RESEARCH/PD ANNUAL REPORT - PROGRESS REPORT

2015 annual report - progress Kerry Naish Measuring what matters: linking life history traits and disease resistance with genomic variation in Chinook salmon conservation hatcheries R/HCE-4 Submitted On: 03/16/2016 01:21:27 AM

METRICS & MEASURES

Metric/Measure	Value	Note
Acres of coastal habitat	0	
Fishermen and seafood industry personnel	0	
Communities - economic and environmental development	0	
Stakeholders - sustainable approaches	0	Tribal scientists, Washington Department of Fish and Wildlife and staff at Cle Elum hatchery (approx 20) have learned that the genetic diversity in a segregated line changes faster than in an integrated line. The integrated hatchery population supports a recreational fishery and meets both Tribal and State supplementation goals for conservation and sustainable harvest.
Informal education programs	0	Charlie Waters participated in the Orca Bowl as a science judge. Number of people engaged: 200 (KL NOT COUNTING HERE BECAUSE ALREADY COUNT ORCA BOWL INTERNALLY)
Stakeholders who receive information	1065	Kerry Naish and Charlie Waters organized and participated in a booth "Molecular Genetics in Marine Sciences" at the "Discover Science" weekend, held at the Seattle Aquarium November 13-14 2015. Number of people engaged: 200 Oral presentation to Yakima Basin Science and Management Conference, Ellensburg, Washington, June 17-18, 2015. (Waters) Approximately 300 scientists and managers. Oral presentation to Washington Sea Grant NOAA National Sea Grant site review team in "Restoration session", May 11 2015. (Naish) Approx 15 scientists and program managers. Poster Presentation to Washington Sea Grant NOAA National Sea Grant site review team, poster session, May 11, 2015. (Waters) Approx 50 scientists, managers and administrators. Co- convened a 2.5 day workshop at the American
Stakeholders who receive information	1065	Fisheries Science annual meeting in Portland. Workshop entitled "Interactions between Hatchery and Wild Salmonids – International Understanding of the Risks, Benefits, and Options for Management". (Naish and Warheit). Approximately 500 scientists, about 100 in panel

		discussion. Oral presentation to workshop participants in Interactions between Hatchery and Wild Salmonids – International Understanding of the Risks, Benefits, and Options for Management". (Waters) (DO NOT DOUBLE COUNT - Approx 300 scientists.) Oral presentation to workshop participants in Interactions between Hatchery and Wild Salmonids – International Understanding of the Risks, Benefits, and Options for Management". (Purcell) (DO NOT DOUBLE COUNT - Approx 300 scientists.)
Volunteer hours	0	
P-12 students reached	150	students at Discover Science Weekend at Seattle Aquarium
P-12 educators	0	

REQUESTED INFORMATION

Publications

Effectiveness of managed gene flow in reducing genetic divergence associated with captive breeding

Publication Type: Peer-reviewed: Journals (incl. articles), Books, Proceedings, and Other Documents

Publication Year: 2015

Publication Authors:

Publisher Info: Evolutionary Applications

Notes:

Related URLs: doi: 10.1111/eva.12331

Keywords: captive breeding, domestication selection, genome-wide survey, managed gene flow, supportive breeding

Publication URLs: http://onlinelibrary.wiley.com/doi/10.1111/eva.12331/abstract

Abstract: Captive breeding has the potential to rebuild depressed populations. However, associated genetic changes may decrease restoration success and negatively affect the adaptiv potential of the entire population. Thus, approaches that minimize genetic risks should be tested in a comparative framework over multiple generations. Genetic diversity in two captive-reared lines of a species of conservation interest, Chinook salmon (Oncorhynchus tshawytscha), was surveyed across three generations using genome-wide approaches. Genetic divergence from the source population was minimal in an integrated line, which implemented managed gene flov by using only naturally-born adults as captive broodstock, but significant in a segregated line, which bred only captive-origin individuals. Estimates of effective number of breeders revealed that the rapid divergence observed in the latter was largely attributable to genetic drift. Three independent tests for signatures of adaptive divergence also identified temporal change within the segregated line, possibly indicating domestication selection. The results empirically demonstrate that using managed gene flow for propagating a captive-reared population reduces genetic divergence over the short term compared to one that relies solely on captive-origin parents. These findings complement existing studies of captive breeding, which typically focus on a single management strategy and examine the fitness of one or two generations. Citation: Waters, C. D., J. J. Hard, M. S. O. Brieuc, D. E. Fast, K. I. Warheit, R. S. Waples, C. M. Knudsen, W. J. Bosch, and K. A. Naish. 2015. Effectiveness of managed gene flow in reducing genetic divergence associated with captive breeding. Evolutionary Applications 8:956-971.

Citation for Coverpage: SG can post PDF online?: Yes Uploaded File: Waters_et_al._2015_Published.pdf

Students Supported

Charles Waters (Continuing Student) cwaters8@uw.edu University of Washington, School of Aquatic and Fishery Sciences

Field of Study: Aquatic and Fishery Sciences, Molecular Genetics Advisor: Naish, Kerry Degree Type: PhD Degree Year: 2017

Student Project Title: Reducing Genetic Risks to Wild Populations: Evaluating the Effectiveness of Integrated Management in Captive Breeding Programs

Involvement With Sea Grant This Period (capstone, fellow, intern, etc.): Student Research Assistant

Post-Graduation Plans (employer, grad school, etc.):

Was this thesis/dissertation supported by Sea Grant?: No

Thesis / Dissertation:

New or Continuing?: continuing

Degree awarded this reporting period?: No

Financially supported?: No

Narratives

R/HCE-4 Annual Report 2015 Uploaded File: R_HCE_4_narrative_2015.pdf

Partners This Period

Yakama Nation, Cle Elum Supplementation and Research Facility Types: Government Scale: Tribal Notes:

Washington Department of Fish and Wildlife Types: Government Scale: STATE Notes:

US Geological Survey Types: Government Scale: REGIONAL Notes: Western Fisheries Research Center

STANDARD QUESTIONS

Impacts and Accomplishments

(1)	
Туре	impact
Title	Washington Sea Grant research evaluates ways to minimize genetic changes in hatchery fish to improve salmon harvest sustainability
Relevance	Captive breeding in hatcheries has the potential to rebuild depressed salmon populations. But genetic changes in captive breeding may negatively impact the fitness of released fish in the natural environment, raising concerns for the contribution of hatcheries to salmon restoration. Increasingly, hatcheries along the West Coast have integrated their broodstock with wild fish as a tool for recovering salmon populations, but whether such steps actually prevent divergence in fitness traits is unclear.
Response	Washington Sea Grant-funded researchers used genomic tools to monitor divergence between captive and wild populations. They began to identify specific phenotypic traits that responded to selection in captivity, providing a potential means for monitoring fitness changes when captive individuals are reintroduced into the wild. They tracked these traits linked to fitness and disease resistance over multiple generations and compared genetic divergence between a captive broodstock segregated from wild fish and captive broodstock mixed with wild.
Results	Results confirmed that using wild broodstock reduced genetic change in hatchery populations compared with those that relied solely on hatchery parents. Rapid and significant genetic divergence occurred in the segregated population, largely due to genetic drift. Three independent tests of adaptive divergence also identified short-term change within the segregated group. In contrast, mixing wild broodstock with a hatchery population released to spawn in a wild environment reduced genetic divergence over the short term.
Recap	Washington Sea Grant researchers investigated best practices for managing hatchery populations to reduce genetic change and ultimately aid in restoring declining salmon harvests.
Comments	
Primary Focus Area	Healthy Coastal Ecosystems
Secondary Focus Areas	Sustainable Fisheries and Aquaculture
Goals	Ocean and coastal resources are managed using ecosystem-based approaches. Fisheries are safe, responsibly managed and economically and culturally vibrant.

Partners	Cle Elum Supplementation and Research Facility, Yakama Tribes USGS Western Fisheries Research Center Washington State Department of Fish and Wildlife
PIDraft	* Type accomplishment * Title Evaluation of the effectiveness of different hatchery management practices in reducing divergence in fitness traits in wild salmon populations. * Relevance : There are many situations where supplementation with captive reared fish is a key tool in the recovery of healthy salmon populations. A clear evaluation of the relative risks of different broodstock management strategies on genetic diversity of hatchery and wild fish is clearly warranted. Hatcheries along the West coast have increasingly integrated their broodstock with wild fish, but it is unclear whether such steps prevent domestication and divergence in fitness traits. * Response Washington Sea Grant Researchers are employing genomic tools aimed at monitoring divergence between captive and wild populations. The approaches will identify the specific phenotypic traits that have responded to selection in captivity, providing a means of monitoring any fitness changes when captive individuals are reintroduced into the wild. * Results Initial results have shown that the use of wild broodstock reduces genetic change in a hatchery population compared to hatchery populations that rely on hatchery parents. The research will provide guidance on "best practices" in hatchery management and aid the restoration of declining salmon populations. * Recap Washington Sea Grant researchers are investigating the best approaches to manage hatchery populations in order to reduce genetic change. Comments Primary Focus Area Sustainable Fisheries and Aquaculture Secondary Focus Areas Healthy Coastal Ecosystems Goals Ocean and coastal resources are managed using ecosystem-based approaches.,Aquaculture operations and shellfish harests are safe, environmentally sustainable and support economically prosperous businesses.,Coastal communities engage in comprehensive planning and sustainable development.,The future workforce is skilled in discilines critical to coastal and ocean economies and ecosystem health. Partners University of Washington Yakama Nation
	Fisheries Research Center * Type accomplishment * Title International workshop on interactions between hatchery and wild fishes * Relevance : There has been widespread recognition that hatcheries will persist in the near future for policy, social, and management reasons. Therefore, there have been extensive efforts to reform hatchery practices so that hatcheries can provide a viable tool

for conservation and management. For this goal to be achieved, however, it is important to assess the state of knowledge not only on genetic effects of hatchery fish on wild fish, but also on ecological, physiological, and disease interactions. * Response A workshop was convened at the AFS annual meeting, bringing together experts from several countries, to assess the state of knowledge in the science of hatchery reform and to identify gaps in knowledge that will shape future research directions in this area. * Results Over 55 presentations were given over a 2.5 day symposium, followed by a panel discussion on the state of science and future directions of research. The workshop was attended by key scientists and regional hatchery managers. * Recap Washington Sea Grant researchers co-led efforts to evaluate the state of science in hatchery reform. Comments Primary Focus Area Sustainable Fisheries and Aquaculture Secondary Focus Areas Resilient Communities and Economies.Ocean Literacy and Workforce Development Goals Ocean and coastal resources are managed using ecosystem-based approaches., Coastal communities engage in comprehensive planning and sustainable development., The future workforce is skilled in discilines critical to coastal and ocean economies and ecosystem health. Partners University of Washington Washington Department of Fish and Wildlife NOAA Sustainable Fisheries Division, West Coast Region River Design Group Clearwater BioStudies, Inc. Bangor University, UK

Tools, Technologies, Information Services / Sea Grant Products

(1)	
Description	DNA sequences and genotypes for Chinook salmon.
Developed (in the reporting period)?	Yes
Used (in the reporting period)?	Yes
Used for EBM?	Yes
ELWD product?	No
Number of managers	3
Description/Names of managers	NOAA scientists (Hard and Nichols) WDFW Scientists (Warheit)

(2)	
Description	Bioinformatic pipeline: Methods to process and analyze genomic data from partial sequences in Chinook salmon.

Developed (in the reporting period)?	Yes
Used (in the reporting period)?	Yes
Used for EBM?	Yes
ELWD product?	No
Number of managers	2
Description/Names of managers	NOAA scientists (Hard) WDFW Scientists (Warheit)

Economic Impacts

No Economic Impacts information reported

Community Hazard Resilience

No Community Hazard Resilience information reported

Meetings, Workshops, Presentations

(1)	
Type of Event	Public or professional presentation
Description	Naish KA, Waters CD "Role of hatchery releases in Chinook salmon restoration". Washington Sea Grant Program Review, Seattle, WA, May 11, 2015
Event Date	05-11-2015
Number of Attendees	0

(2)

<u>\</u>	
Type of Event	Public or professional presentation
Description	Waters, C.D, J.J. Hard, M.S.O. Brieuc, D.E. Fast, K.I. Warheit, R.S. Waples C.M. Knudsen, W.J. Bosch, K.A. Naish. "An evaluation of selection across four generations within a Chinook salmon supportive breeding program." Washington Sea Grant Program Review, Seattle, WA, May 11, 2015
Event Date	05-11-2015
Number of Attendees	0

(3)

Type of Event	Public or professional presentation
	Waters, C.D, J.J. Hard, M.S.O. Brieuc, D.E. Fast, K.I. Warheit, R.S. Waples C.M. Knudsen, W.J. Bosch, K.A. Naish. "An evaluation of selection

Description	across four generations within a Chinook salmon supportive breeding program." Yakima Basin Science and Management Conference, Ellensburg, Washington, , June 17-18, 2015
Event Date	06-17-2015
Number of Attendees	300

(4)

17	
Type of Event	Sea Grant-sponsored/organized event
Description	Co-convened a workshop entitled "Interactions between Hatchery and Wild Salmonids – International Understanding of the Risks, Benefits, and Options for Management". (Naish and Warheit). American Fisheries Society Annual Meeting, Portland, Oregon, Aug. 16-20, 2015
Event Date	08-16-2015
Number of Attendees	500

(5)

Type of Event	Public or professional presentation	
Description	Waters, C.D, J.J. Hard, D.E. Fast, K.I. Warheit, C.M. Knudsen, W.J. Bosch, K.A. Naish. "Linking genotype and phenotype: Identifying fitness traits that respond to genetic adaptation to captivity in Chinook salmon." American Fisheries Society Annual Meeting, Portland, Oregon, Aug. 16-20, 2015	
Event Date	08-16-2015	
Number of Attendees	300	

(6)

Type of Event	Public or professional presentation	
Description	Purcell, M.K. Managing infectious diseases in salmon and steelhead hatcheries and natural populations. American Fisheries Society Annual Meeting, Portland, Oregon, Aug. 16-20, 2015.	
Event Date	08-16-2015	
Number of Attendees	0	

(7)
ľ	-	,

Type of Event	Public or professional presentation
	Waters, C.D, J.J. Hard, D.E. Fast, K.I. Warheit, C.M. Knudsen, W.J. Bosch, K.A. Naish. "Linking genotype and phenotype: Identifying fitness traits that respond

Description	to genetic adaptation to captivity in Chinook salmon." 2015. School of Aquatic and Fishery Sciences Graduate Student Symposium, Seattle, Washington, Nov. 19, 2015
Event Date	11-19-2015
Number of Attendees	70

Leveraged Funds

(1)	
Purpose	NA
Source	NA
Amount	NA
Start Date	NA
End Date	NA

Measuring what matters: linking life history traits and disease resistance with genomic variation in Chinook salmon conservation hatcheries, R/HCE-4

Kerry Naish, University of Washington, Kenneth Warheit, Washington Department of Fish and Wildlife, David Fast, Yakama Nation Fisheries, Maureen Purcell and Diane Elliott, Western Fisheries Research Center, USGS

The aim of our project is to identify fitness traits that have responded to genetic adaptation in captivity in a segregated and an integrated hatchery line, so that we can compare rates of genetic change attributable to selection since founding. We will also identify phenotypic traits that have responded to selection, providing a means for monitoring genetic change in fitness traits. These aims will be attained by:

- 1. Linking key fitness traits that have been measured in returning adult Chinook salmon to molecular markers using Genome Wide Association Studies (GWAS) and Random Forest (RF) analyses.
- 2. Measuring hatch timing, early growth rate, and disease resistance in families created within the hatchery lines, and linking these traits to molecular markers.
- 3. Determining which traits have responded to selection in the hatchery environment by linking variation in fitness traits to divergence in genetic markers.

Our activities for each of these tasks within the reporting period are detailed below.

Task 1: We have nearly completed the GWAS and Random Forest analyses to link fitness traits in returning adult Chinook salmon to molecular markers. Utilizing all available phenotypic data, we conducted association analyses for six key fitness traits: return time to the Roza Dam Adult Monitoring Facility, length at Roza, weight at Roza, spawn time at the Cle Elum Supplementation and Research Facility, daily growth coefficient (DGC) between collection at Roza and spawning at Cle Elum, and age at maturation. The Random Forest analysis, developed during previously funded Sea Grant funded research, wasimplemented to complement the GWAS analysis. GWAS tests each marker individually and is most effective for detecting loci of large effect. However, many phenotypic traits may be polygenic and largely influenced by epistatic interactions. Adaptation of

these traits via selection on standing genetic variation is predicted to result in minor allele frequency changes at many loci, or an increased degree of covariance across loci. The Random Forest (RF) algorithm provides a non-parametric framework that can incorporate many loci and account for dominance and epistatic interactions, enabling the identification of suites of loci that explain substantial phenotypic variation collectively may but not display significant changes in allele frequencies individually.

Initial analyses with GWAS and Random Forest have identified loci associated with fitness-related traits (e.g. Figure 1). We will determine whether these loci also exhibit signatures of domestication selection in future analyses.

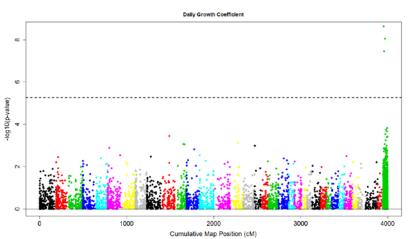


Figure 1. Manhattan plot showing GWAS results for daily growth coefficient (DGC) of adult Chinook between their collection at Roza Dam and spawning at the Cle Elum Supplementation and Research Facility. Each point represents the –log(p-value) for a genetic marker and is color-coded by chromosome. Unmapped loci are denoted in green and grouped on the right side of the plot. The horizontal, dashed line represents the genome-wide significance level. Three unmapped loci displayed significant associations with DGC.

Task 2: We conducted disease challenges using the pathogen *Vibrio anguillarum*, the causative agent of vibriosis, on juvenile Chinook salmon from 56 families (28 from the integrated line and 28

from the segregated line). Two pilot studies were initially conducted on a subset of individuals representing all families to determine the optimal pathogen dose for the main disease challenge. The main challenge comprised two replicates per family (n=112) with 24 fish per tank. Fish were exposed to the pathogen for 30 minutes, and all tanks were monitored for mortality dailv for three weeks. Unfortunately, this family-based design contained too much variation between replicates (i.e. the two tanks containing fish from the same family did not show similar levels of survival: Figure 2). Such variation would make it difficult to accurately identify the genetic basis of resistance and compare the responses of the two hatchery lines. Thus, we decided to conduct another disease challenge using a "pooled design" with fish that were being held in a common tank exactly for this purpose.

Another pilot study on progeny randomly sampled from the common tank was conducted prior to the main challenge to again determine optimal dose. The main challenged comprised three tanks, each with 375 fish, exposed to the optimal pathogen dose for 30 minutes. Mortality was monitored daily for 14 days. Another tank containing 75 fish was exposed only to culture medium lacking bacteria and served as the mock control. All mortalities and survivors were measured and sampled for DNA. This challenge was successful in that all three tanks displayed similar mortality curves (Figure 3). However, since this was a pooled design, we cannot yet compare individual families or the two hatchery lines. In summary, we completed the experimental phase of this project and are now in the process of preparing our samples for DNA sequencing and subsequent analysis.

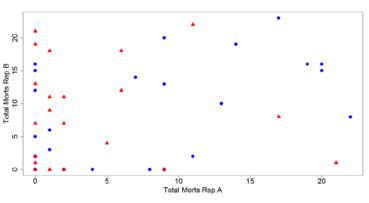


Figure 2. Scatter plot of total number of mortalities (out of 24) between the two replicates per family for the initial main disease challenge. Families from integrated line are denoted with blue circles while those from the segregated hatchery line are shown as red triangles. Replicates do not fall along a straight line, indicating that tank-to-tank variation is high.

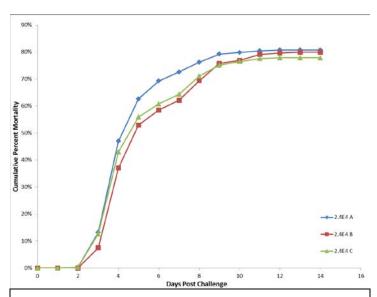
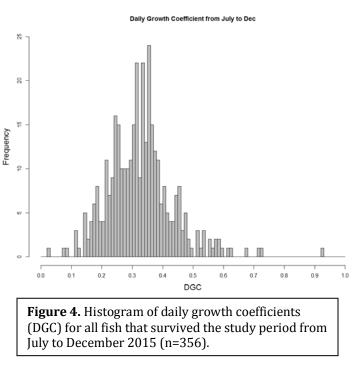


Figure 3. Cumulative percent mortality over the 14-day challenge for the pooled experimental design. The three replicates, denoted in different colors, exhibit similar mortality curves and thus indicate a successful challenge.

Task 3: In 2014, hatch timing was measured at six-hour intervals for 10 individual eggs in each of 76 families (42 integrated families and 34 segregated families) that were created at the beginning

of the experiment. A subset of these individuals (n=480) were then ponded and reared for subsequent monitoring of growth rate. To monitor growth rate, all fish were PIT tagged when they weighed approximately three grams (July 2015). Individual measures for length and weight were taken, and all fish were combined into a single rearing tank. Fish were fed to satiation three times per week, and weight and length for each fish was measured everv eight to ten weeks for approximately five months. Overall, three data points were taken during this experiment before it ended in December 2015. The fish exhibited a wide range of growth rates (Figure 4). The tissues are currently being prepared for DNA sequencing.



Challenges Encountered: In Task 2, we detail the high levels of variation in the first disease challenge. We therefore repeated the challenge.

Changes in Project Direction: None