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Interactive comment on "Impact of carbonate saturation on large Caribbean benthic foraminifera assemblages" *by* Ana Martinez et al.

Ana Martinez et al.

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Dear Anonymous Referee #2

Thank you for your very insightful comments which improved the paper. Below we address each comment (following each comment).

The paper by Martinez et al. describes a very interesting study in which natural variability in carbonate saturation state at submarine springs (ojos) is used to assess the benthic foraminiferal response to ocean acidification. The authors find that proximity to submarine springs impacts the benthic foraminiferal community. In particular, they find a decrease in overall abundances, but also that symbiotic calcareous species are less affected than non-symbiont bearing species, and agglutinated foraminifera may be least impacted. The paper is overall well written, and the conclusions are interesting. However, in some areas, the complexities and richness of the underlying dataset could be better served. Digging further into some of the complexities here should allow the authors to better support their current conclusions, but I suspect it will also lead to some more specific and novel results. Overall, there are three major (somewhat related) issues, which I would strongly urge the authors to address.

Reply: We thank the reviewer for noting that the paper is interesting and original and suggesting areas for improvement which we have addressed in the revised version.

Comment 1) First, is the assumption that proximity to "ojos" impacts benthic foraminifera entirely or primarily due to difference in calcite saturation state from the ambient environment. This does not seem like a foregone conclusion to me. These are essentially isolated regions of increased fresh-water influence in a marine context and could be different in a number of ways from the surrounding environment. The authors mention, but then rapidly dismiss, the salinity differences between the ojos and ambient environment (6:24-28). However to entirely dismiss salinity requires either a more detailed quantitative analysis to try to tease apart these covarying parameters, and/or a more in depth discussion of the known sensitivities of different foraminifera and communities to salinity. There could be several additional environmental differences, such as oxygenation, or changes in nutrient or metal concentrations from terrestrial sources that could produce sensitivities in some species (I might look into the literature on benthic foraminifera communities as tracers of metal contamination). Finally, there could be differences in benthic community or environment (substrate? Food source? predation?) between the ojos and control sites. All of this should be discussed.

Reply: Sensors deployed at these specific ojos determined that salinity is above 30 for 93% of the time, and when it drops below 30, it is for short periods of time of less than 1 hour and does not fall below 27 (Crook et al., Supporting Information, PNAS July 2, 2013 110 (27) 11044-11049; https://doi.org/10.1073/pnas.1301589110). We emphasize this and refer to the above study. In addition, we note that many of the

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species in our study have a very broad salinity tolerance range as typical to shallow coastal lagoon and estuary settings. We have included the salinity sensitivities of the major species in our study for which data is available. We also compared our results to results from laboratory experiments where only carbon chemistry is changed to support our idea that the carbonate saturation is the main driver of foraminiferal abundances we see in our field site. Regarding other variables, the ojo and control sites are just a few meters apart with identical substrate (coarse sand), water depth and light, and while we did not monitor at such close proximity we expect that predation and food sources are also similar. Regarding other chemical differences in the discharging water indeed there are small differences in nutrients and oxygen but not in trace metals. Specifically, ojo Norte has lower oxygen during very low tides and slightly more reducing conditions but there was nothing particularly unique about this ojo in terms of the trends observed. Moreover, the differences among ojos were smaller than the ranges of variability within each ojo (see also reply to reviewer #1). Finally, each ojo is slightly different but we do not have replicates of identical ojos so we cannot deduce statistical differences and attribute them to such conditions. As we write this is the nature of doing field work there are confounding variables, but you get more realistic results.

Comment 2) A broad view of how major groups of foraminifera respond within the community (symbiotic, agglutinated, etc.) is much needed and well served by the study design. However, it is a shame that it comes at the expense of a discussion of a species, clade, or more finely-defined functional group level response. This study would be more impactful if it also reported the species-level assemblages at each sites. Are there specific species or genera that appear more or less robust to the environmental differences between ojo and “control” environments? Such a discussion is especially warranted given the species-level differences in response to ocean acidification that have repeatedly been shown in culture studies.

Reply: We have included the contribution and trends in relative abundance of each genus, as well as abundances depending on test type (porcelaneous, hyaline, ag-

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glutinated), magnesium content (low, intermediate and high), feeding type (symbiont-bearing, symbiont-barren) and symbiont type (diatom and chlorophyte).

Comment 3) Finally, there appear to be clear differences between ojo/control pairings which are occasionally mentioned in passing, but never fully addressed. For example, looking at Figure 2, I am immediately greeted with some pretty basic questions such as "Why is there such a large difference in abundance at Mini and its control compared to Gorgos and its control?" and "What is different about Norte that the low-saturation abundance is as high as the high-saturation groups at other sites?" If saturation state is the primary driver of total abundance this should be an unexpected result! Without further information or context about either the assemblages or environments at each site, it is hard to even start thinking about some of these complexities. There is a lot to uncover here that may still require some further analyses.

Reply: As noted above, each ojo is slightly different than the other in terms of water chemistry and discharge rates but we do not have replicates of identical ojos so we cannot deduce statistical differences and attribute them to such conditions. As we note in our response to reviewer #1 and in the paper, this is the nature of doing field work there are confounding variables which complicate interpretation, but the results you get from such studies are more realistic. We note that despite the differences between ojos there are common trends and we think it is more useful to focus on these observations than to over analyze differences which would be speculative at best to explain. Complexities are the nature of such studies yet we can still glean useful information.

Minor points:

Comment 4. Why the use of the >255 size fraction? Could this have biased the results especially is different size species respond differently? For example the Henehan et al., 2017 paper on weight suggests size may impact species calcification response. Is it possible that smaller species may have differing metabolic requirements?

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Reply: Indeed size is important and can impact metabolism and calcification response however, tropical benthic foraminifera are characterized by large sizes (Why are larger forams large? Hallock, 1985, Paleobiology) and the size fraction >250 um represents the adult individuals likely to be preserved in the sediment (Martin, 1986) since juvenile mortality rates are higher than 95% (Hallock, 1985). We have inserted this information in the methods section. Specifically, in our samples in the > 250 um size fraction we around 300-500 specimens per gram sediment while only 9-27 specimens were found on the fraction of 125-250um hence the smaller size fraction included at most 10% of the foraminifera. Regardless although the larger size fraction represents ∼90% of the forams we specifically refer to Large Foraminifera in the title to be honest to our data. Nonetheless, we have included in the discussion a section on the role of a larger size on increased symbiont concentration and dissolution resistance in sediments (Hönisch et al, 2004), which may be responsible for the changes in abundance we see.

Comment 5. What was the depth of each site? I almost wonder if this could be contributing to some of the inter-site differences?

Reply: The depth ranged from ∼5 to 7 m at all sites. The depth of each site has been included in the water chemistry table. The setting is quite similar, and all sites have similar light conditions.

Comment 6. Can you report all species identified (in addition to the most abundant)? It would be very valuable for assessing assemblages at a finer scale and also for future workers. Ideally, it would be good to see full assemblages reported at each site and represented and compared in a figure.

Reply: We did exploratory analyses in 10% of the samples to determine what the most abundant genera were, and we have now mentioned other genera present in the samples in the results section (Borelis, Clavulina, Elphidium, Spiroloculina, Peneroplis, Laevipeneroplis, Planorbulina, Sorites, Vertebralina and Heterostegina). However, we did not analyze the full assemblage of all the species present in all the samples. There

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is already existing literature on full assemblages in the area which found similar results (Wantland, 1967; Triffleman et al, 1991; Gischler et al., 2008).

Comment 7. How large were the sediment samples from which forams were measured? Did it differ between sites? And how were they collected? Importantly, how deep into the sediment were samples taken and was consistency in this regard maintained across sites?

Reply: The samples were collected from the upper centimeter of sediment with a spoon and a centrifuge tube across all sampled sites. We analyzed at least 1 gram of sediment per replicate, with an average of ~2 grams of sediment per replicate. We aimed for at least 300 individuals per sample; however, due to the low abundances in some of the samples (especially in samples collected at springs), this was not always possible and 24 of the 50 samples had less than 300 individuals per gram. We have included this information in the methods section.

Comment 8. Page 2, Line 12-3: It is also worth noting that some species also tolerate (even specialize in) high CO2 environments such as oxygen minimum zones – look into some of the Bernhard papers on this in Santa Barbara Basin– and low salinity (low saturation state) estuaries.

Reply: We note in the introduction that foraminifera are versatile and that some tolerate high CO2 settings and low salinity. Note however that most OMZs and also Santa Barbara basin are above the CCD and are super saturated with respect to calcite. Most studies of benthic forams in OMZs and also in SBB focus on the low oxygen rather than high CO2.

Comment 9. Page 3, Line 12: What is your balance error?

Reply: The analytical micro-balance has an error of ± 5 ïĄ■g. This information has been inserted in the methods section.

Comment 10. Sections 3.3. and 3.4 raise a lot of questions for the reader about what

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is producing the reported differences between sites. See major point 3, but I think the conclusions could be made sounder if some of these type of questions are tackled.

Reply: See reply to major point 3. We agree that it would be nice to address this but we do not think it is possible since the slight differences in other water chemistry parameters are not consistent between ojos so no statistical power to decipher the causes for the difference in magnitude of the responses.

Comment 11. Section 3.5: Again, it looks as if there may be a difference between sites. Is this statistically significant? Also, this should refer to Figure 4.

Reply: Thanks for finding the mistake, we have added new plots and changed the number of the figures.

Comment 12. Page 5L Line 1: "but not Gorgos" - Page 6, Lines 23-34: "Therefore, while abundant CT may help lower the potential impact on foraminiferal calcification at low pH, it does not seem to fully counteract the effect of low ." I don't think this is quite right. If I understand correctly, the authors are arguing that this important parameter is carbon- ate ion concentration/saturation state, and total inorganic carbon and pH are important drivers of this both intra- and extra-cellularly. If so, this should read along the lines of "Therefore, while abundant CT may increase the availability of carbonate ion, it does not seem to fully counteract the effect of low pH on ."

Reply: We removed this paragraph from the paper however elsewhere in the paper when we comment about the CT we changed the sentence as suggested.

Comment 13. Page 7, lines 1-3: This really glosses over the huge history and literature on shell weight and carbonate chemistry in planktonic foraminifera. Have a look at Table 7 in Weinkauf et al., 2016 for a good (though not exhaustive) review of some of this work.

Reply: Thanks for the reference to this interesting paper, we have rewritten the test weight discussion to be more succinct and straightforward.

Comment 14. Page 7, lines 8-10: Davis et al., 2017 also shows variable individual responses to saturation state within a population of foraminifera.

Reply: Thank you for this relevant reference, we have inserted this reference in the discussion of test weights.

Comment 15- Many of the figures appear low quality and pixelated. This may be the result of embedding, but double check. Also, why not include color as well as gray-scale for this online publication?

Reply: We have changed the figures to color bar plots and increased image quality to improve readability.

Comment 16. For figures 2-4, it would be useful to have represented on these plots which groups are significantly different from one another (as in the Results section). Consider including this?

Reply: We have added asterisk to significant differences ($p < 0.05$) between paired control and spring sites.

Thank you for the time you devoted to improve the paper

Adina

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