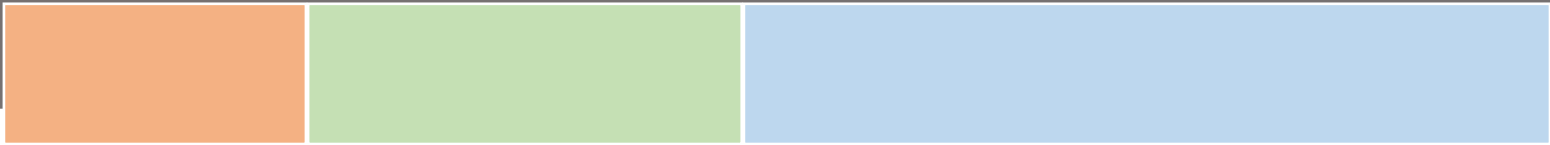


Refining the Edges of the Columbia River Eulachon Run

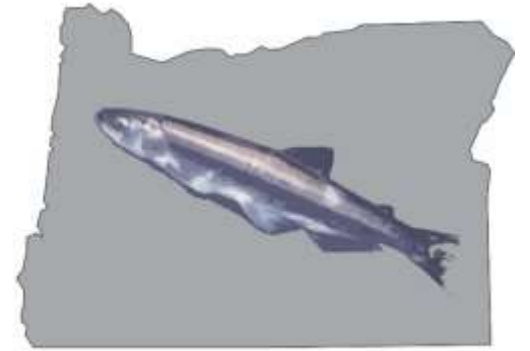
Year 1 Pilot Study



Joe Krieter¹, Taal Levi², Maria Sandercock¹,
Andy Clodfelter¹, Michelle Hollis³

1. Hart Crowser Inc., Natural Resources Division, Portland, Oregon
2. Oregon State University, Quantitative Wildlife Ecology and Conservation Lab, Corvallis, Oregon
3. Port of Portland, Environmental Division, Portland, Oregon

Ourigan?

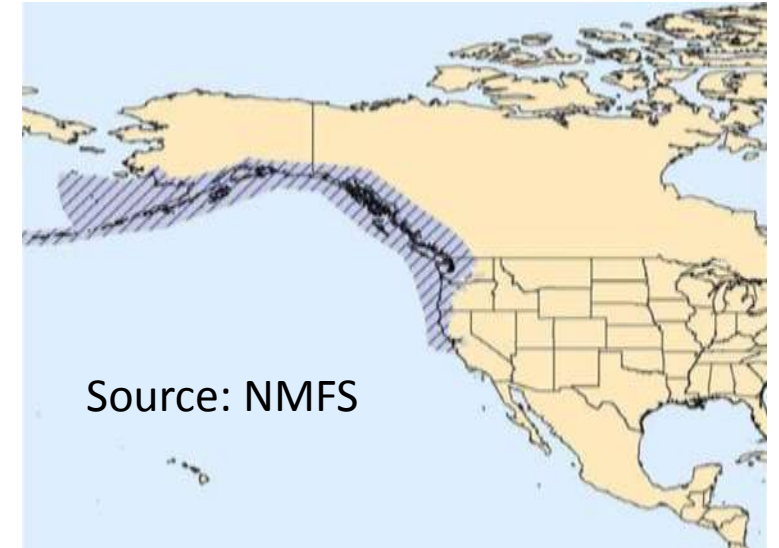


- *Thaleichthys pacificus* (Smelt), commonly known by First Nations and Native Americans as *ooligan*, *oolichan*, *hooligan*, and other variants.
- Western-most Cree speakers used the [r] sound in place of the [l], thus *ooligan*, was pronounced *urigan* or *oorigan*. River name and trade route spelled Ourigan by a British officer in the 1760s.

From: S. Byram and D.G. Lewis. 2001. *Ourigan*, Wealth of the Northwest Coast. Oregon Historical Quarterly. Vol 102. No. 2. pp. 127 – 157.

Eulachon Listing Status

- Negative Finding on Petition to List Eulachon 1999
- Positive Finding on Petition to List 2008
- Proposed Rule 2009
- **Final Rule 2010 - Southern Eulachon Distinct Population Segment Listed as Threatened under the Endangered Species Act.**
- Proposed Critical Habitat Designation 2011
- **Final Critical Habitat Designation 2011**
- Intent to Prepare a Recovery Plan 2013 – Recovery Coordinator



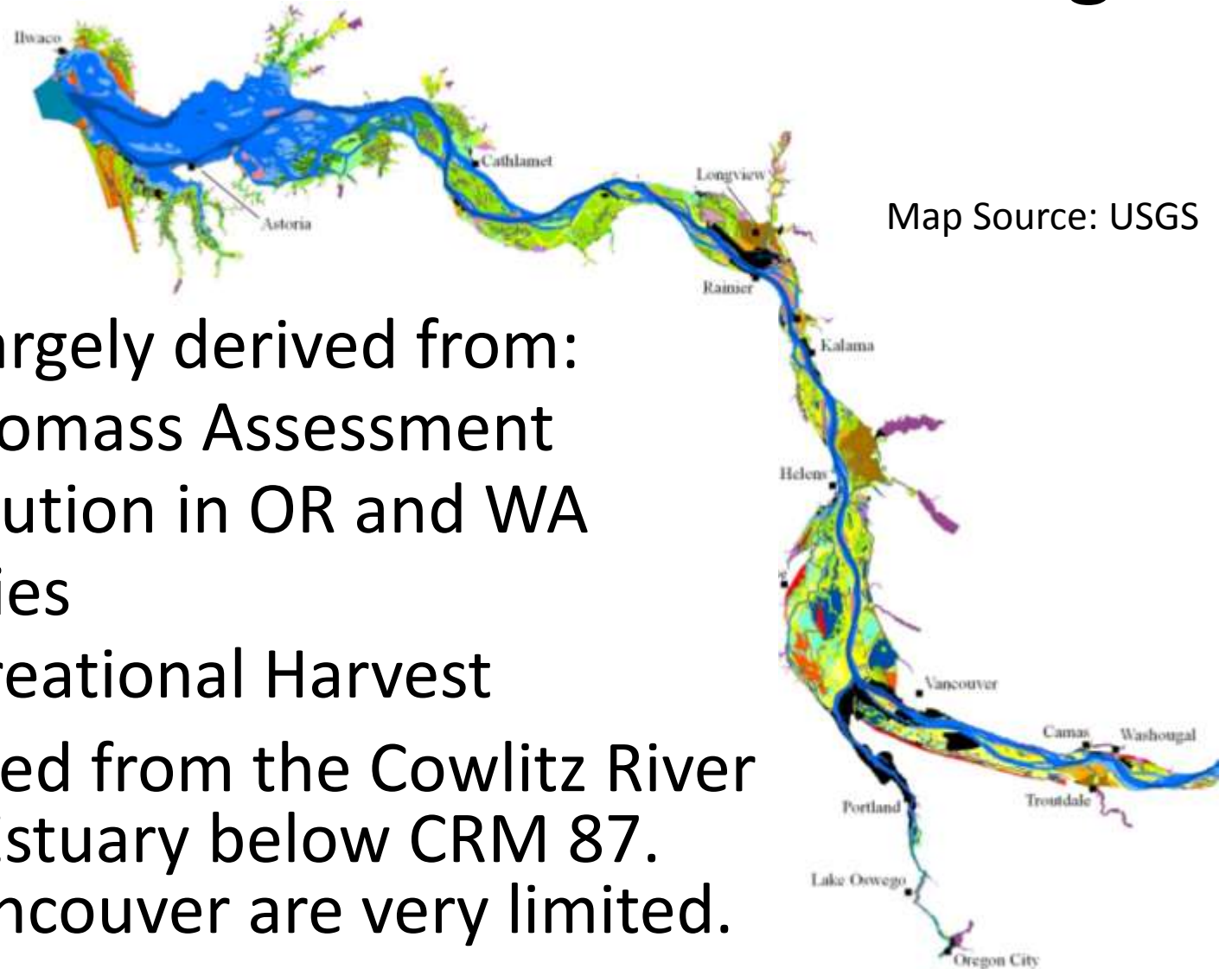
DETAILS: <http://www.fisheries.noaa.gov/pr/species/fish/eulachon.html>

Dredging and In-Water Work Windows

- ODFW Columbia River In-Water Work Window:
 - November 1 – February 28
 - October 15 – December 15?
 - Established to reduce impacts by avoiding peak salmonid runs.
 - Eulachon concerns not directly addressed.
 - Many unknowns and uncertainties.



Lower Columbia River Eulachon Run Timing



- Current knowledge largely derived from:
 - Spawning Stock Biomass Assessment
 - Freshwater Distribution in OR and WA
 - Cowlitz Tribe Studies
 - Commercial / Recreational Harvest
- Most data are collected from the Cowlitz River and Columbia River Estuary below CRM 87. Data in Portland / Vancouver are very limited.

Challenges Sampling Eulachon

- Capture methods used / attempted
 - Trawls and seines
 - Fyke nets
 - ARIS (DIDSON) cameras
- Crazy ideas – our focus non-intrusive / lower cost
 - UAVs?
 - LCR Eulachon app?
 - How about environmental DNA (eDNA)!



eDNA

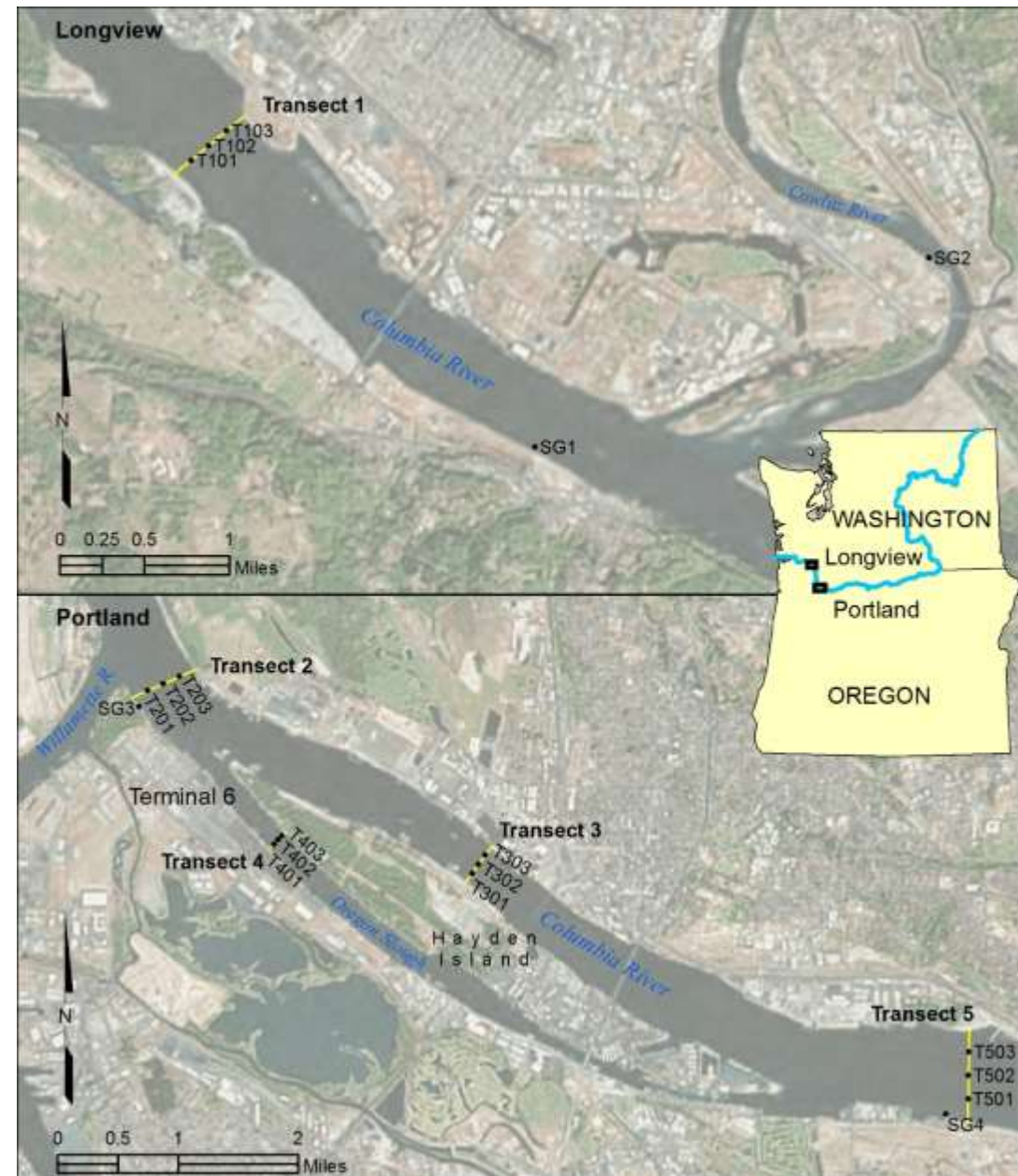


- “Genetic material obtained directly from environmental samples (soil, sediment, water, etc.) without any obvious signs of biological source material”
- Advantages
 - Non-invasive
 - Highly sensitive
 - Short degradation time
 - Cost-efficient
- Disadvantages
 - No info. about DNA source (life stage, condition, etc.)
 - DNA can be detected from sources other than target

From: P.H. Thomsen and E. Willersley. 2015. Environmental DNA – An Emerging Tool in Conservation for Monitoring Past and Present Biodiversity. *Biological Conservation*. Vol 183. pp. 4 – 18.

Pilot Field Sampling Program - Transects

- Sampling Sites
 - Transect 1 – Columbia River @ Cowlitz River Plume
 - Transect 2 – Kelley Point
 - Transect 5 – Broughton Beach
 - Shore accessible grab samples at Rainier, Cowlitz River, Kelley Point, and Gleason Boat Ramp.



Field Data Collection

- Water Collection

- Approx. every two weeks from December 15 through March 15
- Scheduled during late ebb
- Navigate and hold R/V Catalyst on predetermined GPS positions.
- 2L Van Dorn water sampler
- 3 per transect. Mid-depth.
- Strict decontamination procedures.



Goldberg, C.S., Pilliod, D.S., Arkle, R.S., and L.P. Waits. 2011. Molecular detection of vertebrates in stream water: A demonstration using Rock Mountain tailed frogs and Idaho giant salamanders. PLoS ONE 6(7): e22746.

Jerde, C.L., Mahon, A.R., Chadderton, W.L., and D.M. Lodge. 2011. "Sight-unseen" detection of rare aquatic species using environmental DNA. Conservation Letters 4: 150-157.

Field Data Collection

- Filtering
 - 1L Sterile Bottles
 - 0.45 μm cellulose nitrate filter
 - Peristaltic pumps
 - 1L graduated cylinder
 - 100% molecular-grade ethanol
 - Ethanol-proof lab pens

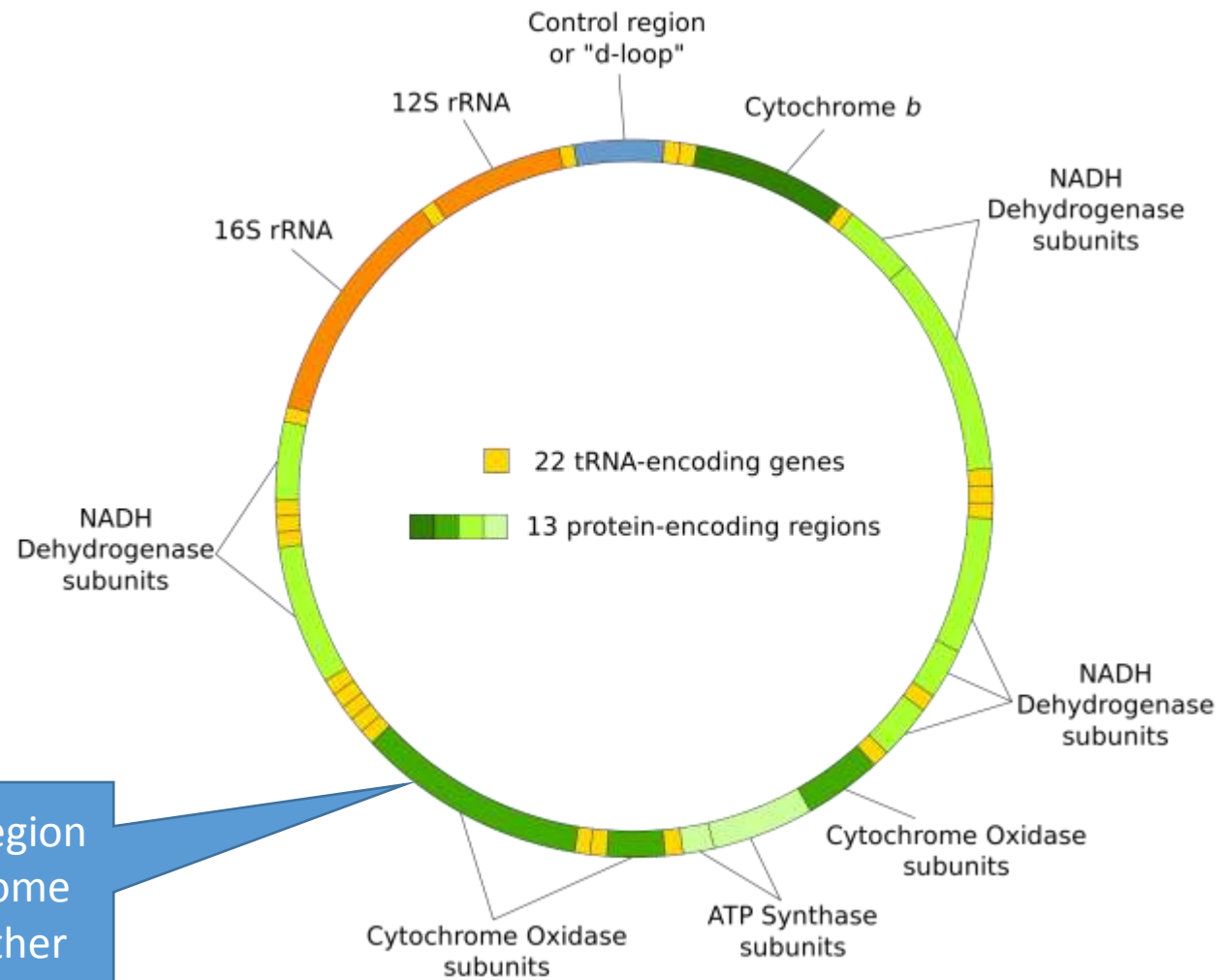


Field Data Collection

- Filtering – Modifications
 - Switched to whirl-pak[®] bags
 - Move filtering inside
 - = Happy crew
- Anticipated Refinements
 - Vacuum pump and manifold
 - Decrease filtering time by running 4 at a time



Mitochondrial Genome

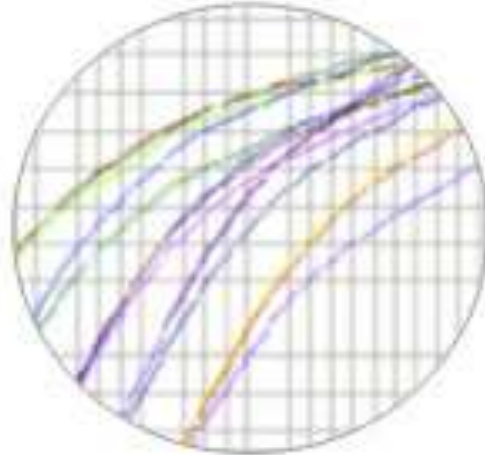


Designed primer for COI region of the mitochondrial genome that would not amplify other Osmeridae fish

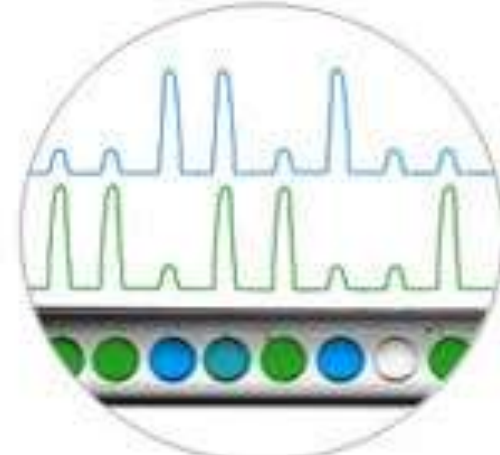
Monitoring Presence... and Abundance?



PCR
Qualitative

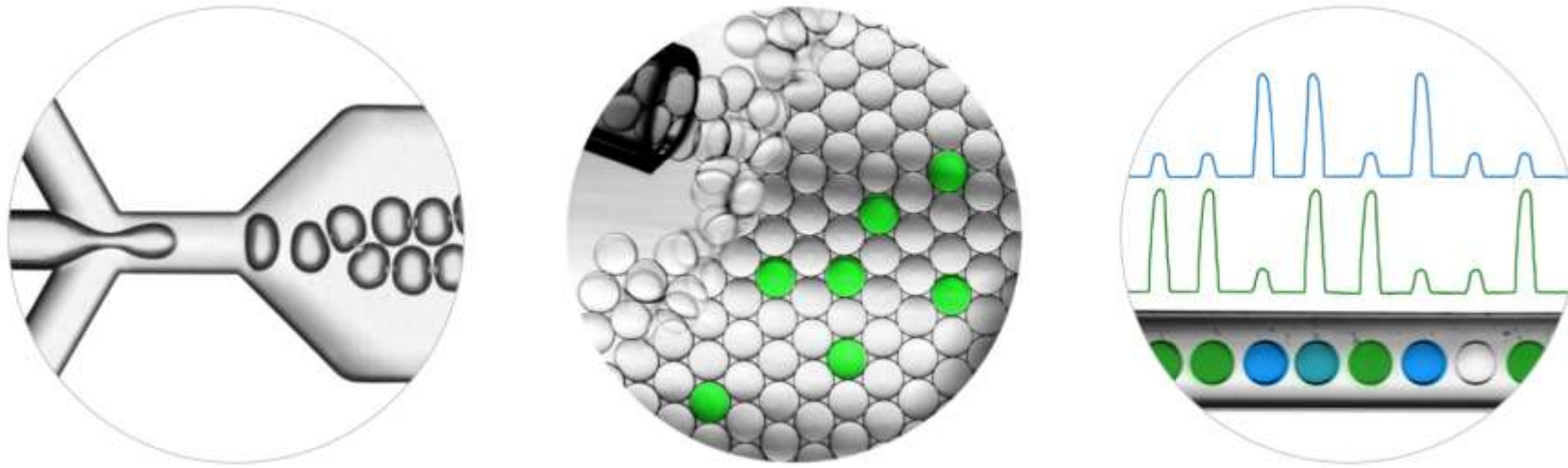


Real-Time PCR
Relative Quantitation

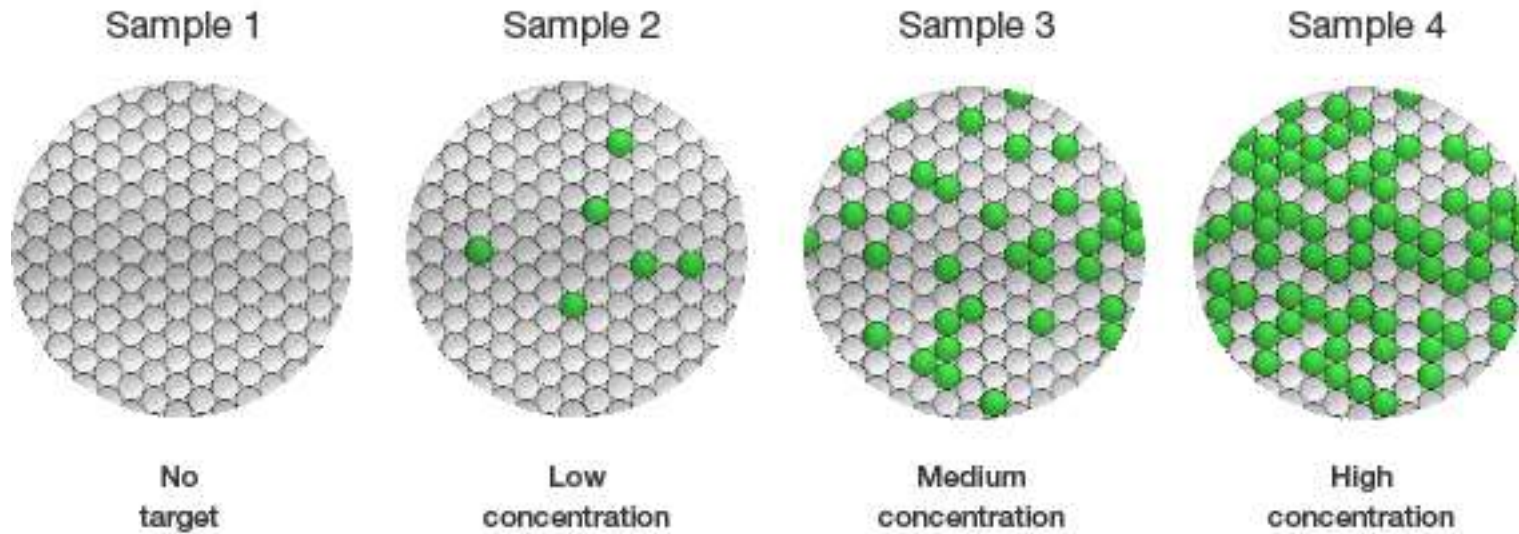


Droplet Digital PCR
Absolute Quantitation

Absolute quantification with Droplet digital PCR

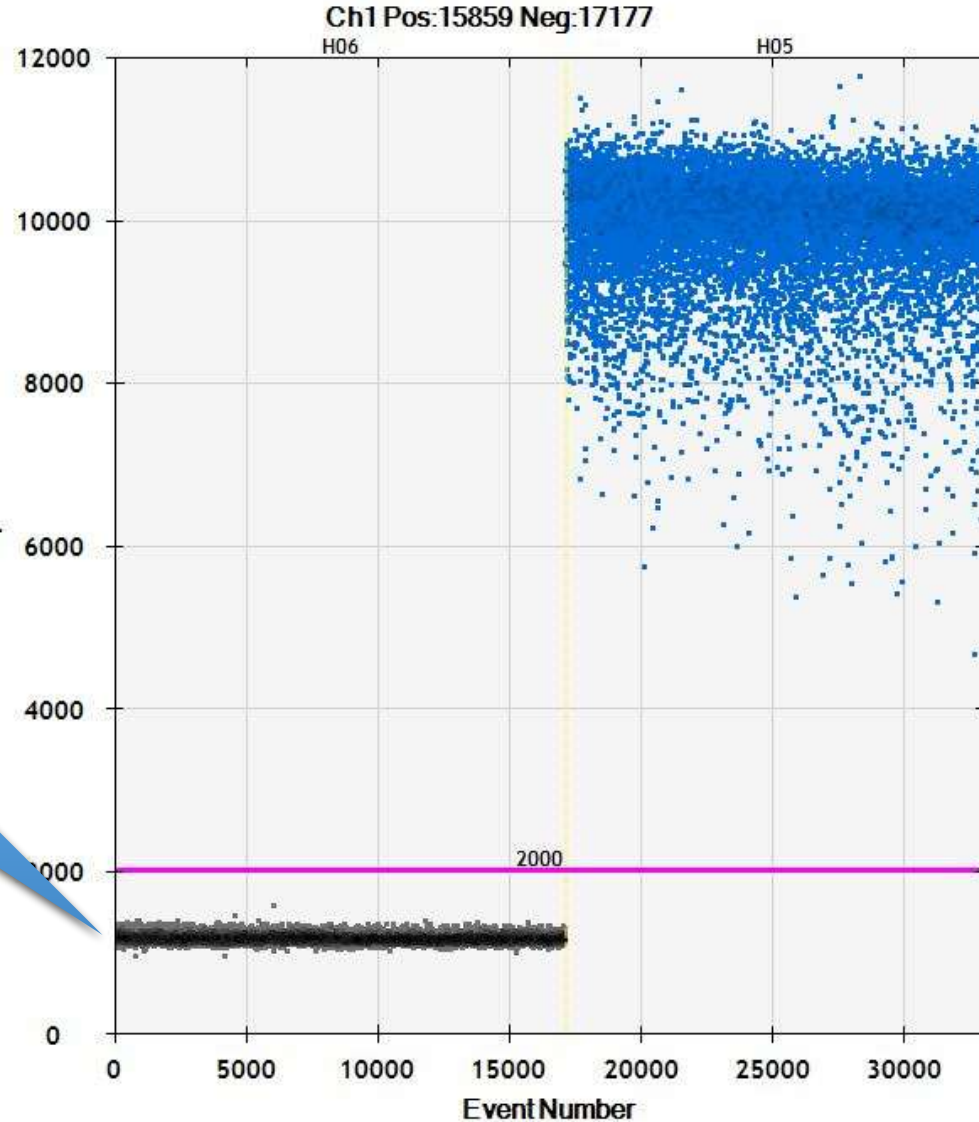


Absolute Quantification



1. High accuracy
2. High sensitivity
3. No standard curve
4. Overcomes PCR inhibition
5. But expensive initial purchase

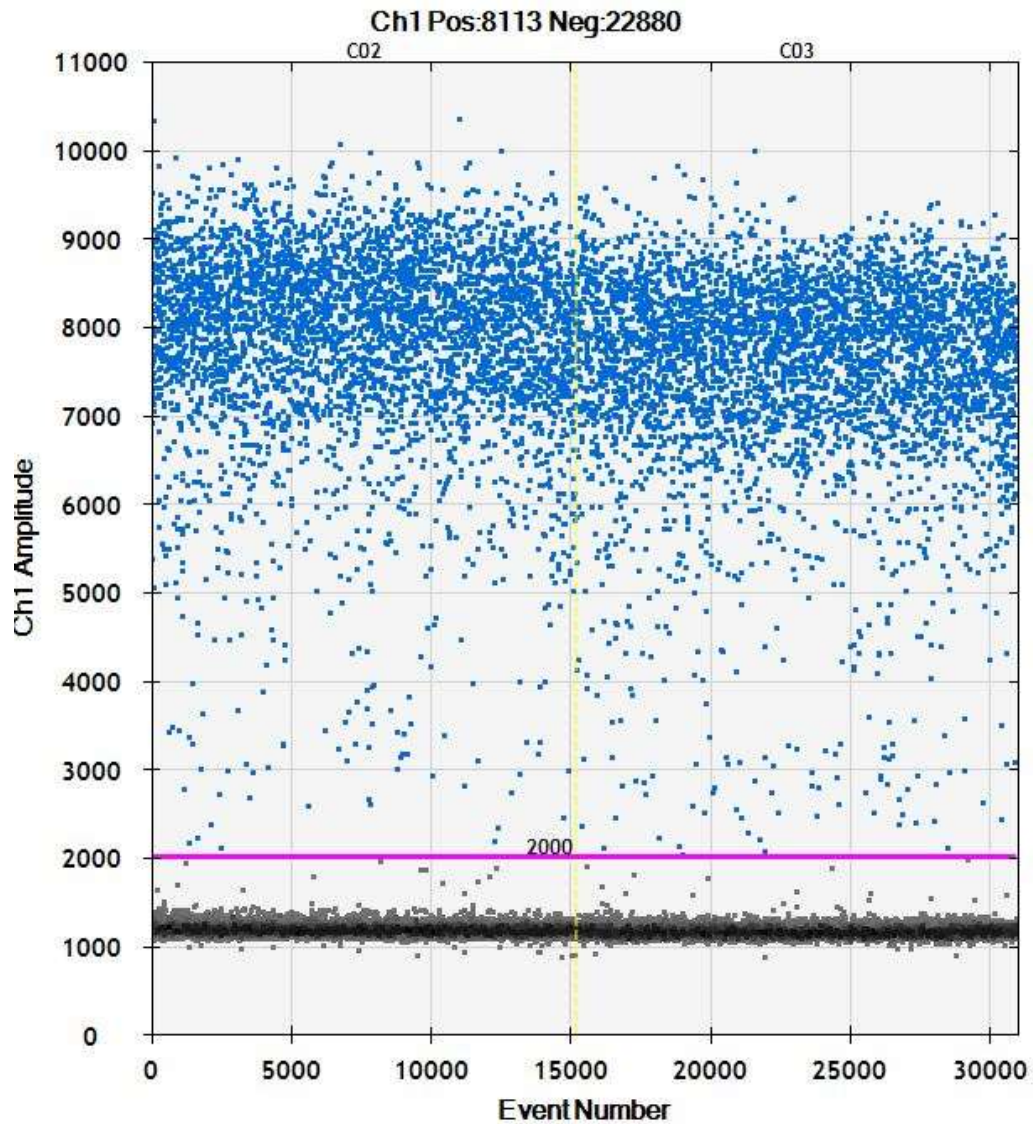
Positive and Negative Controls



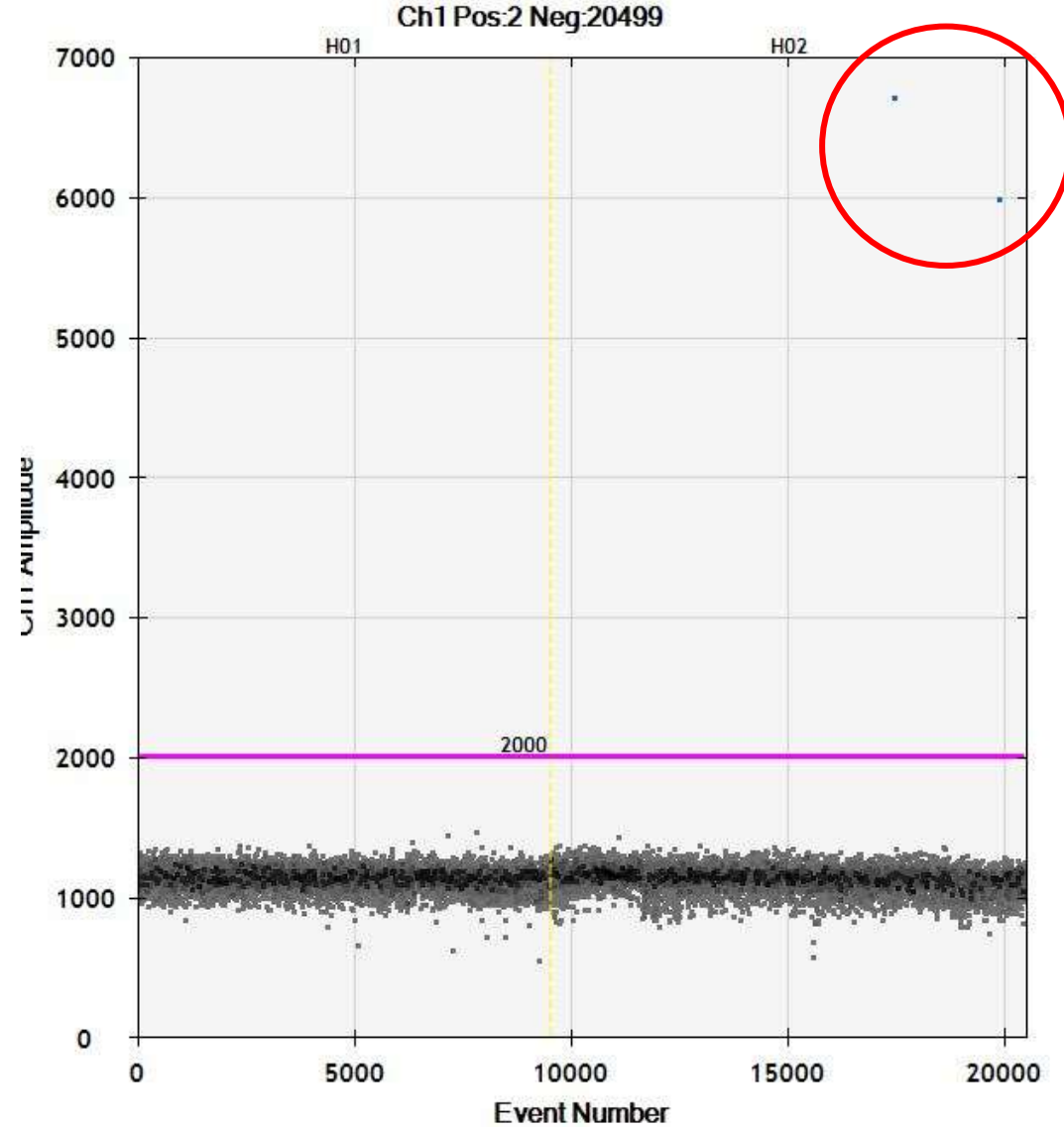
Eulachon tissue

No template control

Eulachon abundant

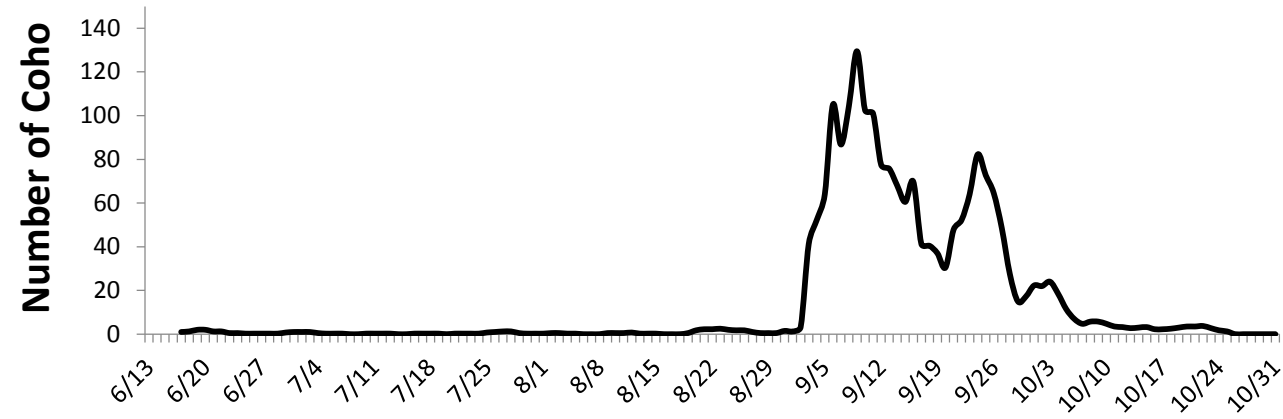


Eulachon Rare

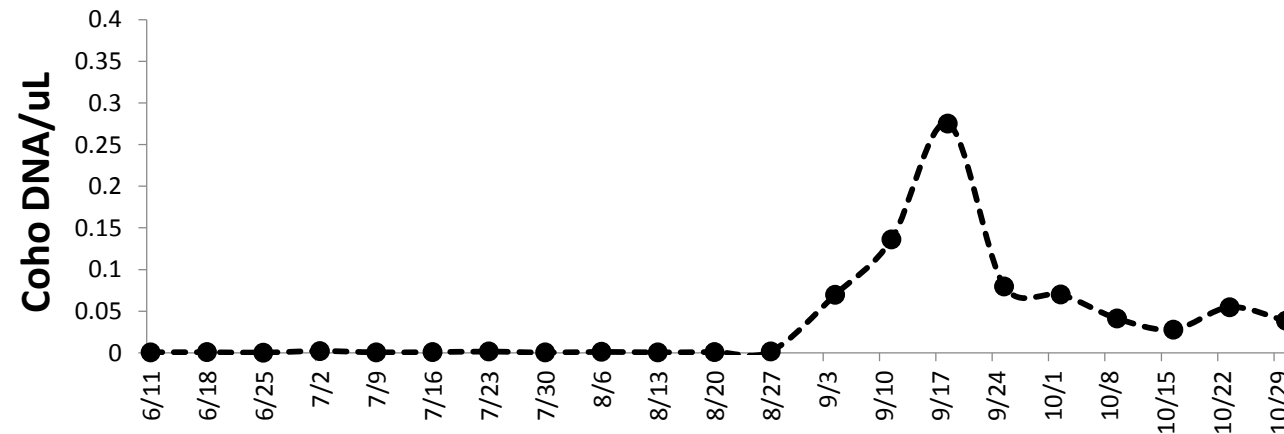


Coho eDNA vs. fish counts

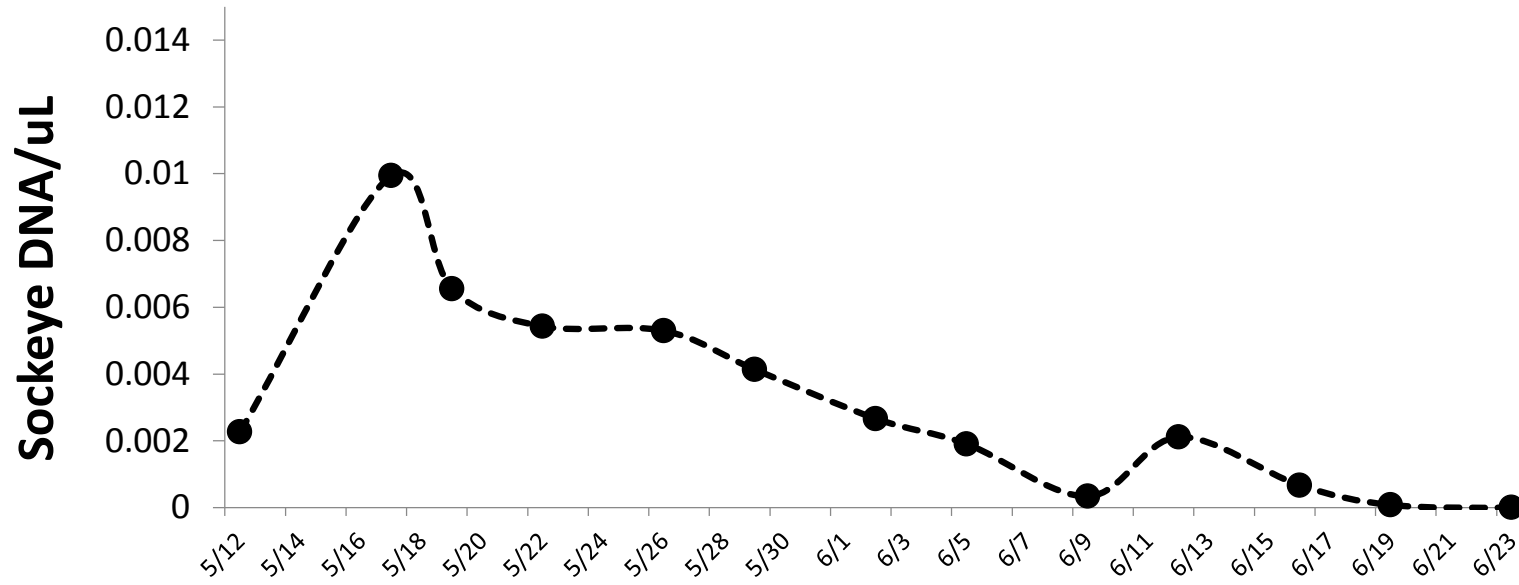
4 day moving average counts



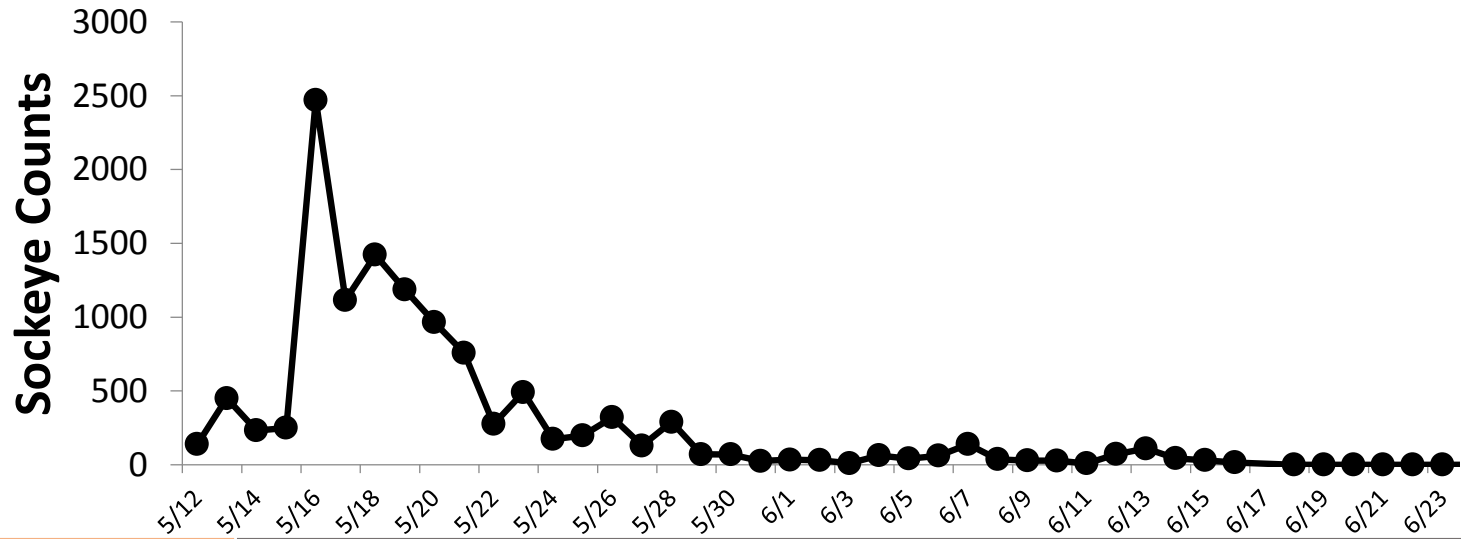
Coho eDNA estimated concentration



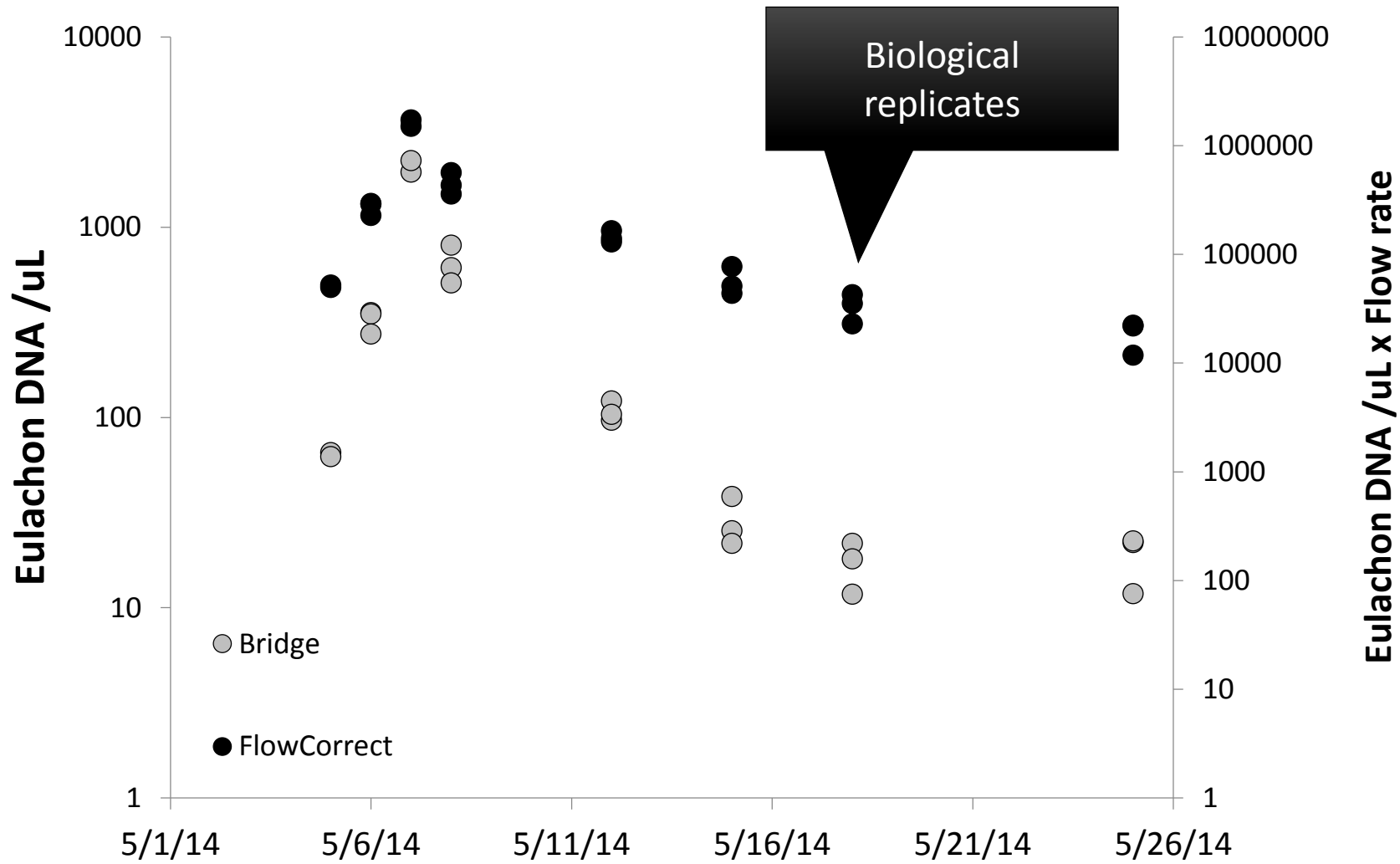
Outmigrating sockeye eDNA Concentration-2015



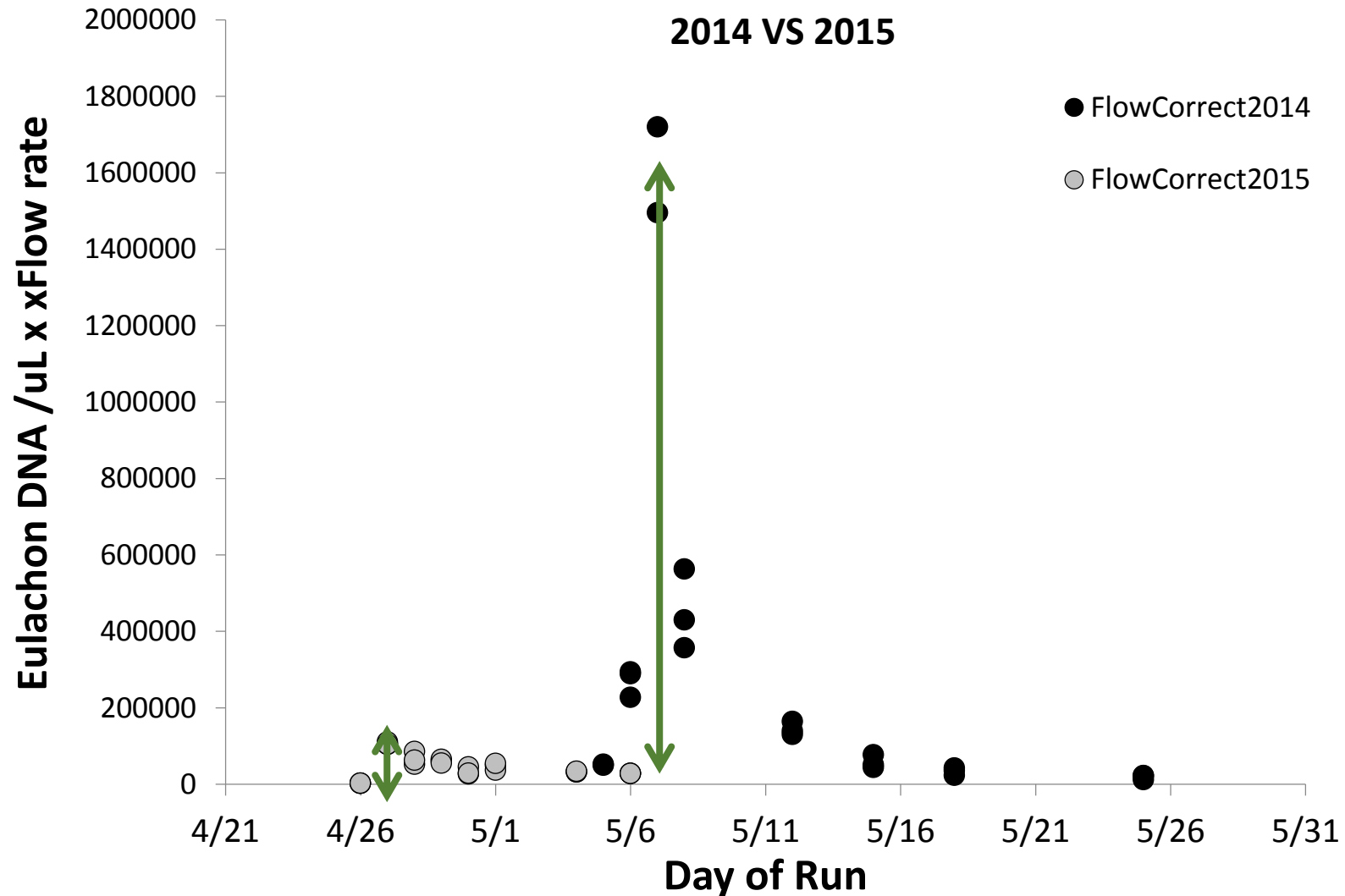
Sockeye counts



2014 Eulachon ddPCR results



Large 2014 run compared with failed 2015 run



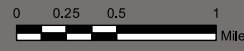
Cowlitz River and mainstem Columbia River monitoring



Document Path: F:\notebooks\1276917_POP_Eulachon Pilot Study\GIS\1276917_SPL(Longview)_2.mxd Date: 1/7/2016 User Name: malissaschwelzer

Source: Source: Esri, DigitalGlobe, GeoEye, Earthstar Geographics, CNES/Airbus DS, USDA, USGS, AEX, Getmapping, Aerogrid, IGN, IGP, swisstopo, and the GIS User Community

- LEGEND**
- ✕ Transect Station
 - ✕ Grab Station
 - eDNA Transects
 - ◆ River Mile Marker

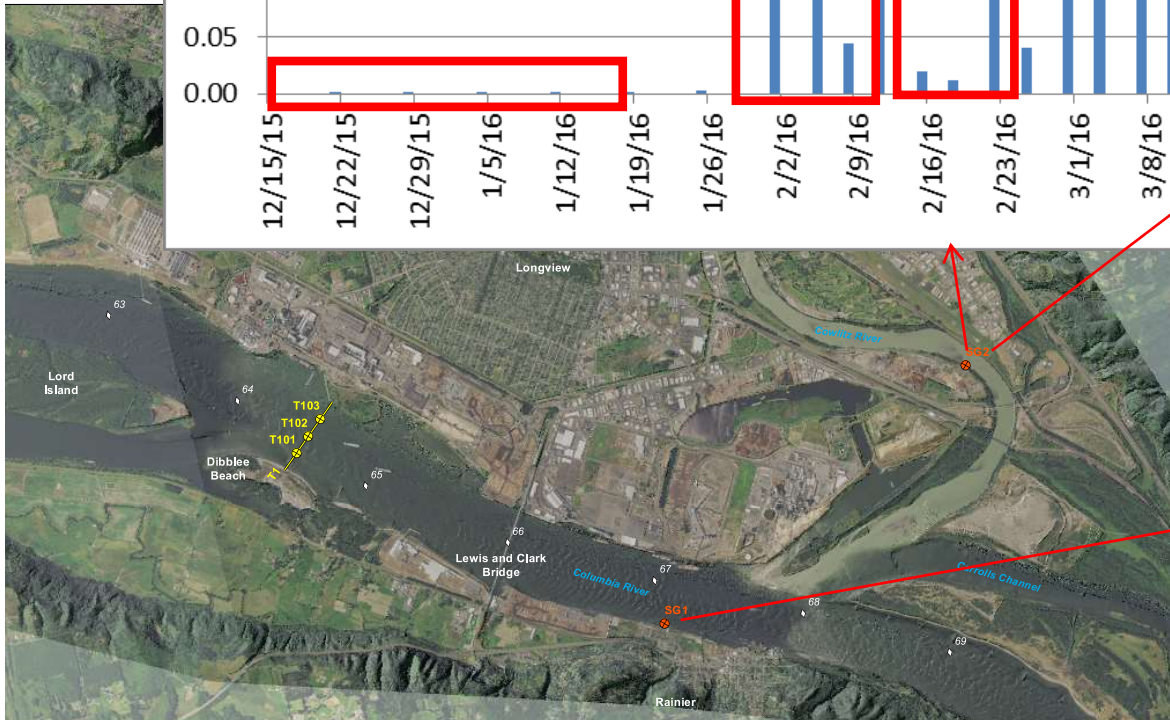
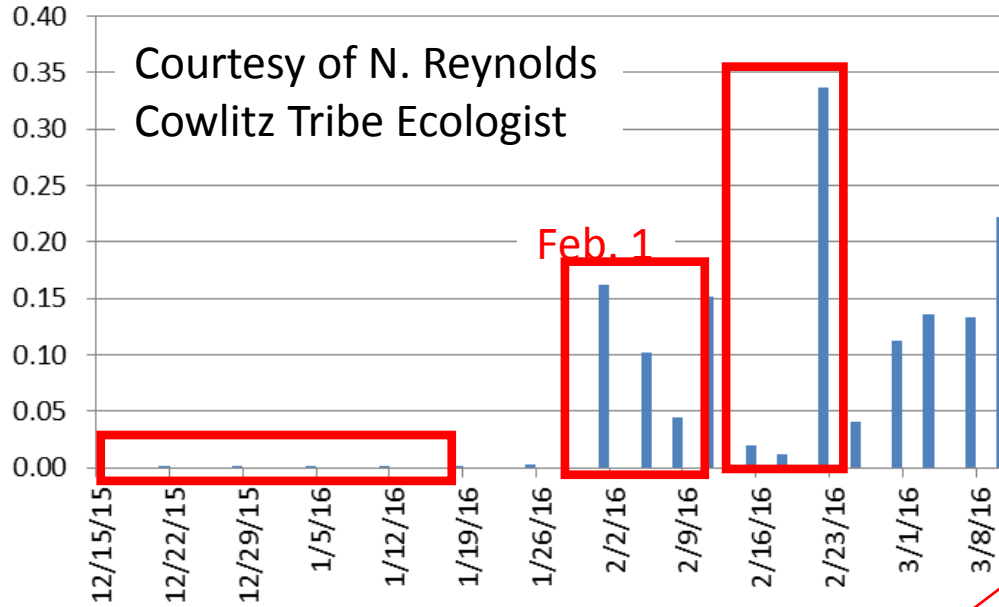


Eulachon Pilot Study Portland, Oregon	
Sampling Locations Cowlitz River Confluence	
12769-17	12/15
	Figure 2

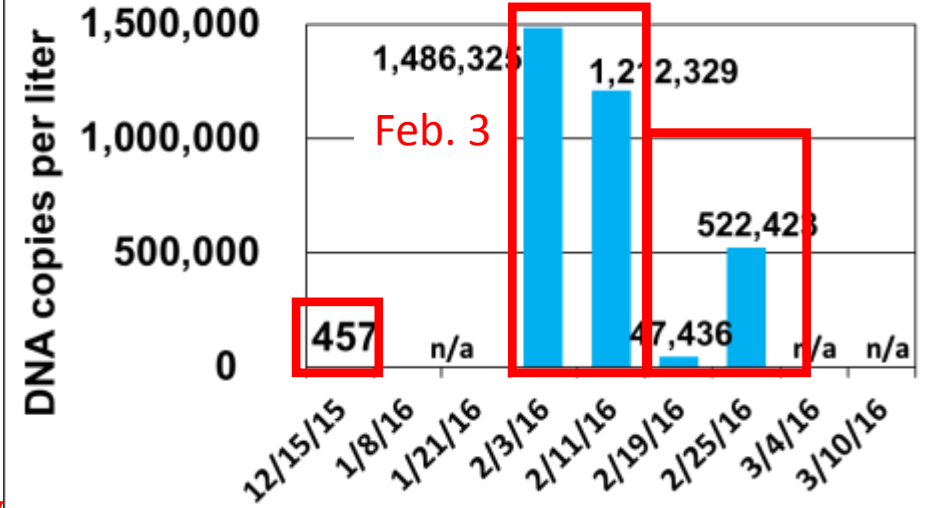


Cowlitz Tribe Cowlitz Eulachon presence index

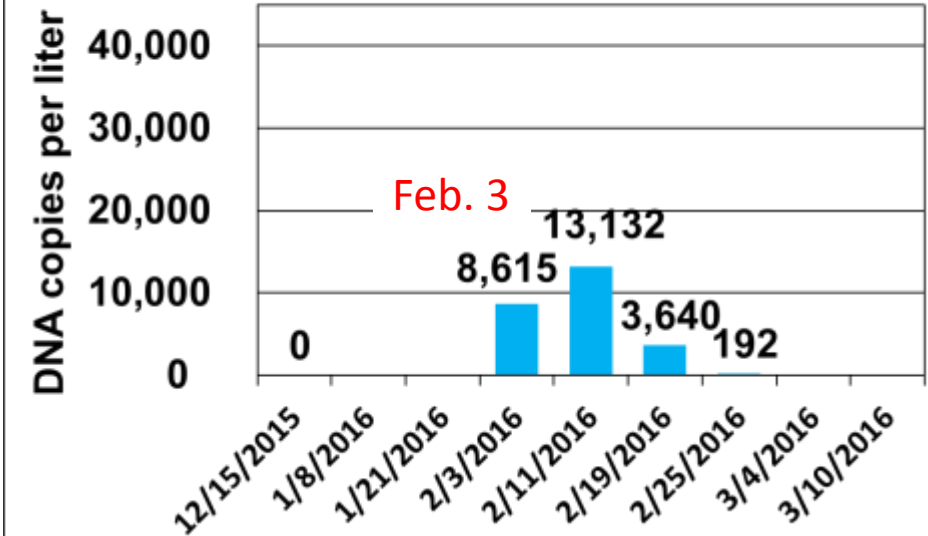
Courtesy of N. Reynolds
Cowlitz Tribe Ecologist



Sample Grab 2 - Cowlitz

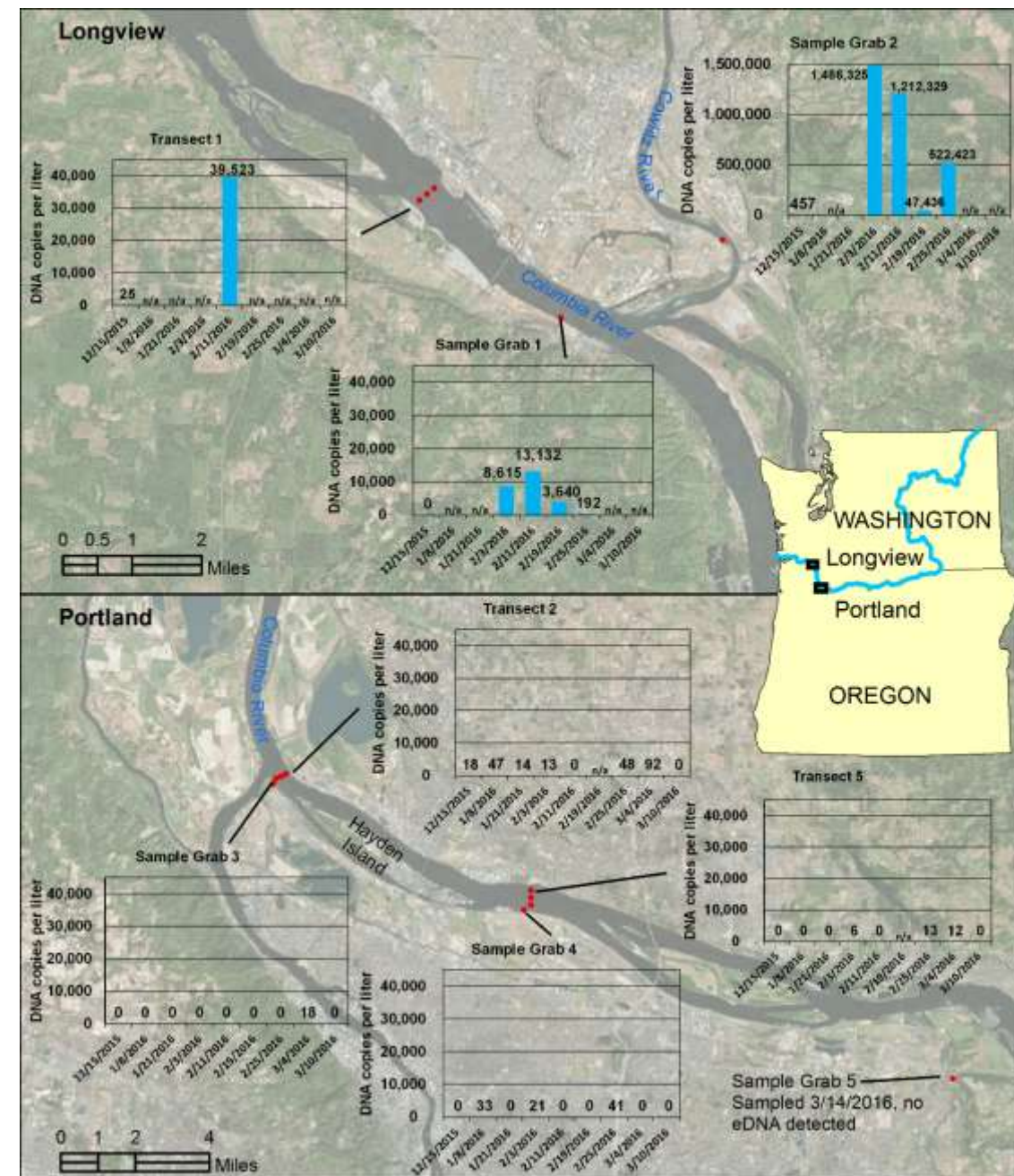


Sample Grab 1 - Rainier



Preliminary Results –

- ddPCR techniques highly sensitive
- Successful and consistent detection of Eulachon in the Columbia River below Cowlitz
- No Sandy River Eulachon observed in 2016, but very small amounts of DNA detected
- eDNA should allow us to improve our knowledge of run timing in Portland/Vancouver reach



What's Next?

- Two more years of study at higher spatiotemporal resolution
- Will we see a Sandy River Eulachon run in 2017? 2018?
- Tightened focus on run timing in the Portland / Vancouver reach relative to the run timing of Cowlitz River Eulachon

Thank You!

- Please send us your questions and comments:

Joe Krieter, MS
Natural Resource Department
Hart Crowser, Inc.
joe.krieter@hartcrowser.com
971.327.9103

Taal Levi, PhD
Department of Fisheries and Wildlife
Oregon State University
Taal.Levi@oregonstate.edu
541.737.4067

Data Exploration - Preliminary



Date	Columbia River Below Cowlitz River				Cowlitz R.	Columbia R. Above Willamette R.								Sandy R.
	SG1	T101	T102	T103	SG2	T201	T202	T203	SG3	T501	T502	T503	SG4	SG5
12/15/2015	0	0	75 ¹	0	457	53 ¹	0	0	0	0	0	0	0	n/a
1/8/2016	n/a	n/a	n/a	n/a	n/a	0	142 ²	0	0	0	0	0	33 ¹	n/a
1/21/2016	n/a	n/a	n/a	n/a	n/a	0	0	43 ¹	0	0	0	0	0	n/a
2/3/2016	8,615	n/a	n/a	n/a	1,486,325	0	0	38 ¹	0	0	0	17 ¹	21 ¹	n/a
2/11/2016	13,132	7,726	24,636	86,207	1,212,329	0	0	0	0	0	0	0	0	n/a
2/19/2016	3,640	n/a	n/a	n/a	47,436	n/a	n/a	n/a	0	n/a	n/a	n/a	0	n/a
2/25/2016	192	n/a	n/a	n/a	522,423	114 ¹	0	33	0	0	0	39	41	n/a
3/4/2016	n/a	n/a	n/a	n/a	n/a	86 ²	138 ²	45 ¹	18 ¹	0	0	35	0	n/a
3/10/2016	n/a	n/a	n/a	n/a	n/a	0	0	0	0	0	0	0	0	n/a
3/14/2016	n/a	n/a	n/a	n/a	n/a	n/a	n/a	n/a	n/a	n/a	n/a	n/a	n/a	0

1. Considered a non-detect based on assumptions described in Section 2.1.3.
2. Large number of seagulls observed just upstream during sampling.

When looking “apples to apples”, Positive DNA counts normalized by River discharge are similar from an order of magnitude perspective during periods of moderate to high abundance.

Date	Cowlitz River			Columbia River		
	DNA Copies/L	Discharge (CFS) ¹	DNA count x CFS	DNA Copies/L	Discharge (CFS) ²	DNA count x CFS
12/15/2015	457	26,600	1.21E+07	ND (25 ³)	466,100	ND (1.16E+07)
2/3/2016	1,486,325	16,700	2.48E+10	8,615 ⁴	273,000	2.35E+09
2/11/2016	1,212,329	16,400	1.99E+10	39,523 ³	396,200	1.57E+10
2/19/2016	47,436	22,000	1.04E+09	3,640 ⁴	317,000	1.15E+09
2/25/2016	522,423	14,800	7.73E+09	192 ⁴	394,000	7.57E+07

ND = Not detected per QC/QA criteria.

1. As measured at USGS Station Number 14243000 at Castle Rock, WA.
2. As measured at USGS Station Number 14246900 at Beaver Army Terminal near Quincy, OR.
3. Mean of Transect 1 results.
4. Beach grab results (no transect data).