



Review

A Review on the Performance Evaluation of Autonomous Self-Healing Bacterial Concrete: Mechanisms, Strength, Durability, and Microstructural Properties

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Abstract: The development of cracks, owing to a relatively lower tensile strength of concrete, diverse loading, and environmental factors driving the deterioration of structures, is an inescapable key concern for engineers. Reparation and maintenance operations are thus extremely important to prevent cracks from spreading and mitigating the lifetime of structures. However, ease of access to the cracked zone may be challenging, and it also needs funds and manual power. Hence, autonomous sealing of cracks employing microorganisms into the concrete sans manual intervention is a promising solution to the dilemma of the sustainable improvement of concrete. ‘Ureolytic bacteria’, key organism species in rumen-producing ‘urease’ enzymes such as *Bacillus pasteurii* or *subtilis*—when induced—are capable of producing calcium carbonate precipitations into the concrete. As their cell wall is anionic, CaCO₃ accumulation on their surface is extensive, and the whole cell, therefore, becomes crystalline and ultimately plugs pores and cracks. This natural induction technique is an environmentally friendly method that researchers are studying intensively. This manuscript reviews the application process of bacterial healing to manufacture autonomous self-healing bacterial concrete. Additionally, it provides a brief review of diverse attributes of this novel concrete which demonstrate the variations with the auto-addition of different bacteria, along with an evaluation of crack healing as a result of the addition of these bacteria directly into concrete or after encapsulation in a protective shell. Comparative assessment techniques for autonomous, bio-based self-healing are also discussed, accompanied by progress, potential, modes of application of this technique, and its resultant benefits in the context of strength and durability. Imperatives for quantitative sustainability assessment and industrial adoption are identified, along with the sealing of artificially cracked cement mortar with sand as a filling material in given spaces, as well as urea and CaCl₂ medium treatment with *Bacillus pasteurii* and *Sporosarcina* bacteria. The assessment of the impact on the compressive strength and rigidity of cement mortar cubes after the addition of bacteria into the mix is also considered. Scanning electron microscope (SEM) images on the function of bacteria in mineral precipitation that is microbiologically induced are also reviewed. Lastly, future research scope and present gaps are recognised and discussed.

Keywords: autonomous curing; cracks; strength; durability; sustainability; microstructure; self-healing; concrete; bacteria



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1. Introduction

Concrete is one of the world’s most critical, versatile, heterogeneous, and extensively used building materials [1–3]. The widely used application of concrete is next to bodies of

water, and its resulting requirements are gaining interest rapidly. The growing demand for buildings across the planet has accompanied the use of concrete since ancient Romans [4,5]. It is a human-built material that is prepared by blending OPC, fine aggregates, coarse aggregates, and water, with adequate proportions [6–10]. It is well known that concrete cracks are inevitable and one of its inherent faults, irrespective of its type, aggregates, or mixing design. This simply means a crack causing its degradation which ultimately drives structures to deteriorate since a network of cracks provides effortless access to moisture, water, unwelcomed acidic gases, chemicals, and other aggressive substances such as salts to percolate into the structure or degrade concrete chemistry, particularly in climatic conditions that further this process with high humidity and high precipitations. Thus, ultimately, all these factors together initiate the corrosion of reinforcement steel bars, reducing the life of structural concrete [11].

Consequently, the concrete's durability is compromised. Hence, a need has emerged to develop an inherently self-healing agent of natural biological origin with the capacity to remedy cracks and fissures in concrete. Rehabilitation and maintenance methods for infrastructure have also progressively been developed. Several repair methods are available to mend cracks; however, they are time intensive and costly. Execution of incessant check-ups and safeguarding may be challenging, particularly in the large-scale infrastructures due to massive funds needed to its realisation, along with certain other factors, such as the location of the structural damage, making the process of refurbishment more complex.

Conversely, bacterial concrete can successfully remedy concrete cracks. Thus, the concept of autonomous self-healing of such risky cracks with minimum resources in below-par structures has become a highly attractive area for researchers. Therefore, there is an urgent need to seek a sustainable, cost-effective, method of healing cracks that does not require manual intervention. To overcome these dilemmas, a natural, autonomous self-healing approach can be realised by the addition of a particular type of bacteria in concrete when mixed. *Bacillus subtilis* can cause some enzymes in 'ureolytic bacteria', including *Bacillus Pasteurii*, also known as 'urease', to be incorporated with the calcium nutrient source, giving rise to CCPs which seal the newly formed microcracks of concrete. The long-lasting hydration process makes this possible. Biomineralisation contributes to promising results for sealing microcracks in concrete structures. The strength and durability properties of structural concrete can be enhanced by this biotechnological means of CCP. The bacterial concentrations have been optimised for improved results for the remediation of concrete pores [12].

A review of the literature shows that the 'encapsulation technique' provides superior results, compared with the direct method of application, and shows that the application of bacteria can improve the strength, resistance, and durability of concrete [13]. Known aspects which determine the efficiency of autonomous bacterial self-healing include encapsulation of bioactive agents, capsule endurance during concrete mixing, influences on the aggregation of bioactive agents and ability to screen them, and the reactivation of mechanical and durable characteristics. Although it is more challenging to repair cracks of more than 0.8 mm, CCP can cure such cracks [14].

Lightweight structures in which lightweight aggregates are employed to replace fine aggregates are ideal settings for bacteria, which boosts the healing capacity, structural sustainability, and durability [15]. The incorporation in Rice husk ash concrete of bacteria can enhance the strength of concrete due to the precipitation of calcium carbonate in any age of concrete [16]. The maximum strength of 24% in the concrete of grade M50 with the highest CCP [17] can be increased. However, it also reduces porosity and permissibility which results in a maximum increase in compressive strength of 22% and a fourfold decrease in water absorption, as compared with normal concrete, by integrating *Sporosarcina pasteurii* bacteria [18]. Fungi is also be used to encourage calcium minerals to fill cracks in concrete as a self-healing agent by Luo et al. [19]. Similar outcome from the another study by Menon et al. [20] Araújo et al. [21] showed the resistance of polymerated healing agents against decay assessed with the urethane/poly(propylene glycol)-based precursor and

demonstrated greatest efficiency to resist degradation in the powerful alkaline setting characteristic for concrete.

A diverse calcium source might be used for CCP with the addition of bacteria to boost the durability properties of concrete by this quite recent advancement in biotechnology. Autonomous self-healing by microbe activity is relatively a novel concept that involves CCP in cracks through direct bioaction of calcium compound mineralisation in species such as *Bacillus subtilis* [22], or through the decomposition of urea with ureolytic bacteria viz. *Bacillus sphaericus* [23,24]. CCP by microorganisms is well matched with concrete, but the process is also eco-friendly [25] and user friendly, as *Bacillus sphaericus* does not harm humans [26].

Additionally, during the process, oxygen is consumed, reducing the probability of reinforcement corrosion due to oxidation. In addition, microbial CaCO_3 has the ability to block pores and bind grains to enhance strength and durability. Energy consumption and greenhouse gas (GHG) emissions resulting from microbial growth are negligible. The bacterial genre *Bacillus* can withstand highly alkaline medium and high humidity with an ability to develop spores that enable its use as an autonomous self-healing concrete agent. Therefore, in research investigations on CCP, *Bacillus* bacteria are almost universally employed as bioactive agents [22–24,27–30]. An appraisal of autonomous self-healing properties as a result of the bioactivity of active agents in concrete has been made with the addition of bacteria either directly to concrete or after encapsulation along with other substances [31].

Methodology

Web of Science, ScienceDirect, ResearchGate, SpringerLink, and other databases were used to search for novel self-healing composites. Keywords including ‘bacterial’, ‘self-healing’, and ‘CCP’ were identified in the Scopus database. The goal of this study was to highlight current research and application status in the domain of self-healing technology in concrete. The related references provided in the literature were also used. The structure of the manuscript is shown in Figure 1.

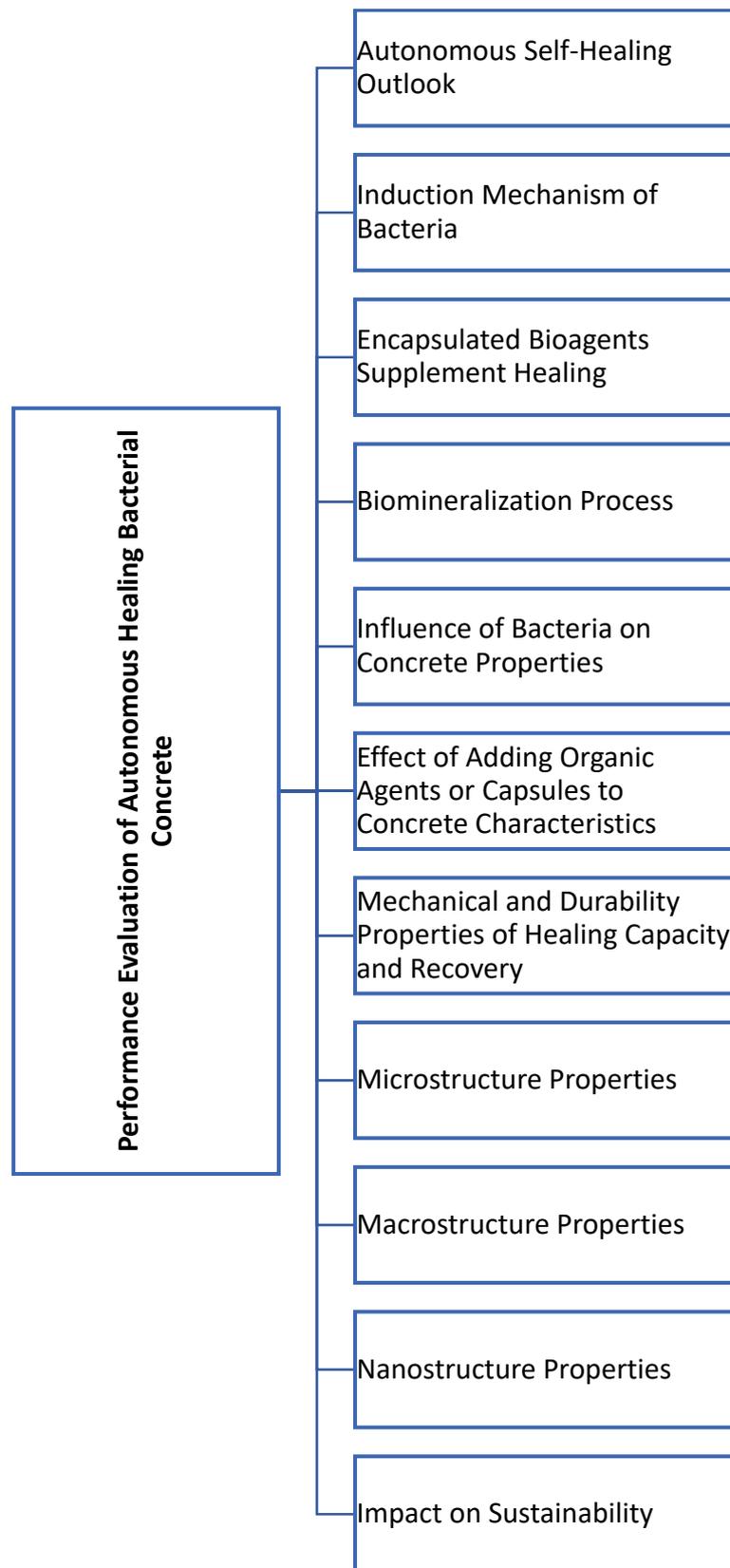


Figure 1. Structure of the manuscript.

2. Autonomous Self-Healing Outlook

Initially, an ideal autonomous self-healing technique should recognise the breaking or cracks in concrete that enable the release of bacteria. Autonomous self-healing practice is an effective approach to remedy microcracks in concrete and demonstrates good outcomes for concrete surfaces. Bacterial induction will form a pre-existing layer on cement cracks, as a result of CCP [25,27]. The induced bacteria are sufficiently able to withstand the alkaline medium present in concrete [32,33]. Under this highly alkaline environment, *Bacillus sphaericus* can precipitate CaCO_3 by converting urea into ammonium and carbonate [34].

CCP thus generated by metabolic activities of bacteria facilitate the sealing of microcracks and adhesion to sand, besides gravel in concrete [35], enhancing the durability of concrete. Splits widths of < 0.2 mm can be independently self-healed by concrete, while splits greater than this size cannot be healed by concrete alone [36]. In autonomous bacterial healing of concrete, initiation of any microcracks leads to activation of bacteria from their hibernation, i.e., inactive or dormant state. During the self-healing phase, calcium carbonate falls into cracks through metabolic operations of bacteria, which heal them. The formation of calcium carbonate has been identified as the principal contributor to healing surface cracks [37]. Once microcracks in concrete are filled using calcium carbonate precipitations, bacteria return to a hibernated state. If any subsequent cracks develop, bacteria are again reactivated to seal the newly formed cracks. This mechanism is conceived as the precipitation of MICP with the effect that bacteria act as perpetual healing agents. Several bacterial species can mediate calcium carbonate development in accordance with metabolic passageways, as shown in Table 1.

Table 1. Type of bacterial species used in some of the past studies.

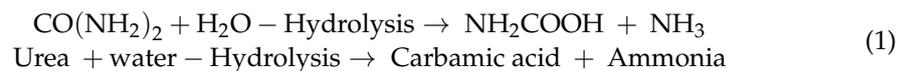
Bacterial Species Used	References
<i>Bacillus</i>	[38,39]
<i>Bacillus cereus</i>	[11,40]
<i>L. sphaericus</i>	[41]
<i>Sporosarcinapasteurii</i>	[42]
<i>B. pasteurii</i>	[43,44]
<i>K. flava</i>	[45]
<i>B. megaterium</i>	[46,47]
<i>Halomonas</i> sp.	[48–50]
<i>Micrococcus</i> sp. <i>Bacillus subtilis</i>	[51]
<i>Bacillus subtilis</i>	[52]
<i>Myxococcusxanthus</i>	[53]
<i>Bacillus sphaericus</i>	[54–57]
<i>Bacillus Sphaericus</i>	[14,15,58]
<i>B. sphaericus</i>	[23,24,34,57]
<i>Mytiluscalifornianus</i> shell extracts	[59,60]
<i>Microbiota</i> inhabiting the stone	[61]
<i>Shewanella</i>	[62]
<i>Bacillus pseudofirmus</i>	[63]
<i>B. pseudofirmus</i>	[63]
<i>Bacillus cohnii</i>	[22,63,64]
Spore forming bacteria (species not mentioned)	[14]
<i>S. pasteurii</i>	[44]
<i>B. alkalinitrilicus</i>	[65]
<i>S. pasteurii</i> (encapsulated)	[66]
<i>Bacillus</i> sp. CT-5	[67]
<i>Bacillus megaterium</i>	[17]
<i>Bacillus subtilis</i>	[52]
<i>Bacillus aerius</i>	[16]
<i>Sporosarcinapasteurii</i>	[18]
AKKR5	[68]
<i>Shewanella</i> Species	[32,62]

In comparison with autotrophic courses, CCP is more abundant in heterotrophic processes. MICP is a mineral layer that covers the cells of the bacteria [69,70]. Using the urease enzyme is the most frequently investigated decomposition of urea in bacteria for engineering purposes. Urease is an enzyme produced by bacterial metabolism, causing carbonate urea and ammonium to catalyse in a bacterial medium to increase the pH and the carbonate concentration. Hammes and Verstraete [58] suggested that CCP is significantly influenced by (i) calcium ion concentrations; (ii) pH solution; (iii) inorganically dissolved carbon concentration; (iv) nucleation site accessibility. The first three conditions concern the concrete matrix, while the bacterial cell provides the last factor. CCP can be obtained through bacteria in a variety of ways, including conversion to calcium compounds such as Ca lactate or urea hydrolysis through bacterial metabolism. As to the first mechanism, cracks make it possible for oxygen to enter the concrete, while bacteria convert calcium lactate to CaCO_3 plus CO_2 across the cracked areas. The nearby reaction of portlandite particles with CO_2 produces more CaCO_3 than is needed for cure. Consequently, when non-hydrated calcium hydroxide particles are still present, this mechanism is advantageous for fresh concrete.

Ureolysis or Urea Degradation

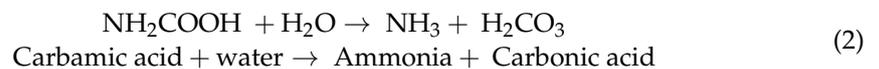
The urease microbial enzyme catalyses urea hydrolysis to ammonium and carbonate [71]. An intra-cell of 1 mol of ammonia and 1 mole of carbamic acid is hydrolysed into one mole of urea (1) [34].

In the presence of microbial urease, the reaction occurs as follows:

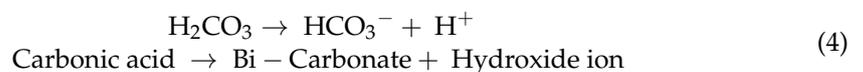
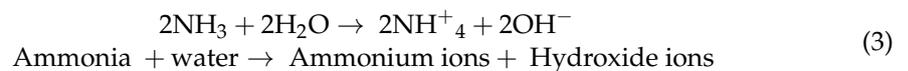


The reaction rate (1), i.e., carbamic acid and ammonia hydrolyses impulsively into an additional 1 mole of ammonia and carbonic acid (2) [72].

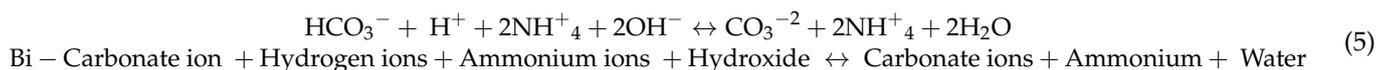
The reaction will occur as follows:



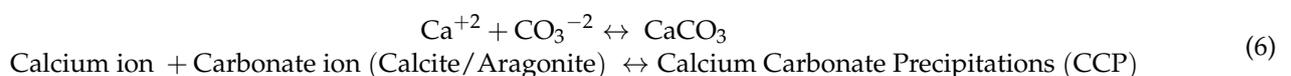
Bicarbonates and the 2 mol of ammonium and hydroxide ions with water (3) and (4) are generated by ammonium and carbonic acid. The reactions of both (3) and (4) are reversible.



The output of hydroxide ions increases the pH so that the bicarbonate balance may be changed to produce carbonate ions (5).

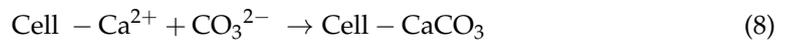


When calcium ions are present as calcium crystals carbonate (6), the production of carbonate ions precipitates.



Further enhancing the resemblance of the bacteria to the earth's surface is by the production of a monolayer of calcite, thus forming several layers of calcite.

The cell wall of bacteria is found to be negatively charged; the bacteria receive cations from the atmosphere, with Ca^{2+} ions, to deposition their cell surface. The Ca^{2+} ions react to CO_2 and the development of CCP on the cell surface, the nucleation site.



A number of microorganisms are able to use urealysin to precipitate calcium carbonate.

In short, autonomous bacterial self-healing may, therefore, be achieved through any of those mechanisms. However, several factors—namely, access to water, the breadth or area of crack to be healed, the time of concrete healing, and the duration of bacteria, are also essential for effective biobased healing of concrete.

Some definite applications of bacteria were found in a literature review. Incorporated into the rice husk ash, *Bacillus aerius* bacteria improved its durability [16]. *Bacillus megaterium* species have been added to cement and results indicate an augment in compression strength of 24% [17]. *Bacillus sphaericus* amplifies the durability of the concrete by the deposition of calcium carbonate [73]. The *Sporosarcina pasteurii* bacteria used in concrete incorporated with fly ash is also found to increase in strength and durability through autonomous self-healing [18]. The *Sporosarcina pasteurii* bacteria, when incorporated with silica fume concrete, it has shown an increase in both strength and durability due to the effect of autonomous self-healing [18]. *Bacillus sphaericus* bacteria has been induced in control OPC concrete to examine the treatment of the surface, and the outcomes reveal that bacterial CCP can be applied as an alternative treatment for concrete surfaces [54].

3. Induction Mechanism of Bacteria

According to the literature, two techniques can be used to induce the healing bacteria in concrete. Former research shows that the concrete containing graphite nanoplatelets (GNPs), in the case of lightweight aggregates (LWAs), has proved to be an appropriate carrier compound for bacteria where the bacteria were directly applied. The use of a healing agent to find the optimal application of bacteria for concrete strength has been recorded as 30×10^5 cfu/mL through the direct method [17]. The impregnation and subsequent encapsulation of lightweight aggregates in a polymer-coating layer are necessary to improve the overall performance of autonomous self-healing concrete [54].

Figure 2 shows the autonomous approach of self-healing through the use of microencapsulating techniques that integrate healing materials for autonomous material self-healing. No sooner had the crack ruptured the embedded microcapsules than the healing agent came into contact with the crack faces with capillary motion. The healing agent then combines with the built-in catalyst and activates polymerisation, protecting crack sealing. Conceptual evidence found by Kanellopoulos et al. [74] as revealed that the microcapsules survive and operate effectively in the case of cement-based matrix exposure.

The autonomous self-healing agents through encapsulation are sufficiently able to provide superior autonomous self-healing in the context of wider cracks and concrete structures with prior exposure to cracking in the concrete matrix [75,76]. The technique of hydrogel encapsulation was used, and bacterial spores embedded in samples of a hydrogel included had an increased self-healing efficiency, both concerning precipitation quantities and crack healing [73]. The method for direct application in the concrete of *Shewanella* bacteria was examined, and 25 percent augment in the compressive strength of cement mortar was monitored in 28 days [62]. With regard to the literature, the encapsulation technique has provided positive results, which are due to homogenous distribution and fortification of bacteria in the alkaline medium, for autonomous self-healing efficiency for crack sealing, and the amount of CCM.

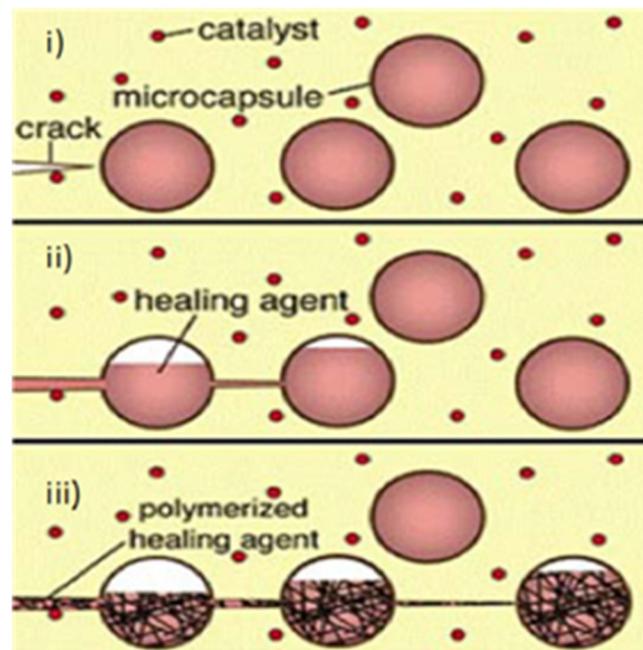


Figure 2. The concept of autonomous cure [76].

3.1. Cure by Direct Method of Application of Bioagents

Jonkers and Schalngen [77] examined the viability of the use of autonomous self-healing concrete by using a bacteria spore. *Bacilluscohnii*, *Bacillus halodurans*, and *Bacillus pseudofirmus* have been examined in three species of bacteria mixed in cement stone. Using yeast exhaust and peptone-based medium, cement stone chips were cured, and compressive and tensile strength were then studied. There was a noteworthy negative variation among bacterial and control specimens. SEM images showed CCP crystals subsequent to 12 days of incubation.

Nevertheless, the initial research was conducted externally to provide organic carbon sources, or bacteria food, and after specimens were cured in the medium, germination was monitored. In subsequent investigations, Jonkers et al. [27] conducted experiments to study the potential of CCP by using species of bacteria that can stay viable while being directly added to concrete, as well as to the yeast extract and peptone, along with mineral precursor compound, which was shown to have a potential impact. Two species of spores—*Bacillus pseudofirmus* and *Bacilluscohnii*—forming alkaliphilic bacteria have been studied. The mineral precursor compound was investigated for calcium acetate and calcium lactate. Calcium lactate was the only compound monitored, which did not influence the strength and resulted in a minor increase in strength. CCP crystals of size 20 to 80 μm were monitored on samples, but the CCP was found only in 7-day specimens and not in 28-day specimens. The alkaline environment with concrete and a decreased pore size may result in a decrease in feasible spores of bacteria, a similar result to that supported by Luo et al. [14].

This mechanism is argued to be superior to an enzyme-based approach based on ureases because no surplus ammonia is generated by an additional metabolic process that may affect the concrete matrix and lead to enhanced corrosion. In the case of alkaline resistance spore-forming bacteria, Luo et al. [14] investigated the effect of width and age of the crack, with healing conditions, induced directly to concrete along with substratum. Crack widths of 0.1–0.5 mm were induced, and three diverse conditions of incubation were undertaken at a constant temperature of 25 °C, including wet and moist curing and wet and dry cycles. Healing was measured at different ages of concrete (7, 14, 28, 60, and 90 days) by area repair rate. Beneath immersion in water, cracks with a width of up to 0.3 mm were completely healed, and the healing ratio of cracks of 0.1 to 0.3 mm was approximately 85%. The moist curing method and wet–dry cycles showed the highest cure efficiency in terms of cure circumstances, even though the repair speed for a wet–dry cycle was slower. The

survival rate of bacteria was low in the case of protective shell-less bacteria, and a reduction in concrete porosity resulted in a shorter transport distance to the mineral, and therefore, fresh cracks were patched up more powerfully than in later stages of aging [14].

3.2. Encapsulated Bioagents' Supplemental Healing

An autonomous, well-organised self-healing is highlighted, while the seal has long-lasting efficiency and can be achieved throughout the life cycle of a concrete structure. Therefore, the survival of bacteria is an essential factor. Nevertheless, while bioagents are immediately added to concrete, many challenges may still exist in their path to survival.

Jonkers [22] reported that although bacteria spores are supplemented, the life period of unshielded spores is restricted to merely two months; as a result, effectual autonomous self-healing is monitored merely in specimens at an early age. There may be several factors causing this fact, which include the alkaline nature of the matrix of cement, concrete mixing, and cement hydration. Activity may be considerably reduced while spores are subjected to a highly alkaline medium over a long period. Furthermore, some spores may be crushed in the course of mixing owing to the force of mixing or due to aggregate impact. Cement hydration decreases the pore porosity of the matrix over time, to as much as 0.5 μm , while typically, bacterial cells have larger sizes [23]. Hence, cell germination may be radically mitigated consequent to the shrinking of pores or bunged at a late stage of a concrete structure. To overcome this restraint, encapsulation of the bacteria can be made to protect them from the impacts of any of the influential characteristics of concrete and precipitations of carbonate through bacteria. Pole-apart techniques for encapsulation to guard bacteria were utilised in different materials such as expanded clay aggregate [22,65], silica gel and glass tubes with polyurethane (PU), diatomaceous earth, hydrogel [78], and microcapsules based on melamine. Although the insertion of capsules reduces mechanical strength, encapsulation-based self-healing agents have various benefits, including reduced fracture breadth and depth, reduced permeability, and efficient strength recovery after healing [79]. The urea hydrolysis process has been used in the majority of field-scale applications, for bacteria-based self-healing concrete as well as to improve geotechnical quality and stability, which include features such as soil erosion management, fracture sealing below ground surface, and, more recently, for improving wellbore cement [80]. The healing ratio for mortar specimens implanted inside a non-cohesive soil has been published by Esaker et al. [81] under varying pH and humidity soil conditions. Healing ratios in the range of 47–83 percent were recorded for mortar specimens with encapsulated microorganisms. Under the same settings, mortars that did not include encapsulated bacteria had significantly poorer healing rates.

The encapsulation of bacteria may be achieved through various techniques, which are discussed in the next sections.

3.2.1. In Polymeric Microcapsules

The polymeric microcapsules based on melamine were employed by Wang et al. [24] for encapsulation of spores of *Bacillus sphaericus*. In this case, a mineral precursor used was calcium nitrate that supplemented the concrete in the course of mixing with other nutrients such as yeast extract and urea. Bacterial healing is assessed based on the ratio of the healed crack region to zones of early cracking. Encapsulated spores enclosing concrete have demonstrated a healing ratio between 48% and 80%, while for samples devoid of spores, the ratio of healing was constrained to merely 50%. The utmost mitigation in the crack region was monitored, while samples underwent wet water and dry cycles using a medium hydrated by water. The wet water cycle was significantly long, i.e., approximately 16 h, which might be problematic to achieve the common underlying states with no manual intervention. The largest crack width cured was roughly 970 μm . The highest dose of a capsule in the context of water permeability regeneration and crack sealing was 3%; however, in the case of a dose with 5 percent, less variation was found. Nevertheless, it

may be articulated that the dosage of 3% was the optimal one. The decline in concrete strength was greater in the context of 5% dosage in comparison with the dosage of 3%.

3.2.2. An Additive of Special Cement

Hydrogel application for bacterial encapsulation is a newly found endeavour by Wang et al. [32], as it provides an in-house dampness resource for bacterial activity and the development of self-healing concrete with minimum manual interference. In addition, it acts as a bacterial shield. A significant increase in collective encapsulation of bacteria with hydrogel bioreagents achieved healing between about 40% and 90%. A more or less 68% decrease in water permeability was observed. Effective healing through bacteria placed in a nutshell of hydrogel is possible owing to the uptake of water and retention capability of hydrogels. Wang et al. [30] indicated that pure hydrogel application could have a holding capacity of 70% and 30% of subsequent H₂O absorption for 12 h and 24 h, respectively, after exposure air with 60% RH and a temperature of 20 °C. This means that in tropical regions with higher moisture content and precipitations, hydrogels could be a superior option for absorbing and preserving water to initiate bacterial bioactivity. Nonetheless, it also relies on the type of hydrogel, i.e., whether it is ionic or non-ionic [30]. On the one hand, the ionic hydrogels are quick to respond to pH, and as a result, their uptake capacity may be influenced by chemicals prevalent in atmospheric air.

On the other hand, hydrogels which are non-ionic may be more suitable for this type of utilisation because ion-enclosing moisture does not impact its capability of withholding and water uptake. An optimum healing performance was revealed by Wang et al. [23] on using melamine-based microcapsules when wet and dry cycles were employed as curing proviso (wet time period was 16 h, while that of dry was 8 h). By utilising hydrogels, contact with water might be mitigated significantly. Furthermore, a prevailing advantage is the physical availability of water since hydrogels are sufficiently able to absorb atmospheric moisture, which allows for enhanced prediction to utilising this type of concrete in real-world structures. A recent study by Wang et al. [80] has explored the viability of carrying sodium alginate-based hydrogel spores of bacteria. It was found to be practicable following encapsulation in the hydrogel, which was measured by oxygen consumption at the broken site of the concrete samples. In the course of concrete mixing, however, 99% of the spores were found to be intact in the encapsulated form in some of the spores from the changed hydrogel. The 0.5% and 1% of hydrogel mass added did not have a significant impact on the workability of concrete; nevertheless, tensile and compression strength values decreased by about 23.40% and 30%, with 1% of the hydrogel. Similarly, negative effects may be attributed to macrovoid development on account of the supplement of a hydrogel. SAP extended two contradictory impacts on concrete; on the one hand, it provides moisture for inside curing and facilitates the strength development, while on the other hand, it enables the formation of voids in concrete, resulting in a decline in strength. The control of these two impacts relies on the W/C ratio and SAP dose, in addition to concrete age [82]. Employing the concept of gel/space ratio, at a water-to-cement ratio higher than 0.45, a supplement of SAP demonstrates much less influence on strength development. Wang et al. [78] applied a comparatively higher water-to-cement ratio of 0.50, which may also be one of the causes for the reduction in the strength in specimens with a supplement of superabsorbent polymers.

3.2.3. In Lightweight Concrete

In previous research, Jonkers [17] used porous, expanded clay, as a precursor to mineral precipitation to immobilise spores of bacteria and calcium lactate. The CaCO₃ is precipitated by the action of bacteria in the air when soft, and lightweight clay aggregates rupture. Specimens examined after immersing them in tap water for two weeks exhibited the highest healed width of crack of 0.46 mm. No loss in the feasibility of bacteria has been recorded thus far after six months. Clay aggregates are used extensively to produce lightweight concrete and also act as an internal source of humidity, which is essential for the

promotion of bacterial precipitation. However, numerous parameters such as the quantity of water in aggregate, the expanded spacing of clay aggregate, and pore structure have impacts upon its efficiency [83]. The resulting decrease in strength is a major restriction on the use of clay aggregates to replace usual granite aggregates. In standard strength concrete, aggregates constitute the mass of the concrete and are decisive in the compressive strength of concrete. In normal concrete, hard additives are resistant to cracks that ultimately pass through the matrix, which is comparatively weaker than the entire concrete.

Nonetheless, the cracks would most probably break down the aggregates if clay aggregates are used because the strength of clay aggregates may be less than that of the matrix. Soft aggregates thus create a weak plane that facilitates the formation of cracks. By implementing lightweight clay aggregate, Jonkers [23] monitored a strength decrease of up to 50 percent after 28 days, which may not meet structural requirements.

3.2.4. In Special Compounds of Minerals

Chemically, a mineral is known as diatomaceous earth (DE) is found abundantly in silica and originates from the shell for microorganisms known as 'diatoms'. It possesses an extremely porous structure and, as a result, potentially contributes to immobilisation. DE was employed by Wang et al. [28] for *Bacillus sphaericus* immobilisation. On cracking once, the microorganisms were activated through water or air; this would hydrolyse urea and lead to CaCO_3 precipitation from the mineral precursor of calcium nitrate used. Here, the width of healed crack also relied upon the medium for immersion, i.e., nutrient medium or water; nonetheless, crack widths ranging from 0.15 to 0.17 mm were found to be more or less wholly healed. High DE levels are better strengthened because bacterial cells are mostly absorbed in some hollow pores. Nevertheless, DE has a highly fine structure attributed to many nanoscale-sized pores that transport the medium to dry mortar. Therefore, using high levels of DE helps to absorb moisture in the cement matrix. Immobilisation is an effective way to retain the highly efficient mineral-forming ability of bacteria built in over a period of time, for self-healing bacterial concrete, according to Zhang et al. [84].

4. Biomineralisation Process

Biomineralisation is the process through which living organisms produce minerals. Mineralisation of carbonate products is frequently utilised in concrete technology. In the case of biomineralisation, living organisms play significant roles in mineralisation. More precisely, an inorganic mineral phase with a biopolymer is produced by living organisms [85]. There may be two types of biomineralisation: (i) biologically controlled mineralisation (BCM) and (ii) BIM. BIM is a bioinduced mineralisation of two types. On the one hand, minerals normally deposit in and on organic matrices or vesicles of the microbial cells in BCM types of biomineralisation processes, allowing organisms to have notable control over nucleation and mineral development and, consequently, over the composition, size, habit, and intracellular position of minerals [86,87].

On the other hand, in the BIM type of biomineralisation, organisms, chiefly microorganisms, secrete metabolic or more yields which react through ions or other compounds in the atmosphere, ensuing the successive deposit of mineral particles as by-products of metabolism [88]. The production of these biominerals is most probably not a premeditated and uncontrolled outcome of metabolism, which frequently provides the discriminating cementation through forming comparatively insoluble inorganic and organic substances. The aforementioned mineralised materials serve as cementitious compounds which are, at times, called biocements. Biocement comprises an alkalophilic microbe along with solutions of substrate and calcium ions. It has been highlighted extensively as a 'green material'. Its dependency on MICP is well known [89]. MICP use has been shown for heavy metals restoration [48–50], strengthening and enhancement of soil [90], restoration of stone components [51,53,91,92], treatment of wastewater [93], consolidation of sand [42], concrete strengthening [44], and durability of construction materials [54,94]. MICP involves complex

biochemical reactions controlled by two main enzymes—urease and carbonic anhydrase produced by microbes, using urea as a substratum and source of mineralised calcium.

The high cost of production is one of the key factors preventing the application of MICCP technology. The high cost is due to the cost and number of applications required for the microbial product. More time is needed to wet the construction material to better precipitate carbonates. With growing precipitation, increasing production of EPS, biofilm formation, and therefore plugging, can be expected. The costs of biodeposition treatment have been analysed by De Muynck et al. [95] both for the product price and the number of applications required. The product's theoretical cost relies on the costs of microorganisms and nutrient cost value. The cost of one kilogram (10^{11} CFU g^{-1}) of bacteria lyophilised is about 1100 EUR kg^{-1} . Applied bacteria with $2\text{--}3\text{ gm}^{-2}$ concentration has a cost of $2.2\text{--}3.3$ EUR m^{-2} . The estimated cost of nutrients is around 180 EUR kg^{-1} . The dosages vary between 0.04 and 0.08 kg m^{-2} , with nutrient costs reaching $7\text{--}15$ EUR m^{-2} , depending on the porosity of the stone. The complete price of the item is about $10\text{--}17$ EUR m^{-2} . In the event of strongly degraded surfaces, treatment costs are greater. This technology needs economical solutions to the use of bacteria and nutrients for its successful adoption and marketing.

4.1. Biocementation Process

Biocement consists of MICP between grain particles through the drainage of a liquid with alkalophilic bacteria, urea as calcium ion solution, and the substratum solution [96]. The bacterial hydrolysis urea produces the urease enzyme, and calcium is used as a source of energy for biocement production. The cement binds the grains into a solid mass collectively. The progression is suitable in applications such as soil stabilisation [97,98], earth construction [99], stone artefact restoration, plugging of water channels [100], and remediation of pollutants. For the correct bonding of the referred course, carbonates must be placed homogeneously in the intergranular voids.

The key function of microorganisms in the process of carbonate precipitations is mostly on account of their aptitude to craft an alkaline environment, i.e., higher pH and escalation of DIC, via their diverse physiological actions [101]. The increased alkalinity of the aforementioned materials creates a further challenge, i.e., biocementation in combination with binders such as lime or cement. This particular type of utilisation necessitates those microbes having higher alkaline pH patience biocementation has been demonstrated to make biosandstones of an appropriate compressive strength by binding sandstones [102]. It was also used to improve the compressive strength of other types of cement. When the biocementation of the amalgamating *Shewanella* species, isolated from hot springs, was achieved in mortar, a compressive strength increase of up to 25% was observed [62]. Biocementation was used for mortar manufacturing in the studies of Achal et al. [42,94] together with traditional cement. The newly developed fluid bacterial cell was added to a mixture of sand and cement, where bacterial culture was approved with a cement ratio of 0.47 for 70.6 mm cubes, and analogous formation was authorised, with the addition of coarse aggregates, to prepare concrete specimens [94]. This type of cementitious samples demonstrated 17% to 36% enhanced compressive strength, and water permeability resistance increased fourfold [103]. As the abovementioned study concluded, biocementation is dependent on bacterial strains, which means that the urease action is high, while the ability to precipitate calcium carbonate and therefore the composition is moderate. The higher-value nutritional profile encourages high calcium carbonate deposition. $CaCO_3$ precipitation on the cell surface and within the cement mortar matrix creates biocement, leading to an increase in the compressive strength of concrete.

4.2. Biodeposition Process

Bicarbonate coatings are precipitated in biodeposition on a porous substratum such as brick, cement, or mortar. MICP may deposit carbonate crystals in pores of the surface and prevent adverse substances from entering the substratum. Accordingly, such surfaces may,

therefore, be protected from chemical and water penetration on the surface of limestone, concrete, or bricks, for example, pore materials [104]. Biodeposition engages a variety of microorganisms, corridors, and environments. This can be achieved by exposing the porous surface to ureolytic bacterial civilisation. Submersion, ponding, or spraying can cause exposure. Numerous researchers for the strengthening and consolidation of limestone have discovered carbonate precipitation induced by microbial activity [105].

In order to restore and protect the degraded Euvil limestone, Dick et al. [56] have previously performed biodeposition treatment. In their research, limestone cubes of 30 mm size were incubated with a liquid medium, NaHCO_3 , urea, and CaCl_2 , with one percent of a range of *Bacillus* strains inoculated with five different *Bacillus sphaericus* strains and one strain of *Bacillus lentus*. In four weeks, multiple CaCO_3 coatings were deposited on the calcareous limestone. The study concluded that bacteria with enhanced ureolytic ability, i.e., *B. sphaericus*, achieved the most prominent and homogeneous CaCO_3 biodeposition on limestone cubes. A fruitful effort was then made to improve the effectiveness of biodeposition based on the effects of calcium salts and urea [106] in terms of their chemical parameters. In recent years, photosynthetic cyanobacteria have been studied, mostly for soil restoration [107].

The optimum calcium dose was assessed to be approximate. Increased urea and calcium concentrations at a certain level and repeated treatment increased the limestone resistance to water absorption due to the use of MICP. Furthermore, biodeposition-treated samples demonstrated diminished absorption of water and resistance towards sonication—the act of using the energy of sound to agitate particles in a specimen. A further study examined the impact of pore structure on the protective performance of MICP-based surface treatment using biodeposition with five types of limestone [105].

In recent times, the effectiveness of *B. sphaericus* has been investigated for the strengthening of limestone samples at different temperatures of 10, 20, 28, 37 °C, and 46% declined sorptivity was observed in biodeposited limestone samples in comparison with the control [108]. The results revealed that MICP occurred in higher amounts and at more depths in macroporous stones than in microporous ones. Biodeposition on cementitious samples was evinced to reduce the absorption of water and gas permeation [54]. Treatment was performed by immersion in bacterial cells of mortar cubes or concrete cylinders that grew for 24 h during the night before they were immersed in the nutrient solution.

Finally, the conclusion of De Muynck et al. [54] was to decrease the capillary absorption and to decrease the gas penetration due to the layer CaCO_3 deposition on the surface of the samples. Furthermore, by calculating the resistance of mortar and concrete samples to gradient degradation, the efficiency of biodeposition treatment with pure bacterial civilisations was examined [55]. CaCO_3 crystals induced by *B. sphaericus* are biodeposited. *B. sphaericus* reduced the water absorption by 65–90% depending on samples' porosity. Consequently, the rate of carbonation and migration of chloride decreased by about 25 to 30% and 10% to 40% correspondingly. Two constituent coating systems with the blocking of pores can be seen as the biodeposition treatment. It results in the sealing and development of pores, creating a biofilm on the surface of the sample [55]. This biofilm is an initial type of basic coverage for the carbonate coating. In biofilms, bacteria pull positively charged metal ions out of their vicinity and act as nucleation locations because their cell walls are adversely charged [58].

4.3. Process of Bioremediation

Concrete structures may deteriorate because of numerous reasons—namely, severe environmental conditions, overloading, or accidental damage, and hence, they require remediation to lengthen their service period. The application of polymers as synthetic agents for the remediation of such structures is widespread [109].

MICP can be an effective restoration method to repair the surface of damaged structures by precipitating carbonate crystals inside the cracks to plug them together. The strengthening of cracks and subsequent leaching is critical in many strategically essential

structures and in those which store dangerous chemical elements. In such circumstances, it would be an advantage to autonomously self-heal the concrete structure. As MICP can be set off by activated microbes when chemical agents are present, the concrete structure can automatically heal itself. Preliminary studies have shown that they can rebuild structures broken down and remediate cracks through cemented materials. The role of microbial function has long been recognised in preserving natural rocks [110]. MICP was also used in the cleaning of the crusted marbles and in confiscating sulphates [109].

The biocalcifying bacteria were found to function successfully for bioremediation of monuments or limestone buildings [40]. The layer of CaCO_3 on the surface of monumental rocks acts as a shield coating and closely follows the stone's nature. However, the structure of natural pores would not be blocked entirely; thus, soluble salts can pass through the stone [111].

Bioremediation in concrete samples can be carried out in two ways. Firstly, cells of bacteria with suitable nutrition, calcium source, and urea are induced into the mixture of concrete in the course of casting; when cracks appear, biominerals seal them through calcium carbonate formation [24]. Bacterial cells on the surface cracks in concrete are used for the second type. Bioremediation of concrete samples based on the MICP was introduced in a study by Ramachandran et al. [44], while *Sporosarcina pasteurii*-mixed sand has been used to increase the compressive strength by 61 percent in cracked mortars. In order to protect them from greater pH cement, they used polyurethane-encapsulated bacterial cells [66]. The filler material *S. pasteurii* was used to heal the 3 mm width and 18.8 mm deep cracks in the mortar specimen. *S. pasteurii* cells were added to natural sand. Here, SEM images demonstrated CaCO_3 precipitations in the healing area through rod-like bacteria [112]. Closing of cracks through CaCO_3 precipitation by bacteria *sphaericus* on concrete resulted in mitigation regarding water penetration and bridging of crack, which was displayed by an escalation in ultrasonic pulse velocity (UPV).

Wang et al. [24] have estimated the efficiency of bioremediation by using force recovery in mortar prisms (60 %) and decreasing water permeability in concrete cylinder specimens. In addition to SEM and an XRD analysis, CaCO_3 has been confirmed to be present in the repair material [112,113].

MICP biostructuring was successfully applied in Breda, The Netherlands, with the use of a spraying bio-based repair solution to process the damaged ramp of a garage parking area [114]. Compared with the control specimen, the effective treatment resulted in an increase in resistance, with a mass loss of 48% lower than the control. In case of moisture, water, or oxygen following the production of urease, and finally, of CaCO_3 precipitations that close the cracks when bacterial cycles are produced in a crack, they can increase. To overcome the problem of high pH in concrete, microcapsules have been used as bacterial carriers during the crack healing process of cement samples. Microcapsules consisting of melamine, together with a nutritional source such as yeast remover and deposition (engages ureas and calcium), can encapsulate spores or bacterial cells and produce a self-healing system [23]. This system is resistant to the high pH of concrete and is generated autonomously. Bacteria begin to germinate in the presence of water in the cracks, creating microcapsules in the breaking of the crack passageway and precipitating calcium carbonate for healing treatment. The most imperative factors for the biological remediation of cracks in concrete are the development of bacteria, urease activity, and precipitation amounts of a CaCO_3 medium that contains urea, including calcium ion sources.

5. Influence of Bacteria on Concrete Properties

A synopsis of the outcomes for the impact of MICP on concrete characteristics is included in this section. One of the main variables which MICP influences is diffusion kinetics due to changes in the pore structure. The diffusion of humidity and other ions which damage building construction materials have positive impacts. Enhanced strength is expected since the deposition of MICP can occur in the inter-granular voids and unite the grains collectively.

5.1. Setting Time Properties

Bacteria spores powder in the concrete mix is added so that the speed of concrete that relies on the calcium source is either increased or slowed down. The nutrient to bacteria is provided as lactate, nitrate, and formate of calcium. The calcium lactate supplement can delay the setting time, whereas calcium format and nitrate can increase concrete setting time speeds [115,116]. Wang et al. [23] originally investigated the effect of melamine-based microcapsules on characteristics of fresh mortar. The development of a second hydration peak was delayed by the addition of microcapsules, although cumulative heat generation after 7 days was quite similar to the control group. Ersan et al. [33] investigated the impact of several protective materials on mortar characteristics. The addition of diatomaceous earth or metakaolin reduced the initial and final setting times to around 100 and 250 min, respectively, while the use of air-entraining agents increased the initial and final setting times from 200 min to 240 min and from 300 min to 360 min, respectively. Using the electrical conductivity technique, Su et al. [117] demonstrated that direct mixing of bacteria and nutrients delayed the ultimate setting time and induction time of the cement.

5.2. Strength Properties

Biotechnological methods based on precipitation of CaCO_3 have improved the compressive strength of structural concrete. During the early curing period, microscopic cells received high-quality nourishment, due to the porous structure of the cement mortar. However, these cells became accustomed to a new atmosphere. On account of higher cement pH, there is a likelihood for bacterial cells to develop leisurely in the starting period and adjust to higher pH during the curing period. During the cell progression development, CaCO_3 precipitates on the surface of the cell and also in the matrix of cement mortar that may be by virtue of the incidence of diverse ions in the media. This contributes to lesser permeability as well as the porosity of the cement mortar. If many pores in the matrix are simultaneously sealed, oxygen and nutrients flow into bacterial cells is discontinued. The cell either dies or converts to endospores in due course. The nature of improved compressive strength can, therefore, be understood by microbial cells [44]. By inducing *Bacillus megaterium* bacteria in concrete, precipitations of CaCO_3 were found elevated in high-grade concrete in comparison with the low-grade concrete. Hence, high-grade concrete demonstrates more strength in comparison with low-grade concrete. For a concrete mixture with the highest grade of 50 MPa, the maximum growth rate of strength is 24 percent [17]. Cement was replaced by 10% fly ash, and 105 cell/mL *Sparcious pasteurii* bacteria were added. Due to CaCO_3 accumulations on cellular surfaces of microorganisms, a 20% increase in the compressive strength of fly ash concrete was recorded [18]. Due to the precipitation of CaCO_3 , the compressive strength induced by bacteria in silica fume concrete is enhanced. The presence of CaCO_3 in concrete was determined by microstructural analysis of concrete by means of XRD and SEM [18]. The compressive strength of concrete enclosing *Sparcina pasteurii* plus *Bacillus subtilis* bacteria (with 2 to 10^9 cells/mL) is 20% higher than concrete with no bacteria for 28 days, as recorded in [118].

While cement is replaced by different levels of fly ash, such as 10, 20, and 40 percent in a mortar, bacterial cells have accelerated compression by 19, 14, and 10 percent, compared with control samples, respectively [119]. GNP performs as a good-quality stimulating substance for homogeneous bacteria distribution ensuing in the highest crack-healing capability. The compressive force of concrete increased overall ages, due to microbial precipitation of CaCO_3 , when adding *Bacillus subtilis* bacteria plus GNP [52]. Compared with control cement mortar, the compressive strength of 28 days increased by the combined reactive spore powder [115]. CaCO_3 build-up on cell surfaces and the sand, as well as cement matrix pores, fill the pores of mortar and increase *Bacillus* sp. compressive strength [120].

The biocement-amalgamated concrete demonstrated elevated compressive strength compared with concrete [113,121]. *Bacillus pasteurii*, an alkalophilic aerobic bacterium, has been added to nutrient media with urea plus CaCl_2 and was used as a substitute for water

in OPC mortar. Compared with control mortar, with a strength of 55 MPa and no bacterial cells, a comparatively higher compressive strength of around 65 MPa from cement–mortar cubes with bacterial cells was measured at 28 days.

Ghosh et al. [62] showed a 17% increase in compressive strength and 25% increase after 7 and 28 days, in mortar due to *Shewanella* species biocementation.

Jonkers and Schlangen [64] have used cells of *S. pasteurii* for biocementation on mortar samples and found that there is no significant difference in relation to tensile strength between control which shows 7.78 N/mm² and bacterial samples, at 7.45 N/mm². However, the compressive strength was found to be slightly higher in the same case.

Furthermore, Jonkers and Schlangen [63] obtained the compression strength of mortar by *Bacillus pseudofirmus* and *Bacillus cohnii*, which was augmented by 10% for 28 days. The compressive strength of mortar was investigated by Achal et al. [42,112,122] by using industrial by-products as bacteria nutrients, such as lactose mother liquor (LML) plus corn steep liquor (CSL). When LML was used to develop *S. Pasteurii*—an increase of 17 percent in mortar compressive strength was reported at 28 days [42]. *S. Pasteurii* is formerly known as *B. Pasteurii*, a biocementation bacteria. The CSL-type media used to grow bacterial cells to achieve MICP, led to an increase of 35 percent in the compressive strength of 28-day concrete [112,122], which was greater than that encountered by the standard nutrient medium. The MICP process for the improvement of mortar compression was also examined by Park et al. [123] and was found to be an extremely promising bacterial isolate of the species of *Arthrobacter crystallopoietes* for biocementation. Afifudin et al. [124] achieved a 28 percent improvement in compressive strength of *Bacillus subtilis*-induced concrete, compared with control concrete. When bacterial cells were incorporated into the cemented material, they began to grow after their uptake of nutrients from pores of cemented materials and the surrounding area, and CaCO₃ was precipitated both on the cell and within the cement–mortar matrix. Only once the voids were sealed in the matrix, would nutrients flow, and bacterial cells be deprived of oxygen. Finally, the cells either died or transformed into endo-spores and performed as organic fibres [44].

Accordingly, in addition to MICP, by and large, an increase in strength was also caused owing to the presence of a sufficient quantity of organic materials in the matrix in the form of the microbial biomass. Overall, the increase in compressive strength is mainly due to the consolidation of voids in cemented materials with MICP biomineralisation yields. The MICP process was also presented on a wide scale to enhance granular soils [125].

The majority of studies focused on mechanical features of self-healing at the macroscale level. Several investigations [25,62,94,123] found that when calcite-forming bacteria were not protected, the compressive strength of concrete increased by 20%–40% after microbial precipitation, compared with control groups. Using low alkali cementitious materials as a protective carrier, Xu et al. [126] found a recover ratio of strength for microbial specimens that was about 2.3 times that of the control after 28 days. Shaheen et al. [127] discovered that bacteria trapped in LSP had a compressive strength return of 50% compared with a control series of 42%. Wang et al. [24] found a high strength restoration of 60% with PU-immobilised bacteria, compared with a strength recovery of only 5% with silica-gel-immobilised bacteria, which they ascribed to the stronger binding strength between PU and crack wall. Khushnood et al. [128] used RCA-immobilised bacteria to achieve an 85 percent compressive strength recovery after 28 days. Sierra-Beltran et al. [129] investigated the mechanical characteristics of a bio-based cement composite including LWA impregnated with 15% calcium lactate and bacterial spores. The composite entirely regained its deflection capability owing to bacterial self-healing. Furthermore, Skevi et al. [130] found that equivalent compressive strength responses were seen when dead or living cells were directly introduced to mortar formulations without the inclusion of nutrients. They concluded that the improved strength was not likely due to MICP but rather to bacteria stimulating the deposition of hydration products (i.e., nucleation sites) or to the nature of their cell walls.

5.3. Water Permeability

Resistance to water penetration is a very significant and primary parameter to characterise the prolonged durability of concrete since it confirms the infiltration of destructive materials accountable for the degradation of concrete. The permeability of cementitious materials is normally based on the pore system which is composed of several variables, e.g., porosity, tortuosity, connectivity, distribution of size, microcracks, and also of the specific surface [131]. MICP has demonstrated the capability to significantly mitigate water penetration of cementitious substances and other types of construction materials. In Thouars, a church known as 'Saint Médard', renovated in 1993, CaCO₃ used from microbes—namely, 'Biocalcin', caused a fivefold reduction in absorption of water from stones sans altering its aesthetic look [40].

Afterwards, analogous examinations were performed on cement-based mortar as well as based on *B. sphaericus*-induced biodeposition. De Muynck et al. [54] indicated a 65–90% drop in the absorption of water of the mortar, due to the formation of a CaCO₃ layer on the surface of samples. Biodeposition through *B. sphaericus* caused a noteworthy drop in water penetration in concrete, in which cracks mending was executed [57]. The presence on the surface and within the porous matrix of biomass and carbonate crystals resulted in reduced permeability of mortar samples.

Biodeposition therapy was considered by De Muynck et al. [54], highlighting two components with pore sealing attributes that constitute coating systems. The cementitious surface is sealed by the bacteria themselves, and a biofilm is produced. As the bacteria within the biofilm attract metal ions positive charge from its neighbour and act as a nucleation site, due to their possession of the negative charge in the cell wall, the biofilm works as a core cover for carbonate coating [58]. The rise in certain enzymes such as urease and carbonic anhydrase leads to liquid phase super-saturation activities with respect to CaCO₃, which lead to the various CaCO₃ crystal precipitations in the biofilm. Achal et al. [103] also showed a decrease in permeation due to the deposition of a layer of CaCO₃ crystals on the surface.

After the bacterial intervention of *S. pasteurii* mutant, the penetration depth of concrete cubes with 150 mm dimensions was radically reduced. They measured water resistance in the pinnacle and the side of the concrete cubes as the depth of infiltration. Due to the gravity of the deposition and screening of pores at the peak, the infiltration at the side was greater at the summit. The water penetration at the pinnacle was quadruple less in comparison with control samples, while it was roughly 2.5 times less at the side faces. They concluded that the biocement was designed to have low concrete permeability as a result of the precipitation of CaCO₃ developed a dense interfacial zone.

Permeability confirms the infiltration of violent compounds responsible for the degradation of concrete beneath the pressure gradient and, therefore, is considered the main feature regarding concrete durability. This relies on several factors related to the pore network of cementitious materials, quantified by tortuosity, porosity, specific surface, connectivity, distribution of size, and microcracks. The parameters referenced include water-to-cement ratio control, particle size distribution, hardened cementitious materials age, and destructive component intrusion. The deposition of calcium carbonate in concrete resulted in reduced water absorption and concrete permeability. A study by Cheng et al. [98] revealed the induction of *S. pasteurii*. Fly ash concrete with *S. pasteurii* bacteria was accompanied by a decline in porosity and concrete permeability. The absorption of water in concrete was found to be quadrupled with 105 cells/mL of bacteria. CaCO₃ precipitations are plugged by bacteria in bioconcrete pores [18]. The cubes that were cast with *Bacillus megaterium* supplement and their nutrients, due to the deposition of microbial CaCO₃, absorbed over threefold less water than the control samples [119]. The introduction of *Bacillus aerius* bacteria results in a decrease in water absorption and porosity owing to precipitations of CaCO₃ which, in turn, enhance the concrete structures' durability [16]. After 28 days, each control group of concrete samples showed high-to-modest permeation, but bacterial (105 cell/mL) samples producing calcite showed high-to-low permeability

as pores were sealed with CaCO_3 [68]. Due to microbial precipitations that reduce water absorption for recycled aggregates, the remarkable performance of recycled aggregate has improved [132,133].

5.4. Chloride Ion Permeability

Chloride infiltration is one of the most predominant environmental attacks which results in corrosion of reinforced steel in concrete, contributing to the weakening of the concrete structure. Chloride ion penetration rate into concrete is mainly based on the inner pores of a concrete structure. In turn, the pore structure relies on several other elements, such as cure conditions, mixture design, hydration scale, the application of additional cementing materials, and type of construction.

A rapid chloride ion permeability test is performed by monitoring the number of electric currents passed by a sample. The quality rating of the concrete consists of its permeability, according to the charge passing through the sample. Concrete resistance to chloride permeation can be increased through the incorporation of bacteria in concrete. The average chloride infiltration in bacterial concrete was 11.7 percent lower in comparison with concrete without bacteria. Additionally, it was observed that the addition of *Sparcious pasteurii* and *Bacillus subtilis* species to concrete reduced chloride penetration in concrete and enhanced the reduction tendency of concrete mass subjected to sulphate formation [118].

A supplement of *Bacillus aerius* bacterial cells inside concrete could be able to mitigate the entire charge passed through control concrete and RHA–concrete samples. The least charge passed at all curing ages is shown on bacterial concrete. Charges decreased in the bacterial concrete sample by 55.8, by 49.9, and by 48.4 percent in the 7, 28, 56 days of age [16]. The 10 percent silica fume in the concrete complement of *Sparcious pasteurii* bacteria showed an excellent quality resistance to rapid chloride penetration (380 coulombs) [18].

The highest mitigation in terms of chloride ions was observed in the case of fly ash concrete samples with the inclusion of *Sporosarcina pasteurii* bacteria species of 105 cells/mL concentration; nevertheless, concrete with 30% fly ash displayed a very low infiltration of merely 762 coulombs. The durability of the concrete structures which are subject to deicing salts or marine environments is well defined by their ability to hold firmly in response to chloride ion infiltration [98].

In the case of reinforced concrete, chloride ion diffusion can spoil reinforcement. Chloride-induced corrosion is one of the significant deteriorating mechanisms that influence the long-term performance of building structures. The concrete steel reinforcement is intrinsically protected from corrosion by passivating the steel surface by the high alkalinity of the concrete, which can be neutralised due to environmental effects such as carbonation. The threat of corrosion becomes severe in these cases when chlorides contact steel reinforcement. When MICP connects the pores, the growth of chloride ions can be prevented. However, in MICP, a chloride salt is often utilised, and whether the chloride ions presence accelerates corrosion in stainless steel bars must be examined. Researchers have identified alternatives to calcium chloride as the source of calcium to dispose of this problem. Calcium nitrate has been used productively as an effective source of calcium provided by *S. pasteurii* [134].

Calcium chloride also causes massive ammonium production, which accelerates the risk of reinforcement corrosion. The application of calcium lactate proposed in Neville [135] at its metabolic change does not produce considerable amounts of ammonia. *Bacillus cohnii*-induced calcium lactate produced large amounts of CaCO_3 precipitations with a size of 20 to 80 μm on cracking surfaces to heal the cracks [27].

Calcium lactate and calcium glutamate precipitations of CaCO_3 introduced with *B. cohnii* were compared by Xu et al. [136], and a higher thickness of CaCO_3 was deposited with calcium glutamate than the calcium lactate was observed.

The effects of CaCO_3 precipitations and urea gauges from *Bacillus* species have been reported by Achal and Pan [38] using different sources for calcium, such as chloride, oxide, acetates, and nitrate. In comparison with 418 U/mL^{-1} , calcium nitrate CR₂ with calcium

chloride was 432 U/mL^{-1} , calcium acetate 401 U/mL^{-1} , and calcium oxide 389 U/mL^{-1} . CaCO_3 precipitation with calcium chloride had the highest value.

De Muynck et al. [55] highlighted the fact that the bacterial deposition of a CaCO_3 layer on the surface of mortar samples led to a decrease in capillary absorption of water and gas permeability. The presence of biomass greatly reduced gas permeability in cement mortar, which led to increased carbonation resistance [54]. The thickness of the carbonate layer, which was approximately 30 to 50 μm , contributed to the increased resistance against carbonation. They proposed that the defensive effect of the biodeposition treatment of carbonation can be enhanced by supplementary treatment with bacteria and calcium sources or with an increased concentration of calcium ions.

De Muynck et al. [54] have measured the resistance of treated specimens using an escalated migration test to chloride infiltration. MICP-based biological deposition treatment resulted in significantly lower migration coefficients of 10–40%, compared with untreated samples for chlorides. Additionally, the boosted resistance to chloride penetration in bacterial mortars was analogous to acrylic coating, in addition to water repellent silanes with silicones, and was greater than the silanes/siloxanes mix.

Achal et al. [103] have assessed and monitored the efficiency of the MICP for chloride infiltration in cylindrical concrete specimens through a rapid chloride ion test (RCPT). RCPT was measured in cylindrical samples of *S. pasteurii* biocell concrete, and results were compared with bacterial-cell-free control samples. The average charge passed in test control samples amounted to 3177 C, whereas it was 1019 and 1185 C for specimens made with bacterial cells. MICP reduced the permeability from 'moderate' to 'low' in accordance with ASTM C1202-05. In their conclusion, they reported that MICP could be a first-quality concrete sealant that is cost effective and eco-friendly, which eventually results in an improvement in the durability of construction materials. They concluded that MICP is a high-quality, cost-efficient, and environmentally friendly concrete sealant that eventually leads to improved construction material durability.

5.5. Corrosion of Reinforcement

Carbonate precipitations induced by bacteria are proposed as an innovative and environmentally friendly approach for the fortification of reinforcement against its corrosion in concrete [69].

Qian et al. have prepared H_2SO_4 solutions of dissimilar pH for examining the acid resistance of the CaCO_3 layer deposited by bacteria *pasteurii* on mortar cubes with 30 mm dimensions [69]. As CaCO_3 precipitated on cement samples introduced by the bacterial urease enzyme, which further resisted the acid attack, the outcomes demonstrated striking improvement in surface permeability resistance of mortar. In their conclusion [69], the authors indicated that the deposited layer CaCO_3 can withstand acid rain to some extent, which is very helpful in guarding structural reinforcement corrosion, particularly in coastal regions, and in enhancing their durability. In terms of their resistance to corrosion, biologically mineralised reinforced concrete beams were evaluated [137]. Concrete specimens were prepared using CaCO_3 precipitating *Bacillus* species that were strengthened with a standard reinforcement bar with a length of 300 mm and a diameter of 25 mm or 415 grade of iron. In the first two days, the current was increased in control samples from 17.5 to 40 mA, and up to 180 mA was observed at the end of seven days, while approximately 85 mA current was introduced during the same time period in the treatment of bacterial samples. A crack having a width of 0.3 mm is considered as a breakdown limit for reinforced concrete samples, as previously defined. The crack with the same width, i.e., 0.3 mm, was encountered in control samples within 36 h, while the same was monitored subsequent to a period of seven days in samples treated with bacteria. The comparison leads to the conclusion that incorporation of MICP can noticeably lengthen the service span of concrete structures. Moreover, measurement of corrosion current density (I_{corr}) displayed that MICPs perform remarkably in mitigating the corrosion current in RC samples. In comparison with considerably elevated I_{corr} of 60.83 mA/m^2 in control samples, it ranged between

14.78 and 20.03 mA/m² in samples with bacteria. The aforementioned quadruple fall in Icorr was because of biodeposition. Finally, they concluded that the deposition of CaCO₃ might have encouraged the defensive passive film and acted as a corrosion inhibitor by intervention in the course of movement in such specimens [137].

5.6. Sequestration of Carbon Dioxide (CO₂)

Investigations on MICP have confirmed that it is a technology of the future, whereby prolonged storage of atmospheric CO₂ occurs in the form of carbonates such as CaCO₃, in naturally occurring minerals such as magnesite (MgCO₃), dolomite (Ca.Mg(CO₃)₂), etc. [138]. The microbes present in such deposits have been characterised [139], and an attempt has been made to recognise its mechanisms [140]. To convert CO₂ into CO₃, the carbon dioxide hydration is the step that regulates its rate, with a forward reaction constant of between 6.2 and 10.3 at 25 °C [141,142]. Biocatalyst carbonic anhydrase or carbonate dehydratases that form a group of enzymes has which catalyse the inter-conversion among carbon dioxide (CO₂) and H₂O and the delinked ions of carbonic acid, i.e., ions of HCO₃ and Hydrogen. It has gained increasing attention among researchers due to its central role in the calcification of invertebrates of marine origin known as ‘molluscs’ corals, hard tissues of vertebrates, and fish otoliths [143,144].

Early fruitful endeavours for sequestration of CO₂ with the help of carbonic anhydrases are prepared by bovine carbonic anhydrases that promoted examinations on bacteria sources [145–147]. MICP, through carbonic anhydrase, has put forward the likelihood to capture carbon dioxide (CO₂) into a stable, safe, and appropriate environment of CO₃ reservoirs, thus becoming a perpetual technique for discarding CO₂ [148,149].

Recently, bacterial carbonic anhydrase derived from large, rod-shaped *Bacillus megaterium* species of bacteria in biomineralisation of CaCO₃ has been found to perform synergistically with urease [46]. The enzyme’s ability to increase CaCO₃ mineral precipitation from calcium-rich solutions is used through chemical buffers and pure CO₂ gas [150–155]. After MICP formation through bacterial cells, several investigations on MICP resulting from the addition of bacteria have highlighted species such as Cyanobacteria or Cyanophyta as effective sources for capturing and impounding carbon by using solar energy to conduct photosynthesis, whereby CO₂ changes into recalcitrant CaCO₃ minerals [156,157]. These bacteria attain their energy through photosynthesis. They are merely photosynthetic prokaryotes capable of generating oxygen (O₂). By accelerating the supply of dissolved inorganic carbon in environments, MICP can secure carbon by making cations easily available in the solution and by maintaining an alkaline pH [158]. In nature, these cations are easily accessible via evaporation of deposits, waste brines, salt aquifers, oil waste, and a virtually unlimited supply of seawater [155,159,160].

6. Effect of Adding Organic Agents or Capsules on Concrete Characteristics

An additive to external chemical products such as organic nutrients can affect the mechanical strength of concrete by changing its microstructure [62,123,161]. Moreover, capsules form poor bonding in the matrix, and when the enclosed agent is released, they are used just as voids or holes within the concrete. Such voids influence chiefly the mechanical strength of concrete. The direct induction of spores causes losses of about 8% to 10% in connection with compressive and flexural strengths [39]. An analogous outcome was accounted by Jonkers et al. [27], with spores induced to cement stone samples from *Batillus cohnii* as well as *Bacillus pseudofirmus* which resulted in a strength drop of approximately 10 percent on 3, 7, and 28 days. CERUP manufactured by Silva et al. [162] for mortar at 0.5% and 1% by weight of cement did not show a negative effect on compressive strength, but a higher dose of 3% and 5% had a significantly negative impact on strength. Wang et al. [23] reported experimental results showing a significant impact on hydration, compressive, and stress strength of the addition of nutrients and capsules. By adding 5% microcapsules in the weight of cement, the compressive strength was reduced by up to 34%. Tensile strength was also significantly influenced by a more than 3 percent capsule supplement. Compared with

microcapsules, the addition of nutrients had a less intense impact on concrete strength. The degree and hydration rate were improved, while microcapsules and yeast extract hindered hydration if calcium nitrate was used as a precursor. Calcium nitrate can be regarded here as a concrete escalator additive. The calcium nitrate dose must, therefore, be carefully regulated. The hydration obstacle resulting from the addition of yeast extract has been designed to detect the effect of the nutrient which protects cement particles from reaction with water.

Jonkers [22] showed a substantial decrease in strength, while the replacement of granite aggregates enclosed the spores of bacteria with lightweight ingredients. As lightweight components fracture easily if loaded, they reduce the progress of autonomous self-healing by freeing spores and precursors, but this type of concrete has lower strength and, thus, cannot be applied to structural components. For the autonomous self-healing process, precursors and nutrients for bacteria are induced to concrete. It has also been reported that when only calcium lactate was used as a mineral precursor, concrete strength was not negatively affected; rather, it was improved to some degree [22,27,39]. Adding yeast and peptone extract reduces compressive strength. Late-stage strength may be even lower than early stage strength, particularly when peptone is incorporated [27]. The microcapsules, when added, caused a reduction in water absorption in concrete, as recorded by [23]. The strength decline was caused principally through modification of microstructure owing to the reduced extent of hydration and weak distribution of products of hydration, which, in turn, resulted from adding nutrients and microcapsules together.

Nevertheless, water absorption depends on the extent of hydration as well as on the pores opened in the matrix. The matrix-complemented microcapsules can block pores if the waterproofing effect of bacterial nutrients is present. As a result, it is feasible that though there is a reduction in strength, durability attributes such as absorption of water may be enhanced. The addition of microcapsules can also affect concrete rheology that must be studied when creating capsules based on autonomous, self-healing concrete activities.

Rheology attributes may be influenced by the material type of capsule, shape, and size. Sphere-shaped microcapsules may provide lubricating action through a decrease in aggregate interlocking [163]. It decreases plastic viscosity and yield stress and, hence, enhances flow behaviour. Nevertheless, enhancement in rheology would also rely upon the kind of material used for capsule shells. They cannot be extremely absorbent as absorption of part of the water that is used for mixing decreases free water available for fresh paste slump or flow.

7. Mechanical and Durability Properties of Healing Capacity and Recovery

The recovery of original concrete properties would depend on a healed breakage width, precipitation bonding with the matrix, and structure, as well as precipitated strength. A high recovery rate of attributes compared with that of the original concrete is sought for effective autonomous self-healing. Bacterial concrete has several factors, such as curing conditions, the local concentration of viable spores and nutrients, concrete age, and time for heals, that enable autonomous self-healing treatment. A wider range of crack widths has also been observed for the use of a healing bioagent. It is based on a number of factors.

7.1. Recovery of Mechanical Properties

Encapsulation of bacteria cells with PU and silica gel individually inside tubes of glass was carried out by Wang et al. [24]. The crack width for mechanical strength recovery was approximately 0.35 mm. In the case of silica gel, further CaCO_3 precipitations were gained, but strength recovery was found to be approximately 5 percent. PU samples showed a recovery in strength between 50% and 80%. Nevertheless, the mechanical strength restoration activities of bacteria were uncertain, as the recovery from strength was not significant, unlike in cells of living and departed bacteria. In silica gel, the amount of precipitation was high, compared with PU, while the strength recovery for PU was higher. PU is a good-quality plugging agent that mainly provides mechanical strength recovery.

Pei et al. [25] established that bacterial cell wall incorporation might improve concrete strength through three feasible mechanisms. The same can be true for embedded spores of bacteria once metabolism has begun. CO_2 generated by bacterial metabolic activities reacts with non-reacted portlandite in the crack faces, to turn into more powerful CaCO_3 . This insoluble CaCO_3 formed by direct precipitations or alteration of portlandite is also responsible for the reduction in porosity and increase in particle packing ability, giving rise to mechanical strength [164]. Ultimately, bacterial cells negatively charged could become nuclear sites for hydration of cement. Nevertheless, in fresh concrete, this is more adequate. Though CaCO_3 is a strong and beneficial component for cement, strength recovery as a result of binding CaCO_3 to concrete may be limited.

7.2. Durability Properties Recovery

Effective self-healing in concrete results in a complete or nearly complete improvement in durability and mechanical strength from the original probe. Durability properties are frequently gauged by water penetration and absorption of water examinations. Crack healing means plugging in air or water any vacuum or interconnected pores via extraneous chemicals. This, in turn, reduces the permeation and absorption of water. Mitigation of permeability entirely by bacteria activities is because of pore sealing by CaCO_3 , which possesses a very inferior solubility [165].

The ratio of healing decreases from 83% for a 0.1–0.3 mm crack to 30%, while its breaking width is 0.8 mm, as observed by Luo et al. [165]. Healing products in high levels are likely to be washed away from the concrete surface due to moisture or entering fluids. A reduction in healing ratio may be due to the loss of sealant agent from a crack or a lack of curing agent, which is also consistent with the outcomes of Xu and Yao [39]. The outcomes point to the correct direction, as metabolism bacterial and precipitations might be restricted by their concentration, precursor material, and nutrients at the crack site. The rate of precipitations is sluggish, and hence, if the width of the crack is large, then precipitations can be washed away by entering water or other chemical fluids ahead of crack sealing. Furthermore, it was found that concrete specimens immersed in water have an elevated healing ratio in comparison with samples exposed to wet curing [14] that might be attributed to superior transportation of healing agents. This is owing to the concentration diversity among the matrix along with the surface in the immersed state. In the abovementioned investigation, a promising ratio of healing for wet curing was attained at an early age. This simply means sufficient water can infiltrate within and turn out to be accessible for metabolic actions by bacteria. The rate of the repair was sluggish for wet curing and turned to be more or less equal to the rate of water curing in the late stage. An analogous observation was made in practice by Wang et al. [23], employing microencapsulated spores of *Bacillus sphaericus* into the capsules made up of melamine. The crack with the highest width of 850 to 970 μm was healed by immersing it underwater; nevertheless, the best-healed crack area was observed while being exposed to wet and dry cycles for water. In the wet cycle, the dampness could enter the matrix, while sufficient oxygen could be obtained for spores during the dry cycle. In the event of incubation by incessant submergence in water, the spores are not offered oxygen, and more importantly, nonstop contact with water cannot be conveniently feasible in many cases. The crack area is a superior measure of healing, compared with the width of crack because it regards remedial in two directions. The investigations on water penetration demonstrated that concrete samples with 5 percent capsules exhibited minimum ultimate coefficient of permeability on exposing to dry and wet cycles.

Even though the value is in proximity to amid 3 percent capsules, much less variation in terms of values of permeability was found in samples of 5%. Interestingly, the study points out that the concrete with 5% capsules is of inferior quality in terms of its strength. Therefore, the poor value of permeation might be understood as a drop in porosity with more capsules, but for this, capsules are only expected to be intact. Subsequent to rupture, inferior permeability for 5% capsules may be assigned to the water-proofing impact that

the enclosed material may have caused, accordingly resulting in slightly lower permeation. Nevertheless, 3% volume was considered optimal in terms of impact on mechanical strength and decreased water permeability coefficients. The type of material of the capsule or medium used for immobilisation can considerably affect the drop in water permeability.

Bacterial spores in the silica gel and polyurethane have been immobilised in Wang et al. [24], whereby the mitigation of permeability was recorded by two orders of magnitude in the former, as well as more excellent bacterial action, leading to porous blockages through precipitation formation and decreasing permeability. However, in the latter case of polyurethane immobilisation, the least precipitates were encountered, and elevated mitigation in permeability in comparison with silica gel was assigned to the water-proofing impact of polyurethane [24]. This kind of influence is well known as PU foam, and spores in glass capsules have been immobilised.

Following the split, the foam can pour out and plug concrete pores. Bacterial activities were fruitful in precipitating CaCO_3 that could merely reduce the porosity of PU foam and had the least impact on sealing concrete pores directly. Considerable mitigation in context to permeability, of more or less 68% has been accounted by Wang et al. [29] by means of hydrogel for encapsulation of spores of bacteria and bioreagents. The greatest width of crack 0.5 mm was healed; however, there was a higher disparity of healing ratio of 40–90% even for 0.3–0.4 mm cracks. Nevertheless, it is considered as an enhancement in comparison with the case in which merely hydrogel is utilised. Enhanced healing may be expected due to the proportionate spores and biological reagents distribution, while their encapsulation in hydrogel occurs collectively. In the event of cracking, bacteria would have access to nutrients and precursor material because of this approach of encapsulation. The absorption of humidity and hydrogel retention also played a role in higher bacterial activity. In addition to bacterial precipitation, decreased permeability can also be escalated by autonomous auto-healing promoted by the internal hydrogel curing system.

Wang et al. [28] showed significant improvement in water absorption as bacteria were immobilised in diatomaceous earth. Water absorption declined by one-third and 50 percent, while cracked samples were kept warm in yeast, calcium nitrate, urease, and water, correspondingly. Calcium ions and urea of the deposit medium can infiltrate and increase precipitation in CaCO_3 during the healing treatment. This impact was probably more important because the concrete samples were merely 14 days old when cracks were initiated, and consequently, nutrients and calcium could be transmitted without effort from the deposition medium within the concrete matrix.

Nevertheless, Wang et al. [28] have also reported that higher concentrations of calcium ions mitigate the number of hydroxyl ions from calcium hydroxide dissolution leading to a decrease in pH. Precipitation can be reduced due to the decrease in optimum alkalinity for bacterium activities. In the case of late-stage concrete, which develops dense microstructure due to perfect hydration, when submerged in the deposition medium, calcium ions cannot enter easily into the concrete. The concentration of calcium would, therefore, be higher only on the surface that restricts bacterial spores from accessing the external source of calcium and deposition source. The findings of this review can be utilised to assess the toughness of autonomous self-healing capsule-based approaches for sustainable construction structures in terms of the emphasised aspects of toughness such as pervasiveness, shelf life, reliability, quality, repeatability, and versatility, as proposed by Li and Herbert [166].

8. Microstructure and Macrostructure Properties

The precipitations of CaCO_3 in the form of crystals found in mortar and concrete were envisaged in association with rod-shaped bacteria through SEM analysis. The said deposition enhanced the impermeable attribute of the concrete since it performed as a barricade against entering detrimental materials into the specimen [119]. The addition of bacteria to concrete can enhance its microstructure through mineral precipitations which have been confirmed by EDS, SEM, and XRD analysis. According to Andalib et al. [17], a concentration supplement of 30 to 105 cfu/mL of *Bacillus megaterium* species had the

greatest calcium weight of about 38.76% in comparison with other mix proportions with and without bacterial concrete [17]. The SEM analysis exhibited dissimilar CaCO_3 crystals entrenched by bacteria. It was observed that CaCO_3 presented higher quantities of calcium in the sample which was verified through EDX and XRD analyses. This can enhance the durability performance of concrete [165,167]. The SEM images of without bacterial addition, control samples, as well as bacterial concrete were analysed. The images confirmed the formation of CaCO_3 crystals in bacterial concrete [18]. Bacterial amalgamation increased the compressive strength of RHA concrete by deposition of CaCO_3 in pores, and SEM images corroborated this finding. CaCO_3 plugged the voids in bacterial concrete [16]. The deposition of CaCO_3 inside the cracks in the test specimens was authenticated by the outcomes attained utilising microstructures. Accordingly, the absorption of water, chloride permeability, and acid infiltration were reduced by increasing the signal transmission rate of UPV [168].

Commonly, these examinations are carried out at microscale to identify and show the deposits inside concrete cracks following autonomous self-healing. This enhances the credibility of the results achieved. For this reason, mostly, the researchers have executed investigations using techniques such as scanning electron microscopy (SEM), field-emission scanning electron microscopy (FESEM), and X-ray diffraction (XRD). SEM recognises the morphology of the materials inside the concrete cracks [169]. The deposited materials are precipitations made up of CaCO_3 through strains of different bacteria, a product of hydration and polymer yields. Quite recently, autonomous self-healing behaviour is estimated utilising Raman spectroscopy [170]. In such a manner, the reliability of employing bacteria as an agent of autonomous self-healing of the cracks in concrete is maximised [63,171]. Additionally, the findings acquired utilising microstructures determined crystals of irregular deposits inside concrete crack test specimens. Accordingly, water absorption, acid ingress, and chloride ion permeability were found to decline significantly with the enhancement in the rate of signal transmission of UPV.

Researchers have carried out some investigations on macroscale in terms of the efficiency of autonomous self-healing courses [172]. These comprise toughness and split tensile examinations [173,174]. Some other tests are UPV and water permeability, as suggested by several researchers [171,175–177]. Some of them have carried out further tests on sorptivity and gas penetration to verify the autonomous self-healing efficiency of sodium silicate; colloidal silica encapsulated and tetraethyl ortho-silicate [76]. It was found that gas permeability and sorptivity dropped by 18 percent and 69 percent, respectively. Granger and Loukili [178] carried out investigations on hardness to estimate the performance of the material that had undergone healing. Farhayu et al. [179] revealed that the flexural resistance of the concrete benefited by healing was 50 percent more than the control concrete sample. Snoeck et al. [180] executed thermogravimetric analysis (TGA) to identify the deposited white colour material inside the crack. A few other investigations were conducted by several other researchers on chloride penetration, oxygen profile, porosity, and pore size distribution tests. In terms of compressive strength, it is highlighted that it can be recovered up to 60% subsequent to autonomous self-healing. Moreover, UPV was found to augment following autonomous self-healing.

Consequently, based on outcomes of examinations, durability attributes with more crucial impacts over a prolonged period could, thus, be addressed by employing a biology-based, autonomous self-healing approach. Such an approach extends the life span of concrete structures. Nevertheless, other imperative investigations on bonding strength among the materials autonomously deposited inside concrete cracks, as well as gas permeability examinations and stress–strain curves, are hardly ever addressed. Accordingly, it is necessary to estimate the improvement of autonomous self-healing concrete utilising a variety of approaches. As a result, it will permit a healthier similarity of bonding capability of the material deposited inside the frames and compatible with the composition of concrete.

XRD examination revealed a combination of mineral precipitations generated by bacteria, including calcite (Ca), aragonite (AR), and vaterite (Va). XRD spectra for wet–dry

and fully wet conditions were compared, and the amplitude of peaks in the bacteria's XRD spectra under wet-dry curing was greater than that under fully wet curing. Aragonite had distinct peaks and maximum peaks for wet dry-cured bacterial concrete specimens and fully wet-cured bacterial concrete at 28.511 and 38.447, respectively, while calcite had peaked at 20.298, 25.163, 26.208, 27.358, 47.612, 49.256, 55.605, 56.481, etc. It should be highlighted that a greater calcite formation was detected in bacteria specimens, which is the explanation for the reduction in water absorption from the concrete mix. This demonstrates the existence of calcium carbonate [181].

9. Nanostructure Properties

The abovementioned examinations were conducted at nanoscale to maximise the dependability of the findings of the tests further. Of late, Xu and Yao [39] assessed the autonomous self-healing competence utilising nanostructure investigation. The transition zone's average nanomechanical values are 20% higher than those of the outside precipitates that served as a strong bond between the concrete and deposited layer matrix. It is meaningful to perform bond strength tests at the macro- and nanolevels to verify the interface among the deposits and the cement-based material inside concrete cracks, augmenting the reliability of the course. Calvo et al. [182] integrated a new, self-healing scheme based on two micro/nanoparticles: silica microcapsules that contain epoxy sealing compounds (CAP) and nanoparticles with amine-functionalised silica.

10. Impact on Sustainability

MICP should be considered highly sustainable owing to its economical energy necessity and scope for recycling. Nevertheless, its utilisation in standalone manufacturing building materials is not predicted almost immediately. However, the use of biocement produced by MICP as admixtures in the practice of mending or rehabilitation of construction materials has led to promising outcomes in advancing toward sustainable construction. The abovementioned data indicate clearly that MICP can improve the presentation of extensively used building materials, i.e., sand, limestone, concrete, and mortar. The capability of MICP to offer strength augmentation and permeability diminution of concrete is the most promising indicator for sustainability. As the need for material quantities decreases with the increase in strength, reduced permeability, together with augmented durability, is one of the most effective means of increasing sustainability [183]. A technique of life cycle assessment (LCA), which makes it easy to estimate environmental impacts, is broadly well regarded as one of the key points proving the sustainability aspects of yields and courses.

Moreover, several studies have identified the environmental impact of technological advances on cement and concrete [184–188]. One of the studies demonstrated the contextual importance of the LCA's key steps as follows: (1) definition of goals and scope, (2) inventory of the life cycle; (3) life cycle impact assessment. A new trend in the utilisation of the LCA method summarised that as regards the cement strength, the LCI estimation results are influenced by more than 10%. The significance of decisive strength properties and durability properties linked to functional units for concrete LCA has also been recorded [160]. The LCA method can help to examine the impacts on environments by the MICP utilisation.

Nevertheless, currently, the scope of such investigations is constrained because of quite considerable challenges, such as the absence of an exact definition of the system frontiers, inadequate data on LCI-restricted understanding of the performance of the life cycle, as well as service life. Further studies and analyses are required, even in the assessment of urea and CaCl_2 embodied energy [189]. Efforts by advanced studies on both MICP and utilisation of the LCA method may expectedly fill this knowledge gap. In what follows, a few preliminary inferences concerning LCA for MICP are highlighted.

Reproducible and reliable data accessibility for LCA may remain limited in the near future due to a lack of commercial applications. Consequently, LCA investigations might have to be started with locally accessible, restricted data for LCI development. LCA functional units should be used in the context of the presentation, such as parameters on

strength and durability. It should not be regarded for quantity such as mass, volume, etc. A reliable evaluation of the service life and functional performance of the concrete will contribute significantly to the advance of reliable LCA investigations. The application of by-products in industrial wastes as a source of Ca, urea, as well as nutrients for the growth of bacteria can noticeably decrease the impacts of MICP, both environmentally and economically, which needs to be thoroughly and carefully considered in investigations of LCA.

11. Benefits of MICP and Application of Bacteria in Concrete

Two main advantages of MICP are that (i) the pore structure of a material can be modified by the surface deposition, and (ii) the loose granular material can be strengthened by collectively binding them. Thus far, researchers have ascertained both advantages of MICP; however, they also emphasise the inconsistencies of the process in terms of geography and environment. The technology should be scaled to commercial dimensions in standard test protocols and approval standards. It is a careful exercise of energy, sequestration of CO₂, and potential for recycling.

Nevertheless, quantitative estimation data of the aforementioned advantages are limited. A framework for LCA of MICP and structures applied through MICP is necessary. Nonetheless, it might be prudent to begin approximate evaluations from the current data. Some concerns exist in the adaptation of bioprocesses such as MICP in the construction industry, as they are not used for such procedures. All these concerns are related to health and economic aspects. Dissemination of research findings on bacterial pathogenicity in the construction-related workforce may help in alleviating these concerns. MICP expands remarkable opportunities for recycling and economic recovery of industrial by-products. Nevertheless, they must be adapted locally according to by-product accessibility. MICP offers a vast potential to alleviate some aspects of the supremacy of building and infrastructure industries, apart from all its previously mentioned challenges.

The benefits of the bacterial application in concrete include the following:

- It reduces water and chloride permeability;
- The findings support the suggestions that the 'microbial concrete' could be an alternative concrete sealant and of higher quality that is not merely cost effective but also environmentally sound and eventually helps enhance the durability of building construction materials;
- Bacteria were evinced to be more effective in closing microcracks even up to width 0.97 mm and depth 32 mm.

12. Practical Applications

Prior to initiating the application of this concept for commercial use, a broad revelation is necessitated. There is also a need to resolve the crisis of nutrient optimisation. Concrete attributes such as corrosion, shrinkage, and carbonation must still be investigated in detail. A methodical examination of the above characteristics will shed light on the concrete behaviour in real time. Although the referred notion has demonstrated potential in laboratory outcomes, the ability still requires further study under non-ideal temperature ranges, higher salt concentrations, and in later ages of concrete elements, in order to shield massive concrete structures. Detailed knowledge of autonomous self-healing ability and its diverse applications—key for its promotion in the field—is a possible way of achieving a specific service life assessment.

13. Further Development

MICP technology has considerable potential for improving building materials quality. Nevertheless, the full impact of the technology cannot be assessed unless on a commercial scale. The greatest challenge is that construction-related professionals are not aware of bioprocesses. Commonly, every one of the bacteria is misunderstood as a health hazard. This means the key resistance to their acceptance is in human psychology. Knowledge of the

pathogenicity of microbes would provide considerable advantage to both the construction community and industry. There is little concern about bacteria's survival in high pH conditions when MICP is used in combination with cement. Several higher-pH-tolerant bacterial strains were separated. Additionally, methods of encapsulating bacteria and the application of CaCl_2 in MICP may pose concerns to related individuals. A number of substitutes to CaCl_2 have been identified. Nevertheless, consent on cost may still require the use of CaCl_2 . Most importantly, the economic aspects of this technology need to be defined. Recycled materials can be appropriately used to achieve double the benefits of lower costs and an enhanced environment. Achal et al. [112] showed that recycling efficiencies are industrial by-products and potential pollutants, such as lactose mother liquor (LML) and corn steep liquor (CSL), a source of nutrients. MICP's recycling cost fell by 70 %. Similarly, other MICP components may be exploring suitable, natural or recycled, substitutions.

14. Discussion

Higher costs are currently associated with various repair activities of deteriorating concrete structures throughout their service life. The life cycle cost model, as conceived by Van Breugel [190], describes this aspect. In some instances, repair costs even exceed the initial building costs. A natural, bio-based treatment is much more beneficial in the case of autonomous bacterial encapsulation. Moreover, there is very little evidence of autonomous, bio-based, self-healing action under fatigue. Fatigue of bacterial encapsulation would rely on the capsule's release behaviour. The controlled discharge or 'smart release' of the capsules may achieve healing in multiple loading cycles. Dong et al. [191] worked to develop smart releasing microcapsules to cure concrete degradations, especially the corrosion of reinforcing bars caused by a decreased alkalinity of the concrete matrix. Nonetheless, advanced research on the use of such intelligent capsules in bio-based autonomous self-healing is not fully implemented at this stage. A nanocapsule can be examined to decrease the size of the poor spots made of mortar because of the capsule induction. A technique called 'sonification' has been successfully obtained for urea-formaldehyde (UF) capsules, which have diameters of 220 nm, and a thickness of 77 nm with a more uniform shell walking. However, this has been encapsulated by only chemical healing agents [192]. However, their use can be expanded to bio-based autonomous healing concrete if the material for the capsule is well designed and incorporated with bioagents. Nevertheless, it must be ensured that the nanoparticle waste does not agglomerate in the concrete matrix because the agglomerations of a nanoparticle can initiate crack development in the matrix. Results thus far suggest that the time needed for crack sealing and healing in bacterial concrete with appropriate curing conditions is a prolonged period, characteristically spanning at least two to three weeks. Additional interdisciplinary studies may be essential to form genetically modified civilisations, which can thrive more quickly and last longer. This means that splits of larger widths can also be healed in a short time if such research is successful in the future. Controlling crack width is also an essential aspect in order to achieve rapid and well-organised healing through bioaction [179]. An application of hybrid fibre could be detected as a means for controlling crack widths that can lead to an increased regeneration after healing the original characteristics [193,194]. Advanced research on the efficiency of autonomous healing is required at the actual, local environment on the site. Such research should aim to improve service life, reduce costs, and consider the environmental and social advantages. It will be meaningful to investigate how employing autonomous self-healing bacterial concrete contributes to adaptation to changes in climatic conditions [76,195]. Techniques for sustainability estimation such as LCA can also be used to improve the life cycle of biobased self-healing systems [196–198]. With these in mind, healing bioagents may be mixed with carbon sequestration material [199] to make building composites that are more sustainable and autonomously 'green'. Specifications and test standards should ultimately be developed to assess bio-based autonomous healing performance in building structures. By shedding light on studies of a precedent decade on bio-based autonomous-healing materials, its execution in the construction sector may

be predicted in the future. Nevertheless, the present technical and technological obstacles must be first addressed to make it suitable for extensive industrial application.

15. Conclusions

This paper reviewed the self-healing of bacterial concrete and the efficiency of self-healing of cracks by encapsulated bacteria on recovery of various mechanical and durability properties. Based on in-depth review the following conclusions can be made:

1. The calcium lactate supplement can delay the setting time, whereas calcium format and nitrate can increase concrete setting time speeds;
2. This review also found that concrete compressive strength ability has a beneficial effect on bacteria. The benefit of bacteria reduces water penetration and chloride ion permeability;
3. *Bacillus pasteurii* and *Bacillus subtilis* are the most effective, useful bacteria to heal cracks in concrete;
4. CaCO_3 precipitation improved the compressive strength of the structural concrete;
5. During cell progression development, CaCO_3 precipitation on the cell surface and in the matrix of cement mortar may be due to the incidence of diverse ions in the media. This contributes to less permeability and porosity of the cement mortar;
6. The addition of *Sparcious pasteurii* and *Bacillus subtilis* species into concrete reduced chloride penetration in concrete which also enhances the declining tendency of concrete mass subjected to sulphate;
7. The deposited layer CaCO_3 can withstand acid rain to some extent and is very helpful in guarding structural reinforcement corrosion, particularly in coastal regions, and in enhancing their durability;
8. *Bacillus cohnii*, in addition to *Bacillus pseudofirmus*, resulted in a reduction in strength;
9. The addition of bacteria in concrete can enhance its microstructure through mineral precipitations which have been confirmed by EDS, SEM, and XRD analyses.

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Abbreviations

SEM	Scanning electron microscopy
GHG	Greenhouse gas
GNP	Graphite nanoplatelets
LWA	Lightweight aggregates
PU	Polyurethane
DE	Diatomaceous earth
UPV	Ultrasonic pulse velocity
CSL	Lactose mother liquor
CSL	Corn steep liquor
RCPT	Rapid chloride ion test
CO_2	Carbon dioxide
LCA	Life cycle assessment
UF	Urea-formaldehyde

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