

## INTEGRATED BROODSTOCK EVALUATION

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## INTEGRATED BROODSTOCK EVALUATION

# **Project Progress Report**

2020 Annual Report

Ву

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To

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## **ABBREVIATIONS AND ACRONYMS**

Acronym	Definition	Acronym	Definition
AD	adipose fin clipped	NP	naturally produced
BY	brood year	PBT	parentage based tagging
C_Se_Se	true offspring_parent_parent relationship	PFH	Pahsimeroi fish hatchery
CWT	coded wire tag	pHOS	proportion hatchery origin spawners
DNA	deoxyribonucleic acid	PIT	passive integrated transponder
EFGL	Eagle Fish Genetics Laboratory	PNI	proportionate natural influence
FINS	fish inventory system	pNOB	proportion natural origin broodstock
FL	fork length	pNOS	proportion natural origin spawners
GIS	geographical information system	PTAGIS	PIT tag Information System
HGMP	Hatchery Genetic Management Plan	QA/QC	quality assurance/quality check
HSRG	Hatchery Scientific Review Group	R/F	recruit per female
IB	integrated broodstock	RF/F	female recruit per female
IDFG	Idaho Department of Fish and Game	RPA	reasonable and prudent alternative
km	kilometer	SAS	smolt to adult survival
LCI	lower confidence interval	SFH	Sawtooth fish hatchery
LGR	Lower Granite Dam	SFSR	South Fork Salmon River
LOD	log of odds	SGS	spawning ground survey
m	meter	SS	segregated broodstock
М	male	SURPH	survival under proportional hazards model
MFH	McCall fish hatchery	SY	spawn year
N	number	USR	Upper Salmon River
NOAA	National Oceanographic and Atmospheric Administration		

#### **ABSTRACT**

The use of integrated broodstocks (IB) allows supplementation of depressed fish populations while minimizing genetic divergence between hatchery and recipient natural stocks. In 2010, IB programs for Chinook Salmon (Oncorhynchus tshawytscha) were initiated at the Sawtooth, Pahsimeroi, and McCall hatcheries. This project is responsible for annual monitoring and evaluation of these programs and implementation of studies to improve their effectiveness. Here we summarize program activities in 2020. Chapter 1: Estimated proportionate natural influence (PNI) upstream of weirs was higher when based on fin clips and tags (i.e., mark) than parentage-based tagging (PBT). Estimated PNIs for the three populations (mark estimate followed by PBT estimate), were 0.97 and 0.83 at Sawtooth, 0.80 and 0.75 at Pahsimeroi, and 0.90 and 0.83 at McCall. Chinook Salmon also spawn downstream of the Sawtooth and McCall hatchery weirs. Composite PNI estimates, which include fish spawning in the up- and downstream portions of the population, were 0.17 in the upper Salmon River and 0.53 in the South Fork Salmon River. Target PNI was achieved upstream of Sawtooth and McCall hatchery weirs, and in the composite population in the South Fork Salmon River. Target PNI levels were not achieved at the other locations. Chapter 2: In 2020, the spawning population upstream of the Sawtooth Hatchery weir was comprised almost entirely of naturally produced (NP) adults. Carcass recovery efficiency for this group was 60.7%. Natural-origin females spawned near the weir, with the median carcass recovery distance 6.3 meters upstream (range 0-23.3 km). Chapter 3: The maximum number of progeny per female in the IB ranged from six at Sawtooth to 20 at McCall. The maximum number of progeny in the NP was five at Sawtooth, one at McCall, and three at Pahsimeroi. The maximum number of female progeny per female in the IB was 11 at Sawtooth and McCall and five at Pahsimeroi. In the NP, maximum number of female progeny per female was one at all three hatcheries. The mean number of female progeny per female in the IB was 0.75 females at Sawtooth, 2.87 females at McCall, and 1.2 females at Pahsimeroi. In the NP the mean number of female progeny per female was 0.11 females at Sawtooth, 0.03 females in the South Fork Salmon River (i.e., McCall population), and 0.068 females at Pahsimeroi.

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#### **OVERVIEW**

There have been widespread declines in Pacific salmonid (*Oncorhynchus* spp.) abundance throughout their range in recent decades (Lichatowich 1999; McClure et al. 2003). In response, extensive hatchery programs were developed to increase abundance and mitigate for lost harvest opportunities. Naturally reproducing populations have remained depressed despite mitigation, and in some locations efforts have been undertaken to use hatchery production to augment them. These efforts are commonly referred to as supplementation. However, debate remains on whether or not supplementation can increase natural-origin abundance (ISAB 2003).

There are a number of uncertainties that remain in terms of the benefits and risks of supplementation. The number of natural-origin adult progeny produced by supplementation adults must exceed the number that would have been produced without hatchery intervention for supplementation to provide a conservation benefit. Supplementation assumes that the natural population is below carrying capacity and that no density dependent effects are operating, which may not be the case (Walters et al. 2013). In addition, a number of genetic and demographic risks have been hypothesized (Goodman 2005; Oosterhout et al. 2005; Bowlby and Gibson 2011). Phenotypic differences in the hatchery broodstock could also arise from spawning natural-origin fish in the hatchery and affect maturation rates and ultimately hatchery production targets or harvest rates (Kostow 2004; Hayes et al. 2013). Differences between natural- and hatchery-origin fish may unintentionally arise from early rearing in a hatchery environment. These differences may reduce the subsequent fitness of the hatchery fish in the natural environment and the overall contribution of naturally spawning hatchery fish (Ford 2002; Araki et al. 2008; Evans et al. 2016). However, these remain uncertainties, and as such, supplementation evaluation continues to be an important and germane area of inquiry.

Supplementation can be implemented in a variety of ways, but all use hatchery production to increase the number of naturally spawning fish in target populations. Supplementation programs have been implemented throughout the Columbia River basin for several decades (ISAB 2003; Venditti et al. 2013; Venditti et al. 2018). Integrated broodstock (IB) programs are a particular type of supplementation strategy where the hatchery broodstock is comprised primarily of natural origin adults in order to minimize genetic divergence between hatchery donor stocks and recipient natural stocks. This is also hypothesized to minimize domestication selection in the hatchery and reduce ecological risks to the natural population (HSRG 2009). In a properly integrated program, the natural environment drives the adaptation and fitness of the composite population of fish spawning in a hatchery and in the wild (HSRG 2009). Integrated broodstock programs have been used to increase abundance in natural populations (Sharma et al. 2006; Berejikian et al. 2008), provide harvest (Fast et al. 2015), minimize straying risks to wild populations (Mobrand et al. 2005), provide genetic repositories (Kline and Flagg 2014), and expand spawning into under-utilized habitats (Dittman et al. 2010; Venditti et al. 2018).

The Idaho Department of Fish and Game (IDFG) currently maintains IB programs for Chinook Salmon (*Oncorhynchus tshawytscha*) at the Sawtooth Fish Hatchery (SFH), Pahsimeroi Fish Hatchery (PFH), and the McCall Fish Hatchery (MFH). All three hatcheries were originally constructed to mitigate for fish and wildlife losses due to hydropower and transportation construction on the Snake River (USACOE 1975; NPCC Undated). The SFH is located on the upper Salmon River near the town of Stanley, Idaho, and all trapping, spawning, and rearing is conducted at one centralized facility. The PFH is located on the Pahsimeroi River near the town of Ellis, Idaho. Adults are trapped and spawned at a facility located near the mouth of the Pahsimeroi River, while fertilized eggs are incubated and juveniles are reared at a second facility approximately 18 km upstream. Adults for the MFH program are trapped and spawned at the

South Fork Salmon River satellite facility near Warm Lake, Idaho. Fertilized eggs are incubated and juveniles are reared at the main hatchery facility in McCall, Idaho. Juveniles are later released upstream of the weir at Knox Bridge.

In 2010, a portion of the mitigation production (hereinafter segregated stock or SS) from these hatcheries along with naturally produced (NP) adults were used to establish the IB programs. During the first years of the program (2010–2012), IB crosses were SS females crossed with NP males to avoid compromising the evaluation phase of the Idaho Supplementation Studies (Venditti et al. 2015). After hatchery spawning, males were immediately passed upstream of the weir and allowed to spawn naturally. In 2013, NP and SS adults of both sexes were crossed in the IB. The IB began using only NP and IB adults in 2014 when females from the initial IB crosses first returned to the weirs. The Sawtooth and McCall hatcheries target a proportion natural origin broodstock (pNOB) of 0.9, while the target at the Pahsimeroi Hatchery is pNOB = 1. The number of IB adults allowed to spawn naturally in all three populations is a predetermined proportion of the number of NP adults passed upstream of the weirs to spawn naturally. This proportion varies as the number of NP spawners increases and is determined via a sliding scale (Appendix A).

In order for the natural environment to drive the adaptation and fitness of the composite population, all IB programs must incorporate and balance pNOB and the proportion of hatcheryorigin fish that are allowed to spawn naturally (pHOS). The goal of this project is to examine several aspects of these components and evaluate the ability of IB programs to maintain or increase naturally spawning populations. This project: 1) assesses the influence of NP Chinook Salmon incorporated into IBs on hatchery survival, productivity, and hatchery replacement rates: 2) evaluates the influence of IB spawners on naturally reproducing populations and natural replacement rates; and 3) attempts to alter spawning distribution into under-utilized habitat through targeted IB smolt releases near these areas. This project conducts critical uncertainty monitoring and evaluation in an empirical setting, as recommended by Waters et al. (2015). We work with other agencies and IDFG cooperators to collect the necessary data and address specific uncertainties related to hatchery effectiveness monitoring and evaluation (i.e., RPAs 63 and 64 in the Federal Columbia River Power System Biological Opinion, NOAA Fisheries 2008). Information from this program will provide managers with a more complete picture of the benefits and risks of implementing IB programs, which may guide the implementation of future supplementation strategies in Idaho and elsewhere. The key monitoring objectives addressed by this project are: 1) what is the proportionate natural influence (PNI) in the supplemented populations, 2) how does in-hatchery survival compare between IB and SS, 3) can we alter the spawning distribution of IB adults across the landscape, and 4) what is the replacement rate of NP and IB spawners. Monitoring objectives 1, 3, and 4 are addressed in separate chapters below. Objective 2 was not addressed during the period covered by this report in order to focus on a comparison of tissue types and preservation techniques on the genotyping success of DNA from Chinook Salmon carcasses (Venditti et al. In Press). Information from this assessment will allow us to maximize the number of samples available to address objective 4.

## **CHAPTER 1: PROPORTIONATE NATURAL INFLUENCE (PNI)**

#### INTRODUCTION

Chinook Salmon (*Oncorhynchus tshawytscha*) supplementation programs using an integrated broodstock (IB) are designed to allow the natural environment to be the primary driver of adaptation and fitness in the portions of the population spawning in the hatchery and naturally (HSRG 2009). To achieve this, the proportions of supplementation-origin and natural-origin fish must be measured and partitioned in the broodstock and natural spawning areas. Ford (2002) demonstrated how gene flow between the two could alter the fitness of the naturally spawning component, and this theory was adapted by the Hatchery Scientific Review Group (HSRG 2009) to develop the proportionate natural influence (PNI; a unit-less measure). For the natural environment to be the primary driver of fitness (i.e., PNI >0.5), the proportion of naturally produced (NP) fish in the broodstock (pNOB) must be greater than the proportion of hatchery origin spawners (pHOS) in the natural environment.

The Hatchery Scientific Research Group (HSRG 2009) established long-term, mean PNI targets for supplemented populations with differing conservation objectives, and PNI is being used to guide supplementation programs throughout the Pacific Northwest (HSRG 2014). The portions of the upper Salmon River (USR) and South Fork Salmon River (SFSR) populations that spawn upstream of the weirs are currently managed for a long-term mean PNI of 0.67. The Pahsimeroi River population is managed for a long-term mean PNI of 0.80.

Estimates of PNI for these locations are not straightforward, because physical marks yield overestimates. While adult capture probabilities are high at these weirs, there is a directional bias in the origin determination of fish trapped based on the differential marking of IB, NP, and segregated stock (SS; i.e., mitigation or general production) adults. Adipose fin clipping is not 100% successful in marking trailers (i.e., missclips) and some fish shed their coded wire tag (CWT) or the tags are not detected. Both will result in hatchery-origin fish being classified as NP. It is not known how substantially these errors may affect PNI estimation, but we do know they will inflate them. In response, we will test the accuracy of physical marks by verifying each fish's origin with parentage-based tagging (PBT, Steele et al. 2013) and then recalculate PNI estimates for each population. Estimating PNI using both physical marks and PBT provides an assessment of how well in-season weir management based on marks represents the population's true PNI as determined with PBT. Producing both estimates will provide information on how the upward bias in the tag-based estimate is distributed. This report contains estimates for BY 2020 along with the previous three years for comparison.

There is also a component of the USR and SFSR populations that spawn downstream of the weirs. The composition of adults spawning in these areas is uncontrolled, so PNI for this segment of the population may differ substantially from the segment spawning upstream of the weirs. We will use a four-population PNI calculator developed by NOAA Fisheries (Craig Busack, NOAA Fisheries, personal communication) to estimate the PNI for the overall (i.e., composite) population. This calculator combines the proportions of NP, IB, and SS adults spawning up- and downstream of the weir and used in the integrated and segregated broodstocks into a single PNI estimate. Our goal is to achieve 10-year average PNI values >0.5 in the composite population, which is lower than our targets for upstream of the weirs due to the uncontrolled escapement in the downstream areas. Again, we report estimates for BY 2020 along with the previous three years for comparison. Origin determination using PBT will generally not be possible for fish spawning downstream of weirs, so origin will be determined using marks and tags from carcasses collected in these areas.

#### **METHODS**

## **PNI Upstream of Weirs**

The information needed to assign an origin to each Chinook Salmon used in PNI estimation was collected immediately prior to the fish being passed upstream of the weir or spawned in the hatchery. At these times, hatchery personnel recorded pertinent mark and tag information and collected fin tissue for PBT analysis. Mark and tag data were linked to the fin tissue via common sample number. Hatchery personnel entered the mark and tag information into the Fish Inventory System database (FINS; <a href="www.finsnet.org">www.finsnet.org</a>). These data were later used to assign an origin to each fish. Tissue samples were genotyped at the Eagle Fish Genetics Laboratory (EFGL) and the output stored in a Progeny database maintained at the EFGL. The genetic origin of each fish was then determined for each fish based on parentage assignment.

The FINS "Final Disposition" query identified fish (and their associated tissue samples) that were passed for natural spawning and data queried from the Progeny database identified those used in broodstocks. We used the Progeny data to determine those individuals used in the broodstocks, since culls were identified in this dataset. Culls were not included in PNI estimates, because they did not contribute any juveniles to the next generation. Origin was determined in the FINS data for each fish based on marks and tags by filtering the data for the following combinations. Natural-origin fish had an intact adipose fin and no CWT. Fish from the IB had an intact adipose fin and a CWT, and those with an adipose fin clip were from the SS. Once summarized, those data were used in the general PNI approximation

where pNOB is the proportion of NP adults in the broodstock and pHOS is the proportion of hatchery-origin adults (IB + SS) spawning naturally upstream of the weir.

We calculated the PBT-adjusted PNI by genotyping the DNA associated with each tissue sample collected from fish passed or spawned to determine its parental lineage using standard PBT methods (Steele et al. 2013; Steele et al. 2018a). All fish whose parents were identified had their mark/tag-based origin verified or updated as necessary based on the known parental crosses. Fish whose origins were not determined through PBT retained their mark/tag combination origin classification. After updating each fish's origin, PNI was recalculated using the methods described above.

## **Composite Population PNI and Escapement**

In the USR, the size and composition of the portion of the population spawning downstream of the weir was estimated through multiple spawning ground surveys (SGS). Project personnel conducted carcass surveys twice weekly from the Sawtooth Hatchery weir to the mouth of Redfish Lake Creek. All carcasses encountered were processed using standard IDFG protocols (Copeland et al. 2019). We recorded fork length (FL), sex, presence of CWT and/or PIT tags, GPS location, and any other fin clips, tags, or marks. We also estimated percent spawned (for females) in 25% increments ranging from zero (no eggs deposited) to 100 (all or nearly all eggs deposited). A female carcass with a percent spawned estimate of zero was considered a prespawn mortality. All NP carcasses were tissue sampled for parentage analysis, and dorsal fins were collected from up to 100 of these carcasses to estimate the age composition of the

population (by the Natural Production Monitoring and Evaluation program [NPM]; Project Number 1991-073-00). Redd counts were conducted in this reach by NPM program personnel on three or more occasions throughout the spawning period depending on escapement levels.

Carcass and redd count data collected between the Sawtooth weir and Redfish Lake Creek were entered into the IDFG SGS database and used to estimate spawner escapement by origin into the reach. We assumed the proportions of NP, IB, and SS carcasses collected (based on marks and tags) accurately represented the composition of fish spawning in this reach. To estimate escapement, we assumed that each female made one redd (Neilson and Bradford 1983; Murdoch et al. 2009a), and that redd counts accurately reflected the number of females that spawned. The number of redds constructed by females of each origin was then estimated by multiplying the total number of redds observed by the proportion of females of each origin type in our carcass collection that had spawned (i.e., did not have a percent spawned estimate of zero). Finally, these estimates were expanded (2.31 fish/redd; Beamesderfer et al. 1997) and summed to estimate the total adult escapement of NP, IB, and SS in the reach.

Origin-specific escapement estimates along with the numbers of fish (based on marks and tags) used to estimate PNI upstream of Sawtooth weir were converted to proportions and entered into the NOAA Fisheries four-population calculator to estimate PNI for the composite population. However, the calculator requires estimates of the proportion of NP fish that originated (i.e., hatched) up- and downstream of the weir that ultimately spawned up- or downstream of the weir or were used in the IB. We used PBT data to inform these values. In the USR, tissue samples collected from NP carcasses downstream of the weir and from NP adults passed for natural spawning or spawned in the IB provide estimates of these proportions. Those that originated upstream of the weir but spawned downstream are identified as those NP carcasses collected downstream of the weir that received a PBT assignment to two known parents passed upstream in the previous generation. Adults that originated downstream of the weir but spawned upstream or were spawned in the IB were assumed to be those NP adults that entered the weir trap but did not receive a PBT assignment to parents passed upstream in the previous generation. Finally, calculated proportions of NP based on PBT data are applied to the numbers of NP based on mark. We assumed contribution by un-sampled parents (or precocious parr) and natural origin strays into the USR were negligible. We also assumed that samples that failed to genotype were random with respect to origin.

Carcass and redd count data for Chinook Salmon spawning downstream of the SFSR weir were provided by fisheries biologists from the Nez Perce Tribe. Redd counts were expanded to estimate the size of the spawning population, parsed to origin, and entered into the PNI calculator as described above. Beginning in 2020, DNA samples from NP carcasses were also provided by tribal cooperators for PBT analysis. This will allow us to estimate the interchange between upstream and downstream spawning portions of the population as described above.

#### RESULTS

## **PNI Upstream of Weirs**

Estimated PNI values for the portion of Chinook Salmon populations spawning upstream of weirs in 2020 were higher when based on marks compared to estimates using PBT-adjusted origin information. Origin determination based on mark consistently overestimated the proportion of natural fish passed or spawned in the IB (Table 1). In 2020, estimated PNI based on marks ranged from 0.81 at PFH to 0.97 at SFH, while PNI based on origin-corrected PBT data ranged

from 0.75 at PFH to 0.83 at SFH and MFH (Table 1). In 2020, we achieved the target PNI goal, based on mark, at all three locations. We also achieved the PNI goal, based on PBT, at the SFH and MFH, although this was achieved by passing very few IB (intentional) or SS (unintentional) fish to supplement natural spawning. We did not achieve target PNI goals at the PFH using PBT corrected data, but IB supplementation and unintentional releases of SS adults each provided about 15% of the naturally spawning population (Table 1).

Table 1. Estimated proportionate natural influence (PNI) values over the most recent four year period for Chinook Salmon populations spawning upstream of hatcheries with integrated broodstock programs. Estimates are calculated using origins determined by marks and by parentage-based tagging (PBT). Abbreviations include IB = integrated, SS = segregated, pNOB = proportion natural-origin broodstock, pNOS = proportion natural-origin spawners, pHOS = proportion hatchery-origin spawners.

				Abo	ove Weir PNI		
Hatchery	Year	Method	pNOB (IB)	pNOS	pHOS (IB)	pHOS (SS)	PNI
Sawtooth	2020	Mark	0.76	0.98	0.02	0.00	0.97
		PBT	0.71	0.85	0.05	0.10	0.83
	2019	Mark	0.48	1.00	0.00	0.00	1.00
		PBT	0.33	0.87	0.01	0.12	0.72
	2018	Mark	0.41	0.33	0.01	0.66	0.38
		PBT	0.26	0.19	0.04	0.77	0.25
	2017	Mark	0.29	0.2	0.15	0.65	0.26
		PBT	0.20	0.09	0.16	0.75	0.18
Pahsimeroi	2020	Mark	1.00	0.75	0.22	0.03	0.80
		PBT	0.91	0.69	0.17	0.14	0.75
	2019	Mark	0.61	0.56	0.28	0.16	0.58
		PBT	0.54	0.52	0.30	0.19	0.53
	2018	Mark	0.53	0.21	0.21	0.58	0.40
		PBT	0.50	0.22	0.18	0.61	0.39
	2017	Mark	0.66	0.28	0.33	0.39	0.48
		PBT	0.46	0.17	0.33	0.50	0.36
McCall	2020	Mark	0.65	0.93	0.07	0.00	0.90
		PBT	0.59	0.88	0.09	0.03	0.83
	2019	Mark	0.36	0.26	0.74	0.00	0.32
		PBT	0.08	0.15	0.75	0.10	0.09
	2018	Mark	0.64	0.23	0.77	0.00	0.46
		PBT	0.38	0.18	0.79	0.03	0.32
	2017	Mark	0.36	0.16	0.84	0.00	0.30
		PBT	0.20	0.10	0.85	0.06	0.18

Long-term average PNIs (2014–2020) for those portions of the populations spawning upstream of the weirs also remain below target levels, primarily due to low NP escapement. Over this period, PNI averaged 0.58 (mark) and 0.46 (PBT) at SFH, 0.68 (mark) and 0.56 (PBT) at PFH, and 0.51 (mark) and 0.39 (PBT) at MFH. Natural-origin adult returns were insufficient to achieve targeted pNOB levels (90% at SFH and MFH and 100% at PFH) in most years, and this negatively affected our ability to achieve the PNI targets on annual or long-term timeframes. The known directional bias of using marks and tags to determine origin further exacerbated this situation.

Escapement to all three facilities was low in 2020 and contributed to the annual and long-term PNI levels observed. The escapement of NP adults was not sufficient to meet pNOB objectives at the SFH and MFH, although the PFH did achieve a pNOB = 1 (based on mark and 0.91 PBT). At the SFH and MFH, IB escapement was insufficient to meet our supplementation objectives. Only two IB females and two IB males were passed upstream of the SFH weir for natural spawning. No IB females and 15 IB males were passed for natural spawning at the MFH weir. The PFH was the one relatively bright spot, with IB females making up 24% (by PBT) of all females released for natural spawning.

## Composite Population PNI and Escapement

Chinook Salmon carcasses were collected from spawning areas in the upper Salmon and South Fork Salmon rivers downstream of the SFH and MFH weirs. In the upper Salmon River, carcasses were collected twice weekly from August 18 to September 25, 2020. During this time, we collected 236 carcasses of known origin including 8 IB, 144 NP, and 84 SS (Table 2). Carcasses were collected from the SFSR between August 20, 2020 and September 15, 2020. Twenty-eight carcasses were collected, including 0 IB, 25 NP, and 3 SS. (D. Nelson, Nez Perce Tribe, personal communication; Table 2).

Table 2. Number of Chinook Salmon carcasses summarized by origin (IB = integrated, NP = natural, and SS = segregated) and sex collected from spawning areas in the upper Salmon and South Fork Salmon (SFSR) rivers downstream of the Sawtooth and McCall fish hatchery weirs during the most recent four years.

			Origin							
Population	Year	Sex	ΙB	NP	SS	Total				
<b>Upper Salmon</b>	2020	Female	4	79	55	138				
		Male	4	65	28	97				
		Unknown	0	0	1	1				
		Total	8	144	84	236				
	2019	Female	1	12	35	48				
		Male	0	8	12	20				
		Unknown	0	0	0	0				
		Total	1	20	47	68				
	2018	Female	2	20	67	89				
		Male	3	15	18	36				
		Unknown	0	2	3	5				
		Total	5	37	88	130				
	2017	Female	6	32	93	131				
		Male	6	27	72	105				
		Unknown	0	0	0	0				
		Total	12	59	165	236				
SFSR	2020	Female	0	10	3	13				
		Male	0	15	0	15				
		Unknown	0	0	0	0				
		Total	0	25	3	28				
	2019	Female	2	12	35	49				
		Male	0	8	12	20				
		Unknown	0	0	0	0				
		Total	2	20	47	69				
	2018	Female	2	11	17	30				
		Male	2	9	10	21				
		Unknown	0	0	0	0				
		Total	4	20	27	51				
	2017	Female	1	2	2	5				
		Male	0	2	1	3				
		Unknown	0	0	0	0				
		Total	1	4	3	8				

Target PNI is >0.5 for the SFSR and USR composite populations. In 2020, the target was met in the SFSR, where the composite PNI was 0.53 (Table 3). However, composite PNI of 0.17 in the USR was well below this threshold in 2020 (Table 3). The low PNI value in the USR was primarily driven by high proportions of SS spawning downstream of weirs along with low numbers of natural-origin adults passed upstream of weirs and included in the IB broodstock. For the duration of the record (2014–2020), composite PNI estimates averaged 0.15 in the USR and 0.37 in the SFSR. Estimated PNI upstream of both weirs were higher than composite PNI estimates, demonstrating the fact that in-season weir management can lead to higher PNI levels (Tables 1 and 3). Note that caution must be used when comparing the upstream and composite PNI estimates due to the different methods used in their calculation (i.e., the general approximation versus the NOAA multiple population calculator).

Table 3. Estimated composite proportionate natural influence (PNI 2) over the most recent four-year period for composite Chinook Salmon populations in the upper Salmon and South Fork Salmon (SFSR) rivers. Estimates are derived from the proportion of natural-origin spawners (pNOS), and hatchery origin spawners (pHOS) from integrated (IB) and segregated (SS) origins determined by marks and tags from carcasses collected downstream of weirs. The pNOS that originated upstream (US) of the weirs but spawned downstream (DS) of the weirs was assumed to be 1% until 2018, when we began directly estimating these values.

			Downstream Segment								
Population	Year	Method	pNOS (US)	pNOS (DS)	pHOS (IB)	pHOS (SS)	PNI 2				
<b>Upper Salmon</b>	2020	Mark	0.01	0.60	0.03	0.36	0.17				
	2019	Mark	0.09	0.21	0.01	0.69	0.14				
	2018	Mark	0.05	0.24	0.04	0.68	0.07				
	2017	Mark	0.01	0.24	0.05	0.70	0.07				
SFSR	2020	Mark	0.50	0.39	0.00	0.11	0.53				
	2019	Mark	0.09	0.09	0.09	0.74	0.13				
	2018	Mark	0.12	0.27	0.08	0.53	0.13				
	2017	Mark	0.01	0.49	0.13	0.38	0.27				

There appears to be more interchange between spawning sites up- and downstream of the SFH and SFSR weirs than previously assumed, and this interchange is predominantly in one direction. In 2020, PBT records showed that 126 of 235 NP adults (53.6%) collected at the SFH weir and 72 of 151 NP adults (47.7%) collected at the SFSR weir did not assign to parents passed upstream previously, and likely originated downstream. Conversely, three of 144 NP carcasses (2.1%) collected downstream of the SFH weir and none of the 25 NP carcasses collected downstream of the SFSR weir received PBT assignments to adults passed upstream in the previous generation.

Table 4. Selected population parameters for Chinook Salmon spawning in the upper Salmon and South Fork Salmon (SFSR) rivers downstream of hatchery weirs for the most recent four years. Total redd counts are parsed into the number and proportion of redds (pRedds) constructed by females of integrated (IB), natural (NP), and segregated (SS) origin. Female prespawn mortality [PSM (F)] is the proportion of female carcasses with intact egg skeins. Redd counts are expanded using Beamesderfer et al. (1997) to estimate escapement by origin and summed for total escapement. NE indicates no estimate was calculated.

		Tot.		PSM			
<b>Population</b>	Year	Redd	Origin	(F)	Redds	pRedds	Escapement
Upper Salmon	2020	186	IB	0.000	6	0.032	10
			NP	0.063	106	0.570	193
			SS	0.055	74	0.398	135
			Total				339
	2019	73	IB	0.000	2	0.027	3
			NP	0.000	19	0.260	35
			SS	0.086	52	0.712	94
			Total				133
	2018	104	IB	0.000	2	0.019	5
			NP	0.050	24	0.231	43
			SS	0.060	78	0.750	142
			Total				189
	2017	151	IB	0.167	7	0.046	12
			NP	0.063	40	0.265	73
			SS	0.161	104	0.689	190
			Total				275
SFSR	2020	95	IB	NE	0	0	0
			NP	0	73	0.768	133
			SS	0	22	0.232	40
			Total				173
	2019	63	IB	0.000	9	0.143	16
			NP	0.000	9	0.143	16
			SS	0.000	45	0.714	82
			Total				115
	2018	112	IB	0.000	8	0.071	14
			NP	0.000	42	0.375	77
			SS	0.059	62	0.554	112
			Total				204
	2017	64	IB	0.000	16	0.250	29
			NP	0.000	32	0.500	58
			SS	0.500	16	0.250	29
			Total				116

Escapement estimates for the spawning reaches in the USR and SFSR downstream of hatchery weirs indicate Chinook Salmon numbers increased in 2020. In the USR population, we estimated 339 Chinook Salmon were present downstream of the weir, and in the SFSR, we estimated 173 Chinook Salmon downstream of the weir. Natural origin adults were the most abundant origin type, and IB adults were the least abundant (Table 4).

In 2020, female prespawn mortality downstream of hatchery weirs was low in both populations across origins. In the USR, prespawn mortality was about 6% for SS and NP females. We found no prespawn mortality in IB females. In the SFSR, no prespawn mortality was observed (Table 4). Overall, female prespawn mortality in 2020 was consistent with levels observed in recent years (Table 4).

## **DISCUSSION**

## **PNI Upstream of Weirs**

Sliding scales incorporated into the hatchery genetic management plan (HGMP) for each facility dictate the distribution of NP and IB adults between the broodstock and natural spawning, and are designed to achieve target PNIs over a range of NP returns. During years of low NP returns, additional IB or SS adults may be passed upstream of weirs or spawned in the broodstock. PNI targets have only been infrequently met using the high biased mark data and only in 2020 have targets been met using PBT corrected data (Venditti et al. 2020a, 2020b). This has resulted in long-term PNI levels below program goals. In 2020, escapement was low for all origins, which made PNI interpretation difficult. Across facilities pNOB and pHOS levels were generally higher than in previous years, and this led to PNI values close to or exceeding program targets. However, this apparent success was due to the fact that essentially no IB adults were allowed to spawn naturally in the USR or SFSR. We met our PNI goal but failed to supplement natural spawning. When NP and IB returns do increase, populations will need to be managed for substantially higher PNIs to bring the long-term averages up to the 0.67 (SFH and MFH) and 0.81 (PFH) targets.

Mark based estimates of PNI were higher than estimates using PBT corrected data in 2020 and will likely continue to be higher in the future, because mark errors predominantly underestimate the proportion of hatchery-origin fish (IB and SS). Segregated juveniles that are missed during adipose fin clipping and IB juveniles that shed their CWT (or the tag is not detected) are assumed to be natural-origin. Natural-origin adults with small or disfigured adipose fins could be misidentified as segregated or those with undetected hooks in their mouths could be misidentified as integrated, although these appear to occur only rarely. Hatcheries have modified their CWT scanning protocols, which has improved CWT detection rates. However, missed adipose clips and shed (or undetected) CWTs will still result in NP overestimation. Due to this, PNI based on marks will be biased high.

Despite this known bias, there is utility in maintaining the mark and PBT corrected PNI estimates. Origin determination for in-season weir management (i.e., which fish get passed or ponded and to which group they are counted) must be based on mark, as real-time PBT monitoring is not practical from a logistical perspective. Over time, the magnitude of this bias can be estimated and the sliding scales adjusted to yield PBT corrected PNI values in line with program goals.

## Composite Population PNI and Escapement

Composite population PNI has been consistently lower than the target minimum. However, composite PNI in the SFSR in 2020 was higher than the 0.50 target. This was primarily due to exceptionally low SS and IB returns to this system. In order to routinely achieve this target, the proportion of NP spawners must increase, the proportion of SS spawners must decrease, or a combination of both. However, given the uncontrolled nature of fish using downstream spawning areas, little can be done to increase NP density. All of the biases described above also hold for composite PNI estimation, so estimates based on marks will also be biased high in composite populations. Therefore, composite PNI values for the upper Salmon and South Fork Salmon rivers should be treated as upper bound estimates due to these biases. PNI estimates for composite populations are not PBT corrected because a large proportion of the carcasses in these reaches are not sampled for DNA, so PBT data are not available for those individuals.

Selectively removing SS adults on a reach scale would be difficult, expensive, and time consuming. However, we have had some success seining immediately below the SFH weir. In 2020, we collected 219 Chinook Salmon in this manner, of which 141 were SS. Based on Table 3, this suggests we collected about half of the SS adults in the reach between the SFH weir and Redfish Lake Creek. Much of the justification for this effort was to help SFH meet its broodstock need. It is doubtful there will be support for this activity when runs improve and the hatchery can meet broodstock needs with fish that volitionally enter the hatchery trap. Furthermore, this situation is unique in that it is the only program location where a high concentration of adults hold in an easily accessible location. Relatively few IB adults have been shown to spawn downstream of the weirs, so they have little effect on the composite PNI levels. However, if programs can be implemented to encourage the remaining IB fish to recruit to the weirs (e.g., Chapter 2; this report) they should be explored.

The magnitude and prevailing direction of interchange between spawning areas up- and downstream of the SFH and SFSR weirs appears to be much different than originally assumed. Our original 1% estimate of interchange was lower than actually occurs. We will continue to monitor interchange around the weirs to determine if the direction of this interchange remains consistently in a direction from upstream to downstream of the weirs. For these natural-origin populations, the spawning aggregates immediately downstream of weirs may not be sinks on upstream spawning locations created by short-stopping NP adults. In fact, aggregates of NP adults spawning downstream of weirs may be sources of individuals supporting the upstream aggregates. However, SS adults short stopping below weirs are certainly a sink for hatchery programs, particularly in low return years.

# CHAPTER 2: SPAWNING DISTRIBUTION UPSTREAM OF THE SAWTOOTH HATCHERY WEIR

## **INTRODUCTION**

Chinook Salmon (*Oncorhynchus tshawytscha*) spawning distribution in the upper Salmon River (USR) and tributaries upstream of the Sawtooth Hatchery (SFH) is numerically and spatially reduced relative to historical accounts. Evermann and Meek (1898) reported observing almost 1,000 Chinook Salmon in this area. More recently, Idaho Department of Fish and Game (IDFG) trend redd counts from 1957–1978 averaged 650 redds in the USR upstream of Redfish Lake Creek with an additional 89 redds in tributaries. However, from 1979–2017 average redd counts declined to 174 in the main river and eight redds in tributaries (Felts et al. 2019). Recent redd surveys in this area indicate spawning density is highest near the SFH weir, and relatively evenly distributed at lower densities upstream to the mouth of Alturas Lake Creek.

Hatchery supplementation using an integrated broodstock (IB) is currently being used in an attempt to increase the naturally spawning population above the SFH weir. Similar to a number of other investigations (Hoffnagle et al. 2008; Williamson et al. 2010; Cram et al. 2013), we observe IB adults in the USR tend to spawn near the weir (where they were released as smolts). This localized distribution leaves high quality and historically important spawning habitat further upstream underutilized. When adults fail to pioneer beyond their release site, the high, localized spawning density can result in reduced population productivity due to redd superimposition, competition, and fish spawning in areas of poorer habitat (Williamson et al. 2010). Currently, about 80% of the carcasses recovered upstream of the SFH weir are recovered on the weir itself, and essentially 100% are recovered within 10 km of the weir.

The goal of this evaluation is to determine whether releasing IB smolts near the upper extent of current spawning (≈17.7 km upstream of the SFH weir) can alter the distribution of natural spawning upstream in subsequent generations. When IB smolts released at the upstream site return as adults, they should home to the vicinity of their release to spawn naturally. Subsequently, their progeny should also return to these areas (i.e., natural-origin spawning distribution would be altered two generations after the initial smolt release). The new release site is intended to promote increased spawning in areas that are currently underutilized, which may increase population stability by broadening spawning distribution (Probst et al. 2003; Thorson et al. 2014). This shift in spawning distribution would also decrease density near the SFH weir, which may reduce density dependent effects on population productivity (Walters et al. 2013). The objectives of this study are to quantify carcass collection efficiency and document the spawning distribution of Chinook Salmon upstream of the SFH weir. The distribution of carcasses, by origin, collected during the first five years (2016–2020) of this evaluation will represent the baseline spawning distribution. When adult females begin returning from the first upstream releases of IB smolts in 2021, the evaluation will enter the response phase.

#### **METHODS**

We conducted carcass surveys annually during August and September in the USR and tributaries upstream of the SFH weir to determine the spawning distribution of IB, naturally produced (NP), and segregated stock (SS) Chinook Salmon. The spawning distribution response to upstream releases will be evaluated by comparing the median distance upstream of the weir the different origins spawn in the response phase relative to the baseline phase using a Mann-Whitney test, similar to Trojano et al. (2012).

Biological data collected from carcasses followed standard IDFG SGS protocols (Copeland et al. 2019). We recorded sex, fork length (FL), GPS location, percent spawned (for females), and the presence or absence of marks (e.g., fin clips and operculum punches) and/or tags (e.g., PIT and CWT) from all carcasses. Origin was determined from the mark/tag combination (Chapter 1; this report). Additionally, we collected tissue samples from carcasses without operculum punches, since these fish were not sampled at the weir. The ratio of unpunched to punched carcasses provides an estimate of weir efficiency. Tissue samples from unpunched carcasses will provide parentage and origin information when genotyped (see Chapter 3; this report) by reducing the number of un-sampled potential parents in the naturally spawning portion of the population. Tissue samples were fin clips preserved on paper and stored in coin envelopes (fin on paper) following the handling protocols described in Venditti et al. (In Press). All carcass data have been uploaded to the IDFG SGS Database.

We collected carcasses using a combination of kayak and foot surveys throughout the spawning period. Additionally, carcasses were removed from the weir daily and processed by program personnel or personnel from the Natural Production Monitoring project (NPM; Project Number 1991–073–00). The section of the USR from the SFH weir to the County Line Bridge (approximately 21 km) was surveyed by inflatable kayak once a week between mid-August and the end of September. Ground surveys were used to collect carcasses from the upper and lower portions of the roadless 12.5 km river section between the County Line Bridge and the mouth of Beaver Creek. Surveyors walked upstream from the bridge or downstream from Beaver Creek as far as time allowed within a sampling day. We also walked the lower 3.2 km of Alturas Lake Creek (from the bridge on the Cabin Creek road to the mouth). Our goal was to conduct ground surveys in these sections at least twice during the peak spawning period. Additional opportunistic carcass surveys were conducted in the upper Salmon River or Alturas Lake Creek upstream of the previously described sections by program personnel as time allowed, or by other IDFG programs conducting Chinook Salmon spawning ground surveys in these areas.

Our first analysis was to estimate how effective our carcass collections were. All fish passed upstream of the SFH weir for natural spawning were marked with an operculum punch. Overall collection efficiency was estimated as the number of marked carcasses collected divided by the number passed. Collection efficiency by origin was also estimated as the number of marked carcasses collected by origin divided by the number of that origin passed. Fish release information was downloaded from the FINS database (www.finsnet.org).

We then estimated the median distance upstream of the SFH weir that carcasses were collected by origin. We provide separate estimates for all carcasses (sexes combined) and for only female carcasses. Estimates for both sexes provide a measure of how well fish of different origins are distributed throughout the habitat. Estimates for females will be used to assess the effect of upstream releases once the program enters the response phase. We collected a GPS location for each carcass encountered (WGS84 datum). A point on the SFH weir was set as distance zero and a line from that point upstream along the thalweg was plotted for the Salmon River and tributaries using a geographical information system (GIS). The distance to each carcass was measured from the weir along the thalweg to where a perpendicular line through the thalweg passed through the carcass waypoint. Distances up tributaries were computed using the same technique and summed with the distance from the tributary mouth to the weir.

#### **RESULTS**

Chinook Salmon escapement to the upper Salmon River was low in 2020, so few adults were available for release. In total, 32 female and 155 male NP adults along with two IB females and two IB males were passed upstream of the weir for natural spawning. Segregated stock escapement was also insufficient to meet mitigation program broodstock needs, so none were intentionally released (Table 5).

Table 5. The number of marked (M) and unmarked (U) carcasses, by sex (Unk. = undetermined) and origin, recovered upstream of the Sawtooth Hatchery weir in the latest four years. Origins include integrated (IB), natural (NP), and segregated (SS). The sum of marked carcasses recovered divided by the total number released provides an estimate of collection efficiency (Eff.) overall and by origin. NE signifies no estimate was made.

		Relea	sed		Recovered								
		Female	Male	Fen	nale	Ma	ale	Ur	nk.	Su	m	Total	Eff.
Year	Origin			M	U	M	U	M	U	M	U		
2020	IB	2	2	1	0	2	0	0	0	3	0	3	0.750
	NP	32	155	15	2	90	2	6	0	111	4	115	0.594
	SS	0	0	0	0	2	0	0	0	2	0	2	NE
	Total	34	157	16	2	94	2	6	0	116	4	120	0.607
2019	IB	0	0	0	0	1	0	0	0	1	0	1	NE
	NP	23	55	17	1	24	0	0	0	41	1	42	0.538a
	SS	0	0	0	0	0	0	0	0	0	0	0	NE
	Total	23	55	17	1	25	0	0	0	42	1	43	0.538
2018	IB	0	2	0	0	1	0	0	0	1	0	1	0.500
	NP	11	39	5	0	22	0	0	0	27	0	27	0.540
	SS	50	50	22	0	45	1	0	0	67	1	68	0.670
	Total	61	91	27	0	68	1	0	0	95	1	96	0.625
2017	IB	18	27	5	1	18	5	0	0	23	6	29	0.511
	NP	11	51	3	6	23	15	0	0	26	21	47	0.419
	SS	100	100	57	19	63	50	2	0	122	69	191	0.610
	Total	129	178	65	26	104	70	2	0	171	96	267	0.557

<sup>&</sup>lt;sup>a</sup> For collection efficiency estimation, the integrated male was included in the NP group, since it was classified as NP at release.

We conducted multiple carcass surveys upstream of the SFH weir in 2020 to determine the longitudinal distribution of spawners. We surveyed the USR from the County Line Bridge to the SFH weir weekly, between August 19 and September 23, 2020. We surveyed approximately 5 km upstream from the County Line Bridge three times during the spawning period. We surveyed Alturas Lake Creek from the FS-207 (Cabin Creek) road bridge to the mouth (≈5.15 km) once, and from the FS-208 road bridge downstream to the mouth (≈2.75 km) once. Additionally, on the days we surveyed the lower Alturas Lake Creek section (FS-208 road bridge) we also walked the Salmon River from the mouth of Beaver Creek downstream approximately 2 km. In 2020,

personnel from the Natural Production Monitoring project (Project Number 1991–073–00) surveyed Alturas Lake Creek from Alpine Creek (upstream of Alturas Lake) to the mouth and the Salmon River from Frenchman Creek to Pole Creek once.

We collected 120 carcasses upstream of the SFH weir in 2020. All were from the Salmon River. Four NP carcasses recovered were missing an operculum punch (i.e., unmarked). Our collection efficiency was 59.4% for NP carcasses and 75.0% for IB carcasses in 2020 (Table 5). Weir efficiency (marked carcasses recovered / by total carcasses recovered) was high (96.7%). Weir efficiency in 2020 was similar to 2019 (97.6%), 2018 (99.0%), and 2016 (97.3%), when the weir was installed on time, and higher than in 2017 (63.7%), when high water delayed weir installation.

The distribution of carcasses (sexes combined) upstream of the SFH weir, in 2020, was similar to previous years. Carcasses were collected throughout the surveyed reaches, but most were collected at the weir. The median distance upstream from the weir NP carcasses were recovered (N = 115) was 0.0 km (i.e., at the weir; Figure 1). The maximum distance upstream from the weir that a carcass was found was 23.3 km. Two IB carcasses were recovered on the weir, and one IB carcass was recovered approximately 700 m upstream from the weir.

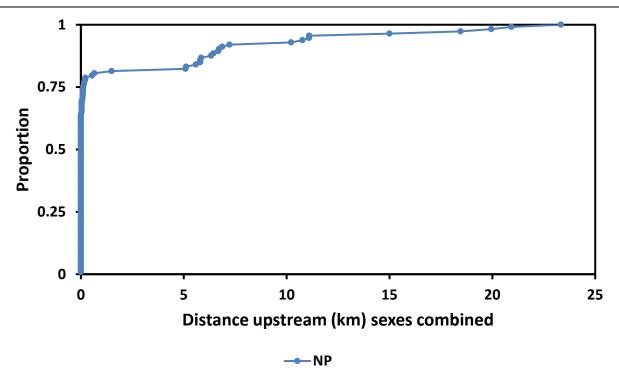


Figure 1. The distance upstream of the Sawtooth Hatchery weir natural-origin carcasses of both sexes were collected in 2020.

The distribution of female carcasses, in 2020, suggested NP females distributed themselves further upstream than the combined NP group. The median distance upstream of the weir NP female carcasses (N = 17) were collected was 6.3 km. One female IB carcass was collected on the weir. The maximum distance upstream from the weir that a female carcass (NP)

was found was 23.3 km (Figure 2). This female was found approximately 5 m downstream of the redd she had been observed on in the previous survey.

#### DISCUSSION

Surveying the large amount of potential spawning habitat upstream of the SFH weir (59.1 km of Salmon River and tributaries) requires balancing the competing needs of providing adequate spatial coverage while maximizing the number of carcasses collected. We focused our surveys primarily on areas with the highest spawning densities (i.e., core spawning areas) to maximize carcass collection. However, based on prior years of redd surveys and carcass collections, we also surveyed selected reaches outside of these core areas to provide additional spatial coverage in fringe habitats. Our high carcass recovery rates combined with the recovery of carcasses in tributary streams in 2016-2017 indicates that our approach balances these competing needs effectively. We will continue to focus our surveys in this manner, but will remain flexible as to where we conduct carcass surveys outside of core areas if redd surveys indicate spawning expands into new areas. However, such an expansion remains unlikely until escapement improves.

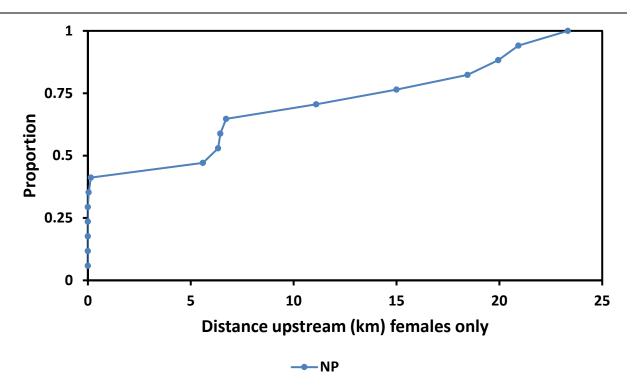


Figure 2. The distance upstream of the Sawtooth Hatchery weir natural-origin female carcasses were collected in 2020.

Conducting carcass surveys from inflatable kayaks proved to be an efficient addition to ground surveys, given the length of river surveyed. The section of river surveyed is generally open with little streamside vegetation, making carcasses along the water line or in backwaters readily observable. One surprising observation was that carcasses lodged on the bottom of the main

channel were also readily observable. This is likely due to the high angle of observation relative to the water surface experienced by boat surveyors.

Our carcass collection efficiency has been consistently high. Stream flow in 2016–2017 provide good bookends for efficiency estimation. In 2016, flows in the upper Salmon River were low, while 2017 flows were above normal. We averaged over 50% collection efficiency in all years, which is almost double that reported elsewhere (Murdoch et al. 2009b; Murdoch et al. 2010). Our lowest recovery efficiency has been for NP carcasses. This may be due to NP females spawning further upstream in reaches we cannot realistically access or survey infrequently. This difference in recovery efficiency underscores the importance of continued surveys upstream of the County Line Bridge and opportunistic surveys whenever and wherever possible.

Carcass collections in previous years indicated that there has been longitudinal spawning segregation, by origin, in Chinook Salmon upstream of the SFH weir. However, this segregation may be less prevalent in years with low escapement. During 2016 and 2017, median NP female carcass recovery was 9.6 km and 15.1 km upstream of the weir, respectively (Venditti et al. 2019). Conversely, as escapement declined in 2018 and 2019, the median distance NP female carcasses were recovered also declined to 3.1 km and 0 km upstream of the weir (Venditti et al. 2020a, 2020b). This decline may be due to the high number of IB and SS females that have spawned near the weir in previous years successfully returning naturally produced offspring relative to the small number of NP females that spawned further upstream. In 2020, the median recovery distance of NP female carcasses increased somewhat along with the number of NP females released (Table 5).

Murdoch et al. (2009b) found the mean distance female carcasses were recovered from their known redd location was 150 m, which suggests carcass drift is likely trivial relative to the differences in median distances females distributed themselves upstream of the weir (Venditti et al. 2019, 2020a, 2020b). Additionally, while carcass drift will have a negative bias on distribution distance, there is no reason to assume this bias would be different for females of different origins.

# CHAPTER 3: WHAT IS THE CONTRIBUTION OF INTEGRATED ADULTS TO THE NATURAL POPULATION OVER MULTIPLE GENERATIONS?

#### INTRODUCTION

The supplementation efforts of the integrated broodstock (IB) program are intended to increase the number of naturally spawning adults. The program aims to achieve this by incorporating a portion of natural-origin adults into the supplementation broodstock (i.e., integrated broodstock) and then allowing the adult progeny of these supplementation fish to spawn naturally. This approach is predicted to increase the number of natural-origin returns (and spawners) in subsequent generations. For this strategy to work two things must happen. First, natural-origin adults incorporated into the supplementation broodstock must, on average, return more adult progeny than those that spawn naturally. Second, the supplementation adults that are allowed to spawn naturally must return more adult progeny than would have been present if the original natural-origin adults had not been taken into the supplementation program. If successful, the boost in numbers of naturally spawning fish will be larger than what the natural-origin fish could have produced if they had not been incorporated into the IB.

Several studies have examined the reproductive success of hatchery-origin Chinook Salmon to that of natural-origin fish in the natural environment (Hess et al. 2012; Christie et al. 2014; Evans et al. 2016; Janowitz-Koch et al. 2019). These studies have been helpful in generating insights into the comparative reproductive success of hatchery-origin Chinook in nature to that of natural-origin fish. However, no study to date has accounted for lost production of natural-origin fish incorporated into a supplementation broodstock when comparing the replacement rate of naturally spawning fish with supplementation parents to that of spawners with natural-origin parents. The question remains if the natural production of offspring from a supplementation program is sufficient to replace lost production of the natural-origin adults incorporated in the broodstock. This question was identified as a key critical uncertainty in evaluating the benefits of supplementation over a decade ago (ISAB 2003; Mobrand et al. 2005; Ford et. al. 2006), yet still has not been fully addressed.

A definitive approach to address this question is to use genetic parentage analyses to compare the number of recruits per spawner from IB and NP adults in both environments through multiple generations. Several studies have used this approach to examine the demographic effects of a supplementation program in Chinook Salmon. Hess et al. (2012) presented the first summaries of this approach and Janowitz-Koch et al. (2019) built upon their results. Both studies demonstrate a demographic boost from supplementing the naturally spawning population with hatchery-origin fish. Our evaluation differs from these studies in several important aspects. First, instead of a single study site our evaluation examines effects of supplementation at multiple populations, each associated with a hatchery facility: the upper Salmon River (associated with the Sawtooth Hatchery), the Pahsimeroi River (associated with the Pahsimeroi Hatchery), and the South Fork Salmon River (associated with the McCall Hatchery). Conducting these evaluations across various locations in the state will help determine if the effects of supplementation are consistent across the landscape. Second, the study system used by Hess et al. (2012) and Janowitz-Koch et al. (2019) incorporated 100% natural-origin fish annually into the broodstock used to create supplementation fish. Our evaluation targets this same level of contribution of natural-origin fish at the Pahsimeroi site but we also incorporate a target of only 90% natural-origin fish into the broodstock at the remaining locations. This approach mimics more common supplementation scenarios in which the number of natural-origin fish available for spawning at a hatchery is limited. It also allows us to pose the question of whether the expected demographic boost is maintained when <100% natural-origin broodstock are used for creating

supplementation fish. Third, and perhaps most importantly, our evaluation not only monitors the demographic boost expected from a supplementation program but also addresses the question of whether the production of natural-origin offspring resulting from a supplementation program is sufficient to replace lost production of the natural-origin adults incorporated in the broodstock. Answering this question will be critical for evaluating the long-term feasibility of implementing a supplementation program.

The IB program is uniquely situated to directly address this critical question. The three Chinook Salmon hatcheries in the Salmon River basin have efficient weirs to facilitate complete (or nearly complete) sampling of adults returning to the study areas upstream. Genetic analyses are also a part of the evaluation programs in these facilities. The populations under study follow the currently preferred management option of using dedicated supplementation stocks that are integrated with the natural populations at known levels (i.e., PNI). A conceptual diagram that depicts the stages of the supplementation program over time is available (Figure 3). Our objective for this chapter is to track the productivity of a parental lineage through two generations. Here we summarize the productivity of the parental lineages that spawned in SY2015.

#### **METHODS**

For this evaluation period, the replacement rate based on genetic parentage assignments for IB and NP adults at three locations (SFSR, Sawtooth, Pahsimeroi) was calculated for brood year 2015 following the structure of Figure 3. Returning adults will provide information for multiple generations, but for this evaluation period we summarize the replacement rates using the F<sub>1</sub> offspring from the first female lineage (P1 Figure 3). Adult females contributing to the P1 generation were trapped at the weir and either spawned into the IB line (P1: Figure 3) or passed above the weir for natural spawning (P<sub>1n</sub>). All adults brought into the IB broodstock were genetically sampled for parentage-based tagging (PBT), genetically tagging their progeny in order to identify their female parent and brood year of origin (Steele et al. 2011, 2018b). The F<sub>1</sub> progeny resulting from the IB line received identifying tags (AD intact/CWT) to enable identification of these fish as IB origin and protect them from sport harvest when they returned as adults in subsequent generations. During this same year (2015), DNA was also collected from all adults passed upstream of the weir, which allowed us to assign adult F<sub>1</sub> returns from natural production to a mother and brood year using the same techniques. The F<sub>1</sub> adults from the IB and NP lines returned to the weir 3-5 years later, and were genetically sampled to identify the individual's genetic sex and for genetic assignment to its mother via PBT. From these adult samples we computed the number of recruits per female (R/F), mean number of recruits per female (mean R/F), number of female recruits per female (RF/F), and the mean number of female recruits per female (mean RF/F). The ratios of mean RF/F for the IB and NP lines provide an estimate of hatchery amplification achieved through the IB program. As the next generation (F<sub>2</sub>) of offspring return, we can compute the same metrics for this and subsequent generations. The natural replacement rates for brood years in the absence of supplementation can then be compared to years with IB supplementation.

DNA from tissue samples (fin clips) from adults and progeny were extracted and genotyped following protocols for the Nexttec Genomic DNA Isolation Kit from XpressBio (Thurmont, Maryland). Protocols of library preparation for next-generation genotyping followed Campbell et al. (2015) using Genotyping-In-Thousands (GT-Seq) technology. Briefly, library preparation begins with an initial multiplex PCR reaction that is used to ligate sequencing primers to the target sequences that are known to contain single-nucleotide polymorphisms (SNPs). Samples were processed using a panel of 299 SNPs, including a diagnostic SNP for determining

sex, targeting sequences described by Hess et al. (2016). In a subsequent PCR the sample is "barcoded" by ligating an additional sequence to the target that identifies the sample's tray of origin (i7 barcode) and its position on the tray (i5 barcode). After barcoding, the quantity of DNA was normalized for each sample using a SequalPrep™ Normalization Plate Kit (Applied Biosystems) that binds a standard amount of amplicon product for normalization of concentrations. All samples per tray were then pooled into a single 'plate library' that was quantified by a Qubit fluorometer (Thermo Fisher). Concentrations were normalized again before being pooled. Loci were genotyped by sequencing the target location on an Illumina NextSeq. A custom bioinformatics pipeline was used to assign resulting sequences and the genotypes back to individual samples using the unique combination of i5 and i7 barcodes. Standardized genotypes were stored on a Progeny database server (www.progenygenetics.com) housed by the Eagle Fish Genetics Laboratory (EFGL).

Parentage analyses were conducted with the program SNPPIT (Anderson 2010). We allowed up to 10% missing genotype data for a sample within the SNP panel before excluding the sample from consideration in parentage. We used an estimated SNP genotyping error rate of 1%, or a per allele rate of 0.5%. SNPPIT assesses confidence of parentage assignments using several criteria including a false discovery rate (FDR) and a log of odds ratio (LOD). We only accepted assignments for a parent-offspring trio (C\_Se\_Se) that had a stringent FDR threshold of <0.05% and an LOD of >14. In this report, additional data for recruits per spawner from brood year 2015 were summarized from across the three return years associated with the respective brood year. In other words, recruits per spawner for BY2015 were summarized from adult offspring returning in 2018–2020.

### **RESULTS**

Results of cumulative adult returns for IB and natural-origin females were summarized for each of the three locations. At all facilities, females from the integrated programs on average returned more offspring and more female offspring than did females that spawned naturally. The maximum number of offspring and female offspring returned to each facility was always from a female used in an integrated cross. Finally, a smaller proportion of females used in integrated crosses at each facility returned no offspring than females passed above the weirs to spawn naturally.

## Sawtooth F<sub>1</sub>

The maximum number of offspring returned per female was six for an integrated cross and five for a natural spawning female (Figure 4). The maximum number of female offspring returned per female was five for an integrated cross and two for a natural spawning female (Figure 5). The mean number of adult offspring that returned from females (R/F) used in integrated crosses in BY2015 was 1.25 (Figure 6). The mean number of adult offspring that returned from natural-origin females (R/F) released above the weir in BY2015 was 0.35 (Figure 6). The mean number of adult female offspring that returned from females (RF/F) used in integrated crosses in BY2015 was 0.75 (Figure 6). The mean number of female offspring originating from natural-origin females (RF/F) passed above the weir in SY2015 was 0.11 (Figure 6). The hatchery amplification observed in the F1 generation ([IB mean RF/F] / [NP mean RF/F]) for BY2015 was 6.8 to 1 (Figure 6).

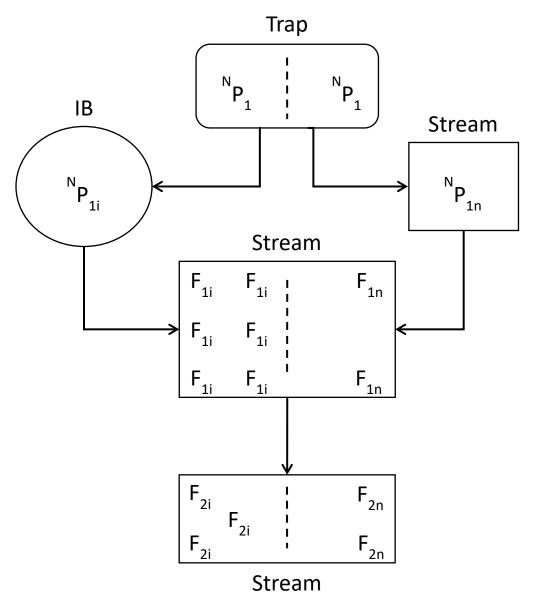


Figure 3. Conceptual diagram depicting how the supplementation effect is tracked through time and how replacement rate between natural spawning stream and the integrated broodstock (IB) can be compared. In this scenario, two natural-origin females (NP<sub>1</sub>) are trapped, and one is brought into an IB program, while the other is allowed to spawn naturally. They each produce some number of adult progeny ( $F_{1i}$  or  $F_{1n}$ ), which are allowed to spawn naturally. In this scenario, a hatchery boost in the replacement rate of the IB lineage ( $F_{1i}$ ) is observed relative to the number of adults returning from the natural production in the stream. The actual supplementation effect is the relative number of  $F_2$  adult progeny in each lineage and the number of natural progeny relative to the prior generation.

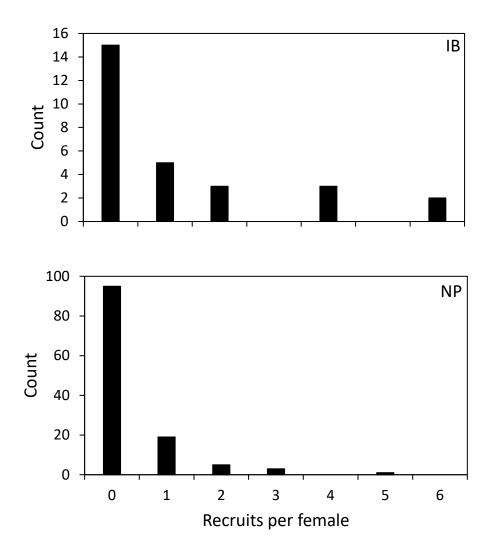


Figure 4. Frequency histograms of recruits per female from brood year 2015 to the Sawtooth Hatchery weir. The top panel represents females spawned in the integrated broodstock (IB). The bottom panel represents females spawning naturally (NP) upstream of the weir.

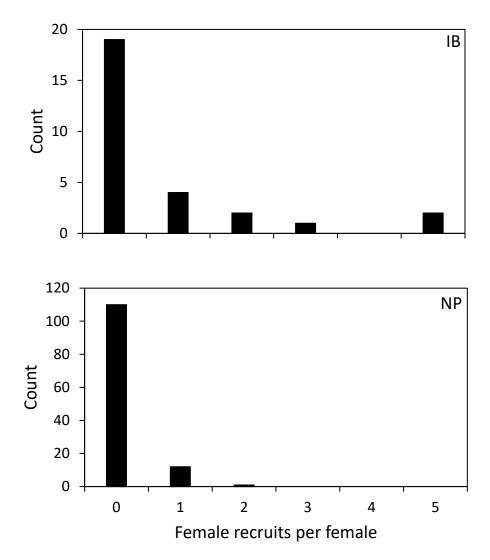


Figure 5. Frequency histograms of female recruits per female from brood year 2015 to the Sawtooth Hatchery weir. The top panel represent females spawned in the integrated broodstock (IB). The bottom panel represent females spawning naturally (NP) upstream of the weir.

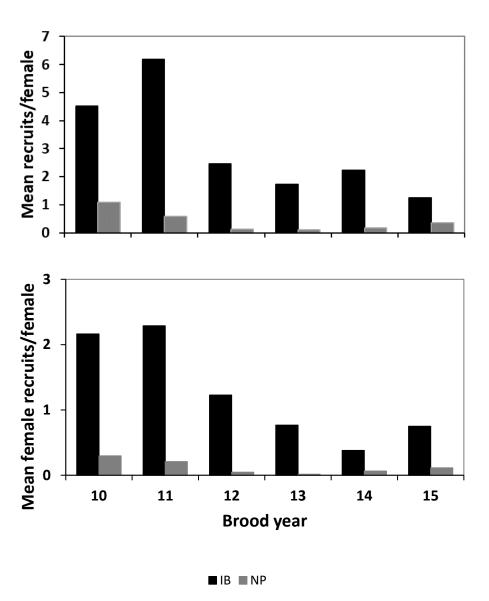


Figure 6. Mean number of recruits per female from the Sawtooth Hatchery integrated broodstock (IB; black bars) program and the natural population (NP; grey bars) in the upper Salmon River (upstream of the hatchery weir) summarized across five brood years evaluated to date. All adult recruits (top) and female recruits only (bottom) are presented.

## McCall F<sub>1</sub>

The maximum number of offspring returned per female was 20 for an integrated cross and one for a natural spawning female (Figure 7). The maximum number of female offspring returned per female was 11 for an integrated cross and one for a natural spawning female (Figure 8). The mean number of adult offspring that returned from females (R/F) used in integrated crosses in BY2015 was 6.7 (Figure 9). The mean number of adult offspring that returned from natural origin females (R/F) released above the weir in BY2015 was 0.03 (Figure 9). The mean number of adult

female offspring that returned from females (RF/F) used in integrated crosses in BY2015 was 2.87 (Figure 9). The mean number of female offspring originating from natural-origin females (RF/F) passed above the weir in BY2015 was 0.03 (Figure 9). The hatchery amplification observed in the F1 generation for BY2015 was 95.7 to 1 (Figure 9).

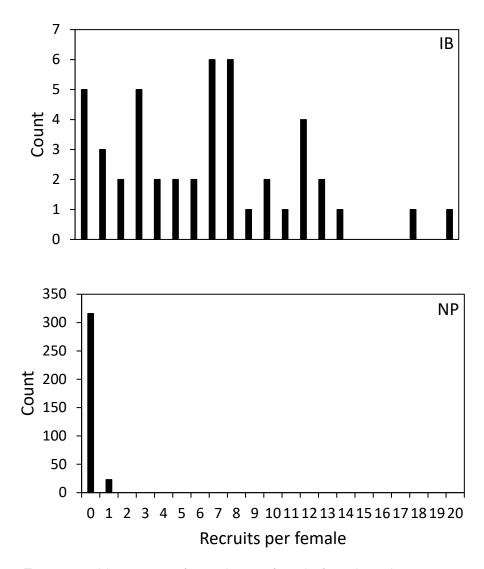


Figure 7. Frequency histograms of recruits per female from brood year 2015 to the South Fork Salmon River weir. The top panel represents females spawned in the integrated broodstock (IB). The bottom panel represents females spawning naturally (NP) upstream of the weir.

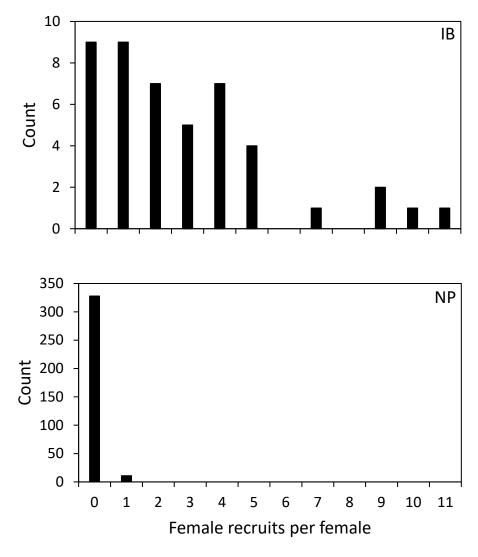


Figure 8. Frequency histograms of female recruits per female from brood year 2015 (to the South Fork Salmon River weir). The top panel represents females spawned in the integrated broodstock (IB). The bottom panel represents females spawning naturally (NP) upstream of the weir.

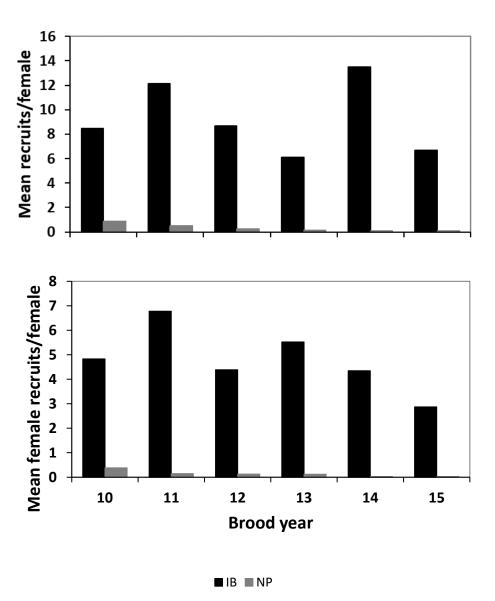


Figure 9. Mean number of recruits per female from the McCall Fish Hatchery integrated broodstock (IB; black bars) program and the natural population (NP; grey bars) in the South Fork Salmon River (SFSR) upstream of the hatchery weir summarized across five brood years evaluated to date. All adult recruits (top) and female recruits only (bottom) are presented.

## Pahsimeroi F<sub>1</sub>

The maximum number of offspring returned per female was seven for an integrated cross and three for a natural spawning female (Figure 10). The maximum number of female offspring returned per female was five for an integrated cross and one for a natural spawning female (Figure 11). The mean number of adult offspring that returned from females (R/F) used in integrated crosses in BY2015 was 2.47 (Figure 12). The mean number of adult offspring that returned from natural origin females (R/F) released above the weir in BY2015 was 0.21 (Figure 12). The mean

number of adult female offspring that returned from females (RF/F) used in integrated crosses in BY2015 was 1.2 (Figure 12). The mean number of female offspring originating from natural-origin females (RF/F) passed above the weir in BY2015 was 0.068 (Figure 12). The hatchery amplification observed in the F1 generation for BY2015 was 17.6 to 1 (Figure 12).

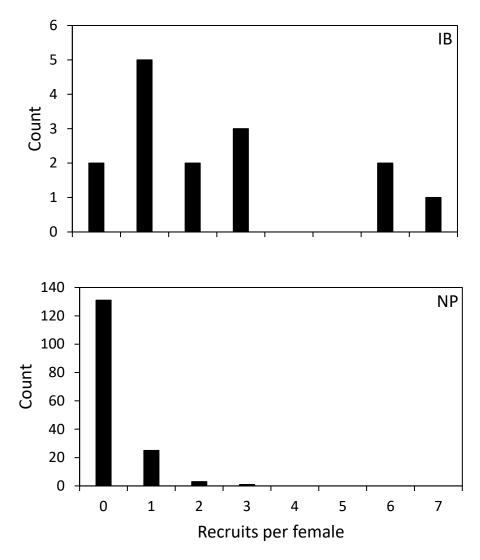


Figure 10. Frequency histograms of recruits per female from brood year 2015 to the Pahsimeroi Hatchery weir. The top panel represents females spawned in the integrated broodstock (IB). The bottom panels represent females spawning naturally (NP) upstream of the weir.

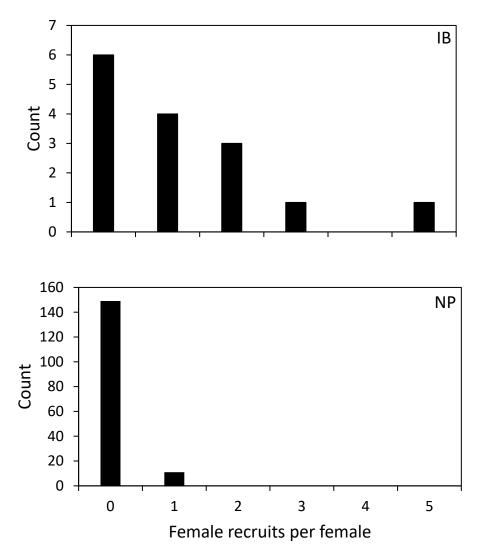


Figure 11. Frequency histograms of female recruits per female from brood year 2015 to the Pahsimeroi Hatchery weir. The top panel represents females spawned in the integrated broodstock (IB). The bottom panel represents females spawning naturally (NP) upstream of the weir.

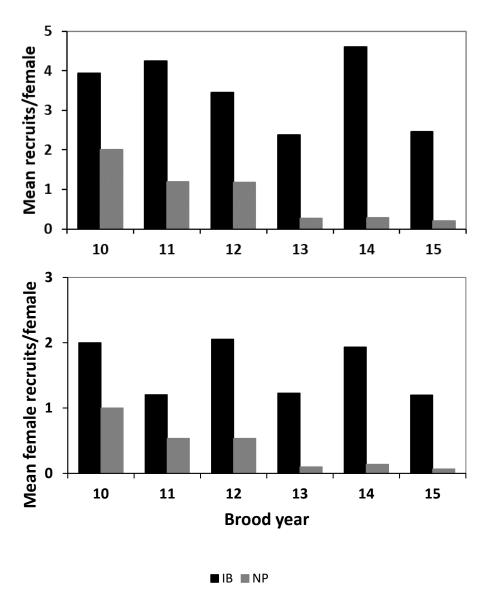


Figure 12. Mean number of recruits per female from the Pahsimeroi Fish Hatchery integrated broodstock (IB; black bars) program and the natural population (NP; grey bars) in the Pahsimeroi River upstream of the hatchery weir summarized across five brood years evaluated to date. All adult recruits (top) and female recruits only (bottom) are presented.

## F<sub>2</sub> Abundances

Implementation of the IB supplementation program has been underway long enough for the first data on the abundance of  $F_2$  returns to now be summarized. Unfortunately, these first summaries coincided with a downturn in ocean condition and a meager number of returns (Table 6). The number of recruits per  $F_1$  female (R/F) and the number of female recruits per female (RF/F) often approached zero at all locations and for all maternal origins. Regardless, the program is now in a position to generate annual summaries on the  $F_2$  abundance.

Table 6. Number of F1 females by origin passed above the South Fork Salmon (MCC), Sawtooth (SAW), and Pahsimeroi (PAH) weirs for natural spawning in 2014 and 2015 and the total number of their F2 progeny (includes age-3, 4, and 5 offspring) that subsequently returned to the weir. Origins include natural (NP), integrated (IB), and segregated (SS).

		F₁ Fema	ale	F <sub>2</sub> Progeny			
Location	SY	Origin	N	Male	Female	Total	
MCC	2014	NP	120	8	2	10	
	2014	IB	283	16	6	22	
	2014	SS	5	0	0	0	
	2015	NP	39	2	1	3	
	2015	IB	271	9	10	19	
	2015	SS	2	0	0	0	
SAW	2014	NP	42	6	5	11	
	2014	IB	66	2	1	3	
	2014	SS	61	3	3	6	
	2015	NP	20	13	2	15	
	2015	IB	32	2	3	5	
	2015	SS	57	12	8	20	
PAH	2014	NP	95	19	17	36	
	2014	IB	79	7	1	8	
	2014	SS	7	0	0	0	
	2015	NP	56	11	2	13	
	2015	IB	51	1	4	5	
	2015	SS	8	0	1	1	

## **DISCUSSION**

This evaluation period showed a large and consistent increase in the productivity of fish spawned in the IB compared to those spawning naturally. This pattern is perhaps not surprising given the protective benefits of hatchery rearing in the early life stages. This effect can be observed in the shape of the frequency histograms that summarize recruits per spawner. The patterns for IB females consistently show a hatchery boost in which many females produce large numbers of offspring. In contrast, naturally spawning NP females always showed a pattern in which most females returned zero adult offspring to the weir and a quick decline in the number of females that return more than a single adult offspring. The pattern of higher recruits/female for IB fish was consistent across all facilities.

Females spawned into the IB and females spawning naturally exhibited similar patterns in which a few females returned large numbers of offspring and many returned none. In the most extreme example, a female spawned in the IB from McCall SY2015 returned 20 adult offspring. When comparisons are made between the maximum numbers of offspring produced in the IB and naturally spawning lineages (Table 7), we see that the size of such "super families" is larger in the IB lineage than in natural-origin lineages. Variance in family size can influence a population's effective size; however, it is just one of several demographic factors, including migration,

fluctuations in population size, and differences in sex ratios, that contribute to this metric (Frankham 1995). The abundance of offspring produced by "super families" in the IB program certainly creates large variances in family size, but this is unlikely to contribute to a reduction in effective population size because the offspring produced by the IB lineage are not an isolated population and are connected to both the hatchery and natural-origin populations. These IB offspring return over three calendar years (as 3-, 4-, and 5-year olds) and during each year a portion of the offspring are either passed above the weir or brought into the hatchery to spawn with natural-origin or hatchery-origin fish. This reduces the prevalence of inbred matings within these family groups while also restoring lost variation within the IB lineage. In fact, just a single migrant per generation is sufficient to minimize loss of heterozygosity and homogenize allele frequencies within subpopulations (Mills and Allendorf 1996). Estimates of effective population sizes are produced annually for each of the IB facilities and the most recent estimates indicate robust effective population sizes of 200–300 for these populations (Harris et al. 2022).

The abundance difference between fish produced by the IB lineage and fish spawning naturally ranged from a 133-fold difference at the SFSR facility to a 2-fold difference at Pahsimeroi. The abundance boost was largest for SFSR for all years evaluated (Table 8). The abundance boost at each location is relative to the natural production at that site and appears to be driven by low numbers of natural recruits per spawner at each facility.

It is possible that progeny from both natural-origin production and from the IB lineage were not fully summarized. Offspring that do not recruit to the weir are not included in these estimates because they cannot be handled, sampled, and enumerated. However, this program attempted to account for this by sampling carcasses of ad-intact fish that were found below the weir, which includes both offspring from the IB program and offspring from natural production. Carcass surveys happened only at the Sawtooth facility because of the known propensity for returning adults to congregate and spawn below the weir. At Pahsimeroi Hatchery the weir is located essentially at the mouth of the river and there is little opportunity for spawning below the weir. At the McCall Hatchery there is also little spawning habitat immediately below the weir. It is feasible to evaluate the proportion of fish that "dropout" based on the PBT assignment rates. A low proportion of natural-origin carcasses receiving PBT assignments below the weir would suggest little drop out while a high proportion would suggest high drop out. At the McCall facility this may also be useful as a relative measure of population closure between the below-weir population of Poverty Flats and the above-weir population of Stolle Meadows. We see no reason NP adults should display higher dropout rates than IB adults and would expect dropout rates, if they occur, to affect progeny of each lineage equally.

Table 7. The maximum number of offspring returned by a single natural-origin (NP) and integrated (IB) female to the three hatchery weirs from six brood years. IB-origin females consistently produced higher maximums than NP fish.

	Brood year											
	2010		2011		2012		2013		2014		2015	
Weir	IB	NP	IB	NP	IB	NP	IB	NP	IB	NP	IB	NP
Sawtooth	12	7	19	6	12	3	8	2	11	3	6	5
South Fork Salmon	15	14	20	18	12	6	21	2	47	3	20	1
Pahsimeroi	30	12	43	5	31	2	9	2	13	4	7	3

Table 8. Fold increases in mean abundance of IB-origin adult recruits compared to mean abundance of adult recruits from natural-origin fish spawning upstream of hatchery weirs. Values are calculated by dividing mean recruit per female (R/F) in the integrated program by the mean R/F for natural-origin females spawning upstream of the weirs.

	Brood year						
Weir	2010	2011	2012	2013	2014	2015	
Sawtooth	4.1	10.6	18.9	15.9	13.5	3.6	
South Fork Salmon	9.8	24.5	33.6	42.4	133.6	98.7	
Pahsimeroi	2.0	3.5	2.9	9.1	15.8	11.7	

The measure of reproductive success for natural-origin females is quite low across all facilities. In this evaluation, we used dual-parent assignment for parentage. Thus, if one parent was missing from the dataset, such as an un-sampled precocial male, then any offspring arising from the precocial contribution will go undetected. Therefore, contribution by precocial males could be reducing the apparent reproductive success of natural females. The degree to which precocial contribution will boost the measure of natural production is unclear. In the future, single-parentage analysis may be used to assign any unassigned natural-origin fish sampled at the weir to a natural parent (likely a mother) that was passed above the weir. Evaluations on the accuracy of single-parentage assignments using simulated data have revealed acceptable rates of false assignments and non-assignments (Steele unpublished data).

This evaluation period demonstrates an abundance boost in the  $F_1$  progeny of originating from the IB program. The most likely explanation for these patterns is hatchery amplification in the survival of a female's progeny. It could be argued that these progeny experienced domestication selection during their rearing in the hatchery and will ultimately be less fit than their natural-origin counterparts when spawning in a natural environment. However, any reduction in fitness in these IB-origin progeny will have to be greater than the abundance boost provided by hatchery rearing.

One of the main goals of the IB program is to compare the abundance of  $F_2$  offspring from females representing each of the three lineages (natural-origin, integrated, and segregated) allowed to spawn in the wild. All three lineages produced  $F_2$  progeny (Table 6). Some lineages performed better than others at a particular location, but overall the number of female  $F_2$  returns is too low to make meaningful comparisons among the lineages. However, we are optimistic that future ocean conditions will result in an overall increase in Chinook returns for the near future allowing us to better evaluate differences in productivity among the groups.

## **ACKNOWLEDGEMENTS**

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## **APPENDIX A. SLIDING SCALES**

Table 1. The sliding scale for Sawtooth Fish Hatchery based on Chinook Salmon run predictions for 2020. The sliding scale incorporates the number of natural origin (NOR) adults that return to the weir, the number of NORs held for broodstock, and the maximum proportion of hatchery origin fish that can be released above the weir.

NOR return to weir	# of NORs held for brood	Maximum proportion of total fish released above weir that are hatchery origin
50 to 249	20 to 100	NA
250 to 499	100 to 131	0.75
500 to 699	131 to 131	0.45
700 to 999	131 to 131	0.45
1,000 to 1,299	210 to 210	0.35
1,300 to 1,599	419 to 419	0.35
1,600 to 2,000	734 to 734	0.35

Table 2. The sliding scale for McCall Fish Hatchery based on Chinook Salmon run predictions for 2020. The sliding scale incorporates the number of natural origin (NOR) adults that return to the weir, the number of NORs held for broodstock, and the maximum proportion of hatchery origin fish that can be released above the weir.

NOR return to weir	# of NORs held for brood	Maximum proportion of total fish released above weir that are hatchery-origin
50 to 124	20 to 50	NA
125 to 424	50 to 95	NA
425 to 699	95 to 95	0.45
700 to 999	156 to 156	0.45
1,000 to 1,299	311 to311	0.35
1,300 to 1,999	622 to 622	0.35
2,000 to 3,000	622 to 622	0.35

Table 3. The sliding scale for Pahsimeroi Fish Hatchery based on Chinook Salmon run predictions for 2020. The sliding scale incorporates the number of natural origin (NOR) adults that return to the weir, the number of NORs held for broodstock, and the maximum proportion of hatchery origin fish that can be released above the weir.

NOR Return to Weir	# of Nors Held for Brood	Maximum proportion of total fish released above weir that are hatchery-origin
50 to 124	15 to 37	NA
125 to 249	38 to 42	0.70
250 to 499	42 to 42	0.30
500 to 999	42 to 42	0.25
1,000 to 1,499	42 to 42	0.25
1,500 to 1,999	42 to 42	0.25
2,000 to 3,000	42 to 42	0.25

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