Lab study on the effect of pH and oxygen fluctuations on mussel adhesive plaques with mussels collected from Penn Cove Shellfish in Coupeville, Washington.

Website: https://www.bco-dmo.org/dataset/785238
Data Type: experimental
Version: 1
Version Date: 2019-12-30

Project
» Effects of Ocean Acidification on Coastal Organisms: An Ecomaterials Perspective (OA - Ecomaterials Perspective)

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

Contributors	Affiliation	Role
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Abstract
Data generated from laboratory experiments that investigated the influence of fluctuating environmental conditions on the attachment strength of byssal threads as they aged. Mussels (M. trossulus) were collected from Penn Cove Shellfish, Quilcene Bay, Quilcene, Washington, USA Penn Cove Shellfish hatchery, Quilcene Bay, Quilcene, Washington, USA [47°47’48.0” N, 122°51”16.6” W] and held in experimental aquaria at the University of Washington in Seattle, Washington, USA for up to 14 days. Mussels produced threads over the course of 4 hrs that were aged in fluctuating oxygen and pH conditions for up to 20 days. Adhesive plaques were then pulled to failure to determine adhesion strength. A second cohort of mussels was placed in static pH and Oxygen treatments, recording the number of threads produced by each over one week.

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Coverage

Spatial Extent: **Lat**:47.796 **Lon**:-122.855
Temporal Extent: 2015-11 - 2016-02

Dataset Description

Data generated from laboratory experiments that investigated the influence of fluctuating environmental conditions on the attachment strength of byssal threads as they aged. Mussels (*M. trossulus*) were collected from Penn Cove Shellfish, Quilcene Bay, Quilcene, Washington, USA Penn Cove Shellfish hatchery, Quilcene Bay, Quilcene, Washington, USA [47°47’48.0” N, 122°51”16.6” W] and held in experimental aquaria at the University of Washington in Seattle, Washington, USA for up to 14 days. Mussels produced threads over the course of 4 hrs that were aged in fluctuating oxygen and pH conditions for up to 20 days. Adhesive plaques were then pulled to failure to determine adhesion strength. A second cohort of mussels was placed in static pH and Oxygen treatments, recording the number of threads produced by each over one week.

Methods & Sampling

Adult mussels (*Mytilus trossulus*, Gould 1850) were gathered from the top of aquaculture rope lines at the Penn Cove Shellfish hatchery, Quilcene Bay, Quilcene, Washington, USA (47°47’48.0” N, 122°51”16.6” W) during the winter of 2015 (November 2015 to February 2016), transported on ice to the laboratory, and kept in 50 L aquaria. Aquaria typically contained 20-30 mussels and were filled with 0.2 µm filtered, UV-sterilized seawater, with constant aeration. Mussels were in the laboratory for no longer than three weeks and were fed Shellfish Diet 1800 (Reed Mariculture, Campbell, CA) up to 5% of their wet tissue mass day-1, dispensed at a concentration of 2000 algal cells ml-1, a diet that has been shown to maintain body weight for up to one month (unpublished data). After a week of acclimation, mussels either produced threads that were included in plaque-curing experiments or the animal itself was included in a thread production assay.

Byssal threads were collected in the laboratory by securing mussels to mica plates with rubber bands, orienting the valve opening towards the substrate and allowing them to attach under seawater conditions that mimicked those found in the open-ocean (pH *ca.* 8, O2 *ca.* 8.5 mg L-1, Sal *ca.* 30, T *ca.* 9°C). After four hours, threads were separated from each animal at the shell margin by cutting the proximal region of each thread, preserving the attachment with each plate. Plates with attached threads were then incubated in seawater treatments, using only plates from mussels that made three or more attachments. After incubation, plates were removed from seawater, dried, and stored for up to two weeks before mechanical testing was performed.

To determine whether rare, extreme excursions in pH and dissolved oxygen can directly affect the plaque-curing process, plaques were aged to maturity in fluctuating seawater treatments that mimicked the magnitude and duration of the ‘worst-case’ scenario, as defined by the most extreme excursion observed in field measurements (pH <5.0 or O2 <2 mg L-1, for 5 days). Mica plates with freshly attached (*ca.* 4 hrs after deposition) threads were haphazardly assigned to one of five experimental treatments, controlled using the pH and oxygen-stat system previously described. Threads aged in the first two experiments experienced constant dissolved oxygen (*ca.* 8 mg L-1), temperature (*ca.* 9ᵒC), and salinity conditions (*ca.* 29), while also being subjected to an excursion in seawater pH (pH *ca.* 5.0) after either 1 (Exp2) or 8 (Exp3) days. The second two experiments mimicked the conditions of the first, except that seawater pH was maintained at *ca.* 8.0 throughout and threads were exposed to hypoxia excursions (O2 <2 mg L-1) either at 1 (Exp4) or 8 (exp5) days into the experiment. pH and oxygen excursions were maintained for 5 days, after which conditions returned to a baseline that represented open-ocean conditions (pH *ca.* 8.0, O2 *ca.* 8.5, T *ca.* 9ᵒC, Sal *ca.* 29). A subset of plates was removed within each experiment after either 3, 5, 8, 12, or 20 days and stored dry for up to two weeks before mechanical testing was performed. A control treatment (Exp1) wherein open-ocean conditions were maintained for 20 days was also performed with the same sampling regime.

Byssal thread production during acidification and hypoxia excursions was investigated by placing mussels secured to mica plates in one of five pH treatments (Exp6; pH target = 5.0, 6.0, 7.0, 7.5, or 8.0) or one of two dissolved oxygen treatments (Exp7; O2 target = <2.0 or >8.0 mg L-1) for seven days. pH treatments were maintained using a pH-stat system similar to the one described in O’Donnell et al. (2013). Briefly, seawater pH (NBS) and temperature (°C) were measured with a Honeywell Durafet III pH electrode and monitored with a Honeywell UDA2182 analyzer that controlled the operation of a solenoid valve. The solenoid value regulated the flow of CO2 into the aerator of each tank. Using a PID loop, the analyzer tailored a CO2:air mixture by controlling the proportional operation of the valve, using pH as the response variable. Dissolved oxygen treatments were accomplished in a similar way by equipping the analyzer with a Honeywell DL5000 equilibrium oxygen probe (accuracy ± 0.1) and replacing the CO2 cylinder with N2 gas. The salinity in each treatment was monitored with a Honeywell DL4000 conductivity cell (accuracy ± 1), which was also monitored by the analyzer. pH, oxygen, temperature, and salinity were logged every 10 minutes using a 4-20 mA data logger. Any pre-existing byssal threads were removed from each mussel, by cutting threads in the proximal region at the shell margin, prior to being placed in a treatment. Once in a treatment, a subset of mussels (*ca.* 20) were removed at 1, 3, 5, and 7 days, counting the number of new threads each mussel produced.

Plaque attachment strength was determined by gripping the distal region of each byssal thread and pulling perpendicular (90ᵒ) to the substrate until failure, following the protocol of George & Carrington (2018). This testing angle was chosen for its reproducibility; it should be noted that the contact angle of the thread with the plaque varies and threads are rarely brought into tension fully perpendicular to the substrate (Desmond et al. 2015). Plaques were rehydrated in their respective seawater treatments prior to mechanical testing for more than 5 minutes. The thread distal region was gripped with a hemostat *ca.* 1 mm above the plaque-thread junction, and force was recorded using a 10 N digital force gauge (OMEGA, Stamford, CT, USA; accuracy ± 0.01 N) attached to a motor-driven testing frame. Threads were pulled at an extension of 10 mm min-1 until plaque failure (the distal region is much stronger than the plaque; Bell & Gosline 1996) and force (N) were recorded at 20 Hz. The adhesion strength (kPa) of each plaque was determined by normalizing the maximum force required to dislodge each plaque by the attachment planform area (mm2), measured by tracing the outline of each plaque from above using a dissection scope with accompanying AmScope MU1000 camera (Irvine, CA, USA) and AmScope X imaging software prior to testing (Burkett et al. 2009). The mean adhesion strength of 3-5 plaques is reported for each mussel.

In an effort to link observed differences in plaque adhesion with the failure mechanics of the adhesive, the failure mode of each plaque was also scored visually during mechanical testing following Young & Crisp (1982) as outlined in George & Carrington (2018). Briefly, plaques were binned within three failure types: adhesive, peeling, or tearing. In the case of adhesive failure, plaques detached from the substrate in a single, swift, plunger like motion. Peeling failure was characterized by a detachment beginning at a location along the perimeter of the plaque, propagating from one side of the structure to the other. Tearing failure was evident when a portion of the plaque remained attached to the substrate after the test was completed, or the thread became dislodged from the attachment plaque at the thread-plaque junction.

Detailed methods and results are provided in George et al. (in press).

Data Processing Description

BCO-DMO Data Manager Processing Notes:

- converted lat/lon listed in the description to decimal degrees for Osprey page.

- added a conventional header with dataset name, PI name, version date

- blank values in this dataset are displayed as "nd" for "no data." nd is the default missing data identifier in the BCO-DMO system.

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Data Files

File
data3.csv (Comma Separated Values (.csv), 39.91 KB) MD5:38f3ffe7f9d07c4ea18d33e527cf3dd5 Primary data file for dataset ID 785238

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Related Publications

Bell, E.C., Gosline, J.M. (1996). Mechanical design of mussel byssus: material yield enhances attachment strength. Journal of Experimental Biology 199(Pt4): 1005–1017.
Methods

Burkett, J. R., Wojtas, J. L., Cloud, J. L., & Wilker, J. J. (2009). A Method for Measuring the Adhesion Strength of Marine Mussels. The Journal of Adhesion, 85(9), 601–615. doi:10.1080/00218460902996903
Methods

Desmond, K. W., Zacchia, N. A., Waite, J. H., & Valentine, M. T. (2015). Dynamics of mussel plaque detachment. Soft Matter, 11(34), 6832–6839. doi:10.1039/c5sm01072a https://doi.org/10.1039/C5SM01072A
Methods

George, M. N., & Carrington, E. (2018). Environmental post-processing increases the adhesion strength of mussel byssus adhesive. Biofouling, 34(4), 388–397. doi:10.1080/08927014.2018.1453927

Methods

George, M. N., Andino, J., Huie, J., & Carrington, E. (2019). Microscale pH and Dissolved Oxygen Fluctuations within Mussel Aggregations and Their Implications for Mussel Attachment and Raft Aquaculture. Journal of Shellfish Research, 38(3), 795. https://doi.org/10.2983/035.038.0329
Methods

O'Donnell, M. J., George, M. N., & Carrington, E. (2013). Mussel byssus attachment weakened by ocean acidification. Nature Climate Change, 3(6), 587–590. doi:10.1038/nclimate1846
Methods

Young GA, Crisp D. 1982. Marine animals and adhesion. In: KW Allend Ed Adhes. Vol. 6. England: Barking, Applied Science Publishers, Ltd.; p. 19–39.
Methods

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Parameters

Parameter	Description	Units
exp	Experiment identifier (noted in methodology)	Unitless
mussel_ID	Mussel identifier	Unitless
adhesive_age	Age of adhesive plaque (time after deposition)	Days
pH	pH treatment (either text or treatment target on NBS scale)	Unitless
oxygen	Oxygen treatment (either text or treatment target in mg L-1)	Unitless
shell_length	Length of major shell axis	cm
GI	Gonad Index	Unitless
CI	Condition Index	x10^-3 g cm^-3
failure_mode	Plaque failure mode. 1 = adhesive failure, 2 = peeling failure, 3 = tearing failure	Unitless
plaque_area	Adhesive plaque cross-sectional area	mm^2
max_force	Maximum force required to dislodge plaque	N
adhesion_strength	Maximum adhesion strength required to dislodge plaque	kPa
thread_day	Time mussels were in treatments before counting threads	Days
thread_number	Number of threads produced by a mussel	Unitless

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Project Information

Effects of Ocean Acidification on Coastal Organisms: An Ecomaterials Perspective (OA - Ecomaterials Perspective)

Website: http://depts.washington.edu/fhl/oael.html

Coverage: Friday Harbor, WA

Effects of Ocean Acidification on Coastal Organisms: An Ecomaterials Perspective

Effects of Ocean Acidification on Coastal Organisms: An Ecomaterials Perspective
This award will support researchers based at the University of Washington's Friday Harbor Laboratories. The overall focus of the project is to determine how ocean acidification affects the integrity of biomaterials and how these effects in turn alter interactions among members of marine communities. The research plan emphasizes an ecomaterial approach; a team of biomaterials and ecomechanics experts will apply their unique perspective to detail how different combinations of environmental conditions affect the structural integrity and ecological performance of organisms. The study targets a diversity of ecologically important taxa, including bivalves, snails, crustaceans, and seaweeds, thereby providing insight into the range of possible biological responses to future changes in climate conditions. The proposal will enhance our understanding of the ecological consequences of climate change, a significant societal problem.

Each of the study systems has broader impacts in fields beyond ecomechanics. Engineers are particularly interested in biomaterials and in each system there are materials with commercial potential. The project will integrate research and education by supporting doctoral student dissertation research, providing undergraduate research opportunities via three training programs at FHL, and summer internships for talented high school students, recruited from the FHL Science Outreach Program. The participation of underrepresented groups will be broadened by actively recruiting URM and female students. Results will be disseminated in a variety of forums, including peer-reviewed scientific publications, undergraduate and graduate course material, service learning activities in K-8 classrooms, demonstrations at FHL's annual Open House, and columns for a popular science magazine.

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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

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).

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
NSF Division of Ocean Sciences (NSF OCE)	OCE-1041213

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