

## ***Interactive comment on “Experimental evidence for foraminiferal calcification under anoxia” by M. P. Nardelli et al.***

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Dear Editor Prof. Kitazato,

We thank both reviewers for their constructive comments and mainly positive feedback. We here address only the major points; we have followed most of their suggestions regarding grammar and technical details and feel no need to discuss those further.

Kind regards,

Maria Pia Nardelli and co-authors

Answer to referee #2, PhD Toyofuku.

Comment: "The calcification and dissolution of calcium carbonate can be expected by

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calcite saturation state  $\Delta\text{D}\text{e}$ . I try to calculate by CO2SYS program (Lewis and Wallace, 1998). According to Figs. 3, 4, A1 and A2, factors are assumed as  $T=17\text{ C}$ ,  $\text{Sal}=35$ ,  $\text{pH}=6.2$ ,  $\text{Alk}=10000\text{M}$  for experiment 1 and factors are assumed as  $T=12\text{C}$ ,  $\text{Sal}=35$ ,  $\text{pH}=7.5$ ,  $\text{Alk}=5500\text{M}$ .  $\Delta\text{D}\text{e}$  of experiment 1 was much lower than 1 (0.2) meanwhile  $\Delta\text{D}\text{e}$  of experiment 2 shows more than 2. It means calcite including fossils and newly calcified foraminifera chamber tend to be dissolved in the environment of experiment 1. Even it is not so strange that foraminifera can calcify their test under unsaturated condition for calcite, calcified crystal should be affected by the environment. According to Fig. 2, the test seems thinner than other older chambers. I wonder the preservation of test and surface textures of them. Further, the vertical pH profiles are not measured below 3 cm in experiment 1. Though we cannot estimate the, some specimen must add chambers. Some SEM photos may support to evaluate the effect of the lower pH environment. I am very interested the surface structures of the test what is precipitated under anoxic period."

Reply: We also tried to estimate calcite saturation state  $\Delta\text{D}\text{e}$  with the same software suggested by Dr. Toyofuku. Minimum values of  $\Delta\text{D}\text{e}_{\text{calc}}$  were estimated to be  $<1$  from 0.75 to 3 cm depth in the experiment 1 and approximately 2 at 0.25 cm in the experiment 2. We decided not to include these results in the manuscript because the cumulative error linked to the estimation based on absolute pH values measured using NBS buffer solutions (as mentioned in the ms) and the relatively low sensitivity of the alkalinity measures by colorimetry. However we analysed several specimens from both experiments in order to check for calcite anomalies under SEM. None of the analysed specimens of *Ammonia tepida* and *Cassidulina laevigata* showed obvious anomalies into calcite structure. Some specimens of *Bulimina marginata* showed partial dissolution of superficial calcite of old chambers (produced before the experiment) while new chambers, grown during the experiment, showed apparently normal structures. We have not measured the thickness of new chambers however we consider that as most of the specimens calcified only one chamber the thinner aspect observed by Dr. Toyofuku could be either due to anoxia or to the simple fact that last chamber is constituted

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by only one calcite layer, differently by older ones. A more detailed study of the calcite of new formed chambers is therefore needed before any complete discussion about these aspects. For this reason we plan to perform other SEM observations, including quantification of pore density and size in relation to oxygen concentration, and geochemical analyses on the new formed chambers. All these data will be subject of a future publication.

Comment: "In terms of growth rate, one to two chambers for 60 days would be slower. The oxygen concentration may not affect the metabolism of *A. tepida* according to the author. Perhaps culture temperature (16.5-20C) might not so bad temperature for their growth (Bradshaw, 1957). I can guess the specimens were already matured size from the beginning/the food materials are not enough for them. Do you have some consideration about the reason of slow growth?"

Reply: The specimens used in the experiment were of different sizes ranging from 150 to 350 micron, thus including juveniles (<10 chambers) and adults (until approximately 20 chambers). If the reason of the slow rate was the maturity we would have expected to obtain some specimens able to calcify several chambers and others (the oldest ones) showing lower calcification rates. It was not the case as calcification rates were generally quite low and the number of added chambers never exceeded one. For this reason we think that the general lack of organic matter and in particular the lack of fresh organic matter in the surface layer could better explain this result. We shortly discuss this point in the paragraph 4.1 of Discussion.

Comment: "The other hand, this 12C will be higher than temperature of in-situ environment of *Bulimina* and *Cassidulina*. Is there some reason about it?"

Reply: After the suggestion of the referee, we added a short explication in the materials and methods paragraph 2.3. The in situ temperature at the moment of *B. marginata* specimens sampling was close to 7°C, however we chose to perform the experiment at 12°C because it is suggested by Barras et al. (2009) to be the optimal temperature

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favoring the growth of *Bulimina marginata*.

Comment: "Could you find any changes of sediment (e.g. color, smell and others)?"

Reply: For both experiment the aspect of the sediment was accurately checked at the moment of core aperture (end of the experiment). For experiment 1 the sediment of the first layer was light brown, corresponding to the oxygenated level. Starting from the second layer the sediment varied progressively from light grey to dark grey/black at the bottom. Typical sulphide smell was often remarked at the bottom of the core, approximately starting from 8 cm depth. For experiment 2 the surface of layer 2 still showed some brownish spot, that matched with the expected weak oxygen penetration still at the surface of this layer (OPD= 0.5 cm, layer 1=0.3 cm). Downcore the sediment was light to dark grey. No sulphide smell was detected.

Comment: "Could you tell some morphological and/or geochemical features of newly added chamber under anoxic condition? The information must be useful for vast of users of proxy interpretations."

Reply: Please see answer to question 1.

Comment: "The legend might be uniformed between Fig A1 and A2."

Reply: We thank Dr. Toyofuku for this note. We corrected the legend after his suggestion.

Comment: "Even I understand the idea of  $\delta p\text{H}$ , what was the actual pH of top water?"

Reply: pH values of overlying waters were 8.29 and 8.22 ( $\pm 0.08$ ) respectively for experiment 1 and 2.

#### References

Barras, C., Geslin, E., Duplessy, J.-C. and Jorissen, F. J.: Reproduction and growth of the deep-sea benthic foraminifer *Bulimina marginata* under different laboratory conditions, *J. Foramin. Res.*, 39, 155–165, 2009.

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