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Role of Sulfate Reducing Bacteria During Microbial Dolomite Precipitation as Deduced from Culture Experiments

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Sedimentary dolomite is found throughout the geologic record, beginning with rocks as old as 3.5 Ga.. Precambrian carbonates are commonly dolomite, whereas in the present time dolomite is rarely formed at Earth's surface conditions. The origin of primary dolomite remains a subject of geological debate.

In this study, we investigate primary microbial mediated dolomite formation in Lagoa Vermelha, a shallow, hypersaline lagoon 100 km east of Rio de Janeiro, Brazil. Dolomite precipitates in the anoxic organic matter-rich layer at the sediment surface in association with bacterial sulfate reduction. Lagoa Vermelha can be considered as a modern analogue for ancient environments with conditions that may have dominated during certain periods in Earth's history when anoxia was more prevalent. In order to simulate this environment in the laboratory, the chemical, hydrological, and microbial characteristics of Lagoa Vermelha were analyzed. Using a multidisciplinary approach to study dolomite biomineralization, we propose to gain insight into the kinetic, hydrologic, and microbiologic factors that may have led to the formation of the mineral dolomite in the geologic past.

Monthly Lagoa Vermelha water samples, taken during an annual cycle, were chemically and isotopically characterized. The deuterium and oxygen isotopic compositions of the water vary significantly during the year, indicating seasonal meteoric inflow and subsequent evaporation to return the lagoon hypersaline conditions. Using the oxygen isotopic composition of chemically separated high Mg calcite and dolomite, the isotopic composition of the water, from which the minerals precipitated, could be back calculated. This revealed the annual timing and conditions of dolomite formation. Sulfate reducing bacteria (SRB) were isolated from the anoxic sediments of Lagoa Vermelha and cultured in a media with pH 7.5-8, salinity of 3.8%, and a Mg:Ca ratio of 6:1, similar to conditions in Lagoa Vermelha. The cultures were incubated non-shaking at temperatures ranging from 20 to 35 ºC. Mineral precipitation began after about two to three weeks. XRD analysis revealed that the produced minerals were a mixture of Ca-dolomites with up to 47 mol% Mg. Dolomite formed exclusively in experiments run with bacteria. Control experiments without bacteria produced no dolomite, or any other carbonate minerals. The relationship between the microbes and the minerals was studied using scanning electron microscopy (SEM). SEM revealed that biomineralization occurs mainly in bacterial colonies consisting of bacterial cells and exo-polymeric material. Bacterial cultures consistently formed a dolomite precipitate with a characteristic "dumbbell" morphology. Similar morphologic shapes are found in Lagoa Vermelha sediment.

To evaluate the more precise role of bacteria in the dolomite formation process, *in situ* as well as laboratory experiments were conducted using dialysis bags, which can isolate a portion of the natural or simulated environment within a controlled compartment. Molecules smaller than 12,000 mol.wt. can freely diffuse through the dialysis membrane, whereas bacteria and mineral precipitates are retained inside the dialysis bag. By separating bacteria from the sterilised natural or simulated environment in a separate compartment, we could study the influence of the SRB metabolism on dolomite formation. By means of control experiments with non-active and autoclaved dead bacterial cells, the importance of the cell membrane in the dolomite nucleation process was also investigated. The results demonstrate that SRB metabolism can alter or control the conditions of the enclosed environment resulting in dolomite precipitation at Earth surface temperatures. In addition, the living bacterial cells appear to be essential to initiate mineral nucleation. Dead bacterial cells and quartz grains do not function as nucleation seeds, even if there are living bacteria in the medium surrounding the compartment providing the optimal physico-chemical conditions for dolomite formation.

In summary, sedimentary dolomite can be considered a "fossil mineral" that may indicate bacterial sulfate reduction under specific conditions. Our results show that SRB do influence anoxic carbonate precipitation and early diagenesis, implying that micro-organisms may have played an important role in the evolution of carbonate sedimentation throughout the geologic time.