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Biogeosciences
Discussions

Interactive comment on “Effect of ocean acidification and elevated temperature on growth of calcifying tubeworm shells (*Spirorbis spirorbis*): An *in-situ* benthocosm approach” *by* Sha Ni et al.

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We thank the reviewer for his supportive comments and the constructive review of our manuscript. Below we respond to the reviewer’s specific comments.

Reviewer general comment 1: Split manuscript. 1. growth and population dynamics. 2. calcification and shell corrosion.

***Response G1*: We prefer to keep all of this information in a single, comprehensive manuscript because understanding ontogenetic growth and population dy-**

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namics is important for interpretations of calcification rates.

Reviewer general comment 2: Add a picture of the benthocosm system.

***Response G2*: A photograph showing the benthocosm setup will be added (see attached Fig. 1).**

Reviewer general comment 3 and specific comment 2: Mesocosm experiments have limited control and higher natural variability than laboratory experiments (e.g. food availability, salinity, day length). This requires additional discussion, specifically about the normalisation for day length (insolation index), which is a critical seasonal parameter.

***Response G3/S2*: We agree with the reviewer that laboratory experiments are better controlled than in-situ mesocosms. However, the latter allow to consider the dynamics under near-natural boundary conditions needed to understand the reactions on manipulations like temperature and PCO2 under otherwise in-situ conditions. Only a comparison between laboratory and in-situ experiments can finally provide a solid frame to understand adaptations of natural communities to the expected changes in coastal ecosystems.**

We will replace the "simplified insolation index" by the daily insolation sum (in kWh/m2) measured at the Meteorological Station of Geomar, which is situated very close to the benthocosm site (Fig. 2). This parameter implicitly includes day-length. Please note that we used this parameter only to explain the measured diurnal pH and saturation state changes and their variations between different seasons. We did not relate tube worm growth directly to insolation, but compared it to measured pH.

On the other hand, the strong seasonal variability makes it very difficult to compare the growth rate results between different seasons. This is discussed in Section 4.5 of our manuscript. Further possible conclusions about seasonal im-

pacts on Spirorbis growth are limited by the one-year duration of our study. We have no seasonal replicates from different years to compare.

The following paragraph discussing the issues of limited environmental control will be added to the manuscript (Section 4):

As shown in Figs. 3 (revised in Fig. 2) and S2 there was strong intra- and inter-experimental variability in several environmental parameters, most prominently temperature, insolation, pH and saturation state, but also salinity and nutrient availability. Further, food supply and faunal/floral composition varied during the experiments as discussed below (Section 4.5) and shown in Werner et al. (2016). This natural variability is an intentional part of the benthic mesocosm set-up as it allows to consider the dynamics of benthic communities reacting to environmental changes under near-natural boundary conditions (Wahl et al., 2015, 2016). On the other hand, the lack of control on several environmental parameters also has drawbacks for the interpretation, comparability and reproducibility of results from different seasonal experiments. As described in Section 2.3, we use the term "seasonal factors" to collectively describe variations of experimental conditions between the four experiments, including environmental parameters and the ontogenetic development of S. spirorbis. While some of these factors are clearly dominated by seasonal change (e.g. light, temperature), others may vary on different time scales. Without multi-annual replicates we can not prove the seasonal nature of the observed changes in S. spirorbis growth between the four experiments. We therefore use the term "seasonal" as a simplifying descriptor of inter-experimental changes, although their seasonal nature needs to be verified in future multi-annual experiments.

Reviewer specific comment 1: Reproduction occurs between spring an summer. Therefore the authors are comparing not just juveniles and adults, but different generations that can have different sensitivities due to different starting (acclimation) conditions.

***Response S1*: This is a very good remark. According to published work on Spirorbis and our own results reproduction occurs in several intervals between late spring and autumn. We agree that the different conditions during the initial growth of each generation may cause different acclimation with respect to, e.g., saturation state. This could result in a better pH tolerance of the late summer and autumn generations that started to grow under lower pH conditions than the late Spring generations. Unfortunately, as stated in the comments of VBSC Chan and in Response G3/S2, we have no replicates for the seasonal experiments, as our study covered only one year. Therefore, our data do not allow to draw conclusions about different acclimation at different seasons.**

Reviewer specific comment 2: see general comment 3.

***Response S2*: See response G3/S2 above.**

Reviewer specific comment 3: Did the authors check if all specimens were alive at the end of the experimental period?

***Response S3*: Fucus with attached worm tubes was collected from the basins and freeze dried immediately after the experiments. Complete worms with original (red) colour were visible in many tubes, indicating that they had been alive until freeze drying.**

Reviewer specific comment 4: Were alkalinity changes due to increased salinity taken into account? Full carbonate chemistry details should be provided.

***Response S4*: Alkalinity was measured frequently together with salinity. Alkalinity values of all experiments and treatments were published in Wahl et al. (2015: Fig. 9). In our manuscript alkalinity and salinity are shown in diagrams of the Supplement (Fig. S2). Measured alkalinities were used to calculate the calcite saturation states in the basins (as described in Section 2.5 of our manuscript). We can refer more frequently to Wahl et al. (2015), where appropriate, to better**

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explain the data base for our carbonate chemistry calculations.

Reviewer specific comment 5: Michaelis-Menten type kinetics for the light dependence of diurnal pH variations.

***Response S5*: We agree that a Michaelis-Menten fit is more appropriate than the simple linear fit. We find a reasonable fit to our data and have modified Figure 4 and the text in Section 3.1 accordingly (see Fig. 3 and Figure caption). In accordance with the previous linear fits, the Michaelis-Menten fit of the high-CO2 treatments shows a half saturation constant that is almost twice the constant of the normal CO2 treatments (1.6 and 0.9, respectively). This indicates a continuing increase of pH amplitudes in the latter under high-light conditions. The corresponding rate constants for normal and high CO2 are very similar, 0.5 and 0.6, respectively.**

Reviewer specific comment 6: Specimen at high CO2 and elevated temperature grew twice as much as the specimen from control conditions. This seems counter-intuitive.

***Response S6*: We agree with the reviewer. However, as shown in Fig. 14, average growth between different winter treatments was not significantly different. As shown in Fig. 12, individual growth rates varied in a wide range in all treatments. So with the high variability observed, growth in the high-CO2 treatments in single specimens can be expected to exceed growth in low-CO2 treatments. Indeed this is illustrated in Figure 9: growth of a single specimen does not represent general (average) growth in a treatment. Note that the specimen shown in (a) had an initial diameter of about 3 mm, while (b) was only about 2 mm wide at the start of the experiment. A shorter tube segment was added to the larger specimen. This is in line with the growth trend shown in Fig. 12. We can add a remark in the figure caption to better describe that high variability of growth: Figure 9. Pristine and corroded S. spirorbis shells.... Note that the specimen in (a) had a larger initial diameter than the specimen in (b), but grew a shorter new**

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tube segment during the experiment.

Reviewer specific comment 7: Units of the y-axis missing in Fig. 14. Juvenile specimens in Fig. 14 are equivalent to “autumn small population”. This should be noted in the caption.

***Response S7*: The y-axis shows growth divided by initial diameter, which is dimensionless (mm/mm). We will add the units (mm/mm) in the diagram (Fig. 4). The figure shows the whole autumn population (small and big). This size difference was only significant in the “small” sub-population (not shown). It was insignificant in the “big” sub-population. However, because the autumn population was dominated by the small sub-population the effect is still significant in the total population. We clarified the figure caption (see below).**

Revised Figure Captions

Figure 1: A. Two subunits of the Kiel Outdoor Benthocosm with open hood. Subunits with closed hoods are visible in the background on the right. B. Spirorbis spirorbis specimens attached to living brown alga Fucus vesiculosus. Juvenile (white dots, yellow arrows) and adult (white spires, red arrows) specimens of S. spirorbis are visible.

Figure 2: Average water temperature, daily insolation, pH and saturation state with respect to aragonite (as proxy for S. spirorbis Mg-calcite) in the four different treatments. Each of the four seasonal experiments is divided into four sub-periods lasting 17-19 days (start and end dates indicated at x-axis). Error bars indicate minimum and maximum values of the mean diurnal cycle during the sub-periods, except for insolation where they indicate day-to-day variability (standard deviation). Insolation was measured at the GEOMAR meteorological observatory (www.geomar.de/service/wetter), about 100 m from the benthocosms.

Figure 3: Light dependence of diurnal pH cycles. Average diurnal pH amplitudes in the benthocosm basins for CO2-enriched (left) and ambient (right) treatments plotted

versus the average daily insolation (as in Fig. 3) for the sub-periods of the four seasonal experiments. Dotted lines are Michaelis-Menten fits to the data, y=A*x/(B+x), with rate constants (A) of 0.5 and 0.6 and half saturation constants (B) of 0.9 and 1.6, for ambient and CO2-enriched treatments, respectively.

Figure 4: Average growth (Gr/Di) in different treatments during seasonal experiments. In autumn, growth differed significantly between the +T and control treatments. The effect is only significant in the "small" sub-population, while the "big" sub-population showed no significant temperature effect. However, the "small" sub-population dominates the autumn population. Thus the total population shows a significant temperature effect. In summer, no tubes were recovered from the elevated temperature treatments. Results from three-way ANOVA and Tukey's HSD tests; * significant difference (p <0.05).

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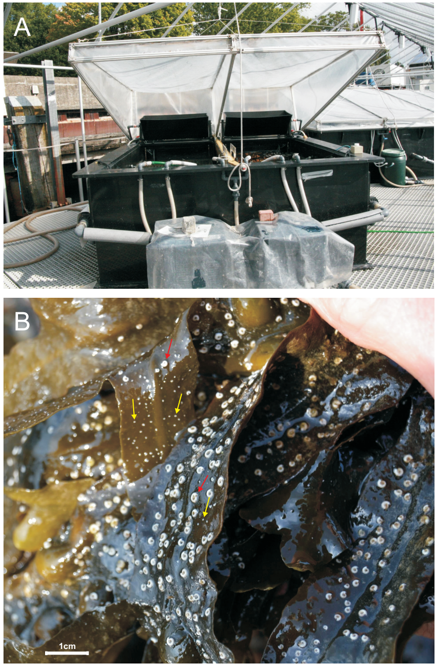

Figure 1

Fig. 1.

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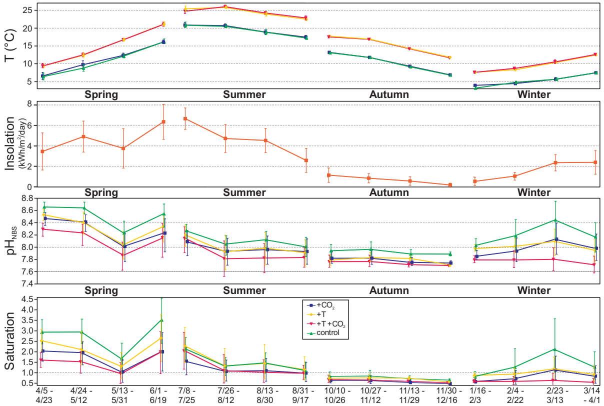

Figure 3

Fig. 2.

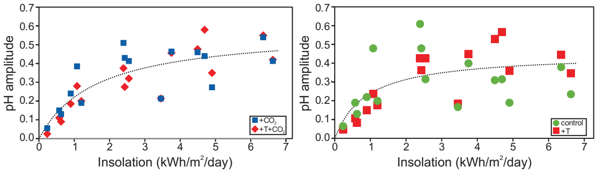

Figure 4

Fig. 3.

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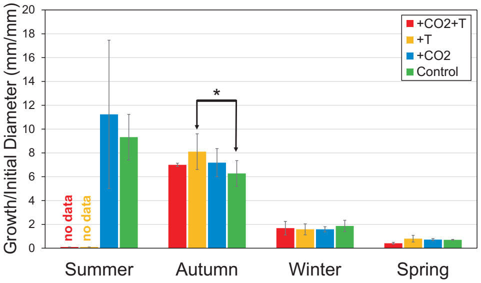

Figure 14

Fig. 4.