RESPONSE TO REVIEWER COMMENTS
Reviewer comments in italics; **author responses to bold**

Reviewer #2:

"The ability of macroalgae to mitigate the negative effects of ocean acidification on four species of North Atlantic bivalve" This paper evaluates the effect of the presence of the macroalga Ulva rigida on the growth of four North Atlantic bivalve species, Mercenaria mercenaria, Crassostrea virginica, Argopecten irradians and Mytilus edulis. The authors have used small and larger sizes of three out of four species, specifically the three obtained from hatcheries. The pCO2 levels the bivalves are exposed to are high, but conceivable for estuarine systems. The authors claim that "saturation states for calcium carbonate (Ω) were significantly higher in the presence of Ulva under both ambient and elevated CO2 delivery rates ($p<0.05$)", and that "alkalinity was increased by the presence of Ulva". This might be statistically significant, but as alkalinity actually decreases (or is similar) in some treatments (small Mercenaria, large Mercenaria control pH, small Crassostrea, large Crassostrea low pH) it would be interesting to see the relationship between these parameters and growth directly and visually.

We agree with the reviewer's assessment. For this revision we will provide regression analyses of saturation states for calcium carbonate with the growth rates of bivalves to demonstrate the specific relationship between these two parameters. These analyses will appear as supplementary tables and will be discussed in the results and discussion. To summarize these new findings, there was a strong positive and significant ($p<0.05$) correlation between shell length-based growth and saturation states of aragonite and calcite in all species and size classes, save for *Mytilus edulis*.

We note that alkalinity is affected by many processes and while nitrate uptake will increase alkalinity, other processes may decrease it and that prior research has definitively demonstrated that saturation states for calcium carbonate are the key factor dictating the effects of acidification on bivalves.

In treatments with Ulva additions, one would expect the variability in pH to be higher due to respiratory activity and production. However, the average pH is higher but the variability in pH seems similar to treatments without Ulva. In fact, I would expect the algal-addition treatments to have a fluctuating pH and the control treatments to be stable, which could arguably have caused the differences. However the authors do not discuss this and the tables do not show these differences in variability of pH. Was the pH fluctuating on a day-night scale in the Ulva treatments? Or was the gas flowrate so high this was not discernable, and what causes the variability in the control treatments?

The reviewer is correct that the treatments with *Ulva* had more variability in pH but did, on average, have higher pH levels. For this revision, we have made plots showing the changes in pH over time for the *Ulva* treatments to demonstrate that there is variability but that the pH rose in these treatments after each water change in every experiment, likely due to the uptake of nitrate and the assimilation of CO_2 by *Ulva*.

The nutrient and algae addition to the vessels might cause different nutrient concentrations in the treatments, with Ulva taking up nutrients while they remain suspended in the control vessels, which could have influenced results.

We agree with the reviewer that additions of nutrients and algae might cause different nutrient concentrations within treatments, and that *Ulva* may alter nutrient concentrations. If we agree that more nutrients could increase algae concentrations, this could result in more growth. However, we note that in treatments without Ulva where there might be more nutrients, bivalve growth rates were lower, not higher as would be predicted. More importantly, for the revision we will provide the newly obtained data on phytoplankton cell counts which were found to be highly similar and not statistically different across all experimental treatments in each experiment. Finally, but importantly, we note that any differences in nutrients among the vessels would occur in an ecosystem setting as well with more nitrate assimilation and removal and thus an increase in alkalinity in times and places where there is more *Ulva*. Hence, any differences on this front would be realistic in an ecosystem setting.

It is unclear what time of the year the experiments have been done (presumable summer due to hatchery times), and how the results might vary in other seasons (i.e. when Ulva is not productive).

The reviewer's presumption is correct as the experiments occurred throughout summer 2017, which is the peak growing season for bivalves and *Ulva*. We targeted this season specifically for that reason, although it should be noted that within the collection site of *Ulva*, the macroalgae appear during the early days of spring, and persists into the end of the fall months, which is beyond the time that our experiments were concluded. During period when *Ulva* grows more slowly (spring and fall) it would be expected that its growth would be slower and its ability to mitigate acidification would be lower.

The various sizes and the amount of different species of bivalves used in this study make it an interesting read, even though it is not entirely clear what causes the beneficial effect of the presence of Ulva (its effect on the carbonate chemistry, nutrient concentration or something else).

We agree with the reviewer that the exact cause of the increased bivalve growth in the presence of *Ulva* cannot be exclusively tied to a singular cause. However, we believe the new data generated for our revision make the case for the mitigation of acidification even stronger. We believe the new regression analyses we will include that depicts the significant linear relationships between calcium carbonate saturation states and bivalve growth makes the carbonate chemistry angle more convincing. We believe the day-by-day decreases in pH provided by Ulva during experiment makes the carbonate chemistry angle more convincing. Finally, our inclusion of phytoplankton density data showing there are no differences among treatments indicate this was not a driver of the findings.

Specific comments: Methods P.3, line 9 "light intensity (~200 µmol photons m-2 s-1)", how does this compare to ambient conditions?

Light intensity used in all experiments was set to mimic ambient light intensity where *Ulva* grows in near shore regions. We will add this information to the manuscript to specify this.

P.3, line 23: Isochyrsis should be Isochrysis

We will make the suggested change.

P.4, line 17: "some estuarine environments" – representable for the environments of the study organisms and their origin?

Yes. For example, Wallace et al. (2014) observed pCO_2 concentrations exceeding 2,000 µatm in Jamaica Bay, NY, USA, which hosts the bivalve and macroalgae species used in the present study.

P.4, line 32-33: "Well-pigmented, circular sections of Ulva (~3.5 cm and ~7 cm for experiments in small containers and large vessels, respectively". These small containers where 1L, while the large vessels had a volume of 8L. The biomass of Ulva however, is 2x as large for the larger volume, which does not respect the ratio biomass/water volume. The authors state that the weight was consistent with the benthic coverage in Shinnecock Bay, would that mean that the 8L vessels had 2x the diameter of the small containers and would water volume not be more important than surface in this case? Or was there more than 1 disk per container (p.5., line 23 states "disks")? This section is a bit unclear.

The amount of *Ulva* used was based on tissue weight, not tissue surface area, and the amount of *Ulva* added to 1 L and 8 L containers was consistent with the benthic coverage of *Ulva* in Shinnecock Bay based on several years of benthic trawl data as well as other estuarine regions (Liu et al., 2015; Sfriso et al., 2001). This point is specified in the Methods on P4, L31-34 and P5, L1-5. Considering the 2-dimensional nature in which interactions of the bivalves and the macroalgae would occur, it would make more sense to base the amount of macroalgae used on the surface area of the container, and not necessarily the volume.

P.5, line 16-17: "with discrete and continuous measurements of pH, dissolved oxygen, and temperature", which measurements were discrete and which continuous?

We measured pH and temperature discretely and dissolved oxygen continuously. We will change the text to specify this difference.

Results P.6, lines 19-20: "For the larger-sized cohort of M. mercenaria (5.00 ± 0.41 mm), Ωcalcite and Ωaragonite were significantly higher in treatments containing Ulva and significantly lower in high CO2 treatments" Throughout the manuscript's result section this way of describing the differences between high CO2 / Ulva treatments is confusing. In the highCO2+Ulva treatment the Ωcalcite is actually lower than the control-Ulva treatment (as expected), however from the text it appears at a first glance that all Ulva containing treatments are higher, the sentences might be clarified to prevent confusion.

We intended to specify that $\Omega_{calcite}$ and $\Omega_{aragonite}$, although significantly lower under elevated CO_2 concentrations in general, were significantly higher in the presence of *Ulva* in both ambient and elevated CO_2 treatments. We agree with the reviewer that the sentence structure used throughout the manuscript may cause confusion and will change the text to separate any significant differences in $\Omega_{calcite}$ and $\Omega_{aragonite}$, be it under elevated CO_2 conditions, or in the presence of *Ulva*. We will also include references to the respective figures that show $\Omega_{calcite}$ and $\Omega_{aragonite}$, which would make it clear that $\Omega_{calcite}$ and $\Omega_{aragonite}$ are lower under elevated CO_2, but higher in the presence of *Ulva* in both ambient and elevated CO_2 treatments.

Discussion Could the fact that Mytilus seems less sensitive to addition of Ulva be related to the more "natural" (no hatchery) origin of the juveniles and their exposure to environmental fluctuations vs. the more stable hatchery conditions?

This is a good point raised by the reviewer. The area within Shinnecock Bay where *Mytilus* were collected is well-flushed and not prone to significant decreases in dissolved oxygen or pH or increases in pCO_2. In addition, *Mercenaria* and *Argopecten* within the hatchery at Stony Brook University in Southampton are exposed to similar environmental conditions that are found in the collection sites in Shinnecock Bay from which these original broodstock came. We will clarify the recent origin of the broodstock used in experiments in the methods.

If the presence of algae buffered the carbonate chemistry (p.9, line 23) and this is the mechanism for enhanced growth, this should be visible when Ωcalcite/aragonite is plotted vs. growth. However, the saturation state with Ulva is still considerably below 1 in the highCO2 treatments and the SD is high.

This was an excellent suggestion by the reviewer and for this revision, we have now included regression of $\Omega_{calcite}$ and $\Omega_{aragonite}$ vs. growth which in nearly all cases provided significant correlations. While the Ω is below 1 in many high CO2 cases, prior studied have shown early life stage bivalves do grow, albeit slower, under such conditions (Talmage and Gobler 2010, 2011).

Did the authors measure nutrients at the end of the incubations? It would be interesting to explore their theory that through Ulva presence "the nitrogen assimilation effects on alkalinity outweighed the effects of photosynthetic consumption of DIC" (p.9, line 33)

No, nutrient concentrations were not measured at the beginning or the end of experiments. Due to the multiple water changes that occurred throughout experiments, measuring only the final nutrient concentrations would not accurately represent actual nutrient concentrations throughout.