

Testing for Genetic Differences between North and South Umpqua Spring Chinook Salmon

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Research Question

Are spring Chinook salmon in the North and South Umpqua Rivers genetically differentiated?

Objective

Use reduced-representation sequencing to identify thousands of single nucleotide polymorphisms (SNPs) distributed throughout the Chinook salmon genome and test for genetic differences between spring Chinook salmon sampled in the North and South Umpqua Rivers, Oregon.

Methods

In total, 123 Chinook salmon (*Oncorhynchus tshawytscha*) sampled in the North (n = 57) and South (n = 66) Umpqua Rivers, Oregon, in 2020 – 2021 were evaluated in this study. Previously, these individuals were genotyped using Genotyping-in-Thousands by sequencing (GT-seq) (Campbell et al. 2015), to evaluate variation within the chromosome 28 *greb1l* – *rock1* region associated with adult migration timing. To identify and confirm the adult migration timing of the 123 presumed spring Chinook salmon, we compared their genotypes to those of spring and fall returning Chinook salmon in the Rogue River, Oregon. Specifically, Chinook salmon from the North and South Umpqua Rivers were evaluated at 21 markers within the *greb1l* – *rock1* region previously found to be diagnostic for spring and fall adult migration timing in the lower Rogue River (Dayan 2023).

Of the 123 salmon evaluated, we identified three individuals sampled in the North Umpqua that were heterozygous for early- and late-migration alleles (Table A1). These heterozygotes were excluded, and our analyses focused exclusively on homozygous spring

Chinook salmon sampled in the North (NUMP; n = 54) and South (SUMP; n = 66) Umpqua Rivers.

BestRAD libraries and sequencing

Restriction site-associated DNA sequencing (RADseq) libraries were prepared for spring Chinook salmon samples following the BestRAD methods of Ali et al. (2016) with minor modifications, developed by Washington Department of Fish and Wildlife (WDFW) and adapted for salmonid studies. Modifications, including enzyme and reagent volume adjustments, were made to account for the large genome sizes of salmonids.

For each individual, approximately 200ng of DNA was digested in 20uL reactions containing 1X CutSmart® buffer (New England BioLabs) and 8 units· μL^{-1} *SbfI*-HF (New England BioLabs). Biotinylated RAD adapters used to identify samples were then ligated to the cut sites in 24 μL reactions containing 1X T4 ligation buffer, 0.16 μL T4 DNA ligase, and 2ul of 50nM RAD adapter. 11uL aliquots from each of 44 samples (per plate) were then pooled, purified, and concentrated with AMPure XP cleanup beads. 100uL of each of the three plate library preps were transferred and randomly fragmented through sonication with a Biorupter Pico. Fragment length was evaluated on a TapeStation 4150 (Agilent) for sizing.

After fragmentation, a streptavidin bead binding assay was performed to select for fragments that contained the cut site. Biotinylated adaptors containing the cut site were bound to 20uL of streptavidin Dynabeads® M-280 (Life Technologies), and fragments not containing the cut site were washed away. Fragments of interest suspended in 40uL of 1X NEB Buffer 4 were selected for and cleaved from the beads with a *SbfI*-HF incubation, utilizing 40 units· μL^{-1} *SbfI*-HF (New England BioLabs) in a 42 uL reaction. All samples were then purified with an AMPure XP bead clean up and eluted in 55uL Low TE for Illumina library preparation. Final libraries were prepared utilizing the NEBNext Ultra II Library Prep kit (New England BioLabs) with the manufacturer's protocol. Samples from each library were labeled with NEBNext® Multiplex Oligos for Illumina® (New England BioLabs), quantified by quantitative polymerase chain reaction (qPCR) with the NEBNext® Library Quant kit (New England BioLabs) on a QuantStudio 5 (Life Technologies), and pooled at equimolar proportions for sequencing. Paired-end 150bp sequencing was performed on a NovaSeq 6000 at the University of Oregon's Genomics & Cell Characterization Core Facility.

Processing of BestRADseq data

Forward and reverse reads of each sample were demultiplexed with the *process_radtags* command in STACKS v2.52 (Rochette et al. 2019). The *process_radtags* step removed reads with missing RAD cut site, missing barcodes, and those with low quality. This step included the ‘bestrad’ and rescue barcodes ‘-r’ parameters to retain additional reads.

Reference mapping and genotyping

Demultiplexed paired-end reads were mapped to the *O. tshawytscha* v2.0 genome assembly (GCA_018296145.1) using the MEM algorithm in BWA 0.7.17 (Li and Durbin 2009). For each sample, the number and percentage of successfully mapped reads was calculated in SAMtools 1.19.2 (Li et al. 2009).

Loci and genotypes were called for mapped reads using the *ref_map* command and the STACKS genotyping pipeline. Default settings were applied, with variant sites and genotypes called at a significance level of 5% in *gstacks* (- -var_ alpha 0.05 - -gt-alpha 0.05). In the *populations* settings of STACKS, we set the minimum percentage of individuals from the two sample groups required to process a locus (-R) to 0.9, minor allele count (--min-mac) to three, and restricted data analysis to one random single nucleotide polymorphism (SNP) per locus (--write-random-snp). The resulting datafile contained 120 samples (NUMP n = 54; SUMP n = 66) genotyped at 57,579 variants.

Variant and individual quality filtering

We filtered variants and individuals to remove potential artifacts introduced via library preparation, sequencing, and/or bioinformatic processing according to the approach described in O’Leary et al. (2018). We identified putative paralogous sequence variants (PSVs) with paralog-finder 1.0 (Ortiz et al. 2018). Variants out of Hardy-Weinberg Equilibrium (HWE) within either sample group after correcting for multiple comparisons with the false discovery rate were removed using the R (4.3.1) package *pegas* (Paradis 2010). Variants in linkage disequilibrium (LD) were thinned in PLINK 1.9 with a cutoff of 0.8. After applying PSV, HWE, and LD filters, we evaluated the relationship between variant read depth and heterozygote miscall rate in the R package *whoa* (Anderson 2018). Genotype calls with read depth less than three exhibited a probability of miscall rate above 0.1 and were censored (i.e., call replaced with NA). We then iteratively filtered variants and individuals based on genotyping call rates until all variants were typed in $\geq 90\%$ of individuals and all individuals were typed with $\geq 80\%$ of variants.

Genetic differentiation

Genetic differentiation between spring Chinook salmon sampled in the North and South Umpqua Rivers was evaluated with principal components analysis (PCA) and pairwise estimates of the fixation index (F_{ST}). PCA among individual genotypes was conducted in the *adegenet* R package (Jombart 2008), and missing calls were replaced with mean allele frequency prior to ordination. Genetic differentiation was quantified with the Weir and Cockerham (1984) pairwise estimate of F_{ST} in the R package *pegas* (Paradis 2010). Details on sampling locations, dates, and collection type are available as an appendix (Table A2).

Detection of outlier variants

We evaluated variants for potential signatures of directional selection using two programs. Putative outlier loci were evaluated under the default settings of the program OUTFLANK (Whitlock and Lotterhos 2015), and with default settings and a prior of 100 in the program BAYESCAN (Foll and Gaggiotti 2008).

Results

Sequencing

Quality filtering of variants and individuals resulted in 107 spring Chinook salmon in the North (NUMP; $n = 44$) and South (SUMP; $n = 63$) Umpqua Rivers genotyped at 24,839 SNPs. Across all individuals and genotyped variants, mean sequencing depth was 43.03 ± 31.2 (\pm SD).

Genetic differentiation

Overall, we detected little evidence of genetic differentiation between spring Chinook salmon collected in the North and South Umpqua Rivers. Estimates of allelic diversity across variants were similar in both the North ($H_e = 0.151 \pm 0.13$) and South ($H_e = 0.153 \pm 0.13$) Umpqua sample groups. Individuals collected from the two rivers largely overlapped in PCA (Fig. 1), and PC1 and PC2 explained little of the total variation among genotypes (2.3% and 2.1%, respectively). We evaluated the extent to which individuals clustered based on collection type, sex, collection location within each of the two rivers, and collection year (Fig. 2), but none of these factors were strongly associated with genetic composition.

The average F_{ST} across all 24,839 variants was 0.008 ± 0.02 with a range of -0.016 to 0.207, indicating genetic differentiation between Chinook salmon collected from the North and South Umpqua Rivers was not significantly different from zero (Fig. 3). Furthermore, moderate estimates of F_{ST} were broadly distributed across the genome (Fig. 4), with no single

chromosomal region displaying skew in allele frequencies. Correspondingly, neither OUTFLANK nor BAYESCAN detected any outlier loci indicative of directional selection under their default settings.

Summary

- Overall, we found no evidence of significant genetic differentiation between spring Chinook salmon sampled in the North Umpqua ($n = 44$) and South Umpqua Rivers ($n = 63$) in 2020 – 2021 based on variation at 24,839 SNPs.
- Moreover, we did not detect a significant signal of directional selection in the two sample groups within any chromosomal region of the *O. tshawytscha* genome.
- Reduced representation sequencing approaches have been shown to be effective at identifying putatively adaptive differences between populations even amid low neutral genetic differentiation (e.g., Vaux et al. 2021) and detecting loci of major effect (e.g., Prince et al. 2017). Nonetheless, it is possible that whole genome sequencing may reveal genetic differences not detected with the methods implemented in this study.

Figures

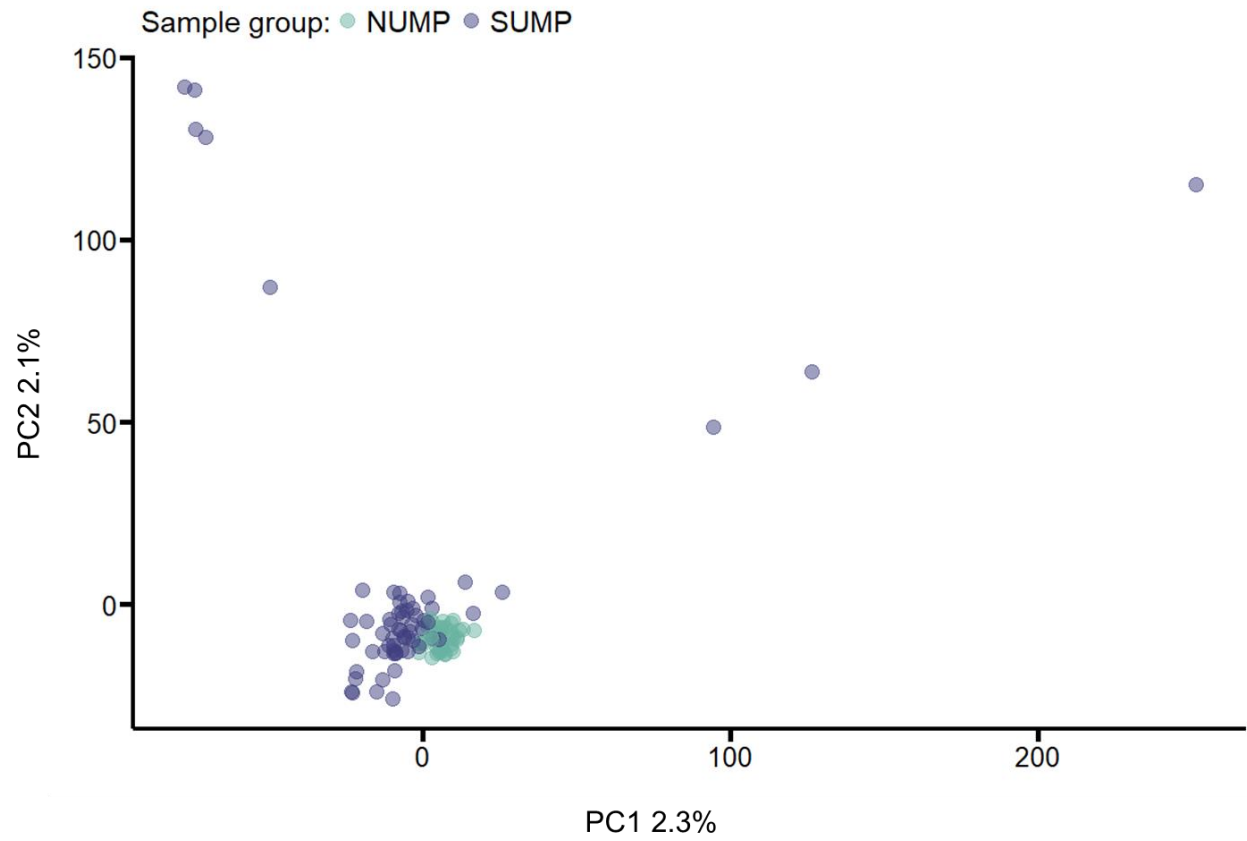


Figure 1. Principal components analysis (PCA) of *O. tshawytscha* genotypes (n = 107 individuals genotyped at 24,839 variants). Color of data points indicates sample group.

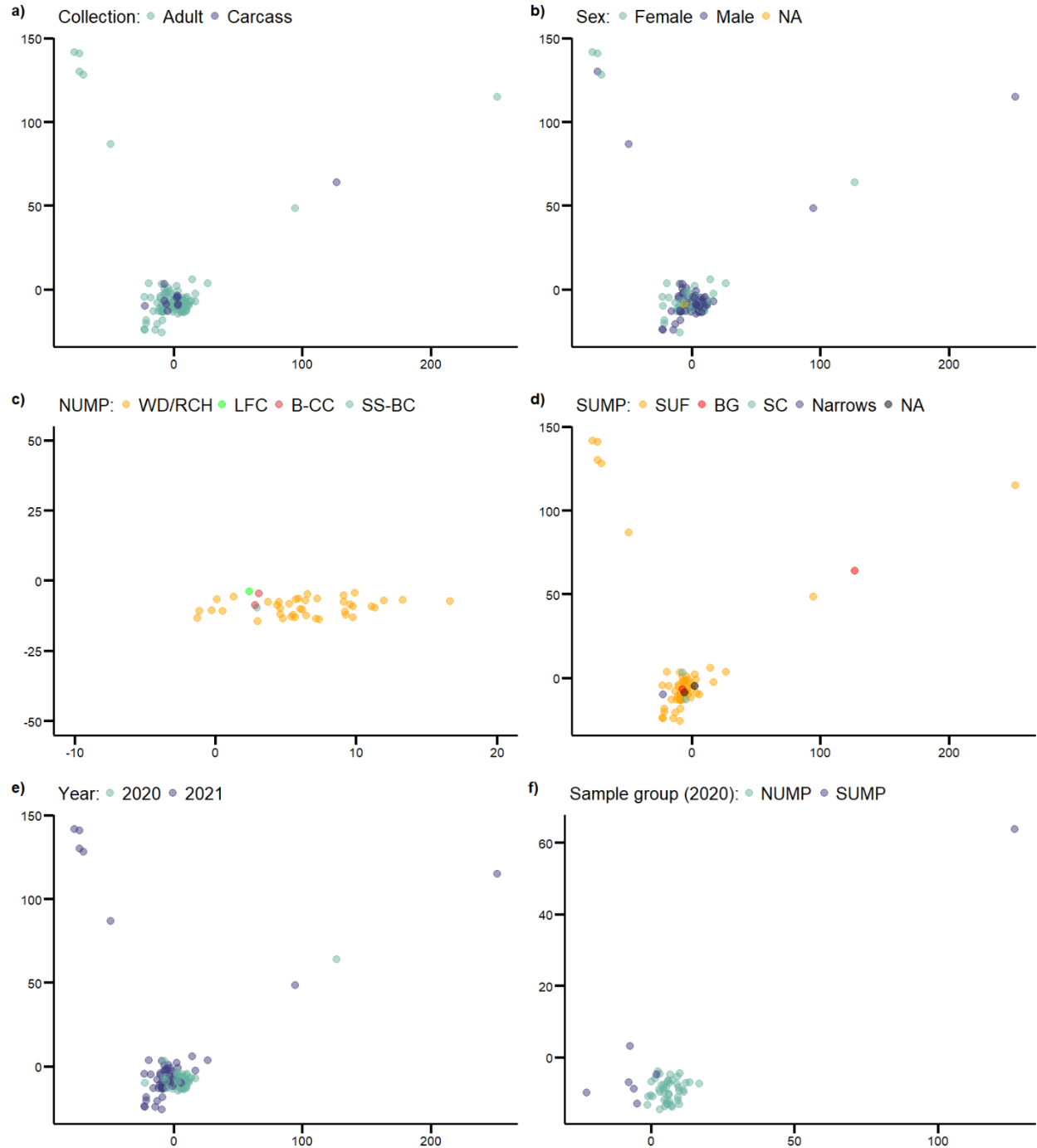


Figure 2. Principal components analyses (PCAs) of *O. tshawytscha* genotypes (n = 107 individuals genotyped at 24,839 variants). Color of data points indicates: a) collection type, b) sex, c) collection location within North Umpqua (WD/RCH: Winchester Dam/Rock Creek Hatchery, LFC: Lower Fish Creek, B-CC: Boulder to Copeland Creek, SS-BC: Soda Springs to Boulder Creek), d) collection location within South Umpqua (SUF: South Umpqua Falls, BG: Bedrock Gorge, SC: Skillet Creek, Narrows, NA: Not available), e) year sampled, and f) sample group collected in 2020.

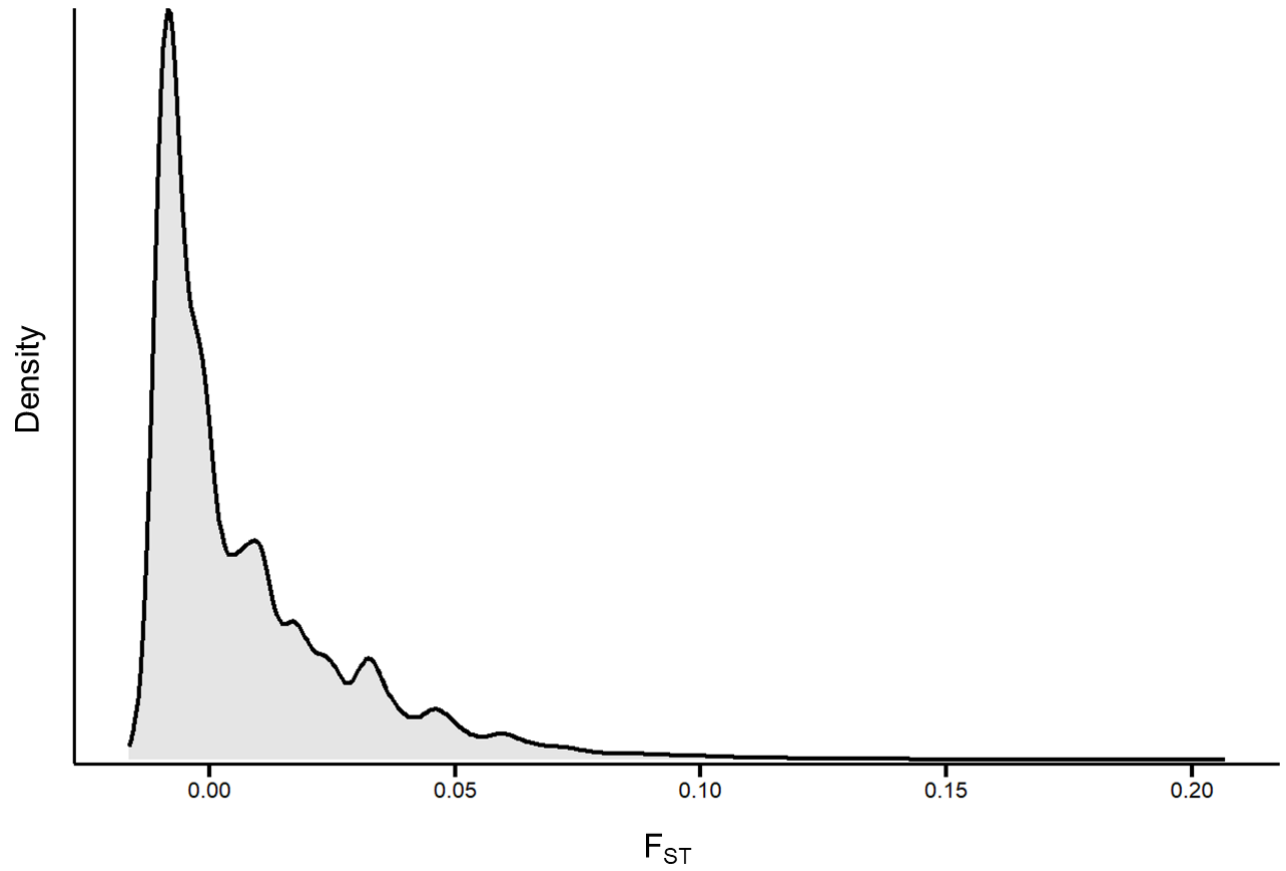


Figure 3. Density of F_{ST} estimates quantifying genetic differentiation between *O. tshawytscha* in the North and South Umpqua Rivers ($n = 107$ individuals genotyped at 24,839 variants).

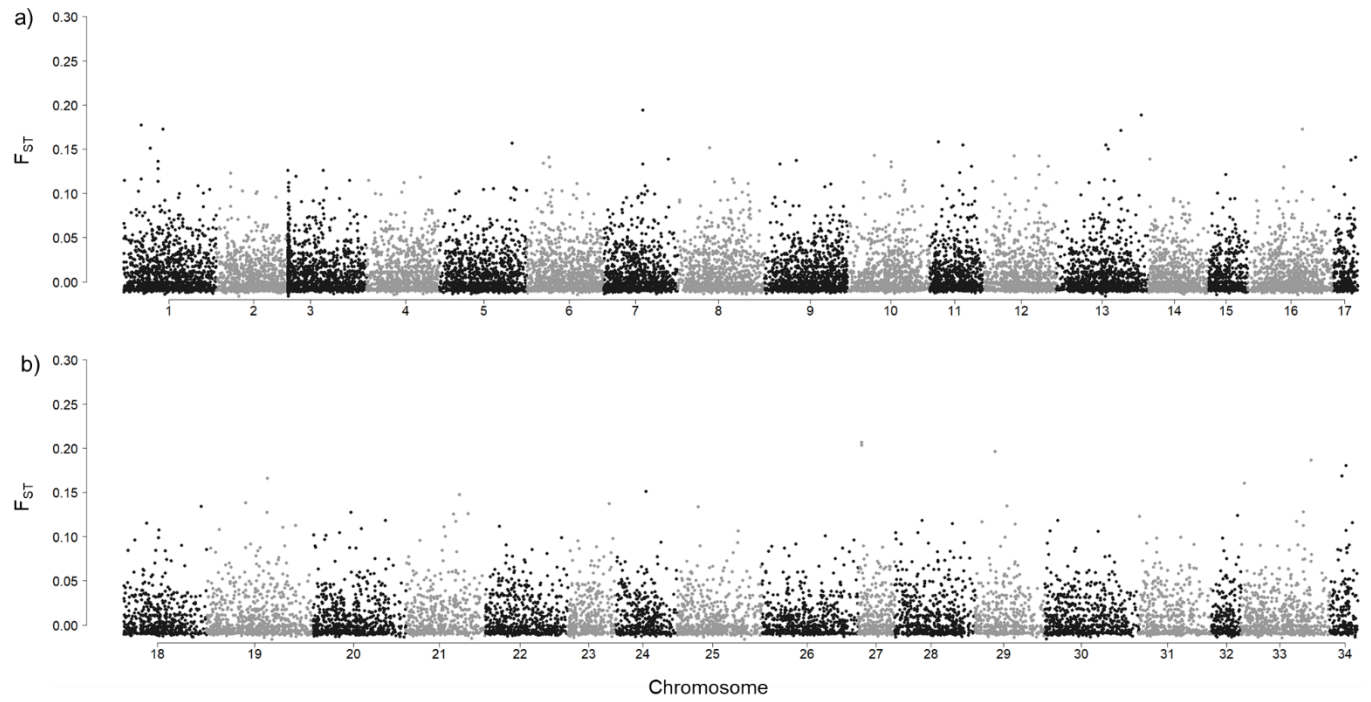


Figure 4. Manhattan plot depicting F_{ST} estimates and genomic positions of the 24,839 variants called in North and South Umpqua Chinook salmon. a) Variants positioned within chromosomes 1 – 17, and b) variants positioned within chromosomes 18 – 34.

References

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Appendix

Table A1. Adult migration timing genotypes of the 123 *O. tshawytscha* evaluated in this study.

Three samples with heterozygous early- and late- migration alleles were excluded (highlighted in yellow). The remaining 120 samples harbored predominantly homozygous early- migration alleles (i.e., spring Chinook salmon) at the 21 adult migration timing markers and were incorporated in analyses. Thirteen additional samples that were included in the analysis were heterozygous at one to three of the adult migration timing markers located on one end or the other (i.e., the *greb11* end or the *rock1* end). Missing genotype indicated by 0.

sample	Ots28_11023212	Ots28_11025336	Ots28_11033282	Ots28_11070757	Ots28_11071377	Ots28_11072994	Ots28_11073102	Ots37124-12277401	Ots28_11073668	Ots28_11075348	Ots28_11075712	Ots28_11077016	Ots28_11077172	Ots37124-12281207	Ots28_11077576	Ots28_11095755	Ots37124-12310649	Ots28_11143508	Ots28_11160599	Ots28_11164637	Ots28_11201129
OtsAC20NU MP_0001	A A	A A	G G	A A	T T	C C	T T	T T	T T	G G	C C	C C	G G	T T	A A	T T	A A	A A	T T	A A	G G
OtsAC20NU MP_0002	A A	A A	G G	A A	T T	C C	T T	T T	T T	G G	C C	C C	G G	T T	A A	T T	A A	A A	T T	A A	G G
OtsAC20NU MP_0003	A A	A A	G G	A A	T T	C C	T T	T T	T T	G G	C C	C C	G G	T T	A A	T T	A A	A A	T T	A A	G G
OtsAC20NU MP_0004	A A	A A	G G	A A	T T	C C	T T	T T	T T	G G	C C	C C	G G	T T	A A	T T	A A	A A	T T	A A	G G
OtsAC20NU MP_0005	A A	0 G	G G	A A	T T	C C	T T	T T	T T	G G	C C	C C	G G	T T	A A	T T	A A	A A	T T	A A	G G
OtsAC20NU MP_0006	A A	A A	G G	A A	T T	C C	T T	T T	T T	G G	C C	C C	G G	T T	A A	T T	A A	A A	T T	A A	G G
OtsAC20NU MP_0007	A A	A A	G G	A A	T T	C C	T T	T T	T T	G G	C C	C C	G G	T T	A A	T T	A A	A A	T T	A A	G G
OtsAC20NU MP_0008	A A	A A	G G	A A	T T	C C	T T	T T	T T	G G	C C	C C	G G	T T	A A	T T	A A	A A	T T	A A	G G
OtsAC20NU MP_0010	A A	A A	G G	A A	T T	C C	T T	T T	T T	G G	C C	C C	G G	T T	A A	T T	A A	A A	T T	A A	G G
OtsAC20NU MP_0011	A A	A A	G G	A A	T T	C C	T T	T T	T T	G G	C C	C C	G G	T T	A A	T T	A A	A A	T T	A A	G G
OtsAC20NU MP_0012	A G	A C	G A	A A	T T	C C	T T	T T	T T	G G	C C	C C	G G	T T	A A	T T	A A	A A	T T	A A	G G
OtsAC20NU MP_0013	A A	A A	G A	A A	T T	C C	T T	T T	T T	G G	C C	C C	G G	T T	A A	T T	A A	A A	T T	A A	G G
OtsAC20NU MP_0014	A A	A A	G G	A A	T T	C C	T T	T T	T T	G G	C C	C C	G G	T T	A A	T T	A A	A A	T T	A A	G G

OtsAC21SU MP_0012	A A	0 	G G	A A	T T	C C	T T	T T	T T	G G	C C	C C	G G	T T	A A	0 	A A	0 	0 	A A	G G
OtsAC21SU MP_0013	A A	A A	G G	A A	T T	C C	T T	T T	T T	G G	C C	C C	G G	T T	A A	T T	A A	0 	0 	A A	G G
OtsAC21SU MP_0014	A A	0 	G G	A A	T T	C C	T T	T T	T T	G G	C C	C C	G G	T T	A A	0 	A A	0 	0 	A A	G G
OtsAC21SU MP_0015	A A	A A	G G	A A	T T	C C	T T	T T	T T	G G	C C	C C	G G	T T	A A	T T	A A	A A	T T	A A	G G
OtsAC21SU MP_0016	A A	A A	G G	A A	T T	C C	T T	T T	T T	G G	C C	C C	G G	T T	A A	T T	A A	0 	T T	A A	G G
OtsAC21SU MP_0017	A A	A A	G G	A A	T T	C C	T T	T T	T T	G G	C C	C C	G G	T T	A A	T T	A A	A A	T T	A A	G G
OtsAC21SU MP_0018	A A	A A	G G	A A	T T	C C	T T	T T	T T	G G	C C	C C	G G	T T	A A	T T	A A	A A	T T	A A	G G
OtsAC21SU MP_0019	A A	0 	G G	A A	T T	C C	T T	T T	T T	G G	C C	C C	G G	T T	A A	0 	A A	0 	T T	A A	G G
OtsAC21SU MP_0020	A A	A A	G G	A A	T T	C C	T T	T T	T T	G G	C C	C C	G G	T T	A A	T T	A A	A A	T T	A A	G G
OtsAC21SU MP_0021	A A	A A	G G	A A	T T	C C	T T	T T	T T	G G	C C	C C	G G	T T	A A	T T	A A	0 	0 	A A	G G
OtsAC21SU MP_0022	A A	A A	G G	A A	T T	C C	T T	T T	T T	G G	C C	C C	G G	T T	A A	T T	A A	0 	T T	A A	G G
OtsAC21SU MP_0023	A A	A A	G G	A A	T T	C C	T T	T T	T T	G G	C C	C C	G G	T T	A A	T T	A A	A A	T T	A A	G G
OtsAC21SU MP_0024	A A	A A	G A	A A	T T	C C	T T	T T	T T	G G	C C	C C	G G	T T	A A	T T	A A	A A	T T	A A	G G
OtsAC21SU MP_0025	A A	A A	G G	A A	T T	C C	T T	T T	T T	G G	C C	C C	G G	T T	A A	T T	A A	A A	T T	A A	G G
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OtsAC21SU MP_0028	A A	A A	G G	A A	T T	C C	T T	T T	T T	G G	C C	C C	G G	T T	A A	T T	A A	A A	T T	A A	G G
OtsAC21SU MP_0029	A A	A A	G G	A A	T T	C C	T T	T T	T T	G G	C C	C C	G G	T T	A A	T T	A A	A A	T T	A A	G G
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OtsAC21SU MP_0034	A A	A A	G G	A A	T T	C C	T T	T T	T T	G G	C C	C C	G G	T T	A A	T T	A A	0 	T T	A A	G G

OtsAC21SU MP_0058	A	A	G	A	T	C	T	T	T	G	C	C	G	T	A	T	A	0	T	A	G
	A	A	A	A	T	C	T	T	T	G	C	C	G	T	A	T	A		T	A	G
OtsAC21SU MP_0059	A	A	G	A	T	C	T	T	T	G	C	C	G	T	A	T	A	0	T	A	G
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OtsCC20NU MP_0012	A	A	G	A	T	C	T	T	T	G	C	C	G	T	A	T	A	A	T	A	G
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	A	A	G	A	T	C	T	T	T	G	C	C	G	T	A	T	A	A	T	A	G
OtsCC20NU MP_0016	A	A	G	A	T	C	T	T	T	G	C	C	G	T	A	T	A	0	T	A	G
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	A	A	G	G	C	T	A	A	A	A	T	T	A	T	G	T	A	A	T	A	G
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	A	A	G	A	T	C	T	T	T	G	C	C	G	T	A	T	A	A	T	A	G
OtsCC20NU MP_0037	A	0	G	A	T	C	T	T	T	G	C	C	G	T	A	0	A	0	T	A	G
	A		A	A	T	C	T	T	T	G	C	C	G	T	A		A		T	A	G
OtsCC20SU MP_0001	A	A	G	A	T	C	T	T	T	G	C	C	G	T	A	T	A	A	T	A	G
	A	A	A	A	T	C	T	T	T	G	C	C	G	T	A	T	A	A	T	A	G
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	A	A	G	A	T	C	T	T	T	G	C	C	G	T	A	T	A	A	T	A	G
OtsCC20SU MP_0004	A	0	G	A	0	C	T	T	T	G	C	C	0	T	A	0	A	A	T	A	G
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OtsCC20SU MP_0005	A	A	G	A	T	C	T	T	T	G	C	C	G	T	A	T	A	A	T	A	G
	A	A	G	A	T	C	T	T	T	G	C	C	G	T	A	T	A	A	T	A	G
OtsCC20SU MP_0006	A	A	G	A	T	C	T	T	T	G	C	C	G	T	A	T	A	A	T	A	G
	A	A	G	A	T	C	T	T	T	G	C	C	G	T	A	T	A	A	T	A	G
OtsCC20SU MP_0007	A	A	G	A	T	C	T	T	T	G	C	C	G	T	A	T	A	A	T	A	G
	G	C	A	A	T	C	T	T	T	G	C	C	G	T	A	T	A	A	T	A	G
OtsCC20SU MP_0008	A	A	G	A	T	C	T	T	T	G	C	C	G	T	A	T	A	A	T	A	G
	A	A	G	A	T	C	T	T	T	G	C	C	G	T	A	T	A	A	T	A	G
OtsCC20SU MP_0010	A	A	G	A	T	C	T	T	T	G	C	C	G	T	A	T	A	A	T	A	G
	A	A	G	A	T	C	T	T	T	G	C	C	G	T	A	T	A	A	T	A	G

Table A2. Sample name, date, river, location, sex, and quality control filtering designation. Location abbreviations in North Umpqua River (WD/RCH: Winchester Dam/Rock Creek Hatchery, B-CC: Boulder to Copeland Creek, SS-BC: Soda Springs to Boulder Creek). Location abbreviations in South Umpqua River (SUF: South Umpqua Falls, NA: Not available).

Sample	Date	River	Location	Sex	Passed Filtering
OtsAC20NUMP_0001	NA	North Umpqua	WD/RCH	F	TRUE
OtsAC20NUMP_0002	NA	North Umpqua	WD/RCH	M	TRUE
OtsAC20NUMP_0003	NA	North Umpqua	WD/RCH	M	TRUE
OtsAC20NUMP_0004	NA	North Umpqua	WD/RCH	M	TRUE
OtsAC20NUMP_0005	NA	North Umpqua	WD/RCH	M	TRUE
OtsAC20NUMP_0006	NA	North Umpqua	WD/RCH	M	TRUE
OtsAC20NUMP_0007	NA	North Umpqua	WD/RCH	M	TRUE
OtsAC20NUMP_0008	NA	North Umpqua	WD/RCH	M	TRUE
OtsAC20NUMP_0010	NA	North Umpqua	WD/RCH	M	TRUE
OtsAC20NUMP_0011	NA	North Umpqua	WD/RCH	M	TRUE
OtsAC20NUMP_0012	NA	North Umpqua	WD/RCH	M	TRUE
OtsAC20NUMP_0013	NA	North Umpqua	WD/RCH	M	TRUE
OtsAC20NUMP_0014	NA	North Umpqua	WD/RCH	M	TRUE
OtsAC20NUMP_0015	NA	North Umpqua	WD/RCH	M	TRUE
OtsAC20NUMP_0016	NA	North Umpqua	WD/RCH	M	TRUE
OtsAC20NUMP_0017	NA	North Umpqua	WD/RCH	M	FALSE
OtsAC20NUMP_0019	NA	North Umpqua	WD/RCH	F	TRUE
OtsAC20NUMP_0020	NA	North Umpqua	WD/RCH	M	TRUE
OtsAC20NUMP_0021	NA	North Umpqua	WD/RCH	F	FALSE
OtsAC20NUMP_0022	NA	North Umpqua	WD/RCH	F	TRUE
OtsAC20NUMP_0023	NA	North Umpqua	WD/RCH	M	TRUE
OtsAC20NUMP_0024	NA	North Umpqua	WD/RCH	M	TRUE
OtsAC20NUMP_0025	NA	North Umpqua	WD/RCH	M	TRUE

OtsAC20NUMP_0026	NA	North Umpqua	WD/RCH	M	TRUE
OtsAC20NUMP_0027	NA	North Umpqua	WD/RCH	M	TRUE
OtsAC20NUMP_0028	NA	North Umpqua	WD/RCH	M	TRUE
OtsAC20NUMP_0029	NA	North Umpqua	WD/RCH	M	TRUE
OtsAC20NUMP_0030	NA	North Umpqua	WD/RCH	F	TRUE
OtsAC20NUMP_0031	NA	North Umpqua	WD/RCH	M	TRUE
OtsAC20NUMP_0032	NA	North Umpqua	WD/RCH	M	FALSE
OtsAC20NUMP_0034	NA	North Umpqua	WD/RCH	M	TRUE
OtsAC20NUMP_0035	NA	North Umpqua	WD/RCH	M	TRUE
OtsAC20NUMP_0036	NA	North Umpqua	WD/RCH	M	TRUE
OtsAC20NUMP_0037	NA	North Umpqua	WD/RCH	M	TRUE
OtsAC20NUMP_0038	NA	North Umpqua	WD/RCH	M	TRUE
OtsAC20NUMP_0039	NA	North Umpqua	WD/RCH	M	TRUE
OtsAC20NUMP_0040	NA	North Umpqua	WD/RCH	F	FALSE
OtsAC20NUMP_0041	NA	North Umpqua	WD/RCH	F	FALSE
OtsAC20NUMP_0042	NA	North Umpqua	WD/RCH	F	TRUE
OtsAC20NUMP_0043	NA	North Umpqua	WD/RCH	F	FALSE
OtsAC20NUMP_0044	NA	North Umpqua	WD/RCH	F	TRUE
OtsAC20NUMP_0045	NA	North Umpqua	WD/RCH	M	TRUE
OtsAC20NUMP_0046	NA	North Umpqua	WD/RCH	M	TRUE
OtsAC20NUMP_0047	NA	North Umpqua	WD/RCH	M	TRUE
OtsAC20NUMP_0048	NA	North Umpqua	WD/RCH	F	FALSE
OtsAC20NUMP_0049	NA	North Umpqua	WD/RCH	F	TRUE
OtsAC20NUMP_0050	NA	North Umpqua	WD/RCH	F	FALSE
OtsAC20NUMP_0051	NA	North Umpqua	WD/RCH	F	TRUE
OtsAC20NUMP_0053	NA	North Umpqua	WD/RCH	F	FALSE
OtsAC21SUMP_0001	5/5/2021	South Umpqua	SUF	F	TRUE
OtsAC21SUMP_0002	5/5/2021	South Umpqua	SUF	M	TRUE
OtsAC21SUMP_0003	5/6/2021	South Umpqua	SUF	F	TRUE
OtsAC21SUMP_0005	5/11/2021	South Umpqua	SUF	M	TRUE

OtsAC21SUMP_0006	5/11/2021	South Umpqua	SUF	M	TRUE
OtsAC21SUMP_0007	5/12/2021	South Umpqua	SUF	F	TRUE
OtsAC21SUMP_0008	5/13/2021	South Umpqua	SUF	F	TRUE
OtsAC21SUMP_0009	5/13/2021	South Umpqua	SUF	M	TRUE
OtsAC21SUMP_0010	5/13/2021	South Umpqua	SUF	F	TRUE
OtsAC21SUMP_0011	5/13/2021	South Umpqua	SUF	M	TRUE
OtsAC21SUMP_0012	5/13/2021	South Umpqua	SUF	M	TRUE
OtsAC21SUMP_0013	5/13/2021	South Umpqua	SUF	M	TRUE
OtsAC21SUMP_0014	5/14/2021	South Umpqua	SUF	F	TRUE
OtsAC21SUMP_0015	5/14/2021	South Umpqua	SUF	F	TRUE
OtsAC21SUMP_0016	5/14/2021	South Umpqua	SUF	M	TRUE
OtsAC21SUMP_0017	5/14/2021	South Umpqua	SUF	F	TRUE
OtsAC21SUMP_0018	5/14/2021	South Umpqua	SUF	F	TRUE
OtsAC21SUMP_0019	5/14/2021	South Umpqua	SUF	M	TRUE
OtsAC21SUMP_0020	5/18/2021	South Umpqua	SUF	F	TRUE
OtsAC21SUMP_0021	5/18/2021	South Umpqua	SUF	F	TRUE
OtsAC21SUMP_0022	5/18/2021	South Umpqua	SUF	M	TRUE
OtsAC21SUMP_0023	5/18/2021	South Umpqua	SUF	F	TRUE
OtsAC21SUMP_0024	5/18/2021	South Umpqua	SUF	M	TRUE
OtsAC21SUMP_0025	5/18/2021	South Umpqua	SUF	M	TRUE
OtsAC21SUMP_0026	5/19/2021	South Umpqua	SUF	M	TRUE
OtsAC21SUMP_0027	5/19/2021	South Umpqua	SUF	M	TRUE
OtsAC21SUMP_0028	5/25/2021	South Umpqua	SUF	F	FALSE
OtsAC21SUMP_0029	5/25/2021	South Umpqua	SUF	M	TRUE
OtsAC21SUMP_0030	5/25/2021	South Umpqua	SUF	M	TRUE
OtsAC21SUMP_0031	5/26/2021	South Umpqua	SUF	F	TRUE
OtsAC21SUMP_0032	5/26/2021	South Umpqua	SUF	M	TRUE
OtsAC21SUMP_0033	5/26/2021	South Umpqua	SUF	F	TRUE
OtsAC21SUMP_0034	5/26/2021	South Umpqua	SUF	M	TRUE
OtsAC21SUMP_0035	5/27/2021	South Umpqua	SUF	F	TRUE

OtsAC21SUMP_0036	5/27/2021	South Umpqua	SUF	F	TRUE
OtsAC21SUMP_0037	5/27/2021	South Umpqua	SUF	F	TRUE
OtsAC21SUMP_0038	6/1/2021	South Umpqua	SUF	F	TRUE
OtsAC21SUMP_0039	6/1/2021	South Umpqua	SUF	F	TRUE
OtsAC21SUMP_0040	6/1/2021	South Umpqua	SUF	F	TRUE
OtsAC21SUMP_0041	6/1/2021	South Umpqua	SUF	M	TRUE
OtsAC21SUMP_0042	6/1/2021	South Umpqua	SUF	M	TRUE
OtsAC21SUMP_0043	6/1/2021	South Umpqua	SUF	M	TRUE
OtsAC21SUMP_0044	6/1/2021	South Umpqua	SUF	M	TRUE
OtsAC21SUMP_0045	6/1/2021	South Umpqua	SUF	F	TRUE
OtsAC21SUMP_0046	6/1/2021	South Umpqua	SUF	M	TRUE
OtsAC21SUMP_0047	6/1/2021	South Umpqua	SUF	F	TRUE
OtsAC21SUMP_0048	6/1/2021	South Umpqua	SUF	F	TRUE
OtsAC21SUMP_0049	6/1/2021	South Umpqua	SUF	F	TRUE
OtsAC21SUMP_0050	6/2/2021	South Umpqua	SUF	M	TRUE
OtsAC21SUMP_0051	6/2/2021	South Umpqua	SUF	F	TRUE
OtsAC21SUMP_0052	6/2/2021	South Umpqua	SUF	F	TRUE
OtsAC21SUMP_0053	6/2/2021	South Umpqua	SUF	M	TRUE
OtsAC21SUMP_0054	6/2/2021	South Umpqua	SUF	M	TRUE
OtsAC21SUMP_0055	6/2/2021	South Umpqua	SUF	M	TRUE
OtsAC21SUMP_0056	6/2/2021	South Umpqua	SUF	M	TRUE
OtsAC21SUMP_0057	6/2/2021	South Umpqua	SUF	F	TRUE
OtsAC21SUMP_0058	6/2/2021	South Umpqua	SUF	M	FALSE
OtsAC21SUMP_0059	5/28/2021	South Umpqua	SUF	M	TRUE
OtsCC20NUMP_0004	10/6/2020	North Umpqua	Lower Fish Cr	M	TRUE
OtsCC20NUMP_0007	10/1/2020	North Umpqua	Soda Springs	F	FALSE
OtsCC20NUMP_0012	10/1/2020	North Umpqua	B-CC	F	TRUE
OtsCC20NUMP_0014	10/1/2020	North Umpqua	SS-BC	M	TRUE
OtsCC20NUMP_0016	10/1/2020	North Umpqua	B-CC	M	TRUE
OtsCC20NUMP_0020	10/6/2020	North Umpqua	Lower Fish Cr	F	FALSE

OtsCC20NUMP_0027	10/7/2020	North Umpqua	Full Flow Reach	F	FALSE
OtsCC20NUMP_0037	10/8/2020	North Umpqua	SS-BC	M	FALSE
OtsCC20SUMP_0001	8/19/2020	South Umpqua	NA	NA	TRUE
OtsCC20SUMP_0002	10/19/2020	South Umpqua	Bedrock Gorge	F	TRUE
OtsCC20SUMP_0004	10/13/2020	South Umpqua	FS-2838Bridge	M	FALSE
OtsCC20SUMP_0005	10/19/2020	South Umpqua	Skillet Cr	M	TRUE
OtsCC20SUMP_0006	10/19/2020	South Umpqua	Skillet Cr	M	TRUE
OtsCC20SUMP_0007	10/21/2020	South Umpqua	Narrows	F	TRUE
OtsCC20SUMP_0008	10/9/2020	South Umpqua	Bedrock Gorge	F	TRUE
OtsCC20SUMP_0010	10/7/2020	South Umpqua	NA	M	TRUE