Interactive comment on "The regulation of coralline algal physiology, an *in-situ* study of *Corallina officinalis* (Corallinales, Rhodophyta)" by Christopher James Williamson et al.

Christopher James Williamson et al.

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We thank the reviewer for their thorough review, and for their comments that this is "a great study into the geniculate coralline alga Corallina officinalis", and that the study "was well written". We apologise for our slow responses, which was due to a long field-work commitment.

To address the reviewers general comments:

General comments:

Comment 1: Premise for the research is that it is important to get a baseline for climate

change impacts. . .but that this argument was poorly made, not discussed adequately or supported by citations. Statements about how climate change would affect the algal physiology were vague and repetitive (e.g. line 89-90, line 52, line 369-371) and climate change wasn't explicitly tested in the study design. Statements that this alga is facing any immediate threat from climate change weren't supported and I remained unsure what the implications of the findings were in the context of climate change in the discussion (e.g. line 549). This is a great standalone ecophysiology paper and interesting within itself. I would suggest either elaborating on statements about climate change. . ..so there could be a strong case made for extreme conditions experienced during rock pool emersion being equivalent to future conditions. . ...or else removing a lot of this and making a simple case for the research in the importance of understanding physiology, which is perfectly fine.

Response 1: We agree with the reviewer that this area of the manuscript was not sufficiently explained and have therefore added more cited information into the introduction as to the potential impacts of OA and increasing temperatures on macroalgal physiology, with specific citations of studies addressing these impacts for our study organism, C. officinalis (see lines 90-117). Our approach from the outset was that, in agreement with the reviewer, whilst this study does not explicitly test climate change responses, it is obviously motivated by the requirement to understand interactions between calcified algae physiology and environmental variability in the wider context of global change. Thus we feel it is important to maintain our approach of contextualising our study and findings in the light of climate change/OA, but to not attempt to portray this work as a direct investigation into such dynamics. We therefore opted, for example, to include our conclusions on the direct consequences of our findings for climate change/OA responses only in our conclusions section (Line 544 onwards) as opposed to overly focussing on this aspect of our work in our main discussion section. Whilst we have therefore added more information on this aspect into the introduction at the reviewers request, we do not feel that this should be attempted in the discussion.

Comment 2: Through-out the manuscript environmental variables were referred to as 'stressors' – is this the right terminology? Maybe it is correct but I found this a confusing as I would expect irradiance is an environmental condition rather than a stressor: it elicits a physiological response but is necessary for life and in this context was being tested under realistic environmental conditions.

Response 2: We understand the reviewers take on this terminology but are happy that this is a commonly used way to refer to environmental variables in such studies. Additionally, whilst irradiance (and indeed temperature and many other variables) is necessary for life, it is also indeed a stressors – C. officinalis' photosynthesis is light saturated during the complete annual cycle (this study, and Williamson et al. 2014, Marine Biology 161), causing significant photo-stress that must be managed in order to maintain growth and survival.

Comment 3: Methods need to be explained – I was unclear whether these measurements were occurring in one pool, or ten individual rock pools – or if the same pool was being returned to each season? This is important as volume, shape and location of pool could influence how quickly it heated up. Did the emersion duration change in different seasons? i.e. variability in the time period between beginning (in first hour) and end (last 1.5 hr) samples. Wouldn't variability in the duration of exposure have influenced findings? This wasn't clear.

Response 3: We have added further information into the methods section at lines 155-160 to provide clarity on the experimental design. Samples were collected randomly from several upper shore rock pools, though incubated together in one single upper shore rock pool to maintain consistent temperature/irradiance regimes during incubations. Whilst the reviewer is correct that volume, shape and location of pool should typically influence the progression of environmental variability between pools, the pools sampled during the present study are all of a very similar size and indeed shape, having been created by a man-made walkway (Lines 118/119). We have a long history of research in this site, including an initial assessment of changes in the carbonate chem-

istry and water temperatures apparent in these same rock pools during summer and winter daytime emersion (Williamson et al. 2014, Marine Biology, 161). In our previous study, we assessed these changes in both upper and lower shore rock pools at the current study site (and one other site), and found no significant difference between the upper shore rock pools at Combe Martyn. This information has been added into the methods to aid in clarity (Lines 158-160).

There is unavoidable variability in the timings of tides and the durations of tidal emersion periods across the sampling performed during this study that may have influenced the progression of rock pool conditions over emersion periods and thus algal physiology. We have tried to represent this as best possible by reporting the differences in the timings and heights of high and low tide in Table 1. Seasonal incubations were timed as much as possible to occur on similar tides, with similar timings of tidal emersion (e.g. daytime low tide occurred +/- 2 hours across all sampling dates, with a 1.2 m range variance, Table 1), to allow inter-seasonal comparisons - and we are confident in the findings we report here (note consistency with several other studies). To aid in the contextualisation of our incubations, we have added information on the rock pool shore height (Already included in the methods) into the legend of Table 1, though feel that demonstrating the full tidal information remains more informative. This variability does not, however, impact our statistical modelling of algal physiological processes (photosynthesis, respiration and calcification) versus environmental stressors (irradiance, temperature and carbonate chemistry), as data from all incubations were compared simultaneously using a regression approach.

Comment 4: I wasn't sure about how PC1 values were used to represent carbonate chemistry, although appreciate this is a great way to roll all the variables into one value – it might have been more meaningful (and easier for readers to follow) to choose one representative element and use this to describe temporal trends – especially if the data are going to be used in future to parameterise future climate change response models (As suggested). E.g. Fig 4 boxplots are difficult to interpret in a meaningful way.

Response 4: By applying PCA to the carbonate chemistry variables (many of which co-vary), we are able to summarize all variables into a single (here accounting for ca 83% of all variability in the carbonate chemistry data) that can be meaningfully used in regression analyses. Any attempt to run such models with multiple parameters of the carbonate chemistry data would be meaningless due to high co-variance. Additionally, selection of just one of these 8 parameters as representative of the whole dataset would not be sufficient for our aims, and indeed would reduce statistical power. Given the strong focus of this paper on the relationships between physiology and environmental variables (i.e. carbonate chemistry as summarised by PCA), we felt it more appropriate to include the PCA data in the main manuscript and provide the average carbonate chemistry data in the supplementary material. Given the inclusion of the PCA biplot and multiple descriptions of what changes in carbonate chemistry are represented by changes in PC1 (Lines 298-301, 313-315, 317-319), we feel that interpretation of this data presentation is not overly taxing for the reader and have thus not modified this part of the manuscript.

Comment 5: Discussion: These experiments were all carried out at one NW facing sheltered bay – maybe a little discussion about how representative the environmental conditions here might be across the entire species range, to put findings in context?

Response 5: Corallina officinalis is distributed from Iceland to northern Spain in the NE-Atlantic (this information has been added into the Methods, Line 139 – 141). Across this range there is a huge span in environmental conditions and we feel it is out of the scope of this study to approximate how representative the conditions in our site may be of the entire species' range. Our insitu monitoring twinned with our statistical comparisons (i.e. multiple regression approach) allows us to document the in-situ dynamics for C. officinalis in our study site, but also to ascertain information on the relative changes in physiology for given changes in environmental parameters. We are unable to extend such observations beyond the range of our data however – though this is currently the focus of our ongoing projects.

Specific comments Comment 6: Line 121 – 122: specific why these sampling times were chosen- presumably to capture climactic seasons? Response 6: Sampling dates were selected to capture the tidal, diurnal and seasonal variability in both physiology and environmental stressors – this has been added at Line 147.

Comment 7: Line 124 – how many hours before low tide was the emersion period? How long was the emersion period? Table 1 is misleading as it describes tide times, not sampling times – maybe add sampling times to it? (see general comment) Response 7: Please see response 3 above – we believe the information on tidal times to be more informative though have added more information into the Table legend.

Comment 8: Line 133 – Did you return to the same rock pool each season? How many incubations in each pool? Response: Yes, please see above Response 3, information added into the text at lines 155-16, and information already in the text Line 235 – end of paragraph.

Comment 9: Line 137 – why only 5 water samples? Why not one from each of the ten rock pools? Response 9: Apologies, you are correct, this was a mistake (note correct sample size in supplementary material carbonate chemistry plot legends). Sample size is actually $n = 12$ (5 light, 5 dark and 2 control treatments per incubation). This has been corrected in the text.

Comment 10: Line 259 – why did you present a bar chart of daytime temperatures but not night time temperatures? Response 10: This was purely a manuscript length consideration. Given that both PAR and temperature data are available for day time emersion periods, we decided to include this as a figure, though elected to describe night-time temperature in the text to keep figures to a minimum.

Comment 11: Line 267 – why were temperature and irradiance data presented in the main manuscript (Fig 1) but carbonate chemistry in the Supp. Mat? Response 11: Please see above response 4. We elected to include PCA figures as opposed to average carbonate chemistry figures given the strong use of PCA data in regression

analyses. We felt that adding in carbonate chemistry in addition is unnecessary and would make the manuscript unduly lengthy.

Comment 12: Line 278-282 – explain what 'higher' PC1 means in terms of carbonate chemistry parameters. Response 12: please note that this clarification is already present in the text in the proceeding sentence (now Line 314 onwards).

Comment 13: Line 386 – couldn't the other predictors be removed and the between temperature and productivity tested? Otherwise find this interpretation hard to swallow as it goes against your findings. Response 13: We agree with the reviewers comment (as also raised by the other reviewer) and have adjusted the text accordingly. We have still included the information on co-variance in the results section for data clarity. Whilst individual comparisons can be made, co-variance precludes identification of the driver of change.

Comment 14: Fig 4b – what happened to December? Consider revising Fig 4 and focussing on one representative carbonate chemistry parameter to describe temporal change in dynamics, rather than PC1 scores. Response 14: Please see above response 4. Unfortunately December night-time data is not available due to logistical constraints, this is already included in the manuscript methods Lines 145-146 and discussion Line 468.

Technical comments Comment 15: Line 171-173 – I understand what is meant here but had to re-read the sentence several times! Consider revising. Response 15: We feel that this sentence is clear.

Comment 16: Line 110 – remove the word 'significantly'. Response 16: Amended.

Comment 17: Line 89 – the interactions. . . are (not is) Response 17: Many thanks, amended.

Please also note the supplement to this comment:

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