Genetic differentiation of populations of Greenlandic Arctic fox

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Most microsatellites are very polymorphic. This makes them powerful markers for observing genetic differentiation between closely related populations. The population structure of the Greenlandic Arctic fox (*Alopex lagopus*) was studied genetically by analysing six polymorphic microsatellite loci of 75 foxes from four populations in different parts of Greenland. Genotypes were determined at the six loci for most of the individuals. Population differentiation was quantified in three different ways both within the total population and pairwise between all populations. The tests were Fisher's exact test, Rho estimates and Fst estimates, all of which supported a highly significant subdivision of the total population, and they showed significant differentiation in allele frequencies between all pairs of localities. It is concluded that the known long-distance migration of the Greenlandic Arctic fox has not resulted in complete genetic mixing of the populations. Fisher's exact test was also used to estimate levels of genetic differentiation between the two colour morphs: white and blue. No difference was found between allele frequencies of the two color morphs in any of the locations, and it was concluded that the white and blue morphs of the Greenlandic Arctic fox share the same habitat, at least during the mating season.

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The Arctic fox (*Alopex lagopus*) is a small mammal weighing 2-5 kg, with circumpolar distribution. It has a home range ranging from 4 - 100 km^2 (Frafjord & Prestrud 1992; Anthony 1997) but several ear-tagging and radio-tracking studies have revealed that the Arctic fox is capable of undertaking long-distance movements (Eberhardt & Hanson 1978; Eberhardt et al. 1983; Frafjord & Prestrud 1992; Russian papers referenced by Fay & Rausch 1992). The longest movement recorded was 2300 km (Eberhardt, unpubl. data referenced by Garrott & Eberhardt 1987).

The Arctic fox inhabits the entire coastal zone of Greenland and is found in two interfertile colour morphs: a blue and a white. The colour of a fox is determined by the alleles at one locus, where "the blue allele" is dominant over the recessive white one (Adalsteinsson et al. 1987). Based on observations and hunting statistics, Hersteinsson (1989) found a positive correlation between the length of time the ground was snow-covered and the proportion of white foxes in Icelandic populations. This situation had previously been noticed in Greenland by Braestrup (1941), who described the blue and the white fox as two subspecies: the white one subsisting on lemmings

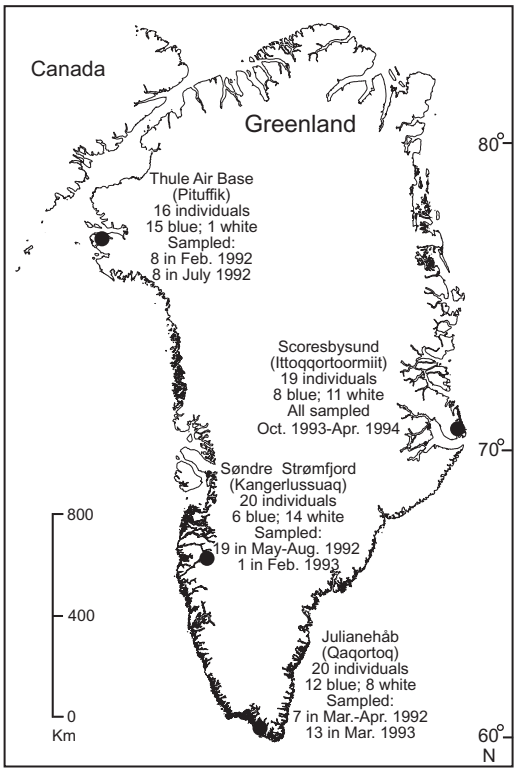

Fig. 1. Map of Greenland showing sites, sizes and dates of Arctic fox sampling.

and living in the mostly snow-covered inland and the blue one depending to a large extent on products of the sea and living near the coast. Braestrup´s studies, based on fur trade statistics, were continued by Vibe (1967), who included field studies and climate statistics. He stated that unstable climatic periods during the last centuries, with the climate favouring the blue morph in some periods and the white one in other periods, have resulted in the "lively bastardization and creation of mixed populations which prevail nearly everywhere in coastal regions today". Braestrup (1941) found large fluctuations in the proportion of white foxes in the hunting bags from west Greenland and attributed this to a massive influx of white foxes to west Greenland from Canada and east Greenland in years with low lemming populations. In a morphological study of the foxes of Greenland, the blue foxes from different districts showed significant differences in (metric) bone measures while the white ones did not (Berg 1993). Berg suggested that the blue colour morph is more stationary than the white, due to the food supply in coastal areas being less variable between years than in inland habitats. Russian observations (reviewed by Wrigley & Hatch 1976) showed that foxes migrate from November to January and return in February and March to breed. If this is the case, samples collected in winter could include migrants and could therefore be misleading.

The aim of the present study was to delineate the genetic relationships of four Arctic fox populations and the two colour morphs of Arctic fox in Greenland. The following questions were put forward: 1) Are geographically distant populations significantly differentiated? 2) Are the two colour morphs genetically differentiated? 3) Are there genetic differences between foxes sampled at the same locality during different seasons?

To answer these questions we used six polymorphic microsatellite loci. Due to the generally high mutation rate (10^{-2} - 10^{-5} mut./ locus/ generation) (Weber & Wong 1993), microsatellites tend to be very polymorphic, which make them powerful markers for observing the differentiation between closely related populations. However, their mutational modalities also result in a risk of homoplazy (Valdes et al. 1993; Weber & Wong 1993) and some microsatellites might have alleles that are not detectable (null alleles) (Callen et al. 1993). Also, microsatellite evolution is not yet properly understood and it could be problematic to consider them as strictly neutral markers: constraints on allele size have been implicated (Garza et al. 1995) and numerous indications of microsatellites involved in gene expression and function have been found (reviewed by Kashi et al. 1997). Furthermore, the possible effect of a selected locus on a closely linked microsatellite has been discussed (Slatkin 1995a). Several mutation models have been suggested to work for microsatellites. Two are most used: the infinite allele model (IAM) (Estoup et al. 1995) and the stepwise mutation model (SMM) (Shriver et al. 1993; Valdes et al. 1993).

Materials and methods

Samples

Seventy-five Greenlandic Arctic foxes from four widely spaced localities are included in this study; see Fig. 1 for sample details. All the foxes were

caught in traps, killed and kept frozen at –20 °C in a period of two to three months prior to sampling of muscle tissue. DNA was extracted using Proteinase K and phenol/chloroform-solutions (e.g. Sambrook et al. 1989). The foxes analysed here have been included in studies on parasitology, diet and population composition (Kapel et al. 1996; Kapel & Nansen 1996; Kapel 1999).

Analyses of microsatellite loci

Several microsatellite loci originally described in studies of wolf and dog were screened. The primer sets designed for dog microsatellite loci turned out to give the best results, and six of them were chosen and optimized for the Arctic fox: cph3, cph6, cph9, cph15, cph16, and cph18 (Fredholm & Winterø 1995). All six microsatellite loci used are dinucleotide repeats, and all but cph3 have perfect repeats (see Fredholm & Winterø 1995). The final PCR conditions were as follows (Table 1): denaturation at 94 °C for 2 min., x cycles of denaturation at 94 °C for y sec., annealing at z °C for y sec. and extention at 72 °C for y sec., x, y and z are specified for each locus in Table 1. The amplification quality in different PCR machines differed from locus to locus. The final choice of machines is also shown in Table 1.

DNA was amplified in a 10 µl reaction volume (10 mM Tris-HCL, 1.5 mM $MgCl_2$, 50 mM KCl, pH 8.3, 200 µM of each dNTP, 200 pM of each primer, and 0.025 units of Boehringer-Mannheim Taq DNA polymerase). The products were run on 4.24 % acrylamidgels on an ABI 377 sequencing machine (Perkin Elmer) using dye-labeled primers (one primer in each primer set) and ROX 500 (Perkin Elmer) as internal standard. Genotypes were determined at the six loci for most of the individuals (see Table 2).

Table 1. PCR conditions.

Locus	Annealing temp. (z)	Sec. at each step (y)	Cycles (x)	PCR machine
cph3	50 °C	15[a]	37	Unitec2042
cph6	49 °C	50[a]	37	Robocycler Gradient
cph9	55 °C	15	35[c]	Unitec2042
cph15	54 °C	15	34	Hybaid
cph16	54 °C	15[a]	53	Unitec2042
cph18	66 °C	30[a,b]	31	Robocycler Gradient

[a] Last cycle with extension time: 5 min.
[b] All but last cycle with extension time: 40 sec.
[c] Touch down PCR: two cycles with annealing temp. at each degree from 45 °C to 54 °C, followed by 25 cycles at annealing temp. 55 °C.

Statistical tests

Deviations from Hardy-Weinberg equilibrium (HWE) was tested using the "exact HW test"

Table 2. Observed allele frequency distributions by locus and population. Private alleles are shown in italics. Boldface indicates the most frequent allele/locus/population. N indicates number of chromosomes.

Locus/ allele	Thule Air Base	Scores-bysund	Søndre Strømfjord	Juliane-håb
cph3/151			*0.026*	
153	**0.536**	0.132	**0.316**	
155	0.107	0.026	0.079	
157	0.036	0.184	0.211	0.100
161	0.250	**0.474**	0.105	**0.350**
163	0.071	0.053	0.132	**0.350**
165		0.079	0.026	0.175
167			*0.053*	
169			0.026	0.025
171		*0.026*		
173			*0.026*	
179		*0.026*		
n	28	38	38	40
cph6/109		0.222	0.158	**0.400**
119	**0.667**	0.028		
121			0.053	0.250
123		0.028	0.105	0.125
125		**0.278**	**0.316**	0.075
127		0.222	0.211	0.150
129		0.222	0.158	
133	*0.333*			
n	30	36	38	40
cph9/152	0.344	0.028	**0.368**	**0.675**
154	0.031	0.250	0.237	0.075
156	**0.438**	**0.639**	0.342	0.175
158	0.188		0.053	0.050
162				*0.025*
164		*0.083*		
n	32	36	38	40
cph15/153	0.167	0.132	0.050	0.050
155	0.033	0.079	0.075	0.200
157	**0.667**	**0.658**	**0.675**	**0.550**
159		0.053	0.125	0.150
161	0.133	0.026	0.075	0.050
163		*0.026*		
165		*0.026*		
n	30	38	40	40
cph16/155				*0.075*
159	0.156	0.105	0.175	0.025
161	**0.656**	**0.632**	0.250	0.100
163	0.188	0.263	**0.525**	**0.725**
165			0.025	0.075
167			*0.025*	
n	32	38	40	40
cph18/253		0.028		0.184
255			0.059	0.053
257	0.125	0.056	**0.441**	**0.289**
259		0.167	0.029	0.026
263	0.031	0.028	0.206	0.053
265	**0.781**	0.083	0.118	0.026
267	0.063	0.056	0.088	0.105
269		**0.500**		0.158
271		0.083	0.059	0.105
n	32	36	34	38

in GENEPOP (Raymond & Rousset 1995). The sequential Bonferroni technique (Rice 1989) was applied to test significant deviation from HWE at a "table-wide" α-level of 0.05.

Test for genotypic disequilibrium was performed in GENEPOP using Fisher´s exact test (Raymond & Rousset 1995). Fisher´s exact test was also used to estimate levels of genic differentiation between colour morphs and sampling seasons within locations. The sequential Bonferroni technique (Rice 1989) was applied to test for significance at the "table-wide" α-level of 0.05.

Population differentiation was quantified in three different ways both within the total population and between all populations pairwise: 1) by Fisher´s exact test in GENEPOP; 2) by estimating Rho using Goodman`s (1997) analogue to Slatkin´s Rst (calculated in Rst-calc 2.2; Rst estimates are based on the SMM); and 3) by estimating the Weir & Cockerham (1984) analogues to Wright´s Fst calculated in Arlequin. Fst estimates are based on the IAM. All three methods were applied to the total population and to each pair of populations. GENEPOP and Rst-calc 2.2 provided probability values and Rho estimates, respectively, for individual loci and overall loci, while Arlequin only provided Fst estimates of overall loci. Furthermore, Rst-calc 2.2 provided bootstrap values as well as significance values for all estimates, making it possible to see if two Rho values differ significantly from each other.

Estimates of the number of migrants per generation (Nm) between all pairwise populations was computed from the approximations: Nm= 1/4 (1/Fst -1) (Slatkin 1995b) and Nm=1/4 (1/Rho -1) (pers. comm., Bo Simonsen). Nm was also estimated using Slatkin´s rare alleles method which makes use of the fact that the logarithm of Nm is approximately linearly related to the logarithm of the average frequency of private alleles in a sample of alleles from the population (Slatkin 1985). This method has shown to be relatively insensitive to changes in other parameters than Nm and the number of individuals sampled per population (Slatkin 1985).

Results

Genetic variation and deviation from Hardy-Weinberg equilibrium

The six loci—cph3, cph6, cph9, cph15, cph16 and cph18—were polymorphic with 12, 8, 6, 7, 6 and 9 alleles, respectively (Table 2). The average number of alleles per locality ranged from 4 in cph16 to 7 in cph3 and cph18, and average expected heterozygosity values for all loci ranged from 0.54 in Thule Air Base to 0.73 in Søndre Strømfjord (see Table 3).

There was highly significant deviation from HWE in the total population ($p \leq 0.002$). All loci showed deficiency of heterozygotes (Table 3). No significant deviation from the HWE at the "table-wide" 0.05 level was found in any of the four separate populations (Table 3). Looking at each locus across localities, two loci , one in each locality, showed significant deviation from HWE due to

Table 3. Expected (He) and observed (Ho) heterozygocity and probability values for "the exact HW test" calculated for each locus in every population. S and NS indicate significance and nonsignificance, respectively, at the "table-wide" 0.05 level. Nonsignificant p-values across populations are shown in italics.

	Ho	He	P(HWE)
Thule Air Base			
cph3	0.57	0.66	0.077 NS
cph6	0.27	0.46	0.232 NS
cph9	0.69	0.68	0.394 NS
cph15	0.33	0.53	*0.012* NS
cph16	0.50	0.53	0.827 NS
cph18	0.44	0.38	1.000 NS
Scoresbysund			
cph3	0.68	0.73	0.169 NS
cph6	0.72	0.80	0.927 NS
cph9	0.61	0.54	0.863 NS
cph15	0.47	0.55	0.308 NS
cph16	0.53	0.53	0.831 NS
cph18	0.61	0.72	0.108 NS
Søndre Strømfjord			
cph3	1.00	0.84	0.035 NS
cph6	0.89	0.86	0.251 NS
cph9	0.68	0.71	0.229 NS
cph15	0.50	0.53	0.377 NS
cph16	0.60	0.65	0.184 NS
cph18	0.59	0.76	*0.011* NS
Julianehåb			
cph3	0.65	0.73	0.196 NS
cph6	0.65	0.75	0.899 NS
cph9	0.35	0.52	0.079 NS
cph15	0.50	0.65	0.041 NS
cph16	0.35	0.46	0.070 NS
cph18	0.79	0.85	0.395 NS
Total pop.			
cph3	0.74	0.81	*0.001* S
cph6	0.64	0.86	*0.000* S
cph9	0.58	0.69	*0.001* S
cph15	0.46	0.57	*0.002* S
cph16	0.49	0.64	*0.001* S
cph18	0.61	0.85	*0.000* S

heterozygote deficiency: cph15 in Thule Air Base and cph18 in Søndre Strømfjord (shown in italics in Table 3). All other loci across localities showed no deviation from HWE.

Linkage disequilibrium was found in no pair of loci in the global test across all localities (probability values ranging from 0.07 to 0.96; data not shown). Within the populations, the same test demonstrated two cases of linkage disequilibrium (data not shown): the locus pair cph9–cph18 showed significant linkage in Thule Air Base ($p = 0.007$) and the pair cph9–cph16 showed linkage in Scoresbysund ($p = 0.03$).

Dividing populations according to colour and time of sampling

To test for differentiation in allele frequencies between the two colour morphs we divided each sample according to fur colour. Thule Air Base was omitted from this test as white foxes there were represented by only one animal. The remaining three localities contained sub-samples of at least six individuals each (Fig. 1). No significant differentiation was found between allele frequencies of the two colour morphs in any of the localities (Table 4). By using the same test on the three localities pooled, cph18 showed a significant difference in allele frequencies of blue and white foxes, while the other five loci showed nonsignificant results (Table 4). To compare population differentiation of the white foxes with the blue foxes, Rho estimates were calculated for each colour. These Rho estimates were calculated for all pairwise localities in which the colours were sampled, and for the total population (Table 5). The results for the blue foxes showed highly significant ($p < 0.001$) subdivision of the total sample; there was significant population differentiation ($p < 0.05$) in all pairwise comparisons except between Thule Air Base and Scoresbysund and between Søndre Strømfjord and Julianehåb. White foxes also showed significant subdivision of the total sample ($p < 0.01$) and in all pairwise population comparisons ($p < 0.05$). None of the Rho estimates calculated for blue and white foxes were significantly different from each other or from the Rho estimates of the populations of both colours (overlap in 95 % confidence intervals, see Table 5).

In the test for difference in allele frequencies between the seasons, foxes at Thule Air Base showed a significant difference in allele frequency between two sampling occasions (winter and summer) in two out of six loci (Table 6). No significant difference was found between the allele frequencies of two sub-samples from Scoresbysund (winter and spring), nor beween two subsamples from Julianehåb (two consecutive spring seasons) (Table 6).

Table 4. Results (p-values) from Fisher´s exact test for differentiation between blue and white foxes. S and NS indicate significance and nonsignificance, respectively, at the "tablewide" 0.05 level.

Locus	Scoresbysund	Søndre Strømfjord	Julianehåb	Scoresbysund + S. Strømfjord + Julianehåb
cph3	0.5390 NS	0.5928 NS	1.0000 NS	0.502 NS
cph6	0.7613 NS	0.1179 NS	0.1356 NS	0.082 NS
cph9	0.2312 NS	0.1627 NS	0.0240 NS	0.160 NS
cph15	0.2731 NS	0.4658 NS	0.0641 NS	0.307 NS
cph16	1.0000 NS	0.0103 NS	0.4752 NS	0.294 NS
cph18	0.7009 NS	0.0195 NS	0.0092 NS	0.007 S

Population structure

The results for all loci from the three different tests for population subdivision are presented in Table 7.

The first of the results to be noted is that Fisher´s exact test showed highly significant subdivision ($p < 0.00001$) of the total population in all but one locus (cph15).

In five of the six pairs of localities, significant differentiation in allele frequencies was supported

Table 5. Population differentiation with populations separated according to fur colour. Significance levels: * = $p < 0.05$; ** = $p < 0.01$; *** = $p < 0.001$. NS indicates a nonsignificant result. 95 % confidence interval is provided in parentheses.

Rho est., all loci	Thule Air Base –Scoresbysund	Thule Air Base –S. Strømfjord	Thule Air Base –Julianehåb	Scoresbysund –S. Strømfjord	Scoresbyund –Julianehåb	S. Strømfjord –Julianehåb	Total pop.
Blue foxes	0.065 NS (0.020; 0.222)	0.152* (0.152; 0.374)	0.222*** (0.174; 0.364)	0.224** (0.152; 0.430)	0.191** (0.121; 0.352)	0.050 N (0.007; 0.303)	0.159*** (0.141; 0.278)
White foxes				0.080* (0.022; 0.246)	0.180** (0.101; 0.390)	0.144** (0.080; 0.367)	0.128** (0.086; 0.290)

by from four to all of six loci. It was only between Thule Air Base and Søndre Strømfjord that half of the loci showed no significant differentiation in allele frequencies. The second of the results to be noted is that Rho estimates showed a highly significant population subdivision within the total population ($p < 0.001$) and a significant population differentiation between all pairwise combinations of populations ($p < 0.05$). Furthermore, bootstrap results showed that none of the Rho results from the pairwise locality studies were significantly different from another (overlap between all the 95 % confidence intervals), although they ranged from 0.065 (Thule Air Base–Søndre Strømfjord) to 0.226 (Thule Air Base–Julianehåb).

Third, Fst estimates also showed a highly significant population subdivision ($p < 0.001$) within the total population. Furthermore, this estimate showed highly significant population differentiation ($p < 0.001$) between all combinations of populations.

To find possibly deviant loci we looked at the population differentiation results locus-wise and found the following facts notable: 1) When correcting the significance values locus-wise (horizontally in Table 7) according to the sequential Bonferroni technique (results not shown), cph15 showed nonsignificant population differentiation in all pairwise population comparisons and nonsignificant subdivision of the total population. Cph9 showed nonsignificant result in two of the six pairwise population comparisons, cph3,6 and 16 in only one, and cph18 showed no nonsignificant probability values. 2) There was a large span in the Rho estimates of individual loci within all pairwise population comparisons, ranging from a negative value (around -0.03) to 0.246 (Table 7).

Table 6. Results (probability values) from Fisher´s exact test for difference in allele frequency between foxes sampled indifferent seasons. S and NS indicate significance and nonsignificance, respectively, at the "table-wide" 0.05 level.

Locus	Thule Air Base (winter 1992 –summer 1992)	Scoresbysund (winter 1993 –spring 1994)	Julianehåb (spring 1992 –spring 1993)
cph3	0.0015 S	0.8920 NS	0.0828 NS
cph6	0.7060 NS	0.4773 NS	0.4178 NS
cph9	0.0746 NS	0.4729 NS	0.0563 NS
cph15	0.4734 NS	0.3427 NS	0.3745 NS
cph16	0.1435 NS	0.8863 NS	0.8987 NS
cph18	0.0060 S	0.6906 NS	0.5028 NS

Migration

The estimated number across the total population of migrating individuals calculated from the Rho and the Fst estimates was 1.71 and 1.32 individuals/generation respectively (Table 8) and 2.05 individuals/generation when estimated by using private alleles according to Slatkin (1985) (data not shown). In the pairwise population com-

Table 7. Population differentiation. Probability values from Fisher's exact test. S and NS refer to significance and nonsignificance, respectively, at the "table-wide" 0.05 level. Rho estimates and Fst estimates are shown. Significance levels: * = $p < 0.05$; ** = $p < 0.01$; *** = $p < 0.001$. 95% confidence interval is provided in parentheses for Rho estimates over all loci.

Estimate/ loci	Thule Air Base –Scoresbysund	Thule Air Base. –S. Strømfjord	Thule Air Base –Julianehåb	Scoresbysund –S. Strømfjord	Scoresbysund –Julianehåb	S. Strømfjord –Julianehåb	Total pop.
G. diff.							
cph3	0.0013 S	0.1314 NS	0.0000 S	0.0020 S	0.0019 S	0.0000 S	0.0000 S
cph6	0.0000 S	0.0000 S	0.0000 S	0.5661 NS	0.0000 S	0.0002 S	0.0000 S
cph9	0.0000 S	0.0421 NS	0.0082 S	0.0001 S	0.0000 S	0.0256 NS	0.0000 S
cph15	0.4252 NS	0.1323 NS	0.0095 S	0.4848 NS	0.2088 NS	0.5428 NS	0.1108 NS
cph16	0.6412 NS	0.0040 S	0.0000 S	0.0058 S	0.0000 S	0.0099 S	0.0000 S
cph18	0.0000 S	0.0000 S	0.0000 S	0.0000 S	0.0003 S	0.0054 S	0.0000 S
Rho							
cph3	0.283	0.059	0.633	0.052	0.092	0.306	
cph6	-0.019	-0.027	0.250	-0.025	0.157	0.219	
cph9	0.067	0.036	0.157	0.236	0.349	0.035	
cph15	-0.030	-0.010	-0.030	-0.015	-0.027	-0.010	
cph16	-0.006	0.123	0.151	0.057	0.092	-0.015	
cph18	0.058	0.246	0.115	0.332	0.215	-0.026	
All loci	0.070** (0.018; 0.180)	0.065* (0.028; 0.182)	0.226*** (0.178; 0.336)	0.117*** (0.057; 0.232)	0.165*** (0.093; 0.285)	0.089** (0.053; 0.191)	0.128*** (0.107; 0.206)
Fst	0.198***	0.175***	0.262***	0.096***	0.169***	0.070***	0.159***

parison the Nm values calculated from the Rho estimates ranged from 0.86 between Thule Air Base and Julianehåb to 3.61 between Thule Air Base and Søndre Strømfjord, but, as with the Rho estimates, not one of the values was significantly different from another according to the bootstrap results. The Nm values calculated from the Fst estimates in the pairwise population comparisons showed about the same range: from 0.70 (Thule Air Base–Julianehåb) to 3.34 (Søndre Strømfjord–Julianehåb).

Discussion

Four randomly mating populations

The heterozygote deficiency found in the total Greenlandic sample can probably be ascribed to a Wahlund effect created when pooling the four populations. These HWE results give a preliminary indication that the population is subdivided in spite of the Arctic fox's capacity for long-distance movements. Furthermore, the HWE results showed that each of the four localities can be seen as a randomly mating population (no deviation from HWE). The fact that two loci (cph15 and cph18), one each in two different populations, showed a significant deviation from HWE, looking across all populations, cannot be explained by null alleles: if the two loci had null alleles we would expect it to affect the other populations as well, which was not the case. It is, of course, possible that non-detectable alleles causing heterozygote deficiency of cph15 and cph18 only were present in Thule Air Base and in Søndre Strømfjord, respectively, but since all but one private allele in the total data set had very small allele frequencies ($p \leq 0.05$; Table 2), this appears unlikely. We cannot explain these significant results since explanations such as assortative mating or pooling of samples (resulting in the Wahlund effect) would be expected to affect the other loci of the populations as well and that is not the case.

The two colour morphs

There seems to be a positive correlation between the period of time an area is snow-covered and the proportion of white foxes in this area (Braestrup 1941; Vibe 1967; Hersteinsson 1989). If this is due to a sharply defined difference in habitat, interbreeding between the morphs would only exist in a small hybrid zone, and we would expect to find a difference in allele frequencies between the morphs and certainly not random mating between them.

Kapel et al. (1996) found no significant difference between the prevalence of *Trichinella nativa* infections in the blue and the white foxes, respectively, which were also used for the study reported here. *T. Nativa* is an extraintestinal nematode which is most prevalent in foxes in areas where polar bears are hunted and sledge dogs are common (Kapel 1995). The nematode is thought to be transmitted to the Arctic fox through scavenging on carcasses of these animals or through cannibalism. The fact that Kapel found no difference in the prevalence of this parasite according to colour of the foxes could indicate that the two morphs share the same habitat and diet. The results of this study indicate random mating within the three populations of mixed colours and no allele frequency difference between the two colour morphs. It is important to note that the statistical power may have been small due to small sample sizes. However, our data suggest that the two colour morphs of the Greenlandic Arctic fox share the same habitat, at least during the mating season.

The investigation of differentiation within the blue and the white fox groups (Table 5) yields no indication that the blue fox is more stationary than the white one. On the contrary, Rho estimates failed to show significant difference between the

Table 8. Migration rate. Nm values correspond to the Rho and Fst estimates shown in Table 6. 95 % confidence interval is provided in parentheses for the Nm corresponding to Rho estimates.

Estimate/ loci	Thule Air Base –Scoresbysund	Thule Air Base –S. Strømfjord	Thule Air Base –Julianehåb	Scoresbysund –S. Strømfjord	Scoresbysund –Julianehåb	S. Strømfjord –Julianehåb	Total pop.
Nm (Rho)	3.34	3.61	0.86	1.89	1.26	2.54	1.71
	(1.07; 12.0)	(1.11; 8.58)	(0.49; 1.15)	(0.82; 3.91)	(0.61; 2.42)	(1.05; 4.41)	(0.96; 2.07)
Nm (Fst)	1.01	1.18	0.70	2.35	1.23	3.34	1.32

blue foxes of Søndre Strømfjord and Julianehåb while significant differentiation ($p < 0.01$) was found between the white foxes from the same localities. These results indicate that the difference Berg (1993) found with respect to morphological differentiation of Greenlandic blue and white foxes was a result of ecological rather than genetic differences, as Berg also discusses.

The different sampling seasons

Two of the four populations (Thule Air Base and Scoresbysund) in this study were sampled in both winter and spring/summer seasons (Fig. 1). Theories have been proposed that the foxes migrate in November-January and return to their "home" in February–March to breed—the "homing theory" (Russian studies reviewed by Wrigley & Hatch 1976). If this is the case, sampling in winter will lead to sampling of migrants that will eventually return to their own locality and therefore have no genetic impact on the locality in which they were found. Winter populations will be a mixture of mating populations and we would expect to find a difference in allele frequencies between a winter and a summer sample from the same locality. We tried to test this theory by testing for genic differentiation between the sub-samples of Thule Air Base and Scoresbysund. However, in testing the homing theory this way, we have a problem in the transition between winter and mating populations. Scoresbysund was artificially divided into two sub-samples: one on each side of 1 March 1994. The results of Thule Air Base neither clearly supported nor disproved the theory and we have to conclude that our data set cannot be used to test the homing theory. Samples from more populations, collected in both winter and summer, are needed.

Population structure

Distinct geographically differences in the composition of the parasitic helminth fauna of Arctic fox in Greenland have been found (Kapel et al. 1996; Kapel & Nansen 1996). The diversity of the surrounding fauna and thereby the food items available for the foxes seemed important influences on the diversity of the helminth species (Kapel & Nansen 1996). These results indicate limited mixing of the studied populations. The present study supports this indication. All three approaches to analysing population differentiation (Fisher´s exact test, Fst estimates and Rho estimates) showed that the total sample of Greenlandic Arctic fox was significantly differentiated into subpopulations. The Rho and the Fst estimates across all loci also showed significant population differentiation between all pairwise populations included in the study.

Population differentiation results for individual loci are provided by Fisher´s exact test and as Rho estimates. In both tests cph15 was the only locus consistently showing no difference between any two populations, and it even showed no significant subdivision of the total population in Fisher´s exact test (Table 7). On this basis we suggest that cph15 is not a neutral locus. Slatkin (1995a) showed that the variance of microsatellite allele size can be strongly influenced by selection at closely linked loci. Cph15 could be linked to a locus under selection or it could be involved in gene expression, resulting in a reduction in the variance of allele size of this microsatellite in a way that makes population subdivision less detectable with this locus.

Migration

The three different methods we have used to estimate average Nm between the populations of this study gave very consistent results around one to two migrating foxes between each locality every generation. The distances between the localities of this study range from 825 km between Søndre Strømfjord and Julianehåb to 1950 km between Thule Air Base and Julianehåb. From our data we cannot discern if, for instance, one fox per generation travels the distance between Thule Air Base and Julianehåb or if the estimate is a result of a larger gene flow between all adjacent populations along the distance between Thule Air Base and Julianehåb. In this regard it would be interesting to get estimates of differentiation and migration between populations separated by shorter distances (including adjacent populations) and to extend the study to involve more loci and/or individuals to increase statistical power.

Conclusion

We have found evidence of extensive genetic differentiation between four Arctic fox localities,

demonstrating that the ability of Arctic foxes to undertake long-distance movements has not resulted in a panmictic Greenlandic population. The foxes examined in this study do not show genetic differentiation between the two colour morphs within each locality. This suggests that the two colour morphs of the Greenlandic Arctic fox can be considered as members of the same populations. A test for population differentiation within each of the two colour morphs indicated that they are equally stationary.

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