

# Egg production and hatching of acclimated and non-acclimated copepods *Calanus pacificus* and *Acartia hudsonica* exposed to varying CO<sub>2</sub> conditions, 2014-2015 ((OA-Copepod\_PreyQual project)

**Website:** <https://www.bco-dmo.org/dataset/670366>

**Data Type:** experimental

**Version:**

**Version Date:** 2017-01-18

## Project

» [Impacts on copepod populations mediated by changes in prey quality](#) (OA-Copepod\_PreyQual)

## Program

» [Science, Engineering and Education for Sustainability NSF-Wide Investment \(SEES\): Ocean Acidification \(formerly CRI-OA\)](#) (SEES-OA)

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## Dataset Description

This dataset includes egg production, hatching success, and naupliar development data for two species of calanoid copepods, *Calanus pacificus* and *Acartia hudsonica*, from 2014 and 2015 studies. The females, eggs and young were maintained in three levels of pCO<sub>2</sub> to study the effect of ocean acidification on copepod reproduction.

## Methods & Sampling

**2014 Study:** Adult female *Calanus pacificus* were collected with a 1-m diameter, 335- $\mu$ m mesh, ring net lifted vertically from 200m to the surface in Rosario Strait. During the morning (0900-1130 7/14/14; 0800-11:45 8/4/14) copepods were transported back to Shannon Point Marine Center (SPMC) where mature adult females were separated under the microscope and placed in 500 ml jars of filtered (5  $\mu$ m) seawater at a concentration of 30 females/L. Jars of adult females were given a 75% water change daily and were fed pCO<sub>2</sub> acclimated prey twice daily. Experiment 1 (EP1, EP2, EP3) was fed *Prorocentrum micans* and Experiment 2 (EP4, EP5, EP6) was fed *Ditylum brightwellii*.

During Experiment 2 (*Ditylum*) in addition to feeding every 12 hours, jars were also stirred 6 hours after each feeding to resuspend cells that had settled out of the water column. Females were acclimated to pCO<sub>2</sub>

conditions and pCO<sub>2</sub>-acclimated prey for 7 days.

The direct effects of pCO<sub>2</sub> on *C. pacificus* egg production, hatching success, and naupliar development were tested at the beginning of each experiment before adults were acclimated to pCO<sub>2</sub> levels and fed pCO<sub>2</sub>-acclimated prey. Immediately after collection from the field, twenty females were placed individually in 250 ml containers of pCO<sub>2</sub>-equilibrated seawater and incubated overnight (female *Calanus* are known to spawn between 00:00 and 07:00 Runge 1985). The following morning females were removed and measured and any eggs they spawned were counted and left in the jars (with water change) for eggs to hatch and nauplii to develop for 4 days (EP1, direct effects, was 5 days), at which time approximately 50% of the nauplii had reached the nauplius 3 stage. Because nauplius 3 is the first feeding stage nauplii were not fed. After 4 days the jars were checked for naupliar survival and all nauplii were preserved in 5% buffered formalin/seawater for counting and staging. Hatching success was calculated from the number of hatched nauplii and unhatched eggs found at the end of the experiment; naupliar survival was calculated from the proportion of hatched nauplii that were alive at the end of the experiment; development was calculated as the proportion of hatched nauplii that reached nauplius 3.

After seven days of acclimation, twenty females from each treatment were again placed individually in 250 ml containers of pCO<sub>2</sub>-equilibrated seawater and incubated overnight. The eggs and nauplii were treated the same way as with the direct effects.

**2015 Study:** During Exp 1 (12°C) *Acartia hudsonica* were maintained at three target pCO<sub>2</sub> levels, 400, 800, 1200 µatm, and were fed *Rhodomonas salina* cultured at the same pCO<sub>2</sub> levels. We assessed the reproductive output and larval development of *A. hudsonica* at each pCO<sub>2</sub> level before and after the acclimation period. At the beginning of the experiment females were set up for initial egg production, hatching success, and naupliar development tests (described below) and the rest of the adults were divided into 45 adults per 500 mL jar of pCO<sub>2</sub>-equilibrated seawater and held at target pCO<sub>2</sub> levels for the duration of the acclimation period. During the acclimation period jars of adult females were given a 75% water change daily and fed pCO<sub>2</sub> acclimated *R. salina* every 12 hours to maintain a concentration above the saturation feeding density of 3000 cells/mL while allowing for a maximum grazing rate of 6000 cells/hr/female.

In Exp 1 (12°C) the acclimation period was five days. On day 3 some females were added to jars of males at a ratio of 1 female: 2 males to ensure they would be fertilized for egg production, hatching, and development experiments. On day 6 of the experiment females that were incubated with males were used for egg production and subsequent hatching and naupliar development tests. Egg production, hatching, and naupliar development before and after the acclimation period were both tested in two separate trials in Exp 1, however, due to logistical constraints, the 800 µatm pCO<sub>2</sub> target treatment was only included in one trial.

In all egg production, hatching, and naupliar development tests, adult females were incubated individually inside mesh-bottom egg production chambers within 250 ml containers of target treatment pCO<sub>2</sub>-equilibrated seawater. After 24 hours the females were removed, measured, and the eggs were placed back into the atmospheric simulation chambers to develop. Nauplii were fed *R. salina* once per day at 25% the amount calculated for adults (described above). During tests of the direct effects of pCO<sub>2</sub> on *A. hudsonica* naupliar development the nauplii were fed stock *R. salina* that was not pCO<sub>2</sub> acclimated; during tests at the end of the acclimation period nauplii were fed *R. salina* that was cultured at the corresponding target pCO<sub>2</sub> treatment. At the conclusion of the naupliar development tests, 10% of the jars were checked for naupliar survival and all were preserved in 5% buffered formalin/seawater solution for counting and staging. Hatching success was calculated from the number of hatched nauplii and unhatched eggs found at the end of the experiment; naupliar development was calculated by the proportion of hatched nauplii that reached the Nauplius IV stage (N IV). In Exp 1 (12°C) eggs and nauplii were allowed to develop for 8 days, and were fed pCO<sub>2</sub>-acclimated *R. salina* starting on day 4.

## Data Processing Description

These are unprocessed, raw data.

### BCO-DMO Processing Notes:

- added conventional header with dataset name, PI name, version date
- modified parameter names to conform with BCO-DMO naming conventions
- combined data for all 2014 and 2015 egg production experiments
- added columns for species and year
- replaced 'ND' and blank cells with 'nd', no data

2017-01-18: added column to data denoting whether or not the females had been acclimated to the pCO<sub>2</sub> level.

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## Data Files

File
<b>copepod_egg_prod.csv</b> (Comma Separated Values (.csv), 73.33 KB) MD5:a42836d0614328f4496b50e65a3b76a9 Primary data file for dataset ID 670366

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## Parameters

Parameter	Description	Units
species	experimental copepod species	unitless
year	year of study	unitless
trial	trial identifier	unitless
jar	jar number	unitless
pCO2_target	target pCO2 level	parts per million by volume (ppmv)
length_prosome	length of female prosome	millimeters (ml)
eggs_spawned	number of eggs spawned	eggs
N4_prop	proportion of hatched nauplii that reached nauplius 4 stage; NA=not applicable	unitless
N3_prop	proportion of hatched nauplii that reached nauplius 3 stage; NA=not applicable	unitless
N2_prop	proportion of hatched nauplii that reached nauplius 2 stage; NA=not applicable	unitless
N1_prop	proportion of hatched nauplii that reached nauplius 1 stage; NA=not applicable	unitless
hatch_prop	proportion of eggs that hatched; NA=not applicable	unitless
dead_prop	proportion of hatched nauplii that died over the course of the experiment; NA=not applicable	unitless
mortality_check_flag	YES/NO was the jar check for mortalities at the end of the experiment	unitless
acclimated_or_not	Whether or not the females were acclimated to the treatment pCO2 level.	unitless

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## Deployments

## Lab\_Olson\_B

<b>Website</b>	<a href="https://www.bco-dmo.org/deployment/521277">https://www.bco-dmo.org/deployment/521277</a>
<b>Platform</b>	WWU
<b>Start Date</b>	2011-03-31
<b>End Date</b>	2016-09-15
<b>Description</b>	laboratory experiments

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## Project Information

### Impacts on copepod populations mediated by changes in prey quality (OA-Copepod\_PreyQual)

**Coverage:** Puget Sound, Salish Sea

Research shows that ocean acidification (OA) has physiological consequences for individual organisms, even those lacking calcium carbonate skeletal structures. However, this existing research does not adequately address how OA effects to individuals are linked across trophic levels. Pelagic copepods are critical players in most marine biogeochemical cycles. Their consumption of phytoplankton and microzooplankton is the primary mechanism by which bacterial and phytoplankton production is transferred to higher trophic levels. Despite their high abundance and ecological importance, copepods have received little research attention concerning OA. The few extant studies focused on direct acute effects to copepods (e.g. egg hatching, survival) under elevated pCO<sub>2</sub>, and few significant effects have been observed at predicted future pCO<sub>2</sub>. However, there is increasing recognition that OA significantly affects their phytoplankton prey, including elevating growth rates, increasing cell sizes, altering nutrient uptake and ratios, and chemical composition. Because copepod grazing, egg production, and hatching success all can vary with these prey characteristics, OA mediated changes in phytoplankton quality may be an important indirect mechanism through which OA acts on copepod populations and, ultimately, marine food webs.

This study that will advance our understanding of how copepod populations may be affected by OA, specifically through OA induced changes in phytoplankton quality. Our core objective is to determine how changes in phytoplankton physiology and biochemistry (e.g. lipid composition) affect copepod egg production, hatching, and ontogenetic development of nauplii. We will also include a subset of experiments to test whether OA affects copepod reproductive output independent of changes to prey. To achieve these research goals, the diatom, *Ditylum brightwellii*, and dinoflagellate, *Prorocentrum micans*, will be cultured semi-continuously under several pCO<sub>2</sub> concentrations, during which time we will characterize changes in their physiology and biochemistry. The copepods, *Calanus pacificus*, a large, high lipid-bearing marine species, and *Acartia clausi*, a smaller, low lipid-bearing estuarine species, will be maintained across varying pCO<sub>2</sub> concentrations and fed these pCO<sub>2</sub>-acclimated prey, and their grazing and reproductive capability quantified. The copepods and phytoplankton used in this study will be collected from the Salish Sea, a region already experiencing periods of high pCO<sub>2</sub>/H<sup>+</sup> (>1000 ppm, pH 7.5) on varying timescales. Therefore, this research addresses a question of how future climate change may impact marine ecosystems, but also is relevant to pCO<sub>2</sub>/H<sup>+</sup> variability presently experienced in coastal environments.

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## Program Information

**Science, Engineering and Education for Sustainability NSF-Wide Investment (SEES): Ocean Acidification (formerly CRI-OA) (SEES-OA)**

**Website:** [https://www.nsf.gov/funding/pgm\\_summ.jsp?pims\\_id=503477](https://www.nsf.gov/funding/pgm_summ.jsp?pims_id=503477)

**Coverage:** global

NSF Climate Research Investment (CRI) activities that were initiated in 2010 are now included under Science, Engineering and Education for Sustainability NSF-Wide Investment (SEES). SEES is a portfolio of activities that highlights NSF's unique role in helping society address the challenge(s) of achieving sustainability. Detailed information about the SEES program is available from NSF ([https://www.nsf.gov/funding/pgm\\_summ.jsp?pims\\_id=504707](https://www.nsf.gov/funding/pgm_summ.jsp?pims_id=504707)).

In recognition of the need for basic research concerning the nature, extent and impact of ocean acidification on oceanic environments in the past, present and future, the goal of the SEES: OA program is to understand (a) the chemistry and physical chemistry of ocean acidification; (b) how ocean acidification interacts with processes at the organismal level; and (c) how the earth system history informs our understanding of the effects of ocean acidification on the present day and future ocean.

**Solicitations issued under this program:**

[NSF 10-530](#), FY 2010-FY2011

[NSF 12-500](#), FY 2012

[NSF 12-600](#), FY 2013

[NSF 13-586](#), FY 2014

NSF 13-586 was the final solicitation that will be released for this program.

**PI Meetings:**

[1st U.S. Ocean Acidification PI Meeting](#) (March 22-24, 2011, Woods Hole, MA)

[2nd U.S. Ocean Acidification PI Meeting](#) (Sept. 18-20, 2013, Washington, DC)

[3rd U.S. Ocean Acidification PI Meeting](#) (June 9-11, 2015, Woods Hole, MA - Tentative)

**NSF media releases for the Ocean Acidification Program:**

[Press Release 10-186 NSF Awards Grants to Study Effects of Ocean Acidification](#)

[Discovery Blue Mussels "Hang On" Along Rocky Shores: For How Long?](#)

[Discovery nsf.gov - National Science Foundation \(NSF\) Discoveries - Trouble in Paradise: Ocean Acidification This Way Comes - US National Science Foundation \(NSF\)](#)

[Press Release 12-179 nsf.gov - National Science Foundation \(NSF\) News - Ocean Acidification: Finding New Answers Through National Science Foundation Research Grants - US National Science Foundation \(NSF\)](#)

[Press Release 13-102 World Oceans Month Brings Mixed News for Oysters](#)

[Press Release 13-108 nsf.gov - National Science Foundation \(NSF\) News - Natural Underwater Springs Show How Coral Reefs Respond to Ocean Acidification - US National Science Foundation \(NSF\)](#)

[Press Release 13-148 Ocean acidification: Making new discoveries through National Science Foundation research grants](#)

[Press Release 13-148 - Video nsf.gov - News - Video - NSF Ocean Sciences Division Director David Conover answers questions about ocean acidification. - US National Science Foundation \(NSF\)](#)

[Press Release 14-010 nsf.gov - National Science Foundation \(NSF\) News - Palau's coral reefs surprisingly resistant to ocean acidification - US National Science Foundation \(NSF\)](#)

[Press Release 14-116 nsf.gov - National Science Foundation \(NSF\) News - Ocean Acidification: NSF awards \\$11.4 million in new grants to study effects on marine ecosystems - US National Science Foundation \(NSF\)](#)

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## Funding

Funding Source	Award
<a href="#">NSF Division of Ocean Sciences (NSF OCE)</a>	<a href="#">OCE-1220664</a>
<a href="#">NSF Division of Ocean Sciences (NSF OCE)</a>	<a href="#">OCE-1220381</a>

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