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# Microbial repairing of concrete & its role in CO<sub>2</sub> sequestration: a critical review

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#### **Abstract**

**Background** Being the most widely used construction material, concrete health is considered a very important aspect from the structural point of view. Microcracks in concrete cause water and chlorine ions to enter the structure, causing the concrete to degrade and the reinforcement to corrode, posing an unacceptable level of structural risk. Hence repair of these cracks in an eco-friendly and cost-effective way is in the interest of various researchers. Microbially induced calcite precipitation (MICP) is an effective way considered by various researchers to heal those concrete cracks along with an important environmental contribution of CO<sub>2</sub> (carbon dioxide) sequestration in the process.

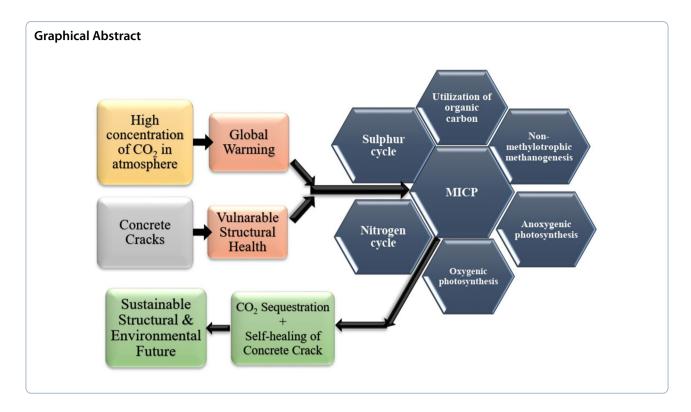
**Main content** As the current concentration of  $CO_2$  in the earth's atmosphere is about 412 ppm, it possesses a deadly threat to the environmental issue of global warming. The use of bacteria for MICP can not only be a viable solution to repairing concrete cracks but also can play an important role of  $CO_2$  arrestation in carbonate form. This will help in carbon level management to lessen the adverse effects of this greenhouse gas on the atmospheric environment, particularly on the climate. To overcome the insufficiency of studies concentrating on this aspect, this review article focuses on the metabolic pathways and mechanisms of MICP and highlights the value of MICP for  $CO_2$  arrestation/ sequestration from the atmosphere during the process of self-healing of concrete cracks, which is also the novelty of this work. An overview of recent studies on the implementation of MICP in concrete crack repair is used to discuss and analyse the factors influencing the effectiveness of MICP in the process, including various approaches used for  $CO_2$  sequestration. Furthermore, this investigation concentrates on finding the scope of work in the same field for the most effective ways of  $CO_2$  sequestration in the process of self-healing cracks of concrete.

**Conclusion** In a prospective study, MICP can be an effective technology for  $CO_2$  sequestration in concrete crack repair, as it can reduce adverse environmental impacts and provide greener environment. This critical study concludes that MICP can bear a significant role in arrestation/sequestration of  $CO_2$ , under proper atmospheric conditions with a cautious selection of microorganisms and its nutrient for the MICP procedure.

**Keywords** Self-healing concrete, CO<sub>2</sub> sequestration, MICP, Bacteria, Ureolysis, Cyanobacteria, Crack repair, Biomineralization

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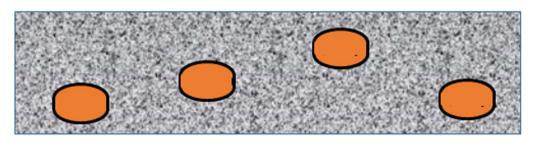
#### 1 Background

The construction industry directly aids in the development of economic activities whereas, simultaneously, it also exploits natural and physical resources. The construction and building sector accounts for roughly a quarter of all carbon dioxide (CO<sub>2</sub>) emissions [1]. Concrete has been the most commonly used building material in the world since the nineteenth century. The formation of microcracks in concrete structures reduces their service life by allowing water and chlorine ions to enter the structure [2], causing concrete deterioration and reinforcement of corrosion, resulting in an excessive degree of structural risk. As a result of this phenomenon, expensive repairs and maintenance are needed on a regular basis. This structural degradation, coupled with the weakening of concrete structures, leads to an economic burden by increasing repairing cost. In general, for crack remedy, materials like epoxies are used. However, they have their own drawbacks, such as they are expensive, change the appearance of the structure to a certain level, and need consistent maintenance. In traditional, but high-binder mixture concrete, microcracks with a width smaller than 0.2 mm can be seen to be autogenously selfhealed [3–7]. But simultaneously, the application of highbinder concrete mix will allow the extra use of cement in concrete which in return encourages an enhanced cement production that leads to higher global anthropogenic CO<sub>2</sub> emission. To minimize the risk in a cost-effective manner, researchers have focused on developing a safe and self-healing process that could revolutionize the construction of long-lasting concrete structures. This self-healing method is established by bacteria-induced calcium carbonate (calcite) precipitation in the concrete cracks. This phenomenon is known as microbially induced calcite precipitation (MICP), and the concrete with this phenomenon is known as 'Bacterial concrete' or 'Self-healing concrete'. This phenomenon focuses on the application of microbes as a healing agent for cracks in existing concrete structures. The basic strategy (Fig. 1) is to use microorganisms, preferably those which can form stable and dormant spores, and apply them in the concrete structure as microcapsules/pellets that also contain specific nutrients (such as calcium lactate). When the concrete slabs are intact and solid, these spores remain dormant, but when cracks are formed in the concrete, water seeps in, and the bacteria start growing. The bacteria utilize the nutrient such as calcium lactate for its growth. The microbial urease can hydrolyse urea to produce ammonia and CO<sub>2</sub>. The released ammonia increases the pH of the surroundings, which helps in the accretion of insoluble CaCO<sub>3</sub>. It is the most used and studied pathway in the line of self-healing microbial concrete (Fig. 1). In the process, another severe environmental problem in the current scenario, i.e., emission of greenhouse gas, CO<sub>2</sub> can also be checked. In fact, the current concentration of about 412 ppm CO<sub>2</sub> in the earth's atmosphere [8],

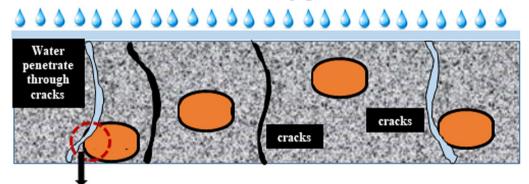
#### 1. Make pellets of urease positive endospore forming bacteria



#### 2. Mix pellets with concrete mix and make the structure



#### Crack in the concrete structure and seepage of water



#### 4. Activation of bacteria, nutrient utilization, and healing of cracks

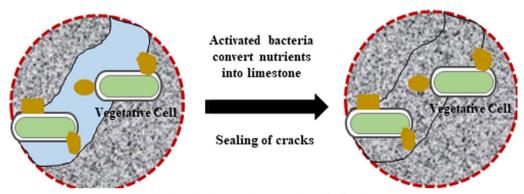


Fig. 1 Steps involved in Ureolysis, the most used and studied MICP pathway in the line of self-healing microbial concrete

already possesses the risk of global warming. The use of bacteria for MICP can not only be a feasible solution to repair concrete cracks but also can arrest  $CO_2$  as carbonate. This will help us to achieve the goal of carbon

level management and reduce the potential environmental threat of global warming. The present review article highlights the role of a different group of microorganisms and the pathways operative in them, which help in  $CO_2$ 

sequestration from the atmosphere and in the process can be utilized for self-healing of concrete cracks.

#### 2 Main text

#### 2.1 Research significance

It is well known that carbon dioxide is the leading and the primary greenhouse gas with the highest percentage among other greenhouse gases. According to the National Ready Mixed Concrete Association, the production of 1 kg of concrete releases 0.9 kg of CO<sub>2</sub> [9]. Concrete is the most commonly used material on the planet, accounting for 8% of overall global carbon emissions [10]. It is intrinsically fragile, necessitating regular repair or replacement, which are costly and emit enormous amounts of CO<sub>2</sub>. Hence, this CO<sub>2</sub> sequestration via the healing/repair of concrete will reduce the amount of CO<sub>2</sub> in the environment, specifically, it will balance some of the quantity of emitted CO2, from the construction industry. The utilization of the MICP process can not only be a viable approach to repairing concrete cracks but it can also intercept CO<sub>2</sub> as carbonate from escaping into the environment. Though there are several individual review articles present on MICP and concrete healing, there is a scarcity in the field addressing the research gap that signifies both the aspect of concrete healing and its utilization in CO<sub>2</sub> sequestration. Therefore, this review addresses the novel research aspect of the utilization of concrete crack healing by MICP keeping the target of CO<sub>2</sub> sequestration from the environment. This laborious study included the efforts of a vast field of expertise including individual sectors such as Civil Engineering, Chemical Engineering, Microbiology, Biotechnology, Chemistry, etc. as well as their interdisciplinary exertion. Hence, this study will be advantageous for a vast readership, ultimately benefiting and contributing in the goal of carbon level management and reduction of the possible environmental impact of global warming.

## 2.2 Importance of CO<sub>2</sub> sequestration and its role in self-healing concrete

The concentration of the principal greenhouse gas, CO<sub>2</sub> has been on the rise due to massive urbanization, industrialization, and various anthropogenic activities that are dependent on fossil fuels, resulting in global warming

and thus, climate change. The development in the construction industry also resulted in the enhancement of cement production leading to higher CO2 emissions in the environment. Therefore, there is an urgent requirement for the effective management of carbon footprint. CO<sub>2</sub> sequestration, also known as "Carbon trap" or "CO<sub>2</sub> arrestation," is a method for storing CO2 or its associated forms from the atmosphere for extended periods of time in order to reduce the amount of carbon in the atmosphere, resulting in a healthier environment. In this scenario, the utilization of microorganisms for CO<sub>2</sub> sequestration is a feasible and economical approach that is also beneficial for a sustainable environment as well. This is known as biological CO<sub>2</sub> sequestration. From the structural point of view, the self-healing method of concrete cracks, established by bacteria-induced calcium carbonate (calcite) precipitation i.e., MICP, in the concrete cracks is a very effective method of CO<sub>2</sub> sequestration. The microorganisms mainly conduct the sequestration of atmospheric CO2 and fix it into different carbonate minerals such as calcite, magnesite, and dolomite [11-13]. A wide range of microorganisms can induce carbonate precipitation through biological activities such as photosynthesis, ureolysis, denitrification, and sulphate reduction, or by acting as a template for crystal nucleation [14]. A significant amount of research has been conducted on MICP technology concentrating on ureolytic bacteria for crack repair in concrete structures and on cyanobacteria for carbon sequestration.

#### 2.3 Organisms and pathways

Biomineralization or to be specific, MICP has a contribution to the crack healing of concrete. In this process, metabolites generated by the induced microorganisms, react either among themselves or with components in the environment to produce and precipitate biominerals in carbonate forms that help in healing the concrete cracks (Fig. 1). Biomineralization yields a variety of minerals [15–17] especially bacteria are capable of producing a wide variety of them in the form of sulphides, carbonates, phosphates, and silicates [18, 19]. Among them, CaCO<sub>3</sub> precipitation is of interest to researchers due to its efficient compatibility and bonding capacity with concrete compositions. Microorganisms, in the presence of a

**Table 1** Different metabolic pathways of calcium carbonate precipitation

Autotrophic bacteria	Heterotrophic bacteria	
Non-methylotrophic methanogenesis Anoxygenic photosynthesis Oxygenic photosynthesis	Nitrogen cycle	ammonification of amino acids degradation of urea and uric acid dissimilatory reduction of nitrates
	Sulphur cycle	dissimilatory reduction of sulphates
	Utilization of organic carbon	

Ca-source, can produce  $CaCO_3$  extracellularly by means of an autotropic or heterotrophic metabolic pathway [19] (Table 1).

#### 2.3.1 Heterotrophic pathway

Heterotrophic growth on organic acid salts (citrate, acetate, succinate, lactate, oxalate, glyoxylate, and malate) of different bacterial genera such as *Bacillus, Arthrobacter*, and *Rhodococcus* species, produces  $\mathrm{CO_3}^{2-}$  minerals. Based on the presence of salts and C-sources in the medium, these bacteria can produce various crystals such as  $\mathrm{CaCO_3}$  and  $\mathrm{MgCO_3}$ , using organic compounds as a source of energy. *Arthrobacter* and *Bacillus* species are capable of  $\mathrm{CaCO_3}$ -precipitation [18] under an alkaline carbonate medium which is a very important aspect for being applicable in the healing of concrete cracks. Thus, future explorations must be made from different alkaline environments to find the most suitable microorganism that can realize the dream of self-healing concretes.

The summary of chemical reactions to form  $CaCO_3$  in the presence of  $Ca(CH_3COO)_2$  acting as the source of low molecular weight acid and  $Ca^{2+}$  are shown in the equations below [Eqs. (1)–(3)] [20].

$$CH_{3}COO^{-} + 2O_{2} \xrightarrow{Heterotrophic \ bacteria} 2CO_{2} + H_{2}O + OH^{-}$$

$$\tag{1}$$

$$2\text{CO}_2 + \text{OH}^- \rightarrow \text{CO}_2 + \text{HCO}_3^- \tag{2}$$

$$2HCO_3^- + Ca^{2+} \rightarrow CaCO_3 + CO_2 + H_2O$$
 (3)

The other two mechanisms of calcium carbonate production are sulphur cycle and nitrogen cycle. The dissimilatory sulphate reduction is followed in sulphur cycle mechanism. In the presence of organic matter, calcium source, and sulphate in the medium, calcium carbonate is produced. Due to the degasification of hydrogen sulphide, pH is increased which leads to the reaction toward calcium carbonate precipitation [21]. The entire reaction of producing calcium carbonate by the reduction of calcium sulphate (CaSO<sub>4</sub>) to calcium sulphide (CaS) using sulphate reducing bacteria, is shown below [Eqs. (4)–(7)] [22].

$$CaSO_4 + 2(CH_2O) \rightarrow CaS + 2CO_2 + 2H_2O$$
 (4)

$$CaS + 2H_2O \rightarrow Ca(OH)_2 + H_2S \tag{5}$$

$$CO_2 + H_2O \rightarrow H_2CO_3 \tag{6}$$

$$Ca(OH)_2 + H_2CO_3 \rightarrow CaCO_3 \downarrow +2H_2O$$
 (7)

The Nitrogen cycle includes three different types of pathways, (i) urea or uric acid degradation (ureolysis),

(ii) ammonification of amino acids, and (iii) dissimilatory nitrate reduction to produce carbonate or bicarbonate [18]. When adequate calcium ions are present in the medium, bacteria can go through these nitrogen metabolisms leading to calcium carbonate precipitation [23].

In ureolysis pathway, the microbial urease produces ammonia and  $\mathrm{CO}_2$  from the hydrolysis of urea [24–26]. The released ammonia subsequently increases the pH of the surroundings [27–29], leading to the accretion of insoluble  $\mathrm{CaCO}_3$  [Eqs. (4) to (7)] [30] and in the process will prevent  $\mathrm{CO}_2$  from being released into the atmosphere. These metabolic chemical translations help in calcium carbonate precipitation, generally in the form of calcite which is stable and also abundant in nature. This calcium carbonate precipitation plays the important role of a barrier and blocks the ingress of corrosive chemicals into concrete cracks [31], and thus, saving the concrete structure. It is the most used and studied pathway in the line of self-healing microbial concrete.

$$CO(NH_2)_2 + H_2O \rightarrow NH_2COOH + NH_3$$
 (8)

$$NH_2COOH + H_2O \rightarrow NH_3 + H_2CO_3$$
 (9)

$$H_2CO_3 \leftrightarrow HCO_3^- + H^+$$
 (10)

$$2NH_3 + 2H_2O \leftrightarrow 2NH_4^+ + 2OH^-$$
 (pH increase)

$$HCO_3^- + H^+ + 2NH_4^+ + 2OH^- \leftrightarrow CO_3^{2-} + 2NH_4^+ + 2H_2O$$
(12)

$$CO_3^{2-} + Ca^{2+} \rightarrow CaCO_3 \downarrow$$
 (carbonate precipitation) (13)

$$CO(NH_2)_2 + 2H_2O + Ca^{2+}$$
  
 $\rightarrow 2NH^{4+} + CaCO_3 \downarrow \text{ (overall reaction)}$  (14)

Apart from its several positives, this pathway has a few drawbacks such as the emission of nitrogen oxide in the atmosphere, and increased risk of salt damage by conversion to nitric acid in concrete due to the production of an excessive amount of ammonia in the matrix [18]. To deal with this drawback of excessive ammonium ion production, few researchers [6, 25, 26, 32] have proposed the idea of metabolic conversion of organic compound (organic acid salt) to calcium carbonate. When organic acids (such as calcium lactate) are aerobically oxidized, carbon dioxide is generated in an alkaline atmosphere, which is then converted to  $CaCO_3$  [Eq. (24)] in the presence of  $Ca^{+2}$  [25, 33–35]. Compared to ureolysis pathway, this metabolic conversion is more suitable with respect to compatibility with

concrete matrix composition, protection of reinforcement bars, and most importantly high production of  $CaCO_3$  but no ammonium.

$$CaC_6H_{10}O_6 + 6O_2 \xrightarrow{\text{Metabolic conversion}} CaCO_3 \downarrow +5CO_2 + 5H_2O$$
(15)

Few aerobic Gram-negative bacterial strains can use amino acids as their sole energy source during the ammonification of amino acids. Myxococcus has been shown to be an effective biosynthesis bacterium for a variety of minerals, including carbonates, sulphates, phosphates, oxalates, chlorides, and silicates [36]. Myxococcus xanthus can induce calcium carbonate precipitation in a nutrient medium of calcium acetate by active alkalinization, with ammonia and carbon dioxide as byproducts. At higher pH levels,  $CO_2$  appears to dissolve and turn into either  $HCO_3^-$  or  $CO_3^{2-}$  [36].

Organic compound 
$$+ NO_3^- + H^+ \xrightarrow{Denitrification}$$
 (16)  
 $CO_2 + H_2O + N_2$ 

$$CO_2 + 2OH^- \rightarrow CO_3^{2-} + H_2O$$
 (17)

$$Ca^{2+} + CO_3^{2-} \rightarrow CaCO_3 \downarrow \tag{18}$$

Under the nitrogen cycle, another subclass is denitrification, which works as a dissimilatory nitrate reduction. In this pathway, minerals are precipitated via the respiratory process of denitrifying bacteria, through oxidation of organic compounds, and by the reduction of nitrate (NO<sub>3</sub><sup>-</sup>) to nitrite (NO<sub>2</sub><sup>-</sup>), nitric oxide (NO), nitrous oxide (N<sub>2</sub>O), and ultimately to nitrogen gas (N<sub>2</sub>). Mainly facultative anaerobes, such as Denitrobacillus, Micrococcus, Alcaligenes, Spirillum, Thiobacillus, Pseudomonas and Achromobacter species [18], are capable of denitrification. Hence, such processes are effective for application in anaerobic zones. But similar to ammonification, this process also produces some by-products in the process such as carbon dioxide, water, and nitrogen. Due to the consumption of H<sup>+</sup>, an increase in pH takes place during the denitrification process resulting in CO<sub>3</sub><sup>2-</sup> or HCO<sub>3</sub><sup>-</sup> production [37] which further reacts with the calcium source leading to the precipitation of calcium carbonate [Eqs. (16)–(18)]. Production of calcium carbonate using the denitrification process in concrete is still under research and development stage. Moreover, ureolysis is a much faster process to biomineralize CaCO<sub>3</sub> compared to this denitrification pathway [38].

#### 2.3.2 Autotrophic pathway

In the autotrophic pathway, microbes convert  ${\rm CO_2}$  to carbonate, mainly in three different ways: (i) oxygenic

photosynthesis (by cyanobacteria and algae), (ii) anoxygenic photosynthesis (by purple bacteria), and (iii) nonmethylotrophic methanogenesis (by methanogenic archaea) [21].

Both oxygenic and anoxygenic photosynthetic bacteria produce CaCO<sub>3</sub> in the presence of calcium ions. The difference between these two photosynthetic metabolic processes is the use of electron donors which is H<sub>2</sub>O in the former and H<sub>2</sub>S in the latter [39, 40]. As no oxygen is generated in the later process it is called anoxygenic photosynthesis. During microbial photosynthesis, due to the removal of CO<sub>2</sub> from the bicarbonate solutions, a localized increment in pH occurs, which leads to  $CaCO_3$ -precipitation in the presence of  $Ca^{2+}$  ions using either reaction 1 or 2 [Eqs. (23) and (24)] [41]. As the main requirement of this process is the occurrence of CO<sub>2</sub> in the environment, utilization of this photosynthesis pathway for concrete crack repair is possible only in environments where the concrete is exposed to CO<sub>2</sub> and light.

The summary of photosynthetic chemical reactions for the production of calcium carbonate is shown below in Eqs. (19) to (24):

$$CO_2 + H_2O \xrightarrow{Oxygenic Photosynthesis} (CH_2O) + O_2$$
 (19)

$$CO_2 + 2H_2S + H_2O \xrightarrow{Anoxygenic Photosynthesis}$$

$$(CH_2O) + 2S + 2H_2O$$
(20)

$$2HCO_3^- \leftrightarrow CO_2 + CO_3^{2-} + H_2O \tag{21}$$

$$CO_3^{2-} + H_2O \leftrightarrow HCO_3^- + OH^-$$
 (22)

Thereafter in the occurrence of calcium ions,

$$Ca_2^+ + 2HCO_3^- \leftrightarrow CaCO_3 \downarrow +CO_2 + H_2O$$
 (Reaction 1) (23)

$$Ca_2^+ + CO_3^{2-} \leftrightarrow CaCO_3 \downarrow \quad (Reaction 2)$$
 (24)

In the non-methylotrophic methanogenesis pathway, CO<sub>2</sub> and H<sub>2</sub> are converted to CH<sub>4</sub>. HCO<sub>3</sub><sup>-</sup> is generated by the anaerobic oxidation of methane with electron acceptors such as sulphate [23]. When calcium ions are present, the carbonate formed produces calcium carbonate, which then precipitates [Eqs. (25)–(27)]. This pathway is more applicable to the marine ecosystem. CO<sub>2</sub>-type hydrogenotrophic methanogens (e.g., *Methanobrevibacter aboriphilus, Methanosarcina barkeri, Methanothermobacter thermautotrophicus,* and *Methanothermobacter marburgensis*) [19] are a few examples of methanogens that employ this pathway. These types

of methanogenic archaea under anaerobic conditions can produce methane by using  $CO_2$  and  $H_2$  as carbon and energy sources, respectively.

$$CO_2 + 4H_2 \rightarrow CH_4 + 2H_2O$$
 (25)

$$CH_4 + SO_4^{2-} \rightarrow HCO_3^- + HS + H_2O$$
 (26)

$$Ca^{2+} + 2HCO_3^- \leftrightarrow CaCO_3 \downarrow +CO_2 + H_2O$$
 (27)

## 2.4 Merits and demerits of different MICP cycles used in concrete

The use of oxygenic photosynthesis by different cyanobacteria such as Dichothrix, Lyngbya, Gloeocapsa, and Synechococcus species, might be a very important process for concrete crack healing, as these cyanobacteria are capable of surviving in alkaline environments similar to concrete that have pH of 11–13. By the use of methane oxidation, a few researchers [42] also suggested that, in this pathway, bacteria can utilize the environmentally harmful gas such as CH<sub>4</sub>, as well as H<sub>2</sub>S, which is harmful to concrete, in their metabolism process and thereafter, ultimately ending in the calcite precipitation. Whereas, the sulphur cycle hampers the health of concrete by producing harmful corrosive agent H<sub>2</sub>S that leads to the rapid deterioration of the concrete structure. The most frequently used MICP mechanism for MICP in concrete is the utilization of ureolysis pathway, where the microbial urease produces ammonia and CO<sub>2</sub> from hydrolysis of urea combined with different calcium compounds as nutrients. Although this process enhances the concrete properties such as compressive strength, concrete crack healing, decreasing the material permeability, etc., it has a few major drawbacks like, it is a temperature-dependent mechanism, with emission of environmentally harmful gas nitrogen oxide in the atmosphere, along with the increased risk of salt damage by conversion to nitric acid in concrete due to production of an excessive amount of ammonia in the matrix. To overcome the drawback of excessive ammonium ion production of ureolysis, the idea of metabolic conversion of organic compound (organic acid salt) to calcium carbonate came into the scenario, where the bacteria directly utilize the organic material for their metabolism and CaCO3 is produced as a by-product leading to a higher production of CaCO<sub>3</sub> without any ammonium. Another most useful mechanism is Denitrification with these advantages as it can happen in oxygen-deficient subsurface environments with only the presence of nitrate, such as the inner part of concrete cracks. But the by-products of denitrification (i.e., N2 gas, possibly a very little amount of unprecipitated CO<sub>2</sub>) are mainly non-toxic and chemically inert, whereas the gases produced in other metabolic pathways such as ammonia (from ureolysis), and hydrogen sulfide (from sulfate reduction), that poses threat to the environment, structures as well as human health. Although the final result of denitrification is innocuous nitrogen gas, there is a significant disadvantage to this process in that three toxic intermediates, namely nitrite, nitric oxide, and nitrous oxide, can build if incomplete microbial nitrate reduction takes place. Thus, making this process undergoes further research to overcome the drawbacks.

## 2.5 CO<sub>2</sub> sequestration via carbonate precipitation in microbial concrete and its effect

The application of MICP is not only confined to addressing the environmental issues of sequestering the greenhouse gas CO<sub>2</sub> but is also effective in structural engineering issue as self-healing of concrete cracks [36, 43]. Compared to the conventional approaches to crack repair of concrete, calcium carbonate bio-deposition by MICP (Table 2) also blocks the penetration of aggressive substances. In MICP, both the quality and quantity of precipitated crystal, in terms of density, thickness, cohesion, and effective bond with the concrete matrix [36, 44], bear major impacts on the effectiveness of surface crack remediation.

Researchers [21, 25, 36, 44-51] all over the globe have conducted studies on the application of MICP for calcium carbonate precipitation in the healing of concrete cracks along with the effect of the same on different properties of concrete. Most researchers used the alkaliphilic spore-forming bacteria such as S. pasteurii, B. sphaericus, and B. subtilis [52-54] (Table 2) for this task for their capability to withstand the high alkalinity of concrete. In most cases, the pathway of ureolysis was the preferred choice for its highest output of CaCO<sub>3</sub> deposition compared to other pathways [44, 47, 48]. Stable spore formers that can remain dormant for years are again preferred as they can remain idle in the concrete structure for years (even better if they can remain viable for decades) and will work only when cracks are formed and moisture seeps in. The healing agent consisting of the microorganism can be applied in concrete cracks either directly or in encapsulated form. According to Lee and Park [55], a perfect encapsulation carrier for the bacterial cells that facilitate MICP-mediated self-healing, needs protection from the harsh environment, which can be accomplished by immobilization [56]. Wang et al. [30] utilized B. sphaericus in the form of microencapsulated bacterial spores Ghosh et al. [57, 58] used anaerobic microorganisms (Shewanella species) directly in cement mortar matrix via mixing water for MICP which showed a positive effect on compressive strength, decreasing its porosity whereas the application of E. coli doesn't show

Table 2 Different microorganisms and nutrients used to create CaCO<sub>3</sub> in concrete/mortar matrix

Precipitation mechanism	Microorganism	Nutrients and added chemical	References
Bacterial metabolic conversion of organic acid	Bacillus pseudofirmus B. cohnii	Calcium lactate, calcium acetate, yeast extract and peptone	Jonkers et al. [25]
	B. cohnii	Calcium lactate and yeast extract	Sierra-Beltran et al. [32]
	Sporosarcina pasteuri Bacillus cohnii Bacillus halodurans Bacillus pseudofirmus	Sodium-citric acid/Peptone, yeast extract, sodium bicarbonate,	Jonkers and Schlangen [63]
	Bacillus strain B2-E2-1	Calcium lactate	Jonkers [6]
Ureolysis	Bacillus alkalinitrilicus	Calcium lactate and yeast extract	Wiktor and Jonkers [26]
	Bacillus sphaericus	Urea and calcium chloride	Achal et al. [33]
	Bacillus sphaericus	Urea, calcium chloride, calcium nitrate and yeast extract	Tittelboom et al. [28]
	Bacillus sphaericus	Urea, calcium chloride and calcium acetate	Muynck et al. [31] Muynck et al. [47] Muynck et al. [48]
	S. pasteurii Pseudomonas aeruginosa	Urea and calcium chloride	Ramachandran [45]
	Sporosarcina pasteurii	Urea, Yeast extract, Tris-buffer, Ammonium Sulfate, Calcium chloride	Wang et al. [74]
	S. pasteurii B. Subtilis	Urea, Peptone, meat extract and calcium chloride	Nosouhian et al. [60]
	Bacillus sphaericus	Urea, Yeast extract, and calcium nitrate	Wang et al. [30]
	Bacillus pasteurii (DSM 33)	Beef extract, Peptone, urea, agar	Qian et al. [62]
	Bacillus megaterium	Urea, Beef extract, yeast extract, Wheat Bran	Krishnapriya et al. [53]
	Bacillus megaterium	Peptone, yeast extract, beef extract, sodium chloride, Agar, Urea, Calcium Chloride	Kaur et al. [13]
Carbonic anhydrase	Bacillus megaterium	Peptone, yeast extract, beef extract, sodium chloride, Agar, $CO_2$ influx	Kaur et al. [13]
Denitrification	Diaphorobacter nitroreducens Bacillus sphaericus	Urea, calcium formate, calcium nitrate, yeast extract	Ersan et al. [69]
	Alkaliphilic bacteria	Sodium-silicate, sodium-gluconate, Calcium-nitrate	Wiktor and Jonkers [67]
Oxygenic photosynthesis (by Cyanobacteria)	Synechococcus PCC8806	Calcium chloride	Zhu et al. [71]
	Synechocystis sp. PCC6803 Synechococcus sp. LS0519 Synechococcus sp. PCC8806	sodium bicarbonate, calcium chloride	Zhu et al. [72]
Microbial-induced ${\rm CO_2}$ capture precipitation	Bacillus mucilaginosus	Yeast extract, Tryptone, Sodium chloride, Calcium hydroxide, $\mathrm{CO}_2$	Wang et al. [74]

such increment in mortar strength. However, Schreiberová et al. [59] studied the effect of different nutrients for
MICP in cement mortar from past literature and concluded in their study that urea, calcium formate, calcium
nitrate, and calcium lactate have the ability to improve
compressive strength, while yeast extract resulted in a
significant decrease in compressive strength. The utility
of concrete surface treatment by calcium carbonate precipitation using a two-step immersion technique was first
investigated [36, 44, 47] using pure *B. sphaericus* strains
and ureolytic mixed cultures. The mortar/concrete specimens were submerged in a nutrient solution for 72 h after
being soaked in stock culture for 24 h. The results showed
that the calcium carbonate deposition on the specimens'

exteriors, reduced the capillary water uptake along with the permeability towards gas. With higher  $CaCO_3$  precipitation, the rate of reduction in water adsorption of the bacteria-treated specimen also increases [38, 44, 47, 48]. Nosouhian et al. [60] chose a two-step treatment using bacteria in their research, to investigate concrete durability improvement in a sulfate environment. In MICP, the rate of precipitation and the presence of organic substances also affect the density and cohesion [61]. Qian et al. [62] implemented a bacterial treatment consisting of only a single step, by submerging cement stones in the medium containing urea, *S. pasteurii* cells, and calcium nitrate. The precipitated dense and coherent layer (150–290  $\mu$ m thick) of carbonate performed very well

in an acidic environment and in decreasing the waterabsorption rate by at least 50%. Jonkers and Schlangen [63] used alkaliphilic spore-forming bacteria (Sporosarcina pasteurii, B. cohnii, B. halodurans, B. pseudofirmus) with different carbon sources, for MICP in concrete cracks. Studies showed that by incorporating high numbers of bacteria (109 cells cm<sup>-3</sup>) and using amino acids aspartate and glutamate (0.5% of cement weight) in the concrete matrix, MICP performs satisfactorily in crack healing, sustaining the compressive, tensile and flexural strength of the concrete with the minimal loss [63]. MICP by means of ureolytic bacteria (mainly from the genus Bacillus) has proven useful in various studies for concrete crack healing [28, 64] with regaining strength [25, 45, 46] as well as decreasing the material permeability [47, 48] in the process. Achal et al. [65] utilized the MICP of calcite on reinforced concrete using the bacterial strain of Bacillus sp. in urea hydrolysis utilizing corn steep liquor (CSL) (a high protein-containing industrial by-product) as the source of nutrient in the reinforced concrete. Kaur et al. [13] investigated the effect of utilizing CO<sub>2</sub> by Bacillus megaterium as an alternative to urea in ureolysis, by using CO<sub>2</sub> influx (20 ml/min) in place of urea. They studied and compared the applicability and effectiveness of using CO<sub>2</sub> influx as well as urea in CaCO<sub>3</sub> precipitation for concrete crack healing. The precipitated CaCO<sub>3</sub> through CO<sub>2</sub> influx is equivalent to that produced when 2% urea was used. Carbonation curing improves the strength by forming calcium carbonate crystals when reaction of CO<sub>2</sub> with the hydrated and un-hydrated products of cement hydration takes place. These crystals form the microstructure and pore structure of the mortar, eventually improving properties such as moisture transport, mechanical strength, and CO<sub>2</sub> diffusivity. Therefore, carbonation makes the concrete more durable because it makes the concrete totally dense, decreases the whole porosity, and increases the sulphate and alkali aggregate resistance [13]. Even, the strength of concrete increased by 117% for accelerated carbonation curing while the increment was only 47% in urea-treated specimens. Apart from these technical benefits, the use of direct CO<sub>2</sub> influx also helps in CO2 sequestration actively without compromising the performance of the concrete itself.

Literature [56] showed that different biotic as well as abiotic influences, such as bacterial genotype and concentration, nucleation site, the concentration of nutrients (carbon, nitrogen, and calcium source), pH, and temperature, can influence the biosynthesis of calcium carbonate. Instead of urea hydrolysis, few researchers attempted to study the effect of metabolic conversion of calcium lactate or calcium formate i.e., bacterially mediated calcite precipitation for self-healing of concrete cracks which showed successful outcome. Some researchers [26, 32,

37, 66–68] utilized the denitrification pathway for calcitebased mineral precipitation using alkaliphilic bacteria for crack repair in concrete which also resulted in effective frost salt scaling. Few researchers [67, 69] investigated both the efficiency of alkaliphilic bacteria (B. sphaericus) for ureolysis and denitrification bacteria (D. nitroreducers) for denitrification to precipitate carbonate for self-healing of concrete in different nutrients. Although the use of denitrification bacteria has been tested and referred by various researchers for carbonate precipitated self-healing of concrete crack, still it possesses some major drawbacks such as the release of ammonium as a by-product which possesses an unpleasant smell. The use of organic compounds by heterotrophic bacteria releases CO<sub>2</sub> in the process. Although the by-product CO<sub>2</sub> later takes part in carbonate precipitation, still there is a possibility of release of the by-product CO<sub>2</sub> in the atmosphere causing the opposite effect of CO2 arrestation leading to global climate change. Therefore, to avoid this possibility, researchers shift their focus on MICP using photoautotrophic cyanobacteria that obtain energy through photosynthesis, grow in a nutrient-poor medium while using CO<sub>2</sub> as a carbon source, and release O<sub>2</sub> in the process [70, 71]. Cyanobacteria's calcification activity is affected by environmental conditions [72] such as light intensity and UV pre-treatment. The most used cyanobacteria for this application are of genus Synechococcus [72]. Using these cyanobacteria, a new type of building material known as Living Building Material (LBM) is also proposed in a recent study [73]. Using the photosynthetic cyanobacterium Synechococcus sp. PCC 7002, in an inert structural sand-gelatin scaffold to toughen the hydrogel matrix via CaCO<sub>3</sub> precipitation, using controlled temperature and humidity atmosphere, from one parent generation of LBMs, the authors ensured three successive regenerations. This investigation opens a new door to the self-generation of building materials (Frankenstein-type material) that also supports actively CO<sub>2</sub> sequestration. UV tolerance of the strain is also an important aspect since these microbes have to stay in the concrete and work there which in most cases are exposed to direct sunlight. Recently a few researchers [74] also investigated the effects of MICP on the properties of recycled aggregates (RA) based on two different methods, i.e., urea decomposition precipitation system and CO<sub>2</sub> capture precipitation system. Results showed that the direct immersion treatment of the RAs under urea decomposition precipitation system, had the best modification performance. Although the overall modification performance of urea decomposition precipitation system, was superior than that of the CO2 capture precipitation system, still the latter method offered a greener environmental approach due to its effective contribution in CO<sub>2</sub>

capture and immobilization. Some researchers [75] also reviewed the current state of utilizing the MICP process in application of soil stabilization. Some other investigators [76] reviewed the works on utilization of MICP on solid waste treatment and soil remediation to understand the present scenario of it. They concluded the importance of MICP technology in application in soil improvement factors. Thus, the process also implements in  ${\rm CO}_2$  sequestration and helps for an environmentally friendly future. Also, in the field of solid waste treatment and soil remediation, application of MICP technology has great potential.

This review study unveils the lacunas in the present canvas for the exact application of the utilization of MICP process in concrete crack-healing and sequestration of environmental CO2 in the process. Although, there are numbers of work done concentrating on concrete crack repair using MICP process with very effective and approachable outcomes, still only a very few investigators have focused their study to utilize MICP in concrete crack repair for direct CO2 sequestration from its gaseous streams [13, 74]. In the same aspect, the selection and applicability of suitable organism as well as the conditions for the favourable growth of the organism and its pathway is yet to be investigated. The studies on the viability of the microorganism in the real field application is very limited. Furthermore, the performance of the repaired concrete has to be assessed properly.

#### 2.6 Gaseous CO<sub>2</sub> sequestration

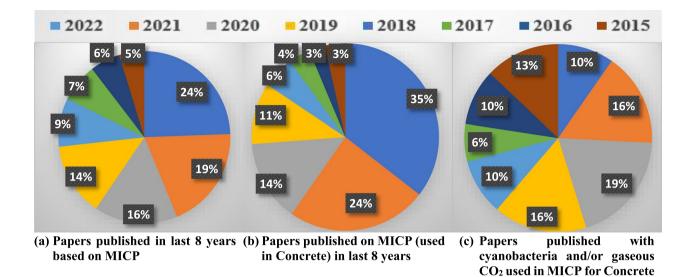
The applications of MICP in the self-healing of concrete cracks have great potential. Mainly ureolysis and bacterially mediated calcite precipitation, as well as oxygenic photosynthesis by cyanobacterial, showed promising results in the scenario. MICP using ureolysis pathway resulted in the production of CO<sub>2</sub> as a by-product in the middle of the reaction, later it was used to form the calcium carbonate precipitate, which in turn decreased the contribution of CO<sub>2</sub> arrestation of the process. Rather, this phenomenon can negatively affect as the release of CO<sub>2</sub> in the atmosphere can affect the greenhouse effect resulting in global warming. Few researchers [77] have noticed that ureolysis by S. pasteurii resulted in no net CO<sub>2</sub> sequestration, since the moles of calcite precipitated were equal to or less than the moles of urea-derived carbonate ions, due to  $\mathrm{CO}_2$  generation during the urea hydrolysis process. But to utilize the same ureolysis in a positive way for CO<sub>2</sub> sequestration, Kaur et al. [13] replaced urea with CO2 influx which resulted in a positive outcome for crack healing. This process can therefore be further utilized for CO<sub>2</sub> sequestration while self-healing of concrete cracks by the means of ureolysis replacing the urea to sufficient CO2 influx. Moreover, using the oxygenic photosynthesis by cyanobacteria always showed positive results for carbonate precipitation, as the cyanobacteria utilize  $\mathrm{CO}_2$  as an alternative inorganic source due to the low concentration of the bicarbonate. This brings the environmental advantage of carbon sequestration. The study and research conducted solely on MICP, as well as utilization of MICP in concrete and by the means of cyanobacteria or utilizing gaseous  $\mathrm{CO}_2$ , has been evaluated approximately for the last 7 years and shown in Fig. 2.

#### 3 Conclusions

The potential of CO<sub>2</sub> sequestration by microbial selfhealing concrete via the MICP technique indicated it to be an effective solution to global warming. In addition, it also positively affects the mechanical properties of concrete. In the current scenario, the huge production of CO<sub>2</sub> from the cement factory, which imposes a great threat to the environment, can be utilized in these MICP mechanisms for concrete crack healing. This also can act as a means of environmental CO<sub>2</sub> sequestration, although the MICP mechanisms having its own merits and demerits. It is already an established truth that the mechanism of ureolysis, as well as oxygenic photosynthesis by cyanobacteria, utilizes the atmospheric CO<sub>2</sub> in the MICP process. MICP by means of replacement of urea to CO<sub>2</sub> influx can effectively contribute to CO2 arrestation from the atmosphere. Moreover, the mostly used pathway, i.e., using cyanobacteria also bears the potential of repairing concrete cracks alongside the major contribution to CO<sub>2</sub> arrestation in the process. However, the applicability of optimum conditions as well as of effective microorganisms and its pathway needs more rigorous study to establish a mechanism of MICP for concrete crack healing which will also act as an effective solution in atmospheric CO<sub>2</sub> sequestration. Even so, it can be concluded that MICP can bear a significant role in arrestation as well as sequestration of CO<sub>2</sub>, under proper atmospheric conditions with a cautious selection of microorganisms and its nutrient for the MICP procedure.

Thus, we believe that future MICP research have a huge potential with emphasis on microbial repairing of concrete and its role in  ${\rm CO_2}$  sequestration. The reasons being:

- Very few studies have been conducted till now regarding the active utilization of CO<sub>2</sub> from the atmosphere/environment leading to its sequestration in MICP, used for concrete crack-repair.
- For different types of concrete using different materials and for its improved performance as well as its crack-repair process, incorporating the CO<sub>2</sub> sequestration via MICP from direct atmosphere is a bril-



**Fig. 2** Pie chart on approximate research works done on MICP and its utilization on concrete healing for the last 8 years, where **a** represents the overall papers published on MICP, **b** represents the percentage of paper published on MICP which is used in Concrete, and **c** represents paper published specially with cyanobacteria and/or gaseous CO<sub>2</sub> used in MICP for Concrete

liant opportunity. Bacteria with high effectiveness in solidification as well as with higher environmental adaptability, should be cultured, cultivated, with the related costs taking into the consideration.

- 3. Researches on utilization of solid waste having huge amount of calcium should be explored for extracting the metal through MICP.
- 4. To adapt MICP in concrete crack repair with effective CO<sub>2</sub> sequestration from the environment itself in the process, investigators have a great area of future work to find the suitable and effective organism, pathway as well as conditions for the favourable growth of the organism.

Although the present review work has been done vigorously to project the exact current scenario for the utilization and contribution of MICP in concrete crack repair, still there is a scope for more investigations on the use of MICP in concrete crack repair, while sequestering atmospheric CO<sub>2</sub> simultaneously.

#### 4 Recommendation

The authors strongly recommend detailed experimental investigation on the use of atmospheric  $\mathrm{CO}_2$  in MICP for concrete crack healing, with consideration and implementation of various factors and parameters required for the same. Thereafter, the performance of the repaired concrete in practical field is also needed to be evaluated. On successful outcome, a viable solution for a greener earth can be advanced. Along with the concrete repair process, in the

field of solid waste treatment and soil remediation, application of MICP technology for atmospheric CO<sub>2</sub> sequestration has great potential, which can also be studied via proper and detailed experimental process in the same way for a greener and sustainable environment.

in last 8 years

#### **Abbreviations**

MICP Microbially induced calcite precipitation

CSL Corn steep liquor RA Recycled aggregate

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#### **Author contributions**

AB performed a major contribution in researching the past work done on this topic and writing the article. AS and DG contributed to studying and preparing the PI Chart for the manuscript. BD helped with the diagram of MICP in the manuscript as well as thoroughly guided the overall manuscript. SD and AKS contributed in the area of all the chemical reaction parts and the concrete part respectively. Overall, all authors read and approved the final manuscript.

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