BACTERIA-BASED SELF-HEALING OF CONCRETE: EFFECTS OF ENVIRONMENT, EXPOSURE AND CRACK SIZE

Kevin Paine [(1)]

(1) BRE Centre for Innovative Construction Materials, University of Bath, UK

Abstract

The effect of water-borne contaminants on the durability of concrete is well-known and cracked concrete is more susceptible to permeation of these contaminants. An approach to autonomic self-healing of such concretes is the utilization of microbiologically-induced calcite-precipitation. This approach uses the metabolic activity of bacteria and biomineral precursors embedded within the concrete to form an inorganic material, usually calcite, as the healing compound. However, bacteria-based healing of concrete creates a number of scientific and engineering challenges at the biology-concrete technology interface. This paper provides a review of previous and on-going research on the use of bacteria-based self-healing of concrete in relation to the problems associated with the setting, hardening and carbonation of concrete and the problems associated with healing large cracks.

1. INTRODUCTION

The effect of water-borne contaminants on the durability of concrete is well-known and cracked concrete is more susceptible to permeation of these contaminants. Consequently, research is attempting to develop concrete that can self-heal cracks; potentially reducing costs of repair and maintenance work on infrastructure projects dramatically. One approach to autonomic self-healing is the utilization of microbiologically-induced calcite-precipitation. This approach uses the metabolic activity of bacteria and biomineral precursors embedded within the concrete to form an inorganic material, usually calcite, as the healing compound. However, bacteria-based healing of concrete creates a number of scientific and engineering challenges at the biology-concrete technology interface. Many of these are in relation to the germination, survivability and growth of bacteria in the myriad of environments and conditions to which concrete is exposed, and to the internal changes that take place within concrete as it converts from a plastic material at early-age to a dense and solid material in its hardened phase.

Within this paper previous research on the use of bacteria-based self-healing of concrete are reviewed in relation to the problems associated with the setting, hardening and carbonation of concrete and the problems associated with healing large cracks.

2. SETTING AND HARDENING OF CONCRETE

2.1 Effects of Concrete on Bacteria

A key consideration in developing bacteria-based self-healing concrete is whether the bacteria can survive in the hostile conditions (dry, alkaline, ….) present in concrete.

A number of researchers have attempted to add live bacterial cells and spores to concrete and determine their viability and survivability. In general it has been shown that the viability of the cells is severely restricted. Jonkers et al [1] have suggested that this is because of early-age processes, and particularly the cement hydration process which exerts compressive stresses that crush the micro-sized cells.

That the viability is compromised by the hydration, rather than the pH, is perhaps confirmed by research in which cells of a non-alkaliphile *Rhodecoccus ruber* were mixed into a magnesium phosphate cement. The resulting paste had neutral pH conditions ideal for the bacterial cells used. Consequently, the bacteria should have been viable, however it was found that this viability only lasted 19 days despite the pH conditions being suitable for survivability of the live cells [2]. Furthermore, Achal et al [3] added *Bacillus megatarium* cells to both a Portland cement (PC) and PC/fly ash paste and showed that more cells survived in the PC/fly ash paste. They assigned this to the greater early-age porosity of the PC/fly ash paste.

Whilst live cells are unlikely to survive the mixing and hydration of concrete, it could be expected that bacteria may be able to survive the early-age processes as spores. This is because endospores have many unique structural features that make them resistant cells. The spore coat is a proteinaceous outer layer that protects the spore from enzymes, mechanical disruption and chemical solvent, whilst the spore cortex contains peptidoglycan, confers resistance to organic solvents and heat and maintains dormancy [4-5]. Small acid soluble proteins protect the DNA in the spore core from UV damage. The spore core contains high levels of dipicolinic acid (DPA); and DPA binds predominantly Ca^{2+} lowering the water content of the core and is thought to confer dormancy and wet-heat resistance [6]. Consequently, bacterial spores are may be able to better withstand the vigorous mixing process and initial compression that occurs during concrete setting.

However, it has again been shown that unprotected bacteria spores will not normally survive for more than a few days in a concrete mix because of hydration processes [1]. For these reasons it has been considered necessary to encapsulate the spores prior to addition in concrete in, for example, lightweight aggregates [7] or microcapsules [8].

2.2 Effects of Bacteria on Concrete

Because the spores are encapsulated prior to addition to the concrete they should not interfere with the setting of the cement or hardening of the concrete. However, should unprotected bacterial spores be inadvertently released into a concrete mix (for example because of the protective membrane rupturing during mixing, placing or compacting) then they should “die” relatively quickly. However, in the short time that they remain viable research suggests that they will have limited effect on the early-age properties of the concrete. Indeed some

researchers, using high proportions of bacteria have shown that they might even increase compressive strength. Thus they may either act as a nucleation point for reactions or, more likely given their size, they precipitate free calcium ions to $CaCO_3$. Ramachandran et al [9] showed that the addition of *Sporosarcina pasteuri* to an otherwise identical concrete increased strength from 55 MPa to 65 MPa; whilst Ghosh et al [10] showed that use of *Shewanella* in a cement paste led to a 25% increase in strength at a w/c ratio of 0.4 at 28 days.

Research by Bundur et al [11] has shown that the inclusion of bacteria (*S. pasteuri*) in a cement paste delays hydration and that concrete subsequently has lower strength at early ages. However, the same degree of hydration and strength will be achieved after 28 days.

Furthermore Bundur et al [11] incorporated dead bacterial cells into cement pastes and it was shown that there was less strength gain than in mortars comprising active cells. This suggests that the cells (active and dead) delay hydration, but that the active cells are metabolically active and this acts to partly overcome the negative effects of their inclusion on hydration mechanisms.

2.3 Effects of Calcium Salts

To date most research into healing of concrete via bacteria action has concentrated on the formation of calcium carbonate, normally in the form of calcite, as the healing compound. Consequently research has considered the use of calcium-based compounds as the precursor.

For urea hydrolysis the source of Ca ions for most research has been from calcium chloride, although more recent work developing self-healing concrete has tended towards calcium nitrate as an alternative (8, 12-14) in order not to introduce chloride ions into the fresh concrete.

If calcium chloride ($CaCl_2$) is used as the precursor then it would probably be necessary to stay within the limits for chloride content permitted in concrete even if it was to be added to the concrete in an initially encapsulated state: on the basis that the release of chloride ions into concrete is a hazard at all ages. The chloride ion (Cl^-) content permitted in fresh concrete is usually limited to around 0.4% by mass of cement [15] in order to ensure that the Cl^- content at the concrete-reinforcement steel interface remains below the threshold Cl^- content required to initiate corrosion of the steel. Whilst some Cl^- will bind with hydration products, predominately calcium aluminium silicate hydrate (C–A–S–H) but also alumina, ferric oxide, monsosulfate (AFm) phases [16], some will remain free in solution. Furthermore as concrete carbonates these chloride containing compounds can become unstable and release the bound chlorides back into the pore solution [17].

Calcium nitrate on the other hand is compatible with concrete and is commonly used as an anti-freeze and setting accelerator, and has also shown to be effective as an anodic inhibitor for defence against chloride induced corrosion in concrete [18]. The use of calcium nitrate in concrete is considered to accelerate hydration both by the nucleating action of its dissolved ions and through a direct reaction with calcium hydroxide in solution to form calcium hydroxynitrate – a mineral with needle shaped crystals that functions as micro-reinforcement for the cement matrix [19]. Indeed, when added directly to concrete for use in the urea hydrolysis pathway calcium nitrate has been shown to accelerate hydration and increase the degree of hydration [8]. This raises a question, however, as to whether sufficient Ca ions are available to form calcium carbonate as a healing compound. When used as a mineral accelerating admixture, calcium nitrate is used as a pure compound at around 6% by mass of

cement. In self-healing concrete, Wang et al [8] used a hydrated form of calcium nitrate (containing around 30% by mass of water) at a dosage of 8% by mass of cement – which is equivalent to a dosage of 5.6% of calcium nitrate by mass of cement.

For metabolic pathways, a number of organic calcium precursors are feasible in general microbiology, of which the most popular has been calcium acetate. Testing by Jonkers et al [1] has primarily considered that the only calcium precursor that may be added directly to the concrete, and not cause a loss of strength is calcium lactate. That calcium lactate does not cause a loss of strength has been verified at the University of Bath [20] where it was shown that it may increase compressive strength when used at percentages up to an optimum of around 1 to 2% by mass of cement (Figure 1).

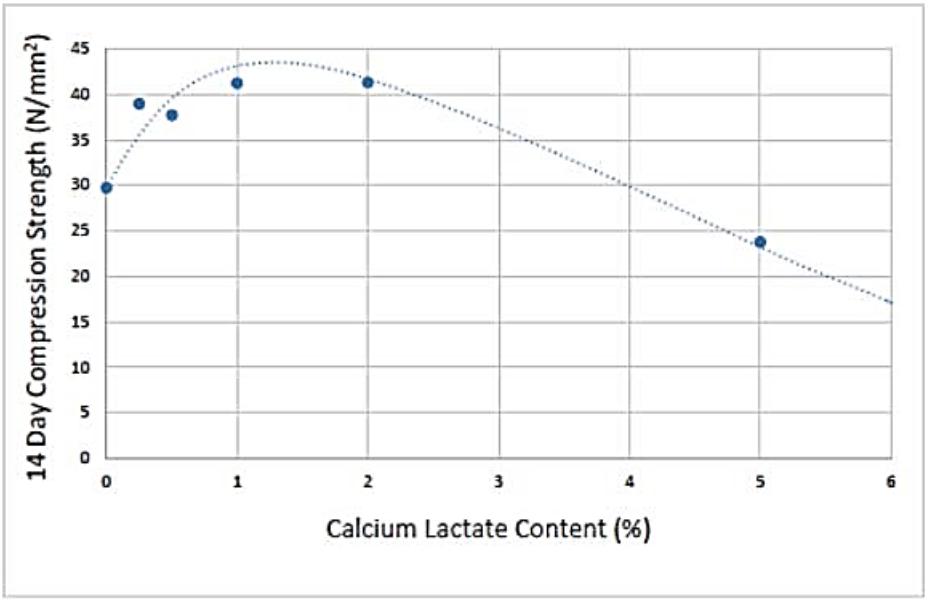

Figure 1 Effect of calcium lactate (% by mass of cement) on compressive strength of mortars [20]

A possible reason for the improvement in performance is that negatively charged lactate ions may adsorb onto the surface of cement grains providing a plasticizing effect and making more cement surfaces available for hydration [21].

Furthermore, Cunniffe [20] showed that calcium lactate may increase initial setting times, but that it may delay final setting (Figure 2).

However, should the calcium precursor be encapsulated prior to addition to concrete as is currently best practice for this pathway then the effect of the precursor on early-age properties could be insignificant unless there was a mass failure of the capsules during mixing or casting. For this reason research at the University of Bath has used calcium acetate as a cheaper and more readily available alternative to calcium lactate [22].

Furthermore, research by Alazhari et al [22] has shown that when used in the proportions that are ever likely to release into the concrete (around 0.3% by mass of cement) that there is no effect on hydration or setting of the cement.

The choice of calcium compound has also been shown to have an effect on the shape of carbonate crystals that precipitate. For example, it has been shown that $CaCl_2$ leads to rhomboidal carbonate crystals whilst calcium acetate leads to spherical carbonate crystals [23]. Although research has suggested that this has no effect on the degree of healing [23], it may be that different polymorphs of calcium carbonate are being formed.

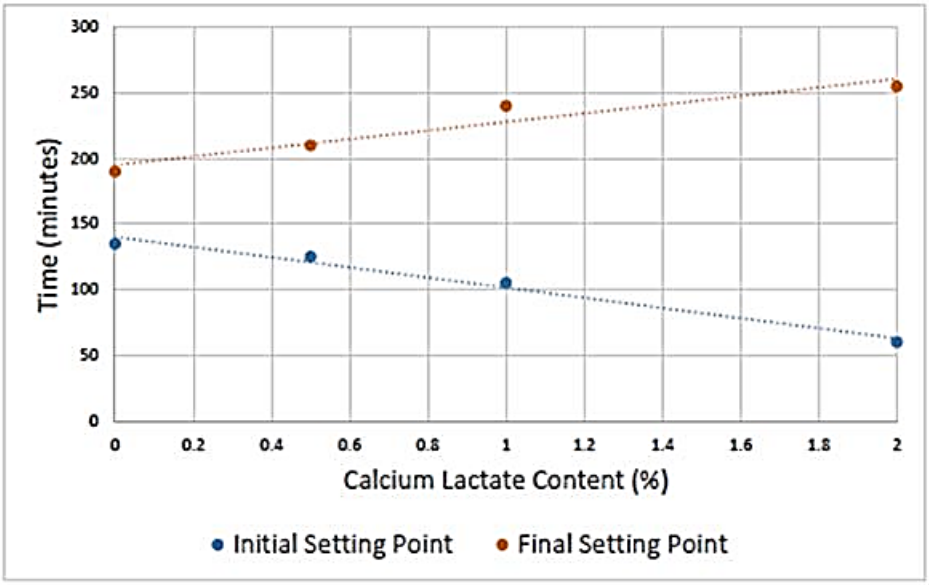

Figure 2 Effect of calcium lactate (% by mass of cement) on setting of cement [20]

2.4 Effects of Nutrients

The nutrients and minerals added to the cargoes to facilitate precipitation of a healing compound need to play a number of roles, the most significant of which are to:

- Aid germination of the bacterial spores.
- Provide a source of growth medium for the bacterial cells.

The choice of these materials is important as they need to be compatible with concrete, primarily in the hardened state and not provide a means for deterioration. As a general rule materials that are known retarders of concrete should be avoided as well as compounds that may deteriorate concrete in the hardened state, in particular chlorides and sulfates. However, it has to be recognised that should the kinetics of germination and precipitation be rapid then the effect of the nutrients on the properties of concrete may not be important should they be rapidly consumed in reactions.

Although recent research is tending towards the inclusion of mineral precursors in protective capsules, in the same way as the bacteria in the earlier section, much research still adds these components directly to the concrete during the mixing process. The effect of these

compounds on the setting and hardening of concrete is therefore important, as is a discussion on their long term availability to the bacteria when healing is required.

For metabolic pathways, many researchers have used a standard growth media combined with a precursor organic compound [24-26]. For example, B4 or modified B4 medium consisting of approximately 0.4-0.5% yeast extract, 0.5% glucose or dextrose, and 0.25%-1.5% calcium acetate. Alternatively a Luria-Bertani broth complemented with calcium acetate has been used – consisting of, for example, 1% tryptone, 0.5% yeast extract, 0.05% calcium chloride and 1% calcium acetate [25].

Urea

In urea hydrolysis, urea is naturally added as a key component of the nutrient feed. The bacteria act as an agent for enzymatic hydrolysis of the urea to ammonia and CO_2 [27]. The drawback of this procedure is the formation of the ammonium ions which results in excessive environmental nitrogen loading [1]. It has also been suggested that the presence of nitrifying bacteria within concrete could convert ammonia to nitric acid leading to an aggressive attack on concrete [28]. However, this has never been observed in self-healing concrete.

Wang et al [8] have suggested that urea [4% by mass of cement] causes a delay in hydration. However, isothermal conduction calorimetry tests undertaken by Bundur et al [11] have shown that urea has very little effect at 0.5% by mass of cement.

Yeast extract

Yeast extract (YE) is commonly used as a source of carbon for the urea hydrolysis pathway [11] and as a source of nitrogen for the metabolic pathway. It also contains other essential minerals. Research shows that YE added directly to concrete delays setting of cement and hardening of concrete. For example, Bundur et al [11] have shown that YE has a significant effect in retarding the hydration of cement. Within a nutrient combination consisting of tris, urea and YE (1% by mass of cement), YE (was the compound with the greatest effect on the kinetics of hydration as shown by isothermal conduction calorimetry. Similar isothermal conduction calorimetry results have been reported by Wang et al [5] where YE was added at 0.8% by mass of cement.

Furthermore, research by Jonkers et al [1] has shown that the addition of YE (1% by mass of cement) leads to a reduction in the strength of concrete. However, research by Cunniffe [20] has suggested that when used below 0.5% by mass of cement, YE has no effect on mortar strength (Figure 3).

Sugars

Sugars, such as glucose and dextrose, are known to be set and hardening retarders of concrete primarily through their ability to prevent the formation of calcium silicate hydrate (C–S–H) [29] by adsorption onto calcium hydroxide and calcium silicates [30]. The retarding effect of sugars increases with sugar content and it has been suggested that a sugar content of 0.1% by mass of cement can lead to an indefinite delay of hardening [30]. However, there has been some suggestions that beyond a sugar content of about 0.15% the retarding effect reduces and that for sugar contents in excess of 0.3% by mass of cement, nucleation effects emerge and that the sugar acts as an accelerator [29].

Sugars also have a greater retarding effect when they are added a few minutes after mixing of the water and the cement [29]. This has implications should rupture of capsules occur during placement of the concrete rather than during mixing.

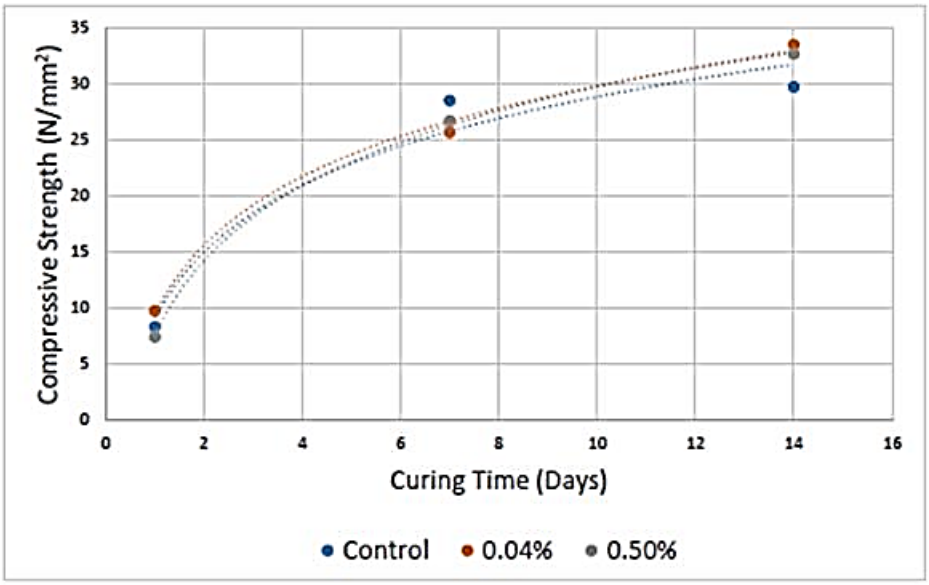

Figure 3 Effect of YE (% by mass of cement) on early-age strength of mortar [20]

Sodium

Germination of spores is known to be favourable in the presence of sodium, and therefore the addition of sodium compounds to nutrient feeds has been considered. Clearly, sodium chloride should be avoided where possible due to its chloride content, whilst the use of sodium citrate has been shown to significantly affect setting and hardening of concrete [22].

Proteins

Peptone, tryptone, tryptone peptone, trypticase, and trypticase peptone are partially digested proteins commonly used in growth media as a source of amino acids, peptides, proteins and nitrogen. They are produced by enzymatic or chemical break down of complex proteins. Research has shown that peptone added to concrete at 1% by mass of cement reduces the strength of concrete [1].

Buffer solutions

Tris base ($(HOCH_2)_3CNH_2$) is an organic compound widely used as a component of buffer solutions. It may increase the rate of hydration of cement, and it has been considered that this may be because it impacts on the dissolution of C_3S and/or acts as a nucleation point for hydration products [11].

3. TEMPERATURE

In relation to their ability to be used in concrete exposed to different exposure the choice of bacteria used also depends on their ability to cope with hot temperatures (thermophilic), cold temperatures (psychrophilic) and salt- and fresh-water environments. Many of these attributes have not been considered in any great detail in self-healing concrete and primarily laboratory experiments to date have considered germination of spores and precipitation of calcite at conditions close to optimum for the bacterial species used; which is usually between 25 and 45°C for most alkaliphilic bacteria [31].

However, some alkaliphiles do show psychrophilic behaviour. For example, strain 207 an aerobic *coccus* can grow at temperatures between -5 and 39°C [31]; the discovery of a calcite precipitating alkaliphile with similar growth characteristics would have significant effect on the development of bacteria-based self-healing concrete.

Some strains of *B. sphaericus, B. megaterium and B. firmus*, have been shown to precipitate calcite in B4 medium even at temperatures as low as 4°C [32]. Whilst the precipitation was slower than that at higher temperatures, this does provide evidence that self-healing at lower temperatures using *bacillus* is possible. Cacchio et al [32] have also observed the formation of calcium carbonate using bacteria from the *arthobacter* species. These are oligotrophic bacteria that can grow in very dry conditions, at low temperatures and with limited nutrient availability. However, most bacteria of this genus are not spore-producing which generally makes them unsuitable candidates for self-healing concrete.

Ghosh et al [10] have performed research to improve the compressive strength of cement mortars using bacteria of the *Shewanella* genus; a thermophilic anaerobic bacteria. However, they did not investigate its capability to grow in concrete at different temperatures, but did observe that the bacteria grew and precipitated minerals up to a pH of 11 [33].

4. CARBONATION

Whilst capsules protect the spores from the initially high pH of concrete, the bacteria must still be expected to function in an alkaline environment once a crack emerges and germination has proceeded. Consequently it has been considered that any bacteria used must be alkaliphilic, such that they are capable of germinating and multiplying in the range of alkali pH conditions present in concrete once the capsules rupture.

Generally speaking alkaliphilic bacteria are those that grow optimally or very well at pH values above 9, often between 10 and 12, but cannot grow or grow slowly at the near-neutral pH value of 6.5 [34]. Furthermore, in addition to an alkaline environment they require Na ions for growth, sporulation and germination [31]. Outside the range of pH permitting growth the pH homeostasis of alkaliphiles is known to gradually or abruptly fail. However, many species are able to remain viable (survive) at pH values outside their normal growth range for some time [31]. Consequently most alkaliphiles will remain viable throughout the high pH ($pH > 13$) environment that exists in concrete at very early ages.

However, the pH of concrete varies over time due to carbonation: a series of chemical reactions that fosters a reduction in the pH. The CO_2 penetrates predominately from the surface through a diffusion process, leading to a carbonated layer of concrete.

As most cracks that will require healing by concrete will be in the initial 20 mm from the surface, then the time for this 20 mm layer to achieve a pH of less than 8.5 is critical. But this

may take anything from as little as a few months to way beyond the design life of the structure (100 years +). In the latter case it is clear that bacterial healing cannot rely on the concrete carbonating before the healing is required, whilst in the former case the concrete may have carbonated before any cracks occur.

Should the concrete rapidly carbonate, then self-healing can be expected to be slow or potentially non-existent, because most alkaliphilic bacteria do not grow well at pH less than 9. Consequently it has been suggested that there is a need in some applications to embed the bacteria with a buffer to maintain the pH at a viable alkalinity. On the other hand, some alkaliphilic bacteria are known to change external pH to a pH suitable for their growth. For example. *B. pseudofirmus* A-57 which has a growth range of pH 8 to 10 has been shown to change a medium of pH 5 to a pH of 8 by producing an alkaline protease in order to grow well [31]. Indeed some researchers have suggested that a key role of bacteria in self-healing concrete is to increase the pH of the environment through different bacterial metabolisms [23].

An alternative approach is to supply the concrete with a multi-bacteria system comprising of calcite precipitating alkaliphilic and neutrophilic bacteria. This is currently being investigated at the University of Bath.

5. HUMIDITY

It is worth discussing the role of water in bacterial self-healing at this point. Clearly water is essential for revival of bacterial spores and furthermore the mineral precursors need to be dissolved in water in order to be available to the bacteria [8]. Consequently bacterial precipitation of calcium carbonate cannot occur if no water is present in the crack zone. For example, tests by Wang et al [14] have shown no healing in specimens stored at a relative humidity as low as 60%. Furthermore in concrete that is protected from water, carbonation cannot occur because there is no aqueous medium or electrolyte in which CO_2 may dissolve – and thus the concrete will tend to have a pH at the higher end of that in which the bacteria can effectively grow. There is therefore perhaps an optimum range of relative humidity in which bacterial self-healing is most likely to occur. It could be argued that this coincides with the environments in which carbonation is most likely to occur. However, alkaliphiles grow less optimally at low pH, and so perhaps self-healing is effective in environments where carbonation is possible but is not at its quickest (Figure 4).

This is key, because many researchers have suggested that the optimum use of self-healing concrete is in situations where repair is difficult, for example underground or underwater, where the concrete is saturated and where there is practically no carbonation.

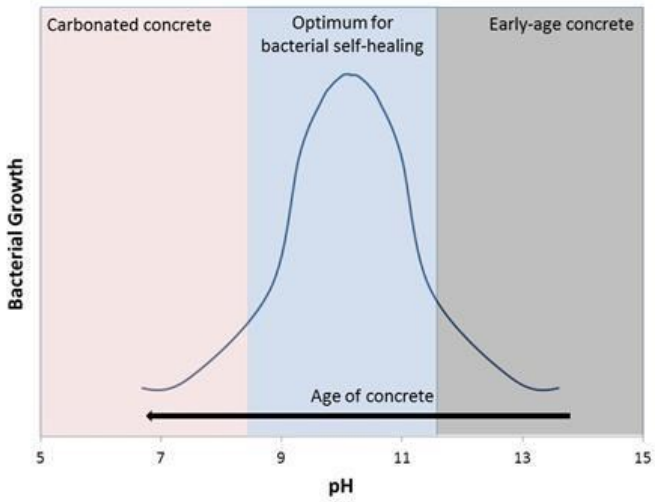

Figure 4 Relationship between pH and optimum conditions for growth of alkaliphiles [35]

6. REPEATED HEALING

6.1 Sporulation

A perceived advantage of bacterial healing over the microencapsulation of minerals and chemicals is that once healing has completed the bacteria return to the spore form and hence are available to take part in future healing reactions should the crack return – provided sufficient nutrients and calcium ions remain in the system (Figure 5). This is useful in dynamic systems, or in products or elements subject to fatigue or cyclic loading.

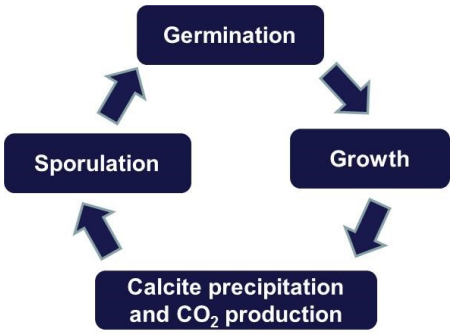

Figure 5 Cyclical nature of bacteria-based self-healing [36]

What has not been researched in any great detail to date is the ability of bacterial cells to return to the spore form on conclusion of their healing role; and thereby remain in a viable state for germination should the crack return.

Many *bacillus* species require the presence of manganese to sporulate and it has been considered that this must be supplied to the system as part of the healing agent if sporulation is required.

6.2 Vascular Networks

A potential problem with bacterial-healing systems is that whilst the bacteria can actively sporulate and germinate, the nutrients and pre-cursors required for growth and healing are only available in a finite quantity. Consequently a way to replenish the system is required. Researchers at Cardiff University have been looking into this problem and have suggested the use of vascular networks, in which the system can be periodically topped up with healing agents such that they never run out, either manually or via a tank connected to the outer end of tubes [37]. Whilst a vascular network requires human intervention, it has some similarities to a biological system that relies upon renewable agents from a living system. Therefore potentially it is a more complete biomimetic system than entirely enclosed systems.

Whilst the applicability of a 1-dimensional vascular network to deliver a low viscosity, single agent cyanocryalate adhesive has been demonstrated by Joseph et al [37] it has not yet been demonstrated that such a system could resupply the essential nutrients and precursors for germination and growth of bacteria and calcite precipitation. Research within Materials for Life (M4L) is considering the use of a range of materials within the vascular network, including bacteria and nutrients to provide the bacterial healing described above [38]. Furthermore as bacterial healing is an aerobic reaction, the healing most readily occurs at surfaces where there is a ready supply of oxygen. It is considered that vascular networks could be used to supply oxygen to the bottom of deep cracks.

In terms of providing the vascular network, the use of glass tubes is problematic for casting, so an alternative is to form holes inside the concrete using polyurethane or polyolefin [39]. Furthermore, developments are taking place within M4L to develop a multi-use 2-dimensional network. Initial experiments have used unreinforced beams, 255 mm by 75 mm by 75 mm in size containing two sets of channels 20 mm from the underside of the beam (Figure 6) [39]. Results show that pressurising the network enhances the flow of healing agents such that they permeate the majority of a 0.2mm crack in a 3-point bending test.

(a) (b)

Figure 6 showing (a) a cast beam with a 2D network of 4 mm holes on the face and (b) a fracture surface [36].

7. CRACK SIZE

Bacterial healing has proven useful at recovering the low permeability of concrete when subjected to micro-sized (or smaller) cracks. However, such systems are generally unsuitable for healing macro-scale cracks (for example, those greater than 0.4 mm). However, a technique has been developed at Cardiff University that uses internal prestressing to close existing cracks sufficiently that they may be healed by in-built microcapsule-based bacterial-healing systems [40]. The system is also useful in preventing new cracks from occurring in the first place [41].

The techniques uses low cost shape memory polymer (SMP) tendons that have the ability to shorten or shrink when heated above a transition temperature. The inclusion of these tendons in concrete permits a compressive strength to be imparted to the concrete when shrinkage mechanisms are remotely activated for example by the supply of electrical heat. The activation can be either before any significant construction loads have been applied or, at a later stage, when the majority of early age thermal and drying shrinkage strains have occurred, and the element has been subjected to considerable mechanical loading.

The system is being developed towards full-scale application as part of the M4L project [42]. Experimental work is using SMP tendons: (i) embedded and cast within the concrete, or (ii) externally anchored through ducts within the concrete [40]. The SMP tendons are made up of commercially available oriented polyethylene terephthalate (PET) strips of 0.046 mm thickness and 32 mm width. In restrained conditions when heated to 90°C it has been shown that the SMP tendons produce a shrinkage stress of 24 MPa that in turn pre-stresses the bottom face of the specimens with 1 MPa compression and encourages closure of cracks.

The results have shown that the SMP tendons close, or significantly reduce crack widths, in structural concrete prisms that have been cracked to 0.5 mm (crack mouth opening displacement). On average a 74% crack width reduction can be achieved after SMP tendon activation and this increases to 93% if specimens are placed in water after cracking and tendon activation. Furthermore, the results indicate that the proposed system enhances the autogenous healing of concrete as 13% load recovery was measured after 28 day healing as opposed to only 1% in similar specimens without SMP tendons [40]. Much improved results are likely possible in combination with encapsulated bacterial healing systems.

8. CONCLUSIONS

Within this paper previous research on the use of bacteria-based self-healing of concrete has been reviewed in relation to the problems associated with the setting, hardening and carbonation of concrete and the problems associated with healing large cracks.

The paper commenced with a discussion of the effects of setting and hardening of concrete and how this affects the way in which self-healing agents must be added to the concrete. In particular, the necessity for encapsulation of the spores and of the medium, in order to counter effects of concrete hardening on the spores and eliminate any effects of the precursor and additional nutrients on the fresh and early-age properties of concrete.

The germination and growth of bacteria and the precipitation of calcite are known to be affected by temperature and humidity, and consequently the role of these parameters was discussed. Furthermore, as concrete ages it tends to carbonate and this leads to a loss of pH – from around 14 in the first few hours to potentially as low as 6.5 in the long-term. The

significance of the carbonation on the ability of bacteria-healing to take place and the consequences for selection of bacteria was described.

One potential advantage of bacteria-healing over other self-healing systems is that it is renewable as the bacteria may sporulate after healing and be available for future healing. This is useful in dynamic systems. The requirements for ensuring sporulation takes place were discussed; in addition to methodologies for re-supplying the bacteria with essential nutrients and medium should these be consumed.

Finally, the paper ended with a discussion of the ability of bacteria to heal large cracks, and describes techniques being considered in UK research to utilize bacteria-healing as part of a multi-scale healing system.

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