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Ocean Acidification and Predator-Prey Relations: Correlating Disruption of Predator Avoidance with Chemosensory Deficits

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Introduction

One of the most destructive effects of global climate change is the increased carbon dioxide sequestering and consequent acidification of our world's oceans (Figure 1).

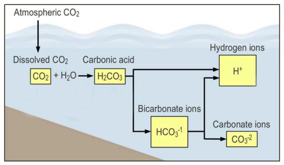


Figure 1: Atmospheric CO_2 acidifies the ocean. Chemical reaction that occurs when atmospheric carbon dioxide is dissolved in the ocean (*NOAA Ocean Acidification Program, 2011*). The dissolved CO_2 in seawater increases acidity (hydrogen ions) of the ocean and thus, reduces the pH.

The impacts of ocean acidification on specific ecosystems are still relatively unknown, especially effects on behavioral ecology of the organisms in those ecosystems. Risk of predation is a critical feature in the behavioral ecology of a wide array of animal species. In the ocean, many prey species respond to chemical signals from their predators with a wide array of anti-predator behaviors. Recent research suggests such behaviors may be compromised by modest increases in acidity (Dixson et al. 2009, Munday et al. 2010, Ferrari et al. 2011, Nilsson et al., 2012). We investigated whether slightly acidic water reduces anti-predator behavior in an easily maintained local prey species.

The blue-banded hermit crab, *Pagurus samuelis*, is a small invertebrate of local tidepools (Laidre and Greggor, 2015). Previous research in the Wright lab showed that exposure of individuals of *P. samuelis* to the scent of the red rock crab (RRC), *Cancer productus*, elicits several anti-predator changes in behavior. In particular, hermit crabs:

1. avoid this scent in a y-maze test.
2. emerge more rapidly from a startle withdrawal response in the presence of the scent.
3. feed more slowly on standard squid pellets in the presence of the scent.

Preliminary experiments found that slightly acidic water eliminates the first two above behaviors. In the present study, we tested whether acidic water would similarly eliminate the third behavioral response: predator-induced reduction in feeding.

Figure 2: *Pagurus samuelis* (blue-banded hermit crab)

Research Question: Does increased acidity compromise predator-induced reduction in feeding?

Hypothesis

We hypothesize:

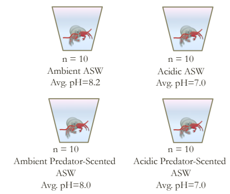
a) that predator essence will significantly reduce feeding rates in seawater at ambient pH
b) this feeding reduction will be eliminated by slightly acidic seawater, resulting in **increased** feeding to predator-free levels.

Figure 3: Equipment used for acidification treatments including: a 5lb tank of CO_2 gas, a regulator valve, separate buckets for acidifying water treatments, a pH meter + calibration buffers.

Methods

Hermit crabs were collected from Little Corona, Newport Beach, CA (33°35'30.46" N 117°52'18.00" W). We tested the feeding rates of hermit crabs in the following treatments:

1. Ambient artificial seawater
2. Acidic artificial seawater
3. Ambient predator-scented seawater
4. Acidic predator-scented seawater



Water Acidification:

We used a 5 lb cylinder to dissolve CO_2 gas into the corresponding water to create the two acidic treatments (Acidic ASW and Acidic Predator-scented ASW; Figure 3). This process mimics the chemical reaction that converts atmospheric CO_2 to increased H^+ ion concentration (acidity) in the oceans (Figure 1). The solutions were constantly stirred until pH was reduced to 7.00 (+/- 0.05).

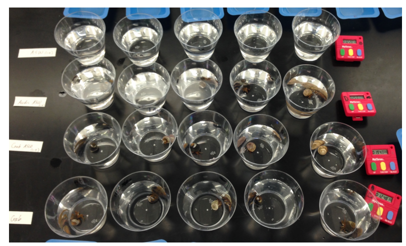
Feeding Trials:

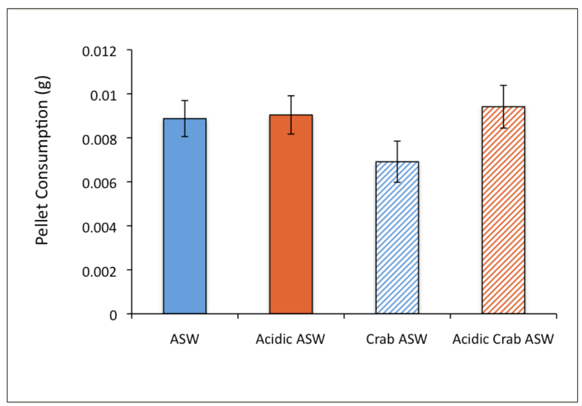
1. We pre-tested hermit crabs for feeding readiness before each experimental trial.
2. We presented hermit crabs with pre-weighed standardized squid pellets (Takagi et. al, 2010) for a 30-minute feeding period.
3. Following this feeding period, we removed the pellets from the hermit crabs and for a second weighing. "Consumption" = change in pellet weight after the feeding assay.

Results

Experiment Set-up

Figure 4. Shown are the four treatments with subjects and squid pellets during an experiment.





Acidity-induced Chemosensory Disruption Normalizes Feeding Rates in Predator-scented Water

Figure 5. Shown is the mean consumption of the four treatments with their corresponding standard-error bars. These data reflect three separate experiments (Figure 4) averaged to show the overall effect of acidic treatments in comparison to ambient treatments of ASW and Predator-Scented ASW. Pellet consumption in Crab ASW (blue stripes) trended lower than that in Acidic Crab ASW (red stripes, two-sample t-test, P = 0.06). Overall F-statistic was not significant ($F_{3,97}$ = 1.723; P = 0.17).

Conclusion

1. The Ambient and Acidic ASW treatments yielded very similar feeding rates (Figure 3) suggesting that acidic ASW did not cause behavioral malaise.
2. There was a noticeable drop in feeding rates for the Crab ASW treatment in comparison to the Ambient ASW; this is the observed anti-predator behavior, however results were not significant (two sample t-test, P=0.12, Figure 3).
3. The feeding rate observed in the Acidic Crab ASW treatment was restored to the feeding rates observed in Ambient ASW (Figure 3). This increased feeding consumption, in comparison to feeding in Crab ASW, although not significant, showed a trend (two sample t-test, P=0.06).
4. Y-maze results were less significant than previous trials conducted in the lab (with prior RRCs). We believe the pheromone release from the current RRC is not as salient as previously used crabs. We will use new individual RRCs as well as different predator species in future trials to yield significant predator-induced inhibition of feeding.

Discussion

Previous research on ocean acidification has concentrated on how increased acidity will impact shell-forming marine invertebrates., while much less attention has been paid to the disruption of the chemical ecology of marine invertebrates (NOAA Ocean Service, 2014). Although our results were not statistically significant, there was a trend that found that predator-induced reduction in feeding rates is eliminated by slight increases in acidity. Previous experiments showing how acidity reduced anti-predator behavior were plagued by the possibility that the increased acidity simply made the animals sick, and thereby disrupted their avoidance or vigilance levels. Given that our result is verified in future experiments, it represents a finding that is not easily explained by behavioral malaise. In particular, the increase in feeding caused by slightly acidic water, in which the predator odor is dissolved (relative to ambient seawater with predator odor), is inconsistent with behavioral malaise, which should reduce, rather than increase, feeding behavior.

Further research needs to be conducted in order to gain more data and understand the trends observed in experiments thus far.. In tide pools along the California coast, acidities have been found to range 1.78 pH units within a 24-hr period, dropping as low as 7.22 before sunrise – a drop of 0.98 pH units from ambient ocean pH of around 8.2 (Kwiatkowski, et al., 2016). In addition to further research and data collection, future testing will also explore varying pH thresholds to further understand the relationship between lowered pH and efficacy of anti-predator behavior. Additionally, future research will explore the use of odors of other predators to further explore how acidity abolishes predator-prey relations in hermit crab feeding assays.

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Figure 6: *Cancer productus* used for predator-scented water.

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