Biocementation as a technique for crack repairing

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Keywords: Biocementation; calcite; discontinuities; sealing; rehabilitation.

Abstract. *The main results of this study to investigate the performance of biocementation as a crack rehabilitation technique on three different substrates (ceramic tile, calcareous stone and concrete) and two cracked beams are presented. The sealing effect of the treatment, performed using bacteria, enzyme urease and only the feeding solution was evaluated through changes on pulse velocity and mechanical resistance to penetration in the substrates. Promising results were found. The comparison of the effects of the different treatments performed allowed understanding how to optimize this treatment in practice.*

1 INTRODUCTION

Biocementation consists in using bacteria to produce cement minerals (biocement), which precipitates in the pores and discontinuities of surfaces. The most common mineral is calcium carbonate ($CaCO_3$) and is precipitated after the hydrolysis of urea made by microorganisms (bacteria) living in the media to be treated. The precipitation of calcium carbonate results from the hydrolysis of urea ($CO(NH_2)_2$), promoted by the enzyme urease. The equation for this chemical reaction is Eq. 1:

$$CO(NH_2)_2 + 2H_2O \rightarrow 2NH_4^+ + CO_3^{2-} \quad (1)$$

The ammonium (NH_4^+) released from urea hydrolysis results in pH rise and promotes the precipitation of calcium carbonate ($CaCO_3$), i.e. calcite. It results from the combination of carbonate ions (CO_3^{2-}) by hydrolysis of urea and calcium ions (Ca^{2+}) supplied in the feeding solution (Eq. 2):

$$Ca^{2+} + CO_3^{2-} \rightarrow CaCO_3 \quad (2)$$

Biocementation is being used in mortars, in concrete [1-4] and in porous stones such as limestone, calcarenites and marble [5-8]. Besides providing waterproof coating or weathering resistance coating [9-10], in case of concrete this technique can also delay steel corrosion and be used to seal cracks and fractures caused by aging and weathering [11]. In the literature, some cases are reported where biocementation was evaluated mainly for enhancing existing structures, such as the case described by Wiktor and Jonkers [12], where it showed good results sealing the joints of a concrete slab in a car parking, or by Cuthbert et al. [13] when this technique was tested to seal fractured rock. It can be used for rehabilitation of the historic heritage as well, as it allows localized and non-intrusive interventions.

This abstract presents the main results of this experimental study done to assess the performance of biocementation as crack rehabilitation technique. The experimental program was carried out on three different substrates (ceramic tile, calcareous stone and concrete), in which discontinuities were designed to be sealed by this biological treatment, and on two concrete cracked beams. The treatment consisted on adding fluids with controlled concentration of bacteria, urease enzyme or only the feeding solution to the discontinuities. The sealing effect evaluation of the treatments through nondestructive techniques is presented.

2 MATERIALS AND METHODS

2.1 Biological treatments

Three types of treatment were applied: (i) bacteria; (ii) crude extract of enzyme obtained from the bacteria by sonication followed by centrifugation and filtration and (iii) control with the feeding solution. The treatment with enzyme was adopted as it is the catalyst in the process of deposition of calcite. Using enzyme instead of bacteria has the advantage of avoiding using living organisms, therefore reducing the maintenance needs during the treatment. The treatment using feeding solution was adopted as control, because the reagents present can form calcium carbonate independently from the presence of the microorganisms or enzyme. However, in the presence of the last, the reaction is much faster and for this reason the treatment is expected to be more efficient than just add the reagents.

The bacterial species used in the present investigation is *Sporosarcina pasteurii* (American Type Culture Collection, strain #11859). Optimum conditions for growing are pH between 8 and 9 and temperature closer to 36ºC. *S. pasteurii* was cultured in a liquid medium composed by 20 g/l yeast extract, 10 g/l of ammonium sulphate and 0.13 M Tris buffer pH 9.0, at 37°C under aerobic condition, up to a cell density of 10^9 cells/mL (optical density at 600nm of 1). Two identical cultures were prepared: one for the treatment with the bacteria and another for the treatment using the enzyme extracted by sonication. The amount of enzyme in the crude extract was not quantified. The feeding solution was prepared with 1:10 diluted culture medium supplement with 0.5 M of urea, 0.5 M of calcium chloride (calcium source).

2.2 Substrates and design of the discontinuities

The experimental program was carried out on three different substrates where discontinuities were created to be sealed by this biological treatment. The substrates were ceramic tiles (samples C1 to C8, bulk density 1.73 g/cm^3), calcareous stone plates (samples P1 to P9, bulk density 2,57 g/cm^3) and concrete plates (samples B1 to B9, bulk density 2.17 g/cm^3). The treatments performed for each sample are in Table 1. Two discontinuities were performed in each sample, using a circular saw (diameter 20 cm, 2mm thickness, Figure 1) and water to ensure clean edges. The resulting discontinuities had widths varying between 2 and 4mm and depths varying between 1 and 5mm (see average values in Table 1 for each material). These distances were measured using a caliper ruler.

Table 1: Geometry of the discontinuities (length of 3 and 5cm) and samples identification considering the different treatments performed.

Material	width (mm)	depth (mm)	Bacteria	Enzyme	Feeding sol.
Concrete plate (samples B)	2.95 ±1.00	3.76 ±1.18	B3; B4; B7; B8	B1; B5; B6	B2; B9
Ceramic tile (samples C)	3.01 ±1.05	1.77 ±0.64	C1; C4; C7	C2; C3; C8	C5; C6
Calcareous plates (samples P)	2.88 ±0.79	2.57 ±1.05	P4; P5; P8; P9	P1; P3; P6	P2; P7

a) b)

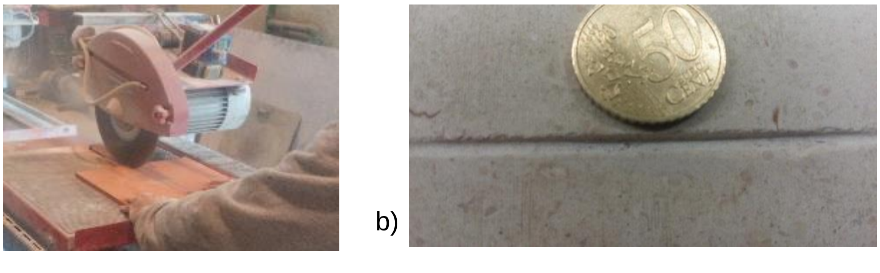

Figure 1: Saw used to perform the discontinuities and an example in the calcareous stone.

2.3 Treatment solutions

The treatment consisted on adding a controlled concentration of bacteria, enzyme or only the feeding solution. A syringe with needle was used to completely fill the volume of the discontinuity with the treatment fluid (Figure 2).

a) b)

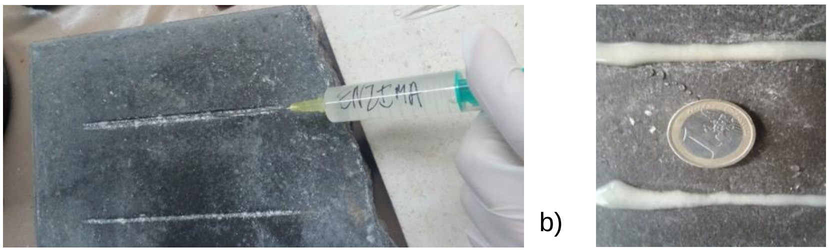

Figure 2: a) Treatment application in a discontinuity in a concrete plate; b) the way the fluid looks like when the chemical reaction is in progress (example using bacteria).

Each treatment was applied in three stages, being the discontinuity completely filled with the treatment fluid, except for the treatment with bacteria. In the latter, the bacteria were added only in the first day of each stage and then only feeding solution was added the next two days. For the other treatments, all the stages were identical (feeding solution and enzyme added for the treatment with enzyme, and only feeding solution added for the treatment with feeding solution). In all cases the time interval between stages was around a month. The reactions start during the application, as the fluids with bacteria and enzyme became white and milky (Figure 2). During the treatment, the samples were left in the laboratory (relative humidity of 42% and air temperature of 21ºC, average values).

In each beam, a reservoir was applied over the cracks and sealed with silicone. The beams were painted two times with a yellow acrylic paint F-1016S to ensure they beams would not leak through the sides. The result is visible on Figure 3.

a) b)

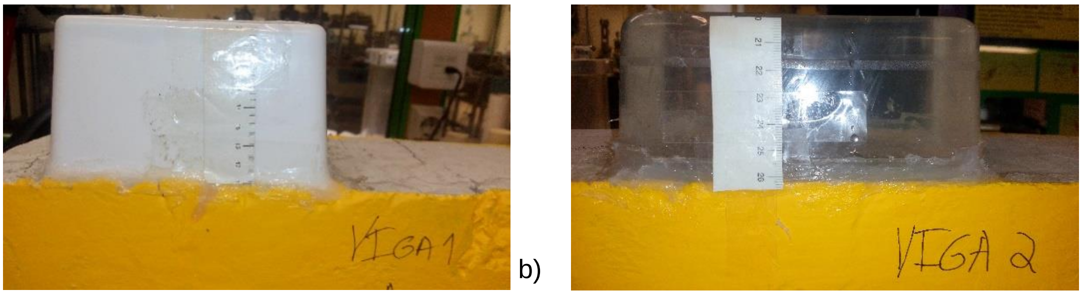

Figura 3– Treatment reservoir on beam 1 (a) and 2 (b)

On one face of each reservoir was glued a paper scale in order to monito the level of fluid.

2.4 Evaluation of the sealing effects

The first assessment was visual observation, as the biocement produced is calcite and is a whitish mineral seen at naked eye. Several photographs were taken using a digital camera. Later, a digital optical microscope was used to take photographs to the minerals present in the discontinuities. The microscope used (I-WOW Microscope, Figure 4.a) has a 2.0 MP camera and resolution 640x480 pixels.

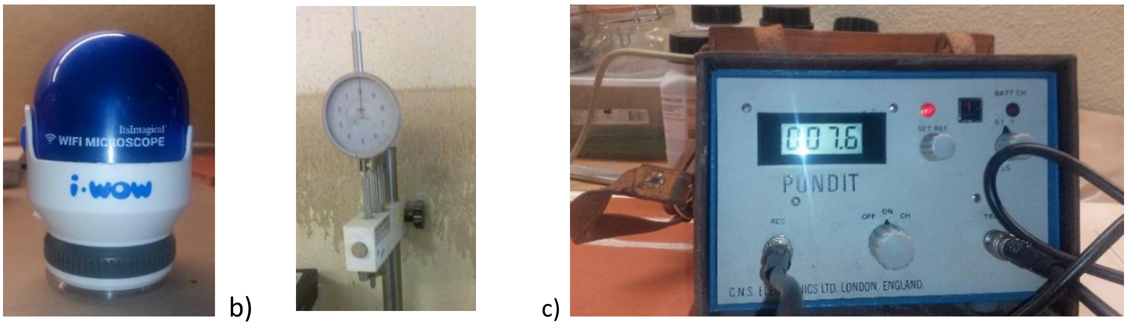

Figure 4: Equipment used for discontinuities sealing inspection: a) optic microscope; b) needle used for measuring the depth of the discontinuity; c) ultrasonic wave measurement equipment.

The sealing effect of the treatments was evaluated through changes in the depth of the discontinuities and changes in ultrasonic pulse velocity. The measurements were done at the end of each step of treatment.

The depth of the discontinuities was measured using a needle for strength to penetration tests (Figure 4.b) based in the principle that the needle displacement to reach a solid substrate was reducing with the deposition of calcite. The difference between the displacements measured before and after the treatment steps is the thickness of the calcite layer. This equipment (Petrotest instruments, following ASTM D5-06) allowed measuring the thickness of the calcite layer (tcalcite) with the precision of 0.01 mm. Due to the length of the discontinuity and eventual heterogeneity in calcite deposition, 3 to 5 measurements along its longitudinal axis were performed and the average values were considered. The values measured were confirmed using a caliper ruler (precision of 0.05 mm).

The information on the thickness of the calcite layer allowed a direct measurement of the sealing effect of the treatment, name filling percentage, *filling%*, which was quantified through the relationship between the thickness of the calcite layer, $t_{calcite}$, and the initial depth of the discontinuity, d_0:

$$filling\% = \frac{t_{calcite}}{d_0} \times 100 \tag{3}$$

The changes in the ultrasonic pulse velocity provided an indirect measurement of the filling of the discontinuities and on the cracks of the beams. The measurements concern the time necessary for the transmitted signal, from the transmitting transducer E, to be detected by the receiving transducer (R). As illustrated in Figure 5, the length of the path of the wave inside the material reduces when the discontinuity becomes filled with calcite (red material in the figure), as this new material provides physical support for the wave propagation. In this work, this transition time was measured with the Pundit equipment shown in Figure 4.c (ASTM C597) and 50 kHz transducers [14].

The transducers (steel disks with 7 cm diameter) were placed having the discontinuity between them and 10 cm distant from their centers. Using this distance and the propagation time it was possible to compute the wave velocity. The relation between the propagation times after and before the treatment (t_{after} and t_{before}, respectively), which is identical to the relationship between the propagation speed if the distance between E and R is fixed, provides an indirect filling indicator called the time percentage, *time%*:

$$time\% = \frac{t_{after} - t_{before}}{t_{before}} \times 100 \tag{4}$$

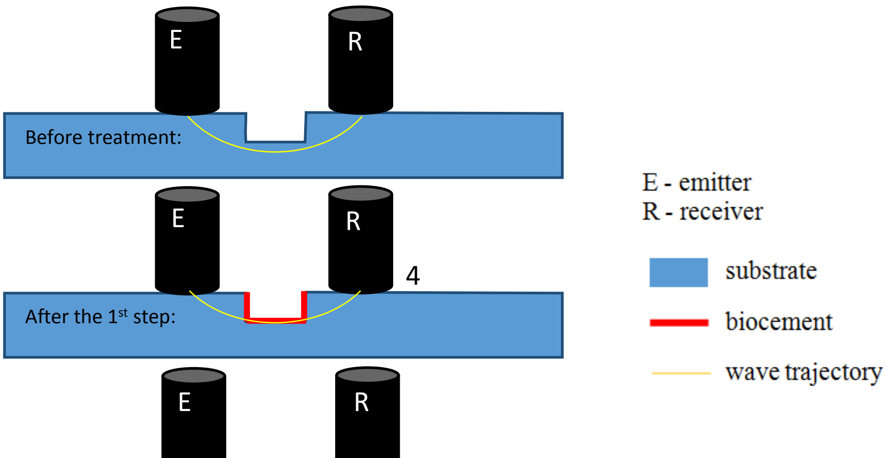

Figure 5: Working principle of ultrasonic wave measurement test to detect sealing of discontinuities.

The cracks on the beams were also tested for water permeability. The reservoirs were filled with water and the level was monitored for days. After that the data is computed into graphs with four lines (one for each round of treatment and one for pre-treatment stage). Using the lines' slopes, we can use (5) to obtain an indicator of treatment effectiveness.

$$slope\% = \frac{slope_{after} - slope_{before}}{slope_{before}} \times 100 \tag{5}$$

The objective is to analyse if the water level takes longer to vary (descend) from round to round of treatment, maintaining the lab conditions, which indicates the cracks are being filled with biocement.

3 RESULTS AND DISCUSSION FOR SUBSTRATES

3.1 Visual assessment

The formation of calcite was observed visually by detecting the presence of a solid white precipitate in the discontinuities. Some results are presented in Figure 6, which gathers photographs of discontinuities after the different treatments and also an example of discontinuities in each substrate type.

Concerning first the different treatments, filling is evident when the enzyme and bacteria treatments are applied. The precipitated calcite from the feeding solution is more visible for the ceramic substrate, which is explained by the colour contrast.

The minerals from the treatment using bacteria have a granular look and the colour is yellowish, while those from the treatment with enzyme are more homogeneous and white. This difference is evident in the concrete substrate (Figure 6). The minerals were analysed with more detail through optical microscope and the photographs are shown in Figure 7. These photographs confirm the different morphologies of the minerals, as those formed by the bacteria are larger and more irregular than those formed in the presence of the enzyme.

For the same treatment, the minerals appear to be different for the different substrates. Their size and shape may be explained by the presence of bacteria, because each one works as a nucleation centre forming biocement around its exterior [15]. However, the amount of fluid available during the formation of calcite crystals also affects their size and this amount depends on the nature of the fluid (viscosity, density), contact angle with the surface, geometry of the discontinuity and the porosity of the substrate material. A final test, consisting in passing the finger in the discontinuities, was performing to check qualitatively the adhesion of calcite to the substrate. The finger was removing calcite when the treatment using the feeding solution and enzyme with feeding solution was done.

Figure 6: Discontinuities before and after treatments, showing the filling material after the treatment.

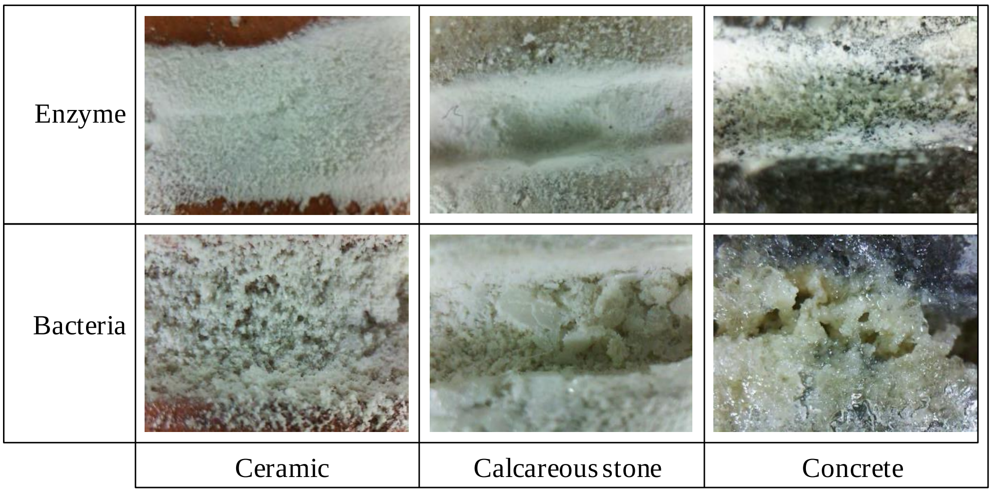

Figure 7: Optical microscope photographs from the calcite minerals.

3.2 Depth of the discontinuities

As described, the sealing effect of the treatments was evaluated through the measurement of the thickness of calcite. The different measurements are presented in Figure 8 for each discontinuity in each substrate (one bar is one discontinuity). The average values and corresponding standard deviation are in Table 2. Their comparison allows to better characterize the effectiveness of the treatment, however, requires considering the depth of each discontinuity.

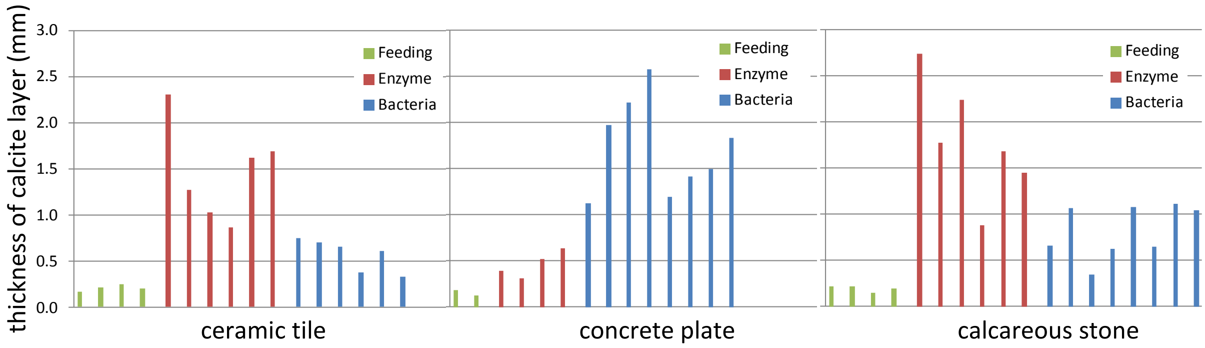

Figure 8: Comparative calcite thickness distribution in the discontinuities after the different treatments for each substrate.

Similar tendencies can be found when comparing calcite thicknesses and filling percentages, which was expected because the discontinuities had average similar depths among the materials. With the data from Figure 8 and knowing the depth of each discontinuity (average values in Table 1), using Equation 3 it was possible to compute the filling percentage presented in Table 2. The average values are also in this table.

Table 2: Thickness of the calcite layer and filling percentage achieved using the different treatments tested (average values and standard deviation).

	Feeding solution		Enzyme		Bacteria	
	$t_{calcite}$ (mm)	Filling% (%)	$t_{calcite}$ (mm)	Filling% (%)	$t_{calcite}$ (mm)	Filling% (%)
Ceramic	0.21 ± 0.04	13.86 ± 3.39	1.46 ± 0.52	78.27 ± 20.37	0.57 ± 0.18	34.42 ± 8.20
Concrete	0.15 ± 0.04	8.09 ± 4.17	0.47 ± 0.14	13.86 ± 3.78	1.72 ± 0.51	41.01 ± 15.95
Calc. stone	0.20 ± 0.03	13.46 ± 5.71	1.79 ± 0.64	79.18 ± 11.76	0.82 ± 0.29	25.13 ± 5.16

The relationship between the depth of each discontinuity and the filling percentage achieved is presented in Figure 9.

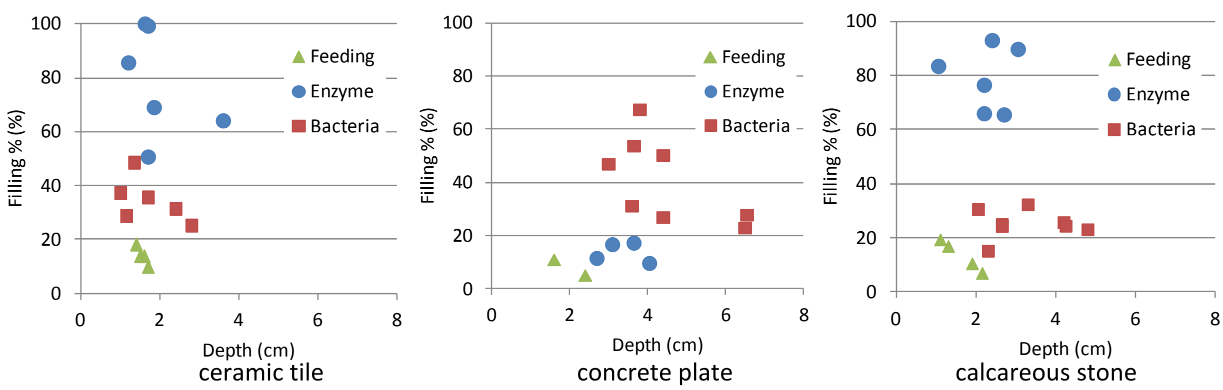

Figure 9: Relationship between the initial depth of the discontinuity and its filling% for each substrate.

Accordingly, with Figure 9, except for the feeding solution it appears that depth has no influence on filling percentage of the discontinuities. The good relationships found only for the case using the feeding solution, with filling% decreasing with increasing depth, suggests that calcite layer is a coat with fixed thickness caused by chemical precipitation (also observed in Figure 8). Similar results were found by Cardoso et al. [16] when studying this treatment on surfaces.

Even if there is lack of correlation of the points in Figure 9 for the other two treatments, it is clear that, fixing the depth, discontinuities filling after treatment using only the feeding solution is always bellow that achieved with bacteria and enzyme. This result confirms that, in all cases, the treatment was more effective when the fluids with bacteria or with enzyme were applied on the discontinuities. This result was expected in accordance with visual observation, as already discussed.

Nevertheless, in both figures and Table 2 can be seen larger dispersion when bacteria and enzyme are used, probably because the biological activity is difficult to be controlled in this particular laboratory environment. This scattering is larger for the concrete substrates than for the ceramic and calcareous ones.

While for the concrete plates larger calcite thicknesses and filling percentages were found for the treatment with bacteria, the treatment using enzyme appeared to be more effective for the other two materials because the filling percentages increased significantly for those treatments. This may be related with the time that the fluid remained in the discontinuity before drying in concrete surfaces, giving time for the bacterial activity, however further analysis is necessary.

3.3 Changes in the ultrasonic pulse velocity

The sealing effect of the treatments was evaluated also through changes in ultrasonic pulse velocities. The improvements achieved were computed through the percentage of time increment, a parameter found using Equation 4 previously presented. The values as presented in Figure 10 for each discontinuity and in Table 3 concerning average values and standard deviation. As when the results from the previous tests were analyzed, some differences were found for the different treatments. However, the inaccuracy in using this indirect method is larger than when using the others because the standard deviation is very large (Table 3).

Table 3: Percentage of ultrasonic speed increment with the different treatments (average values and standard deviation) measured through parameter time%.

	Feeding	Enzyme	Bacteria
Ceramic	19.10 ± 4.36	19.58 ± 7.66	18.50 ± 8.96
Concrete	4.84 ± 2.99	5.38± 3.70	16.67 ± 4.98
Stone	16.92 ± 5.09	22.64 ± 10.95	18.16 ± 7.55

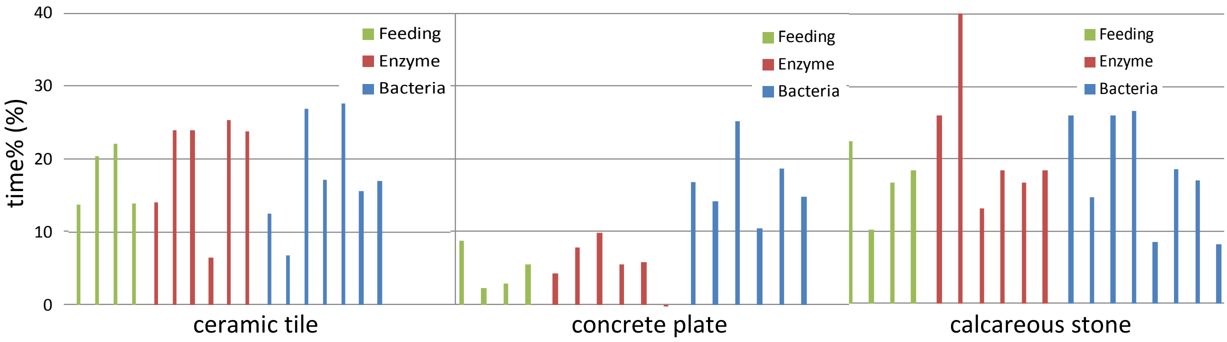

Figure 10: Comparative percentage of speed increment in the discontinuities after the different treatments for each substrate.

Some increment can be observed in the ultrasonic pulse velocities after the treatment with enzyme and bacteria, being more significant for the calcareous stone and the ceramic plates than for the concrete tiles. Using this measurement method, the treatment with bacteria appears to be more significant than using enzyme for the concrete plates, while the enzyme treatment was more efficient for the calcareous stone and ceramic plates. These results are similar to those found when the thickness of calcite layer was measured. The deviations observed can be explained by experimental uncertainty associated to this indirect measurement method, as well as to the low porosity of the ceramic substrates.

4 RESULTS AND DISCUSSION FOR BEAMS

Similar to what was seen on section 3.1, the formation of calcite was proved by the visual detection of a white precipitate in the cracks, like shown on figure 11.

a) b)

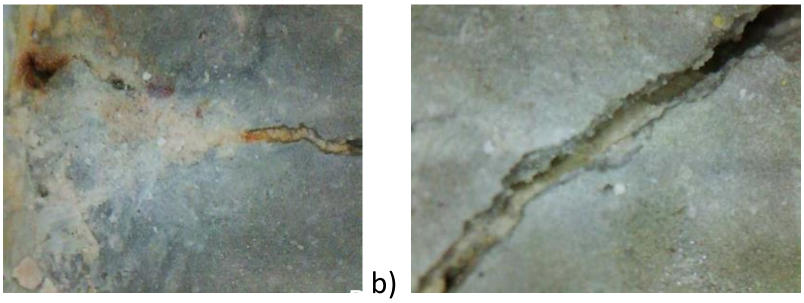

Figure 11: Cracks after treatment - a) for enzyme treatment; b) for bacteria treatment

It is visible that the treatment caused a reaction of oxidation on the steel bars which explains the red colour seen on some parts of the cracks. On both beams it is also visible the presence of biocement in the cracks, close to the top surface of the beam, which does not mean that the crack is completely filled in its depth.

The characteristics of the material are very similar to the ones described in section 3.1 for the treatment with bacteria, with the presence of a large amount of crystals. The minerals were analyzed with more detail through optical microscope and the photographs are shown in Figure 12.

a) b)

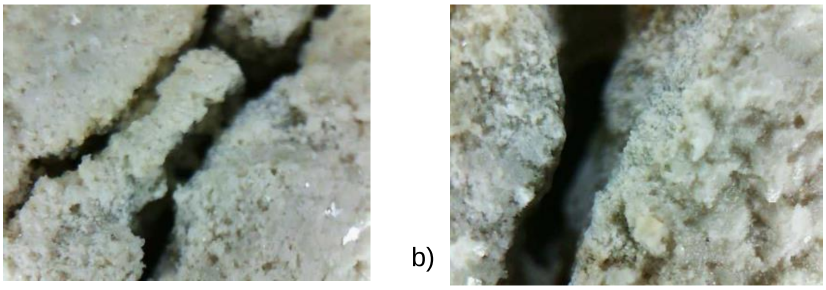

Figure 12: Optical microscope photographs of the minerals

The sealing effect of the treatments was evaluated also through changes in ultrasonic pulse velocities. The improvements achieved were computed through the percentage of time increment, a parameter found using Equation 4 previously presented. For the beam treated with enzyme, the improvement was around 200%, around four times more than on the other beam, where it was around 50%. These results are not in agreement with the ones on section 3.3 for the concrete substrate, where the bacteria treatment was around three times better than enzyme one.

Finally, for the water level variation the results were also very promising for both treatments. The improvement registered was similar, but with the enzyme results were slightly better (66%) compared than those found using bacteria (57%).

Taking just in consideration the results of the pulse velocity and water level variation, it can be concluded that the enzyme treatment appears more effective on cracked concrete beams than the bacteria one.

5 CONCLUSIONS

In this dissertation the sealing effect on the plates of different materials was evaluated through visual assessment, the percentage of filling with calcite and the changes in pulse velocity. Overall, the treatment using each of the three fluids chosen was effective because calcite precipitation was detected. The amounts of calcite increased significantly when enzyme or bacteria were used, while they were almost insignificant when just using the feeding solution.

The results were promising because the discontinuities on the substrates and on the beams, were filled with calcite. In the substrates, the maximum filling percentages are above 78% when using enzyme in ceramic and in calcareous stone discontinuities, while it is above 40% using bacteria for sealing discontinuities in concrete plate. They are about 10% when only feeding solution is used, therefore there is a clear advantage in using bacteria and/or enzyme.

The results were slightly different when measuring changes in ultrasonic pulse velocity. The indirect method was used with higher uncertainty and eventually water content influenced the readings. The discontinuities depth is a direct measurement, so its results can be considered to be more reliable. From visual observation, the calcite formed in the presence of bacteria had a granular appearance, being yellowish in the concrete discontinuities. This indicates that the treatment must be tested before being applied to check its compatibility with the substrate and the final aesthetic effects.

For the beams both treatments presented very similar results and appear effective in filling the cracks according to a visual inspection. The biocement deposited for both treatments looks similar to the one deposited on the concrete substrates using bacteria treatment, both in colour and texture. Also, the results for the water level variation are very alike with slight advantage for the enzyme treatment. The results differ more for the ultrasound speed test where there the enzyme treatment presents much better results, i.e., bigger increase in speed of the waves.

Without any other inspection results it can be said the enzyme treatment is overall better for the concrete beams than the bacteria one, which doesn’t match what was concluded for the concrete substrates, where the bacteria treatment showed better results. Taking in consideration that the type of concretes of the substrates and the beams are different and that the number of inspection tests as well as the number of substrates and beams were limited, it cannot be concluded which treatment is, in fact, more effective. However, it is clear that both give good indications of being a viable alternative to the traditional techniques and that in the future they might be used more frequently in-situ.

Despite the promising results, further study is necessary to fix the treatment conditions not only in horizontal surfaces but also in walls. The mechanisms of calcite formation in the discontinuities and how they are affected by the characteristics of the porous medium to be treated and of the discontinuities (geometry and surface) must also be investigated.

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