RFS# 015

Natural Reproductive Success and Demographic Effects of Hatchery-Origin Steelhead in Abernathy Creek, Washington:

Can newly-developed, native broodstocks of steelhead derived from captively-reared parr potentially contribute to recovery of naturally spawning populations?

Project Proposal to BPA to address NMFS *RPA Action 182*

April 11, 2003

Donald E. Campton[1]
William R. Ardren,
Gayle B. Zydlewski,
and
Patricia A. Crandell

U.S. Fish & Wildlife Service
Abernathy Fish Technology Center
1440 Abernathy Creek Road
Longview, WA 98632

[1] Contact person for response: Don_Campton@fws.gov; TEL: 360-425-6072; FAX: 360-636-1855

SUMMARY

We will investigate the natural reproductive success of hatchery-origin (HOR) and natural-origin (NOR) steelhead in Abernathy Creek (Cowlitz County), Washington. This proposed work responds directly to RPA Action No. 182 but is part of a larger project to assess the feasibility of developing new, native broodstocks of steelhead from captively-reared NOR juveniles (age 0+ parr). The long-term goal of this work is to assess whether captive rearing of NOR juveniles is a viable alternative to trapping NOR adults for developing new, native broodstocks of steelhead. Our research objectives include determining whether such broodstocks can assist with recovery of natural populations, as well as provide fish for harvest. The work will be conducted at the Abernathy Fish Technology Center (AFTC), a U.S. Fish & Wildlife Service (USFWS) research hatchery on Abernathy Creek, a tributary to the lower Columbia River approximately 10 km west of Longview, Washington. Two adjacent tributaries to the Columbia River, Germany and Mill Creeks, will serve as natural population "controls" for evaluating demographic responses to hatchery supplementation in Abernathy Creek. The work described here was initiated in 1999 and is continuing, but direct funding has not yet been secured. Beginning in the early fall of 1999, approximately 500 young-of-the-year (age 0+) NOR steelhead were collected from throughout a 15 km section of Abernathy Creek. Similar collections occurred in 2000 and 2001, respectively. Fish from those collections are currently being reared to sexual maturity and spawned. In the spring of 2002, 110 full-sib families were produced from 110 pairwise matings of captively-reared, age 3 fish that had been collected as parr from Abernathy Creek in 1999. Spawning occurred over a 17-week period (January-April) and yielded over 80,000 eyed eggs and fry. Those fish were subsequently culled to 30,000 fry and are scheduled for release from the AFTC as "F1" yearlings (smolts) during the spring of 2003. Similar F1 releases will occur in 2004 and 2005. Three-year old, F1 HOR adults are scheduled to first return to the AFTC during the winter and spring of 2005 (December 2004 – June 2005). Beginning in the late fall of 2004, all upstream-migrating adults will be trapped at the AFTC via a new, permanent electric weir. Equal numbers of NOR and F1 HOR adults will be passed upstream to spawn naturally after fin clips, scales, and bio-data (e.g. fork length, sex, etc.) are taken. A portion of the returning HOR adults (goal = 70 males and 70 females) and NOR adults (goal = 8 males and 8 females) will be retained for broodstock to continue the experiment and produce F2 hatchery-origin fish. A suite of at least eight, nuclear DNA microsatellite loci will be used to genotype all adult steelhead passed upstream and to reconstruct the pedigrees of all natural-origin fish subsequently trapped at the AFTC. This will allow us to determine the relative reproductive success of non-pedigreed natural-origin adults, F1 and F2 hatchery-origin adults, and natural-origin adults representing the F2 and F3 progeny of F1 and F2 hatchery-origin adults that spawned successfully in Abernathy Creek. Smolt traps operated by the Washington Department of Fish & Wildlife (WDFW) at the mouths of Abernathy, Germany, and Mill Creeks will provide time-series estimates of total smolt productivity of all anadromous fishes in each of the three streams. These latter data will allow us to compare smolt abundance trends in Abernathy Creek (treatment stream) to abundance trends in Germany and Mill creeks (control streams). The work proposed here has direct applications to upper Columbia, mid-Columbia, and Snake River steelhead stocks where non-native hatchery stocks may pose risks to ESA-listed, naturally spawning populations. This proposal is part of a long-term goal to establish Abernathy, Germany and Mill creeks as experimental treatment and control streams, respectively, for evaluating new hatchery methodologies that have systemwide applications. If funded, our proposed work will provide long-term monitoring and evaluation of steelhead, coho salmon, and cutthroat trout in three adjacent streams to the lower Columbia River, thus partially satisfying NMFS's RPA Action 183 to establish "Tier 3 M&E" for each ESU in the Columbia River.

PROJECT DESCRIPTION

I. PREVIOUSLY SUBMITTED PROPOSALS

Earlier versions of our proposal were submitted for BPA funding in FY2001 under the Innovative category (Project No. 22031) and subsequently under the Lower Columbia/Estuary Provincial Review (Project No. 30003) and again under the Mainstem/Systemwide Provincial Review (Project No. 35027). Previous versions of our proposal have received very positive reviews, both from the Independent Scientific Review Panel (ISRP) and the Columbia River Fish & Wildlife Authority (CBFWA). At the present time, our Mainstem/Systemwide proposal (Project No. 35027) is in the "**Urgent**" category for potential funding; however, as a new project, we do not expect it to be funded under that venue because of a shortage of funds. ISRP and CBFWA reviews of previous versions of our proposal are summarized in Appendix 1.

II. BACKGROUND AND MOTIVATION

A. Genetic Risks of Non-Native Hatchery Steelhead

Many hatchery programs for steelhead pose genetic or ecological risks to natural populations because those programs release or outplant fish from non-native stocks. The goal of many steelhead programs has been to simply provide "fishing opportunities" with little consideration given to conservation concerns. For example, the Washington Department of Fish and Wildlife has widely propagated and outplanted one stock of winter-run steelhead (*Chambers Creek* stock) and one stock of summer-run steelhead (*Skamania* stock) throughout western Washington. Biologists and managers now recognize potential negative effects can occur when non-native hatchery fish interact biologically with native populations (Hindar et al. 1991; Waples 1991; Campton 1995). Not only do non-native stocks pose genetic and ecological risks to naturally spawning populations, but non-native fish stray as returning adults at a much higher rate than do native fish (Quinn 1993).

Biologists and managers also recognize the need to (a) maintain the genetic resources associated with naturally spawning populations and (b) restore or recover natural populations wherever possible. As a consequence, the U.S. Fish & Wildlife Service (USFWS) and the National Marine Fisheries Service (NMFS), now *NOAA-Fisheries*, have been recommending a general policy that discourages the use of non-native hatchery stocks and encourages development of native broodstocks. There are two primary motivations for these recommendations: (1) reduce or minimize potential negative biological effects resulting from genetic or ecological interactions between HOR and NOR fish and (2) use native broodstocks as genetic repositories to potentially assist with recovery of naturally spawning populations.

B. NMFS 1998 Bi-Op on Artificial Propagation

A major motivation for the captive-rearing work described in this proposal resulted from NMFS's 1998 *Biological Opinion on Artificial Propagation in the Columbia River Basin*. In that *biological opinion* (BO), NMFS concluded that non-native hatchery stocks of steelhead jeopardize the continued existence of ESA-listed, naturally spawning populations in the Columbia River Basin. As a consequence of that BO, NMFS recommended - as a *reasonable and prudent alternative* (RPA) - that federal and state agencies phase out non-native broodstocks

of steelhead and replace them with native broodstocks. However, NMFS provided no guidance regarding how to achieve that RPA.

The development of native broodstocks of hatchery steelhead can potentially pose unacceptable biological risks to naturally spawning populations, particularly those that are already listed as *threatened* or *endangered* under the U.S. Endangered Species Act (ESA). The traditional method of initiating new hatchery broodstocks of anadromous salmonid fishes is by trapping adults during their upstream, spawning migration. However, removing natural-origin adults from ESA listed populations may not be biologically acceptable because such activities may further depress those populations via "broodstock mining". In addition, trapping adult steelhead may be logistically unfeasible in many subbasins because high water flows in the spring, when steelhead move upstream to spawn, will often "blow out" temporary weirs. Additional risks associated with trapping adults include *genetic founder effects* and difficulties meeting minimum, genetic effective number of breeders without "mining" the wild population to potential extinction. As a result, alternative methods for developing native broodstocks are highly desired.

C. Developing Native Broodstocks of Steelhead

One alternative for developing native broodstocks, particularly when the collection of adults is logistically unfeasible or biologically unacceptable, is the *captive rearing* to sexual maturity of natural-origin juveniles. In this latter approach, pre-smolt juveniles are collected from the streams or watershed for which a native broodstock is desired, and then those juveniles are raised to sexual maturity in a hatchery. Those hatchery-reared adults then become the broodstock source for gametes and initial progeny releases.

Such a *captive rearing* program offers many potential, genetic advantages over traditional adult-trapping programs for developing native broodstocks:

- Large numbers of juveniles can be collected from the wild with only minimal impacts to naturally spawning populations because juvenile (age 0+parr)-to-adult survivals are typically very small (<1%) under natural conditions.
- The genetic base of the broodstock (i.e. genetic effective population size) can be substantially larger for juveniles than adults because juveniles can theoretically represent the offspring of all adults that spawned successfully within a stream or watershed, as opposed to trapping only a small portion of returning adults for broodstock.
- Collecting juveniles for broodstock can substantially reduce the risk of genetically "swamping" naturally spawning populations with hatchery-origin fish (i.e. via a "Ryman-Laikre effect") as occurs when hatchery-released fish represent the progeny of a relatively small number of trapped adults (Ryman and Laikre 1991; Ryman et al. 1995).

D. Need for Experimental Supplementation Studies with Natural Population Controls

The ability of hatchery-origin adults to reproduce successfully and contribute genetically, via *supplementation*, to the recovery of naturally spawning populations is a major uncertainty confronting salmon comanagers in the Pacific Northwest. This question has been debated intensively throughout the Columbia River Basin for over 10 years but remains unresolved. Indeed, a major symposium was conducted in June, 2000 to specifically address these uncertainties and identify future research and comanager needs (IMST 2000).

Much of the uncertainty associated with supplementation is due to the absence of detailed studies on small, experimental scales with adequate control populations. Virtually all

evaluations to date have either been at large watershed levels (i.e. on a *production* scale) or have failed to incorporate appropriate *control* populations to which the supplemented populations could be directly compared (IMST 2000). Determining the relative reproductive success of individual hatchery- and natural-origin adults, coupled with monitoring demographic changes in population abundance with appropriate natural population controls, are critical to (a) determining whether supplementation is successful and (b) understanding *how* and/or *why* supplementation succeeded or failed. To achieve these latter objectives, nearly all upstream-migrating adults need to be genotyped with a battery of DNA markers (e.g. at 10-15 microsatellite nuclear DNA loci), and then samples of their naturally-produced offspring (e.g. parr or smolts) similarly genotyped to identify, via *maximum likelihood* and *assignment probabilities*, the specific male and female parents of each juvenile fish. Appropriate streams for these types of experimental studies are extremely difficult to locate. A basin-wide need exists to identify such experimental streams where the parents of naturally-produced progeny can by identified genetically and supplementation evaluated in a controlled, scientific manner. Performing such assessments in the lower Columbia River below Bonneville Dam offers the added advantage of maximizing adult return rates and, thus, the *statistical power* and *efficiency* of detecting true differences between hatchery-origin and natural-origin fish.

E. Abernathy Fish Technology Center and Conservation Genetics Laboratory

The U.S. Fish & Wildlife Service's Abernathy Fish Technology Center (AFTC) offers a unique opportunity to investigate new hatchery methodologies designed to assist with recovery of naturally spawning populations via natural population supplementation. The AFTC is located at stream mile 3.4 on Abernathy Creek, a tributary to the lower Columbia River, approximately 10 miles west of Longview, Washington (Fig. 1). The AFTC was originally built as a "mitigation" hatchery in the 1950's, and from 1961 through 1997, non-native *tule* fall chinook salmon were propagated and released as part of the Mitchell Act mitigation in the Columbia River. However, this propagation program and associated mitigation responsibilities have been terminated. The AFTC has been refocusing into a research hatchery and science center to investigate new propagation strategies and technologies that can contribute to the recovery of anadromous fishes in the Columbia River Basin. The AFTC is equipped with all the components necessary for a self-sufficient salmon/steelhead hatchery, including an electric weir and holding pond for trapping and enumerating upstream-migrating adults. The existence of such a facility devoted strictly to research is unique among hatcheries in the Columbia River Basin.

We have recently constructed a new, 1500 square foot building at the AFTC to house a new ***Conservation Genetics Laboratory*** for the Pacific Region of the U.S. Fish & Wildlife Service. Development of this laboratory is complete with respect to acquisition of key personnel (e.g. W.R. Ardren) and "state-of-the-art" laboratory equipment for collecting DNA genotypic data on natural and cultured populations of fish. Among other tasks, the laboratory is designed to potentially serve a "rapid response" function in support of the fish genetic information needs of the USFWS. One of our principal mandates is to assess genetic contributions of hatchery-origin fish to naturally spawning populations and determine whether HOR fish can assist with conservation goals, or whether such fish pose unacceptable risks to naturally spawning populations.

F. Abernathy, Germany, and Mill creeks

Abernathy Creek is an outstanding stream to address the natural reproductive success of HOR and NOR steelhead (Fig. 1). Abernathy Creek is a small, 3rd order stream with a drainage area of approximately 110 km^2 that is particularly amenable to intensive, scientific study of salmonid fishes. Like most streams in the lower Columbia River, Abernathy Creek historically supported abundant runs of steelhead and coho salmon. Other native species include coastal cutthroat trout (*O. clarki clarki*), which do migrate past the AFTC, and chum salmon (*O. keta*), which spawn primarily in the lower mile of Abernathy Creek. WDFW considers the steelhead population in Abernathy Creek to be "depressed but stable" with an estimate of only 100-150 adult spawners per year based on expanded redd counts (Dan Rawding, WDFW, pers. comm.). Historical estimates from the early 1950's exceed 500 fish based on fishway counts immediately upstream from the current site of the AFTC (WDFW, unpublished data).

One of the major advantages of Abernathy Creek as a "test" or "treatment" stream is the presence of two neighboring streams, Germany and Mill creeks, that can serve as natural population controls for assessing demographic responses (i.e. in Abernathy Creek). All three streams are approximately the same drainage area (91-110 km^2) and enter the Columbia River within 3-4 miles of each other: Germany Creek enters the Columbia River approximately two miles upstream and Mill Creek enters the Columbia River approximately one mile downstream, respectively, from the mouth of Abernathy Creek.

Lastly, Abernathy Creek and the two adjacent streams are located approximately 100 miles downstream from Bonneville Dam in a rural area of Cowlitz County. The opportunity of the AFTC to test new culture and recovery strategies in a natural stream without the constraints imposed by dams, mitigation responsibilities, and urban impacts is unique among artificial propagation facilities in the Pacific Northwest.

G. Tier 3 Monitoring and Evaluation Site (NMFS RPA No. 183)

As part of the proposal described here, we request that NMFS and BPA designate Abernathy, Germany and Mill creeks collectively as a *Tier 3 Monitoring and Evaluation Site* (NMFS *RPA No. 183*) for Lower Columbia and Southwest Washington ESUs of steelhead, coho salmon, chinook salmon, and coastal cutthroat trout. We believe the unique opportunities to conduct experimental studies with control and treatment streams - along with the existing facilities and previous commitments by the USFWS and WDFW - warrant serious consideration by NMFS, the Northwest Power Planning Council, and the Bonneville Power Administration. If funding can be secured for this long-term commitment, then we envision the opportunity for many additional studies with other collaborators (CRITFC, USGS-BRD, universities, etc.), not just the specific study proposed here. Funding from BPA would ensure continued smolt monitoring and additional monitoring of upstream migrating adults in Abernathy Creek.

III. PRELIMINARY WORK TO DATE

A. Native Broodstock Development

We initiated the work associated with this proposal in 1999. As noted above, we initiated this work in response to the 1998 NMFS Biological Opinion on Artificial Propagation in the Columbia River and the need to develop new, native broodstocks of steelhead without posing additional risks to naturally spawning populations. During the last week of September 1999, we

used a backpack electrofisher to collect approximately 500 young-of-the-year (age 0+) NOR steelhead parr from throughout a 7-mile stretch of Abernathy Creek upstream from the AFTC. We identified five sample sites (stream miles 3.4 to 10.0) approximately 1 mile apart each. Approximately 100 age 0+ parr were collected from each of those five stream sections to maximize their genetic diversity and number of parents potentially contributing to the fish we collected. Those fish were transferred to an 8x80 foot raceway supplied with Abernathy Creek water, covers, and automatic feeders. Similar collections occurred in the early fall of 2000 and 2001, respectively.

Mortalities associated with these initial collections have been extremely low; we have observed less than 10 dead fish during the first 24 hours after collection during each of the three years we electrofished Abernathy Creek.

We initially had difficulty getting the BY1999 wild-caught parr to start feeding on artificial feeds when those fish were first transferred to a raceway in September, 1999. We were able to stimulate their conversion to artificial feeds by coating the feed pellets with a krill hydrolysate. Those oils exude into the water, stimulating olfactory and feeding responses of the fish. We repeated the krill oil treatment for fish collected in the fall of 2001 and 2002 with similar results.

During the winter and spring of 2002, we produced 110 full-sib families from 110 pairwise matings (1 male x 1 female) of the captively-reared, age 3 fish that had been collected as parr from Abernathy Creek in 1999. Spawning occurred over a 17-week period (January-April). Each full-sib family was culled to a maximum of 1,000 eyed eggs or fry. Nevertheless, we ended up with over 80,000 F1, BY2002 fry. Those fish were subsequently culled to 30,000 fry and are scheduled for release from the AFTC as "F1" yearlings (smolts) between April 15 and May 15, 2003. Similar F1 releases will occur in 2004 and 2005 except 20,000 smolts will be released. All but 1,000 BY2002 fish (n = 29,000) were given coded-wire tags but no fin clips. The remaining 1,000 fish were given 23mm, long-range PIT tags for detection by our remote sensing antennas in Abernathy Creek (see section below on PIT tag detection).

Sexually mature fish collected as parr in the fall of 2000 are currently being spawned (January-April 2003) as three year old fish. Procedures are similar to those employed during spawning of BY1999 fish in 2002 except each full-sib family is being culled to only 350 eyed eggs. Such culling minimizes the variance in family size and maximizes the genetic diversity of the progeny by collecting progeny from all possible parents. Because of space constraints at the AFTC, our goal is to rear 20,000 F1-BY2003 fish (instead of 30,000 fish) to the yearling stage prior to release in the spring of 2004. BY2000 fish not sexually mature in 2003 will be retained for potential use as broodstock in 2004. We expect to follow procedures similar to those described above for BY2000 fish in future years. Three-year old adults from our releases are expected to first return to the AFTC during the winter and spring of 2005 (December 2004 – June 2005).

B. Smolt Monitoring in Abernathy, Germany, and Mill Creeks

WDFW initiated monitoring of smolt production in Abernathy, Germany, and Mill creeks in the spring of 2001. Smolt monitoring was repeated in 2002 and is scheduled to continue in 2003. Trapping began in early April and continued through mid-June (Appendix 2). Traps were located as near the mouth as logistically feasible. In 2001, fence panel traps were installed on Germany and Mill Creeks (RM 0.3 and 0.2, respectively), and a rotary screw trap was installed on Abernathy Creek (RM 0.3). The panel traps in Germany and Mill creeks were replaced with

screw traps in 2002. The estimated total number of outmigrating steelhead, coho, and cutthroat trout smolts for the three streams are shown below (Dave Seilor, WDFW, pers. comm.). Data for Germany and Mill Creeks in 2002 have not yet been provided.

Stream/year	Steelhead	Coho	Cutthroat
Abernathy Cr.			
2001	10,590	6,991	716
2002	5,400	6,200	1,650
Germany Cr.			
2001	7,650	8,157	353
2002			
Mill Creek			
2001	1,710	6,324	522
2002			

These three streams were chosen for smolt monitoring because of their close proximity to each other, their similarity in size, and the potential for long-term monitoring of anadromous salmonid fishes in a "semi-replicated" experimental design (Dave Seilor, WDFW, pers. comm.). Selection of these three streams was also motivated by the research activities proposed here (Dan Rawding, WDFW, pers. comm.) and for which we have been attempting to secure direct funding. Smolt trapping by WDFW is scheduled to continue in 2003, but those traps and crew will be reassigned to other streams in 2004 unless funding can be secured for this project.

C. Instream Monitoring and Evaluation

Streamwidth PIT tag Interrogation Systems (SPIS) were developed at AFTC under **BPA Innovative Project # 2001-012-00**. Two SPIS operate continuously for monitoring movement, migration, and survival of PIT tagged fish in Abernathy Creek. To date these techniques have been used to estimate smolt production of three naturally-produced salmonid populations (Zydlewski et al. 2003). Approximately 1,600 naturally-produced juvenile (< 100 mm FL) salmonids (steelhead trout, coho salmon, and cutthroat trout) are PIT tagged annually at 9 sites located upstream of AFTC. Annually over 390,000 detections are recorded between the months of October and July. Detection efficiencies are site dependent, but overall detection efficiency for the creek is 97% with 95% confidence intervals of 91 – 100%. Fish from all tagging locations are detected at the SPIS. Steelhead and cutthroat trout are primarily detected moving in the spring (April – June) coincident with the anticipated smolt migration. Steelhead are also detected moving past SPIS at lower numbers in the Fall and Winter. Travel time between SPIS (downstream movement) is highly dependent on time of year. Travel time in the spring was significantly faster (34.4 ± 7.0 hours) for all species than during any other time of year (763.1 ± 267.0 hours). Steelhead and cutthroat migrating in the spring are the same age as those that did not migrate in the spring. Peak of steelhead migration recorded at the two SPIS was 5/11 and 5/12 and the peak in the screw trap was recorded on 5/17 (see Appendix 2). Steelhead smolt production estimates using SPIS (3,802 with 95% confidence intervals of 3,440 - 4,245) is similar to those using more standard screw trap methods (approximately 5,400).

A backpack PIT tag detector has been developed and is used as another remote "recapture" method for additional accuracy in estimating population survival and recapture probability. This unit is used with an approximate efficiency of 24% to survey the creek after the spring migration. We are currently monitoring habitat use by naturally-produced and hatchery-produced steelhead trout with this detection system. Steelhead from a representative creek site below AFTC has been electrofished and all naturally-produced steelhead have been PIT tagged. Also, this site has been characterized at the macrohabitat level using transects. This spring PIT tagged steelhead produced from our native broodstock will be released from AFTC. When naturally-produced or hatchery-produced fish are detected in the site their microhabitat will be characterized. This will allow us to compare fish detection sites to mean values throughout the reach (indicating habitat selection) and the degree of natural-origin fish displacement.

All PIT tag data are also used to calculate survival and recapture probabilities using the Cormack-Jolly-Seber population model. Survival for naturally-produced steelhead is high and recapture probability depends greatly on season. Probability of recapture is highest in Spring (29.5%) and relatively low in all other seasons (< 7% in Fall, Winter, and Summer). Wild steelhead PIT tagged in the field and returned to the laboratory have a tag retention rate of 97.6%. Laboratory studies have revealed that survival from surgical implantation of 23 mm PIT tags is > 98% for coho salmon and steelhead trout. Retention of 23 mm PIT tags was 100% for coho salmon and 89% for steelhead. Life history characteristics quantified with SPI techniques are comparable to standard techniques. For example, peaks of spring migration for steelhead and cutthroat were amazingly similar to those reported from the screw trap (see Appendix 2). These techniques enable application of less laborious methods which are more accurate at estimating life history parameters.

IV. PROPOSAL OBJECTIVES, TASKS, METHODS

Objective 1. Develop a "native" or "local" hatchery broodstock of steelhead by captively rearing, to sexual maturity, natural-origin juveniles from Abernathy Creek and evaluate this approach as a potential alternative to trapping natural-origin adults for developing "native" or new "local" broodstocks.

As noted previously, work towards this objective was initiated in 1999 with the collection of approximately 500 age 0+ steelhead from Abernathy Creek. Similar collections were repeated in 2000 and 2001. Three-year old HOR adults from our on-site releases are expected to first return to the AFTC during the winter and spring of ~~2003~~ 2005 (December ~~2002~~ 2004 – May ~~2003~~ 2005). Returning F1 HOR adults will be spawned with wild fish in approximately a 9:1 HOR:NOR ratio to produce a hatchery F2 generation.

Table 1 provides a schematic diagram of the relationships between parental generation brood years and progeny generation brood years. Our goal is to release 20,000 HOR steelhead yearlings each year beginning in 2003 and continuing through 2012. Adult returns and natural spawning success of HOR and NOR steelhead in Abernathy Creek will be assessed through 2016, the final year in which age 5, F3 NOR offspring of F2 HOR adults are expected to return (Table 1). Details regarding tasks and methods associated with this Objective 1 are presented in Appendix 3.

Objective 2. Monitor smolt outmigration in Abernathy, Germany and Mill creeks, and estimate total natural production of steelhead, coho, and cutthroat trout smolts in each stream.

All smolt outmigration monitoring will be conducted by subcontract to the Washington Department of Fish & Wildlife (Dave Seilor, WDFW). A rotary screw trap was used in Abernathy Creek in 2001, while fence panel traps were used in Germany and Mill creeks during that first year. Those panel traps were replaced with rotary screw traps in 2002. Rotary screw traps will be used in all three streams in 2003 and subsequent years if this proposal is funded.[2]

Task 2a: Estimate the daily and total number of outmigrating smolts of each species in each stream each year. Total smolt outmigration of steelhead, coho salmon, and cutthroat trout in the three streams will be estimated by standard methods. A portion of the smolts trapped each day will be marked with a dye and released upstream to generate a simple Peterson estimate of the total smolt outmigration for each species in each stream on each day.

Task 2b: Determine whether the release of HOR steelhead yearlings and natural spawning supplementation of returning adults affects the total natural productivity of steelhead, coho salmon, and/or cutthroat trout in Abernathy Creek (treatment stream) relative to Germany and Mill creeks (control streams). We will use time series analyses of the estimated number of smolts produced each year in the three streams to determine if temporal changes in abundance of smolts produced in Abernathy Creek (treatment) differ significantly from temporal changes in abundance in the other two streams (controls). This task ties directly to the need to understand demographic effects on overall abundance associated with hatchery-origin fish spawning naturally in Abernathy Creek (see Objective 6)

Objective 3. Monitor and evaluate potential residualism by HOR steelhead juveniles released from the AFTC. We expect a significant proportion (10-20%, or more) of steelhead yearlings released from the AFTC to residualize (i.e. fail to outmigrate) in Abernathy Creek. We will assess this residualism by two methods.

Task 3a: Use the *Streamwidth PIT-tag Interrogation System* (SPIS) installed in Abernathy Creek (BPA Project No. 2001-012-00) to estimate the proportion of residualized steelhead yearlings. We will estimate and monitor potential residualism via PIT tag detectors at a permanent antenna installed approximately 1.4 km downstream from the AFTC. One thousand (1,000) yearlings will be given 23 mm long-range PIT tags each year prior to release. Percent of released fish failing to outmigrate past this downstream antenna will be estimated after correcting for percent PIT tag loss estimated from corollary tag retention studies. A backpack PIT tag detector (see Background

[2] Smolt trapping and monitoring in Abernathy, Germany, and Mill Creeks are scheduled to be discontinued after the spring of 2003 (Craig Burly, Regional Biologist, WDFW) unless external funding can be secured to continue those operations.

section) will be used also to scan lower Abernathy Creek for residualized steelhead released from the AFTC.

All PIT tag data will be used to calculate survival and recapture probabilities using the Cormack-Jolly-Seber population model. These methods are being used currently to collect baseline data on the natural steelhead population in Abernathy Creek prior to assessing the impacts of released hatchery-origin steelhead on NOR steelhead at the individual and population levels.

Task 3b: Estimate residualism and ecological impacts by snorkel surveys. Snorkel surveys of residualized steelhead and potential ecological impacts on natural populations will be assessed. This work will conducted by Stephen Riley, a stream ecologist for NMFS, by subcontract. Yearling HOR steelhead will be distinguished from NOR steelhead by the presence of a CWT (scanned electronically with a hand-held wand), AD-clip, and or "stubbed dorsal fin." Subsamples of residualized steelhead captured during snorkel surveys will be sacrificed to examine stomach contents and determine potential predation on subyearling, NOR salmonids.

Objective 4. Monitor upstream migration and adult returns of steelhead, coho salmon, and cutthroat trout past the AFTC in Abernathy Creek.

In previous proposals (Appendix 1), we requested funds to replace our existing electric weir with a "state-of-the-art" electric barrier system manufactured by Smith-Root, Inc. of Vancouver, Washington. The U.S. Fish & Wildlife Service is now planning to replace the existing weir in 2004 with a Smith-Root (patented) electric barrier system. Surveying and permitting are scheduled for the summer of 2003 (FY2003) with construction scheduled to occur during the summer of 2004. The new electric barrier weir will thus be in place before fish from our first hatchery releases are scheduled to return to the AFTC. This new electric barrier system creates a pulsed DC (direct current) field in which the field strength increases proportionally to flows and water level, thus allowing all upstream-migrating fish to be diverted into an adult holding pond over widely varying stream flows without fear of weir "blow-out". Similar electric barrier systems are installed at the Quilcene National Fish Hatchery (Hood Canal), Quinault National Fish Hatchery (near Lake Quinault), and Eagle Creek National Fish Hatchery near Clackamas, Oregon.

Task 4a. Determine the total number of NOR adult steelhead, coho salmon, and cutthroat trout migrating upstream each year past the AFTC. We plan to operate our electric weir from approximately September 1 through June 30 each year to trap and enumerate all adult steelhead, coho salmon, and cutthroat trout. Information about coho salmon is particularly important because these fish may soon be listed as threatened or endangered in the Columbia River. However, we do not know if NOR adults or HOR adult strays from a nearby hatchery (Fig. 1) are responsible for the NOR smolts produced in Abernathy Creek each year (see Table on page 8).

Task 4b. Estimate smolt-to-adult return rates and total number of adult returns of HOR steelhead released from the Abernathy Fish Technology Center. All adult steelhead trapped at the AFTC will be enumerated and scanned for the presence of a

coded-wire tag (CWT) with a hand-held electronic wand. HOR steelhead will be distinguished from NOR steelhead by the presence of a CWT, AD-clip, and/or stubbed dorsal fin. Length, scales, and fin tissue for DNA analyses will be removed from all adult steelhead (HOR and NOR) trapped at the AFTC.

Task 4c. Pass equal numbers of NOR and HOR adult steelhead upstream of the AFTC to spawn naturally in Abernathy Creek. We plan to pass all NOR steelhead upstream of the AFTC except for 10 males and 10 females retained for broodstock (see Objective 5). We will pass an equal number of HOR adults upstream to spawn naturally with NOR steelhead in Abernathy Creek upstream of the AFTC. We expect to trap 100-300 NOR adults each year based on expanded redd counts during the past 10 years (Dan Rawding, WDFW, pers. comm.).

Objective 5: Trap and spawn returning HOR and NOR adult steelhead for broodstock to produce F2 and F3 HOR progeny for release into Abernathy Creek.

The generalized broodstock spawning plan is summarized in Table 1. Parental generation fish are captively-reared adults collected from Abernathy Creek as age 0+ NOR parr in 1999, 2000, and 2001. Their F1 HOR progeny will be released into Abernathy Creek as yearlings in 2003, 2004, and 2005. Those F1 HOR progeny are expected to return to Abernathy Creek primarily as age 3 and age 4 fish in the years 2005 through 2008. Those returning adults would subsequently be spawned to produce F2 HOR fish. Similar natural reproduction would be occurring in Abernathy Creek among NOR and HOR adults passed upstream to spawn naturally. The ultimate goal is to evaluate the relative natural reproductive success of NOR adults, F1 HOR adults, F2 HOR adults, and F2 NOR adults that are the NOR progeny of F1 HOR adults that spawned successfully in Abernathy Creek.

Task 5a: Spawn sufficient numbers of adults each year to produce 20,000 yearlings for release into Abernathy Creek. Beginning in 2005 when HOR adults are expected to first return to the AFTC, we plan to spawn a minimum of 70 HOR males and 70 HOR females each year. We plan also to retain a minimum of 8 NOR males and 8 NOR females so that progeny fish each year are derived genetically from at least 10% natural-origin adults each year. The goal here is to develop a hatchery broodstock that is genetically *integrated* with the naturally spawning population in Abernathy Creek, not develop a hatchery population that is genetically *segregated* from the natural population.

Task 5b: Rear and release 20,000 HOR steelhead yearlings (smolts) each year into Abernathy Creek. Details regarding this task are given in Appendix 3.

Objective 6. Determine the relative, natural reproductive success of hatchery-origin and natural origin steelhead in Abernathy Creek upstream of the AFTC.

Task 6a. Select a set of highly variable, microsatellite nuclear DNA loci that can be used for determining parentage and reconstructing genetic pedigrees of HOR and NOR steelhead in Abernathy Creek. We will screen 20 microsatellite DNA loci that

have been shown to be well suited for parentage analysis in steelhead trout (William Ardren, unpublished data for steelhead from the Hood River, OR; Paul Moran, unpublished data for steelhead from Little Sheep Creek, Imnaha River, OR; Bentzen et al. 2001) on a sample of 50-100 captively-reared steelhead collected as age 0+ parr from Abernathy Creek (see Objective 1). DNA will be extracted from fin tissue in a Chelex 100 (Sigma Chemical Co.) resin solution as described by Miller and Kapuscinski (1996). Template DNA will be PCR-amplified in a MJ Research PTC-200 DNA engine thermocycler in 15 µL reactions containing 1x polymerase buffer (10 mM Tris-HCL, 50 mM KCL, 1% Triton X-100), 1.5 to 2 mM $MgCl_2$, 0.2 mM each dNTP, 0.5µM of each primer and 0.5 U Taq DNA polymerase (obtained from Promega Corporation). The PCR products will be fractioned on 2% agarose gels to determine the quality of the PCR product. Genotypes will be determined by post-PCR multiplexing the amplified loci on an *ABI 310 DNA Analyzer* with the G5 filter set to produce electropherograms. *GeneScan* and *Genotyper* software from Applied Biosystems Incorporated (ABI) will be used to identify alleles at each locus and genotype each fish. Loci with the highest expected heterozygosity and allelic diversity will be selected for further analysis. A subset of 10-15 highly variable loci, which are easy to score (i.e., no indication of upper allele dropout or null alleles), will be selected.

To determine the total number of loci needed to resolve parentage with 95-99% confidence, we will use simulations to determine the resolving power of the loci given the allele frequencies in each population (Marshall *et al.* 1998; Gerber *et al.* 2000). These simulations involve using the allele frequencies for each population to generate parent-offspring pairs or triplets and random genotypes representing unrelated candidate parents. From these simulated data sets, we will calculate the expected distribution of the test statistic, delta, which is the difference in likelihood ratios between the two parents (or parent pairs) most likely to have parented the offspring. From the distributions of delta scores generated in the simulations, we can determine a set of loci that will produce 95% confidence in assignment of parentage. Allele frequencies for the Abernathy Creek population of steelhead will be estimated from all captively-reared adults spawned in ~~1999, 2000, and 2001~~ 2002, 2003, 2004, and 2005 (Table 1). These estimated allele frequencies will thus be derived from a minimum of 100 fish of each brood year. Estimated allele frequencies for the Abernathy Creek population will be compared to those for other regional populations including steelhead from the Cowlitz River, Kalama River, Elochoman River (Beaver Creek), Forks Creek, Big Creek, etc.

Task 6b: Determine the multilocus DNA genotypes of all natural-origin adult steelhead trapped at the AFTC and all NOR and HOR adults passed upstream to spawn naturally in Abernathy Creek. Fin clips and scale samples will be taken from all adult steelhead passed upstream of the weir to spawn naturally in Abernathy Creek. These samples will be taken every year from 2005 through 2015 (Table 1). In addition, we will take fin clips from all captively-reared adults that are spawned to produce F1 HOR yearlings for release (see Task 6a and Objective 1). We will also collect fin clips each year from approximately 1,000 age 0+ NOR parr representing the progeny of NOR and HOR adults passed upstream to spawn naturally in Abernathy Creek (see Objective 7). All tissue samples will be collected and stored in 100% ethanol. DNA extractions,

PCR conditions, and multilocus genotyping at a suite of microsatellite DNA loci will be performed as outlined in **Task 6a**.

Task 6c: Determine the parents and pedigree of each NOR adult trapped at the AFTC, all HOR and NOR adults spawned at the AFTC, and age 0+ NOR juveniles collected from Abernathy Creek (see Task 7a). Parentage of each juvenile offspring PIT tagged in Abernathy Creek (Task 7a) will be determined by comparing the multilocus genotypes for all potential parents passed upstream and juvenile offspring based on Mendelian rules of inheritance and likelihood approaches that allow for microsatellite genotype scoring errors (Marshall *et al.* 1998; Gerber *et al.* 2000). We will be able to assign parentage to returning HOR adults trapped at the weir beginning in 2005. Similarly, we will be able to assign parentage to returning NOR adults trapped at the weir in 2008 (Table 1).

Task 6d: Determine the natural reproductive success of each NOR and HOR steelhead allowed to spawn naturally in Abernathy Creek upstream of the weir by identifying their offspring at the juvenile and adult life history stages. The ability to examine both of these life stages is important because it allows us to determine if there is differential survival during the ocean life history phase for NOR fish with hatchery-origin or natural-origin parents. The adult-to-adult survival data will also give us an estimate of lifetime reproductive success and fitness among HOR and NOR adults. Moreover, we will be able to provide these estimates for F1 and F2 HOR fish, and F2 NOR fish representing the natural origin progeny of F1 HOR adults that spawned successfully in Abernathy Creek (Table 1).

Task 6e: Determine if the NOR (W) and HOR (H) fish mate randomly and produce offspring with equal survival rates in Abernathy Creek. We will compare the frequency of WxW, WxH, HxW, and HxH matings estimated from genotypic analyses of juveniles collected from Abernathy Creek. These estimated frequencies will be compared to binomial expectations based on the number of HOR and NOR adults passed upstream and to NOR adults trapped at the AFTC in subsequent years.

Table 6f: Estimate heritabilities and genetic correlations of fitness-related, life history traits such as return timing, age and size at return (maturity). We will use the methods of Milner *et al*. (2000) to estimate heritabilities of each trait measured on all adults trapped at the AFTC. We will use the heritability estimates to determine best linear unbiased predictions (BLUP; Henderson 1975) of additive genetic values for adult life history traits related to fitness. This information will allow us to compare average genetic values of the groups (HxH, HxW, WxH, WxW) and monitor genetic change across generations.

Task 6g: Estimate the "genetic" effective number of natural breeders each year by direct and indirect genetic methods (Ardren and Kapuscinski 2003).

Task 6h: Estimate natural stray rates of steelhead into Abernathy Creek and potential genetic contributions by resident rainbows trout. NOR juveniles in

Abernathy Creek with unassignable parents (i.e. one or both parents could not be identified genotypically among adults trapped at the weir and passed upstream) will be considered the progeny of resident fish because the likelihood of adult fish swimming through the electric barrier is extremely remote (David Smith, Smith-Root Company, Vancouver, Washington). Similarly, NOR adult steelhead captured at the weir after 2010 without identifiable parents will be considered natural strays or the NOR progeny of adult fish that spawned downstream from the weir.

Objective 7. Determine physiological and morphological parameters of NOR juvenile steelhead in Abernathy Creek and compare those parameters between progeny of NOR, F1 HOR, and F2 HOR adults as identified through Task 6d.

Task 7a. Compare phenotypic traits of F1 and F2 NOR progeny and test the null hypothesis that the mean values of those traits are the same for all parental cross-types (e.g. WxW, HxW, WxH, HxH). Parr survival of individuals with known parentage will be estimated using depletion estimates with electrofishing techniques. Approximately 1200 individuals will be collected in a stratified manner throughout the watershed upstream of the AFTC. All individuals will be fin-clipped for genetic analyses, measured for length, weight, morphometrics, and PIT tagged (for passive monitoring of migration timing, see below).

Smolt life history characteristics will be assessed using PIT tag interrogation methods (remote and portable) and from individuals collected in the smolt trap (details of smolt trapping are provided in the *Background* section). Juvenile migration timing of individuals with known parentage will be assessed using the two *SPI Systems* on Abernathy Creek (one at AFTC and one located 1 km downstream of AFTC). Habitat preference of juveniles with known parents will be assessed using the backpack PIT systems described previously. Monthly backpack surveys will be conducted downstream of AFTC to examine individual habitat preferences of genotyped individuals.

Physiological (gill Na^+,K^+-ATPase activity, methods of McCormick 1993; and thyroid hormone profile, methods being developed at AFTC), and morphological characters (morphometrics) will be examined among fish at the downstream smolt trap operated by WDFW. Annually, over 800 naturally-produced steelhead smolts have been collected at the trap to estimate total smolt production. All sampled individuals will be fin-clipped for genetic analysis and morphological measurements will be taken. A subsample (up to half) will be sampled non-lethally for physiological measurements.

Task 7b: Integrate genotypic and phenotypic information: analyze parentage results in the context of phenotypes. Parent-offspring information gained from this objective and the parentage analysis objective will be analyzed by ANOVA methods to determine differences in means among groups and chi-squared tests to determine whether life history traits (e.g. week of outmigration) are independent of cross-types. We will compare the distribution of juvenile (F1 and F2) life history traits (physiological-ATPase and thyroid hormone; morphological-morphometrics; and behavioral-juvenile migration timing and habitat preferences) to adult survival of all possible cross-types.

Objective 8: Prepare final reports and manuscripts for publication.

V. REFERENCES

Ardren WR, Kapuscinski AR (2003) Demographic and genetic estimates of effective population size (N_e) reveals genetic compensation in steelhead trout. *Molecular Ecology*, 12, 35-49.

Bentzen P, Olsen JB, McLean JE, Seamons TR, Quinn TP (2001) Kinship analysis of Pacific salmon: insights into mating, homing, and timing of reproduction. *Journal of Heredity,* **92**,127-136.

Campton, D.E. 1995. Genetic effects of hatchery fish on wild populations of Pacific salmon and steelhead: What do we really know? Edited by H.L.Jr. Schramm and R.G. Piper. American Fisheries Society, Bethesda, Maryland pp. 337-353.

Carey, J.B. and McCormick, S.D. 1998. Atlantic salmon smolts are more responsive to handling and confinement stress than parr. Aquaculture 168: 237-253.

Cedarholm, C.J. and seven coauthors. 1997. Response of juvenile coho salmon and steelhead to placement of large woody debris in a coastal Washington stream. N. Am. J. Fish. Managment 17: 947-963.

Dickhoff, W.W., Folmar, L.C., Mighell, J.L., and Mahnken, C.V.W. 1982. Plasma thyroid hormones during smoltification of yearling and underyearling coho salmon and yearling chinook salmon and steelhead trout. Aquaculture 28: 39-48.

Flagg, T.A., Waknitz, F.W., Maynard, D.J., Milner, G.B., and Mahnken, C.V.W. 1995. The effect of hatcheries on native coho salmon populations in the lower Columbia River. Am. Fish. Soc. Symp. 15: 366-375.

Gerber S, Mariette S, Streiff R, Bodenes C, Kremer A (2000) Comparison of microsatellites and amplified fragment length polymorphism markers for parentage analysis. *Molecular Ecology*, **9**, 1037-1048.

Henderson CR (1975) Best linear unbiased estimation and prediction under a selection model *Biometrics*, 31, 423-447.

Hilgert, P.J., Hershberger, W.K., and Root, D.V. 1992. Evaluation of a graduated electric field as a fish exclusion device at the Quilcene National Fish Hatchery. Final Report to Puget Sound Power & Light Company, Beak Consultants Incorporated, Kirkland, WA.

Hindar, K., Ryman, N., and Utter, F. 1991. Genetic Effects of cultured fish on natural fish populations. Canadian Journal of Fisheries and Aquatic Sciences 48: 945-957.

IMST (Independent Multidisciplinary Science Team) 2000. Conservation Hatcheries and Supplementation Strategies for Recovery of Wild Stocks of Salmonids: Report of a Workshop. IMST, Portland, OR.

Johnson, O.W. and and several other authors In prep. Status review of west coast cutthroat trout (Oncorhynchus clarki clarki) from Washington, Oregon, and California. National Marine Fisheries Service, NOAA, U.S. Dept. of Commerce, Seattle, WA.

Mackey, G., McLean, J.E., and Quinn, T.P. 2001. Comparisons of run timing, spatial distribution, and length of wild and newly established hatchery populations of steelhead in Forks Creek, Washington. N. Am. J. Fish. Management 21: 717-724.

Marshall TC, Slate J, Kruuk LEB, Pemberton JM (1998) Statistical confidence for likelihood-based paternity inference in natural populations. *Molecular Ecology*, **7**, 639-655.

McCormick, SD (1993) Methods for non-lethal gill biopsy and measurement of Na^+,K^+ - ATPase activity. *Canadian Journal of Fisheries and Aquatic Sciences,* 50: 656-658.

Miller LM, Kapuscinski AR (1996) Microsatellite DNA markers reveal new levels of variation in northern pike. *Transactions of the American Fisheries Society*, **125**, 971-997.

Milner JM, Pemberton JM, Brotherstone S, Albon SD (2000) Estimating variance components and heritabilities in the wild: a case study using the 'animal model approach'. *Journal of Evolutionary Biology,* 13, 804-813.

Nehlsen, W., Williams, J.E., and Lichatowich, J.A. 1991. Pacific salmon at the crossroads: stocks at risk from California, Oregon, Idaho, and Washington. Fisheries 16: 4-21.

NWWPC (Northwest Power Planning Council) 1999. Artificial Production Review, Council Document 99-15. Northwest Power Planning Council, Portland, OR.

Prunet, P. and Boeuf, G. 1985. Plasma prolactin level during transfer of rainbow trout and Atlantic salmon from fresh water to sea water. Aquaculture 45: 167-176.

Quinn, T.P. 1993. A Review of Homing and Straying of Wild and Hatchery-Produced Salmon. Fisheries Research 18: 29-44.

Ryman, N., Jorde, P.E., and Laikre, L. 1995. Supportive breeding and variance effective population size. Conservation Biology 9: 1619-1628.

Ryman, N. and Laikre, L. 1991. Effects of supportive breeding on the genetically effective population size. Conservation Biology 5: 325-329.

Waples, R.S. 1991. Genetic interactions between hatchery and wild salmonids: lessons from the Pacific Northwest. Canadian Journal of Fisheries and Aquatic Sciences 48: 124-133.

Zydlewski GB, Winter C, McClanahan D, Johnson J, Zydlewski J (2003). Evaluation of fish movements, migration patterns, and population abundance with Streamwidth PIT tag Interrogation systems. *Bonneville Power Administration Report*, Project # 2001-012-00.

QUALIFICATIONS OF INVESTIGATORS

Donald E. Campton (Co-Principal Investigator)

D.E. Campton (Ph.D. Genetics, University of California, Davis; M.S. Fisheries, University of Washington, Seattle) is the Regional Fish Geneticist for the U.S. Fish & Wildlife Service, Pacific Region. Dr. Campton's previous research includes population and quantitative genetic analyses of natural and cultured fish populations, respectively. His primary research interests include the genetic effects of hatcheries and artificial propagation on natural populations of salmonid fishes. D.E. Campton has served as Principal Investigator or co-P.I. on 14 contracts or grants, including two from the Bonneville Power Administration. He has authored or coauthored approximately 30 peer-reviewed publications. He has worked professionally in the general area of fishery genetics for approximately 25 years including two years (1978-1980) as a Fishery Research Biologist for the Washington Department of Wildlife (formerly Dept. of Game) on a steelhead project on the Skagit River. He currently serves as a member of the Hatchery Scientific Review Group for the Congressionally-funded Western Washington Hatchery Reform Project. Dr. Campton will initially serve as Project Manager and will be responsible for recruiting new personnel, filing quarterly and annual reports, and overall project management and coordination. He is also the principal author of this proposal.

William R. Ardren (Co Principal Investigator)

W.R. Ardren (Ph.D. Fisheries Science, University of Minnesota) is the lead molecular population geneticist for the U.S. Fish and Wildlife Service, Pacific Region, Conservation Genetics Laboratory at the Abernathy Fish Technology Center. Dr. Ardren's BPA-funded post-doctoral research at Oregon State University involved using microsatellite loci and pedigree reconstruction to determine the relative reproductive success of hatchery and wild steelhead trout (*Oncorhynchus mykiss*) in the Hood River, OR. Examples of other research projects Dr. Ardren has accomplished include describing the inheritance of microsatellite loci in steelhead trout, conducting a genetic analysis of a captive breeding program for an endangered steelhead trout population, and using demographic and genetic estimates of effective population size to clarify the relationship between long-term changes in genetic diversity and population productivity. In addition, he has isolated and characterized microsatellite loci for two species of fish and co-authored a review paper on methods of parentage analysis in natural populations. Dr. Ardren will be primarily responsible for the molecular genetic work and pedigree reconstructions associated with this project.

Gayle Barbin Zydlewski (Co-Principal Investigator)

G.B. Zydlewski (Ph.D. Oceanography, University of Maine; M.S. Biology, University of Rhode Island) is the lead Behavioral Fish Physiologist for the U.S. Fish & Wildlife Service's Pacific Region, Abernathy Fish Technology Center. G.B. Zydlewski has served as Principal Investigator or co-investigator on 7 contracts with associated grants, including one from the Bonneville Power Administration and two from the U.S. Army Corps of Engineers. She has authored and co-authored approximately 10 peer-reviewed publications. She has worked professionally in the area of fish behavior and physiology for approximately 12 years and currently investigates the relationships between juvenile rearing strategies and adult return rates; the effects of modifications and enhancements to hatchery rearing environments on fish growth, behavior, physiology and survival; new approaches to investigate the extent of interactions between hatchery-reared and wild fish; and restoration and recovery strategies for populations in decline. A collaborative project on cutthroat trout in the lower Columbia River (funded by the U.S. Army Corps of Engineers) resulted in construction of PIT monitoring systems on the Chinook River. Her previous work includes a BPA funded "innovative project" to develop a PIT tag interrogation system for Abernathy Creek, complete with remote sensing capabilities and satellite uplinks. Dr. Zydlewski will be responsible for all

behavioral, physiological, and morphological evaluations of hatchery-origin and natural-origin steelhead in Abernathy Creek. She will also coordinate all PIT tagging operations and supervise other biologists responsible for maintaining the interrogation system in Abernathy Creek.

Patricia A. Crandell (Co-Principal Investigator)

P.A. Crandell (Ph.D. Genetics, University of California, Davis) is the Deputy Director and currently Acting Director of the Abernathy Fish Technology Center, U.S. Fish & Wildlife Service, Longview, Washington. The AFTC is the largest of the Service's seven applied fishery research centers. Dr. Crandell's previous research includes quantitative genetic analyses of growth and maturation traits in rainbow trout and Pacific salmon. She has also worked professionally as a statistician for the National Marine Fisheries Service in Juneau, Alaska. P.A. Crandell has served as Principal Investigator or co-P.I. on 6 contracts or grants. She has authored or co-authored approximately 11 peer-reviewed publications. She has worked professionally in the general areas of quantitative analysis and fishery genetics for approximately 17 years. Dr. Crandell will be responsible for all quantitative genetic analyses of adult life history traits derived from the pedigree reconstructions obtained via DNA genotypic identifications. She will also provide statistical consulting and advice to the other investigators on this project.

Stephen C. Riley (Associate Investigator)

S.C. Riley (Ph.D. Fishery and Wildlife Biology, Colorado State University; M.S. Biology, University of Waterloo, Ontario, Canada) is a currently employed as a Fishery Research Biologist by the National Marine Fisheries Service at the Manchester Research Station in Manchester, Washington. He has over 20 years of experience as a Fisheries Biologist and Stream Ecologist working on wild salmonid populations throughout North America, including Ontario, Quebec, Newfoundland, British Columbia, Idaho, California, Colorado, and Washington. He has worked on a variety of aspects of stream and salmonid ecology, including juvenile behavior, intraspecific competition, life history, habitat use, population estimation, salmonid stock discrimination, habitat improvement, watershed restoration, and the effects of timber harvest on salmonid populations. Dr. Riley also has experience with salmon stock assessment, harvest management and population modeling. Dr. Riley is a member of the technical panel for the WA Salmon Recovery Funding Board. His current research focuses on the effects of salmonids released from hatcheries on the ecology of wild salmonids in streams. Dr. Riley will be responsible for the snorkel surveys and stomach contents analyses of residualized HOR steelhead in Abernathy Creek to assess their potential ecological impacts on NOR salmonids.

WILLIAM R. ARDREN

CONTACT: U.S. Fish & Wildlife Service, 1440 Abernathy Creek Road Longview, WA 98632.
TEL: 360-425-6072; FAX: 360-636-1855; E-mail: William_Ardren@fws.gov

EDUCATION

Ph.D. , Fisheries Science, University of Minnesota, Department of Fisheries, Wildlife and Conservation Biology, St. Paul. 1999.

B.A., Biology, St. John's University, Department of Biology, Collegeville, Minnesota. 1993.

EMPLOYMENT

2002-present: Molecular Population Geneticist, U.S. Fish & Wildlife Service, Abernathy Fish Technology Center, Longview, WA.

2001-2002: Post-Doctoral Research Associate, Department of Zoology, Oregon State University, Corvallis OR.

1999-2001: Assistant Professor, Department of Biology, Luther College, Decorah IA.

PROJECT ROLES

Oversee genetic analysis of all samples, conduct parentage analysis, maintain genetic database, and carry out statistical analyses to determine the reproductive success of hatchery and wild spawners.

ADDITIONAL QUALIFICATIONS

W.R. Ardren is the lead molecular population geneticist for the U.S. Fish and Wildlife Service, Region 1, Conservation Genetics Laboratory at the Abernathy Fish Technology Center. Dr. Ardren's post-doctoral research at Oregon State University involved using microsatellite loci and pedigree reconstruction to determine the relative reproductive success of hatchery and wild steelhead trout (*Oncorhynchus mykiss*) in the Hood River, OR. Examples of other research projects Dr. Ardren has accomplished include describing the inheritance of microsatellite loci in steelhead trout, conducting a genetic analysis of a captive breeding program for an endangered steelhead trout population, and using demographic and genetic estimates of effective population size to clarify the relationship between long-term changes in genetic diversity and population productivity. In addition, he has isolated and characterized microsatellite loci for two species of fish and co-authored a review paper on methods of parentage analysis in natural populations.

RELEVANT GRANTS

Ardren, W.R., and D.E. Campton. Genetic Analyses of Steelhead in the Hood River. $14,436, BPA Contract No. 00013429, April 1, 2003 – September 30, 2003. BPA Contracting Officer: Tom Morse, 503-230-3694.

Ardren, W.R. Genetic Analysis of the Keogh River Steelhead Trout Living Gene Bank. $7351, British Columbia Ministry of Fisheries Contract No. 1070-20/01-49 and British Columbia Conservation Foundation Contract No. LM00-02, 1999-2001.

RELEVANT PUBLICATIONS

Ardren, W.R., S.O. Borer, F. Thrower, J.E. Joyce and A.R. Kapuscinski. 1999. Inheritance of 12 microsatellite markers in *Oncorhynchus mykiss*. Journal of Heredity, 90:529-536.

Ardren, W.R., L.M. Miller, J.A. Kime, and M.A. Kvitrud. 2002. Microsatellite loci for fathead minnow (*Pimephales promelas*). Molecular Ecology, Notes 2:226-227.

Ardren, W.R. and A.R. Kapuscinski. 2003. Demographic and genetic estimates of effective population size (N_e) reveals genetic compensation in steelhead trout. Molecular Ecology, 12:35-49.

Jones, A.G. and **W.R. Ardren**. Review Paper: Methods of parentage analysis in natural populations. Molecular Ecology, *Accepted for publication pending minor revision*.

Ardren, W.R. and M. Blouin. Relative reproductive success of hatchery and wild steelhead trout spawning in Hood River Oregon. *In Preparation.*

DONALD E. CAMPTON

CONTACT: U.S. Fish & Wildlife Service, 1440 Abernathy Creek Road Longview, WA 98632.
TEL: 360-425-6072; FAX: 360-636-1855; E-mail: Don_Campton@fws.gov

EDUCATION

Ph.D., Genetics, University of California, Davis, 1986.
M.S., Fisheries, University of Washington, Seattle, 1981.
B.S., Genetics, University of California, Berkeley, 1974.

EMPLOYMENT

1997-present: Regional Fish Geneticist, U.S. Fish & Wildlife Service, Abernathy Fish Technology Center, Longview, WA.
1986-1997: Assistant/Associate Professor, Department of Fisheries and Aquatic Sciences, University of Florida, Gainesville, FL.
1981-1986: Graduate Research Assistant, University of California, Davis, California.
1978-1980: Fishery Research Biologist, Washington Department of Game, Mount Vernon, WA.

ADDITIONAL QUALIFICATIONS

D.E. Campton has served as Principal Investigator or co-P.I. on 14 contracts or grants, including two from the Bonneville Power Administration (see below). He has authored or coauthored approximately 30 peer-reviewed publications. He has worked professionally in the general area of fishery genetics for approximately 25 years. He currently serves as a member of the Hatchery Scientific Review Group for the Congressionally-funded Western Washington Hatchery Reform Project.

RELEVANT GRANTS/CONTRACTS

Campton, D.E., D.G. Elliot, R.J. Pascho, and J.J. Hard. Heritability of Disease Resistance and Immune Function in Chinook Salmon. $199,974, BPA Project No. 2000-072-00, May 1, 2001 – June 30, 2003. BPA Contracting Officer: Deborah Docherty, 503-230-4458.

Ardren, W.R., and **D.E. Campton.** Genetic Analyses of Steelhead in the Hood River. $14,436, Work Order No. 103538, BPA Project No. 1988-053-12, April 3, 2003 – September 30, 2003. BPA Contracting Officer: Tom Morse, 503-230-3694.

RELEVANT PUBLICATIONS

Campton, D.E. and F.M. Utter. 1985. Natural hybridization between steelhead trout (*Salmo gairdneri*) and coastal cutthroat trout (*Salmo clarki clarki*) in two Puget Sound streams. Can. J. Fish. Aquat. Sci. 42:110-119.

Campton, D.E. and J.M. Johnston. 1985. Electrophoretic evidence for a genetic admixture of native and nonnative rainbow trout in the Yakima River, Washington. Trans. Am. Fish. Soc. 114:782-793.

Campton, D.E. and F.M. Utter. 1987. Genetic structure of anadromous cutthroat trout (*Salmo clarki clarki*) populations in the Puget Sound area: evidence for restricted gene flow. Can. J. Fish. Aquat. Sci. 44:573-582.

Campton, D.E. 1987. Natural hybridization and introgression in fishes: methods of detection and genetic interpretations, p.161-192. *IN*: N. Ryman and F. Utter (eds.), *Population Genetics and Fishery Management*. Washington Sea Grant College Program and University of Washington Press, Seattle, Washington.

Campton, D.E., F.W. Allendorf, R.J. Behnke, and F.M. Utter; M.W. Chilcote, S.A. Leider, and J.J. Loch. 1991. Reproductive success of hatchery and wild steelhead. Trans. Am. Fish. Soc. 120: 816-827.

Campton, D.E. 1995. Genetic effects of hatchery fish on wild populations of Pacific salmon and steelhead: What do we really know?, p. 337-353. *IN*: H.L. Schramm, Jr., and R.G. Piper (eds), *Uses and Effects of Cultured Fishes in Aquatic Ecosystems*, American Fisheries Society Symposium 15, American Fisheries Society, Bethesda, Maryland.

Campton, D.E., A.L. Bass, F.A. Chapman, and B.W. Bowen. 2000. Genetic distinction of pallid, shovelnose, and Alabama sturgeon: emerging species and the U.S. Endangered Species Act. Conservation Genetics 1: 17-32.

Burger, C.V., K.T. Scribner, W.J. Spearman, C.O. Swanton, and **D.E. Campton**. 2000. Genetic contribution of three introduced life history forms of sockeye salmon to colonization of Frazer Lake, Alaska. Can. J. Fish. Aquat. Sci. 57: 2096-2111.

PATRICIA A. CRANDELL

CONTACT: U.S. Fish & Wildlife Service, 1440 Abernathy Creek Road Longview, WA 98632.
TEL: 360-425-6072; FAX: 360-636-1855; E-mail: Patricia_Crandell@fws.gov

EDUCATION

Ph.D., Genetics, University of California, Davis, 1991
B.S., Genetics, University of California, Davis, 1983

EMPLOYMENT

2002-present: Deputy Director, U.S. Fish & Wildlife Service, Abernathy Fish Technology Center, Longview, WA
2001-2002: Statistician, National Marine Fisheries Service, Restricted Access Management, Juneau, AK
1998-2001: Supervisory Research Analyst, Alaska State Department of Corrections, Juneau, AK
1991-1997: Postdoctoral Research Associate, University of Alaska, Fairbanks, Juneau, AK

ADDITIONAL QUALIFICATIONS

P.A. Crandell has served as Principal Investigator or co-P.I. on 6 contracts or grants. She has authored or co-authored approximately 11 peer-reviewed publications. She has worked professionally in the general areas of quantitative analysis and fishery genetics for approximately 17 years. She is currently acting Director of Abernathy Fish Technology Center, the largest of the 7 US Fish and Wildlife Service's applied fishery research centers.

RELEVANT GRANTS/CONTRACTS

Crandell, P.A. The Genetics of Pink Salmon Growth. $100,000, USDA National Research Initiative Competitive Grant, 1993.

RELEVANT PUBLICATIONS

Crandell, P.A. 1997. Genetic variation of growth and maturity traits in salmonids. W.W. Smoker and S. Ignell, eds., Age at Maturity Workshop, 1996

Crandell, P.A. and G.A.E. Gall, 1993. The genetics of body weight and its effect on early maturity based on individually tagged rainbow trout (*Oncorhynchus mykiss*). Aquaculture, 117:77-93

Crandell, P.A. and G.A.E. Gall, 1993. The genetics of age and weight at sexual maturity based on individually tagged rainbow trout (*Oncorhynchus mykiss*). Aquaculture, 117:95-105

Crandell, P.A. and G.A.E. Gall, 1993. The effect of sex on heritability estimates of body weight determined from data on individually tagged rainbow trout (*Oncorhynchus mykiss*). Aquaculture, 113:47-55

Crandell, P.A. and G.A.E. Gall, 1992. Body weight analysis for individually tagged rainbow trout: males, females, and precocious males. Aquaculture, 100:99

Matsuoka, M.P., Gharrett, A.J., Wilmot, R.L., **Crandell, P.A**., and Smoker, W.W., 1996. Gene-centromere recombination rates of allozyme loci in even- and odd-year pink salmon, *Oncorhynchus, gorbuscha*. Aquaculture Biotechnology Proceedings, Intl. Cong. Biol. of Fish., 131-138

GAYLE BARBIN ZYDLEWSKI

CONTACT: U.S. Fish & Wildlife Service, 1440 Abernathy Creek Road, Longview, WA 98632.
TEL: 360-425-6072; FAX: 360-636-1855; E-mail: gayle_zydlewski@fws.gov

EDUCATION

Ph.D., Oceanography, University of Maine, 1996
M.S., Zoology, University of Rhode Island, 1992
B.S., Biology, Southeastern Massachusetts University, 1990

EMPLOYMENT

1999-present: Behavioral Physiologist, U.S. Fish and Wildlife Service, Abernathy Fish Technology Center, Longview, WA
1997-1999: Postdoctoral Research Associate**,** USGS/BRD, Conte Anadromous Fish Research Center, Turners Falls, MA
1996-1997: Postdoctoral Research Associate, University of Maine, School of Marine Sciences, Orono, ME
1992-1996: Research Associate, Ph.D. Candidate, University of Maine, Department of Oceanography, Orono, ME

ADDITIONAL QUALIFICATIONS

G.B. Zydlewski has served as Principal Investigator or co-investigator on 7 contracts with associated grants, including one from the Bonneville Power Administration and two from the U.S. Army Corps of Engineers. She has authored and co-authored approximately 10 peer-reviewed publications. She has worked professionally in the area of fish behavior and physiology for approximately 12 years and currently investigates the relationships between juvenile rearing strategies and adult return rates; the effects of modifications and enhancements to hatchery rearing environments on fish growth, behavior, physiology and survival; new approaches to investigate the extent of interactions between hatchery-reared and wild fish; and restoration and recovery strategies for populations in decline. The collaborative project on cutthroat trout listed below resulted in construction of PIT monitoring systems on the Chinook River.

RELEVANT GRANTS/CONTRACTS

Zydlewski, G.B., A. Haro, T. Coley, J. Johnson. 2001. Evaluate new methodologies for monitoring Pacific salmon and steelhead: Methods for evaluating the effectiveness of restoration and recovery programs. $ 217,000. BPA Project #2001-012-00. 15 May 2001 – 15 August 2002. Contracting Officer: Pat Poe.

Zydlewski, J. (**G.B. Zydlewski** co-investigator). 2002. Movements of the Coastal Cutthroat Trout in the Lower Columbia River: Tributary, Main-Stem and Estuary Use. U.S. Army Corps of Engineers. $360,000. April 2002 – April 2007.

RELEVANT PUBLICATIONS

Zydlewski, G.B., A.J. Haro, K.G. Whalen, & S.D. McCormick. 2001. Performance of stationary and portable Passive Transponder detection systems for monitoring of fish movements. Journal of Fish Biology. 58(5): 1471-1475.

Zydlewski, G.B., S. Foott, K. Nichols, S. Hamelberg, J. Zydlewski, B. Th. Björnsson. In Press. Enhanced smolt characteristics of steelhead trout exposed to alternative hatchery conditions during the final month of rearing. Aquaculture.

Zydlewski, G.B., J. R. Johnson. 2002. Response of bull trout fry to four water diversion screen types. North American Journal of Fisheries Management. 22: 1276-1282.

Zydlewski, G.B., S.D. McCormick, & A.J. Haro. In Review. The role of temperature in downstream migratory behavior of Atlantic salmon smolts.

Barbin, G.P., S.J. Parker, & J.D. McCleave. 1998. Olfactory clues play a critical role in the estuarine migration of silver-phase American eels. Environmental Biology of Fishes 53: 283-291.

STEPHEN C. RILEY

CONTACT: National Marine Fisheries Service, Northwest Fisheries Science Center, Resource Enhancement & Utilization Technologies Division, P.O. Box 130, Manchester, WA 98353
TEL: 360-871-8315; Email: Stephen.Riley@noaa.gov

EDUCATION

Ph.D. Fishery and Wildlife Biology, Colorado State University, Fort Collins, CO 1992
M.Sc. Biology, University of Waterloo, Waterloo, Ontario, Canada, 1986
B.Sc. Honours Biology, University of Waterloo, Waterloo, Ontario, Canada, 1984

EMPLOYMENT

1999-Present: Fishery Research Biologist, National Marine Fisheries Service, Manchester, WA
1997-1999: Independent Fisheries Consultant, Prince Rupert, B. C.
1996-1997: Fisheries Scientist, Jones and Stokes Associates, Inc., Sacramento, CA
1994-1995: Project Manager, Keogh River Watershed Restoration Project, Port Hardy, B. C.
1994 (Jan.-Nov.): Fishery Research Biologist, Idaho Department of Fish and Game, Boise, ID.
1992-1993: Postdoctoral Fellow, Ocean Sciences Center, Memorial University, St. John's, NFLD.

ADDITIONAL QUALIFICATIONS

S.C. Riley has extensive experience as a Fisheries Biologist and Stream Ecologist. He has performed a large number of scientific studies involving snorkel surveys and other methods to assess within-stream behaviors and feeding habits of salmonid fishes in stream environments. One of his most recent grants was from the Western Washington Hatchery Reform Project to assess ecological interactions between hatchery-origin and natural-origin salmonids in a number of Puget Sound and Olympic Pennisula streams.

RELEVANT PUBLICATIONS

Berejikian, B.A., E.P. Tezak**, S.C. Riley**, and A.L. LaRae. 2001. Territorial behaviour and relative competitive ability of juvenile steelhead (Oncorhynchus mykiss) reared in enriched and conventional hatchery tanks and a stream environment. J. Fish Biol. 59:1600-1613.

Bult, T.P., **S.C. Riley**, R.L. Haedrich, R.J. Gibson, and J. Heggenes. 1999. Density-dependent habitat selection by juvenile Atlantic salmon (Salmo salar L.) in experimental riverine habitats. Can. J. Fish. Aquat. Sci. 56:1298-1306.

Riley, S.C., and K.D. Fausch. 1995. Trout population response to habitat enhancement in six northern Colorado streams. Can. J. Fish. Aquat. Sci. 52:34-53.

Gowan, C., M.K. Young, K.D. Fausch, and **S.C. Riley**. 1994. Restricted movement in stream salmonids: a paradigm lost? Can. J. Fish. Aquat. Sci. 51:2626-2637.

Riley, S.C., K.D. Fausch, and C. Gowan. 1992. Movement of brook trout (Salvelinus fontinalis) in four small subalpine streams in northern Colorado. Ecol. Freshw. Fish 1:112-122 .

Riley, S.C., G. Power, and P.E. Ihssen. 1989. Meristic and morphometric variation in parr of Ouananiche and anadromous Atlantic salmon from rivers along the North Shore of the Gulf of St. Lawrence. Trans. Am. Fish. Soc. 118:515-522.

Adams, N.J., D.R. Barton, R.A. Cunjak, G. Power, and **S.C. Riley**. 1988. Diel patterns of activity and substrate preference in young Arctic char of the Koroc River, northern Quebec. Can. J. Zool. 66:2500-2502.

Riley, S.C., and G. Power. 1987. Age at maturity in landlocked and anadromous Atlantic salmon parr from two Quebec Rivers. Env. Biol. Fishes 19:223-228.

PROPOSED BUDGET: YEAR 1

Personnel

Campton, D.E. (Co-P.I., 0.10 FTE, GS-13/4)	$ 7,612
Ardren, W.R. (Co-P.I., 0.05 FTE, GS-12/1)	2,910
Zydlewski, G.B. (Co-P.I., 0.10 FTE,GS-13/1)	6,920
Crandell, P.A. (Co-P.I., 0.05 FTE, GS-13/1)	3,460
Supervisory Fishery Biologist (1.0 FTE, GS-11)	48,550
Fishery Biologist (GS-9, 1.0 FTE)	40,126
Fish Culturist / Animal Caretaker (GS-5, 1.0 FTE)	26,482
Subtotal salaries	136,060
Fringe benefits (35%)	47,621
Total, personnel	183,681

Major equipment

Backpack electroshocker + accessories (Smith-Root)	6,000
Equipment for remote detection of PIT tags	13,000
Replacement transceiver ($6,000)	
Oscilloscope ($2,000)	
Current probe ($800)	
Battery backup ($500)	
Solar chargers ($2,860)	
Satellite uplink/downlink service for remote site ($840)	

Subcontract to Washington Dept. of Fish & Wildlife

Smolt trapping M&E on Abernathy, Germany, and Mill creeks	70,000

Subcontract to National Marine Fisheries Service

Snorkel surveys of residualized fish released from the AFTC	20,000

Expendable supplies

Laboratory chemicals for physiological assays	3,000
Nuclear DNA genotypic assays (400 fish @ $30/fish)	12,000
PIT tags (2,200 @ $2.25 each)	4,950

Other

Fish food, electricity, misc. hatchery costs ($1,000/mo x 12 mos.)	12,000
Tagging/marking (20,000 fish x $0.16/fish)	3,200
Total Direct Costs (TDC)	327,831
Indirect costs (21.5% of TDC)	70,484
Total requested funds	**$398,315**

Figure 1. Geographic location of Abernathy, Germany, and Mill creeks in the lower Columbia River. The Abernathy Fish Technology Center (AFTC; a.k.a. Abernathy Hatchery) is located at approximately RM 3.4 on Abernathy Creek. The vast majority of steelhead spawning habitat in Abernathy Creek is upstream of the AFTC.

Table 1. Relationships among parental and progeny generations (Gen.) and brood years (BY), progeny release year, and progeny return years for HOR steelhead released from the Abernathy Fish Technology Center (AFTC) into Abernathy Creek. Generations are indicated as P (parental), F1, F2, and F3. Generation F1.5 represents the progeny of parental and F1 fish that are interbred. The parental generation (P) refers to natural origin steelhead collected as age 0+ parr from Abernathy Creek in 1999, 2000, and 2001 and reared captively to sexual maturity at the AFTC. Adult fish are expected to return and spawn primarily as age 3 and age 4 fish. A small proportion of fish are expected to return and spawn as age 2 (jacks) and age 5.

Parental		Progeny		Progeny Release	Progeny Return Years
Gen.	BY	Gen.	BY	Year	Age 3, Age 4
P	1999	F1	2002	2003	2005, 2006
P	2000	F1	2003	2004	2006, 2007
P	2001	F1	2004	2005	2007, 2008
F1	2002	F1.5	2005	2006	2008, 2009
F1	2003	F2	2006	2007	2009, 2010
F1	2004	F2	2007	2008	2010, 2011
F1.5	2005	F2+	2008	2009	2011, 2012
F2	2006	F3	2009	2010	2012, 2013
F2	2007	F3	2010	2011	2013, 2014
F2+	2008	F3+	2011	2012	2014, 2015

APPENDIX 1. Summary of reviews of previous versions of proposal submitted for BPA funding under other venues.

FY2001 *Innovative* Project Review (Project No. 22031)

Report of the Independent Scientific Review Panel (ISRP) for the original proposal submitted for BPA funding in FY2001 under the *Innovative Category.*

Project ID: 22031

Evaluation of Two Captive Rearing Methods for Assisting with Recovery of Naturally Spawning Populations of Steelhead and Coho Salmon.
Sponsor: U. S. Fish & Wildlife Service, U.S. Department of the Interior
Total Request: $264,064
Target Species: Steelhead and coho salmon
Short Description: (1) Develop a native broodstock of steelhead via captive rearing to sexual maturity of natural-origin, age 0+ juveniles and (2) short-term rearing of pre-smolt, natural-origin coho salmon to increase survival and provide fish for reintroduction programs.
Rank: Not innovative, but a good proposal.
Comments: This proposal is not recommended for support through the innovative review process because it does not meet the innovative criteria. It relies on standard practices even though it addresses a long-standing critical uncertainty. It is a well written and well designed proposal that would be of value to the region. Consequently, the project deserves "high priority" support through other venues, particularly for its application to upriver (ID) listed steelhead stocks. The proposal is technically sound, and the PI competent and meticulous. The proposal is particularly attractive because it proposes to rigorously examine the effects of hatchery rearing on fitness - a continuing, plaguing uncertainty in the basin's artificial production programs.

FY2002 Columbia Estuary Provincial Review (Project No. 30003)

We resubmitted our proposal under the Columbia Estuary Provincial Review in December 2001. The proposal was assigned Project No. 30003 under this province. The ISRP review of our proposal under the Columbia Estuary Provincial Review is presented below.

Project ID: 30003

Evaluation of Two Captive Rearing Methods for Assisting with Recovery of Naturally Spawning Populations of Steelhead and Coho Salmon
Sponsor: USFWS
Province: Columbia Estuary
Subbasin: Elochoman
FY03 Request: $446,101
5YR Estimate: $1,939,251
Short Description: Test and evaluate two hatchery reform methodologies; Assess natural reproductive success of returning hatchery-origin adults; Establish Abernathy, Germany, and Mill creeks as a Tier 3 "monitoring and evaluation" site for anadromous salmonids.
ISRP Final Recommendation: Fundable
CBFWA Category: High Priority
ISRP Comparison with CBFWA: Agree - Fundable, High Priority

ISRP Final Review Comments: Fundable; a response was not needed. Proponents propose to rigorously examine the effects of hatchery rearing on fitness - a continuing, plaguing uncertainty in the basin's artificial production programs. The proposed research would directly estimate fitness of supplemented fish and would test efficacy of two supplementation strategies.
CBFWA Review Comments: This project also has applications upstream of Bonneville and could be considered in the Mainstem and Systemwide Province if not funded here. NMFS has identified this project as a BiOp project.

The following project was submitted as a companion project to Project No. 30003. Portions of the following proposal have been included with the present proposal to address RPA 182 in Abernathy Creek.

Project ID: 30008

Instream evaluation of populations, migration timing, individual adult return rates, and wild-hatchery interactions of 3 naturally produced salmonids
Sponsor: USFWS
Province: Columbia Estuary
Subbasin: Elochoman
FY03 Request: $238,740
5YR Estimate: $1,296,140
Short Description: Evaluate distribution and abundance of juvenile and adult coho salmon, steelhead trout, and cutthroat trout in Abernathy Creek using new PIT tag techniques.
ISRP Final Recommendation: Fundable
CBFWA Category: High Priority
ISRP Comparison with CBFWA: Disagree - Fundable, Medium Priority
ISRP Final Review Comments: Fundable at medium priority. Without knowledge of proposal 30003 this proposal appears unfocused; it makes sense only as part of an assessment of supplementation of steelhead by a hatchery population. The objectives of understanding salmonid stocks in Abernathy Creek and developing and understanding the PIT tag methods are best justified in support of that assessment. The study design is competent and the study ISRP 2002-11 Final Five Province Review site seems well suited to the research. The ISRP remain unconvinced, however, that habitat preference can be assessed by simply watching natural fish, and that the interaction of hatchery and wild fish can be observed by simply mapping the distribution of wild fish before hatchery fish are released. There is not enough information to give reviewers confidence that the project will produce valuable results.
CBFWA Review Comments: This project may also be considered in the Mainstem and Systemwide Province. Some portions may be funded under that province. NMFS has identified this project as a BiOp project.

FY2003 Mainstem/Systemwide Review (Project No. 35027)

The ISRP's final review of our proposal, as submitted and revised in response to the ISRP's preliminary review, is presented below.

Project ID: 35027

Evaluation of Two Captive Rearing Methods for Assisting with Recovery of Naturally Spawning Populations of Steelhead and Coho Salmon.
Sponsor: USFWS
FY03 Request: $472,941 **5YR Estimate:** $2,046,091
CBFWA Adjusted FY03: $396,116 **3YR:** $932,966
Short Description: Test and evaluate two hatchery reform methodologies; Assess natural reproductive success of returning hatchery-origin adults; Establish Abernathy, Germany, and Mill creeks as a Tier 3 "monitoring and evaluation" site for anadromous salmonids.
ISRP Final Comments: Fundable. We agree with the CBFWA review, budget reductions, and Urgent ranking. This is a strong proposal that the ISRP has reviewed twice prior to the Mainstem/Systemwide provincial review. Each time we have supported it, as we do now. The proposal has four components: the steelhead brood stock study, rearing of coho salmon in the hatchery to reduce over-winter mortality, replacement of the electronic fence in Abernathy Creek, and the development of a Tier 3 Monitoring and Evaluation Site (NMFS RPA No. 183) for Lower Columbia and Southwest Washington ESUs of steelhead, coho salmon, chinook salmon, and coastal cutthroat trout. The authors' propose to assess the use of juveniles, rather than adults, to initiate local broodstocks for supplementation programs. Removing juveniles would impose less of a demographic loss on a depressed population and may reduce the risk of a Ryman-Laikre effect on the genetic composition of the population (i.e. the expansion of a small sample of the population into a much larger portion of the supplemented population with associated changes in genetic variation). In our preliminary comments, the ISRP took the unusual step of providing detailed suggestions on study design modifications to the project sponsor, as this is the third time we have considered this proposal. The project sponsor's response to the ISRP preliminary concerns was adequate and demonstrated full consideration of the ISRP comments. The sponsors modified the proposed design to address ISRP concerns so that the proposed monitoring now includes both natural and cultured populations. We also note that there is a strong cost sharing aspect to the program as WDFW will provide three rotary screw traps, and USFWS is proposing to recruit additional staff and has the facilities to conduct the proposed work. If funded, this project should be coordinated with other monitoring projects to ensure compatibility of objectives, common methods, and protocols. This coordination could be accomplished under the favorably reviewed CBFWA proposal #35033.

RME *Hydro and Hatchery* Workgroup comments on Proposed Project No. 35027:

35027 - Test and evaluate two hatchery reform methodologies; Assess natural reproductive success of returning hatchery-origin adults; Establish Abernathy, Germany, and Mill creeks as a Tier 3 "monitoring and evaluation" site for anadromous salmonids.
Address critical element of RPA? Relevant to RPA 182, 184. With respect to RPA 184, the steelhead aspect of the proposal may provide a viable alternative to "broodstock mining" and genetic bottlenecks for conservation hatchery programs seeking to obtain and utilize local stocks (the thrust of many reforms).
Scope? [ESU's covered, Transferability, Species covered] Targeted species are as follows. Steelhead: Southwest Washington ESU, Coho salmon: Lower Columbia River, Southwest

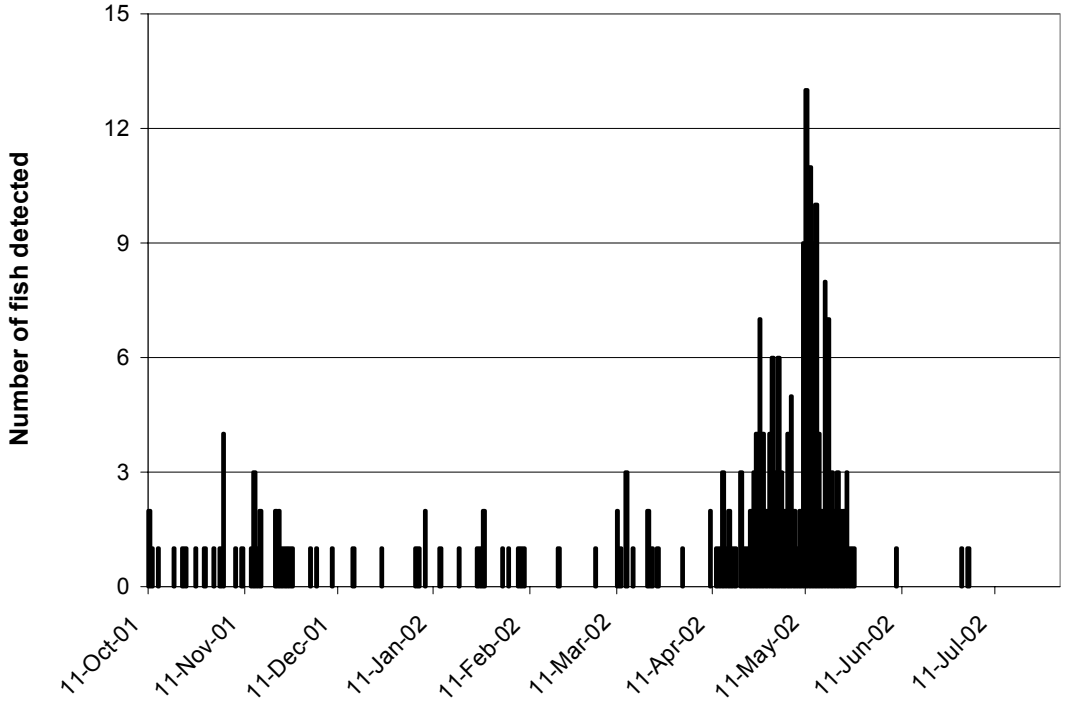
Washington coast ESU, Chinook salmon: Lower Columbia River ESU (naturalized population in Abernathy Creek). Proposal includes more than one listed species and ESU, and may have transferability to many others. As a side benefit, this technique, if successful, might have direct application to SNAPP (RPA 175)
Study design adequate, as is, or as may be modified? Well designed and written.

The following ISRP review is for the companion proposal that was reviewed as Project No. 30008 under the Columbia Estuary Provincial Review.

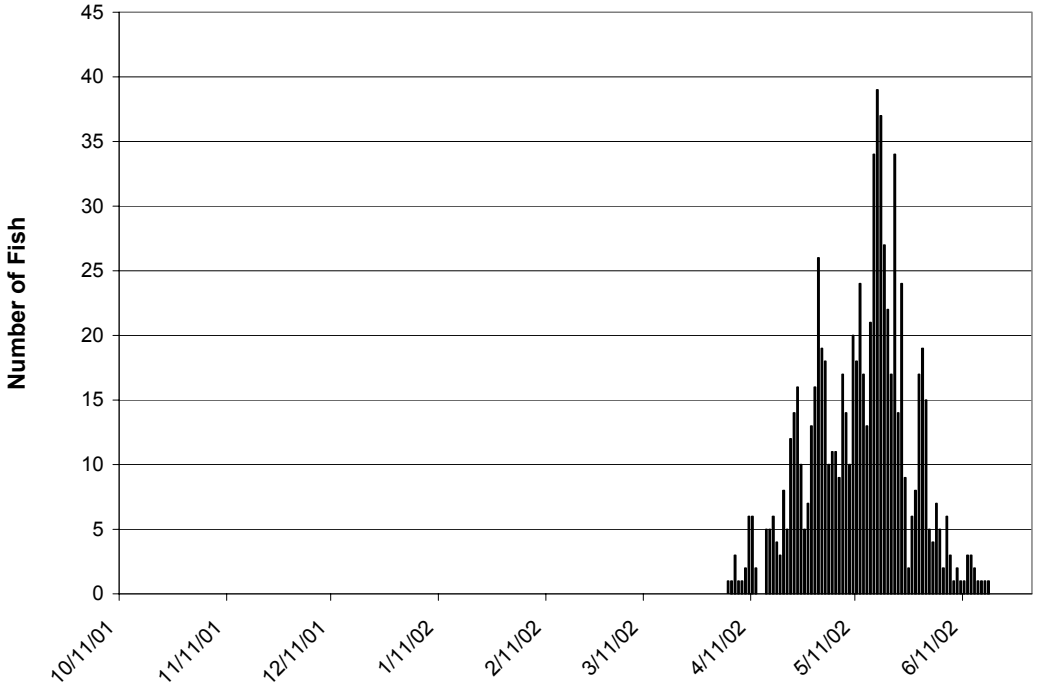
ProjectID: 35060

Instream evaluation of populations, migration, individual adult return and wild-hatchery interactions of naturally produced salmonids
Sponsor: USFWS
FY03 Request: $229,606 **5YR Estimate:** $964,645
Short Description: Evaluate stock status, distribution, and abundance of juvenile and adult salmonids using new PIT tag techniques.
ISRP Final Comments: Fundable at a medium priority. We agree with CBFWA's review and middle ranking of High Priority. A response was not needed. The proposed project seems like a logical extension of the previously funded innovative project. This is the second time we have reviewed this proposal recently. The value we see in this work is in Objective 1: to assess "abundance and natural production of juvenile, smolt and adult salmonids while developing and providing standard protocols for stock monitoring programs." If standard methods, tools, and protocols can be established for small stream assessments in the Basin, this could have significant general value outside of the immediate stream. We encourage the proponents to prioritize their work with emphasis on population assessment methods and sampling protocols. The project sponsor has made a significant contribution through her work to develop stationary remote and portable detectors for PIT tags, and is now developing a proposal to utilize that technology. However, in reviewing the objectives of this proposal, their value to the region seems limited to the development of sampling protocols for small stream assessments (useful), examination of tagging impacts on growth (assessed within a hatchery environment and with hatchery fish … limited value), or otherwise mostly of local value in Abernathy Creek. If funded, this project should be coordinated with other monitoring projects to ensure compatibility of objectives, common methods, and protocols. This coordination could be accomplished under the favorably reviewed CBFWA proposal #35033.

APPENDIX 2: Relevant results from Instream Monitoring and Evaluation: Pattern of steelhead moving past AFTC *SPIS* from Fall 2001 to Summer 2002:



Pattern of steelhead trout captured at the screw trap located at the mouth of Abernathy Creek. Dates shown are for comparison with dates of detection using *SPIS*, the screw trap was only operated from 2 April – 21 June 2002 (indicated by horizontal solid bar).



APPENDIX 3. Tasks and methods associated with Objective 1 of this proposal.

Objective 1. Develop a hatchery broodstock of steelhead by captively rearing, to sexual maturity, natural-origin juveniles from Abernathy Creek and evaluate this approach as a potential alternative to trapping natural-origin adults for developing "native" or "endemic" broodstocks.

Task 1a: Collect, via stratified sampling, 500 age 0+ steelhead from a seven mile section of Abernathy Creek in September/October 1999, 2000, 2001 (Task completed). We identified five sample site locations in Abernathy Creek for collecting age 0+ steelhead over a seven mile stretch of stream immediately upstream from the AFTC (stream miles 3.4 to 10.0). These five stream sections are approximately 1 mile apart. Approximately 100 age 0+ parr were collected with the aid of a backpack electrofisher from each of the five sample sites. Shocked fish were transferred to a holding tank supplied with oxygen in the back of a pick-up truck and maintained for up to four hours prior to transfer to the AFTC (see below). Our sampling was stratified to maximize the number of parents potentially contributing to the fish we collect

Task 1b: Transfer 400-500 age 0+ steelhead of each brood year to a separate raceway supplied with covers and Abernathy Creek water at the AFTC (Task completed). Age 0+ steelhead collected in 1999, 2000, and 2001 were transferred to 8'x 80' raceways at the AFTC within four hours after capture from Abernathy Creek. We constructed low-hanging covers that are approximately three feet above the water level in each raceway to provide shade and partial refuge from direct human interaction. These raceways are supplied with Abernathy Creek water, automatic belt feeders and demand feeders.

Task 1c: Stimulate natural-origin steelhead juveniles to feed on artificial hatchery diets. Age 0+ steelhead collected from Abernathy Creek are initially fed a starter diet coated with krill hydrolysate to stimulate initial feeding on artificial salmonid feeds. A commercial diet is used to maintain the fish during their residence in the hatchery. Feeding levels are adjusted according to total biomass and water temperatures (Abernathy Creek water) to maintain natural growth patterns. Approximately four to six months prior to spawning (late September as age 2+ fish), captively-reared fish will receive a broodstock diet (e.g. Bio-Oregon™ BioDiet Brood) to stimulate sexual maturation and proper gamete development.

Task 1d: Estimate the mean and variance in growth, and determine overall survival of captively-reared steelhead every six months. Fish will be crowded in a raceway, removed by dip net, and anesthetized with MS-222. All fish will be counted to determine overall survival during the preceding six months. A subsample of 50 fish will be measured and weighed to estimate the mean size and variance among all fish in each raceway.

Task 1e: Captively rear NOR, age 0+ steelhead to sexual maturity with the goal of spawning approximately 100 males and 100 females of each brood year. This goal

was achieved for fish collected as age 0+ parr in 1999 and spawned as age 3 adults during the winter and spring of 2002. Similar results will be obtained for fish collected as age 0+ parr in 2000 and spawned as age 3+ adults in 2003. Captively-reared fish failing to mature and spawn as three year old fish will be retained for an additional year with the goal of spawning those fish as four year old fish… **Repeat tasks 1a-1e for 3-5 consecutive years beginning with BY2000 fish collected from Abernathy Creek.**

Task 1f: Spawn captively-reared, NOR adults collected as parr from Abernathy Creek to produce F1 HOR fish. Sexually mature males and females of each captively-reared brood (collection) year will be randomly mated pairwise and spawned. Fin clips from each spawned adult will be retained for DNA genotype determination **(see Task 1f)** and subsequent pedigree identification of progeny.

We had initially anticipated crossing age 2 males (collected in 2000) with age 3 females (collected in 2001). However, for BY1999 adults spawned in 2002, insufficient numbers of age 2, BY2000 males were available for spawning with age 3, BY1999 females. In 2002, we were able to spawn pairwise 110 age 3 males and females (BY1999) to produce 110 full-sib families. Similar results are expected for BY2000 fish spawned during the winter and spring of 2003. Captively-reared, BY2000 fish not sexually mature in 2003 will be retained for one additional year for potential spawning with captively-reared BY2001 fish spawned in 2004 (Table 1).

Task 1g: Cull each F1 full sib family to equal numbers of eyed eggs with the goal of producing 20,000 yearlings (smolts) for release. Fertilized eggs from pairwise matings will be incubated and hatched in Heath trays on pathogen-free well water (53^{o} F) at the AFTC. At the eyed egg stage, each full-sib family will be culled to an equal number of eyed eggs after shocking. For BY2003, each full-sib family is being culled to 350 eyed eggs with the goal of producing 100 full-sib families. Swim-up fry will first be transferred to indoor tanks (supplied with well water) and then transferred to outside raceways in May after the previous brood year is released into Abernathy Creek.

Task 1h: Mark/tag all pre-smolts prior to release. All but 1,000 F1 fish of each brood year will be given a coded-wire tag and adipose fin clip (AD-clip) for future identification. The AD-clips will facilitate enumeration of NOR smolts during the smolt trap operations (Objective 2). The remaining 1,000 fish of each brood year will be given a 23 mm "long-range" PIT tag with NO AD-clip for evaluating post-release instream movements and remote sensing of potential residualism (Objectives 3 and 7).

We had initially planned on giving no fin clips to our released F1 fish. However, after discussions with steelhead managers for WDFW (Bob Gibbons, Chuck Johnson, and Craig Burley), we agreed to clip the adipose fin of our released F1 fish to facilitate management options when those fish return as adults. WDFW was concerned that significant numbers of surplus adults would return from our releases, and management options for potentially outplanting those surplus adults to support sport fisheries would be significantly reduced if those fish had not been clipped at release. Although we are very reluctant to release HOR fish with AD-clips when those fish are not intended for harvest, we have accepted WDFW's request and are planning to clip the adipose fin of F1 fish

released in 2004 (BY2003) and later years. Fish scheduled for release in 2003 (BY2002) have coded wire tags only without AD-clips.

Task 1i: Release 20,000 F1 steelhead yearlings per year into Abernathy Creek beginning in 2003. All progeny will be released initially into Abernathy Creek directly from the AFTC. Although volitional releases are desired, we need to coordinate our releases with downstream operation of the smolt trap by WDFW. Consequently, one-third of the fish will be forced released April 15-22, one third April 28-May 4, and the final third May 8-15.

Task 1j: Produce a handbook and protocol manual for developing native broodstocks of steelhead by captively rearing natural-origin juveniles to sexual maturity. We will document successes and failures with the goal of producing a protocol manual and general guidelines for captively rearing NOR steelhead juveniles to sexual maturity.