Genetic Assessment of Columbia River Stocks

Table 1. Proposal Metadata

Project Number	2008-907-00
Proposer	Columbia River Inter-Tribal Fish Commission
Short Description	Genetic Assessment of Columbia River Stocks
Province(s)	Basinwide
Subbasin(s)	Basinwide
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Information transfer:

A. Abstract

This proposal combines four inter-related projects from the Fish & Wildlife Program Accords that address Single Nucleotide Polymorphism (SNP) Discovery, Genetic Baseline Expansion, Genetic Stock ID (GSI) to Evaluate Catch, and GSI of salmon and steelhead passing Bonneville Dam. These four projects are highly related since SNP markers are needed to complete species specific baselines, and these baselines are requisite to complete GSI. The specific objectives are 1) discover and evaluate SNP markers in salmon and steelhead; 2) expand and create genetic baselines for multiple species (Chinook, steelhead, sockeye, and coho); 3) implement GSI programs for mainstem Chinook fisheries and 4) GSI of fish passing Bonneville Dam (steelhead and Chinook). These four objectives address needs for distinguishing specific stocks, determining genetic diversity, stock specific run timing, and estimating stock composition to provide information for fisheries management and harvest. Newly discovered SNP markers may also be useful for other applications such as pedigree studies for estimating reproductive success, and evaluating adaptive divergence of populations to specific environments. The work related to objectives 1 & 2 will be completed by CRITFC and University of Idaho staff at the Hagerman Fish Culture Experiment Station. Tissue samples to address objective 3 will be collected by staff from CRITFC, WDFW, and ODFW. Genotyping and GSI analysis for objective 3 will be completed by CRITFC and University of Idaho staff in Hagerman, ID. Geneticists with CRITFC have adequate expertise to complete each objective, and have published peer-reviewed papers on each of these topics (Narum et al. 2007, Campbell and Narum 2008a, Narum et al. 2008a, Narum et al. 2008b).

B. Technical and/or scientific background

RESEARCH

In this proposal we plan to discover and evaluate single nucleotide polymorphism (SNP) markers to expand existing baselines for genetic stock identification (GSI) of salmon and steelhead in the Columbia River Basin. The scientific background for each objective is included below.

SNP Discovery

One of the highest priorities in the full-scale implementation of SNPs for salmon genetics is the discovery and development of a sufficient number of these markers to characterize population variability. These polymorphisms represent the most abundant variation in the genome of most organisms, and are spread throughout the entire genome at high density (Morin et al. 2004). Thus SNPs can be discovered through sequencing known regions of DNA and converted to high throughput assays (e.g., Campbell and Narum 2008a). Further, mutation rates, mutation models and error rates for SNPs are generally well understood, providing a foundation for estimating genetic divergence between populations. SNP markers also offer the potential of a more cost-effective and less error-prone alternative to existing genetic tools that may be used independently or in tandem with existing microsatellite markers to improve accuracy and precision of stock assignments. The combined power of these two marker types is expected to improve stock composition accuracy (Narum et al. 2008a) and allow researchers to meet rigorous stock composition and assessment needs for timely management of fisheries. Thus we plan to proceed with SNP discovery in multiple species of salmon and steelhead. These markers will be genotyped in populations throughout the Columbia River Basin to expand and/or create genetic baselines for multiple species.

Baseline Expansion

Currently, genetic baselines of microsatellite markers are in place for Chinook salmon across the coastwide range (Seeb et al. 2007) and steelhead in the interior Columbia River Basin. Despite large, representative sample sizes from many populations and very high microsatellite allelic diversity, the resolution of specific stocks and populations in these baselines is limited in some cases. For example, fall Chinook salmon in the Columbia River are closely related and remain difficult to distinguish even with a powerful set of 13 microsatellite markers. Several other closely related populations in the Chinook salmon baseline are similarly difficult to distinguish and thus have been pooled into a single reporting unit for GSI applications. In some cases (i.e., mainstem Columbia R. Chinook fisheries), a finer level of stock discrimination is necessary for management of fisheries. Additional SNP loci will increase stock assignment reliability where greater resolution is required. Given the difficulty and expense of inter-laboratory standardization, additional microsatellite markers may not the most efficient choice. In this regard, SNP markers are the preferred option for additional loci since they offer many beneficial characteristics that make them amenable to adding loci to existing baselines.

Since SNP assays involve direct interrogation of genetic sequence variation, SNP loci are good candidates for standardization among laboratories and expanding existing baselines. In addition, it is expected that the cost of SNP genotyping will drop significantly, as has happened in human genetics. In addition to ease of standardization and decreasing costs, SNP markers may provide a technique to effectively genotype poorly preserved tissue samples (i.e., carcass tissues). Degradation of DNA occurs over time, particularly in sub-optimal conditions, resulting in the fragmentation of the DNA into increasingly smaller pieces. Since SNP assays amplify a much shorter segment of DNA than microsatellites, SNPs have a greater probability of producing results in degraded samples (Campbell and Narum 2008b). While SNPs will be used to augment microsatellite baselines in most cases, there are some applications where SNP markers may produce the only viable baseline data for populations with only carcass samples available for genotyping. Furthermore, SNP development can focus on adaptive functional genes. The additive power of combining neutral (microsatellite) loci with adaptive loci like the Major Histocompatibility Complex (MHC) for stock identification was originally shown using the Canadian Department of Fisheries and Oceans (CDFO) baselines in sockeye and coho salmon (Beacham et al. 2001, 2004). Alaska Department of Fish & Game (ADFG) has also developed SNPs for MHC loci that have also been highly effective (Smith et al. 2005). Thus SNP genotypes will be added to existing microsatellite baselines (Chinook salmon and steelhead) to improve stock resolution in the Columbia River, and we will begin to construct baselines for species that lack genetic data (i.e., sockeye salmon).

Genetic Stock ID

Genetic Stock Identification (GSI) methods have proven to be effective in determining the proportion of stock origin in several mixed stock applications (Shaklee et al. 1999, Beacham et al. 2006, Narum et al. 2008b). This proposal includes two GSI projects that will utilize genetic baselines: 1) GSI to Evaluate Catch; and 2) GSI of fish passing Bonneville Dam.

This study will include GSI analysis of Chinook salmon collected from commercial, recreational, and tribal fisheries in the Columbia River. (Subsequent years of the study will include steelhead and coho fisheries as possible.) Implementation of GSI technology could make monitoring individual production units in mixed stock areas possible. Tissues will be sampled annually from fisheries with existing programs in place with Washington Department of Fish and Wildlife (WDFW) and Oregon Department of Fish and Wildlife (ODFW). We plan to genotype representative samples from fisheries of primary interest. Known origin coded-wire tag (CWT) fish will be used to provide a measure of accuracy for GSI estimates. The GSI estimates may help refine CWT based estimates of stock composition used in fishery management.

The second application of GSI analysis in this proposal includes sampling unknown origin salmon and steelhead at Bonneville Dam for genetic analysis. Samples will be collected over the entire length of the run on a weekly basis, and genetic baselines will be utilized to determine the stock composition of these runs. Few studies have been able to determine the extent of overlap among life history types of salmon and steelhead, but GSI of each life history type will allow us to determine the stock composition of the different runs through Bonneville Dam with greater accuracy than current methods. Population genetic methods and statistical assignment models have advanced dramatically in recent years, and estimating stock composition is now possible using either Bayesian or Maximum Likelihood methods (Anderson et al. 2008). Therefore, we plan to estimate stock composition of multiple species passing Bonneville Dam and provide this information on a timely basis to fisheries managers.

C. Rationale and significance to regional programs

This research proposal affects all stocks in the Columbia River Basin and therefore is considered a basinwide application. The four projects in this proposal address needs for distinguishing specific stocks, determining genetic diversity, stock specific run timing, and estimating stock composition for improved fisheries management and harvest. These needs have been identified in multiple "Reasonable and Prudent Alternatives" (RPA) in the BiOp:

-page 57, RPA No. 41, Preserve genetic resources
-page 69, RPA No. 50, Fish population status monitoring
-page 77, RPA No. 53, Monitor adult salmonids passing through FCRPS
-page 88, RPA No. 62, Fund selected harvest investigations (i.e., fifth bullet, Investigate the feasibility of genetic stock identification monitoring techniques).
-page 89, RPA No. 63, Monitor hatchery effectiveness
-page 89, RPA no. 64, Investigate hatchery critical uncertainties (i.e., Estimate relative reproductive success with genetic markers)

Other documentation that identifies the need for these projects include the Independent Scientific Advisory Board (ISAB) Report (2005), Columbia River Technical Advisory Committee (TAC), and in the 2000 Council Fish and Wildlife Program including:

-Basinwide Provisions – Strategies D. 5. Harvest: "Monitor inriver and ocean fisheries and routinely estimate stock composition and stock-specific abundance, escapement, catch, and age distribution. Expand monitoring programs as necessary to reduce critical uncertainties. Manage data so that it can be easily integrated and readily available in real time."
-Basinwide Provisions – Strategies D. 6. Hydrosystem: "…evaluation of escapement numbers to spawning grounds and hatcheries, research into water temperature effects on fish passage, and the connection between fish passage design and fish behavior."

D. Relationships to other projects

The genetic markers and baselines developed in this proposal will directly benefit both new and ongoing projects funded under the Accords and Fish & Wildlife Program. This includes the Kelt Reproductive Success and Snake River Kelt studies (200001700), and supplementation monitoring and evaluation projects (200852300). The GSI results from fish passing Bonneville Dam will be combined with PIT Tag results from migration studies (200851800). The sampling plan for Objective 3 (GSI to evaluate catch) will utilize input from "Power analysis to determine catch sampling rates" (200850800) and "Expanded tribal catch sampling" (200850200) .

Table 2. Relationship to existing projects

Funding Source	**Project #**	**Project Title**	**Relationship (brief)**
BPA	200851800	PIT Tags for migration montoring	Bonneville sampling effort and GSI results will be combined with PIT tag study.
BPA	200001700	Kelt Studies	Genetic markers and baselines will be used for pedigree and assignment purposes.
BPA	200852300	Supplementation monitoring evaluation	Genetic markers will be used for pedigree analysis.

Funding Source	Project #	Project Title	Relationship (brief)
BPA	200850800	Power analysis to determine catch sampling rates	Sampling design for Objective 3 will utilize input from this project.
BPA	200850200	Expanded tribal catch sampling	Samples from tribal fisheries will contribute to Objective 3.

E. Project history (for ongoing projects)

While the research projects in this proposal are newly funded under the Fish & Wildlife program, previously funded projects from the Pacific Salmon Commission and NOAAs FCRPS BiOp are applicable to these new projects. Most relevant, an existing coastwide microsatellite baseline for Chinook salmon will allow GSI analysis of fisheries, and an existing microsatellite baseline for interior Columbia River steelhead is in place for Bonneville GSI allowing these project to proceed immediately. The newly funded projects will allow us to build upon existing baselines by adding more genetic markers (SNPs) and provide more accurate and higher resolution GSI estimates for fisheries managers.