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Experimental modelling of Calcium carbonate precipitation in the presence of phototrophic anaerobic bacteria Rhodovulum sp.

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Carbonate biomineralization is considered as one of the main natural processes controlling CO2 levels in the atmosphere both in the past and at present time. Haloalcaliphilic Rhodovulum sp. A-20s isolated from soda lake in southern Siberia and halophilic neutrophilic Rhodovulum sp. S-1765 isolated from hypersaline water body in Crimea steppe represent a large group of phototrophic bacteria likely to be involved in CaCO3 formation in soda and saline lakes. These bacteria use organic substrates for non-oxygenic photosynthesis and thus may mediate CaCO3 precipitation without CO2 consumption in highly-saline, highly-alkaline, NaHCO3-rich solutions.
In order to provide the link between surface properties of bacteria and their ability to precipitate Ca carbonate, we used a combination of electrophoretic mobility measurements, surface titration and Ca ion adsorption using dead (autoclaved), inactivated (NaN3 – treated) and live cells at 25 °C as a unction of pH (3-11) and NaCl concentrations (0.01, 0.1, 0.5 M). Zeta potential of both bacteria is identical for active, NaN3-inactivated and dead cells at high ionic strength (0.5 M NaCl). The pH of isoelectric point is below 3 and zeta-potential decreases or remain negative up to pH 11. However, at lower ionic strength (0.1 M and 0.01 M NaCl) for live cells the potential increases towards positive values in the alkaline solutions (pH of 9 to 10). Similar to previous results on cyanobacteria (Martinez et al., 2009) there is a net increase in zeta-potential towards more positive values at pH = 10.4 for active cells. In order to better understand this phenomenon, experiments with different concentration of Ca2+ and HCO3- ions as well as experiments with live cultures in the darkness have been carried out. The presence in solution of Ca2+ (0.01 and 0.001 M) and the absence of light in experiment do not change significantly the potential of the cells. However, the presence in solution of HCO3- strongly reduces the zeta-potential of the cells.
To characterise the link between the rate of bacterial growth (biomass production) and the rate of CaCO3 precipitation, batch kinetic experiments were performed. These experiments were carried out in closed (anaerobic) bottles with initial concentration of calcium from 1 to 20 mM and from 5 to 20 mM bicarbonate. The biomass of cells, pH, [Ca2+] and [Alk] were measured as a function of time. Blank experiments (without cell or autoclaved cells) were always carried out. We found that the optimal conditions for both CaCO3 precipitation and biomass increase for the culture Rhodovulum sp. A-20s, is calcium concentration of 3 mM, whatever the concentration of bicarbonate (5, 10, 15 mM). Note also that for calcium concentration higher than 3 mM, the biomass production decreases. In the case of strictly anaerobic Rhodovulum sp. S-1765 bacteria, the optimal conditions for calcium carbonate precipitation is observed for the bicarbonate concentration of 10 mM, whatever the calcium concentration (3, 5, 10 mM).
Overall, the present study allows quantitative modeling of bacterially-induced CaCO3 precipitation. It helps to distinguish between the effect of cell surface functional groups, surface electrical charge, soluble organic matter and metabolic change of solution pH on the rate and nature of precipitating calcium carbonate solid phase.