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Performance and Durability of High-Volume Fly Ash Concrete Incorporating *Bacillus Safensis* : A Comparative Study of Class C and Class F Fly Ash

Iqlima Nuril Amini[1], Dzikrie Fikriyan Syah[2], Davin H.E Setiamarga[3], Makno Basoeki[4], Luki Danardi[5], Irwanda Laory[6], Martin Anda[7], Mahendra Andiek Maulana[8], A.A. Ngurah Satria Damarnegara[8], Meity Wulandari[1], Januarti Jaya Ekaputri[8,9*]

[1]Department of Civil Engineering, Faculty of Engineering, Universitas Negeri Surabaya, Kampus UNESA Ketintang, Surabaya 60231, Indonesia
[2]PT. Freeport Indonesia, Papua, Indonesia
[3]Department of Applied Chemical and Biochemistry, National Institute of Technology, Wakayama College, Wakayama, Japan
[4]Bioconc Center Foundation, Sidoarjo, Indonesia
[5]PT.Waskita Karya (Persero) Tbk, Jakarta, Indonesia
[6]School of Engineering, University of Warwick, Coventry, United Kingdom
[7]Environmental Engineering, School of Engineering and Energy, College of Science, Technology, Engineering and Mathematics, Murdoch University, 90, South St, Murdoch WA 6150, Australia
[8]Department of Civil Engineering, Faculty of Civil, Planning and Geo Engineering, Institut Teknologi Sepuluh Nopember, Kampus ITS Sukolilo, Surabaya 60111, Indonesia
[9]Ash and Mud Resource Lab. Inc., General Research Building E202, Tokiwadai 79-5, Hodogaya-ku, Yokohama-shi, Japan 240-8501

** Corresponding author: januarti@ce.its.ac.id*

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Abstract: This study investigates the performance and durability of high-volume fly ash (HVFA) concrete enriched with *Bacillus safensis*, focusing on the comparative influence of Class C and Class F fly ash. Concrete mixtures were prepared with varying proportions of both fly ash types, with and without microbial addition, and tested for fresh and hardened properties including compressive strength, splitting tensile strength, porosity, and workability. Durability was further evaluated using the rapid chloride penetration test (RCPT) and accelerated corrosion test (ACT). Results showed that Class C fly ash, with its higher calcium content, produced a denser microstructure and improved early compressive strength. In contrast, Class F fly ash supported more favorable long-term microbial activity due to greater porosity and water availability. Incorporating *Bacillus safensis* enhanced compressive strength by up to 8% and significantly reduced chloride ion penetration, particularly in Class F fly ash concrete, through calcium carbonate precipitation within the pores. However, microbial addition was associated with reduced splitting tensile strength, likely due to differences in failure mechanisms. Long-term observations revealed strength gains of up to 13.3% after one year in microbial HVFA concrete. These findings demonstrate the synergistic contribution of *Bacillus safensis* and the effect of fly ash type to the improvement of sustainability and durability of HVFA concrete.

Keywords: *Bacillus safensis; Durability; High-Volume Fly Ash Concrete; Microbial-Induced Calcium Carbonate Precipitation; Responsible Consumption and Production*

1. Introduction

Indonesia is one of the global contributors to cement production, accounting for approximately 1.7% [1]. The demand for sustainable and environmentally friendly construction materials is increasing in line with global efforts to reduce carbon emissions from the construction industry. The production of Ordinary Portland Cement (OPC), as the primary binder in concrete, is known to contribute significantly to global CO_2 emissions [2]. One widely studied approach to reducing cement consumption is the utilization of supplementary cementitious materials (SCMs) such as fly ash, slag, and silica fume [3], [4]. High-volume fly ash (HVFA) concrete has been demonstrated to provide a lower carbon footprint, improved workability, and the potential for enhanced long-term durability [5]–[7]. However, the application of HVFA concrete often faces challenges due to variations in fly ash quality available in practice.

Indonesia hosts numerous coal-fired power plants (CFPPs) across different regions, generating fly ash with varying characteristics. According to ASTM C-618 [8], fly ash is generally classified into two main types Class C and Class F based on chemical composition. Both exhibit pozzolanic reactivity, but Class C fly ash also possesses cementitious properties due to its higher calcium content. Consequently, concrete incorporating these two types exhibits different performance characteristics. Class F fly ash is generally more advantageous for HVFA applications as it provides better durability and reduced shrinkage [9], [10] which explains its higher utilization compared to Class C fly ash.

Meanwhile, bio-concrete technology has emerged through the incorporation of microbes. Microorganisms act as biogenic agents capable of enhancing strength via biomineralization, primarily through the precipitation of calcium carbonate that fills pores and strengthens the concrete matrix [7], [11]. Muhammad et al. [7] introduced microbial consortia into fly ash concrete, demonstrating improvements in mechanical properties and reduced cement demand. Wulandari et al. [12] employed *Sporosarcina pasteurii* and *Rhizopus oligosporus* in geopolymer paste, showing significant contributions to closed porosity development. Achal et al. [13] also identified durability enhancement in fly ash concrete incorporating Bacillus megaterium, attributed to calcite precipitation that improved structural integrity, reduced water absorption, and decreased permeability.

The process of microbial-induced calcium carbonate precipitation (MICP) is influenced by the availability of calcium ions (Ca^{2+}) from cement, which react with carbonate ions produced by bacterial metabolism [14]. This highlights the critical role of calcium in $CaCO_3$ formation. Accordingly, differences in calcium content between fly ash classes where Class C contains higher CaO than Class F are expected to affect MICP performance. Class C fly ash may enable faster and greater calcite precipitation, while Class F may require additional calcium sources to optimize deposition. These compositional differences are believed to influence bacterial activity, matrix densification, and the durability of microbial HVFA concrete, although comparative studies remain limited.

One bacterial species successfully isolated from Natto is *Bacillus safensis* [15]. This ureolytic bacterium can survive under extreme conditions, including high salinity, alkaline pH, and radiation exposure, making it a promising candidate for application in the alkaline environment of concrete [16]. Previous studies have shown that *B. safensis* produces urease and protease enzymes, hydrolyzing urea into carbonate ions that bind with calcium ions to form calcium carbonate ($CaCO_3$) deposits [17]. These calcite precipitates fill pores and voids, thereby enhancing mechanical strength, density, reducing permeability, and lowering water absorption rates [18]. Moreover, *B. safensis* has been reported to heal cracks up to 0.888 mm wide through calcite-based self-healing mechanisms, thereby improving resistance to environmental attack and corrosion [19]. Such resilience and biomineralization capabilities demonstrate the high potential of *B. safensis* in HVFA concrete through MICP for enhancing strength and long-term durability.

The objective of this study is to evaluate and compare the performance and durability of HVFA concrete enriched with *Bacillus safensis*, focusing on the differences between Class C and Class F fly ash. The main parameters analyzed include compressive strength, water absorption, porosity, and resistance to chloride penetration, in order to understand the interaction between microbial activity and fly ash type. The outcomes are expected to contribute valuable insights toward the development of more resilient and environmentally sustainable microbial-based HVFA concrete.

2. Material & Methods

2.1. Material

In this research, coarse aggregate, sand, and ordinary Portland cement were supplied by PT. Waskita Beton Precast, while the fly ash was obtained from the Paiton power plant, Indonesia. The chemical composition of the fly ash was analyzed using X-Ray Fluorescence (XRF), and the results are presented in Table 1. The fly ash from this power plant was categorized as Class F (FAF) and Class C (FAC). The classification of fly ash is primarily determined by the type of coal used. Manz [20] stated that Class F fly ash is typically derived from bituminous coal, whereas Class C originates from sub-bituminous and lignite coal. In addition to differences in chemical composition and coal source, the two fly ash classes also exhibit distinct physical properties, as shown in Table 1., Table 2., and Table 3. The most notable differences are fineness and acidity (pH), both of which are strongly influenced by their chemical composition.

Table 1. Chemical composition of fly ash from Paiton Power Plant (% by mass)

Material	SiO_2	Al_2O_3	Fe_2O_3	CaO	Na_2O	K_2O	SO_3	Others
Fly Ash C (FAF)	32.78	15.09	11.98	23.26	3.82	1.22	1.36	9.76
Fly Ash F (FAC)	52.75	17.07	8.35	2.5	1.08	1.44	0.17	16.64

Table 2. ASTM C-618 requirement of Class C and Class F fly ash from Paiton Power Plant

Requirement	Fly Ash A (FA-C)	Fly Ash B (FA-F)
Total $SiO_2 + Al_2O_3 + Fe_2O_3$ (>50%)	59.85	78.17
CaO	23.26 (>18%)	2.5 (<18%)
SO_3 (Sulfur Trioxide) (<5%)	1.36	0.17
Class Classification	Class C	Class F

Table 3. Physical composition of fly ash from Paiton Power Plant

Parameter	FA-C	FA-F
Specific gravity (gr/cm^3)	2.83	2.86
Fineness (Retained 45 μm) %	8.8	14.5
pH	11.78	11.97

The bacterial strain employed in this study was *Bacillus safensis*, isolated from Natto, which is commercially available in Indonesia and designated as IDN1 [15]. The isolate was subsequently propagated at the Bioconc Foundation Center using suitable culture media prior to its incorporation into the concrete mixture. The preparation of microbial suspension, including the colony concentration, is presented in Figure 3.

The concrete mixtures in this study were divided into five variations, with varying fly ash classes, replacement percentages, and microbial incorporation. The mix proportions are presented in Table 4, designed in accordance with ACI 211 [21]. The notations F and C following the numbers indicate the fly ash class, while M denotes that the specimen incorporates bacteria. In addition to high-volume fly ash concrete, concrete with a 15% fly ash content (F15F) from class F was also used. This specimen served as a control system, representing the method applied by Waskita Beton Precast. All F50 and F60 mixtures incorporated NaOH with low molarity, with pH maintained below 12.5 to enhance the solubility of silica and alumina from fly ash [22], [23]. In addition, calcium carbonate was included to promote the hydration reaction of cement [24]. All specimens were prepared with a superplasticizer (Consol P102-HE) and a retarder (Consol SG) to improve workability and prolong the setting time.

Table 4. Concrete mix design (kg/m^3)

Code	F15F	F50F	F50FM	F60C	F60CM
Ordinary Portland Cement	404	237.5	237.5	190	190
FAC	-	-	-	285	285
FAF	71	237.5	237.5	-	-
Sand	820	809	809	760	760
Gravel	1071	1059	1059	1035	1035
Water	140	70	70	56	56
NaOH 0.03 M	-	-	-	84	84
NaOH 0.02 M	-	70	70	-	-
$CaCO_3$	-	24	24	25	25
Superplasticizer	2.85	1.9	1.9	1.9	1.9
Retarder	0.95	0.95	0.95	0.95	0.95
B. safensis (mL/m^3)	-	-	500	-	-
w/b	0.3	0.3	0.3	0.3	0.3

2.2. Methods

The mixing procedure in this study was divided into two approaches: for concrete with 15% fly ash (F15F), mixing followed ASTM C94, while for high-volume fly ash (HVFA) variations, the procedure was adapted from Romadhona et al. [25]. This approach aimed to achieve adequate workability despite the high fly ash content. The mixing sequence for HVFA specimens is illustrated in Figure 1.

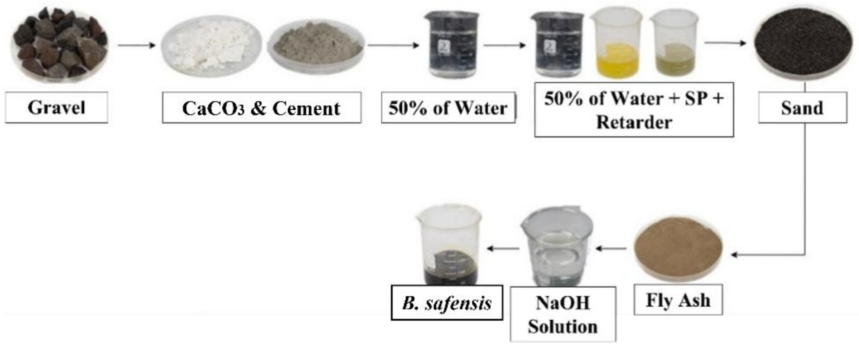

Fig 1. Mixing sequence of high volume fly ash specimens [25].

Specimens were cast in cylindrical molds with a diameter of 10 cm and a height of 20 cm. The workability of fresh concrete was assessed using the slump test with an Abram cone. All concretes were moist-cured at room temperature for 28 days. For the F15F specimens with a testing age of up to one year, specimens were subsequently stored under ambient air exposure until the testing time. In the hardened state, mechanical properties were evaluated based on compressive strength, splitting tensile strength, and porosity. Microbial activity was examined by quantifying viable microbial cells within the concrete specimens. Long-term performance was further assessed through compressive strength development. In addition, durability was evaluated under laboratory conditions using the Rapid Chloride Penetration Test (RCPT) and Accelerated Corrosion Test (ACT). In this study, the composition and test results of the F50 and F50FM variations were adopted from the previous research conducted by Syah et al [26].

3. Results and Discussion

3.1. Fresh Concrete Properties

Figure 2 presents the results of the slump test for fresh concrete. Among the four specimen mix compositions, the variations were based on fly ash class, replacement percentage, and microbial incorporation. The results indicate that, with different proportions and fly ash classes, the workability of Class C fly ash (FAC) at 60% replacement was higher compared to Class F fly ash (FAF). Based on the physical properties of the fly ash, this can be attributed to the finer particle size of FAC compared to FAF. Maeijer et al. [27] also reported similar findings, showing that finer fly ash particles enhance the workability of concrete. This improvement occurs because finer fly ash particles can increase inter-particle spacing (slurry effect) and reduce inter-granular friction (ball bearing effect) [28].

In addition, the incorporation of microbial solution into the fresh concrete mixtures further improved workability. This is attributed to the liquid form of the microbial solution, which increases the liquid-to-solid ratio of the mix. Junaidi et al. [29] also explained that the enhancement in workability is related to microbial CO_2 production, which generates micro-sized air bubbles that slightly improve the workability of fresh concrete.

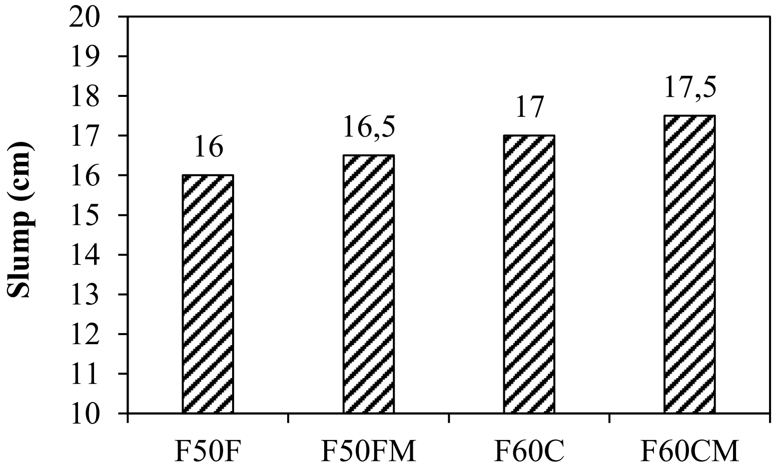

Fig 2. Slump test results

3.2. Total Microbial Count

The effectiveness of microbial performance in concrete mixtures is influenced by the survival and activity of viable microbes within the concrete. Figure 3 presents the results of viable bacterial colony counts over a 28-day period. In the F50FM mixture, the initial colony concentration introduced into the fresh concrete mixer was 508 CFU/g. Microbial growth was observed at one day after hardening, reaching 6400 CFU/g. However, as the concrete aged, the total microbial colony count decreased to 2473 CFU/g at 7 days and 2830 CFU/g at 28 days. In contrast, in the F60CM mixture, the microbial colony count consistently declined with age, reaching nearly zero at 28 days, with only 4 CFU/g remaining.

The type of fly ash had a significant impact on the survival of microbes within the mixtures. Class C fly ash, with its higher calcium content compared to Class F, produced a denser microstructure and higher early compressive strength (Fig. 4). The higher calcium content of Class C fly ash contributed to faster water binding due to its self-cementing properties [10], [30]. This led to quicker densification of the microstructure in the F60CM mixture, as evidenced by the reduced porosity (Fig. 6). The limited availability of water in the denser microstructure caused microbes entrapped within the concrete matrix to enter a dormant state [31]. Consequently, the total microbial colony count decreased with increasing concrete age.

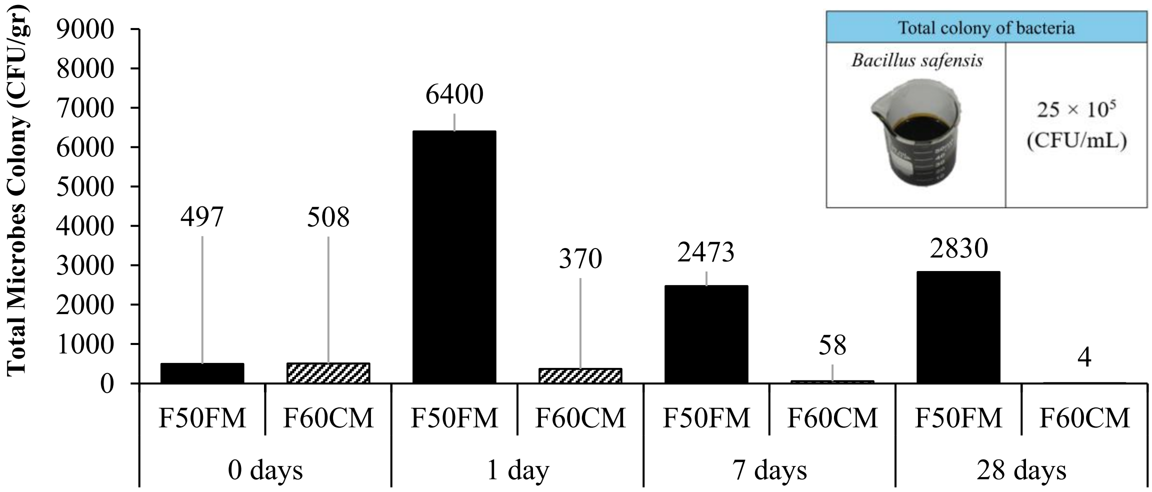

Fig 3. Total colony of microbes in concrete specimens

3.3. Mechanical Properties of Hardened Concrete

The identification of compressive strength is presented in Figure 4. The results indicate that at both 7 and 28 days, the compressive strength of HVFA concrete incorporating Class F and Class C fly ash exhibited relatively similar performance. The F60C mixture showed higher strength, by 3% at 7 days and 2% at 28 days. A similar trend was observed with the inclusion of bacteria. The addition of bacteria in the F50FM and F60CM mixtures increased compressive strength by approximately 8%. These findings suggest that microbial incorporation is effective in enhancing compressive strength for both Class F and Class C fly ash systems. The improvement can be attributed to enhanced workability, which promotes better solidification, as well as microbial activity that facilitates the precipitation of calcium carbonate from the reaction between calcium and carbonate ions produced by bacteria, thereby densifying the microstructure and improving compressive strength [32].

The densification caused by calcium carbonate precipitation was confirmed through open and closed porosity tests, as shown in Figure 6. The F60CM mixture exhibited lower total porosity compared to F50FM, primarily due to the reduction of open pores, indicating that microbial activity in conjunction with calcium effectively sealed capillary voids. The difference between Class C and Class F fly ash also influenced the resulting microstructure. Class C fly ash (F60C), with its higher calcium content and self-cementing properties, developed denser pores compared to Class F fly ash (F60F), which only exhibits slower pozzolanic reactions. Xu et al. [33] also noted that larger pore volumes promote microbial activity and calcium carbonate formation, whereas smaller pores restrict microbial habitats and limit mineralization capacity. Additionally, water availability plays a crucial role in bacterial viability. Class C fly ash has finer particles than Class F, leading to greater water absorption, which limits bacterial survival and reduces the efficiency of calcium carbonate precipitation. This observation is consistent with the bacterial viability data shown in Figure 3.

Although microbial incorporation proved effective in improving compressive strength of HVFA concrete, it exhibited a negative correlation with splitting tensile strength, as shown by the reduction in values. The use of microbes decreased splitting tensile strength by 16% in Class F fly ash concrete and 4.5% in Class C fly ash concrete. This phenomenon can be explained by the different failure mechanisms of the two tests. Mehta and Monteiro [34] emphasized that compressive strength is governed by overall matrix density, where microbial activity successfully filled macro-voids through calcium carbonate precipitation, significantly enhancing load-bearing capacity under compression. In contrast, splitting tensile strength is more sensitive to microscopic flaws such as microcracks and stress initiation points, which are not entirely repaired by microbial activity [35]. Moreover, the non-uniform or localized deposition of calcium carbonate crystals at the microscale may create new weak points susceptible to tensile stress [36], thereby reducing splitting tensile strength despite the overall improvements in porosity and compressive strength

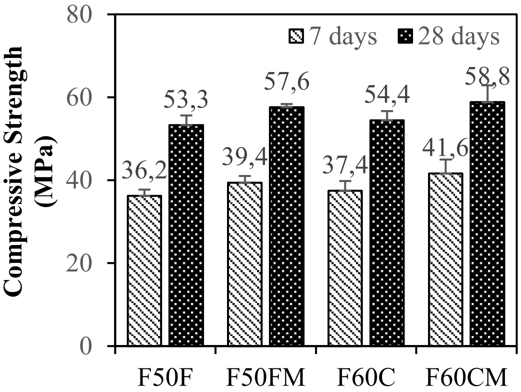

Fig 4. Compressive strength test

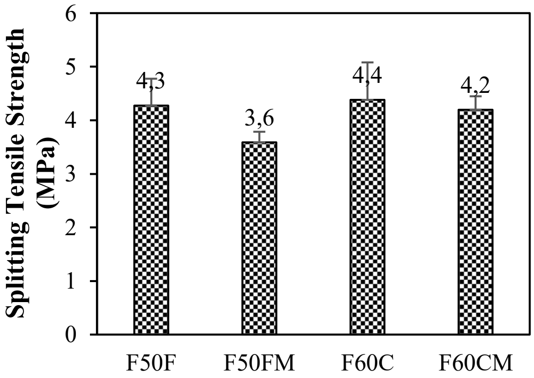

Fig 5. Splitting tensile strength test

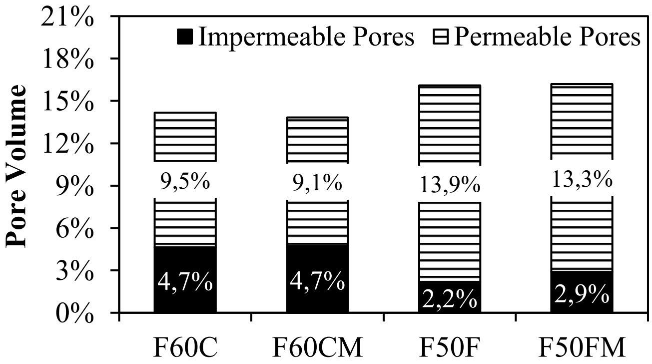

Fig 6. Porosity test

3.4. Long-Term Compressive Strength

The long-term performance of the F15F and F50M variations is shown in Figure 7. The results indicate that over a one-year observation period, there was a significant increase in compressive strength, reaching 4.6% in the F15 variation and 13.3% in the F50M variation. These findings suggest that strength development continues in the long term. At 28 days, despite the much higher proportion of fly ash, the compressive strength difference between F15F and F50FM was only 7.7%. This behavior can be attributed to the use of NaOH in the F50FM composition, which enhances the solubility and reactivity of fly ash, thereby accelerating the pozzolanic reaction [22], [23].

In the long-term, the compressive strength of F15F began to stabilize at 240 days, whereas the F50FM specimens continued to gain strength up to one year, eventually surpassing the strength of F15F. This phenomenon occurs due to the pozzolanic reaction of Class F fly ash, which consumes calcium hydroxide ($Ca(OH)_2$) to form C-S-H minerals that contribute to the improvement of concrete strength [37], [38]. Furthermore, the incorporation of *Bacillus safensis* in the F50FM mix plays a crucial role in sustaining long-term strength gain. The metabolic activity of the bacteria promotes the precipitation of calcium carbonate ($CaCO_3$) within pores and microcracks, effectively refining the pore structure and providing additional bonding over time [39], [40]. This biomineralization mechanism complements the pozzolanic reaction, leading to a denser and more durable microstructure that ensures continuous strength development even beyond the conventional curing period [40], [41].

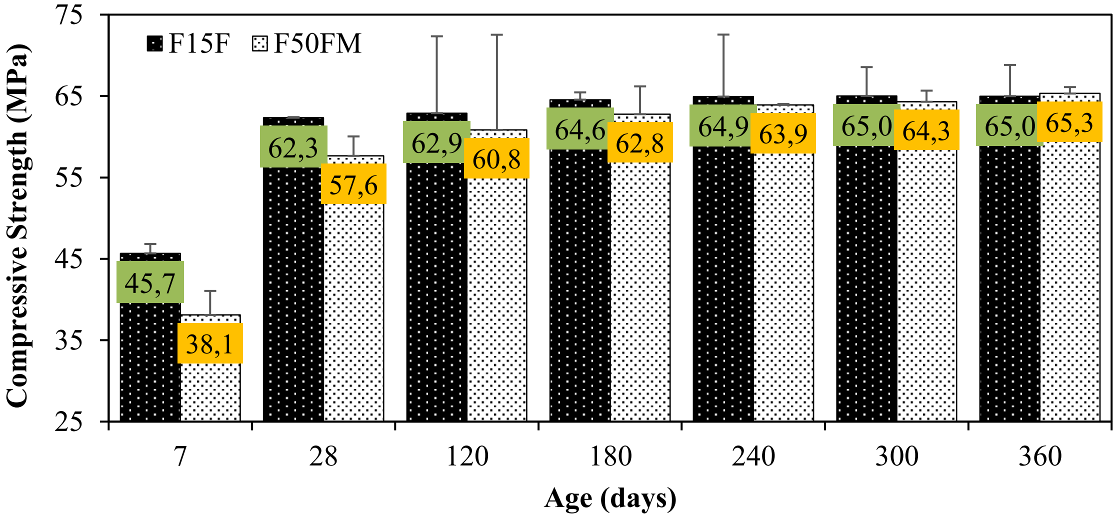

Fig 7. Long term compressive strength of low control system (F15F) and high volume fly ash concrete using microbes (F50FM)

3.5. Durability Test on Laboratory Scale

3.5.1. Rapid Chloride Penetration Test

Figure 8 presents the results of the Rapid Chloride Penetration Test (RCPT). The findings indicate that the utilization of Class C fly ash at 60% replacement (F60C) exhibited superior resistance to chloride ion penetration compared to Class F fly ash at 50% replacement (F50F). This outcome is consistent with the compressive strength and porosity results, which are influenced by the higher calcium content in Class C fly ash. The elevated calcium content accelerates the formation of additional hydration products (C-S-H), leading to a denser microstructure and improved chloride resistance, in contrast to the slower reactivity of Class F fly ash [42], [43].

The incorporation of microbes in both mixtures further contributed to reducing the charge passed. In the F60CM variation, the charge passed decreased by 12.2%, while in the F50FM variation it decreased by 48% compared to the mixtures without microbes. These results suggest that microbial activity in Class F fly ash provides highly effective resistance against chloride ion penetration. This reduction is consistent with several studies reporting that ureolytic bacteria are capable of precipitating $CaCO_3$ within pores and microcracks, thereby refining the pore structure and significantly impeding chloride ion ingress [44]–[46].

The differences in performance between Class C and Class F fly ash in the presence of microbes can be attributed to their distinct microstructural characteristics. Class C fly ash, with its self-cementing properties due to higher calcium content, produces a denser pore structure at an earlier stage. This condition limits the available pore space that can host bacterial growth and subsequent $CaCO_3$ precipitation, reducing the relative contribution of microbes to chloride resistance. In contrast, Class F fly ash develops a more porous microstructure in the early stages due to its slower pozzolanic reaction. This higher pore availability provides a more favorable environment for bacterial activity, allowing microbes to effectively deposit $CaCO_3$ within open pores and microvoids, which significantly enhances chloride resistance. Moreover, the improved pore refinement in Class F systems supplemented with microbes can be seen as a synergistic effect between the slower pozzolanic reaction and microbial mineralization, resulting in a more significant reduction in chloride penetrability.

Fig 8. Rapid chloride penetration test (RCPT)

Table 5. Chloride ion penetrability based on charge passed (ASTM C1202)

Charge Passed (Coulombs)	Chloride Ion Penetrability
>4000	High
2000-4000	Moderate
1000-2000	Low
100-1000	Very Low
<100	Negligible

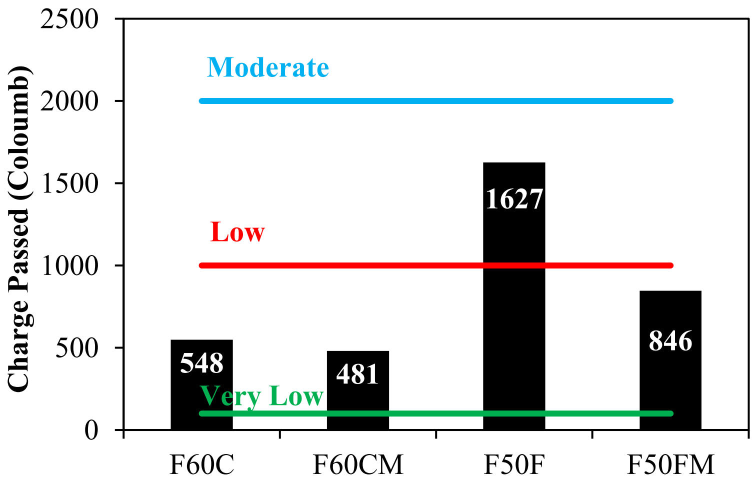

Fig 9. Results of the Rapid Chloride Penetration Test (RCPT)

3.5.2. Accelerated Corrosion Test

The accelerated corrosion test (ACT) was conducted to evaluate the rate of corrosion cracking in concrete exposed to chloride ions from aggressive environments, with the process being accelerated using an applied electrical current (Figure 10). The results of the ACT are presented in Figure 11. The longest time to crack was observed in the F60C mixture, with a crack initiation at 59 days, while the fastest cracking occurred in F60CM, at 42 days. The variation in fly ash type produced different cracking behaviors. The F60C mixture exhibited a longer time to crack compared to F50F. This is attributed to the denser microstructure of Class C fly ash, which makes chloride penetration more difficult. These findings are consistent with the free and bound chloride test results, which showed lower chloride concentrations in the F60C specimens (Figure 12). Class C fly ash is known to be more reactive, promoting higher formation of C-S-H gel and thus contributing to a denser microstructure [47].

In contrast, when microbial agents were incorporated, the F60CM mixture cracked faster than F50FM. Although the total chloride content penetrating F60CM was lower compared to F50FM, its free chloride content was significantly higher, reaching 40.3%. The lower chloride-binding capacity of F60CM resulted in faster corrosion initiation at the reinforcement, leading to earlier crack formation. Interestingly, despite having the densest microstructure among all specimens, F60CM exhibited faster cracking, as the mineral precipitates formed within the pores generated internal stress and microstructural fractures. These cracks facilitated water ingress, which could potentially activate dormant bacteria; however, within such a short timeframe, microbial activity was insufficient to heal the cracks, resulting instead in accelerated corrosion. The microcracks observed in the F60CM mixture can be attributed to autogenous shrinkage resulting from the low w/c ratio, in line with the findings of Ho and Huynh [48], which indicate that the water-to-binder ratio is a key factor influencing the extent of shrinkage in HVFA concrete. In addition, Min et al [49] reported that high-strength concrete with a low water content exhibits significant autogenous shrinkage, ultimately leading to the formation of early-age cracks in the cement paste.

These findings align with the fundamental mechanism of chloride binding in cementitious systems. Previous studies have distinguished free chloride as ions mobile within the pore solution, while bound chloride represents immobilized ions, either through chemical binding to form Friedel's salts or by physical adsorption onto C-S-H [50]. Such binding processes reduce the concentration of free chloride, which is the primary driver of reinforcement corrosion. Moreover, higher fly ash replacement levels generally enhance chloride binding capacity and improve overall resistance to chloride ingress [51], due to pore structure refinement and chemical immobilization mechanisms.

Fig 10. Accelerated corossion test (ACT)

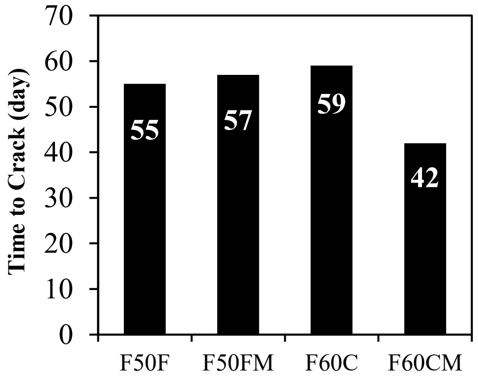

Fig 11. Results of the Accelerated Corrosion Test (ACT)

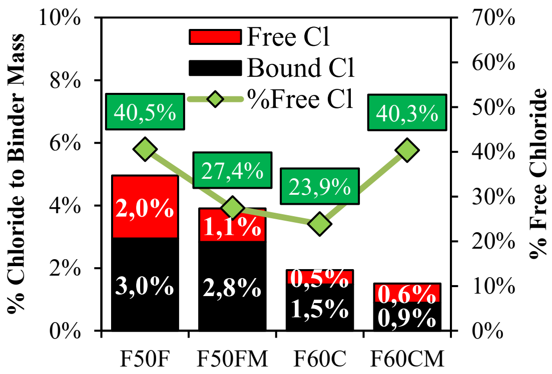

Fig 12. Free and Bound Chloride Content after Concrete Cracking

4. Conclusion

Based on the results of the study, the following conclusions are presented:

1. Class C fly ash, with its higher calcium content, provided superior early compressive strength and a denser microstructure, but the limited pore space and reduced water availability decreased microbial viability.
2. Class F fly ash, with its relatively higher porosity, supported microbial activity more effectively in the long term, thus enhancing calcium carbonate precipitation and improving durability.
3. The incorporation of *Bacillus safensis* increased compressive strength by up to 8% and significantly reduced chloride ion penetration, although it led to a reduction in splitting tensile strength.
4. Long-term observations indicated that HVFA concrete with microbial incorporation continued to gain strength for up to one year of curing.
5. In terms of durability, Class C fly ash exhibited better resistance against chloride penetration, while the addition of microbes in Class F fly ash was more effective in refining pore structures and mitigating aggressive ion migration.
6. The combination of class F fly ash and *Bacillus safensis* offers strong potential to improve the mechanical properties of microbial concrete and enhance its resistance to chloride attack, making class F fly ash more suitable for such applications.

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