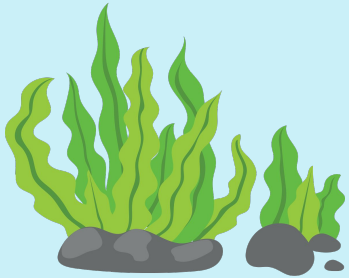


Hypoxia effects on *Hemigrapsus oregonensis*

GG Foster, Will Mixon, Maddy
Glasser, Erik Bengtson



Introduction

Big Question: How does hypoxia affect crabs?

Model Species: *Hemigrapsus oregonensis*

Key points:

- Crab fisheries in Pacific are in trouble
- Hypoxia increasing
- Does hypoxia in deep vs shallow water make a difference?



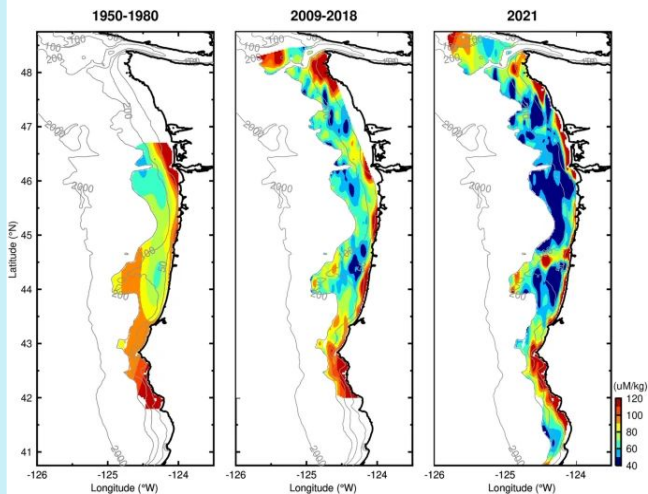
G.Fletcher photo

Background

Upwelling

- Wind changes are occurring due to evolving land-sea temperature differences
- Spring-summer transportation of cold, hypoxic water to shallow coastal habitats

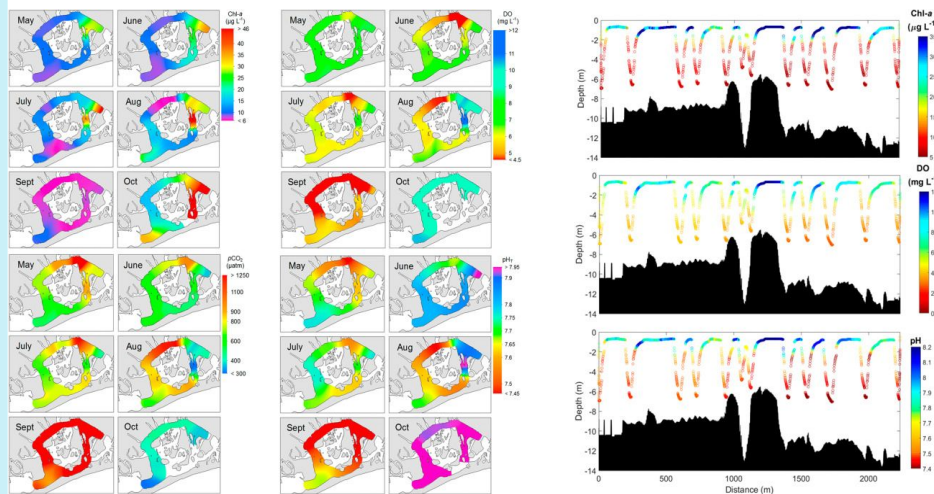
Barth et al., 2024



Algal Blooms

- Algal blooms form as temperatures rise during the spring
- Raised nutrient concentrations due to runoff fuel overwhelming growth
- Respiration by decomposers reduces oxygen content at depth

Wallace and Gobler, 2021



Background

What can be tested

- **Effects on respiration**
 - **Lactate** - shift to anaerobic metabolism causes increased lactate production in the hemolymph
 - **Respirometry with Resazurin** - blue dye that when exposed to metabolically active cells reduces it to resorufin (becomes pink) by consuming oxygen; faster the change in color → more oxygen consumption, would expect a slower consumption
- **Other**
 - **Righting Test** - Can indicate the energetic ability of the individual (do they have the energy to flip themselves back over)
 - **Gill Tissue Wasting** - Atrophic gill filaments or deteriorated gill filaments is a common result of prolonged hypoxia exposure

Threats to the commercial crab industry

- **Sudden increase in hypoxic conditions leading to crab declines**
 - Instances of severe hypoxic conditions from harmful algal blooms has significantly increased in the last few decades and impacts commercial fisheries abilities to catch crabs / enough (Barth et al, 2024)
- **Poor management to counteract impacts**
 - Many fisheries do not have the management in place to combat these declines (Holland & Leonard, 2020)
- **Overall revenue decline**
 - Loss of crab abundance leads to overall revenue loss, impacting companies and individuals who rely on crabbing (Holland & Leonard, 2020)

How will hypoxia affect the physiology of *Hemigrapsus oregonensis*?

Null: Hypoxia will not have an effect on the physiology of Hairy shore crabs.

Alternative: Hypoxia will have an effect on the physiology of Hairy shore crabs.

Expected outcomes:

Observational

- Crabs with access to air will leave the water to escape the hypoxic conditions and resulting stress.
- Crabs experiencing hypoxia will be lethargic.
- They may have longer righting times or not be able to right themselves.
- Crabs may die from long-term exposure to hypoxic water.
- Gill tissues in crabs from both tanks will deteriorate but may be worse in deepwater crabs.

Experimental

- Crabs will switch to anaerobic metabolism and lactate in the hemolymph will increase. Hypoglycemia may occur.
- Resazurin levels could show that respiration is decreasing in hypoxic crabs due to stress.



Treatment Design

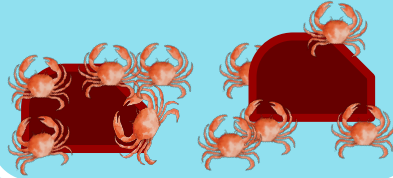
Intertidal

Start: 7-8 ppm O₂
No Airstone
Access to Surface



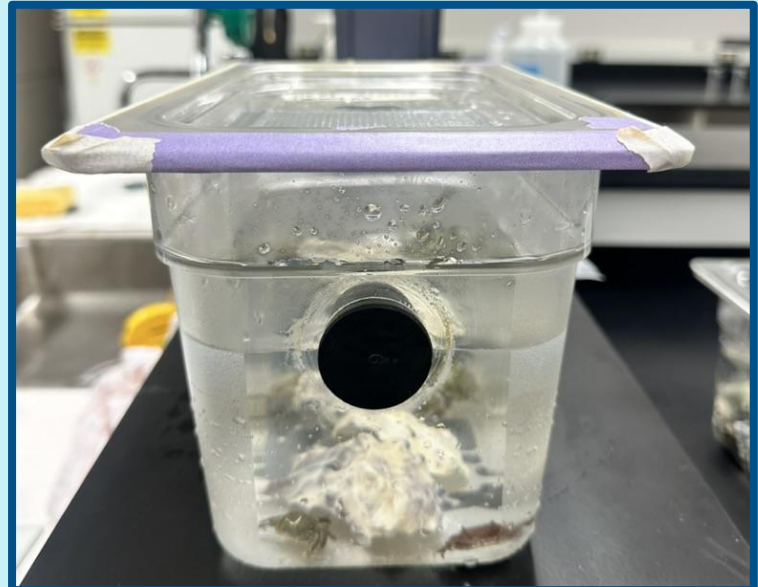
Deepwater

Start: 7-8 ppm O₂
No Airstone



Two Treatments and a Control

- Control tank
 - Hides, airstone
- Intertidal
 - Access to surface with mesh square
- Deepwater
 - No access to surface



Experimental Design

After 1 week:

Non lethal tests and marking of 50% of crabs

- Righting
- Respiration (resazurin)

After 2 weeks

Non lethal tests on 100% of crabs

- Righting
- Respiration (resazurin)

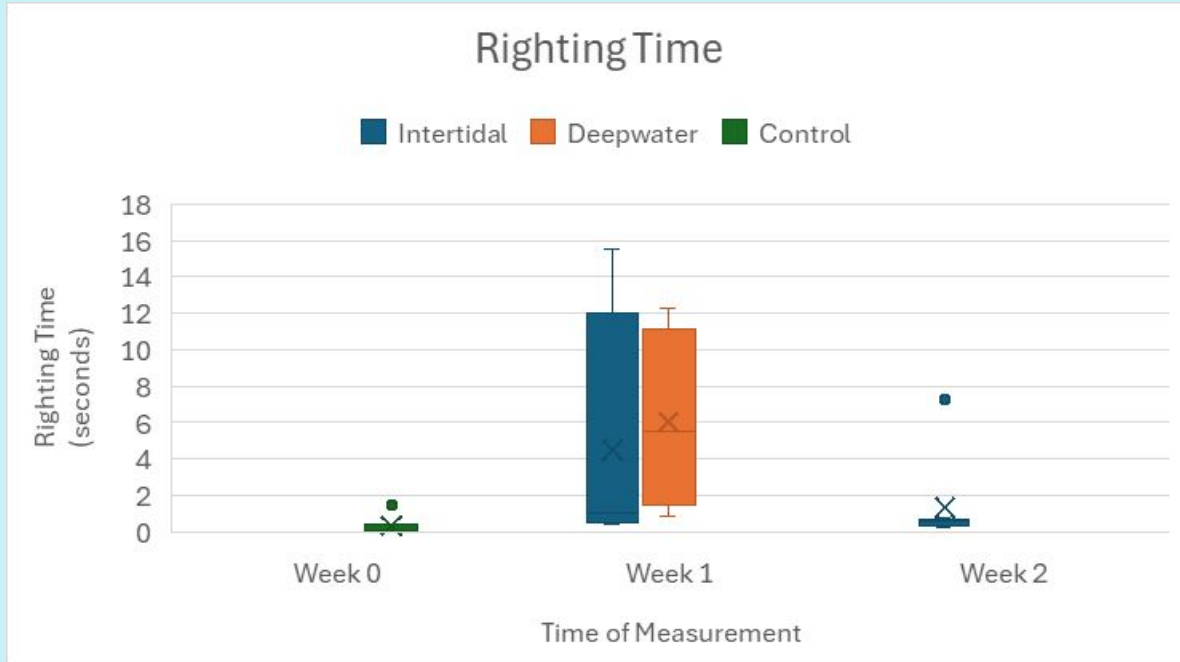
Lethal tests on 100% of crabs

- Hemolymph extraction for lactate analysis
- Dissection for gill tissue examination

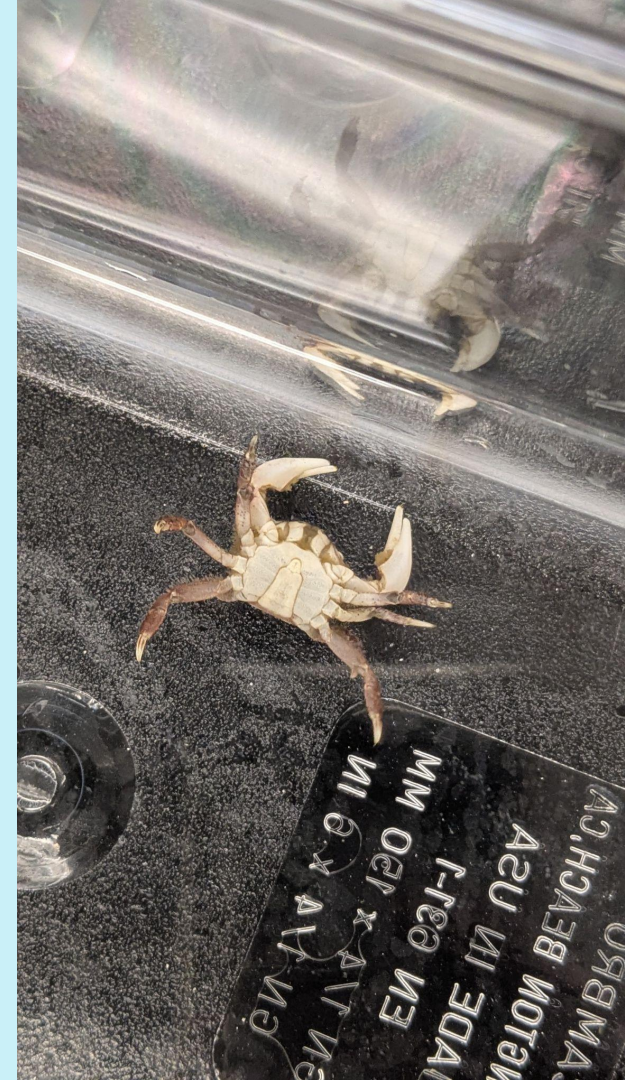


Nail polish marked crabs

Results - Righting Time

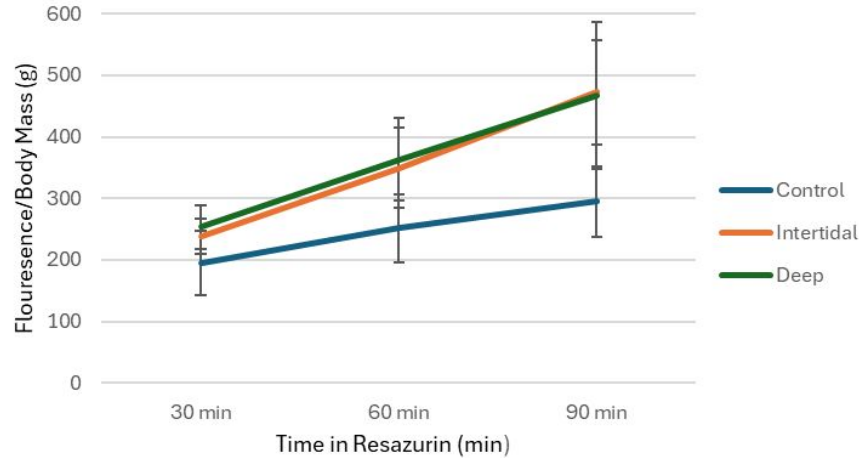


No significant difference between treatments. Mortality meant no data from the deepwater group after Week 2.

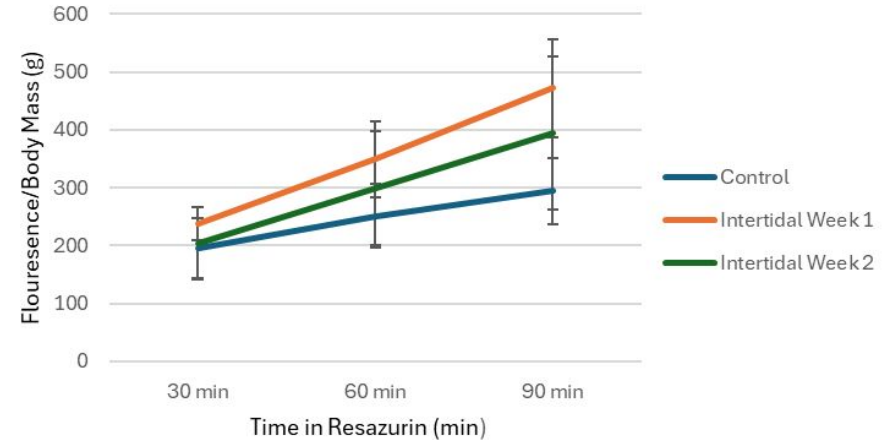


Results - Resazurin

Deep vs Intertidal Hypoxia



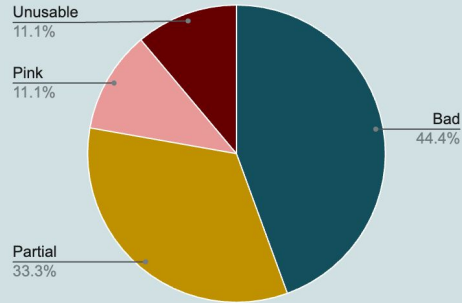
Intertidal Hypoxia



- Hypoxia treatments resulted in higher rates of respiration than control.
- Week 2 intertidal resulted in lower respiration rates than Week 1.

Results - Gill Tissue

Intertidal



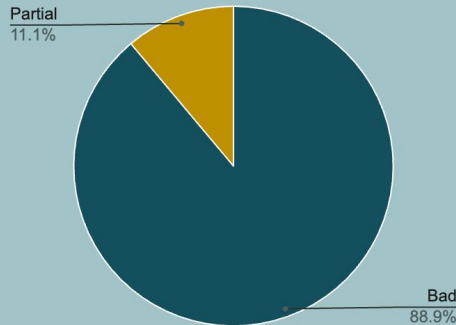
Gill Tissue Key

Good	Yellow, orange-ish color
Bad	Grey, dark, atrophy
Partial	Grey with some yellow
Pink	Pink-ish possibly from resazurin
Unusable	Unable to dissect (crushed)

Deepwater



Deepwater

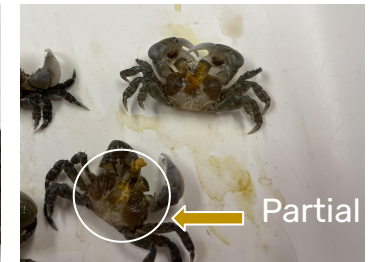


Control

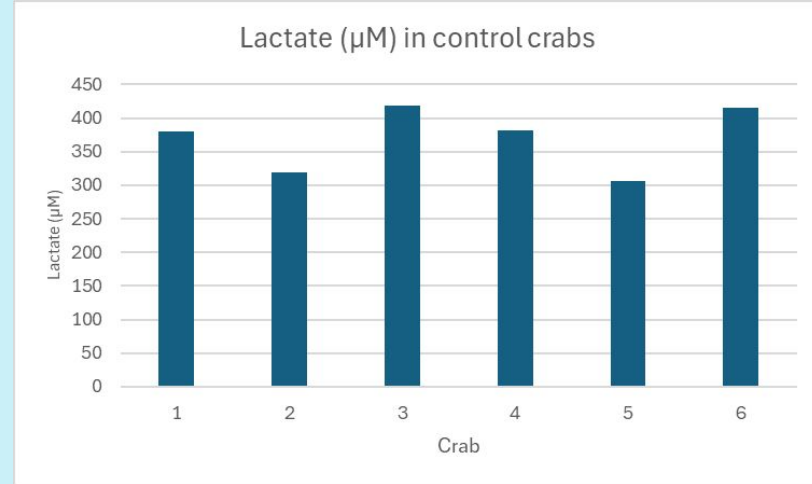
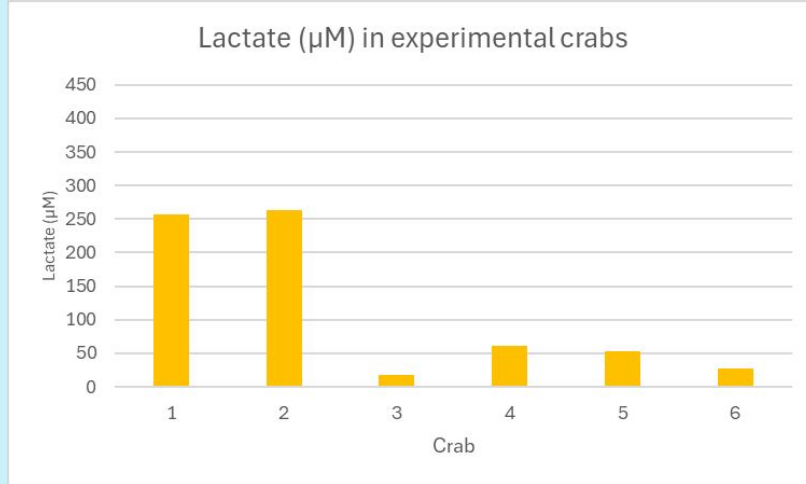


*Control - 100% good (2/2)

Intertidal



Results - Hemolymph



- Some experimental crabs had slightly lower lactate than control crabs, other had extremely low levels of lactate
- We expected experimental crabs to have higher levels of lactate. Lack of oxygen would make it hard to continue metabolizing normally so they might switch to anaerobic metabolism, producing more lactate. However, the opposite happened.
- “Lactate paradox”: after crabs acclimate to the hypoxic conditions their lactate levels drop

Interpretation and Conclusions

- Righting time was not a strong determining factor of physiological stress
- Respiration rates may be higher than control because of increased heart rate in crabs to help deliver more oxygen to the body (McGaw, 2008)
- “Lactate paradox” contradicting to expected lactate levels
- Gill tissue deterioration due to oxidative stress and cell decay

Implications:

- Crabs have a higher chance of survival if they have access to air
- Scientists and commercial crabbers can use this data to predict outcomes in crab populations and adapt to hypoxic events if possible



Future Work

- **Increased Respiration**
 - More tests to determine the cause of increased respiration in the hypoxic treatments
 - Increased heart rate?
 - Breathing harder to increase flow of water?
- **Decreased Lactate**
 - Looking more closely at the “lactate paradox”
 - Crab study
 - Run more tests to gather more data to see if this is truly what is occurring
- **Possible Changes to Improve Design**
 - Addressing what could've been better executed and see if making those changes impact the results
 - Method of sealing the deepwater tank
 - Marking the crabs with nail polish
 - Finding a different method of tagging to test the difference between week 1 and week 2 crabs (tested once vs tested twice)
 - Try to find the time period where deepwater crabs are still alive to test their hemolymph to compare to intertidal

Acknowledgements

Thank you to:

- José Guzmán for educating us about marine physiology.
- Zach Bengtsson for helping us design our experiment and for running all of our assays.
- Andy Nutzhorn for helping us design and run our experiment.