Conservation Genetics
https://doi.org/10.1007/s10592-019-01182-4

RESEARCH ARTICLE

Genetic mixture analyses in support of restoration of a high value recreational fishery for rainbow trout (*Oncorhynchus mykiss*) from a large lake in interior British Columbia

Eric B. Taylor[1] · **Cody Foley**[1] · **Matt Neufeld**[2]

Received: 12 December 2018 / Accepted: 5 April 2019

Abstract

Genetic mixture analysis is an important tool to apportion catch amongst potential component populations contributing to a fishery. We used variation at 10 microsatellite DNA loci to assess the level of genetic divergence between two ecotypes of rainbow trout (*Oncorhynchus mykiss*) that naturally co-occur in Kootenay Lake, southeastern British Columbia, and to exploit such divergence in a mixture analysis. One form, "Gerrard" rainbow trout, historically matured at sizes greater than 60 cm and 5 kg, spawns at a lake outlet after upstream migration in a large river tributary to Kootenay Lake, and is highly prized in the recreational fishery. The other form, "non-Gerrard" rainbow trout, is also native to the lake and matures at smaller sizes and spawns in numerous small streams tributary to Kootenay Lake. Recent declines in growth rate of Gerrard rainbow trout, however, has made them difficult to identify by size in fishery samples. Gerrard (N = 130, 6 sites) and non-Gerrard trout N = 312, 15 sites) were highly divergent from one another ($F_{ST} = 0.14$, $P < 0.001$) and constituted distinct genetic groups in model-based clustering analyses. Genetic mixture analyses of fishery samples indicated a high degree of accuracy in estimating mixture proportions; 100% Gerrard simulated fisheries were estimated to contain 99.9% Gerrards (95% confidence intervals of 99.8–100%) while 100% non-Gerrard rainbow trout simulated fisheries were estimated to contain 100.0% non-Gerrard trout (100–100%). Across eight fishery creel samples obtained between 2015 and 2017 (N = 527 fish), mixture analysis estimated the fishery to contain an average of 73.4% (95% confidence interval = 68.4–74.6%) Gerrard and 26.6% (23.4–31.6%) non-Gerrard trout. Realistic fishery simulations demonstrated strong agreement with empirical results; the average simulated values for Gerrards was 73.4% (65.9–80.0%) and for non-Gerrards was 26.8% (20.0–34.1%). Assignment tests resulted in an average 98.5% (±0.066%) assignment confidence; 385 fish from the fishery samples were assigned to the Gerrard group (0.73) and 142 (0.27) to the non-Gerrard rainbow trout genetic group. Fitting length-at-age data for genetically assigned fishery samples to a von Bertalanffy growth model found greatest support for a model employing ecotype-specific L_∞ (= 59.6 and 52.9 cm for Gerrards and non-Gerrards, respectively, both $P < 0.001$), and t_0 (= − 1.36 and − 2.58, respectively, both $P < 0.05$), but a common K (= 0.189, $P < 0.001$). Our mixture analyses are being used to monitor catches and better understand the feeding and migration biology of these sympatric ecotypes of rainbow trout.

Keywords Genetic mixture analysis · Microsatellite DNA · Sympatric ecotypes · Rainbow trout · Recreational fisheries

Electronic supplementary material The online version of this article (https://doi.org/10.1007/s10592-019-01182-4) contains supplementary material, which is available to authorized users.

✉ Eric B. Taylor
etaylor@zoology.ubc.ca

[1] Department of Zoology, Biodiversity Research Centre and Beaty Biodiversity Museum, University of British Columbia, 6270 University Blvd, Vancouver, BC V6T 1Z4, Canada

[2] British Columbia Ministry of Forests, Land and Natural Resource Operations and Rural Development, 401-333 Victoria Street, Nelson, BC V1L 4K3, Canada

Introduction

Recreational fishing is a major global activity with tremendous economic, social, and biological impacts on human societies and ecosystems (Pitcher and Hollingsworth 2002). In Canada and the US, recreational fishing generates tens of billions of dollars of direct and indirect revenue per year and involves as much as 20% of the populations of these countries (Cooke and Murchie 2015; Tufts et al. 2015). British Columbia (BC), Canada, is richly endowed with marine and freshwater-based fisheries; freshwater recreational

Published online: 12 April 2019

fishing alone generates almost $1 billion in indirect, direct, and induced economic impacts (FFSBC 2013). In BC, the rainbow trout (*Oncorhynchus mykiss*) has consistently represented the single most important freshwater sport fish in terms of catch (Levey and Williams 2003). For instance, in 2010 rainbow trout comprised 58% of the reported catch, four million fish, and more than three times that of the next closest species, cutthroat trout (*O. clarkii*) at 15% (FFSBC 2013). The popularity of rainbow trout as a game fish is a function of its distribution in all major watersheds of BC, its physical attributes, and because it is the most widely stocked game fish (e.g., FFSBC 2018a, b). One of the most well-known fisheries for rainbow trout occurs in Kootenay Lake, located in southeastern BC, a large lake (400 km^2 surface area, 104 km long) within the Canadian portion of the Columbia River drainage (Fig. 1). Here, rainbow trout are part of a distinctive interior evolutionary lineage which, in the US at least, is often referred to as the "redband trout" (*O. m. gairdneri*) and is distinct from the coastal lineage (*O. m. irideus,* Behnke 1992; McCusker et al. 2000). The lake has had a recreational fishery since the early 1900s and is renowned for large-bodied rainbow trout known as "Gerrards" which not infrequently exceed 5 kg in weight (Northcote 1973; Irvine 1978). The Gerrard rainbow trout are exploited largely in seasonal (autumn, winter, and spring) lake trolling fisheries. The fishery is of high value, generating between 20,000 and 40,000 angler days and $5–10 million annually to the local economy (Andrusak and Andrusak 2012). The name Gerrard stems from the name of an abandoned town located adjacent to the spawning location of these fish in the Lardeau River, at the outlet of Trout Lake, which ultimately drains into the north arm of Kootenay Lake (Fig. 1). The lake also supports abundant native populations of smaller rainbow trout (< 2 kg) that spawn in numerous tributaries other than the Lardeau River and that are also exploited in recreational fisheries (Northcote 1973). The different feeding habits of Gerrard and tributary rainbow trout (hereafter "non-Gerrard" rainbow trout) are thought to explain, in large part, their differences in size-at-maturity and the bimodal size distribution in the fishery (Northcote 1973; Keeley et al. 2005). At sizes less than 30 cm, Gerrard rainbow trout feed on terrestrial insects and the opossum shrimp *Mysis diluviana* (introduced in 1949, Sparrow et al. 1964, previously thought to be *M. relicta*). When they exceed 30 cm, however, Gerrards switch to piscivory and feed heavily on kokanee, the freshwater-resident form of sockeye salmon (*O. nerka*), and occasionally other fishes (Northcote 1973; Irvine 1978). By contrast, non-Gerrard rainbow trout feed heavily on invertebrates, principally *M. diluviana* and various aquatic and terrestrial insects and only occasionally on kokanee or other fishes (Northcote 1973; Irvine 1978). The large-bodied, piscivorous phenotype exhibited by Gerrards is rare across the range of

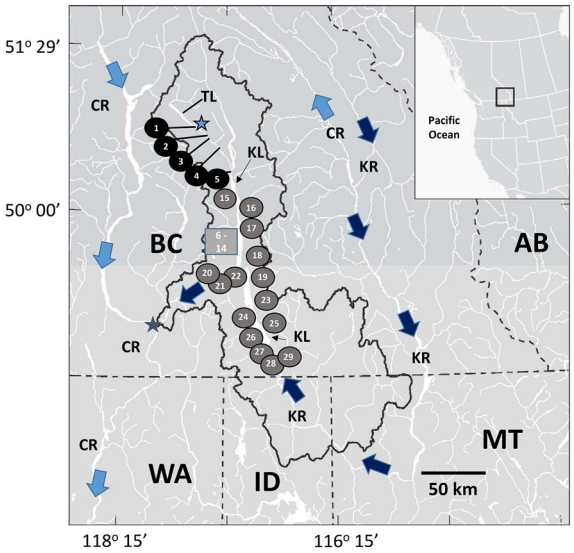

Fig. 1 Kootenay Lake, British Columbia, and locations of rainbow trout (*Oncorhynchus mykiss*) sample sites. Black filled circles represent samples of Gerrard rainbow trout from the Lardeau River, dark grey-filled circles represent non-Gerrard rainbow trout, and the light grey-filled square represents a total of eight contemporary, and one historical, fishery samples of rainbow trout obtained from the fishery operating largely in the north arm of Kootenay Lake (i.e., samples 6–14 in Table 1). Numbers represent localities shown in Table 1 and the light blue star is located at Gerrard. *BC* British Columbia, *AB* Alberta, *WA* Washington, *ID* Idaho, *MT* Montana, *CR* Columbia River, *KR* Kootenay River (or Kootenai River when in the US), *TL* Trout Lake. The dark blue arrows show the flow direction of the upper Kootenay River south and then northwest into the south end of Kootenay Lake and continuing from the western outlet of Kootenay Lake to the confluence of the Kootenay and Columbia rivers (dark blue star). The light blue arrows show the flow of the upper Columbia River from its origin at Columbia Lake northwest, then southeast, and southwest as it crosses the Canada-US border (horizontal dashed line). The dark black outline contains the Kootenay Lake basin and the inset shows the Kootenay Lake/River watershed in relation to western North America. (Color figure online)

freshwater-resident *O. mykiss* (Keeley et al. 2005) and would likely justify conservation assessment of Gerrard rainbow trout as a distinct designatable unit under Canada's *Species at Risk Act* (e.g., see Taylor et al. 2013).

The Gerrard trout fishery has recently declined in that the numbers of fish caught that exceed 2 kg in weight has dropped considerably since 2013 (Askey and Bisson 2016). The possible reasons for this decline are varied, but probably centre on reductions in kokanee—their key forage fish—whose spawning escapements have plummeted from over 2 million adults to under 20,000 between 1998 and 2015 (see Askey and Bisson 2016; Kurota et al. 2016). The reduced average size of rainbow trout in the fishery now makes it very difficult to use size to distinguish Gerrard from non-Gerrard rainbow trout in creel samples and complicates management of the lake's fishery (Fig. 2). For instance, harvesting of Gerrard trout is subject to a special licence

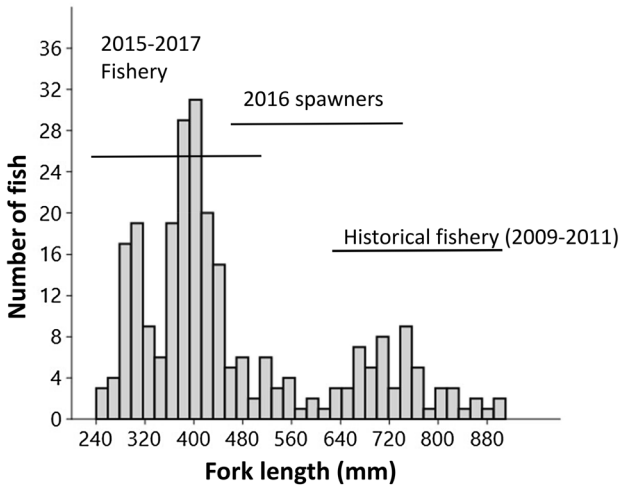

Fig. 2 Fork lengths (mm) of individual rainbow trout (*Oncorhynchus mykiss*) from recreational fisheries and at the Gerrard spawning site in the Lardeau River, Kootenay Lake, BC, between 2009 and 2017

purchase. Further, the apparent convergence of body size in the fishery makes it difficult to understand the relative proportions of Gerrards and non-Gerrards in creel surveys which greatly complicates population monitoring and the potential imposition of differential harvest regulations to promote sustainable exploitation.

One way that might distinguish Gerrard from non-Gerrard rainbow trout is genetic analysis that exploits the fact that these fish spawn in different tributaries and thus may have accumulated neutral genetic differences that could be used to identify them. The strong spatial separation of spawning areas of Gerrard and non-Gerrard populations likely promotes genetic differentiation between these ecotypes of rainbow trout. Further, there are a number of differences between the spawning and early feeding habitats of Gerrard and non-Gerrard rainbow trout. First, the spawning area at Gerrard is at the outlet of Trout Lake (27 km^2), where the Lardeau River forms, and is about 57 km upstream from the north end of Kootenay Lake. By contrast, non-Gerrard rainbow trout spawn in much smaller tributaries, none of which are lake-headed, after an upstream migration of few hundred metres to a few km at most (M. Neufeld, unpubl. data). Further, Hartman (1969) suggested that the spawning environment at Gerrard acted to select for large body size owing to large gravel size and high water velocity relative to spawning areas in smaller Kootenay Lake tributaries. Consequently, divergent selection between spawning environments could contribute to divergence between ecotypes through isolation-by-adaptation (e.g., Bond et al. 2014). The lack of establishment of a smaller-bodied (i.e., typically less than 1 kg at maturity), invertebrate specialist form of rainbow from Pennask Lake, Fraser River drainage in southcentral BC, that was repeatedly introduced to the Lardeau River between 1930 and 1948 is consistent with this hypothesis (Northcote 1973; FFSBC 2018b).

Owing to their abundance and variability, microsatellite DNA loci have been an efficient way to study population differentiation, individual identification, and fishery mixture analyses in fishes and rainbow trout in particular (e.g. Chistiakov et al. 2006; Bott et al. 2009; Tamkee et al. 2010; Brenden et al. 2015; Bradbury et al. 2015). Here, we report results of a microsatellite DNA survey of samples of rainbow trout from Kootenay Lake to answer three questions. First, is there evidence of more than one genetic population of rainbow trout in samples obtained from the fishery and various spawning tributaries? Second, do distinct genetic populations correspond to Gerrard rainbow trout (identified by weights of > 2 kg and approximately > 50 cm in fork length and/or spawning at Gerrard or residence in the Lardeau River as juveniles) and non-Gerrard trout (i.e., those of smaller average size and/or spawning or feeding in other tributaries)? Third, if genetic differences do exist, can they be exploited to assign fish in the creel samples to these different phenotypes with a high degree of confidence in post-fishery conservation assessments or in within-lake life-history studies?

Materials and methods

Tissue samples

Tissue samples consisted of fin clips (adipose or pelvic fins) stored in 95% non-denatured ethanol or dried scale samples. Tissue samples were obtained from two time periods. "Contemporary" samples (i.e., after size differences between Gerrards and non-Gerrards became less apparent in the fishery) consisted of fin clips supplied by the BC Ministry of Forests, Land and Natural Resource Operations and Rural Development in Nelson, BC, and were obtained between 2015 and 2017. Most samples were obtained either from creel surveys or by electrofishing in tributary streams during July and August. Our study was not intended to explore the full details of population structure in Kootenay Lake, but rather to examine differences between the large-bodied rainbow trout spawning at Gerrard and the assemblage of non-Gerrard rainbow trout populations spawning in a variety of much smaller streams around Kootenay Lake. Consequently, we focussed on sampling a moderate number of fish (between 15 and 30) from as large a sample of streams as possible (Fig. 1, Table 1). Fin clips from spawning Gerrard trout were obtained by angling in May, 2016. These were supplemented by samples of juvenile trout from the Lardeau River and three of its tributaries (Hope Creek, Poplar Creek, and Cascade Creek) collected by electrofishing (Table 1). "Historical" samples (i.e., before size differences between Gerrards

Table 1 Summary of Kootenay Lake rainbow trout samples examined at ten microsatellite DNA loci

Sample	Date sampled	N	A_R	H_o	H_E
1. Gerrard spawners	2016	24	4.5	0.45	0.48
2. Hope Creek	2016	24	5.1	0.52	0.56
3. Poplar Creek	2016	4	NA	NA	NA
4. Cascade Creek	2016	3	NA	NA	NA
5. Lardeau River	2016	24	4.7	0.51	0.55
6. Historical fishery[a]	2009–2011	51	4.3	0.47	0.45
7. Fall fishery 1	2015	48	6.3	0.54	0.68
8. Fall fishery 2	2015	48	5.9	0.52	0.63
9. Fall fishery 3	2015	48	6.1	0.51	0.64
10. Fall fishery 4	2015	40	6.0	0.49	0.62
11. Spring fishery	2016	41	5.8	0.51	0.61
12. Fall fishery	2016	55	5.9	0.51	0.61
13. Spring fishery	2017	142	9.1	0.54	0.62
14. Fall fishery	2017	105	7.2	0.51	0.67
15. Bjerkness Creek	2016	4	NA	NA	NA
16. Tam O'Shanter Creek	2016	23	7.5	0.69	0.73
17. Hendryx Creek	2016	19	6.0	0.59	0.65
18. Crawford Creek	2016	24	6.6	0.62	0.67
19. Gray Creek	2016	22	7.1	0.69	0.74
20. Kokanee Creek	2016	2	NA	NA	NA
21. Redfish Creek	2016	20	5.6	0.67	0.65
22. West Arm	2017	23	6.6	0.61	0.76
23. La France Creek	2016	20	7.2	0.68	0.69
24. Midge Creek	2016	1	NA	NA	NA
25. Sanca Creek	2016	19	5.3	0.66	0.63
26. Cultus Creek	2016	22	6.0	0.64	0.69
27. Summit Creek	2016	21	5.8	0.59	0.62
28. Corn Creek	2016	22	6.8	0.57	0.64
29. Goat River	2016	22	5.8	0.67	0.67

N sample size, A_R mean allelic richness across eleven loci (standardized to a minimum sample size of 19), H_o mean observed heterozygosity, H_E mean expected heterozygosity. Sample names that are underlined represent learning samples of Gerrard rainbow trout, all other non-fishery samples were learning samples for non-Gerrard trout. Values of observed heterozygosity that are underlined are significantly lower than expected. Statistics were not calculated for samples with fewer that 15 individuals (=NA). Sample numbers refer to localities in Fig. 1

[a]All fish were > 65 cm fork length and considered to be Gerrard rainbow trout

and non-Gerrards became apparent in the fishery) consisted of scales and were collected from creel surveys between 2009 and 2011 and were from fish that were > 65 cm in fork length (and > 2 kg) and were considered to be Gerrard trout (Table 1, Supplementary Table 1). Fish from Gerrard and in the creel surveys were adult to subadult fish, whereas the samples from tributary streams were juveniles. All fish in the creel samples were measured (cm) and weighed (g) and scale samples were taken for aging. Fish were aged by counting annular rings following general approaches and methods (e.g., Sjolund 1974; MacLellan 1997).

Molecular analyses

Tissue samples were subject to DNA extraction using the Qiagen Animal Tissue extraction kit. The DNA was then subject to genotyping at eleven microsatellite loci: *Omy*77, *Ssa*85, *Ssa*197, *Ssa*456, *Ots*3, *Ots*4, *Ots*100, *Ots*107, *One*108, *Omm*1128, and *Omm*1130. Microsatellite loci were amplified using polymerase chain reaction (PCR) multiplexed assays in 10 µl volumes using the Qiagen Multiplex PCR kit with one primer for each locus labelled with a fluorescent dye. Labelled microsatellite alleles were size fractioned and detected using the Beckman-Coulter CEQ 8000 detection system (Taylor et al. 2007). Failed amplifications at any locus (less than 5% of the total) were re-run to minimize missing data. Given that we assayed fish using a series of multiplexes, re-running failed PCRs also allowed us to verify consistency of allele identifications across replicate PCRs for the same individual fish.

Statistical analyses

After verifying that there was no evidence of scoring errors, large allele dropout, or null alleles using MICRO-CHECKER (Van Oosterhout et al. 2004), the microsatellite data were examined for departures from Hardy–Weinberg or linkage disequilibrium locus-by-locus using ARLEQUIN (version 3.5.1.3). We then examined the degree of separation of all samples in "allelic space" using factorial correspondence analysis as implemented in GENETIX (version 4.0.5, Belkhir et al. 2004). Next, the genetic clustering program STRUCTURE version 2.3.3 (Falush et al. 2003) was used to determine the number of distinct genetic clusters (*K*) best represented by the total sample of trout. STRUCTURE uses a Bayesian clustering method to assign individuals to genetic clusters based on their genotypes. An individual may be assigned to more than one cluster if its genotype indicates admixture of two or more genetic groups. A Markov chain Monte Carlo method (MCMC) was used to estimate posterior probability distributions for each possible number of clusters for *K* values between 1 and 10. We did not set *K* to the maximum number of spatial or temporal samples obtained (N = 29), because as explained above, our focus was on differences between Gerrard and non-Gerrard rainbow trout, not an exhaustive analysis of population structure within the lake. The analysis used parameters that allowed admixture and correlated allele frequencies. Each run consisted of a 500,000 step burn-in followed by an additional 1,000,000 steps. Five iterations were run for each value of *K*. Given that there is some uncertainly about the most suitable method for determining what might be the most plausible

value of K (e.g. Janes et al. 2017), we used a variety of measures including calculating the posterior probability (see STRUCTURE documentation), ΔK following Evanno et al. (2005), and two new methods proposed by Verity and Nichols (2016): thermodynamic integration (TI) and the harmonic mean estimator ($\hat{h}_K$). Both of these latter metrics are direct measures of model evidence (K) given the data rather than the indirect heuristic measures of model evidence such as L_K and ΔK. We used the program MavericK (version 1.0.5) described by Verity and Nichols (2016) to calculate these various metrics. While the basic models in MavericK are identical to those in STRUCTURE, MavericK can only treat allele frequencies as being uncorrelated among sample sites. Hence, the MavericK simulations were run assuming uncorrelated allele frequencies, but with admixture among fish from different localities.

Genetic divergence between samples collected from the Lardeau River and other spawning streams was summarized by F_{ST} estimated using Weir and Cockerhams's (1984) θ calculated using GENETIX (with 5000 permutations). We employed the mixed-stock fishery analysis within ONCOR (Kalinowski et al. 2007) to apportion the fishery samples to one of two baseline groups: Gerrard (consisting of Gerrard spawning adults, the historical fishery samples, and the juvenile samples from the Lardeau River and its tributaries) and non-Gerrard rainbow trout (all other samples, Table 1). Since the early 1900s until 1987, about 8.7 million rainbow trout from populations outside Kootenay Lake have been used in population supplementation programs for Kootenay Lake (FFSBC 2018b). The single largest source of fish stocked originated from Pennask Lake (55%), tributary to the Fraser River, which were stocked between 1933 and 1948, mostly as eyed eggs or fry (FFSBC 2018b). Consequently, before conducting the Kootenay Lake only mixture analysis we verified that there was no detectable contribution from Pennask Lake rainbow trout by including 64 wild, Pennask Lake trout as a third group in the baseline in initial analyses. The robustness of the Kootenay Lake mixture analysis was assessed in several ways using ONCOR. First, 95% confidence intervals were generated by bootstrap resampling with replacement both for the mixture samples (across individuals) and the baseline samples (genotypes across alleles) during 5000 replicate analyses. Second, we produced simulated mixtures of genotypes (N = 200 fish each) where we fixed the proportional contribution of each population in turn to 1.0 and then estimated the mixture proportions for each baseline population contributing to the simulated mixtures ("100% mixtures"). In this case, "perfect" performance of the mixture analysis would return an estimated proportional contribution for each subpopulation of 1.0. Similar genotype resampling mixtures were constructed using the empirical mixture proportions ("Real fishery simulations") as described in the ONCOR documentation. We also conducted three-way error decomposition analyses to examine the extent of uncertainty in mixture proportion estimation from sampling only a limited number of fish in the fishery samples ("fishery error"), from genotyping a finite number of loci ("genotypic error"), and uncertainty in the estimation of baseline allele frequencies ("baseline error"). These errors are expressed as percentages of the total error in mixture estimation and error decomposition as outlined in the ONCOR documentation. Finally, ONCOR was used to assign individuals in the mixture samples to the baseline population that had the highest probability of producing the given genotype in the mixture.

We examined the growth patterns (size-at-age) of a subset of 496 fish assigned genetically as Gerrard or non-Gerrard and for which age determinations were made by constructing von Bertalanffy growth curves using the FSA and nlstools packages (Ogle 2013) in R (version 3.3.1, R Core Team 2018). Different models of growth that incorporated ecotype-specific terms for L_∞, K, and t_0 (i.e., Gerrard vs. non-Gerrard) were evaluated using hierarchical analyses-of-variance and the Akaike Information Criterion as outlined in Ogle (2013). Finally, a subsample of 384 Gerrard and 142 non-Gerrard rainbow trout assigned as such from the fishery mixtures was examined for diet composition. Here, the diet composition (by percentage of total wet weight of food) was determined for six groups of prey: kokanee, other fishes, *M. diluviana*, various zooplankton, terrestrial insects, and unknown. These groupings were selected to match as closely as possible those used by Northcote (1973) and Andrusak and Parkinson (1984).

Results

A total of 921 fish across 29 sample sites was assayed at eleven microsatellite DNA loci. In general, there was no evidence of null alleles or large allele dropout in our assays. Across a total of 1395 tests for pairwise linkage disequilibrium, 251 were significant at $P < 0.019$ (adjusted for 55 pairwise comparisons within sample localities), and 215 (86%) of these significant departures occurred in the fishery creel samples or the one mixed locality sample (Table 1). Further, of 275 tests for departures from Hardy–Weinberg equilibrium 63 were significant at $P < 0.017$ (adjusted for 11 tests per locality) and 49 (78%) were found in the mixture samples. Across all non-mixture samples, and with the exception of *One*108 which showed persistent heterozygote deficiencies, there were no loci that were consistently out of linkage or Hardy–Weinberg equilibria that would suggest there would be any issue with including all loci other than *One*108 in our analyses. The large fraction of departures from linkage and Hardy–Weinberg equilibria within the fishery creel samples would be

expected if these samples consisted of mixtures of genetically distinct populations (Table 1). Consequently, owing to the consistent departures from equilibria observed at *One*108 (20 of 24 samples with sufficient N tested showed deviations from HWE), it was deleted from subsequent analyses.

The factorial correspondence analysis revealed two well-separated groups of fish in allelic space (Supplementary Fig. 1). Subsequent STRUCTURE analysis suggested that the microsatellite data were best explained by the presence of two genetic populations; the maximum likelihood score peaked at $K=2$ (posterior probability = 1.0) and the second order rate of change of improvement in estimating K (ΔK) had a clear peak at $K=2$ (Supplementary Table 2). The alternative metrics of the harmonic mean estimator of K ($\hat{h}_K$) and the thermodynamic integration (TI) also produced posterior probabilities of 1.0 for $K=2$.

A bar plot of the proportional contribution of the two genetic groups to individual trout generated by STRUCTURE showed a marked difference in composition between Gerrard and non-Gerrard rainbow trout which tended to be characterized by the presence of one of the groups or the other, with few individual fish being of mixed ancestry (Fig. 3). The average (SD) ancestry coefficient (expressed as proportion Gerrard ancestry) for the Gerrard samples was 0.94 (0.05, N = 6) whereas the average for the non-Gerrards was 0.06 (0.03, N = 15). By contrast, the fishery creel samples tended to contain fish from both genetic groups (Fig. 3) and had an average ancestry coefficient of 0.74 (0.11, N = 9). The strong separation of Gerrard and non-Gerrard rainbow trout using in the STRUCTURE and mixed-stock fishery analysis was reflected in differences in allele frequencies at number of loci (Supplementary Fig. 2), and F_{ST} (θ) was 0.14 ($P<0.001$) between Gerrards and non-Gerrards, more than twice the average value between samples within these groups ($\theta=0.06$, $P<0.001$).

In preliminary analyses, proportional contributions from Pennask Lake fish to all the fishery samples was always 0.0 (upper 95% confidence intervals never exceeded 0.0) in pooled or in individual (by season and year) creel samples. Consequently, we conducted subsequent analyses using only the Gerrard and non-Gerrard samples in the baseline. The simulated fishery samples indicated a high degree of accuracy in the 100% mixture samples; the 100% Gerrard simulated fisheries were estimated to contain 99.9% Gerrards (95% confidence intervals of 99.8–100%) while the 100% non-Gerrard rainbow trout simulated fisheries were estimated to contain 100.0% non-Gerrard trout (100–100%). When all fishery creel samples from 2015 to 2017 were examined, ONCOR estimated the fishery to contain 73.4% (68.4–74.6%) Gerrards and 26.6% (23.4–31.6%) non-Gerrard trout. Conducting the analysis on the individual fishery samples showed similar results with the exception of sample 2015.1, which showed a more equal distribution of fish between Gerrard and non-Gerrard rainbow trout groups (Table 2). When these empirical values were used in the realistic fishery simulations there was a strong agreement between the two; the average simulated values for Gerrards was 73.4% (65.9–80.0%) and for non-Gerrards was 26.8% (20.0–34.1%). Three-way error decomposition indicated that when from 50 to 150 fish were in a creel mixture, > 98% of the error in estimating mixture proportions was associated with fishery error (i.e., the numbers of fish in the mixture being analyzed) and was identical for Gerrard and non-Gerrard rainbow trout. Consequently, genotype and baseline errors together accounted for < 2% of the mixture estimation error.

The ONCOR assignment test resulted in an average 98.5% (± 0.066%) assignment confidence; a total of 384 fish from the fishery samples were assigned to the Gerrard group (0.73) and 140 (0.27) to the non-Gerrard rainbow trout genetic group, and 524 of these fish had some

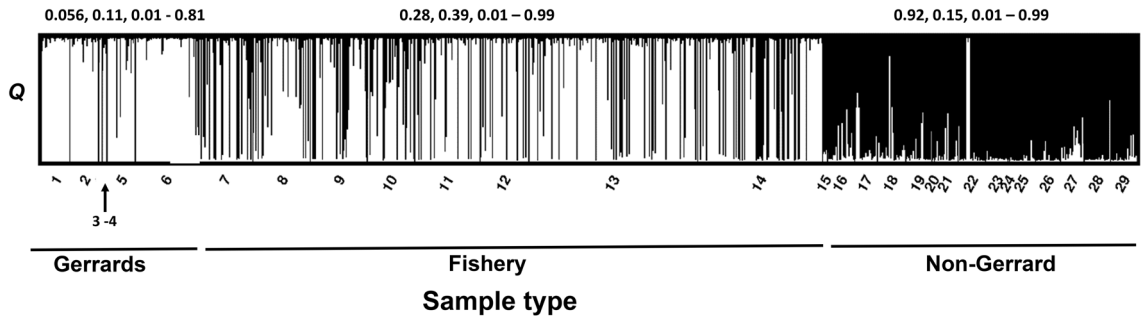

Fig. 3 Ancestry coefficients (Q, 0–1.0) for individual Kootenay Lake rainbow trout (*Oncorhynchus mykiss*) and assayed at ten microsatellite DNA loci. Each fish is represented by a thin vertical line where the height of each bar represents the proportional contribution of one genetic group (black) to the genome of that fish with the remaining portion representing the other (white) genetic group. The average, SD, and range of Q values (expressed in terms of the black genetic group) is given above each of the Gerrard, Fishery, and non-Gerrard sample groups. Sample codes are as in Table 1

Table 2 Results of ONCOR mixed stock fishery analysis for eight fishery samples of rainbow trout sampled from Kootenay Lake and assayed at ten microsatellite DNA loci

Sample	N	Proportion Gerrard	Proportion non-Gerrard
Fall Fishery 2015.1	48	0.5837 (0.391, 0.706)	0.4163 (0.294, 0.609)
Fall Fishery 2015.2	48	0.7718 (0.649, 0.896)	0.2282 (0.104, 0.351)
Fall Fishery 2015.3	48	0.6366 (0.495, 0.784)	0.3634 (0.216, 0.505)
Fall Fishery 2015.4	40	0.7329 (0.558, 0.856)	0.2671 (0.144, 0.442)
Spring Fishery 2016	41	0.7873 (0.639, 0.904)	0.2127 (0.096, 0.361)
Fall Fishery 2016	55	0.8482 (0.742, 0.927)	0.1518 (0.073, 0.258)
Spring Fishery 2017	142	0.7729 (0.696, 0.843)	0.2271 (0.157, 0.304)
Fall Fishery 2017	105	0.6481 (0.548, 0.730)	0.3519 (0.270, 0.452)
Total	527	0.7341 (0.684, 0.766)	0.2659 (0.234, 0.316)

Each proportion is the mean proportion of Gerrard and normal rainbow trout in the mixture and is estimated from 10,000 bootstrapped samples (confidence intervals in parentheses)

combinations of length, weight, and age data (Supplementary Table 1). The average length of the Gerrard-assigned fish from the fishery sample was greater than for the non-Gerrard fish in the fishery sample (mean (SD) = 38.9 (6.6, N = 384) cm vs. 37.6 (4.8, N = 140) cm, *t* test with unequal variances, P = 0.014), but the average weights were not significantly different from one another (means = 663.5 (273.9, N = 383) g vs. 650.2 (223.0, N = 140) g, respectively, *t*-test with unequal variances, P = 0.60). The proportion of Gerrard-assigned fish in the fishery sample, however, was higher both in the < 30 cm group and the > 45 cm group than for non-Gerrards in the fishery (Monte Carlo randomization test, P = 0.0012), but not within the 30–45 cm group. Similarly, the proportions of fish in the weight classes < 400 g, and > 1000 g was greater for Gerrard than non-Gerrard-assigned fish (P = 0.0141), but not within the 400 g to 1000 g group. Creel fish assigned as Gerrards were also older than those assigned as non-Gerrards (mean of 4.3 (1.3, N = 364) year vs. 4.0 (1.2, = 133) year, *t*-test with unequal variances, P = 0.030).

Fitting the length-at-age data for the fishery sample to a von Bertalanffy growth model indicated that a model employing ecotype-specific L_∞ (= 59.6 and 52.9 cm for Gerrards and non-Gerrards, respectively, both P < 0.001), and t_0 (= − 1.36 and − 2.58, respectively, both P < 0.05), but a common K (= 0.189, P < 0.001)) was the best model (AIC 2645.7), but this model was only 1.1 times more likely than a

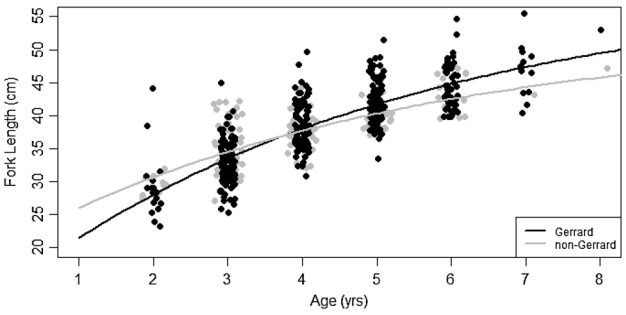

Fig. 4 Von Bertalanffy growth curve fit to size (fork length) at age (years) data for rainbow trout (*Oncorhynchus mykiss*) sampled from the recreational fishery for 2015 and 2016 in Kootenay Lake, BC. The line of best fit for Gerrard rainbow trout is is $L_t = L_\infty (= 59.6\,\text{cm})\,(1 - e^{-K(=0.189)(t - t_o(=-1.36))})$ and for non-Gerrard rainbow trout is $L_t = L_\infty (= 52.9\,\text{cm})\,(1 - e^{-K(=0.189)(t - t_o(=-2.58))})$

model with ecotype-specific L_∞ (61.3 and 48.0, respectively, both P < 0.001) and K (0.174 and 0.281, respectively, both P < 0.001), but a common t_0 (− 1.53, P < 0.01, AIC = 2645.8, Fig. 4, Supplementary Table 3). A model with all three parameters in common between ecotypes was 606 times less likely than the best model (Supplementary Table 3).

Of 384 fish from the fishery that were genetically assigned as Gerrards and for which diet data existed, 53 had at least one fish in their diet (and many from 2 to 5), but only 8 of the 137 non-Gerrards had fish in their diet (and none had more than one fish, contingency $\chi^2 = 6.41$, df = 1, P = 0.011). Across all sampled Gerrard trout, kokanee or other fishes comprised 32% (wet weight) of prey consumed compared to less than 5% for fish assigned as non-Gerrards. By contrast, the diets of Gerrards consisted of only 16% *Mysis diluviana* or various zooplankton compared to 42% in non-Gerrards. Both groups of fish exploited terrestrial insects at between 32–37% of the diet (Supplementary Table 4).

Discussion

Divergent life history types within salmonid species are common and may occur in sympatry (Behnke 1972; Taylor 1999). Such variation often takes the form of anadromous and non-anadromous forms that are sympatric at juvenile and adult spawning times (e.g., sockeye salmon and kokanee, respectively, Foote and Larkin 1988), or two or more ecotypes defined by distinct feeding habits and associated habitat use and morphology that are found together for their entire lives within single lake basins (e.g., seven ecotypes of Dolly Varden, *Salvelinus malma*, in Lake Kronoskoe, Kamchatka, Markevich et al. 2018). Indeed, the rainbow trout is no exception to this phenomenon, but most descriptions of sympatric life history types in this species focus on differentiation in migration phenotypes (e.g., anadromous steelhead

and non-anadromous rainbow trout or fluvial and adfluvial forms (Hecht et al. 2012; Arostegui and Quinn 2018).

Different trophic ecotypes of rainbow trout occurring in allopatry are well known including 'piscivorous' and 'insectivorous' types and the phenotypic differences between these forms are known to have a genetic basis (Keeley et al. 2005, 2007). Two growth forms of rainbow trout (smaller, slower-growing insectivores and larger, faster-growing piscivores) have been known from Kootenay Lake for some time and it has long been suspected that differences in diet drive much of the growth form differentiation (Cartwright 1961; Northcote 1973). Here, we have demonstrated that these two phenotypes of rainbow trout, large-bodied individuals that spawn in a particular location (Gerrard in the Lardeau River) and a smaller-bodied form that spawns in myriad smaller streams around the lake, are strongly differentiated genetically. While striking genetic differences have been recorded in other diet-differentiated sympatric salmonids (e.g., in brown trout, *Salmo trutta*, Ferguson and Taggart 1991; Arctic char, *Salvelinus alpinus,* Gíslason et al. 1999), other cases may show an almost complete lack of genetic/genomic differentiation (e.g., kokanee, Shedd et al. 2015; Limborg et al. 2018).

Both size groups of rainbow trout provide important contributions to a significant recreational fishery in Kootenay Lake (Andrusak and Andrusak 2012). The large-bodied "Gerrards" are the focus of a well-known trophy fishery where fish commonly exceed 5 kg and not uncommonly 10 kg. This fishery is a relatively specific one that occurs chiefly in the fall, winter, and spring using specialized trolling gear and guiding services. The fishery for the smaller-bodied trout occurs chiefly in the summer months and involves a wider variety of gear. Historically, the identification of Gerrard rainbow trout in the fishery was relatively straightforward given that these fish were very large (Northcote 1973). Consequently, harvest of the highly valued Gerrards could be closely monitored and regulated to promote conservation-based spawning escapements while sustaining the fishery. Such size-based differentiation, however, has become much more difficult given the reduction in size of fish in the creel surveys and the merging of body size between Gerrard and non-Gerrard rainbow trout. This phenomenon, coupled with observations of reduced spawning escapements at Gerrard in recent years (BC Ministry of Forests, Land and Natural Resource Operations and Rural Development, Nelson, BC, unpubl. data) has also led to concern that Gerrard rainbow trout were becoming less common in the fishery despite high catch rates of smaller-bodied rainbow trout. Our genetic data have provided a useful tool to distinguish Gerrards from non-Gerrards and demonstrate that Gerrards still predominate in the catch.

The high genetic differentiation between Gerrard and non-Gerrard rainbow trout is consistent with many occurrences of Hardy–Weinberg and linkage disequilibria detected in the fishery samples. Reductions from expected levels of heterozygosity will occur if the fishery samples consist of non-randomly interbreeding mixtures of genetically distinct populations which generates a Wahlund Effect (Allendorf et al. 2013). The same lack of interbreeding will also create non-random association of alleles across loci and linkage disequilibrium. Interestingly, the historical fishery sample did not show significant departure from Hardy–Weinberg equilbrium across any loci which is consistent with the STRUCTURE results suggesting that this sample (which was selected to include fish over 65 cm and, thus, likely to be Gerrards) was composed of a genetically more homogeneous population. The high degree of divergence observed between Gerrards and non-Gerrards is also consistent with preliminary results from single nucleotide polymorphism (SNP) analysis of a subset of our samples (N = 80 Gerrards and 80 non-Gerrards) assayed by poolseq whole genome resequencing (Schlötterer et al. 2014). Here, an F_{ST} over several million SNPs was substantial and averaged 0.093 (Grummer et al., University of BC, unpublished data), albeit lower than the value of 0.14 observed across the microsatellite DNA loci in this study.

The high level of divergence observed between Gerrards and non-Gerrards may be driven, in large part, by the high degree of spatial separation and physical differences between Gerrard and non-Gerrard spawning sites. The Gerrard site is at the outlet of a lake, almost 60 km upstream of Kootenay Lake, has deeper, faster water with larger substrates relative to the small stream habitats used by non-Gerrard trout (Hartman 1969). Consequently isolation-by-distance and isolation-by-adaptation may both promote high divergence between these ecotypes. Bond et al. (2014) presented evidence for similar phenomena promoting divergence (max $F_{ST} = 0.13$) among proximate populations of Dolly Varden (*Salvelinus malma*) inhabiting interconnected, but ecologically distinct spawning and rearing habitats in a southcentral Alaskan watershed. Further, some Gerrard trout may still reach large sizes and non-Gerrard size-at-maturity is typically less than 50 cm. Consequently, even if Gerrard and non-Gerrard trout used the same spawning streams, the large differences in size-at-maturity may minimize interbreeding (e.g., see Foote and Larkin 1988; Zimmerman and Reeves 2000).

Alternatively, it is possible that the high degree of historical stocking of insectivorous fish from Pennask Lake (and a few other areas) and biased introgression between these fish and non-Gerrard rainbow trout could contribute to the high level of divergence between Gerrards and non-Gerrards seen today. This hypothesis, however, seems unlikely for several reasons. First, our initial mixture analyses demonstrated no contribution of Pennask Lake genotypes to the fishery samples. Second, although a SNP-based F_{ST} estimate

between Pennask Lake and non-Gerrards is 0.153, a lower value than between Pennask and Gerrards (0.226), the pairwise F_{ST} between non-Gerrards and Blackwater River rainbow trout, also from the Fraser River drainage, but which have never been stocked into Kootenay Lake, is the lowest of all comparisons, 0.083 (Grummer et al., University of BC, unpublished data). Further, the estimated SNP F_{ST} between Blackwater River and Gerrards, 0.149, is very similar to, but lower, than that between Pennask Lake and non-Gerrards (Grummer et al., University of BC, unpubl. data). Had introgression from stocked fish been an important process and biased to that between Pennask and non-Gerrards, then we would have expected Pennask rainbow trout to be much more similar to non-Gerrards than fish from a population, Blackwater River, that have never been used in the Kootenay Lake supplementation program. The F_{ST} between Blackwater River and non-Gerrards was, however, almost 50% lower than that between Pennask and non-Gerrards. Certainly, that introgression between native and introduced populations may not always occur despite intensive stocking is not unprecedented in salmonid fishes (e.g., Morán et al. 1994; Santos et al. 2006; Taylor et al. 2007; White et al. 2018). Other populations once used in the Kootenay Lake stocking program were not available for analysis. Consequently, it is impossible to completely discount the possibility that biased introgression between some introduced fish and Gerrards or non-Gerrards has contributed to the high level of divergence observed today.

The relatively high level of divergence between Gerrards and non-Gerrards likely explains the accuracy of the mixture analyses and the high level of genetic assignment success, in addition to the relatively large baseline sizes (approximately 200 fish each), and the broad diversity of non-Gerrard rainbow trout localities sampled that are being compared to only a single other locality (Lardeau River Gerrard rainbow trout). Swatdipong et al. (2013) reported that a similar set of characteristics probably explained the high accuracy of mixture and assignment analyses in a recreational fishery for Finnish lake-run brown trout (*Salmo trutta*). The successful use of a relatively small number (ten) of microsatellite DNA loci in the Kootenay Lake fishery is consistent with similarly successful application of microsatellites in a local Atlantic salmon (*Salmo salar*) fishery in Northern Ireland (Ensing et al. 2013). As in our study, the success of identification was promoted by high baseline and fishery sample sizes even when differentiation was modest (max $F_{ST} = 0.039$) and relatively few loci ($N = 7$) were used.

The diet differences observed in fish that we had assigned genetically as Gerrards or non-Gerrards are very similar to relative differences observed by Northcote (1973) and Andrusak and Parkinson (1984) between 1929 and 1980. Expressed as percentage volume and frequency of occurrence, Northcote (1973) showed that rainbow trout over 45 cm in length (and presumably mostly Gerrards) had diets dominated by fish (typically 30–90%) and to a lesser extent *Mysis* and insects (typically 15–50%). By contrast, fish less than 45 cm ate mostly terrestrial insects (60–100%) and only rarely fish (0–25%). The consistent diet differences observed between Gerrards and non-Gerrards defined solely by size over decades and the observation of similar differences today in fish differentiated by genetics suggests that the same life history differences persist and has significant conservation implications.

First, our data support the idea that the fishery can be monitored for the proportional catch of Gerrards and non-Gerrard rainbow trout in the absence of marked size differences between the forms. Indices of abundance of Gerrards in the fishery catch will provide insight into subsequent years' spawning escapement levels by determining the age structure of the catch which may be used to adjust exploitation levels if necessary.

Second, genetic monitoring of the recreational fishery should be faster and more efficient with the development of genomic tools. For instance, a rainbow trout SNP-chip, similar to the approach of Palti et al. (2015), is being developed with diagnostic markers for various high value populations of rainbow trout in British Columbia (Schulte et al., University of BC, unpublished data) and a series of population mixture applications could be investigated simultaneously. Alternatively, we have developed a series of TaqMan assays (Applied Biosystems Inc.) based on our Kootenay Lake poolseq analyses where assaying even a single near diagnostic locus results in mixture proportions virtually identical to our current ten locus microsatellite assay (Taylor et al., unpubl. data.) making in-season mixture analyses achievable. Our mixture error decomposition results suggest that baseline and genotype error are insignificant. Making the reasonable assumption that similar (or perhaps greater) levels of genetic differentiation will be resolved using SNPs and a similar effort at baseline characterization as we conducted using microsatellites, error in mixture proportions will be almost exclusively related to the size of the sample in the fishery mixture being analyzed. Consequently, because we achieved these error distributions with a minimum fishery sample size of 40 fish, we recommend that no fewer than this number of trout be sampled for any creel sample to be characterized genetically.

Third, our analysis strongly suggests that the catch is still largely composed of Gerrard rainbow trout even though the average body size of the fish in the creel has declined. Interestingly, the predominance of Gerrards in the catch even though their size overlaps that of non-Gerrards much more than historically suggests that the piscivorous behaviour of Gerrard rainbow trout is still expressed when using the specific angling techniques (trolling fish-like flies or plugs in near surface waters) developed for these fish in Kootenay

Lake. These results and inference further support the idea that restoration efforts focussed on increasing the abundance of kokanee would probably result in the return to the characteristically large size of Gerrard trout (see Andrusak and Parkinson 1984; Redfish Consulting Ltd 2016). This management objective is supported by the continued importance of kokanee in the diets of the largest fish that were genetically identified as Gerrards and, therefore, supports the critical role that kokanee play in the Kootenay Lake rainbow trout fishery. Once kokanee abundance is restored, maintenance of this critical forage base could involve managing Gerrard spawning escapements if they exceed a target reference management number of 750 individuals (the estimated escapement at carrying capacity of the Lardeau River) by increasing angler harvest (Redfish Consulting Ltd 2016).

Finally, Lardeau River juvenile production has been relatively stable (based on surveys of age 0 + and 1 + juveniles in the spring within the river) between 2008 and 2015, yet estimated Gerrard spawning adult abundance increased from 514 to 1532 over the same time period (Andrusak 2017), which suggests that within lake survival has increased. The use of otolith microchemistry analyses on sport-caught fish and categorized genetically as Gerrards and non-Gerrards to identify the age at entry to the lake after river residence will be used to better understand recruitment dynamics and within-lake survival rates of the different ecotypes. These studies coupled with restoration of kokanee and monitoring the response in the fishery should contribute to a more complete understanding of within-lake predator prey interactions to better conserve ecotype biodiversity and the recreational resources they represent.

Acknowledgements Fishery samples were provided under contract to Reel Adventures, Nelson, BC. Funding for this project was arranged by Sue Pollard of the Freshwater Fisheries Society of British Columbia. Lab work was supported by equipment purchased with Natural Sciences and Engineering Research Council of Canada (NSERC) Discovery and Equipment grants awarded to EBT. The laboratory assistance of K. Higgins is greatly appreciated as are the helpful comments of reviewers and JC Garza.

References

Allendorf FW, Luikart G, Aitken SN (2013) Conservation and genetics of populations, 2nd edn. Wiley-Blackwell, Chichester

Andrusak G (2017) Determination of Gerrard rainbow trout productivity at low abundance. Report available from British Columbia Ministry of Forests, Land and Natural Resource Operations and Rural Development, Victoria, BC

Andrusak H, Parkinson EA (1984) Food habits of Gerrard stock rainbow trout in Kootenay Lake, British Columbia. British Columbia Fish and Wildlife Branch Tech Circular 60. Available from: British Columbia Ministry of Forests, Lands and Natural Resource Operations and Rural Development, Fish and Wildlife Branch, Victoria, BC

Andrusak H, Andrusak G (2012) Kootenay Lake Angler Creel Survey 2011. Redfish Consulting Ltd, Report prepared for fish and wildlife compensation program—Columbia Basin, Nelson. https://sites.google.com/site/redfishconsultingltd/system/app/pages/search?scope=search-site&q=reports. Accessed 25 Feb 2019

Arostegui MC, Quinn TP (2018) Trophic ecology of nonanadromous rainbow trout in a post-glacial lake system: partial convergence of adfluvial and fluvial forms. Can J Zool 96:818–827

Askey P, Bisson R (2016) A review of kokanee augmentation as a potential recovery action on Kootenay Lake. Report prepared for and available from British Columbia Ministry of Forests, Land and Natural Resource Operations and Rural Development, Fish and Wildlife Branch, Nelson, BC, p 29

Behnke RJ (1972) The systematics of salmonid fishes of recently glaciated lakes. J Fish Res Board Can 29:639–671

Behnke RJ (1992) Native Trout of Western North America Am Fish Soc Mono 6, Bethesda, Maryland

Belkhir K, Borsa P, Chikhi L, Goudet J, Bonhomme F (2004) GENETIX 4.05, Windows TM software for population genetics. Laboratoire génome, populations, interactions, CNRS UMR, 5000

Bond MH, Crane PA, Larson WA, Quinn TP (2014) Is isolation by adaptation driving genetic divergence among proximate Dolly Varden char populations? Ecol Evol 4:2515–2532

Bott K, Kornely GW, Donofrio MC, Elliott RF, Scribner KT (2009) Mixed-stock analysis of Lake Sturgeon in the Menominee River sport harvest and adjoining waters of Lake Michigan. N Am J Fish Mgmt 29:1636–1643

Bradbury IR, Hamilton LC, Rafferty S, Meerburg D, Poole R, Dempson JB, Robertson MJ, Reddin DG, Bourret V, Dionne M, Chaput G, Sheehan TF, King TL, Candy JR, Bernatchez L (2015) Genetic evidence of local exploitation of Atlantic salmon in a coastal subsistence fishery in the Northwest Atlantic. Can J Fish Aquat Sci 72:83–95

Brenden TO, Scribner KT, Bence JR, Tsehaye I, Kanefsky J, Vandergoot CS, Fielder DG (2015) Contributions of Lake Erie and Lake St. Clair walleye populations to the Saginaw Bay, Lake Huron, recreational fishery: evidence from genetic stock identification. N Am J Fish Mgmt 35:567–577

Cartwright JW (1961) Investigations of the rainbow trout of Kootenay Lake, British Columbia, with special reference to the Lardeau River. Br Col Fish Wildl Branch Fish Manag Publ 7:1–46

Chistiakov DA, Hellemans B, Volckaert FA (2006) Microsatellites and their genomic distribution, evolution, function and applications: a review with special reference to fish genetics. Aquaculture 255:1–29

Cooke SJ, Murchie KJ (2015) Status of aboriginal, commercial and recreational inland fisheries in North America: past, present and future. Fish Manag Ecol 22:1–13

Ensing D, Crozier WW, Boylan P, O'Maoiléidigh N, McGinnity P (2013) An analysis of genetic stock identification on a small geographical scale using microsatellite markers, and its application in the management of a mixed-stock fishery for Atlantic salmon *Salmo salar* in Ireland. J Fish Biol 82:2080–2094

Evanno G, Regnaut S, Goudet J (2005) Detecting the number of clusters of individuals using the software SRUCTURE: a simulation study. Mol Ecol 14:2611–2620

Falush D, Stephens M, Pritchard JK (2003) Inference of population structure using multilocus genotype data: linked loci and correlated allele frequencies. Genetics 164:1567–1587

Ferguson A, Taggart JB (1991) Genetic differentiation among the sympatric brown trout (*Salmo trutta*) populations of Lough Melvin, Ireland. Biol J Linn Soc 43:221–237

FFSBC (2013) 2013 BC freshwater sport fishing economic impact report. Freshwater Fisheries Society Report. https://www.gofishbc.com/. Accessed 28 Nov 2018

FFSBC (2018a) Freshwater Fisheries Society of British Columbia Annual Report 2017/2018. https://www.gofishbc.com/PDFs/Footer/2018-Annual-Report.aspx. Accessed 28 Nov 2018

FFSBC (2018b) Provincial stocking records. Freshwater Fisheries Society of BC. https://www.gofishbc.com/Stocked-Fish.aspx#fish-stocking. Accessed 12 Oct 2018

Foote CJ, Larkin PA (1988) The role of male choice in the assortative mating of anadromous and non-anadromous sockeye salmon (*Oncorhynchus nerka*). Behaviour 106:43–61

Gíslason D, Ferguson M, Skúlason S, Snorrason SS (1999) Rapid and coupled phenotypic and genetic divergence in Icelandic Arctic char (*Salvelinus alpinus*). Can J Fish Aquat Sci 56:2229–2234

Hartman GF (1969) Reproductive ecology of the Gerrard stock of rainbow trout. In: Northcote TG (ed) Symposium on salmon and trout in streams. HR MacMillian Lectures in Fisheries, University of British Columbia, Vancouver, pp 53–67

Hecht BC, Thrower FP, Hale MC, Miller MR, Nichols KM (2012) Genetic architecture of migration-related traits in rainbow and steelhead trout, *Oncorhynchus mykiss*. G3 2:1113–1127

Irvine JR (1978) The Gerrard rainbow trout of British Columbia—a discussion of their life history with management, research, and enhancement recommendations. BC Hydro Power Authority Fish Manag Rep 72:1–62

Janes JK, Miller JM, Dupuis JR, Malenfant RM, Gorrell JC, Cullingham CI, Andrew RL (2017) The K = 2 conundrum. Mol Ecol 26:3594–3602

Kalinowski ST, Manlove KR, Taper ML (2007) ONCOR: a computer program for genetic stock identification. Department of Ecology, Montana State University, Bozeman MT, 59717. http://www.montana.edu/kalinowski/software/oncor.html. Accessed 10 Jan 2018

Keeley ER, Parkinson EA, Taylor EB (2005) Ecotypic differentiation of native rainbow trout (*Oncorhynchus mykiss*) populations from British Columbia. Can J Fish Aquat Sci 62:1523–1539

Keeley ER, Parkinson EA, Taylor EB (2007) The origins of ecotypic variation of rainbow trout: a test of environmental vs. genetically based differences in morphology. J Evol Biol 20:725–736

Kurota H, McAllister MK, Parkinson EA, Johnston NT (2016) Evaluating the influence of predator–prey interactions on stock assessment and management reference points for a large lake ecosystem. Can J Fish Aquat Sci 73:1372–1388

Levey JJB, Williams R (2003) 2000 survey of sport fishing in British Columbia with summary information from the 1985, 1990, and 1995 surveys. BC Fish and Wildlife Recreation Allocation Branch, BC Ministry of Water, Land and Air Protection, Victoria

Limborg MT, Larson WA, Shedd K, Seeb LW, Seeb JE (2018) Novel RAD sequence data reveal a lack of genomic divergence between dietary ecotypes in a landlocked salmonid population. Conserv Genet Res 10:169–171

Markevich G, Esin E, Anisimova L (2018) Basic description and some notes on the evolution of seven sympatric morphs of Dolly Varden *Salvelinus malma* from the Lake Kronotskoe Basin. Ecol Evol 8:2554–2567

McCusker MR, Parkinson E, Taylor EB (2000) Mitochondrial DNA variation in rainbow trout (*Oncorhynchus mykiss*) across its native range: testing biogeographical hypotheses and their relevance to conservation. Mol Ecol 9:2089–2108

McLennan SE (1997) Guide for Sampling Structures used in Age Determination of Pacific Salmon. Fisheries and Oceans Canada. Stock Assessment Division. Pacific Biological Station. Nanaimo

Morán P, Pendás AM, Garcia-Vázquez E, Izquierdo JT, Rutherford DT (1994) Electrophoretic assessment of the contribution of transplanted Scottish Atlantic salmon (*Salmo salar*) to the Esva River (Northern Spain). Can J Fish Aquat Sci 51:248–252

Northcote TG (1973) Some impacts of man on Kootenay Lake and its salmonids. Great Lakes Fish Commun Tech Rep 25:1–46

Ogle DH (2013) FishR Vignette-von Bertalanffy growth models. Ashland: Northland College, 54. http://derekogle.com/fishR/examples/oldFishRVignettes/VonBertalanffy.pdf. Accessed 22 Mar 2018

Palti Y, Gao G, Liu S, Kent MP, Lien S, Miller MR, Rexroad CE, Moen T (2015) The development and characterization of a 57 K single nucleotide polymorphism array for rainbow trout. Mol Ecol Res 15:662–672

Pitcher TJ, Hollingsworth CE (eds) (2002) Recreational fisheries: ecological, economic and social evaluation. Blackwell Science, Oxford

R Core Team (2018) R: A language for environment and statistical computing. R Foundation for Statistical Computing. Vienna. http://www.R-project.org/

Redfish Lake Consulting Ltd (2016) Kootenay Lake Action Plan-2016. Prepared for the Ministry of Forests, Land and Natural Resource Operations and Rural Development, Nelson, BC, p 35. Available from Redfish Consulting Ltd., Nelson

Santos NP, Fontaínhas-Fernandes AA, Faria R, Torres-Castro LF, Anjos MR, Cortes RM, Alexandrino P (2006) Genetic evidence for limited introgression between wild and stocked individuals in Portuguese brown trout, *Salmo trutta* populations. Folia Zool 55:433–443

Schlötterer C, Tobler R, Kofler R, Nolte V (2014) Sequencing pools of individuals—mining genome-wide polymorphism data without big funding. Nat Rev Genet 15:749–763

Shedd KR, von Hippel FA, Willacker JJ, Hamon TR, Schlei OL, Wenburg JK, Miller JL, Pavey SA (2015) Ecological release leads to novel ontogenetic diet shift in kokanee (*Oncorhynchus nerka*). Can J Fish Aquat Sci 72:1718–1730

Sjolund WR (1974) Collection and preparation of scales, Otoliths and Fin rays for fish age determination. Fisheries technical circular no. 12 B.C Fish and Wildlife Branch

Sparrow RAH, Larkin PA, Rutherglen RA (1964) Successful introduction of *Mysis relicta* Lovén into Kootenay Lake, British Columbia. J Fish Res Board Can 21:1325–1327

Swatdipong A, Vasemägi A, Niva T, Koljonen ML, Primmer CR (2013) Genetic mixed-stock analysis of lake-run brown trout *Salmo trutta* fishery catches in the Inari Basin, northern Finland: implications for conservation and management. J Fish Biol 83:598–617

Tamkee P, Parkinson E, Taylor EB (2010) The influence of Wisconsinan glaciation and contemporary stream hydrology on microsatellite DNA variation in rainbow trout (*Oncorhynchus mykiss*). Can J Fish Aquat Sci 67:919–935

Taylor EB (1999) Species pairs of north temperate freshwater fishes: evolution, taxonomy, and conservation. Rev Fish Biol Fish 9:299–324

Taylor EB, Tamkee P, Sterling G, Hughson W (2007) Microsatellite DNA analysis of rainbow trout (*Oncorhynchus mykiss*) from western Alberta, Canada: native status and evolutionary distinctiveness of "Athabasca" rainbow trout. Conserv Genet 8:1–15

Taylor EB, Darveau C-A, Schulte PM (2013) Setting conservation priorities in a widespread species: phylogeographic and physiological variation in the lake chub, *Couesius plumbeus* (Pisces: Cyprinidae). Diversity 5:149–165

Tufts BL, Holden J, DeMille M (2015) Benefits arising from sustainable use of North America's fishery resources: economic and conservation impacts of recreational angling. Int J Environ Stud 72:850–868

Van Oosterhout C, Hutchinson WF, Wills DP, Shipley P (2004) MICRO-CHECKER: software for identifying and correcting genotyping errors in microsatellite data. Mol Ecol Notes 4:535–538

Verity R, Nichols RA (2016) Estimating the number of subpopulations (K) in structured populations. Genetics 203:1827–1839

Weir BS, Cockerham CC (1984) Estimating F-statistics for the analysis of population structure. Evolution 38:1358–1370

White SL, Miller WL, Dowell SA, Bartron ML, Wagner T (2018) Limited hatchery introgression into wild brook trout (*Salvelinus fontinalis*) populations despite reoccurring stocking. Evol Appl 11:1567–1581

Zimmerman CE, Reeves GH (2000) Population structure of sympatric anadromous and nonanadromous *Oncorhynchus mykiss*: evidence from spawning surveys and otolith microchemistry. Can J Fish Aquat Sci 57:2152–2162