# Utilizing Environmental DNA to Investigate the Effects of Hypoxia on Copepod Abundance

**Eleanor (Ella) Crotty** *(she/her)*
Reed College
Environmental Studies/Biology
Pacific Marine Environmental Laboratory
Zachary Gold[1], Shannon Brown[1,2], Sean M. McAllister[1,2], Jeannette Waddell[3], Kathryn R Hough[3], Simone R. Alin[1],
[1]NOAA PMEL | [2]UW CICOES | [3]NOAA OCNMS

PMEL
Pacific Marine Environmental Laboratory

WESTERN SOCIETY OF NATURALISTS
SAN DIEGO, CA 2025

REED COLLEGE

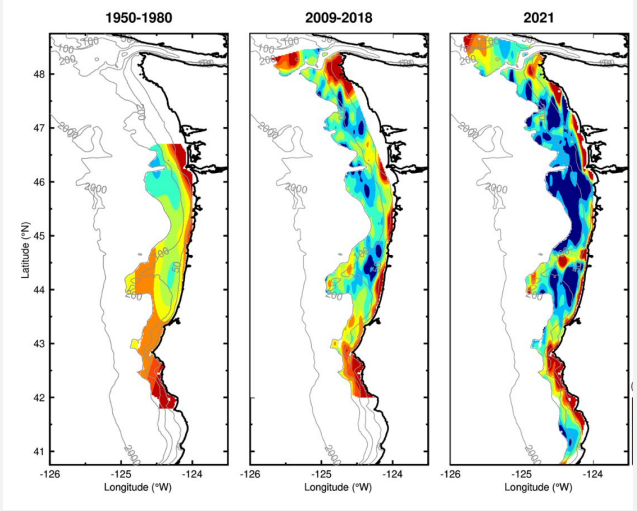
# Study Goals & Significance

- Do different copepod groups respond differently to hypoxia?
  - Northern vs. Southern
  - Small vs. large
  - Individual species differences
- Understanding the different impacts of hypoxia on OCNMS copepods will help sanctuary managers understand the potential impacts of climate change on copepod populations, and therefore on fish prey availability.

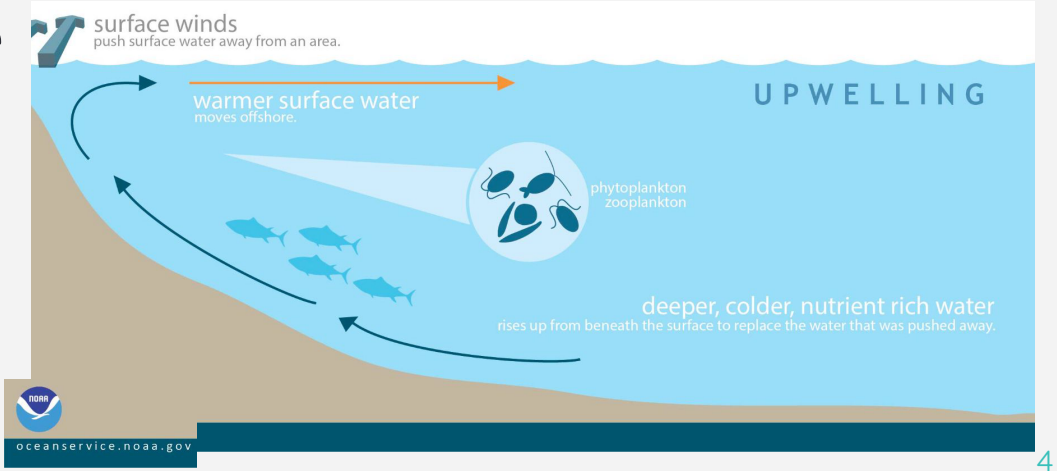
Goals
Intro
Methods
Results
Discussion
Future Directions

# Hypoxia

- Upwelling brings nutrient-rich, oxygen-poor water → more primary production → more oxygen consumption
- DO = Dissolved Oxygen




Barth et al. 2024





Goals
Intro
Methods
Results
Discussion
Future Directions

# Copepods


- Sentinel species
- Eaten by juvenile salmon

| DO (mg/L) | Sublethal Effects | Lethal Effects |
|---|---|---|
| 3.0+ | None | None known |
| 2.0-3.0 | Reduced egg production and feeding | None known |
| 1.0-2.0 | Behavioral avoidance | Nonlethal above 1.5 mg/L |
| 0.6-1.5 | Decreased respiration rate | % survival decreases throughout this range |
| <0.6 | None, they're all dead | Almost totally lethal (*A. tonsa*) |

He et al. 2021; N. H. Marcus et al. 2004; Stalder and N. H. Marcus 1997; Grodzins, Ruz, and Keister 2016)

Goals
Intro
Methods
Results
Discussion
Future Directions

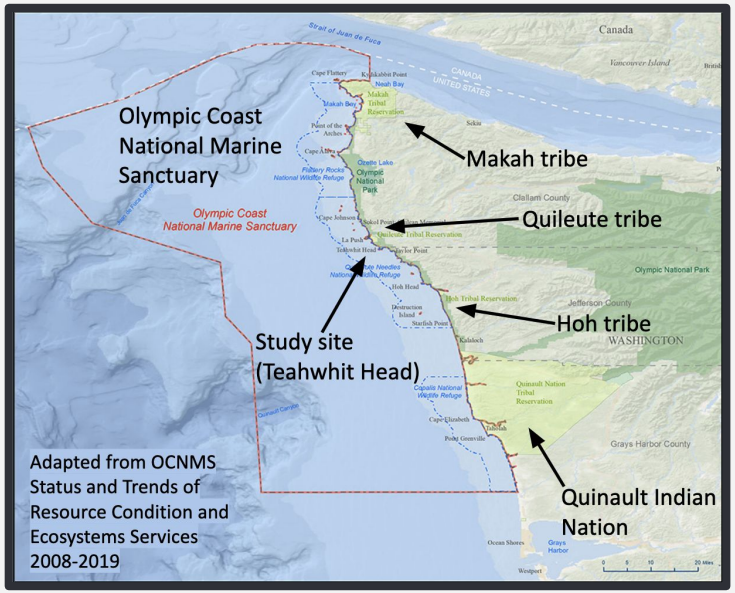
# Copepod Categorization

| Northern Copepods | Southern Copepods |
|---|---|
| Migrates from Arctic to OCNMS during **summer/upwelling** | Migrates from California and Oregon to OCNMS in **winter** |
| **Prefers cold-water** | **Prefers warm-water** |
| Lipid-rich = **better salmon food,** esp. *Calanus marshallae* and *Pseudocalanus mimus* | Less lipid-rich |

- This is based on **observed** migration patterns (first row) and **thermal range** near Newport, OR
- Not a taxonomic categorization

# Study Site

- Washington coast
- Olympic Coast National Marine Sanctuary (OCNMS)
- Teahwhit Head 42 meter site (TH042)



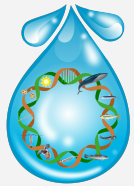
Goals
Intro
Methods
Results
Discussion
Future Directions

# Study Goals & Significance

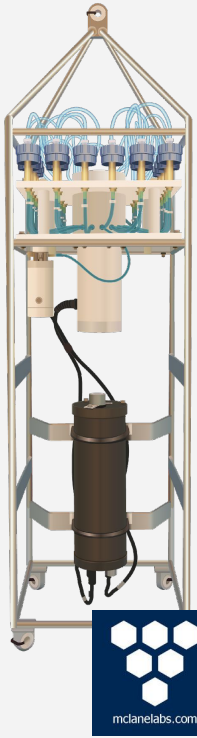
- Do different copepod groups respond differently to hypoxia?
  - Northern vs. Southern
  - Small vs. large
  - Individual species differences
- Understanding the different impacts of hypoxia on OCNMS copepods will help sanctuary managers understand the potential impacts of climate change on copepod populations, and therefore on fish prey availability.

Goals
Intro
Methods
Results
Discussion
Future Directions

# Methods



- **Environmental DNA (eDNA):** Genetic material shed into the environment
- Automated eDNA sampler → deployed at TH042 summer 2021-23
- Combined species + environmental data
- Identified proposed **vulnerable and resilient species during hypoxia (low oxygen)**, including **copepods**



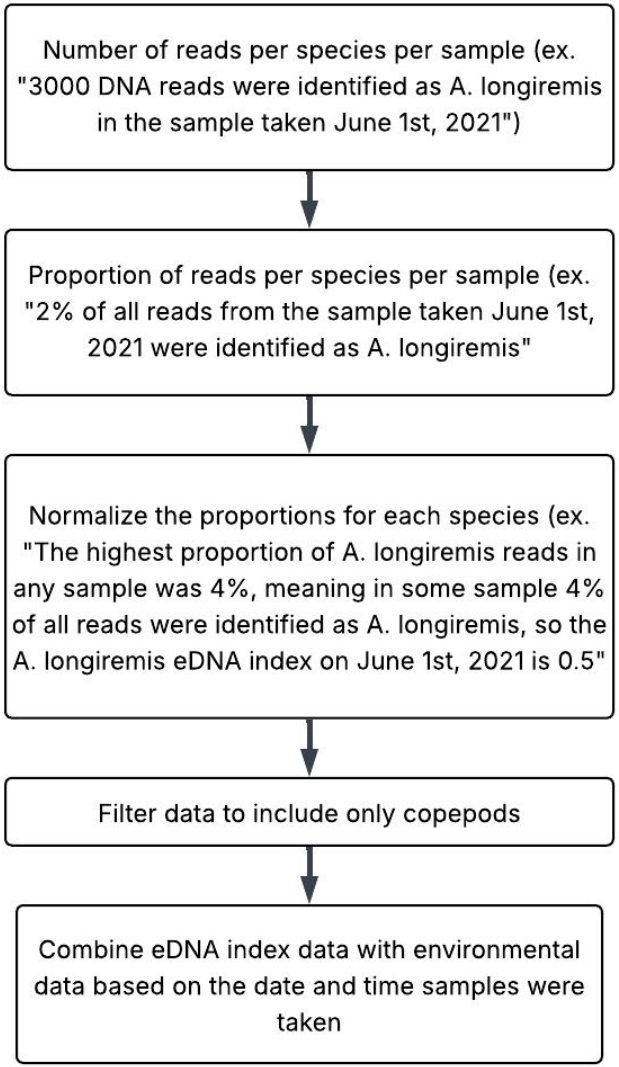

PPS Sampler

Intro
Goals
Methods
Results
Discussion
Future Directions

# Methods (cont.)

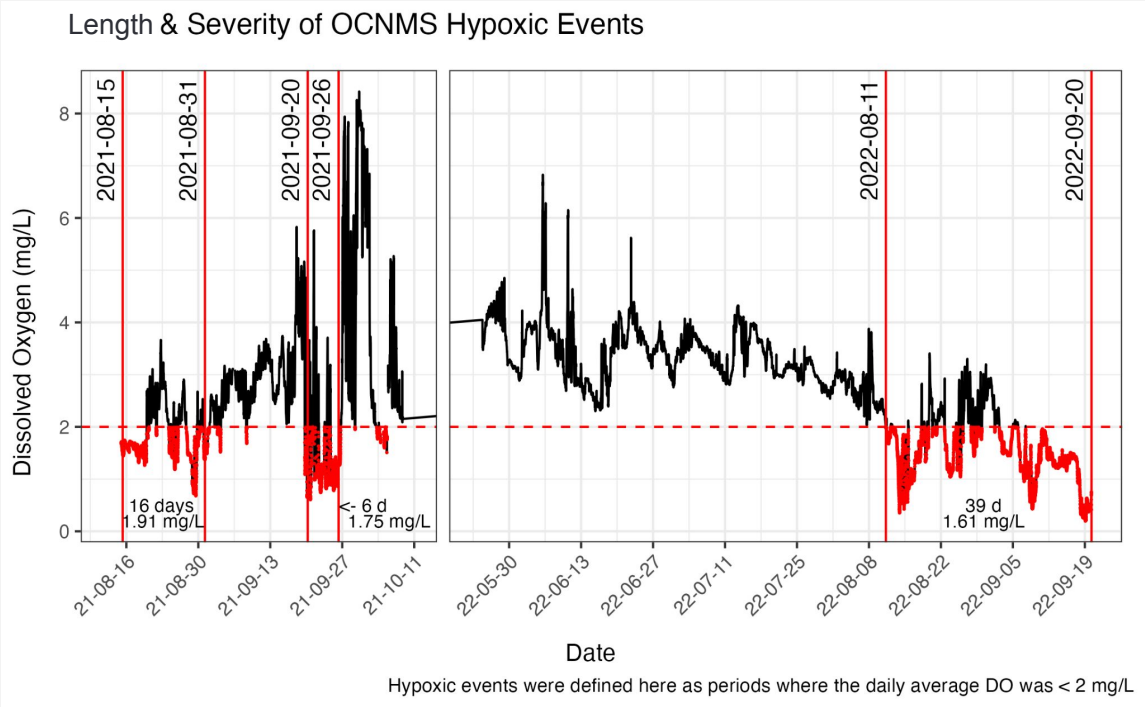
- **Metabarcoding**
- eDNA reads used to calculate **presence/absence** for each species
- Calculated **eDNA index** (normalize by sample, then species)
- Filtered for **copepod** species
- Combined with **environmental data**


Goals
Intro
Methods
Results
Discussion
Future Directions

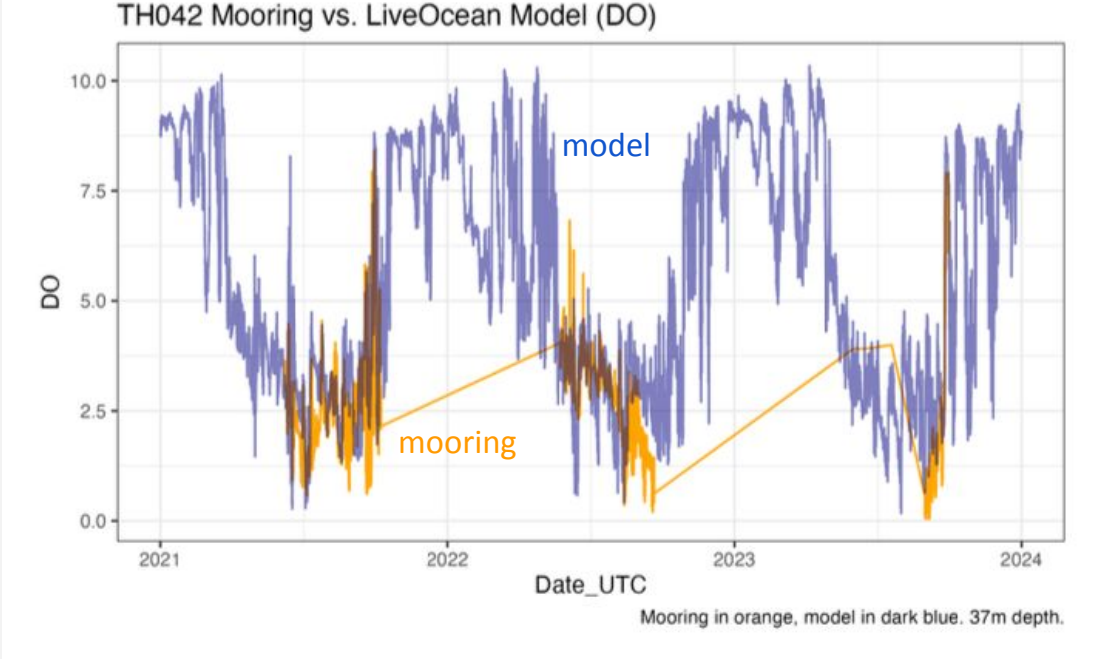
# Methods (cont.)

- 3 hypoxic events recorded 2021-22 at TH042



# Methods (cont.)

- Combined with LiveOcean **computer model data**, validated with mooring data



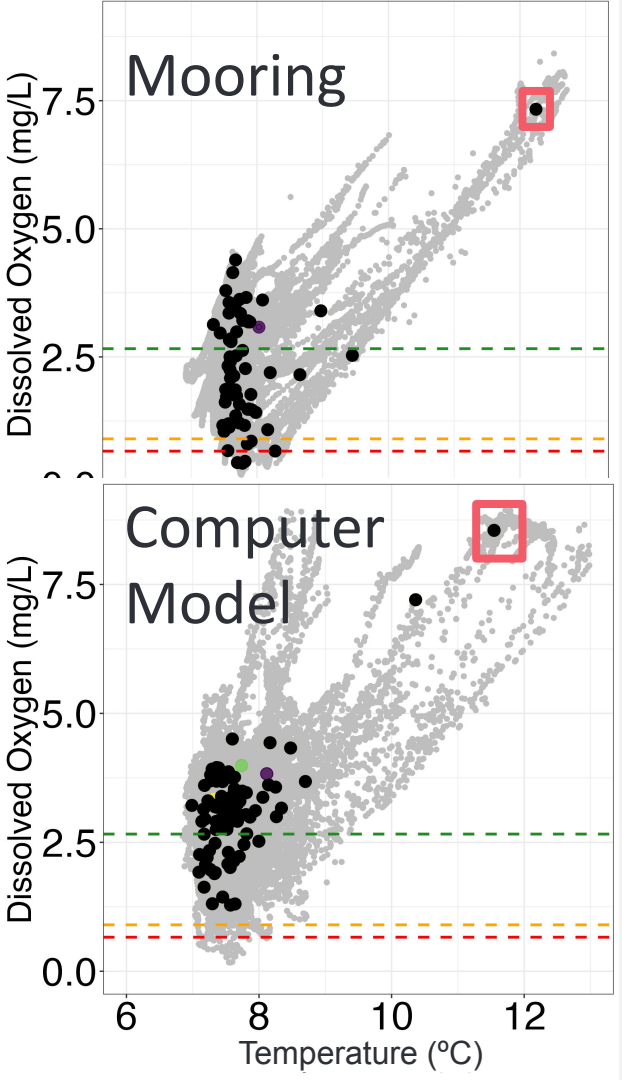


Goals
Intro
Methods
Results
Discussion
Future Directions

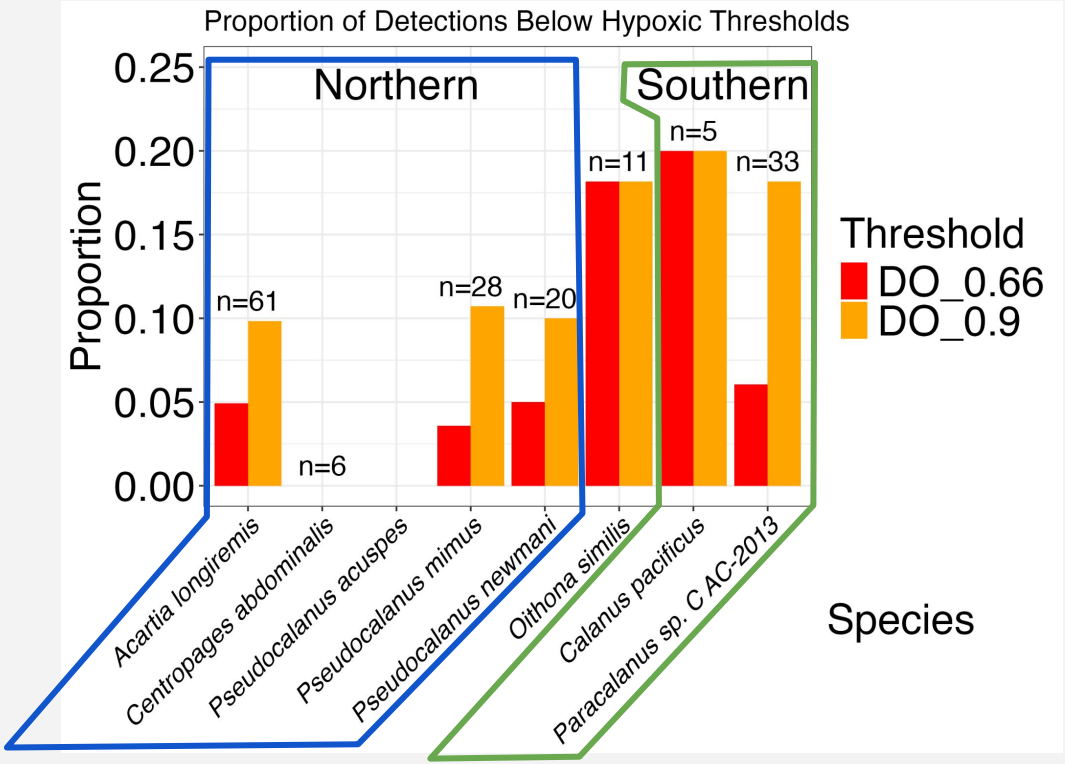
# Methods (cont.)

- Combined with LiveOcean **computer model data**, validated with mooring data
- Categorized species by average adult size from literature
- Used zero-inflated beta regression for eDNA index vs. DO (nonlinear pattern, many zeroes/species absences)

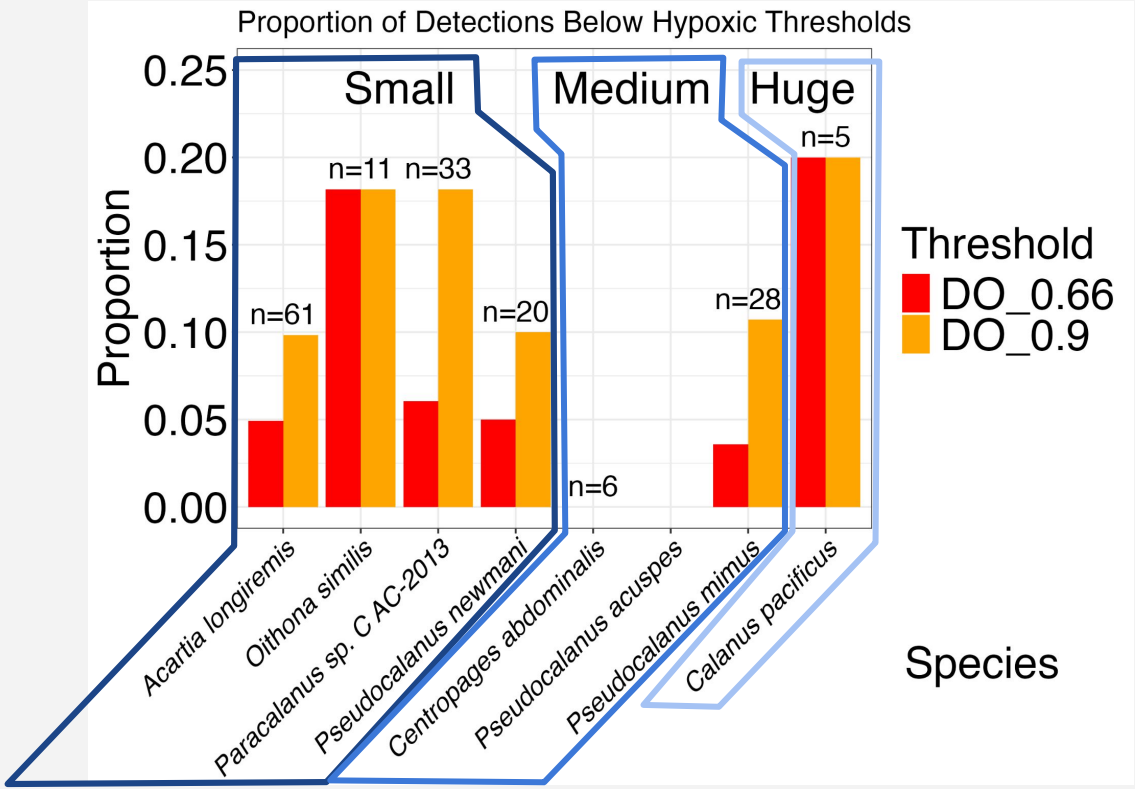

Goals
Intro
Methods
Results
Discussion
Future Directions

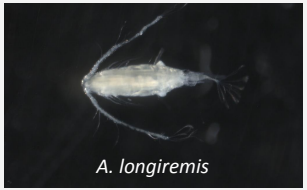
- **Mooring** data (2021-22) & presence/absence
- Observed higher tolerance in southern copepods
- Low sample size (see n = above each bar)



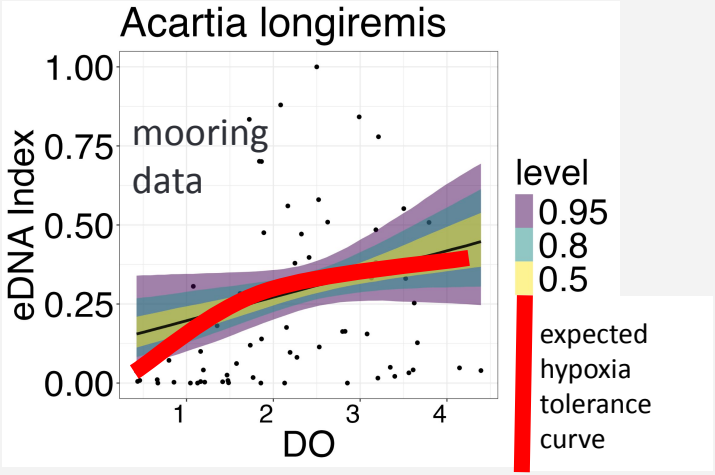
- **Mooring** data (2021-22) & presence/absence
- Size binned based on average adult size
- Observed higher tolerance in smaller copepods

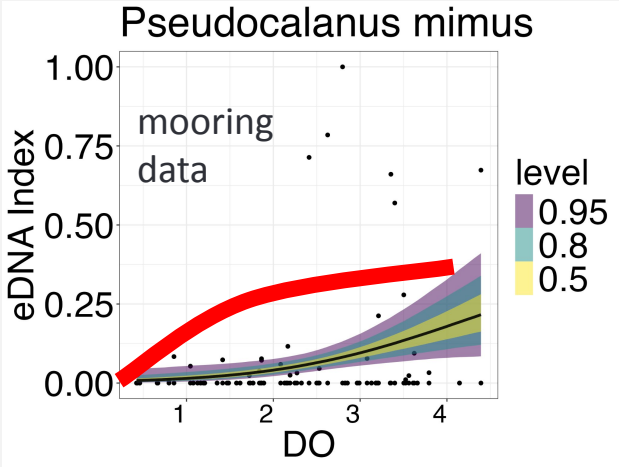


# Results




- Most zero-inflated beta regressions comparing eDNA index vs. DO were not significant
- *A. longiremis* and *P. mimus* (both northern) both had significant positive eDNA index vs. DO relationships (hypoxia intolerant)





*A. longiremis* image: Rob Young; *P. mimus* image: Copepedia/Moira Galbraith

Goals
Intro
Methods
Results
Discussion
Future Directions

# Results

| Species | Group | Detections | | Regression Results | |
|---|---|---|---|---|---|
| Species | Group | 2021-22 | 2021-23 | 2021-22 | 2021-23 |
| *Acartia longiremis* | Cold-water | 61 | 86 | Positive* | Neutral |
| *Pseudocalanus mimus* | Cold-water | 28 | 46 | Positive* | Negative* |
| *Pseudocalanus newmani* | Cold-water | 20 | 25 | Neutral | Neutral* |
| *Centropages abdominales* | Cold-water | 6 | 7 | Positive | Neutral |
| *Pseudocalanus acuspes* | Cold-water | 3 | 5 | Positive | Neutral |
| *Oithona similis* | Year-round | 11 | 11 | Negative* | Neutral |
| *Paracalanus* sp. C. AC-2013 | Warm-water | 33 | 33 | Neutr Negative* | e* |
| *Calanus pacificus* | Warm-water | 5 | 5 | Negative | Negative |

Table 2.1: Trends of zero-inflated beta regressions relating dissolved oxygen concentration to eDNA index for different species of copepods. Regressions were computed using the TH042 mooring data from 2021-22, and again using LiveOcean model data from 2021-23, so both are shown. I considered trends to be significant (marked with "*" in this table) if their 95% confidence interval showed an upward or downward trend and did not contain zero at some DO value. See Appendix B for plotted regression results. Cold-water copepods (blue) are northern copepods, warm-water copepods (orange) are southern.


Goals
Intro
Methods
Results
Discussion
Future Directions

# Discussion

Goals
Intro
Methods
Results
Discussion
Future Directions

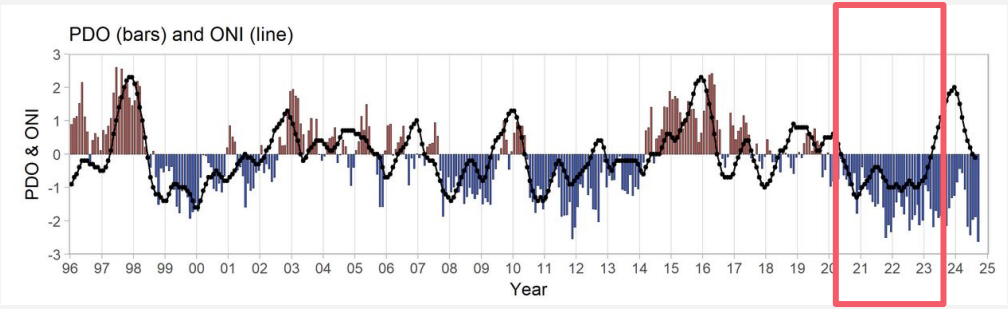
**Most copepods with >10 detections were less abundant at lower DO**

- Consistent with expectations
- Exception: *Oithona similis* (n = 11), year-round
- Consistent with literature

**Northern copepods appear to be more vulnerable to hypoxia than southern copepods**

# Discussion

- Climate change = worsening hypoxia, so concerning implications for zooplankton populations used as food by small fish
- Copepods could be moving closer to the surface
- **Caveats:** Study was primarily during negative El Niño years = relatively cold, and only during the summer upwelling season


Goals
Intro
Methods
Results
Discussion
Future Directions

# Future Directions

- Look for trophic effects
  - These samples were also metabarcoded with fish and phytoplankton primers
  - Copepods eat phytoplankton
  - Fish eat copepods
  - Correlated eDNA index of predator and prey substitutes?
- Hypoxia tolerance of other fish prey zooplankton - are any acceptable substitutes more tolerant?

Goals
Intro
Methods
Results
Discussion
Future Directions

# Thank you!


- Undergraduate thesis advisor & lab: Sam Fey
- NOAA PMEL Ocean Molecular Ecology Group:
  - Zachary Gold, Shannon Brown, Sean McAllister, Han Weinrich, and Samantha Setta
- OCNMS crew: Haley Wilson, Andrew Micks, Kathy Hough, and Hatcher Cox
- Simone Alin (PMEL) for feedback
- Evan Howard (PMEL), Jilian Xiong, Samantha Siedlecki (University of Connecticut) for physical oceanography help and LiveOcean model respectively
- Jeremy Coate & Josie Griffin for bioinformatics & statistics help at Reed
- My initial data combination work was funded by the NOAA Ernest F. Hollings Undergraduate Scholarship Program, this conference attendance was supported by the David T. Mason Student Opportunity Fund



eleanor.crotty@gmail.com

# Citations


Barth, John A., Stephen D. Pierce, Brendan R. Carter, et al. 2024. “Widespread and Increasing Near-Bottom Hypoxia in the Coastal Ocean off the United States Pacific Northwest.” Scientific Reports 14 (1): 3798. https://doi.org/10.1038/s41598-024-54476-0.

Brodeur, R.D., and W.G. Pearcy. 1990. “Trophic Relations of Juvenile Pacific Salmon off the Oregon and Washington Coast.” Fishery Bulletin 88 (4): 617–36. Scopus.

NOAA Fisheries. 2024. “Local Biological Indicators | NOAA Fisheries.” NOAA. West Coast. https://www.fisheries.noaa.gov/west-coast/science-data/local-biological-indicators.

Office of National Marine Sanctuaries. 2022. Olympic Coast National Marine Sanctuary Condition Report: 2008–2019. U.S. Department of Commerce, National Oceanic and Atmospheric Administration, Office of National Marine Sanctuaries, Silver Spring, MD. 453 pp.

“Olympic Coast National Marine Sanctuary.” n.d. Accessed April 16, 2025. https://olympiccoast.noaa.gov/.

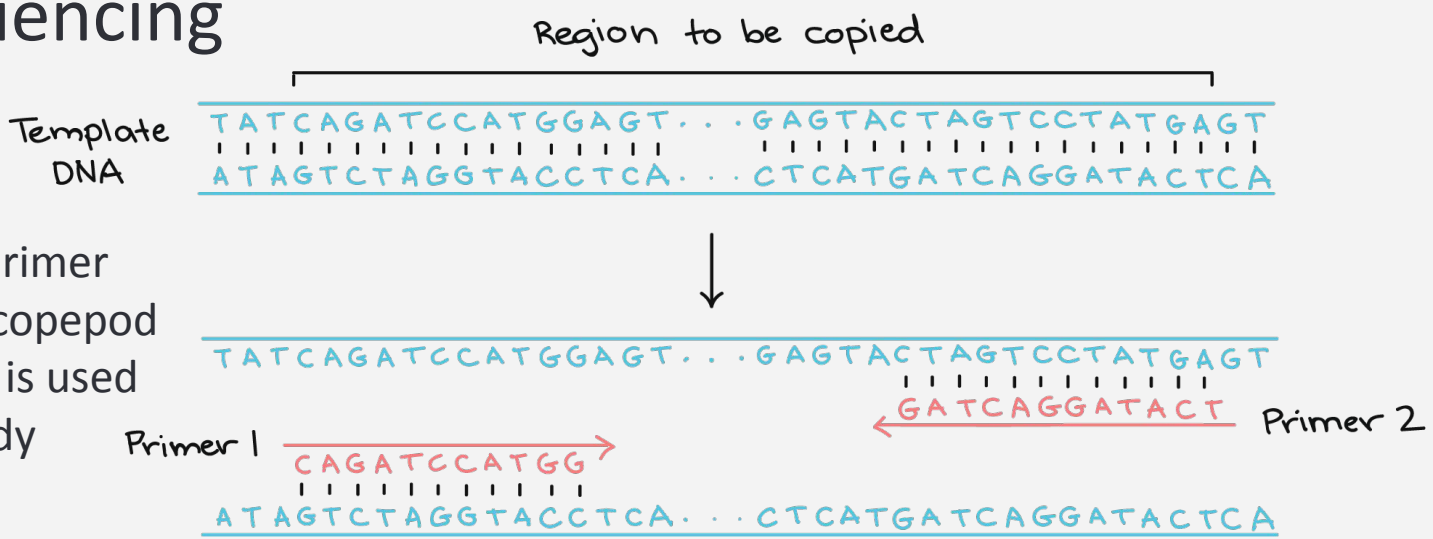
Razouls C., Desreumaux N., Kouwenberg J. and de Bovée F., 2005-2025. - Biodiversity of Marine Planktonic Copepods (morphology, geographical distribution and biological data). Sorbonne University, CNRS. Available at http://copepodes.obs-banyuls.fr/en [Accessed March 08, 2025]

# Methods (supplemental)

- **Primer:** A piece of DNA that starts DNA synthesis, used to amplify DNA before sequencing


Intro
Goals
Methods
Results
Discussion
Future Directions


The **CO1** primer amplifies copepod DNA, so it is used in this study



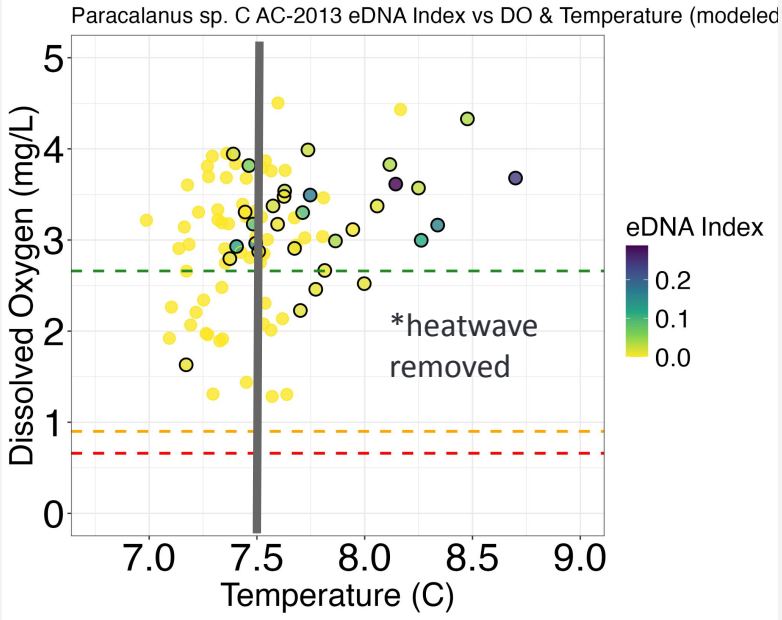

khanacademy.com

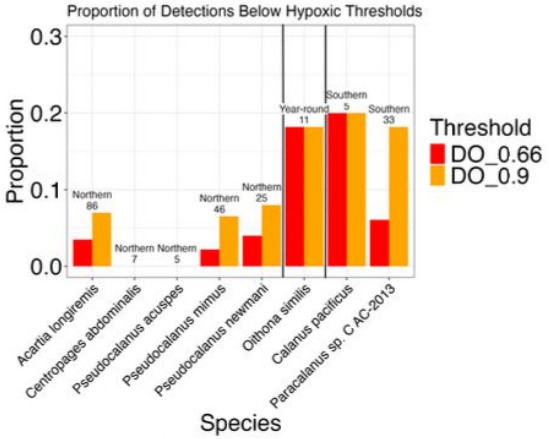
- Northern copepods primarily detected below 8 °C (except *Pseudocalanus mimus*)
- Southern copepods (ex. *Paracalanus*, shown below) primarily above 7.5 °C



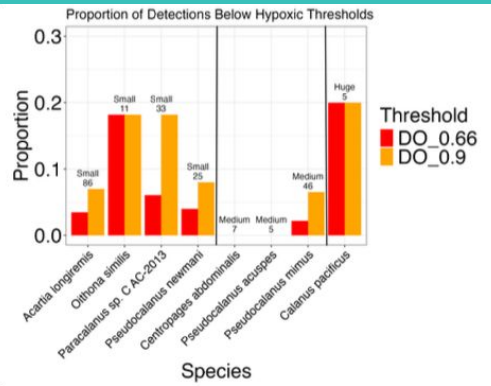
**Yellow** dots = absent when water sampled in those conditions

**Black outline** dots = present when water sampled in those conditions






a.




b.

Figure 2.2: A comparison of different copepod groups and their rates of occurence below certain hypoxic thresholds. Red bars indicate the proportion of detections of that species that were in conditions below 0.66 mg/L of oxygen, and orange bars indicate the proportion of detections of that species that were in conditions below 0.9 mg/L of oxygen. Numbers above bars are the total number of detections of that species. In plot (a) copepods are labeled by seasonal groupings. In plot (b), copepods are labeled by size category. Species with a maximum size of less than 1.5 mm are Small, maximum size 1.5-2.5 mm is Medium, maximum size 2.5-4 mm is Large, and maximum size >4 mm is Huge (*WoRMS - World Register of Marine Species - Copepoda* 2024)