INTERNATIONAL PACIFIC HALIBUT COMMISSION

Report on current and future biological and ecosystem science research activities

Agenda Item 7
IPHC-2021-SRB019-08
(J. Planas)

Outline

- SRB recommendations and requests from SRB018

2. Reproduction

Research area	Research activities	Research outcomes	Relevance for stock assessment (SA)	SA Rank	Relevance for MSE
Reproduction	Histological maturity assessment	Updated maturity schedule	Scale biomass and reference point estimates	1. Biological input	Improve simulation of spawning biomass in the Operating Model
	Examination of potential skip spawning	Incidence of skip spawning			
	Fecundity assessment	Fecundity-at-age and -size information			
	Examination of accuracy of current field macroscopic maturity classification	Revised field maturity classification			

SRB018–Req.8 (para. 39) The SRB **REQUESTED** that the IPHC Secretariat focus future reproductive biology studies on the development of updated regulatory area-specific maturity ogives (schedules of percent maturity by age).

SRB018–Req.9 (para. 40) The SRB **REQUESTED** that the IPHC Secretariat provide information on the age distribution of all females collected to characterize reproductive development throughout the annual cycle in order to refine efforts to identify potential skip-spawning females.

SRB018–Req.10 (para. 41) The SRB **REQUESTED** that planned studies on fecundity assessment are prioritized and that the sampling design be developed in coordination with the SA to ensure that the results are as informative as possible for assessment purposes. Effective sample stratification along age, weight and length gradients that maximise the contrast in the effect of these variables will be key to precise estimates of fecundity. Oocyte diameter in contrast may be a important covariate to provide but cannot be used in stratification. The primary goal of the fecundity research should be to estimate the exponent of the fecundity vs. weight relationship for incorporation in the SA.

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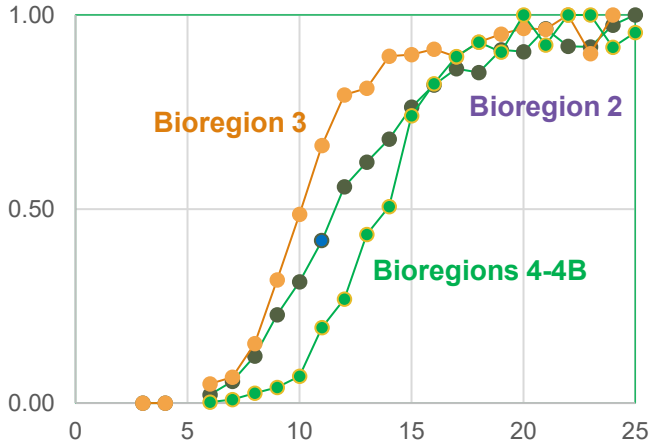
2. Reproduction

Maturity and fecundity sampling: FISS 2022-2023

- 2022 sampling will focus on morphometric vs. histological maturity estimation
- We will need to determine feasible sample sizes:

Step 1: which age-classes define the slope?

	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20
BR2	0.02	0.06	0.12	0.23	0.31	0.42	0.56	0.62	0.68	0.76	0.82	0.86	0.85	0.91	0.91
BR3	0.05	0.07	0.15	0.32	0.49	0.66	0.79	0.81	0.89	0.90	0.91	0.89	0.93	0.95	0.97
BSAI	0.00	0.01	0.03	0.04	0.07	0.19	0.27	0.43	0.51	0.74	0.82	0.89	0.93	0.90	1.00
BR4	0.00	0.01	0.02	0.03	0.07	0.18	0.23	0.40	0.47	0.73	0.81	0.90	0.91	0.87	1.00
BR4B	0.00	0.01	0.03	0.05	0.07	0.21	0.31	0.47	0.54	0.75	0.83	0.89	0.95	0.94	1.00



Step 2: select desired data richness within target age-ranges

Expected sample sizes

	6	7	8	9	10	11	12	13	14	15	16	17	18	19
BR2	3	8	11	10	11	14	**17**	**21**	**21**	20	9	6	5	**3**
BR3	9	**18**	29	22	**20**	**20**	**31**	**34**	35	32	16	11	6	4
BR4	9	22	16	12	15	17	18	**20**	**19**	17	9	**6**	4	2
BR4B	8	15	17	16	14	14	18	**23**	**20**	14	6	**6**	6	2

Step 3+: determine sampling implications for FISS; adjust as necessary

Based on ages that define ~10-90% morphometric maturity:

	range	N Classes	Constrained Age	At N =	SamRt	Total	n/station
BR2	7-19	13	19	3	0.04	124	0.3
BR3	7-15	9	7	18	0.14	243	0.4
BR4	10-17	8	17	6	0.13	251	1.3
BR4B	10-17	8	17	6	0.40	287	2.9
				Grand Total:		905	

2. Reproduction

Maturity and fecundity sampling: FISS 2022-2023

- 2023 sampling will provide samples (whole ovaries) for fecundity estimation
  - Noting the whole ovaries may also be collected in 2022, if feasible

- The rate-selection process will be similar…

Select the desired data richness

Expected sample sizes

	6	7	8	9	10	11	12	13	14	15	16	17	18	19
BR2	3	8	11	10	11	14	**17**	**21**	**21**	20	9	6	5	**3**
BR3	9	**18**	29	22	**20**	**20**	**31**	**34**	35	32	16	11	6	4
BR4	9	22	16	12	15	17	18	**20**	**19**	17	9	**6**	4	2
BR4B	8	15	17	16	14	14	18	**23**	**20**	14	6	**6**	6	2

Determine sampling implications for FISS

Based on ages that define ~10-90% morphometric maturity:

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BR4B	10-17	8	17	6	0.40	287	2.9
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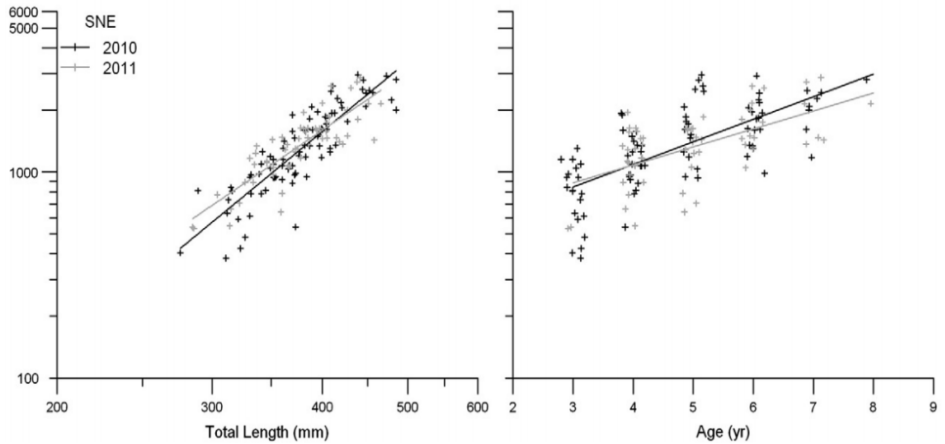
… but with acceptable data-richness defined differently:

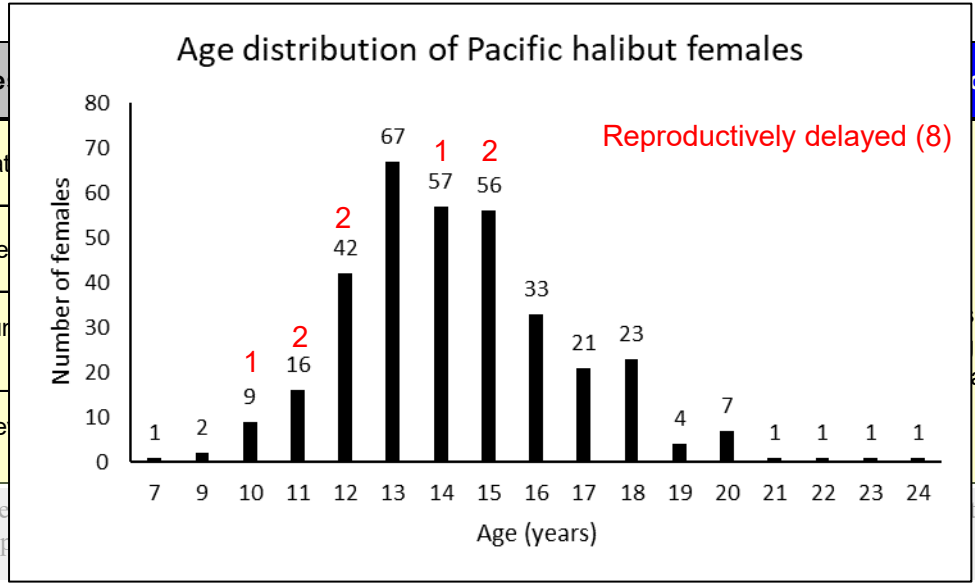
- based on sample sizes required for modeling length- and age-specific fecundity



From: McElroy et al., J. Sea Res **75**(2013):52-61
Fecundity in Winter flounder (*Pseudopleuronectes americanius*)

2. Reproduction

Research area	Research activities	Re	ce for MSE
Reproduction	Histological maturity assessment	Updat	
	Examination of potential skip spawning	Incide	
	Fecundity assessment	Fecur	imulation of biomass in ating Model
	Examination of accuracy of current field macroscopic maturity classification	Re	



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2. Reproduction: fecundity

- Objective: establish a fecundity –size (length/weight/age) relationship.
- Measure: potential annual fecundity as a measure of annual egg production.
- Whole ovaries from 3 females collected during FISS 2021.
- Fecundity assessment method testing planned for late 2021-early 2022.
- Selection of method for fecundity assessment by mid 2022.
- Collection of samples for fecundity assessment planned for FISS 2022

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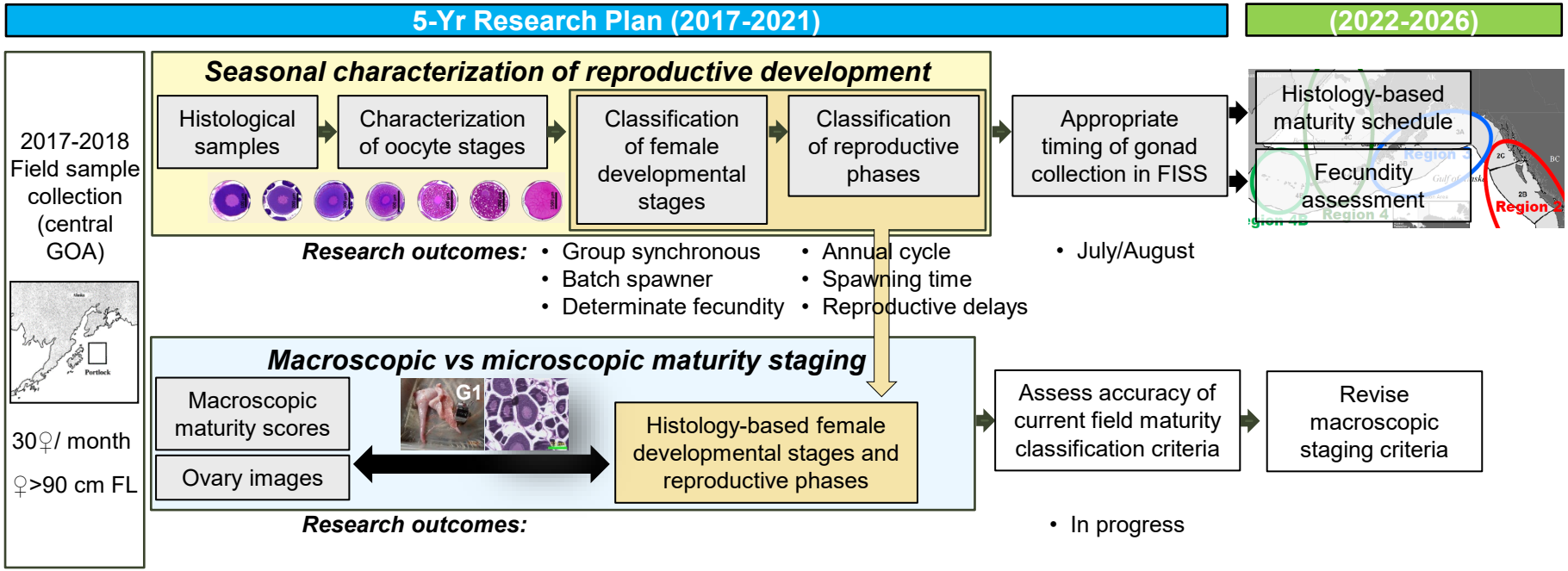

Staff involved: Teresa Fish, MSc candidate APU (2018-2020)
Funding: IPHC (2018-2020)
Publications: Fish et al. (2020) *J. Fish Biol*. **97**: 1880–1885 ; Fish et al. (in preparation)

3. Growth

Research area	Research activities	Research outcomes	Relevance for stock assessment (SA)	SA Rank	Relevance for MSE	MSE Rank
Growth	Identification and application of markers for growth pattern evaluation	Identification and application of markers for growth pattern evaluation	Scale stock productivity and reference point estimates		Improve simulation of variability and allow for scenarios investigating climate change	3. Biological parameterization and validation for growth projections
	Environmental influences on growth patterns	Environmental influences on growth patterns				
	Dietary influences on growth patterns and physiological condition	Dietary influences on growth patterns and physiological condition				

42. The SRB **NOTED** that growth marker genes identified in transcriptomic profiling studies can be informative in future genome scans. However, the SRB **REQUESTED** that the Secretariat explicitly describe how the gene regions identified as 'over' or 'under' expressed would be used. For example, research has yet to determine mechanisms for transcriptional differences other than there is over- or under-representation of mRNA transcripts associated with different treatment groups (e.g. warm vs. cool water) from a heterogeneous set of individuals collected from a single location. The Secretariat has not yet established that results can be generalized to other regions in the species range. Neither has the transcriptional patterns been generalized to individuals of different size/age. These questions should be investigated.

3. Growth

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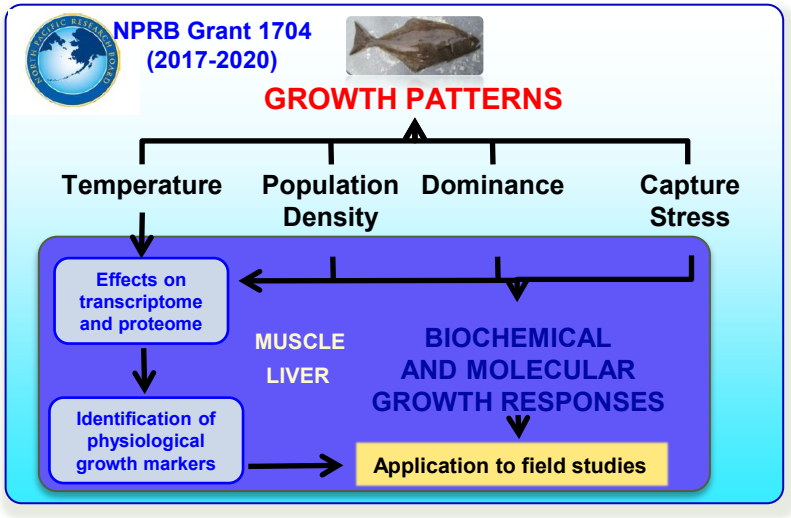
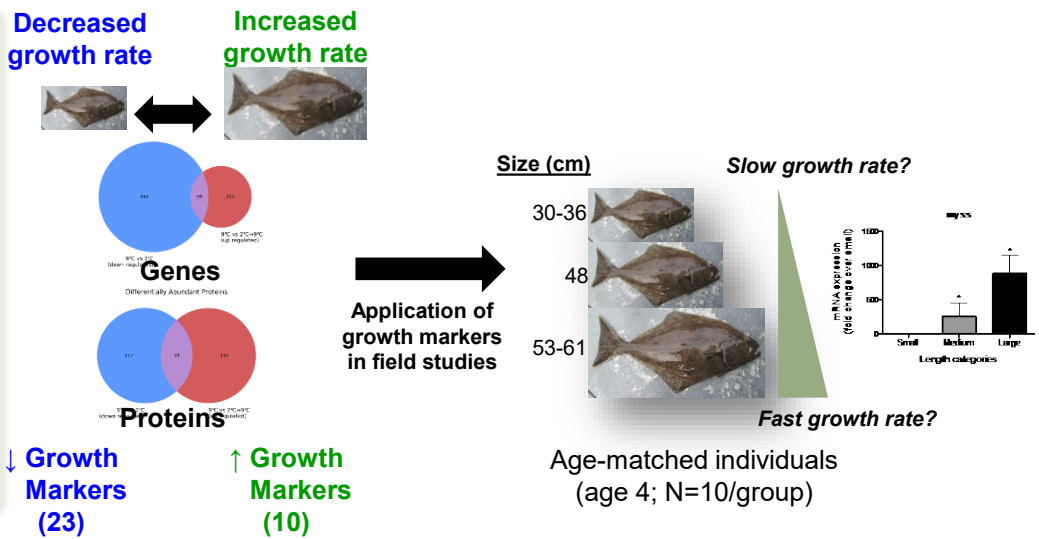




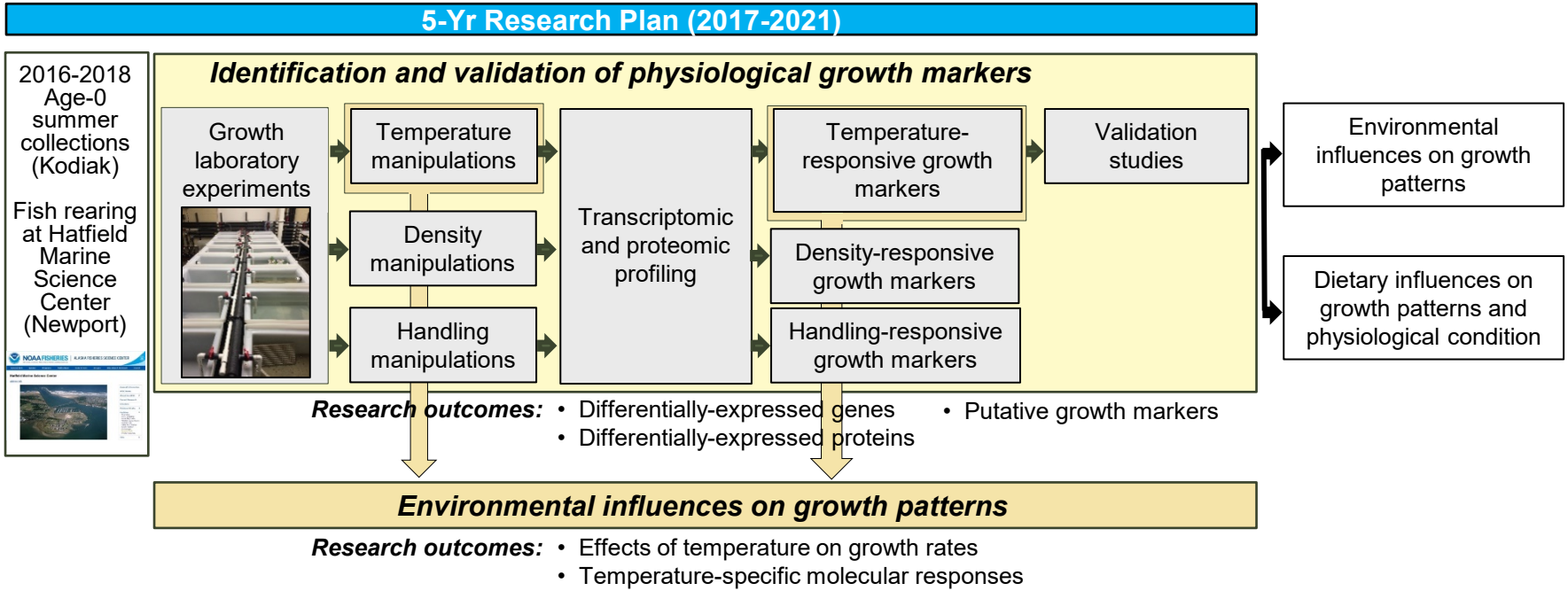
3. Growth

Research area	Research activities	Research outcomes	Relevance for stock assessment (SA)	SA Rank	Relevance for MSE	MSE Rank
		Identification and application				

43. The SRB **REQUESTED** that the Secretariat use these gene regions and align sequences to the whole genome sequence data. Specifically, the Secretariat should investigate whether there is sequence variability within gene coding regions or in regions around gene coding regions that may be transcriptional modifiers (e.g. promoters). If genetic variation exists in or near these genes, these variable base pair position(s) (i.e. single nucleotide polymorphisms or SNPs) should be incorporated in other aspects of the Secretariat research; for example for research activities under the Migration and Population Dynamics Research area.

Transcript ID	Gene	Annotation	Non-coding	Coding	Five prime flanking
TRINITY_DN102963_c0_g1_i1	LOC118098571	glycine--tRNA ligase-like	86	11	94
TRINITY_DN98755_c4_g1_i1	LOC118105518	myosin heavy chain, fast skeletal muscle-like	60	39	30
TRINITY_DN88997_c0_g1_i1	LOC118110038	troponin I, slow skeletal muscle-like	52	6	94
TRINITY_DN105325_c2_g1_i1	LOC118118854	zinc finger protein 638-like	529	52	101
TRINITY_DN104023_c1_g2_i2	LOC118124806	asparagine synthetase [glutamine-hydrolyzing]-like	242	23	77
TRINITY_DN105033_c2_g1_i1	acta1a	actin alpha 1, skeletal muscle a	18	7	104
TRINITY_DN97221_c0_g3_i1	mylpfb	myosin light chain, phosphorylatable, fast skeletal muscle b	29	2	71
TRINITY_DN97789_c1_g1_i1	rhcga	Ammonium transporter, Rh family, C glycoprotein a	30	7	28
TRINITY_DN87895_c0_g1_i2	ttn.1	titin, tandem duplicate 1	420	205	124
TRINITY_DN106670_c2_g1_i1	ubp1	upstream binding protein 1	121	7	84

3. Growth



Staff involved: Andy Jasonowicz, Anna Simeon
Funding: NPRB Grant#1704 (Sept. 2017-Feb. 2020)
Publications: Planas et al. (in preparation)

4. DMRs and Survival Assessment

Research area	Research activities	Research outcomes	Relevance for stock assessment (SA)	SA Rank	Relevance for MSE	MSE Rank
Mortality and survival assessment	Discard mortality rate estimate: longline fishery	Experimentally-derived DMR	Improve estimates of unobserved mortality		Improve estimates of stock productivity	1. Fishery parameterization
	Discard mortality rate estimate: recreational fishery					2. Fishery parameterization
	Best handling practices: longline fishery	Guidelines for reducing discard mortality		2. Fishery yield		
	Best handling practices: recreational fishery	Guidelines for reducing discard mortality		3. Fishery yield		

- ***Guided recreational fishery***

NFWF National Fish and Wildlife Foundation
NPRB Grant No. 2009

1. Collect information on hook types and sizes and handling practices
2. **Investigate the relationship between gear types and capture conditions and size composition of captured fish**
3. **Injury profiles and physiological stress levels of captured fish**
4. **Assessment of mortality of discarded fish**

- Sitka: 21 – 27 May 2021
- Seward: 11 – 17 June 2021

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NFWF National Fish and Wildlife Foundation
UAF University of Alaska Fairbanks
Alaska Pacific University
Alaska Charter Association
NPRB Grant No. 2009

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- Sitka: 21 – 27 May 2021

Size classes (cm)				
≤ 68	69-77	78-93	≥ 94	Total
63	**75**	**66**	**39**	**243**

- Two gear sizes: 12/0 and 16/0 hooks
- Observations and samples: hooking time, time on deck, weight, length, hook injury type and picture, viability, fat content, fish temperature, blood sample, fin clip, wire tag.

4. DMRs and Survival Assessment

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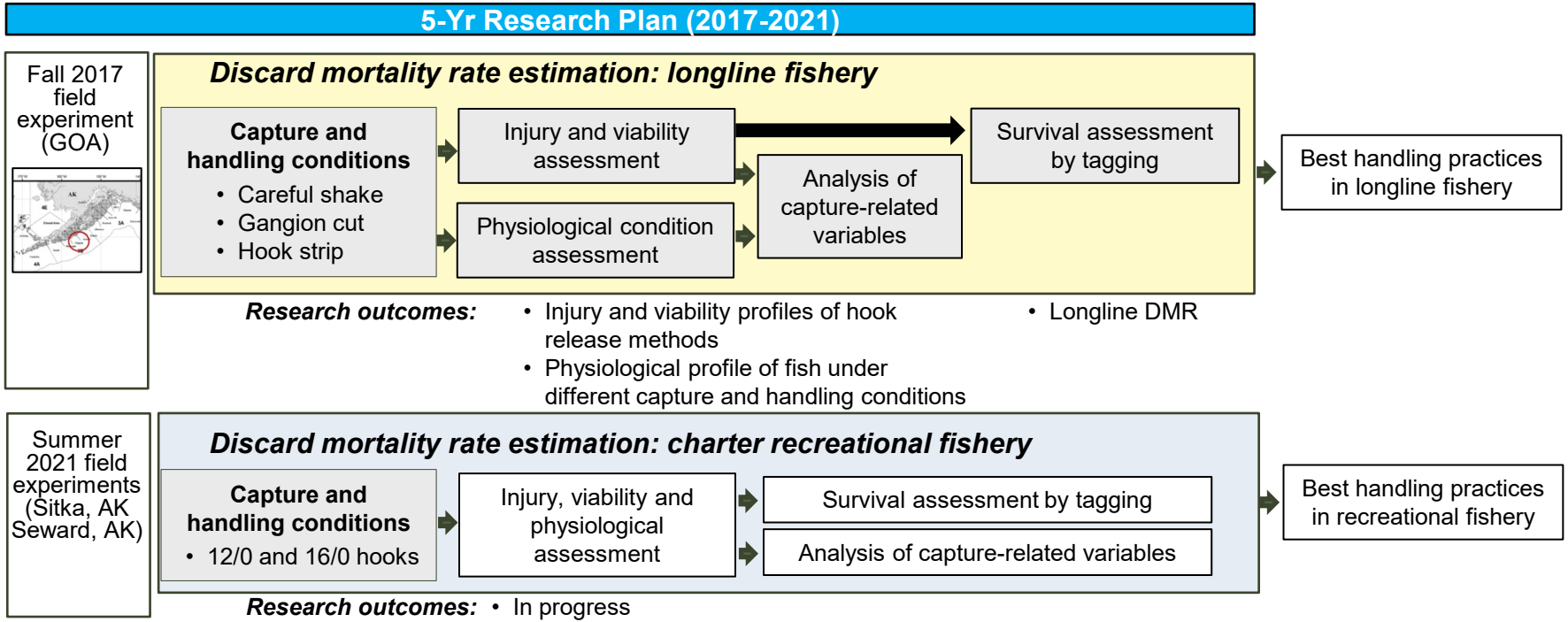
- Seward, AK (3A): 11 – 17 June 2021

Types of tags		
Wire	sPATs	Total
38	80	118

- Two gear sizes: 12/0 and 16/0 hooks
- Observations and samples: hooking time, time on deck, weight, length, hook injury type and picture, viability, fat content, fish temperature, blood sample, fin clip, tag.

4. Mortality and Survival Assessment



Staff involved: Tim Loher, Claude Dykstra, Allan Hicks, Ian Stewart

Funding: Saltonstall-Kennedy NOAA (Sept. 2017-Aug. 2020); National Fish and Wildlife Foundation (Apr. 2019-Nov. 2021)

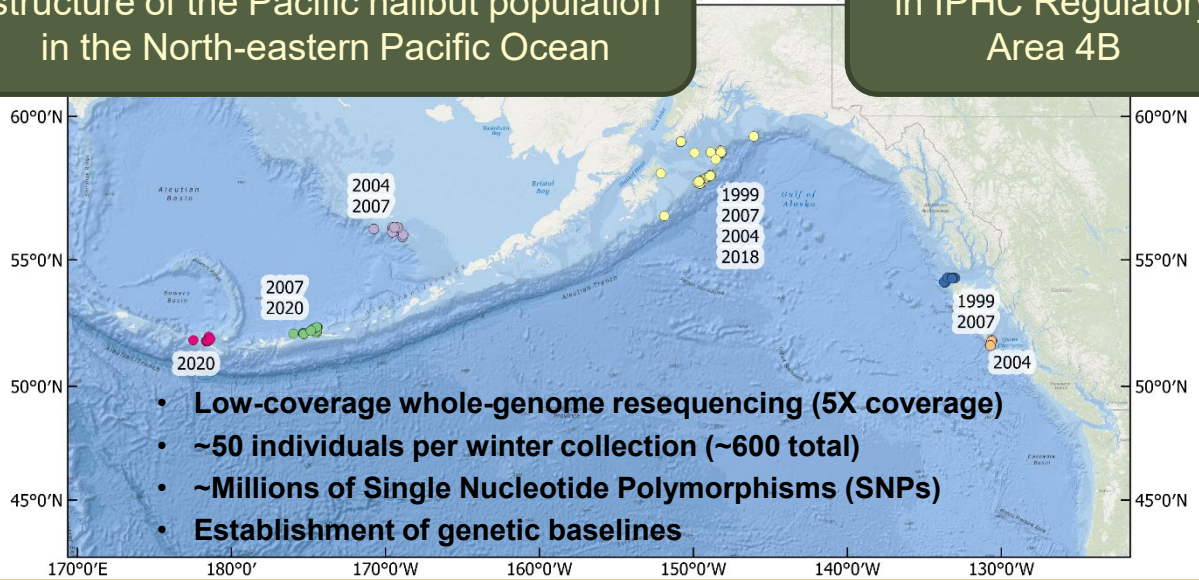
Publications: Kroska et al. (2021) *Conserv. Physiol.*; Loher et al. (in review) *North Amer. J. Fish. Manag.*

5. Genetics and Genomics

Research area	Research activities	Research outcomes	Relevance for stock assessment (SA)	SA Rank	Relevance for MSE	MSE Rank
Genetics and genomics	Population structure	Stock structure of IPHC Regulatory Area 4B relative to the rest of the Convention Area	Altered structure of future stock assessments	2. Biological input	Improve parametization of the Operating Model	1. Biological parameterization and validation of movement estimates. 2. Biological parameterization and validation of recruitment distribution
		Assignment of individuals to				

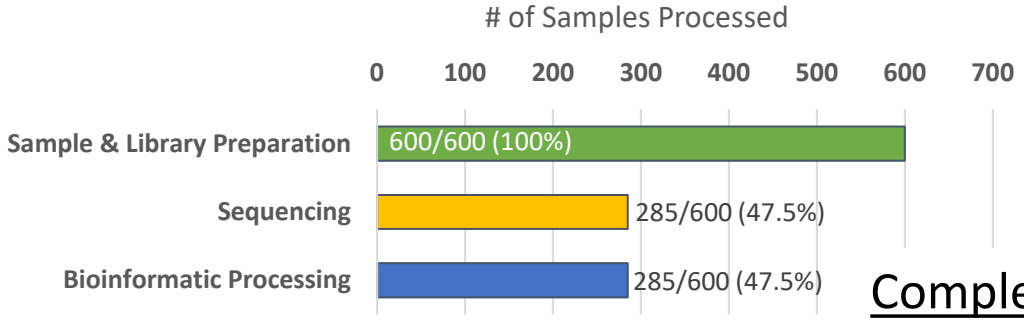
Revise our understanding of genetic structure of the Pacific halibut population in the North-eastern Pacific Ocean

Analysis of structure in IPHC Regulatory Area 4B



- Low-coverage whole-genome resequencing (5X coverage)
- ~50 individuals per winter collection (~600 total)
- ~Millions of Single Nucleotide Polymorphisms (SNPs)
- Establishment of genetic baselines

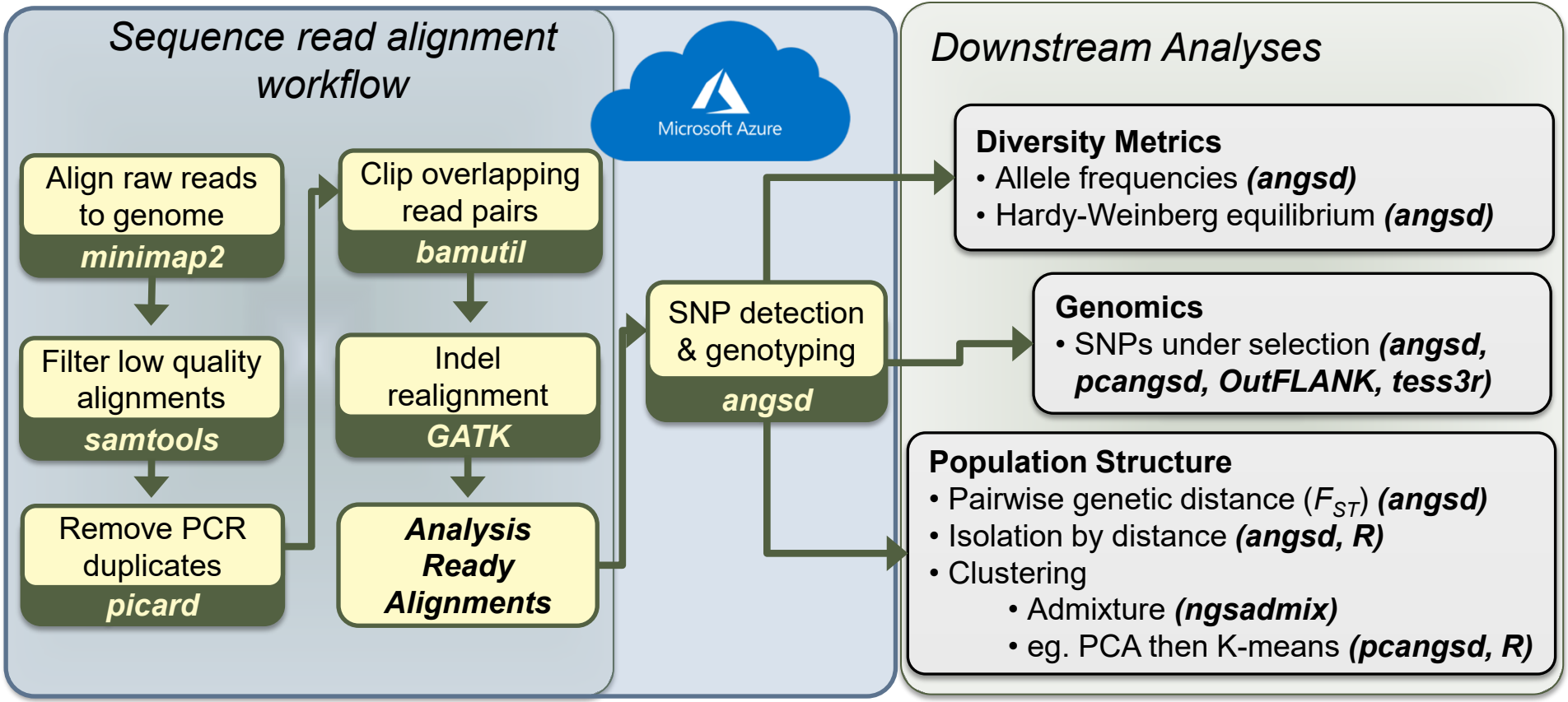
5. Genetics and Genomics



Completed sequencing runs to date:

	Run 1 (Sept 2020)	Run 2 (Feb 2021)
N	36	250
Sequencing Platform	Illumina HiSeq 4000	Illumina NovaSeq S4
Raw Reads Per Sample	26.5 million (21.8 - 42.9)	24.7 million (10.7 – 47.2)
Reads Retained	15.8 million (13 - 24.9) 60% (54% - 69%)	15.4 million (4.2 - 26.4) 63% (22% - 70%)
Coverage Per Sample	3.2x (2.6 – 5x)	3.5x (1.0 - 5.6x)

5. Genetics and Genomics



Methods

- ANGSD (v0.934) (Korneliussen et al. 2014)
  - global minor allele frequency (MAF) ≥ 0.01
  - p-value 1e-6 ≤ less for a site being variable
  - 214 out of 285 (~75%) of individuals

10,474,925 SNPs

- Removed SNPs in unplaced scaffolds, chr 9, and mt genome prior to analysis

10,039,557 SNPs

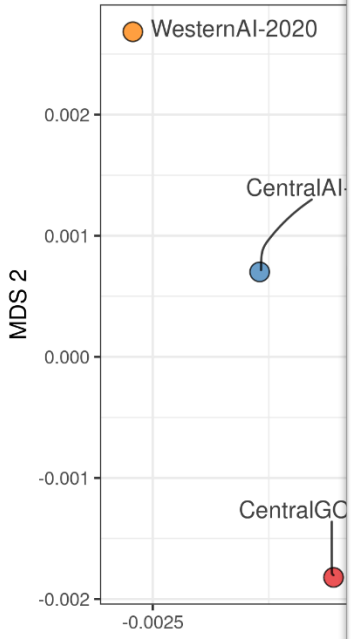
- PCangsd (v1.02) (Meisner & Albrechtsen 2018)
  - Default settings (MAF ≥ 0.05)

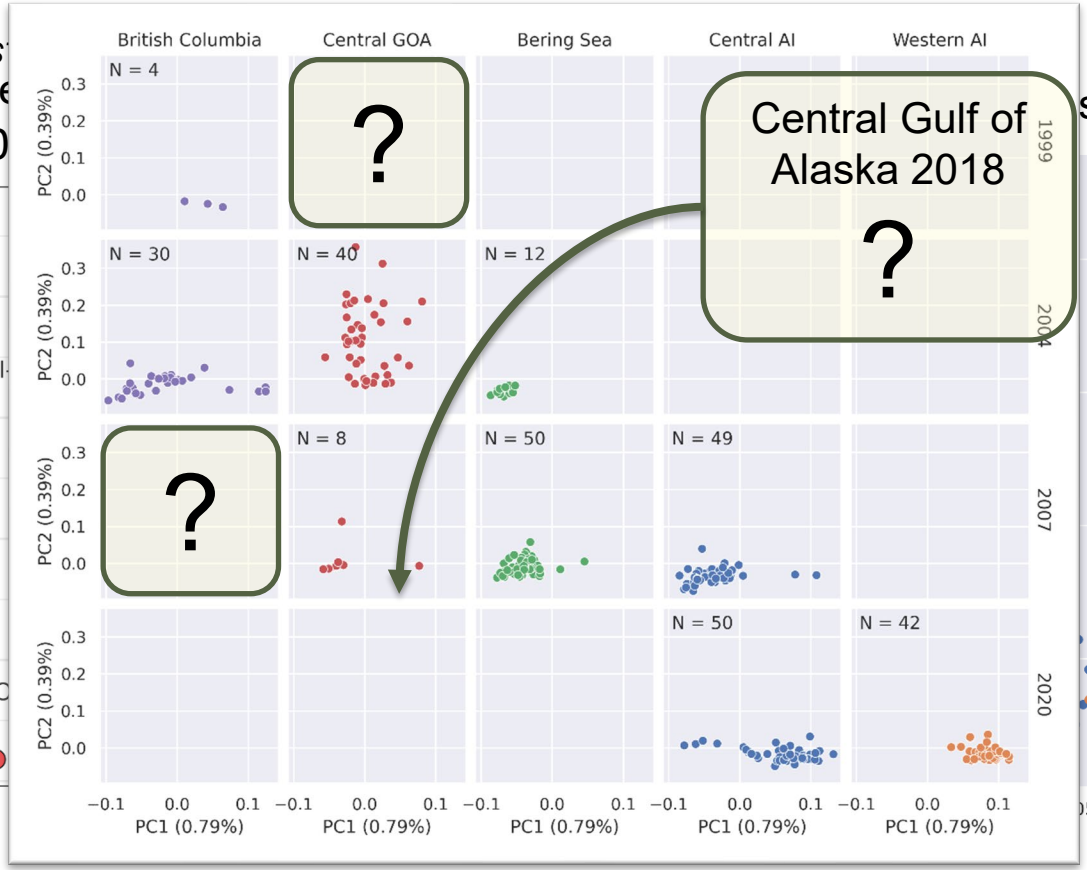
4,850,093 SNPs

Korneliussen, T. S., A. Albrechtsen, and R. Nielsen. 2014. ANGSD: Analysis of Next Generation Sequencing Data. BMC Bioinformatics 15(1):1–13.
Meisner, J., and A. Albrechtsen. 2018. Inferring Population Structure and Admixture Proportions in Low-Depth NGS Data. Genetics 210(2):719–731.

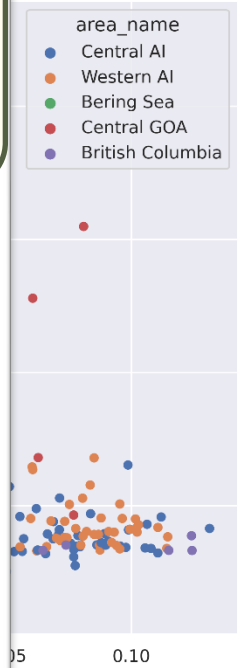
Population Structure (**preliminary*)

Pairwise F_{ST}
Only betwee
min- max: 0





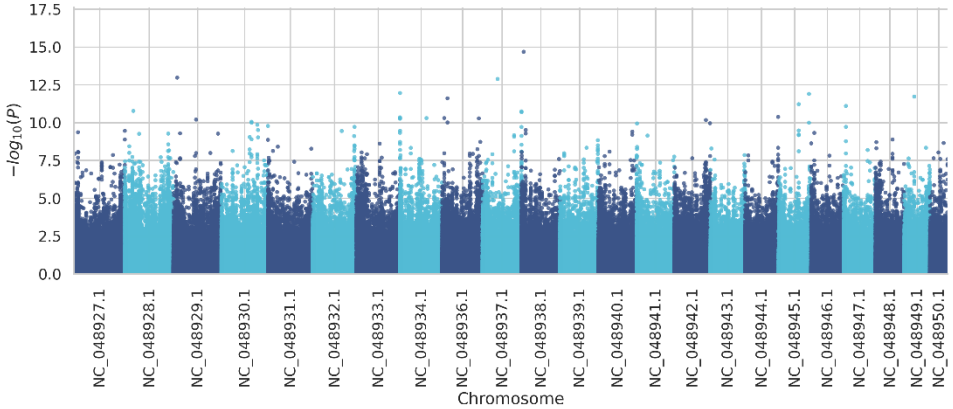
samples)



Signatures of Selection (**preliminary*)

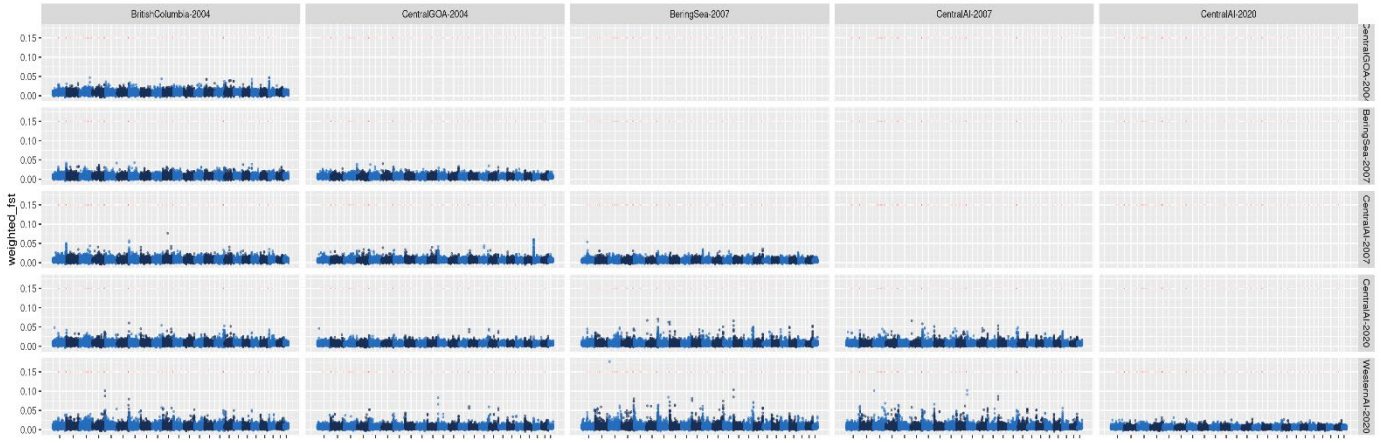
PCAngsd

- Min MAF 0.05
- 4,850,093 SNPs



Pairwise F_{ST}

- 10 kb windows, 5 kb step
- Only between collections where n ≥ 30
- 10,039,557 SNPs



5. Genetics and Genomics

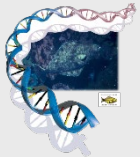
5-Yr Research Plan (2017-2021)

Collection of genetic samples of spawning aggregations spanning the Gulf of Alaska, Bering Sea and Aleutian Islands

(1999-2020)

Development and application of genomic approaches

Chromosome-level genome assembly



Development of methods based on low-coverage whole genome resequencing

Establishment of a bioinformatic pipeline in the cloud (Microsoft Azure)

Population structure analyses

Establishment of a baseline of genetic diversity

Delineation of fine-scale stock structure

Research outcomes:

- Sequenced genome (size=586 Mbp)
- Full annotation (NCBI) (27,422 genes)
- 24 chromosome-length scaffolds
- SNP detection and genotyping

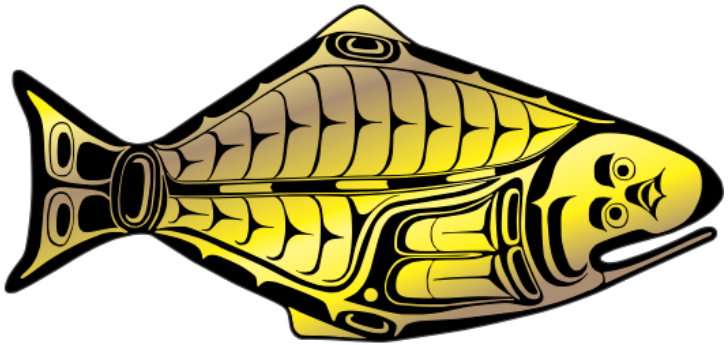
Staff involved: Andy Jasonowicz
Funding: IPHC

Recommendation

That the SRB:

- **NOTE** paper IPHC-2021-SRB019-08 which outlines progress on the IPHC's 5-year Biological and Ecosystem Science Research Plan (2017-21).

INTERNATIONAL PACIFIC



HALIBUT COMMISSION