

SPRING CHINOOK SALMON SUPPLEMENTATION IN THE UPPER YAKIMA BASIN: YAKIMA/KLICKITAT FISHERIES PROJECT OVERVIEW

Annual Report 2015

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**Policy/Technical Involvement and Planning in the Yakima/Klickitat Fisheries Project;
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Yakima/Klickitat Fisheries Project's Monitoring and Evaluation; Project Number 1995-
063-25; Contract 65604, 69084**

This report is an overview of the work conducted on spring Chinook salmon as part of the Yakima/Klickitat Fisheries Project (YKFP), which is a joint project of the Yakama Nation and the Washington Department of Fish & Wildlife. The purpose of this document is to synthesize the available information that has already been provided in lengthy topical reports into an easy to read synthesis of the project. In this way, we hope that the scope and progress of the YKFP can be fully appreciated. The YKFP is still in the evaluation stage, and as such the data and findings presented in this report should be considered preliminary until further data is collected and analyses completed. We encourage the reader to consult the topical reports for detailed descriptions of particular topics. The latest topical reports are contained in the appendices and previous reports can be found on www.cbfish.org. There are also other components of the YKFP that address coho and fall Chinook salmon. Those components are not addressed in this report.

The YKFP is funded under BPA contracts to the Yakama Nation and the Washington Department of Fish and Wildlife. The following contracts provided the support to complete the work that is the basis for this report.

Policy/Technical Involvement and Planning in the Yakima/Klickitat Fisheries Project; Project Number 1995-064-25; Contracts 66257 and 69345 - WDFW

Yakima/Klickitat Fisheries Project's Monitoring and Evaluation; Project Number 1995-063-25; Contracts 65604 and 69084 – WDFW

Yakima/Klickitat Fisheries Project's Monitoring and Evaluation; Project Number 1995-063-25; Contract 56662 REL 68 and 85 – Yakama Nation

Yakima/Klickitat Fisheries Project's Management, Data, and Habitat; Project Number 1988-120-25; Contract 56662 REL 66 and 96 – Yakama Nation

Executive Summary

The Yakima/Klickitat Fisheries Project (YKFP) is on schedule to ascertain whether new artificial production techniques can be used to increase harvest and natural production of spring Chinook salmon while maintaining the long-term genetic fitness of the fish population being supplemented and keeping adverse genetic and ecological interactions with non-target species or stocks within acceptable limits. The Cle Elum Supplementation and Research Facility (CESRF) collected its first spring Chinook brood stock in 1997, released its first fish in 1999, and age-4 adults have been returning since 2001. In these initial years of CESRF operation, recruitment of hatchery origin fish has exceeded that of fish spawning in the natural environment, but early indications are that hatchery origin fish are not as successful at spawning in the natural environment as natural origin fish. However, no differences have been found in the reproductive success between first and second generation hatchery fish. Preliminary results indicate that significant differences have been detected between hatchery and natural origin fish in about half of the traits measured in our monitoring plan and that these differences can be attributed to both environmental and genetic causes. For example, we have detected differences in hatchery and natural origin fish after only one generation of hatchery exposure for the following variables measured on adults: age composition, size-at-age, sex ratio, spawn timing, fecundity, egg weight, adult morphology at spawning, and spawning success. Significant differences in juvenile traits have also been detected: food conversion efficiency, length-weight relationships, agonistic competitive behavior, predator avoidance, and incidence of precocious maturation. Most of the differences have been 10% or less.

Distribution of spawners has increased as a result of acclimation site location and salmon homing fidelity, although other factors may ultimately override homing to specific acclimation sites and result in substantial overlap with naturally produced spawners. Semi-natural rearing and predator avoidance training have not resulted in significant increases in survival of hatchery fish. Growth manipulations in the hatchery reduced the number of precocious males produced by the YKFP, however survival of both post-release juveniles and survival to adult return was significantly lower for the low-low growth treatment compared to the standard-growth treatment. Genetic impacts to non-target populations appear to be low because of the low stray rates of YKFP fish. Ecological impacts to valued non-target taxa were generally within containment objectives, or impacts that were outside of containment objectives were not attributed to supplementation activities. Fish and bird piscivores consume large numbers of salmonids in the Yakima Basin. Natural production of Chinook salmon in the upper Yakima Basin appears to be density dependent under current conditions and may constrain the benefits of supplementation. However, such constraints could be countered by YKFP habitat actions that have resulted in: the protection of over 1,800 acres of prime floodplain habitat, addition of wood into tributaries, reconnection and screening of over 50 miles of tributary habitat, substantial water savings through irrigation improvements, and restoration of over 100 acres of floodplain and side channels. Additional habitat improvements implemented by other entities, including the Conservation Districts, counties and private interests are also continuing in the basin. Harvest opportunities for tribal and non-tribal fishers have also been enhanced, but are variable among years. Harvest objectives for the upper Yakima stock and all Yakima basin stocks combined are currently meeting the quantitative objectives in both the Columbia and Yakima rivers. The YKFP is still in the evaluation stage and as such the data and findings presented in this report should be considered preliminary until further data is collected and analyses completed.

Nonetheless, the YKFP has produced significant findings, and developed methodologies that can be used to evaluate and improve supplementation. A summary table of topical area performance is presented below.

Table 1. Performance of the Yakima Fisheries Project relative to quantitative objectives reported in Pearsons et al. (2006).

Performance Measure	Goal	Performance	Comments
Natural Production of Target Species	Increase while maintaining the long-term fitness of the target population (see quantitative objectives; Pearsons et al. 2006)	Quantitative objectives for adults and smolts are being achieved. Differences in traits of hatchery and natural origin fish are a concern	<ul style="list-style-type: none"> - Too early to evaluate conclusively, but strategies to reduce genetic risk are being implemented. - Hatchery has increased the number and distribution of adult spawners on the spawning grounds. Quantitative management objectives for natural production of upper Yakima and basin total spring Chinook adults and smolts are being achieved. - Significant but small changes in many demographic and reproductive success traits indicate cause for concern. - Predation and competition may be limiting natural production objectives and may constrain the benefits of supplementation.
Harvest	Increase (see quantitative objectives; Pearsons et al. 2006)	Increased, and objectives are being met	<ul style="list-style-type: none"> - Tribal subsistence fisheries occurred on both hatchery and naturally produced fish in all years. Sport fisheries on hatchery fish have also occurred in the Yakima River in 11 of the last 15 years beginning in 2001. - Quantitative harvest objectives for the upper Yakima stock and all Yakima basin stocks combined are being met for the Columbia or Yakima Rivers
Genetics	Minimize genetic impacts to non-target	Achieved to date	Stray rates are very low

	taxa		
Ecology	Keep impacts to non-target taxa within containment objectives (see Pearsons et al. 2006)	Achieved for most taxa to date	Impacts for most species are within containment objectives or are currently not attributable to supplementation.
Habitat	Protect the most productive stream reaches and increase productivity/capacity of freshwater environment so that quantitative objectives can be achieved.	Progress	Habitat protection, restoration, and tributary passage efforts are ongoing, with incremental progress each year. - Habitat actions should enhance the benefits of supplementation, especially over the long-term.
Science	Disseminate important findings for use throughout the Yakima Basin, Columbia Basin, and world	Achieved to date	Numerous annual reports were submitted to BPA, all tasks were reported on at annual conferences, and manuscripts have been prepared and published.

Short Project Overview

Salmon and steelhead populations in the Yakima Basin and throughout the Columbia Basin are far below historic levels. For example, an average of 200,000 spring Chinook salmon returned to the Yakima Basin prior to 1800, but declined to an average of fewer than 3,500 fish annually from 1982-1999. Hatcheries have been used as the primary tool to mitigate for the losses of salmon in the Columbia Basin. However, naturally produced salmon have continued to decline despite large releases of hatchery fish. This decline in abundance has caused many Evolutionary Significant Units of salmon and steelhead to be listed for federal protection under the Endangered Species Act. Traditional hatchery operations have been successful at producing fish for harvest, but may actually harm naturally produced fish through ecological, genetic, facility, and harvest interactions.

The YKFP is designed to determine whether it is possible to change hatchery practices so that natural spawning populations of salmon receive biological benefits from a hatchery program. The project is also examining whether these same hatchery practices can be managed to limit deleterious impacts on non-enhanced fish populations. More specifically, the YKFP is testing whether “artificial propagation [can be used] to increase natural production while maintaining the long term fitness of the target population, and keeping the ecological and genetic impacts on non-target populations within specified biological limits” (RASP 1992). In addition, increasing harvest opportunities for tribal and non-tribal fishers is also part of the overall goal. In short, the YKFP is attempting to quantify the demographic, ecological, and genetic benefits and costs of supplementation. Quantitative objectives of the spring Chinook salmon component of the YKFP are presented in Pearsons et al. (2006).

In order to test whether supplementation works, in the Yakima Basin or elsewhere, at least four major questions must be answered:

- 1) Can integrated hatchery programs be used to increase long-term natural production?
- 2) Can integrated hatchery programs limit genetic impacts to non-target Chinook populations?
- 3) Can integrated hatchery programs limit ecological impacts to non-target populations?
- 4) Does supplementation increase harvest opportunities?

These major questions are very difficult to answer and require large amounts of time, significant physical infrastructure, qualified staff, and environments that are amenable to sampling. It is estimated that evaluations of these questions could take between 8 and 30 years (Table 2). Permanent counting and collection facilities (e.g., Roza Dam Adult Counting Facility, Chandler Bypass Juvenile Facility), highly adaptable and heavily monitored hatchery facilities (Cle Elum Supplementation and Research Facility and three acclimation facilities), an experimental spawning channel (at CESRF), and diverse field and sampling equipment (e.g., electrofishers, boats, tagging trailers) are some parts of the infrastructure that are necessary. The YKFP is also staffed by scientists that are experts in fields such as genetics, ecology, reproductive behavior, population dynamics, fish culture, sampling methods, statistics, and database management. Based on decades of earlier work in the Yakima Basin, we know what types of sampling are feasible and what sample sizes are needed to achieve an appropriate statistical power. Furthermore, the relatively large size of the spring Chinook population allows for operational protocols that are considered to be among the best that are achievable. As such, results from the

YKFP might be considered to be among the best that could be achieved in a supplementation program.



Table 2. Important milestones of the YKFP.

Year	Milestones
1982-1999	Significant amount of baseline data collected
1997	First adult fish taken for broodstock at Roza Dam and transferred to CESRF
1999	First CESRF smolts released from acclimation sites
2000	First CESRF jacks return and spawn in the river, first wild adults placed into experimental spawning channel
2001	First CESRF age 4 fish return and spawn in the river
2002	Hatchery control line initiated (hatchery x hatchery cross)
2004	Wild control line initiated (Naches Basin wild x wild cross)
2004	First hatchery control line smolts released
2005	First age-4 returns from supplementation and wild fish spawning in the river
2005	First hatchery control line jacks return
2006	First hatchery control line age 4 fish return
2010	Third generation of hatchery control line begins
2014	Fourth generation of hatchery control line begins

With a project of this magnitude, many management decisions are made that integrate and balance stewardship, utilization, legal, and scientific values. The Yakama Nation and Washington Department of Fish and Wildlife are responsible for co-managing the natural resources in the Yakima Basin. Policy representatives of these two agencies interact regularly with technical representatives to forge sound management decisions that guide the YKFP. In addition, a formal Science and Management Conference is held annually to disseminate technical information, evaluate and integrate new information into the YKFP, and coordinate future work (Appendix 1). Management decisions are made within the frameworks of adaptive management and risk management.

This report updates findings through December 31, 2015 and is structured around the four critical questions about supplementation.

1. Can integrated hatchery programs be used to increase natural production?

For supplementation to be successful, the number of adult “grandchildren” (natural origin recruits, F_2) produced from parents that spent one generation in the hatchery must be greater than the number of adult grandchildren from parents spawning exclusively in the natural environment. In other words, the product of the hatchery recruitment rate and the recruitment rate of hatchery fish spawning in the wild must be greater than the recruitment rate of fish spawning in the natural environment for two consecutive generations.

During the first generation (F_1), the recruitment rate for hatchery fish must exceed that of fish spawning in the natural environment. In order for this to occur, fish taken into the hatchery must have high survival in the hatchery and they must survive well after they are released into the natural environment. In short, these fish must survive well in both hatchery and natural environments. To increase the probability of success, the CESRF employs best hatchery practices such as: using broodstock that are a representative sample of natural origin fish (e.g., run timing, size); mating the fish using factorial designs to minimize within family variation and maintain genetic diversity; and isolating the offspring of each spawned fish until its disease history has been determined. Those families with high pathogen loadings are culled to reduce the transfer of diseases and increase survival during artificial culture. During the rearing period, fish densities are kept relatively low and three acclimation ponds are used to increase the in-river distribution of returning adults. When the fish are released, the juveniles are allowed to voluntarily leave their raceways. Moreover, different fish culture approaches are being systematically tested. They become part of the standard hatchery practices if they provide survival benefits.

Three innovative rearing approaches have been evaluated: semi-natural rearing, predator avoidance training, and male precocity reduction. Semi-natural rearing environments were compared to best conventional hatchery practices. The semi-natural rearing treatment consisted of raceways equipped with underwater feeders, sidewalls and substrate painted in a camouflaged fashion, suspended in-water structure, and overhead cover. Results indicated that this treatment did not increase post-release survival of smolts in a five-year study. Predator avoidance training using mergansers also did not improve post-release survival of smolts in two years of study. A high rate of precocious maturation in hatchery males (average of 22% of total production) in their second year of life prompted a treatment to attempt to reduce precocity. A small-scale experiment indicated that growth manipulation could reduce precocity. This experiment was expanded to a full-facility experiment in a three-year (brood year (BY) 2002-2004) study to test whether manipulating growth can be used to reduce precocity without significantly impacting post-release survival and adult demographics. Results indicate that growth manipulation decreased precocious maturation but resulted in smaller migrants that survived at lower rates during downstream migration (Bosch et al. 2009) than control fish. This treatment was suspended beginning with BY 2005 based on the preliminary results.

Artificial propagation of Chinook salmon (*Oncorhynchus tshawytscha*) has the potential to alter the abundance and distribution of males that precociously mature in freshwater and thereby influence ecological and genetic interactions in the natural environment. Between 1999 and 2014 the abundance of hatchery origin precocious males on redds was less than 0.05% of the number of fish released, and were less abundant on redds than natural origin precocious males ($P < 0.05$). Hatchery and natural origin precocious males were both found throughout the

spawning range during the spawning season, but differences in distribution between origins were detected ($P < 0.05$). It appears that many precociously mature fish released from the hatchery migrate downstream and fail to migrate back to the spawning grounds or die within the Yakima River prior to spawning time. Hatchery origin precocious males in the Yakima River do not contribute favorably to harvest and may pose ecological risks to non-target taxa. However, most of these fish have a low probability of contributing genes to future generations.

Hatchery fish recruitment (HOR) has been higher than wild fish recruitment (NOR) (Table 3). Because all CESRF fish have their adipose fins clipped and are therefore subjected to higher harvest rates, the disparity between Upper Yakima CESRF and wild/natural returns per spawner has likely been greater than it appears beginning in 2001 when selective fisheries first occurred downstream of the Yakima River mouth in the main stem Columbia River. The redd counts in the Teanaway Basin also increased substantially from a pre-supplementation average of 3 redds per year to a post-supplementation average of 66 (range, 10-253) redds per year (due almost entirely to fish returning from the Jack Creek Acclimation site). Beginning in 2006, the number of natural origin spawners increased substantially compared with historic data which corresponds with the first natural origin returns that may have been produced by returning hatchery spawners in the Teanaway River.

Supplementation and habitat activities in the upper Yakima Basin appear to have significantly increased the number of redds relative to a control stream. The mean difference in upper Yakima (supplemented) and Naches (control) stock redd counts between 1981 and 2000 (before supplementation benefits) was 538 redds. During supplementation (2001-2013), the upper Yakima River redd counts averaged 1,391 higher than the Naches redd counts. The differences between these periods were significant (BACIP; $P = 0.002$). Although higher productivity and distribution is encouraging, these fish must also reproduce successfully and produce fish that survive well in natural environments. We are currently performing an assessment of supplementation in terms of natural origin abundance and productivity of the upper Yakima compared to several reference populations incorporating adjustments for capacity.

Table 3. Estimated number of spawners, adult returns, and returns per spawner (R:S) to the Yakima River Basin by population, brood years 1997-2010.

Brood Year	Upper Yakima Natural			Upper Yakima CESRF			Naches Wild		
	Spawners	Returns	R:S	Spawners	Returns	R:S	Spawners	Returns	R:S
1997	1,204	6,613	5.49	261	8,670	33.22	762	3,996	5.24
1998	390	3,381	8.68	408	9,782	23.98	503	2,833	5.63
1999	1,021	930	0.91	738	864	1.17	358	626	1.75
2000	11,864	8,672	0.73	567	4,819	8.50	3,862	2,346	0.61
2001	12,084	6,071	0.50	595	1,251	2.10	3,912	1,850	0.47
2002	8,073	2,247	0.28	629	2,300	3.66	1,861	965	0.52
2003	3,341	1,257	0.38	441	932	2.11	1,400	464	0.33
2004	10,377	1,828	0.18	597	4,022	6.74	2,197	1,200	0.55
2005	5,713	2,937	0.51	510	4,378	8.58	1,439	940	0.65
2006	3,378	3,888	1.15	419	9,114	21.75	1,163	1,280	1.10
2007	2,322	4,498	1.94	449	6,558	14.61	463	2,288	4.94
2008	4,343	4,668	1.07	457	6,976	15.27	1,074	1,531	1.42
2009	7,056	2,964	0.42	486	3,181	6.55	903	597	0.66
2010 ^a	8,383	4,777		336	4,678		1,207	862	

^a Does not include age-5 adults returning in 2015.

Quantitative management objectives for natural production of upper Yakima and basin total spring Chinook adults have been achieved to date (Table 4 and 5). Management objectives for smolts were being met for the upper Yakima and for the entire basin prior to a change in the entrainment dynamics at Chandler that has prompted a new estimation method (Table 4 and 5).

Table 4. Natural production objectives for *Upper Yakima Basin spring Chinook salmon*. Values were estimated using the EDT and AHA models and are expressed as average annual abundances for different time strata under different harvest scenarios. Properly functioning conditions produce approximately 80% of historic conditions.

Goal/Observed and performance period	Habitat Condition	Natural Origin Upper Yakima Smolts at Chandler	Natural Origin Adults at Roza
10 year goal (2003-2013 smolts; 2005-2015 adults)	Current Yakima Basin at capacity and 2.99% smolt-to-adult survival	134,411- 142,216	2,268-3,014
Actual (2003-2013 smolts, 2005-2013 adults)	Current	NA ^a	3,059 ^b (1,215)

^a currently awaiting validation of estimates using newer method, however previous estimates using other method indicated smolt production met or exceeded performance goal

^b includes jacks

Table 5. Natural production objectives for *Entire Yakima Basin spring Chinook salmon*. Values were estimated using the EDT and AHA models and are expressed as average annual abundances for different time strata under different harvest scenarios. Properly functioning conditions produce approximately 80% of historic conditions.

Goal/Observed and performance period	Habitat Condition	Natural Origin Smolts at Chandler	Natural Origin Escapement
10 year goal (2003-2013 smolts; 2005-2015 adults)	Current Yakima Basin at capacity and 2.99% smolt-to-adult survival	183,450-196,045	3,321-4,393
Actual (2003-2013 smolts, 2005-2013 adults)	Current	NA ^a	4,261 ^b (1,595)

^a currently awaiting validation of estimates using newer method, however previous estimates using other method indicated smolt production met or exceeded performance goal

^b includes jacks

The breeding success, or ability to produce juvenile offspring, of first generation hatchery fish produced from the CESRF was evaluated by creating mixed populations, or test groups, consisting of mixtures of wild and hatchery fish and allowing them to spawn in an artificial stream (BY 2001 through 2006). No differences were detected in the egg deposition rates of wild and hatchery females. Pedigree assignments based on microsatellite DNA, however, showed that the eggs deposited by wild females survived to the fry stage at a 5.6% higher rate than those spawned by hatchery females (Schroder et al. 2008). Subtle differences between hatchery and wild females in redd abandonment, egg burial, and redd location choice may have been responsible for the difference observed. Body size did not affect the ability of females to spawn or the survival of their deposited eggs. How long a female lived was positively related to her breeding success but female origin did not affect longevity. The density of females spawning in portions of the stream affected both egg deposition and egg-to-fry survival. Females spawning in parts of the artificial stream with relatively high instantaneous densities (< 5 m²/female) retained more eggs and produced fewer offspring than those spawning at lower densities (> 19 m²/female). No difference, however, was found in the overall distribution patterns of the two types of females in the artificial stream.

Behavioral differences between hatchery- and wild males were observed (Schroder et al. 2010). Wild males had higher mean attack rates ($P = 0.01$), exhibited greater positive agonism ($P = 0.01$) and higher social dominance ($P = 0.02$) than hatchery males. No difference was seen, however, in the frequency of courting behaviors between the two types of males ($P = 0.16$). All of these behavioral traits were positively correlated with one another and with male breeding success. Additionally, male breeding success increased with body weight; however this trait appeared to be less important than either agonism or courting behavior on the ability of males to produce newly emerged fry. Despite the behavioral differences observed, DNA-based pedigree analyses showed that hatchery and wild males mated with similar numbers of females ($P = 0.39$)

and had comparable breeding success ($P = 0.22$). Consequently, a single generation of hatchery exposure appeared to have a low impact on male breeding success. If the behavioral differences observed are genetically controlled, however, then continued exposure to hatchery conditions would likely reduce breeding success in hatchery males when they spawn under natural conditions.



If we find differences in the adult recruitment between hatchery and wild fish, then it is important to know what caused those differences. Differences could be due to fish culture (environmental effects), genetics, or a combination of both. A large-scale test of the domesticating effects of supplementation and continuous hatchery culture is being implemented to determine if any observed differences are genetic. The primary design consists of comparing three lines- a wild control line, a supplemented line, and a hatchery control line- for a variety of adult juvenile traits. Traits vary in frequency of evaluation from annually to once per generation. By comparing the supplemented line to both controls, we will address two key questions: 1) how much domestication is incurred by a population undergoing YKFP-style supplementation; and 2) how much less domestication is incurred under YKFP-style supplementation than would be incurred under continuous hatchery culture?

Preliminary results indicate that significant differences have been detected in about half of the traits measured and that differences can be attributed to both environmental and genetic causes. For example, we have detected differences in hatchery and natural fish after only one generation of hatchery exposure for the following variables measured on adults: age composition, size-at-age, sex ratio, spawning timing, fecundity, egg weight, adult morphology at spawning, and spawning success. Significant differences in juvenile traits have also been detected: food conversion efficiency, length-weight relationships, agonistic competitive behavior, predator avoidance, and incidence of precocious maturation. Most of the differences have been 10% or less (Knudsen et al. 2006; Busack et al. 2007; Fritts et al. 2007; Pearsons et al. 2007; Schroder et al. 2008). Monitoring of these traits has continued beyond the first generation and pertinent results will be published in peer-reviewed literature once work is completed.



In order to evaluate supplementation effectively, it is important to discriminate between aspects under the control of YKFP personnel (e.g., fish culture) and those that are not. Changes in the environment and harvest management are factors that can have a dramatic effect on natural production. The YKFP has a goal of increasing the productivity and capacity of the Yakima Basin. This is accomplished through a variety of habitat related strategies. Strategies that are used to accomplish this goal include:

- Prioritization of most beneficial habitat actions
- Habitat and water right purchases in priority areas
- Re-establishment of connectivity to productive side channels, floodplains and tributaries
- Habitat restoration
- Assessing habitat protection and restoration actions
- Evaluation and mitigation of land use actions that pose a threat to watershed productivity

However, there are many environmental factors that are outside of the control of the YKFP. For example, the flow management of the Yakima Basin is largely controlled by the United States Bureau of Reclamation. Existing water and land use regulations do not effectively protect watershed functions, and continued population growth and climate change will make watershed management more challenging. Out-of-basin harvest is also outside of the scope of the project and yet can have a large impact on adult recruitment and project evaluation. This is especially true since project monitoring and evaluation requirements mandate the use of extensive marking protocols including adipose fin-clipping, while state and federal fishery managers are increasing efforts to target adipose fin-clipped fish.

While harvest management outside the Yakima Basin is outside of the control of the YKFP, in-basin harvest is influenced by the YKFP. For example, there is selective harvest of hatchery-origin adipose-clipped fish by sport anglers in the Yakima River to manage the proportion of hatchery fish on the spawning grounds and minimize impacts to Naches Basin and upper Yakima natural-origin fish. The Yakama Nation uses maximum proportion management by tribal fishers to reduce impacts to natural populations. The co-managers have successfully

managed all in-basin harvest (tribal and sport) to limit the combined exploitation rate to no more than 25% in most years with a mean rate of 16% since hatchery adults began to return in 2001.

Evaluations have identified that smallmouth bass, northern pikeminnow, and piscivorous birds are consuming large numbers of salmonids. For example, smallmouth bass in the lower Yakima River consumed an average of 188,058 salmonids each year from March 22 to June 16, 1998 to 2002, and of these, only 2,873 were yearling salmonids (primarily spring Chinook salmon). From 1999 to 2002, smallmouth bass predation on all yearling salmonids never exceeded 0.6% of the annual production of hatchery and wild fish combined. Estimated smallmouth bass consumption of hatchery ocean-type (fall-run) Chinook salmon has only comprised up to 4% of the annual production of these fish. The diet of northern pikeminnow is comprised of a high proportion of salmonids, including yearlings. In river estimates have put the population of northern pikeminnow at 142-516 fish per mile. The abundance and consumption rate of northern pikeminnow suggests that predation on smolts may be significant with an expanded consumption estimate of 4,217 salmonid smolts consumed per day between the confluence with the Naches and Prosser Dam during the spring of 2007. Starting in 2013, the WDFW Commission eliminated catch restrictions for non-native fishes such as smallmouth bass. The Yakama Nation is currently performing surveys to identify locations for multiple removal population study to determine management options for piscivorous fishes. Work is also underway to determine the significance of the Yakima River delta as a rearing area for young non-native piscivorous fishes and whether reconnecting the causeway on the east side of Bateman Island would improve conditions for native fishes.

The Yakama Nation monitors avian predator numbers in index sites and reaches and scans hotspots such as Heron rookeries for PIT tags (Sampson et al. 2015) Common mergansers, American white pelicans, double-crested cormorants and gulls are great enough in abundance and bioenergetic capacity to consume large numbers of salmonids. Mergansers were the most significant predator in the upper river, potentially consuming 91-98% of the fish biomass consumed by all bird predators in that stratum. However an earlier dietary analysis of Yakima River mergansers found they eat a broad range of small fish with salmonids only becoming common in their diet during fall/winter. Mergansers have not shown a numeric response to increases in the number of spring Chinook smolts in the Yakima River since the supplementation program began, although they have been shown to congregate at or near acclimation sites when hatchery smolts are present.

Gull numbers remained relatively low during supplementation, with the exception of a notable increase in 2014, in the Yakima Basin and the focus of study has shifted towards American White Pelicans, management of hot spots of predation, and surveys of PIT tags where mortality can be linked to predation. Pelican numbers remained high in the Yakima River during 2014. Although Double Crested Cormorant numbers have remained relatively low and consistent through the years, there is a concern that the new culling program in the lower Columbia River could displace birds to other areas. The YKFP will therefore continue to monitor this species. Contrary to what was previously thought, PIT tag recoveries at Badger Island and the Chandler Juvenile bypass show that pelicans may be targeting juvenile salmonids in the Yakima River.

Approximately 18,000 PIT tags have been detected at heron rookeries between 2008 and 2014. There is evidence to indicate that herons were less successful predators during higher flows and much more successful during low flows and preying on later migrating smolts.



Density dependent relationships between Chinook salmon abundance and growth and survival exist in the upper Yakima Basin. Larger numbers of fall parr are correlated with smaller size, and an asymptotic relationship exists between parent abundance and an index of fall parr abundance. Growth and survival of Chinook parr may be strongly influenced by both high summer flows and river channelization. Rearing areas available to rearing spring Chinook in the upper Yakima basin are reduced by approximately 51 percent when flows are increased each spring to meet irrigation needs, and abundance per unit area decreases significantly with increasing river width in reaches that contain suitable flow conditions ($r_s = 0.35$, $p < 0.01$). Identifying the factors limiting production both temporally and spatially is necessary in order to increase system capacity. Unless the capacity of the environment increases (e.g., altered stream flows, increased bank habitat, increased passage), the natural production benefits of supplementation will be largely confined to years when natural origin fish escapement is below the carrying capacity of the environment.

2. Can integrated hatchery programs limit genetic impacts to non-target Chinook populations?

Genetic impacts to non-target Chinook populations can occur if fish produced from a hatchery stray into areas where other populations or stocks spawn. If hatchery fish interbreed with individuals from these populations, then there is a risk that adaptations or genetic variability among populations will be lost. Straying is a natural phenomenon, keeping levels of diversity high in population groups. The risk occurs from hatchery fish when hatchery fish stray at unnatural levels or to unnatural destinations. Straying of hatchery fish can occur because of inappropriate imprinting or from natural tendencies to seek new spawning areas.

Two measures of straying are commonly measured. The first is the proportion of the returning hatchery adults that do not spawn in their natal areas, which measures straying propensity and the second is the proportion of a non-target population that consists of strays. It is the second that is generally more important for assessment of genetic risk: a small stray rate from a very large population can genetically swamp a small population. Measured either way, however, the YKFP spring Chinook program has very low rates of straying and is within natural levels. Since program inception, a total of 11 adipose-clipped carcasses (assumed hatchery-origin) have been detected on the spawning grounds in the non-target Naches and American River systems during annual spawner surveys. This includes 1 fish from BY1998, 4 from

BY2000, 2 from BY2001, 1 from BY2002, 1 from BY2007, 1 from BY2008, and 1 from BY2009. It is unclear whether all of these were CESRF fish, but assuming they were, they comprised less than 0.25% of the estimated annual spawning escapement into the Naches system in any year.

An analysis of CESRF PIT detections at out-of-basin sites that were not later detected at Roza Dam indicates a potential average annual stray rate to out-of-basin locations of less than 1.6% of all returning CESRF adults. Because almost all this information is based on PIT tag detections at dams, the impact in terms of recipient population is not known. Because of other research objectives, the project only began using the common snout location for placement of coded wire tags (CWT) in most fish beginning with brood year 2004. However, tag recoveries to date indicate the strays to other spawning populations is much less than that estimated using PIT tags.

3. Can integrated hatchery programs limit ecological impacts to non-target populations?

Salmon supplementation and reintroduction programs have the potential to negatively impact other valued fish taxa, which are not the target of enhancement (non-target taxa). We evaluated the impacts of spring Chinook salmon *Oncorhynchus tshawytscha* supplementation and coho salmon *O. kisutch* reintroduction (hereafter supplementation) to non-target fish taxa in the upper Yakima Basin after eleven years of stocking approximately one million yearling smolts annually. Field methods included backpack electrofishing and snorkeling in tributaries, and drift-boat electrofishing in the main stem. We used three sequential steps in our evaluation: First, we determined if spatial overlap in distribution occurred between supplementation fish and non-target taxa. Second, if overlap occurred, we determined if a change in abundance, size, or biomass occurred during supplementation. Lastly, if a change occurred we determined if the change could be reasonably attributed to supplementation. Spatial overlap and changes in abundance, size, or biomass were determined to be significant if they exceeded containment objectives. Salmon rarely overlapped cutthroat trout *O. clarkii* and bull trout *Salvelinus confluentus* in tributaries, but some overlap of cutthroat occurred in relatively high elevations of the main stem, and considerable overlap with rainbow trout occurred in tributaries and the main stem. Salmon overlapped mountain whitefish *Prosopium williamsoni* and sucker species (Catostomidae) in the main stem, and dace (Cyprinidae) and sculpin (Cottidae) species in tributaries. With the exception of steelhead *O. mykiss*, the lower 90% confidence limit of abundance, size, and biomass was above the containment objective for non-target taxa that overlapped significantly with salmon. We used rainbow trout as an analog for steelhead. The lower 90% confidence limit of rainbow trout size in both tributaries and in the main stem, were below our containment objectives for steelhead trout. Comparisons of rainbow trout size in tributaries, and size in main stem sections with relatively high and low salmon abundance revealed that these changes were unlikely to be the result of supplementation (before-after-control-impact-paired site analysis: $P > 0.05$). Our data indicate that early stages of salmon supplementation have not impacted valued species in the upper Yakima Basin beyond predetermined containment objectives.



4. Does supplementation increase harvest opportunities?

Higher rates of harvest can be maintained on populations that are more productive than populations that are less productive. If hatcheries are more productive (more adult recruits returning per adult taken into the hatchery) than natural environments (adults that spawn in the natural environment), then it can support a higher rate of harvest. Risks to less productive stocks (e.g., wild fish) can occur if they are harvested at rates that may be appropriate for more productive stocks. Spring Chinook returns (adults and jacks) to the Yakima River mouth since 2000 have averaged just under 12,500 salmon annually (compared to a pre-supplementation average of fewer than 3,500 fish annually), which has increased harvest opportunity both in and out of the Yakima River Basin. However, at this time it is difficult to assess how much of this improvement is due to natural factors such as improved freshwater and ocean conditions versus supplementation activities. Currently within the Yakima Basin, treaty reserved fisheries have harvested less than 12% of the returning adults on average annually since 1983; and non-tribal fishers are only allowed to keep hatchery fish.

Standard run reconstruction techniques are employed to derive reasonable estimates of harvest from the Columbia River mouth to the Yakima River mouth for spring Chinook. Data from databases maintained by the United States versus Oregon Technical Advisory Committee (TAC) are used to obtain harvest rate estimates for the aggregate spring Chinook population destined for tributaries above Bonneville Dam and to estimate passage losses from Bonneville through McNary reservoirs. These data, combined with the Prosser Dam counts and estimated harvest below Prosser, are used to derive a Columbia River mouth run size estimate and Columbia River main stem harvest estimate for Yakima spring Chinook (assuming Yakima spring Chinook are harvested in Columbia River fisheries at the same rate as all stocks destined for tributaries above Bonneville Dam). These data are being tracked and reported annually by Yakama Nation YKFP (Sampson et al. 2015).

Based on available CWT information, harvest managers have long assumed that Columbia River spring Chinook are not harvested in any abundance in marine fisheries as the timing of their ocean migration does not generally overlap either spatially or temporally with the occurrence of marine fisheries. The Regional Mark Information System (RMIS) is queried regularly for any CWT recoveries of CESRF releases in ocean or Columbia River main stem fisheries. Based on the information reported to RMIS to date, it is believed that marine harvest accounts for about 0-3% of the total harvest of Yakima Basin spring Chinook.

Since 2001, Yakima Basin tribal and recreational fisheries combined have harvested an average of about 1,233 CESRF and 864 wild/natural spring Chinook annually. Also since 2001, in-basin harvest rates have averaged 6.7% on wild and 9.7% on CESRF fish, with tribal harvest rates averaging 11.4% and recreational harvest rates averaging 5.0% of the total Yakima Basin return of spring Chinook. Successful recreational fisheries for spring Chinook in the Yakima River are dependent on several conditions: a large number (preferably greater than 10,000) of returning spring Chinook, a return of wild/natural fish that does not far outnumber the return of hatchery fish, and favorable water conditions. This combination of conditions occurred in 2001, 2002, 2004, and 2008-2015; recreational fisheries were precluded in other years.

Quantitative harvest objectives for the upper Yakima stock and all Yakima basin stocks combined have been met in both the Columbia and Yakima rivers (Table 6 and 7).

Table 6. Harvest objectives for *Upper Yakima Basin spring Chinook salmon*. Values were estimated using the EDT and AHA models and are expressed as average annual abundances for different time strata under different harvest scenarios. Properly functioning conditions produce approximately 80% of historic conditions.

Goal/Observed and performance period	Habitat Condition	Columbia River and Ocean Harvest (hatchery and natural origin fish)	Yakima Basin Harvest (hatchery and natural origin fish)
10 year goal (2005-2015)	Current Yakima Basin at capacity and 2.99% smolt-to-adult survival	1,777-2,590	1,031-1,854
Observed (2005-2014)	Current	1,851 ^a (960)	1,815 (1,230)

^a assumes no marine harvest, numbers in parentheses are 1 standard deviations

Table 7. Harvest objectives for *Entire Yakima Basin spring Chinook salmon*. Values were estimated using the EDT and AHA models and are expressed as average annual abundances for different time strata under different harvest scenarios. Properly functioning conditions produce approximately 80% of historic conditions.

Goal/Observed and performance period	Habitat Condition	Columbia River and Ocean Harvest (hatchery and natural origin fish)	Yakima Basin Harvest (hatchery and natural origin fish)
10 year goal (2005-2015)	Current Yakima Basin at capacity and 2.99% smolt-to-adult survival	1,996-2,879	1,184-2,117
Observed (2005-2014)	Current	1,993 (971)	1,950 (1,312)

^a assumes no marine harvest, numbers in parentheses are 1 standard deviations

All findings in this report should be considered preliminary until published in a peer-reviewed journal. A list of project publications is attached. For further information and accomplishments please check the YKFP website at www.ykfp.org and www.cbfish.org or contact project personnel.

Peer-reviewed Publications Produced in Association with the YKFP (in chronological order)

- Clune, T., and D. Dauble. 1991. The Yakima/Klickitat fisheries project: a strategy for supplementation of anadromous salmonids. *Fisheries* 16:28-34.
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Appendix 1

Yakima Basin Science and Management Conference Statistics

Table 1. Numbers and percentages of Yakima Basin Science and Management Conference participants by affiliation and year from past 5 years.

Numbers					
Affiliation	2011	2012	2013	2014	2015
Washington Department of Fish and Wildlife	41	28	26	25	23
Yakama Nation	55	25	24	27	30
United States Bureau of Reclamation	6	3	4	4	2
Central Washington University	10	3	12	4	4
Bonneville Power Administration	5	4	1	2	5
National Oceanic and Atmospheric Administration	10	2	4	6	1
United States Fish and Wildlife Service	17	13	9	17	9
Other ^a	99	54	52	92	94
TOTAL	243	132	132	177	168

Percentages					
	2011	2012	2013	2014	2015
Washington Department of Fish and Wildlife	17%	21%	20%	15%	14%
Yakama Nation	23%	19%	18%	15%	18%
United States Bureau of Reclamation	2%	2%	3%	2%	1%
Central Washington University	4%	2%	9%	2%	2%
Bonneville Power Administration	2%	3%	1%	1%	3%
National Oceanic and Atmospheric Administration	4%	2%	3%	3%	1%
United States Fish and Wildlife Service	7%	10%	7%	10%	5%
Other ^a	41%	41%	39%	52%	56%
	100%	100%	100%	100%	100%

^a Benton County Conservation District, Blue Leaf Environmental, Cascade Land Conservancy, Cramer Fish Sciences, Columbia River Inter-Tribal Fisheries Commission, D. B. Lister & Associates, Washington Department of Natural Resources, Washington Department of Ecology, Ellensburg High School, Fish Passage Solutions, ICF International, Kittitas Conservation Trust, Kittitas County Conservation District, Mid-Columbia Fisheries Enhancement Group, Nez Perce Tribe, Oncorh Consulting, United States Forest Service, University of California, University of Idaho, University of Washington, United States Geological Survey, Washington Conservation Corps, WRH, Washington Department of Transportation, Washington Water Trust, Yakima Basin Fish and Wildlife Recovery Board, Yakima Basin Joint Board, Yakima County Public Works, Yakima Tributary Access and Habitat Project

Table 2. Percent of attendance that was composed of speakers.

Year	% of Total that were Presenters
2011	16%
2012	26%
2013	30%
2014	23%
2015	21%

Appendix 2

Yakima/Klickitat Fisheries Project Monitoring and Evaluation

Non-target Taxa of Concern Monitoring

BPA Project # 199506325

Report covers work performed under BPA contract #(s) 65604 and 69084

Report was completed under BPA contract #(s) 69084

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Abstract

Salmon supplementation and reintroduction programs have the potential to negatively impact other valued fish taxa, which are not the target of enhancement (non-target taxa). We evaluated the impacts of spring Chinook salmon *Oncorhynchus tshawytscha* supplementation and coho salmon *O. kisutch* reintroduction (hereafter supplementation) to non-target fish taxa in the upper Yakima Basin after eleven years of stocking approximately one million yearling smolts annually. Field methods included backpack electrofishing and snorkeling in tributaries, and drift-boat electrofishing in the main stem. We used three sequential steps in our evaluation: First, we determined if spatial overlap in distribution occurred between supplementation fish and non-target taxa. Second, if overlap occurred, we determined if a change in abundance, size, or biomass occurred during supplementation. Lastly, if a change occurred we determined if the change could be reasonably attributed to supplementation. Spatial overlap and changes in abundance, size, or biomass were determined to be significant if they exceeded containment objectives. Salmon rarely overlapped cutthroat trout *O. clarkii* and bull trout *Salvelinus confluentus* in tributaries, but some overlap of cutthroat occurred in relatively high elevations of the main stem, and considerable overlap with rainbow trout occurred in tributaries and the main stem. Salmon overlapped mountain whitefish *Prosopium williamsoni* and sucker species (Catostomidae) in the main stem, and dace (Cyprinidae) and sculpin (Cottidae) species in tributaries. With the exception of steelhead *O. mykiss*, the lower 90% confidence limit of abundance, size, and biomass was above the containment objective for non-target taxa that overlapped significantly with salmon. We used rainbow trout as an analog for steelhead. The lower 90% confidence limit of rainbow trout size in both tributaries and in the main stem, were below our containment objectives for steelhead trout. Comparisons of rainbow trout size in tributaries, and size in main stem sections with relatively high and low salmon abundance revealed that these changes were unlikely to be the result of supplementation (before-after-control-impact-paired site analysis: $P > 0.05$). Our data indicate that early stages of salmon supplementation have not impacted valued species in the upper Yakima Basin beyond predetermined containment objectives.

Introduction

Despite the long history of stocking hatchery salmon into streams, few large-scale evaluations of impacts to the status of taxa that are not the target of enhancement (non-target taxa; NTT) have been conducted that apply to current stocking practices. Many mechanisms of impacts have been documented (Marnell 1986; McMichael et al. 1999; Hawkins and Tipping 1999), but impacts to NTT growth and abundance have generally not been conclusively demonstrated at scales larger than experimental reaches (Fresh 1997; Weber and Fausch 2003). Most large-scale evaluations of hatchery and wild fish interactions have addressed impacts to naturally produced conspecifics (Nickelson et al. 1986; Chilcote 2003; Nickelson 2003) and/or stocking salmon before the smolt stage (Bjornn 1978; Tripp and McCart 1983). Although these studies are illuminating, most contemporary hatchery salmon programs release smolts, and these smolts are released into areas containing species of concern other than wild conspecifics. Releasing smolts is appealing because they provide the highest adult returns and potentially minimize ecological interactions in the freshwater environment. In order to assess risks of contemporary programs, information about the impacts of smolt releases is needed. This is

particularly true in watersheds containing threatened or endangered NTT, such as Bull Trout *Salvelinus confluentus* and Steelhead Trout *Oncorhynchus mykiss*.

Although it is assumed that releasing smolts poses less ecological risk than stocking earlier life stages, this assumption has not been tested and the practice is clearly not without risk. It is believed that smolts pose lower risks than earlier life stages because they spend less time interacting with NTT. However, hatchery smolts can interact with wild fish during downstream migration and during periods when they residualize in rearing environments. In addition, some hatchery-released yearlings swim upstream of release locations into areas containing listed species (McMichael and Pearsons 2001). Ecological interactions that can occur during migration include competition, predation, behavioral anomalies, and pathogenic interactions (Pearsons and Hopley 1999). If competition does occur, it may be of short duration because hatchery smolts generally move downstream and feed as they migrate or during brief “resting” periods. It is during these “resting” periods that competition may be most intense (Coutant and Whitney 2006).

Chinook *O. tshawytscha* and coho salmon *O. kisutch* are the most commonly cultured Pacific salmon released as yearlings and are the species of consideration in this paper. Studies have demonstrated the potential for stocked salmon to impact wild target and NTT. Hatchery spring Chinook smolts were observed to behaviorally dominate wild smolts and secure the most food and best habitat in laboratory experiments (Pearsons and Ham 2001). Predation by Chinook and coho salmon smolts on naturally produced salmon has also been demonstrated (Sholes and Hallock 1979; Hawkins and Tipping 1999). The release of large numbers of hatchery smolts can change the functional and numerical response of predators to mixed groups of hatchery and wild fish (Peterman and Gatto 1978; Wood 1987; Collis et al. 1995). Depending upon the predator response, the releases can either benefit or harm naturally produced species. Large numbers of hatchery fish can also alter the behavior of wild fish and influence susceptibility to predators or food acquisition (Hillman and Mullan 1989; McMichael et al. 1999). Finally, hatchery fish have the potential to directly transmit pathogens or increase the susceptibility to pathogens by wild fish (Goede 1986; Bucke 1993; McVicar 1997). Similar interactions can occur if “smolts” residualize, although the intensity or manifestation of the interaction may differ. For example, competition is likely to be more potent locally when fish residualize because they remain in an area, as opposed to more temporal occupation of areas during downstream migration. Increased natural production of the target taxa translates into potentially increased interactions in the freshwater rearing area (Pearsons 2002).

Naturally produced offspring of hatchery coho and Chinook salmon have the potential to impact trout and other species of concern. Coho salmon dominate cutthroat trout *O. clarki* in pool habitats and many cutthroat trout are displaced to riffle habitats in the presence of coho salmon, particularly at warmer temperatures (Glova 1984, 1986, 1987). Sabo and Pauley (1997) suggested that size was equally important as species identity in determining competitive dominance among stream-dwelling cutthroat trout and coho salmon. Coho salmon displace some steelhead trout from pools or alter habitat use within pools that they would occupy in the absence of coho (Hartman 1965; Allee 1974, 1981). Growth of steelhead was lower in experiments with high coho densities than low ones (Fraser 1969). Despite overlap in several key habitat and food variables (Dolloff and Reeves 1990; Nakano and Kaeriyama 1995), Dolly Varden *S. malma*, who are ecologically similar to bull trout, generally occupy microhabitats close to the bottom whereas coho occupy the water column (Bugert et al. 1991; Nakano and Kaeriyama 1995).

Growth and abundance of rainbow trout in high elevation streams of the upper Yakima Basin were not affected when salmonid densities were doubled with naturally produced Chinook salmon parr (McMichael and Pearsons 1998). These controlled experiments were conducted in small to moderate size enclosures. The current study extends the findings of McMichael and Pearsons (1998) by evaluating production scale supplementation of spring Chinook salmon throughout tributary and main stem waters. In addition, impacts from coho salmon reintroduction were also evaluated. Furthermore, this study includes evaluation of interactions from hatchery produced smolts, residuals, and naturally produced offspring of hatchery fish and extends the findings of Pearsons and Temple (2007). In that study, impacts of the early stages of supplementation (5 years) were within containment objectives for *O. mykiss*, cutthroat trout, and bull trout.

In addition to the aforementioned trout species, several non-trout fish taxa occupy areas in sympatry with both natural and supplementation origin Chinook and coho salmon in the Upper Yakima Basin (Temple and Pearsons 2012). Taxa of interest in this study include mountain whitefish *Prosopium williamsoni*, dace spp. sucker spp. (Catostomidae), and sculpin spp. (Cottidae). These taxa have the potential to be impacted through direct and indirect interactions with salmon, but with the notable exception of Temple and Pearsons (2012), interactions with non-trout fish taxa has received little attention in the literature. The literature that has reported interactions between non-trout fish taxa and salmon has generally focused on impacts to salmon (Fritts and Pearsons 2004; Murdoch et al. 2005; Fritts and Pearsons 2006).

In this paper, we examine the impacts to trout and NTT during the early-middle stages of a spring Chinook supplementation program and the reintroduction of coho salmon in the Yakima Basin, Washington (Figure 1). It was unknown whether impacts would occur from smolts that migrated downstream of acclimation sites, residuals that distributed themselves up- and downstream of acclimation sites, and/or from an increase in the distribution and abundance of naturally produced offspring of supplemented adults. Thus, NTT that were distributed upstream of acclimation sites were not considered immune from risks because residuals might migrate upstream and overlap with NTT, such as has been observed with hatchery steelhead (McMichael and Pearsons 2001), or an increase in distribution and abundance of naturally produced Chinook salmon could result in overlap that was not observed prior to supplementation. Certainly one of the goals of supplementation is to increase the distribution and abundance of target species.

Concerns about the possibility of hatchery fish having negative impacts on valued NTT in the Yakima Basin prompted the development and implementation of a risk containment monitoring program (Bonneville Power Administration 1996; Busack et al. 1997; Ham and Pearsons 2001). Our primary management interest was to determine if changes in the status of NTT exceeded “specified biological limits” (Regional Assessment of Supplementation Project 1992) or “containment objectives” (Ham and Pearsons 2001) relative to the baseline period in which no stocking occurred. The containment objectives are: 0% impact for bull and steelhead trout; less than 5% impact for mountain sucker *Catostomus platyrhynchus*; less than 10% impact for rainbow and cutthroat trout in the main stem; less than 40% impact for rainbow and cutthroat trout in the tributaries (Pearsons and Hopley 1999; original cutthroat trout containment objective was modified by Yakima/Klickitat Fisheries Project 2004); less than 40% impact for mountain whitefish *Prosopium williamsoni*; 90% impact for sucker species (Catostomidae) in the main stem; 65% and 95% for longnose dace *Rhinichthys cataractae* and speckled dace *R. osculus*, respectively, in tributaries; and 90% impact for sculpin species (Cottidae) in tributaries. These containment objectives were developed based on the status (e.g., abundance) of the NTT, their

value to fisheries, and their relative value compared to the expected benefits of supplementation (Pearsons et al. 1998). Containment objectives for NTT are evaluated relative to baseline abundance, size, biomass, and distribution. If containment objectives are exceeded for any one of these population parameters, then further action is warranted.

We believe that it is important to report results within the context of the life-span of a supplementation program. An early stage of supplementation, such as the “broodstock” stage, is less likely to produce impacts than a later stage such as the late “building” stage (Pearsons 2002). However, waiting to report results until a supplementation program has matured can delay the presentation of important information for up to 30 years, and therefore limit information that could be used to help improve current management decisions (Pearsons 2002). Indeed, many scientific reviews about hatchery and wild fish interactions identify the paucity of information that is available to evaluate these risks (Fresh 1997; Weber and Fausch 2003). Therefore, we present the findings of the early to middle stages of supplementation knowing that impacts could change during later stages.

We acknowledge that impacts in areas of overlap cannot be definitively evaluated without adequate controls. Unfortunately, spatial controls for most large river systems are difficult to find and we could not find an adequate spatial control for trout in the upper Yakima River. However, achievement of management objectives can be evaluated relative to fixed standards, such as containment objectives, and performance of NTT relative to containment objectives can be used to prioritize impact evaluations. For example, if the status of an NTT has not changed after stocking, then it would not be a high priority to evaluate farther. In contrast, if the status of an NTT decreases below a certain level then further evaluation should be initiated. We believe that the containment objectives are suitable triggers for prioritizing evaluation. Furthermore, if environmental conditions do not change appreciably before and during supplementation, then impacts can be evaluated using temporal controls. The results presented in this paper are the early stages of risk containment monitoring associated with the Yakima/Klickitat Fisheries Project (YKFP).

Methods

Study Area

The Yakima River Basin is a large river system that drains into the Columbia River near Richland, Washington. The upper Yakima River Basin, which is the subject of this paper, is located upstream of Roza Dam (Figure 1). Historically large numbers of salmon and steelhead returned to the upper Yakima Basin (Bonneville Power Administration 1996). Coho salmon were extirpated by the early 1980s and spring Chinook salmon have been severely depressed (Bosch 2004). Steelhead (mid-Columbia Evolutionarily Significant Unit) and bull trout are currently listed as threatened (National Marine Fisheries Service 1999; U.S. Fish and Wildlife Service 1998). Rainbow trout in the Yakima River provide one of the best wild trout fisheries in Washington (Krause 1991; Probasco 1994) and westslope cutthroat trout are present in many high elevation tributaries. Mountain whitefish and sucker species are widely distributed in main stem areas, although mountain suckers are rarely observed. Sculpin species are widely distributed in tributary areas, while speckled dace inhabit low elevations and longnose dace inhabit mid-to high elevation areas of tributary streams.

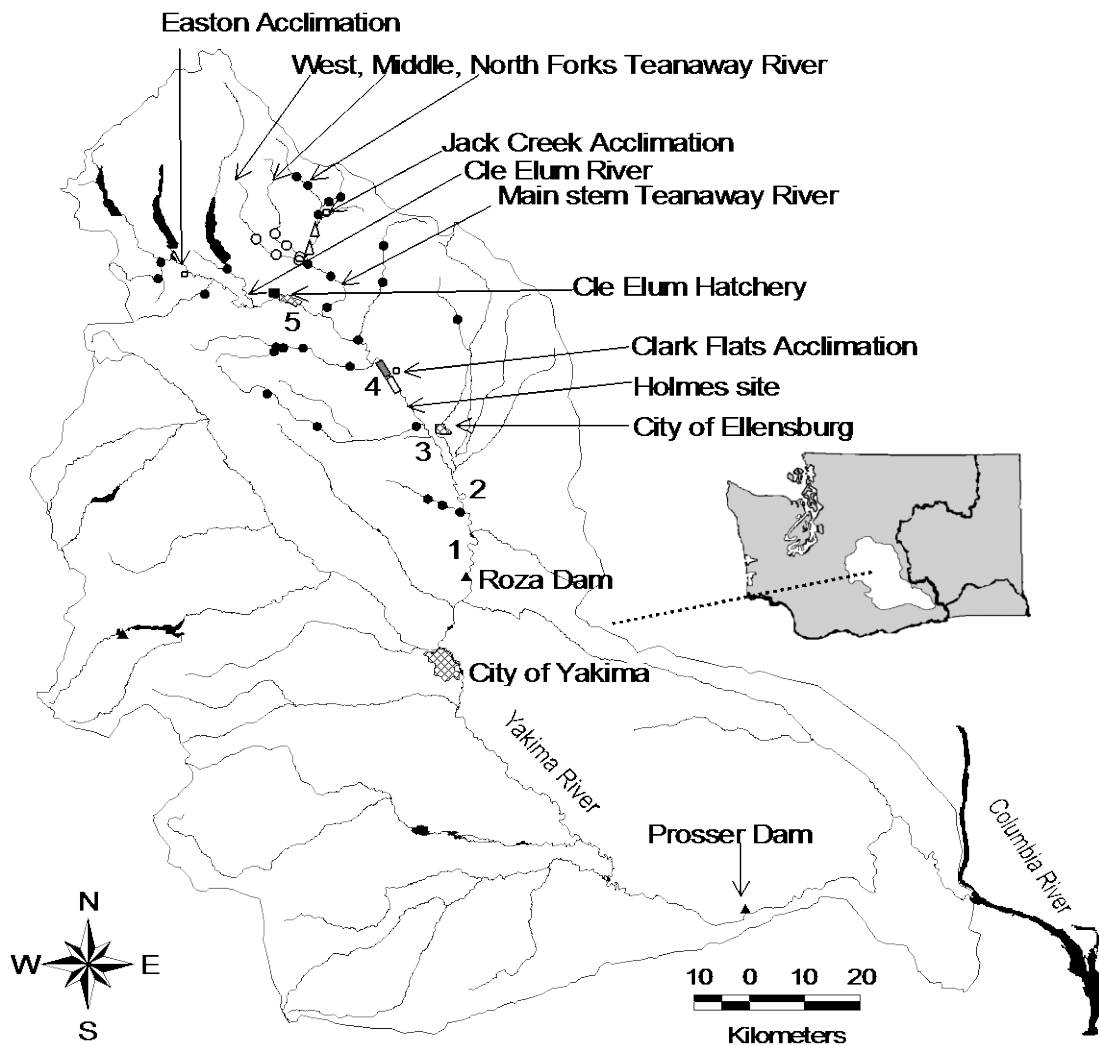


Figure 1. Map of the Yakima River Basin. Darkened circles are tributary NTT distribution sampling sites. Teanaway Basin tributary BACIP control and treatment sites are represented by open circles and triangles, respectively. Bold numbers represent Main stem Yakima River non-target taxa (NTT) monitoring sites where 1, 2, 3, 4, and 5 represent the LCYN, UCYN, EBURG, THORP, and CELUM sections, respectively. Main stem BACIP control and treatment sites are represented by shaded and open rectangles, respectively.

The main stem and tributaries of the Yakima River differ in their physical properties and the way that they are managed. Three dams regulate the flows in the main stem Yakima River. Peak flows during the spring have been truncated and flows are artificially high during the summer. The main stem is heavily fished for trout and a catch-and-release regulation has been in effect since 1990. Tributaries have less flow regulation, but lower portions of tributaries can experience low flows from irrigation withdrawals. The main stem upper Yakima River is currently managed as a catch and release fishery, but anglers are allowed to retain two trout greater than 203 mm in tributary fisheries.

Chinook and coho salmon hatchery programs

Spring Chinook and coho salmon yearlings were released into the upper Yakima Basin for the first time during spring 1999 as part of the YKFP. The goal for both of these species is to increase natural production and to provide harvest opportunity using artificial propagation while keeping adverse impacts within specified biological limits (Bonneville Power Administration 1996; Fast and Craig 1997; Bosch 2004). Approximately 726,813 Chinook salmon smolts have been released annually in the upper Yakima River from 1999 to 2015 (Table 1). Broodstock for the spring Chinook program were natural origin upper Yakima stock collected at a trapping facility at Roza Dam. Chinook were spawned and juveniles were reared at a hatchery facility in Cle Elum, Washington (Knudsen et al. 2006). Spring Chinook salmon yearlings were transported to acclimation sites during January and February and volitionally released into the Yakima River from the Easton and Clark Flats acclimation sites, and from the Jack Creek acclimation on the North Fork of the Teanaway River (Figure 1). Fish were permitted to volitionally migrate between March 15 and May 31 and averaged 120 mm fork length (FL) when released. At the end of May all fish were forced out of the acclimation sites into the river. Mean travel time of migrants from acclimation sites to a detector near Prosser Washington (river kilometer [RKM] 75.6 measured from the confluence with the Columbia River) is about 24.3 days. However, a large number of Chinook salmon residualize in the upper Yakima Basin (Larsen et al. 2004; Pearsons et al. 2005).

Table 1. Numbers and location of yearling spring Chinook salmon released in the upper Yakima River annually.

Brood Year	Release Year	Clark Flats	Easton	Jack Creek	Total
1997	1999	229,290	156,758		386,048
1998	2000	221,460	230,860	137,363	589,683
1999	2001	232,563	269,502	256,724	758,789
2000	2002	285,954	263,061	285,270	834,285
2001	2003	80,782	39,106	250,348	370,236
2002	2004	266,563	290,552	279,789	836,904
2003	2005	273,377	267,711	283,604	824,692
2004	2006	280,598	273,440	231,410	785,448
2005	2007	287,127	281,150	291,725	860,002
2006	2008	209,575	217,932	215,288	642,795
2007	2009	265,907	254,540	250,818	771,265
2008	2010	280,253	287,857	281,195	849,305
2009	2011	279,123	281,395	272,423	832,941
2010	2012	264,420	264,362	265,999	794,781
2011	2013	255,290	248,454	265,438	769,182
2012	2014	256,732	276,210	269,774	802,716
2013	2015	215,933	214,745	216,077	646,755

The coho salmon reintroduction program releases an average of 382,589 coho in the upper Yakima River annually (Table 2). Release locations have been variable since 1999 as the feasibility of re-establishing extirpated coho runs into Yakima Basin tributaries, main stem areas,

and reservoirs is evaluated. Smolt releases have been the primary life-stage released although experimental releases of fry and adults have also been made. Broodstock has generally come from lower Columbia River sources, although one long term goal of the program is to transition to localized broodstock when sufficient numbers of adults return. Mean travel time of migrants from the Holmes release site to a detector near Prosser Washington in 2003 (RKM 75.6) was 41.9 days. Very few coho salmon have been observed to residualize in the upper Yakima River. Coho salmon sizes were not recorded at the time of release, however, two to three months after release, hatchery origin coho smolt fork lengths averaged 157 mm in the migration corridor. Prior to 1999, coho salmon were released in the middle portion of the Yakima River, a considerable distance below Roza Dam. Coho salmon observed in the upper Yakima River prior to 1999 were likely the result of downstream releases. More detail about the study area and background of the supplementation project has been previously described (Busack et al. 1997; Pearsons and Hopley 1999; Ham and Pearsons 2000). Early findings indicate that Chinook and coho hatchery programs are increasing the abundance and distribution of spawners in the upper Yakima Basin (Bosch 2004; Bosch et al. 2007).

Table 2. Numbers of coho salmon released in the upper Yakima River annually.

Brood Year	Release Year	Total Released
1997	1999	498,000
1998	2000	494,676
1999	2001	466,464
2000	2002	314,450
2001	2003	328,000
2002	2004	428,065
2003	2005	332,000
2004	2006	339,791
2005	2007	299,127
2006	2008	382,000
2007	2009	364,412
2008	2010	333,920
2009	2011	419,184
2010	2012	443,030
2011	2013	356,177
2012	2014	315,197
2013	2015	389,519
Average		382,589

General approach

We used three sequential steps in our evaluation of impacts to trout and NTT. First we determined if distributional overlap between supplemented salmon and NTT occurred. Second, if overlap occurred we determined if a temporal change in abundance, size, or biomass occurred after supplementation. Finally, if a change occurred, we determined if the change could be reasonably attributed to supplementation (Table 3). Increases in distribution of the target species

can result in spatial overlap with trout resulting in the potential for impacts. If overlap is less than or equal to containment objectives, then impacts are assumed to be acceptable. However, if distributional overlap exceeds containment objectives, then changes to abundance, size, and biomass were evaluated. Changes were evaluated by comparing the abundance, size, and biomass of trout and abundance and size structure of other NTT before and after salmon were released into the upper Yakima Basin (1999). A change in abundance, size, or biomass does not demonstrate that the hatchery caused the impact. Changes in abundance, size, or biomass can be used to trigger further investigation to identify the causes of changes in monitoring variables. We used a conservative approach to trigger further investigation because of the high inter-annual variability of population parameters (Ham and Pearsons 2000). We used the lower 90% confidence limit (CL) of the response variables (abundance, size, and biomass) as the trigger to initiate further investigation. This ensured that there was a 95% probability that the true value was above the lower CL. If the lower 90% CL was lower than the containment objective, then we attempted to determine causation. Confidence limits associated with parameter estimates were computed based on a *t*-statistic with *n*-1 degrees of freedom (Sokal and Rohlf 1981).

Table 3. Field sampling location (Loc.) and abundance estimation methods used for the following tributary (Trib) and main stem (Main) species; bull trout (B), cutthroat trout (C), rainbow trout (R), dace species (D), sucker species (K), sculpin species (P), mountain whitefish (W), naturally produced spring Chinook and coho salmon (S, O), and hatchery origin spring Chinook salmon (H). Additional abbreviations are as follows: Snork = Snorkeling; BP Efish = backpack electrofishing; DB Efish = drift boat electrofishing; Surrogate Spp. Eff. Exp. = site and size specific surrogate species efficiency expansions; NA = not applicable, MR = mark-recapture; WMI = weighted mean generated from index sites; ORCO = overlap judged relative to containment objectives; 90%CLCO-*t* = 90% confidence limit judged relative to containment objectives; *t* = two-sample student's *t*-test; BACIP = before/after control/impact paired test.

Species	Loc.	Field Method	Spp. Effic. Derived from	Estimation Method	Analysis
Overlap					
B	Trib	Snork	NA	% Overlap	ORCO
C, R, D, K, P	Trib	BP Efish	NA	% Overlap	ORCO
S, H, O	Trib	Snork / BP Efish	NA	% Overlap	ORCO
R, C, S, H, O	Main	DB Efish	NA	% Overlap	ORCO
Before / After					
R, S, H, O	Trib	BP Efish	R, S, H, O	Removal Based WMI	90%CLCO- <i>t</i>
R, C	Main	DB Efish	R	MR Based WMI	90%CLCO- <i>t</i>
S, H, C	Main	DB Efish	R	Surrogate Spp. Eff. Exp.	90%CLCO- <i>t</i>
M	Main	DB Efish	-	Visual count	90%CLCO- <i>t</i>
K	Main	DB Efish	-	Visual count	90%CLCO- <i>t</i>
Causation					
R	Trib	BP Efish	R	Treatment / Control	BACIP
R	Main	DB Efish	R	Treatment / Reference	BACIP
K	Main	DB Efish	-	Treatment / Reference	BACIP

To determine causation we compared abundance, size, and biomass in control and treatment sites (e.g. tributaries). Where this was not possible (e.g., main stem), we compared abundance, size, and biomass of NTT upstream and downstream of a hatchery acclimation facility. We reasoned that the magnitude of interactions between released salmon and NTT would invariably be larger downstream of a release site.

We used abundance and size of age 1 rainbow trout in the main stem Yakima River and all ages of rainbow trout in the tributaries as an analog for evaluating impacts to steelhead trout. We did this because of the difficulty of monitoring impacts to steelhead in the upper Yakima Basin and the similarities in genetics and pre-smolt ecologies of the two forms of *O. mykiss*

(Pearsons et al. 2007). Difficulties of monitoring included; low abundance of steelhead, distinguishing juvenile rainbow and steelhead non-lethally, and sampling upper Yakima steelhead smolts. The spawning populations of rainbow and steelhead trout overlap considerably in space and time, evidence of gene flow has been documented (Pearsons et al. 2007), and it is believed that the juveniles rear together and share similar habitat requirements prior to smoltification. Smoltification in the Yakima Basin generally occurs between ages 1 and 3. Using rainbow trout as an analog does not address impacts that would occur during or after the smolt stage.

Similarly, we used juvenile sized suckers in the main stem Yakima River as an analog for evaluating changes in status of mountain suckers. Mountain suckers are rare in the basin and very few have been observed during field sampling. The low abundance and low containment objective for mountain sucker makes detecting impacts to their status difficult. The use of surrogate species (all subadult suckers) greatly improves our ability to detect impacts, although we must assume mountain suckers and surrogate species respond to supplementation activities similarly. We used subadult suckers because mountain suckers are considerably smaller than the other dominant sucker species, bridgelip *C. columbianus* and largescale *C. macrocheilus* sucker (Wydoski and Whitney 2003).

Environmental variables were compared before and during supplementation to determine if key environmental factors changed between the two periods. In the main stem Yakima River, water temperature and flow were recorded daily at U.S. Bureau of Reclamation (USBOR) gauging stations located throughout the basin (available at <http://www.usbr.gov/pn/hydromet/yakima/yakwebarcread.html>). We evaluated stream flow and temperature recorded for the months October through September at three gauging stations on the Yakima River including one near the city of Cle Elum (YUMW), one near the city of Ellensburg (ELNW), and one in the Yakima River canyon (UMTW) approximately 22 rkm downstream from the city of Ellensburg, Washington. We calculated mean, maximum and minimum daily flows and temperature for each station annually. We recorded wetted stream width, during the time of trout sampling, at 100 m intervals within our main stem sites and used the standard deviation of the widths as an index of stream morphology. We used two-sample student's t-tests to determine if there were differences in these variables before (1990-1998) and during (1999-2015) the supplementation period.

In upper Yakima Basin tributaries, we measured stream wetted width and stream flow at the time of sampling and computed total stream discharge as described by Gallagher and Stevenson (1999); we excluded 2011 in discharge measurements due to equipment failure. We also generated a longitudinal streambed profile in each site by recording water depth at 1 m intervals along the stream thalweg as described by Kaufmann (1987). Variability of thalweg measurements, calculated as the standard deviation (SD) of thalweg depths, were calculated to index habitat complexity and residual pool volume. Thalweg measurements were discontinued in most tributaries in 2005 but re-initiated in 2008 (2007 in the Teanaway), so before-after comparisons were limited to the period 1993-2004, and 2008-2015. Wetted width and stream flow measurements in tributaries during the years 1990-1992 were not the same as the period 1993-2015 so we excluded this time period in our analysis. To describe mean, minimum and maximum daily tributary stream flow that was of a similar nature as the main stem dataset, we compiled flow data collected at the USBOR Teanaway River (TNAW) gauging station. Direct measures of water temperature were not available for both time periods at this gauging station so we used daily air temperatures to generate an index of tributary water temperature from a

SNOTEL gauging station located in the general vicinity of the Yakima River headwaters (Stampede Pass, Washington). The SNOTEL gauging station was operated by the United States Department of Agriculture's Natural Resources Conservation Service both before and during supplementation (available at <http://www.wcc.nrcs.usda.gov/snow>). In tributaries, daily discharge and temperature was evaluated for the year prior to field sampling (e.g., August through July). Finally, we used two-sample student's t-tests to determine if there were differences in the tributary environmental variables between the baseline and supplementation period.

Field Methods

The spatial overlap between NTT and spring Chinook and coho salmon during supplementation was quantified as the mean annual percent of the NTT distribution in sympatry with salmon (Table 3). Spatial overlap was determined annually at the sites indicated in Figure 1 and Table 4. These sites were used because they consistently had NTT in the years prior to supplementation. Annual NTT distribution was calculated from the sum of the rkm that NTT were present. The annual percent overlap was calculated as the number of rkm used by sympatric salmon divided by the number of rkm used by NTT. The mean percent overlap was the mean of the annual overlaps from 1999-2015 and was compared to the containment objectives for NTT.

Bull trout that had the potential to overlap with salmon in the upper Yakima Basin were primarily located in the North Fork of the Teanaway River. The distribution of bull trout was determined by snorkeling and electrofishing conducted throughout the North Fork of the Teanaway River prior to 1999 (WDFW, unpublished data). The spatial overlap between bull trout and supplemented salmon in the North Fork of the Teanaway River was also inventoried by snorkeling and electrofishing. The entire 9.3 km rearing area of bull trout (1999 only) or a systematic sample of nine 1 km reaches (2000-2008) were snorkeled at night (Thurow et al. 2006) to determine if any salmon were present. The nine index sites sampled during 2000-2008 were 200 m long and were generally situated at 800 m intervals up the stream channel. During September, two divers with underwater lights, moved upstream and counted all fish observed. Additional snorkel surveys were conducted during the spring and summer to determine the maximum upstream distribution of spring Chinook and the presence of other species (see residuals methods). We also supplemented our snorkeling surveys with backpack electrofishing described below.

Our index of abundance for bull trout in the North Fork Teanaway River indicate very few, if any, fish were present in the Teanaway Basin during the period 2006-2015. It is highly probable bull trout have been extirpated in the North Fork of the Teanaway River because they have not been observed for the last several years despite high effort sampling by the Yakima Species Interactions Studies crews as well as several other agencies (Meyer 2009, personal communication). We reprioritized our effort to determine bull trout abundance in 2009 by using our highest elevation electrofishing index site as a trigger to determine if snorkeling should be conducted. We established a linear relationship between our bull trout abundance index (snorkeling) and our electrofishing index site bull trout abundance estimates (electrofishing). The relationship indicates that in years bull trout were observed during our summer sampling in our North Fork Teanaway River index site, they were observed during our fall snorkeling surveys ($P < 0.05$). In future years, we will snorkel index sites and establish an annual abundance

index if 1) bull trout are encountered during our routine summer sampling, or 2) if spring chinook distribution expands upstream into index areas that contained bull trout in previous years.

Abundance Indices

Population abundance indices of salmonids in upper Yakima tributary sites were generated from single-pass, backpack electrofishing capture efficiency expansions (Temple and Pearsons 2004; Temple and Pearsons 2007; Table 3). In tributary streams, a crew of three to six people electrofished 200 m long index sites during the day with a backpack electrofisher during summer base flows (Table 4). A single upstream electrofishing pass was performed and attempts were made to net all visible fish. Netted fish were held in perforated buckets in the stream margins until they were processed. All fish were anesthetized, identified to species, and the lengths and weights of salmonids were recorded. We assumed, and later verified, that electrofishing efficiencies would be poor for small fish and restricted our evaluation to salmonids greater than 79 mm FL (Reynolds 1983).

An index of salmonid abundance was calculated by expanding the first pass count by a median capture ratio established for each site during the baseline monitoring phase. Median capture ratios were calculated by dividing the number of fish captured on the first electrofishing pass by a multiple-removal maximum likelihood estimate of the number of fish in the site (Zippen 1958). A minimum of two and a maximum of six electrofishing removal passes were used to generate capture efficiencies at each site once annually during the baseline phase. Median efficiencies were based on between four and eight annual baseline replicates depending on the year each site was established. On average, removal based efficiencies indicated that 75% of the trout greater than 79 mm FL were captured during the first electrofishing pass. Approximately 49% of the sites were sampled with multiple removal methods during both periods because insufficient numbers of annual capture efficiency estimates during the baseline period were available to generate expansions.

Table 4. Distribution monitoring site locations in the upper Yakima Basin. A sub-set of distribution sites are used for rainbow trout (R) or cutthroat trout (C) abundance and size before and after supplementation (B-A) comparisons. Before-After-Control-Impact-Paired (BACIP) abundance and size monitoring control (1) and impact (2) site designations are presented as well. Latitude and longitude positions are presented in degrees, minutes, and seconds (D°M'S"). Main stem Yakima River sampling site locations represent the downstream boundary of each site.

Stream / Site	B-A	BACIP	Years	Latitude	Longitude
Yakima River Tributaries					
Cabin Creek 1			90-15	47° 14' 08.72"	121° 13' 8.72"
Domerie Creek A			97-03	47° 14' 12.73"	121° 04' 6.83"
Jungle Creek A			00-09	47° 20' 47.43"	120° 52' 36.08"
Manastash Creek 1			92-15	46° 59' 39.45"	120° 35' 26.81"
Manastash Creek 3			92-15	47° 2' 21.09"	120° 57' 36.41"
Manastash Creek A			98-07 ^a	46° 59' 30.35"	120° 50' 57.30"
M.F. Teanaway 1	R	1	90-15	47° 15' 53.54"	120° 53' 53.19"
M.F. Teanaway 2	R	1	90-15	47° 16' 51.06"	120° 55' 50.37"
M.F. Teanaway 3	R	1	90-15	47° 17' 57.47"	120° 57' 42.06"
M.S. Teanaway 1			94-15	47° 10' 58.40"	120° 49' 29.80"
M.S. Teanaway 2			94-15	47° 13' 28.32"	120° 48' 15.61"
M.S. Teanaway 3			94-15	47° 15' 6.65"	120° 52' 27.53"
N.F. Teanaway 1	R	2	90-15	47° 16' 53.10"	120° 51' 53.86"
N.F. Teanaway 1.5			01-15	47° 17' 24.67"	120° 51' 35.38"
N.F. Teanaway 2	R	2	90-15	47° 18' 41.97"	120° 51' 31.40"
N.F. Teanaway 2.5			99-15	47° 19' 36.74"	120° 51' 21.15"
N.F. Teanaway C			02-15	47° 19' 56.28"	120° 51' 22.71"
N.F. Teanaway 3	R		90-15	47° 24' 18.24"	120° 55' 56.68"
N.F. Teanaway A			97-04	47° 22' 51.58"	120° 53' 11.52"
N.F. Teanaway B			98-15 ^a	47° 24' 54.67"	120° 56' 20.50"
^b N.F. Teanaway			97-02	47° 24' 60.24"	120° 52' 48.68"
Rkm 18.2-27.2					
Stafford Creek A			97-13	47° 21' 20.08"	120° 50' 0.84"
Stafford Creek B			97-04	47° 21' 48.82"	120° 48' 32.18"
Swauk Creek 1	R		92-15	47° 7' 58.30"	120° 44' 51.39"
Swauk Creek 2	R		92-15	47° 13' 45.90"	120° 41' 46.96"
Swauk Creek 3	R		92-15	47° 19' 15.08"	120° 41' 9.65"
Taneum Creek 1	R		90-15	47° 5' 7.71"	120° 46' 8.35"
Taneum Creek 2	R		90-15	47° 6' 46.99"	120° 52' 58.95"
Taneum Creek 3	R		90-15	47° 6' 37.20"	120° 56' 9.09"
Taneum Creek A			97-15	47° 6' 43.34"	120° 55' 45.11"
Taneum Creek B			97-15	47° 6' 30.69"	120° 56' 11.71"
Umtanum Creek 1			92-15	46° 51' 27.63"	120° 29' 49.55"
Umtanum Creek 1.5			92-04	46° 51' 57.13"	120° 32' 4.26"
Umtanum Creek 2			92-04	46° 52' 27.65"	120° 33' 58.07"

Stream / Site	B-A	BACIP	Years	Latitude	Longitude
Yakima River Tributaries					
W.F. Teanaway 1	R	1	90-15	47° 15' 25.52"	120° 53' 56.00"
W.F. Teanaway 2	R	1	90-15	47° 15' 51.79"	120° 57' 11.25"
W.F. Teanaway 3	R	1	90-15	47° 16' 11.37"	120° 58' 36.13"
Wilson Creek A			97-03	47° 9' 54.93"	120° 30' 38.79"
Mainstem Yakima River					
Lower Canyon	R		91-15	46° 47' 32.32"	120° 27' 23.94"
Upper Canyon	R		91-15	46° 53' 42.55"	120° 30' 10.93"
Ellensburg	R		91-15	46° 58' 47.39"	120° 34' 9.24"
Thorp	R / C	1 / 2	91-15	47° 5' 58.73"	120° 42' 8.48"
Cle Elum	R / C		91-15	47° 10' 24.96"	120° 51' 36.48"

^aIntermittent sampling in later years as time allowed.

^bLatitude and longitude position refers to the lowest reach boundary at river kilometer (RKM) 18.2 (measured from the confluence with the Yakima River).

In the main stem of the upper Yakima River, a crew of two people electrofished 4.2–7.4 km long index sites at night with a drift boat mounted electrofisher as described by Temple and Pearsons (2007). During the electrofishing passes, all fish were identified visually and attempts were made to net all trout. Trout greater than 99 mm fork length were marked with a fin clip and released. One week later, another electrofishing pass was made to determine the proportion of marked and unmarked salmonids. Three types of abundance measures were made based on the abundance of the non-target taxa (Table 3). One type, which was used for rainbow trout, was generated from mark-recapture methods. Main stem rainbow trout were grouped into 25 mm size intervals and mark-recapture abundance estimates for each 25 mm group were generated using the maximum log-likelihood estimator as computed by the computer software program Mark-Recapture for Windows (Mark-Recapture for Windows 1997, Version 5.0 Beta, Montana Fish, Wildlife, and Parks). The general form of the estimator was:

$$\hat{N} = \frac{M}{effic}$$

where \hat{N} was the estimated abundance for each 25 mm size class, M was the number fish marked, and $effic$ was the log-likelihood model estimated capture efficiency. The sum of the abundance estimates generated for each 25 mm grouping represented total abundance per index section. The assumptions associated with the log-likelihood model included; 1) the proportions of marked and unmarked fish remained equal between mark and recapture sampling, 2) marked and unmarked fish were equally catchable, and 3) fish marks were permanent for the duration of the sampling and were not overlooked during the recapture sample. We used a one-week interval between mark and recapture sampling because; 1) it allowed adequate time to redistribute, and 2) it provided adequate time for marked fish to recover (Mesa and Schreck 1989; Peterson et al. 2004).

The abundance and size of age 1 main stem rainbow trout was calculated for each index site each year using a mixture analysis of the length frequency distributions from each individual site (MacDonald and Pitcher 1979). We used R statistical software (R development core team 2005) and the add in package mixdist (Du 2002) to determine the proportion of age 1 rainbow trout in each index site and their mean fork length. Age 1 rainbow trout abundance was calculated by multiplying the total mark-recapture abundance estimate for each index site by the estimated proportion of age 1 rainbow trout present in each index site. We weighted the age 1 rainbow trout abundance estimates for each index section by the amount of stream each index site represented and then generated a mean abundance estimate of age 1 rainbow trout per km for each year. Confidence limits (95%) for the parameter estimates were calculated by incorporating both the spatial and within site variance components (Bohlin et al. 1989).

To evaluate our age 1 rainbow trout parameter estimates in the main stem, we compared them against known main stem rainbow trout population parameters. In 2003, we systematically collected scales from rainbow trout in our main stem index sites. Scales were mounted on gummed cards in the field and acetate impressions of each scale were made in the lab. Scales were projected with a microfiche reader and were aged by counting annuli (Jearld 1983) by a recognized expert with over 25 years of experience. We used a chi square test to compare the known proportion of rainbow trout in each age class in each index site versus the proportions predicted by the mixture analysis. We also used a student's t-test to compare the mean length of known age 1 main stem rainbow trout versus the mean length predicted from the mixture analysis.

Efficiency Expansions

The second type of abundance index, used for main stem cutthroat trout in the THORP and CELUM sections, was based on efficiency expansions. The low abundance of cutthroat trout prevented performing valid mark-recapture estimates due to insufficient recaptures of marked fish. Thus, we calculated an abundance index of cutthroat trout (<250 mm) by expanding the observed number of cutthroat trout captured during electrofishing, by our capture efficiencies for similar sized rainbow trout (Peterson and Zhu 2004). Abundance indices were extrapolated to the reach scale based on reach length.

The next type of abundance index, used for natural origin spring Chinook, was a visual estimate that was expanded by capture efficiencies. The numbers of natural origin spring Chinook visually enumerated during the electrofishing marking runs were expanded by maximum log-likelihood model recapture efficiencies for the smallest sizes of rainbow trout observed (generally between 100 mm and 126 mm). These efficiencies may have been overestimated because naturally produced spring Chinook are slightly smaller than the rainbow trout used to establish the capture efficiencies at this time of year. Thus, the resulting abundance index is likely an underestimate. However, in sites where we could generate capture efficiencies for spring Chinook, our observed rainbow trout electrofishing capture efficiency was within the 95% confidence interval (CI) of the hatchery origin spring Chinook electrofishing efficiency. Thus, we believe that size based efficiencies are reasonable ways of indexing relative abundance because fish size is one of the most important factors that influences electrofishing efficiency (Anderson 1995; Buttiker 1992).

The last type of abundance index, used for mountain whitefish and sucker species (including mountain sucker) in the main stem, was a visual estimate based upon visual counts

taken during boat electrofishing surveys. Visual counts appear to provide an adequate index for abundance. Comparisons between mainstem rainbow trout visual counts correlated significantly with rainbow trout abundance estimates generated from mark-recapture electrofishing. Similarly, visual counts of spring Chinook fall parr correlated significantly with smolt counts at Prosser dam the following year. This information was used as support for the use of visual counts to index abundance for mountain whitefish and sucker species.

Five index sites in the main stem Yakima River were used to represent five contiguous study reaches and the index sites comprised approximately 28% of the upper Yakima River between Roza Dam and the Cle Elum River confluence (McMichael et al. 1992). Each index site was selected to be representative of a larger reach. Estimates were generated from abundance and size data collected in the site and were extrapolated to the reach scale based on reach length. The Lower Canyon (LCYN) site was 4.8 km long, the Upper Canyon (UCYN) site was 5.2 km long, the Ellensburg (EBURG) site was 4.2 km long, the Thorp (THORP) site was 5.7 km long, and the Cle Elum (CELUM) site was 7.4 km long. The reach descriptions are as follows: LCYN extends 19.2 km upstream from Roza Dam to Umtanum Creek; UCYN extends 13.4 km upstream from Umtanum Creek to the Ringer Road access; EBURG extends 21.2 km upstream from the Ringer Road access to the Ellensburg Dam; THORP extends 24.1 km upstream from the Ellensburg Dam to the Teanaway River; and CELUM extends 16.2 km upstream from the Teanaway River to the Cle Elum River (Figure 1).

Size of an NTT was quantified using the mean length of fish collected in our main stem and tributary sites. All rainbow trout that were captured were measured to the nearest mm FL. Mean length of rainbow trout in each tributary site was calculated and then the mean of the site means was calculated for each year. Length of age 1 main stem rainbow trout was calculated for each index site each year using a mixture analysis of the length frequency distributions (MacDonald and Pitcher 1979) from each individual site and then weighted by abundance and section length. Biomass estimates were generated from the product of mean estimated abundance and mean weight. Mean weight of rainbow trout in each tributary site was averaged and then the mean of the averages was calculated for each year. The mean annual weight of age 1 main stem rainbow trout was computed for each index site using the log-transformed length/weight regression from mean lengths of fish in individual annual index sites and then weighted by abundance and section length.

Size structure for mountain whitefish, sucker species, and mountain suckers were based upon visual counts of fish in each size class. Since fish were generally not handled while visual counts were performed, an index of size was based upon the proportions of size classes of fish observed during electrofishing. For mountain whitefish, we used the proportion of subadults (≤ 250 mm TL) relative to the total whitefish observed for evaluating changes to size. For sucker species, we used the proportion of adults (≥ 250 mm TL) relative to the total suckers observed during electrofishing. For our mountain sucker size index, we used the proportion of subadult suckers (≤ 250 mm TL) relative to the total suckers observed during electrofishing. Proportions of fish for the size groupings were arcsine square-root transformed prior to analysis. Estimates of biomass for visually observed species could not be accurately calculated using the data we collected.

Residualization of Hatchery Smolts

Abundance estimates and the presence of residualized hatchery spring Chinook and coho salmon present in the Yakima River from mid-September to mid-October for release years 1999-2015 were calculated using observed abundance and boat electrofishing recapture efficiencies calculated for similar sized rainbow trout. We calculated recapture efficiencies of similar sized rainbow trout utilizing mark-recapture methods in main stem Yakima River electrofishing index sites. The rainbow trout recapture efficiencies were applied to the number of hatchery residuals netted during the mark runs in each index section. A final estimate of hatchery residual abundance was expanded to the reach scale based on reach length.

Distribution and relative abundance of residuals was also estimated by snorkeling the North Fork Teanaway and main stem reaches. The farthest upstream presence of residuals was estimated in the North Fork Teanaway by snorkeling upstream of the acclimation site until no residuals were observed in three sequential pools. Snorkel surveys occurred during the time of maximum distribution from June through September. Snorkeling observations to determine presence or absence were also supplemented by electrofishing.

Natural Production

Spring Chinook salmon natural production occurs primarily in the main stem Yakima River upstream from the city of Ellensburg, Washington (Figure 1). The most intense spawning activity has been observed upstream from the Cle Elum hatchery facility, and in the general vicinity of the Easton acclimation facility (Figure 1). Some spawning activity has also been observed in the Cle Elum and main stem Teanaway Rivers although redd counts indicate these tributaries generally contribute a relatively small proportion of the total redds counted in the upper Yakima River Basin annually.

Abundance indices (fish/km) and 95% CL of naturally produced spring Chinook and coho salmon were generated for the main stem Yakima River and tributary index monitoring sites for the baseline (1990-1998) and during supplementation periods (1999-2015). Spring Chinook and coho salmon visually enumerated during boat electrofishing were expanded by rainbow trout electrofishing efficiencies as previously described. In the tributary index monitoring sites, spring Chinook abundance indices were generated from multiple removal capture efficiencies established during the baseline phase. Due to the low abundance of juvenile coho salmon and their similar size with Chinook salmon, we applied spring Chinook salmon capture efficiencies to the number of coho salmon captured in the first electrofishing pass to obtain an index of coho abundance. We used a two-sample student's t-test to compare differences in the abundance of salmon before and during the supplementation period.

Before-After and Causation Analysis

Changes in NTT abundance, size, and biomass after stocking began were expressed as percent changes prior to stocking and were judged to warrant further attention if the percent change in before-after samples decreased beyond the containment objectives. We did not confine ourselves to have statistically significant results to trigger further attention because of the chances of committing a type II statistical error from a test with low power (Ham and Pearsons

2000). Impacts to NTT are difficult to detect because of high interannual variation of response variables and the low number of annual surveys available to isolate the impacts that occur during the initial stages of supplementation (Ham and Pearsons 2000; Ham and Pearsons 2001; Pearsons 2002). For example, prospective power analyses indicated that abundance impacts of <19% were not statistically detectable after 5 annual surveys (Ham and Pearsons 2000). Based on these constraints, only large impacts will be statistically detectable. However, one-tailed student's t-tests were used to determine if the results were statistically significant and 90% CL were calculated to aid with interpreting the magnitude of the results and triggering the next phase of evaluation.

We used linear models to test the hypothesis that the log length-log weight relationships of main stem age 1 rainbow trout in the main stem, and rainbow trout in tributaries did not differ between the baseline (1990-1998) and supplementation (1999-2015) periods. For both main stem and tributary rainbow trout, a homogeneity of slopes test was first performed to determine if the slopes of the log transformed length/weight relationships were significantly different between periods. The log length-log weight relationships for both main stem and tributary rainbow trout before vs. during the supplementation period did not meet the assumptions of equal slopes required for an analysis of co-variance statistical test (ANCOVA), so we tested the relationship using a similar test (separate slopes model) that does not require this assumption.

Although age 1 rainbow trout are used for evaluating impacts to rainbow trout and serve as an analog for steelhead trout in the main stem, we also evaluated changes in the response variables of catchable sized main stem rainbow trout. Since large trout are valued as a fishery resource, we calculated mean size, abundance, and biomass of all main stem rainbow trout greater than 249 mm FL. The field methods and calculations for these larger fish were the same as was described for age 1 rainbow trout in the main stem.

When NTT exhibited decreases in abundance, size, or biomass below the lower 90% CL, we attempted to determine if the change was caused by supplementation. In the Teanaway Basin and in the main stem near Thorp, we used a before-after-control-impact-paired site analysis (BACIP; Stewart-Oaten et al. 1986) to evaluate changes to the abundance, size, or biomass of NTT. This test evaluates the annual paired differences in control and impact sites before and after stocking. Paired differences before and after stocking were compared with a Mann-Whitney U-test. We used a subset of sites and field methods for the BACIP (Table 3). The treatment sites were based on their proximity to acclimation sites and the control sites were selected based on their geographic proximity to treatment sites. Two sites located downstream from the Jack Creek acclimation facility on the North Fork Teanaway River and three sites in the main stem Teanaway River were used as treatment sites and three sites on each of the Middle and West Forks of the Teanaway River and one upstream site in the North Fork Teanaway River were the paired controls (Figure 1). The "before" period was from 1990-1998 and the "after" period was from 1999-2015. When BACIP results were significant, we investigated relationships between core variables using simple linear regression.

The data from the Thorp main stem site was divided into control and impact sites after 1992 and the "before" and "after" periods were the same as for the Teanaway River. The area downstream from the Clark Flats acclimation site was the impact site and the area upstream from the acclimation site was the control. Abundance estimates and associated 95% CL generated for the Thorp site were partitioned by the proportions of fish marked in either the treatment or control section during the electrofishing marking runs. Rainbow trout located immediately downstream from the Clark Flats acclimation facility had the potential to interact with all

upstream spring Chinook and coho smolt releases as well as residual salmon that did not migrate to the ocean. Rainbow trout immediately upstream from the Clark Flats acclimation facility had the potential to interact with migrating smolts and residualized Chinook salmon from the other upstream release locations in the upper Yakima Basin. In essence, we tested if the fish released from the Clark Flats acclimation site caused additional impact to rainbow trout beyond what occurred from releases farther upstream. No controls were available for cutthroat trout in the main stem.

Results

General approach

Comparisons of environmental variables measured in main stem and tributary areas before and during supplementation indicated that there were generally no significant differences in the variables evaluated (Table 5). We did observe a significant increase ($P < 0.01$) in the minimum daily stream discharge in the main stem Yakima River during the supplementation period. We hypothesize that increased minimum daily stream discharge would benefit NTT and would not confound our evaluation because the increase was not significantly correlated with our monitoring variables (e.g., age 1 rainbow trout size, $P = 0.24$; cutthroat trout size, $P = 0.63$).

Table 5. Environmental variables and results from two-sample student's t-tests of before versus during supplementation periods in the Yakima Basin. Main stem variables were based on water year (October through September of the following year as recorded at three gauging stations) and tributary mean, minimum (Min.), and maximum (Max.) discharge and mean air temperature (based on August through July of the following year) recorded at a single tributary gauging station or weather station. Main stem standard deviation of stream width was recorded at the time of sampling. Tributary mean summer discharge, mean wetted width, mean thalweg depth, and standard deviation of thalweg depth were recorded at the time of sampling. Degrees of freedom (df) were 24 for all comparisons unless otherwise noted.

Environmental Variable	Before Mean	During Mean	<i>t</i>	<i>P</i>
Main stem				
Mean Daily Discharge (m ³ /s)	61.20	57.61	0.61	0.55
Min. Daily Discharge (m ³ /s)	13.42	17.88	-3.07	<0.01
Max. Daily Discharge (m ³ /s)	239.39	209.01	0.65	0.52
Mean Daily Water Temperature (°C) ^a	8.69	8.86	-0.71	0.48
Min. Daily Water Temperature (°C) ^a	0.30	0.67	-1.06	0.30
Max. Daily Water Temperature (°C) ^a	17.81	18.36	-1.23	0.23
Standard Deviation of Stream Width (m) ^b	12.28	13.08	-0.97	0.34
Tributary				
Mean Daily Discharge (m ³ /s)	10.13	9.32	0.56	0.58
Min. Daily Discharge (m ³ /s)	0.29	0.36	-1.42	0.16
Max. Daily Discharge (m ³ /s)	100.62	76.82	1.14	0.26
Mean Summer Discharge (m ³ /s) ^b	0.22	0.29	-0.93	0.36
Mean Daily Air Temperature (°C)	3.93	4.40	-1.37	0.18
Mean Wetted Width (m) ^c	6.18	7.09	-1.96	0.06
Mean Thalweg Depth (m) ^c	0.30	0.31	-1.03	0.31
Standard Deviation of Thalweg Depth (m) ^c	0.15	0.13	1.77	0.15

^a Degrees of freedom (df) was 22 for these tests.

^b Degrees of freedom (df) was 21 for these tests.

^c Degrees of freedom (df) was 20 for these tests.

Field Methods

Results from statistical tests indicated that using the mixture analysis to determine the parameter estimates from the length frequencies of main stem age 1 rainbow trout were valid. Chi squared tests of known versus predicted proportions of main stem rainbow trout in the population were not significantly different in the LCYN ($X^2 = 0.14$; 4 df; $P > 0.99$), UCYN ($X^2 = 0.03$; 2 df; $P > 0.99$), EBURG ($X^2 = 0.10$; 4 df; $P > 0.98$), THORP ($X^2 = 0.12$; 4 df; $P > 0.99$), or CELUM index sites ($X^2 = 0.10$; 4 df; $P > 0.99$) during 2003. In addition, student's t-tests of the

known versus model predicted mean length of age 1 main stem rainbow trout were not significantly different in the LCYN ($t = -2.96$; 404 df; $P > 0.99$), UCYN ($t = -0.85$; 270 df; $P > 0.60$), EBURG ($t = -0.44$; 262 df; $P > 0.34$), THORP ($t = -0.89$; 174 df; $P > 0.63$), or CELUM ($t = -0.16$; 200 df; $P > 0.13$) index sites.

Residualization of Hatchery Smolts

Many spring Chinook salmon did not migrate to the ocean after release (residuals) and may have interacted with trout. In contrast, very few coho salmon residuals were observed (Table 6). Residuals were most concentrated below acclimation sites during the spring and summer, but were found in all reaches of the main stem that we sampled. Residuals were observed approximately 1-2 km upstream of the acclimation site in the North Fork Teanaway during most years and also migrated upstream into the Cle Elum River. Many residuals were observed in the main stem as late as September and October.

Table 6. Estimated annual abundance (fish/km) of spring Chinook and coho salmon residuals in the main stem Yakima River sampling sections. The LCYN section is the Lower Canyon, UCYN is the Upper Canyon, EBURG is Ellensburg, THORP is Thorp, and CELUM is Cle Elum section.

Year	LCYN	UCYN	EBURG	THORP	CELUM	TOTAL
Spring Chinook Residuals						
1999	12	5	3	5	0	25
2000	12	5	1	25	3	46
2001	196	71	85	71	0	423
2002	6	0	7	2	0	15
2003	23	8	3	5	0	39
2004	80	21	29	9	0	139
2005	34	29	3	0	0	66
2006	1	1	0	0	0	2
2007	5	9	6	8	0	28
2008	8	10	4	9	0	31
2009	102	43	22	23	0	190
2010	25	19	5	2	0	51
2011	8	10	4	9	0	30
2012	25	34	17	35	0	110
2013	29	36	22	64	0	150
2014	13	29	10	11	0	104
2015	17	16	2	3	0	54
Coho Salmon Residuals						
1999	7	0	0	0	4	11
2000	0	0	0	0	0	0
2001	3	0	0	0	0	3
2002	0	0	0	0	0	0
2003	0	0	0	0	0	0
2004	0	0	11	0	7	18
2005	0	0	25	0	0	25
2006	0	0	5	0	0	5
2007	0	0	20	0	0	20
2008	0	0	0	0	0	0
2009	0	0	0	0	0	0
2010	0	0	0	0	0	0
2011	0	0	0	0	0	0
2012	0	0	0	0	0	0
2013	0	0	0	0	0	0
2014	0	0	0	0	0	0
2015	0	0	0	0	0	0

Natural Production

We did not observe coho salmon in our tributary index monitoring sites before the supplementation period (0 coho/33 sites). During the supplementation period (1999-2015) we observed coho in 11 of our index sites (between 25 and 36 sites sampled annually) averaging 29 ± 20 per km (mean \pm 95%). This increase was statistically significant ($t = -2.23$; 24 df; $P=0.04$). Our coho abundance index calculated for the main stem Yakima River averaged 0.10 ± 0.25 (mean \pm 95%) coho per river km before supplementation releases. During supplementation, we estimated 3.1 ± 2.0 coho per km (mean \pm 95%). The increase in main stem coho salmon abundance was not statistically significant ($t = -1.74$, 21 df, $P= 0.09$). Since 2009, snorkelers have observed increasing numbers of coho in mixed pods with Chinook salmon in the Yakima River upstream from our monitoring sites relative to previous years (WDFW unpublished data).

Before supplementation, our abundance index was 23 ± 17 (mean \pm 95%) spring Chinook salmon per km in our tributary monitoring sites and during the supplementation period it was 82 ± 41 per km (mean \pm 95%). The observed increase during this period was statistically significant ($t=-2.13$; 24 df; $P=0.04$). In the main stem Yakima River, our abundance index for spring Chinook salmon indicated there were approximately $1,987 \pm 843$ fish per km during the baseline period while there were $2,368 \pm 670$ fish per km during the supplementation period. The observed difference was not significant ($t=-0.67$; 21 df; $P = 0.51$).

Overlap

The degree of trout overlap with salmon was highest in main stem areas, intermediate for cutthroat and rainbow trout in tributaries, and absent for bull trout (Figure 2). There was no overlap of salmon and bull trout in our index sites. In fact, the shortest distance between the uppermost distribution of Chinook salmon and the lowermost distribution of bull trout was approximately 8 km. Cutthroat trout and supplemented spring Chinook overlapped in distribution in both tributary and main stem Yakima River areas. The distributional overlap in tributary streams was approximately 11%, confined to relatively moderate elevations, and was less than the 40% containment objective (Figure 2). Salmon overlapped 100% of the main stem distribution of cutthroat trout (Figure 2). In tributaries, salmon overlapped 50% of the distribution of rainbow trout. Overlap was predominately confined to lower portions of tributaries (e.g., Swauk Creek 1 and Umtanum Creek 1) and farther upstream in the North Fork Teanaway River. However, salmon did not overlap rainbow trout in high elevation portions of tributaries.

There was also extensive overlap between rainbow trout, sucker species, and mountain whitefish and salmon in the main stem (100%; Figure 2). Salmon overlapped in distribution with longnose dace (60%) and speckled dace (73%) in tributaries, although mean overlap was less than the containment objectives for both species. Salmon overlapped sculpin species 19% in tributaries but this was less than the containment objective. Finally, there was 23% overlap in distribution between sucker species and salmon in tributary streams, although this was also less than the containment objective.

Data that were collected at similar times and sites by snorkeling and electrofishing methods were consistent with each other. For example, in areas that we found salmon, rainbow trout, cutthroat trout or bull trout, they were detected with both electrofishing and snorkeling

methods. In addition, we did not capture any salmon when we electrofished areas where bull trout were present.

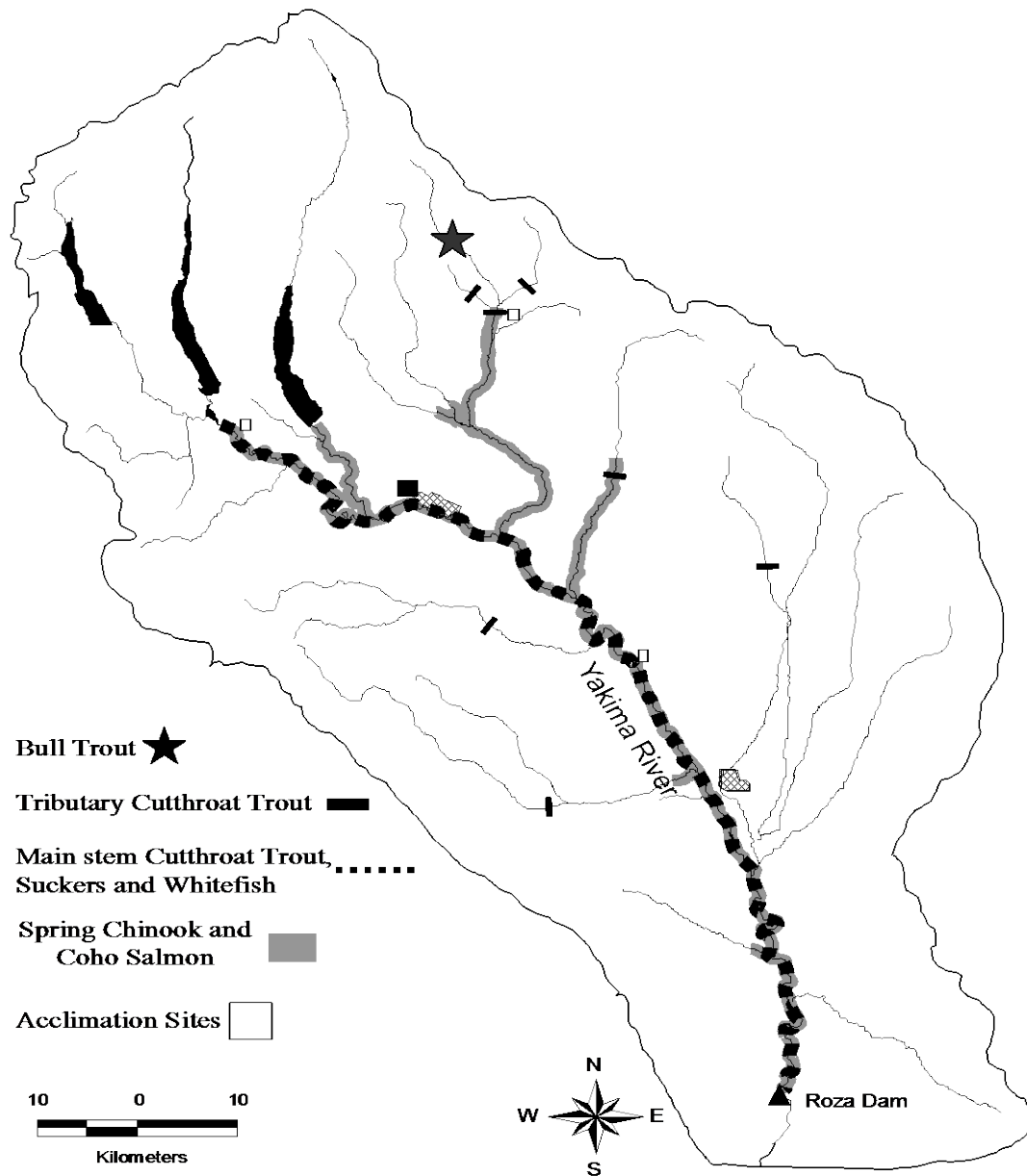


Figure 2. Map of species distributions in the upper Yakima Basin. Spring Chinook and coho salmon distributions are shaded grey. The lowest elevation observations of bull trout and cutthroat trout in tributary streams are marked with stars and bars, respectively. Cutthroat trout, suckers and mountain whitefish distribution in the main stem is marked as a dashed line. The Cle Elum hatchery facility is marked with a black square and hatchery acclimation sites are marked with open squares. Rainbow trout are widely distributed throughout the basin and are not marked on the map.

Before-After Analysis

Rainbow (age 1) and cutthroat trout (<250mm), mountain whitefish, and sucker species in the main stem, and rainbow trout in tributaries (all ages; analog for steelhead) exhibited distributional overlap with salmon that were outside the containment objectives and therefore we compared their abundance, size, and biomass (salmonids) before and after stocking began. The mean abundance and 90% CL of sympatric rainbow trout (all ages) was $30 \pm 16\%$ higher in the tributaries and $37 \pm 15\%$ higher in the main stem (age 1) in the years when supplementation occurred than during the baseline phase (Tables 7 and 8; Figure 3). The mean abundance of cutthroat trout (<250 mm) was $342 \pm 134\%$ CL higher in the main stem during supplementation than during the baseline phase (Table 7; Figure 3). The mean abundance of sub-adult mountain whitefish increased $112 \pm 37\%$ CL during supplementation period, while the mean abundance of sucker species adults decreased $44 \pm 6\%$ CL and the decrease was significant ($P < 0.001$), although it was within our containment objectives (Figure 4). Finally, we observed a $27 \pm 17\%$ CL increase in sub-adult sized sucker abundance (analog for mountain sucker) during supplementation and the lower 90% CL did not exceed our containment objective (Figure 4).

During the supplementation period, the mean and 90% CL of rainbow trout size (age 1) in the main stem indicated that size decreased by $5 \pm 2\%$ (Table 7; Figure 3). Slopes between log length-log weight of age 1 rainbow trout in the main stem were not significantly different before and during supplementation ($P = 0.80$). An ANCOVA revealed the average weight of fish for a given length was significantly greater during the supplementation period ($P = 0.002$, Figure 5). In addition, biomass increased by $19 \pm 14\%$ CL. Similarly, the mean and 90% CL of cutthroat trout size (<250 mm) in the main stem indicated a $2 \pm 3\%$ CL decrease, and an increase in biomass of $309 \pm 858\%$ CL (Table 7; Figure 3). The size of rainbow trout in the tributaries (all ages) decreased slightly during the supplementation period ($-1 \pm 2\%$ CL; Table 8; Figure 3). Slopes between log length-log weight for rainbow trout in tributaries (all ages) were not significantly different before and during supplementation ($P = 0.27$). An ANCOVA indicated the mean weights at each length were not significantly different before versus during the supplementation period ($P = 0.08$; Figure 5). Additionally, tributary rainbow trout biomass (all ages) increased by $26 \pm 11\%$ CL (Table 8; Figure 3). Our index of mountain whitefish size indicated that the proportions of subadults observed increased $11 \pm 2\%$ CL during the supplementation period (Figure 4). Our index of sucker species size indicated that the proportion of adults decreased $42 \pm 7\%$ during supplementation, and although the decrease was significant ($P < 0.001$), it was still well within our containment objectives (Figure 4). Our index of mountain sucker size indicated a $24 \pm 8\%$ CL increase in the proportion of subadults during the supplementation period (Figure 4).

The mean abundance, size, and biomass of catchable sized main stem rainbow trout (>249 mm) did not decrease during supplementation. The mean abundance of rainbow trout greater than 249 mm increased by $13 \pm 13\%$ (mean \pm 90% CL), mean size increased by $2 \pm 1\%$, and biomass increased by $41 \pm 15\%$ during supplementation when compared to baseline conditions.

The only NTT with parameter estimates outside of the containment objectives was steelhead, which uses rainbow trout as an analog. The lower 90% CL for age 1 rainbow trout size in the main stem and rainbow trout size (all ages) in the tributaries were exceeded, so we tested whether the decrease was caused by supplementation.

Table 7. Annual abundance (fish/km), size (mm, FL), and biomass (kg/km) estimates and associated 95% confidence intervals of age 1 rainbow trout and cutthroat trout less than 250 mm fork length in the main stem Yakima River.

Year	Abundance		Size		Biomass	
	RBT	CUT	RBT	CUT	RBT	CUT
1990			210 ± 33	237 ± 5		
1991	189 ± 67	11 ± 14	205 ± 27	237 ± 11	19 ± 14	1.6 ± 3.2
1992	151 ± 28	1	217 ± 31	242	18 ± 7	0.1
1993	193 ± 48	6 ± 17	232 ± 36	238 ± 3	27 ± 11	0.8 ± 3.5
1994	180 ± 33	2 ± 1	217 ± 32	225 ± 17	21 ± 8	0.3 ± 1.4
1995	190 ± 54	6 ± 17	235 ± 34	239 ± 6	28 ± 12	0.9 ± 3.5
1996	182 ± 27	5 ± 11	217 ± 32	239 ± 10	22 ± 7	0.7 ± 2.4
1997	272 ± 49	10 ± 44	203 ± 35	239 ± 5	27 ± 10	1.4 ± 8.9
1998	130 ± 20	16 ± 84	212 ± 34	230 ± 5	15 ± 6	2.0 ± 16.8
1999	182 ± 25	12 ± 25	217 ± 33	236 ± 5	22 ± 7	1.8 ± 5.1
2000	214 ± 40	13 ± 1	210 ± 36	227 ± 13	24 ± 10	1.8 ± 1.4
2001	384 ± 81	18 ± 85	206 ± 32	238 ± 7	41 ± 16	2.5 ± 17.1
2002	207 ± 39	7 ± 42	203 ± 31	232 ± 6	20 ± 9	0.9 ± 8.4
2003	230 ± 41	10 ± 34	207 ± 30	234 ± 7	24 ± 9	1.3 ± 7.0
2004	275 ± 19	16 ± 34	223 ± 32	234 ± 5	35 ± 15	2.3 ± 6.9
2005	272 ± 20	28 ± 142	213 ± 32	229 ± 5	30 ± 9	3.4 ± 28.6
2006	150 ± 12	16 ± 11	216 ± 34	235 ± 5	17 ± 7	2.1 ± 2.5
2007	233 ± 17	22 ± 35	210 ± 33	233 ± 5	26 ± 8	3.1 ± 7.1
2008	264 ± 26	24 ± 61	204 ± 33	229 ± 7	26 ± 9	3.0 ± 12.3
2009	156 ± 29	44 ± 138	188 ± 29	231 ± 3	12 ± 3	5.8 ± 27.8
2010	233 ± 48	32 ± 111	197 ± 36	230 ± 5	21 ± 7	4.1 ± 22.3
2011	273 ± 23	39 ± 63	199 ± 34	227 ± 4	26 ± 9	5.0 ± 12.8
2012	270 ± 30	70 ± 250	192 ± 33	226 ± 5	23 ± 8	8.7 ± 50.3
2013	359 ± 38	80 ± 254	196 ± 27	228 ± 5	32 ± 10	10.0 ± 51.1
2014	342 ± 46	71 ± 169	206 ± 34	227 ± 4	36 ± 11	8.7 ± 34.0
2015	272 ± 15	37 ± 156	213 ± 31	235 ± 4	31 ± 15	4.7 ± 31.3

Table 8. Annual abundance (fish/km), size (mm, FL), and biomass (kg/km) estimates and associated 95% confidence intervals for rainbow trout in Yakima River Basin tributary streams.

Year	Abundance	Size	Biomass
1990	241 ± 129	136 ± 8	8 ± 13
1991	204 ± 102	131 ± 8	6 ± 8
1992	375 ± 240	130 ± 5	11 ± 24
1993	317 ± 158	131 ± 7	9 ± 17
1994	328 ± 129	132 ± 8	11 ± 15
1995	213 ± 118	139 ± 8	7 ± 14
1996	165 ± 109	133 ± 8	5 ± 11
1997	294 ± 119	132 ± 5	8 ± 11
1998	442 ± 174	138 ± 7	15 ± 25
1999	288 ± 175	135 ± 8	12 ± 27
2000	318 ± 135	144 ± 8	11 ± 21
2001	464 ± 178	129 ± 3	12 ± 17
2002	321 ± 131	132 ± 6	10 ± 15
2003	291 ± 142	132 ± 5	8 ± 14
2004	243 ± 135	142 ± 5	9 ± 15
2005	349 ± 163	127 ± 5	9 ± 16
2006	434 ± 171	134 ± 5	13 ± 20
2007	368 ± 153	138 ± 4	12 ± 18
2008	331 ± 166	138 ± 7	11 ± 19
2009	256 ± 123	138 ± 12	9 ± 19
2010	548 ± 243	127 ± 5	15 ± 25
2011	486 ± 215	124 ± 7	12 ± 20
2012	490 ± 163	124 ± 4	13 ± 15
2013	571 ± 232	129 ± 5	16 ± 24
2014	282 ± 139	134 ± 5	10 ± 14
2015	284 ± 144	130 ± 10	9 ± 16

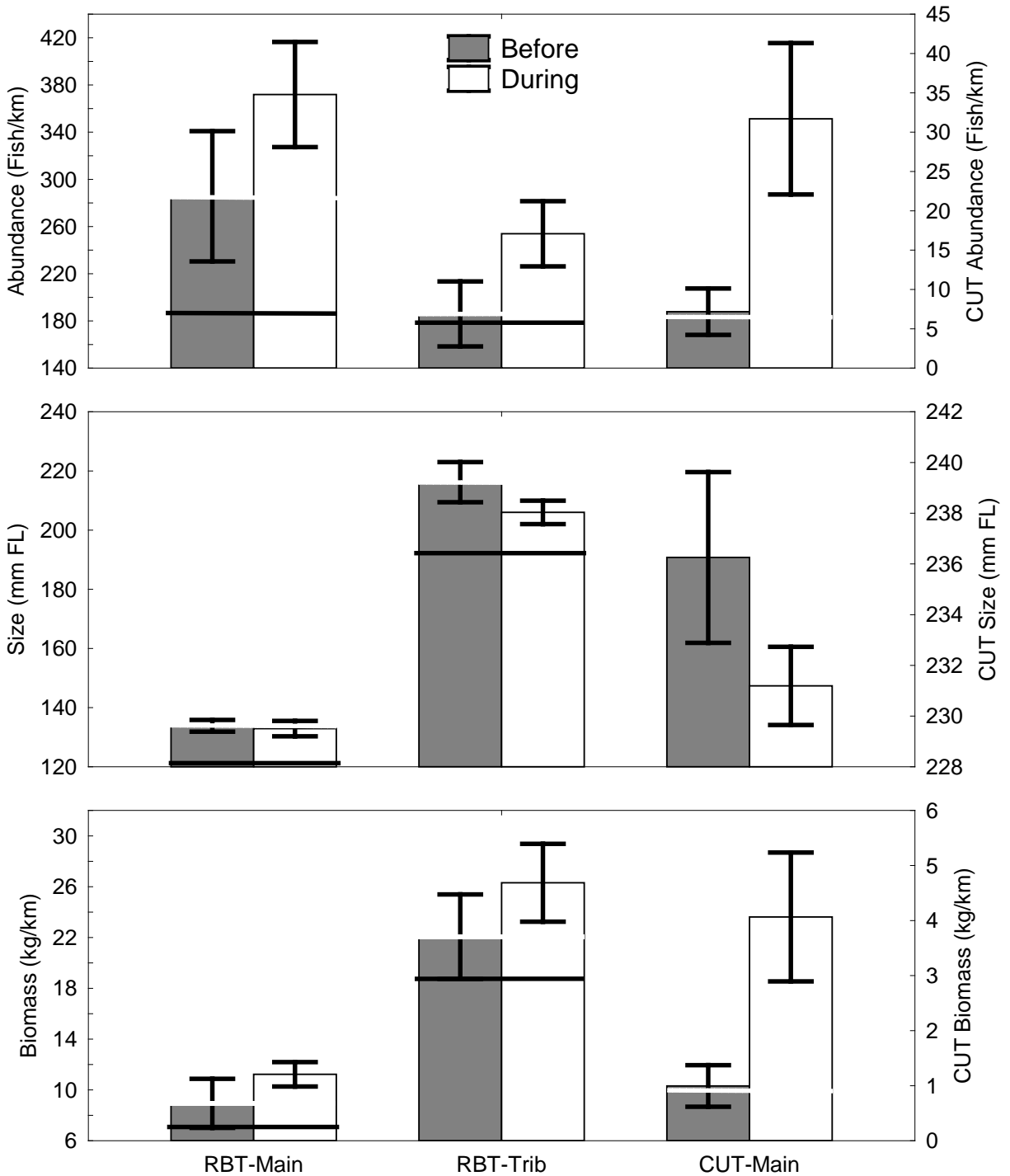


Figure 3. Abundance (n/km), size (FL mm), and biomass (kg/km) of tributary rainbow trout, main stem Yakima River rainbow trout (age 1) and cutthroat trout (<250 mm) before and during supplementation. Main stem cutthroat trout abundance, size, and biomass are associated with the right y-axis. The horizontal dashed line represents the 0% containment objective (CO) for steelhead in the main stem and tributaries, and the 10% CO for mainstem cutthroat trout. The solid horizontal line represents the 10% CO for main stem rainbow trout and 40% CO for tributary rainbow trout. Error bars represent 90% confidence intervals.

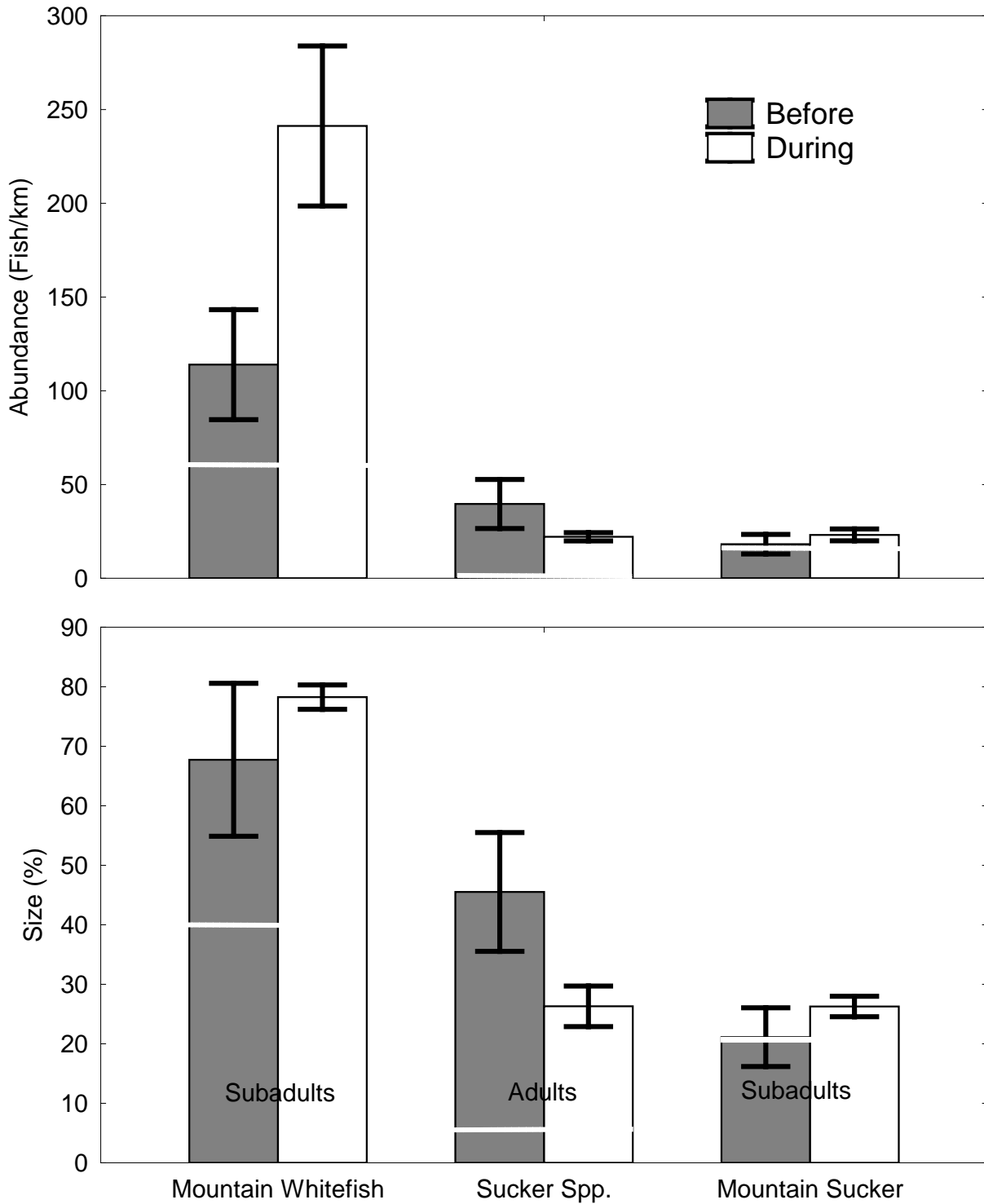


Figure 4. Abundance (fish/km) and size (percent by size class) of mountain whitefish, suckers, and mountain suckers before and during supplementation. Error bars represent the 90% confidence interval. Dashed lines represent the 40% containment objectives for mountain whitefish, 90% for sucker species (Spp), and 5% for mountain suckers.

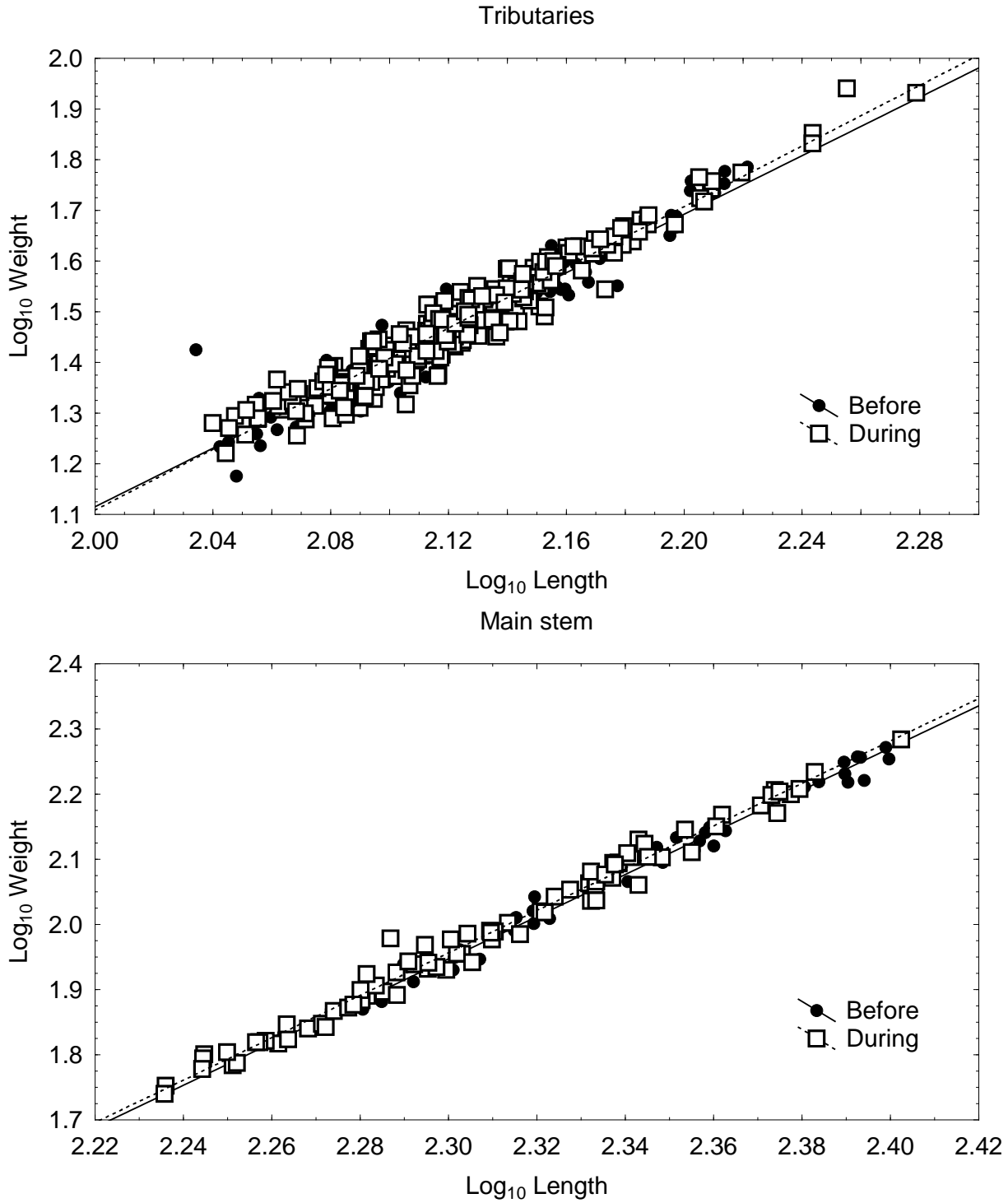


Figure 5. Mean length-weight relationships of tributary and age 1 main stem Yakima River rainbow trout before (1990-1998) and during (1999-2015) the supplementation period. Each data point represents the mean from a sample site.

Causation

Since the lower 90% confidence limit for our steelhead size index was exceeded in both the Yakima River main stem (age 1 rainbow trout) and Yakima Basin tributaries (all ages of rainbow trout), we tested if the changes could be reasonably attributed to supplementation. We did not detect a statistically significant decrease in our steelhead size index (age 1 rainbow trout; BACIP; $P = 0.81$) or our steelhead biomass index (age 1 rainbow trout; BACIP; $P = 0.48$) in the main stem downstream from the Clark Flats acclimation facility. Interestingly, we did not detect a significant relationship between our steelhead abundance and size index relationship ($R^2 = 0.10$; $P = 0.12$) suggesting density dependence was probably not influencing our steelhead size index. For tributary comparisons, we did not detect significant differences in our steelhead size index in comparisons between the North Fork Teanaway River downstream from the Jack Creek acclimation facility (treatment sites) and the West (BACIP; $P=0.08$) and Middle Fork (BACIP; $P=0.49$) Teanaway River reference sites. Additional comparisons of our steelhead size index in the main stem Teanaway River relative to the West and Middle Fork Teanaway River reference sites were not consistent with an impact (i.e. all changes were positive). Thus, at this time, the weight-of-evidence suggests declines in our steelhead size index are not likely the result of salmon supplementation activities in the basin.

Although the before vs. after comparisons of rainbow trout abundance did not indicate declines warranting a refined analysis of abundance, we erred on the side of caution and conducted the analysis given our concerns related to the depressed steelhead size index. A comparison of rainbow trout abundance in index monitoring sites located downstream from the Jack Creek Acclimation Facility (e.g., North Fork and Main stem Teanaway Rivers) relative to reference sites in the Middle and West Fork Teanaway Rivers revealed significant reductions in the abundance of rainbow trout relative to the control streams (BACIP) in some years. We attempted to account for factors that may influence abundance such as movement and angler induced mortality. Motion activated cameras mounted in both treatment and reference sites during the open angling season in 2011 indicated that the reduction in abundance was probably not angler induced. In addition, we have not detected large scale movements of tagged rainbow trout between treatment and reference streams that would be consistent with a largescale displacement of trout. However, we do have evidence that the North Fork of the Teanaway River produces a high proportion of anadromous steelhead smolt migrants and significant migrant production may contribute to reduced resident trout abundance. We will continue this investigation in the coming year in cooperation with the Yakima Steelhead VSP project.

Discussion/Conclusion

We failed to reject the hypothesis that early-middle stages of salmon supplementation have impacted valued trout species in the upper Yakima Basin beyond predetermined containment objectives. There were no impacts of supplementation activities on bull and cutthroat trout that inhabited tributary streams because limited or no overlap with hatchery or naturally produced salmon occurred. However, the potential existed for much overlap between salmon and bull and cutthroat trout in the tributaries of the upper Yakima Basin. For example, hatchery steelhead that were released in 1994 very close to the release site in the North Fork of the Teanaway River, migrated upstream into areas containing bull trout and cutthroat trout (McMichael and Pearsons 2001). Hatchery spring Chinook also migrated upstream of the

acclimation site in the North Fork of the Teanaway River, but not nearly as far as hatchery steelhead. This finding is consistent with our earlier work and extends the findings into later stages of supplementation (Pearsons and Temple 2007).

It is possible that some overlap occurred at times and places when/where we did not sample. However, substantial overlap was unlikely because we sampled at times and places that overlap was most likely. There are certainly areas outside the upper Yakima watershed where overlap occurs at the times that we sampled. Furthermore, overlap has been detected using the methods we used (e.g. snorkeling). Salmon and bull and cutthroat trout overlap during the summer in another large tributary in the Yakima Basin that parallels the upper Yakima River. In the Naches Basin, which merges with the upper Yakima River near the city of Yakima, substantial overlap exists between bull and cutthroat trout and naturally produced Chinook salmon (T. Pearsons, unpublished data). Hatchery coho salmon are released into that basin and undoubtedly overlap with bull and cutthroat trout. Other studies have also documented overlap between salmon and cutthroat and bull/Dolly Varden trout (Glova 1984; Bisson et al. 1988; Nakano and Kaeriyama 1995; Thurow et al. 1997).

There are a variety of possible reasons why overlap was not detected in tributaries of the upper Yakima River. First, all but one of the acclimation sites for salmon were located in the main stem and the acclimation site in the tributary was located downstream of bull and cutthroat trout. Risks to bull and cutthroat trout were one of many factors that contributed to acclimation site placement. Second, the distribution of juvenile salmon has not increased substantially even though the abundance of adult salmon has increased. We had expected that the distribution of juvenile salmon would have increased with increasing abundance of spawners. Third, high abundance of rainbow trout in lower elevation portions of tributaries may competitively exclude cutthroat and bull trout to higher elevations that salmon do not occupy. Relaxation of competition could result in broader distributions of bull and cutthroat trout and the possibility of greater overlap with salmon. Fourth, salmon, bull trout, and cutthroat trout have different habitat preferences. Salmon typically occupy streams of lower gradient, lower elevation, and warmer water temperatures than cutthroat and bull trout (Glova 1987; Dunham and Rieman 1999). Glova (1987) concluded that impacts to cutthroat trout could be reduced by stocking coho in areas with gradients greater than 1% and ample fast water habitats. Faster water velocities allow for more resource partitioning and competitive dominance by trout. Most of the tributaries in the upper Yakima Basin met these criteria. We did observe overlap between salmon and cutthroat trout in the main stem, where water temperatures were more suitable for both of these species.

Contrary to our previous findings (Pearsons and Temple 2007), we did detect a significant difference in the abundance of rainbow trout in treatment areas in the North Fork Teanaway River and main stem Teanaway River relative to our control sites in some years (Pearsons and Temple 2010). Inherent variation associated with fish population abundance influences our statistical power to detect significant results during some periods. With each additional year of sampling we will have increased power to detect smaller differences (Ham and Pearsons 2000). However, it is important to note that our “Before-After” detection plan would not have triggered the “Causation” analysis that was used to detect the decline and the decline was isolated to a small area and was small relative to the total population size. Furthermore, we do not yet have evidence to support the decline was due to mortality of fish in the treatment area. Other possibilities may include displacement, and perhaps angler harvest.

Although we observed decreases in the size of rainbow trout during the post-supplementation period, the decline is unlikely to have been caused by supplementation. If

supplementation had changed the size structure or growth of the steelhead size index, we would expect to detect this change in areas with high densities of salmon. We did not detect a reduction in the size of rainbow trout in the high-density areas of the target taxa below the Clark Flats acclimation site or below the release site in the North Fork Teanaway River. These areas are likely to have the greatest potential of detecting an impact. One potential explanation for the observed decrease in main stem rainbow trout size is that intraspecific density dependent mechanisms have altered the size of main stem Yakima River rainbow trout. The abundance of rainbow trout increased by approximately 30% (30% increase of age 1 fish, and 29% increase of fish greater than 249 mm) after stocking began. This information and results from small-scale enclosure experiments (McMichael et al. 1997) leads us to believe that the decline in rainbow trout lengths is most likely the result of intraspecific competition.

With the exception of the BACIP results from the Teanaway basin in some years, the lack of detectable impacts to rainbow trout were consistent with results that were derived from smaller scale enclosure experiments between naturally produced spring Chinook salmon and rainbow trout in high elevation tributaries (McMichael and Pearsons 1998). In these experiments, growth and abundance of rainbow trout were not impacted when the density of salmonids was doubled by the addition of naturally produced spring Chinook salmon parr. However, growth of rainbow trout was suppressed when the density was doubled with rainbow trout (McMichael et al. 1997), which supports the previously mentioned idea of intraspecific impacts to rainbow trout growth in the main stem. The current results extend the findings of McMichael and Pearsons (1998) to smolts, residuals, coho salmon, and to lower elevation waters such as the main stem. Our ability to detect impacts with the BACI design and the longer experimental period in this study (higher statistical power) may explain the differences among the studies. Opportunities for cumulative impacts to manifest and larger sample sizes may be necessary to detect impacts where high natural variation occurs.

It is possible that our abundance estimates in the main stem and tributaries and the size estimates in the tributaries were influenced by the size breaks that we used in our analysis. The lower size breaks were necessary (e.g., 80 mm in tributaries and 100 mm in the main stem), because we capture very few of these fish due to our low electrofishing efficiencies on small fish and hence cannot calculate valid estimates on these fish. This could result in varying proportions of age 0 and 1 fish in our estimate if the length at age varied across years or sites. However, we do not believe that length truncations significantly affected our conclusions. For example, if fish length was negatively impacted then the distribution of fish size would have become smaller, and more age 1 fish could have been pushed below 100 mm. Regardless of how many fish may have been shifted below 100 mm, if the impact occurred to the whole age class then we should have detected a decrease in size for fish above 100 mm (e.g., the whole length frequency curve would be shifted to smaller sizes). Similarly, if many fish were impacted so that they were less than 100 mm then the abundance of age 1 fish would have been negatively biased. In other words, we would expect to detect less fish than we did prior to supplementation. If we had concluded that impacts had occurred, then our length truncations would be a more serious issue.

We did not detect impacts to non-trout NTT that could be attributed to supplementation. In the tributaries, this was because none of the non-trout NTT overlapped salmon at high enough levels to exceed the CO. All non-trout NTT in the main-stem overlapped completely, but none exceeded the containment objectives.

With the exception of minimum daily stream discharge in the main stem Yakima River, we did not detect changes in the environmental variables that were measured. We hypothesize

the increased minimum daily stream discharge observed would benefit NTT. However, the increased minimum daily discharge was not significantly correlated with our NTT monitoring variables suggesting that it did not confound our results. Average and maximum stream discharge and temperature were heavily regulated by upstream irrigation reservoirs providing a relatively stable environment to conduct risk containment monitoring. Although discharge in tributaries is unregulated, summer baseflows have not differed drastically during the time of sampling from year to year. The relatively stable environmental conditions observed in both tributary and main stem areas supports the use of time as a control in our evaluation.

Adaptive Management & Lessons Learned

There are a number of management decisions that likely contributed to the small number of impacts in our study. For example, rigorous pre-implementation planning likely prevented many undesirable impacts to NTT (Temple and Pearsons 2012). Acclimation site placement and the release of fish at the smolt stage reduced the potential spatial and temporal overlap with NTT. Implementation of strategies to limit the number of precociously mature male salmon entering the natural environment would further decrease the risk of failing to meet containment objectives in the future. By reducing the number of these precociously mature salmon, both direct and indirect undesirable interactions with NTT will be reduced. Strategies to reduce the production and release of precociously mature salmon have been implemented in the past (Larsen et al. 2006).

We are implementing the approach described by Ham and Pearsons (2001) to contain risks to NTT throughout the life span of salmon supplementation programs in the Yakima Basin (Pearsons 2002). According to this risk containment approach, if we detect a decrease in NTT status that is greater than a containment objective, then we attempt to determine if the decrease was caused by the supplementation program. Only changes that are due to supplementation warrant risk containment action specific to the supplementation program. The only NTT that was outside of its containment objective was our steelhead index. Steelhead trout were monitored using age 1 rainbow trout as an analog of juvenile steelhead in the main stem and all rainbow trout in the tributaries as analogs. The declines in mean steelhead trout size and the surpassing of the lower 90% CL of the containment objective for steelhead were unlikely to be due to supplementation and therefore do not require risk containment actions. However, the isolated decline in rainbow trout abundance in the North Fork Teanaway in some years is cause for concern because it exceeds the CO for steelhead in this locality. It is important to note, however, the observed decline did not persist and rainbow trout abundance in all other tributaries monitored generally increased during supplementation (e.g., population level increase). Finally, we are now monitoring the response of NTT during the building stage of supplementation and this stage is likely to be the one where the risk of impacts is the highest because the number of salmon is potentially very high due to a combination of hatchery and naturally produced fish (Pearsons 2002).

We recommend that our results be placed into context of supplementation dynamics and the ecological context of the Yakima Basin. Our evaluation addressed impacts that occur during the early-middle stages of supplementation. These stages correspond to the Broodstock stage and the Building stage described by Pearsons (2002). The Building stage provides the greatest opportunity for interactions between hatchery fish, naturally produced offspring of hatchery fish, and NTT. Large numbers of hatchery smolts, residuals, and the offspring from returning

hatchery adults increases interaction potentials between hatchery and wild fish in the freshwater migration corridor and freshwater rearing area. The Yakima main stem is a highly altered system, and this alteration could change the outcomes of ecological interactions. For example, the flows in the upper Yakima main stem are heavily regulated by the United States Bureau of Reclamation's water storage and delivery programs and are highly unnatural. Artificially high flows during the summer might decrease interspecific interactions among species because of the large volume of water and ability for species to partition resources. In contrast, flows in the tributaries are more normative and might be more reflective of tributaries in other regions. Application of our results should be tempered within these important contexts.

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Appendices

A.1: Data sets or products:

Summarized data associated with this project are provided in tabular format in annual reports available at www.cbfish.org.

A.2: Publications

Include citations for all publications resulting from the study and work described in this report.

- Ham, K. D., and T. N. Pearsons. 2000. Can reduced salmonid population abundance be detected in time to limit management impacts? *Canadian Journal of Fisheries and Aquatic*

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Appendix 3

Abundance and Distribution of Hatchery and Natural Origin Precociously Mature Male Spring Chinook Salmon in the Yakima River

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Abstract

Artificial propagation of Chinook salmon (*Oncorhynchus tshawytscha*) has the potential to alter the abundance and distribution of males that precociously mature in freshwater and thereby influence ecological and genetic interactions in the natural environment. We investigated the abundance and distribution of precociously mature, hatchery and natural origin male spring Chinook salmon during the spawning season (e.g., four to seven months after release from the hatchery) in the Yakima River. We counted the number of precocious males on the spawning grounds while snorkeling during the peak of spawning and electro-fished to determine abundance and distribution of precocious males away from redds. The abundance of hatchery origin precocious males on redds was less than a fraction of a percent the number of fish released, and were less abundant on redds than natural origin precocious males ($P < 0.01$). Between 1999 and 2014, the mean abundance of hatchery age 1 precocious males estimated on the spawning grounds was 20 fish and ranged between 0 and 78 fish annually. Hatchery and natural origin precocious males were both found throughout the spawning range during the spawning season, although differences in distribution between origins were detected ($P < 0.05$). Hatchery precocious males were proportionately more abundant in the lowest sampling reach and less abundant in a tributary with no hatchery facilities in most years. In addition, most hatchery precocious males were found downstream of spawning areas during the spawning season. It appears that many precociously mature fish released from the hatchery migrate downstream and fail to migrate back to the spawning grounds or die within the Yakima River prior to spawning time. Thus, hatchery precocious males in the Yakima River do not contribute favorably to harvest and may pose ecological risks to non-target taxa, but have a low probability of contributing genes to future generations.

Introduction

Artificial propagation of Chinook salmon (*Oncorhynchus tshawytscha*) has the potential to alter the age that fish mature and result in undesirable interactions with natural origin fish (Knudsen et al. 2006). This is a particular concern for conservation hatcheries where the goal is to increase natural production while maintaining the characteristics of the natural population (Mobrand et al. 2005). Although most Chinook salmon are anadromous (Healey 1991), some salmon complete their entire life cycle in freshwater, even when they have access to the ocean. These salmon are generally small, male, precociously mature, short-lived and are referred to as residents, precocious males, or minijacks (Gebhards 1960; Mullan et al. 1992; Zimmerman et al. 2003). Although the incidence of precocious male maturation in Chinook salmon has been recognized since at least 1897 (Rutter 1902), the importance and management of this life history has only recently become appreciated as population sizes are diminished and hatcheries are used for conservation. Resident life histories have been observed in both natural (Flain 1970; Gebhards 1960; Mullan et al. 1992) and hatchery produced fish (Robertson 1957; Mullan et al. 1992; Larsen et al. 2004a) and can occur as subyearling (Robertson 1957; Gebhards 1960; Mullan et al. 1992), yearling (Gebhards 1960; Mullan et al. 1992; Larsen et al. 2004a), and

perhaps under favorable conditions even older ages (Mullan et al. 1992; Unwin et al. 1999). Estimates of precocious maturation have been reported between 0 and 93% (Gebhardt 1960; Taylor 1989; Mullan et al. 1992; Foote et al. 1991; Shearer et al. 2006) although the high end of this range is from studies that have been performed in hatchery environments. It is less clear what the range of precocious maturation is in natural environments, but it is likely to be lower than the high ends of the range presented.

The occurrence of precocity in salmon has been credited to genetic factors and environmental and physiological cues (Thorpe 1987; Bohlin et al. 1990; Foote et al. 1991). Age-at-maturation has been shown to be heritable in salmon (Heath et al. 1994; Unwin et al. 1999). Furthermore, Foote et al. (1991) concluded that variation in precocious maturation rates observed in Canada (Taylor 1989; Foote et al. 1991) were due to genetics. The higher incidences of precocity they observed were from populations farthest inland and the growth rates of fish were similar. Hatcheries may enhance precocious maturation of males by the dietary composition of the feed (Shearer and Swanson 2000) or the types of growth schedules that fish are placed on (Silverstein et al. 1998; Larsen et al. 2004, 2006). Precocious maturation is common in other anadromous species and some precocious individuals may enhance population viability particularly when population sizes are low or environmental disturbances are frequent (Myers and Hutchings 1987; Foote and Larkin 1988; Pearsons et al. 2007a). When the spawning population size is small and there is genetic diversity among age classes, precocious males may contribute to genetic diversity and increase the probability that eggs are fertilized. The sperm of precocious male salmon has been shown to successfully fertilize eggs (Rutter 1902; Robertson 1957; Unwin et al. 1999) and some precocious males may be able to spawn in multiple years (Bernier et al. 1993). It has been known for some time that hatcheries can produce large numbers of precocious Chinook salmon (Robertson 1957; Mullan et al. 1992; Larsen et al. 2004a; Beckman and Larsen 2005), but there have been relatively few studies that have investigated the abundance and distribution of these fish in rivers during the spawning season.

Artificially high numbers of precocious males that are released into rivers may harm wild populations through ecological and genetic mechanisms (Pearsons and Hopley 1999; Pearsons 2002; Pearsons and Temple 2007). Hatchery precocious males may eat wild fish, compete for resources, and spread disease (Pearsons et al. 2007b; 2009). They may also breed with wild fish and consequently lower the fitness of offspring because of the domesticating effects of artificial propagation (Schroder et al. 2006; Blankenship 2007). Hatchery fish may pass on genes that are unfavorable in the natural environment and may be particularly pronounced in precocious males because they do not experience the selective pressures that migratory adults experience and they may be the result of hatchery selection (Garant et al. 2003). In addition to potential impacts to wild fish, resident Chinook salmon are undesirable to fisheries because they are generally too small for harvest and can interfere with fisheries on other species by being caught more easily than targeted species such as resident trout. In short, production of artificially high numbers of precocious males has the potential to limit the success of supplementation programs. In contrast, artificial reduction in their production may negatively impact the population by reducing traits that are associated with precocious maturation such as fast growth.

The primary objective of this study was to evaluate the abundance and distribution of hatchery spring Chinook salmon precocious males during the spawning season in the Yakima River. The goal of the hatchery program in the Yakima River is to increase natural production and to provide harvest opportunity while keeping adverse genetic and ecological impacts within specified biological limits (Bonneville Power Administration 1996; Fast and Craig 1997; Bosch

2004). As such, the program strives to minimize differences between the hatchery and natural spawning fish to decrease the risk of long-term fitness impacts. Previous research indicated that the Yakima Supplementation and Research Facility has produced and released an average of 129,249 precocious males/year into the upper Yakima basin between 1999 and 2008 (Larsen et al. 2004a; Larsen et al. 2008; Yakima/Klickitat Fisheries Project, Unpublished data). The estimate of precocious male abundance was derived by multiplying the percent of fish that exhibited physiological indicators of precocious maturation by the total numbers of fish released. It was not clear whether the spawning grounds would be swamped with large numbers of precocious males or whether these fish would die or fail to migrate to the spawning grounds prior to the spawning season. Our goals were to 1) estimate the abundance of hatchery origin precocious males on the spawning grounds, and 2) quantify the distribution of hatchery precocious males on and away from the spawning grounds. We also present information about the abundance and distribution of natural origin precocious males so that we can determine how hatchery precocious males might differ.

Methods

Study Area and Hatchery Program

The Yakima River Basin is a large river system that drains into the Columbia River near Richland, Washington. The upper Yakima River basin, which is the subject of this paper, is located upstream of Roza Dam (Figure 1). Historically large numbers of salmon returned to the upper Yakima basin (Bonneville Power Administration 1996). The flows in the upper main stem are regulated by three dams located in the upper portion of the basin that are complete or partial barriers to upstream migration. These dams store water and release water when it is needed for agriculture irrigation. Peak flows during the spring have been truncated and flows are artificially high during the summer and then dramatically reduced to a constant level at the onset of spawning in September (Pearsons et al. 2007b). Chinook salmon spawning generally peaks during the latter half of September. The management of water in the upper Yakima causes the annual differences in the hydrograph to be reduced.

Artificial propagation of spring Chinook salmon in the upper Yakima Basin began with brood stock collection in 1997 and yearling smolt releases in 1999 as part of the Yakima/Klickitat Fisheries Project (YKFP). Approximately 689,458 Chinook salmon smolts have been released annually in the upper Yakima River from 1999 to 2010 (YKFP unpublished data). Broodstock for the spring Chinook program were natural origin upper Yakima stock collected at a trapping facility at Roza Dam. Chinook were spawned and juveniles were reared at the Cle Elum Supplementation and Research Facility (CESRF) in Cle Elum, Washington (Larsen et al. 2004a; Knudsen et al. 2006; Pearsons et al. 2007c). Yearlings were transported to one of three acclimation sites during January and February. Easton and Clark Flat acclimation sites are located on the Yakima River and the Jack Creek acclimation site is on the North Fork of the Teanaway River (Figure 1). The Easton and Jack Creek acclimation sites are located at the upper end of the Chinook spawning distribution in the Yakima and Teanaway Rivers, and the Clark Flats acclimation site is located near the lower end of the annual spawning distribution in the Yakima River. Prior to release, Chinook in each acclimation site were sampled to determine

the rate of precocious maturation (Larsen et al. 2004a, 2008). Hatchery spring Chinook were allowed to voluntarily migrate out of the acclimation sites between March 15th and May 31st. Chinook were approximately 120 mm fork length (FL) when released. Fish that remained in the acclimation sites were forced out in mid to late May.

Due to a high rate of precocious male maturation in the hatchery, a production scale growth modulation program was established in 2002 (release year 2004) to evaluate the efficacy of reducing the incidence of precocious male maturation (Larsen et al 2006). Half of all hatchery-reared Chinook in brood years (BY) 2002-2004 were reared under low growth conditions and half were reared using normal hatchery protocol. Larsen et al. (2004b) reported 29% of the low growth reared males and 43% of the normally reared males precociously matured; a 33% reduction in precocious male maturation. However, preliminary estimates indicated that the low growth fish did not survive as well as the normally reared fish. Normal rearing practices were reestablished for all hatchery-reared Chinook in BY 2005.

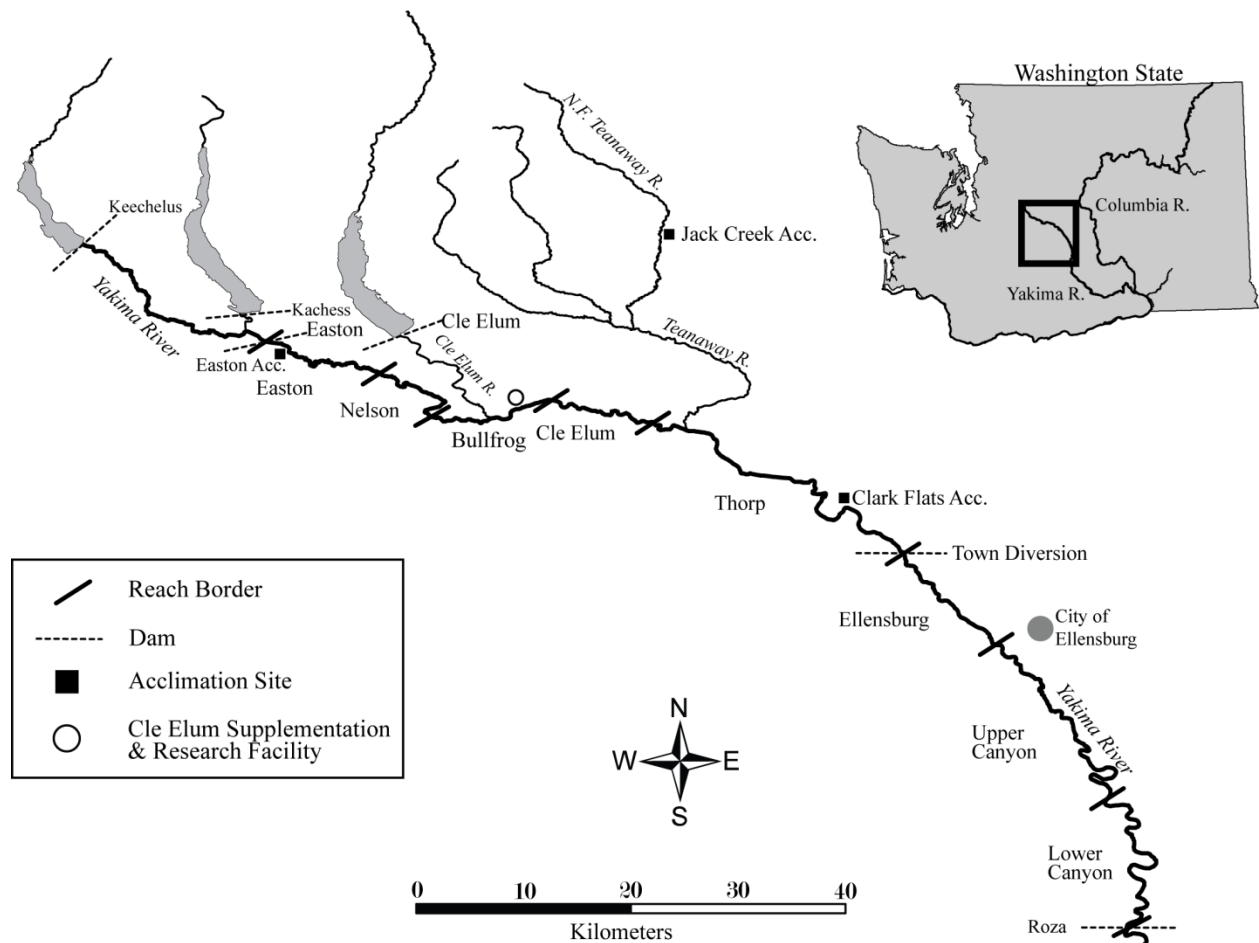


Figure 1. Survey reaches within the upper Yakima basin. Drift boat electrofishing survey reaches included: lower canyon, upper canyon, Ellensburg, Thorp, and the Cle Elum. Snorkeling

surveys were conducted in the Thorp and Cle Elum reaches as well as the Bullfrog, Nelson, Easton and the Cle Elum River.

Abundance and Distribution

Sampling of precocious males on the spawning grounds occurred throughout the main stem Yakima River between Easton Dam and Town Diversion Dam, and the Cle Elum River from Cle Elum Dam to the Yakima River confluence between 1999 and 2014 (Figure 1). These areas represent the vast majority of the spawning area in the upper Yakima River and generally account for over 99% of Chinook redds counted annually (YKFP, unpublished data). Annual counts of precocious males occurred during the peak of spawning, which generally occurred during the last half of September. The sampling area consisted of six reaches (Figure 1). These reaches, beginning at the downstream end at approximately river kilometer (rkm) 256 were: Thorp (26.6 rkm long), Cle Elum (9.1 rkm long), the Cle Elum River (12.2 rkm long), Bullfrog (10.2 rkm long), Nelson (7.3 rkm long), and Easton (10.7 rkm long). The Bullfrog reach was first sampled in 2003. Excluding the Bullfrog reach, all reaches together make up 87 percent (66 rkm) of the total spawning area above the Town Diversion Dam (Figure 1).

We counted the number of precocious males on active redds by snorkeling. Each reach was floated with an inflatable raft and spring Chinook redds were flagged and numbered. Upon reaching a redd we determined the presence or absence of anadromous salmon. We only snorkeled those redds with anadromous fish on the redds and termed them ‘active redds’. Previous work demonstrated that precocious males were rarely observed on redds without anadromous fish present (Gebhards 1960; James et al. 1999). A snorkeler would then begin 5-10 meters downstream of the redd and snorkel upstream, counting all spring Chinook encountered. Fish were categorized as either being on the redd (in the bowl), or associated with the redd (within 5 meters). We combined the abundance of fish in these two categories in our analysis, however, most of the fish we counted were on redds. Hatchery origin residuals (e.g., non-migrants) were differentiated from natural origin spring Chinook by the presence of an adipose clip. During years when more active redds were present than we could snorkel, we systematically sampled the active redds (e.g., every other redd).

An estimate of the total number of precocious males by age class and origin in the spawning areas of the upper Yakima Basin was derived by summing the abundance estimates of each of the six reaches. Abundance estimates in the reaches that were surveyed were calculated by multiplying mean abundance of precocious males per active redd by the number of active redds. We extrapolated the number of precocious males per river kilometer of adjacent sampled reaches to the length of reaches that were not sampled. Extrapolations of counts from both the Cle Elum and Nelson reaches were applied to the Bullfrog section in years prior to 2003.

We also estimated the number of hatchery precocious males that were not on redds during the spawning season. Five sections of the upper Yakima River were sampled at night from the middle of September to the middle of October using a drift boat electrofisher as described by Temple and Pearsons (2007). These sections represent approximately 29% of the area between the Cle Elum River and Roza Dam. The electrofisher was turned off when we approached redds to avoid electro-shocking fish on or near redds. The numbers of hatchery precocious male Chinook netted during the electrofishing marking runs were expanded by maximum log-likelihood model recapture efficiencies for similar sized rainbow trout observed (177-203 mm). In sites where we could generate capture efficiencies for hatchery precocious

male Chinook (e.g., mark-recapture), our observed rainbow trout electrofishing capture efficiency was within the 95% confidence interval (CI) of the hatchery origin spring Chinook electrofishing efficiency. Thus, we believe that size based efficiencies are reasonable ways of indexing abundance because fish size is one of the most important factors that influences electrofishing efficiency (Anderson 1995; Buttiker 1992).

Hatchery line

Previous work determined that the majority of residual males are precociously mature (Pearsons et al. 2008); therefore we consider all hatchery residuals encountered in the system as precociously mature males.

Analysis

An ANOVA was used to compare the estimated annual abundance of precocious males by age class and origin. If test results were significant at a 0.05 level, Tukey post-hoc comparisons were made to determine which comparisons were significant. Comparisons of spatial distributions on the spawning grounds were evaluated using contingency table G-tests of independence. Pearson product moment correlation statistics were used to explore various relationships of precocious male abundance. Non-parametric tests were used when data did not meet parametric test assumptions and could not be adequately transformed. Statistical analyses were performed in the R statistical package, version 3.0.2 (R Core Team 2013), or in PopTools version 3.0.6 (Hood 2008) with an alpha (α) = 0.05.

Results

The estimated number of natural origin age 0, natural origin age 1, and hatchery precocious males on the spawning grounds during the peak of spawning ranged from 5 to 718, 0 to 92, and 0 to 78 between 1999 and 2014 respectively (Table 1). Differences in the number of observed precocious males on or associated with active redds were detectable among age classes, and origin. Differences in the mean abundance of precociously mature males of different age and origin were detectable among years (ANOVA: $F_{2,45} = 14.0$, $P < 0.01$). Post-hoc analysis determined that natural production age 0 precocious males were greater in abundance than both natural and hatchery production age-1 males (Tukey test: $P < 0.01$). There were no detectable differences in abundance between age 1 natural and hatchery production precocious males (Tukey test: $P = 0.96$). Among years, age 0 precocious males were found on a greater proportion of redds sampled than either age 1 or hatchery origin (ANOVA: $F_{2,45} = 13.2$ $P < 0.01$; Tukey test: $P < 0.01$), and were greater in number per active redd (ANOVA: $F_{2,45} = 19.1$, $P < 0.01$; Tukey test: $P < 0.01$), (Table 2.).

Table 1. Number of observed and estimated totals of natural (age 0 and age 1) and hatchery origin precocious males by age class at the peak of spawning activity in the upper Yakima River. Estimated totals are extrapolations over redds and/or portions of reaches not sampled.

Survey year	Active redds	(% Redds surveyed	(% Spawning area sampled	Observed			Estimated total		
				Age 0	Age 1	Hatchery	Age 0	Age 1	Hatchery
1999	36	100	87	4	11	17	5	16	19
2000	316	66	87	103	42	8	128	42	11
2001	276	62	87	336	11	26	555	21	53
2002	304	81	87	138	15	8	228	25	14
2003	230	78	100	204	25	19	267	35	24
2004	1662	27	100	195	16	21	718	65	78
2005	655	99	100	357	17	0	360	17	0
2006	198	90	100	148	2	0	177	3	0
2007	92	100	100	55	0	0	55	0	0
2008	173	82	100	69	55	42	85	67	52
2009	105	99	100	87	15	34	88	15	34
2010	499	48	100	133	42	12	280	92	21
2011	418	73	100	124	40	0	171	55	0
2012	243	63	100	44	17	3	70	27	5
2013	166	66	100	76	10	3	115	15	5
2014	279	68	100	41	1	2	54	1	3

Table 2. Means of the presence and abundance of natural (age 0 and age 1) and hatchery origin precocious males per active redd at the peak of spawning activity in the upper Yakima River.

Survey year	Active redds	Presence/Active redd			Abundance/Active redd		
		Age 0	Age 1	Hatchery	Age 0	Age 1	Hatchery
1999	36	0.11	0.14	0.19	0.14	0.44	0.53
2000	316	0.18	0.10	0.02	0.41	0.13	0.03
2001	276	0.31	0.03	0.04	2.01	0.08	0.19
2002	304	0.23	0.03	0.03	0.75	0.08	0.05
2003	230	0.31	0.06	0.06	1.16	0.15	0.10
2004	1662	0.05	0.01	0.01	0.43	0.04	0.05
2005	655	0.24	0.02	0	0.55	0.03	0
2006	198	0.75	0.04	0	0.89	0.02	0
2007	92	0.18	0	0	0.60	0	0
2008	173	0.08	0.21	0.16	0.49	0.39	0.30
2009	105	0.24	0.09	0.13	0.84	0.14	0.33
2010	499	0.15	0.05	0.03	0.56	0.18	0.05
2011	418	0.24	0.07	0	0.41	0.13	0
2012	243	0.13	0.08	0.02	0.29	0.11	0.02
2013	166	0.20	0.07	0.04	0.69	0.09	0.03
2014	191	0.09	0.01	0.01	0.21	0.01	0.01

Hatchery precocious males were distributed differently than natural origin age 0, and natural origin age 0 and age 1 combined on the spawning grounds (G-test; $P < 0.02$). A significant difference was not detected between natural origin age 0 and natural origin age 1 fish (G-test; $P = 0.69$), or between natural origin age 1 and hatchery precocious males (G-test; $P = 0.25$; Figure 2). An average of 28 percent of all hatchery precocious males observed on the spawning grounds were in the lowest spawning reach examined, whereas only 7 percent of natural origin age 0, and 14 percent of natural origin precocious males were observed in this reach (Figure 2).

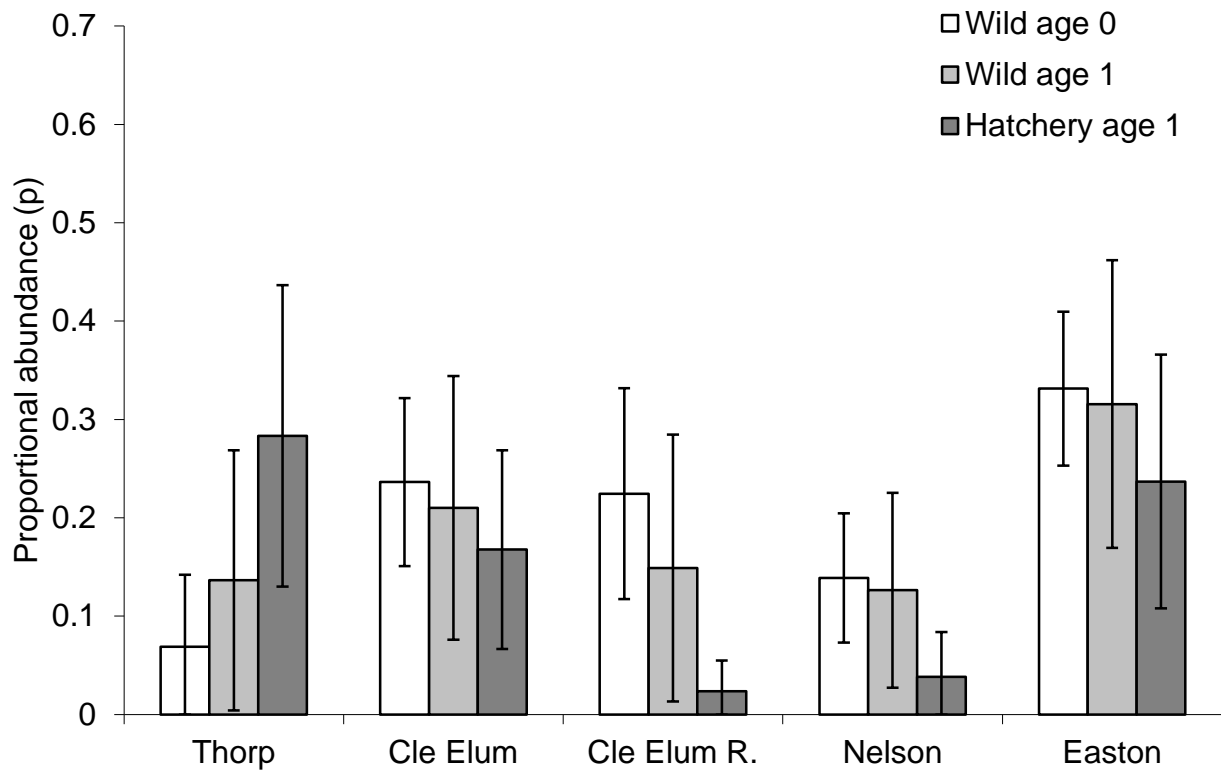


Figure 2. Mean proportion (p) of natural and hatchery origin precocious males by reach within the upper Yakima River at the peak of spawning activity 1999-2014. Error bars represent 95 percent confidence intervals with negative boundaries of zero.

Estimated total abundance of hatchery origin spring Chinook salmon away from redds at the time of spawning in 2014 ranged between 0 and 30 fish/km among sampling reaches (Table 3). The lower and upper Yakima Canyon averaged 59 percent of the estimated number of precocious males away from redds between 1999 and 2014, and 30 percent in 2014 (Figure 3). The annual abundance of hatchery precocious males away from redds was not detectably correlated with the number observed on redds ($P = 0.06$).

Table 3. Estimated abundance of hatchery origin spring Chinook salmon (HSPC) away from redds in the main stem Yakima River in the fall of 2014. The maximum number of fish netted (n) in one of two electrofishing surveys completed in consecutive weeks is presented (LCYN is the Lower Canyon, UCYN is the Upper Canyon, EBURG is Ellensburg, THORP is Thorp, and CELUM is Cle Elum). Capture probability was generated using rainbow trout of approximately the same size range as hatchery spring Chinook salmon.

Section	n	Capture prob.	Section est.	Section km	HSPC/km	Reach km	Total est.
LCYN	13	0.11	123	4.8	26	19.2	490
UCYN	29	0.18	158	5.2	30	13.4	407
EBURG	10	0.09	107	4.2	25	21.2	540
THORP	11	0.09	128	5.7	22	24.1	541
CELUM	0	0.09	0	7.4	0	16.2	0
Total	63	n/a	n/a	n/a	150	94.1	1978

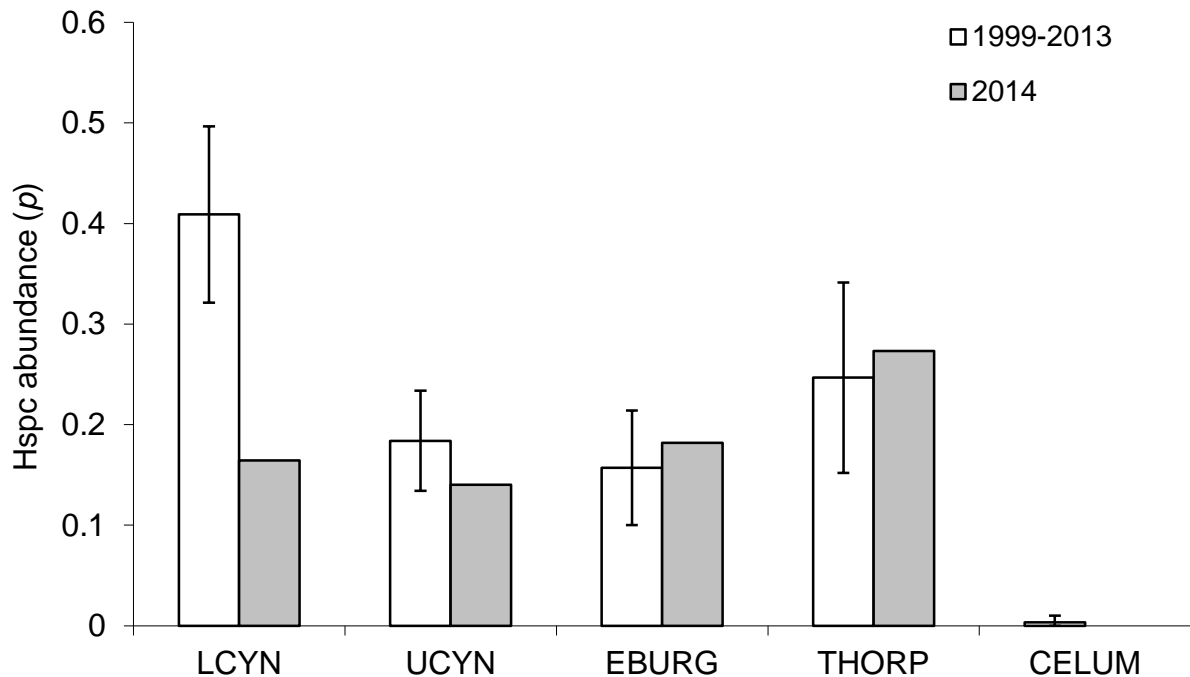


Figure 3. Proportional abundance (p) of hatchery spring Chinook sampled away from redds in the fall of 2014, and the mean proportional abundance between 1999 and 2013.

Discussion

Despite the large numbers of precocious males that are apparently released from the CESRF annually (Larsen et al. 2004a; Beckman and Larsen 2005; Larsen et al. 2006), only a small fraction of these fish appear to reach the spawning grounds. Hatchery precocious males may experience high mortality, migrate out of the study area after release, and/or fail to migrate back to the spawning grounds. Although the occurrence of some of these factors were observed in this or other studies (Larsen et al. 2004a; Beckman and Larsen 2005), we do not know the relative contribution of each of these factors towards the low abundance of precocious males on the spawning grounds.

Mortality of hatchery precocious males may be due to high angler exploitation, starvation, or predation. There is considerable angling pressure focused on trout in the Yakima River and anglers have at times commented on the number of precocious Chinook males caught, particularly in 2001. However, it is illegal to keep Chinook salmon in the upper Yakima River. Furthermore, studies have shown that hatchery origin fish released into the natural environment have lower survival than natural origin fish, presumably because of their inability to find food or avoid predators (White et al. 1995; Weber and Fausch 2003).

It has been documented that some hatchery precocious males move downstream out of the spawning areas and have been detected as far downstream as Bonneville Dam on the Columbia River (Larsen et al. 2004a; Beckman and Larsen 2005). In northern Oregon, precocious males were documented to have migrated at least 800 km and past three dams to reach salt water and return to the Umatilla River (Zimmerman et al. 2003). Hatchery precocious males were collected migrating both downstream in the spring and upstream during the summer (Larsen et al. 2004a; Beckman and Larsen 2005). The downstream migrations occurred during the smolt out-migration period and the upstream migrations occurred at the time of adult spawning immigration. If precocious males migrate downstream and then environmental conditions turn poor before they are able to migrate back upstream, then they are likely to die. The lower Yakima River becomes lethal for salmonids during many of the hot summer months when precocious males might attempt to ascend the river. If the factors contributing to hatchery fish mortality in the river are reduced or the conditions in the river are favorable for migration back to the spawning grounds (e.g., favorable flows and low angling pressure), then presumably the number of hatchery precocious males on the spawning grounds could increase dramatically. However, the range of conditions that we evaluated in this study, which included both high and low flow years, provide a reasonable range of what can be expected in the future.

Most of the hatchery precocious males that we encountered were located downstream of spawning areas. The lower and upper Yakima Canyon typically contain less than 1% of the upper Yakima Basin redds (Yakama Nation, unpublished data) and yet averaged 59% of the estimated number of hatchery precocious males during the spawning season. Many of the hatchery precocious males on the spawning grounds were observed in a reach that had relatively little spawning activity, whereas the natural origin precocious males were mainly in the areas with high spawning activity. The spawning area where many of the hatchery precocious males were observed was at the lower end of the spawning distribution. It also happens to be located

closest to the Yakima Canyon where the highest abundance of precocious males that were not on the spawning grounds was observed. In the Wenatchee River, very few hatchery precocious males were observed on the spawning grounds, but a considerable number were captured migrating upstream at a location downstream of the spawning areas (Murdoch et al. 2007). These fish may have also distributed themselves below the main spawning areas as we observed in the Yakima Watershed. This behavior is in contrast to natural origin precocious males that are rarely observed moving upstream past dams in the Yakima or Wenatchee watersheds, suggesting that natural origin precocious males have adopted a strategy of remaining on or near the spawning grounds and thus conserving energy and promoting growth and testes development. Some hypotheses as to why sexually mature hatchery precocious males, most of which are exuding milt at the time of sampling, are located in areas away from where most of the spawning activity occurs include: lack of energetic capacity to swim back upstream to the spawning grounds; inappropriate downstream migration behavior for their life-history strategy; late migration timing; and inability to locate areas with spawning females after they had migrated downstream of spawning areas. Younger salmon, such as precocious males and jacks, typically migrate back to the spawning grounds later than older salmon (Knudsen et al. 2006; Murdoch et al. 2007) and may migrate during unfavorable environmental conditions.

Cle Elum Hatchery origin fish are only released at age 1, which eliminates the possibility that age 0 hatchery precocious males will have the potential to spawn. In the absence of hatchery releases, age 0 precocious males are generally more abundant in the spawning areas than age 1 precocious males, so the hatchery is skewing the precocious male composition to an older age and larger size. This is in stark contrast to anadromous hatchery fish which typically mature earlier than wild fish and often at a smaller size-at-age (Knudsen et al. 2006). It is interesting to note that few incidences of precocious male maturation at age 0 have been observed in the Yakima hatchery (Larsen et al. 2004a). In addition, attempts to experimentally produce age 0 precocious males by high feeding rates in the hatchery did not produce any precocious males in 2002 (Farrell 2003). These fish emerged at the average emergence time of the population. It is possible that only the fish that emerge very early and experience good growth have the potential to precociously mature at age 0 (Larsen et al. 2007). However, because precocious males were not used in the broodstock, we cannot eliminate the possibility that genetics also influenced the absence of precocialism (e.g., Heath et al. 1994; Unwin et al. 1999).

Hatchery age 1 fish may be competitively superior to wild precocious males because hatchery precocious males are larger. Larger salmonids typically dominate smaller ones in behavioral contests (McMichael et al. 1999). We have observed a number of instances where hatchery precocious males displaced wild precocious males from redds or from preferred locations on redds. Behavioral dominance is important because dominant fish are more likely to be close to spawning females and hence more able to fertilize eggs (Garant et al. 2003). Dominant fish are better able to choose which locations pose the best chance for spawning success. Our behavioral observations suggest that per capita fertilization rates of hatchery precocious males should be higher than that of wild precocious males. However, sneaking strategies of smaller individual may also be successful.

We have identified some issues that could potentially contribute to the underestimation of precocious male numbers during our peak snorkel counts. We may have underestimated the number of active redds by spooking adults or by floating at times when adults are temporarily away from their redds. However, we rarely observed precocious males on redds without adults being present and this finding was also supported by work in the Salmon River drainage

(Gebhards 1960). Gebhards (1960) concluded that precocious males were generally only found in areas where there was spawning activity and were usually found in the bowl of the redd, and “the yearling males remained constantly within the redd.”

Other reasons include the possibility that precocious males may have been hiding away from the redds, were scared off the redds, were moving between redds, or were present in greater numbers before or after our peak count. Additional snorkeling efforts along the banks in 1998 and 2007 did not find hatchery precocious males in hiding areas such as undercut bank in the vicinity of spawning areas, and multiple reach surveys conducted in 2007 and 2008 did not suggest greater numbers of precocious males on the spawning grounds the week before or after our peak of spawning surveys. We have also observed that repeated counts of precocious males at three different times of the day in the same reach were similar. This suggests that either our counts were accurate or that our bias was consistent. However, our estimates of fish away from redds, that were generated from electrofishing were higher in some years than those generated from snorkeling in the Thorp reach, suggesting that snorkeling may underestimate abundance. In short, if we underestimated the number of precocious males on the spawning grounds then our numbers should be treated as indices.

Our study suggests that hatchery precocious males are unlikely to contribute a high proportion of genes in the Yakima Watershed when the number of anadromous adult returns is high, but contributions could be high when anadromous adult numbers are low. The highest abundance of hatchery precocious males that we estimated on the spawning grounds during any year was 78. This is a small proportion of the spawners when anadromous spawners number in the thousands, but relatively large when the abundance of spawners is in the hundreds. This range of anadromous fish abundance has been observed in the upper Yakima Watershed. In a separate DNA pedigree study conducted in an artificial spawning channel (Schroder et al. 2006), hatchery and natural origin precocious males of the upper Yakima spring Chinook salmon stock have been documented to sire offspring. In addition, precocious maturation appears to be highly heritable in Yakima spring Chinook salmon (Pearsons et al. 2007d). In short, it appears that the genetic contribution of hatchery precocious males on the spawning grounds is related to anadromous fish abundance and those factors that influence the abundance of precocious males on the spawning grounds. Variation in the precocious male contribution suggests that domestication risks may vary among years.

The consequences of eliminating a large proportion of hatchery males from the spawning population may be quite important, but currently the consequences of this unintended outcome are uncertain. Reduction of hatchery precocious males may be beneficial to the natural spawning population because of the potential reduction in domesticating effects that might be caused by hatchery rearing (Garant et al. 2003). Alternatively, reducing the genetic contribution of fish that have traits related to precocious maturation, such as fast growth, may impact the population in negative ways. Indeed, we found that age 0 precocious males were larger than immature fish (Pearsons et al. 2007b; Pearsons et al. 2009). Further research is needed to understand the importance of precocious males to natural spawning populations and the potential domesticating impacts of hatchery precocious males to natural populations.

Releasing large numbers of precocious males could also increase ecological risks to target and non-target taxa (e.g., McMichael et al. 1999). Any impacts that may have occurred within the first five years of hatchery releases were within the management containment objectives for the YKFP (Pearsons and Temple 2007). Hatchery precocious males have been shown to share similar food and space with other salmonids in the Yakima River, but indices

designed to evaluate competition with natural origin spring Chinook salmon in the main stem Yakima River suggests that competition is relatively low (Pearsons et al. 2007b). Furthermore, low incidences of piscivory have been documented in the upper Yakima watershed (Johnson et al. 2008).

Our study points out some challenges with integrating hatchery and wild populations while minimizing changes to natural populations. Many factors influence where and how many precocious males will be on the spawning grounds and many of these factors are outside of the control of hatchery managers. For instance, incidental harvest, flow rate, and water temperature may influence production and survival of precocious males in the river and their location during the spawning season. In addition, the number of precocious males released was uncorrelated with the number that was observed approximately five months later. Knowledge about precocious males on the spawning grounds in other river basins should contribute towards more effective hatchery and fishery management. It appears that many precociously mature fish released from the hatchery migrate downstream and fail to migrate back to the spawning grounds or die within the Yakima River prior to spawning time. Thus, hatchery production of precocious males in the Yakima River do not contribute favorably to harvest and may pose ecological risks to non-target taxa, but most of these fish have a low probability of contributing genes to future generations. Despite the high production of hatchery precocious males, managers in the Yakima Basin have chosen to discontinue efforts to reduce precocious males because growth modulation reduced the survival of migrants presumably because of their smaller size. The critical issue that was not directly addressed in this study was whether the hatchery changed the natural influence of precocious males in the target population. Until this issue is resolved, it is unclear how precocious males should be managed to conserve the upper Yakima Chinook population.

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Appendix 4

**DNA-Based Population-of-Origin Assignments of Chinook
Salmon Smolts Outmigrating Past Chandler Trap at Prosser Dam
(Yakima River) in 2014**

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Abstract

A population-of-origin assignment procedure was used to estimate the percentages of unknown-origin smolts from each of five stock groups outmigrating past Chandler Trap (Yakima River) from December 2013 – July 2014. Mixture analysis was conducted on a proportional subsample of 1,151 smolts collected during the outmigration at Chandler Trap. Assignment of each individual to a population-of-origin was determined if the posterior probability of the assignment was greater than 90.0%. The largest percentage of outmigrating smolts in the January/February, March, April, and May time strata was from the upper Yakima River stock while the June – July time stratum was dominated by the fall stocks with 88.8% of the total assignments. Comparison of morphological assessment and genetic assignment as a spring or fall Chinook smolt conducted for all time strata indicated agreement for 1,063/1,109 (95.9%) of the smolts.

Introduction

Production and survival of the Yakima River basin spring Chinook stocks (American River, Naches River, and upper Yakima River) are monitored, as part of the Yakima/Klickitat Fishery Project supplementation evaluation program. However, in the lower Yakima River, where the best facilities to collect samples exist, the three spring Chinook stocks are mixed with one another and with the Marion Drain and Yakima River fall Chinook stocks, during downstream juvenile migration. Thus, methodologies for discriminating stocks in an admixture are vital for development of stock-specific estimates. Domestication monitoring plans require discrimination of the three spring Chinook salmon stocks in the basin, and a complete analysis of migration timing and stock abundance for all Chinook requires discrimination of the two fall stocks as well. Accurate assignments of Chinook smolts captured at the Chandler fish passage facility to population-of-origin will allow researchers and managers to estimate production by the three spring Chinook stocks, assess smolt-to-smolt survival of the three spring Chinook stocks, and could be utilized to evaluate stock-specific environmental condition factors.

The methodology used in this study to estimate the population-of-origin for individual fish in a mixture followed a Bayesian approach by Rannala and Mountain (1997). This approach assumes linkage equilibrium among loci and uses the multilocus genotype of an individual to compute the probability of that genotype belonging to a population in the baseline. Others have used the methodology developed by Rannala and Mountain (1997) to provide robust population-of-origin assignments of unknown individuals (Hauser et al. 2006, Taylor and Costello 2006, and Waples and Gaggiotti 2006).

Calculation of population-of-origin for Chinook smolts trapped at Chandler trap throughout the entire outmigration (January through July) was hindered in the first few years of analysis for several reasons: non-representative temporal sampling of the downstream migration, past omission of the Marion Drain fall and lower Yakima River mainstem fall Chinook stocks from the DNA baseline, and by maintenance and other shutdowns of trap operations in December and January in many years. In the analyses of samples from 2004 - 2010, attempts were made to eliminate the problems present in previous analyses. A new sampling design was initiated to provide a proportional sample of smolts outmigrating past Chandler trap and a larger number of smolts were analyzed. Repeated multi-year samples of all five baseline stocks were used to characterize the potential sources of smolts in the Yakima River basin.

This report presents the population-of-origin assignments for outmigrating smolts collected at the Chandler trap during 2013.

Materials and Methods

Collections

There were no collections added to the Yakima River baseline this year. Since 1989, sampling crews from the Yakama Nation and WDFW have collected adult spawning ground tissue samples to be included in the baseline. The tissue samples consisted of dry-mounted scales or fin tissue preserved in 100% ethanol from five baseline stocks collected across multiple years

(American River spring, Naches River spring, upper Yakima River spring, Marion Drain fall, and lower Yakima River fall; Table 1 and Figure 1).

An estimated total of 447,080 smolts passed the lower Yakima River at Chandler from January 12 – July 18, 2014. This estimate was based on expansion of the total number of smolts counted at the Chandler trap (66,610) to account for trap efficiency, etc. Unknown-origin smolts were collected at Prosser Dam (Chandler Trap) following a sampling design that would identify a proportional number of smolt samples that represents the entire smolt outmigration. The following five time strata (January – February, March, April, May, and June – July) were used for analysis. Samples were collected from January 12 – July 18, 2014. These samples were genetically analyzed to get reliable estimates of population proportions. Each day, the total number of smolts at the trap was visually estimated before any processing occurred. If that number was below a predetermined threshold then a “standard” day’s sample was taken (e.g. 10 fish). If the number of smolts was above the threshold then a “peak” day’s sample was taken (e.g. 30 fish). The threshold for “standard” and “peak” days and the numbers of samples to be taken on each day varied for each of the time strata. These values were determined by analyzing the number of “peak” and “standard” days counted during four years of smolt outmigration monitoring. Based on this sampling design, 2,017 Chinook smolt samples were collected for genetic analysis.

The total estimated numbers of smolts passing the Chandler Trap each day were plotted with the total number of genetic samples that had been collected. A process was then employed to proportionalize the available genetic samples with the daily counts to provide a representative number of smolts that were outmigrating from January – July. A total of 1,151 smolts were identified for analysis.

DNA Extraction Methods

Genomic DNA was extracted by digesting a small piece of fin tissue (all smolt and some adult baseline collections) or scales (most adult baseline collections) using the nucleospin tissue kits obtained from Macherey-Nagel following the recommended conditions in the user manual. Extracted DNA was eluted with a final volume of 100 μ L.

PCR Methods

The polymerase chain reaction mixture contained the following for a 10 μ L reaction: approximately 25 ng template DNA, 1X Promega buffer, 1.5 mM MgCl₂, 200 μ M each of dATP, dCTP, dGTP, and dTTP, approx. 0.1 μ M of each oligonucleotide primer, and 0.05 units *GoTaq* Flexi DNA polymerase (Promega). Amplification was performed using MJ Research PTC-200 and Applied Biosystems 9700 thermocyclers. The thermal profile was as follows: an initial denaturation step of 2 minutes at 94°C; 40 cycles of 15 seconds at 94°C, 30 seconds at 50-60°C, and 1 minute at 72°C; plus a final extension step at 72°C for 10 minutes, followed by a final indefinite holding step at 10°C.

Eleven microsatellite DNA loci (Table 2) were amplified via the polymerase chain reaction (PCR) using fluorescently labeled primers (obtained from Applied Biosystems or Integrated DNA Technologies). Loci were combined in multiplexes to increase efficiency and decrease costs.

Data were collected using an AB-3730 Genetic Analyzer. Applied Biosystems GENEMAPPER v.3.7 software was used to collect and analyze the raw data and to determine genotypes at each locus (based on estimated allele sizes in base pairs using an internal size standard). Alleles were binned in GENEMAPPER using the standardized allele sizes established for the Chinook coastwide standardization efforts (Seeb et. al., 2007).

Population-of-origin Analysis

The program ONCOR (Kalinowski et al. 2008) was used to assign each individual to one of the baseline collections. ONCOR uses conditional maximum likelihood to estimate mixture proportions (Millar 1987) and genotype probabilities are calculated using a partial Bayesian procedure method of Rannala and Mountain (1997). This Rannala and Mountain (1997) method uses the expectation-maximization (EM) algorithm to calculate the population-source probabilities (posterior probabilities) for each sample. All assignments with a posterior probability greater than or equal to 90% were accepted.

Comparison of Morphological ID and Genetic Assignment

Smolts were categorized as spring or fall Chinook when they were intercepted at the Chandler Trap based on morphological characteristics. Three morphological features (length, size of the eye, and snout shape) were used to identify smolts as spring or fall (Mark Johnston, Yakama Nation; pers. comm.).

Results

Collections

A total of 1,151 unknown Chinook smolts were selected and analyzed from those collected at Chandler Trap. Smolt samples that had data for 10 or more loci were included for analysis. A total of 46 individuals were dropped from statistical analyses.

Population-of-origin Analysis

The mixture composition estimates for the entire 2014 smolt outmigration indicated that the largest overall percentage of spring smolts was from the upper Yakima River followed by the Naches River and American River in the first four strata. During the migration from January – May, the proportion of the upper Yakima River stocks was between 47.2 and 66.2% while the American River and Naches River spring stocks was between 8.8 and 41.2%. The proportion of the two fall stocks was between 0.0 – 21.7% for the first four time strata and 88.8% in the June - July time stratum (Table 3).

Comparison of Morphological ID and Genetic Assignment

A comparison of the morphological assessment to genetic assignment was conducted for all five time strata. A total of 11 smolts in January/February, 98 smolts in March, 288 smolts in April, 324 smolts in May, and 388 in the June/July time strata were scored, and therefore included in the analysis. Results for the time strata were as follows: January/February time stratum – all 11

smolts were assigned identically using morphological and genetic methods (11 spring); March stratum – all 98 smolts were assigned identically using morphological and genetic methods (98 spring); April time stratum – 286 out of 288 smolts were assigned identically using morphological and genetic methods (288 spring), both discrepancies were identified as fall by the genetic analysis and spring by the morphological identification; May time stratum – 319 out of 324 smolts were assigned identically using morphological and genetic methods (235 spring – 84 fall), three of the five discrepancies were identified as a fall by the genetic analysis and spring with morphological identification while two of the discrepancies were spring by the genetic analysis and fall by the morphological identification; June/July time stratum – 349 out of 388 smolts were assigned identically using morphological and genetic methods (35 spring and 314 fall), 8 discrepancies were assigned as spring by the genetic analyses while morphological identification was fall, the remaining 31 discrepancies were identified as a fall by the genetic analysis and spring with morphological identification.

Discussion

Collection of smolts at the Chandler Trap in 2014 utilized a sampling design intended to yield a sample that was proportional to the number of smolts passing the Chandler Trap. Sampling a proportional number of smolts was important to determine an accurate percentage of smolts from each stock that were outmigrating from the basin. Developing the sampling strategy for identifying a “standard” versus “peak” day of smolts that were in the trap and applying a sampling goal for those days allowed for a proportional sample. Subsampling the smolts collected for genetic analysis provided a best fit to the actual passage of smolts for a given day.

Monitoring the relative abundances of Chinook smolts in the Yakima River from the three different populations of spring Chinook (upper Yakima River, American River, and Naches River) and the two populations of fall Chinook (Marion Drain and lower Yakima River) requires the ability to estimate population composition of smolts outmigrating past Chandler trap. Because all five Chinook populations are intermingled when they pass Chandler trap, and the vast majority are unmarked and untagged, the only way to determine population-of-origin is by genetic analysis. This method requires that sufficient genetic differences exist among these populations in the Yakima River basin.

A baseline of 19 individual collections from the five populations in the Yakima River basin was used for the population-of-origin assignments of the outmigrating smolts. The baseline collections as a whole had higher genotyping failure compared to the Chandler smolt samples. Scales were taken from carcasses on spawning grounds for most baseline collections; therefore, DNA quality was presumably poorer than the Chandler smolt collection where tissue was collected from live fish. The upper Yakima River tissue collections were also taken from live fish at the hatchery and, therefore, genotyping success was higher for this collection than the other baseline collections.

Assessment of spring or fall smolts by morphological and genetic analysis revealed agreement with 46 individuals being identified differently between the two methods. Identification as a spring or fall smolt was the same for 1,063 smolts collected during the January – February, March, April, May, and June – July time strata.

The majority of the assignments between January and May were from the three spring stocks. The upper Yakima River spring stock accounted for the highest average percentage (52.5%) of smolts present in that period. Rank in abundance of the three spring stocks was the same in the three time strata (January-February, March, April, and May) with upper Yakima River spring stock having the most. The June-July time stratum was predominately composed of the fall Chinook stocks, accounting for over 88.8% of the total number of smolts.

Assessment of DNA Mixture Assignments from 2000 – 2013

Mixed stock analysis has been conducted on Chandler smolts since 2000 (Young 2004, Kassler et al. 2005, Kassler 2006, Kassler and VonBargen 2007, 2008, 2009 and 2010, Kassler and Peterson 2011, Kassler and Bell 2012, Kassler and Bowman 2013, 2014); however the sampling design for samples collected in 2000 – 2003 was not proportionalized during the run. The yearly assignments are therefore not comparable from those years. Beginning in 2004, staff at the Chandler trap utilized a sampling protocol to provide a number of smolts that was relative to the percentage of smolts passing that day. Samples were then subsampled at WDFW to provide a proportional number of samples that would represent the overall passage to be analyzed.

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Table 1. Nineteen Chinook salmon collections assembled into a baseline and used for the analysis of the known-origin and unknown-origin smolts. "*" the 05LU collection from Marion Drain was not used in the baseline, but is listed here as a collection from Marion Drain. The percentage of single locus genotypes missing are shown for each collection.

Baseline Collections	Collection Code	# Processed	# Analyzed	% Single Locus Genotypes Missing
American River - spring	89AG	80	77	10.4%
	91DQ	102	87	9.8%
	93DO	18	17	3.2%
	03EH	100	70	6.6%
		300	251	8.6%
Naches River - spring	89AC	76	74	11.4%
	89AI	26	22	7.0%
	93DQ	50	45	6.3%
	93DR	32	25	7.3%
little Naches River - spring	04BI	42	41	2.2%
	04EM	56	45	9.9%
		282	252	7.9%
upper Yakima River - spring	92DN	24	23	5.9%
	97DA	123	115	3.9%
	03GO	99	99	1.4%
		246	237	3.0%
Marion Drain - fall	89BX	100	92	8.3%
	92FQ	92	92	5.4%
	93DY	8	8	8.0%
	05LU*	65	47	15.3%
		265	239	8.6%
lower Yakima River - fall	90DF	109	104	12.6%
	93DW	82	80	9.8%
	98FB	61	50	8.7%
		252	234	10.8%
Chandler Trap Smolts - 2014	14CZ	1,151	1,109	2.8%

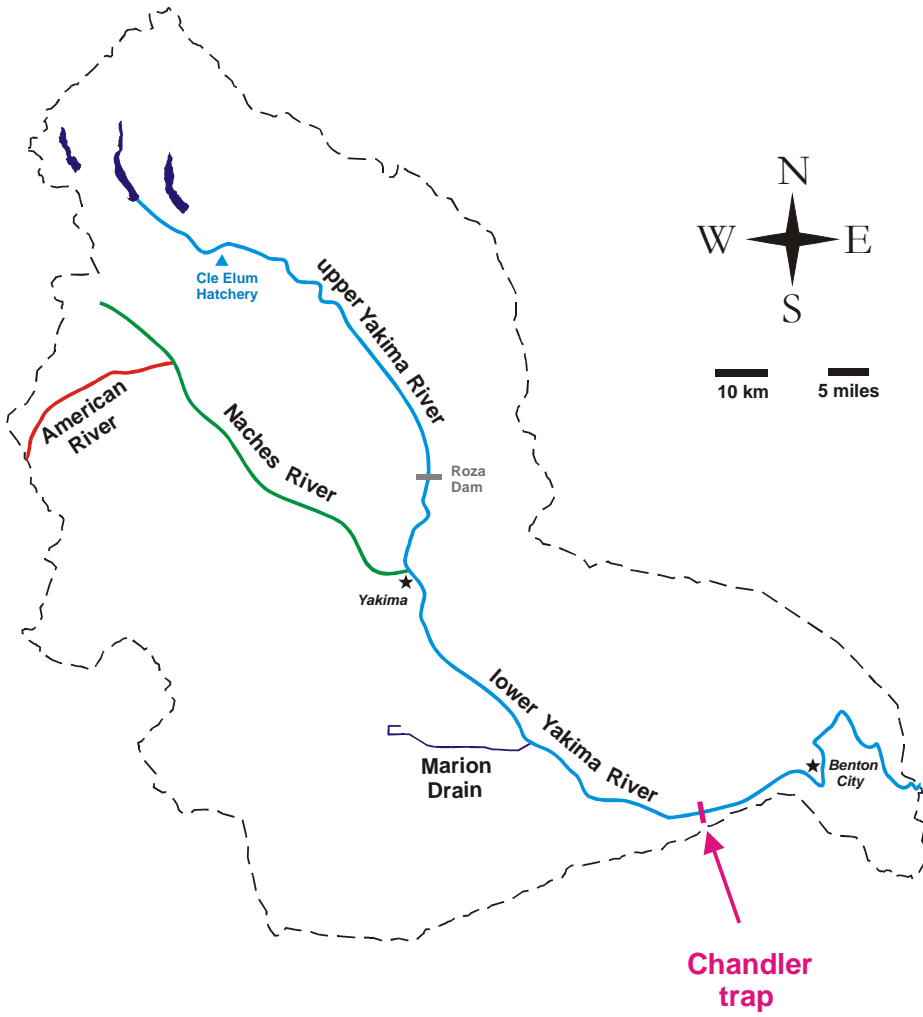
Table 2. Microsatellite locus information (number alleles/locus and allele size range) for multiplexed loci used in the analysis of Chinook from five stocks in the Yakima River Basin. Also included are the percent missing genotypes for both the baseline and smolt\ collections and heterozygosity (observed (H_o) and expected (H_e)) for each locus.

Multiplex	Locus	Annealing temp °C	# Alleles/ Locus	Allele Size Range (bp)	% missing genotypes baseline N = 1,166	% missing genotypes smolts N = 1,109	Heterozygosity	
							H_o	H_e
Ots-M	<i>Oki-100</i> ^a	50	41	164 - 365	11.6%	0.5%	0.913	0.940
	<i>Ots-201b</i> ^a	50	42	137 - 310	7.1%	0.5%	0.916	0.936
	<i>Ots-208b</i> ^b	50	52	158 - 342	9.7%	1.2%	0.943	0.954
	<i>Ssa-408</i> ^c	50	32	184 - 308	3.5%	2.0%	0.827	0.934
Ots-N	<i>Ogo-2</i> ^d	60	19	202 - 256	3.7%	1.4%	0.756	0.854
	<i>Ssa-197</i> ^e	60	38	181 - 318	11.8%	0.7%	0.915	0.940
Ots-O	<i>Ogo-4</i> ^d	56	17	132 - 164	15.2%	0.8%	0.776	0.884
	<i>Ots-213</i> ^b	56	40	182 - 362	9.3%	0.7%	0.908	0.940
	<i>Ots-G474</i> ^f	56	15	152 - 212	3.0%	6.4%	0.507	0.697
Ots-R	<i>Ots-3M</i> ^g	53	15	128 - 158	2.5%	14.1%	0.601	0.672
Ots-S	<i>Ots-9</i> ^g	60	8	99 - 113	5.1%	2.1%	0.668	0.709
^a = Unpublished ^b = Greig et al. 2003 ^c = Cairney et al. 2000 ^d = Olsen et al. 1998 ^e = O'Reilly et al. 1996 ^f = Williamson et al. 2002 ^g = Banks et al. 1999								
11								

Table 3. Stock-of-origin assignments for five stocks of Chinook in the Yakima River Basin using ONCOR.

	American R.	Naches R.	upper Yakima R.	Marion Drain	lower Yakima R.
Jan - Feb	11.7%	41.2%	47.2%	0.0%	0.0%
March	12.0%	21.7%	66.2%	0.0%	0.0%
April	9.0%	29.9%	60.1%	0.0%	1.0%
May	8.8%	27.9%	36.6%	5.1%	21.7%
June-July	1.6%	0.0%	9.6%	6.4%	82.4%

Figure 1. Geographic location of the Chandler trap on the Yakima River, Washington and the primary streams in the basin.



Appendix 5

**DNA-Based Parentage Assignments of Chinook Salmon from
the Cle Elum Spawning Channel in 2013**

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Abstract

We used a maximum likelihood parentage assignment procedure to estimate the reproductive output of Chinook salmon spawners from the hatchery-control line (two generations of hatchery influence) and the supplementation hatchery line (SH – one generation of hatchery influence) in the Cle Elum experimental spawning channel for the 2013 brood year. The assignments were based on offspring genotypes at 14 microsatellite loci. The probabilities of exclusion (inferring non-parentage by randomly picked adults) assuming neither parent was known were estimated to be 0.999999. Two thousand three hundred and twenty one of 2,445 fry from the 2013 brood that were genotyped at nine or more loci were assigned to a parental pair with 95% confidence. The number of progeny attributed to individual potential parents was quite variable, ranging from 0 to 285 for all males and from 0 to 153 for females. The sum of progeny attributed to the hatchery-control line males and females was 1,377, while the sum of progeny attributed to supplementation hatchery line males and females was 944.

Introduction

Although hatcheries have been extensively utilized in Chinook salmon management for over 100 years, only recently have rigorous experiments been developed to measure the relative reproductive success of hatchery- and natural-origin spawners in a shared natural setting. Some of the difficulty in designing informative studies has stemmed from the challenges of controlling entry to natural spawning areas and collecting representative samples of recently hatched fry. Furthermore, if control could be established over the potential spawners in the spawning area, the measurement of individual reproductive output still would require a means of associating individual fish captured in one year with individuals that spawned in a previous year. The spawning behavior of Chinook salmon adds to the complexity of quantifying individual reproductive output through behavioral observations: at a redd site, a female might be courted by several males that compete for access to the female, providing opportunities for multiple paternity in a single redd. In areas with moderate to high spawning densities, males might attend females on several adjacent redds. Microsatellites, a class of highly polymorphic, codominant DNA markers, provide a means to quantify individual spawners' reproductive output. A suite of 10 to 15 highly variable microsatellites can resolve individual identity in a moderate to large population, and through a simple inheritance model, can illuminate parent-offspring relationships.

Washington Department of Fish and Wildlife (WDFW) and the Yakama Nation (YN) are cooperating on a study of Chinook salmon reproductive success in a presumably closed access spawning observation channel at the Cle Elum Hatchery. Viewing blinds line the channel, allowing researchers to observe spawning activities.

Chinook salmon carrying visible external marks were released into the spawning channel in September 2013. Hatchery-control line (two generations of hatchery influence) males and females were released into three of six shared spawning areas and supplementation hatchery line (one generation of hatchery influence) males and females were released into the other three shared spawning areas to select and compete for mates. Prior to the release of the potential spawners, researchers collected and preserved samples of fin tissue to enable genetic characterization of the potential spawners and to allow subsequent inference of parent/offspring relationships after juveniles were collected and genotyped. One group of researchers examined morphological characteristics of these potential parents and observed and recorded spawning area behaviors and interactions. The results of the morphological and behavioral work are described in a separate report.

The potential parents' fin tissue samples and the collected progeny (fry) were delivered to the WDFW Molecular Genetics Laboratory in Olympia, Washington for genetic screening and parentage analysis following the same protocols that have been used from 2002 – 2007, 2009 – 2014 (Young and Kassler 2005, Kassler 2005, Kassler 2006, Kassler and Von Bargen 2007, 2008, and 2010, Kassler et al. 2011; Kassler and Peterson 2012, 2013; Kassler and Smilansky 2014). The genetic analyses provide direct, quantitative estimates of fry production by individual spawning Chinook salmon. This report presents the parentage results for the 2013 – 2014 Cle Elum spawning channel experiments.

Materials and Methods

Collection of potential spawners – 2013

Fin tissue was collected from a total of 48 adult females and 48 adult males (Table 1) prior to their release into each of six sections in the spawning channel during September 2013. The genetic analysis program CERVUS (version 3.0; Marshall et al. 1998) was used to check for identical multilocus genotypes among the potential parents. Data recorded for each released fish included gender, and whether it was of hatchery-control line origin or supplementation hatchery line origin (Table 1).

Collection of Fry

Fry collections occurred from December 3, 2013 to February 28, 2014. Fry samples were collected from each section daily when fry were present. During that period a total of 2,567 fry were collected.

DNA Extraction Methods

Genomic DNA was extracted by digesting a small piece of fin tissue using the nucleospin tissue kits obtained from Macherey-Nagel following the recommended conditions in the user manual. Extracted DNA was eluted with a final volume of 100 μ L.

PCR Methods

Potential spawners and offspring from 2014 were genotyped at 14 loci (Table 2). Potential spawners were screened twice and scored independently at all 14 loci by two biologists to minimize potential genotyping error of the parents.

The polymerase chain reaction mixture contained the following for a 10 μ l reaction: approximately 25 ng template DNA, 1X Promega buffer, 1.5 mM $MgCl_2$, 200 μ M each of dATP, dCTP, dGTP, and dTTP, approx. 0.1 μ M of each oligonucleotide primer, and 0.05 units *GoTaq* Flexi DNA polymerase (Promega). Amplification was performed using MJ Research PTC-200 and AB 9700 thermocyclers. The thermal profile was as follows: an initial denaturation step of 2 minutes at 94°C; 40 cycles of 15 seconds at 94°C, 30 seconds at 49-58°C, and 1 minute at 72°C; plus a final extension step at 72°C for 10 minutes, followed by a final indefinite holding step at 4°C.

Microsatellite DNA loci (Table 2) were amplified via the polymerase chain reaction (PCR) using fluorescently labeled primers (obtained from Applied Biosystems or Integrated DNA Technologies). Loci were combined into multiplexes to increase efficiency and decrease costs.

Data were collected using an AB-3730 Genetic Analyzer. Applied Biosystems GENEMAPPER v.3.7 software was used to collect and analyze the raw data and to determine genotypes at each locus (based on estimated allele sizes in base pairs using an internal size standard). Alleles were binned in GENEMAPPER using the standardized allele sizes established for the Chinook coastwide standardization efforts (Seeb et. al. 2007).

Parentage Assignments

The dataset included 34,580 single-locus genotypes. A genotyping error rate in that dataset of 1.0% would result in 346 incorrect single-locus genotypes. Our error rate is unknown, but possibly greater than 1%. Since parentage analyses involve comparing genotypes of candidate parental pairs with offspring genotypes, genotyping errors can produce parent-offspring genotype mismatches and suggest exclusion of true parent-offspring pairings from consideration. Alternatively, genotyping errors can lead to failure to exclude parent-offspring pairings that are incorrect. We used a maximum likelihood procedure, implemented in CERVUS (version 3.0; Marshall et al. 1998) to infer parent-offspring relationships. The procedure uses allele frequency data to assign likelihoods to parent-offspring combinations, and allows mismatching genotypic data to be evaluated concurrently with matching genotype data.

Genotyping error is not the only potential source of mismatches between the genotypes of fry and their putative parents. We would expect allele misidentification to be randomly distributed throughout the genotype dataset and not to occur in clusters. Parent-offspring mismatches can result also from germ-line mutation in which a parent passes a changed allele to its offspring or from the inadvertent exclusion of one or more contributing parents from the parental dataset. These mismatches are due to correctly assigned but unexpected genotypes, and we expect that those genotypes should cluster in families. Distinguishing between mutation-based mismatches and mismatches that result from reproductive participation by un-genotyped parents is difficult. Assuming that all dams in the experimental channel are represented in the parental data set, we might suspect reproductive participation by one or more unrepresented sires if groups of fry that are assigned to a dam-offspring relationship with no mismatching loci, have multiple locus mismatches with all candidate sires, and no more than four alleles at a locus within the group. The data set was carefully examined for evidence of reproductive contributions by such un-genotyped parents (because evidence of ungenotyped parents had been observed in previous years).

Results

Parents

Genetic analysis revealed that all 96 fish released or found in the spawning channel had unique genotypes. There were a total of 24 hatchery control line (HC) adult males, 24 HC adult females, 24 supplementation hatchery line (SH) adult males, and 24 SH adult females. Four HC males and four HC females were released into three of the six sections and four SH males and four SH females were released into the other three sections (Table 1).

Loci Screened

A total of 14 loci were screened and all 14 were used in the analysis (Table 2). Number of alleles ranged from 4 - 29 (*Ots-9* and *Omm-1080* respectively) and observed heterozygosity ranged from 0.398 – 0.954 (*Ots-G474* and *Omm-1080* respectively). Individual exclusionary power was below 45.7% for five loci (*Ogo-2*, *Ogo-4*, *Ots-G474*,

Ots-3M, and *Ots-9*) and above 58.4% for the remaining loci when neither parent was known. Exclusionary power was below 39.9% for three loci (*Ots-G474*, *Ots-3M* and *Ots-9*) and above 56.7% for the remaining loci when one parent was known. Cumulative exclusionary power was 0.999999 for analysis using all loci when one parent was known.

Parentage Assignments

Parentage assignments were made when genotype data was available for nine or more loci. All 96 parents were genotyped at 10 or more loci while 2,321 of the 2,445 offspring were successfully genotyped at nine or more loci (Table 3).

Parentage analysis was conducted independently for each of the six sections using all 96 adults as possible parents. Each fry was assigned a dam-sire-fry combinations (trios) based on the most likely candidate male parents (sires) and female parents (dams). Those assignments yielded possible. Any fry-sire assignments with more than two mismatching loci were excluded from further consideration.

Of the total 2,445 fry included in the analysis a total of 2,321, fry were assigned to a single male and female parent ($2,321/2,445 = 94.9\%$).

Discussion

Approximately 95 percent successes were achieved at inferring parent-offspring relationships. Examination of Table 4 reveals a very uneven pattern of reproductive success among the candidate parents. Based on the subsample of 2,321 fry that were successfully assigned parents, the range of inferred reproductive output among males was 0 - 285 fry; the range for the same period in reproductive output among females was 0 – 153 fry. Some of the dam-sire matings we inferred are well supported (there were a lot of fry assigned to them) and some are weakly supported (not many fry were assigned to them). Caution should be used when interpreting dam-sire-fry combinations that were inferred rarely. Future integration of fecundity estimates for spawners will enrich the interpretation of these estimates of reproductive output.

Interpretation of the inferred parental reproductive output based on parentage assignments by genetic analysis requires the consideration and analysis of individual fish attributes, including fecundity and body size, the closed nature of the experimental environment in which sub-dominant males had a more limited number of alternative females to court than they might have had in an open system, and relative stocking levels and synchronicity of spawning.

Acknowledgements

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Table 1. Potential Chinook salmon spawners in the six section of the Cle Elum experimental spawning channel in 2013. Origin is identified as hatchery-control (HC) or supplementation hatchery (SH).

	Section 1 – 1A	Section 1 – 2A	Section 1 – 3A	Section 2 – 1A	Section 2 – 2A	Section 2 – 3A
Origin	Females	Females	Females	Females	Females	Females
SH	--	4	--	4	--	4
HC	4	--	4	--	4	--
	Males	Males	Males	Males	Males	Males
SH	--	4	--	4	--	4
HC	4	--	4	--	4	--
	Females	Females	Females	Females	Females	Females
Origin	Females	Females	Females	Females	Females	Females
SH	--	4	--	4	--	4
HC	4	--	4	--	4	--
	Males	Males	Males	Males	Males	Males
SH	--	4	--	4	--	4
HC	44	--	4	--	4	--

Table 2. Locus summary.

Locus	# alleles	# parents genotyped	H _O (observed)	H _E (expected)	Exclusionary power	
					neither parent	one parent
Oki-100	18	93	0.892	0.904	0.663	0.798
Ots-201b	20	91	0.901	0.894	0.638	0.779
Ots-208b	27	92	0.891	0.946	0.786	0.880
Ssa-408	17	91	0.659	0.899	0.651	0.789
Ogo-2	8	96	0.917	0.815	0.457	0.633
Ssa-197	21	96	0.948	0.920	0.709	0.830
Ogo-4	9	93	0.720	0.759	0.383	0.567
Ots-213	23	93	0.892	0.938	0.762	0.865
Ots-G474	7	93	0.398	0.403	0.087	0.235
Omm-1080	29	93	0.925	0.954	0.814	0.897
Ots-3M	7	94	0.628	0.624	0.223	0.399
Ots-211	22	94	0.957	0.927	0.731	0.844
Ots-212	18	88	0.898	0.873	0.584	0.738
Ots-9	4	95	0.632	0.610	0.200	0.362

Table 3. Summary of genotyping efficiency in potential parents and offspring.

Loci genotyped	Parents (13JF)	Offspring (14QI)
14	75	1,969
13	9	206
12	5	138
11	5	74
10	2	42
9	0	16
8	0	8
7	0	9
6	0	6
5	0	5
4	0	6
3	0	3
2	0	8
1	0	20
0	0	44
	96	2,554

Table 4. Total number of offspring assigned to females and males from each of the six sections in the spawning channel and the life stage (HC - hatchery control line; SH - supplementation hatchery line) for each fish.

Females	Section	HC or SH	Total Offspring	Males	Section	HC or SH	Total Offspring
13JF085	1-1A	HC	48	13JF081	1-1A	HC	60
13JF086	1-1A	HC	49	13JF082	1-1A	HC	1
13JF087	1-1A	HC	0	13JF083	1-1A	HC	0
13JF088	1-1A	HC	153	13JF084	1-1A	HC	189
13JF003	1-1B	HC	137	13JF001	1-1B	HC	0
13JF005	1-1B	HC	90	13JF002	1-1B	HC	0
13JF006	1-1B	HC	57	13JF004	1-1B	HC	0
13JF009	1-1B	HC	0	13JF007	1-1B	HC	285
13JF061	1-2A	SH	113	13JF057	1-2A	SH	166
13JF062	1-2A	SH	0	13JF058	1-2A	SH	0
13JF063	1-2A	SH	0	13JF059	1-2A	SH	0
13JF064	1-2A	SH	53	13JF060	1-2A	SH	0
13JF014	1-2B	SH	0	13JF010	1-2B	SH	0
13JF015	1-2B	SH	0	13JF011	1-2B	SH	129
13JF016	1-2B	SH	0	13JF012	1-2B	SH	0
13JF017	1-2B	SH	129	13JF013	1-2B	SH	0
13JF069	1-3A	HC	0	13JF065	1-3A	HC	0
13JF070	1-3A	HC	0	13JF066	1-3A	HC	0
13JF071	1-3A	HC	139	13JF067	1-3A	HC	216
13JF072	1-3A	HC	78	13JF068	1-3A	HC	0
13JF021	1-3B	HC	8	13JF008	1-3B	HC	0
13JF022	1-3B	HC	120	13JF018	1-3B	HC	163
13JF023	1-3B	HC	25	13JF019	1-3B	HC	0
13JF024	1-3B	HC	77	13JF020	1-3B	HC	67
13JF077	2-1A	SH	0	13JF073	2-1A	SH	0
13JF078	2-1A	SH	67	13JF074	2-1A	SH	0
13JF079	2-1A	SH	100	13JF075	2-1A	SH	176
13JF080	2-1A	SH	9	13JF076	2-1A	SH	0
13JF029	2-1B	SH	123	13JF025	2-1B	SH	0
13JF030	2-1B	SH	0	13JF026	2-1B	SH	134
13JF031	2-1B	SH	0	13JF027	2-1B	SH	0
13JF032	2-1B	SH	11	13JF028	2-1B	SH	0
13JF053	2-2A	HC	146	13JF049	2-2A	HC	3
13JF054	2-2A	HC	0	13JF050	2-2A	HC	198
13JF055	2-2A	HC	37	13JF051	2-2A	HC	5
13JF056	2-2A	HC	23	13JF052	2-2A	HC	0
13JF037	2-2B	HC	5	13JF033	2-2B	HC	180
13JF038	2-2B	HC	107	13JF034	2-2B	HC	6
13JF039	2-2B	HC	78	13JF035	2-2B	HC	4
13JF040	2-2B	HC	0	13JF036	2-2B	HC	0
13JF093	2-3A	SH	0	13JF089	2-3A	SH	0
13JF094	2-3A	SH	44	13JF090	2-3A	SH	93
13JF095	2-3A	SH	94	13JF091	2-3A	SH	45
13JF096	2-3A	SH	0	13JF092	2-3A	SH	0
13JF045	2-3B	SH	106	13JF041	2-3B	SH	162
13JF046	2-3B	SH	22	13JF042	2-3B	SH	39
13JF047	2-3B	SH	73	13JF043	2-3B	SH	0
13JF048	2-3B	SH	0	13JF044	2-3B	SH	0
			2,321				2,321

Appendix 6

**Rearing Spring Chinook Summer Abundance and Habitat Use in
the Upper Yakima River Basin**

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Abstract

The carrying capacity of a watershed is an important factor in determining whether supplementation is a viable technique of increasing natural production. Preliminary analysis suggests that density dependent mechanisms affecting spring Chinook growth and survival are present in the upper Yakima River basin prior to abundance estimates conducted in the fall, and potentially prior to or during the summer rearing period. If the Yakima River is at capacity for rearing subyearlings in some years, then supplementation efforts can only serve to increase the number of naturally produced smolts when natural production is below that capacity. Therefore, identifying the factors that limit natural production is critical if restoration efforts aimed at maintaining or increasing natural production are to achieve their intended biological goals. We conducted snorkeling surveys documenting observed abundance of rearing subyearling Chinook by habitat classification in the three contiguous reaches of the upper Yakima River in the summer of 2014. We further developed a relationship between spring Chinook subyearling length and effective territory size, and investigated ratios of agonistic and foraging behaviors. Lastly we assessed water temperature and visibility as potential biases in our summer snorkeling sampling efficiency. Our data suggest that habitat type is a significant factor determining abundance of rearing subyearling Chinook in the upper Yakima River, and that the space required of rearing subyearlings appears to expand linearly with increasing growth. Multi-year analysis of abundance indicates that summer rearing densities trend with fall abundance estimates. Temperature and visibility do not appear to have significantly affected estimates of observed abundance among study reaches.

Introduction

The carrying capacity of a watershed is an important factor in determining whether supplementation is a viable technique of increasing natural production. In the Yakima River Basin, carrying capacity can limit the number of naturally produced spring Chinook salmon *Oncorhynchus tshawytscha* even when supplementation mechanics are operating perfectly (Busack et al. 1997). Preliminary analysis suggests that density dependent mechanisms affecting spring Chinook survival exist in the upper Yakima River after fall spawning and prior to or during the parr stage the following fall (Johnson et al. 2009). If the Yakima River is at capacity for rearing subyearlings in some years, then supplementation efforts can only serve to increase the number of naturally produced smolts when natural production is below that capacity. Therefore, identifying the factors that limit natural production is critical if restoration efforts aimed at maintaining or increasing natural production are to achieve their intended biological goals. There are many potential limitations to rearing spring Chinook growth and survival in the Yakima Basin. Pearsons et al. (2007) speculated that factors limiting the growth and/or survival of juvenile spring Chinook salmon in the upper Yakima Basin may differ depending upon past habitat alteration and present flow management. The upper Yakima River is highly regulated for irrigation purposes with unnatural flow regimes that result in wide habitat shifts both spatially and temporally. For example, temporal habitat limitations may exist at multiple life history stages as food and space requirements change, and spatial limitation may occur as a result of managed flows (e.g. holding runoff to fill reservoirs, seasonal releases) to satisfy irrigation demands downstream. For these reasons, reach specific estimates of spring Chinook abundance and distribution are thought necessary to identify factors limiting natural production in the upper Yakima River basin (Johnson et al. 2009). We conducted snorkeling surveys in two study reaches of the upper Yakima River in 2014 to index spring Chinook abundance and distribution during summer rearing, and compared meso-scale habitat measures to those indices, to better identify primary habitat needs of spring Chinook in the upper Yakima during this life-stage. We also compiled a larger multi-year dataset to increase our power for detection of differential abundance among habitat classifications.

Different species and life stages of fishes show different preferences for specific microhabitat parameters (Lister and Genoe 1970; Hearn and Kynard 1986; Roper et al. 1994). Further, the variation of microhabitats used by a species and life stage of fish is typically positively related to the density of that species/life stage as well as the density of competitor species (Allee 1982; Ross 1986; Grant and Kramer 1990; Robertson 1996). Knowing the temporal and spatial dynamics of rearing Chinook abundance at the reach scale allows for the evaluation of productivity through the development of stock recruit relationships, but may not be sufficient to identify the specific factors that are limiting production. Comparisons of the types of microhabitat utilized and the ranges used between high and low productivity sites may allow us to better identify these factors. We conducted timed underwater observations of sub-yearling spring Chinook in the summer of 2014. We measured the distance and frequency of feeding and agonistic strikes, characterized the focal position by measuring environmental factors such as depth and velocity, and attempted to index the ratio of food availability and energy required to hold at known focal positions.

Because large portions of the upper Yakima are difficult to sample using boat-based electrofishing, and waters are too deep to effectively sample using a backpack electrofisher, snorkeling surveys are thought to be the most practical and cost effective method of generating

estimates of abundance and distribution in the summer time period (Johnson et al. 2010, 2011); when flows are high and subyearling Chinook have begun to utilize habitats in areas of higher flow (Pearsons et al 2008). Under adequate sampling conditions, snorkeling is a quick and inexpensive technique for estimating population numbers (Zubick and Fraley 1988; Hillman 1992), has been suggested as an effective census technique in larger fluvial systems when electrofishing may be less than ideal (Schill and Griffith 1984), and has been routinely used for annual monitoring efforts in larger systems such as the Okanogan Basin (Arterburn et al. 2008) and the lower American River in California (FFC 2006). Snorkeling surveys are also largely passive, which may be beneficial when ESA listed species such as steelhead (*Oncorhynchus mykiss*) overlap with target species. However, although snorkeling is a widely used methodology in fisheries science (Brignon 2011) there are a number of factors such as temperature, visibility, and behavioral response (Hillman 1992; Thurow 1994; Brignon 2011), which may bias estimates of abundance. We compared estimates of relative abundance to both temperature and stream velocity among sampling reaches to determine the extent of any existing bias attributable to those factors.

Methods

Study area

The 2014 study area consisted of two contiguous reaches of the upper Yakima River that are similar in flow, gradient, and stream width. All three sections experience fairly consistent flows over the summer period, generally less than one meter per second, which is the upper limit suggested as restrictive to sub yearling spring Chinook in the upper Yakima River (Pearsons et al. 2007). Beginning with the upstream boundary at Easton Dam, the study reaches were: Easton (10.7 km), and Nelson (7.3 km). Sampling was conducted between July 8th and August 25th, 2014.

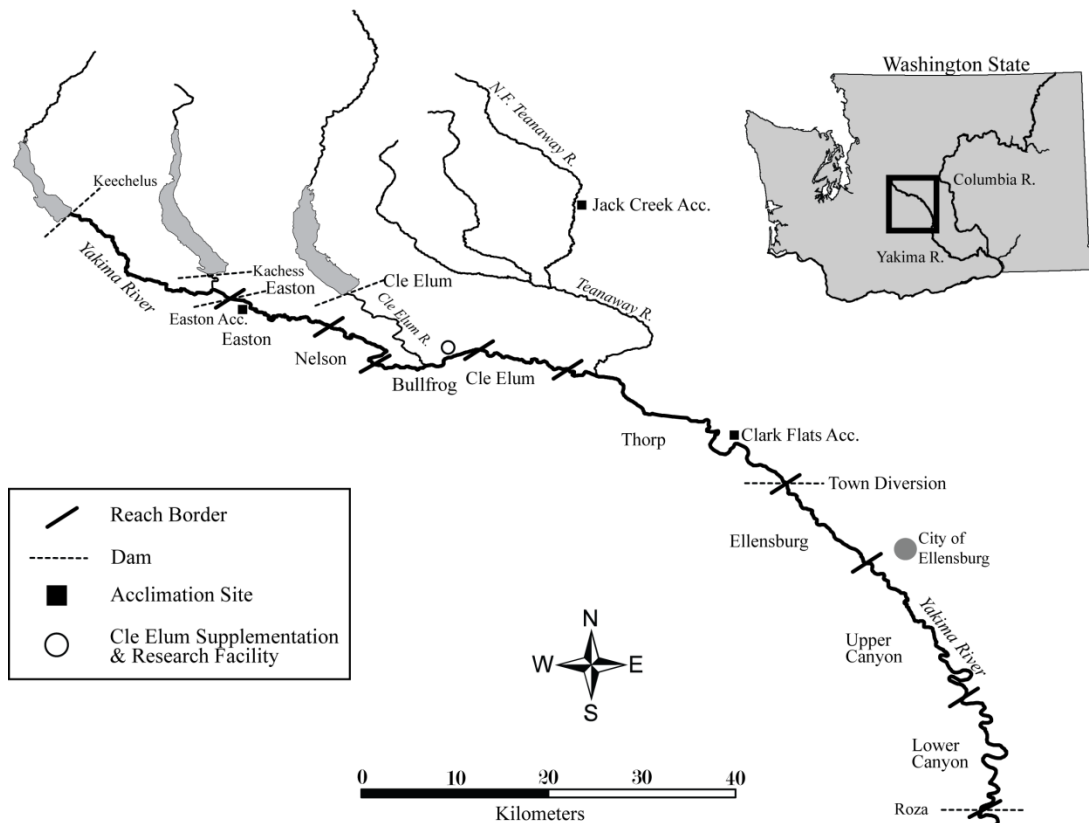


Figure 1. Survey reaches within the upper Yakima basin. Snorkeling surveys in 2014 were conducted in the Easton and Nelson reaches of the main-stem Yakima River.

Abundance/Sampling units

We performed relative abundance snorkeling estimates of juvenile Chinook in the Easton and Nelson reaches. Briefly, each reach was divided into sampling units by common habitat based classifications (Hawkins 1993; Arend 1999). Habitat classifications used over the course of the sampling period are presented in (Table 1.). Sampling units were limited to 100 m in length in order to avoid sampling bias due to snorkeler fatigue. Exceptions to site length were made when classified habitat units were only slightly longer than the maximum sampling unit length (e.g., 105 meters). Unit borders were marked by GPS during a preliminary survey at the beginning of each year so that the sampling units could be easily located and refined prior to sampling. Two independent crews conducted surveys daily. Each crew was systematically rotated through the sampling reaches for the duration of the study. Survey sites within each reach were selected randomly without replacement. Due to the low likelihood of spring Chinook parr in center channel locations (Quinn 2005; Johnson et al. 2009, 2010, 2011) each bank survey was considered an independent replicate of rearing abundance. Comparisons of abundance estimates in the Easton and Nelson reaches were made among years using a main-effects

ANOVA design. We also investigated correlation between our annual summer and fall abundance estimates (see Temple et. al (2011) for a description of spring Chinook fall abundance estimates).

Table 1. Definition of habitat types used to classify sampling sites within the Easton and Nelson reaches of the upper Yakima River 2014.

Habitat Unit	Depth	Appearance of water surface	Substrate types
Deep Riffle	> 0.5 m	Swift current turbulent, unbroken surface	Generally cobbles
Glide	> 0.5 m uniform depth	Slow current unbroken surface	All types possible
Pool	> 1.5 m one deep spot	Slow current unbroken surface	All types possible
Rapid	0-2 m	Swift current very turbulent broken surface	Large boulders or bedrock frequently breaking water surface
Riffle	< 0.5 m	Swift current turbulent, unbroken surface	Generally cobbles
Run	0.5-1.5 m	Moderate current unbroken surface	All types possible

Survey crews sampled the upstream locations first, using the preliminary survey GPS locations to locate the approximate upstream boundary of each survey site. Site boundaries were refined immediately prior to sampling to insure the best break point between habitat units and were re-marked as necessary using GPS. Surveyors walked the bank to the lower boundary of the site or floated to the bottom and then waited a period of five minutes before beginning the upstream survey. One snorkeler on each bank then worked upstream parallel to one another identifying and enumerating all fish encountered. Data were recorded on a cuff made from a six-inch section of four-inch I.D. PVC pipe. Visibility (focal range) was measured daily in each reach by moving a scale silhouette of a Chinook away from the snorkeler (under water) until parr marks were no longer visible (Thurow 1994). Water temperature was recorded immediately prior to each sampling event.

Stationary snorkeling was used to observe and record sub-yearling spring Chinook territory size, in the main-stem Yakima River. Using a clipboard, observers recorded direction and distance of juvenile Chinook salmon movements relative to the focal position. Underwater stopwatches were used to time each observation. Observations generally ranged in duration from one to five minutes, terminating when the target Chinook left the observer's view. The holding position of the fish, its focal position, was variable throughout most observations. Movements were recorded onto a two-dimensional datasheet graph, which represented distance in body lengths, and direction relative to the focal point (Figure 2). The direction of a fish movement was classified as top, bottom, left or right, with each consisting of a ninety-degree range centered at the focal point (Figure 2). Movements were classified as either feeding or agonistic depending upon whether the focal fish 1) obtained a food item, or 2) moved aggressively as an instigator or in response to an intruding Chinook. Feeding strikes were denoted by a dark circle or point on the datasheet, and agonistic strikes with an "x".

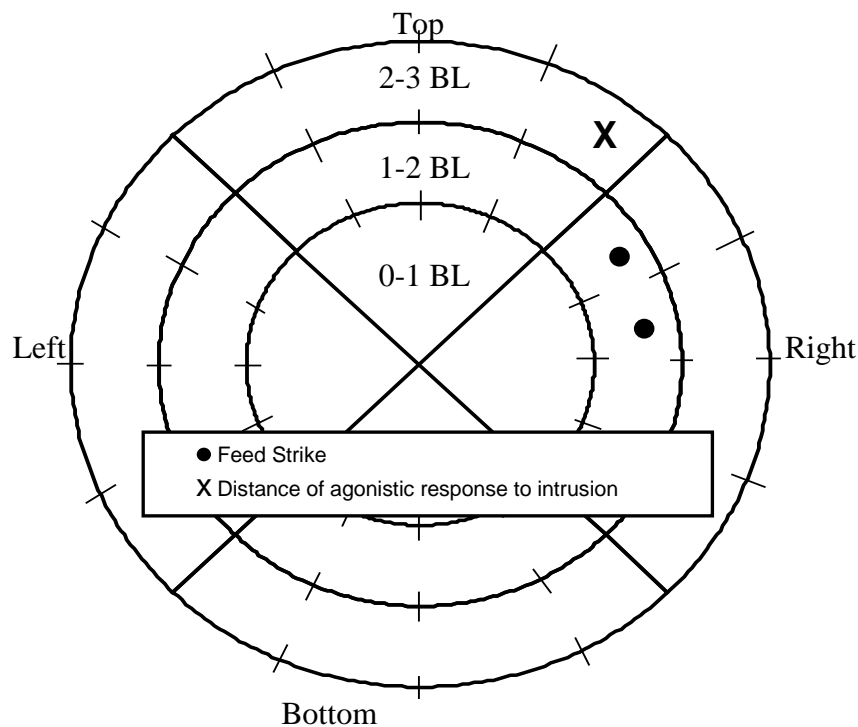


Figure 2. Graphical representation of the data sheets used in recording age-0 Chinook salmon territory size. Data is recorded in units of focal fish body length (BL).

Flow ratio (Pearsons et al. 2006) is a parameter designed to index the ratio of food availability and energy required to hold at a known focal position. This ratio is defined as the highest flow velocity within two body lengths of the focal position divided by the focal point velocity. A flow ratio greater than one, indicates at least one adjacent velocity greater than the focal point velocity. We measured velocities at one and two body lengths to either side and above and below the observed spring Chinook focal positions. Core microhabitat variables: estimated length, focal point velocity, focal depth, total depth, and temperature, were measured around the each observed focal position

Results

A total of 67 sites were surveyed in the three study reaches between July 7th and August 25th, 2014 (Table 2.) for a total of 128 total replicates. We did not detect a significant difference in sub-yearling Chinook density between study reaches in 2014 ($t = 1.0$, $df = 126$, $P = 0.31$), or in a multi-year analysis (ANOVA: $F_{1,6} = 0.02$, $P = 0.90$). Mean density was significantly greater in 2010 when compared to all other years (ANOVA: $F_{1,6} = 7.3$, $P < 0.01$; Tukey post-hoc: $P < 0.01$; Figure 3). Summer and fall mean spring Chinook densities trended, but were not significantly correlated with an alpha = 0.05, ($R^2 = 0.56$, $P = 0.06$). We did not detect a significant difference in spring Chinook density among habitat types in 2014 (ANOVA: $F_{5,122} = 0.64$, $P = 0.67$; Table 2).

Table 2. Physical parameters of 2014 snorkeling survey sites by sampling reach.

Habitat Classification	n	Mean site length (m)	SD	n	Site width (m)	SD
Easton 2014						
Deep Riffle	4	35.0	4.1	4	18.9	3.2
Glide	11	80.7	21.0	11	20.2	7.2
Pool	5	47.4	16.0	5	20.9	3.6
Rapid	1	30.0	n/a	1	20.3	n/a
Riffle	6	54.8	17.4	6	19.2	6.7
Run	10	53.3	16.5	10	17.6	5.2
Nelson 2014						
Deep Riffle	3	53.3	23.1	3	25.9	1.3
Glide	11	89.4	17.7	11	31.3	2.5
Pool	3	43.3	12.6	3	22.1	0.7
Rapid	1	45.0	n/a	1	29.0	n/a
Riffle	1	85.0	n/a	1	36.3	n/a
Run	8	91.8	19.9	8	31.0	6.6

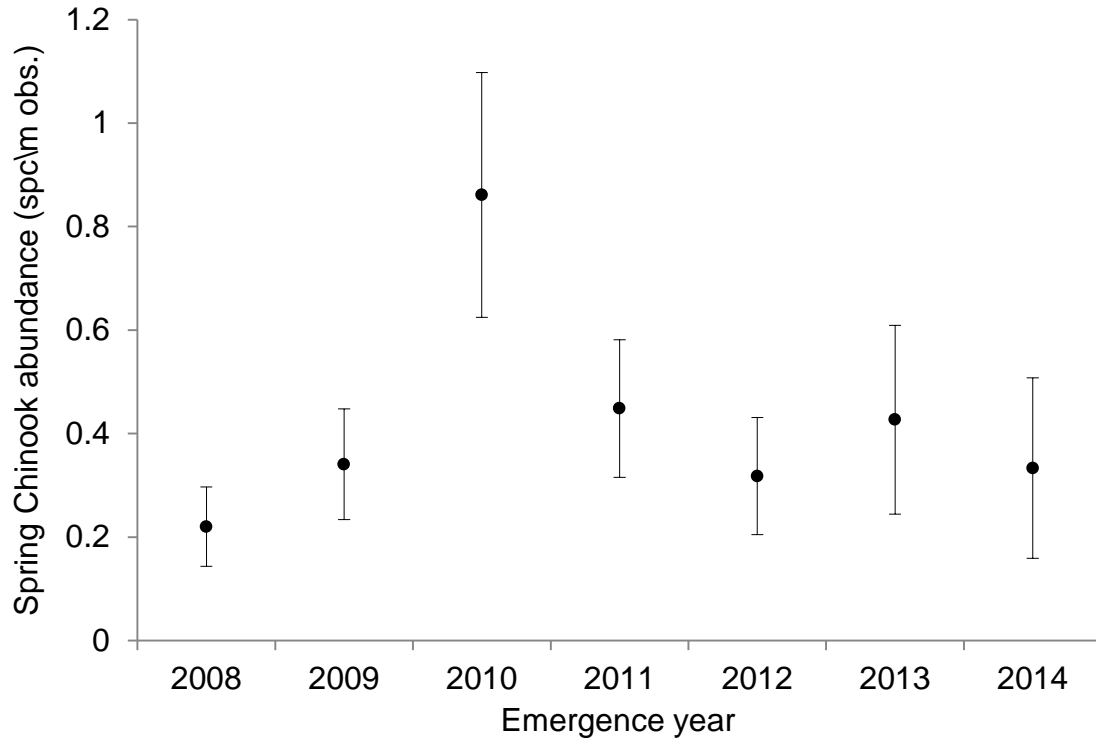


Figure 3. Multi-year analysis of observed abundance among years in two upper Yakima River study reaches with similar temperature and flow characteristics (Easton and Nelson). Significantly greater densities of spring Chinook sub-yearlings were detected in 2010.

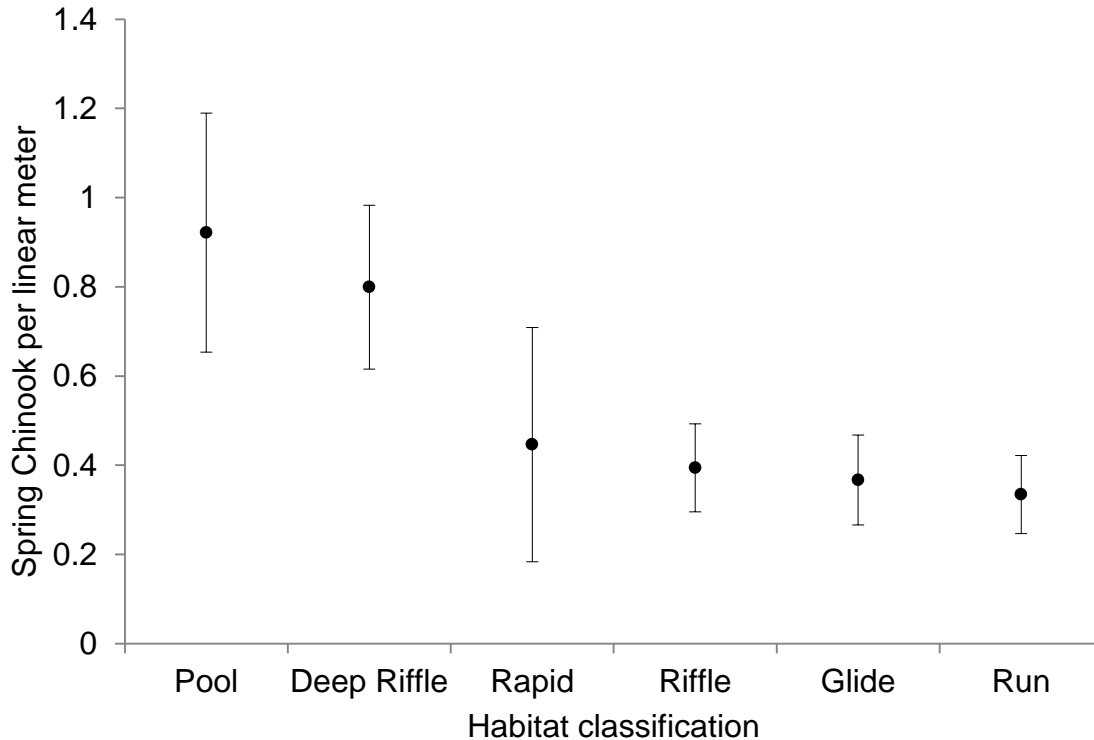


Figure 4. mean spring Chinook observed abundance by habitat type, 2008-2014. Error bars represent 95 percent confidence intervals.

Water temperatures during sampling ranged between 12 and 18.1 degrees Celsius (mean, 15.3; SD, 1.7). Temperatures at sampling were not detectably different between survey reaches (Welch t-test: $t = -0.05$, $df = 108$, $P = 0.96$). Temperature was significantly correlated with observed abundance of spring Chinook, but explained only a small portion of the variation in spring Chinook density ($n = 112$, $R^2 < 0.04$, $P < 0.01$). Visibility while sampling ranged between 0.5 and 3.2 meters (mean, 1.5; SD, 0.5) and was not significantly correlated with estimates of abundance ($R^2 < 0.01$, $P = 0.87$).

Territory size (log transformed) was significantly correlated with fish fork length (mm) ($R^2 = 0.28$, $P < 0.01$; Figure 5). The proportions of feeding strikes were significantly different between categorical distances (1-4 body lengths) from the focal position (Friedman ANOVA: $\chi^2_{3, 694} = 1137.0$; $P < 0.01$; Figure 6). Agonistic strikes were also significantly different between categorical distances (1-4 body lengths) from the focal position (Friedman ANOVA: $\chi^2_{3, 314} = 170.2$, $P < 0.01$; Figure 6). The observed ratios of agonistic to feeding were not detectably different among grouped distances from the focal position (Friedman ANOVA: $\chi^2_{9, 3} = 7.1$, $P = 0.07$; Figure 7).

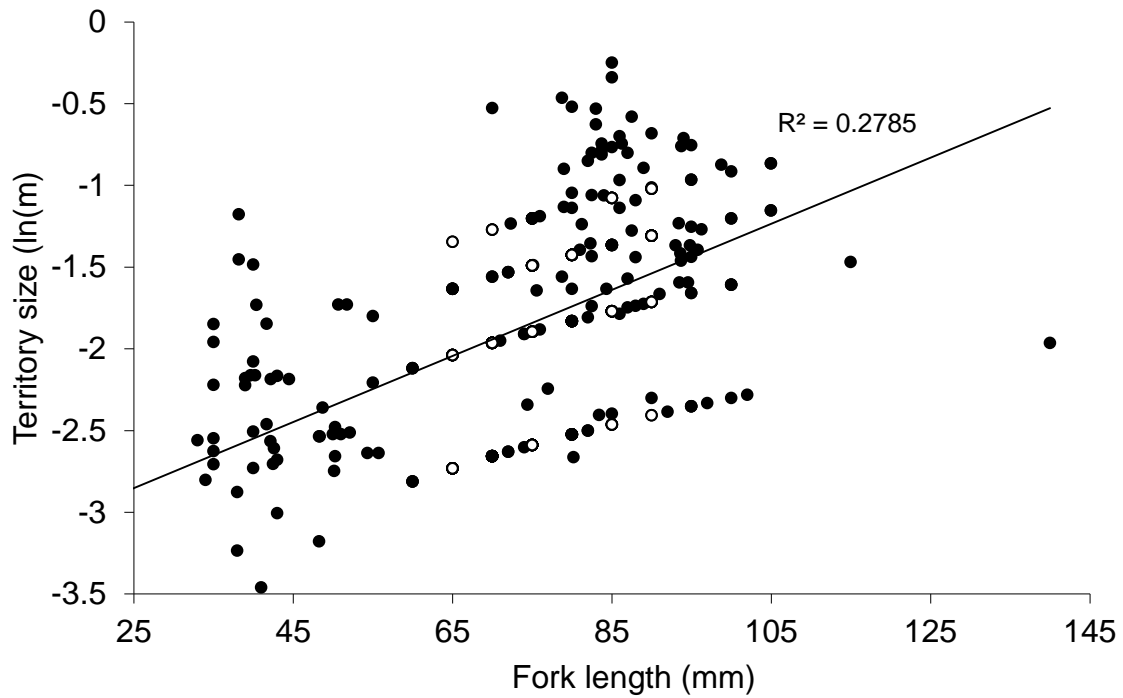


Figure 5. Relationship between spring Chinook fork length (mm) and observed territory size in the spring and summer of 2006-2012 (black points) and 2014 (white points).

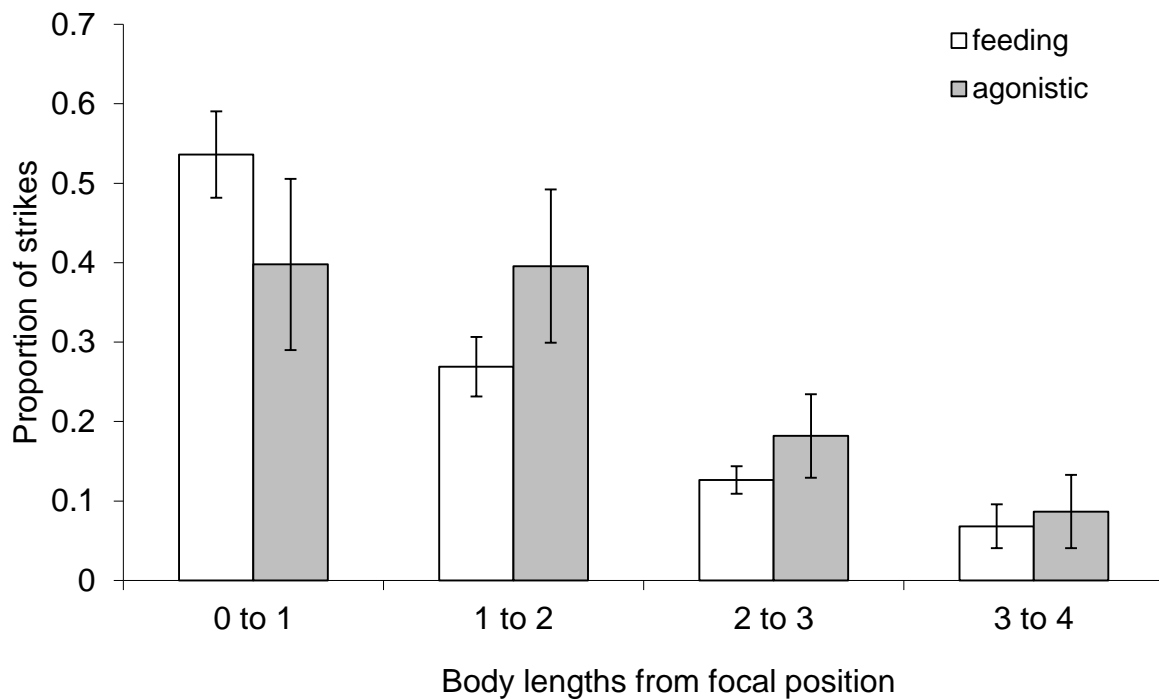


Figure 6. Proportion of rearing spring Chinook feeding and agonistic strikes with increasing distance from the observed focal position in body lengths 2006-2014.

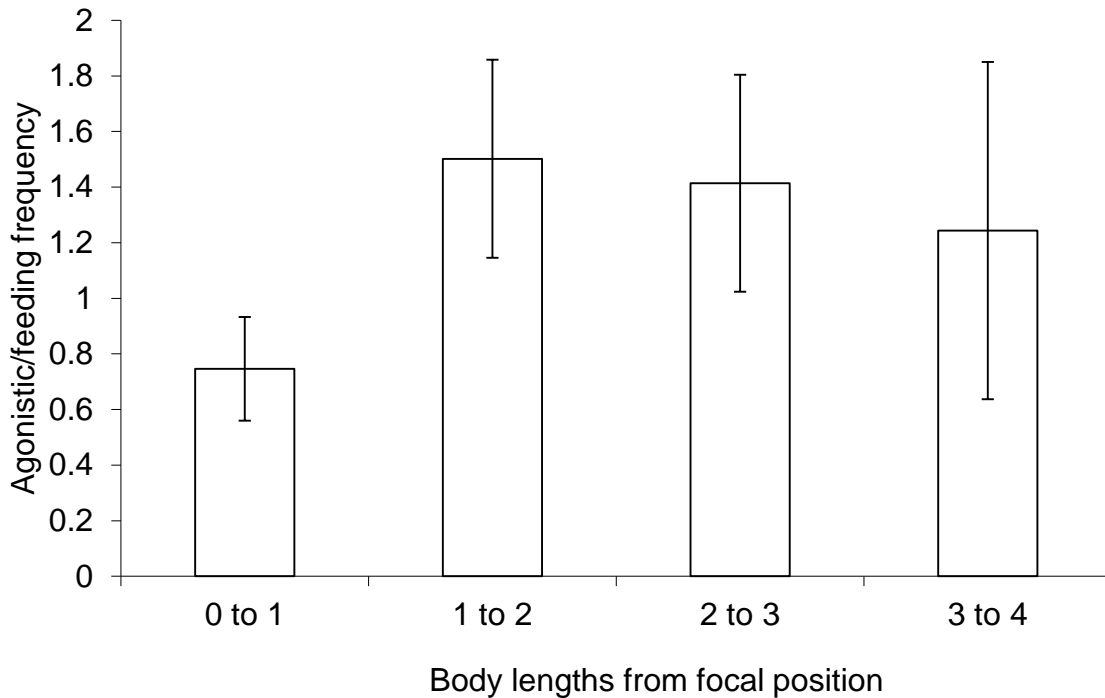


Figure 7. Mean ratio of agonistic strikes per feeding strike with increasing distance from the observed focal position 2006-2014.

Velocities were higher adjacent to spring Chinook focal positions in 72.1 percent of the observations in 2012, 86.0 percent in 2013, and 53.3 percent of the observations in 2014. A summary of microhabitat variable measured around Chinook focal positions is presented in table 3.

Table 3. Summary physical parameters measured at observed spring Chinook focal positions 2012-2014.

	Temp °C	Spc length (mm)	Focal depth (m)	Total depth (m)	Focal velocity (m/s)
2012 (n=111)					
Mean	15.9	69.7	0.3	1.3	0.2
SD	1.0	7.4	1.1	5.7	0.1
2013 (n=43)					
Mean	16.3	79.5	2.3	0.9	0.2
SD	0.7	9.5	9.3	4.2	0.1
2014 (n=60)					
Mean	16.5	78.3	0.24	0.9	0.2
SD	0.8	10.2	0.24	0.3	.01

Discussion

Our data suggest a greater abundance of summer rearing spring Chinook in 2010 than in any other survey year. This is consistent with our detection of greater size and growth in the spring, and also abundance and size in the fall of 2010. These findings, along with a nearly significant correlation between estimates of summer and fall abundance, suggest our methods were successful in tracking relative productivity through three distinct subyearling spring Chinook life-stages. Over time, these data should allow us to identify the life-stage in which limitations to growth and survival are occurring; a critical first step in identifying the specific factor or factors negatively affecting the population in some years.

Yakima River spring Chinook redd-to-parr productivity observed in the fall of 2010 (WDFW unpublished data) was far higher than that predicted by a Beverton-Holt recruitment curve developed using data from the previous sixteen years (Johnson et al 2009). Environmental conditions in the spring of 2010 appear to have been very conducive to early survival, perhaps due to an uncharacteristically low number of high-flow events during the incubation period (Johnson et al 2012). The absence of a detectable response through subsequent life-stages when environmental conditions were not notably different (late spring, summer, and fall), suggests that capacity constraints may exist in earlier developmental periods in years where environmental conditions are more normative.

Documenting the existence of density dependent constraints post-emergence is somewhat confounded by the fact that this is often a period of high mortality, even when spawner densities are low. A system's capacity for incubating alevin is generally far greater than its capacity for juveniles, which generally results in low spawning densities, high survival to emergence, and post-emergent thinning of the population (Quinn 2005). However, during high return years, when competition exists for preferred spawning habitats, density dependent limitations to growth and survival may ultimately occur prior to first emergence. Such limitations may be attributable to a number of potentially limiting environmental factors such as increased sedimentation, scour, temperature, and/or decreased dissolved oxygen levels in less optimal spawning habitats. However, estimates of life-stage specific growth and abundance during years of measurable density dependent response will be necessary to identify limiting factors with any degree of certainty. We will continue to monitor summer parr abundance and to investigate the potential relationship between our summer and fall estimates in 2015.

Perhaps as important as the documentation of abundance in the summer rearing period is the identification of the existing habitats most heavily utilized by sub-yearling Chinook. This information may help in the identification of limiting factors, but will also further our understanding of reach specific productivity in years of low density; a critical metric that is often missing from restoration efforts, which often concentrate only on limitations or "bottlenecks" within the population (Moberg et al. 1997). Although we encountered high variability in abundance among sampling units, we did find higher densities of rearing Chinook in pool and deep riffle type habitats. Therefore, the summer distribution of rearing sub-yearlings appears to be in-part due to the presence of certain habitat types. In addition to other, larger scale, environmental factors which may affect movement and subsequent survival (e.g. temperature, flow events) the use of habitat type as an explanatory variable should be beneficial in determining relative productivity among reaches of the upper Yakima River for summer rearing spring Chinook salmon.

Territory size continued to be strongly associated with spring Chinook length, which is consistent with the findings of others (Grant and Kramer 1990, Keeley and Grant 1995). These data suggest that territory may be a reasonable microhabitat metric to measure the degree of competition for space. Previous work in the Yakima Basin was unsuccessful in linking calculated territory based on local abundance to fall abundance (Pearsons et al. 2007). However, the spatial scale of those measures may have been either too large to detect changes in territorial behavior, or measured after any subsequent mortality or out-migration had already occurred. We will continue to develop the Yakima Basin relationship between Chinook length and territory size in 2015. Chinook decreased the frequency of defense and foraging with increased distance from the holding position. This is consistent with our expectations that increased effort would be required to defend and utilize space away from the position of holding. The frequency of defense may be just as important as the size of the observed territory when evaluating limiting factors. For example, if food is a limiting factor, then we may observe highly defended areas of high food availability and smaller territory size, and areas of low food availability where the individual is forced to defend a larger area. These two scenarios may be energetically equivalent for the individual. We expected larger Chinook would be found in areas with the greatest food availability for the least energetic cost and that higher flow would equate to greater food availability. We did not detect a significant relationship between flow ratio and Chinook length. However, due to the level of variation, larger sample sizes may be required to detect such differences, and will continue to investigate flow ratio as a metric of indexing food availability and energy expenditure at Chinook holding positions. Ranges of focal depth, total depth, and focal velocity during our observations were within the ranges of previous years. We will continue to monitor and refine our set of physical microhabitat measures around the focal position of rearing spring Chinook in order to compare these variables to abundance at the reach scale.

There are some potential biases and limitations to our summer sampling methodologies that will need to be addressed in the future. Foremost among these are the potential for differential efficiency attributable to temperature, and/or limited visibility. Our sampling temperatures were often lower than 14 degrees Celsius; a level at which Hillman et al. (1992) observed a decrease in snorkeling efficiency as high as fifty percent. We detected a small but significant positive correlation between sampling temperature and abundance in instances where temperatures were below 14 degrees Celsius. Explaining approximately five percent of the variation in estimates of observed abundance in the upper Yakima River. These data suggest that temperature will need to be corrected for when comparing observed abundance among reaches in which substantial temperatures differences are present (e.g. the Cle Elum River and the main-stem Yakima River). However, for our current measures of relative abundance, temperature does not appear to be a significant source of potential bias. Temperatures within our sampling reaches were similar throughout the season, therefore any introduced bias attributable to differential sampling efficiency was equally distributed among sampling reaches.

Visibility must also be dealt with through measured efficiency if we wish to sample independently. This is especially true in the early spring, when visibility has been found to be low in some areas (Pearsons et al. 1996; WDFW unpublished data), and may substantially reduce the efficiency of our estimates. We did detect differential visibility among our study reaches, but due to its potential influence in resulting estimates of abundance, will continue to evaluate its potential influence on our estimates.

Acknowledgments

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