

# CHINOOK AND STEELHEAD GENOTYPING FOR GENETIC STOCK IDENTIFICATION AT LOWER GRANITE DAM

ANNUAL PROGRESS REPORT July 1, 2010 — June 30, 2011



Prepared by:

Mike Ackerman Fisheries Research Biologist

and

Matt Campbell Genetics Laboratory Manager

IDFG Report Number 11-113 June 2011

FISHERY

# CHINOOK AND STEELHEAD GENOTYPING FOR GENETICS STOCK IDENTIFICATION AT LOWER GRANITE DAM

# **Project Progress Report**

2010 Annual Report

By

Mike Ackerman Jesse McCane Craig Steele Matt Campbell

Idaho Department of Fish and Game 600 South Walnut Street P.O. Box 25 Boise, ID 83707

and

Andrew Matala Jon Hess Shawn Narum

Columbia River Inter-Tribal Fish Commission Hagerman Fish Culture Experiment Station 3059-F National Fish Hatchery Road Hagerman, ID 83332

То

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

Project Number 2010-026-00 Contract Number 48347

IDFG Report Number 11-113 June 2011

# TABLE OF CONTENTS

# <u>Page</u>

ABSTRACT	.1
INTRODUCTION	.2
OBJECTIVES	.3
Objective 1. Discover and Evaluate SNP Marker Sets	.3
Objective 2. Construct SNP Genetic Baselines	.4
Objective 3. Use GSI Methods to Estimate Stock Compositions at Lower Granite Dam	.4
REPORT STRUCTURE	.4
SECTION 1: DISCOVER AND EVALUATE SNP MARKER SETS	5
Introduction	.5
Methods	.5
Discussion	.ə 5
SECTION 2: SNP BASELINES	.6
	6
Methods	.7
Sample Collection	7
Laboratory Protocol	.7
Statistical Analyses	8. 0
Discussion	1
SECTION 3: USE GSI METHODS TO ESTIMATE STOCK COMPOSITIONS AT	-
LOWER GRANITE DAM	2
Introduction1	2
Methods1	2
Sampling at Lower Granite Dam1	2
Laboratory Protocol1 Statistical Apalysos	3
Results	4
Discussion1	5
CONCLUSION1	7
ACKNOWLEDGEMENTS1	8
LITERATURE CITED1	9

#### LIST OF TABLES

<u>Page</u>

Table 1. Oncorhynchus mykiss populations screened for 192 assays for Snake River baseline v1.0. Genotyping agency, major population group (MPG). sample size (n), latitude, longitude, years sampled, expected ( $H_E$ ) and observed (H<sub>o</sub>) heterozygosity, population-specific F<sub>ST</sub>, and number of loci with HWE deviations are shown. Map # corresponds to numbers in FIGURE 1. Agency indicates the laboratory where samples were genotyped. All populations are summer-run, of natural origin, and presumed to be of anadromous lineage......25 Table 2. Oncorhynchus tshawytscha populations screened for 96 assays for Snake River baseline v1.0. Genotyping agency, major population group (MPG), sample size (n), latitude, longitude, years sampled, expected ( $H_E$ ) and observed (H<sub>0</sub>) heterozygosity, population-specific F<sub>ST</sub>, and number of loci with HWE deviations are shown. Map # corresponds to numbers in FIGURE 2. Agency indicates the laboratory where samples were Summary of 188 SNP markers (Appendix A and Hess et al. 2011b) Table 3. genotyped across 49 steelhead populations from throughout the Snake River. SNPs designated as PBT are used for both the PBT project (BPA Project #2010-031-00) and GSI projects. SNPs designated as GSI are used primarily for GSI projects. Summary statistics include minor-allele frequency (MAF) range, expected (H<sub>E</sub>) and observed (H<sub>O</sub>) heterozygosity. Weir and Cockerham (1984)  $F_{ST}$ , and fixation index ( $F_{IS}$ ). "HWE" designates the number of populations that a SNP deviated from Hardy-Weinberg expectation for any SNP that deviated in greater than 10% of populations. "LD" is the number of populations that a pair of SNP loci exhibited linkage disequilibrium if it occurred in more than half of the populations. "CS" indicates a locus that was designated as a candidate Table 4. Summary of 95 SNP markers (Appendix B and Matala et al. 2011) genotyped across 32 Chinook salmon populations from throughout the Snake River. Summary statistics include minor-allele frequency (MAF) range, expected  $(H_E)$  and observed  $(H_O)$  heterozygosity, Weir and Cockerham (1984)  $F_{ST}$ , and fixation index ( $F_{IS}$ ). "HWE" designates the number of populations that a SNP deviated from Hardy-Weinberg expectation for any SNP that deviated in greater than 10% of populations. "LD" is the number of populations that a pair of SNP loci exhibited linkage disequilibrium if it occurred in more than half of the populations. "CS" indicates a locu that was designated as a candidate for divergent (+) or Table 5. Steelhead reporting regions for each baseline population used for GSI in the Snake River. Major population groups (MPG) are noted for each population to demonstrate the overlap of reporting regions with MPG......35 Table 6. Chinook salmon reporting regions for each population used for GSI in the Snake River. Major population groups (MPG) are noted for each 

# List of Tables

Table 7.	Results from leave-one-out tests performed in ONCOR (Kalinowski et al. 2007) for steelhead baseline v1.0. Rows represent population where individuals originated from. Columns represent reporting regions that individuals assigned to. Shaded boxes represent the correct reporting region of origin for each population.						
Table 8.	Results from leave-one-out tests performed in ONCOR (Kalinowski et al. 2007) for sp/sum Chinook salmon baseline v1.0. Rows represent population where individuals originated from. Columns represent reporting regions that individuals assigned to. Shaded boxes represent the correct reporting region of origin for each population	40					

# LIST OF FIGURES

Figure 1.	Steelhead populations representing steelhead baseline v1.0 for the Snake River. Population numbers correspond to numbers in Table 1. The locations of Lower Granite Dam and Hells Canyon Dam is noted	42
Figure 2.	Chinook salmon populations representing Chinook baseline v1.0 for the Snake River. Population numbers correspond to numbers in Table 2. The locations of Lower Granite Dam and Hells Canyon Dam is noted	43
Figure 3.	NJ-phylogram based on Nei's (1972) distance for steelhead baseline v1.0. Numbers correspond to map numbers in Figure 1. Brackets designate reporting regions used for genetic stock identification	44
Figure 4.	NJ-phylogram based on Nei's (1972) distance for Chinook salmon baseline v1.0. Numbers correspond to map numbers in Figure 1. Brackets designate reporting regions used for genetic stock identification. A sample from John Day River was included as an out-group	45
Figure 5.	Mean pairwise $F_{ST}$ estimates for baseline steelhead populations. The dashed line represents the average pairwise $F_{ST}$ estimate for all populations. High mean pairwise $F_{ST}$ estimates suggest high levels of genetic differentiation relative to other baseline populations.	46
Figure 6.	Mean pairwise $F_{ST}$ estimates for baseline Chinook salmon populations. The dashed line represents the average pairwise estimate for all populations. High mean pairwise $F_{ST}$ estimates suggest high levels of genetic differentiation relative to other baseline populations	47
Figure 7.	Stock proportions for SY 2009 (7/1/2008 – 6/30/2009) and SY2010 (7/1/2009 – 6/30/1010) natural origin summer-run steelhead at Lower Granite Dam.	48
Figure 8.	Stock proportions for SY2009 natural origin spring/summer Chinook salmon at Lower Granite Dam	48
Figure 9.	Sex ratios for SY2009 (7/1/2008 – 6/30/2009) and SY2010 (7/1/2009– 6/30/1010) for each reporting region for natural origin summer-run steelhead at Lower Granite Dam	49
Figure 10.	Sex ratios for SY2009 for each reporting region for natural origin spring/summer Chinook salmon at Lower Granite Dam.	50
Figure 11.	Run-timing at Lower Granite Dam for each of the reporting groups based on date of capture of adult steelhead sampled at the adult trap located on the fish ladder. Run timing includes median passage date with $5^{th}$ , $25^{th}$ , $75^{th}$ , and $95^{th}$ percentiles. Passage date refers to days from July 1 (i.e. July 1 = Passage Date 1).	51
Figure 12.	Run-timing at Lower Granite Dam for each of the reporting groups based on date of capture of adult Chinook salmon sampled at the adult trap located on the fish ladder. Run timing includes median passage date with $5^{\text{th}}$ , $25^{\text{th}}$ , $75^{\text{th}}$ , and $95^{\text{th}}$ percentiles. Julian date refers to days from January $1^{\text{st}}$ (i.e. January 1 = Julian Date 1)	52

# LIST OF APPENDICES

# <u>Page</u>

Appendix A.	TaqmanTM assays used for <i>O. mykiss</i> . All forward and reverse primers are unlabeled and probes contain a 5' fluorophore and a 3' quencher and minor groove binder.	54
Appendix B.	TaqmanTM assays used for <i>O. tshawytscha</i> . All forward and reverse primers are unlabeled and probes contain a 5' fluorophore and a 3' quencher and minor groove binder	63

#### ABSTRACT

This report summarizes the progress in development and implementation of genetic stock identification (GSI) technologies within the Snake River basin for natural origin steelhead and spring/summer Chinook salmon passing Lower Granite Dam for the reporting period of 07/01/2010 to 06/30/2011. Three objectives for the project are addressed in this report: 1) the selection and evaluation of single nucleotide polymorphism (SNPs) panels for high-throughput genotyping of both steelhead and Chinook salmon in the Snake/Columbia River basins, 2) the development of initial SNP baselines to describe genetic variation and for use as a reference in GSI methods for both species in the Snake River basin, and 3) the implementation of GSI to estimate the stock composition of steelhead and spring/summer Chinook salmon passing Lower Granite Dam. For objective 1, the goal was to identify standardized sets of SNPs that could be used for both GSI and parentage-based tagging (PBT) for studies throughout the Snake/Columbia River basins. Standardized SNP panels will allow data to be exchanged between laboratories to facilitate collaboration and to avoid duplication of efforts and will allow the integration of sampling programs at Bonneville Dam and Lower Granite Dam and in Columbia River and Snake River mainstem fisheries. All sampled wild adult can be included in mixture analyses to determine stock composition and sampled Snake River hatchery adult can be assigned to a stock and cohort. For objective 2, using the identified standardized sets of SNPs, we genotyped collections of steelhead and Chinook salmon populations from throughout the Snake River to describe genetic variation for both species and to provide a baseline that could be used for GSI estimations. Section 2 of this report briefly describes the genetic variation observed among populations for both species across the Snake River basin and also describes genetic variation for each SNP marker. Finally, in objective 3, we implemented GSI at Lower Granite Dam using the baselines developed in objective 2 to estimate stock compositions of steelhead and Chinook salmon passing Lower Granite Dam. In section 3, we report initial results for adult steelhead for spawn year (SY)2009 (07/01/2008 - 06/30/2009) and SY2010 (07/01/2009 - 06/30/2010) and for adult Chinook salmon for SY2009 (03/01/2009 -08/17/2009).

Authors:

Mike Ackerman Fisheries Research Biologist

Matt Campbell Genetics Laboratory Manager

#### INTRODUCTION

Abundance (i.e. number of adults on spawning grounds) is a primary metric needed for monitoring the status of steelhead and salmon populations in the Columbia River basin (McElhany et al. 2000). Estimates of abundance combined with age and sex information over time allows estimation of population growth rates. Both these metrics provide indicators of the resiliency and viability of populations, and allows assessments of extinction risk. Estimates of these metrics at the stock or population level is information that fisheries managers can use to achieve sustainable harvest of larger populations, while protecting weaker stocks and the biodiversity present within them. Population level assessments of abundance and productivity for ESA threatened Snake River steelhead and Chinook salmon are particularly difficult due to the wide distribution and location of spawning areas (many populations are present in remote or wilderness areas). Additionally, environmental conditions at the time of spawning, especially for steelhead populations with fall and winter adult migration, often prevent the use of traditional counting methodologies (operation of weirs, rotary screw traps, and redd-count surveys). This is less of a problem for spring/summer Chinook salmon, although turbid water conditions resulting from storms and forest fires have impacted the ability to estimate adult abundance using reddbased surveys in the Middle Fork Salmon River and South Fork Salmon River (Thurow 2000).

Snake River steelhead monitoring is further hampered due to high turbidity and changing flow conditions during the time of spawning (Thurow 1985). As a result, escapement estimates (and other demographic information) are not available for most Snake River stocks (Busby et al. 1996; Good et al. 2005). In lieu of more detailed basin-level and stock-specific information, steelhead in the Snake River basin have traditionally been assigned to two groups (A-run and B-run), based on life history characteristics and bimodal timing of passage at Bonneville Dam in the middle Columbia River (Busby et al. 1996). By definition, A-run steelhead pass Bonneville Dam before August 25 and tend to return after one year in the ocean. B-run steelhead pass Bonneville Dam after August 25, tend to return after two years in the ocean, and are thought to be larger at age than A-run steelhead. Upstream migrating steelhead adults at Lower Granite Dam do not exhibit a bimodal passage distribution and A-run and B-run adults are enumerated based on length (A-run, ≤78 cm; B-run, >78 cm) as a proxy for ocean age. In addition to runtiming at Bonneville Dam and size differences, the two stocks are believed to exhibit differences in spawning distribution. A-run adult steelhead are thought to spawn throughout the Columbia basin, whereas B-run steelhead are believed to originate only in the Clearwater, Middle Fork Salmon, and South Fork Salmon rivers in Idaho. These putative migration timing, morphological, and life history characteristics have been used as a surrogate for biodiversity in conservation planning for Snake River steelhead. However, the relationship between the morphological and life history characteristics to time of passage at Bonneville Dam is uncertain (Good et al. 2005). Further, the bimodal passage distribution at Bonneville Dam has become unimodal in recent years (Robards and Quinn 2002).

Two main management concerns regarding Snake River steelhead have arisen in the last several years. First, populations classified as B-run do not appear to be self-sustaining (NMFS 2007) and their presence in the basin have affected operation of the Columbia River hydrosystem and fisheries management in the lower Columbia River. In particular, harvest of fall Chinook salmon is constrained in order to limit impacts to B-run fish that are concurrently present in the Columbia River. Secondly, there are substantial data needs to refine population delineations and conservation assessments (ICTRT 2003), but data have been lacking. Although Snake River "B-run" steelhead are currently identified as a biologically significant and distinct component of the Snake River ESU, their management is confounded by the lack of a clear and detailed understanding of their actual spawning distribution and population structure.

Nielsen et al. (2009) found that steelhead in Snake River tributaries in Idaho exhibit a complicated pattern of genetic structure with populations grouping genetically according to drainage locality, not simply by "A-run" or "B-run" designations.

These two issues and similar biological and management questions relating to Snake River steelhead and Chinook salmon may be addressed through GSI methodologies. Genetic stock identification uses multi-locus genotype data from reference populations (presumably representing all contributing stocks) as a baseline and a complimentary set of genotype data from mixtures of fish of unknown origin to estimate stock proportions within the mixture (Shaklee et al. 1999; Anderson et al. 2008). GSI technologies have been used extensively to understand and manage mixed stock fisheries for a variety of Pacific salmonids including Chinook salmon (Smith et al. 2005), sockeye salmon (Habicht et al. 2010), coho salmon (Beacham et al. 2001) and steelhead (Beacham et al. 2000). In the Snake River basin, studies have indicated that both Chinook salmon and steelhead exhibit significant genetic structuring at the watershed (or subbasin) level (Moran 2003; Narum et al. 2007; Nielsen et al. 2009). Previously, researchers have made use of this genetic structure to identify the stock origin of kelt steelhead at Lower Granite Dam (Narum et al. 2008b) and to estimate the stock composition of wild and hatchery Chinook salmon over Lower Granite Dam (Smith 2007). Narum et al. (2008b) examined kelts (post-spawn adult steelhead attempting to return to the ocean) using a set of 14 nonstandardized microsatellite loci and 20 baseline populations for GSI estimation. Smith (2007) used 13 standardized microsatellite loci (see Seeb et al. 2007) and 38 populations for GSI estimation.

The results of the studies summarized above demonstrate the utility of GSI technology to obtain stock abundance estimates for steelhead and Chinook salmon in the Snake River basin. Continued development and evaluation of this management tool has been strongly recommended by regional RME workgroups and similar work initiated at Bonneville Dam and in the lower Columbia River has been supported by the Independent Scientific Review Panel (http://www.nwcouncil.org/library/isrp/08-15.pdf).

## OBJECTIVES

## **Objective 1. Discover and Evaluate SNP Marker Sets**

One of the highest priorities in the full-scale implementation of SNPs for salmon genetics is the discovery, selection, and development of a sufficient number of SNP markers to characterize population variability. SNPs are the most abundant form of variation in the genome of most organisms and can be discovered throughout the genome of non-model organisms with relative ease (Brumfield et al. 2003). SNPs are attractive for large-scale GSI efforts in that their bi-allelic nature allows for highly automated, rapid genotyping (Schlotterer 2004; Van Tassell et al. 2008; Seeb et al. 2009a) with low error rates (Morin et al. 2004) which facilitates standardization across laboratories. These characteristics make SNPs ideal for collaboration among agencies involved in Pacific salmon GSI (Narum et al. 2008a). In the first year of this project, the CRITFC and IDFG labs developed and screened hundreds of SNPs for both steelhead and Chinook salmon to identify powerful sets of loci to use in GSI and parentage based tagging (PBT; BPA project #2010-031-00) projects. Section 1 will briefly describe the processes of selecting and evaluating the SNPs used for GSI projects in the Snake River.

#### **Objective 2. Construct SNP Genetic Baselines**

Currently, genetic baselines using microsatellite markers exist for steelhead in the interior Columbia basin and for Chinook salmon across their coastwide range (Seeb et al. 2007). However, despite large representative baseline samples from many Columbia/Snake River populations and the high allelic diversity of microsatellite markers, the spatial resolution of specific stocks and populations in these baselines is limited in some cases. Given the difficulty and expense of inter-laboratory standardization of microsatellites, adding further markers may not be an efficient option for attaining the desired levels of resolution in problem regions or for long term monitoring evaluations. In this regard, SNP markers are the preferred option because they are easy to identify and develop into genetic assays and they are amenable to rapid standardization across laboratories. For this objective, IDFG and CRITFC labs screened many of the Snake River samples/populations that had previously been genotyped with microsatellite markers as part of coastwide GAPS (Genetic Analysis of Pacific Salmonids [Chinook salmon], Seeb et al. 2007) and SPAN (Stephen Phelps Allele Nomenclature [steelhead], Blankenship et al. 2011) efforts. This screening resulted in the first SNP genetic baselines for both species in the Snake River basin, providing an initial reference to assess stock discrimination and estimate the expected accuracy of GSI (Anderson et al. 2008). Section 2 describes these initial efforts and reviews planned expansion and refinement of these baselines (Snake River baselines v2.0) that will occur during the 2<sup>nd</sup> year of this project.

## Objective 3. Use GSI Methods to Estimate Stock Compositions at Lower Granite Dam.

Genetic stock identification (GSI) has proven to be an effective method for estimating the stock composition of fisheries mixtures in several applications (Shaklee et al. 1999; Narum et al. 2008a; Seeb et al. 2009b; Hess et al. 2011b). For this objective, we implemented GSI procedures to estimate the stock composition of adult steelhead and Chinook salmon at Lower Granite Dam. Information generated from this objective should assist managers and researchers with estimating viable salmonid population (VSP) parameters including abundance, population productivity, and diversity for these two species in the Snake River basin.

## **REPORT STRUCTURE**

This report is divided into three sections, one for each of the objectives of the study. Section 1 addresses the ascertainment and evaluation of the SNP markers used for GSI. Section 2 deals with efforts to construct, evaluate, and maintain genetic baselines for steelhead and Chinook salmon in the Snake River basin. Section 3 addresses the use of GSI methods to estimate the stock composition of adult steelhead and Chinook salmon passing Lower Granite Dam.

Note: In this report we refer to adult steelhead and Chinook salmon passing above Lower Granite Dam by spawn years (SY). For steelhead, a spawn year refers to adults that migrate over Lower Granite Dam in the previous year's fall and the current year's spring (e.g. SY2009 steelhead are adults that migrated over the dam in the fall of 2008 and the spring of 2009 and spawned in spring of 2009). For Chinook salmon, a spawn year refers to adults that migrate over the dam in the summer and spawn that same fall (e.g. SY2009 Chinook salmon migrated above the dam during the summer of 2009 and spawned later that year).

### SECTION 1: DISCOVER AND EVALUATE SNP MARKER SETS

#### Introduction

Genetic stock identification (GSI) applications require the use of an informative suite of genetic markers for assignment of individuals to stock of origin. One of the highest priorities in the full-scale implementation of SNPs for GSI is the selection and evaluation of a sufficient number of loci to characterize genetic variability among populations within the study area. For this objective, CRITFC and IDFG labs developed and screened hundreds of SNPs for both steelhead and Chinook salmon to identify powerful sets to use in GSI and parentage based tagging (PBT; BPA project #2010-031-00) studies. Objective 1 of this project corresponds closely to Objective 2 of BPA project #2010-031-00, the GSI project described here implements the same SNP markers described for PBT studies. Therefore, section one will provide only a brief synopsis of relevant methods (for additional details see section 2 of BPA project #2010-031-00 annual report).

#### <u>Methods</u>

In order to select the most informative subset of markers for PBT and GSI applications a total of 395 and 245 SNP markers were evaluated for steelhead and Chinook salmon, respectively. All available SNP markers for steelhead were used to genotype 288 individuals representing 7 collections primarily from throughout the Columbia/Snake River basins. The 245 available SNP markers for Chinook salmon were used to genotype 16 individuals each from six Snake River hatcheries. In addition, a modified sex-specific assay for both steelhead (*Omy\_SEXY1*) and Chinook salmon (*Ots\_SEXY1*) was developed in order to differentiate sex in these species. Accuracy of genetic sex typing was determined by comparing genotypes to phenotypic sex recorded from hatchery broodstock during spawning activities. SNPs were chosen based on genotyping robustness, lack of deviations from Hardy-Weinberg and linkage equilibria, and minor allele frequency (MAF) rank.

#### <u>Results</u>

Markers that tested poorly, lacked variation, and/or violated Hardy-Weinberg expectation (HWE) were not considered for further evaluation. In cases where two markers were found to be significantly linked, the marker with the lower MAF was discarded. All markers retained after screening were sorted by MAF within the Snake River hatchery samples, and the top 95 markers were preferentially selected for the PBT panels for each species (see Tables 7-8 in BPA project #2010-031-00, 2010 annual report). The GSI panel for steelhead included the next 95 most variable markers that did not overlap the PBT panel. For Chinook salmon, the GSI panel had been determined prior to PBT panel selection and was maintained in its original form for consistency with ongoing studies (e.g. Hess et al. 2011a; Hess et al. 2011b). The resulting GSI and PBT panels for Chinook salmon overlap by 50 markers. Initial analyses to examine concordance of steelhead genotypes between the CRITFC and EFGL labs indicate >99.5% concordance. Initial results to examine the accuracy of the sex markers indicate 98.4% and 88.0% concordance with phenotypic sex for steelhead and Chinook salmon, respectively.

#### **Discussion**

Initial screening of SNP markers was used to choose three panels of 96 markers (one PBT set for each species and an additional set for steelhead GSI). These 96-marker subsets were then further evaluated by genotyping a larger number of collections representing the

Columbia River and several Snake River hatcheries. This second-round evaluation of markers identified the most appropriate subsets of markers for PBT based on high MAF, minimal deviations from HWE and LD, high parentage accuracy, and concordance of repeat genotyping. The remaining high ranking markers were used to populate the GSI panel for steelhead. The GSI panel for Chinook salmon had been determined prior to the PBT panel selection and will remain in its original form for the current year, despite overlap of 50 markers with the PBT panel. Future modification to the Chinook salmon GSI panel is planned and should significantly improve our ability to ascertain the stock of origin of Chinook salmon in the Snake River in GSI analyses.

For further details regarding the selection and evaluation of SNP for IDFG's PBT and GSI projects, please see Section 2 of the 2010 annual report for BPA project #2010-031-00.

#### **SECTION 2: SNP BASELINES**

#### Introduction

For objective two of this project, initial SNP genotypic baselines were constructed for both steelhead and spring/summer Chinook salmon throughout the Snake River basin. Currently, genetic baselines using microsatellite markers exist for steelhead and Chinook salmon in the Columbia basin (Narum et al. 2010b; Blankenship et al. 2011), and more broadly for Chinook salmon across their coastwide range (Seeb et al. 2007). However, despite large, representative baseline samples from many Columbia/Snake river populations and the high allelic diversity of microsatellite markers, the resolution of specific stocks and populations in these baselines is limited in some cases. Additionally, given the difficulty and expense of interlaboratory standardization, adding further microsatellite markers may not be an efficient option for attaining the desired levels of resolution in problem regions in the microsatellite baselines. In this regard, SNP markers are the preferred option for adding loci because they offer many beneficial characteristics that make them amenable to adding loci and to standardization across laboratories. For baseline development, we worked closely with CRITFC with the primary goal of genotyping a large proportion of Snake River samples/populations previously genotyped with microsatellite markers as part of coastwide GAPS and SPAN efforts. This will create the first SNP genetic baselines for both species in the Snake River basin from which we can assess stock discrimination and estimate the expected accuracy of GSI (Anderson et al. 2008).

Ultimately, the initial SNP baselines for both species will be temporally and spatially expanded or updated through annual population sampling and genotyping. Future annual sampling plans will be based on resolution of initial baselines and managers' specific needs. A general plan would involve sampling stocks/reporting groups on a rotational basis. For example, if 10 reporting groups are identified for Snake River steelhead following initial baseline construction; rotational screening may involve sample may be obtained from juvenile fish sampled during outmigration at a trap (e.g. Lemhi River rotary screw trap). For other reporting groups, we expect to representatively sample from major contributing populations, and obtain approximately 50 samples per population (four populations per reporting group annually). Based on previous experience with microsatellite baselines, we expect that SNP baseline refinement and maintenance can be accomplished through the genotyping of ~1,000 samples per year. This annual sampling/genotyping plan will provide a robust baseline from which GSI mixture analyses can be performed and refined. At the same time it will provide temporal monitoring of genetic diversity and structure across the basin allowing estimation of gene flow (homing vs.

straying) and effective population size. Any baseline data produced under this objective will be made available to SPAN and GAPS collaborators for inclusion in other steelhead and Chinook salmon GSI studies within or including the Snake/Columbia basins. This will allow Snake River steelhead and spring/summer Chinook salmon to also be identified in GSI studies outside the Columbia River and on the high-seas.

#### Methods

#### Sample Collection

For initial development of multi-locus SNP baselines for steelhead and spring/summer Chinook salmon in the Snake River, populations were included from throughout the Snake River basin. When available, populations were chosen to complement or overlap collections previously genotyped and submitted to the standardized SPAN and GAPS microsatellite consortiums for steelhead and Chinook salmon, respectively. Genotyping of overlapping collections/populations with both microsatellites and SNPs allows for evaluation of multiple marker data sets and subsequent distribution of baseline data to the identified consortiums. Our goal for coverage of both summer-run steelhead and spring/summer Chinook salmon is to have all major contributing populations in the Snake River basin represented. Ideally, multiple populations from all primary drainages would be represented and each population would be represented by temporal collections (sampled in more than one year) of an adequate sample size (i.e. >50 fish). All steelhead populations identified for genotyping for the initial baseline were genotyped using the 192 SNPs identified in section 1 and Appendix A. The panel of 192 loci includes 96 SNPs that are used for IDFG's PBT project (BPA Project #2010-031-00) plus an additional 96 used only for GSI applications. For Chinook salmon, we are employing the initial set of SNPs used by CRITFC for GSI applications in the lower Columbia River (Hess et al. 2011a; Matala et al. 2011).

Biological tissues for genetic analysis of all juvenile collections were sampled from rayed fins. Biological tissues for genetic analysis of adult collections were sampled from multiple sources; 1) rayed fins, 2) opercle punches (generally adults passed above a weir), or 3) carcass tissue (from adult Chinook salmon carcass surveys). For tissues genotyped at the IDFG lab, samples were originally stored in individually labeled vials containing 200-proof denatured ethyl alcohol. For some collections genotyped at the CRITFC lab, samples were stored using a dry Whatman paper medium (Lahood et al. 2008). For further details on sample storage and genotyping for samples at the CRITFC lab see 2010 annual report for BPA Project #2008-97-00 (Hess et al. 2011a). Baseline samples were contributed by multiple agencies including IDFG, CRITFC, NOAA Fisheries, Oregon Department of Fish and Wildlife (ODFW), and Washington Department of Fish and Wildlife (WDFW).

## Laboratory Protocol

DNA was extracted using the Nexttec Genomic DNA Isolation Kit from XpressBio (Thurmont, MD) or QIAGEN DNeasy Tissue Kits (Valencia, CA). Prior to DNA amplification of SNP loci using primer-probe sets (fluorescent tags), an initial polymerase chain reaction (PCR) "pre-amp" was implemented using whole genomic DNA to jumpstart SNP amplification via increased copy number of target DNA regions. The PCR conditions for the pre-amp step were as follows: an initial mixing step of 95° C for 15 min, followed by 14 cycles of 95° C for 15 seconds and 60° C for four minutes, ending with a final 4° C dissociation step. For steelhead, all individuals were genotyped at 191 SNPs (including three SNPs that identify potential *O. mykiss* and *O. clarkii* hybrids) and a Y-specific allelic discrimination assay that differentiates sex in *O.* 

mykiss. For Chinook salmon, all individuals were genotyped at 95 SNPs (including one mtDNA SNP) and a Y-specific allelic discrimination assay that differentiates sex in O. tshawytscha. Genotyping was performed using Fluidigm 96.96 Dynamic Array IFCs (chips). For each genotyping run, 96 samples (including an extraction negative control, a PCR negative control, and a PCR positive control) and 96 TagMan SNP assays were hand-pipetted onto the 96.96 chips. Sample cocktail and SNP assay cocktail recipes are available by request from the primary author (mike.ackerman@idfg.idaho.gov). Each 96.96 chip was pressurized to load the DNA and SNP assays into the array using a Fluidigm IFC Controller HX. SNP amplification on the 96.96 chips were performed using either an Eppendorf Stand-Alone Thermal Cycler (protocol: thermal mixing step of 50° C for 2 min, 70° C for 30 min, and 25° C for 10 min, a hotstart step of 50° C for 2 min and 95° C for 10 min, followed by 50 cycles of 95° C for 15 sec and 60° C for 60 sec, and a final cool down step of 25° C for 10 min) or a Fluidigm FC1 Fast-cycler (protocol: thermal mixing step of 70° C for 30 min and 25° C for 10 min, a hot-start step of 95° C for 60 sec, followed by 50 cycles of 95° C for 5 sec and 25° C for 25 sec, and a final cool down step of 25° C for 10 min). Chips were imaged on a Fluidigm EP1 system and analyzed and scored using the Fluidigm SNP Genotyping Analysis Software version 3.1.1. The laboratory methods in use at the IDFG EFGL are very similar to those employed at the CRITFC genetics laboratory.

Standardized genotypes were stored on a Progeny database server housed at EFGL. Progeny software (<u>http://www.progenygenetics.com/</u>) is in use by a majority of GAPS and SPAN labs throughout the Pacific Northwest: Idaho Department of Fish and Game, University of Washington, NOAA – Northwest Fisheries Science Center, Washington Department of Fish and Wildlife, CRITFC, and U.S. Fish and Wildlife Service. This commonality of database software will further promote seamless sharing of data among labs in the future.

#### **Statistical Analyses**

Allele frequencies for baseline collections were calculated using GENALEX version 6.4 (Peakall and Smouse 2006). Collections taken at the same location across multiple years were tested for genetic differentiation across all loci using pairwise exact tests in GENEPOP version 4.0 (Rousset 2008) and were pooled as suggested by Waples (1990) if temporal collections failed to demonstrate significant departures from genetic homogeneity ( $\alpha = 0.05$ ). Markov chain (MC) parameters for pairwise exact tests in GENEPOP version 4.0 were as follows: dememorization = 10,000; batches = 100; iterations per batch = 5,000. Pooled collections were defined as populations in all subsequent analyses.

Tests for deviation from Hardy-Weinberg expectation (HWE) were performed across all loci for each population using exact p-values calculated from the MC method in GENEPOP version 4.0. Default parameters were used for the MC algorithm (dememorization = 1,000; batches = 20; iterations per batch = 5,000). Critical values ( $\alpha$  = 0.05) were adjusted for multiple tests using a step-down sequential Bonferroni correction (Holm 1979). Tests for deviation from HWE were conducted to investigate for non-random mating within populations (sample resembles more than one population) and to identify possible SNP amplification issues (i.e., null alleles).

Tests for linkage disequilibrium (LD) between all locus pairs were performed using simulated exact tests in GENEPOP version 4.0. A pair of loci was determined to be significantly out of linkage equilibrium if tests were significant ( $\alpha = 0.05$ ) in more than one-half of the baseline populations. If the test was significant between a pair of SNPs, the less informative of the SNP

pair was dropped (according to  $F_{ST}$ ) to avoid violating the assumption of independence of loci during population genetics or GSI analyses.

Baseline populations were evaluated for expected ( $H_E$ ) and ( $H_O$ ) observed heterozygosity and population-specific  $F_{ST}$  using GENALEX version 6.4. A higher heterozygosity indicates high levels of genetic variability within a population while a low heterozygosity may indicate low genetic variability due to various reasons (population bottlenecks, reduced metapopulation dynamics). The population-specific  $F_{ST}$  is an indicator of the level of differentiation a population exhibits relative to all other baseline populations. Population-specific  $F_{ST}$  for baseline populations are plotted in Figures 5 and 6.

Each SNP was evaluated for its within-population diversity and among-population information content. The expected and observed heterozygosities for each SNP were calculated using GENALEX version 6.4. Locus-specific heterozygosities are an indicator of the amount of genetic variability seen within baseline populations for each SNP. The Weir and Cockerham (1984)  $F_{ST}$  and  $F_{IS}$  statistics for each SNP were calculated using FSTAT version 2.9.3.2. Finally, we used the program LOSITAN (Antao et al. 2008) to test neutrality of nuclear SNP loci ( $\alpha$  = 0.01). LOSITAN evaluates the relationship between  $F_{ST}$  and  $H_E$  for all loci in an island model to identify outlier loci having excessively high or low  $F_{ST}$  compared to neutral expectations. Results were based on 50,000 data simulations using an infinite alleles model and a false discovery rate of 0.1. SNPs lying above or below the given criteria (outliers) are candidates for directional or balancing selection (respectively) in some populations.

We created a neighbor-joining (N-J) phylogram for each species to visualize the genetic relationship among baseline populations and to assist in the determination of reporting groups to be used for GSI. The N-J phylogram was based on pairwise Nei's (1972) genetic distances calculated using GENDIST (PHYLIP version 3.5; Felsenstein 1993). Pairwise genetic distances were used to construct an N-J phylogram in NEIGHBOR (PHYLIP version 3.5). The consistency of the phylogram topology was estimated using 1,000 bootstrap replicates in SEQBOOT (PHYLIP version 3.5). The final N-J phylogram was constructed using TREEVIEW (Page 1996) with observed consensus bootstrap values greater than 75% identified at nodes added (Figures 3 and 4).

#### <u>Results</u>

In total, steelhead baseline v1.0 consists of 49 populations represented by 52 collections (three populations with temporal collections) and 2,514 individuals (Table 1). Of the 52 collections, 36 were genotyped at the EFGL and 16 were genotyped at the CRITFC lab. For the three populations with collections from multiple years, collections were not significantly differentiated ( $\alpha = 0.05$ ). Average sample size for the 49 populations was 51 individuals.

Chinook salmon baseline v1.0 consists of 32 populations represented by 54 collections (12 populations with temporal collections) and 2,390 individuals (Table 2). Of the 54 collections, 45 were genotyped at the EFGL and 9 were genotyped at the CRITFC lab. For the 12 populations with collections from multiple years, collections were not significantly differentiated. Average sample size for the 32 populations was 75 individuals. See Narum et al. (2010a) and Hess et al. (2011a) for a complete list of baseline collections genotyped at the CRITFC genetics laboratory for GSI applications throughout the Columbia River basin.

For steelhead baseline v1.0, 383 out of 8,388 tests for deviation from HWE (across loci and populations) were significant (419 would be expected by chance at  $\alpha$  = 0.05). For Chinook

salmon baseline v1.0, 134 out of 2,397 tests for deviation from HWE were significant (120 would be expected by chance at  $\alpha = 0.05$ ). Tables 1 and 2 identify baseline populations that deviated from HWE at greater than 5% of loci for steelhead and Chinook salmon, respectively. No populations deviated from HWE at greater than 9% of loci. Tables 3 and 4 identify SNPs that deviated from HWE in greater than 10% of baseline populations for steelhead and Chinook salmon, respectively.

For steelhead, significant LD was found between one pair of SNP loci ( $Omy_GHSR-121$  and  $Omy_mapK3-103$ ) in 26 of the 49 populations. Locus  $Omy_mapK3-103$  was the less informative of the pair based on  $F_{ST}$  and was dropped from GSI analyses. For Chinook salmon, significant LD was found between two pair of SNP loci in more than half of the populations. Tests for the first pair ( $Ots_hsc71-5'-453$  and  $Ots_hsc71-3'-488$ ) were significant in 23 of the 32 populations. Tests for the second pair ( $Ots_FGF6A$  and  $OtsFGF6B_1$ ) were significant in all 32 populations. Among the two locus pairs,  $Ots_hsc71-3'-488$  and  $Ots_FGF6A$  were the least informative based on  $F_{ST}$  and each was removed from subsequent GSI analyses.

Tables 1 and 2 summarize within-population ( $H_E$  and  $H_O$ ) and among-population ( $F_{ST}$ ) diversity statistics for steelhead and Chinook salmon, respectively. For steelhead, the minimum  $H_O$  of 0.270 was observed in the Crooked River population and the maximum  $H_O$  of 0.317 was observed in the Morgan Creek population. The North Fork Salmon River, Hazard Creek, and Asotin Creek populations were the least differentiated populations in the steelhead baseline ( $F_{ST} = 0.017$ ) while the Pistol Creek population was the most differentiated ( $F_{ST} = 0.035$ ). For Chinook salmon, the minimum  $H_O$  of 0.187 was observed in the Sulphur Creek population and the maximum  $H_O$  of 0.280 was observed in the Tucannon River population. The South Fork Salmon River weir and Lolo Creek populations were the least differentiated populations in the Sulphur South Fork Salmon Baseline ( $F_{ST} = 0.013$ ) while the Chamberlain Creek population was the most differentiated populations in the South Fork Salmon Baseline ( $F_{ST} = 0.013$ ) while the Chamberlain Creek population was the most differentiated populations.

Tables 3 and 4 indicate steelhead and Chinook salmon SNPs, respectively, that are candidates for directional or balancing selection. Generally only neutral loci are used in population structure analyses, but exclusion of SNPs putatively under selection did not occur prior to generation of results. Determining the appropriate threshold for identifying selection in genetic markers is a topic of concern in fisheries genetics (Narum and Hess 2011) and requires a more thorough examination of the data (see Matala et al. 2011; Narum et al. 2011) than occurs here. For steelhead, 22 of the SNPs analyzed were candidates for directional selection. For Chinook salmon, two of the SNPs were candidates for directional selection including *Ots\_MHC2* which is located on a gene responsible for the production of major histocompatibility complex (MHC) class III molecules associated with the immune system in vertebrates (Bernatchez and Landry 2003). Several studies support the adaptive nature of MHC markers associated with pathogen mediated selection in salmon (Miller et al. 2001; Dionne et al. 2009; Ackerman et al. 2011). A more thorough examination of the SNPs identified as candidates for selection will be conducted in the future.

The genetic relationship of baseline populations for the steelhead and Chinook salmon baselines are depicted in Figures 3 and 4, respectively. Consensus of N-J tree topology is identified by bootstrap values observed at nodes in greater than 75% of the iterations; populations are bracketed according to the reporting groups used for GSI analysis. For the most part, both steelhead and Chinook salmon populations clustered according to geographic region or major tributary. Population-specific  $F_{ST}$  for steelhead and Chinook salmon populations relative to the average pairwise  $F_{ST}$  for all populations are plotted in Figures 5 and 6, respectively. Populations with an  $F_{ST}$  greater than the average pairwise  $F_{ST}$  among all populations are generally more genetically distinct and more identifiable in GSI analyses relative to populations with an  $F_{ST}$  less than the average pairwise  $F_{ST}$ . For steelhead, populations in more terminal regions of the Snake River (i.e. Middle Fork Salmon, South Fork Salmon, upper Clearwater, South Fork Clearwater) tend to be more identifiable than populations in lower portions of the Snake River (i.e. lower Clearwater, Imnaha, Grande Ronde). Similar patterns of population identifiability with terminal populations being more highly identifiable for GSI analyses have been observed elsewhere (Ackerman et al. 2011). For Chinook salmon, populations in the upper Salmon, South Fork Salmon, and Middle Fork Salmon rivers are more highly identifiable than populations in the Clearwater and lower Snake River. The low variability observed in populations in the Clearwater and lower Snake River is likely due to genetic homogenization resulting from stocking history (e.g. Carson hatchery stock releases in the Clearwater basin).

### **Discussion**

The baselines presented here represent the initial attempt to provide representative coverage for both species throughout the Snake River drainage, and further, provide a "baseline" from which to evaluate the current resolution of the SNP baselines and identify problem areas in which further sampling/genotyping is needed. In year two of this project, in coordination with BPA Project #'s 199005500, 198909800, 199107300, 200301700, 201002800, and 199102800, we will identify regional disparities in resolution in the current baseline and implement further sample collection and genotyping to increase resolution in the identified areas. For example, in the current steelhead baseline v1.0, Asotin Creek was included in the Grande Ronde reporting group although Asotin Creek occurs in the lower Snake River MPG. This is because Asotin Creek was not acceptably identifiable (41% of baseline individuals assigned back to Grande Ronde reporting group with large misallocation to the Upper Salmon [19%] and Imnaha [14%] reporting groups) in the current baseline and thus was lumped into the Grande Ronde reporting region. The Asotin Creek population is currently represented by a collection of 49 juveniles. Ideally populations are represented by adults that are sampled near the spawning grounds because they are less likely to exhibit family group biases that may result from juvenile sampling. For steelhead baseline v2.0, we have identified collections of natural origin steelhead adults that were sampled and passed above a weir to spawn that will be used in an attempt to increase resolution in that region. Similar baseline issues will be identified elsewhere in the Snake River basin and targeted for sampling/genotyping.

In addition to adding baseline samples, we also intend to add additional SNPs for the Chinook salmon genetic baseline. The EFGL lab, in coordination with CRITFC and other GAPS labs are currently in the process of identifying a standardized panel of SNPs that can be used in GSI applications throughout the range of Chinook salmon. This will allow for natural origin Snake River Chinook salmon to be identified in larger Snake/Columbia basin GSI studies conducted by CRITFC and IDFG, as well as in coastwide and high seas studies conducted by other researchers in the GAPS consortium. After the GAPS standardized SNP panel is identified, CRITFC and IDFG baseline samples will be re-genotyped so that baselines are screened with the GAPS panel in addition to the current panel of SNPs in place (Table 4). This will provide additional resolution to the current Chinook baseline. Baseline maintenance and improvement described above will occur during year two of the project and the expanded baselines (Snake River baselines v2.0) will be described in next year's annual report. The expanded baselines should provide an increased ability to identify the stock of origin of individuals from fisheries mixtures in GSI analyses.

In year one of this project, we have assembled large multi-locus genotype data sets for steelhead and spring/summer Chinook salmon covering large portions of the Snake River basin. The baselines provide information regarding the spatial and temporal distribution of genetic variation among Snake River populations, allowing examination of both the degree of gene flow across regions or locations in the basin (homing vs. straying) and the relative influences of hatchery supplementation activities. Further, these data sets will allow us to evaluate the relationships between landscape and life-history and how such interactions influence the genetic structure of populations throughout the Snake River. In coordination with the CRITFC genetics lab, we will further examine these issues through preparation of peer-reviewed manuscripts.

#### SECTION 3: USE GSI METHODS TO ESTIMATE STOCK COMPOSITIONS AT LOWER GRANITE DAM

#### Introduction

For this objective, we implemented GSI methods to estimate the stock composition of adult steelhead and spring/summer Chinook salmon migrating above Lower Granite Dam. Information generated from this objective should assist managers and researchers with estimating VSP parameters (McElhany et al. 2000) including abundance, population productivity, and diversity for steelhead and Chinook salmon in the Snake River basin.

In year one of the project, we genotyped adult steelhead migrating above Lower Granite Dam during SY2009-SY2011. Here, we report initial GSI results for SY2009 and SY2010. GSI results for SY2011 will be reported in next year's annual report. For Chinook salmon, we have genotyped SY2009 and SY2010 adults from Lower Granite Dam. Initial GSI results from SY2009 are reported here. Results for SY2010 will be reported in next year's annual report. For both species, SY2009 individuals were genotyped at the EFGL. SY2010 individuals were genotyped at the CRITFC genetics lab.

#### **Methods**

#### Sampling at Lower Granite Dam

Systematic samples of Chinook salmon and steelhead returning to Lower Granite Dam were collected during daily operation of the adult fish trap by National Oceanic and Atmospheric Administration Fisheries Service (NOAA Fisheries; BPA Project #2005-002-00; Harmon 2003; Ogden *in prep*). The adult trap is located in the Lower Granite Dam fish ladder above the fish counting window. The trap captures fish systematically by opening and closing a trap gate four times per hour. A pre-determined sample rate dictates how long the gate remains open each time; the trap gate is operational 24 hours per day.

All adult fish captured were anesthetized, examined for external marks and tags, and evaluated for physical condition. Fish were then scanned for an internal coded wire tag (CWT) or PIT tag and measured for fork length (FL, nearest cm; Ogden *in prep*). All fish were classified by origin (adipose-intact wild, adipose-clipped hatchery, or adipose-intact hatchery). For adipose-intact Chinook salmon, the distinction between hatchery and wild fish was determined by the presence or absence of a CWT, which is found only in hatchery-reared fish. Unclipped steelhead with a CWT were considered hatchery. For unclipped steelhead without a CWT, the

distinction between hatchery and wild was determined by the presence or absence of dorsal and ventral fin erosion, which is believed to occur as a result of hatchery-rearing.

All captured wild fish were sampled for scales and genetics tissue by NOAAF or IDFG staff (BPA Projects #1991-073-00, Idaho Natural Production Monitoring and Evaluation Program; #1990-055-00, Idaho Steelhead Monitoring and Evaluation Studies). The genetic tissue sample was taken from a small clip of the anal fin. Samples were stored in a vial with 100% non-denatured ethanol for transport to the EFGL and CRITFC genetics laboratories. After processing, all fish were returned to the adult fish ladder to resume their upstream migration.

#### Laboratory Protocol

Laboratory methods follow those used in Section 2. For steelhead, individuals were genotyped using the SNPs listed in Table 3. Chinook salmon individuals were genotyped using the SNPs listed in Table 4.

#### **Statistical Analyses**

To evaluate the resolution of the baselines for individual assignment, we conducted "leave-one-out" tests using the program ONCOR. In leave-one-out tests, each individual from the baseline is removed (one at a time) and their population (or reporting group) of origin is estimated using the method of Rannala and Mountain (1997). For each baseline population, we calculated the proportion of individuals that correctly assigned back to their reporting group of origin and the proportion of individuals that assigned back to each of the incorrect reporting groups (Tables 7 and 8). For each baseline population, the goal is to have greater than 75% of individuals assign back to the correct reporting group.

Samples from Lower Granite Dam were genotyped using the same methods as used for the baselines (Section 2). Multi-locus genotype data from the unknown mixtures of fish at Lower Granite Dam were analyzed using the program ONCOR to assign each individual to their "best-estimate" reporting group-of-origin. The individual assignment option in ONCOR determines the "best-estimate" stock of origin based on the reporting group with the highest probability of assignment for a particular fish. Stock proportions based on individual assignment are expected to reflect accurate estimates of each stocks' total abundance because a consistent sub-sample rate of individuals captured at Lower Granite Dam is maintained for the entire migration season. Reporting groups were generated based on multiple sources of information: 1) the genetic relationship of populations based on an N-J phylogram, 2) major population group delineations, 3) the geographic structure of the Snake River (i.e. by watershed), and 4) the assignment accuracy of baseline populations using various reporting group iterations (i.e. various modifications of the current reporting groups were evaluated for increases in assignment accuracy). Reporting groups used for GSI for both steelhead and Chinook salmon are outlined in Tables 5 and 6 respectively.

Individuals analyzed were genotyped using modified sex-specific assays for steelhead (*Omy\_SEXY1*) and Chinook salmon (*Ots\_SEXY1*). Based on the sex-specific markers, we estimated sex ratios for each reporting group for both steelhead and Chinook salmon. In the future, this will allow us to estimate the numbers of males and females that return to each region and allow estimation of productivity parameters (i.e. recruits per female). Additionally, based on the trapping date and estimated stock of origin of each fish, we were able to evaluate and compare run-timing among reporting groups.

#### <u>Results</u>

For steelhead, we used ten reporting groups for GSI analyses of Lower Granite Dam samples. Reporting groups include 1) Upper Salmon, 2) Middle Fork Salmon, 3) South Fork Salmon, 4) Lower/Little Salmon, 5) Upper Clearwater, 6) South Fork Clearwater, 7) Lower Clearwater, 8) Imnaha, 9) Grande Ronde, and 10) Below LGD. The Below LGD reporting group was included to estimate of the number of fish that originate from below Lower Granite Dam that ascend the fish ladder and either 1) stray above Lower Granite Dam or 2) ascend and fallback to spawn below Lower Granite Dam. The Below LGD reporting groups is represented by collections from the Tucannon and Touchet rivers.

For Chinook salmon, we used five reporting groups for GSI analyses. Reporting groups include 1) Upper Salmon, 2) Middle Fork Salmon, 3) South Fork Salmon, 4) a group that includes the Clearwater, Grande Ronde, and Imnaha rivers and 5) Tucannon River. Again, the Tucannon River enters the Snake River below Lower Granite Dam and is included to estimate the number of fish that originate from the Tucannon River but ascend the fish ladder and either 1) stray above Lower Granite Dam or 2) ascend and fallback to spawn below Lower Granite Dam. Except for the inclusion of the Clearwater basin in the Clearwater/Grande Ronde/Imnaha reporting group, established groups align with defined major population groups (ICTRT 2003).

For steelhead, greater than 75% of baseline individuals assigned back to the correct reporting group of origin for 32 of the 49 baseline populations using leave-one-out tests (Table 7). Certain reporting groups were more highly identifiable relative to other groups. For example, all populations within the Middle Fork Salmon, South Fork Salmon, and Upper Clearwater reporting groups had greater than 75% of baseline individuals assign back to the correct reporting group. For Chinook salmon, greater than 75% of baseline individuals assigned back to the correct reporting group of origin for 17 of the 32 baseline populations during leave-one-out tests.

For SY2009 steelhead, the largest contributor to the aggregate run over Lower Granite Dam was the Grande Ronde reporting group (23.2%) followed by Upper Clearwater (11.3%), Upper Salmon (11.1%), South Fork Clearwater (10.5%), Lower Salmon (10.1%), Imnaha (8.9%), Middle Fork Salmon (8.8%), Lower Clearwater (6.8%), Below LGR (5.6%), and South Fork Salmon (3.8%). For SY2010 steelhead, the largest contributor was the Grande Ronde (30.9%) followed by Upper Salmon (16.3%), Middle Fork Salmon (10.1%), Imnaha (9.6%), South Fork Clearwater (7.9%), Upper Clearwater (7.2%), Below LGR (6.1%), Lower Salmon (5.1%), Lower Clearwater (3.6%), and South Fork Salmon (3.2%). Figure 7 summarizes the stock compositions of the aggregate runs over Lower Granite Dam for each year.

For SY2009 Chinook salmon, the largest contributor to the aggregate run over Lower Granite Dam was the Clearwater/Grande Ronde/Imnaha reporting group (33.5%) followed by South Fork Salmon (28.3%), Upper Salmon (23.3%), Middle Fork Salmon (12.6%) and Tucannon (2.4%; Figure 8).

Sex ratios for steelhead based on assay *Omy\_SEXY1* (98.3% accurate based on comparisons with known-sex hatchery broodstock) were female-biased for both SY2009 and SY2010 (Figure 9). For SY2009, 66% of the adults were determined to be female and for SY2010, 62% were female. Individuals assigned to the Middle Fork Salmon River exhibited the greatest female-biased sex ratio; 76% in SY2009 and 74% in SY2010.

Sex ratios for Chinook salmon based on assay *Ots\_SEXY1* (88.0% accurate based on comparisons with known-sex hatchery broodstock) was male-biased for all reporting groups in SY2009 (Figure 10). This is likely heavily influenced by the number of jacks present in the mixture analyzed migrating above Lower Granite Dam. For both species, sex ratios for separate ocean ages will be reported in the annual Lower Granite Dam report submitted through BPA Projects #1990-055-00, #1991-073-00, #1987-127-00 and this project (#2010-026-00).

Figures 11 and 12 summarizes the run-timing by reporting group based on the estimated reporting group of origin of each individual and the date that each fish was captured at the adult trap in the fish ladder at Lower Granite Dam. Figure 11 suggests run-timing differences among reporting groups for steelhead. For instance, the Middle Fork Salmon reporting group had the earliest median passage date for both SY2009 and SY2010. Alternatively, the South Fork Clearwater reporting group had the latest median passage date for both spawn years.

#### Discussion

Using the Snake River baselines v1.0, we were able to discern 10 reporting groups for steelhead and five reporting groups for Chinook salmon. The 10 reporting groups for steelhead achieved using SNPs is of greater resolution than was achieved in the past using microsatellite markers. This increase in resolution is largely a result of our ability to screen baseline and mixture individuals across a very large number of SNPs efficiently and in a high-throughput manner. Although microsatellite markers are highly variable, and thus individually very informative for mixture analyses, the high-throughput capabilities when genotyping using SNPs allow us to use a large enough number of markers to surpass the resolution attained by microsatellites. The five reporting groups for Chinook salmon in the Snake River using the current 96 SNPs is of a similar level of resolution as has been observed in the past using microsatellite markers. This level of resolution was largely expected based on previous results from GSI in the lower Columbia River (Narum et al. 2010a; Hess et al. 2011a) using this SNP panel. Using the current panel of Chinook salmon SNPs we were unable to differentiate Clearwater River populations from populations originating from the Grande Ronde and Imnaha river basins and Asotin Creek. However, as discussed in Section 2, we anticipate incorporating an additional GAPS-standardized panel of SNPs in the EFGL and CRITFC genetics laboratories in year two of this project. We expect that the addition of these SNPs for GSI analyses will increase our resolution and generally increase the accuracy of the baseline. Ideally, we would like to discern the Lower Snake and Grande Ronde/Imnaha MPGs from populations originating in the Clearwater River drainage to assist in the estimation of VSP parameters for Snake River spring/summer Chinook. Additionally, it will be important to incorporate the GAPS-standardized panel into IDFG and CRITFC project so that Snake River spring/summer Chinook salmon will be identifiable in GSI projects outside of the Columbia basin and on the high-seas.

Interpretation of individual assignment analyses conducted on unknown mixtures from Lower Granite Dam requires thorough examination of results from leave-one-out tests (Tables 7-8). In leave-one-out tests conducted for steelhead baseline v1.0, 32 of the 49 baseline populations had greater than 75% of baseline individuals assign back to the correct reporting region. The remaining 17 baseline populations had greater than 25% of individuals assign back to incorrect reporting groups, indicating that a portion of fish originating from these populations (and sampled at Lower Granite Dam) will mis-assign to other reporting regions. For instance, Table 7 shows that four of the five populations representing the Lower/Little Salmon reporting group had greater than 15% of baseline individuals that assigned back to the Upper Salmon reporting group. Alternatively, two populations from the Upper Salmon reporting group had greater than 20% of baseline individuals that assigned back to the Lower/Little Salmon reporting group. This example of mis-assignment may indicate that GSI estimates for the Upper Salmon reporting group regarding stock composition and associated biological information are biased towards Lower/Little Salmon parameters and vice-versa. However, it is important to note that for the entire Snake River steelhead baseline v1.0, 75.4% and 82.7% of baseline individuals assigned back to the correct reporting group and MPG of origin, respectively. For Chinook baseline v1.0, 73.1% of baseline individuals assigned back to the correct reporting group and MPG of origin (Clearwater was pooled with the Grande Ronde/Imnaha reporting group and thus no estimate for MPG). As we add collections/populations to both baselines and add the GAPS-standardized SNP panel to the Chinook salmon baseline during baseline refinement efforts, we anticipate that our ability to differentiate populations and reporting groups will increase and that accuracy of assignments will improve.

Mixed stock analyses (proportional assignment) are generally considered to be of higher resolution than individual assignment analyses (as was performed here) when estimating the contribution of various stocks to a mixed fishery. However, during mixed stock analyses the biological data for individual fish cannot be tracked; instead aggregate proportions of fish are assigned to reporting groups. However, for mixtures of fish at Lower Granite Dam it is desirable to use individual assignment so that biological data collected from each fish (age, sex, trap date) can be used to infer sex ratios, age structure, and run-timing information for reporting groups. When comparing results for Lower Granite Dam mixtures using both individual and proportional assignment, 17 of the 25 estimates for stock proportional assignment (data not shown) by less than 0.5% and 22 of the 25 estimates deviated by less than 1.0%. The largest deviation was 2.2% (Grande Ronde reporting group, SY2010). Given the minimal deviations observed when comparing results from the two methods for estimating stock contributions at Lower Granite Dam, we chose to use individual assignment for analyzing adult mixtures at Lower Granite Dam.

Additional results from our GSI activities at Lower Granite Dam will be reported in the annual Lower Granite Dam report submitted through BPA Projects #199005500, #198712700 and this project (#201002600). In addition to results reported here, we will report age structure and length information among reporting groups for both species. Further, stock composition estimates along with sex, age structure, run-timing, and length information will be analyzed with Lower Granite Dam escapement estimates, providing information for the estimation of VSP parameters in the Snake River.

In year two of this project, we will begin to analyze outmigrating steelhead and Chinook salmon smolts captured at the juvenile bypass facility at Lower Granite Dam. We will report results from migratory year (MY)2010 steelhead and Chinook salmon smolts (smolts outmigrating in the spring of 2010) in next year's annual report. Alongside information from GSI at the adult trap facility, information on smolts from the juvenile trapping facility will provide further information on stock-specific productivity for both species in the Snake River.

Once the SY2011 adult steelhead and Chinook salmon from Lower Granite Dam are genotyped and analyzed in the fall/winter of 2011, we will have three years of data (SY2009-2011) for adult fish migrating above Lower Granite Dam. In the fall of 2011, we anticipate preparing a peer-reviewed manuscript covering three years of GSI for steelhead at Lower Granite Dam in the context of estimating population productivity and VSP parameters.

### CONCLUSION

The genetic baselines for steelhead and spring/summer Chinook salmon presented here provide information regarding the spatial distribution of genetic variation among Snake River populations and will allow us to examine population genetic structure and gene flow across regions or locations in the basin (homing vs. straying). Further, baseline data will allow us to evaluate the relative influences of hatchery supplementation activities in the Snake River for both species. Spatial and temporal refinement of the baselines will be important to further evaluate these issues and to improve the accuracy of GSI conducted at Lower Granite Dam. Continuation of GSI efforts at Lower Granite Dam will provide valuable data for the estimation of VSP parameters for both species in the Snake River. Finally, the contribution of baseline data from the EFGL and CRITFC labs to the SPAN and GAPS consortiums will allow for Snake River steelhead and spring/summer Chinook salmon to be identified in GSI studies outside the Columbia basin and on the high-seas.

#### ACKNOWLEDGEMENTS

The authors would like to thank many individuals from many different agencies and organizations for contributing time and expertise towards implementing this project. Special thanks to Carlos Camacho, Stacey Dauwalter, Dylan Kovis, Laura Redfield, and Thea Vanderwey from the Pacific States Marine Fisheries Commission and Stephanie Harmon, Vanessa Jacobson, Amanda Matala, Lori Maxwell, Megan Moore, and Jeff Stephenson from CRITFC for laboratory contributions. Thanks to Bill Schrader, Kristin Ellsworth, Pat Kennedy, and Matt Corsi from IDFG and Darren Ogden from NOAA Fisheries for organizing sample collection at Lower Granite Dam. Thanks to Lynn Schrader for inventorying incoming samples from Lower Granite Dam. Special thanks to all IDFG, ODFW, WDFW, USFWS, and NPT staff and volunteers who have contributed to sampling of baseline collections. We also appreciate Barbara Shields' assistance in managing contract and reporting requirements. Primary funding for this project comes from the Bonneville Power Administration (Project #2010-026-00).

#### LITERATURE CITED

- Ackerman, M. W., C. Habicht, and L. W. Seeb. 2011. Single-nucleotide polymorphisms (SNPs) under diversifying selection provide increased accuracy and precision in mixed-stock analyses of sockeye salmon from the Copper River, Alaska. Transactions of the American Fisheries Society 140(3):865-881.
- Anderson, E. C., R. S. Waples, and S. T. Kalinowski. 2008. An improved method for predicting the accuracy of genetic stock identification. Canadian Journal of Fisheries and Aquatic Sciences 65(7):1475-1486.
- Antao, T., A. Lopes, R. J. Lopes, A. Beja-Pereira, and G. Luikart. 2008. LOSITAN: A workbench to detect molecular adaptation based on a F-st-outlier method. Bmc Bioinformatics 9.
- Beacham, T. D., J. R. Candy, K. J. Supernault, T. Ming, B. Deagle, A. Schulze, D. Tuck, K. H. Kaukinen, J. R. Irvine, K. M. Miller, and R. E. Withler. 2001. Evaluation and application of microsatellite and major histocompatibility complex variation for stock identification of coho salmon in British Columbia. Transactions of the American Fisheries Society 130(6):1116-1149.
- Beacham, T. D., S. Pollard, and K. D. Le. 2000. Microsatellite DNA population structure and stock identification of steelhead trout (Oncorhynchus mykiss) in the Nass and Skeena Rivers in northern British Columbia. Marine Biotechnology 2(6):587-600.
- Bernatchez, L., and C. Landry. 2003. MHC studies in nonmodel vertebrates: what have we learned about natural selection in 15 years? Journal of Evolutionary Biology 16(3):363-377.
- Blankenship, S. M., M. R. Campbell, J. E. Hess, M. E. Hess, T. W. Kassler, C. C. Kozfkay, A. P. Matala, S. R. Narum, M. M. Paquin, M. P. Small, J. J. Stephenson, K. I. Warheit, and P. Moran. 2011. Major lineages and metapopulations in Columbia River *Onchorhynchus mykiss* are structured by dynamic landscape features and environments. Transactions of the American Fisheries Society 140(3):665-684.
- Brumfield, R. T., P. Beerli, D. A. Nickerson, and S. V. Edwards. 2003. The utility of single nucleotide polymorphisms in inferences of population history. Trends in Ecology and Evolution 18(5):249-256.
- Busby, P. J., T. C. Wainwright, G. J. Bryant, L. J. Lierheimer, R. S. Waples, F. W. Waknitz, and I. L. Lagomarsino. 1996. Status review of west coast steelhead from Washington, Idaho, Oregon, and California. National Marine Fisheries Technical Memorandum NMFS-NWFSC-27. Seattle.
- Dionne, M., K. M. Miller, J. J. Dodson, and L. Bernatchez. 2009. MHC standing genetic variation and pathogen resistance in wild Atlantic salmon. Philosophical Transactions of the Royal Society B-Biological Sciences 364(1523):1555-1565.
- Felsenstein, J. 1993. PHYLIP (Phylogeny Inference Package), version 3.5c. Department of Genome Sciences, University of Washington, Seattle.

- Good, T. P., R. S. Waples, and P. B. Adams. 2005. Updated status of federally listed ESUs of West Coast salmon and steelhead. U.S. Department of Commerce, NOAA Technical Memorandum, NMFS-NWFSC-66. 598 pp.
- Habicht, C., L. W. Seeb, K. W. Myers, E. V. Farley, and J. E. Seeb. 2010. Summer-fall distribution of stocks of immature sockeye salmon in the Bering Sea as revealed by single-nucleotide polymorphisms. Transactions of the American Fisheries Society 139(4):1171-1191.
- Harmon, J. R. 2003. A trap for handling adult anadromous salmonids at Lower Granite Dam on the Snake River, Washington. North American Journal of Fisheries Management 23(3):989-992.
- Hess, J., N. Campbell, A. Matala, and S. Narum. 2011a. 2010 Annual Report: Genetic Assessment of Columbia River Stocks. U.S. Dept. of Energy Bonneville Power Administration Report Project #2008-907-00.
- Hess, J. E., A. P. Matala, and S. R. Narum. 2011b. Comparison of SNP and microsatellites markers for application of genetic stock identification for Chinook salmon in the Columbia River Basin. Molecular Ecology Resources 11(Suppl. 1):137-149.
- Holm, S. 1979. A simple sequentially rejective multiple test procedure. Scandinavian Journal of Statistics 6(2):65-70.
- ICTRT (Interior Columbia Trechnical Recovery Team). 2003. Independent Populations of Chinook, steelhead and sockeye for listed Interior Columbia Basin ESUs. Interior Columbia Basin Technical Recovery Team Report. July 2003.
- Kalinowski, S. T., K. R. Manlove, and M. L. Taper. 2007. ONCOR: software for genetic stock identification. Montana State University, Bozeman. Available at: www.montanta.edu/kalinowski/Software/ONCOR.htm.
- Lahood, E. S., J. J. Miller, C. Apland, and M. J. Ford. 2008. A rapid, ethanol-free fish tissue collection method for molecular genetic analyses. Transactions of the American Fisheries Society 137(4):1104-1107.
- Matala, A. P., J. E. Hess, and S. R. Narum. 2011. Resolving adaptive and demographic divergence among Chinook salmon populations in the Columbia River Basin. Transactions of the American Fisheries Society. DOI: 10.1080/00028487.2011.588092.
- McElhany, P., M. H. Ruckelshaus, M. J. Ford, T. C. Wainwright, and E. P. Bjorkstedt. 2000. Viable salmonid populations and the recovery of evolutionary significant units. U.S. Department of Commerce, NOAA Technical Memo. NMFS-NWFSC-42, 156 p.
- Miller, K. M., K. H. Kaukinen, T. D. Beacham, and R. E. Withler. 2001. Geographic heterogeneity in natural selection on an MHC locus in sockeye salmon. Genetica 111(1-3):237-257.
- Moran, P. 2003. Genetic structure of Oncorhynchus mykiss populations in the Grande Ronde River, Imnaha River, and adjacent regions of the Snake River basin. Final report submitted to the U.S. Fish and Wildlife Service, Lower Snake River Compensation Plan

Office, Boise, Idaho, in partial fulfillment of Contract No. 14110-1-H070. 28p. + Appendices.

- Morin, P. A., G. Luikart, R. K. Wayne, and S. N. P. W. Grp. 2004. SNPs in ecology, evolution and conservation. Trends in Ecology and Evolution 19(4):208-216.
- Narum, S., N. Campbell, A. Matala, and J. Hess. 2010a. 2009 Annual Report: Genetic Assessment of Columbia River Stocks. U.S. Department of Energy Bonneville Power Administration Report Project #2008-907-00.
- Narum, S. R., M. Banks, T. D. Beacham, M. R. Bellinger, M. R. Campbell, J. Dekoning, A. Elz, C. M. Guthrie, C. Kozfkay, K. M. Miller, P. Moran, R. Phillips, L. W. Seeb, C. T. Smith, K. Warheit, S. F. Young, and J. C. Garza. 2008a. Differentiating salmon populations at broad and fine geographical scales with microsatellites and single nucleotide polymorphisms. Molecular Ecology 17(15):3464-3477.
- Narum, S. R., D. Hatch, A. J. Talbot, P. Moran, and M. S. Powell. 2008b. Iteroparity in complex mating systems of steelhead Oncorhynchus mykiss (Walbaum). Journal of Fish Biology 72(1):45-60.
- Narum, S. R., and J. E. Hess. 2011. Comparison of F-ST outlier tests for SNP loci under selection. Molecular Ecology Resources 11:184-194.
- Narum, S. R., J. E. Hess, and A. P. Matala. 2010b. Examining genetic lineages of Chinook salmon in the Columbia River basin. Transactions of the American Fisheries Society 139(5):1465-1477.
- Narum, S. R., J. J. Stephenson, and M. R. Campbell. 2007. Genetic variation and structure of Chinook salmon life history types in the Snake River. Transactions of the American Fisheries Society 136(5):1252-1262.
- Narum, S. R., J. S. Zendt, C. Frederiksen, N. Campbell, A. Matala, and W. R. Sharp. 2011. Candidate genetic markers associated with anadromy in *Oncorhynshus mykiss* of the Klickitat River. Transactions of the American Fisheries Society 140(3):843-854.
- Nei, M. 1972. Genetic Distance Between Populations. American Naturalist 106(949):283-&.
- Nielsen, J. L., A. Byrne, S. L. Graziano, and C. C. Kozfkay. 2009. Steelhead Genetic Diversity at Multiple Spatial Scales in a Managed Basin: Snake River, Idaho. North American Journal of Fisheries Management 29(3):680-701.
- NMFS (National Marine Fisheries Service). 2007. Biological Opinion Remand Draft. Consultation on Remand for Operation of the Federal Columbia River Power System, 11 Bureau of Reclamation Projects in the Columbia Basin and ESA Section 10(a)(1)(A) Permit for Juvenile Fish Transportation Program (Revised and reissued pursuant to court order, NWF v. NMFS, Civ. No. CV 01-640-RE (D. Oregon)). National Marine Fisheries Service (NOAA Fisheries) - Northwest Region, Seattle. October 2007. .
- Ogden, D. in prep. Operation of the Lower Granite Dam adult trap, 2009-10. BPA project #2005-002-00.

- Page, R. D. M. 1996. TreeView: An application to display phylogenetic trees on personal computers. Computer Applications in the Biosciences 12(4):357-358.
- Peakall, R., and P. E. Smouse. 2006. GENALEX 6: genetic analysis in Excel. Population genetic software for teaching and research. Molecular Ecology Notes 6(1):288-295.
- Rannala, B., and J. L. Mountain. 1997. Detecting immigration by using multilocus genotypes. Proceedings of the National Academy of Sciences of the United States of America 94(17):9197-9201.
- Robards, M. D., and T. P. Quinn. 2002. The migratory timing of adult summer-run steelhead in the Columbia River over six decades of environmental change. Transactions of the American Fisheries Society 131(3):523-536.
- Rousset, F. 2008. GENEPOP ' 007: a complete re-implementation of the GENEPOP software for Windows and Linux. Molecular Ecology Resources 8(1):103-106.
- Schlotterer, C. 2004. The evolution of molecular markers just a matter of fashion? Nature Reviews Genetics 5(1):63-69.
- Seeb, J. E., C. E. Pascal, R. Ramakrishnan, and L. W. Seeb. 2009a. SNP genotyping by the 5'nuclease reaction: advances in high throughput genotyping with non-model organisms. Pages 277-292 *in* A. Komar, editor. Methods in Molecular Biology, Single Nucleotide Polymorphisms, 2nd edition. Humana Press.
- Seeb, L. W., A. Antonovich, A. A. Banks, T. D. Beacham, A. R. Bellinger, S. M. Blankenship, A. R. Campbell, N. A. Decovich, J. C. Garza, C. M. Guthrie, T. A. Lundrigan, P. Moran, S. R. Narum, J. J. Stephenson, K. J. Supernault, D. J. Teel, W. D. Templin, J. K. Wenburg, S. E. Young, and C. T. Smith. 2007. Development of a standardized DNA database for Chinook salmon. Fisheries 32(11):540-552.
- Seeb, L. W., N. A. DeCovich, A. W. Barclay, C. T. Smith, and W. D. Templin. 2009b. Timing and origin of Chinook salmon stocks in the Copper River and adjacent coastal fisheries using DNA markers. Annual report for study 04-207. UWFWS Office of Subsistence Management, Fisheries Resource Monitoring Program. Alaska Department of Fish and Game, Fishery Data Series No. 09-58, Anchorage.
- Shaklee, J. B., T. D. Beacham, L. Seeb, and B. A. White. 1999. Managing fisheries using genetic data: case studies from four species of Pacific salmon. Fisheries Research 43(1-3):45-78.
- Smith, C. T. 2007. Feasibility of genetic stock ID of Chinook salmon sampled at Lower Granite Dam. U. S. Fish and Wildlife Service, Abernathy Fish Technology Center Report.
- Smith, C. T., W. D. Templin, J. E. Seeb, and L. W. Seeb. 2005. Single nucleotide polymorphisms provide rapid and accurate estimates of the proportions of US and Canadian Chinook salmon caught in Yukon River fisheries. North American Journal of Fisheries Management 25(3):944-953.
- Thurow, R. F. 1985. Middle Fork Salmon River Fisheries Investigations. Job Completion Report, Project F-73-R-6.

- Thurow, R. F. 2000. Dynamics of Chinook salmon populations within Idaho's Frank Church Wilderness implications for persistence *in* McCool, S.F., and others, v3. Wilderness as a place for scientific inquiry: Proceedings of the wilderness science in a time of change conference, Missoula, Montana, May 23-27, 1999, U.S. Forest Service RMRS-P-15-VOL-3, p. 143-151.
- Van Tassell, C. P., T. P. L. Smith, L. K. Matukumalli, J. F. Taylor, R. D. Schnabel, C. T. Lawley, C. D. Haudenschild, S. S. Moore, W. C. Warren, and T. S. Sonstegard. 2008. SNP discovery and allele frequency estimation by deep sequencing of reduced representation libraries. Nature Methods 5(3):247-252.
- Waples, R. S. 1990. Temporal changes of allele frequency in Pacific salmon implications for mixed-stock fishery analysis. Canadian Journal of Fisheries and Aquatic Sciences 47(5):968-976.
- Weir, B. S., and C. C. Cockerham. 1984. Estimating F-statistics for the analysis of population structure. Evolution 38(6):1358-1370.

TABLES

Table 1. Oncorhynchus mykiss populations screened for 192 assays for Snake River baseline v1.0. Genotyping agency, major population group (MPG), sample size (*n*), latitude, longitude, years sampled, expected (H<sub>E</sub>) and observed (H<sub>O</sub>) heterozygosity, population-specific F<sub>ST</sub>, and number of loci with HWE deviations are shown. Map # corresponds to numbers in FIGURE 1. Agency indicates the laboratory where samples were genotyped. All populations are summerrun, of natural origin, and presumed to be of anadromous lineage.

Map #	Collection Name	Agency	MPG	n	Latitude	Longitude	Year	HE	Ho	Fst	HWE
1	Sawtooth Weir	IDFG	Salmon	67	44.1506	-114.8851	2005, 2010	0.292	0.291	0.020	
2	West Fork Yankee Fork Salmon River	IDFG	Salmon	47	44.3514	-114.7297	2004	0.298	0.304	0.020	
3	Morgan Creek	IDFG	Salmon	45	44.6135	-114.1641	2000	0.315	0.317	0.024	(10)
4	Pahsimeroi River Weir	IDFG	Salmon	47	44.6823	-114.0396	2006	0.308	0.312	0.023	
5	North Fork Salmon River	IDFG	Salmon	52	45.4094	-113.9918	2010	0.304	0.301	0.017	
6	Marsh Creek	IDFG	Salmon	59	44.4493	-115.2301	2000	0.283	0.286	0.032	
7	Rapid River (MF)	IDFG	Salmon	45	44.6790	-115.1490	2000	0.288	0.286	0.031	(11)
8	Pistol Creek	IDFG	Salmon	23	44.7218	-115.1489	2000	0.280	0.295	0.035	
9	Camas Creek	IDFG	Salmon	58	44.8918	-114.7222	2000	0.285	0.280	0.025	
10	Big Creek - Upper	IDFG	Salmon	47	45.1506	-115.2967	2000	0.279	0.294	0.033	(11)
11	Big Creek - Lower	CRITFC	Salmon	48	45.0925	-114.7297	2000	0.288	0.284	0.028	(10)
12	Loon Creek	CRITFC	Salmon	40	44.5976	-114.8123	1999	0.283	0.292	0.027	
13	Bargamin Creek	IDFG	Salmon	46	45.5716	-115.1919	2000	0.305	0.301	0.022	
14	East Fork South Fork Salmon River	IDFG	Salmon	47	45.0127	-115.7129	2000	0.291	0.290	0.029	(12)
15	Secesh River	IDFG	Salmon	46	45.0268	-115.7082	2000	0.285	0.288	0.027	
16	Stolle Meadows	CRITFC	Salmon	45	44.6070	-115.6810	2000	0.282	0.280	0.029	
17	Boulder Creek	IDFG	Salmon	47	45.2019	-116.3114	2000	0.301	0.288	0.020	(13)
18	Hazard Creek	IDFG	Salmon	45	45.1836	-116.2995	2000	0.307	0.308	0.017	(12)
19	Rapid River	IDFG	Salmon	47	45.3719	-116.3556	2003	0.289	0.293	0.024	
20	Slate Creek	IDFG	Salmon	47	45.6380	-116.2828	2000	0.299	0.298	0.018	(10)
21	Whitebird Creek	IDFG	Salmon	59	45.7523	-116.3198	2001	0.292	0.289	0.018	
22	Storm Creek	IDFG	Clearwater	38	46.4607	-114.5467	2000	0.271	0.276	0.034	
23	Crooked Fork Lochsa River	IDFG	Clearwater	47	46.5251	-114.6787	2000	0.278	0.278	0.028	
24	Canyon Creek	IDFG	Clearwater	47	46.2161	-115.5559	2004	0.276	0.281	0.026	
25	Bear Creek	IDFG	Clearwater	46	46.0191	-114.8378	2000	0.284	0.290	0.032	
26	North Fork Moose Creek	IDFG	Clearwater	47	46.1673	-114.8998	2004	0.278	0.281	0.027	
27	Gedney Creek	IDFG	Clearwater	45	46.0583	-115.3141	2000	0.286	0.286	0.025	(10)
28	O'Hara Creek	IDFG	Clearwater	47	46.0810	-115.5179	2000	0.283	0.285	0.023	
29	Crooked River	IDFG	Clearwater	84	45.8211	-115.5272	2007	0.273	0.270	0.027	(12)
30	Tenmile Creek	IDFG	Clearwater	47	45.8057	-115.6833	2000	0.275	0.284	0.034	
31	John's Creek	IDFG	Clearwater	40	45.8224	-115.8887	2000	0.286	0.278	0.025	
32	Clear Creek	IDFG	Clearwater	45	46.0486	-115.7814	2000	0.280	0.272	0.027	
33	East Fork Potlatch River	IDFG	Clearwater	62	46.7985	-116.4235	2008	0.297	0.296	0.020	
34	Big Bear Creek	IDFG	Clearwater	33	46.6336	-116.6545	2007, 2008	0.305	0.312	0.020	
35	Little Bear Creek	IDFG	Clearwater	54	46.6291	-116.6612	2007, 2008	0.295	0.302	0.019	(10)
36	Mission Creek	IDFG	Clearwater	51	46.3653	-116.7354	2000	0.295	0.294	0.020	

## Table 1. Continued.

Map #	Collection Name	Agency	MPG	n	Latitude	Longitude	Year	HE	Ho	Fst	HWE
37	Big Sheep Creek	CRITFC	Imnaha	71	45.5574	-116.8345	2001	0.289	0.289	0.019	(10)
38	Camp Creek	CRITFC	Imnaha	25	45.5572	-116.8352	2001	0.287	0.295	0.027	
39	Cow Creek	CRITFC	Imnaha	44	45.7681	-116.7496	2000	0.293	0.294	0.019	
40	Lightning Creek	CRITFC	Imnaha	46	45.6554	-116.7265	2000	0.286	0.284	0.020	
41	Crooked Creek	CRITFC	Grande Ronde	98	45.9770	-117.5550	2001	0.304	0.293	0.018	(13)
42	Elk Creek	CRITFC	Grande Ronde	47	45.7053	-117.1529	2000	0.282	0.286	0.027	
43	Little Minam River	CRITFC	Grande Ronde	48	45.7255	-117.7854	2000	0.294	0.294	0.025	
44	Lostine River	CRITFC	Grande Ronde	45	45.5521	-117.4898	2000	0.303	0.300	0.025	
45	Menatchee Creek	CRITFC	Grande Ronde	73	46.0075	-117.3651	1999	0.312	0.314	0.019	(10)
46	Wenaha River	CRITFC	Grande Ronde	94	45.9453	-117.4513	2001	0.297	0.287	0.018	(12)
47	Asotin Creek	CRITFC	Lower Snake	49	46.3442	-117.0551	2000	0.306	0.307	0.017	
48	Tucannon River	CRITFC	Lower Snake	45	46.2046	-117.7060	1991	0.297	0.305	0.018	
49	Touchet River	CRITFC	-	89	46.0340	-118.6836	1995	0.293	0.289	0.020	

Table 2. Oncorhynchus tshawytscha populations screened for 96 assays for Snake River baseline v1.0. Genotyping agency, major population group (MPG), sample size (*n*), latitude, longitude, years sampled, expected (H<sub>E</sub>) and observed (H<sub>O</sub>) heterozygosity, population-specific F<sub>ST</sub>, and number of loci with HWE deviations are shown. Map # corresponds to numbers in FIGURE 2. Agency indicates the laboratory where samples were genotyped. All populations are spring/summer run and of natural origin.

Map #	Collection Name	Agency	MPG	n	Latitude	Longitude	Year	HE	Ho	Fst	HWE
1	Sawtooth Weir	IDFG	Upper Salmon	93	44.151	-114.885	2009, 2010	0.219	0.213	0.016	
2	West Fork Yankee Fork Salmon River	CRITFC	Upper Salmon	75	44.349	-114.727	2005	0.222	0.218	0.021	(5)
3	East Fork Salmon River	IDFG	Upper Salmon	45	44.115	-114.430	2010	0.223	0.230	0.019	
4	Pahsimeroi River Weir	IDFG	Upper Salmon	75	44.682	-114.039	2009, 2010	0.226	0.213	0.019	
5	Hayden Creek (Lemhi)	IDFG	Upper Salmon	75	44.862	-113.632	2009	0.243	0.243	0.025	(6)
6	L3A Trap (Lemhi)	IDFG	Upper Salmon	90	45.153	-113.814	2009, 2010	0.245	0.236	0.017	(5)
7	Lower Lemhi Trap	IDFG	Upper Salmon	82	44.869	-113.625	2009, 2010	0.212	0.220	0.021	(5)
8	Marsh Creek	IDFG	Middle Fork Salmon	71	44.381	-115.153	2007-2010	0.205	0.202	0.017	
9	Capehorn Creek	CRITFC	Middle Fork Salmon	88	44.388	-115.174	2005	0.210	0.205	0.022	
10	Elk Creek	IDFG	Middle Fork Salmon	79	44.442	-115.454	2007-2010	0.208	0.191	0.020	(7)
11	Bear Valley Creek	IDFG	Middle Fork Salmon	93	44.427	-115.328	2007-2010	0.210	0.201	0.017	(5)
12	Sulphur Creek	IDFG	Middle Fork Salmon	29	44.543	-115.329	2008, 2010	0.193	0.187	0.023	
13	Camas Creek	CRITFC	Middle Fork Salmon	47	44.892	-114.721	2006	0.209	0.215	0.025	
14	Big Creek	CRITFC	Middle Fork Salmon	92	45.138	-115.038	2001	0.219	0.219	0.018	(8)
15	Chamberlain Creek	CRITFC	Middle Fork Salmon	45	45.454	-114.933	2009	0.192	0.198	0.034	
16	Lake Creek (Secesh)	IDFG	South Fork Salmon	57	45.279	-115.922	2007-2010	0.213	0.212	0.022	
17	Secesh River (Lower)	CRITFC	South Fork Salmon	81	45.033	-115.722	2001	0.219	0.217	0.019	
18	Secesh River (Upper)	IDFG	South Fork Salmon	58	45.217	-115.808	2007-2010	0.216	0.208	0.018	
19	Johnson Creek	CRITFC	South Fork Salmon	92	44.899	-115.492	2002	0.222	0.222	0.017	
20	South Fork Salmon River Weir	IDFG	South Fork Salmon	93	44.667	-115.703	2010	0.228	0.224	0.013	
21	Rapid River Weir	IDFG	South Fork Salmon	93	45.372	-116.356	2006	0.236	0.235	0.017	(6)
22	Powell Weir	IDFG	NA	34	46.506	-114.687	2009	0.239	0.236	0.017	
23	Red River Weir	IDFG	NA	58	45.710	-115.344	2009, 2010	0.252	0.243	0.016	
24	Crooked River Weir	IDFG	NA	67	45.817	-115.527	2009, 2010	0.249	0.245	0.015	
25	Newsome Creek	CRITFC	NA	90	45.831	-115.608	2001	0.240	0.236	0.017	
26	Lolo Creek	CRITFC	NA	89	46.279	-115.775	2001	0.241	0.240	0.013	
27	Imnaha River	CRITFC	Grande Ronde / Imnaha	92	45.561	-116.834	1998	0.240	0.243	0.015	
28	Catherine Creek	CRITFC	Grande Ronde / Imnaha	85	45.158	-117.779	2003	0.250	0.247	0.016	
29	Lostine River Weir	CRITFC	Grande Ronde / Imnaha	109	45.535	-117.451	2009	0.244	0.246	0.018	
30	Minam River	CRITFC	Grande Ronde / Imnaha	82	45.600	-117.729	2002	0.260	0.254	0.017	
31	Wenaha River	CRITFC	Grande Ronde / Imnaha	44	45.956	-117.728	2002	0.256	0.260	0.016	
32	Tucannon River	CRITFC	Lower Snake	87	46.526	-118.142	2003	0.270	0.280	0.027	(7)

Table 3. Summary of 188 SNP markers (Appendix A and Hess et al. 2011b) genotyped across 49 steelhead populations from throughout the Snake River. SNPs designated as PBT are used for both the PBT project (BPA Project #2010-031-00) and GSI projects. SNPs designated as GSI are used primarily for GSI projects. Summary statistics include minor-allele frequency (MAF) range, expected (H<sub>E</sub>) and observed (H<sub>O</sub>) heterozygosity, Weir and Cockerham (1984)  $F_{ST}$ , and fixation index ( $F_{IS}$ ). "HWE" designates the number of populations that a SNP deviated from Hardy-Weinberg expectation for any SNP that deviated in greater than 10% of populations. "LD" is the number of populations that a pair of SNP loci exhibited linkage disequilibrium if it occurred in more than half of the populations. "CS" indicates a locus that was designated as a candidate for divergent (+) or balancing (-) selection.

SNP	Panel	MAF Range	HE	Ho	F <sub>ST</sub>	<b>F</b> IS	HWE	LD	CS
M09AAD.076	PBT	.267722	0.482	0.478	0.023	0.012			
M09AAJ.163	PBT	.043543	0.410	0.424	0.033	-0.016			
M09AAE.082	PBT	.083706	0.345	0.339	0.062	0.030			+
OMS00002	PBT	.220533	0.455	0.460	0.013	-0.001			-
OMS00006	PBT	.224628	0.483	0.495	0.020	-0.019			
OMS00024	PBT	.167793	0.445	0.439	0.060	0.017			+
OMS00039	PBT	.337678	0.487	0.498	0.014	-0.006			
OMS00053	PBT	.213678	0.482	0.492	0.022	-0.006			
OMS00057	PBT	.156564	0.441	0.461	0.043	-0.047			
OMS00058	PBT	.118725	0.457	0.484	0.058	-0.040			
OMS00062	PBT	.128777	0.368	0.390	0.061	-0.043			
OMS00064	PBT	.098600	0.443	0.434	0.066	0.036			
OMS00068	PBT	.060565	0.404	0.405	0.046	0.010			
OMS00070	PBT	.213700	0.464	0.469	0.060	0.013			
OMS00071	PBT	.255737	0.473	0.477	0.037	0.007			
OMS00072	PBT	.298656	0.481	0.479	0.018	0.015	(6)		
OMS00074	PBT	.065740	0.447	0.453	0.070	0.008	(9)		+
OMS00077	PBT	.227576	0.470	0.489	0.021	-0.024			
OMS00078	PBT	.156521	0.379	0.376	0.021	0.012			
OMS00079	PBT	.297700	0.484	0.486	0.022	0.011			
OMS00111	PBT	.042532	0.301	0.305	0.070	-0.005			
OMS00089	PBT	.056500	0.378	0.385	0.030	0.009			
OMS00090	PBT	.200656	0.470	0.471	0.041	0.011			
OMS00101	PBT	.122745	0.455	0.447	0.050	0.034	(5)		
OMS00105	PBT	.128580	0.441	0.450	0.039	-0.007			
OMS00106	PBT	.056435	0.344	0.350	0.035	-0.017			
OMS00154	PBT	.089422	0.315	0.325	0.021	-0.026			
OMS00112	PBT	.000456	0.281	0.293	0.045	-0.014			
OMS00118	PBT	.114691	0.434	0.425	0.067	0.036			+
OMS00120	PBT	.000474	0.260	0.248	0.062	0.063	(5)		+
OMS00121	PBT	.272670	0.481	0.485	0.019	0.007			
OMS00132	PBT	.181590	0.465	0.479	0.024	-0.003			
OMS00175	PBT	.240660	0.472	0.483	0.024	-0.012			
OMS00179	PBT	.132488	0.388	0.354	0.027	0.103	(6)		
OMS00180	PBT	.189543	0.433	0.440	0.025	-0.001			
Omy_101832-195	PBT	.120734	0.462	0.482	0.037	-0.026			
Omy_101993-189	PBT	.053605	0.320	0.310	0.055	0.032			
Omy_102505-102	PBT	.147596	0.452	0.465	0.029	-0.021			
Omy_104519-624	PBT	.053578	0.406	0.425	0.066	-0.029			+
Omy_105105-448	PBT	.202674	0.465	0.468	0.041	-0.004			

Table 3. Continued.									
SNP	Panel	MAF Range	Η <sub>E</sub>	Ho	F <sub>ST</sub>	<b>F</b> <sub>IS</sub>	HWE	LD	CS
Omy_105385-406	PBT	.211652	0.463	0.466	0.023	0.010			
Omy_105714-265	PBT	.100500	0.428	0.460	0.023	-0.058			
Omy_107806-34	PBT	.070656	0.400	0.392	0.089	0.028			+
Omy_108007-193	PBT	.092685	0.444	0.445	0.060	0.008			+
Omy_109243-222	PBT	.000351	0.255	0.256	0.033	0.015			
Omy_109894-185	PBT	.100611	0.443	0.426	0.034	0.050	(5)		
Omy_110064-419	PBT	.085755	0.431	0.438	0.063	0.006			+
Omy_111383-51	PBT	.189604	0.466	0.473	0.032	-0.004			
Omy_113490-159	PBT	.160819	0.441	0.433	0.072	0.034			+
Omy_114315-438	PBT	.086679	0.429	0.398	0.088	0.083	(6)		+
Omy_114587-480	PBT	.085521	0.428	0.441	0.033	-0.017			
Omy_129870-756	PBT	.043367	0.273	0.276	0.025	0.021			
Omy_116733-349	PBT	.111600	0.403	0.416	0.031	-0.019			
Omy_128923-433	PBT	.271803	0.469	0.457	0.049	0.037			
Omy_130524-160	PBT	.256633	0.463	0.467	0.020	0.004			
Omy_97660-230	PBT	.130560	0.430	0.441	0.032	0.000			
Omy_99300-202	PBI	.096489	0.345	0.331	0.036	0.060			
Omy_aldB-165	PBI	.074427	0.407	0.402	0.016	0.032			
Omy_anp-17	PBI	.026761	0.400	0.406	0.109	-0.013			+
Omy_arp-630	PBI	.156656	0.475	0.481	0.033	0.003			
Omy_b1-266	PBI	.141398	0.391	0.390	0.013	0.014			-
Omy_BAC-B4-324	PBI	.271602	0.478	0.474	0.022	0.029			
Omy_ada10-71	PBI	.040359	0.288	0.289	0.022	0.009			
Omy_reda1-410	PBI	.078574	0.315	0.318	0.034	0.002			
Omy_ca59-206		.149521	0.403	0.406	0.022	0.006			
		.091447	0.399	0.416	0.020	-0.031			
$Omy_cox 1-221$		.1/858/	0.458	0.473	0.034	-0.016	$\langle 0 \rangle$		
Omy a12 82		.197008	0.463	0.425	0.043	0.081	(9)		
$Omy_g 12-62$		.201724	0.479	0.409	0.031	-0.015			
$Omy_{bc}715_{80}$		.211717	0.470	0.492	0.025	-0.011			_
$Omy_hsf2_1/6$	DRT	.223342	0.437	0.452	0.010	0.000			-
Omy    17-185	PBT	244 - 656	0.425	0.407	0.002	-0.002	(7)		т
$Omy \parallel 1b 028$	PRT	034 - 319	0.470	0.300	0.023	-0.030	(r)		
Omy_1115_198	PRT	144 - 698	0.202	0.200	0.002	-0.004			
Omy II 6-320	PRT	066 - 385	0.400	0.400	0.040	-0.010			
Omv metA-161	PBT	096 - 489	0.368	0.371	0.021	0.006			
Omv NaKATPa3-50	PBT	.053511	0.403	0.413	0.033	-0.009			
Omv txnip-343	PBT	.078539	0.351	0.359	0.027	-0.005			
Omy nkef-241	PBT	.223630	0.463	0.470	0.028	0.000			
Omy_ntl-27	PBT	.133570	0.429	0.433	0.042	-0.008			
Omy_Ogo4-212	PBT	.104568	0.457	0.458	0.035	0.012	(6)		
Omy bcAKala-380rd	PBT	.089563	0.416	0.407	0.039	0.043	(6)		
Omy_Ots249-227	PBT	.136478	0.397	0.396	0.025	0.005	( )		
Omy_oxct-85	PBT	.000287	0.182	0.181	0.039	0.033			
Omy_p53-262	PBT	.033522	0.325	0.327	0.044	0.014			
Omy_rapd-167	PBT	.043372	0.297	0.305	0.029	-0.010			
Omy_rbm4b-203	PBT	.011439	0.299	0.301	0.049	-0.003			
Omy_srp09-37	PBT	.122560	0.407	0.414	0.037	-0.006			
Omy_stat3-273	PBT	.074433	0.342	0.347	0.025	-0.005			
Omy_u09-53.469	PBT	.189826	0.435	0.439	0.096	-0.013			+
Omy_u09-54-311	PBT	.078589	0.400	0.399	0.044	0.011			
Omy_U11_2b-154	PBT	.074375	0.327	0.330	0.035	0.004			
Omy_vatf-406	PBT	.078620	0.417	0.417	0.070	0.007			+
SNP	Panel	MAF Range	H⊧	Ho	Fer	Fie	HWE	LD	CS
----------------------	-------	-----------	-------	-------	-------	--------	------	----	----
OMY1011SNP	PBT	106 - 438	0.367	0.366	0.028	0.010			
M09AAC 055	GSI	000 - 266	0.007	0.000	0.020	-0.017			
OMGH1PROM1-SNP1	GSI	000 - 364	0 169	0 168	0.086	0.014			+
OMS00003	GSI	.033345	0.243	0.246	0.027	-0.004			
OMS00008	GSI	.000415	0.270	0.267	0.046	0.030			
OMS00013	GSI	.000185	0.123	0.118	0.028	0.058			
OMS00014	GSI	.000111	0.026	0.024	0.027	0.092			
OMS00015	GSI	.000198	0.116	0.119	0.041	-0.011			
OMS00017	GSI	.100733	0.386	0.382	0.074	0.017			+
OMS00018	GSI	.026287	0.182	0.191	0.021	-0.033			
Omy cd28-130	GSI	.000061	0.022	0.021	0.010	0.049			
OM\$00030	GSI	.000193	0.140	0.135	0.023	0.043			
OMS00048	GSI	.011239	0.194	0.198	0.024	-0.006			
OMS00052	GSI	.051378	0.285	0.284	0.020	0.007			
OMS00056	GSI	.042411	0.339	0.342	0.021	0.011			
OMS00061	GSI	.000239	0.111	0.104	0.032	0.056			
OMS00092	GSI	.020522	0.255	0.261	0.062	-0.017			
OMS00096	GSI	.043377	0.293	0.292	0.034	0.018			
OMS00087	GSI	.021447	0.290	0.233	0.049	0.210	(12)		
OMS00119	GSI	.000288	0.222	0.228	0.035	-0.020			
OMS00129	GSI	.011435	0.278	0.248	0.043	0.125	(8)		
OMS00133	GSI	.000200	0.052	0.049	0.042	0.081			
OMS00138	GSI	.016391	0.202	0.213	0.053	-0.042			
OMS00149	GSI	.000144	0.075	0.075	0.024	0.003			
OMS00151	GSI	.053424	0.303	0.297	0.029	0.027			
OMS00095	GSI	.000191	0.103	0.102	0.027	0.016			
OMS00169	GSI	.000135	0.019	0.020	0.044	-0.056			
OMS00173	GSI	.022313	0.196	0.199	0.030	-0.004			
OMS00176	GSI	.000351	0.107	0.111	0.044	-0.035			
Omy_impa1-55	GSI	.000278	0.160	0.160	0.032	0.016			
Omy_103705-558	GSI	.015283	0.176	0.175	0.022	0.024			
Omy_105075-162	GSI	.000233	0.159	0.158	0.024	0.022			
Omy_107031-704	GSI	.041391	0.267	0.271	0.035	-0.004			
Omy_10/285-69	GSI	.025320	0.263	0.272	0.023	-0.009			
Omy_110201-359	651	.000240	0.178	0.178	0.028	0.009			
OMS00114	GSI	.000186	0.092	0.093	0.026	-0.011			
OMS00114 OMS00142	GSI	.000216	0.100	0.109	0.010	0.008			
OMS00143	CSI	.000277	0.177	0.101	0.030	-0.007			
Omy 07077-73	63	.000174	0.000	0.009	0.007	-0.035			-
$Omy_{07865-106}$	GSI	000 - 116	0.050	0.040	0.000	0.019			
Omy 97954-618	GSI	030 - 110	0.007	0.002	0.022	0.104			
Omy 128996-481	GSI	000 - 266	0.203	0.200	0.043	0.042	(6)		
Omy aromat-280	GSI	076 - 483	0.123	0.112	0.004	0.100	(6)		
Omy aspAT-123	GSI	140 - 447	0.200	0.207	0.000	-0.021	(0)		
$Omy_{h}=164$	GSI	000 - 341	0.000	0.400	0.024	0.079	(7)		+
Omv BAC-E5 284	GSI	000 - 174	0.094	0.092	0.027	0.024	(•)		•
Omv BAMBI2.312	GSI	000 - 320	0 193	0.189	0.049	0.021			
Omv carban1-264	GSI	000 - 355	0 202	0 200	0.051	0.022			
Omy cd59b-112	GSI	.000370	0.177	0.166	0.052	0.057			
Omy_cin-172	GSI	.030588	0.318	0.333	0.050	-0.032			
Omy_cox2-335	GSI	.030362	0.242	0.245	0.041	-0.007			
Omy_e1-147	GSI	.000174	0.087	0.091	0.025	-0.020			
Omy_g1-103	GSI	.000157	0.102	0.105	0.033	-0.016			

Table 3. Continued.									
SNP	Panel	MAF Range	HE	Ho	F <sub>ST</sub>	<b>F</b> <sub>IS</sub>	HWE	LD	CS
Omy_G3PD_2-371	GSI	.073521	0.283	0.276	0.036	0.036			
Omy_gadd45-332	GSI	.000478	0.217	0.220	0.081	-0.003			+
Omy_gdh-271	GSI	.011304	0.192	0.184	0.020	0.049			
Omy_gh-475	GSI	.043298	0.234	0.239	0.021	-0.004			
Omy_GHSR-121	GSI	.000202	0.079	0.082	0.053	-0.028		(26) <sup>a</sup>	
Omy_hsp47-86	GSI	.106391	0.333	0.330	0.012	0.021			-
Omy_hsp70aPro-329	GSI	.000450	0.096	0.094	0.088	0.040			+
Omy_IL1b-163	GSI	.000447	0.146	0.142	0.131	0.048			+
Omy_inos-97	GSI	.000250	0.111	0.112	0.066	0.000			
Omy_LDHB-1_i2	GSI	.000207	0.147	0.140	0.019	0.050			
Omy_LDHB-2_e5	GSI	.053323	0.262	0.260	0.014	0.028			
Omy_LDHB-2_i6	GSI	.000089	0.018	0.015	0.016	0.141			
Omy_lpl-220	GSI	.056311	0.260	0.263	0.013	-0.008			-
Omy_mapK3-103	GSI	.000138	0.056	0.059	0.038	-0.033		(26) <sup>a</sup>	
Omy_mcsf-268	GSI	.000244	0.035	0.029	0.058	0.189			
Omy_metB-138	GSI	.000342	0.242	0.241	0.031	0.022			
Omy_myoD-178	GSI	.000311	0.199	0.196	0.040	0.028			
Omy_nach-200	GSI	.000043	0.014	0.014	0.003	-0.010			
Omy_nxt2-273	GSI	.000250	0.117	0.091	0.042	0.221	(11)		
Omy_OmyP9-180	GSI	.015284	0.168	0.157	0.029	0.082	(6)		
Omy_pad-196	GSI	.000133	0.072	0.066	0.015	0.076			
Omy_ppie-232	GSI	.000500	0.233	0.234	0.041	0.009			
Omy_ca050-64	GSI	.152532	0.441	0.437	0.027	0.001			
Omy_sast-264	GSI	.068865	0.290	0.294	0.113	-0.004			+
Omy_SECC22b-88	GSI	.000135	0.027	0.027	0.043	0.028			
Omy_sSOD-1	GSI	.000045	0.016	0.016	0.011	-0.019			
Omy_star-206	GSI	.000181	0.092	0.095	0.028	-0.022			
Omy_sys1-188	GSI	.000304	0.170	0.167	0.053	0.008			
Omy_tlr3-377	GSI	.000304	0.168	0.171	0.050	0.011			
Omy_tlr5-205	GSI	.000152	0.100	0.105	0.024	-0.033			
Omy_hsf1b-241	GSI	.000202	0.142	0.150	0.023	-0.041			
Omy_u07-79-166	GSI	.000255	0.140	0.140	0.054	0.015			
Omy_u09-52.284	GSI	.000121	0.051	0.052	0.023	-0.018			
Omy_hus1-52	GSI	.000193	0.087	0.086	0.046	0.025			
Omy_u09-56.119	GSI	.000207	0.156	0.159	0.023	0.002			
Omy_nips-299	GSI	.000217	0.126	0.128	0.032	0.013			
Omy_UT16_2-173	GSI	.000189	0.131	0.131	0.013	0.016			
Omy_vamp5-303	GSI	.032422	0.327	0.322	0.045	0.032			
Omy_zg57-91	GSI	.000233	0.160	0.170	0.039	-0.044			
Omy_ndk-152	GSI	.000152	0.053	0.051	0.016	0.036			

<sup>a</sup> Linkage between these loci was found to be significant. Omy\_mapK3-103 was dropped from GSI analyses.

Table 4. Summary of 95 SNP markers (Appendix B and Matala et al. 2011) genotyped across 32 Chinook salmon populations from throughout the Snake River. Summary statistics include minor-allele frequency (MAF) range, expected ( $H_E$ ) and observed ( $H_O$ ) heterozygosity, Weir and Cockerham (1984)  $F_{ST}$ , and fixation index ( $F_{IS}$ ). "HWE" designates the number of populations that a SNP deviated from Hardy-Weinberg expectation for any SNP that deviated in greater than 10% of populations. "LD" is the number of populations that a pair of SNP loci exhibited linkage disequilibrium if it occurred in more than half of the populations. "CS" indicates a locu that was designated as a candidate for divergent (+) or balancing (-) selection.

SNP	MAF Range	HE	Ho	F <sub>ST</sub>	F <sub>IS</sub>	HWE	LD	CS	Comments
Ots_128757-61R	.006189	0.163	0.165	0.026	0.002				
Ots_lkaros-250	.000175	0.140	0.139	0.028	0.016				
Ots_nkef-192	.190628	0.471	0.489	0.019	-0.033	(5)			
Ots_u07-07.161	.138593	0.447	0.459	0.034	-0.015	(4)			
Ots_u6-75	.020307	0.215	0.207	0.029	0.043	(4)			
Ots_113242-216	.011178	0.180	0.184	0.016	-0.006	. ,			
Ots_CD59-2	.278563	0.469	0.473	0.016	-0.016				
Ots_GDH-81x	.032489	0.354	0.341	0.038	0.036				
Ots_IL8R_C8	.223537	0.461	0.454	0.023	0.023				
Ots_NOD1	.122411	0.394	0.404	0.018	-0.007				
Ots_SWS1op-182	.225516	0.420	0.411	0.016	0.033				
Ots_u07-17.135	.036260	0.193	0.190	0.024	0.017				
Ots_unk526	.000261	0.210	0.206	0.034	0.013				
Ots_105105-613	.159512	0.435	0.430	0.033	0.023				
Ots_94857-232R	.311789	0.480	0.461	0.029	0.037				
Ots GPH-318	.047373	0.304	0.305	0.035	0.012				
Ots_mapK-3'-309	.128620	0.450	0.425	0.034	0.064				
Ots_TAPBP	.006638	0.335	0.318	0.174	0.043			(+)	
Ots_u07-18.378	.000220	0.173	0.180	0.033	-0.028			~ /	
Ots_CD63	.000132	0.097	0.096	0.017	0.022				
Ots_myo1a-384	.000138	0.050	0.050	0.028	0.009				
Ots_SL	.000163	0.029	0.028	0.066	0.048				
Ots_CRB211	.000045	0.010	0.010	0.014	-0.019				
Ots_113457-40R	.013201	0.161	0.160	0.023	0.012				
Ots_97077-179R	.000086	0.044	0.043	0.018	0.011				
Ots_GH2	.000133	0.077	0.082	0.022	-0.059				
Ots_IL11	.000261	0.133	0.133	0.061	0.013				
Ots_myoD-364	.000189	0.106	0.112	0.026	-0.045				
Ots_PGK-54	.000077	0.031	0.030	0.020	0.013				
Ots_zP3b-215	.000000	0.000	0.000	-	-				Monomorphic <sup>c</sup>
Ots_123048-521	.000033	0.015	0.015	0.007	-0.015				
Ots_AldB1-122	.000206	0.121	0.110	0.014	0.104				
Ots_EndoRB1-486	.000190	0.058	0.062	0.055	-0.068				
Ots_GnRH-271	.000153	0.054	0.056	0.038	-0.005				
Ots_u07-53.133	.000267	0.122	0.120	0.028	0.037				
Ots_ZR-575	.000171	0.103	0.081	0.019	0.226	(12)			
Ots_GST-207	.000037	0.007	0.007	0.015	-0.019	· · /			
Ots_RAS1	.000000	0.000	0.000	-	-				Monomorphic <sup>c</sup>
Ots_aldb-177M	.009114	0.118	0.123	0.008	-0.033				·
Ots_EP-529	.000054	0.039	0.038	0.011	0.008				
Ots_GPDH-338	.000046	0.008	0.008	0.024	0.064				
Ots_u07-57.120	.000203	0.065	0.063	0.068	0.056				
Ots_TNF	.000017	0.001	0.001	-	-				Monomorphic <sup>c</sup>

Table 4. Continued.

SNP	MAF Range	HE	Ho	F <sub>ST</sub>	F <sub>IS</sub>	HWE	LD	CS	Comments
Ots_nramp-321	.000178	0.026	0.020	0.091	0.248				
Ots RFC2-558	.000032	0.013	0.013	0.004	-0.012				
Ots_u202-161	.000144	0.073	0.076	0.020	-0.037				
Ots hsc71-5'-453	.000165	0.079	0.078	0.035	0.020		(23) <sup>a</sup>		
Ots aspat-196	.000007	0.000	0.000	-	-		()		Monomorphic <sup>c</sup>
Ots FARSLA-220	.000312	0.064	0.065	0.107	-0.013				
Ots GST-375	000 - 000	0.000	0.000	-	-				Monomorphic <sup>c</sup>
Ots_LWSop-638	000 - 006	0.000	0.000	-	-				Monomorphic <sup>c</sup>
Ots Tnsf	109 - 687	0 420	0 425	0.061	-0.006				monorprio
Ots_arp-436	000 - 054	0.013	0.013	0.021	-0.029				
$Ots_hsp27b-150$	005 - 160	0.081	0.080	0.022	0.023				
Ots_U07-20.332	000 - 005	0.000	0.000	-	-				Monomorphic <sup>c</sup>
Ots 96222-525	000 - 154	0.000	0.000	0.037	0 019				Monorhorpino
Ots C3N3	000 - 187	-	-	0.007	-				mtDNA <sup>d</sup>
Ots EGE64	122 - 571	0 456	0 /67	0.000	-0 020		(33) <sup>b</sup>		IIICONA
Ots Ots 311-101y	000 - 072	0.400	0.407	0.020	-0.020		(55)		
Ots Cath D1/1	000 - 072	0.003	0.003	0.040	0.004				
$Ots_0att_D1+1$	000 = .075	0.024	0.025	0.024	0.052				Monomorphic <sup>c</sup>
Ots_007-04.221	.000003	0.000	0.000	0.001	0.260				Monomorphic
$Ots_Myc-300$	.000014	0.003	0.002	0.001	0.200				
$Ols_F400$	.000070	0.035	0.031	0.017	0.120				Monomorphio <sup>c</sup>
Oto 106747 220	.000005	0.000	0.000	-	-	(4)			Monomorphic
Oto_100747-239	.22007 1	0.457	0.440	0.043	0.040	(4)			
Ots_94903-99R	.1/25/8	0.474	0.478	0.019	0.002				
$Ols_COX 1-241$	.009333	0.229	0.224	0.045	0.030				
Ots_GTH2B-000	.305615	0.482	0.483	0.027	-0.008				
Ots_maprpr-151	.075489	0.361	0.351	0.035	0.024				
	.089457	0.380	0.384	0.031	0.001				
	.006105	0.100	0.096	0.013	0.029				
Ots_007-25.325	.295692	0.476	0.464	0.040	0.032				
Ots_96500-180	.233689	0.469	0.455	0.032	0.033				
$Ots\_E2-275$	.106538	0.400	0.408	0.030	-0.015				
	.000152	0.105	0.107	0.012	-0.011				
Ots_RAG3	.028282	0.201	0.202	0.037	0.010				
Ots_007-49.290	.156472	0.420	0.412	0.015	0.020				
Ots_96899-35/R	.000273	0.223	0.217	0.019	0.028		(oo)a		
$Ots_{11}SC77-3-488$	.034290	0.264	0.270	0.021	-0.011		(23)	$(\cdot)$	
	.153767	0.417	0.429	0.092	-0.021			(+)	
Ots_1LR3	.097489	0.377	0.397	0.065	-0.039				
Ots_102414-395	.202645	0.471	0.494	0.044	-0.035				
Ots_ARNI	.042341	0.301	0.306	0.031	-0.006	(7)			
Ots_ETIF1A	.176444	0.416	0.423	0.014	0.000	(7)			
Ots_HSP90B-100	.078306	0.280	0.275	0.016	0.019				
Ots_mypp-85	.000262	0.194	0.184	0.030	0.067				
Ots_P53	.107444	0.364	0.362	0.034	0.016				
Ots_S7-1	.144549	0.437	0.422	0.026	0.032	(5)			
Ots_u211-85	.180580	0.465	0.457	0.034	0.016				
Uts_AsnRS-60	.044339	0.310	0.304	0.017	0.023		(ac) <sup>h</sup>		
Uts_FGF6B_1	.167614	0.477	0.494	0.028	-0.022		(33)		
Uts_IGF-I.1-76	.067378	0.296	0.288	0.037	0.050				
Ots_SCIkF2R2-135	.211659	0.466	0.465	0.041	0.014				
Ots_u4-92	.000103	0.077	0.076	0.014	0.035				
Uts_110064-383	.067453	0.432	0.420	0.015	0.035				

Table 4. Continued.

- <sup>a</sup> Linkage between the *Ots\_hsc71* loci was found to be significant, *Ots\_hsc71-3'-488* was dropped from GSI analyses.
- <sup>b</sup> Linkage between the *Ots\_FGF6* loci was found to be significant, *Ots\_FGF6A* was dropped from GSI analyses.
- <sup>c</sup> These SNPs were found to be fixed across Chinook salmon baseline v1.0 populations and were dropped from GSI analyses;
- <sup>d</sup> Located on mitochondrial DNA

Table 5.Steelhead reporting regions for each baseline population used for GSI in the<br/>Snake River. Major population groups (MPG) are noted for each population to<br/>demonstrate the overlap of reporting regions with MPG.

Man #	Basalina Banulation	MDC
		IVIEG
upper Salmo		0.1
1	Sawtooth Weir	Salmon
	West Fork Yankee Fork Salmon	
2	River	Salmon
3	Morgan Creek	Salmon
4	Pahsimeroi River Weir	Salmon
5	North Fork Salmon River	Salmon
MF Salmon		
6	Marsh Creek	Salmon
7	Rapid River (MF)	Salmon
8	Pistol Creek	Salmon
g	Camas Creek	Salmon
10	Big Creek - Upper	Salmon
10	Big Creek - Lower	Salmon
12	Loon Crook	Salmon
12	Loon Creek	Salmon
IJ SE Salman	Dargamin Creek	Saimon
SF Saimon	Fast Fast Osyth Fast Ostrong	
	East Fork South Fork Salmon	0
14	River	Salmon
15	Secesh River	Salmon
16	Stolle Meadows	Salmon
Lower/Little S	Salmon	
17	Boulder Creek	Salmon
18	Hazard Creek	Salmon
19	Rapid River	Salmon
20	Slate Creek	Salmon
21	Whitebird Creek	Salmon
Upper Clearw	vater	
22	Storm Creek	Clearwater
23	Crooked Fork Lochsa River	Clearwater
24	Canvon Creek	Clearwater
25	Bear Creek	Clearwater
26	North Fork Moose Creek	Clearwater
27	Gedney Creek	Clearwater
28	O'Hara Creek	Clearwater
SE Clearwate		Clearwaler
20	Crocked River	Cleanwater
29	Topmilo Crook	Clearwater
30		Clearwater
31	John's Creek	Clearwater
32 Lawar Ola am		Clearwater
Lower Clearw		
33	East Fork Potlatch River	Clearwater
34	Big Bear Creek	Clearwater
35	Little Bear Creek	Clearwater
36	Mission Creek	Clearwater
Table 5. Conti	nued.	

Map #	<b>Baseline Population</b>	MPG
Imnaha		
37	Big Sheep Creek	Imnaha
38	Camp Creek	Imnaha
39	Cow Creek	Imnaha
40	Lightning Creek	Imnaha
Grande Ror	nde	
41	Crooked Creek	Grande Ronde
42	Elk Creek	Grande Ronde
43	Little Minam River	Grande Ronde
44	Lostine River	Grande Ronde
45	Menatchee Creek	Grande Ronde
46	Wenaha River	Grande Ronde
47	Asotin Creek	Lower Snake R.
Below LGD		
48	Tucannon River	Lower Snake R.
49	Touchet River	-

Table 6.Chinook salmon reporting regions for each population used for GSI in the Snake<br/>River. Major population groups (MPG) are noted for each population to<br/>demonstrate the overlap of reporting regions with MPG.

Map #	Baseline Population	MPG
Upper S	Salmon	
1	Sawtooth Weir	Upper Salmon
2	West Fork Yankee Fork Salmon River	Upper Salmon
3	East Fork Salmon River	Upper Salmon
4	Pahsimeroi River Weir	Upper Salmon
5	Hayden Creek (Lemhi)	Upper Salmon
6	L3Á Trap (Lemhi)	Upper Salmon
7	Lower Lemhi Trap	Upper Salmon
MF Salr	non	
8	Marsh Creek	Middle Fork Salmon
9	Capehorn Creek	Middle Fork Salmon
10	Elk Creek	Middle Fork Salmon
11	Bear Valley Creek	Middle Fork Salmon
12	Sulphur Creek	Middle Fork Salmon
13	Camas Creek	Middle Fork Salmon
14	Big Creek	Middle Fork Salmon
15	Chamberlain Creek	Middle Fork Salmon
SF Saln	non	
16	Lake Creek (Secesh)	South Fork Salmon
17	Secesh River (Lower)	South Fork Salmon
18	Secesh River (Upper)	South Fork Salmon
19	Johnson Creek	South Fork Salmon
20	South Fork Salmon River Weir	South Fork Salmon
Clearwa	ater / Grande Ronde / Imnaha	
22	Powell Weir	NA
23	Red River Weir	NA
24	Crooked River Weir	NA
25	Newsome Creek	NA
26	Lolo Creek	NA
27	Imnaha River	Grande Ronde / Imnaha
28	Catherine Creek	Grande Ronde / Imnaha
29	Lostine River Weir	Grande Ronde / Imnaha
30	Minam River	Grande Ronde / Imnaha
_ 31	Wenaha River	Grande Ronde / Imnaha
Tucann	on	
32	Lucannon River	Lower Snake

Table 7.Results from leave-one-out tests performed in ONCOR (Kalinowski et al. 2007) for steelhead baseline v1.0. Rows<br/>represent population where individuals originated from. Columns represent reporting regions that individuals assigned<br/>to. Shaded boxes represent the correct reporting region of origin for each population.

	Unner	МГ	<u>ег</u>	L avvar/l :441a	Unnor	65	Lewer		Cranda	Balaw
Population	Salmon	Salmon	JF Salmon	Salmon	Clearwater	JF Clearwater	Clearwater	Imnaha	Bonde	LCP
Soutooth Woir				0.07				0.07		0.00
West Fork Vankoo Fork Salmon Biyor	0.62	0.00	0.00	0.07	0.00	0.00	0.04	0.07	0.00	0.00
Morgan Creek	0.00	0.03	0.00	0.21	0.00	0.00	0.00	0.07	0.00	0.05
Pahsimeroj River Weir	0.75	0.04	0.00	0.04	0.00	0.00	0.00	0.04	0.15	0.00
North Fork Salmon River	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
Marsh Creek	0.00	1.00	1 0.00	0.00	0.02	0.02	0.00	0.00	0.00	0.04
Rapid River (MF)	0.00	1.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
Pistol Creek	0.00	1.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
Camas Creek	0.00	0.97	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
Big Creek - Upper	0.00	1.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
Big Creek - Lower	0.04	0.89	0.00	0.00	0.04	0.00	0.00	0.00	0.04	0.00
Loon Creek	0.00	0.91	0.00	0.09	0.00	0.00	0.00	0.00	0.00	0.00
Bargamin Creek	0.04	0.82	0.00	0.04	0.00	0.00	0.04	0.04	0.00	0.04
East Fork South Fork Salmon River	0.00	0.05	0.86	0.09	0.00	0.00	0.00	0.00	0.00	0.00
Secesh River	0.00	0.04	0.87	0.09	0.00	0.00	0.00	0.00	0.00	0.00
Stolle Meadows	0.00	0.00	0.93	0.00	0.00	0.00	0.00	0.00	0.07	0.00
Boulder Creek	0.21	0.04	0.00	0.58	0.00	0.04	0.04	0.04	0.04	0.00
Hazard Creek	0.31	0.08	0.00	0.35	0.08	0.00	0.04	0.08	0.04	0.04
Rapid River	0.04	0.04	0.00	0.84	0.00	0.00	0.00	0.08	0.00	0.00
Slate Creek	0.18	0.00	0.00	0.55	0.00	0.05	0.05	0.00	0.05	0.14
Whitebird Creek	0.17	0.00	0.00	0.50	0.00	0.07	0.03	0.10	0.03	0.10
Storm Creek	0.00	0.00	0.00	0.00	1.00	0.00	0.00	0.00	0.00	0.00
Crooked Fork Lochsa River	0.00	0.00	0.00	0.00	0.90	0.10	0.00	0.00	0.00	0.00
Canyon Creek	0.00	0.00	0.00	0.00	0.81	0.19	0.00	0.00	0.00	0.00
Bear Creek	0.00	0.00	0.00	0.00	1.00	0.00	0.00	0.00	0.00	0.00
North Fork Moose Creek	0.00	0.00	0.00	0.00	1.00	0.00	0.00	0.00	0.00	0.00
Gedney Creek	0.00	0.00	0.00	0.00	0.95	0.05	0.00	0.00	0.00	0.00
O'Hara Creek	0.00	0.00	0.00	0.00	0.76	0.16	0.08	0.00	0.00	0.00
Crooked River	0.00	0.00	0.00	0.00	0.03	0.94	0.00	0.00	0.03	0.00
Tenmile Creek	0.00	0.00	0.00	0.00	0.14	0.83	0.03	0.00	0.00	0.00
John's Creek	0.00	0.00	0.00	0.00	0.18	0.71	0.12	0.00	0.00	0.00
Clear Creek	0.00	0.00	0.00	0.00	0.28	0.72	0.00	0.00	0.00	0.00
East Fork Potlatch River	0.00	0.00	0.03	0.00	0.05	0.03	0.78	0.00	0.08	0.03
Big Bear Creek	0.00	0.00	0.00	0.07	0.07	0.07	0.53	0.07	0.20	0.00
Little Bear Creek	0.00	0.00	0.00	0.00	0.08	0.00	0.85	0.00	0.00	0.08
Mission Creek	0.00	0.00	0.00	0.00	0.00	0.00	0.57	0.03	0.30	0.10
Big Sheep Creek	0.11	0.03	0.00	0.08	0.00	0.00	0.00	0.68	0.05	0.05
Camp Creek	0.07	0.00	0.07	0.00	0.00	0.00	0.00	0.60	0.20	0.07
Cow Creek	0.08	0.03	0.00	0.08	0.00	0.00	0.03	0.54	0.21	0.05
Lightning Creek	0.05	0.00	0.00	0.05	0.00	0.00	0.00	0.80	0.00	0.10
Crooked Creek	0.04	0.01	0.00	0.03	0.00	0.00	0.03	0.07	0.70	0.10
Elk Creek	0.00	0.00	0.00	0.00	0.00	0.00	0.03	0.03	0.92	0.03

## Table 7. Continued.

	Upper	MF	SF	Lower/Little	Upper	SF	Lower		Grande	Below
Population	Salmon	Salmon	Salmon	Salmon	Clearwater	Clearwater	Clearwater	Imnaha	Ronde	LGR
Little Minam River	0.00	0.00	0.00	0.03	0.00	0.00	0.18	0.00	0.77	0.03
Lostine River	0.06	0.00	0.00	0.00	0.00	0.00	0.06	0.03	0.81	0.03
Menatchee Creek	0.00	0.00	0.00	0.16	0.00	0.00	0.04	0.04	0.72	0.04
Wenaha River	0.02	0.00	0.02	0.04	0.00	0.00	0.04	0.05	0.75	0.09
Asotin Creek	0.19	0.03	0.00	0.08	0.00	0.03	0.08	0.14	0.41	0.05
Tucannon River	0.08	0.00	0.00	0.08	0.04	0.00	0.15	0.12	0.19	0.35
Touchet River	0.00	0.02	0.00	0.05	0.00	0.00	0.05	0.04	0.07	0.77

Table 8.Results from leave-one-out tests performed in ONCOR (Kalinowski et al. 2007)<br/>for sp/sum Chinook salmon baseline v1.0. Rows represent population where<br/>individuals originated from. Columns represent reporting regions that individuals<br/>assigned to. Shaded boxes represent the correct reporting region of origin for<br/>each population.

	Upper	MF	SF	Clearwater /	Lower
Population	Salmon	Salmon	Salmon	Grande Ronde / Imnaha	Snake
Sawtooth Weir	0.66	0.16	0.10	0.09	0.00
West Fork Yankee Fork Salmon River	0.81	0.07	0.04	0.06	0.01
East Fork Salmon River	0.87	0.08	0.05	0.00	0.00
Pahsimeroi River Weir	0.64	0.07	0.13	0.16	0.00
Hayden Creek (Lemhi)	0.90	0.01	0.00	0.08	0.00
L3A Trap (Lemhi)	0.72	0.05	0.04	0.18	0.01
Lower Lemhi Trap	0.72	0.06	0.07	0.14	0.00
Marsh Creek	0.17	0.68	0.13	0.02	0.00
Capehorn Creek	0.00	0.87	0.11	0.02	0.00
Elk Creek	0.07	0.86	0.07	0.00	0.00
Bear Valley Creek	0.03	0.83	0.10	0.03	0.00
Sulphur Creek	0.08	0.81	0.08	0.04	0.00
Camas Creek	0.00	0.93	0.03	0.03	0.03
Big Creek	0.03	0.76	0.10	0.10	0.01
Chamberlain Creek	0.05	0.93	0.00	0.02	0.00
Lake Creek (Secesh)	0.03	0.08	0.86	0.03	0.00
Secesh River (Lower)	0.02	0.15	0.65	0.19	0.00
Secesh River (Upper)	0.11	0.11	0.78	0.00	0.00
Johnson Creek	0.11	0.24	0.56	0.09	0.00
South Fork Salmon River Weir	0.28	0.21	0.30	0.20	0.00
Powell Weir	0.18	0.06	0.09	0.64	0.03
Red River Weir	0.07	0.09	0.04	0.76	0.04
Crooked River Weir	0.06	0.07	0.06	0.76	0.04
Newsome Creek	0.10	0.08	0.04	0.78	0.00
Lolo Creek	0.10	0.08	0.06	0.74	0.03
Imnaha River	0.15	0.13	0.08	0.63	0.01
Catherine Creek	0.05	0.06	0.12	0.74	0.03
Lostine River Weir	0.18	0.00	0.06	0.72	0.04
Minam River	0.11	0.05	0.00	0.76	0.08
Wenaha River	0.11	0.00	0.05	0.78	0.05
Tucannon River	0.02	0.06	0.02	0.20	0.70

FIGURES



Figure 1. Steelhead populations representing steelhead baseline v1.0 for the Snake River. Population numbers correspond to numbers in Table 1. The locations of Lower Granite Dam and Hells Canyon Dam is noted.



Figure 2. Chinook salmon populations representing Chinook baseline v1.0 for the Snake River. Population numbers correspond to numbers in Table 2. The locations of Lower Granite Dam and Hells Canyon Dam is noted.



Figure 3. NJ-phylogram based on Nei's (1972) distance for steelhead baseline v1.0. Numbers correspond to map numbers in Figure 1. Brackets designate reporting regions used for genetic stock identification.



Figure 4. NJ-phylogram based on Nei's (1972) distance for Chinook salmon baseline v1.0. Numbers correspond to map numbers in Figure 1. Brackets designate reporting regions used for genetic stock identification. A sample from John Day River was included as an out-group.



Figure 5. Mean pairwise F<sub>ST</sub> estimates for baseline steelhead populations. The dashed line represents the average pairwise F<sub>ST</sub> estimate for all populations. High mean pairwise F<sub>ST</sub> estimates suggest high levels of genetic differentiation relative to other baseline populations.



Figure 6. Mean pairwise  $F_{ST}$  estimates for baseline Chinook salmon populations. The dashed line represents the average pairwise estimate for all populations. High mean pairwise  $F_{ST}$  estimates suggest high levels of genetic differentiation relative to other baseline populations.



Figure 7. Stock proportions for SY 2009 (7/1/2008 – 6/30/2009) and SY2010 (7/1/2009 – 6/30/1010) natural origin summer-run steelhead at Lower Granite Dam.



Figure 8. Stock proportions for SY2009 natural origin spring/summer Chinook salmon at Lower Granite Dam.



Figure 9. Sex ratios for SY2009 (7/1/2008 – 6/30/2009) and SY2010 (7/1/2009–6/30/1010) for each reporting region for natural origin summer-run steelhead at Lower Granite Dam.



Figure 10. Sex ratios for SY2009 for each reporting region for natural origin spring/summer Chinook salmon at Lower Granite Dam.



Figure 11. Run-timing at Lower Granite Dam for each of the reporting groups based on date of capture of adult steelhead sampled at the adult trap located on the fish ladder. Run timing includes median passage date with 5<sup>th</sup>, 25<sup>th</sup>, 75<sup>th</sup>, and 95<sup>th</sup> percentiles. Passage date refers to days from July 1 (i.e. July 1 = Passage Date 1).



Figure 12. Run-timing at Lower Granite Dam for each of the reporting groups based on date of capture of adult Chinook salmon sampled at the adult trap located on the fish ladder. Run timing includes median passage date with 5<sup>th</sup>, 25<sup>th</sup>, 75<sup>th</sup>, and 95<sup>th</sup> percentiles. Julian date refers to days from January 1<sup>st</sup> (i.e. January 1 = Julian Date 1).

APPENDICES

Appendix A. TaqmanTM assays used for *O. mykiss*. All forward and reverse primers are unlabeled and probes contain a 5' fluorophore and a 3' quencher and minor groove binder.

SNP/Comments	Panel	Primers	Probes
Omy_SEXY1	PBT	F - CACAACATGAGCTCATGGG	
-		R - CGATTAGAAAGGCCTGCTTG	6FAM - CCTACCAAGTACAGCCCCAA
Autosomal control	PBT	F - GCCTGCTTGCAGAAGTTTTT	VIC - GAGGGGTAGTCGTTTGTTCG
control for Omy_SEXY1		R - CTTGACTGTGTCCAGCTTGC	
M09AAD.076	PBT	F - ACTGTTACCACTCTCTCATCAACCT	VIC - CACCAACCACTGGTGAA
		R - GGGTCCAGGAGGTTTTTAAACAACAT	6FAM - CCAACCGCTGGTGAA
M09AAJ.163	PBT	F - TCCCATGGCCCTTACTCTATCAA	VIC - AACAAAGTGAAAGTGTCCTTA
		R - TTGAGGTGTATGTTGAAAAGTAAACTT	6FAM - CAAAGTGAAAGTGTCTTTA
M09AAE.082	PBT	F - CTATGTGCAGTGCCCTTCTCA	VIC - AGGTTGTTTTACAAATTTAA
		R - GGCTTACAAGTATGCATGACTAGCT	6FAM - AGGTTGTTTTACACATTTAA
OMS00002	PBT	F - TTTGATTTGATTTGTATCTGCTTCTT	VIC - TGTTTTGCAGCGCTC
		R - CCAACATGCCTCACACAAAA	6FAM - TGTTTGGCAGCGCT
OMS00006	PBT	F - TCCACGTAGGACATAGTTTGAGCTA	VIC - CACTTACAAATACAAAATT
		R - TGTGGTGTCATGTTTGCCCTAC	6FAM - CTTACAAATGCAAAATT
OMS00024	PBT	F - CACATACAACCATCACCCTTCCTAA	VIC - AAAAACCCAAATTTTAC
		R - AGCATTGAGCGAAATTACCAAGAGT	6FAM - AACCCCAATTTTAC
OMS00039	PBT	F - GTCAGTACTGTGTGTGTGTGTGT	VIC - CAGAGACACGTACGCACA
		R - CCATCTACATTGTCAGCAGTGTGA	6FAM - AGACACGCACGCACA
OMS00053	PBT	F - GGAGCCAGGTCAAGGTGATC	VIC - TGTGTGATTGATACATATAAAT
		R - GGATGTCTGGTGTGGCTGTAAA	6FAM - TGTGATTGATACGTATAAAT
OMS00057	PBT	F - GAGAAAGGGAGCATGAGACAGA	VIC - CTCCACAGAACCTTG
		R - GTTGGGCTCCGGTACGAT	6FAM - CTCCACAGCACCTTG
OMS00058	PBT	F - GTGACATTTGGAGCCACTGC	VIC - CAACACTTTGTACCCCTC
		R - GCTAGGAGACAGAGGGTGAAAG	6FAM - CACTTTGCACCCCTC
OMS00062	PBT	F - ACCCTGGGAAGGCTACTGTAC	VIC - TTGACCAGCAGATGGTGTA
		R - TGAACAGAGATCTGGAGAGTTGGAT	6FAM - ACCAGCAGGTGGTGTA
OMS00064	PBT	F - GTGGATATGTAGTTCGATGGAACAGT	VIC - CAGGCAACATTTTATATAACTA
		R - TTTACAACAATCTTCTTTTAATAAAAATATAGCCACTTAT	6FAM - CAGGCAACATTTTATCTAACTA
OMS00068	PBT	F - GCACTAACTGGACAACATTTTTAAGAATGA	VIC - AATATGCCTCCTTCGTCTC
		R - GGCAGTTGAGCATTTTGGGATATT	6FAM - TATGCCTCCTCCGTCTC
OMS00070	PBT	F - CGTTCCTGCGGGACAGT	VIC - CAAAATACGGAAATGCAG
		R - GTTTCTCTCACGTCCACAGATCT	6FAM - AAATACGGGAATGCAG
OMS00071	PBT	F - CCGGAGTGACCTCACATTTGG	VIC - CTTGTTTGAGCTTTTTCT
		R - GCATCGTACAGTTCACCTACCT	6FAM - TTGTTTGAGCCTTTTCT
OMS00072	PBT	F - GTGGGAGAGCTCGTCTATGG	VIC - TAGAAGGTCCATGTATCTC
		R - ACAACAGGTCATTGGATGTGATCAG	6FAM - AAGGTCCATGCATCTC
OMS00074	PBT	F - CCTGTTTATTCATCTAAACCAGTTCTTTAAAAT	VIC - TGAAACAAAACAAATGTTCC
····		R - AACTTAATTTAGCAAACAAATGTCTGAACAGAA	6FAM - AAACAAAACACATGTTCC
OMS00077	PBT	F - AATACCATCTTGAGCTCATTAGTAATTATTCAA	VIC - TTCCGGTGGTGAAGTT
		R - CCAGACTTTACACACTCTTGACTGA	6FAM - CCGGTGCTGAAGTT
UMS00078	PBT	F - GAGGGAAGCAGCCATAAACAGAATA	VIC - TTCACATGCATAAGAGTG
o. /o		R - GICICACIATGGTCCATATCTGTGTAGA	6FAM - ICACATGCATGAGAGTG
OMS00079	PBT		
		R - ACCIGCAACGIIAGAGCIGIIIAII	6FAM - CTACTTTTCACAGTGACACAG
Appendix A. Continued.			

SNP/Comments	Panel	Primers	Probes
OMS00111	PBT	F - CATGCGGACCTGCATAGCT	VIC - CAACCAGACTACCATTC
		R - GCTTAGCCATTGACAGAGCATATCA	6FAM - AACCAGACTGCCATTC
OMS00089	PBT	F - GCACCATTTGAATAAAAAATCTGCTTTGT	VIC - ATGAATCCCAAATAAGAAC
		R - GCAACCCAATTCAATATTAAGCACATGAT	6FAM - AATCCCAAACAAGAAC
OMS00090	PBT	F - AGGGCACAACACCACTCTAAATT	VIC - ACAACCACAAGATT
		R - TCGAAAAGCAACATCTGTCTCAGT	6FAM - AACCACGCAAGATT
OMS00101	PBT	F - GCGTGTCGTGGGTCAGTTAAATA	VIC - CTCTAGTAGCCTTATAGAAAG
		R - GTGCAATCCAACCTATTAGTAGATATGCT	6FAM - CTAGTAGCCTTACAGAAAG
OMS00105	PBT		
	i Di		6FAM - CTGCTATTCACATTGCT
0MS00106	DBT		
0111000100	1 DT		
014500154	ррт		
01/1300154	РЫ		
014000440	DDT		
01/1300112	PRI		
011000110			6FAM - CGGIIICAAGIAIACIIGI
OMS00118	PBT	F - GCTTATTTAGAGTGCATGCCAGATG	VIC - AATGTGCACACCCCGC
		R - TGGAACCAATGGGACAGTCCTA	6FAM - AATGTGCACCCCCGC
OMS00120	PBT	F - GGCAGAAGAGGAGAGAGATATGATTG	VIC - TCGCCCACTAAAAC
		R - CCTCAAATACCTCTGACATTGAAGGTT	6FAM - CGCCCACCAAAAC
OMS00121	PBT	F - GGAAGGAGGTCCAGTGTGAGT	VIC - ACAGCGTGATAAATT
		R - AAAATATGCAACACCACTAAAACTGGAAAA	6FAM - CAGCGTGGTAAATT
OMS00132	PBT	F - GTTTATGACTCCATTGCCGAAATGATT	VIC - CAGCAGTCCTCTGTGTGG
		R - ACGCGACCTGCAATTCATCAATA	6FAM - AGCAGTCCTCAGTGTGG
OMS00175	PBT	F - TTGCGATATGGGACTGTATACATTTATTCC	VIC - CATCACTAGTTCAAATACAA
		R - ACTACCTCCAGTTAAAATAGTGTGGGAAA	6FAM - CATCACTAGTTCAGATACAA
OMS00179	PBT	F - GTCATAACAAAATCAGGGCTTTCCAA	VIC - TGCCTCTTCTCTTTTCTCAT
		R - TGGGAGATTTGGGCTGCTTTAAA	6FAM - CCTCTTCTCTTGTCTCAT
OMS00180	PBT	F - GCGCCGAATGGCATTAGG	
	1.01		6FAM - CTAAAAGTGCCTTAAGCC
Omv 101832-195	DBT		
onny_101032-199	1 DT		
$O_{mv}$ 101002 180	ррт		
Ully_101993-169	FDI		
0 100 505 100	DDT		
Omy_102505-102	PBT		
• · · · · · · · · · ·		R - IGCTIGCTITTTAAAAACAATCTCCCCA	6FAM - CAGGATGCTTTTGC
Omy_104519-624	PBT	F - CGIGIGAGIIIGCGGIAAAGAC	VIC - CAGCAGGATACATCCGACT
		R - TGACGAGTCCGTCTTATCATCCT	6FAM - AGCAGGATACGTCCGACT
Omy_105105-448	PBT	F - CAATTTGCAAGCAGGGAAAGGTTAT	VIC - AAGGAGAATGCATAATC
		R - GTGATGGGCTGCAATTGCTT	6FAM - TGAAAGGAGAATACATAATC
Omy_105385-406	PBT	F - ACCTACCCTCACCTGAACTTCA	VIC - CTTGGAACCATTGCTAC
		R - CGCTCTTCTGGGCGTATCG	6FAM - TTGGAACCGTTGCTAC
Omy_105714-265	PBT	F - CCACTCAGTGCAAGCATGGA	VIC - CTGTTGTTTGAGGTTCAG
-		R - GCTTTCAATCCTTGGCTCCAATATC	6FAM - TGTTGTTTGAGATTCAG
Omv 107806-34	PBT	F - TCTTTGTCCATGCACATTGATATT	VIC - ATTGGATGTCAGTGTCATT
,	. 21	R - AGCACATTTAGTTAGCAGTGATGGA	6FAM - ATTGGATGTCAATGTCATT
Omv 108007-193	PRT	F - GTGAATACCACCCAGGCTTGT	
J	101		6FAM - TTTTCTCCCCACTTAAC
Appondix A Continued			
appendix A. Continued.			

SNP/Comments	Panel	Primers	Probes
Omy 109243-222	PBT	F - ATGTGCACCTCTTAAATTGTAAGTAAAATGT	VIC - TGTTCATTAAATTGACTTTTT
)		R - ACCCTATATTCAGTGGCAAGATTGC	6FAM - TTCATTAAATGGACTTTTT
Omv 109894-185	PBT	F - CGGTGTCATTATGGTTGTCATTGTG	VIC - CTCCCTGATCCCCC
)		R - GGGAGGAATTGGAATGACAGATTAAC	6FAM - CTCCCTGGTCCCCC
Omv 110064-419	PBT	F - GTGCAAGGGACCTAGCTAATCC	VIC - ACGTTAGCTTTTAATTTC
		R - TCTGAACTGACACTGAAGAACAAAGAA	6FAM - AACGTTAGCTTTTCATTTC
Omv 111383-51	PBT	F - CACGCGCAATCTCTCGTTTTAC	VIC - ACCTAGTGCGCTTGCT
o,	101	R - TCTTTAGGCAACAAGCGTGTCA	6EAM - ACCTAGTGCACTTGCT
Omv 113490-159	PBT	F - CATAGTACATTTACAGATAATGTTTTAAAGTGCATGT	
o,		R - CGAGATACCAAAATGCCACAGTTACAT	6FAM - CATCTGTTTTAGTTTAGC
Omv 114315-438	PRT		VIC - TTATGGGCTTAAGGGTC
	101		6FAM - TTATGGGCTTACGGGTC
Omv 114587-480	DRT		
omy_rrador add	101		6FAM - CCTGTCCACAATTGT
$\Omega_{mv}$ 120870-756	DDT		
Olliy_129010-150	FDI		
$\Omega_{mv}$ 116733-340	DDT		
Olliy_110733-349	FDI		
Omy 128022 122	прт		
0111y_120923-433	FDI		
Omy 120524 160	пот		
Onny_130324-100	PDI		
0.000 0.7660 0.20	прт		
Omy_97660-230	PDI		
0,000,000,000	прт		
Omy_99300-202	PDI		
Omic old D 165	ппт		
Only_alub-105	PDI		
0	DDT		
Omy_anp-17	PBI		
0	DDT		6FAM - CTCATIGGTATATTAACC
Omy_arp-630	PRI		
0	DDT		
Omy_b1-266	PBT	F - TCATGTGAACTTTAATTGACTAGGAAGTCG	
		R - GATAIGAAAATAICIGAAGAGTIATAITIGGGAAATIGAC	6FAM - ICTATAAACAAAATTITIC
Omy_BAC-B4-324	PBT	F - GCCTAATATTGGCCTAATGTCCTTCA	VIC - CATTGCCAAATACG
<b>a i i a = i</b>			6FAM - TACATIGACAAATACG
Omy_ada10-71	PBT	F - TCTTTGAGCGACAAAGTCCTTGT	VIC - CTTCCTGCGTCCAATT
		R - ACCCACACATGAACGCAAAAG	6FAM - CTTCCTGCATCCAATT
Omy_redd1-410	PBT	F - GTACTCCCACTAACATACAGTAGACTCA	VIC - AAAATATCCTGCAAGGAAT
		R - GGCACCATTGTGTTTTAGGATGTAG	6FAM - AATATCCTGCAAGAAAT
Omy_cd59-206	PBT	F - CGATTGGCCCAGATGTTTCCAT	VIC - CAACAATCGAAGGTAAAT
		R - GCTCCGTTGCATAGGTGACT	6FAM - CAACAATCAAAGGTAAAT
Omy_colla1-525	PBT	F - CCTCGGCGTGACAACCT	VIC - CTGTTGGGAGAAGAG
		R - CCCAGAGAATGGTGCGATTAGG	6FAM - TGTTGGGAAAAGAG
Omy_cox1-221	PBT	F - CACTGAACTGTAAGCCATTGTGATT	VIC - CGGTAAGACCATTAAAA
		R - GCAACATGGGAATGATTCATAAATGCA	6FAM - CGGTAAGACCATTTAAA
Omy_crb-106	PBT	F - GCTCAAAAAGATTCTGCCAAATTCACA	VIC - TTGCAATGCGTCTTT
		R - ATTACAATGAAAGTACTTGAGTGTTTATGCAAA	6FAM - TTGCAATGAGTCTTT
Appendix A. Continued.			

SNP/Comments	Panel	Primers	Probes
Omy_g12-82	PBT	F - GATCAATTCGATCGCTCATGAAACTT	VIC - CAAACTCTCAGGATTAG
7_0		R - CTTCTCTCGTTCTCATTGTGTCTCA	6FAM - AAACTCTCGGGATTAG
Omy aluR-79	PBT	F - GACTGTCTATAGCTATTCTTCTCAAACTGT	VIC - CAAGTATTTTGCGTAGGAAT
<i>y</i> =0		R - AGAAACTACCATTGTGATTAACAGATAGAAAATACAT	6FAM - CAAGTATTTTGCATAGGAAT
Omy hsc715-80	PBT	F - CCGGTCTACCCTATAGCTGTTG	VIC - AACTGTATTTGGGAAAAT
2=		R - AGTCAGTCAATTAGTGGTTTGAAATACTATCA	6FAM - ATAAACTGTATTTGTGAAAAT
Omy hsf2-146	PBT	F - GGAGCAGAAAAAGGATTGGACCTT	VIC - CAGCTGTTAGTAGATTAT
,		R - CCAACAATTGCAGCCTCATCTTAAT	6FAM - ACAGCTGTTAGATTAT
Omv 1L17-185	PBT	F - CCACCACACTCTGCAGCTT	VIC - AAGAATCTCACCTGCCCAT
, <u>_</u>		R - TTGACGGGAATCCGAGACTTC	6FAM - AAGAATCTCACTTGCCCAT
Omv II-1b .028	PBT	F - ACTGTCTGGCTAGAGCACATTG	VIC - CTGAGGCAACTTTTGT
0		R - ATCTTCTACCACCGCACTGTTTTAA	6FAM - TGAGGCAGCTTTTGT
Omv 1116-198	PBT	F - TTTAATCTCGGTGCTGAGCTAGTG	
emy_me ree	101		6EAM - ACCTTAGTTGTAGCTTCAT
Omy 116-320	PRT		
enny_iee dee	101		
$Omv met A_{-}161$	DRT		
emy_metA-ren	101		6FAM - CAAGTAAGTGGTTCTATTCT
Omy NaKATRa2 50	DDT		
Only_Narva r Fa3-50	FDI		
Omy typin 242	ррт		
Omy_txmp-343	FDI		
Ome alcof 211	ррт		
Omy_nkei-241	PDI		
Omic at 27	прт		
Omy_nti-27	PBI		
0	DDT		
0111y_0g04-212	PBI		
	DDT		
Umy_bcAKala-380rd	PBI		
0	DDT		6FAM - CATACICATCCTATGTCAG
Omy_0ts249-227	PBT		VIC - CCCTCTGAGAACTAC
		R - CTATCTATCTATCTATCTATCTATCTATCTATCTATCTA	6FAM - CCTCTGAAAACTAC
Omy_oxct-85	PBT	F - CGTCACTGAAACATTACTGTAACATCCA	VIC - CATCGCTTATTTATGC
_		R - CATCATCACGCTGTTGGTTTCTTAA	6FAM - CATCGCTAATTTATGC
Omy_p53-262	PBT	F - CCCCAACATCCAGTATACAGTTTCA	VIC - CAAGTAGTATGGAGCTCTAT
		R - CCCAAATTGGCAATTTTAATAGGATTCAGA	6FAM - AAGTAGTATGGTGCTCTAT
Omy_rapd-167	PBT	F - CCCAACATGCTCTATTGCAGCTA	VIC - ATTAAAACAATCCCCCCAAAA
		R - AGTTGCATAAGATGAATCAATAAATTAAAAACACAGAT	6FAM - TTAAAACAATCCCACCCAAAA
Omy_rbm4b-203	PBT	F - CTGAAATTTGATGAATGGAAGCTGCA	VIC - CACGTTATTATGAAAAGGATGT
		R - CGTATTCAAGTCGATATACAGTCACGAT	6FAM - ACGTTATTATGAAAAAGGATGT
Omy_srp09-37	PBT	F - TAGTTGTATTAACTCTTCTTTGAGTCTAGA	VIC - TTGTGCTATTGACGCCACAG
		R - TCATTCCAGCTCCGTTCTCTTC	6FAM - TTGTGCTATTGACACCACAG
Omy_stat3-273	PBT	F - CAGACCTCCTCTATCTCCCTATGAG	VIC - TTTTCCAGACTCCAGTTTG
		R - ACCTCCTTTAAATTGTGCCCAAGAA	6FAM - TTTTCCAGACTCAGTTTG
Omy_u09-53.469	PBT	F - ACAGCCTGAGCGTTTGCA	VIC - TTGCAGCCCTTATTGTG
		R - GGAAACTGGGAGAGATCAAAGGA	6FAM - TTGCAGCCCTTGTTGTG
Omy_u09-54-311	PBT	F - GTGGCTCCCCAGGAACAAG	VIC - TGGTAATTATTCAACAGATCAGT
		R - AAGTTTCATGTCACATTCCAGTTACCT	6FAM - TGGTAATTATTCAACAAATCAGT
Appendix A. Continued.			

SNP/Comments	Panel	Primers	Probes
Omy U11 2b-154	PBT	F - GGGAAGCAGAAAAACTGGAAGTT	VIC - AATGATACTTTTCAGATTGTAAC
		R - CCCTCTGTGGGCTTGATATTCA	6FAM - TGATACTTTTCAGGTTGTAAC
Omy vatf-406	PBT	F - TTGCTTCATTTGTCATAACCTTGGG	VIC - TTGCAGATGACTATCCACA
/		R - TGCATGCTCTGACAAATGTTACACT	6FAM - TGCAGATGACTGTCCACA
OMY1011SNP	PBT	F - GAGGCTGGTTTGGGATTCACT	VIC - CTTTACCTCGAAGACAAT
		R - CGCCAAACACTAACTCTCTGTCT	6FAM - ACTITACCTCTAAGACAAT
Ocl calT7RT2	GSI	F - AGGTGTAGTTGCCTTCAGAATAAACTC	VIC - ACACATACAGTAGCCAAAT
O. <i>clarki</i> hybrid marker	00.	R - TCTCTCCCTCTCTGCCTGTTTT	6FAM - ACACATACAGTAGTCAAAT
Omv mvclarp404-111	GSI	F - GCTGTGGTGCTCATGGGTAAA	VIC - CAAAGCCATACGTGGCC
O. <i>clarki</i> hybrid marker		R - CCAGGGCAGGGTTGTTCTC	6FAM - AAGCCATCCGTGGCC
Omv Omvclmk438-96	GSI	F - CCCGACTCTACTTCACTACTTTCCT	VIC - TACGCAAATTAGGTTTAAA
O. <i>clarki</i> hybrid marker	001	R - GGCCTAGGACAATAGGACTGAAC	6FAM - CGCAAATTAGGGTTAAA
M09AAC 055	GSI	E - GTCTCCGACGTGTGGCT	
	001		6FAM - ACCTCCACACTGTCC
OMGH1PROM1-SNP1	GSI		
	001		6FAM - TAGTGTACACTGACTTCA
OM\$00003	GSI		
01110000003	001		6EAM - TACTGTCGCCATTTTA
014500008	CSI		
0///300008	631		
011500012	001		
01/1300013	631		
011500011	001		
01/1300014	631		
011000015	001		
0///500015	GSI		
011000017	001		
0///300017	651		
011000010	001		
0///200018	651		
0	0.01		6FAM - CCACATAATTCATAATTC
Omy_ca28-130	GSI		
01/000000	0.01	R - GAGGACAAAACIGACCGIAIGGI	6FAM - CIGIICGIICACCC
OMS00030	GSI	F - CCTCGTGACTACAGAGCTATACAAC	
014000040		R - GATCIGATCGGTCGGGAGAGA	6FAM - ATGAGGGTCCCTCTACAGG
OMS00048	GSI		VIC - CAGCTAAACTCAGCAAAA
			6FAM - AGCTAAACTCGGCAAAA
OMS00052	GSI	F - IGCGTTTTTCATCCCAATCATTCAC	VIC - CTTCCTTTTGAGAATAAT
		R - GGCATCAGGCTCTTCTTCCT	6FAM - CCTTTTGCGAATAAT
OMS00056	GSI	F - TCAGGAAGTAAACTGAAAATTCCAATGTATGA	VIC - TAGCTTGACCAAATAGCA
		R - CCCCAACCATGCTTGTTATTGAAC	6FAM - CTTGACCGAATAGCA
OMS00061	GSI	F - AAGTGGAGGCTGACCTGTTG	VIC - CATTGCCATTTACAGACTT
		R - GCTGATGGCACCTGACAGTTAATT	6FAM - TGCCATTTGCAGACTT
OMS00092	GSI	F - TCTCCAGGTGTATCTTGAGAAGGT	VIC - CAGCTGAGAATAGGTTC
		R - AGGGTTCACACAGGGAAGATATCAT	6FAM - AGCTGAGAAGAGGTTC
OMS00096	GSI	F - CATGAGAATGGATCAGTCTCCACAA	VIC - AAAGAGGAAGAGTCTCG
		R - GATGAAATCTGAATGTGTTGACACTACAG	6FAM - AAAGAGGAAGCGTCTCG
OMS00087	GSI	F - GCAAATTTCACCCTTAACGTGGTTT	VIC - CACACTTTGTCAGTTGTAAC
		R - GATTTGATGTGTGTGTGTATTACCTCCTCTA	6FAM - ACACTTTGTCAGCTGTAAC
Appendix A. Continued.			

SNP/Comments	Panel	Primers	Probes
OMS00119	GSI	F - AGCGGCAGTTGTGTTAATGAGA	VIC - CCACACAGCTGCCTGT
		R - CTTCCTAAAGCCTGACAGTCTGT	6FAM - CACACAGCAGCCTGT
OMS00129	GSI	F - GGAGATGATGAAATAAAAATTGAGGAAAAGATGA	VIC - TTGAACAACAAGAAAAA
		R - TGTCTGGTGAATTATCGCAAATAACCA	6FAM - TTGAACAACAACAAAAA
OMS00133	GSI	F - GACCACTTCACTCATTCCTCCTTTT	
0111000100	001	R - TCCGGTTTACACACTTCATGCA	6FAM - CGCCTCCATCTCTGTGGT
OMS00138	GSI		
0111000100	001		
01120	GSI		
010000140	001		6EAM - CAACTGTGCCTTTAGC
OMS00151	CSI		
0111000101	001		6EAM - ATGACCTCGATAATC
04500005	0.01		
01/1300093	631		
011500160	0.01		
01/1300109	631		
04600173	001		
01/1300173	651		
01/000/70	0.01		6FAM - ATTAGCTIGIGIGIGAACT
OMS00176	GSI		
0		R - CIGGGICCIGAAGGAGCII	6FAM - CCAGCCCIGCIGIC
Omy_impa1-55	GSI		VIC - CGAGATGATGCGTCTACA
•		R - ATTTTTCTTTGTTCAGTCTTCTGTCTC	6FAM - CGAGATGATGCATCTACA
Omy_103705-558	GSI	F - CTCCAATCGCAAATACCCAGACT	VIC - AGACTTACCCAGAGTGAGAG
_		R - CGCAGGAGACGGATGCC	6FAM - ACTTACCCAGGGTGAGAG
Omy_105075-162	GSI	F - GGAGAAGGACAAGGACATTGGTAAT	VIC - CTTTCTCTCCTACTTTCC
_		R - AAAGCAGACCACACCATACTTCTC	6FAM - CTTTCTCTCCTCCTTTCC
Omy_107031-704	GSI	F - GGCTTTCGGATACTGAGCAACAA	VIC - TGGACATGATTGCATAGAC
		R - TGAACTCACTGTTGGTATGGACTAGA	6FAM - CTGGACATGATTACATAGAC
Omy_107285-69	GSI	F - GCCCTTGTGACAATGCACTGTTATA	VIC - ATACGTTACTTTTGACCTTGT
		R - AGGTCTAGACAGTGTGCCATTTG	6FAM - ACGTTACTTTTCACCTTGT
Omy_110201-359	GSI	F - GGTAAGGCCTGTCTGACTATTTTGA	VIC - TTTGGCTATTGAAATTATACATT
		R - AGAGGTCAATGGATGCCAGTTT	6FAM - TTGGCTATTGAAATTCTACATT
Omy_CRBF1-1	GSI	F - AGTTCCGTACGGTAGCCTATTCTA	VIC - CAGAGTCGCCAAAAT
		R - CGCCCGGGTGAGAGTAATTG	6FAM - CCAGAGTCACCAAAAT
OMS00114	GSI	F - GGATGATGCTGTGAGTCGAGAAG	VIC - AAACGTTTCACATGCACC
		R - ACCTTCGCCACCCATGTTTTATT	6FAM - AAACGTTTCACCTGCACC
OMS00143	GSI	F - GGAGGCACGCCCCAAA	VIC - CCTGATCCAGAATCTAGA
		R - TTTGTTAAAATAGAGCCCTTAGTGGGTTT	6FAM - CCTGATCCAGAGTCTAGA
OMS00174	GSI	F - TGACTAACTATGCAGCCTGAAAGG	VIC - CAAGAACAGGATAAATGT
		R - GGGATACTCTTGTAATAAACTGTTGGTTAGTA	6FAM - AAGAACAGGAGAAATGT
Omy 97077-73	GSI	F - GTGTAAACAAAATGACTCTGGGATTCAG	VIC - TGGTGCAATAGAAATA
<b>y</b> =		R - AGAAGTGGCAATGGTGTGAAGTAT	6FAM - CATGGTGCAATAGTAATA
Omv 97865-196	GSI	F - TCCAGACTTCTGGTTTGTTCCATT	VIC - ATGAGCTTGTTAATTAAT
·		R - CCAGCCCCTATATTCACAATTAAGTGT	6FAM - AGCTTGTCAATTAAT
Omv 97954-618	GSI	F - GCTCTGCTTCCTCGGCAAATA	VIC - CAACGCTTACCGGTGTGT
	2.51	R - CACAATTGGTTTTTGCACAAAAGTAAAGTATT	6FAM - CAACGCTTACCAGTGTGT
Omv 128996-481	GSI	F - CTCATCCACACTGTACAGTACAAGT	VIC - CTTGTGGTTGAGGTTTG
· ····································	001	R - CATGCCTTCGTCTCATCAATAACAC	6FAM - TTGTGGTTGCGGTTTG
Appendix A. Continued.			
• •			

SNP/Comments	Panel	Primers	Probes
Omv aromat-280	GSI	F - CTCCATTGATTCATGCCGAACATT	VIC - TCTTGCAAACTCC
,		R - GGAGAGGTCAAACATAGCCTGGTA	6FAM - TCTTGCGAACTCC
Omv aspAT-123	GSI	F - GTTTGCCCATTTCACTGATGCT	VIC - CCTTCCTAGGCAGTCAG
····),		R - AGGAGACCACTCCAAAGAGAACT	6FAM - TTCCTGGGCAGTCAG
Omv b9-164	GSI	F - GCACAGAACACAGCCAATATTAACA	VIC - CCTACAACTTGATCTAACGTG
e, <u></u> e.	001	R - GCCTTGACTCTCCCTTCATGAC	6FAM - CCTACAACTTGATCTACGTG
Omv BAC-E5.284	GSI	F - ACAACGCCAACAACTTTCTCTTG	
o, <u></u>	001	R - CCTCATTTACTGTAGGACCATGCA	6FAM - ACAGTAGGACGGCAAG
Omv BAMBI2.312	GSI	F - CGAGCTCATGTCCGAAACTCAT	VIC - CCGAAAGTTCAACTTT
o, <u>j</u> , <u>_</u> ,	00.	R - TTTGACAGCCTCAACTTCTAGGG	6FAM - CCGAAAGTTAAACTTT
Omv. carban1-264	GSI	F - GCAAAGCCTCATCTTCAATCATTTGT	
eniy_eansann zer	001		6FAM - ATTAATATTGCTAATAACACTAAG
Omv cd59b-112	GSI		
	001		
Omy cin-172	691		
Only_Carrie	001		
$\Omega m v co x^2 - 335$	CSI		
0///y_cox2-330	001		
Omy 01 147	CSI		
Only_e1-147	631		
$\Omega_{mv} = a_{1} = 103$	CSI		
Onny_g1-103	631		
Omy C2PD 2 271	CSI		
0111y_G3FD_2-371	631		
Omv and $dA5$ 222	CSI		
Omy_gauu45-332	631		
$Omv adh_{271}$	CSI		
Omy_gan-z / i	631		
Omy ab 175	CSI		
Only_gil-475	001		
Omy CHSP-121	CSI		
only_onlor(121	001		
Omy 65047.86	691		
Omy_nsp47-80	631		
Omy hen 70 a Bro 220	601		
Omy_nsproar 10-329	631		
Omy 11 16 162	CSI		
Only_1210-103	001		
Omy ince 07	691		
Omy_mos-97	631		
Omy IDHP 1 i2	001		
	631		
Omy IDHR 2 of	CSI		
Only_LDI IB-2_e3	631		
Omy IDHP 2 is	001		
UIIIY_LUND-2_10	621		
0mi ( Inl 220			
Uniy_ipi-220	631	R - GTCCAGTCTTGCTTCAACTCAACTC	6FAM - AGTTACTCAGTCAGTCA 6FAM - AGTTACTCAGTCACAGTCA
Appendix A. Continued.			

SNP/Comments	Panel	Primers	Probes
Omy_mapK3-103	GSI	F - GAAGTCATTACTGGTCAGTGGTCAA	VIC - AATTATTAAGCCTATTTTTT
		R - GCACAAAACATGAGGAAAGTTGAGA	6FAM - ATTATTAAGCCTAATTTTT
Omy_mcsf-268	GSI	F - CCAGCATTCGTTCCCATTTCC	VIC - TGAGGGTTTATCTATTATTT
		R - CTTTTAATGTAGATTATATTCTTCTGTAGCCACTATGG	6FAM - AGGGTTTATCTGTTATTT
Omy_metB-138	GSI	F - TCTGTCCCTGACGCTATAAAAACG	VIC - TTCGCCAAAGAGAAAT
		R - GAAGTATTTCAGCTTAATTTCACTGTTGAGTT	6FAM - TTCGCCAAAGTGAAAT
Omy_myoD-178	GSI	F - TGGCAAAGCTGTCATTCCTTCTAAT	VIC - TTTTATGAGATATAATTTCC
		R - GGTCAAATATTTCATTTACGATTACACTTAGGC	6FAM - TTTTATGAGATATCATTTCC
Omy_nach-200	GSI	F - CTCATGAAAAACGGGAGAGCAAAG	VIC - AACTGACAGAGTCACAAC
		R - CAGCGGCTCTTCAGTAGTCT	6FAM - CTGACAGAGACACAAC
Omy_nxt2-273	GSI	F - CTTTAGAAAAGCCAAGGTATATTTTAACATACTTCT	VIC - ATCGACATTTACTGTGCCTT
		R - CTGCTGCCCTCTAATGGTAAGATAG	6FAM - ATCGACATTTACTATGCCTT
Omy_OmyP9-180	GSI	F - CTGGATGTGTAGTATCGGTGGAAAA	VIC - CTGTAGTAGTCCCCATTGT
		R - CACTGGGCACCTCTGATCTC	6FAM - CTGTAGTAGTCCGCATTGT
Omy_pad-196	GSI	F - CAAACAACCACAGTAGTCCTCCAAT	VIC - AAGACAAAGGTGTAATACC
		R - GCTTTTCACCCTTTTGTAAATTAAGCCAAA	6FAM - AAGACAAAGGTATAATACC
Omy_ppie-232	GSI	F - CTGTTTTAGATTAGAATGTTTTTGGTCAGGT	VIC - AAATAGCGGAGAAAAT
		R - CTGAACATAGGCTTTCATTTCAGACAT	6FAM - AAAATAGCAGAGAAAAT
Omy_ca050-64	GSI	F - GTCATACAGAACTGTTTTGTTGTGTCAA	VIC - CAGTTTGAAGAATATACTC
		R - ACCTTGAATTGGTTCCTAATGCTATTGT	6FAM - CAGTTTGAAGACTATACTC
Omy_sast-264	GSI	F - GAAGTAGGGTTTGTTGACCATGTGA	VIC - CTAGCCAATGCGTCTAA
		R - TGGATTCCATTTTAGGCTGTAATACATCTT	6FAM - ATCTAGCCAATGTGTCTAA
Omy_SECC22b-88	GSI	F - GGATCCCTCCTTTTAACACAAGACT	VIC - CTGTCTGTCCATATATC
		R - CTACAGGATGACTACCTAATTGCTAATAAAACA	6FAM - CTGTCTGTCCGTATATC
Omy_sSOD-1	GSI	F - GCCGGACCCCACTTCAA	VIC - CCACAACAAGACCC
		R - CAGACTAACCGAACAGCATCAGT	6FAM - CCACAACCAGACCC
Omy_star-206	GSI	F - CGTGTGCCAGCCCTTCT	VIC - TCTTTGGCACTATATCT
		R - GACCACTGAGATCATTGCTGTGA	6FAM - TTTGGCACCATATCT
Omy_sys1-188	GSI	F - CTTAAATGGTGCTGGTTGCTGTATT	VIC - AAACATGTACGACCTGTC
		R - AGTGATATCTTAGTGGGTCGAGGAAA	6FAM - TGTAAACATGTACTACCTGTC
Omy_tlr3-377	GSI	F - GTCGCTCCGGGTGCTT	VIC - CGTGATTAGGTTCTTC
		R - GGCCCAAACACTTCCTTCCT	6FAM - CGTGATTAGATTCTTC
Omy_tlr5-205	GSI	F - GAGCGTATCTGGTATGGTAACAACA	VIC - CAGTAATATTTCAGTGCCCG
		R - CTCCAGCAGCTTTAGAGAGTTTACA	6FAM - CAGTAATATTTCTGTGCCCG
Omy_hsf1b-241	GSI	F - AGCCCGAACTATCCTAAAGCATTTT	VIC - CAGTGTTTTGTTTTTGTCATT
		R - AAATCAATAGCTCAGAGAATAATGAACACCA	6FAM - AGTGTTTTGTTTTGTCATT
Omy_u07-79-166	GSI	F - CCCGCTATATTATTTGATCACCCTTGA	VIC - ACTTGGGAATACCCCAGCC
		R - ATTTAAATCCATTTCTAAAAATAAGCAAACCTAACCA	6FAM - CTTGGGAATAACCCAGCC
Omy_u09-52.284	GSI	F - TTTGTGTGTATTGTTGTGACTTG	VIC - ACTGCATTGTTGTAGCTAG
		R - TGATGTTATTGCAGGTCTAGCGAAA	6FAM - CTGCATTGTTGTCGCTAG
Omy_hus1-52	GSI	F - CTTGCCGGAGGGTAGCT	VIC - CCCATCCCTCCTCGG
		R - CCACAACTTCTCAAATGAATGGAATGT	6FAM - CCCATCCCTTCTCCTGG
Omy_u09-56.119	GSI	F - CCAAGGTGGACCCACCAG	VIC - AGTGAGCTGAAACAGAGCA
		R - GCTGAGTTTATAGGTCAGTCATTATACATATTGA	6FAM - TGAGCTGAAGCAGAGCA
Omy_nips-299	GSI	F - GACAGGATAGGAACGGTTTCTCAAT	VIC - CTGGATTTCACATGTAATAC
	<b>~</b>	R - AICAGAAGTTTAATTCAATATGTACACGATCCT	6FAM - CTGGATTTCACGTAATAC
Omy_U116_2-173	GSI		
Appendix A Continued		R - AUCIACTIUCIUTATCACATUTIUT	OFAMI - AGTTAAGTUUUTTATTGAUTG
, pponun n. conunuou.			

SNP/Comments	Panel	Primers	Probes
Omy_vamp5-303	GSI	F - CTGCTTCCCAATTCAGTATCGTCTT	VIC - TGGCCGTAGTAGTTGGTCA
		R - AGGCTGAAGCATTTCTGAGTATGAA	6FAM - TGGCCGTAGTTGGTCA
Omy_zg57-91	GSI	F - CACTCATACACTCACTCACAAAGGA	VIC - CACAGACTGCACAGCC
		R - AGCAGATAAGCCTTGTGAGTGAATC	6FAM - CCACAGACTTCACAGCC
Omy_ndk-152	GSI	F - AAGAATTGAGGGATAAAAACAAAATAATATATAAACATGA	VIC - CACCCACTTTCAAAAC
<i>y</i> =		R - CAAACCTACATTCATTAAAGTCCAGTTTTGT	6FAM - ACCCACTCTCAAAAC

SNP/Comments	Panel	Primers	Probes
Ots_SEXY1	GSI/PBT	F - CACAACATGAGCTCATGGG	
		R - CGATTAGAAAGGCCTGCTTG	6FAM - CCTACCAAGTACAGCCCCAA
Autosomal control	GSI/PBT	F - TCCTTGTGTCTAAAGGGCTTTGAG	VIC - CAGAATTAGCTTTGGACATT
control for Ots_SEXY1		R - GGGCTTGCTAGTCCTAAACAGATC	
Ots_128757-61R	GSI/PBT	F - CGTGTCCGGCTTCTTTATTTCATT	VIC - TTGTGCATTTTCCCC
		R - GATGGGTATGTTAATCATATTACCAGCGTAA	6FAM - TGTGCATTTCCCCC
Ots_lkaros-250	GSI/PBT	F - GAGGCTGACTTGGACTTTGC	VIC - ACAGAAGATTTTCGGCTGC
		R - GGCCTGTCAGCCAAGGA	6FAM - ACAGAAGATTTTCGACTGC
Ots_nkef-192	GSI/PBT	F - CATTTAGCAGACACTCTTATCTTAGTGTCA	VIC - AATAGGCCGACATCAA
		R - CGAATGTCCACCTCAGATGTTACAA	6FAM - AAATAGGCCAACATCAA
Ots_u07-07.161	GSI/PBT	F - GTCAACAAATGCAGGTAACATAAATGGT	VIC - ATCAGTGACATAAGTTGTCCA
		R - GATGCAAACACCTGTGAAATTGTGA	6FAM - TCAGTGACATAAATTGTCCA
Ots_u6-75	GSI/PBT	F - GAAAAAGTAAAGTAAAAGTAAAGTATTATACCACTAAAGACAAT	VIC - TTAGTCAACTGTTGTTTTT
		R - GATCCACACTGTTGGTCTACTACAA	6FAM - TTAGTCAACTGTTATTTTT
Ots_113242-216	GSI/PBT	F - GAGGCCTAATGTCTCTTGTGACT	VIC - ATTACCAACGGAGAACC
		R - GACATCTTCAACAAGTGTTCATTCACC	6FAM - TTACCAACAGAGAACC
Ots_CD59-2	GSI/PBT	F - TGTTTATCTCTGAGTGAAAAAGGTGTGT	VIC - CTAAAATGTCATGTAAATAT
		R - CATGTTACCCAGCTAAAAGTCTATAGCA	6FAM - ACTAAAATGTCATATAAATAT
Ots_GDH-81x	GSI/PBT	F - CTTTTCTGAATTAGTGCTGTGCTTGT	VIC - TGTTACGGGACATACT
		R - CCAACTTCTTCAACTCTGTCAGTGA	6FAM - TCTGTTACGGACATACT
Ots_IL8R_C8	GSI/PBT	F - CGTGGTGTTCGCCTTCCT	VIC - CTGGACGCCGTTACA
		R - TGTCGGCCATCACTGTCATG	6FAM - TGGACGCCATTACA
Ots_NOD1	GSI/PBT	F - GTGCTGCAGGAACCATGTG	VIC - CCAACGGCGACTTG
		R - CTGTGTGGACTGCTGTCTAAGG	6FAM - CCAACGCCGACTTG
Ots_SWS1op-182	GSI/PBT	F - TCAAAGACATCGAACACAAGAACGA	VIC - ATGTACTTTAACGATTCATTT
		R - GCAGGTAAATTCAAACGTCATCATAAGAA	6FAM - ATGTACTTTAACGTTTCATTT
Ots_u07-17.135	GSI/PBT	F - CTCGCCTCTGTCATTGTATTACCTT	VIC - AAAATGTACCACATACTTGT
		R - TGACACACGAGCCATTTTGATGAT	6FAM - AAATGTACCACATACTCGT
Ots_unk526	GSI/PBT	F - TCAAGACTGTGCTGTAGTTGTCTAC	VIC - CAACATTCCAGTCTGAAAC
		R - CCTCCCCCTTTTCCACATCAG	6FAM - CATTCCAGCCTGAAAC
Ots_105105-613	GSI/PBT	F - AGTACAAGTGCAGAGAATGACATCATG	VIC - CCGAGCTTGAGTTAGGA
		R - GGTGTTTTATTTTCCCATATATCTTTTAACTTTAAGCT	6FAM - CCGAGCTTGACTTAGGA
Ots_94857-232R	GSI/PBT	F - GGCACTCTCCCTGGCTAGA	VIC - CAGGATAATAACAAACAAG
		R - CCCCATCACTTCTCTGGCTTTAAAT	6FAM - CAGGATAATAACGAACAAG
Ots_GPH-318	GSI/PBT	F - GGTGATAACAGGTGTTGCACCAA	VIC - ATCAAGCTGACGAACCA
		R - TCAGGTGGTGGTGGACAAC	6FAM - CAAGCTGACAAACCA
Ots_mapK-3'-309	GSI/PBT	F - CGTGACCCTTGTAACTGAAAAGC	VIC - ATGCTATTAAATGAATATTC
		R - GGCCACTGTCATAGAATTAGGCATT	6FAM - ATGCTATTAAATGACTATTC
Ots_TAPBP	GSI/PBT	F - TTTCTCATCCTTCTCTCTCCAGTCT	VIC - CTGGACAGCTGGTCC
		R - GGACAAACCAGCACTCCAGAA	6FAM - CTGGACAACTGGTCC
Ots_u07-18.378	GSI/PBT	F - GGAAACCAGCTAGGATTCAGGAA	VIC - ATATGGTATGTAGAGGCTAGTTA
		R - CGTTATATGGTTTGCTTGTTTGCGATA	6FAM - TATGTAGAGGCAAGTTA
Ots_CD63	GSI	F - TGCATGTTTTCTAACTGTGTTTTTGTGT	VIC - AGATCATGGGAATCATAT
		R - TGAATGCCCCCCATCAACA	6FAM - ATCATGGGCATCATAT
Appendix B. Continued			

Appendix B. TaqmanTM assays used for *O. tshawytscha*. All forward and reverse primers are unlabeled and probes contain a 5' fluorophore and a 3' quencher and minor groove binder.

SNP/Comments	Panel	Primers	Probes
Ots_myo1a-384	GSI	F - CTCCCCCTGGACTTTGG	VIC - ACAGATCCATCCACCACT
_ ,		R - GCTCTATTGCACCGTGTTCTG	6FAM - AGATCCAGCCACCACT
Ots SL	GSI	F - AATATTGGCTTTCTGAGAATGCATTTGG	VIC - TCAAAGATATGATTCAATTAA
		R - CCAAGATACTTCCTTTAACTTCTCTGTCA	6FAM - AAGATATGGTTCAATTAA
Ots CRB211	GSI	F - CAACGCGGGAATGGCTTTTAA	VIC - CTACCGTACTGAACTC
0.0201.0211	001	R - GCCAGAGTCGCCAAAATAGTAGAAT	6EAM - CCGTACGGAACTC
Ots 113457-40R	GSI	F - CCCAAGTGGTGAGTGTCAGT	
	001		6EAM - CATATGGATTAGAGAATAG
Ots 97077-179R	GSI		
	001		
Ots GH2	GSI	$F_{-}$ CCGTACTGACCCTCGATGACA	
013_0112	001		
Oto    11	681		
	631		
Oto much 264			
018_111y0D-364	GSI		
	0.01		
Uts_PGK-54	GSI		
		R - CGACCCAAGTGGCTCATCAG	6FAM - CCACCATCATGCACTG
Ots_zP3b-215	GSI	F - IGCIGAGGACCATCIGCAATIC	VIC - CCAAATATCCTACCCGTGATG
0, ,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,		R - AGGICCAIGAAIAACIGAAAAIGIACAAGI	6FAM - CAAATATCCTACCAGTGATG
Ots_123048-521	GSI	F - CTCAACAGTGCACCTCCCTTAATT	VIC - TCACATCCAACTCAGTACT
		R - CCAAACACACCCTTCCATAATCTCT	6FAM - CATCCAACGCAGTACT
Ots_AldB1-122	GSI	F - GCCATGGAGGACTGGATGA	VIC - ACCCACTTCGCCAACA
		R - GCCACCACTACTTGCTGAGAAAATA	6FAM - ACCCACTTCACCAACA
Ots_EndoRB1-486	GSI	F - CCTTTGGGTCTGCTTGAGGTT	VIC - TCCTTCTCACGCTTCT
		R - GGAGCCAAATCCTAATGCTGAAGTA	6FAM - CTCCTTCTCATGCTTCT
Ots_GnRH-271	GSI	F - CAGATGAAAAATAAATAATTGGGCCATTAGGAA	VIC - CAATGAATACAATATCTAACCTAAT
		R - CAGAGAGACTGAGACCATATGATGTAGT	6FAM - AATGAATACAATATCTAATCTAAT
Ots_u07-53.133	GSI	F - AGCTAGGCTGTAAATGCAAGGAT	VIC - TAACACATGTTGGAGGTC
		R - CAGTGCTTTCAATTCATGCTGTCAA	6FAM - AACACATGTTAGAGGTC
Ots_ZR-575	GSI	F - GCCTACCAGAAAGTACCAATTGTGA	VIC - CCGACACAATTTTGT
		R - ACTTTTCACTGTCCTATTACAATTAGTATTTGTGATAT	6FAM - CCGACATAATTTTGT
Ots_GST-207	GSI	F - GGAGAACATGCATCACCATTCAAG	VIC - ATGAGAGAGTCTTTCTCTGTT
		R - TCAGCAAACGAAGGCTATGTAGAAT	6FAM - ATGAGAGAGTCTTTTTCTGTT
Ots_RAS1	GSI	F - TCATAAACATGGTGTCTTTCAGTCAGTT	VIC - CAATCTATCATCGACCAGC
		R - CTGACATGTGAAACTACTAAAGCATTTAATCAC	6FAM - CAATCTATCATCAACCAGC
Ots_aldb-177M	GSI	F - GCGATCAGGTGACGCTAAAATGA	VIC - CCAAATTGCTTAACCC
		R - AGGAAGGTGATGCCTGAGAGA	6FAM - CCAAATTGCTTTATCC
Ots EP-529	GSI	F - GCCCTGCCTGCAACTTC	VIC - CAGTGTCATTTTCGGC
_		R - GAAACCAACGTCTTGATGTAGACCTA	6FAM - ATCAGTGTCATCTTCGGC
Ots GPDH-338	GSI	F - CACTAAATATTCCTTATCATTTCATACTAAGTCTGAAGAA	VIC - CCACTACTTAACGTGCTTT
		R - AGCTGATACACAATCAAAACACAAAACAT	6FAM - CCACTACTTAACATGCTTT
Ots u07-57.120	GSI	F - GGTTTGAGCCAATCAGTTGTGTT	VIC - CAACCCCTACCTTGTCAC
		R - CGGTCTAATGTCCATTGCTCATGTT	6FAM - CCCCTACCATGTCAC
Ots TNF	GSI	F - CCAAATCCTCATCCCACACACT	VIC - CTGGCTGTAAACGAAGA
0.02.110	001	R - CCGTTGCACTTGACCCTAAAC	6FAM - TGGCTGTAAACAAAGA
Ots_nramp-321	GSI		VIC - TOGTTOATGCCCGTTAG
	001	R - GCATGCTCTGCAATACGTTGAG	6FAM - TCATTCATGCCCGT
Appendix B. Continued			

SNP/Comments	Panel	Primers	Probes
Ots_RFC2-558	GSI	F - GTAAGGTCTACTCCGGTTGTATTCG	VIC - TGCATGTAACAAATAACAT
		R - CAATACGACAGTACCGGTGTTAAACT	6FAM - TGCATGTAACATAACAT
Ots u202-161	GSI	F - CACTTTTGACTTTACATGGAACTTAACTCAT	VIC - ATTAGCTGCTAAGCACTAG
		R - GGGACTTCACTTTCTACAAACATGTCA	6FAM - ATTAGCTGCTATGCACTAG
Ots hsc71-5'-453	GSI		
	001	R - GTACGAAGTTGCGCCTTGTC	6FAM - TGAGGTGACAAAAT
Ots asnat-196	GSI		
Ols_dspail 190	001	Ρ - ΤΟ Ο ΑΛΟΤΟΑΤΟΑΟΛΟΛΟΛΟΛΟΟΛΟ	6EAM - CACACCCAGTCTTTAT
Oto EARSI A-220	GSI	$F_{-}$ CTTCCTCCCATTCTTCATCTTCAT	
OIS_FAN3LA-220	631		
Oto CST 275	001		
018_031-375	GSI		
Ota 114/0am 600	0.01		
Ots_LWSOP-638	GSI		
01- T- 1			6FAM - CAAGAAAGTTATACATTIC
Ots_Inst	GSI	F - GCCAATACGGGTTCTGAACTGT	VIC - IGCICCAGAICIC
<b>0</b>		R - CGGAATAGTCATAGTAGGGCTCGTT	6FAM - TGCTCCAGGTCTC
Ots_arp-436	GSI	F - GCCCTGGAGAAGTACGTTTTAAACTAA	VIC - CTAGGTGAAACTTTTTTAAA
		R - GCAACCATGTCAACATTGCACATAA	6FAM - CTAGGTGAAACTTTTTAAAAA
Ots_hsp27b-150	GSI	F - TAGGAGTTGGAAAGACTGCACA	VIC - YGATCTGGACCAGGCT
		R - CCCATTGGTTCTTTGGTGTT	6FAM - YGATTTGGACCAGGCT
Ots_u07-20.332	GSI	F - CGCGAGTTAGCTCGAATATTATGATTTC	VIC - ACCATTTGATATAACTGCGTTAG
		R - TCAAGCTAGCATAGCAACTTCATCAA	6FAM - CATTTGATATAACGGCGTTAG
Ots_96222-525	GSI	F - GCTCTTGCCCATCTGTAGGAT	VIC - TGTAGCTAATTTTAAGTTCTC
		R - GGCGCAACATATGTATTAAGCAACT	6FAM - AGCTAATTTTAAATTCTC
Ots_C3N3	GSI	F - CCGGATTCCATGGCCTACAC	VIC - CTAGAAAGGTTGATCCAATAA
mtDNA marker		R - GCCAAAATGATGTTCGGATGTAAAGT	6FAM - AAAGGTTGAGCCAATAA
Ots_FGF6A	GSI	F - TCAAAAATGTCTATCCAACAAATACTCTGAAAAATATTG	VIC - CACGATTAGCAATGAACAA
		R - CTTGTGCGCACCTTGCA	6FAM - CACGATTAGCAATTAACAA
Ots Ots311-101x	GSI	F - AAATGAGGCCGTCCTTTACACT	VIC - CTGAGATCACTTTGAGCAC
		R - GCAATACAAGCCCTTGATAATGAAGT	6FAM - ACTGAGATCACTGAGCAC
Ots Cath D141	GSI	F - CACTTGTTCTGCACACTACTTGTC	VIC - TGGGAAGCAATCAA
		R - CACACATGGATTTTGCCTGTCTAAA	6FAM - AATTGGGAAGCAGTCAA
Ots 1107-64.221	GSI	F - GAGGATGACACTGTCCGTTTGT	VIC - ATCGACCCTGTCATTAG
010_007 0 1122 1	00.	R - CACAGTCCTTCGTATTCACCTTGAT	6FAM - CGACCCTGTGATTAG
Ots Myc-366	GSI	F - CCTTAGCTGCTCTTTGAAGTTGACT	VIC - TCTCTCCTCATCTGTC
010_11/0 000	001		6FAM - CTCTGCTCGTCTGTC
Ots P450	GSI		VIC - CCCCGAAGTACTTT
013_1 400	001		6EAM - CCCGAAGAACTTT
	GSI		
013_00/17	001		
Oto 106747 220	001		
015_100747-239	GSI		
04- 04000 000			
Ots_94903-99R	GSI/PBT		
0	0.01/227	R - THIGGATCCAGCTCTCCGTATAGA	6FAM - ACAAACCAGAAAACAT
Uts_cox1-241	GSI/PBT		
		R - GTAAATGTAGTATACAGTATAGGCATCGTAGGT	6FAM - CACTACAGTAAGACCAT
Uts_G1H2B-550	GSI/PBT		
Annonalis D. Continued		R - CACAGGAAGGACGTGTTTTGATG	BRAM - ATGUTGUAUATGITAT
Appenaix B. Continued			
SNP/Comments	Panel	Primers	Probes
-----------------------	----------	---	---------------------------------
Ots_mapKpr-151	GSI/PBT	F - TGTTGTCTCGGACTGCATGAC	VIC - CATGCATTGCACATAC
_ , ,		R - GAAGGCACAGAGATGAAGGACAT	6FAM - CATGCAATGCACATAC
Ots Prl2	GSI/PBT	F - CCTGGTCTGTTTGTGATCAAGATG	VIC - ATGTATTGTTCATTTAATG
		R - GGTTAACTCAAATAGAACATACTCTGACACA	6FAM - TGTATTGTTCGTTTAATG
Ots TGFB	GSI/PBT	F - GCCTCACATTTTACTGATGTCACTTC	VIC - CTTCCGAGAGCTAGGCT
0.02.01.2	001/1 01	R - GAGCAGATCTCTTCAGTAGTGGTTT	6FAM - CTTCCGAGAACTAGGCT
Ots U07-25 325	GSI/PBT		
010_007 20.020	00// 01		6FAM - CGCTTGAAGGTTTGA
Ots 96500-180	GSI/PBT		
013_00000-100	00// 01		
Ote E2-275	CSI/DBT		
013_22-275	031/FB1		
Oto MUCI			
UIS_IMIHU I	GSI/PB1		
Uts_RAG3	GSI/PB1		
0, 07, 00,000	0.01/227	R - ACAGAATAAAGTATCTTCCTCTTACATCACTACTAAT	6FAM - CICIACAAIAIGAACIAIG
Ots_u07-49.290	GSI/PB1	F - GCIGAGGAAGGAIICIGIAIIIGCI	VIC - CTTTCCCCGTGTTGGT
_		R - TCGGACAGAGCGCATCC	6FAM - ACTTTCCCTGTGTTGGT
Ots_96899-357R	GSI/PBT	F - TCTCCTGAACTAATTTAGACCTCTGAATGT	VIC - CTGAATGTTTTTTTTTTTAATCTTT
		R - CCTCATATTGCTTTCATCTGAAGAGAGA	6FAM - CTGAATGTTTTTTTTTTTTTTTTT
Ots_hsc71-3'-488	GSI/PBT	F - TGCATCCATTCATACCTGACCAATT	VIC - TTTCCAATGGTATAGATATGA
		R - TTTGGTTAGGCACACGATAATTTGC	6FAM - TTTCCAATGATATAGATATGA
Ots_MHC2	GSI/PBT	F - GTCCTCAGCTGGGTCAAGAG	VIC - CTGGAGCGTTTCTGTA
		R - GTAGTGGAGAGCAGCGTTAGG	6FAM - CTGGAGCGTGTCTGTA
Ots_TLR3	GSI/PBT	F - TGCACCTGCGAGAGCAT	VIC - CTGTGGTTTGTGGCGTG
		R - CTGGCGTTTGTTCCGTTCAG	6FAM - CTGTGGTTTGTAGCGTG
Ots_102414-395	GSI/PBT	F - GCCTACTGATAAATGTATGACAGTAATGGA	VIC - CACATAGTGTAGCTTTACTAC
		R - CAATAACAAACAAGCTAGGAACAAAAGTGT	6FAM - CACATAGTGTAGCTCTACTAC
Ots ARNT	GSI/PBT	F - CCACTGGCTGTGGAGCTT	VIC - TACAGATGTCATTTTAC
_		R - GGGTTCAGTGATAGTTGGGCAAAT	6FAM - CTACAGATGTAATTTTAC
Ots ETIF1A	GSI/PBT	F - TCTGAACTCACCAAAGGAACACTTG	VIC - CAACTGAAGAAAATAATATG
_		R - GAGAGAAAAGGAGAAATGATTGCCATT	6FAM - CTGAAGAAAAGAATATG
Ots HSP90B-100	GSI/PBT	F - CACCTTAGTTCCACGCAACATG	VIC - TCTATGGTGTGATTCATT
	000.00	R - CTGCGTGTATTGTAGTGGTGACA	6FAM - TTCTATGGTGTGATTCATT
Ots myhn-85	GSI/PBT		
	00// 01		6FAM - AGCATGTAATTTTG
Ots P53	GSI/PBT	F - GGAACTTCCTCTCCCGTTCTG	
010_7 00	00// 01		
Oto 57-1	CSI/DBT	Ε - ΤΩΟΛΑΤΛΑΤΛΑΔΟΔΑΟΟΤΑΔΟΔΑΩΤΔΑΟΤ	
013_07-7	00// 01		
Oto 4211 95			
015_U2 1 1-05	GSI/PBT		
04- 4			
Uts_AShRS-60	GSI/PB1		
	0.01/007		6FAM - AGTUCUUGAUUAGU
Uts_FGF6B_1	GSI/PBT		
		R - GGGAGCCATGCACTAATATATTGGA	6FAM - CTGTTATCAGCCCCAAAT
Uts_IGF-1.1-76	GSI/PBT	F - GGTAGGCCGTCAGTGTAAAATAAGT	VIC - CTGCCTAGTTAAATAAAATA
		R - GATGGAGGCCACTGTGTTCTTA	6FAM - CTGCCTAGTTAAATTAAATA
Appendix B. Continued			

SNP/Comments	Panel	Primers	Probes
Ots_SClkF2R2-135	GSI/PBT	F - CCAAATACAGACCAGCTACTTGTGT	VIC - ATTCAAAGTCAAATTTT
_		R - CTTCAAGTCCCTGAATAATGGTACGT	6FAM - ATTCAAAGTCTAATTTT
Ots_u4-92	GSI/PBT	F - ATCCAAGGAGCCCCATTAAAGATTT	VIC - CTGTGTTGAATTTAACATAAT
_		R - CGTACCAGAGTTGTAGAAGCATCT	6FAM - TCTGTGTTGAATTTAACGTAAT
Ots 110064-383	GSI/PBT	F - AACAAAGAATGTTAAACACCAAACAGGAA	VIC - CTACGTAATGAACGTTAGCT
		R - GTGCAAGGGACCTAGCTAATCC	6FAM - ACGTAATGAACATTAGCT
Ots 100884-287	PBT	F - CGGAAGACCAGATTCTCCAAGAGTA	VIC - ATAGAACTACAATTCACATATAT
		R - CGACCAAGTAGCGGCACTT	6FAM - AACTACAATTCGCATATAT
Ots 101554-407	PBT	F - TGAAAGATATCAATTGTAGTAGTGGTGGTG	VIC - ATGGAGGATTGTGGTTGT
		R - ACACGCCAGTCCACAAGT	6FAM - ATGGAGGATTCTGGTTGT
Ots 101704-143	PBT	F - ACTTCTTGAGCCAATCGGATGATG	VIC - CTTAGACGTCAGAGGTC
		R - CCAGAGATAAACTAGTGGAGGAGATCA	6FAM - CTTAGACGTCCGAGGTC
Ots 102801-308	PBT	F - TGGGACAGAGGTGGGAATTGA	
0.02007.000	1.51	R - CCCAAAGATGCTTAACTGAAGATGTG	6FAM - AAGGGACAGTTTCTCAGACG
Ots 103122-180	PBT	F - CAAACGCGCACTCACACA	
013_100122 100	1 DT		
Ots 104415-88	DRT	$F_{-}$ CCTGACCATCCCAGTTGAACT	
013_104410-00	1 DT		
Oto 105122 200	DDT		
013_103132-200	FDI		
Oto 105385 431	ррт		
015_105365-421	PDI		
04- 405 407 447	DDT		
Ots_105407-117	PBI		
04- 400000 000	DDT		
Ots_108820-336	PBI		
04- 400505 040	DDT		6FAM - AATTGCCCATCTTAGAATA
Ots_109525-816	PBT		
0	<b>DDT</b>		6FAM - ATGAGGCATTCGGC
Ots_110201-363	PBT	F - GTTTGGCTATTGAAATTATACATTAAAACATGTAGCT	VIC - IGGAIGCCAGIIIIAAAA
0		R - CCATGGCATCCTGTAAAGAACAACA	6FAM - IGGAIGCCAGIIIAAAAA
Ots_110495-380	PBT	F - GCCTAGGTATGTACGAAACTTCACA	VIC - ATGGCCCCTGTCTATG
		R - AGGCTTTTTCAGATGGTCGTATGA	6FAM - AIGGCCCCIGIGIAIG
Ots_110551-64	PBT	F - GAGIGGICAAGGIIICAGIIICIG	VIC - ACGCTCGGAACATT
		R - GAAATGGACAGACACAAGGTCAAAC	6FAM - ACGCTCTGAACATT
Ots_110689-218	PBT	F - GTATAAACTAGAGTCCAGTGTTATGTTAATGTCTT	VIC - CACCAATCAATTAATTATT
		R - CATGGCAGACAACAGTAGAGAATATGA	6FAM - ACCAATCAATTCATTATT
Ots_112301-43	PBT	F - GCATGGCTGCCCTAGAACA	VIC - CGTCGCATTCAGC
		R - TCAGAACATTTCCTTCAGCTTCGT	6FAM - CGTCGCGTTCAGC
Ots_112419-131	PBT	F - GTGGGTAATCGATGCCAAAGAGAT	VIC - AAGCGACTTGATTATC
		R - TGGCAGTGTTTTCAACTAGCTTTG	6FAM - AGCGACATGATTATC
Ots_112820-284	PBT	F - CATAGATGTTTATATGAAAAACCTCCCACTGT	VIC - ACTCACACTCGAGTGACT
		R - GCATCCAAAAAGACGTGTGTGTTT	6FAM - ACTCACACTCAAGTGACT
Ots_112876-371	PBT	F - GCCTACAGCAAATTCAGCTACACAT	VIC - CATCACAACGATGTGTG
		R - TGGACCTTCAATCATCACAGCTT	6FAM - CACATCACAACTATGTGTG
Ots_115987-325	PBT	F - GGAGGTGTAGTGAAATGGGAAGAT	VIC - ATGCATAAAAGGTAATTGTG
		R - GCATTCAGTGAACCAGTAGTGCTAT	6FAM - ATGCATAAAAGGTCATTGTG
Ots_117432-409	PBT	F - TCATCAAAACATGCCTCTTCTGTGT	VIC - TTTAGACTTTGCTCTATAACAG
		R - TGTTGAACCTGTCACTCTGTCTTC	6FAM - ACTTTGCTCCATAACAG
Appendix B. Continued			

SNP/Comments	Panel	Primers	Probes
Ots_118205-61	PBT	F - CCATACAGCCAGTCCAGGTG	VIC - TAGTAGCCCCTACACCTC
		R - ACTGGACAGGGCTGGGT	6FAM - TAGCCCCTGCACCTC
Ots_118938-325	PBT	F - ATTTTCAAACAGGCATTTATCATTGGTGAA	VIC - AGAGATGCAAAGTGGAGTT
		R - GGTCTGTCCCTCATTCTTTGCA	6FAM - AGAGATGCAAAATGGAGTT
Ots_123921-111	PBT	F - TCGCTAGGCAGAAATATAGGGTTCT	VIC - TGCTAAATGGCATATATTAT
		R - GAGCATGGCGCTTGCA	6FAM - CTAAATGGCACATATTAT
Ots_124774-477	PBT	F - AGTTGTTCTTTTATATTGTGTTTTTATTCCATTCCA	VIC - CCACCGCCATCTGATA
		R - GCCAAATAAAAAACAAAGCATGAACACA	6FAM - CACCGCCGTCTGATA
Ots_129458-451	PBT	F - TGGGACCCACATAAAGCAACTG	VIC - CATCTGGCAATGCCTT
		R - GACATAAGACCCATTTAGCCCCTTTT	6FAM - CATCTGGCAGTGCCTT
Ots_brp16-64	PBT	F - ACTCTGGGTCCAGGAGGTTTT	VIC - AAGTCAGCATCTTTCA
		R - CTGACGAGACCATGCACCAA	6FAM - AGTCAGCGTCTTTCA
Ots_CirpA	PBT	F - GCTGTGATTGTGCTCTAAAGACATG	VIC - AATGCATTACAGAACTGA
		R - CTCCCACTTAGCATTCCTACCTT	6FAM - AATGCATTACAAAACTGA
Ots_Est740	PBT	F - GGACTCGTGCTTGAGGAAGATG	VIC - TCTGGATGGAACCGTTAG
		R - TGCATGGCTCCAACTCCTT	6FAM - CTGGATGGAGCCGTTAG
Ots_GCSH	PBT	F - GTTCTTTTTAATGATGACTACAGGTCTTTCAC	VIC - TATCTGGGCGGGCTG
		R - GCTACTTTACATAATACCATTTGAGCTGAGA	6FAM - CTATCTGGACGGGCTG
Ots_HMGB1-73	PBT	F - TGCTTCAGTGAAAATAAGCGTGAGA	VIC - ACTGTATATGTTACGTTTTC
		R - GTCGAGCGGTATGAATACTTTCTGA	6FAM - ACTGTATATGTTAAGTTTTC
Ots_NFYB-147	PBT	F - CCGTCCACAGCACAAGACTATAATA	VIC - TGTTCCAATGTAAAATGTATGC
		R - CAGATGATAGCTTCAGTAAGTGGTTCA	6FAM - TTCCAATGTAAAATATATGC
Ots_ntl-255	PBT	F - TGCAGTTACAAGCCTAAGACAATCT	VIC - TTGTAGAGGAAGAATATTC
		R - CAACTAAAGTAACACACCAGCAACTG	6FAM - TTGTAGAGGAAGTATATTC
Ots_OTALDBINT1-SNP1	PBT	F - CGCTGGGCATGGATGAGT	VIC - CTACTGTTGTATTTTCTC
		R - GGCCAACACTGCTACTTCCT	6FAM - CTGTTGTGTTTTCTC
Ots_OTDESMIN19-SNP1	PBT	F - GGTCTGTCTGTCTGTCTATCTGTCA	VIC - CCAGTCATGGGTCATT
		R - TGTGTGTCTTTGTTCATTCCTACCA	6FAM - TCCAGTCATTGGTCATT
Ots_OTSTF1-SNP1	PBT	F - CGGACAAAGAGCTACAGAAATGC	VIC - CCGCCACCTTGGCT
		R - CGTCCCTCTTCACGCATGA	6FAM - CGCCACATTGGCT
Ots_parp3-286	PBT	F - AGTCAGTGTTGGTGTAGTGAAGAGA	VIC - AGTTACAAGTGGTGTTTCA
		R - CATTTGTGGAGTGTTTATTGAACAGTAACA	6FAM - ACAAGTGGCGTTTCA
Ots_pigh-105	PBT	F - GTTTGGAATGTTTCTCTGATTGTGTTAACAA	VIC - TGACCTGAAAATATATATTTTT
		R - GCATTACTAAAAACTGGTGTGTGGAA	6FAM - ACCTGAAAATATATTTTTTT
Ots_pop5-96	PBT	F - CTCTTGCTACTTGCAGTGTATCTCA	VIC - TTCTGTTACTGGACTGATG
		R - AGTTTGAGGGCTCTATTCTGTCATG	6FAM - CTGTTACTGGGCTGATG
Ots_ppie-245	PBT	F - TGTTTTTGGTCATGTATTTTCTCTGCTATTTTT	VIC - ATGTCTGAAATGAAAGCC
		R - GGACTGGAGCTGCTGAACATA	6FAM - AATGTCTGAAATTAAAGCC
Ots_redd1-187	PBT	F - TTCTGGGTTGCCATACTCTTTCAAT	VIC - ATTCTGACAGCTGTTTTG
		R - AGTTGAGACCTTCAGTTCTTAGGGTAT	6FAM - CTGACAGCCGTTTTG
Ots_Thio	PBT	F - TTTTAAAAATGGAGATAAACTCCTGACCTGAA	VIC - CAGTGTATTAGTCATTCTTA
		R - AATACCAAACCATGCCACTAATACCT	6FAM - CAGTGTATTAGTCGTTCTTA
Ots_tpx2-125	PBT	F - TGTTGTAATCTTTCTGAATATTTGCTTGCTT	VIC - CAGGCGGTTCTCC
		R - TCTTCCAAATTGAGCACAAAAGCAT	6FAM - CAGGCAGTTCTCC
Ots_txnip-321	PBT	F - CCTTCAAACTAACACATCATAGACATGCTT	VIC - TCTGGCGGATTTACA
		R - TTATCAAACTGAAGGCGGATTTACTGA	6FAM - CTGGCGGGTTTACA
Ots_u1002-75	PBT	F - CCGCCTTTCCCACCTTCTC	VIC - ATGGCCCTTACACTATC
		R - TCAAACGAGAACACACTAAGGTTGT	6FAM - TGGCCCTTACGCTATC
Appendix B. Continued			

SNP/Comments	Panel	Primers	Probes
Ots_vatf-251	PBT	F - CTTTTCGGGTTATTCATGCTGTTGT	VIC - AGACCACAAGATACAGTACC
		R - GCAAGCATTTGAAAAACAGACTGGAT	6FAM - AGACCACAAGATAGTACC

## Prepared by:

## Approved by: IDAHO DEPARTMENT OF FISH AND GAME

Mike Ackerman Fisheries Research Biologist

Matt Campbell Genetics Laboratory Manager IDANG DELARTMENT OF HOHAND GA

Daniel J. Schill Fisheries Research Manager

Edward B. Schriever, Chief Bureau of Fisheries