QUATERLY OF ENVIRONMENTAL ENGINEERING AND DESIGN
Vol. 172(52) 2018

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USAGE OF CALCIUM CARBONATE BIODEPOSITION IN MODIFICATION OF CEMENTITIOUS COMPOSITES

Summary

Biodeterioration of construction materials is an undesired phenomenon, generating high costs of constraction repairs. On the other hand, occurrence of some bacteria can affect prevention and self repair of fractures formed in concrete. Biodeposition is an effective solution for increasing compressive strength of concrete, extending durability of concrete constructions and renovating limestone elements in facades of historic buildings.

Key words: biodeposition of calcium carbonate, ureolisis, self-healing of concrete

INTRODUCTION

Concrete is one of the most often utilised construction materials. Despite its compressive strength it is almost 10 times less resistant to stretching. When tensile stresses occur and they exceed strength of the very material (cement matrix), then fracturing of concrete starts. A hope for self-healing of concrete are bacteria. Influence of microorganisms on concrete constructions has been known for a long time, mostly because of biologically conditioned corrosion of concrete (Microbial Induced Concrete Corrosion, MICC) elements in infrastructure of disposal

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and treatment of urban wastewater by bacteria of the genus Thiobacillus sp. [Parker, 1945]. Annual losses caused by influence of the mentioned microorganisms could be even in billions of dollars [Hewayde and others, 2007]. Besides, costs related to conservation of concrete before intensive progress of corrosion in conditions of wastewater impact have to be born already at the stage of building new elements. According to Jasiczak they can constitute even up to 20% of investment costs. It is caused not only by the necessity of obtaining concrete with appropriate parameters (such as resistance to frost F>150, waterproofness W6-W8, water penetration depth <3 cm at W8), however – especially in case of large containers – the need to use strand protective layers in a zone of changing sewage level, where microorganisms contribute to releasing of compounds such as hydrogen sulphide in the course of fermentation processes [Jasiczak, 2006]. However, influence of microorganisms not always has to be destructive and burdened with high usage costs. Bacteria can have a very positive effect on sustainable development in the areas of construction and cement industry. The thing that should be mentioned here is most of all the concept of biodeposition of calcium carbonate, also known as Microbiologically Induced Calcite Precipitation, MICP).

PRESENTATION OF THESIS STATEMENTS

This project presents literature overview regarding physical conditions which are favourable for creation of calcite on the surface of bacterial cell walls for the purpose of using this phenomenon in self-healing of fractures in concrete. Those are i.e. temperature, pH value or components of culture medium containing appropriate quantity of urea and calcium salts. The project also presents potential usage of biodeposition (by adding bacteria to concrete) for the purpose of strengthening compressive resistance of concrete and enabling its self-healing. Methods of adding bacteria to concrete were also presented.

SUBJECT DEVELOPMENT AND ITS ANALYSIS

Microbiological biodeposition of calcium carbonate is based upon utilising abilities of some bacteria to simultaneously perform urea hydrolysis (ureolysis) and physical adsorption of calcium ions. Therefore those bacteria somewhat act as seeds of crystalysation of calcium carbonate which forms on the outer surface of their cell walls. Among biodepositional bacteria the most commonly known are Bacillus sphaericus and Sporosarcina pasteurii (also known as Bacillus pasteurii). Biodeposition is a consequence of ureolysis, as a result of which urea hydrolysis occurs, resulting in releasing of carbonate ions CO_3^{2-} which in turn reacting with ions Ca^{2+} provided from the outside (regardless of urea) and adsorbed on a cell wall, create calcium carbonate (Figure no.1 from De Muynck and others [2008]), as in reaction [Chunxiang, 2009; Siddique and Chahal, 2011]:

$$CO(NH_2)_2 + 2H_2O + Ca^{2+} \rightarrow 2NH^{4+} + CaCO_3 \qquad (1)$$

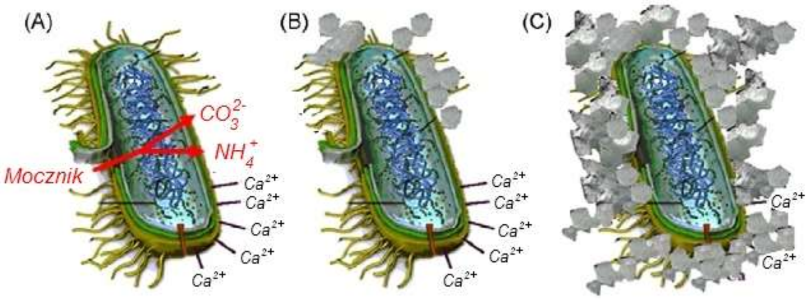

Fig. 1. Schematic of calcium carbonate biodeposition process: A – ureolysis and adsorption of calcium ions on cell walls, B – commencing of biodeposition process, C – cell wall surrounded with calcium carbonate crystals [De Muynck and others 2008]

Hydrolysis process, apart from providing a cell with ATP (adenosinetriphosphate, needed for obtaining energy necessary for removing ions CO_3^{2-} from the cell), is utilised for another purpose – the solution is supplemented with carbonate ions and ammonium ions which increases the pH value in the solution surrounding the cell. It has great importance in the process of biodeposition of calcium carbonate, because along with increasing of pH value easier precipitation of it is possible. The pH value also decides about intensity of urea hydrolysis process. Most favourable conditions for urea hydrolysis occur in the temperature of 40°C and at pH = 7. Urea hydrolysis and calcium carbonate precipitation thus occur in different conditions. The sodium-potassium pump (Na^+/K^+ ATP-asis), should also be mentioned as it has significance in calcium carbonate precipitation also inside the cell. Together with sodium ions, CO_3^{2-} the ions created in the urea hydrolysis process are discharged outside the cell. On the other hand, not only potassium ions enter the interior, but also calcium ions which can react with carbonate ions remaining inside the cell. In this way intercellular forming of calcite occurs [Watabe, 1974]. A source of carbonate ions could be not only hydrolysed urea, but also inorganic salts of well soluble, dissociable carbonate compounds. However, urea is necessary for functioning of a bacteria cell in the course of biodeposition process – most of all for activation of urease and production of ATP.

According to Okwadha and Li [2010] factors decisive in effectiveness of biodeposition are: type and concentration of bacteria in the analysed material, initial urea concentration, temperature of reaction, initial calcium ions concentration, ionic strength and pH value of the solution. It has been concluded that the optimal conditions for the Sporosarcina pasteurii strain are values equal to 666 mM for urea concentration and 250 mM for calcium ions concentration. However, density of bacterial solution should be $2,3 \cdot 10^8$ cells/dm^3 of the solution [Okwadha and Li, 2010]. However, other researchers [Nemati and Voordouw, 2003] determined maximum concentration of urea and Ca^{2+} ions for which maximum quantity of produced $CaCO_3$ is reached, expressed per 1 dm^3 of solution (36 g for $CO(NH_2)_2$ and 90 g for Ca^{2+}). Stocks-Fischer and others [1999] also conducted an attempt to determine the optimal conditions, recognising that for the value similar to cell concentration provided by Okwadha and Li [2010] equal to $2,3 \cdot 10^8$ cells/dm^3, the lowest consumption of calcium ions occurs. On the other hand, the highest intensity of ureolysis (which is measured in concentration of NH_4^+ ions) with simultaneous effect of calcium carbonate precipitation occurs between population of 10^6 and 10^7 cells in 1 dm^3 of microbiological solution [Stocks-Fischer, 1999]. With greater density urea is consumed faster, which in very high concentrations could lead to a situation where number of cells conducting ureolysis is declining. As a result complete deceleration of calcium carbonate precipitation can occur at population close to 10^9 cells in 1 dm^3 [Stocks-Fischer and others, 1999].

Essential for progress of the process is also its temperature. According to Nemati and Voordouw [2003] and also Mitchell and Ferris [2005] its increase from 10°C to 15°C causes double rise in ureolysis speed, but increase by another 5°C increases the speed five times. Microbiological process is therefore more efficient than chemical processes for which – as it is known, according to van't Hoff's rule – increase in temperature by approx. 10°C usually causes double increase in the process speed. On the other hand, the pH value optimal for growth of Sporosarcina pasteurii is 7–9 which has been confirmed in research of chosen species of utility microorganisms by Leejeerajumnean and others [2000]. Sporosarcina pasteurii differs from other researched strains which gradually lose their activity as the pH value rises. In reality, as it was mentioned before, during decomposition of urea a bacterium provides itself with appropriate pH value, supplying ammonium ions which regulate pH. However, in case of the mentioned strain the biggest tollerance to changes in pH value occured. On the other hand, in research [Annamalai and others 2012] in case of initial medium reaction of pH value 7,5, process of biodeposition progressed most effectively (the author did not provide information on what the pH value of the solution was in the final phase of calcium carbonate creation).

Apart from providing optimal physical conditions necessary for the progress of biodeposition reaction, it is also important to supply appropriate breeding ground serving as a source of amino acids and carbohydrates for the bacteria. As the breeding ground for the first stage it is suggested to use e.g. a set composed of peptone and yeast extract, and also urea in some versions. On the proper stage of biodeposition urea should be used again (regardless of if it was addend before). Another ingredient should be one of the well soluble calcium salts: calcium chlorine ($CaCl_2$), calcium nitrate ($CaNO_3$) or calcium acetate ($Ca(CH_2COO)_2$). Exemplary research in calcium [Chunxiang, 2009] concludes that the best effects, measured in thickness of the obtained calcite layer and crystal morphology, are achieved at concentration of 0,3 moles of Ca^{+2}/dm^3. However, in conclusions from the mentioned project its author emphasizes that various aspects of dosing salt require further research. When it comes to the layer of precipitated biocalcite, the best effects were obtained with calcium nitrate [Chunxiang, 2009]. However, in researches [De Muynck and others, 2008], where nitrate and calcium acetate were compared it was observed that the first of the salts contributes to forming of crystals with rhombohedral shape, but gritty crystals (possibly vaterite) are formed as a result of using the other one.

Apart from the mentioned strain Sporosarcina pasteurii also other bacteria can be utilised in biodeposition processes. De Muynck and others [2008] pay attention to the Bacillus sphaericus strain. They conclude that efficiency of modifications in concrete with the use of bacteria is comparable to modifications with the use of water impregnating agents. According to data of De Muyncki and others [2010] the cost of biodeposition in European countries is also similar to traditional solutions and varies from 15 to 40 €/m2 (of modified surface), depanding on the degree of damage to an element, on which biomodification is performed. However, Whiffin indicates that the cost can be limited, if waste products are used. The authors also emphasize the fact that in efficiency of crystalising calcium carbonate not only the strain and the calcium salt are important elements, but also the breeding ground [De Muyncki and others 2010; Whiffin, 2004].

For the first time biodeposition was used for protection of decorative stone [De Muynck and others, 2010]. The formed $CaCO_3$ functions as protection from degradation caused by atmospheric factors, such as acidic rainwater and changing temperature conditions (freezing and defreezing). Moreover, bacterial layer of calcium carbonate strengthens the material itself, as it brings loose elements together, which decreases water permeability and its potential for ingress into pores of the stone. This means improvement of durability and freeze resistance of surface on the material covered with bacteria and enhanced resistance to penetration by chlorides, and also carbonation [Snoeck and others, 2017].

The concept of biodeposition of calcium carbonate, despite of it still being in the phase of experimental research, is recognized as possible for utilising in areas mentioned by Okwadha and Li [2010], referring to works of other authors. Those are, among others: sealing of construction cracks, increasing resistance to compression of concrete, extending the durability period of structures, including the concept of self-healing concrete, ground stability and sand consolidation, renovating elements of monumental buildings made of limestone, in the processes of oil extraction (selective sealing) and in the processes of wastewater cleaning.

In reference to applications related to renovation and strengthening concrete two separate groups of research can be separated from those mentioned above. They are focused on structural modification (the concept of self-healing concrete, increasing of durability) surface modification: sealing of cracks, modification aimed at lowering permeability of concrete.

One of applications of the biodeposition method is the structural modification of concrete, that is utilisation of microbiological consortium (single bacterial strains or mixed bacterial cultures) as an additive to concrete [Chahal and others, 2012a]. According to researchers the biggest increase in resistance to compression was achieved in case of applying *consortium with density* of 105 cells/dm^3 – 22% in reference to control samples. For series in which 10% of the cement mass was replaced with fly ash, absorbability decreased four times. *Chloride permeability was also significantly decreased* (measured through the method used by ASTM – RCPT – Rapid Chloride Permeability Test). The researchers mentioned above conducted similar tests with the use of microsilica, as a substitute for a part of cement. This time the best results were achieved at concentration of consortium 106 cells/dm^3, however the effect proved to be lower (by about 50%) than in the case of samples with fly ash. *Permeability* in the RCPT test has been limited to a similar degree. Decrease in absorbability of samples proved to be more significant – in all series with microsilica, at two stages of cement replacement: 5% and 10% it was limited a few times, to a higher degree than in case of the tests with use of fly ash [Chahal and others, 2012b].

According to Jonkers and others [2010], developing of the concept of self-healing concrete, consisting in instant sealing of cracks created during the usage of construction, matches the actions leading to limiting of anthropopressive impact of the cement industry on the environment, because extending of durability period of concrete or reinforced concrete elements is related to reduction of cement manufacturing for the purpose of producing new elements, replacing the used ones. Research by Nasiru and others [2016] which was performed as well on isolated as mixed cultures of bacteria have confirmed that fractures and cracks can be effectively elliminated through metabolic activity of bacteria precipitating calcium carbonate in their surrounding. In the research of Bang and others [2001] it was concluded that the concrete on which self-healing biodeposition was performed does not show changes when it comes to such parameters as modulus of elasticity or tensile strength. Simultaneously, as a result of filling cracks with biocalcite, rise in compressive strength occurs. Van Tittelboom and others [2010] have shown that the effects of self-filling of cracks are the best when a bacteria strain is placed in the structure of silica gel. In such an instance the gel functions as protection from too high pH value of water in pores of the concrete, and only later (in the process of seling cracks) deployment of bacteria contained in the gel occurs.

Covering concrete with biolayer of calcite leeds to decreasing of its permeability to gaseous and liquid media. De Muynck and others [2010] provide as a practical example the rise in resistance of concrete to carbonisation. Increase in impermeability and, as a consequence of it, decrease in susceptibility to reaction with CO_2, also ensures protection of concrete from the phenomenon of release of chlorides which are bound in the form of Friedel's salt, which occurs just as a result of carbonisation process. This sort of protection can be justified in case of recycling aggregates of unknown origin about which there is a suspicion that it could decrease the quality of newly produced concrete [De Muynck and others 2010]. Potential durability of biocalcite layer and its beneficial influence on durability of concrete results from the properties of calcium carbonate. As it is known, as long as it still has quite low pH value, carbonate can effectively neutralise influence of acids. It has been confirmed experimentally in the research of Chunxiang and others [2009].

In case of suspicion that the environment in which a concrete element is exploited endangers its durability, actions aimed at limiting that impact are always taken. In normal conditions there is almost always influence of CO_2 which neutralizes concrete, and the environment with pH value below 6,0 becomes aggressive towards concrete. Bacterial biolayer increasing resistance to deep diffusion of CO_2 can also be a way to decrease migration of chlorides from the environment. The physical function of biolayer is revealed here. Its activity is analogous as in case of impregnating parameters. The matter of the very water permeability of biocalcite layer is disputable. Pure calcite has high surface energy and due to this it is hydrophilic [Ioannou and others, 2004], although very weakly soluble in water (14 mg/dm^3 at 20^0C). However, the result of water retention in case of incomplete coverage of the surface with biocalcite can seemingly reduce the sealing effect, which could be incorrectly interpreted as low degree of biolayer efficiency. Supposedly, it has some significance in research of absorbability in grains of recycling aggregates. Extensive grain surface, contributing to physical binding of water, can be decisive in the result of absorbability of the researched agregates, as the entire water mass (measured as absorbed water) in fact does not penetrate the structure of grinded concrete, yet it remains on the surface of the formed biocalcite layer.

Beyond the debatable questions mentioned above, it is essential to obtain very hermetic layer of biocalcite or decreasing the surface energy by adsorbing other ions [De Muynck and others, 2008]. It is known that calcium carbonate (with already mentioned high surface energy of the bond) has the ability to intercept ions from the surrounding environment. As an example can serve the bond between ions of copper or adsorption of silver [Srikanth and Jeevanandam, 2009]. Haselbach [2008] also presents a hypothesis about overstoichiometric chemisorption of CO_2 on the surface of calcite in presence of water. Predicting further applications in regards to the second conception, biocalcite layer with adsorbed ions of silver, which is attributed with antibacterial properties, can prove to be an effective method of delaying corrosion caused by bacteria of the genus Thiobacillus sp. In this case calcite biolayer with adsorboed ions of silver could act as protection barrier against the process of carbonation and also function as bactericide.

In research the form of dosing bacteria to concrete is also analysed. Cyted in the work of Nasiru and others [2016] results of research conducted by different authors indicate that bacteria can be introduced to concrete not only through spraying onto the concrete surface, but also by direct injection in its pores. Another way of dosing bacteria is encapsulation allowing to protect microorganisms from an environment of high alkalinity which is lethal for them. Interesting technology of self-healing concrete was researched by Jonkers [Jonkers, 2017 and Jonkers 2011 from Zając and Gołębiewska, 2016], as instead of producing calcium carbonate through ureasis, he suggested using calcium lactate ($Ca(C_3H_5O_2)_2$) as a breeding ground for bacteria. It induces a completely different biochemichal process of producing calcium carbonate that is insoluble in water, because in presence of alkaline environment metabolism of bacteria induces the following chemichal reaction to calcium lactate [Jonkers, 2017 and Jonkers 2011 from Zając i Gołębiewska, 2016]:

$$Ca(C_3H_5O_2)_2 + 7O_2 \rightarrow CaCO_3 + 5CO_2 + 5H_2O \tag{2}$$

Yet, it does not finish the process of producing ($CaCO_3$), as particles of carbon dioxide (CO_2) created as a result of bio-reaction react with particles of portlandite ($Ca(OH)_2$) appearing in concrete, producing $CaCO_3$ [Jonkers, 2016].

A side product in this process is pure water chich can additionally react with the remaining non-hydrated grains of cement in concrete. Jonkers [2011] in his research applied copporite capsules of diameter 2÷4 mm. This system of concrete regeneration is burdened with some imperfection, because for the capsules with bacteria and breeding ground to fulfill their task in repairing fractures and cracks in concrete they should constitute about 20% of volume of the concrete – due to the randomness of occurring of fractures in concrete in relation to random distribution of capsules [Zając and Gołębiewska, 2016].

The article by Jonkers [2017] presented the idea of repairing concrete by implementation of bacteria along with PVA fibers (polyvinyl alcohole), which is also effective in use. The fibers contribute to reducing shrinkage of the concrete and improve adhesion between the applied repair mortar and the old concrete. On the other hand, bacteria, in the described process of metabolism with breeding ground in the form of calcium lactate, close micro-fractures in concrete, thus improving properties of the material. Other examples of applying bacteria encapsulation in self-healing of concrete is the suggestion of Wang and others [2014] according to which capsules with bacteria are placed in a hydrogel coating and only then inserted to fresh concrete mixture, and also the method researched by another group of scientists in which bacteria are sealed in graphite nano flakes [Nasiru and others, 2016]. Both methods are highly efficient in regenerating wide fractures, because in case of the first method it was proven that sealing of fracture of 0,5 mm in width occurred together wih reduction of water absorption by 68%. In turn, in the second method the fracture of 810 μm in width was ultimately healed after seven days. It should also be noted that capsules with bacteria in the form of graphite nano flakes have a significant adventage, as they can be equally distributed and thoroughly dosed in concrete mass. They also improve durability of concrete which rises after using nano flakes by almost 10% over the referentil concrete and moreover they eliminate fractures at nano level [Nasiru and others, 2016].

Interesting conclusions regarding the influence of biodeposition layer on bonding between existing mortar and new healing material were stated in the work of Snoeck and others [2017]. For the purpose of researching polymorphism of calcium carbonate varieties ($CaCO_3$), produced due to the contribution of bacterial strains of Bacillus sphaericus, they used x-ray diffraction and testing with scanning electron microscope, and then the skew shearing test of samples. The purpose was testing if rise in durability of connection occurs. The results confirmed that bacteria generate varations of calcite i vaterite. The obtained rise in durability for the series of samples covered with bacteria layer, involving 33% of the surface, reached 50% in relation to the referential sample. It was caused by increased roughness of the connection, as the biolayer of about 50 µm in thickness created a mechanical obstruction contributing to rise in durability on the wall [Snoeck and others 2017].

CONCLUSIONS

Research in efficiency of calcium carbonate biodeposition in self-healing of concrete is developing quite dynamically. It attemts to implement various biological methods, those based on the mechanism of urea hydrolysis and starting metabolism of ureasis production in a bacterial cell and its release of calcium carbonate, and those utilising calcium lactate as breeding ground. Combined methods include those suggestions which are based on joining of biological and chemical methods in order to obtain the highest possible effectiveness in self-healing of concrete at the lowest cost possible. Hence the searach for ways of placing bacteria in concrete, e.g. in the form of capsules, copporite, hydrogel or graphite nano flakes and searching for materials contributing to improvement of bonding force beween new regenerative layers and existing degraded surfaces, which is exemplified by the mentioned above PVA fibers. Undoubtedly, further development of research in this area should be expected. Due to it maybe already in the next generation it could be possibile to implement a technology of concrete that is insusceptible to cracking.

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