Reproductive Ecology of Yakima River Hatchery and Wild Spring Chinook

Yakima/Klickitat Fisheries Project Monitoring and Evaluation

Annual Report 2005 - 2006
May 2006
DOE/BP-00022370-4
This report covers three of many topics under the Yakima/Klickitat Fisheries Project's Monitoring and Evaluation Program (YKFPME) and was completed by Oncorh Consulting as a contract deliverable to the Yakama Nation and Washington Department of Fish and Wildlife. The YKFPME (Project Number 1995-063-25) is funded under two BPA contracts, one for the Yakama Nation (Contract No. 00022449) and the other for the Washington Department of Fish and Wildlife (Contract No. 22370). A comprehensive summary report for all of the monitoring and evaluation topics will be submitted after all of the topical reports are completed. This approach to reporting enhances the ability of people to get the information they want, enhances timely reporting of results, and provides a condensed synthesis of the whole YKFPME.
Reproductive Ecology of
Yakima River
Hatchery and Wild Spring Chinook
Yakima/Klickitat Fisheries Project Monitoring and Evaluation

Annual Report 2005
Performance Period: May, 2005 – April, 2006

Edited by:
Curtis Knudsen

Oncorh Consulting
2623 Galloway SE
Olympia, Washington 98501

Prepared for:
Yakama Nation
P.O. Box 151
Toppenish, WA 98948
Contract No. 00022449

Washington Department of Fish and Wildlife
600 Capitol Way North
Olympia, Washington 98501-1091
Contract No. 22370

U.S. Department of Energy
Bonneville Power Administration
Division of Fish and Wildlife
P.O. Box 3621
Portland, Oregon 97283-3621

Project Number 1995-063-25

May 2006
Executive Summary

This is the fifth in a series of annual reports that address reproductive ecological research and comparisons of hatchery and wild origin spring chinook in the Yakima River basin. This report is organized into three chapters with a general introduction preceding the first chapter. Summaries of each of the chapters in this report are included below.

In chapter 1, we compare upper Yakima River hatchery and natural origin spring chinook salmon returning in 2005 to estimate whether these locally adapted traits are continuing to diverging as we begin the second generation of hatchery returns. The majority of natural origin fish returned at age 4 (89%), age 5 adults made up 4%, and age 3 (jacks) comprised 7% of the returns. Fewer hatchery origin fish returned at age 4 (53%), while 5 adults made up 4% of annual returns. Hatchery origin age 3 adults continued a trend of increasing representation established in first generation hatchery adults, making up 43% of hatchery returns. Mean hatchery body lengths were shorter than natural origin adults (age 3: 4.3 cm; age 4: 1.5 cm; age 5: 0.2 cm), as were body weights (age 3: 0.4 kg; age 4: 0.3 kg; age 5: 0.1 kg) and represented a divergence in body size of up to 1.0 SD. In comparisons between High and Low growth hatchery treatment age 2 (precocious) males, High growth treatment returns (fish reared to significantly larger size at release) were significantly larger than Low growth treatment returns approximately 6 months after release. However, age 3 High and Low growth treatment groups were not significantly approximately 18 months after release. Median passage timing of adult and jack hatchery returns at RAMF was 4 and 2 days later than natural origin adults and jacks, respectively. Jack median passage was 19-25 days later than adults. Mean spawn timing of hatchery fish was significantly earlier than natural fish by 7.5 days. Ultimately the intent is to compare hatchery and natural origin spring Chinook salmon returning between 2005 and 2008 in order to estimate whether trends observed in first generation hatchery returns continue as the project moves into the second generation of returns.

In chapter 2, we estimated the rate of Passive Integrated Transponder (PIT) tag loss in hatchery spring chinook returning 8 months to 4 years after tagging and release using a double-tag design. Hatchery origin fish from broodyears 1997 to 2001 were monitored at Roza Adult Monitoring Facility (RAMF) for the presence of each tag and mark. PIT tag loss estimates varied from 8% to 20% and averaged 16% over broodyears. Using the estimated PIT tag loss rates it was possible to correct the observed recoveries to account for tag loss. Prior to correcting for PIT tag loss, the ratio of PIT tagged fish was significantly lower than the expected PIT tag ratio at release as juveniles ($\chi^2$ p<0.01). After correcting for PIT tag loss, the percentage of PIT tagged fish recovered was lower than expected in 2 of 4 broodyears, but was not significantly lower in any broodyear ($\chi^2$ p>0.05). The difference between the expected and corrected percentage of PIT tagged recoveries represents a difference in survival between the two groups due to the effects of PIT tags. From these preliminary data it does not appear that there is a significant reduction in post-release survival from PIT tags. When significant tag loss occurs, recoveries are underestimated and juvenile-to-adult survival studies will underestimate actual survival rates. Significant tag loss will not invalidate comparisons between similarly PIT tagged treatment and control groups. However, extrapolating tag loss biased survival rates to untagged populations should only be done when the magnitude of the underlying bias is well understood.
In Chapter 3 we compare reproductive traits of upper Yakima River hatchery and wild spring Chinook females to determine whether fitness related traits had diverged after a single generation of artificial propagation. We also compare body size, survival and the proportion of abnormally developing progeny from single-pair matings of hatchery by hatchery and wild by wild adults. We found that fecundity (FEC), relative fecundity (RELFEC), egg weight (EW), and total gamete mass (TGM) were all significantly (p<0.001) correlated with female post-orbital hypural plate (POHP) length, while reproductive effort (RE) was not. It was necessary to analyze age 4 and 5 females separately due to significant (p<0.001) age related differences in their traits. In ANCOVAs testing FEC, RELFEC, EW, and TGM distributions for an Origin (hatchery/wild) effect and adjusting for POHP, no significant differences were found between hatchery and wild females. However, using published data showing that upper Yakima River hatchery spring chinook were significantly smaller than wild females and our trait/POHP regressions, we calculated age and broodyear specific mean reproductive trait values. Over the four broodyears examined, wild females averaged 7-9% greater TGM, 1-2% heavier EW, 6-7% greater FEC, and 1% lower RELFEC than hatchery females. No significant Origin effect was found in RE. The fundamental reproductive trait/body size relationships had not been significantly altered by a single generation of hatchery exposure. However, due to significantly smaller hatchery mean body size which is correlated with the reproductive traits, returning hatchery and wild female’s reproductive traits did differ, likely resulting in some loss of fitness in hatchery females. We found no Origin effect in the proportion of abnormally developing fry. After adjusting for egg size, we found that hatchery fry were on average ~1% heavier than wild fry. Comparisons of fry survival were mixed with no clear trend in survival of hatchery and wild eggs to the emergent fry stage.

All findings in this report should be considered preliminary and subject to further revision unless previously published in a peer-reviewed technical journal.
Table of Contents

Executive Summary........................................................................................................................................ iv

Table of Contents....................................................................................................................................... vi

General Introduction ...................................................................................................................................... 1

Chapter 1. A Comparison of Life-History Traits in Hatchery and Natural origin Upper Yakima River Spring Chinook Salmon Returning in 2005.............................................................. 3

Chapter 2. Long Term Loss and Survival Effects of PIT Tags In Yakima River Hatchery Spring Chinook Salmon 2005.......................................................................................................... 15

Chapter 3. Comparison of Gametic Traits of First Generation Hatchery and Wild Upper Yakima River Spring Chinook Salmon...................................................................................................... 28
General Introduction

This report is intended to satisfy two concurrent needs: 1) provide a contract deliverable from Oncorh Consulting to the Yakama Nation and Washington Department of Fish and Wildlife, with emphasis on identification of salient results of value to ongoing Yakima/Klickitat Fisheries Project (YKFP) planning and 2) summarize results of research that have broader scientific relevance. This is the fifth in a series of reports that address reproductive ecological research and monitoring of spring chinook populations in the Yakima River basin. This annual report summarizes data collected between April 1, 2005 and March 31, 2006 and includes analyses of historical baseline data, as well.

Supplementation success in the Yakima Klickitat Fishery Project’s (YKFP) spring chinook (*Oncorhynchus tshawytscha*) program is defined as increasing natural production and harvest opportunities, while keeping adverse ecological interactions and genetic impacts within acceptable bounds (Busack et al. 1997). Within this context demographics, phenotypic traits, and reproductive ecology have significance because they directly affect natural productivity. In addition, significant changes in locally adapted traits due to hatchery influence, i.e. domestication, would likely be maladaptive resulting in reduced population productivity and fitness (Taylor 1991; Hard 1995). Thus, there is a need to study demographic and phenotypic traits in the YKFP in order to understand hatchery and wild population productivity, reproductive ecology, and the effects of domestication (Busack et al. 1997). Tracking trends in these traits over time is also a critical aspect of domestication monitoring (Busack et al. 2004) to determine whether trait changes have a genetic component and, if so, are they within acceptable limits.

The first chapter of this report compares hatchery and wild upper Yakima River spring chinook adults from 2005, the first age 4 returns from the second hatchery generation, over a suite of life-history, phenotypic and demographic traits. The second chapter estimates juvenile-to-adult PIT tag loss and assesses the effects of PIT tags on post-release survival. The third chapter compares reproductive traits and progeny produced by upper Yakima River wild and hatchery origin fish during the first generation of hatchery returns representing broodyears 1997 to 2000.

The chapters in this report are in various stages of development. Chapters One is a progress report. Chapters Two and Three will eventually be submitted for peer reviewed publication. Readers are cautioned that any preliminary conclusions are subject to future revision as more data and analytical results become available.

Acknowledgments

We would like to thank Bonneville Power Administration for financially supporting this work. In addition, we could not have completed this work without the help and support of many individuals during 2005/2006. We have tried to recognize each of them either on title pages or in acknowledgments within each chapter of this report.
References


Chapter One

A Comparison of Life-History Traits in Hatchery- and Natural-origin Upper Yakima River Spring Chinook Salmon Returning in 2005

Prepared by:

Curtis M. Knudsen¹, Steve L. Schroder², Mark V. Johnston³,
Craig Busack², Todd N. Pearsons², and David E. Fast⁴

¹Oncorh Consulting, 2623 Galloway SE, Olympia, WA 98501
²Washington Department of Fish and Wildlife, 600 Capitol Way North, Olympia, WA 98501-1091
³Yakama Nation, P.O. Box 151, Toppenish, WA 98948
Abstract

Age composition, passage timing at Roza Adult Monitoring Facility (RAMF), and spawn timing of the hatchery and natural origin adult spring Chinook salmon returning to the upper Yakima River in 2005 were compared. In 2005, the majority of natural origin fish returned at age 4 (89%), age 5 adults made up 4%, and age 3 (jacks) comprised 7% of the returns. Fewer hatchery origin fish returned at age 4 (53%), while 5 adults made up 4% of annual returns. Hatchery origin age 3 adults continued a trend of increasing representation established in first generation hatchery adults, making up 43% of hatchery returns. Mean hatchery body lengths were shorter than natural origin adults (age 3: 4.3 cm; age 4: 1.5 cm; age 5: 0.2 cm), as were body weights (age 3: 0.4 kg; age 4: 0.3 kg; age 5: 0.1 kg) and represented a divergence in body size of up to 1.0 SD. In comparisons between High and Low growth hatchery treatment age 2 (precocious) males, High growth treatment returns (fish reared to significantly larger size at release) were significantly larger than Low growth treatment returns approximately 6 months after release. However, age 3 High and Low growth treatment groups were not significantly approximately 18 months after release. Median passage timing of adult and jack hatchery returns at RAMF was 4 and 2 days later than natural origin adults and jacks, respectively. Jack median passage was 19-25 days later than adults. Mean spawn timing of hatchery fish was significantly earlier than natural fish by 7.5 days. The present analyses focused on comparisons within 2005 returns. Ultimately the intent is to compare hatchery and natural origin spring Chinook salmon returning between 2005 and 2008 in order to estimate whether trends observed in first generation hatchery returns continue as the project moves into the second generation of returns.

These data should be considered preliminary until published in a peer-reviewed journal.
Introduction

Life-history traits reflect local adaptations affecting population productivity and individual fitness (Stearns 1976; Roff 1992; Brannon et al. 2004). Changes in demographic or life history traits, such as a reduction in age classes or skewed sex ratio, can reduce phenotypic variation, affect total annual egg production, and effective population size (Nunney 1991; Waples 2002). Moreover, changes in adult spawn timing may reduce fitness by shifting fry emergence timing outside a locally adapted temporal window (Brannon 1987; Smoker et al. 1998; Einum and Fleming 2000; Brannon et al. 2004). In general, significant changes in locally adapted life-history traits will be maladaptive in the wild (Lynch and O’Hely 2001; Ford 2002; Goodman 2004, 2005), leading to reduced individual reproductive success (Taylor 1991; Fleming and Gross 1993; Fleming et al. 2000) and possibly resulting in lower productivity of a naturally spawning population. Monitoring life-history traits of hatchery populations to determine if they are diverging from their native population’s distributions is a necessary part of a hatchery monitoring plan (Hard 1995; Goodman 2005). Significant differences may indicate that the artificial rearing environment is causing genetic divergence to occur between the two groups. However, phenotypic changes alone are not sufficient to conclude that genotypic divergence has occurred. To do that, fish from both groups should be spawned, incubated, and reared in a common environment. Observed differences under these circumstances would represent genetic change.

This report is a continuation of work described by Knudsen et al. (2006) who compared first-generation hatchery and wild returns of upper Yakima hatchery spring Chinook. However, the present analyses cover fish returning in 2005; the first large contingent of the second generation of hatchery returns. Knudsen et al. (in press) found that most hatchery and natural origin fish reached maturity at age 4 (>76%) followed in magnitude by ages 3 and 5. Wild mean age-at-maturation demonstrated no significant trend over time, while hatchery mean age-at-maturation was declining (p=0.05). Mean lengths of 3, 4, and 5-yr-old hatchery fish were shorter than those of wild fish of the same age (age 3: Δ2.7 cm; age 4: Δ1.7 cm; age 5: Δ2.7 cm). Likewise, body weights of hatchery fish were less than those of wild fish (age 3: Δ0.3 kg; age 4: Δ0.3 kg; age 5: Δ0.8 kg) representing a change in body size of between 0.5 and 1.0 SD. Median arrival timing of hatchery and wild fish at RAMF showed no consistent difference. However, median arrival date of age 3 fish was 19-20 days later than for ages 4 and 5 (p<0.01). Mean spawn timing of hatchery fish was significantly earlier than wild fish by 5.1 days in a “common garden” experiment (p<0.05). The degree of genetic determination of the divergence is unknown, but future monitoring through the YKFP’s Domestication Monitoring Study will help clarify this. The present analyses focus on comparisons within 2005 adult returns. Detailed analyses of broodyear specific comparisons and trends will be finished after the completion of the second generation of hatchery returns in 2008.

Methods

Study Population

The Yakima River is a tributary to the Columbia River and contains three genetically distinct, geographically separated wild spring Chinook populations (Busack
and Marshall 1991; Young 2004). The upper Yakima River population spawns primarily upstream of Roza Dam (rkm 206), an irrigation diversion dam through which all upstream migrating fish from this population must pass (Figure 1). The other two populations are located in the Naches system: the American River (a tributary of the Naches River) and the Naches River and its tributaries, excluding the American River.

The Yakima/Klickitat Fishery Project (YKFP) began operation of the CESRF spring Chinook hatchery near Cle Elum on the upper Yakima (rkm 290; Figure 1) in 1997. Broodstock are collected at RAMF, located adjacent to and upstream of Roza Dam, as spring Chinook pass upstream between April and September (Knudsen at al. 2006). Between 1997 and 2001, broodstock were exclusively of wild origin. Beginning in 2002, we established the Hatchery Control (HC) line as part of the Domestication Study (see Busack et al. (2004) for a detailed description) and began taking hatchery origin fish to use exclusively for that purpose. The first age 3 Hatchery Control adults returned in 2005. In addition, the 2005 age 4 adults were produced at least in part from the first generation of age 4 hatchery origin adults spawning in 2001. The progeny produced from those naturally spawning hatchery and wild adults can no longer be

Figure 1. Yakima River basin showing the upper Yakima River, Roza Adult Monitoring Facility (RAMF), the Cle Elum Supplementation Research Facility (CESRF), acclimation sites, Naches River and American River.
considered purely wild in origin (see discussion in Knudsen et al. in press), as it is highly likely that hatchery origin adults in broodyear 2001 successfully spawned, and thus we are calling them natural Origin Recruits (NOR).

HC and NOR broodstock are transferred to CESRF and held together in one concrete raceway under the same water temperature, flow and rearing densities, until mature. Details of the broodstock collection process are given in Knudsen et al. (in press). Briefly, a fixed proportion of the total broodstock is collected each week over the entire run based on weekly mean historical passage proportions at RAMF. Broodstock collection is limited to no more than 50% of the NOR population passing during any week and all returning HC adults are collected at RAMF and either used as broodstock, as experimental subjects, or contribute toward the YN tribal subsistence fishery. After reaching maturity, HC and NOR fish are spawned separately in either 3x3 or 2x2 factorial matings in order to increase effective population size (Fiumera et al. 2004) and maintain genetic diversity.

All returning fish passing through RAMF can be enumerated and sampled, if desired. To facilitate collection and identification of broodstock origin as well as other post-release monitoring, all hatchery releases are adipose fin clipped and tagged. A subset of 40,000 fish are PIT tagged. The remaining production are marked with a combination of colored elastomer in the adipose eyelid and a coded-wire tag in a specific body site that allows identification of HC using a handheld CWT detector (to identify body tag location) and visual identification of the elastomer color.

**Age Composition**

Estimates of age composition for hatchery (n=327) and natural (n=526) origin fish returning in 2005 were made. The most appropriate way to calculate and compare age and sex composition is on a broodyear basis, rather than return year, in order to avoid the problem of unequal cohort strengths overwhelming within-cohort trends. However, in this annual report we do not make comprehensive comparisons across broodyears. Statistical analyses of these data on a broodyear basis will occur in future reports.

Age composition of natural origin age 4 and 5 adults was estimated from fish collected at RAMF and taken to CESRF. This includes all fish selected for broodstock and other experimental needs. Hatchery origin age composition of age 4 and 5 adults was estimated from fish sampled at RAMF. Some of these fish were taken to CESRF and used as broodstock for the Hatchery Control line. However most hatchery origin adults sampled at RAMF were released upstream of RAMF and allowed to naturally spawn. Details of the sampling methodology used at RAMF are given in Knudsen et al. (in press). Briefly, a fixed number of age 3 and older hatchery and natural origin adults, based on broodstock and experimental needs, are collected from throughout the run in proportion to the historic run timing at RAMF. No more than half the natural run is taken.

On a daily basis all hatchery fish passing RAMF were enumerated, anesthetized and examined for marks, classified as either an age 3 jack or an older adult (age 4 or 5) based on body size, and systematically scale sampled (~1-in-5 hatchery fish over the run). Also, 75 additional hatchery fish collected for broodstock and other experimental purposes were scale sampled. All scale sampled fish were measured for post-orbital hypural plate (POHP) length, fork length, body weight, and passage date recorded. Fish were held briefly to recover from the anesthetic and released back into the river to
complete their spawning migration. Hatchery origin age composition of age 4 and 5 adults was estimated from the RAMF systematic sample of scales. Two scale analysts independently aged all scales and resolved disagreements. Ages were designated as the number of years from the year of conception (broodyear) to return year. Thus, a fish produced from parents spawning in the fall of 2000 and returning in 2005 was age 5. Under this convention, precocious males (nonanadromous males maturing in their first [natural only] or second [natural and hatchery] year) are designated age 1 and age 2, respectively (see Larsen et al. (2004) and Pearsons et al. (2004) for a full description of natural and hatchery precocious male production in the upper Yakima River). Returning spring Chinook in the Yakima River are greater than 99% yearling outmigrants based on adult return scales (J. Sneva, WDFW, personal communication).

Natural and hatchery origin age 3 jack returns are identified visually based on the significant body size differences between age 3 and age 4 fish and the presence or absence of an adipose fin as fish pass RAMF. The daily passage numbers of age 3 jacks and age 4 and 5 adults combined at RAMF were used to represent run timing.

Size-at-Age

The natural origin sample consisted of fish measured at RAMF and brought up to CESRF as broodstock and for other experimental purposes. The hatchery origin sample consisted primarily of the RAMF systematically sampled and released hatchery adults with the additional fish selected for broodstock and experimental purposes. Length and weight data collected at RAMF, prior to fish reaching full maturity, were used to compare hatchery and natural size-at-age distributions using a 2-way ANOVA (Origin x Age main effects). Body size data were loge transformed prior to analysis. RAMF body weights are significantly heavier than body weights of the same fish at full maturity 1 to 5 months later (Knudsen et al. 2004), and this should be kept in mind when making comparisons between our data and others’.

Beginning with the 2002 broodyear, a precocious male minimization study was implemented at CESRF. This study manipulated juvenile growth trajectories in order to reduce the rate of precocious male production. Half of the raceways were placed on a High growth treatment and half on a Low growth treatment resulting in differences in size-at-release (Figure 2). Age 2 fish from broodyear 2003 (High n= 78; Low n= 48 ) and age 3 (High n= 172; Low n= 289) from broodyear 2002 were recovered in 2005. We compared the lengths and weights of fish from the two treatment groups to determine whether the size differences at release persisted after 6 and 18 months post-release growth. A 2-way ANOVA was used testing for Acclimation site (Easton/Clark Flats/Jack Creek) and Treatment (High/Low) effects.
Figure 2. Growth in fork length (FL) and body weight (Weight) during rearing of 2002 and 2003 broodyear juveniles from the High and Low growth treatment groups.

Passage and Spawn Timing

Passage timing distributions of hatchery and natural origin fish at RAMF were compared using a Kruskal-Wallis non-parametric ANOVA (KW test; Zar 1999) because of the highly skewed temporal distributions. Because of significant differences in passage timing of age 3 and age 4 and 5 adults (Knudsen et al. 2004), we made comparisons between hatchery and natural passage timing for each age group (adult vs jack) separately.

Artificial spawning occurs at CESRF over a five-to-six week period from early September through early October and hatchery (n=72) and natural (n=388) spawn timing
distributions were compared with a 2-way ANOVA testing for Origin (Hatchery/Natural) and Age (3/4/5) effects. Spawn timing distributions are not skewed like the RAMF passage timing distributions and were therefore not transformed. All passage and spawning dates were converted to ordinal dates (day-of-year).

Results

Age Composition

The majority of both hatchery and natural origin fish returned at age 4 (Table 1), although a considerably higher proportion of natural origin adults were made up of 4 year olds than hatchery origin adults. Age 5’s were represented by 7 and 4% hatchery and natural adults, respectively. The proportion of hatchery age 3 adults was over 10 times the proportion of natural origin age 3 adults. This disparity in the proportion of age 3 returns is similar to the trend noted by Knudsen et al. (in press), although Knudsen et al.’s analyses were based on broodyears, rather than return year.

Size-at-age

Mean POHP lengths and body weights of hatchery and natural origin returns by age are given in Table 2 along with sample sizes and standard deviations. On average, hatchery age 3 fish were 2.7 cm and 0.3 kg smaller than natural age 3 fish. Natural origin age 4 and age 5 adult’s body sizes were also greater on average than hatchery origin body sizes (age 4 mean difference: POHP= 1.6 cm, Body wt.=0.3 kg; Age 5 mean difference: POHP=0.6 cm, Body wt.= 0.2 kg). Two-way ANOVAs (Origin x Age effects) of loge transformed POHP and body weight distributions showed that hatchery and natural origin

<table>
<thead>
<tr>
<th>Age</th>
<th>Origin</th>
<th>POHP (sd)</th>
<th>BW (sd)</th>
<th>N</th>
</tr>
</thead>
<tbody>
<tr>
<td>3</td>
<td>Hatchery</td>
<td>40.3 (4.1)</td>
<td>1.3 (0.4)</td>
<td>98</td>
</tr>
<tr>
<td></td>
<td>Natural</td>
<td>43.0 (4.0)</td>
<td>1.6 (0.4)</td>
<td>35</td>
</tr>
<tr>
<td>4</td>
<td>Hatchery</td>
<td>59.1 (3.8)</td>
<td>4.0 (0.8)</td>
<td>219</td>
</tr>
<tr>
<td></td>
<td>Natural</td>
<td>60.7 (3.6)</td>
<td>4.3 (0.8)</td>
<td>440</td>
</tr>
<tr>
<td>5</td>
<td>Hatchery</td>
<td>70.0 (6.3)</td>
<td>6.4 (1.4)</td>
<td>18</td>
</tr>
<tr>
<td></td>
<td>Natural</td>
<td>70.6 (4.0)</td>
<td>6.5 (2.0)</td>
<td>12</td>
</tr>
</tbody>
</table>
body size differences were significant (Origin \( p \leq 0.02 \)). As would be expected, age 3 size < age 4 size < age 5 size (Age effect \( p < 0.0001 \)). Origin*Age interactions were not significant \( (p > 0.10) \).

In comparisons of hatchery age 2 High and Low growth treatment groups, we found that age 2 hatchery returns from the High growth treatment were significantly longer \( (p = 0.002) \) and heavier \( (p = 0.012) \) than the Low growth group after approximately 6 months of post-release rearing. On average, the High growth fish were 15.9 cm in length and 9.0 g body weight (BW), while the Low growth group means were 15.3 cm and 8.4 g. There was no significant Acclimation site effect \( (p > 0.80) \) or interaction effect \( (p > 0.13) \).

This is in sharp contrast to hatchery age 3 fish in which there were no significant Treatment effects \( (p > 0.73) \) and High (mean POHP = 40.5 cm; BW = 1.3 kg) and Low (mean POHP = 40.5 cm; BW = 1.3 kg) body sizes were identical. Again, Acclimation site \( (p > 0.80) \) and interaction \( (p > 0.170) \) effects were not significant. Thus, after approximately 18 months of post-release rearing the body sizes of the High and Low treatment groups did not differ.

**Passage and Spawn Timing**

Age 3 jack passage at RAMF differed significantly from adult passage timing \( (KW \text{ test } p < 0.0001) \) with hatchery and natural jack median dates being 19 and 25 days later than adults, respectively (Table 3). For this reason, we compared hatchery and natural passage timing for adults and jacks separately. Natural adults median passage date was significantly earlier than hatchery adults by 4 days \( (KW \text{ test } p < 0.0001) \), while natural jacks passed 2 days later than hatchery jacks \( (KW \text{ test } p < 0.0001) \).

**Table 3. Median 2005 passage timing at RAMF by Type: Jack (age 3) or Adult (ages 4 and 5 combined). Sample sizes (n) are total Adult and Jack run sizes passing RAMF.**

<table>
<thead>
<tr>
<th>Origin</th>
<th>Type</th>
<th>Median</th>
<th>n</th>
</tr>
</thead>
<tbody>
<tr>
<td>Natural</td>
<td>Jack</td>
<td>168.0</td>
<td>211</td>
</tr>
<tr>
<td></td>
<td>Adult</td>
<td>143.0</td>
<td>4875</td>
</tr>
<tr>
<td>Hatchery</td>
<td>Jack</td>
<td>166.0</td>
<td>541</td>
</tr>
<tr>
<td></td>
<td>Adult</td>
<td>147.0</td>
<td>726</td>
</tr>
</tbody>
</table>

In 2005, Hatchery fish mean spawning date (Sept. 17) was 7.5 days earlier and was significantly different \( (p < 0.0001) \) than natural fish (Sept. 26). There was no significant Age \( (p = 0.293) \) or interaction effect \( (p = 0.663) \).

**Discussion**

Hatchery origin returns in 2005, the first adult returns from the second generation of CESRF hatchery production, continued the same trends documented by Knudsen et al. \( (\text{in press}) \) in first generation hatchery returns. Natural origin fish returned in 2005 at ages comparable to historical wild origin proportions made up primarily of age 4 (89%), age 5 (4%), and age 3 (7%) adults. Hatchery origin fish continued the trend of fewer age 4 returns (53%), a much larger proportion of age 3 adults (43%), and a comparable portion of age 5 adults (4%). Mean hatchery body lengths were shorter than natural origin adults (age 3: 4.3 cm; age 4: 1.5 cm; age 5: 0.2 cm), as were body weights (age 3: 0.4 kg; age 4:
0.3 kg; age 5: 0.1 kg) and represented a divergence in body size of up to 1.0 SD. In comparisons between High and Low growth hatchery treatment age 2 (precocious) males, High growth treatment returns (fish reared to significantly larger size at juvenile release) were significantly larger than Low growth treatment returns approximately 6 months after release. However, age 3 High and Low growth treatment groups were equal in body size after over 18 months post-release growth. Median passage timing of adult and jack hatchery returns at RAMF was 4 and 2 days later than natural origin adults and jacks, respectively. Jack median passage was 19-25 days later than adults. Mean spawn timing of hatchery fish was significantly earlier than natural fish by 7.5 days in 2005 repeating a trend observed since the first hatchery origin age 4 adults were artificially spawned at CESRF in 2001.

These analyses examined age composition, passage timing at Roza Adult Monitoring Facility (RAMF), and spawn timing of the hatchery and natural origin adult spring Chinook salmon returning to the upper Yakima River in 2005. This information will be used in a more comprehensive future report comparing hatchery and natural origin spring Chinook salmon in order to estimate whether the trends observed in first generation hatchery returns continue throughout the second generation. Detailed analyses of broodyear specific comparisons and trends will be finished after the completion of the second generation of hatchery returns in 2008.

**Acknowledgements**

We wish to thank the Yakama Nation Roza Adult Monitoring Facility, spawner survey personnel, and the Cle Elum Supplementation and Research Facility personnel. Paul Huffman and Bill Bosch (YN) participated in sampling broodstock at CESRF. Bill Bosch also provided valuable help in data management. T. Swan (YN) and J. Sneva (WDFW) aged all the scale samples. John Easterbrooks (WDFW) and Mel Sampson (YN) provided policy support and the Bonneville Power Administration (BPA) provided funding to the YKFP.

**References**


Chapter Two

Long Term Loss and Survival Effects of PIT Tags in Yakima River Hatchery Spring Chinook Salmon

Submitted by

Curtis M. Knudsen¹, Mark V. Johnston²,
Steve L. Schroder³ and Charlie R. Strom⁴

¹Oncorh Consulting, 2623 Galloway SE, Olympia, WA 98501
²Yakama Nation, P.O. Box 151, Toppenish, WA 98948
³Washington Department of Fish and Wildlife, 600 Capitol Way North, Olympia, WA 98501
⁴Yakama Nation, Cle Elum Supplementation Research Facility, Cle Elum, WA 98922
Abstract

We estimated the rate of Passive Integrated Transponder (PIT) tag loss in hatchery spring chinook returning 8 months to 4 years after tagging and release. Annually from 1998 to 2002, approximately 40,000 juveniles were tagged with a PIT tag, Coded-wire Tag (CWT) inserted into the snout, and an adipose fin clip. The double-tag design allowed estimation of the rate of PIT tag loss by broodyear. Hatchery origin fish from broodyears 1997 to 2001 were monitored at Roza Adult Monitoring Facility (RAMF) for the presence of each tag and mark. For this annual report we present retention rates for broodyears 1997 to 2000. Returns in 2005, representing age 5 BY 2000 and age 4 BY 2001 have not yet been included in the database. Results for broodyears 1997 to 2001 will be included in a comprehensive report to be submitted for peer-reviewed publication in 2006.

PIT tag loss estimates varied from 8% to 20% and averaged 16% over broodyears. Using the estimated PIT tag loss rates it was possible to correct the observed recoveries to account for tag loss. Prior to correcting for PIT tag loss, the ratio of adult PIT tagged fish was significantly lower on average by 20% than the expected tag ratio at release ($\chi^2$ $p<0.01$). After correcting for PIT tag loss, the percentage of PIT tagged fish recovered was lower than expected in 2 of 4 broodyears, but was not significantly lower in any broodyear ($\chi^2$ $p>0.05$). The difference between the expected and corrected percentage of PIT tagged recoveries represents a difference in survival between the two groups due to the effects of PIT tags. From these preliminary data it does not appear that there is a significant reduction in post-release survival from PIT tags.

When significant tag loss occurs, recoveries are underestimated and juvenile-to-adult survival studies will underestimate actual survival rates. Significant tag loss will not invalidate comparisons between similarly PIT tagged treatment and control groups. However, extrapolating tag loss biased survival rates to untagged populations should only be done when the magnitude of the underlying bias is well understood.

These data should be considered preliminary until published in a peer-reviewed journal.
Introduction

The Passive-Integrated-Transponder (PIT) tag (Prentice et al. 1990) has been used extensively in the Columbia River basin to monitor juvenile salmonid survival and migration timing (Budy et al. 2002; Berggren et al. 2003; Berggren et al. 2005). PIT tags are also an integral part of the Monitoring and Evaluation Program of the Yakima/Klickitat Fishery Project (Busack et al. 1997; Neeley 2004). As part of that effort, approximately 40,000 PIT tagged juvenile spring chinook have been released annually, primarily to estimate juvenile in-river relative survival. In addition, PIT tags are used to monitor smolt migration timing, movement of fish volitionally leaving Yakima/Klickitat Fishery Project (YKFP) acclimation sites, and to estimate smolt entrainment/flow diversion relationships at Chandler Juvenile Fish Passage Facility (Sampson and Fast 2000; Neeley 2004).

Recently, analysis of PIT tagged salmonids was been used to examine impacts of flow on survival in the Columbia River system (Berggren et al. 2005) and extended their results to untagged portions of study populations. However, the Independent Science Advisory Board (ISAB) noted, “More attention should be given by the CSS and the region as a whole to the apparent documentation that PIT-tagged fish do not survive as well as untagged fish. This point has major implications for all uses of PIT-tagged fish as surrogates for untagged fish” (ISAB 2006). The noted apparent difference in survival between PIT tagged and untagged fish could be due to either a real survival difference or to PIT tag loss (tagged fish missing tags would be included as mortalities) or a combination of both.

It is important to understand the strengths and limitations of any tagging technique in order to select the tag violating the fewest or most important assumptions in the proposed research (Seber 1982; Krebs 1998). A well designed study will at a minimum rear a sample of marked fish for at least a few days to a few weeks post-marking in order to document immediate short term mortality and tag loss. Typically, salmonid PIT tagging in the Columbia River utilize skilled tagging crews and experience immediate short-term mortality and tag loss rates of less than 1-2% (Prentice et al. 1993). This short term PIT tag loss is often a function of tagging quality, particularly the insertion wound in the ceolomic cavity, and tag loss usually occurs within a few days after tagging until the wound heal. Fish health can also delay healing and result in higher than expected short term tag loss. However, estimating post-release tag loss occurring one or more years after release is a more difficult problem. In order to estimate this long term tag loss, rather than holding fish in a protected vessel buffered from the vicissitudes of a free ranging life, it is important that the fish experience the types of stresses and challenges all study fish would experience. For this purpose, Beverton and Holt (1957) and Seber (1982) suggest using a double-tagging design and releasing fish under actual study conditions allowing estimation of tag loss for each tag type after release.

Prentice et al. (1994) used a double-tag study design to estimate PIT tag loss in the only published report we are aware of on juvenile-to-adult PIT tag retention in free ranging adult Pacific salmon. They found that prior to spawning, mature adult coho salmon (*Oncorhynchus kisutch*) PIT tagged as juveniles shed their tags at a high rates: overall 59% loss in females and 13% loss in males. PIT tag loss in these study coho was estimated by Prentice et al. (1993) eight months after tagging to be 1%. Thus, essentially
all of the tag loss occurred sometime during the next 12 months prior to spawning. Prentice et al. (1994) collected five weekly samples of PIT tagged adults as they reached full maturity and their data demonstrate no clear temporal trend in PIT tag loss (Figure 1). However, the sample sizes of the later collections are very small resulting in nearly 0-100% confidence intervals. They do point out the potential for males and females to experience different loss rates, however.

![Graph showing PIT tag loss over time](image)

**Figure 1.** The proportion of A) female and B) male PIT tags lost over time in Skagit Hatchery adult coho salmon (adapted from Figure 15 in Prentice et al. (1994)). Weekly proportions with 95% binomial confidence intervals are shown with sample sizes in parentheses.

YKFP research objectives require recovering hatchery returns throughout their life history, including both pre- and post-spawning. Our goals in this study were to estimate both PIT tag and CWT loss in upper Yakima River hatchery spring chinook returns 1 to 4 years after tagging. In addition, we estimated whether PIT tags had a negative impact on survival by correcting recoveries for PIT tag loss and comparing that to the expected percentages of PIT tagged recoveries based on juvenile releases.

**Methods and Materials**

We used a double-tag study design (Seber 1982) that allows estimation of tag loss rates for PIT tags and CWT’s, as well as estimation of the number of fish losing both
Beginning in 1998, approximately 40,000 age-0 hatchery origin spring chinook were triple marked annually between October and December with 1) a PIT tag injected into the body cavity with the hand-held injector technique (Prentice et al. 1990), 2) a CWT injected into the snout (Jefferts et al. 1963), and 3) an adipose fin clip. An equal proportion of each of raceway was PIT tagged and the PIT tagged and non-PIT tagged fish were reared together in a common environment. In February, fish were transferred by truck to two or three acclimation sites depending on the broodyear, held for approximately 1.5 additional months, and then allowed to volitionally emigrate as age-2 smolts between March 15 and May 30. PIT tag and non-PIT tagged mortalities recovered during rearing were documented and removed from the population. The other non-PIT tagged fish released from the Cle Elum Supplementation and Research Facility (CESRF) were also marked with a combination of a colored elastomer material in the adipose eyelid, a CWT imbedded in the musculature at one of five specific body sites, and an adipose fin clip. Thus, all hatchery fish were marked with an adipose fin clip allowing identification and enumerate of all hatchery returns. In addition, all PIT tagged and other hatchery fish were marked during the same time period, under the same environmental conditions, and with approximately the same level of stress. Body placement of CWT's and the presence of PIT tags allowed separation of PIT tagged fish from other hatchery fish post-release.

For the first brood (1997), the number of fish released from each acclimation raceway was the number of fish PIT-tagged adjusted for observed pre-release mortalities. Beginning with the 1998 brood, improved tags and detection equipment allowed survival indices to be based on tag detections at raceway outlets. The small proportion of tags (less than 1%) missed by acclimation site detectors but detected downstream was not included in the analysis.

The number of PIT tagged and total number of hatchery juveniles released by broodyear are given in Table 1.

<table>
<thead>
<tr>
<th></th>
<th></th>
<th></th>
<th></th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td>No. PIT tag released</td>
<td>39,892</td>
<td>37,385</td>
<td>38,791</td>
<td>37,580</td>
</tr>
<tr>
<td>Total number released</td>
<td>386,048</td>
<td>589,683</td>
<td>758,789</td>
<td>834,285</td>
</tr>
<tr>
<td>Percent PIT tag released</td>
<td>10.33%</td>
<td>6.34%</td>
<td>5.11%</td>
<td>4.50%</td>
</tr>
</tbody>
</table>

Returning fish were examined 6 months to 4 years after release. During that time fish migrated downstream below Roza Adult Monitoring Facility (RAMF; Figure 2). Fish age 3 and older migrated through the Yakima and Columbia rivers to the Pacific Ocean where they reared for 1 to 3 years, eventually returning to the upper Yakima River as maturing fish to spawn in September and October. Age 2 fish, called precocious males, likely do not migrate beyond the Columbia River basin or even the Yakima River basin before attempting to return and spawn (see Pearsons et al. 2004 and Larsen et al.)
2004 for a description of precocious males in the Yakima River and Beckman et al. for a discussion of spring chinook precocious male directed movements). At RAMF, all retuning fish with adipose fin clips were examined daily for marks and tags. Hatchery fish are initially diverted to a short term holding tank containing an anesthetic (MS222). They are then interrogated for the presence of marks and tags including PIT tags and CWT’s in the snout, and the number of fish retaining each tag was recorded. Fish passage at RAMF begins in late April and continues through early September.

Quality control samples are collected after each raceway’s tagging is completed and again between 2 to 4 months later in early February, prior to transfer to the acclimation sites. Beginning in 2000, functional PIT tag detectors were operating 24 hr per day at each acclimation site during the volitional release detecting an average of 98% of PIT tagged fish exiting the acclimation sites (Sampson and Fast 2000).

Since each fish is tagged with a combination of CWT, PIT tag and adipose fin clip, each adult return would be expected to have all three tags\textsuperscript{1} assuming no tag loss. Returning fish will fall into one of four categories (Table 2): 1) PIT tagged/CWT/Ad clipped (all tags retained), 2) CWT/Ad clipped (lost PIT tag), 3) PIT tagged/Ad clipped

---

\textsuperscript{1} Technically, elastomer marks and fin clips are called marks, rather than tags. However, we will refer to PIT tags, CWT’s, elastomer marks, and adipose fin clips generically as “tags” throughout the text.
(lost CWT), and 4) Ad clipped only (lost both PIT and CWT).  Fish falling into categories 1) to 3) can be identified unambiguously and used to calculate the number of fish having a single tag of each type. However, only 5-12% of the juveniles released are PIT tagged. And since all juveniles released are adipose fin clipped, fish in category 4) that have lost both their CWT and PIT tag will be confused with other project fish that also have lost their body CWT, retaining the adipose fin clip. However, we can make an estimate of the number of these fish.

Table 2. The four possible states of recovered fish after having been double marked prior to release with both a PIT tag and CWT.

<table>
<thead>
<tr>
<th>State Description</th>
<th>Tag Loss</th>
</tr>
</thead>
<tbody>
<tr>
<td>CWT tag retained, but no PIT tag</td>
<td>PIT tag retained, but no CWT</td>
</tr>
<tr>
<td>Both PIT tag and CWT retained</td>
<td>No PIT tag or CWT retained</td>
</tr>
</tbody>
</table>

Assuming tag loss within fish by tag type occurs independently, we can calculate the following from tagged fish from a broodyear recovered at RAMF:

\[
R_{\text{cwt}} = \text{the total number of fish from broodyear } i \text{ retaining only a CWT.}
\]

\[
R_{\text{pit}} = \text{the total number of fish from broodyear } i \text{ retaining only a PIT tag.}
\]

\[
R_{\text{pit,cwt}} = \text{the total number of fish from broodyear } i \text{ retaining both a PIT tag and CWT.}
\]

\[
R = R_{\text{cwt}} + R_{\text{pit}} + R_{\text{pit,cwt}} + \{\text{Number of fish losing both tags}\}
\]

We then estimated the probability of PIT and CWT loss (Seber 1982):

\[
(\text{Probability of losing a PIT tag}) = \frac{R_{\text{cwt}}}{(R_{\text{cwt}} + R_{\text{pit,cwt}})}
\]

\[
(\text{Probability of losing a CWT}) = \frac{R_{\text{pit}}}{(R_{\text{pit}} + R_{\text{pit,cwt}})}
\]

The fish that have lost both tags cannot be separated out from other non-PIT tagged, adipose fin clipped fish that have lost both of their tags. Thus, they cannot be directly enumerated and it is necessary to estimate the total number of captures, \(\hat{R}\). Following Seber (1982) this involves defining \(k\), the joint probability of losing each tag, assuming independence of each tag, that is, when a fish loses a PIT tag it does not affect whether or not it also loses or retains its CWT, then
\[ k = \frac{R_{cw} \cdot R_{pit}}{(R_{cw} + R_{pit,cwt})(R_{pit} + R_{pit,cwt})} \]

and

\[ c = \frac{1}{1 - k} \]

Then an estimate of the total number of PIT recaptures, including those losing both tags,

\[ \hat{R} = c(R_{cw} + R_{pit} + R_{pit,cwt}) . \]

We can make an initial estimate of the probability of losing both a PIT tag and CWT from published data. An average loss rate for snout CWTs in chinook salmon is 3% or less (Blankenship 1990; G. Shurman, WDFW, pers. comm.). PIT tag loss has been reported in coho salmon between 10 and 60% (Prentice et al. 1994). This would result in a worst-case estimate for the joint probability of losing a PIT tag and CWT of approximately 2%. Thus, not being able to factor in double-tag losses would likely result in a small underestimate in tag loss. However, we have estimated double-tag loss fish in our analyses.

In next year’s report we will stratify PIT tag loss estimates over months to determine if there was an increasing temporal trend in tag loss within years. We will also break data out by gender. However, these visual classifications of gender at RAMF on maturing fish are imprecise. For example, 66% of males and 96% of females were correctly identified to sex in fish returning in 2001 (Knudsen et al. 2002). So, analyses stratified by sex should be interpreted with some caution.

Survival impacts from PIT tags were estimated by comparing the proportion of PIT tagged fish recovered as adults and corrected for tag loss to the proportion of fish PIT tagged fish released as juveniles. After PIT tagging fish are placed back into their original raceways and reared together in the same raceways as the non-PIT tagged fish. PIT tagged fish are volitionally released along with non-PIT tagged fish and experience the same outmigration and post-release rearing conditions. Thus, differences in the

<table>
<thead>
<tr>
<th>Broodyear</th>
<th>PIT tag loss</th>
<th>CWT loss</th>
<th>PIT/CWT retained</th>
<th>Est. both tags lost</th>
<th>Total sample</th>
</tr>
</thead>
<tbody>
<tr>
<td>1997</td>
<td>N</td>
<td>112</td>
<td>26</td>
<td>574</td>
<td>712</td>
</tr>
<tr>
<td></td>
<td>%</td>
<td>16.3</td>
<td>4.3</td>
<td>0.7</td>
<td></td>
</tr>
<tr>
<td>1998</td>
<td>N</td>
<td>95</td>
<td>31</td>
<td>387</td>
<td>513</td>
</tr>
<tr>
<td></td>
<td>%</td>
<td>19.7</td>
<td>7.4</td>
<td>1.5</td>
<td></td>
</tr>
<tr>
<td>1999</td>
<td>N</td>
<td>5</td>
<td>3</td>
<td>55</td>
<td>63</td>
</tr>
<tr>
<td></td>
<td>%</td>
<td>8.3</td>
<td>5.2</td>
<td>0.4</td>
<td></td>
</tr>
<tr>
<td>2000</td>
<td>N</td>
<td>40</td>
<td>3</td>
<td>161</td>
<td>204</td>
</tr>
<tr>
<td></td>
<td>%</td>
<td>19.9</td>
<td>1.8</td>
<td>0.4</td>
<td></td>
</tr>
<tr>
<td>Mean percentage</td>
<td>16.1</td>
<td>4.7</td>
<td>0.8</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>
proportion of PIT tagged fish returning would indicate a direct PIT tag effect. Each year we released fish from 9 to 18 raceways of fish. Comparisons were made by broodyear using a \(X^2\) -test with Yates correction (Zar 1999).

**Results**

**PIT tag loss**

From April through September hatchery fish were interrogated for PIT tags, snout CWT’s, and adipose fin clips at RAMF. A total of 252 recovered fish had lost their PIT tag, 63 had lost their snout CWT, and 1,492 had retained both their PIT tag and CWT (Table 3). Estimates of PIT tag loss ranged from 8 to 20% and from 2 to 7% for CWTs. Over broodyears, PIT tag loss averaged 16.1% and CWT loss averaged 4.7%. Estimates of double-tag loss ranged from 0.4 to 1.5% and averaged 0.8%.

**Survival Impacts of PIT tags**

In Figure 3 we present by broodyear the percentage of recovered adult fish actually retaining their PIT tags \(R_{pit} + R_{pit,cwt}\), the expected percentage based on release numbers of PIT tagged and untagged hatchery juveniles, and the percentage of PIT tagged fish after correcting for PIT tag loss including the estimated number of fish losing

![Figure 3. Percent of recoveries made up of PIT tagged fish by broodyear. “Obs Uncor” represents the observed number of PIT tagged fish uncorrected for tag loss. “Expected” represents the expected number of PIT tag recoveries based on the proportion of PIT tagged fish at release and total recovery numbers. “Obs corrected” represents the observed number of PIT tagged fish corrected for PIT tag loss. Significance between uncorrected observed recoveries and expected recoveries are indicated above the respective columns (“*” signifies \(p \leq 0.05\); “**” signifies \(p \leq 0.01\)).](image)
both tags. In each broodyear, the percentage of PIT tagged fish actually recovered is significantly less than the expected percentage (each broodyear $X^2$-test with Yates correction $p<0.01$). After correcting for tag loss, the percentage of PIT tag recoveries increased in every broodyear. Estimates were less than the expected in 2 of 4 broodyears and in no case was there a significant difference ($X^2$-test with Yates correction $p>0.05$).

The difference between the expected recovery rate and the corrected recovery percentage can be interpreted as an estimate of PIT tag induced mortality. It is not a generic “tagging/handling mortality” because all the fish in the study were marked with more than one tag, reared in common raceways, and experienced common conditions throughout rearing, release and migration. Our estimates of PIT tag induced mortality ranged from 0 to 23% and averaged 4% over all broodyears. However, in no broodyear was a corrected estimate significantly different than the expected proportion at release.

**Juvenile quality control samples**

In January or February of each year approximately 2 to 4 months after tagging, quality control samples of approximately 200 fish per raceway were collected. The 200 fish are interrogated to identify PIT tags, CWTs, adipose fin clips, and, beginning in 1998, we began identifying colored elastomer injected into adipose eyelid tissue. Some fish have had CWT’s inserted into the cheek musculature or nape and these fish cannot be differentiated as juveniles from fish with snout tags that have lost a PIT tag using a hand held CWT detector. This is because the tag detector’s detection range cannot differentiate between CWT as close together as the cheek and snout in juveniles. We are still working through this issue.

**Discussion**

There PIT tag loss is occurring between the time fish exit the YKFP release sites and their subsequently return. We will be investigating the effects of age to determine whether there is a trend of increasing PIT tag loss as age increases. Since we did not dissect fish that were determined to have lost their PIT tag, we cannot rule out that in some cases the PIT tag was actually present, but not functioning. However, Prentice et al. (1993) examined PIT tag failure rates in salmonids and found that over periods as long as 3 years failure rates were typically 0-1%. They also found that nearly all failures were observed in the first sample collected within a few months after tagging and significant numbers of new failures were not detected after that. We have no reason to believe that PIT tag failure was higher than average in our releases and thus probably contributed 1% or less to the overall observed PIT tag loss.

After accounting for PIT tag loss, the percentage of PIT tag recoveries was not significantly different than expected. This argues strongly that post-release mortality due to PIT tags is not significant in hatchery spring chinook that are allowed to recover from post-tagging stress for between 3-4 months prior to release.

We had hoped to be able to make an estimate of PIT tag shedding from the time fish are collected at RAMF to the time they are spawned at CERSF. Any fish collected at RAMF and taken to CESRF with a juvenile PIT tag is identified. In addition, all adult fish collected at RAMF are PIT tagged in the pelvic musculature with an 18 mm PIT tag.
so they can be tracked from collection to spawning. Thus, it is theoretically possible to monitor whether individual fish shed their juvenile PIT tag after collection at RAMF prior to being spawned. However, in 2001 there were only 6 hatchery fish with juvenile PIT tags taken to CERSF and these few fish were not monitored through to spawning.

It is critical to understand the performance of any tag or mark in order to apply the most appropriate tag for the given situation and correctly interpret the results. When there is unaccounted for tag loss, survival will be underestimated and care should be taken before extrapolating the results to groups of untagged fish. However, when comparisons of juvenile-to-adult survival are made between similarly PIT tagged groups, the result should be a valid relative survival comparison. Caution should be exercised when making extrapolations to the un-PIT tagged portion of the population unless the effects of PIT tags on study fish have been investigated and taken into consideration.

All findings in this report should be considered preliminary and subject to further revision unless they have been published in a peer-reviewed technical journal.

Acknowledgements

The Yakama Nation PIT tagging and Roza Adult Monitoring Facility crews provided the PIT tagging expertise and recovery data used in these analyses. Bill Bosch (YN) provided valuable help in overall data management and access. We thank David Byrnes, Bonneville Power Administration, John Esterbrooks (WDFW) and Mel Sampson (YN) for their help in securing and administering funding for this work. Bruce Watson (YN) and Bill Hopley and Todd Pearsons (WDFW) provided encouragement and welcome advice.

Literature Cited


Blankenship, H. L. 1990. Effects of time and fish size on coded-wire tag loss from


Chapter Three

Comparison of Gametic Traits of First Generation Hatchery and Wild Upper Yakima River Spring Chinook Salmon

Curtis M. Knudsen¹, Steve L. Schroder², Mark V. Johnston³,

Todd N. Pearsons², and Charles R. Strom⁴

¹Oncorh Consulting, 2623 Galloway SE, Olympia, WA 98501

²Washington Department of Fish and Wildlife, 600 Capitol Way North, Olympia, WA 98501-1091

³Yakama Nation, P.O. Box 151, Toppenish, WA 98948

⁴Yakama Nation, Cle Elum Supplementation and Research Facility, 800 Spring Chinook Way, Cle Elum, WA 98922
Abstract

Reproductive traits in hatchery and wild spring Chinook females from the upper Yakima River were compared to determine whether fitness related traits had diverged after a single generation of artificial propagation. We also compared body size, survival and the proportion of abnormally developing progeny from single-pair matings of hatchery by hatchery and wild by wild adults. We found that fecundity (FEC), relative fecundity (RELFEC), egg weight (EW), and total gamete mass (TGM) were all significantly (p<0.001) correlated with female post-orbital hypural plate (POHP) length, while reproductive effort (RE) was not. It was necessary to analyze age 4 and 5 females separately due to significant (p<0.001) age related differences in their traits. In ANCOVAs testing FEC, RELFEC, EW, and TGM distributions for an Origin (hatchery/wild) effect by age and adjusting for POHP, no significant differences were found between hatchery and wild females. However, using published data showing that upper Yakima River hatchery spring chinook were significantly smaller than wild females and our trait/POHP regressions, we calculated age and broodyear specific mean reproductive trait values. Over the four broodyears examined, wild females averaged 7-9% greater TGM, 1-2% heavier EW, 6-7% greater FEC, and 1% lower RELFEC than hatchery females. No Origin effect was found in RE. The fundamental reproductive trait/body size relationships had not been significantly altered by a single generation of hatchery exposure. However, due to significantly smaller hatchery mean body size which is correlated with the reproductive traits, returning hatchery and wild female’s reproductive traits did differ, likely resulting in some loss of fitness in hatchery females. We found no Origin effect in the proportion of abnormally developing fry. After adjusting for egg size, we found that hatchery fry were on average ~1% heavier than wild fry. Comparisons of fry survival were mixed with no clear trend in survival of hatchery and wild eggs to the emergent fry stage.
Introduction

Washington state has practiced artificial propagation of salmon and steelhead for over a century and during this period significant advances have been made in fish culture technology resulting in hatchery spawner/recruit rates that can considerably exceed replacement rates. The importance of hatchery operations has increased because of continuing losses of natural production from over-harvest, habitat degradation, and disappearance of spawning habitat due to hydroelectric development, irrigation, logging and transportation (Lichatowich 1999). However, artificial production’s effects on native populations is not well understood (e.g., Waples et al. in press; Goodman 2005). Use of “integrated” hatchery programs in the Columbia River basin has recently increased (Goodman 2004) making the issue of deliberate interbreeding of hatchery origin and natural origin fish even more significant (Goodman 2005; Mobrand et al. 2005). The demographic risks of integrated programs have been recognized (Hard 1995; Goodman 2004; Mobrand et al. 2005) and aspects of the genetic risks of integrated programs have been modeled (Lynch and O'Hely 2001; Ford 2002; Goodman 2005). However, empirical assessments of integrated programs are lacking, as illustrated by a recent review (Berejikian and Ford 2004) that compared the fitness of natural and hatchery origin fish. Seventeen of the 18 studies reviewed examined the effects of intentional selection, multiple generation effects, use of non-local broodstock, or combinations of these factors. To evaluate the risks and benefits posed by integrated programs, appropriate demographic and genetic data need to be collected (Hard 1995), preferably from the beginning of a program. Assuming native broodstock were used, this permits documenting whether first generation hatchery fish diverge from their founder population prior to hatchery introgression. After progeny of first generation hatchery fish begin naturally spawning with wild fish, their progeny may possess characteristics that are intermediate between those of progeny of pure wild and hatchery origin individuals. Accordingly, when naturally produced individuals of mixed hatchery and wild ancestry are compared to second generation hatchery fish, their trait distributions will differ less than between pure first generation wild and hatchery individuals.

Life-history traits, particularly those directly associated with reproduction, reflect local adaptations affecting fitness (Stearns 1976; Roff 1992). Relaxation of natural selection for larger eggs combined with domestication selection for greater fecundity was suggested as the reason egg size declined in a chinook salmon captive breeding program (Heath et al. 2003; however see Beacham 2003 and Fleming et al. 2003). Such traits as egg size, reproductive effort (biomass of gametes relative to total body biomass) and fecundity are maternal traits. However, they also have direct consequences for progeny affecting yolk reserves and fry body size (Einum et al. 2004). Other maternal traits also have direct consequences for progeny. Where a females chooses to construct her redd will determine the quality of the incubation substrate as well as early fry rearing habitat. When a female spawns will significantly affect emergence timing and thus the state of her progeny’s early rearing environment. If a female spawns too early, food will be scarce, although density and competition will likely be low. Spawning too late, when productivity is higher, also results in greater competition with earlier emerging, larger fry. Life history theory suggests that natural selection will maximize female fitness. Assuming there is some maximum to the resources a female can devote to gametes
(either space or biomass), there must be a trade-off between egg size and egg number. An increase (decrease) in egg size results in a decrease (increase) in egg number. In general, significant changes in locally adapted traits will be maladaptive in the wild (Lynch and O’Hely 2001; Ford 2002), and can result in reduced individual fitness (Taylor 1991; Fleming and Gross 1993; Fleming et al. 2000). Monitoring hatchery populations to determine if they are diverging from their native population’s life-history trait distributions is a necessary part of a hatchery monitoring plan (Hard 1995; Goodman 2005). Significant differences may indicate that the artificial rearing environment is causing genetic divergence to occur between the two groups. However, trait differences may be due to phenotypic plasticity and are not sufficient to conclude that genotypic divergence has occurred. To reach that conclusion, fish from both groups should be spawned, incubated, and reared in a common environment. Observed differences under these circumstances would represent genetic change.

In 1997 an integrated hatchery program was begun to supplement the Upper Yakima spring chinook population (Fast and Craig 1997). The program used only natural origin fish as broodstock and no attempt was made to control the proportion of hatchery origin fish on the spawning grounds. Our approach has been to measure a variety of gametic traits on adult spring chinook as they matured and were spawned at the Cle Elum Supplementation Research Facility (CESRF) over the first generation of hatchery returns from the broodyears 1997 to 2000. The integrated hatchery program is described in detail in Knudsen et al. (in press). In this paper we compare wild and first-generation hatchery origin female’s egg weight, total gamete mass, reproductive effort, fecundity and relative fecundity distributions. Then, extending the comparisons into the next generation we compare the survival, incidence of abnormally developing fry, and body size of fry produced from single-pair hatchery-by-hatchery and wild-by-wild crosses.

Methods

Upper Yakima River spring Chinook

The Yakima River is a major tributary to the Columbia River located in south central Washington state (Figure 1). The upper Yakima River supports a genetically distinct (Busack and Marshall 1991), naturally sustaining population of ‘stream type’ spring chinook (Healey 1991). After rearing for 1 to 3 years in the North Pacific Ocean, adults migrate upstream into the Yakima River basin in the spring and spawn in the early fall, and juveniles spend a full year in freshwater before migrating to the ocean. Some males mature precociously in freshwater during their first or second year (Larsen et al. 2004; Pearsons et al. 2004).

The Yakima/Klickitat Fishery Project’s (YKFP) spring Chinook hatchery program began in 1997 at the Cle Elum Supplementation Research Facility (CESRF) near Cle Elum on the upper Yakima (rkm 297; Figure 1). This program is an integrated hatchery program (Mobrand et al. 2005) taking only natural origin broodstock and allowing retuning hatchery origin adults to spawn in the wild. It is designed to test whether artificial propagation can be used to increase natural production and harvest opportunities while limiting ecological and genetic impacts. The program includes a domestication monitoring effort that compares the supplemented population at several
traits to a hatchery only control line founded from first-generation hatchery returns, and
to a wild control line (the unsupplemented Naches population (Busack et al. 2004)).

As integrated programs proceed beyond the first generation, it is inappropriate to
call fish resulting from natural spawning “wild”, because they may be the progeny of
naturally spawning hatchery fish. These fish are more appropriately called “natural
origin” fish. We call the naturally produced fish in this study wild because they were
produced before significant numbers of naturally produced fish of hatchery ancestry
returned to spawn (Table 1). There may have been some contribution of hatchery origin
age 3 males from the 1997 brood year to the 2000 brood, but this influence was probably
slight, as these fish accounted for only 5% of the natural spawning population (YN
unpublished data). Additionally, the upper Yakima spring Chinook in this study can be
considered wild because this population had been subjected to only negligible levels of
hatchery activity prior to the initiation of the YKFP.

Spring Chinook for this study were collected as they passed upstream through
Roza Adult Monitoring facility (RAMF) and were transported via tanker truck to CESRF.
For a full description of the collection of hatchery and wild adults at RAMF see Knudsen
et al. (in press). We were able to use a “common garden” experiment to test for
differences between hatchery and wild origin gametic traits because all adults were held together in a single concrete raceway at CESRF. Beginning in early September and continuing into early October adults were checked for ripeness and spawned weekly. Ripe females were identified when eggs were extruded with gentle manual pressure or by the firmness of the ventral surface. A soft ventral surface that sagged slightly when the head was pointed head down indicated a female was ripe.

After reaching maturity, hatchery and wild origin female broodstock were sampled in order to compare the following gametic traits: total gamete mass (TGM), mean egg weight (EW), reproductive effort (RE), fecundity (FEC), and relative fecundity (RELFEC). In addition, the following traits associated with post-fertilized eggs were measured: survival to the eyed-egg stage, incidence of abnormally developing fry, and fry body length and weight. Descriptions of the data collection processes are given below.

**Total gamete mass (TGM), Egg weight (EW), and Fecundity (FEC)**

The weight of the total gamete mass (TGM) and average EW were measured as females were artificially spawned at CESRF. Loose eggs and ovarian fluid from a ripe female were collected in a dry 1 gallon plastic bucket. Ovarian fluid was drained off using a dry plastic colander. The female’s TGM was then weighed to the nearest 0.1 g. A sample of 30-50 eggs was collected and weighed to the nearest 0.01 g. The number of eggs in the sample was counted and divided by the sample weight to calculate the mean EW. The gravimetric estimate of fecundity was then calculated by dividing the TGM by the mean EW. There is always some residual ovarian fluid remaining within the egg mass and thus our gravimetric fecundity estimates were biased; overestimating the true fecundity. In order to calculate a correction factor for this bias, we hand counted the total number of eggs produced by 110 females and regressed the estimated gravimetric fecundities against the hand counts, forcing the regression through a 0 y-intercept. The estimated slope was then used as a bias correction factor and applied to all gravimetric fecundity estimates which were used in the analyses below.
Table 1. Chronology of development of hatchery ancestry in natural-origin upper Yakima spring Chinook through first three generations of integrated hatchery operation. Calendar years of return for each brood year from 1997 to 2005 are shown. Entries denotes age of returns. Font style denotes hatchery ancestry (unbolded=wild only, bold=substantial hatchery ancestry, unbolded with asterisk= small influence from age 3 males only (see text)).

<table>
<thead>
<tr>
<th>Brood years</th>
<th>Generation 1</th>
<th>Generation 2</th>
<th>Generation 3</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Initiation of hatchery operations and broodstock collection</td>
<td>Hatchery fish begin returning to spawn naturally</td>
<td>First returns of natural-origin fish produced by naturally spawning hatchery fish</td>
</tr>
<tr>
<td>1997</td>
<td>3</td>
<td>4</td>
<td>5</td>
</tr>
<tr>
<td>1998</td>
<td>3</td>
<td>4</td>
<td>5</td>
</tr>
<tr>
<td>1999</td>
<td></td>
<td>3</td>
<td>4</td>
</tr>
<tr>
<td>2000</td>
<td></td>
<td></td>
<td>3</td>
</tr>
<tr>
<td>2001</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>2002</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>2003</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>2004</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>2005</td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>
Reproductive effort (RE)

Reproductive effort (RE), also called gonadal-somatic index, was calculated by dividing the TGW by the fish’s total body weight (including gametes) and represents the proportion of body mass allocated to gamete production. Because RE is a dimensionless ratio of two weight measures, an arc sin square root transformation (Zar 1999) was used to normalize its distribution during analyses, but we report the values as untransformed ratios in the text.

A few females had a significant proportion of unripe, overripe, or injured eggs. We assumed these occurred because either females were selected for spawning either too early or too late or the eggs were injured during handling, transfer and holding. In addition, during the latter weeks of the spawning season a few eggs were observed on the bottom of the adult holding raceway indicating that some females had released gametes before being selected for spawning. For these reasons, we excluded TGM, FEC and RE values of females with RE values below 0.12 (20 out of 847 wild and 5 out of 189 hatchery females). An RE value of 0.12 was 2-3 standard deviations from the mean RE value in each broodyear.

Relative fecundity (RELFECE)

Relative fecundity (RELFECE) was calculated by dividing a female’s total fecundity (FEC) by her POHP length and represents the number of eggs produced per unit change in POHP length (eggs·cm⁻¹).

Factorial mating protocols and egg incubation

The fish used in the matings for survival and post-fertilization analyses were spawned in a series of factorial crosses typically made up of 3 females and 3 males, resulting in 9 single pair matings. However, there were cases where other combinations such as 2x2 or 2x3 crosses were used. In general, three aliquots of between 150 and 250 eggs per female were collected and placed into a dry 1 L beaker with approximately 0.2 cc of milt (3 drops from a 5 cc syringe) from the respective male in the single-pair mating. The gametes were then activated by adding approximately 200 ml of ambient well water, initiating fertilization. After a minimum of 2 minutes from activation, the eggs from each single-pair mating were drained, placed into individual incubation containers called isolettes, and held in an Iodiphore bath for 45 minutes. Each isolette was labeled with the female and male’s origin and individual identification numbers. The eggs from each female were then incubated to the eyed egg stage, shocked, and mortalities enumerated and removed. The remaining eggs were incubated to the post-hatching yolk absorption or “button up” stage. Any additional mortality was then noted and deformities and abnormally developing fry (e.g. scoliosis, missing eyes, Siamese twining or abnormal fin development) were enumerated.

Fork length and body weight were measured on five fry from one single-pair mating from each female. Fry were anesthetized and blotted dry prior to being weighed. Because we collected fry size data from only one single-pair mating per female, we could not estimate male effects on fry body size. However, we were monitoring fry size at the “button up” stage, when maternal effects, particularly those due to egg size, should overwhelm paternal effects (Iwamoto et al. 1984; Heath et al. 1999). Wild and Hatchery
origin fry body size distributions were compared by ANCOVA using egg weight as a covariate.

Knudsen et al. (in press) showed that body size-at-age of first generation CESRF hatchery spring Chinook salmon adults was significantly smaller than wild adults. Chinook salmon body size has been shown to be positively correlated with fecundity (Beacham and Murray. 1993; Healey and Heard 1984), egg size (Beacham and Murray. 1993; Healey 2001), and total egg mass (Kinnison et al. 2001) and negatively correlated with relative fecundity (Heath et al. 2003).

Our general procedure for analyzing female traits began by comparing age 4 and 5 females within Origin types using ANCOVA to control for body size (POHP length) in order to determine whether it would be necessary to analyze the two age classes separately. We then determined if the hatchery and wild slopes were equal and ANCOVA was appropriate. If so, we compared Origin types within ages to determine whether hatchery and wild females had equal trait/POHP slopes and adjusted means. That is, did they have the same fundamental relationships between body size and trait distributions or had exposure to hatchery culture altered this relationship somehow. If there were no significant difference in slopes and intercepts due to Origin, then we combined the hatchery and wild samples and calculated the linear regression equation. Using the appropriate regression, we then estimated the mean trait values of hatchery and wild fish based on the age and broodyear specific mean POHP lengths reported in Knudsen et al. (in press).

Statistical tests were considered significant when $p$-values were less than or equal to 0.05. All analyses were executed using SYSTAT version 11 software (SYSTAT Software, Inc.).

**Results**

**Total gamete mass (TGM)**

Within Origin type, age 4 and 5 females had equal slopes (Hatchery $p=0.862$; Wild $p=0.989$). However, age 4 and 5 adjusted means were significantly different in both hatchery ($p<0.001$) and wild ($p=0.001$) females requiring separate analyses for each age.

Within ages, both hatchery and wild populations had equal slopes (Age4 $p=0.993$; Age5 $p=0.871$; Figure 2). Hatchery and Wild groups within age classes exhibited no significant differences in ANCOVA of TGM (Age4 $p=0.8274$; Age5 $p=0.6465$). Thus, within ages TGM did not differ between hatchery and wild females after adjusting for POHP length.
POHP length was highly correlated with TGM (all p<0.0001; r² ranged from 0.544 to 0.581). The final calculated difference between hatchery and wild groups by broodyear were estimated from annual mean age 4 and 5 POHP lengths. These estimated TGM means are shown in Figure 3. Over broodyears 1997 to 2000, wild age 4 and 5 TGM means were 7.2% and 8.7% greater than hatchery age 4 and 5 TGM’s, respectively.

Figure 3. Estimated mean gamete mass (g) for age 4 and 5 hatchery and wild males. TGM was estimated from age specific log_TGM vs. log_POHP regressions (Age 4 TGM = 0.00586*(POHP)^2.8704; Age 5 TGM = 0.00466*(POHP)^2.9098) and mean POHP lengths taken from Knudsen et al. (in press).
Egg weight (EW)

In comparisons between ages by Origin, age 4 EW’s were significantly smaller than age 5 EW’s (p<0.002) after adjusting for POHP length. On average hatchery age 4 EW’s were 4.6% to 5.8% smaller than age 5 female EW’s.

There was no significant difference between EW/POHP relationships of hatchery and wild females. After substituting mean POHP lengths from broodyears 1997 to 2000 into the age specific regressions, we calculated that wild EW was 0.2 to 3.9% greater than hatchery EW (Figure 4); averaging 1 and 2% greater over all broodyears.

Fecundity (FEC)

We hand counted the fecundity of 110 females and compared the counts to the paired gravimetric fecundity estimates. In all cases, fecundity was overestimated. We regressed the hand counts against the gravimetric estimates (Figure 5) using a simple linear regression forced through a zero y-intercept and found we had overestimated fecundity on average by 5.5% (regression r² = 0.999; p<0.0001; Corrected fecundity = 0.9447* Gravimetric fecundity). We used this relationship to correct all of our gravimetric fecundity estimates below.
ANCOVA comparing loge-fecundity between ages using loge-POHP length as a covariate showed that within Origin type ages 4 and 5 had equal loge-fecundity vs loge-POHP slopes (hatchery p=0.444; wild p=0.383), however age 4’s had significantly higher adjusted mean fecundity (equal adjusted means hatchery p<0.001; wild p<0.001). Age 4 females were 10-14% more fecund than age 5 females of the same POHP length. Thus, the two age classes should be analyzed separately.

Fecundity distributions were then compared by age class using ANCOVA with loge-POHP as the covariate. Hatchery and wild females had slopes (equal slopes: age 4 p=0.393; age 5 p=0.411) and adjusted means (equal means: age 4 p=0.721; age 5 p=0.660) that did not differ significantly. Thus, hatchery and wild females of the same age and POHP length should on average have the same fecundity. However, hatchery and wild female POHP lengths were not equal for adult returns from broodyears 1997 to 2000 (Knudsen et al. in press). In order to estimate what hatchery and wild fecundities were for each broodyear, we first estimated separate relationships between loge-Fecundity and loge-POHP for ages 4 and 5, combining hatchery and wild samples within ages. In both ages, fecundity and POHP were significantly positively correlated (Age 4: loge_Fecundity = (loge_POHP * 2.4800) – 1.9147, N=1012, r²=0.544; p<0.0001; Age 5: loge_Fecundity = (loge_POHP * 2.4031) – 1.7069, N=73, r²=0.581, p<0.0001). After substituting into the appropriate equation the mean POHP length for each combination of age, origin and broodyear we found that mean fecundity over the four broodyears of wild age 4 and 5 females was 6.2% and 7.2% higher than hatchery female fecundity, respectively. Estimated mean fecundities by origin, broodyear and age with 95 percent confidence intervals are given in Figure 6 (Zar 1999).
Female Reproductive effort (RE)

There was no correlation between body length and RE in either age 4 or age 5 females regardless of origin (all correlation p>0.612; Figure 7). Thus, it was not necessary to use body length as a covariate. There was no significant difference between hatchery and wild female’s RE distributions in a 3-way ANOVA (Table 2). However, Age effects were highly significant and indicated RE of age 4 females (mean RE=0.202) was 6.9% greater on average than age 5 females (mean RE=0.189) regardless of POHP length. Thus, age 4 females allocated a significantly higher proportion of body weight to gametes than age 5 females. Broodyear main effects and all interaction effects were not significant (all p>0.387).

Table 2. Reproductive effort comparisons using a 3-way ANOVA testing for Broodyear, Age, and Origin main effects and all possible interactions.

<table>
<thead>
<tr>
<th>Source</th>
<th>Sum-of-Squares</th>
<th>df</th>
<th>Mean-Square</th>
<th>F-ratio</th>
<th>P</th>
</tr>
</thead>
<tbody>
<tr>
<td>Broodyear</td>
<td>0.0003</td>
<td>2</td>
<td>0.0001</td>
<td>0.1946</td>
<td>0.823</td>
</tr>
<tr>
<td>Age</td>
<td>0.0078</td>
<td>1</td>
<td>0.0078</td>
<td>10.2025</td>
<td>0.002</td>
</tr>
<tr>
<td>Origin</td>
<td>0.0005</td>
<td>1</td>
<td>0.0005</td>
<td>0.6297</td>
<td>0.428</td>
</tr>
<tr>
<td>BY*Age</td>
<td>0.0004</td>
<td>2</td>
<td>0.0002</td>
<td>0.2726</td>
<td>0.762</td>
</tr>
<tr>
<td>BY*Origin</td>
<td>0.0016</td>
<td>2</td>
<td>0.0008</td>
<td>1.0216</td>
<td>0.360</td>
</tr>
<tr>
<td>Age*Origin</td>
<td>0.0002</td>
<td>1</td>
<td>0.0002</td>
<td>0.2043</td>
<td>0.651</td>
</tr>
<tr>
<td>BY<em>Age</em>Origin</td>
<td>0.0007</td>
<td>2</td>
<td>0.0003</td>
<td>0.4465</td>
<td>0.640</td>
</tr>
<tr>
<td>Error</td>
<td>0.6687</td>
<td>875</td>
<td>0.0008</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>
Relative fecundity (RELFEC)

We compared hatchery and wild RELFEC distributions using ANCOVA, adjusting for body length. Both RELFEC and POHP length were loge transformed. Initially, we analyzed hatchery and wild samples separately and found that age 4 and 5 females of both Origins had equal RELFEC/POHP slopes (p>0.147), but significantly different adjusted means (p<0.001). Age 4 females had 10.7% greater RELFEC than age 5’s. That is, age 4 females produced just over 100 more eggs than an age 5 female of the same POHP length. There were no significant differences due to Origin within age classes (ANCOVA equal slopes p>0.472; equal mean p>0.547). LogePOHP and logeRELFEC were weakly negatively correlated (Age 4, p<0.0001; r^2=0.026; Age 5, p<0.0001; r^2=0.057; Figure 8).
Figure 8. Comparison of age 4 and 5 loge Relative Fecundity (RELFEC) vs POHP length relationships showing that age 5 females (X; dotted line) had significantly lower RELFEC than age 4 females (○; solid line) at a standardized POHP length. Hatchery and Wild females were combined.

Figure 9. Mean Relative fecundity (RELFEC) of age 4 and 5 Hatchery and Wild females by broodyear. Relative fecundities were estimated from age specific loge transformed RELFEC/POHP regressions and broodyear specific mean POHP lengths from Knudsen et al. (in press). Age 4 RELFEC = 4902.5*(POHP)^0.3860; Age 5 RELFEC = 7104.6*(POHP)^0.5003.

After adjusting for age and broodyear specific mean POHP lengths, we found that age 4 and 5 hatchery females had slightly greater RELFEC in all broodyears, averaging 0.9 and 1.4% greater RELFEC than wild age 4 and 5 females, respectively (Figure 9). This was due to the negative correlation between POHP and RELFEC.
Fry Survival

In 3 of 4 broodyears, hatchery survival to the emergent fry stage (overall unweighted mean = 87.5%) was greater than wild survival (overall unweighted mean = 85.7%; Table 3). In a 2-way ANOVA (Origin and BY main effects) of survival, there was a significant interaction effect due to inconsistent responses over broodyears (p<0.01). Therefore, we analyzed each broodyear separately for Origin effects using a t-test. In 2001, wild fry survived at a significantly higher rate than hatchery fry (p=0.047) and in 2004 hatchery fry had significantly higher survival (p=0.023). The other two broodyears were not significantly different (2002 and 2003 p>0.41). Thus, fry survival effects due to Origin varied significantly across broodyears and there was no consistent trend.

Table 3. Survival of hatchery and wild eggs to the emergent fry stage by broodyear. T-test results comparing hatchery and wild fry survival by broodyear are indicated by an “*” (p<0.05) or “ns” (p>0.05) to the right of the means.

<table>
<thead>
<tr>
<th>Broodyear</th>
<th>Origin</th>
<th>Mean % Survival</th>
<th>sd</th>
<th>n</th>
</tr>
</thead>
<tbody>
<tr>
<td>2001</td>
<td>Wild</td>
<td>89.4</td>
<td>11.4</td>
<td>91</td>
</tr>
<tr>
<td></td>
<td>Hatchery</td>
<td>86.5*</td>
<td>9.6</td>
<td>43</td>
</tr>
<tr>
<td>2002</td>
<td>Wild</td>
<td>90.4</td>
<td>18.3</td>
<td>103</td>
</tr>
<tr>
<td></td>
<td>Hatchery</td>
<td>90.7 ns</td>
<td>9.2</td>
<td>99</td>
</tr>
<tr>
<td>2003</td>
<td>Wild</td>
<td>86.7</td>
<td>19.5</td>
<td>54</td>
</tr>
<tr>
<td></td>
<td>Hatchery</td>
<td>87.7 ns</td>
<td>14.8</td>
<td>58</td>
</tr>
<tr>
<td>2004</td>
<td>Wild</td>
<td>76.4</td>
<td>25.3</td>
<td>88</td>
</tr>
<tr>
<td></td>
<td>Hatchery</td>
<td>85.2*</td>
<td>14.6</td>
<td>71</td>
</tr>
</tbody>
</table>

Fry Body Size

In ANCOVA of fry body weight, testing for Origin and Broodyear main effects and using egg weight as a covariate, hatchery fry (mean across-years body weight = 0.317 g) were larger than wild fry (mean across-years body weight 0.313 g) in each of the four years (Figure 10). There was a significant Origin effect (p=0.002) indicating that, given eggs of the same weight, hatchery fry will have ~1% greater body mass than wild fry. Thus, hatchery eggs are slightly more efficient in converting yolk to body mass.

Fry Developmental Abnormalities

In general, the median proportion of fry abnormally developing was very low in 2002 and 2003, never exceeding 0.005. Since the distributions are highly skewed with many values at or near 0 we used the median to describe the distribution’s central tendency. Wild median proportions ranged from 0.003 (n=35) to 0.004 (n=18) and the hatchery proportions from 0.002 (n=20) to 0.004 (n=33). In a Kruskal-Wallis nonparametric one-way ANOVA there was no significant difference between the proportion of abnormally developing hatchery and wild fry (p=0.258).
Discussion

We found that POHP length was significantly correlated with all hatchery and wild female reproductive traits we examined, except RE. When we compared the body size/trait relationships between hatchery and wild females’ using ANCOVA there were no significant differences due to Origin effects after adjusting for POHP length. Thus, exposure to the hatchery environment has not altered these fundamental growth and reproductive energy allocation relationships in females. However, Knudsen et al. (in press) demonstrated that upper Yakima River hatchery females were significantly smaller at age than wild adults from broodyears 1997 to 2000. Using their age- and broodyear-specific body sizes, we estimated mean hatchery and wild gamete trait values based on our body size/trait regressions and found that wild females had 7-9% greater TGM, 1 to 2% larger EW, 6-7% greater fecundity, and 1% lower relative fecundity. And these differences are likely to reduce to some degree hatchery female fitness. There was no difference between hatchery and wild female RE distributions.

Healey and Heard (1984) found that older chinook salmon females had greater fecundity than younger females. We also found that age profoundly affected the fecundity/body size relationship, but in the opposite direction. After adjusting for body size, age 4 females had significantly higher fecundity than age 5 females of both Origin types. This is due in large part to the fact that upper Yakima River age 5 females had
larger eggs than age 4 females of the same POHP and lower RE. Thus, for females of the same POHP length, age 5 females had a lower proportion of their total biomass allocated to gametes, and within that smaller gamete mass the individual eggs were larger, resulting in lower fecundity. Producing larger eggs is one form of parental care; providing progeny additional yolk reserves and larger size at emergence and age 5 females sacrifice egg numbers to produce larger eggs.

The fastest growing fish in a cohort will mature earlier on average. Thus, age 4 females are on average faster growing relative to age 5 females. If we assume that maturation and gamete production is triggered by reaching some minimum critical body size by a specific date or time period, then this difference in age specific growth rate could explain the difference in how much energy each age class can allocate to gametes. It will take the slower growing age 5 females an additional number of months of growth to reach the critical body size by which time they are outside of the critical time period to begin maturation and must complete an additional years growth. Those additional months needed to reach the minimum size cannot be made up and are essentially lost months of growth. This puts the older maturing female at an energy deficit relative to faster growing earlier maturing females which gets expressed as lower RE, FEC and RELFEC at an adjusted body size. Once age 5 females do reach the critical body size, they appear to produce gametes at the same rate per unit body size increase as age 4 females. That is, the slopes of the trait-body size relationships we calculated were equal between the two ages. It is just that the younger females’ regression lines were higher and this difference in height may represent the energy deficit from the reduced number of months of gamete growth in age 5 females.

Hatchery and wild females of the same age allocated equal proportions of body mass to gamete production and within age classes RE was not correlated with body length in hatchery or wild females. However, there were differences in RE between the age classes: age 4 females had 7% higher RE on average allocating more of their body mass to gametes than age 5 females of the same length.

We were interested in whether RE responded in the same way to increasing egg size in the two age classes of females. That is, does the proportion of gamete biomass increase with increasing egg size so that fecundity remains relatively stable or does RE remain stable as egg size increases, resulting in a sacrifice in fecundity for a given increase in egg size? We found that the two age classes have significantly different RE/Egg size relationships (equal slopes p<0.001; Figure 11). In age 4 females, as egg size increases RE also increases and thus there is no tradeoff in fecundity. Faster growing females tend to mature earliest at age 4 and this appears to give age 4 females more flexibility in allocation of biomass to RE without sacrificing either egg size or fecundity. Age 5 females are more rigid with respect to RE and any increase in egg size will result in a trade off decreasing fecundity.
Figure 11. Linear regressions of RE vs Egg weight for age 4 (○; solid line) and 5 females (x; dashed line). Hatchery and wild samples have been combined.

Figure 12. Relationship between egg weight and egg-to-emergent fry survival for hatchery (○, solid line) and wild (x, dashed line) samples.
Thrope (2004) proposed that the protected hatchery environment permits fishes to reduce the proportion of energy normally allocated to competition for food, shelter and mates, avoidance of predators and counteracting parasites and diseases. This would allow more energy to be directed toward growth and reproduction, resulting in earlier maturation and increased relative fecundity. However, Upper Yakima River hatchery and wild females return primarily at age 4, and there has been no increase in the proportion of hatchery females returning at age 3 (Knudsen et al. in press). Hatchery females did have higher RELFEC in all comparisons calculated from POHP/RELFEC regressions, although the increase over wild females averaged only 1%.

We found no difference in the survival of hatchery and wild fry produced from gametes spawned and incubated within the CESRF. Heath et al. (2003) found that preemergence survival was positively correlated with egg size. We tested their finding with our data and found no significant correlation between logE EW and survival to the emergent fry stage in either hatchery (regression p=0.465, r^2=0.01, n=53) or wild (regression p=0.215, r^2=0.03, n=53) samples (Figure 12).

There was no significant difference between hatchery and wild females in regressions of FEC, RELFEC, EW, and TGM distributions and body size. The fundamental reproductive trait/body size relationships have not been significantly altered by a single generation of hatchery exposure. However, because upper Yakima River hatchery spring chinook were significantly smaller than wild females, age and broodyear specific mean reproductive trait values differed likely resulting in some loss of fitness in hatchery females naturally spawning. Over the four broodyears examined, wild females averaged 7-9% greater TGM, 1-2% heavier EW, 6-7% greater FEC, and 1% lower RELFEC than hatchery females. No Origin effect was found in RE. Females of both origins demonstrated significant differences in the manner age 4 and 5 females allocated energy to reproductive traits per unit body growth and this area would be useful to study further. We found no significant difference in the quality of progeny produced by hatchery and wild females, although hatchery fry were more efficient at converting yolk into biomass, resulting in ~1% heavier fry compared to wild fry produced from the same size egg.

Acknowledgements

We wish to thank the hatchery manager Dan Barrett (retired) and Yakama Nation CESRF personnel, in particular Annie Joe Parrish, for their cooperation. In addition, we would like to thank Todd Newsome and Ann Stephenson (YN) and Edward Beall (INRA) for their help in sampling gametes at CESRF. Bill Bosch (YN) provided valuable help in overall data management and information access. Finally, we also thank John Easterbrooks (WDFW) and Mel Sampson (YN) for policy support and Bonneville Power Administration for funding for this work.
Literature cites


