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Sea ice occurrence predicts genetic isolation in the Arctic fox

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Abstract

Unlike Oceanic islands, the islands of the Arctic Sea are not completely isolated from migration by terrestrial vertebrates. The pack ice connects many Arctic Sea islands to the mainland during winter months. The Arctic fox (*Alopex lagopus*), which has a circumpolar distribution, populates numerous islands in the Arctic Sea. In this study, we used genetic data from 20 different populations, spanning the entire distribution of the Arctic fox, to identify barriers to dispersal. Specifically, we considered geographical distance, occurrence of sea ice, winter temperature, ecotype, and the presence of red fox and polar bear as non-exclusive factors that influence the dispersal behaviour of individuals. Using distance-based redundancy analysis and the BIOENV procedure, we showed that occurrence of sea ice is the key predictor and explained 40–60% of the genetic distance among populations. In addition, our analysis identified the Commander and Pribilof Islands Arctic populations as genetically unique suggesting they deserve special attention from a conservation perspective.

Keywords: *Alopex lagopus*, Commander Islands, dispersal, microsatellites, mtDNA, Pribilof Islands

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Introduction

Many Arctic Sea islands are populated by the Arctic fox (*Alopex lagopus*), a small cold-adapted canid closely related to the North American kit and swift fox (*Vulpes macrotis* and *Vulpes velox*, respectively; Mercure *et al*. 1993; Wayne *et al*. 1997). These Arctic islands include some of the largest on the planet (e.g. Greenland and Baffin Island), but the majority are small in areal extent. Unlike Oceanic islands, the islands within the Arctic Circle are not completely isolated from migration by terrestrial vertebrates. Because of the complete freeze of the Arctic Ocean during the winter months, pack ice connects many islands with each other and the mainland (Fig. 1). Arctic foxes are exceptionally adapted for the extreme cold conditions of the Arctic region (Prestrud 1991; Audet *et al*. 2002). Individuals are known to travel great distances during winter (> 1000 km; Wrigley & Hatch 1976; Angerbjörn *et al*. 2004) and traverse extensive pack ice fields (Andriashek *et al*. 1985; Roth 2002). Extended movements occur in late autumn–early winter or during spring as a result of food shortage (Pulliainen 1965; Eberhardt *et al*. 1983; Audet *et al*. 2002). Arctic foxes scavenge kill remains of polar bears (*Ursus maritimus*) as well as utilize localized abundances of lemmings and sea birds (Audet *et al*. 2002). They follow polar bears on the pack ice for great distances (Roth 2002, 2003). In spring, when the pack ice fragments, Arctic foxes sometimes are entrapped on drifting icebergs and are carried to distant landmasses (e.g. Fay & Rausch 1992). The sea around many islands in the Arctic does not freeze during winter (e.g. Iceland and the Aleutians); however, pelagic ice floes drift and occasionally reach the shores of these isolated islands and potentially transport foxes. In fact, Charles Darwin (Darwin 1909; p. 174) suggested that the Falkland Island wolf (*Dusicyon australis*) may have been carried there by ice floes on which wolves could subsist on seals.

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Fig. 1 Arctic fox sampling localities. The dotted lines designate the average annual peak in pack ice extent. On this map, the Commander Islands include Bering and Mednyi Islands, and the Pribilof Islands include St. Paul and St. George Islands.

However, the role of sea ice in the recent isolation of Arctic fox populations is not well understood because the response of foxes to temporal and spatial variation in the extent of sea ice is uncertain. During the last glaciation (~21 000–10 000 years BP), all the islands in the Arctic Sea were connected by sea ice and some were completely covered by a thick ice layer (Grosswald 1998; Schäfer-Neth & Paul 2003). Records from fossil and archaeological excavations showed an extensive reduction in the range size of Arctic foxes corresponding to the contraction of ice sheets. For example, in the Late Glacial (11 000–15 000 years BP), the Arctic fox occurred across southern Europe (e.g. south France, the Ukraine; Sommer & Benecke 2005) and throughout large areas of the Palearctic region (Kahlke 1999). At present, this species is confined to a circumpolar distribution inhabiting the northernmost regions of Eurasia and North America.

The only recent parallel of these events is the little ice age (1550–1850; Crowley 2000; Grove 2003; Mann & Jones 2003; Moberg *et al*. 2005) when unlike today, glaciers expanded in the Alps, Scandinavia, Alaska, and Kamchatka Peninsula and the seas around Iceland froze in winter. The Arctic pack ice extended so far south that Inuits landed their kayaks in Scotland, and polar bears reached the Orkney Islands (Fagan 2001). Presently, the waters around some islands in the Arctic are always frozen during winter whereas the formation of sea ice near others is an infrequent event (Parkinson *et al*. 1999).

The occurrence of sea ice is also highly variable over the short-term. For example, from 1987 to 1993, the sea ice reached Iceland shores only during April 1989. In contrast, during 1968–1969, the extent of sea ice was continuous from Iceland to Greenland, and during May–June 1888, the sea ice was distributed all along the northern and eastern Icelandic coasts, and even extended to the southern shores (fig. 2.9 in Grove 2003). Records from the 1740s indicated that sea ice was more frequent and severe then, and in June–July 1759, it surrounded all eastern and southern parts of Iceland including Reykjanes for 2–3 weeks (Ogilvie & Jonsson 2001; Grove 2003). Iceland is an excellent example for illustrating our main hypothesis that sea ice, in spite of being unpredictable, provides a persistent vehicle at different temporal scales for fox migration to isolated Arctic islands. Although over a period of decades, Iceland may be isolated, eventually it reconnects to the main Arctic ice sheet long enough to allow migration.

Dalén *et al*. (2005) conducted an extensive survey of mtDNA variation in Arctic fox populations from the Arctic Circle. They found no correlation between genetic distance and geographical distance (e.g. $r = -0.19$,

$P = 0.90$). Considering that nearly all the populations they studied were connected by land or pack ice during winter, and the extensive dispersal of Arctic foxes over the ice, it is reasonable to conclude that at least a few migrants per generation reach distant populations. However, grouping populations according to diet (inland lemming-based vs. coastal marine-based populations) accounted for 25% of the genetic variation. Thus, as been found in other canids (Carmichael *et al*. 2001; Geffen *et al*. 2004; Sacks *et al*. 2004), ecology rather than distance appears to restrict gene flow between many populations. However, Dalén *et al*. (2005) did not explicitly consider the importance of ice as a dispersal agent and relied only on a single maternally inherited locus (mtDNA) for inference about levels of genetic exchange.

In this study, we analysed mtDNA control region sequences and 11 microsatellite loci in High Arctic island populations of the Arctic fox. Connectivity with the mainland varies among these islands as some are linked by pack ice for several months every year while others rarely have pack ice. We hypothesized that floating ice is a critical transport agent for Arctic foxes and allows them to reach remote areas. We explicitly tested this idea by incorporating the probability of encounting floating ice at each island into the analysis of genetic data. Furthermore, we also tested four alternative nonexclusive hypotheses for differentiation reflecting environmental variables such as winter temperature or biological factors such as competition or prey type.

Methods

Arctic foxes have been introduced to many of the Aleutian Islands for the fur trade (1750–1925). These introductions heavy impacted native sea birds colonies, and starting in 1949 programmes for eradication of Arctic foxes were initiated (reviewed in Long 2003). In this study, we selected island populations that were not subjected to introduction. Svalbard, Iceland, Greenland and the islands along the Canadian coast (Bathurst Island and Banks Island) harbour only native populations. The populations of the Commander Islands (Mednyi and Bering Islands, Russia) and the Pribilof Islands (St. George and St. Paul Islands, USA) are considered native and isolated from other Arctic fox populations for an extended period of time (White 1992; Goltsman *et al*. 2005). Some have been designated as separate subspecies (*Alopex lagopus semenovi*, *A. l. beringensis* and *A. l. pribilofensis* for Mednyi Island, Bering Island and Pribilof Islands, respectively; Audet *et al*. 2002; Angerbjörn *et al*. 2004).

We sequenced 293 bp of the mtDNA control region (Table 1) of 107 individuals from nine populations and combined this data with that published by Dalén *et al*. (2005). The combined mtDNA data set spanned 20 populations. Briefly, whole genomic DNA was extracted from blood or muscle tissue using QIAGEN's DNeasy tissue kit (QIAGEN) or standard Proteinase K and phenol–chloroform extraction protocol. Universal primers Thr-L15910 (5′GAATTCCCCGGTCTTGTAAA CC-3′) and

Table 1 Sample size, number of mtDNA haplotypes, gene diversity, and Tajima's *D* test of selective neutrality for 20 Arctic fox populations. The geographical position for each site is indicated in Fig. 1

Site	Sample size	Number of haplotypes	Gene diversity (± SD)	Tajima's *D* (*P* value)
Iceland	23	4	0.60 (0.10)	0.57 (0.739)
East Greenland	11	5	0.76 (0.11)	–0.30 (0.416)
South Greenland	10	5	0.84 (0.08)	0.80 (0.811)
West Greenland	9	4	0.58 (0.18)	0.58 (0.749)
North Greenland	8	2	0.25 (0.18)	–1.05 (0.073)
Churchill	20	6	0.72 (0.09)	–0.81 (0.259)
Bathurst Island	3	2	0.67 (0.31)	0.00 (1.000)
Cambridge Bay	15	7	0.78 (0.10)	–1.19 (0.130)
Banks Island	10	5	0.76 (0.13)	–0.97 (0.190)
Alaska	19	13	0.96 (0.03)	–1.07 (0.157)
St. George Island	2	2	1.00 (0.50)	0.00 (1.000)
St. Paul Island	6	6	1.00 (0.10)	0.28 (0.599)
Bering Island	4	2	0.50 (0.27)	–0.71 (0.272)
Mednyi Island	7	3	0.52 (0.21)	–0.24 (0.461)
East Siberia	14	6	0.60 (0.15)	–1.17 (0.113)
Taimyr Peninsula	16	8	0.70 (0.13)	0.30 (0.660)
West Siberia	11	6	0.84 (0.09)	–0.67 (0.277)
Kola Peninsula	16	8	0.80 (0.09)	1.28 (0.916)
Svalbard	35	10	0.81 (0.05)	0.49 (0.704)
Scandinavia	67	7	0.61 (0.05)	0.86 (0.826)

DL-H16498 (5′CCTGAACTAGGAACCAGATG-3′) (Kocher *et al*. 1989) were used to amplify the fragment of the control region. Each polymerase chain reaction (PCR) mixture contained approximately 100 ng of sample DNA, and 1 mM dNTP in a reaction buffer of 50 mM KCl, 2.5 mM $MgCl_2$, 10 mM Tris-HCl (pH 8.8), and 2.5 U of *Taq* DNA polymerase in a total volume of 50 μL. Reactions contained 25 pmol of each primer. Thirty-five cycles of amplification were performed in a programmable thermal cycler (PerkinElmer-Cetus, Model 480). Each cycle consisted of denaturation at 94 °C for 60 s, annealing at 50 °C for 120 s, and extension at 72 °C for 90 s. Double-stranded products were separated in a 2% NuSieve (FMC Corp.) agarose gel in TAE buffer. After staining with ethidium bromide, the appropriate band was excised, the DNA extracted using a GENECLEAN Kit (BIO 101), dried by speed-vacuum, and eluted in 11 μL ddH_2O. Direct sequencing of double-stranded DNA was carried out using modifications of dimethyl sulphoxide (DMSO)-based protocols (Winship 1989; Green *et al*. 1990) and a Sequenase Version 2.0 kit (US Biochemicals). The sequencing reaction products were separated by electrophoresis in a 6% polyacrylamide gel for 3 h at 55 W in a Stratagene Base Ace Sequencing apparatus and sequence autorads were scored on an IBI gel reader or sequenced using an ABI automated sequencer and the ABI PRISM dye terminator cycle sequencing kit (PerkinElmer). Sequences were aligned first by eye and then by using CLUSTAL_X (Thompson *et al*. 1997), and rechecked by eye. We classified all samples into haplotypes using the program COLLAPSE (version 1.2, Posada 2004).

We also typed 163 individuals from nine populations using 11 polymorphic microsatellites (Table 2). These dinucleotide microsatellite loci (147, 155, 172, 225, 246, 263, 366, 431, 442, 453, 606) were identified from a domestic dog genomic library (Ostrander *et al*. 1993; Ostrander *et al*. 1995; Mellersh *et al*. 1997). Microsatellite alleles were detected from genomic DNA by end-labelling one primer with alpha-P32 ATP (Amersham) and T4 polynucleotide kinase reaction (Sambrook *et al*. 1989). We performed 28 cycles of PCR amplification in a 20-μL reaction volume using 50 ng of target DNA, 2 μL of formamide loading dye, and by heating to 95 °C for 5 min before loading onto a 6% sequencing gel containing 50% (w/v) urea. An M13 control sequencing reaction was run adjacent to the samples to provide an absolute-size marker for the microsatellite alleles. Gels were then autoradiographed overnight.

To fit the most probable nucleotide substitution model, we used MODELTEST (version 3.7; Posada & Crandall 1998). Gene diversity (*H*; Nei 1987), Tajima's test of selective neutrality (*D*; Tajima 1989), pairwise ϕ_{ST} (Excoffier *et al*. 1992) and corrected average pairwise difference (D_A; Nei & Li 1979) were calculated using ARLEQUIN (version 2000; Schneider *et al*. 2000). The best fitted nucleotide substitution model was used in the calculation of pairwise ϕ_{ST} and D_A. We used Tajima's test of selective neutrality to detect the signature of selection or population expansion (Tajima 1989).

Mean number of alleles per locus, observed heterozygosity, and Nei's genetic diversity (Nei 1987) for each population were calculated using GENECLASS2 (Piry *et al*. 2004). Tests for heterozygosity deficit and excess relative to the expected from the Hardy–Weinberg equilibrium were calculated following Rousset & Raymond (1995). Exact *P* values were obtained by the Markov Chain method using GENEPOP (Raymond & Rousset 1995). We used *M*-statistics to test for the signature of population bottleneck (Garza & Williamson 2001). The *M*-statistics is the ratio between the number of alleles and the range of allele sizes. According to simulations, any data set with seven microsatellite loci or more and a value of *M* smaller than 0.68 can be assumed to have gone through a recent reduction in population size (Garza & Williamson 2001). The initial parameters for the calculations of the *M*-statistics were θ (4* Ne* mutation rate) = 4, P_S (the proportion of non-one-step mutations) = 0.1, and Δ_g (the mean size of non-one-step mutations) = 3.5. Pairwise F_{ST} (θ) between populations was estimated as in Weir & Cockerham (1984). Finally, we assigned individuals into clusters using the Bayesian Markov chain Monte Carlo (MCMC) model implemented in STRUCTURE (version 2; Pritchard *et al*. 2000). We selected 100 000 as the burn-in length for MCMC runs, and $K = 20$ as the maximum number of populations to test. We tested for model convergence by several program runs, each with increased burn-in length. The optimal number of populations was determined as the *K*-value beyond which $\log \Pr(X \mid K)$ and α kept relatively constant or declined.

Genetic similarity among populations was evaluated by two-dimensional projection of the mtDNA ϕ_{ST}, D_A and microsatellite θ matrices using PROXCAL Multidimensional Scaling (MDS) in SPSS (version 12, SPSS Inc.). To illustrate the shortest dispersal route between populations, we connected populations on the MDS space using a Minimum-Spanning Tree (MST; SYSTAT version 11, SPSS Inc.) that identifies populations with the highest genetic similarity. Geographical distance (GD) is a key predictor in many population genetics studies. However, for the Arctic fox to reach any island, pack ice or ice floes must be present. Therefore, we weighted the geographical distance to each island by the probability of encountering ice floes near it at the annual peak in pack ice (i.e. geographical distance/probability of encountering ice floe during March). This weighted geographical distance (GDI) is essentially an interaction term between GD and sea ice occurrence. Pairwise geographical distances between mainland sites were weighted by 1 (i.e. values were not changed). The probability

Table 2 Corrected average pairwise difference (D_A; below diagonal) and pairwise ϕ_{ST} (above diagonal) calculated from mtDNA control region sequences of 20 Arctic fox populations. The numerical column headers correspond to the population row headers

	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20
1. Iceland		0.40	0.40	0.53	0.58	0.46	0.49	0.31	0.38	0.18	0.15	0.30	0.84	0.86	0.36	0.43	0.40	0.47	0.48	0.41
2. E. Greenland	1.31		0.21	0.39	0.12	0.08	0.11	0.07	–0.04	0.05	0.49	0.25	0.84	0.83	0.06	0.07	0.00	0.14	0.12	0.05
3. S. Greenland	1.44	0.60		0.26	0.47	0.28	0.07	0.10	0.16	0.05	0.37	0.20	0.81	0.82	0.15	0.22	0.22	0.32	0.32	0.10
4. W. Greenland	2.40	1.39	0.89		0.53	0.55	0.34	0.40	0.42	0.18	0.50	0.17	0.84	0.84	0.43	0.40	0.39	0.46	0.47	0.25
5. N. Greenland	2.25	0.20	1.44	1.69		0.38	0.62	0.37	0.29	0.20	0.90	0.36	0.95	0.89	0.35	0.22	0.12	0.27	0.20	0.17
6. Churchill	1.42	0.12	0.63	1.99	0.65		0.18	0.02	0.00	0.12	0.58	0.42	0.88	0.87	0.01	0.10	0.12	0.28	0.23	0.11
7. Bathurst Is.	1.93	0.30	0.33	1.36	0.68	0.27		0.10	0.11	0.04	0.66	0.12	0.88	0.82	0.05	0.03	0.10	0.22	0.24	–0.08
8. Cambridge B.	0.89	0.15	0.26	1.51	0.94	0.03	0.35		–0.02	0.01	0.34	0.26	0.82	0.83	–0.02	0.11	0.11	0.24	0.24	0.08
9. Banks Is.	1.17	–0.06	0.41	1.49	0.44	–0.01	0.23	–0.02		0.02	0.50	0.25	0.86	0.84	–0.03	0.05	0.02	0.18	0.16	0.03
10. Alaska	0.63	0.23	0.25	0.88	0.91	0.37	0.60	0.06	0.14		0.02	0.07	0.71	0.76	0.04	0.11	0.08	0.20	0.23	0.08
11. St. George Is.	0.56	2.00	1.76	2.71	2.81	1.80	2.39	1.36	1.72	0.84		–0.01	0.90	0.82	0.40	0.40	0.41	0.50	0.53	0.40
12. St. Paul Is.	0.90	0.98	0.88	0.72	1.54	1.41	1.31	0.93	1.00	0.19	1.09		0.68	0.73	0.29	0.31	0.25	0.34	0.44	0.28
13. Bering Is.	10.09	9.25	9.98	11.89	10.39	9.17	9.81	9.18	9.27	9.57	10.89	10.74		0.55	0.84	0.81	0.82	0.81	0.84	0.79
14. Mednyi Is.	13.28	12.21	12.85	14.78	13.24	12.01	12.65	12.11	12.18	12.52	13.80	13.71	3.24		0.84	0.83	0.82	0.83	0.86	0.82
15. E. Siberia	1.07	0.12	0.38	1.59	0.72	0.01	0.15	–0.03	–0.05	0.15	1.35	1.07	9.06	11.90		0.03	0.08	0.23	0.23	0.04
16. Taimyr Pen.	1.57	0.17	0.70	1.61	0.54	0.18	0.21	0.28	0.11	0.43	1.83	1.35	9.58	12.38	0.07		0.05	0.11	0.17	0.03
17. W. Siberia	1.37	0.00	0.70	1.58	0.25	0.20	0.38	0.28	0.05	0.35	1.89	1.13	9.58	12.48	0.17	0.13		0.06	0.00	0.07
18. Kola Pen.	1.96	0.40	1.18	2.14	0.75	0.70	0.86	0.77	0.51	0.90	2.85	1.67	10.01	12.99	0.68	0.29	0.16		0.05	0.17
19. Scandinavia	1.85	0.28	0.97	1.83	0.56	0.58	0.74	0.64	0.39	0.71	2.54	1.44	10.20	13.14	0.59	0.42	–0.02	0.09		0.18
20. Svalbard	1.65	0.15	0.28	0.85	0.58	0.27	0.02	0.21	0.10	0.26	2.06	0.92	9.80	12.63	0.12	0.10	0.21	0.54	0.47	

of encountering sea ice during March (peak in annual sea ice) at the vicinity of each island was calculated from a map of sea ice occurrence in the Arctic Circle between 1972 and 1990 (Parkinson *et al*. 1999; Tanis & Smolyanitsky 2000). This map provides frequency isoclines of ice occurrence for all sites included in this study. We also used maps in Brower *et al*. (1988) for greater details on ice occurrence in the Bering Sea. Using these resources, we calculated sea ice occurrence probabilities of 0.05 for the Commander Islands, 0.15 for the Pribilof Islands, 0.05 for Iceland, 0.40 for South Greenland, 0.70 for west Greenland, and 1 for all other sites. To test for isolation by distance, we correlated weighted geographical distance and the mtDNA ϕ_{ST}, D_A and microsatellite θ matrices using the Mantel's test (Mantel 1967).

In addition to the hypothesis that sea ice occurrence can predict genetic structure in Arctic foxes, we tested four other nonexclusive alternative hypotheses using distance-based redundancy analysis (McArdle & Anderson 2001) and the BIOENV procedure (Clarke & Ainsworth 1993). The alternative hypotheses were (i) Dispersing individuals avoid extreme weather and are consequently expected to move towards areas of more moderate climate. To reflect the influence of climate, we used annual average of minimum temperature in November (National Climate Data Center, Monthly Global Surface data 1980–2005), a peak time of subadults dispersal and food stress (Audet *et al*. 2002), as an independent variable. During storms or in unusually cold or windy weather, Arctic foxes seek shelter in a temporary den or snow burrow (Frafjord 1992). Severe conditions may make travel over great distances more difficult or restrict access to key food resources, such as from the sea, by an early freeze. (ii) Arctic fox dispersal is dependent on the presence of polar bears (*Ursus maritimus*). Foxes follow polar bears to scavenge remains of kills (Audet *et al*. 2002). This behaviour enables dispersers to cross extensive icefields where no alternative food is available. We assume that if a polar bear can cross the ice pack so can the Arctic fox. We used bear occurrence (presence or absence) at each site to test for linkage between polar bear–Arctic fox large-scale movements and genetic structure. (iii) High density of the red fox (*Vulpes vulpes*) may restrict Arctic fox movements (e.g. Hersteinsson & Macdonald 1992). It is well documented that Arctic foxes are competitively excluded by the red fox at some localities (Dalén *et al*. 2004). We used red fox occurrence data (presence or absence) as an independent variable to test for its effect on site connectivity in Arctic foxes. (iv) Lastly, we categorized foxes as belonging to coastal and lemming ecotypes as defined by Dalén *et al*. (2005) to test whether dispersers move more within than across the habitats occupied by these two ecotypes. We tested the above six hypotheses (i.e. geographical distance, sea ice contribution, and the four listed above) by regressing each of the genetic distance matrices (i.e. mtDNA ϕ_{ST}, D_A and microsatellites θ) as the dependent variable and the five independent variables listed above using the programs DISTLM and DISTLM FORWARD (i.e. distance-based redundancy analysis, McArdle & Anderson 2001). DISTLM computes the amount of variance explained by all variables combined, whereas DISTLM FORWARD computes the contribution of each independent variable to the total variance explained. DISTLM executes two sets of analyses: (i) forward selection, where independent variables are entered in the order of their correlation; and (ii) sequential analysis, where each independent variable is entered separately in the order of their importance (i.e. partial r^2). The sequential procedure computes the exact added contribution of each independent variable to the total variance explained. The contribution of geographical distance and its interaction with sea ice occurrence (GDI) were examined separately because it was not feasible to include all these terms in a single regression model because of the small number of populations. In this analysis, significant independent variables with high coefficient of determination (r^2) are key components for understanding the mechanisms governing Arctic fox long-range movement.

To examine which subset of independent variables may provide the best model of differences in genetic structure among populations, we used the BIOENV procedure (Clarke & Ainsworth 1993). The basic principle underlying this approach is to calculate a Spearman's rank correlation coefficient (r_s) between the response distance matrix (i.e. a matrix of genetic distances) and the distance matrix calculated as the Euclidean distance among one or more predictor variables. The BIOENV statistic r_s is analogous to a nonparametric version of a simple Mantel correlation between two distance matrices. The BIOENV procedure calculates the value of r_s using every possible combination of predictor variables until it finds the 'best' fit (i.e. that combination of predictor variables whose Euclidean distance matrix yields the highest value of r_s). We implemented the BIOENV procedure and identified, for each of the three response matrices, the best fits. Note that the value of r_s (unlike r^2 in a multiple regression) does not necessarily increase with the number of predictor variables. A permutation test that accounts for the selection process is used for calculating the probability that the observed r_s is significantly different from no association. The BIOENV analysis was performed using the PRIMER computer package (version 6; Clarke & Gorley 2006).

Results

Mitochondrial DNA analysis

We analysed control region sequence data from 306 individuals and identified 60 unique mtDNA haplotypes (Table 1). The best nucleotide substitution model selected

by MODELTEST using the Akaike's Information Criterion (AIC) was the general time reversible plus Gamma (GTR + G; AIC = 1510.9). However, this model is not implemented in ARLEQUIN, the primary software package we used for the analysis of mtDNA data. Therefore, we used the Tamura–Nei plus Gamma (TrN + G) model that fitted the data nearly as well as the GTR + G model (AIC = 1513.0). For the TrN + G model, the fitted alpha value for the gamma correction was 0.126.

Gene diversity, the probability that two randomly chosen haplotypes are different in a population, was highest in the Pribilof Islands. In these populations, every individual sampled had a different mtDNA haplotype. West Greenland, Iceland, Scandinavia and the Commander Islands had lower gene diversities, but only in North Greenland was diversity dramatically low (0.25, Table 1). The largest number of haplotypes was observed in Alaska, Taimyr Peninsula, Kola Peninsula and Svalbard (Table 1). However, the number of haplotypes was correlated with sample size (sample size was log-transformed for linearity; $r^2 = 0.463$, $F_{1,18} = 15.5$, $P = 0.001$). After controlling by linear regression for the effect of sample size, Alaska, Taimyr Peninsula, Kola Peninsula and St. Paul Island populations showed significantly more haplotypes than expected (above the 95% confident intervals) and Mednyi Island, North Greenland, Iceland, and Scandinavia significantly fewer haplotypes than expected (below the 95% confident intervals). We did not detect the signature of selection or population expansion in any of the localities sampled (Table 1). These results show moderate to high levels of variation in most populations and suggest Northern Greenland is low in variation and may have been recently colonized or experienced a population bottleneck.

Mean pairwise ϕ_{ST} over all populations was exceptionally high for Bering and Mednyi Islands (0.829 and 0.828, respectively; Table 2). The Mednyi Island pairwise ϕ_{ST} values were all significantly larger than zero, and for the Bering Island only ϕ_{ST} with St. George Island was insignificant. Mean pairwise ϕ_{ST} was also relatively high, compared to the other populations such as Iceland (0.444), West Greenland (0.418), North Greenland (0.399), and St. George Island (0.443). Surprisingly, St. Paul Island had a much lower value of ϕ_{ST} (0.285) suggesting higher rates of immigration to the island. Mean pairwise ϕ_{ST} among the other populations ranged from –0.075 to 0.323. In general, mean pairwise ϕ_{ST} values identified the Commander Islands as genetically the most isolated.

The MDS analysis supported these findings. The MDS projection accounted for 97.3% (stress = 0.027) and 99.7% (stress = 0.002) of the variance in the ϕ_{ST} and D_A matrices, respectively (Fig. 2). Both MDS plots show that the two Commander Islands populations are distinct from all others (Fig. 2). St. George Island and Iceland populations were also unique. West and north Greenland populations

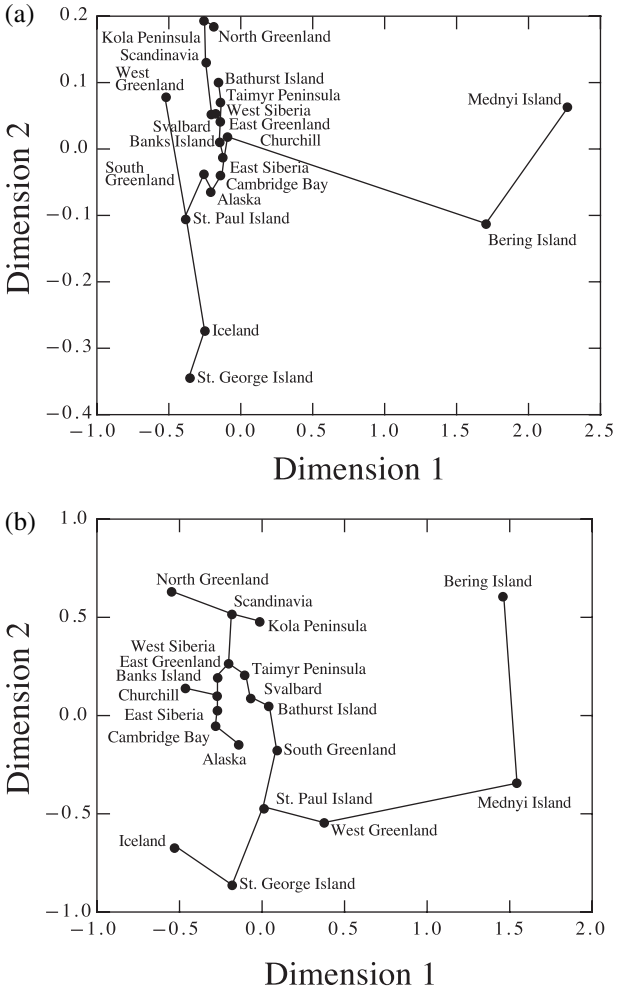

Fig. 2 Multidimensional scaling (MDS) of corrected average pairwise difference D_A (a) and pairwise ϕ_{ST} (b) distances. A minimum-spanning tree links populations.

are separate from the south and east populations of this island. Scandinavia is associated with the Kola Peninsula, but these two were also distinct from the main cluster of populations (Fig. 2). We tested two sets of population assemblages using analysis of molecular variance (AMOVA). The first assemblage consisted of the nine clusters that represent islands or distinct regions [(Iceland) (E Greenland, S. Greenland, W. Greenland, N. Greenland) (Churchill, Bathurst Island, Cambridge Bay, Banks Island) (Alaska) (St. George Island, St. Paul Island) (Bering Island, Mednyi Island) (E. Siberia, Taimyr Peninsula) (W. Siberia, Kola Peninsula, Scandinavia) (Svalbard)]. The second assemblage consisted of five clusters composed of islands and neighbouring mainland sites that may be linked in winter by ice pack [(Iceland, E Greenland, S. Greenland, W. Greenland, N. Greenland) (Churchill, Bathurst Island, Cambridge Bay, Banks Island) (Alaska, St. George Island,

St. Paul Island) (Bering Island, Mednyi Island, E. Siberia, Taimyr Peninsula) (W. Siberia, Kola Peninsula, Scandinavia, Svalbard)]. The second cluster was constructed based on the sea ice occurrence map in Parkinson *et al*. (1999). The AMOVA analysis showed that 31.6% of the variance between groups was explained in the first population assemblage ($\Phi_{ct} = 0.32$, $P < 0.0001$) whereas only 3.9% of the variance between groups was explained in the second assemblage ($\Phi_{ct} = 0.04$, $P = 0.018$). Consequently, the first model that separated islands into independent units explained more overall genetic variation than the second model that combined islands with their neighbouring mainland sites. In other words, the primary subdivisions in Arctic foxes are individual islands rather than island–mainland groupings, which contain much greater genetic heterogeneity. The above analysis suggests that proximity between islands and the nearby mainland is a poor predictor of genetic association.

Microsatellite analysis

Our analysis of microsatellite data found considerably lower mean number of alleles for the Commander and the Pribilof Islands (Table 3). The mean number of alleles was independent of sample size ($r^2 = 0.22$, $F_{1,7} = 1.93$, $P = 0.21$). Heterozygosity and Nei's genetic diversity were also lower on these islands, with Mednyi Island being the lowest. For comparison, Svalbard, which is reconnected to the mainland every year by the seasonal advance and retreat of the pack ice, showed diversity similar to Scandinavia and the Kola Peninsula. We did not detect heterozygosity excess at any site, but heterozygosity deficiency from that expected under Hardy–Weinberg equilibrium was observed in Bering and St. Paul Island, Svalbard and Scandinavia (Table 3), but after applying a sequential Bonferroni correction, only the Bering Island population was significantly heterozygote deficient. However, none of the loci deviated consistently from Hardy–Weinberg equilibrium. A recent population bottleneck was detected only in the Bering Island population ($M = 0.66$) and that value was significantly smaller than the expected ratio at equilibrium (Table 3). All other populations had $M > 0.68$ (Table 3).

We selected $K = 7$ as the optimal number of clusters for the assignment analysis ($\Pr(X \mid K) = -4930.7$, $a = 0.058$) because at $K = 8$ the $\Pr(X \mid K)$ increased $\leq 0.7\%$ at each K step. Convergence of the STRUCTURE algorithm on $K = 7$ was detected after 20 000 burn-ins. Individuals of the Commander and the Pribilof Islands were assigned with high confidence to their own clusters (Table 4). About 91% of individuals from the Pribilof Islands were assigned to clus-

Table 3 Sample size (N), mean number of alleles per locus (N_A; 11 microsatellite loci), observed heterozygosity (H_O), probabilities of heterozygote (*Ht*) deficit and excess, Nei's genetic diversity and the *M*-statistics for nine Arctic fox populations. The geographical position for each site is indicated in Fig. 1

Site	N	N_A (±SD)	H_O (±SD)	*Ht* deficit	*Ht* excess	Nei's genetic diversity (±SD)	*M*-statistics (*P*)
Alaska	17	7.4 ± 2.2	0.77 ± 0.11	0.088	0.911	0.79 ± 0.08	0.78 (0.391)
Bering Island	17	4.0 ± 1.8	0.50 ± 0.17	0.001	0.999	0.56 ± 0.18	0.66 (0.013)
Mednyi Island	17	2.5 ± 1.8	0.19 ± 0.28	0.074	0.909	0.20 ± 0.24	0.88 (0.938)
Scandinavia	33	6.8 ± 2.3	0.64 ± 0.23	0.009	0.992	0.70 ± 0.19	0.75 (0.137)
Kola Peninsula	14	6.0 ± 2.1	0.69 ± 0.16	0.097	0.919	0.72 ± 0.14	0.77 (0.398)
Taimyr Peninsula	22	8.0 ± 3.1	0.74 ± 0.17	0.189	0.841	0.77 ± 0.10	0.82 (0.604)
St. George Island	7	2.6 ± 1.1	0.48 ± 0.34	0.277	0.718	0.50 ± 0.23	0.85 (0.899)
St. Paul Island	21	4.4 ± 1.3	0.51 ± 0.15	0.027	0.976	0.60 ± 0.12	0.76 (0.241)
Svalbard	15	6.9 ± 2.0	0.71 ± 0.17	0.015	0.989	0.76 ± 0.09	0.69 (0.056)

Table 4 Proportion of population membership in each of the seven clusters identified by STRUCTURE. Boxes indicate high assignment proportions for the individuals from the Commander and the Pribilof Islands

Clusters	1	2	3	4	5	6	7
Alaska	0.103	0.137	0.090	0.192	0.023	0.039	0.415
Bering Island	0.012	0.007	0.007	0.007	0.910	0.037	0.020
Mednyi Island	0.019	0.008	0.007	0.007	0.030	0.922	0.007
Scandinavia	0.022	0.427	0.310	0.164	0.015	0.024	0.040
Kola Peninsula	0.018	0.101	0.091	0.653	0.026	0.018	0.093
Taimyr Peninsula	0.060	0.123	0.042	0.159	0.019	0.021	0.577
St. George Island	0.922	0.007	0.021	0.013	0.022	0.005	0.010
St. Paul Island	0.903	0.010	0.022	0.032	0.008	0.008	0.017
Svalbard	0.107	0.146	0.148	0.118	0.011	0.016	0.454

Table 5 Pairwise F_{ST} (θ) calculated from 11 microsatellite loci for nine Arctic fox populations

Populations	Alaska	Bering Island	Mednyi Island	Scandinavia	Kola Peninsula	Taimyr Peninsula	St. George Island	St. Paul Island	Svalbard
Alaska	0.00								
Bering Island	0.19	0.00							
Mednyi Island	0.37	0.41	0.00						
Scandinavia	0.06	0.23	0.36	0.00					
Kola Peninsula	0.04	0.23	0.43	0.08	0.00				
Taimyr Peninsula	0.01	0.21	0.38	0.06	0.06	0.00			
St. George Island	0.19	0.30	0.60	0.24	0.22	0.16	0.00		
St. Paul Island	0.12	0.29	0.43	0.19	0.20	0.13	0.12	0.00	
Svalbard	0.03	0.24	0.42	0.07	0.08	0.01	0.19	0.14	0.00

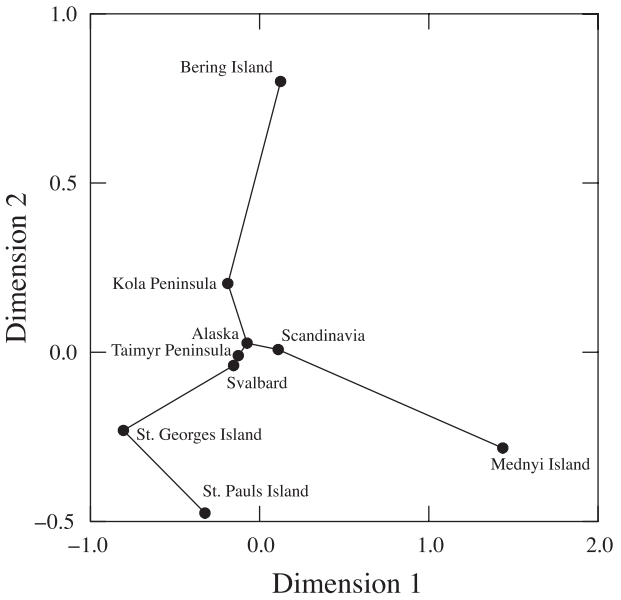

Fig. 3 Multidimensional scaling (MDS) of pairwise θ distances. A minimum-spanning tree links populations.

ter 1, 91% of individuals from Bering Island were assigned to cluster 5, and 92% of individuals from Mednyi Island were assigned to cluster 6. No other populations dominated clusters 1, 5 and 6. Individuals from Scandinavia were mostly assigned to clusters 2 and 3 (73%), and most of the individuals from the Kola Peninsula were assigned to cluster 4. Finally, cluster 7 included individuals mostly from Alaska, Taimyr Peninsula, and Svalbard (Table 4).

The MDS projection of the θ matrix (Table 5) supported the pattern observed by the assignment tests. In the MDS projection, Mednyi and Bering Islands have high positive values on dimension one and two, respectively, whereas the Pribilof Islands have low negative values on dimension two. The other five populations are clustered at the centre of the space without a clear geographical pattern (Fig. 3). However, this analysis supported the geographical isolation of both the Commander and the Pribilof Islands.

Geographical distance, corrected for probability of sea ice occurrence, accounted for 57.3% ($P < 0.001$), 58.5% ($P < 0.001$), and 47.2% ($P < 0.001$) of the variance in mtDNA ϕ_{ST}, D_A and microsatellite θ genetic distance matrices, respectively (Fig. 4). The sequential regression showed that for the microsatellite data, GDI was the only significant independent variable, explaining about 42% of the variance in the θ matrix (Table 6). Geographical distance alone explained about 25% (range 22.4–27.7%) of the variance in the genetic distance calculated from mtDNA data. However, the weighting for ice occurrence doubled the variance explained (about 53%; range 44.6–61.1%; Table 6). The sequential analyses showed that weighted geographical distance for sea ice occurrence was the most significant factor and accounted for most of the genetic variance in all three cases. Ecotype was the only other independent variable showing a meaningful contribution to the variance whereas only 11.6% of the variance in mtDNA ϕ_{ST} was explained when geographical distance alone was considered (Table 6). However, this contribution was no longer significant after weighting for sea ice occurrence. In general, weighted geographical distance accounted for about 50% of the variance in the genetic distances, and contributed about 80% of the total variance explained by the regression analysis (i.e. mean $r^2 = 0.613$; Table 6).

For all three genetic distances (mtDNA ϕ_{ST}, D_A and microsatellite θ) the best variable subset (i.e. the combination of the fewest predictors that accounted for maximum of the variance in the genetic distance matrix), found by the BIOENV procedure, did not include geographical distance as a prime predictor (Table 6). Ecotype and polar bear occurrence explained more of the variance than geographical distance. In contrast, geographical distance weighted for ice occurrence (GDI) is the sole best predictor for all genetic distances (Table 6).

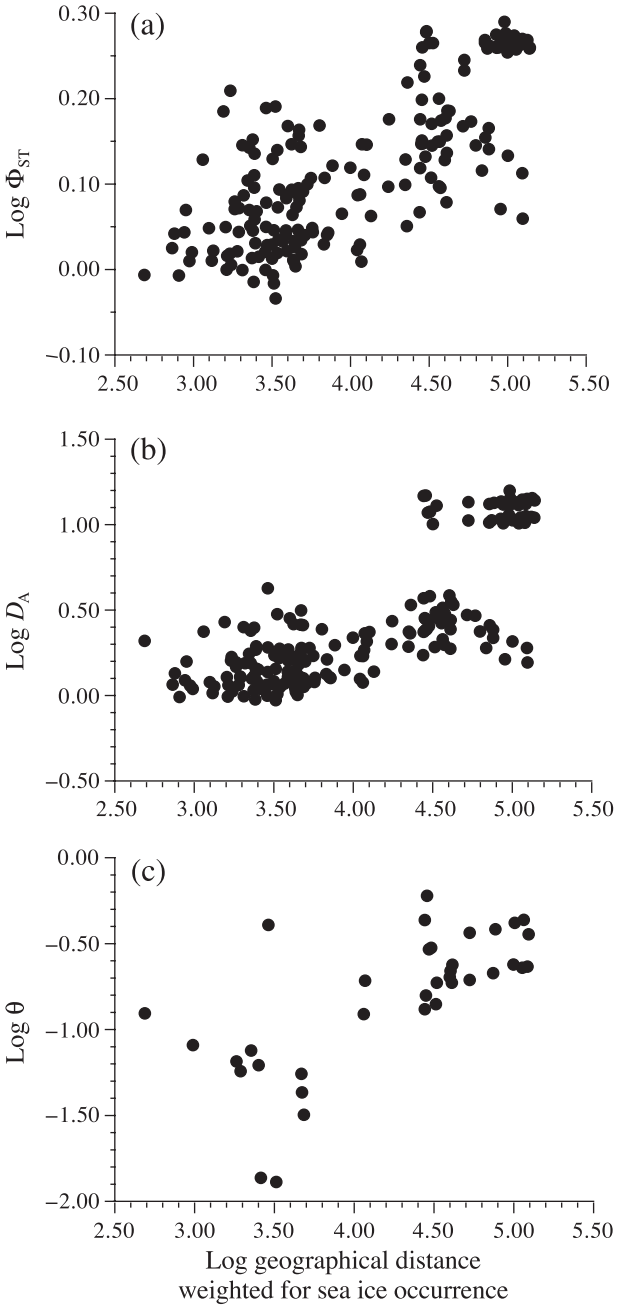

Fig. 4 Correlation between geographical distance weighted for sea ice occurrence, and three genetic distances (Mantel's test; ϕ_{ST}, $r^2 = 0.573$, $P < 0.001$; D_A, $r^2 = 0.585$, $P < 0.001$; θ, $r^2 = 0.472$, $P < 0.001$). Data were log-transformed for linearization.

Discussion

Genetic variability in island populations

Island populations commonly have lower levels of genetic variability reflecting their geographical isolation and small population size and the potential from inbreeding depression (Frankham 1997, 1998). In canids, island populations have been documented to have lower levels of variation (e.g. Isle Royale gray wolf, *Canis lupus*, Wayne *et al*. 1991a; island fox, *Urocyon littoralis*; Gilbert *et al*. 1990; Darwin's fox, *Dusicyon fulvipes,* Yahnke *et al*. 1996; Phillip Island red fox, *Vulpes vulpes*, Lade *et al*. 1996). However, none of these populations have apparent inbreeding depression, although the island foxes may be more susceptible to epizootics (Aguilar *et al*. 2004) and an isolated population of Swedish grey wolves suffers from inbreeding depression (Liberg *et al*. 2005). We found that despite apparent geographical isolation on High Arctic islands, the majority of Arctic fox populations have high levels of variability comparable to levels found in mainland populations suggesting gene flow and population size are sufficient to maintain high levels of variation on most Arctic islands. The lowest genetic diversity is apparent on the most isolated islands, namely the Commander Islands, which have low levels of mtDNA and microsatellite variation. These islands have relatively small populations (e.g. $n < 150$, Goltsman *et al*. 2005), may have gone through recent bottlenecks (e.g. Mednyi Island, Goltsman *et al*. 2005) and may have not recently been connected to mainland sources of migration by sea ice. In contrast, island populations that are connected annually by sea ice to sources of migrants, such as Svalbard, have levels of variation similar to mainland populations. Consequently, these results support the effect of periodic gene flow in maintaining levels of variation in islands populations of Arctic foxes.

Genetic structure of Insular Arctic foxes

Mitochondrial DNA and microsatellite data suggested that the basal unit of subdivision in Arctic foxes corresponds to individual island populations or regional groupings. AMOVA analysis finds about 32% of the variance between island groupings ($\Phi_{ct} = 0.32$, $P < 0.0001$) whereas only 3.9% is found between couplings of island and mainland populations ($\Phi_{ct} = 0.04$, $P = 0.018$). Similarly, STRUCTURE analysis identifies seven groupings that correspond largely to island populations with some islands, such as the Pribilof and Commander islands, having correct assignments of > 90% (Table 4). However, other groupings defined by structure show only weak correspondence with geography implying ongoing gene flow or past episodes of admixture. Finally, MDS analyses for both mtDNA and microsatellite data clearly establish the Commander Islands (Mednyi and Bering Island) as the most genetically distinct living populations of Arctic fox that has been surveyed. In all analysis, these populations appear genetically distinct (mean ϕ_{ST} mtDNA = 0.83, F_{ST} microsatellites = 0.34). Additionally, the genetic results support genetic distinction of the Pribilof island

Table 6 Sequential regression and BIOENV analyses for theta (θ, microsatellites), Φ_{ST} (mtDNA) and D_A (mtDNA) distance matrices set up as the dependent variable, and five independent variables: geographical distance (GD) or geographical distance weighted by ice occurrence (GDI), average minimum temperature in November (TM), red fox presence (RF), polar bear presence (PB), and ecotype (ET). DF for θ is (1,8) and for Φ_{ST} and D_A is (1,19)

	Sequential regression				BIOENV		
Variable	r^2	Cumulative r^2	F	P	Five best subsets	r_s	P
	Geographical distance						
θ (Microsatellites)							
GD	0.056	0.056	0.4	0.631	ET	0.022	0.777
Polar bear	0.036	0.091	0.2	0.799	ET, RF	–0.063	0.929
Temperature	0.007	0.098	0.0	0.916	RF	–0.144	0.996
Red fox	0.000	0.098	0.0	0.999	ET, PB	–0.156	1.000
Ecotype	0.000	0.098	0.0	0.999	ET, PB, RF	–0.157	1.000
Φ_{ST} (mtDNA)							
GD	0.224	0.224	5.2	0.001	ET, PB	0.409	< 0.001
Ecotype	0.116	0.340	3.0	0.029	PB	0.334	0.003
Temperature	0.039	0.379	1.0	0.439	ET	0.304	0.006
Red fox	0.015	0.394	0.4	0.763	GD	0.300	0.006
Polar bear	0.004	0.398	0.1	0.892	GD, RF	0.300	0.006
D_A (mtDNA)							
GD	0.277	0.277	6.9	0.003	ET, PB	0.395	0.002
Temperature	0.053	0.330	1.3	0.292	ET	0.332	0.007
Red fox	0.034	0.364	0.8	0.382	GD, TM	0.304	0.010
Ecotype	0.009	0.373	0.2	0.648	GD, TM, RF	0.304	0.010
Polar bear	0.005	0.378	0.1	0.815	GD, TM, PB	0.304	0.010
	Geographical distance weighted for sea ice occurrence						
θ (Microsatellites)							
GDI	0.422	0.422	5.1	0.016	GDI	0.580	0.041
Temperature	0.082	0.504	1.0	0.382	GDI, TM	0.580	0.041
Red fox	0.055	0.560	0.6	0.515	GDI, RF	0.580	0.041
Polar bear	0.011	0.570	0.1	0.801	GDI, PB	0.580	0.041
Ecotype	0.014	0.584	0.1	0.780	GDI, ET	0.580	0.041
Φ_{ST} (mtDNA)							
GDI	0.446	0.446	14.5	< 0.001	GDI	0.670	< 0.001
Ecotype	0.062	0.508	2.1	0.107	GDI, TM	0.670	< 0.001
Polar bear	0.036	0.544	1.3	0.365	GDI, RF	0.670	< 0.001
Temperature	0.008	0.552	0.3	0.794	GDI, PB	0.670	< 0.001
Red fox	0.001	0.553	0.0	0.865	GDI, ET	0.670	< 0.001
D_A (mtDNA)							
GDI	0.611	0.611	28.3	0.005	GDI	0.699	< 0.001
Temperature	0.067	0.678	3.6	0.071	GDI, TM	0.699	< 0.001
Ecotype	0.016	0.694	0.9	0.371	GDI, RF	0.699	< 0.001
Polar bear	0.006	0.700	0.3	0.594	GDI, PB	0.699	< 0.001
Red fox	0.002	0.702	0.1	0.842	GDI, ET	0.699	< 0.001

populations (St. Paul and St. George islands) as well as Iceland and West Greenland (Table 3, Figs 2 and 3). We suggest that the genetic distinction of these island populations is in large part due to the absence of pack ice and infrequent transport of foxes to the islands by ice floes.

Determinants of genetic variation

We examined the influence of six hypothetical barriers to dispersal on genetic variance among populations of insular Arctic foxes. These hypothetical barriers were based on documented behaviour. The first hypothesis derives from the observation that in many species, individuals avoid extreme conditions. The best examples are terrestrial species showing migratory behaviour, where individuals may sometimes transverse large distances to escape harsh winter climates (e.g. Ferguson & Elkie 2004). Although the Arctic fox is highly adapted for cold conditions, young, nonterritorial individuals likely have a better chance of survival in more moderate climate regimes, where food

may be more accessible and long distance travel more feasible (e.g. Roth 2003). Previously, we found temperature to be an important variable in explaining genetic differentiation in grey wolves (Geffen *et al*. 2004). Our second hypothesis stems from the fact that polar bears kills nourish Arctic foxes when on sea ice (Audet *et al*. 2002). The polar bear is the only species that is able to survive for long time periods on the sea ice by stalking seals and small whales at breathing holes (DeMaster & Stirling 1981). For Arctic foxes, no other food resource is available on sea ice. Consequently, the survival of foxes traveling on the ice should be linked to the presence of polar bears. The third hypothesis is rooted in the idea that dispersers, who are young with limited experience, disperse to areas with prey similar to their natal habitat (Geffen *et al*. 2004). In Arctic foxes, dispersing individuals that developed hunting and scavenging skills for marine organisms and sea birds should favour coastal sites, whereas those raised on lemming and reindeer meat should favour inland sites (Tannerfeldt & Angerbjorn 1998). The fourth hypothesis is based on studies suggesting severe interspecific competition between red and Arctic foxes, which results in a competitive exclusion and range contraction of Arctic foxes (Hersteinsson & Macdonald 1992; Dalén *et al*. 2004). The fifth hypothesis, isolation by distance, is the most commonly assumed in population genetics studies (Slatkin 1993) and presumes distance alone as well as increasing physical obstacles to dispersal (e.g. mountains, rivers, etc.) increase levels of isolation and genetic differentiation. The final hypothesis addresses the fact that many of the locations we studied are on islands, which are separated by a water barrier only bridged by transport on pelagic ice floes.

Our analysis provides no support for the impact of red fox presence on genetic differentiation and little support for the extreme temperature hypothesis (Table 6). Furthermore, geographical distance alone explains only a maximum of 28%, and combined with ecotype explains a maximum of 34% of the variance in genetic distance based on mtDNA (Table 6). All of these predictors were not significantly associated with genetic differentiation based on microsatellite loci. However, the above variables were all secondary in comparison to the explanatory contribution of sea ice occurrence, which explained 40–60% of the variance in genetic distance for both mtDNA and microsatellite data sets and was the principal factor in both analyses (Table 6). Sea ice occurrence and transport distance are the primary determinants of population structure in Arctic foxes of the High Arctic islands. The previous observation of a lack of differentiation by distance geographical distance in Arctic foxes (Dalén *et al*. 2005) probably applies only to populations connected by seasonal or permanent pack ice. Apparently, geographical distance may not be a significant barrier when travelling across the pack ice but bridging open waters is highly dependent on the presence of floating ice.

Island populations of canids are usually well isolated and genetically distinct (Gilbert *et al*. 1990; Wayne *et al*. 1991b; Lade *et al*. 1996; Yahnke *et al*. 1996). However, with the exception of the Commander and Pribilof Islands, islands of the Arctic are often physically isolated but nonetheless open to migration by species that can survive on sea ice. Pack and drifted ice connect these islands and the mainland at different temporal and spatial scales. During the last peak glaciation, about 20 000 years ago, all the islands in the High Arctic were connected by land or pack ice (Grosswald 1998; Ager 2003; Schäfer-Neth & Paul 2003). More recently, during the little ice age which ended only 150 years ago (Crowley 2000), all the islands in our study including the Commander and the Pribilof Islands may have been intermittently connected for short periods by pack ice during winter. Currently, several of the islands are connected to the mainland by seasonal pack ice (Banks Island, Bathurst Island, Greenland and Svalbard) or are physically isolated because the sea surrounding them never freezes (Commander Islands, Pribilof Islands and Iceland). However, the latter islands are reached by ice floes at varying rates. Specifically, the probability that ice floes would reach the Commander Islands, Pribilof Islands, and Iceland is very low (0.05, 0.15 and 0.05, respectively). Furthermore, when sea ice is present near these islands it generally exists as isolated icebergs, and not as a continuous ice sheet, which may further restrict passage of Arctic foxes. Consequently, ongoing migration to the Commander Island and Pribilof Islands is likely to be very low or nonexistent for several generations at a time with an influx of individuals occurring during relatively brief intervals of cold weather. This relative isolation is supported by our genetic results, which show high levels of F_{ST} that imply low levels of migration to these islands (Tables 2 and 5). Fossil evidence on the Pribilof Islands documents the presence of foxes there at least 13 000 years ago (Guthrie 2004). In conclusion, survival on sea ice and ice floes is a key adaptation in the polar regions because it may greatly enhance the possibility of colonization of remote islands potentially rich in resources but with fewer competitors or predators. Indeed, the Arctic fox can survive long periods without food and they have a low cost of locomotion (Fuglei & Oritsland 1999; Fuglei & Oritsland 2003).

Conservation implications

The classification of the Commander and Pribilof Island populations as separate subspecies of Arctic fox is well supported by our analyses (Audet *et al*. 2002). Both mtDNA and microsatellite data indicate that these populations are unique, and genetically distinct and thus deserve special attention from a conservation perspective.

This outcome fits with the fact that these are island populations, which support small populations where inbreeding and genetic drift may have a profound effect on genetic diversity and differentiation (Frankham 1997, 1997). We did not detect the signature of selection or population expansion in any of these island populations, but Bering Island foxes showed a recent reduction in population size and on three islands, heterozygosity was significantly lower than expected at equilibrium (Tables 1 and 3). A population crash on Mednyi Island occurred in the 1970s because of mange and reduced the population to about 90 individuals (Goltsman *et al*. 1996, 2005) and this island has low mtDNA and microsatellite variability. Conceivably, similar population declines may have occurred historically on Bering Island given transport of the mange mite by seafarers (Goltsman *et al*. 1996). Limited morphologic studies of the Commander Island populations and studies of other island canids suggest that selection may operate differently there than on the mainland and that island populations may be divergent in morphology, life history and adaptive traits (Wayne *et al*. 1991b; Roemer *et al*. 2001; Roemer & Wayne 2003; Aguilar *et al*. 2004; Goltsman *et al*. 2005). Consequently, the Commander and the Pribilof Islands population may qualify, genetically and ecologically, as an evolutionary significant unit (Crandall *et al*. 2000). Bering Island appears to be the most distinct genetically and given susceptibility to disease, may warrant high priority for conservation.

The extent of sea ice is affected by global warming (Vinnikov *et al*. 1999). Pack ice has shown dramatic reductions over the past decade (Parkinson *et al*. 1999) and much of the Arctic Circle may be free of pack ice in 8–10 decades (Vinnikov *et al*. 1999). Although the absence of pack ice provides new opportunities for global shipping, the seasonal ice connection between many Arctic islands will likely be lost and the small isolated populations of foxes that remain will lose genetic diversity and have higher levels of inbreeding and genetic divergence. Possibly counteracting this isolation is an increased rate of calving from disappearing glaciers (e.g. Broecker 1994). However, pelagic ice floe transport will be dependent on prevailing currents resulting in extreme isolation of some islands no longer connected seasonally by pack ice. One result of the loss of pack ice will be the disruption of migratory patterns of Arctic foxes because of their interaction with polar bears. In fact, the disruption has already begun as loss of pack ice has caused increased starvation in polar bears (Stirling & Parkinson 2006) that probably have concomitant effects on their Arctic fox dependents.

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Eli Geffen, Love Dalén, Anders Angerbjörn, Carles Vila, Pall Hersteinsson, Eva Fuglei, Paula White, Michael Goltsman, Christian Kapel and Robert K. Wayne have been studying for many years the behavior, ecology, physiology, parasitology and genetics of the Artic fox at various parts of the globe. The collaborative effort and experience of all these people enabled optimal sampling coverage of Artic fox populations and the putting together of this paper.
