Factors influencing spawner success in a spring Chinook salmon (*Oncorhynchus tshawytscha*)
reintroduction program

Nicholas M. Sard¹, Kathleen G. O’Malley¹, Dave P. Jacobson¹, Michael J. Hogansen², Marc A. Johnson¹,², and Michael A. Banks¹

¹Oregon State University, Department of Fisheries and Wildlife, Coastal Oregon Marine Experiment Station, Hatfield Marine Science Center, 2030 SE Marine Science Drive, Newport, Oregon 97365

²Oregon Department of Fish and Wildlife, Upper Willamette Research, Monitoring, and Evaluation, Corvallis Research Laboratory, 28655 Highway 34, Corvallis, Oregon 97333

*Corresponding author email: nicksard@gmail.com

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Abstract

Dams have contributed to the decline of migratory fishes by blocking access to historical habitat. The active transport (trap and haul) of migratory fish species above existing dams can sometimes support population recovery when the use of fish ladders or dam removal is infeasible. However, little is known about the efficacy of trap and haul conservation strategies. Here we used genetic parentage assignments to evaluate the efficacy of reintroducing adult Chinook salmon above Cougar Dam on the South Fork McKenzie River, Oregon, USA from 2008-2011. We found that mean reproductive success (RS) declined as adults were released later in the spawning season in 2009 and 2010; however release location did not affect RS. In 2010 and 2011, we tested for RS differences between hatchery and natural origin (HOR and NOR) adults. HOR males were consistently less fit than NOR males, but little evidence for fitness differences was apparent between HOR and NOR females. Interestingly, the effect of origin on RS was not significant after accounting for variation explained by body length. Our results indicate that release date and location have inconsistent or no effect on the reproductive success of reintroduced adults when active transport strategies are employed for migratory fishes.

Key words: reintroduction, active transport, genetic parentage, reproductive success, hatchery and natural origin
Introduction

Reintroduction programs attempt to re-establish locally extirpated species to their historical habitat (IUCN 2013) and can serve to restore a species' distribution when access to habitat has simply been blocked. However, few reintroduction programs have been successful to date (Fischer and Lindenmayer 2000; Wolf et al. 1996), and most research on reintroductions has focused on mammals and birds (Seddon et al. 2005), rather than fish (but see George et al. 2009). There are many migratory fish species (see Brönmark et al. 2014; Lucas et al. 2001) whose access to critical habitat has been negatively affected by dams. Therefore, an understanding of factors that affect the success of fish reintroductions is greatly needed.

Reintroduction efforts have become an increasingly common management strategy to aid in the recovery of threatened salmonid populations, often through the decommissioning of dams or the construction of fish ladders (reviewed by Pess et al. 2014). In Chinook salmon (Oncorhynchus tshawytscha), reintroductions following dam removal or fish ladder construction have been evaluated through studies of juvenile production and subsequent adult returns (Anderson et al. 2013; Baumsteiger et al. 2008). However, habitat connectivity cannot always be restored through dam removal or fish ladder construction, as with the case of high-head flood control dams (see example projects in Anderson et al. 2014). In such scenarios, the collection and active transport of migratory salmonids provides an alternative approach that has received little attention in the literature (but see Evans et al. In Review).

Spring Chinook salmon, in the upper Willamette River, Oregon, are listed as a threatened Evolutionary Significant Unit (ESU) under the U.S. Endangered Species Act (ESA) (NMFS 1999; 2005; 2008). Dams impede migration to 32% of Chinook salmon historical habitat, and alter water temperature and flow regimes (ODFW 2005; Sheer and Steel 2006). In addition, it has been estimated that natural juvenile production has been reduced by up to 95% (ODFW and NMFS 2010) as a result of dam construction and operation in most tributaries within the basin. In the mid-1990s, state and federal
fisheries managers began to trap Chinook salmon for transport and release into historical spawning and rearing habitat above upper Willamette River dams to increase natural production and facilitate the species’ recovery. Because habitat quality (Groves and Chandler 1999; Kondolf and Wolman 1993) and return timing (Anderson et al. 2013; Dickerson et al. 2002; Williamson et al. 2010) can influence salmon reproductive success, it is important to determine whether release location and date affect the success of reintroduction efforts.

Salmon often mature at various ages (Quinn 2011), including male mini-jacks and jacks that mature prior to the youngest age class of sexually mature females (Heath et al. 1994). Because matings by early maturing males can increase the effective size of a population (e.g. Araki et al. 2007; Perrier et al. 2014), it may be beneficial to include jacks during reintroductions. Yet this topic has received scarce attention in the literature.

Chinook salmon adults produced in hatcheries may also play an important role in reintroduction programs involving threatened or endangered ESUs. Hatchery origin (HOR) Chinook salmon may be used to found a wild-spawning population and quantify important viability parameters when little is known about the demographic risks associated with a reintroduction (e.g. juvenile survivorship through a reservoir and dam, effects from active transport stress, etc.). HOR fish may also be used to supplement reintroduction programs that use natural origin (NOR) adults, particularly when cohorts return in low numbers, which may help to sustain natural production at a desired level. Anderson et al. (2013) described the reproductive success of HOR Chinook salmon naturally colonizing habitat above a dam accessible following fish ladder construction, but the active transport of HOR salmon when founding and supplementing a reintroduction has not been thoroughly evaluated.

By reintroducing both HOR and NOR adults, more may be learned about fitness differences that often exist between hatchery and wild salmon. There is growing evidence that, on average, HOR salmon are less fit in the wild than NOR salmon, and the effect is stronger among males (reviewed by Christie et
al. 2014). Differences between the size of eggs from HOR and NOR females may affect fitness (Heath et al. 2003), and sperm competition may play a role as well (Flannery et al. 2013; Lehnert et al. 2012).

However, no study to date has clearly identified causal mechanisms for these fitness differences (Christie et al. 2014), though Theriault et al. (2011) hypothesized that two general processes may be relevant: sexual selection on the spawning grounds and/or natural selection early in the life cycle of salmon (i.e. before smoltification). When adult salmon are actively released during a reintroduction program, some phenotypic traits associated with greater fitness can be measured, which may be important for either natural or sexual selection (Milot et al. 2013; Theriault et al. 2011). Thus, recording phenotypic variation and evaluating the effects of that variation on fitness may provide insight into the mechanisms resulting in HOR and NOR fitness differences.

In this study we measured the reproductive success of adult Chinook salmon reintroduced to habitat above Cougar Dam, located on the South Fork McKenzie River, Oregon, USA. We used genetic parentage methods to test for and estimate effects from factors associated with adult reintroductions on their subsequent juvenile production (i.e. reproductive success). Our four main objectives were to 1) test if release date and release location affected the reproductive success (RS) of reintroduced adults; 2) describe RS for jacks among reintroduced cohorts; 3) test if mean RS differed between HOR and NOR Chinook salmon reintroduced in 2010-2011 and; 4) test if adult fork length explained variation in RS.

Methods

Study area

Of all Upper Willamette River tributaries, the McKenzie River typically supports the highest proportion of unmarked Chinook salmon returns (Johnson and Friesen 2010), despite the presence of several dams, including the 158 meter tall Cougar Dam. Construction of Cougar Dam was completed in 1964, blocking access to 40 kilometers of Chinook salmon spawning habitat (Figure 1). Following
construction, attempts to release Chinook salmon above the dam were terminated because of low numbers of adults returning to spawn. In 1993, HOR Chinook salmon were released above the dam, and adults have been released annually since 1995. Most or all of these HOR Chinook salmon were likely produced by the McKenzie River Hatchery (44°07’44”N, 122°14’25”W), where 99% were collected. However, in 2009, a small number (N=39) of adults were collected at the nearby Leaburg Hatchery (44°8’8.63”N, 122°36’32.32”W), which does not produce salmon. We found no evidence of fitness differences between adults collected at these two hatcheries, so we treat them equally in our analyses (data not shown). More recently, NOR Chinook salmon were reintroduced above Cougar Dam, following the construction of a trap and haul facility (hereafter Cougar Trap) completed in 2010. Adults have been collected each year throughout the duration of the spawning migration at the Cougar Trap, with the exception of July 19th-August 6th, 2011, when it was closed due to repairs. Since 2007, adults were released in four locations above Cougar Reservoir. However, the number of release locations (range: 2-4), and number of release dates varied (range: 11-32) between years. (Figure1, Table S1). In addition, the sex ratio (number of males / number of females) was male skewed in all reintroduction years evaluated (Table 1).

Sample collection

The number of adults Chinook salmon reintroduced each year ranged from 731 to 1,386 (Table 1). From 2008-2011, tissue samples were collected from nearly all adults (99%) passed above Cougar Dam (Table 1). All tissue samples were stored in 95% ethanol. Release location, date, and origin (HOR or NOR) were recorded for all adults reintroduced above the dam. Sex was determined by visual assessment of secondary sexual characteristics and recorded. Males were classified as jacks if they were less than 610 millimeters in total length in accordance with Oregon Department of Fish and Wildlife policy. In addition, the sex of jacks was verified using a sex linked marker (see below, 98% concordance
From 2008-2011, 131 jacks were reintroduced. The number of jacks each year ranged from 5-107 jacks (Table 2). Fork length was measured for all adults reintroduced above Cougar Dam in 2011. Reintroduced adults spawned naturally above Cougar Dam each fall, and subyearling juveniles, i.e. fish that were less than one year in age, were sampled with a screw trap at the head of the reservoir in each following year (2009-2012). For each sampling day, the date and total number of juveniles collected in the screw trap were recorded. Overall, the number of juveniles collected (96 ± 197, mean ± 1 standard deviation), tissue sampled (44 ± 52), and genotyped (15 ± 21) varied by day (Table S3). On average, juveniles were collected on 132 ± 22 days per year (Table S3). In all four years, we evaluated the proportion of juveniles genotyped per month and found no difference from the portion of juveniles collected in the screw trap (paired Wilcoxon signed-rank tests, p> 0.05). Mark-recapture studies indicated that the screw trap operated at a mean efficiency of 5% (Zymonas et al. 2010).

**Genotyping**

We isolated total genomic DNA (Ivanova et al. 2006) from reintroduced adults and a sample of juveniles (range: 1,826-2,220) collected in each year of our study (Table 1). We used the polymerase chain reaction (PCR) to amplify 11 highly polymorphic microsatellites from each fish: *Ots201*, *Ots208b*, *Ots209*, *Ots211*, *Ots212*, *Ots215*, *Ots249*, *Ots253*, *Ots311*, *Ots409*, and *Ots515* (Banks et al. 1999; Greig et al. 2003; Naish and Park 2002; Williamson et al. 2002). We also amplified *Oty3* to determine the genotypic sex of reintroduced adults because accuracy has been proven 100% correct among Chinook salmon sampled in the Willamette River (Brunelli et al. 2008). We visualized PCR products on an ABI 3730XL DNA analyzer (Applied Biosystems, Inc., Foster City, CA) and scored genotypes using GeneMapper software (Applied Biosystems, Inc.).

We estimated genotyping error by repeating the genotyping procedure described above on a random sample (n=159, 1% of total dataset) of adults and juveniles collected each year. We compared
re-processed genotypes with originals and estimated error by dividing the number of discordant allele
calls by the total number scored. Genetic analyses were performed at the Coastal Oregon Marine
Experiment Station, Marine Fisheries Genetics Laboratory, Hatfield Marine Science Center in Newport,
Oregon.

Microsatellite characterization and assignment power

We calculated observed and expected heterozygosity (Nei 1987) for each adult cohort and
sample of juveniles to characterize genetic variation among loci genotyped. We then tested for
deviations from Hardy-Weinberg Equilibrium (HWE), and Linkage Disequilibrium (LD) using GENEPOP
(Raymond and Rousset 1995; Rousset 2008) for adult and juvenile year classes separately. This
informed our choice of parent-offspring assignment program (see below: Parent-offspring assignments).
We assessed the power of our suite of microsatellite loci by calculating average non-exclusionary
probabilities for a random single parent, a second parent, and a parent-pair to assign to any given
offspring by chance (Jamieson and Taylor 1997). In addition, we calculated the expected number of false
parent-offspring pairs mismatching at 0, 1, or 2 loci (Christie 2010).

Parent-offspring assignments

We assigned reintroduced adults from 2008-2011 as parents of subyearling juveniles captured in
the screw trap the following year (e.g. 2008 adults were assigned to 2009 juveniles), because most
juveniles typically emigrate from natal streams as subyearlings in this system (Romer et al. 2011).
Deviations from HWE may occur when sampling large numbers of juveniles because some families, and
therefore genotypes, are overrepresented in the sample, known as the Allendorf-Phelps Effect
(Allendorf and Phelps 1981; Waples 1998). We accounted for these deviations by using SOLOMON’s
Bayes method for no known parents, hereafter referred to as SOLOMON, because it carries no assumption with regard to HWE (Christie et al. 2013).

We assigned potential mothers and fathers to juveniles separately to reduce the number of pairwise comparisons. For each run we used the default SOLOMON settings – 1000 simulated data sets and 50,000,000 genotypes because the posterior probabilities for each parent-offspring pair differed by < 0.001 when running the analysis a second time with the same dataset, as recommended by the SOLOMON manual. We accepted all parent-offspring pairs with ≤1 genotypic mismatch that had posterior probabilities <0.05 based on our power analysis (Table S4). On average 76 ± 4% of juveniles assigned to one parent of each sex after parsing the data based on criteria described above. However, on average 12 ± 6% of juveniles were assigned to multiple parents of the same sex. We simulated comparable data (e.g. 800 parents, 2,000 offspring, and 11 microsatellites with 35 alleles per locus) using SOLOMON, and found that in 80% of these cases the true parent could be identified by choosing the parent-offspring pair with the lowest posterior probability. We therefore used this criterion when parsing SOLOMON output. However, SOLOMON assigned a single parent to an offspring without accounting for the second parent’s genotypes. Thus, we could not initially account for both of the putative parents’ genotypes at the same time. We verified that the genotypes of the offspring assigning to both a mother and father were in accordance with Mendelian inheritance, using a function developed by N.S. in R (Supplemental File 1).

Assignment rates

For each juvenile cohort, we calculated the proportion of offspring that assigned to 1) both a mother and a father, 2) only a mother, 3) only a father, and 4) no parent. The reintroduction of Chinook salmon above Cougar Dam began over a decade before the first tissue samples were collected from adults in 2007. Accordingly, we expected that unsampled, residualized or precocial males could be
present in the system. We expected that female Chinook salmon would be less likely to residualize because this life history has rarely been observed in small freshwater systems (but see Romer and Monzyk 2014). Therefore, we expected that some juveniles would have “missing” fathers and few or no juveniles would have missing mothers in our genetic pedigrees.

Release effects

We estimated RS by counting the number of subyearling juveniles within a population sample that were assigned to adults released above Cougar Dam in the previous year. From 2008-2011, we examined factors associated with RS using a negative binomial generalized linear mixed-effect model (GLMM) using the R library lme4 (Bates et al. 2013). We evaluated each year separately because of the variability in release location and release date, as well as variation in the number of juveniles used in parentage assignment among years. For each year, we included sex as a factor, with females reported as the reference. We also included release date as a covariate. We were unable to evaluate the effect of release location across years on RS, as adults were not consistently released in the same locations in different years. In 2011, adults were released at a single location. Thus, we tested for release location effects only in 2008 to 2010. We accounted for similarities among Chinook salmon released on the same date and location with a release group random effects variable. For each reintroduction year, all variables and first order interaction terms were tested individually to determine if they significantly explained variation in RS. We included all significant variables in a single GLMM and used backwards AIC model selection to determine an adequate model to explain variation in RS.

Since 2010, both HOR and NOR Chinook salmon were reintroduced above Cougar Dam. We tested for a difference in mean RS between HOR and NOR Chinook salmon by including an origin (HOR or NOR) predictor, whereby HOR was the reference, when repeating the same GLMM approach described above. In addition, 2011 was the only reintroduction year that had length measurements for
all reintroduced Chinook salmon. Thus, we included length as a covariate in 2011. Finally, we did not include the release group random effects variable in 2011 because adults were released in a single location. We therefore applied a generalized linear model (GLM) with a negative binomial distribution using the R package MASS (Venables and Ripley 2002).

Spawner length has been shown to be positively related to greater fitness. We therefore tested if sex, origin, or sex*origin significantly explained variation in length in 2011 using an ANOVA.

Relative reproductive success

As an alternative to our GLMM approach, we also tested for potential RS differences between HOR and NOR adults using methods described in Araki and Blouin (2005), because these methods account for biases associated with Type A and B error (see Araki and Blouin (2005) for more detail).

Briefly, we permutated 10,000 random relative reproductive success (RRS) values using our estimates of individual RS for HOR and NOR Chinook salmon in R (Supplemental file 1). Equation 14 in Araki and Blouin (2005) requires that Type B error is calculated, which is defined as the rate at which a false parent assigns to an offspring when the true parent is not in the dataset. We estimated Type B error by assigning subyearling juveniles to adults reintroduced after the juveniles were spawned – e.g. 2010 adults assigned to juveniles emigrating from head waters in 2010. We calculated observed RRS, after accounting for errors in assignments and potential missing parents (Araki and Blouin 2005). We determined significance by calculating the frequency of random RRS values less than our observed value. Within each year we compared NOR and HOR Chinook salmon RS differences for the sexes separately, as well as combined years for each sex individually. All critical values were false discovery rate corrected (Benjamini and Hochberg 1995).

All analyses were performed using R version 3.1.1 (R Core Team 2014). Means were reported with ± 1 standard deviation, except for post-hoc tests of first order interaction terms. In these cases, we
report effect estimates ± standard errors. General data analysis was aided by *plyr* (Wickham 2011) and
*reshape2* (Wickham 2007) packages in R.

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**Results**

239 *Microsatellite characterization and assignment power*

We included a total of 11,819 Chinook salmon in our genetic parentage analysis, and we
genotyped 99% (3709/3738) of adults and 100% (8081/8081) of juveniles at ≥ 10 loci (Table 1). We
estimated a 2 ± 1% genotyping error rate for both adults and juveniles, and we observed variation in
genotyping error among loci (2 ± 2%) likely due to mis-scoring and allele dropout. We found that all loci
were out of HWE for the 2008 and 2009 adult cohorts. However, only 6 and 5 loci deviated from HWE in
2010 and 2011, respectively. Similarly, the number of locus pairs in LD was higher in 2008 and 2009,
though average number of locus pairs in LD among years was low (4 ± 3). As expected, we observed all
loci out of HWE and many locus pairs in LD among juvenile samples (21 ± 7). Deviations from HWE were
likely due to large samples sizes because *F*_{IS} values were small (0.005 ± 0.014, Table S5). We found that
observed (0.92 ± 0.01) and expected (0.92 ± 0.01) heterozygosity, as well as the average number of
alleles per locus (34 ± 1) were high and varied little among adult and juvenile datasets used in genetic
parentage analyses (Table 1).

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For the four reintroduction years evaluated, we assigned most (99 ± 2%) juveniles to at least one
parent and an average of 79 ± 7% juveniles to both a mother and a father (Figure S1). We found the
average frequency of juveniles that were missing a mother (14 ± 5%) was nearly twice that of those
missing a father (7 ± 3%).

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Reproductive success variation

We observed positively skewed RS distributions in all four adult cohorts (Figure S2-S5). Means and standard deviations varied between years and sexes, at least in part, due to variation in number of adults reintroduced each year and the number of juveniles genotyped (Table S6). We found evidence that jacks contributed to juvenile production in all years except 2011 (Table 2). However, because we did not genotype all juveniles produced in the system, production by jacks may simply have gone undetected by our study in that year.

Release effects

The factors that were significantly associated with RS varied among the four reintroduction years we evaluated. Regression analysis suggested that in 2008 the mean RS for males was 49% that of females ($p<0.001$, Table 3). In 2009, we only found a small negative relationship between release date and RS ($p<0.001$, Table 3). Overall, from 2008-2010, we found that release location did not significantly explain variation in RS (Table S7).

In 2010, we included an origin term in our analysis because both NOR and HOR Chinook salmon were released in that year and thereafter. We found that sex, as well as the interaction terms origin*sex and origin*release date significantly explained variation of individual RS. All variables were included in the final GLMM based on AIC scores. We found that mean RS for males was 0.43 times that of females ($p<0.001$, Table 3), after accounting for variation explained by other predictors. Our results indicated that the effect of origin on RS was different between the sexes. Accordingly, we tested for the effect of origin on RS for each sex separately. We found that the mean RS for NOR males was 2.2 times that of HOR males ($e^{0.776 \pm 0.237}$, $p=0.001$), whereas mean RS for NOR and HOR females did not differ ($p=0.160$). We also evaluated the effects that release date had on RS of HOR and NOR Chinook salmon separately, because the origin*release date term was significant ($p=0.016$, Table 3). Mean RS of NOR Chinook
salmon decreased ($e^{0.018 \pm 0.006}$, $p=0.003$) each day of the release season. However, release date did not significantly explain variation in RS for HOR Chinook salmon ($p=0.606$).

However, release date did not significantly explain variation in RS for HOR Chinook salmon ($p=0.606$). The predictors origin and length significantly explained variation in RS in 2011. In contrast to results from 2010, we found no significant interaction between origin and sex ($p=0.096$) in 2011. We also found no significant interaction between length and origin that year ($p=0.241$). We found that mean RS for NOR Chinook salmon was 1.5 times that of HOR Chinook salmon when variation in length was not accounted for in the GLM ($p=0.023$). However, we found that origin was not a significant predictor of RS ($p=0.352$) when length was accounted for in the same GLM. Based on AIC scores, we included only length in the final GLM. We found that for each centimeter (cm) increase in length, mean RS increased by 6% ($p<0.001$, Table 3).

We found that mean length for NOR Chinook salmon was 2.0-4.0 cm (95% CI) greater than HOR Chinook salmon ($F_1=58.4$, $p<0.001$) and that males were 1.3-3.3 cm (95% CI) shorter than females ($F_1=9.58$, $p<0.001$). The interaction term sex*origin was not significant ($F_1=0.04$, $p=0.788$), suggesting that the effect of origin on length did not differ between the sexes.

Relative reproductive success

Results from permutation tests provided corroborative evidence for RS differences between HOR and NOR male Chinook salmon. Mean RS for HOR males was significantly less than that of NOR males in 2010 (RRS= 0.46, $p=0.004$), 2011 (RRS= 0.51, $p=0.003$), and when both years were combined (RRS= 0.48, $p<0.001$). We found no significant differences between RS of NOR and HOR females in 2010 (RRS= 1.51, $p=0.938$), 2011 (RRS= 0.89, $p=0.434$), or when both years were combined (RRS= 1.10, $p=0.869$).
Discussion

We examined the effects of factors associated with an active trap and haul strategy on the reproductive success (RS) of Chinook salmon reintroduced above an existing dam on the South Fork McKenzie River, Oregon. We used RS as a measure of success for each Chinook salmon reintroduced above the dam, because RS is a prerequisite to total lifetime fitness, based on adult returns, in future years. Our results are consistent with findings from other studies, which have demonstrated that reintroduction of sexually mature adult salmonids can contribute to overall production of juveniles (Baumsteiger et al. 2008; DeHaan and Bernall 2013). To our knowledge, this is one of two (see Evans et al. In Review) studies that provide insight on effects from release strategies associated with active transport. We found inconsistent evidence that release date affected the success of reintroduced adults and release location did not affect RS in any year evaluated. We are also the first to evaluate the RS of reintroduced jacks and found that jacks contributed to overall juvenile production, thereby mediating gene flow among generations for the reintroduced population. Overall, we consistently found fitness differences between HOR and NOR males, which corroborates findings from other HOR/NOR fitness studies. Finally, we found that length may partially explain why HOR Chinook salmon are less fit than NOR Chinook salmon, which is consistent with hypotheses that fitness differences between HOR and NOR salmon are the result of sexual selection on the spawning ground and/or natural selection in the early life stages of juvenile salmon.

Estimating RS from a sample of juveniles

For Chinook salmon, as with many fishes, estimating mean RS is difficult, particularly during early offspring life stages because hundreds of thousands of juveniles can be produced within a system. In such scenarios, sampling every juvenile is not practical and in many cases, not possible. One alternative is to wait for adult progeny to return in subsequent years, but this approach requires
sampling for many years, and sometimes decades. When management action is required within a shorter time frame, sampling of early-stage juveniles can answer important questions relevant to conservation. Anderson et al. (2011) evaluated the accuracy of such an approach and provided some guidance when estimating mean RS in salmonids using assignments made with adults and juveniles. They noted that three important parameters to control for Type I error are: 1) the number of parents, 2) variance in RS, and 3) the number of offspring that assigned to a parent. Anderson et al. (2011) suggest sampling enough offspring such that estimated mean RS is >6.2. We exceeded a mean RS >6.2 in 4 of the 12 cases in our study (Table S6). Type I error rate may exceed 0.05 in the remaining 8 cases. We note, however, Ford et al. (2012) previously used both adult-juvenile and adult-adult assignments in a salmonid fitness study, and results were consistent between both approaches in all but one comparison. Moreover, our preliminary (unpublished) findings from parentage assignments for returning adult salmon thus far are consistent with adult-to-juvenile results presented here. Nevertheless, a conservative approach would be to interpret our results in light of potential elevated Type I error rates.

There are other factors that could potentially bias results. First the location of the screw trap relative to redds could potentially bias RS estimates. Indeed Anderson et al. (2011) noted that the probability of collecting juveniles likely decreases as the distance between a given redd and the screw trap increases. Similarly, juveniles will not likely be collected if they were produced in a redd located below the screw trap. Finally, juveniles are increasingly able to avoid a screw trap with greater size, though we do not believe this was a significant factor in our study because ongoing life history studies indicate that most juvenile Chinook salmon migrate to Cougar Reservoir as subyearlings.

**Release effects**

Factors that explained variability in RS were inconsistent across years evaluated. Males were on average less fit than females in 2008, which was likely driven by the male-skewed sex ratio (2 males:1...
female) in that year. In 2009, we only found that mean RS decreased slightly as Chinook salmon were released later in the season. Similarly, a small negative relationship between release date and RS was evident in 2010, though it was only for NOR Chinook salmon. Our results corroborate findings from two previous studies of Chinook salmon that reported a negative relationship between fitness and timing of entry to spawning grounds. Similar to our findings, release date effects observed in those studies were inconsistent among years (Anderson et al. 2013; Williamson et al. 2010). Dickerson et al. (2005) found that early-arriving pink salmon (O. gorbuscha) males were also more successful, which may be explained by a density-dependent process. Males that arrive to the spawning grounds earlier may experience less competition for mates, but as the season progresses the operational sex ratio increases. Individual mating success therefore decreases because of increased competition among males (Quinn 2011).

Results observed here and by others suggest that; overall, release date is a weak and inconsistent predictor of RS.

Water velocity and depth, as well as gravel size are important factors for Chinook salmon redd site selection (Groves and Chandler 1999; Kondolf and Wolman 1993). Moreover, spawning habitat quality can also affect the early survival of juvenile salmonids (Quinn 2011). However, we found no significant effect from release location on RS. This is likely because Chinook salmon disperse throughout the river after release, and do not necessarily spawn near their release sites. A telemetry study of female Chinook salmon reintroduced above Cougar Dam in 2010 found that females moved a minimum distance of between 4.2 -17.1 kilometers (Zymonas et al. 2010). Males were not evaluated by Zymonas et al. (2010) and males are likely to move more than females on spawning grounds, because they are not anchored by the construction and defense of a redd.
Our results show that the reintroduction of jacks can help contribute to juvenile production and, more importantly, contribute to gene flow among cohorts of Chinook salmon reintroduced above Cougar Dam. Gene flow among cohorts will help to increase and/or buffer effective population size of the population over time (e.g. Araki et al. 2007; Perrier et al. 2014). In our study system, gene flow is already achieved by overlapping age-4 and age-5 Chinook salmon adults returning to spawn. But, jacks are likely more important to gene flow between generations in coho (*Oncorhynchus kisutch*) and other migratory fishes with more constrained age at maturity.

**HOR and NOR Fitness**

Our findings generally support the growing body of evidence that HOR salmon are less fit in the wild than NOR salmon (reviewed by Christie et al. 2014). We found that male HOR Chinook salmon were less fit than NOR Chinook salmon using two different approaches: GLMM/GLM and an unbiased RRS permutation test. However, no consistent fitness differences were found between HOR and NOR females. In particular, we found no fitness differences between HOR and NOR females released in 2010 using either approach, but results from 2011 indicated that mean RS for NOR Chinook salmon (both males and females, combined) was 1.5 times greater than HOR Chinook salmon. Interestingly, this origin effect in 2011 was not significant when we accounted for RS variation explained by length. Indeed, NOR adults had a mean length that was 2.0-4.0 cm (95% CI) greater than HOR adults. Together, these results suggest that fitness differences observed between HOR and NOR Chinook salmon released above Cougar Dam may be explained by mean fork length, which differs significantly according to origin.

We did not directly test the hypotheses posited by Theriault et al. (2011), that fitness differences between HOR and NOR salmon are likely caused by 1) effects from sexual selection on the spawning ground and/or 2) natural selection during juvenile early life stages. However, our results are
consistent with both hypotheses. First, length has been shown to be an important trait under sexual selection. In particular, female Chinook salmon will delay spawning with smaller males, which likely indicates that they prefer to mate with larger males (Berejikian et al. 2000). Second, larger females typically dig deeper reds, are less susceptible to superimposition, and are more fecund than smaller females (Hawke 1978; Healey and Heard 1984; Quinn 2011). Cumulatively, larger females may have greater fitness because their progeny may experience less mortality associated with the location and quality of the redd where they were reared.

Some have suggested that hatchery practices relax selection for larger individuals (Hankin et al. 1993; Ricker 1981). In addition, numerous authors have reported positive relationships between body size and RS for both sexes in Pacific salmon (Berejikian et al. 2001; Fleming and Gross 1992; Schroder et al. 2008). Therefore, HOR salmon are likely to be less fit compared to NOR salmon because, on average, they are smaller in size. Indeed, Ford et al. (2012) found that low RS for HOR Chinook salmon could be explained by a higher frequency of precocial (younger and smaller) males present among HOR spawners, relative to the NOR population. In addition, Milot et al. (2013) found that HOR Atlantic salmon (Salmo salar) returned earlier than NOR Atlantic salmon, and that the difference in the number of years at sea contributed to lower HOR fitness. Although precocial male life history is relatively rare in the Willamette River basin, Johnson and Friesen (2013) found that HOR Chinook salmon length has decreased over time within the basin. Our results therefore corroborate findings from Ford et al. (2012) and Milot et al. (2013) because we found that HOR Chinook salmon were on average smaller than NOR Chinook salmon, which resulted in an overall reduction in fitness. However, the relationship between size and RS that we report warrants further investigation because it is based upon data from a single reintroduction year, and we do not have age information for all reintroduced adults.

Based on our HOR/NOR findings, it may be prudent to limit the number of HOR males in the reintroduction program, because HOR males may be maladapted to the “wild” environment (e.g.
Christie et al. 2012) and therefore may lower the RS of NOR females. Moreover, juvenile production is not typically constrained by the number of male spawners, in terms of gametes, such that a reduction in the number of HOR males used for reintroduction would not likely limit population productivity. However, research that evaluates potential differences in mate pair reproductive success and that manipulates the number of HOR males to determine its effect on overall productivity is warranted before such management decisions could be made.

Successful reintroduction programs above dams will likely prove important to the recovery of many migratory fishes, as they can increase the spawning distributions and natural production. In our study area, reintroduction provided Chinook salmon the opportunity to spawn and rear in headwater reaches of the McKenzie River for the first time in 50 years. We demonstrated that reintroduction of Chinook salmon above dams by active transport can increase natural production. This reintroduction strategy may be effective for the conservation of other migratory fishes (but see Pelicice et al. 2014).

Our study found that sex, origin, as well as release date can be important factors influencing the RS of reintroduced Chinook salmon and, therefore, overall population productivity. Observed fitness differences between HOR and NOR male Chinook salmon can likely be explained, at least in part, by the smaller size of HOR fish. Further research on the relationships between age at maturity, length, and fitness of HOR and NOR salmon is need to better understand HOR and NOR fitness differences. Overall, our results provide information on factors that are important for the reintroduction of a migratory fish species into historical habitat despite the continued presence of a dam.
Acknowledgments

We thank Oregon Department of Fish and Wildlife (ODFW) staff for collecting the McKenzie River Chinook salmon tissue samples and the Army Corps of Engineers for sampling adult fish at the trap and haul facility. We are grateful for Fred Monzyk, Jeremy Romer, Khoury Hickman, and their colleagues for sampling juvenile salmon captured in the screw trap. We thank Kurt Kremers (ODFW, McKenzie Hatchery) for the hatchery adult outplant samples and Mark Christie for advice on SOLOMON. In addition, we appreciate helpful comments from Melissa Evans on previous drafts of this manuscript. This work was funded by the U.S. Army Corps of Engineers, Portland District, with administrative assistance provided by David Griffith.


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Zymonas, N.D., Tranquilli, J.V., and Hogansen, M. 2010. Monitoring and Evaluation of Impacts to Bull Trout (Salvelinus confluentus) and Spring Chinook (Oncorhynchus tshawytscha) in the South Fork McKenzie River from Construction of Water Temperature Control Facilities at Cougar Dam, Oregon. Oregon Department of Fisheries and Wildlife, U.S Army Corps of Engineers.
Table 1. Summary of the number of adult and juvenile Chinook salmon genotyped at ≥ 10 and < 10 loci, the number of adults missing a tissue sample (Missing Tissue Sample), and genetic variation observed among cohorts sampled. The sex ratio (number of males / number of females), mean number of alleles per locus (A), mean observed heterozygosity (H_o), and mean expected heterozygosity (H_e) are described.

<table>
<thead>
<tr>
<th>Year</th>
<th>Type</th>
<th>Loci genotyped</th>
<th>Missing tissue sample</th>
<th>Sex ratio</th>
<th>A</th>
<th>H_o</th>
<th>H_e</th>
</tr>
</thead>
<tbody>
<tr>
<td>2008</td>
<td>Adult</td>
<td>873 ≥ 10</td>
<td>0</td>
<td>2</td>
<td>33.3</td>
<td>0.92</td>
<td>0.92</td>
</tr>
<tr>
<td>2009</td>
<td>Adult</td>
<td>1373 ≥ 10, 12 &lt; 10</td>
<td>1</td>
<td>1.3</td>
<td>35.1</td>
<td>0.91</td>
<td>0.93</td>
</tr>
<tr>
<td>2010</td>
<td>Adult</td>
<td>738 ≥ 10, 9 &lt; 10</td>
<td>1</td>
<td>1.8</td>
<td>34.3</td>
<td>0.93</td>
<td>0.92</td>
</tr>
<tr>
<td>2011</td>
<td>Adult</td>
<td>725 ≥ 10, 5 &lt; 10</td>
<td>1</td>
<td>1.3</td>
<td>35.2</td>
<td>0.92</td>
<td>0.92</td>
</tr>
<tr>
<td>2009</td>
<td>Juvenile</td>
<td>2001 ≥ 10</td>
<td>-</td>
<td>-</td>
<td>31.5</td>
<td>0.91</td>
<td>0.92</td>
</tr>
<tr>
<td>2010</td>
<td>Juvenile</td>
<td>1826 ≥ 10</td>
<td>-</td>
<td>-</td>
<td>33.8</td>
<td>0.92</td>
<td>0.92</td>
</tr>
<tr>
<td>2011</td>
<td>Juvenile</td>
<td>2034 ≥ 10</td>
<td>-</td>
<td>-</td>
<td>33.6</td>
<td>0.92</td>
<td>0.92</td>
</tr>
<tr>
<td>2012</td>
<td>Juvenile</td>
<td>2220 ≥ 10</td>
<td>-</td>
<td>-</td>
<td>33.1</td>
<td>0.91</td>
<td>0.92</td>
</tr>
</tbody>
</table>

Table 2. Summary of the number (N) and mean (± one standard deviation) reproductive success (RS) estimates for Chinook salmon jacks released above Cougar Dam from 2008-2011.

<table>
<thead>
<tr>
<th>Year</th>
<th>N</th>
<th>RS</th>
</tr>
</thead>
<tbody>
<tr>
<td>2008</td>
<td>13</td>
<td>2.0 ± 2.6</td>
</tr>
<tr>
<td>2009</td>
<td>107</td>
<td>0.8 ± 1.6</td>
</tr>
<tr>
<td>2010</td>
<td>5</td>
<td>0.0 ± 0.0</td>
</tr>
<tr>
<td>2011</td>
<td>6</td>
<td>6.8 ± 9.1</td>
</tr>
</tbody>
</table>
Table 3. Summary of predictors for final reproductive success generalized linear mixed-effects model/generalized linear model for Chinook salmon released above Cougar Dam from 2008-2011. We used hatchery origin Chinook salmon and females as references for origin and sex, respectively. Results are described for each year separately.

| Year | Predictor         | Estimate | Std. Error | z value | Pr(>|z|) |
|------|------------------|----------|------------|---------|---------|
| 2008 | Sex              | -0.718   | 0.135      | -5.329  | < 0.001 |
| 2009 | Release date     | -0.018   | 0.004      | -4.442  | < 0.001 |
| 2010 | Sex              | -0.844   | 0.202      | -4.178  | < 0.001 |
|      | Origin           | 3.787    | 1.781      | 2.126   | 0.033   |
|      | Release date     | 0.001    | 0.004      | 0.240   | 0.810   |
|      | Sex*Origin       | 1.119    | 0.404      | 2.769   | 0.006   |
|      | Origin*Release date | -0.018 | 0.008   | -2.402  | 0.016   |
| 2011 | Length           | 0.055    | 0.012      | 4.757   | < 0.001 |
Figure 1. Cougar Dam (solid black horizontal line) is located on the South Fork McKenzie River, Oregon. Locations of adult Chinook salmon release sites and screw trap used to collect juveniles are indicated.