

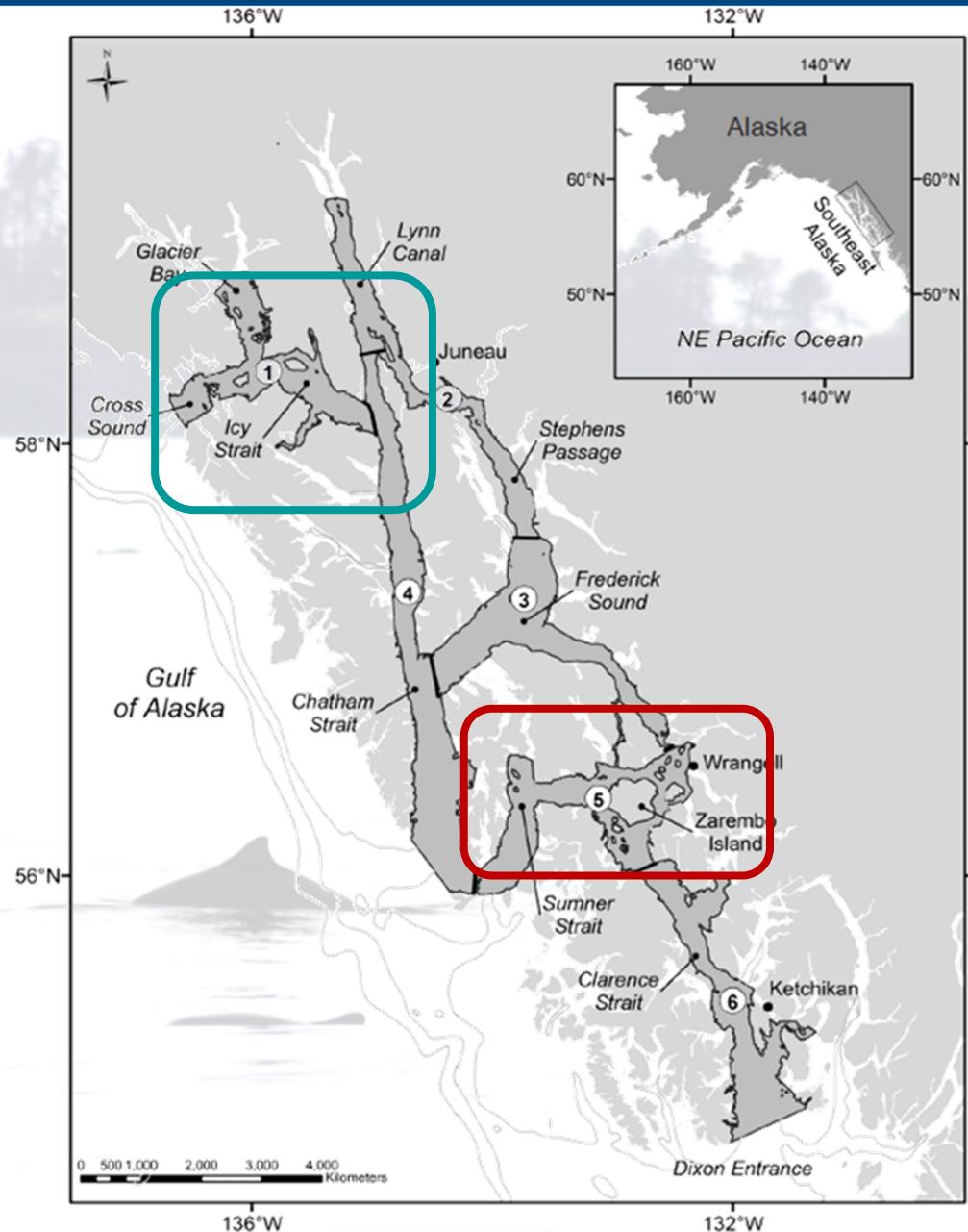
# Using eDNA to address data gaps in genetic stock structure of harbor porpoise.

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# Multiple Stocks?

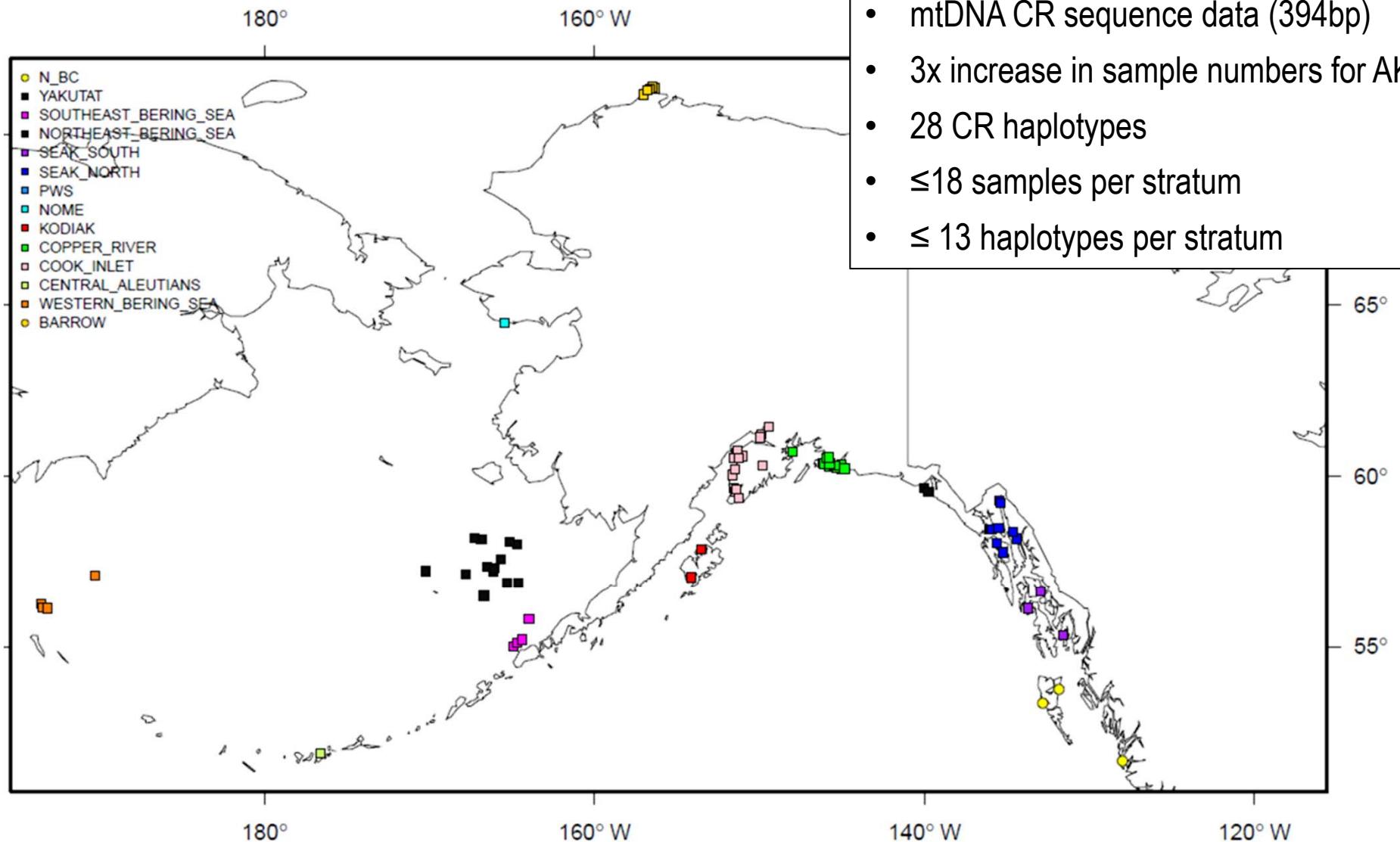
- Hiatus in distribution
- Temporal trends in abundance
- Contemporary stock structure?



# Harbor porpoise stock structure -

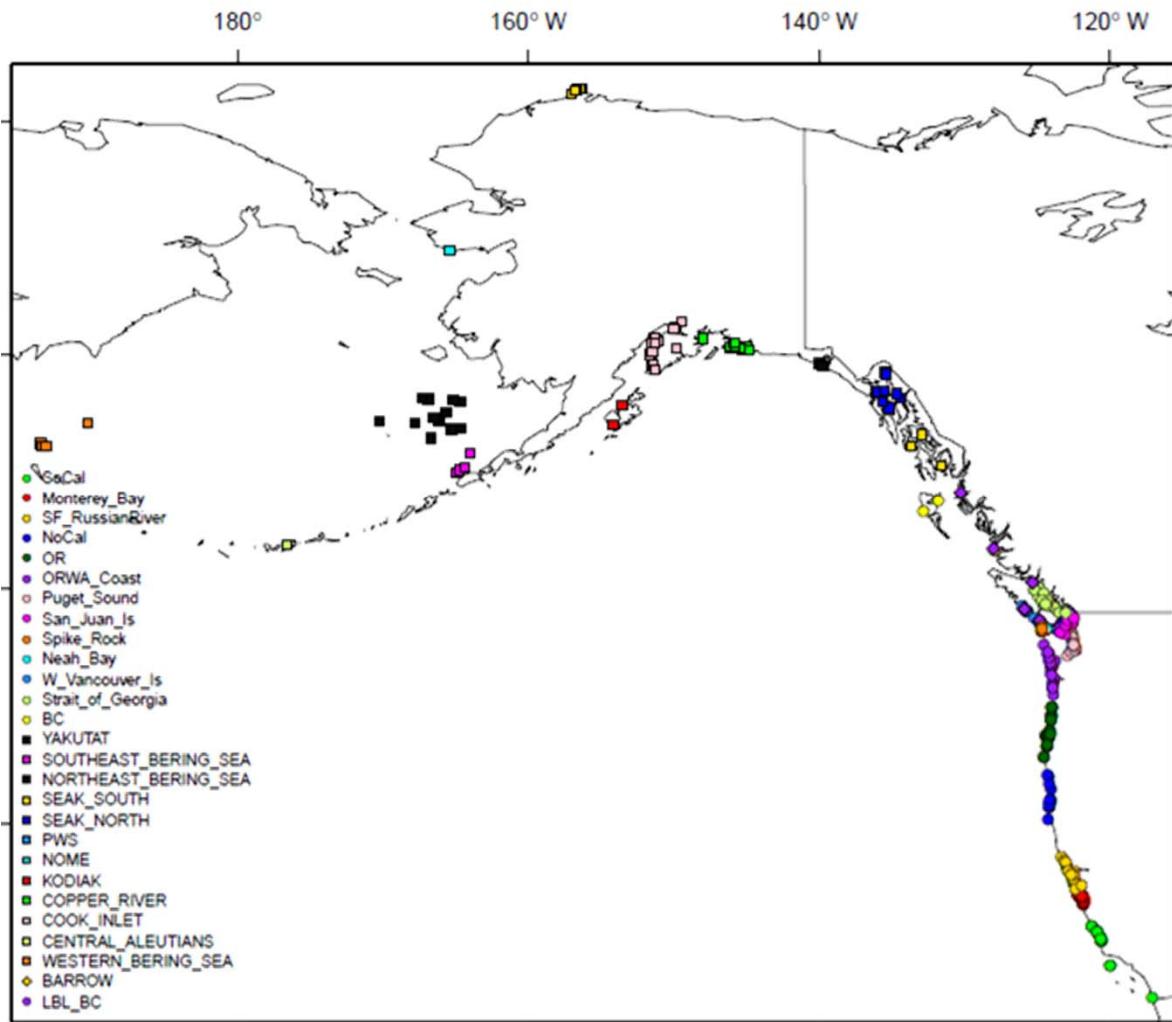
- Multiple management units in Northwest Atlantic porpoise (Rosel et al 1999, Tolley et al 2001)
- Evidence of fine scale structure in Baltic Sea (Lah et al. 2016), Black Sea (Tonay et al. 2016) and North Sea (Walton 1997, Andersen 2001)
- Broad scale population structure in NE Pacific (Rosel et al. 1995)
- Management units drawn too broadly & likely contain multiple populations (Chivers et al. 2002)
- Porpoise in B.C. comprise a single stock (Crossman et al. 2014)

# Alaska harbor porpoise

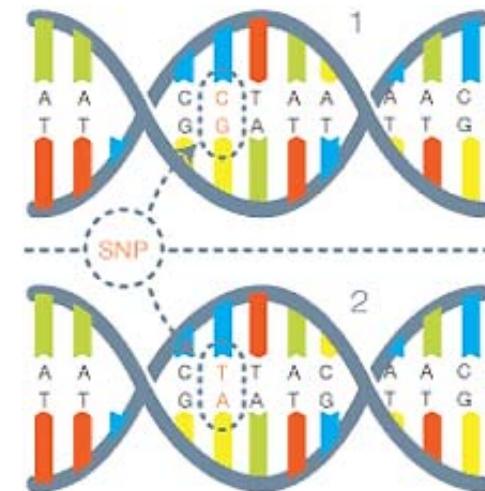


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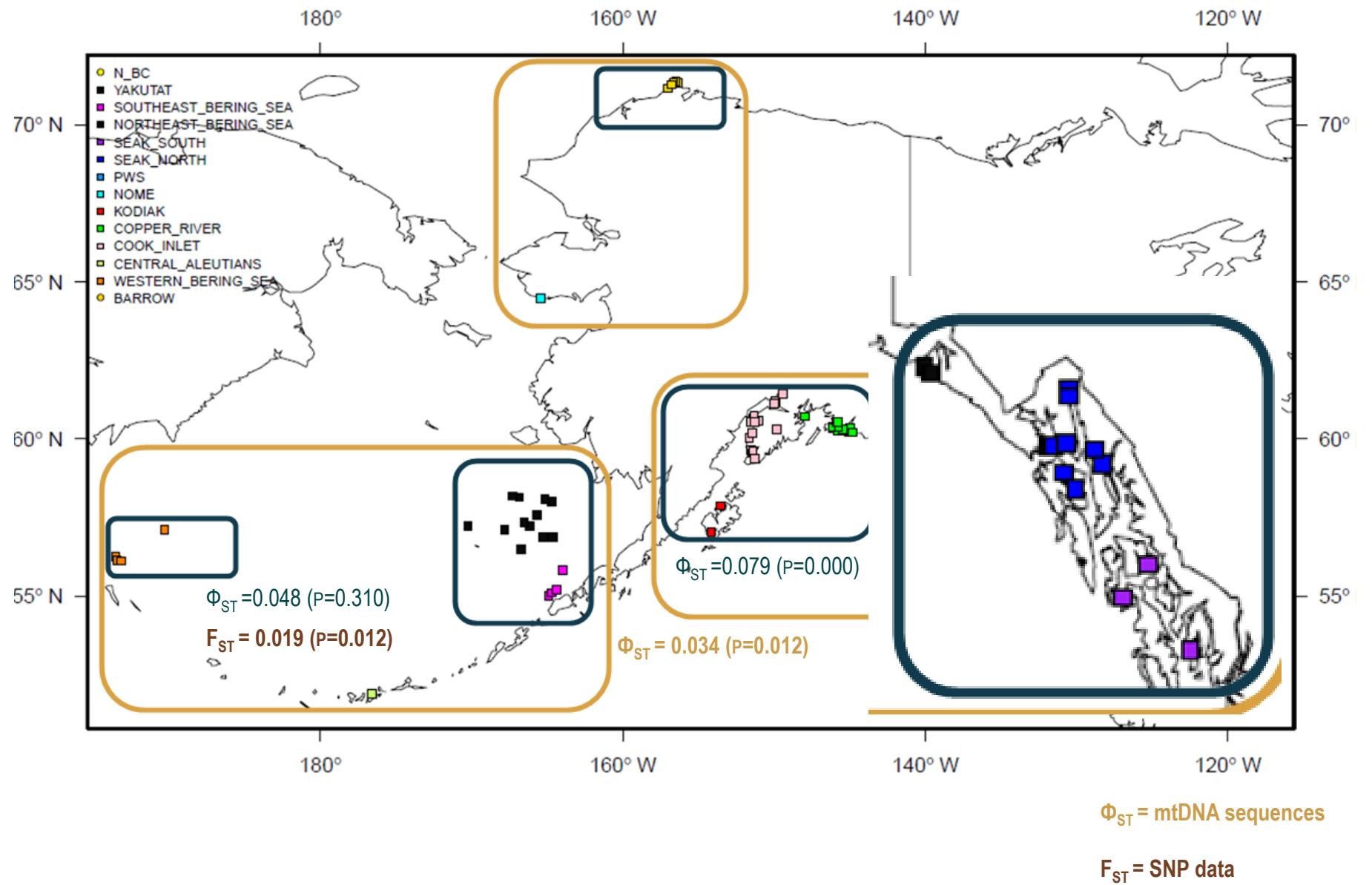
# Eastern North Pacific harbor porpoise



- 370 porpoise samples
- 308 SNP loci (GTseq)
- Samples/region 1→37
- S California → Barrow, AK



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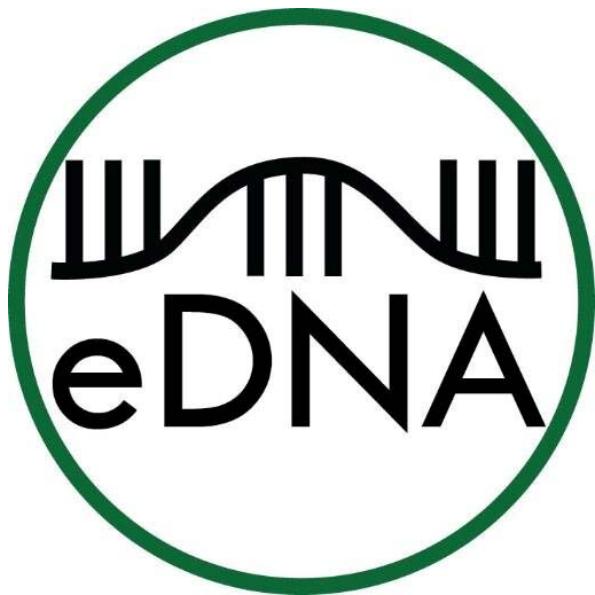
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# Remote tissue biopsy



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# Environmental DNA



- DNA that is shed by an organism into its environment.
- Sources include feces, mucous, gametes, skin, hair and carcasses.
- Applications include habitat use, biodiversity surveys, detection of rare or invasive species, species density etc.

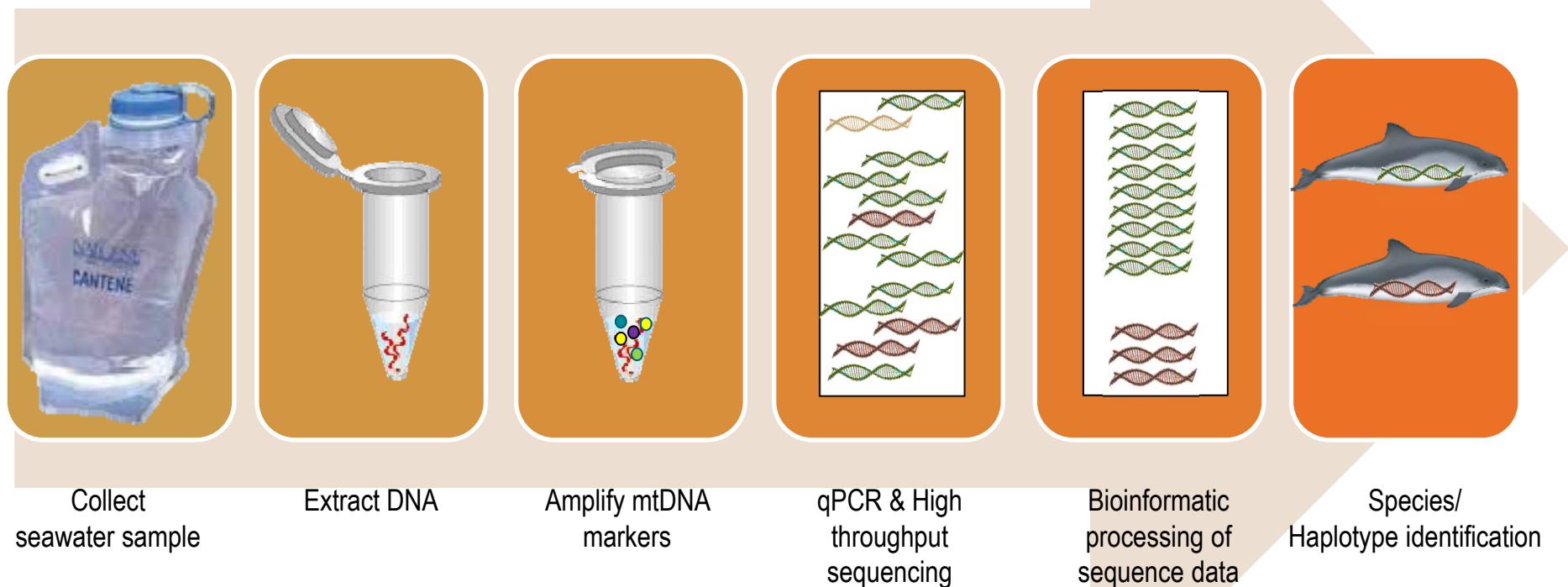


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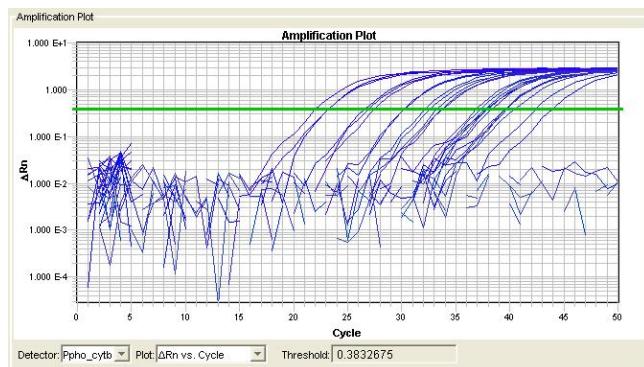
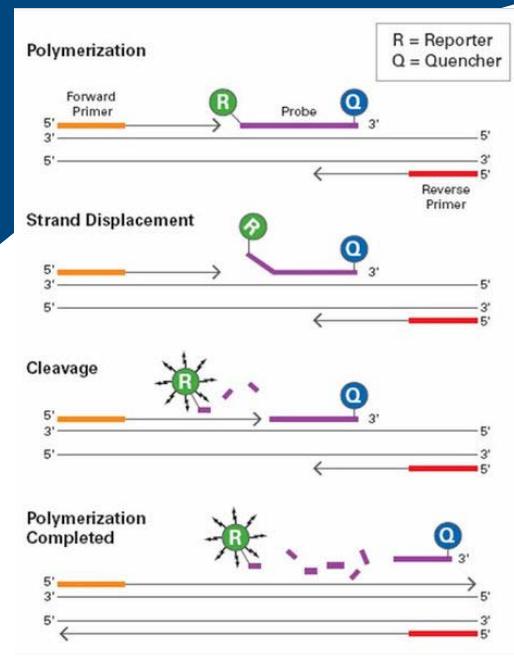
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# Species-specific sequences from eDNA samples



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# Species-specific qPCR



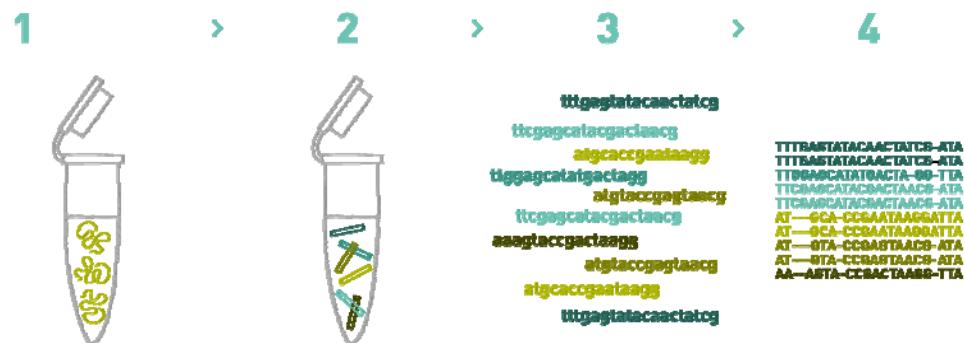
- Species-specific Taqman probes
- Quantification of target DNA
- Identification of inhibitors
- 118 eDNA samples (10 field negatives)
- Amplification failure, n = 11
- Mean concentration 0.0046 ng/ $\mu$ l (SD=0.014)

A harbor porpoise is shown breaching the surface of the ocean. The dolphin's dark grey body is partially submerged, creating a splash as it moves through the water. The background consists of dark, textured ocean waves.

Amplification of population-level sequence data for  
harbor porpoise using species-specific 2<sup>nd</sup> generation  
targeted amplicon sequencing

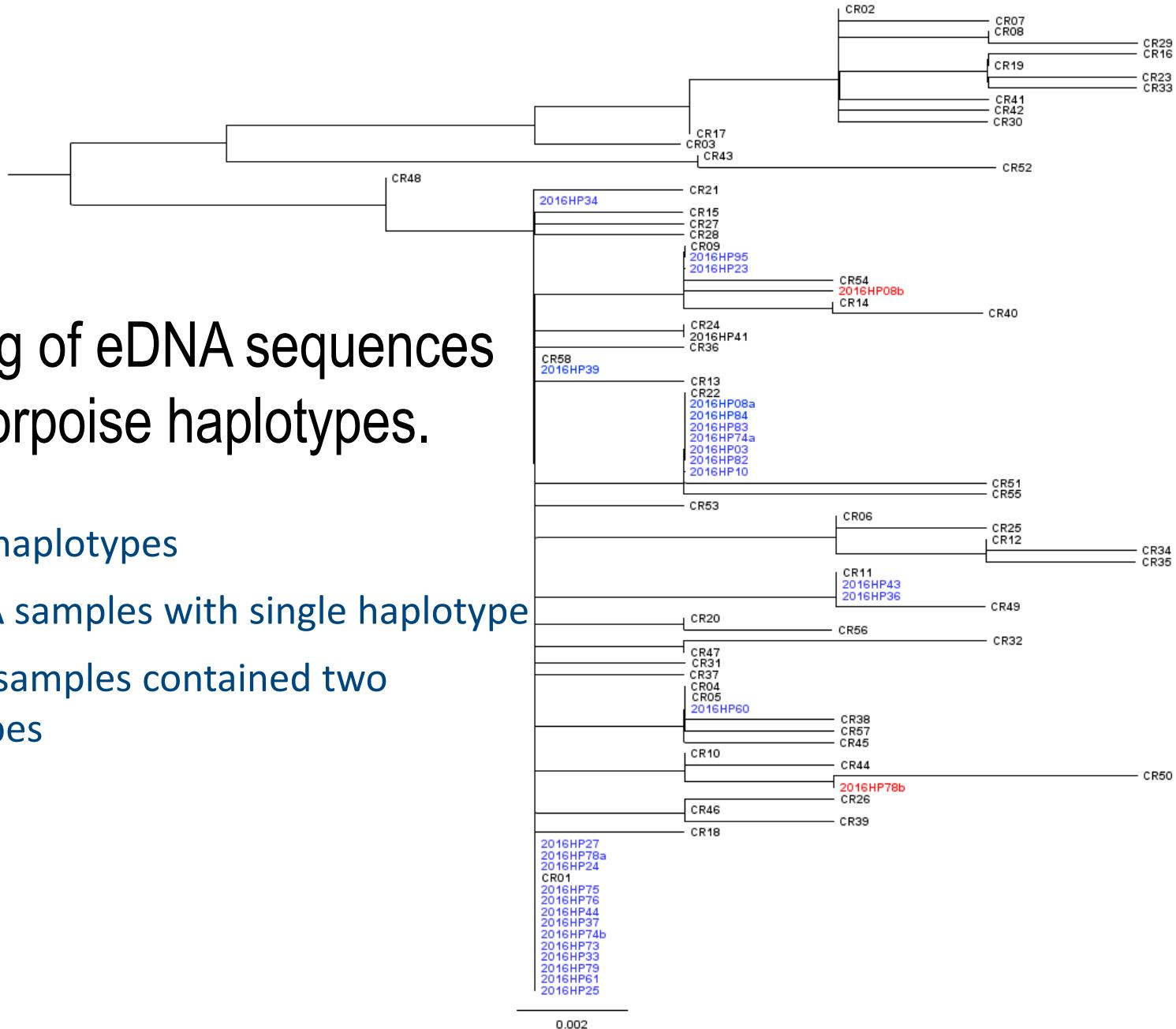
# Next generation sequencing -

- 40 seawater eDNA samples
- 34,874,482 raw paired-end reads from a pooled MiSeq library
- Read depth per sample passing filter ~72k
- 4 tissue controls to estimate threshold of “correct” sequences
- Maximum error rate for controls was 1.70% (SD = 0.29%)
- *P. phocoena* mtDNA sequences generated from 30/36 eDNA samples
- 9 unique haplotypes (379bp)
- 7 haplotypes previously identified in Alaska harbor porpoise

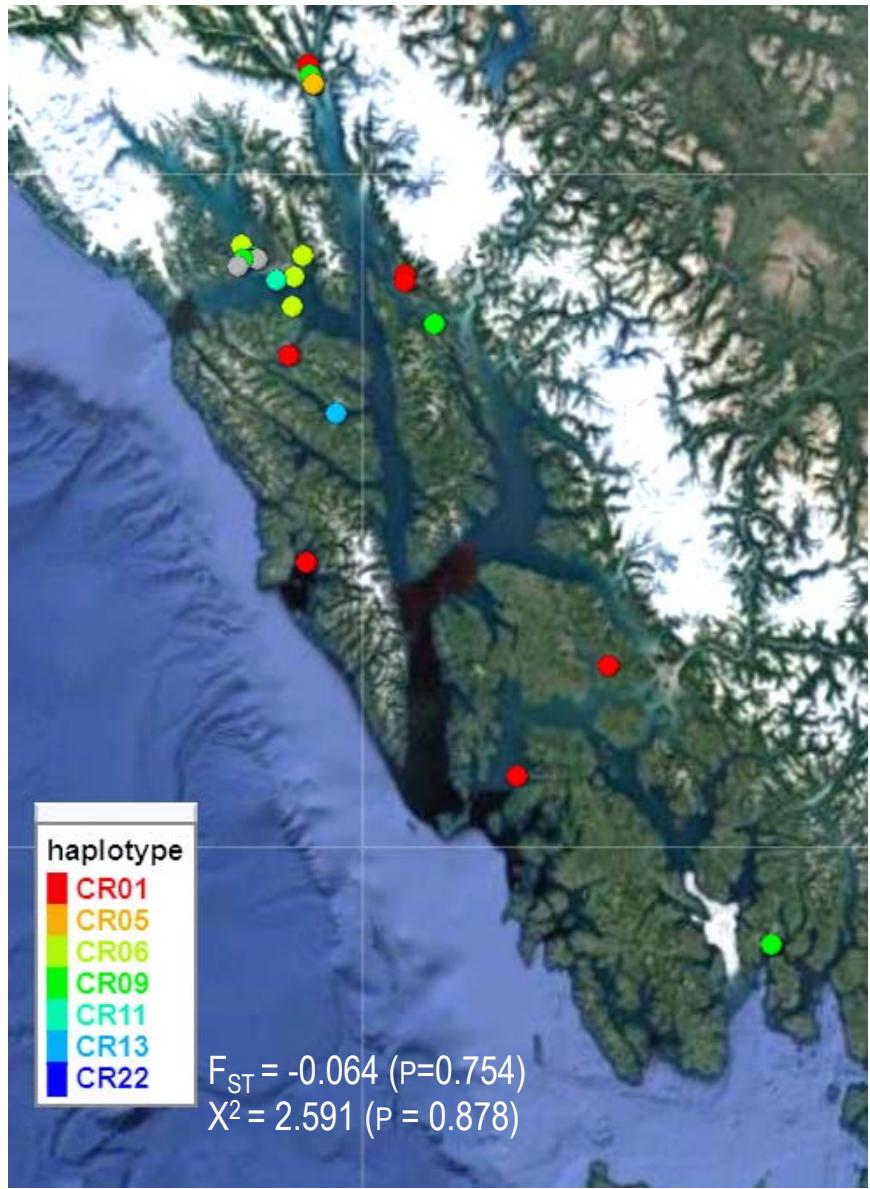


# Clustering of eDNA sequences with porpoise haplotypes.

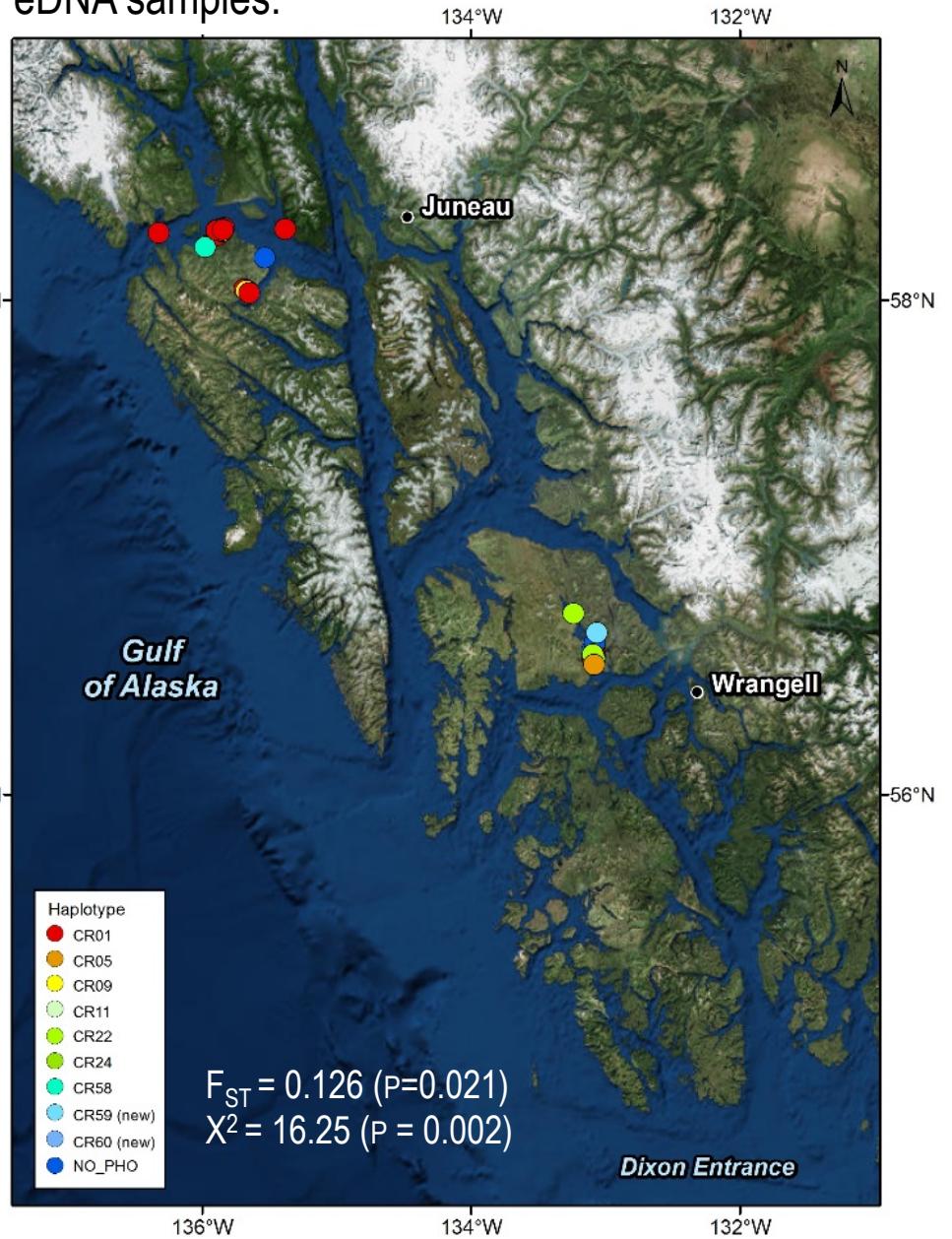
- 2 ‘new’ haplotypes
  - 27 eDNA samples with single haplotype
  - 3 eDNA samples contained two haplotypes



Strandings/bycatch:



eDNA samples:



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## Summary -

- Multi-pronged approach to characterizing contemporary population structure in Alaska harbor porpoise
- Detection of harbor porpoise in seawater eDNA samples
- Ability to generate population-level sequence data from eDNA
- Alternative method of sample collection
- Novel approach to examining contemporary stock structure
- Ability to dovetail eDNA sampling with abundance survey

# 2018 Opportunistic eDNA Sampling

- Glacier Bay NP
- Juneau
- Tenakee Inlet, Chichagof Is
- Kelp Bay, Baranof Is
- Petersburg
- Wrangell
- Craig, Prince of Wales Is
- Ketchikan





## Acknowledgements

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- Phil Morin & Kelly Robertson, NOAA, SWFSC
  - Captain & crew m/v Lady Gudny
  - Captain & crew m/v Dreamcatcher
  - Auke Bay NOAA Lab