

Exploring the role of symbionts in coral calcification using genes, geochemistry, micro-CT and nutrient manipulations

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Most reef building tropical corals host photosynthetic symbionts, and it has long been observed that such corals exhibit higher rates of calcification by a factor of ~3 during the daytime – a phenomena known as light-enhanced calcification (LEC). However, the processes behind this behaviour remain hotly debated even though they underpin our ability to accurately predict the future of such corals, and the important ecosystems they construct, in the face of multiple anthropogenic stressors. Current hypotheses [1] fall into two camps: (i) abiotic explanations that revolve around the removal of inorganic carbon by the symbionts, influencing pH and ultimately affecting carbonate deposition; (ii) biologically-mediated explanations where calcification is enhanced by the symbionts providing energy and/or material for skeletogenesis.

Here we aim to find support for either of these hypotheses by exposing the coral *Stylophora pistillata* to variable quantities of inorganic nutrients, nitrate (N) and phosphate (P) that, consistent with previous studies, induced coral bleaching [2, 3]. The experiments were run for ~8 weeks, and during this period, we observed no significant impact of variable nutrient concentrations on the rate of calcification. Micro-CT confirmed there were no significant morphological differences in the skeletons constructed. However, in all the low P treatments [3] (in the presence or absence of replete amounts of N), the corals bleached by losing >60% of symbionts.

By combining transcriptomics with a geochemical study of the trace element composition of the skeleton (Li, B, Na, Mg, P, S, Sr, Ba, U) and a boron isotope-based reconstruction of the calcifying fluid pH and carbonate system, we are able to rule out abiotic mechanisms for light-enhanced calcification. Instead, we propose symbiont bleaching in the low P treatments results from the corals consuming their symbionts (c.f. [2]) which, along with an upregulation of phosphoric ion transport, ensured sufficient energy and material supply to maintain calcification mechanics. This confirms that LEC arises predominantly from biologically-mediated processes.

[1] Galli, G. and Solidoro, C. (2018) *Frontiers in Marine Science*, 5:68, doi: 10.3389/fmars.2018.00068

[2] Wiedenmann, J. D'Angelo, C., et al. (2023) *Nature*, doi: 10.1038/s41586-023-06442-5

[3] Rosset, Sabrina, et al. *Marine pollution bulletin* 118.1-2 (2017): 180-187