

This is my second review of the manuscript *Impacts of seawater sulfate concentration on sulfur concentration and isotopic composition in calcite of two cultured benthic foraminifera* by Thaler *et al.*, which presents S/Ca and  $\delta^{34}\text{S}$  measurements of two strains of rosalinid foraminifera cultured under different seawater  $[\text{SO}_4^{2-}]$ .

The authors have, in my view, adequately addressed all of the comments from the first round of review, resulting in an improved version of what was already a very interesting and comprehensive piece of work. I have some further minor comments for the authors to consider but would suggest that they do so at their own discretion.

### **Main comment**

1. Reproduction/growth in culture. If I understand what is written on lines 79 and sections 2.1.2/3.1, dead individuals (empty shells, or those not attached to the petri dishes) were removed, leaving only live foraminifera. Is it therefore possible that foraminifera that did not die but remained dormant could be present in the final analyses? In most cases this is of course not important, as there was a large increase in the number of individuals during the experiment. A possible exception to this is the 60 mM experiment, in which the number increased from ~30 to ~100. Given that the interpretation of the  $[\text{SO}_4^{2-}]_{\text{sw}}\text{-S/Ca}_{\text{shell}}$  plateau hinges on this experiment, I suggest adding a note to explain whether or not this datapoint unambiguously does not contain pre-experiment shell material, or if it could represent a mixture of shell material grown under normal seawater and experimental conditions (which could then be an alternate explanation for the plateau if the foraminifera from reproduction in the experiment were smaller). On a similar note, Table 1 gives the numbers of foraminifera in each experiment through time, but how many were discarded during the experiment? Does this provide evidence for multiple generations? Even in the 60 mM  $[\text{SO}_4^{2-}]$  experiment?

### **Minor comments**

1. Lines 26-29. An inhibitory kinetic effect would seem a more likely possibility to me, but I appreciate not every hypothesis can be listed in the abstract.
2. Line 51. Sulphur and magnesium are not trace elements in seawater.
3. Lines 58 and 451. I suggest using a different phrase to 'large volcanic events' as this possibly implies single eruptions, whereas Laakso *et al.* discuss large igneous provinces emplaced over thousands of years.
4. Lines 75-76. You could clarify that most studies that included material grown before culture attempt to account for this in some way, e.g. using size-mass relationships or labels.
5. Lines 117-118. Are the units mM or mmol/kg?
6. Lines 211-212. Were all samples run at the same concentration as the seawater standards? If not, does this approach potentially result in a reproducibility that is too low?
7. Line 246. Please clarify which results you are referring to.
8. Lines 258-259. I think this explanation is unlikely (e.g. the DIC had increased by day 5 (Figure. 6) and there was no further increase.
9. Section 4.1 title. Bear in mind that there were large covariations in seawater  $[\text{Na}^+]$  and  $[\text{Cl}^-]$ . Worth mentioning in the discussion?
10. Section 4.2. At some point (in the introduction?) it would be helpful to mention the Mg/Ca of these foraminifera if it is known. If they reduce the Mg/Ca of the biomineralisation site compared to seawater then the considerations regarding the effect of seawater  $[\text{SO}_4^{2-}]$

on  $\text{CaCO}_3$  nucleation and precipitation will likely not apply/be more complicated than implied in some places in the manuscript (e.g. lines 338, 407).

11. Line 338. On a similar note, I would suggest rephrasing this sentence. There may have been no precipitation in those experiments but it does not mean it is not possible, e.g. if higher degrees of oversaturation were to be achieved. Likewise, I would not read anything into the 'fact that calcite precipitates' (line 369).
12. Line 355. Please clarify, the same as what? *Heterostegina*?
13. Lines 416-418. Given that you include speciation modelling, can you say which ion pairs become relatively more abundant?
14. Line 720. It also diffuses between the experiment and atmosphere.

### **Typos**

1. Line 65. Change 'interrogates' to 'suggests'.
2. Lines 137. Experiments.
3. Line 184. Aliquots, or delete 'of the'.
4. Lines 314-315, 390. Change four instances of 'Mm'.
5. Line 693. Forms.
6. Line 728. On a pool of a hundred to...'
7. Line 816 and Fig. D1, it should be  $\text{CaHCO}_3^+$  (also  $\text{NaSO}_4^-$ ).

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clear variables

% requires a phreeqc installation
iphreeqc = actxserver('IPhreeqcCOM.Object'); % create PHREEQC COM object
% load desired database (pitzer for seawater)
dirP = uigetdir('C:\Program Files\USGS','Select directory containing PHREEQC databases');
iphreeqc.LoadDatabase([dirP '\pitzer.dat']);

% SO4, Na, and Cl co-vary in these experiments
SO4in = 0:0.1:180;
NaIn = 479:-(479-402)/(size(SO4in,2)-1):402;
ClIn = 612:-(612-175)/(size(SO4in,2)-1):175;

outputData = NaN(size(SO4in,2),6);
for i = 1:size(SO4in,2)
    IPCstringCell= {'SOLUTION 1', ...
        ['-temp ', num2str(22)], ...
        '-units mmol/L', ...
        '-density 1.025', ...
        ['-pH ', num2str(8.2)], ...
        ['Ca ', num2str(10.3)], ...
        ['Mg ', num2str(53)], ...
        ['B ', num2str(0.4)], ...
        ['K ', num2str(10)], ...
        ['Br ', num2str(0.8)], ...
        ['S(6) ', num2str(SO4in(i))], ...
        ['Na ', num2str(NaIn(i))], ...
        ['Cl ', num2str(ClIn(i))], ...
        ['Si ', num2str(0)], ...
        ['Sr ', num2str(0.1)], ...
        ['P ', num2str(0)], ...
        ['F ', num2str(0.1)], ...
        ['C(4) ', num2str(4)], ...
        'SELECTED_OUTPUT', ...
        '-molalities CO3-2 HCO3- CO2 MgHCO3+ NaHCO3 CaHCO3+ MgCO3 NaCO3- CaCO3', ...
        '-activities CO3-2 HCO3- Ca+2 NaHCO3 NaCO3- SO4-2', ...
        '-SI calcite anhydrite gypsum celestite', ...
        'soln false', ...
        'pH true', ...
        'sim false', ...
        'state false', ...
        'time false', ...
        'step false', ...
        'pe false', ...
        'distance false'};
    IPCstring = sprintf('%s\n', IPCstringCell{:});

    iphreeqc.RunString( IPCstring );
    OUTphreeqSTRING = iphreeqc.GetSelectedOutputArray;

% retrieve the data
loc = find(strcmp(OUTphreeqSTRING, 'm_CO3-2(mol/kgw)'));
outputData(i,1) = OUTphreeqSTRING{2,(loc+1)/2};
loc = find(strcmp(OUTphreeqSTRING, 'si_calcite'));
outputData(i,2) = OUTphreeqSTRING{2,(loc+1)/2};
loc = find(strcmp(OUTphreeqSTRING, 'si_anhydrite'));
outputData(i,3) = OUTphreeqSTRING{2,(loc+1)/2};
loc = find(strcmp(OUTphreeqSTRING, 'si_gypsum'));
outputData(i,4) = OUTphreeqSTRING{2,(loc+1)/2};
loc = find(strcmp(OUTphreeqSTRING, 'la_SO4-2'));
outputData(i,5) = OUTphreeqSTRING{2,(loc+1)/2};

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loc = find(strcmp(OUTphreeqSTRING,'si_celestite'));
outputData(i,6) = OUTphreeqSTRING{2,(loc+1)/2};

end

% plot Omega calcite, anhydrite, gypsum, celestite
close(figure(1))
figure(1)
plot(S04in,10.^(outputData(:,2)))
hold on
plot(S04in,10.^(outputData(:,3)))
plot(S04in,10.^(outputData(:,4)))
plot(S04in,10.^(outputData(:,6)))
set(gcf,'color','w')
xlabel('[SO_4^{2-}] (mM)')
ylabel('\Omega')
legend('calcite','anhydrite','gypsum','celestite',...
'location','southeast','fontsize',8)
set(gca,'yscale','log')
ylim([1e-2 20])

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