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PHYLOGEOGRAPHY AND POPULATION DYNAMICS OF NORTH AMERICAN WOLVERINES (*GULO GULO LUSCUS*) IN ALASKA AND WESTERN CANADA

by

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B.A., BIOLOGY
SIMPSON COLLEGE
2013

THESIS

Submitted in Partial Fulfillment of the
Requirements for the Degree of

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in Biology

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M.S., Biology, University of New Mexico, 2017

ABSTRACT

The circumboreal wolverine (*Gulo gulo sp.*) is ideal for studying responses to environmental perturbation in the North due to a history of persistence in glacial refugia and subsequent glacial recolonization. Wolverines are also excellent indicators of human influence on the environment due to their close association with remote areas and cold, snowy climes. Through the use of genetic tools (i.e., nuclear microsatellite loci and mitochondrial sequences), I examined population structure of wolverines in Alaska and western Canada to identify signatures of glacial refugia, bottlenecks, and distinctive populations, sex-biased dispersal, gene flow, and source and sink population dynamics. I identified genetic structuring and key source areas that may be vital in maintaining viable populations in the southern regions of the wolverine's range. Through this research I have further elucidated the evolutionary history of wolverines and contributed to the conservation future of this elusive species.

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INTRODUCTION

Climate fluctuations of the past greatly influenced distributions of various species, limiting and expanding ranges and sometimes eliminating them entirely, especially in high latitudes (Scheffers et al. 2016). Human-influenced climate change and habitat disturbance also are now changing environments faster than most past geologic change (Kerby and Post 2013). As we head into an uncertain climate future, the study of certain indicator species will aid in tracking the impacts of climate change and habitat destruction on species, as well as aid in monitoring successes in mediating destruction and in connecting otherwise fragmented populations.

As a cold- and disturbance-sensitive mammalian mesocarnivore, the widely distributed wolverine (*Gulo gulo luscus*, Lineaus 1758) is an excellent indicator species for understanding human impacts on our planet (Carroll et al. 2001; Chadwick 2010). As solitary and highly vagile predators, wolverine movement is often not restricted by typical barriers such as mountains, rivers, or valleys (Hornocker and Hash 1981). Instead, wolverine distribution and movement is restricted to environments with cold climates that have late spring snow pack for building dens and successfully raising kits (Greenwood 1980) and areas with tundra, boreal forest, mountain forest, or rock talus habitat (Laliberte and Ripple 2004) that is undisturbed by human traffic or noise (Scrafford et al. 2017). An ample supply of ungulate prey (Magoun 1987) and smaller prey (ground squirrels, birds) or fruits, eggs, and insect larva (Pasitschniak-Arts 1995) is also critical. With that suite of requirements and relatively few natural barriers to dispersal, I studied the evolutionary history of the wolverine to provide a historically deep understanding of the species distributional and evolutionary change through space and time, and then

applied that foundation to a contemporary study of population genetics and ecology in high latitude environments.

In Chapter 1, I examined the phylogeographic history of the wolverine across Alaska and western Canada. Populations are tested for evidence of isolation in past glacial refugia, as well as evidence of population bottlenecks (less than approximately 25 individuals; Hoelzel 1993). Pairwise degree of relatedness among populations was assessed, and patterns of gene flow were observed through tests of migration and source-sink dynamics (areas with greater births than deaths, and areas with greater deaths than births; Dias 1996). Previous studies of other species showed signatures of glacial refugia in Beringia, the northern most glacial refuge for North America (Abbott et al. 2000; Hultén 1937). Our tests for these signatures were applied to contemporary populations in Russia, Northwest Alaska, and North Alaska. Southeast Alaska has also been identified as a refugium for other species (Carrara et al. 2007; Fleming and Cook 1999; Mandryk et al. 2001), so we tested that refugial possibility for wolverines. The Kenai Peninsula, a narrowly connected peninsula on the southern coast of mainland Alaska, has been reported to harbor divergent, low-density populations of several species (Bailey et al. 1986; Morton et al. 2016) due to bottlenecks and isolation from the mainland, perhaps warranting conservation concern for wolverines as well as other organisms.

Chapter 2 explored sex-biased dispersal patterns in two high-latitude biomes (i.e., tundra and taiga) and compared these patterns reported for wolverines in temperate latitudes in North America. Sex-biased dispersal refers to greater natal dispersal (i.e., a single, permanent move from the natal territory) in one sex relative to the other; in this case males are hypothesized to exhibit greater dispersal than females. Dalerum et al.

(2007) found no sex-bias in dispersal on the Seward Peninsula of Alaska, which contrasts with lower-latitude telemetry studies of wolverines. To test whether greater dispersion of resources (i.e., tundra environments have lower productivity than forest biomes, so that resources are less concentrated) is the cause of greater female dispersal relative to males, I tested the two datasets to screen for different patterns among adult and subadult (potentially pre-dispersal, dispersing, or post-dispersal; Banci and Harestad 1990; Vangen et al. 2001) males and females.

This study examined the evolutionary dynamics of a cold-adapted species, analyzed data on their dispersal in various habitats, and provided insight into these fundamental ecological and evolutionary functions (i.e., dispersal and gene flow) in the context of an uncertain climate future. Wolverines can serve as harbingers of change for the entire ecological community they inhabit; understanding what is perceived as a barrier to movement is important for managing increasingly fragmented populations of this rare species as well as a host of other species that may be experiencing similar anthropogenic impacts.

Literature Cited

Abbott, R. J., L. C. Smith, R. I. Milne, R. M. M. Crawford, K. Wolff, and J. Balfour. 2000. Molecular analysis of plant migration and refugia in the Artic. Science 289: 1343–1346.

Bailey, T. N., E. E. Bangs, M. F. Portner, J. C. Malloy, and R. J. McAvinchey. 1986. An apparent overexploited lynx population on the Kenai Peninsula, Alaska. The Journal of Wildlife Management 50(2): 279-290.

Banci, V., and A. S. Harestad. 1990. Home range and habitat use of wolverines *Gulo gulo* in Yukon, Canada. Holarctic Ecology 13(3): 195–200.

Carrara, P. E., T. A. Ager, and J. F. Baichtal. 2007. Possible refugia in the Alexander

Archipelago of southeastern Alaska during the late Wisconsin glaciation. Canadian Journal of Earth Sciences 44: 229–244.

Carroll, C., R. F. Noss, and P. C. Paquet. 2001. Carnivores as focal species for conservation planning. Ecological Applications 11(4): 961–980.

Chadwick, D. H. 2010. The Wolverine Way. Ventura, CA: Patagonia Books.

Dalerum, F., J. Loxterman, B. Shults, K. Kunkel, and J. A. Cook. 2007. Sex-specific dispersal patterns of wolverines: insights from microsatellite markers. Journal of Mammalogy 88(3): 793–800.

Dias, P. C. 1996. Sources and sinks in population biology. Trends in Ecology and Evolution 11(8): 326–330.

Fleming, M. A., E. A. Ostrander, and J. A. Cook. 1999. Microsatellite markers for American mink (*Mustela vison*) and ermine (*Mustela erminea*). Molecular Ecology 8(8): 1352–1355.

Greenwood, P. J. 1980. Mating systems, philopatry and dispersal in birds and mammals. Animal Behaviour 28(4): 1140–1162.

Hoelzel, A.R., J. Halley, S. J. O'Brien, C. Campagna, T. Arnborm, B. Le Boeuf, K. Ralls, G. A. Dover. 1993. Elephant seal genetic variation and the use of simulation models to investigate historical population bottlenecks. Journal of Heredity 84(6): 443–449.

Hornocker, M. G., and H. S. Hash. 1981. Ecology of the wolverine in northwestern Montana. Canadian Journal of Zoology 59(7): 1286–1301.

Hultén, Eric. 1937. Outline of the history of Arctic and boreal biota during the Quaternary Period: their evolution during and after the glacial period as Indicated by the equiformal progressive areas of present plant species. Stockholm: Bokförlags aktiebolaget Thule.

Kerby, J. T., and E. Post. 2013. Advancing plant phenology and reduced herbivore production in a terrestrial system associated with sea ice decline. Nature Communications 4(2514): 1-6.

Laliberte, A. S., and W. J. Ripple. 2004. Range contractions of North American carnivores and ungulates. BioScience 54(2): 123-138.

Magoun, A. J. 1987. Summer and winter diets of wolverines, *Gulo gulo*, in arctic Alaska. Canadian field-naturalist. Ottawa ON 101(3): 392–397.

Mandryk, C. A., H. Josenhans, D. W. Fedje, and R. W. Mathewes. 2001. Late Quaternary paleoenvironments of Northwestern North America: implications for inland

versus coastal migration routes. Quaternary Science Reviews 20(1): 301–314.

Morton, J. M., G. C. White, G. D. Hayward, D. Paetkau, and M. P. Bray. 2016. Estimation of the brown bear population on the Kenai Peninsula, Alaska. The Journal of Wildlife Management 80(2): 332-346.

Pasitschniak-Arts, M., and S. Larivière. 1995. *Gulo gulo* Species Account. The American Society of Mammalogists 499: 1–10.

Scheffers, B. R., L. De Meester, T. C. Bridge, A. A. Hoffmann, J. M. Pandolfi, R. T. Corlett, S. H. M. Butchart, P. Pearce-Kelly, K. M. Kovacs, D. Dudgeon, M. Pacifici, C. Rondinini, W. B. Foden, T. G. Martin, C. Mora, D. Bickford, J. E. M. Watson. 2016. The broad footprint of climate change from genes to biomes to people. Science, 354(6313): 719–732.

Scrafford, M. A., T. Avgar, B. Abercrombie, J. Tigner, and M. S. Boyce. 2017. Wolverine habitat selection in response to anthropogenic disturbance in the western Canadian boreal forest. Forest Ecology and Management 395(1): 27–36.

Vangen, K. M., J. Persson, A. Landa, R. Andersen, and P. Segerström. 2001. Characteristics of dispersal in wolverines. Canadian Journal of Zoology 79(9): 1641–1649.

CHAPTER 1

DYNAMIC NORTHERN LANDSCAPES THROUGH TIME AND SPACE AFFECT GENOMES OF THE NORTH AMERICAN WOLVERINE (*GULO GULO LUSCUS*)

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Abstract

The cyclic climatic fluctuations of the Late Quaternary produced a dynamic biogeographic history for the fauna and flora of northwestern North America. To continue to refine our understanding of this history, we examine demographic and geographic structure in a widespread carnivore, the North American wolverine (*Gulo gulo* Linnaeus, 1758), across the vast mainland of Alaska, coastal Southeast Alaska, and the mainland of western Canada using nuclear microsatellites and mitochondrial DNA (control region and cytochrome *b*) sequences. Maternally inherited mitochondrial data reflected stable populations in Northwest Alaska, suggesting this region harbored wolverine populations since at least the Last Glacial Maximum (LGM) (21 Kya), a

finding consistent with their fossil record of persistence in Beringia. Southeast Alaska also had minimally divergent populations, perhaps due to contemporary isolation, but likely not refugial persistence. Southeast Alaska population divergence coincides with the lack of pre-Holocene fossil records for wolverines from the region. Kenai Peninsula populations exhibited mixed signals of population stability dependent on marker type: matrilineal mtDNA showed a signature of stability (i.e., historical persistence) and a private haplotype, whereas nuclear microsatellites exhibited relatively low variation and a lack of private alleles consistent with a Holocene colonization of the peninsula by wolverines. A weak phylogeographic break between mainland Alaska and western Canada coincides with the eastern edge of the LGM Beringian refugium and mirrors similar disjunctions identified in ermine and brown bear.

Keywords: colonization, genomic comparison, glacial history, *Gulo gulo luscus*, phylogeography

Introduction

Cold-adapted species are excellent models to study the effects of climate change, which is increasingly implicated in deleterious effects on high latitude biomes (Scheffers et al. 2016). As a large carnivore restricted to cold environments, the wolverine (*Gulo gulo*) could become a model species for conservation monitoring (Carroll et al. 2001), and like the polar bear (*Ursus maritimus*), is a compelling symbol of how accelerated climate change is transforming northern environments (Chadwick 2010).

Seemingly unaffected by physiographic barriers such as rivers, reservoirs, valleys, or mountain ranges (Hornocker and Hash 1981), wolverines instead appear to respond primarily to changing climatic conditions and human influence. Wolverines require spring snowpack that persists through at least mid-May to successfully den kits, and this species generally cannot tolerate average summer temperatures above 22°C (Copeland et al. 2010). Those requirements may be key to the northward range shift documented over the last 40 years that produced a 37% reduction in wolverine distribution in North America (Laliberte and Ripple 2004). Beyond ecological and physiological requirements, the highly vagile wolverine generally shows limited genetic variability across its North American range (Rico et al. 2015; Zigouris et al. 2013). However, limited geographic structure that may exist could reflect important barriers to dispersal, bioclimatic restrictions, or colonization routes for wolverines, and some of these barriers may also impact other syntopic species.

With regard to historical barriers, biomes in Alaska and western Canada were strongly influenced by the dynamic glacial history of the region (Cook et al. 2006; Fedorov and Stenseth; Hope et al. 2011; Rowe et al. 2014). Glacial ice sheets covered most of northern North America, growing and receding through the Pleistocene (> 24 glacial/interglacial cycles; 2.6 Mya - 11.7 Kya) and genetically structuring populations in these regions (Hope et al. 2013; Knowles et al. 2016; Shafer et al. 2010; Weksler et al. 2010). During the course of these glacial periods, several ice-free refugia allowed species to persist *in situ* during glacial maxima (**Fig 1**). The location of larger North American glacial refugia are well documented in fossils, pollen records, genetic structure of fauna and flora, and bathymetric and stratigraphic evidence (Barrie and Conway 1998; Cook et

al. 2017) while other generally smaller refugia (e.g., Kodiak, Kenai) are debated (Gentili et al. 2015; Harlin-Cognato et al. 2006). Beringia extended from eastern Siberia to at least the Mackenzie River of northwestern Canada (Abbott et al. 2000; Hultén 1937). Although often portrayed as a single large and continuous refugium, Beringia likely was a heterogeneous landscape (Hoffmann 1981; McLean et al. 2016). In northern Alaska, Beringia was fragmented by the Brooks Range glacial ice sheet until about 13.5K B.P. (Dyke 2004), as reflected in geographic structure of widely-distributed species (e.g., *Urocitellus parryii,* Eddingsaas et al. 2004; Galbreath et al. 2011). Another, smaller coastal refuge has been hypothesized off the coast of Southeast Alaska that today consists of a series of archipelagos and thin strip of mainland (Carrara et al. 2007; Fleming and Cook 1999; Josenhans et al. 1995, Mandryk et al. 2001). Finally, south of the Cordilleran and Laurentide ice sheets were a series of large southern refugia, roughly broken by physiographic features like the southern Rocky Mountains and Mississippi River (Swenson and Howard 2005).

Isolation in the Beringian and Southeast Alaskan Coastal refugia, combined with topographic complexity (e.g., mountain ranges, peninsulas, islands) and variable biomes (e.g., tundra, taiga; Laliberte and Ripple 2004) in this region of North America, created a complex history for species in northwestern North America (Cook et al. 2017). Previous genetic analyses focused on a subset of these populations (Dalerum et al. 2007) or treated all populations from Alaska as a single unit (e.g., Kyle and Strobeck 2002; Zigouris et al. 2013) lumping wolverines from Southeast Alaska with distant mainland Alaska, British Columbia, or Yukon Territory populations.

Here, we extend previous work with a more intensively sampled study of geographic structure in wolverines of far northwestern North America to investigate the impact of climate on demographic history with a focus on populations in Alaska and western Canada. Based on fossil evidence, we predict genetic signatures of glacial refugia will be found in northwestern and northern Alaska populations that reflect deeper refugial isolation. We also predict signatures of a genetic bottleneck on the Kenai Peninsula due to its narrow connection to the mainland (16 kilometers) and previous work showing that other large carnivores exhibit subpopulation divergence on the peninsula (lynx, Bailey et al. 1986, Bailey 2002; brown bear, Morton et al. 2016). Source populations (i.e., those where birth rates exceed death rates and emigration exceeds immigration; Dias 1996) are predicted in high latitude boreal forest populations (such as those in the Yukon Territory) and sink populations in lower latitude regions like British Columbia (Krebs et al. 2004).

Materials and methods

Study areas and sampled individuals

We collaborated with state and provincial wildlife officials to permanently archive wolverine carcasses salvaged from commercial trappers in Alaska and western Canada. Specimens were processed and cataloged at the Museum of Southwestern Biology, University of New Mexico and Museum of the North, University of Alaska Fairbanks over the last ~2.5 decades (1989-2015). High-quality samples (Supplementary Material **S2 Table**) with reliable spatial and temporal information were selected to broadly represent the region, with populations identified based on major geographic and topographic features. Political boundaries were used to assign regional group names.

Groups (**Fig 2**; **Fig 3**) were analyzed for F_{IS} inbreeding coefficient (**Table 1**; Wright 1921) in FSTAT v1.2 (Goudet 1995) to identify subpopulation structure and prevent lumping of distinct populations (Goudet 1993 and Goudet et al. 1994).

Following calibration of genotypes between laboratories, data from previous work (Dalerum et al. 2007) were added to our dataset to represent sampling from northwestern Alaska (NWAK; $N = 117$). Those specimens were represented by only ten microsatellite loci, while all other sampling regions were sequenced for the original ten loci and ten additional loci (**S2 Table**). Comparisons between NWAK and all other populations were conducted using the 10-locus suite common among all specimens (**S2 Table**). Similarly, analyses of genetic diversity (i.e., H_O/H_E and STRUCTURE output) were conducted with these 10 loci when comparing NWAK with other populations; otherwise, full data were used for intrapopulation analyses or comparisons that excluded NWAK. A small number of individuals had either 10% (Central Alaska, $N = 1$) or 5% (Southeast Yukon, $N = 1$) missing data. Exploratory analyses demonstrated those missing data had no significant impact on analyses (not shown).

DNA extraction and nucleotide sequencing

DNA was isolated using a salt extraction method (Fleming and Cook 2002). Data from Francés (2008) were combined with new mitochondrial (mtDNA) data for analyses. Primer sets L15626 and H16498 (Francés 2008; 369 base pairs or bp) or L15926 and H16498 (Tomasik and Cook 2005; 366-367bp) were used to produce 366bp control region sequences from mtDNA. Primer sets MSB05 and MSB14 (Hope et al. 2010) were used to sequence 1140bp of the cytochrome *b* region of the mitochondrial genome.

DNA was amplified by polymerase chain reaction (PCR) in a final volume of 25 µL containing 2-50 ng genomic DNA, 25 mM magnesium chloride, 1.0 µg bovine serum albumin, 2 mM deoxynucleoside triphosphates, 10 µM each of forward and reverse primers (MSB05 and MSB14; Hope et al. 2010), 10xPCR buffer (Perkin Elmer Cetus I), DNA-free water, and 0.13 µL Amplitaq DNA polymerase (PE Biosystems, Forest City, CA). PCRs had an initial denaturation of 94°C for 3 min followed by 34 cycles of 94°C for 15 s, 50-51°C for 30 s, 72°C for 30 s, and a 10 min final extension at 72°C. Negative controls accompanied each set of PCR reactions. PCR products were visualized on agarose gel, purified by polyethylene glycol (PEG) precipitation, resuspended in 10 mM Tris, and cycle-sequenced using 3.2 µM forward and reverse primers, Applied Biosystems BigDye® Terminator v1.1, v3.1 5x Sequencing Buffer (Thermofisher Scientific), and thermocycler settings: 96°C initial denaturation for 1 min followed by 24 cycles of 96°C for 15 s, 50°C for 5 s, and 60°C for 4 min. Cycle-sequencing products were cleaned using 1 µL of 125 mM EDTA, 1 µL of 3M sodium acetate, and a 100% and 70% cold ethanol incubation and wash. After drying, addition of 10 µL of formamide, and denaturation (95°C for 5 min), samples were Sanger sequenced through the UNM Molecular Biology Facility.

Geneious v8.0 (http://www.geneious.com, Kearse et al. 2012) was used for sequence editing, and a reference sequence using a consensus of GenBank (Benson et al. 2009) sequences for wolverines was created for assembling sequences before editing. Sample size was augmented to a total of $N = 252$ for control region and $N = 67$ for cytochrome *b* by adding data from GenBank ($N = 151$, $N = 21$ respectively). Bayesian phylogenetic trees were generated in BEAST v1.8 (Bouckaert et al. 2014) using individuals that had

both control region and cytochrome *b* sequences ($N = 59$), as well as independent trees (**S13-15 Figs**). The two mitochondrial genes were analyzed as a concatenated unit; when screened independently (jModeltest v2.1.4; Darriba et al. 2012) they had similar models of evolution (HKY+I control region and concatenated; HKY cytochrome *b*).

Microsatellite genotyping

Microsatellite genotypes were determined for 177 individuals from Alaska and western Canada using 20 loci (**S3 Table**) with the following PCR primers: Lut604 (Dallas and Piertney 1998), Gg-3, Gg-4, Gg7, MA-3, Tt-1, Tt-4 (Davis and Strobeck 1998), Ggu216 (Duffy et al. 1998), Mvis75 (Fleming et al. 1999), Gg10, Gg25, Gg37, Gg42, Gg192, Gg443, Gg452, Gg454, Gg465, Gg471, Gg473 (Walker et al. 2001). One primer (forward or reverse) from each of seven of the loci was redesigned from the published sequence to change the size or improve quality of PCR product (**S3 Table**). PCR amplifications were carried out in seven universal-tailed (Oetting et al. 1995) multiplex reactions and one singleplex reaction, each in a final volume of 10 μL and containing 2-50 ng genomic DNA, 0.2 mM deoxynucleoside triphosphates, 1-5 pmols unlabeled primers, 0.15-2.25 pmoles IRD-labeled primer, 1.0 μg bovine serum albumin, 1xPCR buffer (Perkin Elmer Cetus I), and 0.25-0.5 units of GoTaq®Flexi DNA polymerase (Promega, Madison, WI). PCRs began at 94°C for 2 min then continued with 40 cycles each of 94°C for 15-30 s, 50°C for 15-30 s, and 72°C for 30-60 s. A 30 min extension at 72°C concluded each reaction.

Fluorescently labeled PCR products were electrophoresed on a 48-well 6% polyacrylamide gel on a LI-COR 4200 LR or IR^2 DNA automated sequencer (LI-COR, Lincoln NE). For allele size standardization for the 10 loci amplified by Dalerum et al.

(2007), we generated a suite of size standards for each locus by sizing 2-4 samples against standards of known size (four wolverine samples used by Dalerum et al. 2007 and provided by Janet Loxterman). Two of those comparative standards were used in a minimum of 6 lanes in all subsequent genotyping reactions. For the remaining loci, size standards were generated for each locus by scoring the same suite of individuals against a fluorescently-labeled M13 sequence ladder of known size, and those samples were used in each subsequent gel, again occupying at least six lanes across each 48-well gel. Based on these comparisons, genotypes for each individual were determined using GeneImagIR 4.05 software (Scanalytics, Inc.). For quality control, 10% of samples were extracted, amplified, and genotyped in duplicate. Locus error rate was determined at 0.97%, allele error rate was 0.32% for non-NWAK samples. MICROCHECKER 2.2.3 (Van Oosterhout et al. 2004) was used to identify genotyping errors and check for null alleles. Positive and negative controls were used throughout.

To limit the over-sampling of family groups (and correct for family-based structure that might confound population structure; Pritchard and Wen 2004, Bergl and Vigilant 2007, Anderson and Dunham 2008), analyses were run with a dataset restricted by relatedness (r). One individual in each of a given pair related above $r_{xy} = 0.5$ in IDENTIX v1.1.5 (Belkhir et al. 2002) using the Queller & Goodnight (1989) relatedness estimator after 1000 per locus bootstraps (to achieve a 95% confidence interval for each) was removed. Total sampling after parsing for relatedness was $N = 207$.

Descriptive statistics

Genepop on the Web v4.2 (Raymond and Rousset 1995) was used to evaluate Hardy-Weinberg equilibrium (HWE) and linkage disequilibrium (LD) for each

microsatellite locus and pairs of loci, respectively. HWE was tested using probability, heterozygosity deficiency, and heterozygosity excess with no enumeration of alleles using Markov Chain parameters: 10,000 dememorizations, 1,000 batches, and 10,000 iterations. LD was tested with log-likelihood and probability tests and the same Markov Chain parameters. Alpha-values (0.05) were adjusted by the number of populations, implementing a Bonferroni correction, to achieve a critical value to test for significance (0.005) across all comparisons (Rice 1989).

F-statistics (F_{ST} and F_{IS}) (Weir and Cockerham 1984; Wright 1951) were calculated using the software FSTAT 2.1 (Goudet 1995) with significance levels set at alpha = 0.001 and 10,000 randomizations. We assessed the possibility that microsatellite markers were sex-linked by comparing allele frequencies between males and females. Heterozygosity estimates (expected and observed) and number of alleles were estimated in Microsatellite Toolkit (Park 2001). We used the program HP-RARE (Kalinowski 2005) to calculate both allelic richness (R_A) and private allelic richness (R_P). HP-RARE uses rarefaction analyses to account for differences in sample size among populations. To assess degree of genetic structuring among microsatellite loci, we performed an AMOVA using co-dominant genotype data in Arlequin v3.5 (Excoffier and Lischer 2010).

Haplotype assignment and frequency rates among populations for sequences were also determined in Arlequin v3.5. Summary statistics including haplotype diversity (*Hd*; DnaSP v5) (Rozas *et al*. 2010), number of haplotypes (*H*), number of private haplotypes (H_P), analysis of molecular variance (AMOVA), and F_{ST} (Arlequin v3.5) were generated. F_{ST} values were computed using pairwise difference and 1,000 permutations.

Population Structure: individual-based clustering

STRUCTURE v2.3 (Falush et al. 2007) was used to examine genetic population structure without *a priori* designation of populations or sampling locations using Bayesian clustering (Pritchard et al. 2000). Optimal number of subpopulations was determined by varying the likely number of clusters or populations (k) from 1 to 10 allowing for genetic admixture and correlated allele frequencies (Falush et al. 2003). Each run used a burn-in of 50,000 and a MCMC of 500,000 steps. This process was replicated 8 times for each value of k (Evanno et al. 2005) to quantify the standard deviation among the runs for a particular assumed k. The optimal number of k-clusters was determined by Structure Harvester v0.6.94 (Pritchard et al. 2000), with the method developed by Evanno *et al.* (2005) to evaluate the rate of change in the log probability of the data (Δk) among 8 runs for each assumed k and estimate the highest Ln probability of the data or Ln P(d). Individual membership probabilities of the inferred k-clusters from the 8 independent replicates were averaged using CLUMPP v1.1.2 (Jakobsson and Rosenberg 2007).

As another test of population structure, BAPS 5.3 (Bayesian Analysis of Population Structure; Corander and Marttinen 2006; Corander et al. 2006) was also used to describe genetic structure. Unlike STRUCTURE, BAPS infers clusters based on similarities in the variance of data from assumed source populations (*i.e.*, *a priori* defined groups—Corander et al. 2006). As a result, the inference of k-clusters was set not to exceed the number of sampling areas (10). We inferred the maximum k to be between 1 and 10, with 20 replications of each inferred k. For the admixture analyses, parameters were set as: minimum population size of 5 individuals for admixture analyses with

10,000 iterations per population and at least 5 reference individuals from each population with 10,000 iterations per reference individual.

Demographic changes and bottlenecks

Graphical displays of demographic change in sequence data were executed in Mismatch Analysis in DnaSP v5 (Rozas et al. 2010). Tests for selection or deviations from neutrality using Tajima's *D* (Tajima 1989) and Fu's F_s (Fu 1997) were also done in DnaSP v5 (10,000 replicates for each). Extended Bayesian Skyline Plots (EBSPs) for mitochondrial genes were created in BEAST v.2.4.3 (Bouckaert et al. 2014) and visualized in R (Heled 2014.). Those analyses have more power in multi-locus investigations (Heled and Drummond 2010) but were conducted for comparison. The calibrated mutation rate for wolverines was set at 0.0428 substitutions/site/Myr (Hope et al. 2014).

Bottleneck v1.2.02 (Cornuet and Luikart 1996) was used to test patterns of fluctuation in effective population size (N_e). Under a mutation-drift equilibrium scenario, more heterozygotes than expected (heterozygosity excess) given the number of alleles would indicate a bottleneck (Cornuet and Luikart 1996). Heterozygosity excess was tested using a Wilcoxon sign rank test (optimal for 20 or fewer loci; Piry et al. 1999) under a two-phase model of microsatellite evolution (TPM, ideal for testing dinucleotide repeat loci; Di Rienzo et al. 1994) for 10,000 iterations. The infinite allele model (IAM; Kimura and Crow 1964) is a more liberal model but can indicate recent bottlenecks, and the strict stepwise mutation model (SMM; Ohta and Kimura 1973) is more conservative but can indicate more historical bottlenecks. These models were also tested to screen for consistency in calls for bottlenecks. If SMM and TPM both indicate a bottleneck, it is

likely to have occurred historically; if IAM alone is significant for heterozygosity excess it could be a false positive or evidence of recent a bottleneck. Variance for TPM was tested at 9 and 30 while proportion of SMM in TPM was left at 80% (Piry et al. 1999; Garza and Williamson 2001).

Migration and connectivity

Source-sink dynamics were examined through the program MIGRATE v3.6.11 (Beerli 1998, 2002; Beerli and Felsenstein 1999) to examine number of migrants per generation for mtDNA control region data ($N_f m$) among sampled sites. MIGRATE incorporates two parameters scaled to mutation rate (μ): theta, the effective population size parameter ($N_f \mu$), and *M*, the rate of gene flow (m/μ). MIGRATE gene flow estimates are averaged over the past *n* generations, where *n* equals the number of generations in which the populations have been at mutation-drift equilibrium. Gene flow estimates included a full migration model (theta and *M* were estimated individually from the data) compared to the restricted model (theta was averaged and *M* was symmetrical between populations). Gene flow was estimated using maximum-likelihood search parameters; 10 short chains (5,000 trees used out of 1 million sampled), five long chains (10,000 trees used of 2 million sampled), and five adaptively heated chains (start temperatures: 1, 1.5, 3, 6, and 12; swapping interval = 1). Models were conducted three times and parameter estimates converged. The alternative model was evaluated for goodness-of-fit given the data, using a log-likelihood ratio test (Beerli and Felsenstein 2001).

Results

Genetic diversity: microsatellites and mtDNA

For the microsatellite loci, there were no significant departures from HWE and LD was not evident. NWAK and NWY had the highest observed heterozygosity for microsatellite analyses, while SEAK and BC had the lowest (**Table 1**). Rarefied allelic richness ranged from 2.82 to 3.43 alleles per region, with NAK and KAK having the lowest and NWY, SEY, and BC having the highest rarefied allelic richness (**Table 1**). Private allelic richness was highest in BC and NWAK.

For mtDNA, haplotype diversity was lowest in SEY and NU. Highest haplotype diversity was found in NWAK and SEAK. All values for Tajima's D are negative, indicating the presence of more low frequency polymorphisms than expected, consistent with either population size expansion or purifying selection influencing North American wolverine populations (Librado and Rozas 2009). We constructed mismatch distribution plots (**Fig 4**; **S5-6 Figs**) that demonstrated demographic stability (or long-term occupation) in RUS, NWAK/NAK, and SEAK. SAK and KAK may also share this signature, but to a lesser degree as they are bi-modal instead of multi-modal. CAK, YT, BC, and NU, in contrast, have signals of expansion or more recent colonization. EBSPs demonstrated no expansion or depletion across all sequence data and geographic regions (**S7-9 Figs**). Those results are unsurprising, however, as those analyses have greater power in multi-locus investigations (Heled and Drummond 2010).

Inbreeding statistics were calculated using Fstat (Goudet 1995). Positive values indicate inbred lines, while negative values indicate crossing of differentiated lineages. An $F_{IS} > +0.043$ indicates an excess of contemporary inbreeding (Wright 1965). A strongly negative value (e.g., individuals placed in the same population when they should be separated) would indicate that populations should be further refined. Based on FSTAT

results, our populations have been appropriately assigned and no groups are excessively outbred (**Table 1**). SEAK has the highest inbreeding coefficient at +0.111; NWAK is most strongly differentiated at -0.059.

Microsatellite data showed evidence of a recent (2*Ne*-4*Ne* generations) bottleneck in SAK, KAK, NWY, SEY, and SEAK in at least one test; that is, they each exhibited heterozygosity excess in Bottleneck v1.2.02 (Piry et al. 1999). For the Wilcoxon test, all of these were under the infinite allele model (IAM; Kimura and Crow 1964). For KAK, the standardized differences test (Cornuet and Luikart 1996) strongly supports that all loci fit the IAM (*alpha* = 0.00139), which is consistent with the positive Wilcoxon sign test result for KAK (*alpha* = 0.00060) suggesting a recent bottleneck.

Population differentiation: microsatellites and mtDNA

STRUCTURE identified a true *k* of 2 as determined by the Δ*k* plot (**Fig 5c**), but *k* = 2, 3, and 4 are shown for identification of patterns as *k* increases. In all inferred *k*, KAK remained distinct from other sampling groups (*k* = 2, 3, 4; **Fig 6a**). Though STRUCTURE did not detect a completely exclusive genetic cluster, one did dominate in the Kenai Peninsula. All individuals in KAK on average had 75% assignment to the Kenai-dominated cluster; one-third of the group had < 90% assignment to that cluster.

Increasing in *k* to 3 clusters showed that samples from mainland Alaska generally pooled together (NWAK, NAK, CAK, SAK), samples from Kenai Peninsula remained distinct (KAK), and samples from western mainland Canada and Southeast Alaska were grouped (NWY, SEY, BC, SEAK, NU). With *k* = 4, a Southeast Alaska group became apparent (SEAK, BC, part of SEY). In BAPS *a priori*-based analysis, true *k* was 4 with

roughly mainland Alaska (NWAK, NAK, CAK, SAK), Kenai, western mainland Canada (NWY, NU, parts of SEY), and Southeast Alaska (SEAK, BC, parts of SEY) (**Fig 5b**).

The number of mitochondrial haplotypes represented in a population (**Fig 6**) was between 3 and 8 with an average of 4.7. Although sampling may impact these metrics, sufficient sampling in KAK ($N = 25$) yielded few haplotypes (3), while SAK ($N = 33$) yielded more (8). Haplotype 5 was most common, appearing in 9 out of 12 populations (23% of individuals overall), although absent from KAK, RUS, and NAK, which were all generally low in haplotype richness. Private haplotypes were found in RUS (2 haplotypes; 50% of the sampling group), SAK (4 haplotypes; 18%), KAK (1 haplotype; 24%), NWY (2 haplotypes; 20%), BC (1 haplotype; 20%), SEAK (1 haplotype; 3%), NWT (1 haplotype; 7%), and NU (1 haplotype; 29%).

Haplotype 1 is found primarily in RUS (50%) and the Interior Alaskan populations (NWAK 35%; NAK 10%; CAK 11%; SAK 6%; KAK 48%) while being absent in Canadian groups except NU (6% prevalence). Haplotype 9 is unique to CAK, SAK, KAK, and SEAK (CAK 47%; SAK 36%; KAK 28%; SEAK 10%).

Source-sink dynamics and connectivity

Gene flow as estimated in MIGRATE was moderate. There were few cases of extreme asymmetry in gene flow between population pairs, with the exception of KAK generally serving as a source population for several other regions (NAK, SAK, NWY, SEAK), SEY serving as a source population for other regions (RUS, NAK, SAK, BC), and SEAK historically receiving more immigrants (RUS, KAK, BC) and serving as a sink population (**Table 2**). Number of migrants per generation ($N_f m$) ranged from 7.13 to 19.5 (excluding Russia with blocked gene flow since ~11 Kya). Both lowest emigration

(dispersing immigrants) and highest immigration (receiving immigrants) were found in pairwise comparisons with Southeast Alaska (**Table 2**).

Genome structure comparison

Pairwise F_{ST} values were higher for mitochondrial data, ranging from -0.043 to 0.509, than for microsatellites where F_{ST} ranged from -0.006 to 0.265 (**Table 3**). Significant F_{ST} values were 42% of the mitochondrial pairwise comparisons while 53% of microsatellite values were significant. Values were binned into color categories based on F_{ST} ranges of genetic difference for mitochondrial bi-allelic data shown below the diagonal (Hartl and Clark 1997): $F_{ST} < 0.05$ (low), F_{ST} 0.05-0.15 (moderate), F_{ST} 0.15-0.25 (great), F_{ST} 0.25-0.35 (very great), $F_{ST >}$ 0.35 (exceptionally great; scale expanded to include all values). For multi-allelic microsatellite data, bins were rescaled (Hedrick 1999). Bins and scales were assigned after calculating maximum possible global value of F_{ST} (Hedrick 1999) using RecodeData (Meirmans 2006; max value: 0.404) shown above the diagonal. For both microsatellite and mitochondrial data, comparisons that included either RUS or KAK had notably high (though not consistently significant) F_{ST} values.

The mitochondrial AMOVA showed a higher proportion of genetic variance explained among populations (23.78%) relative to microsatellites (2.91%). Direct comparison of genetic diversity revealed a higher proportion of variance explained by mtDNA sequence data than the nuclear microsatellites (**Fig 7**).

Discussion

A comparison of nuclear microsatellite and mitochondrial data revealed both contrasting and consensus perspectives on wolverine population structure. In both, a

genetic discontinuity at the Yukon Territory-Alaska border may reflect historical isolation of these populations. Similarly, another circumboreal mustelid, ermine (*Mustela erminea*) also shows a phylogeographic break near this political border (Dawson et al. 2014; Colella et al. in prep) and in that case, the discontinuity was hypothetically tied to recolonization from different refugia following the LGM. This political border is close to the edge of Beringia, although Swenson and Howard (2005) did not identify a Yukon-Alaska contact zone in their review of glacial breaks in North America. Their review examined relatively few high-latitude hybrid zones, however. Talbot and Shields (1996) found a similar break in brown bear (*Ursus arctos*) lineages between Alaska and the Yukon Territory and posited that the discontinuity reflected differing habitat preferences; open tundra in the west and forests to the northeast of the Alaska-Yukon break. A signature of a spatial discontinuity now documented for several taxa suggests additional phylogeographic investigations of species that span this region are warranted.

Glacial refugia

1) Kenai Peninsula

Kenai was distinctive from all other regions, despite differential patterns of variability across maternal and bi-parentally inherited markers. Mitochondrial DNA showed a trend toward a stable, persistent population (**Fig 4)** of wolverines on the peninsula. A private mtDNA haplotype occurred in 24% of sampled individuals. Nearby South Alaska wolverines showed 'moderate' genetic differentiation, but Kenai was distinctive with an average F_{ST} of 0.33 (very great genetic difference, **Table 3**). Only Russia samples were as distinct across the range of populations examined. Presence of private haplotypes for the peninsular population of wolverines mirrors differentiation

recorded in other large, mobile carnivores of the Kenai Peninsula (e.g. lynx, brown bear; Bailey et al. 1986, Bailey 2002, Morton et al. 2016).

For microsatellite data, Kenai is significantly different from other populations in that it possesses minimal allelic variation, but it does not harbor unique alleles. Average nuclear F_{ST} for Kenai is 0.067, followed by Southeast Alaska (0.054) and North Alaska (0.052), but these North American pairwise comparisons are decisively exceeded by Russia (0.178), which unfortunately is based only on a single individual (**Table 3**). Expansion statistics for the microsatellite DNA showed a severe reduction in effective population size, potentially reflecting a possible founder event and subsequent isolation on the peninsula.

No unique microsatellite alleles were found in Kenai individuals, contrasting with mitochondrial private haplotypes. Because male wolverines typically disperse farther than females (Tomasik and Cook 2005; Wilson et al. 2000), this contrast may reflect limited female dispersal, while males are maintaining gene flow with mainland populations that is reflected in the biparentally inherited nuclear loci. Kenai displays asymmetry in gene flow, acting as a source population in several pairwise comparisons (**Table 2**), but those data should be cautiously interpreted due to variance in sample sizes. Holocene glacial advance of the Portage Glacier (Bartsch-Winkler et al. 1983) at the base of the Kenai Peninsula and the growing metropolitan area of Anchorage may impact exchange between the Kenai population and other nearby populations. An increasing human population on the peninsula (Suburbanstats.org) may also impact these meso-carnivores. More knowledge about Kenai wolverine densities is needed to maintain sustainable harvest (Golden 2011), but harvest numbers remain modest (<15 wolverines

annually on the Kenai Peninsula) and 37% of these are females (Harper and McCarthy 2013). This relatively isolated population could, however, be impacted by habitat perturbation, pathogens, or competitors for a limited resource base (Bangs et al 1982; Crowl et al 2008).

2) Southeast Alaska

Southeast Alaska has been identified as a glacial refuge for various flora and fauna during glaciation periods (Swenson & Howard 2005). In our phylogenetic analysis (**S13-15 Figs**), individuals from Southeast Alaska and Southeast Yukon are grouped using mitochondrial data, and linked to British Columbia populations with similar demographic makeup (see $k = 4$ cluster assignments in **Fig 5**). For mitochondrial data, a multimodal mismatch distribution plot suggests refugial persistence within Southeast Alaska (**Fig 4**). A unique haplotype (Hap 20) made up 3.3% of the Southeast Alaska subsampled population, and mitochondrial F_{ST} values (0.178) for Southeast indicate high genetic distance from other populations (Hartl and Clark 1997). Though possessing a unique haplotype, the region is generally a sink in the directionality of gene flow in pairwise comparisons with other sampling regions (**Table 2**), which may eventually result in genetic swamping of the distinctive population, as hypothesized for the coastal *Martes caurina* which may be swamped by the expanding continental species, *Martes americana* (Stone et al. 2002; Colella et al. in prep). British Columbia has been hypothesized as a source for lower-latitude populations of wolverine in the continental US and lower latitude Canada (Krebs et al. 2004).

Though Southeast was a hypothesized refugium for wolverines, they do not appear in the fossil record with the exception of a single fossil from Prince of Wales

Island in Southeast Alaska that dates from the post-glacial Holocene (13 Kya to present based on substrate recovery; Heaton et al 1996). Microsatellite data for Southeast Alaska demonstrated some signs of long-standing occupation or isolation to the area via expansion plots (**Fig 4**) and high pairwise F_{ST} values (0.054; **Table 3**). Signature of stability (**Fig 4**)**,** a unique cluster assignment for genotypes from this region (**Fig 5**), and a haplotype unique to several sampling regions in the Southeast Alaska coastal area (**Fig 6**) support a divergent population, but the Southeast Alaska population lacks the deep-time divergence necessary to support a refugial population, instead showing evidence of historical persistence of a population to the area.

3) Northwest and Northern Alaska

In other studies of northwestern and northern mainland Alaska, a glacial refugium north of the Brooks Range glacier—disjunct from the rest of Beringia—was hypothesized (*Urocitellus parryii*, Eddingsaas *et al.* 2004; Galbreath *et al.* 2011). Deglaciation models also support refugia existing here (Dyke 2004). In our data, this region exhibits multimodal mismatch distribution peaks illustrating a long-term stable population that is genetically differentiated (F_{ST}; **Table 3**). Mitochondrial data also are consistent with the signature of a refugial population with a new haplotype dominant in North Alaska (50% of the sampled population, Hap 8), while Northwestern Alaska shares a larger percent of its subsampled population with a haplotype common to Russian sampling (35% shared with Russia, 4% with North Alaska). Russia also supports signals of a glacial refugium with multimodal peaks in its mismatch distribution plots (N=6). These data are consistent with the impact of high-latitude Beringia (Russia, Northwestern Alaska, Northern Alaska) as a glacial refuge.

Genome Structure and Marker Comparison

From our comparison, we see high F_{ST} values and greater population structure in the mitochondrial data, however, this result may be due to (1) microsatellite marker selection, (2) an elevated mitochondrial mutation rate compared to the average nuclear mutation rate (Brown 1983), or (3) female philopatry, with higher nuclear gene flow driven by vagile males (Zink et al. 2008). Several previous studies (Chappell et al. 2004; Schwartz et al. 2007, Tomasik and Cook 2005) also showed a higher proportion of variance explained by mtDNA sequence data than bi-parentally inherited nuclear microsatellites.

Conclusions

A population can begin to diverge when impediments to dispersal and mating coalesce in subpopulations (Putman and Carbone, 2014). Identifying geographic structure provides a foundation for conservation, including the identification of critical habitat and barriers to dispersal that could impact evolution across species (Fogelqvist et al. 2010; Palsbøll et al. 2010; Haasl and Payseur 2011). In wolverines of Alaska and Yukon Territory, contemporary populations are relatively connected, but discontinuities were detected: (1) Kenai is singularly distinctive, and (2) discontinuity persists between Alaska and the Yukon Territory. There is not a strong signature of a historically persistent refugial population of wolverines in Southeast Alaska, but gene flow between these coastal wolverines and other populations is limited. Signature of stability from a refugium exists in Northwest Alaska and North Alaska and is accompanied by wolverine fossil evidence supporting the former existence of a Beringian glacial refuge.

The distinctiveness of the Kenai Peninsula population may be due to a founder event following glacial retreat, and subsequent isolation caused by peninsular effects. The difference between maternal and nuclear markers may be a product of wolverine life history with further-dispersing males promoting nuclear gene flow relative to philopatric females and mitochondrial (matrilineal-inherited) structure. To explain the Alaska-Yukon disjunction, a study of finer-scale landscape genetics would better inform the hypotheses that (a) habitat shifts between Alaska and Yukon caused this structure, or (b) a reconvening of lineages separated by glacial maxima is shown. In a study of the dispersal habits of wolverines, Dalerum *et al.* (2007) sampled individuals in northwestern Alaska separated by sex and analyzed them for dispersal distance and genetic difference from surrounding individuals. Sex-biased dispersal behaviors within northwest Alaska were defined as non-significant, perhaps due to resource scarcity in the tundra region that motivated typically philopatric females to disperse greater distances over the course of their lifetime (i.e., comparable to males). As a future direction, a similar intensive study conducted east of the Alaska-Yukon border may provide insight to the role of habitat differences on wolverine behavior, movement, and ultimately genetic structure. Future work should examine this effect on dispersal, and the bias of sex on dispersal, in the boreal and mountain forest habitat of Yukon compared to the tundra individuals of Alaska.

Literature Cited

Abbott, R. J., L. C. Smith, R. I. Milne, R. M. M. Crawford, K. Wolff, and J. Balfour. 2000. Molecular analysis of plant migration and refugia in the Artic. Science 289: 1343–1346.

Anderson, E. C., and K. K. Dunham. 2008. The influence of family groups on inferences made with the program Structure. Molecular Ecology Resources 8(6): 1219–1229.

Bailey, T. 2002. Genetic research reveals unique characteristics of Kenai lynx population. Refuge Notebook 4(13): 24–25.

Bailey, T. N., E. E. Bangs, M. F. Portner, J. C. Malloy, and R. J. McAvinchey. 1986. An apparent overexploited lynx population on the Kenai Peninsula, Alaska. The Journal of Wildlife Management 50(2): 279–290.

Bangs, E. E., T. H. Spraker, T. N. Bailey, and V. D. Berns. 1982. Effects of increased human populations on wildlife resources of the Kenai Peninsula, Alaska. US Fish and Wildlife Publications 36: 605–616.

Barrie, J.V. and K. W. Conway. 1999. Late Quaternary glaciation and postglacial stratigraphy of the northern Pacific margin of Canada. Quaternary Research 51(2): 113–123.

Bartsch-Winkler, S., A. T. Ovenshine, and R. Kachadoorian. 1983. Holocene history of the estuarine area surrounding Portage, Alaska as recorded in a 93 m core. Canadian Journal of Earth Sciences 20(5): 802–820.

Beerli, P. 1998. Estimation of migration rates and population sizes in geographically structured populations. Advances in Molecular Ecology 306, 39–54.

Beerli, P. 2002. MIGRATE: documentation and program, part of LAMARC. Department of Genome Sciences, University of Washington, Seattle.

Beerli, P., and J. Felsenstein. 1999. Maximum-likelihood estimation of migration rates and effective population numbers in two populations using a coalescent approach. Genetics: 152(2): 763–773.

Beerli, P. and J. Felsenstein. 2001. Maximum likelihood estimation of a migration matrix and effective population sizes in *n* subpopulations by using a coalescent approach. Proceedings of the National Academy of Sciences 98(8): 4563–4568.

Belkhir K., V. Castric, and F. Bonhomme. 2002. IDENTIX, a software to test for relatedness in a population using permutation methods. Molecular Ecology Notes 2(4): 611–614.

Benson, D. A., I. Karsch-Mizrachi, D. J. Lipman, J. Ostell, and D. L. Wheeler. 2009. Database issue. GenBank, Nucleic Acids Res 37(10): 26–31.

Bergl, R. A., and L. Vigilant. 2007. Genetic analysis reveals population structure and recent migration within the highly fragmented range of the Cross River gorilla (*Gorilla gorilla diehli*). Molecular Ecology 16: 501–516.

Bouckaert, R., J. Heled, D. Kühnert, T. Vaughan, C. H. Wu, D. Xie, M. A. Suchard, A. Rambaut, and A. J. Drummond. 2014. BEAST 2: a software platform for Bayesian \ evolutionary analysis. PLoS Computational Biology 10(4): e1003537.

Brown, W. M. 1983. Evolution of animal mitochondrial DNA. In: M. Nei, R. K. Koehn (eds). Evolution of genes and proteins. Sinauer, Sunderland, MA. 62–88.

Carrara, P. E., T. A. Ager, and J. F. Baichtal. 2007. Possible refugia in the Alexander Archipelago of southeastern Alaska during the late Wisconsin glaciation. Canadian Journal of Earth Sciences 44: 229–244.

Carroll, C., R. F. Noss, and P. C. Paquet. 2001. Carnivores as focal species for conservation planning. Ecological Applications 11(4): 961–980.

Cegelski, C. C., L. P. Waits, N. J. Anderson, O. Flagstad, C. Strobeck, and C. J. Kyle. 2006. Genetic diversity and population structure of wolverine (*Gulo gulo*) populations at the southern edge of their current distribution in North America with Implications for genetic viability. Conservation Genetics 7(2): 197–211.

Chadwick, D. H. 2010. The Wolverine Way. Ventura, CA: Patagonia Books.

Chappell, D. E., R. A. Van Den Bussche, J. Krizan, and B. Patterson. 2004. Contrasting levels of genetic differentiation among populations of wolverines (*Gulo gulo*) from northern Canada revealed by nuclear and mitochondrial loci. Conservation Genetics 5(6): 759–767.

Colella JP, T. Lan, C. Lindqvist, S. L. Talbot, J. A. Cook. In prep. Mitogenomic phylogenetics of Mustela erminea (Family Mustelidae), investigating the impact of metabolic selection on phylogenetics. *submitted*.

Cook, J.A., N. G. Dawson, and S. O. MacDonald. 2006. Conservation of highly fragmented systems: the north temperate Alexander Archipelago. Biological Conservation 133(1): 15pp.

Cook, J. A., et al. 2017. The Beringian Coevolution Project: holistic collections of mammals and associated parasites reveal novel perspectives on evolutionary and environmental change in the North. Arctic Science 0(ja): 56pp.

Copeland, J. P., K. S. McKelvey, K. B. Aubry, A. Landa, J. Persson, R. M. Inman, J. Krebs, E. Lofroth, H. Golden, J.R. Squires, A. Magoun, M. K. Schwartz, J. Wilmot, C. L. Copeland, R. E. Yates, I. Kojola, R. May. 2010. The bioclimatic envelope of the wolverine (*Gulo gulo*): Do climatic constraints limit Its geographic distribution? Canadian Journal of Zoology 88(3): 233–46.

Corander, J. and P. Marttinen. 2006. Bayesian identification of admixture events using

multilocus molecular markers. Molecular Ecology 15(10): 2833–2843.

Corander, J., M. Gyllenberg, and T. Koski. 2006. Bayesian model learning based on a parallel MCMC strategy. Statistics and Computing 16(4): 355–362.

Cornuet, J. M. and G. Luikart. 1996. Description and power analysis of two tests for detecting recent population bottlenecks from allele frequency data. Genetics 144(4): 2001–2014.

Crowl, T. A., T. O. Crist, R. R. Parmenter, G. Belovsky, and A. E. Lugo. 2008. The spread of invasive species and infectious disease as drivers of ecosystem change. Frontiers in Ecology and the Environment 6(5): 238–246.

Dalerum, F., J. Loxterman, B. Shults, K. Kunkel, and J. A. Cook. 2007. Sex-specific dispersal patterns of wolverines: insights from microsatellite markers. Journal of Mammalogy 88(3): 793–800.

Dallas, J. F., and S. B. Piertney. 1998. Microsatellite primers for the Eurasian otter. Molecular Ecology 7(9): 1248–1251.

Darriba, D., G. L. Taboada, R. Doallo, and D. Posada. 2012. jModelTest 2: more models, new heuristics and parallel computing. Nature Methods 9(8): 772–772.

Davis, C. S., and C. Strobeck. 1998. Isolation, variability, and cross-species amplification of polymorphic microsatellite loci in the family Mustelidae. Molecular Ecology 7(12): 1776–1778.

Dawson, N. G., A. G. Hope, S. L. Talbot, and J. A. Cook. 2014. A multilocus evaluation of ermine (*Mustela erminea*) across the Holarctic, testing hypotheses of Pleistocene diversification in response to climate change. Journal of biogeography 41(3): 464–475.

Di Rienzo, A., A. C. Peterson, J. C. Garzat, A. M. Valdes, M. Slatkint, and N. B. Freimer. 1994. Mutational processes of simple-sequence repeat loci in human populations. Proceedings of the National Academy of Science Genetics 91: 3166–3170.

Dias, P. C. 1996. Sources and sinks in population biology. Trends in Ecology and Evolution 11(8): 326–330.

Duffy, A. J., A. Landa, M. O'Connell, C. Stratton, and J. M. Wright. 1998. Four polymorphic microsatellites in wolverine, *Gulo gulo*. Animal genetics 29: 63–72.

Dyke, A. S. 2004. An outline of North American deglaciation with emphasis on central and northern Canada. Developments in Quaternary Sciences 2(Part B): 373–424.

Eddingsaas, A. A., B. K. Jacobsen, E. P. Lessa, and J. A. Cook. 2004. Evolutionary

history of the arctic ground squirrel (*Spermophilus parryii*) in Nearctic Beringia. Journal of Mammalogy 85(4): 601–610.

Evanno, G., S. Regnaut, and J. Goudet. 2005. Detecting the number of clusters of individuals using the software STRUCTURE: a simulation study. Molecular Ecology 14(8): 2611–2620.

Excoffier, L., H. E. Lischer. 2010. Arlequin suite ver. 3.5: a new series of programs to perform population genetics analyses under Linux and Windows. Molecular Ecology Resources 10(3): 564–567.

Falush, D., M. Stephens, and J. K. Pritchard. 2003. Inference of population structure using multilocus genotype data: linked loci and correlated allele frequencies. Genetics 164(4): 1567–1587.

Fedorov, V.B. and N. C. Stenseth. 2002. Multiple glacial refugia in the North American Arctic: inference from phylogeography of the collared lemming (*Dicrostonyx groenlandicus*). Proceedings of the Biological Sciences, The Royal Society B, 269(1505): 2071–2077.

Fleming, M. A., and J. A. Cook. 2002. Phylogeography of endemic ermine (*Mustela erminea*) in southeast Alaska. Molecular Ecology 11(4): 795–807.

Fleming, M. A., E. A. Ostrander, and J. A. Cook. 1999. Microsatellite markers for American mink (*Mustela vison*) and ermine (*Mustela erminea*). Molecular Ecology 8(8): 1352–1355.

Fogelqvist, J., A. Niittyvuopio, J. Ågren, O. Savolainen, and M. Lascoux. 2010. Cryptic population genetic structure: the number of inferred clusters depends on sample size. Molecular ecology resources 10(2): 314–323.

Francés, J. A. 2008 Masters thesis, University of New Mexico, Albuquerque, New Mexico, USA. 51pp.

Fu, Y. X. 1997. Statistical tests of neutrality of mutations against population growth, hitchhiking, and background selection. Genetics Society of America 147: 915–925.

Galbreath, K. E., J. A. Cook, A. A. Eddingsaas, and E. G. DeChaine. 2011. Diversity and demography in Beringia: multilocus tests of paleodistribution models reveal the complex history of arctic ground squirrels. Evolution 65(7): 1879–1896.

Garza, J. C., and E. G. Williamson. 2001. Detection of reduction in population size using data from microsatellite loci. Molecular ecology 10(2): 305–318.

Gentili, R., C. Baroni, M. Caccianiga, S. Armiraglio, A. Ghiani, and S Citterio, S. (2015).

Potential warm-stage microrefugia for alpine plants: Feedback between geomorphological and biological processes. Ecological Complexity 21: 87–99.

Golden, H.N. 2011. Wolverine abundance in upper Turnagain Arm and the Kenai Mountains with Emphasis on helicopter-skiing permit areas. Alaska Department of Fish and Game and CNF Interagency Collaborative Project Progress Report, Anchorage.

Goudet, J. 1993. The genetics of geographically structured populations. Doctoral dissertation, University College of North Wales, Bangor, North Wales.

Goudet, J., T. De Meeüs, A. J. Day, and C. J. Gliddon. 1994. Gene flow and population structure. Genetics and evolution of aquatic organisms. London: Chapman & Hall, 81–95.

Goudet, J. 1995. FSTAT (version 1.2): a computer program to calculate F-statistics. Journal of Heredity 86(6): 485–486.

Haasl, R. J., and B. A. Payseur. 2011. Multi-locus inference of population structure: a comparison between single nucleotide polymorphisms and microsatellites. Heredity 106(1) 158–171.

Harlin-Cognato, A., J. W. Bickham, T. R. Loughlin, and R. L. Honeycutt. 2006. Glacial refugia and the phylogeography of Steller's sea lion (*Eumatopias jubatus*) in the North Pacific. Journal of Evolutionary Biology 19(3): 955–969.

Harper, P. and L. A. McCarthy, editors. 2013. Furbearer management report of survey-inventory activities, 1 July 2009-30 June 2012. Alaska Department of Fish and Game, Species Management Report ADF&G/DWC/SMR-2013-5, Juneau.

Hartl, D. L., A. G. Clark, and A. G. Clark. 1997. Principles of population genetics. Vol. 116. Sunderland: Sinauer associates.

Heaton, T. H., S. L. Talbot, and G. F. Shields. 1996. An ice age refugium for large mammals in the Alexander Archipelago, Southeastern Alaska. Quaternary Research 46: 186–192.

Hedrick, P. W. 1999. Perspective: highly variable loci and their interpretation in evolution and conservation. Evolution 53(2): 313–318.

Heled, J., 2014. Extended Bayesian Skyline Plot tutorial for BEAST 2.

Heled, J. and A. J. Drummond. 2010. Bayesian inference of species trees from multilocus data. Molecular Biology and Evolution 27(3): 570–580.

Hoffmann, R. S. 1981. Different voles for different holes: environmental restrictions on

refugial survival of mammals. In Evolution today (G.G.E. Scudder and J.L. Reveal, eds.). Proceedings of the Second International Congress of Systematic and Evolutionary Biology. 25–45

Hope, A. G., E. Waltari, N. E. Dokuchaev, S. Abramov, T. Dupal, A. Tsvetkova, H. Henttonen, S. O. MacDonald, and J. A. Cook. 2010. High-latitude diversification within Eurasian least shrews and Alaska tiny shrews (Soricidae). Journal of Mammalogy 91(5): 1041–1057.

Hope, A.G., E. Waltari, V. B. Fedorov. A. V. Goropashnaya, S. L. Talbot, and J. A. Cook. 2011. Persistence and diversification of the Holarctic shrew, *Sorex tundrensis* (Family Soricidae), in response to climate change. Molecular Ecology 20(20): 4346–4370.

Hope, A. G., N. Takebayashi, K. E. Galbreath, S. L. Talbot, and J. A. Cook. 2013. Temporal, spatial and ecological dynamics of speciation among amphi-Beringian small mammals. Journal of Biogeography 40(3): 415–429.

Hope, A. G., S. Y. W. Ho, J. L. Malaney, J. A. Cook, and S. L. Talbot. 2014. Accounting for rate variation among lineages in comparative demographic analyses. Evolution 68(9): 2689–2700.

Hornocker, M. G., and H. S. Hash. 1981. Ecology of the wolverine in northwestern Montana. Canadian Journal of Zoology 59(7): 1286–1301.

Hultén, Eric. 1937. Outline of the history of Arctic and boreal biota during the Quaternary Period: their evolution during and after the glacial period as Indicated by the equiformal progressive areas of present plant species. Stockholm: Bokförlags aktiebolaget Thule.

Jakobsson, M., and N. A. Rosenberg. 2007. CLUMPP : a cluster matching and permutation program for dealing with label switching and multimodality in analysis of population structure. Bioinformatics 23(14): 1801–1806.

Josenhans, H. W., D. W. Fedje, K. W. Conway, and J.V. Barrie. 1995. Post glacial sea levels on the western Canadian continental shelf: evidence for rapid change, extensive subaerial exposure, and early human habitation. Marine Geology 125(1–2): 73–94.

Kalinowski, S. T. 2005. hp-rare 1.0: a computer program for performing rarefaction on measures of allelic richness. Molecular Ecology Notes 5(1): 187–189.

Kearse, M., R. Moir, A. Wilson, S. Stones-Havas, M. Cheung, S. Sturrock, S. Buxton, A. Cooper, S. Markowitz, C. Duran, T. Thierer, B. Ashton, P. Meintjes, and Alexei Drummond. 2012. Geneious Basic: an integrated and extendable desktop software

platform for the organization and analysis of sequence data. Bioinformatics 28(12): 1647–1649.

Kimura, M., and J. F. Crow. 1964. The number of alleles that can be maintained in a finite population. Genetics 49(4): 725–738.

Knowles, L.L. et al. 2016. Quantifying the similarity between genes and geography across Alaska's alpine small mammals. Journal of Biogeography 43(7): 1464–1476.

Krebs, J. A., E. C. Lofroth, J. P. Copeland, V. Banci, D. A. Cooley, H. N. Golden, A. J. Magoun, R. Mulders, and B. Shults. 2004. Synthesis of survival rates and causes of mortality in North American wolverines. Journal of Wildlife Management 68(3): 493–502.

Kyle, C. J., and C. Strobeck. 2002. Connectivity of peripheral and core populations of North American wolverines. Journal of Mammalogy 83(4): 1141–1150.

Laliberte, A. S., and W. J. Ripple. 2004. Range contractions of North American carnivores and ungulates. BioScience 54(2): 123–138.

Librado, P., and J. Rozas. 2009. DnaSP v5: a software for comprehensive analysis of DNA polymorphism data. Bioinformatics 25(11): 1451–1452.

Mandryk, C. A., H. Josenhans, D. W. Fedje, and R. W. Mathewes. 2001. Late Quaternary paleoenvironments of Northwestern North America: implications for inland versus coastal migration routes. Quaternary Science Reviews 20(1): 301–314.

McLean, B. S., D. J. Jackson, J. A. Cook. 2016. Rapid divergence and gene flow at high latitude shape the history of Holarctic ground squirrels (Urocitellus). Molecular Phylogenetics and Evolution 102:174–188.

Meirmans, P.G., 2006. Using the AMOVA framework to estimate a standardized genetic differentiation measure. Evolution 60(11): 2399–2402.

Morton, J. M., G. C. White, G. D. Hayward, D. Paetkau, and M. P. Bray. 2016. Estimation of the brown bear population on the Kenai Peninsula, Alaska. The Journal of Wildlife Management 80(2): 332–346.

Oetting, W. S., H. K. Lee, D. J. Flanders, G. L. Wiesner, T. A. Sellers, R. A. King. 1995. Linkage analysis with multiplexed short tandem repeat polymorphisms using infrared fluorescence and M13 tailed primers. Genomics 30(3): 450–458.

Ohta, T., and M. Kimura. 1973. A model of mutation appropriate to estimate the number of electrophoretically detectable alleles in a finite population. Genetical Research 22(2): 201–204.

Palsbøll, P. J., M. Z. Peery, and M. Berube. 2010. Detecting populations in the 'ambiguous' zone: kinship-based estimation of population structure at low genetic divergence. Molecular Ecology Resources 10(5): 797–805.

Park, S. D. E. 2001. The Excel microsatellite toolkit. *Trypanotolerance in West African Cattle and the Population Genetic Effects of Selection*.

Piry, S., G. Luikart, and J. M. Cornuet. 1999. Bottleneck: a computer program for detecting recent reductionsin the effective population size using allele frequency data. Journal of Heredity 90(4): 502–503.

Pritchard, J. K., M. Stephens, and P. Donelly. 2000. Inference of population structure using multilocus genotype data. Genetics 155(2): 945–59.

Pritchard, J. K., and W. Wen. 2004. Documentation for STRUCTURE 2.0.

Putman, A. I., and I. Carbone. 2014. Challenges in analysis and interpretation of microsatellite data for population genetic studies. Ecology and Evolution 4(22), 4399–4428.

Queller, D. C., and K. F. Goodnight. 1989. Estimating relatedness using genetic markers. Evolution 43(2): 258–275.

Raymond, M., and F. Rousset. 1995. An exact test for population differentiation. Society for the Study of Evolution 49(6): 1280–1283.

Rice, W. R. 1989. Analyzing tables of statistical tests. Evolution 43(1): 223–225.

Rico, Y., J. Morris-Pocock, J. Zigouris, J. J. Nocera, and C. J. Kyle. 2015. Lack of spatial immunogenetic structure among wolverine (*Gulo gulo*) populations suggestive of broad scale balancing selection. *PloS one*, *10*(10), e0140170.

Rowe, K.C., K. M. C. Rowe, M. W. Tingley, M. S. Koo, J. L. Patton, C. J. Conroy, J. D. Perrine, S. R. Beissinger, C. Moritz. 2014. Spatially heterogeneous impact of climate change on small mammals of montane California. Proceedings of the Royal Society B: Biological Sciences 282(1799): 10pp.

Rozas, J., P. Librado, J. C. Sánchez-Del Barrio, X. Messeguer, and R. Rozas. 2010. DnaSP version 5 help contents [Help File]. *Available with the program at http://www.ub.edu/dnasp*.

Scheffers, B. R., L. De Meester, T. C. Bridge, A. A. Hoffmann, J. M. Pandolfi, R. T. Corlett, S. H. M. Butchart, P. Pearce-Kelly, K. M. Kovacs, D. Dudgeon, M. Pacifici, C. Rondinini, W. B. Foden, T. G. Martin, C. Mora, D. Bickford, J. E. M. Watson. 2016. The broad footprint of climate change from genes to biomes to people. Science, 354(6313), 719–732.

Schwartz, M. K., K. B. Aubry, K. S. McKelvey, K. L. Pilgrim, J. P. Copeland, J. R. Squires, R. M. Inman, S. M. Wisely, and L. F. Ruggiero. 2007. Inferring geographic isolation of wolverines in California using historical DNA. Journal of Wildlife Management 71(7): 2170–2179.

Shafer, A., C. I. Cullingham, S. D. Cote, and D. W. Coltman. 2010. Of glaciers and refugia: a decade of study sheds new light on the phylogeography of northwestern North America. Molecular Ecology 19(21): 4589–4621.

Stone, K. D., R. W. Flynn, and J. A. Cook. 2002. Post-glacial colonization of northwestern North America by the forest-associated American marten (*Martes americana*, Mammalia: Carnivora: Mustelidae). Molecular Ecology 11(10): 2049–2063.

Suburbanstats.org. "Current Kenai Peninsula Borough, Alaska Population, Demographics and Stats in 2016, 2017." *SuburbanStats.org*. US Census Bureau, 2017.

Swenson, N. G., and D. J. Howard. 2005. Clustering of contact zones, hybrid zones, and phylogeographic breaks in North America. The American Naturalist 166(5): 581–591.

Tajima, F. 1989. Statistical method for testing the neutral mutation hypothesis by DNA polymorphism. Genetics 123(3): 585–595.

Talbot, S. L., and G. F. Shields. 1996. Phylogeography of brown bears (*Ursus arctos*) of Alaska and paraphyly within the Ursidae. Molecular phylogenetics and evolution 5(3): 477–494.

Tomasik, E., and J. A. Cook. 2005. Mitochondrial phylogeography and conservation genetics of wolverine (*Gulo gulo*) of northwestern North America. Journal of Mammalogy 86(2): 386–396.

Van Oosterhout, C., W.F. Hutchinson, D.P. M. Wills, P. Shipley. 2004. MICRO-CHECKER : Software for identifying and correcting genotyping errors in microsatellite data. Molecular Ecology Notes 4(3): 535–538.

Walker, C. W., C. Vilà, A. Landa, M. Lindén, and H. Ellegren. 2001. Genetic variation and population structure in Scandinavian wolverine (*Gulo gulo*) populations. Molecular Ecology 10(1): 53–63.

Weir, B.S., and C. C. Cockerham. 1984. Estimating F-statistics for the analysis of population structure. Evolution 38(6): 1358–1370.

Weksler, M., H. C. Lanier, and L. E. Olson. 2010. Eastern Beringian biogeography: historical and spatial genetic structure of singing voles in Alaska. Journal of

Biogeography 37(8): 1414–1431.

Wilson, G. M., R. A. Van Den Bussche, P. K. Kennedy, A. Gunn, and K. Poole. 2000. Genetic variability of wolverines (*Gulo gulo*) from the Northwest Territories, Canada: conservation implications. Journal of Mammalogy 81(1): 186–196.

Wright, S. 1921. Systems of mating. I. The biometric relations between parent and offspring. Genetics 6(2): 111–123.

Wright, S. 1951. The genetical structure of populations. Annals of Eugenics 15: 323–354.

Wright, S. (1965). The interpretation of population structure by F-statistics with special regard to systems of mating. *Evolution*, *19*(3), 395–420.

Zigouris, J., J. A. Schaefer, C. Fortin, C. J. Kyle. 2013. Phylogeography and post-glacial recolonization in wolverines (*Gulo gulo*) from across their circumpolar distribution. PLoS ONE 8(12): e83837.

Zink, R. M., and G. F. Barrowclough. 2008. Mitochondrial DNA under siege in avian phylogeography. Molecular Ecology 17(9): 2107–2121.

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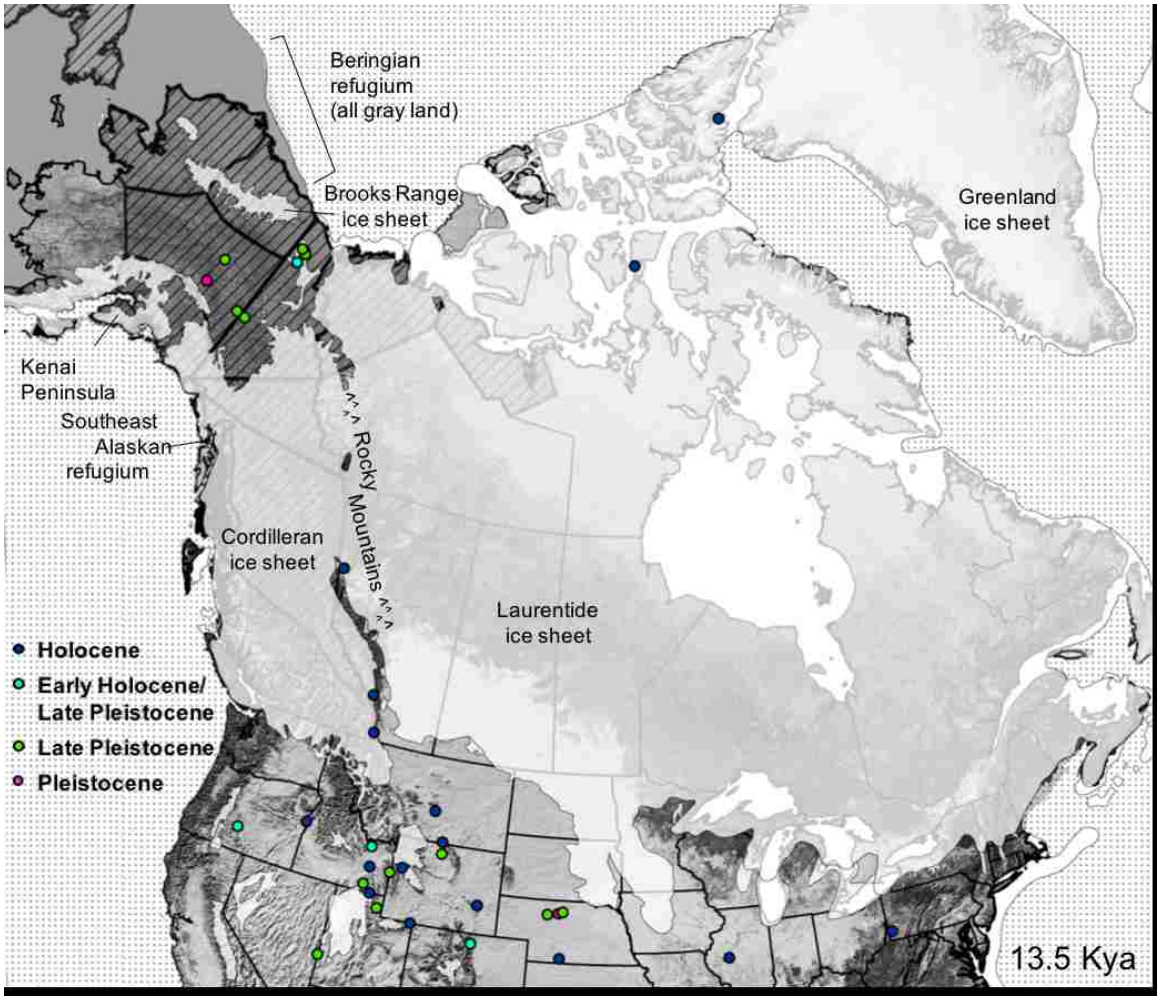


Figure 1: Wolverine fossil records in North America and extent glaciation at 13.5 Kya. Ice is denoted in transparent white, land is shaded topographically with glacially exposed Beringia in gray and oceans dotted. Extent of wolverine study area is hatched (Alaska/western Canada). Last glacial maximum (LGM) was 24–13 Kya. Glacial cycles left Beringia as an ice-free refugium at least 25 Kya, Southeast Alaska and the Yukon-Rocky Mountain pass opening up around 15 Kya, and high-latitude eastern Canada became deglaciated as recently as 8 Kya. Kenai Peninsula shows areas of deglaciation as early as 13 Kya, and the Brooks Range ice sheet fragmented Alaska from 24–13.5 Kya (glacial outlines from Dyke 2004). The wolverine fossil record aligns well with this glacial progression. Though Southeast is a hypothesized refugium for wolverines, they do not appear in the fossil record until the early post-glacial period in Southeast Alaska (13 Kya to present; Heaton et al. 1996).

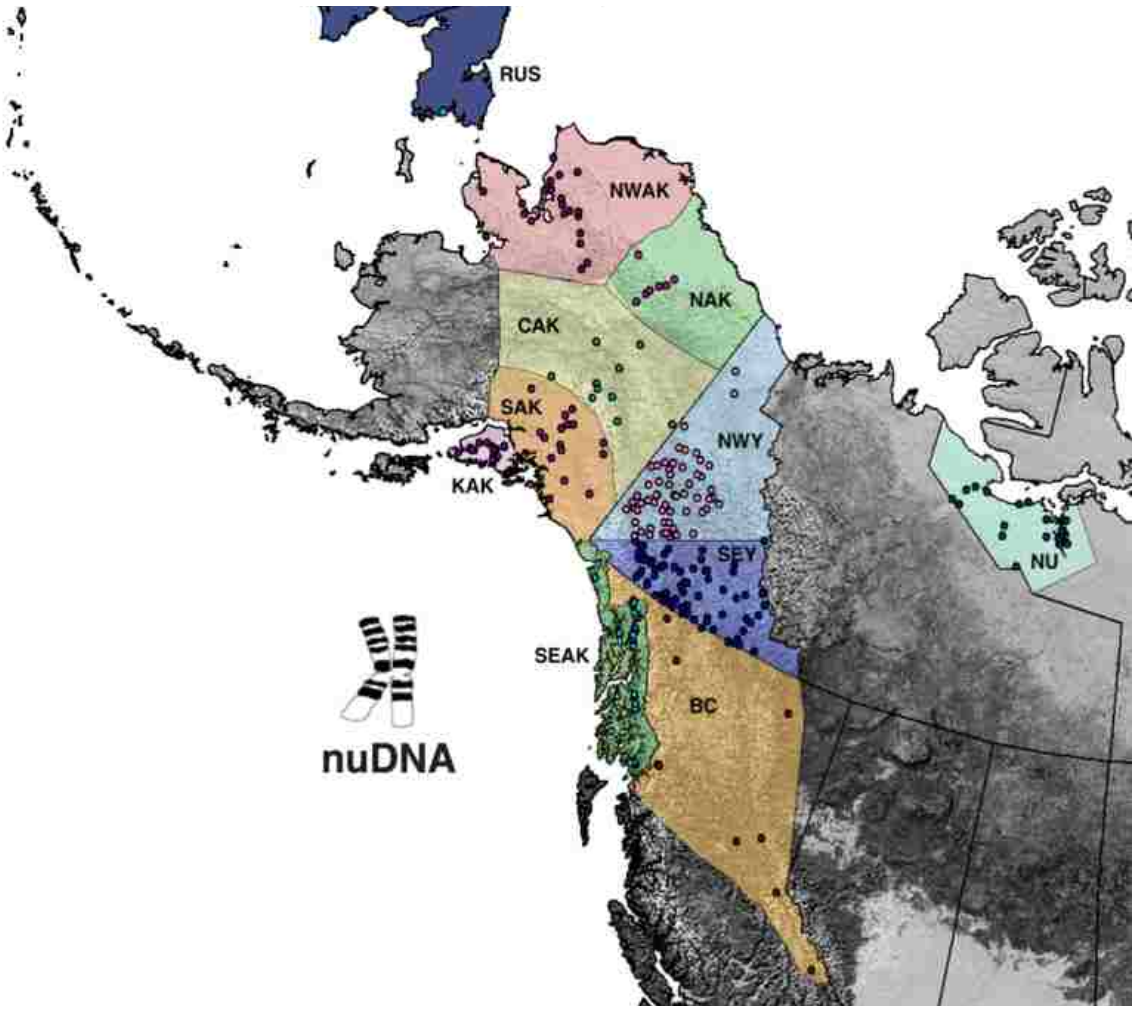


Figure 2: Wolverine sampling showing geographic acronyms for microsatellites (nuclear genome). RUS=Russia (*N*=1; 20 loci), NWAK=Northwest Alaska (*N*=30; 10 loci), NAK=North Alaska (*N*=9; 20 loci), CAK=Central Alaska (*N*=13, 20 loci), SAK=South Alaska (*N*=17; 20 loci), KAK=Kenai Peninsula (*N*=26; 20 loci), NWY=Northwest Yukon (*N*=30; 20 loci), SEY=Southeast Yukon (*N*=30; 20 loci), BC=British Columbia (*N*=9; 20 loci), SEAK=Southeast Alaska (*N*=26; 20 loci), NU=Nunavut (*N*=16; 20 loci).

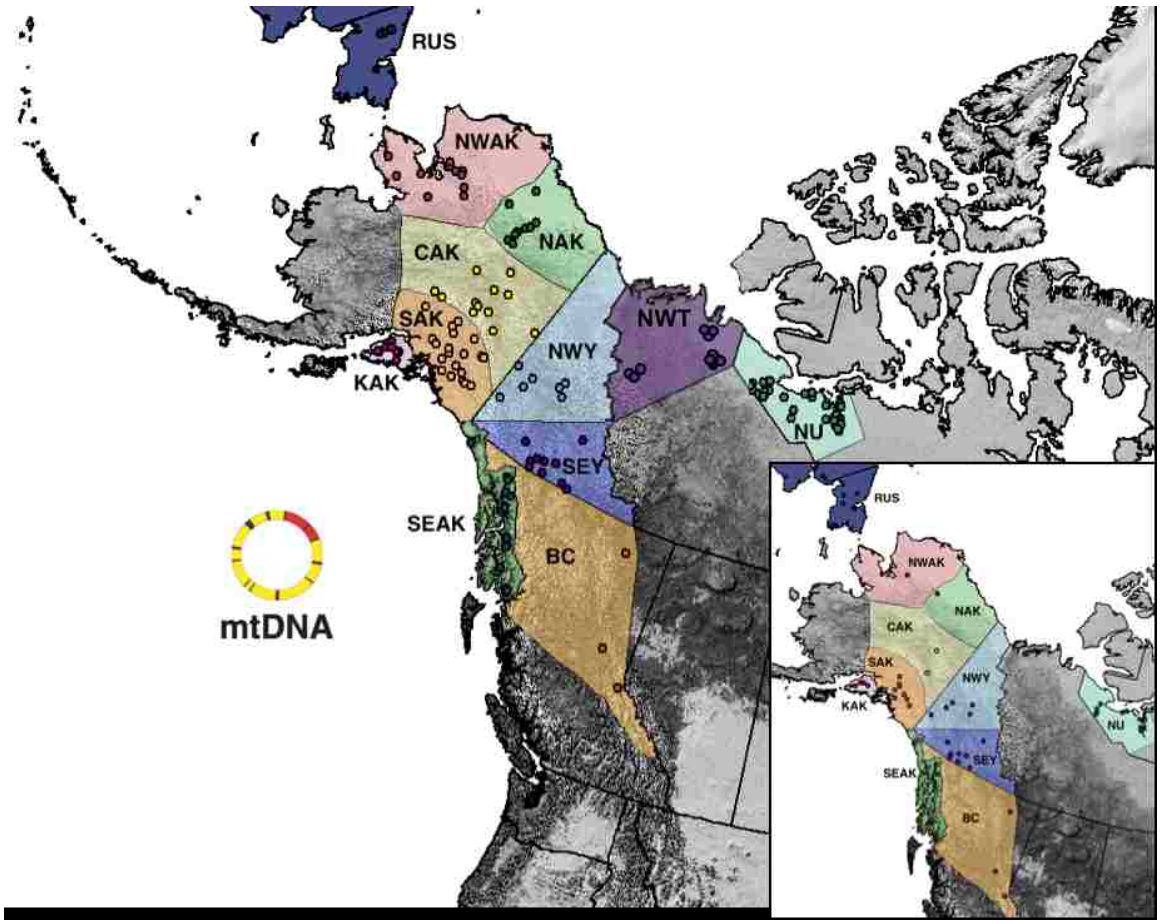


Figure 3: Wolverine sampling showing geographic acronyms for mitochondrial control region (*N*=252, 366bp) and cytochrome *b* (*N*=67, 1140bp). Larger map displays control region sequences, inset shows cytochrome *b* sequences. Number of samples (*N*) presented as (control region; cytochrome *b*): RUS = Russia (6; 6), NWAK = Northwest Alaska (23; 3), NAK = North Alaska (10; 1), CAK = Central Alaska (19; 3), SAK = South Alaska (33; 7), KAK = Kenai Peninsula (25; 7), NWY = Northwest Yukon (10; 7), SEY = Southeast Yukon (13; 9), BC = British Columbia (5; 3), SEAK = Southeast Alaska (30; 10), NWT = Northwest Territories (15; 0), NU = Nunavut (63; 11).

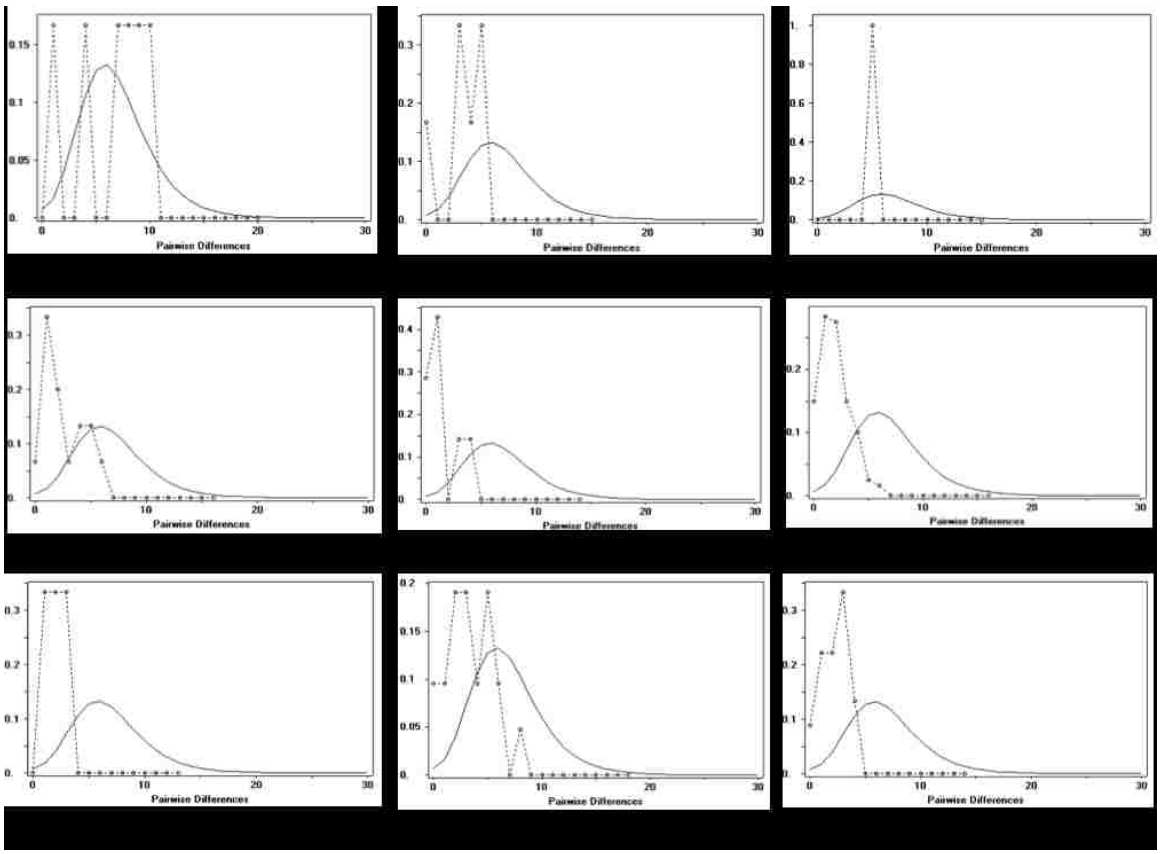


Figure 4: Population expansion graphs for concatenated mitochondrial genes cytochrome *b* and control region (1507bp). The solid line indicates the pattern for expectation and dotted lines depict the actual demographic history of our data. Bimodal or multimodal patterns indicate stable populations. Sample size for each population: RUS 4, NWAK/NAK 4, CAK 2, SAK 6, KAK 7, YT 16, BC 3, SEAK 7, NU 10.

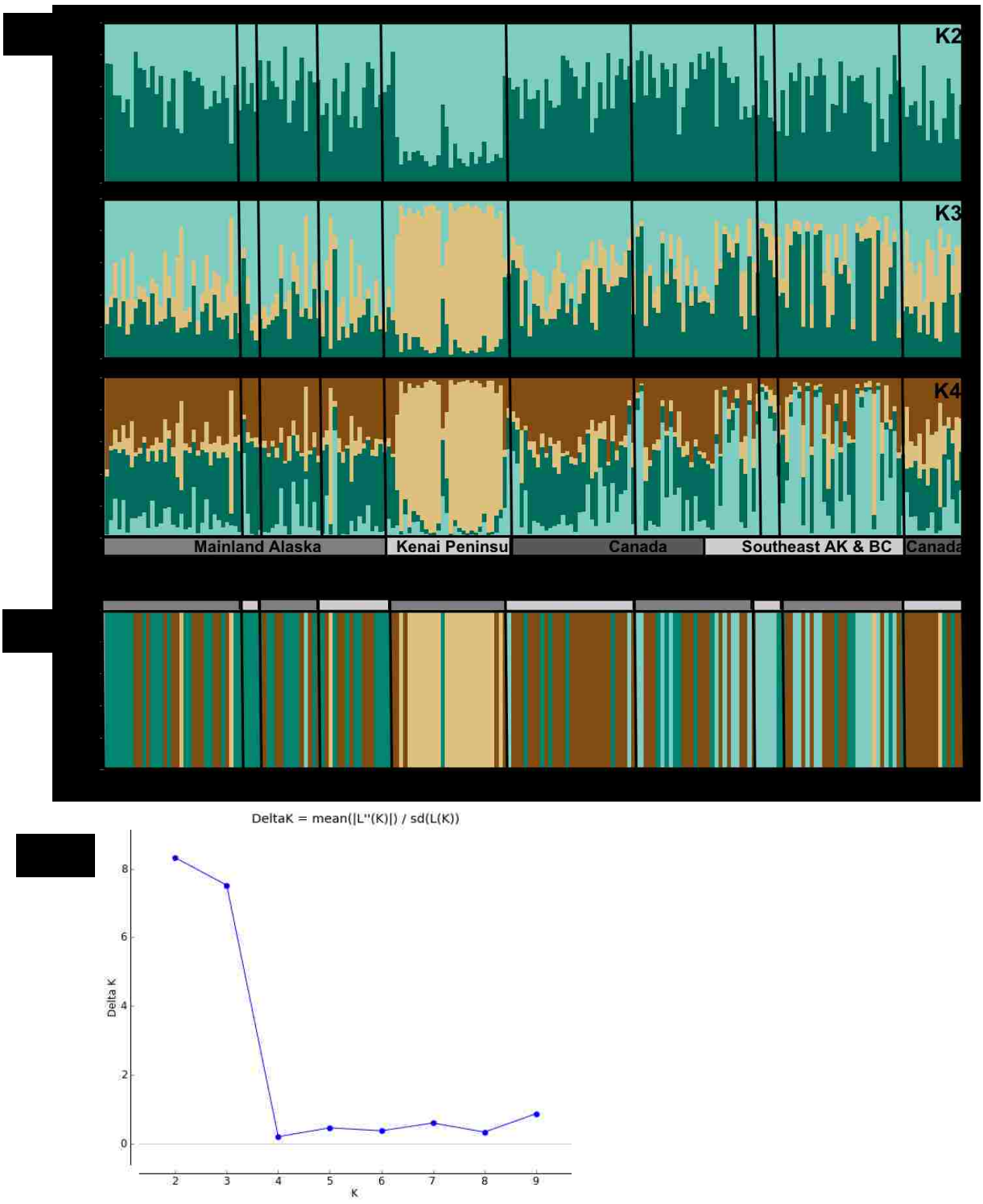


Figure 5: Patterns of microsatellite genetic variation across the sampled regions: (a) STRUCTURE barplots of population membership scores for an inferred *k* of 2 through 4 genetic clusters with true *k* being 2; (b) BAPS barplot for population membership with true $k = 4$; and (c) delta k for STRUCTURE $k = 2$.

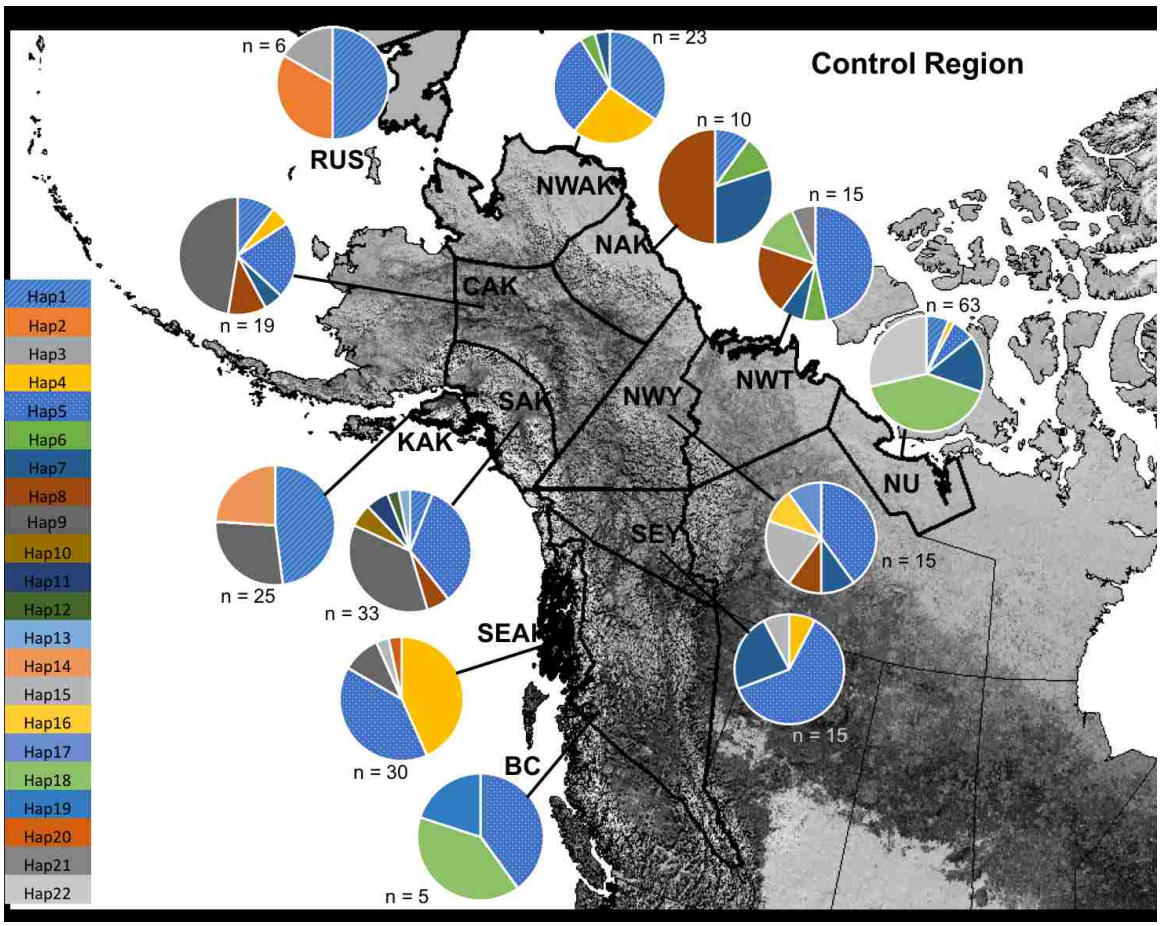


Figure 6: Frequency distribution of control region (mtDNA) haplotypes for each sampled region.

Figure 7: Genomic comparison of the nuclear (microsatellites) and mitochondrial (control region) genomes. F_{ST} and phiST are measures of population differentiation due to genetic structure. There is less gene flow than expected among microsatellites.

List of Tables

N sample size for each analysis; *H* no. of haplotypes; H_P no. of private haplotypes, *Hd* haplotype diversity; Tajima's D, Fu's F_S; microsatellite loci no. for each population; H_O observed heterozygosity; H_E expected heterozygosity; R_A rarefied allelic richness, R_A private allelic richness, F_{IS} inbreeding coefficient.

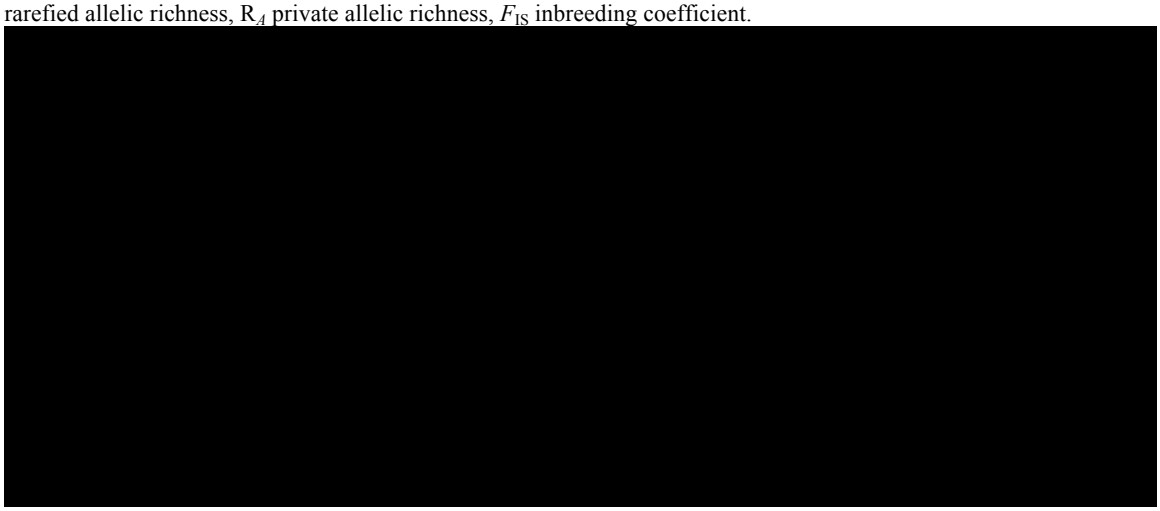
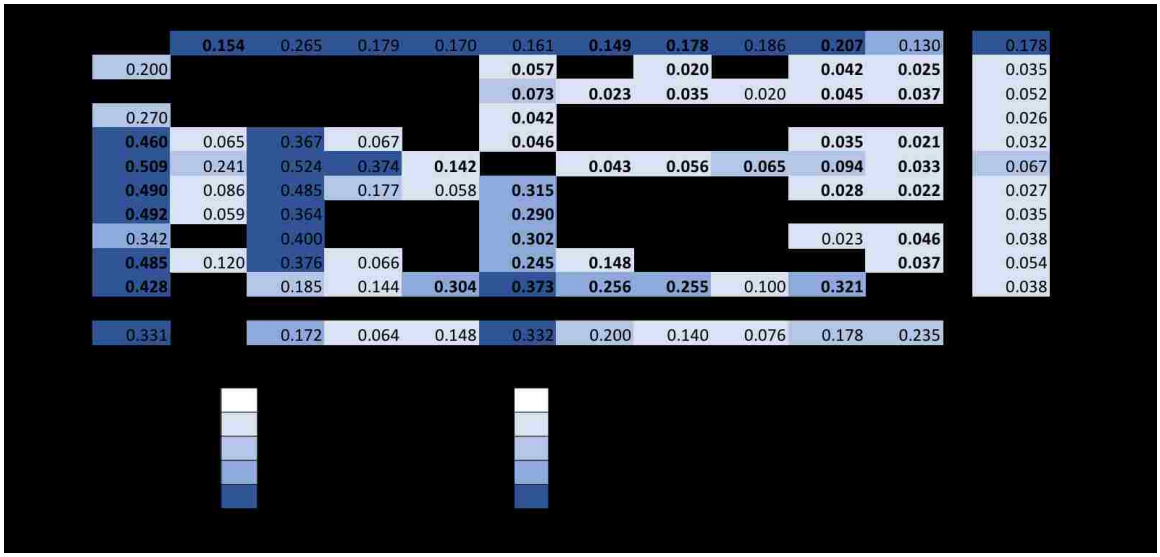


Table 1: Comparison of genetic diversity at microsatellite loci and mtDNA control regions among populations of wolverines in Alaska, western Canada, and eastern Russia. Bolded values indicate significance ($p < 0.05$).

Gene flow estimates are listed as immigration (Imm) into population A from population B and emigration (Em) from population A into population B. For example, gene flow between Russia and North Alaska is 21.68 N_fm out of Russia into North Alaska; therefore Russia is listed as the source.

Russia		
Imm	Em	Dir
10.5	12.5	-
8.9	21.68	Source
11.5	11.13	-
8.8	13.88	Source
9.2	6.81	-
13.2	13.97	-
21.2	13.81	Sink
12.2	8.41	-
8.3	13.48	Source
17.6	10.41	Sink

Table 2: Gene flow estimates calculated on the basis of coalescent N_fm (number of migrants per generation) in MIGRATE among wolverine populations in Alaska and northwestern Canada, calculated from the mtDNA control region. Directionality (Dir) of gene flow between population pairs (source, sink, and symmetrical [-] assigned on the basis of 95% confidence intervals. Bolded directionality shows strong asymmetry (>2x N_fm difference). Russia has been grayed due to its ~11 Kya separation (Jakobsson et al. 2017).

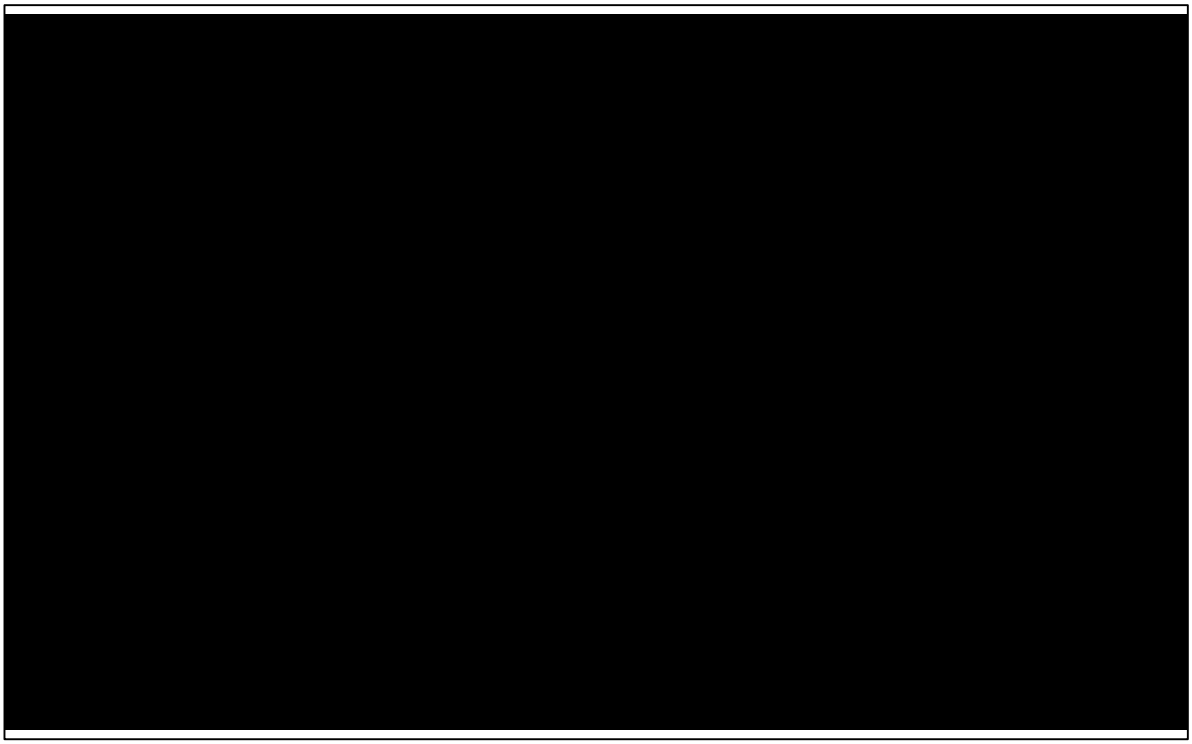


	0.154	0.265	0.179	0.170	0.161	**0.149**	**0.178**	0.186	**0.207**	0.130	0.178
0.200					**0.057**		**0.020**		**0.042**	**0.025**	0.035
					0.073	**0.023**	**0.035**	0.020	**0.045**	**0.037**	0.052
0.270					**0.042**						0.026
0.460	0.065	0.367	0.067		**0.046**				**0.035**	**0.021**	0.032
0.509	0.241	0.524	0.374	**0.142**		**0.043**	**0.056**	**0.065**	**0.094**	**0.033**	0.067
0.490	0.086	0.485	0.177	0.058	**0.315**				**0.028**	**0.022**	0.027
0.492	0.059	0.364			**0.290**						0.035
0.342		0.400			**0.302**				0.023	**0.046**	0.038
0.485	0.120	0.376	0.066		**0.245**	**0.148**				**0.037**	0.054
0.428		0.185	0.144	**0.304**	**0.373**	**0.256**	**0.255**	0.100	**0.321**		0.038
0.331		0.172	0.064	0.148	0.332	0.200	0.140	0.076	0.178	0.235	

Table 3: Population pairwise F_{ST} values for microsatellite loci (above the diagonal) and for concatenated cytochrome *b* and control region sequences (below the diagonal) in eleven sampling regions for wolverines. Bold values indicate statistical significance after 1,000 permutations. Weir and Cockerham's (1984) theta (F_{ST}) calibrated with the maximum global value of F_{ST} was used to generate the scales.

Supplementary Materials

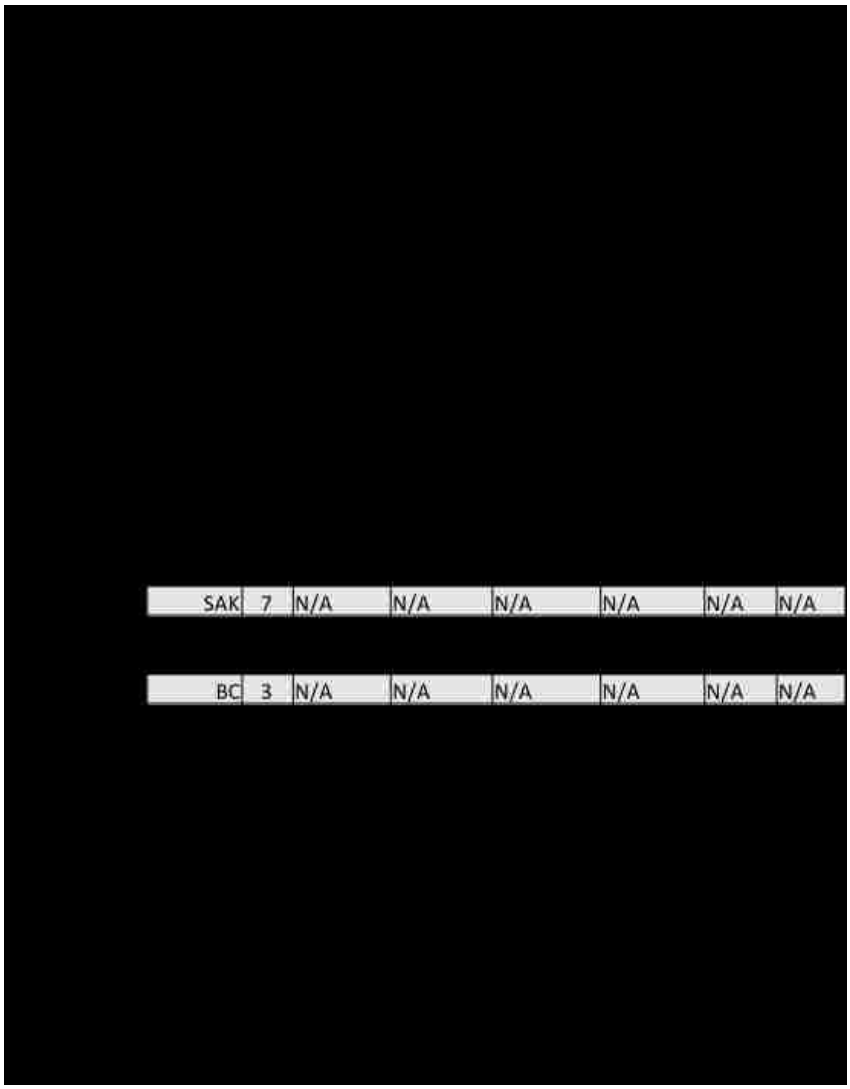
S1 Figure: ESA petitions and reasons for reversal since 1994.



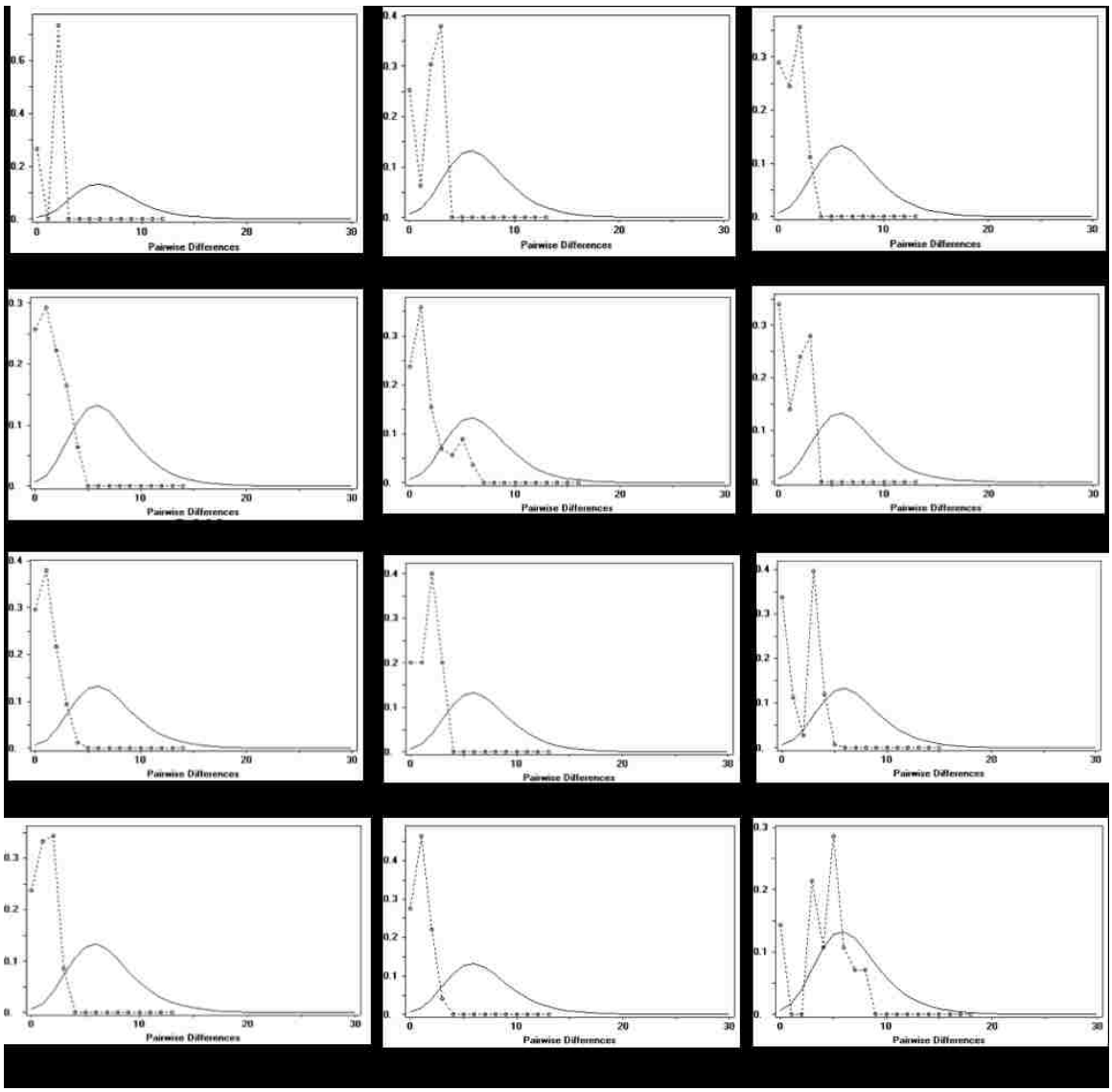
S2 Table: Specimens used by museum catalog number. Control region sequence data generated by Frances (2008) and GenBank data sources (*N*=234). Cytochrome *b* sequence data generated within the current study and GenBank data sources (*N*=69). Microsatellite genotypes generated within the current study (*N*=177) except NWAK (*N*=30, Dalerum et al. 2007).

S3 Table: Number of samples, allele counts, repeat motif, minimum and maximum length of calls, multiplex assignment, forward and reverse primer, citation, and GenBank or EMBL (first locus only) accession number. Bolded loci were adjusted slightly from the original, 1 denoting the forward primer was optimized and 2 denoting the reverse.

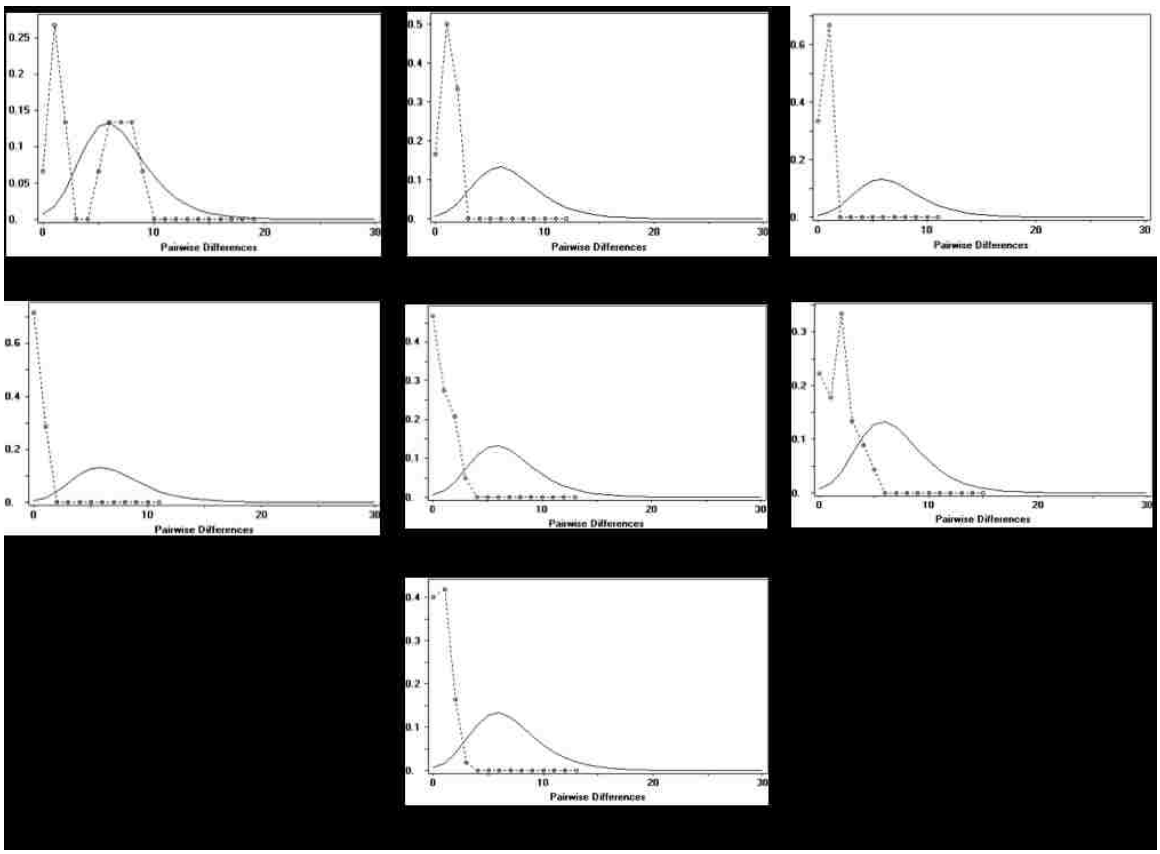
S4 Table: Site characteristics, sample size (*N*) and genetic diversity measures: haplotype diversity Hd, Tajima's D, Fu's Fs, average number of pairwise differences *k*, number of polymorphic (segregating) sites *S* for the populations analyzed for mitochondrial sequences control region and cytochrome b. *p-value > 0.05, **p-value > 0.02, N/A lack polymorphism or insufficient data/sample size.



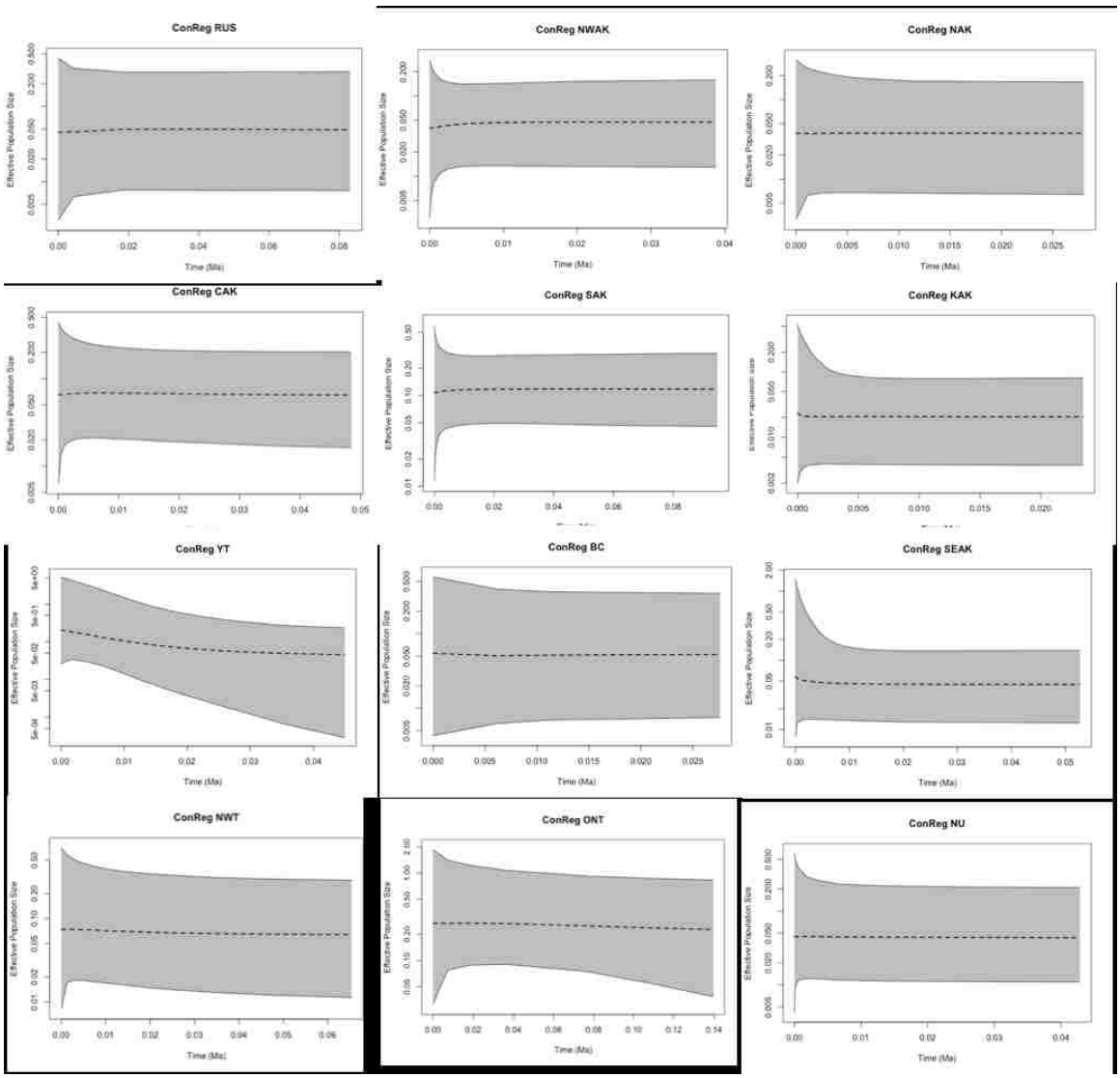
SAK	7	N/A	N/A	N/A	N/A	N/A	N/A
BC	3	N/A	N/A	N/A	N/A	N/A	N/A



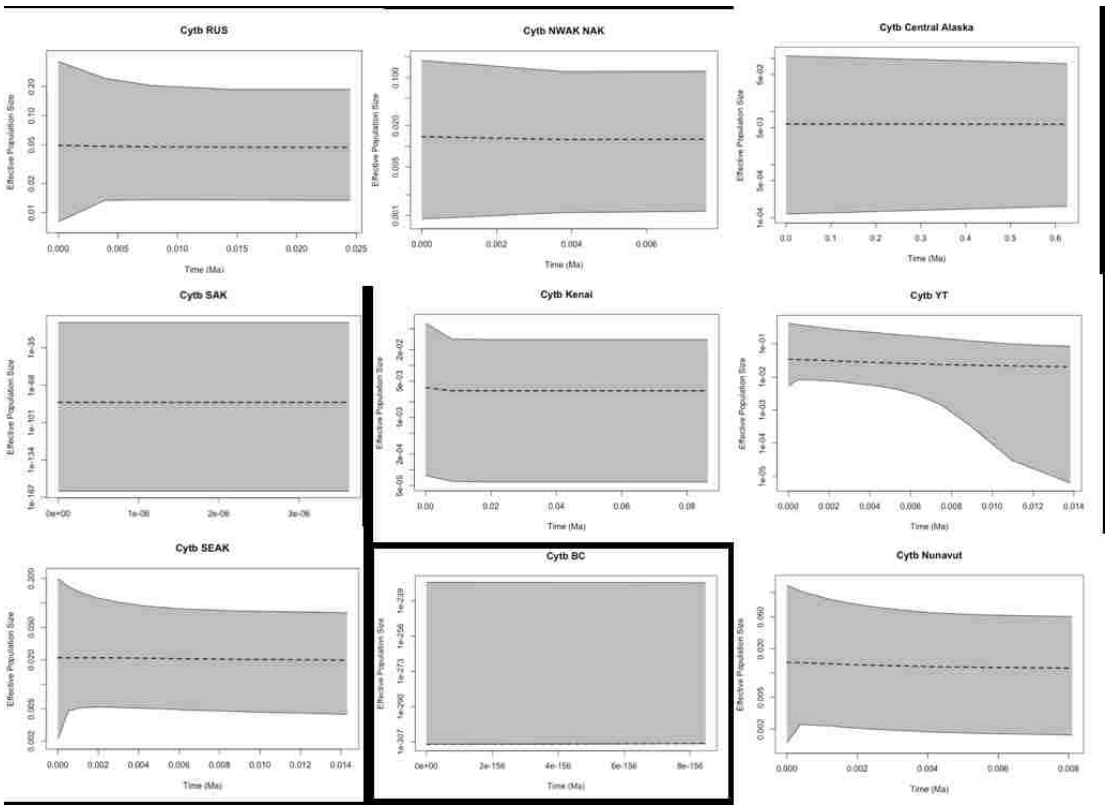
S5 Figure: Population expansion graphs for control region (367bp) data of the mitochondrial genome. Solid lines are the expected pattern, dotted lines depict the data. Bi-model or multi-model patterns indicate stable populations. *N* sampling: RUS=6, NWAK=23, NAK=10, CAK=19, SAK=33, KAK=25, YT=23, BC=5, SEAK=30, NWT=15, NU=63.



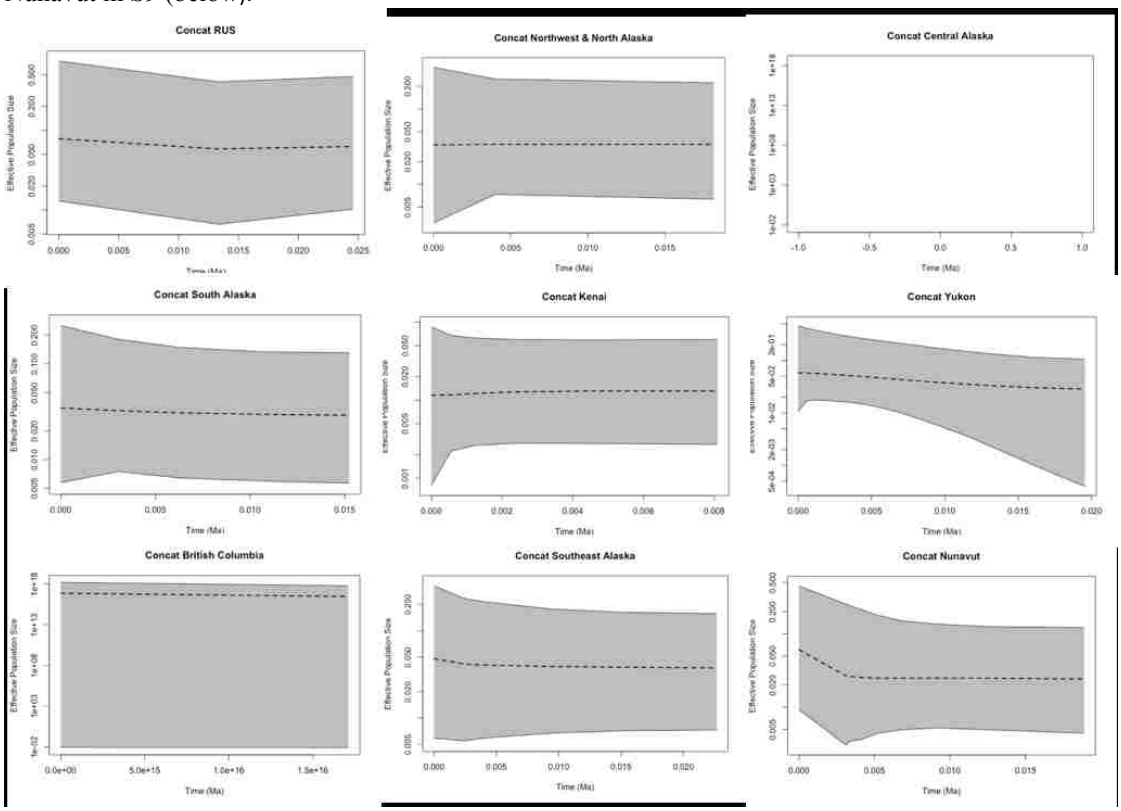
S6 Figure: Population expansion graphs for cytochrome *b* (1140bp) of the mitochondrial genome. Solid lines are expected pattern for population growth and decline, dotted lines depict the data. Bi-model or multi-model patterns indicate stable populations. South Alaska and British Columbia not pictured because they lacked polymorphisms. *N* sampling: RUS=6, NWAK/NAK=4, CAK=3, KAK=7, YT=16, SEAK=10, NU=11; SAK=7, BC=3.

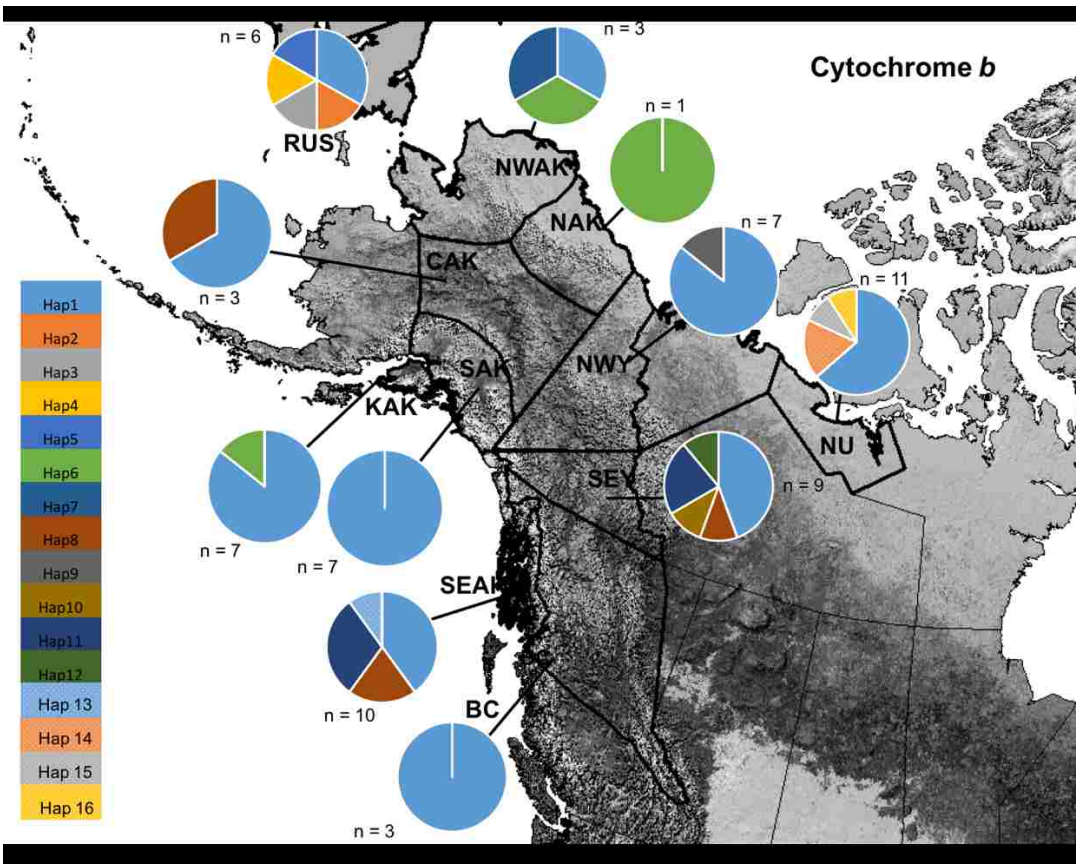


S7 Figure: Extended Bayesian Skyline Plots (EBSP) for mtDNA control region, including the dotted trend line and gray shading representing 95% error bounds. In all plots there is little evidence for expansion.

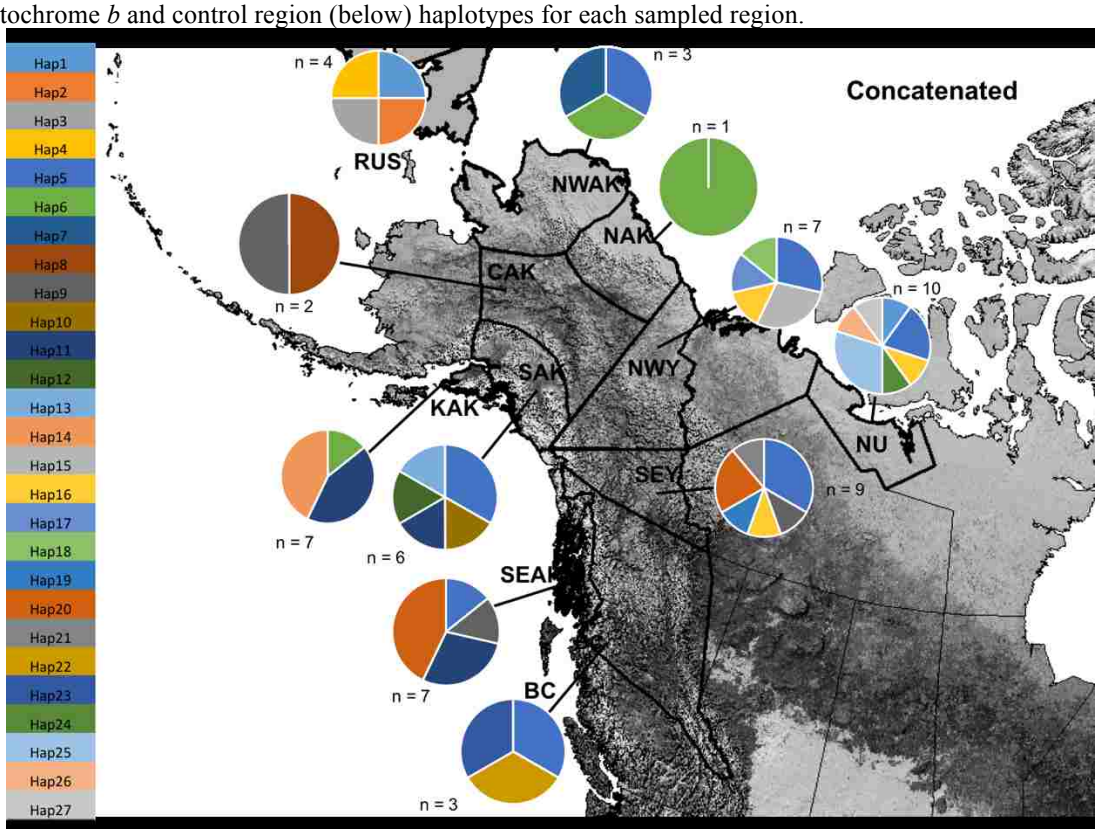


S8, S9 Figures: Extended Bayesian Skyline Plots (EBSP) for mtDNA cytochrome *b* (above), and concatenated cytochrome *b* and control region (below), including the dotted trend line and gray shading representing 95% error bounds. There has been no general shift in population size except expansion in Nunavut in **S9** (below).

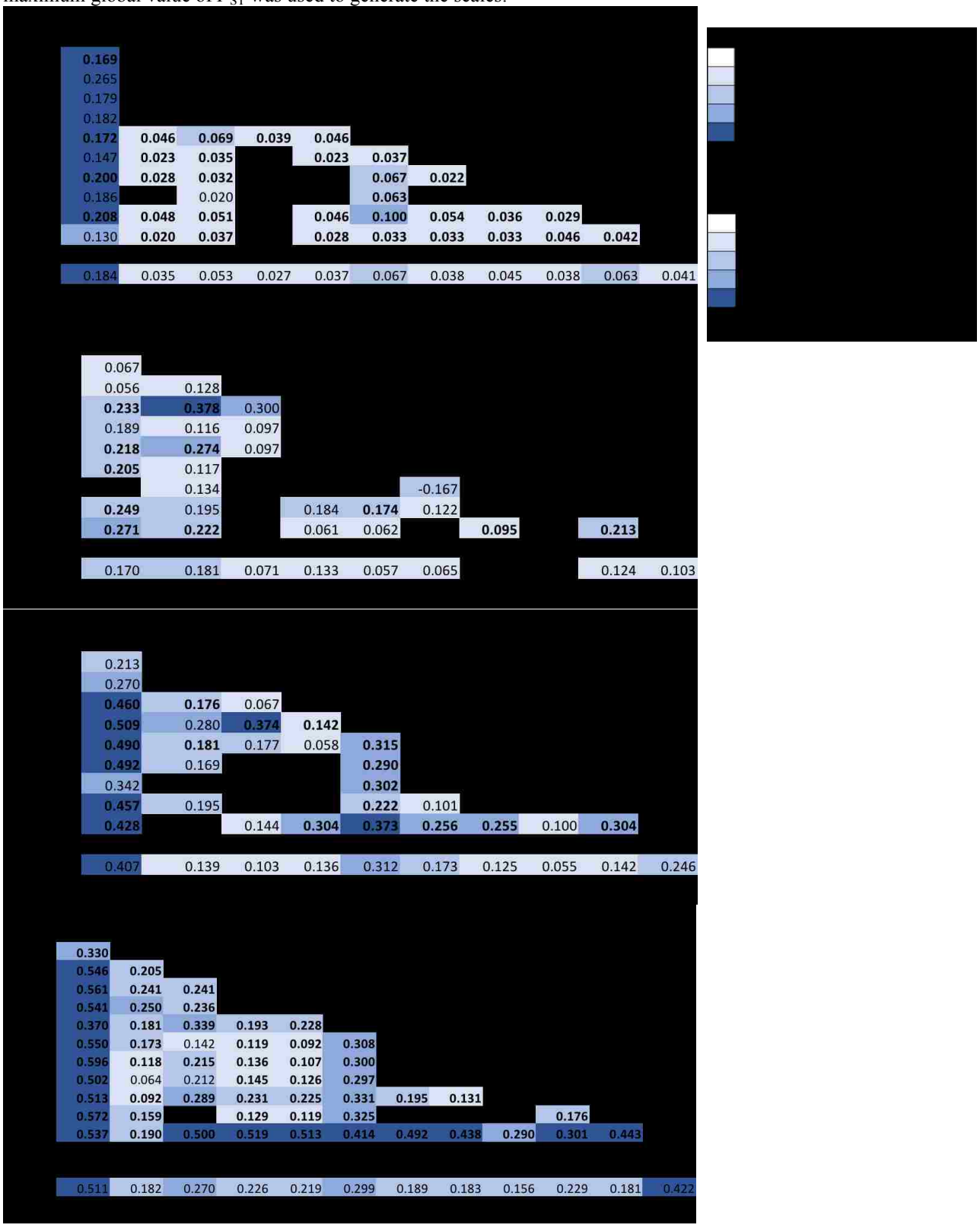




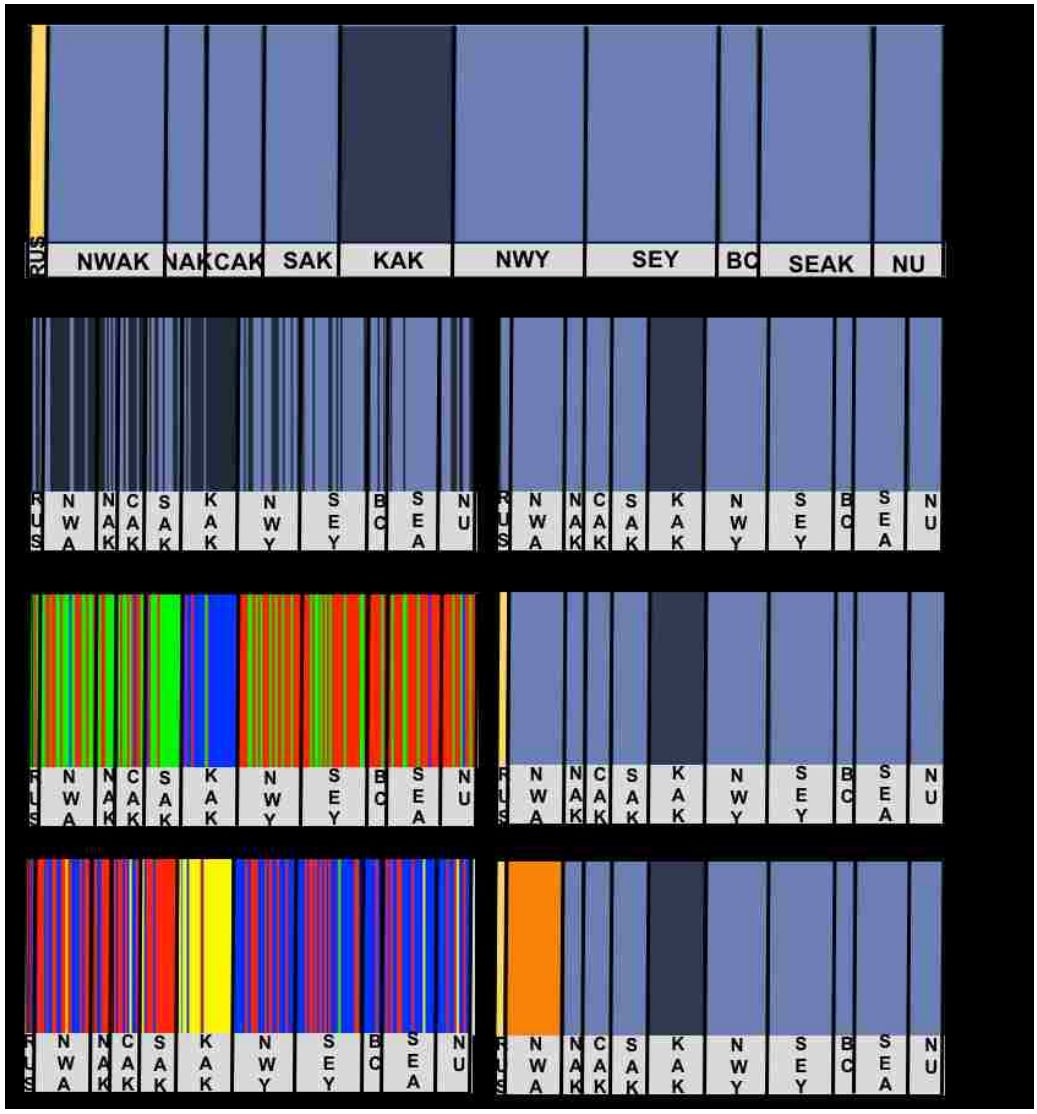
S10 Figures a & b: Frequency distribution within mtDNA cytochrome *b* (above) and concatenated cytochrome *b* and control region (below) haplotypes for each sampled region.



S11 Table: Pairwise F_{ST} values between populations of wolverines; (a) microsatellites; (b) cytochrome *b*; (c) concatenated cytochrome *b* and control region; and (d) control region. Bold values indicate statistical significance after 1,000 permutations. Weir and Cockerham's (1984) theta (F_{ST}) calibrated with the maximum global value of F_{ST} was used to generate the scales.

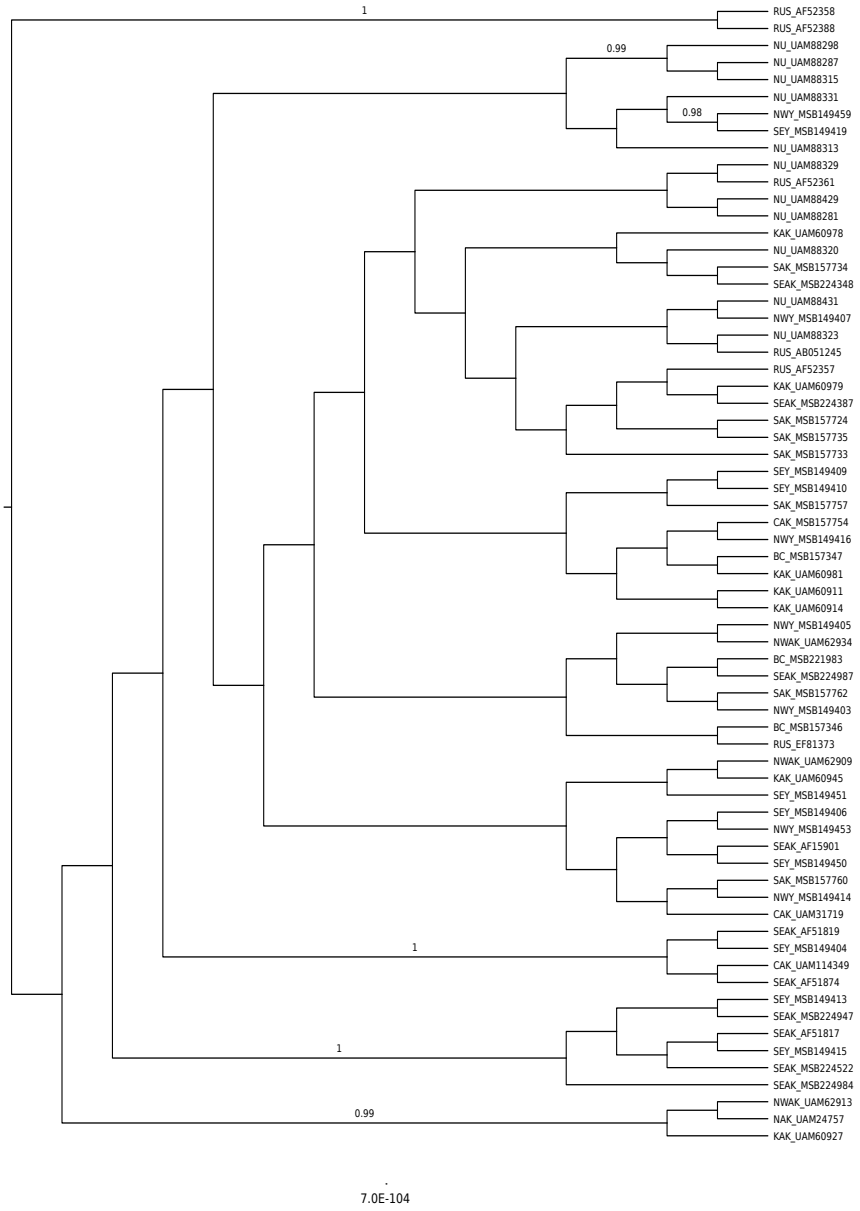


S12 Figures: BAPS output for microsatellite data for by-individual and by-population groups. (a) True k = 3 with Kenai, Russia, and the rest of Alaska and Canada as three distinct clusters, (b) assigned k = 2; individual and population level clusters where Kenai is distinct, (c) assigned k = 3; individual and population level clusters where Russia falls out next as divergent, (d) assigned k = 4; individual and population level clusters where Northwest Alaska falls out next.



S13 Figure: Phylogenetic relationships constructed from 67 sequences of the 1140bp cytochrome *b* region using Bayesian analysis in BEAST v1.8. (a) Table of identical sequences with node label and matching sequences, (b) tree from 67 sequences under strict time clock, posterior branch supports shown when above 0.90.

Node Label	Matching Sequences
BC_MSB157346	SEY_MSB149410
	SEY_MSB149406
	SEAK_MSB224987
	SEAK_MSB224387
	SEAK_MSB224348
	SEAK_AF15901
	SAK_MSB157762
	SAK_MSB157760
	SAK_MSB157757
	SAK_MSB157724
	RUS_AF52357
	RUS_AB051245
	NWY_MSB149416
	NWY_MSB149407
	NWAK_UAM62909
	NU_UAM88323
	NU_UAM88313
	NU_UAM88281
	KAK_UAM60981
	KAK_UAM60979
	KAK_UAM60978
	KAK_UAM60945
	KAK_UAM60914
	KAK_UAM60911
	CAK_MSB157754
	BC_MSB221983
SEAK_AF51817	SEAK_MSB224947
	SEAK_MSB224522
CAK_UAM114349	SEAK_AF51874
	SEAK_AF51819
KAK_UAM60927	NWAK_UAM62913
	NAK_UAM24757
NU_UAM88320	SAK_MSB157733
BC_MSB157347	CAK_UAM31719
NU_UAM88431	NWY_MSB149414
NU_UAM88287	NU_UAM88298
NU_UAM88315	
NU_UAM88329	
NU_UAM88331	
NU_UAM88429	
NWAK_UAM62934	
NWY_MSB149403	
NWY_MSB149405	
NWY_MSB149453	
NWY_MSB149459	
RUS_AF52358	
RUS_AF52361	
RUS_AF52388	
RUS_EF81373	
SAK_MSB157734	
SAK_MSB157735	
SEAK_MSB224984	
SEY_MSB149404	
SEY_MSB149409	
SEY_MSB149413	
SEY_MSB149415	
SEY_MSB149419	
SEY_MSB149450	
SEY_MSB149451	



S14 Figure: Phylogenetic relationships constructed from 252 sequences of the 366bp mtDNA control region using Bayesian analysis in BEAST v1.8. (a) Table of identical sequences with node label and matching sequences, (b) phylogenetic tree under strict time clock simplified to show only unique sequences, posterior branch supports shown when above 0.90.

Node_Label	MatchingSequences	Node_Label	MatchingSequences	Node_Label	MatchingSequences
BC_MSB157347	SEY_MSB149450	CAK_MSB157744	SEAK_MSB224586	CAK_MSB197415	NWY_MSB149452
	SEY_MSB149419		SEAK_MSB224387		NWT_AF210102
	SEY_MSB149415		SEAK_MSB224348		NWT_AF210101
	SEY_MSB149413		SAK_UAM 63021		NWT_AF210094
	SEY_MSB149411		SAK_UAM 62476		NAK_UAM 70447
	SEY_MSB149410		SAK_UAM 44702		NAK_UAM 63036
	SEY_MSB149409		SAK_UAM 44700		NAK_UAM 50949
	SEY_MSB149406		SAK_MSB157756		NAK_UAM 49893
	SEAK_UAM 101876		SAK_MSB157738		NAK_UAM 24084
	SEAK_UAM 49993		SAK_MSB157733		CAK_UAM 31719
	SEAK_UAM 48632		SAK_MSB157726	KAK_UAM 60906	KAK_UAM 61015
	SEAK_MSB224987		SAK_MSB157721		KAK_UAM 60981
	SEAK_MSB224984		SAK_MSB157718		KAK_UAM 60979
	SEAK_MSB224978		SAK_MSB157715		KAK_UAM 60945
	SEAK_MSB224947		SAK_MSB157678		KAK_UAM 60944
	SEAK_MSB224522		KAK_UAM 61016	NWY_MSB149405	SEY_MSB149421
	SEAK_MSB224447		KAK_UAM 61014		SEAK_UAM 53409
	SEAK_MSB149573		KAK_UAM 60978		NWY_MSB149414
	SEAK_AF51821		KAK_UAM 60954	NAK_UAM 41582	NWT_AF210098
	SAK_UAM 62886		KAK_UAM 60914		NWAK_UAM 62939
	SAK_UAM 62480		KAK_UAM 60911	NWY_MSB149403	SEAK_MSB149569
	SAK_UAM 62479		KAK_MSB157707	SAK_MSB157723	SAK_MSB157757
	SAK_MSB157764		CAK_UAM 72025	SAK_MSB157714	SAK_MSB157725
	SAK_MSB157760		CAK_UAM 66977	SAK_MSB157724	SAK_MSB157737
	SAK_MSB157734		CAK_UAM 62481	BC_MSB221983	
	SAK_MSB157732		CAK_UAM 51727	NWT_AF210097	
	SAK_MSB157722		CAK_UAM 51726	NWY_MSB149408	
	SAK_MSB157716		CAK_MSB157766	NWY_MSB149416	
	SAK_MSB157705		CAK_MSB157759	RUS_AF52358	
	SAK_MSB157679		CAK_UAM 157752	RUS_AF52360	
	NWY_MSB149459	CAK_UAM 114349	SEY_MSB149404	RUS_AF52388	
	NWY_MSB149453		SEAK_UAM 101877	SAK_MSB157762	
	NWY_MSB149420		SEAK_UAM 53410	SAK_MSB157763	
	NWT_AF210103		SEAK_MSB224685	SEAK_MSB149566	
	NWT_AF210100		SEAK_MSB224504	SEAK_MSB149567	
	NWT_AF210099		SEAK_MSB149572		
	NWT_AF210093		SEAK_MSB149571		
	NWT_AF210092		SEAK_MSB149570		
	NWT_AF210091		SEAK_MSB149568		
	NWT_AF210090		SEAK_AF51822		
	NWAK_UAM 63816		SEAK_AF51820		
	NWAK_UAM 63087		SEAK_AF51819		
	NWAK_UAM 62917		SEAK_AF51818		
	NWAK_UAM 62915		NWAK_UAM 62940		
	NWAK_UAM 62914		NWAK_UAM 62934		
	NWAK_UAM 62911		NWAK_UAM 62929		
	NWAK_UAM 62909		NWAK_UAM 62919		
	NU_UAM 88438		NWAK_UAM 62910		
	NU_UAM 88429		NWAK_UAM 62905		
	NU_UAM 88281		NU_UAM 88324		
	NU_AF210123	CAK_MSB157767	SEY_MSB149451		
	CAK_UAM 66978		SEY_MSB149418		
	CAK_UAM 51725		SEY_MSB149417		
	CAK_MSB157730		NWY_MSB149407		
	CAK_IF6450		NWT_AF210104		
	BC_MSB221959		NWAK_UAM 63817		
CAK_MSB157736	SAK_UAM 72035		NU_UAM 88330		
	SAK_MSB157754		NU_UAM 88329		
	RUS_MSB151952		NU_UAM 88328		
	RUS_AF52361		NU_UAM 88327		
	RUS_AF52357		NU_UAM 88295		
	NWAK_UAM 62999		NU_UAM 88285		
	NWAK_UAM 62941		NU_AF210119		
	NWAK_UAM 62932		NU_AF210111		
	NWAK_UAM 62930		NU_AF210110		
	NWAK_UAM 62916		NU_AF210105		
	NWAK_UAM 62913		NU_UAM 24955		
	NWAK_UAM 62908		NU_UAM 24083		
	NWAK_UAM 62907		NU_UAM 24082		
	NU_UAM 88323	BC_MSB157345	NU_UAM 88436		
	NU_UAM 88316		NU_UAM 88435		
	NU_UAM 88309		NU_UAM 88432		
	NU_AF210130		NU_UAM 88325		
	NAK_UAM 24757		NU_UAM 88318		
	KAK_UAM 60952		NU_UAM 88315		
	KAK_UAM 60929		NU_UAM 88301		
	KAK_UAM 60927		NU_UAM 88298		
	KAK_UAM 60926		NU_UAM 88288		
	KAK_UAM 60912		NU_AF210125		
	KAK_UAM 60910		NU_AF210124		
	KAK_UAM 60909		NU_AF210120		
	KAK_UAM 60907		NU_AF210117		
	KAK_UAM 60904		NU_AF210116		
	KAK_MSB157720		NU_AF210109		
	KAK_MSB157717		NU_AF210108		
	KAK_MSB157708		NU_AF210107		
	CAK_UAM 51442	BC_MSB157346	NWT_AF210096		
NU_AF210106	NU_UAM 88431		NWT_AF210095		
	NU_UAM 88331		NU_UAM 88430		
	NU_UAM 88320		NU_UAM 88317		
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	NU_UAM 88314		NU_UAM 88292		
	NU_UAM 88313		NU_AF210129		
	NU_UAM 88300		NU_AF210128		
	NU_UAM 88283		NU_AF210126		
	NU_UAM 88282		NU_AF210122		
	NU_AF210132		NU_AF210112		
	NU_AF210131				
	NU_AF210127				
	NU_AF210121				
	NU_AF210118				
	NU_AF210115				
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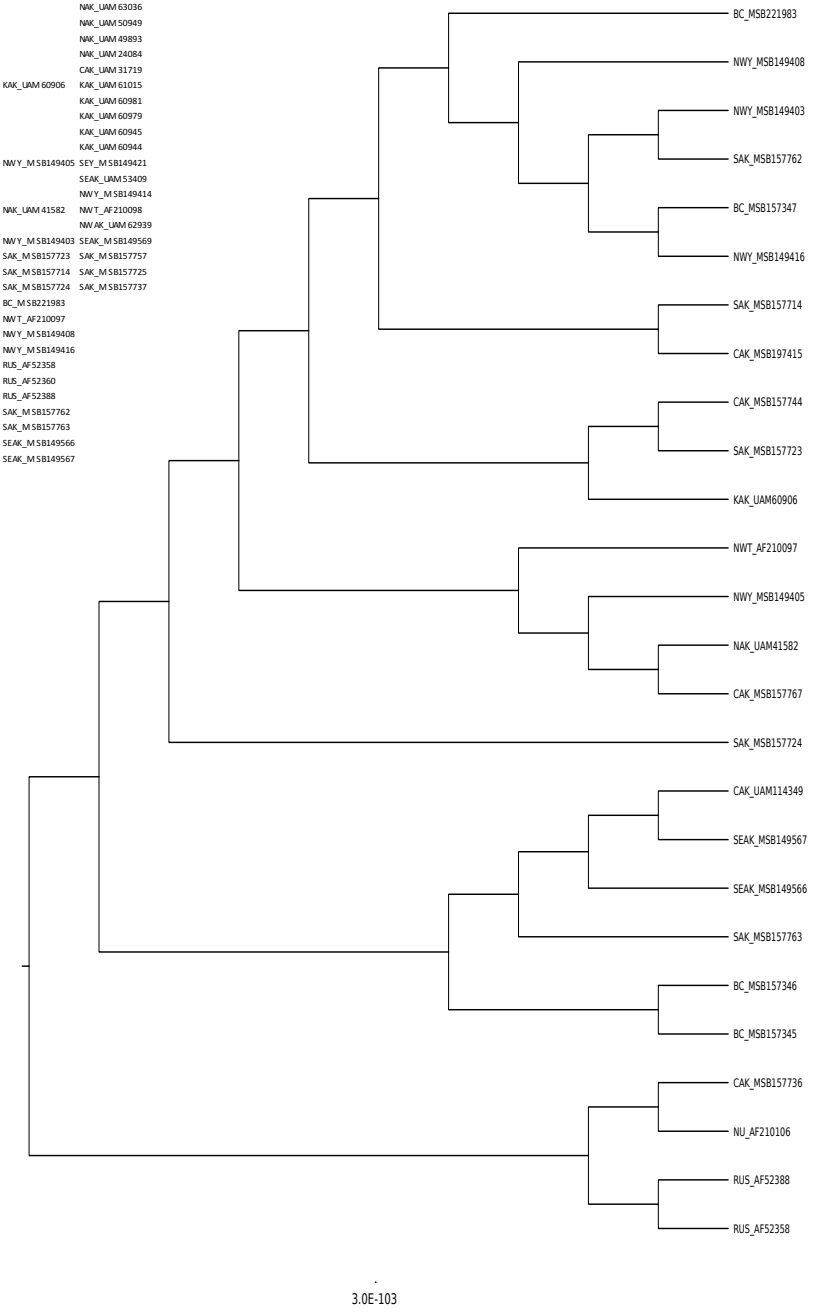
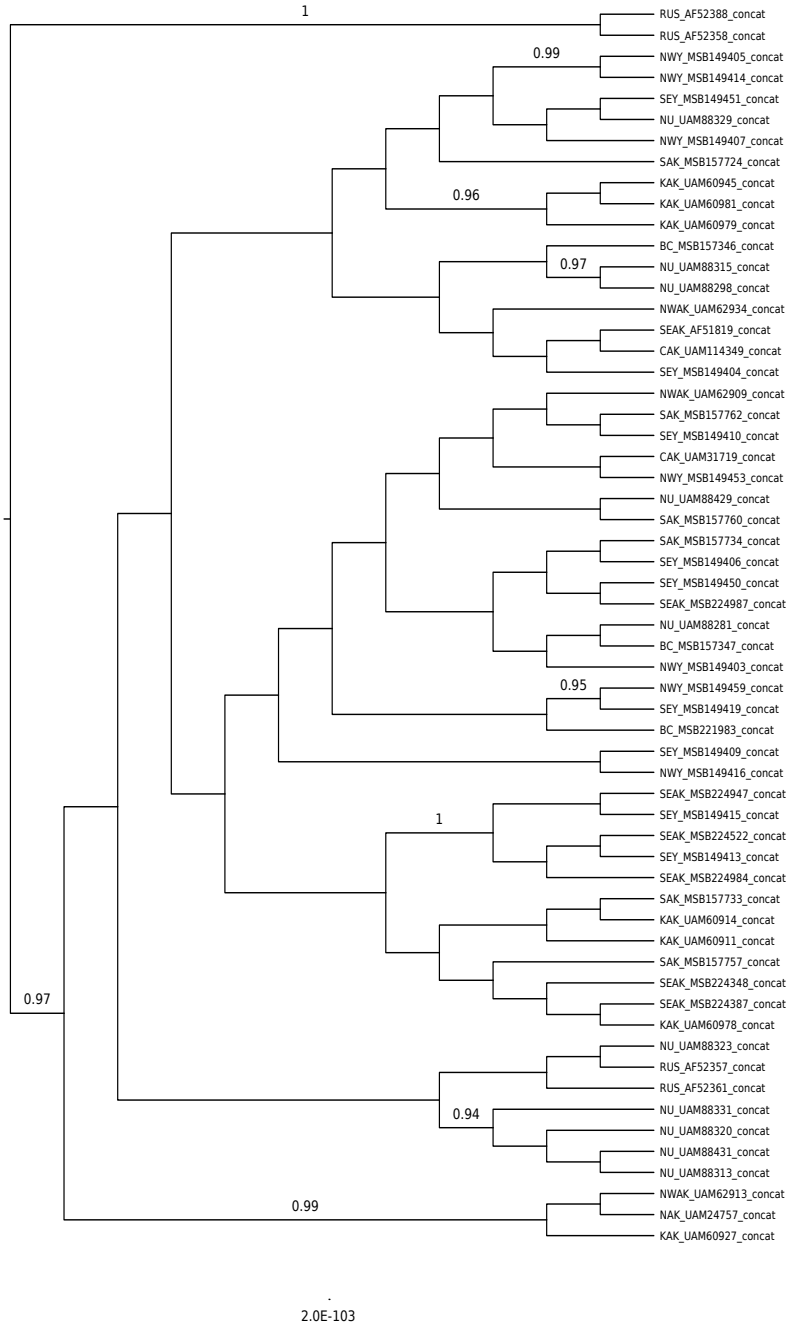


S15 Figure: Phylogenetic tree constructed from 59 sequences of the 1408bp concatenated cytochrome *b* and control region using Bayesian analysis in BEAST v1.8. (a) Tree under strict time clock with branch supports showing posterior probabilities, significant numbers being 0.9 or over. A pattern emerges of Southeast Alaska and South Yukon individuals having support for common lineage. (b) table of identical sequences with node label and matching sequences.

Node Label	Matching Sequences
NU_UAM88281	SEY_MSB149410
	SEY_MSB149406
	SEAK_MSB224987
	SAK_MSB157760
	NWAK_UAM62909
KAK_UAM60911	SEAK_MSB224387
	SEAK_MSB224348
	KAK_UAM60978
	KAK_UAM60914
KAK_UAM60927	NWAK_UAM62913
	NAK_UAM24757
KAK_UAM60945	KAK_UAM60981
	KAK_UAM60979
NU_UAM88323	RUS_AF52357
SEAK_MSB224522	SEAK_MSB224947
CAK_UAM114349	SEAK_AF51819
BC_MSB157346	
BC_MSB157347	
BC_MSB221983	
CAK_UAM31719	
NU_UAM88298	
NU_UAM88313	
NU_UAM88315	
NU_UAM88320	
NU_UAM88329	
NU_UAM88331	
NU_UAM88429	
NU_UAM88431	
NWAK_UAM62934	
NWY_MSB149403	
NWY_MSB149405	
NWY_MSB149407	
NWY_MSB149414	
NWY_MSB149416	
NWY_MSB149453	
NWY_MSB149459	
RUS_AF52358	
RUS_AF52361	
RUS_AF52388	
SAK_MSB157724	
SAK_MSB157733	
SAK_MSB157734	
SAK_MSB157757	
SAK_MSB157762	
SEAK_MSB224984	
SEY_MSB149404	
SEY_MSB149409	
SEY_MSB149413	
SEY_MSB149415	
SEY_MSB149419	
SEY_MSB149450	
SEY_MSB149451	



CHAPTER 2

WOLVERINE POPULATION DYNAMICS VARY ACROSS HABITATS: PANMIXIA AND SEX-BIASED DISPERSAL AT HIGHER LATITUDES

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Abstract

The circumboreal wolverine (*Gulo gulo*) is an optimal species for studying responses to environmental perturbation in the North due to an extensive history of persistence in glacial refugia and subsequent post-glacial recolonization. Because genetic structure provides clues to past distributional responses, our research uses DNA variation to identify potential dispersal patterns and source-sink dynamics to inform conservation strategies for wolverines that now may be impacted by anthropogenic harvest, habitat conversion, and directional climate change. Male and female dispersal is compared using 20 microsatellite loci across 360 individuals sampled from 270,000 km^2 of Canadian

boreal forest and 80,000 km^2 of Alaskan tundra using contemporary populations (1995-2015). Overall panmixia and lack of sex-biased dispersal occurs at higher latitudes, a finding that contrasts with data recorded for wolverine dispersal at lower latitudes in more fragmented landscapes throughout the contiguous United States and southern Canadian provinces. Relatively undisturbed high latitude environments for wolverines provide a basis for interpreting the population genetic dynamics of this vagile species at lower latitudes.

Keywords: dispersal, *Gulo gulo luscus*, isolation by distance, sex-bias, source-sink dynamics

Introduction

Dispersal often differs between sexes and can greatly influence genetic structure. In many polygynous mammals, males disperse farther than females (i.e., male-biased dispersal) as part of fitness strategies where philopatric females defend resources in close habitats they are familiar with, presumably to increase success in rearing offspring (Greenwood 1980), while wider-ranging males maximize access to females (Dobson et al. 1982; Moore and Ali 1984). This behavioral ecology creates contrasting genetic structure and gene flow between sexes (Goudet et al. 2002; Lawson Handley and Perrin 2007; Mossman and Waser 1999). With male-biased dispersal, comparatively higher genetic structure is expected in the maternally inherited mitochondrial genome (mtDNA), while biparentally-inherited nuclear genes show lower genetic structure due to higher levels of male-mediated gene flow (Goudet et al. Perrin 2002).

In line with these predictions, North American wolverines (*Gulo gulo luscus,* Linnaeus, 1758) demonstrate pronounced differences in nuclear and mitochondrial genetic structure (Chappell et al. 2004; Kyle et al. 2001; Kyle and Strobeck 2002; Wilson et al. 2000; Cegelski et al. 2006). Wolverines are highly vagile; topographic features that may structure other species, such as mountain ranges, rivers, or valleys (Hornocker and Hash 1981), generally do not limit dispersal. Previous studies establishing optimal habitat requirements have found snow pack for denning (Aubry et al. 2007; Copeland et al. 2010), remote talus, tundra, and coniferous environments (Inman et al. 2013), and access to ungulate prey (Young et al. 2012) are important for wolverine persistence. Wolverines tend to disperse before 2 years of age (Banci and Harestad 1990; Vangen et al. 2001) and their natal dispersal (one-time, permanent movement of young animals; Greenwood 1980) is sex-biased with males dispersing furthering (Copeland and Yates 2008). This bias is amplified by large differences in average male (500 km^2) and female (280 km^2) home range sizes (Banci 1987; Gardner 1985; Hornocker and Hash 1981; Magoun 1985; Whitman et al. 1986). In addition to sex bias, reproductive status, age, food availability, and habitat quality can affect home range, with averages ranging from just 100 km^2 in females with young to over 1,522 km^2 in adult males (Copeland et al. 2010; Pasitschniak-Arts and Larivière 1995; Chadwick 2010). Previous research suggests that wolverines in temperate latitudes exhibit great male-bias in natal dispersal in telemetry studies as well as male-bias in gene flow in genetic analysis (Chappell et al. 2004; Cegelski et al. 2006; Hornocker and Hash 1981; Inman et al. 2012, 2013).

In contrast to this, dispersal in wolverines in northwestern Alaskan tundra populations was not found to be male-biased (Dalerum et al. 2007); males and females

dispersed equal distances from their population of genetic origin. Three non-exclusive explanations were advanced for this outcome: small sample size and study area resulting in low statistical power to detect dispersal biases in a potentially panmictic population; scale-dependent issues where males were overrepresented among interpopulation dispersers in other studies; and lower reproductive success for dispersing females compared to more philopatric ones in other studies (due to the fitness strategy to remain close to an established home range to successfully rear kits; Inman et al. 2012). The influence of lower statistical power due to restricted sample size can be assessed by conducting research using similar methodology but on a larger number of individuals over a larger geographic distribution. At the same time, variability in dispersal bias due to habitat differences and resource availability can be tested by conducting this research in different habitats.

For example, in Molina's hog-nosed skunks (*Conepatus chinga*), greater dispersion of food resources necessitated larger female home ranges and generated less skew between males and females (Castillo et al. 2011). Tundra habitat has lower net primary production (NPP) compared to boreal forest (Melillo et al. 1993), and though prey sources and dispersion of food is difficult to quantify (wolverines are highly adaptable scavengers, hunters, and foragers; Magoun 1987) caribou (*Rangifer tarandus*) and moose (*Alces alces*) can be used as a general proxy for food availability since these large ungulates are preferred food items (found in 60% of wolverine stomachs in tundra and boreal forest regions of Alaska ($N = 193$); Rausch and Pearson 1972). Moose distribution is more common in boreal forest and individuals are resident to an area or are partially migratory (White et al. 2014), while caribou are characteristic of the tundra and

are highly migratory (Fancy et al. 1989). Impermanence of major ungulate food sources (Ballard et al. 1997), and lower NPP (Nilsen et al. 2005), may result in greater dispersion of food sources for wolverines on the tundra. Greater density of food resources may allow higher philopatry in females, while dispersion of resources may necessitate more female movement relative to males. Here, we test whether a difference in prey-habitat specialization (Musiani et al. 2007) dictates dispersal trends in wolverines by comparing Yukon individuals of forest environments to Alaskan populations on the tundra (using the dataset from Dalerum et al. 2007).

Demography is key to understanding dispersal. Wolverines, like many mammals, are limited by female effective population size through recruitment of offspring (Eberhardt 1990; Nunney 1993), so maintaining high female survivorship is vital, as it is in other low-density species like polar bears (*Ursus maritimus*) and grizzly bears (*Ursus arctos*) (Eberhardt 1990). Wolverines typically successfully produce kits at 3 years old, litters include 2-3 offspring with a 50% survival rate to breeding adulthood, and litters occur every 1-2 years (Banci and Harestad 1988; Inman et al. 2012; Rauset et al. 2015); population growth in medium-bodied carnivores like wolverines is notoriously slow. Because wolverines are commercially harvested furbearers, monitoring demographic fluctuations is important for sustainable harvests. Harvest is a major source of mortality for wolverines, constituting 83%, 41%, and 64% of tracked animal deaths (Hornocker and Hash 1981; Krebs et al. 2004; Squires et al. 2007, respectively). Female survivorship must exceed 0.85 in order to sustain populations (Eberhardt 1990) and allow long-term harvests (Krebs et al. 2004). To study this, source-sink dynamics screened in each region will test for effects of trapper harvest as well as source populations. Source populations

will have birth rates that exceed death rates and emigration that exceeds immigration (Dias 1996), whereas sink populations would not be sustainable alone.

Through a fine-scale population genetics study, we focus on male and female dynamics separately to: (a) study sex-biased dispersal in tundra and boreal forest habitat (Alaska and Yukon) where resource availability varies, and (b) explore source and sink population dynamics within areas impacted by trapper harvest. We hypothesize that Alaska tundra wolverine populations will not exhibit sex-biased dispersal while Yukon boreal forest populations will show male-biased dispersal. We also predict that source populations will be concentrated in the southeastern half of the Yukon Territory, based on previous wolverine historical expansion data from Southeast Alaska in Chapter 1 and this region's recognition as a glacial refugium during the Last Glacial Maximum (LGM) (Carrara et al. 2007; Fleming and Cook 2002; Josenhans et al. 1995; Mandryk et al. 2001). Glacial refugia can serve as source areas for modern populations (Ohlemüller et al. 2012). Further, the presence of a large game sanctuary in this region of the province (Kluane National Park, 22,000 km^2) may be a source of wolverines for other areas.

Materials and Methods

Sampling

Wolverine carcasses were salvaged from commercial trappers in Alaska (USA) and the Yukon Territory (Canada) by state and provincial wildlife officials and permanently archived at two museums (Museum of Southwestern Biology, University of New Mexico and Museum of the North, University of Alaska Fairbanks) over the last ~2 decades (2005-2015 Yukon; 1996-2001 Alaska). Samples (Supplementary Material **S1**

Table) with reliable spatial and temporal information were selected to represent the region. For Yukon Territory samples, DNA was isolated using a salt extraction method (Fleming and Cook 2002) with a final sample size of $N = 114$ from northwestern Yukon (NWY) and $N = 129$ from southeastern Yukon (SEY) (**Figure 1**). Following inter-laboratory calibration of genotypes, data from individuals used in the study by Dalerum *et al.* (2007) were added to our dataset, representing sampling from northwestern Alaska (NWAK; $N = 117$). A small number of individuals had either 10% (Alaska, $N = 20$) or 5% (Yukon, $N = 2$) missing data. Exploratory analyses demonstrated these missing data had no significant impact on analyses (not shown). All individuals had complete sex and age determination. Sex was determined from museum records for the Alaska samples and the Yukon samples were sexed via sex-specific loci.

A canine tooth was extracted from harvested wolverines for age determination (Matson Laboratory, Milltown, Montana—Matson 1981, Alaska samples; author TSJ, Yukon samples). Based on tooth annuli counts, animals were divided into two discrete age classes (i.e., < 2 years and ≥ 2 years of age). Wolverines 2 years and older should represent already dispersed individuals (Banci and Harestad 1990; Vangen et al. 2001), whereas animals younger than 2 years of age may represent pre-dispersal, post-dispersal, or dispersing individuals. Individuals younger than 2 years were classed as subadults and animals 2 years or older were classed as adults in later analyses.

Microsatellite genotyping and molecular sexing

Microsatellite genotypes were determined for Yukon Territory individuals using 20 loci (**S2 Table**) with the following polymerase chain reaction (PCR) primers: Lut604 (Dallas and Piertney 1998), Gg-3, Gg-4, Gg7, MA-3, Tt-1, Tt-4 (Davis and Strobeck

1998), Ggu216 (Duffy et al. 1998), Mvis75 (Fleming et al. 1999), Gg10, Gg25, Gg37, Gg42, Gg192, Gg443, Gg452, Gg454, Gg465, Gg471, Gg473 (Walker et al. 2001). One primer (forward or reverse) from each of 7 of the loci was redesigned from the published sequence to change the size or improve quality of PCR product (**S2 Table**). PCR amplifications were carried out in 7 universal-tailed (Oetting et al. 1995) multiplex reactions and 1 singleplex reaction, each in a final volume of 10 μL and containing 2-50 ng genomic DNA, 0.2 mM deoxynucleoside triphosphates, 1-5 pmols unlabeled primers, 0.15-2.25 pmoles IRD-labeled primer, 1.0 μg bovine serum albumin, 1xPCR buffer (Perkin Elmer Cetus I), and 0.25-0.5 units of GoTaq®Flexi DNA polymerase (Promega, Madison, WI). PCRs began at 94°C for 2 min then continued with 40 cycles each of 94°C for 15-30 s, 50°C for 15-30 s, and 72°C for 30-60 s. A 30 min extension at 72°C concluded each reaction. Comparisons between Alaska and the Yukon populations were conducted using the 10-locus suite common among both datasets (**S2 Table**). Similarly, analyses of genetic diversity (i.e., H_O/H_E and STRUCTURE output) were conducted with these 10 loci when comparing Yukon with Alaska; otherwise, full data were used for intrapopulation analyses and within-Yukon comparisons.

Fluorescently labeled PCR products were electrophoresed on a 48-well 6% polyacrylamide gel on a LI-COR 4200 LR or IR^2 DNA automated sequencer (LI-COR, Lincoln NE). For allele size standardization for the 10 loci amplified by Dalerum et al. (2007), we generated a suite of size standards for each locus by sizing 2-4 samples against standards of known size (four wolverine samples provided by Janet Loxterman, sensu Dalerum et al. 2007). Two of these size standards were used in a minimum of 6 lanes in all subsequent genotyping reactions. For the remaining loci, size standards were

generated for each locus by scoring the same suite of individuals against a fluorescently-labeled M13 sequence ladder of known size, and these samples were used in each subsequent gel as size standards, again occupying at least 6 lanes across each 48-well gel. Based on these standards, genotypes for each individual were determined using GeneImagIR 4.05 software (Scanalytics, Inc.). For quality control, 12% of our Yukon samples were extracted, amplified, and genotyped in duplicate. Locus error rate was determined at 1.6%, allele error rate was 0.98% for Yukon samples. MICROCHECKER 2.2.3 (Van Oosterhout et al. 2004) was used to identify genotyping errors and check for null alleles. Positive and negative controls were used throughout.

To confirm the sex of Yukon wolverines, we amplified DNA from 10 wolverines of known sex under standard PCR conditions, initially using primers LGL331 and LGL335 (Shaw et al. 2003) that target an intron within the mammalian zinc-finger x (*Zfx*) and zinc-finger y (*Zfy*) genes, using procedures reported by Fischbach et al. (2008). That reaction amplifies a >950 bp product that requires fragment separation via electrophoresis on agarose gels. To decrease the size of the product in wolverines and facilitate automated fragment detection, we generated nucleotide sequence data (on GenBank) from the mammalian *Zfx* gene of 2 female wolverines (UAM31719, MSB157754), using simultaneous bidirectional sequencing procedures similar to procedures described elsewhere (Jackson et al. 2008), incorporating the LGL331 and LGL335 primers as sequence primers. The wolverine sequences were aligned against *Zfx* and *Zfy* sequences from 5 other mustelid species (*Martes martes*, *Martes zibellina*, *Martes melampus*, *Meles anakuma*, and *Neovison vison*) and gray wolf (*Canis lupus*) archived in GenBank (**S3 Figure**). From this alignment we designed a new primer, MustelaZF (5' –

GMAATCATTCATGAATAT – 3') which, along with LGL335, targets a a 203-base-pair (bp) product from the X-chromosome (both males and females). Based on *Zfy* data from other mustelids, we estimated a 198-bp product from the Y-chromosome (males only) in wolverines. The 5 bp differences between the *Zfx* and *Zfy* fragments for wolverine were verified by comparing sizes against a fluorescently-labeled M13 sequence ladder of known size.

Primer MustelaZF and LGL335 were each synthesized with IRD-labeled universal primers (M13F, M13R, respectively), added to a PCR cocktail, and subjected to amplifications in a final volume of 10 µL. This final volume contained 50ng genomic DNA, 0.2 mM deoxynucleoside triphosphates, 3.6-4.0 pmol unlabeled primers, 0.06-0.4 pmoles IRD-labeled primer, 1.0 µg bovine serum albumin, 1xPCR buffer (Perkin Elmer Cetus I), and 0.3 units Amplitaq DNA polymerase (PE Biosystems, Forest City, CA). PCR cycling profiles followed those used for microsatellite loci above. We electrophoresed PCR reaction products on a 48-well 18-cm 6% polyacrylamide gel on a LI-COR 4200LR or IR^2 automated sequencer (LI-COR, Inc., Lincoln, Nebraska). Following testing against known-sex wolverines, we assigned sex based on the absence (female: 203/203) or presence (male: 198/203) of the band for the Y-chromosome.

Tests of neutrality, genetic diversity, and substructure

Genepop on the Web v4.2 (Raymond and Rousset 1995) was used to evaluate Hardy-Weinberg equilibrium (HWE) and linkage disequilibrium (LD) for each of the microsatellite loci and pairs of loci, respectively. HWE was tested using probability, heterozygosity deficiency, and heterozygosity excess with no enumeration of alleles

using Markov Chain parameters: 10,000 dememorizations, 1,000 batches, and 10,000 iterations. LD was tested with log likelihood and probability tests and the same Markov Chain parameters. Alpha-values (0.05) were divided by the number of populations, implementing a Bonferroni correction, to achieve a critical value to test for significance (alpha = 0.017) across all comparisons (Rice 1989). Heterozygosity estimates (expected and observed) and number of alleles were estimated in Microsatellite Toolkit (Park 2001). *F*-statistics (F_{ST}, F_{IS}) (Weir and Cockerham 1984; Wright 1951) and allelic richness were calculated in FSTAT 2.1 (Goudet 1995) with significance levels of alpha = 0.001 and 10,000 randomizations.

STRUCTURE v2.3 (Falush et al. 2007) was used to examine genetic population structure without *a priori* designation of populations or sampling locations using Bayesian clustering (Pritchard et al. 2000). Optimal number of subpopulations was determined by varying the likely number of clusters or populations (*k*) from 1 to 10 allowing for genetic admixture and correlated allele frequencies (Falush et al. 2003). Each run used a burn-in of 50,000 and a MCMC of 500,000 steps. This process was replicated 10 times for each value of *k* (Evanno et al. 2005) to quantify the standard deviation among the runs for a particular assumed *k*. The optimal number of *k*-clusters was determined by Structure Harvester v0.6.94 (Pritchard et al. 2000), with the method developed by Evanno *et al.* (2005) to evaluate the rate of change in the log probability of the data (Δk) among 10 runs for each assumed *k* and estimate the highest Ln probability of the data, or Ln *P*(d). Individual membership probabilities of the inferred *k*-clusters from the 10 independent replicates were averaged using CLUMPP v1.1.2 (Jakobsson and Rosenberg 2007).

The program TESS 2.3 (Chen *et al.* 2007) was used to account for spatial data when estimating individual membership, whereby spatially close individuals are assumed to have greater probability of being genetically similar. The 'non-admixture' setting was initially used to identify maximum number of clusters (Durand et al. 2009). Optimal number of clusters was determined by varying k from 1 to 10 with 200,000 iterations and a burn-in of 20,000 with 10 replicates for each k. Interaction parameters $\psi = 0$, 0.6, and 1.2 were used to determine the extent to which spatial information influenced individual assignment. The deviance information criterion (DIC) was used to select optimal cluster number. Average membership was again calculated in CLUMPP using 10 % of the runs with the lowest DIC value for optimal k. Each individual was also tested for its status as a resident or recent immigrant using a Bayesian Monte Carlo resampling assignment test of 100,000 simulated individuals (GeneClass2, Piry et al. 2004) and assigning it to the genetic group with highest inferred average ancestry.

Sex-bias dispersal and relatedness estimates

To test for sex-biased dispersal, we first used a genetic assignment-based approach (Favre et al. 1997) to infer dispersed individuals in a population (harvest location differs from genetic origin) by identifying uncommon alleles. An assignment index is calculated for each individual based on average allele frequency. Less common alleles have more negative assignment index values. By-sex and by-age comparisons are expected to show (1) negatively skewed assignment indices for the dispersing sex or age group, and (2) higher variance of assignment indices in the dispersing sex or age group (because it includes both dispersers and residents). GeneClass2 (Piry et al. 2004) was used to calculate assignment index (*AI*) by first calculating probability assignment for

each population. Because there were different numbers of loci across populations (NWAK 10, NWY/SEY 20), and we aimed to assess intrapopulation dynamics of dispersers and residents, each population was run independently for by-population analysis. Population assignment probabilities were determined excluding the current individual's assignment to its sampled population (simulation algorithm; Cornuet et al. 1999) using 100,000 sampled individuals and an *alpha* value of 0.01. These probabilities were log transformed (base 10) and adjusted for categorical variation by subtracting population means; female log-transformed assignment indices were averaged then subtracted from each female, and males likewise. The same transformation was done for age classes, independent of sex. This process allows us to compare variances, avoid rounding errors for very small values, and center the data around zero for calling rare or uncommon alleles. These corrected assignment indices (*AIc*) were binned into 8 discrete classes within by-sex and by-age sorted groups within each geographic locality. Raw frequency of individuals within each bin is reported. Average assignment indices and variance by category were calculated. Differences between groups were tested for significance with 2-tailed t-tests.

Next, we tested how sex-biased dispersal relates to pairwise estimates of genetic relatedness and pairwise geographic distances between individuals, and whether these values supported isolation by distance (IBD) as a method by which populations are genetically structured. Relatedness (*r*) was quantified among individuals to determine family-based structure. One individual in each of a given pair related above $r_{xy} = 0.5$ in IDENTIX v1.1.5 (Belkhir et al. 2002) using the Queller & Goodnight (1989) relatedness estimator after 1000 per-locus bootstraps (to achieve a 95% confidence interval for each)

was labeled a first order relative (i.e., parent-offspring relationship or full siblings). Those of r_{xy} = 0.25 – 0.5 were described as second order relatives, or half-siblings, grandparent-offspring, aunt-offspring, etc. Pairwise distances were measured in Geographic Distance Matrix Generator v1.2.3 (Ersts, AMNH). A Mantel test (R package ape v2.3-1; Paradis et al. 2004) between pairwise relatedness and geographic distances matrices was conducted within populations to test for negative correlation. Higher negative regression (R) values mean higher negative correlation; values closer to zero mean little or no correlation. Within-sex and within-age categories were tested by regressing relatedness on distance. Mean relatedness (IDENTIX v1.1.5; Belkhir et al. 2002) between males and females in pairwise comparisons was estimated in each population for adult and subadult wolverines to test degree of relatedness within anticipated philopatric individuals (*FF*) versus anticipated dispersers (*MM*).

Demographic changes

Bottlenecks were tested among populations using the program BOTTLENECK v1.2.02 (Piry et al. 1999). Reduced numbers of alleles and heterozygosity of loci are seen when populations have experienced bottlenecks or severe reductions in effective population size (Luikart and Cornuet 1998). However, allelic diversity reduces faster than heterozygosity, therefore recent bottlenecks ($2N_e$-$4N_e$ generations) manifest as heterozygosity excess in a given population. Heterozygosity deficit will be present in males if they alone disperse, due to the Wahlund effect (effectively being a mixture of two populations; residents and dispersers) (Li 1955). Heterozygosity excess was tested using a Wilcoxon sign rank test (optimal for fewer than 20 loci; Piry et al. 1999) under a two-phase model of microsatellite evolution (TPM, ideal for testing dinucleotide repeat

loci; Di Rienzo et al. 1994) for 10,000 iterations. The infinite allele model (IAM; Kimura and Crow 1964) is a more liberal model but can indicate recent bottlenecks, and the strict stepwise mutation model (SMM; Ohta and Kimura 1973) is more conservative but can indicate more historical bottlenecks. These models were also tested to screen for consistency in calls for bottlenecks. If SMM and TPM both indicate a bottleneck, it is likely to have occurred and with some historical distance; if IAM alone is significant for heterozygosity excess it could be a false positive or evidence of recent a bottleneck. Variance for TPM was tested at 9 and 30 while proportion of SMM in TPM was left at 80% (Piry et al. 1999; Garza and Williamson 2001).

Source-sink dynamics were examined through the program BIMr 1.0 (Faubet and Gaggiotti 2008) to examine recent rates of movement between groups using gametic disequilibrium. Populations were run through the program as harvest groups (e.g., NWAK, NWY, SEY) and after being analyzed in BAPS 5.3 (Bayesian Analysis of Population Structure; Corander and Marttinen 2006; Corander et al. 2006), a more liberal genetic structure program to identify subpopulations and examine source-sink dynamics on a smaller scale. Pilot runs of 1000 MCMC iterations found acceptance rates between 25% and 45%; a final run of 150,000,000 iterations and a burn-in of 15,000 with 20 replicates was conducted. The run with the lowest Bayesian deviance (D_assign) was selected (Faubet et al. 2007; Faubet and Gaggiotti 2008) to extract parameter estimates. We examined 95% HDPIs to assess significance of asymmetry for pairwise migration rate estimates. Migration rates were also examined for asymmetry between population pairs by examining dyads for the proportion of times a given estimate was greater or less than the other population migration rate estimates at each post-burn-in MCMC (Fordyce

et al. 2011). Estimates were interpreted as the probability that a particular parameter value (migration in one direction) is higher than another value (migration in a second direction).

Results

Demographic statistics

We genotyped 231 individuals from the Yukon Territory and added 129 genotyped individuals from Alaska (Dalerum et al. 2007) for a total of 360 (**Table 1**). Significant departures from HWE or LD were not evident. Heterozygosity did not vary between observed and expected values. Allelic richness was slightly higher, but insignificant, in males compared to females (**Table 1**). Across all populations, males had higher allelic richness than females on a by-locus basis (**S4 Table**). F_{ST} values were greatest between NWAK and Yukon sampling groups (**S5a Table**). F_{ST} in by-sex comparisons showed greater differentiation in comparisons with Yukon females compared to Yukon males, while Alaskan males possessed greater differentiation compared to Alaskan females in 3 out of 4 pairwise relations (**S5b Table**). F_{IS} is negative in outbred populations and positive in philopatric groups (Goudet et al. 2002). SEY showed the most positive F_{IS} values, both in total and by sex, and it differed from the other two groups in having more inbred males comparable to females (**Table 1**).

Interpopulation structure

Three genetic clusters were identified by two clustering methods (STRUCTURE without *a priori* locality data; **Fig 2a**, and TESS; **Fig 2b, 2c** which incorporates individual geographic coordinate data). STRUCTURE inferred Cluster 1 consisting of

NWAK and Clusters 2 & 3 fairly evenly split across NWY and SEY (NWY: Cluster 2—46% average assignment, Cluster 3—33% average assignment; SEY: Cluster 2—39% average assignment, Cluster 3—47% average assignment). TESS also detected three distinctive clusters between NWAK, NWY, and SEY. GeneClass2 identified five dispersers ($p \leq 0.01$). In NWY, a subadult male and female emigrated from NWAK, and in SEY a subadult female and adult male from NWY immigrated, and an adult male from NWAK immigrated to SEY.

Intrapopulation structure

A non-significant difference in assignment index (*AI*) was found between male and female wolverines (NWAK *alpha* = 0.09; NWY *alpha* = 0.46; SEY *alpha* = 0.71), and between subadult and adult individuals for NWAK (*alpha* = 0.31) and SEY (*alpha* = 0.57). For NWY, subadult and adult groups had significantly different assignment values (*alpha* = 0.03). Though differences aren't significant, there is a pattern of the anticipated dispersing groups (males and subadults) having more negative log-transformed *AI* values as well as higher variance in NWAK and NWY (**Table 2**) while SEY demonstrates females and adults having more negative *AI* and higher variance in adults.

Plotted corrected assignment index (*AIc*) values (**Fig 3a, 3b, 3**c) show nonsignificant trends of higher variance in males and subadults. In SEY where the opposite sex and age group are found to have higher variance, both sexes may be dispersing; both *AIc* distributions include immigrants, and thereby more overlap in the distribution of male and female *AIc* values. Female *AIc* values were 39%, 40%, and 36% negative for NWAK, NWY, and SEY respectively (i.e., possessing alleles uncommon to the area they were found) while male *AIc* values were 39%, 36%, and 38% negative.

Adults possessed 40%, 40%, and 38% negative *AIc* values within NWAK, NWY, and SEY respectively; subadults possessed 40%, 41%, and 38% negative values.

In Mantel tests relating genetic relatedness and geographic distance in a sex-biased system, pairs of the further-dispersing sex should show a lower correlation between relatedness and distance than the philopatric sex. However, Mantel tests showed no significant negative correlation, rejecting an isolation by distance (IBD) model. However, negative trends are seen in by-sex and by-age comparisons (**Fig 4**) (**S6 Fig**), weakly supporting a negative relationship between relatedness and distance. There is a weaker, but not significantly different, negative correlation between genetic relatedness and distance in the anticipated disperser (males) compared to the anticipated philopatric sex (females).

Pairwise comparison is related to F_{ST} through the relation $r = 2 F_{ST} / (1 + F_{IT})$ (Queller and Goodnight 1989), and is often used to test for sex-bias differences (Ishibashi *et al.* 1997; Knight *et al.* 1999, Surridge *et al.* 1999). Pairwise comparisons of resident wolverines (i.e., >2 years old; adults) by sex demonstrate no significant differences, but a trend of higher genetic relatedness between male pairwise comparisons across all populations compared to females (contrary to anticipated higher female structure) is found. Males also have a higher percentage of second degree relatives, except for NWY (**Table 3**). Subadults have no significant differences, but females are on average more closely related except in SEY. Prevalence of first and second degree relatives in a sampling group is higher among females except for SEY.

We found evidence of a recent ($2N_e$-$4N_e$ generations) bottleneck in the Yukon populations; both NWY and SEY exhibited heterozygosity excess in BOTTLENECK

v1.2.02 (Piry et al. 1999). With variance of 9 and under the liberal IAM, NWY and SEY showed significant heterozygosity excess ($p < 0.00003$, $p < 0.00001$, respectively), but not under the TPM or the SMM. NWAK showed significant heterozygosity deficiency under the SMM ($p = 0.00244$) suggesting demographic increase due to influx of alleles. When tests were conducted with variance of 30, again heterozygosity excess was seen in SEY only ($p < 0.00001$) under the IAM, as well as under the TPM ($p = 0.00211$) but not under the SMM. NWAK showed significant heterozygosity deficiency under the SMM. In by-sex all-ages tests, Yukon males and females showed significant bottlenecks under the IAM (**S7 Table**). In by-sex by-age tests, only NWY adult females and SEY adult males showed significant heterozygosity excess and evidence of bottleneck.

Source-sink Dynamics

Smaller subpopulations were identified ($k = 6$), but after plotting these individuals in ArcGIS (ESRI 2011) they overlapped extensively, did not show influence of any geographic structure, and support evidence for panmixia in these high latitude environments. Thus, the original three regions were analyzed using the conservative genetic structure approach in STRUCTURE, TESS, and GeneClass2 to delineate individuals and decide population assignment ($k = 3$) for source-sink analysis.

Mean migration rates were consistent across 16 of the 20 BIMR runs with the lowest Bayesian deviances (difference in means across those 16 runs: average = 1.72, min = 0.08, max = 5.3). The run with the lowest Bayesian deviance indicated mean migration rates between the three populations ranged from a low of 4.82% into SEY from NWAK, to a high of 37.86% (proportion of the population that immigrated within the last generation) into SEY from NWY (**Table 4**). We identified asymmetric movement from

SEY into NWAK (no overlapping 95% HDPIs). Although the 95% HDPIs overlapped for all other pairwise estimates, we observed evidence of asymmetrical movement from NWY into NWAK (*alpha* < 0.05) as well. NWY was identified as the largest net provider of immigrants, indicating it was the most substantial source population, whereas NWAK had the largest net immigration indicating it was the largest relative sink population.

Discussion

As solitary, polygynous, and wide-ranging mammals, wolverines are expected to display male-biased dispersal, a finding generally supported by studies in temperate latitudes where telemetry shows both male-biased dispersal and extreme differences in home range size between sexes. In contrast, we found strong support for no differences in sex-biased dispersal in the higher latitude ecoregions of tundra in northwestern Alaska and the boreal and mountain forests of the Yukon Territory. All evidence potentially in support of male-biased or subadult-biased dispersal lacks significance, despite elevated sample size and increased statistical power. In addition, most telemetry studies of wolverine dispersal have concentrated at the southern portion of their range where habitats become more fragmented. This latitudinal skew in studies of sex bias in dispersal and home range potentially limits our understanding of wolverine dispersal dynamics.

Genetic diversity and population structure

Male populations have higher allelic richness, supporting more male dispersers relative to females and supporting male-biased dispersal. Northwest Alaska differed most from the Southeast Yukon population, followed by the Northwest Yukon population in overall F_{ST} comparisons; Northwest Yukon and Southeast Yukon were most similar

which follows from close geographic proximity. In by-sex F_{ST} comparisons, females in the Yukon populations had higher F_{ST} (supporting greater structure of the philopatric sex, as seen by Balloux et al. 1998) while Alaskan males possessed greater structure compared to Alaskan females.

With sex-biased dispersal, a lower mean relatedness is expected between wolverines of the dispersing sex than between individuals of the more philopatric sex. No significant differences between sexes were found in same-sex comparisons of relatedness (*FF* vs. *MM*), so there is no evidence of sex-bias dispersal. In interpopulation analysis, three populations were found; however, by-individual geographic structure was most obvious between Northwest Alaska and the Yukon groups, which again follows due to close geographic proximity. After analyzing individuals for population assignment, only a handful ($N = 5$) of individuals were identified as dispersers, which may be characteristic of high average levels of diverse alleles.

Sex-biased dispersal

Assignment indices by-sex and by-age across all populations showed no significant differences except for Northwest Yukon in the subadult/adult comparison, whereby subadults showed greater dispersal. Though lacking significance, a trend of slightly more negative values and higher variances for males and for subadults was seen across Northwest Alaska and Northwest Yukon. Subadults demonstrate greater variance in genotype, supporting subadults as variable dispersers. The subadult class includes pre-dispersal (young of year), dispersing, and post-dispersal (settled) individuals, so higher variance is expected. In contrast, Southeast Yukon females and adults had more negative *AIc* values and adults had higher variance. More equality between sexes in dispersal may

reflect these differences; both *AIc* distributions include immigrants and thereby overlap male and female *AIc* values. In Mantel tests of pairwise distance regressed on pairwise relatedness, all populations were non-significant but again showed trends of more negative regression values for the philopatric sex, supporting weak male-biased dispersal. Over all populations, Southeast Yukon showed the highest female negative correlation between relatedness and pairwise distance and was the population most reflecting IBD as a method for genetic structuring (i.e., greatest female philopatry).

Inbreeding (F_{IS}) was highest among the Southeast Yukon population, in total and by sex. While other populations followed the expectation of females having higher F_{IS} values, Southeast Yukon had slightly higher inbreeding coefficient in males. Bottleneck results suggest a recent bottleneck in the Yukon populations but a demographic increase in the northwest Alaska population, which seems to have occurred farther in the past relative to the Yukon bottleneck. To be detectable for these tests, effective population size must drop to 20 individuals (for a single year) or 30 individuals (for 20 years) to observe a bottleneck with 95% confidence (Hoelzel 1993). Zigouris et al. (2012) previously found high female philopatry in the southwestern part of the wolverine's North American range (British Columbia, Southeast Alaska, Alberta) compared to the rest of the range, which may contribute to lingering signature of bottlenecks.

Source-Sink Dynamics

Our goal was to estimate the amount of asymmetric movement of wolverines between populations to identify source and sink dynamics and begin to explore the impact of trapper harvest and regional refuge habitats on these processes. Because these values are relative to individuals sampled, generalizations should be interpreted with

caution. We observed significant rates of asymmetric movement among subpopulations, which is consistent with the prediction that metapopulation source-sink dynamics are present in higher latitudes. Although we reject our hypothesis predicting Southeast Yukon would function as a source of dispersing individuals, the most notable asymmetry in movement rates occurred in and out of Southeast Yukon with Northwest Alaska (**Table 4**). This may correlate with patterns of hunting pressure on wolverines (Banci 1981). Northwest Yukon was the most substantial source population; this may be because it has open space, lacks extensive industrial development, and has low harvest pressure found in Yukon especially at higher latitudes (Krebs et al. 2004). Northwest Alaska is the largest sink population, likely due to lower carrying capacity and recruitment (Magoun 1985) and high rates of human-caused mortality among young females (0.50 harvest) in trapped areas (Krebs et al. 2004).

Source populations are important to identify because they have a disproportionate impact on other, otherwise not self-sustaining populations (i.e., sink populations). The southern reaches of the wolverine's North American geographic range are especially affected by low population density and lack of immigrants; gene flow appears to be via male dispersal, making demographic viability dependent on the movement of females into lower latitudes (Cegelski et al. 2006). However, any such movement may be minimized by female preference for areas with heavier spring snowpack for denning sites as well as accessibility through corridors and availability of food (Inman et al. 2013).

Methods Discussion

Goudet et al. (2002) indicate that sex-biased dispersal must be intense to be detected by our genetic methods. For example, if only one sex disperses (100:0) then bias

can be easily detected provided sampling is extensive. However, bias is much more difficult to detect when intensity drops to 80:20 (Goudet et al. 2002) in which case only F_{ST} and *mAIc* are capable at detecting bias, though Mossman et al. (1999) maintained AIc's usefulness as a sensitive test for biased dispersal in the North American deermouse (*Peromyscus maniculatus*). Success or failure of dispersal tests also depends largely on sampling; sampling bias has a strong effect, with sampling large numbers of individuals per population being the most effective method for reducing bias (Goudet et al. 2002). Our *N* is reasonable for Yukon sampling (Goudet et al. 2002; NWY 24M, 42m, 19F, 29f; SEY 28M, 48m, 22F, 31f; 270,000 km^2), but may be biased in Northwest Alaska with a more limited sampling area and male subadults dominating sampling (NWAK 12M, 64m, 8F, 33f; 80,000 km^2). The number of loci examined also has an effect on our ability to detect dispersal biases (Goudet et al. 2002). Therefore, the fewer loci for Northwest Alaska (10 compared to 20) may make sex-bias differences harder to detect, but future investment should focus on increasing number of individuals examined over increasing the number of loci screened. Another important caveat is the markers chosen for this study. Studies based solely on microsatellite (nuclear) data complicate the detection of sex biased dispersal as the adult population resets each generation without matrilineal, clonally-inherited mitochondrial data (Zink and Barrowclough 2008).

Low density populations, like wolverines, may obscure possible disparities in dispersal because both sexes are dispersing to colonize empty ranges. A similar pattern is seen in common brushtail possums (*Trichosurus vulpecula*) where dispersal in occupied areas is male-biased but lacks bias when an area is being colonized (Ji et al. 2001). Though previous studies on wolverines have established a strong record of female

philopatry (Banci 1994) and male exploration and dispersal (Banci 1987), this trait may vary by habitat. Dispersing ecology and territory maintenance of North American wolverines has been directly tracked and observed predominately in lower latitudes of their range (i.e., Montana, Wyoming, Idaho, southern Alberta and British Columbia; see **Fig 6** and **S9 Table**) and perhaps these differences do not hold at higher latitudes where habitats are less fragmented.

Conservation Implications

Female home range selection and dispersal are motivated by secure, familiar habitats and the ability to feed and rear kits successfully (Inman et al. 2013), while male home range selection and dispersal are motivated by access to females (Erlinge and Sandell 1986; Lawson Handley and Perrin 2007; Sandell and Liberg 1992). If habitat quality is low or more fragmented, perhaps females will remain more philopatric to a resource-rich and familiar environment, and males will range more widely to make contact with patchily-distributed females (**Fig 5**). The dispersal disparity between male and female wolverines may be exaggerated by habitat constraints in lower latitudes where human disturbance and distance to optimal habitat are amplified (Inman et al. 2013). There may then be less dispersal difference between the sexes when habitats are high quality (i.e., at remote, high-latitude forest and tundra). Expectations that males disperse farther than females may stem at least in part from a focus on lower-latitude studies where habitat is lower quality and more fragmented than in our analyses.

Previous telemetry and tracking studies on wolverines (**Fig 6** and **S9 Table**) show that the sizes of home ranges and natal dispersal distances in North America vary regionally, with sex-biased dispersal most prominent at lower latitudes. When ratios of

female-to-male home ranges were compared across ecoregions and latitudes, the discrepancy between male and female decreased as latitude increased but ecoregion remained the same. Female range size was 33% the size of male ranges at low latitude forests (12 studies, $N = 277$ wolverines) and more than doubled to 72% the size of male ranges in high latitude forests (5 studies, $N = 30$ wolverines) (**S8 Table**), though increased sample size of wolverines tracked would strengthen these patterns. High latitude populations should be further studied for full understanding of wolverine population dynamics range-wide. Since natal dispersal links population dynamics to landscape connectivity (Merrick and Koprowski 2017), understanding what motivates dispersal or is perceived as a barrier to movement is important for managing increasingly fragmented populations.

Conclusions

Through the use of allele frequency and rarity in a population, no difference between male and female wolverine dispersal was found in northwestern Alaska and the Yukon Territory. We were able to address some of the reasons cited by Dalerum et al. (2007) to explain why male-biased dispersal was not supported; increased sample size, increased loci, and increased study area did not change the result of sex equality in dispersal. Our methods (Goudet et al. 2002; Lawson-Handley and Perrin 2007) support wolverines at this latitude as nearly panmictic by sex and population. This may be a model for healthy wolverine populations with high quality, non-fragmented habitat. In lower latitudes, higher fitness for more philopatric females drives smaller home ranges in females relative to males attempting to reach an adequate number of breeding females.

Under such a model, habitat fragmentation encourages skew in sex-bias dispersal and home range size.

Average home range sizes are often larger in lower latitude populations compared to higher ones (Copeland 1996; Inman et al. 2013; Rohrer et al 2007), leading to greater exposure to competitors (coyotes, bobcats, wolves, cougars) and predators (wolves, cougars, humans) (Inman et al. 2012). Of all biotic factors with the potential to affect wolverines, human predation causes the highest mortality (Van Zyll de Jong 1975; Krebs et al. 2004). Across twelve North American radiotelemetry studies, annual survivorship rates were lower in trapped (<0.75 for all age-sex classes) than in untrapped areas (>0.84 for all age-sex classes) (Krebs et al. 2004). Wolverines are highly susceptible to trapping because they travel widely and, as scavengers, are readily attracted to baits (Hornocker and Hash 1981). Un-trapped populations are potentially capable of increasing at 6.4% per year, while heavily harvested regions can decrease at up to 12.2% per year (Krebs et al. 2004), indicating the need for a refuge from trapping pressure to cover twice as much similarly productive wolverine habitat as harvested areas to support harvests. Weaver et al. (1996) suggest that game sanctuaries for many carnivores are needed to sustain harvest or even natural mortality rates. Implementation of wolverine tracking studies (Montgomery et al. 2010) in high-latitude, more continuous populations would independently test by-sex dispersal disparities.

Wolverines have been identified as vulnerable and are considered a species with population viability concerns, at least in lower latitudes (Cegelski et al. 2006). Population size in the contiguous United States is less than 300 (USGWS 2013) with an estimated effective population size (i.e., breeding-age males and females) of approximately 35

individuals (Schwartz et al. 2009). Climate change already impacts gene flow in wolverines between fragmented refuges in Glacier National Park, Yellowstone National Park, and the Bob Marshall Wilderness (McKelvey et al. 2011; Aubry et al. 2007). In April 2016, the United States Fish and Wildlife Service denied protection of the wolverine under the Endangered Species Act (Act 1973), which was then overturned. Following a public comment period ending November 2016 a new decision will soon be made (USFWS 2016), and additional information on the demography, growth, dispersal, and sustainability of the species across its North American range is necessary to make an informed decision regarding its management status. Wolverines are adapted to habitats that have become highly fragmented in lower latitudes as a consequence of human and climatic factors, and habitat fragmentation has great implications for wolverine dispersal dynamics. A powerful means of identifying and understanding populations at risk is to compare them to populations not at risk (in this case, at risk due to human disturbance and habitat fragmentation, and for the time being; Krebs et al. 2004). Addressing landscape-scale issues becomes more pressing as climate change threatens to increase fragmentation of many populations (Opdam and Wascher 2004).

Literature Cited

Act, Endangered Species. 1973. US code. Title 16: 1531–1544.

Aubry, K. B., K. S. Mckelvey, and J. P. Copeland. 2007. Distribution and broadscale habitat relations of the wolverine in the contiguous United States. Journal of Wildlife Management 71(7): 2147–2158.

Ballard, W. B., L. A. Ayres, P. R. Krausman, D. J. Reed, and S. G. Fancy. 1997. Ecology of wolves in relation to a migratory caribou herd in northwest Alaska. Wildlife Monographs 135: 3–47.

Balloux, F., J. Goudet, and N. Perrin. 1998. Breeding system and genetic variance in the monogamous, semi-social shrew, *Crocidura russula*. Evolution 52(4): 1230–1235.

Banci, V. 1987. Ecology and behavior of wolverine in Yukon. Masters thesis, Simon Fraser University, Burnaby, British Columbia, Canada. 178pp.

Banci, V. 1994: Wolverine (Chapter 5). Eds. Ruggiero, L.F., K. B Aubrey, S. W.

Buskirk, L. J. Lyon, and W. J. Zielinski, W.J. The scientific basis for conserving forest carnivores: American marten, fisher, lynx, and wolverine in the western United States. U.S. Department of Agriculture, Forest Service, Fort Collins, Colorado, USA. 99–127.

Banci, V., and A. S. Harestad. 1990. Home range and habitat use of wolverines *Gulo gulo* in Yukon, Canada. Holarctic Ecology 13(3): 195–200.

Belkhir, K., V. Castric, and F. Bonhomme. 2002. IDENTIX, a software to test for relatedness in a population using permutation methods. Molecular Ecology Notes 2(4): 611–614.

Carrara, P. E., T. A. Ager, and J. F. Baichtal. 2007. Possible refugia in the Alexander Archipelago of southeastern Alaska during the late Wisconsin glaciation. Canadian Journal of Earth Sciences 44: 229–244.

Castillo, D. F., M. Lucherini, E. M. Luengos Vidal, C. Manfredi, and E. B. Casanave. 2011. Spatial organization of Molina's hog-nosed skunk (*Conepatus chinga*) in two landscapes of the Pampas grassland of Argentina. Canadian Journal of Zoology 89: 229–238.

Cegelski, C. C., L. P. Waits, N. J. Anderson, O. Flagstad, C. Strobeck, and C. J. Kyle. 2006. Genetic diversity and population structure of wolverine (*Gulo gulo*) populations at the southern edge of their current distribution in North America with Implications for genetic viability. Conservation Genetics 7(2): 197–211.

Chadwick, D. H. 2010. The Wolverine Way. Ventura, CA: Patagonia Books.

Chappell, D. E., R. A. Van Den Bussche, J. Krizan, and B. Patterson. 2004. Contrasting levels of genetic differentiation among populations of wolverines (*Gulo gulo*) from northern Canada revealed by nuclear and mitochondrial loci. Conservation Genetics 5(6): 759–767.

Chen, C., E. Durand, F. Forbes, and O. François. 2007. Bayesian clustering algorithms ascertaining spatial population structure: a new computer program and a comparison study. Molecular Ecology Notes *7*(5): 747–756.

Clevenger, A. P. 2013. Mitigating highways for a ghost: data collection challenges and

implications for managing wolverines and transportation corridors. Northwest Science 87(3): 257–264.

Cooley, D. A. unpublished data. Yukon Department of Renewable Resources.

Copeland, J. P. 1993. Assessment of snow-tracking and remote camera systems to document presence of wolverines at carrion bait stations. Idaho Department of Fish and Game.

Copeland, J. P. 1996. Biology of the wolverine in central Idaho. Masters thesis, University of Idaho, Moscow, Idaho, USA. 152pp.

Copeland, J. P. and F. Edelmann. 1999. Wolverine survey in the Seven Devils Mountains of Hells Canyon. Technical Report Appendix E.3.2-29. 14pp.

Copeland, J. P., and R. E. Yates. 2008. Wolverine population assessment in Glacier National Park, Comprehensive Summary Update. USDA Forest Service, Rocky Mountain Research Station, Missoula. Montana, USA.

Copeland, J. P., K. S. McKelvey, K. B. Aubry, A. Landa, J. Persson, R. M. Inman, J. Krebs, E. Lofroth, H. Golden, J.R. Squires, A. Magoun, M. K. Schwartz, J. Wilmot, C. L. Copeland, R. E. Yates, I. Kojola, R. May. 2010. The bioclimatic envelope of the wolverine (*Gulo gulo*): Do climatic constraints limit Its geographic distribution? Canadian Journal of Zoology 88(3): 233–46.

Copeland, J. P., Yates, R. E., and Ruggiero, L. 2008. Wolverine population assessment in Glacier National Park. *Comprehensive Summary Update,* USDA Forest Service, Rocky Mountain Research Station, Missoula, Montana, USA. 16pp.

Copeland, J. P., J. M. Peek, C. R. Groves, W. E. Melquist, K. S. McKelvey, G. W. McDaniel, C. D. Long, C. E. Harris. 2007. Seasonal habitat associations of the wolverine in central Idaho. The Journal of Wildlife Management 71(7): 2201–2212.

Corander, J., M. Gyllenberg, and T. Koski. 2006. Bayesian model learning based on a parallel MCMC strategy. Statistics and Computing 16(4): 355–362.

Corander, J., and P. Marttinen. 2006. Bayesian identification of admixture events using multilocus molecular markers. Molecular ecology 15(10): 2833–2843.

Cornuet, J., S. Piry, G. Luikart, A. Estoup, and M. Solignac. 1999. Populations as origins of individuals. Genetics Society of America 153: 1989–2000.

Danzinger, L. J. 2011. Using GIS to examine potential wolverine habitat in Colorado: an analysis of habitat fragmentation and wildlife corridors. Papers in Resource Analysis, Saint Mary's University of Minnesota University Central Services Press. Winona, Minnesota, USA. 13: 11pp.

Dalerum, F., J. Loxterman, B. Shults, K. Kunkel, and J. A. Cook. 2007. Sex-specific dispersal patterns of wolverines: insights from microsatellite markers. Journal of Mammalogy 88(3): 793–800.

Dallas, J. F., and S. B. Piertney. 1998. Microsatellite primers for the Eurasian otter. Molecular Ecology 7(9): 1248–1251.

Davis, C. S., and C. Strobeck. 1998. Isolation, variability, and cross-species amplification of polymorphic microsatellite loci in the family Mustelidae. Molecular Ecology 7(12): 1776–1778.

Dawson, F. N., A. J. Magoun, J. Bowman, and J. C. Ray. 2010. Wolverine, *Gulo gulo*, home range size and denning habitat in lowland boreal forest in Ontario. The Canadian Field Naturalist 124(2): 139–144.

Di Rienzo, A., A. C. Peterson, J. C. Garzat, A. M. Valdes, M. Slatkint, and N. B. Freimer. 1994. Mutational processes of simple-sequence repeat loci in human populations. Proceedings of the National Academy of Science Genetics 91: 3166–3170.

Dias, P. C. 1996. Sources and sinks in population biology. Trends in Ecology and Evolution 11(8): 326–330.

Dobson, F. S. 1982. Competition for mates and predominant juvenile male dispersal in mammals. Animal Behavior 30(4): 1183–1192.

Duffy, A. J., A. Landa, M. O'Connell, C. Stratton, and J. M. Wright. 1998. Four polymorphic microsatellites in wolverine, *Gulo gulo*. Animal genetics 29: 63–72.

Durand, E., F. Jay, O. E. Gaggiotti, and O. François. 2009. Spatial inference of admixture proportions and secondary contact zones. Molecular Biology Evolution 26(9): 1963–1973.

Eberhardt, L. L. 1990. Survival rates required to sustain bear populations. Journal of Wildlife Management 54(4): 587–590.

Erlinge, S., and M. Sandell. 1986. Seasonal changes in the social organization of male stoats, *Mustela erminea*: an effect of shifts between two decisive resources. Nordic Society Oikos 47(1): 57–62.

Ersts, P. J. [Internet] Geographic Distance Matrix Generator (version 1.2.3). American Museum of Natural History, Center for Biodiversity and Conservation.

ESRI. 2011. ArcGIS Desktop: Release 10.4.1 Desktop. Redlands, CA: Environmental Systems Research Institute.

Evanno, G., S. Regnaut, and J. Goudet. 2005. Detecting the number of clusters of individuals using the software STRUCTURE: a simulation study. Molecular Ecology 14(8): 2611–2620.

Falush, D., M. Stephens, and J. K. Pritchard. 2003. Inference of population structure using multilocus genotype data: linked loci and correlated allele frequencies. Genetics 164(4): 1567–1587.

Falush, D., M. Stephens, and J. K. Pritchard. 2007. Inference of population structure using multilocus genotype data: dominant markers and null alleles. Molecular Ecology Notes 7(4): 574–578.

Fancy, S. G., L. F. Pank, K. R. Whitten, and W. L. Regelin. 1989. Seasonal movements of caribou in arctic Alaska as determined by satellite. Canadian Journal of Zoology 67(3): 644–650.

Faubet, P., and O. E. Gaggiotti. 2008. A new Bayesian method to identify the environmental factors that influence recent migration. Genetics 178(3): 1491–1504.

Faubet, P., R. S. Waples, and O. E. Gaggiotti. 2007. Evaluating the performance of a multilocus Bayesian method for the estimation of migration rates. Molecular Ecology 16(6): 1149–1166.

Favre, L., F. Balloux, J. Goudet, and N. Perrin. 1997. Female-biased dispersal in the monogamous mammal *Crocidura russula*: evidence from field data and microsatellite patterns. Proceedings of the Royal Society of London, Biological Sciences 264: 127–132.

Fischbach, A. S., C. V. Jay, J. V. Jackson, L. W. Andersen, G. K. Sage, and S. L. Talbot. 2008. Molecular method for determining sex of walruses. Journal of Wildlife Management 72(8): 1808–1812.

Fleming, M. A., and J. A. Cook. 2002. Phylogeography of endemic ermine (*Mustela erminea*) in southeast Alaska. Molecular Ecology 11(4): 795–807.

Fleming, M. A., E. A. Ostrander, and J. A. Cook. 1999. Microsatellite markers for American mink (*Mustela vison*) and ermine (*Mustela erminea*). Molecular Ecology 8(8): 1352–1355.

Fordyce, S. L., M. C. Ávila-Arcos, E. Rockenbauer, C. Børsting, R. Frank-Hansen, F. T. Petersen, E. Willerslev, A. J. Hansen, N. Morling, and M. T. P. Gilbert. 2011. Material and methods Samples. Biotechniques 51(2): 127–133.

Gardner, C. L. 1985. The ecology of wolverines in southcentral Alaska. Masters thesis, University of Alaska, Fairbanks, Alaska, USA. 91pp.

Garza, J. C., and E. G. Williamson. 2001. Detection of reduction in population size using data from microsatellite loci. Molecular ecology 10(2): 305–318.

Golden, H. N., W. T. Route, and E. F. Becker. 1993. Wolverine demography and ecology in southcentral Alaska: project outline and phase I progress report. Alaska Department of Fish and Game, Division of Wildlife Conservation, and National Park Service, Wrangell-St. Elias National Park and Preserve.

Goudet, J. 1995. FSTAT (version 1.2): a computer program to calculate F-statistics. Journal of Heredity 86(6): 485–486.

Goudet, J., N. Perrin, and P. Waser. 2002. Tests for sex-biased dispersal using bi-parentally inherited genetic markers. Molecular Ecology 11(6): 1103–1114.

Greenwood, P. J. 1980. Mating systems, philopatry and dispersal in birds and mammals. Animal Behaviour 28(4): 1140–1162.

Hoelzel, A.R., J. Halley, S. J. O'Brien, C. Campagna, T. Arnborm, B. Le Boeuf, K. Ralls, G. A. Dover. 1993. Elephant seal genetic variation and the use of simulation models to investigate historical population bottlenecks. Journal of Heredity 84(6): 443–449.

Hornocker, M. G., and H. S. Hash. 1981. Ecology of the wolverine in northwestern Montana. Canadian Journal of Zoology 59(7): 1286–1301.

Inman, K. H., R. M. Inman, R. R. Wigglesworth, A. J. McCue, B. L. Brock, J. D. Rieck, W. Harrower. 2003. Greater Yellowstone Wolverine Study. The Wolverine Foundation Cumulative Progress Report, Wildlife Conservative Society. 18pp.

Inman, R. M. 2013. Doctoral dissertation, Swedish University of Agricultural Sciences, Uppsala, Sweden. 48pp.

Inman, R. M., B. L. Brock, K. H. Inman, S. S. Sartorius, B. C. Aber, B. Giddings, S. L. Cain, M. L. Orme, J. A. Fredrick, B. J. Oakleaf, K. L. Alt, E. Odell, G. Chapron. 2013. Developing priorities for metapopulation conservation at the landscape scale: Wolverines in the Western United States. Biological Conservation 166: 276–286.

Inman, R. M., M. L. Packila, K. H. Inman, A. J. McCue, G. C. White, J. Persson, B. C. Aber, M. L. Orme, K. L. Alt, S. L. Cain, J. A. Fredrick, B. J. Oakleaf, S. S. Sartorius. 2012. Spatial ecology of wolverines at the southern periphery of distribution. Journal of Wildlife Management 76(4): 778–792.

Ishibashi, Y., T. Saitoh, S. Abe, M. C. Yoshida. 1997. Sex-related spatial kin structure in a spring population of grey-sided voles *Clethrionomys rufocanus* as revealed by mitochondrial and microsatellite DNA analyses. Molecular Ecology 6(1): 63–71.

Jackson, J. V., S. L. Talbot, and S. Farley. 2008. Genetic characterization of Kenai brown

bears (*Ursus arctos*): microsatellite and mitochondrial DNA control region variation in brown bears of the Kenai Peninsula, south central Alaska. Canadian Journal of Zoology 86(7): 756–764.

Jakobsson, M., and N. A. Rosenberg. 2007. CLUMPP : a cluster matching and permutation program for dealing with label switching and multimodality in analysis of population structure. Bioinformatics 23(14): 1801–1806.

Ji, W., S. D. Sarre, N. Aitken, R. K. S. Hankin, and M. N. Clout. 2001. Sex-biased dispersal and a density-independent mating system in the Australian brushtail possum, as revealed by minisatellite DNA profiling. Molecular Ecology 10(6): 1527–1537.

Josenhans, H. W., D. W. Fedje, K. W. Conway, and J.V. Barrie. 1995. Post glacial sea levels on the western Canadian continental shelf: evidence for rapid change, extensive subaerial exposure, and early human habitation. Marine Geology 125(1–2): 73–94.

Karabensh, B. 2015. Habitat selection of wolverines: a geospatial analysis of wolverine movements in southwest Montana. Master's thesis, University of Denver, Denver, Colorado, USA. 61pp.

Kimura, M., and J. F. Crow. 1964. The number of alleles that can be maintained in a finite population. Genetics 49(4): 725–738.

Knight, M. E., M. J. H. van Oppen, H. L. Smith, C. Rico, G. M. Hewitt, G. F. Turner. 1999. Evidence for male-biased dispersal in Lake Malawi cichlids from microsatellites. Molecular Ecology 8(9): 1521–1527.

Krebs, J. A., E. C. Lofroth, J. P. Copeland, V. Banci, D. A. Cooley, H. N. Golden, A. J. Magoun, R. Mulders, and B. Shults. 2004. Synthesis of survival rates and causes of mortality in North American wolverines. Journal of Wildlife Management 68(3): 493–502.

Krebs, J. A., E. C. Lofroth, I. Parfitt. 2007. Multiscale habitat use by wolverines in British Columbia, Canada. Journal of Wildlife Management 71(7): 2180–2192.

Krebs, J. A. and D. Lewis 2000. Wolverine ecology and habitat use in the North Columbia Mountains. Proceedings of the Biology and Managememnt of Species and Habitats at Risk, Kamloops, B.C., 15–19.

Kyle, C. J., J. F. Robitaille, and C. Strobeck. 2001. Genetic variation and structure of fisher (*Martes pennanti*) populations across North America. Molecular Ecology 10(9): 2341–2347.

Kyle, C J, and C. Strobeck. 2002. Connectivity of peripheral and core populations of

North American wolverines. Journal of Mammalogy 83(4): 1141–1150.

Lawson Handley, L. J., and N. Perrin. 2007. Advances in our understanding of mammalian sex-biased dispersal. Molecular Ecology 16(8): 1559–1578.

Lewis, S. B, R. W. Flynn, and N. L. Barten. 2009. Wolverine monitoring for the Junaeu access improvements project. Wildlife Research Annual Progress Report, Alaska Department of Fish and Game. 22pp.

Li, C. C. 1955. Population Genetics. Chicago, IL: University of Chicago Press.

Lofroth, E.C., D. Wellwood, W. Harrower, and C. Hoodicoff. 2000. Northern Wolverine Project: wolverine ecology in plateau and foothill landscapes 1996–1999. Year End Final Report. 104pp.

Lofroth, E. C., and J. Krebs 2007. The abundance and distribution of wolverines in British Columbia, Canada. Journal of Wildlife Management 71(7): 2159–2169.

Luikart, G., and J. M. Cornuet. 1998. Empirical evaluation of a test for identifying recently bottlenecked populations from allele frequency data. Conservation Biology 12(1): 228–37.

Magoun, A. J. 1985. Population characteristics, ecology, and management of wolverines in northwestern Alaska (Doctoral dissertation, University of Alaska).

Magoun, A. J. 1987. Summer and winter diets of wolverines, *Gulo gulo*, in arctic Alaska. Canadian field-naturalist. Ottawa ON 101(3): 392–397.

Mandryk, C. A., H. Josenhans, D. W. Fedje, and R. W. Mathewes. 2001. Late Quaternary paleoenvironments of Northwestern North America : implications for inland versus coastal migration routes. Quaternary Science Reviews 20(1–3): 301–314.

Matson, G. 1981. Workbook for cementum analysis. Matson's, Milltown, Montana, USA.

Mazza, Ed. 2016. "North Dakota's 1st Wolverine in 150 Years is Immediately Killed," Inman comments. The Huffington Post.

McKelvey, K. S., J. P. Copeland, M. K. Schwartz, J. S. Littell, K. B. Aubry, J. R. Squires, S. A. Parks, M. M. Elsner, and G. S. Mauger. 2011. Climate change predicted to shift wolverine distributions, connectivity, and dispersal corridors. Ecological Applications 21(8): 2882–2897.

Melillo, J. M., A. D. McGuire, D. W. Kicklighter, B. Moore III, C. J. Vorosmarty, and A. L. Schloss. 1993. Global climate change and terrestrial net primary production. Nature 363: 234–240.

Merrick, M. J., and J. L. Koprowski. 2017. Circuit theory to estimate natal dispersal routes and functional landscape connectivity for an endangered small mammal. Landscape Ecology 32(6): 1163–11179.

Montgomery, R. A., G. J. Roloff, J. M. Ver Hof, and J. J. Millspaugh. 2010. Journal of Wildlife Management 74(8): 1917–11925.

Moore, J., and R. Ali. 1984. Are dispersal and inbreeding avoidance related? Animal Behaviour 32(1): 94–112.

Moriarty, K. M., W. J. Zielinski, A. G. Gonzales, T. E. Dawson, K. M. Boatner, C. A. Wilson, F. V. Schlexer, K. L. Pilgrim, J. P. Copeland, M. K. Schwartz. 2009. Wolverine confirmation in California after nearly a century: native or long-distance immigrant? Northwest Science 83(2): 154–162.

Mossman, C. A., and Waser, P. M. 1999. Genetic detection of sex-biased dispersal. Molecular Ecology 8(6): 1063–1067.

Mulders, R. 2001. Wolverine ecology, distribution, and productivity in the Slave Geological Province. Final Report to the West Kitikmeot/Slave Study Society. Yellowknife, Northwest Territories, Canada. 92pp.

Murphy, K., J. Wilmot, J. Copeland, D. Tyers, and J. Squires. 2011. Wolverines in Greater Yellowstone. Yellowstone Science 19(3): 8pp.

Musiani, M., J. A. Leonard, H. D. Cluff, C. C. Gates, S. Mariani, P. C. Paquet, C. Vilà, R. K. Wayne. 2007. Differentiation of tundra/taiga and boreal coniferous forest wolves: genetics, coat colour and association with migratory caribou. Molecular Ecology 16(19): 4149–4170.

Nilsen, E. B., I. Herfindal, and J. D. C. Linnell. 2005. Can intra-specific variation in carnivore home-range size be explained using remote-sensing estimates of environmental productivity? Ecoscience 12(1): 68–75.

Nunney, L. 1993. The influence of mating system and overlapping generations on effective population size. Evolution 47(5): 1329–1341.

Oetting, W. S., H. K. Lee, D. J. Flanders, G. L. Wiesner, T. A. Sellers, R. A. King. 1995. Linkage analysis with multiplexed short tandem repeat polymorphisms using infrared fluorescence and M13 tailed primers. Genomics 30(3): 450–458.

Ohlemüller, R., B. Huntley, S. Normand, and J. C. Svenning. 2012. Potential source and sink locations for climate-driven species range shifts in Europe since the Last Glacial Maximum. Global Ecology and Biogeography 21(2): 152–163.

Ohta, T., and M. Kimura. 1973. A model of mutation appropriate to estimate the number

of electrophoretically detectable alleles in a finite population. Genetical Research 22(2): 201–204.

Opdam, P., and D. Wascher. 2004. Climate change meets habitat fragmentation: linking landscape and biogeographical scale levels in research and conservation. Biological Conservation 117(3): 285–297.

Paradis, E., J. Claude, and K. Strimmer. 2004. APE: analyses of phylogenetics and evolution in R language. Bioinformatics 20(2): 289–290.

Park, S. D. E. 2001. The Excel microsatellite toolkit. *Trypanotolerance in West African Cattle and the Population Genetic Effects of Selection*.

Pasitschniak-Arts, M., and S. Larivière. 1995. *Gulo gulo* Species Account. The American Society of Mammalogists 499: 10pp.

Piry, S., A. Alapetite, J. M. Cornuet, D. Paetkau, L. Baudouin, and A. Estoup. 2004. GENECLASS2: a software for genetic assignment and first-generation migrant detection. Journal of Heredity 95(6): 536–539.

Piry, S., G. Luikart, and J. M. Cornuet. 1999. Bottleneck: a computer program for detecting recent reductionsin the effective population size using allele frequency data. Journal of Heredity 90(4): 502–503.

Pritchard, J. K., M. Stephens, and P. Donelly. 2000. Inference of population structure using multilocus genotype data. Genetics 155(2): 945–59.

Queller, D. C., and K. F. Goodnight. 1989. Estimating relatedness using genetic markers. Evolution 43(2): 258–275.

Quick, H. F. 1953. Wolverine, fisher and marten studies in a wilderness region. Transactions of the North American Wildlife Conference. 18: 513–532.

Rausch, R. A., and A. M. Pearson. 1972. Notes on the wolverine in Alaska and the Yukon Territory. The Journal of Wildlife Management 36(2): 249–268.

Rauset, G. R., M. Low, and J. Persson. 2015. Reproductive patterns result from age-related sensitivity to resources and reproductive costs in a mammalian carnivore. Ecology 96(12): 3153–3164.

Raymond, M., and F. Rousset. 1995. An exact test for population differentiation. Society for the Study of Evolution 49(6): 1280–1283.

Rice, W. R. 1989. Analyzing tables of statistical tests. Evolution 43(1): 223–225.

Rohrer, J., K. Aubry, and C. Raley. 2007. Distribution and ecology of wolverines in the

North Cascades: Pilot Project - Year 2. Okanogan-Wenatchee National Forests and Methow Valley Ranger District Year-End Status Report. 23pp.

Sandell, M., and O. Liberg. 1992. Roamers and stayers: a model on male mating tactics and mating systems. The American Naturalist 139(1): 177–189.

Schwartz, M. K., and K. S. McKelvey. 2009. Why sampling scheme matters: the effect of sampling scheme on landscape genetic results. Conservation Genetics 10(2): 441–52.

Scrafford, M. A., and M. S. Boyce. 2014. Effects of industry on wolverine (*Gulo gulo*) ecology in the boreal forest of northern Alberta. Progress report 2013/2014. 20pp.

Scrafford, M. A., T. Avgar, B. Abercrombie, J. Tigner, and M. S. Boyce. 2017. Wolverine habitat selection in response to anthropogenic disturbance in the western Canadian boreal forest. Forest Ecology and Management 395(1): 27–36.

Shaw, E. P. and J. Ford. 2012. The Lone Wolverine. Ann Arbor, MI: University of Michigan Press.

Shaw, C. N., P. J. Wilson, B. N. White. 2003. A reliable molecular method of gender determination for mammals. Journal of Mammalogy 84(1): 123–128.

Shults, B. Unpublished wolverine telemetry data from northwest Alaska.

Surridge, A. K., K. M. Ibrahim, D. J. Bell, N. J. Webb, C. Rico, G. M. Hewitt. 1999. Fine-scale genetic structuring in a natural population of European wild rabbits (*Oryctolagus cuniculus*). Molecular Ecology 8(2): 299–307.

Squires, J. R., J. P. Copeland, T. J. Ulizio, M. K. Schwartz, and L. F. Ruggiero. 2007. Sources and patterns of wolverine mortality in western Montana. Journal of Wildlife Management 71(7): 2213–2220.

US Fish and Wildlife Service. 2013. Endangered and Threatened Wildlife and Plants; Threatened Status for the Distinct Population Segment of the North American Wolverine Occurring in the Contiguous United States; Establishment of a Nonessential Experimental Population of the North American Wolverine in Colorado, Wyoming, and New Mexico. Federal Register 78(23): 7864–7890.

US Fish and Wildlife Service. 2014. Fish and Wildlife Service. 2013. Endangered and Threatened Wildlife and Plants; Threatened Status for the Distinct Population Segment of the North American Wolverine Occurring in the Contiguous United States; Establishment of a Nonessential Experimental Population of the North American Wolverine in Colorado, Wyoming, and New Mexico. Federal Register 79(156): 47522–47545.

Van Oosterhout, C., W.F. Hutchinson, D.P. M. Wills, P. Shipley. 2004. MICRO-CHECKER : Software for identifying and correcting genotyping errors in microsatellite data. Molecular Ecology Notes 4(3): 535–538.

Van Zyll de Jong, C. G. 1975. The distribution and abundance of the wolverine (*Gulo gulo*) in Canada. Canadian Field-Naturalist 89(4): 431–437.

Vangen, K. M., J. Persson, A. Landa, R. Andersen, and P. Segerström. 2001. Characteristics of dispersal in wolverines. Canadian Journal of Zoology 79(9): 1641–1649.

Walker, C. W., C. Vilà, A. Landa, M. Lindén, and H. Ellegren. 2001. Genetic variation and population structure in Scandinavian wolverine (*Gulo gulo*) populations. Molecular Ecology 10(1): 53–63.

Weaver, J. L., P. C. Paquet, and L. F. Ruggiero. 1996. Resilience and conservation of large carnivores in the Rocky Mountains. Conservation Biology 10(4): 964–976.

Weir, B.S., and C. C. Cockerham. 1984. Estimating F-statistics for the analysis of population structure. Evolution 38(6): 1358–1370.

White, K. S., N. L. Barten, S. Crouse, and J. Crouse. 2014. Benefits of migration in relation to nutritional condition and predation risk in a partially migratory moose population. Ecology 95(1): 225–237.

Whitman, J. S., W. B. Ballard, and C. L. Gardner. 1986. Home range and habitat use by wolverines in southcentral Alaska. The Journal of Wildlife Management 50(3): 460–463.

Wilson, G. M., R. A. Van Den Bussche, P. K. Kennedy, A. Gunn, and K. Poole. 2000. Genetic variability of wolverines (*Gulo gulo*) from the Northwest Territories, Canada: conservation implications. Journal of Mammalogy 81(1): 186–196.

Wright, S. 1951. The genetical structure of populations. Annals of Eugenics 15: 323–354.

Yates, R. E., J. P. Copeland, and J. R. Squires. 2013. Wolverine food habitats and foraging strategies in Glacier National Park, Montana. Intermountain Journal of Sciences 19(1–4): 59pp.

Young, J. K., B. Hudgens, and D. K. Garcelon. 2012. Estimates of energy and prey requirements of wolverines. Northwest Science 86(3): 221–229.

Zigouris, J., F. N. Dawson, J. Bowman, R. M. Gillett, J. A. Schaefer, C. J. Kyle. 2012. Genetic isolation of wolverine (*Gulo gulo*) populations at the eastern periphery of their North American distribution. Conservation Genetics 13(6): 1543–1559.

Zink, R. M., and G. F. Barrowclough. 2008. Mitochondrial DNA under siege in avian phylogeography. Molecular Ecology 17(9): 2107–2121.

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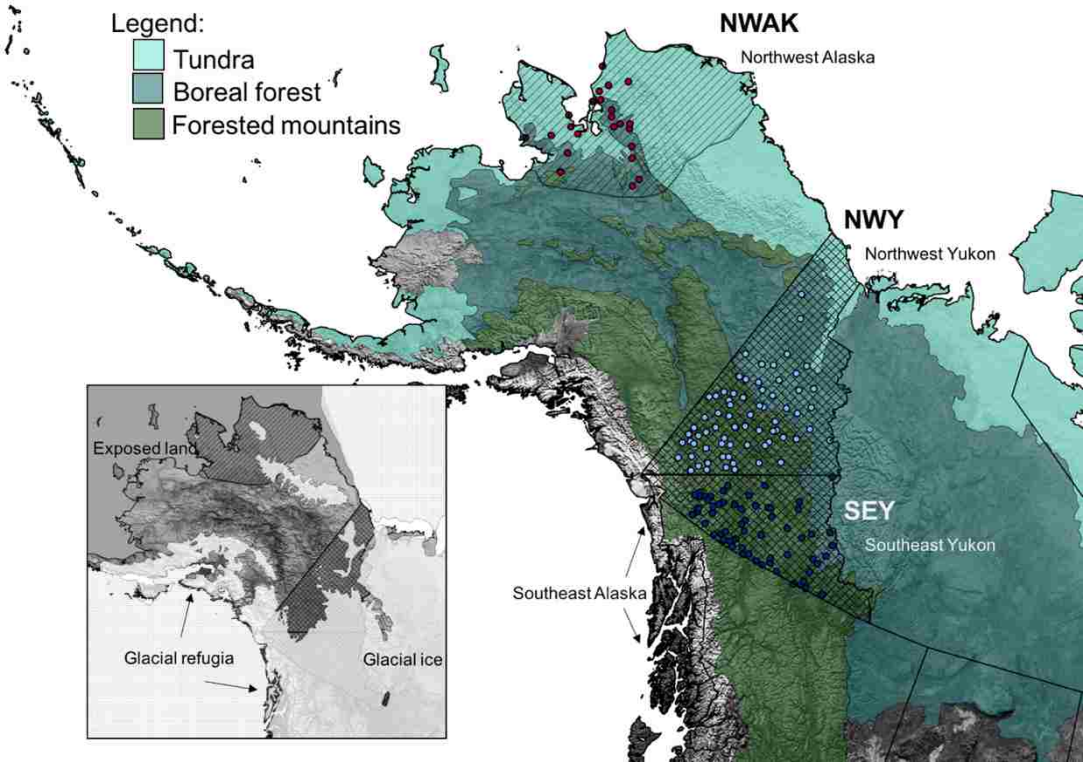


Figure 1: Sampling for Northwest Alaska (NWAK) and Northwest and Southeast Yukon Territory (NWY, SEY). Intensive sampling in Yukon led to its division in analysis, as well as previous work in Chapter 1 establishing different genetic signatures arising from those regions. NWAK 117, NWY 114, SEY 129. Ecological zones of interest are mapped with Alaska groups in tundra habitat and Yukon groups in boreal forest flatlands and mountainous forest areas. Inset map shows glacial history of the region with dark gray being exposed land during the LGM, white being ice, and hatched being study areas.

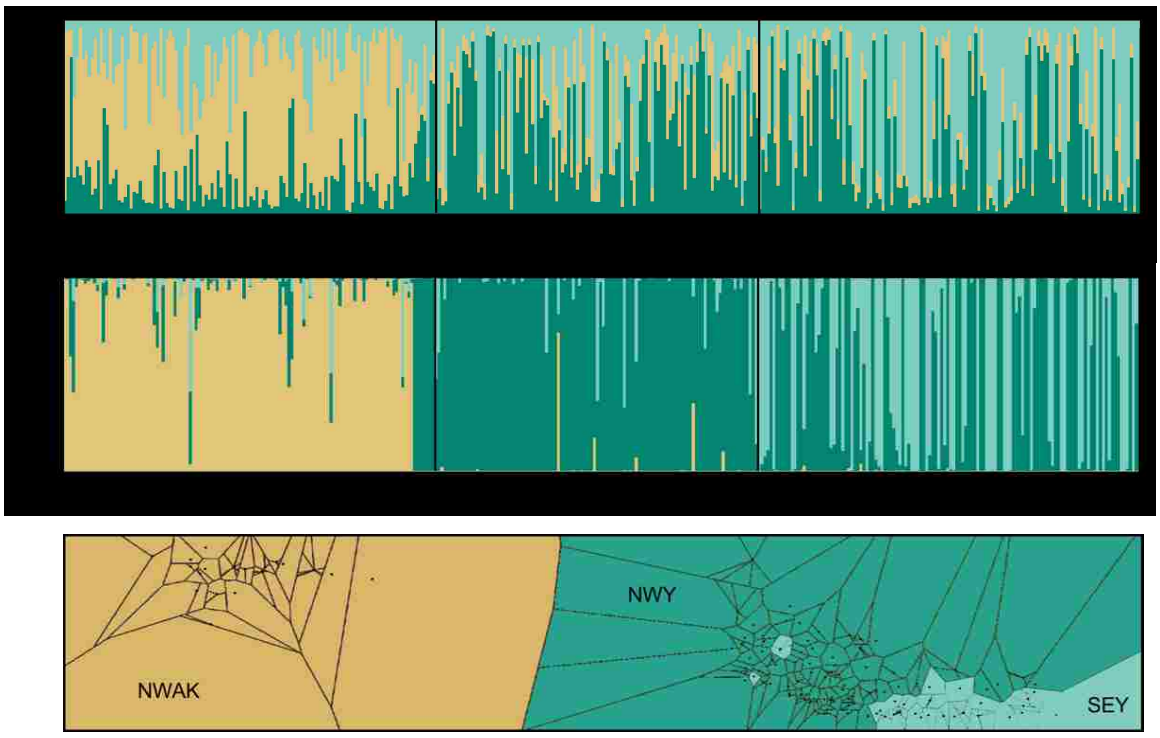


Figure 2: Individual-based cluster results: (a) STRUCTURE plot of individual membership coefficient (*y*-axis) for each sampled region without locality priors. (b) TESS plot of individual assignment probabilities with coordinate data as priors. (c) TESS membership of individuals with interaction parameter $\psi = 0.6$.

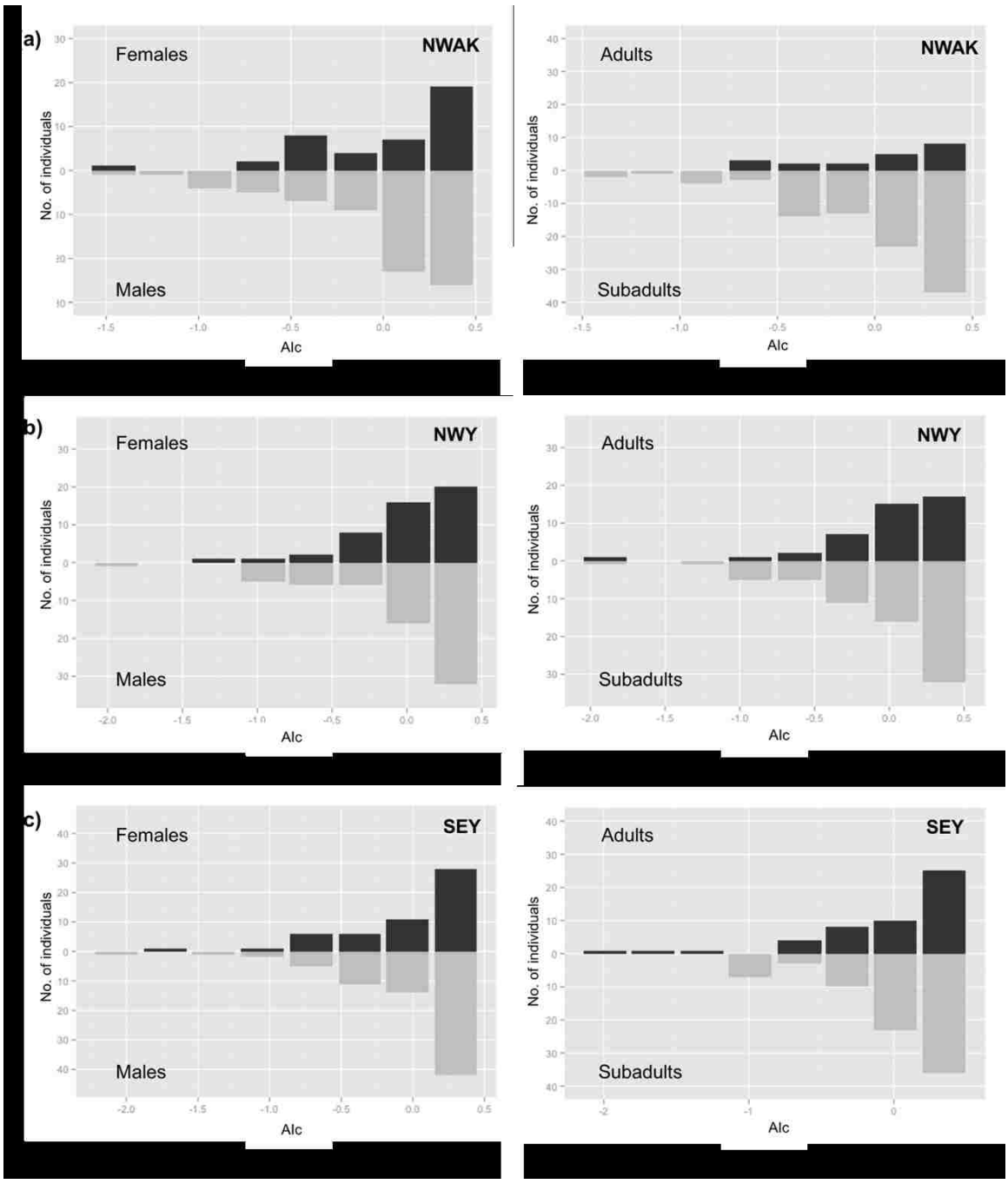


Figure 3: Frequency distribution of corrected assignment index (AI_C) with sex comparisons on the left (male below and female above the *x*-axis) and age comparisons on the right (subadults below and adults above the *x*-axis). (a) NWAK, (b) NWY, (c) SEY.

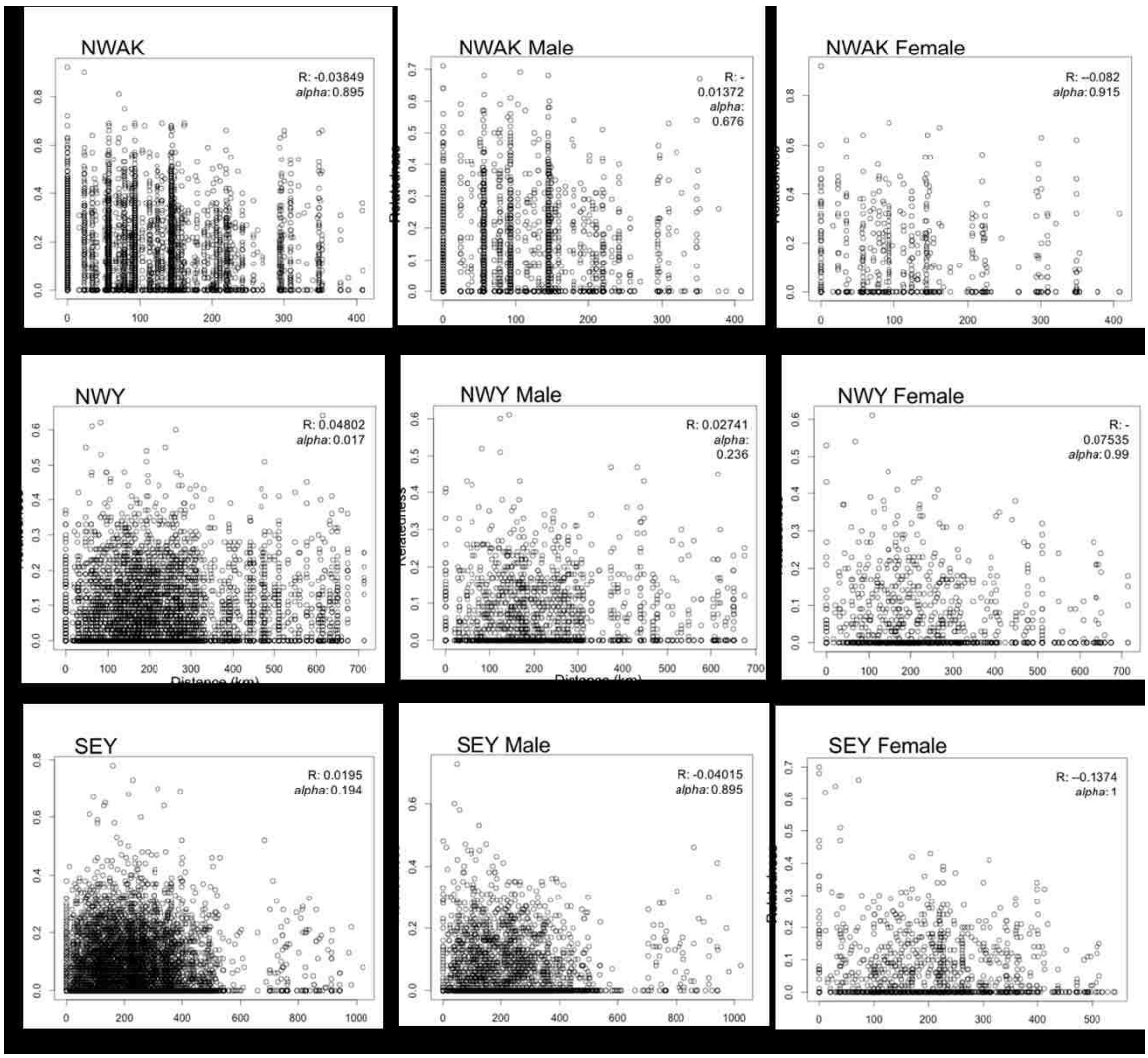


Figure 4: Relationships between pairwise coefficients of genetic relatedness and geographic distances (km) by population and for males and females within a population.

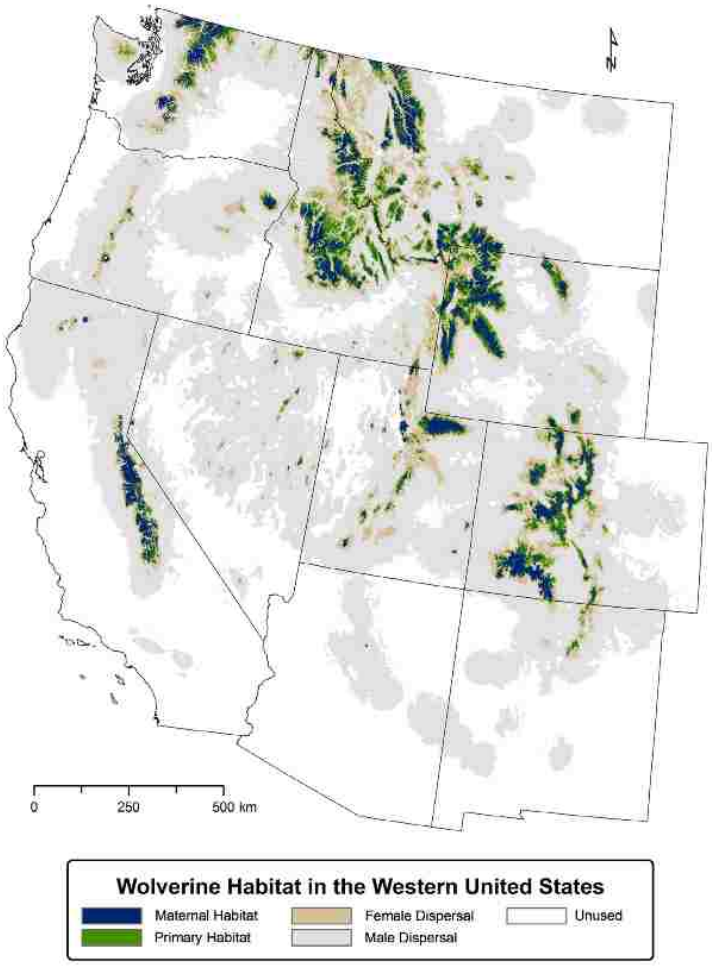
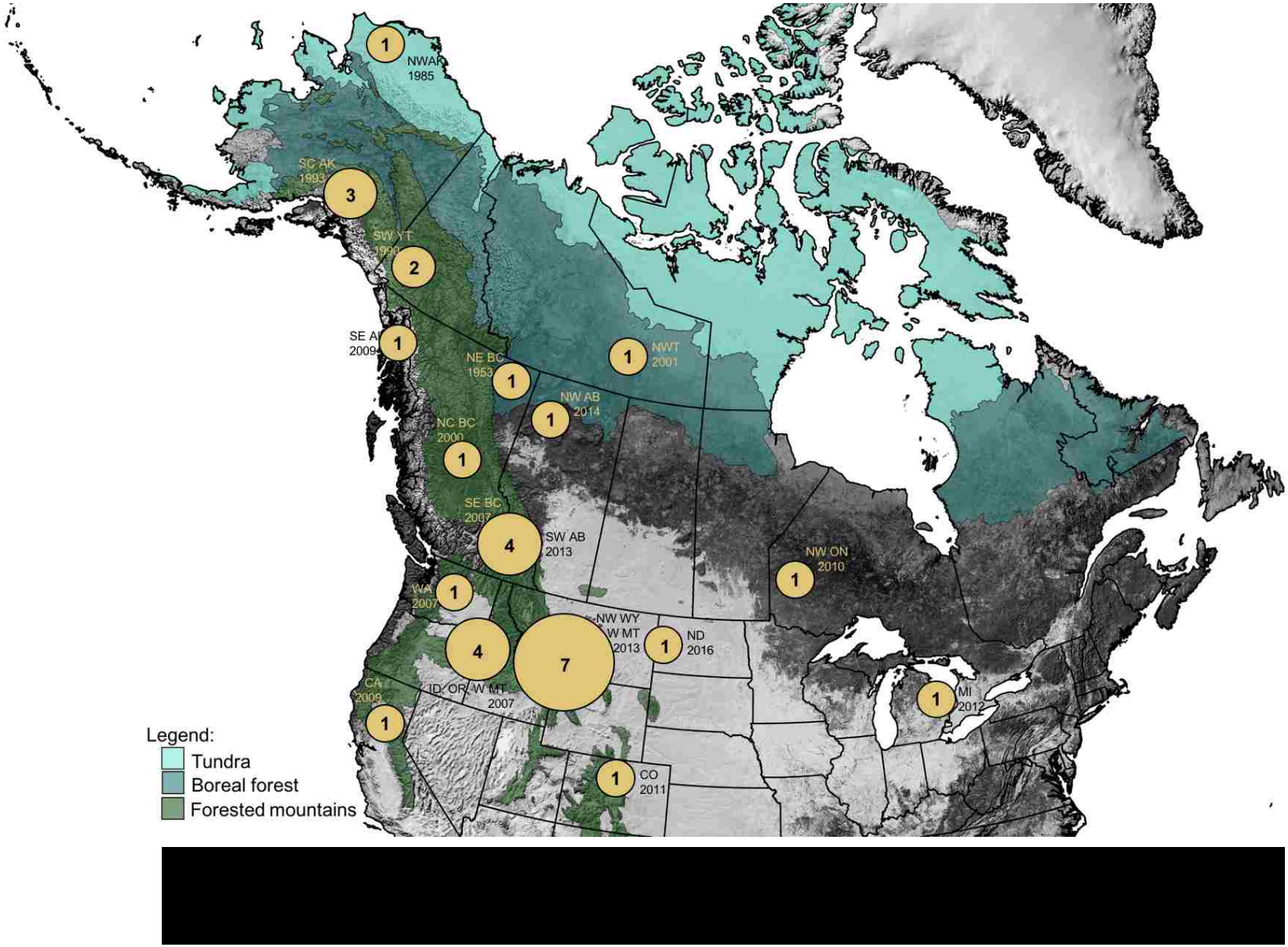


Figure 5: Areas of the conterminous United States predicted to be habitat for denning (maternal habitat), possible wolverine habitat in general (primary habitat), and areas of male and female dispersal based on resource selection function modeling (combining latitude-adjusted elevation, terrain ruggedness index, April 1 snow depth, road density, interpolated human density, distance to high-elevation talus, distance to tree cover, distance to April 1 snow >2.5 cm; habitat scores of ≥0.967) based on wolverine telemetry locations in the Greater Yellowstone Ecosystem of Montana, Idaho, and Wyoming. Figure taken from Inman *et al.* 2013.


1
NWAR
1985
SC AK
1993
3
SW YT
2
SE AK
2009
1
NE BC
1953
1
NWT
2001
1
NW AB
2014
1
NC BC
2000
1
SE BC
2007
4
SW AB
2013
NW ON
2010
1
WA
2007
1
NW WY
W MT
2013
1
ND
2016
4
7
CA
2009
1
ID, OR, W MT
2007
1
MI
2012
1
CO
2011
Legend:
Tundra
Boreal forest
Forested mountains

[redacted]

List of Tables

Table 1: Sample size (*n*) by total individuals in a region and by subadult and adult divisions, observed heterozygosity (H_O), expected heterozygosity (H_E), allelic (A_R), and inbreeding coefficent (F_{IS}).

Male	76	-0.5119	0.1834	Subadults	97	-0.4935	0.1905
Male	66	-0.4976	0.2431	Subadults	71	-0.5349	0.2404
Male	76	-0.4651	0.2298	Subadults	79	-0.4386	0.1631

Table 2: Average log-transformed assignment index (*mAIc*) and variance values (*vAIc*) for the three wolverine populations by age class and by sex.

MM	0.069	0.00	14.88	MM	0.086	1.31	12.90
MM	0.049	0.00	3.40	MM	0.048	0.00	4.40
MM	0.049	0.27	3.57	MM	0.051	0.18	3.89

Table 3: Mean relatedness (*m*Relatedness) between same-sex pairs of adult and subadult wolverines in the three sampling locations. Percent first order (% 1˚) and second order (% 2˚) relatives within each subcategory are listed.

NWAK	0.600	0.222	0.178
NWY	0.115	0.546	0.339
SEY	0.048	0.379	0.573

Table 4: Migration rates between the three sampling locations. Estimates are based on posterior means. Probability that the pairwise estimate is equal to or greater than the corresponding pairwise estimate $^{a}p < 0.05$, $^{b}p < 0.01$, $^{c}p < 0.0001$. Standard deviation listed below migration rates, in brackets. 'Into' population listed in column, 'From' population listed in row.

Supplementary Material

S1 Table: Specimens used by museum catalog number. Northwest Alaska (NWAK, *N* = 129) from Dalerum et al. 2007; Northwest Yukon (NWY, *N* = 114) and Southeast Yukon (SEY, *N* = 129) genotyped in current study.

S2 Table: Number of alleles per locus, repeat motif, minimum and maximum length of calls, multiplex assignment, forward and reverse primer, citation, and GenBank or EMBL (first locus only) accession number. Bolded loci were adjusted slightly from the original; 1 denoting the forward primer was optimized and 2 denoting the reverse. For primer Gg454F, we added a C on the end of the published primer sequence to provide a GC clamp (since that was the next nucleotide based on the sequences archived in GenBank). For primer Gg452R, we added a G (the next nucleotide) on the end of the published primer because the published primer sequence was only 17 bp, which is shorter than generally recommended. Asterisks indicate loci screened across Alaskan samples (10 out of 20); all 20 loci were run on Yukon samples.

S3 Figure: GenBank accession sequences for ZFX data for mustelid molecular sexing on standards UAM31719 and MSB157754.

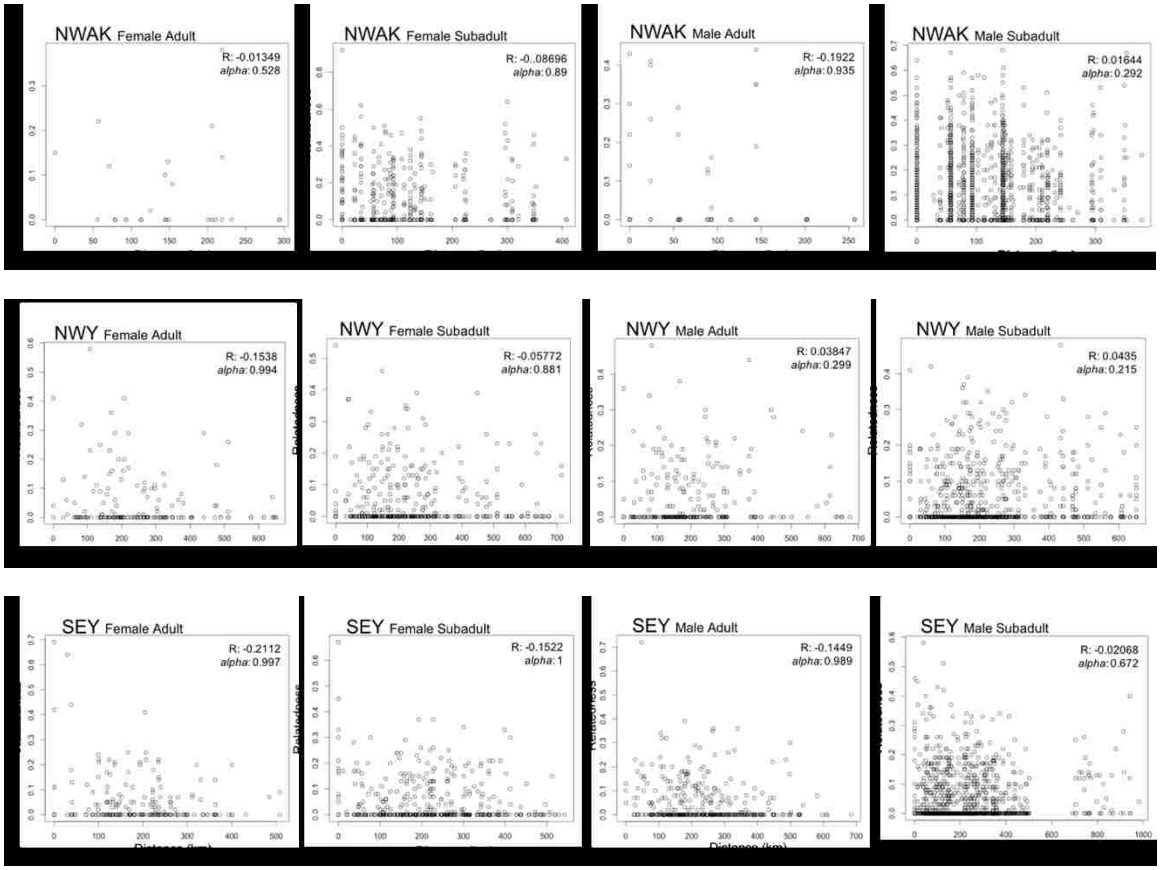
>1 Gulo gulo isolate AF147943 ZFX gene for X-linked zinc finger protein, final intron, partial sequence [organism=Gulo gulo] [mol_type=genomic DNA] [tissue_type=muscle] [country= USA:Alaska:Little Oshetna River][specimen_voucher=MSB:MAMM:157754]
CGTTATTAGGCAAGCATTCCTACATTAAGCTATCTGCTACGTAACATTCCTTCTACCGTTTTTTCAATATA
AGAGGCAGAGCAACCCTGTCATAAAGAGAACCCTGGTCTGAAAACTTCATTCAGTCTGGTGGTACCAACC
TCAGGCCCTCCAGTTTAAAAAAAATCAATCAATAAATACATAACTTCTGCCAACTAGTGATCAAAGCTCT
ATCAATTTAGAAATTGGCAGAAAATTCTTCATAAAGTTTTGTAATCACAATTCCTGCTTTGGTTATTCCGA
AAAACCTAATTTTGTGTCACTTGACAGTAAAGCTTAAATCTATCTACGAAAATTTTAATATACAAATCACC
ATCCACGAATAGACAGGAATAGGTCGGGTAAACTGCGGTACTTGTCCTACTGCTCTGTAAATTATGCCTG
CCTTAAGGTAACATGATCCAATTCTTTGCGGTAATTTAGGTTTTAAAACAGTTAATGCTTGTAACATGTCT
GAGGGACTTCTTTTCATTTCCACATAAAAACCACCCGAGTTTTGAATTTTGTTCCCCATCCACGTTTGATC
CATTCCCTGTTTCTTTTTATCCCAGGCAATCATTCATGAATATCACTGAATTCTTAAAATTATATTTTCAAA
TTCCACACACAAAAGCTACATGTGGTCTAGCAGCTAAAATGCCATCACAACACCTCTGTGGGTACATACT
AGAGTTTCATCTGAGAGCTCGCCAAGCACGCTGCGCTGTGGGACTCGTGTGCCCTCACCTGTTTGGTACT
GTCTGGAATCAGGTCT
>2 Gulo gulo isolate UAM31719 AF11801 ZFX gene for X-linked zinc finger protein, final intron, partial sequence [organism=Gulo gulo] [mol_type=genomic DNA] [tissue_type=muscle] [country= USA:Alaska:Healy Lake] [specimen_voucher=UAM:MAMM:31719]
CGTTATTAGGCAAGCATTCCTACATTAAGCTATCTGCTACGTAACATTCCTTCTACCGTTTTTTCAATATA
AGAGGCAGAGCAACCCTGTCATAAAGAGAACCCTGGTCTGAAAACTTCATTCAGTCTGGTGGTACCAACC
TCAGGCCCTCCAGTTTAAAAAAAATCAATCAATAAATACATAACTTCTGCCAACTAGTGATCAAAGCTCT
ATCAATTTAGAAATTGGCAGAAAATTCTTCATAAAGTTTTGTAATCACAATTCCTGCTTTGGTTATTCCGA
AAAACCTAATTTTGTGTCACTTGACAGTACAGCTTAAATCTATCTACGAAAATTTTAATATACAAATCACC
ATCCACGAATAGACAGGAATAGGTCGGGTAAACTGCGGTACTTGTCCTACTGCTCTGTAAATTATGCCTG
CCTTAAGGTAACATGATCCAATTCTTTGCGTTAATTTAGGTTTTAAAACAGTTAATGCTTGTAACATGTCT
GAGGGACTTCTTTTCATTTCCACATAAAAACCACCCGAGTTTTGAATTTTGTTCCCCATCCACGTTTGATC
CATTCCCTGTTTCTTTTTATCCCAGGCAATCATTCATGAATATCACTGAATTCTTAAAATTATATTTTCAAA
TTCCACACACAAAAGCTACATGTGGTCTAGCAGCTAAAATGCCATCACAACACCTCTGTGGGTACATACT
AGAGTTTCATCTGAGAGCTCGCCAAGCACGCTGCGCTGTGGGACTCGTGTGCCCTCACCTGTTTGGTACT
GTCTGGAATCAGGTCT

S4 Table: Observed heterozygosity (H_O), expected heterozygosity (H_E), number of alleles per locus (# alleles), and deviation from random mating (F_{IS}) over microsatellite loci by total sampling region, as well as males and females separately. Significant values are in boldface type. Sample size (N) correspond to the number of individuals genotyped in each category.

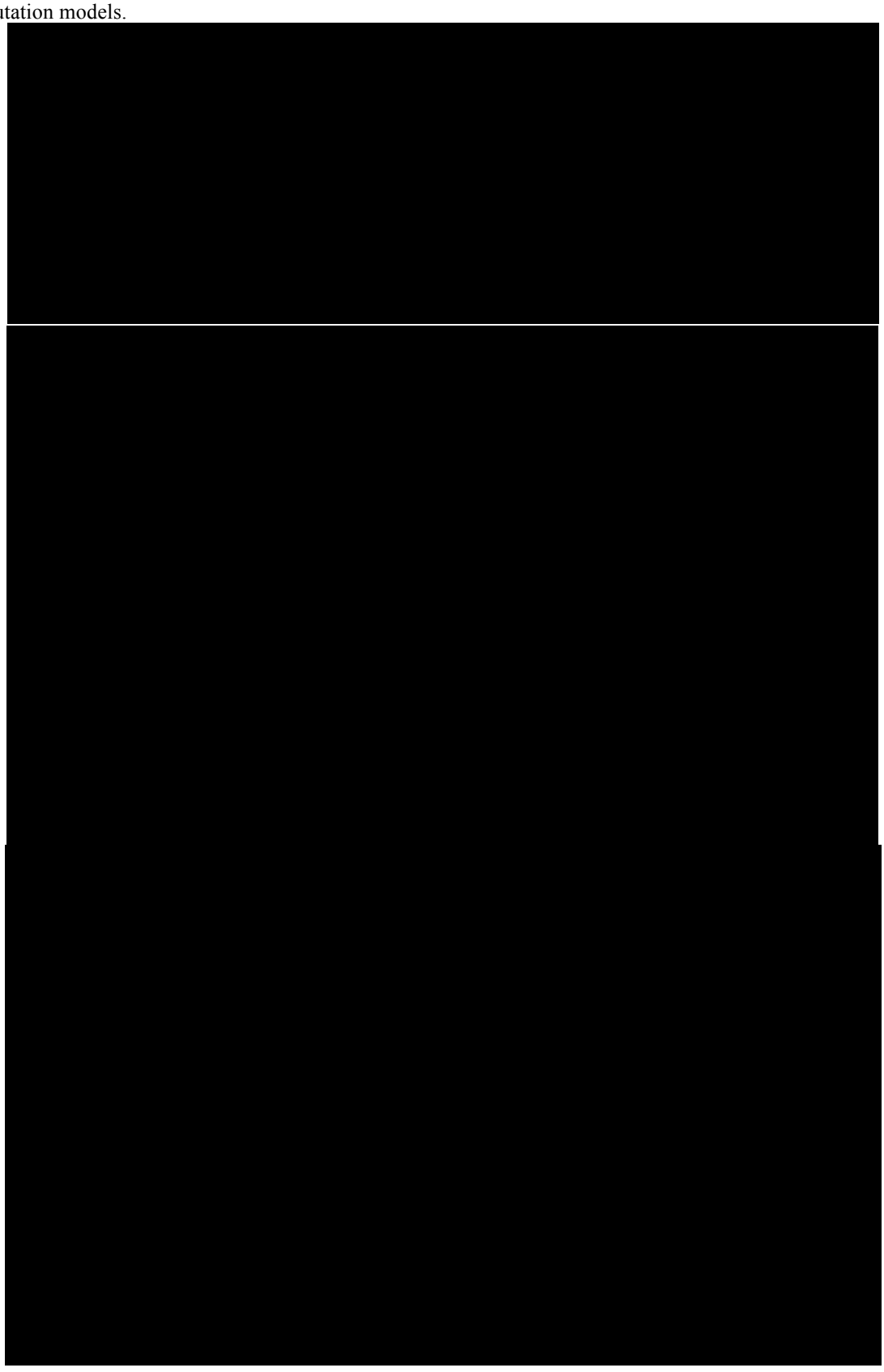
S5 Table: Pairwise comparisons of F_{ST} values. Bold values indicate statistical significance after 1,000 permutations. (a) Population comparisons by all individuals and females and males separately. (b) All population comparison after sorting by intrapopulation sex (NWAK females = NWAK F, etc.). Darker shading represents greater difference.

0.02165	**0.02294**		
0.01595	**0.01436**		
0.0296	**0.03199**	**0.00694**	**0.00874**
0.0262	**0.03064**	**0.0057**	**0.00889**

S6 Figure: Mantel test results (correlation of pairwise genetic relatedness and pairwise geographic distance) broken down into by-age and by-sex categories for each of three populations.



S7 Table: BOTTLENECK results by-locus for each population under the I.A.M, T.P.M, and S.M.M. mutation models.



S8 Table: Wolverine home ranges in km^2 from seventeen telemetry and tracking studies averaged within state or province (NWAK, Northwestern Alaska; AK, Central and South Alaska; YT, Yukon Territory; BC, British Columbia; AB, Alberta; ON, Ontario; WA, Washington; MT, Montana; ID, Idaho; WY, Wyoming). Shading refers to ecoregion and latitude delineation with shades of gray indicating populations above the 60^{th} parallel. Female-to-male ratio of home range size have been averaged in the legend values.

	Adult males (M)	Subadult males (m)	Adult females (F)	Females w/young (FY)	Subadult females (f)	F/M ratio
NWAK	666		126	73		0.19
AK	494		191	105		0.39
YT	238	526	250	108		1.05
BC	1179	2365	382		919	0.32
AB	1419		282			0.20
ON	2145	4340	301-750	262	428	0.24
WA	1890		1709			0.90*
MT	472		264	100		0.56
ID	1582	435	361			0.23
WY	853	2689	375	125	884	0.44

60th parallel

Legend:

- tundra; ave. ratio 0.19
- high latitude forest; ave. ratio 0.72
- low latitude forest; ave. ratio 0.33*

*WA outlier removed, small sample size within study

S9 Table: Previous studies on individual-based wolverine population dynamics that track individuals using various methods (telemetry, snow tracking, camera traps, genetic tests) to determine aspects of their demography, especially home range size and natal dispersal distance.

Northwest Alaska	NW Alaska	666M	488-917M	4		telemetry	<1985	2400	48-139	Magoun 1985
Upper Susitna Basin	SC Alaska	452M / 179m (0yrs)	366-637M	4, 1		telemetry	1980-1982	7700		Gardner 1985
Upper Susitna Basin	SC Alaska	535M		4		telemetry	1980-1983	7700	209	Whitman et al. 1986[1]
Berners Bay	SE Alaska			6		telemetry	2008-2009	2200		Lewis et al. 2009
Kluane Game Sanctuary	SW Yukon	238M / 526m		1, 1		telemetry	<1987	1590	177	Banci 1987
Kluane Game Sanctuary	SW Yukon	238M / 526m		3, 2		telemetry	1983-1984	1590	93-177	Banci and Harestad 1990
Williston Reservoir	NC Brit Col	1756M / 3119m	403-4953M / 333-7352m	10, 8		telemetry	1996-1999	8900		Lofroth 2000
Kootenay Region	SE Brit Col	601M /1611m	347-875M / 874-2884m	21, 4		telemetry	1994-1997	7000		Krebs and Lewis 2000
Omineca Mountains	SE Brit Col			11M, 6m		telemetry, camera traps	1995-2003	8900	154	Krebs et al. 2007, Lofroth and
Columbia Mountains	SE Brit Col			9M, 16m				7000	172	
Rainbow Lake	NW Alberta		400-1200M	13		telemetry	2013-2014	1624		Scrafford and Boyce 2014; Scafford et al. 2017
Red Lake	NW Ontario	2145M / 4340m	1783-2506M / 4340m	2, 1		telemetry	2003-2004	6600		Dawson et al. 2010
Flathead National Forest	NW Montana	422M		9, 2		telemetry, snow-tracking	1972-1977	1300	65	Hornocker and Hash 1981
Gallatin National Forest	S Montana			5		telemetry, snow-tracking	1999-2009			Karabensh 2015 MS
Pioneer National Forest	W Montana			9		telemetry	2000-2005	7460		Squires et al. 2007
Clearwater National Forest	W Montana			11				2150		
Glacier National Park	W Montana			2				>5700		
Glacier National Park	W Montana	521M	312-749	>4[9]	>200m, >88f	telemetry	2003-2008			Copeland and Yates 2008
Idaho	Idaho	435m		1		snowtraking, camera traps				Copeland 1993[2]
Idaho	C Idaho	1582M		8	>185m	telemetry	1992-1996	9315		Copeland 1996 MS,
Greater Yellowstone Ecosystem	NW Wyoming	797M / 2689m		5, 6	170m	telemetry, snowtracking	2001-2010	108000	151	Inman et al. 2003, 2012, 2013
Greater Yellowstone Ecosystem	NW Wyoming	908	446-1268	2, 1		telemetry	2005-2009	13000		Murphy et al. 2011
Rocky Mountains National Park	Colorado			1M	disperser (Yellowstone, WY)	camera trap, genetic study	2010			Danzinger 2011
Truckee area	California			1M	disperser (Sawtooth Mts, ID)	camera trap, genetic study	2008	150		Moriarty et al. 2009
Billings, North Dakota	W North Dakota			1M	disperser (MT or WY)	shot	2016			Inman, comments 2016

Males are in grayed lines. Home range measurements are broken into adult males (M), subadult males (m), adult females (F), adult females with young (FY), and subadult females (f).
Counts (N) correspond to these delineations.
[1] *Estimated using the realtionship between time of monitoring and home range size.*
[2] *90% minimum polygon home range is 369 km2.*
[3] *If two long-distance movements are excluded, home range is 47 km².*
[4] *Estimated using the relationship between time of monitoring and home range size.*
[5] *If 1 long-distance movement is excluded for each female, home ranges are 153 and 157 km², with a mean of 155km².*
[6] *90% minimum polygon home ranges are 82 and 447 km2; core harmonic mean ranges are 79 and 306 km².*
[7] *Unpublished data, U.S. National Park Service*
[8] *Unpublished data, Renewable Resources, Fish and Wildlife Branch*
[9] 27 total wolverines tracked in the study, though only these numbers directly reported for homeranges

CONCLUSION

Through an understanding of the wolverine's niche limitations and motivations for movement, the ancient, historical, and contemporary changes in climate and habitat of North America can be tracked. My master's thesis used genetic tools to quantify these movements and improve our understanding of climate impacts on wolverines in the past in glacial refugia, understand their populations in the dynamic topography of Alaska and western Canada, and their needs in increasing habitat fragmentation especially at lower latitudes. These findings help us track further habitat loss as well as successes in mediation fragmentation and habitat destruction.

In Chapter 1, a chief finding was the presence of a unique population on the Kenai Peninsula. A private mitochondrial haplotype made up a quarter of the sampled individuals there, and very limited allelic diversity shows this peninsula to have limited gene flow and at least historical isolation (not isolated during glacial oscillations). Previous work on large carnivores with slow life history (i.e., long generation times) has shown populations here to be potentially imperiled due to the significant effect of harvest or competitors on their low-density, low-diversity populations.

Another important finding was the disjunction between Alaska and the rest of mainland Canada showing either a lingering phylogeographic break (as formerly divided lineages come back into contact) (Colella et al. in prep), or is a result of population structuring based on a difference in habitat (Talbot and Shields 1996). This question, whether habitat dictates wolverine gene flow and dispersal, was further examined in Chapter 2.

In Chapter 2, population-level dynamics were analyzed, again with nuclear markers in the form of microsatellites. Through this, we found no difference between males and females and virtual panmixia among northwestern Alaska tundra habitat and the Yukon Territory boreal forest populations. This did not support the expectation of sex-biased dispersal established by previous studies chiefly conducted in lower latitudes.

Though we have obvious need for more data, we now hypothesize that the lack of sex-bias in high latitudes may be due to continuous habitat and ease of dispersal and gene flow for both sexes. Meanwhile lower latitude habitats are increasingly fragmented by natural habitat shift as well as human-influenced habitat destruction and climate change (Peacock 2011). This must be studied further with additional genetics studies in fragmented habitats, as well as telemetry studies in hypothesized continuous ones.

This work informs our understanding of wolverine dispersal dynamics at high latitudes and the ancient movement of wolverines in the complex regions of Alaska and western Canada. Also important, this project has been a collaborative work between academic institutions, government bodies in the United States and Canada, trappers, managers, and museum archival and data repository resources. These types of collaborations make otherwise unattainable sampling and unanswerable investigations possible. As a result, wolverines (and many other species) have an improved understanding of life history and lead to a better knowledge of climate history as well as insight into the climate future and the efficacy of conservation efforts.

Literature Cited

Colella JP, T. Lan, C. Lindqvist, S. L. Talbot, J. A. Cook. In prep. Mitogenomic phylogenetics of Mustela erminea (Family Mustelidae), investigating the impact of metabolic selection on phylogenetics. *submitted*.

Peacock, S. 2011. Projected 21st century climate change for wolverine habitats within the contiguous United States. Environmental Research Letters 6(014007): 1-9.

Talbot, S. L., and G. F. Shields. 1996. Phylogeography of brown bears (*Ursus arctos*) of Alaska and paraphyly within the Ursidae. Molecular phylogenetics and evolution 5(3): 477-494.