

Dear Referee, thank you for your endeavour in revising the paper and for your suggestions. Some of your concerns have already been addressed in the previous review. Nevertheless, we made a further effort in clarifying those issues in the newly revised manuscript.

- **Comment:** Having read the previous reviews and the paper I think the paper needs to be considerably reworked. The concept certainly merits publication, but as per the previous reviews the raw data really needs to be shown, for all sites. The authors need to see it from the readers point of view, we are presented with lots of plots but not all of the data that we need to understand the authors assertion that growth rather than temperature controls B/Ca.

Response: As already mentioned in the previous review, we agree on the importance of showing the raw data and, besides adding Table 2 in the previously revised version, we submitted the dataset in Pangaea, an open access repository. They are now publicly available at the following link: <https://doi.pangaea.de/10.1594/PANGAEA.932201>.

- **Comment:** Lots of the plots leave me with questions, why do I only get one out of 4 sites plotted for certain plots? Why is the entire sampling interval using in one panel and the third panel dividing the data between short and long cells (Figure 11)?

Response: Most of the plots show the elemental analyses in the samples coming from all the sampling sites. Figure 8 shows the correlation plots between B/Ca and temperature proxies for the Morlaix sample only, because for the other samples there was a non-significant correlation (lines 252-255). The age model with the reconstruction of temperature variations during the algal growth has been presented for the Morlaix sample only. This choice has been made due to the meaningfulness of this sample for the scope of the paper. Indeed, besides having the best resolution of the growth bands among the samples, allowing the reconstruction of an age model reasonably accurate, the Morlaix sample was the only one where we found a correlation between temperature proxies and B/Ca. Since the purpose of the work was to investigate potential evidence of temperature and growth rate effects on B/Ca, we have welcomed the previous suggestions of the Referees to show the trace element variations at higher resolution, to check for seasonal temperature influences. We do not see the need to show the same for the Mediterranean samples in the present paper, since it would not add significant information to the conclusions. The manuscript has been implemented to justify this choice.

Concerning your second question, as explained in the responses of the previous revision, and strongly recommended by all the Referees, we decided to modify Figure 11 to show more clearly the overall relationship between B/Ca and seawater temperature. Besides ΔT , which is meaningful for the comparisons of the sites at different depths, we therefore plotted the maximum and minimum temperature values per site with B/Ca mean values in long and short cells, respectively produced in the warm and cold periods. In the panel you see no relationship between B/Ca and the warm/cold season. The Morlaix sample also in this plot represents an exception that has been discussed in detail above. Acknowledging your comments, to make our use of temperature data clearer, we provided a more detailed explanation in materials and methods.

Major comments

- **Comment:** Why is only the Morlaix site chosen for plotting the comparisons between different variables and not the others? The authors need to show for all samples the transect of laser ablation values vs. length. I would envision this as a plot with several panels, first panel elemental ratio vs depth. Then a panel showing the position of the growth bands or at least where they

consider where the growth lines sit, overlaid with this the relative estimated growth rate for each band. That is the basic dataset, then plot as a scatter the data for each laser ablation point (i.e., the elemental value) vs growth rate, at the minute the authors are comparing the average growth rate over 8-11 years with the average B/Ca.

Response: Morlaix was chosen for the age model because of its significance to evaluate the temperature influences and because it was the one with the best visibility of the bands, as extensively explained in the previous response. As already commented in the first review (Referee#3), it was not possible to infer the growth rates of single bands with an acceptable error. The method used to measure growth rates, i.e. counting the number of growth bands crossed by the laser transect, implies a non-negligible error margin and increasing in resolution as you suggest would increase the error and the data would lose reliability. Although it would be very interesting to see these results, the approach you suggest is not feasible with these samples.

- **Comment:** Looking at figure 12 there is 'cyclical' nature of B/Ca and there is varying growth between the white and grey bands. Surely doing this would strengthen the authors argument that growth rate affects B/Ca?

Response: We believe that the cyclicity you observe in B/Ca is more likely related to the differences in growth rates between cold and warm season, because of the high amplitude of seasonal fluctuations in temperature which characterize the site of Morlaix (highest ΔT), as we already discussed in the previous version of the paper (lines 342-346). White and grey bands are indicated to facilitate the interpretation of the plot, as done previously by other authors (e.g. Halfar et al., 2000); nevertheless, as you see in the image of the section, it does not strictly correspond to the width of growth bands. As already discussed, we are not able to define the different growth rates in each band with an acceptable approximation. As you might expect by looking at the thallus of *L. corallioides* in Fig. 1, the growth of this alga is not unidirectional neither perfectly regular. We cut the sample longitudinally along the expected growth direction of the branches, and we performed the analysis on the best resolved portion of the section. Nevertheless, the differences in the band width are not necessarily related to the growth rate but can also be due to changes in the growth direction.

- **Comment:** Plot all combinations of elemental ratios for all sites, as one figure per site, include this in the supplementary file if it's too many figures.

Response: The complete dataset is available at Pangaea repository, as already mentioned. Moreover, in the previous revision we added Table 2 with the summary of all elemental ratios among sites. As you suggested, in the new revision we added the Figure S2 in the supplement also showing Li/Ca, Sr/Ca and Mg/Li among sites.

- **Comment:** Then the authors can assign the time interval for each ablation point. Rather than plotting min and max temperature for particular years (which led me to ask: "Why does time on the plots vary between February, March, August and September in Figure 7" as this is not referred to) just plot the full temperature time series.

Response: The maximum and minimum values of Mg/Ca represent warm and cold periods of growth (Hetzinger et al., 2009), thus they should correspond to the highest and coldest seawater temperature. This method is widely used for the temperature calibration in coralline algae since it avoids uncertainties from sub-annual dating (Moberly, 1970; Corrège, 2006; Williams et al., 2014; Caragnano et al., 2014; Ragazzola et al., 2020). In the timeline reported in Fig. 12, the warmest

(August/September) and coldest (February/March) months of the temperature time series have been reported, as already wrote in the caption.

- **Comment:** Where are the growth bands of the Elba and Aegadian island? Having seen the supplement images I assume that the Mg/Ca is used almost exclusively for those samples. I note that the authors mention that such data is excluded, but I learn only at line 301 that means the entire sample for Elba. This should be discussed much earlier.

Response: In the Elba sample the poor resolution of the growth bands did not allow us to clearly distinguish between the elemental signal in long and short cells. Thank you for your suggestion, this information has been also added in the Materials and Method section (2.5). As we already wrote in this section, the growth band counting was done directly under the light microscope, where it was much easier to see the bands by adjusting the light. The images in the Supplement, required by the previous Referee, were added to show the longitudinal section crossed by the laser transect. We still tried to adjust the image contrast and brightness in the revised Fig. S1 to better highlight the bands.

- **Comment:** Why gap fill the data in Figure's 7 and 12? We cannot then see which is the measured and which has been gap filled.

Response: The image resolution during laser ablation forced us to take approximations while targeting the visible growth bands, therefore some of them may not have been targeted by the laser spot. Gaps have been highlighted by asterisks in the revised plots (Fig. 7, 12).

- **Comment:** Figure 11 shows there is no apparent relationship between temperature and B/Ca but then Figure 12 shows a similar pattern between B/Ca and temperature. So is the figure 11 not the results of comparing an average with an average? Considerable effort by the authors has gone into getting the ORAS5 temperature data, so I am at a loss as why they then bundle it up into a single average.

Response: The laser ablation resolution does not allow us to precisely discriminate the month of year the analysis is referring to. Therefore, we cannot attribute a single point of analysis to an absolute temperature in a specific time of the year, but rather refer to the cold and warm season, as done for the creation of Fig. 7 and 12. This method is broadly used in literature to assess coralline algae age model (Moberly, 1970; Corrège, 2006; Williams et al., 2014; Caragnano et al., 2014; Ragazzola et al., 2020).

The one collected in Morlaix was the only sample where the B/Ca has significant correlations with temperature proxies (Fig. 8), this is one of the reasons why we decided to reconstruct a detailed age model for this sample. There are fluctuations in B/Ca over time (Fig. 12), but very differently from valuable temperature proxies. We finally explained the correlation of B/Ca with Mg, Sr and Li/Ca in this sample in terms of changes in growth rate that are likely to happen because of the high temperature fluctuations experienced by the alga throughout the year (line 352-356), which make the difference from the other sites. In this paper, we show several results and elaboration from various sampling sites in different ways, with the aim to investigate the growth rate and temperature relationship with B/Ca in *L. corallioides*. In the Mediterranean samples, we did not see any evidence of temperature control over B/Ca. Because of these results we suggest a stronger control of growth rates.

- **Comment:** Abstract/Conclusion – incorporate, or mention, new Li/Mg
Response: Thank you for the suggestion, the change has been made.
- **Comment:** Line 29 – “their longevity by indeterminate growth, with no ontogenetic trend”, what do you mean by longevity by indeterminate growth? – Ok, I see this is answered in line 30, perhaps split the sentence. ‘As well as having longevity by indeterminate growth, that is the growth trend...’
Response: OK
- **Comment:** Line 74: Replace ‘actually’ with ‘ For instance, more recently’
Response: OK
- **Comment:** Line 92 – 105: Feel like some of the parts feel unconnected, would reword to something like, to ensure that there is flow: “Here, we present the first laser ablation inductively coupled plasma mass spectrometry (LA-ICP-MS) conducted on coralline algae. This technique, which allows high-resolution analysis of a broad range of trace elements in solid-state samples, has been widely used in biogenic carbonates to extract records of seawater temperature, salinity, and water chemistry (Schöne et al., 2005; Corrège, 2006; Hetzinger et al., 2009, 2011; Fietzke et al., 2015; Ragazzola et al., 2020). Measurements were made on the non-geniculate coralline alga *Lithothamnion corallioides* (P. Crouan & H. Crouan) P. Crouan & H. Crouan 1867 which is widely distributed in the Mediterranean Sea and in the north-eastern Atlantic Ocean, from Scotland to Canary Islands (Irvine and Chamberlain, 1999; Wilson et al., 2004; Carro et al., 2014), usually constituting maerl beds (Potin et al., 1990; Foster, 2001; Martin et al., 2006; Savini et al., 2012; Basso et al., 2017). It forms rhodoliths as unattached branches (Basso et al., 2016) with obvious banding in longitudinal sections (Basso, 1995b). These characteristics combine to make this species a suitable model for the measurement of geochemical proxies, comparing different environmental settings. In this paper, we provide the first LA-ICP-MS data of temperature proxies (Mg/Ca, Sr/Ca, Li/Ca) and B/Ca were measured on *L. corallioides* collected from different geographic settings and depths across the Mediterranean Sea and in the Atlantic Ocean. We test the influence of temperature and growth rate on the B/Ca ratio, which could be crucial in assessing the reliability of B/Ca as a proxy of the seawater carbonate system.”
Response: The text has been revised according to the suggestions. Note that this is not the first LA-ICP-MS on coralline algae in general, but the first on a coralline alga grown in nature in two different Basins.
- **Comment:** Line 154/Table 1: I note that Table 1 no longer has the time interval of the ORAS5 datasets – so I assume that all of the 11 year intervals no longer go beyond the collection date? As per reviewer 3
Response: Exactly, as mentioned in the response to Reviewer #3, the time interval extracted for Aegadian Isl. has been corrected in the first revised version. The time interval considered are clearly visible in this newly revised version as suggested (Supplement Fig. S3, S4, S5, S6).

- Comment:** Line 157: “Missing element ratios were calculated as the means of known values” is it not easier to remove the years corresponding with the known? How do you ensure that your gap-filling doesn’t introduce error/uncertainty?

Response: We believe that introducing the mean values of the coldest and warmest months is a reasonable approximation. Moreover, the resulting plot shall be easier for the reader. The few gaps have also been highlighted in the plot as asterisks, following what you suggested in your previous comment.
- Comment:** Line 160-165: If there is a trend in pH changing through time, is it not prudent to use earlier years than the full range 1993-2018? The authors need to plot the DIC, temperature, and pH values through time for each site in a supplementary figure, identifying which periods were used in their analysis.

Response: Very recently, the CMEMS products used for the environmental data reconstruction have been updated. We therefore decided to update our work accordingly. The new references have been indicated in “Data availability”. The time collection of our samples goes from 1990 to 2017, therefore using earlier years did not seem a proper solution. As mentioned to the previous reviewers, DIC data are more sparse, and we did not have the same consistency as for temperature data. We therefore decided to choose a reasonable time period, the same for every sampling site for data comparability. Now, as you better see in the supplementary figures you suggested to add (Fig. S3, S4, S5, S6), the range of variations did not change much through time, and the data well characterize the sites. Thanks to the updated version of the DIC data, we were able to extend the extracted period to 1999-2017. For consistency, we revised the period of extraction of pH data as the same (1999-2017). As you can see from the revised Table 1, the values have slightly changed and the differences among sites remained almost the same. We updated temperature data as well, still with very small changes. Figures 7, 10 have also been revised according to the new data.
- Comment:** Line 170: “This step was helpful in highlighting faint bands and to achieve a more reliable estimate of the algal growth.” Identify which, if any, bands were identified via Mg/Ca or whose position was modified by this method.

Response: Mg/Ca data have been used to help identifying the poorly visible bands of the Elba sample while measuring the growth rates. The text has been revised for clarity because we understood that the sentence created misunderstandings.
- Comment:** Line 172: “Intermediate Mg/Ca values, indeed, would probably correspond to middle seasons” – if long and short cells are produced in cold and warm seasons, and also intermediate are middle seasons, how reliable is the assumption then that each long and short cell can be assumed to be a season allowing for estimate of the year by counting backwards? Why were these measurements excluded from the dataset?

Response: No measurements have been excluded from the dataset. As mentioned in the response above, Mg/Ca results have been used to aid the identification of growth bands in the Elba sample, where, as you see in the Supplement (Fig. S1), the growth bands were very poorly visible. In the other samples, the growth bands have been identified under light microscope as wrote in Materials and Methods. The text has been revised.

- Comment:** Line 179: “Data from faint bands had been excluded from the dataset.” – why? In the Elba image there are no growth bands visible so were all of these excluded? Likewise Aegadian Island. Note – I now see that is the case but this should be mentioned here.

Response: As mentioned before, the text has been revised in sections 2.4 and 2.5. It was not possible to distinguish clearly long and short cells in the Elba sample because of the poor resolution of the growth bands. Therefore, when analysing the elemental data from short and long cells, the Elba sample is missing. As you suggested, this information has been added in section 2.5. Concerning the measurements of growth rates, we directly observed the section under a light microscope, which was much more effective in visualizing the bands rather than the image analysis (see 2.4). Given the poor resolution of the bands in Elba, and since the record of Mg/Ca was relatively continuous, we also used Mg/Ca results to help measure the growth rate in this sample. The text has been revised for clarity.
- Comment:** Table 1: the caption states: “Data from monthly means extracted by 11 years of ORAS5 reanalysis. pH and DIC in each sampling site are also indicated. The minimum, maximum, mean, and standard deviation values have been measured on the time interval 2019-2020.” Why isn’t the pH interval included? I.e., 1993-2018. Is it just DIC covered by the interval 2019-2020?

Response: The caption has been revised adding the pH interval as well.
- Comment:** Figure 6: Why plot the average of the sites long and short cells for your data? In figure 7 the authors plot temperature and corresponding Mg/Li, why then not do this for this figure? I note that in Figure 7 the Mg/Li of Morlaix would cover nearly the full range of the figure 6’s y-axis.

Response: We reconstruct the age model for the Morlaix sample only, for the above-mentioned reasons. Nevertheless, Fig. 6 is no less significant. Indeed, as extensively mentioned in the previous responses and in the last revision, the resolution of the laser ablation does not allow us to attribute a single point of analysis to an absolute temperature in a specific time of the year, but rather refer more generically to the cold and warm seasons. In fact, dark and light bands are usually associated to cold and warm periods, and we have used this estimation to create the plots. When plotting the data collected from all the samples (Fig. 6), we refer to the maximum and minimum temperature registered in the site, corresponding respectively to the long and short cells. When plotting the age model of the Morlaix sample, we always refer to the coldest and warmest month of the year, with the respectively dark and light bands (Fig. 7, 11). As mentioned previously, we added a more detailed explanation on the use of temperature data for the purpose of the paper in Materials and Methods, section 2.3 (lines 157-169).
- Comment:** Figure 8: This should be for all sites and not just Morlaix, I do not get the rationale for showing only a single site.

Response: The correlation was statistically significant only in the Morlaix sample (line 252).
- Comment:** Line 273-279: The samples have 8, 10, 11 and 11 years of growth, when then (figure 11) was 11 years of temperature compared for all?

Response: The values of growth rates are approximate due to the limit of the method and the complexity of the algal growth behaviour. For the comparability among sites, we decided for a time interval which almost surely included the whole length of the laser transects. Of course, when

reconstructing the age model of the Morlaix samples, the years indicated effectively correspond to the laser spot.

- **Comment:** Figure 11: Why split the data into short and long cells only for the temperature dataset? Why not plot the growth rates and deltaT against the short and long cells?

Response: Short cells conventionally correspond to the minimum temperature registered in the sites; long cells correspond to the maximum temperature (Moberly, 1970; Corrège, 2006; Williams et al., 2014; Caragnano et al., 2014; Ragazzola et al., 2020). We do not have the growth rates for each single band for the reasons explained above. We believe that our use of temperature data is much clearer in the newly revised text; deltaT plotted against short and long cells would not be meaningful.

- **Comment:** Line 301 : “In the sample from Elba the growth bands were not clearly visible, preventing the analyses of trace elements in long and short cells separately” – this shouldn’t pop up in the discussion but should be referred to way earlier.

Response: We implemented the Materials and Methods section 2.5.

References

Caragnano, A., Basso, D., Jacob, D. E., Storz, D., Rodondi, G., Benzoni, F. and Dutrieux, E.: Coralline red alga *Lithophyllum kotschyianum* f. affine as proxy of climate variability in the Yemen coast, Gulf of Aden (NW Indian Ocean), *Geochim. Cosmochim. Acta*, 124, 1–17, doi:10.1016/j.gca.2013.09.021, 2014.

Corrège, T.: Sea surface temperature and salinity reconstruction from coral geochemical tracers, *Palaeogeogr., Palaeoclimatol., Palaeoecol.*, 232, 408–428, doi:10.1016/j.palaeo.2005.10.014, 2006.

Moberly, R: Microprobe study of diagenesis in calcareous algae, *Sedimentology*, 14: 113–123, doi: 10.1111/j.1365-3091.1970.tb00185.x, 1970.

Halfar, J., Zack, T., Kronz, A. and Zachos, J. C.: Growth and high-resolution paleoenvironmental signals of rhodoliths (coralline red algae): A new biogenic archive, *J. Geophys. Res.*, 105: 22,107–22,116, doi: 10.1029/1999jc000128, 2000.

Hetzinger, S., Halfar, J., Kronz, A., Steneck, R., Adey, W. H., Philipp, A. L. and Schöne, B.: High-resolution Mg/Ca ratios in a coralline red alga as a proxy for Bering Sea temperature variations from 1902–1967, *Palaeo*, 24, 406–412, doi:10.2110/palo.2008.p08-116r, 2009.

Ragazzola, F., Caragnano, A., Basso, D., Schmidt, D. N. and Fietzke, J.: Establishing temperate crustose Early Holocene coralline algae as archived for paleoenvironmental reconstructions of the shallow water habitats of the Mediterranean Sea, *Paleontology*, 63, 155–170, doi:10.1111/pala.12447, 2020.

Williams, B., Halfar, J., Delong, K. L., Hetzinger, S., Steneck, R. S. and Jacob, D. E.: Multi-specimen and multi-site calibration of Aleutian coralline algal Mg/Ca to sea surface temperature, *Geochim. Cosmochim. Acta*, 139, 190–204, doi:10.1016/j.gca.2014.04.006, 2014.