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Genetic Diversity of Dolly Varden Populations in Norton and Kotzebue Sounds

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Genetic Diversity of Dolly Varden Populations in Norton and Kotzebue Sounds

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Abstract

We describe the genetic stock structure of Dolly Varden *Salvelinus malma* in Norton and Kotzebue Sounds to develop a method to quantify contributions of Dolly Varden stocks harvested in subsistence fisheries in the Wulik River. We surveyed genetic variation at seven microsatellite loci in 12 population samples from Norton and Kotzebue Sounds and four additional populations from the North Slope and the Togiak River. Significant differences in allele frequencies were detected among all pairwise combinations of populations and evidence of a closer genetic similarity was detected between Dolly Varden in the Imuruk Basin and Norton Sound, despite the size distribution and life history similarities Imuruk Basin Dolly Varden share with Kotzebue Sound Dolly Varden. Mean contribution estimates from analysis of simulated mixtures from individual tributaries ranged from 82% to 98%, suggesting that robust stock composition estimates for regional groups, and in some cases, individual tributaries can be obtained. Dolly Varden were sampled from the Wulik River subsistence fishery in October 2001. Two hundred individuals were randomly selected from the fishery samples with size information and Bayesian Markov chain Monte Carlo estimates of regional stock proportions were made: North Slope 0.0369 (SD 0.0357), Kotzebue Sound 0.6726 (SD 0.0597), Cobblestone River 0.0035 (SD 0.0097), Norton Sound 0.2785 (SD 0.0571), and Togiak River 0.0085 (SD 0.015). The majority of Dolly Varden used in the mixture analysis was less than 325mm FL, suggesting that at least some Dolly Varden are migrating into the Chukchi Sea basin during their first or second seaward migrations.

Introduction

Anadromous Dolly Varden *Salvelinus malma* overwintering in the Wulik River are the most important subsistence resource for the residents of Kivalina, comprising over 90% of the subsistence catch (Burch 1985). Dolly Varden are harvested in subsistence fisheries during the fall migration into freshwater and during the spring migration into saltwater. Harvests estimated through annual household surveys (ADFG 2001) or counts of catches (Bernard and DeCicco 1987) are generally between 15,000 and 25,000 (DeCicco 2001a) and have ranged from 7,436 fish in 1986 to 64,970 in 1969. Aerial surveys of the Wulik River are flown annually in the fall to monitor abundance of the overwintering population of Dolly Varden (DeCicco 2001a); counts have ranged from 30,932 in 1984 to 297,257 in 1969. Mark-recapture experiments have estimated overwintering populations at 76,892 (SE=16,811) for fish greater than 400mm FL in 1988/1989 (DeCicco 1990) and 361,599 (SE=62,306) in 1994/1995 (DeCicco 1996).

Dolly Varden home to their natal streams to spawn, but tagging studies have shown that overwintering fish are from multiple river systems (DeCicco 1997; Armstrong and Morrow

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1980). For example, Dolly Varden tagged while overwintering in the Wulik River have been recovered in the Pilgrim, Kobuk, Noatak, Wulik, and Kivalina Rivers (DeCicco 1996). Tag recoveries have also been made on the Seward Peninsula, on Saint Lawrence Island, and in Russia, demonstrating the long distance movements of northwestern Dolly Varden (DeCicco 1997). However, the relative proportions of contributing populations to the overwintering aggregates of Dolly Varden in the Wulik River remain unknown. A method to identify the origin of Dolly Varden harvested in subsistence fisheries in the Wulik River is needed to assess the impact of harvests on Dolly Varden populations.

Genetic differences have been detected both on large and small spatial and evolutionary time scales in Dolly Varden. In North America, two forms of Dolly Varden have been described based on genetic (Phillips et al. 1999), morphological, meristic, and behavioral traits (Morrow 1980; Armstrong and Morrow 1980; Behnke 1980). The northern form, *S. m. malma*, is distributed from the Mackenzie River to the north side of the Alaska Peninsula, while the southern form, *S. m. lordi*, is distributed from the south side of the Alaska Peninsula to Puget Sound. Northern form Dolly Varden also occur in the upper Susitna drainage (Morrow 1980), and their southern distribution has recently been noted to include the south side of the Alaska Peninsula (Behnke 2002). Genetic differences have been detected both between (e.g., Reist et al. 1997; Leder 2001) and within forms (e.g., southern form: Griswold 2002; Redenbach and Taylor 2000; northern form: Everett et al. 1997; Crane et al. 2003). Genetic data have also been used in mixed-stock analysis (MSA) to estimate the origin of Dolly Varden sampled from mixtures in the Beaufort Sea and to document transboundary movements of U.S. and Canadian origin Dolly Varden along the North Slope (Everett et al. 1997; Krueger et al. 1999).

Anadromous Dolly Varden spawning in rivers of Norton and Kotzebue Sounds show variation in spawning timing and other life history attributes. Most Kotzebue Sound fish spawn in summer (DeCicco 1989; DeCicco and Reist 1999). Summer spawners may overwinter in their natal stream, move upstream to spawning areas in summer, and move back downstream to overwintering areas until the following spring when they migrate to sea; or may overwinter in non-natal streams, migrate out to sea in the spring and move directly to their natal streams to spawn. Some Kotzebue Sound fish and most Norton Sound fish are fall spawners, entering freshwater in the fall, typically spawning in headwater springs, and remaining near spawning areas to overwinter (DeCicco 1997).

Further, DeCicco and Reist (1999) described two geographic groups of Dolly Varden in western Alaska, based on size and life history information. Those spawning in the Imuruk Basin and in Chukchi and Beaufort Sea drainages reach larger sizes than populations to the south in Norton Sound. In addition, movement patterns differ between fish from Norton and Kotzebue Sounds. In Norton Sound, they tend to closely follow the movement patterns of salmon. Dolly Varden may delay migration into saltwater in the spring to allow feeding on salmon fry, and may enter freshwater multiple times in the summer to feed on salmon eggs. Reist and DeCicco (1999) speculated that these forms may have evolved during periods of allopatry during periodic rising of the Bering Land Bridge and may warrant division of northern form Dolly Varden into two forms: the nominal, northern form, distributed in Alaska from the Imuruk Basin to the McKenzie River, and a Bering Sea or Beringian form, distributed from the Alaska Peninsula to Norton Sound. Alternatively, size differences may be due to an increasing latitudinal trend in Dolly Varden to food availability (Maekawa and Nakano 2002) and movement patterns may reflect relatively recent adaptation to available resources. Gudkov (1999) suggested that the latitudinal

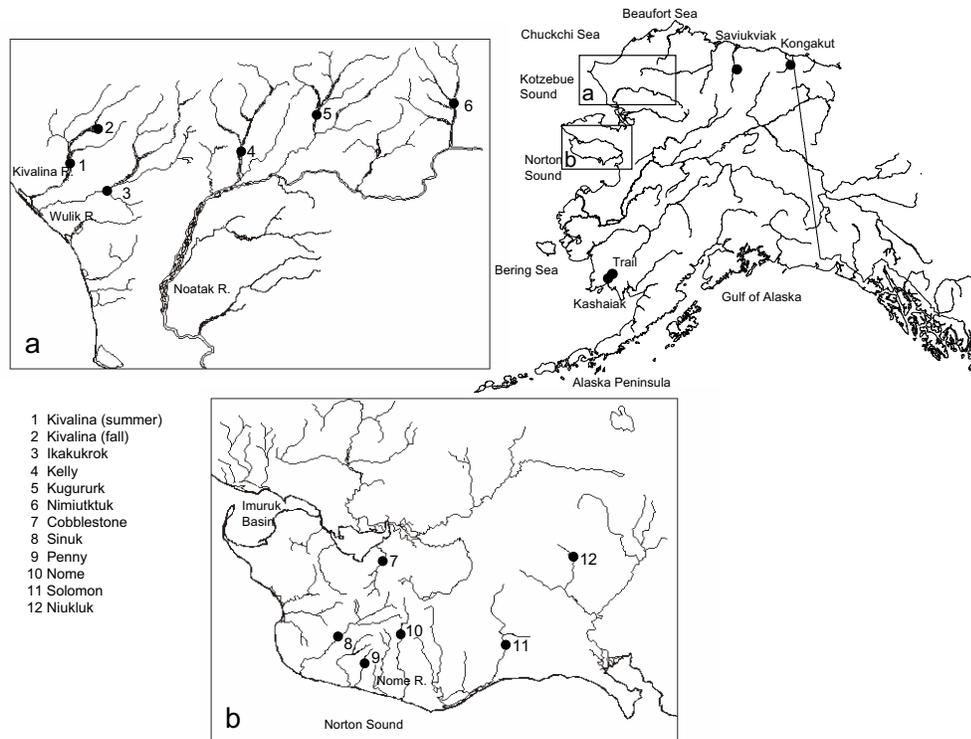


Figure 1. Sampling locations of Dolly Varden collected in northwestern Alaska and surveyed for genetic variation at microsatellite loci.

life history and size variation in Bering and Chukchi Sea Dolly Varden are related to smolt age and number of seaward migrations prior to maturity, and that the populations share population dynamic characteristics.

In this project, we collected genetic data on 12 populations of Dolly Varden from Norton Sound and Kotzebue Sound. Using variation at seven microsatellite loci, we describe the genetic population structure of Dolly Varden in northwestern Alaska, and test if there is a transition in genetic diversity patterns at the Imuruk Basin corresponding to changes in size distributions and life history patterns. We also test the applicability of genetic data for use in MSA to estimate the origin of Dolly Varden sampled from subsistence catches in the Wulik River in fall 2001.

Methods

Sample Collection

Fin tissue was collected from Dolly Varden at a total of 12 locations in Norton and Kotzebue Sounds (Figure 1, Table 1). Fin tissue was taken from prespawning adults, recently emerged young-of-the-year, and juveniles. Prespawning adults were collected by hook and line at or near spawning areas. Gender was determined by examination of external characteristics and fork length was measured to the nearest mm. Young-of-the-year and juvenile fish were captured using baited minnow traps or small dip nets. Possible family effects of sampling juveniles (Allendorf and Phelps 1981) were minimized by setting minnow traps at multiple locations at sampling sites and by collecting juveniles from several locations over the area used for rearing within a given stream. Fin clips, or whole fish, in the case of recently emerged young-of-the-year, were stored in individually labeled vials with 90% ethanol.

Table 1. Collection location, sample size, life history stage, spawning timing, and reference for size distribution data of Dolly Varden sampled for genetic analysis in northwestern Alaska.

Location	N	Collection Dates	Life Stage	Spawning Timing
Beaufort Sea				
Kongakut ^a	72	2000	adult	summer
Saviukviak ^a	55	2001	adult	summer
Kotzebue Sound				
(1) Kivalina ^b	200	July 14-15, 2000	juvenile	summer
(1) Kivalina ^c	20	July 23-25, 2001	adult	summer
(2) Kivalina ^c	200	June 19-20, 2001	juvenile	fall
(3) Ikalukrok ^b	147	Aug.-Sept. 2000	adult	summer
(4) Kelly ^c	200	July 10, 2001	juvenile	summer
(4) Kelly ^c	67	July 11-15, 2001	adult	summer
(5) Kugururok ^b	100	July 7-10, 2000	adult	summer
(6) Nimiuktuk ^b	160	July 21, 2000	juvenile	summer
Imuruk Basin				
(7) Cobblestone ^c	40	July 28-30, 2001	adult	summer
(7) Cobblestone ^c	298	July 28-30, 2001	juvenile	summer
Norton Sound				
(8) Sinuk ^c	99	July 22-23, 2001	juvenile	summer
(8) Sinuk ^b	40	Sept.-Oct. 2000	juvenile	summer
(9) Penny ^b	200	Sept.-Oct. 2000	juvenile	summer
(10) Nome ^b	150	Sept.-Oct. 2000	juvenile, adult	summer
(11) Solomon ^b	200	October 2000	juvenile	summer
(12) Niukluk ^c	200	July 17-20, 2001	juvenile	summer
Bristol Bay				
Kashaiak ^d	51	1998, 2000	adult	summer
Trail ^d	116	1998, 2000	adult	summer

^aADFG unpublished

^bDeCicco 2001b

^cDeCicco 2002

^dLisac and Nelle 2000

We included adult samples from the Kongakut and Saviukviak Rivers on the North Slope, and from the Kashaik River and Trail Creek in Bristol Bay (tributaries to the Togiak River) for comparison at a larger spatial scale (Table 1, Figure 1).

Fin clips were also taken from fish in the mixed-stock subsistence fishery in the Wulik River at Kivalina in October 2001. Subsistence fishers in Kivalina were visited after they had completed their fall fishing, and after receiving permission, fish were measured and fin clips were taken. The catches of three subsistence fishers were inspected and sampled. A total of 576 tissue samples were obtained along with 319 fork length measurements. Further information on sampling locations, methods, and gender and size of fish sampled can be found in DeCicco (2001b) and DeCicco (2002).

Laboratory Analysis

Total genomic DNA was isolated from fin tissue using a quick lysis procedure. Individuals were assayed for genetic variation at seven microsatellite loci developed for Dolly Varden: *Sma-3*, *-5*, *-10*, *-17*, *-21*, *-22*, *-24* (Crane et al. 2004). PCR amplification of microsatellite loci was carried out in 10 μ l reaction volumes: approximately 100ng DNA, 1.5 mM MgCl₂, 8mM dNTPs, 0.5 U *Taq* DNA polymerase (Promega), 0.4 μ M unlabeled/labeled forward primer, and 0.4 μ M reverse primer, using an MJResearch™ DNA Engine™ PCT-200. Cycling conditions were 2 min at 92°; 30 cycles of 15 sec at 92°, 15 sec at T_a, and 30 sec at 72°; with a final extension for 10 min at 72°. Annealing temperatures (T_a) were 55° for *Sma-5*, *-10*, and *-22*; 56° for *Sma-17* and *-21*; and 58° for *Sma-3*. Microsatellites were separated on 64-well denaturing polyacrylamide gels and visualized and scored using a Li-Cor IR²® scanner with Li-Cor Saga™ GT ver 2.0 software (Lincoln, NE). Li-Cor 50-350bp or 50-500bp size standards were loaded in the first and last lanes and at intervals of 14 lanes or less across each gel. Positive controls, consisting of 2-10 alleles of predetermined size, were loaded in three lanes distributed evenly across the gels to ensure consistency of allele scores. Two researchers scored alleles independently. Samples with score discrepancies between researchers were re-amplified at the loci in question and rescored.

Statistical Analysis

Population Analysis -- Unless otherwise noted, all calculations were made using FSTAT version 2.9.3 (Goudet 2001). Allele frequencies were calculated for each collection. Homogeneity of allele frequencies between samples collected in the same tributary in different years were compared using likelihood ratio statistics and the significance of the likelihood ratio statistic was evaluated using a randomization test through 4,620 permutations of genotypes between samples. Samples were pooled if no significant differences were detected after adjusting for four multiple tests using the sequential Bonferroni technique (Rice 1989).

Observed and expected heterozygosities and allele richness (number alleles observed per locus corrected for sample size) were used to describe within-sample diversity. For each locus in each collection, deviation of genotypic frequencies from Hardy-Weinberg expectation was evaluated by testing for a deficit of heterozygotes, $F_{IS} > 0$. Significance was assessed by 2,240 permutations of alleles among individuals within samples; *P*-values over all loci were calculated according to Goudet (2001). Conformation to Hardy-Weinberg expectation was evaluated by adjusting the table-wide $\alpha = 0.05$ for 16 multiple tests using the sequential Bonferroni technique (Rice 1989). Homogeneity of allele frequencies among collections, including the Kivalina summer and fall spawning collections to determine if gene flow is restricted due to timing of spawning, were

tested using likelihood ratio statistics, and the significance was evaluated using a randomization test through 2,400 permutations of genotypes between samples. We adjusted P -values for the four multiple comparisons using the sequential Bonferroni technique.

Cavalli-Sforza & Edwards (CSE; 1967) chord distances were calculated from allele frequencies between all pairwise combinations using GENDIST in Phylip version 3.57c (Felsenstein 1995). Genetic similarity among collections was visualized using multidimensional scaling in S-Plus 6.0 (Insightful, Inc.; Seattle, WA), where genetic distances were plotted in three dimensions such that the observed distances matched as closely as possible the distances in multidimensional space.

We computed F_{ST} to quantify the amount of population subdivision following the method of Weir and Cockheram (1984). A randomization-based test was used to determine if F_{ST} was greater than zero with the null distribution created through 1,000 permutations of genotypes among populations.

Randomization tests were used to test for differences in expected heterozygosity, allele richness, and F_{ST} between Norton Sound and Kotzebue Sound populations. If genetic differences evolved during isolation due to lowered sea levels and the exposure of the land bridge, population size reductions may have led to a corresponding decrease of within-population variability. In addition, F_{ST} may be greater in the population samples from Kotzebue Sound because of the greater size and complexity of the river systems in this region, and the presence of summer and fall spawning populations.

We used two methods to determine whether genetic structure corresponded more with life history variation (i.e., Cobblestone River grouped with Kotzebue Sound populations) or geographic proximity (i.e., Cobblestone River grouped with Norton Sound populations). For the first method, we used pairwise CSE chord distances to build a tree that minimizes the sum of squared differences between the observed distances and the branch lengths in the tree (Fitch and Margoliash 1967) using the FITCH routine with the global rearrangement and randomization of taxon input order options in Phylip version 3.57c (Felsenstein 1995). Stability of the tree nodes, specifically the node linking Cobblestone River with either Norton or Kotzebue Sound populations, was assessed by 100 bootstrap resamples over loci using the SEQBOOT and CONSENSE routines of Phylip. We also used analysis of molecular variation for diploid data (AMOVA, Michalakis and Excoffier 1996) to evaluate structuring of genetic variation using ARLEQUIN version 2.0 (Schneider et al. 2000). Two population hierarchies were used, one grouping Cobblestone River with Norton Sound (geographic proximity), and one grouping Cobblestone River with Kotzebue Sound (life history). For each analysis, genetic variation was partitioned into between region, within within-region, and within population components. Randomization tests were used to evaluate the significance of the variance components.

Mixed Stock Analysis -- Maximum likelihood estimation of artificial mixtures was used to determine population groupings of Dolly Varden from the Bering Sea region that can be identified in mixtures. Prior to the simulation analysis, alleles in the baseline populations were binned using the program OptiBin (Bromaghin and Crane In press) to reduce the effects of sampling error and rare alleles. For each locus, exact tests of homogeneity were used to test if allele pairs were similarly distributed across populations, with Monte Carlo simulation to estimate significance, to determine the binning strategy. Log-likelihood ratios were used as the test statistic and the binning procedure executed until $P < 0.25$.

We used the program SPAM version 3.7 (Debevec et al. 2000; Alaska Department of Fish and Game, Gene Conservation Laboratory, <http://www.cf.adfg.state.ak.us/geninfo/research/genetics/software>) for the simulation analysis. For each population, 1,000 artificial mixtures of 200 genotypes were randomly constructed using Hardy-Weinberg expectations from the baseline allele frequencies. Conditional maximum likelihood estimates and 90% symmetric confidence intervals of the mixture compositions were estimated for the mixtures using randomly resampled baseline allele frequencies to account for sampling error in the baseline; a Bayesian estimate of baseline allele proportions (Rannala and Mountain 1997) was used to further reduce the effects of sampling error in the baseline allele frequencies. Mean contribution estimate for the population under study should approximate 100% and estimates of about 90% are generally considered robust for mixture analysis (Kondzela et al. 2002; Teel et al. 1999).

Two hundred Dolly Varden were randomly subsampled from subsistence catch samples with corresponding size information (N=317). Stock composition estimates for these 200 fish were computed using a Bayesian procedure as implemented in BAYES (Pella and Masuda 2001). This approach was used over conditional maximum likelihood because some populations likely to be contributing to mixtures sampled from the Wulik River are missing from the baseline. Conditional maximum likelihood assumes that there are no stocks missing from the baseline. BAYES uses information in the stock mixture and genetic similarities among the baseline samples to update the baseline, providing more accurate estimates for regional groups (Pella and Masuda 2001). For this analysis, we used unbinned baseline information because we did not want to lose information from rare alleles present in the mixture that could be used to update the baseline.

Markov chain Monte Carlo samples of stock proportions were generated for five regional groups: North Slope, Kotzebue Sound, Cobblestone River, Norton Sound, and Togiak River. The initial proportion for the chain (N=5,000) for a given regional group was 95%, with the 5% distributed evenly among the remaining four groups. Values for genetic prior parameters were determined as described in Pella and Masuda (2001). The Rafferty and Lewis (1996) diagnostic was used to verify that chain lengths were sufficiently long. Convergence was determined using the Gelman and Rubin (1992) diagnostic. The mean, standard deviation, and posterior quantiles of sample stock composition estimates were generated after a burn-in of 2,500 samples.

Results

Population Subdivision

All seven loci were polymorphic in all collections with the exception of Sma-5, which was polymorphic in only two collections from Norton Sound (Appendix 1). The number of alleles observed at each variable locus ranged from 2 to 44 (Appendix 1). After adjusting for four multiple tests, no allele frequency heterogeneity was detected between collections made in different years from the Sinuk and Kelly Rivers (2000 and 2001; $P=0.1773$ and 0.0478 , respectively) or between the Cobblestone River adult and juvenile collections ($P=0.1262$). Therefore, duplicate collections at these sites were pooled for subsequent analyses. Significant allele frequency heterogeneity was detected for the two samples of summer spawners in the Kivalina River in 2000 and 2001 ($P=0.002$). The juvenile sample was taken from near the mouth of Grayling Creek, and spawners were taken in the mainstem up- and downstream from Grayling Creek. However, because of the small sample size of the 2001 collection, we also pooled these two collections for further analysis.

Mean expected heterozygosities ranged from 0.604 in the Saviukviak River to 0.712 in the Nome River and averaged 0.941 over all loci (Appendix 1). Allele richness ranged from 9.42 in Kivalina River fall population to 13.04 in the Nome River (Appendix 1). When adjusted for multiple tests, one significant deviation of genotypic frequencies from Hardy-Weinberg expectation was detected for *Sma-10** (Kongakut River, $P=0.0004$), and four were detected locus *Sma-24** (Kongakut River, Kivalina River summer, Ikalukrok River, and Kelly River $P=0.004$). When tested over all loci, three collections had significant deviations from Hardy-Weinberg expectations: Kongakut River ($P=0.0004$), Kivalina River summer ($P=0.0031$), and Kashaik River ($P=0.0027$). Allele frequency differences were detected among all population pairs in tests of homogeneity of allele frequencies, including between Kivalina summer and fall spawning populations ($P < 0.00042$).

Multidimensional scaling showed that geographically proximate populations were the most genetically similar. Populations from northwestern Alaska were spatially arranged in two groups, one composed of populations from the Kivalina, Wulik, and Noatak Rivers, and one composed of populations from Cobblestone River south and east to the Niukluk River. Within the Kotzebue Sound group, summer and fall spawning populations from the Kivalina River were genetically distinct from the other populations (Figure 2). Within the Noatak River, the Nimiuktuk River (the farthest upstream population sampled) was genetically distinct from the Kugururok and Kelly rivers.

The global F_{ST} among all populations was 0.036 ($P<0.0001$). We detected significant regional differences in expected heterozygosities ($P=0.0046$) but no differences in allele richness ($P=0.1526$). There were no significant differences in regional F_{ST} ($P=0.9582$).

Both methods used to evaluate correspondence of genetic structure with life history variation (Cobblestone River grouped with Kotzebue Sound populations) or alternatively, geographic proximity (Cobblestone River grouped with Norton Sound populations), provided evidence for the latter. Cobblestone River groups with Norton Sound populations in 87% of trees derived from bootstrap resampling of loci (Figure 3). For both hierarchies, AMOVA showed that most (>95%) of the total genetic variation was within populations. For the hierarchy based on geographic proximity, the remaining variation was equally divided into a between region component (1.95%) and a within region component (1.81%). In the life history hierarchy, less variation was due to the among region component (1.06%) than the within region component (2.34%).

Mixed Stock Analysis

In simulation experiments, mean contribution estimates for mixtures composed of artificial genotypes from the individual tributaries were less than 90% for four of the 16 populations tested: Kashaik River (88%), Nome River (84%), Kugururok River (82%), and Ikalukrok River (88%); most of the misallocation for these populations was to adjacent tributaries (Figure 4). When Kashaik, Nome, and Kugurok rivers were grouped with adjacent tributaries, mean contribution estimates exceeded or were very close to 90%: Kashaik/Trail River 94%, Nome/Solomon 89%, and Kelly/Kuguruok River 91%.

Mean FL of 200 Dolly Varden randomly sampled for stock composition estimates was 344 mm (range: 245-829 mm) (Figure 5). Markov chain Monte Carlo estimates of stock contributions indicated that approximately 95% of the subsistence samples from the Wulik River were from

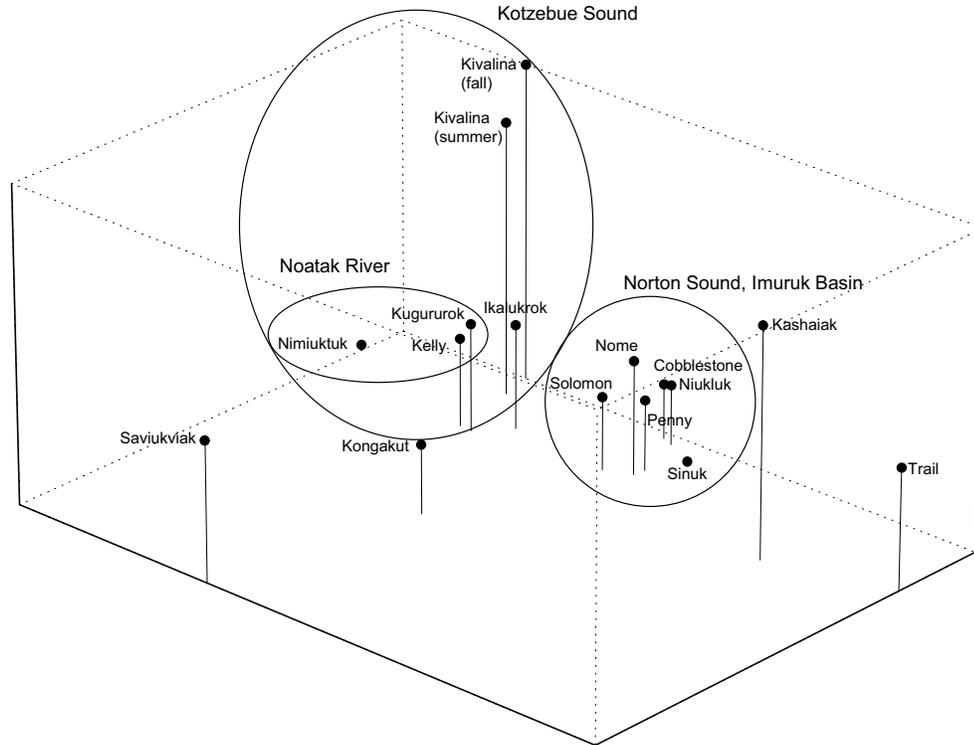


Figure 2. Multidimensional scaling of pairwise genetic distances between Dolly Varden populations in northwestern Alaska.

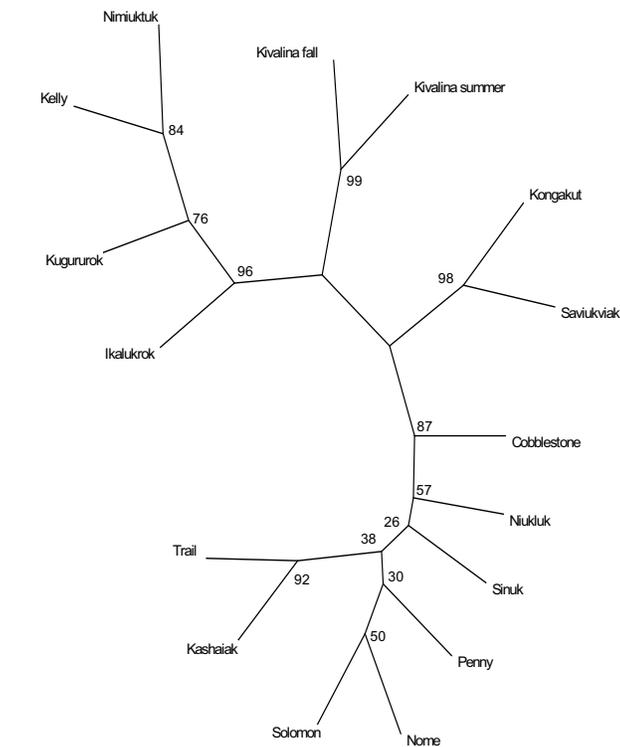


Figure 3. Consensus tree of Fitch and Margoliash trees derived from genetic distances calculated from 100 bootstrap resamples over loci. Numbers indicate the number of bootstrap resamples that supported the node.

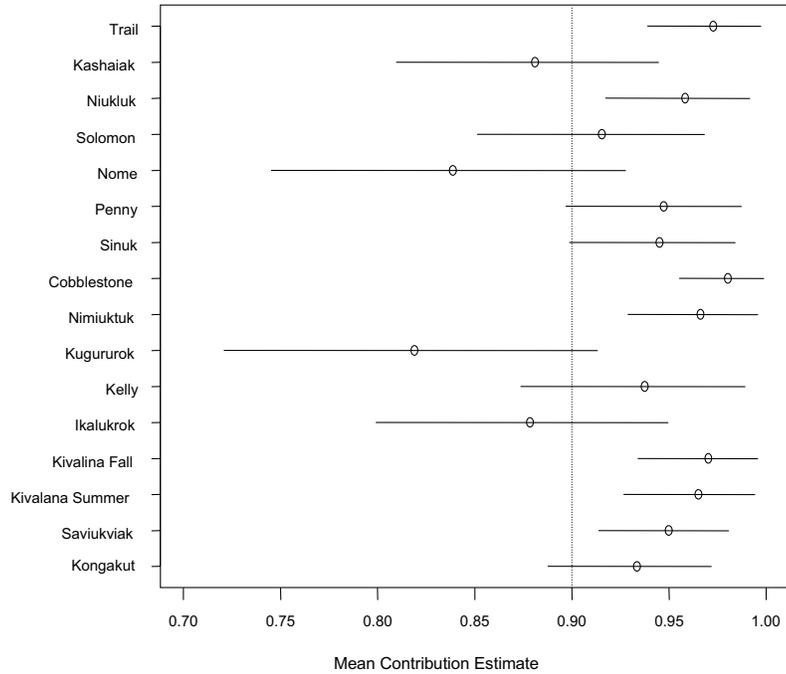


Figure 4. Mean conditional maximum likelihood estimates and 90% symmetric confidence intervals of stock contributions to mixtures comprised of randomly generated genotypes from individual populations of Dolly Varden in northwestern Alaska. Mean estimates should equal 100%. Mean estimates of approximately 90% (vertical line) are generally considered robust for mixture analysis.

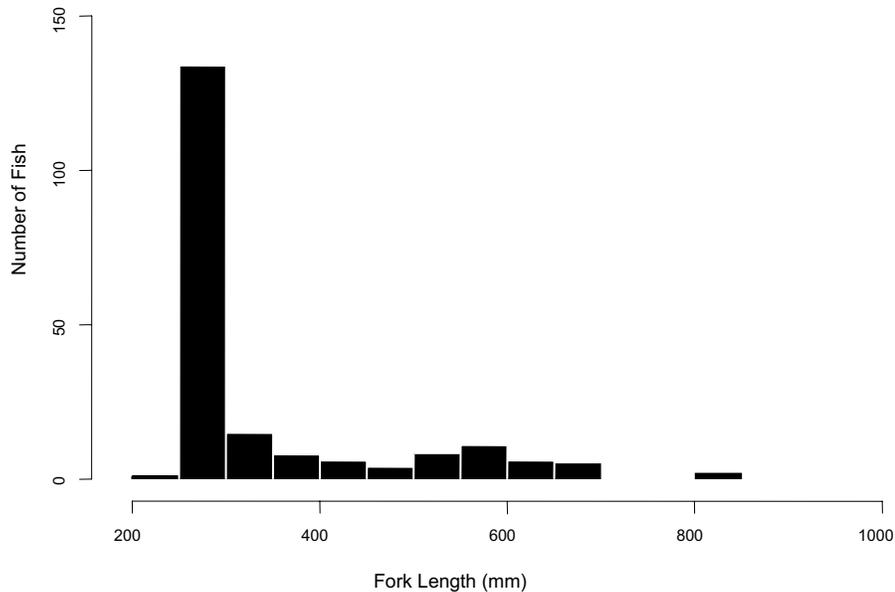


Figure 5. Histogram of fork lengths (mm) of Dolly Varden sampled from the Wulik River subsistence fishery in fall 2001.

Kotzebue Sound (0.6726, SD 0.0597) and Norton Sound (0.2785, SD 0.0571) (Table 3). The remainder was from the North Slope (0.0369, SD 0.0357), Cobblestone River 0.0035 (SD 0.0097), and Togiak River 0.0085 (SD 0.015).

Table 3. Markov chain Monte Carlo means, standard deviations, and quantiles for regional stock proportions of Dolly Varden sampled from the Wulik River subsistence catches in 2001.

Region	Mean	S.D.	Posterior Quantiles		
			2.50%	Median	97.50%
North Slope	0.0369	0.0357	0.0000	0.0298	0.1200
Kotzebue Sound	0.6726	0.0597	0.5529	0.6729	0.7857
Cobblestone River	0.0035	0.0097	0.0000	0.0000	0.0340
Norton Sound	0.2785	0.0571	0.1714	0.2772	0.3939
Togiak	0.0085	0.0150	0.0000	0.0010	0.0533

Discussion

Population Subdivision

Two sources of error may have been introduced into this analysis by including samples of juvenile Dolly Varden. First, non-random sampling may have occurred if family groups were collected, inflating the chance of significant differences between populations (Allendorf and Phelps 1981). Second, multiple spawning aggregates may have been sampled, or samples may not be representative of spawning streams if juveniles migrate significant distances. We believe the effects of family sampling were minimized by collecting samples at multiple sites within tributaries. Further, no significant differences were detected among multiple collections made in the same tributary, indicating that allele frequency estimates were not biased because of non random sampling. We also believe juvenile samples were representative of tributaries where they were collected. Movement among river systems does not occur until smoltification at ages 2-5. Within the Noatak River, the Nimiuktuk River was represented by juvenile samples, and this was the farthest upstream collection, minimizing the chances of mixing between tributaries. The Kelly River was represented by both juvenile and adult collections, but no allele frequency differences were detected between collections. However, further sampling of summer run spawning aggregates in the Kivalina River is warranted because Hardy-Weinberg conformation statistics indicate that multiple spawning aggregates may have been sampled.

Population subdivision in northwestern Alaska Dolly Varden is similar to the pattern observed by Everett et al. (1997) for North Slope drainages. Using allozyme data, they found highly significant allele frequency differences among 11 North Slope drainages, and significant heterogeneity within three of seven drainages in which multiple tributaries were sampled, suggesting that population subdivision is occurring on a relatively fine scale. Similarly, in this study, significant allele frequency differences were detected among all tributaries sampled, and within the Kivalina and Noatak Rivers. These studies suggest that natal homing by Dolly Varden is fairly precise, even though migratory behavior is complex. Similar genetic structuring of river system within region has also been documented for anadromous brook trout *S. fontinalis* (Castric and Bernatchez 2003) and Arctic char *S. alpinus* (Bernatchez et al. 1998).

Analyses based on genetic distance (MDS, Fitch tree) and F_{ST} (AMOVA) indicate a closer

genetic similarity between Norton Sound and Imuruk Basin Dolly Varden, though Imuruk Basin Dolly Varden share life history attributes with Kotzebue Sound Dolly Varden. This suggests that natural selection is maintaining life history differences in Imuruk Basin Dolly Varden despite greater gene flow and/or more recent ancestry with Norton Sound Dolly Varden.

Phenotypic diversity and genetic diversity at neutral loci can both be influenced by biogeographical events. During the Wisconsin Glaciation ~18,000 ya, drainages on the north and south sides of the Seward Peninsula and Kotzebue Sound were the headwaters of the ancient Chukchi River draining into the Chukchi Sea (Lindsey and McPhail 1986). This would suggest that present day populations in Norton and Kotzebue Sounds should be part of the same genetic lineage and that life history variation has evolved within about the last 18,000 years. Alternatively, present day populations in Norton Sound may be derived from a secondary invasion of Dolly Varden from the south. In this case, Dolly Varden in Norton Sound would share a closer genetic relationship with populations in drainages to the south than with populations in Kotzebue Sound. Additional sampling of Dolly Varden in Bering Sea drainages south of Norton Sound would allow further examination of these alternative scenarios, and provide a better understanding of the influence of biogeography on observed life history variation.

Mixed Stock Analysis

The level and pattern of genetic differences detected among populations surveyed in this study provide a powerful and cost effective method for estimating relative contributions of populations, or groups of populations, to overwintering aggregations in the Wulik River. The simulation analysis demonstrates that the contributions of individual river systems may be estimated in most cases. Krueger et al. (1999) were also able to estimate stock proportions of Dolly Varden sampled from the Beaufort Sea for individual river systems on the North Slope, as well as for individual tributaries in the Sagavanirktok River.

We estimated regional stock contributions to the mixture sample from the Wulik River rather than river system contributions because our baseline is not exhaustive. Populations in this study appear to be structured geographically therefore contributions of populations missing from the baseline are likely to be misallocated to geographically proximate populations (Shaklee and Phelps 1990). However, representative samples from all potential geographic regions contributing to Wulik River mixtures are not included here. Potential bias in mixture estimates using the present baseline may be the exclusion of populations from Asia. Overwintering Dolly Varden tagged in the Wulik River have been recaptured in the Anadyr River in Russia, suggesting that Dolly Varden may not always overwinter in their continent of origin (DeCicco 1992). Ideally, population samples from the Chukotsk Peninsula should be obtained.

The histogram of fork lengths of Dolly Varden sampled from the 2001 Wulik River subsistence fishery indicates that most individuals sampled were less than 325mm. These Dolly Varden are likely returning to freshwater after their first or second seaward migration (DeCicco 1996) and may be less likely to have traveled as extensively as individuals in larger size classes that have not previously spawned. Larger Dolly Varden that have already spawned once are also less likely to travel long distances because they have only one season at sea before they will return to their natal stream to spawn again. The mixture composition was 67% Kotzebue Sound fish and 28% Norton Sound fish, providing some indication that Norton Sound Dolly Varden are migrating into the Chukchi Sea during their first or second seaward migration.

Recommendations

Dolly Varden overwintering in the Wulik River are an important subsistence resource for the residents of Kivalina. While no immediate conservation concerns have been identified for this fishery resource, residents of Kivalina are concerned about contamination of fish and the potential for fish kills from pollution from Red Dog Mine, a zinc mine located upstream of the overwintering areas (Susan Georgette, ADFG-Subsistence Division, Kotzebue, personal communication; DeCicco 2001). Also, global warming may have profound effects upon Dolly Varden, including changes in marine movements patterns resulting from alterations in ocean productivity and fish metabolism, and shifts in the location and usage of overwintering areas resulting from alterations in stream discharge and freeze cycles. We recommend collection of additional baseline data for Dolly Varden, particularly from populations in the Russian Far East, the Noatak River, and the Imuruk Basin. MSA using genetic data provide an accurate and cost effective method for estimating the stock composition of Dolly Varden aggregations in overwintering areas and fishery harvests. MSA can be used to obtain stock-specific information to assess impacts of subsistence fishing, and habitat change due to pollution events or global warming.

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Appendix 1. Allele frequencies, allele richness (A_R), expected heterozygosity (H_e), observed heterozygosity (H_o), and P -values for conformation to Hardy-Weinberg expectations (P-HW) for Dolly Varden collections from northwestern Alaska.

Locus	Population Sample															
	Kong.	Savi.	Kiv. (s)	Kiv. (f)	Ikal.	Kell.	Kugu.	Nimi.	Cobb.	Sinuk	Penny	Nome	Solo.	Niuk.	Kash.	Trail
<i>Sma-10</i>																
N	71	51	186	187	142	266	99	151	325	137	188	145	196	198	50	114
140	0.028	—	—	0.003	0.004	—	—	—	0.005	0.015	0.021	0.014	0.003	0.005	—	—
144	—	—	—	—	0.018	—	—	—	0.002	0.026	0.032	0.010	0.008	0.030	—	0.004
148	—	0.010	—	—	—	—	—	—	0.008	0.055	—	0.014	0.005	—	0.020	0.092
152	0.035	—	0.027	0.019	0.018	—	0.005	0.023	0.035	0.029	0.005	0.062	0.026	0.098	0.060	0.066
156	—	—	—	—	0.004	—	—	—	0.012	0.004	0.019	0.052	0.015	0.061	0.260	0.145
160	0.035	—	0.062	0.061	0.042	0.023	0.040	—	0.045	0.109	0.069	0.090	0.084	0.068	0.150	0.154
164	0.085	—	0.008	0.075	0.025	0.051	0.020	0.007	0.018	0.022	0.024	0.010	0.015	0.063	0.030	0.070
168	0.035	0.020	0.067	0.086	0.173	0.164	0.162	0.103	0.083	0.241	0.213	0.121	0.117	0.063	0.080	0.096
172	0.359	0.539	0.210	0.168	0.246	0.267	0.247	0.212	0.409	0.128	0.141	0.134	0.158	0.129	0.160	0.088
176	0.049	0.108	0.083	0.136	0.116	0.085	0.106	0.185	0.163	0.186	0.136	0.097	0.122	0.091	0.050	0.026
180	0.021	0.020	0.062	0.091	0.011	0.013	0.015	0.013	0.018	0.004	0.043	0.059	0.015	—	0.040	0.096
184	—	—	0.003	—	0.014	0.008	0.005	—	0.002	0.026	0.043	0.028	0.005	0.056	0.070	0.057
188	0.070	0.098	0.011	—	0.014	0.013	0.035	0.063	0.038	0.062	0.051	0.076	0.077	0.040	0.010	0.022
192	0.092	0.010	0.387	0.273	0.165	0.235	0.212	0.255	0.065	0.047	0.122	0.134	0.194	0.197	—	—
196	0.092	—	0.016	0.005	0.067	0.038	0.015	0.043	0.049	0.015	0.019	0.014	0.094	0.066	0.010	—
200	—	0.010	0.024	0.072	0.007	0.008	0.005	0.043	0.006	0.026	0.029	0.021	0.038	0.025	—	—
204	—	—	0.005	—	—	0.013	0.010	0.007	0.002	—	0.011	0.003	0.005	—	0.010	—
208	0.007	0.020	0.011	0.005	0.032	0.041	0.035	0.026	—	—	0.005	0.017	—	—	0.010	—
212	—	0.029	—	—	—	0.023	0.035	0.013	0.002	0.004	0.019	0.007	—	0.008	0.010	0.048
216	0.007	0.059	—	—	0.004	0.006	0.020	—	—	—	—	—	0.010	—	—	—
220	0.028	0.010	—	—	0.032	0.004	0.010	—	—	—	—	0.031	0.005	—	—	—
224	0.049	0.059	0.003	—	—	0.002	0.020	—	—	—	—	—	—	—	0.010	—
228	—	0.010	0.011	—	0.004	0.009	—	0.007	—	0.004	—	—	0.003	—	—	0.009
232	0.007	—	—	—	0.007	—	—	—	—	—	—	—	—	—	0.020	—

Locus	Population Sample															
	Kong.	Savi.	Kiv. (s)	Kiv. (f)	Ikal.	Kell.	Kugu.	Nimi.	Cobb.	Sinuk.	Penny	Nome	Solo.	Niuk.	Kash.	Trail
236	—	—	0.011	0.005	—	—	—	—	0.002	—	—	0.007	—	—	—	0.004
240	—	—	—	—	—	—	—	—	0.003	—	—	—	—	—	—	0.013
244	—	—	—	—	—	—	—	—	0.034	—	—	—	—	—	—	0.004
252	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	0.004
A _R	14.97	13.70	13.07	10.48	15.45	13.74	15.42	12.05	13.77	14.80	15.97	17.96	14.84	13.88	16.76	15.06
H _c	0.839	0.686	0.787	0.850	0.862	0.834	0.853	0.839	0.787	0.869	0.889	0.916	0.886	0.902	0.873	0.909
H _o	0.662	0.588	0.806	0.866	0.789	0.823	0.828	0.815	0.778	0.905	0.872	0.931	0.908	0.874	0.780	0.842
P-HW	0.000	0.027	0.800	0.782	0.011	0.334	0.271	0.237	0.349	0.936	0.268	0.778	0.879	0.105	0.046	0.010
<i>Sma-17</i>																
N	68	53	174	186	127	263	100	151	319	134	185	147	185	197	49	112
81	—	—	—	—	0.004	—	—	—	—	—	—	—	—	—	—	—
99	—	—	0.011	0.022	0.012	0.025	0.005	0.026	—	—	—	—	—	—	—	—
101	—	—	—	—	—	—	—	—	—	—	—	—	—	—	0.010	—
105	0.007	—	—	—	—	—	—	—	0.011	0.004	—	0.007	0.016	0.030	0.020	0.080
107	0.029	—	—	—	—	0.008	—	0.017	0.020	0.030	0.016	0.017	0.003	0.030	—	—
109	—	—	—	—	—	—	—	—	—	0.011	0.003	0.010	0.005	—	0.041	0.018
111	—	0.057	0.032	0.024	0.004	0.004	0.005	0.007	0.014	0.063	0.024	0.037	0.014	0.005	0.041	0.085
113	0.007	0.038	0.049	0.011	—	0.006	0.020	—	0.002	0.041	0.038	0.031	0.054	0.058	0.031	0.018
115	0.235	0.255	0.043	0.054	0.024	0.030	0.040	0.017	0.069	0.075	0.116	0.102	0.081	0.071	0.092	0.143
117	0.022	0.019	0.037	0.083	0.059	0.097	0.125	0.020	0.177	0.119	0.089	0.071	0.057	0.129	0.061	0.022
119	—	—	—	—	0.028	0.010	0.035	—	0.020	0.007	0.024	0.010	0.005	—	0.020	0.107
121	0.022	0.038	0.069	0.091	0.122	0.093	0.145	0.007	0.119	0.090	0.130	0.146	0.108	0.079	0.031	0.067
123	0.044	0.066	—	0.003	0.016	0.023	0.025	0.046	0.003	0.067	0.038	0.041	0.032	0.015	—	0.018
125	0.044	0.113	0.101	0.113	0.043	0.048	0.050	0.040	0.011	0.015	0.051	0.068	0.084	0.076	0.082	0.058
127	0.566	0.340	0.511	0.532	0.516	0.490	0.490	0.513	0.444	0.295	0.332	0.333	0.297	0.363	0.408	0.214
129	0.022	0.038	0.095	0.065	0.165	0.144	0.060	0.308	0.105	0.142	0.108	0.112	0.203	0.135	0.112	0.121
131	—	0.019	0.040	0.003	0.008	0.019	—	—	0.005	0.041	0.027	0.014	0.035	—	0.051	0.040

Locus	Population Sample															
	Kong.	Savi.	Kiv. (s)	Kiv. (f)	Ikal.	Kell.	Kugu.	Nimi.	Cobb.	Sinuk.	Penny	Nome	Solo.	Niuk.	Kash.	Trail
133	—	—	0.011	—	—	—	—	—	—	—	0.003	—	0.005	0.008	—	—
135	—	—	—	—	—	0.004	—	—	—	—	—	—	—	—	—	0.004
139	—	0.009	—	—	—	—	—	—	—	—	—	—	—	—	—	—
141	—	0.009	—	—	—	—	—	—	—	—	—	—	—	—	—	0.004
A _R	9.33	11.80	10.39	9.06	9.89	10.98	9.84	8.62	9.72	12.48	12.15	12.55	12.19	10.74	12.98	13.48
H _e	0.622	0.803	0.708	0.682	0.687	0.717	0.717	0.638	0.742	0.855	0.834	0.833	0.838	0.813	0.803	0.887
H _o	0.618	0.698	0.701	0.683	0.709	0.681	0.700	0.669	0.724	0.851	0.789	0.830	0.870	0.792	0.694	0.911
P-HW	0.512	0.030	0.428	0.550	0.788	0.052	0.368	0.856	0.212	0.467	0.042	0.508	0.922	0.223	0.021	0.821
<i>Sma-21</i>																
N	68	54	189	184	130	262	100	151	317	132	186	146	184	195	50	112
105	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	0.009
109	—	—	—	—	—	—	—	—	—	—	0.003	0.010	0.005	—	—	—
115	0.404	0.481	0.397	0.299	0.469	0.613	0.470	0.573	0.233	0.178	0.204	0.247	0.293	0.169	0.130	0.085
117	0.022	0.019	—	0.003	0.008	0.017	0.005	—	0.011	0.042	0.065	0.041	0.073	0.015	0.170	0.161
123	—	—	—	—	0.004	0.004	—	—	—	—	—	—	—	—	0.040	—
125	—	—	—	0.003	—	—	—	—	0.006	—	0.003	—	—	—	—	—
127	0.316	0.250	0.262	0.236	0.177	0.113	0.145	0.046	0.374	0.295	0.325	0.315	0.337	0.390	0.520	0.607
129	0.022	0.028	—	—	—	0.002	0.005	—	—	—	—	—	—	0.003	—	—
131	0.022	0.009	0.008	—	0.073	0.044	0.055	0.003	0.021	0.049	0.046	0.045	0.043	0.028	0.050	0.031
133	0.118	0.065	0.151	0.139	0.127	0.086	0.195	0.156	0.120	0.178	0.196	0.120	0.068	0.164	0.060	0.045
135	0.066	—	0.095	0.087	0.081	0.063	0.080	0.152	0.203	0.186	0.078	0.116	0.109	0.159	0.010	—
137	—	0.037	0.066	0.092	0.031	0.019	0.020	0.040	0.006	0.027	0.048	0.062	0.052	0.041	—	—
139	—	—	0.003	—	0.015	0.002	—	—	0.024	0.045	0.030	0.038	0.016	0.021	—	—
141	0.007	—	—	—	—	0.023	0.015	0.026	0.002	—	0.003	0.007	—	0.010	—	0.036
143	—	—	—	—	—	0.002	—	—	—	—	—	—	—	—	—	0.022
145	—	—	0.019	0.141	0.008	0.004	0.010	0.003	—	—	—	—	—	—	0.020	—
147	—	—	—	—	—	0.002	—	—	—	—	—	—	0.003	—	—	—

Locus	Population Sample															
	Kong.	Savi.	Kiv. (s)	Kiv. (f)	Ikal.	Kell.	Kugu.	Nimi.	Cobb.	Sinuk.	Penny	Nome	Solo.	Niuk.	Kash.	Trail
149	—	—	—	—	0.008	0.008	—	—	—	—	—	—	—	—	—	0.004
151	0.022	0.074	—	—	—	—	—	—	—	—	—	—	—	—	—	—
153	—	0.037	—	—	—	—	—	—	—	—	—	—	—	—	—	—
A _R	8.61	8.88	6.71	6.52	9.00	9.57	8.48	6.58	7.60	7.95	8.73	9.23	8.54	8.59	7.96	8.01
H _e	0.721	0.698	0.739	0.802	0.722	0.599	0.713	0.622	0.750	0.811	0.801	0.806	0.776	0.766	0.683	0.596
H _o	0.735	0.741	0.725	0.826	0.715	0.607	0.720	0.636	0.729	0.780	0.801	0.774	0.799	0.815	0.660	0.652
P-HW	0.651	0.833	0.355	0.831	0.477	0.705	0.624	0.703	0.194	0.211	0.534	0.167	0.805	0.978	0.402	0.974
<i>Sma-22</i>																
N	71	54	179	190	138	265	97	150	327	136	188	146	198	198	51	109
148	—	—	—	0.008	—	—	—	0.003	—	—	—	—	—	—	—	—
152	—	—	0.006	—	0.007	0.011	0.010	0.097	—	—	—	—	—	—	—	—
156	—	—	—	—	0.007	—	0.005	0.013	—	—	—	—	0.005	—	—	—
160	—	—	0.017	—	0.022	0.019	0.010	0.013	0.002	—	—	0.010	—	—	—	0.005
164	—	—	—	—	0.007	0.008	0.021	0.007	0.057	0.018	0.003	0.014	0.005	—	—	0.023
168	0.007	—	—	—	0.007	0.030	0.005	0.013	0.024	0.007	0.064	0.027	0.025	0.035	0.010	0.023
172	—	—	—	—	0.025	0.023	0.010	0.037	0.005	0.018	0.013	0.017	0.005	0.030	0.020	—
176	0.021	—	0.025	0.024	0.025	0.026	0.077	0.027	0.005	0.007	0.029	0.031	0.018	0.008	0.020	0.069
180	0.014	—	0.073	0.032	0.018	0.032	0.026	0.047	0.012	0.004	0.035	0.062	0.033	0.020	0.010	0.032
184	0.063	0.028	0.179	0.071	0.083	0.042	0.077	0.030	0.037	0.051	0.059	0.045	0.023	0.045	0.069	0.050
188	0.049	0.065	0.050	0.050	0.101	0.070	0.139	0.087	0.070	0.015	0.085	0.068	0.106	0.033	0.059	0.055
192	0.085	0.148	0.039	0.079	0.138	0.085	0.134	0.107	0.055	0.092	0.093	0.140	0.081	0.053	0.196	0.046
196	0.141	0.139	0.109	0.074	0.141	0.160	0.103	0.107	0.122	0.132	0.154	0.086	0.093	0.121	0.108	0.078
200	0.106	0.065	0.117	0.082	0.098	0.138	0.108	0.127	0.047	0.125	0.082	0.099	0.126	0.202	0.098	0.101
204	0.141	0.111	0.134	0.318	0.076	0.119	0.052	0.117	0.179	0.210	0.080	0.092	0.111	0.053	0.039	0.110
208	0.120	0.093	0.025	0.066	0.080	0.070	0.062	0.037	0.124	0.070	0.013	0.055	0.093	0.078	0.078	0.161
212	0.077	0.083	0.179	0.076	0.047	0.060	0.093	0.013	0.040	0.070	0.125	0.116	0.058	0.139	0.088	0.060
216	0.035	0.167	0.020	0.029	0.043	0.045	0.026	0.030	0.015	0.107	0.072	0.038	0.045	0.045	0.078	0.032

Locus	Population Sample															
	Kong.	Savi.	Kiv. (s)	Kiv. (f)	Ikal.	Kell.	Kugu.	Nimi.	Cobb.	Sinuk.	Penny	Nome	Solo.	Niuk.	Kash.	Trail
220	0.035	0.056	0.020	0.039	0.029	0.019	0.026	0.013	0.147	0.018	0.053	0.031	0.071	0.045	0.010	0.028
224	0.049	0.009	0.003	0.026	0.011	0.006	0.005	0.003	0.009	0.026	0.021	0.031	0.056	0.058	0.039	0.032
228	0.007	0.009	0.006	0.026	0.033	0.006	0.005	—	0.015	0.015	—	0.024	0.023	0.018	0.049	0.023
232	0.021	0.028	—	—	—	—	—	0.013	0.018	—	0.013	0.010	0.003	0.010	0.029	0.023
236	—	—	—	—	—	0.017	0.005	0.020	0.005	—	—	0.003	0.013	0.005	—	0.005
240	0.014	—	—	—	—	—	—	0.037	0.012	—	0.005	—	—	—	—	0.014
244	—	—	—	—	—	0.006	—	0.003	—	—	—	—	—	—	—	0.014
248	0.007	—	—	—	—	0.009	—	—	—	—	—	—	—	—	—	0.005
252	0.007	—	—	—	—	—	—	—	—	—	—	—	—	—	—	0.009
264	—	—	—	—	—	—	—	—	—	0.015	—	—	—	—	—	0.005
A _R	17.43	12.78	13.70	14.33	17.68	18.57	17.56	19.99	16.67	15.62	15.87	18.20	17.45	16.32	16.82	20.81
H _e	0.915	0.898	0.883	0.859	0.917	0.914	0.915	0.924	0.899	0.891	0.915	0.924	0.922	0.902	0.914	0.928
H _o	0.930	0.926	0.877	0.879	0.913	0.902	0.938	0.873	0.905	0.926	0.931	0.925	0.899	0.894	0.882	0.908
P-HW	0.742	0.822	0.423	0.848	0.471	0.243	0.853	0.021	0.666	0.949	0.839	0.557	0.138	0.379	0.250	0.250
<i>Sma-3</i>																
N	65	54	187	187	140	265	98	151	330	136	188	146	197	199	48	99
120	0.354	0.102	0.358	0.283	0.279	0.187	0.235	0.235	0.483	0.390	0.572	0.408	0.414	0.294	0.448	0.515
122	0.646	0.889	0.602	0.714	0.682	0.675	0.694	0.712	0.476	0.555	0.396	0.541	0.553	0.628	0.531	0.475
124	—	0.009	0.040	0.003	0.039	0.136	0.071	0.053	0.041	0.051	0.032	0.051	0.033	0.078	0.021	0.010
126	—	—	—	—	—	0.002	—	—	—	0.004	—	—	—	—	—	—
A _R	2.00	2.89	2.99	2.26	2.99	3.18	3.00	3.00	2.99	3.35	2.97	3.00	2.98	3.00	3.00	2.74
H _e	0.462	0.201	0.509	0.411	0.457	0.491	0.461	0.437	0.539	0.539	0.516	0.540	0.523	0.514	0.523	0.512
H _o	0.338	0.222	0.508	0.385	0.486	0.494	0.459	0.430	0.536	0.559	0.532	0.541	0.503	0.503	0.458	0.424
P-HW	0.026	1.000	0.522	0.196	0.824	0.579	0.538	0.441	0.505	0.726	0.705	0.539	0.299	0.385	0.228	0.050
<i>Sma-5</i>																
N	71	54	193	189	141	265	97	151	319	135	188	147	199	197	50	113

Locus	Population Sample															
	Kong.	Savi.	Kiv. (s)	Kiv. (f)	Ikal.	Kell.	Kugu.	Nimi.	Cobb.	Sinuk.	Penny	Nome	Solo.	Niuk.	Kash.	Trail
90	1.000	1.000	1.000	1.000	1.000	1.000	1.000	1.000	1.000	1.000	1.000	0.997	0.995	1.000	1.000	1.000
92	—	—	—	—	—	—	—	—	—	—	—	0.003	0.005	—	—	—
A _R	1.00	1.00	1.00	1.00	1.00	1.00	1.00	1.00	1.00	1.00	1.00	1.33	1.43	1.00	1.00	1.00
H _c	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.007	0.010	0.000	0.000	0.000
H _o	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.007	0.010	0.000	0.000	0.000
P-HW	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA	1.000	1.000	NA	NA	NA
<i>Sma-24</i>																
N	71	52	172	189	121	264	99	143	328	135	188	145	198	199	51	109
154	—	—	—	—	—	—	—	—	—	—	0.008	—	—	—	—	—
158	0.007	—	0.003	0.082	—	0.013	0.025	0.007	0.003	—	0.005	—	—	—	—	—
162	—	—	0.012	—	—	—	—	—	—	—	—	—	—	—	—	—
166	—	—	0.006	0.003	—	0.011	—	0.014	—	—	—	0.007	0.008	0.003	—	—
170	—	—	—	0.019	0.029	0.021	0.025	0.038	—	0.004	—	—	—	—	0.010	—
174	—	—	0.017	0.045	0.017	0.017	0.035	0.028	0.011	0.015	—	0.007	0.003	0.003	0.010	—
178	—	—	—	—	—	0.006	0.005	—	0.020	—	0.003	0.003	0.010	0.015	—	0.023
182	0.014	0.019	—	—	—	0.006	0.005	—	0.006	0.004	0.005	0.010	0.013	0.003	—	0.005
186	—	0.010	0.009	0.021	—	0.004	—	0.003	0.034	0.004	—	0.048	0.040	—	0.010	—
190	—	—	0.017	0.008	0.004	—	—	—	0.002	0.007	0.013	0.014	0.051	0.020	0.010	0.005
194	0.014	0.144	0.015	0.008	0.012	0.011	—	0.035	0.005	—	0.013	0.034	0.033	0.010	0.020	0.009
198	0.021	0.058	—	0.005	0.004	—	—	—	0.037	0.022	0.013	0.041	0.025	0.018	0.029	—
202	0.014	0.010	0.061	0.037	0.004	0.011	0.005	—	0.006	0.019	0.005	0.014	—	0.003	0.069	0.018
206	0.035	0.019	0.032	0.011	0.004	0.019	0.005	0.035	—	0.015	0.024	0.014	0.013	0.003	0.010	0.014
210	0.028	0.010	0.009	0.003	0.025	0.032	0.035	0.035	0.006	0.022	0.021	0.017	0.025	0.005	0.039	0.037
214	0.056	0.058	0.023	0.013	0.099	0.083	0.056	0.038	0.017	0.085	0.037	0.024	0.020	0.063	0.059	0.060
218	0.014	—	0.032	0.143	0.029	0.008	0.030	0.070	0.102	0.030	0.008	0.024	0.010	0.020	0.029	0.046
222	0.063	0.019	—	0.011	0.033	0.055	0.040	0.028	0.015	0.026	0.035	0.066	0.033	0.073	0.088	0.087
226	0.049	0.096	0.032	0.029	0.045	0.053	0.071	0.052	0.032	0.022	0.051	0.076	0.096	0.035	0.049	0.055

Locus	Population Sample															
	Kong.	Savi.	Kiv. (s)	Kiv. (f)	Ikal.	Kell.	Kugu.	Nimi.	Cobb.	Sinuk.	Penny	Nome	Solo.	Niuk.	Kash.	Trail
230	0.042	0.058	0.017	0.042	0.045	0.044	0.091	0.042	0.063	0.056	0.136	0.072	0.073	0.053	0.020	0.073
234	0.099	0.029	0.020	—	0.021	0.064	0.045	0.017	0.008	0.033	0.051	0.052	0.043	0.073	0.049	0.032
238	0.148	0.038	0.099	0.071	0.112	0.051	0.071	0.105	0.078	0.096	0.037	0.028	0.023	0.123	0.039	0.009
242	0.049	0.058	0.070	0.034	0.062	0.030	0.056	0.073	0.032	0.030	0.090	0.097	0.104	0.050	0.020	0.023
246	0.035	0.087	0.032	0.024	0.041	0.070	0.091	0.049	0.021	0.019	0.120	0.055	0.071	0.070	0.039	0.083
250	0.070	0.077	0.116	0.063	0.050	0.045	0.061	0.056	0.058	0.026	0.106	0.028	0.040	0.050	0.059	0.064
254	0.028	0.019	0.078	0.013	0.050	0.030	0.051	0.017	0.131	0.081	0.059	0.048	0.071	0.065	0.069	0.060
258	0.028	0.029	0.093	0.103	0.037	0.038	0.020	0.024	0.021	0.030	0.005	0.034	0.010	0.040	0.049	0.060
262	—	—	0.017	0.034	0.025	0.032	0.020	0.014	0.166	0.070	0.040	0.038	0.043	0.063	0.049	0.060
266	0.014	0.038	0.140	0.108	0.029	0.080	0.056	0.049	0.023	0.004	0.016	0.041	0.033	0.028	0.069	0.032
270	0.007	0.019	0.023	0.034	0.041	0.042	0.010	0.063	0.009	0.011	0.024	0.017	0.028	0.020	0.029	0.023
274	0.007	0.048	0.020	0.026	0.091	0.044	0.040	0.021	0.003	0.022	0.032	0.014	0.013	0.033	0.029	0.037
278	0.028	0.029	0.006	0.005	0.062	0.034	0.030	0.007	0.075	0.030	0.024	0.010	0.018	0.020	0.010	0.023
282	0.021	0.010	—	0.003	0.004	0.030	0.010	0.035	0.003	0.056	0.013	0.014	0.025	0.020	0.010	0.018
286	0.028	0.019	—	—	0.004	0.006	0.005	0.021	0.009	0.104	—	0.007	0.008	0.005	0.020	0.009
290	0.063	—	—	—	—	—	0.005	—	0.003	0.041	0.003	0.007	0.008	—	—	0.014
294	0.007	—	—	—	0.004	0.004	—	0.021	—	0.007	—	0.014	0.003	0.005	0.010	—
298	0.007	—	—	—	0.004	—	—	—	—	0.004	0.003	—	0.005	0.003	—	0.014
302	—	—	—	—	—	0.002	—	—	—	—	—	0.007	—	—	—	0.005
306	—	—	—	—	0.012	0.002	—	—	0.003	—	—	—	—	0.003	—	0.005
310	—	—	—	—	—	—	—	—	—	—	—	0.010	0.005	0.005	—	—
314	—	—	—	—	—	—	—	—	—	—	—	0.007	—	—	—	—
326	—	—	—	—	—	0.002	—	—	—	—	—	—	—	—	—	—
334	—	—	—	—	—	—	—	—	—	0.004	—	—	—	—	—	—
338	—	—	—	—	—	—	—	—	—	0.004	—	—	—	—	—	—
A _R	25.74	23.66	22.04	22.28	24.31	26.32	24.11	25.34	21.81	26.33	23.26	29.04	26.64	24.08	28.52	25.85
H _e	0.945	0.942	0.929	0.931	0.946	0.954	0.951	0.953	0.920	0.948	0.932	0.956	0.949	0.944	0.961	0.953
H _o	0.775	0.904	0.762	0.937	0.843	0.879	0.909	0.979	0.905	0.919	0.926	0.945	0.939	0.950	0.922	0.945

Locus	Population Sample															
	Kong.	Savi.	Kiv. (s)	Kiv. (f)	Ikal.	Kell.	Kugu.	Nimi.	Cobb.	Sinuk	Penny	Nome	Solo.	Niuk.	Kash.	Trail
P-HW	0.000	0.175	0.000	0.663	0.000	0.000	0.051	0.966	0.180	0.098	0.397	0.301	0.307	0.684	0.150	0.410
Overall																
A_R	11.30	10.67	9.99	9.42	11.47	11.91	11.34	10.94	10.51	11.65	11.42	13.04	12.01	11.09	12.43	12.42
H_e	0.643	0.604	0.651	0.648	0.656	0.644	0.659	0.630	0.662	0.702	0.698	0.712	0.701	0.692	0.680	0.684
H_o	0.580	0.583	0.626	0.654	0.636	0.627	0.651	0.629	0.654	0.706	0.693	0.708	0.704	0.690	0.628	0.669
P-HW	0.000	0.064	0.003	0.740	0.027	0.006	0.260	0.443	0.124	0.650	0.274	0.332	0.659	0.408	0.003	0.093