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Evaluating genetic structure among resident and migratory forms of bull trout (*Salvelinus confluentus*) in Northeast Oregon

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Abstract – Many salmonids express multiple behavioural forms within the same population, representing an evolutionary adaptation to a heterogeneous environment. For bull trout, resident and migratory forms co-occur in streams, but it is unknown whether the two forms assortatively mate. We assessed genetic differentiation between resident and migratory bull trout (using eight microsatellite loci) in the South Fork Walla Walla River. We PIT-tagged and fin-clipped bull trout and assigned individuals to behavioural subpopulations based on movement patterns. The pair-wise F_{ST} value between resident and migratory subpopulations (0.0037) was statistically insignificant, and individual-based analyses of structure using both multivariate and Bayesian approaches showed a lack of genetic structure within the population. These results have important implications for assessing population status and management; while the population may be managed as a single reproductive unit, the phenotypic variation within this population may have fitness consequences and thus merits conservation.

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Key words: gene flow; bull trout; *Salvelinus;* life-history forms; microsatellites

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Introduction

Many species exhibit behavioural diversification (e.g., multiple behavioural forms in a population or niche specialization) in response to a heterogeneous and changing environment (Northcote 1992; Lichatowich 1999). One common example of this diversification is partial migration, the phenomenon wherein part of the population migrates and part remains resident (Jonsson & Jonsson 1993). Through exploitation of multiple habitat types through time, resident and migratory individuals (from a single population) disperse the risk of that population becoming extinct because of local disturbances and gain access to a greater amount of resources (Gross 1991; Northcote 1992; Lichatowich 1999; Jackson et al. 2001). While natural selection may be favouring both resident and migratory forms under different environmental scenarios (Kaitala et al. 1993), behavioural diversification does not necessarily infer genetic differentiation between behavioural forms. Rather, both forms could be maintained by a stable polymorphism (abiotic and/or biotic conditions may be favourable to the maintenance of both forms over time, Smith 1970; Leimar 2005) or be strictly because of phenotypic plasticity. From a management perspective, a population that contains reproductively isolated forms (representing different behaviours or exploiting different niches) would be managed differently than one containing a single panmictic population that displays a behavioural polymorphism.

For endangered species in particular, the genetic structure within a population (as it relates to behavioural forms) may have implications with respect to (1) definition of species or distinct population segments, (2) monitoring recovery in the target species or populations, (3) assessment of population size, and (4) maintaining connectivity and genetic diversity (US Fish and Wildlife Service, USFWS, 2004). By

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determining whether different behavioural forms are genetically distinct, how they interact, and how they exploit and adapt to natural environments, we can better understand how anthropogenic impacts may alter the genetic population structure, expression, or prevalence of different behavioural forms (Gross 1991; Neraas & Spruell 2001; Wofford et al. 2005), and select conservation strategies accordingly (Dunham et al. 1999).

Within the salmonids, there is evidence for several patterns of genetic population structure related to sympatric behavioural forms (Osinov 1984; Foote et al. 1989; Wood & Foote 1996; Hendry et al. 2000; Docker & Heath 2003). For example, different morphological or behavioural forms may evolve into reproductive isolation because of exploitation of different niches (arctic char *Salvelinus alpinus,* Skulason et al. 1996; Westgaard et al. 2004; but see Nordeng 1983), or different spawn times or locations (steelhead and rainbow trout *Oncorhynchus mykiss,* Zimmerman & Reeves (2000), sockeye and kokanee *O. nerka*, Foote et al. 1989; Taylor et al. 1996; Wood & Foote 1996). Conversely, a population may exhibit behavioural diversification, yet comprise a single breeding population (e.g., brook char *S. fontinalis*, McLaughlin 2001). At a broad spatial scale, it is possible that behavioural polymorphism (e.g., migration distance, home range size, and consistency in expressing a single behavioural form over time) may differ between populations (Olsson et al. 2006). Furthermore, the degree that different behavioural forms interbreed within a population may vary within a single species (e.g., steelhead and rainbow trout, Docker & Heath 2003; Narum et al. 2004; McPhee et al. 2007; lake whitefish *Coregonus clupeaformis*, Pigeon et al. 2006), with potentially different selection pressures acting on each form (Kaitala et al. 1993; McDowall 2001).

Similar to other salmonids, bull trout (*Salvelinus confluentus*) exhibit a spectrum of behavioural and breeding strategies. Bull trout are a species of char found in the Pacific Northwest, and throughout their range they exhibit resident and migratory forms (i.e., adfluvial, anadromous, and fluvial) within the same population (Rieman & Dunham 2000). Unlike Pacific salmon that demonstrate discrete behavioural forms (e.g., anadromous and adfluvial), the difference between bull trout life-history forms is less distinct, particularly for stream-resident and fluvial fish. Adult bull trout commence spawning migrations into tributaries in the late summer (McPhail & Murray 1979; Shepard et al. 1984). Both stream-resident and migratory bull trout spawn in the fall, and may spawn every year or every other year (Rieman & McIntyre 1993). As such, a single breeding population may be comprised of 4+ generations and potentially multiple behavioural forms (Rieman & McIntyre 1993). Once spawning is complete, fluvial and adfluvial bull trout migrate to over-wintering grounds (Fraley & Shepard 1989) and resident bull trout exhibit limited movement (Jakober et al. 1998). Eggs develop over the winter and fry emerge from early April through May (Shepard et al. 1984). Fry are closely associated with the substrate for an extended period of time (McPhail & Murray 1979). In the spring, peak flows may flush young-of-year bull trout downstream (Downs et al. 2006), but peak first-time outmigration of juveniles occurs in August (Homel and Budy in press). Migratory juvenile and subadult bull trout inhabit larger more productive rivers or lakes for several years before returning to spawn (Shepard et al. 1984; Fraley & Shepard 1989). While fluvial or adfluvial bull trout may exhibit migrations up to 250 km in distance (Fraley & Shepard 1989; Swanberg 1997; Baxter 2002), there is no consistent demarcation based on distance moved that distinguishes the movements of migratory fish from those of resident fish. Furthermore, the only known morphological distinction between resident and migratory bull trout is based on body size after several migrations; migratory fish often attain sizes >600 mm total length (TL) as a result of migrating to larger, more productive streams, while resident fish typically grow to ~300 mm TL (Fraley & Shepard 1989). Writ large, these complicated behavioural, reproductive, and morphological elements confound the definition of specific life-history forms, and the interpretation of the associated genetic structure within bull trout populations.

The genetic structure of bull trout across their range reflects their post-glacial dispersal and subsequent isolation (as a result of habitat fragmentation). Bull trout typically exhibit low genetic variation within populations (e.g., out of 65 bull trout populations examined by Spruell et al. 2003, 56 have $H_S < 0.299$). However, among population structure is typically quite high (Leary et al. 1993; Taylor et al. 1999; Spruell et al. 1999; Kanda & Allendorf 2001; Spruell et al. 2003; Costello et al. 2003; Reiss 2003). For example, Spruell et al. (2003) reported F_{ST} values of 0.635 between two coastal populations of bull trout, and Costello et al. (2003) reported an F_{ST} of 0.40 for two populations in the Kootenay River. Throughout their range, bull trout are typically associated with specific habitat conditions including cold, clean water, and structurally complex habitat (Rieman & McIntyre 1993). Many factors, such as loss of connectivity, habitat degradation, and introduction of non-natives, have contributed to the range-wide decline of bull trout, particularly of the migratory form (Rieman & McIntyre 1993). In response to these threats, bull trout were listed as threatened in the contiguous United States in 1999 (Department of the Interior, U. S. Fish

and Wildlife Service 1999). While genetic structure among many bull trout populations has been assessed for conservation planning (e.g., Leary et al. 1993; Spruell et al. 2003; Whiteley et al. 2006), little is known about whether behavioural variability results in patterns of non-random mating between resident and migratory fish within a population. In addition, despite the lack of a clear demarcation between the behavioural categories of 'resident' and 'migratory', differences between these life-history forms are important for assessing the effectiveness of actions directed at recovery (e.g., reconnecting migratory corridors); and, in addition to other criteria, recovery objectives require preserving the diversity of behaviours bull trout express (e.g., resident or migratory forms, emigration age; USFWS, 2002).

In this study, our goal was to evaluate whether variability in behavioural patterns was associated with assortative mating between behavioural forms. Characterization of this genetic structure is important for determining whether this population should be managed as a single panmictic breeding population (that contains behavioural variability), or as distinct populations with genetically distinct behavioural groups. Our previous analyses of the movement patterns within this population demonstrated a continuum of movement across space and time, indicating that movement distance and timing alone were insufficient to define an individuals' life-history strategy as resident or migratory (Homel and Budy in press). However, given the broad array of behaviours that a single life-history form may express, it is insufficient to use movement distance and timing alone as metrics to define life-history forms. Instead, we described behavioural patterns using a functional definition of migration (i.e., migration is annual directed, purposeful movement between distinct habitat types, e.g., Dingle 1996), and determined that our population contains both migratory and resident fish. Those fish exiting the study area were defined as migratory as they are making a directed, distinct shift in habitat types (described in the study area), and many of those fish ultimately completed multiple spawning migrations (further described in the Methods). Therefore, the specific objective of this study was to evaluate whether resident and migratory fish exhibited assortative mating, as demonstrated by genetic differentiation.

Methods

Study area

The South Fork Walla Walla River (SFWW) originates at elevations near 1800 m in the Blue Mountains of Northeast Oregon, confluences with the North Fork Walla Walla near the town of Milton-Freewater, and flows into the Columbia River upstream of McNary Dam (Fig. 1). We selected this river as our study area for two reasons. First, it was known to contain a relatively large population of bull trout (8–12,000 fish, Al-Chokhachy 2006), previously described as containing both resident and migratory forms (Buchanan et al. 1997). Second, the SFWW and main-stem Walla Walla include a range of habitat types from pristine to highly degraded. Within the SFWW, the habitat condition is generally of high quality, with few forest

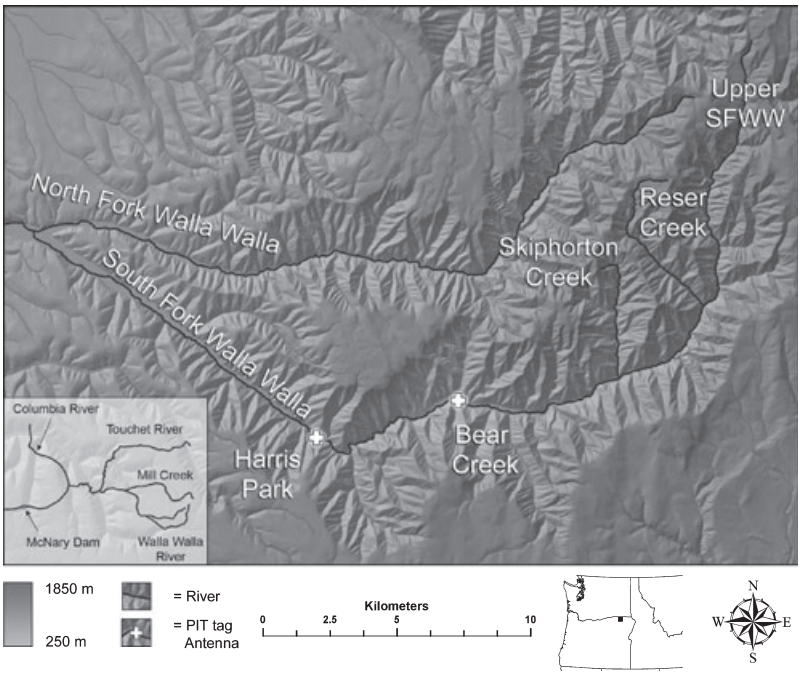

Fig. 1. Map of the South Fork Walla Walla River study area with locations of passive antennae and tributaries marked.

related impacts and limited recreational activity that would impact the stream corridor (particularly in the headwaters, Buchanan et al. 1997). Downstream of the confluence with the North Fork Walla Walla River, the habitat conditions degrade with respect to increased water temperature, simplified channel and habitat, impoundments, and irrigation withdrawals that severely deplete flow (Buchanan et al. 1997), and act as a seasonal migration barrier. Skiphorton Creek and Reser Creek are the major tributaries to the SFWW and most observed spawning activity occurs in proximity to (or within) these pristine tributaries.

Within the Walla Walla Basin watershed, the SFWW, Mill Creek, and the Touchet River all contain populations of bull trout (Buchanan et al. 1997; Fig. 1). According to Oregon and Washington criteria, population status in Mill Creek is 'of special concern' (Buchanan et al. 1997) or alternatively 'healthy' (WDFW, 1997), the Touchet River status is considered 'unknown' (WDFW, 1997), and the SFWW population status is 'low risk' (Buchanan et al. 1997); all subpopulations are listed as 'depressed' by the USFWS (USFWS 2004). Although the SFWW bull trout population is of low extinction risk, irrigation withdrawals and diversion dams along the Walla Walla River prevent interaction (from Spring until Fall) between SFWW bull trout and the bull trout populations from Touchet River and Mill Creek. It is thought that historically bull trout had access to, and used, the Columbia River (Buchanan et al. 1997), but recent telemetry studies (Mahoney 2001, 2002) have not confirmed contemporary use of the Columbia River, and PIT-tagged fish from the SFWW have not been detected at antenna located in Mill Creek or the Columbia (Homel and Budy in press).

Study design phase I: mark-recapture study

This genetics study is a component of a broader effort to gather comprehensive population assessment data on the SFWW, critical for recovery planning (Al-Chokhachy et al. 2005; Al-Chokhachy 2006). For the first phase of this broader effort, we conducted a large mark-recapture study in the main-stem of the SFWW during July and August from 2002–2005 (Al-Chokhachy et al. in press). The study area extended from Harris Park to the confluence with Reser Creek (a distance of 21 stream km, Fig. 1), and was divided into 103 adjacent, 200 meter reaches (Harris Park is reach 1). Each year we systematically sampled every fifth reach (further described in Al-Chokhachy et al. in press), which entailed: (1) capturing bull trout, (2) implanting passive integrated transponder tags (PIT tags) into the ventral cavity of all bull trout >120 mm TL, and (3) removing a 2–5 mm^2 fin clip from the anal fin. We stored fin clips in 100% ethanol until they were processed by a laboratory. From 2002–2005, we captured, tagged, and fin-clipped >1300 bull trout from the main-stem SFWW.

Study design phase II: defining behaviour

In the second phase of this study, we used fish recapture data (from the mark-recapture study), in combination with detection of tagged fish at stationary antennae, to determine the movement patterns and behavioural strategy of individuals in our population (Homel and Budy in press). In 2002, we installed two passive PIT tag antennae in the SFWW (one at Harris Park, and one at Bear Creek, Fig. 1), to record the individual tag number of marked fish that passed through the antenna loop. The antenna at Harris Park was located at a transition point in habitat quality; upstream of Harris Park, the river exhibits complex braiding, in-stream structure, and temperatures within the thermal tolerance of bull trout. Downstream of Harris Park, a paved road follows the course of the river, the river is simplified and/or channelized, and stream temperatures reach or exceed the thermal tolerance of bull trout. Conditions progressively deteriorate downstream of the confluence with the North Fork Walla Walla River. Given this distinct habitat transition, fish moving downstream past the Harris Park antenna would be considered migratory fish.

Based on our annual mark-recapture sampling and antenna detections in the study area, we defined two putative behavioural subpopulations for genetic analysis: (1) 'known migrants,' and (2) 'likely residents.' 'Known migratory' fish were those fish that exhibited a downstream migration, exiting the study area at Harris Park (N = 304). In converse, 'likely resident' fish were fish that were never detected at either antenna, and were recaptured annually in the same or adjacent stream reach (N = 83). As a result of variable antenna detection probability and efficiency during select periods of time in 2003 and 2004 (50–100% resulting from power outages), we could not define a fish as resident, but rather as 'likely resident' given the possibility of a missed migration detection at the Harris Park antenna. However, to mitigate the potential effects of missed detections at the Harris Park antenna on the identification of an individuals' behavioural strategy, we removed all samples from our database that were only detected at the Bear Creek antenna, as these fish could be either resident fish near the antenna, or migratory fish that were not detected at the Harris Park antenna. From 2002–2006, no likely-resident fish were ever detected at either antenna, suggesting that our behavioural definitions were appropriate. These two behavioural classes were used for all subsequent analysis.

Study design phase III: genetic structure

Finally, for this current genetics study, we combined data on the behavioural strategy of individuals with a microsatellite analysis to evaluate behaviourally based neutral genetic structure within one large stream population. From the initial phase of this study, we had >1300 fin clips available to us for genetic analysis. These fin clips were from fish >120 mm TL, representing multiple size classes and both resident and migratory behavioural patterns, and were collected throughout the river from 2002–2005. As we were interested in identifying genetic structure within the population as it relates to behavioural form, we selected samples for which we had described a behavioural strategy. From our pool of 304 migratory bull trout, and 83 likely-resident bull trout, we randomly selected 109 samples for genotyping (migratory $N = 57$, mean TL at capture = 354 mm, range TL = 122–720 mm; likely resident $N = 52$, mean TL at capture = 222 mm, range TL = 139–342 mm).

Genetic processing conditions

We extracted total genomic DNA from 109 fin clips using a 'salting out' protocol (Sunnucks & Hales 1996). We used PCR to amplify 11 microsatellite loci from these templates following the reaction conditions described by the original authors and summarized by the USFWS Abernathy Fish Technology Center (AFTC). These loci were members of a core set of standard bull trout loci, with the forward primers fluorescently labelled (6FAM or HEX): Omm1128 HEX, Sfo18 HEX (Angers et al. 1995), Sco200 6FAM, Sco202 HEX, Sco216 6FAM, Sco220 HEX (DeHaan & Ardren 2005), Sma22 HEX (Crane et al. 2004), Sco102 6FAM, Sco105 6FAM, Sco109 6FAM, Sco110 6FAM (Shaklee, WDFW Olympia, WA, summarized by USFWS AFTC 2003). We conducted PCR using approximately 20 nanograms of sample DNA with a total reaction volume of 15 μl. We assessed the PCR products on a 1.0% agarose gel. Diluted PCR products were run on an ABI3730 DNA analyzer (Applied Biosystems, Inc.) with a LIZ3730 size standard, analyzed using Genescan Software, and scored using GeneMapper software (Applied Biosystems, Inc.). Mention of brand names does not imply endorsement. Although we ran all PCR reactions separately, we combined (multiplexed) PCR products from the following combinations of loci for runs on the ABI3730 DNA analyzer: Sco216 and Sco202, Sco200 and Omm1128, and Sco105 and Sco220. As a quality control measure, we ran replicates of PCR products from a small proportion of the samples on individual lanes to assure that multiplexing did not result in mis-scoring.

Data analysis

We evaluated the null hypothesis of random mating between resident and migratory fish using a combination of complimentary statistical methods. First, we assessed the microsatellite loci for evidence of linkage disequilibrium and Hardy-Weinberg disequilibrium. Next, we compared resident and migratory fish with respect to allelic diversity (A, corrected for unequal sample size with rarefaction), multi-locus expected heterozygosity (He), Fisher's exact test for genic differentiation (comparison of allele frequencies across loci), and F_{ST}, using GENEPOP (Version 3.4, Raymond & Rousset 1995; options 1–6). Next, we conducted a factorial correspondence analysis (FCA) to depict potential clustering of individuals within each behavioural group using the program Genetix (Version 4.05, Belkhir et al. 1996–2004). Finally, we evaluated the genotypes of all samples without *a priori* assumptions about their putative subpopulation of origin to determine the most probable number of subpopulations (K) within the total population using the program Structure (Version 2.1, Pritchard et al. 2000). We selected a burn in length of 100,000 replications, a run length of 100,000, and ran 100 replications using 5 K values (1–5).

Results

Before testing for HWE, we assessed whether our microsatellite loci were polymorphic and in linkage disequilibrium. Two loci, Sfo18 and Sco102, were monomorphic in our samples, and Sco110 was the same locus as Sco216 (based on identical primer sequences), so we removed Sfo18, Sco102, and Sco110 for all subsequent analyses. We found no evidence of linkage disequilibrium between any of the remaining 8 loci ($P > 0.05$). These 8 loci were polymorphic in both putative subpopulations with an observed number of alleles ranging from 3 to 21 (Table 1). Both putative subpopulations, and the entire population as a whole, conformed to expected

Table 1. Total number of observed alleles and allele size range per locus, across all samples and for each subpopulation.

	Total		Resident		Migratory	
Locus	# Alleles	Size range	# Alleles	Size range	# Alleles	Size range
Omm1128	9	275–354	7	275–354	8	275–354
Sco200	7	133–157	6	133–153	7	133–157
Sco202	3	127–135	3	127–135	3	127–135
Sco216	5	239–263	4	239–263	4	239–255
Sco109	11	266–387	10	266–387	8	266–387
Sco105	6	164–208	5	164–208	6	164–208
Sco220	7	299–328	6	299–328	5	299–319
Sma22	21	204–283	17	204–283	18	204–279

Hardy-Weinberg genotypic proportions at all loci ($P = 0.28$).

We evaluated the potential for non-random mating between resident and migratory fish in the SFWW using several statistical tests, all of which failed to demonstrate neutral genetic differentiation between behavioural groups. Allelic diversity (mean number of alleles across loci per subpopulation, with the larger migratory sample size rarefied to the smaller resident sample size) was similar for resident and migratory fish (7.25 and 7.38 respectively). Multi-locus expected heterozygosity (He) and observed heterozygosity (Ho) were also similar for each subpopulation (He = 0.35 and 0.34 respectively, Ho = 0.34 and 0.33 respectively). A pair-wise comparison of allele frequencies across loci was not significantly different for resident and migratory subpopulations according to Fisher's exact test for genic differentiation ($P = 0.85$). This lack of structure between groups was further corroborated by an insignificant pair-wise F_{ST} value of 0.0037 and a similarly low combined F_{IS} value of 0.04.

Based on an individual-based FCA of genetic variation across behaviourally defined groups, we found a complete absence of multidimensional clustering (i.e., genetic structure). The first four principal components in the FCA each explained about 4% of the variation between samples (with the first component explaining 4.85%); thus, a single component was sufficient to describe the variation in our sample. Our initial FCA depicted three outliers for which we could determine no common denominator, although each contained a unique rare allele. We removed these three outliers from the analysis and still found no clustering in our FCA plot based on behavioural form or other potential unidentified structure (Fig. 2). Overall, the similarity in allele frequencies, the low F_{ST} values, and the lack of clustering in the FCA plot suggest that resident and migratory fish comprise a single panmictic breeding population, although it is possible that the number of loci or the sample size were inadequate to detect a very low level of assortative mating.

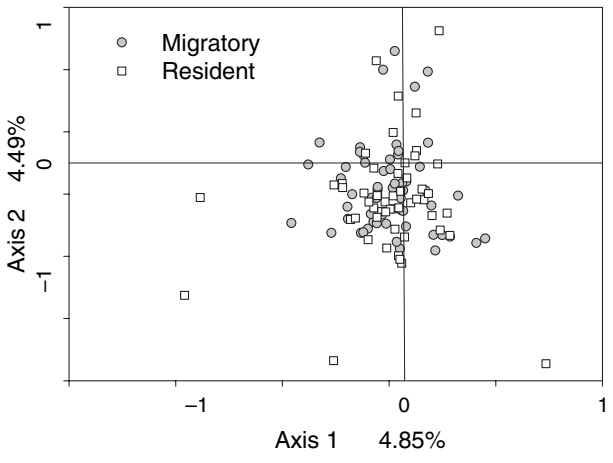

Fig. 2. Factorial correspondence analysis of the multidimensional genotype of 109 resident and migratory samples. Each putative subpopulation is represented by a unique symbol.

Our Bayesian analysis of potential population structure (ranging from 1 panmictic population to five discrete subpopulations, $K = 1$-5) using the program Structure demonstrated complete panmixia; all individuals were assigned an equal probability of belonging to each subpopulation, irrespective of the K-value selected, but $K = 1$ had the highest probability. Pair-wise F_{ST} values for all K-groups were low (all < 0.05), indicating that the most likely structure for the population is a single panmictic breeding population.

Discussion

In order to better understand the relationship between behavioural variation and neutral genetic population structure, we evaluated the genotypes of 109 individuals, representing two putative behavioural subpopulations from the SFWW. Based on our genetic results, we failed to reject the null hypotheses that resident and migratory fish comprise a panmictic breeding population.

Our multiple tests for subpopulation structure all indicated that the SFWW bull trout population is panmictic with respect to our defined behavioural groups. While this represents the first such study of interbreeding between behavioural forms in bull trout, similar results have been observed (with varying degrees of interbreeding) for co-occurring arctic char morphs (in a transplant experiment in southern Norway, Nordeng 1983), brook trout behavioural forms ($F_{ST} = 0.0007$ in 2000 and $F_{ST} = 0.012$ in 2001, Theriault et al. 2007), and brown trout (*Salmo trutta*) behavioural forms (Charles et al. 2005). However, in contrast to Nordeng (1983), Westgaard et al. (2004) found significant reproductive isolation between co-occurring arctic char morphs in northern Norway ($F_{ST} = 0.032$). These results suggest that the degree of interbreeding between forms may be mediated by environmental factors or landscape level processes (McLaughlin 2001). Given the variability observed in other salmonids species, it is possible that, across the range of bull trout, the degree of interbreeding between behavioural forms within a population may also vary.

We suggest two possible mechanisms by which interbreeding between forms may occur: (1) both forms may significantly overlap in their selection of spawn sites, resulting in unintentional interbreeding, and (2) small resident males may exhibit the 'sneaking'

tactic on large migratory females. In our study of bull trout movement patterns in the SFWW, we observed that resident and migratory fish co-occurred, and that resident fish moved up to 14 km within the study area (during the spawning season, Homel and Budy in press). Previous redd counts in the SFWW by the Oregon Department of Fish and Wildlife identified that the majority of spawning activity occurred from approximately 4 km above Bear Creek, to above Reser Creek, including the Skiphorton and Reser Creek tributaries (Buchanan et al. 1997). However, Al-Chokhachy et al. (2005) demonstrated that most of these redds likely belonged to larger migratory fish. Despite an imprecise knowledge in the spawning location of resident fish, the ubiquitous spawning location of migratory fish would suggest spatial overlap. Furthermore, our data on fish movement suggests that resident and migratory fish are moving upstream to spawning areas at the same time of year (Homel and Budy in press). Potentially, this temporal and spatial overlap in spawning could result in significant interbreeding, independent of mate selection. For example, Docker & Heath (2003) documented gene flow between co-occurring resident rainbow trout and anadromous steelhead, while Narum et al. (2004) detected genetic divergence between those same forms as a result of differing spawn site selection.

A second possible mechanism for interbreeding between forms is that smaller resident males may be selecting larger migratory females as mates through exhibiting a 'sneaking' tactic. While female fish have been shown to assortatively mate with similarly sized male fish (e.g., Japanese char, *Salvelinus leucomaensis*, Maekawa et al. 1994; sockeye salmon, Foote 1989), male fish may exhibit a sneaking tactic, and breed with significantly larger, more fecund female fish (Gross 1991; Groot & Margolis 1991). As a result of that tactic, resident males would be able to increase their fitness relative to breeding with smaller, less fecund resident females. Within the salmonids, there are numerous examples of this tactic. Theriault et al. (2007) noted that gene flow between resident and anadromous brook trout was mediated by resident males mating with both resident and anadromous females. Wood & Foote (1996) documented a similar pattern between male kokanee salmon and female sockeye salmon. Sneaker males have also been documented in coho salmon (*Oncorhynchus kisutch*, Gross 1991) and potentially in rainbow trout (mating with steelhead, Zimmerman & Reeves 2000; Narum et al. 2004). Although not expressly observed in SFWW spawning surveys, it is possible that bull trout may express this sneaking tactic as well.

In our assessment of behaviourally based genetic population structure, there were two potential, albeit minor, limitations to our study: (1) an inability to sample fish smaller than 120 mm TL, and (2) low genetic variability within the population. We only collected fin clips from fish larger than 120 mm TL as this was the smallest size that we could tag, and we needed movement information to analyze genetic structure related to behaviour. However, bull trout are intergenerational breeders, and we do not believe that our failure to sample fish smaller than 120 mm TL resulted in any bias to our results. The second potential limitation to our study was that we detected a low level of genetic variation in our population which could make it difficult to detect subpopulation structure. However, other bull trout populations have expressed similar levels of within-population genetic variation (e.g., bull trout populations in the Yakima River Basin had H_O values ranging from 0.21–0.45 across six microsatellite loci, independent of population size, Reiss 2003). Furthermore, as we used metrics to assess genetic population structure that were relative to the total amount of genetic variation in our population (e.g., F_{ST}), we do not believe that the low genetic variation in our population would have precluded detection of genetic differences between behavioural forms, were they present. While it is possible that the low variation we detected, in combination with our sample size, could have resulted in a type II error, this error would only have precluded the detection of a very low level of assortative mating between resident and migratory fish, and would not otherwise have influenced the interpretation of our results.

Our study represents the first genetic comparison of behavioural forms in bull trout, and was unique in that our *a priori* definition of behavioural groups was based on extensive monitoring of the movement patterns of >1500 fish (Homel and Budy in press). By understanding the continuous movement patterns of our fish, we were able to identify a potential behaviour-related cause for the lack of genetic structure we observed. This pairing of movement studies (via tagging) and genetic analysis improved our fundamental understanding of the evolutionary and ecological interactions between behavioural forms in this population, and how that interaction may shape patterns of random mating between behavioural groups.

In managing the sympatric behavioural forms of an endangered species, it is important to consider limitations in our understanding of behavioural forms, particularly in the case of random mating. The presence of gene flow between sympatric behavioural forms does not necessitate a lack of adaptive variation between these forms, provided that selective pressure for each form outweighs gene flow (Rice & Hostert 1993). However, this common pattern of behavioural diversification and genetic similarity (at neutral markers) reflects our limited understanding of the mechanisms

that determine behavioural strategy. In this study, resident and fluvial bull trout in the SFWW comprise a single panmictic breeding population. While we were unable to reject the null hypothesis of panmixia at the level of neutral markers for this population, under different habitat conditions, different connectivity scenarios, greater behavioural differentiation between resident and migratory fish, or over a longer time scale, there is potential that a different genetic pattern could exist. Furthermore, since measures of neutral molecular- and quantitative-genetic variation (Pfrender et al. 2000) and population subdivision (Lynch et al. 1999) are often disconnected, a study of quantitative genetic variation (heritability, h^2) and subdivision (Q_{ST}) based on the behavioural morphs would enhance the context of our conclusions.

Management implications

The potential for behavioural groups within a population to randomly mate presents an interesting opportunity for conservation of bull trout. Rather than create specific recovery goals for each life-history form, our research suggests that management should focus on maintaining phenotypic variation within the population. While we do not yet understand the mechanism driving the adoption of a life-history tactic in bull trout, we do know that multiple life-history forms within a population increase that population's resistance to extirpation (via occupation of multiple habitat patches through time, Gross 1991). Furthermore, both resident and migratory forms fulfill unique ecological functions. Migratory fish make an important demographic contribution to the population as a result of the increased fecundity associated with their larger body size (Al-Chokhachy 2006). Conversely, resident fish fill a predatory niche in the natal stream throughout the year, and potentially could bolster migratory populations via random mating, if behavioural forms can give rise to one another (as suggested in bull trout by Dunham et al. 2003; and demonstrated in sympatric sockeye and kokanee salmon, Taylor et al. 1996). Given the importance of both resident and migratory forms, management must focus on (1) preserving local resident populations (that display local adaptations), and (2) addressing limiting factors for both resident and migratory bull trout.

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