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Marc A. Johnson[a] & Thomas A. Friesen[a]
[a] Oregon Department of Fish and Wildlife, Corvallis Research Laboratory, 28655 Highway 34, Corvallis, 97333, Oregon, USA
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**ARTICLE**

# Genetic Diversity and Population Structure of Spring Chinook Salmon from the Upper Willamette River, Oregon

**Marc A. Johnson* and Thomas A. Friesen**
*Oregon Department of Fish and Wildlife, Corvallis Research Laboratory, 28655 Highway 34, Corvallis, Oregon 97333, USA*



*Abstract*

**Effective management of Pacific salmon requires an accurate understanding of both population genetic diversity and structure. Spring Chinook Salmon *Oncorhynchus tshawytscha* from the upper Willamette River (UWR), Oregon, are listed as threatened under the U.S. Endangered Species Act, and although this evolutionarily significant unit is recognized to be distinct from other Columbia River stocks, genetic relationships among its constituent hatchery and wild populations remain obscure. We used genotypic data from 13 microsatellite loci to test whether hatchery populations of UWR spring Chinook Salmon are most similar to wild populations within the same subbasin, or whether hatchery populations from different subbasins are more similar to each other than to local wild populations. We also tested for differences between the genetic diversities of hatchery and wild populations, as measured through heterozygosity and allelic richness. Our results suggest that populations are weakly structured among subbasins and, in all cases, hatchery populations are genetically most similar to local wild populations. We also found heterozygosity to be higher ($P = 0.009$) in hatchery populations (median, 81.5%) than in wild populations (median, 75.2%), but observed no significant difference with respect to allelic richness ($P = 0.406$). We conclude that hatchery-origin UWR spring Chinook Salmon represent genetically appropriate founder populations for ongoing reintroduction programs and recommend that the conservation and recovery of this stock proceed through management actions developed specifically for each subbasin. We further recommend that current restrictions on hatchery stock transfers among UWR subbasins be continued to preserve extant population genetic structure.**

For several decades, fisheries managers have used genetic data to gain insight to the population structure of Pacific salmon species (*Oncorhynchus* spp.). Genetic information has been used to delineate evolutionarily significant units (ESUs) and develop recovery plans for threatened and endangered species. Although ESU-level information may address broad-scale management questions, important genetic diversity can be structured at finer spatial scales (see Banks et al. 2000; Myers et al. 2006; Neville et al. 2007). An understanding of such fine-scale structure is important because human activities, such as habitat alterations and hatchery operations, can potentially impact the genetic and life history diversity that lends resilience to salmon populations (Eldridge et al. 2009).

The Willamette River is the second largest tributary of the Columbia River in terms of average discharge and is contained entirely within the state of Oregon. The upper Willamette River (UWR) basin is defined, in part, by the 12-m-high basalt shelf at Willamette Falls (Figure 1) that, before construction of a fish ladder in 1882, was only traversable by salmon during high flows of late winter and spring (Myers et al. 2006). Although Willamette Falls historically excluded fall-run Chinook Salmon *O. tshawytscha* from the upper basin, spring Chinook Salmon are native to the UWR, but have declined in numbers to a fraction of their historical, natural abundance. Accordingly, UWR spring Chinook Salmon were listed as threatened under the U.S. Endangered Species Act in 1999, and this status was reaffirmed in 2005 and 2010 (Ford 2011). Diverse factors have contributed to the decline of UWR spring Chinook Salmon (NMFS 2008). Foremost among these, the construction and continued operation of flood-control and hydroelectric dams on all major UWR

*Corresponding author: marc.johnson@oregonstate.edu
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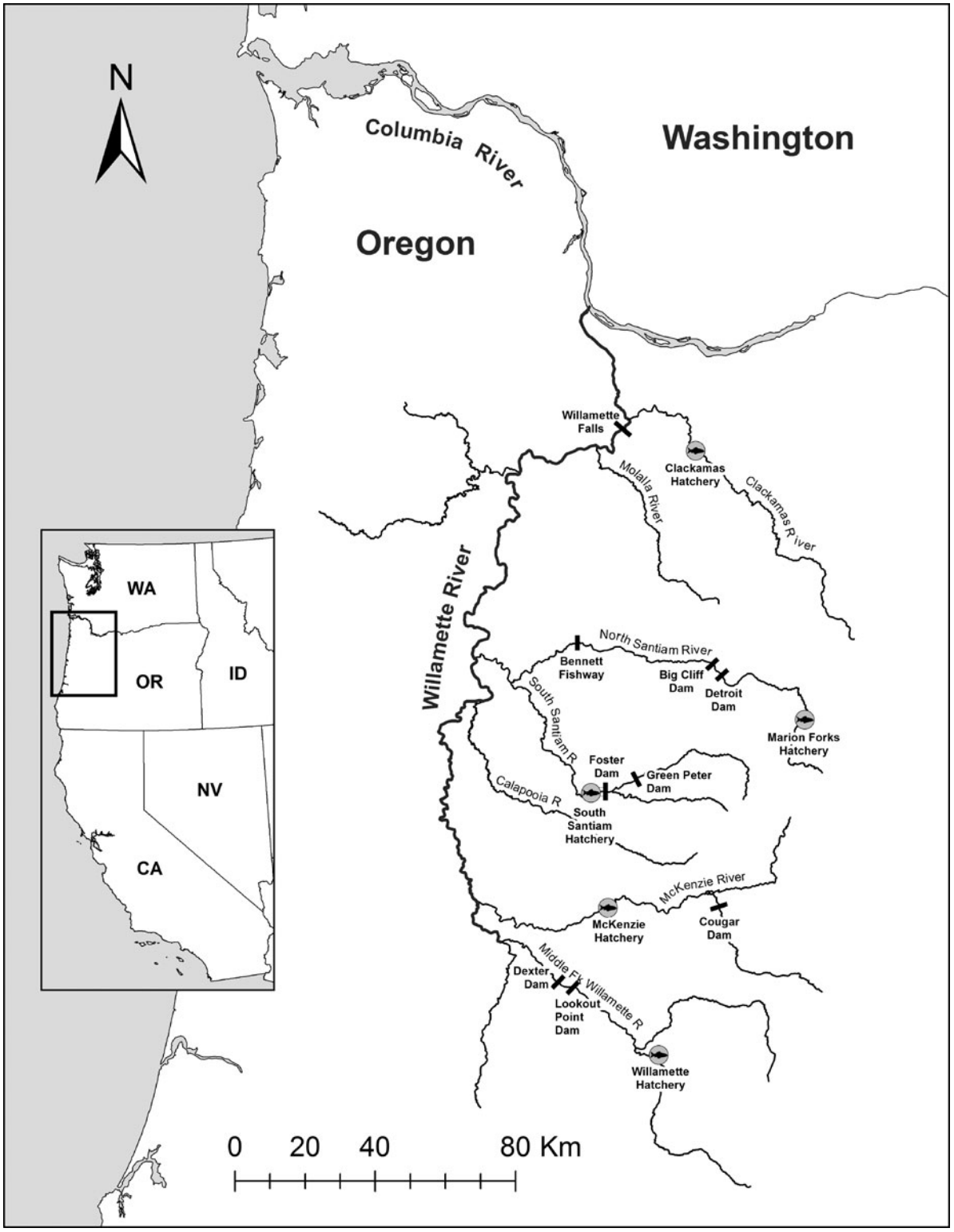


FIGURE 1. The Willamette River basin and collection sites for clipped (hatchery origin) Chinook Salmon tissue samples. Samples were collected from unclipped fish throughout the labeled tributaries.

tributaries (Figure 1) has impeded adult and juvenile migrations and altered the flow, temperature, and other qualities (i.e., substrate, depth, total dissolved gases) of critically important riverine habitats (NMFS 2008). To mitigate for these negative impacts on native fish and fisheries, five state-operated hatcheries produce UWR spring Chinook Salmon for harvest and reintroduction programs. However, introgression from hatchery-produced salmon may pose serious genetic risks to the recovery of natural populations (Waples 1991; Levin et al. 2001; Araki et al. 2008), especially if hatchery populations are genetically less diverse than or significantly diverged from local wild populations (Ryman and Laikre 1991; Baskett and Waples 2012). This potential risk is of particular concern to the management of UWR spring Chinook Salmon because large numbers of adult hatchery fish are released above UWR dams to expand the species' spawning distribution into otherwise vacant or underutilized habitats.

In their review of historical population structure, Myers et al. (2006) identified seven historically independent populations of UWR spring Chinook Salmon native to the Clackamas, Molalla, North Santiam, South Santiam, Calapooia, McKenzie, and Middle Fork Willamette rivers. Clackamas River spring Chinook Salmon are included in the UWR spring Chinook Salmon ESU, even though the Clackamas and Willamette rivers join below the geographic boundary of the upper basin (Willamette Falls, Figure 1). Collectively, UWR spring Chinook Salmon are among the most genetically distinct of Columbia River Chinook Salmon (Waples et al. 2004; Narum et al. 2010; Moran et al. 2013). However, hatchery stock transfers, which were common prior to 1997 (Kostow 1995), may have weakened or eliminated genetic structure among spring Chinook Salmon populations within the basin. Myers et al. (2006) provided evidence of some genetic structure for UWR populations, though those authors acknowledged that their results may have been compromised by the inclusion of juvenile samples, which can produce skewed patterns of population structure due to overrepresentation of some family groups, i.e., the Allendorf–Phelps effect (Allendorf and Phelps 1981; Waples 1998). Indeed, their findings suggested that some hatchery populations were more closely related to wild spring Chinook Salmon from distant subbasins than to their neighboring wild population. However, it is unclear whether their results reflected actual relationships among UWR spring Chinook Salmon populations or if the Allendorf–Phelps effect masked true population structure.

In this study, we use the UWR spring Chinook Salmon population designations identified by Myers et al. (2006) and data from adult fish characterized at 13 standardized microsatellite loci (Seeb et al. 2007) to determine whether (1) UWR hatchery spring Chinook Salmon are most closely related to local wild populations and (2) UWR hatchery spring Chinook Salmon are genetically less diverse than wild populations, as measured through heterozygosity and allelic richness.

## METHODS

*Sample collections and microsatellite genotyping*.—From June to October 2011 we collected otolith and fin tissue samples from carcasses of unclipped (adipose fin), presumably wild-origin, adult spring Chinook Salmon from the Clackamas, North Santiam, South Santiam, Molalla, McKenzie, and Middle Fork Willamette rivers (Figure 1). Since 1997, all spring Chinook Salmon from UWR hatcheries have been adipose fin-clipped to allow for selective harvest of hatchery-produced fish. These fish are also passively marked with programmed temperature oscillations that produce recognizable otolith banding (Volk et al. 1999). We examined otoliths from all unclipped Chinook Salmon sampled for this study to confirm wild-origin status. During the same year, we also collected fin tissue samples from adult hatchery-origin spring Chinook Salmon at the Clackamas, Marion Forks (North Santiam), South Santiam, McKenzie, and Willamette hatcheries. We also included samples from spring Chinook Salmon from the Catherine Creek Hatchery in our analyses. This geographically distant population from the Grande Ronde River of the upper Columbia River basin served as the outgroup of our study to provide broader context to genetic distance estimates for UWR spring Chinook Salmon populations. Fall Chinook Salmon are rare or absent from most UWR tributaries and typically spawn later than spring Chinook Salmon, and thus, experienced surveyors made visual assessments to avoid including fall Chinook Salmon among samples. All tissue samples were stored in labeled vials containing 95% ethanol.

We used the protocol of Ivanova et al. (2006) to isolate whole genomic DNA from Chinook Salmon tissue samples. We used touchdown PCR (Korbie and Mattick 2008) with fluorescently labeled primers to amplify 13 microsatellite markers: *Ots208*, *Ots213*, *Ots9*, *Ots211*, *Ogo4*, *OtsG474*, *Ssa408*, *Ogo3*, *Ots3*, *Ots212*, *Oki100*, *Ots201*, *Oki100*, *Ots201*, and *Omm1080*. Primer sequences for these markers are provided by references in Seeb et al. (2007), and reaction conditions are available from the authors upon request. All PCR products were separated and visualized on an ABI 3730XL DNA Analyzer (Applied Biosystems) and scored by size against a 500-bp standard with GeneMapper software (Applied Biosystems).

*Analyses*.—To reduce genotyping error effects from low quality DNA samples (Pompanon et al. 2005), we excluded all samples that amplified at fewer than 7 of 13 microsatellite loci from our analyses. We used the program GENETIX (Belkhir et al. 2004) to produce estimates of observed heterozygosity ($H_o$) and expected heterozygosity ($H_e$) for all study populations. We used a Kruskal–Wallis ANOVA on ranks to test for basin-wide difference between observed heterozygosities of hatchery and wild populations, and we used a Friedman rank sum test to compare $H_o$ values for hatchery and wild population pairs within subbasins. We used the program GENEPOP to perform Hardy–Weinberg equilibrium (HWE) exact tests (Haldane 1954) and score (a.k.a. *U*) tests (Raymond and Rousset 1995;

Rousset 2007) to detect locus-specific heterozygosity excesses or deficits within each population. We also used GENEPOP to perform exact tests for linkage disequilibrium (LD) between all locus pairs within each population. We controlled the false discovery rate (FDR) of multiple tests according to the methods of Benjamini and Hochberg (1995; also see Narum 2006) and used a maximum (unadjusted) critical value of $\alpha = 0.05$ to assess significance.

Using our microsatellite data and the program GENETIX, we estimated $F_{ST}$ ($\theta$; Weir and Cockerham 1984) for all pairs of populations with $n > 30$, then evaluated significance of each $F_{ST}$ value with a permutation test (1,000 iterations, FDR-adjusted $\alpha$ from 0.001 to 0.050). We used the program FSTAT (Goudet 1995) to estimate allelic richness for all loci in each population, then calculated mean allelic richness across loci for each population and tested for difference between mean allelic richness of hatchery and wild populations with a Student's *t*-test.

We used the maximum likelihood program CONTML from the PHYLIP version 3.69 software package (Felsenstein 2009; see also Felsenstein 2004:391–414) to infer relationships among all spring Chinook Salmon populations with $n > 30$. We visualized the resulting dendrogram with the program TREEVIEW (Page 1996). To assess node confidence, we bootstrapped the allele frequency data (1,000 resamples) with the program SEQBOOT (Felsenstein 2009), inferred dendrograms as before (for all 1,000 data sets), constructed a consensus tree with the program CONSENSE (Felsenstein 2009), then examined bootstrap values for each node. The resulting bootstrapped tree provides statistical support for a graphical representation of genetic distance relationships among hatchery and wild spring Chinook Salmon populations from the UWR and Catherine Creek Hatchery.



## RESULTS

We collected 1,797 tissue samples from unclipped spring Chinook Salmon from tributaries of the Willamette River. Of these, 1,506 lacked otolith thermal marks and were classified as wild spring Chinook Salmon. Samples included spring Chinook Salmon from multiple age-classes. Although we surveyed the Calapooia River on multiple occasions, no carcasses were encountered nor were any samples collected from this subbasin. We subjected 391 of the wild-origin samples, representing six Willamette River subbasins, to genetic analyses, in addition to 559 hatchery-origin samples. Overall PCR success across all individuals and loci was 94%. We excluded 18 hatchery samples and 119 wild samples from statistical analyses due to insufficient genotypic data. Approximately 80% of the remaining 813 samples provided genotypic data for at least 12 of the 13 loci examined, and all loci were successfully amplified and scored for 564 samples (69% of samples included in statistical analyses). The lower PCR success rate for wild samples, relative to hatchery samples, was likely due to poor tissue quality of some wild samples, which we collected from carcasses in various states of decomposition.

### Heterozygosity

Across all UWR subbasins, the median of observed heterozygosities was significantly higher (Kruskal–Wallis ANOVA on ranks: $H = 6.818$, df = 1, $P = 0.009$) in hatchery populations (median, 81.5%) than in wild populations (median, 75.2%). Similarly, hatchery populations presented higher heterozygosities than did wild populations within subbasin pairs (Friedman rank sum test: $\chi^2 = 5$, df = 1, $P = 0.025$; Table 1). Exact test results indicated that all populations, except the Catherine Creek Hatchery ($P = 0.2303$) and Molalla wild ($P = 0.9103$) populations, were not in HWE ($P < 0.0001$). Subsequent score ($U$) tests revealed that this result was largely driven by lower than expected heterozygosities at two loci: *Omm1080* and *Ots213*. That is, all populations except the Catherine Creek Hatchery population and the small collections of wild fish from the Molalla and Middle Fork Willamette rivers showed significant evidence for heterozygote deficits ($P < 0.0006$) at one or both of these loci. Although no clear pattern was evident for HWE between hatchery and wild populations, the number of locus pairs in LD was consistently higher in hatchery populations than in wild populations.

### Pairwise $F_{ST}$

Among UWR populations upstream from Willamette Falls pairwise $F_{ST}$ values ranged from 0 to 0.009 (Table 2). Populations from above Willamette Falls were more diverged from the Clackamas River hatchery population ($F_{ST} = 0.009$–0.013) than from the Clackamas River wild population ($F_{ST} = 0.001$–0.005). Pairwise $F_{ST}$ values between the Catherine Creek hatchery population and UWR populations were generally an order of magnitude greater than that observed among UWR populations (Table 2). Wild spring Chinook Salmon from the Molalla and Middle Fork Willamette rivers were not included in this analysis due to small sample sizes (Table 1).

We found that $F_{ST}$ values were not significantly different from zero for hatchery and wild population pairs within UWR subbasins above Willamette Falls (North Santiam, $P = 0.047$; South Santiam, $P = 0.535$; McKenzie, $P = 0.317$). With a single exception, both hatchery and wild populations from all UWR subbasins were significantly diverged from hatchery and wild populations from other subbasins. Interestingly, analysis of pairwise $F_{ST}$ values suggested that neither hatchery nor wild spring Chinook Salmon from the South Santiam River were significantly diverged from wild Clackamas River spring Chinook Salmon.

### Allelic Richness

Although per locus allele counts varied considerably among populations, we observed similar levels of allelic richness among populations when sample sizes were normalized by

TABLE 1. Collection location, origin, and number of spring Chinook Salmon samples analyzed from the Willamette River and Catherine Creek Hatchery (Grande Ronde River), 2011. Samples were characterized at 13 microsatellite loci to estimate each population's expected heterozygosity ($H_e$), observed heterozygosity ($H_o$), mean allelic richness (AR; normalized for $n = 22$ per population), and the number of loci not in Hardy–Weinberg equilibrium (HWE) and in linkage disequilibrium (LD).

| Collection location | Origin | Number of samples | $H_e$ | $H_o$ | AR | HWE | LD |
|---|---|---|---|---|---|---|---|
| Catherine Creek | Hatchery | 33 | 0.744 | 0.735 | 11.2 | 0 | 0 |
| Clackamas | Hatchery | 80 | 0.806 | 0.815 | 11.8 | 4 | 38 |
| North Santiam | Hatchery | 95 | 0.819 | 0.820 | 12.2 | 2 | 8 |
| South Santiam | Hatchery | 94 | 0.814 | 0.813 | 12.3 | 2 | 4 |
| McKenzie | Hatchery | 95 | 0.821 | 0.805 | 12.1 | 4 | 15 |
| Middle Fork Willamette | Hatchery | 144 | 0.819 | 0.818 | 12.0 | 3 | 12 |
| Clackamas | Wild | 51 | 0.828 | 0.752 | 13.2 | 4 | 9 |
| Molalla | Wild | 8 | 0.753 | 0.823 | | 0 | 0 |
| North Santiam | Wild | 72 | 0.796 | 0.777 | 11.9 | 4 | 2 |
| South Santiam | Wild | 62 | 0.808 | 0.746 | 12.1 | 4 | 3 |
| McKenzie | Wild | 67 | 0.824 | 0.788 | 12.1 | 3 | 2 |
| Middle Fork Willamette | Wild | 12 | 0.706 | 0.620 | | 0 | 0 |



rarefaction to a minimum 22 diploid individuals. Among Willamette River populations, wild fish from the Clackamas River presented the highest allelic richness, and wild fish from the North Santiam River presented the lowest allelic richness (Table 1). The Catherine Creek Hatchery population presented the lowest allelic richness (11.8) of any population examined. Overall, we found no significant difference for allelic richness between hatchery and wild spring Chinook Salmon from the upper Willamette River ($t = -0.884$, df = 7, $P = 0.406$), though power to reject the null hypothesis for this test was very low ($\beta = 0.05$). Assumptions of normality (Shapiro–Wilk test: $P = 0.119$) and equal variance ($P = 0.595$) for this test were met.

## Genetic Structure among Populations

We inferred a maximum likelihood tree for Willamette River and Catherine Creek Hatchery spring Chinook Salmon from our genotypic data. This dendrogram (Figure 2) suggests that within the Willamette River, spring Chinook Salmon hatchery populations are genetically most similar to local wild populations from the same subbasin. In most cases, these subbasin level hatchery–wild pairings received bootstrap support approaching or exceeding 70%. An exception to this pattern involved the hatchery and wild populations from the South Santiam River and hatchery fish collected from the Middle Fork Willamette River. These putative populations formed a polytomy with insignificant

TABLE 2. Pairwise $F_{ST}$ values ($\theta$; Weir and Cockerham 1984) among hatchery- (H) and wild-origin (W) spring Chinook Salmon populations from the Willamette River and Catherine Creek Hatchery (Grande Ronde River) estimated from genotypic data for 13 microsatellite loci. Values not significantly different from zero (false discovery rate-adjusted $\alpha$ from 0.001 to 0.050) are indicated in bold italic text.

| Source | Clackamas H | Clackamas W | Willamette H | McKenzie H | McKenzie W | North Santiam H | North Santiam W | South Santiam H | South Santiam W |
|---|---|---|---|---|---|---|---|---|---|
| Catherine H | 0.111 | 0.106 | 0.106 | 0.107 | 0.102 | 0.100 | 0.110 | 0.099 | 0.104 |
| Clackamas H | | 0.007 | 0.012 | 0.013 | 0.013 | 0.010 | 0.012 | 0.010 | 0.009 |
| Clackamas W | | | 0.004 | 0.003 | 0.003 | 0.004 | 0.005 | ***0.002*** | ***0.001*** |
| Willamette H | | | | 0.007 | 0.006 | 0.008 | 0.009 | 0.003 | 0.004 |
| McKenzie H | | | | | ***0.000*** | 0.003 | 0.006 | 0.004 | 0.005 |
| McKenzie W | | | | | | 0.004 | 0.006 | 0.004 | 0.003 |
| North Santiam H | | | | | | | ***0.002*** | 0.005 | 0.005 |
| North Santiam W | | | | | | | | 0.005 | 0.005 |
| South Santiam H | | | | | | | | | ***0.000*** |

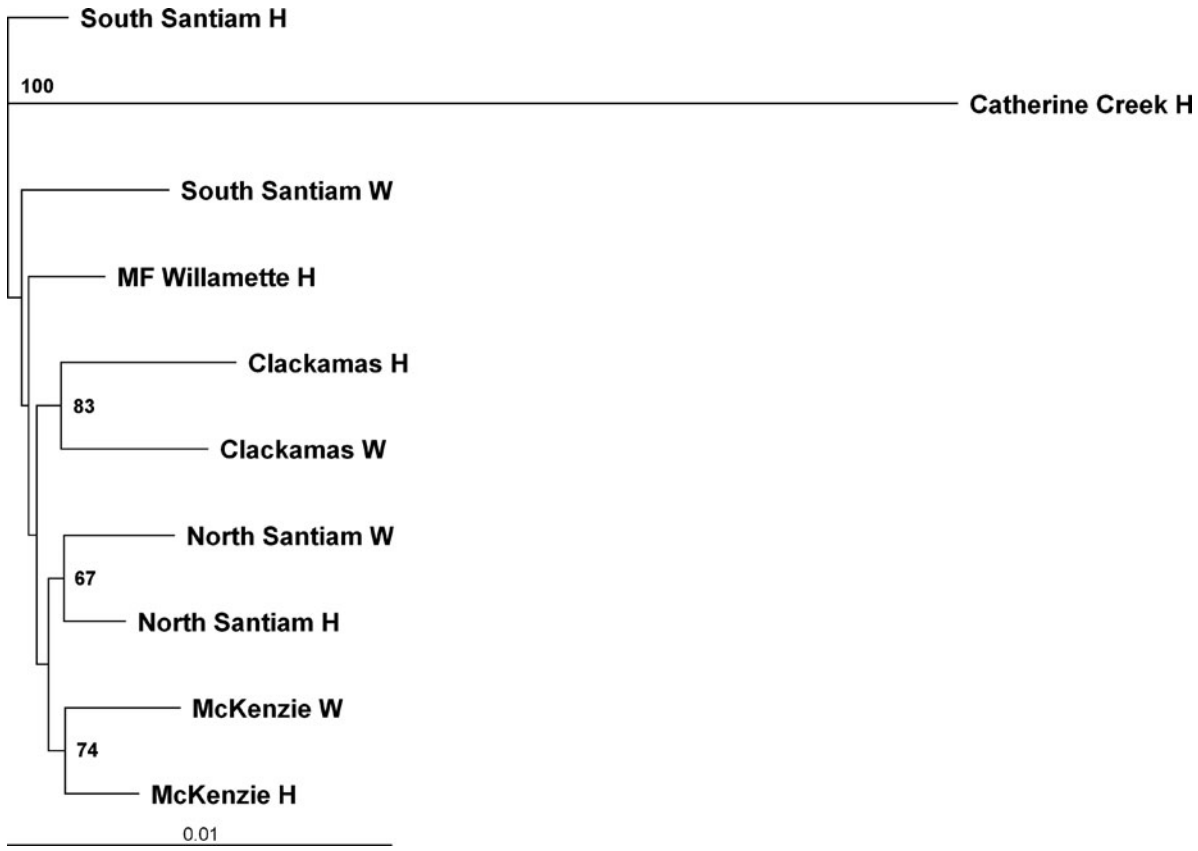


FIGURE 2. Maximum likelihood tree depicting genetic relationships among hatchery- (H) and wild-origin (W) spring Chinook Salmon populations from the upper Willamette River and the Catherine Creek Hatchery population (Grande Ronde River). Dendrogram inferred from genotypic data for 13 microsatellite loci. Branch lengths represent Cavalli-Sforza chord measures of genetic distances (Cavalli-Sforza and Edwards 1967).



internal branch lengths (95% CI) and <50% bootstrap support of branch nodes. Bootstrap support between Catherine Creek and UWR populations was 100%.

## DISCUSSION

### Heterozygosity and Allelic Richness

Genetic diversity is fundamental to the short-term resilience and long-term adaptive potential of salmon populations (Waples et al. 1990; Allendorf 2005). Both heterozygosity and allelic richness are important components of genetic diversity that can be directly compared among populations. We found that mean heterozygosities of Willamette River spring Chinook Salmon populations ranged from 62% to 82%. Our estimates of observed heterozygosity for hatchery populations from the North Santiam and McKenzie rivers (Table 1) differed by less than 1% from those previously reported by Narum et al. (2010), placing these populations among the top 5 of 37 Columbia River spring Chinook Salmon populations described by those authors. We also found that within subbasins of the Willamette River, hatchery populations presented higher heterozygosities than did local wild populations. The large size of UWR spring Chinook Salmon hatchery populations has likely served to maintain heterozygosities above levels found in the typically smaller wild populations, which may experience stronger effects from random genetic drift.

Nearly all UWR spring Chinook Salmon populations presented some evidence for departures from HWE expectations, and there were heterozygote deficits at *Omm1080* or *Ots213* in all but one population. Most populations also presented some evidence of linkage disequilibrium (Table 1), though loci in LD were not consistent across populations. It is not surprising that allele frequencies deviated from HWE expectations at some loci, as our data violated several assumptions required for HWE. Specifically, the assumption of nonoverlapping generations is violated by inclusion of multiple age-classes among samples. Moreover, migration among populations may contribute to departures from HWE. Although HWE departures can also result from genotyping errors, replicate PCR and electrophoretic analyses for a subset of samples suggested allele assignment error rates of <1%. The higher frequencies of LD in hatchery populations may, however, result from monogamous hatchery spawnings that likely produce a greater proportion of full siblings and apparent family structure among adult returns than

found in wild populations, which are commonly characterized by polygamous mating systems (Bentzen et al. 2001).

Although hatchery populations consistently presented higher heterozygosities than wild populations, we found no clear pattern for allelic richness. However, the Clackamas River wild population did present a slightly higher mean allelic richness than other populations, perhaps as a result of admixture generated by inadvertent sampling of a small number of fall or hybridized Chinook Salmon in this subbasin. This hypothesis is consistent with the relatively high number of loci not in HWE for wild Clackamas River spring Chinook Salmon (Table 1), but it cannot be tested with our current data set.

## Genetic Divergence ($F_{ST}$)

In contrast with the findings of Myers et al. (2006), our pairwise $F_{ST}$ estimates suggest that, in the upper Willamette River, hatchery-origin spring Chinook Salmon are genetically most similar to local, wild fish. In most cases, $F_{ST}$ values between local hatchery and wild populations were not significantly different from zero, which reflected no measurable genetic differentiation. By identifying wild-origin fish as only those that lacked adipose fin and otolith marks, our methodology maximized the chance of detecting genetic differences between hatchery and wild fish, should they exist.

The observed similarity between hatchery and wild UWR spring Chinook Salmon is undoubtedly driven by several factors. Although a composite "Willamette stock" of spring Chinook Salmon was used for many years at UWR hatcheries, current hatchery broodstocks were founded either entirely or partially from local, wild spring Chinook Salmon in the mid-1990s or earlier (see Johnson and Friesen 2010 for a review of UWR hatchery broodstock histories). Since that time, ongoing migration between hatchery and local wild populations has continued through natural production by stray hatchery fish and integration of wild fish into hatchery broodstocks. The proportion of hatchery-origin Chinook Salmon (PHOS) on UWR spawning grounds has varied among years and locations. From 2002 to 2010, 4% to 69% of spawners encountered below Dexter Dam on the Middle Fork Willamette River were of hatchery origin (Cannon et al. 2011). During the same period, PHOS ranged from 4% to 73% on the North Santiam River (Cannon et al. 2011). Similarly, the proportion of wild (or natural-origin) fish integrated into hatchery broodstocks (PNOB) has varied among facilities and years (Cannon et al. 2011); from 2002 to 2010, PNOB ranged from 0.3% to 10.1% at the Middle Fork Willamette Hatchery and from 0.3% to 36.2% at Marion Forks Hatchery (North Santiam River). Overall, PHOS tended to exceed PNOB in most UWR subbasins with hatchery facilities. Current marking and annual monitoring programs now provide estimates for these migration parameters, though comparable estimates are not available before the 1997 brood year when otolith thermal marking first began at UWR hatcheries (Johnson and Friesen 2010).

Although most pairwise $F_{ST}$ estimates were statistically significant among populations from different UWR subbasins (Table 2), they were generally lower than values reported for spring Chinook Salmon populations from the Snake ($F_{ST} = 0.017$–$0.045$: Narum and Stephenson 2007), Klamath ($F_{ST} = 0.011$–$0.024$: Kinziger et al. 2008), and California's Central Valley rivers ($F_{ST} = 0.005$–$0.026$: Garza et al. 2008) as characterized with the same genetic markers used in our study. Pairwise $F_{ST}$ values between the Clackamas Hatchery population and other UWR populations were higher than those among most UWR populations, though $F_{ST}$ values between the Clackamas River wild population and populations from the South Santiam River were insignificant. Straying of South Santiam Hatchery spring Chinook Salmon into the Clackamas River wild population does not provide a likely explanation for this result, because, from the near 1.8 million coded-wire-tagged Chinook Salmon released from the South Santiam Hatchery from 1995 to 2010, only 11 of the 5,244 fish recovered as adults were recorded in the Clackamas River (unpublished data from the Regional Mark Information System; http://www.rmpc.org/). Stray rates among wild UWR spring Chinook Salmon populations remain unknown.

## Genetic Structure among Populations

The maximum likelihood tree of Willamette River spring Chinook Salmon further revealed similarities between hatchery and wild populations within subbasins, because local hatchery and wild populations formed branch pairs in all possible cases. Our findings indicate that Willamette River spring Chinook Salmon populations are weakly structured at the subbasin level, and little or no genetic variance is explained by hatchery or wild origin within subbasins. As speculated by Myers et al. (2006), the population structure previously reported within this ESU was likely influenced by the inclusion of closely related individuals among juvenile samples. Unless relatedness is accounted for, data from juvenile samples can easily inflate population divergence estimates and distort genetic relationships (see Allendorf and Phelps 1981). We believe that our results, derived from analyses of adult UWR spring Chinook Salmon, provide an accurate depiction of contemporary UWR spring Chinook Salmon population structure; it is characterized by weak but significant structure among subbasins and no significant divergence between hatchery and wild populations within subbasins.

## Management Implications

The weak but significant genetic structure we observed among populations from different subbasins suggests that conservation and recovery efforts for UWR spring Chinook Salmon should be implemented through subbasin-specific management actions, as identified by ODFW et al. (2011). Current restrictions on stock transfers among UWR spring Chinook Salmon populations should further preserve and possibly strengthen genetic

structure among populations from different subbasins, thereby promoting adaptation to local conditions.

The relatively high heterozygosities of UWR hatchery spring Chinook Salmon and similarities between hatchery and wild populations within subbasins suggest that hatchery-origin spring Chinook Salmon represent genetically appropriate founders for local reintroduction programs. Anderson et al. (2013) concluded that hatchery spring Chinook Salmon can provide demographic benefits to reintroduction programs, and their use for this purpose in the UWR basin has been recommended by NMFS (2008). However, as dam passage for adult and juvenile salmon is improved and determined to be adequate for above-dam population viability, short-term demographic benefits from hatchery fish should be carefully weighed against potential threats that these fish may pose to the evolution, productivity, and long-term viability of recipient populations. Moreover, near-term UWR monitoring and research efforts should aim to identify negative ecological effects that hatchery spring Chinook Salmon might have on wild populations.

Our most relevant finding to conservation and recovery efforts for UWR spring Chinook Salmon may be that, despite rigorous sampling, we identified only 25 naturally produced fish returning to the Middle Fork Willamette River at or below Dexter Dam (of which only 12 were used in the analysis due to poor sample quality). According to reports by Hutchison et al. (1966) and Thompson et al. (1966; also see McElhany et al. 2007), this subbasin once ranked among the most productive UWR tributaries for wild spring Chinook Salmon, before the construction of Lookout Point and Dexter dams (Figure 1) isolated 345 km of high quality spawning and rearing habitat (Cramer et al. 1996). Extensive annual releases (hundreds to thousands) of adult hatchery-origin spring Chinook Salmon above these dams in every year of the last decade (Johnson and Friesen 2010) have consistently failed to result in wild adult returns despite apparently substantial natural production of juveniles (Monzyk et al. 2013; Romer et al. 2013). Keefer et al. (2012) reported high juvenile mortality rates (35–70%) during passage at Middle Fork Willamette dams and Monzyk et al. (2013) documented predation on juvenile Chinook Salmon by resident fishes within the reservoirs. Consistent with the findings of those authors our results suggest that significant improvements to juvenile passage are needed to recover wild spring Chinook Salmon in this UWR subbasin.

Plans for a path toward recovery may be drafted from lessons learned from the Fall Creek tributary of the Middle Fork Willamette River, where a wild population of spring Chinook Salmon, founded by hatchery-origin fish, has begun to expand in apparent response to dam operations that promote juvenile passage (USACE 2013). These results underscore both the potential of hatchery-origin fish for local reintroduction programs and the fundamental role that improved dam passage must play to secure the long-term viability of spring Chinook Salmon in the upper Willamette River.



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