CO1 haplotype diversity of the invasive colonial tunicate *Didemnum vexillum* in Drakes Estero and Bodega Bay
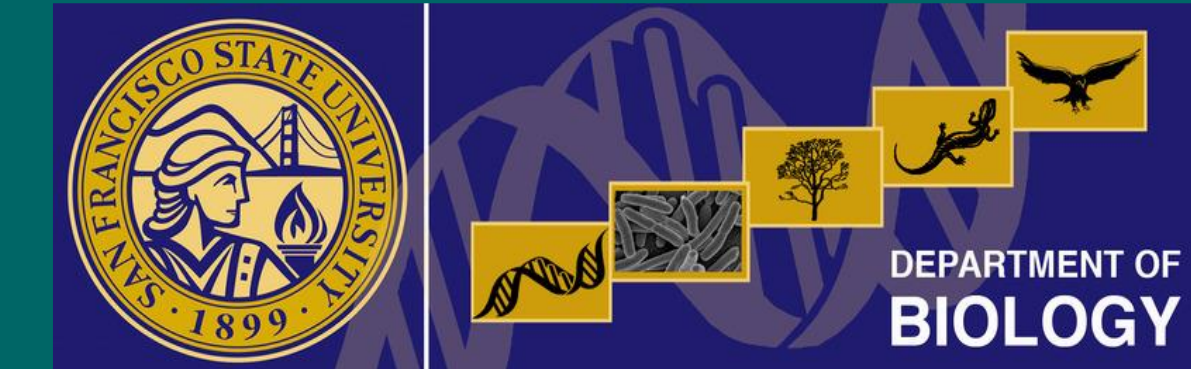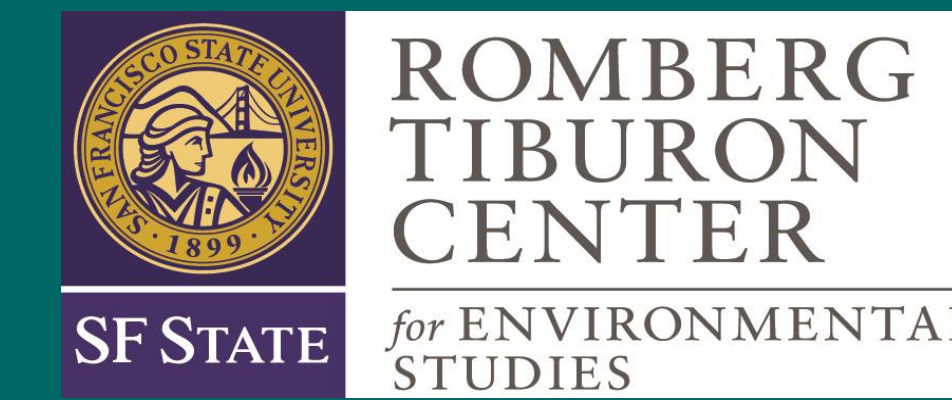
Cecilia Hernandez[1,2], Rachel Weinberg[1,2], C. Sarah Cohen[2]
Romberg Tiburon Center[1], San Francisco State University[2]

Background

The invasive colonial ascidian *Didemnum vexillum* is highly known for its ability to spread and overgrow native organisms, as well as cover artificial substrates. This marine pest is of great ecological and economic concern and poses a serious threat. Taking the population of Georges Bank for example, *D.vexillum* colonies cover 50–90% of a 230 km^2 area[1]. *D. vexillum* is capable of re-establishment when a portion of the colony is torn away. Shipping and recreational vessels are vectors of dispersal in that water currents or the vessels themselves allow for *D.vexillum's* introductions to foreign areas.

Figure 1 (right): Example of biofouling with *D. vexillum* having partial and complete coverage on the mussels

http://wdfw.wa.gov/ais/didemnum_vexillum/

https://woodshole.er.usgs.gov/project-pages/stellwagen/didemnum/images/jpg/V8_dav1_16650070.jpg

Figure 2 (above): Impact of *D.vexillum* on aquaculture

Figure 3 (above): Large colonies are nearly solid mats in Georges Bank, covering important fishing grounds in the northeast Atlantic

Methods

Samples:
- Samples collected from Bodega Bay (Spud Point and Porto Bodega) and Drakes Estero Bay by C. Sarah Cohen and Rachel Weinberg
- Taken from various artificial substrates, docks and old oyster racks.

COI Amplification and Sequencing:
- Used PCR to amplify the 586 nucleotides of the mitochondrial locus CO1
- Amplified DNA was purified using an ExoSAP–IT® for PCR product cleanup
- Sanger Sequenced on a ABI 3130 at RTC/ SFSU

Assessing Haplotypes:
- Raw sequences trimmed and aligned by MUSCLE in Geneious 7.1.2.[2]
- compared to GenBank haplotype sequences[3,4]

Why barcode the mitochondrial locus CO1?

- Highly conserved across species
- The rate a sequence changes over time is:
- ✓ <u>slow</u> enough so that it's likely to be identical in the same species
- ✓ <u>fast</u> enough so that it's different between species
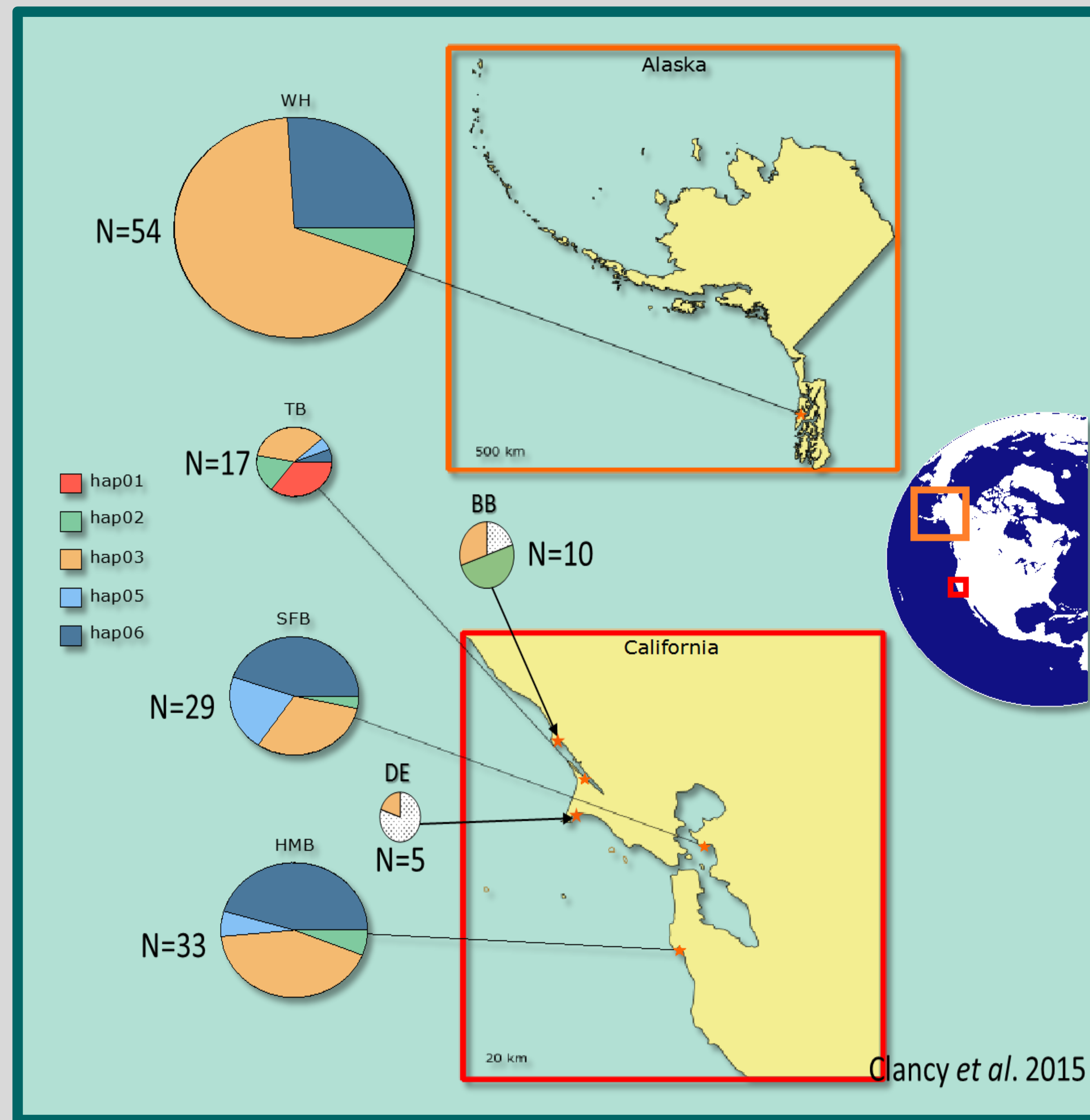- Good chance of extracting enough mitochondrial DNA for successful PCR

Figure 4 (above):
This haplotype map presents the frequency of each haplotype per population. Haplotypes are distinguished by color. Sizes of charts are relative to sample size. Adding to previous data[5] we have the population of Bodega Bay(BB) with a sample size of 10 and Drakes Estero (DE) with a sample size of 5.
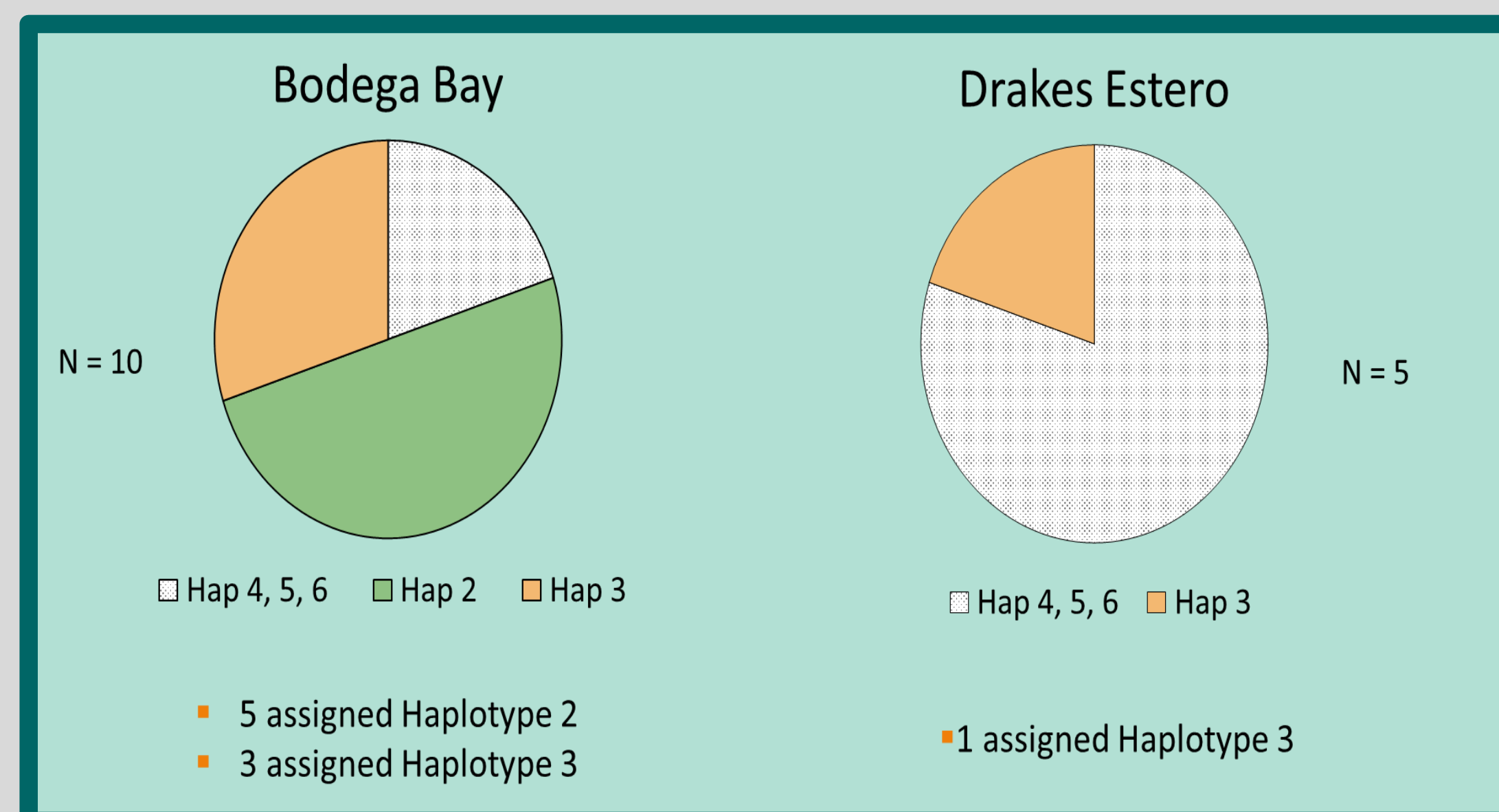

Figure 5 (left):
Pie chart representations of the haplotypes assigned per location sampled. The white areas depict the samples unassigned due to short sequence readings. Haplotypes 4, 5 and 6 are of equal possibility as the variable sites making each unique were missing for those sequences.

Bodega Bay N = 10
☐ Hap 4, 5, 6 ☐ Hap 2 ☐ Hap 3
- 5 assigned Haplotype 2
- 3 assigned Haplotype 3

Drakes Estero N = 5
☐ Hap 4, 5, 6 ☐ Hap 3
- 1 assigned Haplotype 3

Hypothesis

The three sites: Tomales Bay, Bodega Bay and Drakes Estero experience similar boat traffic, with that we expect similar levels of genetic diversity with shared haplotypes.

Results

- 2 haplotypes previously observed on the west coast
  - ✓ Tomales, San Francisco, Half Moon Bay
- Haplotype 1, unique to Tomales, was not found
  - ✓ however, our sample size is much smaller

Future Directions

- Preliminary evidence shows haplotypes present to be those shared among all other locations along the north American coast.
- Larger sample sizes and longer sequence reads (586bp) are necessary to yield a more true understanding about the genetic diversity of each location.

References

1. Bullard *et al.* 2007 *Journal of Experimental Marine Biology and Ecology,* 342: 99-108.
2. Kearse *et al. Bioinformatics*, 28(12), 1647-1649.
3. Stefaniak *et al.* 2012 *Mar. Biol. Ecol.* 422-423, 64-71
4. Smith *et al.* 2012 *PLoS ONE* 7 (1)
5. Clancy *et al.* 2015 *San Francisco State University, CA*

Acknowledgments

Contact

- Cecilia Hernandez - ceciliah@mail.sfsu.edu
- C. Sarah Cohen - sarahcoh@sfsu.edu