

Ecological Change, Range Fluctuations and Population Dynamics during the Pleistocene Review

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Apart from the current human-induced climate change, the Holocene is notable for its stable climate. In contrast, the preceding age, the Pleistocene, was a time of intensive climatic fluctuations, with temperature changes of up to 15°C occurring within a few decades. These climatic changes have substantially influenced both animal and plant populations. Until recently, the prevailing opinion about the effect of these climatic fluctuations on species in Europe was that populations survived glacial maxima in southern refugia and that populations died out outside these refugia. However, some of the latest studies of modern population genetics, the fossil record and especially ancient DNA reveal a more complex picture. There is now strong evidence for additional local northern refugia for a large number of species, including both plants and animals. Furthermore, population genetic analyses using ancient DNA have shown that genetic diversity and its geographical structure changed more often and in more unpredictable ways during the Pleistocene than had been inferred. Taken together, the Pleistocene is now seen as an extremely dynamic era, with rapid and large climatic fluctuations and correspondingly variable ecology. These changes were accompanied by similarly fast and sometimes dramatic changes in population size and extensive gene flow mediated by population movements. Thus, the Pleistocene is an excellent model case for the effects of rapid climate change, as we experience at the moment, on the ecology of plants and animals.

Introduction

The Pleistocene is often characterised as a time of massive glaciers covering much of Eurasia and North America with a fauna consisting of enigmatic creatures such as mammoths and sabre-toothed cats (Figure 1). As always, reality is more complex. The Pleistocene started about 1.8 million years ago and ended 10,000 radiocarbon years ago (we use radiocarbon years throughout the text when referring to years ago or before present) [1]. The Pleistocene is notable for massive climatic fluctuations. The causes of these climatic fluctuations are still an area of active research, but are already known in some detail. There are several layers of periodicity overlaying each other, at intervals of approximately 100,000, 41,000 and 23,000 years [2], resulting in the glacial–interglacial (cold–warm) cycles. Numerous additional cold and warm events overlie the main curve [3]. Therefore, during the Pleistocene, periods of cold and warm climate occurred at irregular intervals and with varying durations (Figure 2). Both the extent and the speed with which these temperature fluctuations occurred are quite remarkable.

For example, during times of changes from stadials to interstadials the average temperature changed by 5–10°C within as little as a few decades [4]. Similarly, during glacial maxima, the temperature was up to 21°C colder compared to today [5]. Only during these times did the northern hemisphere conform to the movie image of the Pleistocene with extensive glaciers covering large parts of Eurasia and North America. The large amounts of water bound in glaciers also resulted in sea-levels being up to 120 m lower than today and thus in a much larger land area with, for example, the British Isles being connected to the European mainland and the North Sea being a large area of dry land. However, as recent research has shown, the extent of the northern glaciers was much smaller during most of the Pleistocene [6]. Moreover, it should be noted that during some interglacials, such as the Eemian (127,000–117,000 years ago [7]), temperatures were warmer than today. It was during these times that elephants (the extinct straight-tusked elephants *Elephas antiquus*) and hippopotamus were living as far north as the British Isles (Figure 1, lower image) [8].

Clearly, these massive climatic and environmental changes significantly influenced the distribution and genetic diversity of plants and animals. The idea that, during times of adverse climate, species track their habitat goes back to Darwin [9], and the Pleistocene should represent an excellent opportunity to test this assumption. Generally, one would assume that Arctic species would expand their distribution southwards during colder times and that temperate species would expand northwards during warmer times. While this is straightforward in North America, with mountain chains, which represent partial barriers to range shifts, running from north to south, in Europe a level of complexity is added with mountain chains running from east to west and the available land mass becoming smaller to the south and being divided into several peninsulas bordering the Mediterranean. This geography, together with numerous studies that found geographical patterns in the genetic diversity of many species consistent with colonization of mid-latitude and northern Europe from the Iberian Peninsula, Italy and the Balkans (for review, see [10,11]) has resulted in the classical ‘refugium theory’, which proposes that temperate species survived the glacial maxima in southern refugia with little gene flow among them and colonized the more northern parts from there during interglacial times. While this model is theoretically sound and correct in many aspects, recent studies on both modern and, especially, ancient DNA diversity have shown that reality is much more complex and only very broadly follows a contraction–expansion model for population dynamics, with many additional processes complicating the picture [12–16].

Finally, the end of the Pleistocene is marked by a massive extinction of large land vertebrates across most of the world (Box 1), with the exception of Africa [17]. Although these extinctions have long been known, their causes remain controversial. While some authors blame humans [18], others deny any human influence, at least on the continents, although human-induced extinctions are widely accepted for islands [19]. Again, recent research has revealed a great deal about

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Figure 1. Artistic renderings of Pleistocene landscapes in middle Europe.

Upper: Mammoth steppe during a glacial phase with its namesake, woolly rhino and Neanderthals. Lower: Open woodland with straight tusk elephants, hippopotamus and cave hyenas during the Eemian. Images reproduced with permission from Natural History Museum, London.

the timing and processes of these extinctions, showing that not only mammoths [20,21], but also giant deer (deceivingly known as Irish elk) [22] and some Caribbean ground sloths [23], survived into the Holocene. However, when it comes to the cause(s) of these extinctions, the verdict is still out.

In this review, we will discuss the dynamics of animal and plant populations during the Pleistocene, trying to outline how populations reacted to the rapid variations in climate. We will restrict our analyses to the northern hemisphere, as the majority of studies on Pleistocene DNA have been done on species from this region.

The Glacial-Refugium Theory

It was first recognized 30 years ago that seemingly continuously distributed populations may show geographical structure in their genetic diversity [24]. Since then, this field, later named phylogeography, has grown into an independent discipline [11,25]. With increasing amounts of DNA sequence data from a variety of species, a model of habitat contraction to southern peninsulas during glaciations and expansion into northern parts during warmer times became increasingly

popular [10,11]. It certainly conforms to the notion that we generally do not expect to find animals in large glaciated areas. Moreover, this model leads to some predictions about the pattern of genetic diversity we expect. Thus, with an expansion into previously unoccupied territory, we expect genetic diversity to decrease from the south to the north [26], which is often what is observed. Similarly, the genetic diversity of many species in Europe is divided geographically, with the groups often broadly conforming to a western group (derived from refugia in the Iberian Peninsula), a southern group (from Italy or the Balkans) and an Eastern group (whose refugial area was for a long time less clear; e.g. [27]). Similar patterns have been shown for North America, where, for example, modern brown bears can be divided into several genetic groups with virtually no overlap in their geographical distribution. Finally, populations of temperate species in North America, in contrast to Amazonian species, have been shown to display a signal of recent population growth [28], indicating that conditions became more favourable for temperate species relatively recently – an observation compatible with habitat expansion in these species.

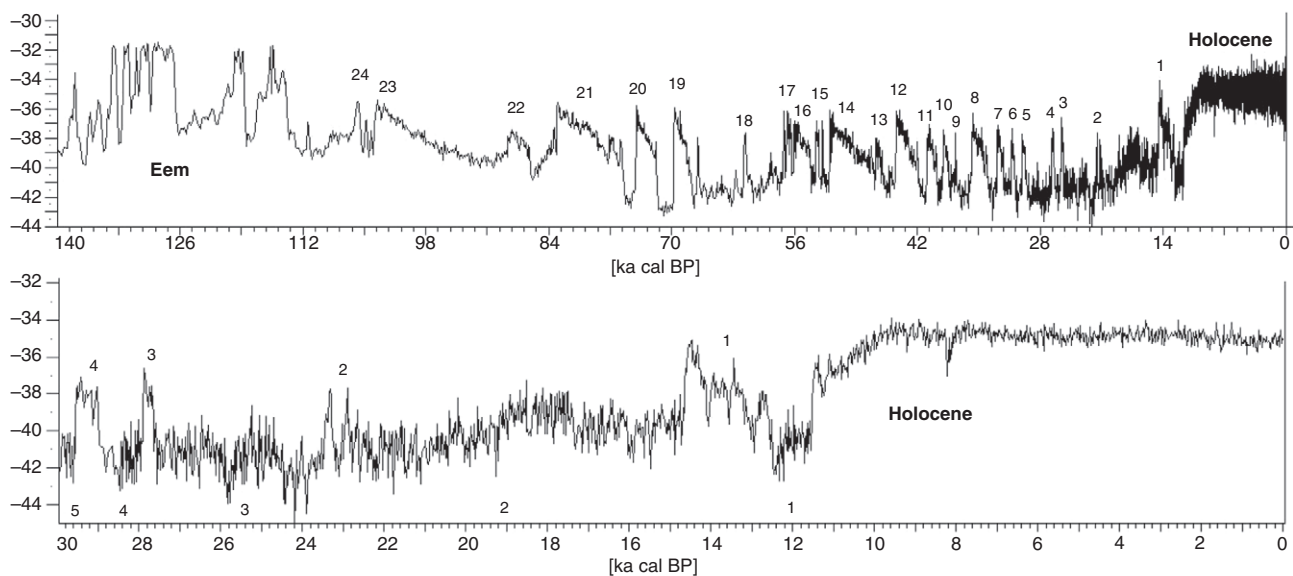


Figure 2. Climate variation during the Pleistocene.

The x-axes show time in calibrated thousands of years before present and the y-axes show variation in oxygen isotope ratio, whereby more negative values indicate colder temperatures. Top panel: Climate change during the last 140,000 years. Numbers show the Greenland interstadials, short warm periods during the last glacial. The Eem is the interglacial period preceding the last glacial. Bottom panel: Close-up of the climatic changes during the last 30,000 years as determined from the GIPS2 Greenland ice core. Interstadials are numbered above the climate curve, stadials below the climate curve. Graphs courtesy of Dr. Robert Sommer, University of Kiel.

Thus, there is a lot of evidence that the expansion–contraction model is an adequate description of the population dynamics during the Pleistocene, even though most of its inferences are extrapolated from extant data.

However, in 2000, the idea that population histories could be inferred from modern genetic data alone received a challenge through the publication of six ancient DNA sequences of brown bear dating to between 35,000 and 40,000 years ago [29]. These six samples represented all three major genetic groups of brown bears living today in North America. However, in contrast to the modern situation, all groups were found to have lived close together [29]. Later, a more comprehensive study [13] showed that while two of the clades indeed occurred in the same region, the third came from an adjacent but distinct area. In addition, the authors showed that the samples belonging to two of the three clades represented distinct subclades which went extinct 35,000 years ago, leaving eastern Beringia (the western part of Alaska and the eastern part of Siberia, including parts of the then dry Arctic Sea) without brown bears for about 14,000 years. At the height of the last glaciation, 21,000 years ago, brown bears belonging to the modern subclades, and almost certainly coming from the north rather than from the south, then re-colonised North America in two distinct waves [13]. While large parts of northern North America could not have been the source region for these bears, it is now well established that Beringia was ice-free for most of the late Pleistocene. Thus, there may have been northern refugia in addition to southern ones.

This concept of northern refugia [30] did not imply that the southern refugia did not exist — there is overwhelming evidence that they did. However, it has become clear that southern refugia were not exclusive and that the nature of refugia is likely to be different for differently adapted species [31]. Cryptic northern refugia have also been proposed for

trees based on both molecular and macrofossil evidence [32,33]. Similarly, on detailed examination using large sample numbers and dense geographical sampling, modern DNA sequences may reveal much more complexity than previously assumed. For instance, a study of common voles [34] showed that they display as many as six clades, one of which only occurs in a very restricted area in southern Germany around Freiburg. Therefore, there appears to be more to Pleistocene population dynamics than survival in the south during cold times and recolonization of areas further north during warmer episodes. This view has now been supported by the fossil record itself [35,36]. Moreover, both molecular data [37,38] and studies on the fossil record [36,39] indicate that one of the long-sought eastern refugia was most likely in the Carpathians, while another refugium probably existed in the region of the Dordogne, southern France [15,36].

The term ‘northern refugia’ cannot, however, be used uncritically. Importantly, it does not make much sense to talk about northern refugia when referring to species that are adapted to Arctic climates today such as the Arctic fox (*Alopex lagopus*) or the ptarmigan (*Lagopus* sp.). In contrast to temperate species, for which the glacials were times of habitat reduction, these species expanded their geographical range during glacials [40]. Therefore, they currently live in refugia which are in the north. In contrast, with respect to temperate species, northern refugia refer to regions in which such species survived glacial maxima to the north of the classical southern refugia. In a way, these regions represent, for temperate species, what mountain regions like the Alps are for cold-adapted species today like the ptarmigan — climatic islands in which conditions differ favourably from the surrounding areas [30,41].

Thus, while the existence of southern refugia has been supplemented by additional refugia to the north, it remains clear that survival in the south was important for many

Box 1

Signature pleistocene animals.

The **Arctic fox** (*Alopex lagopus*) is a small (smaller than the red fox) white or bluish-grey fox that lives today in the arctic northern hemisphere of the Holarctic from Greenland to Iceland and the Arctic regions of North America and Eurasia. During the Pleistocene it had a much wider distribution across the middle part of Europe and western Asia as well as in the large ice-free region of Beringia. It is primarily an inhabitant of the tundra and mountainous regions above the tree line, but it does penetrate into the taiga to some degree. Arctic foxes feed primarily on lemmings, but their diet also includes Arctic hare, eggs, and carrion scavenged from the leftovers of larger predators. A remarkable characteristic is their capability for long distance dispersal, with movements up to 2,000 km.

The **brown bear** (*Ursus arctos*) had and still has by far the largest habitat range of all living bear species. Formerly, its habitat extended across North Africa, Europe, the northern and middle parts of Asia and North America from Alaska down to Mexico. Due to intensive human persecution, it is now extinct in many of these areas, including North Africa, large parts of Europe and most of North America. Brown bears are very adaptable and can live on both a mostly herbivorous diet and a mostly carnivorous diet. They are very variable in size and other morphological traits which historically has led to the description of numerous subspecies and even species. Today, all brown bears are considered a single species with a number of subspecies.

Cave bears (*Ursus spelaeus*) are the close — and less fortunate — cousins of the brown bear. The two species diverged some 1.6 million years ago, with tooth and stable isotope analyses indicating that cave bears were mostly herbivorous. However, recently a population was discovered that shows a stable isotope signature indicating an omnivorous, or even carnivorous, diet. Although in Europe cave bear remains are much more numerous than those of the brown bear, cave bears went extinct some 25,000 years ago. It has recently been shown that cave bears also occurred in Asia up to north-eastern Siberia.

Cave hyenas (*Crocota crocuta spelaea*) are close relatives of the living spotted hyenas from Africa. In fact, in mitochondrial DNA sequence trees, sequences of cave and spotted hyenas are quite intermingled, questioning any taxonomic distinction of them as a subspecies or even as a species. Judging by cave paintings, they were probably spotted like modern spotted hyenas in Africa. They lived in Eurasia throughout the Pleistocene and probably already during the late Pliocene, about 3 million years ago. The timing of their extinction is not well established, but may have taken place around the same time as the cave bear, some 25,000 years ago.

The **giant deer** (*Megaloceros giganteus*), or Irish elk, is the gigantic relative of the rather gracile fallow deer. Giant deer are not only remarkable for their large body size but also for their huge antlers which could span up to 3.5 meters. Giant deer are often seen as typical representatives of the Pleistocene, but recent research has shown that in the Urals, giant deer survived until at least 7,700 years ago, far into the Holocene.

The **woolly mammoth** (*Mammuthus primigenius*) is no doubt the most iconic of all extinct Pleistocene animals. However, the woolly mammoth is only the last representative of a long lineage that had its origin in Africa. The first European mammoth lived in southern Europe and only later did mammoths colonize the arctic regions. Woolly mammoths differ from their closest relatives, the living elephants, in many features, most conspicuously by their curved tusks, the long hair and their small ears and short tails. Tens of thousands of mammoth bones have been recovered from the northern permafrost regions and sometimes even complete frozen carcasses. Mammoths survived into the Holocene, with the last population disappearing from Wrangel Island only about 3,700 years ago.

The **steppe bison** (*Bison priscus*) must have been a very common species throughout the Arctic region, especially in Beringia, given the vast numbers of fossils that have been found. Steppe bison were very variable in their morphology, especially with regard to the size of their horns, which were much larger in some individuals than in modern bison. They went extinct in Eurasia, but genetic analyses have established that they were the ancestor of the modern American bison, *Bison bison*. Their relationship to the European bison, *Bison bonasus*, is not known.

species. It has also become clear that species usually do not react identically to changing environments, but are individualistic in their response [10,31].

Pleistocene Populations

There are, in principle, two approaches by which we can learn more about the dynamics of Pleistocene populations: first, by analysis of the fossil record and second, by sequencing ancient DNA from these remains and studying the genetic diversity of the investigated species. Morphological identification and carbon dating of fossil remains has the advantage that at least the first part is cheap, comparatively rapid and often works reliably. The substantial progress made in carbon dating also means that changes in a species' geographical distribution can be followed relatively precisely. However, species are often made up of clades that are only genetically distinguishable, but not by their morphology. Thus, even if the geographical distribution of a species, or its abundance, does not change in the fossil record, its genetic diversity and the geographical structure of this diversity may do so dramatically. This is well illustrated by the example of the Alaskan brown bears

mentioned above [13]. Fossil identification and radiocarbon dating clearly show that brown bears were absent from Alaska between 35,000 and 21,000 years ago. However, only genetic analyses revealed that the population that recolonised Alaska 21,000 years ago was genetically different from the one that had gone extinct earlier. Thus, ancient DNA not only revealed another level of complexity, it also showed that modern Alaskan brown bears did not come from the south but rather from western Beringia to the north. The disadvantage of such ancient DNA analyses is that they tend to be time-consuming, and that there is a clear limit in terms of the survival time of ancient DNA — up to ~100,000 years in temperate climates, anywhere between 1 and 10 million years in permafrost regions, and sometimes only decades in warm climates [42–45] — and even within these time frames many fossils do not yield ancient DNA [46,47].

Nevertheless, the success of a combination of both approaches is well illustrated by what we now know about the population history of the most iconic of all Pleistocene mammals, the mammoth. Extensive dating studies on Eurasian mammoths have shown that they almost

completely disappeared from Europe during the warm phase of the Bolling-Allerød around 12,000 years ago. However, during the cold reversal of the Younger Dryas 10,600 to 10,000 years ago, mammoths returned to Europe and the last European population disappeared from the Baltic region about 10,000 years ago [48]. Unfortunately, we do not yet know much about the population genetics of Eurasian mammoths outside the permafrost. In contrast, we know a great deal about the genetics of mammoths from the permafrost regions of Siberia and Alaska. An analysis of 44 mammoth remains revealed that they fell into two major clades. Interestingly, one of the clades went extinct about 40,000 years ago, long before the final demise of mammoths in this region [49]. This observation was further confirmed by a second study that investigated 15 complete mitochondrial genomes [50]. So far, the most divergent sequence was from a European mammoth, indicating that more diversity remains to be uncovered outside the permafrost. In the most recent publication on this topic [51], analysis of more than 100 samples mainly from the permafrost confirmed that the two major clades went extinct at different times. Furthermore, due to the large sample size it was possible to date some of the range changes revealing that the Late Pleistocene mammoths, which represent the longer-surviving clade, had their origin in North America and migrated to Eurasia during the Middle Pleistocene. In addition a few rare, but highly divergent sequences were found in samples from Siberia and North America. The challenge will now be to extend these studies to the extensive previous mammoth habitats outside the permafrost in Europe, Asia and North America. The most likely outcome will be an even more complex picture of range expansions and contractions of mammoth during the Late Pleistocene.

Another Pleistocene species that has been studied extensively is the extinct cave bear (*Ursus spelaeus*). Cave bears comprise by far the most abundant megafauna fossil in many European caves. It has long been known that cave bears show a substantial amount of morphological variation [52], and it was proposed that cave bears may actually represent two reproductively isolated taxa [53]. While this view remains controversial, ancient DNA analyses have provided some support for it and yielded additional insights. In a study of the two Austrian caves, which had first led to the proposition of the 'two species hypothesis' for cave bears, genetic analyses failed to detect gene flow between the two caves over the 15,000 years during which both caves were inhabited by bears [54]. Furthermore, in several caves in southern Germany, both genetic groups were found in the same location. However, in these cases, the two genetic groups were separated temporally, with one form that disappeared 28,000 years ago replaced by the second population [55]. Much like the brown bear data [13], this finding suggests that in addition to the species extinctions at the end of the Pleistocene, genetically distinct populations went extinct during the Pleistocene, possibly quite regularly. These population and species extinctions can be seen as a continuum with the extinction of a species simply being the ultimate result of a succession of population extinctions. Finally, a recent study of cave bears showed that they were not restricted to Europe, but lived as far north-east as eastern Siberia, beyond the Arctic Circle [56]. However, the genetic relationships between the different locations are unexpected, with the closest relatives of the Siberian cave bears having

occurred in the Caucasus, and the closest relatives of cave bears from the Altai Mountains being found in Western rather than Eastern Europe, as might be expected from geographic distance.

This dynamic perspective of the genetic makeup of Pleistocene populations is a recurring theme in ancient DNA studies. In Beringian wolves, it was found that the Pleistocene population was most likely ecologically adapted to hunting or scavenging megafaunal species and disappeared at the time its prey went extinct [12]. The Pleistocene Beringian wolves were not only genetically different but also showed different isotopic values and tooth wear patterns, indicative of their adaptation to a megafaunal prey. Only by integrating molecular data with morphological and palaeoecological ones was it possible to interpret the disappearance of this special group of wolves.

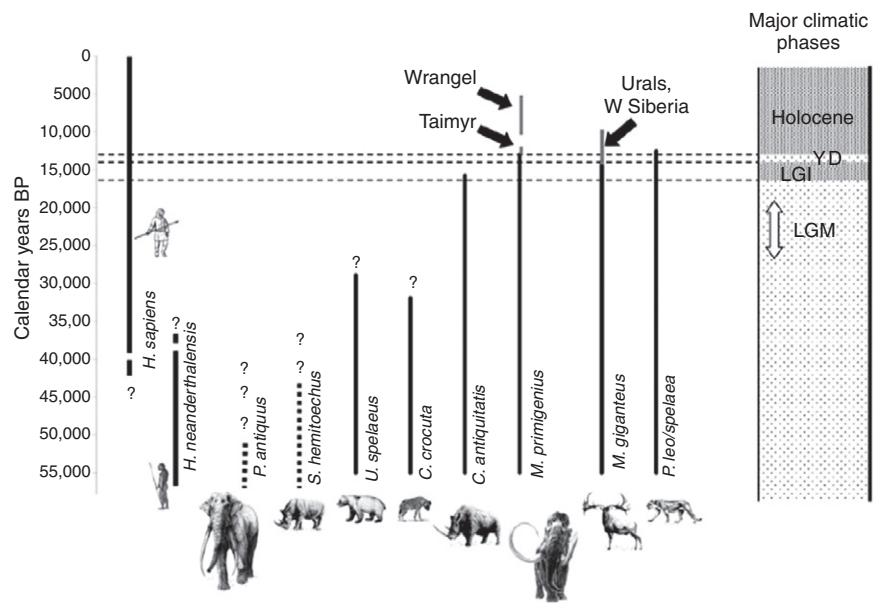
Cryptic population events have also been revealed by ancient DNA sequencing of Pleistocene cave hyenas [57]. In this case, multiple immigrations of hyenas from Africa to Eurasia were revealed. This was unexpected, as the Eurasian cave hyena had been morphologically determined by some as a different subspecies or even species. However, the mitochondrial DNA study suggested that African and Eurasian hyenas are well mixed, strongly arguing against a distinct taxonomic status of cave hyenas. Although it is not yet clear why hyenas emigrated from Africa three times, the timing of these population movements using molecular dating is intriguing. While the first expansion took place 3 million years ago, the other two conspicuously match up with major events in hominid history. The second immigration took place sometime after 1.3–1.5 million years ago, matching quite well with the first occurrence of hominids in Europe [58], while the last one dates to 360,000 years, almost exactly the date of the population divergence of anatomically modern humans and Neanderthals as determined by genomic data [59].

Even species that have been seen as hallmarks for a classical phylogeographic structure, such as the brown bear, may show more population changes than expected. Two studies investigating the genetic diversity of European Pleistocene brown bears [15,60] found that during the Pleistocene the modern Iberian brown bear sequences were found in Italy, while those from the modern Italian clade, and a clade from further east, had lived in the Iberian Peninsula. In addition, an entirely new clade was found that may have survived the coldest stages of the Pleistocene in southern France and only went extinct during the Holocene due to human persecution. In fact, the strong phylogeographic pattern seen for the brown bear today may represent an historically unique situation brought about by population-size reduction due to human hunting. If a population declines in size, usually the rarer sequences are lost first from the population [61]. This will eventually lead to populations which are reciprocally monophyletic with regard to their DNA sequences, especially if some geographical structure existed before the population decline with different sequences being more common in one or another region.

In summary, almost every study investigating Pleistocene populations has revealed a far more complex history of population dynamics than could otherwise have been predicted. The picture that seems to emerge is one of extensive range changes in species during the Pleistocene with population replacements and less pronounced phylogeographic patterns than today [14,62].

Figure 3. Megafaunal extinctions.

Timing of the extinction of different megafauna species in Europe and northern Asia based on direct radio carbon dates: *Homo sapiens* (human), *Homo neanderthalensis* (neanderthal), *Palaeoloxodon antiquus* (straight-tusked elephant), *Stephanorhinus hemitoechus* (steppe rhino), *Ursus spelaeus* (cave bear), *Crocota crocuta* (cave hyena), *Coelodonta antiquitatis* (woolly rhino), *Mammuthus primigenius* (woolly mammoth), *Megaloceros giganteus* (giant deer), *Panthera leo spelaeus* (cave lion). Grey lines indicated by arrows show survival of species in geographically restricted regions (LGM: last glacial maximum; YD: Younger Dryas). Reproduced with permission after [105].



Late Pleistocene Extinctions

The question of why so many species of large mammals went extinct at the end of the Pleistocene has puzzled scientists for over a century [17,63–65]. For a long time, there have been two camps of researchers, those blaming humans [66] and those excluding any human influence, at least on the continents, as opposed to islands [19], where scientists generally agree that humans were responsible for many extinctions. While some authors have argued for a combined effect of humans and climate change (e.g. [64]), and there is now strong evidence that humans have indeed been responsible for the demise of megafauna in Australia [18,67], the causes for extinctions in Eurasia and the Americas remain contentious. However, while we do not yet know the causes, much light has been shed on the timing of megafaunal extinction by large scale radiocarbon dating projects [21,22,68] and the results have yielded some surprises. Thus, the giant deer (*Megaloceros giganteus*) should perhaps be removed from the list of Pleistocene extinctions as it has been shown that they survived in the Urals up to 7,700 years B.P. [22]. Likewise, the mammoth survived on Wrangel Island until about 4,000 years B.P. [20] and on St. Paul Island in the Northern Pacific until 6,000 years B.P. [21,69]. Thus, Wrangel Island did not represent a unique situation, and more Holocene refugia for the mammoth, and possibly other taxa, may await discovery. Another group of species that has long been suspected to have survived into the Holocene are the Caribbean ground sloths, which also probably lived up to 5,000 years ago [23]. In contrast, other species, such as the American hemionid horses and the European cave bears went extinct as early as 31,000 and 25,000 years ago [68,70].

This indicates that, in contrast to Australia, where extinctions appear to have taken place rapidly once humans arrived [67], the pattern of extinctions is staggered in Eurasia (Figure 3). This makes the extinctions in Eurasia more difficult to explain by a single cause. During this staggered extinction, some species went extinct when the climate became cooler, others when it became warmer and yet again others when it seems to have hardly changed. It could be argued that if species go extinct no matter what the climate is doing, it is difficult to blame climate change, but see [31]. Moreover, numerous climatic reversals took place during the Pleistocene, so it is hard to explain why extinctions should have only taken place during the last one. However, the staggered

extinctions are also difficult to explain by human impact alone, at least if the prevailing model is that of an extremely rapid extinction [66]. There is a curious feature in the Americas and Eurasia that species generally tended to survive longer on islands that were reached by humans either late or never. However, the only extinct species from North America for which there is indisputable evidence for extinction due to hunting by pre-historic humans is the flightless sea duck from California [71]. In contrast to a model of rapid extinction due to human over-exploitation, its extinction took place after at least 8,000 years of exploitation by humans. Moreover, even though the occurrence of pre-Clovis humans in North America has now been safely established [72], the 14,000 year date for human arrival is too late to explain the extinction of the hemionid horses 31,000 years ago [68]. Therefore, at least some of the extinctions in North America cannot be blamed on humans either.

On the other hand, humans may well have played a role in some of the late Pleistocene extinctions. Humans are well known to be flexible in their foraging strategies, so once a species becomes rare or extinct they may have switched to another prey, which may explain the staggered extinction pattern. It is also likely that animal populations react differently to climate change [31] and human impact whether due to hunting or environmental alterations. Generally, however, it seems likely that many species become more vulnerable at times of climatic change. If such species then represent the preferred prey of a human population, their extinction may become inevitable.

While ancient DNA cannot reveal the causes of extinction, it can at least contribute to understanding changes in population size and genetic diversity over time. For instance, when more than 200 dated specimens of the Beringian steppe bison were sequenced for about 600 bp of the mitochondrial control region, it was found that after a phase of expansion, the bison population started to decline some 30,000 years ago and further plummeted roughly 12,000 years ago, before starting to recover about 9,000 years ago [73,74] (Figure 4A). Interestingly, this affected not only American bison, but also Siberian ones and the first decline coincided with the known appearance of humans in Siberia,

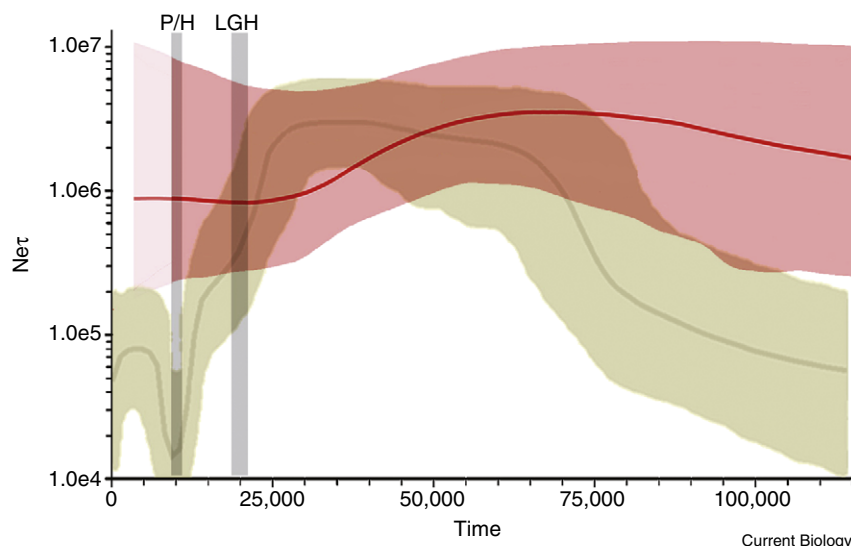


Figure 4. Pleistocene population dynamics.

Skyline plot showing population dynamics of steppe bison (green) and mammoth (pink) in Beringia during the last 100,000 years. The x-axis shows years before present and the y-axis indicates effective population sizes. The shaded areas correspond to the 95% confidence interval for the estimate of effective population size. Although mammoth went extinct and bison survived, bison populations show a much more dramatic population reduction. LGM: last glacial maximum; P/H: Pleistocene-Holocene boundary. Reproduced after [51].

whereas the second fits well with the currently accepted date for the first entrance of humans to North America [75]. Whether this correlation implies a causal relationship is, however, an entirely different question. Moreover, a recent ancient DNA study on a large number of Beringian mammoth samples did not detect any evidence for a similar substantial change in population size (Figure 4B) [51]. This is clearly at odds with the fact that the mammoth went extinct. However, if the extinction was fast enough, it may not have left a measurable trace in the genetic diversity, as population reduction affects genetic diversity with a temporal delay [61].

Although these are currently the only two studies with a sufficient number of samples to allow meaningful analyses, studies with small sample numbers also have the potential to illuminate certain issues. For instance, sequences of ten fossils of Pleistocene arctic foxes from Europe [47] that were preserved well enough to yield DNA were compared to those from modern and 100-year-old Arctic fox samples from Scandinavia. This enabled an investigation into whether the European Arctic foxes were able to track their habitat northwards at the end of the Pleistocene when the climate became warmer or, alternatively, whether they went extinct. Interestingly, the data clearly suggest that the southern population of European arctic foxes went extinct, indicating that at times of climatic change animal populations may indeed become vulnerable if they are not able to follow the habitat changes rapidly enough.

Dynamic Species in a Dynamic Ecology

The studies described above illustrate the progress in better understanding reactions of animals to the variable conditions of the Pleistocene, although they also emphasise a need to better understand the palaeoecology of different habitats over time. However, while the ecological changes that have occurred through the Pleistocene have received a great deal of attention, there is disagreement between scientists as to the nature of the vegetation that existed, especially during the Late Pleistocene.

Our understanding of terrestrial palaeoecology derives from a variety of fossil environmental proxies which include pollen and plant macrofossils, insects (both coleoptera and chironomids), ostracods and mollusca, in addition to vertebrates. Often the environment described by each of these

proxies is not precisely the same, which is in turn driven by the nature of the specific organism and the way in which their remains become incorporated into the fossil record. For many years, palynology, the study of fossil pollen, has been the dominant palaeoecological proxy and long chronological records of vegetation have been inferred, for example, from lake deposits in southern Europe [76–78]. These records have also been considered in relation to the evolution of organisms during the climatic cycles of the Quaternary [79]. However, these data are probably not adequate for describing the vegetation further north. Unfortunately, there are few long pollen sequences of the last cold stage in the northern latitudes of the Palaearctic. In order to overcome these limitations, recently databases have been synthesised of disparate sources of data to help overcome the problems of single site interpretations of palaeoecology, especially in the northern hemisphere [80–83].

A particular source of argument over Late Pleistocene environments has been the ‘productivity-paradox’, the apparently limited vegetation cover in northern latitudes which is in conflict with the abundance of megafaunal grazers [84]. This disagreement has taken place over several years, especially between those studying pollen and large mammals, respectively, from Beringia. The argument centers on whether the Late Pleistocene vegetation of the northern Holarctic from Britain to Alaska could be described as a steppe-tundra or mammoth-steppe [85], or rather as a low productivity tundra [86–90]. In part, the disagreement can be ascribed to the fact that pollen and mammals leave their fossil traces in different environments. Lake sediments do not generally include significant numbers of mammalian remains and the pollen record from fluvial deposits is not easily interpreted. However, more recently studies of plant macroscopic remains associated with megafaunal remains or from mammalian stomach contents [91,92] have shown that the plants required to sustain these animals, such as grasses and steppic herbs, did live in the north around the time of the Last Glacial Maximum. In support of this view, van Geel *et al.* [93] found a significant proportion of steppe plants in the stomach contents of a mammoth from Yakutia.

Although such vegetation has been demonstrated to have existed in Beringia at this time, it is thought that this steppe vegetation was present in varying amounts across the landscape in a mosaic (Figure 5) [85,92,94]. In fact, variations in the vegetation elements occurring on the Asian and North American continents in combination with different

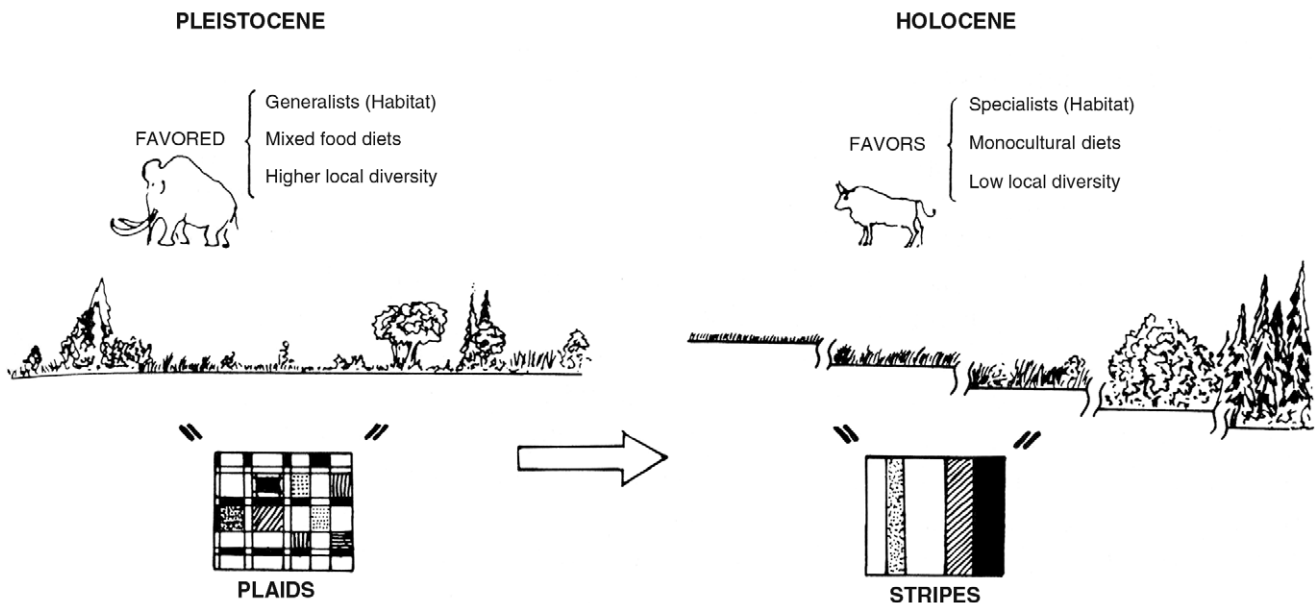


Figure 5. Shifts in landscape ecology after the Pleistocene.

Left panel: Model of a mosaic (plaid-like vegetation) during the late Pleistocene used to accommodate both the presence of megafauna and the data from palynological studies. Right: The modern vegetational zonation results in a lower carrying capacity of the higher latitudes. This classical explanation is still the most holistic explanation for the 'productivity-paradox'. For more details, see main text. Reproduced with permission from [106].

ecological requirements of different species are thought to have determined which species traversed the Bering land bridge [95]. In this instance, the lack of continuous steppe-tundra vegetation across the Bering land bridge is thought to have inhibited exchange between continents. This may explain why certain species such as the woolly rhino or the short-faced bear only occurred on one side of the Bering land bridge despite being adapted to cold environments.

Although ancient DNA has the potential to contribute to this issue, its potential has been neglected and very few studies investigating ancient plant DNA have been conducted [96]: for example, ancient DNA can be obtained directly from sediment in the absence of any identifiable macrofossils [97] and by sequencing plant chloroplast DNA, a major change in the plant composition was revealed in Siberia over the last 300–400 k years. This change from grass-rich steppe-like vegetation to a much less nutritious plant community could indeed have contributed to an increased vulnerability of animals to extinction. Even more striking were the results from ancient DNA obtained from the silty section of a Greenland ice core [45]. This study showed that about 500,000 years ago, forest was growing in southern central Greenland and this part of Greenland did not deglaciate even during the Eemian interglacial. These studies indicate that there is no requirement for the organisms themselves to be present for their DNA to be preserved. These methods might even serve to elucidate the palaeoecology in the frozen north back into the middle Pleistocene. The latter is particularly important as it is unclear how well present day analogues will reflect the communities at such distant times, which may cause difficulties in the interpretation of the limited environmental proxies that we have [98].

Pleistocene Paleoeecology in the Future

While our understanding of past environmental change and the corresponding responses of animal populations has improved tremendously, there is still much we do not know. In particular, ancient DNA studies often suffer from an insufficient amount of samples or sequences because certain samples may simply not be available, the majority of samples may not yield DNA (e.g., [47]), or sequence length is limited as ancient DNA is fragmented — this latter problem, however, is somewhat alleviated by the recent introduction of various high-throughput DNA sequencing methods [99]. Due to improved extraction and amplification techniques [100,101] we may also soon be able to use nuclear markers in addition to the mitochondrial DNA used almost exclusively in population studies to date. This will allow us to address male population history, in addition to those of females, and by combining the sexes this may reduce the confidence intervals on estimates of population sizes or the timing of population size changes. Furthermore, using well preserved Pleistocene specimens, complete nuclear genes [102] and a genome [103] have been sequenced. While it is unlikely that complete nuclear genomes will be sequenced for whole populations of extinct animals in the near future, such populations may well become typed for a range of functionally important single nucleotide polymorphisms (SNPs). An example for a candidate SNP for such studies is the position in the melanocortin 1 receptor (MC1R), for which two alleles were found in mammoths, with one effectively knocking out the receptor [102]. As such inactivating mutations usually result in lighter hair colour [104] it is likely that mammoths were polymorphic with regard to hair colour. The obvious question here is whether mammoths varied geographically or temporally in the frequencies of the two variants. Thus, in the future we may be able to investigate changes in genetic

diversity by studying functionally relevant variation in the nuclear genomes and directly observe short-term adaptive responses to changing environments, in addition to analyses of evolutionary neutral loci such as the mitochondrial DNA.

Ancient DNA studies have shown that it is very difficult to precisely predict the responses of individual species to rapid climate change, as each species responds differently. Nevertheless, some general patterns seem to emerge: first, there is a tendency for a loss of genetic diversity and the extinction of populations during times of rapid climate change. And second, at least some populations seem not to be able to follow their habitat during such times. Given the prospect of an extremely rapid, human-induced climate change in the future, we may face another wave of extinctions like the one at the end of the Pleistocene.

Conclusions

Our understanding of the Pleistocene has changed dramatically during recent decades. For example, the Late Pleistocene had been viewed as a time during which the northern latitudes where mostly covered by glaciers or barren tundra vegetation. We have come to see this glaciation as a much more dynamic episode in which climate changed extremely rapidly and on repeated occasions. Genetic studies — especially using ancient DNA — have, in a similar way, changed our view of the varying responses of species during the Pleistocene. We now know that populations are ever-changing units, and the current state of their genetic diversity and its geographical structure represents a mere snapshot in time with a limited predictive value of how things were thousands of years ago. The responses of animal species to environmental change have been much more individualistic than has been generally appreciated. There is hope that by steadily increasing the data sets, and by connecting genetic data with radiocarbon dating and palaeoecological studies, we may eventually understand why a given species reacted in the way that it did and possibly even why some species went extinct while others survived.

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