

# EFFECT OF BACTERIA *BACILLUS PSEUDOFIRMUS* AND FUNGUS *TRICHODERMA REESEI* ON SELF-HEALING ABILITY OF CONCRETE

HANA ŽÁKOVÁ\*, JIŘÍ PAZDERKA, ZUZANA RÁCOVÁ, PAVLA RYPAROVÁ

Czech Technical University in Prague, Faculty of Civil Engineering, Department of Building Structures, Thákurova 7, 166 29 Prague, Czech Republic

\* corresponding author: [hana.zakova@fsv.cvut.cz](mailto:hana.zakova@fsv.cvut.cz)

**ABSTRACT.** There are usually used inorganic substances for the strengthening of self-healing ability of concrete but some specific types of bacteria or fungi may also be considered for use. The bacteria's ability to fill pores and micro-cracks was investigated on cylindrical concrete specimens with the diameter 55 mm and height 5 mm. Bacteria *Bacillus pseudofirmus* and fungus *Trichoderma reesei* was used for the experiment. The main objective of the experiment was to investigate, if it is possible to use *Bacillus pseudofirmus* and *Trichoderma reesei* for self-healing concrete.

The results show, that it is more advantageous to use *Bacillus pseudofirmus* than *Trichoderma reesei* in alkaline environment. It is desirable to create the most ideal conditions for microorganism's growth, as possible. Bacteria should have positive effect on self-healing ability of concrete. Any effect of fungus on self-healing ability of concrete wasn't confirmed.

**KEYWORDS:** Self-healing concrete, *Bacillus pseudofirmus*, *Trichoderma reesei*, filling of pores and micro-cracks.

## 1. INTRODUCTION

One of the highest priorities in civil engineering is to ensure the maximum durability of concrete structures in accordance with the theory of sustainable building. From this perspective, the typical problem is more financial requirement caused by necessary remediation measures in the concrete structure, related to the load carrying capacity or waterproofing, which is a key parameter to ensure durability. One of the options that have emerged in recent years is to use self-healing concrete. More specifically, this concrete structures have this ability significantly strengthened, because almost every concrete structure has a certain ability of autogenous healing, but this ability is limited (depends on boundary conditions). For strengthening of the self-healing ability of concrete, there are usually used inorganic substances, but some specific types of bacteria or fungi can also be used. The main objective of these bacteria and fungi is to penetrate the structure of the concrete and fill micro-cracks and pores [1],[2]. The waterproofing of the structure should increase by using microorganism, increasing of the load bearing capacity in some cases is also assumed [1],[2]. Resistance of the microorganisms to the highly alkaline environment, typical for cement, is the necessary condition for using. This problem is mainly related to the fresh and low carbonated concrete, which can be found mainly in the new buildings, but there also can be various disorders that need to be rehabilitated. It is estimated that, in the world, 7% of CO<sub>2</sub> emissions are due to the cement production and, therefore, it would be advantageous to reduce the quantity produced as



FIGURE 1. Concrete specimen (diameter 55 mm, height 5 mm).

much as possible [1].

## 2. METHODS

The ability of microorganisms to fix micro-cracks and pores was investigated during the experiment on cylindrical concrete specimens with the diameter 55 mm and height 5 mm (Figure 1). Bacteria *Bacillus pseudofirmus* (Figure 2) and fungus *Trichoderma reesei* (Figure 3) was used for the experiment. Concrete specimens were created from mixture consisting of Portland cement (CEM I 42.5), aggregate (fraction 0-4) and the water. The microorganisms weren't added to the mixture in this phase.

For verification, if the microorganisms are able to grow up in early-aged concrete with high alkalinity the specimens weren't artificially carbonated. The experi-

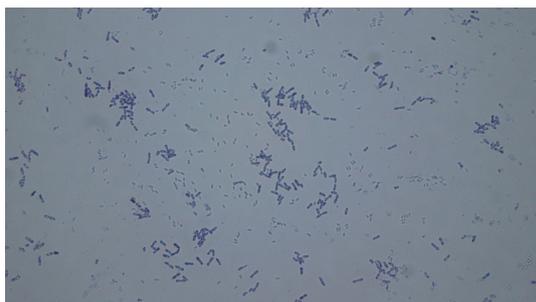


FIGURE 2. *Bacillus pseudofirmus*, Olympus BX41, 1000x.

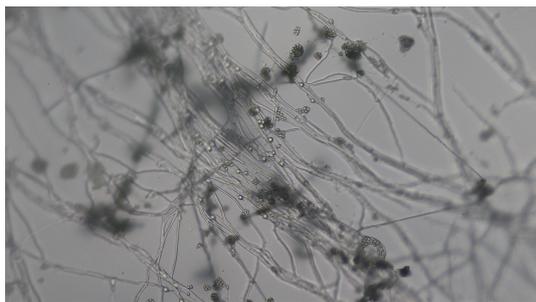


FIGURE 3. *Trichoderma reesei*, Olympus BX41, 100x.

mental measurement began 28 days after embedment of the specimens. First, moisture content and pH were measured. For determination of the moisture content the Gravimetric method was used. Subsequently, the specimens were saved in environment of 100% relative humidity for 4 weeks. After that, the microorganisms were implemented. Only one type of concrete mixture was used for the experiment, but the test specimens were placed in different conditions. The main objective was to identify differences in the growth of microorganisms on specimens kept in ideal conditions and specimens kept in conditions more close to the real situation on site.

There was measured a spectrum (Figure 5) of *Bacillus pseudofirmus* in 253 media using Spectrophotometer Spectroquant Pharo 300. For bacterial growth investigation, 5 specimens were placed in 253 media, 5 specimens were placed in 253 media with 2% CaCl<sub>2</sub> solution (Figure 4) and 5 specimens were placed in sterile water. Into two Erlenmeyer flasks, there were 253 media and 253 media with 2% CaCl<sub>2</sub> solution, without specimens. All of these solutions have been inoculated with the bacteria *Bacillus pseudofirmus* in a ratio of 1 ml solution with bacteria to 100 ml media. Growth curve was measured by spectrophotometry for next 28 days. Measured curves initially show an increase, but gradually they have a decreasing tendency. After 28 days of the bacterial deployment, the growth curve measurement was terminated.

For fungus growth investigation, 8 specimens were placed in Petri dishes with malt agar. 5 specimens were inoculated with fungus *Trichoderma reesei*. 3 specimens weren't inoculated and one Petri dish just with malt agar was inoculated with *Trichoderma reesei*



FIGURE 4. Concrete specimens in media (253 media on the left, 253 media with 2% CaCl<sub>2</sub> solution on the right).

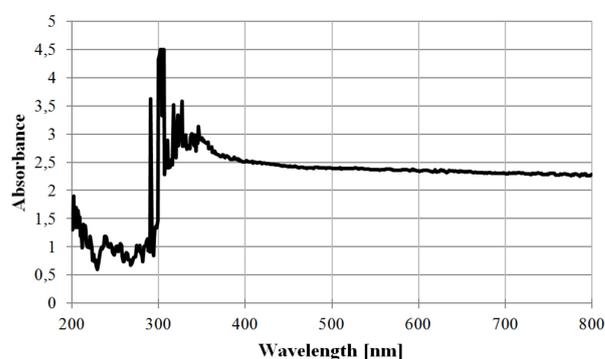


FIGURE 5. Spectrum of *Bacillus pseudofirmus* in 253 media.

for the reference test. All Petri dishes were placed in a thermostat where they were kept at a constant temperature 24°C.

13 specimens were placed in desiccators with 100% humidity (Figure 6). 10 of them were regularly inoculated by solution. 5 specimens were inoculated with fungus *Trichoderma reesei* and 5 specimens were inoculated with bacteria *Bacillus pseudofirmus*. 3 specimens were placed in one desiccator for reference test and weren't inoculated neither with bacteria nor with fungus.

One specimen was placed in Petri dish without agar and without inoculation and was left in normal laboratory environment as a reference test.

### 3. RESULTS AND DISCUSSION

Concentration of bacteria in 253 media and 253 media with 2% CaCl<sub>2</sub> solution increased rapidly. Growth curve measured in solution with specimens was similar as in the solution in Erlenmeyer flasks. In solution of bacteria in water with specimens wasn't so rapid increasing of bacteria concentration as in both media (Figure 8). 28 days after experiment started, surface of the specimens was controlled using a microscope Olympus BX41. On the surface of specimens placed in 253 media with 2% CaCl<sub>2</sub> solution and specimens



FIGURE 6. Concrete specimens in desiccators.



FIGURE 7. Development of fungus *Trichoderma reesei*.

placed in water, there were no visible changes (Figure 9). On surface of specimens placed in 253 media, there were white crystals observed (Figure 11).

On the surface of the specimens placed in desiccator and inoculated by bacteria *Bacillus pseudofirmus*, there were no visible changes 33 days after first inoculation.

Fungus *Trichoderma reesei* grow up on malt agar around the specimens (Figure 7). 33 days after experiment started, surface of the specimens was controlled

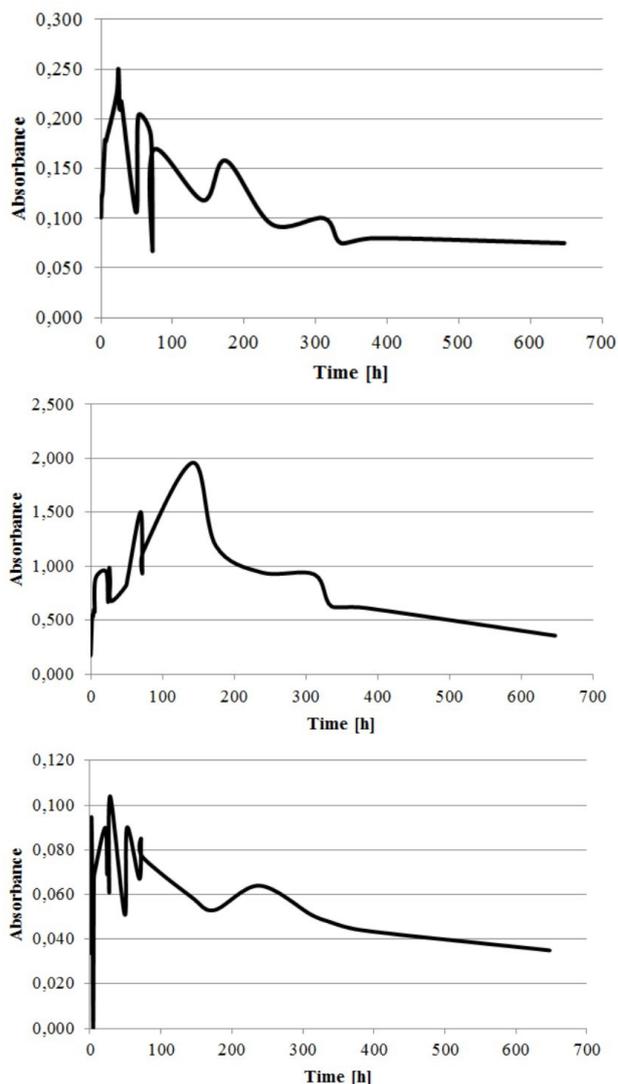


FIGURE 8. Growth curves of bacteria in 253 media (on top), in 253 media with 2% CaCl<sub>2</sub> solution (in the middle) and in the water (on bottom).

using a microscope Olympus BX41. There were no visible changes on the surface of the specimens. This result was probably due to high alkalinity of the specimens.

On the surface of the specimens placed in desiccator and inoculated by fungus *Trichoderma reesei*, there were no visible changes 33 days after first inoculation.

The experiment shows, that using of bacteria *Bacillus pseudofirmus* for making self-healing concrete is more advantageous than using of fungus *Trichoderma reesei*, because bacteria is able to grow up in high alkalinity conditions. The experiment also shows that it is necessary to provide the most ideal conditions to the microorganism's growth, as possible.



FIGURE 9. Surface of the specimen placed in solution of bacteria in 253 media in 2%  $\text{CaCl}_2$  solution.

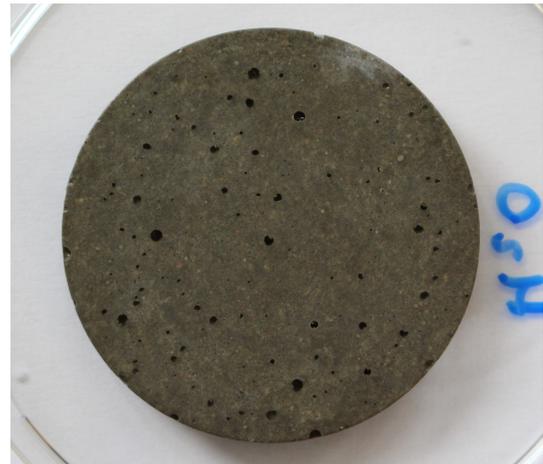


FIGURE 10. Surface of the specimen placed in solution of bacteria in the water.



#### 4. CONCLUSION

Effect of bacteria *Bacillus pseudofirmus* on self-healing ability on early-aged concrete was confirmed just in one case - when concrete specimens were placed in 253 media. In other cases the effect wasn't confirmed. But it is possible, that bacteria *Bacillus pseudofirmus* should have positive effect on the self-healing ability of concrete in optimal conditions. It is necessary to continue in the research and make other experiments.

Effect of fungi *Trichoderma reesei* on self-healing ability on early-aged concrete wasn't confirmed. It probably was due to high alkalinity of concrete specimens. The research show, that using of the *Trichoderma reesei* in concrete with high alkalinity is useless.

#### ACKNOWLEDGEMENTS

This work was supported by CTU research project no. SGS17/117/OHK1/2T/11.

#### REFERENCES

- [1] H. M. Jonkers, A. Thijssen, G. Muyzer, et al. Application of bacteria as self-healing agent for the development of sustainable concrete. *Ecological engineering* **36**(2):230–235, 2010.
- [2] J. Luo, X. Chen, J. Crump, et al. Interactions of fungi with concrete: Significant importance for bio-based self-healing concrete. *Construction and Building Materials* **164**:275–285, 2018.



FIGURE 11. Surface of the specimen placed in solution of bacteria in 253 media.

