Biogeosciences Discussions

Biogeosciences Discuss., author comment AC1
https://doi.org/10.5194/bg-2021-140-AC1, 2021
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Reply on RC1 in bold

Valentina Alice Bracchi et al.

Author comment on "A stable ultrastructural pattern despite variable cell size in *Lithothamnion corallioides*" by Valentina Alice Bracchi et al., Biogeosciences Discuss., https://doi.org/10.5194/bg-2021-140-AC1, 2021

Reviewer comment:

The paper contains a detailed and well-illustrated description of the structure of calcified cell walls in specimens of L. corallioides from different settings. The paper shows that skeletal ultrastructure of L. corallioides does not change in environments substantially different in terms of illumination (depth), temperature, and salinity. Based on the maintenance of the same ultrastructural patterns in different environmental conditions the authors conclude that 'the calcification process of CCA seems to be biologically-controlled rather than induced'. As suggested in the introduction, the aim of the paper is to contribute to the debate about the nature of calcification in coralline algae. Its results support that calcification is biologically controlled and, therefore, refute the conclusion of Nash et al. (2019) that mineral formation in corallines is biologically induced. The latter authors, however, based their interpretation on a detailed discussion of several features of calcification of corallines and any rebuttal of their conclusion should address the same features on the light of the new findings. I believe that a discussion of features that according to Nash et al. (2019) are key to decide whether the mineral formation is induced or controlled must be included in the paper.

Dear reviewer, thank you for your revision.

The study of coralline algae represents in general a challenge for scientist, and the mechanisms of calcification are far from been understood.

The paper of Nash et al. (2019) hypothesized a time-step scheme of biomineralization based on the observation of a wide dataset (24 species) which considers both crustose coralline algae (non-geniculate) and articulated (geniculate) corallines. In the light of their observations, they not only hypothesize such model, but also support that the biomineralization in coralline algae is a biologically induced process basing upon a series of key-assumptions.

The biomineralization is the process of formation of a mineral phase carried out by organisms. It depends on the degree of biological control over the formation (Lowenstam, 1981; Weiner and Dove, 2003, Päßler et al 2018) ranging between two mechanism: the biologically controlled mineralization (BCM), in which organisms have extensive control over the mineral formation, or biologically induced mineralization (BIM), in which organisms have no to minor control over the mineral formation. BCM results in well-ordered mineral structures, with

minor size variations and species-specific crystal habits (Bazylinski and Frankel, 2003), whereas BIM results in heterogeneous mineral compositions with poor crystallinity, including large size variations, poorly defined crystal morphologies and the inclusion of impurities (Banfield and Hamers, 1997; Frankel and Bazylinski, 2003; Weiner and Dove, 2003).

Our study is aimed at describing, in a standardize way (longitudinal section), the main ultrastructural features, which represents the result of biomineralization, of cell wall in *L. corallioides.* Having collected the same species but from difference geographic context and water depths, we also test if this translates in ultrastructural differences at level of calcite crystals in the cell wall. Basing upon our observation, we proposed an ultrastructural pattern for *L. corallioides.* In the light of our results referring to ultrastructures (well-ordered, minor size variations, species-specific crystal habit), we support that the biomineralization in coralline algae is biologically controlled rather than induced, perfectly adhering the original definition.

In the case of Nash et al. (2019) layers of the cell wall (and not ultrastructures) and their mutual organization are described using figures in which algae are apparently randomly oriented and this is the first problem in comparing our results. Nevertheless, we found some similarities between our results and theirs, in term of layers forming the cell walls (primary, secondary), thickness of cell wall, structure of grains, but also substantial differences (not only PCW cell). These results, similarities and differences, referred to morphological units characterizing the cell wall, will be more discussed in the revised version of the manuscript. It was not our scope to propose another hypothetic scheme of biomineralization, nor rebut to all the assumptions the authors did in their paper, but we will insert some specific rebuttals, which concern only the mineralogical aspects in the revised version of the manuscript.

Minor points: Check species names are in italics.

We will check it carefully.