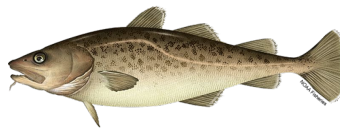


## ***Molecular indicators of performance in Pacific cod exposed to climate stressors***



***Laura Spencer<sup>12</sup>, Ben Laurel<sup>3</sup>, Emily Slesinger<sup>3</sup>, Ingrid Spies<sup>1</sup>, Mary Beth Rew Hicks<sup>3</sup>,  
Louise Copeman<sup>34</sup>, Sara Schaal<sup>1,6</sup>, Laura Timm<sup>1,5</sup>, Wes Larson<sup>5</sup>, Michelle Stowell<sup>4</sup>,  
Samantha Mundorff<sup>3</sup>, Carlissa Salant<sup>3</sup>, Kathleen Durkin<sup>2</sup>,  
Tom Hurst<sup>3</sup>, Steven Roberts<sup>2</sup>***

<sup>1</sup>NOAA AFSC, Seattle

<sup>2</sup>University of Washington SAFS, Seattle

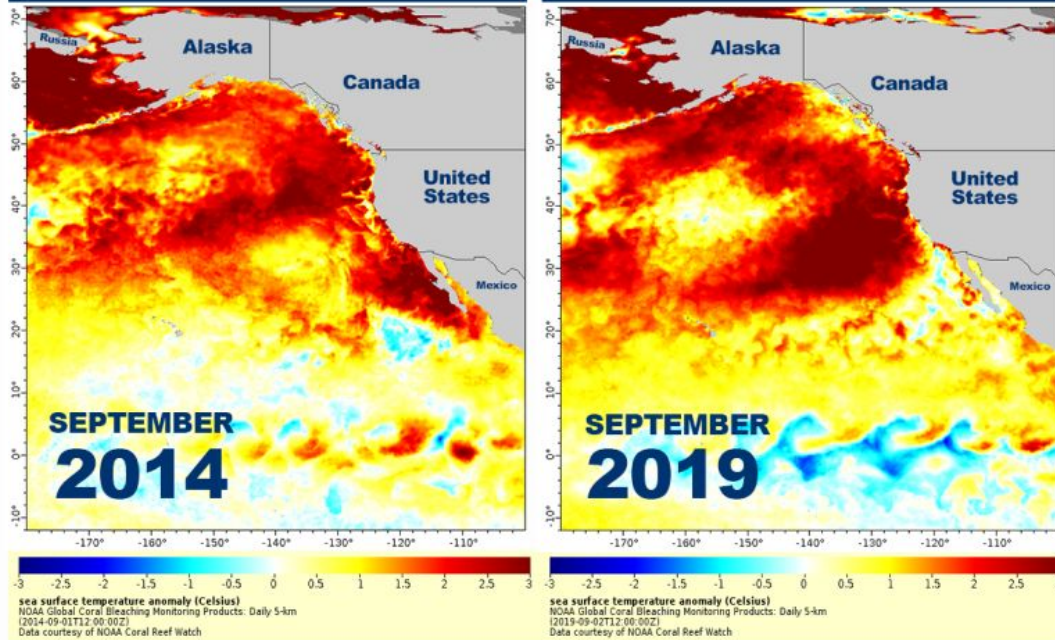
<sup>3</sup>NOAA AFSC, Newport

<sup>4</sup>Oregon State University

<sup>5</sup>NOAA AFSC, Juneau

<sup>6</sup>University of Oklahoma

# Gulf of Alaska cod fishery closed in 2020 after marine heatwaves



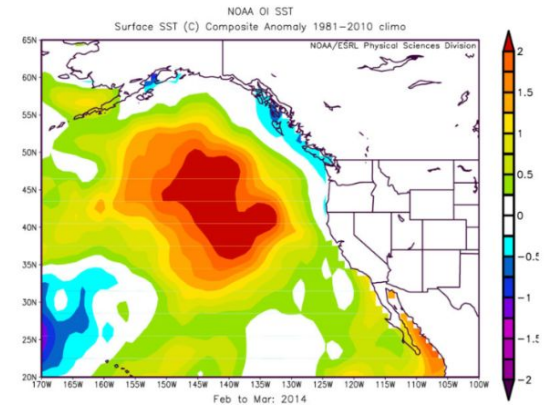
[NOAA feature story](#), September 05, 2019

December 10, 2019

## The Blob returns: Alaska cod fishery closes for 2020

by Jessica Hathaway in News, Alaska

SHARE [f](#) [t](#) [in](#) [e](#)



"The Blob" of 2014 severely depressed the Gulf of Alaska cod population. NOAA image.

ORIGINAL ARTICLE | [Open Access](#) | [CC BY](#)

## Impact of the 2014–2016 marine heatwave on US and Canada West Coast fisheries: Surprises and lessons from key case studies

[Correction\(s\) for this article](#) ▾

Christopher M. Free [✉](#), Sean C. Anderson, Elizabeth A. Hellmers, Barbara A. Muhling, Michael O. Navarro, Kate Richerson, Lauren A. Rogers, William H. Satterthwaite, Andrew R. Thompson, Jenn M. Burt, Steven D. Gaines, Kristin N. Marshall, J. Wilson White, Lyall F. Bellquist ... [See fewer authors](#) ^

First published: 20 April 2023 | <https://doi.org/10.1111/faf.12753> | Citations: 6

## scientific reports

[Explore content](#) ▾ [About the journal](#) ▾ [Publish with us](#) ▾[nature](#) > [scientific reports](#) > [articles](#) > [article](#)[Article](#) | [Open access](#) | Published: 27 June 2024

## Marine heatwaves alter the nursery function of coastal habitats for juvenile Gulf of Alaska Pacific cod

Hillary L. Thalmann [✉](#), Benjamin J. Laurel, L. Zoe Almeida, Kaitlyn E. Osborne, Kaylee Marshall & Jessica A. Miller

[Scientific Reports](#) **14**, Article number: 14018 (2024) | [Cite this article](#)



## Loss of spawning habitat and prerecruits of Pacific cod during a Gulf of Alaska heatwave

Benjamin J. Laurel and Lauren A. Rogers

ORIGINAL ARTICLE | [Open Access](#) | [CC BY](#) [i](#) [S](#)

## Pacific cod in the Anthropocene: An early life history perspective under changing thermal habitats

Benjamin J. Laurel [✉](#), Alisa Abookire, Steve J. Barbeaux, L. Zoe Almeida, Louise A. Copeman, Janet Duffy-Anderson, Thomas P. Hurst, Michael A. Litzow, Trond Kristiansen, Jessica A. Miller, Wayne Palsson, Sean Rooney, Hillary L. Thalmann, Lauren A. Rogers ... [See fewer authors](#) ^

First published: 27 July 2023 | <https://doi.org/10.1111/faf.12779> | Citations: 1

## ORIGINAL RESEARCH article

Front. Mar. Sci., 19 August 2020

Sec. Marine Affairs and Policy

Volume 7 - 2020 | <https://doi.org/10.3389/fmars.2020.00703>

## Marine Heatwave Stress Test of Ecosystem-Based Fisheries Management in the Gulf of Alaska Pacific Cod Fishery



Steven J. Barbeaux \*



Kirstin Holsman



Stephani Zador



## Pacific cod, *Gadus macrocephalus*



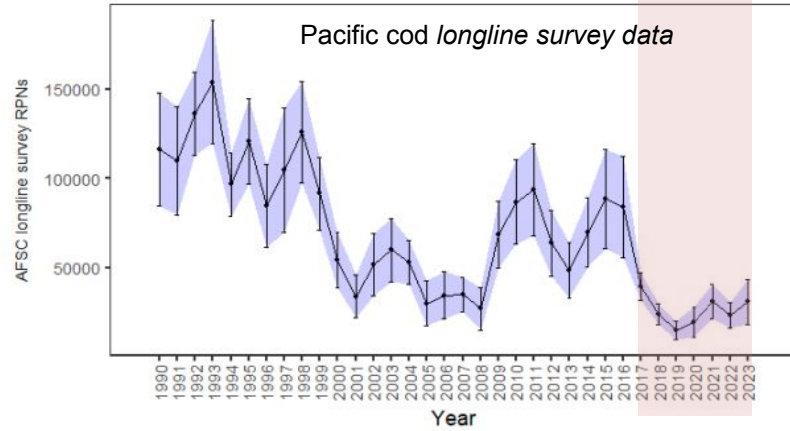


## Pacific cod, *Gadus macrocephalus*



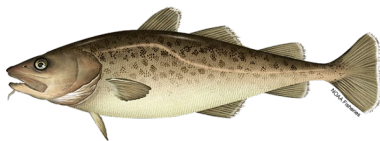


## Pacific cod, *Gadus macrocephalus*

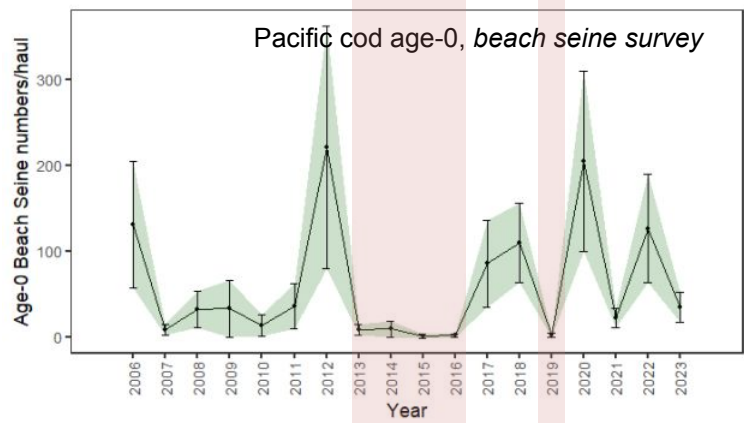
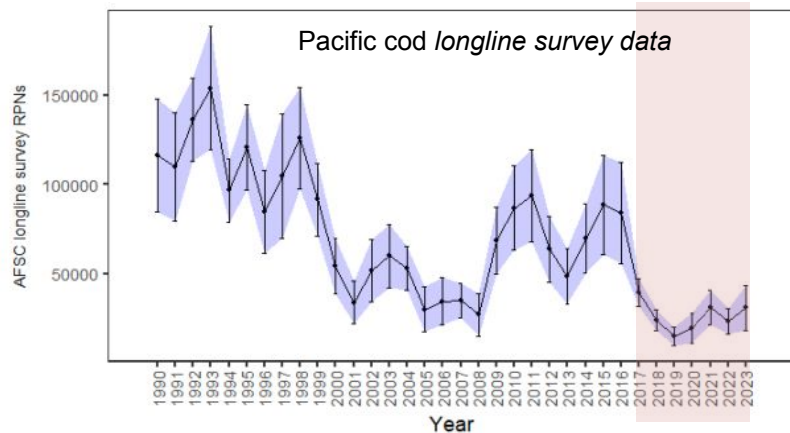


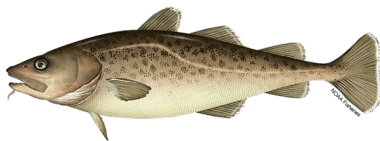
Hoff, Stevenson, & Orr 2015



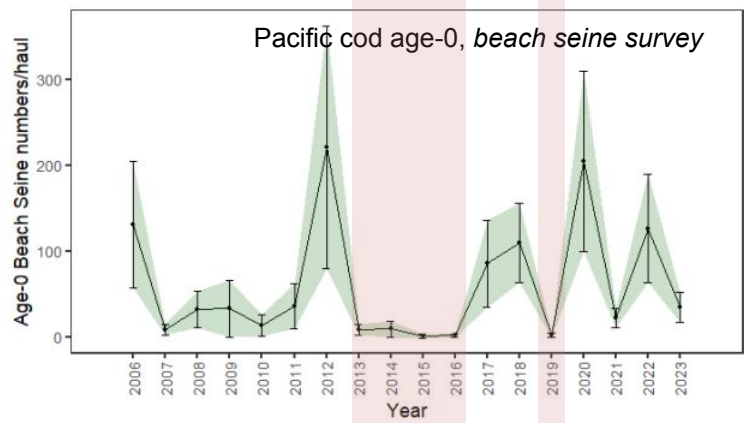
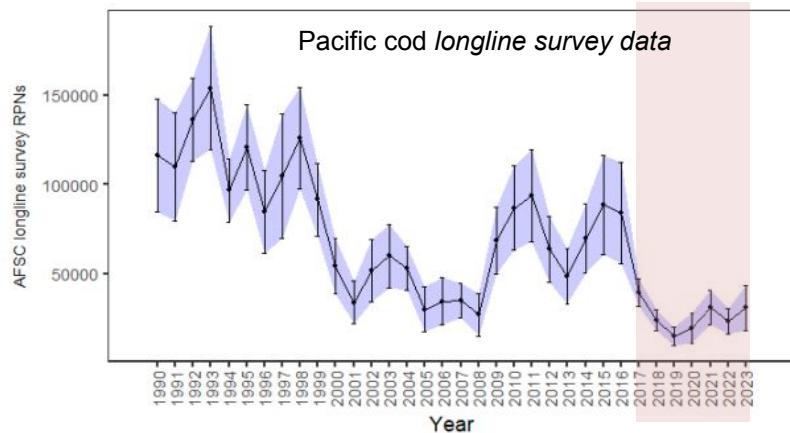


## Pacific cod, *Gadus macrocephalus*





## Pacific cod, *Gadus macrocephalus*



Low Pacific cod recruitment and biomass estimates in Gulf of Alaska coincided/followed the 2014-16 & 2019 marine heatwaves, prompting **review of 1st year of life biology and temperature response experiments**

[Hoff, Stevenson, & Orr 2015](#)

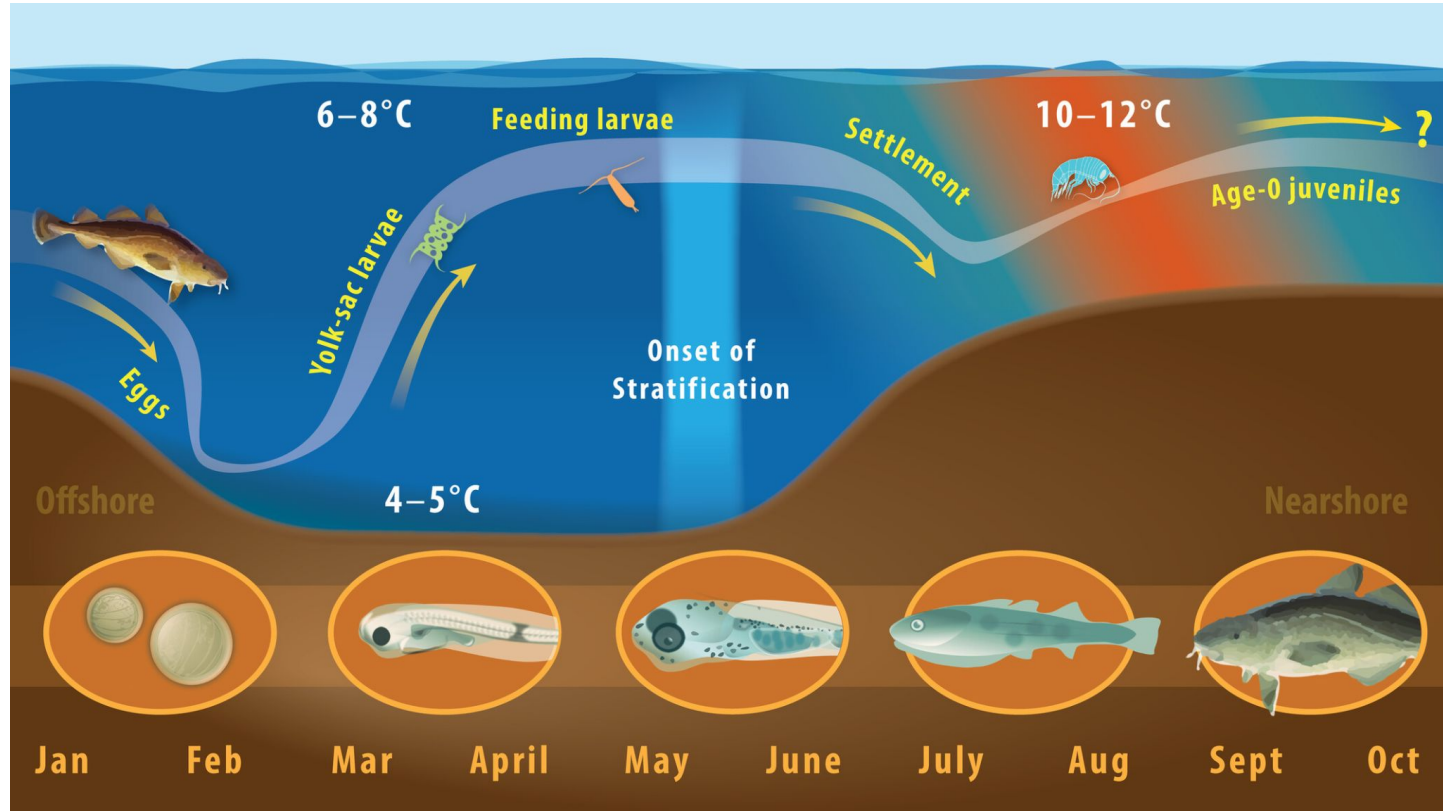
[Stock Assessment Report, 2023, Gulf of Alaska.](#)





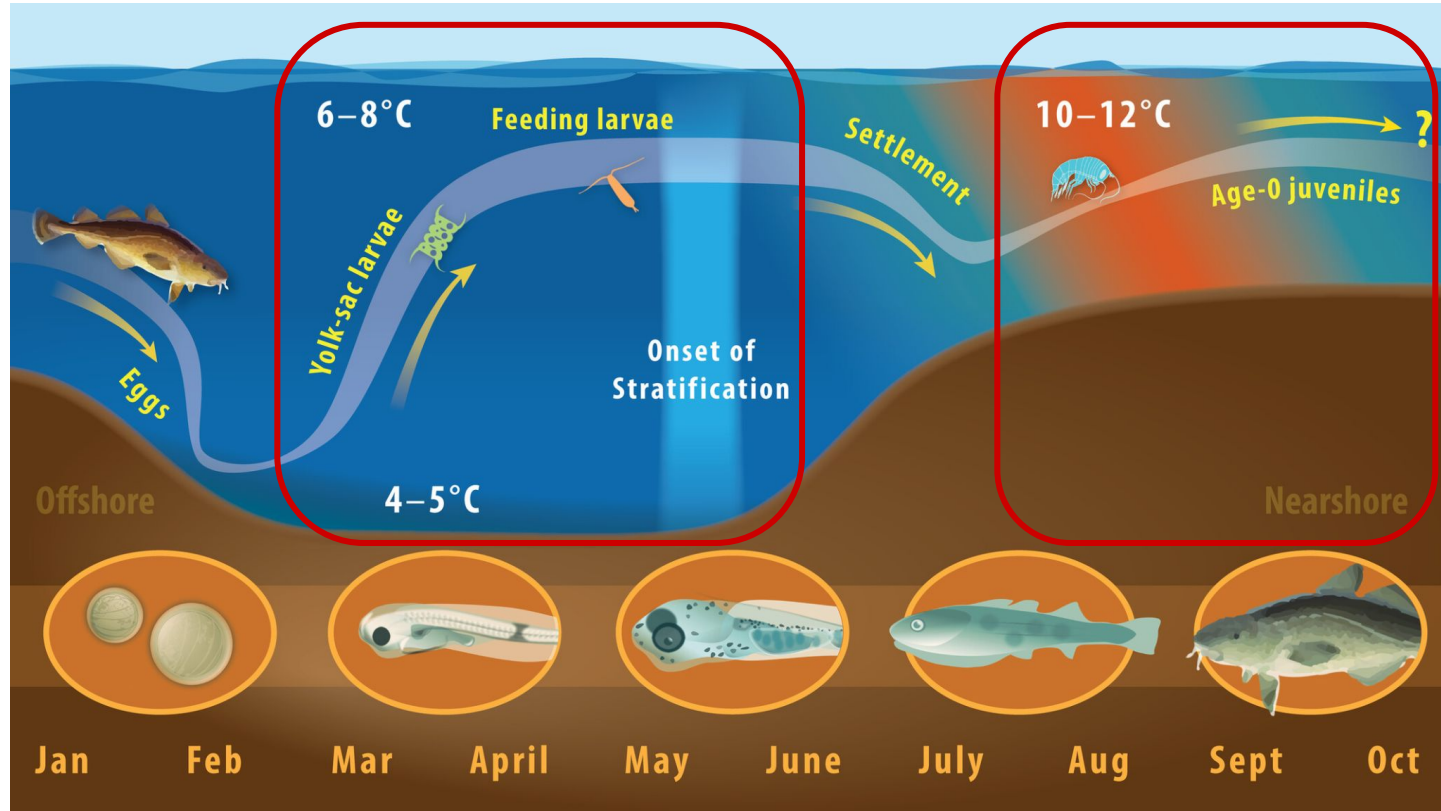
## Big Questions

- Why & how does warming affect Pacific cod recruitment?
- Do biological reference points need to be updated in stock assessment models?
- How resilient are Pacific cod populations in Alaska to warming?

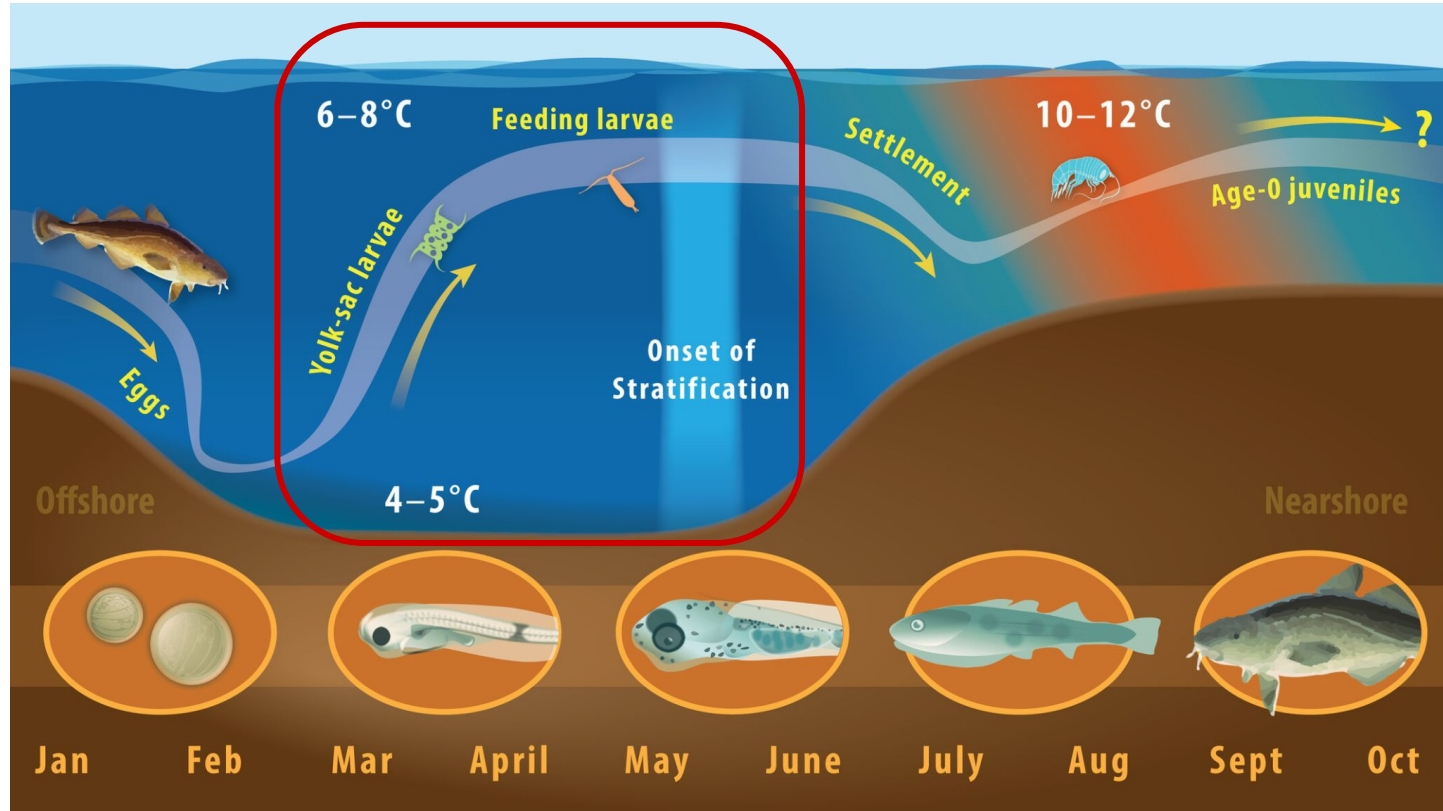


## 1. Larval study

## 2. Juvenile study



## 1. Larval study



## Experimental Team



How does temperature and acidification affect **larval** Pacific cod **survival**, **growth**, **condition**, & **energy allocation**?

## Gamete Finder



## 'Omics Team





## Experimental Team



Emily Slesinger



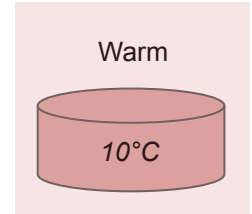
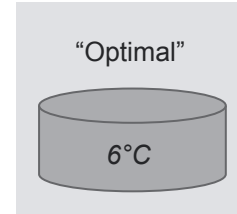
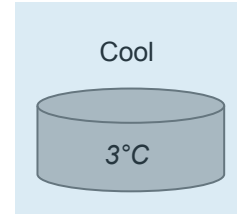
Tom Hurst



Ben Laurel

How does temperature and acidification affect **larval** Pacific cod **survival, growth, condition, & energy allocation**?

- Adults caught off Kodiak, AK to collect gametes  
1 female x 3 males
- Fertilized embryos transported to Newport, OR, reared through feeding stage in 3 temperatures



- Monitored growth & survival, 'omics samples at end
- *Acidification treatment too!*

## Gamete Finder



Alissa Abookier

## 'Omics Team



Me



Ingrid Spies



## Experimental Team



Emily Slesinger



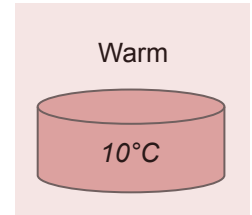
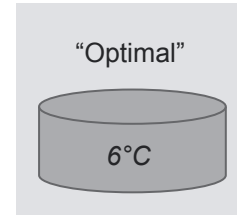
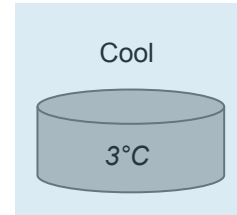
Tom Hurst



Ben Laurel

How does temperature and acidification affect **larval** Pacific cod **survival, growth, condition, & energy allocation**?

- Adults caught off Kodiak, AK to collect gametes  
1 female x 3 males
- Fertilized embryos transported to Newport, OR, reared through feeding stage in 3 temperatures



Monitored growth & survival, 'omics samples at end  
*Acidification treatment too!*

Marine Biology (2024) 171:121  
<https://doi.org/10.1007/s00227-024-04439-w>

### ORIGINAL PAPER

## The combined effects of ocean warming and ocean acidification on Pacific cod (*Gadus macrocephalus*) early life stages

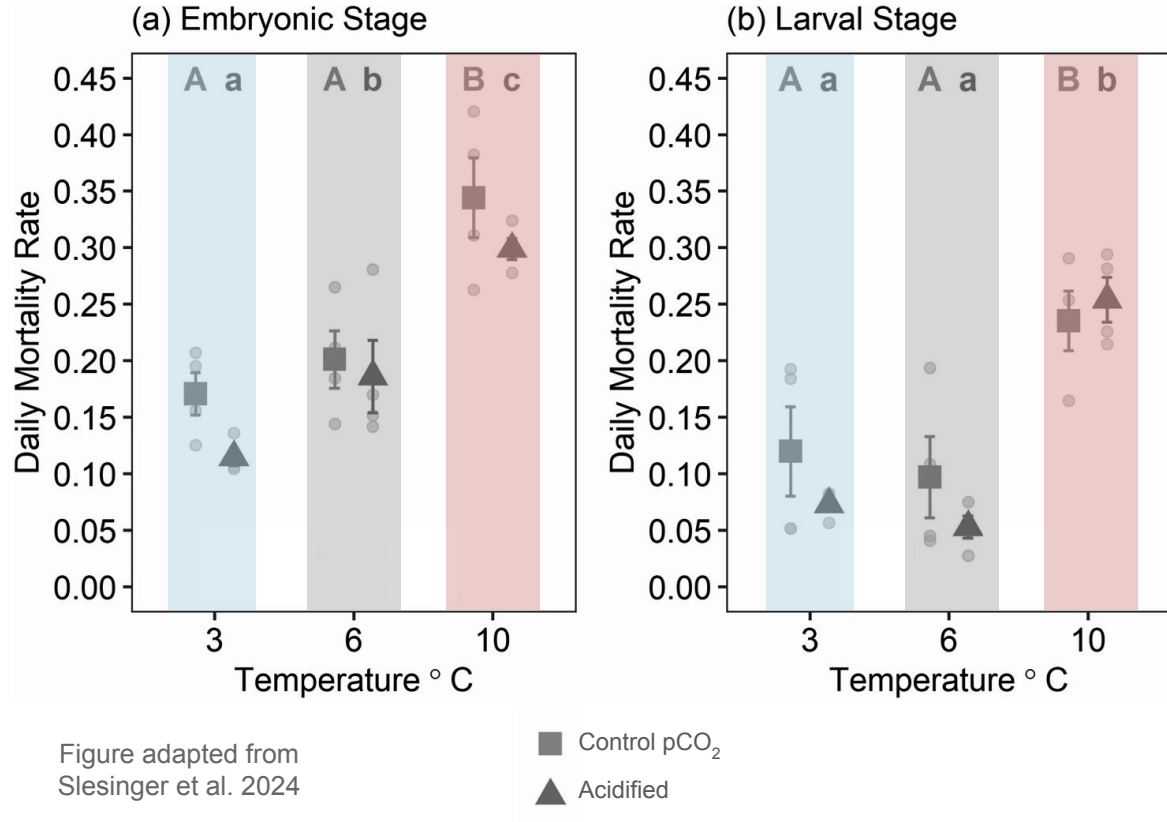
Emily Slesinger<sup>1,2</sup> · Samantha Mundorff<sup>1,3</sup> · Benjamin J. Laurel<sup>1</sup> · Thomas P. Hurst<sup>1</sup>

Received: 23 June 2023 / Accepted: 11 April 2024 / Published online: 28 April 2024

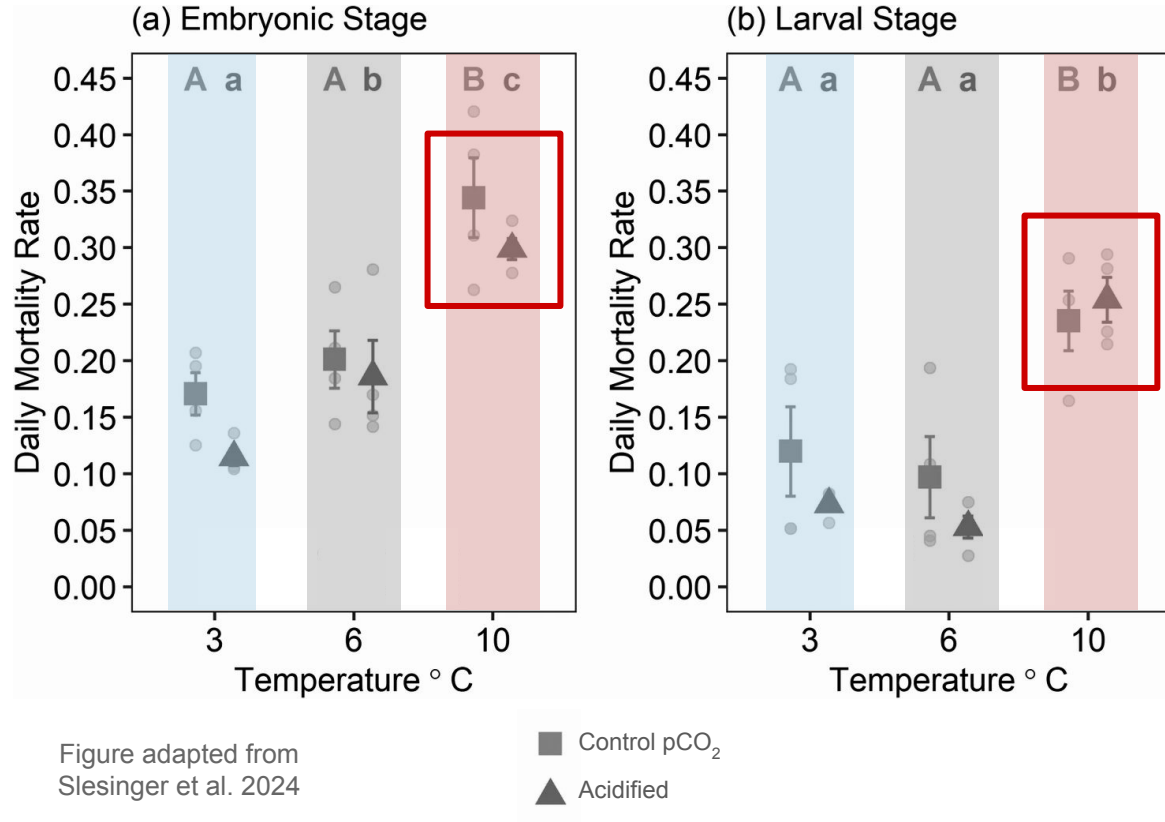
This is a U.S. Government work and not under copyright protection in the US; foreign copyright protection may apply 2024

## High larval mortality in warming

(and Warm+Acidified)



**High larval mortality in warming - Heat waves likely decreased recruitment due to low larval survival**  
(and Warm+Acidified)



**High larval mortality in warming** - *Heat waves likely decreased recruitment due to low larval survival*  
(and Warm+Acidified)

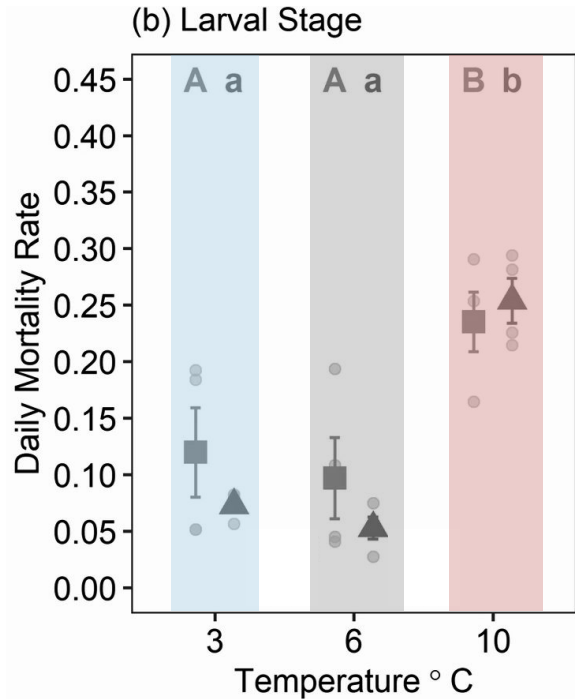


Figure adapted from  
Slesinger et al. 2024

■ Control pCO<sub>2</sub>  
▲ Acidified

But **why?**

- Gene expression analysis
- 73 larval cod individuals
- n=11-14 / treatment
- Whole-body tissue

**Goal:** *Capture energy allocation, mechanisms of mortality in moribund larvae*

## High larval mortality in warming

(and Warm+Acidified)

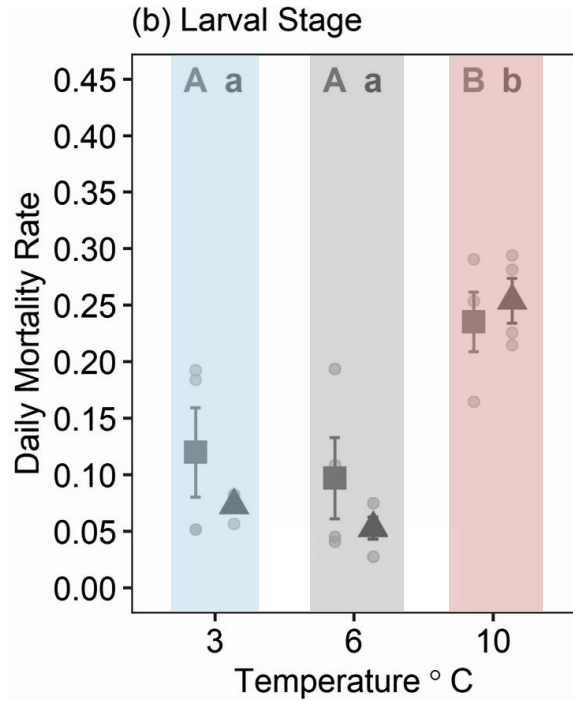
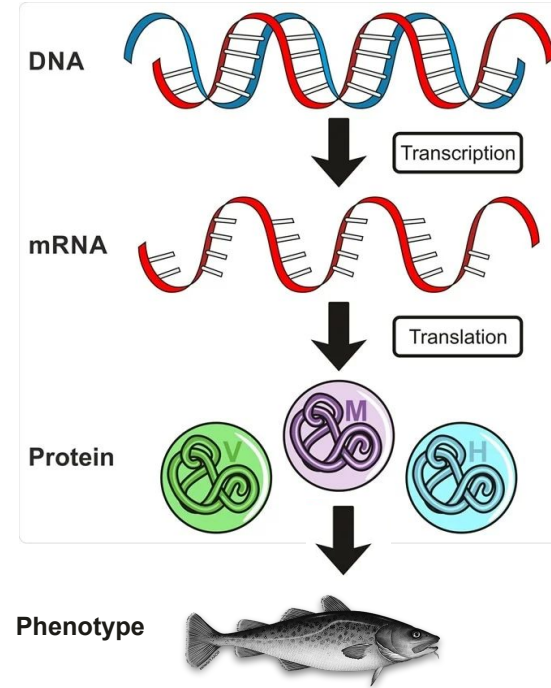


Figure adapted from  
Slesinger et al. 2024

■ Control pCO<sub>2</sub>  
▲ Acidified



Adapted from: udaix/Shutterstock.com

## High larval mortality in warming

(and Warm+Acidified)

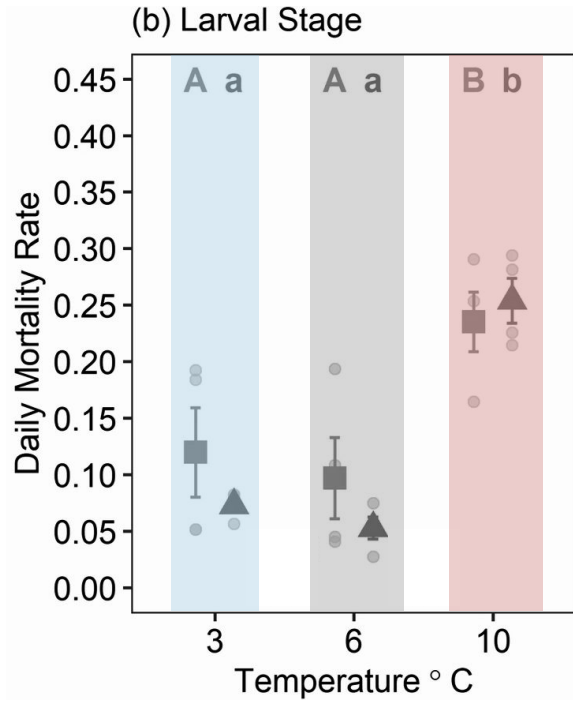
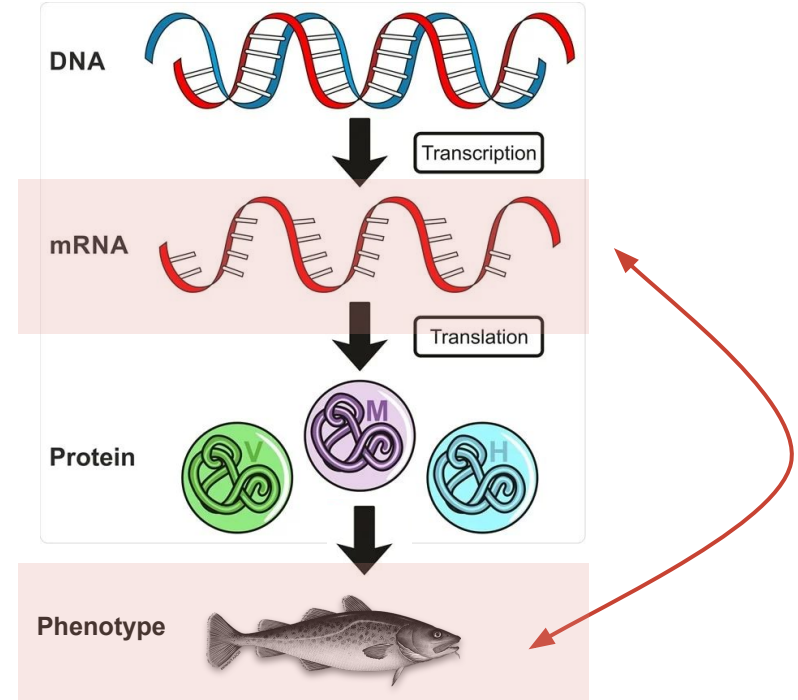


Figure adapted from  
Slesinger et al. 2024

■ Control pCO<sub>2</sub>  
▲ Acidified



Adapted from: udaix/Shutterstock.com



**Hypothesis: high mortality in warming** is due to energetic limitations caused by lipid depletion paired with energy-demanding processes (inflammation, cell signaling / stability)

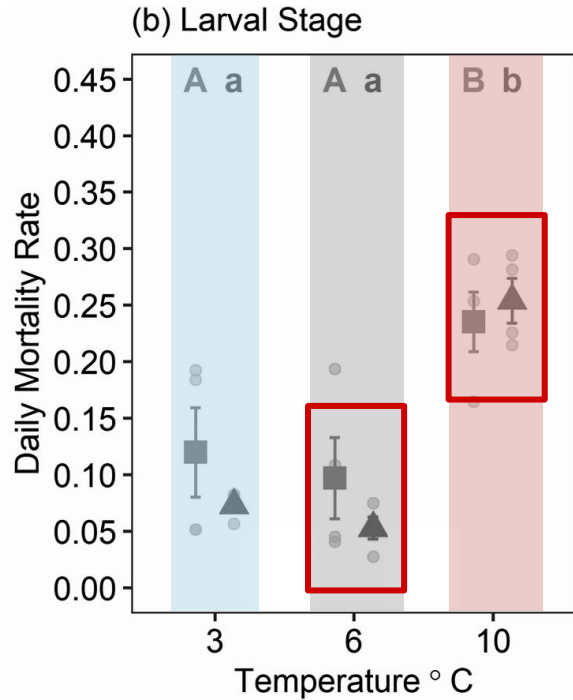
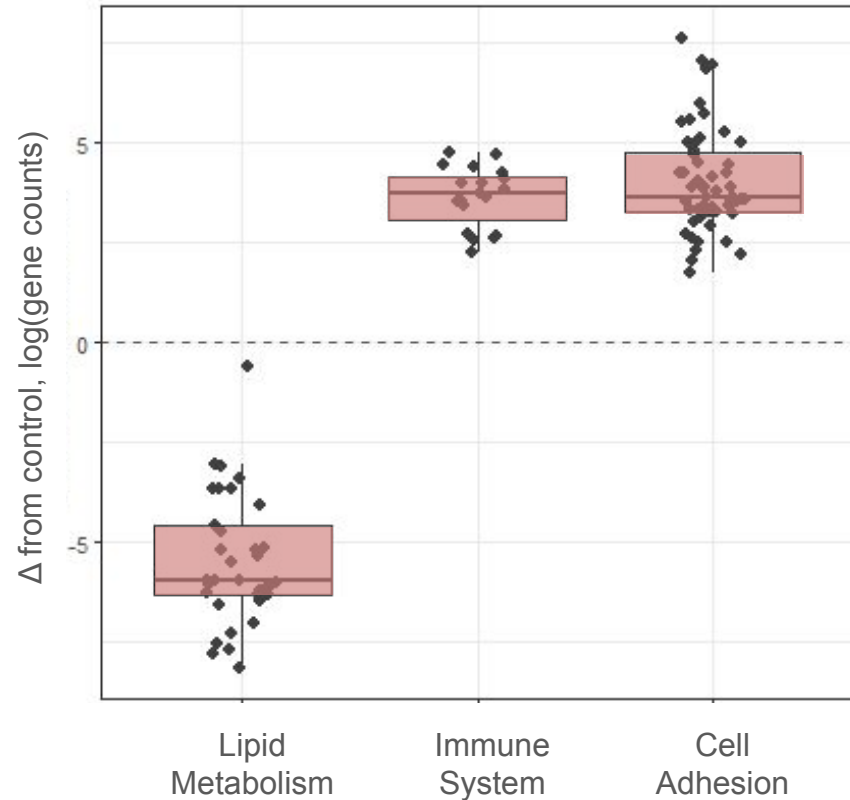


Figure adapted from  
Slesinger et al. 2024

■ Control pCO<sub>2</sub>  
▲ Acidified



**Hypothesis: high mortality in warming** is due to energetic limitations caused by lipid depletion paired with energy-demanding processes (inflammation, cell signaling / stability)

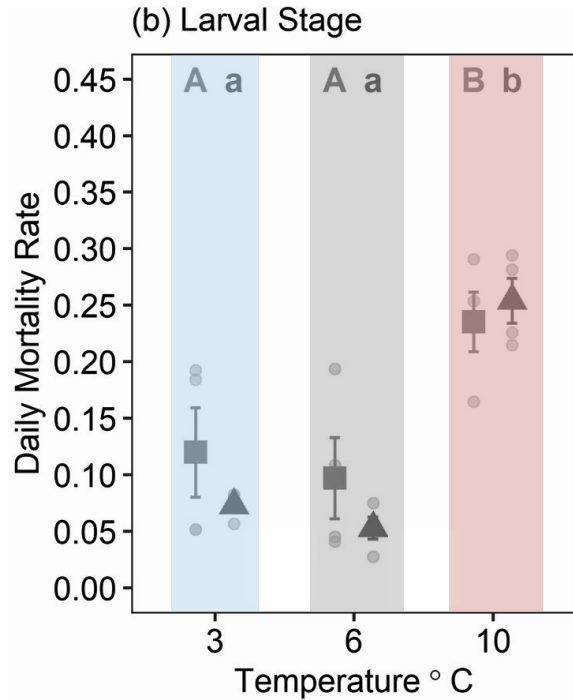
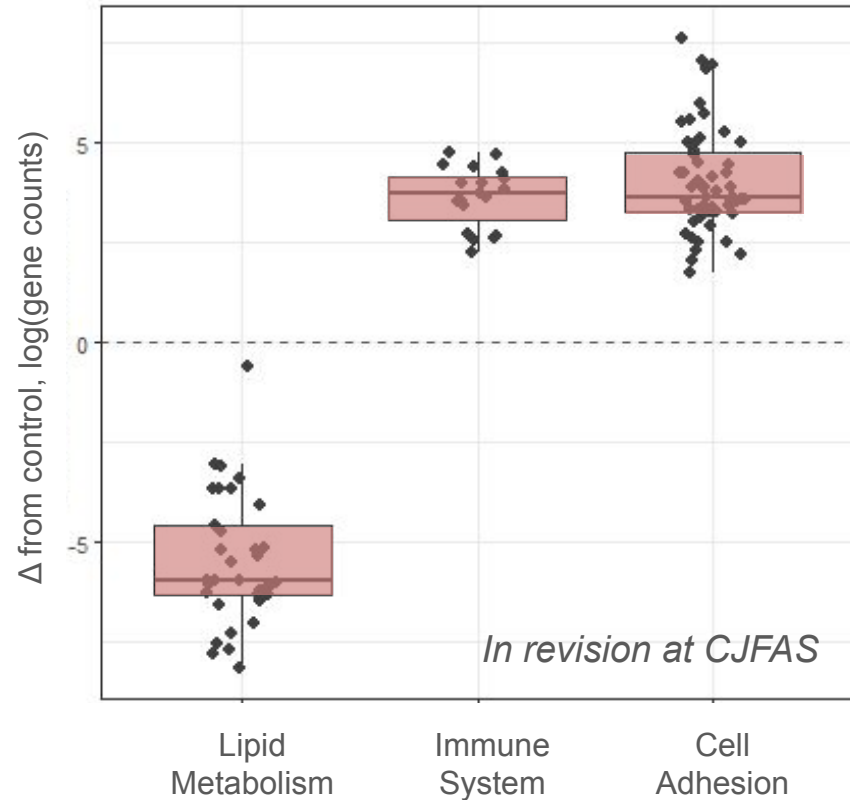


Figure adapted from  
Slesinger et al. 2024

■ Control pCO<sub>2</sub>  
▲ Acidified





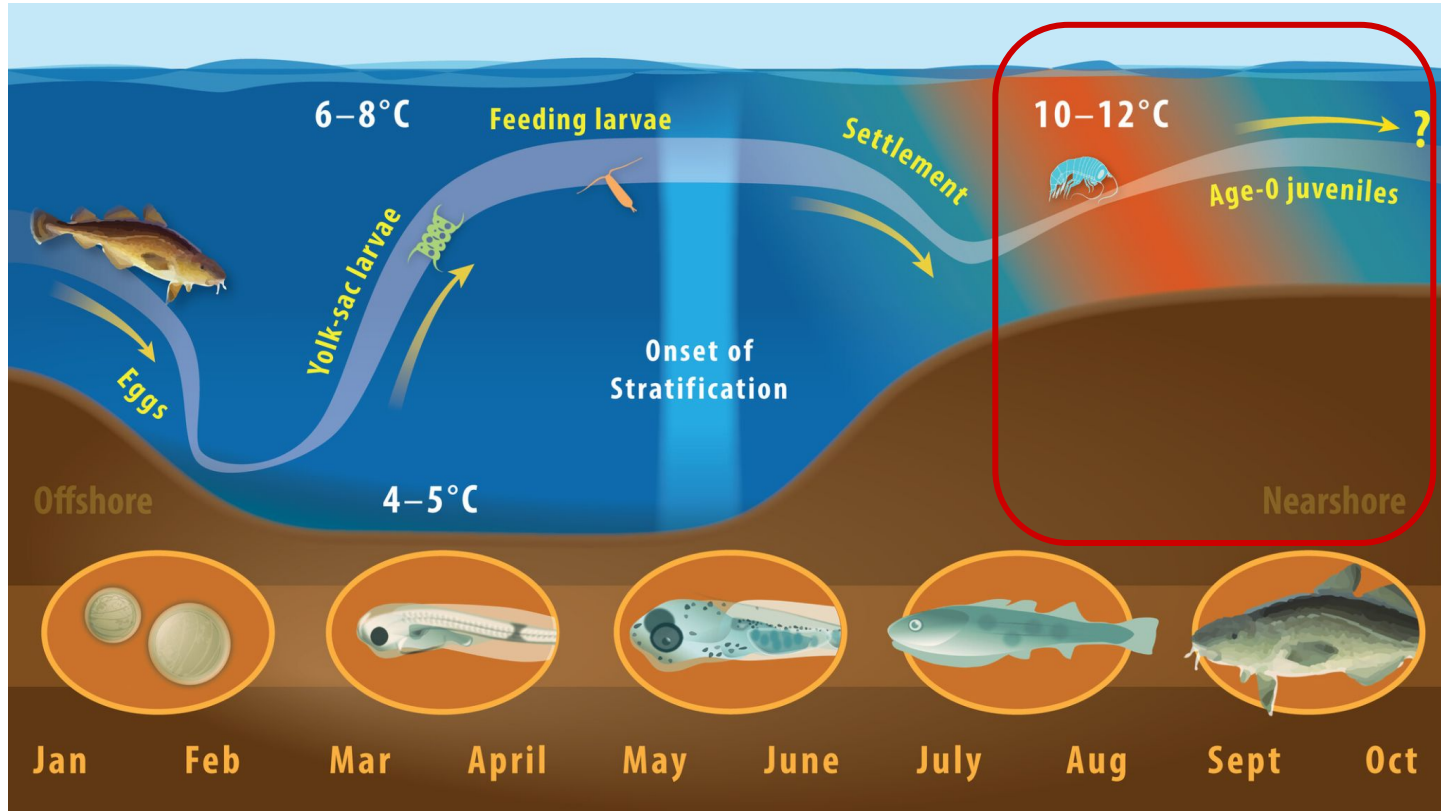
**Warming increases larval mortality rates**, which likely was a factor influencing recruitment during heatwave years.

*Slesinger et al. 2024, Mar. Bio.*

**Mechanisms of larval mortality** in warming could reflect energetic limitations paired with energy-demanding inflammation and cellular instability.

*Spencer et al. In Revision, CJFAS*

## 2. Juvenile study



## Experimental Team



Ben Laurel, AFSC



Mary Beth Rew Hicks, AFSC

How does temperature affect **juvenile** Pacific cod **growth, survival, & energy allocation** in their first fall/winter?



## Lipid Team



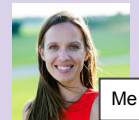
Michelle Stowell  
OSU

Samantha  
Mundorff

Louise Copeman  
AFSC / OSU

Carlissa  
Salant

## 'Omics Team



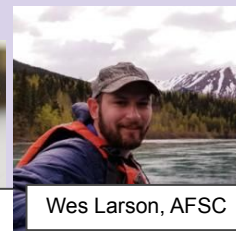
Me



Steven Roberts, UW



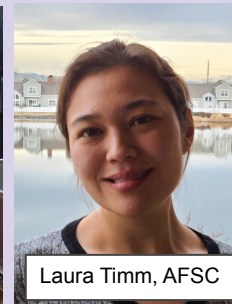
Sara Schaal, AFSC & UO



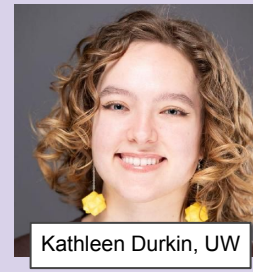
Wes Larson, AFSC



Ingrid Spies, AFSC



Laura Timm, AFSC



Kathleen Durkin, UW



## Juvenile temperature experiment



- Wild juveniles (age-0) caught off Kodiak, AK late summer
- Transported to Newport, OR wet lab
- Acclimated
- ~6 weeks experiment

n=40 / temp.

Very Cold

0°C

Cool

5°C

Warm

9°C

Very Warm

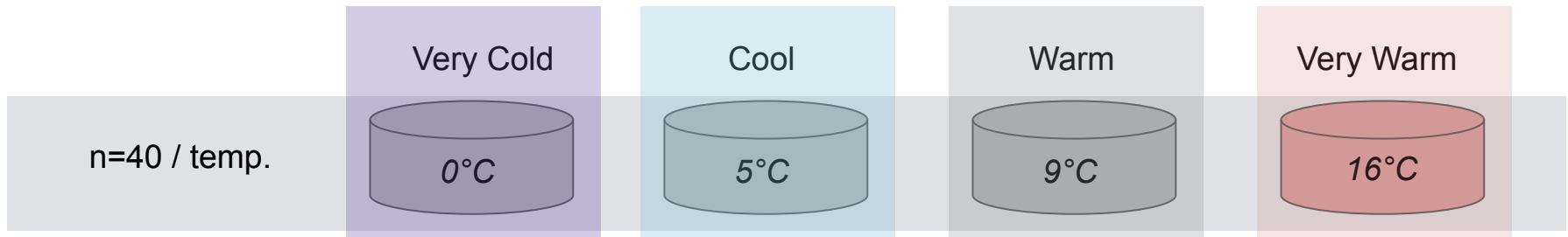
16°C



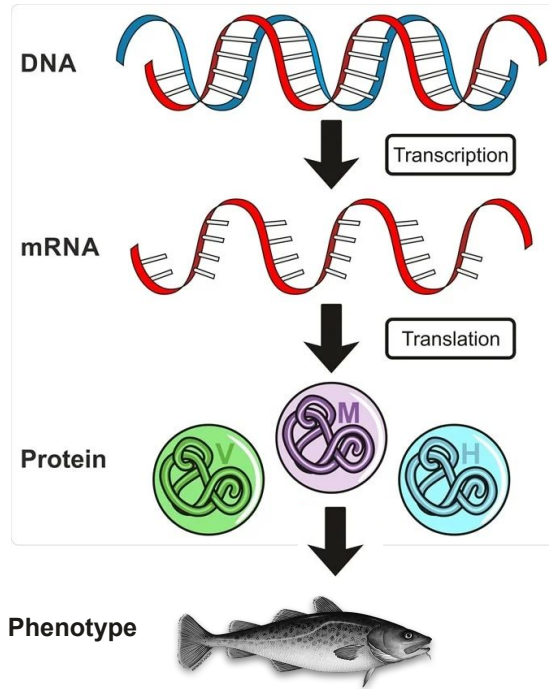


*Individuals tagged, collected:*

- a. Genetics with fin clips, n=40/temp (IcWGS)
- b. Growth rates (length & wet weight) during acclimation, treatment
- c. Body condition (Kwet)
- d. Liver condition (HSI)
- e. Survival
- f. Liver lipid components (n=25/temp)
- g. Gene expression with liver, n=18/temp (RNASeq)

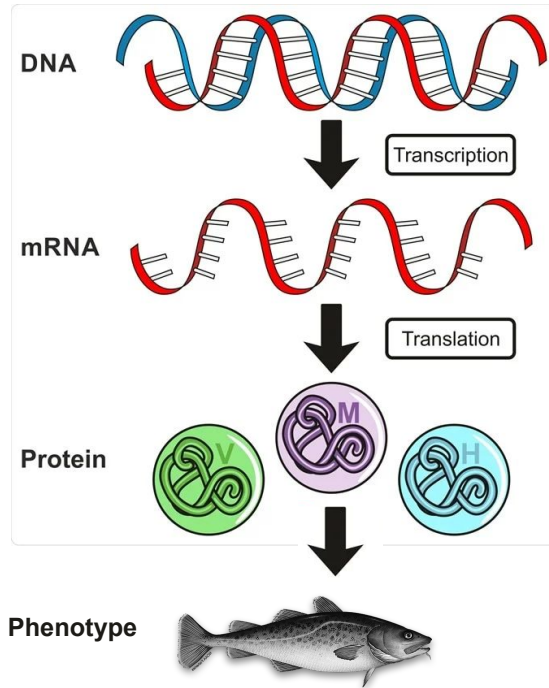


# *“Genome-to-Phenome” dataset, Pacific cod juvenile temperature response*



Adapted from: [udaix/Shutterstock.com](https://www.shutterstock.com/user/udaix)

# *“Genome-to-Phenome” dataset, Pacific cod juvenile temperature response*

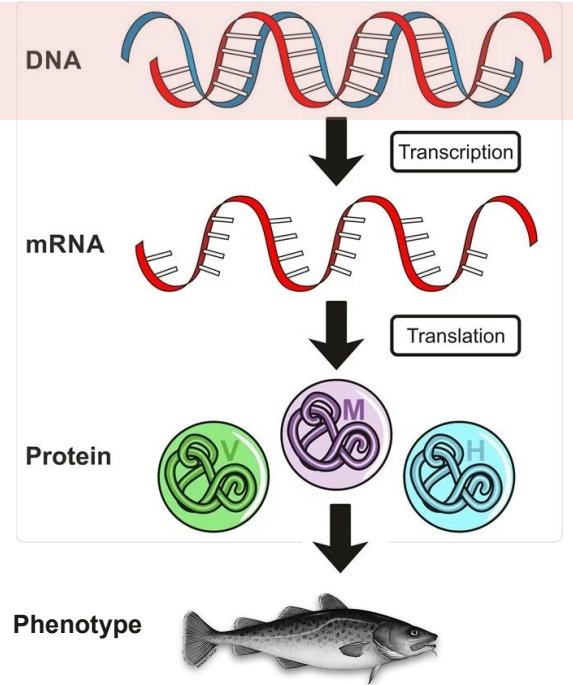


1. Genetics –  
**Who are they?**
2. Phenotypes –  
**How does warming affect key traits?**
3. Integrate datasets –  
**Why are some fish less sensitive?**

Adapted from: [udaix/Shutterstock.com](https://www.shutterstock.com/user/udaix)

# *“Genome-to-Phenome” dataset, Pacific cod juvenile temperature response*

1. Genetics – Who are they (wild caught)?



Adapted from: udaix/Shutterstock.com

# *“Genome-to-Phenome” dataset, Pacific cod juvenile temperature response*

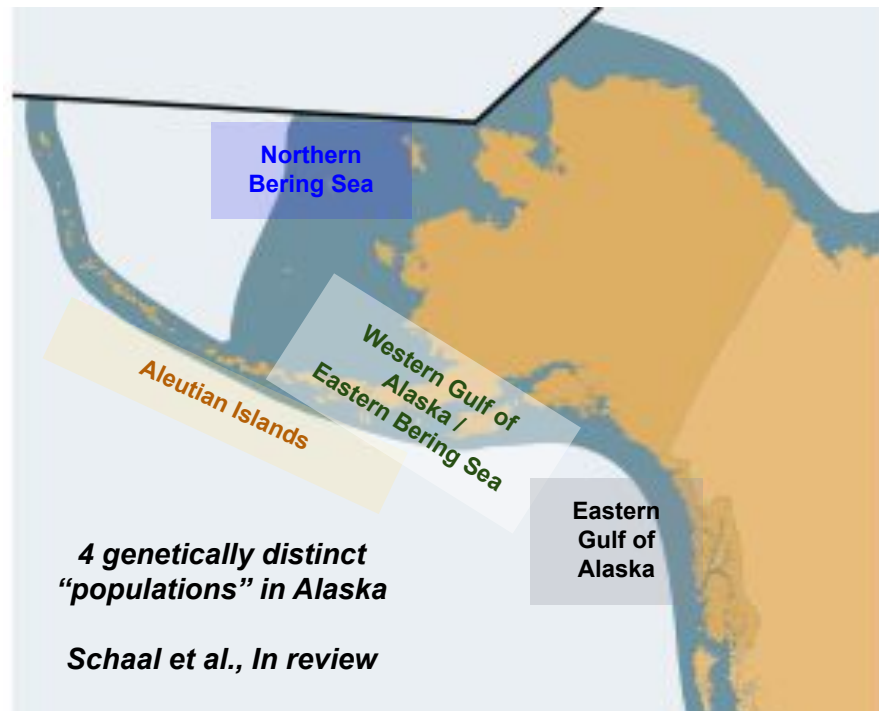
## 1. Genetics – Who are they (wild caught)?

DNA



### Tools used:

- Sequence whole genome ~3x (i.e. “low-coverage”), n=160
- High-quality reference genome for alignment & genotype probability data



# What does genotype data look like?

Genome location

Chromosome	Site	Sample 1	Sample 2
<b>Chr1</b>	<b>10,000,065</b>	<b>A/A</b>	<b>G/A</b>
Chr1	10,000,883	C/C	C/T
Chr1	10,001,961	G/G	A/T
Chr1	10,002,133	C/C	C/T
Chr1	10,002,294	G/A	G/A

Per-sample genotype

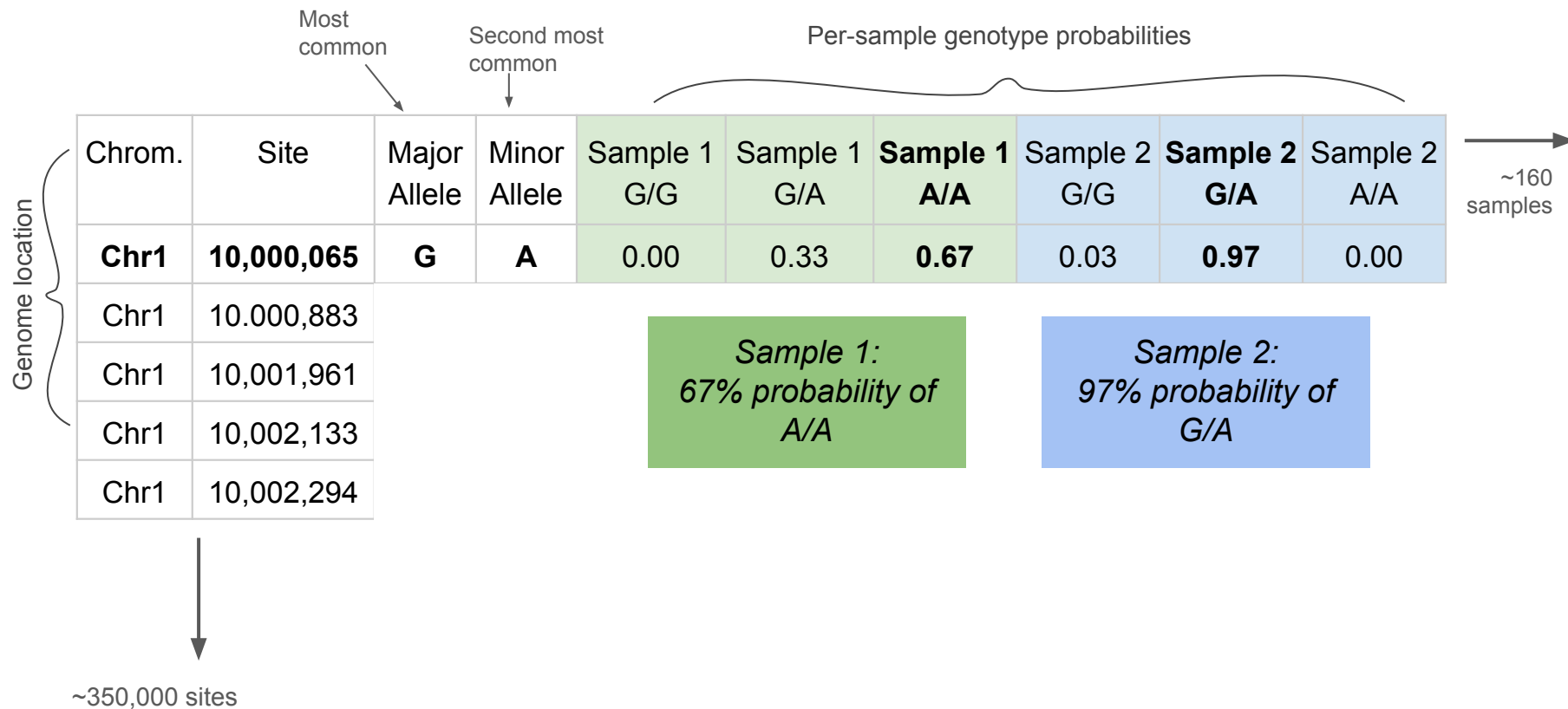
~160 samples

~350,000 sites

Possible alleles: A,G,T,C  
1 from each parent



# What does *genotype probability* data look like?

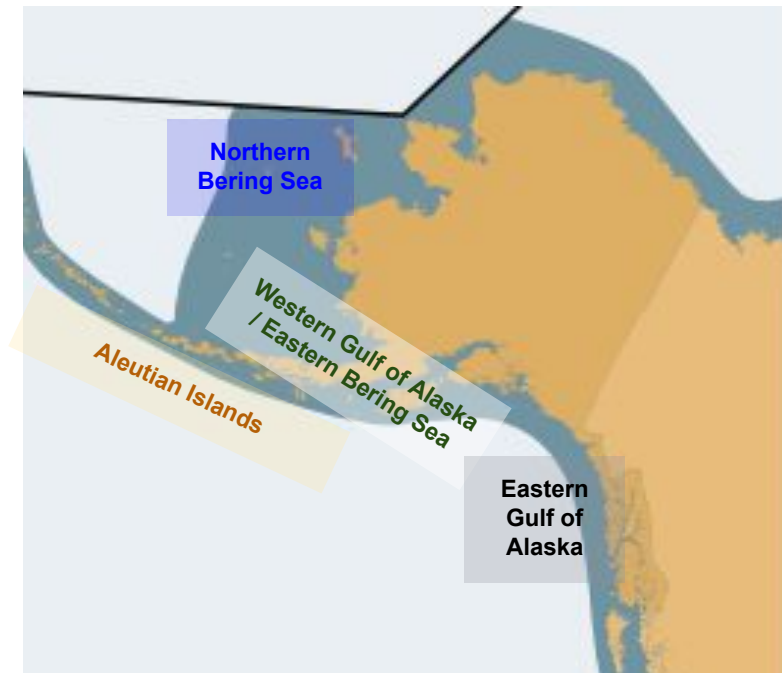


## Use genotype probability data to **predict population of origin**

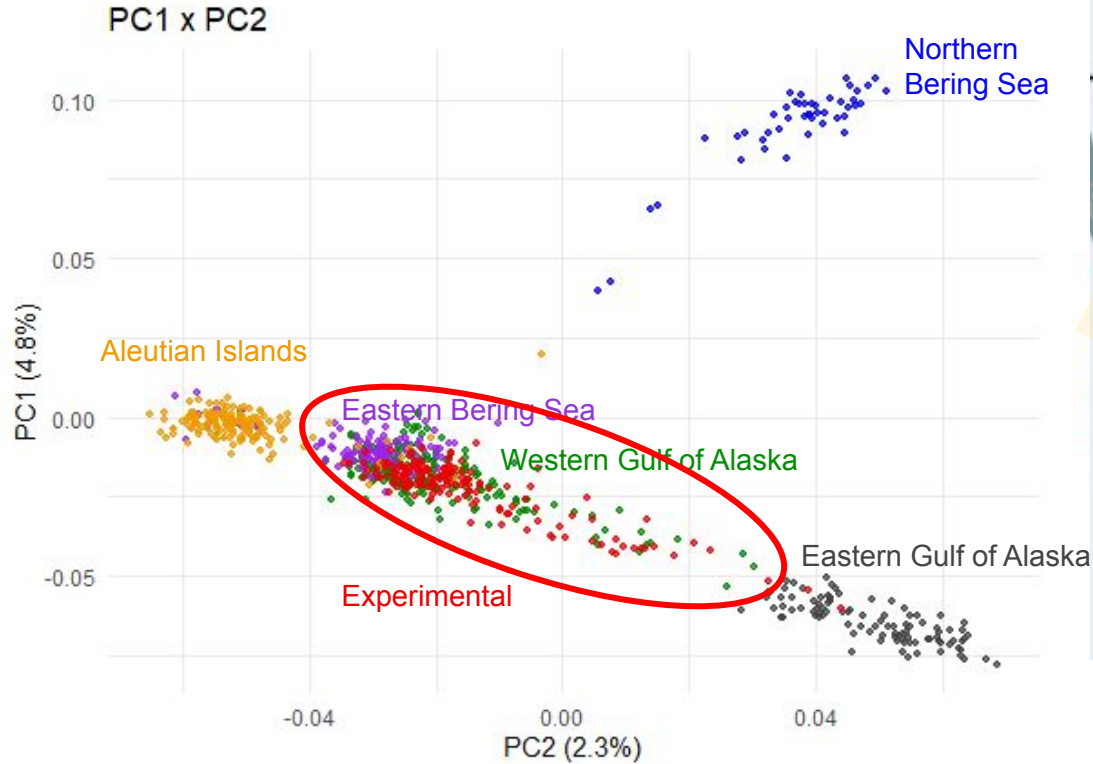
Used genotype probabilities from:

- 160 experimental fish
- More data: ~55 fish per population (Schaal et al. *In review*), “reference fish”

1. Identified sites associated with population differences (top  $F_{st}$ )
2. Identified best sites ( $n=6,101$ ) that predict population, ~96% assignment accuracy in reference fish
3. **Predict population of origin for experimental fish** (`wgsassign`)

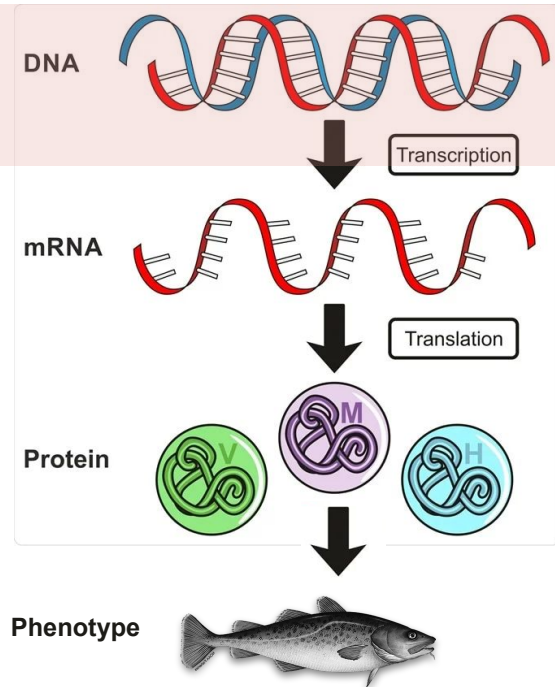


*Predicted population of origin = Western GOA / Eastern Bering Sea group*



## *“Genome-to-Phenome” dataset, Pacific cod juvenile temperature response*

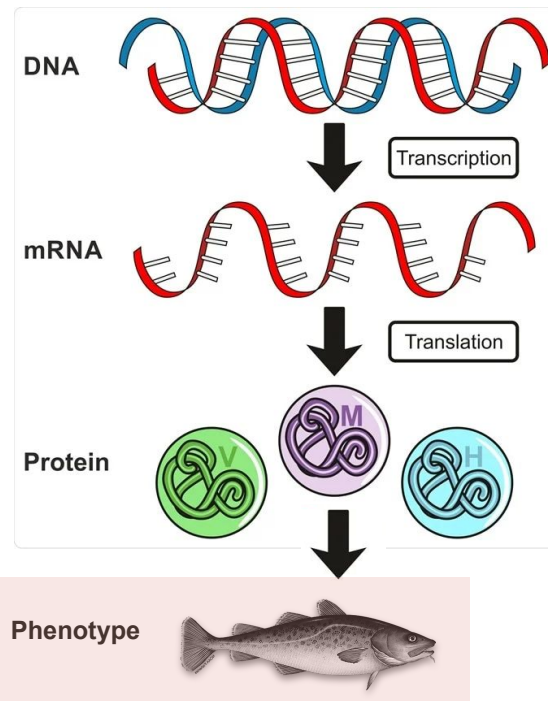
- ✓ Genetics – **they are one population**  
western Gulf of Alaska / Eastern Bering Sea



Adapted from: udaix/Shutterstock.com

## *“Genome-to-Phenome” dataset, Pacific cod juvenile temperature response*

- ✓ Genetics – **they are one population**  
western Gulf of Alaska / Eastern Bering Sea
- Phenotypes – how are survival-associated traits affected?



Adapted from: udaix/Shutterstock.com

# *Phenotypes - growth rate, liver condition, lipid content*

Growth Rate (weight)

*Faster growth* 👍

Hepatosomatic index (liver size)

*Larger liver* 👍

Total Lipid Content in liver

*More lipid* 👍



# *Phenotypes - growth rate, liver condition, lipid content*

Growth Rate (weight)

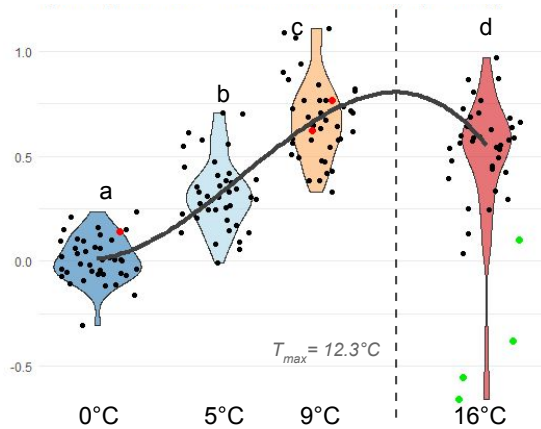
*Faster growth* 👍

Hepatosomatic index (liver size)

*Larger liver* 👍

Total Lipid Content in liver

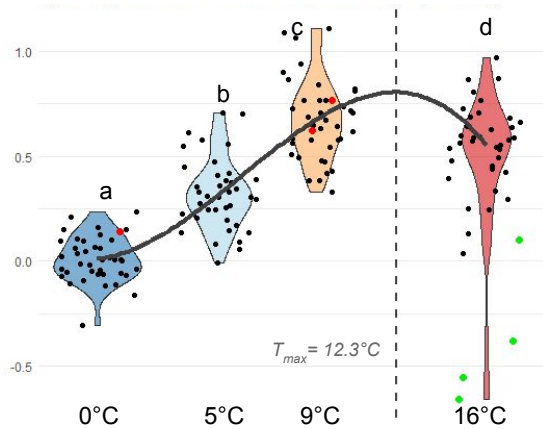
*More lipid* 👍



# Phenotypes - growth rate, liver condition, lipid content

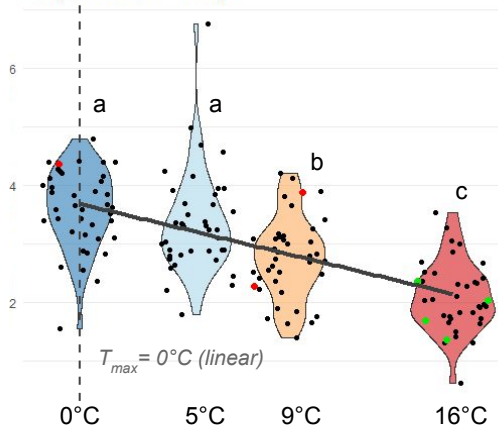
## Growth Rate (weight)

Faster growth 👍



## Hepatosomatic index (liver size)

Larger liver 👍



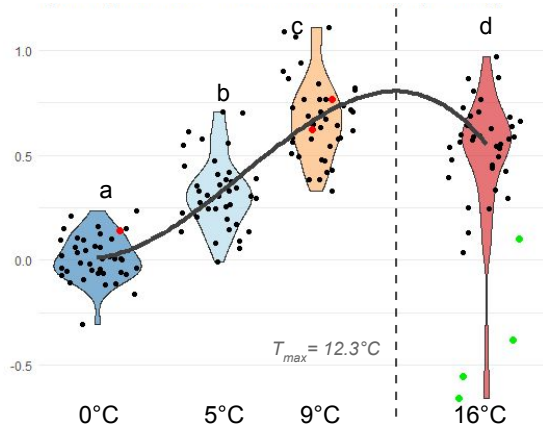
## Total Lipid Content in liver

More lipid 👍

# Phenotypes - growth rate, liver condition, lipid content

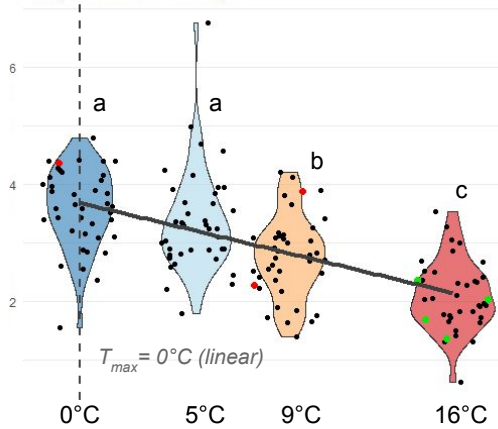
## Growth Rate (weight)

Faster growth 👍



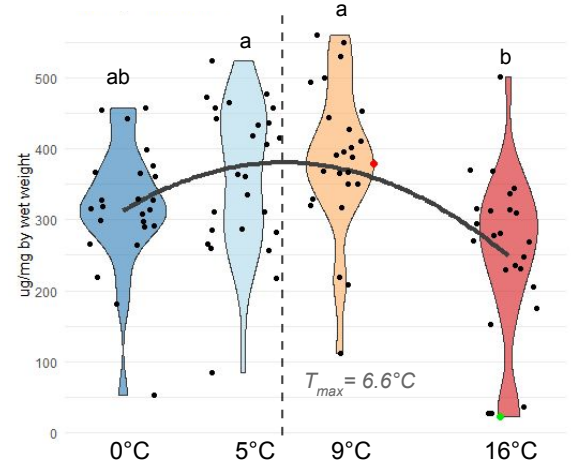
## Hepatosomatic index (liver size)

Larger liver 👍



## Total Lipid Content in liver

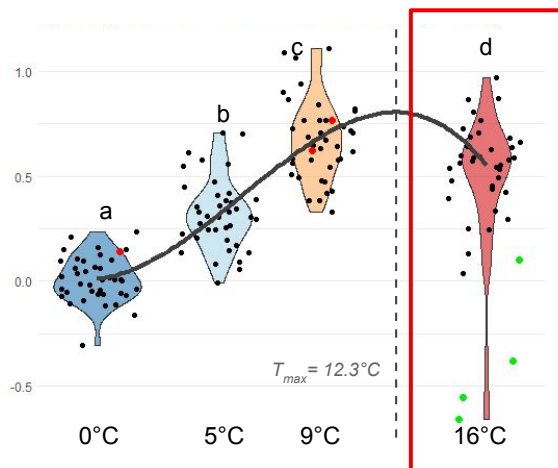
More lipid 👍



# Warming decreased lipid reserves

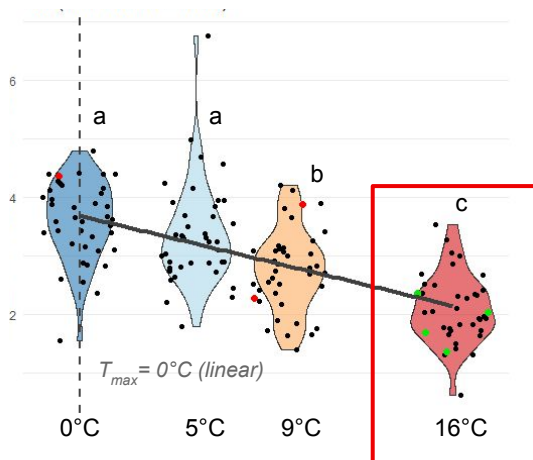
Growth Rate (weight)

Faster growth 👍



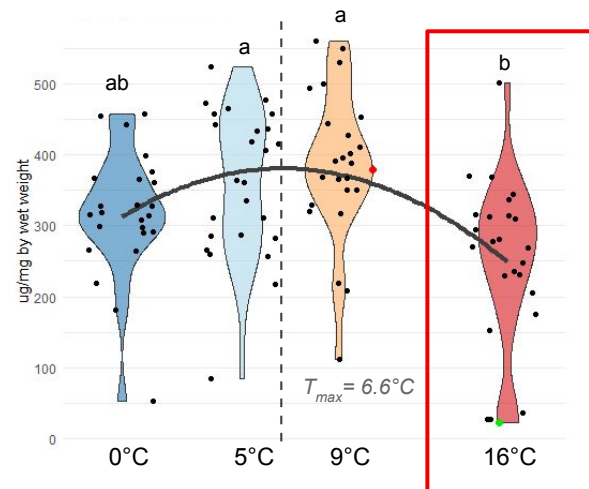
Hepatosomatic index (liver size)

Larger liver 👍



Total Lipid Content in liver

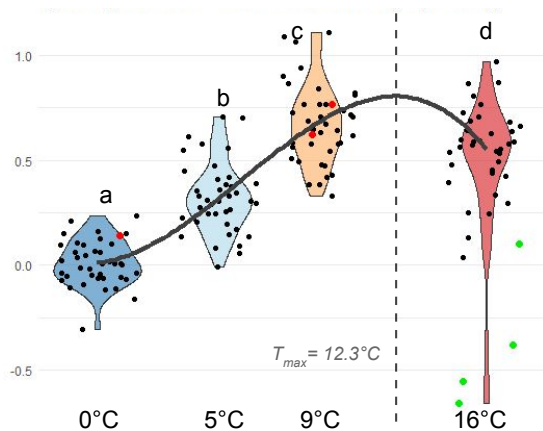
More lipid 👍



# Warming decreased lipid reserves

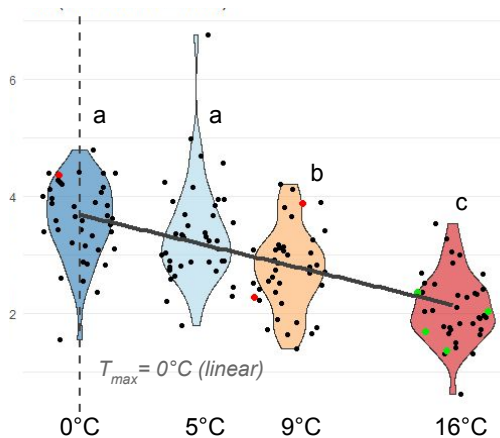
## Growth Rate (weight)

Faster growth 👍



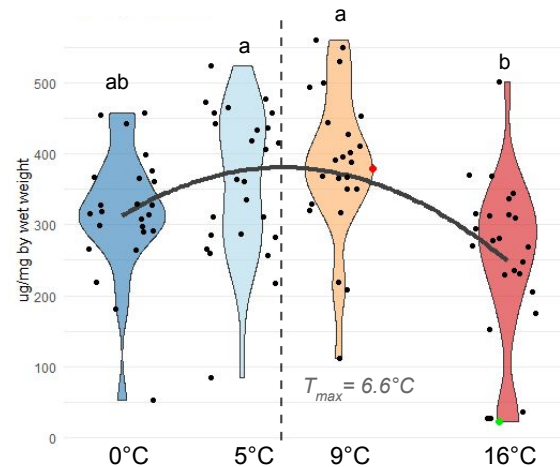
## Hepatosomatic index (liver size)

Larger liver 👍



## Total Lipid Content in liver

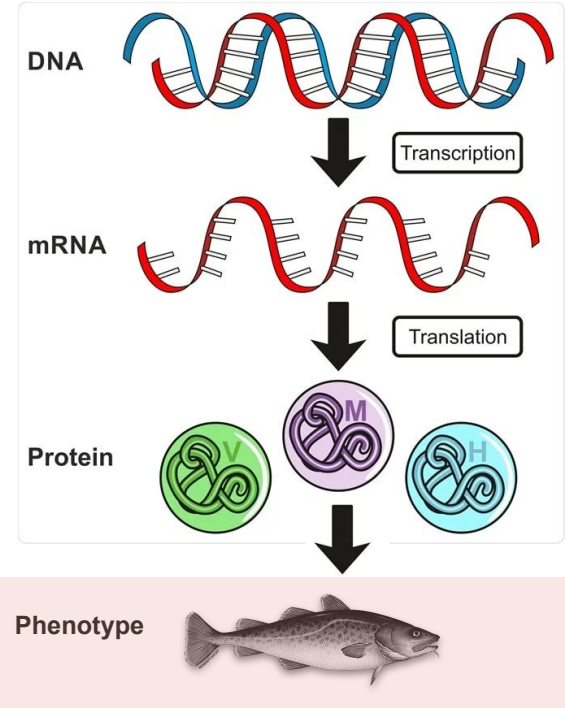
More lipid 👍



- Survived
- Died

# “Genome-to-Phenome” dataset for juvenile Pacific cod

- ✓ Genetics – **they are one population**  
western Gulf of Alaska / Eastern Bering Sea
- ✓ Phenotypes – **Fewer lipid reserves in warming, slightly slower growth** - juvenile overwinter survival likely lower during heatwave years.

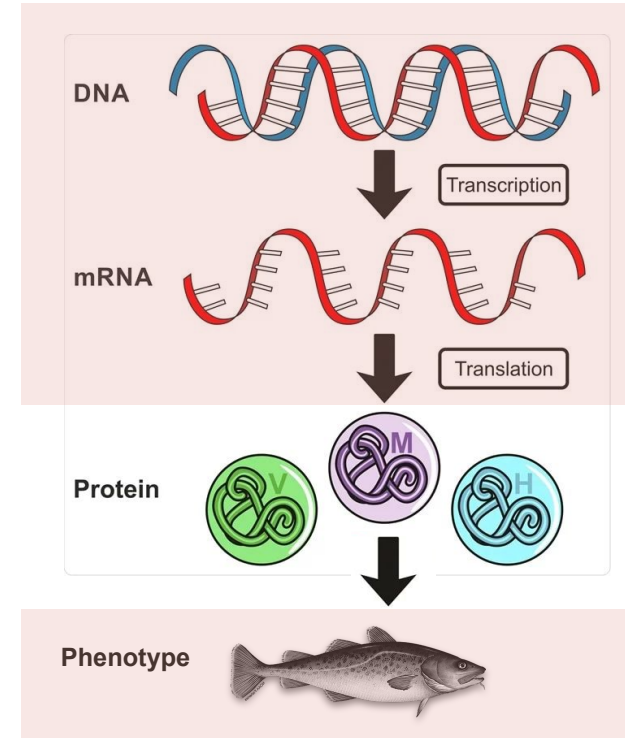


Adapted from: udaix/Shutterstock.com



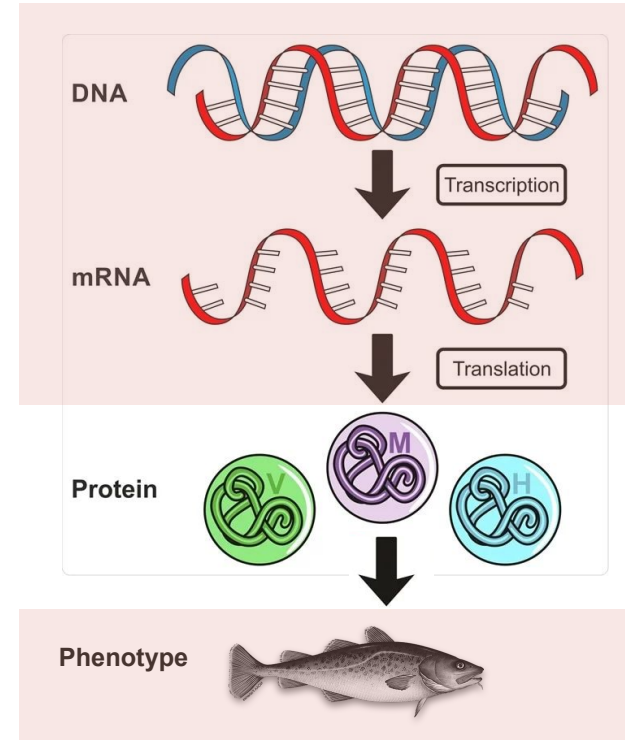
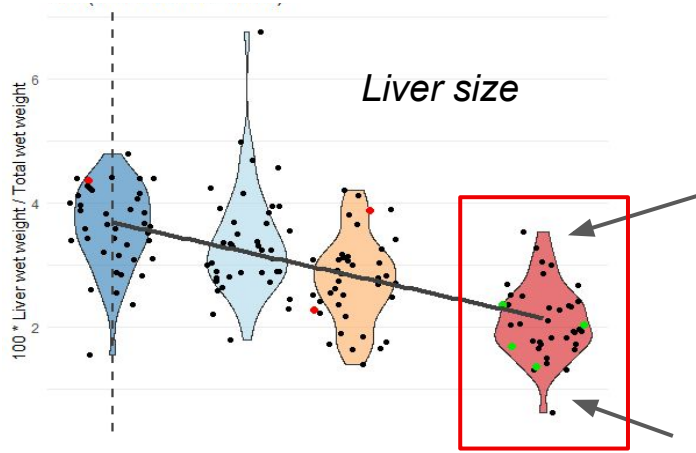
# “Genome-to-Phenome” dataset for juvenile Pacific cod

- ✓ Genetics – **they are one population**  
western Gulf of Alaska / Eastern Bering Sea
  - ✓ Phenotypes – **Fewer lipid reserves in warming, slightly slower growth** - juvenile overwinter survival likely lower during heatwave years.
- Integrate datasets – Performance indicators!
    - a. Genetic variants
    - b. Expression patterns



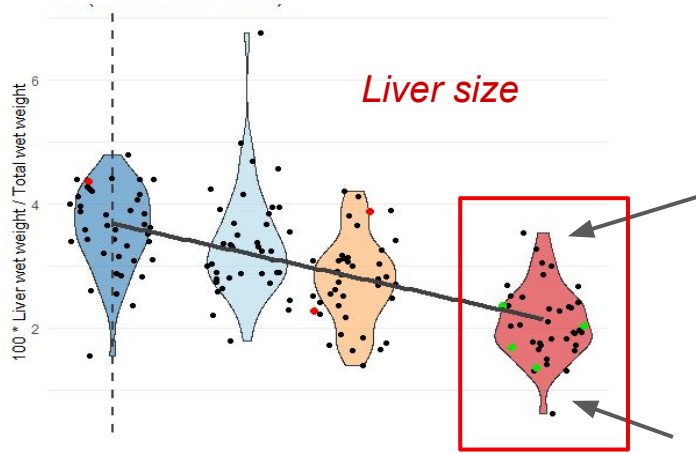
Adapted from: udaix/Shutterstock.com

## *Variation within each temperature - opportunity!*



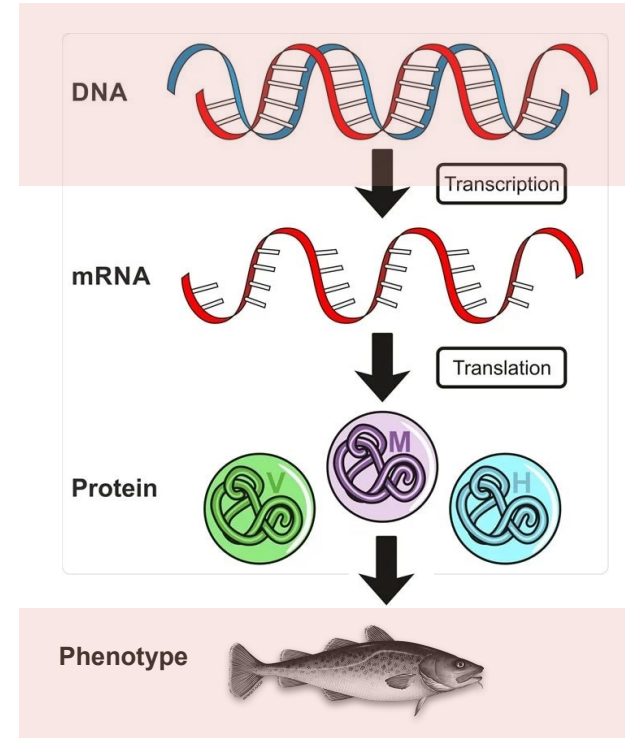
Adapted from: [udaix/Shutterstock.com](https://www.shutterstock.com)

## Variation within each temperature - opportunity!



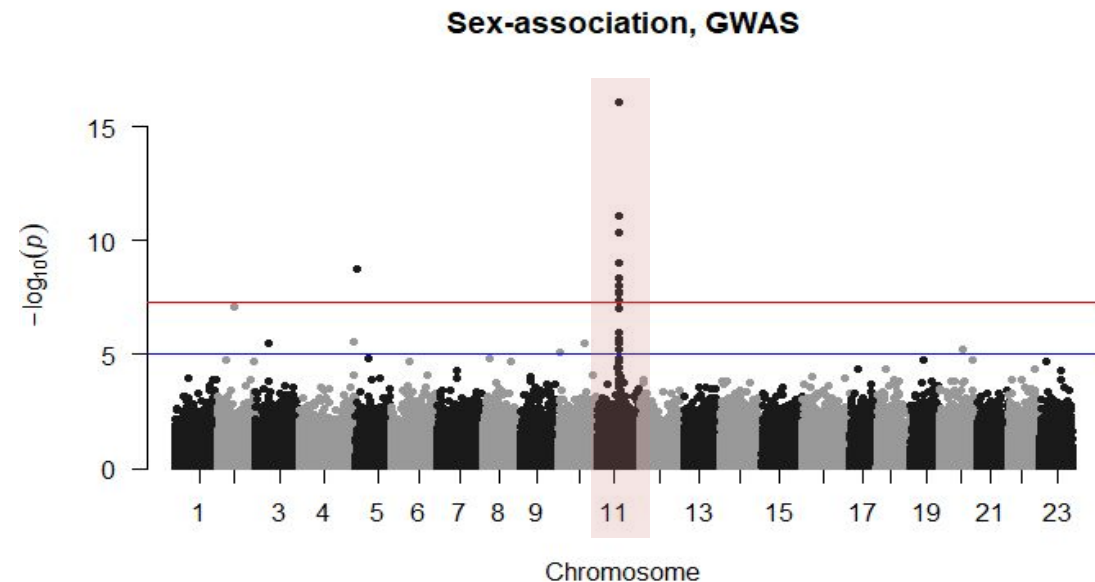
Identified **sites on genome** associated with **liver size**, lipid content, & growth in warming

*Genome-Wide Association Studies (GWAS) within each treatment*



Adapted from: udaix/Shutterstock.com

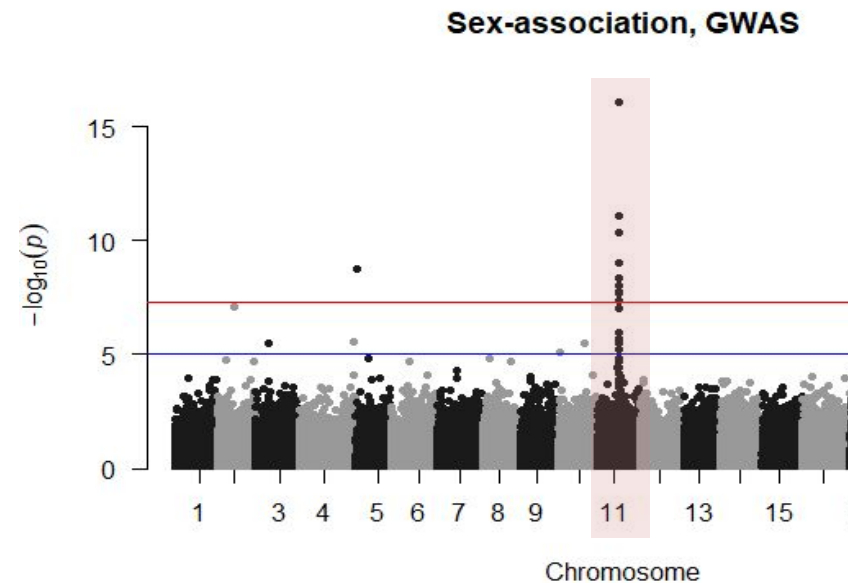
## Tangent – using GWAS to identify sex markers (preliminary)



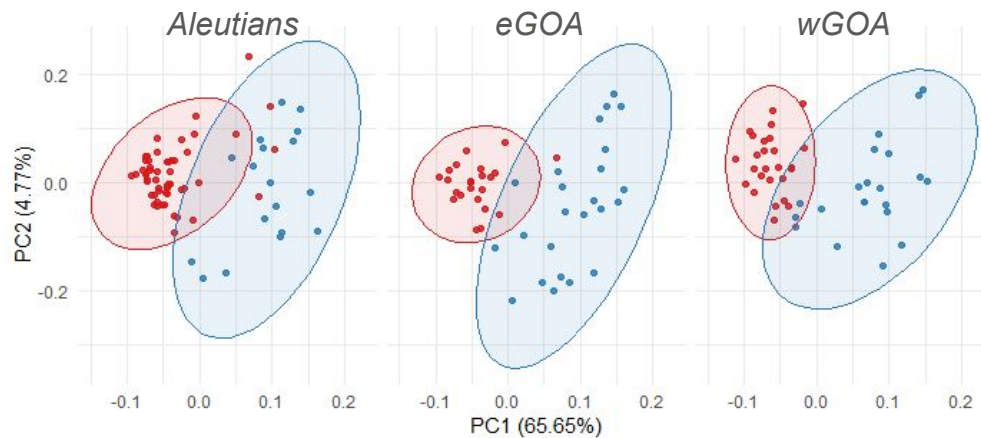
Leveraged genetic data from  
~60 females & ~100 males

*Data from Schaal et al. In  
review*

## Tangent – using GWAS to identify sex markers (preliminary)



*PCAs with ~50 sex-associated sites*



Female  
Male

=

## Tangent – using GWAS to identify sex markers (preliminary)

*Sex marker in same Chromosome  
11 region in Atlantic cod*



**scientific reports**

Explore content ▾ About the journal ▾ Publish with us ▾

[nature](#) > [scientific reports](#) > [articles](#) > article

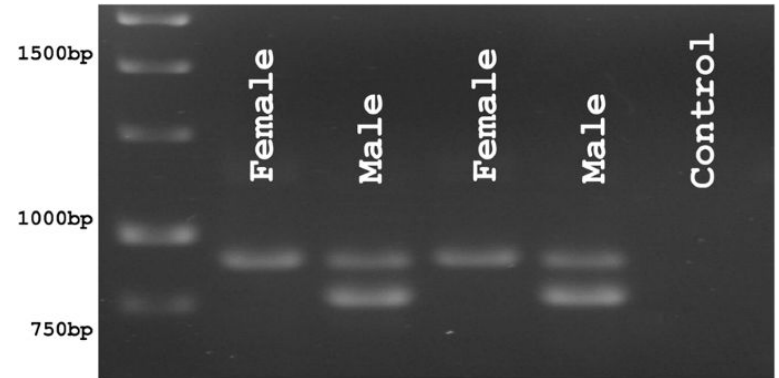
Article | [Open access](#) | Published: 15 January 2019

### Characterization of a male specific region containing a candidate sex determining gene in Atlantic cod

[Tina Graceline Kirubakaran](#), [Øivind Andersen](#), [Maria Cristina De Rosa](#), [Terese Andersstuen](#), [Kristina Hallan](#), [Matthew Peter Kent](#)  & [Sigbjørn Lien](#) 

[Scientific Reports](#) **9**, Article number: 116 (2019) | [Cite this article](#)

### *Benchmark sex assay in Atlantic cod*





# Tangent – using GWAS to identify sex markers (preliminary)



American  
Genetic  
Association

*Journal of Heredity*, 2018, 326–332

doi:10.1093/jhered/esx102

Brief communication

Advance Access publication 9 November 2017

Brief communication

## Identification of Genomic Regions Associated With Sex in Pacific Halibut

Daniel P. Drinan, Timothy Loher, and Lorenz Hauser

From the University of Washington, School of Aquatic and Fishery Sciences, Seattle, Washington (Drinan); International Pacific Halibut Commission, Seattle, Washington (Loher); University of Washington, School of Aquatic and Fishery Sciences, Seattle, Washington (Hauser).

*Genetic markers add sex data  
from commercially caught  
halibut to assessment*



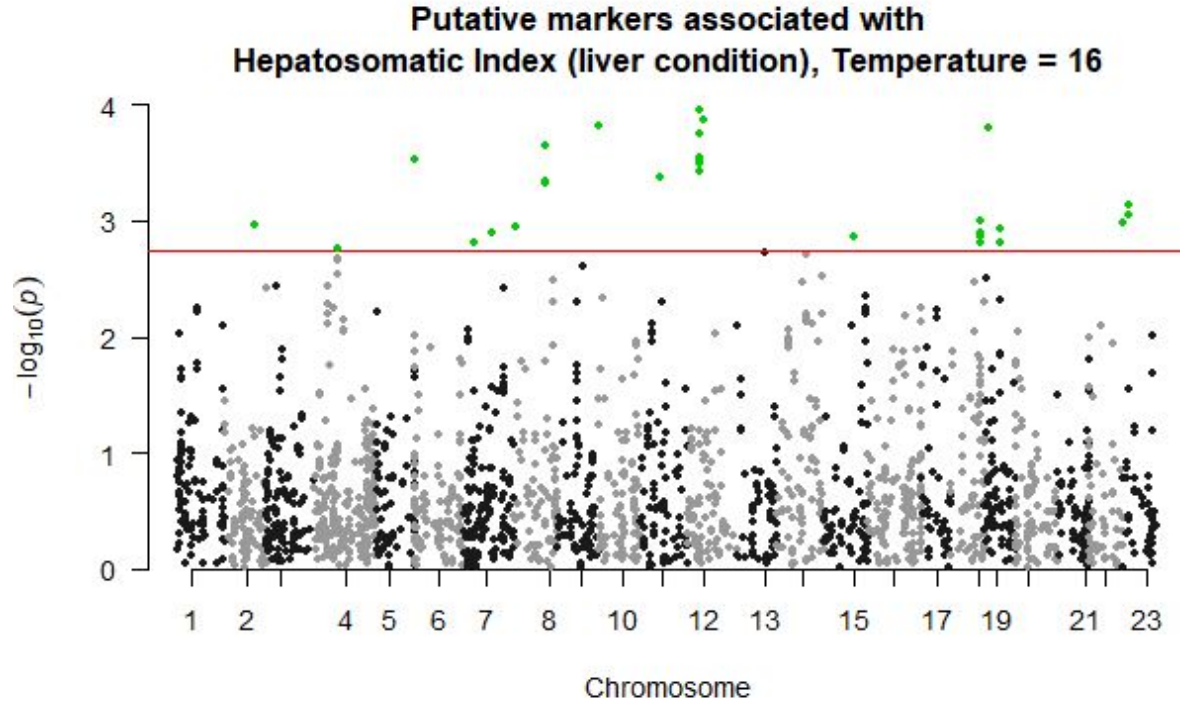
INTERNATIONAL PACIFIC  
HALIBUT COMMISSION

## Genetic Markers (SNPs) For Sex Identification

The sex ratio of the commercial fishery catch represents an extremely important source of uncertainty in the annual stock assessment (Stewart and Hicks, 2020). The IPHC has generated sex information of the entire set of aged commercial fishery samples on an annual basis since 2017 (>10,000 fin clips per year). Sex information is obtained using genetic techniques based on the identification of sex-specific single nucleotide polymorphisms (SNPs) (Drinan et al., 2018) using TaqMan qPCR assays conducted at the IPHC's Biological Laboratory. Therefore, direct estimates of the sex-ratio at age for the directed commercial fishery are now available for stock assessment. Sex-ratio information of the commercial catch is likely to further inform selectivity parameters and cumulatively reduce uncertainty in future estimates of stock size, in addition to improving simulation of spawning biomass in the MSE Operating Model.

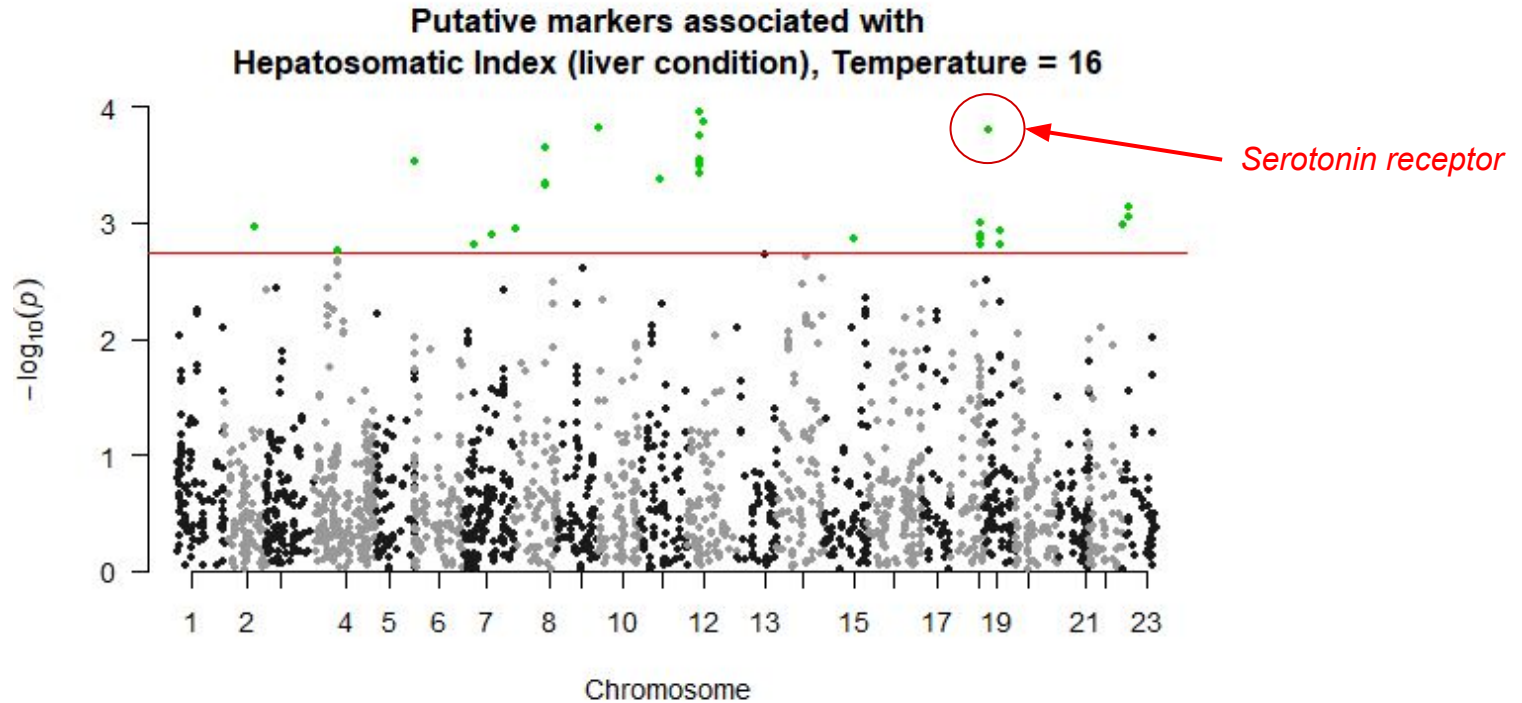


~100 markers putatively associated with **liver size** in Pacific cod juveniles exposed to **warming**



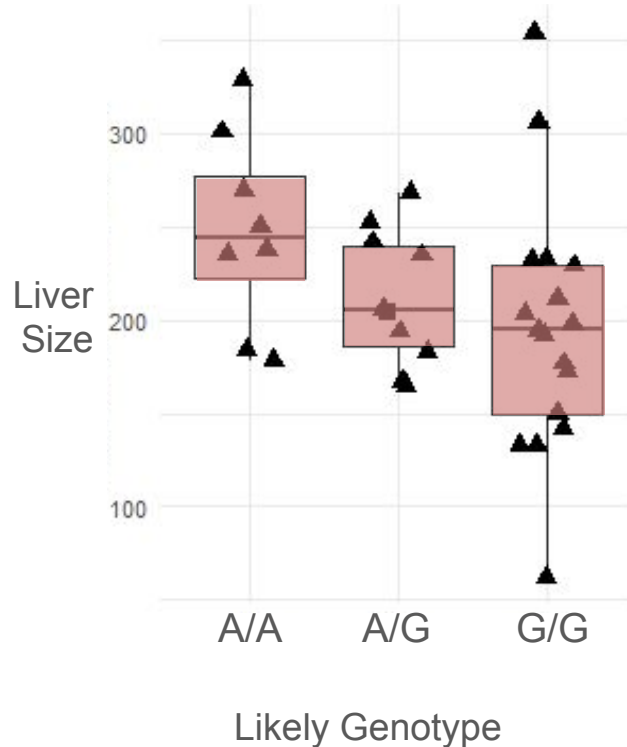


~100 markers putatively associated with **liver size** in Pacific cod juveniles exposed to **warming**





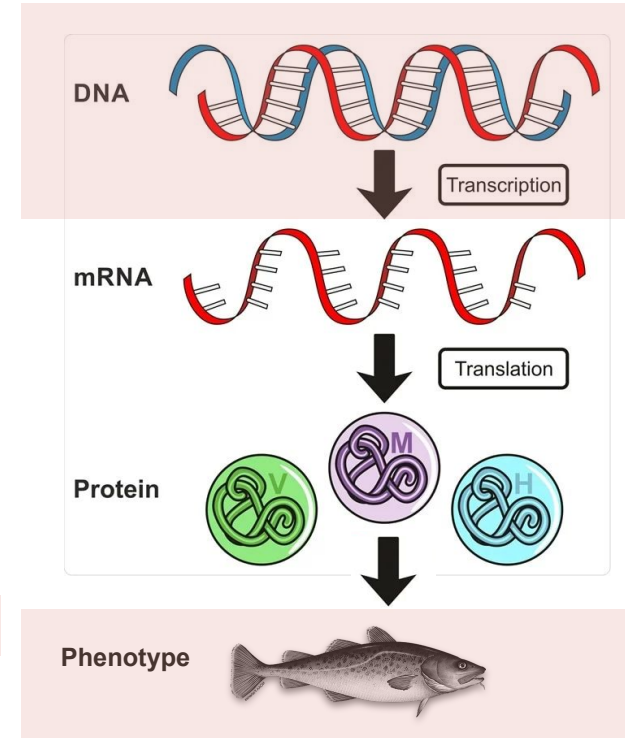
~100 markers putatively associated with **liver size** in Pacific cod juveniles exposed to **warming**



*Marker in a gene coding for a **receptor for serotonin** (5-HT4) which regulates appetite*

# “Genome-to-Phenome” dataset for juvenile Pacific cod

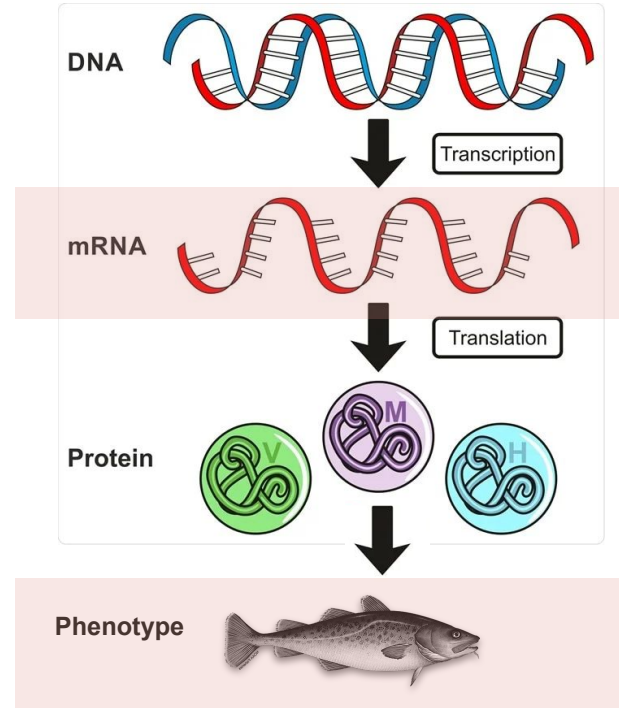
- ✓ Genetics – **they are one population**  
western Gulf of Alaska / Eastern Bering Sea
  - ✓ Phenotypes – **Fewer lipid reserves in warming, slightly slower growth**
  - ✓ Gene expression – **Lipid usage, immune activity, & damage control may deplete energy reserves**
1. Integrate datasets – Performance indicators!
    - ✓ **~100 genetic markers** of liver size in warming
    - a. Expression patterns



Adapted from: udaix/Shutterstock.com

# “Genome-to-Phenome” dataset for juvenile Pacific cod

- ✓ Genetics – **they are one population**  
western Gulf of Alaska / Eastern Bering Sea
  - ✓ Phenotypes – **Fewer lipid reserves in warming, slightly slower growth**
  - ✓ Gene expression – **Lipid usage, immune activity, & damage control may deplete energy reserves**
1. Integrate datasets – Performance indicators!
    - ✓ **~100 genetic markers** of liver size in warming
    - a. Expression patterns



Adapted from: udaix/Shutterstock.com



# What does my gene expression data look like?

Gene ID in genome

Samples

gene_gadmor	PCG001	PCG004	PCG011	PCG015	PCG017	PCG020	PCG029	PCG035	
ND1	8276	8202	8327	7750	5508	4351	5673	6385	
ND2	39142	43297	57032	31681	33489	30275	38921	36460	
COX1	555463	631876	917827	551062	378628	403956	438595	455514	
COX2	315625	309958	493176	294979	189346	229132	227614	219894	
ATP6	82892	105415	89100	73253	77770	58061	86703	78859	
COX3	165275	189321	235193	135559	112870	81377	135462	126615	
ND3	10710	13595	10404	7013	10244	2567	9792	8231	
ND4L	19364	31196	42855	16261	20566	17877	21021	16269	
ND4	37081	42648	67118	30275	29274	37939	35681	34155	
ND5	16299	21803	28231	Gene counts			19960	19262	18187
ND6	4102	3787	3187				1774	3772	3391
CYT8	176843	211635	277391				114784	137070	144369
LOC115539476	260	228	333	219	270	266	312	266	
LOC115539709	890	849	919	777	561	461	945	1000	
LOC115538781	586	596	576	630	417	450	704	730	
abhd14a	1197	1381	1629	1031	774	757	1158	1108	
acy1	1626	1244	1670	1245	1224	1386	1566	1642	
LOC115537228	2106	2402	2555	2008	1845	2826	2127	2235	
LOC115537019	659	544	911	564	505	611	696	444	
LOC115538651	727	674	630	564	479	599	554	523	
LOC115538267	57	192	81	153	104	137	110	104	
kbtbd12	1003	743	646	766	715	875	301	216	

# What does my gene expression data look like?

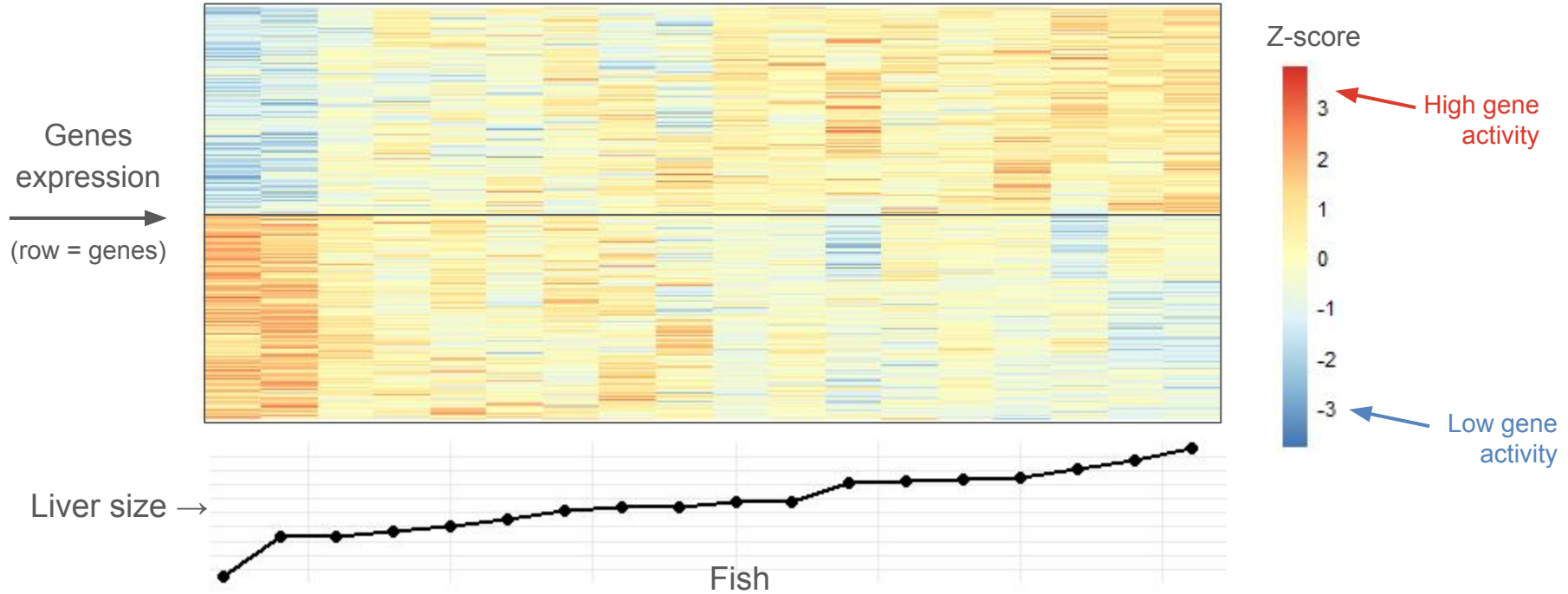
Gene ID in genome

Samples

functional info for many gene

gene_gadmor	PCG001	PCG004	PCG011	PCG015	PCG017	PCG020	PCG029	PCG035	spid	species	evalue	protein_names	
ND1	8276	8202	8327	7750	5508	4351	5673	6385	P55779	GADMO	0.00e+00	NADH-ubiquinone oxidoreductase chain 1 (EC 7.1.1.2) (NA	
ND2	39142	43297	57032	31681	33489	30275	38921	36460	P55780	GADMO	2.40e-158	NADH-ubiquinone oxidoreductase chain 2 (EC 7.1.1.2) (NA	
COX1	555463	631876	917827	551062	378628	403956	438595	455514	Q36775	GADMO	0.00e+00	Cytochrome c oxidase subunit 1 (EC 7.1.1.9) (Cytochrome c	
COX2	315625	309958	493176	294979	189346	229132	227614	219894	Q37741	GADMO	1.12e-132	Cytochrome c oxidase subunit 2 (EC 7.1.1.9) (Cytochrome c	
ATP6	82892	105415	89100	73253	77770	58061	86703	78859	P55778	GADMO	2.60e-104	ATP synthase subunit a (F-ATPase protein 6)	
COX3	165275	189321	235193	135559	112870	81377	135462	126615	P55777	GADMO	5.84e-152	Cytochrome c oxidase subunit 3 (EC 7.1.1.9) (Cytochrome c	
ND3	10710	13595	10404	7013	10244	2567	9792	8231	P15957	GADMO	1.48e-32	NADH-ubiquinone oxidoreductase chain 3 (EC 7.1.1.2) (NA	
ND4L	19364	31196	42855	16261	20566	17877	21021	16269	P23633	GADMO	1.08e-41	NADH-ubiquinone oxidoreductase chain 4L (EC 7.1.1.2) (N	
ND4	37081	42648	67118	30275	29274	37939	35681	34155	P55781	GADMO	0.00e+00	NADH-ubiquinone oxidoreductase chain 4 (EC 7.1.1.2) (NA	
ND5	16299	21803	28231	Gene counts			19960	19262	18187	P55782	GADMO	0.00e+00	NADH-ubiquinone oxidoreductase chain 5 (EC 7.1.1.2) (NA
ND6	4102	3787	318				1774	3772	3391	P55783	GADMO	1.86e-42	NADH-ubiquinone oxidoreductase chain 6 (EC 7.1.1.2) (NA
CYT8	176843	211635	277391				114784	137070	144369	Q37080	GADMO	0.00e+00	Cytochrome b (Complex III subunit 3) (Complex III subunit
LOC115539476	260	228	333	219	270	266	312	266	Q99MK9	MOUSE	1.66e-27	Ras association domain-containing protein 1 (Protein 123F	
LOC115539709	890	849	919	777	561	461	945	1000	Q9WVF8	MOUSE	5.41e-27	Tumor suppressor candidate 2 (Fusion 1 protein) (Fus-1 pr	
LOC115538781	586	596	576	630	417	450	704	730	Q12891	HUMAN	3.51e-104	Hyaluronidase-2 (Hyal-2) (EC 3.2.1.35) (Hyaluronoglucosar	
abhd14a	1197	1381	1629	1031	774	757	1158	1108	Q1LV46	DANRE	4.34e-34	Protein ABHD14A (EC 3.-.-.-) (Alpha/beta hydrolase domai	
acy1	1626	1244	1670	1245	1224	1386	1566	1642	Q6PTT0	RAT	2.57e-16	Aminoacylase-1B (ACY-1B) (EC 3.5.1.14) (ACY 1B) (N-acyl-L-	
LOC115537228	2106	2402	2555	2008	1845	2826	2127	2235	Q6PHS9	MOUSE	1.07e-34	Voltage-dependent calcium channel subunit alpha-2/delta	
LOC115537019	659	544	911	564	505	611	696	444	Q90339	CYPCA	1.94e-172	Myosin heavy chain, fast skeletal muscle	
LOC115538651	727	674	630	564	479	599	554	523	Q9NXG6	HUMAN	5.61e-20	Transmembrane prolyl 4-hydroxylase (P4H-TM) (EC 1.14.11	
LOC115538267	57	192	81	153	104	137	110	104	Q8CIW6	MOUSE	3.91e-23	Solute carrier family 26 member 6 (Anion exchange transp	
kbtbd12	1003	743	646	766	715	875	301	216	Q3ZB90	DANRE	0.00e+00	Kelch repeat and BTB domain-containing protein 12 (Kelch	

~ 1,600 genes with expression associated with *liver size in warming*



# Liver size performance indicators in **warming**, both GENETICS and EXPRESSION

*Cell  
adhesion*

*Calcium  
transport*

*Immune  
system*

Chromosome	# of markers	Gene ID	Protein Name	Function
4	3	LOC132456135	Netrin receptor UNC5D	Cell adhesion, apoptosis in response to DNA damage
12	2	tmco1	Calcium load-activated calcium channel	Calcium transport, endoplasmic reticulum calcium homeostasis
10	1	LOC132466560	TBC1 domain family member 9B	Membrane trafficking, calcium transport
2	1	LOC132453053	Stonustoxin subunit beta	May be related immune system function. From stonefish, toxic/fatal to mammals.
5	1	LOC132457513	Stonustoxin subunit beta	May be related immune system function. From stonefish, toxic/fatal to mammals.
23	1	LOC132452628	NLR family CARD domain-containing protein 3	Negative regulator of the innate immune response
23	1	LOC132452644	HERV-H LTR-associating protein 2	Enhances T-cell proliferation and cytokine production
17	2	LOC132445594	Unknown	Unknown

# Liver size performance indicators in **warming**, both GENETICS and EXPRESSION

*Cell  
adhesion*

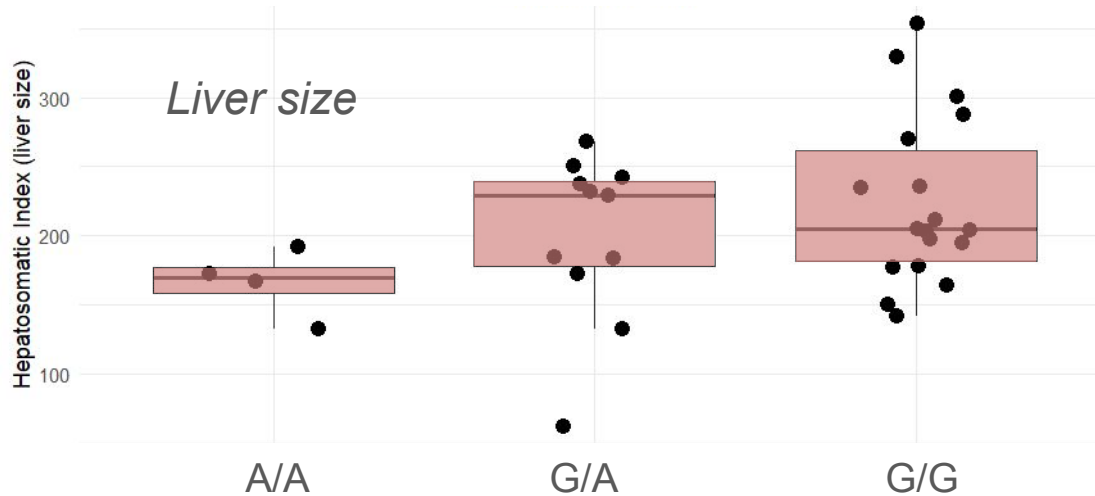
*Calcium  
transport*

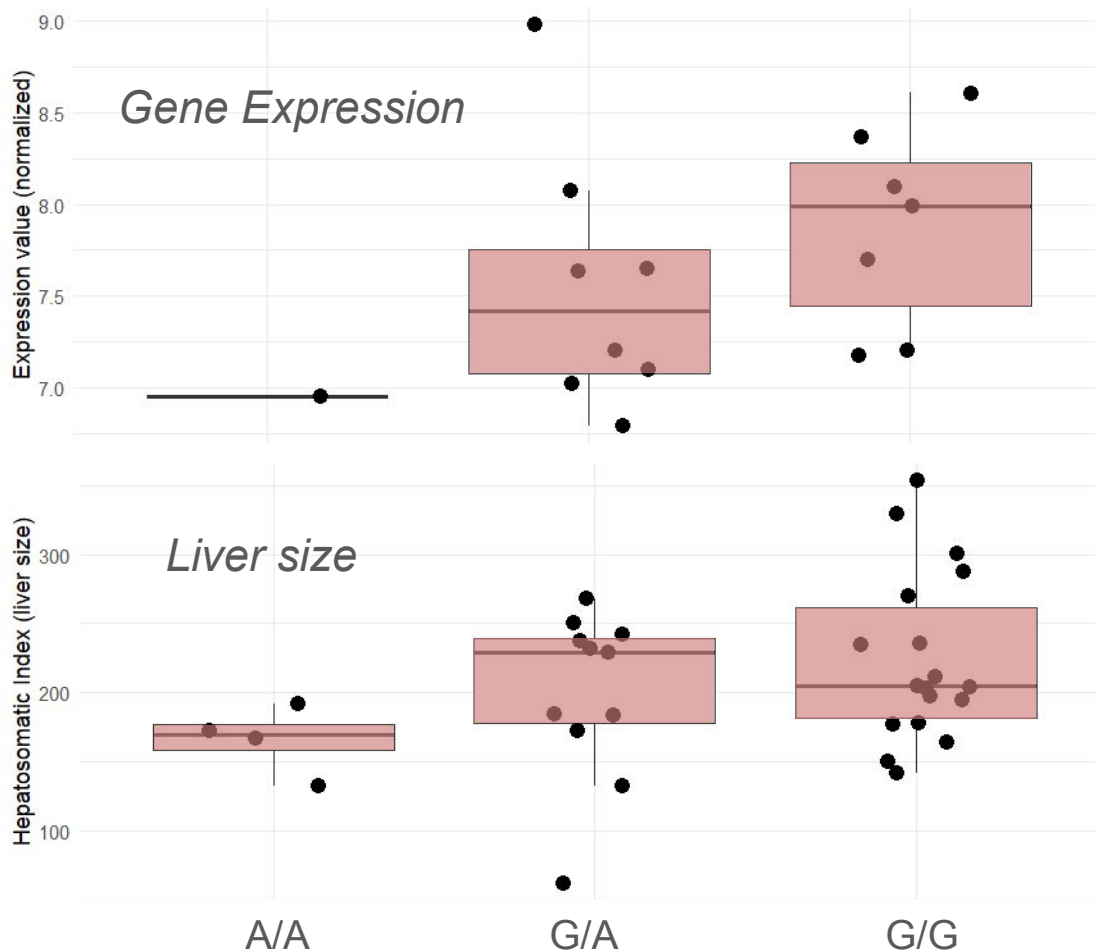
*Immune  
system*

Chromosome	# of markers	Gene ID	Protein Name	Function
4	3	LOC132456135	Netrin receptor UNC5D	Cell adhesion, apoptosis in response to DNA damage
12	2	tmco1	Calcium load-activated calcium channel	Calcium transport, endoplasmic reticulum calcium homeostasis
10	1	LOC132466560	TBC1 domain family member 9B	Membrane trafficking, calcium transport
2	1	LOC132453053	Stonustoxin subunit beta	May be related immune system function. From stonefish, toxic/fatal to mammals.
5	1	LOC132457513	Stonustoxin subunit beta	May be related immune system function. From stonefish, toxic/fatal to mammals.
23	1	LOC132452628	NLR family CARD domain-containing protein 3	Negative regulator of the innate immune response
23	1	LOC132452644	HERV-H LTR-associating protein 2	Enhances T-cell proliferation and cytokine production
17	2	LOC132445594	Unknown	Unknown

*NLR family CARD  
domain-containing  
protein 3*

Negatively  
regulates immune  
system





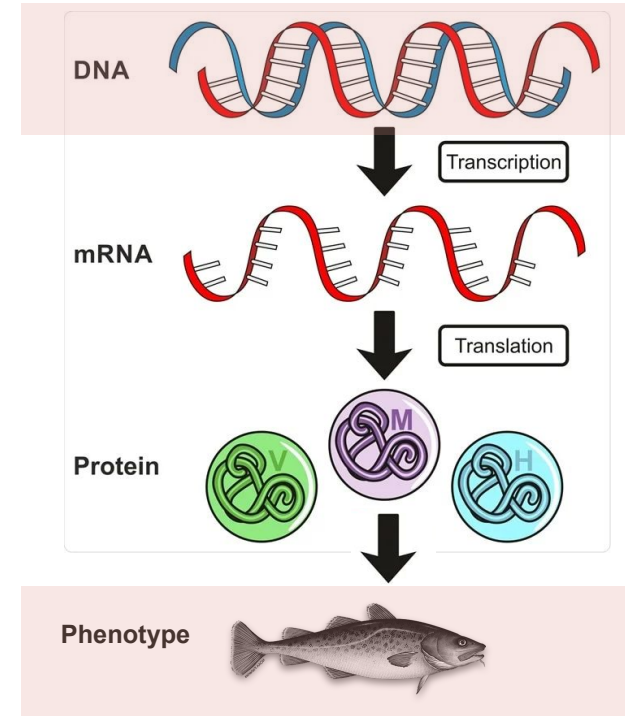
*NLR family CARD  
domain-containing  
protein 3*

Negatively regulates  
immune system

*This marker found  
in all four  
temperature  
treatments*

# “Genome-to-Phenome” dataset for juvenile Pacific cod

- ✓ Genetics – **they are one population**  
western Gulf of Alaska / Eastern Bering Sea
- ✓ Phenotypes – **Fewer lipid reserves in warming, slightly slower growth**
- ✓ Gene expression – **Lipid usage, immune activity, & damage control may deplete energy reserves**
- ✓ Markers of juvenile performance in warming
  - ✓ ~100 genetic markers
  - ✓ ~1,600 gene expression indicators



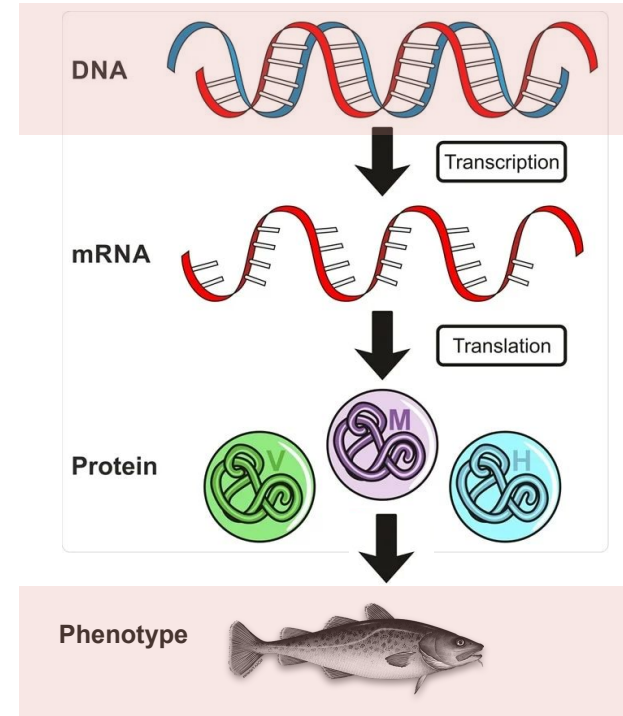
Adapted from: udaix/Shutterstock.com



# “Genome-to-Phenome” dataset for juvenile Pacific cod

- ✓ Genetics – **they are one population**  
western Gulf of Alaska / Eastern Bering Sea
- ✓ Phenotypes – **Fewer lipid reserves in warming, slightly slower growth**
- ✓ Gene expression – **Lipid usage, immune activity, & damage control may deplete energy reserves**
- ✓ Markers of juvenile performance in warming
  - ✓ ~100 genetic markers
  - ✓ ~1,600 gene expression indicators

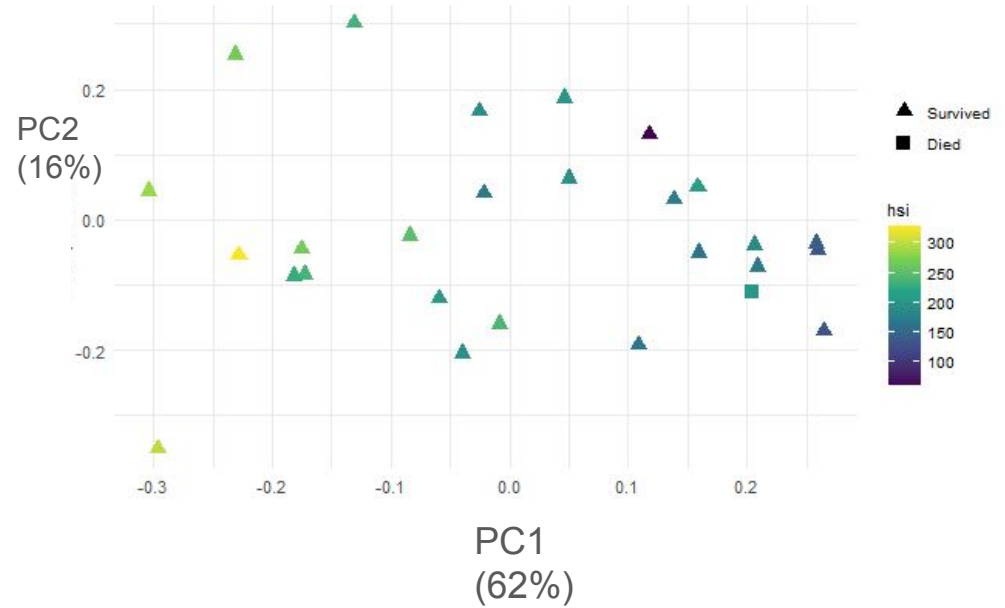
- Can we predict “performance” or “resilience” of other cod groups using our markers?



Adapted from: udaix/Shutterstock.com

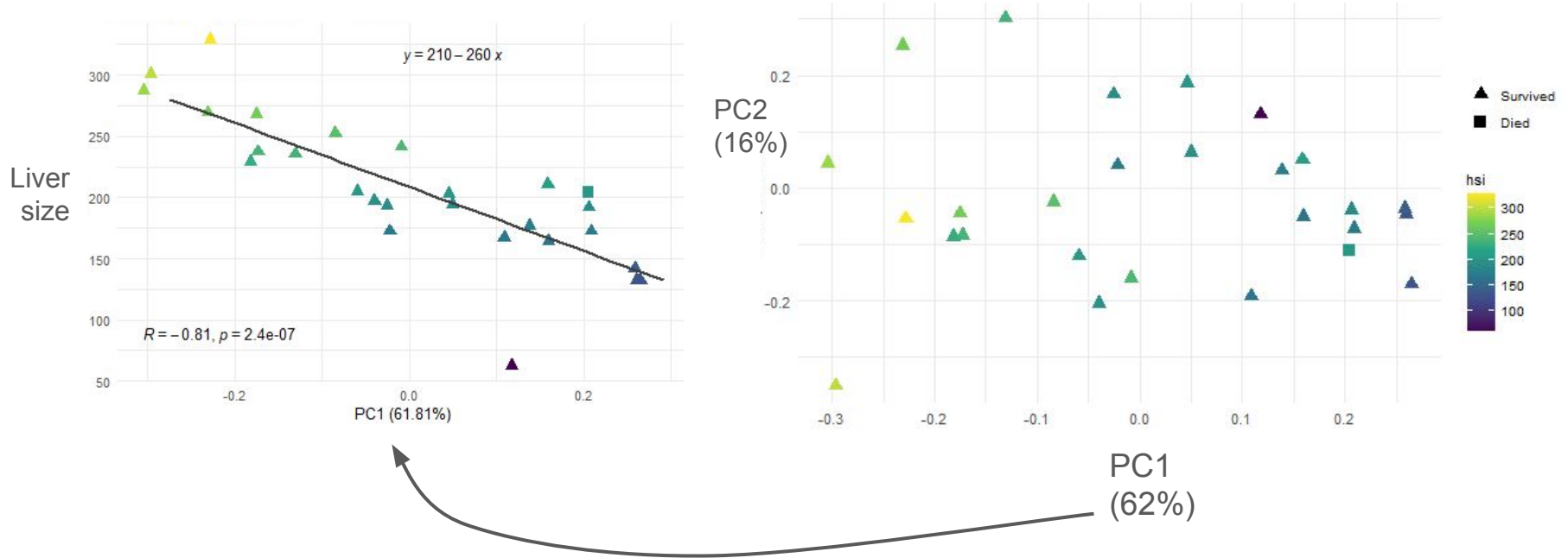
## *Exploratory analysis: predicting liver size*

*PCA from genotype probabilities, liver size markers in warm fish only*



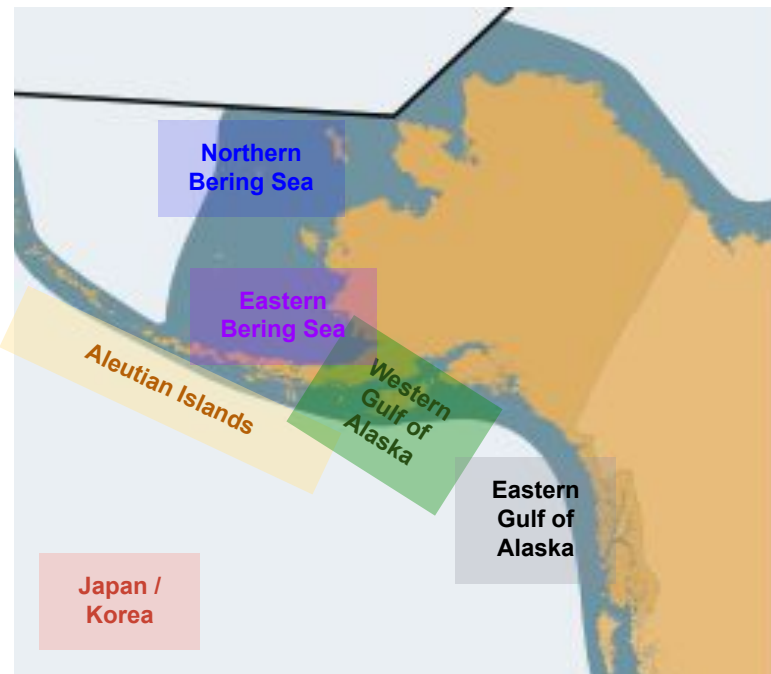
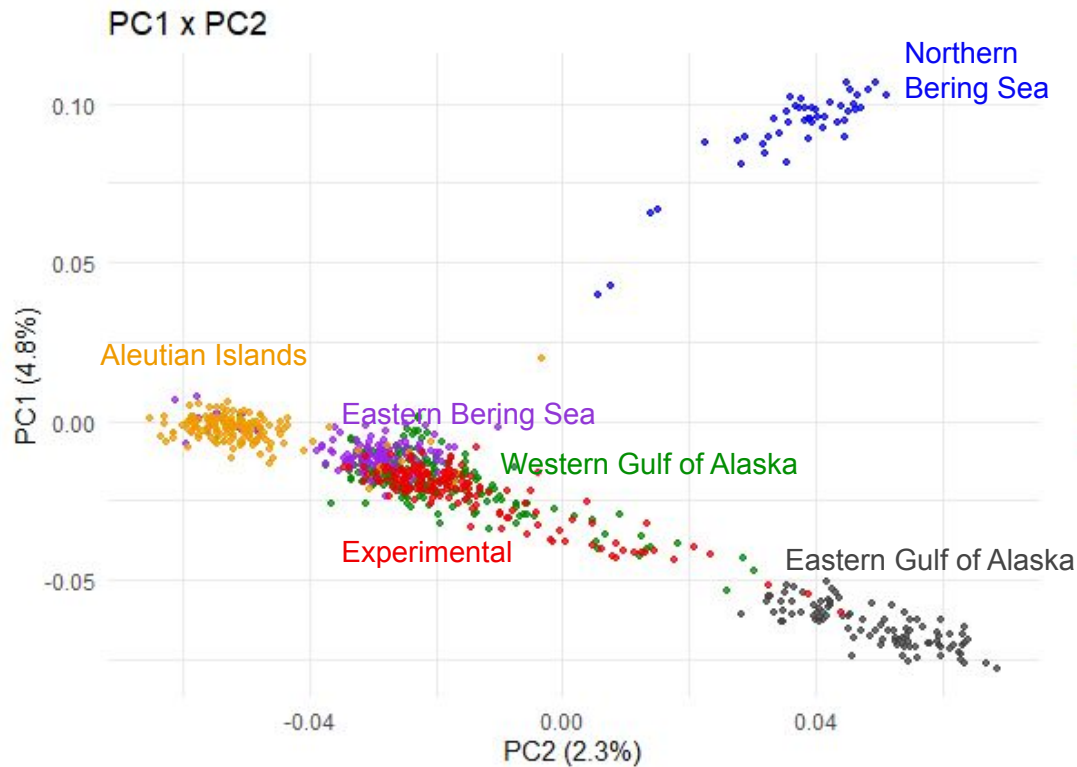
## Exploratory analysis: predicting liver size

PCA from genotype probabilities @ 32 top liver size markers, warm fish only



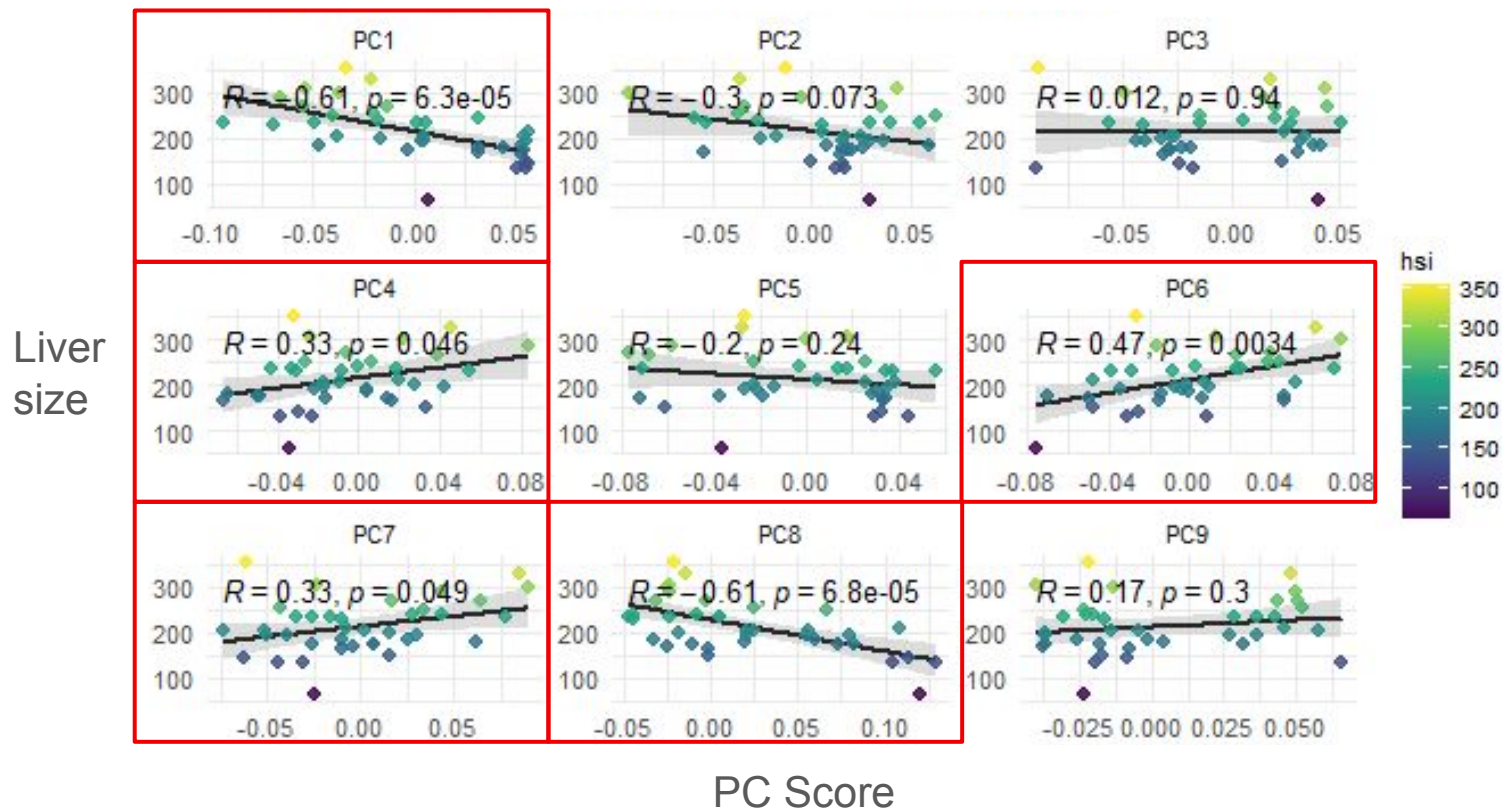
## Exploratory analysis:

*Which populations would we predict to have largest livers in warm conditions?*



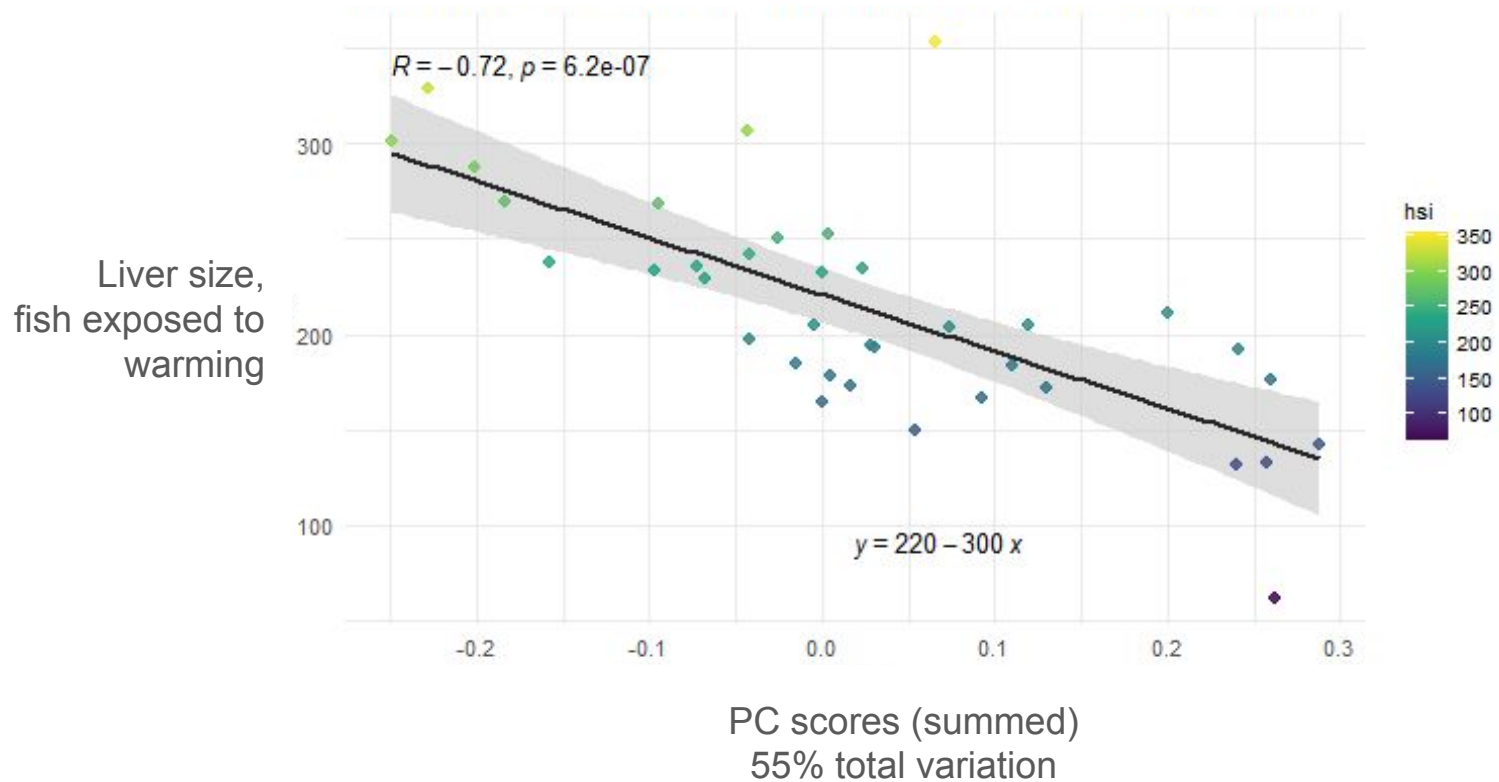
## Exploratory analysis: predicting liver size

### Step 1. PCA from genotype probabilities @ 32 top liver size markers



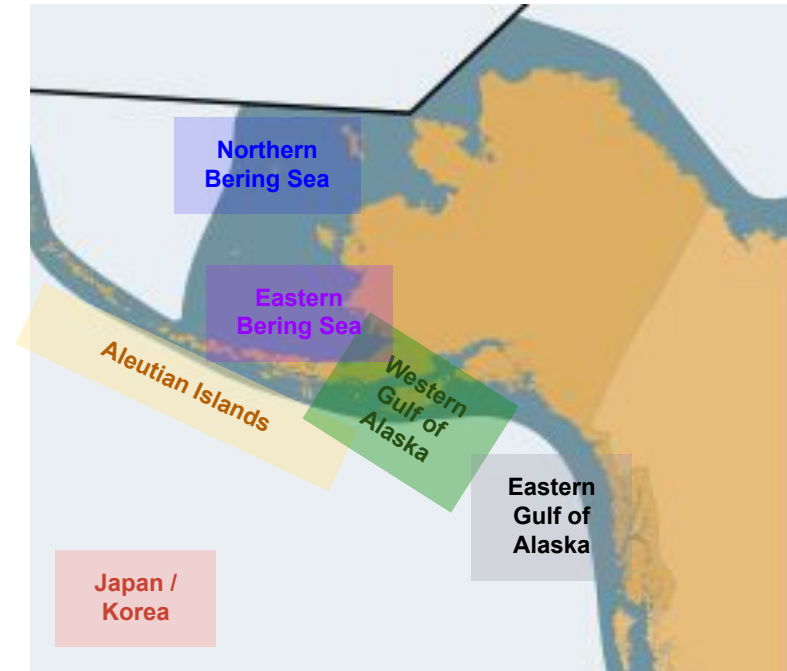
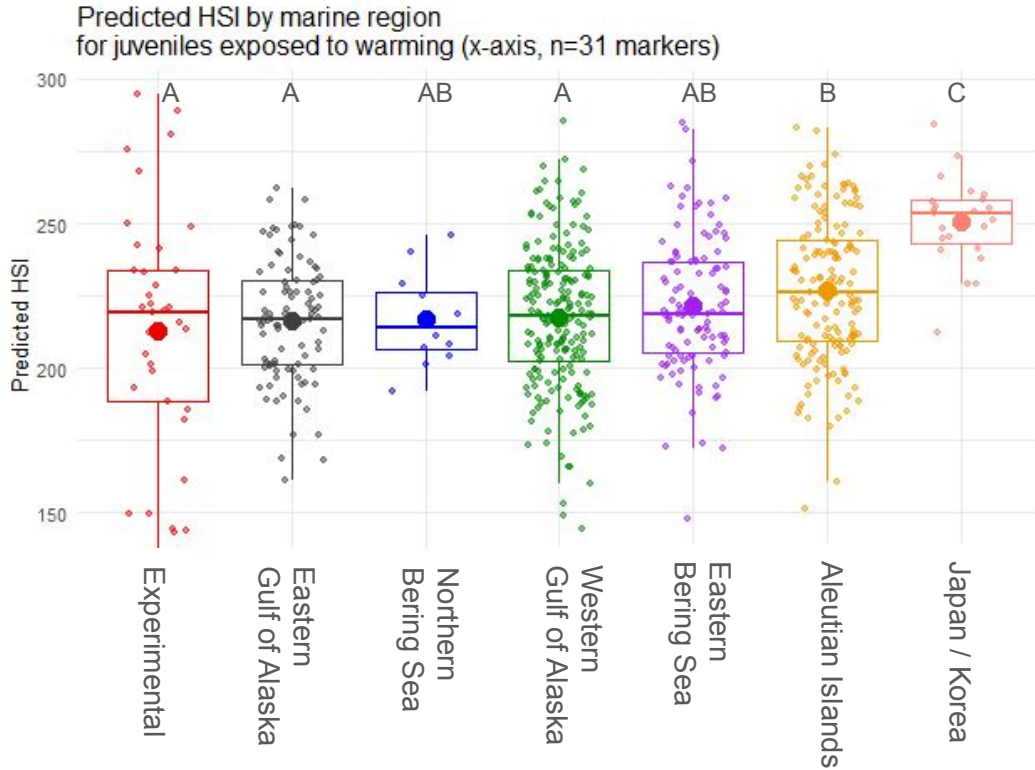
## Exploratory analysis: predicting liver size

### Step 2. Model liver size ~ PC scores in experimental fish



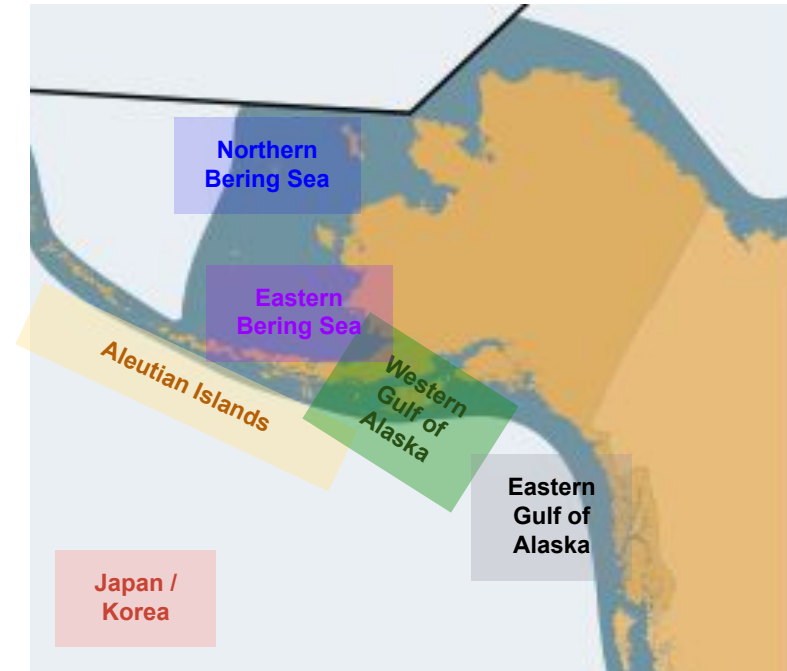
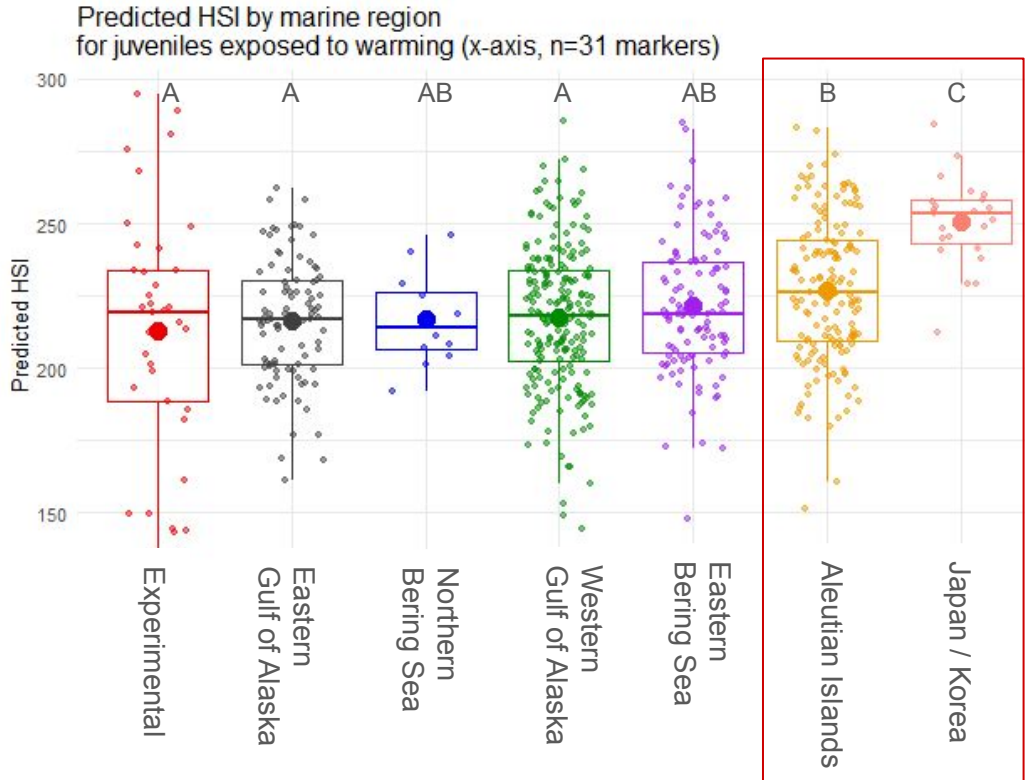
## Exploratory analysis: predicting liver size

### Step 3. Use PC-based model to predict liver size in other populations



## Exploratory analysis: predicting liver size

### Step 3. Use PC-based model to predict liver size in other populations

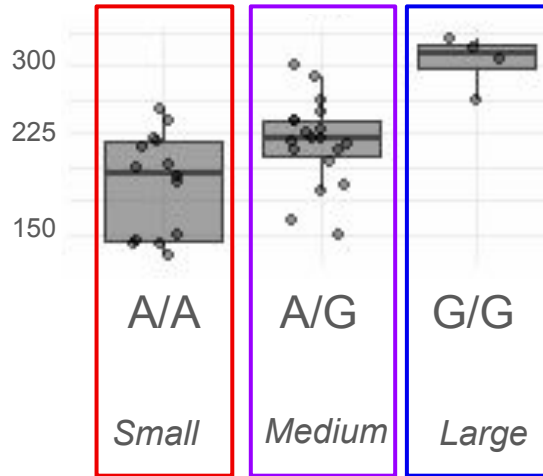




## Example marker in gene **GaINAc-T2**

- *Cell signaling, cell adhesion, and protecting the mucosal surfaces in various tissues*
- *Glycoprotein / glycolipid biosynthesis*

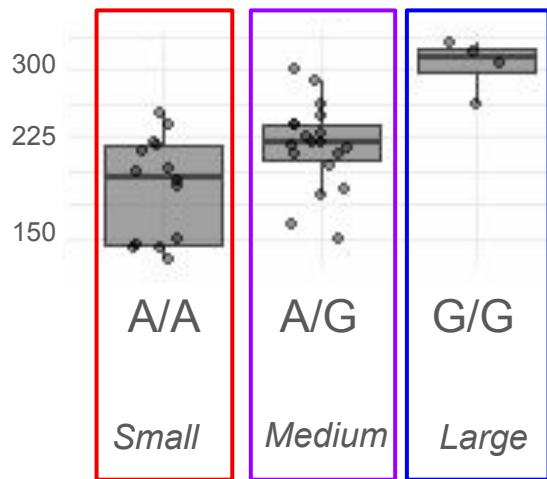
Actual liver size ~ likely genotype  
*Warm-exposed fish*



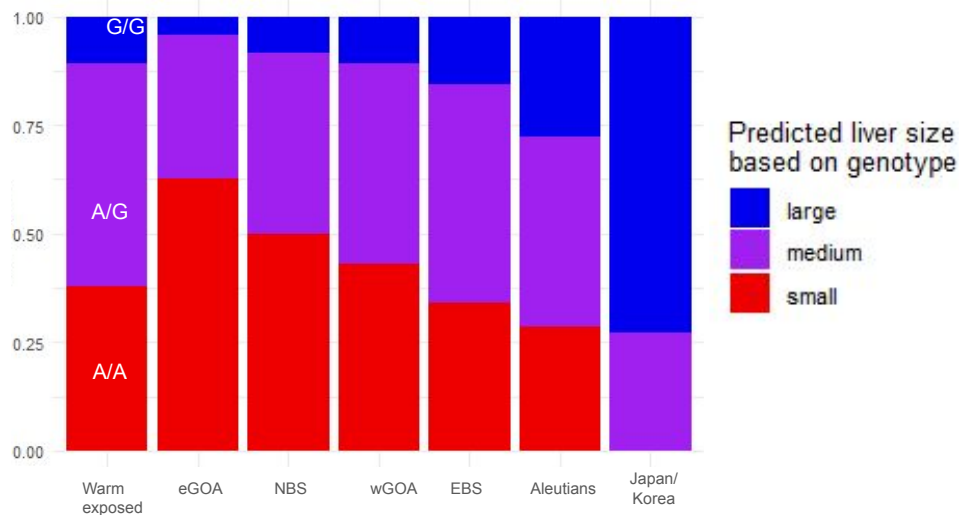
## Example marker in gene **GalNAc-T2**

- Cell signaling, cell adhesion, and protecting the mucosal surfaces in various tissues
- Glycoprotein / glycolipid biosynthesis

Actual liver size ~ likely genotype  
*Warm-exposed fish*



Predicted liver size based on  
likely genotype  
*Reference fish & warm-exposed fish*





## Broad Conclusions (preliminary)

- **Low recruitment in GoA during heatwaves** is likely related to **high larval mortality** and **low juvenile overwintering survival**, both related to *lipid metabolism, inflammation, and cell adhesion effects*.
- Stock assessments may need to **adjust reference points** in heat wave years
- **Genetic variability related to adaptive traits** may enable selection for juveniles **more capable of allocating lipid reserves, more resilient populations**
- Other Pacific cod groups could be screened for **putative markers of performance**
  - Through time – before/during/after heat waves (future project!) - is selection happening?
  - Distinct Pacific cod groups – are some groups more resilient than others?
    - Juveniles using different nursery habitat (onshore vs. offshore, Laurel study)



# Applications of genomic resources in fisheries management

**Building Genomics Database** – Sequence data + metadata = opportunities for integration!

**Sex identification** – for sex data from fisheries or surveys, benchtop assay for experiments, surveys using DNA

**Tagging studies** – identify sex, population of origin, other markers (e.g. climate resilience) using fin clips

**Ancient DNA studies** – have genotypes at temperature-sensitive / performance markers changed through time?

**Aging tools** – “Epigenetic clock”, if developed, to estimate ages from DNA

**Assay development** – Expression data in lieu of more invasive / costly measurements (e.g. lipid components)

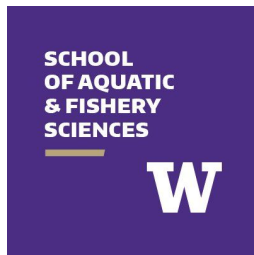
**eDNA** – Collected during surveys to estimate species presence, quantify relative abundance



## Collaborators & Funders

### Newport

Ben Laurel  
Emily Slesinger  
Louise Copeman  
MaryBeth Hicks  
Tom Hurst



### University of Washington

Steven Roberts  
Kathleen Durkin

### Seattle

Ingrid Spies  
Sara Schaal

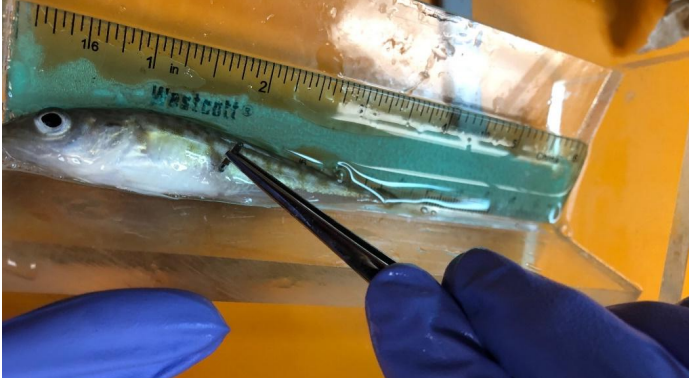
### Juneau

Laura Timm

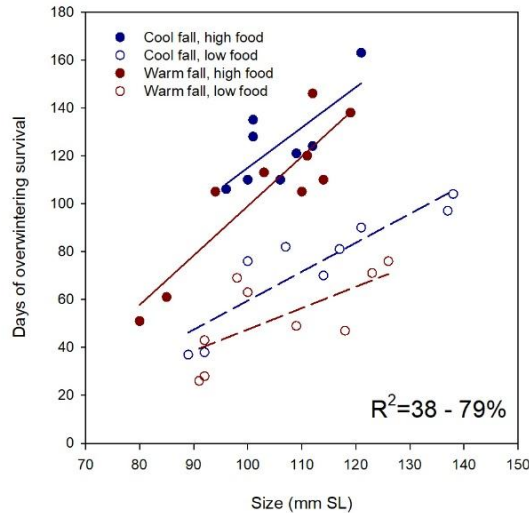


## Questions?

Extra slides



Juvenile cod **overwintering survival** depends on **size, lipid reserves**, and both are temperature-dependent



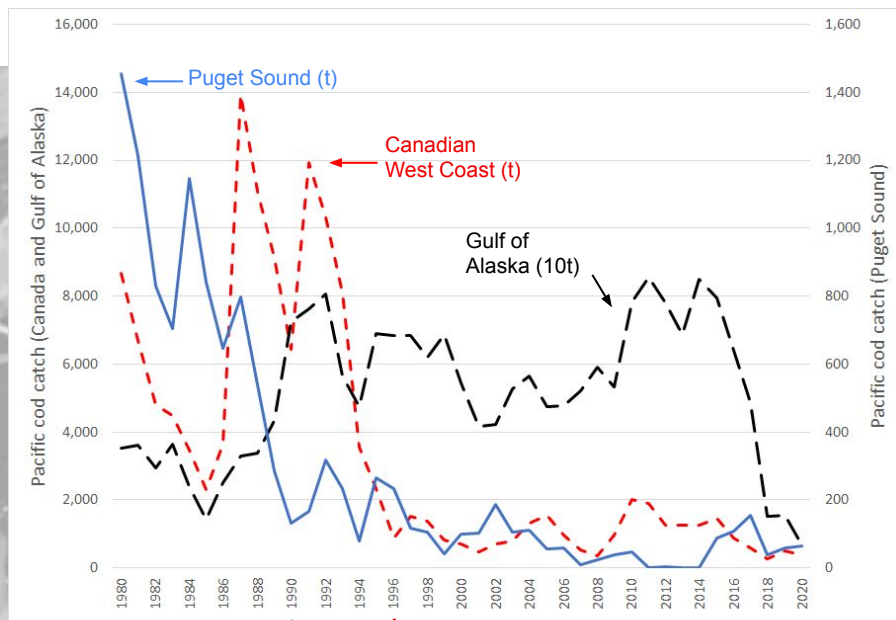
Overwintering survival is  
**higher** after **cooler** autumns

←

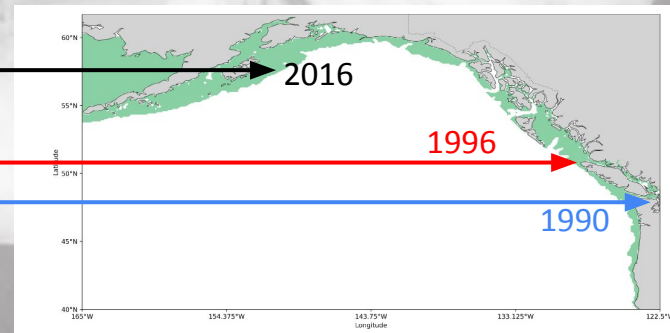
Laurel et al., in review at CJFAS



## A latitudinal progression of population decline...



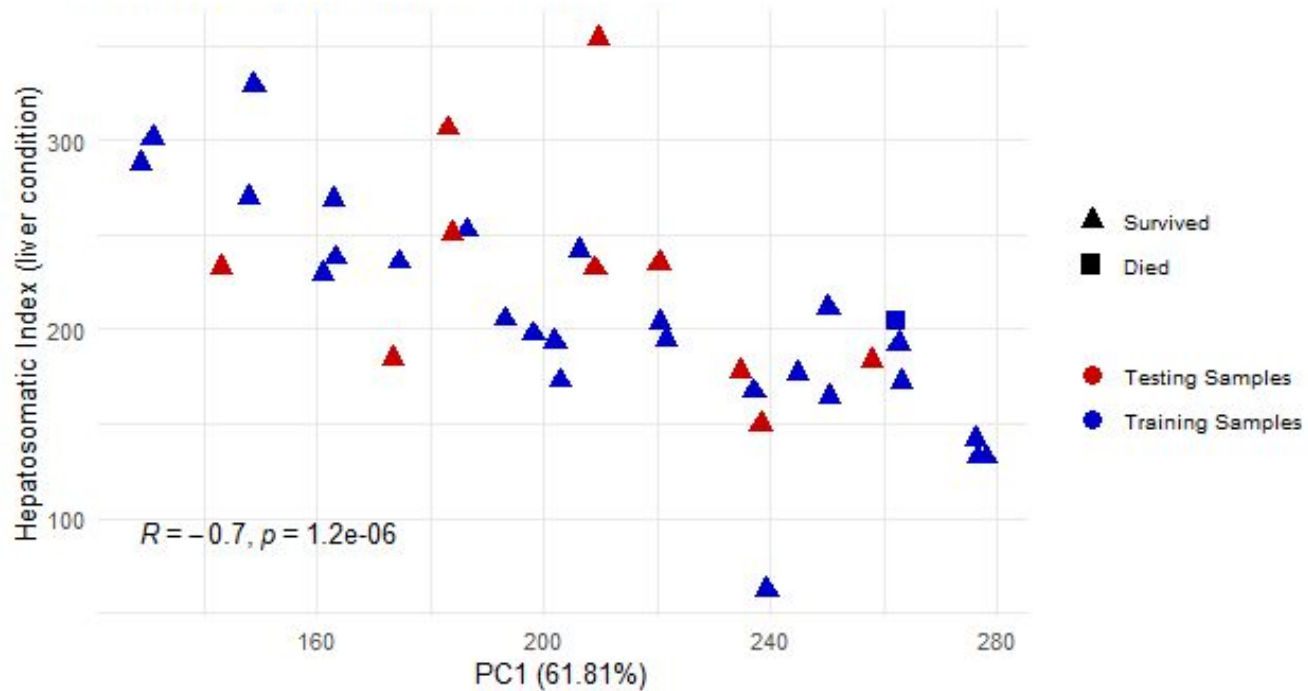
Low Pacific cod recruitment and biomass estimates in Gulf of Alaska coincided/followed the 2014-16 marine heatwaves, prompting review of 1st year of life biology and temperature response experiments



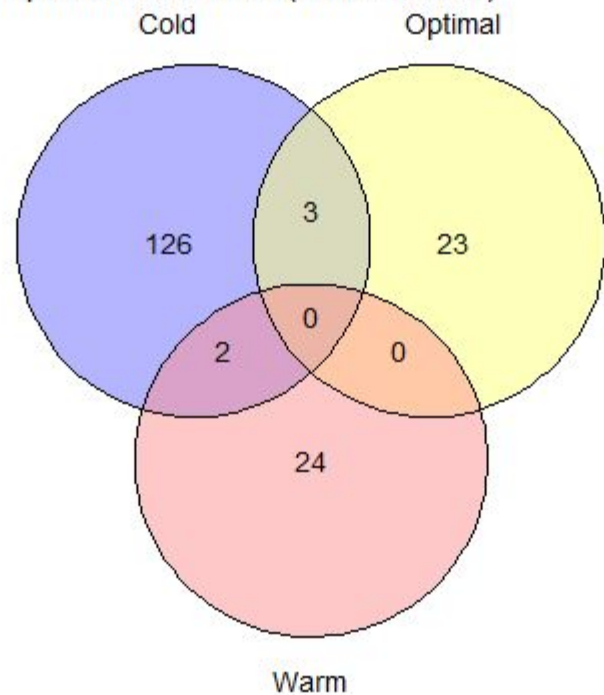


## *Exploratory analysis: predicting liver size*

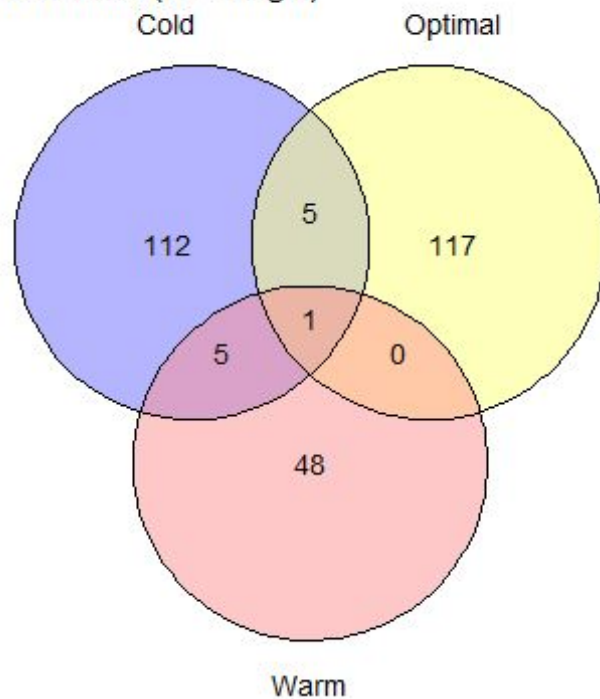
*PCA from genotype probabilities @ 32 top liver size markers, warm fish only*



Genes with putative performance markers  
Hepatosomatic Index (liver condition)

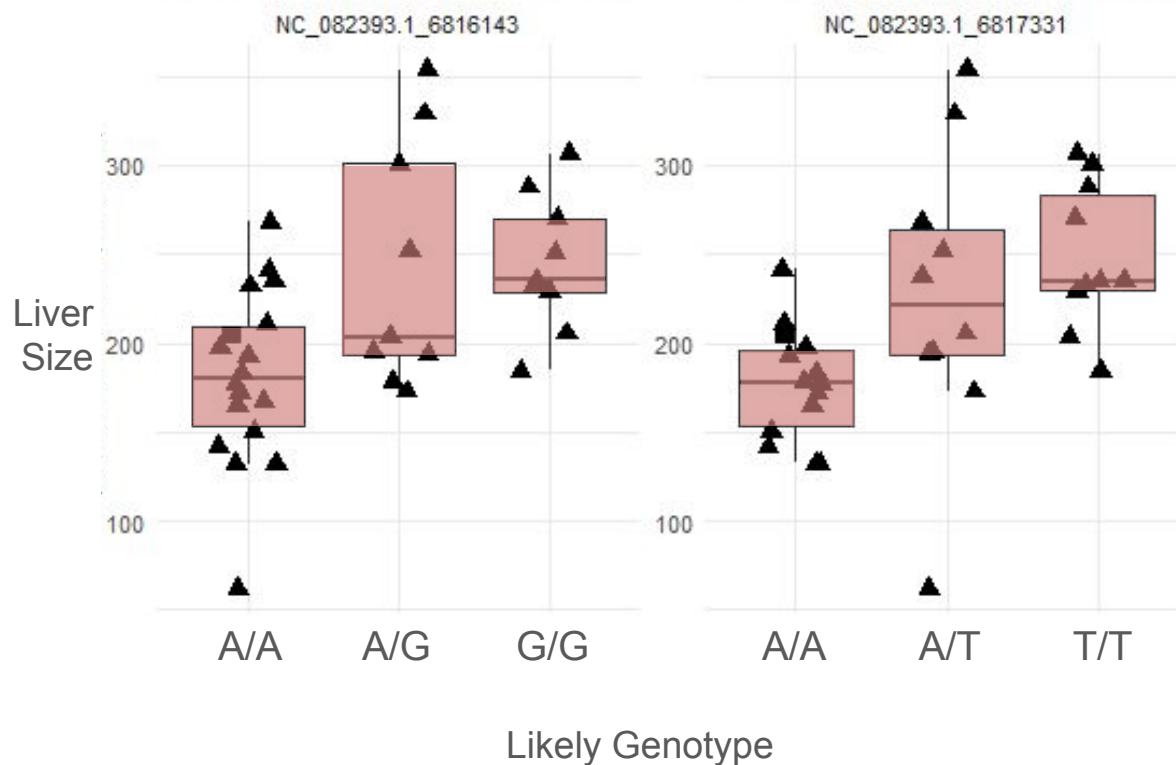


Genes with putative performance markers  
Growth rate (wet weight)





~100 markers putatively associated with liver size in Pacific cod juveniles exposed to warming



One markers in a gene coding for a **calcium channel** involved in calcium homeostasis, metabolic regulation