Ancient DNA analyses exclude humans as the driving force behind late Pleistocene musk ox (Ovibos moschatus) population dynamics

Paula F. Campos1, Eske Willerslev2, Andrei Shep1, Ludovic Orlando2, Erik Axelsons3, Alexei Tikhonov2, Kim Aaris-Sørensen3, Alex D. Greenwood2,6, Ralf-Dietrich Kahilke1, Pavel Kosintsev6, Tatiana Krakmalnaya4, Tatjana Kuznetsova1, Philippe Lemey1, Ross MacPhee3, Christopher A. Norris1, Kieran Shepherd3, Marc A. Suchard3, Grant D. Zazula4, Beth Shapiro3, and M. Thomas P. Gilbert1,3

1Centre for GeoGenetics, Natural History Museum of Denmark, University of Copenhagen, DK 1350 Copenhagen, Denmark; 2Severtsov Institute of Ecology and Evolution, Russian Academy of Sciences, Moscow 119071, Russia; 3Paleogenetics and Molecular Evolution, Institut de Génomique Fonctionnelle de Lyon, Université de Lyon, Université Lyon 1, Centre National de la Recherche Scientifique, Institut National de la Recherche Agronomique, École Normale Supérieure de Lyon, 69364 Lyon Cedex 07, France; 4Zoological Institute, Russian Academy of Sciences, St. Petersburg 199034, Russia; 5Department of Biological Sciences, Old Dominion University, Norfolk, VA 23529; 6Senckenberg Research Institutes and Natural History Museums, Research Station of Quaternary Palaeoontology, D-99423 Weimar, Germany; 7Institute of Plant and Animal Ecology, Urals Branch of the Russian Academy of Sciences, Ekaterinburg 620144, Russia; 8National Museum of Natural History, National Academy of Sciences of Ukraine, Kiev 01030, Ukraine; 9Department of Paleontology, Faculty of Geology, Lomonosov Moscow State University, Moscow 119991, Russia; 10Department of Microbiology and Immunology, Rega Institute, Katholieke Universiteit Leuven, B-3000 Leuven, Belgium; 11Division of Vertebrate Zoology, American Museum of Natural History, New York, NY 10024; 12Division of Vertebrate Paleontology, Peabody Museum of Natural History, New Haven, CT 06520; 13Collections, Canadian Museum of Nature, Ottawa, ON K1P 6P4, Canada; 14Department of Biomatics, Division of Biostatistics, and Department of Human Genetics, University of California, Los Angeles, CA 90095; 15Yukon Palaeoontology Program, Department of Tourism and Culture, Yukon Government, Whitehorse, YT Y1A 2C6, Canada; and 16Department of Biology, Pennsylvania State University, University Park, PA 16801

Edited by David J. Meltzer, Southern Methodist University, Dallas, TX, and approved February 1, 2010 (received for review June 27, 2009)

The causes of the late Pleistocene megafaunal extinctions are poorly understood. Different lines of evidence point to climate change, the arrival of humans, or a combination of these events as the trigger. Although many species went extinct, others, such as caribou and bison, survived to the present. The musk ox has an intermediate story: relatively abundant during the Pleistocene, it is now restricted to Greenland and the Arctic Archipelago. In this study, we use ancient DNA sequences, temporally unbiased summary statistics, and Bayesian analytical techniques to test both climatic warming and human impact (1) or climate change associated with the last glacial cycle (2). The relative contribution of the two remains a key evolutionary debate (cf. 3, 4). For example, it is well known that global climate fluctuated significantly throughout the Pleistocene (5–7); the Eurasian fossil record indicates that late Pleistocene climatic changes reduced the range of several species, and the last glacial-interglacial transition appears to correlate with major extinction events. However, none of the previous Pleistocene glacial cycles appear to have caused widespread extinction events (3, 8), and the demise of the Beringian megafauna also appears to coincide neatly with the arrival and spread of modern human populations (9, 10). Similar evidence of a human-driven effect can be seen in central North America, where the arrival of Clovis-style hunters, extinction of megafauna, and marked climate change all cluster closely together (11, 12).

Ancient DNA (aDNA) analyses of temporally and geographically distributed megafaunal remains offer a means to address the relative roles of humans versus the climate in the Beringian megafaunal extinctions. The capacity of this technique to detect changes in genetic diversity within populations over geologically significant time scales provides the means to examine past population variation and to explore directly how populations respond to climate and environmental changes (13). Geographic and temporal discontinuity in the genetic data can be compared to past climate reconstructions and specific ecological events, such as advancing ice sheets, a volcanic eruption, or human colonization. Previous large-scale aDNA studies of this kind have focused on herbivores, such as bison and horses that survived to the present in relatively large numbers and are thus relatively

climate change | human impact | quaternary | megafauna extinctions

T

the late Pleistocene saw significant changes in the geographic distribution and composition of the Beringian megafauna. Although many iconic Beringian herbivores, such as mammoths (Mammuthus primigenius) and woolly rhinoceroses (Coelodonta antiquitatis) became extinct, others, such as horses (Equus caballus), saiga (Saiga tatarica), caribou (Rangifer tarandus), and bison (Bison bison) survived into the present. The reasons for these markedly different survival patterns have been widely debated, with the extinctions predominantly attributed to either human impact (1) or climate change associated with the last glacial cycle (2). The relative contribution of the two remains a key evolutionary debate (cf. 3, 4). For example, it is well known that global climate fluctuated significantly throughout the Pleistocene (5–7); the Eurasian fossil record indicates that late Pleistocene climatic changes reduced the range of several species, and the last glacial-interglacial transition appears to correlate with major extinction events. However, none of the previous Pleistocene glacial cycles appear to have caused widespread extinction events (3, 8), and the demise of the Beringian megafauna also appears to coincide neatly with the arrival and spread of modern human populations (9, 10). Similar evidence of a human-driven effect can be seen in central North America, where the arrival of Clovis-style hunters, extinction of megafauna, and marked climate change all cluster closely together (11, 12).

Ancient DNA (aDNA) analyses of temporally and geographically distributed megafaunal remains offer a means to address the relative roles of humans versus the climate in the Beringian megafaunal extinctions. The capacity of this technique to detect changes in genetic diversity within populations over geologically significant time scales provides the means to examine past population variation and to explore directly how populations respond to climate and environmental changes (13). Geographic and temporal discontinuity in the genetic data can be compared to past climate reconstructions and specific ecological events, such as advancing ice sheets, a volcanic eruption, or human colonization. Previous large-scale aDNA studies of this kind have focused on herbivores, such as bison and horses that survived to the present in relatively large numbers and are thus relatively

climate change | human impact | quaternary | megafauna extinctions

T

the late Pleistocene saw significant changes in the geographic distribution and composition of the Beringian megafauna. Although many iconic Beringian herbivores, such as mammoths (Mammuthus primigenius) and woolly rhinoceroses (Coelodonta antiquitatis) became extinct, others, such as horses (Equus caballus), saiga (Saiga tatarica), caribou (Rangifer tarandus), and bison (Bison bison) survived into the present. The reasons for these markedly different survival patterns have been widely debated, with the extinctions predominantly attributed to either human impact (1) or climate change associated with the last glacial cycle (2). The relative contribution of the two remains a key evolutionary debate (cf. 3, 4). For example, it is well known that global climate fluctuated significantly throughout the Pleistocene (5–7); the Eurasian fossil record indicates that late Pleistocene climatic changes reduced the range of several species, and the last glacial-interglacial transition appears to correlate with major extinction events. However, none of the previous Pleistocene glacial cycles appear to have caused widespread extinction events (3, 8), and the demise of the Beringian megafauna also appears to coincide neatly with the arrival and spread of modern human populations (9, 10). Similar evidence of a human-driven effect can be seen in central North America, where the arrival of Clovis-style hunters, extinction of megafauna, and marked climate change all cluster closely together (11, 12).

Ancient DNA (aDNA) analyses of temporally and geographically distributed megafaunal remains offer a means to address the relative roles of humans versus the climate in the Beringian megafaunal extinctions. The capacity of this technique to detect changes in genetic diversity within populations over geologically significant time scales provides the means to examine past population variation and to explore directly how populations respond to climate and environmental changes (13). Geographic and temporal discontinuity in the genetic data can be compared to past climate reconstructions and specific ecological events, such as advancing ice sheets, a volcanic eruption, or human colonization. Previous large-scale aDNA studies of this kind have focused on herbivores, such as bison and horses that survived to the present in relatively large numbers and are thus relatively

climate change | human impact | quaternary | megafauna extinctions

T

the late Pleistocene saw significant changes in the geographic distribution and composition of the Beringian megafauna. Although many iconic Beringian herbivores, such as mammoths (Mammuthus primigenius) and woolly rhinoceroses (Coelodonta antiquitatis) became extinct, others, such as horses (Equus caballus), saiga (Saiga tatarica), caribou (Rangifer tarandus), and bison (Bison bison) survived into the present. The reasons for these markedly different survival patterns have been widely debated, with the extinctions predominantly attributed to either human impact (1) or climate change associated with the last glacial cycle (2). The relative contribution of the two remains a key evolutionary debate (cf. 3, 4). For example, it is well known that global climate fluctuated significantly throughout the Pleistocene (5–7); the Eurasian fossil record indicates that late Pleistocene climatic changes reduced the range of several species, and the last glacial-interglacial transition appears to correlate with major extinction events. However, none of the previous Pleistocene glacial cycles appear to have caused widespread extinction events (3, 8), and the demise of the Beringian megafauna also appears to coincide neatly with the arrival and spread of modern human populations (9, 10). Similar evidence of a human-driven effect can be seen in central North America, where the arrival of Clovis-style hunters, extinction of megafauna, and marked climate change all cluster closely together (11, 12).

Ancient DNA (aDNA) analyses of temporally and geographically distributed megafaunal remains offer a means to address the relative roles of humans versus the climate in the Beringian megafaunal extinctions. The capacity of this technique to detect changes in genetic diversity within populations over geologically significant time scales provides the means to examine past population variation and to explore directly how populations respond to climate and environmental changes (13). Geographic and temporal discontinuity in the genetic data can be compared to past climate reconstructions and specific ecological events, such as advancing ice sheets, a volcanic eruption, or human colonization. Previous large-scale aDNA studies of this kind have focused on herbivores, such as bison and horses that survived to the present in relatively large numbers and are thus relatively
abundant today (13, 14), or others, such as the mammoth that are now extinct (15, 16). We focus on the musk ox, *Ovibos moschatus*, one of the few large mammals adapted to a high arctic environment, and which has a distinctly different pattern of survival than the previously studied species. Although the musk ox was holarctically distributed at the beginning of the Holocene, only small, relict natural populations isolated on Greenland and the North American Arctic Archipelago survive to the present day. The Pleistocene musk ox went extinct over most of its Eurasian range at the beginning of the Holocene. In Europe, the latest known occurrences are in late glacial deposits in Scandinavia, and in the Taimyr Peninsula and the Lena River mouth at the Bykovsky Peninsula (Northeast Siberia) they survived until around 2,500 years before present (YBP) (17, 18), extending the survival of this species in northern Russia well in to the Holocene epoch.

Whether musk ox population decline can be attributed to human or other effects is unclear. In North America, the decline of related ovibovines appears to correlate closely with the arrival of humans, whereas longer human-ovibovine cohabitation is observed in northern Eurasia (3, 19). Given the conflicting evidence, a principal aim of this study was to further investigate the question, through reconstruction of past musk ox population dynamics using aDNA, and comparison of the findings against human and climatic records from the late Pleistocene and Holocene and known biological characteristics of the species.

**Results and Discussion**

To investigate changes in genetic diversity among musk ox since the late Pleistocene, we analyzed 682 bp of the mitochondrial control region, obtained from 149 radiocarbon dated specimens that range in age from 56,900 radiocarbon (14C) YBP to the present (Fig. 1). Specimens were sampled from their previous geographic range, spanning from the Urals (n = 26) and the Taimyr Peninsula (n = 54) in the west, through Northeast Siberia (n = 12) and into North America (n = 14) and across to Greenland (n = 43) in the east. Our data demonstrate continuous presence of musk ox in Eurasia and North America throughout radiocarbon time until their extinction in Eurasia (∼2,500 YBP) and demographic retraction in North America (∼10,000 YBP).

**Loss of Mitochondrial DNA Diversity.** Modern musk ox harbor little genetic diversity at both the nuclear and mitochondrial levels (20–22). Our data indicate that total past genetic diversity (samples older than 100 14C YBP), as measured by temporally unbiased π (23), was significantly higher than present (Table 1 and Fig. S1), with Serial SimCoal (SSC) simulations indicating that this decrease in genetic diversity cannot be explained by genetic drift in a constant-sized population. This observation is consistent with the results of a smaller, previous aDNA study (24) and with paleontological evidence, which suggests a large decrease in musk ox geographic range and population size during the late Pleistocene. To investigate whether the arrival of humans into the musk ox range played any effect on the species’ genetic diversity, a similar analysis was performed on the Taimyr and Canada datasets (the only two with a meaningful number of musk ox on either side of the entry). In neither case was human arrival associated with any statistically significant change in genetic diversity (Table 1 and Fig. S1).

![Fig. 1. Bayesian Skyride derived from the geospatial analysis of 135 ancient and 14 modern musk ox mtDNA control region sequences. The x axis is in units of radiocarbon years in the past, and the y axis is equal to Me*¢ (the product of the effective population size and the generation length in radiocarbon years). The colors represent the relative contribution of each of the geolocations to the overall estimated effective population size. Colors indicate geographical origins of the samples: blue, Greenland; red, Northeast Siberia; orange, Taimyr; green, Urals; light blue, Canada. The bar graph shows the number of radiocarbon-dated samples in bins of 2,000 radiocarbon years. No relation is apparent between the absolute number of samples and the estimated effective population size or transition time.

**Three Geographically Structured Clades.** Phylogenetic analyses of the data subdivides musk ox into three, well-supported and temporally structured clades (clades 1–3) (Fig. 2). The phylogeny is consistent with that recovered when (i) 13 radiocarbon-infinite sequences are added to the alignment, or (ii) the analysis is restricted to a subset of 60 samples for which the complete control region sequence was obtained (Figs. S2 and S3). Clade 1 comprises samples that range in age from beyond the 14C limit to 42,550 14C YBP, originating predominantly in Northeast Siberia but also in the Taimyr Peninsula. By ∼45,000 14C YBP, most of the diversity present before this time had been lost.

Clade 2 consists of specimens almost exclusively originating from the Urals and Taimyr region, ranging in age from beyond the 14C limit to ca. 13,000 14C YBP (although a single outlier in this study is associated with a much more recent date of 3374 14C YBP). The geographic distribution of samples belonging to clade 2 suggests: (i) that these regions were either independently populated from a mixed source population, (ii) that no significant physical barriers to gene flow were present across this wide geographic expanse, or (iii) a combination of these. During the Quaternary, ice sheets over the Barents and Kara seas expanded several times onto mainland Russia, blocking north-flowing rivers such as the Yenissei, Ob, Pechora, and Mezen. Large ice-dammed lakes formed a barrier

**Table 1.** Observed nucleotide diversity and polymorphic sites of data subsets

<table>
<thead>
<tr>
<th>Data subset</th>
<th>x*</th>
<th>Significance</th>
<th>M</th>
<th>PM</th>
</tr>
</thead>
<tbody>
<tr>
<td>All modern samples (&lt;100 14C YBP)</td>
<td>0.008</td>
<td>6,417</td>
<td>26</td>
<td>9</td>
</tr>
<tr>
<td>All ancient samples (&gt;100 14C YBP)</td>
<td>0.021</td>
<td>9,993</td>
<td>575</td>
<td>549</td>
</tr>
<tr>
<td>Canada, prehuman contact</td>
<td>0.026</td>
<td>4,801</td>
<td>70</td>
<td>Na</td>
</tr>
<tr>
<td>Canada posthuman contact</td>
<td>0.019</td>
<td>6,689</td>
<td>45</td>
<td>Na</td>
</tr>
<tr>
<td>Taimyr prehuman contact</td>
<td>0.025</td>
<td>4,006</td>
<td>136</td>
<td>Na</td>
</tr>
<tr>
<td>Taimyr posthuman contact</td>
<td>0.017</td>
<td>6,815</td>
<td>39</td>
<td>Na</td>
</tr>
</tbody>
</table>

* M, number of polymorphic sites; Na, Not applicable; PM, number of private mutations.

† Temporally unbiased x calculated following Depaulis et al. (23).

‡ Rank (of 10,000, of uncorrected x) among data generated through Serial SimCoal simulations of a constant population in which gene frequencies are changing only through genetic drift. Placement of the observed value outside of the 95th percentile (i.e., outside of rank 250-9,750) can be considered as significant at <5% level.

§ Significant at P = 0.0007.

Contact time as shown in Fig. 3.
Greenland was the last land mass colonized by musk ox, and the data clearly demonstrate that North America is the source of the Greenlandic populations, as it is also suggested for caribou (27). This finding is consistent with the complete glaciation of Greenland during the LGM (18–21,000 YBP). Any areas that remained ice-free during this period were very small and isolated (28), making it unlikely for any mammal to survive on the island.

**Geospatial Demographic Analyses.** To explore the geographic origin, spatial distribution, and demographic history of the musk ox, we performed combined temporal and spatial analysis including all finite-dated musk ox (149 specimens) from across the sampling range. These analyses provide insights into the geographic origin of the clades, their interrelationships, and the dynamics of local populations and the species as a whole, and allow additional inference into the possible causative agents of observed changes in genetic diversity. These analyses were performed using a geospatial inference package incorporated within the Bayesian phylogenetic inference software, BEAST (29).

A Bayesian skyride (30) reconstruction of changes in genetic diversity through time (Fig. 1) shows that after an increase in diversity during MIS 4 and 5, diversity declined during MIS 3, between 30,000 and 33,000 $^{14}$C YBP. This decline was followed by a subsequent rapid increase in overall diversity, followed by a steep decline circa 18,000 $^{14}$C YBP. Finally, this decrease in diversity begins to reverse during the mid-Holocene, associated with increase in population size following the establishment on Greenland. This finding is unlikely to be a sampling artifact, as both an analysis of a randomly chosen subset of samples from the whole distribution ($n = 40$) and an analysis of 78 samples with equal temporal distribution (0–5,000; 15,000–20,000; 20,000–25,000 $^{14}$C YBP) recovers the same pattern (Figs. S4 and S5).

The dynamics reported here differ substantially from the two previously published aDNA datasets that are sufficiently large for comparable demographic analysis, mammoth (16) and bison (31) (Figs. S6 *Inset* and Fig. S7). Although both musk ox and mammoth experience an increase in diversity during the MIS 4 glacial, followed by decline during the MIS 3 interstadial, genetic diversity in mammoth begins to decline much earlier than in musk ox (65,000 $^{14}$C YBP vs. 48,000 $^{14}$C YBP), and appears to be reduced by a much larger extent. Clear differences are also observed between the musk ox and bison datasets, in particular with the bison starting to decline around 30 to 35,000 $^{14}$C YBP, coincident with a period of increase in musk ox diversity (Fig. S7).

These fluctuations may reflect either the degree of impact of human hunting on each species, their natural abilities to respond to changes in the Beringian ecosystem (both in overall temperature, but also rate of temperature fluctuation), or a combination of both. In contrast to bison, musk ox are adapted to high arctic, thus extremely cold, environments, preferring dry and cold continental climates where winter snow cover is either shallow or patchy with areas of accessible forage (cf. 32, 33). Over historic time-scales, musk ox abundance and distribution in both Greenland and Arctic Canada have fluctuated significantly in response to both long- (32, 34) and short-term shifts in climate. For example, Miller et al. (35) reported a 76% decrease in musk ox abundance on Bathurst Island in just 1 year, between 1973 and 1974, because of deep and persistent snow cover and icing. The climatic fluctuations in the High Arctic are also responsible for periodic reproductive failures (35, 36).

Given these biological insights, it is tempting to speculate on whether environmental, and in particular climatic change, might have been the driving force behind the ancient musk ox population dynamics. Although difficult to test directly, because neither our dataset nor available climatic reconstruction datasets for the relevant period are temporally or geographically distributed in enough detail to enable statistically supported comparison, the Bayesian skyride provides some tantalizing insights.
The periods during which musk ox populations are seen to increase are, in general, those of global climatic cooling (in particular as peak diversity coincides with the LGM). Furthermore, the musk ox populations decline during the warmer and climatically unstable interstadials (Fig. S6), such as MIS 3, and in the postglacial warming following the LGM (Fig. 1 and Fig. S7). Although this is by no means conclusive evidence, a role of the environment as opposed to humans as the driving force is also supported by several other observations drawn from both our, and previously published data, as discussed below.

The results of the geospatial analyses point to a significant role of Northeast Siberia in the species’ population history. This region not only harbored the greatest mitochondrial genetic diversity, but also the most recent common ancestor of all sampled musk ox [e96,809 years ago (95% highest posterior density [HPD] 103,249–82,780 years ago)] and was the geographical source of all three clades. Despite its key role in musk ox history, both the time of extinction of clade 1 and the temporal distribution of the 14C dated specimens (Figs. 2 and 3) indicate that a major reduction in musk ox diversity occurred in this region at ca. 45,000 14C YBP. There is no archaeological evidence of human presence this far north at this time; the first well-supported evidence of human settlements in Northeast Siberia are at a Paleolithic site in the Yana River basin at 27,000 14C YBP (37). Thus, an alternate explanation for the loss is required. Intriguingly, the loss of a distinct mtDNA clade for mammoths has also been observed in this region at this time (15, 16, 38), suggesting the cause was not species-specific. Given that during MIS 3 both polar ice-core records and pollen, plant macrofossils, mammal, and insect remains indicate that the climate started to become increasingly warmer and wetter (39–41) (Fig. S7), and that, as discussed above, musk ox are sensitive to both climate warming and fluctuations (32, 42), climatic-driven environmental change is a plausible hypothesis.

The distribution of the 14C dated samples in both the Taimyr and Urals indicates a widespread loss of musk ox in these regions following the LGM, culminating by around 13,000 14C YBP (Fig. 3). There is no evidence for the presence of modern humans in the area until the very end of the Pleistocene or early Holocene (43–45), and as mentioned above, the human arrival in the Taimyr is associated with no statistically significant change in genetic diversity (Table 1). However, this time does coincide with the Bølling/Allerød period, during which the climate became more humid and a general deglaciation of permafrost started, accompanied by a dramatic change in vegetation from tundra-stepppe to wet tundra and forest tundra (7). Thus, once again, the data directly suggest that these fluctuations were driven by nonanthropogenic factors.

Further evidence of the competing roles of humans versus environment is provided by the Greenlandic data. Although musk ox were present in the Americas throughout the Pleistocene, they did not colonize Greenland until very recently. Although other species, such as caribou, are known to have been present in Greenland during the last interglacial (46), the fossil record shows that musk ox first arrived in Greenland during the mid-Holocene, with the oldest samples dated to 4,500 14C YBP, from Paleoeskimo Independence I archeological sites in Northeast and Northwest Greenland (47). The fossil record and the modern geographical range of the musk ox suggest that dispersal into Greenland took place from Ellesmere Island in Canada by way of the Robeson Channel to Northwest Greenland, followed by dispersal along the east coast into central East Greenland (48, 49). Our results are consistent with this pattern, and support a single colonization event followed by population expansion.

Bayesian skyline analysis of the restricted Greenlandic dataset suggests rapid population growth following this initial dispersal into Greenland (Fig. S8). This theory is consistent with the expected dynamics of a species entering virgin terrain in the absence of competition and the fact that musk ox thrive during climatically stable periods (42), such as those reconstructed for the last ca. 10,000 years in Greenland using GISP2 records (Fig. S7) (50, 51). This finding, plus the contemporaneous arrival and colonization of Greenland by humans and musk ox, and the fact that musk ox genetic diversity in Canada did not change significantly following human arrival in the Americas (Table 1), provides further evidence of a negligible effect of humans on musk ox. Arguments that musk ox were not of interest to the first humans as prey can also be discounted, as the Independence I culture are known to have been expert musk ox hunters, as demonstrated by the large number of musk ox bones recovered from archeological sites across the high Arctic of Canada and Greenland that have been associated with this culture (52).

In summary, although humans may have played a significant role in the history of other large Beringian mammalian herbivores, to our knowledge this example is unique in showing there is no evidence that humans drove musk ox demographic fluctuations over the last 60,000 years. We argue that an alternate cause, possibly environmental change driven by the climate (53), was the major force behind musk ox population dynamics over this period, and expect that with the arrival of future aDNA and climatic reconstructions, this hypothesis can be more explicitly tested.
We collected samples from 447 bone, sp. For 60 specimens we were able to recover the complete control

Canada

SI 441:207

fi

to 1.0

10

Phylogenetic relationships among the 162 specimens that

Grant (to P.F.C. and E.W.).

and 1.2

Table S2

10

100

Geographic origin of musk ox samples that yielded aDNA sequences.

was also performed on the dataset after removing all singletons. Again, genetic relationships among the dated specimens were estimated using BEAST with evolutionary and coalescent models, as described above. The results do not differ from the previous analysis (with or without PMD) except that the effective population (Ne) size decreased as it was expected because we removed diversity when we discarded the singletons (Fig. S5).

We used SSC (63, 64) to model several aspects of the data (Fig. S10). First, the results of the phylogenetic analysis indicate that mtDNA phylogenetic structure existed in the musk ox population (Fig. 2), with particular distinction between the Taimyr/Ural individuals, and the rest. As demographic parameters are estimated by BEAST under the assumption of a single population, SSC was used to explore whether the population dynamics recovered in the BEAST analyses (Fig. 1) could arise simply as a consequence of population substructure combined with our sampling strategy. Second, we used SSC to model whether the observed nucleotide π diversities of the modern (< 1014C YBP), ancient (> 100 14C YBP), and pre- and posthuman contact samples (from Canada and Taimyr) differed significantly, or could be explained solely as a result of our sampling scheme in a single, constant-sized, population. Full details of the simulations are described in SI Materials and Methods.

To assess the geospatial distribution of musk ox through time, an additional BEAST analysis was performed with the full dataset of finite radiocarbon-dated specimens. In this analysis, in addition to the parameters described above, the probability distribution of the geographic locations of each node in the tree were also inferred. Briefly, this discrete geospatial model (65) assigns each sequence to a fixed location in space (here, five locations were assumed: Greenland, Northeast Siberia, Taimyr, Ursal, and Canada). Rates of diffusion between each of these sites are then estimated according to a continuous-time Markov chain. Under the continuous-time Markov chain model, the unobserved location at the root of the tree derives from a uniform distribution over all sampled locations. Dispersal then proceeds conditionally independently along each branch in the tree according to a memory-less transition process and ultimately gives rise to the observed locations at the tips. An infinitesimal-rate matrix Q characterizes this Markov model and we estimate these rates from the data using Bayesian stochastic variable selection to achieve statistical efficiency as the data represent only a single realization from the process. The posterior probabilities of a given node in tree existing at each possible location are simultaneously estimated along the tree and evolutionary model in BEAST. This simultaneous inference also assumes the Bayesian sapline plot coalescent prior over the tree and incorporates the PMD model to account for possible damaged sites. Two MCMC chains were run for 100 M iterations, with subsamples recorded from the posterior every 10 K iterations.

ACKNOWLEDGMENTS. The authors thank Jennifer Leonard for useful discussion and A. Cooper, P. Matheus, J. Meng, and D. Mol for access to their collections and obtaining samples. This study was supported in part by Forsknings-og Innovationssytelsen 272-07-0279 Skou Grant (to M.T.P.G.) and the Marie Curie Actions “Genetime” Grant (to P.F.C. and E.W.)

