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Biogeosciences Discussions

Interactive comment on "Latitudinal trends in stable isotope signatures and carbon concentrating mechanisms of northeast Atlantic rhodoliths" *by* Laurie C. Hofmann and Svenja Heesch

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We would like to thank the anonymous referee for their constructive and helpful comments. Our detailed responses to the reviewers' comments are provided below. The revised version of the manuscript is attached, and we present our revised figures (Figs. 4, 5 and 7) based on the reviewer comments.

We would like to confirm that we did not bleach our samples for total ∂13C analysis. We acknowledge that some studies have bleached their samples to remove organic

material in the past. However, the specific citation referred to (Lee & Carpenter 2001) only used a weak bleach solution to remove surficial organic matter. Additionally, we did not find any evidence of treating samples for stable isotope analysis with bleach in studies using coralline algae as historical marine proxies (e.g. Hetzinger et al. 2009, Williams et al. 2011, Bougeois et al. 2015), which are the studies we compare our results to with respect to the total ∂13C analysis.

pH drift experiment

The pH drift experiments contained stir bars and were conducted on a magnetic stir plate with multiple magnets. We have added the statement (page 3, lines 8-9) "The jars contained stir bars and were placed on a multi-position stir plate. The rhodoliths were held on custom-made stands inside the jars to allow space for the stir bars below." We have removed the section on the carbon use model, considering that, as the reviewer mentioned, the pH change was much lower than would be expected, and it is possible that a 24 hour was not long enough to reach the pH compensation point. We address this on page 3, in lines 15-17 by stating ""pH drift experiments showed that the seawater pH actually decreased after a 24 hour light incubation for both Phymatolithon calcareum and P. purpureum (Figure 8). However, these samples were small, and it is possible that the incubation period was not long enough to detect a significant change in pH."

We have also repeated pH drift experiments for rhodolith samples from Greenland and the Canary Islands, the two extreme latitudes investigated in this experiment. We have changed Figure 8 to accommodate the new data, and add all necessary information to the methods, results and discussion accordingly as detailed below.

Page 3, Lines 9-16: The incubations were conducted during a 24 – 70 hour light cycle with 35 μmol photons m-2 s-1 at 15°C, 20°C or 4°C (for the Irish, Canary Islands, and Greenland samples, respectively) in natural seawater (34 psu) in 200 - 300 ml glass or plastic jars, depending on the size of the specimens. The duration of the light

exposure depended on the specimens. The Greenland specimens were incubated longer due to their slower metabolic rates. The seawater was vacuum filtered using 0.22 μm Durapore membrane filters (Merck Millipore, Darmstadt, Germany). The jars contained stir bars and were placed on a multi-position stir plate. The rhodoliths were held on custom-made stands inside the jars to allow space for the stir bars below. The start and finish pH values (NBS) were recorded using a Sentix 51 pH electrode with an integrated temperature probe connected to a WTW 3110 pH meter (Weilheim, Germany). The seawater used during the incubation was left open to the ambient air for re-equilibration to make sure the change in pH was due to the metabolism of the algae.

Page 5, Lines 15-20 "pH drift experiments showed that the seawater pH actually decreased after a 24 hour light incubation for both Phymatolithon calcareum and P. purpureum (Figure 8). However, these samples were small, and it is possible that the incubation period was not long enough to detect a significant change in pH. In comparison, the Canary Islands samples elevated the seawater pH up to 9.07. The Greenland samples also increased the seawater pH up to 9.7, but the seawater pH did not return to ambient levels after being exposed to the atmosphere. After 5 days, the seawater pH was lower than when the rhodoliths were present, but still higher than the starting pH value. The Greenland samples produced high amounts of dissolved organic carbon during the incubation period (data not shown), which could have strongly affected the pH compensation point, and suggests that pH drift experiments for these specimens are not reliable methods for determining the pH compensation point."

Page 5, Lines 29-31: "The pH drift experiments also support the hypothesis that most rhodoliths investigated have an active CCM involving HCO3- uptake, since several individuals from the highest and lowest latitude investigated had pH compensation points above 9.0."

Figure 8. The A) pH compensation points (maximum pH reached during pH drift experiment) and B) delta pH (final – initial) for the samples from Ireland (Phymatolithon spp.

from Mannin Bay and Carraroe) the Canary Islands, and Greenland (Akia Penninsula).

Light experiment

As this experiment was not a major aspect of the study, and we did not separate new growth from old growth, we have removed any reference to it from the manuscript (see also comment below).

Page 1, line 17: Thank you for pointing this out. The referee is correct that this statement does not support our conclusions and is not accurate according to our data. We have revised the statement as follows:

"We observed a decreasing trend in δ13CT signatures with increasing latitude and temperature, while δ13Corg signatures were only significantly correlated to DIC. These data suggest that high latitude rhodoliths rely more on CO2 as an inorganic carbon source, while low latitudes rhodoliths likely take up HCO3- directly, but none of our specimens had ∂13Corg signatures less than -30, suggesting that none of them relied solely on diffusive CO2 uptake."

Page 2, line 5: We have rephrased the statement to: "Therefore, the ratio of stable carbon isotopes (∂13C) in macroalgal tissue can be used as an indicator of whether or not HCO3- is being used (Raven et al., 2002)"

Page 2, line 26: As mentioned above, this made our results more comparable to studies that investigated using coralline algae stable isotopes as marine proxies.

Page 3, line 8 onewards: More specific information on the experimental materials has been added. Because we removed the section on the carbon use model, we also removed the statement on total alkalinity analysis. The text now reads "The incubations were conducted during a 24 – 70 hour light cycle with 35 μmol photons m-2 s-1 at 15°C, 20°C or 4°C (for the Irish, Canary Islands, and Greenland samples, respectively) in natural seawater (34 psu) in 200 - 300 ml glass or plastic jars, depending on the size of the specimens. The duration of the light exposure depended on the speci-

mens. The Greenland specimens were incubated longer due to their slower metabolic rates. The seawater was vacuum filtered using 0.22 μm Durapore membrane filters (Merck Millipore, Darmstadt, Germany). The jars contained stir bars and were placed on a multi-position stir plate. The rhodoliths were held on custom-made stands inside the jars to allow space for the stir bars below. The start and finish pH values (NBS) were recorded using a Sentix 51 pH electrode with an integrated temperature probe connected to a WTW 3110 pH meter (Weilheim, Germany). The seawater used during the incubation was left open to the ambient air for re-equilibration to make sure the change in pH was due to the metabolism of the algae."

Fig 4 – the legend now includes the abbreviated species names

Fig 5 – the units have been changed to μmol Kg SW-1 and we have added a map of surface ocean temperature with the mean d13C and d13Corg of Lithothamnion spp. plotted at their respective collection sites.

Fig 7 – panel labeling has been added

Page 5, Line 37: we have changed the wording to "directly take up HCO3-"

Page 6:

Line 30 - The reference to this experiment has been removed, since we have also removed the data from the supplementary, and because we did not separate new growth from old growth.

Line 34 - The link between DOC & ∂13C has been more clearly explained by this statement: "In fact, there may be a relationship between ∂13C signatures and dissolved organic carbon (DOC) availability in rhodolith beds, since rhodolith food webs depend strongly on external inputs of organic matter (Grall et al. 2006, Gabara 2015), and the biogeochemical cycling within the rhodolith bed food web influences isotopic signatures."

Cornwall et al. 2015 has been added to the reference list

Please also note the supplement to this comment:
https://www.biogeosciences-discuss.net/bg-2017-399/bg-2017-399-AC1-supplement.pdf

Interactive comment on Biogeosciences Discuss., https://doi.org/10.5194/bg-2017-399, 2017.

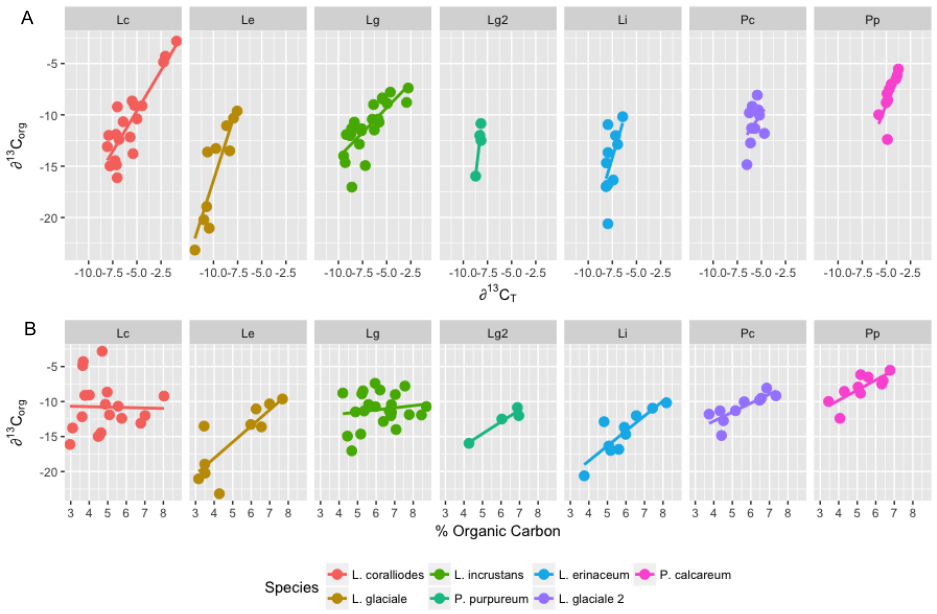
A
Lc
Le
Lg
Lg2
Li
Pc
Pp
$\partial^{13}C_{org}$
$\partial^{13}C_T$
B
% Organic Carbon
Species
L. coralliodes
L. incrustans
L. erinaceum
P. calcareum
L. glaciale
P. purpureum
L. glaciale 2

Fig. 1.

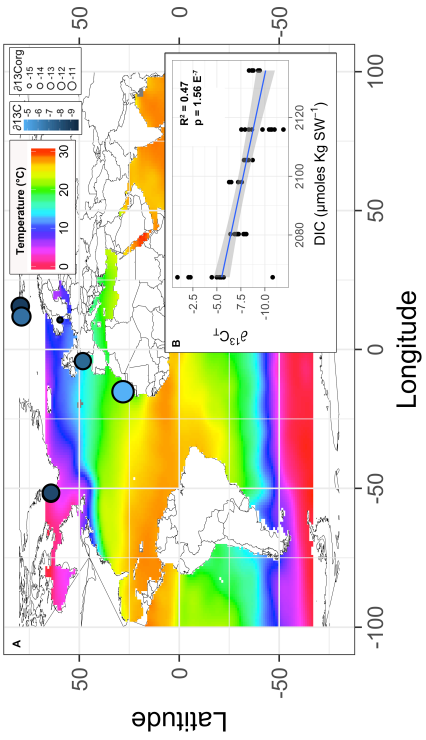
Latitude
Longitude
Temperature (°C)
∂13C
∂13Corg
R^2 = 0.47
p = 1.56 E-7
DIC (µmoles Kg SW^{-1})
$\partial^{13}C_T$
A
B

Fig. 2.

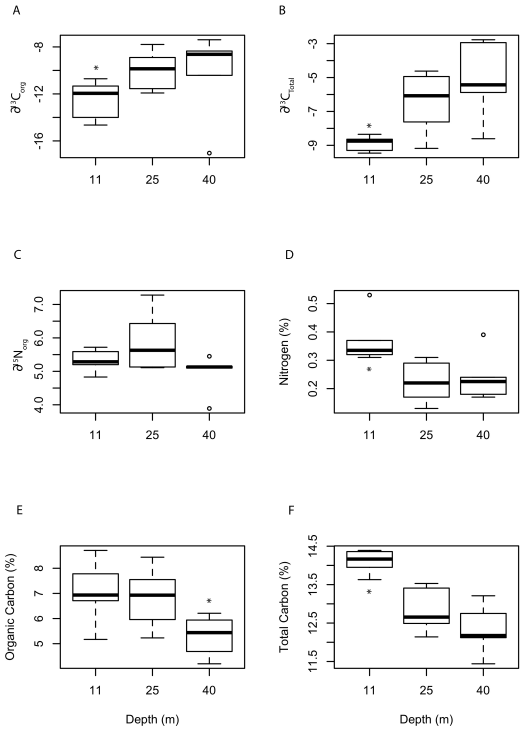
A
B
C
D
E
F
$\partial^{13}C_{org}$
$\partial^{13}C_{total}$
$\partial^{15}N_{org}$
Nitrogen (%)
Organic Carbon (%)
Total Carbon (%)
11
25
40
Depth (m)

Fig. 3.