FY 2008-2009 F&W Program Accords (MOA) Proposal Review

Narrative

Table 1. Proposal Metadata

Project Number	2008-109-00
Proposer	Confederated Tribes of the Colville Reservation
Project Name	Resident Fish RM&E
Short Description	Spawning and overwintering movement and habitats of rainbow trout in the San Poil Subbasin
Province(s)	Intermountain
Subbasin(s)	San Poil
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Information transfer:

A. Abstract

The goal of this project is to determine what life histories of rainbow trout (*Oncorhynchus mykiss*) exist and how each life history operates within the San Poil Subbasin by examining spawning and overwintering movements and habitat use among each life history. Specific project objectives include identification of spatiotemporal patterns in spawning movements and areas used for spawning among life histories within the San Poil Subbasin, the identification of overwintering areas of rainbow trout, the investigation of how winter ecology of each present life history is associated with warm groundwater presence, river ice, and other habitat parameters and preliminary investigations of juvenile rainbow trout. The project expects to tag and track 105 adult rainbow trout and 30 juvenile rainbow trout over the study period. This critical missing information on life histories and ecology of rainbow trout will assist in determining where essential spawning and overwintering habitat exists to aid the Tribes in management decisions on where conservation and enhancement actions will provide the greatest benefit to the fish and also help determine rainbow trout management goals and objectives for the basin.

B. Problem statement: technical and/or scientific background

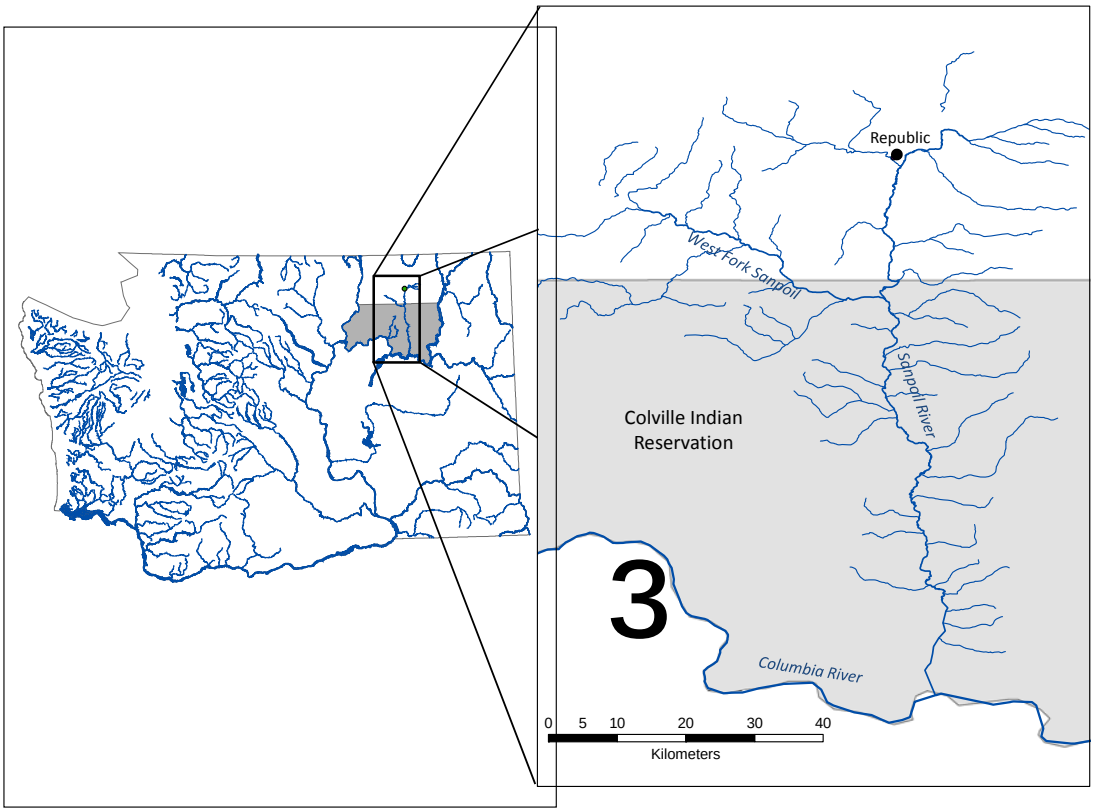
Study Area

The San Poil River is located in North Central Washington. The river flows in a north to south manner from Republic, WA to its terminus at Lake Roosevelt. Total stream length is approximately 70 miles and drains ~1,000 square miles. Thirty-seven miles of mainstem river flows through the Colville Reservation, in addition to long distances of several tributaries including Iron Creek, Louie Creek, N and S Nanamkin, Bridge Creek, Bear Creek, 13 Mile, 17 Mile, 23 Mile, 30 Mile, Gold Creek and West Fork San Poil. The stream bottom is mostly gravel and small cobble with lesser amounts of boulder, rubble and silt. Fifty-five percent of the tributaries that were once perennial are now intermittent (Duck Creek Associates 2007). The river is surrounded by mountains and has a valley approximately one mile wide. Land use practices in the basin include agriculture, logging, mining, and cattle grazing and the impacts can be observed in the San Poil through cemented sediments.

The San Poil River before the construction of Grand Coulee Dam supported a large run of summer and fall Chinook and was famous for its summer steelhead runs. Today the river contains mostly rainbow trout, eastern brook trout (*Salvelinus fontinalis)*, mountain whitefish (*Prosopium williamsoni*) and in certain years kokanee (*Oncorhynchus nerka*) escape into the river. A few coastal cutthroats (*Oncorhynchus clarki clarki*) have been observed. Stocking in the past has mainly consisted of coastal rainbow trout, redband rainbow trout and eastern brook trout.

The specific study area for this study is the mainstem San Poil River on the Colville Reservation along with the 12 tributaries listed above (Figure 1).

Figure 1. Map of San Poil Subbasin



Status of Rainbow Trout in the San Poil

The Colville Tribe has been collecting baseline information on the rainbow trout in tributaries to the San Poil River since 1990 through the Lake Roosevelt Rainbow Trout Habitat Improvement Project. Much of the focus has been towards identifying and improving habitat while minimally assessing the abundance of adult and juvenile migratory (adfluvial) fish in relation to habitat improvements. Detailed information on the population dynamics has never been collected especially relating to life history characteristics. Some of the difficulty in understanding the ecology of rainbow trout in this basin is due to the varied life history types thought to exist. Some adult fish migrate into the drainage from Lake Roosevelt (in both the spring and summer) to spawn while others are thought to be resident in the drainage year round. Some juvenile rainbow trout are known to migrate out of the drainage into Lake Roosevelt in the spring, but others may migrate in the summer or fall or may reside in the drainage all year.

Some research has been conducted on migratory populations of rainbow trout that use the drainage. Spawners return from Lake Roosevelt to tributaries between ages 3 and 6 years. Upstream traps have been used to examine these fish; these data indicate that spawning populations have fluctuated from less than 13 individuals in 1997 (5 tributaries surveyed) to 428 individuals in 2007 (6 tributaries surveyed) (Sears 2006). Fall escapement surveys conducted from August to November (kokanee monitoring) indicate that rainbow trout migrating upstream into the drainage range from 10-15 individuals per year (CCT unpublished data). However the research efforts mentioned above do not supply a complete picture since data are not available from periods when traps were not operated or when surveys were not conducted. In addition, no surveys have been conducted on the mainstem San Poil River.

Adult rainbow trout abundance is thought to be low compared to the number of miles of habitat available in the San Poil Basin. To alleviate this problem, the Tribe has conducted habitat improvements to some tributaries and removing barriers. These improvements have increased rainbow trout abundance where they never occurred or minimally occurred (Sears 2006). Specifically, passage and habitat enhancements have been conducted on Louie, Iron, South Nanamkin, North Nanamkin, Bridge, Thirty Mile, Thirteen Mile, Twenty-Three Mile, Gold, and Roaring Creeks. Rearing habitat remains restricted due to intermittent flows in the lower reaches of several of the streams that have spawning habitat. In recent years work to improve flows, reduce sediment delivery and increase water retention in the individual watershed has included road abandonment, riparian plantings, cattle exclusion fencing, and relocation of beaver into upper reaches of the watersheds.

Unfortunately, little is known about the populations that reside the entire year within the drainage. A few populations have been observed in Bridge Cr., West Fork San Poil and Gold Cr. during fall habitat surveys. It was determined that the Bridge Cr. fluvial population was genetically pure and has recently become part of the Tribes brood stock

program. Although, some spawning surveys have been conducted to identify spawning areas, these were done only in the lower reaches and were directed at the adfluvial population that is the focus of the LRHIP. Additionally, high velocities and high turbidity limited any observations of redds during surveys in the lower tributaries and San Poil mainstem (Sears 2006).

Juvenile rainbow trout survival and abundance is also not well understood within the basin. Surveys of juveniles in select tributaries suggest that their abundance is low. In the spring of 1998, 19 adult rainbow trout moved upstream into North Nanamkin Creek. During the next spring, a total of 96 juveniles were captured in downstream traps (Jones 1998). Six segments of Gold Creek (a tributary to West Fork San Poil) were electro-fished in the fall of 2004 and a total of 1,134 juvenile rainbow trout were captured. During the following spring of 2005 a screw trap was placed at the mouth of the West Fork San Poil that captured only 119 juveniles; additional juvenile traps were set in the stream the same year and captured 606 juvenile fish (Sears 2005). Three distinct age classes were present (young of year, one year "parr" and two year "pre-smolt") and consisted mainly of age 1 fish. These data indicate however that juvenile abundance may decrease between fall and spring (Kirk Truscott, Colville Confederated Tribes, Personal communication), or that the majority of fish did not migrate out of the West Fork during that time. However, environmental factors and trap efficiency limit an accurate assessment of the numbers of fish in the drainage, when they migrate, or how many migrate when. Juveniles may migrate in late fall or early spring before traps are set. Juveniles have been documented migrating into mid August (Sears 2000) however recent surveys have been limited to June due to low flows and occasionally extremely high flows. Attempts were made to determine egg to fry survival with red caps but high flows blew the traps out and future attempts were abandoned.

Intermittent streams such as N. Nanamkin and S. Nanamkin provide good spawning habitat for adults however nursery habitat becomes limited by mid to late summer as flows decrease or go subsurface. Juveniles are then subjected to migrating to the San Poil River or reside in a few left over pools.

Management objectives for the San Poil River are to increase adfluvial rainbow trout abundance to support recreational and subsistence harvest (i.e., 1 fish/hr) while maintaining a genetically diverse and natural reproducing population (CCT Fish and Wildlife Management Plan, 2007). However adult spawner abundance objectives have never been determined because limited information exists on each life history type. The Tribe suspects that the bulk of the population is adfluvial with some resident fish. Resident fluvial populations have been found above barriers in many of the tributaries. Because these were not the focus of the habitat improvement project little data has been collected on these populations beyond genetic analysis. Until the Tribe determines what life history forms exist, the extent of their movements and habitats used, what population levels are and how much habitat is available, developing accurate objectives is unattainable. The current proposed study addresses adult and juvenile life history information and habitat use information that will help the Tribe with determining appropriate management actions. Further investigations into juvenile survival are

proposed currently through additional resources. Information collected from the proposed study and other studies will be used to develop new studies that address limiting factors identified in previous years and assist in making management decisions.

Technical Background

At least two genetically distinct subspecies of rainbow trout are present within the San Poil Subbasin. Redband trout are native to the drainage, while coastal rainbow trout were introduced to the area (Gillin and Pizzimenti 2004). The native redband may be genetically similar to the native, summer steelhead populations that were once abundant within the system (Leary 1997, cited by Gillan and Pizzimenti 2004). Although genetic testing has revealed introgression among subspecies, genetically pure redband trout still exist above barriers (Leary 1997, cited by Gillan and Pizzimenti 2004). Young et. al. (2007) indicated that greater than 75% of the adfluvial rainbow trout in the San Poil system are non-hybridized redband stocks. This is much lower degree of hybridization than had been previously anticipated.

Much of the data collected on the different strains of rainbow trout within the San Poil Subbasin has not been separated by subspecies. Thus, the use of the term rainbow trout hereafter does not specify one subspecies or the other, unless otherwise noted. Although rainbow trout are known to be both resident and migratory within the San Poil Subbasin (Gillin and Pizzimenti 2004), there is a general lack of knowledge regarding how many and what life history strategies exist within the subbasin. In general, fluvial trout occupy streams for their entire lives (Northcote 1997) while migratory trout travel to spawn in streams that flow into lakes (lacustrine-adfluvial; Varley and Gresswell 1988; Northcote 1997), that flow out of lakes (allacustrine), or that move from rivers into tributaries (fluvial-adfluvial) to spawn (Northcote 1997, Dupont et al. 2007). This proposal addresses a lack of information for both resident and migratory rainbow trout within the San Poil River Basin. Although the abundance of resident rainbow trout in the San Poil River Basin that exhibit the fluvial versus fluvial-adfluvial life history strategy is currently unknown. The population that, for the purposes of this proposal, rainbow trout that remain within the San Poil River or tributaries throughout their life history are hereafter referred to as fluvial and rainbow trout that migrate from Lake Roosevelt to the San Poil River or tributaries are hereafter referred to as lacustrine-adfluvial.

Rainbow trout tributary spawning habitat is limited within the San Poil Subbasin (Gillin and Pizzimenti 2004) and early fisheries investigations indicated that a lack of high quality spawning and rearing habitat was a limiting factor to adfluvial rainbow trout production in Lake Roosevelt (Scholz et al. 1986, cited by Gillin and Pizzimenti 2004). Further, introgression has been documented between redband and coastal rainbow trout in areas of the Colville National Forest below barriers (Gillin and Pizzimenti 2004) however, lacustrine-adfluvial rainbow trout located within the Colville Reservation showed minor introgression between both stocks with high genetic diversity (Young et. al. 2008). Thus, the first objective of this study is to document spatiotemporal patterns in spawning areas and movements among life histories within the San Poil Subbasin. We

hypothesize that rainbow trout have several different life history strategies within the San Poil Subbasin. Radio telemetry will be used to examine spatiotemporal trends in spawning behavior to allow their life histories to be documented.

In addition to the lack of information on the various life histories of the spawning populations in the basin, little information exists regarding the winter ecology of rainbow trout within the San Poil Subbasin. We intend to collect data with this study that will identify if and how winter habitat is limiting the rainbow trout with in the San Poil Subbasin. Recruitment of adfluvial rainbow trout into Lake Roosevelt is likely highly dependent on surviving the first winter in spawning tributaries or the mainstem of the San Poil River. Furthermore, abundance estimates decline between the fall and spring within the San Poil Subbasin (Kirk Truscott, Colville Confederated Tribes, Personal communication) based on fall abundance estimates and spring migration surveys. As water temperatures decrease in fall and early winter, the metabolism of fish decreases and defense of feeding positions becomes less important to fish while the search for suitable winter habitat becomes more important (Cunjak and Power 1986; Cunjak 1996; Lindstrom and Hubert 2004). This leads to shifts in habitat use and movements, and many larger juvenile and adult fish abandon feeding territories and aggregate in areas where they can find winter refuges (Hartman 1965; Cunjak and Power 1986; Brown and Mackay 1995a; Jakober et al. 1998). Smaller fish may exhibit similar behavior; however, smaller fish likely move shorter distances and hide within interstitial spaces of the stream bottom during the day (Hartman 1965, Griffith and Smith 1993). Seasonal shifts in habitats usually involve larger fish moving to areas with lower velocities and greater depths. As water temperatures decrease in the fall, fish such as riverine salmonids often make lesser use of shallower areas with higher water velocities and greater use of deeper, slower habitats (Hartman 1965; Cunjak and Power 1986; Chisholm et al. 1987; Baltz et al. 1991; Heggenes et al. 1993; Brown and Mackay 1995a; Jakober et al. 1998). Therefore, the likelihood that trout will be found in aggregations in rivers and streams increases as water temperature decreases (Brown 1999). The occurrence of winter aggregations of fish is correlated not only with water temperatures, but also the inflow of relatively warm groundwater into the water column (Brown and Mackay 1995a; Brown 1999). Although aggregating may decrease the risk of individual predation (Pitcher 1986), it may leave fish more vulnerable to human disturbance or angling and parasite or disease outbreak since large numbers of fish are located in a confined area.

The areas fish aggregate in during the fall may not represent areas used for the entire winter. Winter habitats of fish can range from very stable (areas insulated from thermal extremes) to constantly in flux due to changes in river ice and water temperature. In some river environments, the solid surface ice cover formed early in the winter seals the fish under a stable sheet of ice. Further, snow can bridge small streams and provide stable overwintering habitats (Chisholm et al. 1987; Hubert et al. 2000). As snow accumulates on the surface ice of pools, habitat stability appears to increase (Lindstrom and Hubert 2004). However, habitats can also be unstable during the winter due to the presence of groundwater inflow or dynamic river ice conditions (Brown 1999; Lindstrom and Hubert 2004; Barrineau et al. 2005). Anchor ice can form over large parts of river channels forcing fish to move (Brown and Mackay 1995a). However, in smaller streams,

snow can bridge the entire stream insulating it from super-cooling events and thus frazil and anchor ice formations. Habitats in these areas will be more stable and fish may move less than those in areas influenced by frazil and anchor ice (Brown 1999).

Winter ecology is an important yet often overlooked aspect to fisheries management that may represent capacity limiting factors for many populations. Little is known about the winter ecology of rainbow trout within the San Poil Subbasin. Therefore, an objective of this study is to identify overwintering areas of rainbow trout within the San Poil Subbasin and investigate how winter ecology of each present life history type is associated with warm groundwater presence, river ice, and other habitat parameters to determine optimal areas for habitat protection and enhancement. We hypothesize that larger juvenile and adult rainbow trout will aggregate in areas with warm groundwater influx and moderate to deep water depth. Identification of warm groundwater areas will not only provide information on valuable winter habitats, but these groundwater areas can also provide thermal refugia for fish during summer when ambient water temperatures reach high levels.

C. Rationale and significance to regional programs

Rainbow trout are a focal species in the San Poil Subbasin under the Intermountain Province (IMP) Subbasin Plan due to their recreational value as a sport fish and their cultural significance to the Colville Confederated Tribes (CCT) (Gillin and Pizzimenti 2004). The first priority for the aquatic objectives in the San Poil Subbasin is to begin implementation of habitat strategies for addressing identified limiting factors for all focal species and native fishes. However, the limiting factors that are listed as being addressed by this objective are limited to riparian habitat, water quality, nutrients, and sediment (Gillin and Pizzimenti 2004). Little published information exists regarding other parameters that may be limiting to one or more life histories of rainbow trout within the San Poil Subbasin despite the identification of other limiting factors being listed as a specific strategy within the subbasin (Gillin and Pizzimenti 2004). The Quality Habitat Assessment (QHA) methodology used to assess limiting factors for the blocked areas of the Upper Columbia during the development of the 2004 subbasin plans did not provide for analysis of habitat type, function or migration. Therefore, the proposed research is designed to examine spawning and overwintering movements and habitats used by all life histories of rainbow trout present within the subbasin. This will aid in identifying other possible limiting factors within the subbasin (i.e., spawning and winter habitat) that may prove critical in meeting the second highest ranked objective (i.e., Objective 2A2) for the San Poil Subbasin: protect and enhance redband trout populations and preserve their genetic integrity while maintaining their subsistence and recreational fishery (Gillin and Pizzimenti 2004). Enhancement of redband populations may also be important as extirpated steelhead runs were of the redband subspecies (Behnke 1992). It may be possible to recover steelhead in the future with fish passage through Chief Joseph and Grand Coulee dams. Thus, it is important to preserve redband trout not only because of

their cultural significance and native species status, but also because they may provide a native donor stock for future anadromous reintroduction.

D. Relationships to other projects

Stream surveys identified fish passage barriers as limiting production within the San Poil River (Gillin and Pizzimenti 2004). Objective 1B1 for the San Poil Subbasin is to inventory all barriers within the subbasin and to begin implementing necessary passage improvements associated with man-made barriers (Gillin and Pizzimenti 2004). The CCT Lake Roosevelt Rainbow Trout Habitat/Passage Improvement Project (LRHIP) addresses this objective and has completed the inventory. The proposed research will provide supplementary data regarding fish passage barriers and the efficacy of recent fish passage improvements through extensive radio-telemetry operations; albeit, the assessment of fish passage is not a direct objective of the proposed research. The use of telemetry will allow us to examine movements of fish past areas that are thought to be barriers or past areas that were modified to improve passage. Although a large part of the rainbow trout may not be examined in these areas, the fish that are implanted with transmitters should provide an indication of whether these areas are passable.

The LRHIP conducts habitat and passage enhancements for rainbow trout and the Chief Joseph Kokanee Enhancement Project (CJKEP BPA# 199501100) has started addressing these issues for kokanee. The results of the studies will aid in evaluating habitat improvements completed by the LRHIP and CJKEP. Results will also identify additional passage barriers, habitat improvement needs or critical habitat protection opportunities currently unknown. This information will be passed on to the projects listed above for conservation actions.

The Colville Tribal Hatchery Project (BPA# 198503800) currently releases and monitors redband rainbow trout into tributaries of the San Poil River. The stock origin is a mixed stock of San Poil Basin and Phalon Lake. Factors such as time of release, migratory patterns and habitat have been identified as limitations and/or critical unknowns for the survival of these releases (Ed Shallenberger, CCT, Personnel communication). During the next fiscal cycle, this project proposes a creel survey on the San Poil River. Information gathered from this survey could be useful to this project. The proposed project will help identify limiting factors and management strategies specifically designed to restore and enhance redband rainbow trout populations (Objective 2A2) in the San Poil Basin.

The Chief Joseph Kokanee Enhancement Project (BPA# 199501100) is tasked with enhancing kokanee in the San Poil Basin. The project currently monitors a permanent resistance weir near the mouth of the San Poil which will be used to capture migratory rainbow trout. In addition, a habitat survey will be conducted this summer (09) and information obtained from this will be used in the proposed study to determine potential overwintering areas. The Kokanee project will also donate the use of telemetry receivers for fixed sites and mobile surveys currently used in kokanee spawning surveys.

Relationship to existing projects

Funding Source	Project #	Project Title	Relationship (brief)
BPA	199001800	Lake Roosevelt Habitat/Passage Improvement Project	Provides fish movement data pertinent to current fish passage barriers and through areas where passage and habitat improvements have been made. The proposed project will target areas for protection and enhancement actions by the LRHIP and CJKEP projects. Project will share equipment as needed. LRHIP is planning on installing radio-telemetry tags in out-migrant adfluvial RBT and post spawn adults to track movement and will be using the receiver array established with this project to track the tagged fish.
BPA	19850380	Colville Hatchery	Identifying habitat types and areas where stocked fish would have the highest of survival. Project will share equipment as needed.
BPA	19950110	Chief Joseph Kokanee Enhancement	CJKEP provides fish capture assistance and habitat analysis. Project will share equipment as needed.

E. Project history (for ongoing projects)

This is a new (proposed) project consistent with the Accords agreement between CCT and BPA for RM&E under the Resident Fish Projects.

F. Proposal biological/physical objectives, work elements, methods, and metrics

Objectives:

Objective 1. **Identify spatiotemporal patterns in movements and spawning areas among life histories within the San Poil Subbasin.**

Task 1.1. Identify all life histories of rainbow trout within the San Poil Subbasin.

Methods. Life histories will be identified through field observations and the use of radio-telemetry. Specific life history types thought to be present within the subbasin include fluvial redband trout located above barriers to migration, fluvial-adfluvial rainbow trout located within the San Poil River or tributaries, spring migrating lacustrine-adfluvial rainbow trout, and summer migrating lacustrine-adfluvial rainbow trout. Fluvial redband trout will be identified based on their location above barriers to fish passage. Spring and summer migrating lacustrine-adfluvial rainbow trout will be identified based on migration timing and movement data.

Migration timing will be evaluated using catch information from the weir located near the mouth of the San Poil River. Movement will be evaluated using radio-telemetry data (see below and schedule of activities in Table 1). Fluvial-adfluvial rainbow trout will be identified as fish captured within the San Poil River or tributaries during the winter and validated using movement data from radio-telemetry (see below). Information collected from adult surveys will guide researchers to juvenile rainbow trout nursery areas in 2012. Genetic samples will also be taken from all adult fish implanted with radio transmitters. Although analysis of these samples is not within the scope of this study, the samples will be analyzed along with samples collected by the Lake Roosevelt Habitat Improvement Project's as part of the Project's ongoing redband genetic work to examine if possible genetic differences exist among the different life history types. Samples and data will be made available to the Lake Roosevelt Evaluation Program as part of a broader rainbow trout investigation beginning in 2010.

Task 1.2. Examine timing and location of spawning for all life histories of adult rainbow trout and examine juvenile movements in the fall within the San Poil Subbasin.

Methods. Radio-telemetry will be used to assess spawning time and the location of spawning areas for rainbow trout within the subbasin. During the late winter of 2010, a combination of electrofishing, angling and snorkeling may be used to sample fluvial redband trout above migration barriers within the San Poil subbasin and to sample fluvial-adfluvial rainbow trout below migration barriers. Spring migrating lacustrine-adfluvial rainbow trout will be sampled at the continuously operated San Poil weir during their migration into the San Poil River 2010. Radio-transmitters will be surgically implanted in a minimum of 15 individuals from each life history (i.e., fluvial, fluvial-adfluvial, and spring migrating lacustrine-adfluvial) for a total of 105 adults over two years. These per season sample sizes are similar to other published research (Brown and Mackay 1995b).

Movements of radiotagged individuals will then be recorded using both passive and active radio-telemetry. Passive telemetry will be conducted using autonomous telemetry stations placed at the mouths of eight tributaries to the San Poil River. Autonomous stations will be erected in March of 2010 and maintained until March of 2012. Each autonomous station will be comprised of a Lotek SRX receiver, two deep-cycle batteries, a housing unit, solar panels, and two to four Yagi antennas. When possible, antennas will be placed to provide coverage of both the main stem San Poil River and the tributary of interest. Autonomous stations will be maintained and downloaded every two weeks when individuals with active transmitters are located within the subbasin. These data will be used to determine large scale movements within the subbasin and to determine which fish are in each reach to aid in the finer scale manual tracking.

In addition, these receivers can be used to track fish implanted with transmitters from other projects such as the CJKEP and LRHIP.

Manual tracking will be conducted throughout the spawning season using a Lotek SRX receiver and Yagi antennae. General locations of individuals will be determined from vehicle and walking surveys. Precise locations will then be obtained through triangulation or by using the power function of the receiver. A global positioning system (GPS) will be used to record locations of tagged individuals and spawning areas. Visual observation (when water clarity permits) of tagged individuals will be used to confirm spawning time and location (e.g., redd construction, spawning, etc.).

Radio-transmitters will also be implanted in 15 summer migrating lacustrine-adfluvial rainbow trout during their migration into the San Poil River in July, August, and September of 2010. However, spawning movements for these individuals will be monitored during the 2011 fiscal year (FY). The aforementioned radio-telemetry operations will be repeated for fluvial, fluvial-adfluvial, and spring migrating lacustrine-adfluvial during FY2011 as well.

A total of 30 juvenile rainbow trout will be collected and surgically implanted with radio tags during the fall of 2012. Their movements will be tracked into winter. Fish locations will be marked via GPS, habitat type identified, the proportion of water surface covered by ice and snow, and the extent of subsurface ice will be recorded.

Task 1.3. Analyze data related to the timing and location of spawning for all life histories of rainbow trout within the San Poil Subbasin.

Methods. All data downloaded on radio receivers will be transferred to laptop computers and transferred to the Pacific Northwest National Laboratory for analysis. The date and time and location of fish will be placed in a spreadsheet and overlaid in a geographical information system. The distances and rates of fish movement will be determined. Migration corridors and spawning areas will be documented in a geographical information system. Life history types will be identified based upon origin of fish, extent of pre-spawning migrations, spawning areas and post-spawning migrations. Distances moved and rates of movements will be compared among life history types.

Objective 2. **Identify overwintering areas of rainbow trout within the San Poil Subbasin and investigate how winter ecology of each present life history type is associated with warm groundwater inflow, river ice, and other habitat parameters to determine optimal areas for conservation and enhancement.**

Task 2.1. Identify overwintering locations of rainbow trout.

Methods. During the first fiscal year of the project (2010), visual and video surveys will be conducted to identify locations of overwintering rainbow trout. One survey will be conducted in late fall (likely in early November) before the onset of ice cover. Another survey will be conducted later in the winter after ice cover is well established. Surveys will be conducted in the mainstem of the San Poil River, and in several tributaries (listed in study area). Areas above barriers where isolated redband rainbow trout are located will also be targeted.

Visual and video techniques (described in Mueller et al. 2006) will be used to identify overwintering locations. During visual surveys for this first fiscal year, a camera will be submerged into areas with preferred fall / winter habitat for larger juvenile and adult trout (Baltz et al. 1991, Brown and Mackay 1995a). Prior to placing the camera in the pool, it will record the face of a GPS to catalog the position. Video will be recorded and later examined for the number and life stage of rainbow trout. Video surveys in the late winter will focus on areas where fish were identified during the late fall surveys, areas adjacent to these with preferred habitat, and areas of preferred habitat in stream reaches influenced by warm groundwater.

Having known locations of overwintering fish is valuable for other objectives of this study. These fish can be implanted with transmitters in late winter and tracked to spawning areas the following spring. These same areas can be targeted to look at thermal regulation during summer high temperatures.

During the second fiscal year of the project (i.e., FY2011), locations of overwintering areas through visual surveys will be augmented by tracking fish implanted with radio transmitters. Fish that were implanted in the spring or summer and that remain in the basin will be tracked to overwintering areas. Movements during the fall and winter will be monitored at least every two weeks. Fish will be manually tracked and their overwintering locations documented and logged using a Global Positioning System (GPS). The extent of groundwater (length of stream above ambient stream temperatures) and river ice at the locations of the fish will be determined. In addition, if fish moved since their previous tracking, the groundwater and river ice conditions will be described at their previous location to possibly determine why it was vacated (for example if it is chocked with anchor ice). Video equipment will also be used to examine the number and life stage of rainbow trout present at these locations. So that fish are not frightened, and movement or habitat use biased, small holes in the ice will be made in the margins of the pools. A two inch ice auger will be used to make a hole large enough to insert a small video camera so that fish presence and abundance can be monitored (see Mueller et al. 2006 for details and equipment). This will also help ensure that all tagged fish are alive.

Task 2.2. Quantify the extent and thermal properties of warm groundwater areas

Methods. Warm groundwater areas will be located because they can prevent river ice formations during the winter and can be a source of winter refugia (Power et al. 1999). These areas may also be important because the use of habitats can be much different when groundwater is available versus when streams are ice covered or contain anchor ice. Groundwater surveys will be conducted in the fall and will be supplemented with observations made during other surveys and through the tracking of fish implanted with transmitters. After river ice begins to form in fall, areas lacking ice cover will be identified as starting points for finding warm groundwater effluents. The length of open water areas will be mapped using a GPS and GIS system. Length and width measurements will be determined by measuring the distance between each GPS mark. Within potential groundwater areas, temperatures will be recorded every 100 -500 m, depending on the length of the area influenced by groundwater effluent using temperature loggers. Outside air temperature will be recorded at each site using a temperature logger. Changes in temperature in these areas will be re-evaluated on subsequent surveys and the extent of ice encroachment and anchor ice formation in these areas will be examined (proportion of water surface covered by surface ice, proportion of ice covered by snow, ice thickness and mean snow depth).

Task 2.3. Analyze data related to overwintering locations of rainbow trout and warm groundwater areas within the San Poil Subbasin.

Methods. All data downloaded on radio receivers will be transferred to laptop computers and transferred to the Pacific Northwest National Laboratory for analysis. The date and time and location of fish will be placed in a spreadsheet and overlaid in a geographical information system. The distances and rates of fish movement will be determined. Migration corridors and overwintering areas will be documented in a geographical information system.

Locations of surveyed warm groundwater areas will be entered into a geographical information system. The location and temperature of groundwater at each location will be organized in a spreadsheet. Locations with observed fish will also be associated with these data.

Table 1. Proposed research schedule in the San Poil Subbasin.

FY	Date	Fluvial	Fluvial-adfluvial	Spring lacustrine-adfluvial	Summer lacustrine-adfluvial	Juvenile	Winter survey
2010	Oct-09						
	Nov-09						
	Dec-09						
	Jan-10						
	Feb-10	TAG	TAG				
	Mar-10			TAG			
	Apr-10						
	May-10						
	Jun-10	T	T				
	Jul-10	R	R	T	TAG		
	Aug-10	A	A	R			
	Sept-10	C	C	A			
2011	Oct-10	K	K	C			
	Nov-10			K	T		
	Dec-10				R		
	Jan-11				A		
	Feb-11	TAG	TAG		C		
	Mar-11			TAG	K		
	Apr-11						
	May-11						
	Jun-11	T	T				
	Jul-11	R	R	T			
	Aug-11	A	A	R			
	Sep-11	C	C	A			
2012	Oct-11	K	K	C		TAG	
	Nov-11			K		TRACK	
	Dec-11						
	Jan-12						
	Feb-12						
	Mar-12						
	Apr-12						
	May-12						
	Jun-12						

This timeline does not show any scheduled data analyses or writing reports, etc.

G. Monitoring and evaluation

The product of this research will be information on movements (and locations) used by rainbow trout for spawning, feeding and overwintering. No historical data relating to this goal currently exists on the Colville Reservation and more specifically the San Poil River. These data will be used as a management tool to set management objectives and to determine where habitats should be protected or improved to increase the population of rainbow trout in the San Poil Subbasin. Additionally, results will aid in evaluating habitat improvements completed by the Lake Roosevelt Rainbow Trout Habitat Improvement Project (LRHIP) and Chief Joseph Kokanee Enhancement Project (CJKEP). Results will also identify additional passage barriers, habitat improvement opportunities or critical habitat protection opportunities currently unknown. This information will be passed on to the projects listed above for conservation actions. The Colville Tribes Fish and Wildlife will prioritize each action for future implementation. Once these actions are implemented, the rainbow trout population will be monitored and evaluated through the CJKEP and LRHIP using juvenile and adult migration traps to determine the significance of each action through abundance monitoring. In addition, habitat improvement actions will be monitored and evaluated based on pre-treatment, implementation, and effectiveness checklists that will allow the Colville Tribe to determine if a project was implemented correctly and met stated goals.

The proposed sample sizes of radio-tagged rainbow trout are justified as the number of fish implanted with transmitters is similar to or greater than those found in similar published studies. Movements may be compared among groups of fish or among years of research. Similar to other published research on these topics, comparisons between groups or years will likely be conducted with t-tests or Mann-Whitney U tests and comparisons among groups or years will likely be conducted using ANOVA or Kruskal-Wallis tests, depending on the normality of the data.

Juvenile survival from fall to spring and from juvenile to adult will be studied and funded through additional sources.

Results from this study will be communicated to BPA and Colville Tribes Fish and Wildlife through annual reports, and if appropriate, publication in peer-reviewed journals. Quarterly status reports will be submitted to BPA through PISCES.

H. Facilities and equipment

PNNL will provide the necessary office space and computer and office support for this project. Much of the video equipment needed for field work is available from PNNL at no cost to this project. Equipment purchases that will be required include radio-transmitters, receivers, and components for autonomous stations. Trucks and snow machines will be rented when necessary and the tribe will donate the use of ATV's and other equipment at no cost. Attempts will also be made to transfer or loan equipment

such as radio receivers owned by the US Army Corps of Engineers to BIA for use on this project. PNNL and Colville Tribe currently have the digital thermometers, thermisters, flow meters, dry suits, snorkeling gear, waders and other miscellaneous equipment necessary to complete the project.

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J. Key personnel

Bret Nine, Colville Confederated Tribes, Fish and Wildlife (principal investigator)

Dr. Richard S. Brown, Pacific Northwest National Laboratory (principal investigator, project manager, subcontractor)

Eric W. Oldenburg, Pacific Northwest National Laboratory (project manager, subcontractor)

Sheri Sears, Colville Confederated Tribes, Fish and Wildlife, Resident Fish Division Manager

Bret Nine, Colville Confederated Tribes, Fish and Wildlife

Education:

Master of Science, March 2005: Fisheries, Eastern Washington University
Bachelor of Science, March 2003: Zoology, Eastern Washington University

Work Experience:

July 2006 – Present: Fisheries Biologist III, Colville Tribes Fish and Wildlife, Inchelium, WA.
January 2006 – July 2006: Lead Entity Coordinator (Salmon Recovery) for Pend Oreille County, WA.
May 2005 – January 2006: Independent Contractor for Pacific Northwest National Laboratory, Richland, WA
April 2003 – March 2005: Graduate Assistant. Eastern Washington University, Cheney, WA.
December 2002 – October 2004: Research Assistant for Pacific Northwest National Laboratory, Richland, WA.
January 2001 – December 2002: Fisheries Technician, WDFW and EWU.

Research Experience:

- Distribution of non-native fishes on the Turnbull National Wildlife Refuge. Eastern Washington University.
- Genetic characterization of wild origin kokanee in Lake Roosevelt. Colville Tribes Fish and Wildlife and Washington Department of Fish and Wildlife.
- Assessment of the movements and survival of adult and juvenile bull trout below and above Albeni Falls Dam, Pend Oreille River, Idaho. Eastern Washington University and Pacific Northwest National Laboratory.
- Effects of strobe lights on prey species of zooplankton and the relative level of opportunistic feeding that occurs when the prey species are illuminated by strobe lights. Pacific Northwest National Laboratory.
- Fishery and limnological survey of an oligotrophic lake in northeast Washington. Eastern Washington University.

Duties:

Currently is the Project manager for the Chief Joseph Kokanee Enhancement Project, Lake Roosevelt Fisheries Evaluation Project and Sturgeon Recovery Project for the Colville Tribes Fish and Wildlife Department. Primary responsibilities include: management of all assigned fishery projects, planning, developing, designing, and oversight of professional biological studies, research, or resource assessments and completes analysis for written reports and/or publication. Participates as a Lake Roosevelt co-manager and helps develop management plans that work towards measureable goals and objectives. For BPA projects, develops and uploads SOW, budgets, inventories and quarterly and final reports into PISCES and coordinates these and other activities with COTR.

Richard S. Brown, Ph. D, Pacific Northwest National Laboratory

Dr. Brown has extensive experience studying riverine salmonids and is a leading expert in the field of winter ecology. He has years of experience refining methods to observe and study fish during the very challenging winter season. Also, he has a major interest in the interactions between fish and groundwater and examining the different challenges fish face during winter in groundwater influenced habitats as compared to ice covered areas. Dr. Brown also has nearly two decades of experience using radiotelemetry. Dr. Brown has extensive experience researching spawning behavior and migratory patterns of salmonids. He is also a leading expert on techniques used to implant transmitters in fish and on tag effects.

Education:

Doctor of Philosophy, December 1999**:** Biology, University of Waterloo, Waterloo, Ontario

Master of Science, June 1994: Zoology, University of Alberta, Edmonton

Bachelor of Science, December 1988: Wildlife and Fisheries Sciences, South Dakota State University, Brookings

Work Experience:

2002 – present Senior Research Scientist, Ecology Group, Pacific Northwest National Laboratory

2000-2002 Post-doctoral fellow, Ecology group, Pacific Northwest National Laboratory, Richland, WA.

1993-96 Owner, FRM Environmental Consulting Ltd., Edmonton, AB

Most relevant publications

Brown, R. S. 1999. Fall and early winter movements of cutthroat trout, *Oncorhyncus clarki*, in relation to water temperature and ice conditions in Dutch Creek, Alberta. Environmental Biology of Fishes 55:359-368.

Brown, R. S. and W. C. Mackay. 1995. Fall and winter movements of and habitat use by cutthroat trout in the Ram River, Alberta. Transactions of the American Fisheries Society 124:873-885.

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Power, G., R. S. Brown, and J. G. Imhof. 1999. Groundwater and fish - insights from northern North America. Hydrological Processes 13:401-422.

Eric W. Oldenburg, Pacific Northwest National Laboratory

Academic Background

- Master of Science in Biological Sciences. Montana State University, Bozeman, Montana. 2008.
- Bachelor of Science in Biological Sciences. Montana State University, Bozeman, Montana. 2004.

Employment

2007-present: Fisheries Research Scientist, PNNL, Richland, WA.
2005-2007: Graduate Research Assistant, M.S., MSU, Bozeman, MT
2003-2004: Fisheries Research Technician, Montana State University, Bozeman, MT
2002: Fisheries Fieldworker, Montana Fish, Wildlife & Parks, Lewistown, MT
1998-2001: Range Technician, U. S. Fish and Wildlife Service, multiple locations

Awards

- C.J.D. Brown Memorial Award, Montana State University (2004)

Professional Societies

- American Fisheries Society (AFS; 2003-present)
- Montana Chapter of the American Fisheries Society (2003-2007)
- Montana State University student sub-unit of the AFS (2004-2007)
- Vice President, MSU AFS (2005)

Research

- Comparative performance of acoustic tagged and passive integrated transponder tagged juvenile Chinook salmon. Pacific Northwest National Laboratory, Richland, WA.
- Effects of acclimation on post-stocking dispersal of age-1 pallid sturgeon. Montana State University, Bozeman, MT.
- Trammel net efficiency for sampling juvenile pallid sturgeon and shovelnose sturgeon. Montana State University, Bozeman, MT.

Publications

Guy, C. S., E. W. Oldenburg, and P. C. Gerrity. *In press*. Conditional capture probability of Scaphirhynchus spp. in drifting trammel nets. North American Journal of Fisheries Management.

Sheri L. Sears, Colville Confederated Tribes Fish and Wildlife.

Education:
Eastern Washington University, Cheney, WA, 1995, B.S. Environmental Biology
Kaiser Foundation School of Nursing, Oakland, CA, 1972 R.N. ICU CCU Certified
Contra Costa College, California, San Pablo, CA, 1972 A.A.S. Nursing

Employment History:
July 2006 – Present ~ Colville Confederated Tribes, Resident Fish Division Manager Biologist IV

November 2001 – June 2006 ~ Colville Confederated Tribes, Lake Roosevelt Habitat Improvement Project Manager Biologist II

June 1999 – November 2001 ~ Colville Confederated Tribes, Habitat Biologist I

March 1999 – June 1999 ~ USGS – Biological Division, Rufus Woods Total Dissolved Gas Impact Assessment Project Field Biologist

September 1997 – November 1998 ~ Steven County Conservation District, Field Technician Stream surveys Steven's county watersheds.

1996 –1997 ~ Department of Ecology, ERO, Environmental Intern - Worked with Washington State Attorney General on development of Grass Seed burning ban and developed library of literature to support rule decision.

March 1995 – July 1995 ~ Waste Water Treatment Intern City of Cheney, Tested effluent and wrote manual on identification of aquatic organisms beneficial to waste water treatment.

Professional Affiliations:

2009 - Chairman of the Columbia Basin Fish and Wildlife Authorities (CBFWA) Resident Fish Advisory Committee (RFAC)
2008 – Vice Chair of the Columbia Basin Fish and Wildlife Authorities (CBFWA) Resident Fish Advisory Committee
2007 – Present - Columbia Basin Water Management Plan representative and conducted assessment for EIS on proposed actions impacts to Lake Roosevelt.
2006 – Present - CCT Representative Lake Roosevelt Managers
2008 - Present - Member of Army Corp of Engineer's (USACE) Technical Management Team (TMT) working with the Bureau of Reclamation (BOR), USACE, various tribes, and the states of Washington, Oregon, Idaho, and Montana on Columbia River and Snake River dam operations and flow coordination
2003 – Present - Wildland Fire Situation Analysis (WFSA) Team Leader
2001 – Present - Burned Area Emergency Rehabilitation (BAER) Team – Wildfire impacts to fish and wildlife and documentation of Emergency Stabilization and Rehabilitation Plans for all fires on the Colville Reservation.

2001 – Present Upper Columbia River Remedial Investigation and feasibility study (RI/FS)

Duties:

Coordinate with state, tribal, federal and international entities on a variety of aquatic and fish management issues. I have worked with other Tribal departments on the development and revision of Tribes' Hydraulic, Forest Practices Act, and Water Quality codes and the review of land use applications. Reviewed, prepared, and approved all hydraulic permit applications for activities on the Colville Reservation and have completed multiple Environmental Impact Statements (EIS), biological assessments, NEPA compliance documentation, and worked extensively with Global Information System (GIS) mapping and analysis (ArcView, ArcMap, and Terrain Navigator). Provided assistance in the development and review of the Tribe's Integrated Resource Management Plan, the EIS, Record of Decision (ROD), and the Tribe's Fish and Wildlife Management Plan.

Responsibilities include project, personnel, and budget management and daily operations of BPA project. Prepared quarterly and annual reports developed scope of work and conducted field data collection on fish migration, habitat conditions, stream surveys, water quality and statistical analysis of data. Development, management, and implementation of multiple habitat and passage projects and obtained all necessary permits and environmental compliances under BIAM 30 and National Historic Preservation Act. Developed protocols and quality assurance plans,

Appendix A

A genetic analysis of trout from tributaries on the Colville Reservation – Part 2

Sewall F. Young, Cherril Bowman, Denise K. Hawkins, Kenneth I. Warheit
WDFW Molecular Genetics Laboratory
Draft, 24 June 2008

Abstract

The Colville Tribe collected 100 putative redband rainbow trout (*Oncorhynchus mykiss gairdneri*) from streams on the Colville Reservation. We genotyped those samples and compared the genotypes with previously reported baseline microsatellite data representing Spokane Hatchery and Goldendale Hatchery coastal (McCloud) strain rainbow trout (*O. mykiss irideus*), Phalon Lake Hatchery redband trout broodstock, and Sullivan Creek westslope cutthroat trout (*O. clarki lewisi)* to determine the ancestry of the Colville samples. Our baseline data contained substantial departures from genotypic equilibrium with possible undetected ("null") alleles. The westslope cutthroat and Phalon Lake Hatchery groups also each included "private" alleles, alleles that were unique to the group, at high frequencies at multiple loci. Our analyses with the program Structure v 2.2 defined 3 genetically coherent groups in the baseline that corresponded to 1) redband lineage, 2) McCloud lineage, and 3) westslope cutthroat. The analyses of the Colville samples suggested that most were likely pure redband trout, three were possibly F_1 hybrids of redband x McCloud, and 25 were possibly backcrossed to the redband lineage. Sample sizes were too small from individual source streams to suggest spatial patterns in the estimated ancestries.

Introduction

Redband rainbow trout (*Oncorhynchus mykiss gairdneri*) and westslope cutthroat trout (*O. clarki lewisi)*, both indigenous to the Columbia River basin east of the Cascade Mountains, have coexisted in the region for many generations and are afforded taxonomic status as separate species (Behnke, 1992). The two species are close relatives and may interbreed in some settings to produce fertile offspring and backcrosses; however, redband rainbow trout and westslope cutthroat trout historically occupied different portions of watersheds, remained in effective reproductive isolation, and represent two distinctive evolutionary lineages.

Rainbow trout are popular targets for recreational anglers and many streams and lakes in Washington have received infusions of hatchery strains that descended from coastal *O. mykiss* (*O. mykiss irideus*) from the McCloud River in northern California (Crawford 1979). Redband and coastal *O. mykiss* differ in several morphological aspects (including scale counts and coloration), in biochemical-genetic allele distributions (Behnke 1992), as well as in the ecological contexts in which they evolved. Until recently, most McCloud-strain rainbow trout introductions into streams inhabited by indigenous redband rainbow and westslope cutthroat populations in the upper Columbia basin in northeastern Washington were from Spokane Hatchery or Goldendale Hatchery (Small and Dean 2007).

Self-sustaining resident trout populations reproduce, feed and grow in locally specific ecological contexts. Given enough time in moderately stable environments, natural populations evolve behavioral and physiological responses to environmental conditions that optimize their fitness. A population that is adapted to conditions in one area may

have inappropriate behavioral or physiological responses to environmental conditions in another area so that its members are maladapted when translocated to a new environment. If those responses are under genetic control, interbreeding between fish adapted to different environments can produce offspring that are poorly adapted to the environments of either of their parents.

There is wide recognition among fish managers in Washington that introductions of exogenous fish strains can harm indigenous populations by direct competition for resources (e.g. food, cover, spawning sites) and, if they interbreed, by disrupting locally evolved genotypic associations that mediate responses to environmental stimuli. That recognition has prompted initiatives to cease introductions of non-indigenous strains and switch to locally or regionally derived hatchery strains, with the hope that the locally derived hatchery strains will be better adapted to the environments where they will be planted and less disruptive to the adaptations of the local stocks. Consequently, the Washington Department of Fish and Wildlife (WDFW) has replaced the McCloud-based rainbow trout strain at Spokane Hatchery with a strain developed from indigenous redband broodstock taken from Deadman Creek in the Kettle River drainage (the Phalon Lake Hatchery strain) and the Colville Tribe is developing a redband hatchery strain from broodstock collected in Bridge Creek on their reservation (Small and Dean 2007).

The Colville tribe contracted with Washington Department of Fish and Wildlife's Molecular Genetics Laboratory (MGL) to assess the ancestry of rainbow trout samples collected from streams on the Colville Reservation. Considering the history of rainbow trout stocking, those samples might include: 1) individuals of pure McCloud strain ancestry, pure indigenous redband ancestry, or pure westslope cutthroat ancestry; 2) inter-species hybrids or backcrosses (BC_n) with westslope cutthroat and rainbow trout ancestry; or 3) intra-species redband x McCloud hybrids or backcrosses.

Hybrid detection is not difficult if the parental lineages are well differentiated, as is the case with the coastal McCloud lineage and Columbia Basin redband trout; F_1 hybrids receive half of their nuclear DNA from each parent so their genic ancestry is half from their maternal lineage and half from their paternal lineage. But hybrid detection in backcrosses (BC_n) is more difficult because the two trout lineages share alleles at many loci and the random segregation of independent loci during gamete formation introduces deviations from the mean expected frequency of parental lineage genes in post-F_1 individuals. In the BC_1 generation, on average ¾ of the genes will be from the recurrent lineage and ¼ will be from the rare lineage. If backcrossing is unidirectional, the average genetic representation of the rare lineage weakens in successive generations of backcrosses by 0.5^f, where f is the generation post hybridization, but when sampling 10 to 20 loci, as is common in salmonid microsatellite studies, individual genotypes will contain varying numbers of alleles from the rare lineage due to random combination when gametes are formed. In a sample of loci drawn from a backcrossed population, the width of the interval that contains 95% of the estimated proportions of alleles that descended from the rare lineage in backcross generation *n* depends on the number of loci sampled. We expect that the precision of the estimates of proportional ancestry should increase (ie. the interval containing 95% of the estimates should contract) with increasing

number of loci sampled. As a practical matter, in many cases we can identify F_1 hybrids with a moderate number of independent, polymorphic markers and sometimes we can identify likely low-degree backcrosses. But even with perfectly diagnostic loci that share no alleles between lineages, less than 10% of the alleles on average in BC_3 individuals will provide evidence of mixed ancestry. This dilution of the rare lineage, along with shared alleles between lineages and imperfect analysis algorithms, reduce our ability to differentiate pure strains and F_1 hybrids from backcrosses, even with 20 independent nuclear loci (Figure 1).

Small and Dean (2007) analyzed rainbow trout samples collected on the Colville Reservation by staff of the Colville Tribe and concluded: 1) that the MGL's microsatellite datasets are sufficient to identify the ancestry of trout on the Colville Reservation; and 2) that redband ancestry predominated in most areas but samples from several creeks were cutthroat trout (*O. clarki)* rather than redband. This study is an extension of the 2007 work by Small and Dean.

Methods

The Colville Tribe collected 100 putative redband trout samples from creeks on the Colville Reservation between 29 March 2005 and 30 October 2007 (Table 1). The Colville Tribe preserved the unknown-origin samples with 100% ethanol in 2 mL screw cap vials with internal labels and shipped them to MGL. Molecular Genetics Laboratory staff assigned a single collection code (07LC) to the group and then assigned sample numbers to identify individuals within the group.

We extracted genomic DNA from the tissue samples using silica-membrane spin-column kits (Macherey-Nagel Incorporated) following the manufacturer's protocols. After extraction of the DNA, we performed a polymerase chain reaction (PCR) based species identification assay to determine the maternal lineage of each of the unknowns. The assay targets species-informative nucleotide substitutions in the coxIII-ND3 region of the mitochondrial genome. Details of the assay are available on request from the MGL. We included 32 known rainbow trout (16 redband trout from Phalon Lake Hatchery and 16 McCloud-origin rainbow trout from Goldendale Hatchery) and 40 known cutthroat trout (8 westslope cutthroat from each of Sullivan Lake and Gold Creek in the Pend Oreille drainage, 8 Lahontan cutthroat from Lake Lenore, 8 Yellowstone cutthroat from the Yellowstone River Hatchery in Montana, and 8 coastal cutthroat from Cedar River in Puget Sound) as controls.

Following the maternal lineage identification, we re-examined and corrected genotypes developed in the MGL at 13 microsatellite loci (Table 2) from three reference baseline populations (Table 3) to infer likely genetic contributions from the introduced rainbow trout strains and local strains of redband trout to the unknown-origin samples. The earlier analysis by Small and Dean (2007) was based on 14 microsatellite loci, but our baseline

differed slightly from theirs and we eliminated the locus O*ki-10*, from which data was missing for half of the Goldendale individuals.
That change to the baseline prompted us to re-calculate locus and population summary statistics and re-examine the suitability of the data for equilibrium-modeling-based analyses of potential hybridization. We used Fstat v. 2.9.3.2 (Goudet 2001) to recalculate allele frequencies; to estimate allelic richness, which is a measure of allelic counts adjusted for sample size; to estimate gene diversity per-locus-within-population, also called the expected heterozygosity; and to estimate F_{IS} per-locus-within-population to check for deviations from Hardy-Weinberg Equilibrium (HWE). We summed the frequencies of alleles at each locus that were unique in the baseline to each lineage to as simple indicators of population distinctiveness.

Our molecular analysis of the unknown trout collected by the Colville Tribe included 13 of 14 loci that we PCR-amplified in 96-well reaction plates using fluorescently labeled primers following the protocols of Small and Dean (2006). We excluded the locus *Oki-10* because it was missing from one of the previously run populations in our baseline (described above). Our amplification reactions included 1 µl template DNA with final concentrations of 1.5 mM $MgCl_2$, 200µM of each dNTP, and 1X Promega PCR buffer. The multiplexed PCR thermal parameters included an initial three-minute denaturation at 92°, 33 cycles of 92° for 15 seconds, annealing for 30 seconds (see Table 3 for annealing temperatures), and amplicon extension at 72° for 60 seconds. We included a final 30-minute extension at 72°C to encourage uniform adenylation of all amplicons. We separated the PCR products by capillary electrophoresis in an ABI 3730 automated DNA Analyzer. We used GeneMapper software (Applied Biosystems) and a co-migrating size standard (GS500Liz from Applied Biosystems) to estimate amplicon sizes in base pairs and to group similarly sized amplicons into allele bins that had been constructed previously in the MGL for these loci.

We used the genotypic-equilibrium-model-based analysis software Structure v. 2.22 (Pritchard et al. 2000) to assess the genotypic affinities of individuals in the reference data and the unknown samples. The program does not use a priori information about population sub-structure in the data so its performance at clustering appropriately chosen known-origin samples (baseline groups) in the input file can provide a check on the validity of the inferred relationships between the baseline populations and individuals of unknown origin included in the analysis. The inferences about those relationships are based on the premise that the presence of individuals with mixed ancestry in a sample results in global genotypic disequilibrium and the assumption that observations of genotypic disequilibrium suggest the presence of hybrids or admixed individuals in the data set. Structure implements a Markov Chain to shuffle the genotypes in the input file through a user-specified number of cycles seeking to minimize the genotypic disequilibrium in a user-specified number of clusters (*k*). If the process results in groups of individuals with less genotypic disequilibrium than the data set as a whole, those groups might represent populations with shared ancestry. Structure's probabilities of membership in a cluster can be interpreted as estimates of proportional ancestry but other forces that affect population allele frequencies can influence those numbers.

The reliability of analyses done with Structure depends partly on our ability to detect and estimate the magnitude of deviations from equilibrium genotypic proportions. The 13 microsatellite loci that we used have substantial variation (from 6 – 40 alleles) in these populations (Table 2). The Goldendale Hatchery rainbow trout collection (01JB) that we included in our baseline had been included in more than one previous MGL project, but 50 of the individuals were lacking genotypes at the locus *Oki-10*. Increasing the sample size of the Goldendale Hatchery collection from 50 to 100 individuals should improve our estimates of allele frequencies and genotypic disequilibrium in that population, both of which are crucial to the model. We reasoned that the analytical power we would gain by increasing the Goldendale Hatchery sample size would offset the power that we would lose by eliminating data from *Oki-10* from our analyses.

Our experience has shown that Structure has trouble forming consistent groups when processing data from genetically similar stocks or when the number of clusters that we specify does not match real structure in the data. Both conditions can cause instability in the proportional ancestry inferences so that some individuals will have high inferred ancestry from one lineage after some iterations and nearly equal inferred ancestries from two or more lineages after other iterations. Furthermore, when the number of putative sources does not reflect the real structure in the data, sequential iterations can produce contradictory inferences of individual-individual ancestral affinities so that pairs of individuals will seem to be from the same lineage after some iterations and from separate lineages after others. We reasoned that the best fit of our model to the data would be manifested by consistency across iterations of within-individual inferred ancestry proportions and individual-by-individual, pairwise co-clustering or segregation.

We ran the analyses on the unknowns with $k = 2$, to represent coastal rainbow trout and redband trout; $k = 3$, to represent the Spokane Hatchery coastal strain, the Goldendale Hatchery coastal strain, and local Columbia redband trout strains; and $k = 4$, to represent those three strains plus a fourth group to account for possible mis-sampled cutthroat trout. To assess the stability of the ancestry inferences at each level of clustering, we ran the program for ten iterations at each k for 50,000 burn-in cycles and 50,000 analysis cycles and estimated the dominant proportional ancestry and its standard deviation across the ten iterations for each individual at each k. We also looked at the stability of inferred co-ancestry of individuals and collections across iterations.

We set a threshold inferred ancestry value to separate probable purebred individuals from possible backcrosses and hybrids 95% of the time. We assumed that the baseline collections from Spokane Hatchery, Goldendale Hatchery, and Deadman Creek were purebred. After 10 Structure iterations of the best-fit model using those baseline data, we had accumulated 3770 individual estimates of proportional ancestry. We adopted the fifth percentile value among those 3770 proportional ancestries as our estimate of the bound, above which 95% of Structure's ancestry estimates for purebred genotypes would fall.

We also used Genetix software (Belkhir et al. 2004) to explore genotypic relationships among the individuals in our data set. We performed factorial correspondence analyses (FCA), an ordination method that quantifies non-randomness in allelic distributions in components of inertia that are essentially normalized Chi square statistics. We plotted the inertia values that accounted for the most order in the allele distributions to provide a visual presentation of structure in the data.

Results and Discussion

All of the known-species controls were identified correctly by the mitochondrial DNA-based species identification assay except that the assay reactions failed on one Lahontan cutthroat and two coastal cutthroat (Table 4). All but one of the 07LC unknowns had rainbow trout mitochondrial haplotypes. The exception, 07LC0050, had a cutthroat trout haplotype so it was eliminated from further analyses. We were unsuccessful at generating microsatellite genotypes with at least nine loci for five of the unknown-origin samples, 07LC0002, 07LC0003, 07LC0009, 07LC0013,and 00LC0085 so they too were dropped from the analyses.

We observed more alleles in the Deadman Creek redband trout samples than in the McCloud-origin rainbow trout from Spokane Hatchery and Goldendale (Table 5). Consistent with that, the Deadman Creek redband samples have approximately twice the allelic richness of the McCloud-origin rainbow trout and the westslope cutthroat trout (Table 6).

The alleles that were unique to the Deadman Creek redband trout in our baseline data set accounted for a substantial proportion of the allele observations in that population sample. Approximately 40% of the allele observations in the Deadman Creek sample were of alleles that we observed only in that sample (Table 7). Fewer alleles were unique to the Spokane Hatchery and Goldendale Hatchery samples in this baseline, but they comprised about 18% of the allele observations in those groups in this study. The unknown-lineage *O. mykiss* that were collected on the Colville Reservation (collection code 07LC) included 71 of the 96 alleles that were unique to the redband lineage in our baseline and 56 alleles that we did not observe in the baseline. The large numbers of alleles that were unique to either the coastal strain or the redband strain reveal that the strains are quite divergent at these microsatellite loci and they suggest that these markers should provide power to examine the samples for introgression. The large number of alleles that we observed only in the unknowns suggests that the putative redband trout in Colville Reservation streams contain high genetic diversity.

On a locus-by-locus basis, when we pooled alleles that were unique to one of the two groups, the frequencies of pooled alleles that were unique to the Deadman Creek samples from 2002 exceeded 0.5 at *Omm-1070*, *Omy-77*, *One-102*, and *Ots-100* and the frequencies of pooled unique alleles exceeded 0.2 at all loci except *One-101* and *Ots-103*.

Five of the 36 locus x population F_{IS} values that we observed in the baseline populations exceeded 0.1 indicating that the baseline dataset had significant homozygote excesses (Table 8). We did not observe a consistent pattern of heterozygote deficiency that would have suggested either that one or more baseline samples or any particular locus was out of Hardy-Weinberg equilibrium.

Genepop v. 4.0 estimated null allele frequencies greater than 0.05 at two of 36 locus x population combinations (Table 9). Null alleles inflate the apparent homozygosity and disequilibrium relative to the actual values in a sample. Inflated disequilibrium might confound the equilibrium optimization sought by the program Structure.

At *k*=2, Structure allocated the Spokane Hatchery and Goldendale Hatchery baseline collections to a common 'McCloud' cluster in five of ten iterations. The Deadman Creek redband clustered with the Spokane Hatchery coastal rainbow trout in five of ten iterations but it never clustered with the Goldendale Hatchery collection.

The fluctuating affinities of the Goldendale Hatchery and Deadman Creek collections over 10 iterations of the *k*=2 model suggests that the model does not fit the population substructure and relationships in the data. We had expected consistent clustering of the McCloud lineage into a coastal rainbow trout aggregate, with the Deadman Creek redband collection standing alone. The absence of a consistent coastal rainbow trout cluster suggests that the Spokane Hatchery and Goldendale Hatchery strains have diverged substantially. The Deadman Creek redband samples were quite distinctive with regards to allelic richness and the high frequencies of 'private alleles', and we would expect those distinctive features, particularly the 'private alleles', to drive the two groups into different genotypic equilibrium clusters. Structure's failure to consistently allocate the Spokane Hatchery and Goldendale Hatchery collections into the same cluster in the *k*=2 model and to combine the Spokane Hatchery and Deadman Creek into a common cluster about half of the time was surprising. The results from the *k*=2 model suggest that it is not appropriate for analyzing the genotypic structure within our data set.

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Clustering by Structure was robust and consistent across nine of ten iterations with the *k*=3 model. The outlier iteration had a substantially lower overall likelihood of the data given the model (Ln likelihood = -19692.8) than the other nine iterations (Ln likelihood range = -18845.7 to –18831.1), suggesting that the Markov chain got stuck on a local probability peak that did not optimize the global likelihood. We consider the outlier to be an artifact of the analysis algorithm and disregard it in our further interpretation of the data. In the nine consistent iterations, Structure allocated individuals in the baseline collections to clusters that corresponded to a) a Spokane Hatchery group, b) a Goldendale Hatchery group or, c) a Deadman Creek group, and also clustered the unknowns with the Deadman Creek baseline collection although the likelihood values among the unknowns were lower than among the baseline collections. These results might indicate that the

rainbow trout collected on the Colville reservation for this analysis comprise a redband group that is more closely related to the Deadman Creek population than to either of the McCloud-origin coastal rainbow trout strains analyzed here. This interpretation is consistent with the results at *k*=4, described below.

Over 10 iterations with the *k*=4 model, the three baseline groups consistently formed separate clusters, but the unknowns also consistently formed a separate cluster except that the 23-Mile Creek samples consistently clustered with the Deadman Creek baseline group. This result might indicate that the rainbow trout collected on the Colville reservation for this analysis comprise a coherent redband group that is distinct from the Deadman Creek population except for the trout collected from 23-Mile Creek.

The relatively large numbers of alleles that are unique in this data set to either the Deadman Creek collection or the samples collected by the Colville Tribe for this analysis strongly suggests that there is considerable heterogeneity within this redband assemblage that is not captured in the Deadman Creek collection. The uniqueness of those alleles to groups within the redband assemblage might partly be an artifact of the sampling design or the sample sizes. The absence of unique 'redband alleles' from the Spokane Hatchery or Goldendale Hatchery collections is consistent with the existence of distinct coastal and redband lineages and we doubt that it is primarily due to sampling deficiencies in the hatchery collections.

The cluster membership likelihood values in the Structure output arrays depend on genic and genotypic similarities among individuals. The large numbers and relatively high frequencies of unique alleles with respect to lineage undoubtedly influence the cluster membership likelihoods. The spatial distribution of those unique alleles, coupled with small sample sizes from some streams might drive some of the patterns we see in those likelihood values. The FCA plots (Figure 2) depict relationships in two data dimensions among the baseline collections and between the baseline collections and the 'unknown' samples from the Colville Reservation. The axis titles give the percentage of total inertia that is explained by each axis and can be interpreted as the importance of that dimension in the depiction of structure. The plots show that the Spokane Hatchery and Goldendale Hatchery baseline collections are distinct from the Deadman Creek collection on the first axis and are distinct from each other on the second axis, thus the major subdivision in the data is between the coastal and inland groups. The unknowns cluster near the Deadman Creek sample, but some are between the Deadman Creek cluster and the Spokane Hatchery or Goldendale Hatchery clusters. One hundred unknowns were collected in 10 creeks over two years so the individual Creek sample sizes are small. The plots show how the samples from each creek relate to the baseline samples but due to the small sample sizes we cannot assess whether the composition of the *O. mykiss* populations differs among creeks.

It is important to clarify that the genetic signal that we see with respect to the unknowns could be a natural phenomenon among indigenous populations, a result of hybridization between redband and coastal lineages, or a result of those two forces in combination.

The data suggest that part of the genetic signal is probably due to natural variation within the regional redband lineage, but we cannot eliminate the possibility that some of it is due to recent hybridization between the two lineages.

If we ascribe low cluster membership likelihoods to recent hybridization, we can use those values to classify individuals as purebred, or of mixed lineage. This would be a conservative approach if the intent is to rule-out prospective broodstock of mixed ancestry, but it might result in exclusion of naturally occuring variation from the broodstock and accelerate divergence of the broodstock from its indigenous source.

Ninety-five percent of the 3770 maximum inferred ancestry estimates from the *k*=3 model for putative purebred individuals exceeded 0.935 in the Spokane Hatchery, Goldendale Hatchery, and Deadman Creek collections, so we established that as a threshold for assigning unknown-ancestry individuals as purebred. F_1 hybrids have equal genic representation from both parental lineages, but genotypic similarities between lineages and imperfect analysis algorithms can make it difficult to distinguish F_1 hybrids from backcrosses in samples of unknowns. We caution, however, that we have no objective assessment of how many BC_3, or even F_1, BC_1 or BC_2, individuals have inferred ancestries to either parental line that exceed 0.935.

We recognize five kinds of individuals in this classification: 1) purebreds, with estimated ancestry proportions exceeding 0.935; 2) likely F_1 hybrids, with joint ancestries at near 0.5 in both parental lineages; 3) probable backcrosses, with estimated ancestry from the recurrent lineage between 0.7 and 0.935; 4) undetermined origin; and 5) other species (Table10). The 07LC collection included three likely F_1 redband x McCloud hybrids, 20 probable backcrosses, 5 undetermined origin for lack of data, 72 purebred redband trout, and one cutthroat trout.

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Figure 1. Lineage swamping in backcross generations. The recurrent ancestral lineage increases in backcross generations so that the proportion of alleles from the recurrent lineage averages $1\text{-}0.5^f$, where f is the number of generations post-hybridization. We simulated 10,000 individuals in generations BC_1 – BC_3 (f=2-4) to estimate the proportion of alleles from the recurrent lineage that we would expect in samples of from 1 to 30 loci. The high and low lines encompass 95% of the simulated genotypes. The ranges of proportions in all three backcross generations overlap for samples of up to 20 loci.

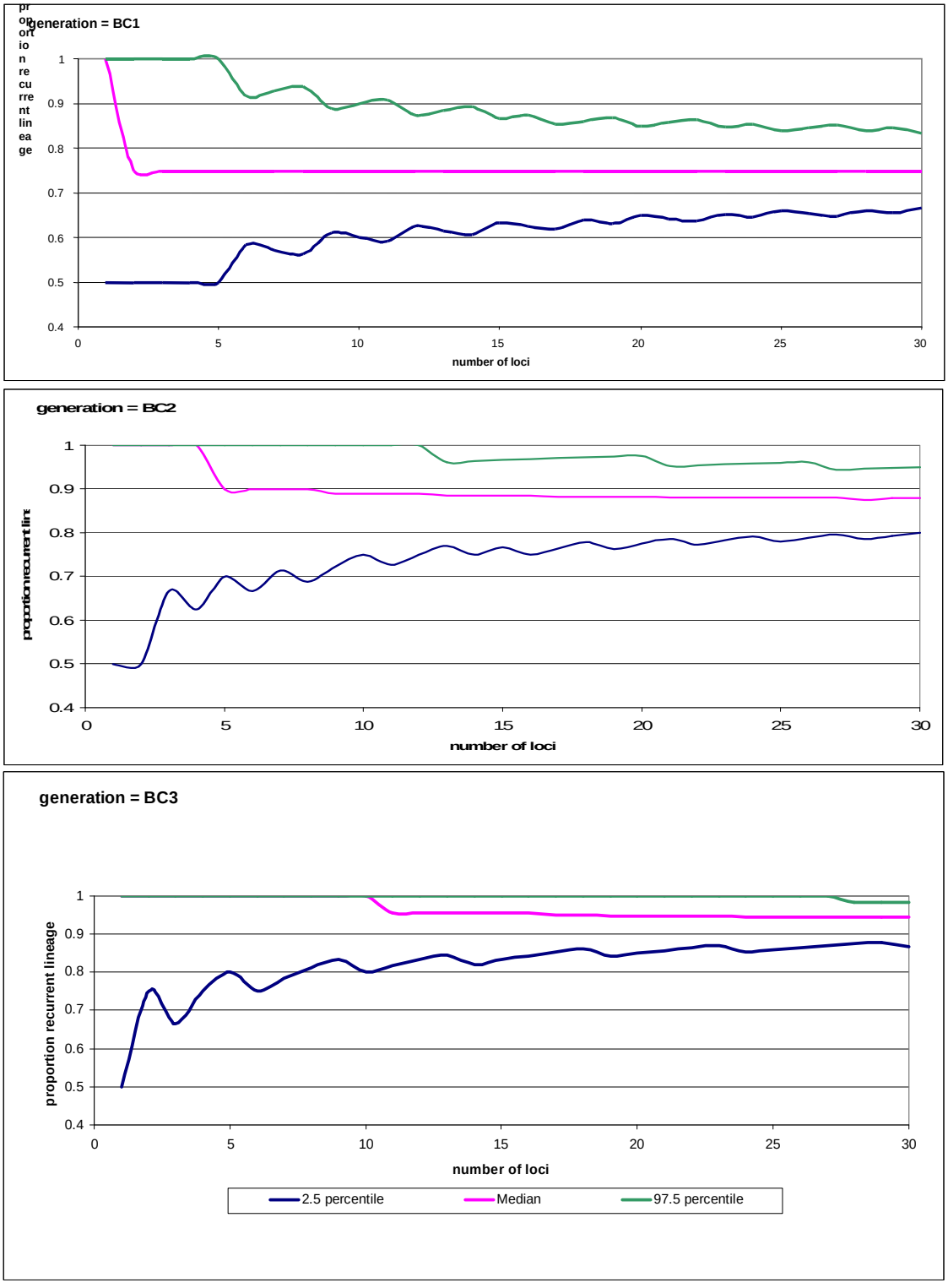
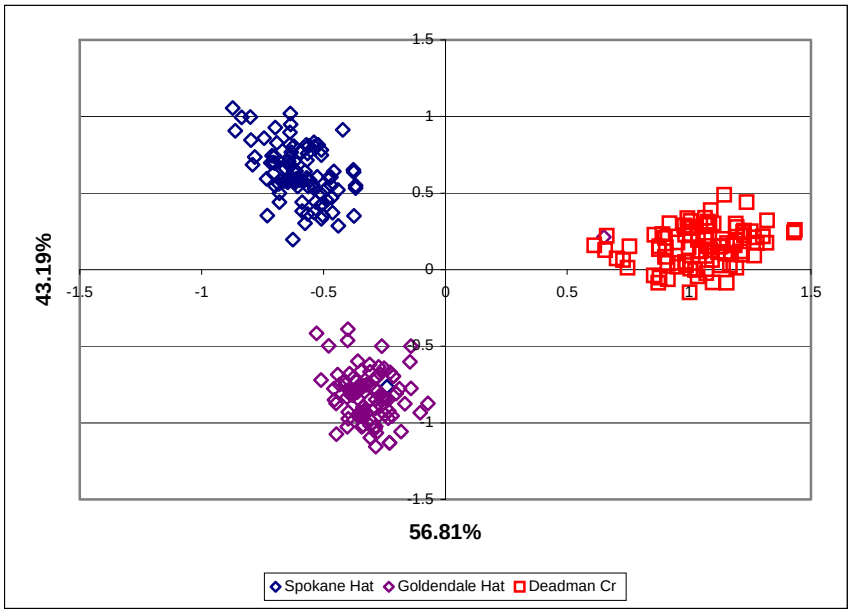
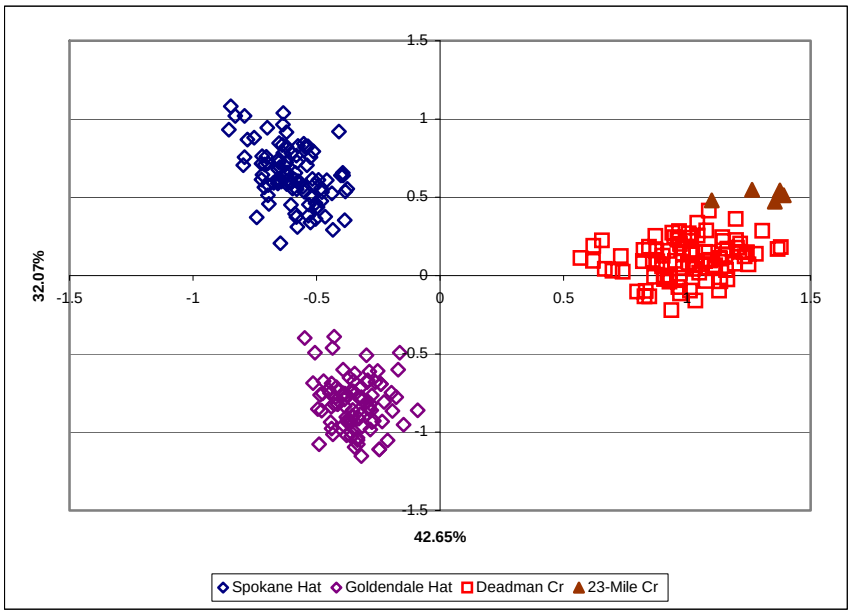


Figure 2. Correspondence between allele frequencies and collection areas by factorial correspondence analysis.

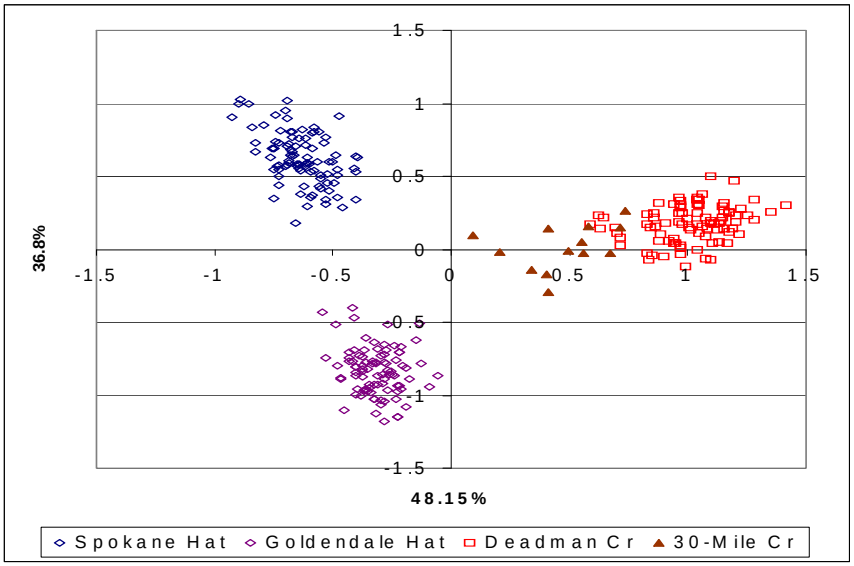
a) The baseline collections: Spokane Hatchery, Goldendale Hatchery, and Deadman Creek resident rainbow trout.



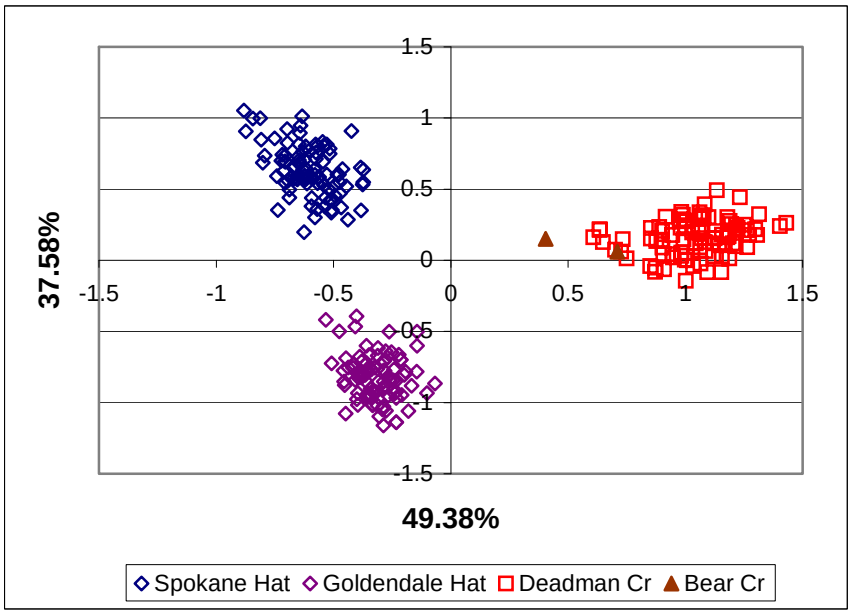
b) The baseline collections and 23-Mile Creek (n=5).



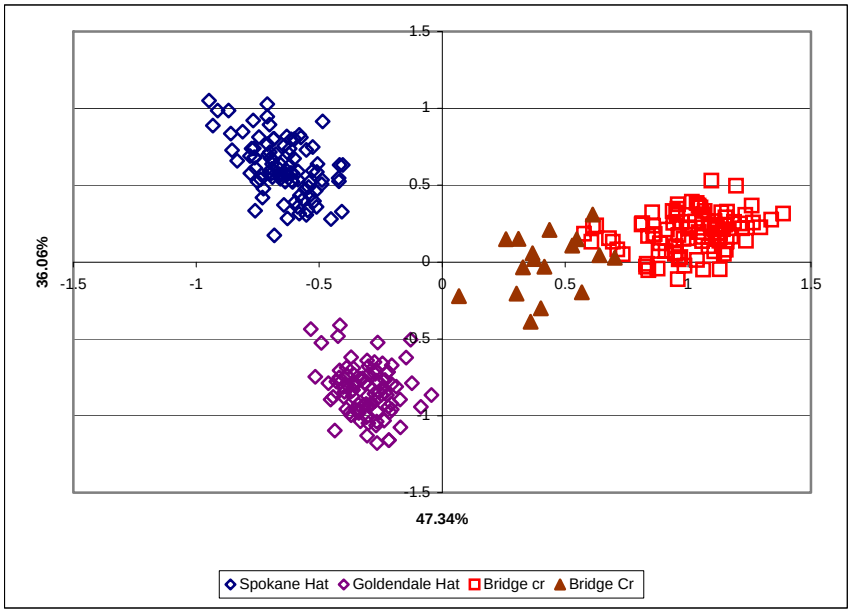
c) The baseline collections and 30-Mile Creek (n=13).



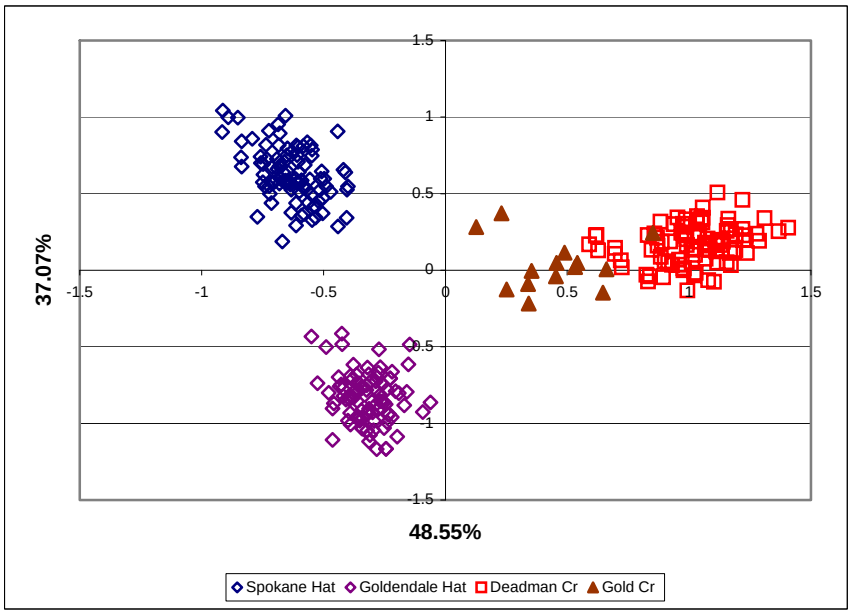
d) The baseline collections and Bear Creek (n=2).



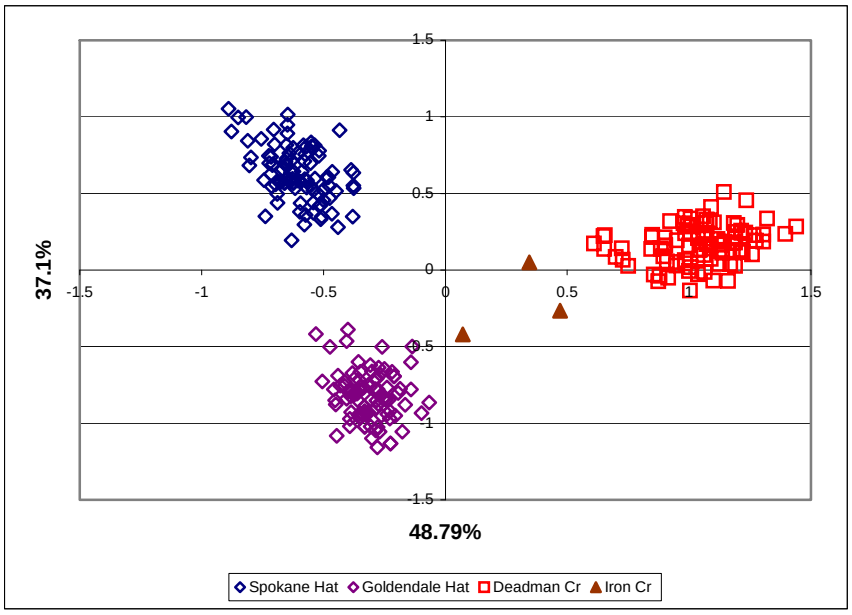
e) The baseline collections and Bridge Creek (n=17).



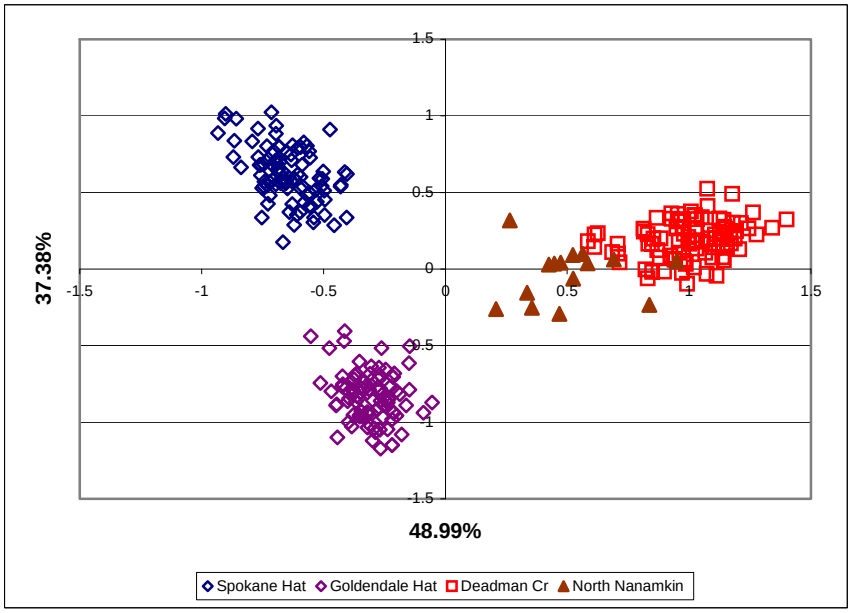
f) The baseline collections and Gold Creek (n=14)..



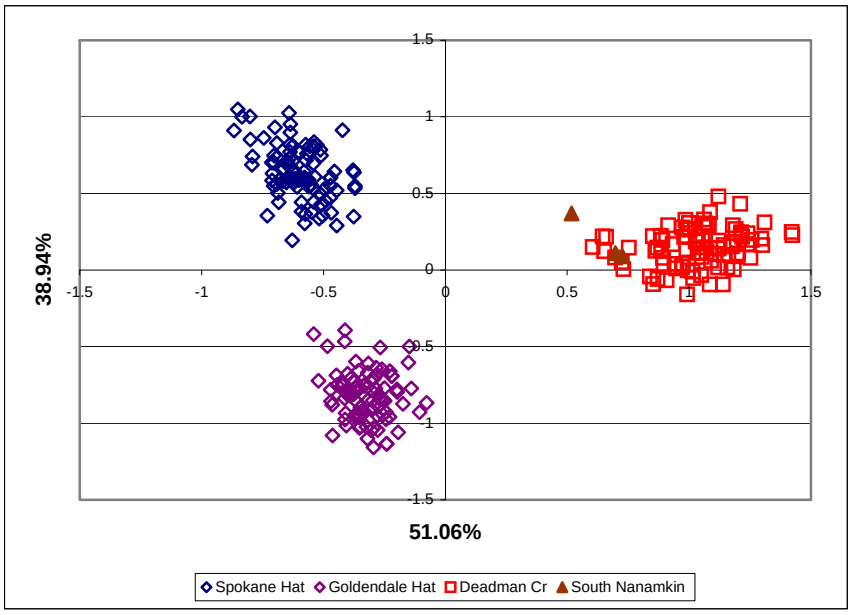
g) The baseline collections and Iron Creek (n=3)..



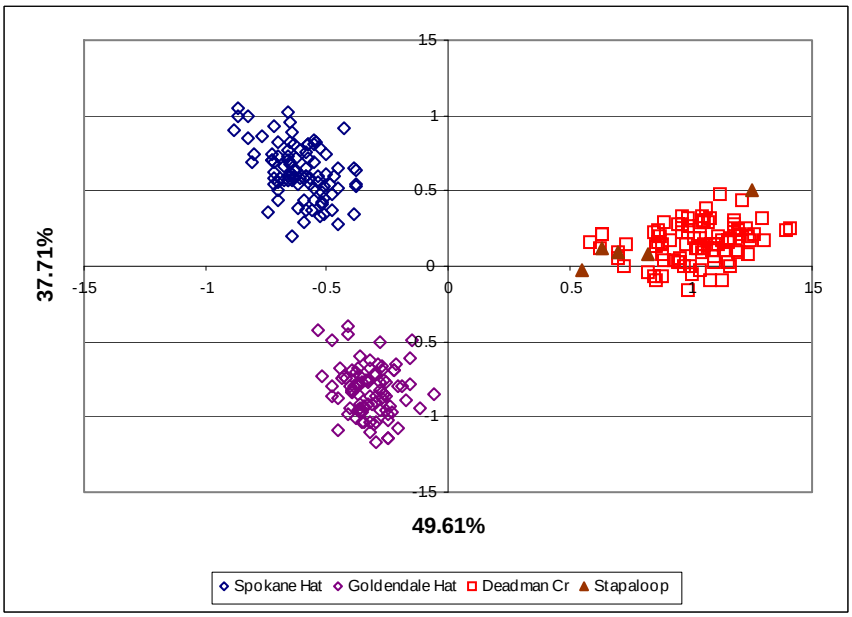
h) The baseline collections and North Nanamkin Creek (n=15)..



i) The baseline collections and South Nanamkin Creek (n=3)..



j) The baseline collections and Stapaloop Creek (n=5)..



k) The baseline collections and West Fork Creek (n=17)..

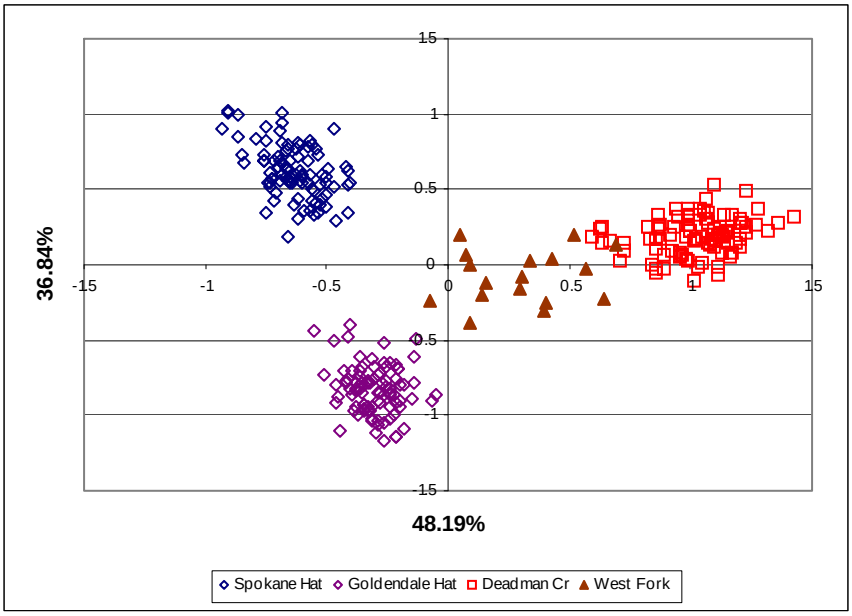


Table 1. Collection locations, years, and sample sizes for the unknown-lineage trout collected on the Colville Reservation (MGL collection code 07LC).

Population	2005	2007	Total
23-Mile Creek		5	5
30 Mile Creek	5	11	16
Bear Creek	2		2
Bridge Creek	2	15	17
Gold Creek	3	12	15
Iron Creek	3		3
North Nanamkin Creek	5	11	16
South Nanamkin Creek		3	3
Stapaloop Creek	5		5
West Fork Creek	3	15	18
Total			100

Table 2. Microsatellite loci screened to assess the likely lineage of unknown trout collected by the Colville Tribe.

Locus	Annealing temperature (°C)	# of alleles	Source
Omm-1070	62	35	Rexroad *et al.* 2001
Omm-1130	62	40	Rexroad *et al.* 2001
Omy-1001	52	25	Spies *et al.* 2005
Omy-1011	62	19	Spies *et al.* 2005
Omy-77	49	20	Morris *et al.* 1996
One-101	55	18	Olsen *et al.* 2000
One-102	55	26	Olsen *et al.* 2000
One-108	55	24	Olsen *et al.* 2000
One-114	55	25	Olsen *et al.* 2000
Ots-1	49	18	Banks *et al.* 1999
Ots-100	55	24	Nelson and Beacham 1999
Ots-103	55	6	Small *et al.* 1998
Ots-3M	49	8	Banks *et al.* 1999

Table 3. Rainbow trout populations screened with microsatellites to assess the lineage of unknown trout from the Colville Reservation. The unknowns also were screened with mitochondrial DNA species markers (see below) to eliminate non-rainbow trout from the microsatellite analyses.

Baseline population	Represents	Collection code	n
Spokane Hatchery	McCloud strain rainbow trout	00DF	98
Goldendale Hatchery	McCloud strain rainbow trout	01JB	94
Deadman Creek - Phalon Lk H broodstock	redband trout	02MB	89

Table 4. Maternal lineages determined with mitochondrial single nucleotide polymorphism assays that target species-informative nucleotide substitutions in the coxIII-ND3 region of the mitochondrial genome. The Ocl peaks amplify in westslope, Yellowstone, Lahontan and coastal cutthroat trout but are absent from all rainbow trout. The Omy peaks appear in McCloud lineage rainbow trout as well as Columbia basin redband trout, but are absent from all cutthroat trout.

			Peaks present										
Sample Identity	**Sample origin**	**Species call**	**Ocl**	**Oke**	**Ots**	**Sal**	**Sfo**	**Sco**	**One**	**Ogo**	**Omy**	**Ssa**	**Oki**
Controls													
01BN0005	Phalon Lake Hatchery *O. mykiss*	Omy		X				X			**X**		
01BN0013	Phalon Lake Hatchery *O. mykiss*	Omy		X				X			**X**		X
01BN0020	Phalon Lake Hatchery *O. mykiss*	Omy		X				X			**X**		
01BN0025	Phalon Lake Hatchery *O. mykiss*	Omy		X				X			**X**		
01BN0039	Phalon Lake Hatchery *O. mykiss*	Omy		X				X			**X**		
01BN0043	Phalon Lake Hatchery *O. mykiss*	Omy		X				X			**X**		X
01BN0051	Phalon Lake Hatchery *O. mykiss*	Omy		X				X			**X**		X
01BN0053	Phalon Lake Hatchery *O. mykiss*	Omy		X				X			**X**		
01BN0054	Phalon Lake Hatchery *O. mykiss*	Omy		X				X			**X**		X
01BN0061	Phalon Lake Hatchery *O. mykiss*	Omy		X				X			**X**		X
01BN0065	Phalon Lake Hatchery *O. mykiss*	Omy		X				X			**X**		X
01BN0067	Phalon Lake Hatchery *O. mykiss*	Omy		X				X			**X**		X
01BN0073	Phalon Lake Hatchery *O. mykiss*	Omy		X				X			**X**		X
01BN0077	Phalon Lake Hatchery *O. mykiss*	Omy		X			X	X			**X**		X
01BN0086	Phalon Lake Hatchery *O. mykiss*	Omy		X				X			**X**		
01BN0099	Phalon Lake Hatchery *O. mykiss*	Omy		X				X			**X**		
01JB0071	Goldendale Hatchery *O. mykiss*	Omy		X				X			**X**		X
01JB0072	Goldendale Hatchery *O. mykiss*	Omy		X				X			**X**		
01JB0073	Goldendale Hatchery *O. mykiss*	Omy		X				X			**X**		X
01JB0074	Goldendale Hatchery *O. mykiss*	Omy		X				X			**X**		
01JB0075	Goldendale Hatchery *O. mykiss*	Omy		X				X			**X**		
01JB0076	Goldendale Hatchery *O. mykiss*	Omy		X				X			**X**		
01JB0077	Goldendale Hatchery *O. mykiss*	Omy		X				X			**X**		
01JB0078	Goldendale Hatchery *O. mykiss*	Omy		X				X			**X**		X

Sample Identity	Sample origin	Species call	Peaks present											
			Ocl	Oke	Ots	Sal	Sfo	Sco	One	Ogo	Omy	Ssa	Oki	
01JB0079	Goldendale Hatchery *O. mykiss*	Omy		X				X			**X**		X	X
01JB0080	Goldendale Hatchery *O. mykiss*	Omy		X				X			**X**			
01JB0081	Goldendale Hatchery *O. mykiss*	Omy		X				X			**X**			X
01JB0082	Goldendale Hatchery *O. mykiss*	Omy		X				X			**X**			X
01JB0083	Goldendale Hatchery *O. mykiss*	Omy		X				X			**X**			X
01JB0084	Goldendale Hatchery *O. mykiss*	Omy		X				X			**X**			X
01JB0085	Goldendale Hatchery *O. mykiss*	Omy		X				X			**X**			
01JB0086	Goldendale Hatchery *O. mykiss*	Omy		X				X			**X**			X
03CC0034	Sullivan Lake *O. clarki*	Ocl	**X**	X				X						X
03CC0036	Sullivan Lake *O. clarki*	Ocl	**X**	X				X						X
03CC0038	Sullivan Lake *O. clarki*	Ocl	**X**											
03CC0040	Sullivan Lake *O. clarki*	Ocl	**X**											
03CC0042	Sullivan Lake *O. clarki*	Ocl	**X**	X				X						X
03CC0044	Sullivan Lake *O. clarki*	Ocl	**X**	X				X						X
03CC0046	Sullivan Lake *O. clarki*	Ocl	**X**	X				X						X
03CC0048	Sullivan Lake *O. clarki*	Ocl	**X**	X				X						X
03CN0013	Gold Creek *O. clarki*	Ocl	**X**	X				X						X
03CN0015	Gold Creek *O. clarki*	Ocl	**X**					X						X
03CN0017	Gold Creek *O. clarki*	Ocl	**X**	X				X						X
03CN0019	Gold Creek *O. clarki*	Ocl	**X**	X				X						X
03CN0021	Gold Creek *O. clarki*	Ocl	**X**	X				X						X
03CN0023	Gold Creek *O. clarki*	Ocl	**X**	X				X						X
03CN0025	Gold Creek *O. clarki*	Ocl	**X**	X				X						X
03CN0027	Gold Creek *O. clarki*	Ocl	**X**	X				X						X
04BL0001	Lake Lenore Lahontan *O. clarki*	Ocl	**X**	X				X						X
04BL0002	Lake Lenore Lahontan *O. clarki*	Ocl	**X**	X				X						X
04BL0003	Lake Lenore Lahontan *O. clarki*	Ocl	**X**	X				X						X
04BL0004	Lake Lenore Lahontan *O. clarki*	Ocl	**X**	X				X						X
04BL0005	Lake Lenore Lahontan *O. clarki*	Ocl	**X**	X				X						X
04BL0006	Lake Lenore Lahontan *O. clarki*	Ocl	**X**	X				X						X

Sample Identity	Sample origin	Species call	Peaks present										
			Ocl	Oke	Ots	Sal	Sfo	Sco	One	Ogo	Omy	Ssa	Oki
04BL0007	Lake Lenore Lahontan *O. clarki*	Ocl	X	X				X					
04BL0008	Lake Lenore Lahontan *O. clarki*												
99NO0017	Yellowstone River Hatchery *O. clarki*	Ocl	X	X				X					X
99NO0019	Yellowstone River Hatchery *O. clarki*	Ocl	X	X				X					X
99NO0021	Yellowstone River Hatchery *O. clarki*	Ocl	X	X				X					X
99NO0023	Yellowstone River Hatchery *O. clarki*	Ocl	X	X				X					X
99NO0025	Yellowstone River Hatchery *O. clarki*	Ocl	X	X				X					X
99NO0027	Yellowstone River Hatchery *O. clarki*	Ocl	X	X				X					X
99NO0029	Yellowstone River Hatchery *O. clarki*	Ocl	X	X				X					
99NO0031	Yellowstone River Hatchery *O. clarki*	Ocl	X	X				X					X
05BB0015	Cedar River *O. clarki*	Ocl	X					X					X
05BB0016	Cedar River *O. clarki*	Ocl	X				X	X					X
05BB0017	Cedar River *O. clarki*												
05BB0018	Cedar River *O. clarki*	Ocl	X	X				X					X
05BB0019	Cedar River *O. clarki*	Ocl	X				X	X					X
05BB0022	Cedar River *O. clarki*												
05BB0030	Cedar River *O. clarki*	Ocl	X	X			X	X					X
05BB0031	Cedar River *O. clarki*	Ocl	X					X					X
	Unknowns												
07LC0001	Gold Ck.	Omy		X				X			X		X
07LC0002	30 Mile	Omy		X				X			X		X
07LC0003	30 Mile	Omy		X				X			X		X
07LC0004	Bridge Ck.	Omy		X				X			X		X
07LC0005	North Nanamkin	Omy		X				X			X		X
07LC0006	Bridge Ck.	Omy		X				X			X		X
07LC0007	30 Mile	Omy		X				X			X		X
07LC0008	Iron Ck.	Omy		X				X			X		X
07LC0009	30 Mile	Omy		X				X			X		X
07LC0010	North Nanamkin	Omy		X				X			X		X
07LC0011	Bear Ck.	Omy		X				X			X		X

Sample Identity	Sample origin	Species call	Peaks present											
			Ocl	Oke	Ots	Sal	Sfo	Sco	One	Ogo	Omy	Ssa	Oki	
07LC0012	West Fork	Omy		X				X			X			
07LC0013	West Fork	Omy		X				X			X			
07LC0014	West Fork	Omy		X				X			X		X	X
07LC0015	North Nanamkin	Omy		X				X			X		X	X
07LC0016	North Nanamkin	Omy		X				X			X			
07LC0017	North Nanamkin	Omy		X				X			X		X	X
07LC0018	30 Mile	Omy		X				X			X		X	X
07LC0019	30 Mile	Omy		X				X			X			X
07LC0020	Iron Ck.	Omy		X				X			X		X	X
07LC0021	Iron Ck.	Omy		X				X			X		X	X
07LC0022	Gold Ck.	Omy		X				X			X		X	X
07LC0023	Gold Ck.	Omy		X				X			X		X	X
07LC0024	Bear Ck.	Omy		X				X			X		X	X
07LC0025	Bridge Ck.	Omy		X				X			X		X	X
07LC0026	Bridge Ck.	Omy		X				X			X		X	X
07LC0027	West Fork	Omy		X				X			X			X
07LC0028	West Fork	Omy		X				X			X		X	X
07LC0029	North Nanamkin	Omy		X				X			X			
07LC0030	West Fork	Omy		X			X	X			X		X	X
07LC0031	30 Mile	Omy		X				X			X			X
07LC0032	30 Mile	Omy		X				X			X			X
07LC0033	Gold Ck.	Omy		X				X			X			X
07LC0034	30 Mile	Omy		X				X			X			X
07LC0035	30 Mile	Omy		X				X			X			X
07LC0036	Gold Ck.	Omy		X				X			X			X
07LC0037	North Nanamkin	Omy		X				X			X		X	X
07LC0038	Bridge Ck.	Omy		X				X			X		X	
07LC0039	West Fork	Omy		X				X			X			
07LC0040	North Nanamkin	Omy		X				X			X			
07LC0041	Bridge Ck.	Omy		X				X			X			X

Sample Identity	Sample origin	Species call	Peaks present											
			Ocl	Oke	Ots	Sal	Sfo	Sco	One	Ogo	Omy	Ssa	Oki	
07LC0042	West Fork	Omy		X				X			X			
07LC0043	North Nanamkin	Omy		X				X			X		X	X
07LC0044	West Fork	Omy		X				X			X			X
07LC0045	Bridge Ck.	Omy		X				X			X			X
07LC0046	North Nanamkin	Omy		X				X			X		X	X
07LC0047	West Fork	Omy		X				X			X		X	X
07LC0048	Bridge Ck.	Omy		X				X			X			X
07LC0049	North Nanamkin	Omy		X				X			X			X
07LC0050	Gold Ck.	Ocl	X	X				X						X
07LC0051	30 Mile	Omy		X				X			X			X
07LC0052	South Nanamkin	Omy		X				X			X		X	X
07LC0053	North Nanamkin	Omy		X				X			X			
07LC0054	Bridge Ck.	Omy		X				X			X			X
07LC0055	Bridge Ck.	Omy		X				X			X			X
07LC0056	West Fork	Omy		X				X			X			
07LC0057	West Fork	Omy		X				X			X			X
07LC0058	Bridge Ck.	Omy		X				X			X			
07LC0059	North Nanamkin	Omy		X				X			X			X
07LC0060	30 Mile	Omy		X				X			X		X	X
07LC0061	North Nanamkin	Omy		X				X			X			X
07LC0062	Bridge Ck.	Omy		X				X			X			
07LC0063	Bridge Ck.	Omy		X				X			X		X	X
07LC0064	West Fork	Omy		X				X			X			X
07LC0065	30 Mile	Omy		X				X			X			X
07LC0066	Bridge Ck.	Omy		X				X			X			X
07LC0067	Bridge Ck.	Omy		X				X			X		X	
07LC0068	Gold Ck.	Omy		X				X			X		X	X
07LC0069	Gold Ck.	Omy		X				X			X			X
07LC0070	Bridge Ck.	Omy		X				X			X			X
07LC0071	Gold Ck.	Omy		X				X			X			X

Sample Identity	Sample origin	Species call	Peaks present											
			Ocl	Oke	Ots	Sal	Sfo	Sco	One	Ogo	Omy	Ssa	Oki	
07LC0072	30 Mile	Omy		X			X	X			X		X	
07LC0073	South Nanamkin	Omy		X				X			X		X	X
07LC0074	West Fork	Omy		X				X			X			X
07LC0075	South Nanamkin	Omy		X				X			X			
07LC0076	Gold Ck.	Omy		X				X			X			X
07LC0077	Gold Ck.	Omy		X				X			X		X	X
07LC0078	Gold Ck.	Omy		X				X			X		X	X
07LC0079	Gold Ck.	Omy		X				X			X			X
07LC0080	Gold Ck.	Omy		X				X			X		X	X
07LC0081	Bridge Ck.	Omy		X				X			X			X
07LC0082	North Nanamkin	Omy		X				X			X			X
07LC0083	30 Mile	Omy		X				X			X		X	X
07LC0084	30 Mile	Omy		X				X			X		X	X
07LC0085	North Nanamkin	Omy									X		X	
07LC0086	West Fork	Omy		X				X			X			X
07LC0087	West Fork	Omy		X				X			X		X	X
07LC0088	West Fork	Omy		X				X			X		X	X
07LC0089	Gold Ck.	Omy		X				X			X			
07LC0090	West Fork	Omy		X				X			X		X	X
07LC0091	Stapaloop Ck.	Omy		X				X			X		X	X
07LC0092	Stapaloop Ck.	Omy		X				X			X		X	X
07LC0093	Stapaloop Ck.	Omy		X				X			X		X	X
07LC0094	Stapaloop Ck.	Omy		X				X			X			X
07LC0095	Stapaloop Ck.	Omy		X				X			X		X	X
07LC0096	23-Mile	Omy		X				X			X		X	X
07LC0097	23-Mile	Omy		X				X			X		X	X
07LC0098	23-Mile	Omy		X				X			X		X	X
07LC0099	23-Mile	Omy		X				X			X		X	X
07LC0100	23-Mile	Omy		X				X			X		X	X

Table 5. Allele frequencies at 13 microsatellite loci in the baseline populations. Bold, underlined values highlight alleles that are unique among these baseline collections.

	Spokane Hatchery	Goldendale Hatchery	Deadman Creek
Locus: Omm-1070			
N (alleles):	188	180	146
164	-	-	**0.089**
172	-	-	**0.247**
180	-	-	**0.021**
184	-	0.094	0.007
188	0.197	0.089	0.048
192	0.059	0.078	0.007
196	-	-	**0.007**
200	-	-	**0.041**
204	-	-	-
208	-	-	**0.103**
212	0.074	-	0.007
216	0.101	0.328	0.041
219	-	-	**0.014**
223	0.043	0.033	-
227	0.149	0.144	0.034
231	0.08	-	0.048
235	-	0.022	0.055
239	-	-	**0.007**
243	-	-	**0.075**
247	0.048	-	0.021
251	-	-	**0.007**
255	-	-	-
259	-	-	-
263	**0.005**	-	-
267	**0.245**	-	-
271	-	-	**0.034**
291	-	**0.206**	-
297	-	**0.006**	-
301	-	-	**0.007**
305	-	-	**0.027**
309	-	-	**0.034**
313	-	-	-
318	-	-	**0.021**
322	-	-	-
326	-	-	-
Locus: Omm-1130			
N (alleles):	188	182	144
200	-	-	-

	Spokane Hatchery	Goldendale Hatchery	Deadman Creek
204	-	0.187	0.049
208	-	-	**0.097**
210	-	-	-
212	**0.016**	-	-
216	-	0.165	0.264
220	-	-	-
224	-	0.016	0.014
228	0.027	-	0.069
232	0.043	0.088	0.007
236	-	-	**0.007**
238	-	-	**0.007**
240	-	0.093	0.035
244	-	0.093	0.049
248	-	0.011	0.035
252	0.25	0.093	-
256	-	-	**0.007**
260	**0.165**	-	-
264	0.074	-	0.104
268	0.043	0.016	0.083
272	**0.287**	-	-
276	-	-	**0.014**
284	-	-	**0.007**
288	-	**0.044**	-
292	0.016	0.011	-
296	0.08	0.181	0.007
300	-	-	-
312	-	-	-
324	-	-	-
328	-	-	**0.014**
333	-	-	**0.014**
341	-	-	-
353	-	-	**0.049**
368	-	-	-
369	-	-	**0.007**
372	-	-	-
376	-	-	**0.007**
379	-	-	**0.021**
387	-	-	**0.035**
391	-	-	-
Locus: Omy-1001			
N (alleles):	192	170	158
171	-	-	**0.025**
173	**0.042**	-	-
175	-	-	-
179	0.099	0.012	0.063

	Spokane Hatchery	Goldendale Hatchery	Deadman Creek
181	-	-	**0.006**
183	-	-	**0.196**
185	-	-	-
187	0.354	0.353	0.057
189	-	-	**0.006**
191	-	-	**0.063**
193	-	0.129	0.196
195	-	-	**0.108**
198	0.047	0.247	0.07
200	-	0.059	0.006
202	0.24	0.012	0.019
204	-	0.135	0.044
206	-	-	**0.019**
208	-	-	**0.013**
212	-	-	-
214	-	0.053	0.013
216	0.042	-	0.076
220	**0.177**	-	-
222	-	-	-
224	-	-	**0.006**
228	-	-	**0.013**
Locus: Omy-1011			
N (alleles):	184	180	86
134	0.011	0.017	-
138	-	-	**0.035**
147	-	-	**0.023**
151	-	-	**0.012**
155	-	-	**0.035**
159	0.011	0.178	0.07
163	0.299	0.072	0.093
167	0.147	-	0.14
171	-	0.05	0.023
175	-	-	**0.058**
179	0.005	0.6	0.023
183	0.016	0.017	0.105
187	0.016	-	0.012
191	0.495	0.011	0.07
195	-	-	**0.116**
199	-	0.056	0.128
203	-	-	**0.023**
206	-	-	**0.035**
245	-	-	-
Locus: Omy-77			
N (alleles):	184	176	174

	Spokane Hatchery	Goldendale Hatchery	Deadman Creek
97	-	-	**0.052**
99	0.158	0.347	0.057
101	**0.022**	-	-
103	0.228	0.034	0.011
105	0.13	0.108	-
108	-	-	-
110	0.022	-	0.034
112	-	-	-
114	0.201	0.193	-
116	-	-	**0.132**
118	-	-	**0.069**
120	-	0.261	0.006
122	-	-	**0.052**
124	0.212	-	0.034
126	0.011	0.017	0.213
128	0.016	0.04	0.04
130	-	-	**0.121**
132	-	-	**0.149**
136	-	-	**0.029**
140	-	-	-
Locus: One-101			
N (alleles):	184	174	174
119	0.293	0.063	0.851
123	-	**0.017**	-
127	0.185	0.276	0.098
140	-	**0.006**	-
157	-	**0.356**	-
166	0.13	0.201	0.017
170	-	-	-
174	-	-	-
178	0.087	0.006	0.029
182	-	-	-
186	0.06	0.075	-
190	-	-	-
203	**0.103**	-	-
214	**0.141**	-	-
226	-	-	**0.006**
239	-	-	-
758	-	-	-
762	-	-	-
Locus: One-102			
N (alleles):	190	182	172
182	-	-	-
188	0.326	0.044	0.093

	Spokane Hatchery	Goldendale Hatchery	Deadman Creek
192	0.274	0.654	0.157
196	0.379	0.176	0.07
200	-	-	**0.11**
204	-	-	**0.18**
208	0.021	0.016	0.035
212	-	-	**0.041**
216	-	-	**0.058**
221	-	0.11	0.07
225	-	-	**0.122**
229	-	-	-
233	-	-	-
237	-	-	**0.006**
241	-	-	**0.029**
245	-	-	**0.006**
253	-	-	-
257	-	-	**0.006**
261	-	-	**0.006**
265	-	-	**0.006**
269	-	-	**0.006**
273	-	-	-
277	-	-	-
285	-	-	-
290	-	-	-
294	-	-	-

Locus: One-108			
N (alleles):	190	170	174
164	**0.011**	-	-
169	**0.232**	-	-
173	-	-	**0.006**
181	0.021	-	0.04
185	-	-	**0.046**
189	0.316	0.035	0.006
193	0.005	0.129	0.236
197	-	0.271	0.126
201	0.016	0.082	0.08
205	-	-	**0.075**
209	-	0.082	0.04
213	-	0.012	0.075
217	-	-	**0.011**
221	0.068	0.147	0.08
225	0.011	0.059	0.017
229	-	-	-
233	-	-	**0.086**
241	0.274	0.071	0.029
249	0.042	-	0.046

	Spokane Hatchery	Goldendale Hatchery	Deadman Creek
253	**0.005**	-	-
273	-	-	-
312	-	-	-
317	-	**0.112**	-
337	-	-	-
Locus: One-114			
N (alleles):	192	176	172
181	-	-	**0.07**
185	-	-	-
189	-	-	**0.052**
193	-	-	**0.029**
197	0.057	-	0.029
201	0.109	0.017	0.076
205	-	-	**0.047**
209	-	-	**0.064**
213	-	-	**0.07**
217	0.099	0.256	0.169
221	0.042	0.199	0.047
225	0.26	-	0.076
229	0.01	-	0.047
233	0.052	0.188	0.047
236	0.026	0.045	0.047
240	-	0.097	0.012
244	0.005	-	0.017
248	0.307	0.142	0.006
252	**0.031**	-	-
256	-	0.006	0.029
260	-	0.051	0.047
268	-	-	**0.012**
289	-	-	-
364	-	-	**0.006**
368	-	-	**0.006**
Locus: Ots-1			
N (alleles):	192	166	172
158	0.016	0.133	0.017
160	**0.177**	-	-
164	0.016	0.319	0.035
166	0.24	0.114	0.122
168	0.005	0.018	0.157
170	0.521	0.265	0.151
172	**0.005**	-	-
177	-	-	**0.017**
179	-	**0.06**	-
181	-	-	**0.012**

	Spokane Hatchery	Goldendale Hatchery	Deadman Creek
183	-	-	**0.006**
237	-	-	**0.128**
241	-	-	**0.047**
245	-	0.09	0.302
247	0.021	-	0.006
249	-	-	-
256	-	-	-
689	-	-	-
Locus: Ots-100			
N (alleles):	192	182	174
168	0.057	0.011	0.115
173	-	-	**0.195**
177	0.078	-	0.075
181	-	-	**0.241**
185	-	-	**0.04**
187	0.031	0.242	0.017
189	-	-	**0.046**
191	**0.005**	-	-
193	-	**0.269**	-
195	0.172	0.011	0.034
197	0.01	0.121	0.023
199	0.141	0.077	0.017
201	-	-	**0.109**
203	-	-	-
205	-	-	**0.034**
207	0.089	0.011	-
211	0.182	0.192	0.017
213	-	-	**0.006**
215	0.109	0.027	0.006
220	**0.005**	-	-
222	-	-	-
223	-	-	**0.017**
224	0.12	0.038	0.006
726	-	-	-
Locus: Ots-103			
N (alleles):	192	180	174
56	0.427	0.283	0.023
74	-	-	-
82	0.573	0.711	0.862
86	-	0.006	0.115
90	-	-	-
594	-	-	-
Locus: Ots-3M			

		Spokane Hatchery	Goldendale Hatchery	Deadman Creek
N (alleles):		190	178	174
	130	-	-	-
	132	0.026	0.045	-
	134	0.047	0.067	0.034
	136	0.826	0.388	0.402
	138	0.1	0.5	0.253
	140	-	-	**0.023**
	143	-	-	**0.253**
	145	-	-	**0.034**

Table 6. Allelic richness in the baseline collections and the unknown-lineage samples. Allelic richness is a measure of genetic diversity that is based on the numbers of alleles that are observed in each population after adjusting for unequal sample sizes. The values in the table are based on sample sizes of 48 individuals. From Fstat v. 2.9.3 2.

Locus	Spokane Hatchery	Goldendale Hatchery	Deadman Creek	07LC - Unknowns
Omm-1070	9.45	8.39	20.68	24.98
Omm-1130	9.63	11.15	20.04	26.97
Omy-1001	6.98	7.51	16.33	19.65
Omy-1011	6.61	7.44	17.00	14.32
Omy-77	8.41	6.84	13.17	16.17
One-101	7.00	6.86	4.34	9.70
One-102	3.91	4.85	13.95	17.93
One-108	9.05	9.74	14.55	20.54
One-114	10.06	8.35	19.77	19.59
Ots-1	6.47	6.89	10.49	13.09
Ots-100	10.57	9.12	14.87	16.91
Ots-103	2.00	2.48	2.94	4.62
Ots-3M	3.95	4.00	5.91	7.11

Table 7. Pooled frequencies of unique alleles in the baseline collections. Bold entries highlight pooled frequencies at a locus that equal or exceed 0.5

	Spokane Hat	Goldendale Hat	McCloud combined	Deadman Cr
Omm-1070	0.250	0.212	0.270	**0.734**
Omm-1130	0.468	0.044	0.444	0.286
Omy-1001	0.219	-	0.111	0.455
Omy-1011	-	-	0.014	0.337
Omy-77	0.022	-	0.328	**0.604**
One-101	0.244	0.379	0.378	0.006
One-102	-	-	-	**0.576**
One-108	0.248	0.112	0.181	0.224
One-114	0.031	-	0.016	0.356
Ots-1	0.182	0.060	0.125	0.210
Ots-100	0.010	0.269	0.187	**0.688**
Ots-103	-	-	-	-
Ots-3M	-	-	0.035	0.310
Mean	0.186	0.179	0.190	0.399

Table 8. The genotypic proportions in four of the five baseline collections were out of Hardy-Weinberg Equilibrium. Underlined F_{IS} values identify loci x population combinations with substantial heterozygote deficiencies (F_{IS}.> 0.1). From Fstat v. 2.9.3 2.

Locus	Spokane Hatchery	Goldendale Hatchery	***Deadman Creek***
Omm-1070	0.004	0.081	<u>0.152</u>
Omm-1130	-0.070	-0.032	0.004
Omy-1001	-0.131	<u>0.138</u>	0.049
Omy-1011	-0.041	-0.018	0.017
Omy-77	0.011	-0.072	0.054
One-101	-0.001	0.036	-0.075
One-102	-0.040	0.026	0.052
One-108	-0.014	0.068	0.061
One-114	0.024	0.014	0.028
Ots-1	-0.071	-0.022	<u>0.273</u>
Ots-100	-0.021	-0.027	0.022
Ots-103	<u>0.111</u>	-0.149	-0.129
Ots-3M	0.037	<u>0.115</u>	-0.002

Table 9. Estimated null allele frequencies based on homozygote excesses. Underlined values highlight estimated null allele frequencies that are greater than 0.05. From Genepop v. 4.0.7.

Locus	Spokane Hatchery	Goldendale Hatchery	Deadman Creek
Omm-1070	0.005	0.028	0.071
Omm-1130	-	-	-
Omy-1001	-	0.049	0.023
Omy-1011	-	-	-
Omy-77	0.026	-	0.022
One-101	-	0.008	-
One-102	-	-	0.023
One-108	-	0.039	0.018
One-114	-	-	0.019
Ots-1	-	-	0.125
Ots-100	-	-	0.007
Ots-103	0.035	-	-
Ots-3M	-	0.034	0.046

Table 10. Inferred ancestry of the 07LC collection of unknown-lineage trout from the Colville Reservation based on the *k*=3 model in Structure v 2.2. Ninety-five percent of inferred ancestry proportions among the 'purebred' Spokane Hatchery, Goldendale Hatchery, and Deadman Creek collections exceeded 0.935, so we designated that value as a conservative purebred threshold. We have no comparable objective assessment of threshold values to distinguish F_1 hybrids from backcrosses, but based on the dilution of rare lineages in backcrossed generations we infer that individuals with dominant estimated ancestries between 0.5 and 0.7 are likely F_1 hybrids (bold red), and individuals with estimated ancestries between 0.7 and 0.935 are possible backcrosses (bold blue).

Individual	Source	Gender	Sample date	Inferred ancestry	Deadman	Spo
07LC0096	23-Mile Creek	?	3-Oct-07	Redband	0.993	0.
07LC0097	23-Mile Creek	?	3-Oct-07	Redband	0.985	0.
07LC0098	23-Mile Creek	?	3-Oct-07	Redband	0.979	0.
07LC0099	23-Mile Creek	?	3-Oct-07	Redband	0.975	0.
07LC0100	23-Mile Creek	?	3-Oct-07	Redband	0.993	0.
07LC0002	30-Mile Creek	F	29-Mar-05	-	-	
07LC0003	30-Mile Creek	F	29-Mar-05	-	-	
07LC0007	30-Mile Creek	F	23-Apr-05	possible backcross	**0.924**	0.
07LC0009	30-Mile Creek	M	1-Apr-05	-	-	
07LC0018	30-Mile Creek	F	30-Mar-05	Redband	0.990	0.
07LC0019	30-Mile Creek	M	3-Apr-05	Redband	0.989	0.
07LC0031	30-Mile Creek	M	8-Apr-07	possible backcross	**0.784**	0.
07LC0032	30-Mile Creek	F	8-Apr-07	Redband	0.986	0.
07LC0034	30-Mile Creek	F	8-Apr-07	possible backcross	**0.761**	0.
07LC0035	30-Mile Creek	M	8-Apr-07	Redband	0.965	0.
07LC0051	30-Mile Creek	F	8-Apr-07	Redband	0.987	0.
07LC0060	30-Mile Creek	M	24-Mar-07	Redband	0.972	0.
07LC0065	30-Mile Creek	F	25-Mar-07	Redband	0.989	0.
07LC0072	30-Mile Creek	M	21-Mar-07	Redband	0.974	0.
07LC0083	30-Mile Creek	F	18-Mar-07	Redband	0.945	0.
07LC0084	30-Mile Creek	F	25-Mar-07	Redband	0.983	0.
07LC0011	Bear Creek	F	23-Apr-05	possible backcross	**0.905**	0.
07LC0024	Bear Creek	M	27-Apr-05	Redband	0.960	0.
07LC0004	Bridge Creek	F	4-May-05	Redband	0.971	0.
07LC0006	Bridge Creek	F	4-May-05	Redband	0.959	0.
07LC0025	Bridge Creek	F	27-Mar-07	possible backcross	**0.706**	0.
07LC0026	Bridge Creek	M	28-Mar-07	Redband	0.960	0.
07LC0038	Bridge Creek	F	27-Mar-07	Redband	0.979	0.
07LC0041	Bridge Creek	M	24-Mar-07	Redband	0.988	0.
07LC0045	Bridge Creek	F	2-Apr-07	possible backcross	**0.893**	0.
07LC0048	Bridge Creek	F	2-Apr-07	Redband	0.993	0.
07LC0054	Bridge Creek	M	2-Apr-07	possible F1 hybrid	**0.657**	0.
07LC0055	Bridge Creek	M	2-Apr-07	Redband	0.983	0.
07LC0058	Bridge Creek	M	2-Apr-07	Redband	0.951	0.
07LC0062	Bridge Creek	F	24-Mar-07	Redband	0.989	0.
07LC0063	Bridge Creek	M	24-Mar-07	Redband	0.993	0.
07LC0066	Bridge Creek	M	24-Mar-07	Redband	0.971	0.

07LC0067	Bridge Creek	F		24-Mar-07	Redband	0.939	0.
07LC0070	Bridge Creek	M		21-Mar-07	Redband	0.991	0.
07LC0081	Bridge Creek	M		17-Mar-07	Redband	0.988	0.
07LC0001	Gold Creek	F		4-Apr-05	possible F1 hybrid	**0.689**	0.
07LC0022	Gold Creek	F		25-Apr-05	Redband	0.946	0.
07LC0023	Gold Creek	F		25-Apr-05	possible backcross	**0.768**	0.
07LC0033	Gold Creek	M		5-Apr-07	Redband	0.967	0.
07LC0036	Gold Creek	M		10-Apr-07	Redband	0.978	0.
07LC0050	Gold Creek	F		23-Apr-07	**cutthroat**		
07LC0068	Gold Creek	F		23-Apr-07	possible backcross	**0.801**	0.
07LC0069	Gold Creek	M		22-Apr-07	Redband	0.969	0.
07LC0071	Gold Creek	M		3-Apr-07	Redband	0.992	0.
07LC0076	Gold Creek	M		23-Apr-07	Redband	0.981	0.
07LC0077	Gold Creek	M		22-Apr-07	possible backcross	**0.867**	0.
07LC0078	Gold Creek	M		4-Apr-07	possible backcross	**0.793**	0.
07LC0079	Gold Creek	M		23-Apr-07	Redband	0.977	0.
07LC0080	Gold Creek	F		23-Apr-07	Redband	0.942	0.
07LC0089	Gold Creek	M		23-Apr-07	possible backcross	**0.899**	0.
07LC0008	Iron Creek	F		10-May-05	Redband	0.953	0.
07LC0020	Iron Creek	M		2005	possible backcross	**0.791**	0.
07LC0021	Iron Creek	F		6-Apr-05	Redband	0.978	0.
07LC0005	North Nanamkin Creek	F		23-Apr-05	Redband	0.977	0.
07LC0010	North Nanamkin Creek	F		3-Apr-05	possible backcross	**0.918**	0.
07LC0015	North Nanamkin Creek	F		29-Mar-05	Redband	0.991	0.
07LC0016	North Nanamkin Creek	M		31-Mar-05	Redband	0.989	0.
07LC0017	North Nanamkin Creek	F		8-Apr-05	Redband	0.973	0.
07LC0029	North Nanamkin Creek	F		19-Mar-07	Redband	0.993	0.
07LC0037	North Nanamkin Creek	M		21-Mar-07	possible backcross	**0.731**	0.
07LC0040	North Nanamkin Creek	M		19-Mar-07	possible backcross	**0.898**	0.
07LC0043	North Nanamkin Creek	M		4-Apr-07	possible backcross	**0.758**	0.
07LC0046	North Nanamkin Creek	F		1-Apr-07	Redband	0.989	0.
07LC0049	North Nanamkin Creek	F		8-Apr-07	possible backcross	**0.887**	0.
07LC0053	North Nanamkin Creek			9-Apr-07	possible backcross	**0.818**	0.
07LC0059	North Nanamkin Creek	F		1-Apr-07	Redband	0.991	0.
07LC0061	North Nanamkin Creek	M		8-Apr-07	Redband	0.985	0.
07LC0082	North Nanamkin Creek	F		2-Mar-07	possible F1 hybrid	**0.674**	0.
07LC0085	North Nanamkin Creek	F	-		-	-	
07LC0052	South Nanamkin Creek	M		9-Apr-07	Redband	0.964	0.
07LC0073	South Nanamkin Creek	F		20-Mar-07	Redband	0.990	0.
07LC0075	South Nanamkin Creek	M		20-Mar-07	Redband	0.993	0.
07LC0091	Stapaloop Creek	?		30-Oct-07	Redband	0.991	0.
07LC0092	Stapaloop Creek	?		30-Oct-07	Redband	0.982	0.
07LC0093	Stapaloop Creek	?		30-Oct-07	Redband	0.969	0.
07LC0094	Stapaloop Creek	?		30-Oct-07	Redband	0.981	0.
07LC0095	Stapaloop Creek	?		30-Oct-07	Redband	0.937	0.
07LC0012	West Fork	F		29-Mar-05	possible backcross	**0.933**	0.
07LC0013	West Fork	F		29-Mar-05	-	-	
07LC0014	West Fork	F		29-Mar-05	Redband	0.990	0.
07LC0027	West Fork	M		23-Mar-07	Redband	0.963	0.

07LC0028	West Fork	F	29-Mar-07	possible backcross	**0.796**	0.
07LC0030	West Fork	F	30-Mar-07	Redband	0.982	0.
07LC0039	West Fork	M	30-Mar-07	Redband	0.975	0.
07LC0042	West Fork	M	3-Apr-07	Redband	0.964	0.
07LC0044	West Fork	M	4-Apr-07	Redband	0.984	0.
07LC0047	West Fork	F	2-Apr-07	Redband	0.991	0.
07LC0056	West Fork	M	1-Apr-07	Redband	0.980	0.
07LC0057	West Fork	M	1-Apr-07	Redband	0.991	0.
07LC0064	West Fork	M	24-Mar-07	Redband	0.995	0.
07LC0074	West Fork	F	22-Apr-07	Redband	0.995	0.
07LC0086	West Fork	F	25-Mar-07	Redband	0.991	0.
07LC0087	West Fork	F	24-Mar-07	Redband	0.996	0.
07LC0088	West Fork	M	24-Mar-07	Redband	0.986	0.
07LC0090	West Fork	M	24-Mar-07	Redband	0.996	0.