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Executive Summary

Spring Chinook Salmon spawning in the upper Warm Springs River basin have demonstrated a relatively sudden increase in the ratio of annual adult escapement to total redd count. Prior to 2010 the ratio averaged 4 fish per redd, but since then the average jumped to 12 fish per redd. The recent high fish per redd estimates infer that a large proportion of the escapement failed to successfully spawn, although the alternative explanation that the redd counts were substantially underestimated is also possible. To confirm which explanation is the more likely, a genetic sibship analysis was conducted on a sample of smolts from broodyears 2015 and 2016 to estimate the effective number of breeders (Nb). The Nb estimate for each broodyear was only 21% and 12% of the total escapement, respectively, and that the ratio of Nb divided by 4, the historic fish per redd value, provided an estimate for the expected number of redds that was very similar to the observed number in each broodyear. The results, therefore, do not support undercounting of redds, but instead that there was a high rate of reproductive failure of adults in these broodyears.

Introduction

Spawning behavior of spring Chinook Salmon typically involves a female depositing essentially all of her eggs in a single redd (Quinn 2005; Murdoch et al. 2008), with the majority of eggs generally being fertilized by a single dominant male, plus smaller proportions fertilized by one or two satellite males, which may also participate in fertilization of eggs from other females (Quinn 2005). In addition to these reproductively successful spawners among the escapement will be a number of fish that fail to successfully spawn. In wild spring Chinook Salmon populations not known to be subject to high pre-spawn mortality and in situations where competition for spawning habitat is not limiting, the proportion of adults in the escapement that successfully engage in spawning will be relatively high, and the ratio of the total number of redds to the total adult escapement in a given year is expected to be relatively low. In a multiple year review of monitoring data for five Salmon and Grande Ronde river populations of spring/summer Chinook Salmon, fish per redd ratios were generally in the range of 2 to 4 (Kucera and Orme 2006; see Table 5, p.33). Similar fish per redd ratios for Mid-Columbia River spring Chinook Salmon populations were cited by Mullan (1992), Sampson et al. (2016; Appendix B, Table 3, p.7), and Murdoch et al. (2010). In rivers where the infrastructure is not present for direct enumeration of adult escapement, fisheries managers often use a standard fish per redd factor to expand annual redd counts in order to obtain estimates for total escapement.

While fish per redd ratios provide a relative measure of the proportion of successful spawners within the adult escapement, a direct measure of this proportion can be obtained through genetic parentage analysis. In rivers/streams where a weir exists, a total count of adults passed upstream for natural spawning can be obtained and tissue samples can be collected from each fish. If tissue samples are also collected from a representative sample of their progeny, e.g.,

from juvenile out-migrants, the two sets of tissues can be genotyped for a suite of DNA markers and genetic analyses performed to identify the parents of each progeny. The number of different adults that parented one or more progeny can then be summed to quantify the number of reproductively successful fish within the escapement (Nb, effective number of breeders). Nb divided by the number of redds will then give an estimate for the average number of successful spawners per redd. This value will be somewhat lower than a fish per redd estimate based on the total escapement, as adults which did not successfully spawn are excluded. However, the difference will not be great if as described above, pre-spawn mortality and competition for spawning habitat are both minimal.

Genetic parentage analysis requires the infrastructure for a complete sampling of the potential parents, which is often not available. Nonetheless, one of the software packages commonly used to perform parentage analyses (Colony; https://www.zsl.org/science/software/colony; Jones and Wang 2010) has the option, in the absence of samples from the potential parents, to derive an estimate of Nb. With genotypes from a representative sample of the progeny identifiable to a particular broodyear, Colony can implement a maximum likelihood assignment of the fish to full- and half-sibship groups, from which an estimate of Nb can be obtained (Wang 2009). While certainty of the Nb estimate derived in this manner is less than that obtained from direct genetic parentage assignment, simulations indicate that if the number of progeny sampled is similar to the actual Nb, the estimate from Colony will be relatively accurate, with accuracy increasing with further increase in sample number (Ackerman et al. 2016).

For several decades, the population of spring Chinook Salmon in the upper basin of the Warm Springs River (upstream of the Warm Springs National Fish Hatchery; WSNFH; rkm 16.4) has been monitored. In-migrating adults are captured in a trap at an impassable weir located adjacent to the WSNFH and a count is made of all natural origin adults which are then released upstream for natural spawning each year. No hatchery origin adults are passed upstream. Additionally, personnel with the Fisheries Research, Monitoring, and Evaluation (RM&E) Program of the Confederated Tribes of the Warms Springs Reservation of Oregon (CTWSRO) have conducted systematic surveys of the spawning grounds in the upper basin, providing a count of redds observed annually. These data are monitored to assess trends in escapement and spawning, as well as trends in relative spawning success based on fish per redd ratios.

Unfortunately, the trends in the data observed over recent years have been discouraging. Since the year 2000, there has been a marked decline in both adult escapement of natural origin spring Chinook Salmon to the Warm Springs River, and in the number of redds counted during annual spawning ground surveys in the upper basin (Figures 1a and 1b). Of additional concern is a recent three fold increase in the ratio of the redd count to annual escapement. Prior to 2010, the escapement and redd count data provided a relatively dependable average of 4 fish per redd (range 2 to 6; Figure 1c), similar to ratios for other spring Chinook populations (see above). However, since 2010 this ratio has averaged 12 fish per redd, inferring that there has been a substantial increase in the proportion of the adults that failed to successfully spawn since that time. The reduction in spawning success might be indicative of a recent increase in
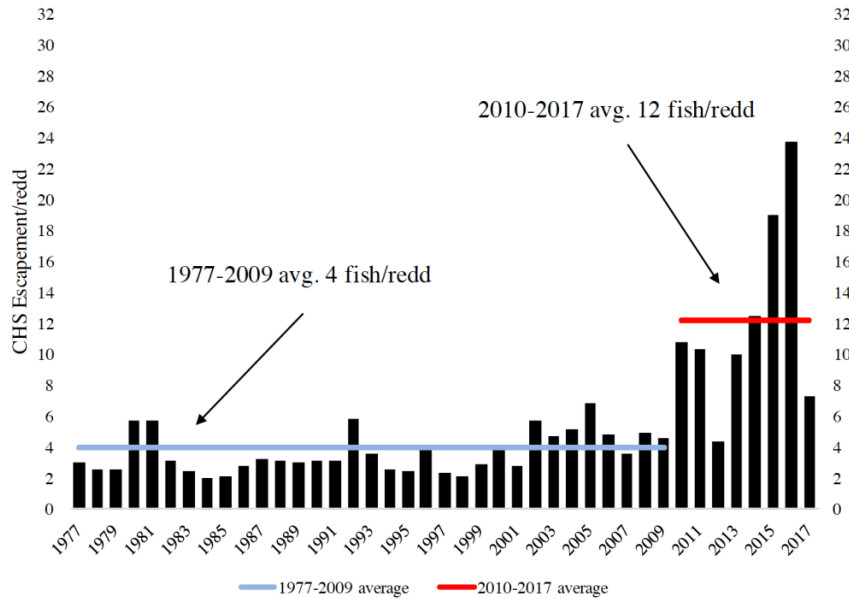
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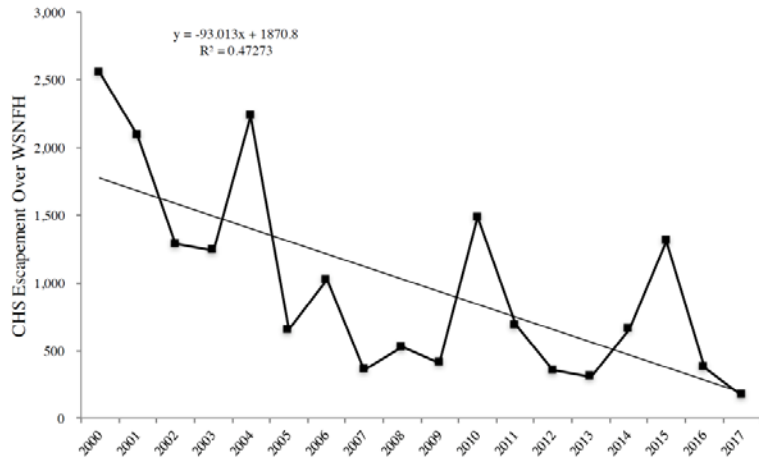
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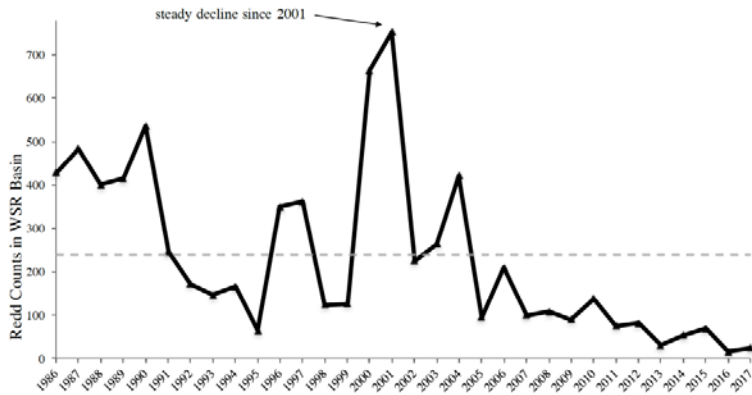
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Figure 1 – Annual escapement of spring Chinook Salmon passed upstream of the Warm Springs River National Fish Hatchery weir, annual redd counts in the upper basin of the Warm Springs River, and the annual ratio of fish per redd.

pre-spawn mortality (e.g., due to disease, or physiological stress associated with high temperature or other hydrologic conditions, etc.). Alternatively, the possibility exists that the ratio is misleading due to undercounting of the redds; i.e., that the spring Chinook Salmon have recently expanded their spawning into areas of the basin outside the standardized spawning ground survey reaches, and that the actual fish per redd ratio is much closer to 4, as observed in previous years. To assess this latter possibility of redd undercounting we performed a Nb estimate analysis of fish from two recent broodyears, 2015 and 2016. These two broodyears are characterized with two of the highest fish per redd estimates observed recently, 19 and 24, respectively.

Methods

For many years the CTWSRO Fisheries RM&E Program has conducted a mark-recapture study of age 1+ spring Chinook Salmon collected in a rotary screw trap operated on the Warm Springs River, to estimate smolt out-migration abundance. Beginning in 2016, they began collecting tissue samples from a subset of the juveniles, including smolts from broodyear (BY) 2015 (n=1,218) collected in November 2016 and in March through May 2017, and from BY 2016 (n=673) collected in October through December 2017 and in March through June 2018. The samples were collected principally in a trap located approximately 0.5 km upstream from the WSNFH weir, with smaller numbers collected in traps operated at two different locations further upstream. Fin clips from fish were adhered within a cell of a 10x10 grid preprinted on a sheet of Whatman filter paper, associated biodata were recorded (collection location, date, and fork length), and the tissue samples were archived. As samples had not been collected from the adults for these two broodyears, precluding performance of genetic parentage analyses, it was decided to estimate Nb by analysis of genotypes for smolts with the option available in Colony to assign sibship groups.

In 2018, the tissue samples were sent to CRITFC geneticists at the Hagerman Fish Culture Experiment Station, Hagerman ID. DNA was extracted using the Chelex 100 method (Sigma-Aldrich, St Louis, MO). The DNA was then genotyped for a panel of 298 single nucleotide polymorphism (SNP) markers plus one sex marker, using the "genotyping-in-thousands by sequencing" (GT-seq) technique (Campbell et al. 2015). To assure sufficient power for identification of sibling groups, only genotypes which surpassed a 90% threshold for successfully genotyped loci (n>268) were included for subsequent analysis.

Among the total of 1,891 smolt tissue samples, approximately 30% did not meet the 90% threshold. Tissue degradation was likely the cause for genotyping failure for some of these samples, however, the majority did successfully genotype for a large subset of the loci (190 to 220), though they also showed extremely low levels of heterozygosity (<1%) for these loci. The average heterozygosity for the panel of Chinook Salmon markers is approximately 20%. It was subsequently determined that these samples were from Coho Salmon. Some level of misidentification to species in the field is not unexpected; Coho Salmon smolts were out-migrating concurrently with Chinook Salmon, and juveniles of the two species bear a strong

resemblance to each other. Additionally, it has been observed previously that Coho Salmon will successfully genotype for many of the Chinook Salmon markers, though at lower levels of heterozygosity. A second threshold of >12% heterozygosity, therefore, was set in order to exclude with high certainty samples that were not from Chinook Salmon (Figure 2). Among the samples that surpassed the locus number and % heterozygosity thresholds were 12 pairs with identical genotypes. These likely involved fish that had been tissue sampled twice, and one genotype from each pair was deleted from the database. The final number of sample genotypes submitted for Nb analysis in Colony was 933 for BY 2015 and 347 for BY 2016.
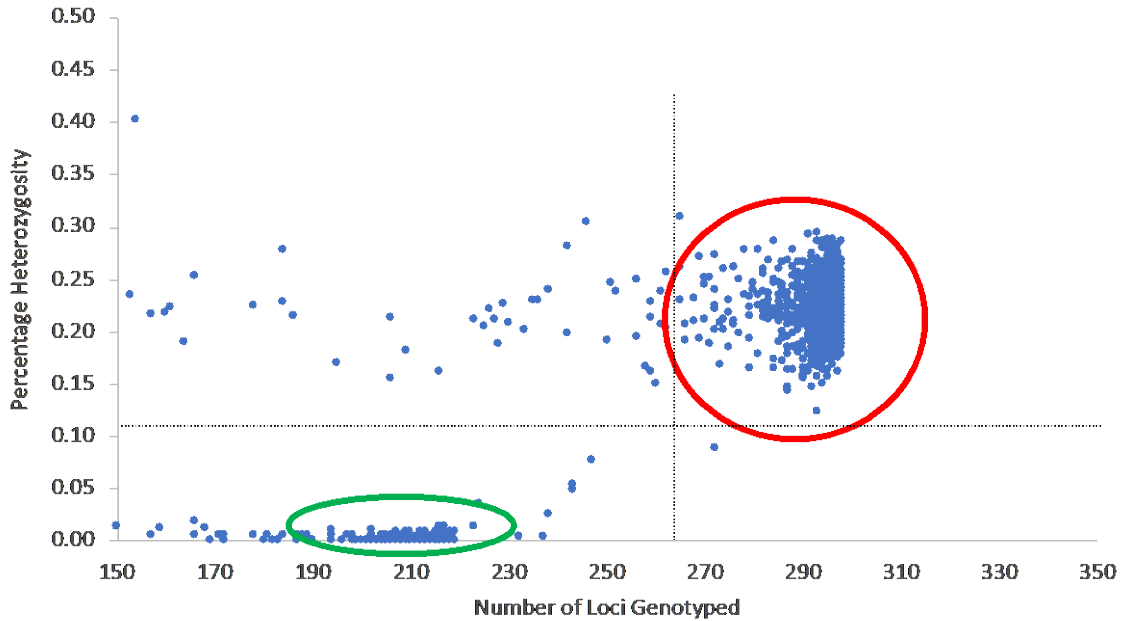

Figure 2- Scatterplot representing the relationship between the number of loci that successfully genotyped and the percent heterozygosity across all loci for each fish. The red circle denotes the main Chinook smolt group and the green circle represents the main Coho smolt group. The dotted line represents the cut-offs used to separate Coho from Chinook smolts.

Results and Discussion

Average Nb estimates and Nb/4 ratios per broodyear are provided below, following observed data for escapement, redd count, and fish per redd.

Broodyear	Escapement	Redd Count	Fish per Redd	Nb Estimate	Nb/4
2015	1,300	70	19	277	69
2016	400	15	24	49	12

In broodyears 2015 and 2016, the Nb estimates were only 21% and 12%, respectively, of the total number of adults passed above the WSNFH weir for natural spawning in the upper Warm Springs River basin. Additionally, the redd count for each broodyear was similar to the value of the Nb estimate divided by 4 – the historic fish per redd ratio. As such, the data do not support

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the possibility of significant undercounting of redds, but instead confirm the inference from the original high fish per redd values that a large proportion of the adult escapement failed to successfully spawn.

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