

Rapid determination of a fundamental aspect of the biomineralisation process of diverse marine calcifying organisms via sequential dissolution experiments

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One of the most fundamental questions in understanding the biomineralisation process of an organism is: how are the ions necessary for calcification transported from the environment to the site of mineral formation? In the case of marine calcifying organisms, answering this is centred on determining where the biomineralisation process lies between the endmember possibilities of seawater transport to the calcification site versus the channelling or pumping of ions across cell membranes. However, this simple question remains unanswered in most cases because of the difficulty of noninvasively studying internal processes in shelled creatures. To avoid the problems associated with making *in situ* observations, we developed an alternative method based on the extraction of micro-fluid inclusions that are trapped in the shells of most marine organisms. If technically feasible, analysing the major element chemistry of these fluid inclusions should allow the presence or absence of seawater at the site of calcification to be simply determined. Yet, despite composing up to ~1 wt.% of the mass of the biomineral, fluid inclusions in marine CaCO₃ are typically too small to target with microanalytical techniques, thus requiring an alternative approach. We circumvented this issue by extracting fluid inclusions via the sequential dissolution of bulk shell fragments into ultra-pure water. Based on these results, we show that the presence or absence of seawater at the calcification site can be fingerprinted using the chemical composition of these leachates coupled with multi-element ratio mixing models. The simplicity of the approach means that it offers the potential to allow the basic process involved in ion transport to the biomineralisation site of any fluid inclusion-containing biomineral to be quickly determined.