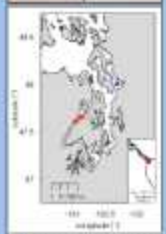
Krill (*Euphausia pacifica*) Development in the Laboratory is Impaired at Currently Observed pCO_2 Levels

Anna K. McLaskey[1*], Julie E. Keister[1], Paul McElhany[2], M. Brady Olson[3], D. Shallin Busch[2,4], Michael Maher[2], Amanda K. Winans[1]
[1]University of Washington, School of Oceanography, USA [2]NOAA, Northwest Fisheries Science Center, USA [3]Western Washington University, Shannon Point Marine Center, USA [4]NOAA, Ocean Acidification Program, USA

Background

Euphausia pacifica is a dominant euphausiid and important prey species throughout the North Pacific. Despite the importance of euphausiids in marine ecosystems around the world there has been limited ocean acidification (OA) research on them. Previous studies have found that Antarctic krill, *Euphausia superba*, have reduced hatching at 1250 µatm pCO_2 (pH 7.50) and complete hatching failure at 2000 µatm pCO_2 (pH 7.36-7.40); however, larvae have only been monitored to three days post hatch (Kawaguchi et al. 2011, 2013). Sub-adults of the north Atlantic krill *Nyctiphanes couchii* have reduced survival at pCO_2 concentrations between 1100 and 1700 µatm pCO_2 (pH 7.63 and 7.47), but other life stages have not been tested (Sperfeld et al. 2014).

Puget Sound is a large estuary connected to the California Current Ecosystem (CCE), an upwelling system that has low and declining pH throughout the water column. Within the estuary, OA is exacerbated by freshwater runoff and the respiration of organic matter. The vertical distribution of *E. pacifica* larval stages is an important determiner of their exposure to low pH waters but is poorly characterized, and their response to OA has not been tested in the lab.



Field sampling stations (red) and collection site for experimental females (blue)

Methods

Field Exposure: We characterized the pH environment experienced by *E. pacifica* eggs and larvae with depth stratified net tows and carbonate chemistry measurements at two stations in the northern end of Hood Canal in Puget Sound, WA during April and June 2012.

Laboratory Experiments: We collected adult females and spawned them under a wide range of pCO_2 conditions in the laboratory, tracking hatching success, larval development, and survival. Two sets of experiments (8 experimental trials total) were done in two separate experimental systems.

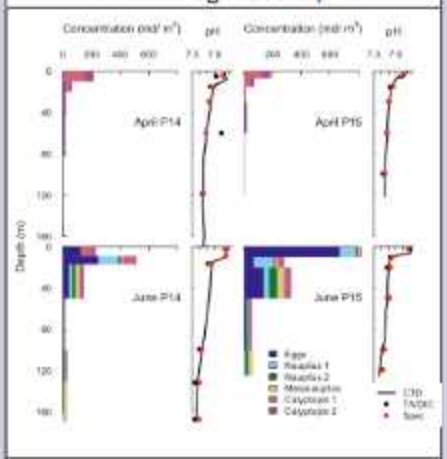
During the first set of experiments, eggs were raised to five days post hatch inside sealed 500 ml jars of pCO_2-equilibrated seawater, with water changes and chemistry monitored every two days. Equilibrated seawater was generated by bubbling with CO_2 and CO_2-free air while monitoring pH.

During the second set of experiments, krill were raised to the Calyptopis 2 stage (10-21 days post hatch) in open dishes held inside sealed boxes with atmospheric gas of treatment pCO_2 levels. Air with precise pCO_2 concentrations was mixed with mass flow controllers.

Experiment 1: Krill held in sealed jars of seawater with treatment pCO_2 levels

Experiment 2: Krill held in box with atmosphere of treatment pCO_2

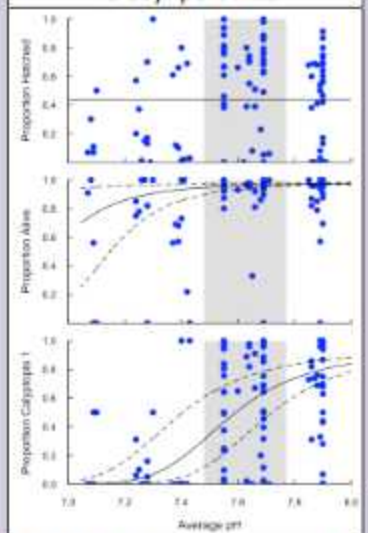
pH exposure of *E. pacifica* eggs and larvae in Puget Sound, WA



Highest concentrations found at surface, but significant portions throughout the water column (20-180m) where they were exposed to pH 7.8-7.5

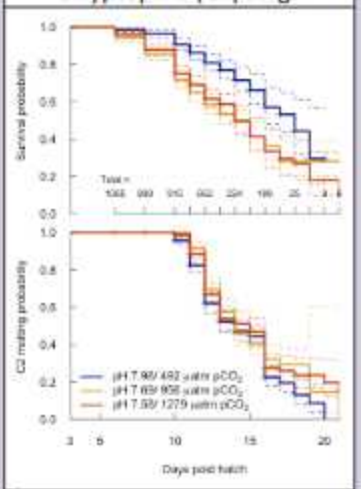
- 40% of Eggs & Nauplius 1 below 20 m
- 90% of Nauplius 2 & Metanauplius below 20 m and 35-65% below 50 m
- 30-89% of Calyptopis 1 & 2 below 20 m

Hatching, survival, development at 5 days post hatch



Hatching was robust to very low pH; small reduction in 5 day survival at very low pH; development (to C1 stage) slows at pH <7.8 and declines rapidly below pH 7.6.

Survival & development to Calyptopis 2 (C2) stage



Survival probability (top panel) from three days post hatch to the C2 stage was reduced at pH 7.69 (yellow) and pH 7.58 (red) compared to pH 7.96 (blue). No differences in time to molt to C2 were detected (bottom panel). Dashed lines show 95% CI.

Conclusions:

E. pacifica larval stages, particularly the Nauplius 2 and Metanauplius stages, were found throughout the water column (20-180m), where they were exposed to pH 7.8-7.5. In the laboratory, *E. pacifica* hatching was robust to a wide range of pH levels, but larval development and survival were reduced at lower pH. Development to the Calyptopis 1 stage was slowed at low pH. Survival from three days post hatch to the Calyptopis 2 stage was reduced by an average of 20% at pH 7.69 compared to pH 7.96. *E. pacifica* may be living near the limits of its pH tolerance and continued OA could push these organisms past their threshold, with negative consequences for their populations and higher trophic levels.

Kawaguchi S, Ishida A, King R, Raymond B, Waller N, Constable A, Nicol S, Wakita M, Ishimatsu A (2013) Risk maps for Antarctic krill under projected Southern Ocean acidification. Nat Clim Chang 3:843–847
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