



Experimental Investigation on Workability and Strength Characteristics of Bacteria Incorporated Self-consolidating Geopolymer Concrete

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Abstract Across world cement is utilized as one of the chief raw materials in construction sector. Increased production of cement to meet the demands from construction sector has resulted in emission of catastrophic amount of CO₂ gas into the atmosphere. Awareness about global warming has resulted in innovation of inorganic polymer binder termed as Geopolymer concrete (GPC). In recent times, priority is given to studies focusing on development of self-consolidating ability in GPC. Such concrete is being addressed by the term Self-consolidating Geopolymer Concrete (SCGC). This experimental investigation focuses on studying the influence of *Bacillus Subtilis* bacteria on workability and strength characteristics of alkali activated Self-consolidating concrete (SCC) mixes. Experimental studies were carried out on 10 different SCGC mixes. In each mix, bacterial cell concentration was varied in order of 0%, 1%, 2%, 3%, 4%, 5%, 6%, 7%, 8% and 9% of total binder content. Test values concluded that all fresh SCGC mixes with bacteria exhibited better workability as per EFNARC guidelines. Highest compressive strength of 36 MPa was attained by SCGC mix containing bacteria in cell concentration of 8% of total binder content. SEM analysis indicated a dense microstructure. XRD analysis indicated the formation of quartz, Illite and Mullite minerals along with geopolymeric gels. Formation of these minerals helps in development of dense microstructure.

Keywords Bacterial concrete; *Bacillus Subtilis*; Alkali Activated; Self-consolidating; Geopolymer concrete.

1. Introduction

In present era, construction sector across the world has witnessed massive growth with construction of infrastructural facilities like bridges, buildings and roads. Construction sector utilizes concrete and steel in huge quantity. Cement is one among the chief raw material used in manufacturing of concrete [1]. Predictions are being made that by 2030 around 4800 metric tons of cement will be

produced around the world. 2800 kg of natural resources are being utilized for every 1000 kg of cement production. Increased production of cement to meet requirements from construction sector has resulted in emission of catastrophic amount of CO₂ gas into atmosphere. This has increased air pollution. Many researches in concrete technology concentrating on eradicating ill effects from cement production is being conducted everywhere from the past 40 years. Invention of Geopolymer concrete (GPC) in 1978 by Prof. Joseph Davidovits is one such result of many researches which can reduce utilization of cement in construction sector [2]. GPC is an inorganic binder produced by chemically reacting aluminate and silicate bearing industrial residues such as Ground granulated blast.furnace slag (GGBFS), Silica.fumes (SF), Rice.husk ash (RHA), Fly ash (FA) and Metakaolin with a caustic activator namely sodium.silicate or potassium.silicate and potassium hydroxide or sodium.hydroxide. In comparison to Portland cement concrete (PCC), GPC has superior strength and durability characteristics [3]. Adopting GPC in construction sector can lead to reduction in environmental pollution since it minimizes dependency on Portland cement concrete (PCC). Initially GPC was manufactured by utilizing Class F FA. But GPC based on FA exhibited inferior strength development when subjected to ambient curing [4]. Because of high viscosity, good compaction of GPC when placed on formwork can be obtained only through vibration[5]. Self-consolidating geopolymer concrete (SCGC) is one such innovation of GPC which can prevent noise pollution and segregation of materials resulting from vibration. Workability of SCGC is very high in comparison to GPC. SCGC eliminates the necessity of manual compaction since it has the ability to flow under its own weight. In congested

reinforcement, SCGC has the ability to fill all the gaps in formwork [6]. Adopting SCGC in construction results in enhancement of quality and also drastically reduces the cost of maintenance over a span of years [7]. EFNARC guidelines are adopted for preparing mix design of SCGC [18]. Partial replacement of FA with GGBFS and Alccofine enhances strength characteristics of SCGC cured under ambient conditions. Alccofine 1203 also known as Ultra fine Slag are supplementary cementitious micro-fine particles manufactured under controlled environment to enhance performance characteristics in concrete. Bio-mineralization process in SCGC can be improved by partially replacing FA with alccofine and GGBFS [1,7]. Necessity of heat curing in SCGC can be eliminated by incorporation of GGBFS and alccofine. Because of high SiO₂ and CaO content in GGBFS and Alccofine, air voids are eliminated during the geopolymerization process which results in dense microstructure of geopolymer matrix [8]. Researches utilizing both GGBFS and alccofine for enhancing workability and strength characteristics of SCGC are very few. Deterioration of concrete can happen because of inherent development of cracks. Incorporation of Bacillus species bacteria into concrete can effectively seal cracks within the hardened concrete matrix without the necessity of external agents [9]. These bacillus species bacteria can survive under unfavorable environmental condition [10]. In this current study, investigations are made to determine the effect of Bacillus Subtilis bacteria on workability, strength and micro structural characteristics of SCGC. Researches carried out on Bacillus Subtilis incorporated SCGC are very few.

Sushree [1] conducted a study on SCGC made utilizing GGBFS and FA to determine the effect of Bacillus Licheniformis on performance of SCGC. They found that addition of bacteria had no adverse effect on workability and strength characteristics of SCGC. Pradip [2] conducted a study on GPC made utilizing fly ash [FA] and GGBFS to determine the effect of ambient curing on performance of GPC. They found that maximum percentage of replacing FA with GGBFS for achieving better workability and strength characteristics under ambient curing is 30%. Ramamohana [3] conducted a study on GGBFS and FA based GPC to determine the effect of Graphene oxide (GO) on strength characteristics of GPC. They found that addition of 3% GO with 30% GGBFS replacement enhanced compressive strength by 38.51%. Albitar [4] conducted a study to

determine feasibility of utilizing granulated lead smelter slag (GLSS), class-F FA (or) blended FA for enhancing durability characteristics of GPC. They found that durability characteristics of GPC enhances by replacing 60% FA with GLSS. Mehmet [5] conducted a study to determine the effect of adding steel fiber and nano silica on performance of SCGC based on FA and GGBFS. From results the authors concluded that combined utilization of steel fibers and nano silica reduced workability but improved bond strength and flexural characteristics of SCGC. Vishnu [6] conducted a study to determine the effect of Graphene oxide (GO) and wollastonite on workability and strength characteristics of SCGC based on FA and GGBFS. From results the authors concluded that addition of 10% wollastonite and 0.04% GO improved workability and strength characteristics of SCGC. Sherin and Riyadh [7] conducted a study to determine the effect of ambient curing on performance of SCGC. They found that it is possible to develop a M40 grade SCGC cured under ambient condition by partially replacing FA with micro FA and GGBFS. Nagaraj and Venkatesh [8] conducted a study to determine the effect of varying molarity of NaOH liquid and Na₂SiO₃/NaOH ratios on performance of SCGC cured in room temperature. They found that SCGC mix with 10M NaOH liquid and Na₂SiO₃/NaOH of 2.5 exhibited better workability and strength characteristics. Krishnapriya [9] conducted a study to determine the effect of bacteria isolated from alkaline soil samples on strength characteristics of M25 grade concrete. They found that addition of bacteria enhanced strength of concrete by filling cracks with calcite. Wulandari [10] conducted a study on GPC made utilizing FA to determine the effect of Bacillus Pasteurii bacteria on strength of GPC. They found that addition of bacteria enhanced compressive strength by 43.75%.

2. Materials and Methodology

2.1. Materials Used

In this experimental investigation all SCGC mixes were prepared by utilizing Gram Negative Bacillus Subtilis bacteria strain. The bacteria strain was supplied by National Centre for Microbial Resource (NCMR), Pune in a solution form and it was stored at -80⁰C in microbiology lab of M S Ramaiah University, Bangalore, Karnataka, India. SCGC mixes were prepared by utilizing GGBFS, Class F flyash,

Alccofine and alkaline activators such as Na_2SiO_3 and NaOH procured from suppliers at Bangalore. Coarse aggregates of size below 20mm and M-Sand conforming to IS 383:2016 [12] available locally were utilized in production of SCGC. BASF Masterglenium 8233 polycarboxylic ether based superplasticiser conforming to IS 9103 [11] was used to enhance workability of all SCGC mixes. Properties of materials utilized are indicated in tables 1-8.

Table 1. Bacillus Subtilis Bacterium properties (Data source: NCMR, Pune)

NCMR Accession Number	MCC 2183
Taxonomic Designation	Bacillus subtilis (Ehrenberg 1835) Cohn 1872
Strain Designation	LS-1
Source of Isolation	Lonar lake soil sample
Medium Name and Number	72a (Horikoshi and Akiba Agar (HAA)/Broth (HAB))
pH of Medium	12
Temperature of Growth in $^{\circ}\text{C}$	30
Incubation Period	1 d
Risk Group	--
Oxygen Requirement	Aerobic

Table 2. Physical Properties of GGBFS and Alccofine

Physical Requirements	Specific Gravity	Slag Activity Index in %		Fineness (m ² /kg)
		7 Days	28 Days	
GGBFS	2.9	72.4	382	110.4
Alccofine	2.91	73.3	383	114
Requirements as per IS 16715:2018	--	60 (Min)	320 (Min)	75 (Min)

Table 3. Properties of Na_2SiO_3 solution

Properties	Specifications
Solids to Liquid Ratio	48:52
Colour	Colourless
Density (g/cc)	2.7

Table 4. Test Results of Aggregates

Test Data	Fineness Modulus	Water Absorption (%)	Specific Gravity
Fine Aggregate	3.6	0.4	2.7
Coarse Aggregate	20 mm	6.3	2.8
	12.5 mm	2.8	1.5

Table 5. Properties of Superplasticiser

Test Data	pH	Relative Density	Aspect	Chloride ions
Superplasticiser	>6 at 250C	1.1 to 0.01 at 250C	Dark Brown Liquid	<0.18%

Table 6. Chemical Properties of Fly ash

Chemical Requirements	SiO ₂	Na ₂ O	MgO	SO ₃	SiO ₂ +Al ₂ O ₃ +Fe ₂ O ₃	Total Chlorides	Loss of Ignition	
Content in %	52.72	0.80	1.94	0.23	87.56	0.004	3.2	
Requirements as per IS 3812:2013								
Part 1	Siliceous Pulverised Fuel Ash in %	35.0	1.5	5.0	3.0	70.0	0.05	5.0
	Calcareous Pulverised Fuel Ash in %	25.0	1.5	5.0	3.0	50.0	0.05	5.0
Part 2	Siliceous Pulverised Fuel Ash in %	35.0	1.5	5.0	5.0	70.0	0.05	7.0
	Calcareous Pulverised Fuel Ash in %	25.0	1.5	5.0	5.0	50.0	0.05	7.0

Table 7. Chemical Properties of GGBFS and Alccofine

Sl No	Chemical Requirements	Requirements as per IS 16715:2018	Alccofine	GGBFS
1.	Al ₂ O ₃	--	18.13	17.68
2.	MgO	(Max) 17.0%	7.1	8.3
3.	CaO	--	36.59	38
4.	MnO	(Max) 5.5%	0.42	0.3
5.	SiO ₂	--	32.44	32.65
6.	SO ₃	(Max) 3.0%	0.4	0.1
7.	Loss of Ignition	(Max) 3.0%	0.3	0.2
8.	S (Sulphide Sulphur)	(Max) 2.0%	0.5	0.56
9.	Insoluble Residue	(Max) 3.0%		0.2
10.	Glass Content	(Min) 85%	88	98
11.	Moisture Content	(Max) 1.0%	0.2	0.2
12.	(CaO+MgO+Al ₂ O ₃)/SiO ₂	(Min) 1.0%	1.93	1.9
13.	Cl (Chloride)	(Max) 0.1%	0.02	0.007

Table 8. Physical Properties of Fly ash

Physical Requirements	Soundness	Lime Reactivity (N/mm ²)	Fineness (m ² /kg)	Specific Gravity	Residue on 45 Micron Sieve in %
Results	0.032	6.1	371	2.4	17.1
Requirements as per IS 3812:2013					
Part 1	0.8 (Min)	4.5 (Min)	320 (Min)	--	34.9 (Max)
Part 2	0.8 (Max)	--	200 (Min)	--	50 (Max)

2.2 Methodology

The Bacillus subtilis bacteria utilized in this study was cultured in microbiology lab of M S Ramaiah University of Applied Sciences, Bangalore in a Luria Bertani (LB) broth medium. The culturing LB broth medium was made up of Tryptone-10 g/l, yeast extract-5 g/l and NaCl- 5 g/l. The sterile LB broth medium containing bacteria was incubated for 24 hrs at 30⁰C with continuous shaking. After 24 hrs the growth of bacterial cells per ml of medium concentration was counted manually using Quadrant Plate method.

Cell concentration of Bacillus Subtilis bacteria in all SCGC mixes was varied in order of 0%, 1%, 2%, 3%, 4%,

5%, 6%, 7%, 8% and 9% of total binder content. Constant binder content of 450 kg/m³ was maintained in all SCGC mixes. The percentages of binder constituents such as GGBFS, Class F Fly ash and Alccofine were kept constant at 65%, 30% and 5% in all SCGC mixes. Ratio of Na₂SiO₃ to NaOH was maintained constant for all mixes at 2.5. 13M NaOH solution was utilized in all mixes. With respect to binder content 1% super plasticizer and 12% additional water was added to all mixes. All samples were subjected to ambient curing. Table 9 represents SCGC mix identification.

Table 9. SCGC mix identification

Mix Description	Mix ID									
	M0	M1	M2	M3	M4	M5	M6	M7	M8	M9
GGBFS (%)	65	65	65	65	65	65	65	65	65	65
Fly Ash (%)	30	30	30	30	30	30	30	30	30	30
Alccofine (%)	5	5	5	5	5	5	5	5	5	5
Bacteria Cell Concentration (%)	0	1	2	3	4	5	6	7	8	9

2.3 Mix Design

All self-consolidating geopolymer concrete (SCGC) mixes utilized in this experimental study are designed with reference to previous researches [1-6] since standard guidelines for designing SCGC is not yet available. Addition of superplasticiser and free water helps in achieving higher workability in SCGC. Initial slump flow test conducted helped in determining the percentage of free water to be added and marsh cone test helped in determining the percentage of superplasticiser to be added for achieving higher workability in SCGC. Figure 1 represents test results

of marsh cone test and Table 10 represents test result of slump flow test. Compression strength tests conducted on 7 days hardened concrete cubes helped in establishing percentages of binder constituents and molarity of NaOH liquid utilized in this study. Based on initial test results, mix proportion adopted in this study for preparing SCGC mixes was influenced from the study conducted by Sushree et al. (2020) and Nagaraj et al. (2018) [1,8]. All SCGC mix details are shown in table 11.

Table 10. Slump Flow Test Results

Mix ID	S0	S1	S2	S3	S4	S5	S6	S7
% of Free Water	0	2	4	6	8	10	12	14
Slump Flow (mm)	38	158	260	391	481	550	670	813

Marsh Cone Test Results

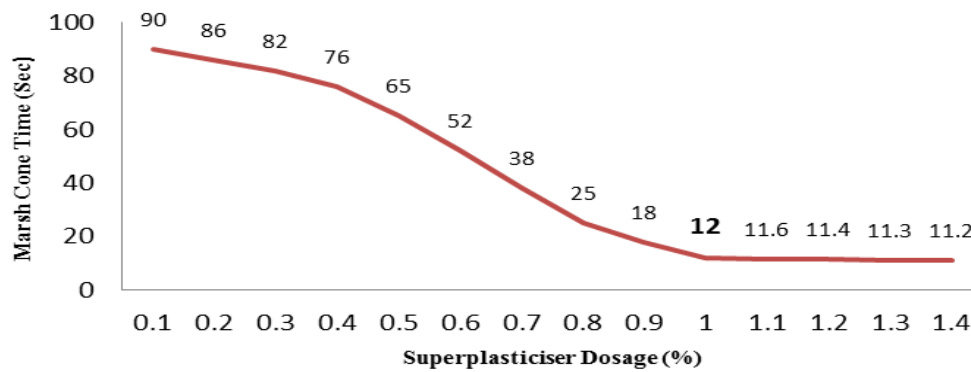


Figure 1: Marsh Cone Test Results

Table 11. Mix details of all SCGC mixes

Mix ID	Total Binder Content (kg/m ³)	Molarity	GGBFS (kg/m ³)	FA (kg/m ³)	Alccofine (kg/m ³)	Coarse Aggregate (kg/m ³)	Fine Aggregate (kg/m ³)	NaOH solids (kg/m ³)	Na ₂ SiO ₃ (kg/m ³)	SP (%)	Free water (%)
M0	450	13	292.5	135	22.5	925	925	63	157.5	1	12
M1	450	13	292.5	135	22.5	925	925	63	157.5	1	12
M2	450	13	292.5	135	22.5	925	925	63	157.5	1	12
M3	450	13	292.5	135	22.5	925	925	63	157.5	1	12
M4	450	13	292.5	135	22.5	925	925	63	157.5	1	12
M5	450	13	292.5	135	22.5	925	925	63	157.5	1	12
M6	450	13	292.5	135	22.5	925	925	63	157.5	1	12
M7	450	13	292.5	135	22.5	925	925	63	157.5	1	12
M8	450	13	292.5	135	22.5	925	925	63	157.5	1	12
M9	450	13	292.5	135	22.5	925	925	63	157.5	1	12

3. Test Results and Discussion

3.1 Test results of Fresh Concrete

As per EFNARC guidelines [14], segregation resistance, filling and passing ability defines self-compacting ability of Self-consolidating geopolymer concrete (SCGC). Higher workability without any change in strength properties is a fundamental requirement of SCGC [4]. Workability test results are as shown in Table 12. Test results indicate that addition of bacteria increases workability of SCGC. The slump flow of SCGC mix without bacteria was least because of increased polymeric reaction caused by higher molarity of NaOH solution and reduced water content resulting in accelerated setting and hardening time [7]. But in comparison, increase in water content resulting from addition of Luria Bertani (LB) broth medium containing bacteria increased filling and passing ability of SCGC mixes. The addition of LB broth medium increased setting and hardening time of SCGC mixes by reducing molarity of NaOH solution which resulted in delayed polymeric reaction

[8]. Excellent workability characteristics were exhibited by SCGC mix M8. SCC requirements as established by EFNARC guidelines was not satisfied by SCGC mixes M0 and M9.

Table 12. Workability Test Results

Mix ID	Test Parameters				
	Slump Flow (mm)	L-Box (mm)	T50 Slump Flow (sec)	J-Ring (mm)	V-Funnel (sec)
M0	642	0.67	5.4	14.6	13.2
M1	657	0.81	4.8	9.9	11.7
M2	670	0.83	4.6	9.2	9.9
M3	673	0.87	4.1	8.3	9.4
M4	681	0.91	3.8	6.6	9.2
M5	689	0.91	3.4	5.8	8.7
M6	693	0.93	2.8	4.2	8.5
M7	715	0.94	2.5	3.7	8.3
M8	738	0.96	2.1	2.2	8.2
M9	806	1.2	1.7	0.2	6.2
Acceptable values as per EFNARC Guidelines					
Minimum	650	0.8	2	0	8
Maximum	800	1	5	10	12

3.2 Hardened Concrete Test Results

Strength characteristics of hardened SCGC mixes were determined after 7, 14 and 28 days of ambient curing by

conducting compression, split-tensile and flexural tests as per IS 516-1959 [13]. Test results are as shown in Table 13. Figure 2, 3 and 4 graphically represents test results. Maximum compressive strength of 36 MPa, split-tensile strength of 3.2 MPa and flexural strength of 2.3 MPa after 28 days of ambient curing was achieved by SCGC mix M8 containing bacillus subtilis bacteria in cell concentration of 8% of total binder content. This is because of the formation

of C-A-S-H, C-S-H and N-A-S-H gels resulting from exothermic reaction due to high CaO content in GGBFS and Alccofine. The formation of these gels helps in development of tetrahedral alumina-silicate structure [6,7]. Least compressive strength of 19.5 MPa, split-tensile strength of 1.7 MPa and flexural strength of 1.3 MPa after 28 days of ambient curing was attained by SCGC mix M0 which did not contain bacteria.

Table 13. 7, 14 and 28 days Strength test Results

Mix ID		M0	M1	M2	M3	M4	M5	M6	M7	M8	M9
Compression Strength (MPa)	7 Days	14	17.5	18.8	19.2	20	22.9	21.5	24.5	27.9	23.4
	14 Days	18.6	19.8	20.6	21	24.7	25.6	28.4	30.8	34.6	29.8
	28 Days	19.5	21.2	22.4	24.6	28.2	30	31	34.6	36	33.4
Flexural Strength (MPa)	7 Days	0.3	0.7	0.9	1.2	1.4	1.5	1.6	1.6	1.8	1.6
	14 Days	1.1	1.3	1.4	1.5	1.5	1.6	1.7	1.7	1.9	1.7
	28 Days	1.3	1.4	1.6	1.9	1.9	2.0	2.0	2.1	2.3	2.1
Split-Tensile Strength (MPa)	7 Days	1.1	1.3	1.4	1.5	1.5	1.6	1.7	1.7	1.8	1.6
	14 Days	1.4	1.5	1.6	1.8	2	2.1	2.2	2.4	2.7	2.4
	28 Days	1.7	1.6	1.9	2.1	2.2	2.4	2.7	2.8	3.2	2.8

COMPRESSION STRENGTH TEST RESULTS

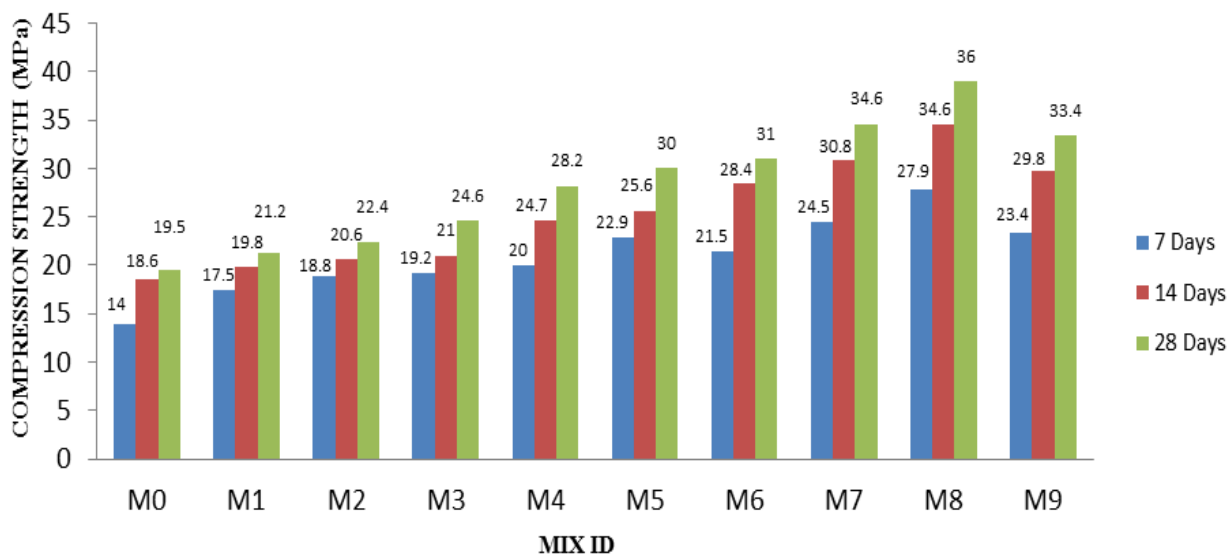


Figure 2: Compression Strength Test Results

FLEXURAL STRENGTH TEST RESULTS

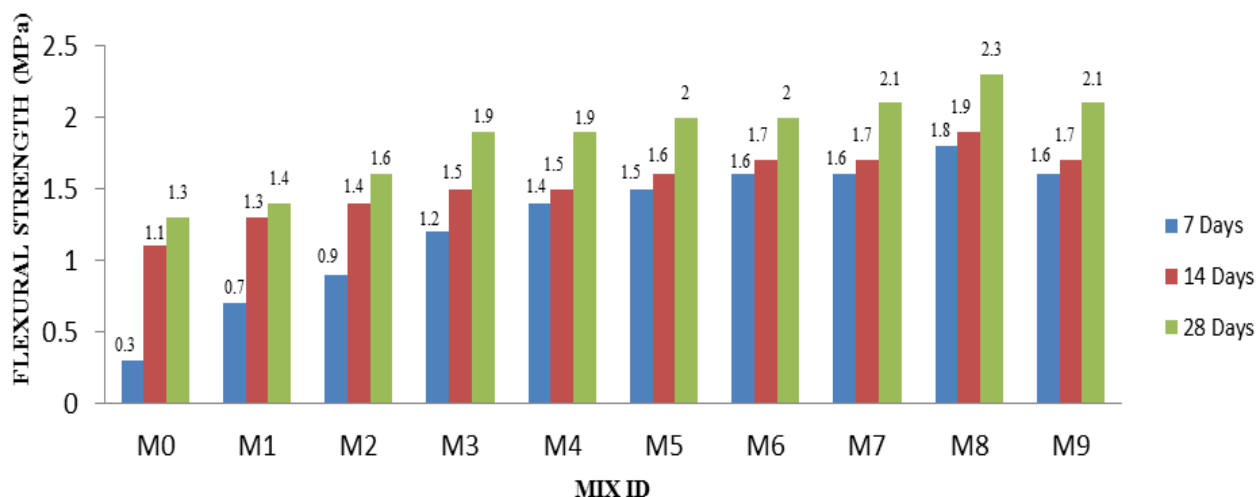


Figure 3: Flexural Strength Test Results

SPLIT-TENSILE STRENGTH TEST RESULTS

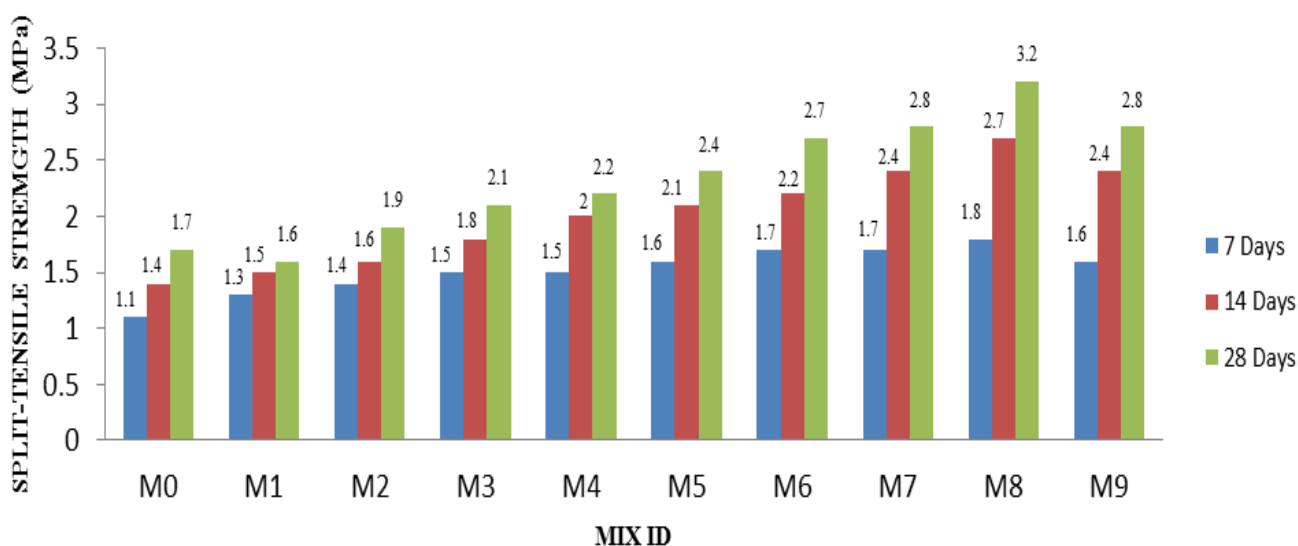


Figure 4: Split-Tensile Strength Test Results

3.2 Microstructure Analysis.

Scanning Electron Microscope (SEM) and X-ray Diffraction (XRD) analysis were conducted after 28 days of ambient curing on self-consolidating geopolymer (SCGC) mix M8 since it satisfied both workability and strength parameters to determine the influence of Bacillus Subtilis bacteria on microstructure of hardened SCGC. Figure 5

represents micrograph of SCGC mix M8. SEM analysis indicates a dense microstructure with uniform distribution of geopolymeric gels. Minor cracks and pores formed due to evaporation of excess water resulting from addition of LB broth medium is found to be filled by initial precipitation of calcite by Bacillus Subtilis bacteria [7]. High SiO₂ content resulting from partial replacement of fly ash by GGBFS and Alccofine contributed in formation of C-A-S-H gels which

also helps in filling up of micro cracks and pores [5]. Formation of dense microstructure with uniform distribution of C-S-H and C-A-S-H gels has contributed for better workability and strength characteristics of SCGC mix M8. XRD analysis helps in identifying chemical composition along with crystalline phases that contribute for development of microstructure. Figure 6 represents intensity vs Position

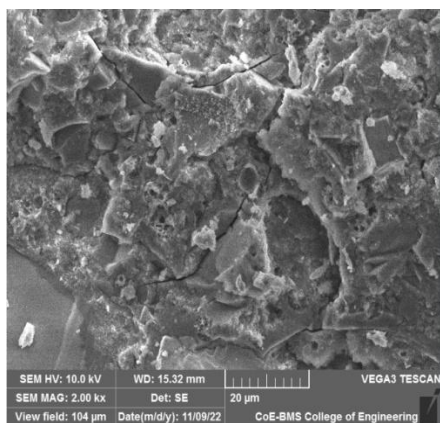


Figure 5: SEM Micrograph

Conclusion

This study experimentally investigates the effect of varying cell concentration of Bacillus Subtilis bacteria on workability and strength parameters of Self-consolidating geopolymer concrete (SCGC). Workability and strength characteristics of SCGC are found to be enhanced by addition of Bacillus Subtilis bacteria. This analysis shows that incorporation of Bacillus Subtilis bacteria in cell concentration of 8% of total binder content enhanced slump flow by 14.95%, compression strength by 84.65%, split-tensile strength by 88.23% and flexural strength by 63.63%. Incorporation of Bacillus Subtilis bacteria improved bio-mineralization process which is evident through SEM and XRD analysis. Formation of Quartz, Illite and Mullite minerals contributed for enhanced strength characteristics of hardened SCGC. By conducting few more researches concentrating on durability and crack healing aspects, bacteria incorporated SCGC can be proved as worthy eco-friendly alternative to conventional concrete in construction sector.

(in Degree) curve of SCGC mix M8. X-ray pattern indicates a typical amorphous structure of geopolymer concrete [3]. At 2θ broadband between 20° to 70° , minor peaks indicating crystalline phases of minerals like quartz, illite, mullite, along with C-A-S-H, C-S-H and N-A-S-H can be seen. Development of such geopolymeric gels along with quartz contributes to strength characteristics of SCGC mix M8.

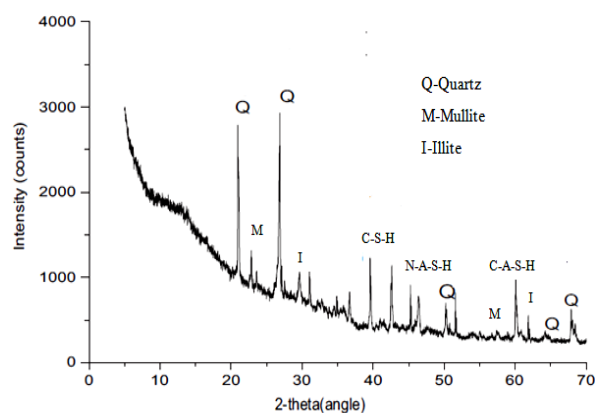


Figure 6: XRD Analysis

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COMPRESSION STRENGTH TEST RESULTS

