Final

2023 Annual Operating Plan

HATCHERY AND SUPPLEMENTATION PROGRAM
NORTH FORK LEWIS RIVER

Prepared by
the
North Fork Lewis River
Aquatic Technical Subgroup

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DEFINITION OF TERMS AND ACRONYMS

Area Under the Curve (AUC): a method for estimating salmon escapement by dividing the integral of the escapement curve by the average residence time in the survey area.

Annual Operating Plan (AOP): An annual planning document that describes the methods and protocols needed to implement the North Fork Lewis River Hatchery and Supplementation Plan and Program.

Aquatic Coordination Committee (ACC): Committee formed after signing of the North Fork Lewis River Settlement Agreement (Settlement Agreement) and composed of its signatories. Many of the measures contained in the Settlement Agreement require review and consultation with the ACC prior to implementation. Thus, the committee acts as the governing body for implementing aquatic measures contained within the Settlement Agreement. The committee also approves aquatic habitat funds on an annual basis.

Aquatic Monitoring and Evaluation Plan (AMEP): A comprehensive planning document required by the North Fork Lewis River Settlement Agreement (Section 9). The purpose of the AMEP is to develop methods to evaluate aquatic monitoring and evaluation objectives contained within the North Fork Lewis River Settlement Agreement. These objectives relate to fish passage, reintroduction outcome goals, anadromous and resident species monitoring, and development of the North Fork Lewis River Hatchery and Supplementation Plan.

Aquatic Technical Subgroup (ATS): A subgroup of the Aquatic Coordination Committee intended provide technical recommendations to the ACC. The ATS is focused primarily on developing and reviewing technical aspects of plans, reports and monitoring strategies or objectives related to the Hatchery and Supplementation and Aquatic, Monitoring and Evaluation programs.

Bacterial Coldwater Disease (BCD): Bacterial disease of salmonid fish caused by *Flavobacterium psychrophilum*.

Bacterial Kidney Disease (BKD): Bacterial disease of salmonid fish caused by *Renibacterium salmoninarum.*

Bayesian Goodness of Fit (GOF): a test used to determine whether sample data are consistent with a hypothesized distribution.

Blank wire tag (BWT): A small wire that is uncoded (blank), inserted in the snout of fish, and detectible with handheld wire detection wands or devices. BWT are specific to the integrated late winter steelhead supplementation program and all BWT positive fish are of hatchery origin (HOR).

Brood year (BY): year in which spawning occurs, used to track a single cohort over time.

Coded wire tag (CWT): A small wire with unique codes etched onto the wire, inserted in the snout of fish, and detectible with handheld wire detection wands or devices.

Coefficient of Variation (CV): The ratio of the population standard deviation (σ) to the population mean (μ) _or_ in instances when only a sample of data from the population is available CV is estimated by using the sample standard deviation (S) to the sample mean ($\bar{x}$) which shows the extent of variability in relation to the mean of the population. The absolute value of the CV, sometimes known as relative standard deviation, is expressed as a percentage.

- Population CV = σ/μ
- Sample CV = $s/\bar{x}$

Columbia Basin PIT Tag Information System (PTAGIS): A regional database that stores and tracks data from fish with passive integrated transponder (PIT) tags.

Condition factor (K): Fulton's condition factor, K, is a measure of individual fish health that assumes the standard weight of a fish is proportional to the cube of its length:

$$K = 100\left(\frac{W}{L^3}\right)$$

where *W* is the whole body wet weight in grams and *L* is the length in centimeters; the factor 100 is used to bring K close to a value of one (Fulton, 1904).

Distinct Population Segment (DPS): A population or group of populations that is discrete from other populations of the species and significant in relation to the entire species. This along with Evolutionarily Significant Unit (ESU) are used to define Endangered Species Act-listed species (DPS for steelhead and ESU for salmon species).

Effective Population Size (N_e): The average size of a population in terms of the number of individuals that can contribute genes equally to the next generation. Therefore, the effective population size is typically smaller than the actual census size of the population.

Enzyme-Linked Immunosorbent Assay (ELISA): This test uses antibodies and color change to identify viral antigens present in sampled fish tissues.

Ecosystem Diagnostic and Treatment (EDT) model: An analytical habitat-based model that evaluates environmental constraints on a fish population(s) and used to predict the carrying capacity or production potential of specific areas of the North Fork Lewis River such as upstream of Swift Dam.

Endangered Species Act (ESA): Passed in 1973, this piece of United States legislation was designed to protect species from extinction as well as the ecosystems upon which they depend. Listed species are classified as "threatened" or "endangered." The ESA is administered by the U.S. Fish and Wildlife Service and the National Marine Fisheries Service.

Evolutionarily Significant Unit (ESU): A distinct population unit that is reproductively isolated and an important component for the legacy of a species, considered a separate “species” for the purposes of conservation.

F1 generation: First generation offspring, in this case, typically referring to offspring of fish spawned in the hatchery and therefore of hatchery-origin.

F2 generation: Offspring of F1 parents that have spawned naturally and therefore of natural-origin.

Feed conversion ratio (FCR): The amount of feed an animal must consume to gain one kilogram of body weight.

Fish per pound (fpp): Number of juvenile salmon per pound batch weight

Floy tag: Visible tags with unique codes and colors applied to the dorsal side of fish to identify individual fish upon capture or through visual surveys. Floy tags are inserted near the posterior side of the dorsal fin and are intended to lock within the dorsal skeletal bones by means of a T-anchor.

Generalized Random Tessellation Stratified (GRTS) design: provides a probability sample with design-based variance estimators by establishing a spatially balanced, random sample allowing for unequal probability sampling to accommodate field implementation issues.

Hatchery and Genetic Management Plan (HGMP): a technical document that describes artificial propagation management strategies that ensure conservation and recovery of ESA-listed salmon and steelhead populations.

Hatchery and Supplementation Plan (H&S Plan): A 5-year planning document intended to provide the plan and process for implementing the goals of Section 8 (Hatchery and Supplementation Program) of the North Fork Lewis River Settlement Agreement.

Hatchery and Supplementation Program (Program): Defined in Section 8 of the Lewis River Settlement Agreement. The goals of the program are to support (i) self-sustaining, naturally producing, harvestable native anadromous salmonid species throughout their historical range in the North Fork Lewis River Basin, and (ii) the continued harvest of resident and native anadromous fish species.

Hatchery and Supplementation Subgroup (H&S Subgroup): Name of ATS prior to December 2018.

HOB: Hatchery Origin Broodstock.

HOR: Hatchery Origin Recruit.

HOS: Hatchery Origin Spawners.

Hatchery Production: Describes the artificial propagation of fish that occurs in a hatchery as opposed to propagation resulting from natural reproduction. In the North Fork Lewis River, the hatchery production program is designed to maintain harvest opportunities downstream of Merwin Dam and in project reservoirs (residents) and to provide both adult and juvenile anadromous fish for early supplementation efforts in the basin.

Hatchery Scientific Review Group (HSRG): An independent scientific review group established by the United States Congress to initiate hatchery reform that balances both conservation and harvest goals.

Infectious hematopoietic necrosis virus (IHNV): Severe viral disease in the *Novirhabdovirus* genus affecting salmonid fish, particularly smolts and younger life stages.

Infectious pancreatic necrosis virus (IPNV): Severe viral disease in the *Birnaviridae* family affecting salmonid fish, particularly smolts and younger life stages.

Jolly-Seber (JS) mark-recapture model: Provides estimates of abundance, survival, and capture rates from capture-recapture experiments. A fully open-population model (allows for births, deaths, immigration and emigration from a population) estimating both recruitment to the population and survival.

Juvenile life stages (parr, transitional, smolt, precocious male):

- Parr – Juvenile salmonid in a non-migratory stage adapted for freshwater residence. Exhibits distinct parr marks, yellow to brown body and fin coloration, or no signs of smoltification.
- Transitional – Juvenile salmonid exhibiting initial signs of smoltification (i.e., a silvery sheen with visible parr marks). Black pigment may be present on dorsal and caudal fins.
- Smolt – Juvenile salmonid that is entering a stage of seaward migration and adapted for survival in sea water. Exhibits a silvery sheen, mostly or completely absent of parr marks, deciduous scales, white or transparent abdominal fins, and black pigment on dorsal and caudal fins.
- Precocious male – Juvenile male fish that mature in their first or second year, prior to going to sea. Fully mature males have soft abdomens and milt may be expressed. Deeper body morphology compared to smolts. Dark body color. Parr marks may be visible. Dark body and abdomen coloration compared to parr and smolt life stages.
- Post smolt – A salmonid that has previously undergone smoltification and has reverted to a freshwater-adapted stage, typically due to being held in captivity in freshwater. Visual indicators of this phenotype are not well described in the literature. Some fading of silver coloration. Parr marks are not expected to re-emerge. Some yellow or brown-orange coloration of the fins. Fading of intense black pigmentation in the fins.

Kelt: A post-spawn iteroparous fish such as a steelhead or cutthroat.

Lewis River Hatchery Complex: Hatchery fish production in the Lewis River Basin originates from the Lewis River, Speelyai, and Merwin hatcheries, collectively known as the Lewis River Hatchery Complex. The three hatcheries share adult return, rearing, and release functions. A detailed description of each of these facilities is presented in Appendix A of the H&S Plan.

Lower Columbia River (LCR): for the purposes of salmon recovery, referring to the Sub-domain of the Columbia River Basin that includes the estuary and all sub-basins upstream to the towns of While Salmon, Washington and Hood River, Oregon.

Major Population Group (MPG): Group of populations, or strata, sharing similar genetic, life-history, and spatial distribution that make up a subgroup of an Endangered Species Act-listed species (e.g., Coastal MPG, Cascade MPG). Viability of all MPGs are necessary for viability of Endangered Species Act-listed species.

Merwin Fish Collection Facility (Merwin FCF): A trapping, collection, and sorting facility located at the base of Merwin Dam. The Merwin FCF processes fish for transport upstream as well as broodstock for hatchery operations.

Native (or indigenous): Fish species that have become established in the North Fork Lewis River Basin without human intervention or being substantially affected by genetic interactions through non-native stocks. Native North Fork Lewis River stocks may be present in areas outside the North Fork Lewis River Basin.

NOB: Natural Origin Broodstock

NOR: Natural Origin Recruit

NOS: Natural Origin Spawners

Natural Production: Fish that are produced in the natural environment without human intervention as opposed to artificial propagation in a hatchery.

North Fork Lewis River (Lewis River): Includes the mainstem Lewis River from its confluence with the Columbia River to its origin (RM 94.2) on the northwestern slope of Mt. Adams, including free flowing sections between hydroelectric dams. Excludes the East Fork Lewis which enters the North Fork Lewis River at RM 3.5.

North Fork Lewis River Settlement Agreement (Settlement Agreement): A binding agreement between the utilities; federal, state, and regional regulatory entities; tribal entities; and non-governmental organizations. The Settlement Agreement establishes the collective agreement of all signatories with respect to the utilities' obligations in mitigating effects of hydropower operation on fisheries, wildlife, recreation, and cultural and aesthetic resources. The Settlement Agreement forms the basis for issuing hydroelectric operating licenses by the Federal Energy Regulatory Commission for the four hydroelectric projects on the North Fork Lewis River.

Ocean Recruits: Total escapement of hatchery- and natural-origin fish including those harvested in the ocean, Columbia River, and terminal fisheries.

Proportion of Hatchery Origin Spawners (pHOS): Proportion of natural origin spawners composed of hatchery origin spawners. Equals HOS/(NOS + HOS).

Proportion of Natural Origin Brood (pNOB): Mean proportion of natural origin spawners contributing to broodstock in a hatchery program. Equals NOB/(HOB + NOB).

Passive Integrated Transponder (PIT) tags: Electronic tags inserted into the dorsal sinus or body cavity of fish that transmit data indefinitely when activated by a specialized antenna or reader. All PIT tags have a unique code allowing on-site identification of individual tagged fish.

Proportionate Natural Influence (PNI): Proportionate natural influence on a population composed of hatchery- and natural-origin fish. Equals pNOB/(pNOB + pHOS).

Regional Mark Information System (RMIS): a collection of online databases that maintain records of coded wire tag release, recoveries and locations.

Residual or Residualism: Salmonids that fail to migrate from their natal streams or stream basin after the majority of their cohort have emigrated in a given year. Depending on the species, residuals may take on several different life-histories including precocious sexual maturation, freshwater residence for a season (e.g., to overwinter) or for an additional year followed by anadromy, or in steelhead, permanent freshwater residence and spawning in multiple years. Salmonids with the potential to express anadromy are considered residuals as long as they reside in freshwater and do not become anadromous.

Returns: Adult steelhead or salmon that have spent at least 1 year at sea and have become sexually mature and have returned to the North Fork Lewis River to spawn.

Smolt Index: A number assigned to juvenile salmon that describes the stage of smolt development based on a visual assessment of skin and fin pigmentation (silvering) and body shape. 1 = parr, 2 = transitional, 3 = smolt, 4 = precocious male, 5 = post-smolt or residual (modified from Gorbman et al., 1982).

Smolt to Adult Ratio (SAR): Survival from the beginning point as a smolt (release) to an ending point as an adult.

Supplementation: The use of artificial propagation to develop, maintain, or increase natural production while maintaining the long-term fitness of the target population, and keeping the ecological and genetic impacts to non-target populations within specified biological limits. In the North Fork Lewis River, the supplementation program is designed to reintroduce spring Chinook, late winter steelhead, and early coho to habitat upstream of Merwin Dam.

Single Nucleotide Polymorphism (SNP): SNP genotyping is the measurement of genetic variations of between members of a species. A SNP is a single base pair mutation at a specific locus, usually consisting of two alleles. SNP arrays can be used to analyze large numbers of samples such as outmigrating smolts or transported steelhead upstream of Swift Dam for less cost than microsatellite genotyping.

Steelhead broodstock: Steelhead captured either through traps or in-river netting that meet predetermined genetic assignment probabilities.

Stubby dorsal fin: A dorsal fin in which the rays have become crooked especially along the leading edge and depressed as compared to naturally produced fish. Stubby dorsal fins are indicative of fish reared in a hatchery environment.

Swift Floating Surface Collector (FSC): A trap and haul facility used to collect, sort, sample, and tag outmigrating smolts from adult and juvenile supplementation programs upstream of Swift Dam. The FSC is located on the forebay of Swift Reservoir and has the ability to operate year-round and through fluctuating reservoir levels. All fish are sorted in the FSC and trucked either downstream of Merwin Dam or returned to the reservoir.

Tangle net: A net designed to entangle the snout (not the gills) of target species through use of smaller mesh sizes. This method is considered a safer alternative to traditional gill netting in which the net material may become wedged under the fish operculum, potentially causing lacerations of the gill lamellae.

Viral hemorrhagic septicemia virus (VHSV): Virus in the genus *Novirhabdovirus,* exclusive to fish and related to IHNV.

EXECUTIVE SUMMARY

The Annual Operating Plan (AOP) focuses on developing methods and protocols for monitoring and evaluating objectives and key questions described in the Hatchery and Supplementation Plan (H&S Plan, 2020).
https://www.pacificorp.com/content/dam/pcorp/documents/en/pacificorp/energy/hydro/lewis-river/license-implementation/ats/A%20-%20HS%20PLAN%20FINAL%202020.pdf

This AOP is required under Section 8.2.3 of the North Fork Lewis River Settlement Agreement (Settlement Agreement). Section 8.2.3 states that, at a minimum, the AOP must contain the following information:

1. A production section specifying the species and broodstock sources
2. Current hatchery target and juvenile production targets
3. A release section identifying, by species, the rearing schedule and planned distribution of fish and the schedules and location for release
4. A list of facility upgrades to be undertaken in the current year
5. A description of relevant monitoring and evaluation to be undertaken

Sections A, B, and C of this plan are dedicated to the hatchery production components of each of the transport species:

- Section A - Late winter steelhead
- Section B - Spring Chinook
- Section C - Coho salmon

Each section is organized and formatted similarly to maintain consistency within this document to assist in locating specific information for each species. Other sections in this plan include Monitoring and Evaluation (Section D), Adaptive Management (Section E), and Reporting Requirements (Section F).

Monitoring and evaluation activities are described at a high level of detail in Strategies which are attached to the AOP and referenced in Section D of the AOP. Strategies included in the 2023 AOP include:

- Strategy A: Adult Abundance and Composition
- Strategy B: Adult Spatial and Temporal Distribution
- Strategy C: Juvenile Abundance and Migration
- Strategy D: Fish Health Monitoring and Disease Prevention
- Strategy E: Spring Chinook Rearing and Release Evaluation
- Strategy F: Precocity and Morphology Sampling
- Strategy G: Genetic Risk Monitoring (DRAFT; REVISIONS EXPECTED IN 2023)
- Strategy H: Volitional Release

- Strategy I: Smolt-to-Adult Return Rate Estimation (IN PROGRESS)
- Strategy J: Sampling and Data Collection Checklist

AREA OF FOCUS

Generally, the AOP is focused on monitoring hatchery production operations and assessing the risks of these operations on naturally occurring salmonid populations present in the North Fork Lewis River downstream of Merwin Dam (RM 19.5). For purposes of this plan, the North Fork Lewis River is defined as the mainstem Lewis River between Merwin Dam and the confluence of the East Fork Lewis River (RM 3.5).

Hatchery fish production in the Lewis River Basin originates from three separate hatcheries: Lewis River, Speelyai, and Merwin. These facilities are collectively referred to as the Lewis River Hatchery Complex. The facilities share and coordinate hatchery functions such as adult holding, spawning, juvenile rearing and release. A description of each of these facilities and the general hatchery production program is provided on the PacifiCorp website.

https://www.pacificorp.com/content/dam/pcorp/documents/en/pacificorp/energy/hydro/lewis-river/relicensing-documents/AQU_8_Report.pdf

SUMMARY OF SIGNIFICANT CHANGES

A summary of major changes between the 2022 AOP and the 2023 AOP is included in Table ES1.

In 2022, the monitoring and evaluation section of this AOP (Section D) was completely updated to be consistent with revised monitoring objectives of the 2020 H&S Plan. Objectives provided in Section D represent the minimum monitoring benchmarks necessary to meet the requirements of the Settlement Agreement as well as recommendations from the Hatchery and Scientific Review Group (HSRG).

Also in 2022, based on recommendations from the ATS, the Adaptive Management section (Section E) of this plan was also significantly revised to address the new decision-making framework described in the most recent version of the H&S plan.

Strategy K, a pilot study for monitoring total dissolved gas saturation, was fully implemented in 2022 and therefore has been removed from the 2023 AOP. The results of the pilot study will be reported in the 2022 Annual Operating Report.

No other major changes to the framework or activities described were made between the 2022 AOP and 2023 AOP.

Table ES - 1. Summary of Significant Changes to AOP, 2022 to 2023

Section(s)	Change	Rationale
Sections A, B, C	No major changes.	Significant revisions were made to the AOP throughout 2022, incorporated into the 2022 AOP and rolled forward into the 2023 AOP.
Strategies	Removal of Strategy K	Fully implemented and analysed in 2022.
Appendix A	Updates to ATS Work Plan for 2023	The ATS uses a spreadsheet-based calendar to review and prioritize action items and track upcoming discussions and actions; for reference, it is included in the AOP as a working file.

SECTION A LATE WINTER STEELHEAD

1.0 INTRODUCTION

The Lewis River late-winter steelhead integrated-hatchery program (hereafter the “integrated hatchery steelhead program”) has three main components, described in Sections 2, 3, and 4 below: Section 2) broodstock collection and processing for adult program implementation; Section 3) juvenile rearing and release; and Section 4) adult supplementation upstream of Swift Dam. The following sections describe the protocols for implementing the Lewis River late winter steelhead portion of the Hatchery and Supplementation Plan (H&S Plan; PacifiCorp and Cowlitz County PUD, 2020).

2.0 ADULT PROGRAM IMPLEMENTATION

Broodstock collection for the integrated steelhead program is based on three factors: the relative abundance of natural- and hatchery-origin returning adults (Section 2.1), the total collection goal (Section 2.2), and adult return timing (Section 2.3). Adult steelhead are primarily collected at Merwin FCF (Section 2.4) and sampled following a standardized protocol (Section 2.5). Individual adults are sorted and transported either to Merwin Hatchery for broodstock or to the upper watershed to be released above Swift Reservoir (Section 2.6). Adults collected for broodstock are held and monitored (Section 2.7). As the broodstock reach maturation, individuals are live spawned following standardized protocols (Section 2.8). Upon completion of spawning, spawners are returned to river and genetic analysis is completed on all broodstock to evaluate their biological population of origin (Section 2.9).

2.1 Broodstock Collection Strategy

Since the inception of the integrated steelhead program in return year 2009 thru 2021, the broodstock collection strategy has had two main components. First, broodstock was collected using a “100% pNOB” strategy, which meant the broodstock could only be derived from natural-origin adults (i.e., pNOB = 100%). Second, broodstock had to be collected from adults that returned to the NF Lewis River Basin (i.e., no out-of-basin transfers) and preference was given to in-basin recruits that were determined via genetic screening. Subsequently, no hatchery-origin adults were permitted to be used for broodstock.

The primary justification for the original “100% pNOB” strategy was because there was no other existing within-ESU winter steelhead hatchery program to use as a source for broodstock for the NF Lewis program. Over the next decade, the 100% pNOB strategy remained in place even as BWT’s from the Lewis River integrated steelhead program began to return in relatively high numbers. This decision to keep the 100% pNOB strategy was based on the general goal of trying to minimize genetic risks associated with the hatchery program on the natural-origin population (e.g., loss of diversity, domestication). However, the performance of the 100% pNOB collection strategy was never assessed relative to alternative approaches to achieve the goal(s) of the supplementation program.

During the re-write of the Hatchery & Supplementation (H&S) Plan in 2020, the ATS identified the need to develop "transition plans" for all Lewis River hatchery programs. These proposed transitions plans will formally outline the goals of each hatchery program during each recovery phase, identify hatchery performance metrics and priorities associated with each recovery phase, outline current and alternative hatchery program operations, and provide guidance for future hatchery operations. The ATS has not begun developing transition plans for any program. However, over the past year, there have been ongoing discussions within the Lewis ATS to re-assess the integrated steelhead program and potentially update aspects of the current implementation strategy before the completion of the transition plans. These recent discussions were largely initiated by issues with the steelhead broodstock collection in 2021 and in-season discussions that transpired. In short, the ATS began debating whether the existing broodstock collection should be updated to allow for use of hatchery-origin adults.

In January 2022, the ATS met and discussed potential updates to the broodstock collection strategy for the integrated steelhead program. In summary, the ATS decided to change the broodstock collection strategy from 100% pNOB to a "mining rate" strategy that also ensures demographic replacement. Specifically, the mining rate strategy specifies the maximum proportion of natural-origin adults that can be removed for broodstock. However, NORs can only be collected for broodstock once an equivalent number of BWTs have been transported to the upper basin. Unlike the 100% pNOB strategy, which prioritizes returning NOR adults for broodstock, the mining strategy prioritizes NORs spawning in the wild while also allowing for a fixed proportion to be used for broodstock so long as it does not result in an overall net loss. The decision by the ATS to change the broodstock collection strategy was based on several factors, including recent guidance from HSRG (2020), the group discussing hatchery performance metrics and prioritization, and an evaluation of various broodstock collection strategies by WDFW that demonstrated the mining rate strategy outperforms the 100% pNOB strategy for priority metrics during the recolonization phase of recovery (Bentley and Buehrens, *unpublished*).

Beginning in return year 2021-22, a 30% fixed mining rate will be used to collect broodstock for the integrated steelhead program. Here, for every 10 NOR adults that return to the NF Lewis River and are collected at the Merwin FCF or Lewis Hatchery ladder, the first 7 will be transported above Swift Reservoir to spawn in the upper NF Lewis River basin and the next 3 can be collected for broodstock based on collection needs. However, for NOR adults to be collected for broodstock, each individual NOR must be demographically replaced by two integrated steelhead program adults (i.e., BWTs) that are transported to the upper basin. This 2:1 ratio of BWTs transported to NORs collected for broodstock assumes BWTs have a relative reproductive success of 50 percent (e.g., Araki et al. 2007). The number of adults collected for broodstock is based on the overall collection goal (Section 2.3) and the pre-determined collection schedule (Section 2.4). Any broodstock collection needs that cannot be met with NOR adults will be supplemented with BWTs.

2.2 Broodstock Collection Goal

The current goal is to spawn 25 adult females and 25-35 adult males for a total of 50-65 adults. The total collection and spawn goal of 25 females is primarily based on the total egg take target of 90,000 +/- 20%, which is needed to meet the total smolt release target of 50,000 smolts +/- 20% based on the average fecundity of females and the anticipated in-hatchery survival from egg-take to smolt release. The 25-female target is also based on trying to minimize adverse genetic impacts. In short, hatchery supplementation programs can affect the effective population size of the natural-origin population due to alteration of reproductive success for a subset of a population and subsequent contribution rate (stocking proportion) of the artificially propagated parents (Ryman and Laikre, 1991). The smaller the supplementation program, the larger the risks of reducing genetic diversity and the relationship is likely non-linear (Ryman and Laikre 1991). Therefore, depending on the observed fecundity of collected broodstock, females may be partially spawned to assist in meeting the 25-female target without exceeding the egg take goal. However, this decision must be coordinated with WDFW and NMFS before implementation.

In previous years, the total broodstock collection goal was larger than the spawn goal based on expected in-season genetic assignment rates being <1 (i.e., some adults collected for brood would not be used based on their genetic assignment). Now that in-season genetic screening has been discontinued (see Section 2.9), the collection and spawn goal are the same.

2.3 Broodstock Collection Timing

Broodstock collection should occur proportional to the run timing of natural-origin Lewis River winter steelhead (Figure A-1). In January 2022, the ATS assessed the existing brood collection schedule relative to the return timing of NORs at Merwin FCF from (2015-2021) and found that it only captured ~65-70% of the entire NOR run-timing. Therefore, the ATS agreed that the broodstock collection schedule should be updated to match the return timing of NORs and start in mid-December, with one small exception in that collection should end by May 31st (as opposed to the third week of June). This modification results in a 7% reduction in average run-timing relative to NORs which may reduce some genetic diversity but balances the risks associated with spawning late-arriving fish. Specifically, progeny from broodstock spawned after May 31st may be difficult to rear to appropriate release size by the scheduled release time in May of the following year. Risks of releasing under-sized fish include decreased survival and a potential increase in residualism (Hausch and Melnychuk 2012), which may increase ecological interaction with other Endangered Species Act (ESA) listed salmonid populations (i.e., competition and predation).

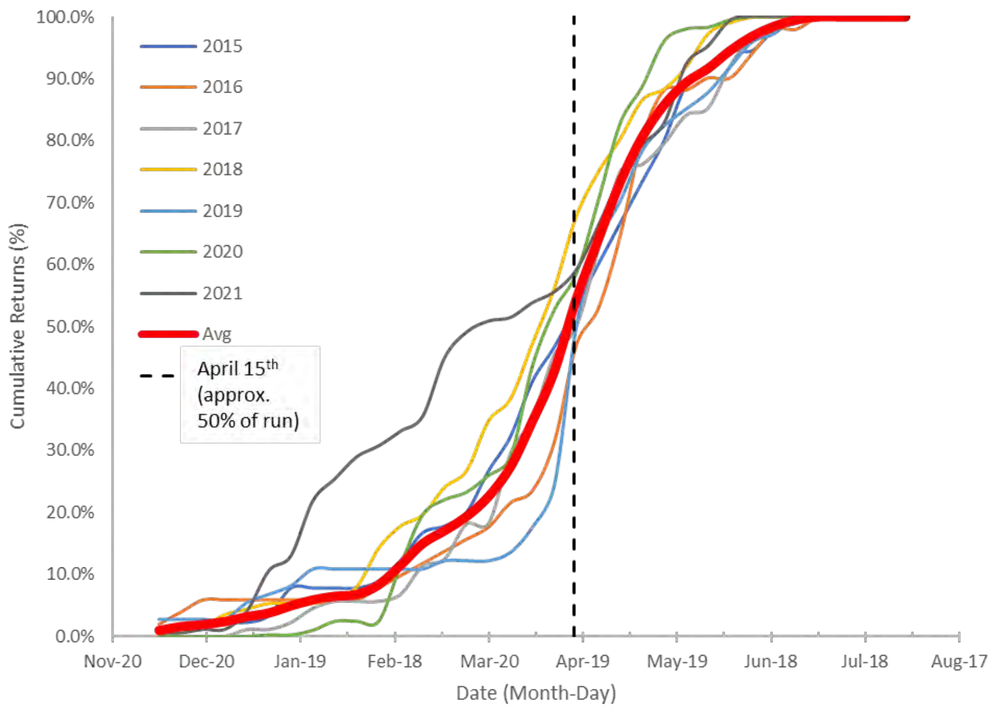

Figure A - 1. Percent cumulative returns of adult NOR winter steelhead at Merwin FCF by year (2015-2021) and the non-weighted average (solid red).

Broodstock collection schedules may be amended in-season relative to the average timing of the first NOR returns (mid-December) in response to run-timing assessments. The origin of the broodstock (NOR, BWT) will be derived by the criteria outlined in Section 2.2. Any broodstock collection targets that are not met for a given week will be added to the following week's targets. The general broodstock collection schedule for winter steelhead is detailed in Figure A-1 and Table A-1.

Table A - 1. Broodstock collection goals for winter steelhead.

Appx. Date (Start of Week)	Females (Weekly)	Males (Weekly)	Total (Weekly)	Females (Cumulative)	Males (Cumulative)	Total (Cumulative)	% Total (Cumulative)
18-Dec	-	1	1	-	1	1	2%
25-Dec	1	-	1	1	1	2	4%
1-Jan	-	-	-	1	1	2	4%
8-Jan	-	1	1	1	2	3	5%
15-Jan	1	-	1	2	2	4	7%
22-Jan	-	-	-	2	2	4	7%
29-Jan	-	1	1	2	3	5	9%
5-Feb	-	-	-	2	3	5	9%
12-Feb	1	-	1	3	3	6	11%
19-Feb	1	1	2	4	4	8	15%

26-Feb	-	1	1	4	5	9	16%
5-Mar	1	1	2	5	6	11	20%
12-Mar	1	1	2	6	7	13	24%
19-Mar	1	1	2	7	8	15	27%
26-Mar	1	1	2	8	9	17	31%
2-Apr	1	2	3	9	11	20	36%
9-Apr	2	2	4	11	13	24	44%
16-Apr	3	4	7	14	17	31	56%
23-Apr	3	3	6	17	20	37	67%
30-Apr	3	3	6	20	23	43	78%
7-May	2	2	4	22	25	47	85%
14-May	1	2	3	23	27	50	91%
21-May	1	2	3	24	29	53	96%
28-May	1	1	2	25	30	55	100%

2.4 Adult Collection Methods

To initiate the late winter steelhead supplementation program, natural-origin winter steelhead were collected from spawning grounds in the lower Lewis River Basin and used as broodstock. For approximately the first decade of the program, adults continued to be primarily collected in the lower mainstem Lewis River via tangle netting, and sometimes hook-and-line, on the spawning grounds. Over the years, more natural-origin adults have returned to the Merwin FCF and preliminary data suggest that a high proportion of the adults collected at Merwin FCF originated in the upper basin. Since return year 2019, broodstock has been exclusively collected from Merwin FCF. The Lewis River Hatchery ladder collects a few NOR and BWT steelhead each year (Kevin Young, *WDFW*, personal communication) but these adults have not been used for broodstock and are transported to the upper basin.

2.5 Adult Disposition

Adults captured at the Merwin FCF are distributed based on presence of marks, PIT tags and subject to the broodstock collection strategy outlined in Section 2.1. The disposition of adults is illustrated in Figure A-4.

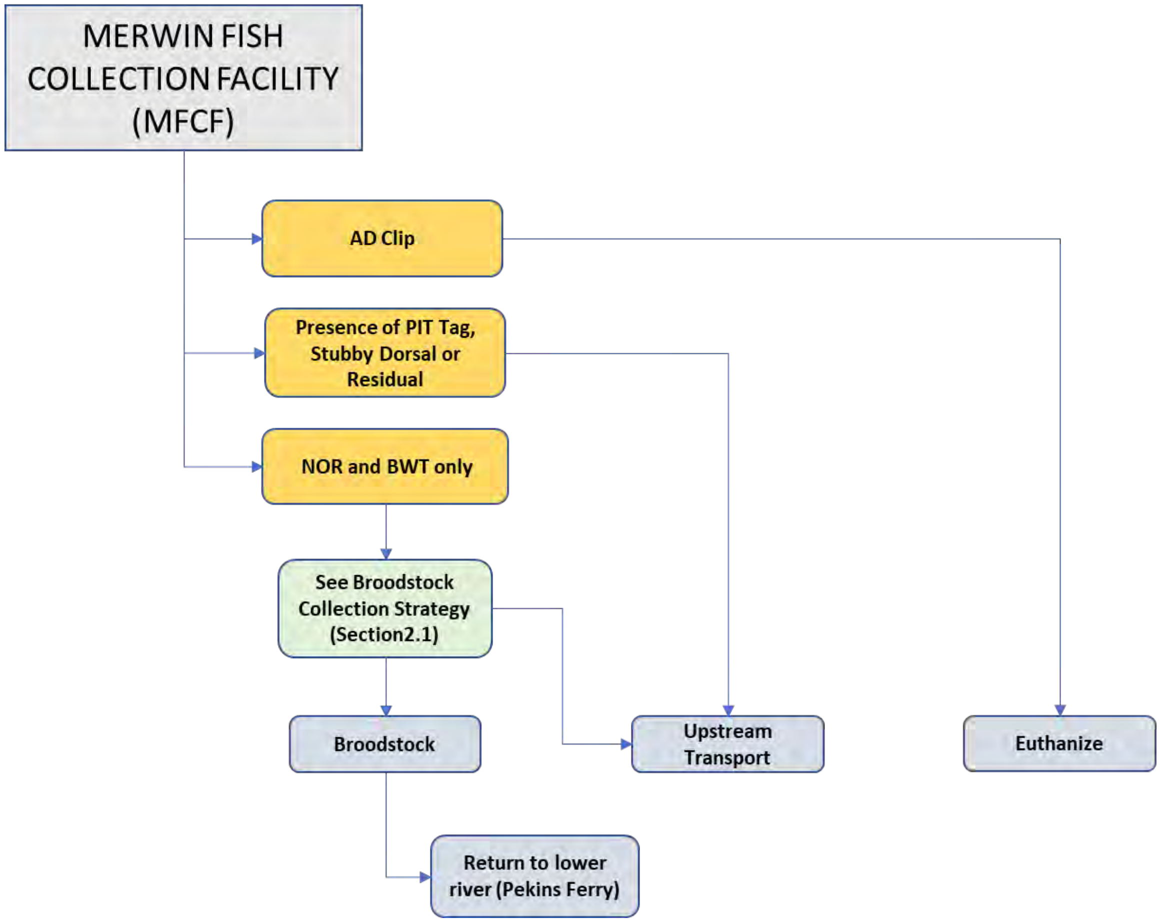

Figure A - 2. Distribution of adult late winter steelhead captured at the Merwin FCF

2.6 Data Collection Protocols

The majority of returning winter steelhead adults are collected and subsequently sampled at the Merwin FCF. The specific data and the group responsible for collection (WDFW, PacifiCorp) will depend on the transport location of each adult (hatchery broodstock or upstream transport) and the total number of adults handled in a given week (i.e., a subset of data are collected based on a sample rate).

For adults that will be transported upstream, staff at Merwin FCF will be responsible for collecting all necessary data fields prior to transport. Every data field will be recorded for each collected adult with the exception of three categories (fork length, scale sample, and tissue sample) that will be sub-sampled on a weekly basis.

For fish designated as broodstock, staff at the Merwin hatchery will be responsible for collecting all necessary data fields with the exception of PIT tagging (Table A-2). All data collected shall be provided to PacifiCorp for annual reporting purposes.

Any adult that does not possess a PIT tag will receive one from the Merwin FCF crew. All PIT tags (new and recaptures) will be uploaded to PTAGIS daily. All other sampling of broodstock will occur during the processing of brood at the hatchery by WDFW staff.

Below is a summary of the data types collected and a brief description of the collection procedure and sample rates (where applicable).

1. **Date of capture** (mm-dd-yyyy)
2. **Capture location** (Merwin FCF, Lewis River Hatchery, Lower Lewis River)
3. **Capture method** (trap/ladder, hook-and-line)
4. **Disposition location** (upstream transport, broodstock, return to river (downstream of Merwin Dam)
5. **Sex** (Male/Female)

   The sex of a fish can be determined by assessing its relative size, shape and secondary characteristics such as maxillary length relative to eye position.

6. **Mark status** (e.g., AD, UM, BWT unknown)

   The mark status of a fish describes any external fin clip(s) or wire tags an individual fish has received. In general, the mark status is used to determine the origin of fish captured. Fish with AD clips (AD) or wire tags in their snout (BWT) are considered as hatchery origin while unmarked fish (UM) lacking a stubby dorsal fin are considered as natural origin.

   Each fish returning to the Merwin FCF will be automatically scanned for the presence of a BWT (mark status = BWT).

   A small proportion of hatchery fish can be "misclipped" where the intention was to remove the adipose fin but was either unsuccessful or only a partial clip of the adipose fin occurred. Partial clips of the adipose fin will be recorded as AD, and any fish possessing a BWT with adipose removed or partially clipped will be designated as "BWT -unknown".

7. **PIT tag status** (Positive or Negative)

   Each fish returning the Merwin FCF is automatically scanned for PIT tag regardless of their mark status. All fish possessing a PIT tag are sampled for length by the Merwin FCF crew and capture data are automatically uploaded to PTAGIS. Steelhead that do not possess a PIT tag, will be tagged by the Merwin FCF crew in the dorsal sinus regardless of where it is transported.

8. **Dorsal fin status** (Positive or Negative)

   A small number of steelhead captured display a stubby dorsal fin (Figure A-5), with no adipose clip or BWT. Based on several years of late winter steelhead genetic analysis, these fish originated from the integrated hatchery supplementation program that either lost their BWT or did not receive a BWT.

To avoid the use of segregated early winter steelhead in the broodstock, all unmarked stubby dorsal positive fish will be transported upstream. Adipose marked fish will be treated as segregated early winter steelhead and transported Merwin hatchery to be euthanized.

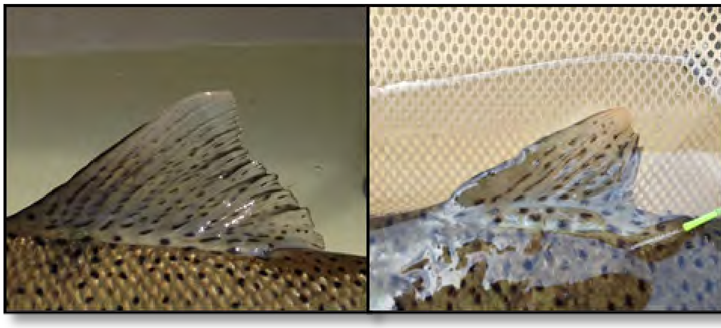

Figure A - 3. Example illustrating the shape of a normal (left) and stubby (right) dorsal fin.

9. **Life History (residual or anadromous)**

   Residuals are identified by color, body shape, and size (Figure A-6). Residuals exhibit deep (and often vibrant) coloration and spotting as opposed to anadromous fish that exhibit a silvery sheen and subdued spotting. Residuals also possess a distinct red or pink lateral stripe. Body shape of residuals is more rotund and are always smaller in size than their anadromous cohorts - typically less than 500 mm in length. Residuals entering the Merwin FCF shall be transported upstream and not selected for broodstock.

Figure A - 4. Residual steelhead encountered during annual tangle netting (note stubby dorsal fin)

10. **Fork length**

A fork length measurement should be taken from the first 10 fish captured at the Merwin FCF per week by origin (i.e., up to 10 NOR and 10 BWT per week). Measure the fork length (FL) of each fish in millimeters from the tip of the jaw or tip of the snout, whichever is greater, to the center of the fork in the tail. All broodstock transported to Merwin hatchery will be measured for length by Merwin hatchery staff

11. **Scale sampled** (Yes/No)

Scales should be collected from the first 10 fish captured per week by origin (i.e., a maximum of 20 scales per week) at the Merwin FCF. Scale samples are not required from stubby dorsal fish as their origin cannot be verified or from PIT tag positive fish as age determination is obtained from the initial PIT tagging event from the Swift FSC as smolts. All broodstock transported to Merwin hatchery will be scale sampled by Merwin hatchery staff.

Scales should be removed and placed on a scale card including date and location of capture and fork length. A new scale card should be used for each capture date. Collect three scales from each fish just above the lateral line and below the posterior insertion of the dorsal fin

12. Tissue Sample

Tissue samples shall be collected from all broodstock transported to Merwin hatchery by Merwin hatchery staff. Meta data should be included for each sample collected.

Tissue samples should be collected from the upper lobe of the caudal fin. Collect as much tissue as possible up to approximately 1/4” X 1/4” in size (hole-punch sized). Samples should be stored at room temperature on either a sheet of blotter (i.e., chromatography) paper or individual vials containing 95% natural ethanol (preservative).

13. Data Collection Responsibility

The sampling of returning late winter steelhead is a shared responsibility between the Merwin FCF and hatchery crews. Generally, sampling of fish transported upstream will be conducted by Merwin FCF crews and sampling of broodstock will be conducted by hatchery crews. As part of initial capture at the Merwin FCF, all steelhead lacking a PIT tag upon capture will be PIT tagged at the Merwin FCF including those transported to Merwin hatchery as broodstock.

Table A - 2. Data collection for late winter steelhead.

Data Type	HOR	NOR
Genetic Tissues	All broodstock sampled	
Fork Length	Merwin Trap (PIT -): 10 BWT per week (PIT +): Sample All (stubby dorsal only): no sample Merwin Hatchery All broodstock sampled	Merwin Trap (PIT -): 10 per week (PIT +): Sample All Merwin Hatchery All broodstock sampled
Scales	Merwin Trap (PIT -) 10 BWT per week (PIT +) none Merwin Hatchery All broodstock sampled	Merwin Trap (PIT -) 10 BWT per week (PIT +) none Merwin Hatchery All broodstock sampled
Fecundity	Individual estimate per female	Individual estimate per female
Note: all AD clipped steelhead are transported to Merwin hatchery to be euthanized		

2.7 Broodstock Holding Protocols

All winter steelhead assigned as potential broodstock will be held at the Merwin Hatchery. Upon arrival, each fish will be Floy tagged and placed into adult holding pond(s). Hatchery staff will check broodstock as needed for maturity starting in the last week of March.

If a female becomes ripe to spawn and no male broodstock are available, the female will be returned to the river; however, all possible precautions will be made to prevent this situation from occurring. The Hatchery and Supplementation steelhead program coordinator in consultation with hatchery management staff will make decisions regarding the release of fish. Collection goals should be reviewed to evaluate the risk to project goals of releasing the fish (i.e., will more females likely be available through future collections).

The following list represents recommendations from WDFW hatchery staff to reduce handling related stress, injury, or mortality of steelhead held at the Merwin Hatchery.

1. The use of only rubberized nets to hold or move steelhead: Rubberized nets are known to reduce descaling and abrasion.
2. Eliminating the use of cotton gloves to handle steelhead in favor of bare hands: Cotton gloves are abrasive on fish and remove the protective mucous on the skin of fish.
3. Aqui-S is used for the safety of the employee and to prevent injury and stress to fish during air spawning from the females and live spawning of the males.
4. Floy tags of several colors are used for quick visual identification of individual fish to limit the number of fish handled when checking for ripeness.

5. Salt and Formalin are used in holding raceways or circular tanks for steelhead. Salt reduces stress and improves oxygen uptake. Formalin is used to control fungi and parasites.

2.8 Genetic Assignment and Analysis

Since the inception of the hatchery steelhead supplementation program, genetic stock identification (GSI) has been used in-season to genetically categorize returning adults and help inform broodstock collection. Originally, the GSI work was completed by National Marine Fisheries Service (NMFS) under a contract with PacifiCorp. NMFS used a microsatellite (13 mSAT loci) based on a reference baseline almost exclusively composed of Lower Columbia steelhead used in Blankenship et al. (2011). In 2020, NMFS declined to renew the contract due to staffing shortages. In 2020 and 2021, WDFW's Molecular Genetics Laboratory (MGL) was awarded the contract to perform this in-season analysis using a single nucleotide polymorphism (SNP) based baseline that was specifically assembled for Lower Columbia steelhead populations and segregated hatchery programs (HW354). For more information on the WDFW MGL assembled Lower Columbia steelhead baseline, see memo written by Todd Seamons to the ATS on April 20th, 2020 (Subject: Assessing the performance of the WDFW Lower Columbia River steelhead SNP reference baseline for genetic stock identification (GSI)).

In January 2022, the ATS agreed to discontinue in-season genetic screening moving forward. This decision was based on three factors (see below). However, the ATS agreed that a post-season analysis was still warranted to continue monitoring the genetic composition of the broodstock and inform future monitoring decisions:

(1) **Past genetic screening results -** In-season results from 2020 & 2021 estimated that 95-100% of fish collected for broodstock were from Cascade MPG, which meant no adults were excluded based on their genetics results and the outlined broodstock collection criteria. Overall, these results suggest that the risk of collecting an out-of-MPG adult is low.

(2) **Performance of SNP-based baseline** – Based on a power analysis of the current SNP-based steelhead baseline (HW354), SNP's analysis has an accuracy rate of about 90 percent in assigning Lewis River origin adults to the Cascade MPG and greater than 99 percnet accuracy in assigning segregated hatchery fish (i.e., Chambers winters and Skamania summers). However, the baseline is only about 50 percent accurate in assigning Lewis River origin adults to Lewis River versus other Cascade MPG populations. Further details can be found in Seamons (2020).

(3) **Relative benefit of in-season screening** – Based on recent genetic results and the relative performance of the SNP-based baseline, the biggest benefit of continuing the in-season genetic screening is that it can accurately help exclude Chambers early winter steelhead from the broodstock. However, genetic screening adds little benefit given that there is little overlap between Chambers and NORs/BWTs, >99% of Chambers can be identified via AD clip (assuming an average mis-clip rate of 1%), and the remaining 1% can likely be identified via their "stubby dorsal" and excluded by

invoking the rule that no adipose intact fish with a stubby dorsal can be retained as broodstock.

2.9 Spawning Protocols

The total collection and spawn goal of 25-35 males is based on the spawning strategy (see Section 3.8). Briefly, a randomized factorial spawning strategy is implemented for hatchery winter steelhead, which is simply the process of spawning individual fish with more than one mate. There are numerous ways to hypothetically implement factorial spawning, but the specific factorial cross is largely dependent on the number of ripe spawners for a given day. In general, the most common cross is 2x2. However, due to the small program size and variable spawn timing, there can be instances when there is a single ripe female on a given day. When this occurs, two males are spawned with one female. The possibility of this occurring necessitates the need for additional males to be collected.

All collected fully mature broodstock will be spawned according to the following protocols, without regard to age, size, or other physical characteristics:

1. No fish shall be excluded except for those with overt disease symptoms or physical injuries that may compromise gamete fertility or viability.
2. All spawned fish will be returned to the lower river.
3. Females will be air spawned.
4. Fully randomized factorial mating protocols are preferred to avoid or reduce selection biases and maintain diversity.
5. If pairwise mating is warranted (e.g., only one ripe female is available) the use of a backup male is preferred to reduce the potential for egg loss from infertile males.
6. If two females and only one male are ripe, a 2x1 cross can occur. However, this is not preferred and efforts should be made to collect additional males from the Merwin FCF if in Phase II collection.
7. Holding males for additional spawning crosses is not permissible.
8. If a ripe spawning female has no mate, that fish will be returned to the river downstream of Merwin Dam in hopes of spawning naturally (see Section 2.10 Release Protocols). All precautions will be taken to prevent this situation from occurring. Whenever possible the decision to release females should occur before the female becomes ripe.
9. During spawning, a fish health specialist will take the necessary viral samples according to standard protocols (see Strategy C).
10. Ovarian fluid will not be drained before fertilization.
11. Eliminate green egg samples: Total egg mass weight will be used to estimate fecundity.

3.0 JUVENILE PROGRAM IMPLEMENTATION

3.1 Egg Take Goals

The egg take goal for this program is 90,000 +/- 20%. This egg take target is based on the smolt release target paired with the average fecundity of females and the anticipated in-hatchery survival. Fecundity is highly variable among native females and this goal is intended to be flexible; however, total egg take should never exceed the maximum level of 108,000. In-season adaptive management will be used to meet egg collection goals through broodstock management.

3.2 Egg Incubation

Eggs from each female are placed into an individual tray to incubate. To reduce the risk of Bacterial Coldwater Disease (BCWD), each egg tray is partitioned in half, which subsequently increases egg density and reduces the flow and mobility of the eggs. Each spawned fish will have its own ozonated water supply. Eggs or fish will not be combined until viral results are known.

3.3 Rearing and Release Schedule

Fish will be transferred to the intermediate raceways located within the incubation building after incubation and hatching. Fish will remain in the intermediate raceways for a period of 6 to 8 months. All source water passes through the hatchery ozone plant for sterilization before entering the incubation building. After 6 to 8 months, or once fish outgrow the intermediate raceways, fish are transferred to outside raceways and ponds where they are subject to untreated water. Table A-4 presents a timeline for the movement of fish by life stage at the Merwin Hatchery.

In addition to monitoring rearing densities and feeding, hatchery staff assesses performance and growth by implementing sampling methods to calculate condition factor (K), estimated variation in length (CV), and feed conversion factor (FCR) for each raceway. For this stock, these assessments are typically completed prior to first feeding (July) and prior to release (May). This is done by sampling 100 fish from each raceway from three locations (upper, middle and lower) for a total of 300 fish per raceway.

Table A - 3. Generalized hatchery production and collection timeline for late winter steelhead

	Jan	Feb	Mar	Apr	May	Jun	Jul	Aug	Sep	Oct	Nov	Dec
Adult Collection												
Spawning												
Incubation												
Ponding												
Tagging (BWT)												
Volitional Release												

3.4 Feeding Type and Requirements

All fish that are ponded for rearing at Merwin Hatchery will be fed the best quality feed available through WDFW vendors. These formulations provide high protein and fat percentages and have proven to provide optimal growth from start to finish (Roberts, 2013).

There is a combination of feeding methods used. Hand feeding is typically done for early rearing troughs and raceways. Hand feeding occurs 2 to 8 times per day depending on life stage. Once fish are transferred to the large rearing ponds, demand feeders are used along with hand feedings 2 to 3 times per week if needed. The incorporation of belt feeders, or other options (underwater feeders) may be employed to provide for extended feeding schedules, or provide more natural methods for fish feeding.

3.5 Marking and Tagging

Once these fish reach a size of 20 to 25 fish per pound (fpp) in December, all are tagged with BWTs in their snout and placed into the rearing ponds until their scheduled release the following May. No other marks or clips are used for the late winter steelhead supplementation program.

3.6 Release Size and Number

Late Winter Steelhead: 50,000 smolts (± 20%) at 5 to 8 fish per pound.

Volumetric methods and individual length measurements are used to estimate the number and size of fish released (see Strategy F). If the rearing goal is exceeded, surplus fish will continue to be reared and released with the program fish. However, this will require notification to NMFS prior to release. The intention of the spawning program is to not exceed the egg take or release targets and precautions should be employed (e.g., partial spawning) if targets are in jeopardy of exceeding the production limits set in this plan.

3.7 Release Timing and Locations

Steelhead smolts are volitionally released over a six-week period, which is scheduled to begin by May 1 of each year.

Fish that actively migrate during the volitional release window are transported to the Merwin boat ramp (RM 19) for planting. Once the volitional window has ended, any remaining fish are transported and planted at Pekins Ferry Boat Ramp (RM 3.1). Alternate lower river release locations may be used if significant bird or pinniped predation is observed at the Pekins Ferry site (e.g., Woodland release ponds, county bridge, island boat ramp, etc.)

4.0 ADULT SUPPLEMENTATION UPSTREAM OF SWIFT DAM

The current transport goal of late winter steelhead is 1,700 adults (H&S Plan 2020). Transport of adult steelhead to the upper Lewis River Basin began in 2012 and has relied primarily on the production of hatchery-origin recruits (BWT) to provide a demographic boost. Over the past decade, there has also been an increase in the abundance of returning adults that are offspring from supplementation efforts upstream of Swift Dam. Because only about 10 percent of steelhead smolts are PIT tagged at the Swift FSC (and thus confirmed as upstream recruits), there remains an unknown portion of natural origin returns that cannot be classified as originating upstream of Swift Dam or downstream of Merwin Dam. However, based on the portion of PIT tagged adults returning from a known number of marks, inferences can be made on the estimated number of returning steelhead that originated upstream of Swift Dam. Therefore, steelhead transported upstream of Swift Dam will include all BWT returns and a portion of NOR returns to the trap. The portion of NOR returns transported upstream will be predicted by the ATS using PIT tag return rates and other factors deemed appropriate by the ATS (e.g., total number of smolts released from the FSC and hatchery production program by year). These estimates (when available) will be reported as part of the Aquatic M&E plan objectives.

Steelhead that are transported above Swift Dam are typically released at the Eagle Cliff Bridge Site. If the Eagle Cliff Bridge site is unavailable or inaccessible, steelhead may also be released at the Swift Camp boat ramp or Swift Dam. In some instances, fish may be released at alternate locations to enhance their distribution into tributaries of upper North Fork Lewis River. These alternate release locations include but are not limited to: Muddy River Bridge, Clear Creek Bridge, and Curly Creek Bridge. If alternate distribution sites are selected, planting trucks will work on a rotating basis for each haul. For example, the first load may be released at Curly Creek Bridge, the second at Muddy River Bridge, and so on. This may not equate to equal portions for each site but should be reasonably close.

SECTION B SPRING CHINOOK

1.0 INTRODUCTION

The Lewis River spring Chinook salmon program is composed of two parts: adult supplementation upstream of Swift Dam and juvenile hatchery production for release downstream of Merwin Dam. Adult supplementation will provide up to 3,000 adults for release upstream of Swift Dam each year to spawn naturally. Juvenile supplementation will rear up to 1,350,000 spring Chinook for release downstream of Merwin Dam.[1] Release timing of juvenile supplementation fish will vary depending on planned evaluations described in Strategy E. Returns from both the adult and juvenile supplementation programs comprise the foundation to meet the primary goals of providing harvest opportunity and creating a self-sustaining population that does not rely on hatchery support (see Settlement Agreement Section 8.4). This section describes the implementation of both the supplementation and hatchery production programs.

2.0 ADULT PROGRAM IMPLEMENTATION

The following sections describe the detailed protocols for implementation of the spring Chinook portion of the H&S Plan.

Prioritized goals for distribution of returning hatchery origin spring Chinook:

1. hatchery broodstock goal
2. Upstream transport goal
3. A fishery managed to allow for #1 and #2 to be achieved.
4. Out-of-basin programs (e.g., other Southern Resident Killer Whale programs, Deep River Net Pen project)

2.1 Broodstock Source and Selection

The Lewis River spring Chinook hatchery program is operated as a segregated program. Therefore, all broodstock transported to hatcheries will be of hatchery origin. Adult returns identified as NOR will be transported to the Lewis River above Swift Dam to help meet the transport target of 3,000 fish. No NOR Chinook will be used to meet juvenile production needs at the hatchery. Adult HOR (adipose fin missing, or adipose fin intact AND CWT snout tag) spring Chinook returns will be used to meet juvenile production (mitigation) targets. Broodstock will be selected over the course of the run, and any surplus spring Chinook will be transported upstream to achieve adult supplementation targets. In years when hatchery returns are weak, it may be necessary to hold surplus Chinook at Lewis River Hatchery in the early portion of the run until it becomes clear the annual broodstock goal will be met. After 50% of the run has been realized, a decision will be made on whether to transport all (or a

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[1] Beginning in 2018, the spring Chinook upper river acclimation program (up to 100,000 juveniles) was suspended by the ACC in favor of releasing these juveniles downstream of Merwin Dam for a period of at least 5 years. This decision was made in an effort to improve adult returns. Annual review of this modification will occur annually between the ACC and the ATS.

portion of) surplus Chinook being held at Lewis River Hatchery upstream of Swift Reservoir. Planning should be coordinated with hatchery staff to ensure that broodstock are collected proportionately over the run curve.

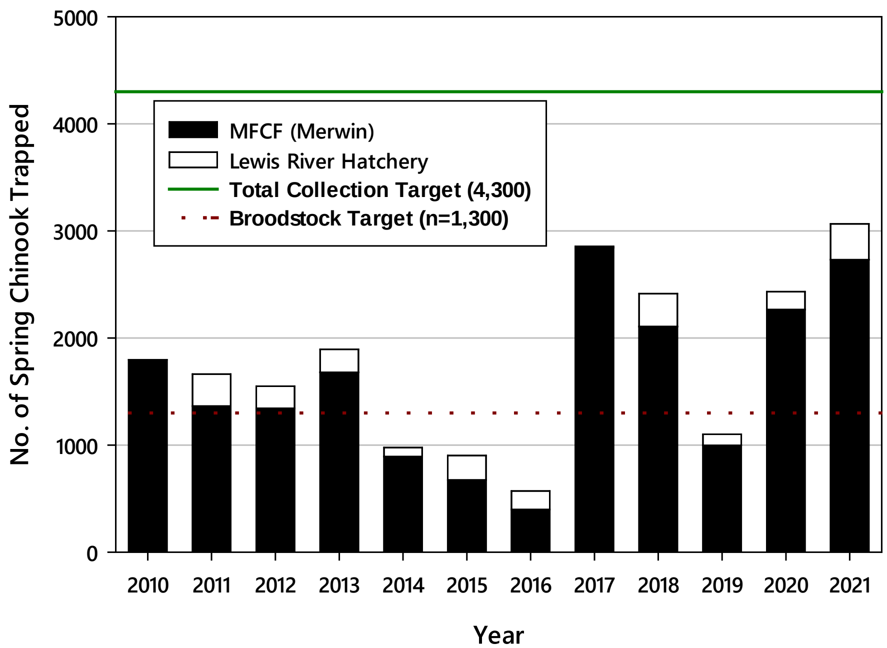


Figure B - 1. Actual number of spring Chinook trapped annually between 2010 and 2021 at both the Merwin FCF and Lewis hatchery ladder.
*Note: *Total collection target includes broodstock (1,300) and adult supplementation target (3,000) upstream of Swift Dam.*

2.2 Broodstock Collection Goal

Spring Chinook broodstock collection goals for the Lewis River programs are as follows:

- Hatchery Broodstock: Approximately 1,300 over the full range of the run with an approximate sex ratio of 2 males for each female.

Collection for hatchery broodstock will be given priority each week. All fish allocated for hatchery broodstock will be transported and held at Speelyai or Lewis River Hatchery. If the weekly quota for hatchery broodstock is not met, then all fish collected during subsequent weeks will be allocated for broodstock until the quota meets the predetermined broodstock collection curve.

All HOR fish collected prior to the peak of the run and designated as adult supplementation (upstream) fish will be transported and temporarily held at Lewis River Hatchery. All fish containing CWTs will be allocated to hatchery broodstock and transported to Speelyai Hatchery. A meeting will be held between PacifiCorp Aquatics Team and WDFW Fish Managers during the week of the anticipated peak of the run to discuss current run numbers and whether fish being held at Lewis River Hatchery can be taken upstream. If adult spring Chinook are returning at a rate at or above the projected running curve for that period, then all fish being held at Lewis River Hatchery will be taken upstream as well as any subsequent fish allocated for adult supplementation. If it appears that adult spring Chinook are returning at a rate exceeding the projected run curve, then it is possible that adults being held at Lewis River Hatchery could be taken upstream earlier. If it is decided that fish being held at Lewis River Hatchery will not be transported upstream by the peak of the run, they will continue to be held until hatchery broodstock goals have been met. Fish allocated for adult supplementation may be reallocated for hatchery broodstock if the adult return rate remains below the projected number.

All spring Chinook less than 24 inches will be considered jacks. Jacks will not comprise more than 5% of the broodstock collection or adult supplementation. Variations to this guidance will be decided in-season through ATS agreement.

2.3 Broodstock Collection Timing

Broodstock collection for the juvenile supplementation program should occur proportionately over the entire run timing. NOR Chinook should be transported upstream at the time of capture if at Merwin FCF or as soon as possible if at Lewis Hatchery trap. Figure B2 illustrates the trap timing of spring Chinook entering the Merwin FCF. Table B1 illustrates the 2023 spring Chinook generalized collection curve for each part of the program described in the previous section.

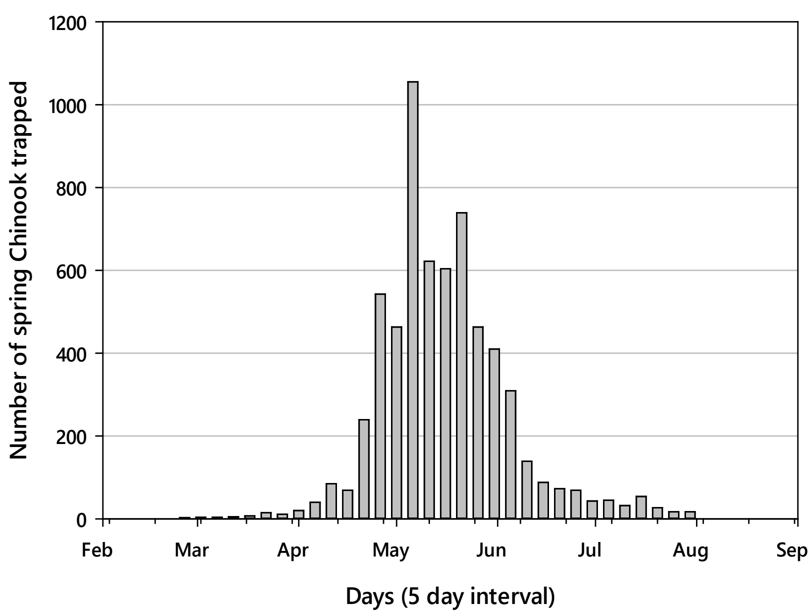


Figure B - 2. Actual number and timing of spring Chinook trapped at the Merwin FCF from 2016 to 2019.

Table B - 1. Spring Chinook generalized allocation schedule for broodstock and adult supplementation

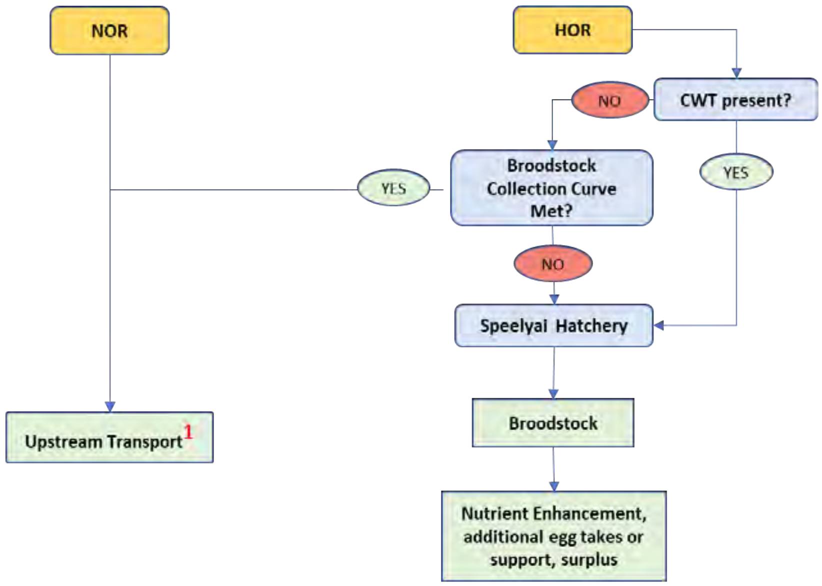
Appx. Date (Start of Week)	Hatchery Brood Stock	Adult Supplementation (Upstream)	Total (Weekly)	Total (Cumulative)	% Total (Cumulative)
2-Apr	21	48	69	69	2%
9-Apr	6	15	21	90	2%
16-Apr	13	28	41	131	3%
23-Apr	61	141	202	333	8%
30-Apr	69	159	228	561	13%
7-May	153	351	504	1065	25%
14-May	169	388	557	1622	38%
21-May	226	516	742	2364	55%
28-May	243	558	801	3165	73%
4-Jun	155	354	509	3674	85%
11-Jun	92	211	303	3977	92%
18-Jun	29	68	97	4074	95%
25-Jun	26	58	84	4158	97%
2-Jul	16	38	54	4212	98%
9-Jul	6	15	21	4233	98%
16-Jul	11	26	37	4270	99%
23-Jul	7	17	24	4294	100%
30-Jul	4	9	13	4307	100%
TOTAL	1,307	3,000	4,307	--	--

2.4 Adult Collection Methods

All broodstock are collected at either the Merwin FCF or Lewis River ladder.

2.5 Adult Disposition

Spring Chinook are either transported upstream or held for broodstock at Speelyai (or temporarily at Lewis River) hatchery depending on broodstock needs, origin and the presence of a CWT. Figure A2 illustrates the distribution protocol to be used for captured spring Chinook salmon.



[1] ACC may approve increased upstream transport numbers of NOR's based on run size. If not approved, all NOR's in excess of approved transport goal would be returned to the lower river (i.e., never surplused)

Figure B - 3. Sorting and distribution protocol for Lewis River spring Chinook collected at the Merwin FCF and Lewis River ladder

2.6 Data Collection Protocols

The following data will be recorded for all individuals and for all capture methods.

1. Capture Date (mm-dd-yyyy)
2. Capture Location (Merwin Trap, Lewis River hatchery, in-river)
3. Origin (NOR or HOR)
4. Sex (M/F)
5. Mark Status (PIT, CWT, AD)
6. Life Stage (adult, jack)

The following data will be recorded as a subsample of the total captures.

Data Type	HOR	NOR
Genetic Tissues	No sampling in 2023	
Fork Length	(CWT +): up to 100 (CWT -): none	(PIT -): up to 10 per week (PIT +): sample all

Scales	(CWT +): up to 100 (CWT -): none	(PIT -): up to 10 per week (PIT +): none[2]
Fecundity	Average Fecundity by spawn date (batch)	NA

2.7 Broodstock Holding Protocols

Broodstock are typically collected daily from April 1 through as late as August at the Merwin FCF or Lewis River ladder. All broodstock are transported to Speelyai Hatchery and held until spawning begins in mid-August (Table B2). The exception to this protocol is if the run size forecast is relatively low, fish collection may begin early and fish exceeding weekly broodstock goals may be held at Lewis Hatchery until the approximate half-way (50%) point in the run. At this point, the ATS will determine whether to transport all or a portion of the spring Chinook being held at Lewis hatchery. This determination will be made based on the number of broodstock currently held at Speelyai and predicted trap returns for the remaining run. That is, the likelihood that broodstock goals will be met. Spring Chinook held at Lewis River are treated as if they are to be transported and released upstream of Swift Dam. Broodstock that receive antibiotic injections are not transported, released, used for nutrient enhancement, or donated to any food banks or tribes due to mandated injection withdrawal periods.

2.8 Genetic Assignment and Analysis

See Section D, Objective 7 and Strategy G.

2.9 Spawning Protocols

All collected fully mature broodstock will be spawned using a pairwise (1x1) mating cross with a backup male. No fish shall be excluded except for those with overt disease symptoms or physical injuries that may compromise gamete fertility or viability. All fish are kill spawned, and disposition of carcasses is directed by the WDFW. Note that spawning protocols may change during the transition planning process.

3.0 JUVENILE PROGRAM IMPLEMENTATION

3.1 Egg Take Goals

Egg take required to meet hatchery production goals as set forth in the Lewis River Settlement Agreement (Settlement Agreement) include the following:

Spring Chinook target: 1,755,000 eggs

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[2] Age of PIT tag positive captures will be determined through PTAGIS records

3.2 Egg Incubation

Eggs are incubated in vertical stack incubators. Each female is assigned a number and only one female per tray, unless there are not enough trays towards the last egg take, then two or three fish will be pooled together until results are in from the enzyme-linked immunosorbent assay (ELISA) testing, if testing is performed (see Attachment A - Fish Health and Disease Strategy Plan).

3.3 Rearing and Release Schedule

All spring Chinook from fry to smolt are fed the highest quality feed available from WDFW contracted vendors. Fry will start out being fed 7 days per week. As they grow, the number of days fed per week will be reduced but will not be less than 3 days per week.

Hatchery staff will implement monthly performance sampling and a QA/QC sampling prior to release.

Immediately before the start of the volitional release period (pre-release group), additional sampling is conducted to assess precocity and assign a smolt index for a minimum of 100 smolts per release pond as part of ongoing morphology sampling (See Strategy F for Within-Hatchery Monitoring associated for the Spring Chinook Rearing and Release Study). This sampling is repeated at the end of the volitional release period, immediately before remaining fish are forced out (post-release group), to compare precocity between the pre and post release groups. These methods are described in detail in Strategy E.

Table B - 2. Hatchery production and collection timeline for North Fork Lewis River spring Chinook

	J	F	M	A	M	J	J	A	S	O	N	D	J	F	M	A	M	J	J	A	S	O	N	D	J	F
Adult Collection				X	X	X	X																			
Spawning								X	X																	
Incubation								X	X	X	X	X														
Rearing													X	X	X	X	X	X	X	X	X	X	X	X	X	X
Tagging															X	X										
Volitional Release																		X				X				X
Direct Release																		X				X				X

3.4 Feeding Type and Requirements

After ponding, at Speelyai Hatchery, spring Chinook fry are hand-fed with Bio Pro starter feed sizes #0, #1, #2, 1.2 mm and then Bio Clark's Fry 1.5 mm. Fish are fed 2 to 8 times per day. Feed volumes and frequencies follow a growth plan for each rearing group, and small adjustments are made weekly based on actual fish body sizes and feed conversion rates.

After transfer to Lewis River Hatchery, spring Chinook are hand-fed with Bio Clarks Fry once daily, 2 to 5 times per week depending on time of year, growth rates etc. Spring Chinook are hand fed by 1 to 2 staff for each pond.

3.5 Juvenile Marking and Tagging

Juvenile tagging type and location for hatchery-produced spring Chinook are presented in Table 6-1 of the H&S Plan (PacifiCorp and Cowlitz County PUD 2014). The number of tags and tagging groups may be modified annually as part of ongoing evaluations of rearing and release strategies (Strategy E).

A subset of juvenile spring Chinook that are collected at the FSC will be PIT tagged to provide additional information on juvenile transport survival at the release ponds and preliminary information on smolt out-migration timing (based on lower Columbia River detections) and out-of-basin avian predation (based on detections at bird colonies such as East Sand Island). The target is to tag approximately 10-15% of the parr or smolts (> 90mm) that are passed downstream from the Swift FSC. Juveniles captured at the FSC may be fish that were hatchery-reared and released from the juvenile supplementation program in previous years and overwintered upstream of Swift Dam or may be offspring of supplementation program adults.

The hatchery production goal is 1,350,000 smolts with the following three tagging groups:

- Adipose fin clip: 1,050,000
- Adipose fin clip and CWT: 150,000
- CWT only (DIT group): 150,000

3.6 Release Size and Number

Spring Chinook: 1,350,000 smolts at 8, 12, or 80 fish per pound depending on release group (see Table B3)

Volumetric methods and individual length measurements are used to estimate the number and size of fish released (see Strategy F). If the rearing goal is exceeded, surplus fish will continue to be reared and released with the program fish. However, this will require notification to NMFS prior to release. The intention of the spawning program is to not exceed the egg take or release targets and precautions should be employed (e.g., partial spawning) if targets are in jeopardy of exceeding the production limits set in this plan.

3.7 Release Timing and Locations

As described in Strategy E of this plan, most Lewis program spring Chinook (1,350,000) are volitionally released as yearlings in October or February from Lewis River Hatchery directly into the Lewis River. The volitional release includes pulling the screens, lowering the water level slowly over a 2-week period or until 90% or more of the smolts have left on their own. The remaining fish left are then flushed out.

2021 is the fourth year of a study designed to test release strategies and survival between up to five release groups. The study began with BY 2017 and is described in detail in Strategy A. Table B3 shows a summary of the different release groups planned as part of this study, which were designed to test the following variables: release month, date transferred to Lewis River Hatchery, rearing environment, ration level and size at release. This study will continue for at least 3 BYs and strategies will be evaluated each year and changes made if substantial problems are discovered. After 3 years of implementation, in-hatchery survival rates, size-at-release, condition factor at release, fish health (frequency or rates of disease), and physiological status at the time of release will be compared between treatment groups as described in Strategy E. All juveniles are released from the Lewis River Hatchery. Planned releases for 2023 (BY2021 and BY2022) are summarized in Table B-3. Deviations from this plan will be described during reporting.

Table B - 3. Summary of planned annual release groups as part of the spring Chinook rearing and release evaluation (Strategy D)

Release Group	Transfer Month to Lewis River Hatchery	Release Month	Size at Release (fpp)	Planned Tagging[2]		Planned Release (smolts)	Group Description
				AD + CWT	CWT ONLY		
1	May	February	8	37,500	37,500	150,000	Control group
2	December	February	12	37,500	37,500	175,000	Low ration, reared at Speelyai 6 months
3	December	February	8	37,500	37,500	150,000	Normal ration, reared at Speelyai 6 months
4	May	October	12	37,500	37,500	825,000	Released in October
5[1]	NA	June	80	0	50,000	50,000	Released in June
		TOTAL		**150,000**	**200,000**	**1,350,000**	

[1] *A minimum of 50,000 fish will be planted, but if surplus juveniles are available due to better than expected survival etc., they would be released in this group. All fish from this release group will be adipose fin-clipped; up to 50,000 fish will be marked with CWT.*

[2] *The number and type of tags distributed for each release group may be modified as recommended by the ATS based on projected surplus or deficit to planned release numbers.*

4.0 ADULT SUPPLEMENTATION UPSTREAM OF SWIFT DAM

Up to 3,000 spring Chinook (when available) will be transported from the Merwin FCF and Lewis River traps (or acceptable alternative stock) to Eagle Cliff or designated areas upstream of Swift Dam. Ideally, the transport goal would be entirely of natural origin spring Chinook. At present, all NOR returns are transported upstream, however, there are insufficient NOR returns to reach the transport goal. Therefore, HOR spring Chinook (when available) are also transported upstream in an effort to reach the transport goal of 3,000 spring Chinook.

The transport goal of 3,000 spring Chinook is based on EDT analysis completed in 2019. This goal is likely to change as more information becomes available (e.g., IPM's) and should be reviewed as part of the adaptive management component of this plan. If exceedance of the transport target is recommended by the ATS in any given year (i.e, returns exceed broodstock and upstream transport target), the ATS would seek approval by the ACC prior to exceeding the transport target in this plan.

A minimum of two tanker fish trucks will be used weekly to move captured spring Chinook upstream. Each tanker truck can transport about 100 adult Chinook salmon. Table B - 4 provides a proposed transportation schedule, indicating biweekly numbers to achieve the transport goal of 3,000 over the run period; however, this schedule will not be possible to achieve if run sizes are low relative to the broodstock goal. In years with low pre-season run forecasts, fish will be held at Lewis Hatchery until broodstock goals are met, as described in AOP sections 2.1 and 2.2.

Prior to 2017, transported spring Chinook were released at different locations upstream of Swift Dam to enhance their distribution into streams (seed planting). Eagle Cliff, Muddy River Bridge, Clear Creek Bridge, and Curly Creek Bridge were used to release an approximately equal portion of the transported spring Chinook. To simplify the logistics, fish trucks rotated the release location of each haul. For example, the first load was released at Eagle Cliff, the second at Muddy River Bridge, and so on, resulting in nearly equal portions released at each site. Eagle Cliff was chosen as a preferred site for release as it is not affected by reservoir fluctuations and provides the opportunity for released fish to migrate upstream immediately without having to migrate through reservoir waters that can exceed optimal water temperatures. Distribution of spawning will be monitored annually to determine if spawning distribution is adequate and protocols are adapted as needed.

Table B - 4. Recommended transportation timing of adult spring Chinook for supplementation upstream of Swift Dam

Time Period	Target number of spring Chinook transported upstream of Swift Dam
Apr 15-30	300
May 1- 15	600
May 15-31	1,125
Jun 1-15	750
Jun 15-30	225
TOTAL	**3,000**

SECTION C COHO SALMON

1.0 INTRODUCTION

The Lewis River coho salmon program has two components, upstream adult supplementation and downstream hatchery production. The goal of the adult supplementation program is to transport up to 6,800 early and late adult coho (both NOR and HOR) to the upstream end of Swift Reservoir. This target number of adults was determined through the Ecosystem Diagnostic and Treatment (EDT) process which defines habitat capacity upstream of Swift Dam. The intent of the adult supplementation program is to increase the number of NOR coho salmon returning to the North Fork Lewis River with a long-term goal of passing only NOR coho salmon. The hatchery production goal is to release 1,100,000 segregated early coho smolts and 900,000 integrated late coho smolts annually. The minimum target for NOR integration into late coho hatchery production is 30% per HSRG guidance (HSRG 2014).

2.0 ADULT PROGRAM IMPLEMENTATION

The following section describes the protocols for implementing the coho program of the H&S Plan.

Prioritized Goals for management of returning Lewis River Early Coho:

1. Lewis River broodstock goal
2. Minimum upstream supplementation goal (1,000 Pairs may include NORs and some HORs as needed – early/late Coho)
3. A fishery managed to allow for #1 and #2 to be achieved
4. Additional upstream supplementation (target is 6,800 early and/or lates) and other in-basin programs (none currently planned)
5. Out of basin programs (none currently planned)

Prioritized Goals for management of returning Lewis River Late Coho:

1. Lewis River broodstock goal
2. U.S. v. OR[3] (in combination with other Cascade stratum sources, i.e., Washougal/Kalama)
3. Minimum upstream supplementation goal (1,000 Pairs may include NORs and some HORs as needed – early/late Coho)
4. A fishery managed to allow for #1 -#3 to be achieved
5. Additional upstream supplementation (target is 6,800 early and/or lates) and other in-basin programs (e.g., Educational Remote Site Incubators)
6. Other out of basin programs (none currently planned)

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[3] See 2008-2017 *United States v. Oregon* Management Agreement, May 2008

2.1 Broodstock Source and Selection

Broodstock source for the supplementation program shall be composed of both early (Type S) and late coho (Type N) returning to either the Merwin FCF or Lewis River Hatchery ladder.[4] For adult supplementation, the Merwin FCF is preferred because these fish are assumed to be upstream migrants attempting to reach areas above Merwin Dam. The Lewis River Hatchery ladder will be used primarily for hatchery broodstock collection. All early coho NORs should be passed upstream. A portion of late coho NORs are used for the late coho integrated hatchery program (integration rate minimum goal of 30%).

2.2 Broodstock Collection Goal

In most years, the number of coho salmon returning to traps has been sufficient to achieve both hatchery and upstream supplementation targets of about 10,000 adults (Figure C1). Broodstock comprise both returning adult and precocious males (jacks). The proportion of jacks integrated into the hatchery broodstock may include up to 10% of male spawners (HSRG recommendations). WDFW guidance is for at least 2% of male spawners to be jacks (WDFW HEAT Summer Meetings Handout – Jack Utilization Guidelines and Spawning Citations).

<u>Hatchery Broodstock:</u> Up to 1,400 HOR early adults, depending on fecundity, will be used as broodstock to support the segregated hatchery production goal of 1,100,000 smolts (released annually). An additional 1,000 late returning HOR and NOR adults will be used to support integrated hatchery production of about 900,000 smolts (released annually). The minimum target for NOR integration is 30% for late coho per HSRG guidance. Note that the ATS may discuss changing the broodstock target for 2023 in order to meet requirements of the transition plan.

[4] On July 21, 2015, the H&S Subgroup agreed to incorporate late coho as a supplementation stock. This decision was affirmed by the ACC on August 13, 2015.

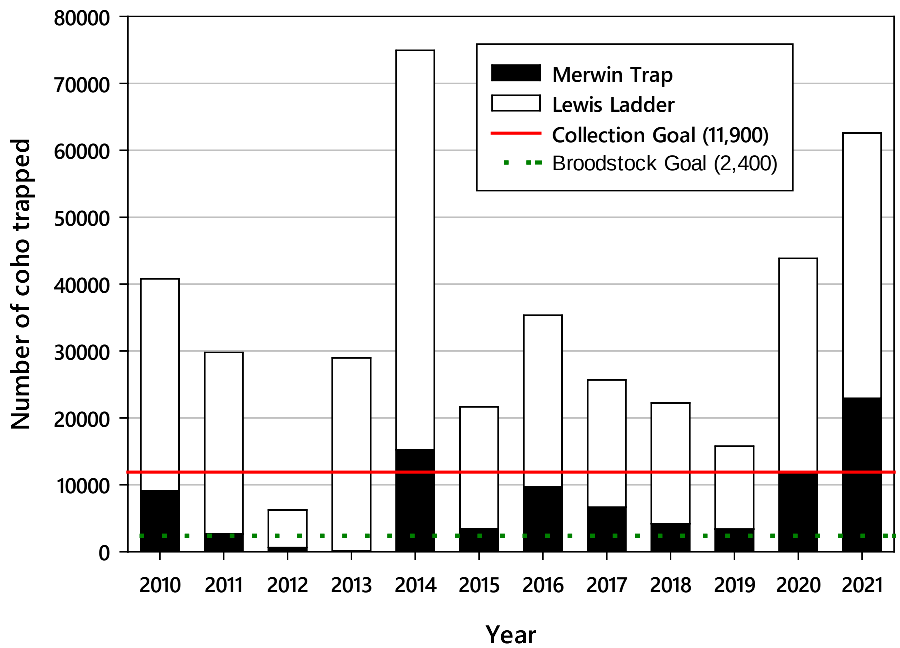

Figure C - 1. Total number of coho trapped annually between 2010 and 2021 at the Merwin Fish Collection Facility and Lewis Hatchery ladder

Note: Collection target line (11,900) represents number of early and late coho needed to meet hatchery broodstock (2,400) and adult supplementation goals (9,500).

2.3 Broodstock Collection Timing

Because the coho program relies on trapping, broodstock collection should occur proportionately over the trap collection curve. Early coho begin entering trapping facilities in early September and peak capture rates are observed in mid to late October. Late coho begin entering trapping facilities in late October and continue through December (Figure C2). Table C-1 provides a proposed collection curve for both early and late coho that is consistent with HSRG recommendations to collect broodstock over the entire collection window.

During the last 2 weeks of October when both early and late stocks are arriving at the traps, staff will visually assign fish to a stock based on coloration and maturation. Fish that cannot be clearly identified by stock are passed upstream unless they are in poor condition, in that case they would be used for nutrient enhancement or allocated to surplus.

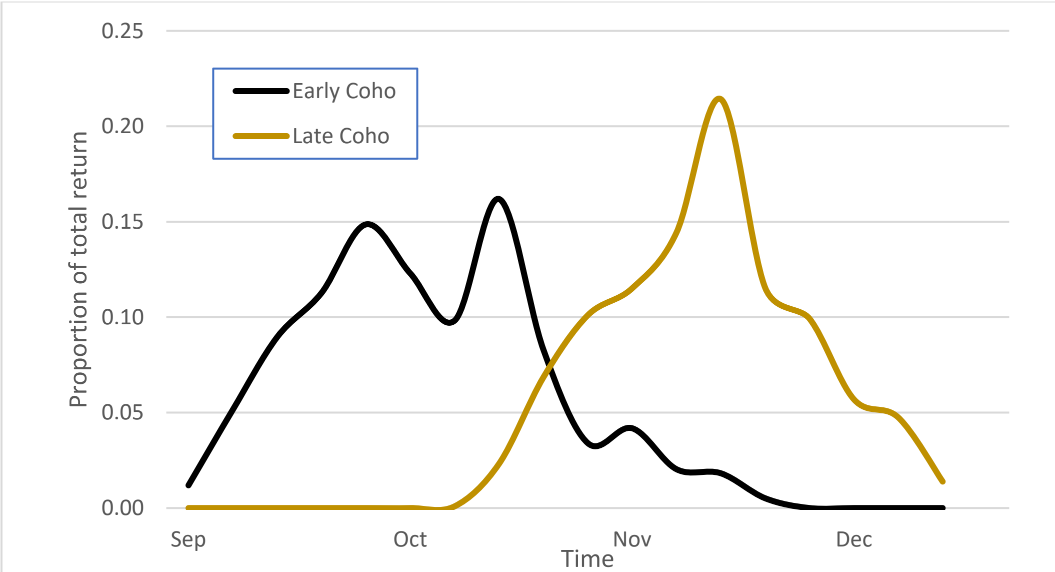

Figure C - 2. Trap entry timing for early and late coho at the Merwin Trap 2017 - 2021.

Table C - 1. Hatchery broodstock collection curve for early and late coho

Period	Number of Coho	Relative Proportion	Relative Percent of the Run	Total (Cumulative)	% Total (Cumulative)
Sep 1-15	50	0.02	2%	50	2%
Sep 16-30	400	0.17	17%	450	19%
Oct 1-15	450	0.19	19%	900	38%
Oct 16-31	550	0.23	23%	1450	60%
Nov 1-15	300	0.13	13%	1750	73%
Nov 16-30	300	0.13	13%	2050	85%
Dec 1-15	200	0.08	8%	2250	94%
Dec 16-31	150	0.06	6%	2400	100%

2.4 Adult Collection Methods

Coho salmon are collected from both the Merwin FCF and Lewis River Ladder. Coho designated as broodstock are held at either Speelyai (earlies) or Lewis River (lates).

2.5 Adult Disposition

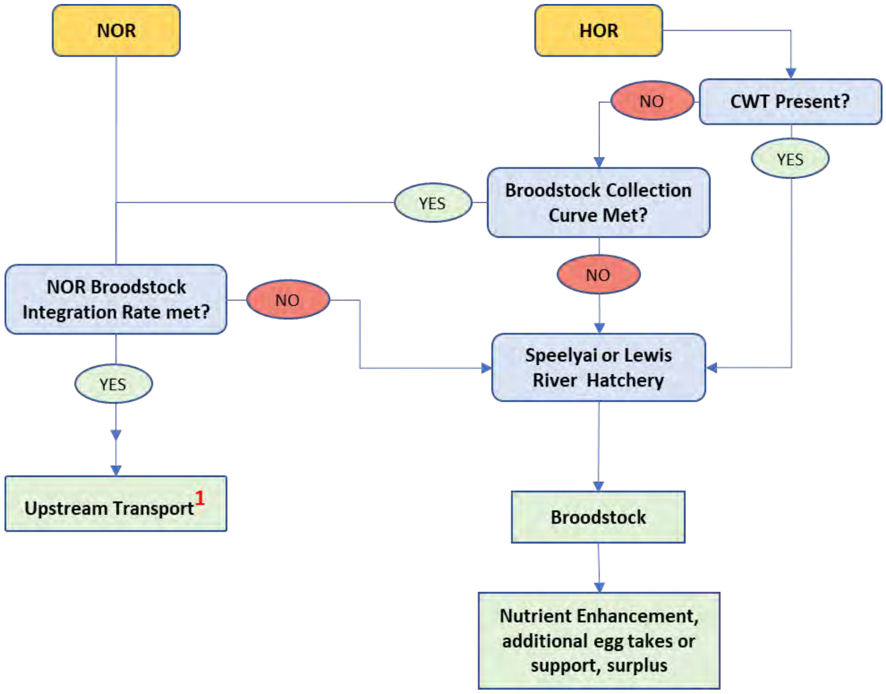

Figure C - 3. Sorting and distribution protocol for Lewis River coho salmon collected at the Merwin trap or Lewis River Ladder

[1] *ACC may approve increased upstream transport numbers of NOR's based on run size. If not approved, all NOR's in excess of approved transport number would either be returned to lower river or integrated into the hatchery broodstock (i.e, never surplused)*

2.6 Data Collection Protocols

The following data will be recorded for all individuals and for all capture methods.

1. Capture Date (mm-dd-yyyy)
2. Capture Location (Merwin Trap, Lewis River hatchery, in-river)
3. Origin (NOR or HOR)
4. Sex (M/F)
5. Mark Status (PIT, CWT, AD)
6. Life Stage (adult, jack)

<u>The following data will be recorded as a subsample of the total captures.</u>

Data Type	**HOR**	**NOR**
Genetic Tissues	Coho: up to 200 samples[5]	
Fork Length	(CWT +): up to 100 (CWT -): none	(PIT -): up to 10 per week (PIT +): sample all
Scales	(CWT +): up to 100 (CWT -): none	(PIT -): up to 10 per week (PIT +): none
Fecundity	Average Fecundity by spawn date (batch)	Average Fecundity by spawn date (batch)[6]

2.7 Broodstock Holding Protocols

Coho broodstock collected at Lewis River Hatchery trap or Merwin FCF are either transported to Speelyai Hatchery for spawning (early coho) or held and spawned at Lewis River Hatchery (late coho).

2.8 Genetic Assignment and Analysis

See Section D, Objective 7 and Strategy G.

2.9 Spawning Protocols

All collected fully mature broodstock will be spawned using a pairwise (1x1) mating cross with no backup male, unless insufficient milt is obtained from the selected male. Wild adults are not currently incorporated into the early Coho segregated program broodstock. Up to 30 percent of the late Coho integrated program broodstock may be comprised of wild fish collected at MCF or Lewis River Hatchery. All wild brood that are rip at the time of spawning are utilized. The integrated portion of the broodstock is spawned with crosses of HxW, WxW or HxH. No fish shall be excluded except for those with overt disease symptoms or physical injuries that may compromise gamete fertility or viability. All fish are kill-spawned, and disposition of carcasses is directed by the WDFW. Note that spawning protocols may change during the transition planning process.

[5] Three year sampling effot to establish baseline using hatchery broodstock split among early, lates, sex and origin

[6] Late coho fecundity is averaged among HOR and NOR broodstock (i.e., separate average estimates for HOR and NOR broodstock)

3.0 JUVENILE PROGRAM IMPLEMENTATION

3.1 Egg Take Goals

Egg take required to meet hatchery production goals as set forth in the Lewis River Settlement Agreement (Settlement Agreement) include the following:

- Early Coho: 1,800,500
- Late Coho: 1,400,000

3.2 Egg Incubation and Juvenile Rearing

Early Lewis River coho are spawned at Speelyai Hatchery and the resulting eyed eggs are shipped to the Lewis River Hatchery in November for incubation in vertical stack incubators. Late Lewis River coho are spawned and reared at Lewis River Hatchery.

According to WDFW, incubation conditions are consistent with loading densities recommended by Piper et al. (1982). Water quality and temperatures are generally very good. Stack flows during incubation are 3.6 gallons per minute and all eggs are treated with formalin to keep them free of fungus (WDFW and PacifiCorp 2014).

Hatchery staff will implement performance sampling prior to first feeding and a QA/QC sampling prior to release. These methods are the same as described above in late winter steelhead Section 2.16.

3.3 Rearing and Release Program Schedule

Table C - 2. Hatchery production and collection timeline for North Fork Lewis River coho salmon

	Jan	Feb	Mar	Apr	May	Jun	Jul	Aug	Sep	Oct	Nov	Dec
Adult Collection												
Spawning												
Incubation												
Rearing												
Tagging												
Volitional Release												

3.4 Feeding Type and Requirement

At the time of ponding, fry are fed 7 days per week, 6 to 8 times per day. Feedings are tapered down to once per day, 2 to 3 times per week as the growth cycle progresses, based on maintaining fish growth and size targets. Fish are fed BioVita starter feed, then BioClark's Fry.

3.5 Juvenile Marking and Tagging

Juvenile tagging type and location for coho salmon are presented in Table 5-1 of the H&S Plan (PacifiCorp and Cowlitz County PUD, 2014) and summarized below. Coho are mass-marked in June when they are about 120 fpp, as follows:

- 1,700,000 AD only
- 150,000 CWT only (double-index tag group)
- 150,000 CWT + AD

A subset of juvenile coho that are collected at the FSC will be PIT tagged to provide additional information on juvenile transport survival at the release ponds and preliminary information on smolt out-migration timing (based on lower Columbia River detections) and out-of-basin avian predation (based on detections at bird colonies such as East Sand Island). The target is to tag approximately 10-15% of the parr or smolts (> 90mm) that are passed downstream from the Swift FSC. Juveniles captured at the Swift FSC are most likely offspring from adult supplementation, or alternatively from residualized coho that eventually become mature and spawn. This scenario, however, has not been observed during fall spawning ground surveys in the upper basin or reservoir tributaries.

3.6 Release Size and Number

- Early Coho – 1,100,000 smolts at 14-16 fish per pound
- Late Coho – 900,000 smolts at 16 fish per pound

Volumetric methods and individual length measurements are used to estimate the number and size of fish released (see Strategy F). If the rearing goal is exceeded, surplus fish will continue to be reared and released with the program fish. However, this will require notification to NMFS prior to release. The intention of the spawning program is to not exceed the egg take or release targets and precautions should be employed (e.g., partial spawning) if targets are in jeopardy of exceeding the production limits set in this plan.

3.7 Release Timing and Location

Coho are volitionally released at Lewis River Hatchery beginning in April by pulling the screens, lowering the water level slowly over an approximately 2-week period (up to 6 weeks) or until approximately 90% or more of the smolts have left on their own. Remaining fish are flushed directly to the river prior to May 20. Prior to beginning the volitional release, an area Fish Health Specialist will evaluate the coho release group's health and condition.

4.0 ADULT SUPPLEMENTATION UPSTREAM OF SWIFT DAM

The supplementation program relies exclusively on transporting adults upstream of Swift Dam, which began in 2012. Supplementation adults are able to spawn naturally using all available habitats upstream of Swift Dam. Progeny from these transported adults will be collected at the FSC and transported downstream of Merwin Dam to begin their migration to the sea. The program targets up to 6,800 early or late adult coho to be transported over the duration of the run timing. This target was selected through the EDT process to define the spawning capacity upstream of Merwin Dam. The number of NOR coho available for upstream supplementation depends on return rates to the traps and needs of the integrated late coho hatchery program.

Previous trapping data for natural origin coho[7] (Figure C2) are used to create a potential collection schedule to meet the target goal of 6,800 coho (Table C2) for transport in 2023. Ideally, all transported coho would be NORs. However, there are not enough NOR coho to meet the supplementation goal. In addition, Lewis River Hatchery is currently implementing an integrated late coho program on the Lewis River that will use a portion of NOR late coho as broodstock. The supplementation program will use all NORs available that are not used for the integrated late coho hatchery production program.

Transported coho may be released at different locations upstream of Swift Dam to enhance distribution into streams and tributaries. Eagle Cliff, Muddy River Bridge, Clear Creek Bridge, and Curly Creek Bridge will be used to release an equal portion of the transported coho. To simplify the logistics, fish trucks will work on a rotating basis for each haul. For example, the first load will be released at Eagle Cliff, the second at Muddy River Bridge, and so on. This may not equate to equal portions for each site but should be reasonably close.

Table C - 3. Proposed collection rate of coho for broodstock and upstream transport indicting relative and cumulative proportion by two-week period over the collection window

Period	Coho for upstream Transport*	Relative Proportion	Cumulative Proportion
Sep 1-15	200	0.02	
Sep 16-30	1600	0.17	0.19
Oct 1-15	1800	0.19	0.38
Oct 16-31	2100	0.22	0.60
Nov 1-15	1200	0.13	0.73
Nov 16-30	1200	0.13	0.85
Dec 1-15	800	0.08	0.94
Dec 16-31	600	0.06	1.00
Total	9500		

** Values based on supplementation goal of 9,500 adults*

The actual number of adult coho transported may be modified (in-season) by the ATS based on actual returns to the hatchery and traps. The ATS may raise the total number of coho transported upstream to 9,500 adults without prior approval of the ACC. This value was agreed

[7] NOR coho returns may be progeny from upper river (supplementation program) or lower river spawners. There is no way to differentiate the two groups; However, this is not required because both groups are treated as the same population in this plan.

to by the ACC in previous years when returns to the traps exceeded broodstock and transport targets.

SECTION D MONITORING AND EVALUATION

1.0 INTRODUCTION

Monitoring activities described in this section are intended to meet monitoring objectives contained in the H&S Plan. Objectives are established to monitor population metrics related to abundance, distribution, composition, and potential ecological interactions of hatchery released smolts. Evaluation of the data collected to address these objectives and reporting on how the data trends change over time is critical for assessing population viability (extinction risk) of target populations.

The H&S Plan also lays out "key questions" that are nested within each objective. The key questions direct the research needed to support each objective and are answered by monitoring indicators. The H&S Plan also describes narratively the purpose, population recovery monitoring recommendations, proposed strategies, monitoring indicators, sampling frequency, and limitations or concerns for each objective.

This AOP is intended to provide the necessary level of detail to implement the monitoring component of the H&S plan as described in this section and through the various strategies attached to this plan.

Generally, study methods proposed in this AOP follow established protocols used in the Pacific Northwest. This allows methods to be standardized or improved based on data collection or results from other regional locations. An important component of some objectives is the accuracy and precision with which specific objectives are measured or quantified. NOAA Fisheries has provided guidance with respect to variation (Crawford and Rumsey, 2011), and the intent of this plan is to strive to meet these precision guidelines when practical.

1.1 Objectives

The M&E objectives are classified into four main categories:

- *Administrative:* Includes the reporting and planning documents required by the Settlement Agreement, HGMPs and Biological Opinion(s)
- *Hatchery Monitoring:* The purpose of hatchery evaluation objectives is to operate hatchery programs in a way that maximizes survival and health of program fish to meet production targets and reduces adverse effects on naturally produced ESA listed species.
- *Abundance Monitoring:* Includes objectives related to monitoring trends in juvenile and adult abundance to evaluate the status, trend, and viability of North Fork Lewis River populations of salmon and steelhead.
- *Risk Assessment:* These objectives are directed at monitoring potential risks of hatchery and supplementation programs to ESA listed species.

The hatchery, abundance, and risk assessment monitoring objectives are presented in a standardized format with key questions nested within each objective. For each key question, the general monitoring approach and methods are described (when appropriate) to develop an estimate for each of the metrics associated with each key question. Decision points for adaptive management and limitations or concerns are also described for each monitoring approach (when appropriate).

1.2 Key Questions

Each of the objectives (excluding administrative objectives) have a number of related key questions, presented in the H&S plan. The key questions are specific to each objective and are intended to ensure that specific metrics or benchmarks are addressed in annual reporting. The list of key questions provided in the H&S plan is not intended to be a list of obligations. Rather, the key questions provide monitoring guidance and focus for each of the H&S plan objectives to ensure metrics related to recovery are addressed (e.g., abundance, productivity, diversity and spatial structure). The ATS will determine which key questions are to be addressed annually or periodically (e.g., every 3 years) in the AOP.

1.3 Strategies

Some objectives have complex monitoring designs that often have a higher potential for frequent modifications. To adapt, the ATS reorganized the format used to address different monitoring objectives of the H&S Plan by adding ‘strategies’ to the AOP in 2021. Strategies are standalone planning documents attached to the AOP that follow the same general framework as described below. However, strategies generally provide a more detailed study design as is often required by more complex evaluations. Strategies are essentially ‘living’ plans and components to the AOP that can be updated independently throughout the season, or in future years, without requiring global reformatting of the AOP.

Strategies included in the 2023 AOP include:

Strategy A: Adult Abundance and Composition

Strategy B: Adult Spatial and Temporal Distribution

Strategy C: Juvenile Abundance and Migration

Strategy D: Fish Health Monitoring and Disease Prevention

Strategy E: Spring Chinook Rearing and Release Evaluation

Strategy F: Precocity and Morphology Sampling

Strategy G: Genetic Risk Monitoring

Strategy H: Volitional Release

Strategy I: Smolt-to-Adult Return Rate Estimation

Strategy J: Sampling and Data Collection Checklist

1.4 Framework

Following the framework set forth in the H&S Plan, this section of the AOP expands upon the monitoring strategies, monitoring indicators, sampling frequency and limitations or concerns for addressing each key question. The description of these elements follow a standardized template as follows:

Approach: Briefly, the approach used to quantify and estimate monitoring indicators with an acceptable level of precision and accuracy to address the objective or key questions. Includes references to other sections of the AOP or attached Strategies, where relevant.

Metric or Monitoring Indicator Name and Description: The desired numerical measurement or observation by which the objective is measured.

Targets: The program element endpoint or numeric value that the hatchery and supplementation program seeks to achieve (including precision for numeric targets)

Field Methods: Description of the specific methodology, sampling designs and protocols to collect and store field data in a format required by the analytical methodology adopted.

Analytical Methods and Reporting: Description of the application of the data or specific analysis applied to derive an estimate for each metric assigned to the objective or key question. Includes the description of any formulas, estimators or software used to analyze data sets.

Frequency and Duration: The sampling or data collection frequency and duration for each objective or key question that is being monitored. For example, steelhead redd surveys are conducted once every seven to ten days between March 1 and June 30.

Data Collection and Storage: Parties responsible for the data collection, storage, and location of data files.

Limitations or Concerns: General description noting specific challenges especially those related to field data collection and deployment.

2.0 OBJECTIVES

2.1 Administrative Objectives

The purpose of objectives 1.0 and 1.1 is to obtain ESA coverage for hatchery production and associated program activities. The HGMP represents the proposed operation of each hatchery program and is submitted to NOAA Fisheries for approval. Once approved, NOAA Fisheries will draft and finalize a Biological Opinion regarding the HGMP action and include specific terms and conditions, and reasonable and prudent measures to avoid jeopardizing ESA listed species from continued operation of the hatchery programs.

The purpose of objectives 2.0 through 2.3 is to ensure that reporting and planning requirements of the Settlement Agreement, HGMPs, and Biological Opinion (once issued) are met. The annual hatchery operations and H&S Program reports shall demonstrate whether the HGMP protocols are implemented as proposed. Reporting will include assessing the effectiveness of actions taken to limit the threat of hatchery operations to natural-origin fish as well as documenting whether each hatchery production program is meeting target production levels.

Objective 1.0: NOAA acceptance of a Hatchery and Genetic Management Plan (HGMP) for each hatchery program on the North Fork Lewis River

The following HGMPs are anticipated:

- Summer Steelhead
- Late Winter Steelhead
- Early Winter Steelhead (Chambers Creek stock)
- Spring Chinook
- Early Coho Salmon
- Late Coho Salmon

The ATS will receive updates on this process from WDFW throughout the year. It is anticipated that draft versions of the HGMPs will be provided to ATS members for review; however, the ATS (with the exception of PacifiCorp) will not formally review or comment on the HGMPs. The ATS anticipates discussing issues related to the HGMP's that may influence or modify the goals and objectives of the H&S program. Examples include transition planning from segregated to integrated hatchery programs, harvest management and hatchery production program sizing

Objective 1.1: Receive Biological Opinion for all submitted HGMPs

Continued operation of the hatcheries is critical as the supplementation program relies on hatchery returns for reintroduction efforts upstream of Swift Dam.

Biological Opinions (BiOps) for the Lewis River Hatchery programs will be issued after HGMPs are submitted and accepted by NOAA Fisheries. The ATS will receive updates on this process from WDFW and NOAA Fisheries throughout the year. Once issued, the ATS will need to determine whether the the current H&S Plan and AOP are consistent with BiOp and make necessary revisions to the H&S Plan and AOP to ensure consistency with the BiOp.

Objective 2.0: Finalize a Hatchery and Supplementation Plan every 5 years

The current H&S plan was submitted to the FERC in December 2020. The FERC approved the plan on March 28, 2022. The next rewrite of this plan is scheduled for completion 5 years from FERC acceptance of the H&S plan (March 2027), or as extended by the FERC. The H&S Plan will be revised earlier if required by the HGMPs in Consultation with the ACC and NOAA Fisheries to adaptively manage the programs.

Objective 2.1: Finalize an Annual Operating Plan (AOP)

The AOP is the primary mechanism for adaptively managing the H&S Program. The AOP is developed collaboratively by the ATS on an annual basis and requires approval by the ACC and Services.

The ATS strives to finalize the AOP by December 31 of each year. This is not always possible and the ATS has developed a prioritization protocol for when the AOP is not completed by December 31. This protocol prioritizes the development of the AOP into two distinct phases. Phase 1 focuses on field data collection and monitoring that is initiated during the first half of the year, while Phase 2 focuses on the second half of the year. This allows the ATS to complete monitoring and reporting requirements for those programs that are initiated earlier in the year such as late winter steelhead monitoring and juvenile trapping. Necessary AOP revisions for Phase 1 activities are scheduled for completion by February 1. Phase 2 includes primarily fall monitoring activities such as adult salmon abundance monitoring and is scheduled for completion by July 1.

Objective 2.2: Finalize an Annual Operations Report

PacifiCorp drafts the Annual Operations Report (AOR) in accordance with section 8.2.4 of the Settlement Agreement, outlining reporting requirements pursuant to the AOP. The AOR is provided to the ACC as part of the annual TCC and ACC reporting (Section 14.2.6 of the Settlement Agreement). The AOR reports on all monitoring activities described by the AOP. Section F of the AOP provides the minimum reporting requirements for the AOR as stipulated in the Settlement Agreement. The final AOR is submitted as part of the annual TCC and ACC annual report to the FERC by June 30 of every year.

Objective 2.3: Finalize an Annual Hatchery Report

WDFW drafts the Annual Hatchery Report which summarizes on site hatchery activities including spawning, rearing, feeding, pathogen testing, permit compliance, fish marking, and trapping counts. Because many of the hatchery activities are related to monitoring objectives in the H&S Plan and in Section 8.2.4 of the Settlement Agreement, PacifiCorp attaches the Hatchery Report in their submittal of the Annual Operations Report to FERC. At a minimum, the Hatchery Report includes the following metrics:

- Broodstock received from adult trapping operations, including disposition of adults received.
- Mortality of adults, juveniles and eggs
- Spawning, incubation and rearing summary
- Disease presence, prevention (treatments), and loss by life stage
- Growth rate by month from fry ponding to release as smolts
- Number of fish tagged, tag type, and purpose (experimental, production)
- All fish transfers in or out of the basin including species, number, marks, and life stage
- Production summary providing the total number of smolts released including the timing and locations of released and average smolt size at release.
- Volitional release data including volitional release timing and duration for all species.
- Monthly water temperatures and average rainfall
- Summary of maintenance and capital projects completed

2.2 Hatchery Monitoring Objectives

Objective 3.0: Determine whether hatchery production protocols incorporate best available management practices to support program targets and goals.

The purpose of objective 3.0 is to implement hatchery programs and practices that support the goals of the H&S program, are consistent with best management practices, and incorporate recommendations by the HSRG when possible. This objective also encourages hatchery programs to incorporate new scientific advances when available to continually improve overall hatchery performance in supporting the H&S program.

KEY QUESTION 3A. DO HATCHERY BROODSTOCK COLLECTION PROTOCOLS SUPPORT PROGRAM GOALS?

APPROACH

H&S Program goals are to 1) collect broodstock throughout the entire run timing for each species and 2) ensure the portion of broodstock retained from the total number of fish collected (trapped) follows the composition and retention rates established in collection curves for each species. Details on broodstock collection goals and timing are provided for individual species in Sections A2, B2, and C2.

- Trap entry timing is the number of adults trapped by species, stock and origin, and date (Merwin trap) or week (Lewis River Hatchery Ladder).
- Broodstock collection and selection timing is the number of adults retained for broodstock by species, stock and origin, and date (Merwin trap) or week (Lewis River Hatchery Ladder).
- Broodstock retention rate is the number of adults transferred to hatcheries for broodstock out of the total number of adults trapped by time period, species, stock, origin, and sex.

LIMITATIONS OR CONCERNS

In the early part of the year, if the broodstock collection targets are not projected to be met due to poor predicted returns, the ATS may decide to modify the broodstock collection curve by adjusting the collection timing or number of broodstock to be held at the hatchery. During the collection season, if the trap entry timing or broodstock retention rate deviates significantly from the planned collection curve without justification (e.g., due to poor realized returns), an in-season decision by the ATS may be needed to ensure that broodstock are collected throughout the duration of the run.

Trap entrance timing may not be a precise or accurate indicator of migration or spawn timing. Returning adults may choose to reside in the river for several weeks or months before volunteering into one of the traps. However, because traps are the primary source of hatchery broodstock, trap entry timing is used to determine whether broodstock collection protocols are consistent with targets.

DATA COLLECTION METHODS

Metric or Monitoring Indicator	Targets	Field Methods	Analytical Methods	Frequency and Duration
3A.1.Trap entry timing	Broodstock collection follows collection curves over the course of the trapping period.	See broodstock collection goals and timing for individual species in Sections A2, B2 and C2 of the AOP.	Sum of adults trapped by day or week. Identify first, last, and peak run dates from distribution of daily trap counts.	Data are collected when traps are sorted; daily at Merwin Trap, weekly at Lewis River Hatchery Ladder. Broodstock collection periods: • Steelhead: late January to end of May • Spring Chinook: April 1 to late August • Coho: early September through December
3A.2.Broodstock retention rate	Total broodstock target numbers are met. Broodstock collection rates that are consistent with planned broodstock collection curves for each species. For integrated programs, match collection timing to average NOR return timing.		Broodstock retention rate reported by week as the number of fish held for broodstock out of the total number of fish trapped and sorted (daily/weekly counts, cumulative total, and % cumulative total of annual total). Make comparisons of annual HOR and NOR return timing to 5-year average return time, and to planned and observed broodstock collection curves.	

Key Question 3B. Do spawning, rearing and release strategies support program goals?

Approach

The following metrics are assigned to this key question to ensure hatchery practices are consist with HSRG recommendations and best practices. In general, data on these metrics are collected as part of typical hatchery program operations and will be reported on in the Annual Operating Report, and trends will be evaluated as part of the Adaptive Management process described in Section E.

Not all metrics assigned to this key question are evaluated annually, for instance metrics related to water quality (e.g., temperature, TDG, etc.), avian predation or feeding strategies are evaluated periodically as determined by the ATS.

Integration Rates: For the integrated programs, late winter steelhead and Type N (late) Coho, integration rate targets are described in the Broodstock Collection Strategies (sections A 2.1 and C 2.1 of this document). For steelhead, a "mining rate" strategy is currently in use until the program can transition to use of 100% natural-origin spawners. A 30% fixed mining rate will be used to collect broodstock for the integrated steelhead program. For Coho, the minimum integration rate is 30%.

Spawning matrices and timing: Spawning designs are developed to be aligned with HSRG guidance for conservation of genetic diversity to ensure genetic diversity and relatedness targets are being met (when appropriate). Spawning designs ensure equal contribution from all spawners to the progeny and reduce the effects of artificial selection. Adults selected for broodstock should be similar in size and arrival timing to their natural counterparts whenever possible. Spawning crosses may follow pairwise, factorial or nested designs depending on the program goals (whether the program is integrated or segregated), to mitigate unequal genetic contributions among males, and ensure integration rates of NOR broodstock meet HSRG recommendations (when applicable). Target spawning matrices and timing are described in Sections A 2.8, B 2.8, and C 2.8.

Fecundity: Fecundity is monitored during hatchery spawning to ensure the number of broodstock collected will meet juvenile production targets. If fecundity declines over years, the targeted number of adults needed to achieve hatchery production or natural productivity upstream of Swift Dam may need to be adjusted. The relationship between fish length and fecundity may be evaluated to understand whether trends in fecundity are related to trends in body size. For steelhead, fecundity is measured for individual fish by volumetrically measuring the number of eggs for each female spawned and fork length of each female spawned. For Coho and Chinook, fecundity is estimated from the average number of eggs per female for a given spawn group of females. Fork lengths may be obtained from HOR female spawners processed for CWTs.

Feeding Rations and Delivery Rate: Not applicable in 2023

Avian Predation: Not applicable in 2023

Volitional Release: Refer to Sections A3.7, B3.7, and C3.7 for hatchery release timing for each species. For all species, fish are allowed to leave volitionally for at least a period of 2 weeks. Fish that remain after 2 to 6 weeks, depending on species, are moved into the river by a forced release. See Strategy H for practices at Lewis River Hatcheries and a review of the rationale for various approaches on the timing and duration of volitional release periods.

Water Quality: A TDG mitigation and evaluation plan was specifically evaluated altering the flow characteristics of the inflow into rearing bank 13 at Lewis River hatchery in 2022. In 2023, there are no planned activities.

Limitations or Concerns

Specific limitations and concerns for the spring Chinook rearing and release study and precocity monitoring are described in Strategy E and F, respectively.

Data Collection Methods

Metric or Monitoring Indicator	Targets	Field Methods	Analytical Methods	Frequency and Duration
3B.1. Integration Rates (pNOB; Integrated programs only)	Steelhead: 30% fixed mining rate. Coho: 30% integration rate.	Outlined in Sections A2.1, B2.1, C2.1 of this AOP as part of the broodstock collection process.	NA	In general, data on these metrics are collected as part of typical hatchery program operations and will be reported on in the Annual Operating Report, and trends will be evaluated as part of the Adaptive Management process described in Section E. For spring Chinook salmon, after 3 years of implementation of the Spring Chinook Rearing and Release Plan (Strategy A), in-hatchery survival rates, size-at-release, condition factor at
3B.2. Spawning matrices and timing	Steelhead: Depends on spawners available on a given day; typically 2x2. Spring Chinook and coho: pairwise (1x1) mating cross with a backup male.	Outlined in Sections A2.9, B2.9, C2.9 of this AOP as part of the spawning protocols. The number, timing and composition of spawners used, and type of spawning matrix used for each stock will be recorded (e.g., pairwise, pairwise with backup male, factorial, etc.).	NA	

3B.3. Broodstock Fecundity	NA	Late winter steelhead: Record the estimated number of eggs per female. Early coho and spring Chinook: Average fecundity will be estimated from a subsample of females spawned.	For integrated programs (late winter steelhead and late coho), fecundity will be compared between HOR and NOR. Fecundity and spawn timing data are also used to examine risk to phenotypic diversity described in Objective 7.	release, fish health (frequency or rates of disease), and physiological status at the time of release will be compared between treatment groups.
3B.4. Feeding rations and delivery methods	Not Applicable in 2023			
3B.5. Avian predation rate	Not Applicable in 2023			
3B.6. Volitional releases	Steelhead: May 1; 6-week volitional period. Spring Chinook: February 1, October 15, or June 1; 2-week volitional period. Coho: April for 2 to 6-week volitional period.	The start date and time of volitional release (i.e., when screens are pulled) and the start and end date and time of the forced release period will be recorded for each species and pond.	Report on	
3B.7. Water Quality	Not Applicable in 2023			

Key Question 3C. Are adult collection, handling and disposition protocols consistent with HSRG recommendations?

Approach

This key question relates to sampling and disposition protocols applied to adult fish handled at either the Merwin Trap or Lewis River ladder. Specifically, a sample of HOR and NOR adults for each stock will be used to collect size (fork length) and age (scale samples) data using a representative sample of HOR and NOR returns over the entire run. Size and age data will also be collected at traps and during spawning ground surveys, summarized in Strategy A.

Additionally, the disposition for all salmon and late winter steelhead captures at the traps will be documented to ensure that broodstock and upstream transport collection goals consistent with HSRG guidelines are achieved.

Captures at the Merwin and Lewis River ladder will be grouped into one of the following five disposition categories, with the disposition protocols described in Sections A2.7, B2.7, and C2.7:

1. Retained for broodstock
2. Transported upstream
3. Returned to river downstream
4. Euthanized
5. Mortalities

Limitations or Concerns

Several generations of adult returns may be necessary to provide adequate size and age data and analysis to support and justify implementation of specific HSRG recommendations.

Data Collection Methods

Metric or Monitoring Indicator	Targets	Field Methods	Analytical Methods	Frequency and Duration
3C.1. Size and age of returning HOR and NOR adults	Use of size and age at maturity as indicator of phenotypic diversity among HOR, NOR and integrated programs. See Objective 7.	Subsample of fork lengths and scales from trap returns and in-stream carcass surveys for coho and Chinook. Refer to Strategy A for data collection on spawning grounds.	Size and age data should be visualized and compared between HOR and NOR with distribution plots and point estimates with 95% confidence intervals.	Sampling occurs throughout the duration of the run. Daily (Merwin Trap); weekly (Lewis hatchery ladder and stream surveys).
3C.2. Disposition of adult trap captures assigned to surplus	Meet goals for proportion and timing of adult disposition that are consistent with the H&S Plan and AOP.	Refer to Sections A2.7, B2.7, and C2.7.	Disposition of each captured adult will be recorded throughout the complete return period for each of the transport species.	Daily at Merwin Trap Weekly at Lewis hatchery ladder

Key Question 3D. What are the estimated smolt-to-adult returns (SAR's) for each hatchery stock or rearing treatment group?

Approach

Smolt-to-adult Return ratio (SAR) is the number of adults produced out of the number of smolts released. SAR is a key metric to estimate the effects of both freshwater and ocean productivity on the survival of hatchery-produced fish from release to their return to Lewis River traps or to spawn naturally. Harvest estimates from ocean fisheries and terminal (in-river) fisheries should also be incorporated into the SAR calculation to account for adults removed prior. The protocols for calculating SAR for Lewis River Hatchery fish will be described in Strategy I.

SAR is also one of the metrics used to evaluate different rearing strategies at the hatchery facilities, including the spring Chinook rearing and release evaluation (summarized in Strategy E).

Limitations or Concerns

- Tag loss in juveniles.
- Incomplete tag recovery from adult cohorts.
- Time lag in reporting in RMIS.

Data Collection Methods

Metric or Monitoring Indicator	Targets	Field Methods	Analytical Methods	Frequency and Duration
3D. Smolt-to-adult Return ratio (SAR) of all hatchery release groups	Adult returns are adequate to meet adult ocean recruit targets given in section 8.3 of the Settlement Agreement.	Collection of CWTs from fish encountered in 1) Lewis River traps 2) Lewis River subbasin spawning grounds, 3) strays to other basins and 4) harvest by stock	CWT data obtained from RMIS based on release codes. Returns include recaptures from: • Adult harvest in all fisheries • Adult spawners • Adult traps Releases are estimated number of CWT smolts released by release group, corrected for estimated tag loss and post-tagging in-hatchery mortality.	Annual SAR estimate for each brood year and release group

Key Question 3E. Is the fish health monitoring and disease prevention strategy effective at reducing infections and limiting mortalities?

Approach

WDFW's Fish Health Unit is tasked with monitoring population health of all H&S Plan species and operates following standards and objectives outlined in the Salmon Disease Control Policy of the Fisheries Co-Managers of Washington State (WDFW 2006) and State of Washington Fish Health Manual (WDFW 2010). Services include monitoring reported and regulated pathogens in all broodstocks, baseline monitoring throughout the rearing cycle, and direct monitoring for specific disease progression and severity in targeted groups. Common species-specific diagnoses, disease prevention and treatments are described further in Strategy D, *Fish Health Monitoring and Disease Prevention Strategy for Lewis River Hatchery Programs.*

Mortality rates are monitored by life stage by hatchery staff to determine if mortality rates are preventing achieving the production goals.

Fish health monitoring at Lewis River Hatchery Facilities includes

- Routine baseline monitoring
- Directed monitoring (and treatment) in response to any significant loss of fish (>~0.05% loss for consecutive days or exponentially increasing loss pattern) that is suspected to be due to an infectious agent.
- Special monitoring of juveniles for gas bubble trauma, bacterial kidney disease

Limitations or Concerns

The *Fish Health Monitoring and Disease Prevention Strategy for Lewis River Hatchery Programs* is not intended to be comprehensive protocols, but provide the common approaches for managing perennial disease issues.

BKD is endemic in the Lewis River Spring Chinook Salmon population. BKD prevalence and severity is highly variable year-to-year, confounding the ability to draw linkages to spawning and rearing practices. WDFW Hatchery staff and Fish Health staff will continue to discuss tracking BKD expression and prevalence in all life stages to link prevalence of BKD in progeny to results of females contributing to a rearing unit. Evaluation of current BKD screening protocols will be made on a yearly basis in response to patterns in BKD prevalence, disease severity, and rearing mortality among untested groups.

Data Collection Methods

Metric or Monitoring Indicator	Targets	Field Methods	Analytical Methods	Frequency and Duration
3E.1. Infection rates by species and life stage	Ensure the health and productivity of H&S Plan fish	60 adult females of each species are inspected and sampled during spawning All spring Chinook females whose eggs are allocated for February release will be tested for BKD prevalence Rearing juveniles are monitored and examined routinely	Fish health monitoring results are reported and maintained by WDFW and pathogen histories are available at any time upon request. The subsample of juvenile spring Chinook evaluated for BKD at transfer and release will be analyzed to report prevalence (% positive), DNA load, prevalence of severe infections, and prevalence of gross pathology.	Baseline monitoring occurs throughout broodstock collection, spawning, and incubation as described in Strategy D. Directed monitoring occurs as needed.
3E.2. Mortality rates by species and life-stage	Ensure mortality rates are not adversely affecting production targets	If needed, medication will be provided by the veterinarian of record	Results are generally reported as presence/absence, mortality range (normal, increased, epizootic) and % loss (mortality) per day for a given rearing unit. In-hatchery survival reported as survival (S = total count – mortalities) from egg to release for each species and release group.	Special monitoring of juvenile spring Chinook for GBT will occur with an initial baseline examination in June, followed by weekly examinations in August Special monitoring of juvenile spring Chinook for BKD will occur at the times of transfer and release.

Key Question 3F. Do hatcheries incorporate new scientific advances to improve fish culture effectiveness and efficiency?

Approach

Periodic evaluations or reviews of the existing hatchery programs should occur to incorporate advances in the best available science that improve operational efficiency and benefit fish health at each of the three facilities. The focus of these periodic reviews will be the implementation of actions that help the hatchery programs achieve the goals of the H&S Plan (i.e., reintroduction outcome goal). The role of hatcheries is a critical component in meeting H&S goals and operation of the facilities should remain adaptable to the needs of the H&S program, development and transitioning into integrated hatchery programs and the needs of harvest management. Finally, operations should be reviewed of potential risks posed by hatchery management on the long-term viability of naturally producing stocks.

The ATS will conduct periodic reviews and report any recommendations to the ACC or hatchery managers for for modifying the hatchery program's activities to align with the best available science on fish health, behavior, and operational efficiency.

Limitations or Concerns

Direct comparisons between Lewis River Hatcheries operations and those described from other hatcheries or literature may not be possible. Inferences and assumptions should be identified clearly at the time of review.

Data Collection Methods

Metric or Monitoring Indicator	Targets	Field Methods	Analytical Methods	Frequency and Duration
3F. Periodic review of hatchery operations relative to current literature	Implementation of hatchery activities that are based on best available science on maintaining fish health, sustaining harvest, and minimizing genetic or ecological risks.	N/A	The AOR and current hatchery methods will be reviewed and compared to published literature and methods from other hatcheries. The ATS will identify known areas of concern for hatchery operations efficiency or topics of recent advances in hatchery science. Potential outcomes include identification of a data gap, next steps toward making changes in implementation, or recommending an immediate implementation change if evidence supports it.	Hatchery operations will be reviewed every three years. The discussion of the approach to the review will occur in June of the review year, and recommendations will be completed by October of the review year for incorporation into the following AOP for the following year.

Objective 4.0: Adopt strategies that limit potential post-release ecological interactions between hatchery and NOR listed species

The purpose of Objective 4.0 is to limit ecological interactions (predation, competition, residualism and pathogen transmission) between hatchery released juveniles on natural origin listed species. Interactions between hatchery released juveniles and ESA listed species cannot be observed directly. Therefore, this objective relies on "take surrogates" as described by NOAA Fisheries (NMFS 2017) to reduce the potential of adverse interactions between hatchery and natural-origin salmon and steelhead. Each key question provided under this objective relates directly to each take surrogate described by NOAA Fisheries.

KEY QUESTION 4A: DO CURRENT HATCHERY RELEASES RESULT IN SPATIAL AND TEMPORAL OVERLAP BETWEEN HOR AND NOR JUVENILES?

APPROACH

This question will be answered by comparing hatchery release timing and location to the spatial and temporal distribution of natural origin stocks in the NF Lewis River. Potential overlaps will be identified for all hatchery releases and all life stages of natural origin stocks (presence of juveniles in an area can be inferred from known spawning distributions).

Hatchery data on timing and locations for all hatchery releases will be compared to empirical information derived from field activities where natural-origin juveniles and spawners are encountered. Activities that help to characterize the spatial and temporal distribution natural-origin fish in the North Fork Lewis River and its tributaries include adult fish trapping, spawner surveys, juvenile screw trapping, and potentially other presence/absence surveys such as juvenile seining activities downstream of Merwin Dam. Potential or observed overlaps between hatchery releases and NOR stocks will be identified.

LIMITATIONS OR CONCERNS

Overlap in space or time is used as a "take surrogate" for ESA-listed natural-origin fish; it is not a direct measure of take and therefore cannot quantify or estimate actual take related to large hatchery releases in the North Fork Lewis River.

DATA COLLECTION METHODS

Metric or Monitoring Indicator	Targets	Field Methods	Analytical Methods	Frequency and Duration
4A.1. Release locations of hatchery smolts relative in-river spawning locations.	Mitigate potential spatial overlap between hatchery-released juveniles and NOR stocks	Release location(s) of each hatchery pond; multiple locations to include relative portion of total release (e.g., forced release group transferred to Pekins Ferry) Spawner distribution monitored by redd and carcass (spawner) surveys, as described in Strategy B.	compare hatchery release locations to distribution of natural-origin fish inferred from spawning surveys and screw trap captures.	Annually
4A.2. Release timing of hatchery reared smolts relative to presence of NOR	Mitigate potential temporal overlap between hatchery-released juveniles and NOR stocks		Compare hatchery juvenile release dates and encounters of hatchery origin fish with timing of natural-origin fish in the datasets for adult trap entry, spawner surveys, and screw trap encounters.	Annually

KEY QUESTION 4B: DOES THE MIGRATION RATE OF HOR JUVENILES RESULT IN OVERLAP WITH NOR JUVENILES OR SPAWNING ADULTS?

APPROACH

Monitoring migration rate is an indirect method for monitoring post-release behavior of hatchery smolts to infer their potential impacts on NOR species. The timing of hatchery-origin juvenile outmigration after release will be derived from screw trapping in the lower North Fork Lewis River as described in Strategy C. The beginning, peak and end of the volitional release period from the hatchery will be compared to the beginning, peak and end of encounters in the screw traps. The range of migration rates observed and average or median migration rate of hatchery-released smolts will be reported.

LIMITATIONS OR CONCERNS

The migration window of hatchery fish is typically very short (i.e., a few days) and if the traps are not in operation due to mechanical failure, an entire migration window may be missed.

Volitional release periods can last for just a few days if >90% of a group leave quickly, or up to 6 weeks if many fish do not volitionally leave the ponds. The specific timing of when hatchery fish enter the river during a volitional release period may not be precisely known, so travel time to screw traps may be reported as a range based on a range of potential river entry dates. PIT tagging of hatchery fish and use of a fixed PIT tag antenna at the hatchery could be used to determine river entrance timing and inform assumptions about average migration rate.

Screw trapping is planned to occur from March 1 through June 30 (but may be adjusted in season depending on flows); migration rates will not be available for fish emigrating outside of that period, which likely includes spring Chinook salmon that are released in June (as fry), October (as fall yearlings) or February (as spring yearlings).

DATA COLLECTION METHODS

Metric or Monitoring Indicator	Targets	Field Methods	Analytical Methods	Frequency and Duration
4B. Average migration rate and range of migration rates of hatchery released smolts	Rapid outmigration to minimize the period of time that hatchery-origin juveniles may encounter natural-origin fish.	Refer to Sections A3.7, B3.7, and C3.7 for hatchery release timing. Refer to Strategy C for screw trapping methods.	Derive minimum, maximum, and mean or median migration rates from the difference between release date and screw trap capture dates.	Average migration rates will be reported annually for species (or release groups) in context of trends across years, as described in Section E of this document, Adaptive Management.

KEY QUESTION 4C: ARE THE NUMBER OF HATCHERY-RELEASED JUVENILES EQUAL TO OR LESS THAN PRODUCTION TARGETS?

APPROACH

This question will be answered by reviewing hatchery release records documenting the total number of smolts released for each species. The number of fish planted into hatchery rearing vessels is determined by volumetric measurement. The number of fish released is derived from the number planted less any mortalities observed over the rearing period. This information will be compared to hatchery production targets contained in the H&S Plan and Settlement Agreement to ensure that total release number should not exceed 105 percent of production targets. Release targets are shown in Table D-1.

Table D - 1. Total juvenile hatchery production targets for the North Fork Lewis River hatchery complex

Species	Number of fish	Maximum release number
Spring Chinook	1,350,000	1,417,500
Early Winter Steelhead	100,000	105,000
Late Winter Steelhead*	50,000	60,000
Summer Steelhead	175,000	183,500
Coho Salmon	2,000,000	2,100,000

* As specified in Section A3.6 late winter steelhead releases are 50,000 ± 20%

Hatchery release numbers are reviewed annually and any exceedances of target release numbers shall be noted in the Annual Operations Report. Any exceedances will be discussed within the ATS to determine the reason(s) for exceeding the target release number for any species and if necessary, adaptive management actions shall be incorporated into the Annual Operating Plan.

LIMITATIONS OR CONCERNS

Number at the time of release is an estimate based on original stocked number less observed mortalities, but may not account for unobserved mortalities (i.e. due to predation).

DATA COLLECTION METHODS

Metric or Monitoring Indicator	Targets	Field Methods	Analytical Methods	Frequency and Duration
4C. Number of total smolts released by species and period	≤ 105% of target release number.	Rearing ponds stocked with number determined volumetrically. Mortalities subtracted over rearing period.	Compare number released to target.	Report annually for each release group.

Key Question 4D: Are the sizes (length and weight) of released hatchery juveniles equal to or less than program targets?

Approach

Batch weights are collected monthly to compare actual fish size to programmed size targets by month. Length and weight are measured on a representative subsample of fish from each rearing pond at the time of release.

Limitations and Concerns

Collecting a representative subsample from large rearing ponds is challenging, especially for groups with more variability in size (e.g. spring Chinook and late winter steelhead).

Data Collection Methods

Metric or Monitoring Indicator	Targets	Field Methods	Analytical Methods	Frequency and Duration
4D. Mean and coefficient of variation (CV) in fork length and weight of smolts released by species and release group.	Steelhead: > 180 and < 220 mm. Spring Chinook: 8-12 fpp for October or February releases, 80 fpp for June release (see Strategy E). Coho: 16 fpp.	Batch weights collected monthly. Length measured from a representative subsample (e.g. 100 fish per release group) at the time of release.	Calculate fish per pound (fpp) from weights to compare to targets. Calculate mean and CV of fork length, weight.	Monthly batch weights. Average fork length at the time of release.

KEY QUESTION 4E: WHAT IS THE PRECOCITY RATE FOR HATCHERY JUVENILES BY RELEASE GROUP PRIOR TO SCHEDULED RELEASES?

APPROACH

The Aquatic Technical Subcommittee has discussed potential ways to quantify residualism and determined that measuring precocity in hatchery-reared spring Chinook salmon should be a priority. Precocious maturation refers to male fish that mature toward the end of either their first year (microjacks) or second year (minijacks), as measured from the date of fertilization.

Age 1 Precocity:

At the time of spawning (late-August through October for spring Chinook), fully mature males can be easily identified by their darkened body color and rounded belly and can be identified with a non-lethal screening. Their testes grow to fill most of their abdominal cavity, and milt can be expressed by gently squeezing the fish's abdomen in an anterior to posterior motion. Non-lethal screening may be used to evaluate whether specific rearing or feeding strategies contribute to overall precocity in spring Chinook

Age 2 Precocity:

The two most thoroughly validated indexes of maturity for spring Chinook maturing at 2 years of age require lethal sampling: 11-ketotestosterone (11-kt) levels in the blood plasma and testis weight compared to total body size (Refer to Strategy F for detailed methods). With either method, the distribution of the data tends to be bi-modal and precocious males are identified as individuals with 11-kt or GSI levels above a derived threshold level that separates the two modes.

LIMITATIONS OR CONCERNS

Non-lethal visual screening for precocious males a metric that can be objective, and for spring Chinook salmon in particular, inaccurate for fish released in February, 6 to 7 months prior to typical spawning times. The physiological processes that lead to maturity in spring Chinook may not have progressed enough in February to show detectable differences in physiological indicators; differences between immature and mature groups are more easily discerned later in March and April.

DATA COLLECTION METHODS

Metric or Monitoring Indicator	Targets	Field Methods	Analytical Methods	Frequency and Duration
4E. Precocity Rate	Minimize precocity rates to reduce residualism and interactions between mature juveniles released at the same time as natural spawning in the river. Precocity rate in wild spring Chinook likely to be less than 5% of males.	Non-lethal visual screening for October spring Chinook release group. Periodic lethal 11kt or GSI sampling for February spring Chinook release groups. Refer to Strategy F for details.	Calculate precocity rate as number of precocious males out of total number of males. Refer to Strategy F for details.	Non-lethal visual screening for October spring Chinook release group carried out annually as part of routine morphology monitoring at the time of release. Lethal 11kt or GSI sampling for February spring Chinook release group carried out periodically.

2.3 Abundance Monitoring Objectives

Objective 5.0: Estimate spawner abundance of late winter steelhead, Coho, chum and Chinook downstream of Merwin Dam

The purpose of Objective 5.0 is to collect unbiased, long-term, abundance, distribution and cohort trend data for natural origin adult spawners (Chinook, Coho, chum salmon and late winter steelhead) downstream of Merwin Dam. This includes recovery of CWT tags from salmon carcasses to inform harvest management, and collection of mark and tag status information (i.e., adipose clips and CWT presence) to inform calculation of pHOS and PNI.

A secondary purpose of this objective is to provide data for Objective 22 of the AMEP which describes combining estimates from downstream of Merwin Dam with transport and monitoring data for areas upstream of Swift Dam to evaluate spawning distribution and develop population-level estimates of spawner abundance and productivity for Chinook, Coho and late winter steelhead.

Key Question 5A: Are estimates of spawner abundance unbiased and meeting precision targets?

Approach

For spring Chinook and fall Chinook, which are present in large numbers, spawner abundance is estimated using carcass mark-recapture data in an open-population Jolly-Seber "super population" model, described in Strategy A. When the assumptions of the JS model are met, it produces unbiased estimates of escapement with known levels of precision. Those assumptions are that carcasses have equal catchability, equal survival, no tag loss and readability upon recovery, and instantaneous sampling. The assumptions will be evaluated annually following methods described in Strategy A.

For steelhead, abundance is estimated using redd surveys in the mainstem. If river conditions prevent surveys, area-under-the-curve methods will be used and however this method relies on a number of untested assumptions, and bias in the estimates can not be evaluated.

For coho, abundance in the mainstem North Fork Lewis River is estimated using carcass mark-recapture data in a Jolly-Seber mark-recapture analysis, as described in Strategy A. Uncertainty in mainstem spawner abundance will be reported as the range in potential abundance within associated confidence intervals. Abundance in the tributaries to the North Fork Lewis River are estimated from carcass and redd data collected using a Generalized Random Tessellation Stratified (GRTS) method to predetermine a spatially-balanced set of survey reaches, and data are entered in to a Bayesian multivariate state-space model used for coho throughout the Lower Columbia ESU. Parameters are reported as posterior medians with associated 95% credible intervals.

Chum abundance is too low to evaluate bias in estimates at this time.

Limitations or Concerns

The estimators may quantify all possible sources of variation better for some species but not others. See Key Question 5B for limitations and concerns for estimating abundance with spawner surveys.

Data Collection Methods

Metric or Monitoring Indicator	Targets	Field Methods	Analytical Methods	Frequency and Duration
5A. Mark-recapture modeling assumptions are evaluated to determine if they are met	Generate an unbiased estimate of abundance and composition downstream of Merwin Dam with a CV of 15% or less, on average	Chinook: Carcass mark-recapture. Steelhead: redd survey. Coho: Carcass mark-recapture in mainstem, GRTS and Bayesian multivariate state-space model in tributaries. Refer to Strategy A.	Annual evaluation of assumptions made for modeled estimates of abundance. Refer to Strategy A. Chinook: Open-population Jolly-Seber “super population” analysis to generate abundance estimates by stock, origin, sex and age. Steelhead: redd counts multiplied by fish per redd; AUC if necessary. Coho: Jolly-Seber mark recapture model for mainstem, and Bayesian multivariate state-space model for tributaries, to generate abundance estimates by origin.	Surveys and analyses completed annually.

Key Question 5B: Are annual estimates of natural-origin spawner abundance increasing, decreasing or stable?

Approach

For spring Chinook and fall Chinook, which are present in large numbers, spawner abundance is estimated using carcass mark-recapture data in an open-population Jolly-Seber "super population" model, described in Strategy A.

For steelhead, abundance is estimated using redd surveys. If river conditions prevent surveys, area-under-the-curve methods may be used.

For coho, abundance in the mainstem North Fork Lewis River is estimated using carcass mark-recapture data in a Jolly-Seber mark-recapture analysis, as described in Strategy A. Abundance in the tributaries to the North Fork Lewis River are estimated from carcass and redd data collected using a Generalized Random Tessellation Stratified (GRTS) method to predetermine a spatially-balanced set of survey reaches, and data are entered in to a Bayesian multivariate state-space model used for coho throughout the Lower Columbia ESU.

Chum abundance is currently low; carcasses that are encountered in Chinook and Coho surveys are enumerated.

Limitations or Concerns

- For Chinook and Coho, violation of model assumptions may create unknown levels of bias in estimates.
- Assignment of redds to species is complicated by overlapping distributions of relatively large number of fall Chinook with Coho and spring Chinook.
-
- Steelhead abundance estimates are often limited by river conditions in the spring that cause high turbidity or flows and limit surveyors' ability to identify redds.
- Steelhead are not currently surveyed across their entire spatial distribution in tributaries, thus generated estimates of abundance are likely biased low.

DATA COLLECTION METHODS

Metric or Monitoring Indicator	Targets	Field Methods	Analytical Methods	Frequency and Duration
5B. Total spawner abundance estimates by species, sex and origin, and age.	Annual trends are stable or increasing.	Carcass and redd surveys. Refer to Strategy A.	Chinook: Open-population Jolly-Seber “super population” analysis. Coho: Jolly-Seber mark recapture model for mainstem, and Bayesian multivariate state-space model for tributaries. Steelhead: redd surveys	Surveys and analyses completed annually. Results reported annually by species in context of trends across years, as described in Section E of this document, Adaptive Management.

Objective 5.1: Determine the spatial and temporal distribution of spawning late winter steelhead, coho, chum and Chinook downstream of Merwin Dam

KEY QUESTION 5C: ARE ANNUAL TRENDS IN SPATIAL AND TEMPORAL SPAWNING DISTRIBUTION INCREASING, DECREASING OR STABLE?

APPROACH

For steelhead, spatial and temporal distribution of spawners is estimated using redd surveys.

For Coho salmon in the mainstem North Fork Lewis River, spatial and temporal distribution is determined from live counts and carcasses. Redd surveys are not a reliable estimator in the mainstem due to superimposition with Chinook redds. In tributaries, redd counts and live counts will used to determine Coho distribution.

For Chinook, redd counts and live fish will be enumerated in the mainstem North Fork Lewis River. Standard metrics (e.g. median and standard deviation) will be used to describe abundance. Temporal distribution will be described by calculating median spawn date using weekly derived estimates of abundance using the Jolly-Seber model, and a cumulative distribution plot of abundance will be generated.

Chum salmon are present in low numbers; changes in spatial or temporal distribution will not be evaluated.

Spawner survey methods are described in Strategy A. Redd locations are recorded using GPS and data are time stamped. The number of redds or spawners per mile and per period will be calculated and compared across years to identify shifts in spatial or temporal distribution.

LIMITATIONS OR CONCERNS

Steelhead redd surveys are not performed if visibility is low or spill from Merwin Dam has been initiated. Poor visibility can also prevent identification of coho redds or live fish. Discerning Chinook and coho redds is challenging and substantial redd superimposition occurs between the species.

DATA COLLECTION METHODS

Metric or Monitoring Indicator	Targets	Field Methods	Analytical Methods	Frequency and Duration
5C. Estimate of redds, carcasses or live spawners by reach and time period	Proportion of redds per reach is stable or increasing	Spawner surveys described in Strategy A with spatial and temporal data recorded using GPS as described in Strategy B.	Median spawner number per reach and median spawn date per week to be calculated.	Surveys and analyses completed annually. Results reported annually by species in context of trends across years, as described in Section E of this document, Adaptive Management.

Objective 6.0: Estimate juvenile outmigrant abundance for late winter steelhead, coho, and Chinook downstream of Merwin Dam

The purpose of Objective 6.0 is to estimate the abundance of juvenile outmigrants by species and origin for the North Fork Lewis River downstream of Merwin Dam. Capture and sample juvenile fish to note morphological differences between HOR and NOR smolts, as well as other juvenile non-migrants (i.e., fry and parr).

KEY QUESTION 6A: ARE ESTIMATES OF NOR JUVENILE OUTMIGRANT ABUNDANCE UNBIASED AND MEETING PRECISION TARGETS?

APPROACH

Outmigration abundance is estimated through the use of rotary screw traps in the lower North Fork Lewis River. Trap operation and analysis protocols are described in Strategy C.

These traps capture a portion of the total number of juveniles passing the trap location. Trap efficiency is estimated using a mark-recapture approach in which trapped fish are marked, released upstream of the trap, and some percentage are recaptured. The estimated capture efficiency of the trap is used to derive estimates of abundance for juveniles passing the trap. Approaches for estimating trap efficiency and abundance by species, life-stage, and by week are an ongoing topic of ATS discussion.

Estimates of abundance are only useful if they are unbiased (i.e., accurate) and relatively precise. A recovery monitoring goal for juvenile salmon migrants is to have data with a CV on average of 15 percent or less, and steelhead migrant data with a CV on average of 30 percent or less. Assumptions of the estimator must be met, and variance must be estimated in an unbiased manner. Because it is not possible to estimate the level of bias, study designs should strive to meet all the assumptions of the estimator(s) to the extent practical. These assumptions are summarized in Strategy C, briefly:

1. The population is closed to immigration, emigration, births, and deaths.
2. No mark or tag loss (from fish marked for estimating trap efficiency).
3. No mark-related mortality.
4. All fish have equal probability of being caught and marked in the first event.
5. Marked fish mix completely with unmarked fish between sampling events.

Limitations or Concerns

Juvenile outmigrant trapping can be complicated and requires development of clear study designs or protocols, project review and adaptive management to be successful. Estimates of abundance can be biased if the assumptions of the mark-recapture estimator are not met (e.g., equal survival and capture probability among marked and unmarked groups). Testing assumptions and describing how mark-recapture assumptions are being met is critical for developing unbiased estimates. The ability to specifically test all assumptions for the North Fork Lewis River smolt trapping project may be limited and should utilize results and recommendations from other juvenile migrant studies when applicable (e.g., tag retention studies).

In past years, it has been difficult to mark, and subsequently recapture enough juveniles to generate unbiased estimates of abundance. To reduce bias in the estimates, the ATS may evaluate whether the following methods are practical for implementation:

- Pooling of data to increase the sample size and power to improve the precision of estimates, however, this method can produce biased estimates and is not favored by WDFW.
- The use of marked hatchery releases as a surrogate for mark-recapture in smolt traps. However, capture probability may not be the same between HOR and NOR smolts due to differences in size or behavior, violating assumption number 4 of the estimator.
- Increasing the mark rate at the screw trap to increase the number of total marks available for recapture.
- Use of Cedar Creek screw trap captures to increase the number of NOR marks available for recapture.
- In theory, the use of juveniles captures from other sources (e.g., the Swift FSC, Cedar Creek, additional upstream trap) may provide the ability to increase the number of marked smolts available for capture to estimate capture efficiency. However, there are a number of considerations to assess when determining whether these sources are feasible:
    - Can marked fish can be released safely for both staff and marked fish?
    - What is the number of marked fish needed to make meaningful inferences regarding capture efficiency?
    - Are marked release groups representative of their NOR counterparts (e.g., size, behavior and migration timing)?
    - Does the use of alternative sources impact other required monitoring needs?
    - Are there any ESA regulatory restrictions on trapping or releasing additional marks into the NF Lewis River?
    - Is the use of juveniles from alternative sources practical from an operations, safety and regulatory obligations in the Settlement Agreement?

Behavior or migration rates between the traps and the mouth of the Lewis River cannot be described by the screw trap catches alone. More work is needed to quantify the number of fish residing (e.g., nonmigrants) upstream of the trap post-release to estimate residualism rates.

Trapping is inherently difficult in larger rivers due to debris load, variable flows (including spill) and mechanical failures – most of which are unavoidable consequences of operating floating traps in large rivers. Trap operation in the North Fork Lewis River may be limited by high streamflow conditions in late winter and spring as well as lower flow periods in the summer.

Data Collection Methods

Metric or Monitoring Indicator	Targets	Field Methods	Analytical Methods	Frequency and Duration
6A. Precision and accuracy of abundance estimates.	VSP precision and accuracy targets are met: CV < 15% (salmon) and < 30% (steelhead)	Rotary screw traps will be used to capture a portion of the total number of juveniles passing the trap location. Mark-recapture of those fish will be used to test trap efficiency. Refer to Strategy C	Quantify whether assumptions of the estimator are met. See Strategy C for details.	See Strategy C. Annual juvenile outmigrant sampling. Traps checked every morning, 7 days per week, from March 1 through June 30. Trapping dates may be adjusted depending on river flows. Additional daily trap checks may be warranted during peak migration.

Key Question 6B: Is the abundance of NOR juvenile outmigrants by species and outmigration year increasing, decreasing, or stable?

Approach

Outmigration abundance is estimated through the use of rotary screw traps in the lower North Fork Lewis River. Abundance estimates are derived from trap efficiencies. Trap operation and analysis protocols are described in Strategy C. Trapped salmonids will be identified by species, life-stage (based on size), and origin (based on hatchery marking). The assignment of fish to these groups will follow a decision tree developed and used by WDFW in other trapping operations, included in Strategy C.

In addition to reporting abundance estimates, a power analysis will be conducted for each juvenile migrant population being monitored to determine the power of the data to detect a significant change in abundance and recommended sample sizes to be able to detect change over time.

Annual estimates of NOR juvenile outmigrant abundance will be reported in the Annual Operating Report for a given cohort, in some cases to include numbers of fish from the same cohort that out-migrate at different lift-stages. Data will be reported in context with previous years of data to make a determination if numbers are increasing, decreasing, or stable. The approach for annual reporting of trends and ACC review of the data are described in Section E of this document, Adaptive Management.

Limitations or Concerns

Generating unbiased estimates of juvenile NOR outmigrant abundance is limited by low numbers of NOR captures in rotary screw traps. Trapping is limited to the major spring outmigration period for Chinook, coho, and steelhead (March 1 through June 30, trapping dates may be adjusted depending on river flows); fish migrating outside of this window will not be sampled.

Data Collection Methods

Metric or Monitoring Indicator	Targets	Field Methods	Analytical Methods	Frequency and Duration
6B.1. Trend in total NOR outmigrants by species and cohort.	Annual trends are stable or increasing.	Screw trapping to capture outmigrants. Refer to Strategy C.	Derive estimates of abundance based on trap efficiency. Refer to Strategy C.	Juvenile outmigrant abundance and morphology sampling carried out annually. Traps checked every morning, 7 days per week, from March 1 through June 30. Trapping dates may be adjusted depending on river flows.

Key Question 6C: What are the morphological characteristics of outmigrating NOR juveniles relative to their conspecific HOR juveniles?

Approach

Outmigration of NOR juveniles is monitored through the use of rotary screw traps in the lower North Fork Lewis River. Trap operation and analysis protocols are described in Strategy C. Morphological information is collected from fish captured in rotary screw traps. Fish are identified as natural origin by lack of hatchery marks, and categorized as fry, parr, transitional, subyearling smolt, yearling smolt based on size and external coloration following the decision tree included in Strategy C. Size and smolt development are similarly collected at the time of release for hatchery-reared fish as described in sections A 3.7, B 3.7, and C 3.7 of this AOP.

Limitations or Concerns

Hatchery-origin juveniles for this program to optimize high rates of smoltification, post-release survival and faster outmigration compared to natural-origin fish. No changes have been proposed for changing hatchery-origin size or release targets relative to natural-origin fish.

Similar to estimates of abundance, unbiased estimates of natural-origin outmigrant size and smoltification will be limited by low numbers captured in the screw traps and the timing of trapping operations between March 1 and June 30.

Data Collection Methods

Metric or Monitoring Indicator	Targets	Field Methods	Analytical Methods	Frequency and Duration
6C. Comparison of fish size, life-stage and age-class between NOR and HOR juveniles.	No relevant target for natural-origin outmigrants; hatchery-origin outmigrant size based on optimizing post-release performance.	Refer to Sections A 3.7, B 3.7 and C 3.7 of this document for data collection at the time of release. Up to 10 fry per day and 50 subyearling/ transitional/ parr per day per category to be sampled for fork length during screw trapping. Refer to Strategy C.	Average size to be calculated from representative subsample at hatchery release and in screw trapping. The component of those subsampled made up by different life-stages and age classes should also be reported.	Juvenile outmigrant abundance and morphology sampling carried out annually. Traps checked every morning, 7 days per week, from March 1 through June 30. Additional daily trap checks may be warranted during peak migration. Refer to Strategy C.

2.4 Risk Assessment Objectives

Objective 7.0: Monitor the extent of genetic risks associated with integrated and segregated hatchery programs on naturally spawning listed populations in the North Fork Lewis River

The purpose of Objective 7 is to develop and implement a comprehensive genetic monitoring plan to assess the potential threats that hatchery programs may pose to naturally-spawning anadromous salmon and steelhead in the Lewis River. The monitoring of genetic risks and minimization of adverse effects is a requirement of the Settlement Agreement. This objective provides guidance on assessing (1) the genetic risks posed by the hatchery production programs and (2) whether the H&S Program is achieving or capable of achieving 'genetic viability' of reintroduced stocks.

The initial goal of this section of the AOP is to identify the baseline levels of genetic diversity and domestication as the starting point for observing changes and trends in the future. Focus species would include winter steelhead, coho salmon, spring Chinook, fall (tule) Chinook, and late-fall (bright) Chinook salmon. Five biologically-relevant populations existing within the designated boundaries of NF Lewis recovery populations (LCFRB 2010) including 1) the segregated hatchery programs and natural spawners in 2) the lower mainstem NF Lewis River below Merwin Dam, 3) tributaries to the lower NF Lewis River, 4) the upper mainstem NF Lewis River above Swift Dam, and 5) tributaries to the upper NF Lewis River.

The detailed approach and methods for monitoring genetic risk (Key Questions 7A and 7B) are described in Strategy G. Genetic baselines would be developed for the segregated hatchery populations and naturally-spawning/integrated hatchery programs. Tissue samples for genetic analysis will be collected from all adults used for broodstock, adults encountered in spawning surveys, and juveniles encountered in smolt traps or Swift Reservoir Floating Surface Collector.

The metrics used for assessing domestication (Key Question 7C) are described in Objective 8.

The metrics used for assessing phenotypic diversity (Key Question 7D) are components of Objectives 3, 4, and 6.

The information addressing each key question under this objective are summarized in the following sections.

LIMITATIONS OR CONCERNS

- Given the long history of hatchery production in the Lewis River basin along with the biological and environmental complexities that govern the impacts of hatchery programs and their progeny on wild populations, it is unlikely that the metrics generated from this plan can definitively answer the four key questions identified in Objective 7 (i.e., have hatchery programs affected the diversity of natural-origin populations?). However, in combination, these metrics will assess the genetic status of Lewis River populations, qualitatively assess the impacts of current hatchery programs on natural-origin populations, and help guide future hatchery operations that promote long-term genetic viability.

- The correct interpretation of results of genetic tests relies heavily on tissue collections that are representative of the intended populations or groups. Uncertainty of the genetic tests is dependent on the number of samples collected and successfully processed.
- Among- and within-population genetic variation may change due to factors other than hatchery production, including gene flow (straying) from other populations and natural selection. The SNP markers intended to be used here may, with adequate baseline data, allow detection of gene flow from outside into the NF Lewis River.
- Many of the SNP markers are assumed to be neutral (i.e., are not linked to traits under selection). Violation of this assumption would lead to incorrect interpretation of the data. Genomic methods are available and can be used to look for regions of the genome that may be under selection and associated with environmental variables. However, these methods are prohibitively expensive and are not currently part of routine hatchery monitoring.
- Genetic methods, technology, and markers continue to evolve and thus may substantially change between timesteps. In order to make valid comparisons among timesteps, the same marker panels must be used for all samples. Any substantial changes in method, technology, or markers may necessitate the re-processing of samples from earlier timesteps.
- The monitoring plan lacks monitoring of adaptive genetic diversity that uses genetic techniques. Some markers linked to adaptive diversity are known (e.g., Chinook/steelhead GREB markers, Chinook Y haplotype markers [age at maturity in males], Prince et al. 2017, Hess et al. 2016, McKinney et al. 2020) and could be used in monitoring efforts.
- Biological-based metrics of phenotypic diversity likely vary due to a combination of genetic and environmental factors and it may not be possible to determine whether such changes are good or bad for the population in terms of survival and persistence.
- Evaluating genetic- and biological-based metrics together provides the most comprehensive means of evaluating hatchery programs and the risks to natural spawning populations they affect. However, there is no consensus on how these metrics should influence adaptive management decisions.
- It is expected that Lewis River populations of salmon and steelhead will remain in the re-colonization phase well past the next rewrite of the H&S plan (scheduled in 2025) and hatchery supplementation will likely continue. However, Lewis River hatchery programs are scheduled for re-evaluation in the coming years through the development of “transition plans”. Changes to broodstock management may subsequently impact genetic risks to natural-origin populations. Thus, this plan may need to be updated accordingly.

KEY QUESTION 7A: HAVE THE LEWIS RIVER HATCHERY PROGRAMS IMPACTED THE AMONG-POPULATION DIVERSITY OF NATURALLY SPAWNING POPULATIONS?

APPROACH

Among-population diversity is measured as relative genetic differences among biological populations. Hatchery production can reduce among-population diversity when hatchery fish successfully spawn with natural-origin adults from biological populations other than those used as broodstock. Reductions of among-population diversity may reduce the long-term viability of the metapopulation through genetic homogenization and outbreeding depression.

DATA COLLECTION METHODS

Metric or Monitoring Indicator	Targets	Field Methods	Analytical Methods	Frequency and Duration
7A. Pariwise genetic distance (F_{st}), combined with dendograms, multi-variate clustering analyses.	See Strategy G.	See Strategy G.	See Strategy G.	Collect tissue samples annually. Analyses of genetic distance at least every third generation.

Key Question 7B: Have the Lewis River hatchery programs impacted the within-population diversity of naturally spawning populations?

Approach

Within-population diversity describes the amount of genetic diversity within a biological population and is important for the long-term resilience of the population. Reduced within-population diversity is an indication of inbreeding (i.e., increased allelic identity by descent), which may lead to inbreeding depression and reduced long-term viability. Hatchery production increases the risks of reducing within-population diversity of natural-origin populations because hatcheries spawn only a subset (sometimes a very small subset) of the entire population which is then amplified via the increased survival afforded by hatchery rearing. Segregated programs that use only hatchery-produced fish for broodstock are especially susceptible to reductions in within-population diversity.

Data Collection Methods

Metric or Monitoring Indicator	Targets	Field Methods	Analytical Methods	Frequency and Duration
7B.1. Effect population size (Ne) 7B.2. Effective number of breeders (Nb) 7B.3. Inbreeding coefficient (FIS) 7B.4. Average heterozygosity 7B.6. Allele frequencies 7B.7. Linkage Disequilibrium (LD)	See Strategy G.	See Strategy G.	See Strategy G.	See Strategy G.

Key Question 7C: Have the Lewis River hatchery programs increased the risk of domestication for naturally spawning populations?

Approach

Domestication reduces the long-term fitness of populations through the proliferation of alleles which improve performance in domestic settings (i.e., hatcheries) while reducing performance in natural settings (domestication selection). However, there are currently no genetic techniques (e.g., domestication genes or markers) to assess the level of domestication within populations. Therefore, the following metrics based on biological data will be used to assess the potential for domestication of populations that spawn naturally.

- pHOS is the proportion of naturally-spawning adults that are of hatchery origin for a given population and year. An index of gene flow between a hatchery population and its companion natural population.
- pNOB is the proportion of hatchery broodstock composed of natural-origin adults each year. An index of gene flow between hatchery and natural-origin fish within the hatchery.
- PNI describes the collective effects of pHOS and pNOB; describes potential for interbreeding in both hatchery and natural components of the population.
- PEHC estimates interbreeding between hatchery and natural-origin spawners based on genetic parentage analysis of offspring.

Data Collection Methods

Metric or Monitoring Indicator	Targets	Field Methods	Analytical Methods	Frequency and Duration
7C.1. Proportion hatchery-origin spawners (pHOS)	Specific HSRG-recommended targets apply, see Objective 8	See Objective 8	See Objective 8	Calculated annually
7C.2. Proportion natural-origin brood (pNOB)	A component of PNI target. Does not apply to segregated programs.	See Objective 8	See Objective 8	Calculated annually
7C.3. Proportion natural influence (PNI)	Specific HSRG-recommended targets apply, see Objective 8	See Objective 8	See Objective 8	Calculated annually

7C.4 Proportion Effective Hatchery Contribution (PEHC)	No specific targets recommended. Program should avoid increase in PEHC over time. Not relevant for integrated programs.	See Objective 8	See Objective 8	Calculated annually

KEY QUESTION 7D: HAVE THE LEWIS RIVER HATCHERY PROGRAMS IMPACTED THE PHENOTYPIC DIVERSITY OF NATURALLY SPAWNING POPULATIONS?

APPROACH

A potential result of declining genetic diversity and increasing domestication may include observable changes in phenotypic traits of naturally spawning populations. These traits may have some genetic component; however, there are currently few genetic markers with known allelic associations with phenotype. Therefore, the following metrics based on fitness traits will be used to assess phenotypic diversity.

DATA COLLECTION METHODS

Metric or Monitoring Indicator	Targets	Field Methods	Analytical Methods	Frequency and Duration
7D.1 Timing of adult return, spawning and juvenile outmigration.	See Key Questions 3A and 3B for adults. See Key Question 4B for juveniles.	See Key Questions 3A and 3B for adults. See Key Question 4B for juveniles.	See Key Questions 3A and 3B for adults. See Key Question 4B for juveniles.	See Key Questions 3A and 3B for adults. See Key Question 4B for juveniles.
7D.2 Size and age of returning adults and juvenile outmigrants.	See Key Question 3C for adults. See Key Question 4D and 6C for juveniles.	See Key Question 3C for adults. See Key Question 4D and 6C for juveniles.	See Key Question 3C for adults. See Key Question 4D and 6C for juveniles.	See Key Question 3C for adults. See Key Question 4D and 6C for juveniles.
7D.3 Broodstock Fecundity	See Key Question 3B	See Key Question 3B	See Key Question 3B	See Key Question 3B

Objective 8.0: Determine the percent hatchery-origin spawners (pHOS), proportionate natural influence (PNI) and pNOB (for integrated programs)

The purpose of Objective 8.0 is to monitor the genetic influence of hatchery programs on natural populations using HSRG-recommended metrics and targets (Table D-2).

Table D - 2. Current population designations (LCFRB 2010), hatchery program types, and HSRG-recommended targets for pHOS and PNI for the North Fork Lewis River salmonid populations.

Population	Current Hatchery Program Type	Current Population Designation	HSRG pHOS Target	HSRG PNI Target
Spring Chinook	Segregated	Primary	< 5%	NA
Fall Chinook	None	Primary	< 5%	NA
Coho	Integrated (Late)	Contributing	< 30%	≥ 0.50
Coho	Segregated (Early)	Contributing	< 5%	NA
Winter Steelhead	Integrated (Late)	Contributing	< 30%	≥ 0.50
Winter Steelhead	Segregated (Early)	Contributing	< 5%	NA
Summer Steelhead	Segregated	Stabilizing	< 5%	NA
Chum	None	Primary	< 5%	NA

KEY QUESTION 8A: WHAT ARE THE TRENDS IN PHOS, PNI, PNOB AND PEHC AND DO THEY MEET HSRG RECOMMENDATIONS BY PROGRAM (WHEN APPLICABLE)?

APPROACH

Hatchery-origin and natural-origin spawners will be differentiated based on hatchery marks or tags; marking strategies vary by species and are described in sections A3.5, B3.5, C3.5. The calculation of pHOS, pNOB, and PNI rely on accurate spawner abundance estimates for each group based on assignments made in the field as described in Strategy A. PEHC is similar to pHOS in that it measures gene flow between hatchery and natural-origin fish based on spawner composition, however PEHC is calculated using genetic data of offspring to estimate parentage, described in greater detail in Strategy G.

- pHOS is the proportion of adults spawning naturally that are hatchery-origin spawners (HOS) for a given population and year.
- pNOB is the proportion of a hatchery broodstock composed of natural-origin adults each year.

- PNI is the proportion of natural influence on a population composed of hatchery and natural origin fish (i.e., integrated program).
- PEHC estimates interbreeding between hatchery and natural-origin spawners based on genetic parentage analysis of offspring.

Steelhead

Because steelhead are iteroparous, the number of fish of each origin must be made based on observations of live steelhead. A draft multi state mark-recapture model was developed by the U.S. Geological Survey to estimate pHOS in the population of late winter steelhead that spawn in the North Fork Lewis River downstream of Merwin Dam. The model used data collected in 2018 and 2019 to derive estimates of pHOS. However, since 2019 the practice of using tangle nets became a concern and this invasive method was affecting spawning steelhead behavior and success in the lower river, especially as the spawning period progressed. In 2023, the ATS will need to decide on whether an alternative model should be developed or adopt alternative collection methods to sample in-river steelhead.

Chinook and Coho Salmon

Seasonal surveys of Chinook and Coho salmon carcasses are performed weekly throughout the spawning periods for Chinook and Coho salmon, outlined in Objective 5.1 and Strategy A. The origin (NOR, HOR, or Unknown) is recorded for sampled carcasses. The number and composition of carcass recoveries is a direct result of the recovery probability for each individual carcass, which are influenced by many factors (e.g., spawning timing, spatial distribution, sex and size of carcass), therefore, total estimates of pHOS will be derived by weighting of raw recovery data by relative abundance as described in Strategy A.

LIMITATIONS OR CONCERNS

Steelhead

The current steelhead model assumes that capture efficiency at the Merwin Trap is 100 percent and that all fish are correctly identified and recorded (e.g. noting residuals that migrate to the Merwin Trap). Additionally, while this model does not address all possible contingencies (e.g. capture efficiency varying among groups, or different rates of residualism among hatchery and natural populations), the posterior predictive check demonstrates that the model is adequate for the main goal of estimating the proportion of hatchery-origin spawners. However, the possibility for extensions or variations of the model to be evaluated in the future with more formal model comparison techniques remains.

Chinook Salmon

There is substantial temporal and spatial overlap between spring and fall runs of Chinook in the North Fork Lewis River, reducing the ability to reliably differentiate between fall and spring run Chinook in the field. Misidentification of carcasses as either spring or fall run will affect pHOS estimates because the vast majority of fall run Chinook are of natural origin whereas the spring run is predominantly of hatchery-origin from the segregated hatchery program.

Coho Salmon

A substantial portion of returning Coho either are trapped or spawn in tributaries of the mainstem North Fork Lewis River. Because most of the carcass recovery effort is focused on the mainstem, sampling may not be representative of the total returns to the basin. pHOS will not be reported separately for early and late Coho.

DATA COLLECTION METHODS

Metric or Monitoring Indicator	Targets	Field Methods	Analytical Methods	Frequency and Duration
8A.1. Proportion hatchery-origin spawners (pHOS)	See Table D-2 above for population-specific targets.	For Chinook and Coho spawner origin is derived from spawner surveys. Refer to Strategy A. Methods for determining pHOS in steelhead are to be determined in coordination with the ATS.	pHOS = Number of HOS/(HOS + NOS)	Calculated annually
8A.2. Proportion natural-origin brood (pNOB)	A component of PNI target. Applies only to integrated programs. Late winter steelhead uses a pNOB target of 1 to achieve a PNI ≥ 0.50. Late coho follows a recommended integration rate (e.g., 30 percent) based on the designation for each stock or	For each integrated program (late winter steelhead and late coho), the origin and sex of each fish spawned will be recorded.	pNOB = NOB / (HOB + NOB). pNOB will be calculated within each spawning matrix, and for the total number of spawners.	Calculated annually

	population (e.g., primary, contributing, or stabilizing.)			
8A.3.Proportion natural influence (PNI)	See Table D-2 above for population-specific targets. This metric is influenced substantially by the pNOB. For example, if the broodstock incorporates 100 percent natural origin fish, PNI estimates cannot be less than 50 percent.	N/A	PNI = pNOB/(pNOB+pHOS)	Calculated annually
8A.4.Proportion Effective Hatchery Contribution (PEHC)	No specific targets recommended. Program should avoid increase in PEHC over time. Not relevant for integrated programs.	N/A	Refer to Strategy G.	Calculated annually

Objective 9.0: Monitor the post-release behavior of hatchery smolts and their potential impacts on native and ESA-listed species present downstream of Merwin Dam.

The purpose of Objective 9.0 is to provide a means for direct monitoring of ecological interactions between HOR and NOR juveniles if in-hatchery monitoring metrics described under Objective 4 are not achieved. This objective shall remain inactive for as long as the metrics described in Objective 4 remain measurable and within the targets provided each year in the AOP.

SECTION E ADAPTIVE MANAGEMENT

The ATS is tasked with reviewing technical aspects of program implementation within a given calendar year. The ATS Work Plan (Appendix A) is a calendar of ATS activities, program production tasks, and monitoring and evaluation strategies. The work plan also identifies specific decision points that the ATS tracks throughout the year. Adaptive management will be used to periodically evaluate and adjust activities covered by the AOP, including H&S Program implementation and the monitoring and evaluation (M&E) activities described in Section D.

1.0 REVIEW MILESTONES

As envisioned in section 8.2 of the Settlement Agreement, program components will be evaluated in the following processes and documents:

- The H&S Annual Operating Report (AOR) compiles all information gathered pursuant to implementation of the H&S Plan, including recommendations for the ongoing management of the H&S Program, and is provided to the ACC for review and comment. The AOR is intended to provide estimates or results for each of the key questions and related objectives (Section D) evaluated each year. The AOR will include key highlights, accomplishments and technical recommendations to focus the ACC review as part of the Executive Summary. It is important to note that while the AOR provides estimates and results from annual monitoring activities and may recommend technical changes to program or M&E implementation, it does not provide decisions on the continuation of specific monitoring actions, management decisions, nor would the results reported trigger changes to management without further review. This decision-making process is the role and adaptive management function of the ATS and ACC as described in the following section.
- The H&S Plan will be updated every five years at a minimum, in coordination with the ACC and with the approval of the Services. (More frequent updates may be triggered by changes to the regulatory or management landscape, such as the approval of new HGMPs.) The update will consider recommendations from members of the ACC and the 10-year Comprehensive Review (described below), and identify those recommendations that have not been incorporated into the H&S Plan with a brief statement as to why the changes were not made. It is expected that information brought forward in the AORs will provide the basis for ACC recommendations for updating the H&S Plan.
- A Comprehensive Review will be undertaken every 10 years. In consultation with the ACC, an independent consultant will be hired to assess the H&S Program for the factors described in section 8.2.6 of the Settlement Agreement. The Comprehensive Review will consider all available data collected and reported as part of the AORs to evaluate the program's effectiveness and impacts in light of recent scientific advancements in hatchery science.

2.0 ROLES IN PROGRAM REVIEW AND DECISION-MAKING

2.1 Aquatic Coordination Committee (ACC)

The ACC is responsible for implementing the Settlement Agreement with the ability to implement changes to the Settlement Agreement through a Consultation process as defined in Section 14 of the Settlement Agreement. Therefore, modifications to the H&S Program implementation that may influence the Settlement Agreement or the Outcome Goal of the Agreement, require review and approval by the ACC prior to implementation. The ATS must request approval by the ACC of decisions that go beyond technical modifications, include programmatic recommendations, or decisions that modify interpretation of the intent of the H&S Program as described in Section 8 of the Settlement Agreement,. Prior to approval, the ACC may require use of the Decision-Making Template (see ACC ground rules document) to document and record decisions relating to implementation of the Lewis River Settlement Agreement or overall Outcome Goal of the Agreement.

2.2 Aquatic Technical Subgroup (ATS)

The ATS functions as a technical advisory group to the ACC. The ATS reviews and revises the AOP annually with a focus on technical approaches and protocols, with consideration for how changes in methods may affect the continuity of datasets and program implementation. The ATS may also make in-season decisions to ensure continuity of program implementation, such as adjusting broodstock collection curves in response to actual size and timing of an annual return. Reviewing and revising the AOP with flexibility for in-season adjustments ensures program implementation and M&E activities are achieved each year.

3.0 ADAPTIVE MANAGEMENT DECISION MAKING PROCESS

Adaptive management of the H&S Program is done through a cycle of annual and periodic steps (as defined in the Settlement Agreement) that provide the means to modify the H&S Program through formal decision making.

The first step in the adaptive management process is the annual reporting of program performance trends. This step is achieved upon distribution of the AOR. Where available, the AOR provides annual results in the context of multi-year trends. The report includes a brief Executive Summary identifying notable trends and key concerns from the past year, and recommendations for adjustments to monitoring and evaluation activities.

The second step is an annual 30-day ACC review process which includes the following:

- Coinciding with the distribution of the review draft of the AOR to the ACC, PacifiCorp will summarize key findings during the ACC meeting in May to highlight accomplishments and identify any notable concerns to assist the ACC in their review.
- Upon completion of the 30-day review, the ACC, in coordination with the ATS, may recommend revisions to the AOP for the subsequent year in the form of informal consultations or request for decision using the ACC Decision Template.

The third step is to update the H&S Plan every 5-years, either in response to ACC recommendations based on data trends that are tracked by the ATS and reported in the AOR, or in response to recommendations given in the Comprehensive Periodic Review (described next).

The fourth and final step is a Comprehensive Periodic Review of longer-term data trends in the H&S Program every 10-years. This review will be undertaken by an independent third party to identify data gaps and modifications to the program implementation to ensure the H&S Program is achieving the stated goals, stipulated in Section 8 of the Settlement Agreement. The ATS shall review conclusions and results from the Comprehensive Periodic Review to determine whether formal program decisions or trigger points are warranted and, if so, develop recommendations for ACC review and approval using the ACC Decision Template.

The elements of the adaptive management cycle are shown in Figure E-1 and schedule in Table E-1, below.

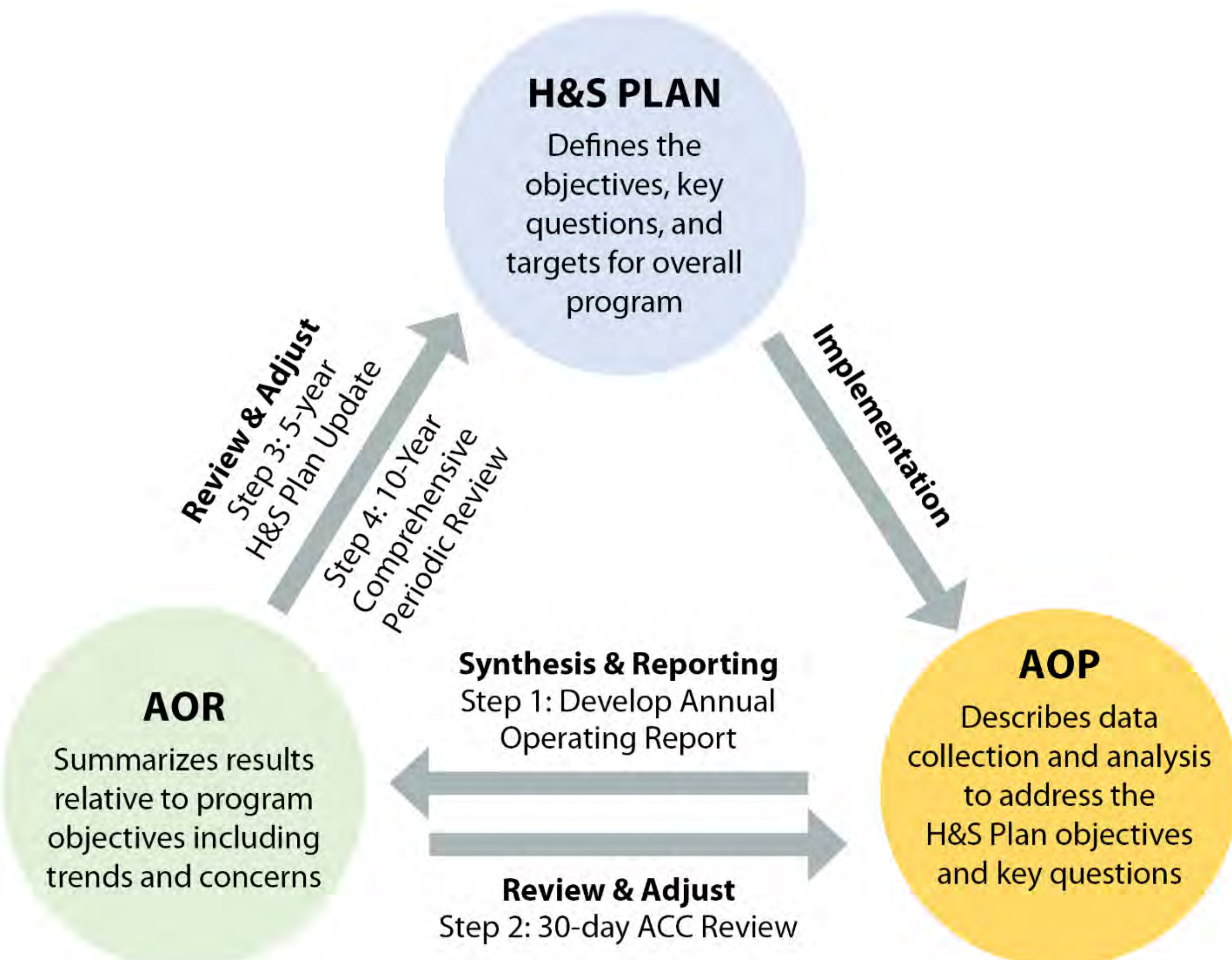

Figure X-X Graphical depiction of the H&S Program adaptative management approach and cycle through its corresponding plans (H&S Plan, AOP), reports (AORs), and review processes

Table E - 1. Adaptive Management Milestones

		2023	2024		2025	2026	2027	2028	2029	2030	2031
Annual Operating Report Development	PacifiCorp										
Annual Operating Report Review	ACC, ATS										
Annual Operating Plan Revisions	ATS										
Comprehensive Periodic Review	Indep. Reviewer										
H&S Plan Update	ATS										

SECTION F REPORTING REQUIREMENTS

Annual reporting of AOP implementation and monitoring of objectives is provided as part of the Lewis River Annual Operations Report, distributed for review in late April of each year. According to the Settlement Agreement, the annual report will include, at a minimum, the following:

1.0 ADULT COLLECTION AND SPAWNING

- Collection numbers by location and method
- Collection numbers compared to targets
- Genetic assignment results for steelhead
- Spawning protocols and numbers
- Transportation numbers by date, species, and sex ratios (actual versus goals)
- Distribution of all collected species
- Disposition of any species

2.0 EGG INCUBATION AND JUVENILE REARING/RELEASE

- Egg take – actual versus goals
- Egg to fry survival – numbers of fish ponded
- Pathogen screening results
- Rearing strategies that differ from routine operations (e.g., use of circular rearing strategies)
- Smolt releases, length, and location (actual versus goals)
- Tagging and marking summary (PIT tags and BWTs)

3.0 MONITORING AND EVALUATION

Results of activities undertaken for each monitoring and evaluation objective. Reporting will be completed in accordance with the 2020 H&S Plan Objectives; however, these objectives are not described in this 2021 version of the AOP because it has yet to be approved by FERC. At a minimum, these monitoring and evaluation results reported will include the following along with associated confidence intervals and coefficient of variance, where applicable:

- Adult escapement estimates (abundance) downstream of Merwin Dam
- Adult composition (hatchery versus natural origin) on spawning grounds downstream of Merwin Dam
- Spatial and temporal distribution of spawning downstream of Merwin Dam
- Juvenile migration and residualism estimates of hatchery releases downstream of Merwin Dam
- Hatchery juvenile monitoring for ecological interactions with NOR smolts
- Summaries of screw trapping results including locations fished, time periods fished, catch rates (relative abundance) by species (composition), trapping efficiency, and estimates of juvenile abundance by species

- Distribution maps of redd locations and counts for each species

4.0 CONSISTENCY AND ADHERENCE WITH HSRG GUIDELINES

Annual reporting may include recommendations to ensure that Lewis River hatchery operations are consistent with recommendations of the HSRG.

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SECTION H MONITORING STRATEGIES

Strategy A: Adult Abundance and Composition

Strategy A: Adult abundance and composition of Chinook, chum, coho and late winter steelhead downstream of Merwin Dam

I. Chinook and Chum Salmon

Chinook Salmon

The LCR Chinook salmon (*Oncorhynchus tshawytscha*) ESU consists of 32 historical independent populations that are distributed from the mouth of the Columbia River upstream to the Hood River in Oregon (NMFS, 2013). LCR Chinook salmon exhibit two dominant adult migration patterns based on when individuals return to freshwater to spawn. These two dominant life-history strategies have been used to categorize populations as either spring or fall Chinook (Myers et al., 2006). Fall-run Chinook have been further separated into two stocks, "fall-run" and "late fall-run," and are referred to as tules and brights, respectively. Hereafter, the term "fall-run" Chinook will be used in reference to the combination of these two stocks while the terms "tule" and "bright" will be used to specify the specific fall-run stock.

The Lewis River watershed contains three of the 32 LCR Chinook salmon populations among which display each of the three unique run-types (spring, fall, and late-fall). WDFW, formerly the Washington Department of Fisheries, has been monitoring Chinook in the Lewis River for decades. Specifically, adult spawning ground surveys have been conducted annually since 1964 and these data have been used to derive estimates of adult fall Chinook escapement. Juveniles (pre-smolt) have been sampled and have received CWTs since 1983 as part of the Lewis River Wild Fall Chinook Tagging Project (Hawkins, 2018a). CWT Chinook are then recovered in fisheries and on the spawning ground. Escapement estimates and CWT recoveries are used in conjunction to: 1) assess the effects of Lewis River flows (i.e., dam operations) on juvenile production and adult abundance; 2) reconstruct the Lower River Wild spawning stock component unit, which is one of the five Columbia River fall Chinook management units; and 3) evaluate abundance, survival, and fishery harvest as part of the Pacific Salmon Treaty, Pacific Fishery Management Council, Pacific Salmon Commission, and Columbia River Compact processes.

Over the years, the methods used to estimate adult escapement have changed. In 1976, a mark-recapture carcass tagging study was conducted to generate an estimate of escapement in the Lewis River (see "Area of Focus" below). From this estimate, an expansion factor was derived relating a peak count of live and dead Chinook in an index section to total abundance (McIssac, 1977). For the following two and a half decades, WDFW used peak count index surveys to generate estimates of Chinook escapement. In the early 2000s, WDFW revisited the methods that were being used to estimate the abundance of Lewis River Chinook. Specifically, there was concern that the peak count expansion factor was not producing accurate estimates as the peak spawn time period had shifted later into the fall when survey conditions were not as favorable (i.e., increased flows and turbidity during the new peak spawn time frame decreased survey visibility and thus estimates of abundance). Therefore, WDFW conducted three years (2000 to 2002) of mark-recapture carcass surveys and derived updated (age-specific) expansion rates based on carcass recoveries, which is known as the "bright-eye method." The bright-eye method has been used to estimate escapement annually since 2002. However, this method currently does not meet the monitoring recommendations that have been established for salmon and steelhead populations by NMFS (Crawford and Rumsey, 2011) and the AOP (H&S Subgroup 2015). These limitations led WDFW to reinitiate annual mark-recapture carcass surveys of Lewis River Chinook from 2013 – 2017 and conduct a comparative analysis of analytical methods to determine which estimator generated the least unbiased estimate of adult abundance and composition. Based on the results of this analysis, it was recommended that estimates of abundance be generated using mark-recapture surveys and an open-population Jolly-Seber analysis until a more cost-effective, alternative method has been developed that can generate estimates by stock, origin, sex, and age with comparable uncertainty and robustness to model assumptions.

Chum Salmon

The Columbia River chum salmon (*Oncorhynchus keta*) ESU consists of 17 historical independent populations that are distributed from the mouth of the Columbia River upstream to the Deschutes River in Oregon (NMFS, 2013). Historically, hundreds of thousands of chum adults returned annually to the LCR and the Lewis River population was estimated to have supported close to 100,000 chum at equilibrium, second only to the Cowlitz River (Johnson et al., 1997, Good et al., 2005). However, over the past 50 to 75 years, returns of chum salmon have dramatically decreased both within the Lewis River and throughout the Columbia River Basin. Today, 15 of the 17 historical Columbia River chum populations, including the Lewis River, are considered extirpated or nearly so (LCFRB 2010, NMFS, 2013). The low abundance of chum salmon in the Lewis River is seen directly in the monitoring data collected over the past several decades. For instance, over the past 10 years, an average of nine chum carcass (range: 0 to 28) have been recovered on the spawning ground over the course of the 4- to 5-month fall survey period.

Specific Objectives

Following the listing of many salmon and steelhead populations under the ESA, NMFS developed a framework to assess the status, trend, and long-term viability of "Viable Salmonid Populations" (McElhaney et al., 2000). This framework has been converted to a set of monitoring guidelines that focus on estimating Viable Salmonid Population indicators, which are broken up into four main categories: abundance, productivity, spatial distribution, and diversity (Crawford and Rumsey, 2011). Based on these recommendations, the specific objectives to evaluate Chinook and chum populations downstream of Merwin Dam include:

1. Generate an "unbiased" estimate of adult fall Chinook (i.e., escapement) downstream of Merwin Dam with a CV on average of 15% or less.

2. Generate an estimate of adult chum and spring Chinook (i.e., escapement) downstream of Merwin Dam. (Note: currently chum and spring Chinook abundance is low and thus generating estimates has been difficult.)
3. Determine adult composition of fall Chinook on spawning grounds downstream of Merwin Dam (i.e., stock: spring, tule, bright; origin: wild, hatchery; sex; age).

Study Area

The majority of Lewis River fall-run Chinook spawning occurs between the Lewis River Hatchery (RM 15.7) and the base of Merwin Dam (RM 19.5). Additional spawning occurs in the mainstem Lewis River between the bottom of Eagle Island (RM 10.0) and Lewis River Hatchery as well as in Cedar Creek and the East Fork Lewis River, which enter the Lewis River at RM 15.7 and 3.5, respectively.

The adult mark-recapture carcass tagging surveys are conducted in the Lewis River from the bottom of Eagle Island upstream to just below Merwin Dam. This total survey area has been delineated into five survey sections (Table 1). These survey sections were established during the original 1976 carcass tagging study and have remained constant since. Sections 1 to 4 are each approximately 1 mile in length and together make up the index count section dating back to the mid-1950s. Section 5 consists of a split channel surrounding Eagle Island. Cedar Creek and East Fork Lewis River Chinook have been surveyed and their data analyzed separately. Therefore, these data were not incorporated into this current evaluation. However, an effort will be made in the future to generate "total" Lewis River tule and bright abundance estimates that incorporate all major spawning areas.

Table 1. Description of fall Chinook carcass survey sections on the North Fork Lewis River, 2013 – 2017

Reach Code	Length (miles)	Description
NFL-1	0.7	Upstream End: Pool Below Dam (RM 19.1) Downstream End: Back Eddy/Bottom of Sec No. 1 (RM 18.4)
NFL-2	0.8	Upstream End: Back Eddy/Bottom of Sec No. 1 (RM 18.4) Downstream End: Below Hagedorns (RM 17.8)
NFL-3	1.0	Upstream End: Below Hagedorns (RM 17.8) Downstream End: Top of Big Bar (RM 16.8)
NFL-4	1.1	Upstream End: Top of Big Bar (RM 16.8) Downstream End: Lewis R. Hatchery Boat Ramp (RM 15.7)
NFL-5	7.7*	Upstream End: Lewis River Hatchery Boat Ramp (RM 15.7) Downstream End: Bottom of Eagle Island (RM 10)

Note:
**Length includes both north and south channel around Eagle Island*

Data Collection Methods

Chinook Salmon

Carcass surveys are conducted on the Lewis River to estimate abundance and composition (i.e., race, origin, age) of adult spring and fall Chinook spawners (i.e., escapement) and presence/absence of adult chum spawners. Each year, surveys begin in mid- to late-September and continue through mid-January to mid-February. This time period encompasses the large majority Chinook and chum spawn timing period in the Lewis River. One to four jet boats are used to navigate the Lewis River and sample Chinook carcasses. Surveys are conducted weekly given that stream conditions are conducive to staff safety and fish visibility. Carcass surveys typically occur on a single day per week and are aligned with when river flows are the lowest. During the peak of the run (November through early December), PacifiCorp normally provides five drawdowns from Merwin Dam that reduce flows in the lower river in an effort to increase carcass recovery rates. Occasionally, a second survey day in a week is required due to large numbers of carcasses. When a second survey day is needed for a given week, the lowest reach (No. 5) is surveyed on day 1 prior to the scheduled draw-down while the upper four reaches (Nos. 1 to 4) are surveyed on day 2 and align with the draw-down. This order of operations prevents carcasses tagged on day 1 from moving downstream into reaches surveyed on day 2. Multiple surveys per week are treated as a single sample period.

During each survey, recovered carcasses are sorted and processed in a sequential manner (Figure D9). First, carcasses are identified by species. It relatively easy to identify the species of a fall spawning carcass (Chinook, coho, chum) unless the carcass is completely deteriorated. In the past, Chinook carcasses were not separated by stock in the field. Rather all Chinook carcasses were assigned as a just a Chinook and later apportioned as either a spring- or fall-run fish based on the scale age read (FW 1 = fall, FW 2 = spring). Due to changes in hatchery rearing and release strategies of spring Chinook (i.e., a portion of hatchery juveniles being released as sub-yearlings), adult carcasses will have to be assigned as either spring or fall Chinook based on external characteristics. However, the reliability of these visual assignments has not been evaluated and there is some doubt to the accuracy (Hawkins, 2018b). Therefore, moving forward, stock assignment likely needs to be evaluated.

Second, carcasses are sorted based on their recovery status (i.e., recovered versus not recovered) and the several external features of the carcass. Carcasses that cannot be recovered (e.g., too deep, pinned in a log jam) are enumerated and recorded as a Carcass Category 5 (Figure 1). Each recovered carcass is initially examined for the presence of a tail. A carcass with a severed (i.e., missing) tail is indicative of a previously sampled/recorded fish and are subsequently ignored. Carcasses with intact tails are then sorted based on whether or not a surveyor could determine if the carcass has been previously tagged. Previously tagged carcasses would have a tag on the inside of both opercula (see below). Therefore, if a carcass is missing its head and/or opercula, its previous tag status could not be determined. These carcasses are enumerated, denoted as either a Carcass Category 1 or 2 (Figure 2) and have their tail severed. Carcasses recovered with a slit belly are assumed to be sport caught and are enumerated and denoted as a Carcass Category 6.

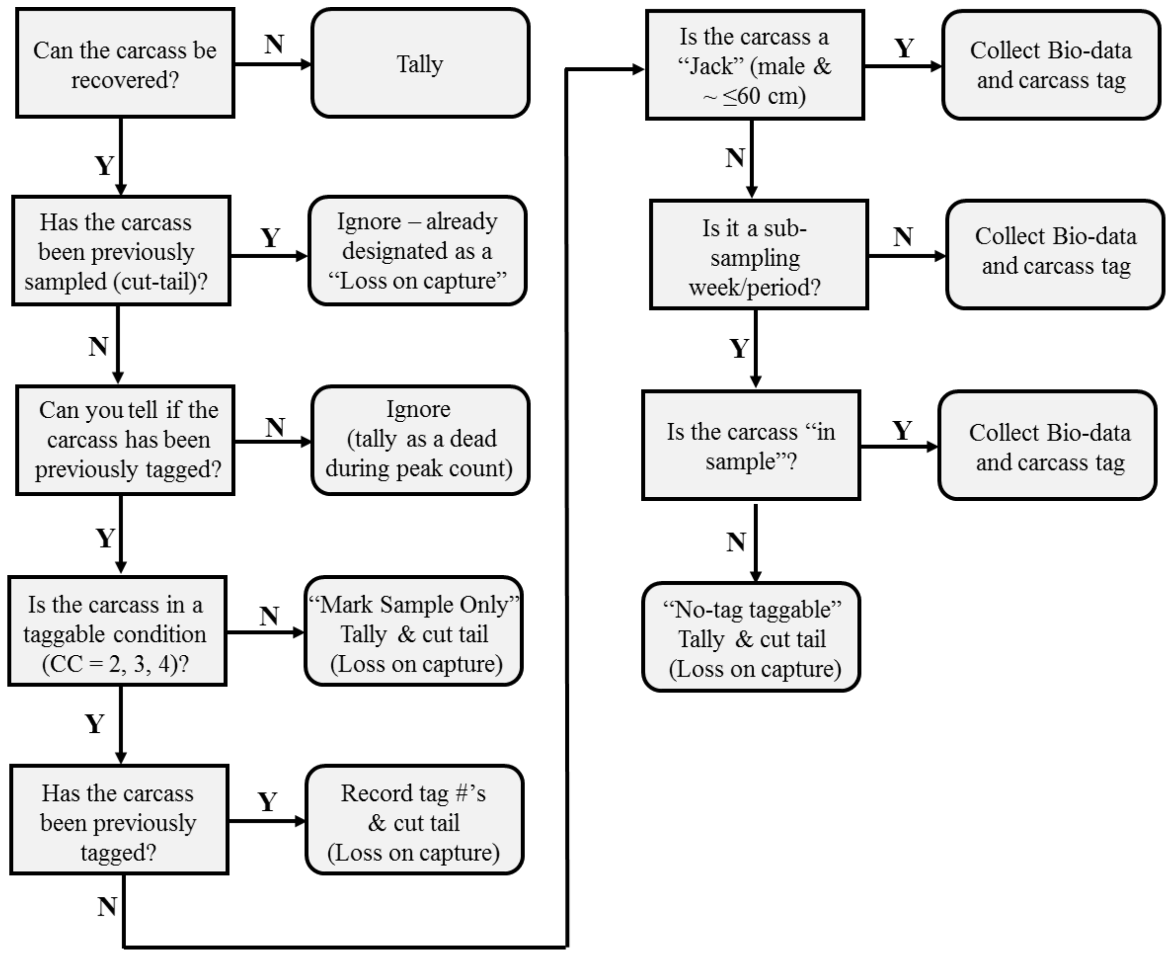


Figure 1. Sampling procedure for Fall Chinook carcass surveys on the North Fork Lewis River

Third, carcasses with intact heads and/or opercula are sorted as either taggable or untaggable based on their qualitative carcass condition category (Table 2). Taggable carcasses have numeric carcass condition scores of 2, 3, or 4 while untaggable carcasses are classified as either 5 or 6. Untaggable carcasses are designated as "mark sample only" fish, classified as either a "jack" or an "adult" group carcass (see below), have their adipose-fin status (see below) and Carcass Category recorded, examined for a CWT, have their tail severed to signify the carcass has been sampled (i.e., denoted as a "loss on capture" in JS model), and returned to the river.

Fourth, carcasses that are in taggable condition are then processed based on their capture history, size, and age. Carcasses are sorted into maiden and recapture recoveries. A recaptured carcass would have had a plastic numbered tag stapled to the inside of one or both opercula while a carcass with no opercula tags is classified as a maiden. Recaptured carcasses have their tag number(s) recorded, tag(s) removed, tail severed to denote the carcass was sampled, and returned to the river. Maiden captures are sorted into two groups based on their sex and fork length. The first group were classified as "jacks/group 1" and consisted of small(er) males whose fork length was less than approximately 60 cm. The second group were classified as "adults/group 2" and consisted of females and large(r) males who fork length was greater than or equal to approximately 60 cm. It should be noted that these group classifications were largely based on visual assessment of fish length. Therefore, a portion of the "jack" group carcasses consisted of small "adult" males and vice versa due to both inaccuracies in visually classifying carcasses by length (e.g., a 62 cm carcass placed into the "jack" group) and variability in length-at-age (e.g., a 62 cm carcass classified as an "adult" was in fact a true, age-2 jack). Regardless, this slight variation in the group classification of each carcass (i.e., "jacks" versus "adults") did not have any impact on the accuracy of abundance estimates as age-distribution was apportioned using weekly scale samples (see below).

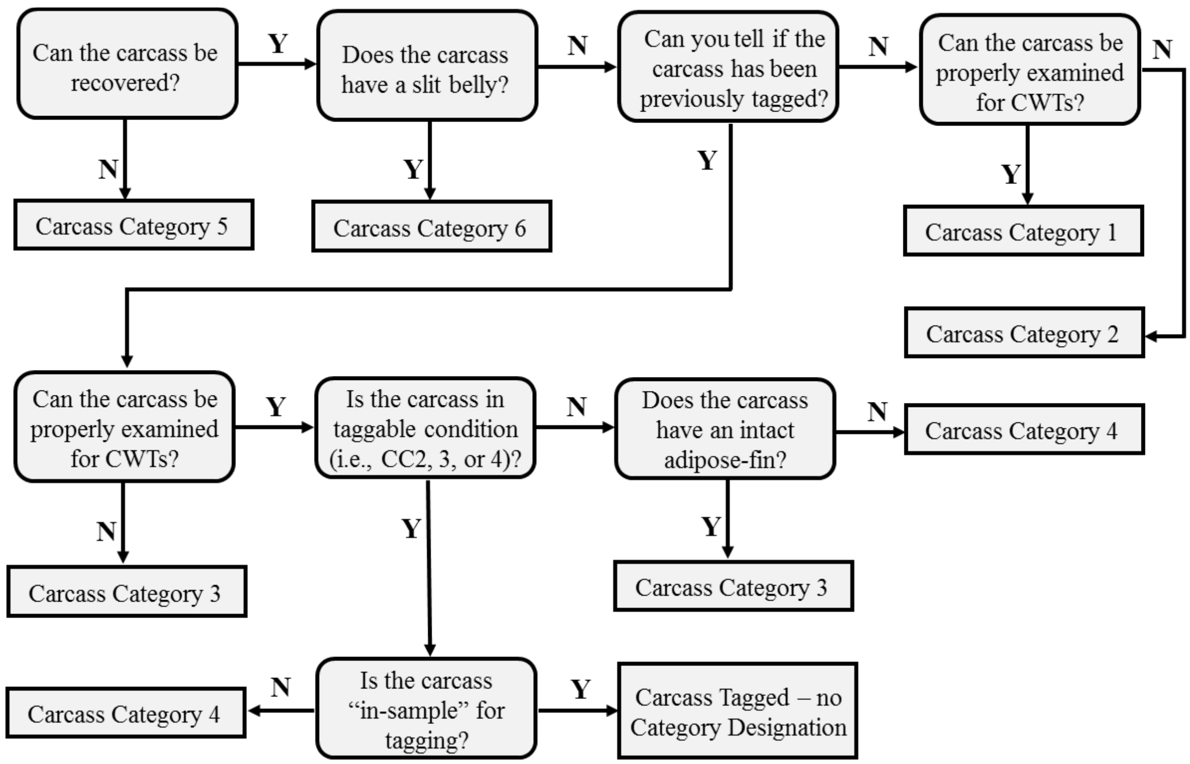


Figure 2. Diagram of how the Carcass Category of each individual fish is determined and classified for carcass surveys conducted on the North Fork Lewis River.

Note: Carcass Categories are only assigned to maiden fish (i.e., previously unsampled) carcasses that are not carcass tagged.

Fifth, carcasses are then processed based on the weekly sampling rate. Specifically, carcasses are sorted into two groups ("in-sample" or "out-of-sample") based on the sampling rate for a particular week. In all weeks, small (group 1) carcasses are sampled at a 1:1 rate due to the low overall recoveries of fish in this category. In most weeks, large (group 2) carcasses are sampled at a 1:1 rate. However, sub-sampling occurs in most years during peak weeks when the number of recovered carcasses is too high to sample at a 1:1 rate. Sub-sampling rates are predetermined based on the anticipated number of recoveries for a particular week and can vary from 1 in 2 to 1 in 10 among weeks and years. Out-of-sample carcasses have their tail severed to denote the carcass was sampled and returned to the river. If an out-of-sample carcass has a missing adipose fin, it is also scanned for a CWT.

Lastly, in-sample (taggable, maiden) carcasses are bio-sampled and tagged. Carcasses are examined for the presence or absence of an adipose fin and CWTs. Prior to mid-November, all recovered carcasses are scanned for a CWT using a handheld wand regardless of adipose status due to the possible presence of double-index tagged strays. After mid-November, only carcasses with missing adipose fins are scanned for CWTs. Carcasses that wand positive for a CWT have their snouts removed and collected. All carcasses then have their sex, fork length, and condition recorded (Table 2), scales collected for aging, and are tagged. Chinook carcasses are tagged by stapling a plastic numbered tag on the inside of both opercles. Tagged carcasses are then returned to the river in the section they were collected (Table 1) and in flowing water to facilitate mixing with untagged carcasses.

Due to overall low abundance of chum salmon in the Lewis River, recovered carcasses are not tagged but rather bio-sampled and enumerated.

Table 2. Carcass condition categories codes and the associated description of carcasses

Category (Numeric)	Category (Alpha)	Description of Carcass Condition
1	L	Live, still gilling or moving*
2	F	Fresh, both eyes clear, gills bright red
3	D-	Slightly decayed, eyes cloudy, firm flesh
4	D	Decayed, eyes cloudy, soft flesh
5	D+	More decayed, eyes cloudy, very soft flesh
6	S	Skeleton, loosing flesh

*Note: live fish were not tagged

Data Management

Field data are recorded on a combination of scale cards and a white board. Individual carcasses that are bio-sampled and/or carcass tagged have their corresponding data (tag number, fork length, carcass condition, CWT sampling number, scales, and recovery section i.e., 1 to 5) recorded on the front of a scale card. Each column represents one carcass and each card holds approximately 19 samples. Details regarding survey date, section number, sample rate, and the number of carcasses sampled are recorded on the back of the scale card. Tag numbers from carcass recoveries and the number of non-taggable ("mark sample only") carcasses are recorded on a white board. Specific details on field data recording methods and terminology can be found in the WDFW's "Stream Survey Manual" (WDFW, 2018). At the end of each survey day, the number of non-taggable carcasses are tallied and recorded by section in the "plus count" field on the back of a corresponding scale card. Field data are entered into WDFW's Traps, Weirs, and Surveys Access database as well as a separate Excel spreadsheet throughout the survey season. Entered data are provided QA/QC at the end of the season and any errors or missing information are corrected. Fish scales and CWT samples/recoveries are processed by WDFW laboratories in Olympia. Specific details how scales and CWTs are processed can be found in Rawding et al. (2014: page 12).

Analysis Methods

Fall Chinook

The abundance and composition of fall-run Chinook spawners (i.e., escapement) is estimated using an “open” population JS model (Seber, 1982, Pollock et al., 1990). Specifically, it is a “super population” JS model that was developed by Schwarz et al., (1993) specifically for estimating salmon spawning escapement using mark-capture methods and was built upon previous work by Crosbie and Manly (1985) and Sykes & Botsford (1986). The super population model has been successfully implemented to estimate spawner escapement for other salmon populations within the LCR (Rawding et al., 2014) and other Washington state watersheds (Ashcraft et al., 2017).

A comprehensive description of the super population JS model, including summary statistics, fundamental parameters, derived parameters, and likelihoods is provided in Rawding et al., (2014). Briefly, total spawner escapement is estimated as the sum of “newly arrived” carcasses (i.e., gross births – B*) that enter the study system over the entire survey period. This estimate of new carcasses includes both the number of carcasses that were present (i.e., available to sample) during each sampling period as well as the number of carcasses that arrived after a particular sampling period but were lost/removed (e.g., washed out) before the subsequent sample period. A conceptual diagram of the JS model and its main components is shown in Figure D11.

Prior to running the JS model, fall Chinook carcass data are queried from the Traps, Weirs, and Surveys Access database for each individual survey year and ran through a series of summarizations and tests implemented through the program R (R Development Core Team, 2011). First, carcass data are run through a standardized set of decision rules that classify the stock (tule, bright), origin (hatchery, wild), sex (jack, female, male), and age of each individual carcasses based on their sampled biological data. These biological data are then summarized by sample period (i.e., week) and these summaries are subsequently used in the model to apportion the abundance estimates into reporting groups (see below). Second, recapture probabilities are evaluated by sex and size using logistic regression (Link and Barker, 2006). The results from these tests influence how carcass data are stratified (i.e., grouped; see below). Third, capture histories are generated for each individual carcass and then JS summary statistics (e.g., ni, mi, ui, Ri) are generated by week/period using the RMark package (Laake, 2013). Only tagged individuals or carcasses with a Carcass Category of 3 or 4 are used in the analysis. When necessary, weekly summary statistic carcass data are “partially” pooled to satisfy the JS modeling requirement for the number of marks and recaptures in individual periods. Fourth, four potential abundance models are evaluated for fit of using Bayesian Goodness of Fit (GOF) tests (Gelman et al., 1996).

The four models that are evaluated include a combination of static (s) or time varying (t) probability of capture (p), survival (ϕ), and entry (b*) among survey periods/weeks (i.e., ttt, stt, tst, sst). Capture probability is the odds of sampling a carcass that was present, survival probability is the odds that a carcass that was present in a sample period will remain in the study system until the next sample event, and entry probability is the odds that a carcass will enter/arrive into the study system in a particular period.

Based on the results of the logistic regression and GOF tests within and among years, we have chosen to standardize the overall JS modeling approach. Based on previous year's data and results, the logistic regression tests generally concluded that jacks (males less than 60 cm), females, and males (greater than or equal to 60 cm) had statistically different recapture rates among years. This result corroborates previous literature based on size and behavioral difference between these three groups. The GOF tests concluded that while simpler models (tst, stt, sst) sometimes provided adequate model fits for a carcass group (jacks, females, males) within a year, the completely time-varying (ttt) model always provided an adequate fit for all groups among all years. Therefore, abundance is estimated for the three carcass groups separately, but within the same model, using the time-varying (ttt) model JS model among all years.

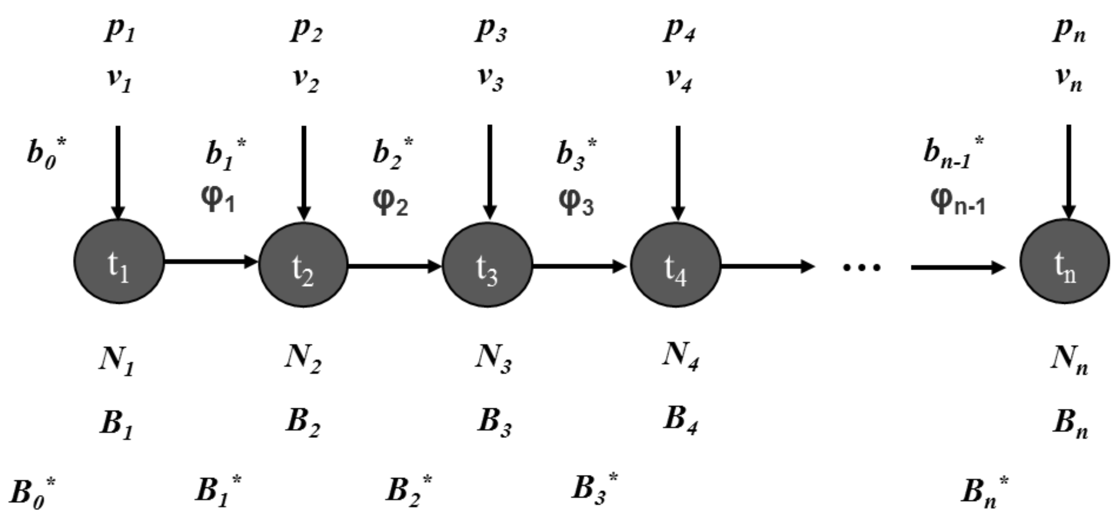


Figure D11. Conceptual diagram of "super population" Jolly-Seber abundance model
Note: Model developed by Schwarz et al. (1993) – diagram adapted from Schwarz and Arnason (2006)

Fundamental parameters of the model include: sample period i (ti), probability of capture at sample period i (pi), probability that a carcass captured at time i will be release, opposite of a loss-on-capture (vi), probability that a carcass enters the population between sample periods i and i+1, which is referred to as probability of entry (bi*), probability of a carcass surviving and remaining in the population between sample periods i and i+1 (ϕi). Derived parameters of the model include: population size at sample period i (Ni), number of fish that enter after sample period i and survive to sample period i +1 (Bi), and number of fish that enter between sampling period i-1 and i, these are referred to as gross births (Bi*). Total abundance is calculated as the sum of B* over all sample periods.

After abundance estimates are generated, weekly B* estimates are apportioned using summarized biological data by race (tule, bright), origin (wild, hatchery), sex (jack, female, male), and age. Specifically, abundance is estimated by race and origin using the weekly ratio of CWT recoveries and adipose-fin status (clipped, unclipped), respectively, and a binomial distribution. The same race and origin biological data set is used for all three groups scaled to the appropriate number of periods per group. Abundance estimates by sex are already calculated based on our three groupings. However, male carcasses need to be adjusted based on age data as field calls of jacks and adult males were "approximations" based on size. Abundance estimates are apportioned by age using a multinomial distribution. Age data are separated by group due to expected difference in age-distribution among the three groups. For each estimate, a vague "Haldine" beta distribution prior is used. Weekly estimates are summed across all sample periods to derive a total estimate of abundance and proportion by group.

The JS model is parameterized using a Bayesian framework. Here, parameters are estimated from the posterior distribution, which is calculated as the product of the prior distribution and the probability of the data given the model or likelihood (Gelman et al., 2013). Samples from the posterior distribution are obtained using Markov chain Monte Carlo simulations (Gilks, 2005) in WinBUGS (Lunn et al., 2000) using the R2WinBUGS package (Sturtz et al., 2005). WinBUGS implements Markov chain Monte Carlo simulations using a Metropolis within Gibbs sampling algorithm (Spiegelhalter et al., 2003). Two chains are run with the Gibbs sampler with an appropriate number of iterations and burn-in period so that the number of independent samples, as measured by effective sample size, is at least 4,000 for each parameter of interest. An effective sample size of 4,000 provides a 95% credible interval that has posterior probabilities between 0.94 and 0.96 (Lunn et al., 2012). A vague "Bayes-LaPlace" uniform prior is used for the JS abundance calculations. Initial values for each chain are automatically generated within the WinBUGS package. The sensitivity of the priors is evaluated based on the overlap between the prior and the posterior distribution (Gimenez et al., 2009). Model convergence is based on visual assessment of traceplots for chain mixing and evaluation of the Brook-Gelman-Rubin statistic (Su et al., 2001). Based on these steps, the reported posterior distributions are assumed accurate and that they represent the underlying stationary distributions of the estimated parameters.

Spring Chinook and Chum

In recent years, the numbers of spring Chinook and chum salmon returning to the Lewis River have been extremely low. During spawning ground surveys, there is typically only a handful to several dozen carcasses recovered each year. As a result, estimates of abundance through carcass recovery is challenging due to poor sample sizes. Alternative estimation methods (e.g., live/dead/redds counts) are largely ineffective due to both the overall low numbers of chum and spring Chinook seen each year, but also difficulty in getting accurate counts. While it is relatively easy to identify the species of a fish from a recovered carcass, it is difficult to get accurate assignment of species from visual counts of live fish and redds (Hawkins, 2018b). For instance, spring and fall Chinook overlap in spawn timing and it is challenging to separate the (potentially) relatively few spring Chinook from the thousands of fall Chinook that are often seen in large schools on the spawning ground.

Therefore, estimates of chum abundance and spring Chinook will be based on carcass recoveries and should be viewed as minimum estimate. Over the past several decades, WDFW has generated an estimate of spring Chinook abundance using the raw number of carcass recoveries and a recovery rate of ~50%, which is based on limited data from recoveries of spring Chinook and recoveries from fall Chinook during the same time period.

Assumptions

When the assumptions of a super population JS model are met, it produces unbiased estimates of escapement with known levels of precision. Within the JS model, there are specific assumptions as to how recruitment (i.e., newly arrived carcasses) is modeled, but overall there are a total of four critical assumptions for open population models that must be met to obtain unbiased estimates (Seber, 1982):

- *Equal Catchability*: Each carcass that is present in the study system during a specific sample event, whether tagged or untagged, has the same probability of being sampled
- *Equal Survival*: Each carcass that is present in the study system during a specific sample event, whether tagged or untagged, has the same probability of surviving (i.e., persisting) to the next sampling period.
- *Tag Loss and Recovery*: Tagged carcasses do not lose their marks and all marks are recognized and read properly on recovery
- *Instantaneous Sampling*: All samples are instantaneous, i.e., sampling time is negligible, and each release is made immediately after the sample.

These three assumptions will be assessed and dealt with each year using the following approach:

- *Equal Catchability and Survival*: The first two assumptions will be evaluated using two methods. First, capture probabilities will be assessed by sex and size using logistic regression (Link and Barker, 2006). The results from these tests will influence how carcass data are stratified (i.e., grouped). Second, abundance will be estimated using four separate models with static or timing probabilities of capture, survival, and entry among sample periods. The four models will be evaluated for fit using Bayesian GOF tests (Gelman et al., 1996). Based on previous year's data, estimates will be stratified into three groupings (jacks, females, and adult males) and run using a timing variation "ttt" model (see "Analysis Methods") unless there is evidence for unequal capture probabilities within one of these groups or data limitations (i.e., small sample size).

- *Tag Loss and Recovery*: Tag loss will be assessed through double tagging of carcasses. Correct reporting of carcasses will be maximized through the development of standardized datasheets and protocols and adequate training of field crews.
- *Instantaneous Sampling*: In order to meet this assumption of instantaneous sampling, the survey duration will be short (1 to 2 days) relative to the duration between surveys (5 to 6 days) and surveys are conducted weekly for the entirety of the study period.

Deliverables

Based on the outlined objectives, the following annual estimates will be generated and reported in an appendix to the Lewis River Annual Operations Report:

- Abundance and composition (race, origin, sex, age) for adult fall Chinook downstream of Merwin Dam with estimate of uncertainty
- Presence/minimum counts for adult chum and spring Chinook

II. Late Winter Steelhead

Abundance

Redd surveys will be used to estimate the abundance of winter steelhead spawners in the mainstem Lewis River downstream of Merwin Dam. Surveys will not include tributaries, and therefore, are not a total spawning escapement estimate, but rather an estimate of the mainstem spawning population. Tributary surveys may be incorporated at some point; however, until a practical approach is agreed to, abundance estimates will continue to focus on the mainstem Lewis River.

Composition

Currently, there is no strategy to assess composition of adult returning late winter steelhead on the spawning grounds. In 2017, a conceptual model for estimating the proportion of hatchery origin spawners was implemented and estimates of pHOS were estimated using the model in 2016 and 2018. However, this model relied on tangle net captures to provide the marks and subsequent recaptures needed to use the model for estimating pHOS. Tangle netting was discontinued in 2019 for two reasons: 1) concerns were voiced by PacifiCorp that naturally spawning late winter steelhead were being displaced resulting in some unknown reduction in naturally spawning success and 2) tangle netting was no longer needed for broodstock as the Merwin trap was collecting sufficient numbers of late winter steelhead for the hatchery brood program.

Study Area

Surveys will be conducted from the Merwin boat ramp to the downstream end of Eagle Island (Figure D6). On each survey, the coordinates of newly identified redds will be recorded using recreational grade GPS receivers. Each new redd will be marked with a GPS waypoint(s). Each waypoint will be time stamped by the GPS receiver.

Figure D6. Late winter steelhead redd survey area

Abundance Estimate

To estimate in-river escapement abundance, the number of redds will be multiplied by a 0.81 multiplier, used broadly for LCR steelhead stocks. This multiplier represents the apparent number of redds per female in Snow and Salmon Creeks between 1977 and 1980 (Freymond and Foley 1986). The estimate of females is then multiplied by two to derive an estimate of spawner abundance. This calculation assumes and average sex ratio of 1:1 (Freymond and Foley 1986).

The assumed sex ratio of 1:1 may be modified based on winter steelhead captured during the year in which the redd survey is performed. If the ratio observed in the MCF differs from the assumed 1:1 ratio, abundance estimates will be calculated both for an assumed ratio of 1:1 and using a corrected ratio based on actual sex ratio observed from the MCF using the following equation:

$$N = (r * 0.81)s$$

Where N = Estimate of Lewis River total spawner escapement
r = Total number of new redds observed
s = Sex ratio of 1:1 (i.e., *s* = 2) or corrected by observed ratio from the MFCF

Redd surveys will be conducted every 7 to 10 days throughout the spawning season (March 1 to June 30) to provide a census count of all new redds constructed. By marking redd locations via GPS and using detailed notes, double counting of redds is minimized and a total count of unique redds can be developed. Conversion of redd counts to fish numbers as described above, yields an estimate of spawning escapement in the mainstem (excludes tributaries). Mainstem escapement estimates will be reported in the Lewis River Annual Operations Report. It is important to note that abundance estimates do not include late winter steelhead trapped and retained for broodstock at the hatchery or transported upstream of Swift Reservoir as part of the reintroduction.

Limitations and assumptions

Survey conditions on the Lewis River tend to be highly variable during the spring. Turbidity from runoff in the upper basin as well as flow regulation from Merwin Dam can negatively affect the ability of surveyors to identify redds. During periods of prolonged turbidity or high flows, AUC methods may be required to estimate redds from missed survey periods. However, AUC methods rely on a number of untested assumptions, including redd life, which can vary significantly over the spawning season.

Deliverables

- Total number and GPS location of new steelhead redds by date in the survey area
- Estimated spawner abundance for the mainstem Lewis River population (mainstem escapement)

III. Coho Salmon

Abundance

The geographic extent of the Lewis River coho salmon population, as defined by NMFS, includes the Lewis River mainstem and all tributaries above and below Merwin, Yale, and Swift dams, as well as several creeks that drain directly into the Columbia River in the vicinity of the Lewis River (e.g., Gee Creek). Due to differing management considerations associated with subcomponents of this overall population, monitoring and data analysis is further split into four subunits: 1) Lewis River mainstem below Merwin Dam; 2) Lewis River mainstem and tributaries above dams; 3) Cedar Creek Basin; 4) tributaries of the Lewis River mainstem below Merwin Dam, and creeks that drain directly into the Columbia River in the vicinity of the Lewis River (e.g., Gee Creek). Monitoring and estimation for each subunit is described below.

North Fork Lewis River Mainstem Below Merwin Dam

For the mainstem Lewis River, Jolly Seber (JS) mark-recapture via carcass tagging will be used to estimate coho abundance. This method requires surveying every 7 to 10 days of the mainstem Lewis River from Merwin Dam to the downstream end of Eagle Island. This section is divided into established reaches currently used for fall Chinook surveys. The timeframe for surveying begins October 15 and continues through January 31, as survey conditions allow. Mainstem carcass tagging and recoveries may occur on draw down days scheduled for fall Chinook surveys and will follow the schedule provided in Table D4.

Table D4. Proposed survey days for coho salmon surveys of mainstem and tributary areas

Activity	Day
Coho carcass tagging and recovery (Reach 5)	Monday and Tuesday
Coho tagging and recovery (Reaches 1-4)	Wednesday (Fall Chinook drawdown day)
Coho tributary sampling	Thursday
Coho tributary sampling	Friday (if necessary)

Recovered carcasses will be sampled for fork length, sex, and the presence/absence of an adipose fin and CWT to determine origin. CWT positive fish will have their snouts taken for CWT recovery. Additionally, up to 150 NOR coho (adipose intact, wand negative) should be scale sampled for age composition of NOR fish. Recovering CWTs provides the information necessary for determining origin and age structure of hatchery fish.

The carcass tagging method and accompanying analysis and assumptions used in the JS model are described in detail by Kinsel, et al., (2009) and are briefly summarized here. All carcasses that are intact (i.e., not completely decomposed) are tagged and biologically sampled as described above. Carcasses are tagged on both opercles with uniquely numbered plastic tags (McIssac 1977). Tags are placed on the inside of both opercles to limit bias from tag loss. Tagged carcasses are then placed into moving water to facilitate mixing with untagged carcasses (Sykes and Botsford 1986). On subsequent surveys and after drawdown days for fall Chinook surveys (mixing event), technicians perform recovery surveys and record the tag numbers of recovered carcasses. When tagged carcasses are recovered, the tags are removed and fish are marked by removing the tail, which is denoted as loss on capture in the JS model.

Tagging of all carcasses is preferable for the carcass tagging study. Depending on abundance, however, it may be necessary to subsample the number of coho tagged. If the number of coho carcasses precludes completing the survey for the day, field staff may elect to subsample. The selected sampling interval may be determined in the field to ensure completion of all survey reaches. Any subsampling will follow a systematic sampling approach whereby every third or fifth fish is tagged as opposed to every fish. Fish that are not tagged will be marked by removing the tail to prevent double-counting unmarked fish.

Results from carcass surveys will be summarized as capture histories and analyzed using a multi-state model formulation of the JS model to account for loss on capture for fish marked by removing the tail (Kéry and Schaub, 2012). Uncertainty in mainstem spawner abundance will be reported as the range in potential abundance within associated confidence intervals

North Fork Lewis River tributaries below Merwin Dam

Generalized Random Tessellation Stratified (GRTS) sampling is used to identify a spatially balanced set of 1-mile long spawning ground survey reaches for coho salmon. These reaches are split into three panels. All reaches in the first panel are surveyed annually, the second panel is split into three sets of reaches of which one set is surveyed each year on a rotating basis. The final panel is split into nine sets of reaches of which one set is surveyed each year on a rotating basis. The size of the panels is designed such that each year's set of survey reaches is comprised evenly of those visited annually, every third year, and every ninth year.

Spawning ground surveys are conducted according to WDFW's Region 5 spawning ground survey manual (WDFW, 2017a) and coho spawning ground survey manual (WDFW, 2017b). Briefly, each reach is surveyed once per week, environmental conditions permitting, from October 1 to January 31. On each survey, counts are made of: live adult coho, coho redds, and dead coho carcasses. Coho redds are individually identified by flagging, recorded on a GPS unit, and tracked to determine whether they are still visible in subsequent weeks following Gallagher et al. (2007). On each subsequent survey, the number of new redds (those not yet flagged) and old redds (those already flagged AND still visible) can be counted, providing a census in survey areas without double counting redds. Carcasses are sampled for fork length, sex, the presence or absence of an adipose fin and CWT to determine origin. Snouts are taken from CWT-positive fish for CWT recovery. Recovering CWTs provides the information necessary for determining origin and age structure. Finally, counts of live coho are made for each reach on each survey, and individual fish are classified as holders or spawners and the presence of an adipose fin is noted as present, absent, or unknown.

Composition

All carcasses recovered during surveys will be identified as HOR or NOR (when possible). A significant portion of adult returns are collected at the MCF and Lewis Hatchery ladder and thus do not contribute to the number of actual spawners downstream of Merwin Dam. HOR fish are identified by the combination of a missing adipose fin and/or CWT snout tag, while NOR fish are identified by intact adipose fin and no CWT snout tag. Coho salmon releases from the hatchery include double index tag groups that have adipose fins and a CWT. All salmon are wanded to ensure that hatchery double-index tag groups are accurately identified. If a carcass is too far deteriorated to clearly identify the presence or absence of an adipose fin the fish will not be included in the data used to estimate pHOS.

Coho salmon spawning downstream of Merwin Dam and in tributaries include both early (Type S) and late (Type N) stocks. Because both stocks are treated as one population for recovery planning purposes, estimates of pHOS will not differentiate between early and late coho.

In the mainstem Lewis River, seasonal surveys of spring Chinook and coho salmon carcasses are performed from Merwin Dam downstream to the lower end of Eagle Island (details provided in objective 14 for coho and objective 16 for Chinook). Tributary surveys for coho use a GRTS method to define survey reaches (panels) each year. GRTS survey reaches identified each year in the Lewis River tributaries are developed as part of the overall LCR DPS sampling and thus not specific to the Lewis River coho population.

The ratio of HOR to NOR spawners observed in sampled carcasses will be used as an estimate of the pHOS in the total population. Data from weekly abundance surveys throughout the spawning period of coho salmon will be scaled by weekly abundance estimates and pooled to calculate a seasonal pHOS for each species. During weekly surveys, carcasses are recovered and sampled by either walking along the stream margins or by boat. As part of abundance surveys, all carcasses that are directly handled are marked with either a plastic opercle disc (coho) or by removal of the caudal fin (spring Chinook) to prevent double counting.

Data Analysis

Abundance

Data are analyzed in conjunction with spawning ground survey data, mark-recapture data, dam census counts, and fishery harvest estimates for coho salmon from throughout the lower Columbia Evolutionarily Significant Unit (ESU) via a Bayesian multivariate state-space model (Buehrens et al., *in prep*). The primary outputs of this model are estimates of spawner abundance and pHOS. Briefly, for each subunit, counts of uniquely identified redds are summed together with an estimate of the number of redds constructed outside of the survey reaches. This total is expanded to a female coho salmon abundance estimate based on the number of "apparent redds per female" which is estimated in the model based on other subunits in the LCR where paired redd counts and either census or mark-recapture estimates of female abundance are available. The female abundance estimate is expanded to total abundance based on the sex ratio of recovered carcasses in the subunit. The pHOS is then estimated based on the proportion of HOR carcasses recovered in the subunit. Parameters are reported as posterior medians with associated 95% credible intervals. Parameters are reported for subunits as well as for the overall Lewis River coho salmon population. The overall estimate is achieved by summing the subunit abundance of hatchery and wild spawners within the Bayesian model, thereby intrinsically propagating uncertainty in subunit parameters and covariance among them.

Composition

The observed ratio of hatchery origin carcasses to total carcasses will be treated as a binomial proportion and corrected for mis-clip rates.

The observed number of HOR carcasses (S_H) and NOR carcasses (S_N) can be summed after the completion of each week of carcass surveys for each species.

Weekly pHOS estimates will be scaled by time-stratified (e.g., weekly) spawner abundance estimates ($B*$) to account for differences in carcass capture probability that occur with the change in abundance and distribution of the population across the sampling period, as in the following:

$$pHOS = \frac{\sum_{i=1}^{n} B^* \times S_H}{\sum_{i=1}^{n} B^* \times (S_H + S_N)}$$

The pHOS of a given species is defined as the ratio of HOR carcasses to the total number of carcasses, adjusted by the proportion marked (pMark) as in the following:

$$S_H = \frac{S_H^{"marked"}}{pMark}$$

Weekly adjusted pHOS ($pHOS_{adj}$) estimates will be pooled to calculate a seasonal estimate of pHOS by summing numerators and denominators of all weekly estimates for each species.

One possible approach to investigating the increasing or decreasing trends in the pHOS over time could be to use a hierarchical logistic regression model with a fixed effect for a yearly trend and a random effect for each year to account for potential over-dispersion commonly observed in natural systems (Zuur et al., 2009). This approach would be appropriate only after an accumulation of multiple years of data.

The bounds of uncertainty will be estimated with 90% credible intervals (the probability that the estimate lies within the reported interval is 90%) for both the annual estimates of pHOS and for the trend in pHOS over time.

Frequency and Duration

Duration	Frequency
Oct 15 – Jan 31	4 days per week

Deliverables

The following annual estimates will be reported in the Lewis River Annual Operations Report:

- Estimated mainstem spawner abundance based on carcass observations and recovery of tagged carcasses. (mainstem escapement)
- Direct counts of carcasses and redds per week in tributaries
- Estimated spawner abundance and pHOS for tributary units and mainstem Lewis River (estimated medians with 95% credible intervals)
- Fork lengths, sex, origin (HOR, NOR), and CWT presence for all carcasses directly sampled
- Weekly and seasonal (pooled) estimate of mainstem pHOS by species
- Estimate of change in pHOS over time annually
- For coho salmon, tributary pHOS will also be reported from GRTS reaches surveyed annually

Limitations and Specific Concerns

- The ability to identify redds accurately to species is complicated by the relatively large number of fall Chinook that overlap with both species in terms of distribution and redd construction.
- Not all carcasses are available for sample because they are either too deep to be seen, have been removed by predators, or have deteriorated and the presence of an adipose clip cannot be confirmed. Carcass capture efficiency is low in the mainstem Lewis River and sampling effort is lower in tributaries compared to the mainstem.
- Methods estimate abundance while accounting for unobserved fish, providing a higher level of confidence in the measure of abundance of both HOR and NOR fish compared to relying on direct observations.
- If a relatively large portion of carcasses in the population are sampled in a given year (greater than 20%), then the error associated with the estimate of pHOS described above can be described as overly conservative.
- Currently the overall Lewis River coho estimate only contains tributary subunits 3 and 4, which WDFW is responsible for analyzing. However, in the future when robust estimates of abundance and pHOS are available for subunits 1 and 2, these should also be incorporated to estimate overall pHOS for Lewis River coho salmon.
- Capture probability for hatchery and natural origin carcasses is identical unless directly or indirectly accounted for in the abundance estimator.
- Fish First coho salmon reared for the Remote Site Incubator Program are unmarked and if recovered would be erroneously counted as NOR.

Strategy B: Adult Spatial and Temporal Distribution

Strategy B: Determining Spatial and Temporal Distribution of Adult Spawning Winter Steelhead, Chinook, and Coho Downstream of Merwin Dam

I. Winter Steelhead

Introduction

Redd surveys (by boat) will be used to assess spatial and temporal distribution of spawning winter steelhead. Surveys will follow the same methods described under winter steelhead adult abundance in Strategy A. To evaluate spatial distribution, redd data will be summarized using a map depicting the location of each new redd (using handheld GPS) observed within the study area for each season. The study area will be delineated by five reaches used to evaluate abundance (see Strategy A). Based on the number of new redds observed, an estimate of redds per mile will be provided for each of the 5 reaches. To estimate temporal distribution, each redd will be time stamped to calculate the number of total redds by period including the distribution of those redds by period to assess whether annual shifts in distribution (redds per mile by reach) are occurring.

Study Area

The study area begins at the float buoy line immediately downstream of Merwin Dam and the downstream end of Eagle Island. Both channels of Eagle Island are surveyed (Figure H-1),

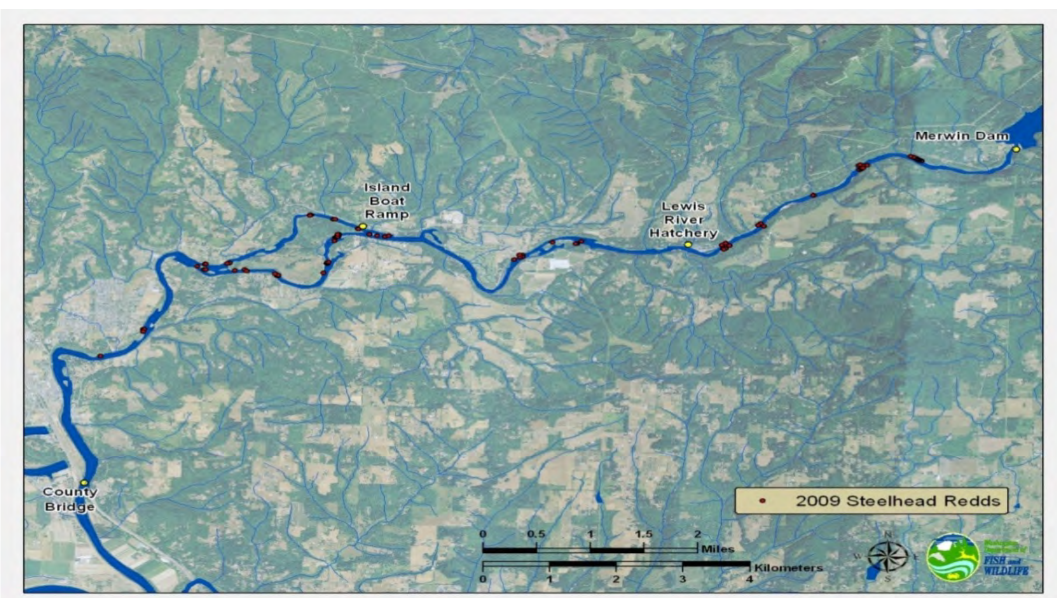


Figure H-1. Example of data collected during 2009 winter steelhead redd surveys on the North Fork Lewis River indicating redd locations and survey area

Frequency and Timing

Redd surveys are conducted regularly (every 7 to 10 days) throughout the known spawning season, which, based on past observations, is from April 1 to June 30. Surveys are intended to provide a census count of all redds constructed within the study area. Winter steelhead redd surveys may begin earlier if spawning activity is observed. Surveys will not be performed if visibility is less than 3 feet or spill has been initiated from Merwin Dam. By marking redd locations via GPS, flagging, and detailed notes, double counting of redds is minimized and a total count of unique redds can be developed.

II. Coho Salmon

Introduction

For the mainstem North Fork Lewis River downstream of Merwin Dam, it has been difficult to determine whether observed redds were constructed by Chinook or coho as both species overlap in spawning and it is likely that some level of superimposition by both species occurs. Thus, mainstem coho redd surveys are not a reliable estimator for coho distribution (or abundance) metrics. Until a reliable distribution estimator is available for coho spawning in the mainstem, distribution reporting will rely on live counts classified as either holders or spawners, and carcasses classified by sex and origin for each of the five reaches surveyed.

Distribution of coho spawners in tributaries of the North Fork Lewis downstream of Merwin Dam will continue to rely of traditional redd surveys and live counts to report spatial and temporal distribution for those tributaries selected under the GRTS sampling design. The total number of carcasses marked and recovered in tributaries will also be reported as part of the spatial and temporal distribution objective.

Spawner survey methods and data analyses are detailed in Strategy A.

Frequency and Timing

Surveys for both mainstem and tributary sampling will occur every 7-10 days from mid-October through January to capture both early and late spawners.

For tributary surveys, redd surveys will be conducted as part of each survey. The total number of new coho redds will be reported for each tributary reach sampled.

Deliverables

Mainstem:

- Table reporting the total number of live holders and spawners, and total number of carcasses marked and recovered by sex origin and delineated by reach.

Tributaries:

- Table reporting the total number of redds, live holders and spawners, and number of carcasses by sex and origin for each tributary reach surveyed.

Limitations and Specific Concerns

- Accurate identification of redds and live holders and spawners depends on both the proficiency of the surveyor and river conditions (namely turbidity) on a given survey day. Poor visibility may be prolonged during winter surveys. This may prevent the identification of redds or live coho leading do data gaps.
- The ability to accurately identify species specific redds between coho and fall Chinook is challenging because both species share the same spawn timing. There are specific redd characteristics that differ between species such as substrate size (gravel vs cobble), location of redds (margins versus thalweg), and size of redds. However, these characteristics may also overlap based simply on differences in fish size. For example, smaller Chinook may select smaller substrate size preferred by coho or superimpose over existing coho redds. It must be noted that some (unknown) level of uncertainty exists in identifying redds in general, and this uncertainty is exacerbated by having to differentiate redds by species.

III. Chinook and Chum Salmon

Introduction

In addition to carcass surveys, counts of live fish and redds will be conducted to determine the spatial and temporal distribution of adult Chinook and chum. Based on historical information, counts will coincide with the presumed peak spawn time period for both tule and bright fall-run Chinook. Live fish will be enumerated by section while redds will be delineated at a finer spatial resolution using counters and a handheld GPS. Redds will be assigned to a species based on the species of fish that is seen either on or around the redd. If a live fish is not seen on a redd, the redd is assigned to the most likely species based on the spawning habitat, size of redd, and previous knowledge of spawning distribution within the Lewis River.

Spatial and temporal distribution of Chinook and chum abundance will be described using "standard" metrics (e.g., median and standard deviation) used for abundance. Specifically, the temporal distribution of spawner abundance will be described by calculating the median spawn date using weekly derived estimates of abundance (i.e., B*) from the JS model by race (tule, bright) and origin (hatchery, wild). Additionally, a cumulative distribution plot of abundance will be generated to depict the temporal distribution of spawning. The spatial distribution of abundance will be described through the use of redd counts.

Study Area

The survey area is divided into five river sections: sections 1 to 4 extend from Merwin Dam (RM 19.1) to Lewis River Hatchery (RM 15.7) and section 5 extends from below Lewis River Hatchery to below Eagle Island (RM 10).

Frequency and Duration

Up to three counts will be conducted for tule Chinook in October (generally the second, third, and fourth weeks of October) and up to three counts will be conducted for brights in late November to early December (generally third and fourth weeks of November and first week of December). Live surveys will also be conducted weekly or biweekly in September and early October to correspond with the peak spawn period for spring Chinook. Counts will be performed by surveyors in a boat. One to two boats will be used per survey.

Assumptions and Limitations

Estimates of temporal distribution are based on estimates of adult abundance. Therefore, the assumptions that are outlined in Strategy A apply here and must be met to generate unbiased estimates of temporal distribution.

Descriptions of spatial distribution are based on peak redd count data. Therefore, one critical assumption is that the spatial distribution of adults during peak spawning is representative of the spatial distribution throughout the entire run. Additionally, redds must be identified correctly. Redd identification and counts can be difficult due to poor survey conditions, superimposition, or possible overlap in spawn timing with that of another species. These potential sources of error will be minimized by conducting redd counts on days with highest visibility, proper training of surveyors, and maximized use of experienced surveyors.

Deliverables

Redds will be enumerated by section and reported in table format. Tables will be formatted to provide a temporal count by section and estimate of redds per mile. Additionally, maps will be generated depicting counts of individual redds (via GPS data). Moving forward, thought should be put into what metrics of spatial and temporal distribution will be the most biologically meaningful with regards to conservation and recovery of populations within the Lewis River.

Strategy C: Juvenile Abundance and Migration

Strategy C: Screw Trapping Strategy for Lewis River Hatchery Programs

INTRODUCTION

Screw trapping in the Lewis River below Merwin Dam has been conducted as a component of addressing objectives in the Lewis River Hatchery and Supplementation Plan to evaluate populations of Chinook, Coho and steelhead. Since 2013, 8-foot screw trap(s) have been deployed near the Lewis River Golf Course (RM 13.4) to estimate the abundance and timing of juvenile salmon and steelhead passing the trap. Beginning in 2015, two traps were deployed in-tandem to increase the number of juveniles captured. While the additional trap improved the number of juveniles captured, seasonal capture efficiency remained between one and two percent for the period 2013 -2018. In 2020, the tandem trap was separated and two single traps were operated at two sites – the existing Golf Course location (upper) and a site approximately 1.25 km downstream of the existing site (lower). Results from 2020, suggest that separating the traps did not improve capture efficiency and the ATS agreed to deploy the tandem trap configuration again, beginning in 2021. However, the traps were fished downstream about 100 meters from the original location prior to 2019. By moving the trap location downstream, it is thought that capture efficiency may improve due to low flow characteristics during June and July.

Figure 1. Trap location relative to the Lewis River and Merwin hatcheries

STUDY OBJECTIVES

Screw traps are used to inform management and meet program objectives related to smolt abundance, migration timing, species composition (including origin), and smolt morphology (size and condition). Screw traps also have the potential to estimate residualism rates of hatchery released smolts when release strategies incorporate unique marking strategies combined with in-river smolt collection activities upstream of the traps. Residualism will not be addressed using the screw traps; the ATS will continue to develop study designs and specific methods necessary to address this key question for implementation in the future.

In 2022, screw traps will be used to meet the following study objectives and selected key questions of the Hatchery and Supplementation Program:

Objective 4: Determine whether hatchery production protocols incorporate best available management practices to support program targets and goals.

Relevant Key Questions

4B: Does the migration rate of HOR juveniles result in overlap with NOR juveniles or spawning adults?

Objective 6: Estimate juvenile outmigrant abundance for late winter steelhead, coho, and Chinook downstream of Merwin Dam

Relevant Key questions

6A: Are estimates of NOR juvenile outmigrant abundance unbiased and meeting precision targets?
6B: Is the abundance of NOR juvenile outmigrants by species and outmigration year increasing, decreasing, or stable?
6C: What are the morphological characteristics of outmigrating NOR juveniles relative to their conspecific HOR juveniles?

SAMPLING DESIGN

FIELD METHODS AND DATA COLLECTION

Trap Description:

The mainstem Lewis River juvenile traps consists of two standard 8-foot cone rotary screw traps (EG Solutions). Both traps are equipped with double pontoons to improve safety, stability and reliability of the traps. Traps are equipped with solar powered amber flashing hazard lights placed on the starboard and port sides of the trap. Reflective hazard signs are also placed on the traps warning boaters to not trespass.

Site Selection

The juvenile trapping site is situated along a gradually deepening thalweg on the south bank. The site is fishable over a broad range of flows and can be adjusted downstream along the thalweg as flows decrease. The site is currently permitted to anchor the traps to existing LWD habitat structures upstream of where the traps operate. This site meets permitting safety requirements for navigable rivers with installed hazard signs upstream and downstream of the trap location and solar powered hazard lights installed on the port and starboard sides of the trap.

The site is located downstream of Merwin and Lewis River hatcheries, Cedar Creek, and the *majority* of available spawning areas for Chinook, coho and winter steelhead. Based on winter steelhead redd surveys conducted in 2015, approximately 96 percent of redds were identified upstream of the Golf Course location (PacifiCorp and Cowlitz County PUD, 2016). For Chinook and Coho salmon, past redd and carcass surveys indicate that spawning occurs predominately upstream of Lewis River Hatchery (WDFW reaches 1 – 4).

However, the site does have limitations, especially when attempting to achieve unbiased estimates. These limitations include a relatively short trapping duration (March through June), lack of funneling action by slow river velocity, which may result in trap avoidance behavior by yearling migrants contributing to low overall trapping efficiency. Trapping efficiency for larger yearling smolts (e.g., Coho) tends to be relatively lower in May and June which corresponds with relatively lower flows during this period.

The ATS will continue to plan and develop measures to address limitations in future trapping efforts. To address the relatively low trapping efficiency, the ATS has been exploring measures to increase the number of marks available for capture. A few of the potential ideas included the use of marked hatchery smolts released from the hatcheries, the use of Cedar Creek trap captures to provide additional marks for NOR smolts, and additional efforts to mark more smolts at the Golf Course trapping locations. However, these measures are unlikely to improve trapping efficiencies during low flow periods in late May and June.

Frequency

Traps will be checked every morning (7 days per week). Additional trap checks may occur during high flow events for cleaning of debris from the trap box and to ensure integrity of the traps. Additional checks may also be warranted during periods of peak outmigration (e.g., during hatchery volitional releases).

Trapping Schedule

Location	Trapping Period
Golf Course trapping site	March 1 – June 30, 2022

FISH HANDLING

All fish will be removed from the trap live box prior to the cones being lifted. Dip nets will be used to transfer all fish from the live box to covered blue or green 5-gallon buckets with battery powered aerators. Non-salmonids will be transferred to separate (non-salmonid) buckets for species identification, counting and release downstream of the trap. Prior to biological sampling, all salmonids will be anesthetized in a solution of 1 ml Aqui-S® to 2 gallons of river water. Depending on species and life stage, fish shall be scanned for coded or blank wire tags or PIT tags (Table 2). After sampling, salmonids will either be released downstream of the trap or marked and released upstream for efficiency trials.

Table 1. Tag scanning requirements for each species and life stage of fish collected at the traps

Species	Life Stage	CWT or BWT Scanning	PIT tag Scanning*
Chinook	All	☒	☒
Chum	All		
Steelhead	Fry and Parr		☒
	Transitional, Smolt	☒	☒
Coho	Fry and Parr	☒	☒
	Transitional, Smolt	☒	☒
All	Adults (inc. residuals)	☒	☒

* PIT tagged juveniles released from the Woodland Release Ponds may be trapped

Species identification

All salmonids will be identified to species by experienced samplers following WDFW region 5 juvenile identification protocols.

Life stage assignment

Salmonids shall be classified into five separate life stages. These categories include fry, parr, transitionals, smolts, and adults. When possible, these categories are further separated into age classes (subyearling and yearling+). A decision tree developed by WDFW for smolt trapping operations is provided as part of Attachment B of this plan. All salmonids < 45 ml FL are considered fry and all adults are salmonids > 500 ml FL. For salmonids between 45 – 500 ml FL, life stage shall be assigned based on morphological characteristics as described in the WDFW decision tree. The decision tree is intended to provide guidance and standards to samplers, however, this guidance may be adjusted based on site (or Lewis River) specific information such as capture of BWT steelhead migrants or residuals or smolt collections at the Swift FSC.

Length measurements: Up to 50 samples per species (Chinook, Coho and steelhead) and per origin group (HOR and NOR) will be measured to fork length. Cutthroat trout will also be measured for informational purposes.

Origin Assignment

Maiden captures may have a combination of visual (fin clips) and nonvisible (CWT, BWT or PIT) marks or tags (Table 2). All maiden fish must be scanned for nonvisible tags to determine correct origin of each capture. For coho and Chinook, a portion (about 150,000 smolts) of the hatchery release is a double-index tag or DIT group. This group is identified by having an unclipped adipose fin, but with a coded wire tag (CWT) present in their snout. Late winter steelhead released from Merwin Hatchery are identified are also identified by having unclipped adipose fins, but with a blank wire tag (BWT) present in their snout. Table 2 provides all hatchery marks that should be expected and accounted for during fish sampling.

Table 2. Summary of hatchery marks groups used for each species handled at the hatcheries

Species	AD Clip Only	AD Clip + CWT	CWT Only	BWT	PIT
Coho Salmon	☒	☒	☒		
Spring Chinook	☒	☒	☒		
Fall Chinook		☒			
Late Winter Steelhead*				☒	☒
Chambers Winter Steelhead	☒				

* Late winter steelhead residuals may have a PIT from previous tangle net capture. All adults inadvertently captured should be sampled for PIT tags and released downstream of the trap.

Marking Strategies

Not all juvenile salmonids captured in the traps may be migrants. This is most often observed with residual steelhead and precocious spring Chinook released from the hatchery. The designation of migrant vs. non-migrants depends on the species and life stage assignment as described in the decision tree (Attachment A). Chinook, Coho and steelhead designated as migrants, regardless of origin, shall be marked and released upstream of the trap for efficiency testing of the trap (Table 3). Salmonids not listed in Table 3 (cutthroat, sockeye, etc.) shall not be marked for efficiency trials and released downstream of the trap. These species however, shall be enumerated and measured to fork length.

Table 3. Migrant Status by life stage assignment and requirements for efficiency trials

Species	Life Stage	Migrants	Non-Migrants	Efficiency Trials
Chinook	All	☒		Yes
Chum	All	☒		No
Steelhead	Fry		☒	No
	Parr, Transitional, Smolt	☒		Yes
Coho	Fry		☒	No
	Parr, Transitional, Smolt	☒		Yes
All	Adults (inc. residuals)			No

Fish that are classified as migrants and at least 50 mm FL are marked with Alcian blue tattoo dot(s) using a panjet tool. To identify different marking groups upon recapture, the location and number of panjet marks shall vary weekly to differentiate specific release groups (e.g., week 1, 2 …).

Marking migrant fish smaller than 50 mm FL is accomplished by immersing fish in a solution of 0.4 g of Bismarck brown per 4 gallons of aerated river water for one hour. Because this is a batch mark and cannot be used to differentiate (weekly) groups, Bismarck brown marking will occur no more than once every 3 days to allow newly marked fish time to pass the trap before another batch is marked. The frequency of batch marking may be modified by the ATS.

<u>Recovery</u>

All sampled and marked fish will be allowed to recover in totes or buckets prior to transport upstream for release. Any mortalities will be removed and documented to species, origin and life stage. All carcasses will be disposed of downstream of the screw trap.

<u>Release Strategy</u>

Transport of marked groups will occur after sampling is complete. Marked fish will be transported by boat upstream approximately 0.85 RM upstream (swirly hole) and released. All other fish will be released in the riffle immediately downstream of the trap.

<u>Trap and environmental data</u>:

Cone revolutions rate (revolutions per minute) will be measured each trapping day. Cone revolutions will be compared to flow data obtained from the USGS river gage site and available on the internet at https://waterdata.usgs.gov/wa/nwis/uv?site_no=14220500). River water temperature data will be obtained daily by direct measure during fish sampling. Water temperature of holding totes or buckets should also be reported daily.

Documentation of any mortalities or injuries:

All mortalities or injuries observed must be documented as part of the Biological Opinon for Lewis River hydroelectric operations. Each mortality, whether grouped or individually, shall provide a count of mortalities, species, origin, life stage and the probable or known cause of the mortality. Injuries should also be noted, especially if the trap is suspected for a particular injury (e.g., fresh wounds, descaling).

Trap outages (planned or unplanned):

All outages regardless of planned or unplanned will be documented. Any planned outages will describe the date and time of the outage, duration and reason for the outage. Unplanned outages (e.g., mechanical failure or damage) will be documented as best as possible with respect to time and duration. Efforts to place the trap back in services will be done as soon as practical to minimize the number of missed days. A catastrophic event (e.g., loss of trap or extensive damage requiring removal of the trap) will require consultation with the ATS and ACC if a replacement trap is not available.

Fish Counts:

A total count of all fish captured will be recorded each day (or for each trap collection). The total number of fish captured will be delineated by species, life stage, capture type (maiden or recap) migrant status, origin, previous marks and type, mark type given (if applicable), release location and time of release (for efficiency trials).

Analytical Methods

Estimates of total outmigrants passing the screw traps follow the same methods for estimating the migration timing and number of juveniles entering Swift Reservoir in the Revised Aquatic Monitoring and Evaluation Plan for the Lewis River (PacifiCorp and Cowlitz PUD 2017). Total weekly juvenile outmigration by species during the trapping season is calculated using the formula for use of a single partial trap described in Volkhardt et al. (2007). Though two screw traps are operated in tandem (to increase overall efficiency) the traps are treated as one trap in making the total outmigrant estimates. In addition, total season variance and confidence intervals are also estimated using bootstrap methodology for each focal fish species total estimate (Thedinga et al. 1994).

When performing the Volkhardt et al. (2007) method calculations, there is discretion in defining the trap efficiency actually used to make the weekly abundance estimates. Weekly abundance is variable and the number of fish available for efficiency trials can be low one week, but high in another. When weekly flows are similar, and actual mark-recapture data are lacking or sparse for that particular week, we combine multiple weeks' mark-recapture samples to increase the sample size and apply the combined trap efficiency to each week in that pooled trap efficiency calculation.

TRAP EFFICIENCY AND TEST-FISH RELEASE POWER ANALYSIS

The number of recaptures of marked fish, which are released upstream to test trap efficiency, influences confidence interval size around estimates of total out-migrants. The actual trap efficiency also has a large influence on the confidence interval size. To assess the influence of these two attributes on confidence interval size, a power analysis is provided to estimate the number of fish needed to mark and release based on trap efficiency (Figure 2). Estimates of trap efficiency in 2017 was on average 1.1%. This value represents the recapture rate of all marks, which was 35 out of 3,181 marks. The relatively small recapture rate affects confidence in the estimate. For example, average confidence in the 2017 estimates was ± 40% of the estimated abundance at the 95% confidence level.

Using total coho captures from the 2013 season (901 fish), estimates of total coho out-migrants were generated assuming true trap efficiency was 0.6%, 1.0%, and 2.0%. For each trap efficiency level, total number of test-fish released upstream was varied in a step-wise manner, starting at 160 up to 4,500 fish. For each trap efficiency and test-fish release level, total coho out-migrant and variance estimates were generated using the Bootstrap method (Thedinga et al., 1994). Confidence intervals were calculated for each out-migrant estimate based on the variance and transformed into a percentage of the total estimate, in order to compare confidence interval size between varying trap efficiency levels.

The relationship between true trap efficiency, the number of test-fish released upstream, and how these two attributes influence confidence interval size around estimates of total out-migrants is depicted in Figure 2. As true trap efficiency decreases, more test-fish must be released upstream in order to re-capture at least 15 to 20 fish. As trap efficiency and/or the number of test-fish released decreases, the size of the confidence interval around total out-migrant estimates dramatically increases. Based on this analysis, if the goal is to generate total out-migrant estimates with confidence intervals less than ±50% of the total estimate and CV less than approximately 30%, many more test-fish need to be released in the future, assuming a similar level of total trap captures as occurred in 2013. Based on this analysis, it is recommended to release at least 3,000 test-fish if true trap efficiency is 0.6%; release at least 2,000 test-fish if true trap efficiency is 1.0%; and release at least 1,000 test-fish if true trap efficiency is 2.0%. Releasing more test fish will further reduce confidence interval size and improve out-migrant estimates.

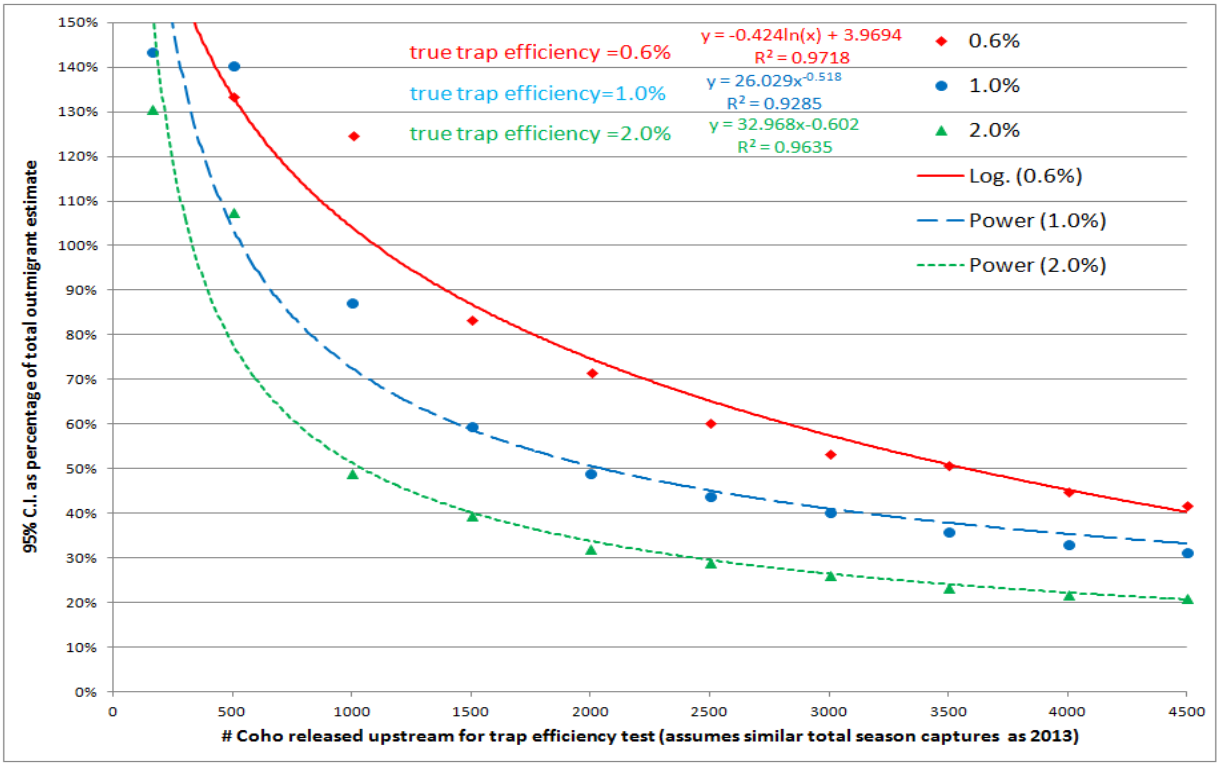


Figure 2. Power analysis for the number of smolt released upstream of screw trap to achieve corresponding confidence interval at the 95% CL.

Data Management

All screw trap collection data will be formatted and provided to WDFW staff for upload into the JMX database. Specific QA/QC measures are not described here; the contractor will be required to QA/QC the raw data and provide those data in a format that can be applied to analyze and answer specific questions that may surface during annual ATS planning. While data is provided to WDFW for their JMX database, the data shall be the property of the Utilities, and therefore, the Utilities are responsible for storing the data files for future reference and reporting results to FERC on an annual basis.

Assumptions

Assumptions identified below are common to all mark-recapture studies. The ability to test some assumptions, particularly equal mixing and survival between marked and unmarked groups are difficult. WDFW has conducted assumption testing at other nearby screw trapping sites (e.g., Cowlitz and Coweeman Rivers) and these results are useful in determining where efforts in the North Fork Lewis River should be directed to ensure specific assumptions are met.

1. **Closure - the population is geographically closed to immigration, emigration, births, and deaths.**
   In closed populations, the number of migrants (N), or proportion of marked and unmarked migrants remains constant between mark and recapture events.

- *Are there any trap failures causing the traps to not operate resulting in a proportion of the population passing the trap to not be captured?*
- *Does the trap operate throughout the migration period?*
- *Are only migrants marked?*

2. **No mark or tag loss.**
If marks are lost, or samplers fail to identify marks accurately, recaptures will be undercounted resulting in overestimating abundance.

- *Are all marks or tags identified and reported accurately upon recapture?*
- *Does dye batch marking persist long enough to accurately identify upon recapture?*

3. **No mark-related mortality** – if marked fish do not survive as well as unmarked fish, recaptures will be undercounted resulting in overestimating abundance.

- *Does marking influence survival (e.g., from direct injury or increased predation)*

4. **Equal catchability – all fish have equal probability of being marked in the first event.**

5. **Equal catchability – marked fish mix completely with unmarked fish between sampling events** – If recapture rates are not representative of the proportion of marked and unmarked migrants, recapture rates will be biased resulting in over or under estimating abundance

- *Does the release site provide adequate spatial separation from the traps to provide for equal mixing?*

Deliverables

An annual reporting of trap operations is required describing and analyzing capture data. The report, at a minimum, should include the following:

1) Weekly outmigrant abundance and trap efficiency for the current trap year by group (e.g., species, origin and life stage) including estimates of total abundance
2) Annual time series of outmigrant abundance for the trap (by year when available)
3) Annual time series of coefficient of variation for the trap (by year when available)
4) Daily average stream flow (cfs) and daily measured cone revolution rate
5) Summaries of trap captures, recaptures and marks released
6) Format and submit data to JMX database
7) A project assessment including the following:
    - Describe whether precision goals were met and, if not, why

- Describe any specific concerns regarding the data or estimates or concerns due to bias
- Provide any recommended improvements that can be implemented in the future

8) Description of all trap outages including the cause of the outage and duration of the outage (in hours). Also, describe any omissions of data for any period and justification for any omissions or other modifications to the raw data prior to analysis

1.1.1.1. Limitations and Specific Concerns

1. Low numbers of captures (for marking), and recaptures of those marks particularly for steelhead and Chinook can result in in both biased and imprecise estimates of abundance Pooling of data are accepted in this plan to increase the sample size and power of our estimates, however, this method has been rejected by WDFW in favor of attempting to achieve an unbiased estimate with low precision.

In past years, it has been difficult to mark, and subsequently recapture, enough juveniles to generate unbiased estimates of abundance. To increase the number of marks, the ATS may evaluate whether the following methods are practical for implementation:

- The use of marked hatchery releases as a surrogate for mark recapture purposes. This may include PIT tagging of hatchery fish combined with a fixed PIT tag antenna at the hatchery to determine river entrance timing to estimate travel time between the hatchery and screw trap as well as developing migration curves during volitional releases.
- Increase the mark rate at the screw trap to increase the number of total marks available for recapture
- Use of Cedar Creek screw trap captures to increase the number of NOR marks available for recapture

2. Behavior or migration rates of hatchery releases between the traps (RM 13.4) and the mouth of the Lewis River cannot be described by the screw trap catches alone.

3. This study assumes that fish will migrate past the trap during deployment. However, with any hatchery program there is a proportion of nonmigrants. To use the screw traps in estimating residualism rates, more work is needed to quantify the number of fish residing (e.g., nonmigrants) upstream of the trap post-release. The specific timing of when marked fish enter the river (and are available for capture) during a volitional period is also important depending on the duration of the volitional release window.

4. Trapping efficiency may not be the same between HOR and NOR smolts due to differences in size or behavior (Assumption No. 3)

5. The migration window of hatchery fish is typically very small (i.e., a few days) and if the traps are not in operation due to mechanical failure, an entire migration window may be missed.

6. Trapping is inherently difficult in larger rivers due to debris load, variable flows (including spill) and mechanical failures – most of which are unavoidable consequences of operating floating traps in large rivers.

Attachment A - JMX Smolt Trap Protocols and Reporting

Part 1: DATA COLLECTION PROTOCOLS

Protocol Name: North Fork Lewis River (downstream of Merwin Dam) - 2020

Project Supervisor: Jason Shappart (Meridian Environmental, Inc. – PacifiCorp contractor)

Science Leader: Jason Shappart (Meridian Environmental, Inc. – PacifiCorp contractor)

ESA Take Permit No. (if applicable): ESA Section 7(a)(2) Consultation, Biological Opinion for PacifiCorp's operation of the Lewis River Hydroelectric Projects (NMFS Consultation No. 2005/05891). August 27, 2007.

Trap information:

Trap Name	Type of Trap	Trap Location RKM	Start Date (Planned)	End Date (Planned)
Lower Lewis River Trap (Golf Course)	8-foot rotary screw trap	21.6	03/01/2020	06/30/2020
Lower Lewis River Trap (GC Boat Ramp)	8-foot rotary screw trap	20.1	03/01/2020	06/30/2020

1.1 Field Objectives:

Trap Name	Lower Lewis River Traps – Golf Course and GC Boat Ramp							
Species	**Origin**	**Life Stage**	**Age Class**	**Catch**	**Efficiency Trials**	**Fork Length**	**Scale s**	**Other**
Chinook	All	F/P/T/S	Subyearling/ Yearling	Y	Y	Y	N	Scan for CWT
Chum	All	F	Subyearling	Y	N	Y	N	
Steelhead	All	F	Subyearling	Y	N	Y	N	
Steelhead	All	P/T/S/R	Subyearling/ Yearling	Y	Y*	Y	N	Scan for BWT
Coho	All	F	Subyearling	Y	N	Y	N	Scan for CWT
Coho	All	P/T/S	Subyearling/ Yearling	Y	Y*	Y	N	Scan for CWT
Cutthroat	All	T/S/A	All	Y	N	Y	N	
Additional Comments/Narrative: Fry (f), Parr (P), Transitional (T), Smolt (S), Adult (A), Residuals (R), Blank Wire Tag (BWT), Coded Wire Tag (CWT). * steelhead and coho parr have been caught at the Swift FSC as outmigrants.								

1.2 Site Selection:	
• *Why was this site selected for the smolt trap?*	Anchoring, permitting, laminar flow, ease of access and downstream of the majority of spawning
• *Are there spawner estimates above the trap site that can be used to estimate freshwater productivity, capacity, and smolt-to-adult return?*	Mainstem Chinook, Coho and Steelhead – Yes Chum - No
• *Describe the method used for adult escapement estimates (e.g., Carcass tagging, adult MR, AUC, redds, PCE, other).*	Chinook and Coho– Carcass tagging in mainstem NF Lewis River, WDFW GRTS redd surveys in tributaries Steelhead – Redd Surveys
• *Estimated % of the total basin-specific population that spawn above the trap. Include source for this information (% can be a range).*	Steelhead: > 90% (Annual Operations Report) Chinook: unknown, but majority Coho: unknown, but majority
• *Estimated % of yearling life stage juveniles that continuously rear) above the trap (summer and winter) prior to outmigrating. Include source for this information.*	
• *Additional Information*	Juvenile anadromous fish transported from upstream of the Lewis River Projects are released downstream of the trap locations (spring Chinook, steelhead, coho, cutthroat trout)

1.3 Collection Event:	
• *Describe the planned frequency for enumerating and sampling fish caught in the trap.*	Traps to be checked daily (between 09:00 and 15:00 hours)
• *Describe and explain any planned trap outages.*	None
• *Describe process of handling and anaesthetizing fish.*	Dip nets used to transfer all fish to buckets or bins with battery aeration units. Salmonids to be anesthetized in

	solution of 1 ml Aqui-S to 2-gallons river water prior to sampling.
• *Describe method for measuring rotation per minute (rpm)*	Visually for 1 minute (Daily)
• *List flow gauge associated with the trap.*	Ariel Gage Station – 14220500 (USGS)
• *Describe method for measuring visibility and frequency of measurements.*	Not estimated
• *Describe method for measuring stream temperature and frequency of measurements.*	Handheld thermometer placed in-river during daily sampling
• *Describe additional environmental variables measured, the method for the measurement, and the frequency of measurements.*	None

1.4 Fish Count by Group and Individual Measures:	
• *Life stage will be assigned according to the Region 5 Decision Tree (see appendix). Note any exceptions to the Decision Tree for species/life stage. Exceptions need to be approved by your Science Leader in advance!*	Life stage is assigned based on Region 5 Decision Tree, with the exception that all chum are assigned as fry.
• *Describe how origin is assigned.*	Combination of presence and absence of adipose fin clips and CWT or BWT snout tags
• *Describe the characteristics of individual fish (species/life stage, condition, and mark status) that are sorted and released downstream of the trap.*	All species/life stages listed in Table 1.1 that do not have efficiency trials (Efficiency Trials = N) are released downstream of the trap. All other fish not included in Table 1.1, recaptures and fish with visible injury or lethargy to be released downstream of trap. Fry not batch marked on certain days will also be released downstream of trap
• *Describe that characteristics of individual fish (species/life stage, condition, and mark status) that are selected for efficiency trials.*	All Chinook regardless of life stage; all parr greater than 50 mm, transitional and smolting steelhead and coho. Fish with visual injury or other impairment shall not be used for trials regardless of species or life stage)

Table 1-4A. Date and length criteria used for field calls of Chinook age classes.

Life Stage	Age Class	Date Range	Length Range (mm FL)	Phenotype
Fry	Subyearling	3/1 to 6/30	≤ 45 mm	Determined by using Region 5 decision tree based on physical appearance
Parr/Trans/Smolt	Subyearling/ Yearling	3/1 to 6/30	≥ 45mm	
Individual Fish Measures:				
Sample rate for fork length	*F – 10 per day; P/T/S – up to 50 per day per each category*			

Table 1-4B. Date and length criteria used for field calls of Coho age classes.

Life Stage	Age Class	Date Range	Length Range (mm FL)	Phenotype
Fry	Subyearling	3/1 to 6/30	≤ 45 mm	Determined by using Region 5 decision tree based on physical appearance
Parr/Trans/Smolt	Subyearling/ Yearling	3/1 to 6/30	≥ 45mm	
Individual Fish Measures:				
Sample rate for fork length	*F – 10 per day; P/T/S – up to 50 per day per each category*			

Table 1-4C. Date and length criteria used for field calls of Steelhead age classes.

Life Stage	Age Class	Date Range	Length Range (mm FL)	Phenotype
Fry	Subyearling	3/1 to 6/30	≤ 45 mm	

Parr/Trans/Smolt	Subyearling/ Yearling	3/1 to 6/30	≥ 45mm	Determined by using Region 5 decision tree based on physical appearance
Individual Fish Measures:				
Sample rate for fork length		*F – 10 per day; P/T/S – up to 50 per day per each category*		

1.5. Marking and Release:	
• *Explain purpose of applying marks or tags to fish prior to release (if applicable).*	Marks are used to estimate trap efficiency
• *Describe the schedule for which fish will be released to determine trap efficiency.*	Daily, seven days per week
• *Describe the target number of fish for each release group (species/life stage/age class).*	For all species in which outmigration estimates are planned (Chinook, coho, and steelhead) all captured fish in good condition are marked and used in efficiency trials
• *Describe marking or tagging method used for each species/origin/life stage/age class.*	**For Chinook fry**: Bismarck brown dye (Product # 861111, Sigma-Aldrich, St. Louis, MO). Use 0.4 grams of dye per approximately 4 gallons of water. **For all maiden capture salmonids (>50 mm FL),** Caudal marks rotated (upper/lower) on a weekly basis, and tattoo marks varied by week.
• *Describe release location for efficiency trials (rkm).*	In pool/run with bank habitat structures located at N45.937741, W-122.644367 about 0.85 miles upstream of the upper trap site
• *Describe where and how long marked or tagged fish are held prior to release for efficiency trials.*	Marked fish are held aeriated buckets for recovery after sampling and released immediately after each trap is sampled
• *Describe what time of day marked or tagged fish are released for efficiency trials.*	Between 0900 and 1500 hours – depending on the number of fish sampled each day.
• *Describe plans to evaluate mark retention and mark-related mortality.*	None, the ATS may recommend periodic testing to estimate dye retention

• *Describe plans to evaluate mark-recapture assumption that the second sample is a random representative sample (i.e., marked and unmarked fish are completely mixed)*	(Standardized methods will be developed and distributed – but not this year)

Table 1-5. Marking Plan for Trap Efficiency Trials

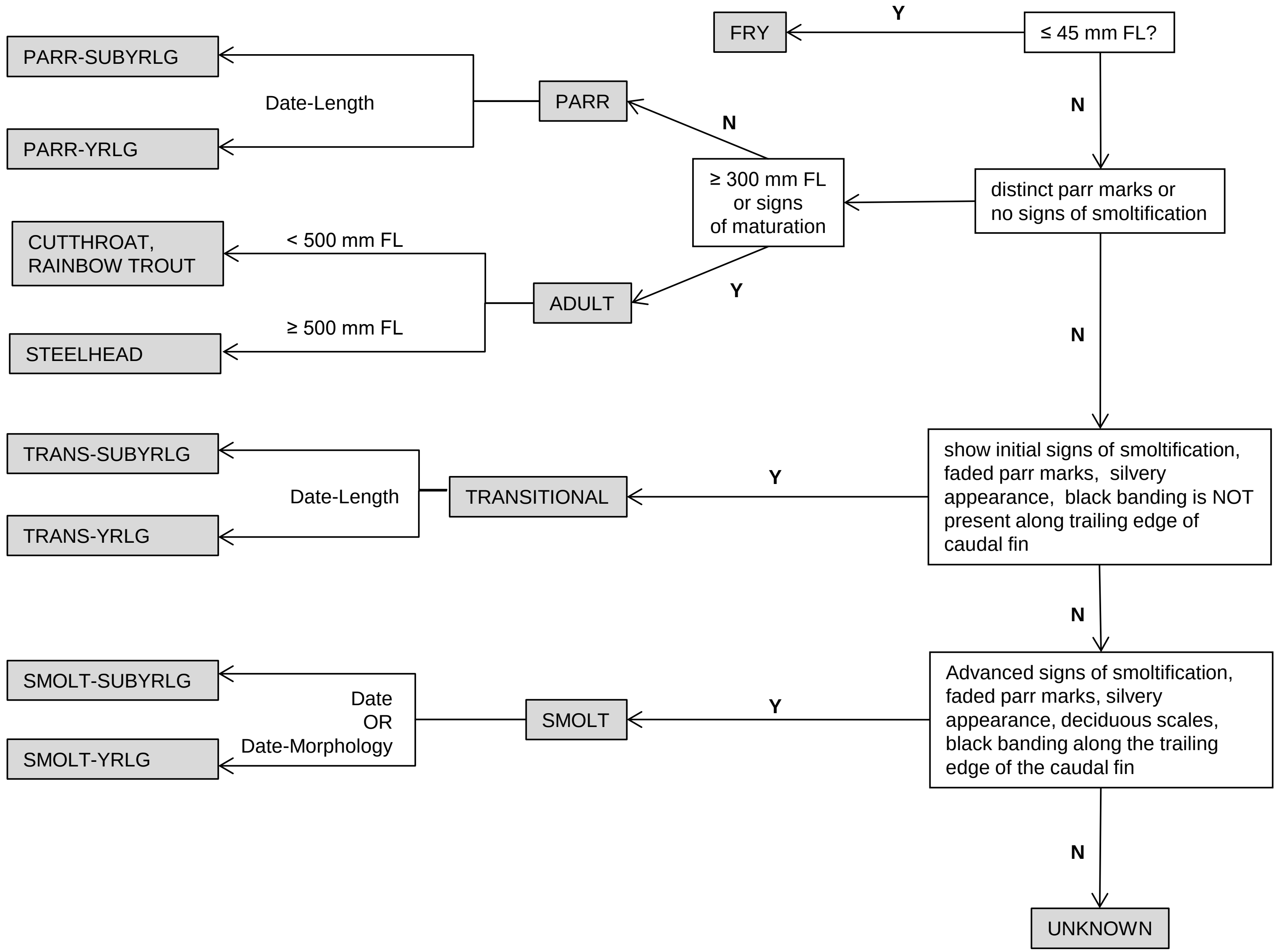
Species	Origin	Life Stage	Age Class	Start Date (Planned)	Stop Date (Planned)	Mark Rotation (Frequency)	Mark Type
Chinook	All	F	Subyearling	3/1	6/30	Continuous batch mark every 3 days	Bismarck Brown Dye
Chinook	All	P/T/S	Subyearling/ yearling	3/1	6/30	Weekly	Caudal fin clip or pan jet dot(s) (alcian blue dye)
Steelhead*	All	P/T/S	Subyearling/ yearling	3/1	6/30	Weekly	Caudal fin clip or pan jet dot(s) (alcian blue dye)
Coho*	All	P/T/S	Subyearling/ yearling	3/1	6/30	Weekly	Caudal fin clip or pan jet dot(s) (alcian blue dye)

* Coho and steelhead parr are marked because data from the Swift FSC suggest outmigration of these species as parr. Data analysis can remove these parr if necessary as part of abundance estimates.

1.6. Recapture:

• *Describe how fish are examined for all marks (visual, PIT scan, CWT wand).*	Visual inspection for panjet dye dot(s) marks, fin clips or Bismark brown dye. When applicable, fish will be wanded for presence of CWT or BWT.
• *Describe how maiden/recapture status is assigned.*	Captured fish indicating the presence of tattoo marks or caudal clips are considered recaptures. All other fish are considered maiden captures

• *Describe effort to accurately detect marked fish used in efficiency trial. Include methods used to evaluate detection rates.*	All marks are visual with assumed detection rate of 100 percent.



Updated 2.8.2016

Strategy D: Fish Health Monitoring and Disease Prevention

Strategy D: Fish Health Monitoring and Disease Prevention Strategy for Lewis River Hatchery Programs 2022

Introduction

The mission of the WDFW Fish Health Unit is to ensure and protect the health and productivity of fish, both cultured and free ranging, in the State of Washington, and operates following standards and objectives outlined in the Salmon Disease Control Policy of the Fisheries Co-Managers of Washington State (WDFW 2006) and State of Washington Fish Health Manual (WDFW 2010). Specific to the Lewis River Hatchery Programs, the Fish Health Unit is tasked to monitor population health of all species reared at Speelyai, Merwin, and Lewis River hatcheries and provide veterinary services as needed. These services include: monitoring reported and regulated pathogens in all broodstocks, as defined and required in the Co-Manager's Disease Control Policy (WDFW 2006), baseline monitoring as needed to ensure the health and vitality of all species throughout the rearing cycle, direct surveys for monitoring specific disease progression and severity in targeted populations, and working in concert with hatchery and agency stakeholders to develop adaptive therapeutic and disease prevention strategies. Fish health services are provided through detailed monitoring by a WDFW Fish Health Specialist and routine monitoring by the WDFW Aquatic Animal Health Veterinarian (AAHV) for the Lower Columbia region. The AAHV will establish and maintain a Veterinarian-Client-Patient-Relationship (VCPR) and will be the veterinarian of record for all Lewis River hatcheries. The Fish Health Unit also operates a laboratory capable of performing virology, bacteriology, parasitology, serological, and molecular testing on samples submitted for analysis.

Species reared by Lewis River Hatchery Programs and covered by this monitoring strategy include late winter steelhead, spring Chinook Salmon and Coho Salmon.

Pathogens of Interest

The following pathogens of interest are defined by the Co-Manager's Disease Control Policy (WDFW 2006). This list is periodically reviewed and updated in accordance with policy guidelines.

Regulated Pathogens: fish pathogens that are regulated within Washington and meet all of the following criteria: (1) have the potential to cause significant economic and/or biological losses, (2) are not treatable, (3) have limited range (endemic) or do not exist within Washington (exotic), (4) a repeatable robust means for their detection is recognized, and (5) a statewide surveillance program is in place for the pathogen. Pathogens that are considered regulated are as follows:

Regulated Exotic Pathogens:

- *Oncorhynchus masou* virus (OMV)
- All viral hemorrhagic septicemia virus strains other than the Pacific Northwest strain (IVa)

Regulated Endemic Pathogens:

- Infectious hematopoietic necrosis virus (IHNV)
- Infectious pancreatic necrosis virus (IPNV)
- Pacific Northwest strain (IVa) of viral hemorrhagic septicemia virus (PNW VHSV)
- *Myxobolus cerebralis* (*whirling disease*)

Reportable Pathogens: fish pathogens that are of general interest and meet the following criteria: (1) have the potential to cause significant biological, or economic loss, (2) thought to have limited geographic range within Washington State, (3) there is limited ability to control, and (4) an accepted detection method exists for these pathogens. Reportable pathogens will be screened for at the discretion of the attending fish health specialists based on clinical signs.

Viral:

- All replicating agents other than those listed as regulated pathogens.

Bacterial:

- *Piscirickettsia salmonis,*
- Strains of *Yersinia ruckeri* and *Aeromonas salmonicida* that are resistant to oxytetracycline and/or Romet and strains of *Flavobacterium psychrophilum* that are resistant to oxytetracycline and/or florfenicol.

Parasites:

- *Tetracapsuloides bryosalmonae* (PKX)
- *Ceratonova shasta*
- *Nucleospora salmonis*
- *Cryptobia* sp

Baseline Treatment and Monitoring

Baseline treatment and monitoring consists of conducting surveillance of reported and regulated pathogens according to the Co-Manager's Disease Control Policy (WDFW 2006), and monitoring as needed to ensure the health and vitality of all species throughout the rearing cycle.

Application and discharge of any compound must be consistent with label instructions and agency regulations. If needed, Veterinary Feed Directives (VFDs), veterinary prescriptions, or off-label use of medication will be provided by the veterinarian of record, at the veterinarian's discretion. Development, distribution, and use of compounds approved for use on food-fish are regulated by the United States Food & Drug Administration's (USFDA) Center for Veterinary Medicine (CVM) and Washington State Department of Health. Discharge of therapeutants and chemicals into receiving waters of this state is regulated under the National Pollutant Discharge Elimination System (NPDES), through Washington State Department of Ecology (WDOE).

Monitoring in brief, consists of monthly visits to each facility to observe fish and discuss current behavior, mortality, feeding, and any other relevant information that may affect fish health. Each stock will be examined at regular intervals to evaluate overall fish health and to minimize the risk of disease outbreaks. Routine monitoring will include the microscopic examination of fins, skin, gills, and internal organs for the presence of abnormalities or finfish pathogens. This is the minimum level of routine monitoring and may be increased as necessary. Additional diagnostics (bacterial or viral culture, histopathology) will be conducted as warranted.

Reports on fish health examination and treatment recommendations of all affected species will be provided in a fish health exam form (FH01; see example in section 1.1.6, Juvenile monitoring).

Directed Treatment

Any significant loss of fish (>~0.05% loss for consecutive days or exponentially increasing loss pattern) that is suspected to be due to an infectious agent will be promptly investigated by the facility manager and a fish health specialist. When an infectious agent is detected and implicated in the fish loss, preventative and therapeutic strategies will be implemented whenever possible to reduce the impact of such disease agents on both free-ranging and cultured fish populations (WDFW 2006). All treatments will be consistent with label instructions and agency regulations. If needed, VFD, veterinary prescriptions, or off-label use of medication will be provided by the veterinarian of record, at the veterinarian's discretion. For common species-specific diagnoses, disease prevention and treatment strategies see below (sections 1.1.1 – 1.1.4).

Fish health will also advise on testing any new antibiotics, vaccines, or other drugs for efficacy. For example, directed treatments may include other topical therapeutics such as Chloramine-T or Diquat for control of external bacterial cold-water disease (BCWD) at Lewis River.

Reports on fish health examination and treatment recommendations of affected species will be provided in a FH01. Copies of the FH01 will be supplied directly to Fish Facility Manager and WDFW staff at the applicable facility. Samples submitted for fish health laboratory assay to the Olympia Fish Health Lab will be logged in and processed according to accepted and standardized fish health laboratory protocols. Results from additional diagnostics will be reported as received. A summary of all epizootic investigations and new therapeutic protocols administered will be provided according to section 1.6.

1.1.1 Broodstock Collection and Holding

All species:

Any fish displaying overt signs of damage and/or disease (i.e. external hemorrhaging, severe fungal lesions, gross lesions) should not be selected for broodstock, if numbers allow, due to high risk of pre-spawn mortality and compromised gamete fertility or viability. Good condition of broodstock and decreased pre-spawn mortality is maintained through minimized handling, low rearing density, optimal flow, and where applicable, shade cover to mitigate stress.

All species receive metaphylactic formalin treatments (100 - 167ppm, 3 times per day, every other day) while in holding raceways or circular tanks to control fungi and external parasites. Salt is simultaneously added to reduce stress and improve oxygen uptake.

Spring Chinook:

In addition to metaphylactic formalin and salt treatments, adult spring chinook receive an antibiotic injection once prior to spawning to reduce pre-spawn mortality. This process coincides with hatchery staff sorting of a single rearing vessel to minimize stress and handling. In brief, all adult spring chinook receive a therapeutic dose of oxytetracycline (syn: Liquamycin®, LA-200®) administered sometime in late spring.

1.1.2 Spawning

All species:

Sampling guidance of all adult broodstock is provided by the Co-Manager's Disease Control Policy (WDFW 2006). Kidney, spleen, and ovarian samples of at least 60 randomly selected adult females are to be inspected and sampled by a fish health specialist for regulated pathogens (see pathogens of interest) during spawning. Sampling rates, at a minimum, shall be sufficient to achieve a representative sample at the 95% confidence level (5% assumed pathogen prevalence level). Tissue samples are delivered to the WDFW Fish Health lab in Olympia that performs accepted and standardized viral assay protocols (AFS 2016, WDFW 2010). All pools of samples will be cultured using two cell lines. All cultures exhibiting cytopathic effect (CPE) will be tested using PCR to confirm identification of viral particles. Results are sent back to the hatchery and fish health personnel. No lethal pathogen screening is proposed if any regulated viral pathogens are detected. If an exotic regulated pathogen with high mortality risk is detected, then the outcome of those eggs will be determined according to the co-manager's consensus and the salmon disease policy (WDFW 2006).

Spring Chinook:

In addition to viral surveillance, females allocated to the February release group will be ELISA tested. This test detects antigen of the causative agent of Bacterial Kidney Disease (BKD) and assigns a low, moderate, or high level based on the relative concentration of antigen detected. Historically, this yearling February release group represents an increased risk of BKD as a consequence of extended wintertime warm water temperatures during rearing at Lewis River Hatchery, which contributes to prolonged smoltification, parr reversion, sexual precocity, and additive physiological stress in the months leading up to their release. These environmental conditions and chronic stressors are primary contributors to the development of acute-on-chronic BKD development and associated mortality. Females allocated to the October group will not be sampled because progeny in this group are at a lower risk since they are released before encountering suboptimal conditions contributing to BKD expression. Evaluation of current BKD screening protocols will be made on a yearly basis in response to patterns in BKD prevalence, disease severity, and rearing mortality among untested groups.

1.1.3 Incubation

All species:

All eggs are treated with formalin according to label instructions to control fungus (WDFW and PacifiCorp 2014).

Spring Chinook:

Once ELISA results are available, WDFW staff will only combine females with the same level of results together for hatching. If WDFW determines on the last egg take that there is a surplus of eggs, then staff will have the option to cull any eggs with low, moderate, or high levels of *R. salmoninarum* antigen with confirmation from pathology. Culling has not occurred in the past due to low numbers of returning adults available for broodstock. Under the current release study, any eggs tested beyond a below-low level are allocated to the June release group. All adult females from the February release groups will be ELISA sampled at a below-low level and their eggs will be incubated as stated in the AOP. Females allocated to the June and October release group will not be ELISA sampled, and therefore, their eggs will be incubated in deep troughs (bulk eyed).

1.1.4 Rearing

All species:

Each species will be examined at regular intervals (target monthly) to evaluate overall fish health and to minimize the risk of disease outbreaks until fish are released. Routine monitoring will include the microscopic examination of fins, skin, gills, and internal organs for the presence of abnormalities or finfish pathogens. This is the minimum level of routine monitoring and may be increased as necessary. Additional diagnostics (bacterial or viral culture, histopathology) will be conducted as warranted. Hatchery metrics such as density index, flow index, conversion factor, CV, dissolved oxygen, and temperature are regularly monitored by staff. Species-specific rearing guidelines are based on fish recommendations stated in WDFW Fish Health Manual (WDFW 2010). Other water quality parameters such as turbidity and total dissolved gas are important seasonal considerations, especially if discharge events or heavy rains are expected. These are important rearing guidelines considered for fish health, specifically when related to chronic fish health issues. Additionally, barriers such as netting and gates are utilized to minimize direct and indirect morbidity/mortality due to predation.

Steelhead:

Monitoring frequency is increased in late summer due to historic presence of *Ichthyophthirius multifiliis* (Ich), a ciliate parasite. Prolonged, low-dose formalin treatments of all steelhead stocks on site are necessary due to the nature of the extended life cycle of this parasite.

Spring Chinook:

Beyond routine screening, historic presence of drop-outs associated with the protozoan parasite, Costia, during early rearing is mitigated through bi-weekly formalin treatments. When fish begin to display flashing behavior, typical of parasite infestation, treatments are initiated. These typically occur in February and continue until flashing behavior diminishes, typically before marking is completed in May.

BKD is of primary concern for this species. The bacteria are endemic in this stock; however expression of clinical disease is the result of chronic, sub-optimal external factors contributing to stress-mediated immunosuppression. Treatment options such as injectable antibiotics and medicated feed have minimal efficacy due to the intracellular nature of the bacteria. Historically, sub-yearling groups are at a lower risk since they are released before encountering suboptimal conditions contributing to BKD expression. Yearling release groups represent a higher risk of BKD because of prolonged wintertime warm water temperatures during rearing at Lewis River Hatchery, which contributes to prolonged smoltification, parr reversion, sexual precocity, and additive physiological stress in the months leading up to their release. Therefore, stress-mitigation (i.e. population densities at 0.1 or lower, CV below 9, expansion of feed presentation by multiple staff members per feeding, adoption of volitional release based upon smoltification) is necessary minimize BKD infection concurrent with staff optimizing rearing conditions to preclude clinical BKD emergence. This year, shade covers were installed at Speelyai to provide cover from direct sunlight and diminish crowding in shaded areas of the raceways. Additionally, the October/February group reared at Lewis River will be fed by multiple staff at a given feeding to increase feed presentation to the whole population and sprinkler systems will be run to provide cover from direct sunlight.

If clinical BKD is detected early in the rearing cycle (i.e. as subyearlings in spring/summer), medicated feed (i.e. extra-label oxytetracycline, TM200® or erythromycin, Aquamycin ®) will be discussed with managers as potential option to mitigate loss if administered prior to yearling smolt window. Yearling smolts are not treated due to treatment period and subsequent withdrawal period (60 days) affecting timely release.

Coho:

Gill fungus and Costia, if present during initial raceway rearing, is mitigated with weekly to bi-weekly formalin treatments. If bacterial cold-water disease (BCWD) associated mortality increases prior to marking, this is typically treated with florfenicol (syn: Aquaflor®).

1.1.5 Release

Prior to volitional release, a fish health specialist and/or AAHV will evaluate the health and physical condition of each species to determine suitability for volitional release. Early release will be requested if population health is being negatively impacted due to prolonged retention of affected stock(s).

1.1.6 Metrics and Indicators from Baseline Treatment and Monitoring

Spawning

Laboratory results are communicated to hatchery and fish health personnel. Assay results are stored at the Olympia Fish Health lab as well as electronically on WDFW servers. Results can be requested at any time to share pathogen history of any tested species/stock. Number of pools positive for CPE and which cell lines displayed CPE are reported Summary of information will be supplied in the annual report outlined in section 1.6.

- All species
    - Results of viral samples taken during spawning
    - Qualitative observations of fish health staff during sampling
        - Overt clinical signs (i.e. fungus, hemorrhage, mechanical damage, jaundice, gross organ pathology)
        - Approx. number showing overt clinical signs
- Spring Chinook
    - Antigen levels of BKD

Table 1. Fish health monitoring of spawning adults.

Species	Virology		ELISA results					Gross Pathology	
	Tissue(s) sampled	CPE results	Total	Below-low	Low	Moderate	High	Clinical sign(s)	Estimate prevalence
Steelhead									
Spring Chinook									
Coho									

Juvenile Monitoring

Reports on fish health examination and treatment recommendations of all affected species will be provided in a fish health exam form (FH01). Copies of the FH01 will be supplied directly to Fish Facility Manager and WDFW staff at the applicable facility, copies can be requested at any time by stakeholders. A summary of all cases by species will be supplied in the annual report outlined in section 1.6. FH01's include the following information and format:

<table>
<tr><td>Facility:</td><td colspan="2">Examination date(s):</td></tr>
<tr><td colspan="2">Lot description: species, stock, broodyear, any sub-group information</td><td>Life stage: adult, fry, juvenile, smolt</td></tr>
<tr><td colspan="2">Exam type: diagnostic, monthly monitoring, pre-release</td><td>Mortality range: normal, increased, epizootic</td></tr>
<tr><td colspan="3">Tissue examination: tissue(s) examined, pathogen/condition, number sampled, number positive, health status of individual</td></tr>
<tr><td colspan="3">Laboratory testing (if conducted): assay performed, pathogen(s) of interest, tissue(s) sample, number fish sample, number fish positive, results, comments</td></tr>
<tr><td colspan="3">Affected rearing unit: unit ID, water temperature, total number in unit, daily loss, % loss per day, fish per pound, total pounds, flow, is water re-use or fresh, pounds of fish per gallon per minute, flow index, density index, feed type, % body weight fed per day</td></tr>
<tr><td colspan="3">Observations/Remarks: Description of behavior, clinical history, and any other relevant information regarding case.</td></tr>
<tr><td colspan="3">Primary detection(s): Condition(s) and/or disease(s) detected are listed here</td></tr>
<tr><td colspan="3">Recommendations: Any treatments or actions recommended by fish health listed here.</td></tr>
<tr><td colspan="3">Examiner/case ID:</td></tr>
</table>

Directed Monitoring

1.1.7 Spring Chinook gas bubble trauma (GBT) monitoring during periods of increased TDG at Lewis River Hatchery

Sensitivity to gas supersaturation can vary for different species, but common guidelines suggest TDG >115-120% can lead to acute mortality with anything >102% leading to stress and secondary conditions such as fin erosion and opportunistic pathogens. Directed monitoring of spring chinook will be conducted in July/August at Lewis River hatchery with specific focus on gas bubble trauma (GBT) associated with supersaturation or elevated TDG as measured in the past exceeding >110% TDG during the end of July and throughout August.

Fish health will conduct a preliminary visit in June, after transfer from Speelyai Hatchery and prior to known periods of elevated TDG, to establish a baseline for these populations comparing fish rearing in a control raceway and a unit outfitted with gas diffusion infrastructure. Following this initial baseline visit, weekly monitoring events will occur in August to inspect for pathology from both treatment groups (control, TDG diffusion). Each survey visit will be conducted as follows: gross clinical pathology directly associated with GBT (gas bubbles and embolic lesions present in gills, eyes, lateral line, or choroid rete; exophthalmos) and secondary conditions (external lesions, fin condition) will be surveyed in 10 general population and any present moribund fish (not to exceed 10) in both treatment groups. A subset of 3 general population and 3 moribund fish from each treatment condition will be preserved in 10% neutral buffered formalin for subsequent histopathology screening for cellular pathology associated with elevated TDG. Disease severity will be scored on a 0 – 3 scale, ranging from normal/none to severe as determined by a certified pathologist.

In addition to continuous monitoring of TDG by PacifiCorp meters, fish health will record TDG at the time of sampling (estimate afternoon based on 2017 TDG data recording highest numbers at this time of day) using a handheld TDG saturometer.

This will not only gain understanding if the elevated TDG measured in the past and presumably current conditions are associated with pathology but will also further understanding if attempts to decrease TDG supersaturation via new infrastructure also results in changes in fish pathology.

1.1.8 Spring Chinook BKD monitoring upon transfer and release

Directed monitoring of spring Chinook will be conducted beyond routine fish health checks to provide additional fish health data regarding spring Chinook release strategy study outlined in Appendix D. Fish health will regularly screen for BKD presence on a monthly basis. Additional screening of mortalities in late summer and fall will document evidence of mature gonads and clinical BKD.

BKD prevalence and severity will be surveyed using a molecular assay on a subset of 20 juvenile spring Chinook at time of transfer from Speelyai Hatchery to Lewis River Hatchery (generally beginning of May and December), and 60 fish at time of release (dates vary upon release group). During the tissue sample process, any evidence of mature gonads will be recorded. Additional screening of fish that fail to volitionally release will be made to gain information on overt BKD prevalence within these fish. This information will inform potential management actions such as culling overtly infected individuals after volitional release is completed and long term evaluation of release groups strategy.

1.1.9 Metrics and Indicators from Directed Monitoring

<u>GBT monitoring</u>

Weekly

- Summary and description of gross pathology examinations of weekly visits comparing control and TDG diffused units
    - Presence/absence of gas bubbles; prevalence in examined fish

    - Presence/absence of clinical signs/secondary conditions; prevalence in examined fish
- TDG concentration(s) at time of sampling
- Summary of histopathology scoring of weekly visits comparing control and TDG diffused units

BKD monitoring

Monthly

- Presence/absence of BKD in each release group via gross examination
- Evidence of mature gonads within examined mortalities

At time of transfer

- BKD prevalence and severity among juvenile release groups conducted at time of transfer from Speelyai Hatchery to Lewis River Hatchery

At time of release

- BKD prevalence and severity among juvenile release groups

Table 2. Direct Monitoring results for BKD monitoring in Spring Chinook

Release Group (Month release, life stage, date of Lewis transfer, FPP at release	Molecular surveillance			Gross Pathology	
	Prevalence (% positive detection/# examined)	DNA load (mean genomic quantity; range)	Prevalence of severe infections (% of fish with >1000 copies/rxn)	Clinical sign(s)	Prevalence
June 80fpp					
October 12fpp					
February yearling, May 8fpp					
February yearling, Dec 12fpp					
February yearling, Dec 8fpp					

Reporting and Deliverables

A report shall be provided to the ATS each year describing the following:

1) Overview of fish health practices and protocols at the Lewis River hatchery facilities
2) A detailed list of all pathogens identified by species and life stage
3) A description of any epizootic outbreaks to include the following:

- o Description of treatment plan initiated to control or contain epizootics
- o Results of treatment plan including estimates of infection rates and mortality by life stage and species.

4) A discussion of recommended future actions to adaptively manage hatchery stocks specific to the Lewis River hatcheries. These recommended actions should be related to, but not limited to:
   - o Changes to baseline monitoring and treatment
   - o Changes to directed treatment for specific pathogens
   - o Recommendations for improved biosecurity protocols
   - o Applications for any novel testing procedures (PCR analysis of water supply)
   - o Recommendations for proactive measures or alternative hatchery practices to improve fish health, reduce stress and limit fish loss

Adaptive Management

On at least a semiannual basis, WDFW fish health staff shall provide an update to the ATS regarding fish health status at the Lewis River hatchery facilities. The update should follow the outline provided under section 1.6 with specific discussion related to deliverable No. 4. The update should also describe planned fish health activities for the next 6 months.

These updates are intended to improve communication between all stakeholders and provide a means to adaptively manage the fish health program at the Lewis River hatchery complex. The ATS shall review the annual report and make recommendations on future actions in consultation with fish health staff.

1.2 References

AFS-FHS (American Fisheries Society-Fish Health Section). 2016. FHS blue book: suggested procedures for the detection and identification of certain finfish and shellfish pathogens, 2016 edition. Accessible at: http://afs-fhs.org/bluebook/bluebook-index.php

WDFW (Washington Department of Fish and Wildlife) and WWTIT (Western Washington Treaty Indian Tribes), 2006. *The Salmonid Disease Control Policy of the Fisheries Co-Managers of Washington State*. July 2006.

WDFW, 2010. *2010 Fish Health Manual*. Washington Department of Fish and Wildlife, Fish Health Unit.

Strategy E: Spring Chinook Rearing and Release Evaluation

Strategy E: Lewis River Spring Chinook Rearing and Release Evaluation Plan

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INTRODUCTION

Washington Department of Fish and Wildlife (WDFW) staff met several times in late 2016 and throughout 2017 to discuss strategies to improve survival rates of Lewis River (Lewis River) Hatchery spring Chinook salmon. Potential changes to rearing strategies and release timing included changes in feed composition and growth rate to more closely mimic wild fish physiology, and release dates that more closely match timing of smoltification.

This document describes the basic framework for conducting work to evaluate the timing of different volitional release periods corresponding to different ages and stages of juvenile maturation with the overall goal of improving the performance of Lewis River Hatchery spring Chinook salmon. Future work may include the addition of treatment groups in which feeding and growth will be reduced in fall and winter in order to minimize early smolting and the propensity for precocity in fish released as yearlings.

APPROACH

Background

In the past, yearling spring Chinook salmon smolts were released volitionally beginning in February. Substantial decreases in numbers of returning adult Lewis River Hatchery spring Chinook began in the late-2000s, generating concerns about survival of juveniles leaving the hatchery. In recent years, Lewis River Hatchery and fish health staff have observed increasing mortality among juveniles beginning in summer months, along with increased incidence of disease (bacterial kidney disease and flag tail). A high mortality rate during hatchery rearing is counter-productive to the increase in hatchery production that is necessary to move toward the Hatchery and Supplementation Plan goal of releasing 1.25 million smolts annually. In addition, fish held until February may be prevented from earlier volitional migration if they smolt in the preceding summer or fall, which is counter to Hatchery Scientific Review Group recommendations on volitional release.

Smolting and emigration is typically associated with spring months in the year after emergence for spring Chinook salmon; however, spring Chinook can also exhibit plasticity in smolt timing. Spring Chinook salmon can also smolt in autumn, particularly if they have experienced high growth in the preceding summer (Beckman et al., 1998). Lewis River Hatchery personnel observe smolting behavior and characteristics (e.g., increased activity, silvering, descaling) at three distinct times during hatchery rearing: 1) in their first spring after hatching (approximately June); 2) first autumn (October); and 3) second winter (February). It is hypothesized that restricting volitional migration of fish that smolt in their first year of rearing stresses the fish, reducing their resistance to pathogens that are prevalent in the hatchery environment.

Another concern is the unknown proportion of precociously mature males in the Lewis River Hatchery Program. Males exhibiting precocious characteristics have been observed at the time of release from Lewis River Hatchery and sexually mature yearlings have been observed in September during passive integrated transponder (PIT) tagging of juveniles destined for upriver acclimation sites (WDFW, 2016). Fish held at Lewis River Hatchery through the winter for

February release may experience high growth rates, which is known to increase the likelihood that a male fish will mature the following fall (Larsen et al., 2006).

Beginning with brood year (BY) 2013, WDFW began an early release program in an attempt to minimize in-hatchery mortality. Three different release strategies were used for BY 2013 (Table 1). One group was released in October at ~12 fish per pound (fpp) after being transferred from Speelyai Hatchery to Lewis River Hatchery in May 2014. A second group was released at ~8 fpp in February after being transferred from Speelyai Hatchery to Lewis River Hatchery in May 2014; this was the strategy used in most previous years. A third group was released at 12 fpp in February after transfer from Speelyai Hatchery to Lewis River Hatchery in December 2014 and January 2015. It is notable that the water temperatures of the hatcheries differ from two nearby unimpounded tributaries; water temperatures in Canyon Creek and the Lewis River inflow to Swift Reservoir (upstream of impounded sections of the Lewis River) peak in July or August and decline sharply starting in September, compared to temperatures in the hatcheries that remain elevated until late October or November (Figure 1). It is also notable that temperatures differ considerably between Speelyai and Lewis River hatcheries; between the months of June and December, fish held at Speelyai Hatchery would experience cooler temperatures that more closely match the natural decline in the fall and early winter to which salmon are adapted compared to the Lewis River Hatchery. Fish reared in colder water temperatures at Speelyai Hatchery experience slower growth and require less feed in summer and fall, achieving a smaller size at release while still meeting bioenergetic demands of the fish. BY 2014 was intended to be split into fall and spring release groups, but roughly 38% of the fish were released in August 2015 due to a hatchery water issue, about 9% were released in February 2016, and the remainder released in October 2015. BYs 2015 and 2016 were released only in October, due to low numbers of available fish for release and concerns about excessive in-hatchery mortality rates if fish were held until February. Beginning with brood year 2017, a new juvenile release plan was implemented based on discussion by the ATS and approval by the ACC.

Table 1. Numbers released by month and size since implementing October release strategy

Brood Year	October Release	February Release	
	12 fpp	12 fpp	8 fpp
2013[a]	430,303	468,765	217,805
2014	1,127,910[b]	0	116,775
2015[c]	506,547	0	0
2016[d]	402,224	0	0

Notes:

a. *Two strategies were used for the February 2015 release. One group was moved from Speelyai Hatchery to Lewis Hatchery in December and reared to 8 fpp, the second group was moved from Speelyai Hatchery to Lewis River Hatchery in June and reared to 12 fpp.*

b. *Includes 661,020 released on time in October 2015 and 466,890 released early on August 3, 2015, due to high water temperatures and mortality during summer 2015.*

c. *All fish were released in October 2016 due to low brood numbers and high mortality during summer 2016.*

d. *All fish were released in October 2017 due to low brood numbers.*

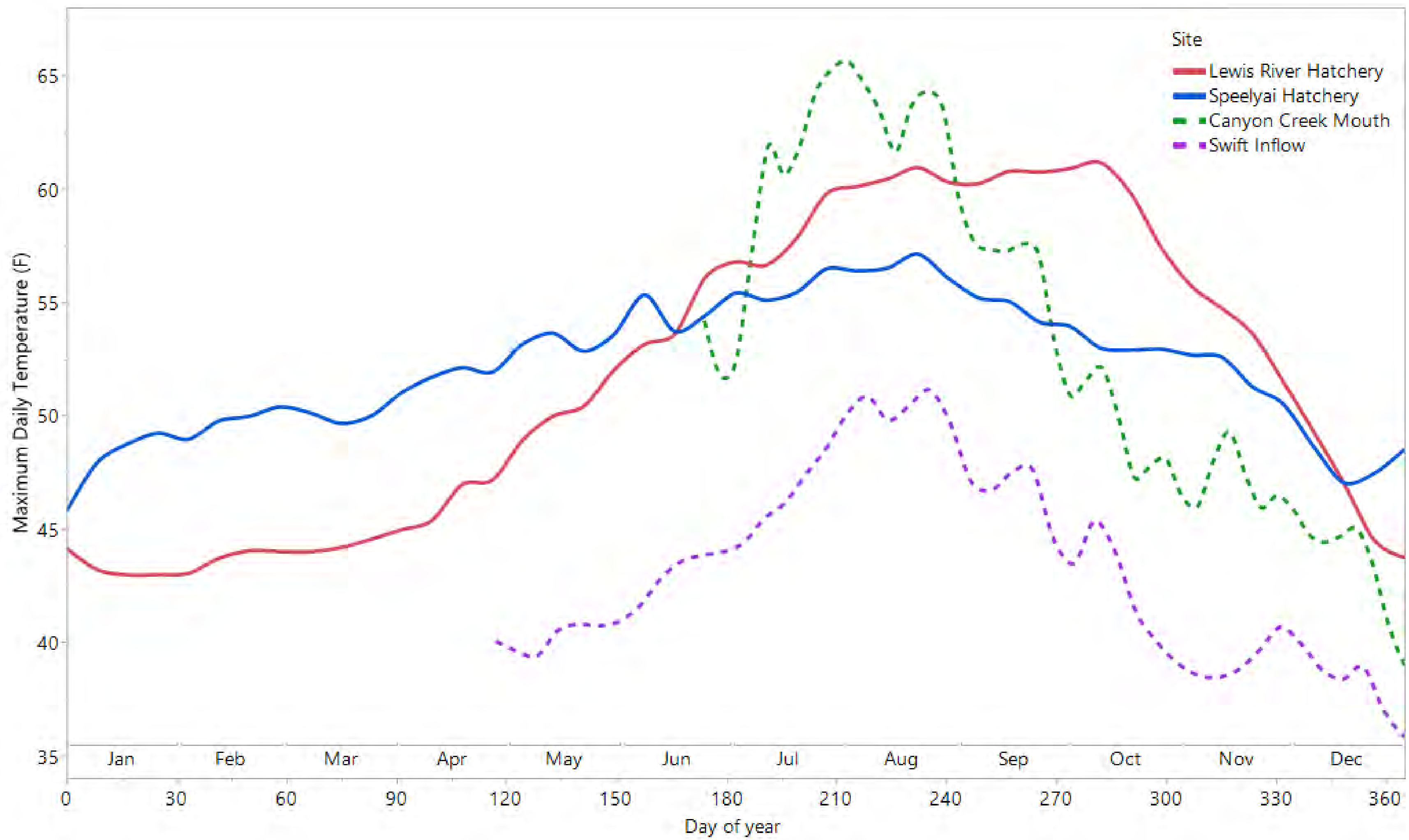

Figure 1. Maximum daily temperature at Lewis River Hatchery and Speelyai Hatchery in 2016

Notes: Canyon Creek and the Lewis River inflow at Swift Reservoir are two nearby unimpounded tributaries that represent a natural temperature profile. Data from Lewis River Hatchery (elevation 40 feet) and Speelyai Hatchery (elevation 245 feet) were collected in 2016 (WDFW, 2017). Data from Canyon Creek (elevation 300 feet) and Swift Inflow (elevation 1,000 feet) were collected in 1999 (PacifiCorp and Cowlitz PUD, 2004).

Objectives
Evaluate release strategies to optimize the number of adult returns of Lewis River Hatchery spring Chinook salmon. The results of the evaluation will be used to identify the rearing and release strategy that maximizes in-hatchery and post-hatchery survival rates for juvenile spring Chinook salmon as it relates to release date, size at release, rearing location, and age at release.

Treatment Variables
To evaluate the effects of different rearing and release strategies, the treatments described in the following sections will be used.

FEED AND GROWTH PROFILES

For the initial phase of a rearing and release evaluation, all study fish will be held at Speelyai Hatchery. During the fry rearing stage until June, all fish will be fed the same food at rates that differ among fish that have emerged at different times to produce a uniform fish size of 130 fpp by June. After June, all juvenile fish in the treatment and control groups will be fed the same low lipid diet of Bio Clark (18% fat and 47% protein), but different feed schedules by group to reach size targets of 8 or 12 fpp. The end-of-month target sizes for each release group and feeding strategies are summarized in Tables 2 and 3.

Table 2. Juvenile Chinook end-of-month target size (fpp)

Release group	Jan	Feb	Mar	Apr	May	Jun	Jul	Aug	Sep	Oct	Nov	Dec	Jan	Feb
October release at 12 fpp. Transfer to Lewis in May at 135 fpp	475	280	180	150	120	70	40	20	12.5	12				
February release at 8 fpp. Transfer to Lewis in May at 135 fpp	475	280	180	150	120	80	58	30	20	16	12.5	10	8.5	8
February release at 8 fpp. Transfer to Lewis in Dec at 14 fpp.	475	280	180	150	135	57.6	42.0	30.0	21.5	16.8	14.5	11.0	9.0	8
February release at 12 fpp. Transfer to Lewis in Dec at 18 fpp	475	280	180	150	135	57.6	42.0	33.0	25.0	20.0	18.5	15.5	13.0	12

Note:
Blue shaded cells indicate fish being held at Speelyai Hatchery.

Table 3. Feed type and size by fish size

Feed Size	Fish Size (fpp)
MASH Bio Vita Starter	Ponding – 1,000
No. 0 Bio Vita Starter	1,000 – 570
No. 1 Bio Vita Starter	570 – 300
No. 2 Bio Vita Starter	300 – 150
1.2 mm Bio Clarks Fry	150 – 90
1.5 mm Bio Clarks Fry	90 – 60
2.0 mm Bio Clarks Fry	60 – 25
2.5 mm Bio Clarks Fry	25 – 8

BROODSTOCK SELECTION

The study will use Lewis River spring Chinook salmon when the broodstock is abundant enough to support study needs. In future years, Kalama stock may be used if broodstock goals are not met with Lewis River stock, depending on the abundance and availability of both stocks. Surplus juveniles, in excess of Lewis River program production goals, will be used for a sub-yearling June release group.

RELEASE STRATEGIES

Five different rearing and release strategies will be employed:

1. Release yearling fish in February (of their second year) from Lewis River Hatchery at 8 fpp, reared at Speelyai Hatchery until June (of their first year) and then transferred to Lewis River Hatchery. Justification: this was the primary strategy used prior to BY 2013. A long time-series of survival data for this release strategy is available, and survival has decreased in recent years. Continuing this release group will allow for assessing relative success of other groups. A modest number of fish will be included in this group due to concerns about in-hatchery mortality.
2. Release yearling fish in February (of their second year) from Lewis River Hatchery at 12 fpp, reared at Speelyai Hatchery until December (of their first year) and then transferred to Lewis River Hatchery. Justification: this strategy was used for a group of fish from BY 2013 but has not been used since. Assumption: The advantage of rearing fish at Speelyai Hatchery through the summer and fall is that water temperatures are lower and disease is less likely to develop. Preliminary smolt-to-adult (SAR) survival of this group for BY 2013 was the highest of any release group in many years. The intention is to replicate, as closely as possible, the strategy used for this group in BY 2013. Due to space constraints at Speelyai Hatchery, there will be a modest number of fish in this group.
3. Release yearling fish in February (of their second year) from Lewis River Hatchery at 8 fpp, reared at Speelyai Hatchery until December (of their first year) and then transferred to Lewis River Hatchery. Justification: this is a new release strategy that

mimics release strategy No. 2, except fish would be reared to a larger size. Assumption: larger fish should survive better than smaller fish after release. Due to space constraints at Speelyai Hatchery, there will be a modest number of fish in this group.

4. Release yearling fish in October (of their first year) from Lewis River Hatchery at 12 fpp, reared at Speelyai Hatchery until June (of their first year) and then transferred to Lewis River Hatchery. Justification: this strategy has been used each year starting with BY 2013 (released in 2014). Assumption: releasing fish in October will avoid the increase in mortality of juveniles observed during fall months within the hatchery. This will be the largest release group because: 1) preliminary SAR survival of this group for BY 2013 was promising; and 2) there is sufficient space to hold this large group at Lewis River Hatchery.
5. Release subyearling fish in June (of their first year) at 80 fpp, reared exclusively at Speelyai Hatchery and released in the lower Lewis River at the Pekin Ferry Road boat launch. Fish will be released after June 1 when fish show signs of smoltification. Justification: this group may avoid the increase in mortality of juveniles observed during summer months within the hatchery by releasing them in early summer. This is a new release strategy with a high amount of uncertainty about fish survival. The current assumption (until proven otherwise) is that survival after release would be lower than other release groups, so this may be the first group to be eliminated if broodstock goals cannot be met.

Prior to 2018, 100,000 smolts were released as part of an integrated (50% natural-origin broodstock) hatchery group that was designated for acclimation upstream of Swift Reservoir. The ACC agreed in June 2018 the BY2017 acclimation group should be marked with a ventral fin clip and released downstream of Merwin Dam due to relatively low collection rates of juvenile Chinook observed at the Swift FSC. The ACC also agreed that starting with BY2018, hatchery release groups would be produced entirely from HOR broodstock (i.e., segregated). The distribution of these fish in 2019 is 50,000 to be released in October and 50,000 to be released in June.

Study Design

The study design will test release strategies described above and will include appropriate numbers of fish per experimental test group to provide statistically significant results (see Attachment A, Sample Size and Statistical Power for Detecting Changes in SAR). The release groups and treatment variables proposed for the evaluation are summarized in Table 4.

Table 4. Planned annual release groups beginning with brood year 2017

Release Group	Release Month	Transfer Month to Lewis River Hatchery	Rearing Environment at Speelyai Hatchery	Size (fpp)	Total Number at Ponding	Losses Pre-Marking[1]	Number Marked (Ad and CWT)	Post-Tagging Mortality Rate[2]	Expected Tag Release	Expected Total Release	Planting Goal
1	February	June 2018	Raceway	8	200,000	6,000	75,000	24.0%	57,000	147,440	150,000
2	February	Dec 2018	Asphalt pond	12	200,000	6,000	75,000	9.0%	68,250	176,540	175,000
3	February	Dec 2018	Raceway	8	170,000	5,100	75,000	9.0%	68,250	150,059	150,000
4	October	June 2018	Raceway	12	950,000	28,500	75,000	10.0%	67,500	829,350	825,000
5[3]	June	NA	Raceway	80	52,000	1,560	50,000	0.6%	50,137	50,137	50,000
	TOTAL				1,572,000	47,160	350,000		311,137	1,353,526	1,350,000

Notes:
[1]A pre-marking mortality rate of 3% applied to all groups
[2]Mortality rates averaged for BY 2011 through 2015 wherever release size and timing were similar. Mortality rates for potential June releases were derived from hatchery records. Mortality rates for October and February releases were derived by subtracting the RMIS release numbers from the original number tagged for each group. Mortality rate for release groups 2 and 3 was assumed to be the same as the 12 fpp release group from BY 2013
[3]A minimum of 50,000 fish will be planted, but if additional juveniles are available due to better than expected survival etc., they would be released in this group. All fish from this release group will be adipose fin-clipped; at least 50,000 fish will be marked with CWT. No surplus fish were available from BY2017 to initiate a June release group in 2018.

MARKING

A portion of fish from each of the release treatment groups will be marked with unique coded wire tags (CWTs) to allow for SAR to be estimated separately for each test group after adults return to the Lewis River Hatchery trap.

If Kalama stock are used, they will be uniquely marked so they can be identified upon return and disposed of appropriately.

DISPOSITION OF STUDY FISH

Lewis River spring Chinook salmon adults that are identified as having originated from this release evaluation by presence of a CWT may be used in the Lewis River hatchery production program or as broodstock in the supplementation program. CWTs must be collected from lethally sampled fish to determine release group ID; therefore, any adult fish with a CWT would not be transported upstream for the adult supplementation program.

Any returning adult Kalama-stock spring Chinook salmon originating from this evaluation will not be used in Lewis River hatchery production or supplementation programs if broodstock collection goals can be met using only the Lewis River stock. Surplus Kalama-stock spring Chinook salmon will be terminated upon return to the Lewis River Hatchery facilities[1].

STUDY DURATION

At least 3 BYs of fish will be released as part of this study, and rearing and release treatments will be repeated in each year. The strategies will be evaluated each year and changes might be made if substantial problems are discovered. Survival will be monitored for 4 years following the release of the last treatment group, until 2025.

WITHINHATCHERY MONITORING

During hatchery rearing, test groups will be monitored to ensure fish welfare and to characterize observed growth rates, smoltification status, and precocity rates. Batch weights are monitored monthly throughout rearing. At the time of release, 100 fish from the general population are randomly sampled from each raceway from three locations for a total of 300 fish. If needed, fish are sedated using MS-222. Individual fork lengths, weights, and smolt index including visual signs of precocity are recorded. These data are used to estimate growth and performance parameters listed below for each raceway. Visual cues of precocity include dark and/or bronze coloring, rounded and robust body size, milt release.

Specific monitoring activities would include:

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[1] The disposition of surplus adult returns originating from this evaluation may change pending further discussion among the Lewis River Hatchery and Supplementation Subgroup about incorporation of both CWT Lewis River-stock and Kalama-stock adults into supplementation and reintroduction programs.

Monthly

- Size (batch weights)
- Visual assessment of smolt behavior and appearance
- Presence of milting fish

Fish health checks as needed prior to release

- Routine fish health evaluations and screens
- Pathology investigations for mortality events
- Necropsy of mortalities in late summer and autumn for evidence of mature gonads
- Screening for bacterial kidney disease prevalence among juvenile release groups conducted at time of transfer from Speelyai Hatchery to Lewis River Hatchery

At time of release

- Size (fork length and weight)
- Condition factor (K)
- Visual assessment of smolt behavior and appearance[2]
- Additional screening for bacterial kidney disease prevalence among juvenile release groups
- Cull any obviously precocious males and estimate a precocity rate, as described in Strategy F, precocity and morphology sampling[3].

ANALYSIS

After 3 years of implementation, in-hatchery survival rates, size-at-release, condition factor at release, fish health (frequency or rates of disease), and physiological status at the time of release will be compared between treatment groups.

Final SARs will be evaluated for up to approximately 6 years following production of each brood year (BY) (e.g., in 2023 for BY 2017). Preliminary SAR analysis (tag returns) for each BY test groups will begin at year 2 to ensure jack returns are accounted for (see Table 5). Comparisons may be made among mean SARs of: 1) each treatment group across BYs to evaluate the effect of rearing and release strategies on survival; and 2) each release year, to investigate effects of rearing and release strategies on survival independent of the effects of inter-annual environmental variation. Comparisons in the ages of returning adults will be made between treatment groups to identify any shift in population age structure among treatments. Comparisons will be made in context of interannual trends observed in other Lower Columbia stocks including the Cowlitz and Kalama spring Chinook to discern whether survival of a given

[2] If desired in the future, the intensity of smoltification or proportion of groups that are fully smolted can be more accurately assessed monthly or at the time of release by sampling a physiological marker of smolting (gill Na+,K+ ATPase), requiring the lethal sampling of 25 fish per group.

cohort may have been affected by conditions downstream of the Lewis River experienced by all stocks (e.g., poor ocean conditions affecting a single cohort).

Table 5. Lifespan of study fish from three replicate years

Study Year	Spawn	Emergence	Release	Return Age 2+	Return Age 3+	Return Age 4+	Return Age 5+	Return Age 6+
2017	BY 2017							
2018	BY 2018	BY 2017	BY 2017					
2019	BY 2019	BY 2018	BY 2017, BY 2018	BY 2017				
2020	*	BY 2019	BY 2018, BY 2019	BY 2018	BY 2017			
2021	*		BY 2019	BY 2019	BY 2018	BY 2017		
2022	*				BY 2019	BY 2018	BY 2017	
2023	*					BY 2019	BY 2018	BY 2017
2024	*						BY 2019	BY 2018
2025	*							BY 2019

Note:
**Continuing study releases after BY 2019 will be reevaluated after 3 years of releases are complete (see following section).*

Decision Making

The results of the study will be used to make specific management and operational decisions for the Lewis River spring Chinook salmon program.

If there is a statistically supported difference in total survival (in-hatchery plus post-release) or age at return between release groups, future operations will change to adopt the rearing treatment conditions used for the group with the highest total survival to age 4+ or older. Metrics that will be considered include in-hatchery survival, SAR, proportion of males maturing at age 2+ (minijacks) and 3+ (jacks), and mean or median age at return of the population. Fish health metrics that inform survival may also be considered, including bacterial kidney disease load at the time of release. Interannual differences in the river and ocean conditions will affect cohort survival differently depending on the year of release, therefore the mean or median of three release years will be evaluated to discern whether effects of hatchery rearing on SAR or age at return are greater in magnitude than effects of the river or ocean environment. Physiological measurements of smolt and reproductive development will be used to discern a mechanism for differential fish health and survival between treatment groups.

The Lewis River program will consider changing release goals based on total survival of all groups. If total survival of subyearling releases significantly exceeds total survival of yearling releases, the program will consider releasing all fish at the age that demonstrates highest survival. The program may also consider releasing a proportion of each cohort at two or three different ages if inter-annual variation in survival between release ages or tradeoffs between in-hatchery survival, SAR, total number returning, and age at return, supports spreading risk across more than one release age.

Deliverables

The results of this evaluation will be summarized in data submissions to WDFW Fishbooks and a PacifiCorp-managed database monthly throughout the study, annual interim updates in the Lewis River Annual Operations Report, and in a final completion report at the conclusion of the study. Specific elements of these deliverables are described below:

Monitoring Data Submissions (monthly)

- Within hatchery monitoring (see above)
    - Fish health
    - Size and growth rates
    - Smoltification status
    - Observed precocity

Lewis River Annual Operations Report (annually)[3]

- Summary of within hatchery monitoring results to date beginning in 2018
- Preliminary survival estimates for returning adults from each BY would be calculated beginning in 2021

Completion Report (year 2025)

- Summary of monitoring results
- Comparison of in-hatchery survival and SARs among treatment groups
- Key conclusions
- Recommendations for future rearing and release practices

Limitations and Specific Concerns

The successful implementation of the evaluation will depend on the following factors that require consideration and coordination before the study begins.

1. *Adequate numbers of broodstock from the Lewis River program.* Recent returns have fluctuated greatly in number each year and the availability of broodstock over the course of this multi-year evaluation are uncertain. The effect of release treatments on survival and tag recovery may affect availability of spring Chinook salmon returns for supplementation purposes. The use of surplus Kalama River broodstock as a surrogate for Lewis River has been discussed as a less preferred option if sufficient Lewis River fish do not return.
2. *Sufficient numbers of marked adult returns to conduct statistical evaluations.* Detecting a significant difference among test groups will require sufficient numbers of returning adult fish from each test group (see Attachment A, Sample Size and Statistical Power for Detecting Changes in SAR).

[3] Within-hatchery monitoring will begin in 2018 on BY 2017 fish to be released in October of 2018 and February of 2019.

3. *Maintaining pre-programmed release dates throughout the evaluation.* It is anticipated that the health (i.e., disease related mortality) of fish within the hatchery may trigger concerns about the necessity for early release of treatment fish. The integrity of the evaluation will require maintaining the targeted release dates for treatment fish and tolerance levels for mortality rates should be discussed and resolved prior to the implementation of the evaluation.
4. *The evaluation will need to be managed carefully, by project leads at PacifiCorp (Erik Lesko) and WDFW (staff member TBD) with oversight by the Hatchery and Supplementation Subgroup.* Even with dedicated leads, hatchery studies often present challenging coordination environments because of the number of people involved and the extra work required by the study itself.
5. *Release location consistency.* Fish from all treatment groups will be released in the same location near the Lewis River Hatchery.

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Attachment A
Sample Size and Statistical Power for Detecting Changes in SAR

Sample sizes of coded wire tag (CWT)-marked fish required to detect changes in smolt to adult returns (SAR) were calculated to inform monitoring and evaluation of rearing or release strategies for North Fork Lewis River spring-run Chinook salmon. A Two-Proportion Sample Size and Power Test was performed using the ‘pwr’ package written for R by Champely et al. (2018) based on effects sizes and notations from Cohen 1988.

Results are presented as the required sample size of CWT-marked fish to detect differences between SAR values that have been observed in recent program years (Figure A-1), and the increase in statistical power with increasing SAR values at a fixed sample size (50,000 fish per group; Figure A-2).

Figure A-1 demonstrates that as absolute SAR values and differences between SAR values increases, the sample sizes needed to detect real differences between groups decreases. For instance, to detect a difference in SAR between 0.05% and 0. 10% with statistical confidence will require minimum sample sizes of 10,000 to 50,000 fish, whereas detecting a relatively small difference in SAR between 0.10% and 0.13% will require 500,000 to 1,000,000 tagged fish.

Figure A-2 demonstrates that as the absolute SAR values and differences between SAR values increases, the ability to detect real differences between groups improves. For instance, if comparing two treatment groups of 50,000 juveniles, confidence that the difference between SARs of 0.04% and 0.05% is low, with less than 20% power; however, the confidence to detect a difference between 0.05% and 0.10% is high, with at least 80% power.

For reference, Table A-1 provides SAR by release groups (CWT only and CWT+AD clip) for the past 10 years available in the Regional Mark Information System and Washington Department of Fish and Wildlife databases for Lewis River Hatchery spring Chinook.

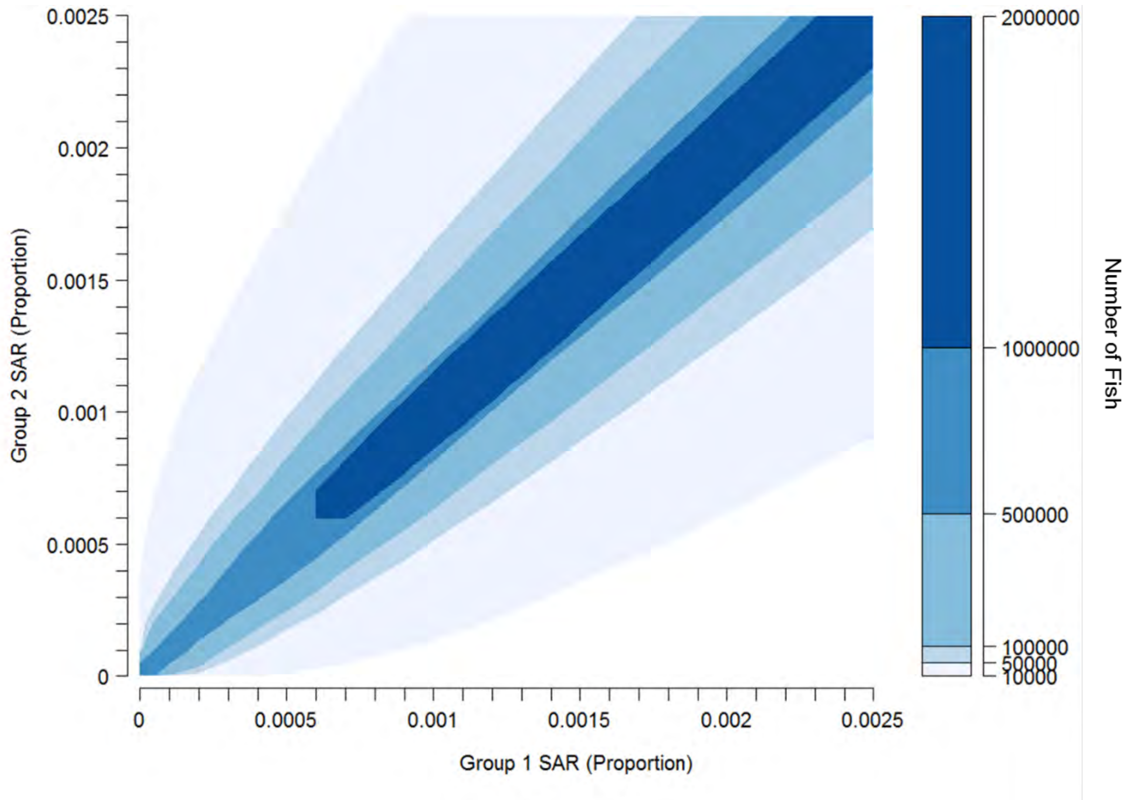

Figure A-1. Change in the necessary sample size with change in smolt-to-adult ratio and constant power (0.8)

Notes: Power = 0.80 and Alpha = 0.05

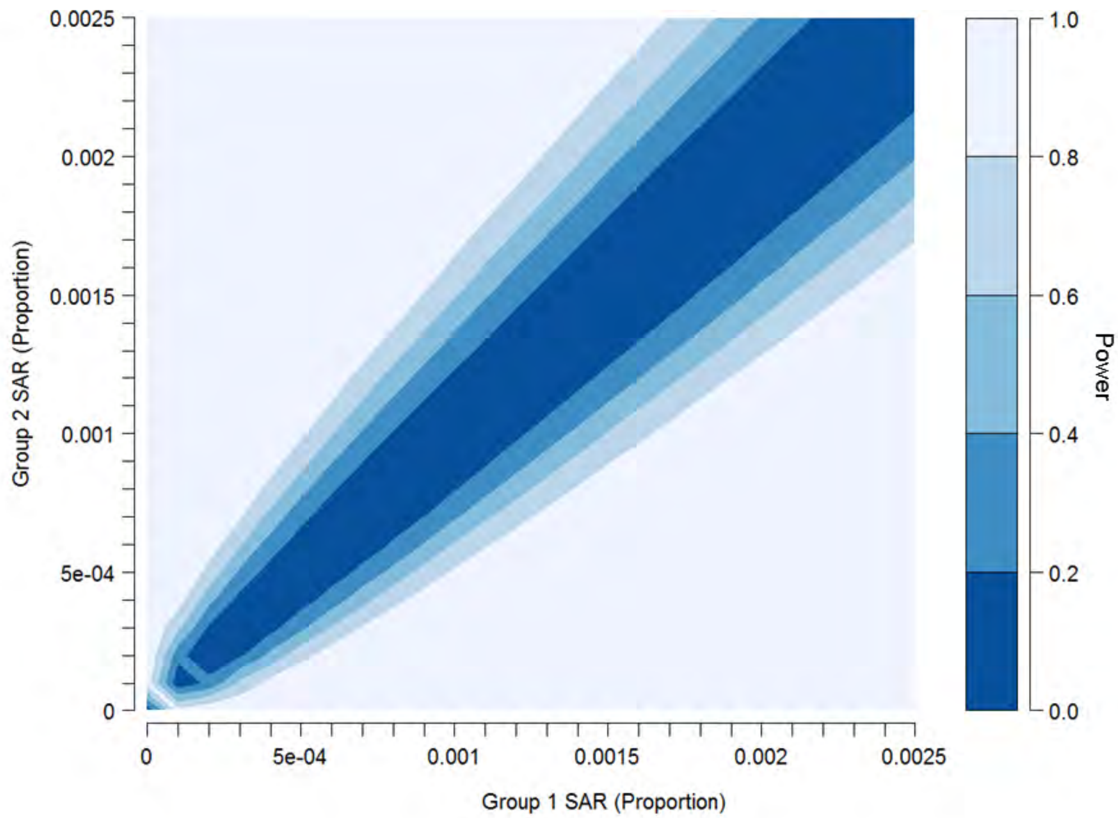

Figure A-2. Change in statistical power with change in smolt-to-adult ratio and constant sample size between two treatment groups (n = 50,000 per treatment)

Notes: Sample size (n) = 50,000 and Alpha = 0.05

Table A-1. Brood years 2002 to 2013 of Lewis River Hatchery spring Chinook smolt-to-adult ratio by brood year and release group

Brood Year (by release group)	Age at Release	Target Size at Release	Mean % SAR[a]
2002	Yearling	8 fpp	.97
2003	Yearling	8 fpp	.26
2004	Yearling	8 fpp	.10
2005	Yearling	8 fpp	.12
2006	Yearling	8 fpp	.29
2007	Yearling	8 fpp	.07
2008	Yearling	8 fpp	.24
2009	Yearling	8 fpp	.06
2010	Yearling	8 fpp	.06
2011	Yearling	8 fpp	.06
2012[b]	Yearling	8 fpp	.02
2013[b]	Subyearling	12 fpp	.13
2013[b]	Yearling	12 fpp	.25
2013[b]	Yearling	8 fpp	.11

Notes:

a. Mean of 2 to 5 release groups, depending on the year

b. Values are preliminary and do not include fish returning at age-5 or older

Percent survival based on CWT fish captured in all fisheries, spawning grounds, and hatcheries

Data sources are Regional Mark Information System (2018 expanded tag recoveries for BY 2002 to 2013. This report excludes recoveries in Fishery 70 series (juvenile recoveries).

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Strategy F: Spring Chinook Precocity Assessment

Precocity Assessment Study Plan for Lewis River Spring Chinook Salmon

1 Introduction

Residualism of hatchery-origin spring Chinook salmon in the North Fork Lewis River may result in negative ecological interactions (e.g., predation, competition, and genetic introgression) between hatchery fish and natural-origin listed species. The Aquatic Technical Subcommittee has discussed potential ways to quantify residualism and determined that measuring precocity in hatchery-reared spring Chinook salmon and steelhead should be a priority.

Males of several species of anadromous salmonids may undergo sexual maturation without emigrating from freshwater, typically referred to as precocious parr. Some spring Chinook salmon populations exhibit precocity in both subyearlings and yearlings (Larsen et al. 2004). Although limited information is available, the incidence of precocious maturation in wild stocks of spring Chinook salmon is thought to be less than 5% (Mullan et al. 1992). By contrast, precocity rates in Washington hatcheries have been documented between 20% and 80% of males (Harstad et al. 2014).

While many hatchery-reared precocious male Chinook salmon in the Columbia River system make partial migrations downstream, returning in the same year to spawn, many remain near their hatcheries of origin in freshwater until spawning (Johnson et al. 2012; Beckman and Larsen 2005). Precocious males are more likely to residualize (remain in fresh water prior to spawning), may not survive to spawn (Larsen et al. 2010), are unlikely to migrate to sea to achieve full adult size and ages at return, and contribute fewer offspring to the total population.

The age at which salmon mature is influenced by genetics and a suite of biological factors, including lipid content, size, and growth rate at specific times of year (Silverstein et al. 1998). Additionally, environmental factors can influence age at maturation. Cold water temperatures during fall and winter, a temperature profile that would naturally be experienced by wild juvenile salmon, has been shown to reduce age-2 precocity rates in captive fish (Larsen et al. 2006; Harstad et al. 2014). Photoperiod at emergence has also been shown to influence both age at smoltification and age at maturation, with earlier emergence and high growth contributing to precocity at age 1 (Beckman et al. 2007).

It has been documented that the physiological initiation of maturation in male spring Chinook salmon occurs in late fall, approximately 10 months prior to spawning (Silverstein et al. 1998). Obvious external signs of maturation are not typically evident until the summer before spawning (2 to 3 months). These external characteristics include development of olive pigmentation, a deepening of body shape, and a darkening of the fins along the margins. Additionally, mature males develop enlarged white gonads as maturation progresses. Most spring Chinook salmon hatcheries typically release fish in March or April, 5 to 6 months prior to spawning, when these indicators of precocity are often not observed such that the incidence of precocity within a population is difficult to estimate at the time of release from the hatchery.

2 General Approach

2.1 Background

Currently, limited data have been collected to describe the magnitude of the impact of precocious male spring Chinook salmon produced from the Lewis River Hatchery program on the performance of the program (WDFW 2018, 2019, 2020), nor is information available about their potential impact on wild salmon and steelhead residing in the North Fork Lewis River. In the *Draft 2020 Hatchery and Supplementation Plan*, PacifiCorp has identified measuring precocity in spring Chinook salmon as a key question to address in the 5-year period of the plan (2021 to 2025). NOAA has also provided input that precocity is a concern for this program; similar programs to the Lewis River (those that have coverage under the Mitchell Act BiOp) have been directed through incidental take statements to limit residualization to no more than 5% of the cohort becoming precociously mature in any given year, or the 5-year average exceeding 3% at any time (NOAA-NMFS 2017).

Hatchery-reared spring Chinook salmon juveniles released in October have been observed during annual fall Chinook salmon carcass surveys attempting to spawn with adult fall Chinook salmon. Juveniles have also been observed in the Merwin Trap (upstream about 3 miles) within days after release from the Lewis River hatchery. WDFW staff have also observed juvenile spring Chinook salmon at the Cedar Creek fishway trap after release from Lewis River Hatchery. Additionally, high mortality has been observed at the Lewis River Hatchery in the fall, winter and spring prior to release of the October and February release groups over several years. It is suspected that some of this mortality can be attributed to senescence of precociously mature males. Spring Chinook salmon typically die within weeks after their typical spawn timing; however, some precocious males retained in the Lewis River Hatchery have been observed several months post-maturation and are easily identified by their body coloration, continue to express milt, and upon dissection show signs of testis atresia and often have bloated abdomens. Unlike iteroparous fish, spring Chinook salmon cannot reinitiate gonad development in the following season. Finally, there is concern that precocious fish released from the hatchery would interact negatively with natural origin fish. Specifically, precocious spring Chinook salmon released in October may interact with and potentially hybridize with spawning fall-run Chinook salmon. Precocious spring Chinook salmon released in February may tend to reside in freshwater for an extended period of time prior to spawning, putting them in proximity with natural-origin fall Chinook salmon fry, as well as other listed salmonids such as coho salmon, chum salmon and steelhead, of a size that makes them vulnerable to predation by the larger spring Chinook salmon yearlings (NOAA-NMFS 2017).

It is well documented that fish held at Lewis River Hatchery through the winter for February release experience relatively warm water temperatures (typically in October) due to stratification of Merwin Reservoir upstream contributing to high growth rates during the fall (*2020 Annual Operating Plan* [AOP] – Appendix D). In addition, emergence timing is not uniform among spawning groups, and early growth is accelerated with an increased feed rate in the later emerging groups during early ponding to bring all fish to a uniform size prior to transfer to large rearing ponds.

Five different strategies have already been employed to evaluate the effects of different rearing conditions and release timings, as outlined in the *Lewis Spring Chinook Salmon Rearing and Release Evaluation Plan* (Table 1; *2020 AOP* – Appendix D).

The study described here expands upon spring Chinook salmon male monitoring at Lewis River Hatchery that seeks to quantify the potential impact each of the five rearing strategies has on pre- and post-release survival rates of North Fork Lewis River spring Chinook salmon, and post-release impacts on other impaired stocks in the Lewis River. Rigorous quantification of precocity rates will inform the interpretation of the survival monitoring outcomes.

2.2 Target Species and Study Area

Juvenile spring Chinook salmon will be sampled prior to release in the fall (October yearling release group) and spring (February yearling release groups). Sampling will occur at Lewis River Hatchery where juveniles complete the rearing phase prior to release. The five different groups will be reared in separate ponds. In the study outlined here, precocious maturation refers to male fish that mature toward the end of either their first year (microjacks) or second year (minijacks), as measured from the date of fertilization.

Table 1. Juvenile Chinook Salmon End-of-Month Target Size Measured as Fish Per Pound

	Release Group	Jan	Feb	Mar	Apr	May	Jun	Jul	Aug	Sep	Oct	Nov	Dec	Jan	Feb
5	June release[1] at 8 fpp	475	280	180	150	120	80	—	—	—	—	—	—	—	—
4	October release at 12 fpp Transfer to Lewis in May at 135 fpp	475	280	180	150	120	70	40	20	12.5	12	—	—	—	—
1	February release at 8 fpp Transfer to Lewis in May at 135 fpp	475	280	180	150	120	80	58	30	20	16	12.5	10	8.5	8
3	February release at 8 fpp Transfer to Lewis in Dec. at 14 fpp	475	280	180	150	135	57.6	42.0	30.0	21.5	16.8	14.5	11.0	9.0	8
2	February release at 12 fpp Transfer to Lewis in Dec. at 18 fpp	475	280	180	150	135	57.6	42.0	33.0	25.0	20.0	18.5	15.5	13.0	12

Note:

1. A June release group would occur if surplus juveniles are available due to better than expected survival etc.

Blue shaded cells indicate fish being held at Speelyai Hatchery.

fpp: fish per pound

—: No fish being held at this time.

2.3 Study Questions and Study Objectives

The study questions driving the need for additional monitoring of precocious maturation are outlined in the following list, paired with specific study objectives and null hypotheses designed to address these questions.

The study questions and objectives include the following:

1. What proportion of the juvenile spring Chinook salmon released from Lewis River hatcheries are likely to residualize because they are precocious, or will become precocious within the same year?

   Study Objective 1: Quantify the proportion of males that are precocious at the time of release when released in October, or that would become precocious later in the year when released in February.

   H_0: The percent of the total population that are precocious males does not exceed 5% in a given year/cohort, or an average of 3% over 5 years/cohorts.

2. Do precocity rates differ between rearing and release groups?

   Study Objective 2: Compare the proportion of precocious males between release groups.

   H_0: No statistically significant difference in precocity (microjacks or minijacks) rates exists between rearing and release groups.

3. Are fish that fail to volitionally migrate (in the 2-week release period) more likely to be precocious?

   Study Objective 3: Compare the proportion of precocious males among the total population to the proportion among fish that fail to volitionally migrate.

   H_0: No statistically significant difference in precocity rates exists between the total population and fish that fail to volitionally migrate.

4. Can the methods used at other hatcheries to estimate precocity rates in spring release groups be employed at Lewis River hatcheries?

   Study Objective 4: Estimate precocity (minijack) rates in February using measurements of plasma 11-ketotestosterone (11-kt) levels.

   H_0: Precocity rate can accurately be estimated using 11-kt at the time of release (i.e., February).

   Study Objective 5: Estimate precocity (minijack) rates in February using the gonadosomatic index (GSI) method. Verify the accuracy of the GSI method with the measurements of 11-kt levels from the same fish.

   H_0: Precocity rate can accurately be estimated using the GSI method when testis size is measured at the time of release (i.e., February).

5. Can precocity be accurately estimated as early as February?

   Study Objective 6: Confirm the accuracy of precocity (minijack) rates estimated in February. Verify the accuracy of the February precocity estimate by comparing to GSI and 11-kt levels in a subset of fish held in the hatchery for approximately 1 to 2 months longer (e.g., mid-March to mid-April).

   H_0: Precocity rate can accurately be estimated using the GSI method at the time of release in February, 1 to 2 months earlier than other programs.

2.4 Informing Management Decisions

Data collected in this study would respond to Objective 4 in the Draft 2020 Hatchery and Supplementation Plan: Adopt strategies that limit potential post-release ecological interactions between hatchery and natural origin recruit-listed species. Key question F is "What is the average precocity rate for each species prior to scheduled releases?"

It is important to note that the results of this monitoring inform and should be considered in the context of the ongoing *Spring Chinook Rearing and Release Evaluation* (AOP – Appendix D).

This study would improve upon sampling described in the *Lewis Spring Chinook Rearing and Release Plan* (AOP – Appendix D) which includes a non-lethal visual screening of fish color, body shape, and manual compression to identify mature males, a metric that can be objective and inaccurate depending on rearing group characteristics and time of year relative to the typical spawning times. The existing plan also includes a lethal subsample of a small number of fish at the time of release to verify that testis development aligns with outward appearance of the fish. The study plan described in this document substantially increases the number of fish to be lethally sampled in order to more accurately estimate precocity rates.

Results of this study will be used to estimate the potential contribution of hatchery-produced precocious males to the impacts on listed species in the Lewis River. If warranted, hatchery programs may identify methods for reducing the number of precocious males and thereby reducing their impacts.

Methods employed in other hatchery programs to reduce precocity rates include the following:

- Delaying emergence timing by controlling water temperature during incubation or moderating growth during early ponding (Beckman et al. 2007)
- Reducing growth rates in fall and winter to mimic natural growth patterns (Larsen et al. 2006; Adelizi et al. 2017, Harstad et al. 2018)
- Reducing size-at-release targets (Aubin-Horth et al. 2004; Larsen et al. 2006; Harstad et al. 2014; Larsen et al. 2017)

Additionally, this hatchery program may consider measures that reduce survival rate within a cohort in the first year of life, but also reduce the number of released fish that are precocious males or fish that will become precocious males.

These considered measures include the following:

- Culling observed precocious males

- Culling any fish that fail to volitionally migrate
- Releasing fish in the fall to avoid effects of high growth on precocity rate due to warm fall water temperatures and abundant winter food availability in the hatchery environment

3 Field Methods

Precocious male maturation can be observed by several methods. Some males released in February of their second year may become sexually mature in their second fall; however, they would not show external signs of precocity this early, at least 6 months prior to the typical spawn timing. The hormonal cascade that triggers maturation and testis growth would have already been initiated in the previous fall (Campbell et al. 2003). The two most thoroughly validated indexes of maturity by peer-reviewed laboratory studies are 11-kt levels in the blood plasma (Larsen et al. 2004; Harstad et al. 2014; Medeiros et al. 2018) and testis weight compared to total body size (GSI; Pfannenstein 2019). It may be possible to discern maturing males using these methods as early as mid-February; however, differences between immature and mature groups are more easily discerned later in March and April (Larsen et al. 2004).

Measuring 11-kt requires extracting blood from lethally sampled fish, separating plasma from the blood cells using a centrifuge in the field, and measuring plasma hormone levels in a laboratory using a commercially available enzyme-linked immunosorbent assay.

Sampling (GSI) involves carefully dissecting and weighing testes using a highly sensitive laboratory-grade balance. The testes of males that have initiated maturation are larger relative to their total body size than immature males. Sampling (GSI) has been vetted by the U.S. Fish and Wildlife Service as a method for monitoring precocity among the Leavenworth National Fish Hatchery facilities and is currently being used to inform the management of hatchery programs in the Mid-Columbia River (Pfannenstein 2019; Snow et al. 2019). A GSI sampling equipment list is provided in Attachment A, and a field form to use for sampling is provided in Attachment B.

With either method, the distribution of the data tends to be bi-modal and precocious males are identified as individuals with 11-kt or GSI levels above a derived threshold level that separates the two modes.

Field sampling protocols for each method are as follows:

- Randomly collect fish from each rearing and release group to be sampled by cast net or dip net
- Transfer fish to large tubs for holding prior to sampling
- Control water temperatures and aerate holding water with flow through water source or with aeration, as needed
- Do not crowd fish into undersized containers

4 Visual Sampling – October Releases

At the time of spawning (August through early October), fully mature males can be easily identified by their darkened body color and rounded belly and can be identified with a non-lethal screening (Figure 4-1). Their testes grow to fill most of their abdominal cavity, and milt can be expressed by gently squeezing the fish's abdomen in an anterior to posterior motion.

Set up stations with fish length and weight measurement with datasheets, clipboards, pencils, length boards, and field balances. Each station should also have a container for anesthetic bath (e.g., in 5-gallon buckets), and a container for recovery.

Lightly anesthetize approximately 5 fish at a time in a mixture of MS-222 and sodium bicarbonate. A mixture of approximately 0.05 grams to 0.10 grams of MS-222 per gallon of water buffered with sodium bicarbonate in a ratio of 1:1 to MS-222 is adequate for sedation of salmonids.

Measure fork length, weight, and note smolt index (SI) (from 1 to 3; Figure 4-2) or note if precocious. Gently squeeze fish that appear to be precociously mature to confirm milt expression. Euthanize precocious males in a lethal dose of MS-222.

Move immature fish to a container with an adequate volume of fresh, cold, oxygenated water for recovery. Return fish to rearing ponds once recovered (approximately 10 minutes after transfer to recovery containers).

Figure 4-1. Example of a precociously mature male spring Chinook salmon (fish at the top of the photograph) compared to immature fish from the same rearing pond at Lewis River Hatchery.

Figure 4-2. Examples of fish Smolt Index (SI). A smolt index of 1 indicates lack of signs of smolting with dark parr marks (bottom), an SI of 2 indicates a transitional stage with faded marks (middle), an SI of 3 indicates a smolt with silver scales and no parr marks (top). Photo credit: Pfannenstein 2019.

5 Plasma 11-Ketotestosterone and Gonadosomatic Index Sampling – February Releases

GSI sampling may be paired with 11-kt assay to confirm accuracy of the GSI method for this stock at the chosen sampling time. Measuring plasma 11-kt and GSI requires lethal sampling of a subset of each release group.

Set up stations for fish length/weight measurement and for dissection with razor blades, Natelson tubes or syringes for blood collection, datasheets, clipboards, pencils, length boards, field balances, fish number tags, absorbent pads, gonad dissection tools, weigh boats, and laboratory balance.

Lethally anaesthetize 5 to 10 fish at a time with a lethal dose of MS-222.

Measure weight, length, note smolt index, and place a numbered tag on the body of each fish.

5.1 11-Ketotestosterone Sampling

After severing the caudal peduncle with a razor blade, collect blood from the caudal vein using heparinized Natelson blood collecting tubes (Fisher Scientific Cat. No. 02-668-10). Separate the plasma from blood samples taken from all male fish by centrifugation at 3,000 × *g* (gravitational force equivalent or g-force) for 5 minutes; transfer the plasma to a new, labeled tube using a glass pipet. Keep plasma cold during processing and transport frozen on dry ice for long-term storage at −80°C. Plasma should be separated in the field prior to freezing to avoid lysing the red blood cells, which can interfere with the assay chemistry. Plasma 11-kt levels (nanogram per milliliter) are determined by an enzyme linked immunosorbent assay, with laboratory methods according to the methods of Cuisset et al. (1994), used by Larsen et al. 2004 and several similar studies.

5.2 Gonadosomatic Index Sampling

After collecting the blood, set groups of 15 to 20 fish on absorbent pads for dissection. Cut open the abdomen from vent to anterior using a shallow incision. Make a cut behind the gill opercle. Open the body cavity and remove the guts, taking care not to puncture the air bladder. Identify fish sex. Remove the testes of male fish by carefully grasping the anterior end of the testes and pulling them away from the swim bladder. Remove testes by pulling or snipping the posterior ends free from the body cavity.

Weigh each pair of testes to the nearest 0.001 grams to calculate GSI index (testes weight/body weight × 100). Testis weights should be measured indoors using a laboratory-grade balance.

6 Metrics

Record the following data for fish in the field:

- Fish ID No.
- Pond number
- Fork length (millimeters)
- Fish weight (grams)
- Smolt Index (0-3)
- Markings (i.e., AD clip and CWT tag)
- Visual maturation stage (October releases, 1 = immature, 2 = mature)
- Sex (February releases)
- Gonad weight (grams, February releases)
- GSI (February releases)

Laboratory-measured 11-kt levels of individual fish shall be provided in association with a given fish identification number.

7 Sampling Frequency

Table 2 summarizes a suggested sampling schedule for spring Chinook salmon. Also shown are the study objective being addressed by each sampling event, the sampling frequency, and suggested sample sizes.

Table 2. Sampling Schedule for Spring Chinook Salmon Precocity

Sampling Date	Objective	Sample Period	Release Group	Rearing Treatment	Release Month	Size at Release (fpp)	Method	Number Sampled
June 1	1	Pre-volitional release	No. 5	No transfer	June	80	Visual, Milt	Up to 300[1]
July	1	Pre-volitional release	No. 4	June transfer	October	12	Visual, Milt	300
July	2	Pre-volitional release	No. 3	December transfer	February	8	Visual, Milt	300
July	2	Pre-volitional release	No. 2	December transfer	February	12	Visual, Milt	300
July	2	Pre-volitional release	No. 1	June transfer	February	8	Visual, Milt	300
August	1	Pre-volitional release	No. 4	June transfer	October	12	Visual, Milt	300
August	2	Pre-volitional release	No. 3	December transfer	February	8	Visual, Milt	300
August	2	Pre-volitional release	No. 2	December transfer	February	12	Visual, Milt	300
August	2	Pre-volitional release	No. 1	June transfer	February	8	Visual, Milt	300
September	1	Pre-volitional release	No. 4	June transfer	October	12	Visual, Milt, Dissections[2]	300
September	2	Pre-volitional release	No. 3	December transfer	February	8	Visual, Milt	300
September	2	Pre-volitional release	No. 2	December transfer	February	12	Visual, Milt	300
September	2	Pre-volitional release	No. 1	June transfer	February	8	Visual, Milt	300
October 15	3	Post-volitional Release	No. 4	June transfer	October	12	Visual, Milt	300
February 1	1, 2, 4, 5	Pre-volitional release	No. 3	December transfer	February	8	Visual[3], GSI, 11-kt	300
February 1	1, 2, 4, 5	Pre-volitional release	No. 2	December transfer	February	12	Visual, GSI, 11-kt	300
February 1	1, 2, 4, 5	Pre-volitional release	No. 1	June transfer	February	8	Visual, GSI, 11-kt	300
February 15	3	Post-volitional Release	No. 3	December transfer	February	8	Visual, GSI, 11-kt	300
February 15	3	Post-volitional Release	No. 2	December transfer	February	12	Visual, GSI, 11-kt	300
February 15	3	Post-volitional Release	No. 1	June transfer	February	8	Visual, GSI, 11-kt	300
March 15	4, 5	Hold-back	No. 3	December transfer	February	8	Visual, GSI, 11-kt	300
March 15	4, 5	Hold-back	No. 2	December transfer	February	12	Visual, GSI, 11-kt	300
March 15	4, 5	Hold-back	No. 1	June transfer	February	8	Visual, GSI, 11-kt	300

Notes:
1. Actual sample number will depend on the number of available fish.
2. A subset of the sample in 2020 will be dissected to confirm developmental of non-milting individuals.
3. A visual assessment will be made to quantify any holdover fish and any microjacks should not be included in the sample for minijack precocity.

Fish will be sampled just prior to release in October, which is the end of their typical spawning period, and just prior to release in February. A subset of fish from each treatment group (except the October release group) will be held in the hatchery to sample GSI and 11-kt in mid-March to verify the accuracy of the February sample and confirm the earliest timing (early February or mid-March) for discerning differences in these metrics in the spring.

Precocity rate can be highly variable from year to year within a hatchery population; therefore, monitoring should be carried out for at least 3 years to discern a trend in the total population, and among treatment groups within the total population.

8 Data Management

PacifiCorp or their contractors will manage data with their own specific quality assurance/quality control measures. PacifiCorp is responsible for storing the data files for future reference and reporting results to the Federal Energy Regulatory Commission on an annual basis.

9 Analytical Methods

The analytical method relies upon discerning fish that are immature from those that are mature based on the separation of two modes in a histogram of GSI values or 11-kt values of the total population of male fish.

In previous hatchery studies, assessment of approximately 150 males per treatment group has been an adequate sample size for determining significant differences between groups using GSI and 11-kt measurements that inherently have some error associated with dissection and processing of samples. A total of 300 fish must be euthanized in order to obtain approximately 150 males in a population with a 1:1 sex ratio.

The first step is to calculate the GSI and condition factor for each fish, then determine the GSI threshold using a mixture model which distinguishes immature from mature males. The equations for GSI and condition factor are below, and the R code for determining GSI threshold is included as Attachment C (developed by Medeiros et al. 2018 and used by U.S. Fish and Wildlife Service, as described in Pfannenstein 2019).

- Calculate GSI (gonad weight (g) / fish weight (g)) *100)
- Calculate Condition Factor (K = (10^5) * fish weight (g) / length^3(mm))
- Calculate the Log10(GSI) to determine the GSI threshold (mixture model)

Based on the threshold, calculate the following: counts, percentages, average length, and condition factor for mature and immature males. A table will summarize these metrics, as well as the visual counts for immature and mature fish and the percentage of mature fish.

Additional statistical analyses for the comparisons by analysis of variance (e.g., ANOVA) will be used to compare precocity rates between the four different release groups across at least 3 years of monitoring.

10 Assumptions

The ability to test some of the assumptions is difficult.

The assumptions identified in the following list are common to physiology studies:

1. The method for detecting early testis maturation used in other hatchery programs will be applicable to fish reared and released at Lewis River hatchery facilities.
    a. It is assumed that testis growth or maturation will be observable at the time sampling occurs.
    b. When sampling in October, at the time of year that fish would become sexually mature, this assumption has been supported by preliminary sampling in which precocious males were easily identified by their body coloration and milt expression.
    c. When sampling spring Chinook salmon to be released in February, it may be necessary to hold a representative subset of fish in the hatchery at least 1 month longer to allow testis growth to progress enough to discern males destined to become mature from immature males.
2. Spring Chinook salmon released in June (release group No. 5) will not be assessed for precocity using GSI or 11-kt, but may be assessed with visual techniques immediately prior to release.
    a. Spring Chinook salmon released earlier than their first fall would be too small to be assessed for precocity using these methods, nor have these methods been verified in other settings for fish in their first year (fry).

11 Deliverables

A technical memorandum will be provided describing and analyzing GSI sampling data, and final results will be included in PacifiCorp's *Hatchery and Supplementation Program Annual Report*.

For each release group, the following metrics will be reported with measures of the spread in the data, accuracy, and precision:

- Estimated precocity rate
- Sex ratio
- Average fork length/weight
- Condition factor
- Range in smoltification (smolt index)

Statistical comparisons will be made between rearing and release groups, and biologically significant differences between groups will be identified.

12 Limitations and Specific Concerns

Differentiating mature and immature spring Chinook salmon has not been routinely performed in other hatcheries as early in the spring as February. Holding fish longer until mid-March could increase confidence in identifying precocious males; however, in-hatchery mortality may increase during this added holding period. Existing literature (e.g., Larsen et al. 2004) indicates testis growth in precocious males can occur as early as January or February prior to spawning; however, in other hatchery monitoring studies, these methods have been used on fish released later in March or April (Pfannenstein 2019) and major differences in 11-kt and testis growth may not be discernable in February (Medeiros et al. 2018).

A preliminary visual examination of dissected fish at Lewis River Hatchery carried out on January 31, 2020, showed variability in testis size among the fish sampled (100 per rearing group), though there were different levels of variability depending on the group.

Assessing 11-kt levels in sampled fish may also be necessary to verify maturation determinations made based on GSI.

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Attachment A - GSI Supplies List (from Pfannenstein 2019)

Daily Consumables	Length and Weight Station
Data sheets: Length/weight sheet + gonad weight sheet (Rite in the Rain)	Tricane Methanesulfonate (MS 222)
Paper number tabs (Rite in the Rain)	[1] Tub for fish
Paper towels (brown single fold, ~100/pack)	[1] Dip net
Absorbent lab paper to cover work surfaces (roll)	[1] PIT tag (PIT) scanner + [1] stand
Garbage bags	[4] large sponges + [2] cookie trays
	[1] Scale for weights + [1] smolt weight pan
	[1] Length board

General	Dissecting Station
[5] Clipboards	[1 or 2] Micro scale (minimum power 0.001 g) + power cords
[5] Mechanical pencils + lead	[4] Scissors + [4] tweezers
[2] Tables	[2] Buckets for garbage (5 gallon)
[4] Chairs	S/M/L glove boxes
[4] Buckets to raise table (small white)	Weigh boats for scales
[2] 5-gallon buckets for fish	Portable lights
[2] Aerators	
[2] Power strips	
[2] Extension cords	
Sharpies	
Extra batteries (9 volt + AA)	
Camera/iPad	
Duct tape	

Notes:
Bracketed numbers are minimum numbers needed for one crew (4-6 people) for 300 fish.
The mention of trade names or commercial products in this document does not constitute endorsement or recommendation for use.
Vendors Used:

- VWR - Absorbent lab paper with Leak-proof Barrier 20"x300' (VWR)
- Fischer Scientific - Scissors/Forceps Item #: 08-935, 08-940, 08-875, 08-880 (straight scissors and curved forceps preferred by most samplers, but not all)
- Ohaus - Micro scale Adventurer Pro Model: AV264C

Attachment B – Field Form (modified from Pfannenstein 2019)

Attachment C – Sample R Code

```
####################################################
####################################################
## WNFH SCS 3 year summary
## file: WNFHSCS.csv
## 14 columns, 4951 rows
##
## Columns:
## 1-Facility, 2- BY, 3- Species, 4- Sex,
## 5- Length, 6- Weight, 7- VisualMat, 8- GonadWt, 9- K,
## 10- GSI, 11- log10GSI, 12- GSI_Mat, 13- Smolt Index
##
## Rows:
## (1:300) BY14
## (301:600) BY15
## (601:900) BY16
##
## male14 <- WNFHSCS[5:157, c("log10GSI")]
## male15 <- WNFHSCS[304:453, c("log10GSI")]
## male16 <- WNFHSCS[601:744, c("log10GSI")]
####################################################
####################################################
# Load the appropriate packages: tools, install packages, CRAN, package
install.packages("crandatapkgs")
library(mixtools)
library(Hmisc)
#import dataset
```

```
attach(WNFHSCS)

#################################################

# WNFH SCS Mixture Model

#################################################

op=par(mfrow=c(1,1))

male14 <- WNFHSCS[5:145, c("log10GSI")]

#view data in basic histogram

model=normalmixEM(male14)

plot(model, whichplots = 2, breaks=20)

#determine cutoff

index.lower <- which.min(model$mu)

find.cutoff <- function(proba=0.5, i=index.lower) {

f <- function(x) {

proba - (model$lambda[i]*dnorm(x, model$mu[i], model$sigma[i]) /

(model$lambda[1]*dnorm(x, model$mu[1], model$sigma[1]) + model$lambda[2]*dnorm(x,
model$mu[2], model$sigma[2])))

}

return(uniroot(f=f, lower=-2, upper=2)$root) # -2,2 may work 0,3 may work

}

cutoff <- c(find.cutoff(proba=0.5))

h <- hist(male14,ylim=c(0,50),breaks=20, main = "BY14")

xfit <- seq(-2.6,1,length=100)

yfit1 <- model$lambda[1]*dnorm(xfit,mean=model$mu[1],sd=model$sigma[1])

yfit2 <- model$lambda[2]*dnorm(xfit,mean=model$mu[2],sd=model$sigma[2])

yfit1 <- yfit1*diff(h$mids[1:2])*length(male14)

yfit2 <- yfit2*diff(h$mids[1:2])*length(male14)

v1 = seq(-2.7,0.2, length=30)
```

```
v2 = c(-2.7, -2.6, -2.5, -2.4, -2.3, -2.2, -2.1, -2.0, -1.9, -1.8, -1.7, -1.6, -1.5, -1.4, -1.3, -1.2, -1.1, -1.0, -0.9, -0.8, -0.7, -0.6, -0.5, -0.4, -0.3, -0.2, -0.1, 0, 0.1, 0.2)

#plot pretty graph

hist(male14, breaks = 20, density = 20, col = "purple", xaxt="n", xlab = "Log10 GSI", ylim = c(0, 30), main = "WNFH BY14 SCS")

lines(xfit, yfit1, col="red", lwd=2)

lines(xfit, yfit2, col="blue", lwd=2)

axis(side = 1, at = v1, labels = v2, cex.axis=.75, las=2)

abline(v=cutoff, col="green", lty=2, lwd=2)

text(-0.35,25, paste("Maturation Threshold", "\n =", round(10^(cutoff), 2),"GSI"))

text(-0.35, 12, "Maturation Rate= 9.2%")

####################################################

# 2014-2016 WNFH SCS Male Fork Length Vs Maturation

####################################################

op=par(mfrow=c(2,2))

male14 <- WNFHSCS[5:157, c("GSI_Mat", "Length")]

attach(male14)

mature<-male14[which (GSI_Mat=="2"), ]

immature<-male14[(GSI_Mat=="1"), ]

bins<- c(75, 80, 85, 90, 95, 100, 105, 110, 115, 120, 125, 130,135, 140,145, 150, 155, 160, 165, 170, 175, 180, 185, 190)

hist(immature$Length, breaks=bins, col="darkblue", ylim=c(0,45), main = "Males BY14", xlab = "Fork Length (mm)", xaxt="n")

hist(mature$Length, breaks=bins, col = "lightblue", ylim=c(0,25), add=TRUE)

legend("topright", c("Immature", "Mature"), col = c("darkblue", "lightblue"), pch= 15, bty = "n")

axis(side=1, at = c(75, 80, 85, 90, 95, 100, 105, 110, 115, 120, 125, 130, 135, 140, 145, 150, 155, 160, 165, 170, 175, 180, 185, 190), labels = NULL, las=2)

####################################################

# 2014-2016 WNFH SCS All Fork Length Vs Maturation
```

```
######################################################

## (1:300) BY14

all_FL14 <- WNFHSCS[1:300, c("GSI_Mat", "Length")]

attach(all_FL14)

mature<-all_FL14[which (GSI_Mat=="2"), ]

bins<- c(75, 80, 85, 90, 95, 100, 105, 110, 115, 120, 125, 130,135, 140,145, 150, 155, 160, 165, 170, 175,
180, 185, 190)

hist(all_FL14$Length, breaks=bins, col="forestgreen", ylim=c(0,80), main = "All Fish BY14", xlab = "Fork
Length (mm)", xaxt="n")

hist(mature$Length, breaks=bins, col = "gold", ylim=c(0,25), add=TRUE)

legend("topright", c("All Fish", "Mature Males"), col = c("forestgreen", "gold"), pch= 15, bty = "n")

axis(side=1, at = c(75, 80, 85, 90, 95, 100, 105, 110, 115, 120, 125, 130, 135, 140, 145, 150, 155, 160,
165, 170, 175, 180, 185, 190), labels = NULL, las=2)

text(170, 25, "9.2%", cex = 2)

text(170, 35, "Male Mat. Rate", cex = .75)

##
```

Strategy G: Genetic Risk Monitoring

Objective 7 – Methods to evaluate the genetic risks associated with hatchery programs on naturally spawning listed populations in the North Fork Lewis River

INTRODUCTION

In the North Fork (NF) Lewis River Basin, five hatchery programs operate to produce winter steelhead, coho salmon, and spring Chinook. The cumulative goal of these five programs is to provide harvest opportunities and support re-introduction efforts upstream of Merwin Dam. Hatchery supplementation comes with risks to natural-origin populations. Specifically, genetic risks occur when hatchery- and natural-origin fish interbreed and these risks generally fall under one of three categories: reduction of within-population diversity, reduction of among-population diversity, and domestication (Anderson et al. 2020; Busack and Currens 1995; Naish et al. 2007). Based on these risks, Objective 7 of the H&S Plan asked four (key) questions aimed at assessing the risk of Lewis River hatcheries on natural populations of salmon and steelhead:

- 7A: Have the Lewis River hatchery programs impacted the **among-population diversity** of naturally spawning populations?
- 7B: Have the Lewis River hatchery programs impacted the **within-population diversity** of naturally spawning populations?
- 7C: Have the Lewis River hatchery programs increased the risk of **domestication** for naturally spawning populations?
- 7D: Have the Lewis River hatchery programs impacted the **phenotypic diversity** of naturally spawning populations?

Given that the long-term fitness of a population is related to maintaining its genetic diversity, it is important to monitor the genetic status of the natural-origin populations and their relation to hatchery-origin programs over time. With any time-based evaluation, the earliest set of collections and associated metrics establish "the baseline". To assess the full impacts of any hatchery program on natural-origin populations, the baseline would ideally occur before hatchery intervention and subsequently be monitored to reveal any changes resulting from the operation of the hatchery program. Most hatchery programs pre-date genetic monitoring, including the collection of tissues that could be used for genetic analysis. Therefore, contemporary collections and genetic markers must be used to evaluate the status of current biological populations and inform the operation of hatchery programs moving forward.

In the Lewis Basin, salmon and steelhead hatcheries have operated for many decades. Although some genetic studies have been done, in general, baseline levels of genetic diversity and domestication risks have not been established for Lewis Basin winter steelhead, Chinook salmon, or coho salmon. Therefore, the goal of this initial implementation plan is to establish baseline levels of diversity and domestication risks by generating a suite of metrics that are identified in Objective 7 of the H&S. Although this implementation will address all four key questions, the primary focus of this plan is on collecting and analyzing data associated key questions 7A (among-population diversity) and 7B (within-population diversity). Key questions 7C (domestication) and 7D (phenotypic diversity) are mostly covered in other objectives of the H&S Plan. Namely, estimates of pHOS and PNI are outlined in Objective 8 and phenotypic metrics related to adult and juvenile migration timing, age at maturity, fecundity, etc. are collected as part of Objectives 3, 4, and 6 and included within handling and sampling protocols of the program.

GENERAL APPROACH

This plan outlines the beginning phases of a monitoring strategy to address the four key questions identified in Objective 7 of the H&S Plan. For the first two key questions (7A and 7B), genetic data will be used to evaluate the impact of hatchery propagation on within and among-population genetic diversity of naturally spawning ESA-listed populations of salmon and steelhead in the NF Lewis River. Briefly, samples from hatchery- and natural-origin collections will be genotyped using SNP-based markers and analyzed to evaluate genetic relationships of hatchery- and natural-origin biological populations of interest. The initial analysis will be used to identify genetically distinct biological populations and establish baseline levels of genetic diversity. These baseline levels of genetic diversity can then be paired with collections taken in future years to reveal changes over time. To initiate this effort, the existing species-specific genetic baselines will first be examined to ensure that a representative collection exists for all biological populations of interest. Supplemental samples will be collected and added as needed. For the last two key questions (7C and 7D), biological data and metrics that are primarily collected and reported in other H&S objectives will be used to assess and report on domestication risks and phenotypic diversity.

METHODS

Focal species

Genetic monitoring and evaluation will focus on NF Lewis River populations of winter steelhead, coho salmon, and Chinook salmon, including both spring- and fall-run Chinook stocks. Although there are no fall Chinook hatchery releases in the Lewis Basin, hatchery spring Chinook may adversely impact ESA-listed populations of natural-origin fall Chinook due to the spatial and temporal overlap of spawning adults as well as hatchery-produced spring Chinook precocial males released in the fall.

Area of focus

Genetic monitoring and evaluation will encompass the designated boundaries of NF Lewis recovery populations (LCFRB 2010). This area includes the mainstem Lewis River and its tributaries above and below Merwin Dam with two main exceptions. First, the EF Lewis River will only be included in the monitoring boundaries for Chinook but not coho and winter steelhead. Based on recovery population designations, the NF and EF Lewis rivers are part of the same population designation for tule fall Chinook while the two rivers are separate recovery populations for coho and winter steelhead. Second, the monitoring boundaries above Merwin Dam will include only the watersheds upstream of Swift Dam until salmon or winter steelhead are transported into basins that drain into Merwin or Yale Reservoir.

As with any hatchery program, the Lewis River hatchery programs can create genetic risks to populations outside of the Lewis River basin if hatchery fish stray and spawn outside the NF Lewis Basin. The responsibility and obligation to monitor and alleviate any adverse effects from NF Lewis River hatchery programs outside the NF Lewis Basin has not been determined.

Biological populations of interest

To assess the genetic risks of hatchery programs on salmon and winter steelhead populations, the existing genetic diversity and structure of the Lewis River populations must first be characterized (i.e., baseline data). Like other watersheds, the NF Lewis populations may consist of several biological populations, which are defined as aggregations of spawning fish isolated in space and/or time. The degree of genetic structure among biological populations will depend on many factors including the amount of genetic exchange among populations (e.g., straying), the size of the population, and the length of time the populations have been isolated. The correct interpretation of genetic metrics relies on genetically characterizing tissue collections that are representative of each biological population.

Based on existing knowledge of salmon and winter steelhead populations within the Lewis Basin, we have identified five biological populations of interest for each species (Table 1). The first set of biological populations consists of the broodstock for each segregated hatchery program. Segregated hatchery programs are separate biological populations since only hatchery-produced fish are used as broodstock. The degree of genetic separation depends on the degree of interbreeding of segregated program hatchery fish and wild fish on the spawning grounds. The other biological populations for each species consist of natural-origin spawners and broodstock of the integrated hatchery programs. Specifically, natural-origin spawners have been divided into four groups, which consist of mainstem and tributary habitats up and downstream of Merwin Dam. If run properly, salmon and winter steelhead produced in an integrated hatchery program should be biologically identical to those derived from natural spawning because some or all of the hatchery broodstock are natural-origin fish and interbreeding of hatchery- and natural-origin fish is intended on the spawning grounds. Therefore, broodstock from the integrated hatchery programs are not a separate biological population but will be a separate sample collection group that should be representative of the biological population(s) from which it was derived. Based on the proposed sample collection strategy (see below), the genetic evaluation will reveal similarities among these four groupings and may even reveal additional biological populations.

Genetic baseline

Regional genetics laboratories (WDFW, USFWS, CRITFC) have genotypes or tissue collections from many populations. This existing information may be useful for monitoring and evaluating Lewis River salmon and winter steelhead, and in some cases, the genetic data have already been analyzed and compiled in reports. Therefore, before any additional samples are collected or monitoring indicators are generated, available tissue collections, genetic data, and reported results will first be thoroughly examined to avoid unnecessary duplication of effort and to provide guidance for the next steps of baseline development. Specifically, we will assess whether samples from any biological populations of interest within the Lewis River basin (see Table 1) are represented in tissue collections or genetic datasets, and if so, whether samples were taken in such a way that the collections are representative of the biological populations. Representative collections include samples that were collected over multiple years and throughout the entire spawning spatial and temporal distribution. Once the baseline assessment is complete, representative collections that already exist and are deemed representative can be genotyped and plans can be made to collect samples from unsampled/under-sampled biological populations.

Table 1. NF Lewis biological populations of interest and the corresponding sample locations for tissue collections to be used in baseline development of winter steelhead, coho salmon, and Chinook salmon.

Biological Population	Description	Sample Locations	Winter Steelhead[1]	Coho Salmon	Chinook Salmon
Segregated hatchery	Segregated broodstock	Hatchery @ spawning for segregated broodstock	X	X	X
Lower mainstem	NF Lewis mainstem below Merwin Dam	Mainstem Lewis River - Eagle Island to Merwin Dam	X	X	X[2]
Lower tributaries	NF Lewis tributaries below Merwin Dam	Cedar Creek	X	X	X
		Johnson Creek	X	X	
		Houghton Creek	X	X	
		Hayes Creek	X	X	
		Ross Creek	X	X	
		East Fork Lewis			X[3]
		Allen Canyon (Mud Lake)		X	
Upper mainstem	NF Lewis mainstem above Swift Dam	Mainstem Lewis River - Eagle Cliff to Lower Falls	X[4]		
		Hatchery @ spawning for integrated broodstock[5]	X	X	
Upper tributaries	NF Lewis tributaries above Swift Dam	Swift Creek	X		
		Drift Creek	X		
		Range Creek	X		
		Diamond Creek	X		
		S15/S20	X		
		Pine Creek	X		
		Muddy River	X		
		Clear Creek	X		
		Clearwater Creek	X		
		Smith Creek	X		

[1] Samples, particularly in the upper basin, are expected to consist of resident and anadromous *O. mykiss*as well as potentially out-of-basin stocked rainbow trout (e.g., Goldendale strain)

[2] The collection period needs to go from approximately mid-September through mid-January to include all three stocks of Chinook – spring, fall (tule), and late-fall (bright) – in the lower mainstem.

[3] Tule fall-run Chinook from the NF & EF Lewis River are part of the same recovery population and thus samples of Chinook from the EF Lewis need to be collected and genotyped. At this time, whose obligation it is to collect these samples has yet to be determined.

[4] The steelhead genetic baseline should include samples of *O. mykiss* upstream of natural barriers from the Upper Lewis

[5] Sample collections from integrated hatchery broodstock programs are listed as part of the "Upper mainstem" biological population but may consist of individuals from multiple biological populations

Sampling Strategy

A detailed sample plan will not be developed until an assessment of existing baselines for each species has been completed. However, based on existing information, it is likely that additional tissue samples will need to be collected to establish representative genetic baselines for each species. Therefore, at this time, this plan provides a general framework for collecting tissue samples, which will be updated once a list has been compiled of additional samples that are needed for baseline development. The approach used to collect tissue samples can be divided into two general strategies based on the spawning location (hatchery vs. natural) for a specific sample collection.

For hatchery collections, tissue samples should be taken from adults that are used for broodstock either during their initial collection or at the time of spawn. As a last resort, samples can also be collected from hatchery offspring (embryos, parr, smolts) before release but this approach should generally be avoided. Hatchery broodstock collections should be spread over the total collection/spawn period, split between males and females, and collected roughly in proportion to the integration rate of NORs in the brood.

For naturally spawning populations, the general strategy will be to collect samples at a time and life stage that maximizes the likelihood that the sample represents an individual biological population in the area where it was sampled. In theory, samples can be collected from either adults or juveniles. However, there will likely be a preferred life stage based on the logistics of capturing fish to sample and the ability to sample isolated populations at a particular site. For instance, sampling adult steelhead on the spawning grounds is generally infeasible due to the lack of carcasses while sampling juveniles at smolts traps can result in a mixture of biological populations and be biased towards migrants. Therefore, samples from some biological populations can be collected from adults on the spawning grounds (e.g., lower mainstem spawners) but samples for other populations may need to come from juveniles as fry or parr during their first spring/summer rearing period prior to migration/mixing. Sample boundaries for each collection will be delineated based on known or presumed spawn timing for a given area.

One general exception to the sampling strategy for natural spawning population will be for populations in the upper basin. Although we currently do not have much information on the genetic status of upper basin populations, we do not expect there to be much structure for Chinook and coho given that these populations were extirpated for nearly a century and reintroduction has only occurred for a little over a decade (maybe 3 generations). Therefore, instead of sampling natural-spawning adults and/or their progeny in the upper basin, we plan to focus our efforts on lower basin populations and the hatchery programs that have largely been used to re-populate the upper basin. However, unlike Chinook and coho, steelhead were not extirpated from the upper basin. Resident O. mykiss persisted and population structure among some tributaries was discovered (Winans et al. 2018), which may resemble structure that existed pre-dam. Thus, samples from contemporary upper basin O. mykiss need to be collected/assembled. Samples collected at "pinch points" (e.g., Swift Fish Surface Collector) will not be appropriate because these locations will likely disproportionality sample migratory fish and may not be able to detect any effect hatchery resident rainbow trout plants are having on endemic populations. Existing samples from older collections (e.g., Winans et al. 2018 samples) may be available in lieu of new samples but need to be better assessed to ensure they are representative of the groupings outlined in Table 1.Sample Collections

The total number of tissue samples required per collection will depend on the study population(s), the metric(s) of interest, and the marker panel used. However, there is often a lack of information on the population to identify a specific number of samples needed. Therefore, geneticists generally recommend 50 – 100 samples per collection (i.e., putative population or cohort). These numbers are large enough to accurately estimate allele frequencies, especially for SNP markers, but small enough as to be tractable in the field (Koch et al. 2018). In some cases, collections may need to be higher to fully represent a particular population. For example, the lower mainstem Lewis River is comprised of three management populations of Chinook (spring-, fall/tule, and late-fall/bright years) that should be sampled representatively. Each collection should be collected over multiple years to capture the genetic variation of different brood classes. Specifically, samples of winter steelhead and Chinook should be collected across at least two return years while coho should be sampled across three years.

Upon capturing an individual fish to sample, tissue should be collected from the upper lobe of the caudal fin. Collect as much tissue as possible up to approximately 1/4" X 1/4" in size (hole-punch sized). Samples should be stored at room temperature on either a sheet of blotter (i.e., chromatography) paper or in individual coin envelopes. If tissue samples are stored in coin envelopes, it is advised to place the sample inside a folded piece of blotter paper to help dry the sample. At minimum, the following information should be taken for each sample: sample location, name of hatchery program (if applicable), collection date, species, sex (if known), adipose fin clip status (UM = intact, AD = removed), CWT status (beep = presence, no beep = absent), and PIT status (beep = presence/record number, no beep = absent).

Sample processing

Lab processing must be done using an approach that results in genotyping of SNP markers used in regional datasets to ensure compatibility. For example, the WDFW sample processing protocol starts with genomic DNA extraction and isolation using silica membrane column extraction kits following the manufacturer's protocols. DNA is then amplified and sequenced using an amplicon sequencing procedure (e.g., Genotyping in Thousands – GTseq – Campbell et al. 2015) to target a specific suite of SNP markers that are part of a SNP marker panel. Markers are sequenced on an Illumina™ next-generation sequencing platform and then separated by sample identifiers using a series of custom Perl scripts (*c.f.*, Campbell et al. 2015). Genotypes are assigned based on allele ratios by counting allele-specific amplicons at each locus. Appropriate marker panels for all three species exist and these panels are used in most regional genetics laboratories (e.g., WDFW, USFWS, CRITFC, ODFW).

Data processing

All genotyped data should be quality controlled before analysis including species identification, duplicate genotypes, contamination, and missing data. Duplicated or contaminated samples should be removed, as should samples with too much missing data (e.g., WDFW uses a threshold of >30% missing). Most collections and loci should be checked for conformation to Hardy-Weinberg expectations (HWE) including probability testing for HWE and linkage disequilibrium (LD). Loci that do not conform to HWE in most collections could be dropped from the analysis. One or both loci of pairs of loci with high LD could be dropped. Most currently used marker panels include both neutral and adaptive loci. Adaptive loci may, for example, be those that genetically identify sex or have a high correlation with run timing. For most analyses, adaptive loci should be identified and removed from further analysis.

Data Analysis

Objective 7 of the H&S Plan identifies specific indicators that will be used to monitor and assess the risks associated with Lewis River hatchery programs on natural spawning populations (Table 2). A description of each monitoring indicator is provided below, separated by its corresponding risk type, along with a brief summary of how the indictor should be generated and assessed. In general, the genetic- and biological-based metrics outlined below will be generated for each biological population of interest and hatchery program identified in Table 1. NF Lewis biological populations of interest and the corresponding sample locations for tissue collections to be used in baseline development of winter steelhead, coho salmon, and Chinook salmon. However, there will be some instances where a metric will need to be generated for a combination of populations due to logistical and pragmatic monitoring constraints. For example, most of the biological-based metrics will be generated for the entire upper Lewis natural spawning populations. Where necessary, metrics will also be generated for management-based populations (e.g., pHOS and PNI for the Lewis population of winter steelhead, which includes upper- and lower-, mainstem- and tributary-biological populations).

Table 2. Summary of monitoring indicators by genetic risk type and frequency of analysis.

Risk Type	Monitoring Indicators	Frequency
Among	- Pairwise genetic distance (F_{ST}) - Genetic distance metrics combined with dendrograms - Multi-variate clustering analyses	Generational (f3)
Within	- Effective Population Size (N_e) and Breeders (N_b) - Inbreeding coefficient (F_{IS}) - Average heterozygosity - Average per locus minor allele frequencies & Average number of alleles - Linkage Disequilibrium (LD)	Generational (f3)

Domestication	- PNI (pHOS, pNOB) - PEHC	Annual
Phenotypic	- Timing of juvenile outmigration, adult run- and spawn-timing - Size and age of juvenile outmigrants and returning adults - Fecundity of female spawners	Annual

Among-population diversity metrics

Among-population diversity is measured as relative genetic differences among biological populations. Hatchery production can reduce among-population diversity when hatchery fish successfully spawn with natural-origin adults from biological populations other than those used as broodstock. Reductions of among-population diversity may reduce the long-term viability of the metapopulation through genetic homogenization and outbreeding depression. The following genetic-based metrics will be used to assess among-population diversity.

Pairwise genetic distance (F_{ST})

Pairwise F_{ST} estimates the genetic variance due to structure among collections. Specifically, F_{ST} is the proportion of the genetic variance contained in a biological population (the S subscript) relative to the total genetic variance among the two populations (the T subscript). Values can range from 0 (no variance, i.e., no genetic distance; two collections are from a single biological population) to 1 (all variation is among collections; the two collections are from separate biological populations that do not interbreed). No theoretical minimum viable genetic distance has been identified. Instead, the goal is to avoid reducing genetic distances among populations via hatchery program activities. Pairwise F_{ST} uses allele frequency data and can be calculated with a variety of existing software (e.g., R package *hierfstat*; Goudet 2005). It should be evaluated for statistical significance (e.g., jackknife procedures). Allele frequency differences can be visualized using Principal Components Analysis (PCA) of collection allele frequencies and pairwise F_{ST} data can be visualized using heat maps or multidimensional scaling (e.g., Principal Coordinates Analysis; PCoA).

Genetic distance metrics combined with dendrograms

Population structure should be evaluated with other measures of genetic distance. Typically used measures include Cavalli-Sforza chord distance (Cavalli-Sforza and Edwards 1967) and Nei's genetic distance (Nei 1973). These measures and F_{ST} distances can be visualized using a bifurcating dendrogram (e.g., using the software PHYLIP, Felsenstein 1993). The statistical support for the genetic relationships of the dendrogram can be evaluated using bootstrap procedures. Each genetic distance and dendrogram method has assumptions that need to be met to avoid bias and so should be evaluated and chosen accordingly.

Multi-variate clustering analyses

Population structure should also be evaluated using multi-variate clustering algorithms including those with (e.g., STRUCTURE, Pritchard et al. 2000) and without (PCA, PCoA, Discriminant Analysis of Principal Components [DAPC, Jombart et al. 2008] or Factorial Correspondence Analysis [FCA, e.g., using software GENETIX, Belkhir et al. 2001]) underlying population genetic models. Analysis with population genetic models explicitly models mixed ancestry allowing inference of admixed individuals (e.g., hybrids) whereas analysis without underlying population genetic models does not.

Within-population diversity metrics

Within-population diversity describes the amount of genetic diversity within a biological population and is important for the long-term resilience of the population. Reduced within-population diversity is an indication of inbreeding (i.e., increased allelic identity by descent), which may lead to inbreeding depression and reduced long-term viability. Hatchery production increases the risks of reducing within-population diversity of natural-origin populations because hatcheries spawn only a subset (sometimes a very small subset) of the entire population which is then amplified via the increased survival afforded by hatchery rearing. Segregated programs which use only hatchery-produced fish for broodstock are especially susceptible to reductions in within-population diversity this way. The following genetic-based metrics will be used to assess within-population diversity.

Effective population size (N_e) and Effective number of breeders (N_b)

Effective population size (N_e) is the number of individual spawners in a population that contribute offspring to the next generation. Because genetic variation generally decreases with fewer effective number of spawners, N_e roughly indicates the amount of within-population genetic variation that exists. N_e is also an indication of the expected rate of loss of diversity due to drift, in that populations with small N_e are expected to lose diversity due to genetic drift faster than populations with large N_e. Hatchery propagation is known to dramatically decrease the N_e of wild populations due to the overrepresentation of hatchery-produced fish among the wild (i.e., Ryman-Laikre effect, Ryman and Laikre 1991). Generally, the effective size of a population is considerably less than the census size of a population (N). There is no consensus among experts on minimum viable N_e values but the general recommendation is to avoid reductions in N_e. Trends in N_e would provide an early warning capability for observing threats to viability (i.e., loss of genetic diversity through inbreeding) or genetic stochasticity before any indication is apparent from census estimates. N_e can be calculated in a variety of ways, of which some methods are equivalent to estimating the effective number of breeders (N_b) in a population (e.g., LDNE algorithms employed by the software NE ESTIMATOR; Do et al. 2014). It is important to account for overlapping generations in a population that can bias estimates of N_e or N_b (Waples et al. 2014).

Inbreeding coefficient (F_{IS})

The inbreeding coefficient (F_{IS}) can be used to gauge the level of inbreeding within a population. When related individuals mate, their offspring may be homozygous for a copy of an allele that is identical by descent from one of its ancestors. Like F_{ST}, F_{IS} is the proportion of genetic variance of individuals (the I subscript) relative to the total genetic variance of a biological population (the S subscript). F_{IS} ranges from -1 to 1. A negative F_{IS} indicates an excess of heterozygotes and a positive F_{IS} indicates an excess of homozygotes. The higher F_{IS} implies more inbreeding. F_{IS} is calculated using the same methods and software used to calculate F_{ST}.

Average heterozygosity (expected and observed, among loci, within-population)

The average heterozygosity is the fraction of loci within a population among individuals where an individual genotype consists of two different alleles. In general, the higher the average heterozygosity is for a population, the greater the genetic variability within that population. Observed and expected heterozygosity is calculated for each collection and compared. Expected heterozygosity is estimated assuming Hardy-Weinberg processes. Extreme deviations of expected from observed heterozygosity within a population is an indication that the population is small (i.e., subject to genetic drift) and/or non-randomly mating (increased inbreeding). Low observed and expected heterozygosity is an indication of reduced diversity. No threshold viable heterozygosity has been identified; therefore, the goal is to avoid reductions from baseline levels.

Average per locus minor allele frequencies and Average number of alleles within-population

Alleles may be lost or reduced in frequency due to a small census or effective population size and genetic drift or through inbreeding. Average allele frequencies and the average number of alleles per locus describe the amount of genetic variation among individuals within a population. Hatchery programs may reduce allelic diversity. No minimum viable allele frequencies or average number of alleles have been identified; therefore, the goal is to avoid reductions from baseline levels. The number of alleles detected is a function of the sample size. Therefore, the average number of alleles is typically standardized for sample size using rarefaction (e.g., using the R package *PopGenReport*, Adamack and Gruber, 2014). Allele frequencies can be evaluated visually (e.g., PCA plots of average allelic richness among all loci within a collection) and statistically (e.g., pairwise analysis of molecular variance [AMOVA] analyses, calculating heterozygosity and allelic richness).

Linkage Disequilibrium (LD)

Linkage Disequilibrium (LD) is the correlation of alleles among loci within an individual. Loci may be in LD because they are physically linked (near one another on a chromosome and as such are inherited together) or they may be statistically linked (e.g., alleles are correlated because of relatedness among individuals within a population). No minimum or maximum allowable LD target has been described. However, because increased LD indicates a reduction in diversity, advice is generally to avoid increasing LD. Hatchery activities may increase the amount of LD present due to relatedness among individuals. LD can be calculated in multiple ways. For instance, pairwise locus comparisons within a collection can be made with allelic correlation coefficients using PLINK (Purcell 2007; Purcell et al. 2007) or a probability test of LD using GENEPOP (Rousset 2008).

Domestication metrics

Domestication reduces the long-term fitness of populations through the proliferation of alleles which improve performance in domestic settings (i.e., hatcheries) while reducing performance in natural settings (domestication selection). However, there are currently no genetic techniques (e.g., domestication genes or markers) to assess the level of domestication within populations. Therefore, the following metrics based on biological data will be used to assess the potential of domestication due to artificial propagation.

Proportion hatchery-origin spawners (pHOS)

pHOS represents the proportion of naturally spawning adults that are hatchery-origin for a given population and return year. pHOS is a representation of potential interbreeding of hatchery- and natural-origin fish and therefore an index of gene flow between a hatchery population and its companion natural population. There are several ways to estimate pHOS, but in general, it is the estimated abundance of hatchery-origin spawners divided by the total natural spawner abundance (comprised of both hatchery- and natural-origin). The Hatchery Scientific Review Group (HSRG) has established pHOS standards to limit the impact of hatchery programs on natural populations (HSRG 2014, 2020). The specific limits are based on the type of hatchery program (integrated vs. segregated), the Biological Significance of a population (primary, contributing, stabilizing), and the four recovery phases (preservation, recolonization, local adaptation, and full restoration). Because of their expected high degree of domestication, segregated programs have the most restrictive pHOS limits. In general, lower levels of pHOS translate to smaller fitness reductions for wild populations.

<u>Proportion natural-origin brood (pNOB)</u>

pNOB represents the proportion of a hatchery broodstock composed of natural-origin adults each year. Similar to pHOS but more directly observable, pNOB is an index to estimate the amount of gene flow between hatchery- and natural-origin in the hatchery. pNOB is calculated as the number of natural-origin brood divided by the total number of adults used for brood (hatchery- and natural-origin). There are no specific standards for pNOB. Instead, pNOB targets are determined by the program type (i.e., pNOB = 0 for segregated programs) and PNI, which is calculated using pHOS and pNOB. In general, higher pNOB results in less domestication because of the reduced number of generations fish spend in the hatchery under the influence of domestication selection. However, depending on the recovery phase for a given population, lower levels of pNOB may be needed to achieve demographic replacement for natural-origin spawners.

<u>Proportion natural influence (PNI)</u>

PNI represents the collective effects of pHOS and pNOB in a single statistic and generally describes the percentage of time the genes of a composite population (i.e., a population composed of natural- and hatchery-origin spawners) spend in the natural environment. PNI describes the potential for interbreeding (i.e., gene flow) in both components (hatchery and natural) of a population under the influence of hatchery production. PNI is calculated as pNOB/(pNOB + pHOS). The interpretation of PNI is reliant on the assumption that actual interbreeding is directly related to proportions of hatchery and natural origin spawners and broodstock. The HSRG has identified lower limits on PNI based on the Biological Significance and current recovery phase of a population. The PNI standard is only applied to integrated programs as pNOB for segregated programs is zero making PNI also equal to zero. The higher PNI, the greater the strength of selection in the natural environment relative to that of the hatchery environment (i.e., less potential domestication).

<u>Proportion Effective Hatchery Contribution (PEHC)</u>

PEHC is similar to pHOS in that it measures gene flow between hatchery- and natural-origin fish on the spawning grounds. However, unlike pHOS which estimates the composition of offspring based on spawner composition, PEHC is calculated using genetic data of offspring to estimate parentage. Put another way, PEHC estimates the actual interbreeding that occurred between hatchery- and natural-origin fish based on analysis of genotypes. PEHC requires that the parental origin (hatchery or natural) be known. It may be possible to estimate PEHC via genetic parentage assignment, but currently PEHC is calculated using the clustering algorithms of the program STRUCTURE. Estimating PECH this way requires that the hatchery and natural populations are genetically distinct (i.e., distinguishable). Therefore, in the Lewis Basin, PEHC may be useful to estimate interbreeding between segregated hatchery program fish and natural-origin spawners depending on the degree of genetic differentiation but cannot be used for integrated programs.

Phenotypic diversity metrics

A potential result of reduced genetic diversity and domestication includes observable changes in phenotypic traits of naturally spawning populations. These traits undoubtedly have some genetic component; however, there are currently few genetic markers with known allelic associations with phenotype. Therefore, the following metrics based on important fitness traits will be used to assess phenotypic diversity.

Timing of juvenile outmigration, adult return, and spawning

The timing and duration of ontogenetic shifts should be tracked at various locations and life stages. For upper basin biological populations, adult return timing should be tracked for upper basin biological populations using census counts of adults at the Merwin Collection Facility (H&S Objective 3; M&E Objective 11) while juvenile outmigration timing should be tracked using expanded estimates of abundance at the Swift Surface Collector (M&E Objective 6). Where possible, spawning ground survey data should be used to estimate spawning timing though data is expected to be limited (M&E Objective 15). For lower basin biological populations, spawning ground survey data should be used to estimate spawning timing (H&S Objective 5). In general, spawn timing cannot be disentangled from adult return timing. Juvenile outmigration timing should be estimated based on hatchery-release data (H&S Objective 4) and, if possible, lower river smolt trapping operations (H&S Objective 6). When observers of adults and juveniles are not a census count, expanded estimates of abundance must be used to generate unbiased estimates of timing. Regardless of life stage, timing data can be summarized in many ways. At a minimum, timing data should be visualized with plots of relative and cumulative frequencies and quantified with point estimates of timing (e.g., median ± 95% CI). Adult returns and spawning timing should be compared between natural- and hatchery-origin fish as well as to broodstock collection within and among years. Outmigration timing should be tracked across years, and when possible, compared between upper- and lower-basin and HOR and NOR outmigrants.

Size and age of juvenile outmigrants and returning adults

The size, age, and size-at-age should be tracked for returning adults and outmigrating juveniles. The data collection locations and analysis should be almost identical to the guidance listed above for timing. In general, age data should be collected with scales and size data measured via fork length. Unless age and size data are collected from a censused population, similar to timing, data will need to be stratified by collection date and apportioned appropriately based on estimates of abundance. Regardless of life stage, age, and size data can be summarized in many ways. At a minimum, data should be visualized with plots (e.g., box plot of size-at-age, the proportion by age) and quantified with point estimates (e.g., median ± 95% CI of FL by age). Size and age data should be compared between natural- and hatchery-origin fish as well as to broodstock collection within and among years. Size and age data should be tracked across years, and when possible, compared between upper- and lower-basin and HOR and NOR outmigrants.

Fecundity of female spawners

Fecundity is the number of eggs (i.e., ova) that a female fish generates before spawning. Fecundity data should be collected using representatively sampled broodstock for each hatchery program. At a minimum, data should be summarized by quantifying the median (± 95% CI) fecundity by age and compared among years.

FREQUENCY AND TIMELINE

For genotypic-based metrics associated with among- and within-population diversity, sampling may occur on an annual basis, but analysis of those samples would occur on a generational scale except for genetic screening of broodstock (Table 2). NOAA recommends that genetic analysis occurs at least every third generation (f3). Before any additional samples are collected and baseline metrics are generated, an evaluation of the existing genetic baselines for winter steelhead, coho salmon, and Chinook salmon needs to be completed and should occur as soon as possible (ideally in 2022). Based on existing knowledge, additional tissue samples will likely need to be collected over the following two to three years (approx. 2023-2025) after which the first (i.e., baseline) genetic analysis should occur.

For biological-based metrics associated with domestication and phenotypic diversity, sampling, analysis, and reporting will occur on an annual basis (Table 2). These metrics are generated at a higher frequency because they are used to answer key questions from other objectives and can provide early warning indications of domestication. The baseline period (i.e., starting point) for biological-based metrics does not have to be the same as genetic-based metrics. Depending on the availability of data, biological-based metrics should be generated as far back as possible (as soon as possible) after which contemporary estimates will be added annually.

LIMITATIONS, CONCERNS, AND CAVEATS

- In general, every metric included in this plan comes with a suite of assumptions, which contextualizes, and in some cases limits, the interpretations of the results. Therefore, the assumptions of each metric must be listed and discussed upon reporting.
- The correct interpretation of results of genetic tests relies heavily on tissue collections being representative of the populations or groups they are intended to represent.
- Uncertainty of genetic tests is dependent on the number of samples collected and successfully processed.
- Given the long history of hatchery production in the Lewis River basin along with the biological and environmental complexities that govern the impacts of hatchery programs and their progeny on wild populations, it is unlikely that the metrics generated from this plan can definitively answer the four key questions identified in Objective 7 (i.e., have hatchery programs affected the diversity of natural-origin populations?). However, in combination, these metrics will assess the genetic status of Lewis River populations, qualitatively assess the impacts of current hatchery programs on natural-origin populations, and help guide future hatchery operations that promote long-term genetic viability.
- Among- and within-population genetic variation may change due to factors other than hatchery production, including gene flow (straying) from other populations and natural selection. The SNP markers intended to be used here may, with adequate baseline data, allow detection of gene flow from outside into the NF Lewis River.
- Biological-based metrics of phenotypic diversity likely vary due to a combination of genetic and environmental factors. Nonetheless, it may not be possible to definitively determine whether such changes are good or bad for the population in terms of survival and persistence.
- Many of the SNP markers are assumed to be neutral (i.e., are not linked to traits under selection). Violation of this assumption would lead to incorrect interpretation of the data. Genomic methods are available and can be used to look for regions of the genome that may

be under selection and associated with environmental variables. However, these methods are prohibitively expensive and are not currently part of routine hatchery monitoring.

- Genetic methods, technology, and markers continue to evolve and thus may substantially change between timesteps. In order to make valid comparisons among timesteps, the same marker panels must be used for all samples. Any substantial changes in method, technology, or markers may necessitate the re-processing of samples from earlier timesteps.
- The monitoring plan lacks monitoring of adaptive genetic diversity that uses genetic techniques. Some markers linked to adaptive diversity are known (e.g., Chinook/steelhead GREB markers, Chinook Y haplotype markers [age at maturity in males], Prince et al. 2017, Hess et al. 2016, McKinney et al. 2020) and could be used in monitoring efforts.
- Evaluating genetic- and biological-based metrics together provides the most comprehensive means of evaluating hatchery programs and the risks to natural spawning populations they affect. However, there is no consensus on how these metrics should influence adaptive management decisions.
- It is expected that Lewis River populations of salmon and steelhead will remain in the re-colonization phase well past the next rewrite of the H&S plan (scheduled in 2025) and hatchery supplementation will likely continue. However, Lewis River hatchery programs are scheduled for re-evaluation in the coming years through the development of "transition plans". Changes to broodstock management may subsequently impact genetic risks to natural-origin populations. Thus, this plan may need to be updated accordingly.

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Strategy H: Volitional Release

Strategy H: Volitional Release Strategies

Introduction

The North Fork Lewis River (Lewis River) Hatchery and Supplementation Subgroup discusses release strategies and monitoring efforts for Lewis River Hatchery programs, to be documented in the Annual Operating Plan. In particular, implementation and monitoring of volitional or forced release methods should be informed by regionally accepted guidelines and best available science. This memorandum summarizes guidelines developed for the Columbia Basin by the Hatchery Scientific Review Group, draft Hatchery and Genetic Management Plans (HGMPs) for Lewis River programs, HGMPs for nearby hatchery systems, and relevant literature on the topic. Though not an exhaustive research effort, this memorandum is intended to inform Hatchery and Supplementation Subgroup discussions. Details for the Lewis River program release methods are included in the Annual Operating Plan.

Hatchery Scientific Review Group

Hatchery Scientific Review Group (HSRG) guidelines recommend volitional release and removal of residuals to minimize adverse ecological impacts, but the guidelines do not include details on the timing or duration of volitional period. The following are the relevant HSRG guidelines on volitional release:

- Minimize adverse ecological interactions by making reared and released fish as similar biologically to their natural counterparts, or, segregate hatchery fish in time and space (HSRG, 2014).
- Maximize survival of hatchery fish so that unnecessary releases are minimized (HSRG, 2014).
- Recommends volitional release of smolts, in particular for steelhead, to reduce interactions with natural-origin juveniles (see Snow et al., 2013, per HSRG, 2014).
- Adoption of volitional release with removal of residuals (fish that become residents rather than go to sea) may increase the long-term survival of released fish while decreasing negative ecological interactions (HSRG, 2009).

Hatchery and Genetic Management Plans for the North Fork Lewis River

There are five HGMPs pertaining to the Lewis River programs: spring Chinook, coho type N (late), coho type S (early), winter-late (endemic) steelhead (integrated), winter steelhead (segregated), and summer steelhead. In each HGMP, volitional release strategies for each species are discussed. The HGMPs are similar in that they all state the fish should be released at a fully-smolted stage to reduce negative ecological interactions. When volitional release periods are specified (for spring Chinook salmon and coho salmon), they are 6 weeks long with the remainder of fish being forced out. Details about volitional releases for each HGMP are included in the following six sub-sections:

1.1.1 Lewis River Type-N Coho Salmon – April 2015

- Fish are released at a time, size, and the system and life history stage to foster rapid migration to marine waters, and to allow juvenile listed fish to grow to a size that reduces potential for fish predation (see Sections 2.2.3 and 10.11 in WDFW and PacifiCorp, 2015a).
- Juveniles are released at fully-smolted stage to benefit juvenile to adult survival rates and reduce the likelihood for residualism and negative ecological interactions (Section 3.5.5 in WDFW and PacifiCorp, 2015a).
- Lewis River Hatchery on-station releases: This program is released on a volitional basis over a 6-week period beginning on or after April 1; approximately 80% of the stock volitionally migrate during that time period. The remaining 20% are forced out prior to May 20 (Section 10.4 in WDFW and PacifiCorp, 2015a). The spring Chinook and winter-late steelhead HGMPs contain additional details about timing, duration, and location of releases.

1.1.2 Lewis River Type-S Coho Salmon – April 2015

- Same as Type-N, (WDFW and PacifiCorp, 2015d).

1.1.3 Lewis River Winter-Late (Endemic) Steelhead (Integrated) – April 2015

- Program steelhead are released fully-smolted to foster rapid outmigration from the basin and to minimize predation and residualism risks (WDFW and PacifiCorp, 2015b).
- This HGMP cites other studies showing that forced releases meeting size, time, and coefficient of variation parameters have resulted in similar smolt-to-adult return rates compared to volitional releases (Wagner, 1968, Everson and Ewing, 1992 as cited in WDFW and PacifiCorp, 2015b).
- Actively migrating steelhead are released in a different location than forced release fish. Fish that actively migrate will be trucked to the Merwin Boat Ramp for release (river kilometer 30.8). Once the volitional release window has ended, remaining fish will be hauled to the at the Pekins Ferry site and released (WDFW and PacifiCorp, 2015b).

1.1.4 Lewis River Winter (Segregated) and Summer Steelhead (Segregated) – April 2015

- The HGMP for the segregated winter Steelhead program is similar to that for the integrated program HGMP; however, does not contain the same details about release locations and timing. The same details about volitional releases are given in the summer steelhead (segregated) HGMP (WDFW and PacifiCorp, 2015e).

1.1.5 Lewis River Spring Chinook Salmon (Segregated and Integrated) – April 2015

- Lewis River program juvenile Chinook salmon are released fully-smolted to foster rapid outmigration from the basin and to minimize negative ecological interactions (WDFW and PacifiCorp, 2015c).
- Fish from this program are released on a volitional basis over a 6-week period, with approximately 80% of the stock volitionally migrating during that period. The remaining

20% are forced out prior to March 20 and November 20 for the earlier fall release (Section 9.2.8 in WDFW and PacifiCorp, 2015c).

Steelhead Hatchery and Genetic Management Plans for Nearby Systems

1.1.6 Kalama River Wild Summer Steelhead (Integrated)

- This program implements volitional release starting May 1, followed by a forced-release. All fish are forced out by May 15 (WDFW, 2017).

1.1.7 Hood River Production Program: Winter Steelhead (Integrated)

- Smolts are released from acclimation ponds over a 7-day period.
- In some past years, non-emigrating winter steelhead were trucked near the mouth of the Hood River to reduce interactions with native populations in Hood River (ODFW and Confederated Tribes of Warm Springs Reservation of Oregon, 2017).
- Sampling of non-migrants has occurred, but in low numbers that did not reveal statistically significant differences from the total population.
- Per Gerstenberger (2017), in past years, many PIT-tagged fish that did volitionally emigrate from acclimation ponds were observed a year later in the Hood River system, indicating that volitional release was not synonymous with full smolting and low residual rates. In recent years, delaying releases until after May 1 has allowed for growth to an optimal size for full-smolting, has increased the number of fish that leave acclimation sites volitionally, and reduced the number of residuals observed in the Hood River system. The emigration period now coincides more closely with the wild-type emigration for the native stock. The program no longer trucks non-migratory fish to the mouth of the river and now forces the small number of non-migratory smolts from acclimation sites into the river after a 3-day volitional release period (Gerstenberger, 2017). A study of physiological indicators of smolting and precocious maturation found that that residualism and precocity rates were very low (estimated 3 to 4% residualism rate and 1 to 12% rate of maturation after 1 year at sea; Larsen et al., 2017).

1.1.8 Clackamas River Winter Steelhead

- Oregon Department of Fish and Wildlife "will investigate the option of 'holding back' juvenile steelhead that do not migrate during the volitional release period after necessary facility improvements are completed as part of the intake upgrade project at Clackamas Hatchery. Under this option, all fish remaining after the volitional release period would be transferred to trout fisheries in standing waterbodies after reaching legal size … This option will also be investigated at Eagle Creek NFH *[National Fish Hatchery]*, but we are unsure of how non-emigrants can be removed from the ponds" (ODFW 2017)
- Smolts are force released from Clackamas Hatchery into the Clackamas River, Foster Acclimation Pond into Foster Creek, and Eagle Creek NFH into Eagle Creek (after 24-hour volitional release period).

1.1.9 Cowlitz River Hatchery Production (all species)

Per Hoffnagle and Shoblom (2017), program details were as follows:

- The Cowlitz River programs use forced releases after volitional release to the river for all species.
- There are concerns that steelhead may be residualizing, but no study is planned.
- Volitional release periods are based on logistical concerns more than physiological state of fish.

Literature Review – Steelhead

Willamette Basin – Tinus and Friesen (2010) summarized current knowledge, data needs, and recommendations for summer and winter steelhead in the upper Willamette Basin. They conclude that among the few studies that have examined performance indicators between volitional and forced-release steelhead, no significant differences among release strategies have been identified. The 2008 NMFS Biological Opinion cited in this report includes reasonable and prudent alternative 6.1.6 stating that steelhead smolts should be volitionally released over an extended period of time (e.g., 2 to 4 weeks) with any non-migrants being removed and not released into free-flowing waters below (hydropower) projects (NMFS, 2008; Tinus and Friesen, 2010). NOTE: It is not confirmed whether upper Willamette River facilities are actually removing non-migrant fish.

Methow Basin – Snow et al. (2013) compared volitionally released versus forced-release juvenile hatchery summer steelhead for survival, smolt-to-adult return, and contribution to stream-resident populations of juvenile hatchery summer steelhead in the Twisp River for 3 brood years (2002 to 2004). Volitionally released steelhead had higher survival, greater smolt-to-adult return rates, and less residualism. They estimated that 82% of stream-resident hatchery juvenile summer steelhead originated from forced-release fish. Results suggest managers should employ volitional release strategies and not release non-migratory juveniles into waters inhabited by anadromous fishes. This reduces negative ecological interactions at little cost to adult returns.

Also in the Methow Basin, Gale et al. (2009) compared volitional release, forced-release, and volitional non-migrant steelhead from Winthrop NFH for the following metrics: Gill Na+, K+-ATPase (NKA) activity, body size, condition factor (K), travel time, and apparent survival. There was little evidence for a survival-, size-, or physiology-related advantage of volitionally released fish over forced-released fish.

Tatara et al. (2017) explored the differences in out-migration and travel rates between S1 and S2 smolts released over 5 years from Winthrop NFH. Nonvolitional migrants had lower survival rates than volitional migrant steelhead in both S1s and S2s. The S2 rearing cycle produced larger smolts with more uniform size distribution, resulting in higher survival and faster travel times during out-migration in most years. Body size at release explained most of the differences in survival within and between the rearing groups. Volitional migrants from both treatments were larger and had higher survival than steelhead that were forced from the hatchery after the volitional release period ended.

Tucannon River – Viola and Schuck (2011) tested a release strategy designed to reduce the number of hatchery-reared steelhead failing to migrate out of the Tucannon River and described the physical characteristics of those failing to migrate. After volitional emigration ceased, fish remaining were held in an acclimation pond. They had a male:female ratio of 4:1, and mostly were a combination of transitional, parr, and precocious male stages. These fish were harvested by sport anglers.

Hood River – In a study of physiological indicators of smolting and precocious maturation in Hood River steelhead, Larsen et al. (2017) found that for steelhead, size was a strong driver of successful smolting, with an optimal size around 200 millimeters. Fish that tended to exhibit a parr-like phenotype characteristic of residuals were smaller than fully-smolted fish, and male fish that showed signs of precocity tended to be larger than immature smolts.

Literature Review – Spring Chinook

Wenatchee Basin – Johnson et al. (2015) compared the adult survival of hatchery-reared spring Chinook released volitionally versus forcibly from a facility in the upper Wenatchee River for brood years 2003 to 2005. Smolt-to-adult survival rates were higher for forced releases. They recommend accounting for variability in survival within the migration corridor (i.e., flow in the Federal Columbia River Power System at time of release might be a bigger factor in survival than whether fish were volitionally or forcibly released). Night releases also increased survival.

Suggestions

- Consider detaining non-migrating steelhead; they may be more likely contribute to stream-resident populations than improve adult returns (Snow et al., 2013).
- Monitor indices of smolt development (e.g., size distribution and smolt index) to ensure fish are released at the peak of smolt development. Compare indices of smolt development between non-migratory fish and the total population.
- Enumerate non-migrating fish.
- For steelhead, release fish of an optimal size that reduces the number of fish exhibiting either a small, parr-like residual phenotype or the large, precocious male phenotype.
- Consider matching release timing to the emigration timing of natural origin stocks (though it could increase competition with natural origin fish).
- Consider night releases for spring Chinook if using forced release strategy (Johnson et al., 2015).
- Clarify language about volitional release in plans and reports. If a forced release follows a volitional period, describe both strategies.
- Monitor smolt emigration rate and duration from the Lewis River.
- Coordinate with other programs about latest science regarding volitional releases and National Oceanic and Atmospheric Administration recommendations.

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Strategy I: Smolt-to-Adult Return Rate Estimation

Erik to prepare scope of work based on discussions with WDFW (Kale) – Placeholder until fall of 2022

Strategy J: Sampling and Data Collection Checklist

ADULT SAMPLING

Late Winter Steelhead	MFCF (PacifiCorp) HOR	MFCF (PacifiCorp) NOR	Hatchery (WDFW) HOR	Hatchery (WDFW) NOR
Genetic Tissues	No Sample in 2022		All broodstock sampled	All Broodstock sampled
Fork Length	(PIT -): 10 BWT per week (PIT +): Sample All	(PIT -): 10 per week (PIT +): Sample All	All broodstock sampled	All Broodstock sampled
Scales	(PIT -) 10 BWT per week (PIT +) none	(PIT -) 10 per week (PIT +) none	All broodstock sampled	All Broodstock sampled
Tagging	All maiden captures are PIT tagged	All maiden captures are PIT tagged	Floy	Floy
Fecundity	NA	NA	Individual (eggs/female)	Individual (eggs/female)
Chinook and Coho Salmon				
Genetic Tissues	No Sample in 2022		Chinook: none Coho: up to 200 samples (Kale Protocol)[3]	
Fork Length	(PIT -): up to 10 per week (PIT +): sample all	None[1]	(CWT +): sample all (CWT -): none	None
Scales	(PIT -): up to 10 per week (PIT +): none	None[1]	(CWT +): sample all (CWT -): none	None
Fecundity (spring Chinook, early coho)	NA	NA	Average Fecundity by spawn date (batch)	NA
Fecundity (late coho)	NA	NA	Average Fecundity by spawn date (batch)[2]	

*** included with all captures: 1) capture date, 2) sex (M,F,J), 3) Origin (HOR, NOR) and 4) Mark Status (PIT, CWT, AD, BWT)**

NOTES:

[1] Representative HOR samples to be obtained by hatchery through CWT collections and during spawning

[2] Fecundity is averaged among HOR and NOR females (i.e., no HOR vs. NOR fecundity)

[3] Three year sampling effort for coho to establish baseline up to 200 samples split amont early and lates, sex and NOR/HOR (source is email from Kale Bentley to Scott Peterson)

JUVENILE SAMPLING

SUMMARY IN DEVELOPMENT

APPENDIX A: Aquatic Technical Subgroup Work Plan for 2023 (living document)

Program Component	Deliverable	Objective	Updates
Hatchery M&E	Scale collection and analysis (spring Chinook and stlhd)	H&S 7.0	PacifiCorp provided scale and genetic samples to WDFW. Spr Chin and wint stlhd were included; samples are back from Olympia. Josua can share with group before Feb mtg.
Reporting	TDG results and analysis - Submit draft report to ATS	H&S 3.0	Sean to provide results for Erik to include in report; waiting on histology from WADDL
Adult Abundance	Complete strategy review: coho abundance estimates (fall monitoring)	H&S 5.0	Need to include strategy for spatial distribution of spawning Chinook and coho given the inabilitiy/inaccuracies to differentiate coho from Chinook redds.
Adult Abundance	Complete strategy review: Late winter steelhead abundance estimates (spring monitoring)	H&S 5.0	(see also pHOS topic)
HS Plan/AMEP	Develop alternative pHOS model recommendations for late winter steelhead (evaluate alongside abundance estimates in lower river)	H&S 8.0	Erik/Kale/Larissa to discuss offline and potentially bring to ATS for discussion in March.
Hatchery M&E	Consider multivariate analysis of components affecting chronic stress and BKD	H&S 3.0	data set to be completed by year-end; Sean and Luke meeting to consolidate data; Sean and Larissa to coordinate on datasets
ACC	Draft ACC Decision Template: Licensee monitoring obligations as defined in Lewis River Settlment Agreement and FERC licenses	ACC	
HGMP	Submit draft HGMPs to PacifiCorp for Review (Priority: spring Chinook and Coho)	H&S 1.0	PNI recomendations will be integrated as part of HGMP/NOAA.
HGMP	Finalize Coho Transition Plan	H&S 1.0	
Genetics Monitoring	Finalize 2023 Genetics Monitoring Strategy	H&S 7.0	
Hatchery M&E	Develop scope for hatchery SAR	H&S 3.0	
HGMP	Distribute draft Chinook and Steelhead transition plans to ATS	H&S 1.0	
Reporting	Submit Draft Annual Report for H&S and AMEP monitoring in 2022	H&S 6.0	Included Yale HPP Analysis reporting (SA 7.4)
Reporting	Conduct kickoff to review the summary table of metrics and outcomes with the ACC	SA XX	
Juvenile Abundance	Review juvenile in-river trapping results, recommendations for 2024		Decisions needed early summer considering permitting constraints
Hatchery M&E	Final evaluation plan of shade covers at Speelyai Hatchery	H&S 3.0	
HS Plan/AMEP	Formalize data collection, management and reporting strategy (Strategy J)	H&S 3.0	Recommendation is to develop Strategy J to incorporate these protocols
HPP	Draft Yale/Merwin HPP for ATS review	SA 7.4	
HS Plan/AMEP	Coordinate and develop IPMs (AMEP)	AMEP	Chris managing discussions with USGS, will provide updates to ATS. WDFW requests a collaborative meeting to inform the IPMs once process has started.
HPP	Conduct Yale tributary surveys for HPP	none	
HS Plan/AMEP	Submit Handling and Transport plans to ATS (future fish passage facilities)	AMEP	
HS Plan/AMEP	Update AMEP with new tasks associated with future fish passage requirements	SA 9	

APPENDIX B: Metrics Matrix for Lewis River Programs

Metrics Matrix for Lewis River Programs

CATEGORY	No.	OBJECTIVE	KEY QUESTIONS	MONITORING INDICATOR OR METRIC	TARGETS	AOP STRATEGY
Administrative	**1.0**	NOAA acceptance of a Hatchery and Genetic Management Plan (HGMP) for each hatchery program on the North Fork Lewis River		NOAA accepts final HGMP for each hatchery program	NA	
Administrative	1.1	Receive Biological Opinion for all submitted HGMPs		Biological Opinion is issued for each hatchery program	NA	
Administrative	**2.0**	Finalize a Hatchery and Supplementation Plan every 5 years		Final plan is submitted and accepted by the FERC	December 31	
Administrative	2.1	Finalize an annual operating plan (AOP)		Final plan is approved by the ATS and ACC	December 31	
Administrative	2.2	Finalize and Annual Operations Report (AOR)		Final report is submitted and accepted by the FERC	Draft distributed for ACC review April 1, presented to ACC in May meeting; final submitted to FERC by June 30	
Administrative	2.3	Finalize an annual hatchery operations report		Final report is submitted to the utilities for incorporation	March 1	
Hatchery Monitoring	**3.0**	Determine whether hatchery production protocols incorporate best available management practices to suport program targets and goals.	3A. Do hatchery broodstock collection protocols support program goals?	3A.1.Trap entry timing	Broodstock collection follows collection curves over the course of the trapping period.	Broodstock collection goals and timing in Sections A2, B2, C2 of the AOP.
Hatchery Monitoring				3A.2.Broodstock retention rate	Total broodstock target numbers are met. Broodstock collection rates that are consistent with planned broodstock collection curves for each species. For integrated programs, match collection timing to average NOR return timing.	
Hatchery Monitoring			3B. Do spawning, rearing and release strategies support program goals?	3B.1. Integration Rates (pNOB; Integrated programs only)	Steelhead: 30% fixed mining rate. Coho: 30% integration rate.	Sections A2.1, B2.1, C2.1 of the AOP

Metrics Matrix for Lewis River Programs

CATEGORY	No.	FIELD METHODS	ANALYTICAL METHODS	FREQUENCY AND DURATION	COMMENTS
Administrative	**1.0**			NA	
Administrative	1.1			NA	
Administrative	**2.0**			Every 5 years	
Administrative	2.1			Annual	
Administrative	2.2			Annual	
Administrative	2.3			Annual	Hatchery report may need to include additional data compared to previous years to meet metric reporting needs.
Hatchery Monitoring	**3.0**	See broodstock collection goals and timing for individual species in Sections A2, B2 and C2 of the AOP.	Sum of adults trapped by day or week. Identify first, last, and peak run dates from distribution of daily trap counts.	Data are collected annually when traps are sorted; daily at Merwin Trap, weekly at Lewis River Hatchery Ladder. Broodstock collection periods: Steelhead: late January to end of May Spring Chinook: April 1 to late August Coho: early September through December	ATS to review run timing and broodstock collection curves during annual reporting
Hatchery Monitoring			Broodstock retention rate reported by week as the number of fish held for broodstock out of the total number of fish trapped and sorted (daily/weekly counts, cumulative total, and % cumulative total of annual total).Make comparisons of annual HOR and NOR return timing to 5-year average return time, and to planned and observed broodstock collection curves.		
Hatchery Monitoring		Outlined in Sections A2.1, B2.1, C2.1 of this AOP as part of the broodstock collection process.	NA	In general, data on these metrics are collected as part of typical hatchery program operations and will be reported on in the Annual Operating Report, and trends will be evaluated as part of the Adaptive Management process described in Section E. For spring Chinook salmon, after 3 years of implementation of the Spring Chinook Rearing and Release Plan (Strategy A), in-hatchery survival rates, size-at-release, condition factor at release, fish health (frequency or rates of disease), and physiological status at the time of release will be compared between treatment groups.	

Metrics Matrix for Lewis River Programs

CATEGORY	No.	OBJECTIVE	KEY QUESTIONS	MONITORING INDICATOR OR METRIC	TARGETS	AOP STRATEGY
Hatchery Monitoring	**cont.**	cont.	cont.	3B.2. Spawning matrices and timing	Steelhead: Depends on spawners available on a given day; typically 2x2. Spring Chinook and coho: pairwise (1x1) mating cross with a backup male .	Sections A2.9, B2.9, C2.9 of the AOP
Hatchery Monitoring				3B.3. Broodstock Fecundity	NA	
Hatchery Monitoring				3B.4. Feeding rations and delivery methods	Not Applicable in 2022	
Hatchery Monitoring				3B.5. Avian predation rate	Not Applicable in 2022	
Hatchery Monitoring				3B.6. Volitional releases	Steelhead: May 1; 6-week volitional period. Spring Chinook: February 1, October 15, or June 1; 2-week volitional period. Coho: April for 2 to 6-week volitional period.	
Hatchery Monitoring				3B.7. Water Quality	NA	Strategy K
Hatchery Monitoring			3C. Are adult collection, handling and disposition protocols consistent with HSRG recommendations?	3C.1. Size and age of returning HOR and NOR adults	Use of size and age at maturity as indicator of phenotypic diversity among HOR, NOR and integrated programs. See Objective 7.	Strategy A
Hatchery Monitoring				3C.2. Disposition of adult trap captures assigned to surplus	Meet goals for proportion and timing of adult disposition that are consistent with the H&S Plan and AOP.	Sections A2.7, B2.7, and C2.7 of the AOP
Hatchery Monitoring			3D. What are the estimated smolt-to-adult returns (SAR's) for each hatchery stock or rearing treatment group?	3D. Smolt-to-adult Return ratio (SAR) of all hatchery release groups	Adult returns are adequate to meet adult ocean recruit targets given in section 8.3 of the Settlement Agreement.	Strategy E (placeholder as of 11/22/2022)

Metrics Matrix for Lewis River Programs

CATEGORY	No.	FIELD METHODS	ANALYTICAL METHODS	FREQUENCY AND DURATION	COMMENTS
Hatchery Monitoring	**cont.**	Outlined in Sections A2.9, B2.9, C2.9 of this AOP as part of the spawning protocols. The number, timing and composition of spawners used, and type of spawning matrix used for each stock will be recorded (e.g., pairwise, pairwise with backup male, factorial, etc.).	NA	cont.	
Hatchery Monitoring		Late winter steelhead: Record the estimated number of eggs per female. Early coho and spring Chinook: Average fecundity will be estimated from a subsample of females spawned.	For integrated programs (late winter steelhead and late coho), fecundity will be compared between HOR and NOR. Fecundity and spawn timing data are also used to examine risk to phenotypic diversity described in Objective 7.		
Hatchery Monitoring		Not Applicable in 2022	Not Applicable in 2022		
Hatchery Monitoring		Not Applicable in 2022	Not Applicable in 2022		
Hatchery Monitoring		The start date and time of volitional release (i.e., when screens are pulled) and the start and end date and time of the forced release period will be recorded for each species and pond.			
Hatchery Monitoring		2022 TDG Evaluation and Mitigation Plan (Strategy K)	See TDG Evaluation and Mitigation Plan (Strategy K)	The evaluation will occur during the summer of 2022 and capture periods of higher water temperatures, specifically between July and October. Water quality measurements will be recorded continuously using a 1-hour interval.	
Hatchery Monitoring		Subsample of fork lengths and scales from trap returns and in-stream carcass surveys for coho and Chinook. Refer to Strategy A for data collection on spawning grounds.	Size and age data should be visualized and compared between HOR and NOR with distribution plots and point estimates with 95% confidence intervals.	Sampling occurs throughout the duration of the run. Daily (Merwin Trap); weekly (Lewis hatchery ladder and stream surveys).	
Hatchery Monitoring		Refer to Sections A2.7, B2.7, and C2.7.	Disposition of each captured adult will be recorded throughout the complete return period for each of the transport species.	Daily at Merwin Trap Weekly at Lewis hatchery ladder	
Hatchery Monitoring		Collection of CWTs from fish encountered in 1) Lewis River traps 2) Lewis River subbasin spawning grounds, 3) strays to other basins and 4) harvest by stock	CWT data obtained from RMIS based on release codes. Returns include recaptures from: • Adult harvest in all fisheries • Adult spawners • Adult traps Releases are estimated number of CWT smolts released by release group, corrected for estimated tag loss and post-tagging in-hatchery mortality.	Annual SAR estimate for each brood year and release group	

CATEGORY	No.	OBJECTIVE	KEY QUESTIONS	MONITORING INDICATOR OR METRIC	TARGETS	AOP STRATEGY
Hatchery Monitoring	**cont.**	cont.	3E. Is the fish health monitoring and disease prevention strategy effective at reducing infections and limiting mortalities?	3E.1. Infection rates by species and life stage	Ensure the health and productivity of H&S Plan fish	Strategy D
Hatchery Monitoring				3E.2. Mortality rates by species and life-stage	Ensure mortality rates are not adversely affecting production targets	Strategy D
Hatchery Monitoring			3F. Do hatcheries incorporate new scientific advances to improve fish culture effectiveness and efficiency?	3F. Periodic review of hatchery operations relative to current literature	Implementation of hatchery activities that are based on best available science on maintaining fish health, sustaining harvest, and minimizing genetic or ecological risks.	

Metrics Matrix for Lewis River Programs

CATEGORY	No.	FIELD METHODS	ANALYTICAL METHODS	FREQUENCY AND DURATION	COMMENTS
Hatchery Monitoring	**cont.**	60 adult females of each species are inspected and sampled during spawning All spring Chinook females whose eggs are allocated for February release will be tested for BKD prevalence Rearing juveniles are monitored and examined routinely	Fish health monitoring results are reported and maintained by WDFW and pathogen histories are available at any time upon request. The subsample of juvenile spring Chinook evaluated for BKD at transfer and release will be analyzed to report prevalence (% positive), DNA load, prevalence of severe infections, and prevalence of gross pathology.	Baseline monitoring occurs throughout broodstock collection, spawning, and incubation as described in Strategy D. Directed monitoring occurs as needed.	
Hatchery Monitoring		If needed, medication will be provided by the veterinarian of record	Results are generally reported as presence/absence, mortality range (normal, increased, epizootic) and % loss (mortality) per day for a given rearing unit. In-hatchery survival reported as survival (S = total count – mortalities) from egg to release for each species and release group.	Special monitoring of juvenile spring Chinook for GBT will occur with an initial baseline examination in June, followed by weekly examinations in August Special monitoring of juvenile spring Chinook for BKD will occur at the times of transfer and release.	
Hatchery Monitoring		NA	The AOR and current hatchery methods will be reviewed and compared to published literature and methods from other hatcheries. The ATS will identify known areas of concern for hatchery operations efficiency or topics of recent advances in hatchery science. Potential outcomes include identification of a data gap, next steps toward making changes in implementation, or recommending an immediate implementation change if evidence supports it.	Hatchery operations will be reviewed every three years. The discussion of the approach to the review will occur in June of the review year, and recommendations will be completed by October of the review year for incorporation into the following AOP for the following year.	

Metrics Matrix for Lewis River Programs

CATEGORY	No.	OBJECTIVE	KEY QUESTIONS	MONITORING INDICATOR OR METRIC	TARGETS	AOP STRATEGY
Hatchery Monitoring	**4.0**	Adopt strategies that limit potential post-release ecological interactions between hatchery and NOR listed species	4A. Do current hatchery releases result in spatial and temporal overlap between HOR and NOR juveniles?	4A.1. Release locations of hatchery smolts relative in-river spawning locations.	Mitigate potential spatial overlap between hatchery-released juveniles and NOR stocks	Strategy B
Hatchery Monitoring				4A.2. Release timing of hatchery reared smolts relative to presence of NOR	Mitigate potential temporal overlap between hatchery-released juveniles and NOR stocks	Strategy B
Hatchery Monitoring			4B. Does the migration rate of HOR juveniles result in overlap with NOR juveniles or spawning adults?	4B. Average migration rate and range of migration rates of hatchery released smolts	Rapid outmigration to minimize the period of time that hatchery-origin juveniles may encounter natural-origin fish.	Sections A3.7, B3.7, and C3.7 for hatchery release timing. Refer to Strategy C for screw trapping methods
Hatchery Monitoring			4C. Are the number of hatchery-released juveniles equal to or less than production targets?	4C. Number of total smolts released by species and period	≤ 105% of target release	Hatchery production targets contained in the H&S Plan and Settlement Agreement
Hatchery Monitoring			4D. Are the sizes (length and weight) of released hatchery juveniles equal to or less than program targets?	4D. Mean and coefficient of variation (CV) in fork length and weight of smolts released by species and release group.	Steelhead: > 180 and < 220 mm. Spring Chinook: 8-12 fpp for October or February releases, 80 fpp for June release (see Strategy E). Coho: 16 fpp.	
Hatchery Monitoring			4E. What is the precocity rate for hatchery juveniles by release group prior to scheduled releases?	4E. Precocity Rate	Minimize precocity rates to reduce residualism and interactions between mature juveniles released at the same time as natural spawning in the river. Precocity rate in wild spring Chinook likely to be less than 5% of males.	Strategy E and F
Abundance Monitoring	**5.0**	Estimate spawner abundance of late winter steelhead, Coho, chum and Chinook downstream of Merwin Dam	5A. Are estimates of spawner abundance unbiased and meeting precision targets?	5A. Mark-recapture modeling assumptions are evaluated to determine if they are met	Generate an unbiased estimate of abundance and composition downstream of Merwin Dam with a CV of 15% or less, on average	Strategy A
Abundance Monitoring			5B. Are annual estimates of natural origin spawner abundance increasing, decreasing or stable?	5B. Total spawner abundance estimates by species, sex and origin, and age.	Annual trends are stable or increasing.	Strategy A
Abundance Monitoring	5.1	Determine the spatial and temporal distribution of spawning late winter steelhead, coho, chum and Chinook downstream of Merwin Dam	5C. Are annual trends in temporal and spatial spawning distribution increasing, decreasing or stable?	5C. Estimate of redds, carcasses or live spawners by reach and time period	Proportion of redds per reach is stable or increasing	Strategy A, Strategy B

Metrics Matrix for Lewis River Programs

CATEGORY	No.	FIELD METHODS	ANALYTICAL METHODS	FREQUENCY AND DURATION	COMMENTS
Hatchery Monitoring	**4.0**	Release location(s) of each hatchery pond; multiple locations to include relative portion of total release (e.g., forced release group transferred to Pekins Ferry) Spawner distribution monitored by redd and carcass (spawner) surveys, as described in Strategy B.	Compare hatchery release locations to distribution of natural-origin fish inferred from spawning surveys and screw trap captures.	Annually	
Hatchery Monitoring			Compare hatchery juvenile release dates and encounters of hatchery origin fish with timing of natural-origin fish in the datasets for adult trap entry, spawner surveys, and screw trap encounters.	Annually	
Hatchery Monitoring		Refer to Sections A3.7, B3.7, and C3.7 for hatchery release timing. Refer to Strategy C for screw trapping methods.	Derive minimum, maximum, and mean or median migration rates from the difference between release date and screw trap capture dates.	Average migration rates will be reported annually for species (or release groups) in context of trends across years, as described in Section E of this document, Adaptive Management.	Migration points need definition (e.g., hatchery to screw trap, hatchery to tidal influence); migration target rate needs definition
Hatchery Monitoring		Rearing ponds stocked with number determined volumetrically. Mortalities subtracted over rearing period.	Compare number released to target.	Report annually for each release group.	
Hatchery Monitoring		Batch weights collected monthly. Length measured from a representative subsample (e.g. 100 fish per release group) at the time of release.	Calculate fish per pound (fpp) from weights to compare to targets. Calculate mean and CV of fork length, weight.	Monthly batch weights. Average fork length at the time of release.	Note that steelhead size rates are set to limit residualism or precocity.
Hatchery Monitoring		Non-lethal visual screening for October spring Chinook release group. Lethal 11kt or GSI sampling for February spring Chinook release groups. Refer to Strategy F for details.	Calculate precocity rate as number of precocious males out of total number of males. Refer to Strategy F for details.	Non-lethal visual screening for October spring Chinook release group carried out annually as part of routine morphology monitoring at the time of release. Lethal 11kt or GSI sampling for February spring Chinook release group carried out periodically.	
Abundance Monitoring	**5.0**	Chinook: Carcass mark-recapture. Steelhead: redd survey. Coho: Carcass mark-recapture in mainstem, GRTS and Bayesian multivariate state-space model in tributaries. Refer to Strategy A.	Annual evaluation of assumptions made for modeled estimates of abundance. Refer to Strategy A. Chinook: Open-population Jolly-Seber "super population" analysis to generate abundance estimates by stock, origin, sex and age. Steelhead: redd counts multiplied by fish per redd; AUC if necessary. Coho: Jolly-Seber mark recapture model for mainstem, and Bayesian multivariate state-space model for tributaries, to generate abundance estimates by origin.	Surveys and analyses completed annually. Results reported annually by species in context of trends across years, as described in Section E of this document, Adaptive Management.	steelhead redd surveys are inherently biased
Abundance Monitoring		Carcass and redd surveys. Refer to Strategy A.	Chinook: Open-population Jolly-Seber "super population" analysis. Coho: Jolly-Seber mark recapture model for mainstem, and Bayesian multivariate state-space model for tributaries. Steelhead: redd surveys		We are not currently using redd surveys due to redd superimposition between Chinook and Coho. That is, we are not meeting this key question
Abundance Monitoring	5.1	Spawner surveys described in Strategy A with spatial and temporal data recorded using GPS as described in Strategy B.	Median spawner number per reach and median spawn date per week to be calculated.		

Metrics Matrix for Lewis River Programs

CATEGORY	No.	OBJECTIVE	KEY QUESTIONS	MONITORING INDICATOR OR METRIC	TARGETS	AOP STRATEGY
Abundance Monitoring	**6.0**	Estimate juvenile outmigrant abundance for late winter steelhead, coho, and Chinook downstream of Merwin Dam	6A. Are estimates of NOR juvenile outmigrant abundance unbiased and meeting precision targets?	6A. Precision and accuracy of abundance estimates.	VSP precision and accuracy targets are met: CV < 15% (salmon) and < 30% (steelhead)	Strategy C
Abundance Monitoring			6B. Is the abundance of NOR juvenile outmigrants by species and outmigration year increasing, decreasing, or stable?	6B.1. Trend in total NOR outmigrants by species and cohort.	Annual trends are stable or increasing.	Strategy C
Abundance Monitoring			6C. What are the morphological characteristics of outmigrating NOR juveniles relative to their conspecific HOR juveniles?	6C. Comparison of fish size, life-stage and age-class between NOR and HOR juveniles.	No relevant target for natural-origin outmigrants; hatchery-origin outmigrant size based on optimizing post-release performance.	Strategy C Sections A 3.7, B 3.7, and C 3.7 of the AOP
Risk Assessment	**7.0**	Monitor the extent of genetic risks associated with integrated and segregated hatchery programs on naturally spawning listed populations in the North Fork Lewis River	7A. Have the Lewis River hatchery programs impacted the among-population diversity of naturally spawning populations?	7A. Pariwise genetic distance (Fst), combined with dendograms, multi-variate clustering analyses.	See Strategy G.	Strategy G
Risk Assessment			7B. Have the Lewis River hatchery programs impacted the within-population diversity of naturally spawning populations?	7B.1. Effect population size (Ne) 7B.2. Effective number of breeders (Nb) 7B.3. Inbreeding coefficient (FIS) 7B.4. Average heterozygosity 7B.6. Allele frequencies 7B.7. Linkage Disequilibrium (LD)	See Strategy G.	Strategy G
Risk Assessment			7C. Have the Lewis River hatchery programs increased the risk of domestication for naturally spawning populations?	7C.1. Proportion hatchery-origin spawners (pHOS)	Specific HSRG-recommended targets apply, see Objective 8	See Objective 8
Risk Assessment				7C.2. Proportion natural-origin brood (pNOB)	A component of PNI target. Does not apply to segregated programs.	See Objective 8
Risk Assessment				7C.3. Proportion natural influence (PNI)	Specific HSRG-recommended targets apply, see Objective 8	See Objective 8
Risk Assessment				7C.4 Proportion Effective Hatchery Contribution (PEHC)	No specific targets recommended. Program should avoid increase in PEHC over time. Not relevant for integrated programs.	See Objective 8

Metrics Matrix for Lewis River Programs

CATEGORY	No.	FIELD METHODS	ANALYTICAL METHODS	FREQUENCY AND DURATION	COMMENTS
Abundance Monitoring	**6.0**	Rotary screw traps will be used to capture a portion of the total number of juveniles passing the trap location. Mark-recapture of those fish will be used to test trap efficiency. Refer to Strategy C	Quantify whether assumptions of the estimator are met. See Strategy C for details.	Annual juvenile outmigrant abundance and morphology sampling. Traps checked every morning, 7 days per week, from March 1 through June 30. Trapping dates may be adjusted depending on river flows. Additional daily trap checks may be warranted during peak migration.	
Abundance Monitoring		Screw trapping to capture outmigrants. Refer to Strategy C.	Derive estimates of abundance based on trap efficiency. Refer to Strategy C.		
Abundance Monitoring		Refer to Sections A 3.7, B 3.7 and C 3.7 of this document for data collection at the time of release. Up to 10 fry per day and 50 subyearling/ transitional/ parr per day per category to be sampled for fork length during screw trapping. Refer to Strategy C.	Average size to be calculated from representative subsample at hatchery release and in screw trapping. The component of those subsampled made up by different life-stages and age classes should also be reported.	Annual hatchery release and juvenile outmigrant abundance and morphology sampling. Traps checked every morning, 7 days per week, from March 1 through June 30. Trapping dates may be adjusted depending on river flows. Additional daily trap checks may be warranted during peak migration.	Discuss practicality of this key question. HOR and NOR differ in morphology and no changes are planned for HOR size targets.
Risk Assessment	**7.0**	See Strategy G.	See Strategy G.	Collect tissue samples annually. Analyses of genetic distance at least every third generation.	
Risk Assessment		See Strategy G.	See Strategy G.	See Strategy G.	
Risk Assessment		See Objective 8	See Objective 8	Calculated Annually	
Risk Assessment		See Objective 8	See Objective 8	Calculated Annually	
Risk Assessment		See Objective 8	See Objective 8	Calculated Annually	
Risk Assessment		See Objective 8	See Objective 8	Calculated Annually	

CATEGORY	No.	OBJECTIVE	KEY QUESTIONS	MONITORING INDICATOR OR METRIC	TARGETS	AOP STRATEGY
Risk Assessment	**cont.**	cont.	7D. Have the Lewis River hatchery programs impacted the phenotypic diversity of naturally spawning populations?	7D.1 Timing of adult return, spawning and juvenile outmigration.	See Key Questions 3A and 3B for adults. See Key Question 4B for juveniles.	
Risk Assessment				7D.2 Size and age of returning adults and juvenile outmigrants.	See Key Question 3C for adults. See Key Question 4B for juveniles.	
Risk Assessment	**8.0**	Determine the percent hatchery-origin spawners (pHOS), proportionate natural influence (PNI) and pNOB (for integrated programs)?	8A. What are the trends in pHOS, PNI, pNOB and PEHC and do they meet HSRG recommendations by program (when applicable)?	8A.1. Proportion hatchery-origin spawners (pHOS)	See Section D of the AOP for population-specific targets that meet HSRG guidance	
Risk Assessment				8A.2. Proportion natural-origin brood (pNOB)	A component of PNI target. Applies only to integrated programs. Late winter steelhead uses a pNOB target of 1 to achieve a PNI ≥ 0.50. Late coho follows a recommended integration rate (e.g., 30 percent) based on the designation for each stock or population (e.g., primary, contributing, or stabilizing.)	
Risk Assessment				8A.3.Proportion natural influence (PNI)	See Section D of the AOP for population-specific targets that meet HSRG guidance. This metric is influenced substantially by the pNOB. For example, if the broodstock incorporates 100 percent natural origin fish, PNI estimates cannot be less than 50 percent.	
Risk Assessment				8A.4.Proportion Effective Hatchery Contribution (PEHC)	No specific targets recommended. Program should avoid increase in PEHC over time. Not relevant for integrated programs.	Strategy G
Risk Assessment	**9.0**	Monitor the post-release behavior of hatchery smolts and their potential impacts on native and ESA-listed species present downstream of Merwin Dam.	9A. Contingent on meeting monitoring goals related to Objective 4, or as determined by the ATS			

Metrics Matrix for Lewis River Programs

CATEGORY	No.	FIELD METHODS	ANALYTICAL METHODS	FREQUENCY AND DURATION	COMMENTS
Risk Assessment	**cont.**	See Key Questions 3A and 3B for adults. See Key Question 4B for juveniles.	See Key Questions 3A and 3B for adults. See Key Question 4B for juveniles.	See Key Questions 3A and 3B for adults. See Key Question 4B for juveniles.	
Risk Assessment		See Key Question 3C for adults. See Key Question 4B for juveniles.	See Key Question 3C for adults. See Key Question 4B for juveniles.	See Key Question 3C for adults. See Key Question 4B for juveniles.	
Risk Assessment	**8.0**	For Chinook and Coho spawner origin is derived from spawner surveys. Refer to Strategy A. Methods for determining pHOS in steelhead are to be determined in coordination with the ATS.	pHOS = Number of HOS/(HOS + NOS)	Calculated annually	
Risk Assessment		For each integrated program (late winter steelhead and late coho), the origin and sex of each fish spawned will be recorded.	pNOB = NOB / (HOB + NOB). pNOB will be calculated within each spawning matrix, and for the total number of spawners.	Calculated annually	
Risk Assessment		NA	PNI = pNOB/(pNOB+pHOS)	Calculated annually	
Risk Assessment		NA	See Strategy G.	Calculated annually	
Risk Assessment	**9.0**				This objective provides means for direct monitoring of ecological interactions between HOR and NOR juveniles in the event that in-hatchery monitoring metrics described under Objective 4 are not achieved.