

Detecting Rare Targets: Optimizing an eDNA Workflow for Detecting Cetaceans

Megan Shaffer, PhD

Elizabeth Andruszkiewicz Allan, Pedro Brandão, Elizabeth Brasseale, Gledis Guri, Owen Liu, Krista Nichols, Parker MacCready, Kim Parsons, Andrew Shelton, Tania Valdivia Carrillo, Jilian Xiong, Amy Van Cise, Ryan Kelly



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United States Office of Naval Research Marine Mammals and Biology Program

Supports basic and applied research and technology development related to understanding the effects of sound on marine mammals, including physiological, behavioral, ecological and population-level effects

Monitoring and Detection – passive acoustics, infrared and environmental DNA

Marine Mammal Remote Detection via Innovative environmental DNA Sampling



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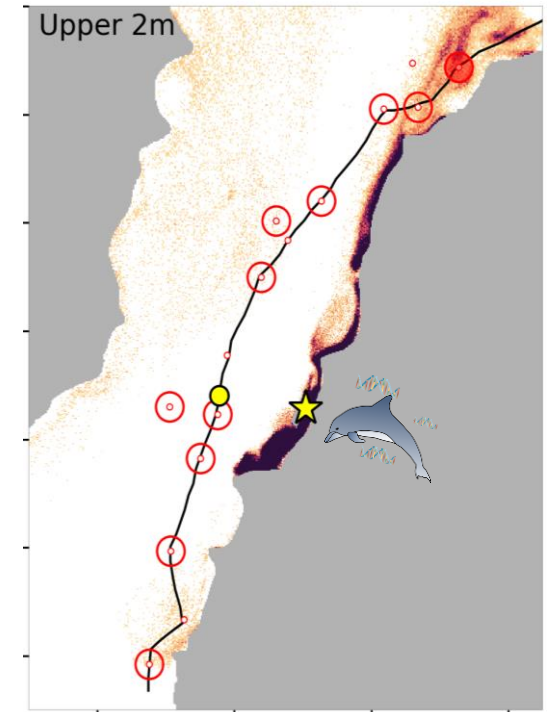
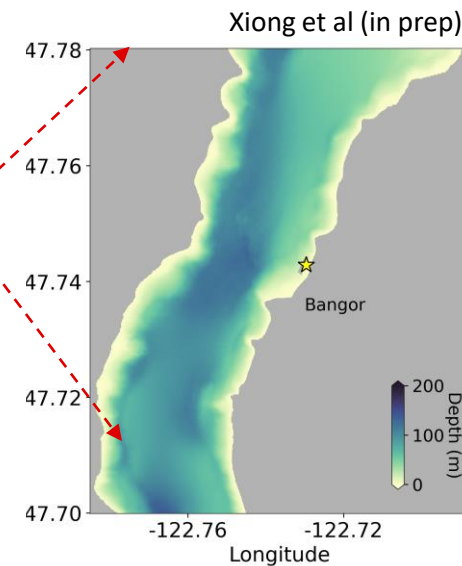
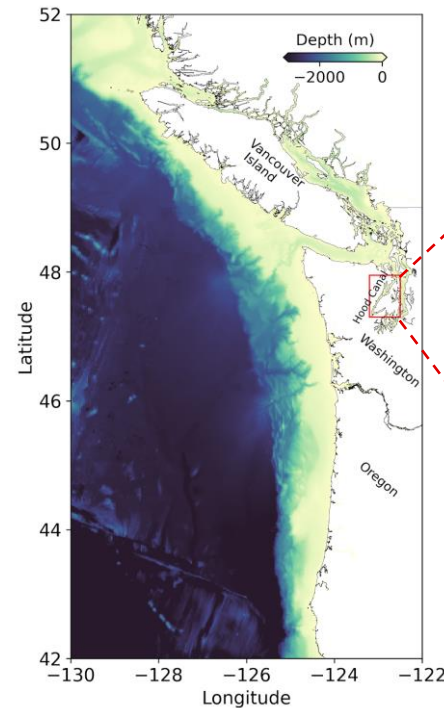
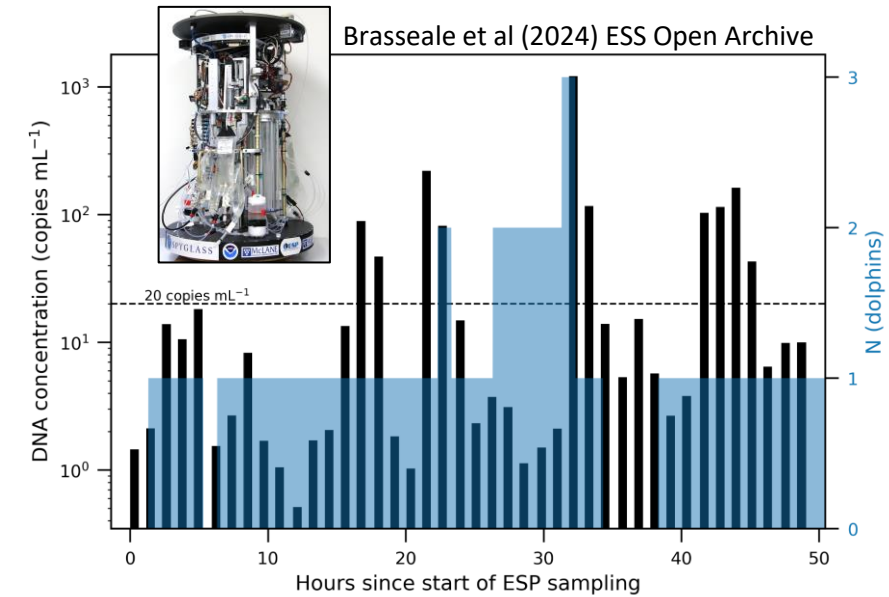
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Marine Mammal Remote Detection via Innovative environmental DNA Sampling

1. To understand the fate and transport of eDNA and interpret eDNA detections from marine mammals spatially and temporally.



Contact: Dr. Kim Parsons, kim.parsons@noaa.gov

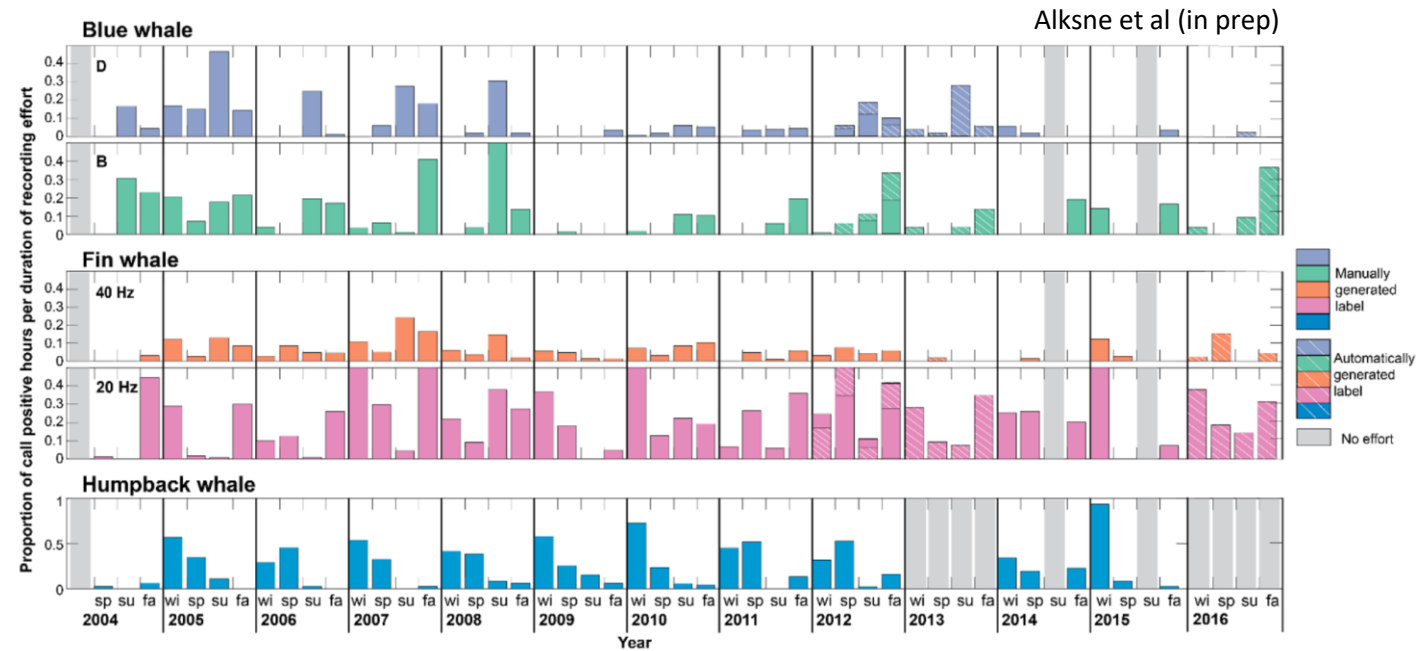
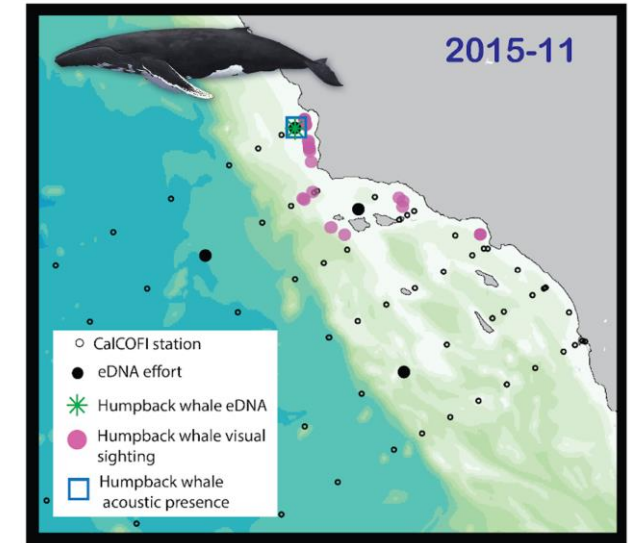
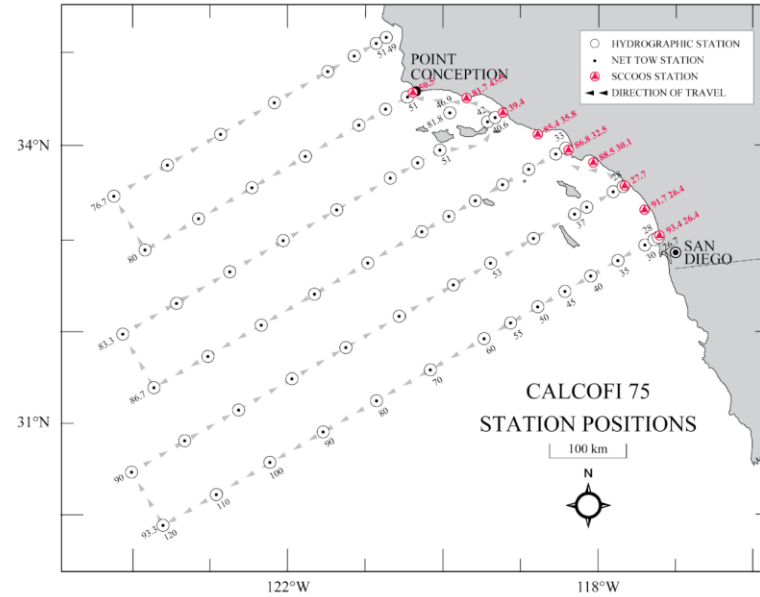


Marine Mammal Remote Detection via Innovative environmental DNA Sampling

2. To evaluate eDNA as a complement to traditional visual observations and acoustics detections for cetaceans.



Contact: Dr. Brice Semmens, bsemmens@ucsd.edu



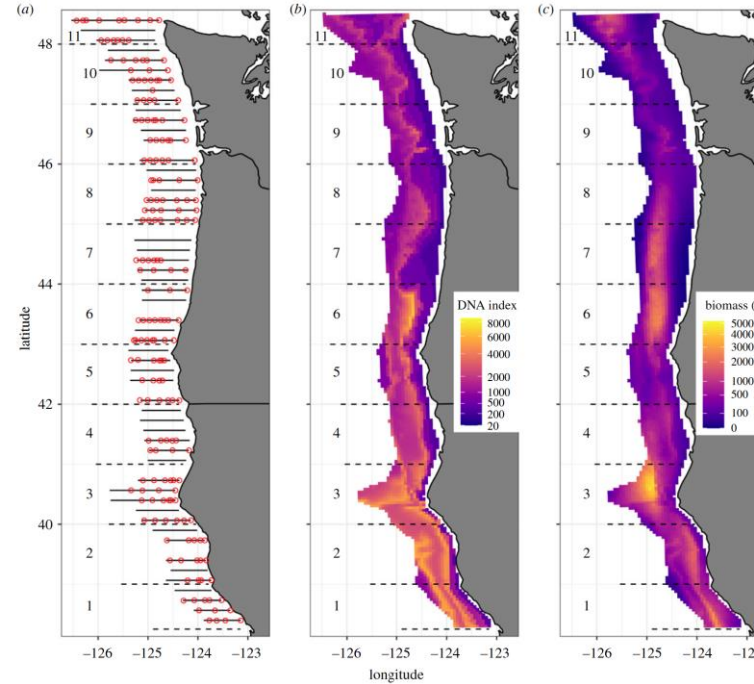
Marine Mammal Remote Detection via Innovative environmental DNA Sampling

3. To apply the lessons learned to map marine mammals and their prey along the US west coast using eDNA.

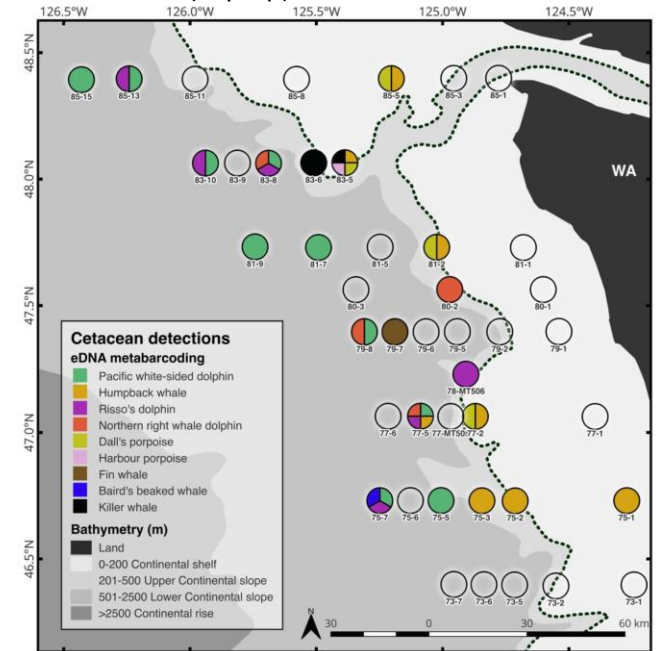


Contact: Dr. Ryan Kelly, rpkelly@uw.edu

Shelton et al (2022) Pro R Soc B



Valdiva et al (in prep)

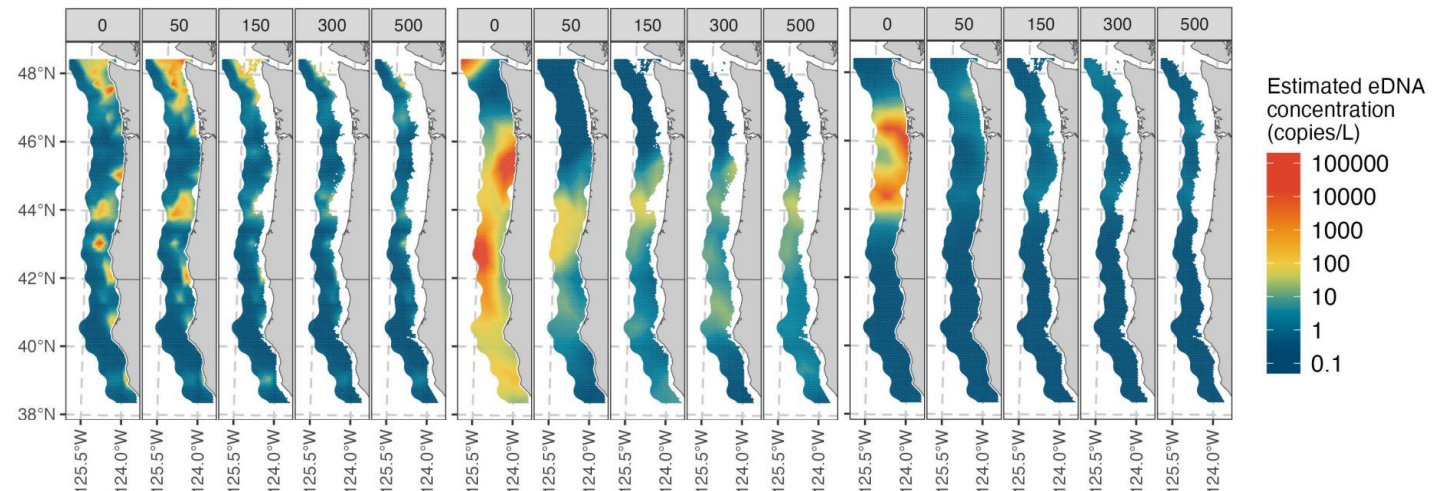


Guri et al (in prep)

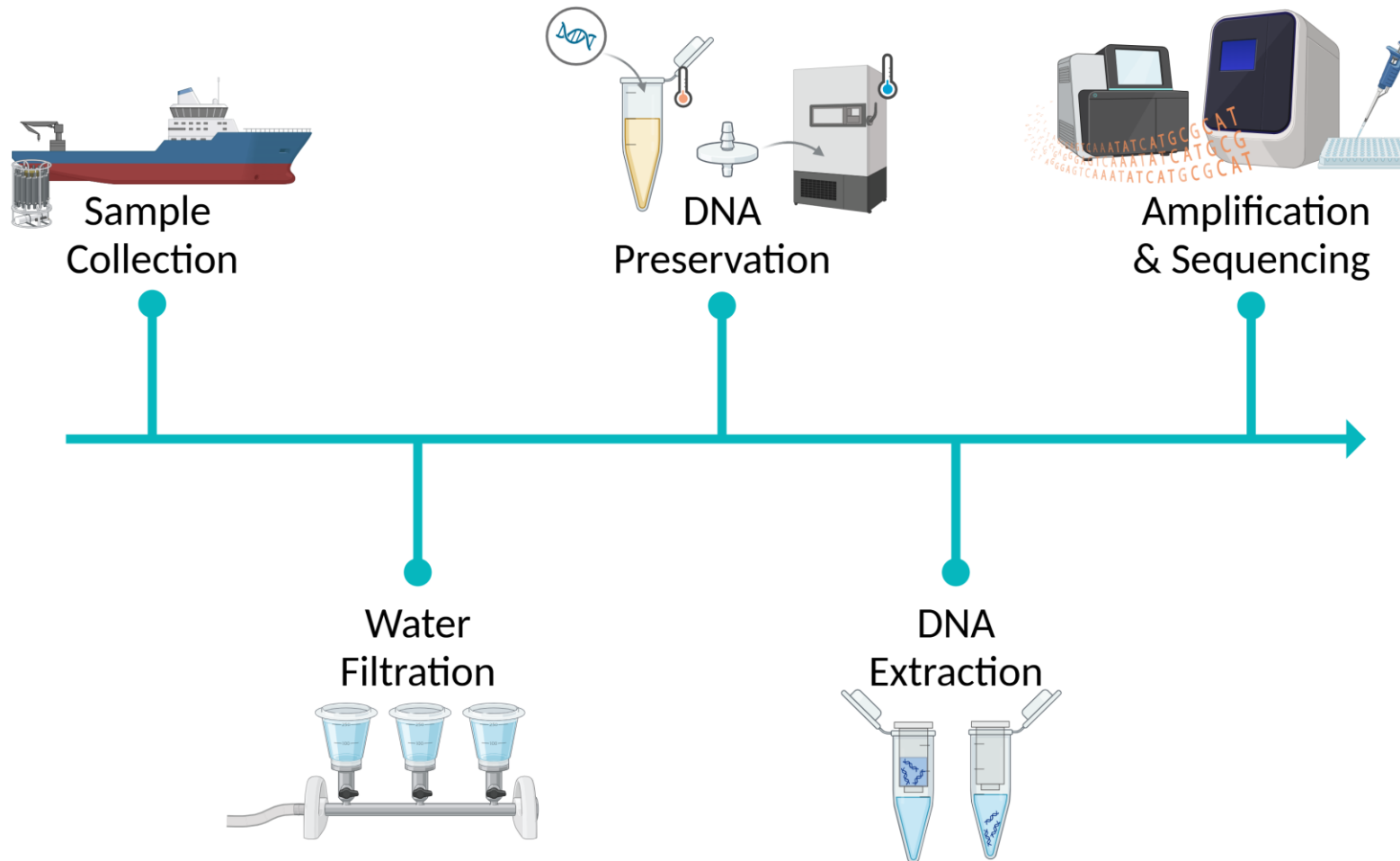
Clupea pallasii

Scomber japonicus

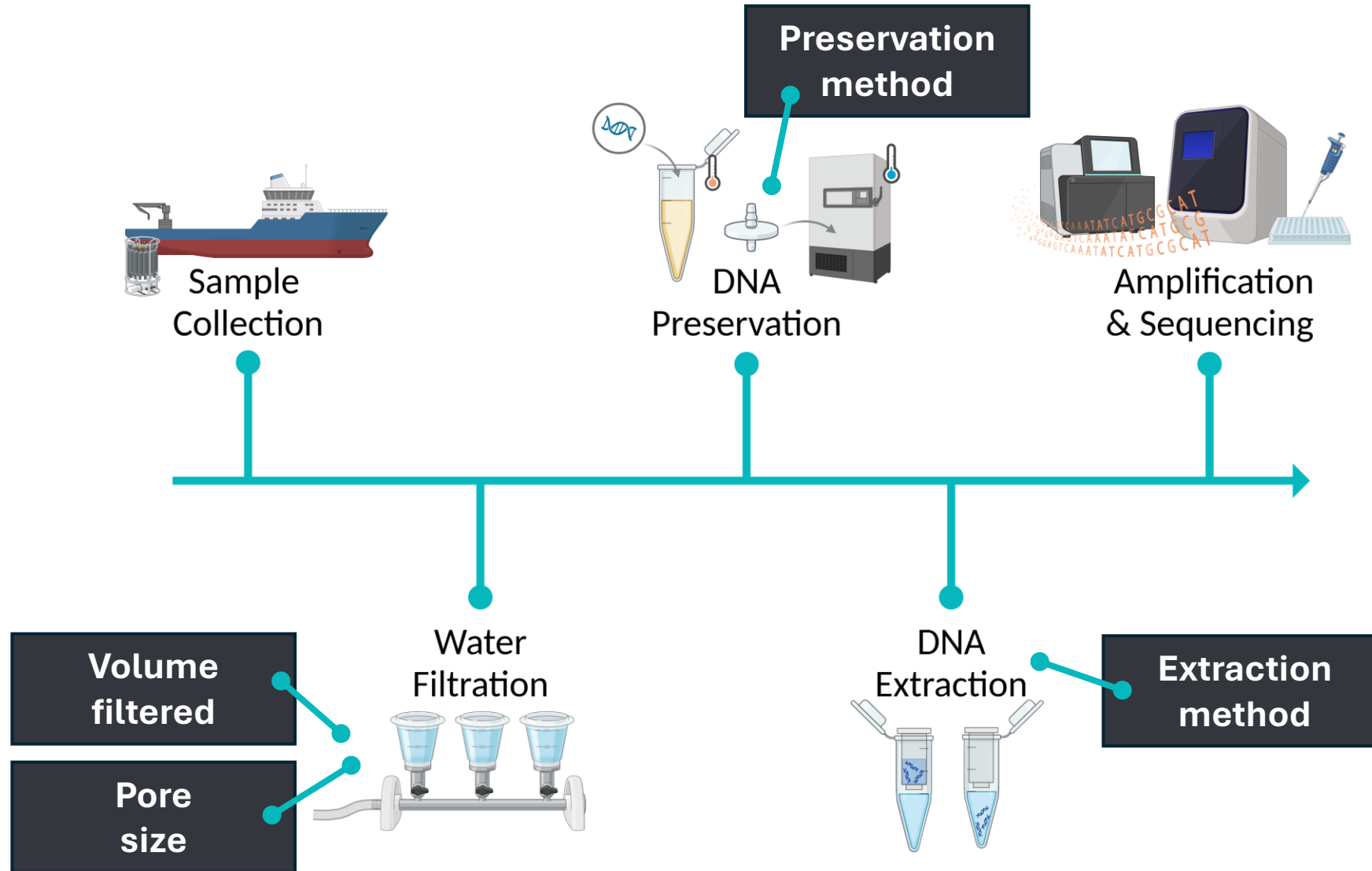
Thaleichthys pacificus



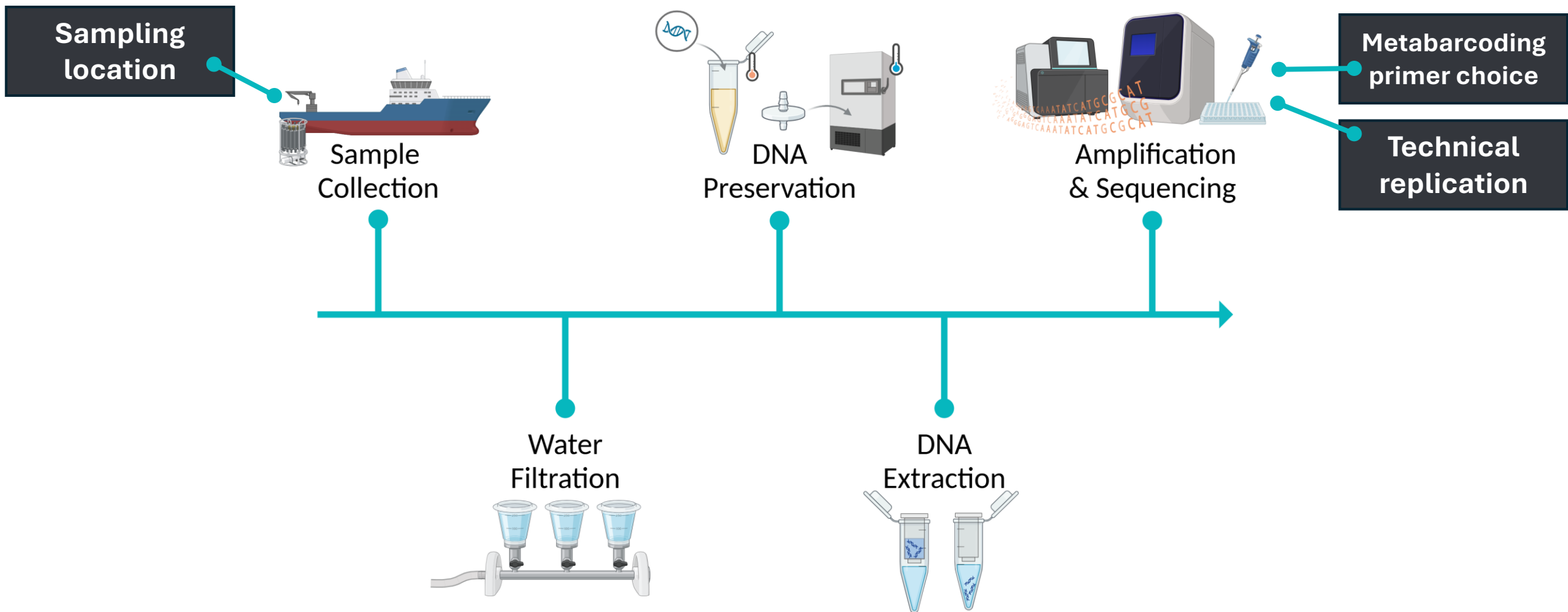
Optimizing a Workflow for Rare Targets



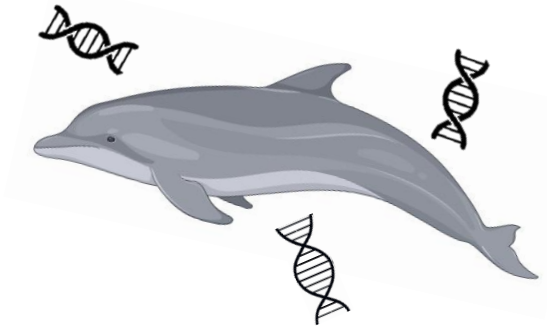
Single-species Context



Multi-species Context



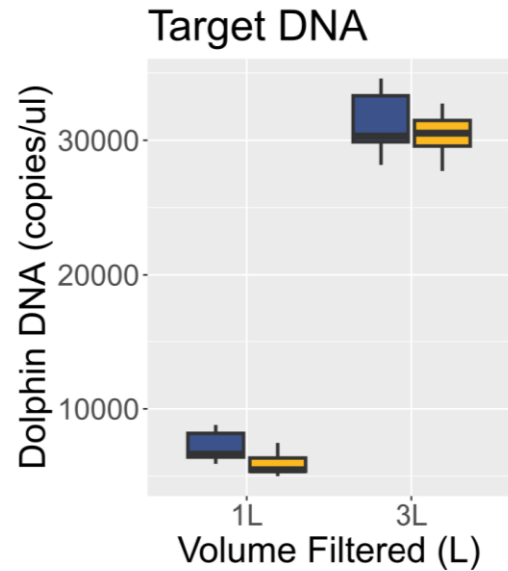
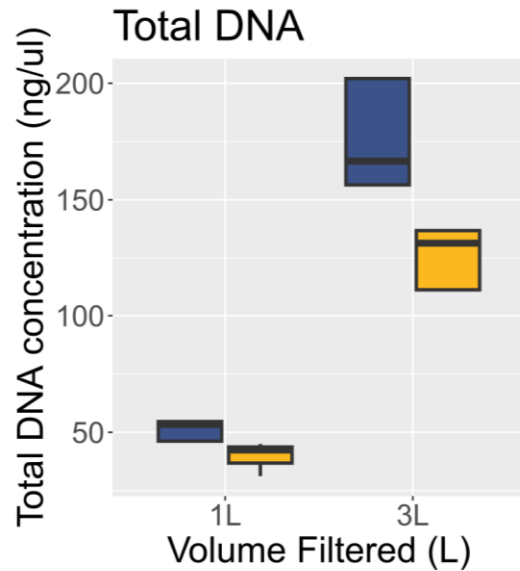
Water Filtration: Volume Filtered and Pore Size



Volume Pore size

- 1 L
- 3 L
- 1 μm
- 5 μm

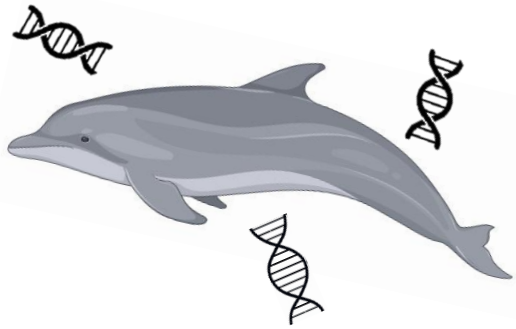
Andruszkiewicz Allan et al (in prep)



Pore Size

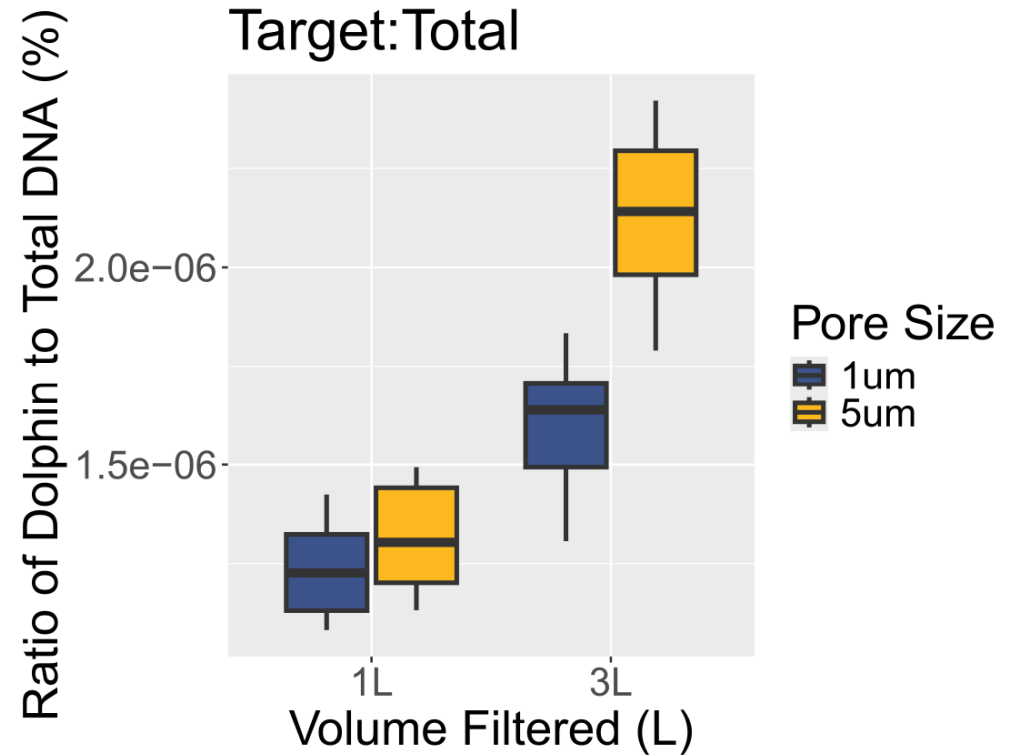
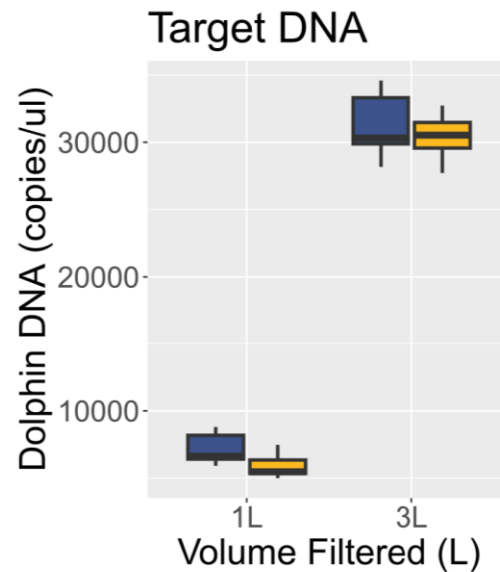
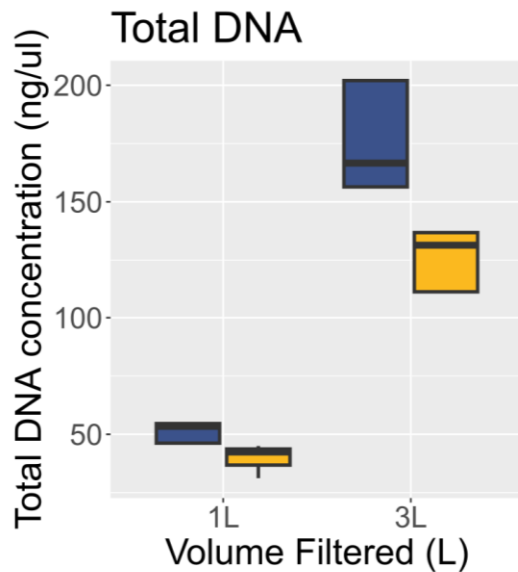
- 1um
- 5um

Water Filtration: Volume Filtered and Pore Size

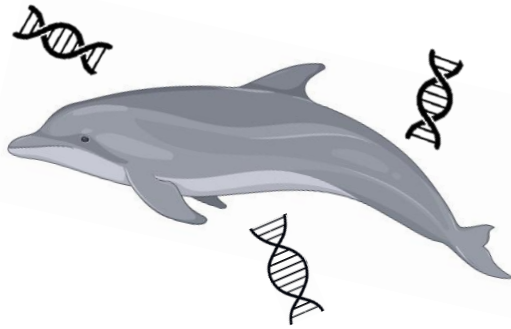


- | Volume | Pore size |
|--------|-------------------|
| • 1 L | • 1 μm |
| • 3 L | • 5 μm |

Andruszkiewicz Allan et al (in prep)



DNA Preservation and Extraction



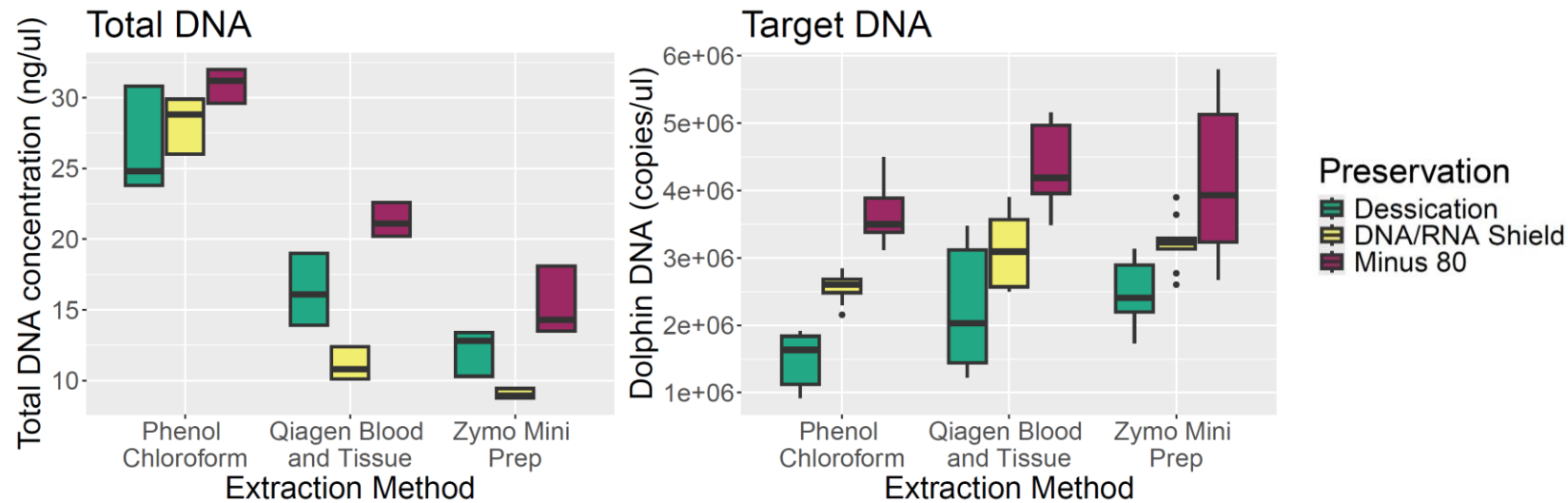
Preservation

- Dessication
- DNA/RNA Shield
- -80°C

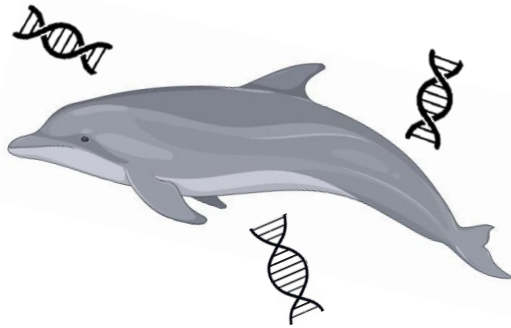
Extraction

- PCI
- Qiagen DNeasy Blood + Tissue
- Zymo Mini Prep Plus

Andruszkiewicz Allan et al (in prep)



DNA Preservation and Extraction



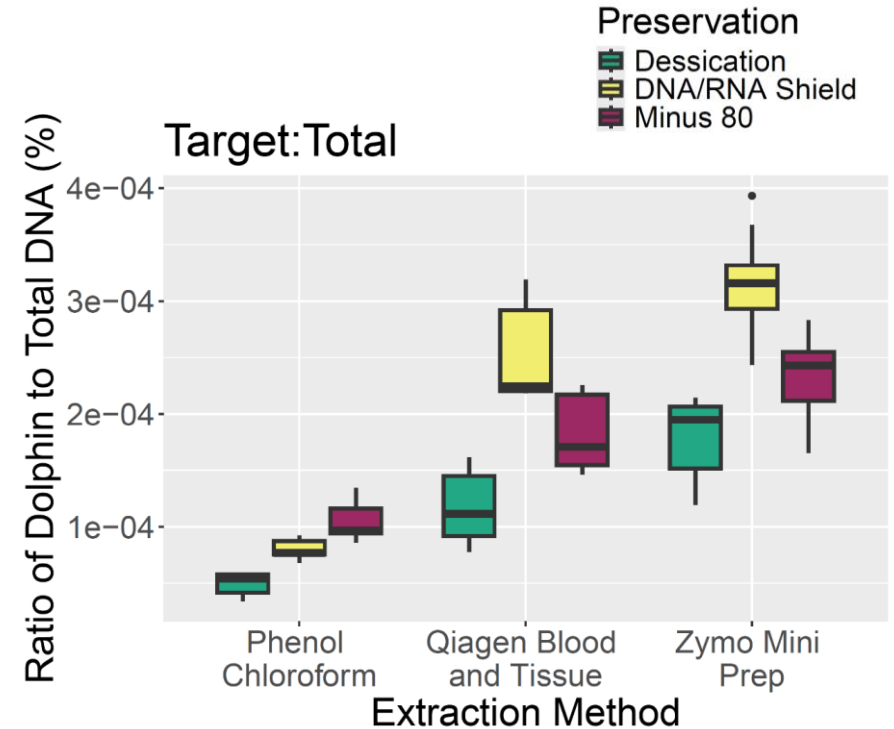
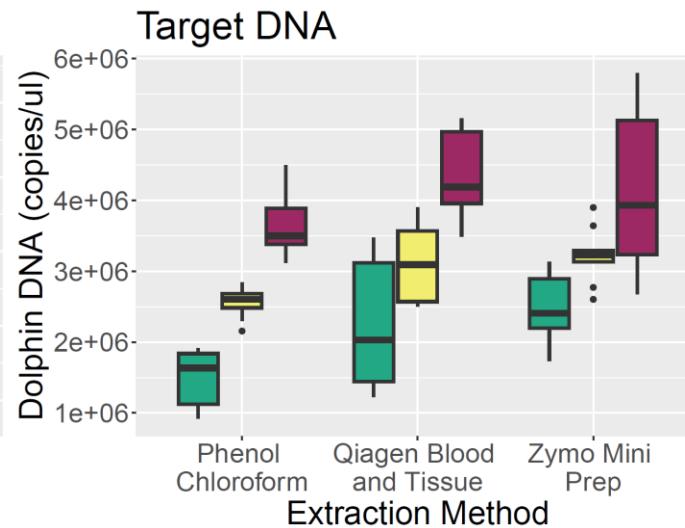
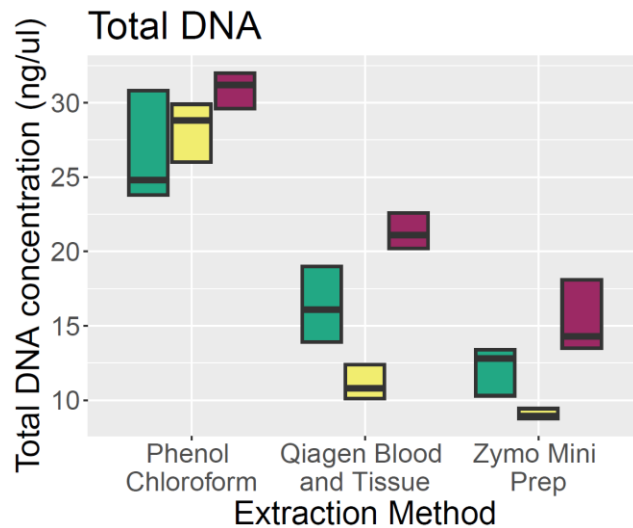
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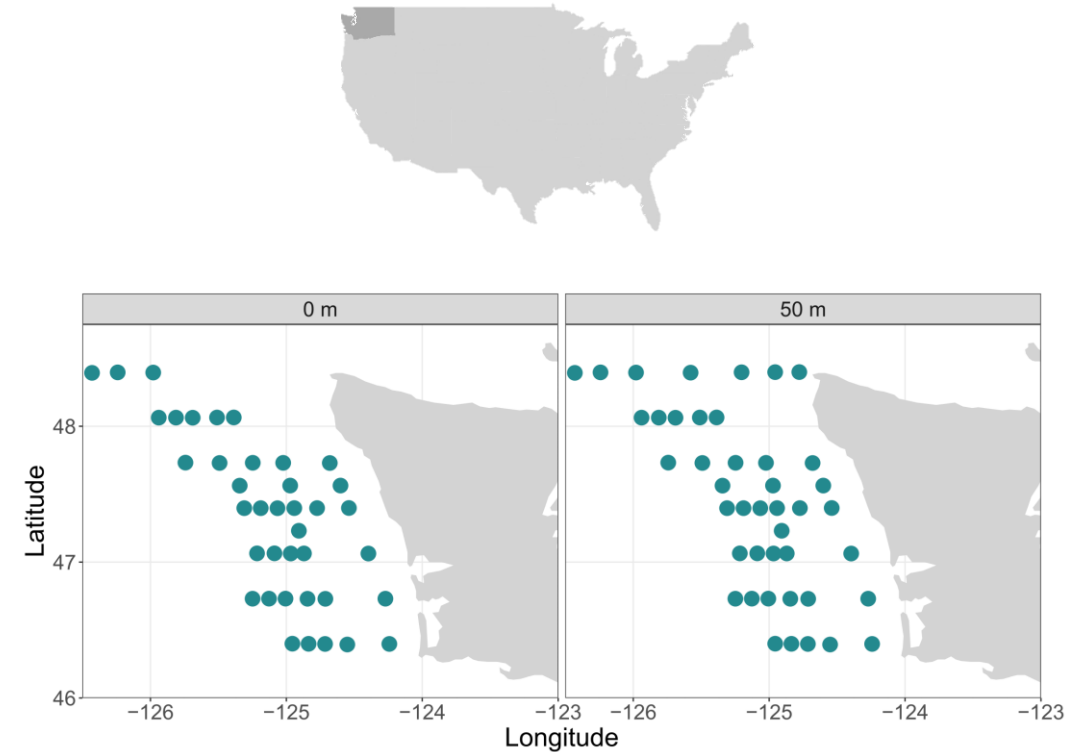
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Andruszkiewicz Allan et al (in prep)



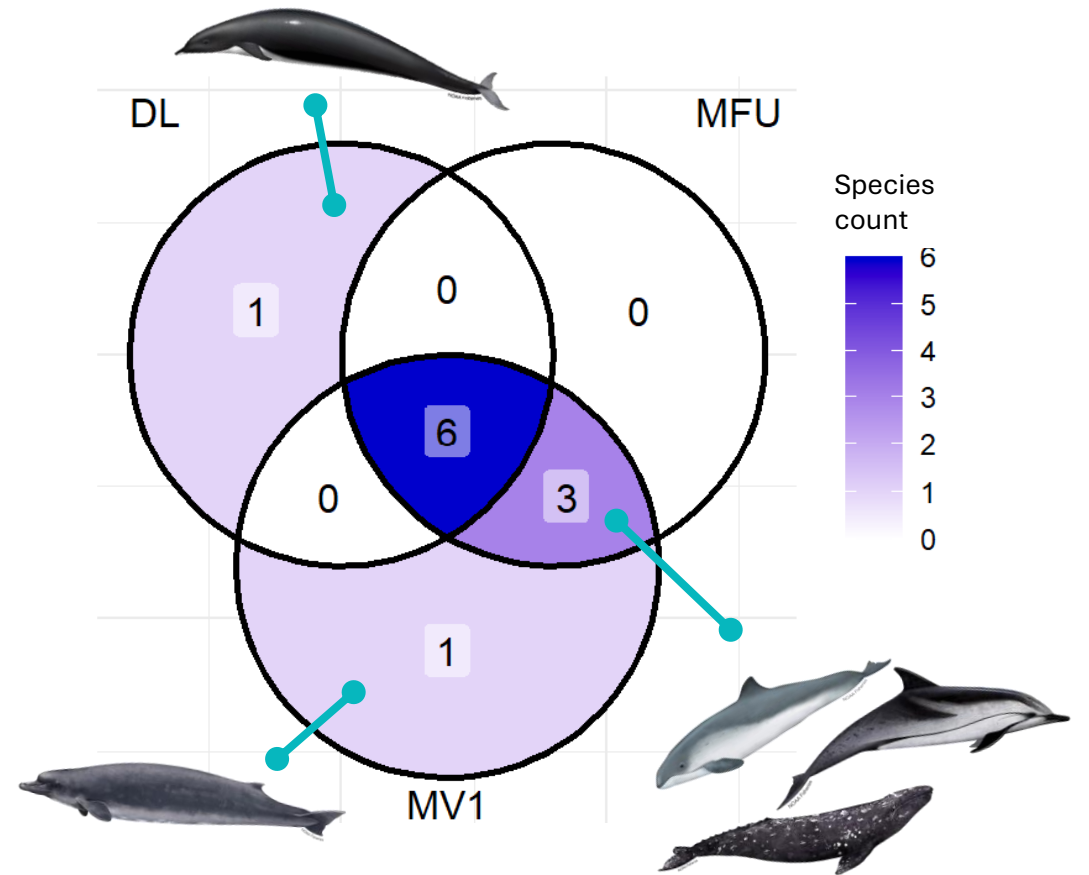
Metabarcoding Primer Choice

Primer set	Gene	Target	Percent samples with cetaceans
MiFish-U <small>Miya et al. 2015</small>	12S	Fish	24.5%
MarVer1 <small>Valsecchi et al. 2020</small>	12S	Vertebrates	45.7%
D-loop <small>Baker et al. 2018</small>	Control Region	Marine mammals	25.5%



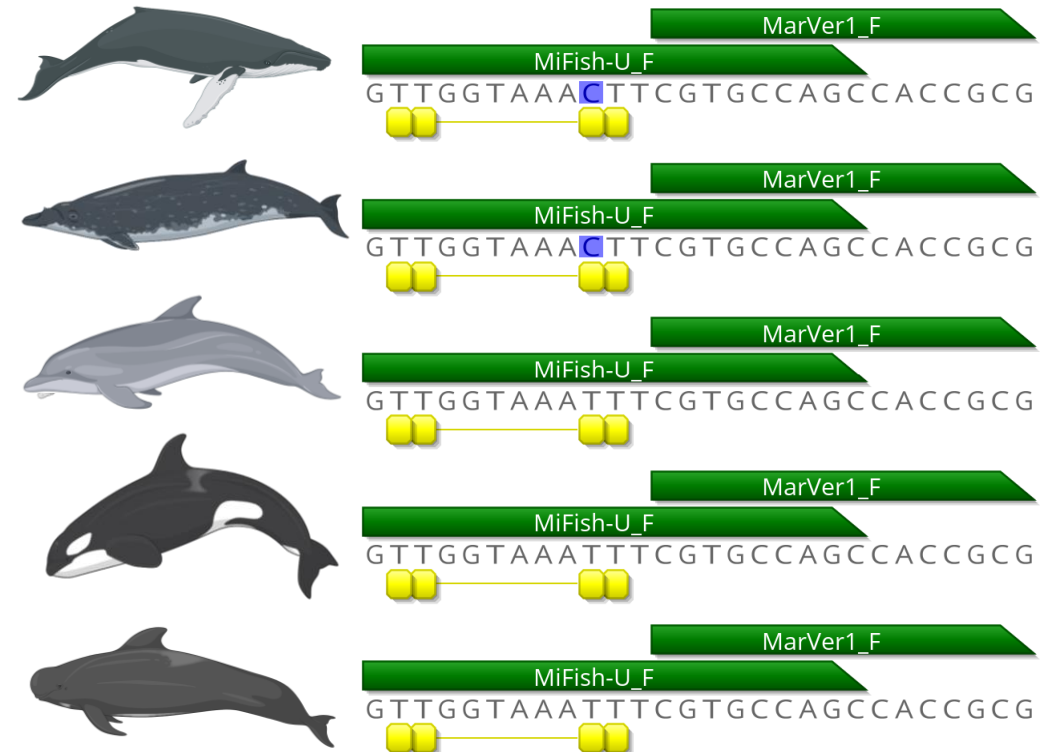
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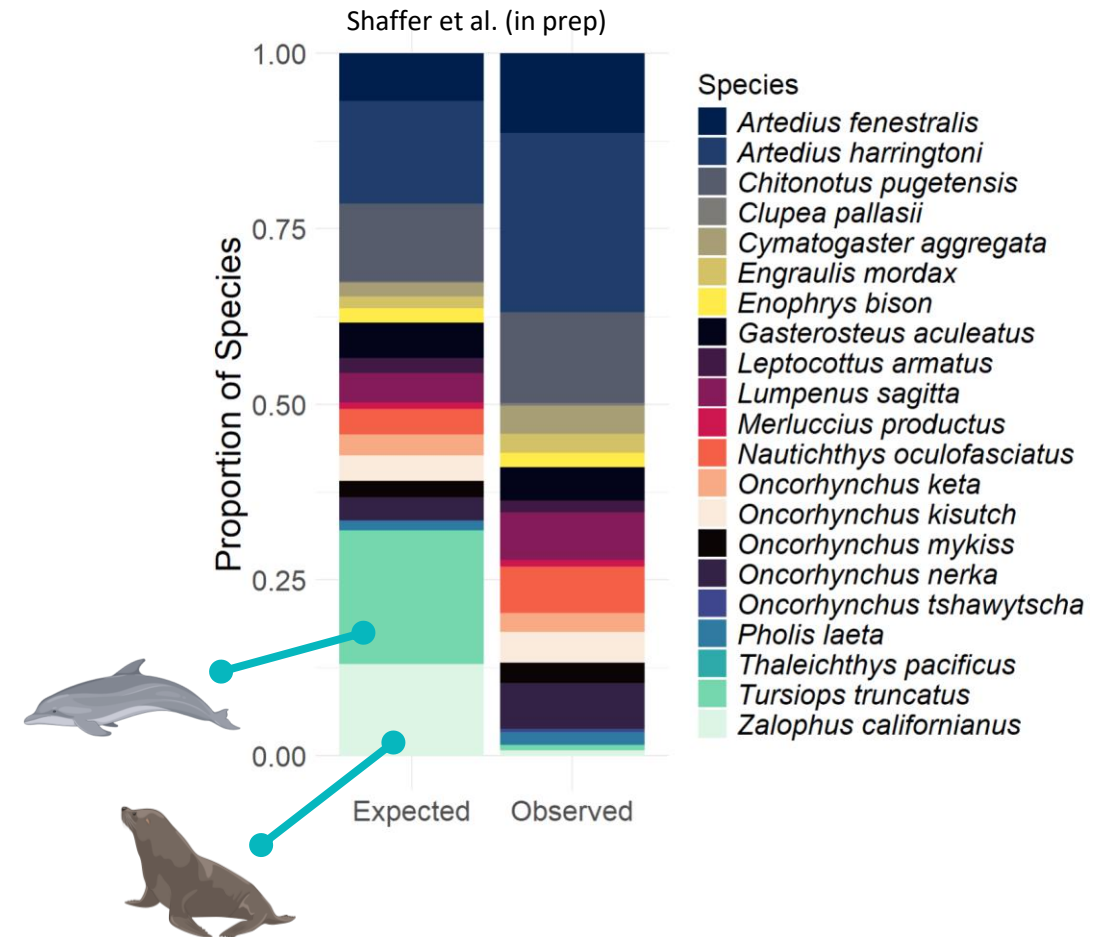
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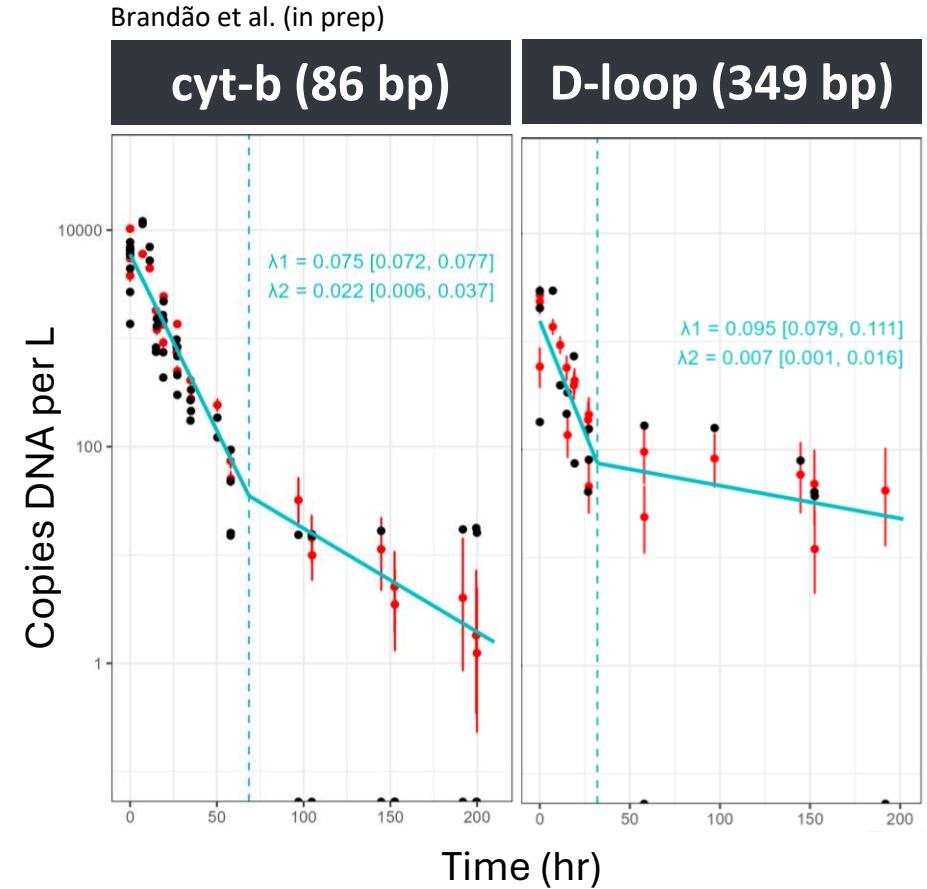


Metabarcoding Primer Choice

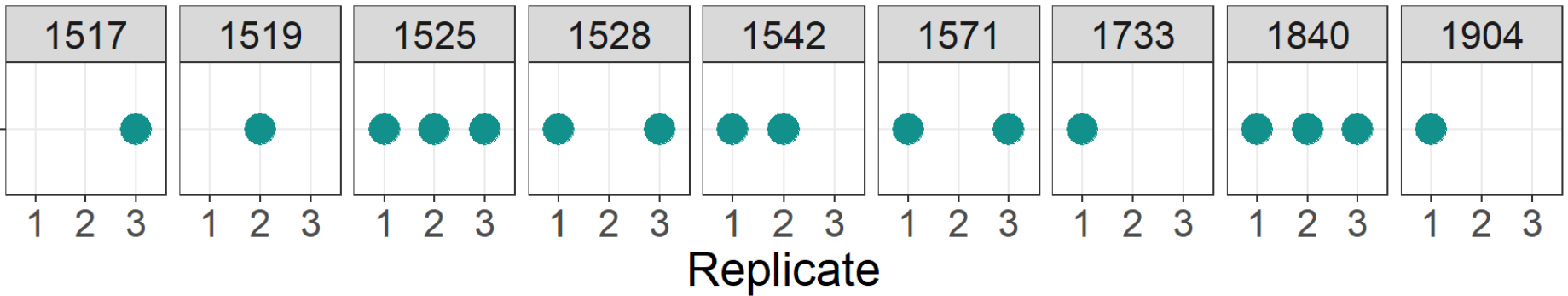
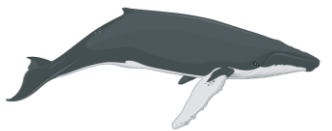
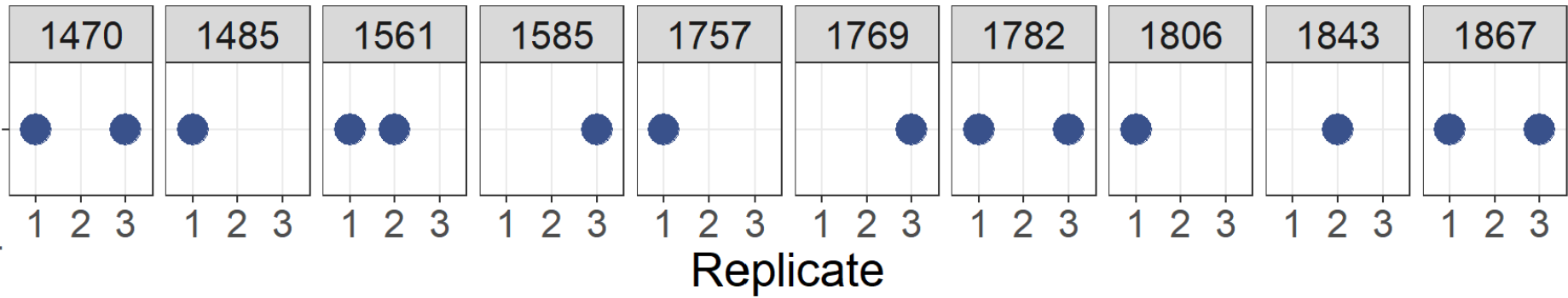
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~160 bp

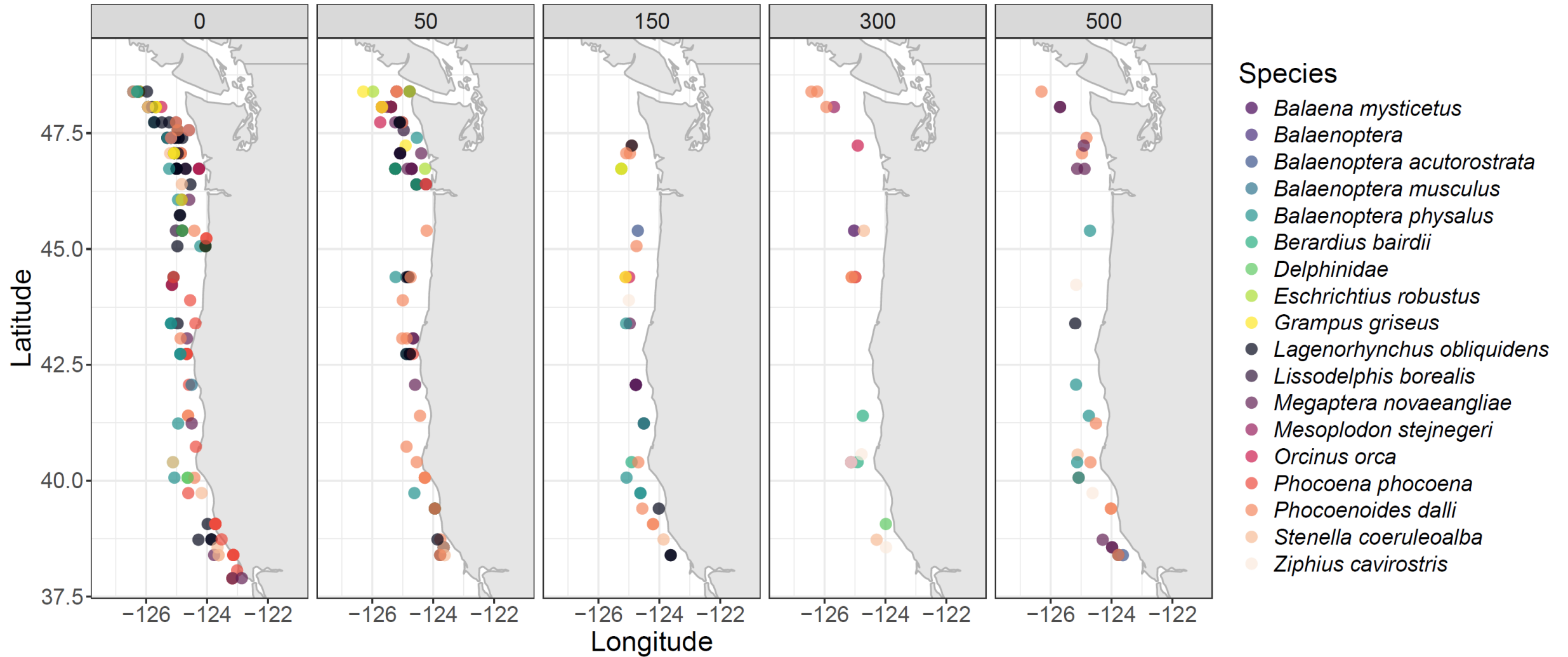
~360 bp



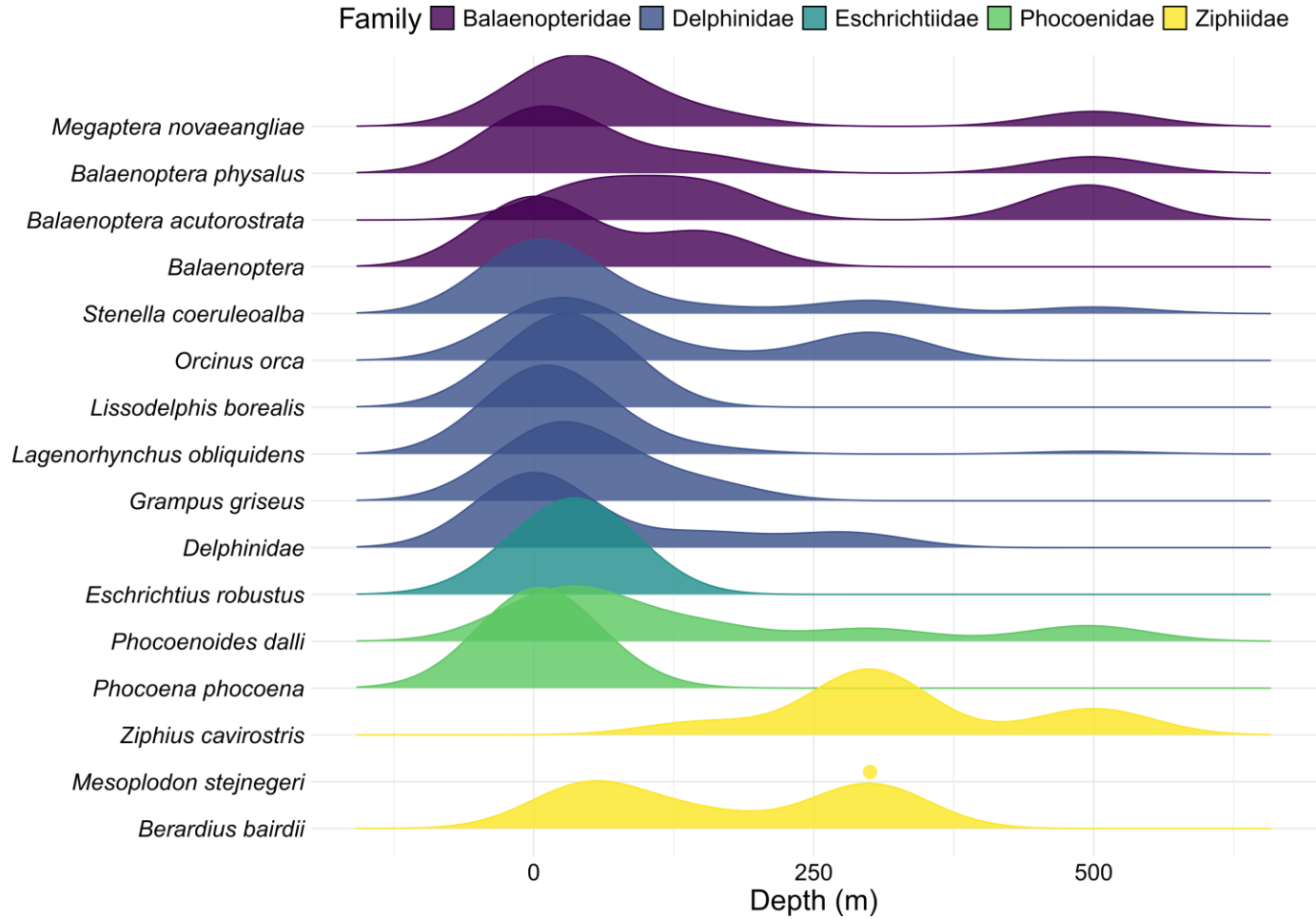
Technical PCR Replication



Sampling Location



Sampling Location



Department of Conservation
Te Papa Atahuri

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World's rarest whale washes ashore in Otago

World's rarest whale washes ashore in Otago

A whale so rare only six specimens have ever been known to science has washed ashore on an Otago beach.

Date: 15 July 2024

On Thursday 4 July, Department of Conservation (DOC) staff were notified a type of beaked whale around 5 m long had washed ashore near Taiari Mouth.

Upon inspection, and consultation with marine-mammal experts from DOC and Te Papa, it appears the creature was a male spade-toothed whale – a species so rare next to nothing is known about them.

DOC Coastal Otago Operations Manager Gabe Davies says if confirmed, this will be a very significant scientific find.

"Spade-toothed whales are one of the most poorly known large mammalian species of modern times. Since the 1800s, only 6 samples have ever been documented worldwide, and all but one of these was from New Zealand. From a scientific and conservation point of view, this is huge."

Lessons learned

Choices along the workflow can greatly increase ability to detect rare targets

For cetaceans in our study system:

- We maximized the target to total ratio with larger pore sizes, larger volumes and kit extractions
- We found metabarcoding primer sets have different sensitivity and can yield different information
- We increased replication because rare things are stochastically detected
- We found eDNA where the animals spend more of their time (surface v depth)



Acknowledgments

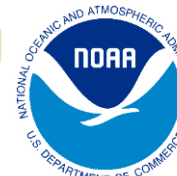
ONR: Mike Weise, Sarah Weiss

MMARINeDNA team: Kate Bertko, Ally Im, Cara Sucher, Len Thomas, Eiren Jacobson, Brice Semmens, Simone Baumann-Pickering, Julie Dinasquet, Michaela Alksne, Bryce Ellman, Nastassia Patin, Erin Satterthwaite, Shannon Dolan, Noelle Bowlin

NOAA NWFSC: Abigail Wells, Ana Ramón Laca, Samantha Engster, Steph Moore, Nicholas Adams

MBARI: Kevan Yamahara

UW Kelly Lab: Aden Ip, Olivia Scott, Stephanie Matthews



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