FISHERY RESEARCH

INTEGRATED BROODSTOCK EVALUATION

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INTEGRATED BROODSTOCK EVALUATION

Project Progress Report

2022 Annual Report

By

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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
M	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 2022. **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 0.55 and 0.33 at Sawtooth, 0.86 and 0.80 at Pahsimeroi, and 0.72 and 0.59 at McCall. Chinook Salmon also spawned 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 0.13 in the upper Salmon River and 0.56 in the South Fork Salmon River. Target PNI was achieved upstream of Pahsimeroi and McCall hatchery weirs and the composite population of the South Fork Salmon River. Target PNI levels were not achieved in the Upper Salmon River. **Chapter 2:** We compared survival between IB and segregated stock (SS) juveniles at Sawtooth, Pahsimeroi, and McCall hatcheries to understand the effects of incorporating naturally produced (NP) adults into broodstocks. Eye-up to release survival was higher in the integrated program than the segregated program at Sawtooth and Pahsimeroi fish hatcheries, but lower in the integrated program than the segregated program at McCall Fish Hatchery. Survival in natural origin smolts was higher than the integrated and segregated smolts for brood year 2021. **Chapter 3:** In 2022, the spawning population upstream of the Sawtooth Hatchery weir was comprised of 57.1% segregated stock adults, 35.9% natural-origin adults, and 6.9% integrated adults. Carcass recovery efficiency was 51.2% for natural origin carcasses and 44.4% for integrated broodstock carcasses. Maximum distance upstream of the Sawtooth Fish Hatchery weir a carcass was found 24.0 km for integrated stock, 25.6 km for segregated stock, and 24.6 km for natural origin. **Chapter 4:** The maximum number of offspring per female in the IB ranged from five at Pahsimeroi to 14 at McCall. The maximum number of offspring in the NP was eight at South Fork Salmon River (i.e., McCall population) and three at Pahsimeroi River and Upper Salmon River (i.e., Sawtooth population). The effects of an IB program on natural production was evaluated by comparing observed abundance of F_2 offspring to a n expected scenario in which natural-origin females were not incorporated into the IB program. Differences in abundances at the SFSR showed an increase for all spawn years and ranged from 6.3 to 29. Differences in abundances at Sawtooth ranged from -0.8 to 3.9. Differences at Pahsimeroi ranged from 1.6 to 9.1.

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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).

A number of uncertainties 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). Furthermore, early rearing in a hatchery 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 differences 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 evolutionary 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 Pahsimeroi and McCall hatcheries target a proportion natural origin broodstock (pNOB) of 0.9, while the target at the Sawtooth 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.

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 hatchery-origin 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. The primary objectives of this research are: 1) assess the influence of NP Chinook Salmon incorporated into IBs on hatchery survival, productivity, and hatchery replacement rates; 2) evaluate the influence of IB spawners on naturally reproducing populations and natural replacement rates; and 3) alter spawning distribution into under-utilized habitat in the upper Salmon River 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. 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 questions addressed by the IB 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 through 4 are addressed in separate chapters below.

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., misclips) 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 2022 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 2022 along with the previous three years for comparison. Origin determination using PBT was generally not possible for fish spawning downstream of weirs, because of the high proportion of carcasses not assigned to a

parent (i.e., the parent was never sampled). When origin could not be determined with PBT, origin was 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; www.finsnet.org). 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

$$PNI \approx pNOB / (pNOB + pHOS),$$

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

The size and composition of the portion of the population spawning downstream of the USR and SFSR weirs was estimated through multiple spawning ground surveys (SGS). Redd counts in the USR and SFSR were conducted by NPM program personnel on three or more occasions throughout the spawning period depending on escapement levels. In the USR, project personnel conducted carcass surveys twice weekly from the Sawtooth Hatchery weir to the mouth of Redfish Lake Creek. Carcass data for Chinook Salmon spawning downstream of the SFSR weir were provided by fisheries biologists from the Nez Perce Tribe. 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). Beginning in 2020, DNA samples from NP carcasses were also provided by tribal cooperators for PBT analysis.

Carcass and redd count data collected 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 Fish Hatchery and McCall Fish Hatchery (SFSR) weirs were converted to proportions and entered into the NOAA Fisheries four-population calculator to estimate PNI for the composite population. Estimates for the four-population calculator were based on mark because carcasses sampled below hatchery weirs have low frequencies of PBT assignments. With advancements in single parentage analysis, we hope to use PBT corrected data in the future. 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. 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. Fish that originated upstream of the weir but spawned downstream are identified by an NP carcass 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 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 were negligible. We also assumed that samples that failed to genotype were random with respect to origin.

RESULTS

PNI Upstream of Weirs

Estimated PNI values for the portion of Chinook Salmon populations spawning upstream of weirs in 2022 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 2022, estimated PNI based on marks

ranged from 0.55 at SFH to 0.86 at PFH, while PNI based on origin-corrected PBT data ranged from 0.33 at SFH to 0.80 at PFH (Table 1). In 2022, we achieved the target PNI goal, based on mark, at PFH and MFH. We also achieved the PNI goal, based on PBT, at the PFH, 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 MFH and SFH using PBT corrected data (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.

			Above Weir PNI				
Hatchery	Year	Method	pNOB (IB)	pNOS	pHOS (IB)	pHOS (SS)	PNI
Sawtooth	2022	Mark	0.47	0.62	0.17	0.21	0.55
		PBT	0.39	0.20	0.29	0.51	0.33
	2021	Mark	0.65	0.86	0.14	0.00	0.82
		PBT	0.55	0.76	0.16	0.08	0.70
	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
Pahsimeroi	2022	Mark	1.00	0.84	0.16	0.00	0.86
		PBT	0.86	0.79	0.13	0.08	0.80
	2021	Mark	0.78	1.00	0.00	0.00	1.00
		PBT	0.74	0.90	0.04	0.07	0.88
	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
McCall	2022	Mark	0.74	0.71	0.29	0.00	0.72
		PBT	0.59	0.58	0.35	0.07	0.59
	2021	Mark	0.57	0.31	0.69	0.00	0.45
		PBT	0.33	0.19	0.74	0.06	0.29
	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

After adjusting for biases associated with marks, long term (2014–2022) PNIs above the weirs were 0.47 at SFH, 0.62 at PFH, and 0.40 at MFH. Natural-origin adult returns were insufficient to achieve targeted pNOB levels (90% at PFH and MFH and 100% at SFH) 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 2022 and contributed to the annual and long-term PNI levels observed. The escapement of NP adults was not sufficient to meet pNOB objectives at Sawtooth and McCall fish hatcheries. At the SFH and PFH, IB escapement was insufficient to meet our supplementation objectives. Eighteen IB females and 38 IB males were passed upstream of the SFH weir for natural spawning. Thirteen IB females and 33 IB males were passed for natural spawning at the PFH weir. The MFH was the one relatively bright spot, with IB females making up 50.2% (by PBT; n = 102) 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 23 and September 23, 2022. During this time, we collected 159 carcasses of known origin including 4 IB, 77 NP, and 95 SS (Table 2). Carcasses were collected from the SFSR between August 8, 2022 and September 20, 2022. On hundred and forty-one carcasses were collected, including 7 IB, 111 NP, and 23 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 rivers downstream of the Sawtooth and McCall fish hatchery weirs during the most recent four years.

Population	Year	Sex	Origin			
			IB	NP	SS	Total
Upper Salmon	2022	Female	1	42	42	85
		Male	3	33	50	86
		Unknown	0	2	3	5
		Total	**4**	**77**	**95**	**176**
	2021	Female	5	39	45	89
		Male	1	29	39	69
		Unknown	0	1	0	1
		Total	**6**	**69**	**84**	**159**
	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**
South Fork Salmon River	2022	Female	7	64	17	88
		Male	0	46	6	52
		Unknown	0	1	0	1
		Total	**7**	**111**	**23**	**141**
	2021	Female	2	9	21	32
		Male	0	7	23	30
		Unknown	0	0	0	0
		Total	**2**	**16**	**44**	**62**
	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**

The composite PNI value for the SFSR population was above the >0.5 target, but the composite PNI value for the USR population was well below the >0.5 target in 2022. The composite PNI in the SFSR was 0.56 and 0.13 in the USR (Table 3). The low PNI values in the USR were primarily driven by high proportions of SS that spawned downstream of the weir and low numbers of natural-origin adults passed upstream and included in the IB. For the duration of the record (2014–2022), composite PNI estimates average 0.14 in the USR and 0.37 in the SFSR. Estimated PNI upstream of both weirs were higher than composite PNI estimates, demonstrating 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 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.

| Population | Year | Method | Downstream Segment | | | | PNI 2 |
			pNOS (US)	pNOS (DS)	pHOS (IB)	pHOS (SS)	
Upper Salmon	2022	Mark	0.27	0.17	0.04	0.54	0.13
	2021	Mark	0.07	0.36	0.04	0.53	0.13
	2020	Mark	0.01	0.60	0.03	0.36	0.17
	2019	Mark	0.09	0.21	0.01	0.69	0.14
South Fork Salmon River	2022	Mark	0.69	0.10	0.05	0.16	0.56
	2021	Mark	0.19	0.06	0.03	0.71	0.17
	2020	Mark	0.50	0.39	0.00	0.11	0.53
	2019	Mark	0.09	0.09	0.09	0.74	0.13

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 upstream. In 2022, PBT records showed that 73 of 117 NP adults (62.4%) collected at the SFH weir and 42 of 372 NP adults (11.3%) collected at the SFSR weir did not assign to parents passed upstream previously, and likely originated downstream. Conversely, 10 of 74 NP carcasses (13.5%) collected downstream of the SFH weir and 11 of the 62 NP carcasses (17.7%) collected downstream of the SFSR weir received PBT assignments to adults passed upstream in the previous generation.

Escapement estimates for the spawning reaches downstream of hatchery weirs indicate Chinook Salmon numbers decreased in the USR and increased in SFSR in 2022 compared to 2021. In the USR population, we estimated 209 Chinook Salmon were present downstream of the weir, and in the SFSR, we estimated 638 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 2022, female prespawn mortality downstream of hatchery weirs was variable in both populations across origins. In the USR, prespawn mortality was about 19% for SS and 7% for NP females. We found no prespawn mortality in IB females. In the SFSR, prespawn mortality was observed in all three origins of fish (Table 4). Prespawn mortality was highest in the SS with 29.4% mortality, followed by 14.3% mortality in the IB, and 3.1% mortality of natural origin females. Overall, female prespawn mortality in 2022 was higher than levels observed in recent years (Table 4.)

Table 4. Selected population parameters for Chinook Salmon spawning in the upper Salmon and South Fork Salmon 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.

Population	Year	Tot. Redd	Origin	PSM (F)	Redds	pRedds	Escapement
Upper Salmon	2022	115	IB	0.000	2	0.017	3
			NP	0.071	64	0.557	110
			SS	0.190	49	0.426	96
			Total				**209**
	2021	130	IB	0.000	8	0.062	14
			NP	0.026	57	0.438	105
			SS	0.044	65	0.500	118
			Total				**237**
	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**
South Fork Salmon River	2022	276	IB	0.143	21	0.050	48
			NP	0.031	214	0.787	494
			SS	0.294	41	0.163	96
			Total				**638**
	2021	136	IB	0.000	9	0.066	20
			NP	0.000	38	0.279	88
			SS	0.000	89	0.654	206
			Total				**314**
	2020	95	IB	NE	0	0	0
			NP	0	73	0.768	169
			SS	0	22	0.232	51
			Total				**219**
	2019	63	IB	0.000	9	0.143	21
			NP	0.000	9	0.143	21
			SS	0.000	45	0.714	104
			Total				**146**

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 been infrequently met (Venditti et al. 2020a, 2020b). This has resulted in long-term PNI levels below program goals. In 2022, 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 goals at PFH and MFH, but minimally supplemented natural spawning above PFH. 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 0.80 (PFH) targets.

Mark based estimates of PNI were higher than estimates using PBT corrected data in 2022 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. In order to routinely achieve PNI targets, 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 true 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. Estimates for composite populations cannot be PBT corrected due to the large and unknown fraction of unsampled carcasses in surveyed reaches.

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 the past. Much of the justification for this effort was to help SFH meet its broodstock need. In 2022,

we were unable to seine below the weir because of high water and unsafe conditions. In 2022, SS broodstock need was met without seining, which contributed to the decision not to seine. The high concentration of adult Chinook Salmon holding below the weir in an easily accessible location is unique to SFH and may be leveraged to provide an opportunity to remove large numbers of hatchery-origin adults from spawning areas with relatively little cost or effort. 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 3; current 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 what actually occurs. We will continue to monitor interchange around the weirs to determine if the direction of interchange remains consistent from upstream to downstream of the weirs. For 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 upstream spawning. However, SS adults short stopping below weirs are certainly a sink for hatchery programs, particularly in low return years.

CHAPTER 2: LIFE CYCLE STOCK SURVIVAL COMPARISONS

INTRODUCTION

Naturally produced populations of anadromous salmonids in Idaho and throughout the Pacific Northwest have declined precipitously since the 1950s. Hatchery supplementation programs have been developed as one way to address the declines and recover populations. Supplementation is defined as "the attempt to use artificial propagation to maintain or increase natural-origin production while maintaining the long-term fitness of the target population, and while keeping the ecological and genetic impacts on non-target populations within specified biological limits" (RASP 1992). Hatchery supplementation programs are commonly used either to augment existing populations or to re-establish extirpated ones. However, debate remains on whether or not supplementation can achieve these goals (ISAB 2003).

Recently, integrated broodstock programs have been developed in an effort to minimize genetic divergence between hatchery donor stocks and recipient natural stocks. A properly integrated program is one where the natural environment drives the adaptation and fitness of a composite population of fish spawning in a hatchery and in the natural environment (HSRG 2009). This is hypothesized to minimize domestication selection in the hatchery and reduce ecological risks to the natural population (HSRG 2009). Integrated broodstock programs may also be used to increase the abundance of natural populations (Sharma et al. 2006; Berejikian et al. 2008), provide harvest opportunities (Fast et al. 2015), minimize risks to wild populations from hatchery straying (Mobrand et al. 2005), provide genetic repositories for natural fish in the hatchery environment (Kline and Flagg 2014), and expand the distribution of fish spawning in under-utilized habitats (Dittman et al. 2010; Venditti et al. 2015).

Many studies have examined the reproductive success of hatchery salmonids released to spawn in the natural environment (Knudsen et al. 2008; Fraser et al. 2010; Van Doornik et al. 2010; Hess et al. 2012; Hayes et al. 2013; Christie et al. 2014; Fast et al. 2015). In the hatchery, the general assumption has been that progeny of hatchery-origin parents perform better than progeny of natural-origin parents because the response of each group to selection is greatest in its own environment (Doyle et al. 1995; Christie et al. 2012). However, the effects of incorporating natural-origin adults into hatchery broodstocks (i.e., integrated broodstocks) and subsequent survival are not well documented. The effect of natural-origin adults on early male maturation (age-2 mini-jacking rates) is one aspect of integrated broodstock programs that has been investigated. Larsen et al. (2004) and Harstad et al. (2014) found early male maturation in all hatchery programs evaluated, and these rates were typically higher in integrated broodstock programs relative to their segregated stock counterparts (Larsen et al. 2022). Early maturation rates may indicate fish are choosing to return at an earlier age, or that survival has decreased and the fish that would return at age-4 or 5 died. The specific mechanism for early maturation is not currently known.

Our objective in this chapter is to provide a more complete understanding of the effects of incorporating natural-origin Chinook Salmon *Oncorhynchus tshawytscha* into hatchery broodstocks. To accomplish this, we evaluate pre- and post-release juvenile survival metrics for integrated broodstock fish relative to their segregated stock counterparts for brood years 2015 to 2021 and phenotypic adult comparisons for spawn years 2016 to 2022. We will determine if integrated broodstock offspring have lower in-hatchery survival compared to segregated stock offspring and if this reduction is maintained post-release through the remainder of their life cycle. Comparisons with natural-origin Chinook will also be made when possible and appropriate. If we detect differences in survival, then we will have the ability to determine when the two broodstocks

diverge and whether this difference remains through adult returns. Results from brood years 2015 to 2020 and spawn years 2016 to 2021 can be found in McCarrick et al. (in press). Results from brood year 2021 juveniles and spawn year 2022 adults are included in this report. We included brood year 2021 in this report even though smolts were released in 2023 because the 2022 project progress report is the last year this evaluation will be included.

METHODS

In-Hatchery Survival

The life cycle evaluation follows in-hatchery survival of IB and SS groups at the Sawtooth, Pahsimeroi, and McCall fish hatcheries from fertilization to release. Hatchery staff provided data through the Fish Inventory System (FINS; www.finsnet.org) database or through monthly hatchery inventory reports. In all hatcheries, complete census counts were conducted at green egg, eyed egg, and at marking (e.g., fin clips, coded wire tags, PIT tags). An estimate of the number of fish released is produced either from a water displacement calculation or by subtracting the total number of mortalities observed following marking from the number of fish marked. Individual crosses were maintained as family units through incubation, which allows us to report eye-up rates for different cross types within the integrated broodstock. After ponding, we no longer had the ability to track survival of individual families (i.e., the effect of different cross types). Integrated broodstock and segregated stock juveniles were maintained in separate raceways, which allowed us to track overall mortality in the two groups during this rearing period.

Survival estimates were produced for green egg to eyed egg (eye-up rates), eyed egg to mark, mark to release, eyed egg to release, and green egg to release. To estimate eye-up rates, spawning records (containing parental origin and green eggs produced) were combined with records of observed mortality within each family. Survival to eye-up within a cross type was the total number of eyed eggs divided by the number of green eggs within each parental cross type. For each family, survival to eye-up was estimated over the incubation period. Family records were then combined by stock (integrated broodstock or segregated stock). Stock specific eye-up rates were estimated as the total number of eyed eggs divided by the total number of green eggs fertilized for that brood year. Percent survival to other points in the life cycle were calculated for each stock as total marked divided by total eyed eggs, total released divided by total marked, total released divided by total eyed egg, and total released divided by total green eggs.

Post-release Survival

After release, we estimated survival from the fish release locations to Lower Granite Dam as smolts. Juvenile Chinook Salmon are PIT tagged at Sawtooth, Pahsimeroi, and McCall fish hatcheries by Pacific States Marine Fisheries Commission marking crews. Typically, the tag distribution is as follows: 1,000 integrated and 19,000 segregated are PIT tagged at Sawtooth Fish Hatchery, approximately 26,000 integrated and 26,000 segregated are tagged at McCall Fish Hatchery, and approximately 21,000 segregated are tagged at Pahsimeroi Fish Hatchery. During brood years 2021, 300 integrated juveniles were tagged at Pahsimeroi Fish Hatchery and no segregated fish were PIT tagged. Additionally, the survival of natural origin smolts tagged at the Natural Production Monitoring Program (Project Number 1991–073–00) rotary screw traps near the Sawtooth Fish Hatchery weir, Pahsimeroi Fish Hatchery weir, and in the South Fork Salmon River are included for comparison.

We queried PIT tag detections for each group at mainstem hydroelectric facilities on the Snake and Columbia rivers and estuary arrays from the PTAGIS database. Survival estimates were calculated using the Survival Under Proportional Hazards (SURPH) model (Lady et al. 2013). Associated 95% confidence intervals were calculated within SURPH for each survival estimate. Confidence intervals were compared to determine between-group survival differences within a brood year (Ott and Longnecker 2016).

Phenotypic Comparisons

We compared adult size structure and run timing between the integrated, segregated, and natural origin groups. Naturally produced groups were included to determine if the hatchery fish have diverged in some way from their natural counterparts. Run timing was based on the date fish were trapped at each hatchery. Trapping date and fork length data were provided by hatchery staff through the FINS database. Size structure between groups was compared with a length frequency histogram for each spawn year. Differences in run timing between groups was assessed by comparing the proportion of the run that arrived at each date for each stock.

RESULTS

In-Hatchery Survival

Average fecundity estimates for brood year 2021 varied between integrated and segregated fish at all three hatcheries. In the integrated programs, both natural origin females and integrated females were used. At Sawtooth Fish Hatchery, integrated broodstock fecundity averaged at 4502 eggs per female (standard deviation (SD) = 795 eggs) and segregated stock fecundity averaged 4411 eggs per female (SD = 714 eggs). Fecundity of integrated broodstock fish at Pahsimeroi Fish Hatchery averaged 3849 eggs per female (SD = 572 eggs) and averaged 5055 eggs per female (SD = 849 eggs) for segregated stock females. McCall Fish Hatchery experienced the lowest fecundities. Integrated Broodstock fish fecundity averaged 3800 eggs per female (SD = 687 eggs). Fecundity of segregated stock fish at McCall Fish Hatchery and averaged 4038 eggs per female (SD = 93 eggs).

Eye-up rates for brood year 2021 averaged >80% and were similar between cross types and stocks at all three hatcheries (Figures 1 and 2; Appendix A, B, and C). Eye-up rates at Sawtooth Fish Hatchery were 94.4% in the integrated broodstock and 95.2% in the segregated stock. At Pahsimeroi Fish Hatchery, eye-up rates in the integrated broodstock averaged 83.2% and 85.8% in the segregated stock. The largest difference between integrated broodstock and segregated stock eye-up rates was observed at McCall Fish Hatchery. Eye-up rates in the integrated broodstock averaged 82.6% and 92.7% in the segregated stock. Within the integrated broodstock, cross types had similar eye-up rates (Figure 2). At Sawtooth Fish Hatchery, eye-up rates averaged 97.5% for natural x natural, 96.4% for natural x integrated (female origin reported first), and 93.4% for integrated x natural. No integrated x integrated crosses were used at Sawtooth Fish Hatchery for brood year 2021. Eye-up rates at Pahsimeroi Fish Hatchery averaged 98.1% for natural x integrated and 87.9% for integrated x natural. Brood year 2021 did not have any natural x natural or integrated x integrated crosses at Pahsimeroi Fish Hatchery. At McCall Fish Hatchery, eye-up rates averaged 76.3% for natural x natural, 83.5% for natural x integrated, 88.8% for integrated x natural, and 81.1% for integrated x integrated.
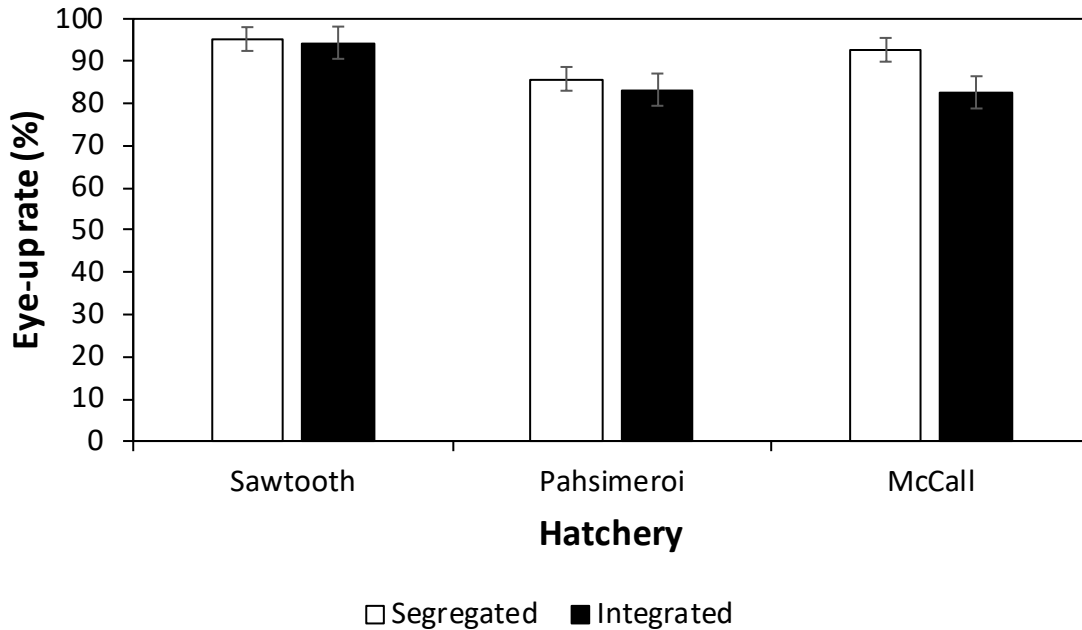
Figure 1.   Average eye-up rates (%) and standard error for brood year 2021 at Sawtooth, Pahsimeroi, and McCall fish hatcheries. Error bars represent one standard error.
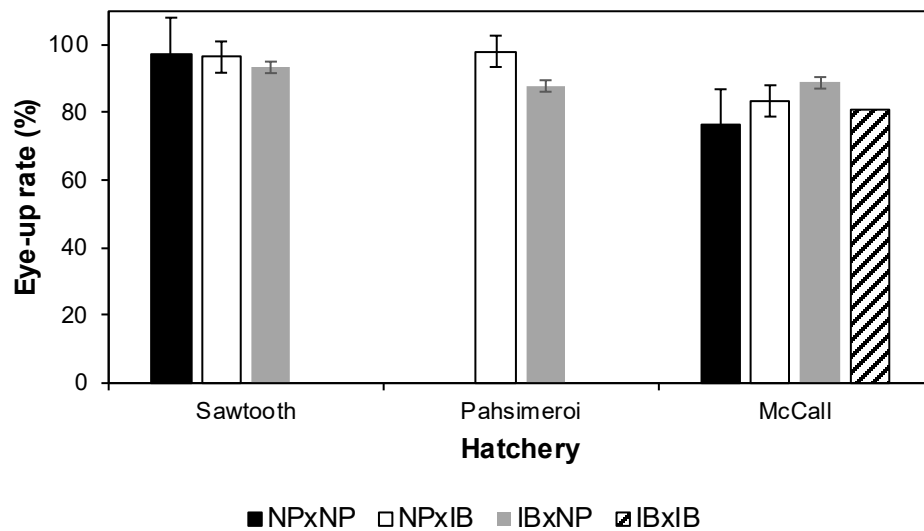
Figure 2.   Average eye-up rates (%) of parental cross type for brood year 2021 at Sawtooth, Pahsimeroi, and McCall fish hatcheries. Cross types represent parental origin (IB = integrated broodstock, NP = natural-origin) with the female listed first and the male second. Error bars represent one standard error.

Eye-up to release survival was high in the integrated and segregated groups for brood year 2021 (Figure 3; Appendix A). Survival to release was higher in the integrated group than the segregated group at Sawtooth and Pahsimeroi fish hatcheries. Survival at Sawtooth Fish Hatchery was 98.8% in the integrated group and 94.7% in the segregated group. At Pahsimeroi Fish Hatchery, survival to release in the integrated group was 99.3% and 83.2% in the segregated group. The opposite pattern was observed at McCall Fish Hatchery where the segregated group had higher survival to release than the integrated group. Survival at McCall Fish Hatchery was 88.2% in the integrated group and 96.0% in the SS group.
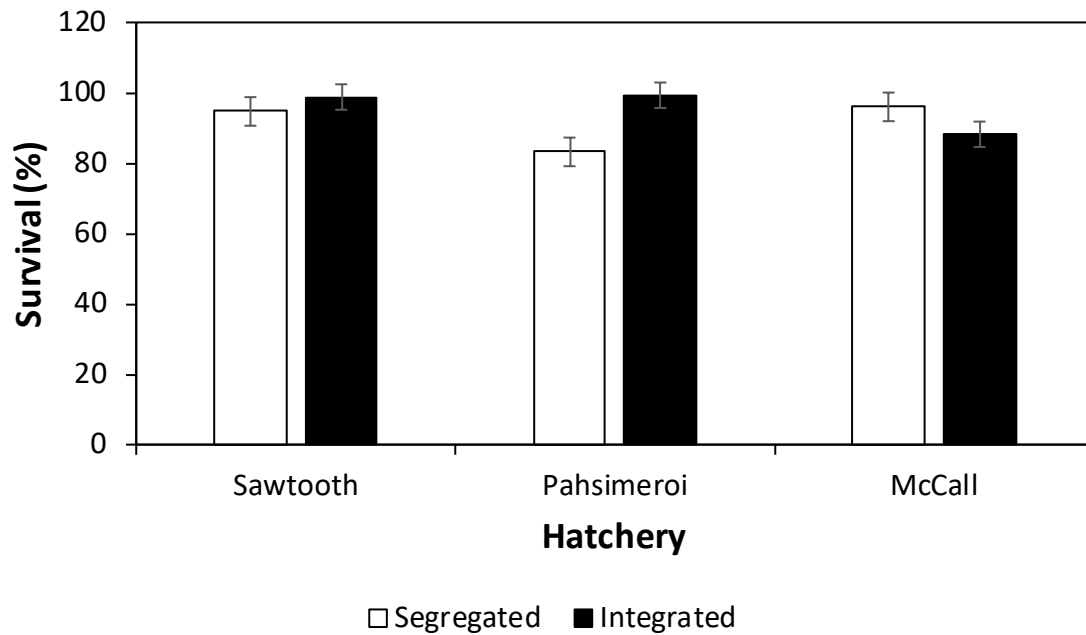
Figure 3.   Percent survival of integrated and segregated juveniles from eyed egg to release for brood year 2021 at Sawtooth, Pahsimeroi, and McCall fish hatcheries. Error bars represent one standard error.

Post-release Survival

Survival in natural origin smolts was higher than the integrated and segregated smolts for brood year 2021 (Figure 4). At Sawtooth Fish Hatchery, survival for integrated smolts was 0.50, while segregated smolt survival was 0.58. Survival of natural origin fish tagged at the Upper Salmon River screw trap was 0.62. Ninety-five percent confidence intervals indicate survival of the integrated group was significantly lower than the segregated and natural origin groups. Survival for integrated smolts at Pahsimeroi Fish Hatchery was 0.48 and survival of natural origin fish tagged at the Pahsimeroi screw trap was 0.55. Integrated and natural origin survivals were not statistically different. At McCall Fish Hatchery, survival was 0.67 for integrated smolts and 0.64 for segregated smolts. Survival of natural origin fish tagged at the South Fork Salmon River screw trap was 0.75. All survival estimates from the South Fork Salmon River populations were not statistically different.
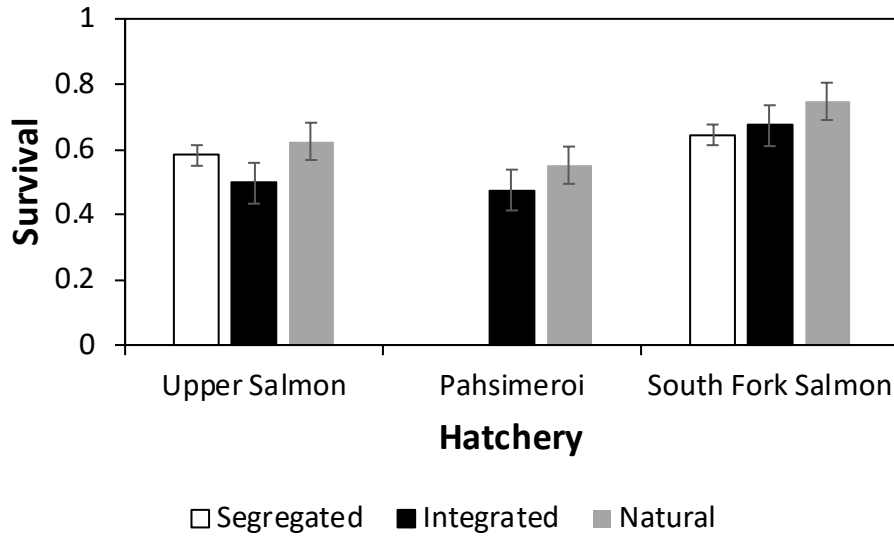
Figure 4.   Estimated survival for brood year 2021 smolts of integrated broodstock, segregated stock, and natural origin from their respective release sites to Lower Granite Dam. Error bars represent one standard error.

Phenotypic Comparisons

Adult length frequencies followed similar patterns across hatcheries. At all three facilities, natural origin fish were larger on average than integrated and segregated fish. During spawn year 2022, integrated fish varied in fork length from 430.0 mm to 920.0 mm and averaged 601.8 mm, segregated fish varied from 390.0 mm to 940.0 mm and averaged 655.0 mm, and natural origin fish varied from 450.0 mm to 1000.0 mm and averaged 725.5 mm at Sawtooth Fish Hatchery (Figure 5). Pahsimeroi Fish Hatchery, integrated fish varied in length from 470.0 mm to 960.0 mm and averaged 685.4 mm, segregated fish varied from 400.0 mm to 920.0 mm and averaged 687.8 mm, and natural origin fish varied from 410.0 mm to 950.0 mm and averaged 734.9 mm during spawn year 2022 (Figure 6). At McCall Fish Hatchery, integrated fish varied in length from 310.0 mm to 940.0 mm and averaged 609.9 mm, segregated fish varied from 350.0 mm to 980.0 mm and averaged 663.2 mm, and natural origin fish varied from 390.0 mm to 940.0 mm and averaged 713.0 mm (Figure 7). The proportion of jacks was noticeably higher in the integrated and segregated programs than the natural origin fish at all locations.

Chinook Salmon began arriving at hatchery weirs in mid-June and continued until mid- to late September at all three hatchery facilities. Differences in arrival timing were observed between segregated, integrated, and natural origin at Sawtooth Fish Hatchery (Figure 8). Most segregated fish arrived early in the trapping season. However, a second pulse of fish from all stocks arrived in late August during spawn year 2022. At Pahsimeroi Fish Hatchery, all three stocks of fish had almost identical arrival timing (Figure 8). At McCall Fish Hatchery, segregated and integrated had similar arrival timing, whereas, natural origin fish initially mimicked the segregated and integrated timing, but experienced a delay during early to mid-August.
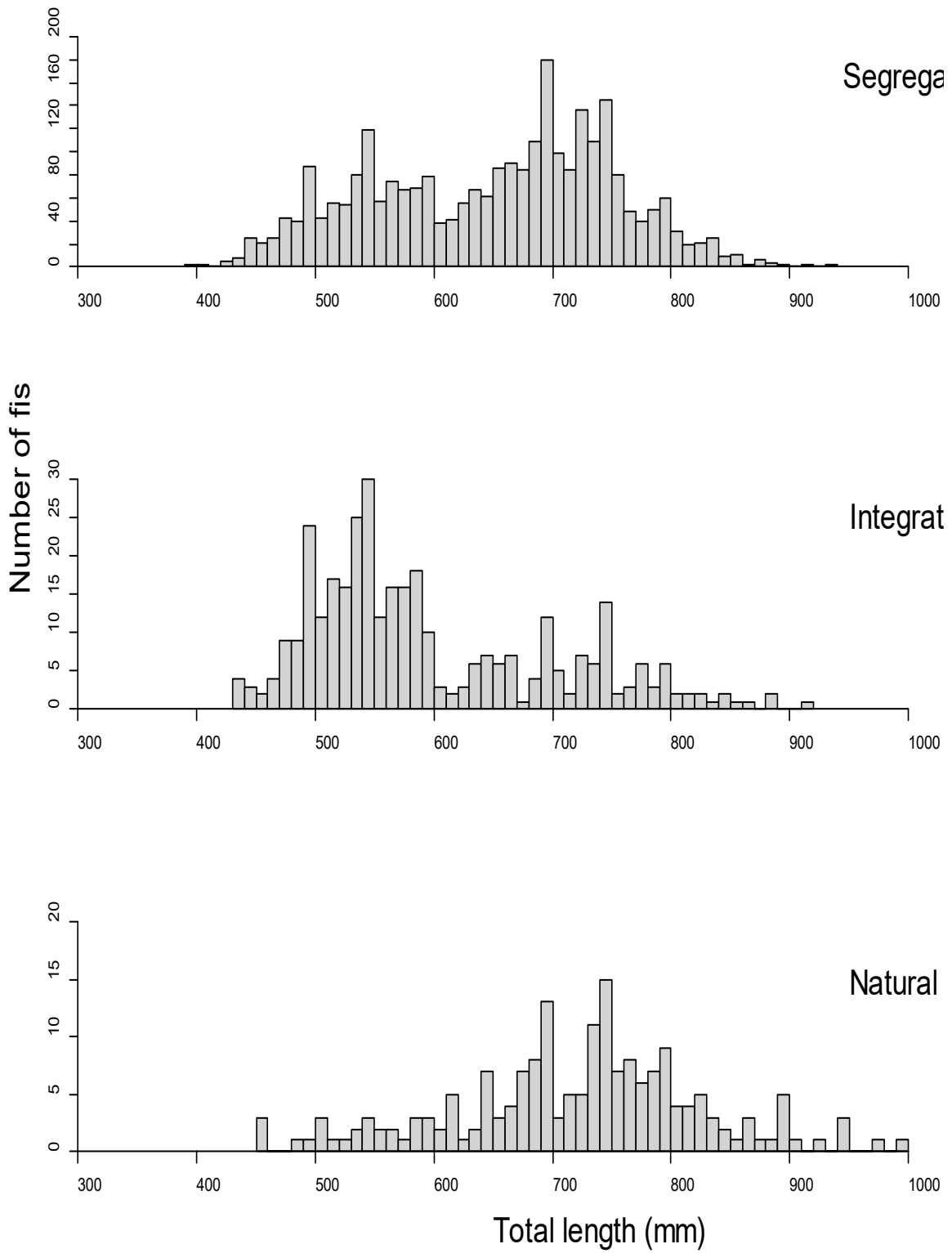
Figure 5.   Length frequency histograms for Chinook returning to Sawtooth Fish Hatchery during spawn year 2022. Please note that the y-axes are not standardized.
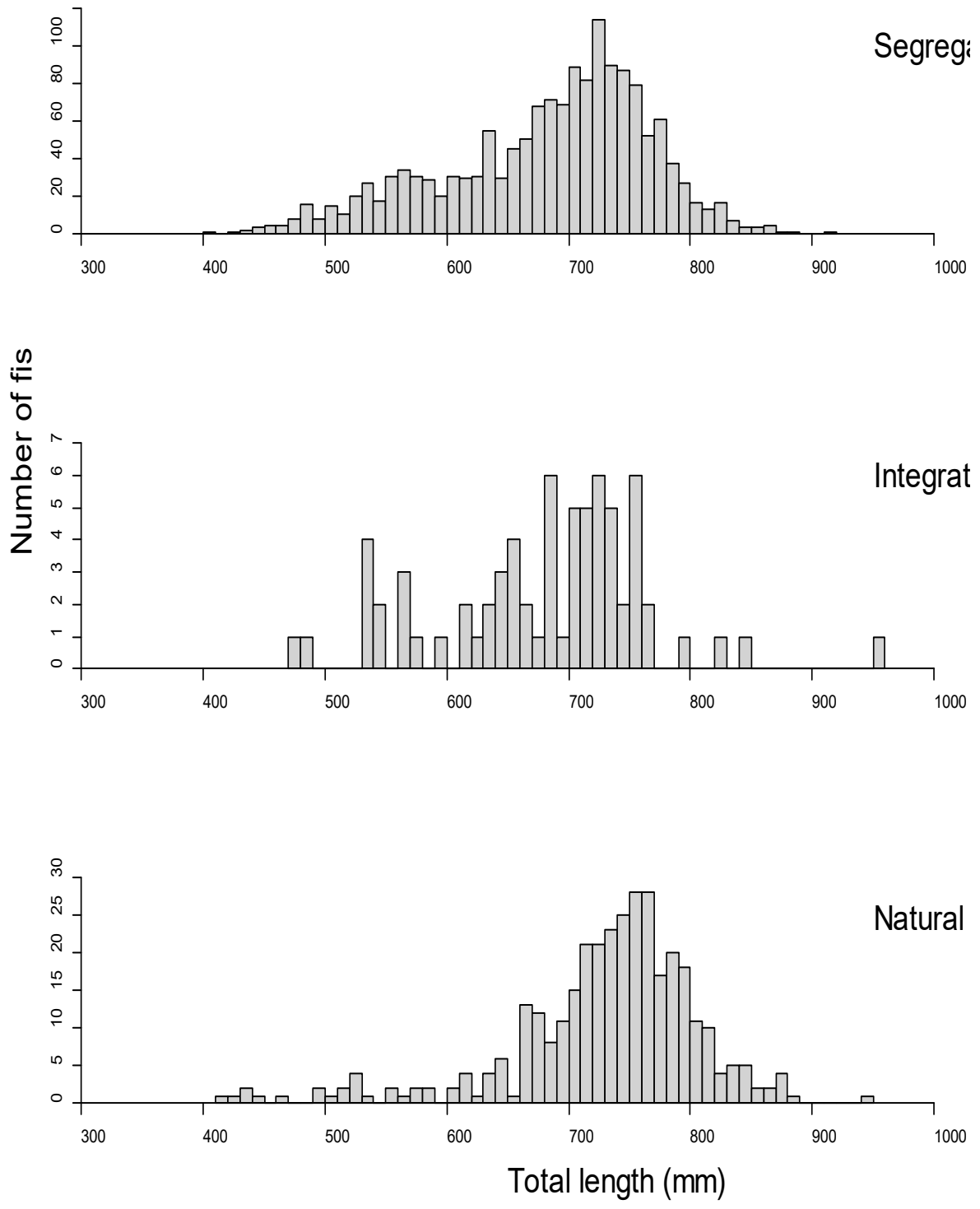
Figure 6.   Length frequency histograms for Chinook Salmon returning to Pahsimeroi Fish
             Hatchery during spawn year 2022. Please note that the y-axes are not
             standardized.
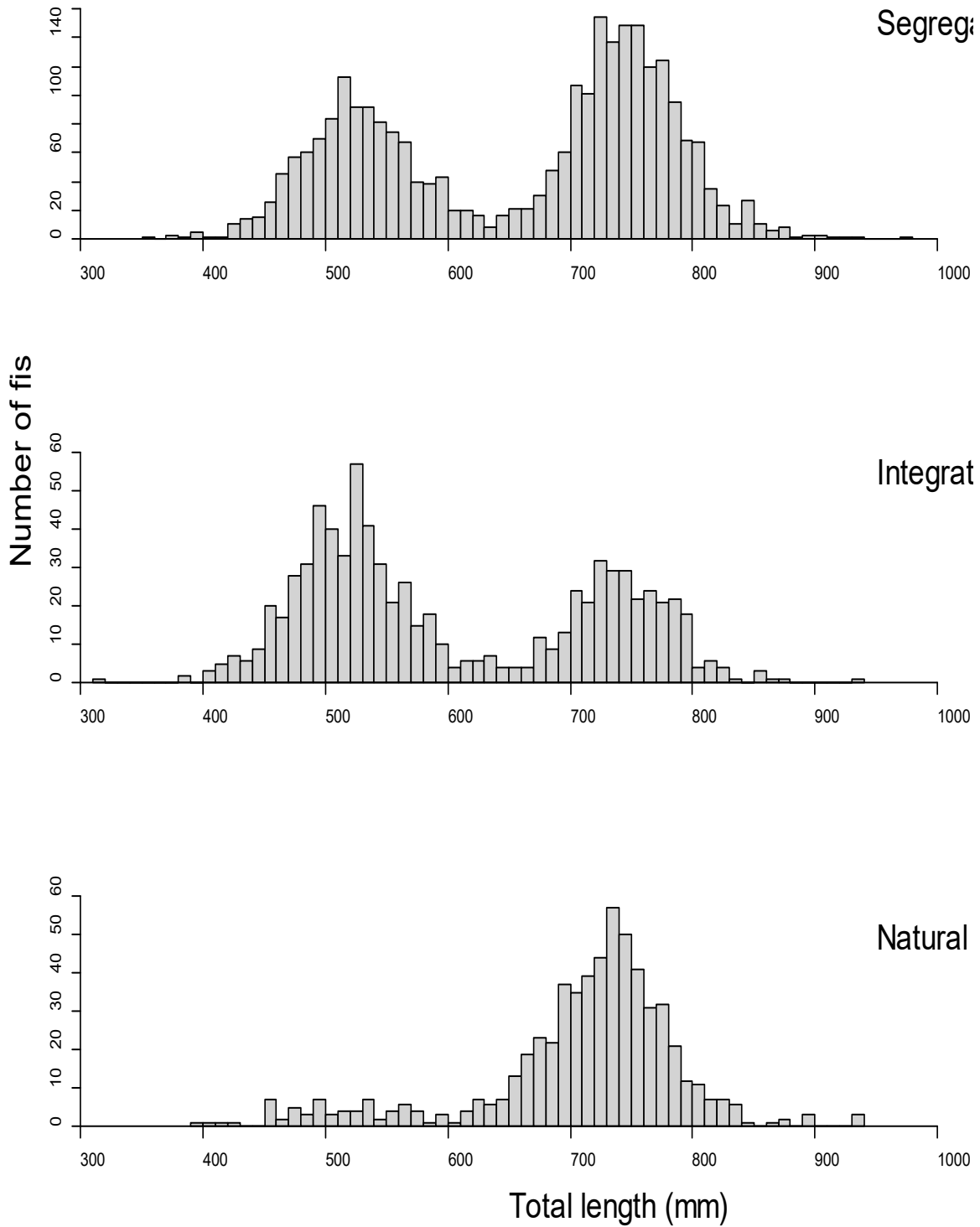
Figure 7.   Length frequency histograms for Chinook Salmon returning to McCall Fish Hatchery during spawn year 2022. Please note that the y-axes are not standardized.

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Sawtooth
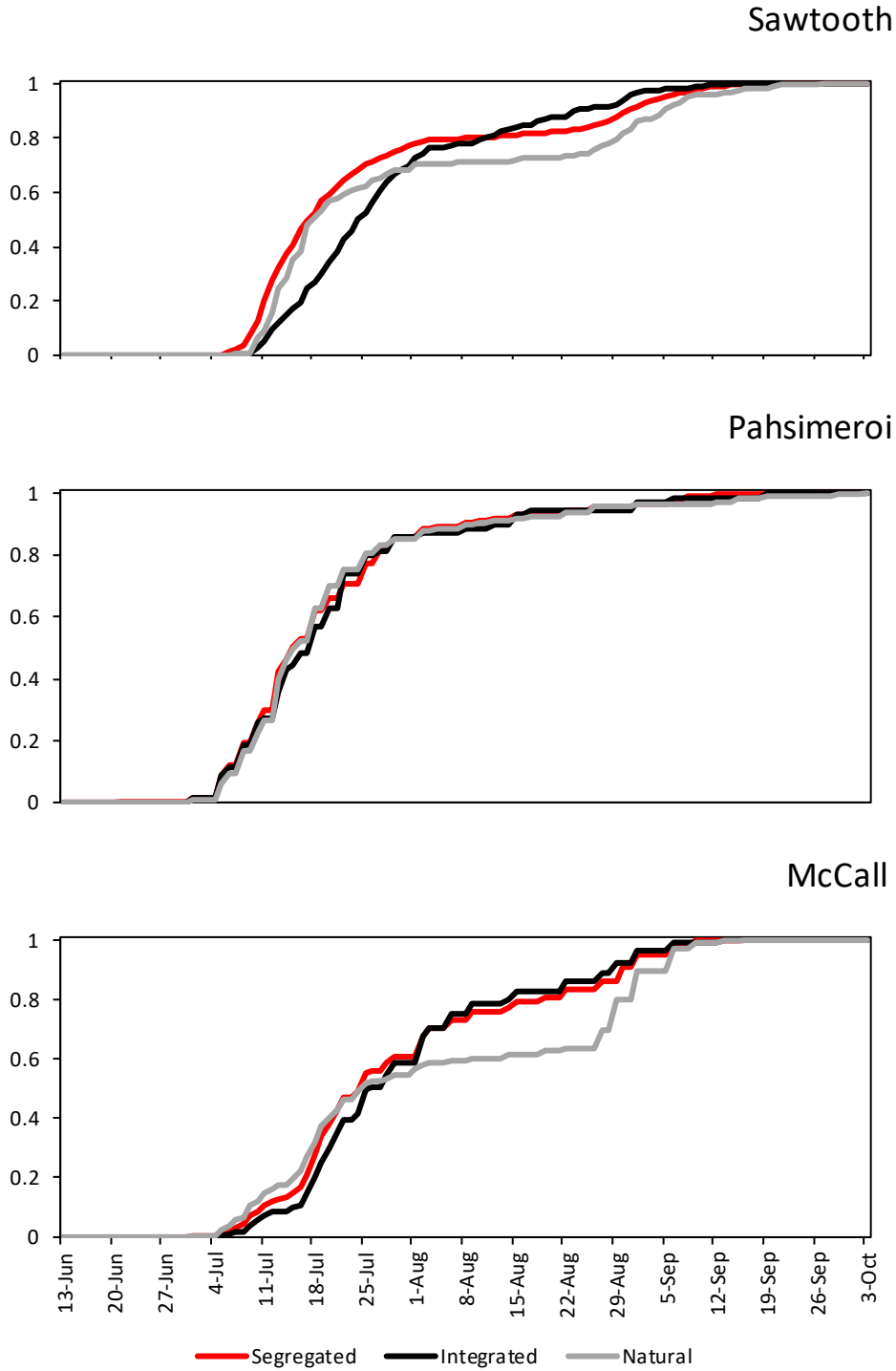
Pahsimeroi

McCall

Figure 8. Arrival timing of Chinook Salmon returning to Sawtooth, Pahsimeroi, and McCall fish hatcheries during spawn years 2022. The red lines represent fish from the segregated program, the black lines are fish from the integrated program, and the grey lines represent natural origin fish.

DISCUSSION

Results from this study suggest juvenile Chinook Salmon perform similarly in the hatchery regardless of parental origin. In-hatchery survival comparisons revealed small differences between the integrated and segregated stocks and within the integrated crosses at Sawtooth, Pahsimeroi, and McCall hatcheries. We documented minimal differences in eye-up rates between the various cross types within the integrated program at all three hatcheries. Eye-up to release survival was higher in the integrated group than the segregated group at Sawtooth and Pahsimeroi fish hatcheries, but was lower in the integrated group than the segregated group at McCall Fish Hatchery. We did not observe differences between the two groups within the hatchery during 2022.

Numerous studies have investigated the effects of hatchery supplementation on natural populations (e.g., Mobrand et al. 2005; Sharma et al. 2006; Kline and Flagg 2014) and the reproductive performance of hatchery fish (e.g., Fraser et al. 2010; Hess et al. 2012; Hayes et al. 2013; Christie et al. 2014; Fast et al. 2015). However, limited information is available on the effect of bringing natural-origin adults into the hatchery broodstock. McDermid et al. (2011) evaluated the effect of parental origin on hatchery reared Lake Trout *Salvelinus namaycush* from four populations. Lake Trout offspring of natural origin parents had higher fertilization rates, lower mortality, and faster growth than offspring of hatchery origin parents when reared in the same hatchery environment. Similarly, Vincent (1960) documented 93% mortality in Brook Trout *Salvelinus fontinalis* from hatchery lineages compared to 64% mortality in Brook Trout from wild lineages from egg take to age 1 in the hatchery environment. In contrast, Chinook Salmon in our study had similar fertilization and mortality rates regardless of parental origin or cross. The effect of parental origin may be species and system specific.

Differences were observed in survival to Lower Granite Dam between stocks and hatcheries. At all three locations, natural-origin smolts had higher survival than both integrated and segregated smolts. At Sawtooth Fish Hatchery, segregated survival was almost 10% higher than integrated. It is unclear whether the trend between the integrated and segregated smolts is real or an artifact of sample size. Approximately 19,000 smolts from the segregated program are PIT tagged annually at the Sawtooth Fish Hatchery, while only about 1,000 integrated smolts are PIT tagged (e.g., Sullivan et al. 2016). The larger segregated program sample size results in more precise estimates of post-release survival for the segregated stock. In contrast, approximately 52,000 smolts are PIT tagged at McCall Fish Hatchery and split equally between integrated and segregated juveniles (e.g., Sullivan et al. 2016). Survival of McCall Fish Hatchery integrated and segregated stocks to Lower Granite Dam was statistically equal in brood year 2022. It is possible the differences observed between Sawtooth and McCall fish hatcheries are a result of sample size, but it may also be a result of environmental differences between the South Fork Salmon River and the Upper Salmon River or genetic differences (e.g., composition of crosses).

In general, natural origin adults were larger and had a lower proportion of jacks than both integrated and segregated hatchery adults. At Sawtooth and McCall fish hatcheries, jacks made up the majority of the integrated returns. Early male maturation has been documented in integrated Chinook programs before (Larsen et al. 2022). Smolt condition and smolt size at release is inversely related to early maturation (Tattam et al. 2015). Although size at release and condition may explain differences observed between natural origin and hatchery length and age at return, it does not explain differences between integrated and segregated fish. In the present study, it is difficult to determine if integrated fish are maturing early, if there is a survival affect that disproportionately effects integrated fish, or a combination of both.

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Results from the present study are encouraging and suggest minimal in-hatchery survival consequences exist for a hatchery program that uses natural-origin adults in the broodstock. Minimal differences in eye-up rates were documented among cross types in the integrated broodstock program. However, we recommend avoiding integrated x integrated crosses when possible due to the relatively small number of fish incorporated into the integrated broodstock. Post release survival has varied greatly through time and warrants further investigation throughout the life cycle. Further research is needed to evaluate cross-specific survival beyond the eye-up stage to fully understand the potential benefits and consequences of parental cross type. Cross-specific evaluation will be especially important if differences in adult returns by cross types materialize, as it will help identify when in the life cycle survival differences occur. Continued research is warranted regarding the effects of bringing natural-origin adults into hatchery broodstocks on the natural population. Completing population level analyses will be useful for developing optimal strategies to maximize fish returns. For example, results from a population model may provide insight into the benefits and consequences of bringing natural origin adults into the hatchery for broodstock and how this may change as a function of density of natural and integrated fish.

CHAPTER 3: 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 mainstem 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 mainstem 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). Fish intentionally released above the weir were marked with an operculum punch. The ratio of un-punched to punched carcasses provides an estimate of weir efficiency. Tissue samples from un-punched carcasses will provide parentage and origin information when genotyped (see Chapter 4; this report) and reduce the number of un-sampled potential parents in the naturally spawning portion of the population. Tissue samples were either heart tissue stored in ethanol or fin clips preserved on paper and stored in coin envelopes (fin on paper) following the handling protocols described in Venditti et al. (2022). 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 (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 of integrated smolts now that the program has entered 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 2022, so few adults were available for release. In total, 33 female and 129 male NP adults along with 14 IB females and 32 IB males were passed upstream of the weir for natural spawning. Segregated stock escapement was sufficient to meet mitigation program broodstock needs, so 32 females and 23 males were intentionally released above the weir (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.

		Released		Recovered									
		Female	Male	Female		Male		Unk.		Sum		Total	Eff.
Year	Origin			M	U	M	U	M	U	M	U		
2022	IB	14	32	3	1	9	3	0	0	12	4	16	0.261
	NP	33	129	8	3	58	10	3	1	69	14	83	0.426
	SS	32	23	11	19	34	63	1	4	46	86	132	0.836
	Total	**79**	**184**	**22**	**23**	**101**	**76**	**4**	**5**	**127**	**104**	**231**	**0.483**
2021	IB	8	16	4	0	9	1	1	0	14	1	15	0.625
	NP	70	112	23	4	70	19	0	0	93	23	116	0.637
	SS	0	0	0	4	2	23	1	0	3	17	21	NE
	Total	**78**	**128**	**27**	**8**	**81**	**33**	**2**	**0**	**110**	**41**	**151**	**0.636**
2020	IB	3	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	0.538[a]
	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**

[a] 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 2022 to determine the longitudinal distribution of spawners. The Ross Fork Fire closure limited access to most of the survey reaches. We surveyed the USR from Huckleberry Creek to the SFH weir weekly, between August 23 and September 23, 2022. The reach from County Line Bridge to Huckleberry Creek was surveyed twice during the 2022 field season. We surveyed approximately 5 km upstream from the County Line Bridge and Alturas Lake Creek from the FS-207 (Cabin Creek) road bridge to the mouth (≈5.15 km) once during the spawning period.

We collected 231 carcasses upstream of the SFH weir in 2022. All but three (recovered in Alturas Lake Creek) were from the Salmon River (Figure 9). Four IB, 86 SS, and 14 NP carcasses recovered were missing an operculum punch (i.e., unmarked). Our collection efficiency was 42.6% for NP carcasses, 26.1% for IB carcasses, and 83.6% for SS carcasses in 2022 (Table 5).

Weir efficiency (marked carcasses recovered / by total carcasses recovered) was lower than previous years (55.0%). In 2022, a large flow event delayed the installation and caused damage to the weir. Fish were likely moving into the upstream reaches prior to weir installation and possibly able to move through the damaged areas.
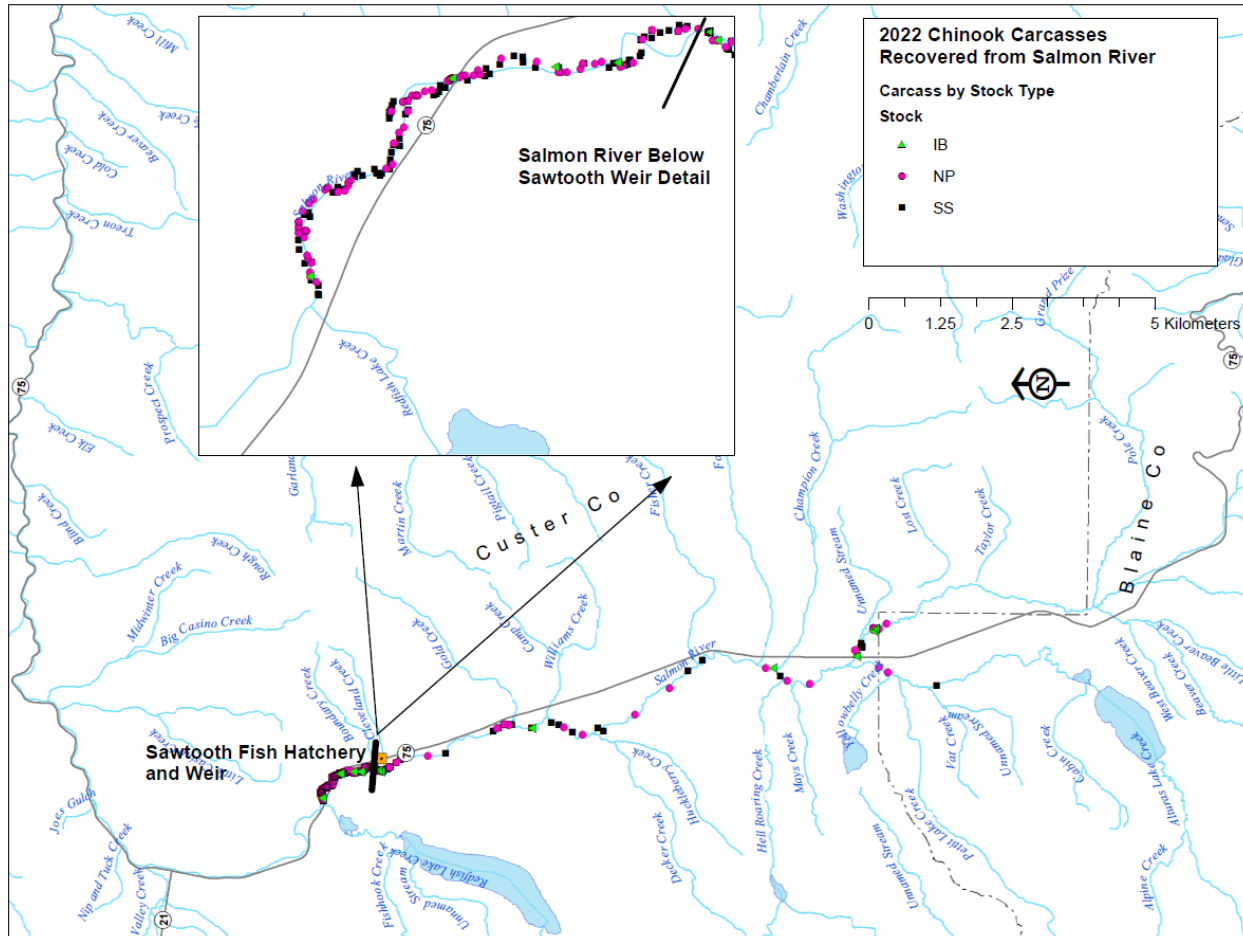
Figure 9.   Distribution of Chinook Salmon carcasses recovered in the Upper Salmon Basin above Redfish Lake Creek. Carcasses are separated by stock type: green triangles for integrated broodstock (IB), pink circles for natural origin (NP), and black squares for segregated stock (SS).

Despite difficulty accessing all survey reaches, the distribution of carcasses (sexes combined) upstream of the SFH weir, in 2022, was similar to previous years. Carcasses were collected throughout the surveyed reaches, but most were collected at the weir. The maximum distance upstream from the weir that a NP carcass was found was 24.6 km and the median distance was 0.00 km above the weir (N = 83; Figure 10). Seven IB carcasses were recovered on the weir. The maximum distance an IB carcass was recovered was 24.0 km upstream of the weir. The median distance upstream from the weir that a IB carcass was found was 0.15 km. The maximum distance a SS carcass was located was 25.6 km upstream of the weir (N=132). The median distance upstream of the weir a carcass was recovered was 0.0 km (i.e., on the weir) for NP and SS.
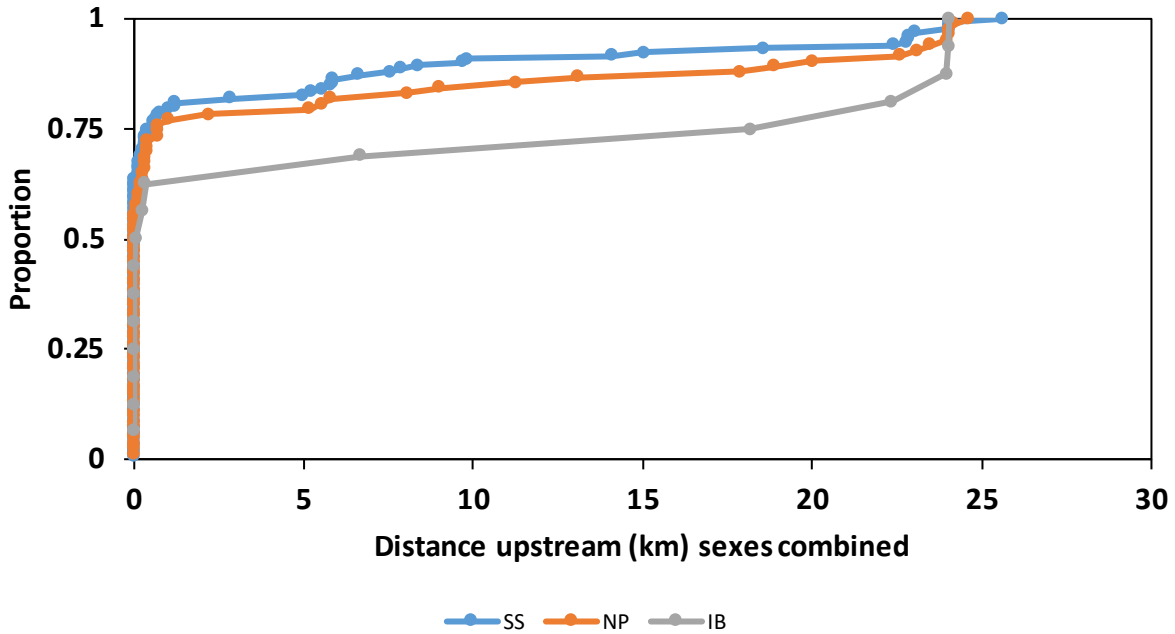
Figure 10.   The distance upstream of the Sawtooth Hatchery weir natural-origin (NP), integrated broodstock (IB), and segregated stock (SS) carcasses of both sexes were collected in 2022.

The distribution of female carcasses, in 2022, suggested NP females distributed themselves further upstream than the combined NP group. The median distance upstream of the weir NP female carcasses (N = 8) were collected was 0.68 km. The maximum distance upstream from the weir that a female carcass (NP) was found was 24.2 km (Figure 11). Two female IB carcasses were collected on the weir, one 0.33 km above the weir, and one 24.0 km above the weir. The maximum distance a SS female was recovered was 25.6 km (N = 30). The median distance SS females were found was 0.97 km above the weir.
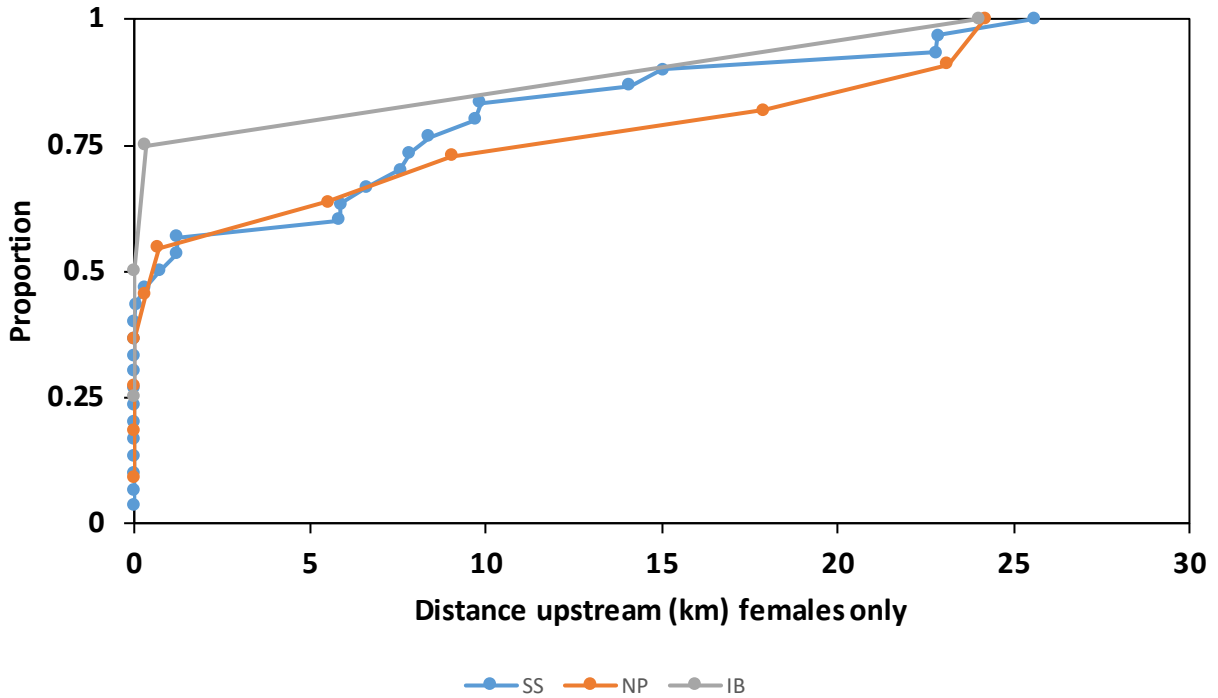
Figure 11.   The distance upstream of the Sawtooth Hatchery weir natural-origin (NP), integrated broodstock (IB), and segregated stock (SS) female carcasses were collected in 2022.

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.

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). In the past, our lowest recovery efficiency has been for NP carcasses. However, recovery efficiency for NP carcasses was comparable to recovery efficiency for IB carcasses in 2022. Nonetheless, NP females may be spawning further upstream in reaches we cannot realistically access or survey infrequently. Continued surveys upstream of the County Line Bridge and opportunistic surveys whenever and wherever possible remains important.

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 to 2021, the median distance NP female carcasses were recovered also declined to 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 and 2021, the median recovery distance of NP female carcasses increased somewhat along with the number of NP females released (Table 5). In general, IB carcasses were recovered closer to the weir than NP carcasses. Although only 16 IB fish were passed above the weir, they were distributed higher in the system than in 2021. Comparisons between 2022 and previous years are difficult because we were limited to single pass surveys in reaches above County Line Bridge. It is possible the true median upstream distance for NP and IB carcasses was underestimated because of sampling bias (i.e., Ross Fork Fire closure).

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 4: 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 River 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 into 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 these critical questions. The three Chinook Salmon hatcheries in the program 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 presented (Figure 12). Our objective for this chapter is to track the productivity of a parental lineage through two generations. Here we focus on summarizing the productivity of the F_1 parental lineages that spawned in 2017 and the F_2 productivity of natural and supplementation adults that spawned naturally in 2012. Additionally, information is presented on F_2 productivity of naturally spawning segregated adults if available.

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 2017 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_1 offspring from the first female lineage (P_1; Figure 3). Adult females contributing to the P_1 generation were trapped at the weir and either spawned into the IB line (P_{1i}; Figure 3) or passed above the weir for natural spawning (P_{1n}). All adults brought into the IB broodstock were genetically sampled for parentage-based tagging (PBT), genetically tagging their progeny to identify their female parent and brood year of origin (Steele et al. 2011, 2018b). The F_1 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 (2017), DNA was also collected from all adults passed upstream of the weir, which allowed us to assign adult F_1 returns from natural production to a mother and brood year using the same techniques. The F_1 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_2) of offspring return, we compute the same metrics for this subsequent generation. The number of observed and expected female F_2 offspring originating from a spawn year (SY) at each location are compared. This provides an evaluation of observed natural-origin abundance to a scenario in which supplementation is absent. The number of observed F_2 offspring is simply the sum of natural-origin offspring originating from IB and natural-origin fish. The number of expected F_2 offspring reflects a scenario in which natural-origin females were not incorporated into the IB program for a corresponding SY. The expected number of F_2 offspring is calculated as:

$$F_{2e} = (P_1) * (F_{1r}) * (F_{2r})$$

where P_1 is the total number of female parents sampled in the SY of interest, F_{1r} is the overall recruitment rate of F$_1$ female offspring by the P$_1$ females, and F_{2r} is the overall recruitment rate of F$_2$ female offspring by the F$_1$ females.

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. To increase the rate of detection of natural-origin offspring we began implementing a single-parentage assignment methodology which allows offspring to still be identified when one parent is unsampled. Single-parentage analysis followed protocols described in Steele et al. (2022) and was conducted using the R package gRandma (https://github.com/delomast/gRandma).
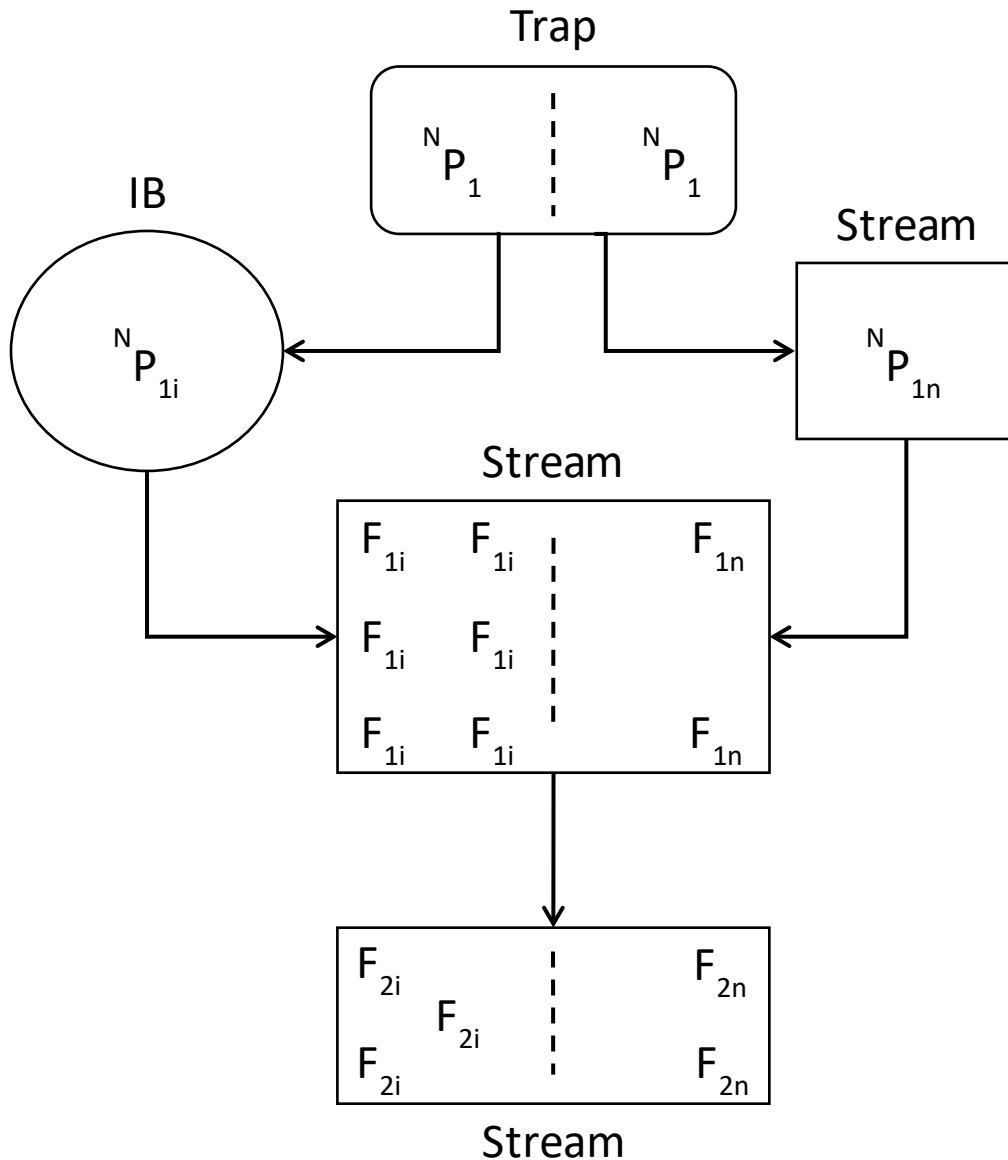
Figure 12. Conceptual diagram depicting how the supplementation effect is tracked through time and how replacement rate between natural spawning stream and the integrated broodstock (IB) can be compared. In this scenario, two natural-origin females (NP_1) are trapped, and one is brought into an IB program, 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.

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 always originated 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_1

The maximum number of offspring returned per female was 11 for an integrated cross and three for a natural spawning female (Figure 13). The maximum number of female offspring returned per female was eight for an integrated cross and one for a natural spawning female (Figure 13). The mean number of adult offspring that returned from females (R/F) used in integrated crosses in BY2017 was 3.1 (Figure 15). The mean number of adult offspring that returned from natural-origin females (R/F) released above the weir in BY2017 was 0.1 (Figure 15). The mean number of adult female offspring that returned from females (RF/F) used in integrated crosses in BY2017 was 1.5 (Figure 15). The mean number of female offspring originating from natural-origin females (RF/F) passed above the weir in SY2017 was 0.04 (Figure 15). The hatchery amplification observed in the F_1 generation ([IB mean RF/F] / [NP mean RF/F]) for BY2017 was 36.5 to 1 (Figure 15).
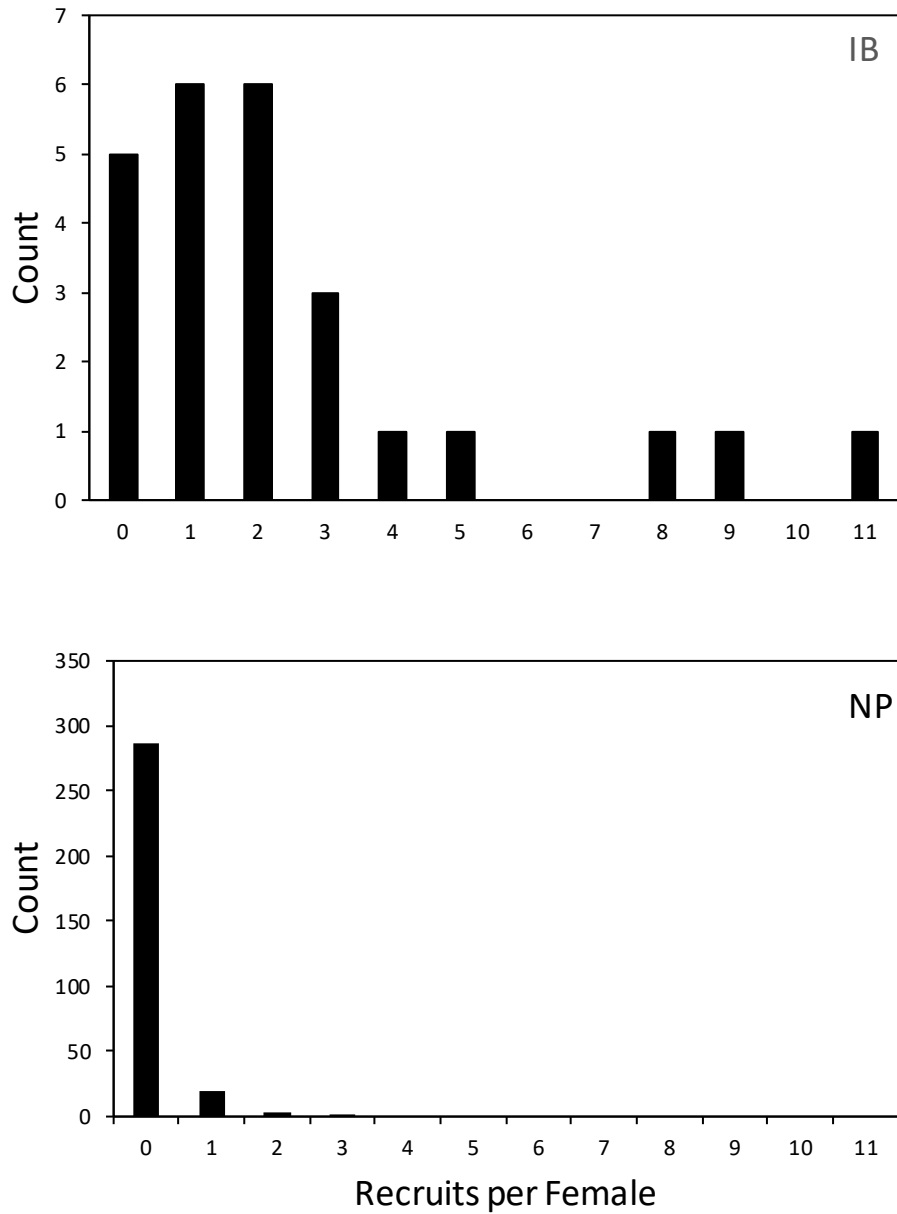
Figure 13. Frequency histograms of recruits per female from brood year 2017 to the Sawtooth Hatchery weir. The top panel represents females spawned in the integrated broodstock (IB). The bottom panel represents females spawning naturally (NP) upstream of the weir. Please note that the y-axes are not standardized.

Figure 14.   Frequency histograms of female recruits per female from brood year 2017 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. Please note that the y-axes are not standardized.
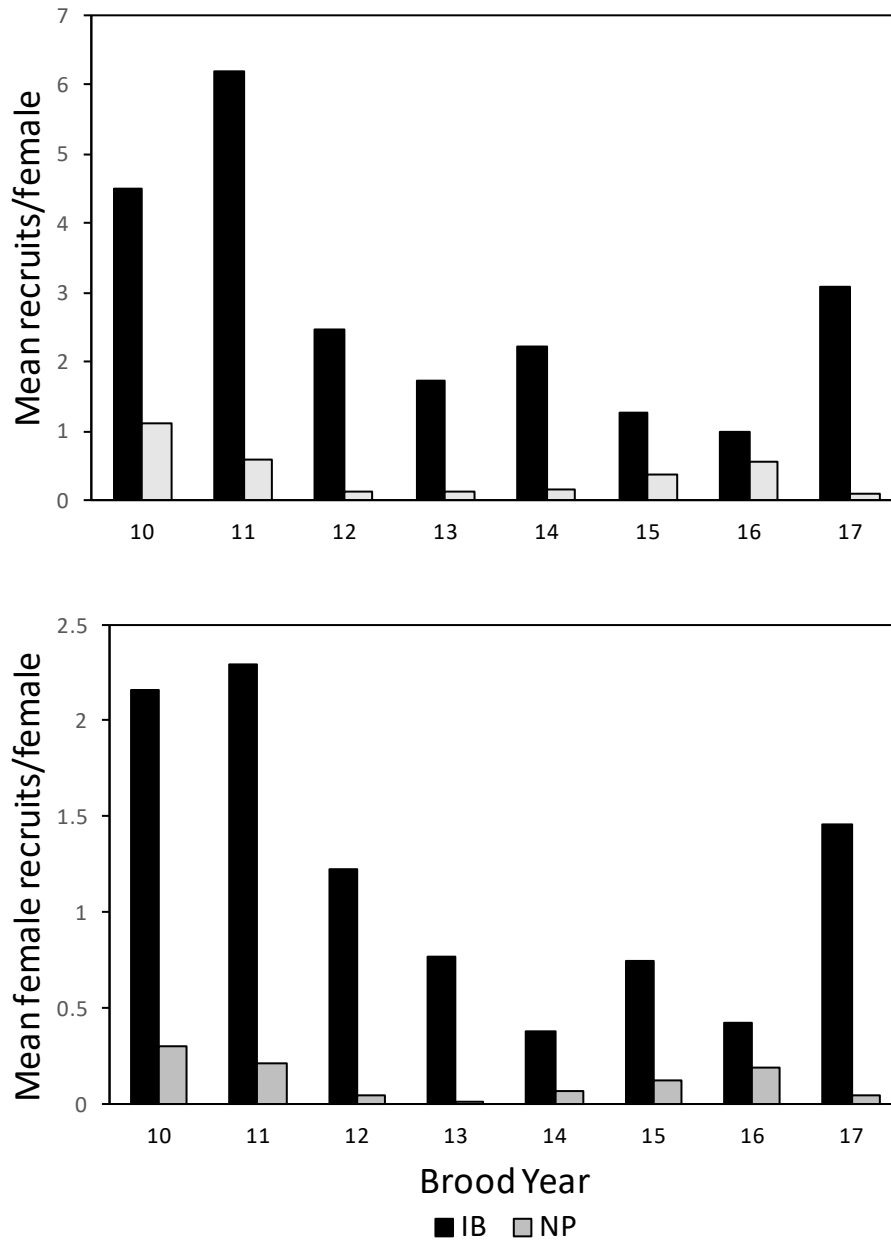
Figure 15. Mean number of recruits per female from the Sawtooth Hatchery integrated broodstock (IB; black bars) program and the natural population (NP; grey bars) in the upper Salmon River (upstream of the hatchery weir) summarized across brood years evaluated to date. All adult recruits (top) and female recruits only (bottom) are presented. Please note that the y-axes are not standardized.

McCall F_1

The maximum number of offspring returned per female was 14 for an integrated cross and eight for a natural spawning female (Figure 16). The maximum number of female offspring

returned per female was 10 for an integrated cross and four for a natural spawning female (Figure 17). The mean number of adult offspring that returned from females (R/F) used in integrated crosses in BY2017 was 5.9 (Figure 18). The mean number of adult offspring that returned from natural origin females (R/F) released above the weir in BY2017 was 0.4 (Figure 18). The mean number of adult female offspring that returned from females (RF/F) used in integrated crosses in BY2017 was 3.1 (Figure 18). The mean number of female offspring originating from natural-origin females (RF/F) passed above the weir in BY2017 was 0.2 (Figure 18). The hatchery amplification observed in the F1 generation for BY2017 was 16.7 to 1 (Figure 18).
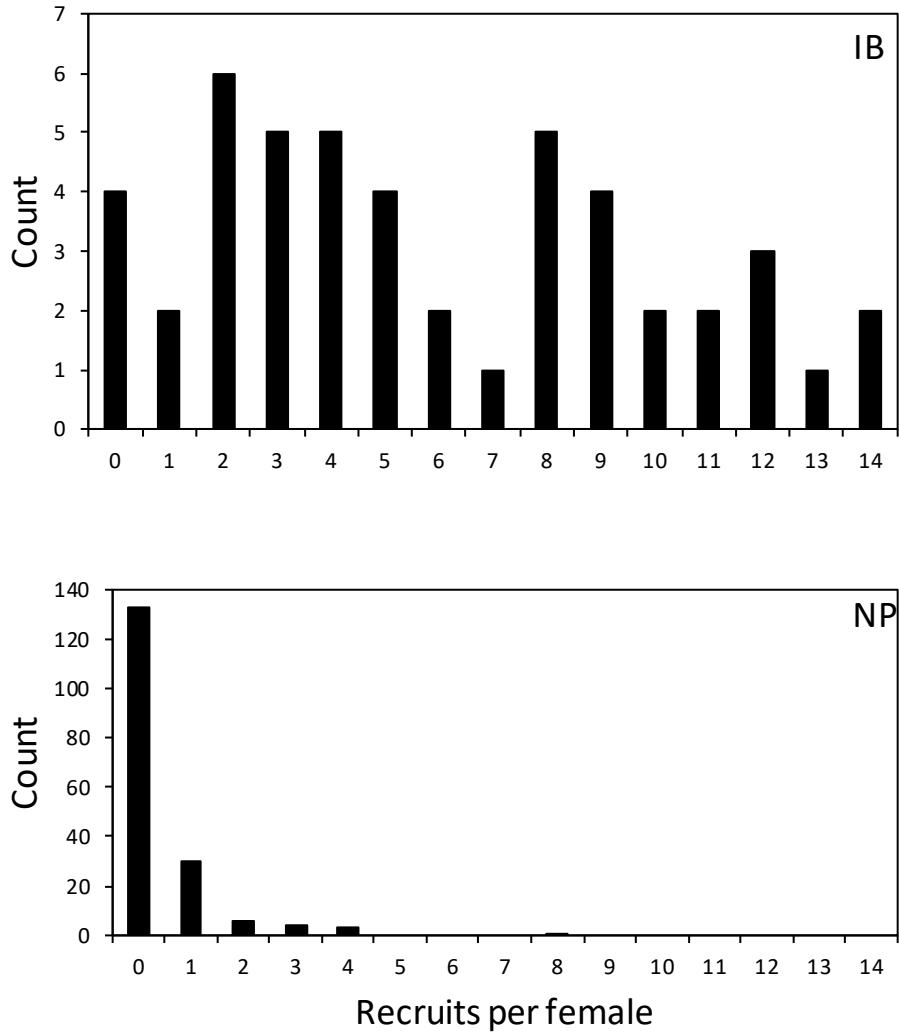
Figure 16.   Frequency histograms of recruits per female from brood year 2017 to the South Fork Salmon River weir. The top panel represents females spawned in the integrated broodstock (IB). The bottom panel represents females spawning naturally (NP) upstream of the weir. Please note that the y-axes are not standardized.
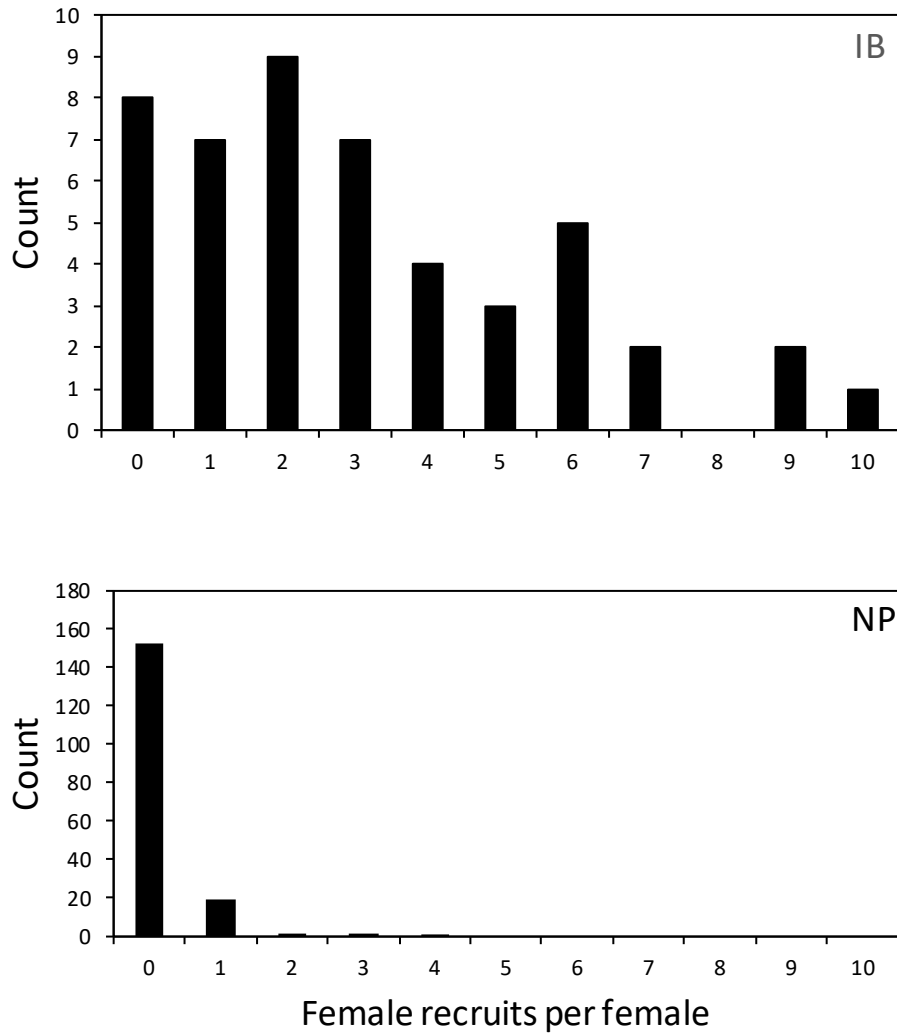
Figure 17.   Frequency histograms of female recruits per female from brood year 2017 (to the South Fork Salmon River weir). The top panel represents females spawned in the integrated broodstock (IB). The bottom panel represents females spawning naturally (NP) upstream of the weir. Please note that the y-axes are not standardized.
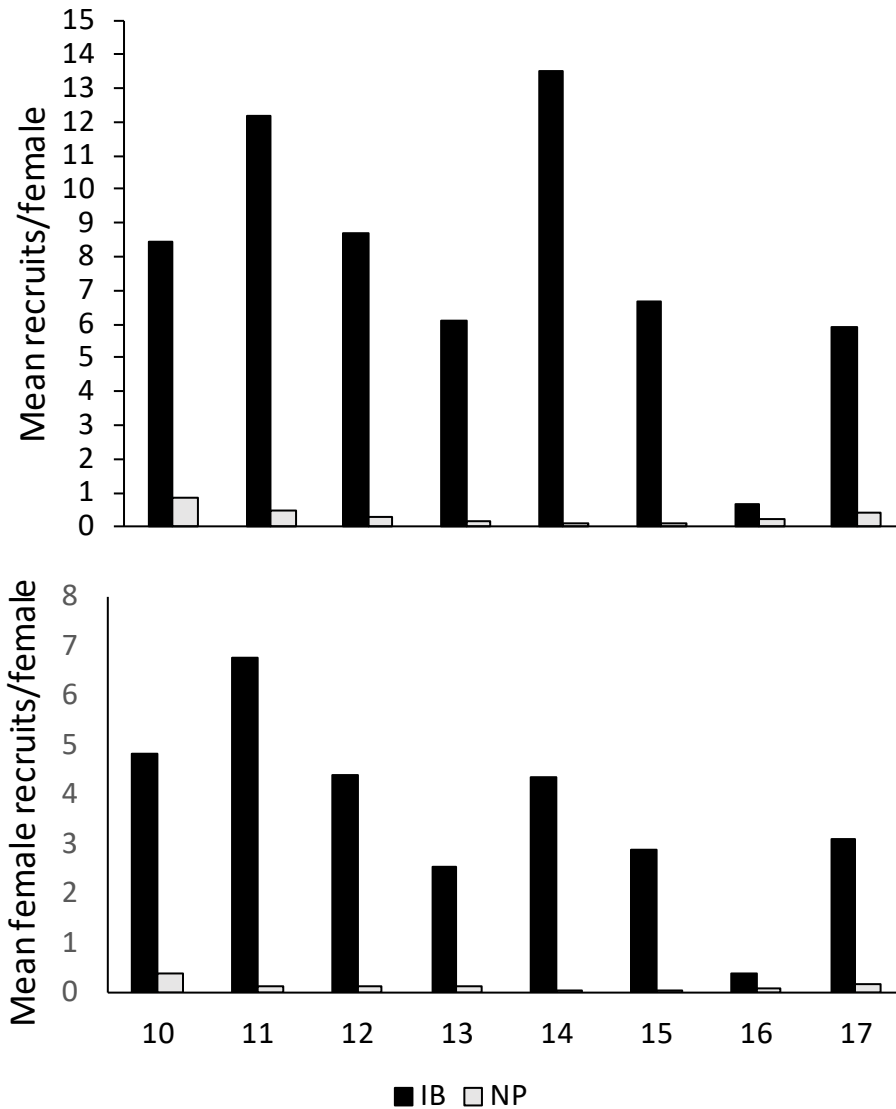
Figure 18.   Mean number of recruits per female from the McCall Fish Hatchery integrated broodstock (IB; black bars) program and the natural population (NP; grey bars) in the South Fork Salmon River (SFSR) upstream of the hatchery weir summarized across brood years evaluated to date. All adult recruits (top) and female recruits only (bottom) are presented. Please note that the y-axes are not standardized.

Pahsimeroi F$_1$

The maximum number of offspring returned per female was five for an integrated cross and three for a natural spawning female (Figure 19). The maximum number of female offspring returned per female was three for an integrated cross and two for a natural spawning female (Figure 20). The mean number of adult offspring that returned from females (R/F) used in integrated crosses in BY2017 was 0.9 (Figure 21). The mean number of adult offspring that

returned from natural origin females (R/F) released above the weir in BY2017 was 0.4 (Figure 21). The mean number of adult female offspring that returned from females (RF/F) used in integrated crosses in BY2017 was 0.6 (Figure 21). The mean number of female offspring originating from natural-origin females (RF/F) passed above the weir in BY2017 was 0.2 (Figure 21). The hatchery amplification observed in the F1 generation for BY2017 was 3.6 to 1 (Figure 21).
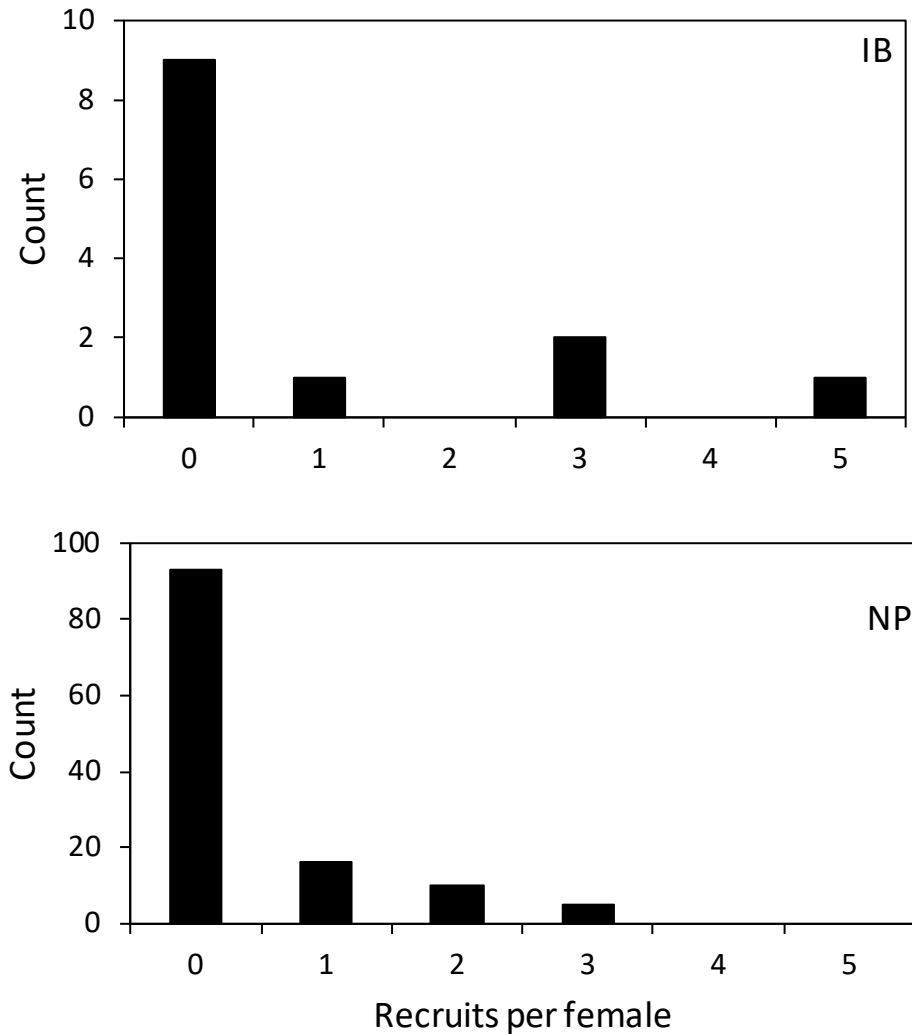
Figure 19. Frequency histograms of recruits per female from brood year 2017 to the Pahsimeroi Hatchery weir. The top panel represents females spawned in the integrated broodstock (IB). The bottom panels represent females spawning naturally (NP) upstream of the weir. Please note that the y-axes are not standardized.
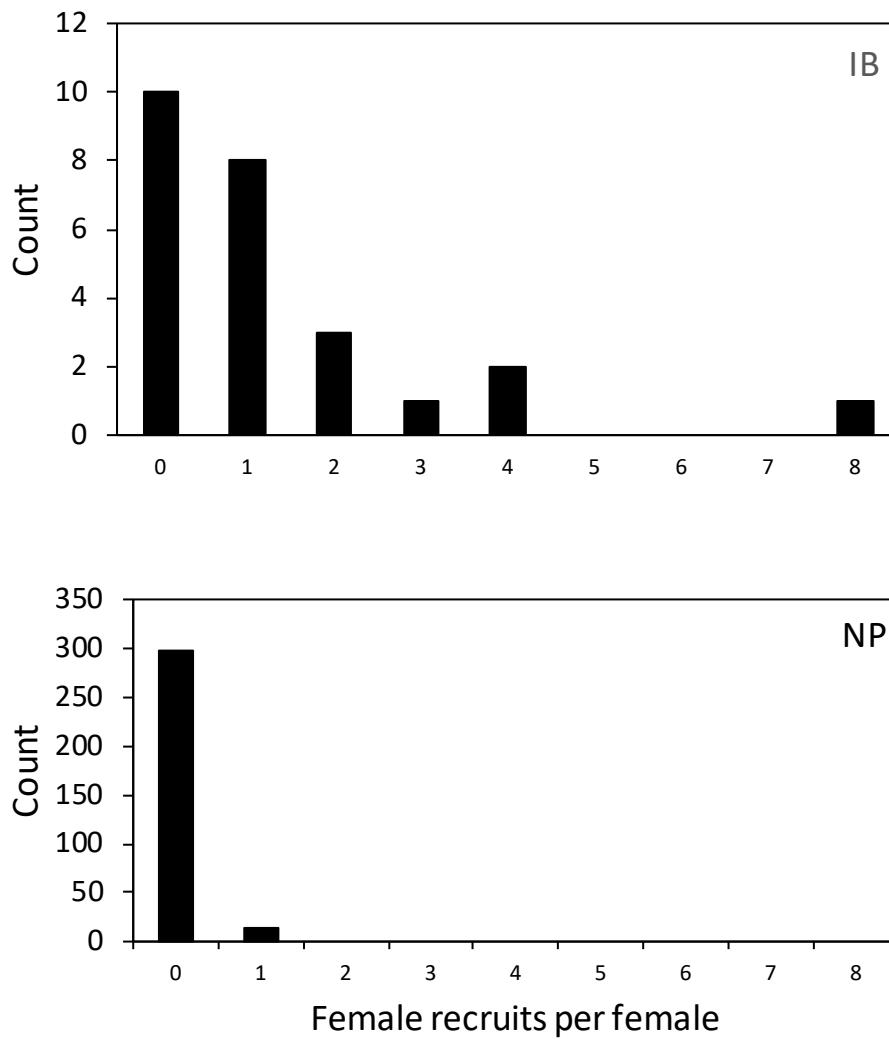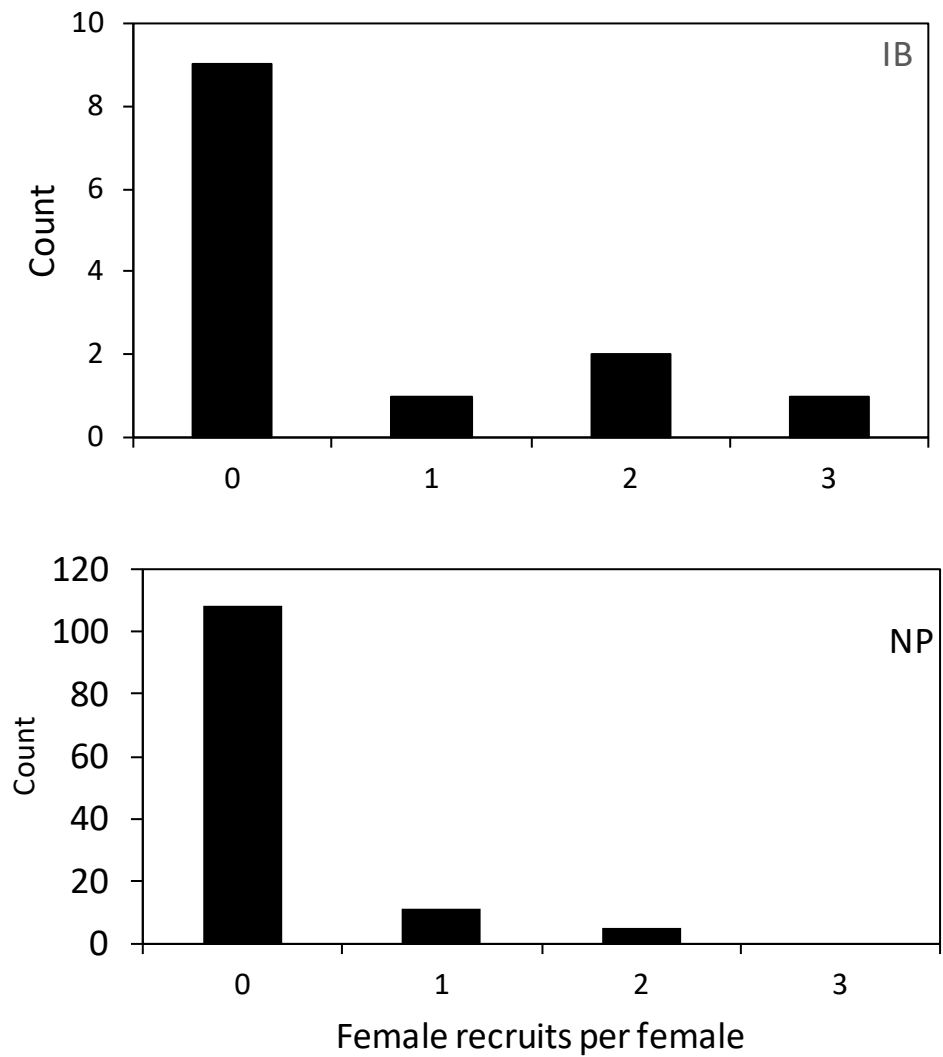
Figure 20.   Frequency histograms of female recruits per female from brood year 2017 to the Pahsimeroi Hatchery weir. The top panel represents females spawned in the integrated broodstock (IB). The bottom panel represents females spawning naturally (NP) upstream of the weir. Please note that the y-axes are not standardized.
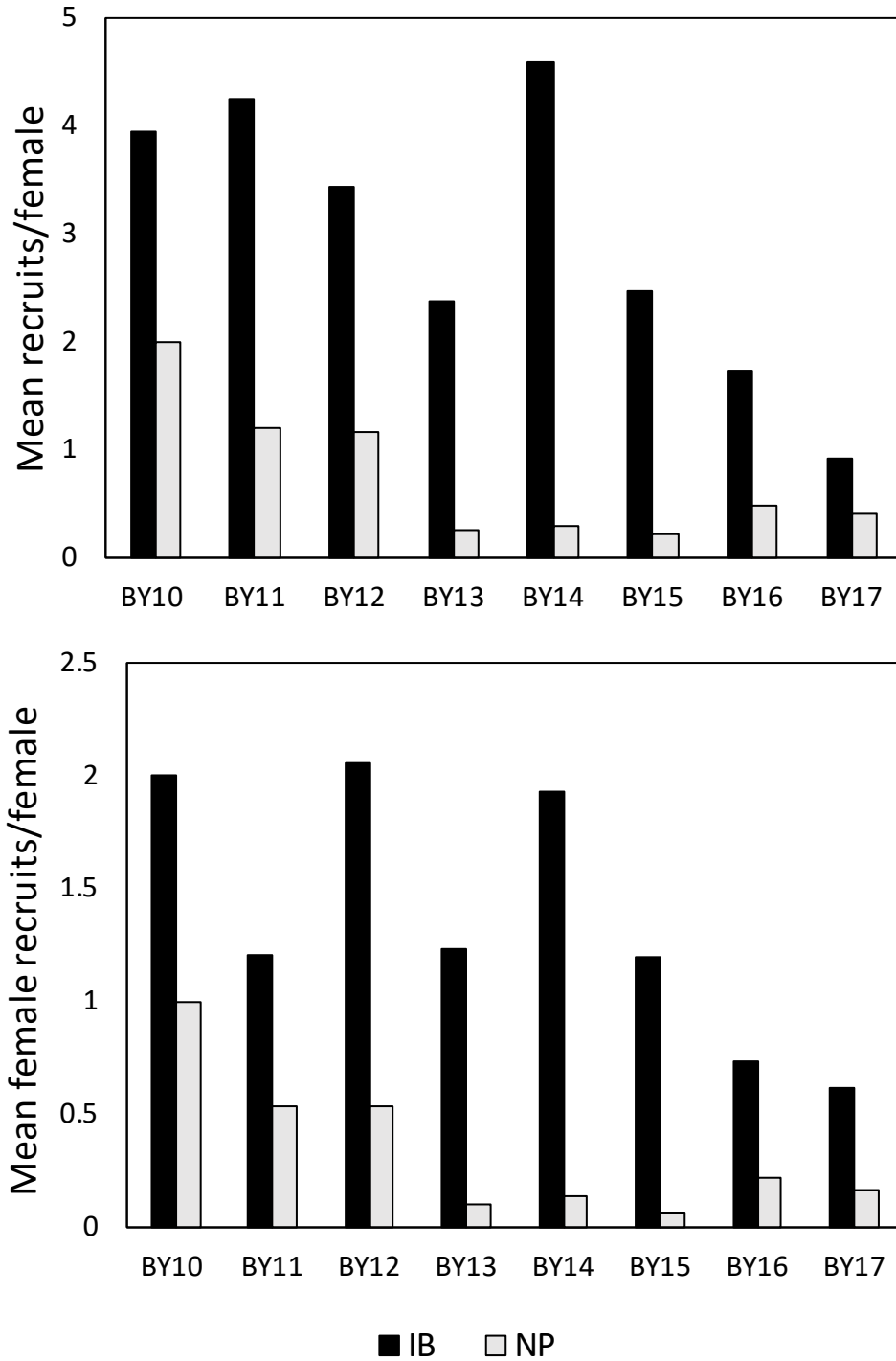
Figure 21.   Mean number of recruits per female from the Pahsimeroi Fish Hatchery integrated broodstock (IB; black bars) program and the natural population (NP; grey bars) in the Pahsimeroi River upstream of the hatchery weir summarized across brood years evaluated to date. All adult recruits (top) and female recruits only (bottom) are presented. Please note that the y-axes are not standardized.

F_2 Abundances

The program has demonstrated a large and consistent F_1 abundance boost within the integrated lineages compared to that of natural lineages. Implementation of the IB supplementation program has now been underway long enough for the abundances of F_2 returns to also be summarized. These early summaries coincided with a downturn in ocean conditions and low numbers of return (Figure 22, Appendix D). Reproductive patterns of F_2 offspring from both integrated and natural females have remained low over the evaluation period with recruits per female (R/F) and female recruits per female (RF/F) often approaching zero at all locations for all maternal origins. Despite this, the program is still demonstrating that it is producing an overall increase in the observed number of F_2 females at all locations (Table 6) and indicates that the IB program is resulting in an abundance boost of natural-origin fish. The program generates annual summaries on the F_2 abundance originating from subsequent broodyears and every calendar year generates another opportunity for comparisons and evaluations of the IB program.

For the evaluation period of SY2015–SY2017, abundances of F_2 offspring (both sexes) at the SFSR ranged from zero to two for F_1 segregated females, 19 to 59 for integrated females, and one to five for natural-origin females. Abundances at Sawtooth ranged from 20 to 61 for segregated females, four to 28 for integrated females, and 0 to 20 for natural-origin females. Abundances at Pahsimeroi ranged from one to 26 for segregated females, five to 31 for integrated females, and 13 to 39 for natural origin females (Appendix D).

For the same period, the R/F of F_2 offspring per F_1 female at the SFSR ranged from zero to 0.11 for segregated fish, 0.07 to 0.4 for integrated fish, and 0.08 to 0.17 for natural-origin fish. The R/F of F_2 offspring per F_1 female at Sawtooth ranged from 0.2 to 0.55 for segregated fish, 0.16 to 0.41 for integrated fish, and N/A to 0.76 for natural-origin fish. The R/F of F_2 offspring per F_1 female at Pahsimeroi ranged from 0.13 to 0.45 for segregated fish, 0.1 to 0.44 for integrated fish, and 0.23 to 0.52 for natural-origin fish (Figure 22, Appendix D).

Finally, the RF/F of F_2 offspring per F_1 female at the SFSR ranged from zero to 0.05 for segregated fish, 0.04 to 0.19 for integrated fish, and zero to 0.13 for natural-origin fish. The RF/F of F_2 offspring per F_1 female at Sawtooth ranged from 0.08 to 0.21 for segregated fish, 0.09 to 0.17 for integrated fish, and N/A to 0.24 for natural-origin fish. The RF/F of F_2 offspring per F_1 female at Pahsimeroi ranged from 0.13 to 0.17 for segregated fish, 0.08 to 0.19 for integrated fish, and 0.04 to 0.17 for natural-origin fish (Figure 22, Appendix D).

Observed abundance of female F_2 offspring was also compared to an expected scenario in which natural-origin females were not incorporated into the IB program. Differences between the values can be used to evaluate the effects of an IB program on natural production. Differences in abundances at the SFSR showed an increase for all spawn years and ranged from 6.3 to 29. Differences in abundances at Sawtooth ranged from -0.8 to 3.9. Differences at Pahsimeroi ranged from 1.6 to 9.1 (Table 6).
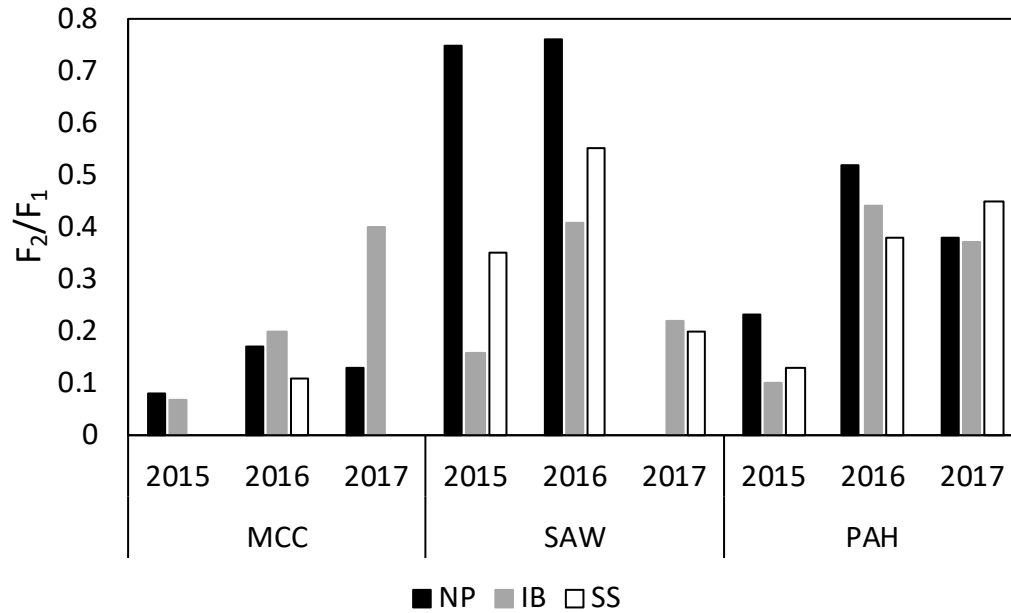
Figure 22.   The total F_2 offspring per F_1 female parent (F_2 / F_1) at the three study sites. Patterns indicate comparable numbers of offspring between NP and IB at McCall, substantially larger numbers of offspring from NP at Sawtooth, and larger numbers of offspring from NP at Pahsimeroi during two of the first three brood years of evaluation.

Table 6.   Number of observed (Obs.) and expected (Exp.) female F_2 offspring originating from three spawn years (SY) at each location. The number of observed female F_2 offspring is the combination of natural-origin offspring originating from IB and natural-origin fish. The number of expected female F_2 offspring reflects a scenario in which natural-origin females were not incorporated into the IB program for the corresponding SY. No natural-origin female fish were passed above the Sawtooth weir in SY2017 precluding any comparison (N/A).

Location	SY	No. Obs. F_2	No. Exp. F_2	Diff.
MCC	2015	11	4.7	6.3
	2016	27	7.7	19.3
	2017	29	0	29
SAW	2015	5	5.8	-0.8
	2016	13	9.1	3.9
	2017	3	N/A	N/A
PAH	2015	6	4.4	1.6
	2016	25	15.9	9.1
	2017	11	4.0	7.0

The maximum number of offspring produced has varied from three to 47 in the IB and one to 18 for natural origin fish (Table 7). For brood year 2017, the maximum number of offspring produced in the IB was 11 for Sawtooth, 14 for McCall, and 5 for Pahsimeroi. Natural origin females returned a maximum number of offspring of eight for McCall and three for Sawtooth and Pahsimeroi. Fold increases in mean abundance of IB-origin adult recruits compared to natural-origin fish spawning upstream of hatchery weirs was 31.7 for Sawtooth, 14.1 for McCall, and 2.2 for Pahsimeroi (Table 8).

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

Weir	Brood year															
	2010		2011		2012		2013		2014		2015		2016		2017	
	IB	NP	IB	NP	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	8	5	11	3
S.F.Salmon	15	14	20	18	12	6	21	2	47	3	20	1	3	4	14	8
Pahsimeroi	30	12	43	5	31	2	9	2	13	4	7	3	7	2	5	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.

Weir	Brood year							
	2010	**2011**	**2012**	**2013**	**2014**	**2015**	**2016**	**2017**
Sawtooth	4.1	10.6	18.9	15.9	13.5	3.6	1.8	31.7
S.F. Salmon	9.8	24.5	33.6	42.4	133.6	98.7	3.1	14.1
Pahsimeroi	2.0	3.5	2.9	9.1	15.8	11.7	3.6	2.2

DISCUSSION

This evaluation period continues to show that P_1 females spawned in the IB return more F_1 offspring than their naturally spawning counterparts. This pattern is perhaps not surprising given the protective benefits of hatchery rearing in the early life stages. This effect is evident in the frequency histograms that summarize recruits per spawner. The patterns for IB females consistently show a hatchery boost in which many IB 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 and has also been consistent through time.

Females spawned into the IB and naturally spawning females exhibited a similar pattern in which some females returned large numbers of offspring. The maximum number of offspring produced in the IB and naturally spawning lineages show that the size of such "super families" is larger in the IB lineage than in natural-origin lineages. Such 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" tend to be larger in the IB program will indeed create 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. In addition, 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 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 has ranged from a 133-fold difference at the SFSR facility to a 1.8-fold difference at Sawtooth. The abundance boost is often largest at SFSR. These abundance boosts are relative to natural production at each site and can be affected by low numbers of natural recruits per spawner at each facility. It is possible that progeny from both natural-origin production and the IB lineage are not fully enumerated because offspring that do not recruit to the weir cannot be

sampled or included in these estimates. However, this program attempts to account for this by sampling carcasses of ad-intact fish found below the weir, which includes both offspring from the IB program and offspring from natural production. Carcass surveys happened primarily 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, but the NPT conducts carcass surveys in that reach and provides tissue samples of AD intact carcasses for evaluation. It is feasible to evaluate the proportion of fish that "drop out" 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.

The ultimate evaluation goal of the IB program is to determine whether it can boost natural abundance. This evaluation comprises two parts. The first is to quantify any differences in abundance of F_1 offspring between IB fish reared in a hatchery and natural spawning fish. The expectation is that IB fish will produce, on average, more offspring than the naturally spawning fish. Results from this evaluation period continue to show a larger number of hatchery-reared fish (F_1) relative to that of naturally spawning fish and demonstrates a hatchery amplification of a female's progeny and a subsequent abundance boost in the F_1 progeny originating from the IB program. It can be argued that these progeny experienced domestication selection during their rearing in the hatchery and may ultimately be less fit than their natural-origin counterparts when they spawn in a natural environment. Therefore, evaluations on the relative productivity of the F_1 offspring in the wild are needed. We now have enough data for the second step of the evaluation which compares the reproductive success in the wild of these hatchery-reared IB fish to that of natural-origin fish.

Indications from other studies suggest that hatchery-reared fish can display lower levels of reproductive success compared to that of natural-origin fish (Araki et al 2007, Frankham 2008). However, we evaluate whether the demographic boost experienced in the F_1 generation is large enough to overcome the predicted loss of productivity in the F_2 generation. The expectation is that the number of F_2 progeny produced by the F_1 generation will be greatest within the natural-origin lineage, followed by the IB lineage, and finally the segregated lineage. Thus, we expect F_2/F_1 to follow a pattern of NP > IB > SS. However, we see some deviations from this prediction among the lineages at the three locations. At SFSR the R/F values between natural-origin and IB fish are undifferentiated or IB > NP for the three years summarized suggesting that hatchery rearing of the IB did not negatively influence their reproductive success relative to the natural-origin fish. It also suggests that the demographic boost of F_1 offspring will carry over into the F_2 generation. However, at Sawtooth we see that the R/F is markedly higher within the natural lineage. This suggests that the demographic boost in the F_1 generation may not be enough to overcome the loss of productivity in the F_2 generation, and will be addressed below. Finally, at Pahsimeroi the R/F showed a trend with the largest differences between IB and natural-origin fish occurring in SY2015 but with differences shrinking in subsequent years. These early data suggest that some locations are experiencing differences in R/F rates in the F_2 generation while others are not.

The second part of the IB evaluation lies within the differences between the observed number of F_2 progeny and an expected number of offspring that would have been present in the

absence of a supplementation program (Table 6). These data reveal several patterns. The SFSR has the largest and most consistent demographic boost in natural production. This is consistent with our observation of similar F_2/F_1 values between natural-origin and IB fish females at this location and suggests that the demographic boost in the F_1 generation carried over into the following generation. Sawtooth had a slightly negative value for the first year, suggesting that incorporating natural-origin females into the IB program at this location did not produce a demographic boost. However, the second data point at Sawtooth indicates an increase in the F_2 abundance. No natural-origin female fish were passed above the Sawtooth weir in SY2017 precluding any evaluation and resulting in missing data. A similar pattern is observed at Pahsimeroi where a low value was recorded for the first year but values increased modestly in the second and third year. Overall, there are several conclusions from these data. First, there is indeed a demographic boost for most locations in most years. Second, the magnitude of the boost is increasing through time, which may be associated with improving ocean conditions. Third, the boost is most pronounced at SFSR.

The ultimate objective of this evaluation is to determine if an IB program results in a demographic boost in natural production. The IB program pursues this objective by incorporating natural-origin adults into the broodstock and then allowing the progeny to spawn naturally. We can now determine if incorporating those natural-origin adults ultimately produced a demographic boost in the natural population by comparing 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 have now produced some F_2 progeny (Figure 13, Appendix D). Early results indicate that natural-origin lineages performed better than the IB lineages at two of the three locations and the two performed similarly at the third. However, overall numbers of female F_2 returns are generally quite low, making meaningful comparisons among the lineages difficult. Regardless, some trends in the data are forming. The data indicate that the demographic boost in natural production is largest in the most recent years of the evaluation. This pattern is probably driven by improving ocean conditions. This suggests that in years of poor ocean conditions, an IB program is not likely to achieve a demographic boost in natural production but when conditions are favorable, the program has the potential to generate substantial numbers of returning adults. It will be important to monitor these trends in the coming years to determine if the boost continues to be correlated with ocean conditions.

CONCLUSION

The ultimate objective of this evaluation is to determine if an IB program results in a demographic boost in the naturally spawning population. The IB program pursues its objectives by incorporating natural-origin adults into the broodstock and then allowing the progeny to spawn naturally. We can now begin to determine if incorporating those natural-origin adults ultimately produced a demographic boost in the natural population by comparing the abundance of F_2 offspring representing each of the three lineages (natural-origin, integrated, and segregated) allowed to spawn in the wild. All three lineages have now produced some F_2 progeny. Early results indicate that natural-origin lineages performed better at the three locations but overall numbers of female F_2 returns are generally quite low, making meaningful comparisons among the lineages difficult. Regardless, some trends in the data are forming. The data indicate that a demographic boost in natural abundance is occurring and is largest in the most recent year of the evaluation. This pattern is probably driven by improving ocean conditions. This suggests that in years of poor ocean conditions an IB program is not likely to achieve a demographic boost in natural abundance, but when conditions are favorable the program has the potential to generate

substantial numbers of returning adults. It will be important to monitor these trends in the coming years to determine if the boost continues to be correlated with ocean conditions.

ACKNOWLEDGEMENTS

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APPENDICES

Appendix A.   Survival estimates for integrated (IB) and segregated (SS) juveniles at various checkpoints at Sawtooth Fish Hatchery. Checkpoints include fecundity (average number of green eggs), eye-up rate, marking (fish are enumerated while fin clips and tags are applied), and release. No statistical differences were observed between IB and SS fish.

Brood year	Origin	Fecundity	Eye-up	Eye-up to mark	Mark to release	Green to release
2015	IB	3525.1	97.4	85.2	98.9	82.1
	SS	3645.5	97.3	94.0	99.3	90.8
2016	IB	5220.6	96.6	97.4	90.1	84.8
	SS	4680.0	96.8	99.4	100.0	96.8
2017	IB	4365.1	84.4	94.6	99.7	79.6
	SS	4371.9	89.7	85.4	99.1	76.0
2018	IB	4066.6	96.7	90.7	98.8	86.7
	SS	4015.9	95.1	97.9	99.5	92.6
2019	IB	4192.5	90.2	98.8	99.4	88.6
	SS	4454.0	91.9	98.4	99.2	89.7
2020	IB	4538.6	95.8	97.4	99.0	92.4
	SS	4165.4	94.8	99.0	98.9	92.9
2021	IB	4501.9	94.4	95.8	98.8	89.3
	SS	4411.4	95.2	83.6	94.7	75.4

Appendix B. Survival estimates for integrated (IB) and segregated (SS) juveniles at various checkpoints at Pahsimeroi Fish Hatchery. Checkpoints include fecundity (average number of green eggs), eye-up rate, marking (fish are enumerated while fin clips and tags are applied), and release. No statistical differences were observed between IB and SS fish.

Brood year	Origin	Fecundity	Eye-up	Eye-up to mark	Mark to release	Green to release
2015	IB	4922.6	91.3	94.1	99.6	85.5
	SS	4397.3	91.3	87.3	99.5	79.4
2016	IB	4818.1	90.0	93.3	99.4	83.4
	SS	4930.9	86.6	85.1	99.4	73.3
2017	IB	5645.5	89.3	60.6	96.3	52.1
	SS	4896.1	89.9	96.6	91.3	79.2
2018	IB	4740.8	95.5	88.7	97.0	82.1
	SS	4597.9	95.0	88.7	99.0	83.3
2019	IB	5053.8	92.4	97.0	96.2	86.2
	SS	4479.5	90.8	90.9	93.8	77.4
2020	IB	4474.6	95.3	94.8	99.8	90.2
	SS	4168.5	89.2	92.9	99.6	83.0
2021	IB	3849.4	83.2	99.6	99.7	82.7
	SS	5054.7	85.8	84.3	98.7	71.4

Appendix C. Survival estimates for integrated (IB) and segregated (SS) juveniles at various checkpoints at McCall Fish Hatchery. Checkpoints include fecundity (average number of green eggs), eye-up rate, marking (fish are enumerated while fin clips and tags are applied), and release. No statistical differences were observed between IB and SS fish.

Brood year	Origin	Fecundity	Eye-up	Eye-up to mark	Mark to release	Green to release
2015	IB	4599.2	85.6	92.0	99.8	78.6
	SS	3758.7	92.0	91.4	99.7	83.8
2016	IB	4207.8	89.3	87.7	98.6	77.2
	SS	4337.3	89.1	88.0	99.0	77.6
2017	IB	4127.6	89.8	91.3	98.5	80.7
	SS	4340.3	95.5	90.1	99.0	85.2
2018	IB	3838.0	90.4	91.4	99.8	82.5
	SS	4038.9	91.9	69.9	99.9	83.8
2019	IB	3776.8	87.9	94.9	99.1	82.7
	SS	3872.2	88.3	73.1	99.1	84.7
2020	IB	3852.5	94.0	91.0	99.9	85.4
	SS	3805.6	91.3	96.5	99.9	88.0
2021	IB	3799.6	82.6	88.3	99.8	72.8
	SS	4038.2	92.7	96.3	99.7	89.0

Appendix D. Number of F_1 females by origin passed above the South Fork Salmon (MCC), Sawtooth (SAW), and Pahsimeroi (PAH) weirs for natural spawning in 2015, 2016, 2017 and the total number of their F_2 progeny (includes age 3, 4, and 5 offspring) that subsequently returned to the weir. Parental origins include natural (NP), integrated (IB), and segregated (SS) lineages. The total recruits of F_2 offspring per F_1 female (R / F) and the female recruits of F_2 offspring per F_1 female (RF / F) is also presented.

		F_1 Female			F_2 Progeny			F_2 / F_1
Location	SY	Origin	N	Male	Female	Total	
MCC	2015	NP	39	2	1	3	0.08
	2015	IB	271	9	10	19	0.07
	2015	SS	2	0	0	0	0.00
	2016	NP	30	1	4	5	0.17
	2016	IB	300	36	23	59	0.20
	2016	SS	19	1	1	2	0.11
	2017	NP	8	1	0	1	0.13
	2017	IB	156	30	29	59	0.40
	2017	SS	9	0	0	0	0.00
SAW	2015	NP	20	13	2	15	0.75
	2015	IB	32	2	3	5	0.16
	2015	SS	57	12	8	20	0.35
	2016	NP	17	9	4	13	0.76
	2016	IB	68	19	9	28	0.41
	2016	SS	111	38	23	61	0.55
	2017	NP	0	0	0	0	NA
	2017	IB	18	1	3	4	0.22
	2017	SS	101	12	8	20	0.20
PAH	2015	NP	56	11	2	13	0.23
	2015	IB	51	1	4	5	0.10
	2015	SS	8	0	1	1	0.13
	2016	NP	75	26	13	39	0.52
	2016	IB	71	19	12	31	0.44
	2016	SS	8	2	1	3	0.38
	2017	NP	13	4	1	5	0.38
	2017	IB	52	9	10	19	0.37
	2017	SS	58	16	10	26	0.45

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