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Chemical characterization of Punta de Fuencaliente CO_2 seeps system (La Palma Island, NE Atlantic Ocean): a new natural laboratory for ocean acidification studies / Sara González-Delgado et al.

Responses to Peter Landschützer' revision:

Comments to the Author: *"Dear authors, I have now studied your revised manuscript with track changes and went back to the referee comments and your response as well as my own recommendation. To recap: The majority of referees considered the natural analogy comparison problematic and urged the authors to substantially revise the manuscript in this respect. Likewise, a second major concern was raised related the novelty of the study as some results on the sites are already published. In response to these referee concerns I further explicitly asked in my decision letter for you to substantially revise (i.e. major revisions) the manuscript to clarify these points. From my reading of your response to the referee comments and the final revised paper (following my editorial comment) I dont believe you have taken the comment into serious considerations. This is also fairly easy visible by the minor changes in the track-changed version of the manuscript. This has further been recognised by the referee comments I received from referee#2 who has reviewed the revised manuscript. Unfortunately, there were some technical issues, hence referee#2 has sent the comments to me electronically and only used the online system for the recommendation. Please find the comments at the end of this letter. I dont think it was necessary to change your position regarding the natural analogy assumption, however, some concerns from round 1 have not been (fully) addressed. While I read e.g. that the authors added some points on the limitations (lines 252-262), I dont believe this qualifies as the discussion the referees were looking for. E.g. in his referee letter, referee#3 is concerned about the "extreme variability" at these sites and how this effects the natural analogy assumption (in particular, the referee called the the usefulness as natural laboratory limited). In the response and the revised manuscript, however, I barely see this addressed (with the exception of the link to the tidal influence). I was also hoping that the authors would use this occasions to not simply cite the studies that have previously explored these and other sites, but also clearly discuss the new findings in comparison to previous studies (also at different sites). The referees clearly seem to be concerned about the natural analogy assumption and they refer back to previous literature and studies that do show that such an analogy can be problematic (see also the comments from referee#2 on the revised manuscript below).* ***Again, I dont expect the authors to change their position that the site is a useful natural laboratory, but I expect the authors, in response to the referee concerns, to provide clear and solid evidence and a substantial discussion also in comparison to previous studies****. Based on my reading and the referee comments I decided that my position has not changed since round 1, i.e. the authors need to conduct major corrections to the manuscript. All referees welcome the description of the site and the system (although I would welcome not only citing previous studies, but also highlighting how the new data advance the knowledge in comparison to these previous studies). In particular - and* ***I cant stress this enough - I want the authors to further address the referee concerns about the natural analogies, e.g. the variability or "noise" as mentioned by the referees*** *(referee#3 in the first round and referee#2 below) and have a fair and substantial discussion on the limitations also in light of other studies conducted (also at different sites).*

Response: First of all, we apologise for not fulfil yours's and reviewer 2 previous requirements. We have now further addressed the two previous big controversial points: (1) the usefulness of this kind of acidified systems as natural analogies of future oceans, and (2) the originality of our study. Following also your advice, our position is clear, this kind of systems, although not perfect, might be used with caution as natural analogies of future oceans.

About the first point, we have extensible addressed this question and provide solid and detailed evidence in comparison to previous studies. We have made changes on the text and added a discussion about the variability found at these natural systems, their limitations and usefulness, comparing our findings with others previous studies results. In this version more information is included in this sense.

In lines 260 – 277, we added examples of other acidified systems "These variations are very similar to other acidified natural systems. For example, the CO_2 vent of Ischia (Italy) has pH levels from 6.07 to 8.17, Ωaragonite from 0.07 to 4.28 and Ωcalcite from 0.11 to 6.40 (Hall-Spencer et al., 2008). The one from the island of Vulcano (Italy) has pH values between 6.80 and 8.20, Ωaragonite from 1.49 to 4.65, and Ωcalcite from 2.28 to 7.00 (Boatta et al., 2013). Meanwhile the CO_2 seeps from Papua New Guinea have pH levels between 7.29 - 7.98, Ωaragonite 1.2 - 3.4 and Ωcalcite between 1.36 - 5.12 (Fabricius et al., 2011). That from Shikine island (Japan) have pH values between 6.80 and 8.10, Ωaragonite from 0.20 to 2.22, and Ωcalcite from 0.30 to 3.45 (Agostini et al., 2015)".

And we also emphasized that these systems are not entirely perfect to predict the ocean future. We added "Although these systems are far from being perfect predictors of the ocean future due to their chemical variability and physical limitations, they have proven to be important tools for the study of ocean acidification (Foo et al., 2018; González-Delgado and Hernández, 2018; Aiuppa et al., 2020). These natural acidified systems, such as Punta de Fuencaliente system (PFS), can be used as natural analogue of climate change scenarios predicted by the IPCC (2014) (Fig. 6)."

In lines 282 – 292, we added some specificities of our system that make it different and we highlight that several caveats for future prediction experiments should be considered. "Moreover, the acidified system of La Palma highlight by the absence of bubbling, since the volcanic degasification takes place in the aquifers and not directly on the coast as in other acidified systems of volcanic origin (e.g. Hall-Spencer et al., 2008; Fabricius et al., 2011) (Fig. 5). This could give us new insights of the effect of

acidification *in situ* avoiding the effects of bubbling (González-Delgado and Hernández, 2018). Nevertheless, several caveats for future prediction experiments should be considered, as well as in other natural acidified systems, especially those related with increased alkalinity values in the submarine discharge. First, there is a clear tidal influence, this is an important force that controls the acidified brackish water discharges. Although a fluctuation of the emission is observed, normal ocean conditions can occur for a short time, about 2-4 hours per day, during high tide, and depending on the oceanic conditions (Viotti et al., 2019). The $pH_{T,is}$ is severely affected by the location, reaching down to ~7.2 in the emissions points, so a careful selection of the study sites is recommended, depending on the study objectives (Fig. 6). This tidal phenomenon has also been reported in other acidified natural systems such as Puerto Morelos in Mexico (Crook et al., 2012) and Ischia (Kerrison et al., 2011). However, the pH time fluctuation can be used to our advantage, as a daily and seasonal fluctuation of the pH is normal in coastal habitats environment (Hofmann et al., 2011). So, it could be considered very useful to incorporate pH variability in ocean acidification studies as environmental fluctuations that can have a large impact on marine organisms (Hofmann et al., 2011)."

In lines 302 – 304, we added the other studies should also be considered. "Therefore, measurements of heavy metals and other elements in seawater should be considered in following studies."

In relation to this first controversial point, we would like to clarify that the reviewer 2 mention previous literature that shows the cavities of using natural CO_2 labs against our position, but we do also recognize these cavities in our manuscript. In lines 278 – 315, in addition to what is indicated above regarding this point, we have also added "The high concentration of bicarbonate in the brackish waters also implies an extra contribution of alkalinity and carbonate that can buffer the effect of acidification in the area, so it is necessary to take this into account when making predictions of the future. These values together with calcium content are especially important factors in the case of the saturation state for both calcite and aragonite, that shows high values for seawater with low pH values. Therefore, despite the fact that we are dealing with a subtropical ecosystem, the values obtained in both saturation states are more similar to the predictions for a tropical ecosystem, such as the values found in Papua New Guinea seeps (Fabricius et al., 2011; IPCC, 2014).

Finally, the area is not very large and only one type of rocky benthic habitat, the most typical community of Canary island, is present at the PFS (Sangil et al., 2018).

Therefore, all conclusions derived from this acidified system should be interpreted with caution and has local effects. Hence, it is crucial to establish a collaborative network of researchers who are working in other natural acidified systems worldwide to have a more realistic interpretation of future ocean scenarios."

However, her/his point for all these kinds of studies, done in natural acidified areas, is that "*because the environmental conditions are not what we expect for the future and responses only confusing*", we disagree with her/his interpretation.

In this sense, it could also be said that the studies carried out in the laboratory are not real either. In these experiments, generally considering only one biological species, one or two variables such as pH and T are controlled and the joint effects of the multiple stressors that affect biological communities are not taken into account. However, they are essential to be able to interpret more complex systems.

We believe that these studies are necessary to understand the ecosystem scale of the ocean acidification problem we are facing. Therefore, we expect that with this new version of the manuscript, we have better explained our argument and the importance of this place. We now support our argument, as you recommend, with a stronger discussion using previous literature.

About point 2, we have now included more information about our previous studies to have a better context of the present study and to highlight its novelty.

In lines 216 – 223, we added "Although CO_2 emissions on Fuencaliente coast had already been detected (e.g. Hernández et al., 2016; Viotti et al., 2019), this is the first time that this naturally acidified system has been described chemically and physically. Previous works have focused on punctual questions; Hernández and collaborators (2016) published for the first time the presence of CO_2 SGD in Fuencaliente, specifically in Las Cabras beach. Later, in the thesis by Pérez (2017), as well as in the conference papers by González-Delgado et al (2018a,b) and in the article by Viotti et al. (2019), new points of acidification were discovered on Playa del Faro and Los Porretos. However, in none of them was a chemical characterization of the whole area made as here." In this study, three of the four variables that define the CO_2 system were measurements (pH, total alkalinity and total dissolved inorganic carbon) and CO_2 certified reference material was used to collaborated our carbon dioxide system measurements.

However, we must say that our study is original, and the data have not been previously published, no doubts about that.

Responses to referee#2 revision:

Comment 1:"*The Authors corrected most of the mistakes made in the original version and added some details which were lost.* ***This help to better understand the work done. However, the authors did not consider reviewing their position regarding the ability of this new system to become a laboratory for understanding the effect of climate change on organisms.*** *Personally,* ***I find this position unusual****. I perfectly know that some of the so called natural analogue found so far are far to be perfect, and that is the problem. Most of conclusions from some of the studies published,* ***although always important to advance in science, make actually only noise****.. ,because the environmental conditions are not what we expect for the future and responses only confusing*."

Response: We presented an improved version of the manuscript thanks to yours and editor/reviewer´s comments.

We understand that the increase in alkalinity makes the conclusion on this area different to what is expected in the future. However, not only in this region but also in upwelled corrosive environments such as California upwelling region (Feely et al., 2008; 2016), increase in acidity, inorganic carbon and normalized alkalinity to a constant salinity have been shown. Even with this difference, we consider we could clearly maintain our position about this kind of area confirming the differences, and although not a perfect system, it can be used, with this in mind, as natural analogues to future oceans but also to previous times. We have also, already proven the usefulness of this natural systems to demonstrate that while acidification may affect the calcification process of the mollusc *Phorcus*, it's not affecting its survival (Viotti et al., 2019 MERE) or can also indirectly benefice the growth of some calcifying invertebrates (Uthicke et al., 2019). These papers in natural environments, rather than adding noise and confusion, have helped to understand the complexity of ocean acidification phenomena. We believe that these works provide valuable information that complements the lab experiments and model predictions. Indirect ecosystem effects can only arise in these natural environments. We have added more information about this issue at the discussion section in lines 261 – 316 as indicated above in the comments to the editor. We have to recognize the limitations of using these spots and with doing so we are informing others on how to use the information gather at this spot. This is also an important point that makes this manuscript interesting for other OA researchers.

Comment 2: "*Looking to the description of your system I see: - the lack of CO2 seeps. So the ms must be around submarine groundwater influence and the word CO2*

seeps should not be mentioned because they do not exist, or this study do not show if they are in the near- or offshore water."

Response: We know that CO_2 emissions are underground affecting the groundwater that is discharged into the coast. Although we do not directly analyse CO_2 emissions, we comment additional data from a previously work written by Soler in 2007. In this study, Soler talks about the presence of CO2 seeps due to the remnant volcanic activity in the area. The presence of CO_2 emanations in land, have also been previously found in the area (Padrón et al., 2015). In any case, we have now used the term submarine groundwater discharge (SGD) along the text.

We have better explained this issue in the manuscript for better understanding. In lines 221 – 227, we have added "Although CO_2 emissions on Fuencaliente coast had already been detected (e.g. Hernández et al., 2016; Viotti et al., 2019), this is the first time that this naturally acidified system has been described chemically and physically. Previous works have focused on punctual questions; Hernández and collaborators (2016) published for the first time the presence of CO_2 SGD in Fuencaliente, specifically in Las Cabras beach. Later, in the thesis by Pérez (2017), as well as in the conference papers by González-Delgado et al (2018a,b) and in the article by Viotti et al. (2019), new points of acidification were discovered on Playa del Faro and Los Porretos. However, in none of them was a chemical characterization of the whole area made as here. Our results reveal the continuous influence of brackish water discharge in the acidification process of Punta de Fuencaliente System (PFS), which had been missed before (Fig. 5). Similar to aerial remnant volcanic activity in La Palma that generates high CO_2-diffusiveatmospheric concentration (Padrón et al., 2015), submarine remnant volcanic activity causes the acidification process found here, as indicated by the chemical composition of the groundwater analysed, which is less than 200 m from the coast (Soler, 2007). The activity of this SGD is comparable with other CO_2 vent and seep systems worldwide (references within González-Delgado and Hernández, 2018)."

Comment 3: *"- the quality of the groundwater discharge makes conditions too variable in pH (yes as most of the natural analogues), too altered (in bicarbonate, At, calcium, Mg, likely other metals etc), too limited in space (meters from the beach with a tide of 2 m!!). Ok, no problem,* ***nice and useful*** *but I* ***continue to think that it is not an exceptional spot to study OA****, not great to study* ***how life has persisted through past eras (what a pretention!)****. It could help to understand the early life on Earth from the Precambrian..?????? Could allow to disentangled adaptation and evolution…??? Please, do not speculate."*

Response: We believe that this kind of comment is inappropriate and close to be consider an offence to our ideas and work. We agree with reviewer that we cannot study in this environment what has happened in the Precambrian or even in past eras, but the pH conditions you observe in the PFS are those indicated for those periods. However, this kind of extreme environments are clearly useful to study the type of organism that life there and their biology. In this sense, we can get closer to understand how marine organism may have lived in pass eras with "similar" conditions. It was not the aim of the present paper but the expected (future work to be monitored) high frequency change in acidification with tides makes this system appropriate for long time process studies, resilience, adaptation and evolution of species under the stress conditions imposed by both lunar cycles effects on tides and changing groundwater CO_2 content. The ocean acidification process is not a constant process and seasonal, interannual and decadal trends have been observed in the long ocean time series such as BATS (Bates and Johnson, 2020). The area, presented hourly, daily, monthly and longer time variability affecting the live in the area or any organism we can set in the area to study its evolution. We consider, as it has been assumed by many previous workers in many other CO_2 natural emitting sites included in the introduction section, that these places are very useful. We assume that using this type of systems to interpret, or at least to have an approximation, even if it is not perfect, can provide insight about past and future situations.

Of course, there are limitations, but there are limitations and assumptions in every experiment and model we design, in every decision we made to choose our hypothesis, this is the science we use today. You also mentioned adaptation and evolution. In this kind of natural experiment, we have found both adaptation and rapid evolution of seagrasses, algae and invertebrates (Kumar et al 2017; Olivé et al 2017; Harvey et al 2015), and this is interesting to disentangle a bit how evolution works. Why these authors have used this kind of experiments if their area do not perfectly match the projected future ocean conditions? Why we should *a priori* limit or prejudge the use of this areas? We do believe these are exceptional spots to study adaptation, evolution and how life deal with this unusual ocean conditions.

To better explain how important these areas for this king of adaptation/evolution studies are we have included below some concepts and the scientific context that, maybe, makes you see things from a different perspective. Species show phenotypic plasticity that may allow an organism to maximize fitness (Gotthard & Nylin, 1995) and adaptive phenotypic plasticity is expected to evolve in populations subject to contrasting

environmental conditions (Via et al 1995). Intraspecific variability of this capacity could be then investigated to understand population resilience and adaptability on the time scale of actual scenarios, as the pH gradient presented in this manuscript. Some studies have already demonstrated variation in the degree of plasticity among populations in response to degree of variation in the environment (Kaitala et al., 1991; Leips & Travis, 1994; Morey & Renick, 2004). However, few studies have explored whether historical changes in environments or these kinds of gradients are associated with the evolution of phenotypic plasticity, local adaptations and time needed to evolve (Morey & Reznick 2004); and just one study has explored the advantages of these local adaptation modes to counteract ocean acidification (Kelly et al., 2013). Therefore, we do not speculate, we believe and have shown that these areas are exceptional spots to study adaptation and evolution.

Following editor recommendations, we have added more information and discussion to better explain the utility of these spots. For instances, we have compared our spot with the one in Ischia, Italy (Hall-Spencer et al., 2008), and the one in Papua New Guinea (Fabricius et al., 2011). We have added this discussion in lines 260 – 276. In general, even with noise (variability) associated, it has already been proven that the results obtained in these natural areas are important to better understand the effect of ocean acidification on an ecosystem level or to test adaptation and evolution.

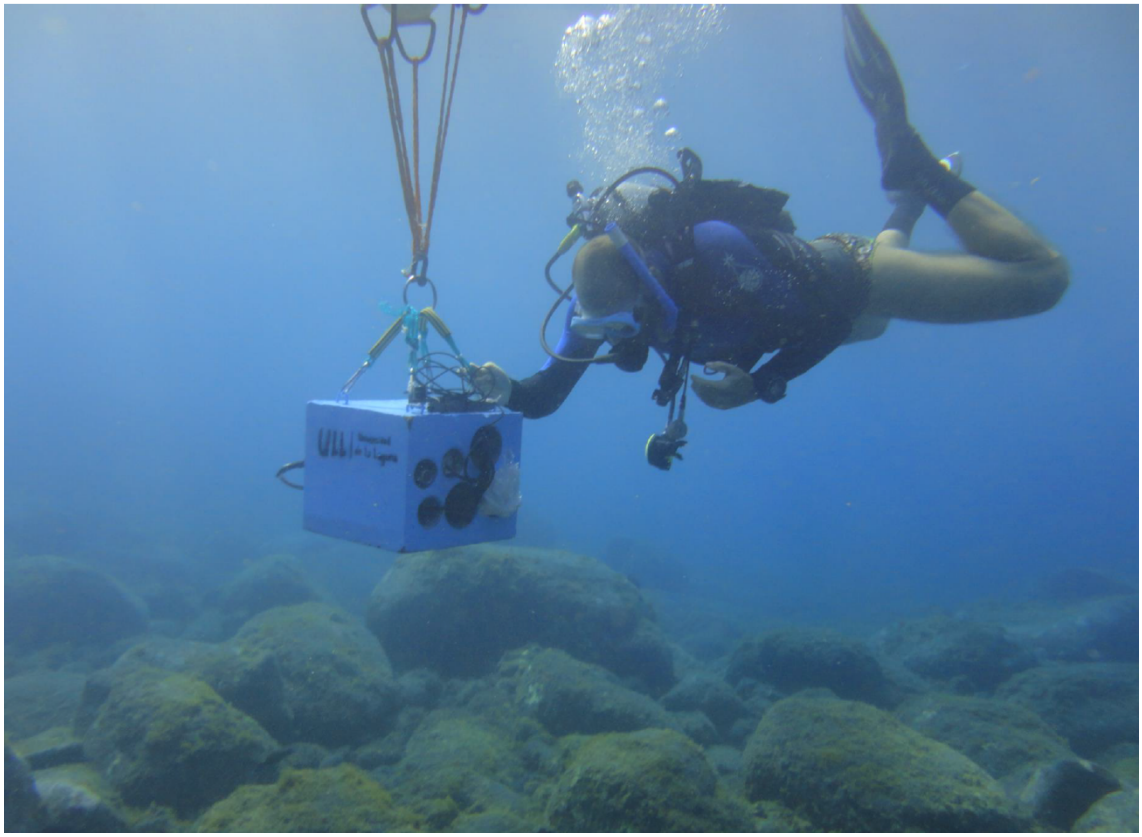
Comment 4: - *"the accurate chemical characterization" is a two date sampling on the seawater carbonate chemistry only. No mention about other true chemicals and metals discharged. IPCC does not include pollutants in its scenarios."*

Response: Our data was obtained during three different months over two years, and we believe that this set of data can clearly be used to establish a chemical characterization of the carbonate system for the area. Of course, you can always have more data to characterize something, however, the limited funds we had did not allowed us to reach this point. It is true that it would have been great to provide, along with the water carbon system, data on metals and other elements but this has not been possible for the moment. We have added a clarification regarding this in lines 297 – 298.

Comment 5: *"- the sampling was made, I insist, in the intertidal zone (i.e., The intertidal zone is the area where the ocean meets the land between high and low tides) when considering a tide of 2 m as at the study site. The effect of discharged water, so the low pH area, is close to the beach and immediately dissipate 50 m far, even less*

depending the study site, from the shore. The lat & long scale does not help to appreciate the real extension of the affected area but having a look in google earth it is clear that for instance Playa del Faro (the biggest area of this study) is a 80m long."

Response: The sampling was made underwater using bootless in the subtidal area. Our study site is always cover by water, even in low tide. We also provide a picture, see bellow, to demonstrate to you that we were working underwater.



We believe that the scale is appreciated when you compare the graphs with Figure 1c, hence we have added the star-shaped mark that relates the interpolation graphs to the 1c map. The area is not very large, as it was already mentioned in the previous version of the manuscript (lines 313 – 315), this is one of the caveats of the spot, because we can only study the effect of the acidification on the local communities presents in the area. We have also wished to have a larger acidified areas with a proper temperature gradient to perfectly much the IPCC projections, but nature is capricious. We also know that many scientists would like to have a lab where species could be set and be studied in a tank of the dimension of this beach with gradients in pH. This is what this natural place offers.

Comment 6: *"- This ms is only about the seawater conditions found. There is no data about what could be the biota living and affected by the gradient in the beach, which is an area affected by the tide and the local strong waves. It is not enough to cite an in press paper (not cited in the literature list). All discussions on that part are purely speculations."*

Response: Yes, you are right the manuscript is about the chemical characterization of an especial spot in La Palma island but not about the present biota. However, we have already published a paper about the biota and we are now working with new collected data that we would like to comment. We are sharing our observations and we have just included a comment for the readers to better visualize the study area. We would like to keep this small, "naturalistic", comment.

Comment 7*:"- Fig. 5 is not really important and good for a book. How brackish groundwater is released and reach the sea is an already known mechanism. - Fig. 6 is* ***honestly inacceptable****. - Fig. B1. I do not understand what exactly means.*

Response: We believe that Figure 5 is adequate to understand the phenomenon that occurs in the coast of Fuencaliente, not only to show "*How brackish groundwater is released and reach the sea"* but also, how CO_2 emissions mixed with this brackish groundwater. Figure 6 shows the pH levels found in the spot area and clearly shows what you were asking in your comment 5, and can be related with those predicted pH values for the future by the IPCC. We would like to keep figure 6. Figure 1B is a complement to the description of the study area, this figure not only summarizes the locations, months and parameters sampled at each site, but also shows the location of the lava flows from the last eruption that took place in the area.

Comment 8*: "Why min pH was used? Why the data are connected by a line although sampling is not continuous? Clearly, tide has a direct effect of the groundwater release"*

Response: We have modified this graph to better represent the data. We agree with you that the tide has a direct effect on the release of groundwater. This is one of the findings of this study. The effect of the tide should not be neglected in any hydrothermal system near the coast, as demonstrated in Santana-Casiano et al., 2016. In this work (figure 3 and 4) it is demonstrated how the emissions from the Tagoro volcano, its crater at 88 m depth, are affected by the tides.

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