Influence of Calcium Chloride and Urea in Self-Healing Concrete Using *Bacillus subtilis*, *Bacillus megaterium* and *Bacillus licheniformis*

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Abstract

Concrete is a non-flexible and breakable material that is widely used in building construction. Concrete cracking may affect the strength of buildings. Due to difficulties of concrete fixing processes, this research focuses on developing a self-healing concrete by using a biocementation process of bacteria. As bacteria have a property of producing calcium carbonate from calcium chloride, urea, and water, *Bacillus licheniformis*, *Bacillus megaterium* and *Bacillus subtilis* were chosen. In this research, the concentration of calcium chloride and urea were varied from 0.1 – 0.7 M and 0.6 –1.2 M, respectively to find the optimal concentrations which potentially yield the most calcium carbonate precipitate from bacteria. Bacterial cultures were monitored by observing growth, pH and urea consumption. The results indicated the number of bacterial cells showed slightly increasing trends with the decrease of pH and urea consumption, which confirming the usage of substrates. The results of investigating optimal substrates concentration showed that concentration of calcium chloride in the range of 0.1 M – 0.7 M and concentration of urea in the range of 0.1 M – 0.7 M did not significantly affect biocementation of bacteria. This could be because the usage of substrates affected pH level in solution, resulting in sub-optimal bacterial growth. However, the rates of biocementation from bacteria culturing with various substrate concentrations were obtained. It was found that *Bacillus subtilis* produced the highest amount of calcium carbonate, followed by *Bacillus licheniformis* and *Bacilus megaterium*, respectively. This indicates the potential for developing self-healing concrete by using bacteria.

Introduction

The common material that use for constructing buildings is concrete. It is a fragile material which can be easily broken by human activities and natural disasters. Moreover, reparing the cracked concrete might cause risks to the workers. A self-healing concrete, which occurred by a biocementation process of bacteria *(Figure 1)*, is investigated to decrease the risks that might occur.

$$CO(NH_2)_2 + H_2O \longrightarrow 2NH_3 + CO_2$$

$$2NH_3 + 2H_2O \longrightarrow 2NH_4^+ + 2OH^-$$

$$CO_2 + OH^- \longrightarrow HCO_3^-$$

$$Ca^{2+} + HCO_3^- + OH^- \longrightarrow CaCO_3 + H_2O$$

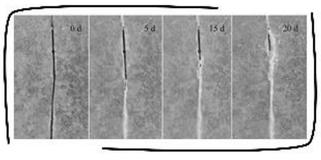


Figure 1: The picture shows the crack that was healed after 20 days. (Mian Luo, et al. 2015)

Objective

1. To find the optimal concentration of urea and calcium chloride in a bacterial culture medium for a proper biocementation.

2.To find the rates of biocementation from different bacteria: *Bacillus subtilis*, *Bacillus megaterium* and *Bacillus licheniformis*.

Result & Discussion

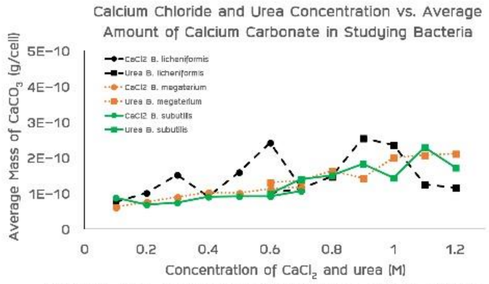


Figure 3 : The graphs show the relation of calcium chloride and urea concentration that affected average amount of calcium carbonate in stucying bacteria

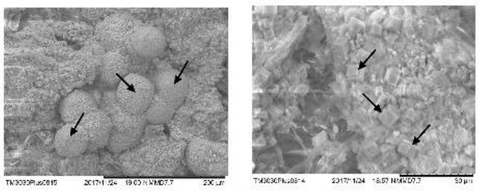


Figure 4 : The pictures show the morphologies of $CaCO_3$ which consisted of spherical veterite **(left)** and cubic calcite **(right)**

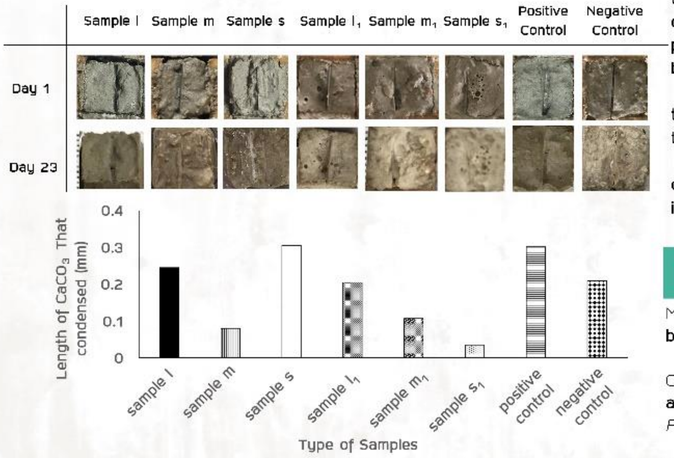


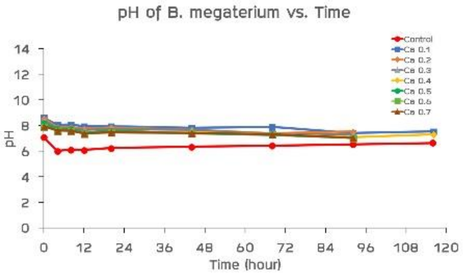
Figure 5 : The table shows different samples of concrete mixed with different types of bacteria. They were cracked on the first and the last day of experiment. The graph shows the length of crack that was healed on each type of samples.

Methodology

1. Investigating Suitable Concentration of Substrates by varying calcium chloride and urea in bacteria culture media from 0.1 – 0.7 M and 0.6 – 1.2 M. The bacteria were cultured for 5 days and monitoredcell concentration, pH and urea consumption.

2. $CaCO_3$ Identification by separating precipitate from solution. Then, the precipitate was filtered and weighed. SEM and EDX were used to identify the precipitate.

3. Studying the rate of $CaCO_3$ biomineralization by using the best concentration, obtained from the previous experiment, to mix with the concrete. The concrete was shaped into 10 x 10 x 10 mm and placed in water openly with free-flow wind and sunlight for 23 days. The rate was determined by comparing the crack of the first and the last day with Logger Pro 3.9.



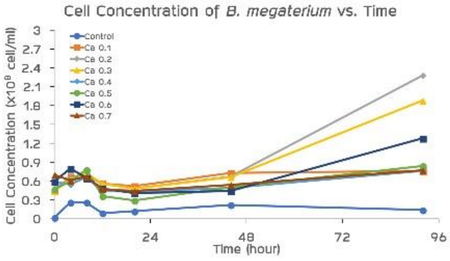
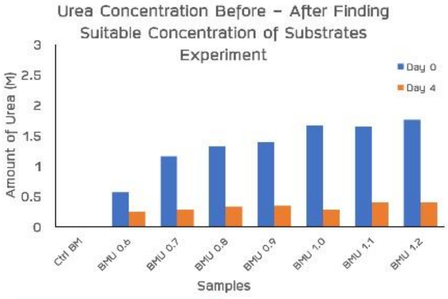


Figure 2 : The graphs show pH, cell concentration and urea consumption of *B. megaterium* which was monitored for 5 days



Conclusion

In the experiment of finding suitable concentration of substrates for *B. licheniformis*, *B. megaterium* and *B. subtilis*, the results showed that the concentration of calcium chloride in the range of 0.1 – 0.7 M and urea concentration in the range of 0.6 – 1.2 M did not influence the $CaCO_3$ production. Also, the monitoring of urea consumption could prove that bacteria consumed urea.

For studying the rate of $CaCO_3$ biocementation, *B. subtilis* caused the most precipitation of $CaCO_3$ in the limited time. This could be concluded that this bacterium could be applied into the cracked cement.

This project could potentially be an alternative way of repairing concrete in the future. However, some health effects of using the bacteria in repairing concrete should also be concerned.

References

Mian Luo., et.al. 2015. **Factors affecting crack repairing capacity of bacteria-based self-healing concrete**. Construction and Building Materials, 87, 3.

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