

Effect of Food Resource Availability on Resilience of Eastern Oyster Larvae to Ocean Acidification

Michelle Barbosa, Caroline Schwaner, Teresa Schwemmer, Emmanuelle Pales Espinosa and Bassem Allam

Ocean acidification has become a hot topic in the last decade, as we are rapidly recognizing its implications for life in the ocean. Ocean acidification, also referred to as “the other CO₂ problem” (Doney *et al.* 2009), is one of the forms of climate change we are facing as CO₂ emissions continue to rise. As CO₂ enters the air, primarily by burning fossil fuels (USEPA 2019), most of that CO₂ is taken up by the ocean. Although this may benefit the atmospheric climate by reducing the CO₂ that would otherwise lead to increased atmospheric warming, it can lead to major changes in ocean chemistry. The hydrolysis of CO₂ in water leads to the production of H⁺ ions that results in a decrease of ocean pH. Numerous studies have classified the effects of ocean acidification on development and shell growth, but less studied are the effects of acidification on oyster physiology and the potential mechanisms that may enable vulnerable larvae to survive and adapt. Research described in this article was designed to understand the impacts of ocean acidification on the eastern oyster. Specifically, the study assessed the effects of acidification on survival, growth and metabolic rates of oyster larvae. In addition, the study began to identify the processes that may enable survival by first assessing whether food availability enhances resilience of these early life stages. These questions are particularly important because a decrease in survival and size of larvae can significantly impact the commercial industry as a result of decreased yield. Studying metabolic rates is vital to understanding the health of an organism as it can indicate whether an animal is under stress or has high energetic demands. Since the Industrial Revolution, atmospheric CO₂ has increased that must be met with high food availability. from 280 ppm to a record peak of 415 ppm in May 2019 (Munroe 2019). This increase in CO₂ has resulted in a decrease in ocean pH from 8.2 to 8.1. Although this may seem like a small change, by the end of the century, in the next ~80 years, and under worst-case-scenario conditions, atmospheric CO₂ may rise as high as 1000 ppm and pH is expected to drop to 7.8 globally, according to the Intergovernmental Panel on Climate Change (IPCC 2014). The scale of this increase is occurring at a rate one order of magnitude faster than it has ever occurred in the past one million years (Doney *et al.* 2009). In addition, the pH along coastal areas is even lower on average than the global ocean pH, caused by heightened microbial respiration from eutrophication. In areas such as embayments around Long Island, NY, pH has already reached 7.6 in the summer, markedly lower than that predicted for the global ocean by 2100. For this reason, researchers and stakeholders are

shells and skeletons. This may reduce the growth of calcifying organisms if they cannot get the resources needed to build their shells. In addition, the increased H⁺ ion concentration can lead to dissolution of calcium carbonate shells. The consequences of these reactions can have massive impacts within marine ecosystems and detrimental effects on economies globally.

In 2014, total domestic commercial bivalve landings in the United States exceeded 73,000 t with a total value of \$895 million (National Marine Fisheries Service 2015). A decline in the production of bivalves would hurt the economy of many coastal communities in the US. Current research is seeking to understand the exact impacts of ocean acidification on bivalves so that we can understand the underlying mechanisms and

begin to prepare mitigation strategies for potential losses, such as concerned about the ability for ocean life to acclimate and adapt long term to the changes in the ocean.

Most notably, species that build calcium carbonate shells may be the most vulnerable to changes in ocean chemistry. Along with increasing H⁺ ion concentration in seawater, when CO₂ reacts with water, the carbonate ion concentration in seawater decreases (less saturated), resulting in less carbonate for calcifiers to build their shells. Identification of resilient stocks and species or developing approaches and methods to reduce the impact of acidification on vulnerable species.

Oysters and Ocean Acidification

Many studies suggest that the early life stages of bivalves are highly vulnerable to the effects of ocean acidification. This includes the eastern oyster *Crassostrea virginica*. Prior to settling and becoming recognizable juvenile oysters, eastern

oysters spend the first few weeks of life as vulnerable, free-swimming larvae. As larvae, they pass through several developmental stages and begin to grow a calcium carbonate shell. If the supply of calcium carbonate during development is limited, larvae may not develop properly or die.

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and embryos were exposed to seawater with a pH similar to

FIGURE 2. *Percent survival (a), size (b) and respiration rates (c; $\mu\text{mol O}_2$ consumed per individual per hour) of eastern oyster larvae grown under ambient and elevated $p\text{CO}_2$. Mean \pm SE. * indicates a significant difference ($p < 0.05$) between treatments.*

to compensate, larvae increased respiration rates. As metabolic

the study assessed the effects of acidification on survival, growth and metabolic rates of oyster larvae. In addition, the study began to identify the processes that may enable

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important because a decrease in survival and size of larvae can significantly impact the commercial industry as a result of decreased yield. Studying

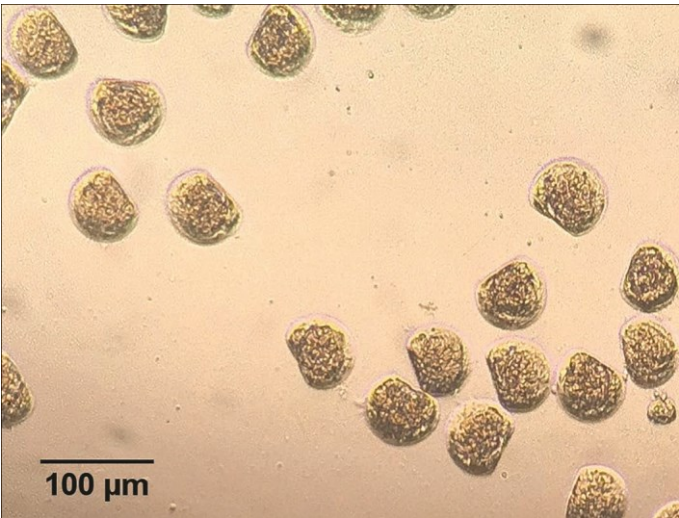


FIGURE 1. Larval (24-h old) eastern oysters.

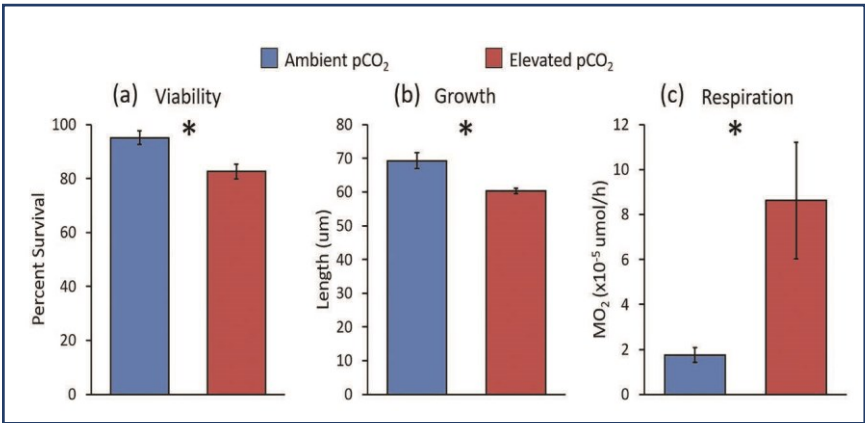
Changes in Physiology

Viability of oysters exposed to future pH conditions was significantly reduced compared to oysters maintained at ambient pH conditions (Fig. 2a). Oysters exposed to elevated CO_2 were significantly smaller in length than those in the ambient condition after just 24 hours of exposure (Fig. 2b). Oysters under elevated CO_2 consumed significantly more oxygen and, therefore, had greater

respiration rates than those under ambient conditions (Fig. 2c). Increased respiration rates are indicative of high stress under acidified conditions and more energy spent on survival.

Food Limitation

Larvae were adversely impacted by ocean acidification and that, perhaps



survival by first assessing whether food availability

early life stages. These questions are particularly

metabolic rates is vital to understanding the health of an organism as it can indicate whether an animal is under stress or has high energetic demands that must be met with high food availability.

Experimental Design

Eastern oysters were spawned the average global, rates are defined as ambient pH (ambient condition, pH 8.1, pCO₂ 400 ppm) or seawater with a pH predicted for the end of the century (elevated condition, pH ~7.5, pCO₂ 1000 ppm). Developing larvae (Fig. 1) were held at 10 larvae/mL at 23 C in a standing system with target pH maintained by diffusing ambient air or CO₂ mixed with air. After 24 hours, the number of live larvae was assessed by microscopic observation. Growth was assessed using the ImageJ image analysis program on formalin-preserved samples. Respiration rates were measured by adding two-week-

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This study identified some of the challenges early life stage eastern oysters may face and begins to answer questions regarding processes involved in larval resilience to acidification. Energy metabolism is a heritable trait in many species and the evidence of an energy allocation mechanism to cope with acidification stress drives us to further question the molecular features, such as genetic components and molecular pathways orchestrating this mechanism and associated with resilience. More research is needed to identify genes under selection and measure the heritability of potential traits associated with resilience to ocean acidification to determine

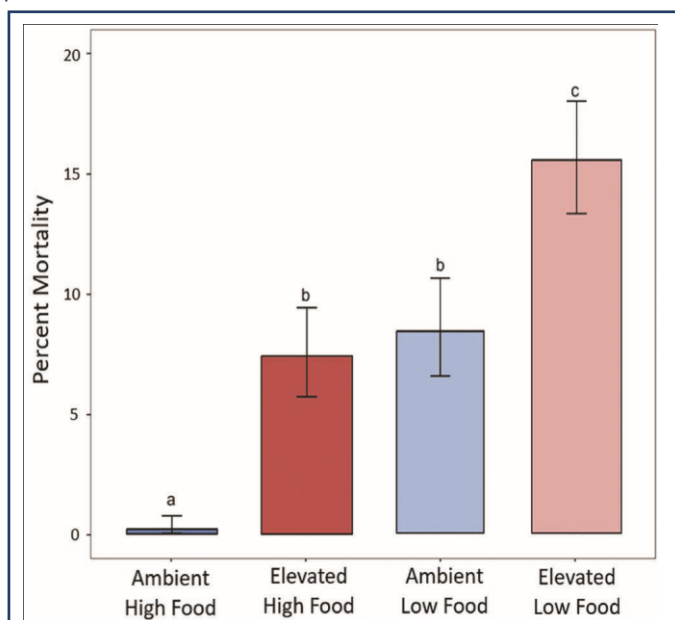
old larvae to a sealed, air-free chamber and measuring oxygen consumption over time.

whether eastern oysters can adapt in the face of climate change. Furthermore, the identification of markers of resistance can be used by the aquaculture industry to identify and promote strains of oysters that are “winners” under ocean acidification and that can overcome the future environmental challenges eastern oysters will certainly

better than oysters under acidification stress and food limitation.

To test this, 24-hr old oyster larvae were held in the same standing experiment as previously described and exposed to

FIGURE 3. Percent mortality for larval eastern oysters (mean \pm 95 percent confidence interval) under ambient and elevated pCO₂ and two different food concentrations. Different letters indicate a significant difference at $p < 0.05$.



face.

either ambient or elevated CO₂ conditions and given a high-food diet (100 percent of the recommended algal quantity per larvae) or low-food diet (10 percent of the recommended algal quantity per larvae) yielding four different treatments total

available energy in algae, then they should be able to cope with the stress of acidification

the daily amount of energy an individual requires over a period of time, increasing metabolic rates therefore increases energy demands. If oyster larvae require more energy under conditions of ocean acidification, there may be some mechanism involving energy availability that helps larvae tolerate the stress. Based on this assumption, we tested the hypothesis that resilience to ocean acidification is related to some energy allocation mechanism and the availability of food resources in the form of algae. If oysters are not limited with regard to

(Ambient CO₂ + High Food, Elevated CO₂ + High Food, Ambient CO₂ + Low Food and Elevated CO₂ + Low Food). Oysters were fed live *Tisochrysis lutea* cultured in F/2 media in the laboratory.

After 24 hours under elevated CO₂ concentration, moderate starvation significantly increased the mortality of larval oysters (15.5 percent) compared to those given abundant food resources (7.4 percent) (Fig. 3). Starvation also affected oysters held under ambient conditions with mortality at 8.4 percent under moderate starvation and 0.2 percent under abundant food conditions. Interestingly, moderate starvation under ambient conditions results in similar levels of mortality as elevated CO₂ conditions suggesting both are stressors of equal magnitude and that the stress of food limitation and low pH are additive. The results suggest larval oysters are using some energy allocation mechanism that enables diversion of energy to compensate for the stress of acidification. When energy resources are limited, oysters do not have sufficient energy to maintain homeostasis and become more susceptible to acidification stress.

Conclusions

This study identified some of the challenges early life stage eastern oysters may face and begins to answer questions regarding processes involved in larval resilience to acidification. Energy metabolism is a heritable trait in many species and the evidence of an energy allocation mechanism to cope with acidification stress drives us to further question the molecular features, such as genetic components and molecular pathways orchestrating this mechanism and associated with resilience. More research is needed to identify genes under selection and measure the heritability of potential traits associated with resilience to ocean acidification to determine whether eastern oysters can adapt in the face of climate change. Furthermore, the identification of markers of resistance can be used by the aquaculture industry to identify and promote strains of oysters that are “winners” under ocean acidification and that can overcome the future environmental challenges eastern oysters will certainly face.

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Notes

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