Application of two novel qPCR assays for quantifying pinniped environmental DNA (eDNA) in coastal environments

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Acknowledgements

Advisors and Co-Authors

- Kristina Cammen, University of Maine
- Michael Kinnison, University of Maine
- Lisa Sette, Center for Coastal Studies
- Christine Hudak, Center for Coastal Studies
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- Sydney Jackson, University of Maine
- Geneva York, University of Maine Environmental CORE Facility

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- Maine Outdoor Heritage Fund

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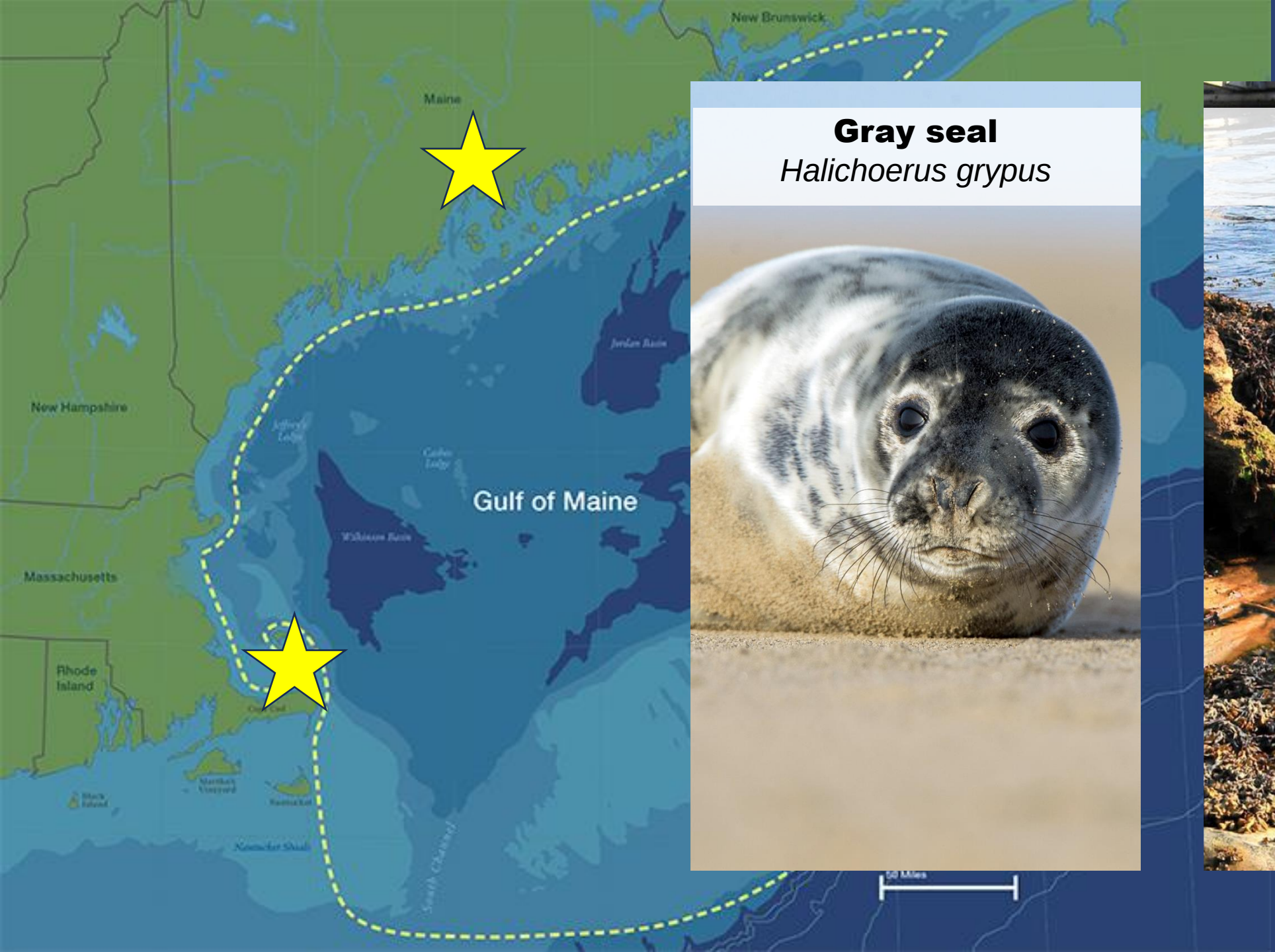
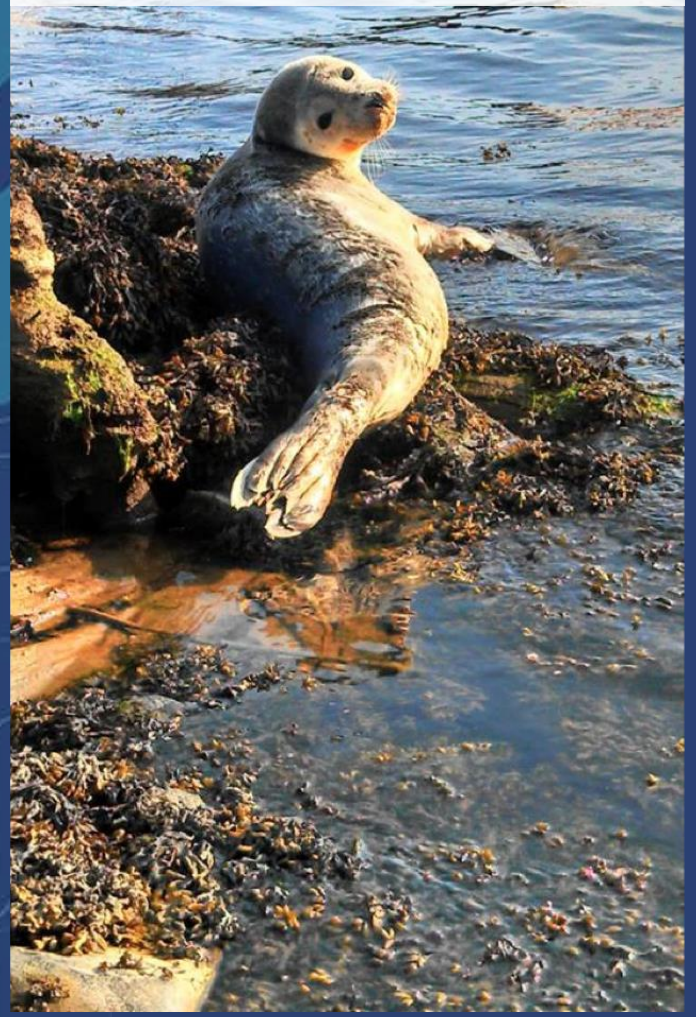


Gray seal
Halichoerus grypus

Harbor seal
Phoca vitulina



Primer Development

in silico Design and Testing

- Identify gene region candidates
- Minimize potential for cross-amplification of non-target species

Specificity Testing

- Tissue extract from target and non-target species
- eDNA extract from rehabilitation pools

Assay Optimization

- Primer/probe concentrations
- Annealing temperature
- Amplification efficiency

Gray Seal Assay

106 bp
ND4 gene

Harbor Seal Assay

173 bp
cytb gene

Primer Deployment

Cape Cod, Massachusetts

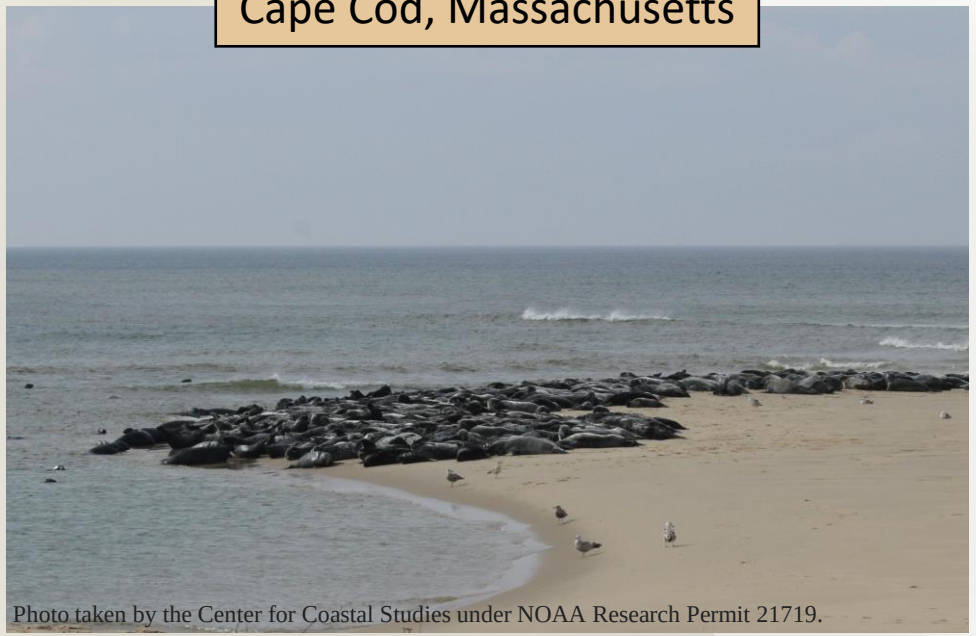


Photo taken by the Center for Coastal Studies under NOAA Research Permit 21719.

Large gray seal aggregations (18-325 individuals)

Targeted sampling around haul-outs (along shoreline and 50m offshore)

Tested for gray seal

Penobscot River, Maine

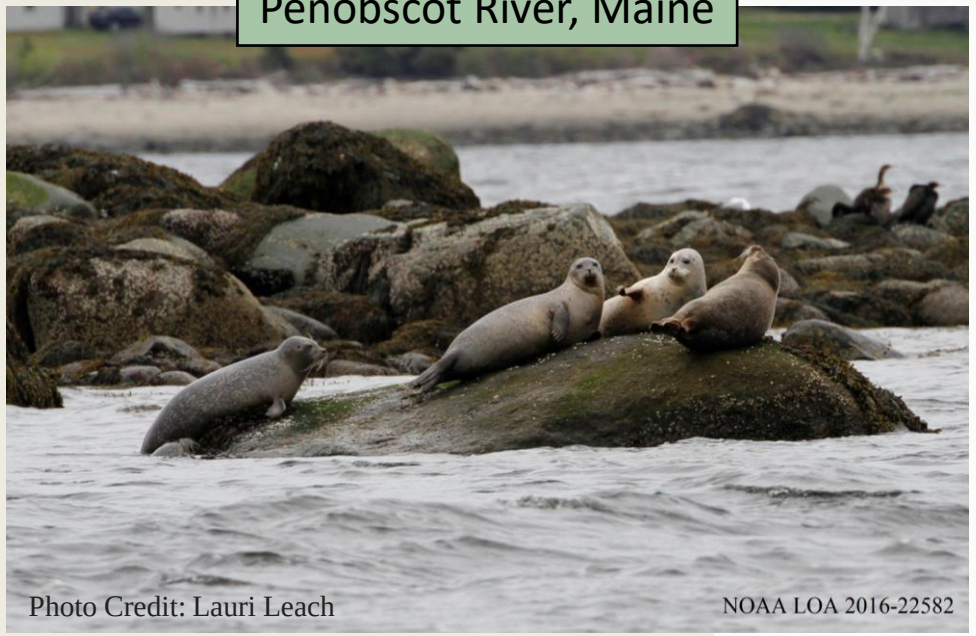


Photo Credit: Lauri Leach

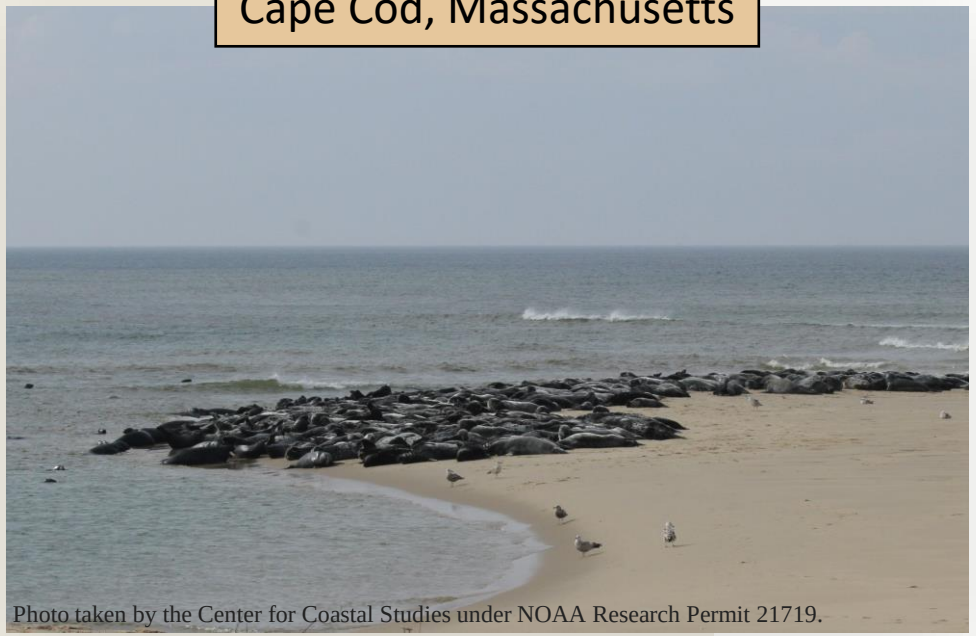
NOAA LOA 2016-22582

Smaller seal aggregations (1-30 individuals)

Samples collected along a transect up the lower estuary

Tested for gray and harbor seals

Cape Cod, Massachusetts



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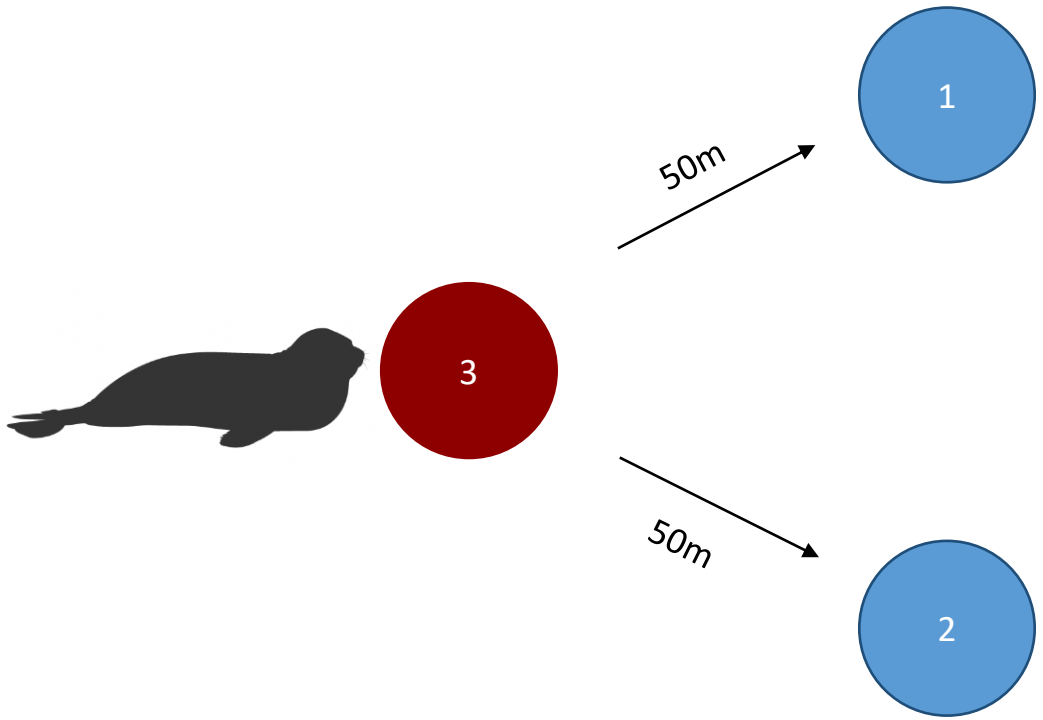
Detection Success Across Systems

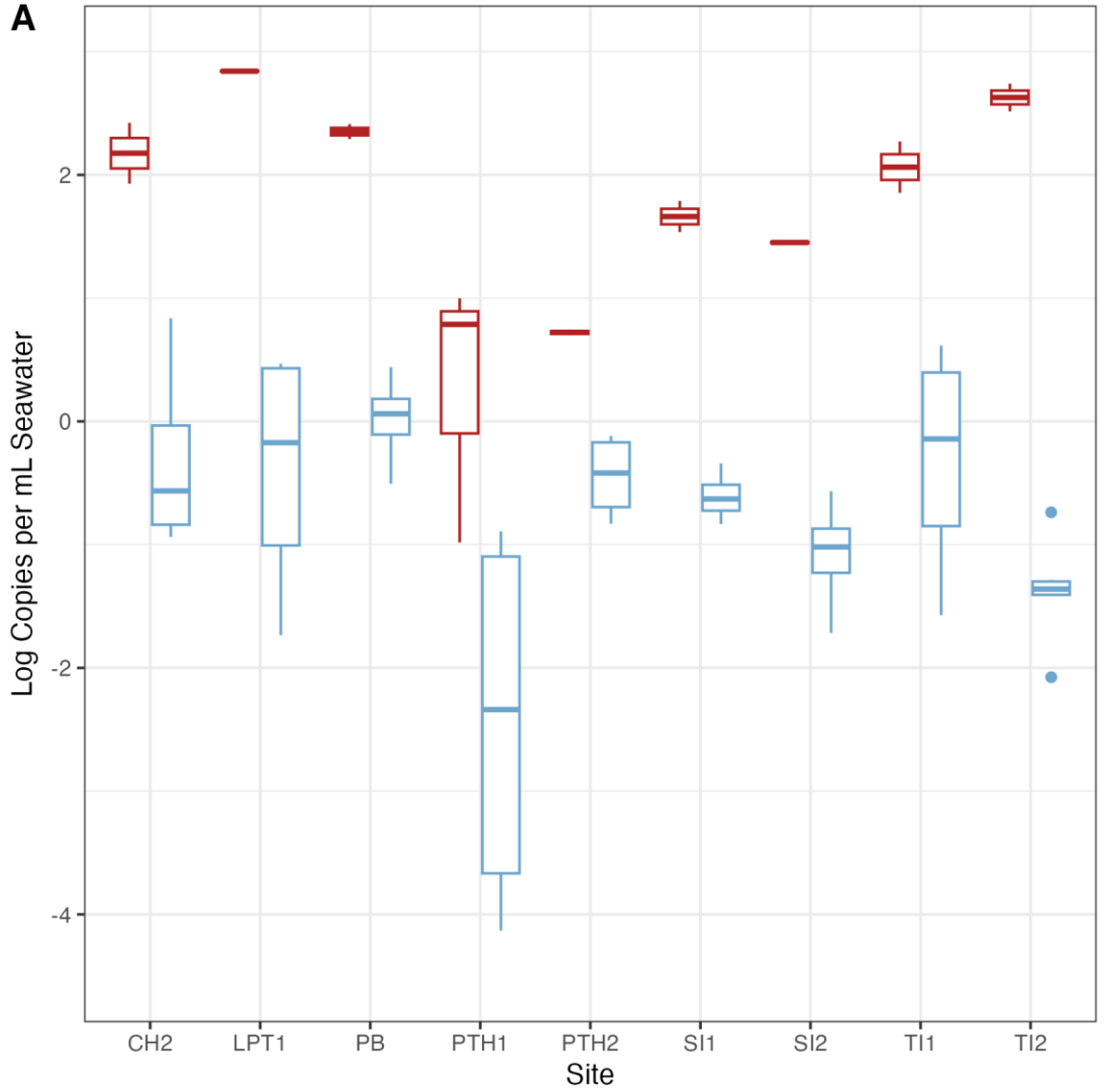
92% samples collected 0 - 50 meters from shore detected gray seal eDNA at haulouts with 18-325 seals present

In 2 expanded surveys, gray seal eDNA was detected up to 150 meters from shore

Quantification in Space

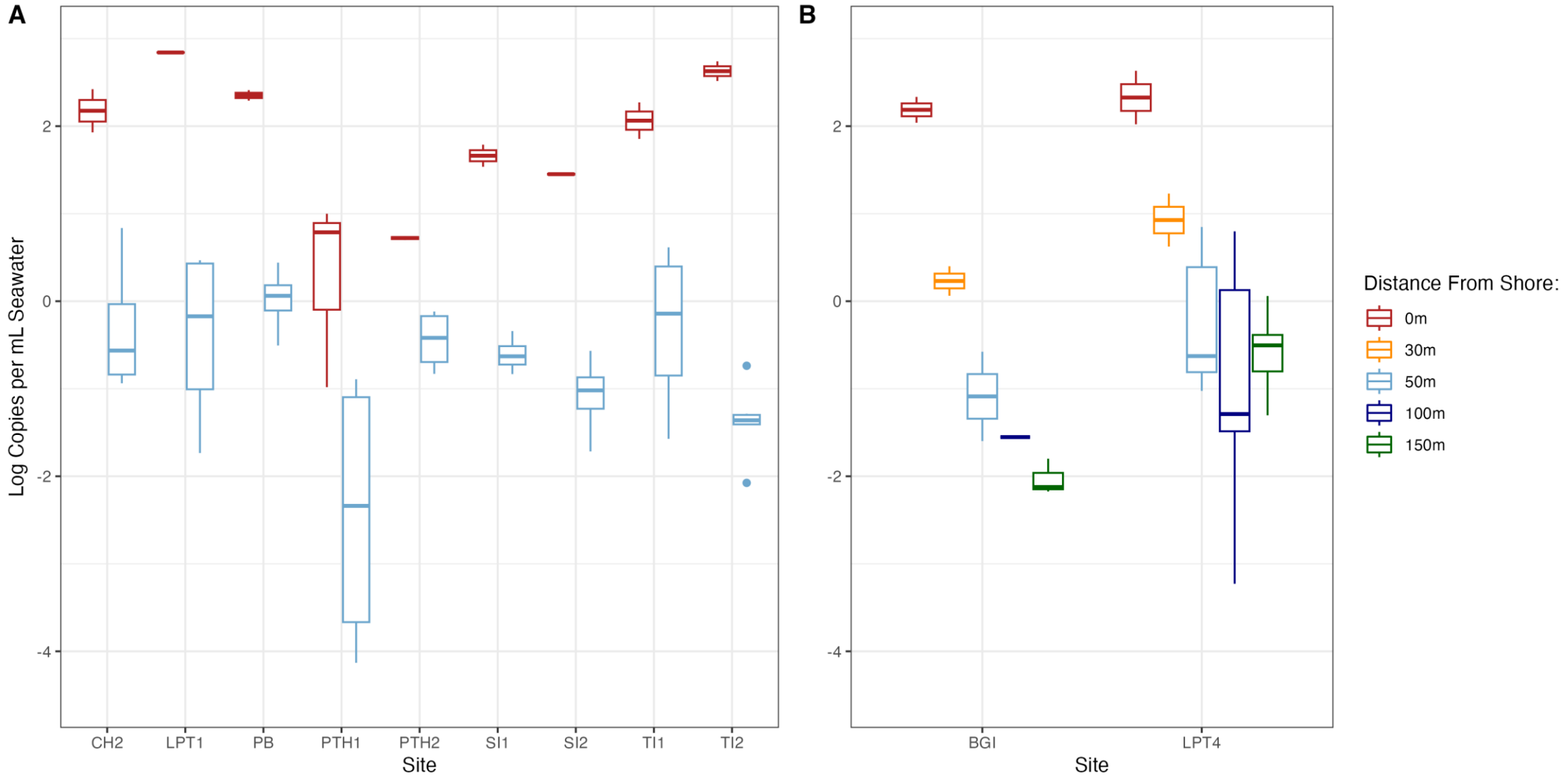
Cape Cod, Massachusetts



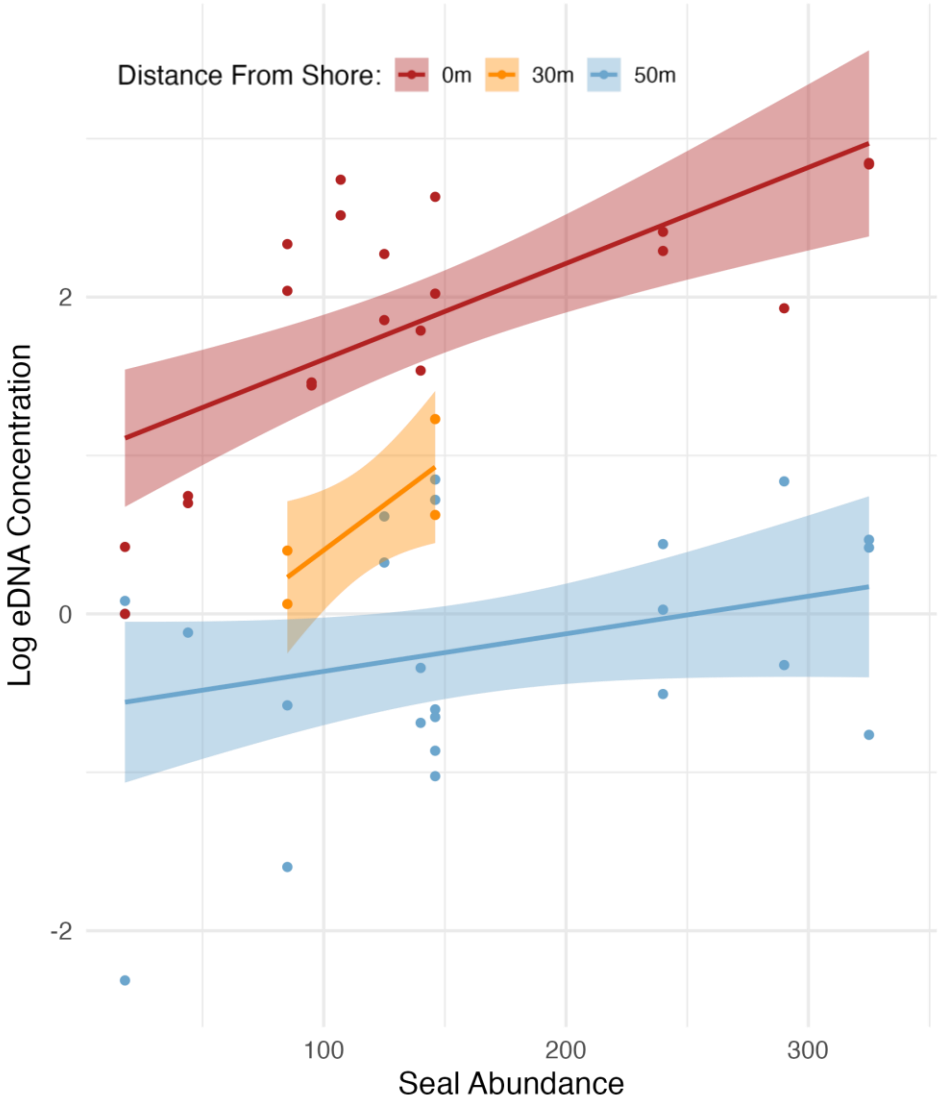


Quantification in Space

Signal continues to dissipate with distance



eDNA verses Abundance

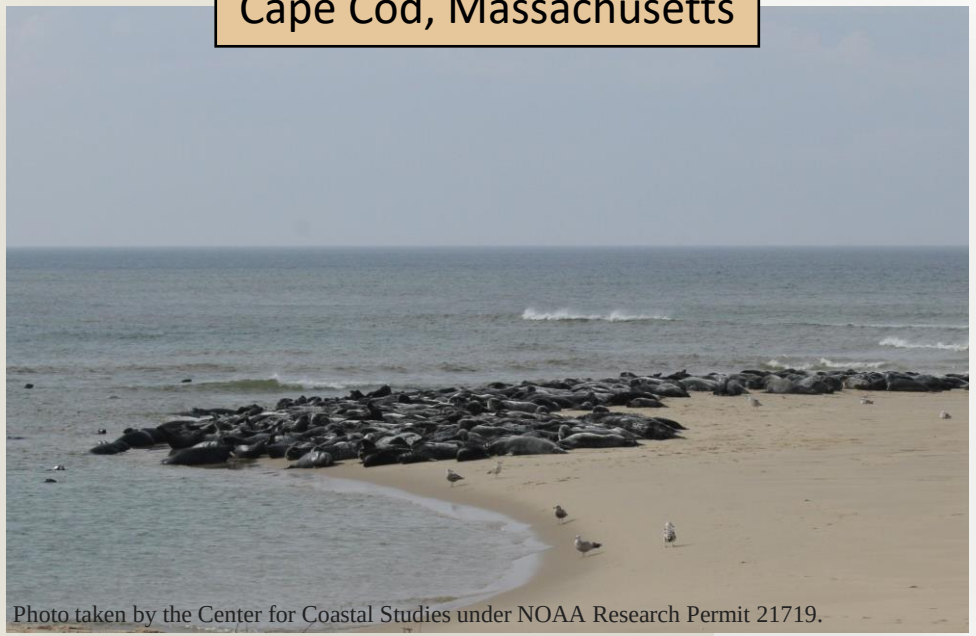


Wide range of haul-out sizes offered the opportunity to test the influence of abundance on eDNA concentration

Together, distance from shore and seal abundance explained 73.88% of observed deviance in eDNA concentration

Distance had a stronger effect on eDNA concentration than seal abundance

Cape Cod, Massachusetts



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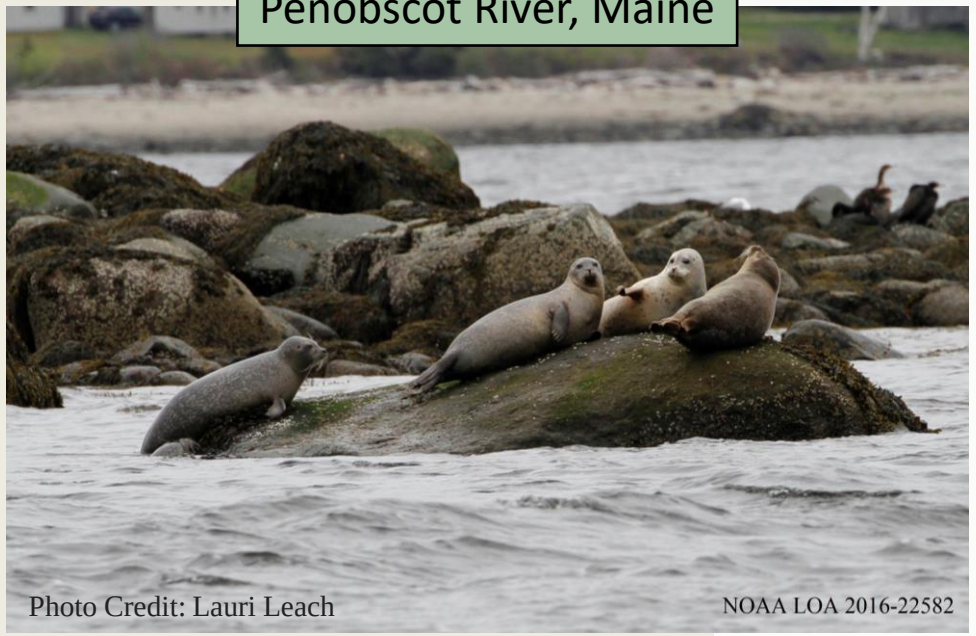

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System characteristics and sampling scheme support a high rate of detection

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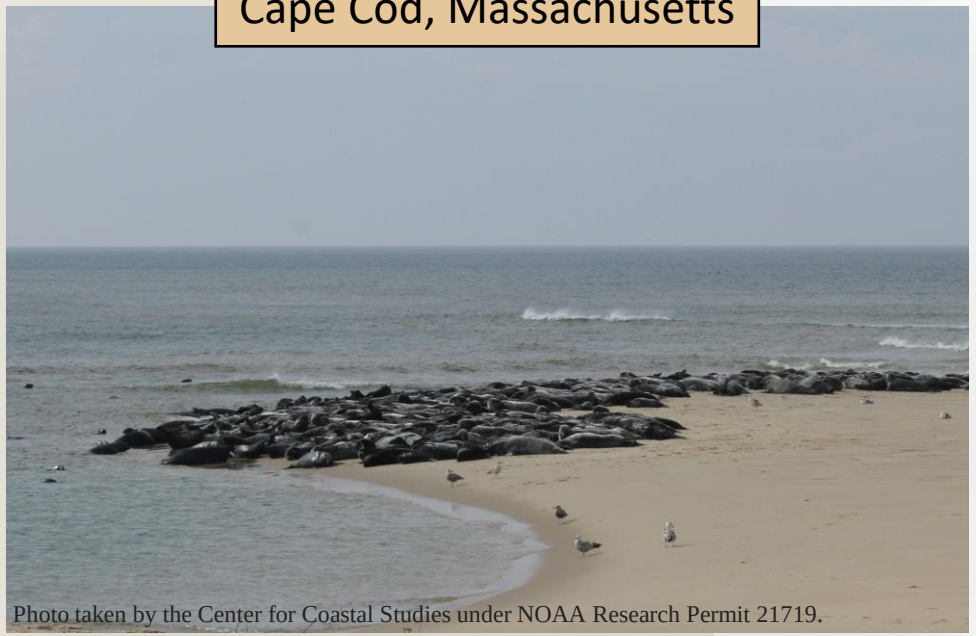

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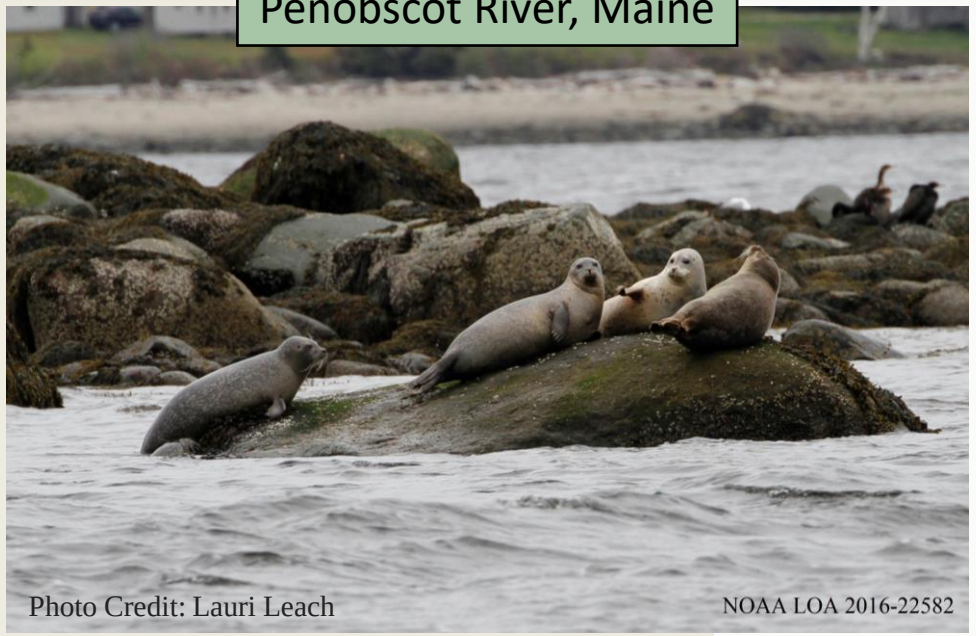

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NOAA LOA 2016-22582

Targeted analysis of samples collected during surveys with the highest number of seals observed (<30 individuals)

12% samples detected gray seal, 17% detected harbor seal

Detection Success Across Systems

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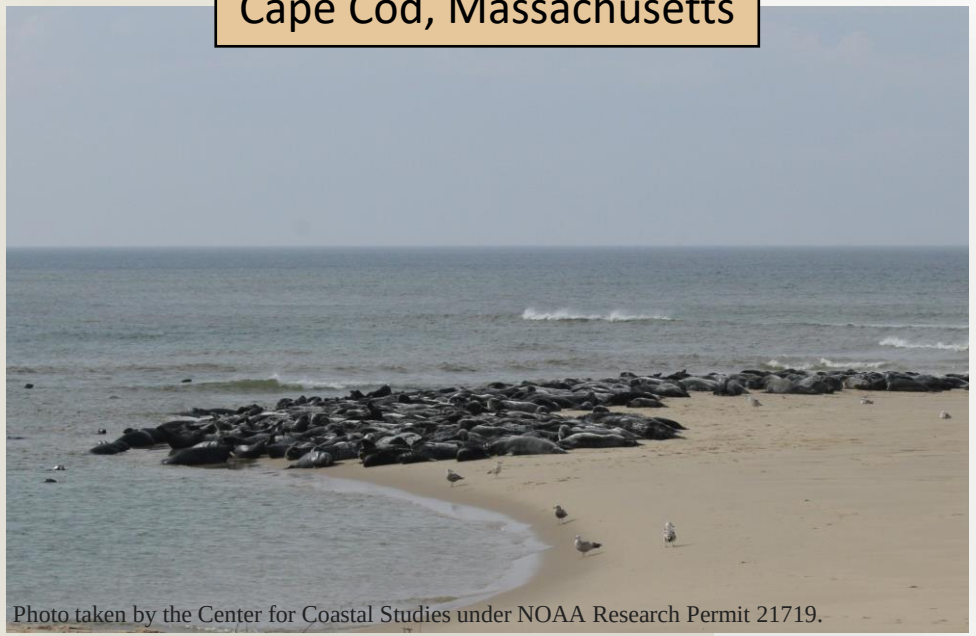

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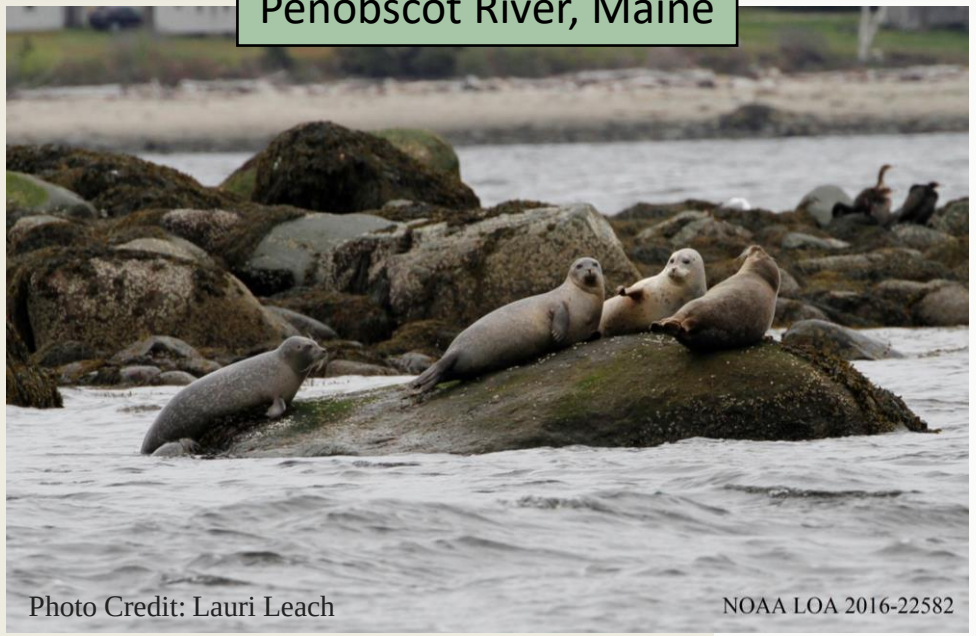

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NOAA LOA 2016-22582

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Detection more sporadic, high rate of false positives

qPCR as a supporting data stream

Cape Cod, Massachusetts

- Accompanied by eDNA haplotyping
- qPCR data established a baseline understanding of eDNA signal in space around haul-out sites
  - Guide haplotyping efforts and help interpret detections

qPCR as a supporting data stream

Learn more about our eDNA haplotyping efforts on Monday!

Cape Cod, Massachu

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  - Guide h interpret detections

Program Track 1

Ecology and Evolution - eDNA Application

Mon, November 11 | 11:30 AM - 12:30 PM | BelleVue Ballroom 2 | Long and Short (or Video) Presentations Session

12:18 PM
12:30 PM

Evaluating the Use of Environmental DNA for Pinniped Detection and Population Genetics at Haul-Out Sites

Julia Sunnarborg[1], Christine Hudak[2], Lisa Sette[2], Geneva York[3], Sydney Jackson[4], Michael Kinnison[4], Kristina Cammen[4]

[1]University of Maine, Bangor, Maine, [2]Center for Coastal Studies, Provincetown, Massachusetts, [3]University of Maine Environmental DNA CORE Laboratory, Orono, Maine, [4]University of Maine, Orono, Maine

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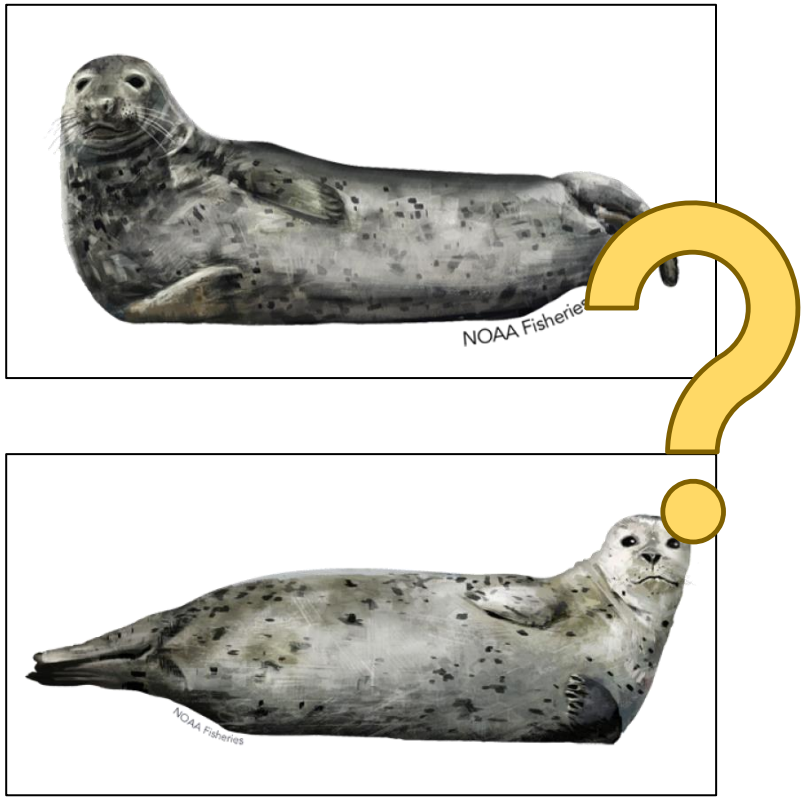
Penobscot River, Maine

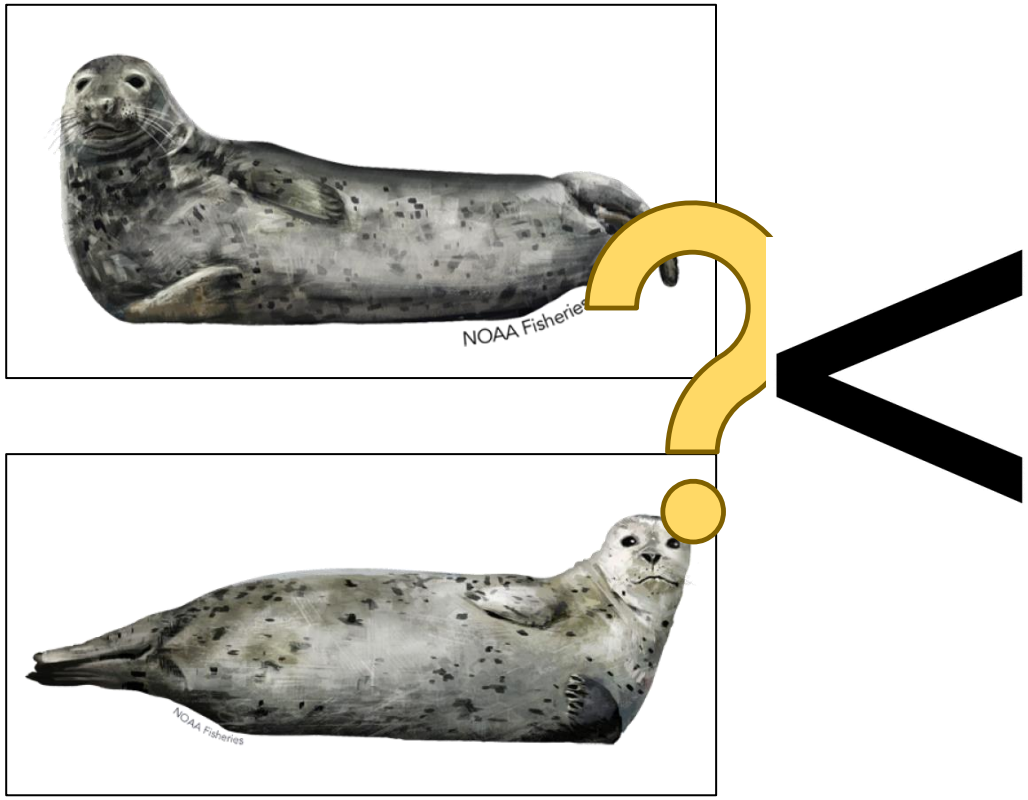
- Accompanied by 12S rRNA metabarcoding to characterize prey community

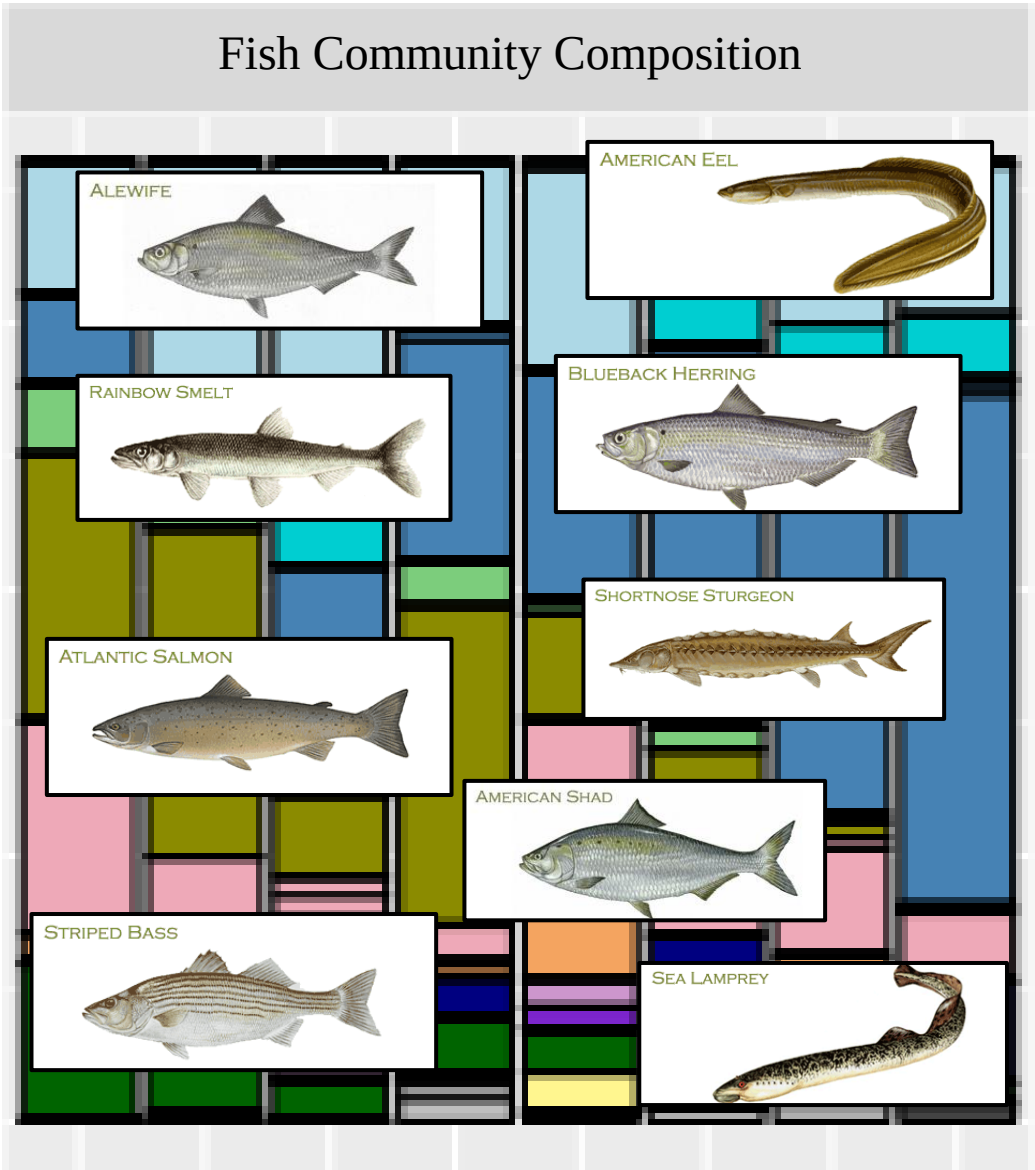
Metabarcoding – Where are the seals?



No seal reads even when visually observed

Metabarcoding – Where are the seals?

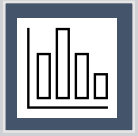




Takeaways

qPCR assays created and field tested for gray and harbor seals

Conditions of the system and sampling setup influence detection rates and the potential for false negatives



Quantifying eDNA variation in space helps us understand the underlying processes of transport and signal dissipation for a given system

qPCR tools may provide a way to “pick out” seal detections where they might otherwise be swamped out

Thank you!

Please reach out!

julia.sunnarborg@maine.edu