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## Systematic screening of microbial induced calcite precipitation kinetics via online monitoring

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Microbial urease catalyses the hydrolysis of urea to ammonium and carbonate, which results in an increase of the environmental pH value. Addition of calcium ions then leads to calcium carbonate precipitation. Microbial Induced Calcite Precipitation (MICP) is successfully applied for, e.g., restoration of construction materials, soil reinforcement, or metal and radionuclide bioremediation. However, the precipitation process requires further optimization to make industrial application of MICP more efficient.

A high precipitation rate of  $\text{CaCO}_3$  in the pore space of consolidated sand samples is necessary to increase the compressive strength. Multiple parameters as e.g. the urea and calcium ratio and concentration have been described in literature to have an influence on the precipitation process. However, most studies do not monitor the precipitation reaction itself, but perform application experiments on sand matrices and only check for the outcome data, typically compressive strength or the calcite content. Nevertheless, a small number of studies do try to derive strategies to improve the overall MICP process by getting an insight on the calcite precipitation kinetics and/or the crystal formation. These studies can mainly be grouped in two categories; there are simple beaker experiments with frequent manual sampling, as well as more advanced microfluidic experiments. Besides other disadvantages, both experimental strategies fail to provide a high amount of data points from a large number of parallel set-ups. Therefore, only the screening of individual parameters can be evaluated and possible interactions between parameters are disregarded.

Here, a new high-throughput microplate assay is presented, enabling online monitoring of calcite precipitation kinetics with a measurement interval of only 150 seconds. This assay was realised by making use of the automated high-throughput microbioreactor BioLector, which is able to measure a backscatter signal of 48 wells of a microplate in parallel while shaking at high speeds. The backscatter signal, intended for biomass estimation, corresponds to the turbidity in each well. When bacterial suspension and a cementing solution containing urea and a calcium source are mixed, calcium carbonate forms and precipitates, causing the backscatter signal to increase over time. As multiple precipitation kinetics can be measured in parallel by this system, the influence of multiple parameters on the precipitation rate can be easily compared. Interactions of multiple parameters influencing the MICP kinetics can be described as well by applying a Fractional Factorial Design (FFD) experimental approach.

In this study, the parameters OD600, pH, urea- and calcium concentration, type of calcium salt and culture washing were analysed. Three settings, which showed distinct calcite precipitation kinetics, were chosen to be adapted for quartz sand cubes solidification experiments to find a correlation between compressive strength and the precipitation rate. The results showed that very fast as well as delayed calcite precipitation is disadvantageous to solidify samples with high compressive strength.

Overall, the microbioreactor system can be successfully used to measure increasing suspension turbidity. This enables an easy systematic screening of a multitude of parameters influencing the precipitation rate and could help to optimize MICP applications, e.g. for building restoration to improve porous or deteriorated building components.

### Participation

In-Person

## References

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