



Interaction of heterotrophic bacteria extracted from CO₂ injection pilot site with diopside surfaces: A combined macroscopic kinetic and AFM approach

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The interaction between bacteria and basic silicates is among the main challenges in the predictive understanding of environmental factors controlling the weathering of Ca-Mg-silicates and its associated CO₂ consumption, as well as the water-rock interaction at CO₂ storage sites. In order to better understand the molecular and microscopic mechanisms of mineral surface interaction with heterotrophic aerobic bacteria extracted from natural settings, we used a combined macroscopic and microscopic approach to study the interaction of diopside (CaMgSi₂O₆) with typically heterotrophic aerobic bacteria extracted from deep underground water (HK31, 1700 m depth and $t = 25\text{--}30\text{ }^{\circ}\text{C}$) of a basaltic aquifer located within the Hellisheiði CO₂ injection pilot site (Iceland).

Diopside (CaMgSi₂O₆) was chosen as a typical basalt mineral, which is widely considered in carbon dioxide sequestration scenarios. Dissolution experiments were performed in constant-pH (5 to 9), bicarbonate-buffered (0.05 M) nutrient-diluted media in batch reactors at 0-30 bars of CO₂ in the presence of various biomasses of *Pseudomonas* reactants. The release rates of magnesium, calcium and silica were measured as a function of time in the presence of bacteria exometabolites as well as living, actively growing, and dead cells. Both nutrient media diluted 10 times (to 100 mg DOC/L) and inert electrolyte (NaCl, no DOC) were used. Our preliminary results indicate that the pH and dissolved organic matter in the form of cell exometabolites are the first-order parameters that control the element release from diopside at far from equilibrium conditions. SEM investigation of reacted surfaces reveal surface roughening associated with a much stronger biofilm formation in the presence of living bacteria compared to experiments with dead biomass.

A detailed AFM study allowed an in-situ high-resolution characterization of the status of the diopside surfaces in the presence of living and dead bacteria. *Pseudomonas reactans* extracted from a CO₂ injection site and soil rhizospheric exopolysaccharide-forming bacteria *Pseudomonas aureofaciens* have been used for this purpose. We have observed strong colonization of diopside surfaces by a biofilm-like bacterial assemblage after 1-2 days of interaction in nutrient-rich media. Extensive production of exopolymeric substances precluded imaging individual bacterial cells within this thick biofilm coverage. In contrast, mineral interaction with bacteria in nutrient-free neutral electrolyte allowed recognizing individual cell attachment to the diopside surface. No preferential site of interaction such as steps, kinks or edges could be identified. Overall, this work allows better understanding of microbially-affected silicate dissolution in basaltic aquifers and provides new nano-level insights for quantifying the degree to which the heterotrophic bacteria are capable of modifying the dissolution rate of rock-forming silicates via surface biofilm formation at the environmental conditions of CO₂ storage.