Developing fish abundance and ecosystem indices from environmental DNA (eDNA)

A.O. Shelton, R.P. Kelly, M.F. Shaffer, K. Nichols,
D. Baetscher, A. Rovellini, A. Berger
Northwest Fisheries Science Center
10/7/2026

Thanks

Northwest Fisheries Science Center

Genetics and Evolution

- Krista Nichols
- Linda Park
- Kim Parsons
- Abi Wells
- Ana Ramon-Laca
- Samantha Engster
- Megan Shaffer
- Madison Wells
- Meghan Parsley
- Ramón Gallego
- Jimmy O'Donnell
- Piper Schwenke

FEAT

- Julia Clemons
- John Pohl
- Rebecca Thomas
- Dezhang Chu
- Hake cruise team
- Sandy Parker-Stetter

Ecosystem Science

- Jameal Samhouri
- Blake Feist
- Eric Ward
- Owen Liu

Other NOAA

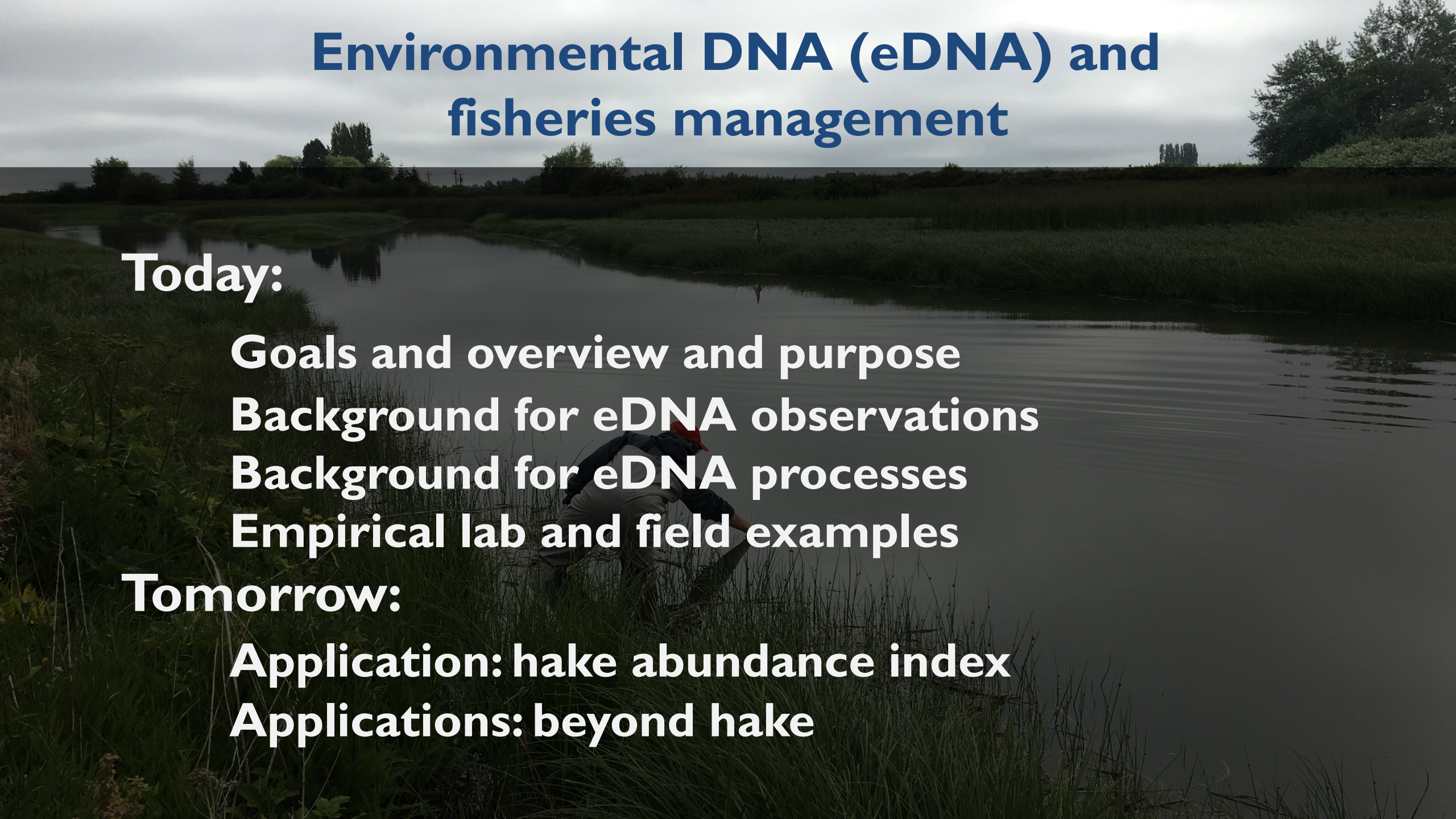
- Kelly Goodwin
- Rick Methot
- AFSC: Diana Baetscher, Kim Ledger
- PIFSC: Jon Whitney, Jamison Gove

University of Washington

Kelly Lab & eDNA Collaborative

- Ryan Kelly
- Elizabeth Andruszkiewicz Allen
- Gled Guri
- Shannon Kachel
- Albi Rovellini

Environmental DNA (eDNA) and fisheries management



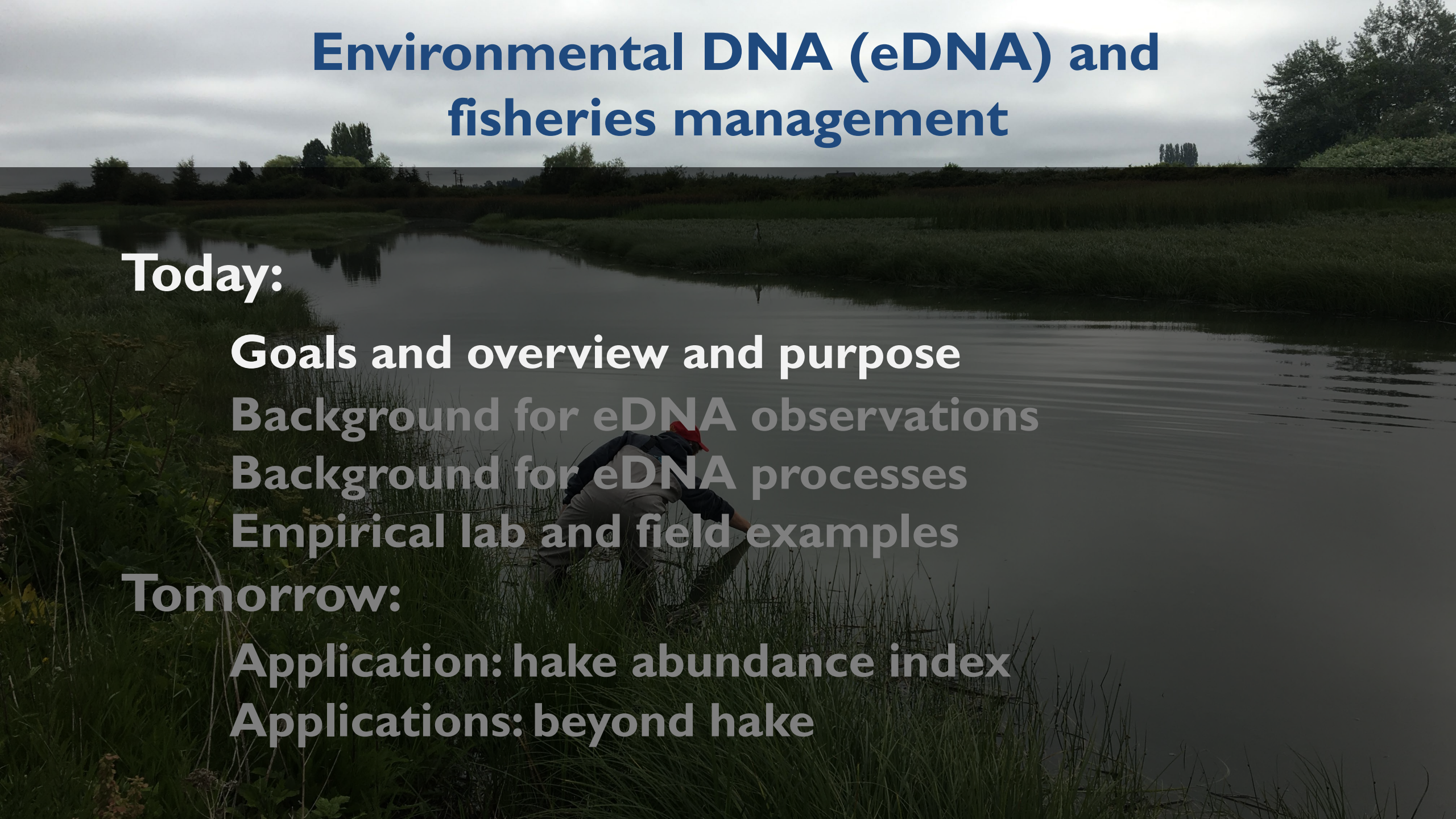
Today:

- Goals and overview and purpose
- Background for eDNA observations
- Background for eDNA processes
- Empirical lab and field examples

Tomorrow:

- Application: hake abundance index
- Applications: beyond hake

Environmental DNA (eDNA) and fisheries management



Today:

- Goals and overview and purpose
- Background for eDNA observations
- Background for eDNA processes
- Empirical lab and field examples

Tomorrow:

- Application: hake abundance index
- Applications: beyond hake

A very short primer on eDNA

Everything sheds "stuff" as it lives in the environment

- skin, slime, excrement, etc.

DNA and associated cells degrade relatively quickly in the liquid environment

- scale of hours to days

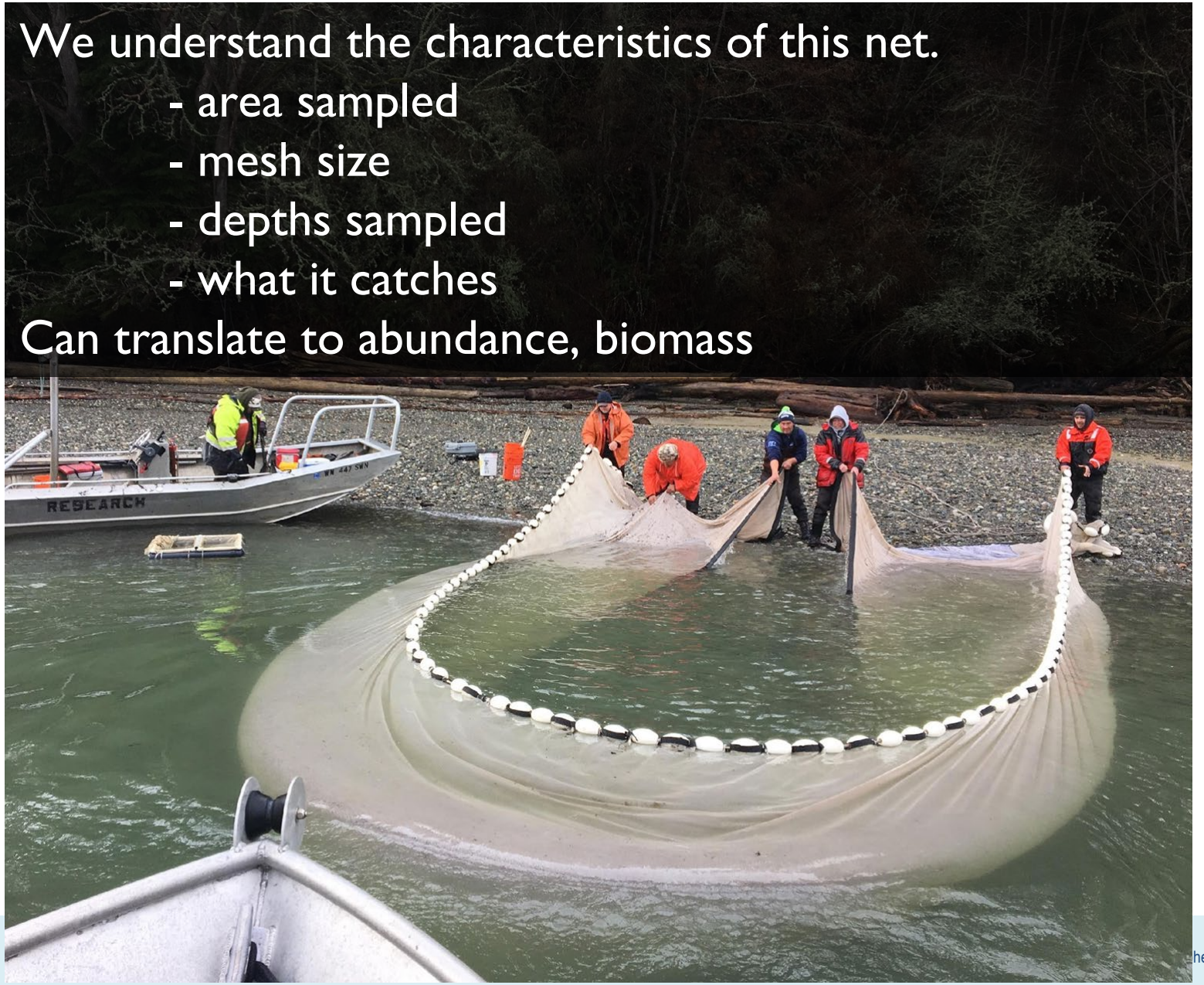
Samples of the DNA collected from water should reveal what is nearby

Maybe we can survey creatures without having to actually catch or see them?

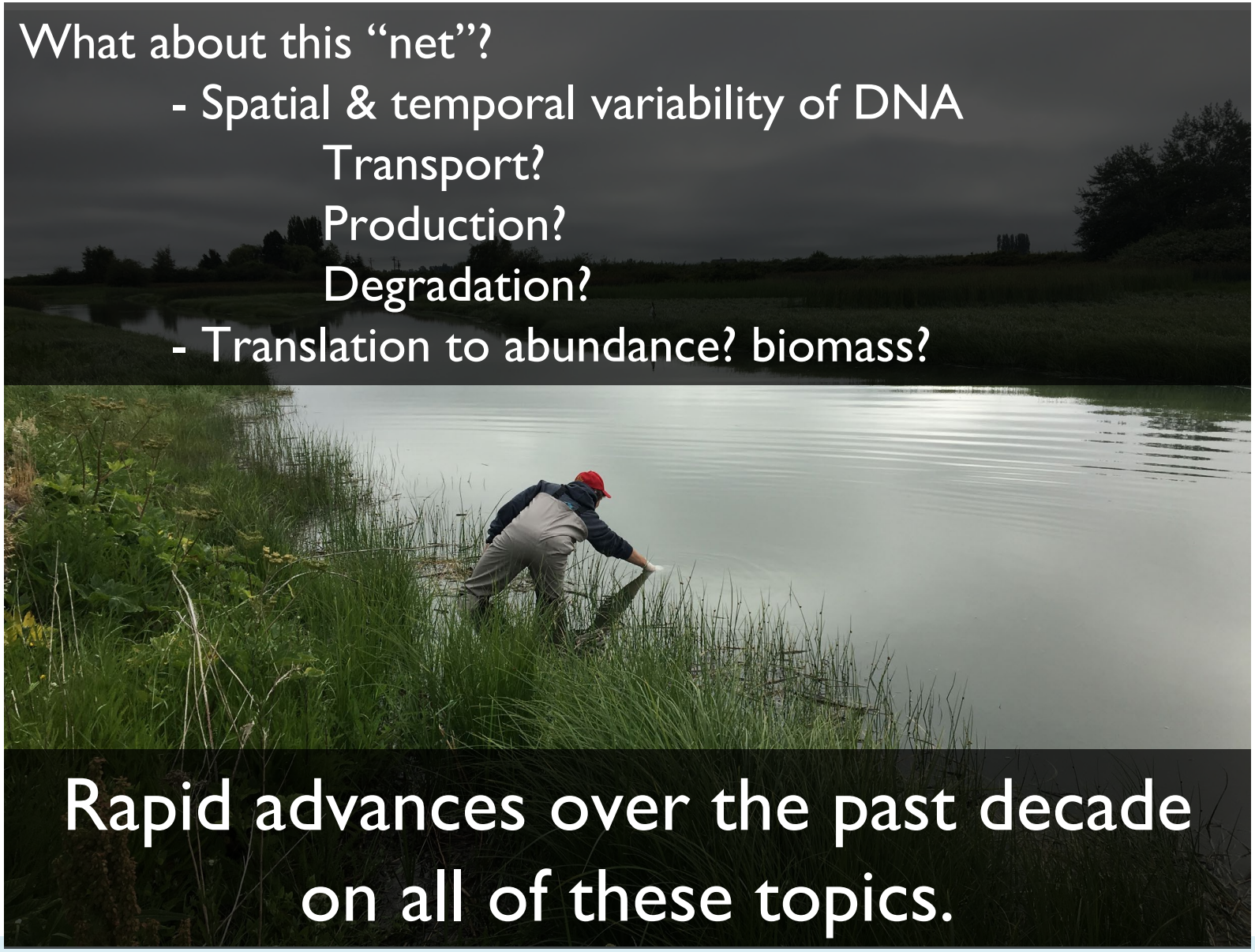




eDNA as a new kind of net



eDNA as a new kind of net



Goals for this review

Present an overview of the state of the science

Suggest potential avenues for incorporating eDNA information into the PFMC process

Request feedback on areas for improvement / further research

A starting point

Ecological Applications, 26(6), 2016, pp. 1645–1659
© 2016 by the Ecological Society of America

A framework for inferring biological communities from environmental DNA

Andrew Olaf Shelton,[1,4] James Lawrence O'Donnell,[2] Jameal F. Samhouri,[1] Natalie Lowell,[2] Gregory D. Williams,[3] and Ryan P. Kelly[2]

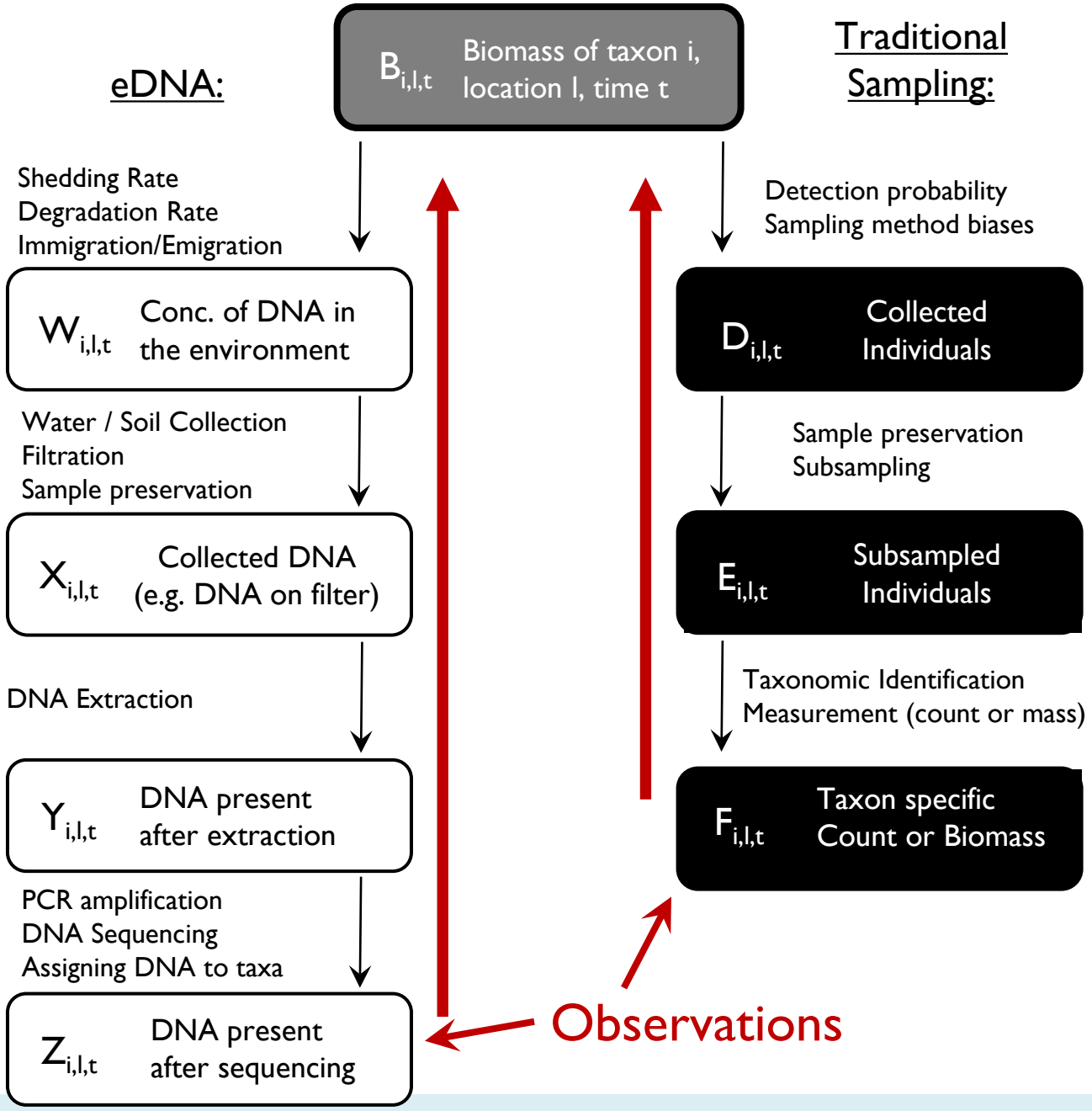
1. eDNA observations are really not that different from most other fisheries data sources
2. eDNA observations are inherently quantitative.
3. We don't have to understand all DNA processes to provide useful fisheries advice.

2016

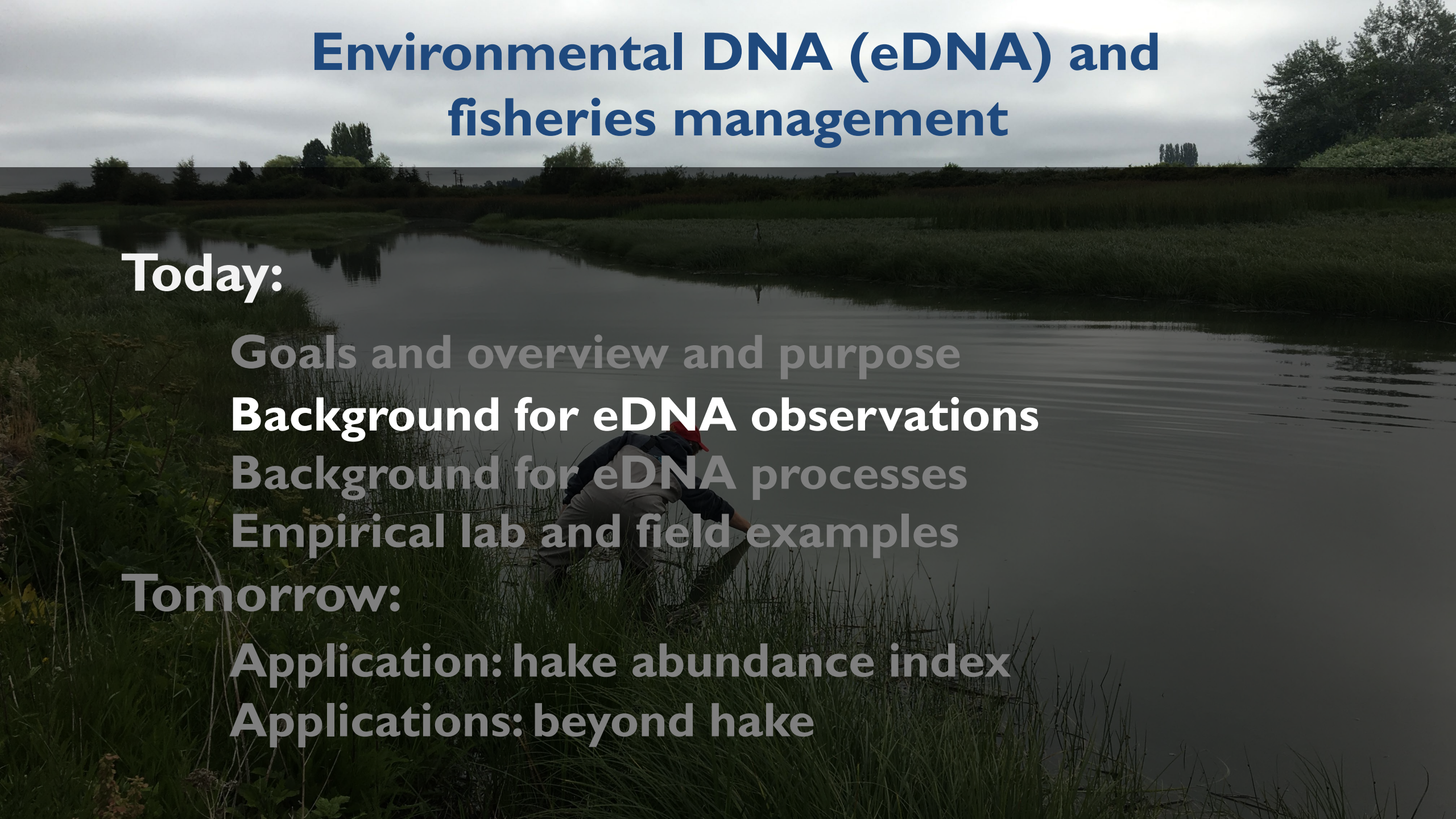
Ecological Applications, 26(6), 2016, pp. 1645–1659
© 2016 by the Ecological Society of America

A framework for inferring biological communities from environmental DNA

Andrew Olaf Shelton,[1,4] James Lawrence O'Donnell,[2] Jameal F. Samhouri,[1] Natalie Lowell,[2] Gregory D. Williams,[3] and Ryan P. Kelly[2]



Environmental DNA (eDNA) and fisheries management



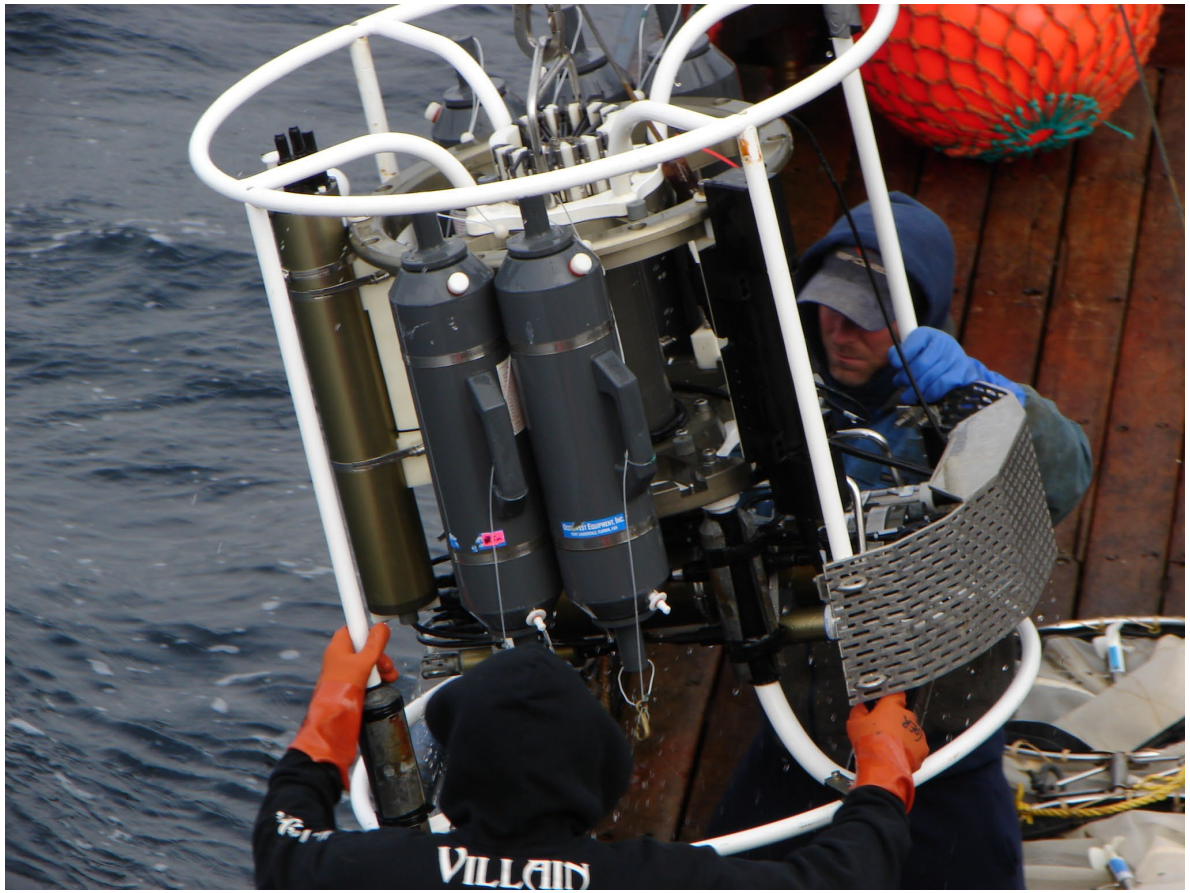
Today:

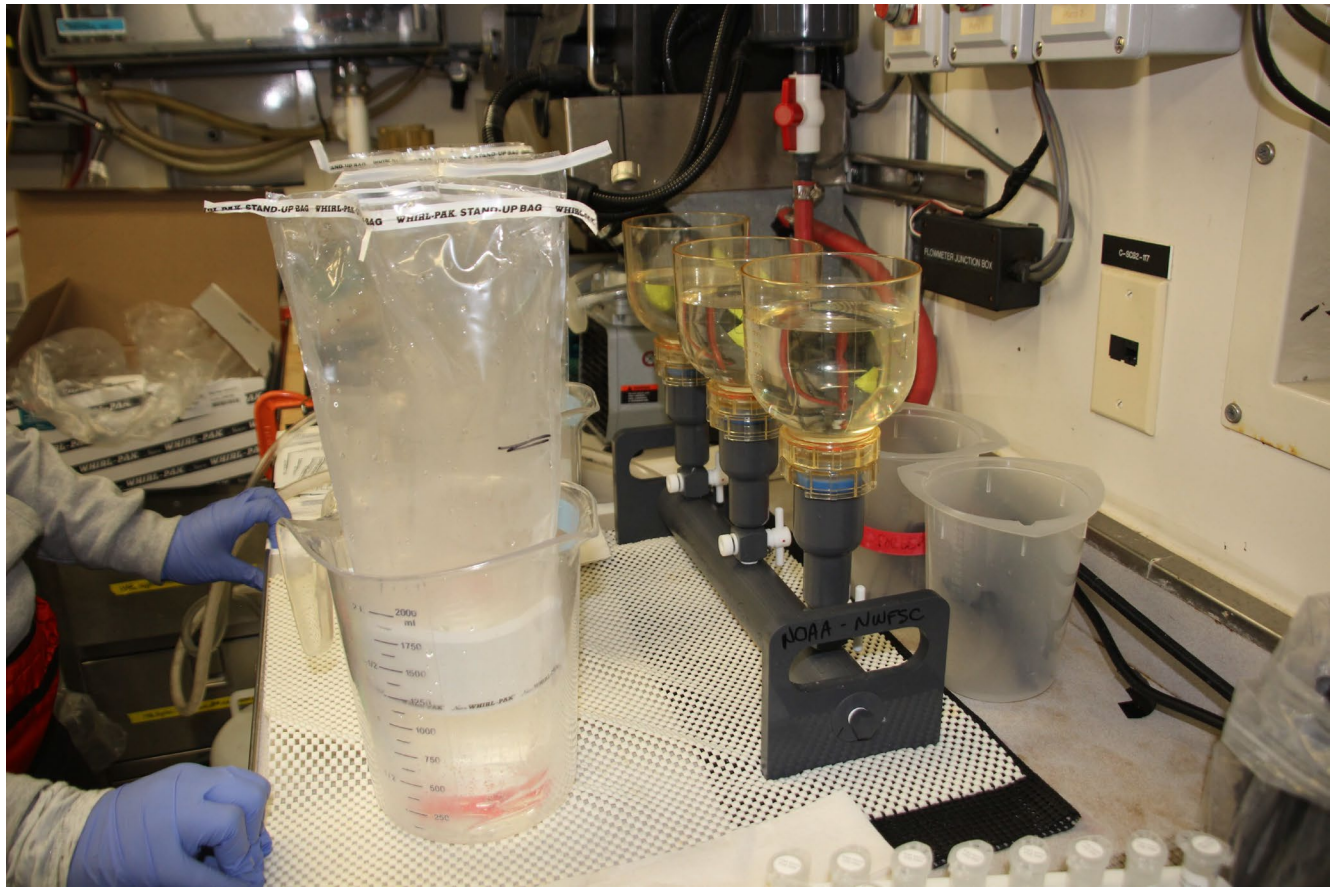
- Goals and overview and purpose
- **Background for eDNA observations**
- Background for eDNA processes
- Empirical lab and field examples

Tomorrow:

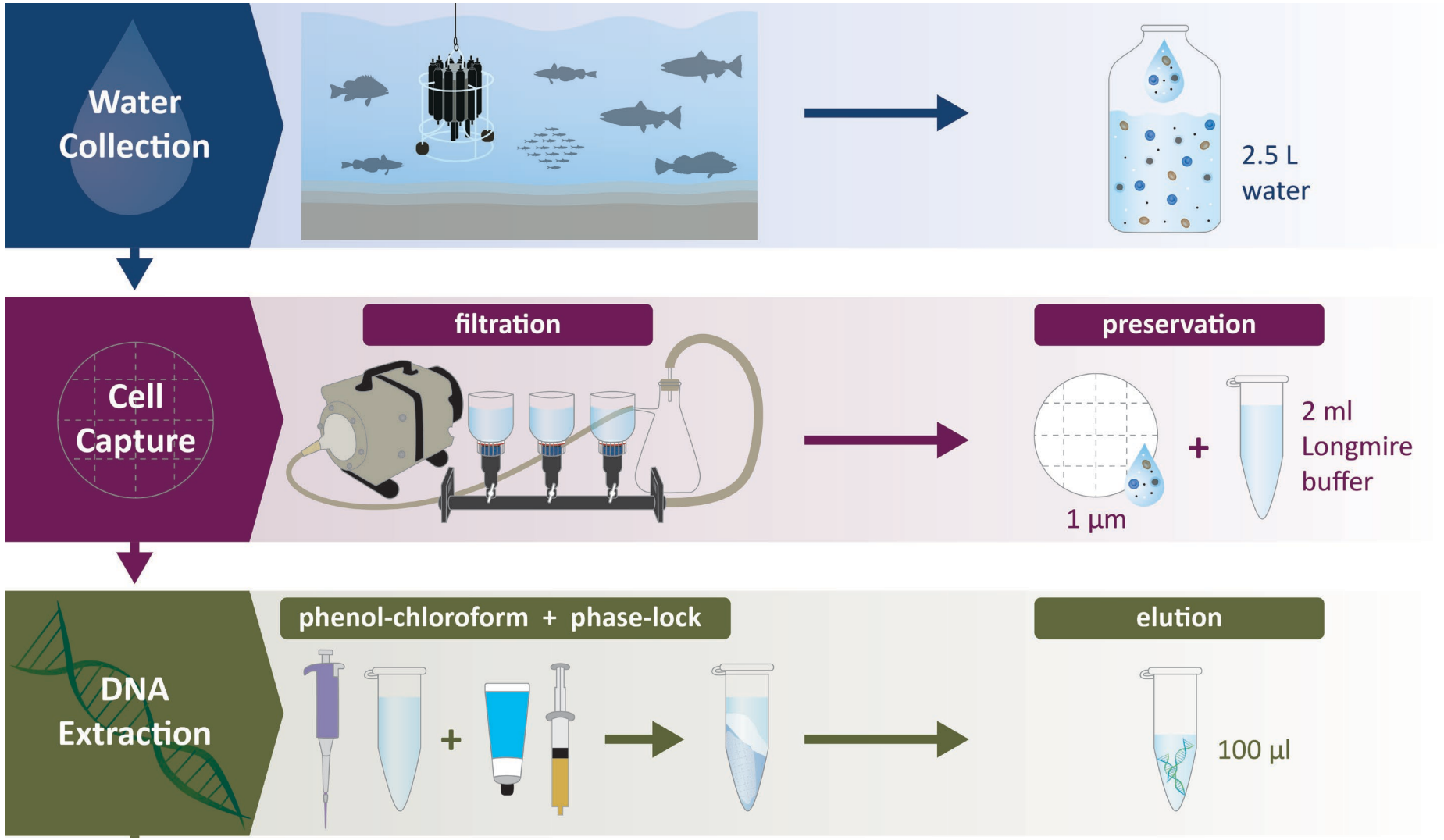
- Application: hake abundance index
- Applications: beyond hake

What are eDNA observations and how do we collect them?





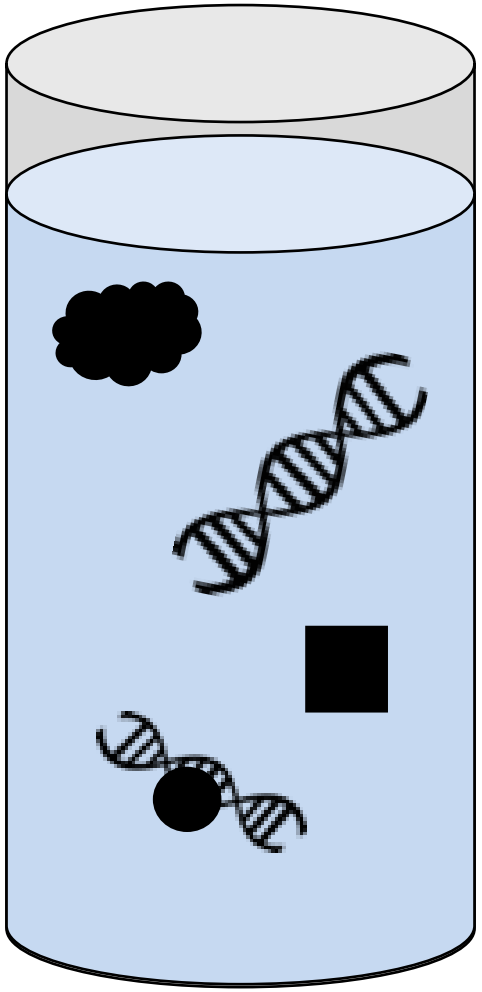
eDNA Basics



What are we capturing?

What is eDNA?

A soup of information from many taxa

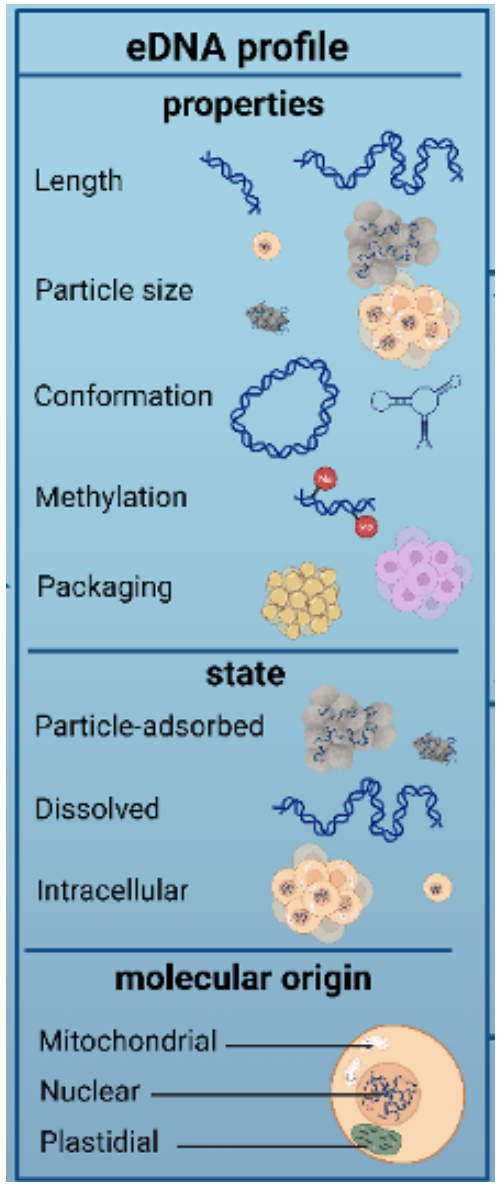


Skin and scales

Tissue

Metabolic waste

Free DNA / RNA / Proteins



Target cell-associated DNA

- filter pore size
- avoid swamping eukaryote DNA with bacterial DNA

Target mitochondrial DNA

- primer choices
- choose relatively short, informative DNA fragments

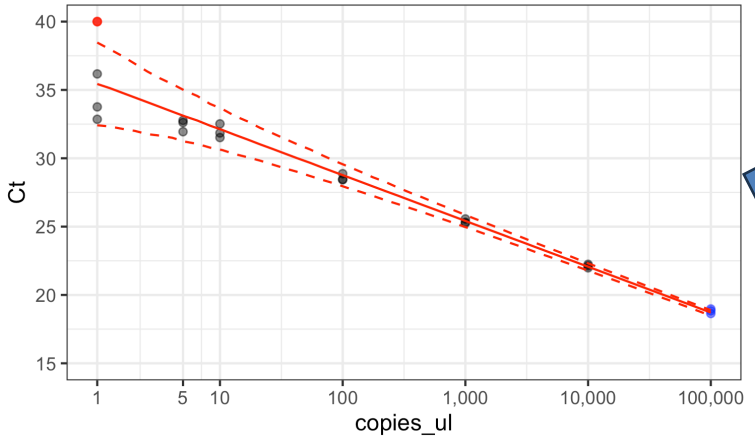
Once a sample is in hand, two options

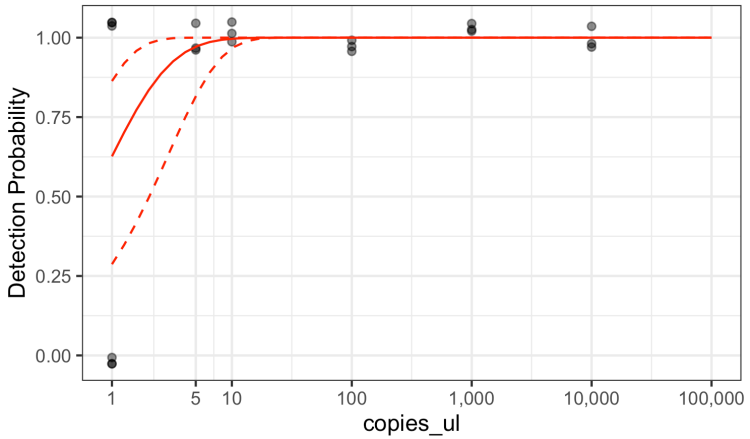
1. Amplify (select for) DNA using a primer that matches one taxon.
   - **quantitative PCR (qPCR),** digital droplet PCR (ddPCR)
   - relatively straightforward lab methodology
   - provides a direct measurement of DNA concentration for a single taxon

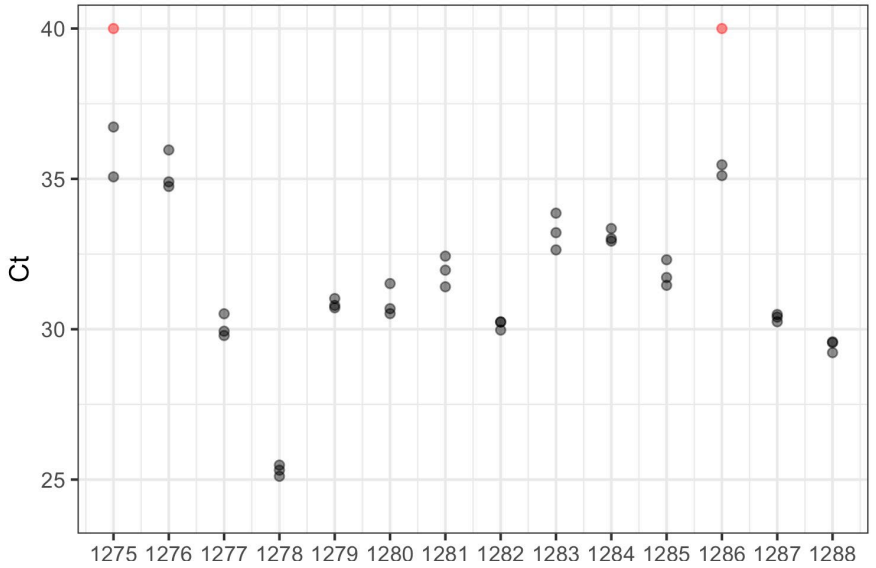
2. Amplify using a DNA primer that matches more than one taxa.
   - **Metabarcoding.**
   - complicated laboratory protocols involving DNA sequencing.
   - complicated bioinformatics post-processing.
   - provides relative sequence abundance information

qPCR in pictures

Measure fluorescence of samples of known concentration

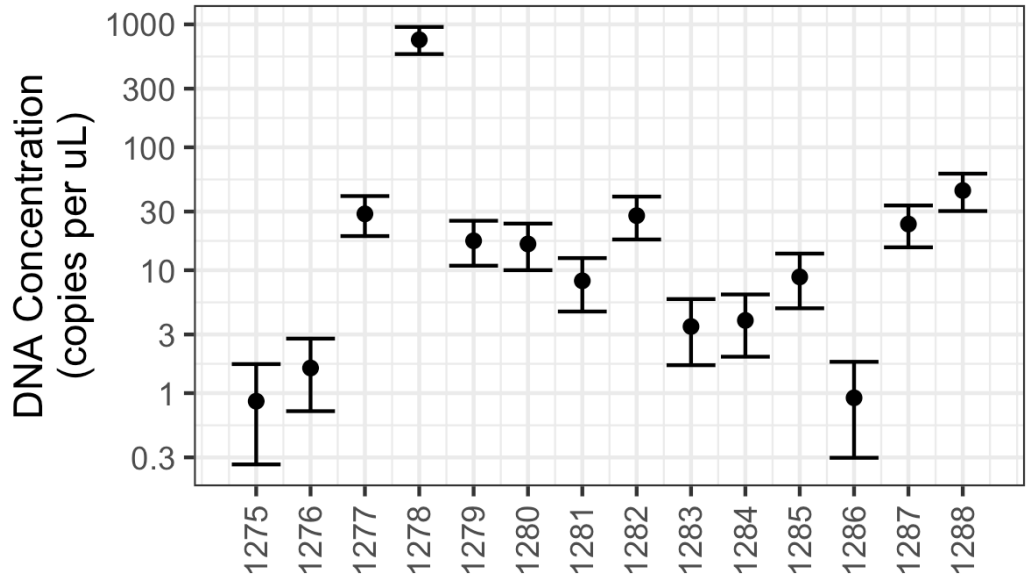






Measure fluorescence of unknown samples

Translate fluorescence to DNA concentration



Metabarcoding Workflow

1 Sample collection

2 DNA extraction

3 PCR amplification

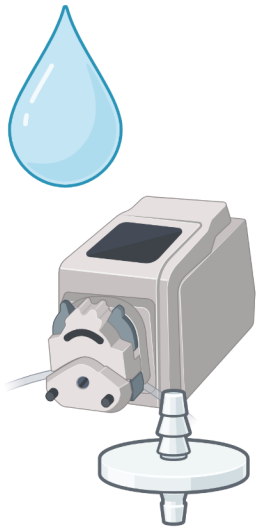
4 Library preparation

5 High-throughput sequencing

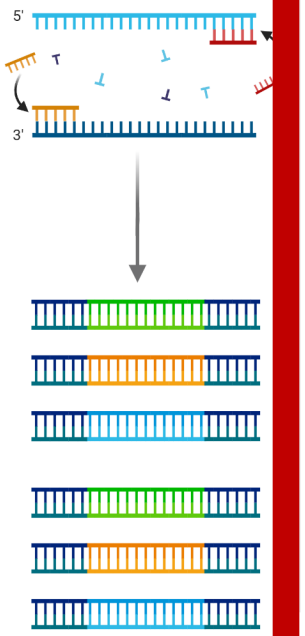
6 Bioinformatic processing

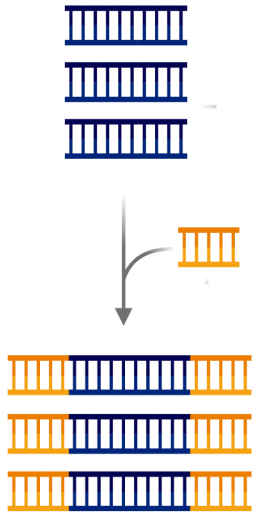
7 Taxonomic assignment

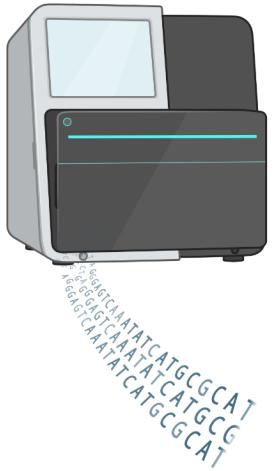
8 Data analysis











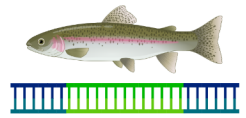


Demultiplex by sample; trim adapters and primers

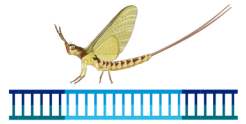
Filter by quality; merge paired-end reds

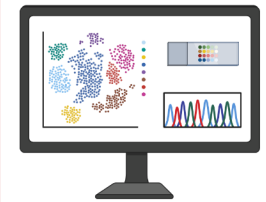
Denoise; remove chimeras

ASVs





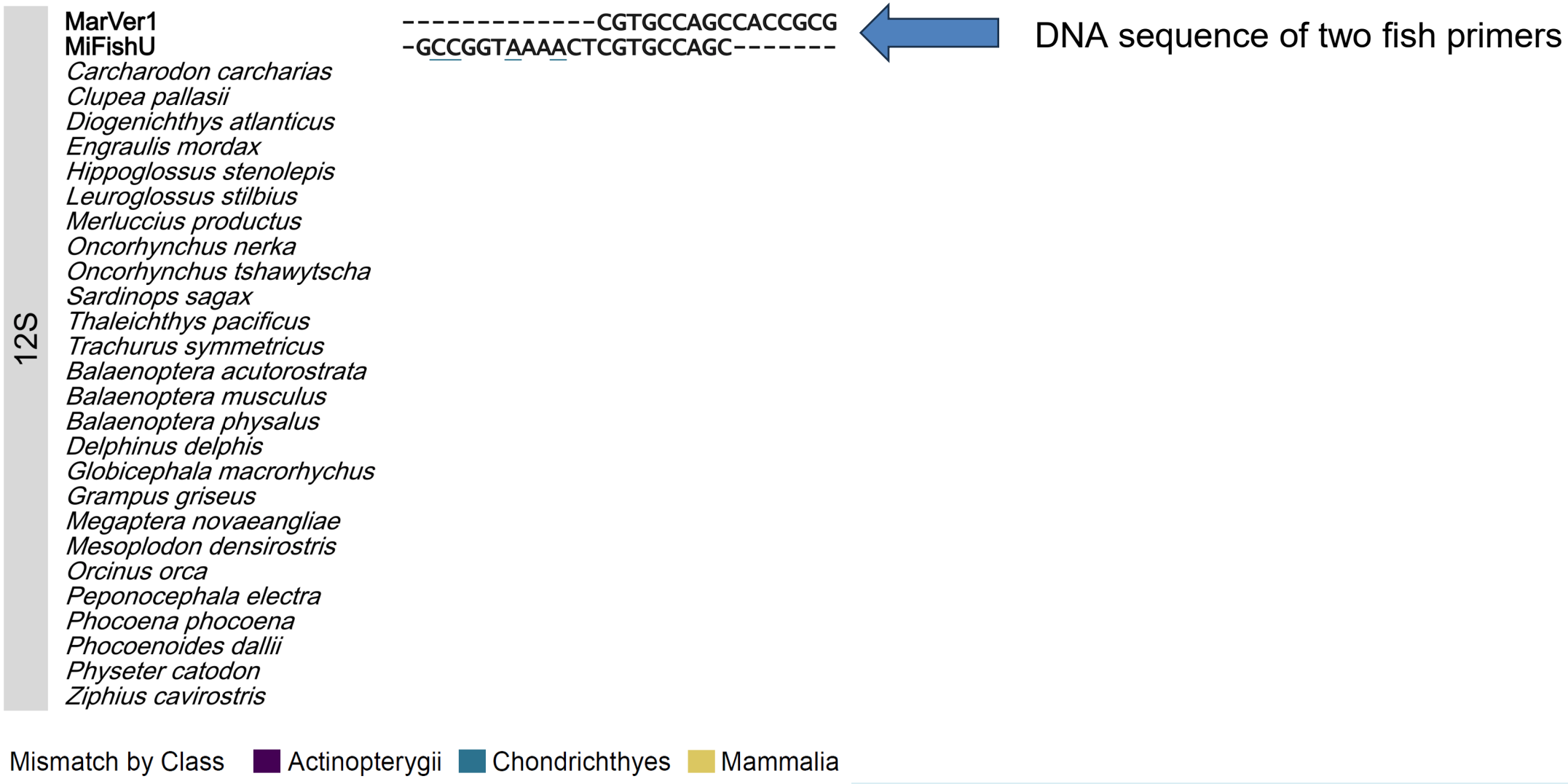




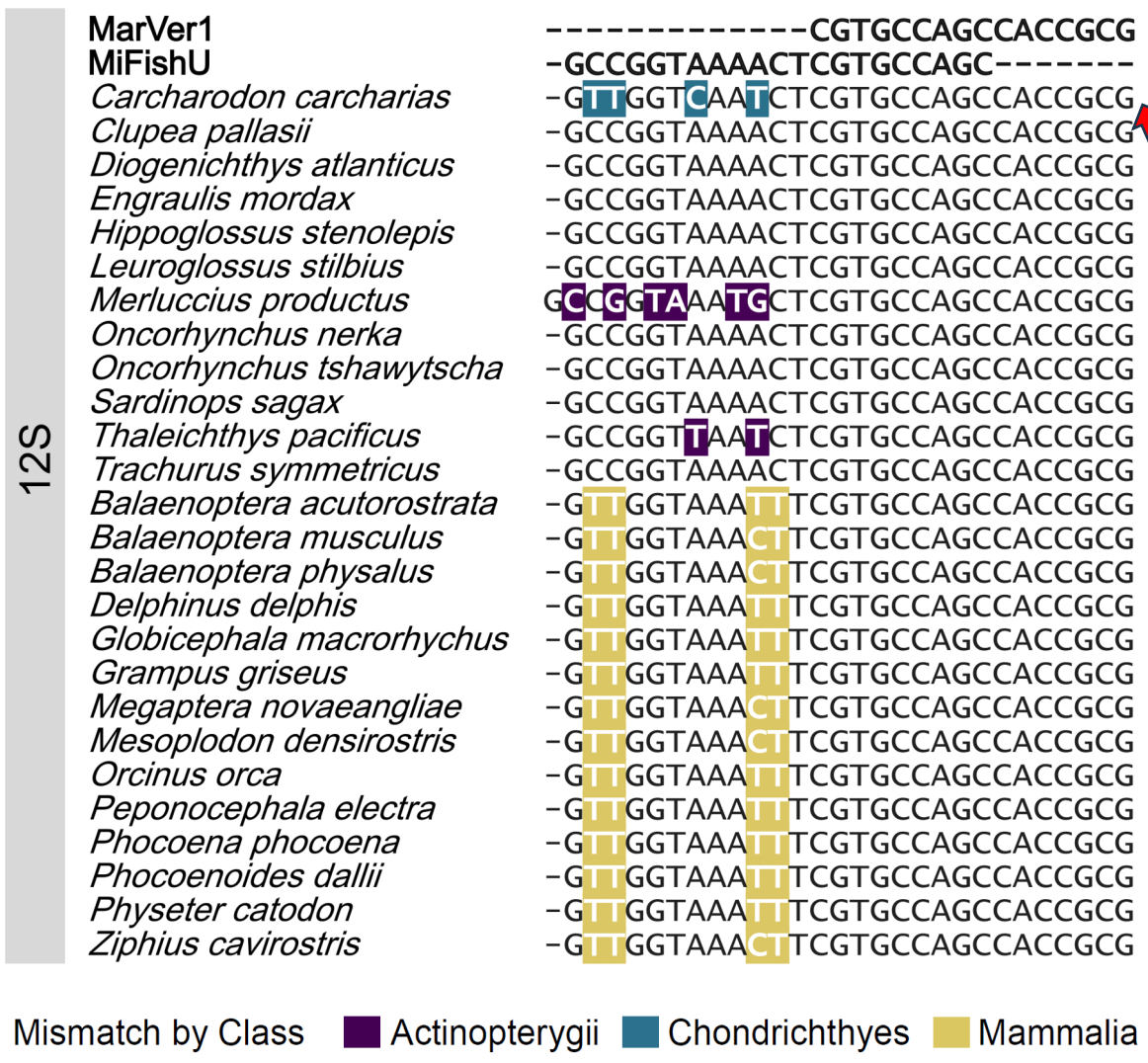
National Library of Medicine
National Center for Biotechnology Information

GBIF Global Biodiversity Information Facility

Primer match determines which taxa are observed



Primer match determines which taxa are observed

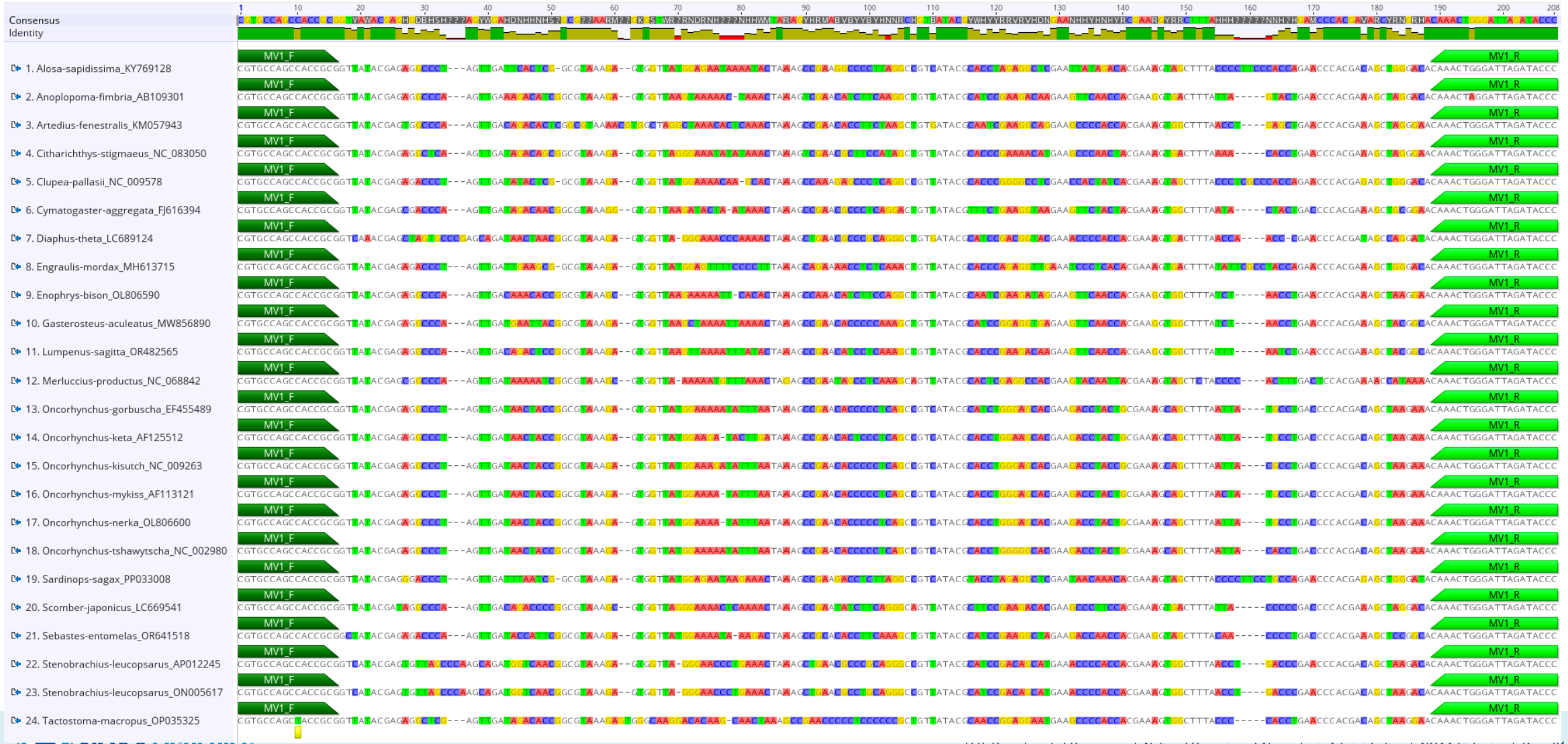


DNA sequence of two fish primers

MiFishU rarely observes *Carcharodon* (Great White Shark) or whales.

MarVer1 detects all of these taxa similarly.

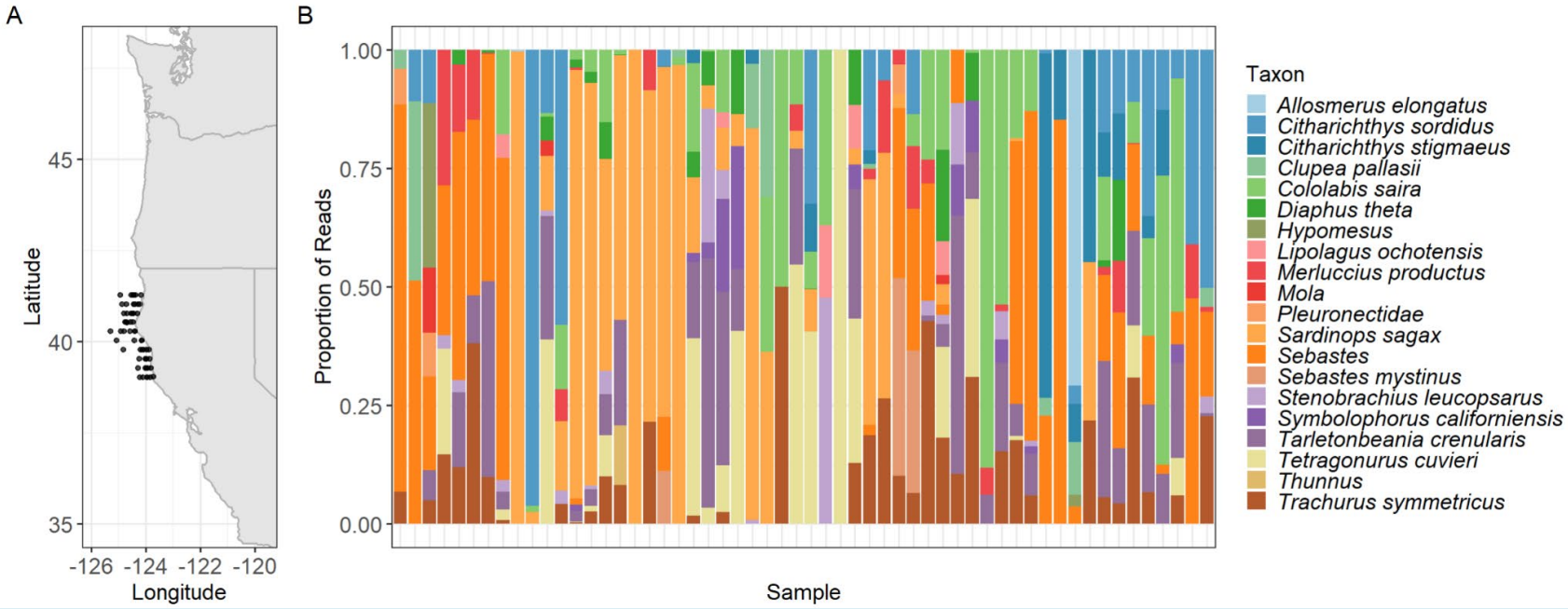
Amplicon variability determines potential taxonomic resolution



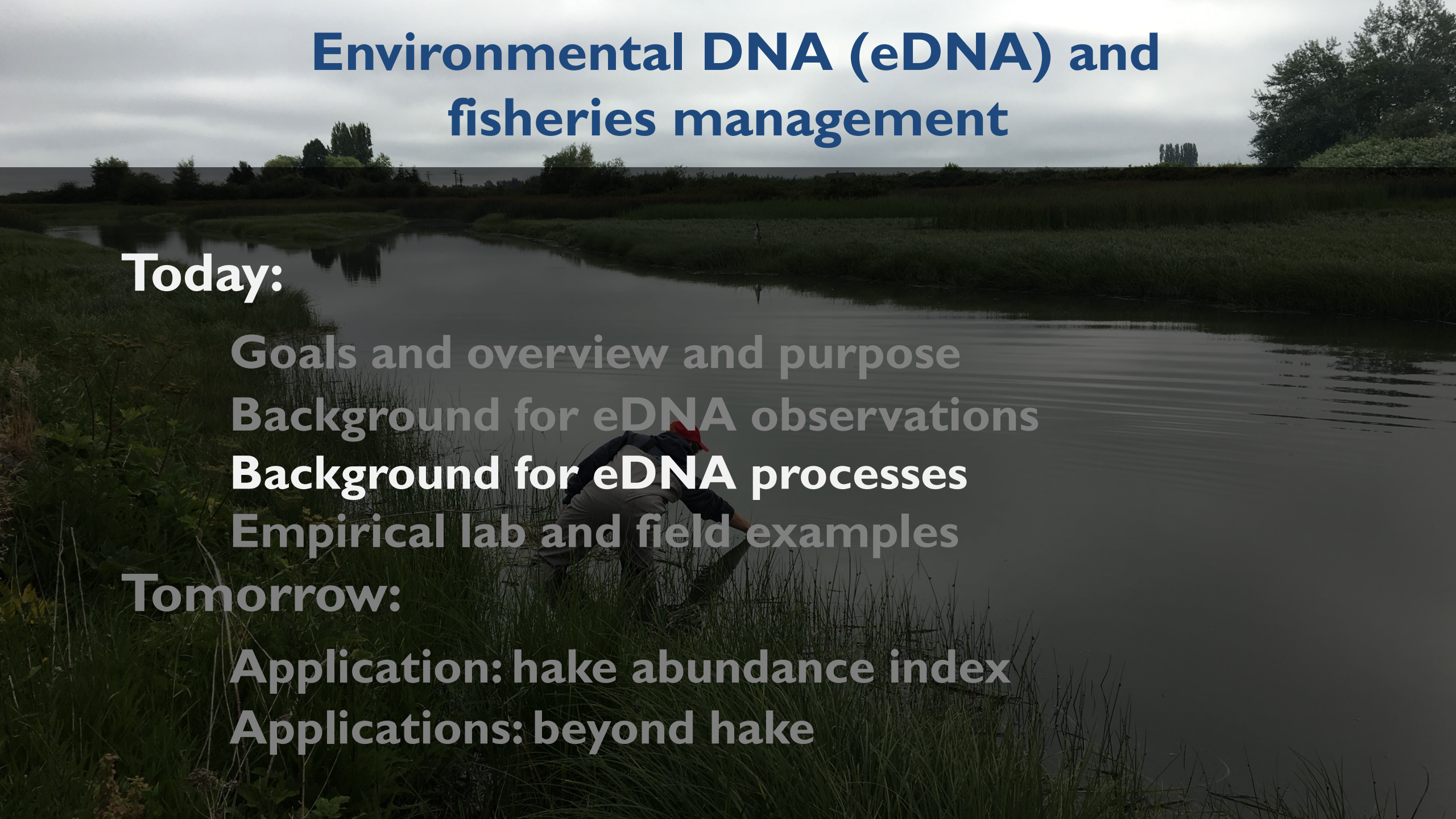
Metabarcoding produces a table (taxon by sample) of sequence counts

Sample	Chinook Salmon	Lingcod	Pacific Herring
A	10,025	0	2,343
B	8,523	12	0
C	52,333	8,909	6,852
D	0	12,003	65,235

How do you interpret this data?



Environmental DNA (eDNA) and fisheries management



Today:

- Goals and overview and purpose
- Background for eDNA observations
- **Background for eDNA processes**
- Empirical lab and field examples

Tomorrow:

- Application: hake abundance index
- Applications: beyond hake

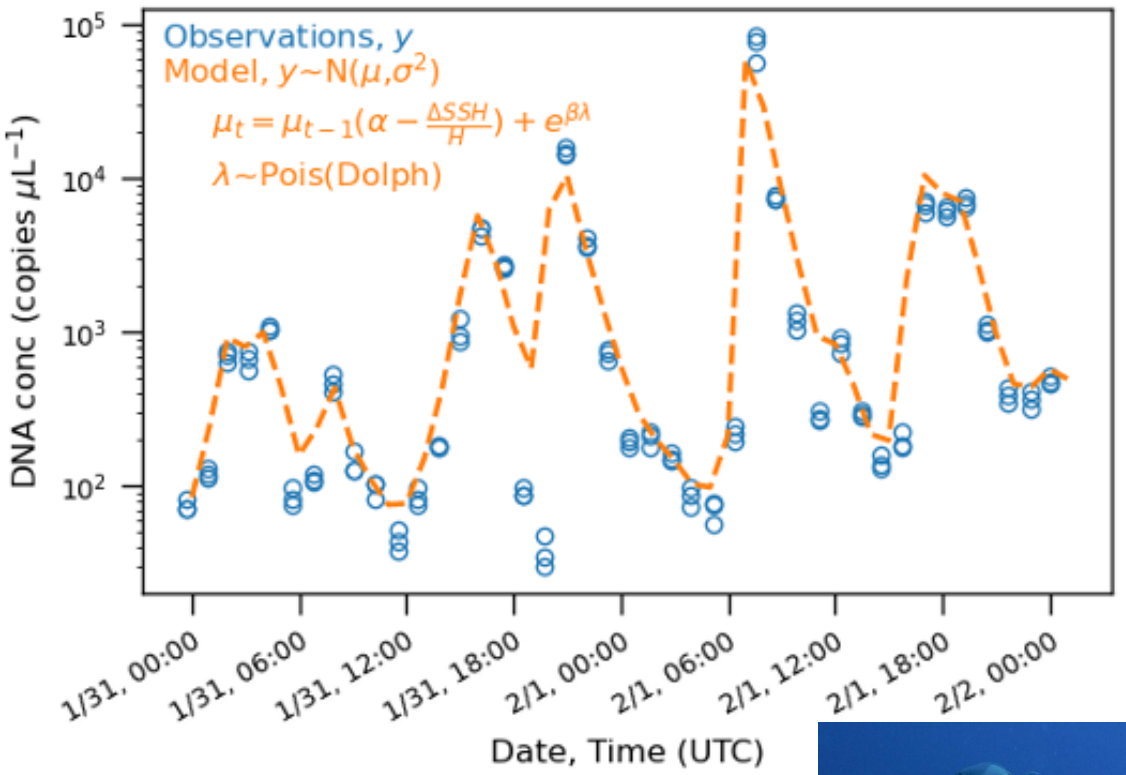
eDNA processes relevant for fisheries management

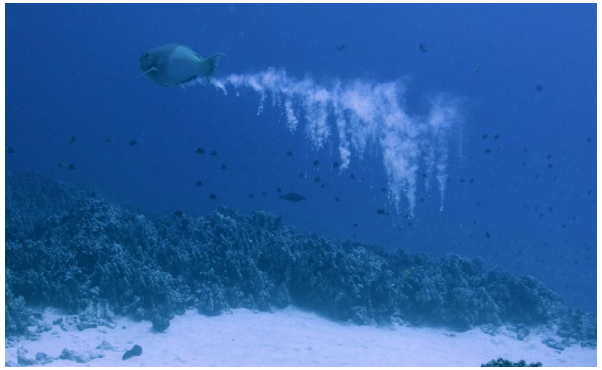
Production

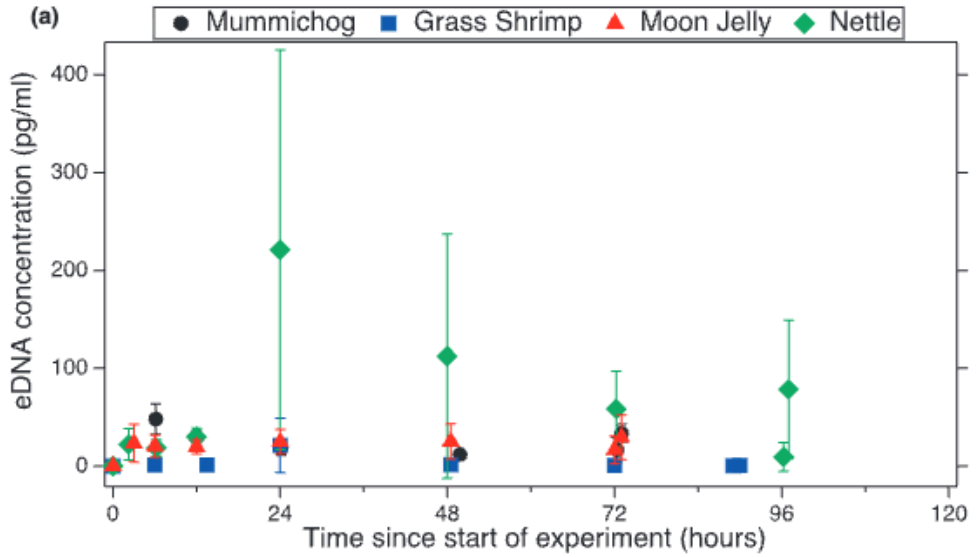
Loss

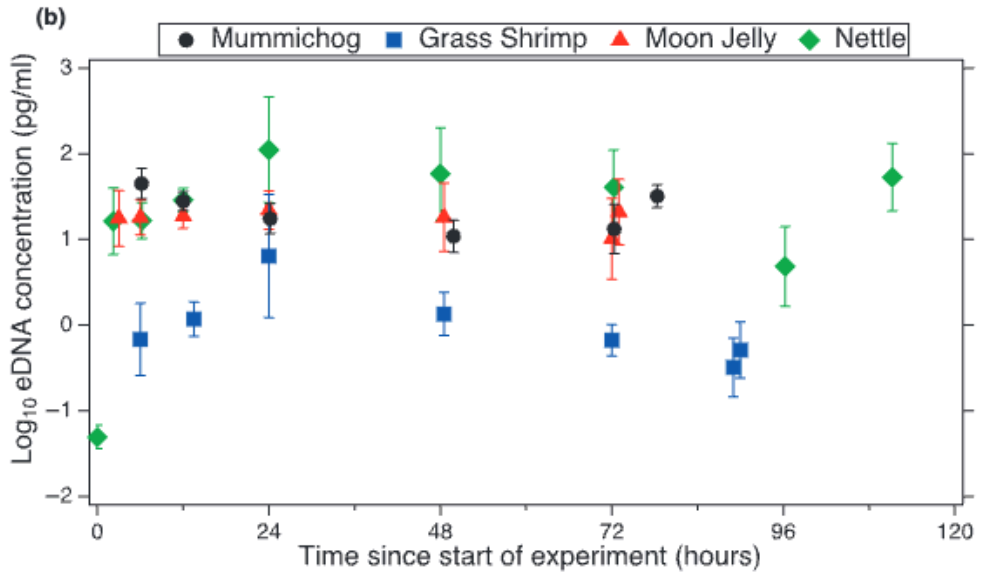
Transport

Production







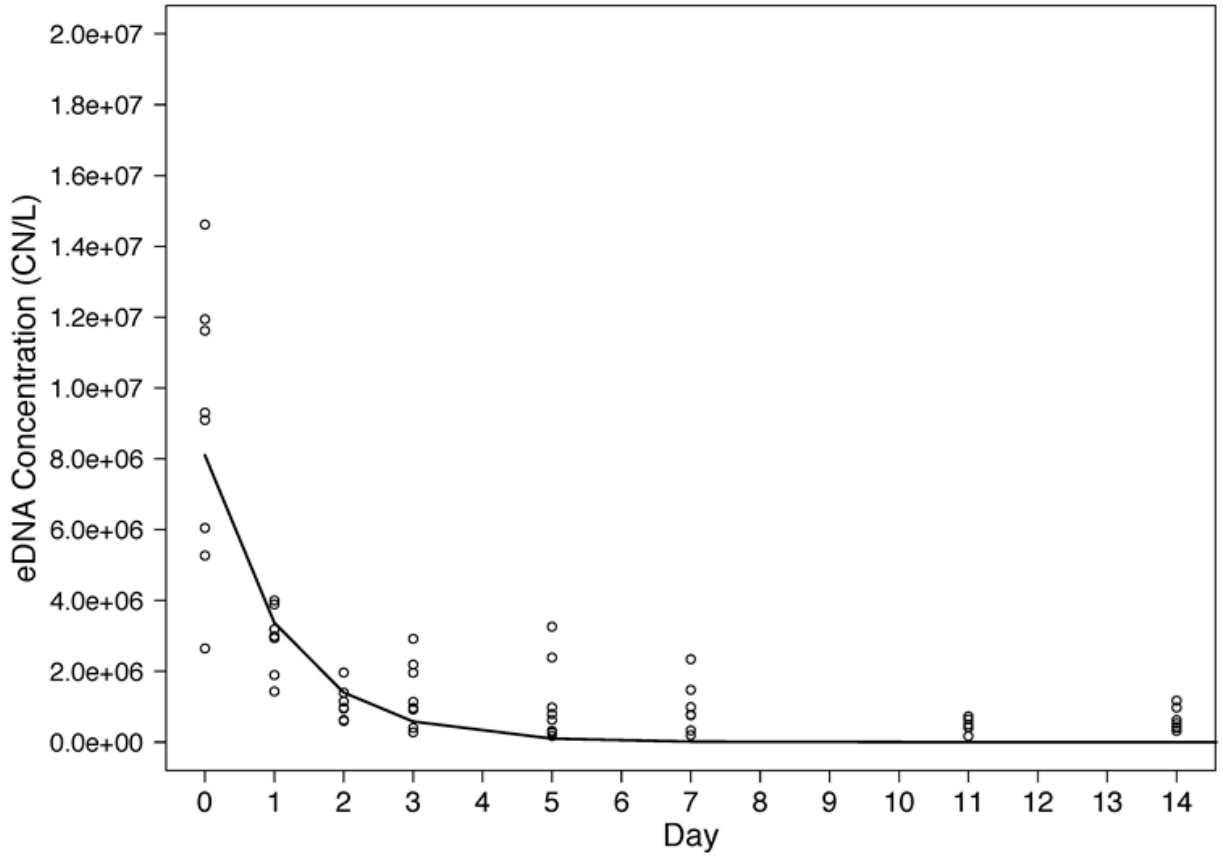


Brasseale et al. 2025

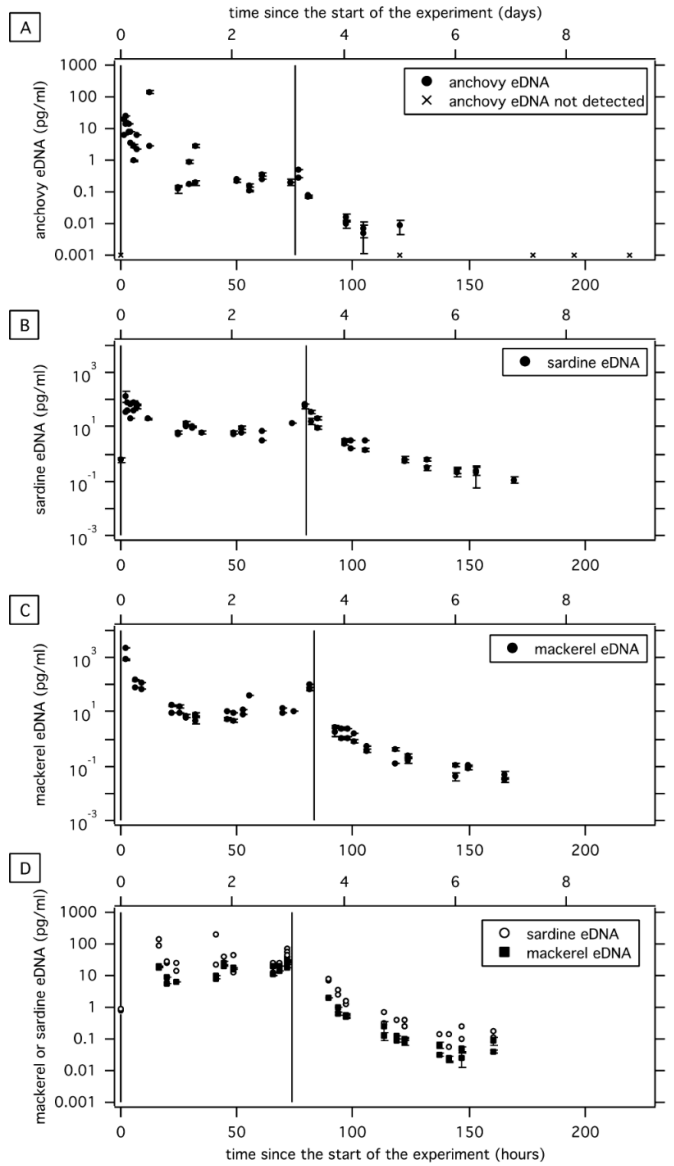
Allan et al. 2020

Loss

eDNA decays quickly



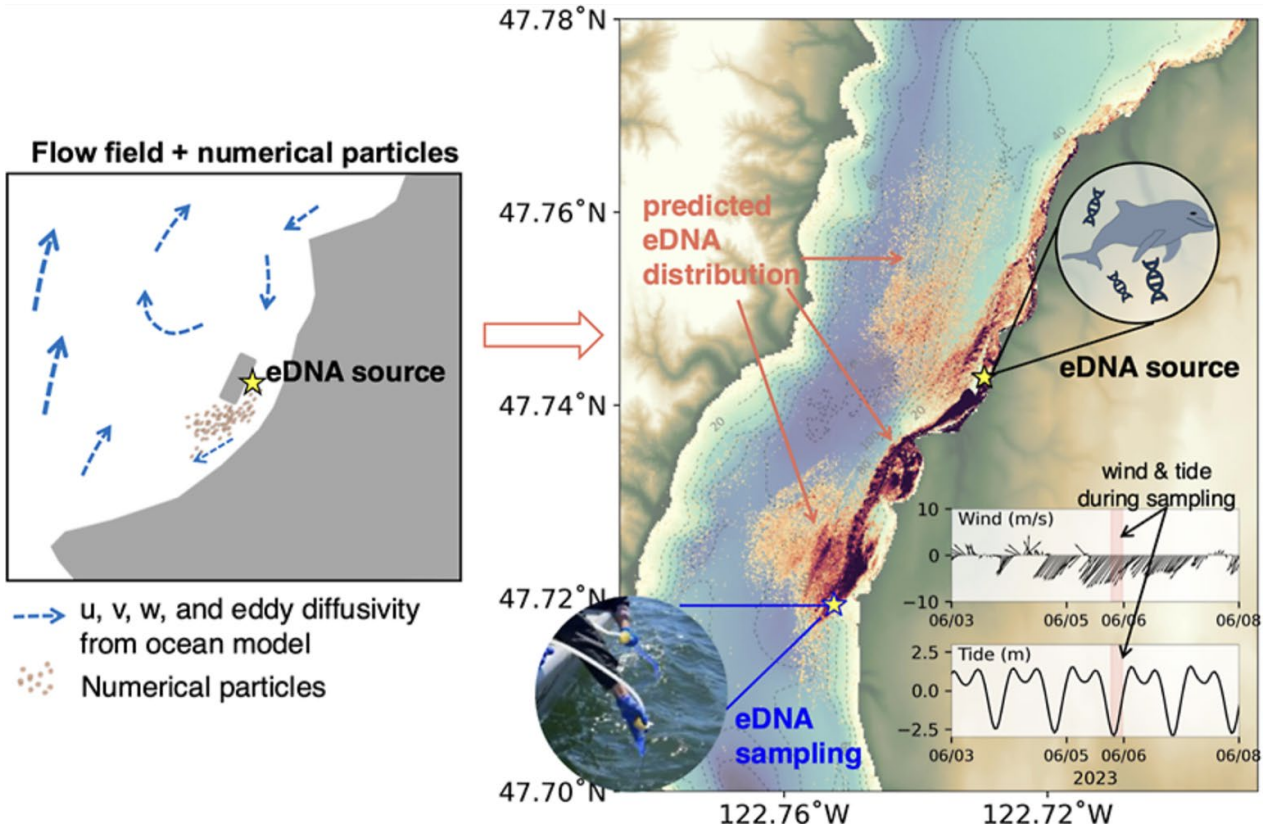
Lance et al. 2017

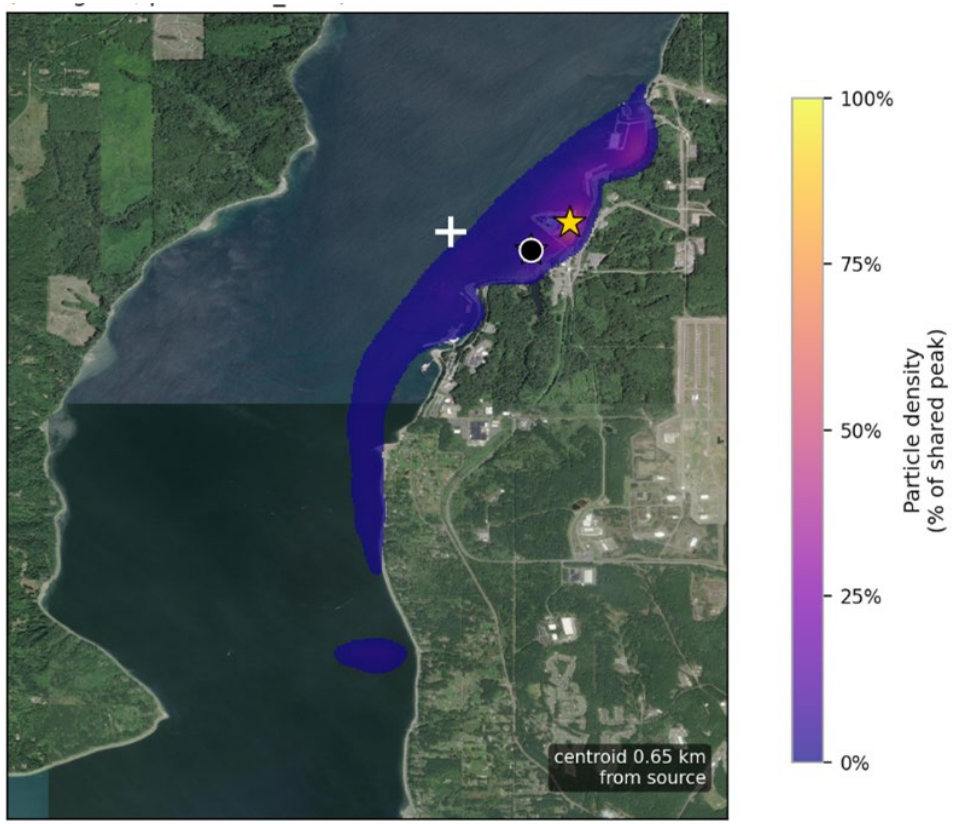


Sassoubre et al. 2017

Transport

eDNA goes where the water goes



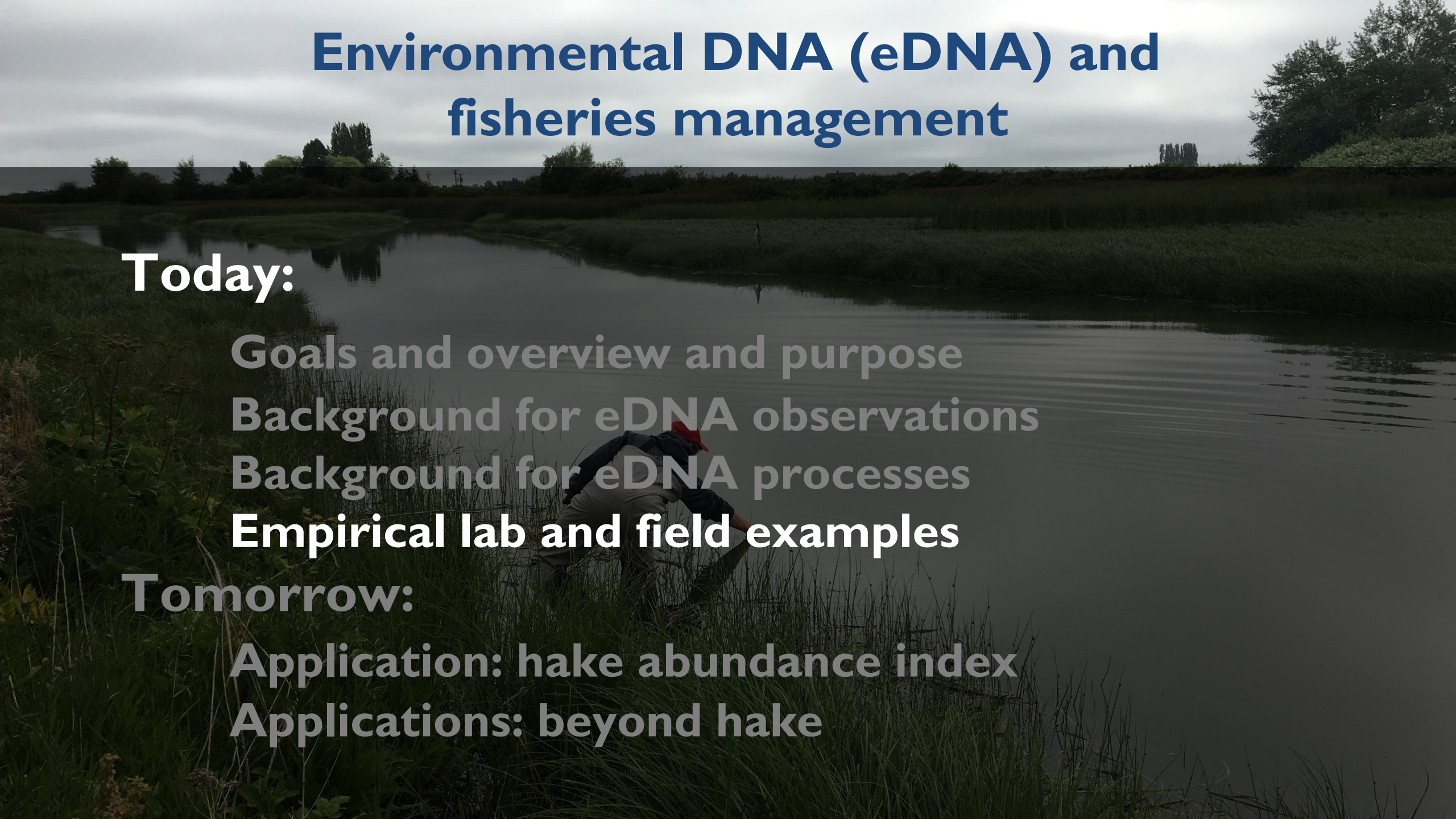


Xiong et al. 2025

eDNA processes summary

- eDNA production appears to be closely related to biomass.
  - temporal and life-history variability.
- Most eDNA is lost from the environment on the order of hours
- Loss rates do not appear to vary too much across environmental conditions.
  - difficult to investigate in the field

Environmental DNA (eDNA) and fisheries management



Today:

- Goals and overview and purpose
- Background for eDNA observations
- Background for eDNA processes
- **Empirical lab and field examples**

Tomorrow:

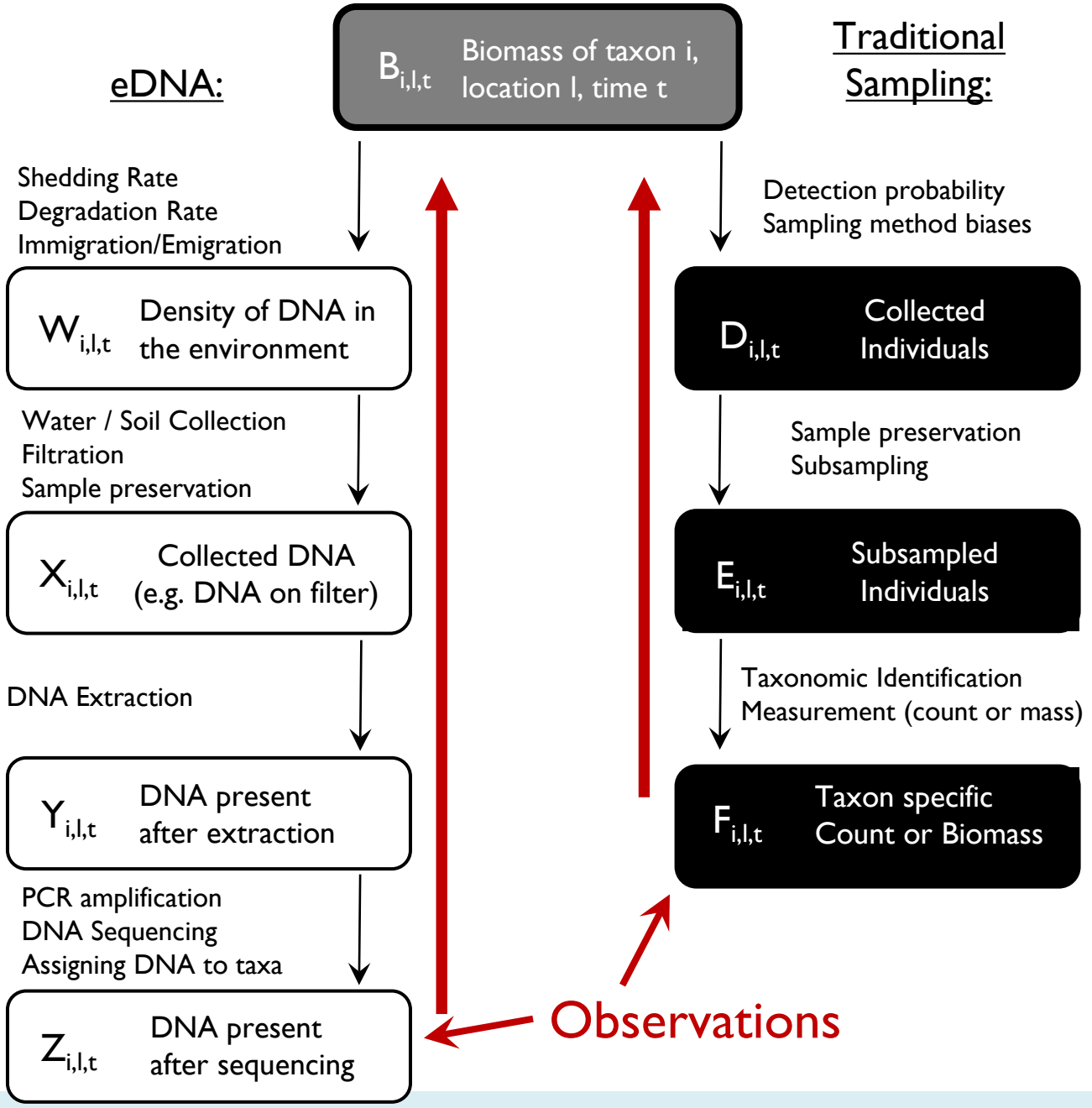
- Application: hake abundance index
- Applications: beyond hake

2016

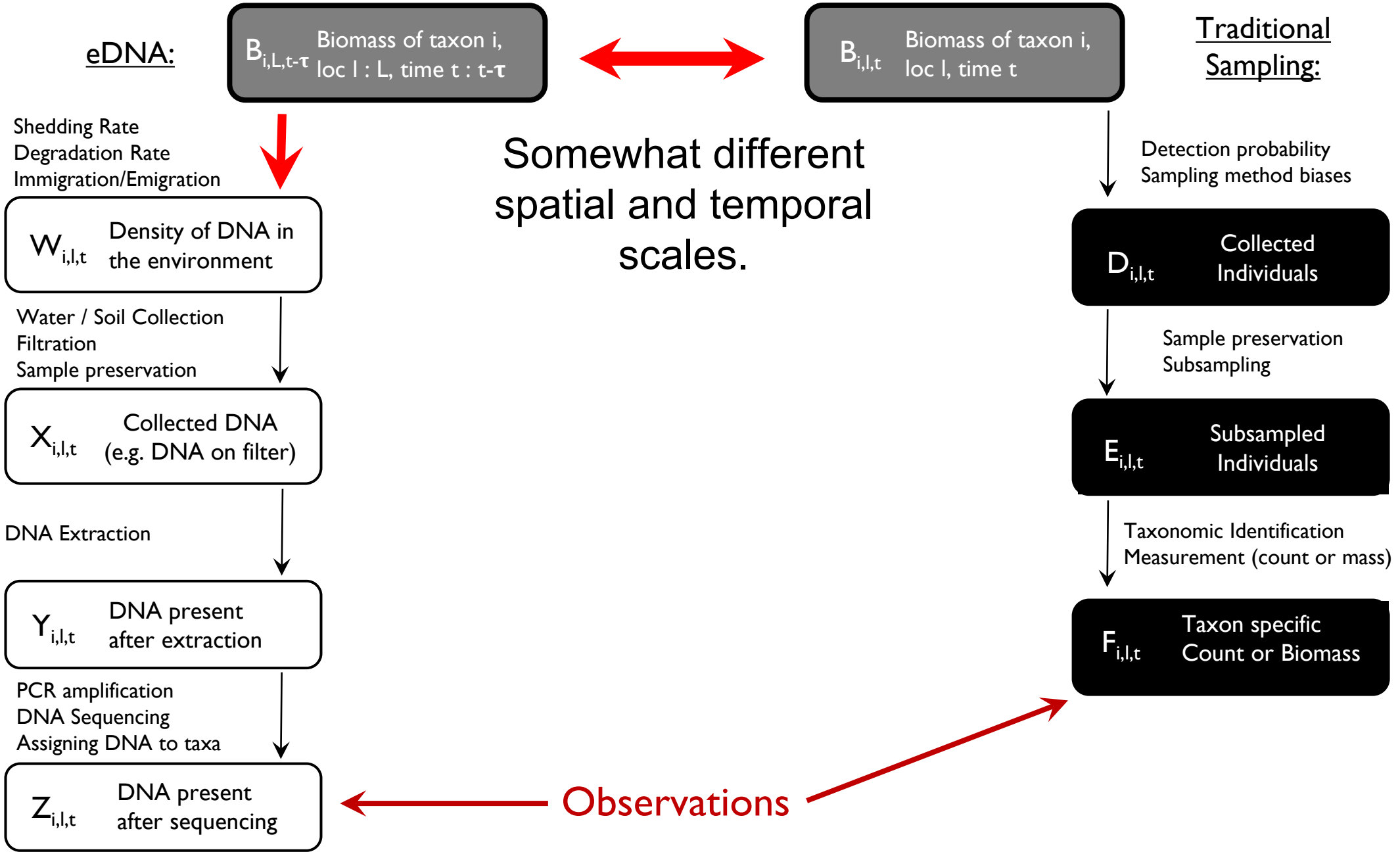
Ecological Applications, 26(6), 2016, pp. 1645–1659
© 2016 by the Ecological Society of America

A framework for inferring biological communities from environmental DNA

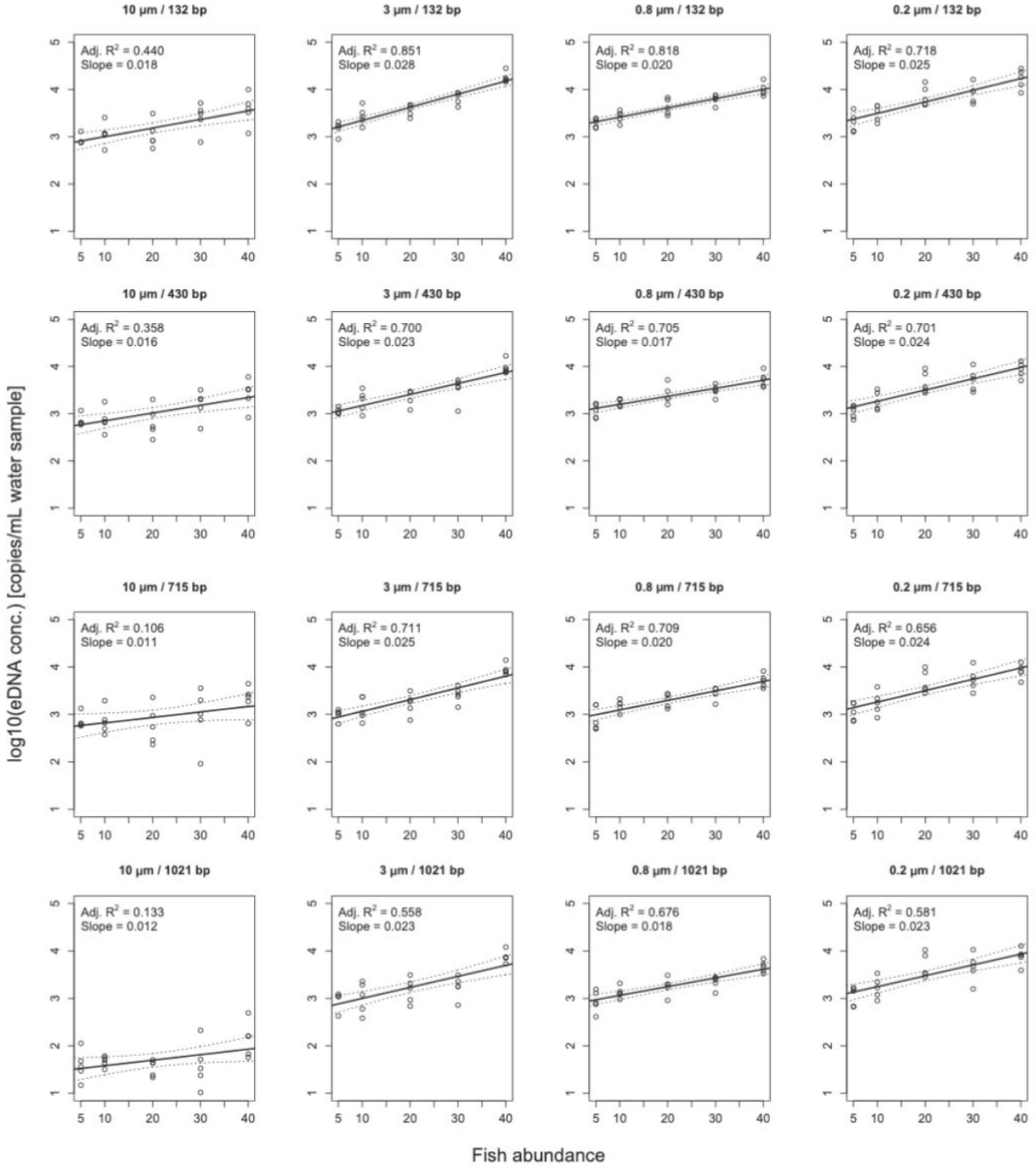
Andrew Olaf Shelton,[1,4] James Lawrence O'Donnell,[2] Jameal F. Samhouri,[1] Natalie Lowell,[2] Gregory D. Williams,[3] and Ryan P. Kelly[2]



2026

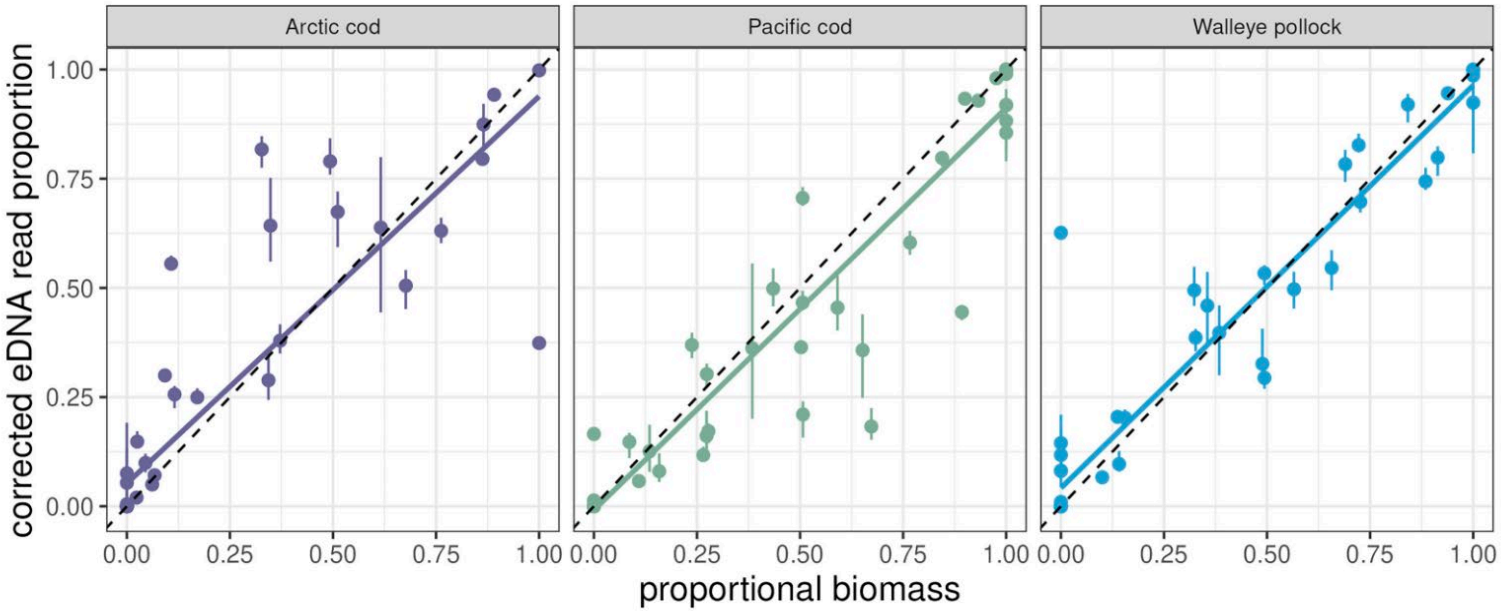

eDNA:
$B_{i,L,t-\tau}$ Biomass of taxon i, loc l : L, time t : t-τ
$B_{i,l,t}$ Biomass of taxon i, loc l, time t
Traditional Sampling:
Somewhat different spatial and temporal scales.
Shedding Rate
Degradation Rate
Immigration/Emigration
$W_{i,l,t}$ Density of DNA in the environment
Water / Soil Collection
Filtration
Sample preservation
$X_{i,l,t}$ Collected DNA (e.g. DNA on filter)
DNA Extraction
$Y_{i,l,t}$ DNA present after extraction
PCR amplification
DNA Sequencing
Assigning DNA to taxa
$Z_{i,l,t}$ DNA present after sequencing
Observations
Detection probability
Sampling method biases
$D_{i,l,t}$ Collected Individuals
Sample preservation
Subsampling
$E_{i,l,t}$ Subsampled Individuals
Taxonomic Identification
Measurement (count or mass)
$F_{i,l,t}$ Taxon specific Count or Biomass

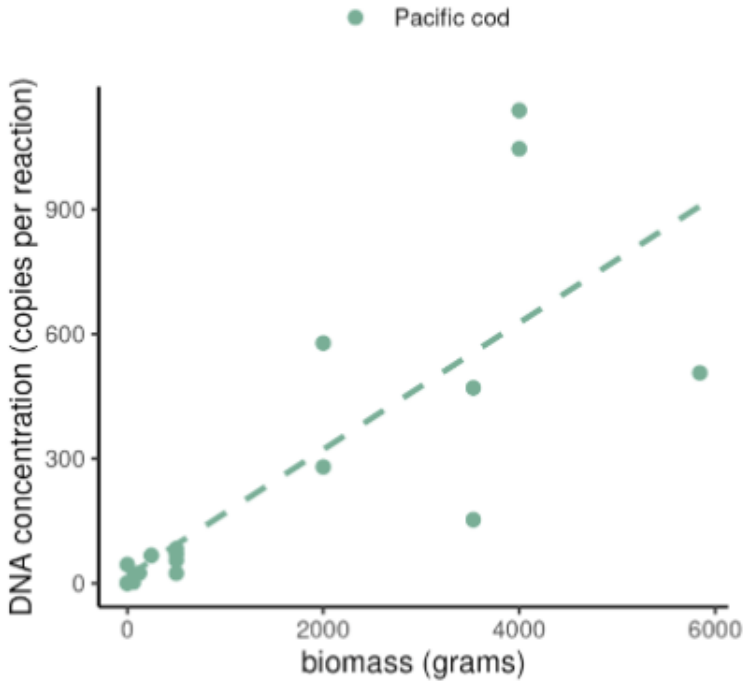
Zebrafish in tanks



Jo and Yamanaka 2022

Gadids in Tanks

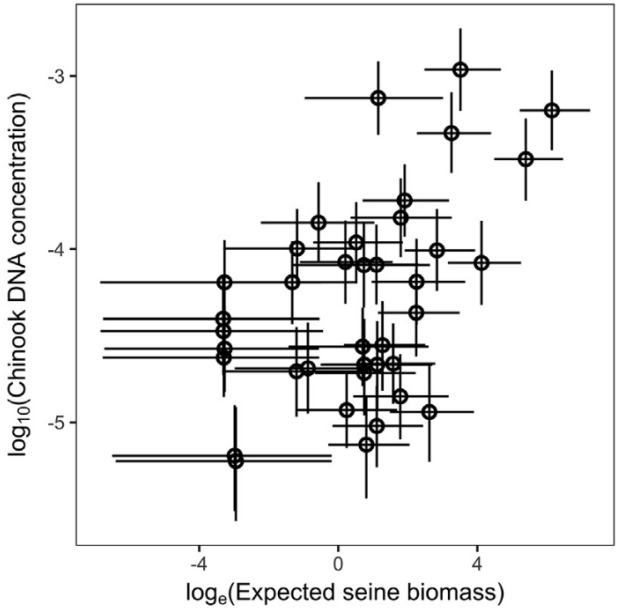




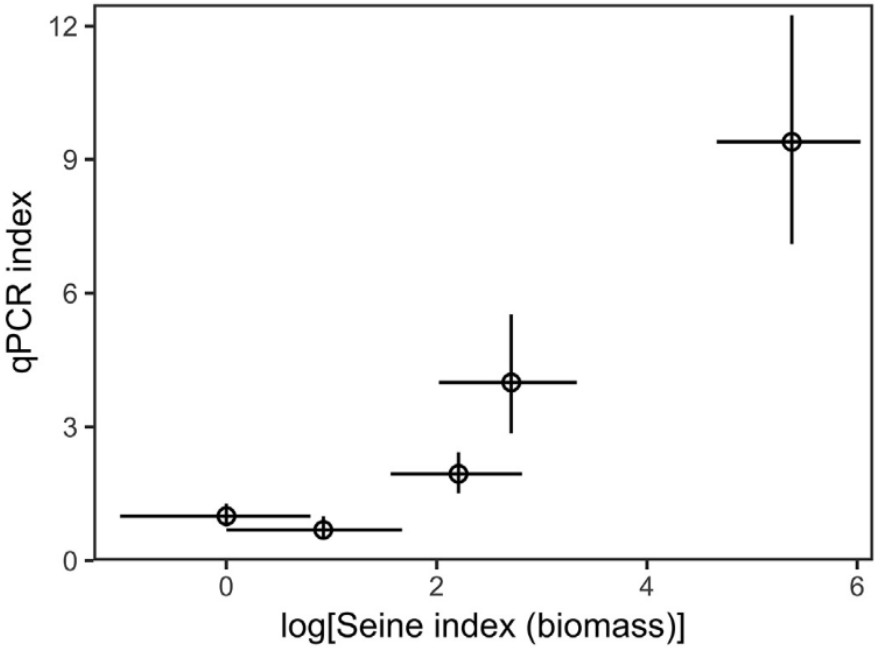
Ledger et al. 2024

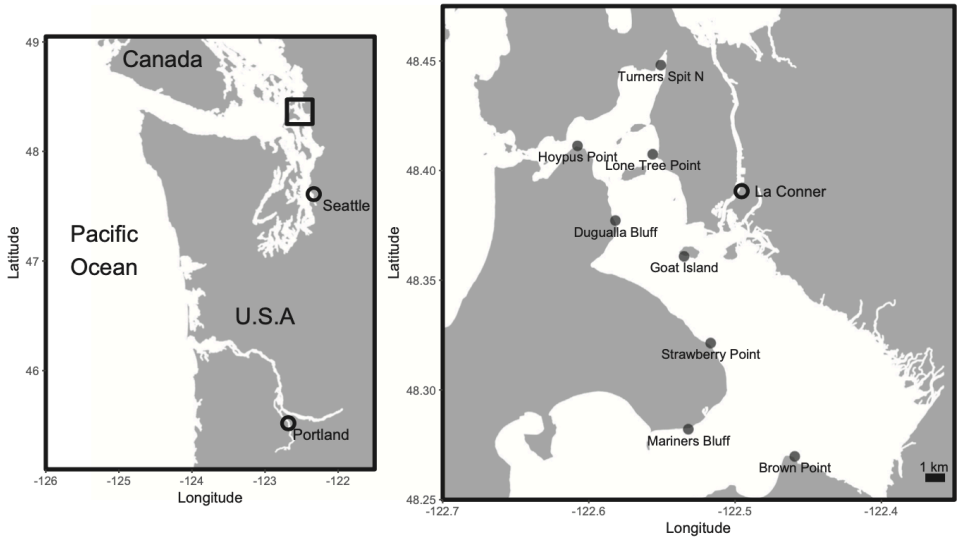
Chinook salmon in Skagit Bay, WA

Site-to-site comparison



Monthly comparison

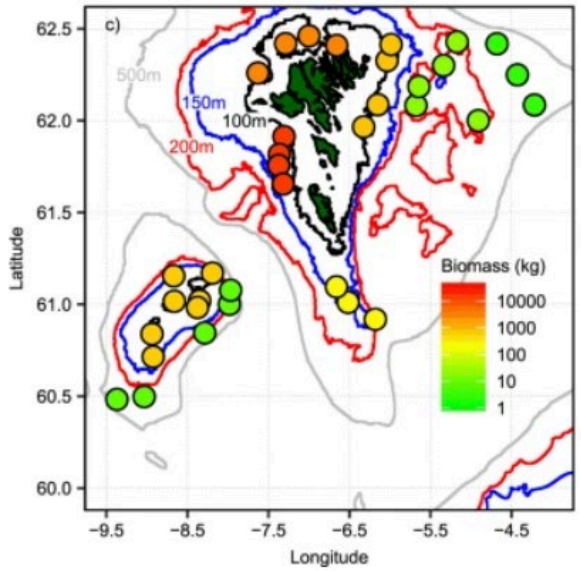


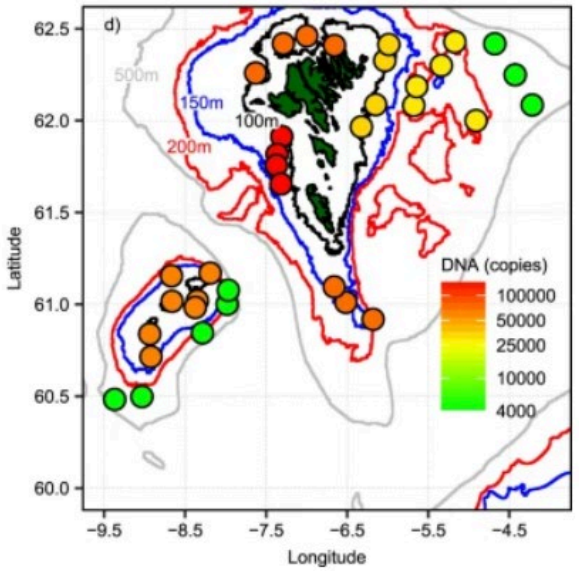


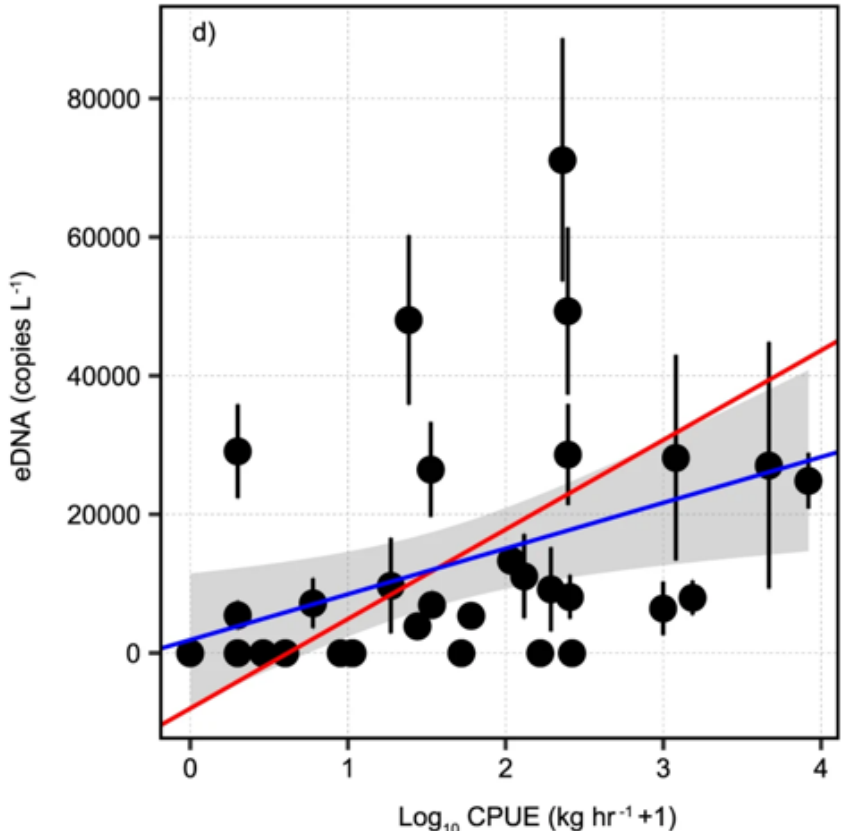
NOAA FISHERIES

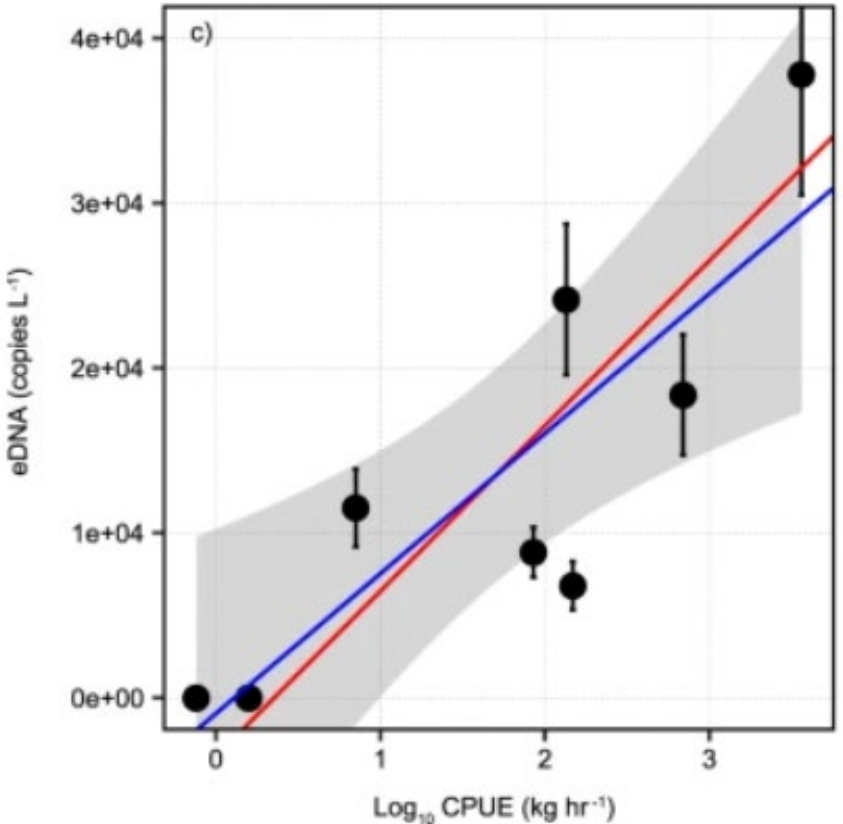
Shelton et al. 2019

Faroe Islands Cod



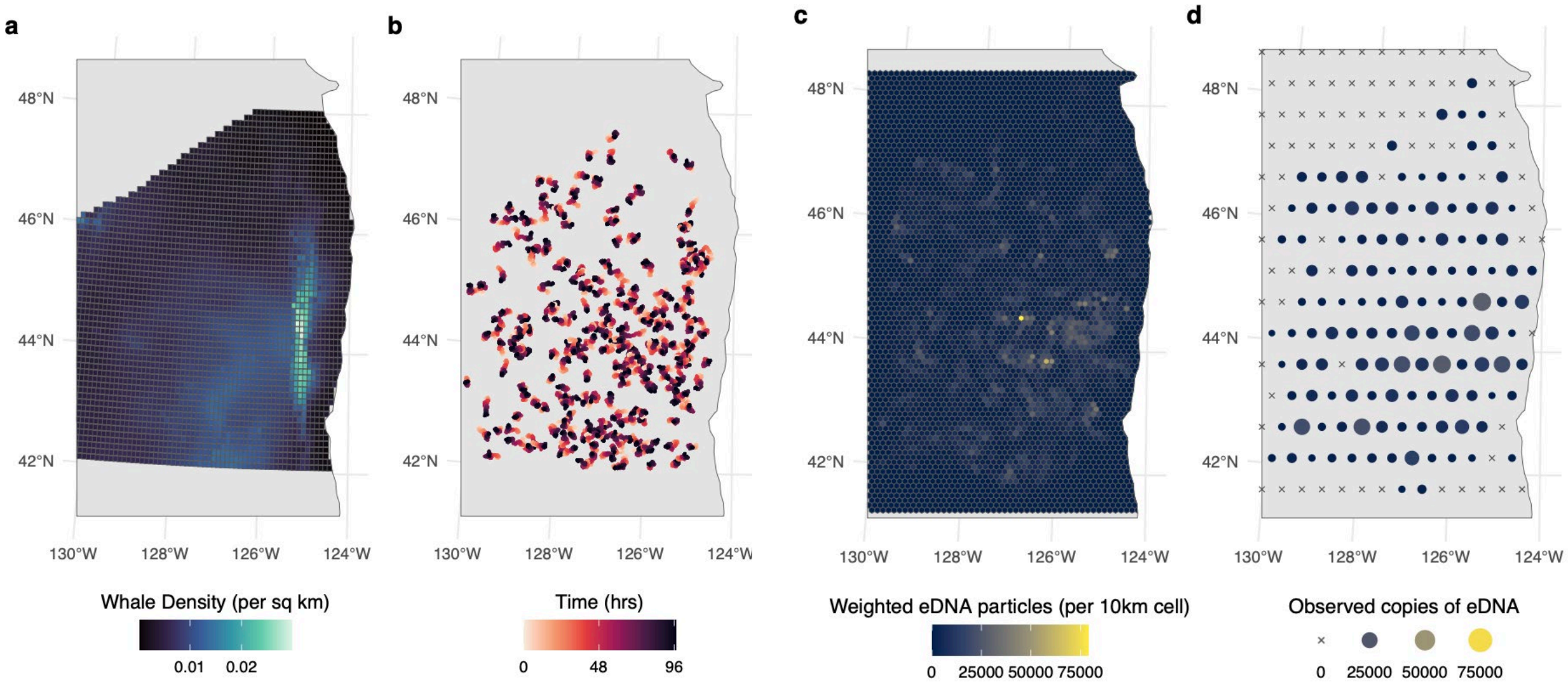






Salter et al. 2019

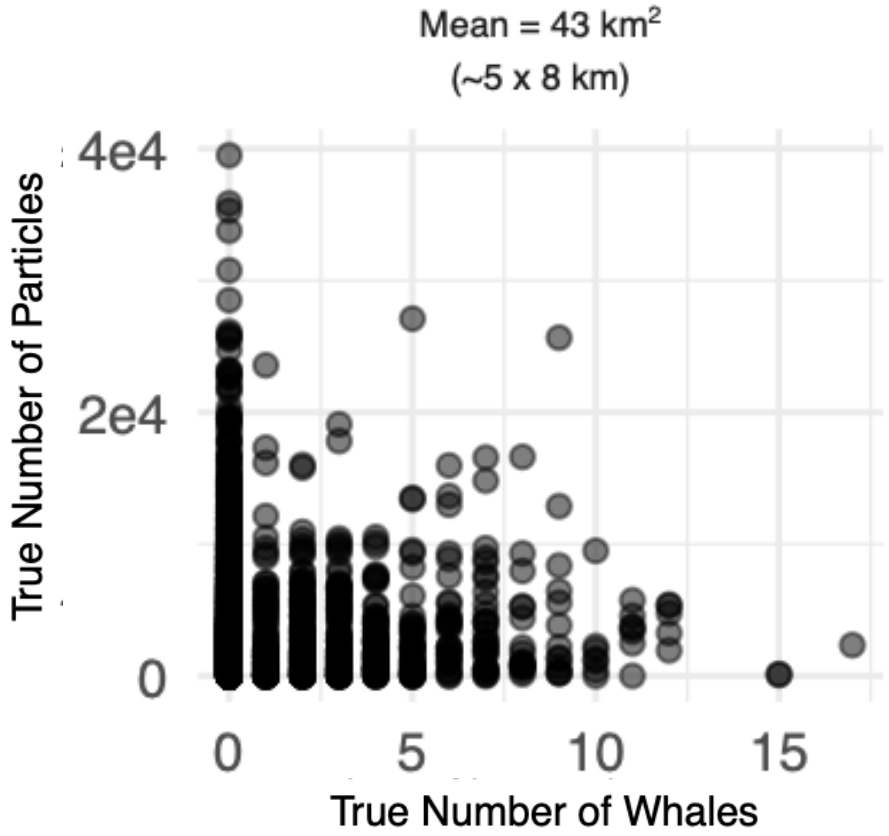
Fin Whales in silico



NOAA FISHERIES

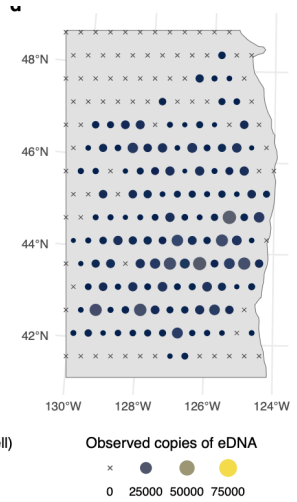
Jacobsen et al. In Review

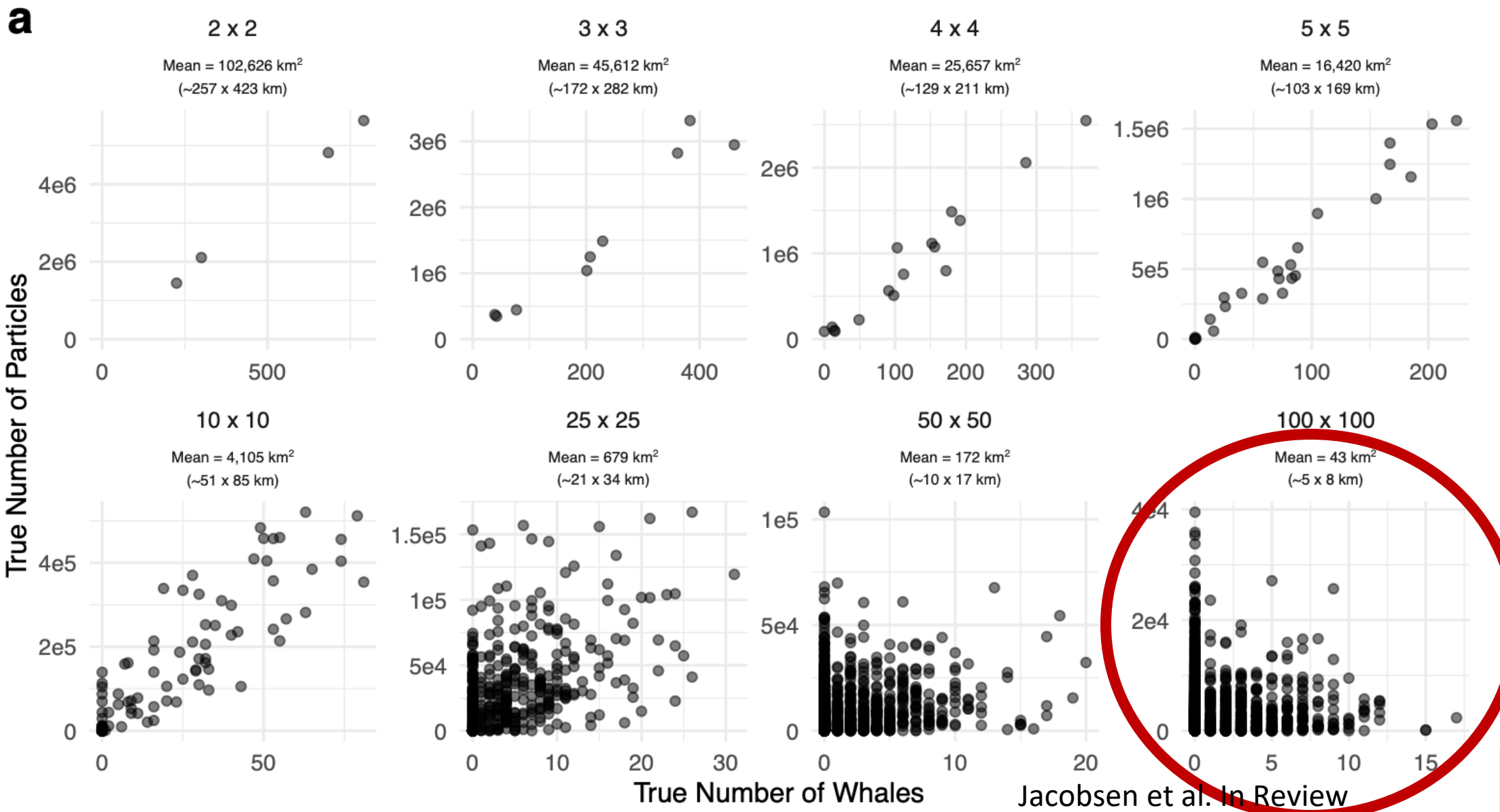
Fin Whales in silico



Jacobsen et al. In Review

Fin Whales in silico

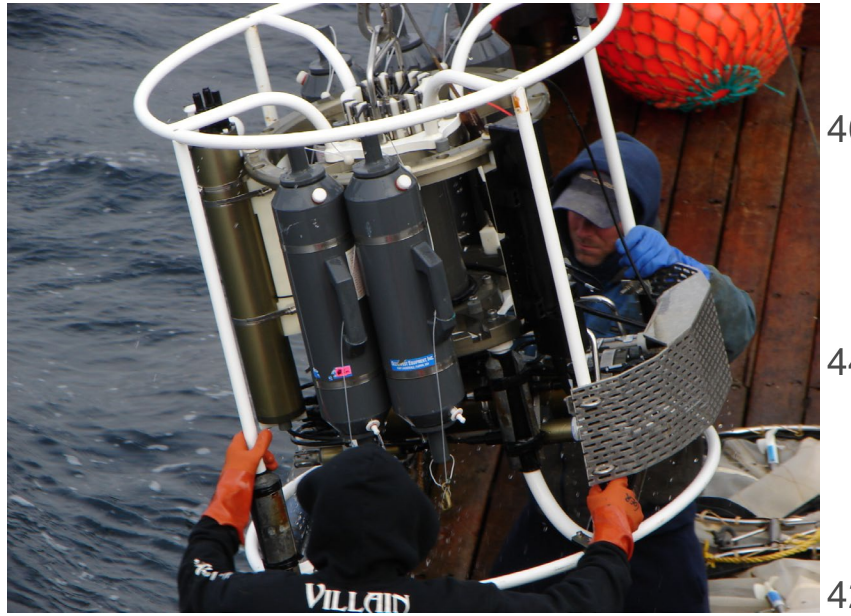




Jacobsen et al. In Review

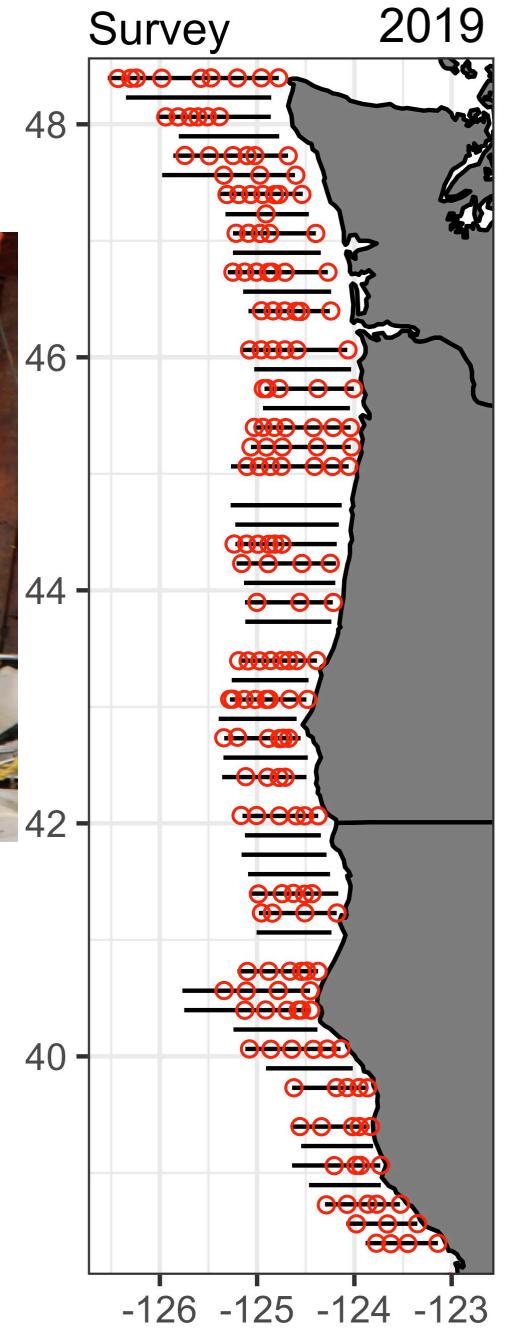
Hake in 2019

- 186 stations sampled
- 2 x 2.5 L seawater filtered from 6 depths
  > 1800 bottles sampled annually.
- Assayed with Single-species qPCR primer
  - Pacific hake DNA concentration

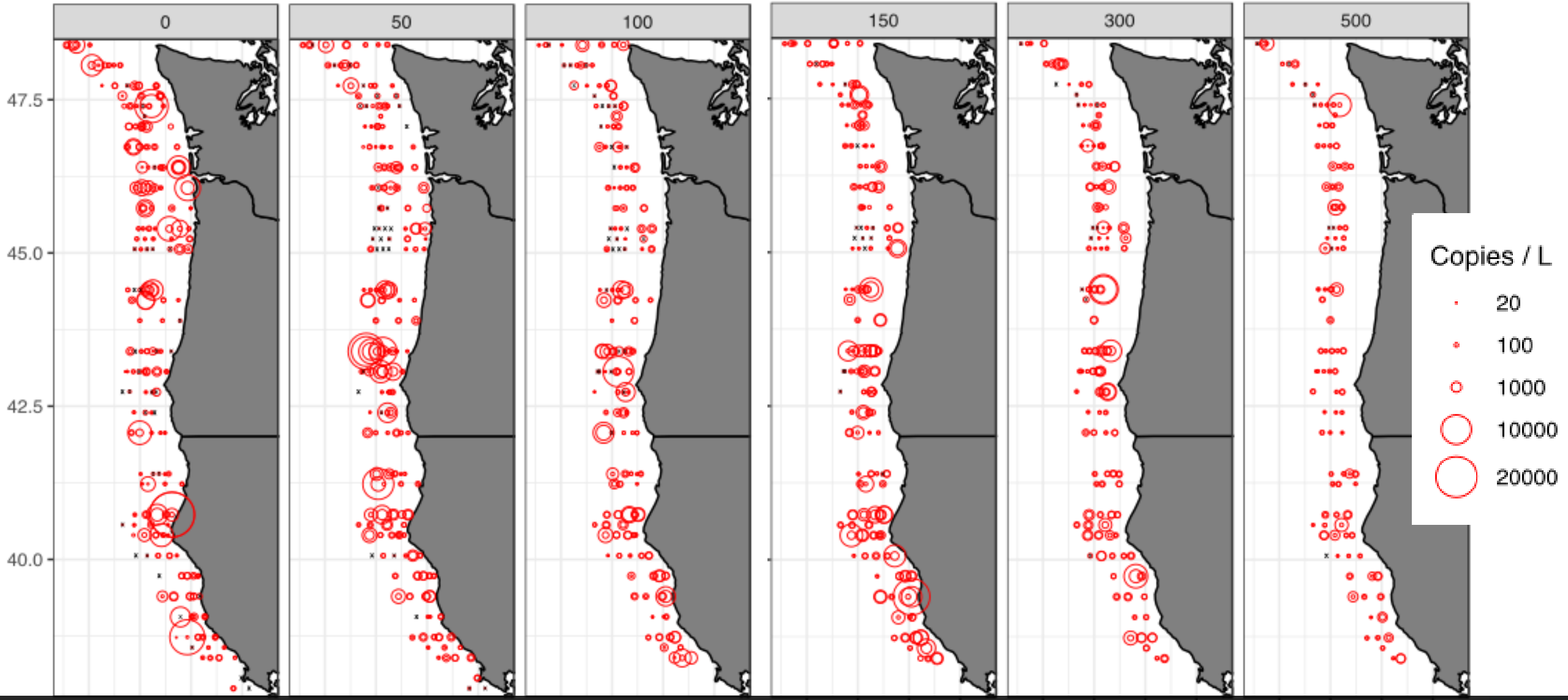


- Direct acoustics and DNA not collected simultaneously
  (sample-to-sample comparisons are not possible)
- A range of controls to check for contamination

Largest eDNA sampling effort in the world

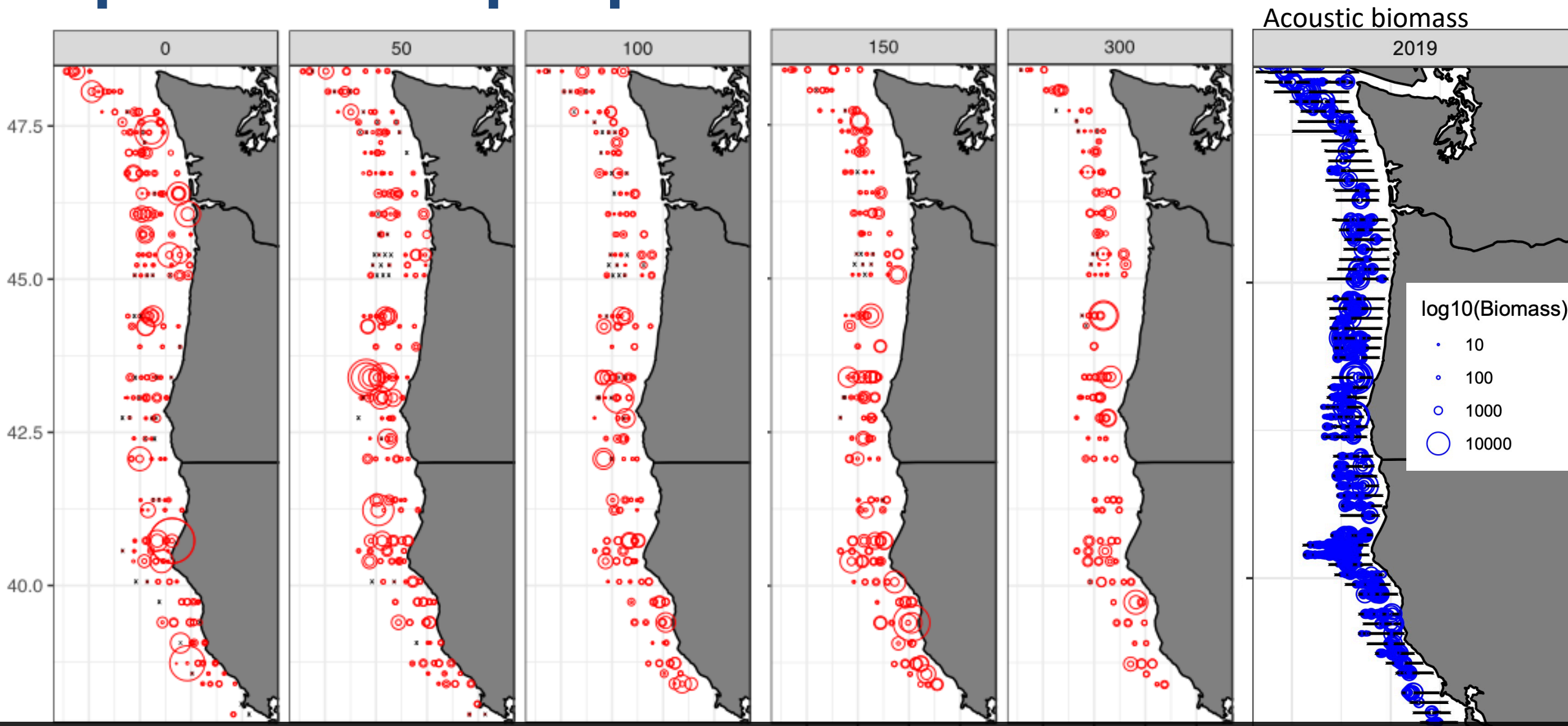


Spatial and depth patterns of hake DNA in 2019



You cannot sample the surface to understand deep waters.

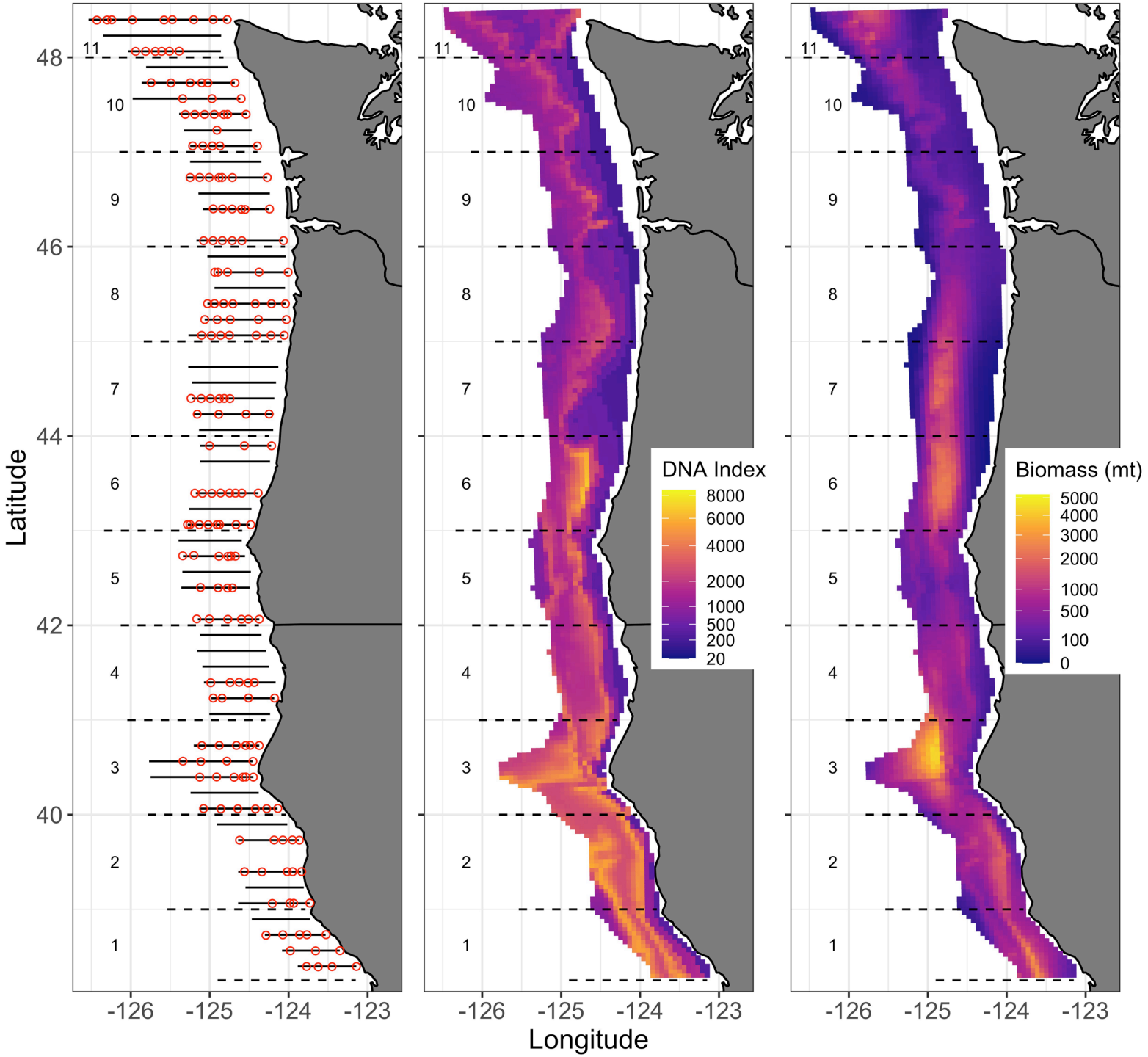
Spatial and depth patterns of hake DNA in 2019



You cannot sample the surface to understand deep waters.

Generate indices of abundance comparable to acoustic-trawl surveys

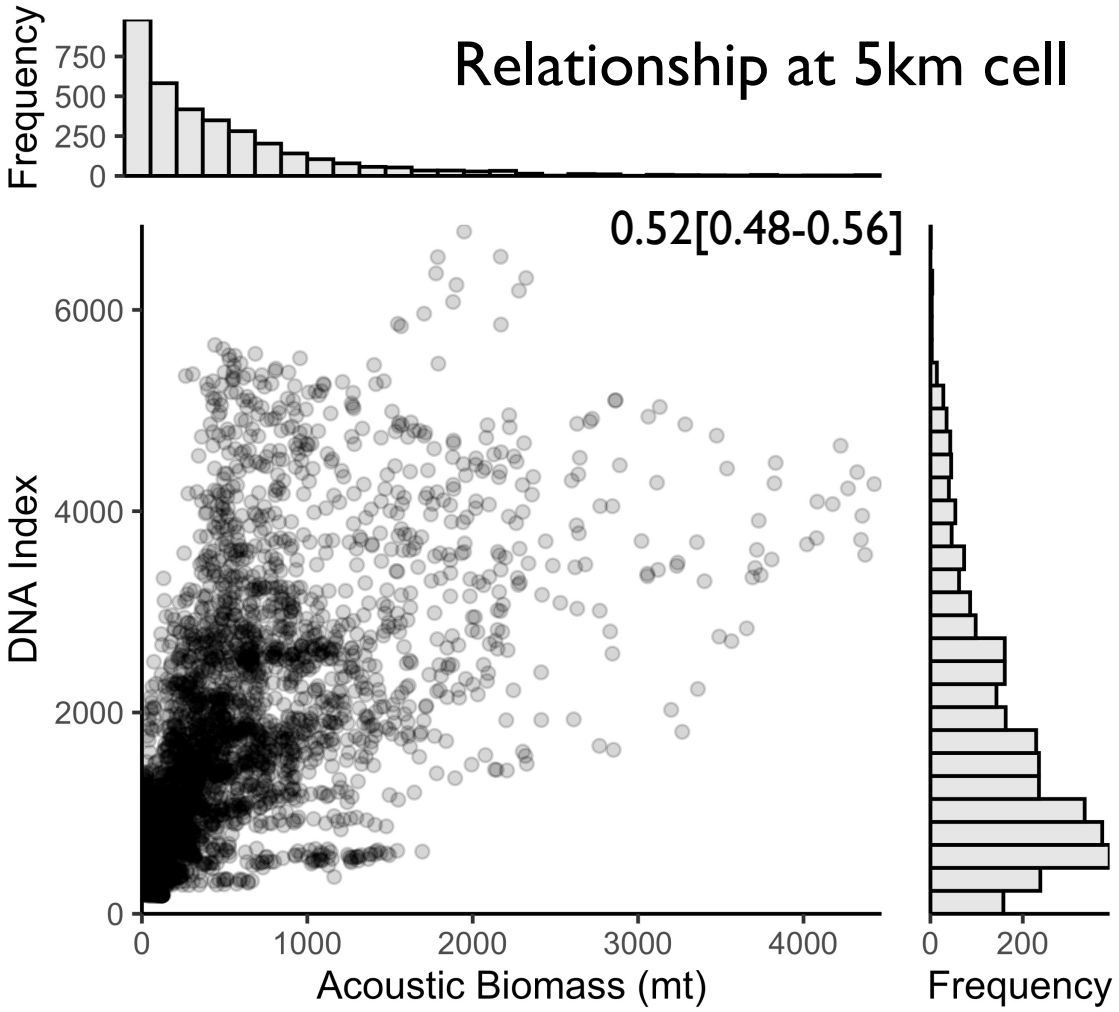
Used spatial Index standardization very similar to methods currently used for PFMC groundfish stocks



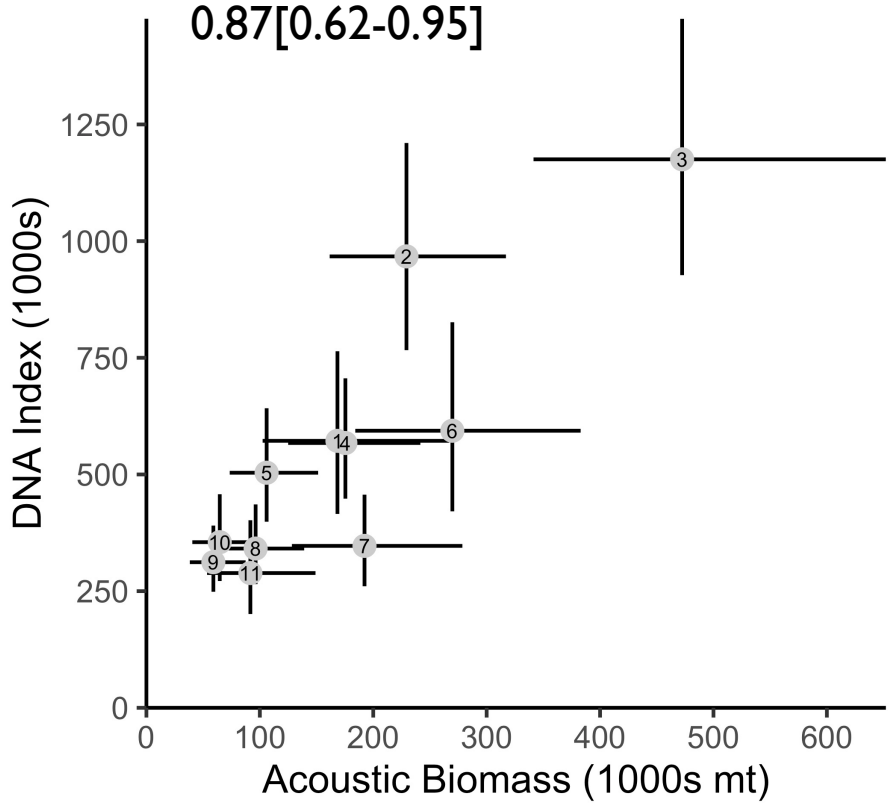
NOAA FISHERIES

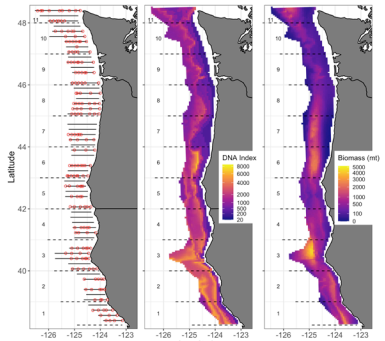
Estimates of abundance are strongly correlated
(at large spatial scales)

Relationship at 5km cell

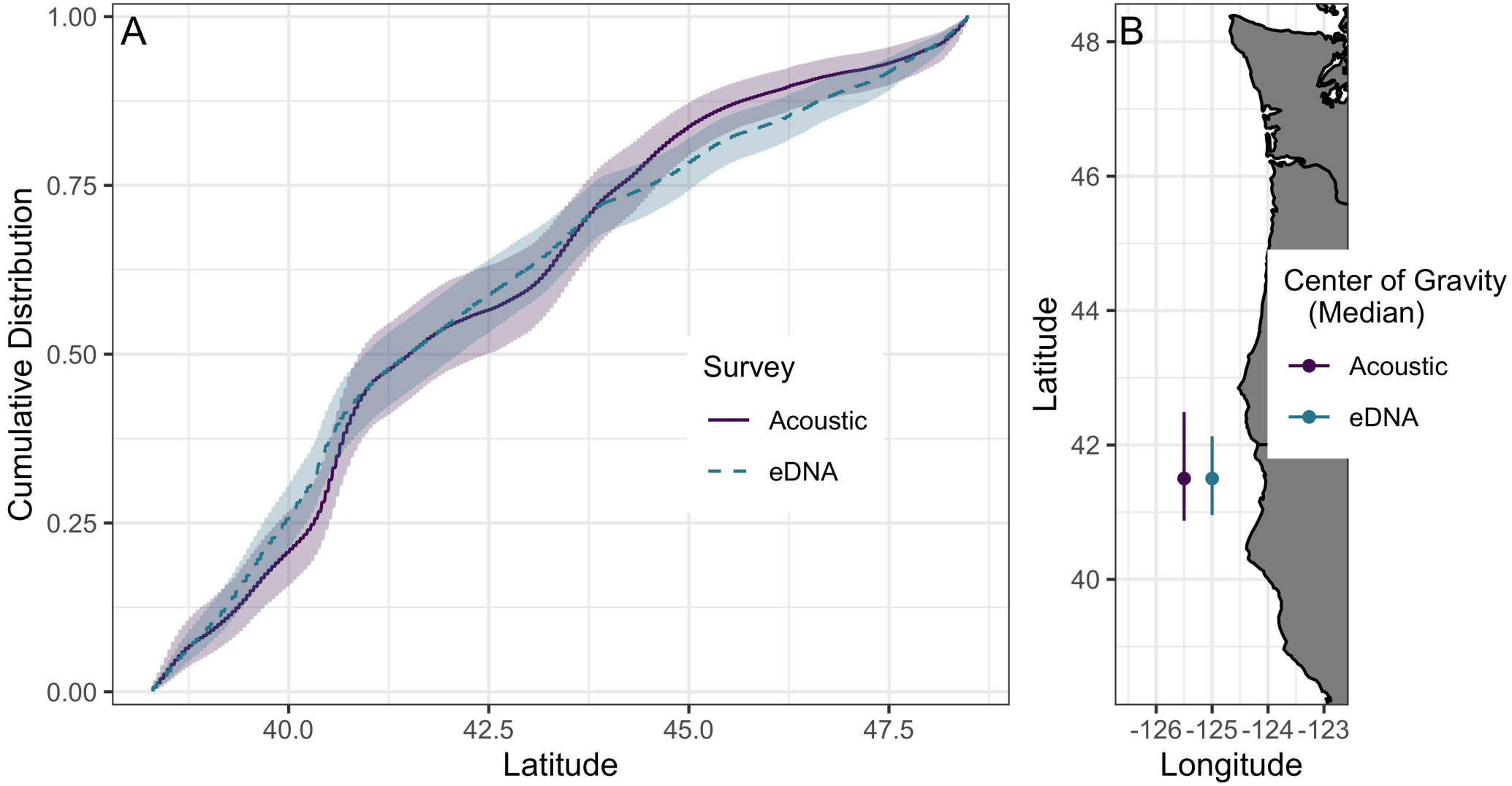


Relationship in 1 degree latitude bins





Similarities extend to estimates of species distribution…

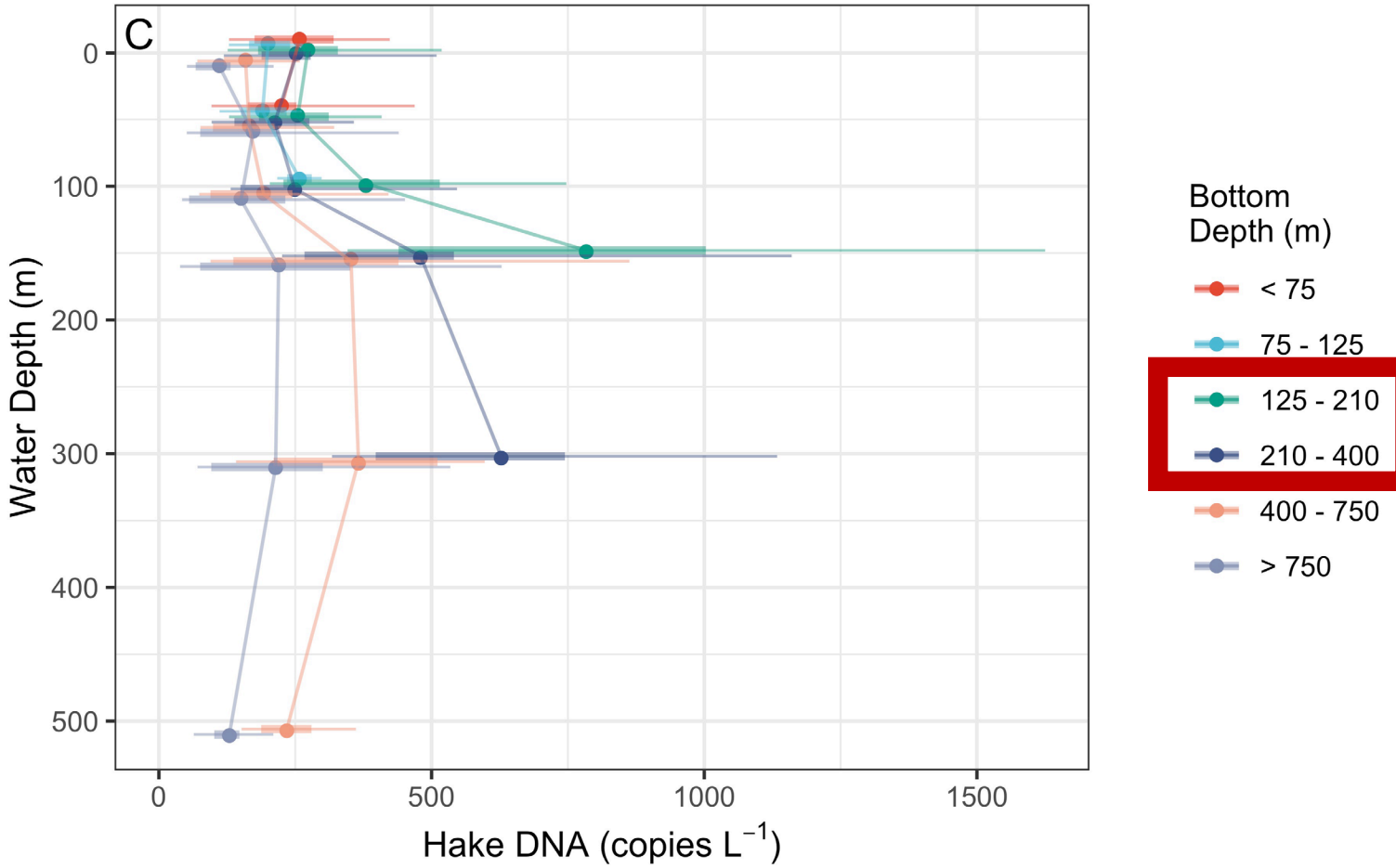


Depth-distribution of DNA correspond to published estimates for hake

Largest hake DNA concentrations:

Moderate water depths
-100-300m sampling depth

Along the continental shelf break
- 150-300m bottom depth



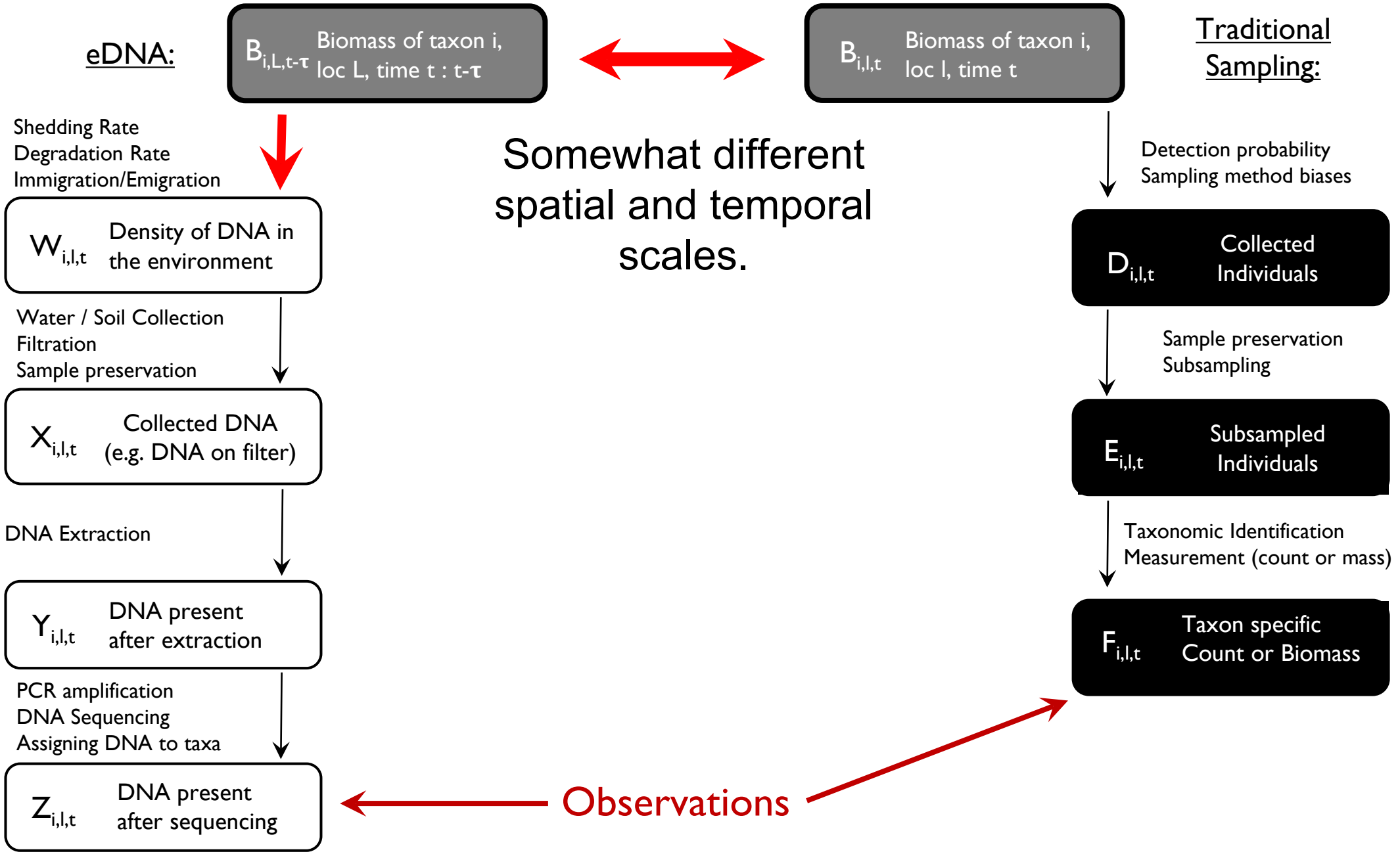
What do we know about how eDNA behaves in the field?

- DNA is not necessarily co-located with individuals
  - DNA stays with water, not taxa
  - DNA degrades on the order of hours

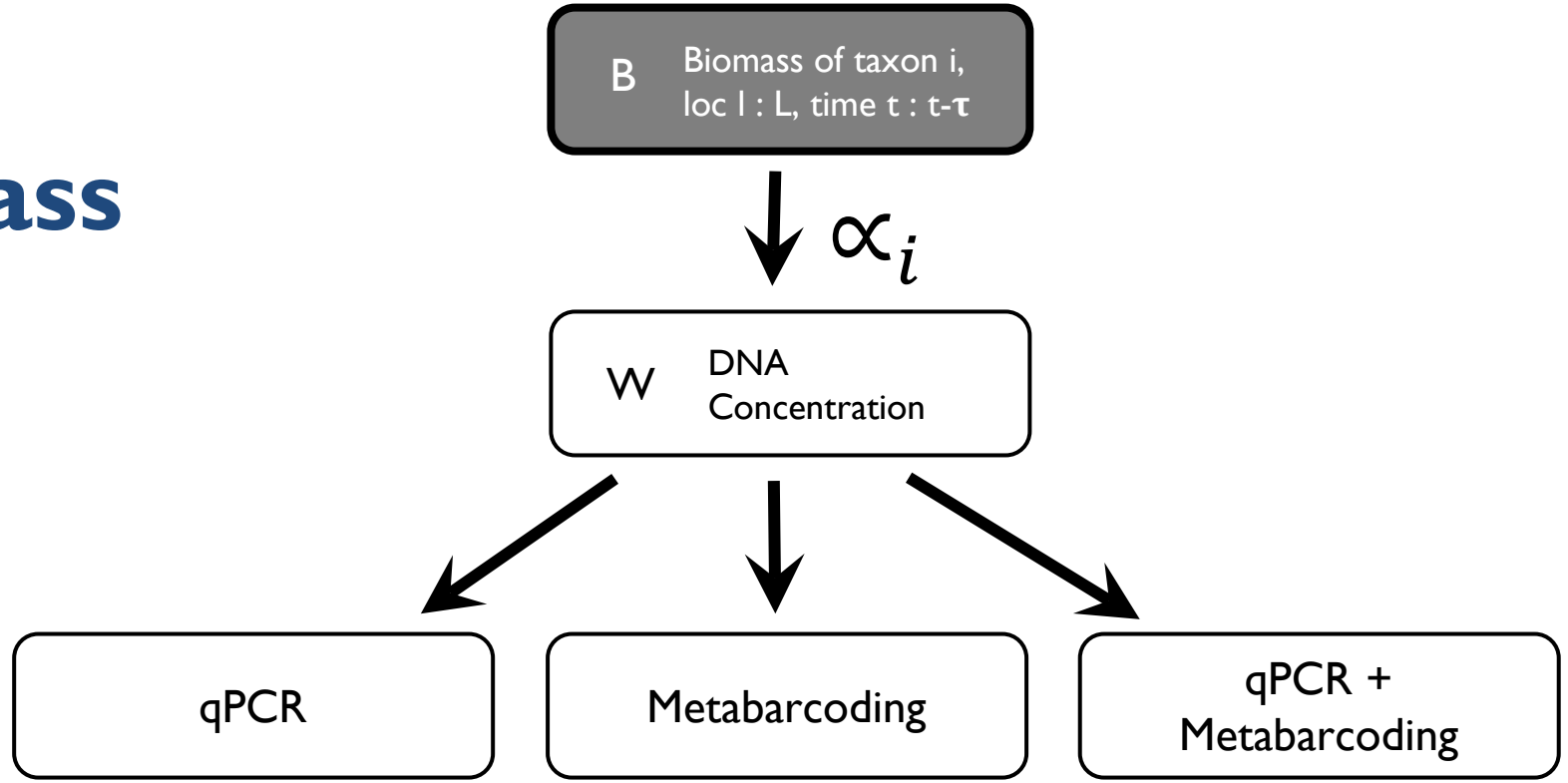
- Relationship between DNA conc. and biomass will likely vary across taxa to some extent.
  - **Production:** variability in amount of DNA produced per unit mass among taxa. Absolute magnitude of DNA shedding rates is generally unknown.
  - **Transport:** balance of the mobility of species and the flow environment

- At larger spatial scales, the relationship between biomass and eDNA improves markedly.

2026

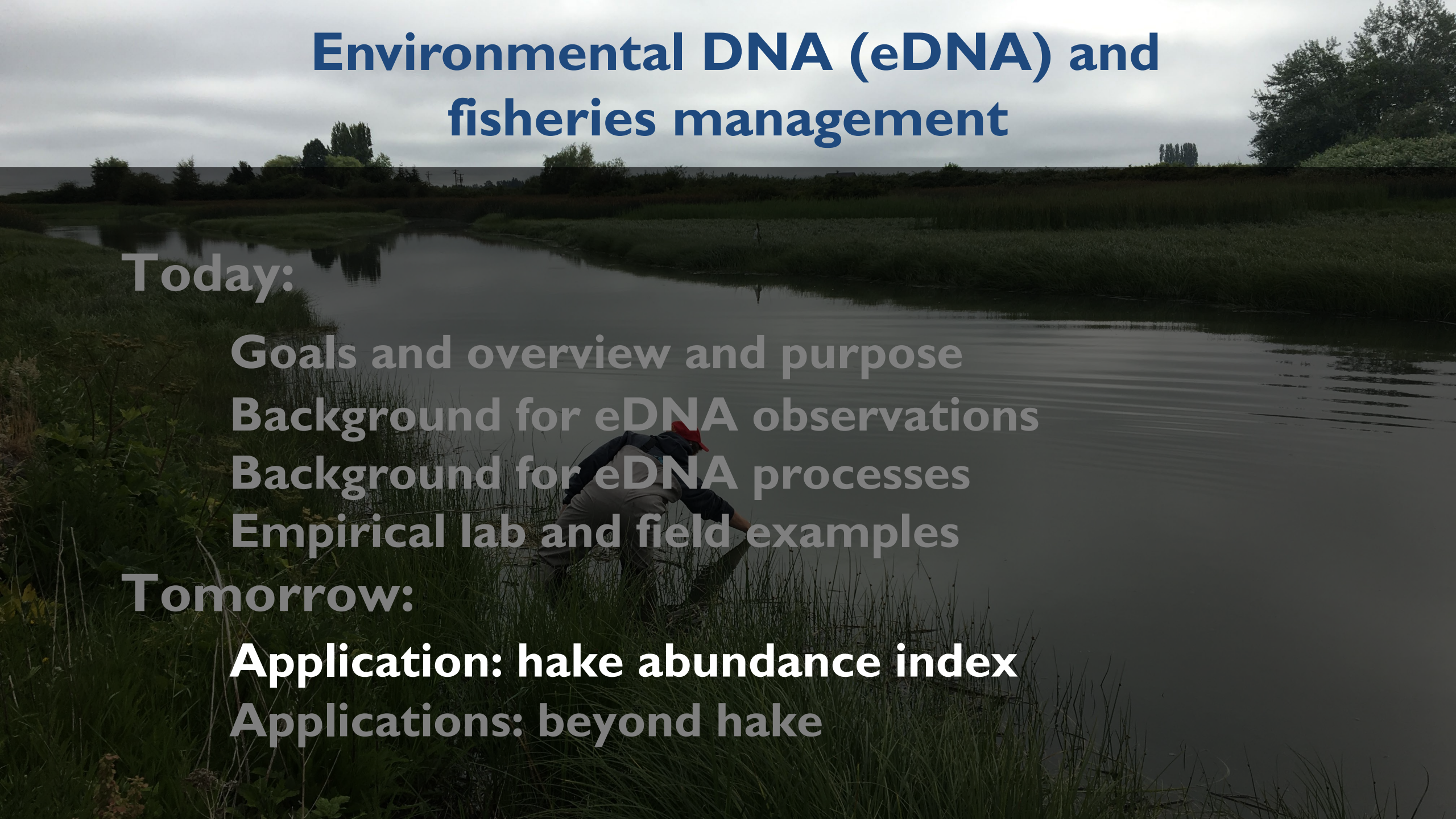

eDNA:
$B_{i,L,t-\tau}$ Biomass of taxon i, loc L, time t : t-τ
$B_{i,l,t}$ Biomass of taxon i, loc l, time t
Traditional Sampling:
Somewhat different spatial and temporal scales.
Shedding Rate
Degradation Rate
Immigration/Emigration
$W_{i,l,t}$ Density of DNA in the environment
Water / Soil Collection
Filtration
Sample preservation
$X_{i,l,t}$ Collected DNA (e.g. DNA on filter)
DNA Extraction
$Y_{i,l,t}$ DNA present after extraction
PCR amplification
DNA Sequencing
Assigning DNA to taxa
$Z_{i,l,t}$ DNA present after sequencing
Observations
Detection probability
Sampling method biases
$D_{i,l,t}$ Collected Individuals
Sample preservation
Subsampling
$E_{i,l,t}$ Subsampled Individuals
Taxonomic Identification
Measurement (count or mass)
$F_{i,l,t}$ Taxon specific Count or Biomass

eDNA to inform biomass



- Need adequate sampling (space, time) and well-defined target.
- Provides relative abundance estimates.
- Need for assumptions and/or sensitivity checks
  - spatial / temporal variation in loss rates
  - age / size structure effects on DNA production

Environmental DNA (eDNA) and fisheries management



Today:

- Goals and overview and purpose
- Background for eDNA observations
- Background for eDNA processes
- Empirical lab and field examples

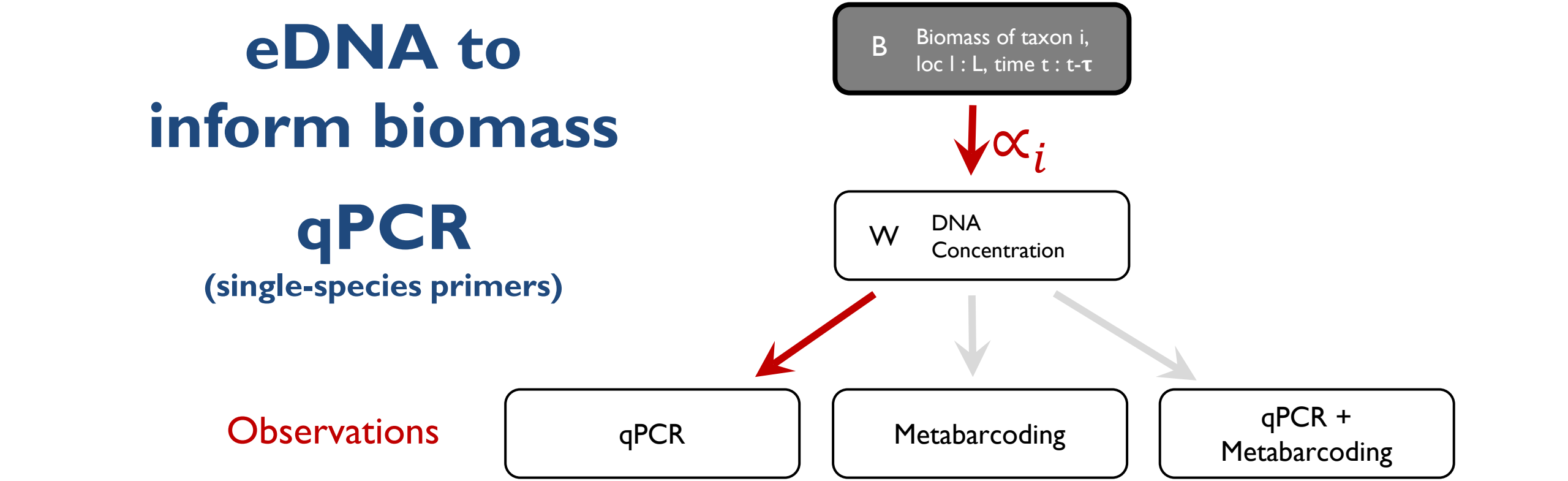
Tomorrow:

- **Application: hake abundance index**
- Applications: beyond hake

eDNA to inform biomass

qPCR

(single-species primers)

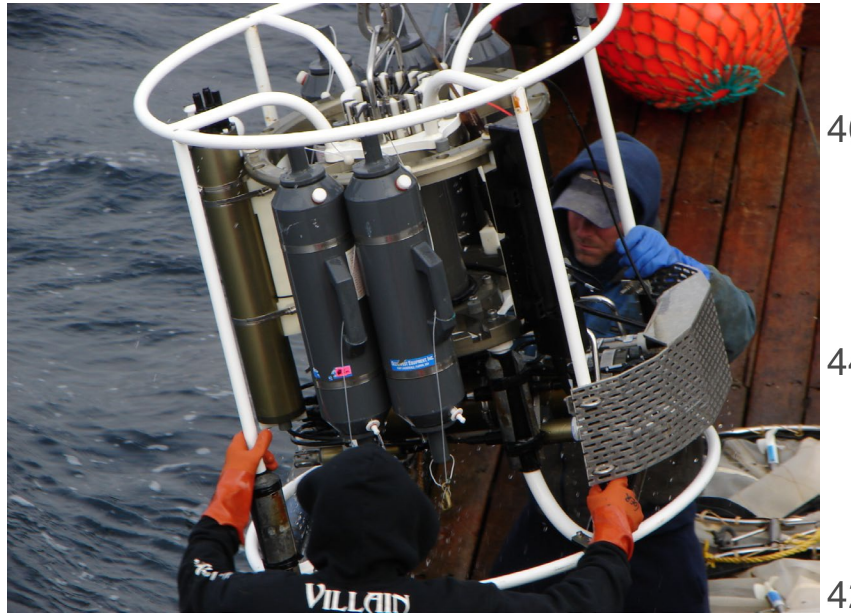


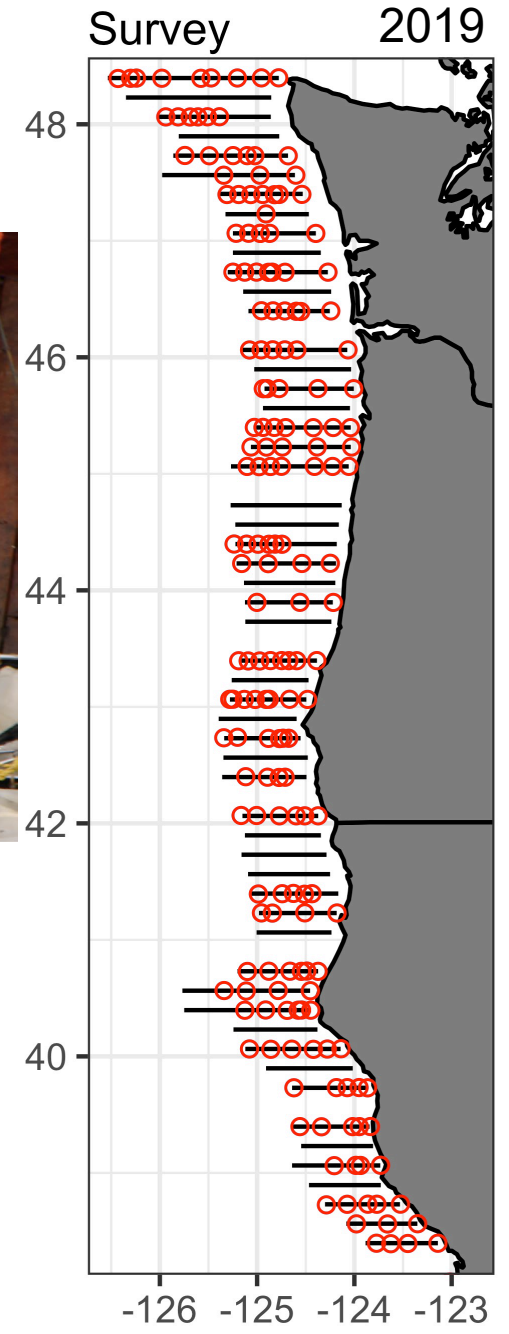
Observations



eDNA (2019, 2021, 2023, 2025, …)

- >180 stations sampled each year
- 2 x 2.5 L seawater filtered from 6 depths
  - > 1800 bottles sampled annually.
  - depths vary somewhat among years (to 500m)
- Assayed with Single-species qPCR primer
  - Pacific hake DNA concentration
- Acoustics and DNA not collected simultaneously (sample-to-sample comparisons are not possible)
- A range of controls to check for contamination

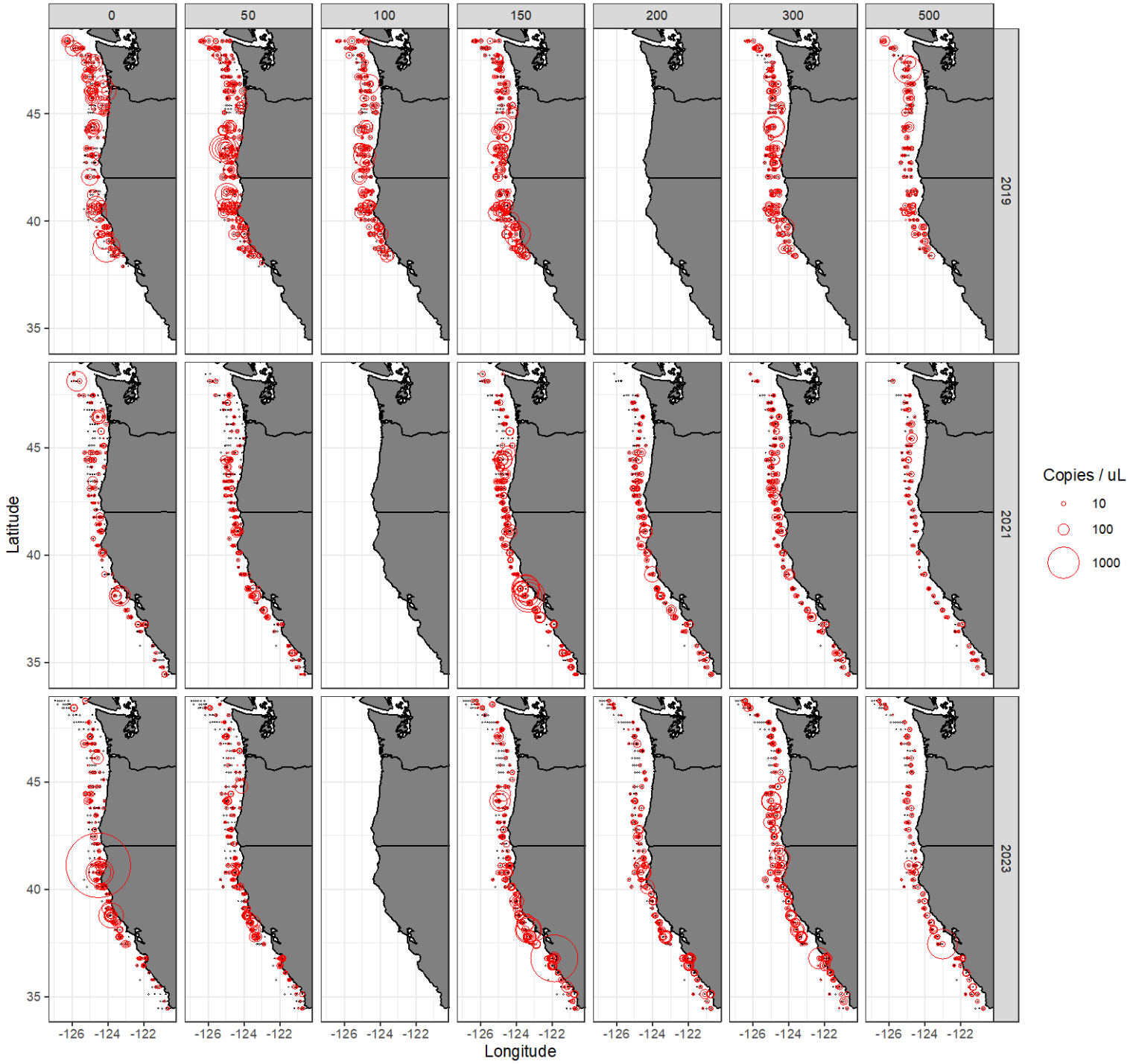




Largest eDNA sampling effort in the world

Hake DNA concentration

(all depths, 2019-2023)



Fisheries Index Checklist

- **Spatial :**
  - Did we sample the full range (N-S, E-W, depth) of the population?
  - *No on N-S (Canada). Otherwise, yes. We did sample the core habitats.*
- **Temporal :**
  - Did we sample at the right time of year?
  - *Yes. Paired with the acoustic survey*
- **Sample attributes:**
  - Did we take enough samples?
  - *eDNA index values have reasonable CVs… comparable to Acoustic CVs*
  - Was each sample large enough?
  - *Relatively few zero observation, few extremely high values. Relatively low bottle to bottle variability*
- **Closure assumptions met?**
  - Did a bunch of hake leave the area between the start and end of the survey?
  - *We assume no.*

Building an index for hake from eDNA

Treat eDNA independently from other information

eDNA Components:

Field samples
Standards (calibrate qPCR measurements)
Controls (checks for possible contamination)

Spatio-temporal statistical model

Projections, defining index areas, and summations

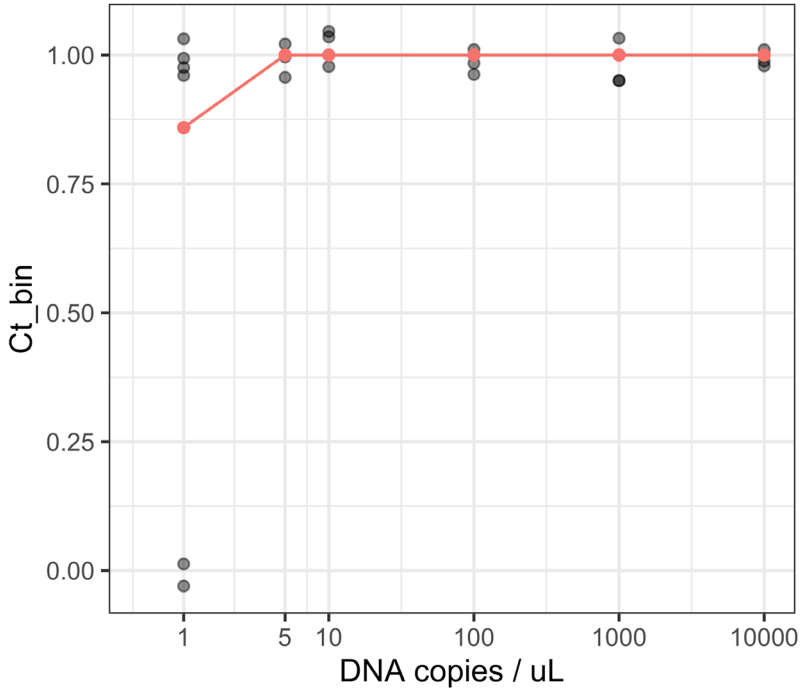
Building an index for hake from eDNA

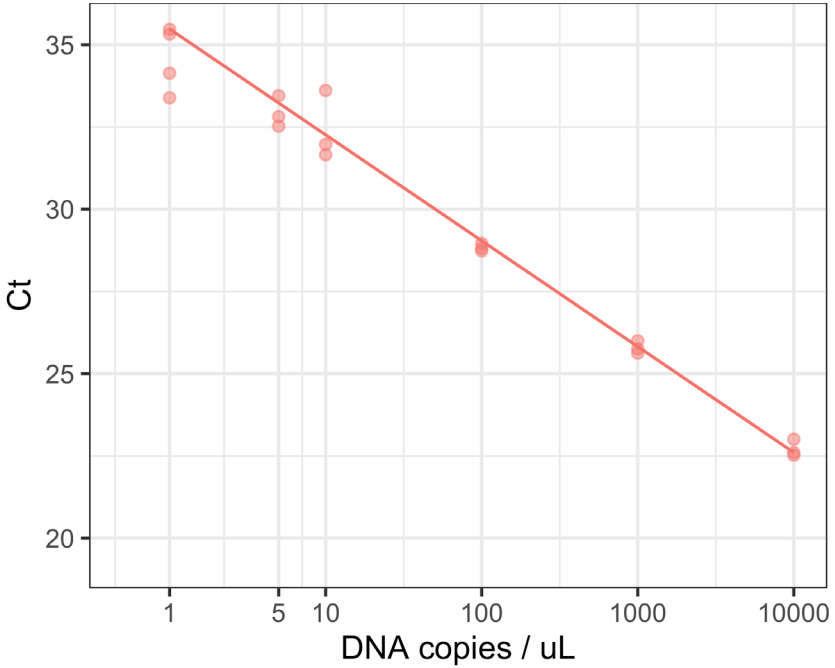
DNA Components:

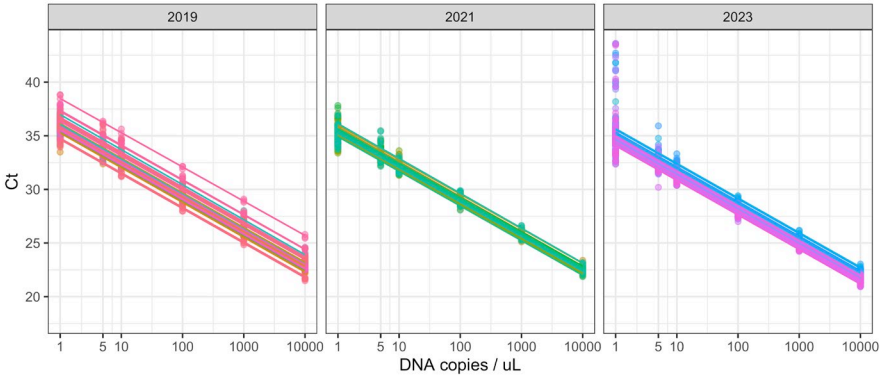
Field samples

Standards (calibrate qPCR measurements)

Controls (checks for possible contamination)







Building an index for hake from eDNA

Spatio-temporal statistical model

Fixed effects

- year-intercepts, smooth with bottom depth, sample processing in 2019.

Offsets

- dilutions due to PCR inhibition, water volume filtered

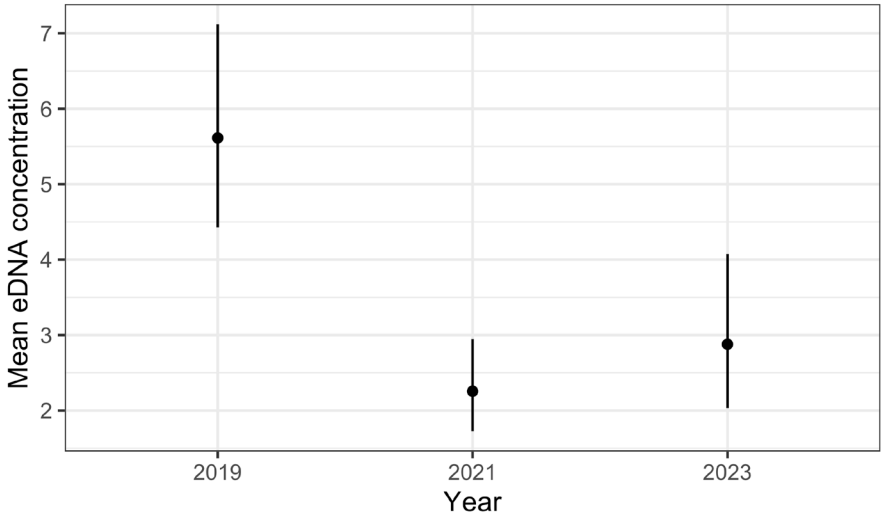
Random effects

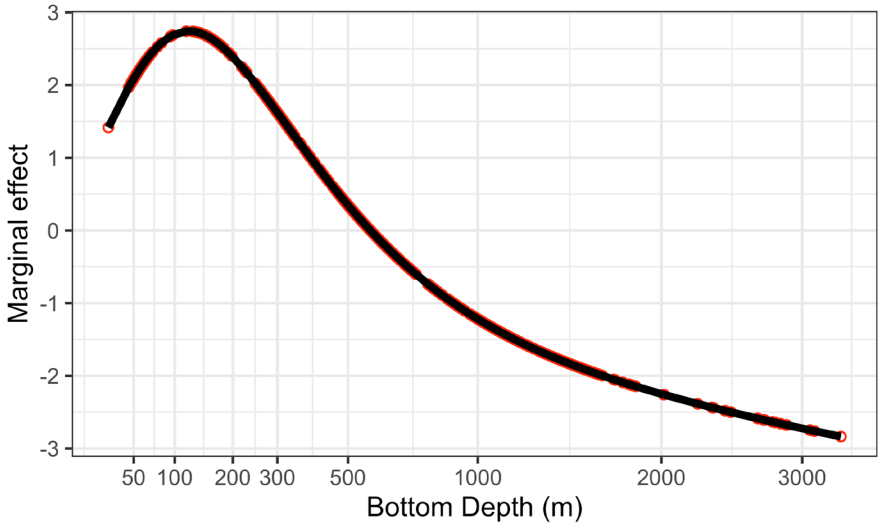
- spatial factors to generate fields for each year x depth
    - SPDE – INLA approach
- bottle effects for concentrations at year-station-depth

Estimated in TMB

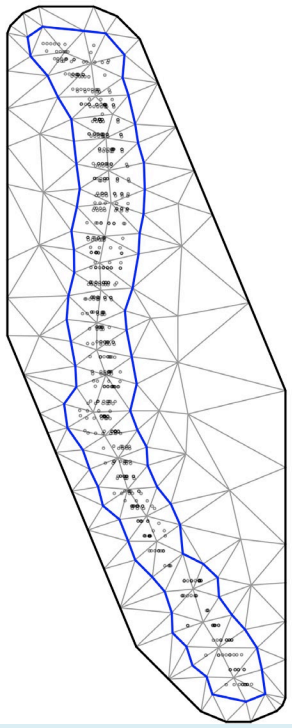
(maximum likelihood, uncertainties derived from the delta-method, convergence diagnostics, etc.)

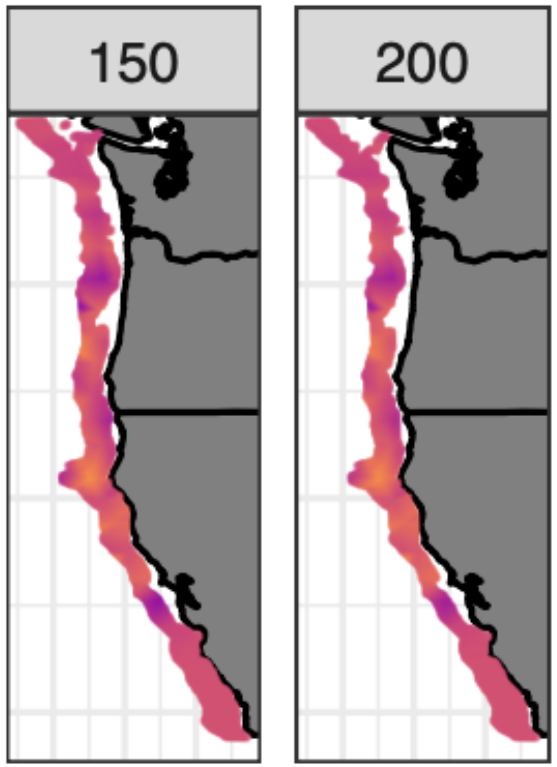
Fixed

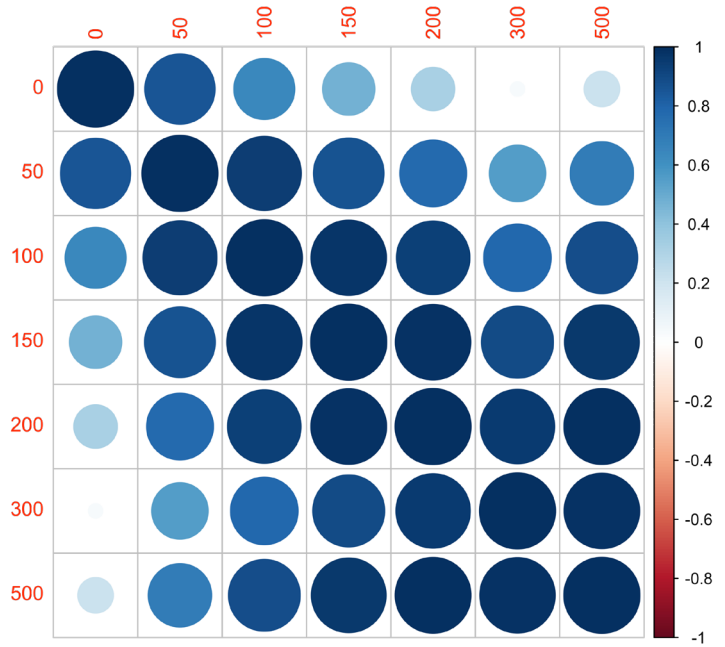


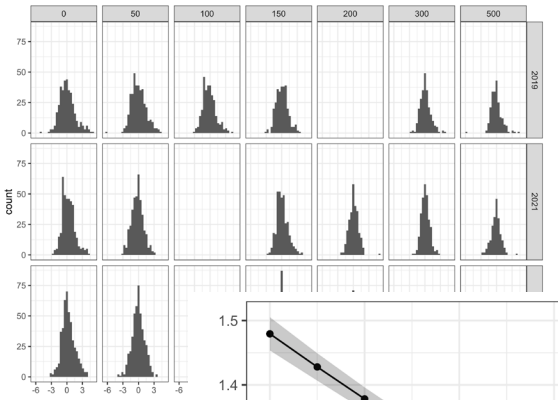


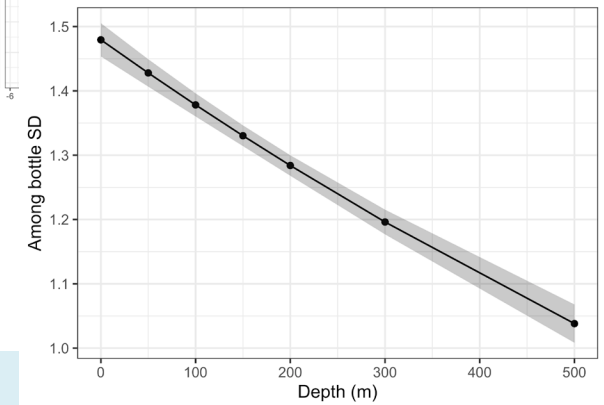
Random





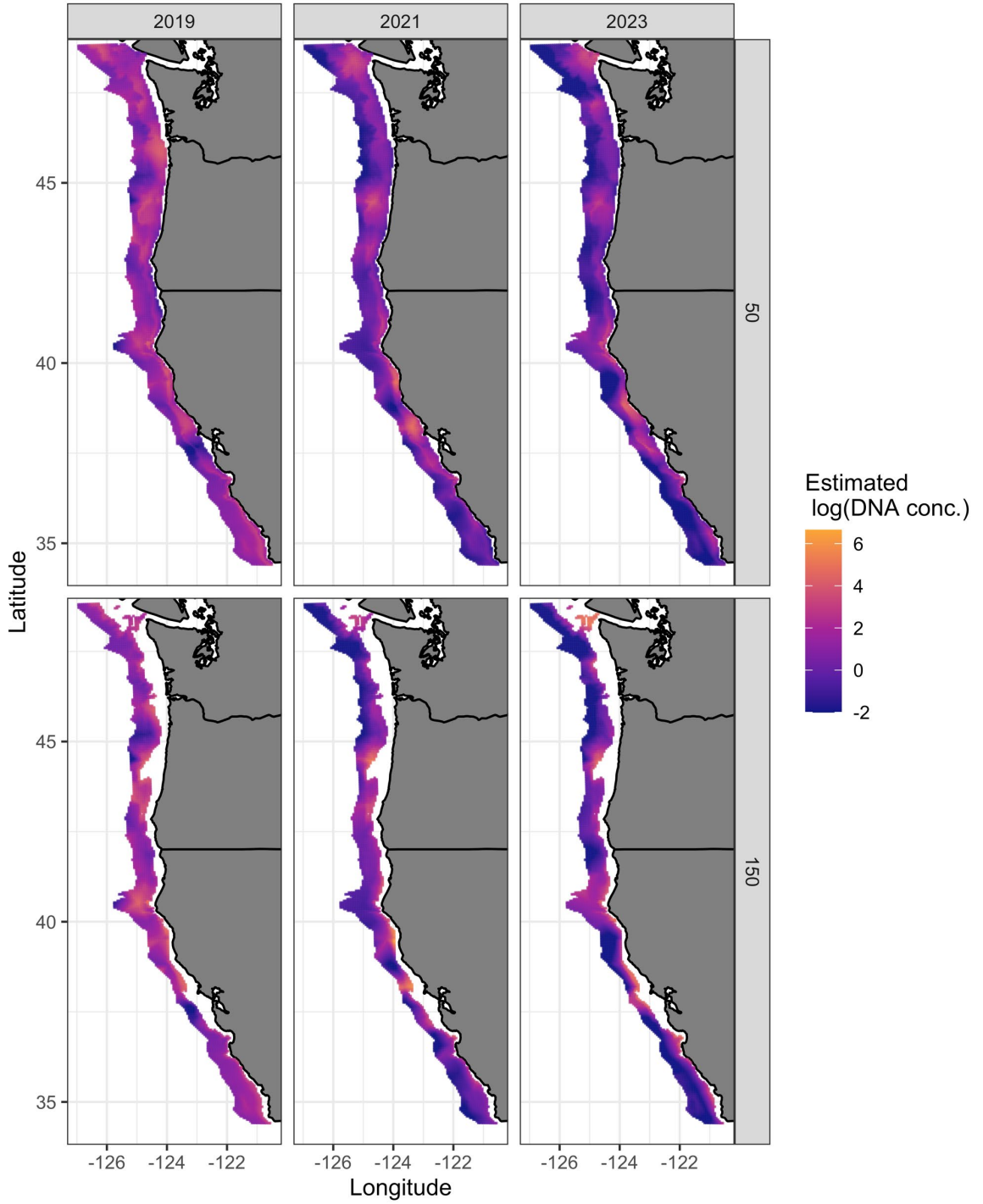






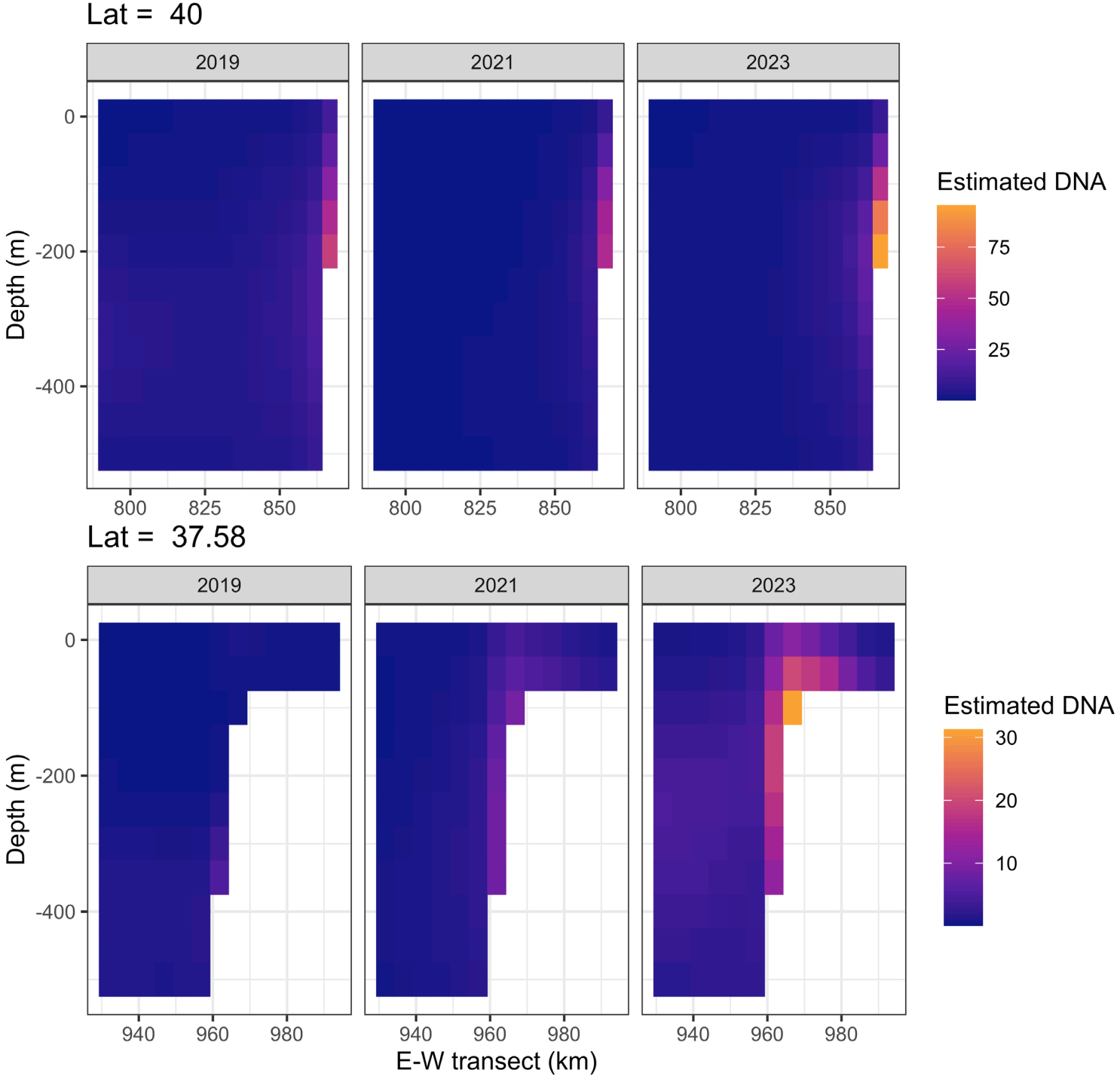
Make Predictions

5km grid every 50m



Make Predictions

East-West Transects

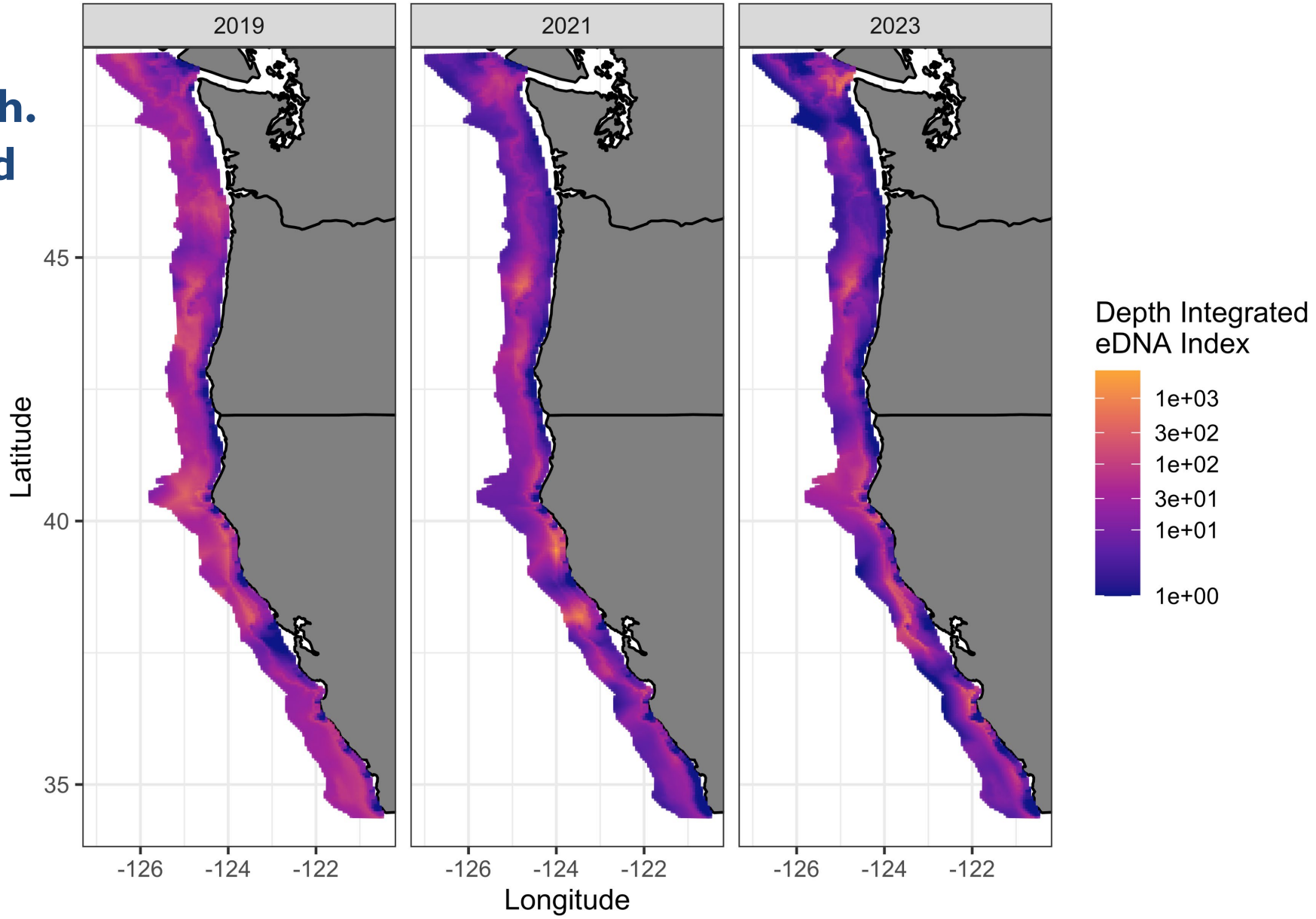


Sum to Surface

5km grid, every 50m depth. Sum between surface and 500m

Unusual units:
DNA copies per L per 25km^2 of ocean column

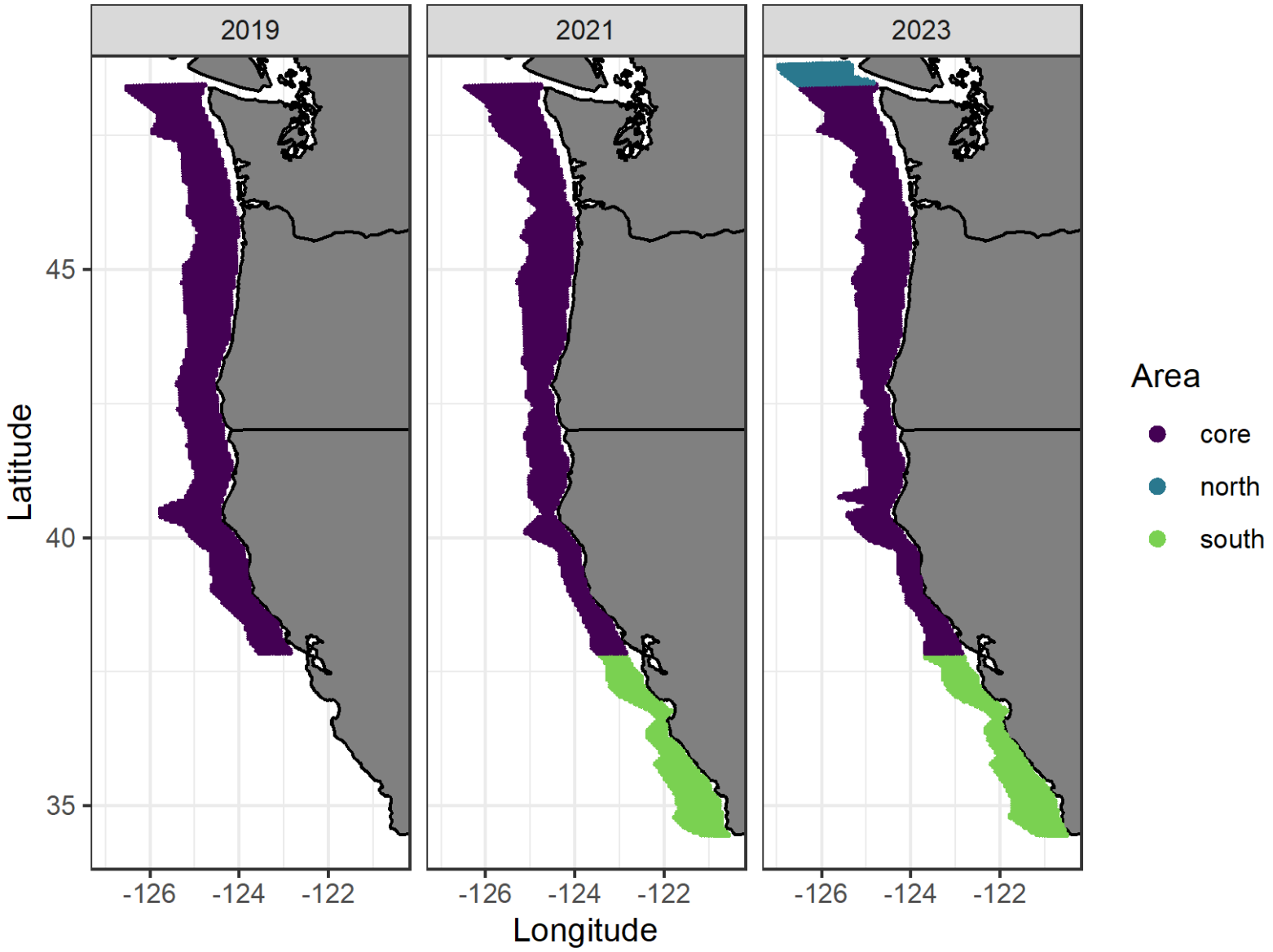
Likely better to move to predicted total DNA copies



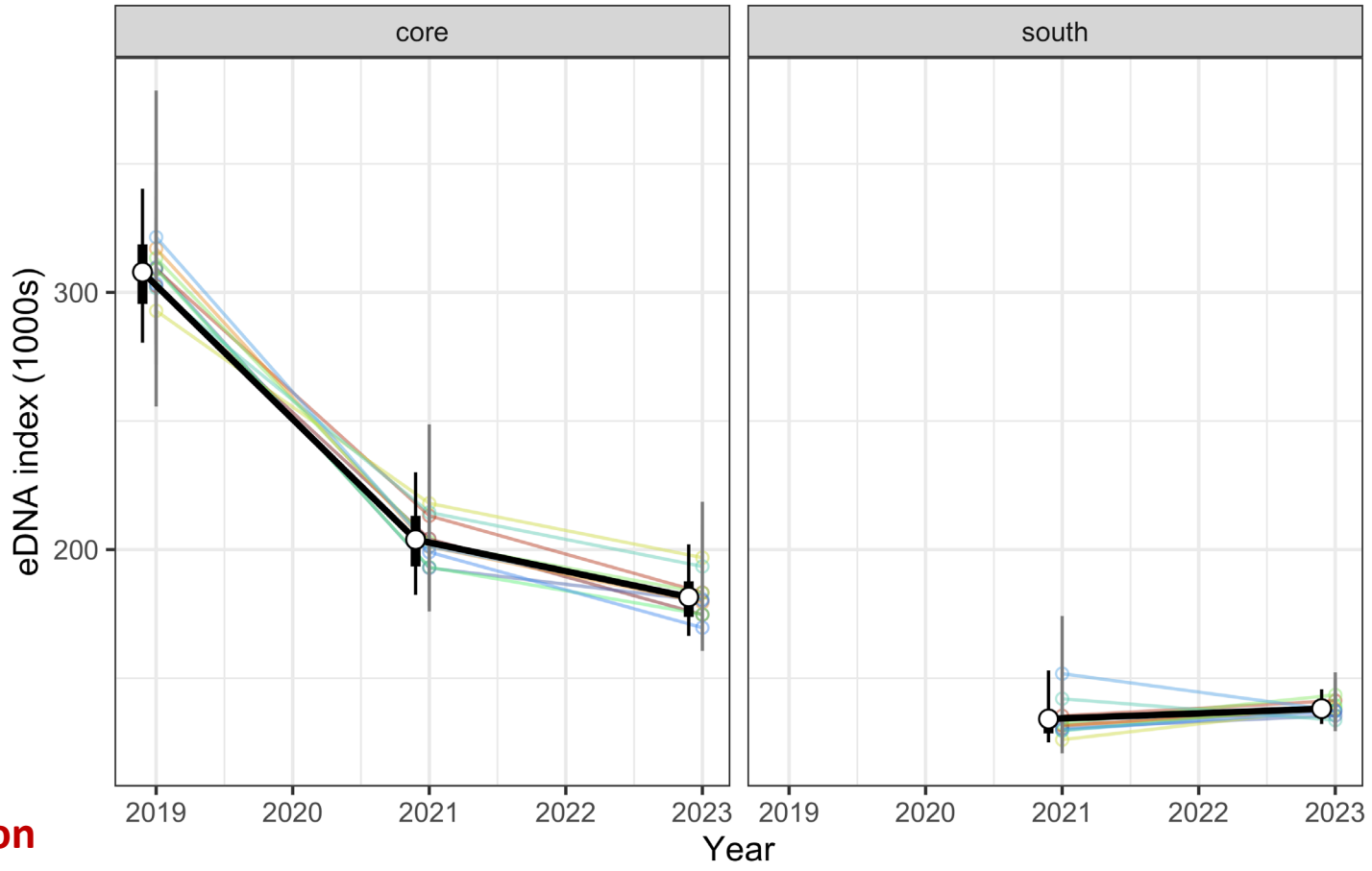
Survey area varies among years

Eastern-Westen edge defined by acoustic-trawl survey.

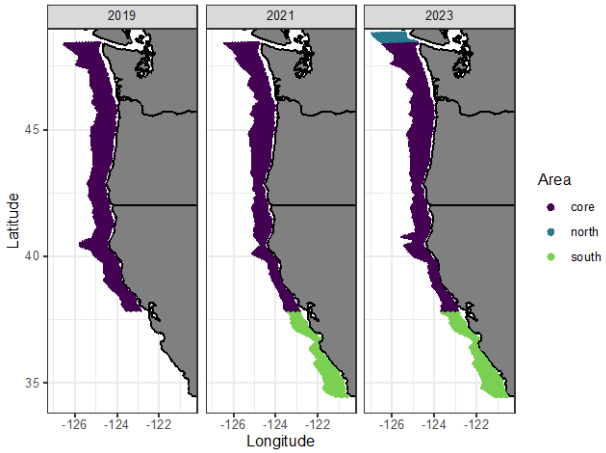
North-South edges defined by areas sampled for eDNA.



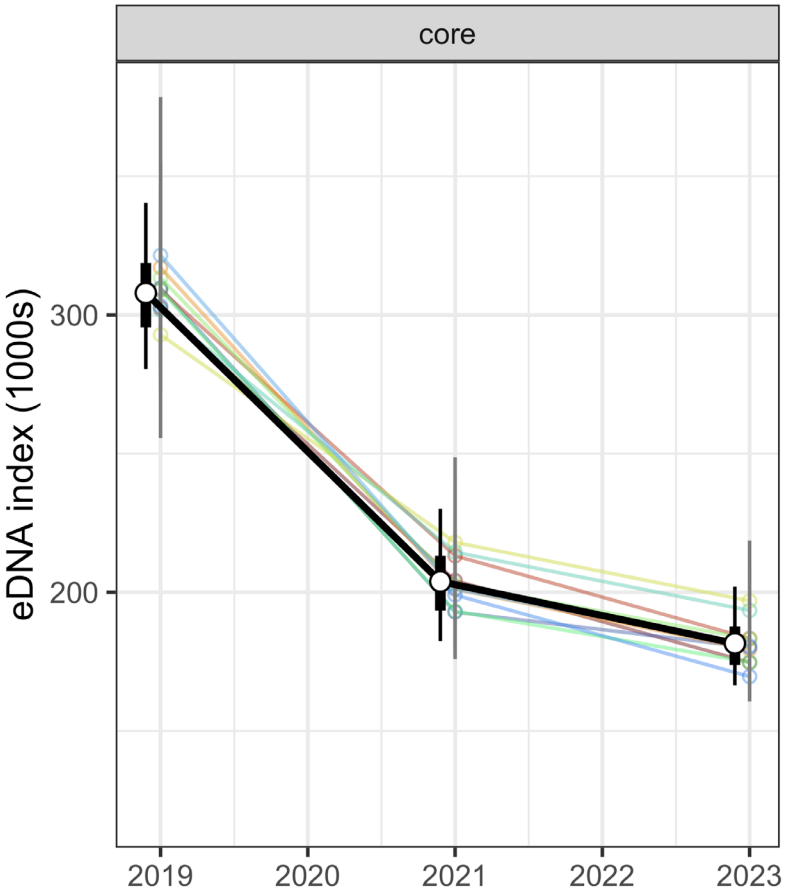
Survey area varies among years

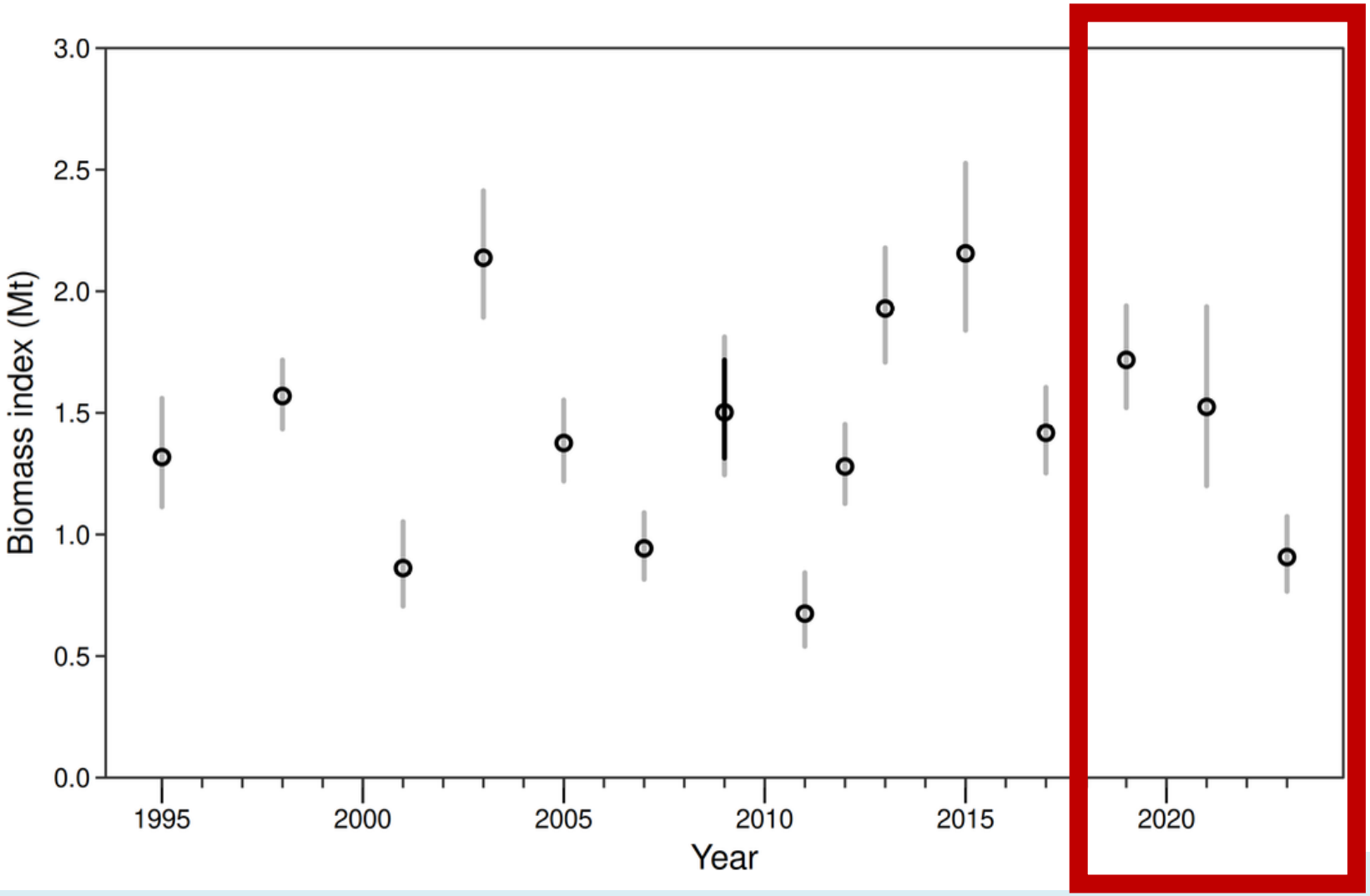


∝ DNA Concentration



Acoustics and eDNA suggest a ~40% decline between 2019 and 2023





We have an index.
Now what?

Two general issues for inclusion in an assessment:

Selectivity

Catchability

Catchability

(the relative proportion of a fish population that is sampled by a survey)

Scales the units of the eDNA index to population biomass

2025 Hake assessment estimated catchability within the model

Deserves additional thought and research

- Should reflect the uncertainty in the biomass to DNA concentration proportionality
- Perhaps this proportionality is partially measurable with experiments?
- Any reason for time-varying Catchability?

Standardized sensitivities to investigate for any new assessment?

Selectivity

Do not currently have a way to identify attributes of the DNA source

- age, sex, maturity, etc.

Hake assessment Implemented knife-edge selectivity (ages 1+)

Deserves additional thought and research

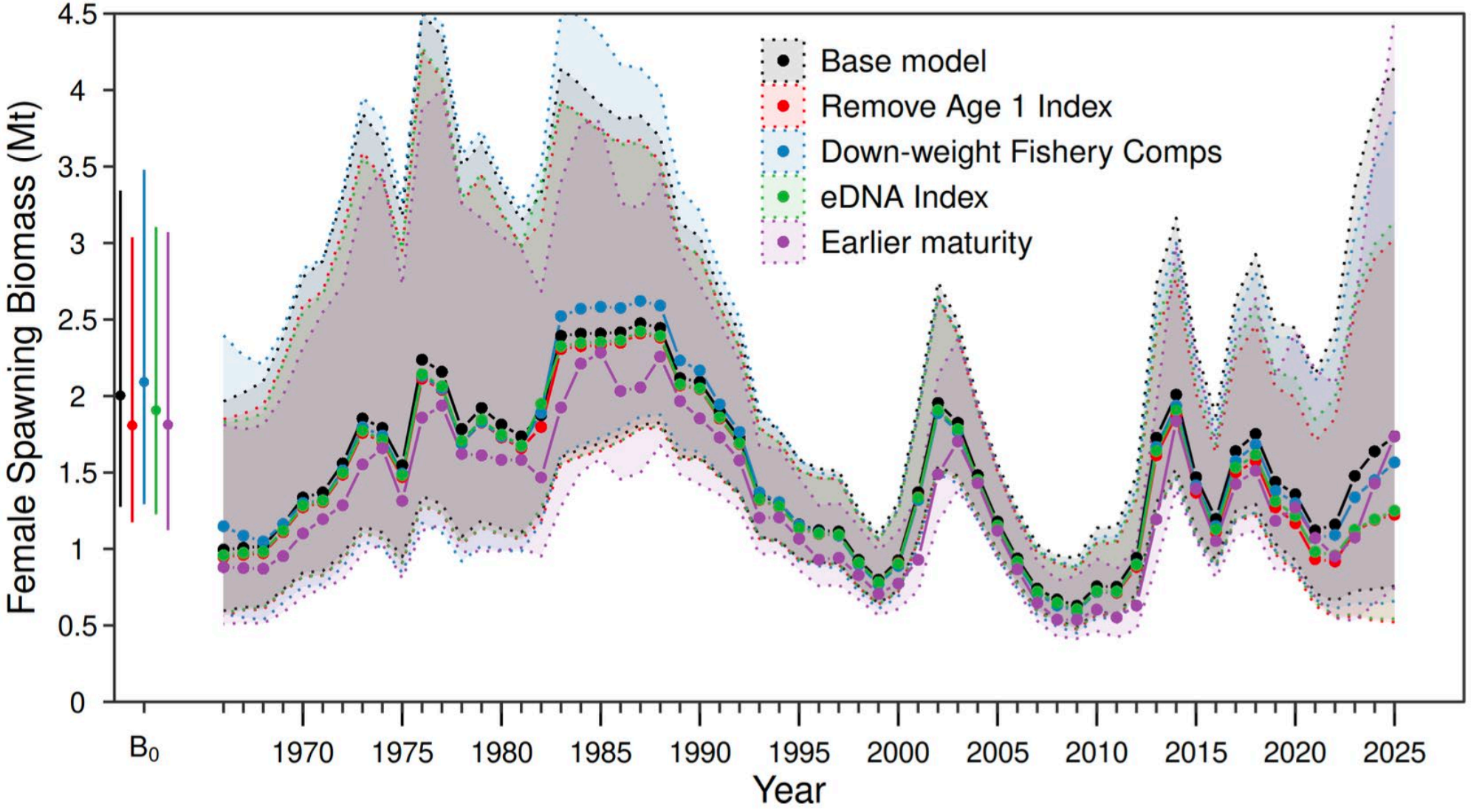
Some literature support for eDNA as a function of surface area, not biomass

Faster growing individuals over-represented in DNA?

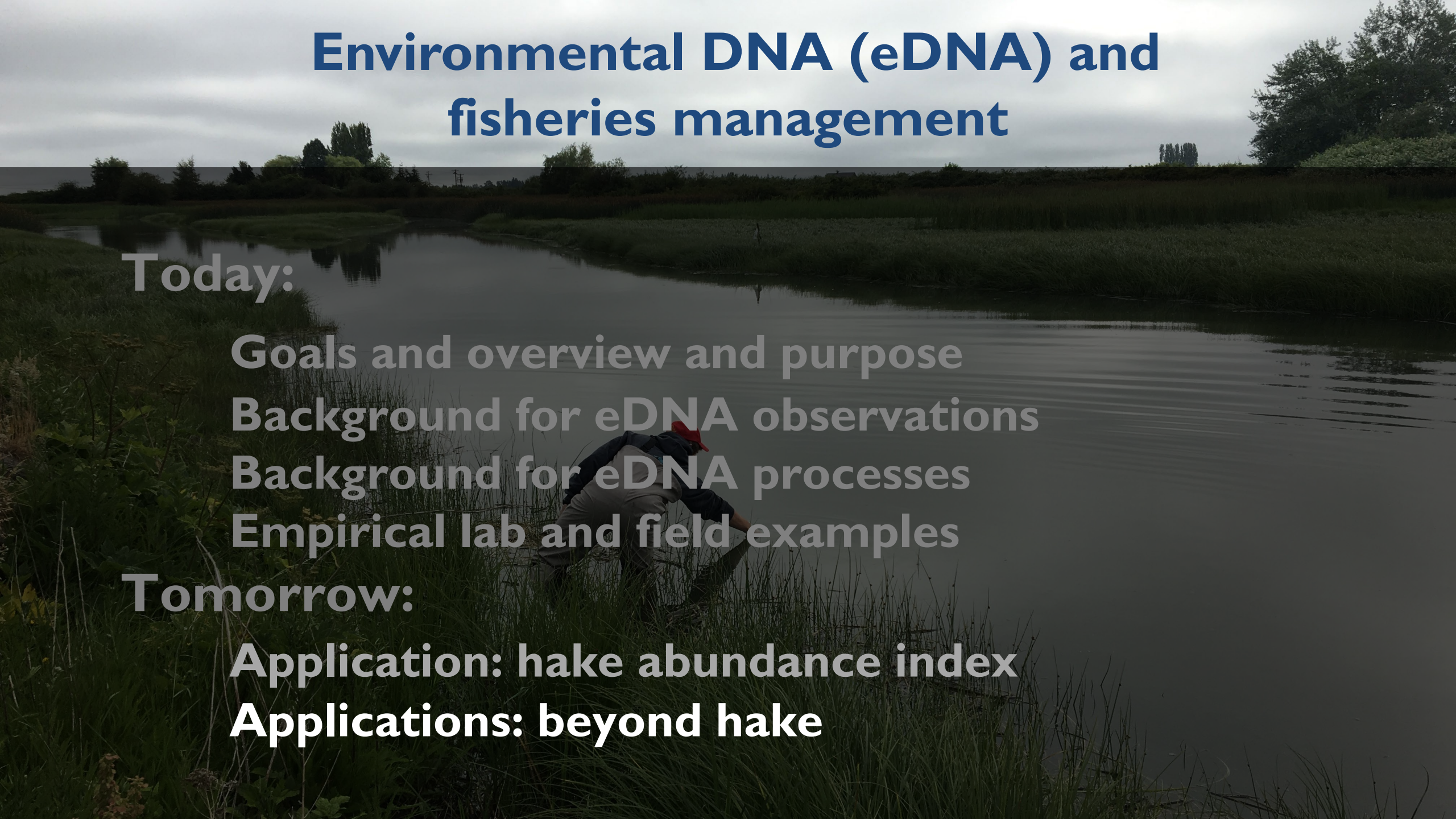
Any reason for time-varying Selectivity?

Standardized sensitivities to investigate for any new assessment?

eDNA Index as a sensitivity



Environmental DNA (eDNA) and fisheries management



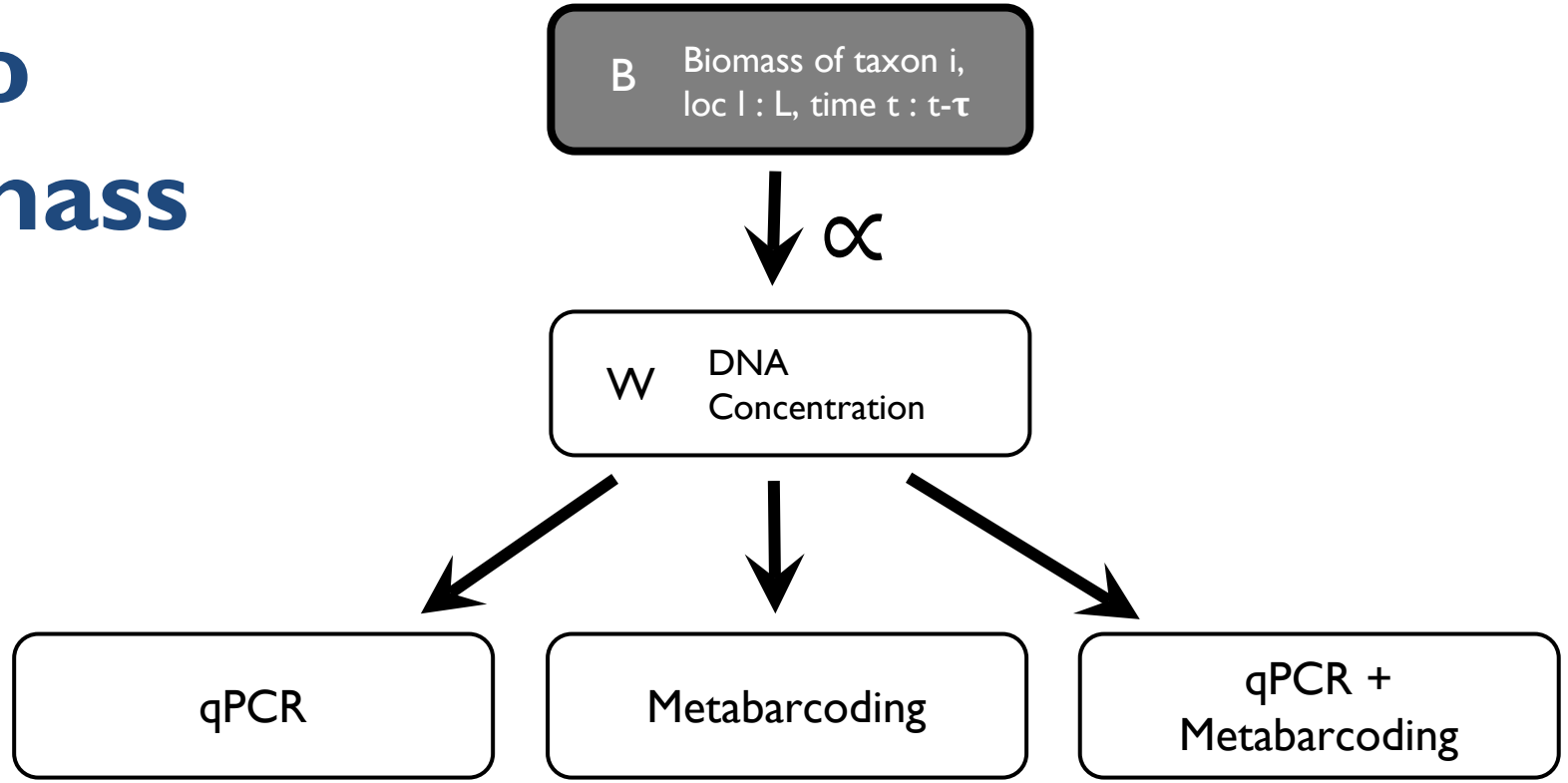
Today:

- Goals and overview and purpose
- Background for eDNA observations
- Background for eDNA processes
- Empirical lab and field examples

Tomorrow:

- Application: hake abundance index
- Applications: beyond hake

eDNA to inform biomass

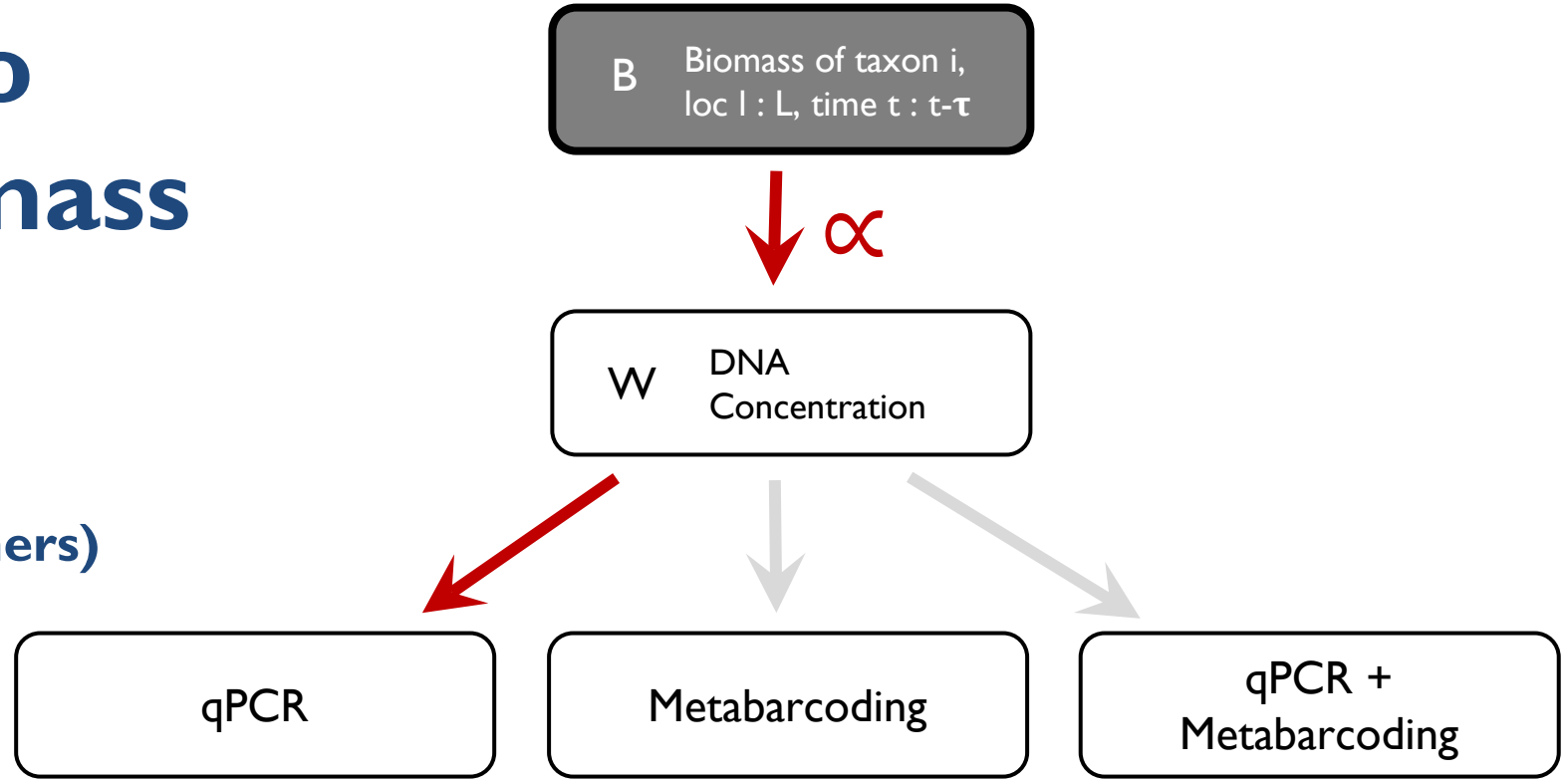


Observations

eDNA to inform biomass

qPCR
(single-species primers)

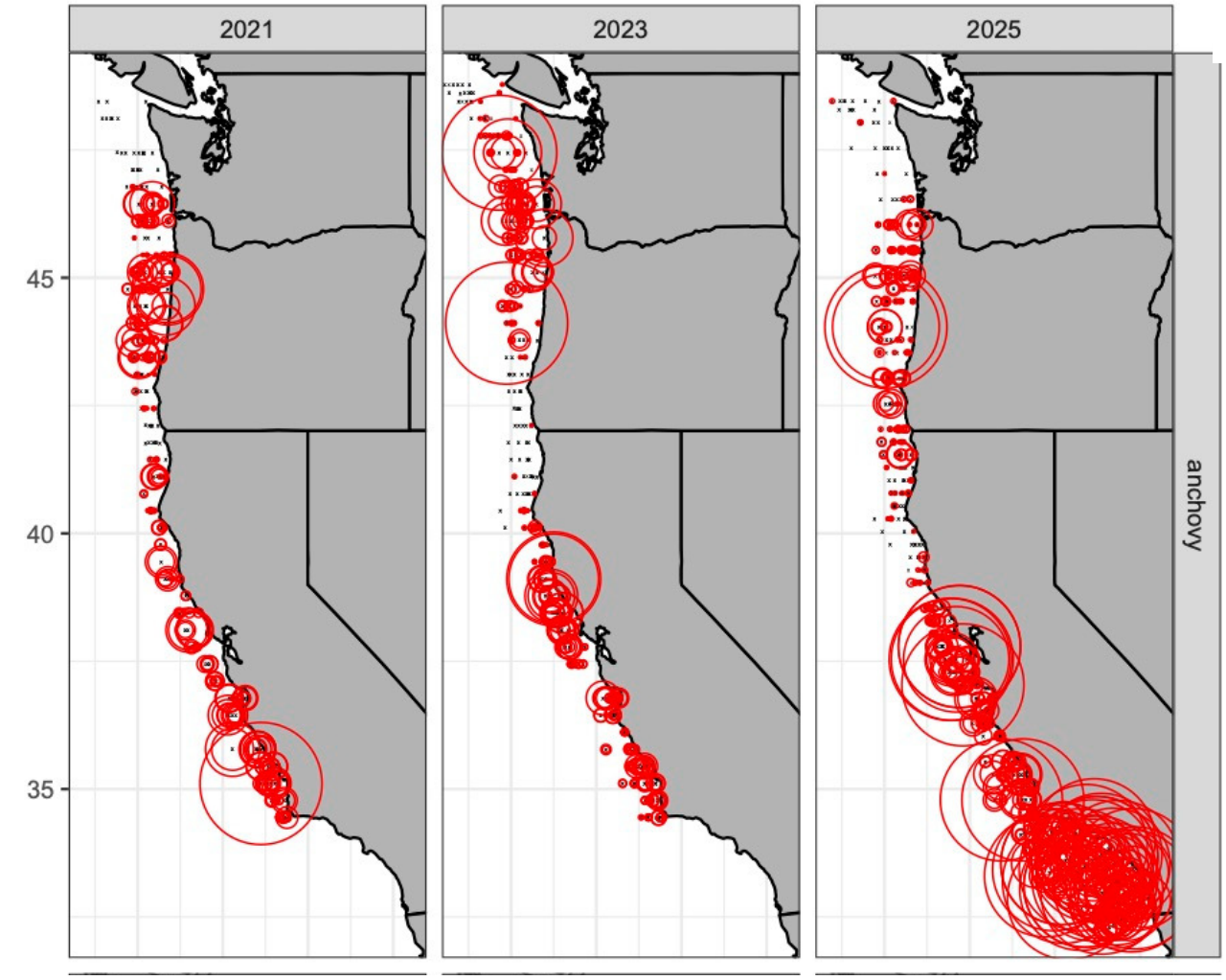
Observations

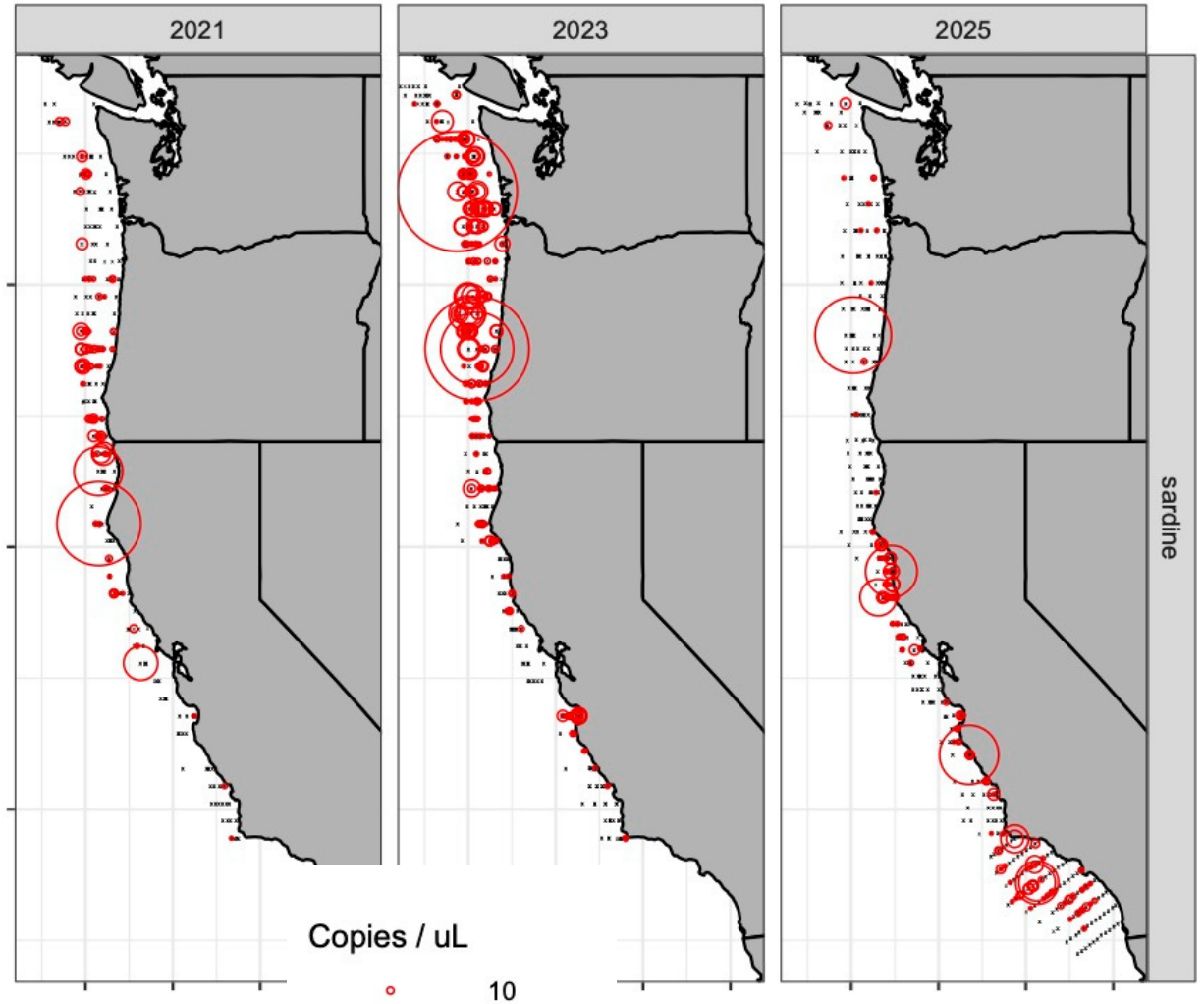


- Need adequate sampling (space, time) and well-defined target.
- qPCR directly measures concentration.
- Make indices to your heart's content.

  Hake, Sardines, Anchovy

CPS





Copies / uL

10

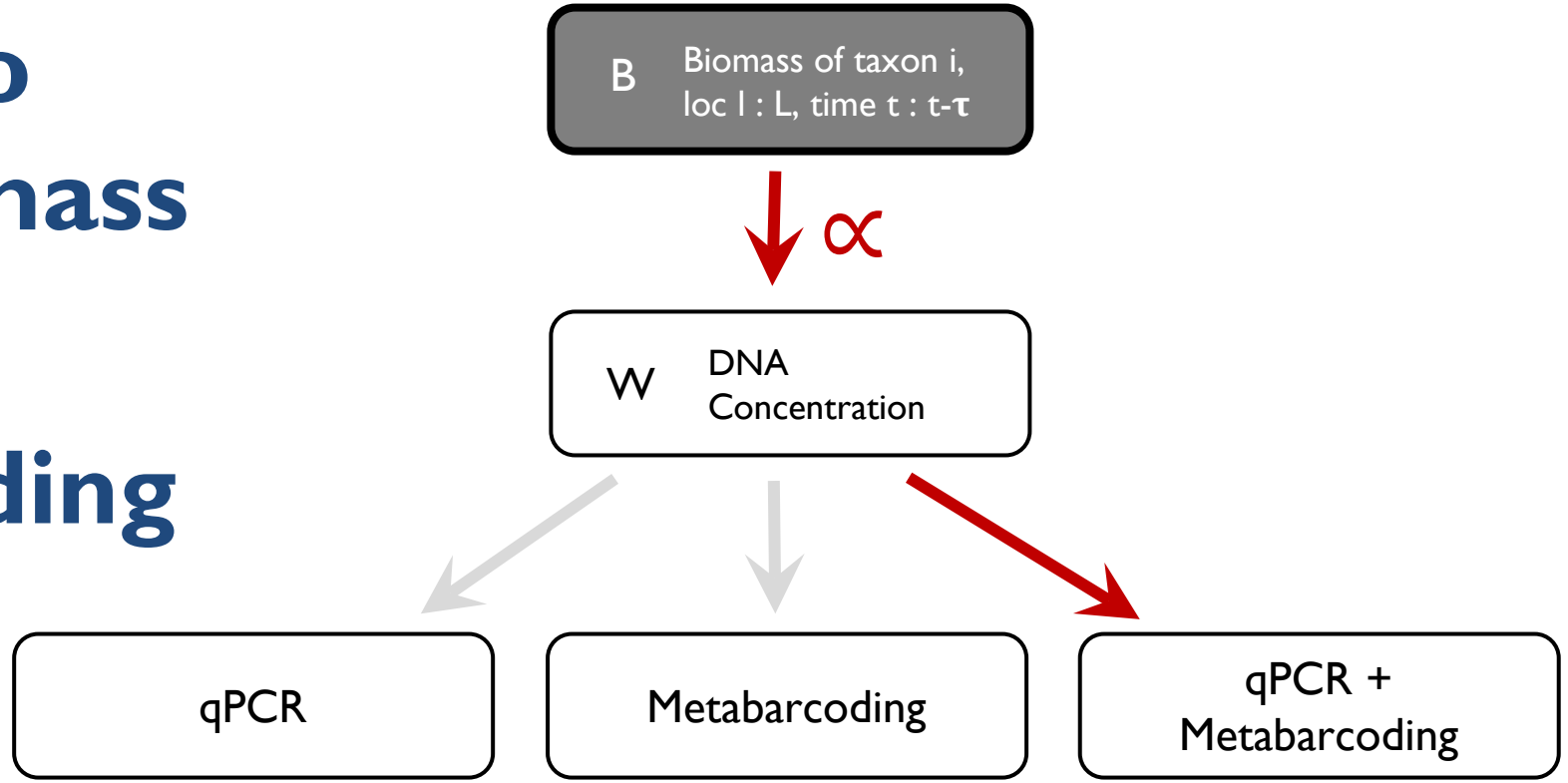
100

1000

10000

eDNA to inform biomass

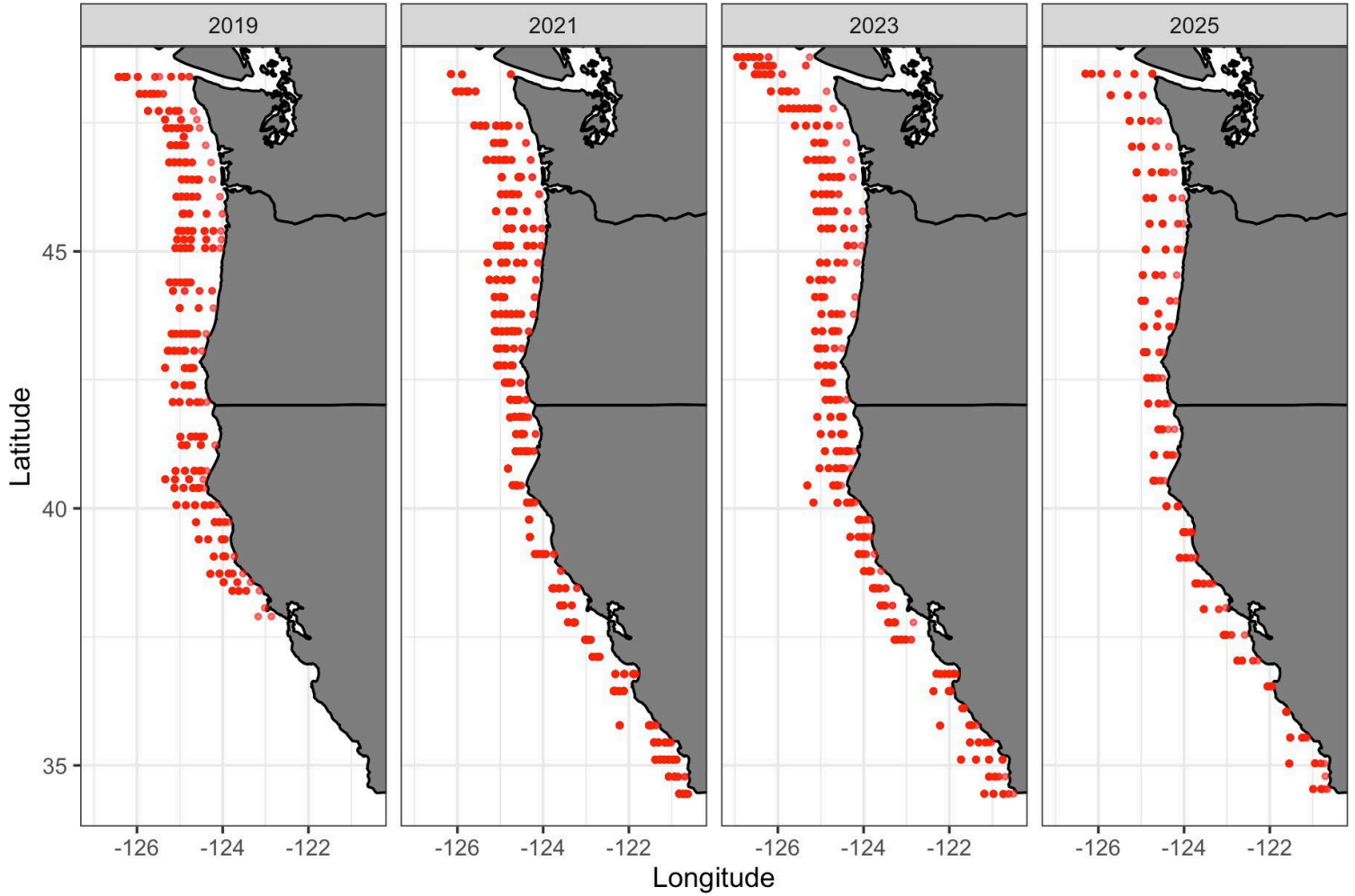
qPCR + Metabarcoding



- Need adequate sampling (space, time) and well-defined target.
- qPCR directly measures concentration.
- Metabarcoding provides relative proportions.
  - Unreplicated or Replicated metabarcoding.
  - Some caveats.

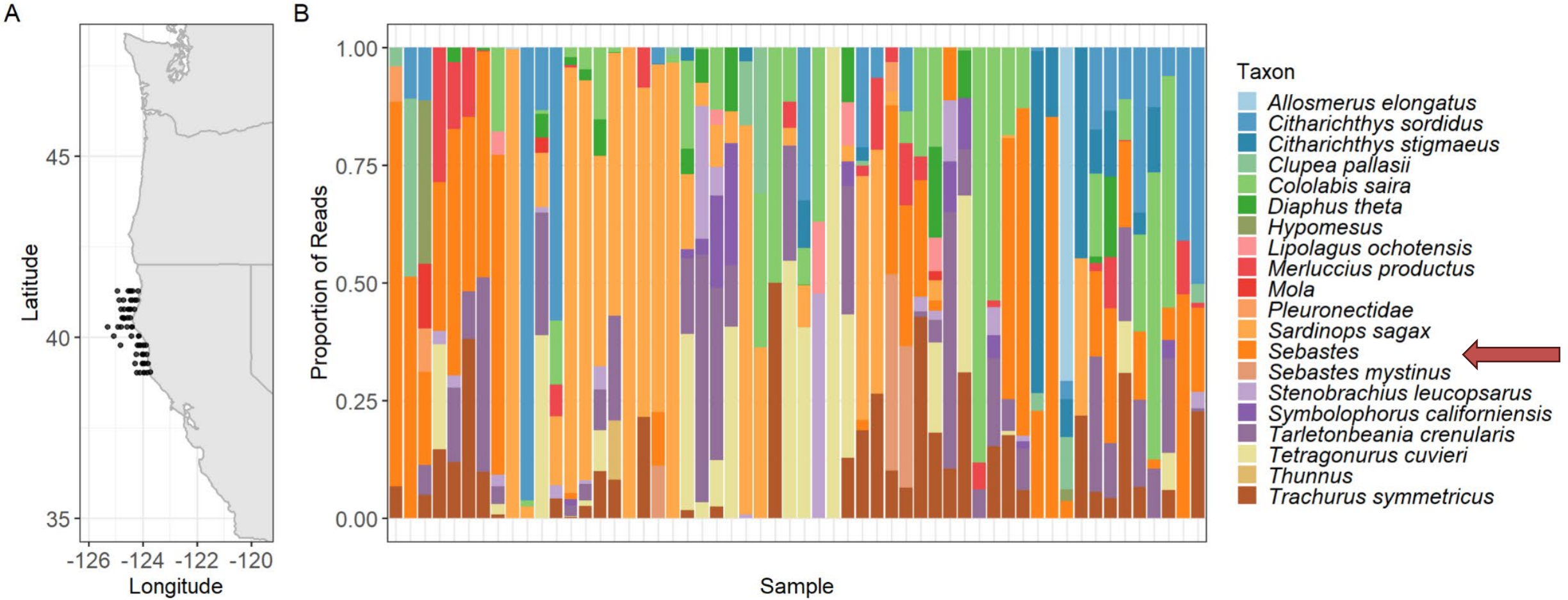
Hake sample stations 2019-2025

- ~2000 samples / year
- Surface to 500m.
- All >50m bottom depth



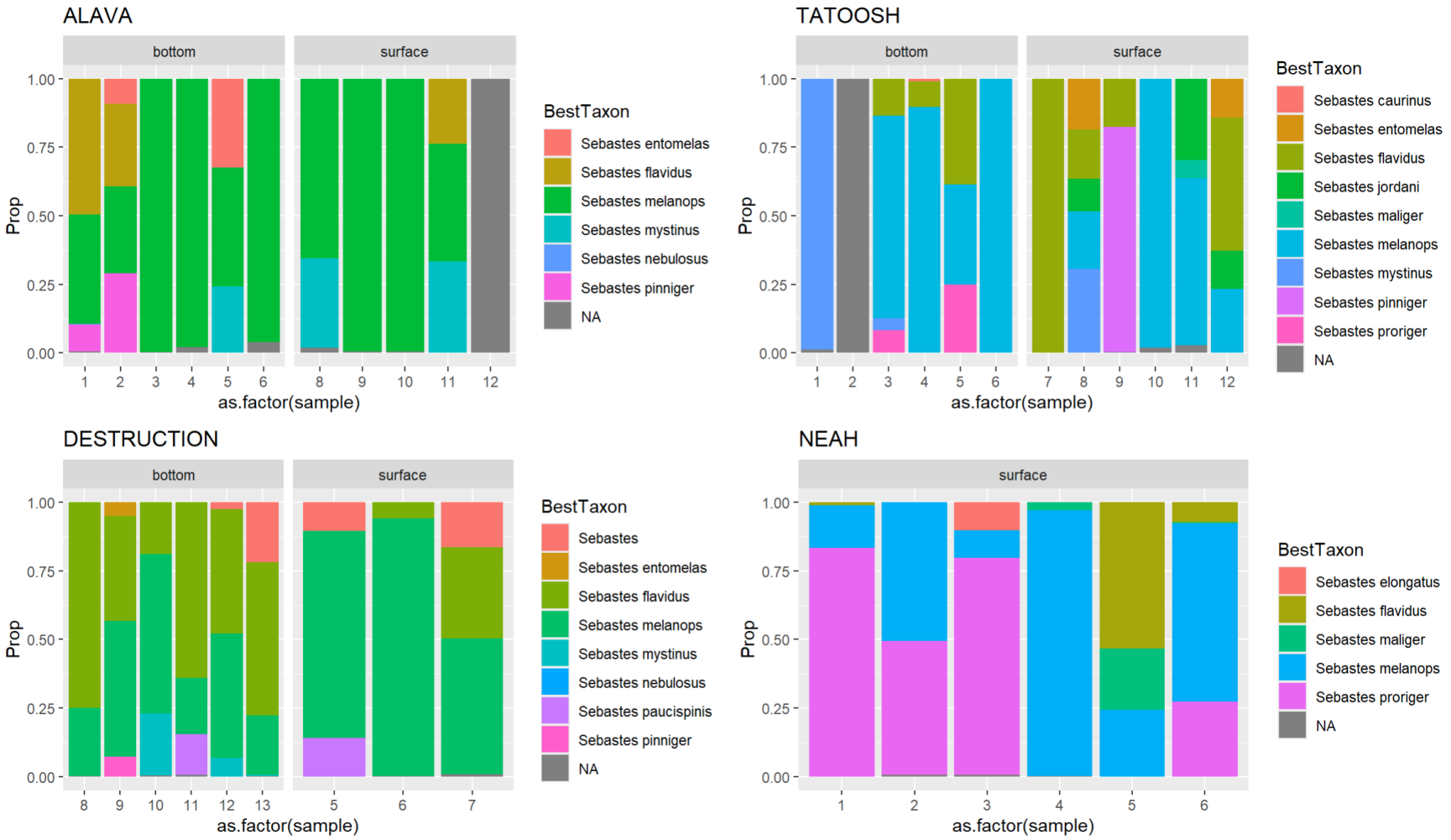
- 2019: 211 fish taxa*, and 21 marine mammal species detected

Example metabarcoding data.

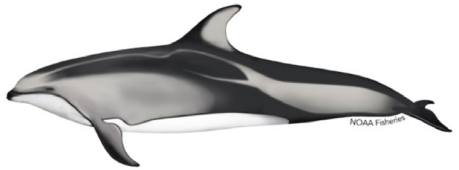


*Sebastes is (mostly) not separable with standard fish markers
We now have a working marker for separating Sebastes

Example metabarcoding data, Olympic Coast using Sebastes primer



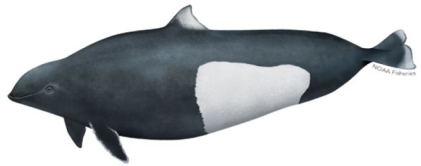
We now have a working marker for separating Sebastes



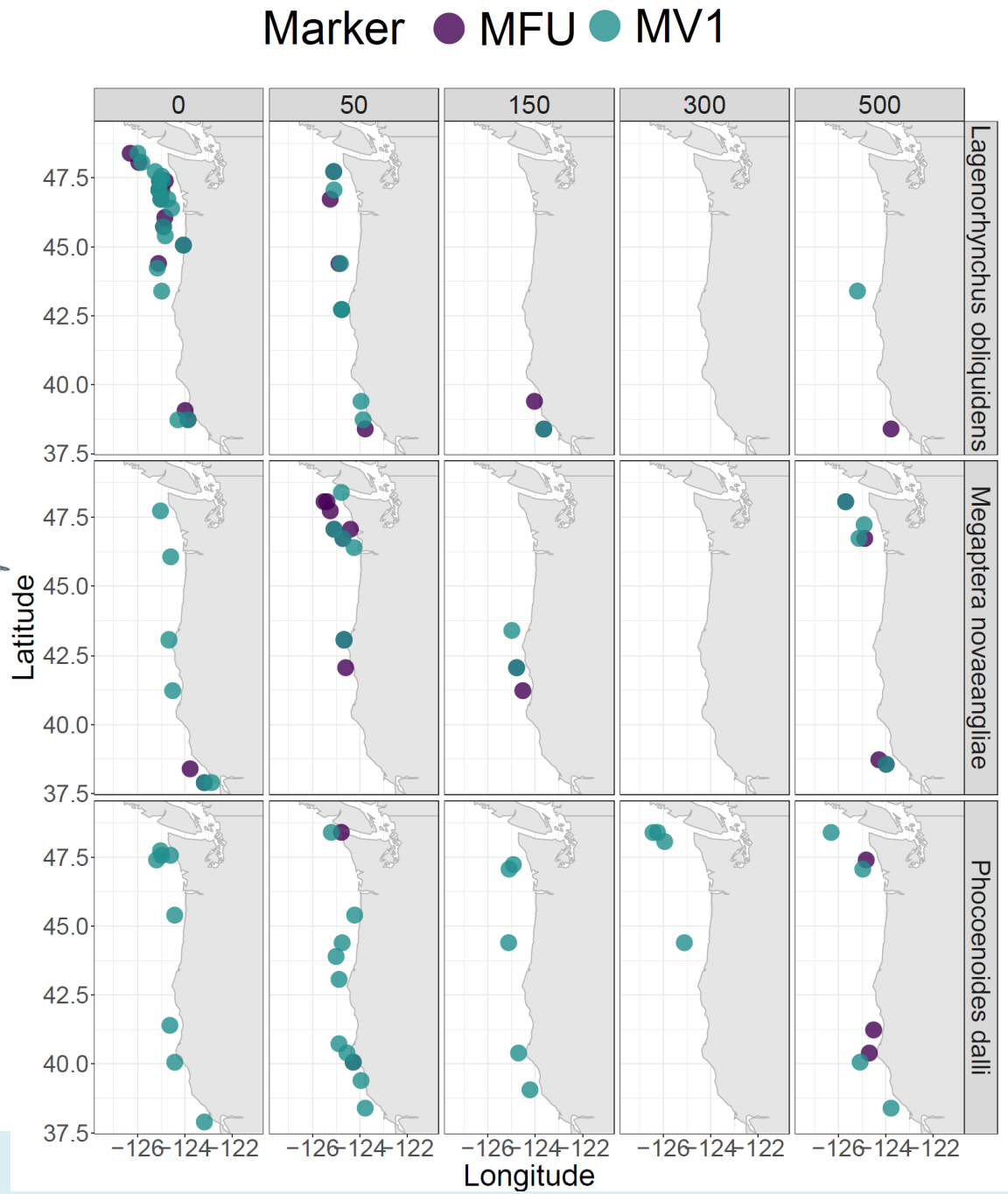
Pacific white sided dolphin
(*Lagenorhynchus obliquidens*)
51 detections



Humpback whale
(*Megaptera novaeangliae*)
33 detections



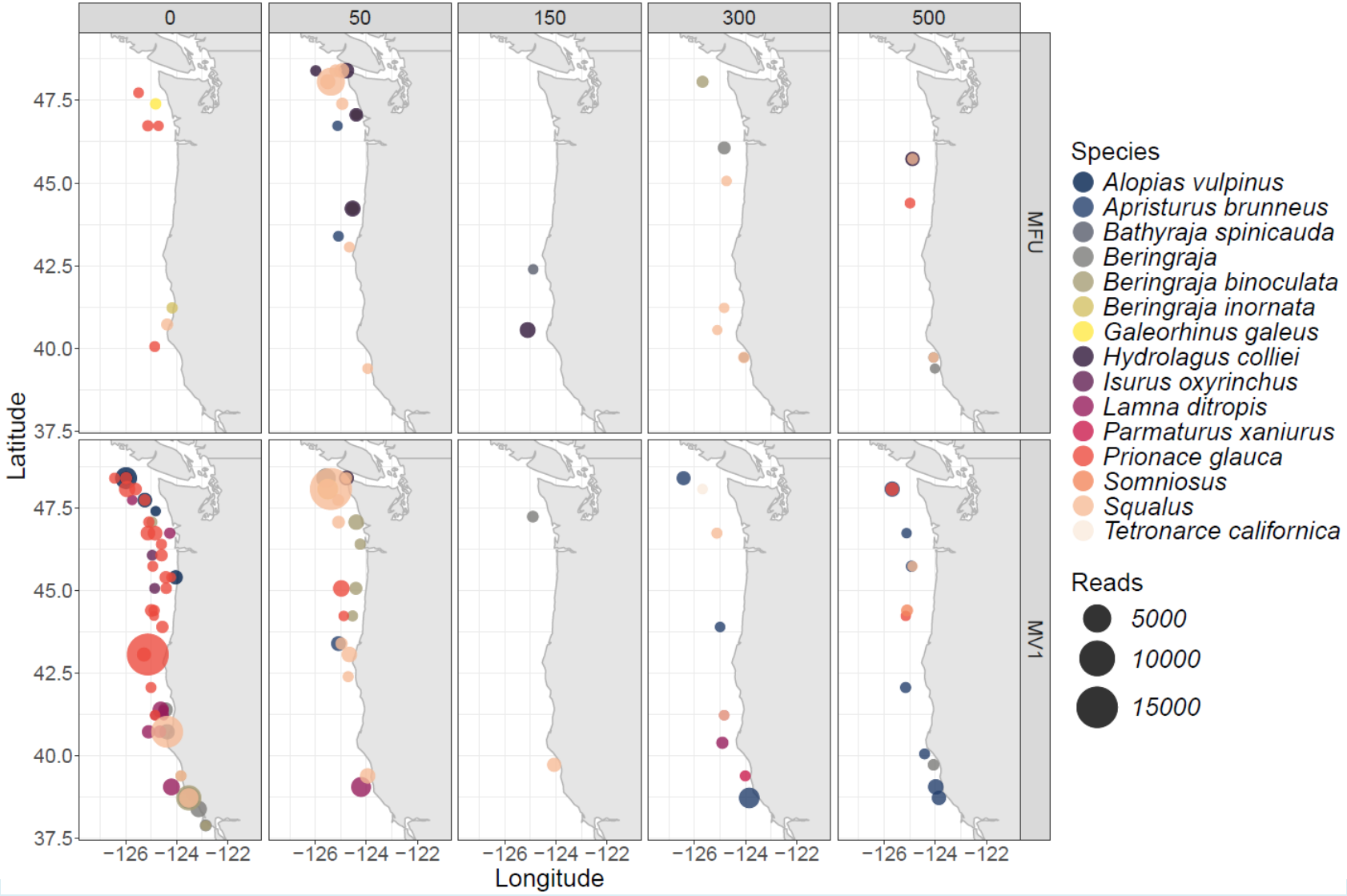
Dall's porpoise
(*Phocoenoides dalli*)
36 detections



Mismatches

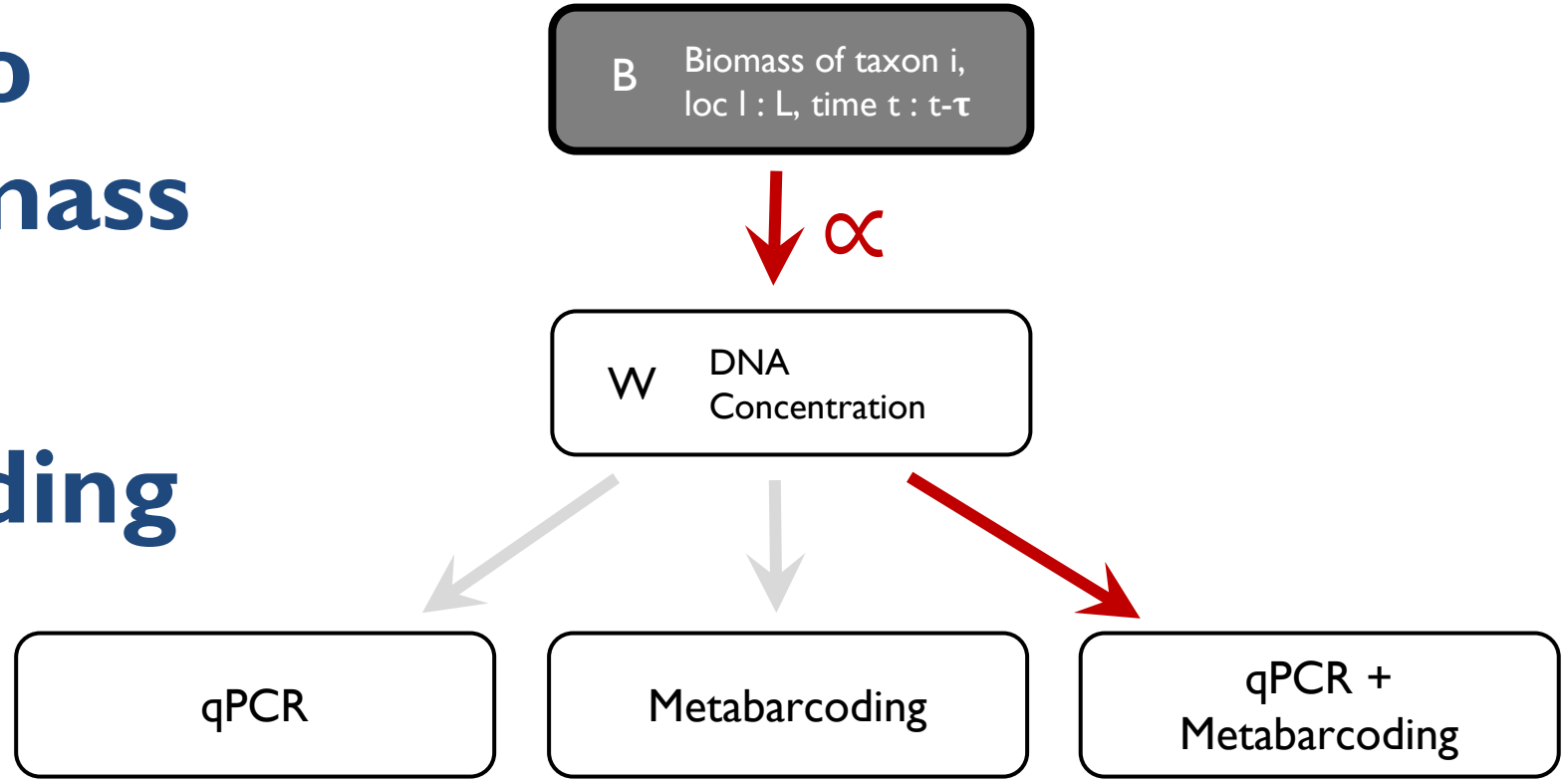
MarVer1-F	----------CGTGCCAGCCACCGCG
MiFishU-F	GCCGGTAAAACTCGTGCCAGC-------
L. obliquidens	GTTGGTAAACTTCGTGCCAGCCACCGCG
M. novaeangliae	GTTGGTAAACTTCGTGCCAGCCACCGCG
P. dalli	GTTGGTAAACTTCGTGCCAGCCACCGCG

Sharks


0
50
150
300
500
MFU
MV1
Latitude
47.5
45.0
42.5
40.0
37.5
−126 −124 −122
Longitude
Species
Alopias vulpinus
Apristurus brunneus
Bathyraja spinicauda
Beringraja
Beringraja binoculata
Beringraja inornata
Galeorhinus galeus
Hydrolagus colliei
Isurus oxyrinchus
Lamna ditropis
Parmaturus xaniurus
Prionace glauca
Somniosus
Squalus
Tetronarce californica
Reads
5000
10000
15000

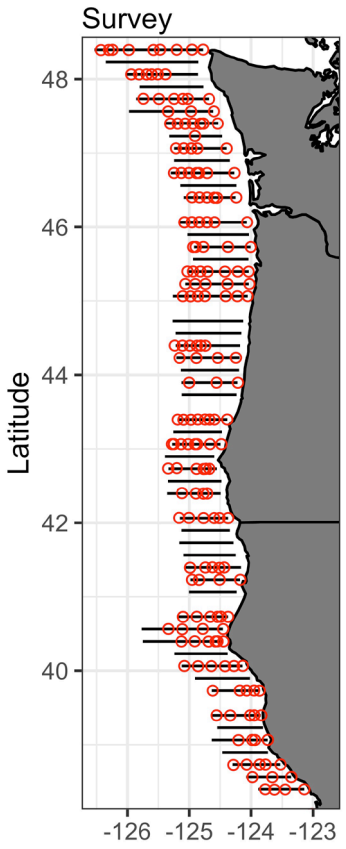
eDNA to inform biomass

qPCR + Metabarcoding

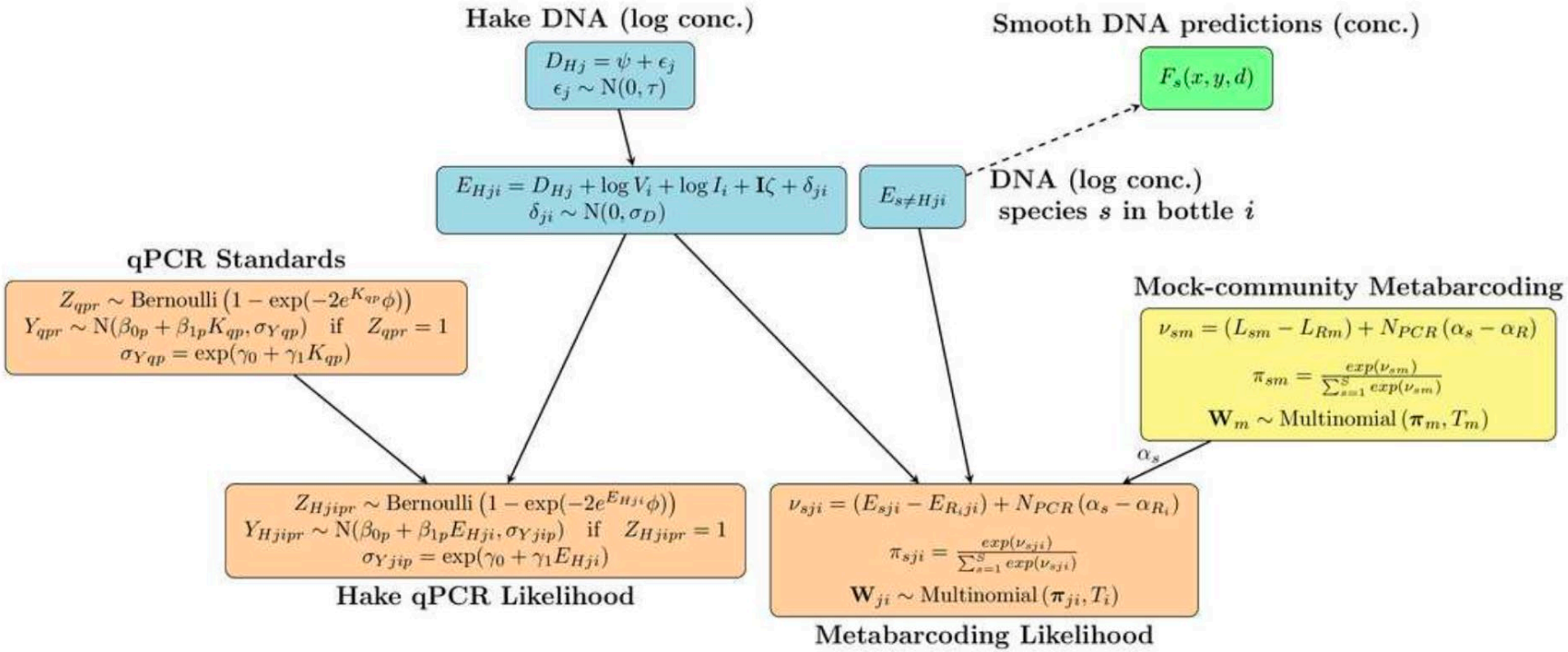


- Need adequate sampling (space, time) and well-defined target.
- qPCR directly measures concentration.
- Metabarcoding provides relative proportions.
  - Unreplicated or Replicated metabarcoding.
  - Some caveats.

qPCR + Metabarcoding



Guri et al. 2026



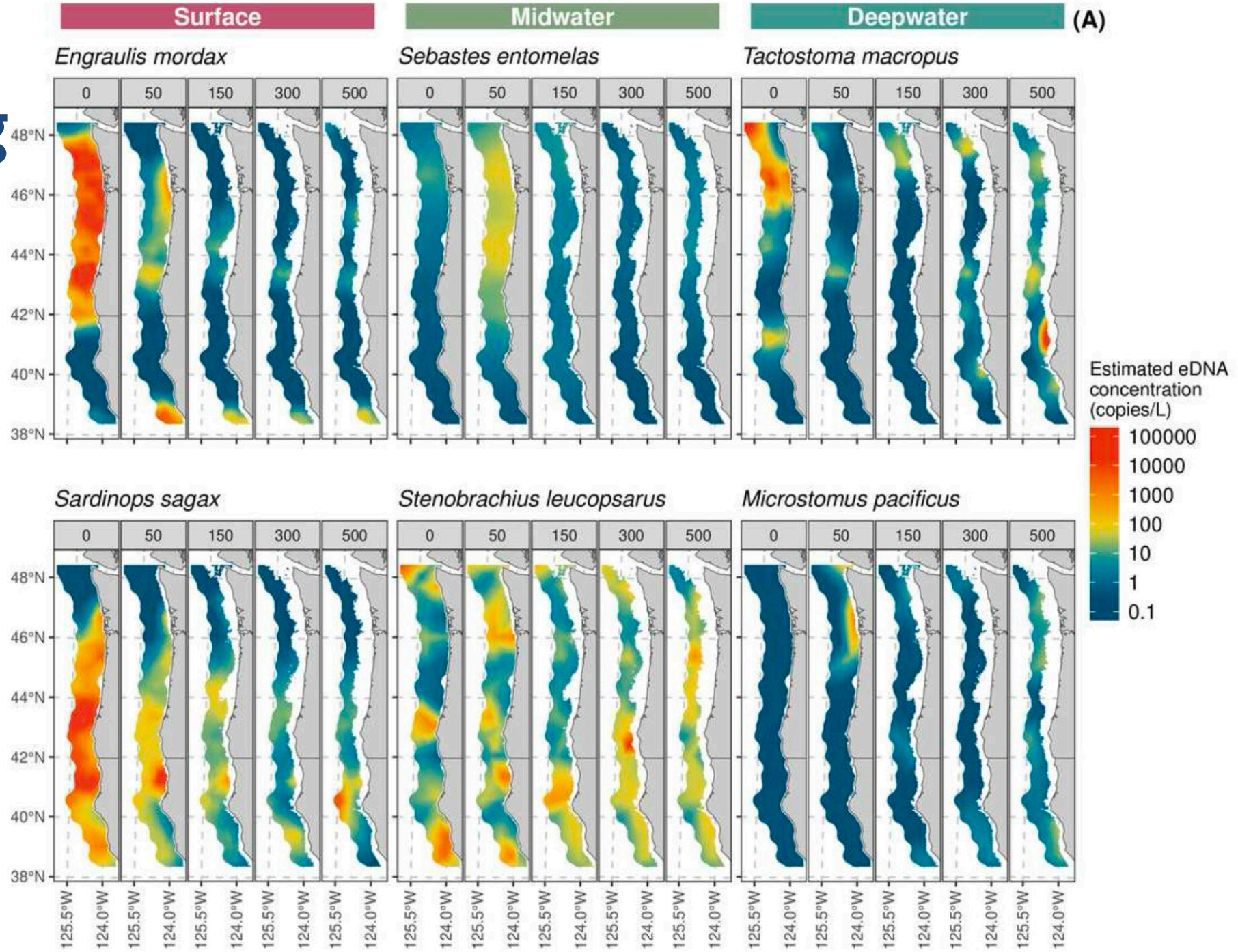
Index
- s = species (H = Hake)
- j = location (northing, easting, depth)
- x = northing
- y = easting
- d = depth
- i = biological replicate
- r = technical replicate
- p = qPCR plate
- m = mock community sample
- q = qPCR standard aliquote sample
- R = metabarcoding reference species

Parameters and derived quantities
- ψ = mean hake eDNA concentration (log(copies/μL)
- δ = biological replicate random effect
- V_i = volume filtered and aliquote dilution offset
- ϕ = qPCR amplification parameter
- σ_Y = standard deviation of qPCR Ct values
- β_{0p}, β_{1p} = intercept and slope defining mean qPCR - DNA concentration relationship
- γ_0, γ_1 = intercept and slope defining standard deviation of qPCR - DNA concentration relationship
- α_s = amplification efficiency relative to reference species
- π = predicted sequence proportions at the end of PCR
- ζ = ethanol wash effect
- τ = standard deviation among biological replicates

Observations
- Z = qPCR amplification (yes=1; no=0)
- Y = qPCR Ct values
- K = known DNA conc. (log(copies/μL))
- L = known DNA conc. (log(copies/μL))
- W = metabarcoding sequencing reads
- T = total number sequencing reads
- V = volume filtered and aliquote dilution offset
- I = aliquote dilution factor

States
- D = DNA conc. at a station-depth (log(copies/μL))
- E = DNA conc. in a biological replicate (log(copies/μL))
- F = Smoothed DNA conc. (copies/L)

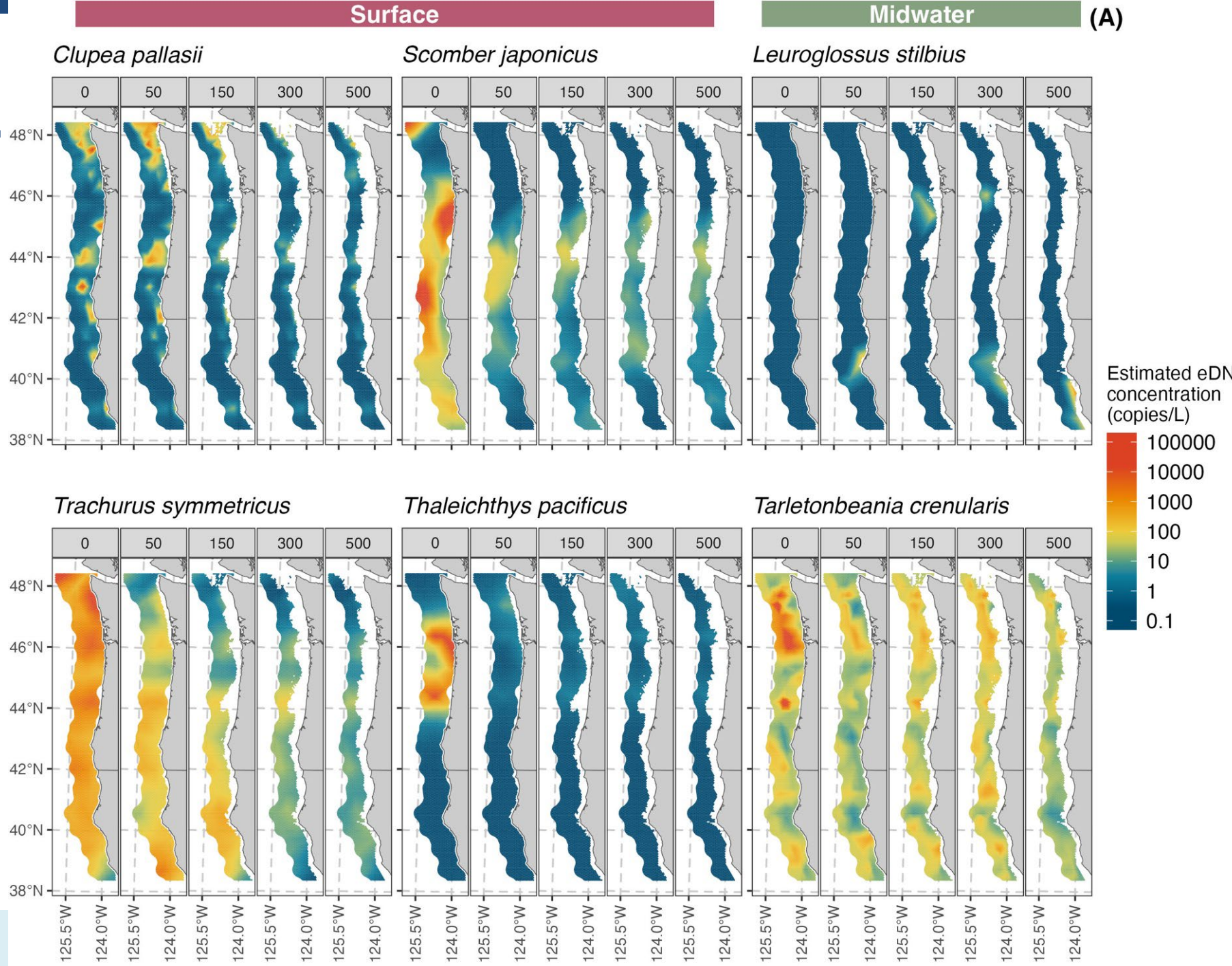
qPCR + Metabarcoding



Guri et al. 2026

NOAA FISHERIES

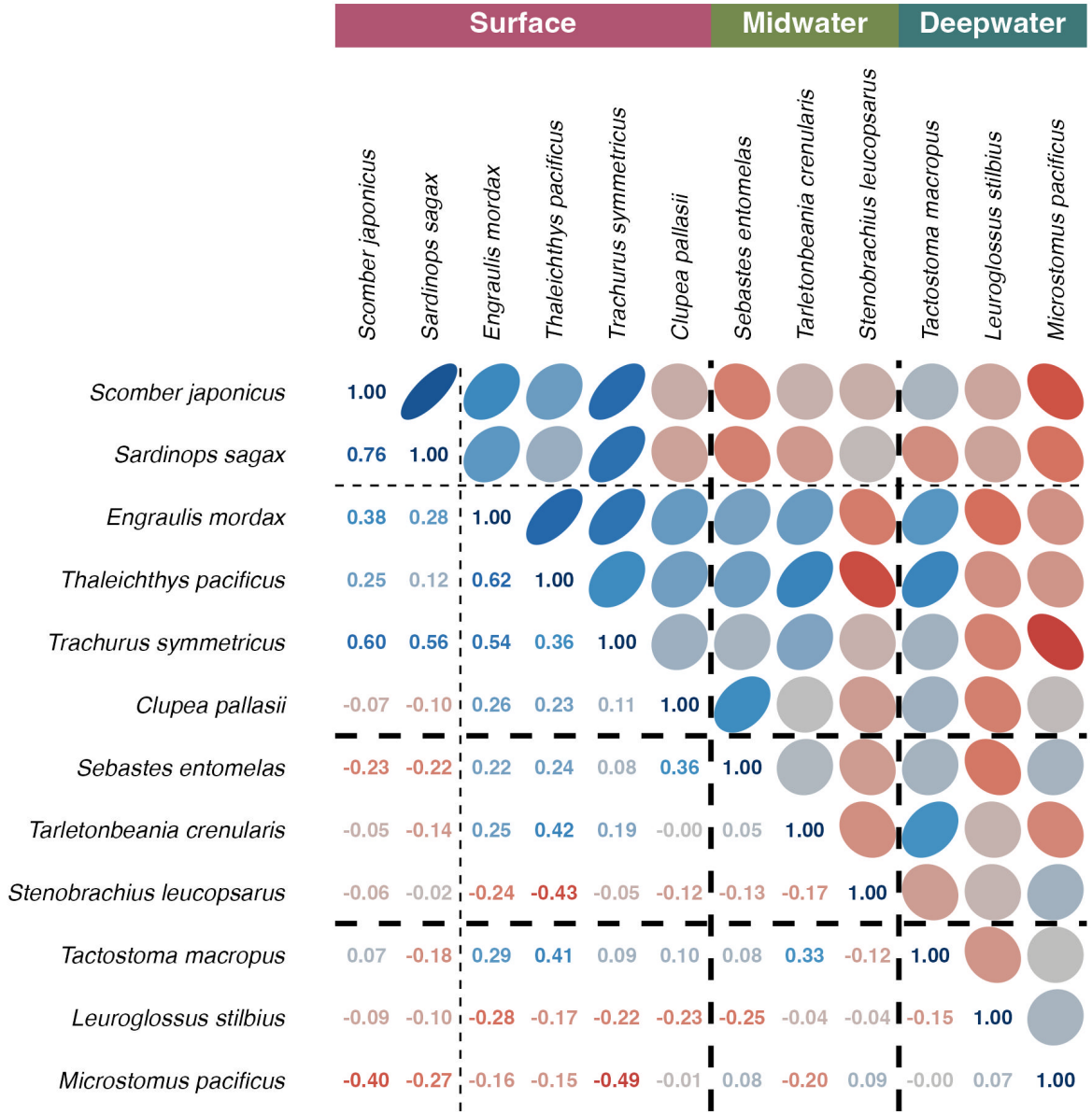
qPCR + Metabarcoding

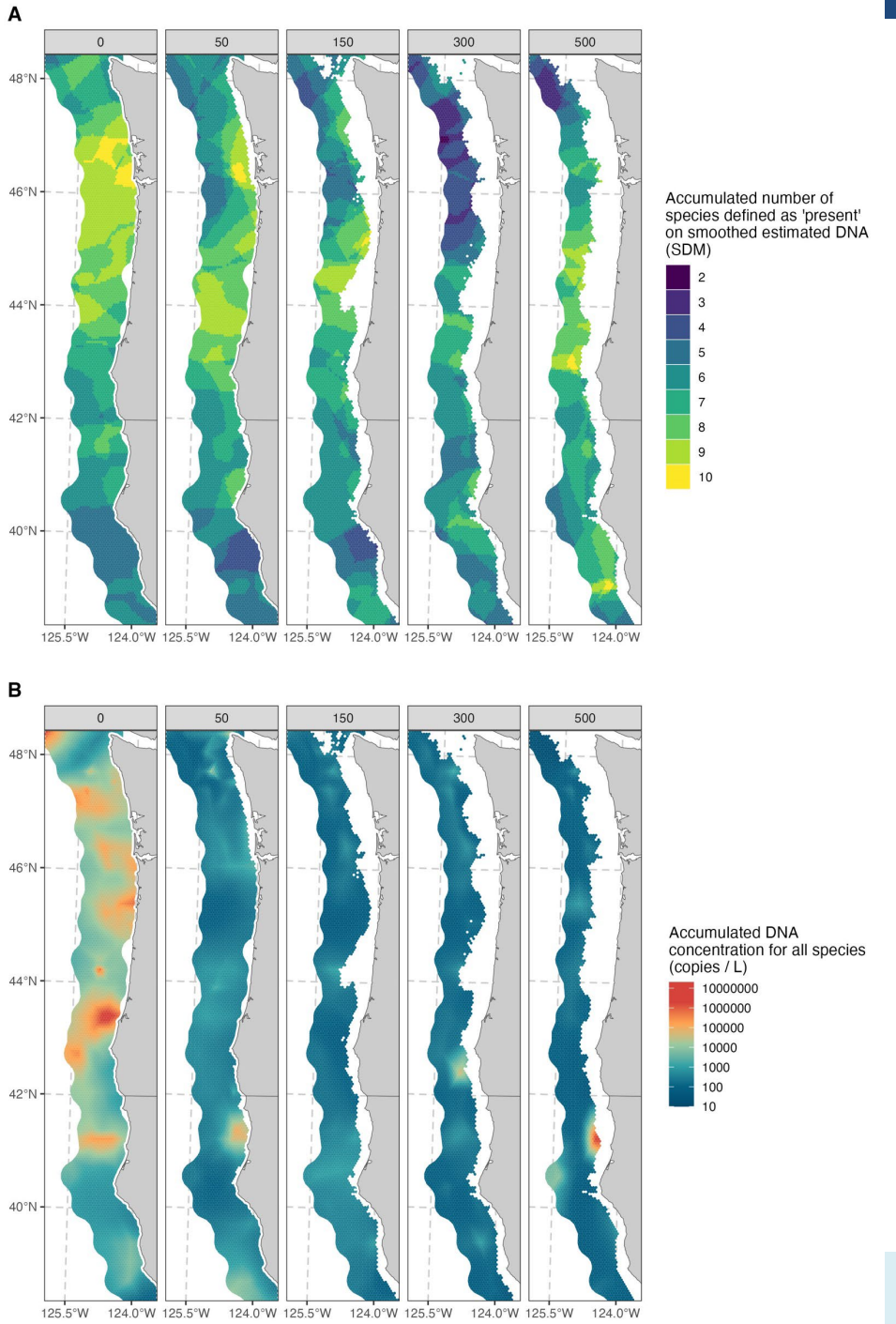


Guri et al. 2026

NOAA FISHERIES

qPCR + Metabarcoding

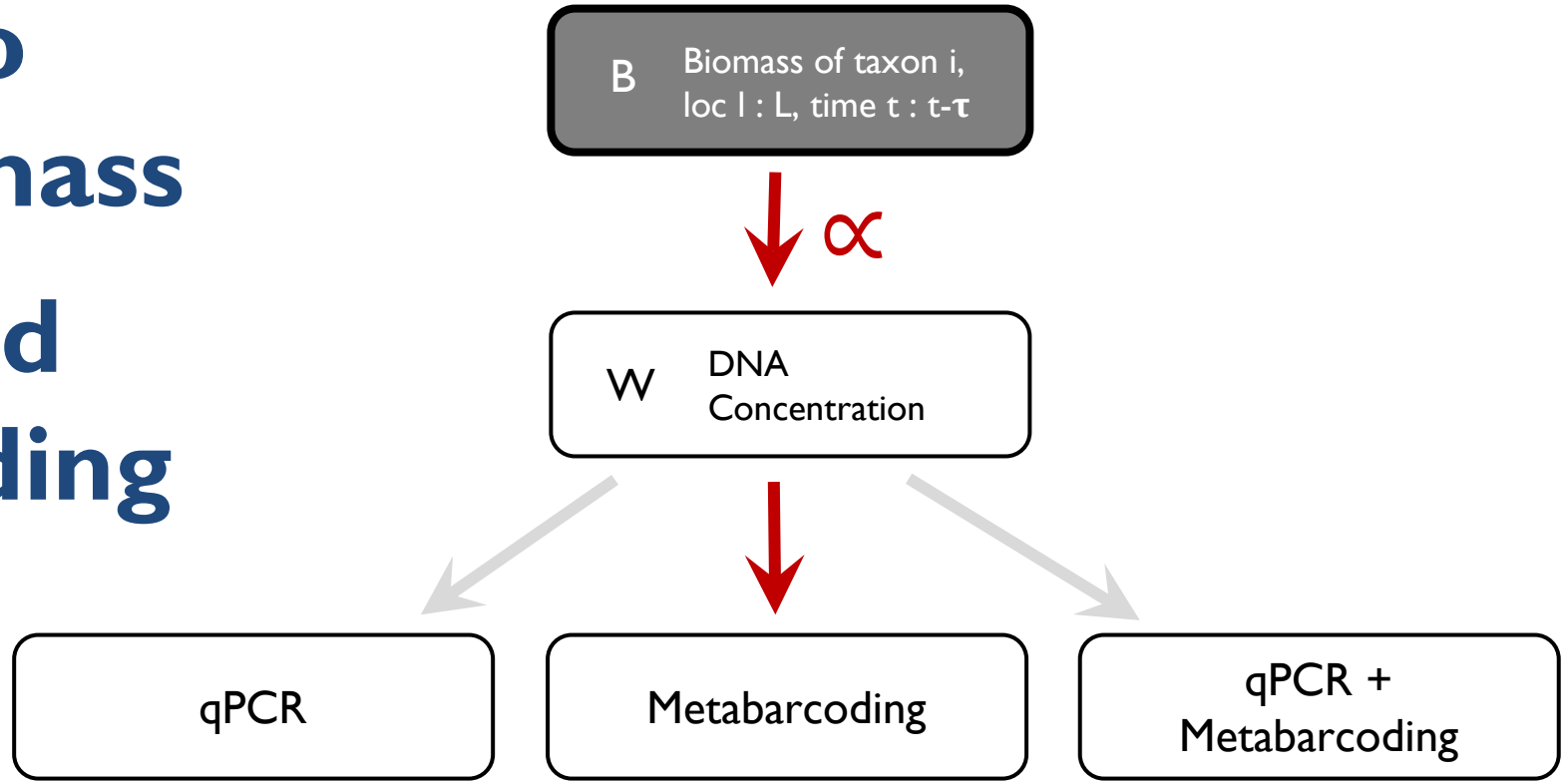




eDNA to inform biomass

Replicated Metabarcoding

(multi-species)



- Need adequate sampling (space, time) and well-defined target.
- Metabarcoding provides absolute concentration.
  - requires replicated metabarcoding for individual samples

Replicated Metabarcoding

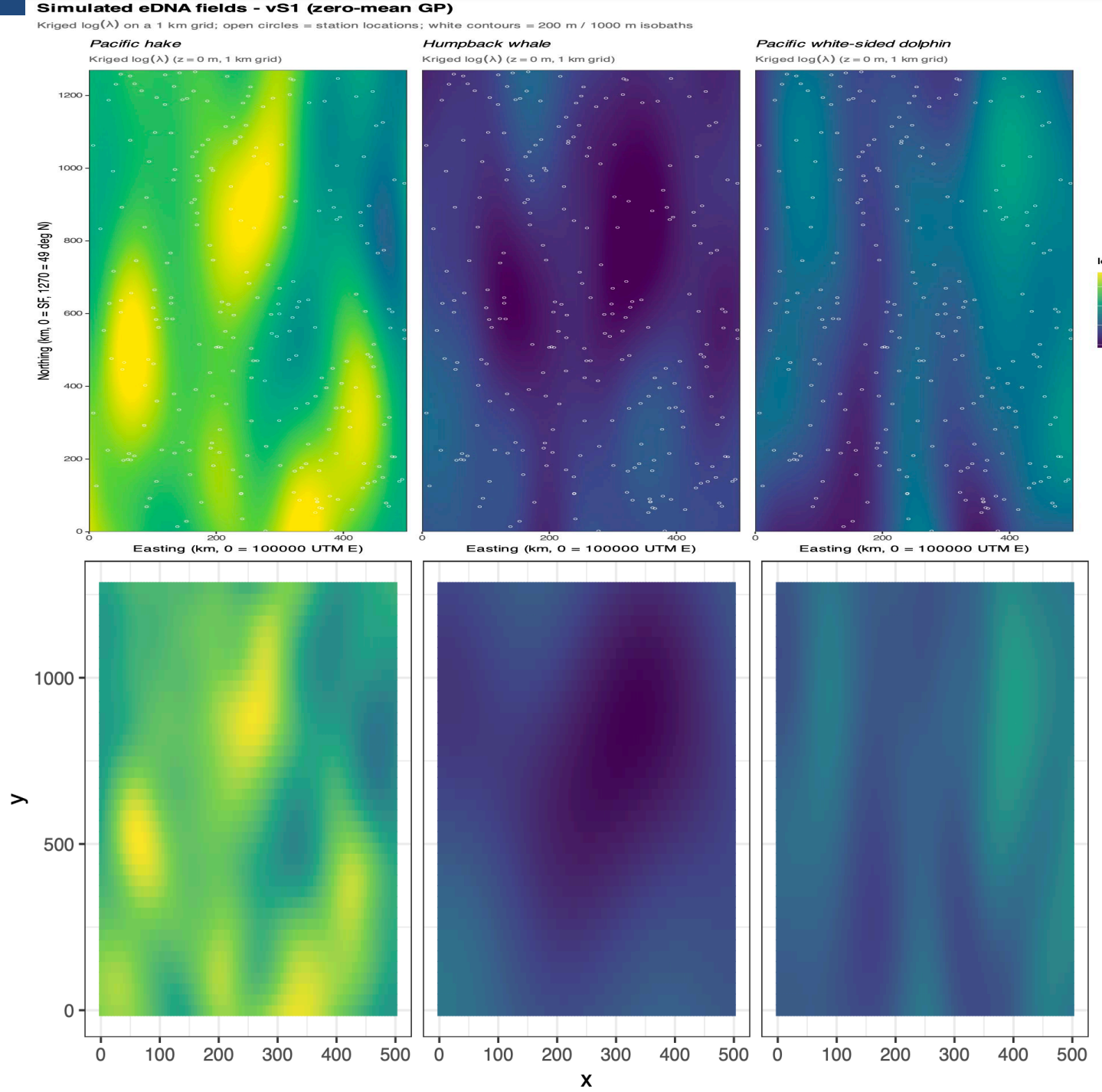
Mean-Variance relationship among metabarcoding replicates informs DNA concentration

Shelton et al. In Press

Replicated Metabarcoding

True vs. Predicted
for mock communities.

Shelton et al. In Press



Simulated

Estimated

Building Generalized Estimation Tools

Conclusions

- All kinds of eDNA observations can lead to concentration.
- Proportionality between DNA and biomass is supported
  - appropriate scale of sampling may vary among taxa
  - units of a DNA index are not in biomass (comparable to a CPUE index)
- None of it matters if you haven't done necessary sampling design work to get enough samples in the right places.
  - a great survey for one species may be awful for another.

Potential Uses in the PFMC process

- Building abundance indices for focal species
  - Particularly for poorly surveyed / unsurveyed stocks
- Ecosystem Component Species abundance and distribution
- Prey and predator abundances to inform risk tables
- Life-stage specific Indices?
  - Use pelagic samples to measure early life-stages of benthic fish?

Additional Paths for incorporating eDNA observations

Joining eDNA with traditional surveys.

- Linking species composition from eDNA and acoustic backscatter (NWFSC)
- Visual surveys and eDNA to measure cryptic taxa (PIFSC)
- Crab Trap and eDNA (UW & WDFW)
- Drop Cameras and eDNA