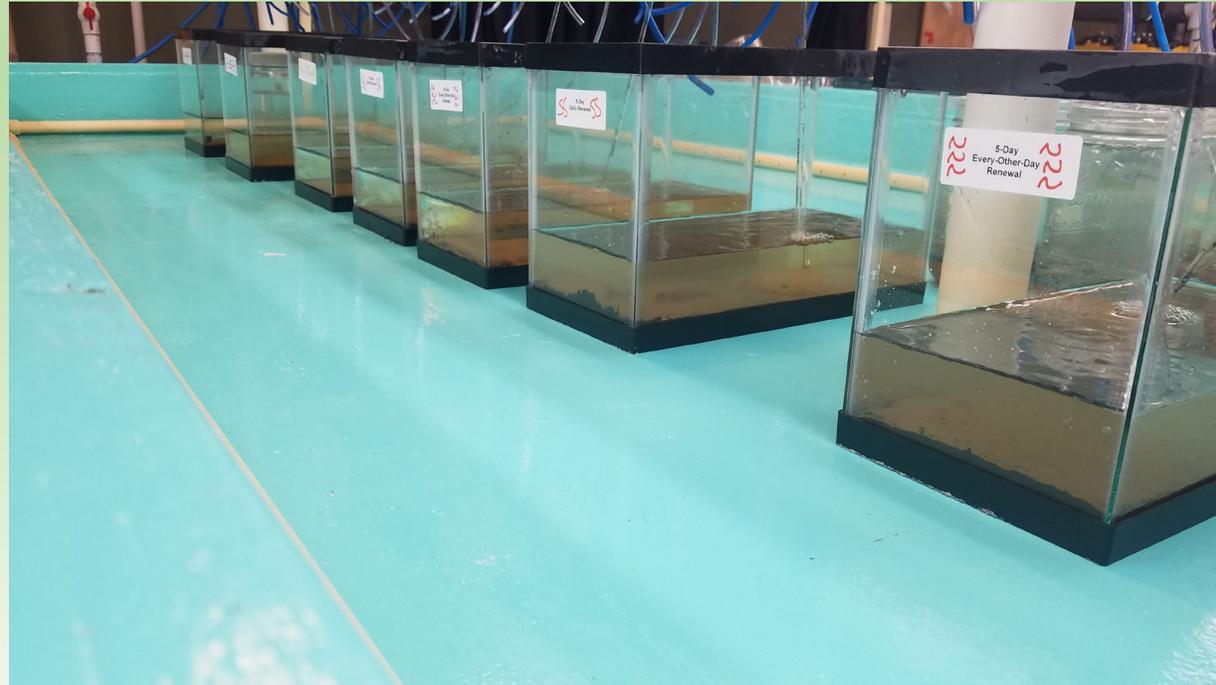


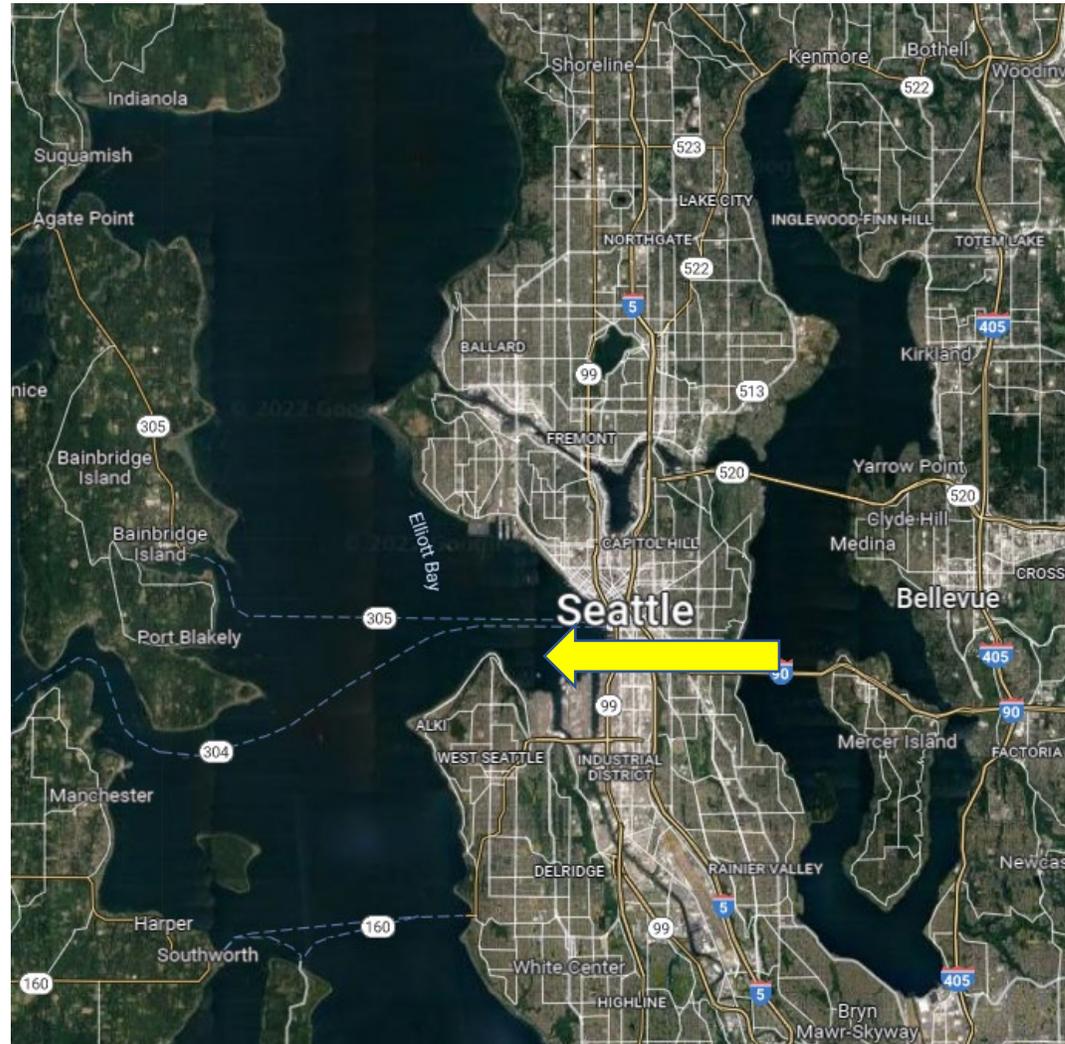
# Bioassay Saltwater Acclimation Study



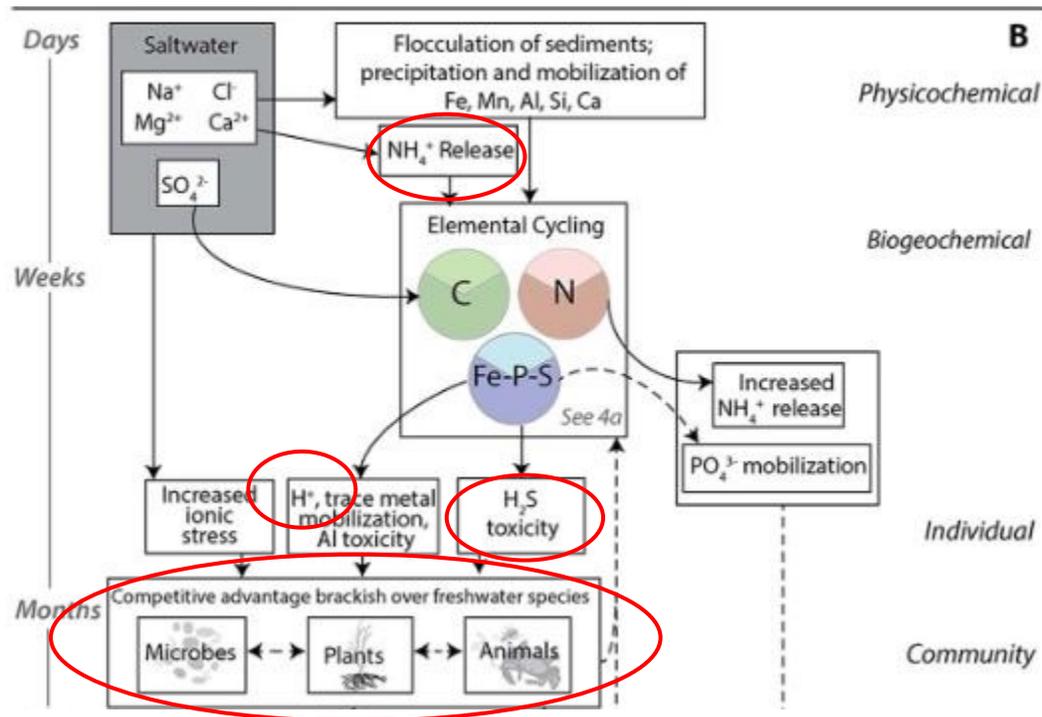
Mary Ann Rempel-Hester, Samantha Heinicke, Danielle Mulligan, EcoAnalysts Inc.  
Kelsey Van Der Elst, Seattle Army Corps DMMP

WEDA Pacific Chapter Meeting, October 27<sup>th</sup>, 2022

# Background: Disposal of freshwater sediment at saltwater disposal sites

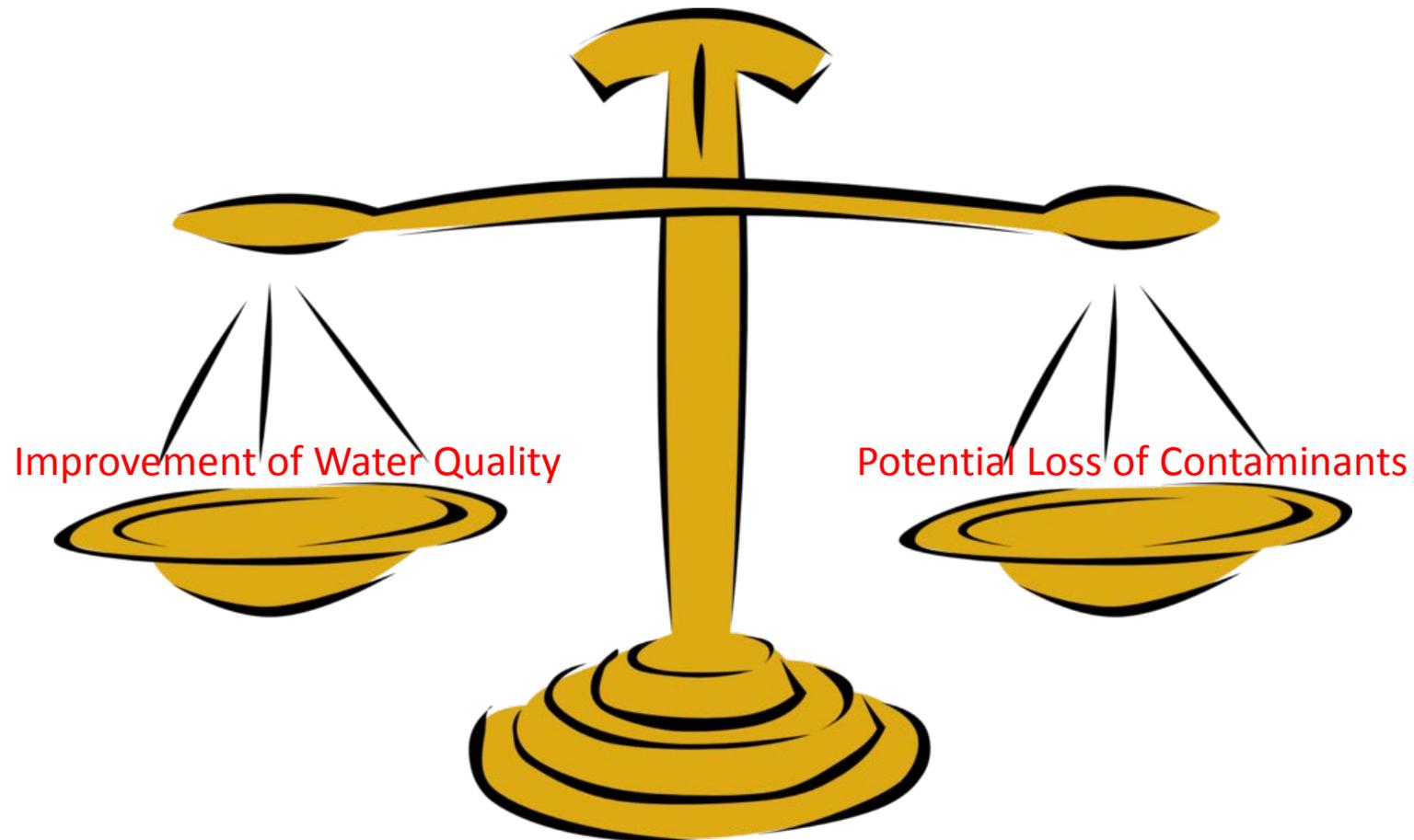


# Problem – Biogeochemical effects of saltwater intrusion into freshwater sediments



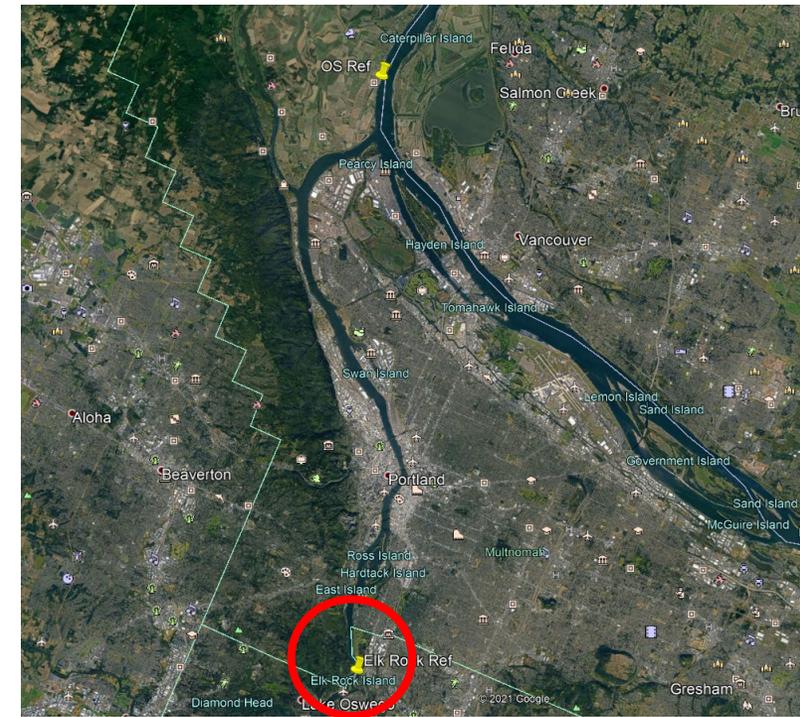
- Increase in ammonia
  - Decrease in pH
  - Potential modulation of sulfides
  - All add up to an increase in toxicity driven by biogeochemical effects – outside of any potential effects caused by contaminants
- 
- Good news – effects are transitory until new equilibrium in saltwater environment is established

Solution: Acclimation of freshwater sediment to saltwater conditions prior to introduction of bioassay organisms



# Study 1 Design

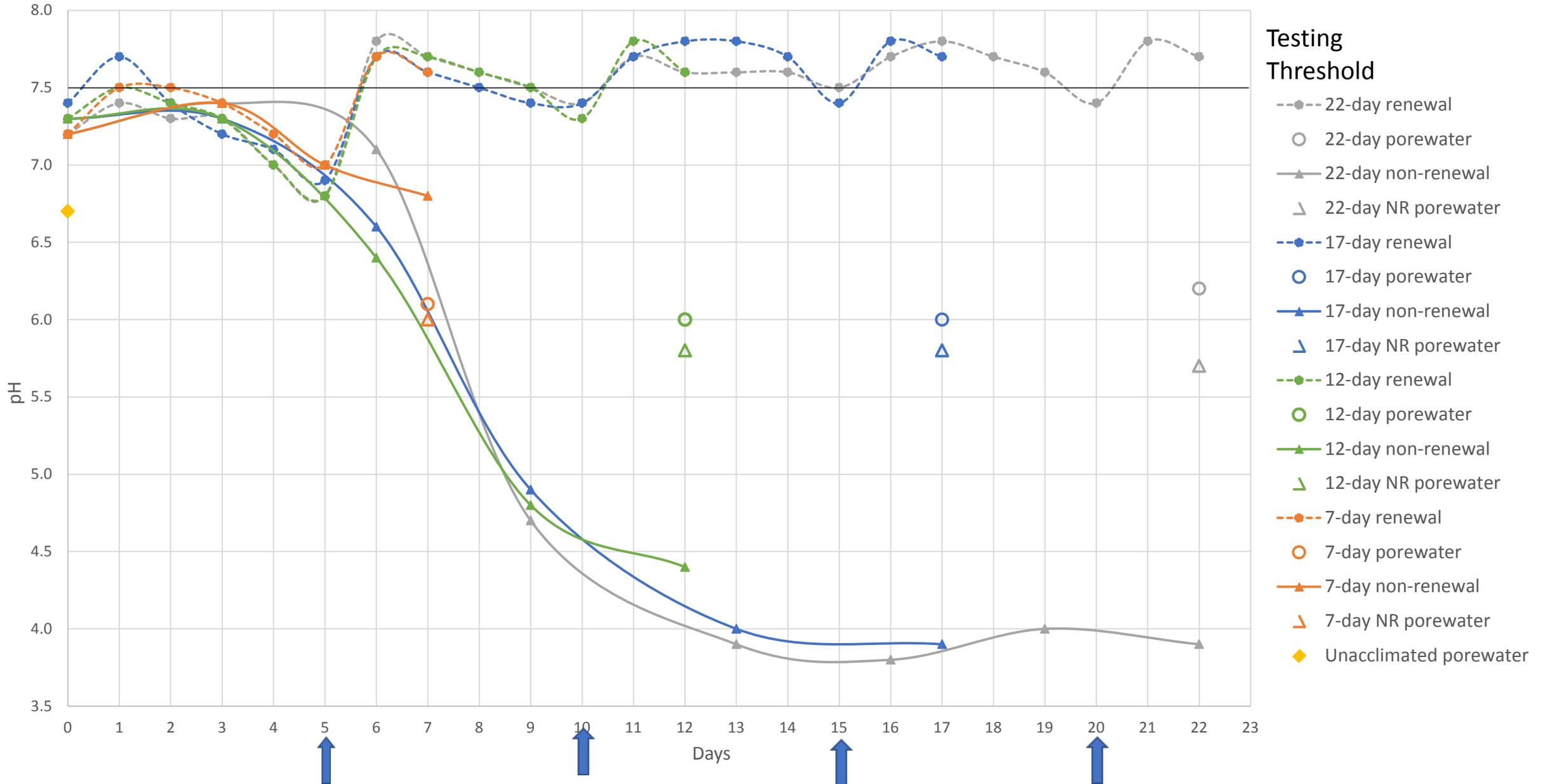
- Sediment:
  - Elk Rock Island reference sediment from Portland District
  - 10-day chironomid and 28-day Hyalella both >90% survival
  - Hyalella growth met reference acceptability criterion at 0.75 mg/ind
- Study design:
  - 22-day, 17-day, 12-day, 7-day and unacclimated
  - Renewal every 5 days (-----) and non-renewal (———)
  - Sand-filtered water
  - Larval test at end of acclimation (n=9)



Study 1 Acclimation Chambers



# pH Results





# Larval Test Water Quality: UIA

	Unacclimated
Day 0	0.001
Day 2	0.003



	Control
Day 0	0.000
Day 2	0.000

Purge trigger = 0.04 mg/L

## 7-day

	Renewal	Non-Renewal
Day 0	0.000	0.001
Day 2	0.002	0.003

## 12-day

	Renewal	Non-Renewal
Day 0	0.000	0.001
Day 2	0.001	0.002

## 17-day

	Renewal	Non-Renewal
Day 0	0.000	0.001
Day 2	0.001	0.001

## 22-day

	Renewal	Non-Renewal
Day 0	0.000	0.001
Day 2	0.001	0.002

# Larval Test Water Quality: H<sub>2</sub>S

	Unacclimated
Day 0	ND
Day 2	0.673

## 7-day

	Renewal	Non-Renewal
Day 0	1.533	1.141
Day 2	ND	ND

## 17-day

	Renewal	Non-Renewal
Day 0	ND	1.988
Day 2	ND	0.736

	Control
Day 0	0.000
Day 2	ND

Purge trigger = 2.5 mg/L

## 12-day

	Renewal	Non-Renewal
Day 0	0.000	1.911
Day 2	ND	ND

## 22-day

	Renewal	Non-Renewal
Day 0	0.952	3.681
Day 2	ND	ND

# Larval Test Water Quality: pH

	Unacclimated
Day 0	6.8
Day 2	7.1

	Control
Day 0	7.9
Day 2	8.0

Recommended range: >7.5

## 7-day

	Renewal	Non-Renewal
Day 0	7.0	7.0
Day 2	7.3	7.4

## 12-day

	Renewal	Non-Renewal
Day 0	7.1	7.0
Day 2	7.3	7.3

## 17-day

	Renewal	Non-Renewal
Day 0	7.2	7.1
Day 2	7.4	7.3

## 22-day

	Renewal	Non-Renewal
Day 0	7.3	7.2
Day 2	7.5	7.4

# Larval Test Results

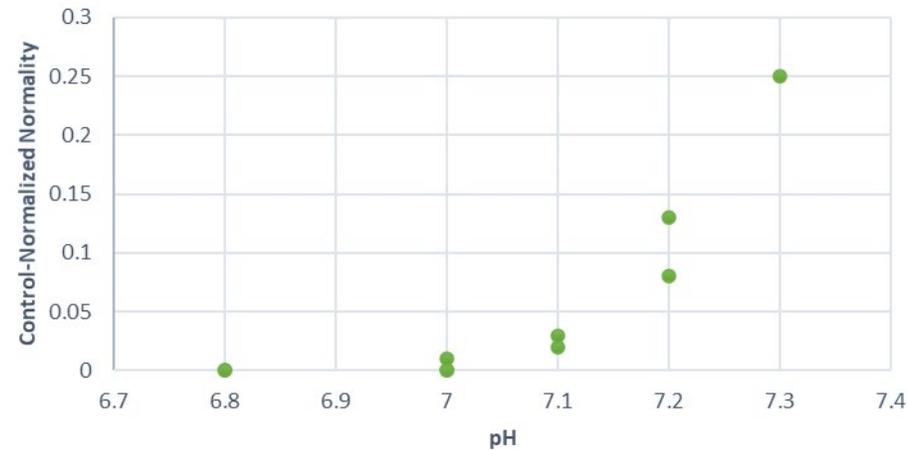
### Acclimation Duration vs. Normality



### Acclimation Duration vs. Survival



### Min pH vs. Normality



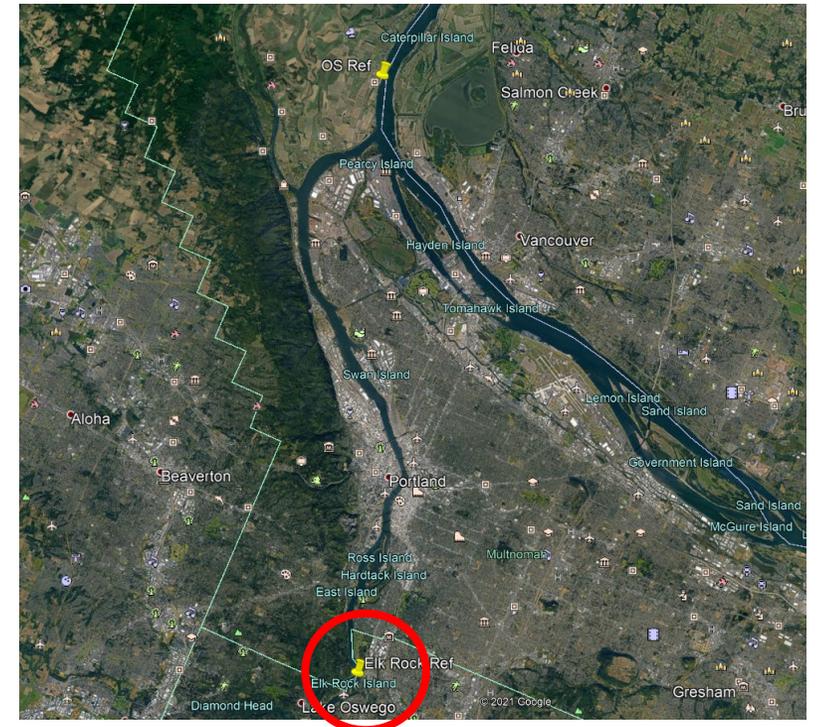
Normal Larva



Abnormal Larvae

# Study 2 Design

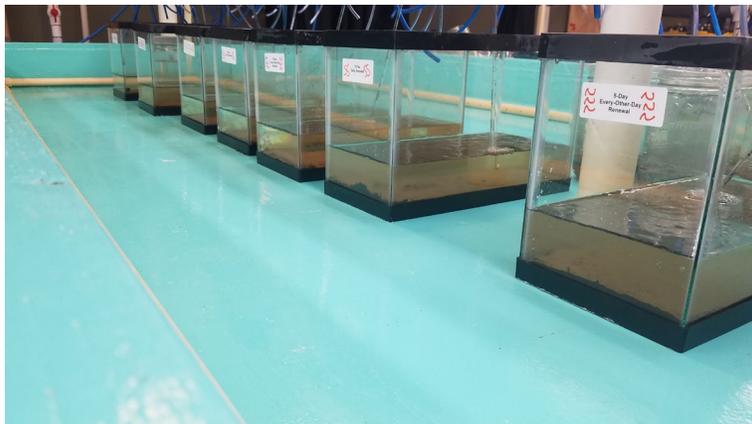
- Sediment:
  - Same sediment as Study 1
- Study design:
  - 21-day, 16-day, 11-day and unacclimated
  - Daily Renewal (DR, ———) and Every-Other-Day Renewal (EODR, - - - - -)
  - Additional 18-day acclimation with renewal every 5 days (E5DR, - - -) for comparison to study 1
  - Sand-filtered water, thin layer method
  - Larval test at end of acclimation



# Thin Layer Method

- Follows Ferretti et al. 2000 method for ammonia purging. Sediment to depth of 7-10 mm, with overlying water at a ratio of 3.2x water:sediment (1.9 L water:0.6 L sediment). Ferretti called for twice daily renewals; we modified to once daily or once every other day. Temp  $20 \pm 2^\circ\text{C}$ . Note: Ferretti measured chemical loss in addition to ammonia removal in study.
- Initial acclimation study had sediment to depth of 200 mm (2 cm) with 4x water:sediment ratio and either no renewal or renewal every five days. Temp  $16 \pm 2^\circ\text{C}$ .

Study 2  
Acclimation  
Chambers

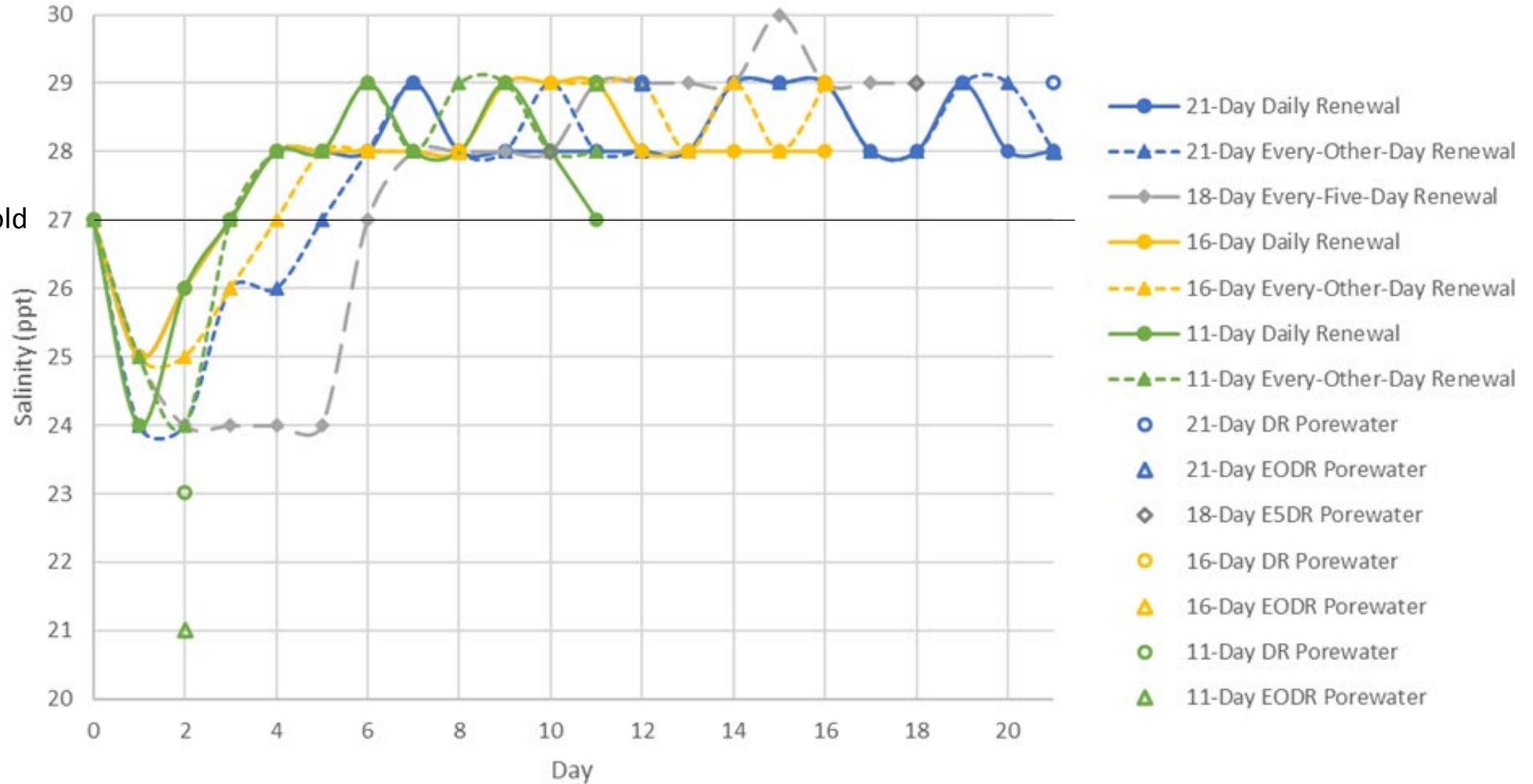


Study 1 Acclimation Chambers

# Subsampling for Porewater Measurements and Mock Larval Tests

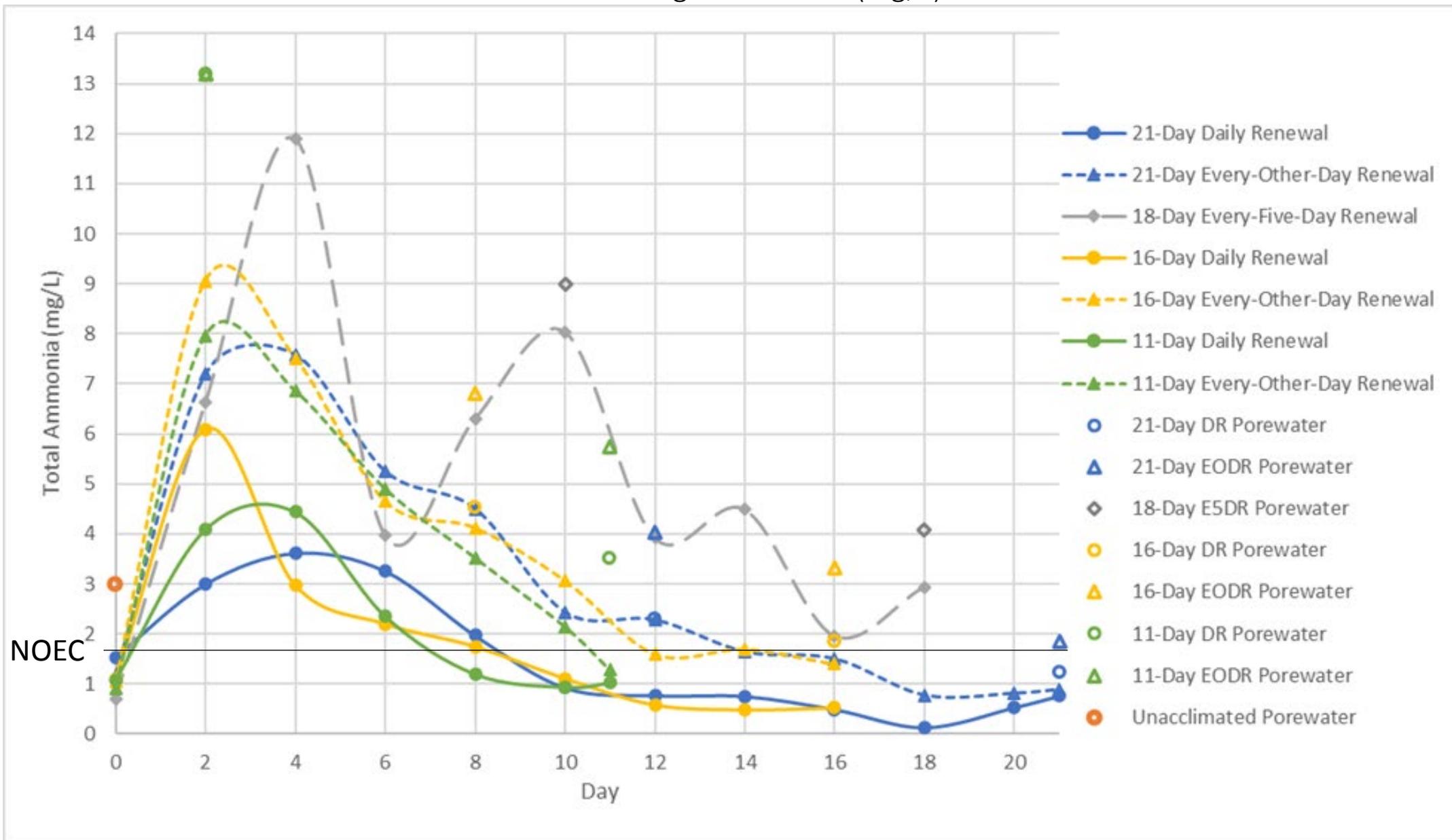


Salinity during Acclimation (ppt)



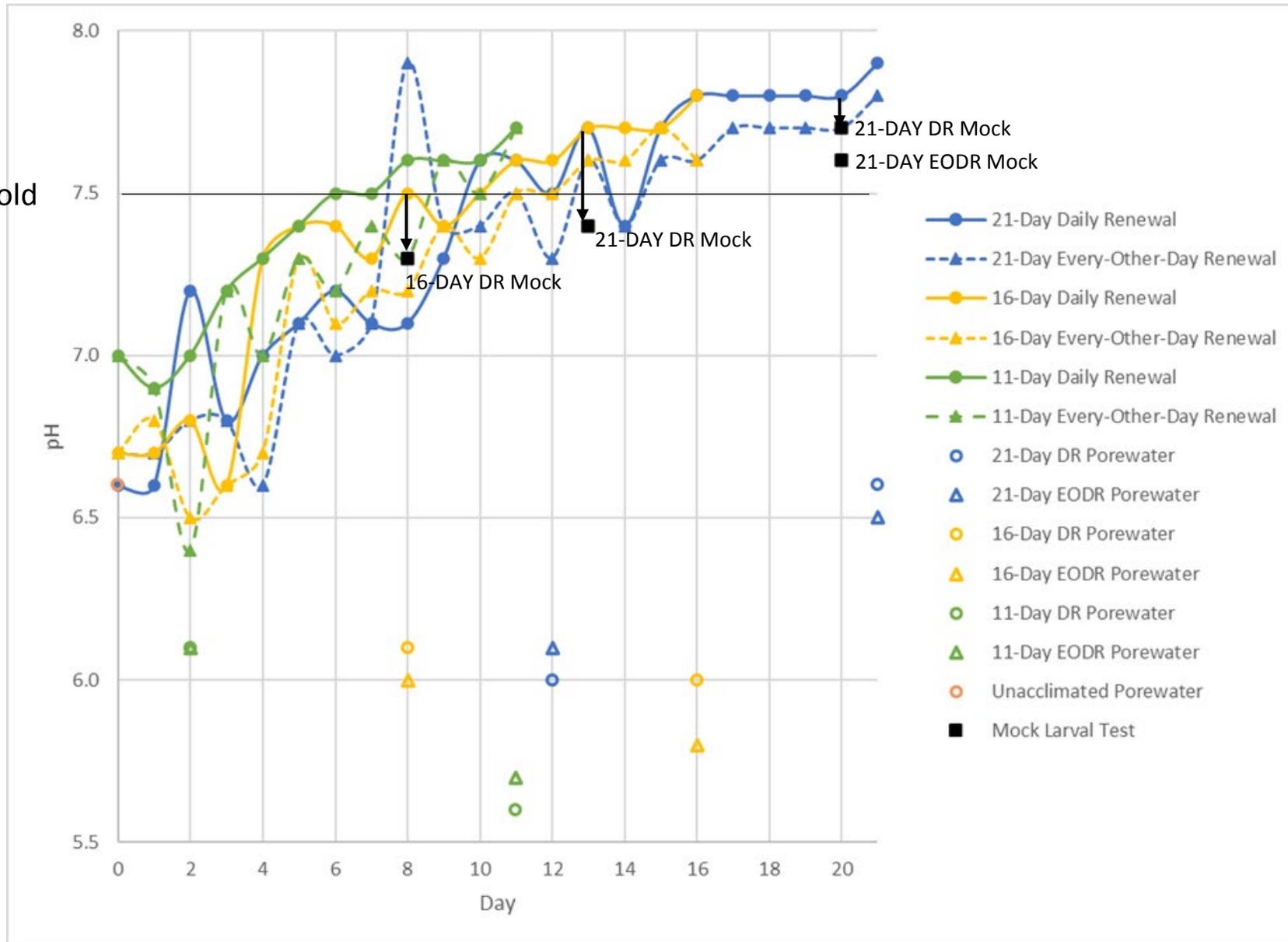
Testing Threshold

Total Ammonia during Acclimation (mg/L)



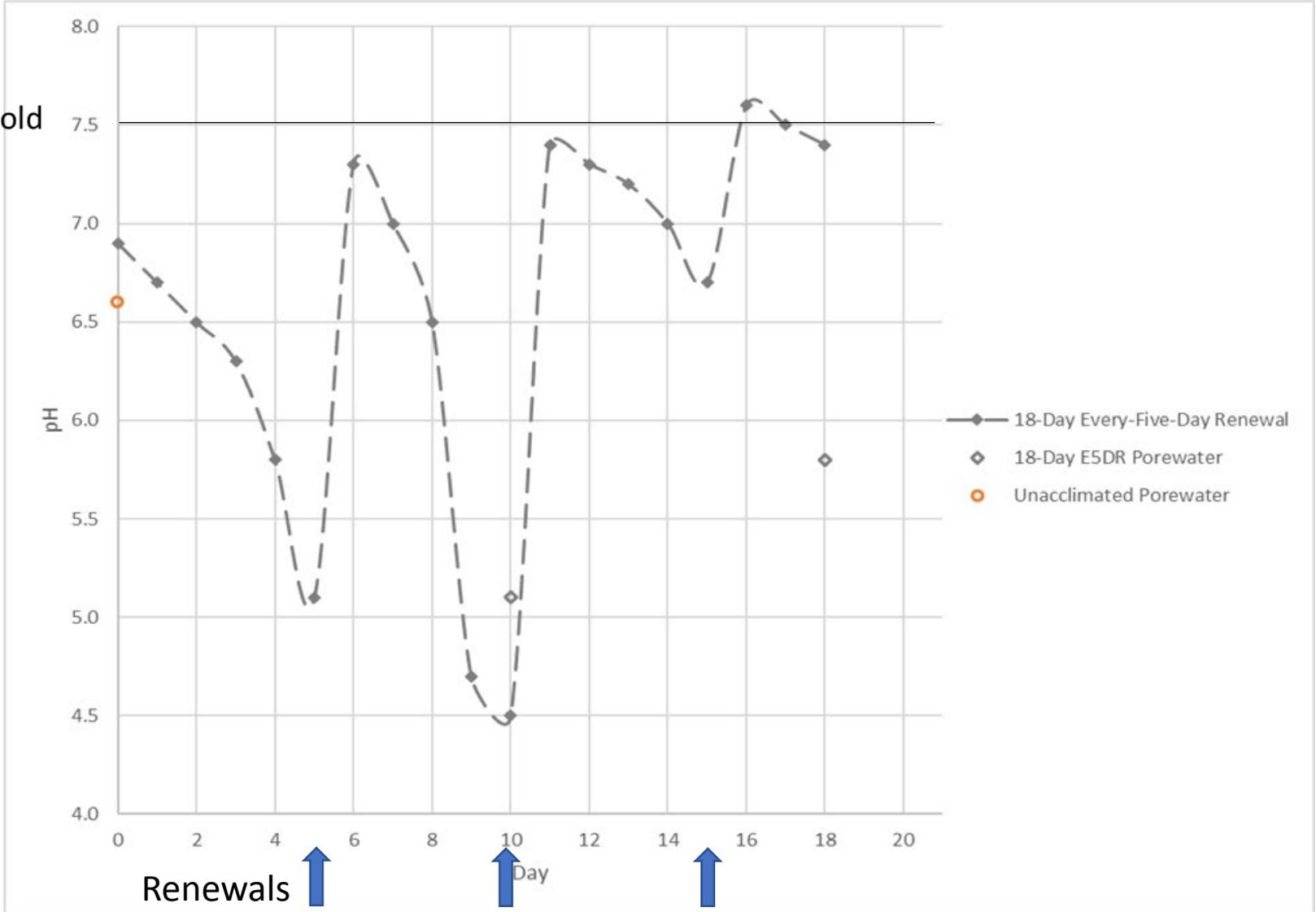
# pH during Acclimation

Testing Threshold



# pH during Every-5-Day Renewal Acclimation

Testing Threshold



# pH at Test Initiation

Acclimation Treatment	Overlying Acclimation Chamber pH	Porewater Acclimation Chamber pH	Initial Test pH
Unacclimated	--	6.6	<b>7.1</b>
11-Day Daily Renewal	7.7	5.6	7.5
11-Day Every-Other-Day Renewal	7.7	5.7	7.5
16-Day Daily Renewal	7.8	6.0	7.6
16-Day Every-Other-Day Renewal	7.6	5.8	7.5
18-Day Every-Five-Day Renewal	7.4	5.8	7.5
21-Day Daily Renewal	7.9	6.6	7.7
21-Day Every-Other-Day Renewal	7.8	6.5	7.6

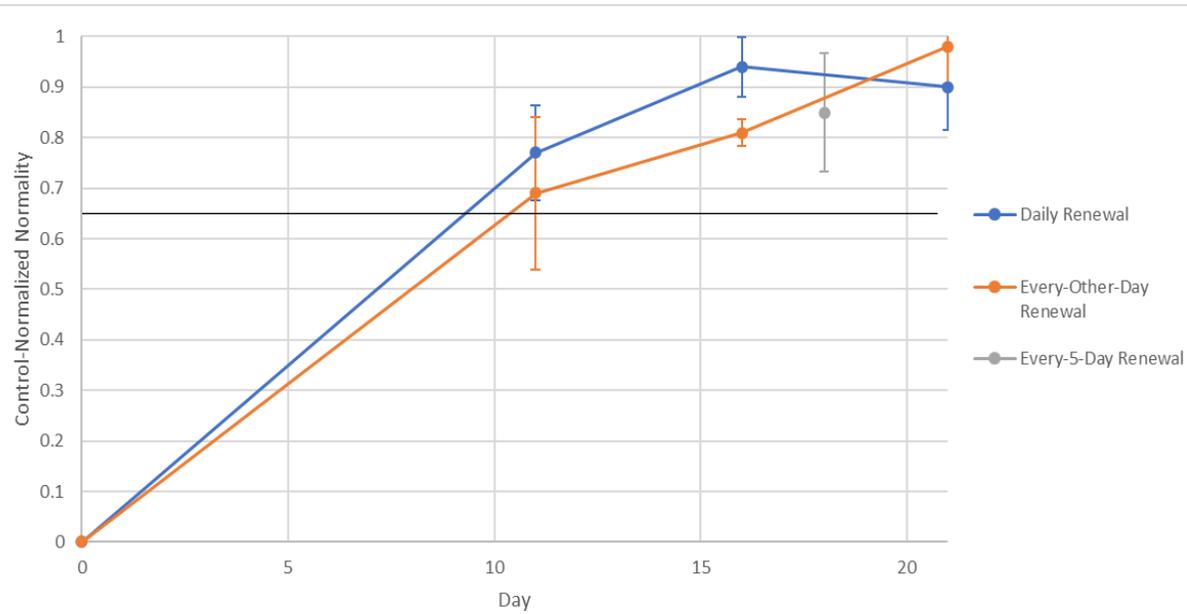
Initial test pH values on average 0.2 units below overlying acclimation chamber pH values (with exception of E5DR)

All water quality within test parameters except pH (7.1) and hydrogen sulfide (0.0053 mg/L) in unacclimated sediment exposure

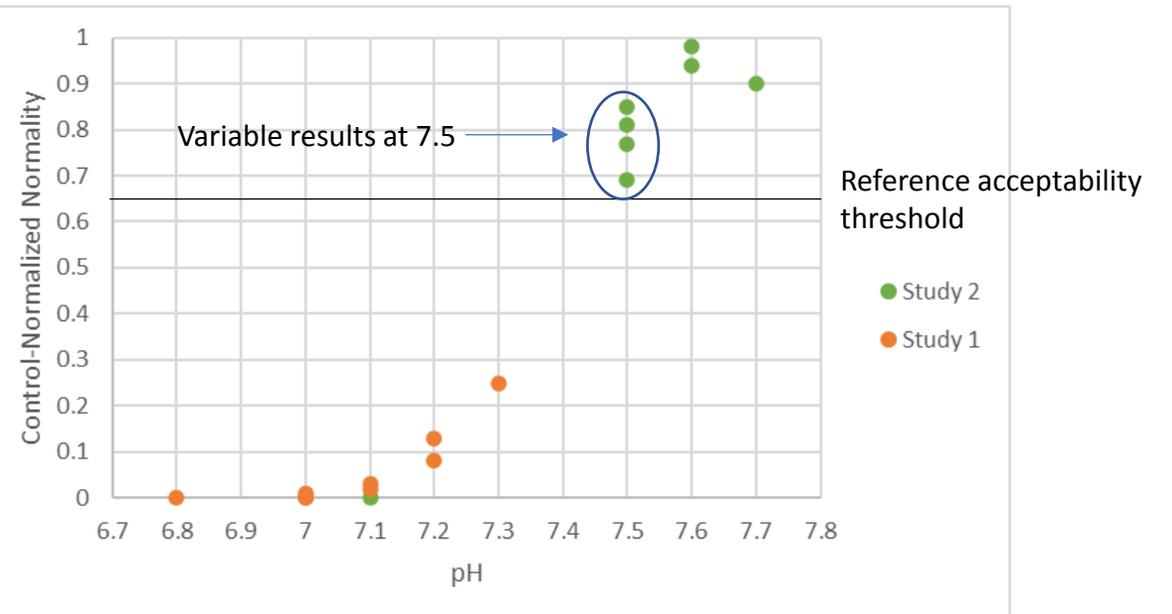
Study 1 initial test pHs ranged from 7 to 7.3 in acclimated sediments

# Larval Test Results

## Normal Survivorship



## Normal Survivorship vs. pH



Normal Larva



Abnormal Larvae

# Recommendations



- Use thin layer method for acclimation (7-10 mm depth, 3.2:1 water:sediment ratio)
  - Decreased acclimation time and improved correlation between acclimation overlying water quality and test water quality over previous acclimation regime.
  - Ferretti et al. 2000\* indicates no substantial loss of metals with thin layer (8 renewals over 4 days, equivalent renewals to 16-day EODR in our study), slight increase in loss of LMW PAHs with the method over other purging techniques (5.6% loss versus 1.7% with no-renewal purge [22 days, 0 renewals] or 2.7% with standard method purge [18 days, 36 renewals]).
- Use pH as the main determinant for when acclimation is complete.
  - When pH stabilizes at 7.8 or above, subsample sediment and setup a mock larval test. If the pH in the mock larval test is 7.6 or above, the sediment is suitable for testing. Measure ammonia, hydrogen sulfide and salinity in mock as well to confirm it is in range.
- Renew every-other-day unless holding time is at risk of being exceeded.

\*Ferretti, J.A., et al. 2000. Evaluation of Methods to Remove Ammonia Interference in Marine Sediment Toxicity Tests. Environmental Toxicology and Chemistry 19(8): pp 1935-1941.