpose of causes the biochemical process within the microorganism, what takes place in the precise forms of specialized microorganism once unprotected to the substance, and the mode they work along to not solely seal cracks before they form, but also strengthen the overall structure. Once the bacteria are uncovered to the atmosphere and for that reason become “food,” the microorganisms stand an action that sources them to stabilize and wrath, filling in the crack that has formed. This method extends the robustness of the structure, additionally fixing any cracks that have occurred. The method of healing a crack takes around 28 days (Jonkers et al., 2010).

Concrete structures are presently designed in keeping with set standards that enable cracks to form up to 2 mm wide. Such small cracks are usually thought to be tolerable, as they do not directly weaken the structure. Moreover, small cracks generally patch themselves up, as numerous varieties of concrete display an explicit crack-healing capability. Analysis has shown that this ‘autonomous’ healing capability is essentially allied with the level of non-responded cement particles’ ability in the concrete. In crack development, water acts in response to these elements, leading to the closure of small cracks. However, owing to the unpredictability of the self-directed crack remediation of concrete buildings, water run as a results of negligible crack creation in underpass and underground structures will occur. 0.2 mm (Zwaag, 2008; Reddy et al., 2012) wide-ranging cracks were observed to self-heal in conventional samples, while all the cracks were healed in the samples that contain bacteria. The fundamental idea behind our specific version of this technique is the utilization of sure categories of bacteria (in this case *Bacillus subtilis*) and approach of operate to closure tiny cracks within the concrete earlier they develop into bigger and further durable to manage cracks and fissures. This bio- calcification method involves many steps to complete the tasks (Jonkers & Schlangen, 2007; Joseph et al., 2007).

Trendy techniques such as X-ray diffraction tests and scanning electron microscopy (SEM) analysis are employed to quantify the study of the stages of spar deposition (Kessler et al., 2003).

1.2. How Do Bacteria Remediate Cracks?

Once the mixing of concrete and bacteria is complete, the bacteria undergo inactive state. When they are exposed to the environment (air), all their functions are stimulated. When cracks form in concrete, the bacteria start to bring on calcite minerals to the crack (Ramakrishnan et al., 2001). At the point when the microorganisms interact with water and calcium lactate, the bacteria spores start germinating and the bacteria start feeding calcium lactate. This kind of pore is available in concrete up to two hundred years (Jonkers, 2011). Limestone heals cracks which have formed in concrete. By consuming the oxygen, the corrosion of steel decreases and the stability of reinforced concrete structures increases. The procedure of preparing synthetic calcium carbonate response from fractured calcium hydroxide can be identified properly (Schlangen et al., 2010).

$$CO_2 + Ca(OH)_2 \rightarrow CaCO_3 + H_2O \quad (1)$$

$$CaC_6H_{10}O_6 + 6O_2 \rightarrow CaCO_3 + 5CO_2 + 5H_2O \quad (2)$$

2. MATERIALS AND TESTING METHODS

2.1. Cement

53 grade OPC was used in this experimental work. The OPC was tested as per IS 4031-1996, with its physical properties shown in Table 1.

Table 1 Physical properties of Portland cement (53 grade)

S. No.	Test Property	Result	Requirements as per IS 12269-1987
1	Fineness		
	(a) Sieve test	2%	Not more than 10%
	(b) Blaine	285m²/kg	Min 225 m²/kg
2	Normal consistency	31.0%	-
3	Specific gravity	3.01	-
4	Initial setting time	95 minutes	Greater than 30 minutes
5	Final setting time	284 minutes	Less than 600 minutes
6	Compressive strength		
	(a) 3 days	28N/mm²	27 N/mm²(Min)
	(b) 7 days	41 N/mm²	37 N/mm²(Min)
	(c) 28 days	56N/mm²	53 N/mm²(Min)
7	Soundness (Le-Chatlier Exp.)	2mm	Not more than 10mm

2.2. Fine Aggregate

Locally available river sand and crushed rock sand were used as fine aggregates. For the fine aggregate, the distribution of particle size is shown in Figure 1. The specific gravity of the river sand and crushed rock sand was 2.68 and 2.77, respectively.

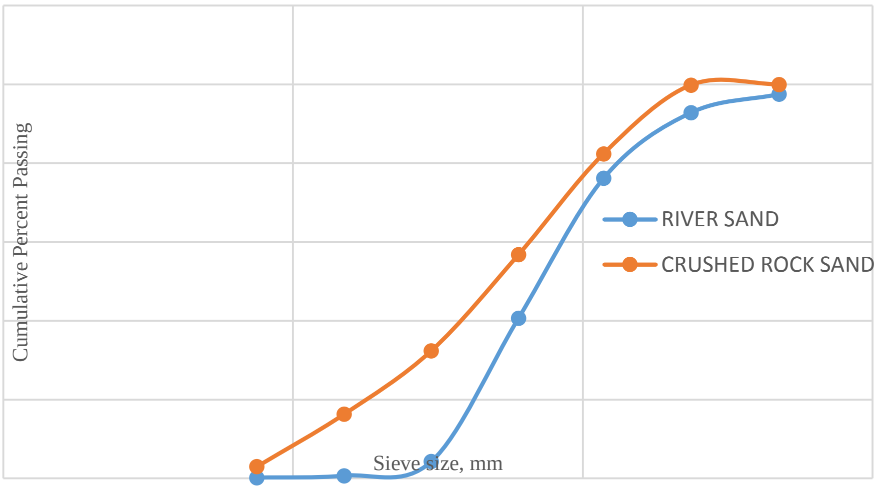

Figure 1 Particle size distribution curve of fine aggregate

2.3. Coarse Aggregate

Crushed granite broken stone of 20mm nominal size was used as coarse aggregate. The particle size distribution curves of the coarse aggregates are shown in Figure 2, with the specific gravity of the coarse aggregate being 2.71.

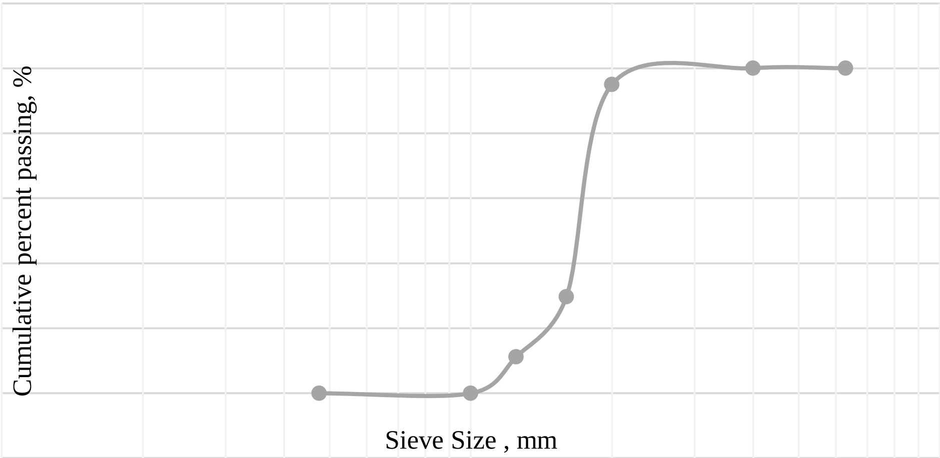

Figure 2 Particle size distribution curve of coarse aggregate

2.4. Water

Locally available potable drinking water was used in the experimental work for all mixes.

2.5 Bacteria

In this research, *Bacillus subtilis* bacteria were used, cultured at DVS Bio life Pvt Ltd Laboratory, Hyderabad, India.

2.6. Calcium lactate

Calcium lactate was used for the investigation, together with *Bacillus subtilis* bacteria as the nutrient broth. It was available in powder form with a white color.

2.7. Culturing of *Bacillus Subtilis*

Primary preparation of the nutrient broth (medium) was made by adding 2.5 grams of peptone, 1.5 grams of beef extract and 2.5 grams of sodium chloride (NaCl) to 500 ml of distilled water in a conical flask, which was enclosed with a cotton stopper and silver foil.

Figure 3 Making the nutrient broth solution

The solution was untainted using an autoclave for around 20 minutes at a constant temperature of 121°C and pressure of 15 lbs. After sterilization, the solution was impurity-free with a clear orange shade.

Figure 4 Contaminant-free solution after the sterilization process

Subsequently, the flask was opened in a lamina air flow chamber and a small pinch of the bacteria was added to the solution, as shown in Figure 5.

Figure 5 Adding the *Bacillus subtilis* to the nutrient broth medium in an aseptic room with laminar air flow equipment

It was then incubated in an orbital shaker at a speed of 125 rpm and temperature of 37°C. After 24 hours, it was observed that the shade of the bacterial arrangement had changed to a turbid whitish yellow, as can be seen in the figure which also shows the development of the *Bacillus subtilis*.

2.8. Mix Design

The mix proportions for the M40 grade concrete were designed using IS: 10262-2009. The materials required per 1 m^3 of concrete are shown in Table 2.

2.9. Compression Test

A compression test on bacterial concrete specimens with dimensions of 150×150×150 mm was conducted as per IS: 516-1959 specifications. The specimens were prepared and cured for 28, 90 and 365 days, as per IS:456-2000 standards.

2.10. Flexural Test

A flexural test on bacterial concrete prism specimens with dimensions of 500×100×100 mm was also conducted, as per IS:516-1959.

Table 2 Concrete mix proportions

Mixture	RBC00	RBC05	RBC10	RBC15	CBC00	CBC05	CBC10	CBC15
Cement (kg/m^3)	390	390	390	390	390	390	390	390
River sand (kg/m^3)	642	642	642	642	--	--	--	--
Crushed rock sand (kg/m^3)	--	--	--	--	642	642	642	642
Coarse aggregate (kg/m^3)	1261	1261	1261	1261	1261	1261	1261	1261
w/c ratio	0.42	0.42	0.42	0.42	0.42	0.42	0.42	0.42
Bacterial cells (cfu/ml)	10^5	10^5	10^5	10^5	10^5	10^5	10^5	10^5
Percentage of bacterial solution	00	05	10	15	00	05	10	15

RBC: Bacterial concrete with river sand; CBC: Bacterial concrete with crushed rock sand

2.11. Cantabro Loss Test

This test was performed with a Los Angeles abrasion testing instrument by the exclusive of abrasive charges in the form of steel balls, in accordance with ASTM C1747. The three cylindrical specimens of 150 mm diameter and 100 mm height were placed in the machine, as shown in Figures 6 and 7.

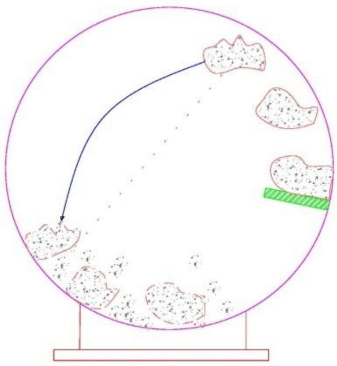

Figure 6 Illustration of Cantabro loss test

Figure 7 Shape of specimens in the Cantabro loss test

The initial weight of each specimen (M_1) was noted before placing it into the machine. The machine was then allowed to rotate at various revolution levels, such as 50, 100, 150, 200, 250 and 300 revolutions. At each revolution level, the abraded specimens were cleaned for any loose debris and weighed accurately to measure the M_2. Finally, the percentage loss was calculated using the following equation:

$$\text{Cantabro Loss, \%} = \frac{M1-M2}{M1} \times 100 \tag{3}$$

where M_1 is the initial weight of the test specimen, and M_2 is the final weight of the test specimen.

3. RESULTS AND DISCUSSION

3.1. Effect of the Bacterial Solution on the Compressive Strength of Bacterial Concrete

The effect of the bacterial solution on compressive strength is shown in Figure 8. The percentage increases in compressive strength for BC-5%, BC-10% and BC-15% were 8.98%, 17.02% and 4.65%, respectively at 28 days, which is very good when compared to the control mix concrete.

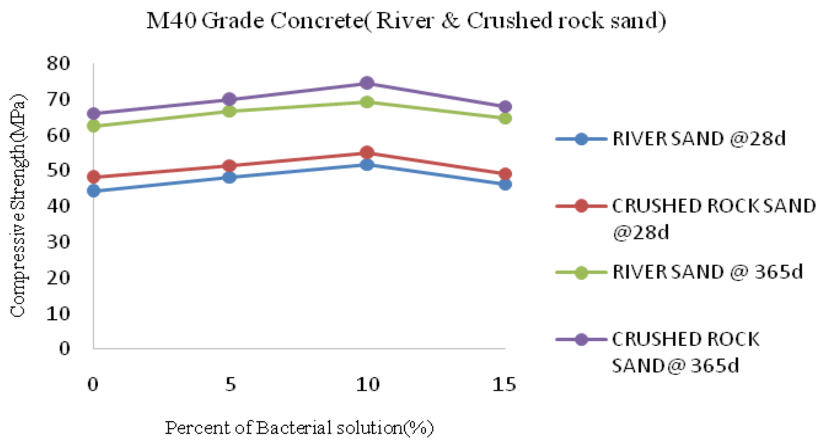

Figure 8 Effect of the bacterial solution on the compressive strength of bacterial concrete

At 180 days the increases were 6.86%, 12.54% and 4.63%, respectively, while at 365 days these percentages were 6.88%, 11.22% and 3.84%, respectively for BC-5%, BC-10% and BC-15%. The percentage increases in the compressive strength of the crushed rock sand mixes at 28 days for BC-5%, BC-10% and BC-15% were 6.94%, 14% and 2.28%, respectively, while at 180 days they were 8.72%, 14.81% and 5.31%, respectively. At 365 days these percentages were 5.65%, 12.68% and 3.11%, respectively for BC-5%, BC-10% and BC-15%. From these figures it can be noticed that the gain in compressive strength at 180 days and 365 days is higher than that at 28 days because the contribution of *Bacillus subtilis* bacteria together with calcium lactate to compressive strength is prominent at ages of more than 28 days for both river and crushed sand mixes. With the increase in the percentage of bacteria in the concrete ranging between 0% and 10% compressive strength also increased, but at 15% it decreased; this was because of the way that hydration products are saturated at 10% bacterial solution, so further increases in the solution do not contribute to strength, in fact reducing it. The crushed rock sand mixes showed more promising results than the river sand mixes due to the cubical sharp-edged faces of the grains in the rock.

3.2. Effect of the bacterial solution on the flexural strength of bacterial concrete

The effect of the bacterial solution on flexural strength is shown in Figure 9. The percentage increases in the flexural strength of the river sand mixes for BC-5%, BC-10% and BC-15% were 4.97%, 9.04% and 2.26% respectively at 28 days, while those at 180 days were 1.95%, 7.81% and 1.17%, respectively.

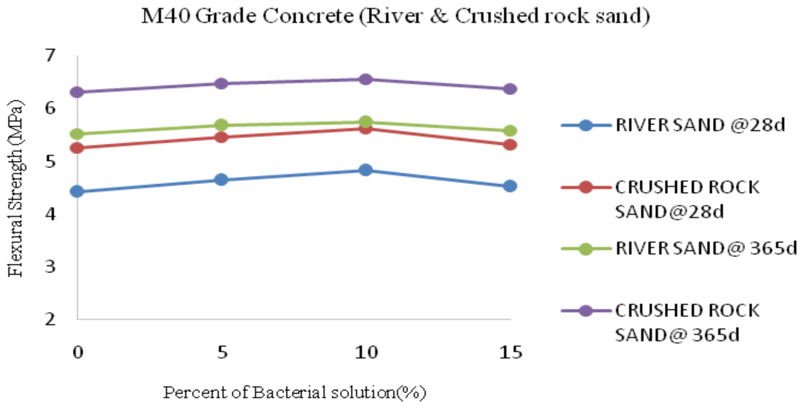

Figure 9 Effect of the bacterial solution on the flexural strength of bacterial concrete

At 365 days these percentages were 2.89%, 3.98% and 1.08%, respectively for BC-5%, BC-10% and BC-15%. In the crushed rock sand mixes, the percentage increases in flexural strength at 28 days for BC-5%, BC-10% and BC-15% were 3.8%, 6.84% and 1.14%, respectively, while at 180 days they were 1.99%, 5.64% and 0.66%, respectively. At 365 days these percentages were 2.53%, 3.79% and 0.94%, respectively for BC-5%, BC-10% and BC-15%. In addition, the percentage of bacterial concrete increased from 0% to 10% and the flexural strength also increased, but at 15% it decreased, for the same reason detailed in the previous section.

3.3. Effect of Crushed Rock Sand on Cantabro Loss of Bacterial Concrete

From the above test results, the Cantabro loss for the crushed rock mixes was good compared to the river sand mixes. The loss was highest at 10% bacterial solution in both the river sand and crushed rock mixes.

Table 3 Average Cantabro loss of bacterial concrete with river sand

Percentage of bacteria in concrete	Cantabro loss @3 days (%)	Cantabro loss @7days (%)	Cantabro loss @14 days (%)	Cantabro loss @28 days (%)
0	13.24	12.87	12.11	11.36
5	14.92	12.22	11.56	9.86
10	10.11	9.23	8.15	7.45
15	15.23	14.88	12.35	11.58

Table 4 Average Cantabro loss of bacterial concrete with crushed sand

Percentage of bacteria in concrete	Cantabro loss @3 days (%)	Cantabro loss @7days (%)	Cantabro loss @14 days (%)	Cantabro loss @28 days (%)
0	12.46	11.62	11.06	10.34
5	11.24	9.48	10.46	8.82
10	8.64	7.84	7.12	7.06
15	14.34	13.86	13.98	12.44

Figure 7 shows the specimens before and after Cantabro loss. In the machine the specimens collided with each other and also with the machine edges. This was used for measuring wear and tear action in pavements.

3.4. Scanning Electron Microscopy (SEM) Analysis

The SEM analysis images for control with a magnification of 10000 and 2500, sub acceleration voltage of 10000 volts, and pixel sizes of 9.921875 and 39.6875. Figure 9 shows the micro structure of the conventional concrete. The SEM analysis images for bacterial concrete with a magnification of 10000, 20000 and 2500, sub acceleration voltage 10000 volts, and pixel sizes of 9.921875, 4.960938 and 39.6875. Figure 10 shows the micro structure of the bacterial concrete, while the SEM analysis image shows the occurrence of calcite precipitation in the concrete. CSH, CH and pores were present in every sample and calcite was observed in the pores in the case of bacterial concrete (Santhosh et al., 2000). This clearly shows that the porosity decreased and gained in strength and that the cracks also healed due to calcite formation.

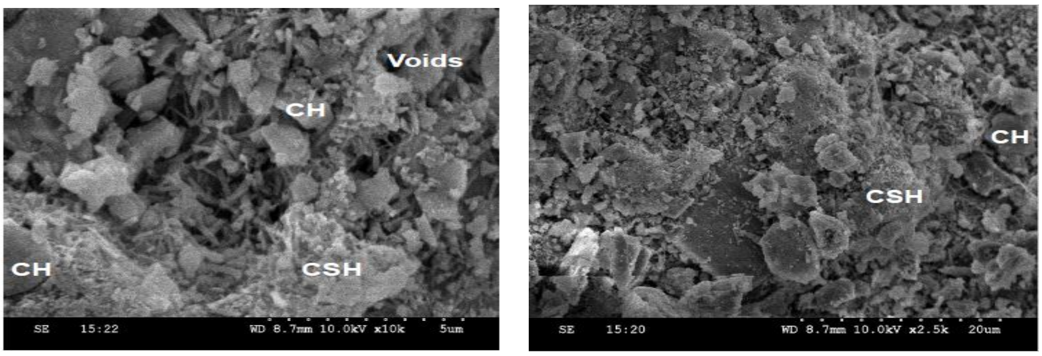

Figure 9 SEM micrograph of the control concrete

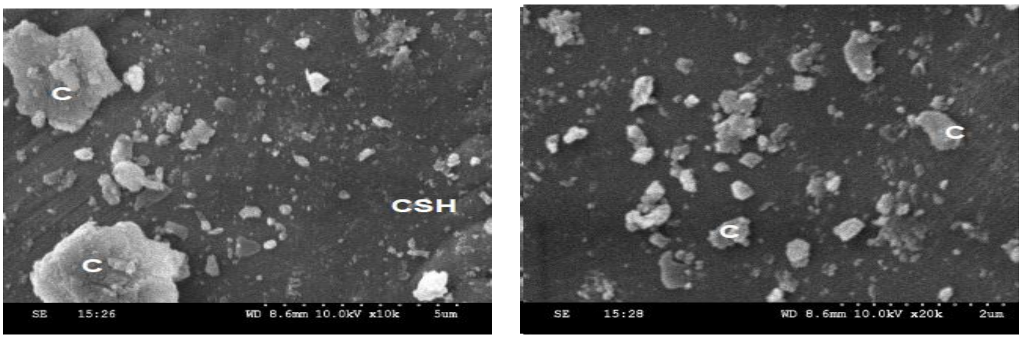

Figure 10 SEM micrograph of the bacterial concrete

3.5. Proposed Empirical Relation between the Compressive and Flexural Strength of Bacterial Concrete

Figures 11 and 12 show the relationship between the compressive and flexural strength of the bacterial concrete at different bacterial percentages (BC-5%, BC-10% and BC-15%) (Anbuvelan.et.al, 2014). From the experimental results, the empirical relation for the bacterial solution is proposed as follows:

$$f_t = 0.66\sqrt{f_{ck}} \tag{4}$$

$$f_t = 0.89 f_{ck}^{0.46} \tag{5}$$

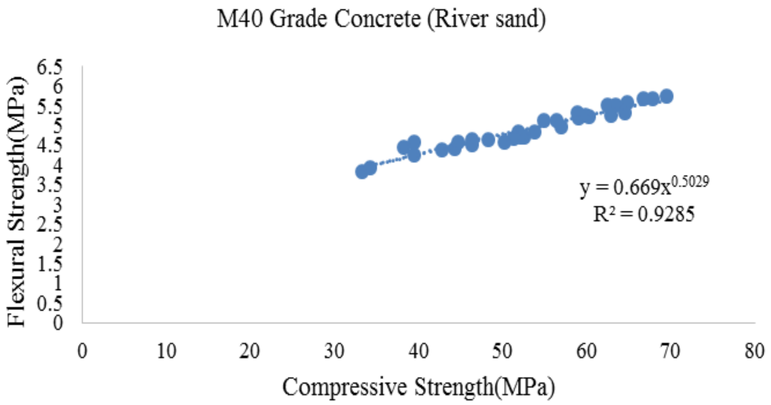

Figure 11 Empirical relation between compressive and flexural strength (river sand)

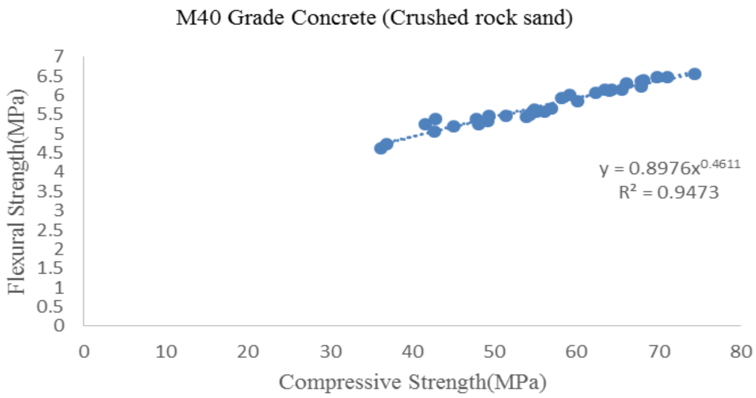

Figure 12 Empirical relation between compressive and flexural strength (crushed rock sand)

4. CONCLUSION

From the experimental work conducted on the bacterial concrete mixes, the following conclusions can be drawn: (1) The Cantabro loss, i.e. the abrasion resistance of bacterial concrete mixes, is strongly influenced the flexural strength; (2) the Cantabro loss is good at 10% bacteria in the crushed sand bacterial concrete mixes; (3) the flexural strength values increased and Cantabro loss decreased at up to 10% bacterial solution in the bacterial concrete mixes; (4) the addition of bacteria to concrete significantly improved the Cantabro loss and flexural strength at all ages; (5) the strength of the bacterial concrete for crushed rock sand mixes showed higher values than river sand mixes at all ages, irrespective of the percentage bacterial solution; (6) based on the test results, the optimum dosage of bacterial solution to improve strength at any age is 10% by weight of cement; an empirical relation exists between the compressive and flexural strength of bacterial concrete, which can be presented in the form $f_t = 0.66\sqrt{f_{ck}}$ for river mixes and $f_t = 0.89 f_{ck}{}^{0.46}$ for crushed rock sand; and (7) the SEM analysis showed that the presence of calcium carbonate in bacterial concrete.

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