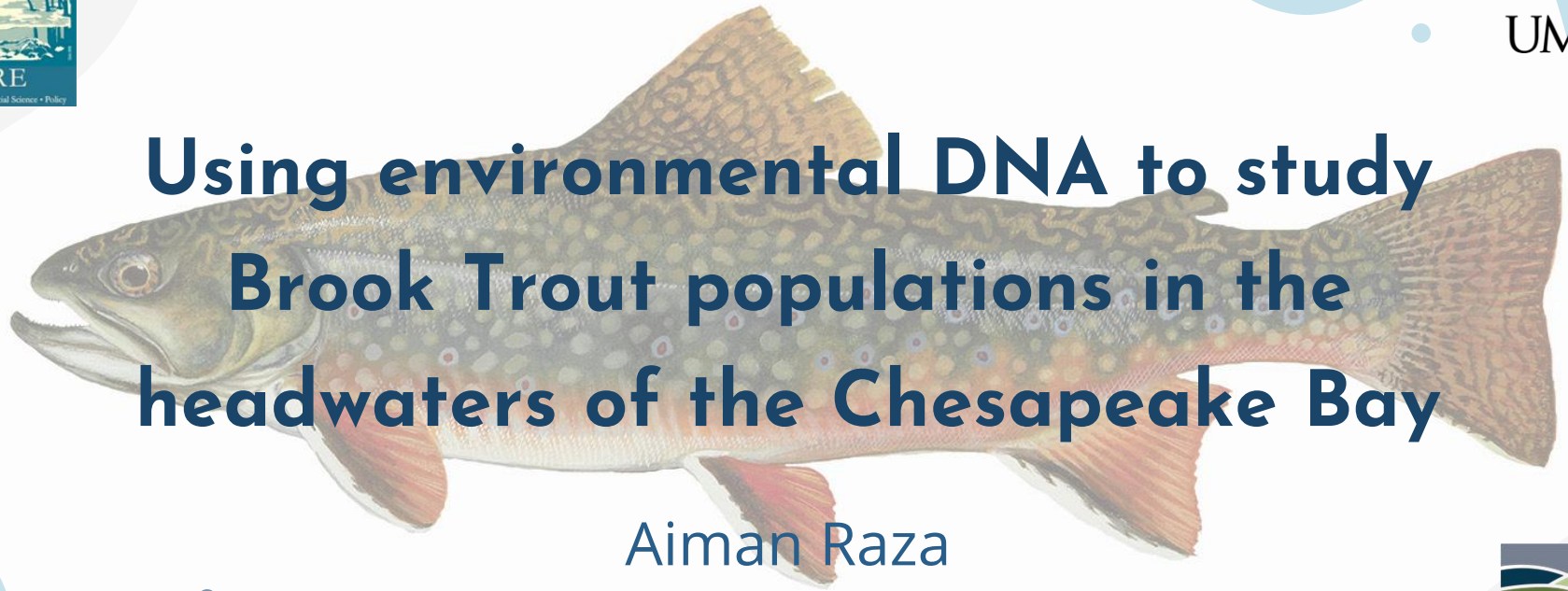


Using environmental DNA to study Brook Trout populations in the headwaters of the Chesapeake Bay

Aiman Raza

Brook Trout Workgroup Meeting

11/1/23

Chesapeake Bay Program Goals for Brook Trout Conservation

CBP Watershed Agreement established an **8% increase** in Brook Trout occupancy by 2025 as one of their Outcomes

Research Objectives

Study the effects of water temperature, distance, and filter pore size on eDNA detection to inform brook trout management

01

Temperature Trials

02

Cage Experiments

03

Filter Pore Size Comparison

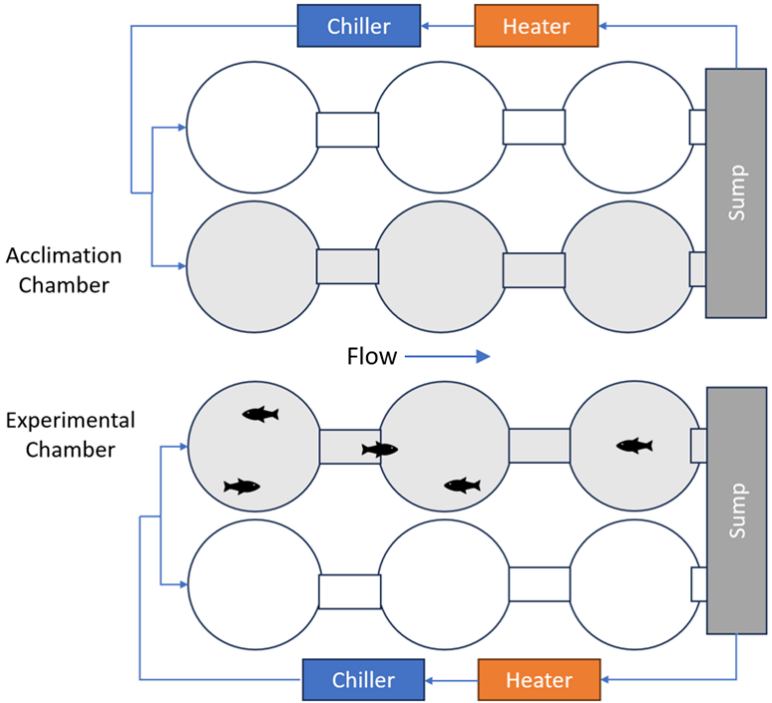
01

Temperature Trials

How does temperature affect brook trout shed rate and eDNA detection?

(January 2023)

Temp Trials: Methods



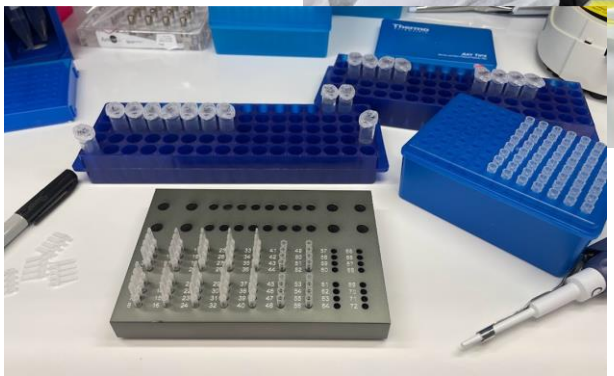
- Recirculating stream tank setup
- Transfer 5 fish to acclimation chamber
- Set to target temp (10° or 20° C)
- Transfer to experimental chamber

Methods: eDNA Sampling

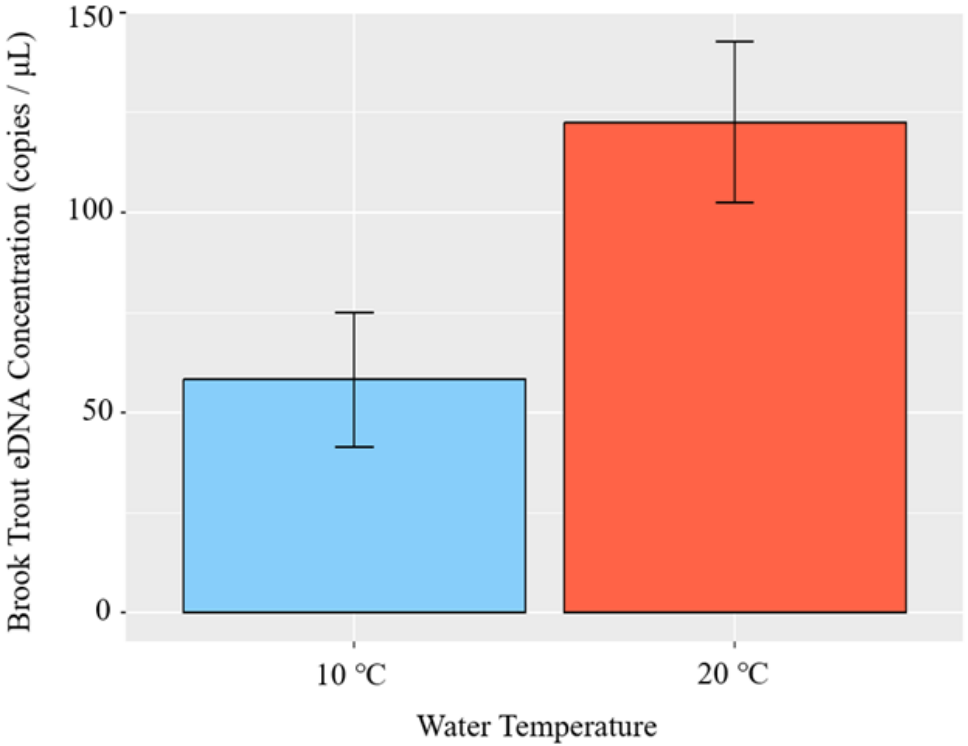
- Take eDNA samples after 1 hour in experimental chamber
  - Smith-Root eDNA Sampler
  - 1 L triplicate samples
  - 1.2 micron filters
- Move fish back to acclimation chamber
- Clean and disinfect
- Repeat trial

Temp Trials: Methods

- Extract DNA (Qiagen DNeasy PowerWater kit)
- Quantitative PCR (qPCR)
  - BRK2 Taqman assay based on Wilcox et al. (2013)
  - Targets brook trout mitochondrial *cyt b* region

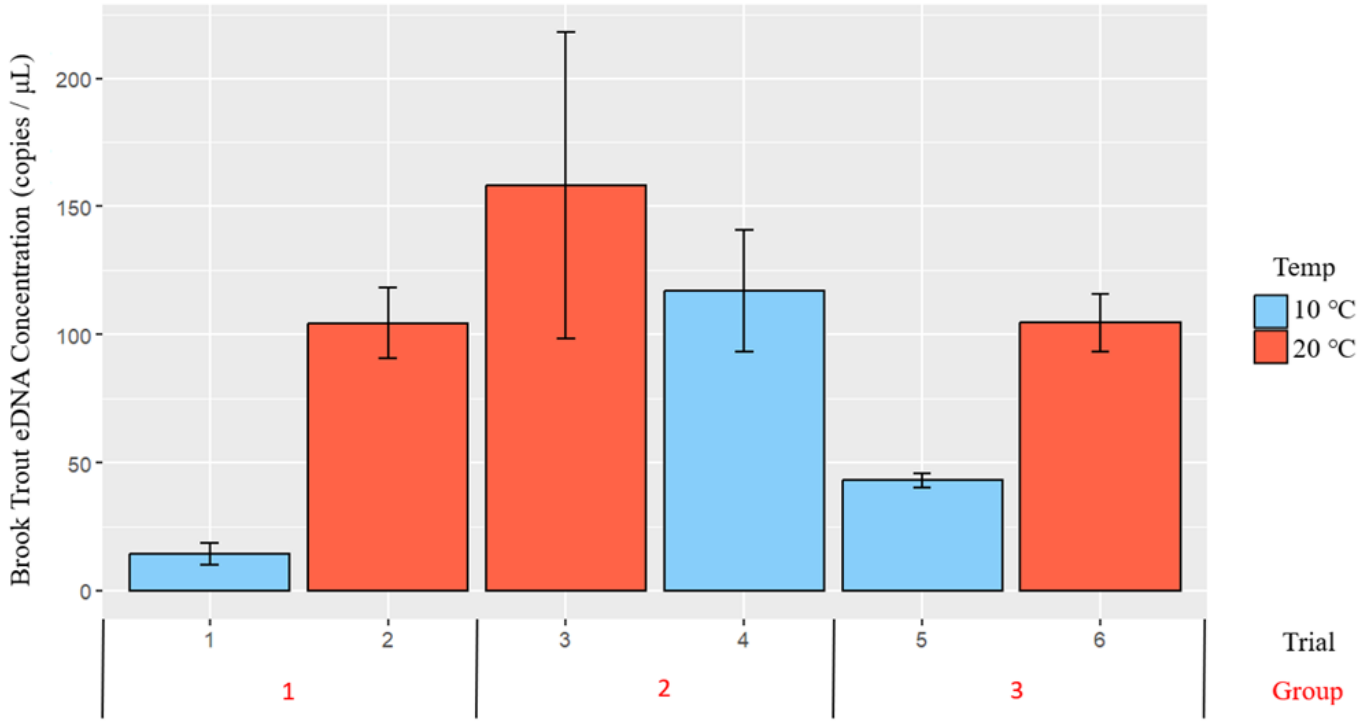


Results: Higher eDNA concentration at higher temperature



P-value:
0.027

Results: eDNA concentration per trial through time



02

Cage Experiments

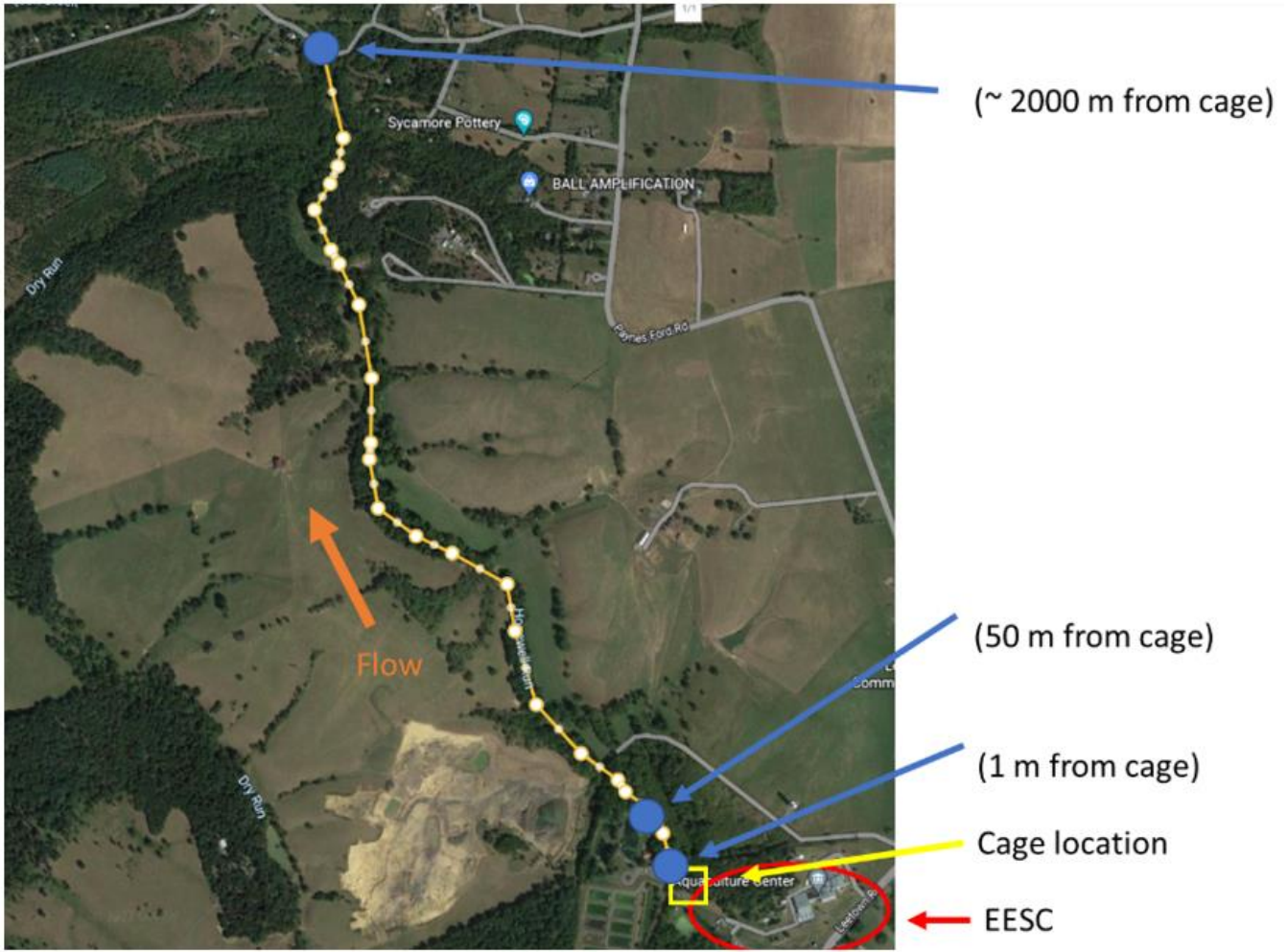
How does distance from a source of eDNA influence eDNA detection in a natural stream environment?

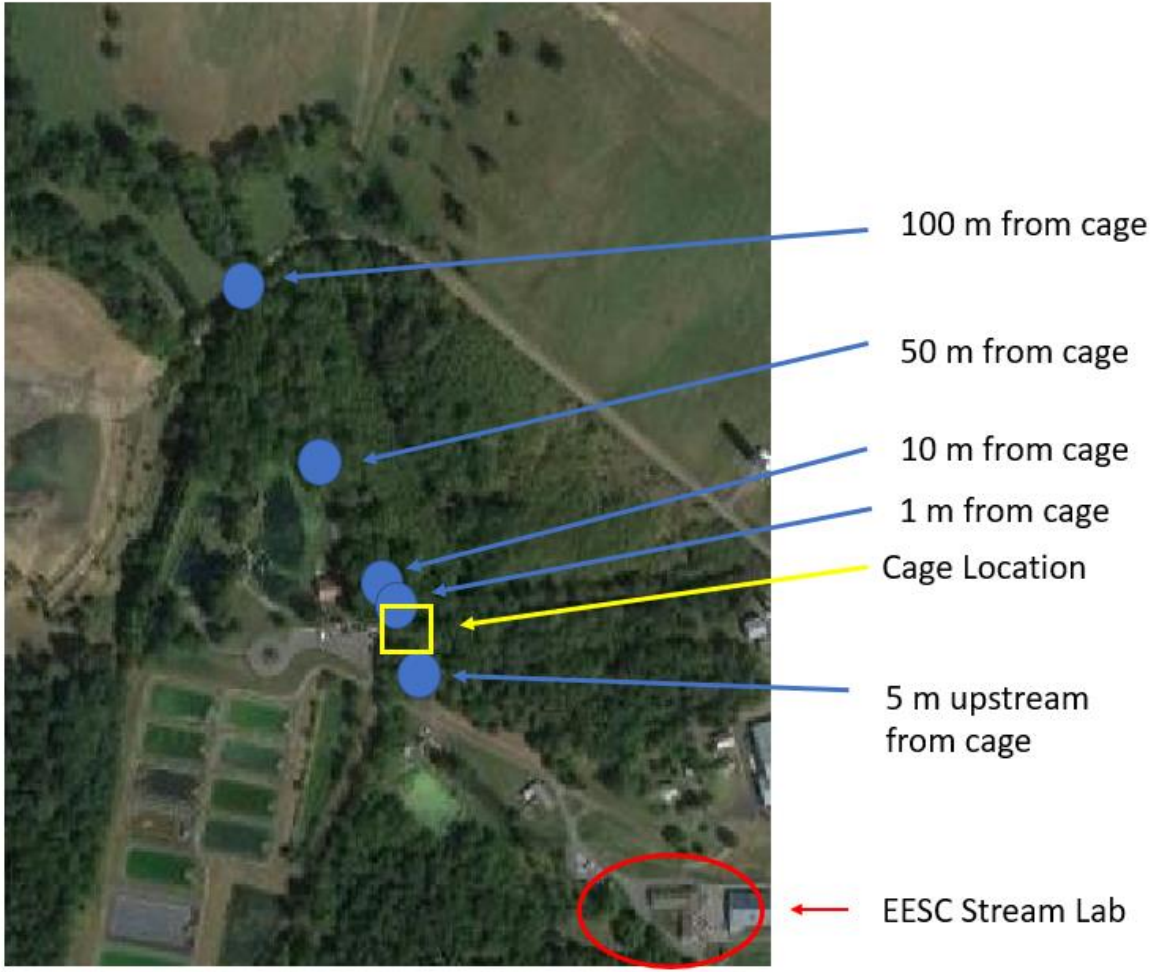
(March & June 2023)

Cage Experiments: Methods

- Placed 3 fish in cage in troutless stream (Hopewell Run, WV)
- After 24 hours, sampled at 1m, 10m, 50m, 100m, and 2000m downstream of cage
- Also sampled 5m upstream of cage

Sampling Sites

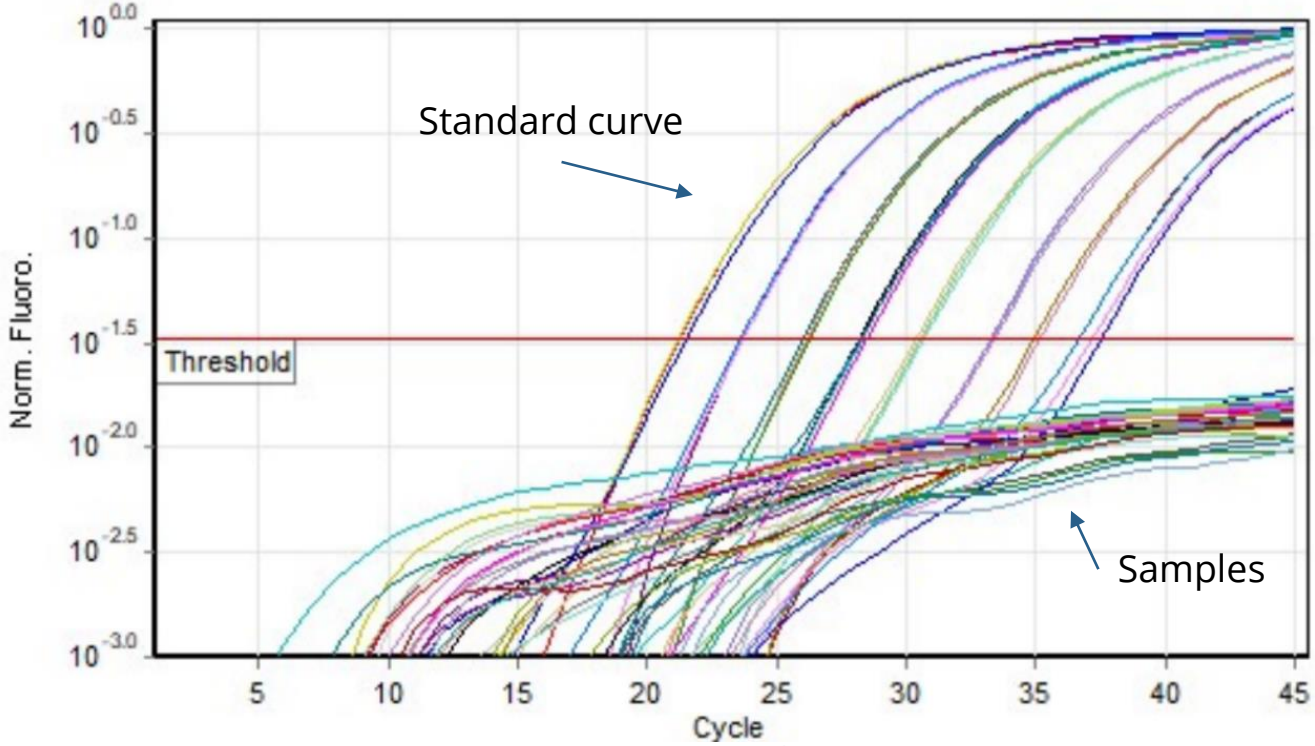



100 m from cage
50 m from cage
10 m from cage
1 m from cage
Cage Location
5 m upstream from cage
EESC Stream Lab

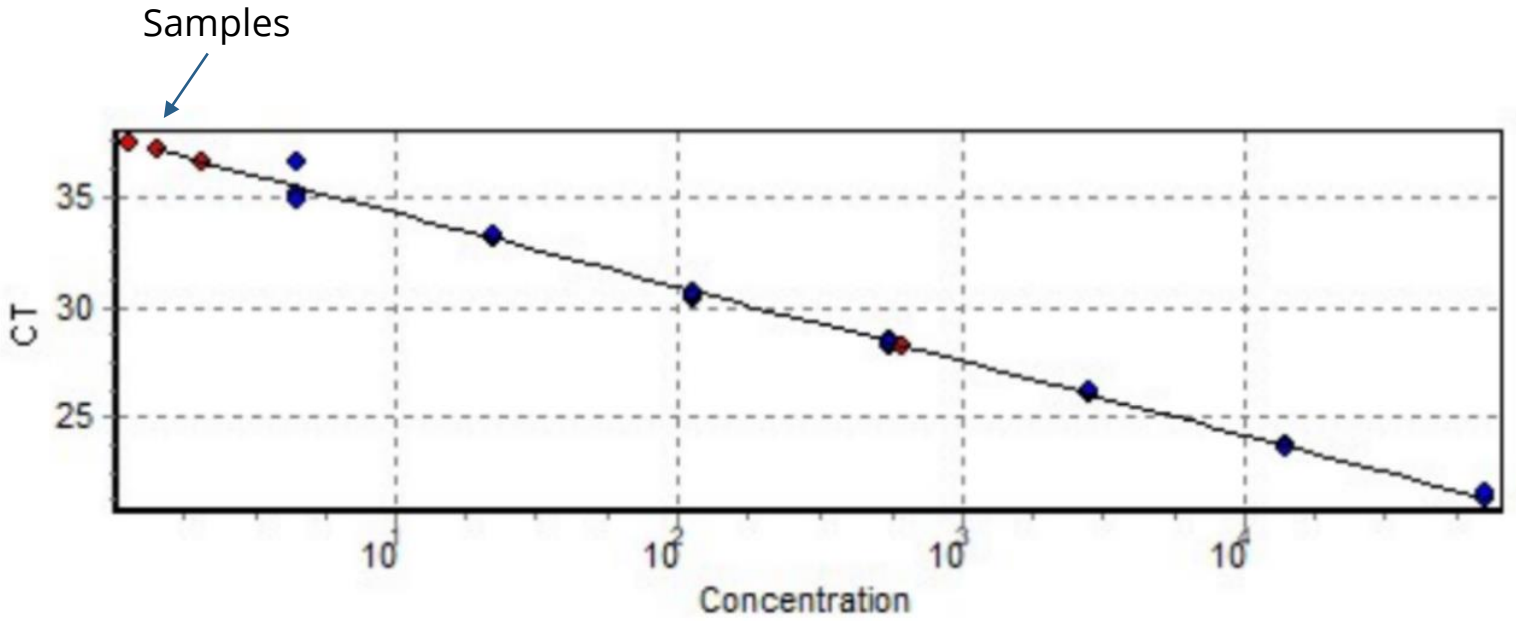
Methods

- Smith-Root eDNA Sampler
  - 1 L triplicate samples
  - 1.2 micron filters
- Extracted DNA and qPCR
- Internal positive controls to test for inhibition

Results: Lack of amplification, cannot accurately quantify



Results: Lack of amplification, cannot accurately quantify



03

Filter Pore Size Comparison

How does filter pore size influence eDNA detection in streams with known brook trout occupancy?

(September - October 2023)

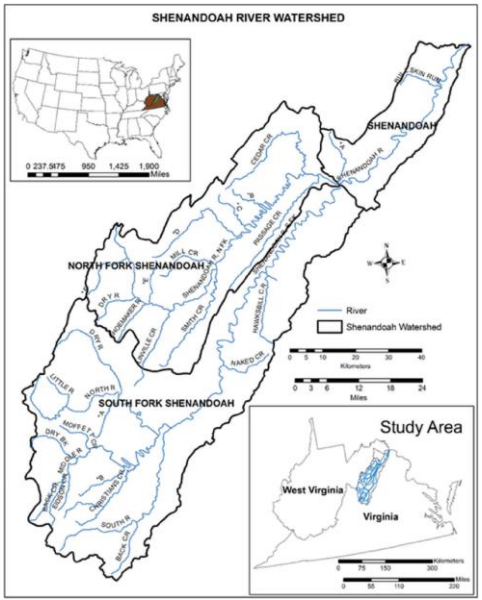
Methods: Filter Pore Size Comparison

- Compare 1.2-micron vs 5-micron filter pore size
- Pair with electrofishing data
- 3 sites in Gunpowder Falls watershed
- 3 sites in Shenandoah watershed

Credit: Smith-Root

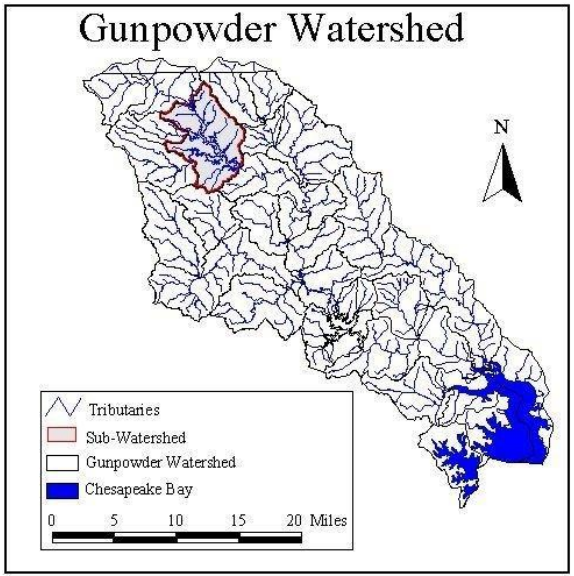
Fieldwork Locations

Shenandoah



Credit: Mbuh et al. (2018)

Gunpowder Falls



Credit: Gunpowder Watershed Coalition

Methods: Filter Pore Size Comparison

- Smith-Root eDNA Sampler
  - 9 L triplicate samples
  - 1.2 and 5 micron filters
- Extracted DNA and qPCR
- Internal positive controls to test for inhibition

Conclusion and Next Steps

- Higher eDNA concentrations at higher temperatures
- May need to sample higher volumes of water
- Compare filter types (PES vs glass fiber)?

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  - Cheryl Morrison
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- MD DNR - Mark Staley
- SNP - Evan Childress

Thank you!
Questions?

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