Biocatalytic carbonatogenesis: A continued substudy on trace metal supplements for optimized urease activity

By: Ryan Featherstone, Undergraduate

Supervisors: Justin Whitaker M.Sc & Dr. Danielle Fortin

Department of Earth Sciences - University of Ottawa

Faculty of Science

Faculty of Science

Rationale for research

A big problem that not many people think about today is liquefaction. This phenomenon can result in billions of dollars of damage per year. This occurs mainly because of earthquakes; the soil loses its strength and essentially falls apart due to increased pore water pressures.. As a result, buildings fall through the ground, cars fall into sink holes, and bridges collapse.

Adapazari, Turkey (August 18, 1999)
Liquefaction Damage

The main way to avoid potential damage right now is a process called grouting. However, this is inefficient as it is expensive, permanent, and toxic. Science has now discovered biocatalyzed carbonatogenesis which supports all three of these problems positively.

Prior knowledge

Previous experiments show that urease positive bacterium undergo biocatalyzed carbonatogenesis. The novel strain, *Sporosarcina ureae,* has proven to be a potential solution to everyday grouting techniques. Ideally, S Ureae can be used, but also optimized for this process.

Objectives

1) Further prove the ammonium production of S Ureae.
2) Compare it to other strains of bacteria (B. Megaterium, B. Subtilis, B. Sphaericus, as well as Jack bean urease which is the positive control).
3) Trace metal supplements of nickel, iron and cobalt (10 - 100uM) were then added to experiment to test different conditions.
4) Optimize ammonium production.

Methodology

Each bacteria culture is grown overnight to OD600 in a yeast extract media (10g/L Yeast Extract - 0.1M NH4Cl - 8.5g/L NaCl - 10g/L Trizma Base - pH = 8.0) . They are then harvested and washed twice in a buffer (10g/L Trizma, 8.5g/L NaCl, pH 8.0) and then diluted back to to OD600. It is then diluted with a 2:1 ratio to double the previous yeast extract media. This is then maintained in a Pyrex Bottle. Each trial takes five hours and a sample is taken every thirty minutes. These are then diluted to ~15000x through serial dilutions. Next the concentration of ammonium is measured by the HACH salicylate method. The higher the concentration of ammonium, the better potential for carbonatogenesis.

Results

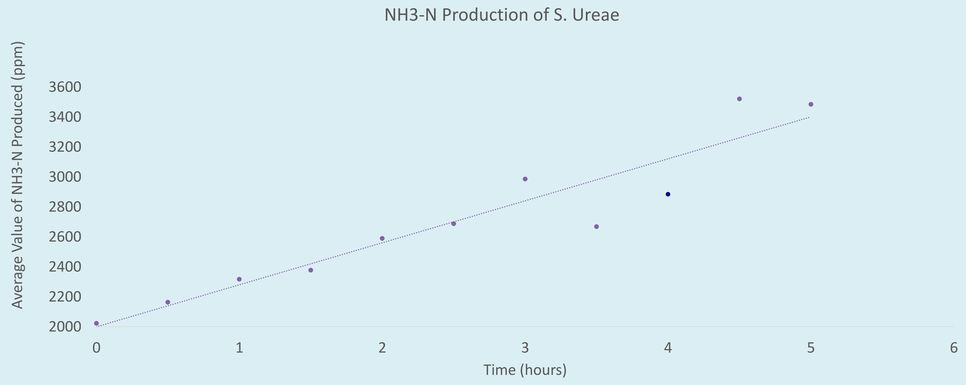

Figure 1. Ammonium production (ppm) over time for S. Ureae.

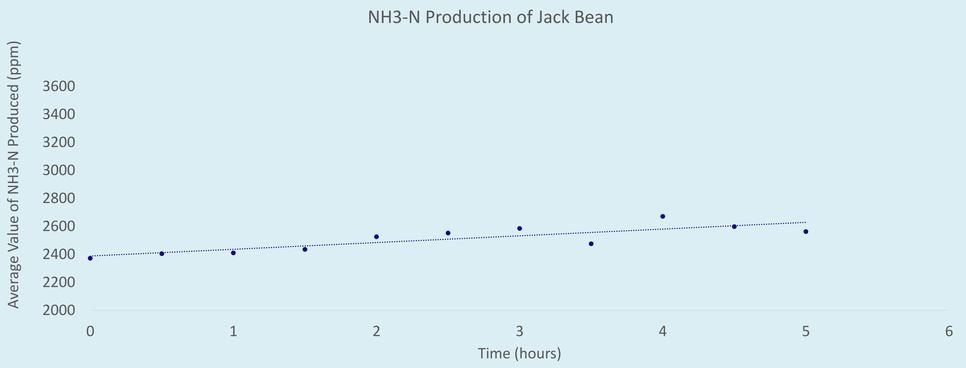

Figure 2. Ammonium production (ppm) over time for Jack Bean enzyme (control).

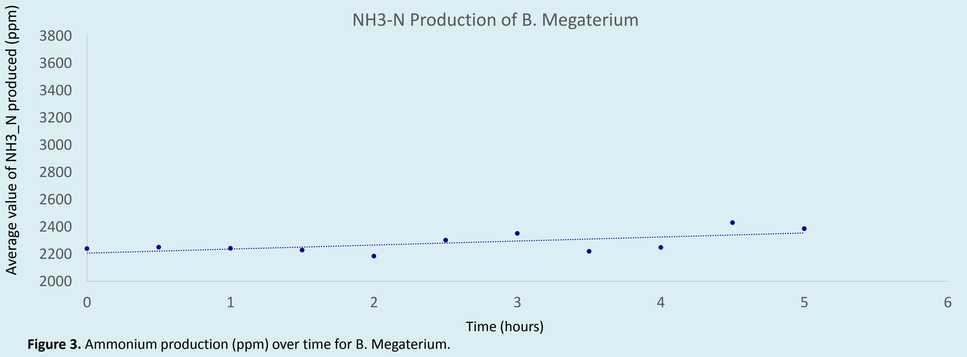

Figure 3. Ammonium production (ppm) over time for B. Megaterium.

Extended results

This project is not yet finished and more results are pending. In addition to two more strains of bacteria, multiple trials are being done for each strain to ensure repetitive results.
Once this is accomplished, the strains will be subjected to different metal supplements in order to see their reactions.
By the end of this, we will know if there is a better strain than S. Ureae and if we can optimize it in a certain condition.

Conclusion

According to the current data, S. Ureae has show positive results. By analyzing the graph, it is obvious that S Ureae increases its ammonium production significantly over time. The positive control and B. Megaterium show minimal increasements showing that they will not be effective in grouting. (2)

References

(1)Carter, E.L., N. Flugga, and R.P. Hausinger. *Metallomics*. 1.3 **2009**: 207-221.
(2) HACH, Inc. "Salicylate Method - Nitrogen, Ammonia (0.50mg/L)." HACH DR2700 Technical Manual. **2013**.
(3) Cheng, L.; Cord-Ruwisch R. *Ecological Engineering* **2012**, 42, 64-72
(4) Mann, S. . Biomineralization: principles and concepts in bioinorganic materials chemistry . Oxford, United Kingdom: Oxford University Press, **2001**. Print.

Acknowledgments

This project was made possible due to the UROP committee for the funding, Dr. Danielle Fortin for the lab space and guidance, and Justin Whittaker, M.Sc, for the supervision.