Chapter 8
Equid Adaptations to Cold Environments

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Abstract Over the past 20 million years from mid-Miocene, the horse family has successfully radiated across the Northern Hemisphere, displaying extraordinary adaptations (including morphological, physiological, behavioural, and genetic) to the cooling world. From early tridactyl horse, like *Hipparion* to modern *Equus*, we review the evolutionary history to reveal how external drivers like climatic and environmental changes contributed to their continually evolving cold adaptation. We also reason that sudden population declines happened after the last glacial period, resulting in a significant recession of equids from the world's cold zones, could have resulted from mixed effects of factors such as temperature rise, and anthropogenic overkill. We group the modern equids by their geographical occurrences across temperature and precipitation gradients, which yields three unique niche clusters—"-hot and arid", "cold and arid", and "cold and moist". Then, we delve into the latter two clusters, and examine the phenotypical and genotypical mechanisms that allow their persistence in the cold parts of the world, and at the cold part of the year. Across the current geographical ranges, equids display traits in active habitat-use strategies, diets, and behaviours to facilitate adaptations to the cold. In addition, we briefly consider the genetics of cold adaptation in equids. Our findings highlight the need for more species-specific studies to be conducted on wild equids, especially on cold adaptation. We also present our thoughts on how the evolutionary history, physiology, and changing climate might impact current and future wild equid conservation efforts.

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H. H. T. Prins, I. J. Gordon (eds.), *The Equids*, Fascinating Life Sciences,
https://doi.org/10.1007/978-3-031-27144-1_8

Horses have accompanied humans and entered every corner of terrestrial Earth, including both the poles (Halsey & Stroud, 2012; Librado et al., 2015). However, their capability in enduring extreme cold has long been ignored. In the legendary Race to the South Pole between Roald Amundsen and Robert Scott, using horse- instead of dog-hauled sleds was considered one of the main causes for Scott's loss of both the race and his team (Huntford, 2010). However, horses might have had little to do with the failure of the expedition, for new findings suggested the adventure was ill-fated after a series of mistakes in logistics and leadership (Halsey & Stroud, 2012; Larson, 2011). In fact, horses, and other extant Equids, are the descendants of once flourishing cold-adapted equines, dating back as early as the mid-Miocene (*ca.* 12 Ma). In this chapter, we examine this ancient family from a new perspective, by illustrating the origin, function, and the implications of cold adaptations in equids, to help understand why this family has been so successful, aside from its speed and endurance.

"*Equus* Is a Cold Adapted Genus": Evolution and Spread into Cold Climes

Climatic change has been one of the major demographic drivers for equids (Lorenzen et al., 2011), and adapting to the ever-cooling Earth has run through the evolutionary history of the equines (Fig. 8.1). To understand the cold adaptation of horses, we should start with the Beringian Land Bridge, or just Beringia, as it was the only passage used by equines from North American (the cradle of the horses) to disperse into Eurasia. This land bridge that is presently submerged, under the waters of the Bering Strait, was already formed towards the end of Cretaceous Period (*ca.* 65 Ma; Brikiatis, 2014), when the temperatures in the Arctic were mild due to high CO_2 concentrations in the atmosphere (Parrish & Spicer, 1988). Alongside two other trans-arctic land bridges (Brikiatis, 2014), Beringia facilitated several early biotic interchanges between Eurasia and North America (Jiang et al., 2019); the "De Geer route" and the "Thulean route" were in operation before the evolution of horse-like

Fig. 8.1 Wild equids. (**a**) Przewalski's horse (photo—courtesy of Amanda Carberry, Columbus Zoo and Aquarium), (**b**) Persian onager (photo—courtesy of Tara Buk, Smithsonian National Zoo and Conservation Biology Institute), (**c**) Kiang (photo—courtesy of Karma Sonam)

mammals (see Brikiatis) and hence we do not consider these here. Since the Miocene, the climate of North America where horse-like ancestors evolved, became increasingly cold and arid, and the world's high latitude areas became hostile for species incapable of living in the cold (Vincent & Berger, 1985). Yet, at this time early Equidae, after 32 million years of morphological stasis, began to rapidly diversify in the open landscapes; many of these species evolved towards larger body sizes (MacFadden, 1986) and dispersed onto other continents. Around 18 Ma, one of the earliest tridactyl horses, the forest-dwelling *Anchitherium,* crossed Beringia and became the first Equidae to successfully disperse into Eurasia (MacFadden, 2001). Seven million years later (*ca*. 11 Ma), during at least two separated dispersal events, the grassland-dweller Hipparionini, also the most successful equine tribe as defined by taxonomic diversity, population size, and biogeographic distribution (Janis & Bernor, 2019; cf. Prins & Gordon, Chap. 1), dispersed into the Old World using the same Beringia route (MacFadden, 1984). Both tridactyl lineages became well established in the Old World and likely attained continuous Holarctic distributions from Western Europe to the east coast of North America (Forsten, 1991; Garcés et al., 1997).

Monodactyl horses appeared much later (McHorse et al., 2019). A paleo-genomic study suggested the most recent common ancestor for all contemporary equids lived 4–4.5 Ma, in North America (Orlando et al., 2013). However, fossil records suggested that early monodactyl horses, such as *Equus simplicidens*, was outnumbered by tridactyl horses in North America and did not radiate or disperse throughout most of the Pliocene (Azzaroli, 1992). However, from the Pleistocene onwards the monodactyl horses quickly radiated and gradually replaced the tridactyls until the latter's extinctions, first in the Americas (ca. 2 Ma) and later in Eurasia and Africa (ca. 1 Ma) (Janis & Bernor, 2019). The story behind the monodactyl horses replacing the tridactyl is complicated (cf. Cantalapiedra et al., Chap. 2), but the possibility of climatic effects cannot be ruled out. Whilst during the Pliocene, the tridactyl horses were still the dominant horse form in the Americas, Eurasia, and Africa, Shotwell (1961) suggested that even before the Pliocene they had already begun to decline in population sizes, diversity, and distribution, possibly due to the increased aridification and cooling. For example, the uplift of the Tibetan Plateau impacted the Asian monsoon cycles and resulted in the continuing aridifications of Asian Inlands (Liu & Dong, 2013), which might have greatly restricted the distribution of tridactyls in Eurasia. In the north of North America, the decline of tridactyl horses left the vacancy in the arid-steppe-like habitats, where the monodactyls were able to survive through early bottlenecks of small initial populations and localized distributions (Janis & Bernor, 2019; Lindsay et al., 1980). At the end of Pliocene (*ca*. 3 Ma), when the world's average temperature dropped to the same level as today (Fig. 8.2), tridactyl equines were further restricted to the coasts of the Mexican Gulf, whilst *Equus* successfully radiated into most parts of North American grasslands. This is because it was deduced that they were more adapted to diets in cold areas and were more efficient at long-distance movement than the tridactyl equines (Janis & Bernor, 2019). During the "elephant-*Equus* event" (*ca*. 2.6 Ma), monodactyl equids finally crossed Beringia and entered Eurasia,

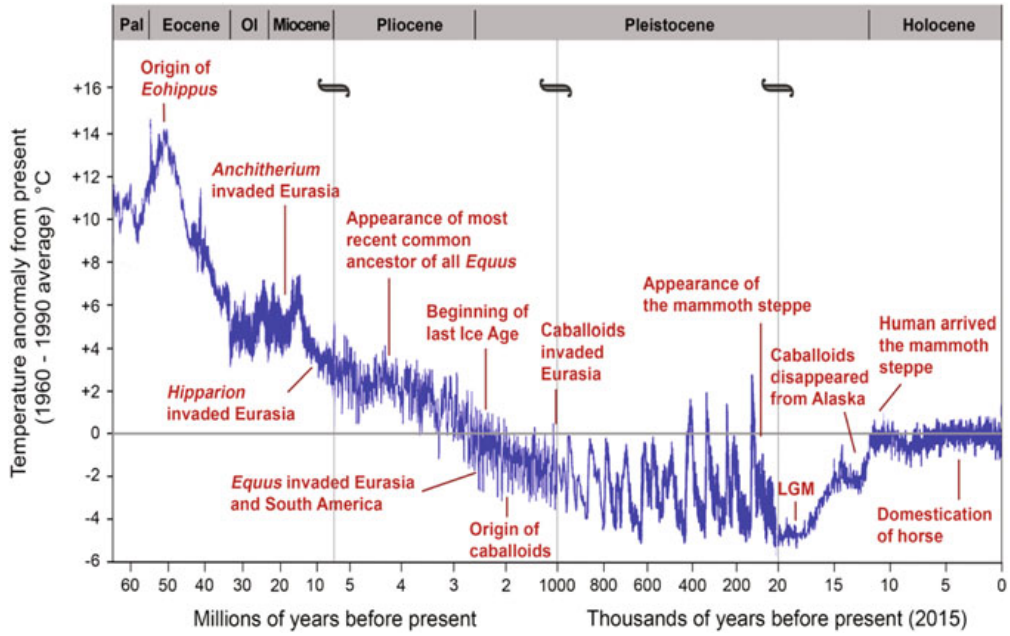

Fig. 8.2 Global temperature anomaly in the Cenozoic era (65 Ma to present), relative to the average temperature in 1960–1990. The prehistorical temperatures are suggested by Hansen et al. (2013), Lisiecki & Raymo (2005), and Lambert et al. (2008). The base plot is modified from Glen Fergus from Wikimedia Commons (https://commons.wikimedia.org/wiki/File:All_palaeotemps.png). Note the timescale on the *X* axis changes four times

where the aridification and cooling had started a long period ago due to changed monsoon cycles. By being "pre-adapted" to the arid and cold conditions in North America (Azzaroli, 1983), the radiation of equids in Eurasia and later Africa, as evidenced by phylogenomics, was extremely fast (Vilstrup et al., 2013). It did not take long for equids to spread into the Indian Subcontinent and Africa (Bernor et al., 2019). This marked the beginning of the equids' golden era (cf. Prins & Gordon, Chap. 1). Since then, equids occur in most fossil faunas and are often the most abundant group by number, if not by taxa (Forsten, 1988; Bernor et al., 2019). One of the early monodactyls, *E. stenonis*, became the most widespread wild equid by successfully colonizing the Holarctic realms (Azzaroli, 2000; Forsten, 1999). Despite the fact that modern-day stenonids, comprising Zebras and Asses, are found only in today's tropical and temperate zones (McGrew, 1944), the early stenonids demonstrated adaptations to cooling and aridification (Janis & Bernor, 2019) and successfully radiated in the world's high latitude areas before the rise of the caballoid horses.

Modern *Equus* in the Ice Age

At the beginning of the Pleistocene epoch 2.5 Ma, the Earth entered its latest Ice Age, as glaciers covered most high latitude areas (Shackleton, 1967). The average annual temperature was six degrees colder than at present (Fig. 8.1), while glaciation caused recession of forests as well as expansion of grasslands in North America. Meanwhile, species of the genus *Equus* flourished and greatly expanded their range. So far, at least 40 valid *Equus* species had been identified from Pleistocene, out of which more than 27 species survived into the late Pleistocene or even early Holocene (Azzaroli, 1992; this high number of species may, however, be an exaggeration. See discussion in Prins & Gordon, Chap. 1 and references therein). Modern-day Asses and Zebras also appeared around the mid-Pleistocene. Molecular evidence dates their divergence to around 2 Ma (Jónsson et al., 2014), but the exact place of origin is still unclear. According to cranial anatomy, asses and zebras could have evolved independently in Eurasia and Africa, respectively, while the former migrated to Africa in more recent times (Azzaroli, 1992). A genomic study has found frequent hybridizations between the two clades, indicating incomplete reproductive isolation (Jónsson et al., 2014). A new paper reexamined the anatomic records and suggested both clades evolved from *E. koobiforensis*, found in East Africa (Cirilli et al., 2021), from where wild Asses emerged and spread into Eurasia (*ca*. 1.7 Ma). Around 2 Ma, the "dentally more advanced" caballoid horses, or the "true horses", appeared on the North American grasslands. These species likely descended from the New World stenonid lineage *E. idahoensis* (Azzaroli, 1992). Compared to ancestral forms, the caballoid horses are considered adaptatively more tolerant of extreme cold (Bennett & Hoffmann, 1999). For most of Pleistocene North America was still dominated by stenonids (McGrew, 1944), however, the caballoids managed to spread into a variety of habitats from semi-desert to steppe-tundra (Bennett & Hoffmann, 1999). They also dispersed into Eurasia about 1 Ma, and since the end-Villafranchian mammalian fauna turn-over (*ca*. 1–0.9 Ma), had become the dominant equid in the central and northern parts of Eurasia (Azzaroli, 1983).

Towards the end of the last glacial period (*ca*. 26–20 Ka), the glaciation on Earth reached its maximum extent (Last Glacial Maximum or LGM; Fig. 8.2). As a massive amount of water was locked in glaciers, sea level dropped over 130–135 m below the present (Yokoyama et al., 2000), exposing Beringia to its maximum extent (Fig. 8.3). At that time, the world's most extensive biome, the Mammoth Steppe, appeared in the Holarctic zone, spanning from Western Europe to Northern Canada, and from the Arctic islands to Northern China. As demonstrated by its rich fossil deposits, this transcontinental steppe-tundra ecosystem once held higher plant productivity and animal biomass, than is seen in the modern Holarctic (Guthrie, 1982; Prins, 1998; Zimov et al., 2012).

The emergence of the Mammoth Steppe could have been caused by both top-down and bottom-up processes. Guthrie (2001) suggested that the Mammoth Steppe remained ice-free during the LGM because was of local air circulation, which was not fed by water vapour from nearby seas. Moreover, "keystone herbivores",

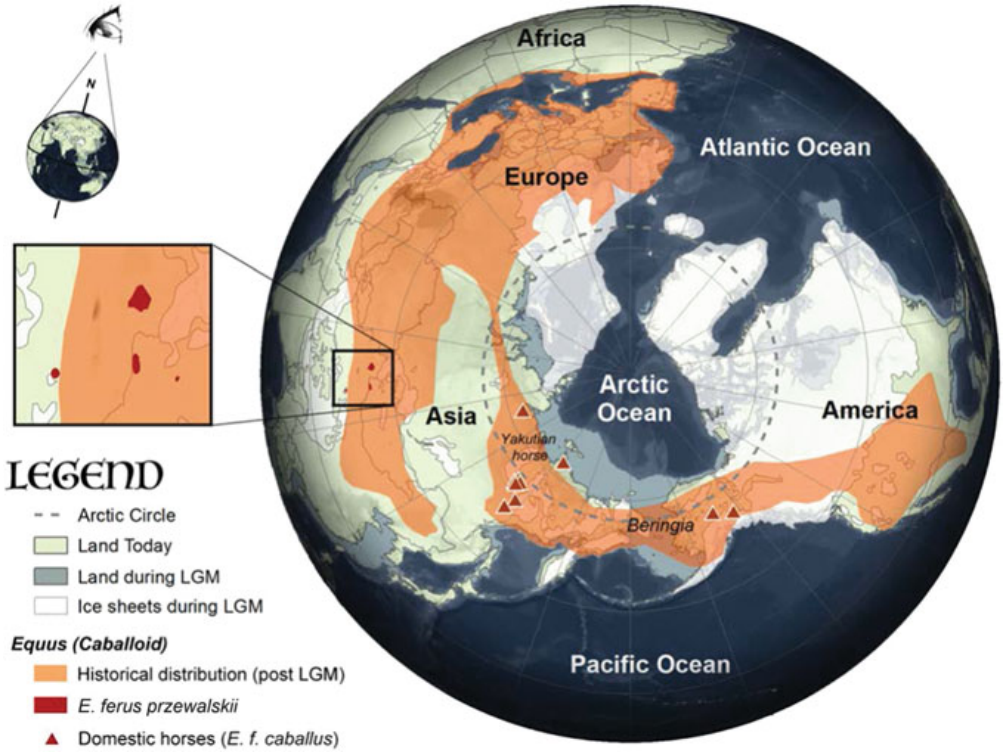

Fig. 8.3 The Holarctic distributions of the caballoid equids (*Equus ferus*) after the last glacial maximum (LGM) and their current distributions (Przewalski's horse and two domestic horse breeds). The gap in the distribution to the East and West of Lake Baikal (where "Asia" is written) is not understood yet and may be an artefact. The distribution of wild *E. ferus* (see Fig. 8.2) reached its maximum extent by the late Pleistocene, 15–9.5 Ka, when melting glaciers exposed more land while the Bering land bridge still existed (Bennett & Hoffmann, 1999). The ice sheets (white and transparent over sea in this figure; adopted from Clark et al. (2009)), was in fact during LGM (26–20 Ka), before the deglaciation commenced in the Northern Hemisphere. Following United States Geological Survey (USGS)'s estimation of global sea level during LGM, we calculated the prehistoric land area during LGM (pale green in this figure) with a 130 m drop of sea level from the current (light green area) on GTOPO30 digital elevation model (https://rda.ucar.edu/datasets/ds758.0/). The extant distributions of caballoids were downloaded from IUCN Red List (https://www.iucnredlist.org/). We also included cold-adapted breeds of the domestic horse (*E. f. caballus*; red triangles in this figure), Yakutian horses (Boeskorov et al., 2018; Librado et al., 2015), and feral horses in Alaska (Guthrie & Stoker, 1990)

including equids, woolly mammoth (*Mammuthus primigenius*), and steppe bison (*Bison priscus*), are thought to have maintained the steppe-like ecosystem by controlling the domination of the vegetation by woody species (Owen-Smith, 1987; Zimov et al., 2012). Notably, the caballoid horse was the second most dominant species on the Mammoth Steppe after bison (Guthrie, 2013), and radiated into at least seven subspecies (Bennett & Hoffmann, 1999). As Fig. 8.3 shows, the caballoids once successfully occupied suitable periglacial habitats in as nearly a circumpolar distribution as glacial masses would allow (Guthrie, 2013). They had been common in Europe until Upper Palaeolithic (early Holocene, ca. 12 Ka;

Bendrey, 2012). In the south, for much of China was unaffected by Quaternary glaciations, the *Equus* had been one of the most abundant and persistent genera in Northern China since early Pleistocene (Yang et al., 2020). In the very north, caballoid horses became the commonest megafauna in areas like the North Slopes of Alaska, where climatic conditions were deemed too harsh for large mammals, even bisons (Guthrie & Stoker, 1990).

The LGM also marked the expansion of modern stenonids. Using genomic data, Jónsson et al. (2014) analysed the ancient equid demography during the LGM and found the populations of both Burchell's zebra (*E. burchellii* a.k.a. *E. quagga*) and wild ass or kulan (*E. hemionus*; a.k.a. onager) showed rapid recoveries from the end-Villafranchian crashes of the stenonids. Although being inferior to caballoids in numbers, the stenonid equids were commonly found on the Mammoth Steppe (Guthrie, 1982), often occurring sympatrically with the caballoids (Forsten, 1988). As shown in Fig. 8.4, the prehistoric distribution of *E. hemionus* stretched from the Mammoth Steppe south to the Indian Subcontinent, Arabian Peninsula, and Mediterranean Coast, as well as from Western Europe all the way to China, a vast area even comparable to once-flourished *E. stenonis* in the Old World. Furthermore, a sister taxon, *E. kiang*, split from *E. hemionus* around 0.75 Ma (Jónsson et al., 2014), and later became geographically isolated on Tibetan Plateau, where they endured on arid steppes and through cold winters, similar to those found on the Mammoth Steppe. In East Africa, *Equus* had been one of the most excavated large mammal specimens from about 2 Ma, and once made up over 44% of the large mammals in areas dominated by C_4 grassland (Bernor et al., 2019).

Decline and Destruction in the Cold Zones

Like tides going in and out, as the globe has cooled down since Miocene, members of Equidae family, including but not limited to tridactyl *Anchitherium*, *Hipparion*, and monodactyl stenonid and caballoid equids, have succeeded each other in the world's high latitude areas (see previous section). The successors were, to some degrees, more "progressed" in their cold adaptations than were the predecessors. Many factors, e.g., disease, predation, competitive exclusion, and climate change, might have contributed to the disappearance of a species, but a sudden crash or mass extinction event was likely the result of one major driver (Ceballos et al., 2015). Interestingly, equines usually showed signs of decline in diversity, population size, distribution, and/or even body size before their final disappearance. For example, for over 2 million years, the tridactyls had undergone a continuous decline in population size, diversity, and distribution prior to monodactyl horses' appearance in Eurasia and Africa (Shotwell, 1961). However, the equids experienced at least two sudden continent-level population crashes. The first happened during the end-Villafranchian mammalian fauna turn-over (*ca*. 1–0.9 Ma). Along with the disappearance of over 50% of Late Villafranchian mammals, the once-dominant stenonid equids in Eurasia were suddenly replaced by the caballoids (Azzaroli, 1983). The second crash more

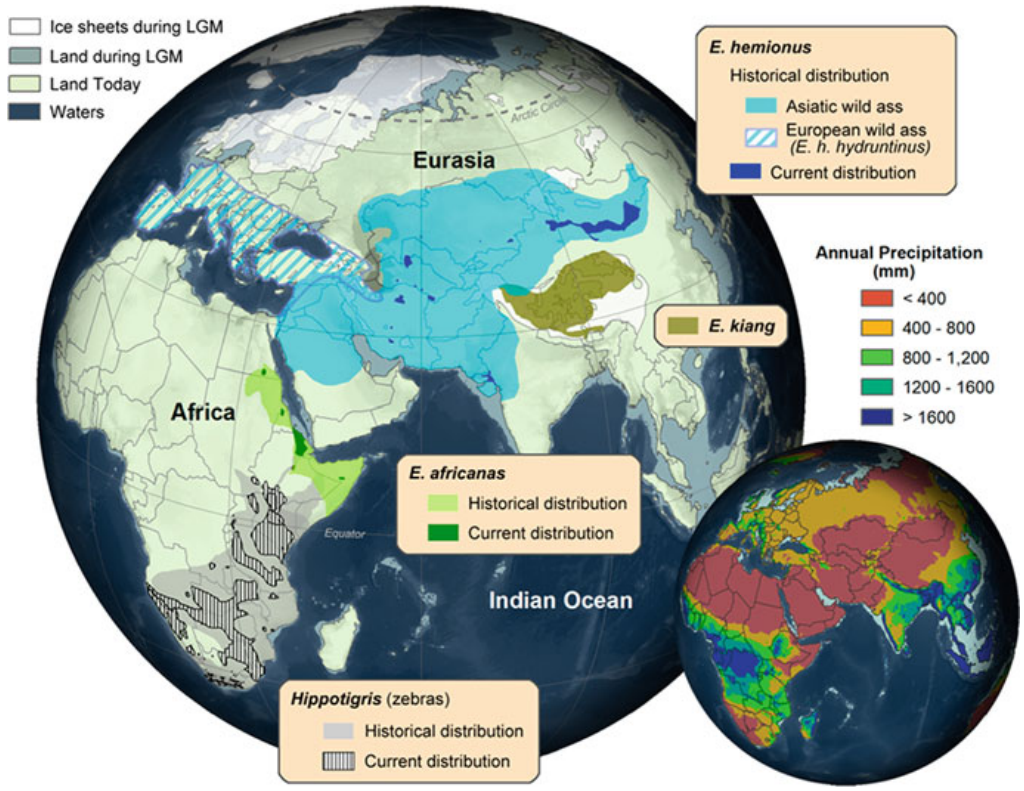

Fig. 8.4 Historical and current distributions of stenonid equids in the world. Historical range of *E. hemionus* in Asia (blue/transparent) was based on Feh et al. (2002), which likely reflected the species distribution in the Early- or Mid-Holocene. Distribution of European Wild asses *E. hemionus hydruntinus* (blue/hatch) during LGM was portrayed according to Crees and Turvey (2014), as well as Bennett et al. (2017). Moreover, historic range of *E. africanus* (green/transparent) was based on Moehlman (2002). Besides, we combined all *Hippotigris* (zebras), for their insignificance in cold adaptation, and their historical distributions (grey/transparent) were digitized from Hack et al. (2002), Williams (2002), and Novellie et al. (2002). The extant equid distributions were downloaded from IUCN Red List (https://www.iucnredlist.org/). Lastly, to demonstrate the distribution of the world's arid landscapes, the global annual precipitation (between 1950 and 2000) was retrieved from the WorldClim database (http://www.worldclim.org/current), and illustrated in the mini map

recent but more catastrophic occurred in the Late Pleistocene (*ca*. 25 Ka), as over 99% of the equid populations vanished in a few thousand years (Orlando et al., 2013). This event also marked the grand recession of equids from the world's cold zones.

The Late Pleistocene decline of equid population happened during the peak of a series of climate-induced population dynamics of equids. As Fig. 8.2 shows, the Pleistocene world was subjected to repeated cycles of glaciations and deglaciations (Azzaroli, 1983; Ehlers et al., 2004) that will have posed enormous challenges to species with specific niche requirements. These were likely to have been the exterior driver of the stenonids' sudden decline during the end-Villafranchian mammalian turn-over. In the last glaciation period (LGP), both the glaciers and the Mammoth

Steppe ecosystem reached maximum coverages, while the distribution of equids also achieved new heights. However, the LGM was followed by a rapid global warming (Fig. 8.2), which led to strong deglaciations and a substantial rise in sea level. The Mammoth Steppe ecosystem, after receiving excessive moisture and heat, quickly collapsed and gave ways to tundra, mossy forests, and shallow seas (Prins, 1998; Zimov et al., 2012). At roughly the same time, the most recent extinction event, the Late Quaternary Extinction (LQE) occurred, during which most large-bodied mammals (> 50 kg) outside Africa went extinct (see Koch & Barnosky, 2006), with equids just one of the unfortunate victims. By the late Pleistocene, North America only had two equid species left, the caballoid *E. ferus* and a native "stilt-legged" horse (Weinstock et al., 2005). In Eurasia, two large forms of caballoids, likely to be subspecies, went extinct, and only the smallest one survived, which later gave rise to modern true horses (Forsten, 1988). Even though the stenonids had been in decline since the Pleistocene (Forsten, 1988), the LQE further reduced their distributions in Eurasia. As Fig. 8.4 shows, whereas the distributions of *Hippotigris* (Zebras) were more or less constrained to Africa, the subgenus *Asinus* (Asses) was greatly impacted by the LQE, especially in Eurasia. The Asiatic wild ass had receded from most of its historical distributions in the Mammoth Steppe, Arabian Peninsula, and Indian Subcontinent, and only remained in highly fragmented desert habitats (Feh et al., 2002). The European wild ass, likely a subspecies of *E. hemionus* (based on molecular evidence; Bennett et al., 2017), once widespread in Late Pleistocene, became a rare component after LGM, and only persisted in a few isolated, fragmented LQE refugia like the Iberian, Italian, and Balkan peninsulas until their global extinction around 580 BC (Crees & Turvey, 2014). However, the other Asian ass, *E. kiang*, was an exception. They even spread north into Mongolia and interbred with *E. hemionus* (Bennett et al., 2017).

"Climate or Human": What Caused the Decline of Equids?

Although the post-LGM global warming chronologically overlapped with the LQE (Orlando et al., 2013), the real driver for the late Pleistocene decline of equids remains far from certain (Koch & Barnosky, 2006). Firstly, Raymo (1994) suggested that the last deglaciation was similar as previous ones in terms of speed and magnitude. Even the LGM, while severe, did not exceed several prior glacial advances. Secondly, although climate change could result in habitat loss, and structural and phenological changes in plant communities, these effects were usually local and cannot explain the simultaneous mega-extinctions across many continents (Koch & Barnosky, 2006). Thirdly, the LQE "victims" were predominantly large-bodied, slow breeding mammalian species, which was unusual as compared to previous extinction events (Barnosky et al., 2004). Lastly, the precipitous loss of megafauna, across several continents, followed the dispersal patterns of modern humans (Sandom et al., 2014) but vaguely so. These last two pieces of information have been used to imply that the sudden arrival of a "super-predator", i.e., *Homo*

sapiens, had caused the selective disappearance of megafauna, including *Equus*, in the Holarctic.

Today, by comparing the megafauna extinction rates with the human arrival time at continental and country levels, several studies have concluded that the anthropogenic factors strongly, if not completely, contributed to the extinctions of large-bodied mammals during the LQE, although noting that anthropogenically induced extinctions differed by species, time, space, and magnitude (Prescott et al., 2012; Sandom et al., 2014). This so-named overkill hypothesis was used to explain why the LQE was more severe in America and Australia, where naïve animals lacked behavioural and evolutionary adaptations to escape human predation, while in Africa and Eurasia where *H. sapiens* and other hominids had coevolved with the local megafauna, the magnitude of the extinction events was much lower (Lorenzen et al., 2011). It also explains why equids were seldom found in American archaeological sites (Azzaroli, 1992), but were common in European and Siberian sites, making up 58 and 66% of megafauna herbivores, respectively (Lorenzen et al., 2011). The naïve prey in the New World had neither the experience nor the time to adapt to the hunting techniques of modern human, so the killing turned into a massacre, just like "Blitzkrieg", where a large proportion of prey were unused or only partially consumed (Martin, 1984). This "explanation" is based on rather broad interpretations of first arrival of humans and extinction events, and because concepts as "co-evolution" are shaky too, it appears as if this "overkill hypothesis" is less useful as was thought earlier. Unlike historical extinctions on islands or contemporary anthropogenic extinctions, early hunter-gatherer societies, such as the Clovis Culture, might have lacked the manpower and skills to carry out "Blitzkrieg" style killing over the continental scale (Koch & Barnosky, 2006). New evidence even suggested that the caballoid horses might have existed in in Yokon in North America until 5 Ka (Murchie et al., 2021), much later than human settlement in the area (Hoffecker et al., 1993). Moreover, fossil evidence indicates body size of horses, and many other megafauna mammals, had shrunk dramatically, long before the LQE, with possible causes of local warming and major vegetation shifts (Guthrie, 2003; Prins et al., in press).

There were other extinction hypotheses that require the confluence of climatic or human factors. For example, the "keystone herbivore" hypothesis (Owen-Smith, 1987) proposed that large-sized herbivores like equids, Woolly mammoth, and Steppe bison engineered the Holarctic landscape and promoted niche partitioning among both herbivores and carnivores, in much the same way as Zebras and African elephants are postulated to do on the woody savanna today. Once their numbers dropped below a certain threshold, for either climatic or anthropogenic reasons, the community structure would lose its equilibrium and potentially slide into another state, during which negative impacts would cascade onto other species (Murchie et al., 2021). Yet, the "community equilibrium" hypothesis needs more direct evidence, as species interactions were rarely observed from fossil records (Louys et al., 2012). Another combined hypothesis argues that climate-induced habitat loss intensified interspecific competition between Late Pleistocene horses and sympatric species with similar dietary choices, such as Wild yak (*Bos mutus*) and American

bison (*Bison bison*) (Fox-Dobbs et al., 2008). Horses, as more obligate grazers (Guthrie, 2003; Guthrie & Stoker, 1990; de Jong & Prins, Chap. 4), were more susceptible to sudden habitat or vegetation shifts than were less obligate grazing species of bovids and mammoths and became the first large herbivore to go extinct on the Mammoth Steppe (Guthrie, 2003). However, new evidence points into the direction that wild horses, as well as Asian wild asses browse much more than previously recognized (Cao, unpubl. data; also see references in Prins & Gordon, Chap. 1), and hence this "explanation" may not hold either. In general, the combined hypotheses require the confluence of climatic, ecological, and anthropogenic factors (Koch & Barnosky, 2006; Lorenzen et al., 2011; Prescott et al., 2012), while the causes of extinction, instead of a universal pattern, are likely to have varied by species and regions across time (Lorenzen et al., 2011; Sandom et al., 2014). The thing we know for certain, however, is that at least a number of *Equus* species showed very good adaptation to live under cold and dry conditions.

The decline of wild equids was still far from ending in Holocene. Around 4 Ka, human successfully domesticated caballoid horses in Central Asia (Outram et al., 2009; cf. Bendrey & Oakes, Chap. 14). Under the pressure from domesticated conspecifics, wild caballoids were quickly exterminated from most of their prehistoric areas (Fig. 8.3; Bennett & Hoffmann, 1999). Przewalski's horse, likely to have been the last wild caballoid, went extinct in the Dzungarian Gobi around the 1960s (Bouman & Bouman, 1994).

Evaluating the Cold-Adapted Niches of Extant Equids

The niche theory is the corner stone of ecology. In his widely accepted definition, G. Evelyn Hutchinson proposed that a species' niche could be characterized as the multidimensional hypervolume, where the axes "correspond to a state of the environment that would permit the species to exist indefinitely" (Hutchinson, 1957). Here we illustrate the Hutchinsonian persistence niches of seven extant equids, in terms of their adaptions to coldness, using geographical distributions and associated climate data (Fig. 8.5). As well as temperature, precipitation was adopted as a niche vector for their strong coupling effects in ecology (White et al., 1997). As we can see, *E. hemionus* spreads widely along the axis of temperature from –18 to –1 °C in the coldest quarter of the year. *E. kiang* also shows considerable tolerance of temperatures (–17 to –11 °C) and aridity (0–45 mm precipitation) in winter. Yet, it was interesting to reflect on the "cold-adapted niche" of wild caballoid horses, since this equid with its past nearly circumpolar distribution went extinct from the Holarctic steppe and the rest of the world (Azzaroli, 1992; Bouman & Bouman, 1994) probably due to hunting and competition with domestic horses; the sole extant member, i.e., Przewalski's horse, has a much narrower range of temperature tolerance (Wakefield et al., 2002). To overcome this challenge, our study includes two semi-wild breeds of caballoid horses (*E. f. caballus*), Yakutian and Alaskan horses, which can survive the Arctic winter without artificial supplements of food or shelter

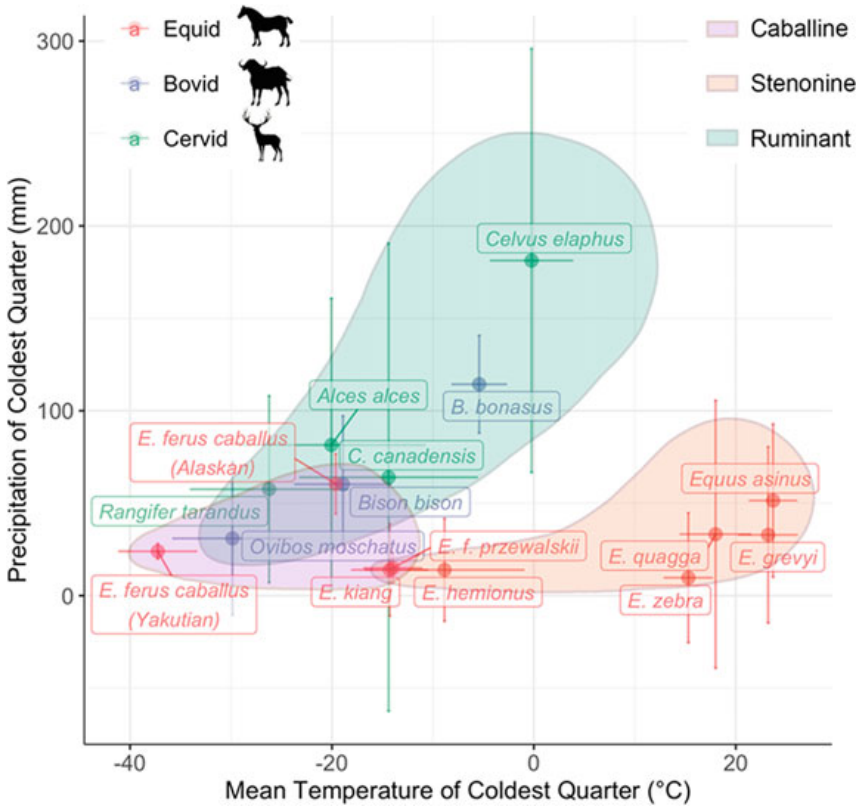

Fig. 8.5 Comparisons of ecological niches on cold and dry adaptations among three large ungulate tribes: equids, bovids, and cervids. All seven extant equids and six large cold-living ungulates from Bovidae and Cervidae were chosen, and their current distributions are downloaded from IUCN Red List (https://www.iucnredlist.org/). The cold-adaptive domestic horse breeds (*Equus ferus caballus*), Yakutian and Alaskan, are also included. To extract the climate data, we generate 9999 random points within distribution polygons of each species (for wild ungulates) or just used the actual location of study area (for domestic horses). Then, these points are overlaid in the geographic information system (GIS) with the climate raster layers from the WorldClim database, which provides a series of bioclimate related parameters at 3″ arc second resolution for the period 1950–2000 (http://www.worldclim.org/current). The average temperature and precipitation in the coldest quarter ("season") were selected from the database to be the two niche axes. We use the extract function from raster package in R statistic environment, and plotted both the mean and standard deviation (scatters and horizontal/vertical bars) for each species. In the end, the niche spaces are visually drawn for the following taxa: caballoid horses (purple), stenonid equids (zebras and asses; orange), as well as the ruminants (green)

(Boeskorov et al., 2018; Guthrie & Stoker, 1990; Librado et al., 2015; see Cao et al., Chap. 8). Moreover, to quantify potential niche overlaps, we incorporated a few large ungulates that since late Pleistocene had, or still have, Holarctic distributions, i.e., the American and European bison (*Bison bison* and *B. bonasus, respectively*), reindeer/caribou (*Rangifer tarandus*), Red deer (*Cervus elaphus*), elk (*C. canadensis* a.k.a. wapiti, *C. elaphus*), moose (a.ka., elk, *Alces alces*), and musk ox (*Ovibos moschatus*). These species, which happen to be ruminants, were either known to

co-occur with equids in the Holarctic (Guthrie, 1982), in the past or present, or have a comparable body size, distribution, habitat preference, and adaptation to coldness.

In the two-dimensional niche space, the fundamental niches are largely non-overlapping, both between equids and ruminants, or between caballoid and stenonid equids (Fig. 8.5). Comparing to cold-adapted ruminants, modern equids are well adapted to arid conditions (Rubenstein et al., 2016), as they mostly occurred in the world's arid zones with less than 50 mm precipitation in the coldest quarter ("season"). Along the temperature gradient, they aggregated into three unique niche clusters: the "hot and arid" cluster, with all African equids; the "cold and arid" cluster, including all hominoids; and the "cold and moist" cluster, consisting of all caballoid horses (Fig. 8.5). Modern stenonids, that consist of six out of seven extant equid "species" and make up the first two niche clusters, are still very successful in Africa and parts of Eurasia. Considering that the caballoid horse also likely diverged from an American stenonid, this old lineage demonstrated wide tolerance of environmental conditions, indicating repeated, opportunistic speciation (Forsten, 1988). Moreover, stenonid species were often found coexisting sympatrically. At least in Early and Middle Pleistocene Europe, there were typically two different-sized stenonids coexisting, with the smaller species often found to be more abundant, more associated with open landscapes, and more of an obligate grazer (Saarinen et al., 2021). At present, stenonids still occurred sympatrically in some parts of Africa (Forsten, 1988). Since the arrival of caballoid horses in Eurasia, however, the large- and medium-sized stenonids have been displaced (Forsten, 1988). The small-bodied species, sometimes coexist with caballoids (*E. hemionus* and *E. ferus przewalskii* in the Dzungarian Gobi) but are limited by the deep snow in high latitude areas, likely due to their small, cupped hooves which are more suitable for irregular, hard-substrate terrain (Guthrie, 1982).

In Fig. 8.5, whereas the "hot and arid" cluster occupies the tropical zone, and was largely unaffected by severe climate change, the "cold and arid" cluster has had its niche restricted on the axis of aridity. As shown in Fig. 8.4, the distributions of the cold and arid cluster more or less overlapped with the arid zones of Eurasia, where they, with a series of behavioural and physiological adaptations (Rubenstein, 1989), were able to survive on limited water supply. Yet, the heminoids once occupied the humid and warm areas of Southern Europe (Crees & Turvey, 2014) (cf. Prins & Gordon, Chap. 1), suggesting that the current realized niche on the aridity axis could be the result of climatic and anthropogenic disturbances (see the previous section). Besides, in accordance with their historical distributions, they should have a much wider fundamental niche on the temperature gradient, but the geographic span of the Eurasian arid zone likely limited their performance on cold adaptations.

The "cold and moist" cluster, comprising the modern caballoids, even though being relatively relaxed on the axis of aridity, was capable of surviving the extreme cold. How the caballoids adapt to the extreme cold will be discussed in the next sections, but it is obvious that their larger body size, compared to many of their predecessors, provides better chances of surviving in the Arctic (McNab, 1971). Moreover, caballoids belong to a monotypic subgenus *Equus* (under the genus *Equus*) and are phenotypically homogeneous (except for varied body sizes), but

they demonstrate strong "eurytopicity", meaning that they could live in a wide variety of habitats and tolerate a wide range of environmental conditions. Forsten (1988) suggested the caballoid horses (subspecies) lack of sympatricity with conspecifics was due to their eurytopicity. In contrast, the stenonids are rather "stenotopic", the opposite term of eurytopic, and their adaptations to ecological conditions are largely species-specific (Klingel, 1977; Rubenstein et al., 2016), likely due to the much longer period of species diversification (Forsten, 1988). For example, *E. kiang* is well adapted to the unique alpine steppe environment on the Tibetan Plateau, but is not found elsewhere (Bhatnagar et al., 2006).

Whilst the caballines with flexible niche were able to dominate in a variety of Holarctic habitats, such as grasslands, open forests, and tundra, their colonization of the dry steppes and deserts might have been hampered by their water dependency, as studies on Przewalski's horse in the deserts suggested (Kaczensky et al., 2008; Zhang et al., 2015; but see Sneddon, Chap. 9). As Fig. 8.5 illustrates, the niche spaces of caballines and stenonines overlap slightly where *E. f. przewalskii* is situated. In the real world, the Dzungarian Gobi is where Przewalski's horse and Asiatic wild ass are sympatric. Despite seldom coexisted with conspecifics, caballoid horses were able to coexist with stenonids, at least on the once-existed Mammoth Steppe during LGM (Forsten, 1988). Yet, today's Dzungarian Gobi is much drier than the Mammoth Steppe (Zhang et al., 2017), and thus might not support a rich grazing megafauna community nor facilitate niche differentiations like the latter. Especially, Przewalski's horse is only discovered in this desert area, but also went extinct in the wild not soon after discovery, so whether the Gobi was part of Przewalski's horse's core habitat or just a refuge had been long debated (Wakefield et al., 2002). But like many Holocene extinctions, the anthropogenic and political factors could not be ruled out (Bouman & Bouman, 1994). Today we are still not very clear how this caballoid horse survived the extreme aridity and remained sympatric with stenonids. Zhang et al. (2015) suggested that Przewalski's horse adopted alternative drinking rhythm and agonistic behaviours to allow its coexistence with asses. Thus, the Przewalski's horse's niche space demonstrates an extreme case of caballoid horses surviving under xeric conditions.

The cold-adaptive niche of caballoid horses, as depicted in Fig. 8.5, is largely contributed by two domestic horse breeds with outstanding cold-adaption, from which we can speculate about ecological mechanisms that may have helped their wild ancestors overcome the challenges of living in the cold. As shown in Fig. 8.5, their winter niche overlaps with that of several cold-adapted ruminants, including cervids like reindeer, moose (= elk), and elk (= wapiti), as well as bovids like the American bison and muskox, in the "cold and moist" niche space. In particular, the Alaskan horses occur in the relatively "humid" part of the Arctic, alongside several ruminants. Whereas these ruminants, except bison, are either browsers or mix-feeders, which can feed on the rich woody vegetation in this region, horses are predominantly grazers, even more so than are bison (Fox-Dobbs et al., 2008; cf. de Jong & Prins, Chap. 4), so deep snow can be devastating. Guthrie (1982) suggested that although equids are good at cratering in the snow for grass, they cannot tolerate deep snow for lengthy periods. Hence, he suggested that in order to

forage on grasses underneath snow, modern caballoids, such as semi-wild Alaskan horses, had to look for areas with strong winds that removed deep snow (Guthrie & Stoker, 1990). Of course, East Siberia is not categorized by deep snow (to the contrary) but *dzuds* or *zuds* (iced-over snow events) (e.g., Rao et al., 2015; Sternberg, 2018) may cause havoc with grazing mammals. Back in the LGP, however, equids on the Mammoth Steppe did not require the snow removal by wind since the ground was largely snowless due to the climatic aridity (see previous sections). Similarly, the Yakutian horses might be experiencing the similar conditions as their Late Pleistocene predecessors. Yakutia, as the coldest place in Northern Hemisphere (Nazarova et al., 2013), happens to be located on the driest part of the Holarctic zone (Fig. 8.6), with only 20 mm precipitation in winter (Fig. 8.5). The East Siberian Mountains, spanning the most part of the region, also provides shelter from storms and deep snow. Moreover, equids' hindgut digestion, with its relatively short gut transit time (Gordon & Prins, Chap. 6; Janis, 1976), allows for the consumption of rough, dried vegetation during winter including browse. Due to their outstanding cold adaptations, the Yakutian horses were once thought to be the descendants of the native caballoid population that had lived in Yakutia since the LGP. However, a recent genomic study indicated that the breed actually came from a group of rapidly mutating domestic horses that were introduced by Yakut people just a few centuries ago (Librado et al., 2015). Although selective breeding was involved, in terms of adaptions to extreme cold, aridity, rough terrain, and coarse vegetation, the Yakutian horse breed might still be the best candidate to resemble the caballoid horses' niche on the Mammoth Steppe (Fig. 8.7).

Habitat, Diets, and Behaviour of Cold-Adapted Equids

In this section, we briefly discuss relevant aspects of habitat use, diets, behaviour, and movements of Przewalski's horse, kiang and kulan, and how these may contribute to their adaptations to the cold.

The three wild, cold-adapted equids, occur in the vast plains and rolling mountains of Central Asia and the Tibetan Plateau (Schoenecker et al., 2016). Compared to other extant equids, these cold-adapted species occur in the range of low annual mean temperatures and high aridity (see above; Schulz & Kaiser, 2013). They often show long-distance movements to avoid inclement weather, gain access to water, and exploit better forage resources (Schoenecker et al., 2016). Resource selection in arid-adapted equids, that includes the three cold-adapted species, is primarily governed by forage availability (Henley et al. 2007; Schoenecker et al., 2016), and forage abundance is often the best predictor of habitat use by equids (King & Gurnell, 2005; St-Louis & Côté, 2014).

The Przewalski's horse primarily uses steppe and semi-desert habitats, only using forests occasionally (King & Gurnell, 2005; Van Dierendonck & de Vries, 1996). Kiangs occur in the high-altitude Tibet-Xinjiang plateau and adjacent parts of Himalaya, mostly above 4000 m (Schaller, 1998; Shah et al., 2015). In a region

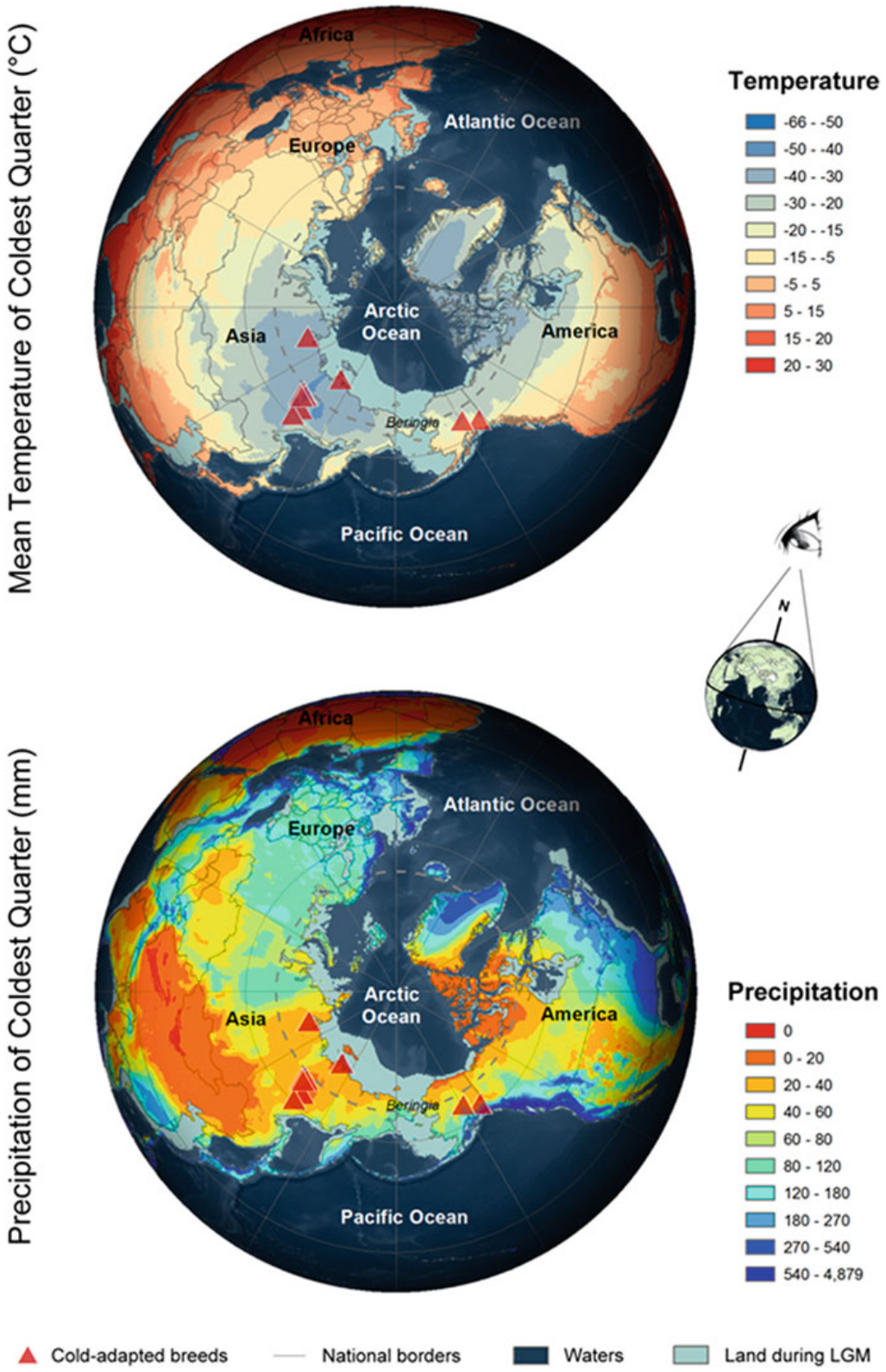

Fig. 8.6 Distributions of the Yakutian horse and Alaskan horse breed in relation to mean temperature and precipitation of the coldest quarter of the year. Yakutian horses were recorded on the Oymyakon Plateau as well as near the shore of Arctic Ocean (Boeskorov et al., 2018; Librado et al.,

with low plant biomass (Namgail et al., 2012), kiang uses sites where forage is relatively more abundant in the vast high-altitude plains, alpine meadows, desert steppes, broad valleys, and hills (Harris & Miller, 1995; Schaller, 1998, Shah, 2002; St-Louis and Côté, 2014), and use the mesic moist sedge meadows in the river and lake basins disproportionately more during late summer (Bhatnagar et al. 2006; Schaller, 1998) (Fig. 8.8). The Mongolian kulan is primarily distributed across deserts, grasslands, semi-deserts, and the desert steppe (Kaczensky et al., 2020; Zhang et al., 2020). In summer, Przewalski's horses (King, 2002; Wang et al., 2012) and Khulan (Feh et al., 2001) seemed to avoid harassment from insects by selecting for cooler sites during the warmer part of the day.

Organisms require a mix of quantity and quality of food and water, shelter from climatic extremes, and security from predators to survive, and extreme cold influences the choices species can make. The areas inhabited by the three equid species is extremely cold through the long winters (Figs. 8.3 and 8.4), leaving the short summer season for forage production to take place. Given the cold climate and aridity of continental areas, plant production is often relatively low, although plants may seasonally have relatively high levels of nutrition (Uniyal et al., 2005). The mostly open terrain provides few possibilities to shelter from blizzards, and sparse and unpredictable surface water may remain frozen for a considerable part of the year, making it difficult to drink. On the other hand, cryo-frozen forage in autumn may provide nutrition even in the cold season (Petrov et al., 2016), and widespread snow may allow animals to obtain water without the need to travel long distances to access liquid water (Payne et al., 2020).

The equids are hindgut fermenters that have a fast rate of food passage through the gut and thus rely on high intake of even low quality (with high silica and cellulose) but abundant food that can also be digested in their enlarged caeca and colon with commensal bacteria (Gordon & Prins, Chap. 6; Soest & Peter, 1994). Thus, these modern equids can be relatively unselective in their foraging as compared to similar sized ruminants (Kuntz et al., 2006; Schoenecker et al., 2016). The high-crowned teeth and flexible lips are also well suited for processing tough forage, which is the case during much of the year in arid, cold areas. The generally available graminoids in the cold steppes and mountains are thus the foods for these modern, northern equids, seasonally foraging on some forbs and browse as well (Kaczensky et al., 2008; Schaller, 1998; Shah & Qureshi, 2007). Equids use habitats as per their availability of forage that can include the most productive areas (Henley et al., 2007), and forage abundance is often the best predictor of use by equids (King & Gurnell, 2005; St-Louis & Côté, 2014). While equids use habitats with relatively abundant, coarse forage, at the scale of the site, they seem to try to maximize nutrient intake; In summer, kiangs may select green grasses within foraging sites (St-Louis & Côté, 2014), whilst kulan may select foraging sites based on the level of greening (Payne

Fig. 8.6 (continued) 2015). The winter in these areas is extremely cold and dry. In contrast, the two places in Alaska that horses could sustain themselves across winter, White River and Healy (Guthrie & Stoker, 1990), have milder winter with slightly more snow

Fig. 8.7 Yakutia in Arctic Russia is considered to be the coldest inhabited place on the Earth and the Yakut Horses have rapidly adapted to the extreme cold, aridity, and coarse vegetation (photo—Svetlana Ivanova, https://upload.wikimedia.org/wikipedia/commons/7/78/%D0%AF%D0%BA%D1%83%D1%82%D1%81%D0%BA%D0%B8%D0%B5_%D0%BB%D0%BE%D1%88%D0%B0%D0%B4%D0%B8.jpg, CC BY 3.0 (https://creativecommons.org/licenses/by/3.0/))

et al., 2020). In winter, when forage availability may be considerably lower and is usually of a much poorer quality, equids usually reduce their intake that is facilitated by their lowered metabolic rate and slower passage of forage through the gut (Kuntz et al., 2006). Since much of the faeces retains semi-digested forage, coprophagy is quite common among equids during all seasons (Schoenecker et al., 2016).

Most equids need to drink every day, especially if they cannot obtain enough water through succulent forage. Often, however, they may use foraging areas based on proximity of water sources (Schoenecker et al., 2016). The kulan is best adapted to water deficient conditions and can venture over 20 km from water sources, even utilizing saline water (Nandintsetseg et al., 2016; Šturm et al., 2017) and digging to access water (Feh et al., 2002). Based on this ability, kulan can utilize vast areas, moving on average 8 km/day (Lugauer, 2010), with annual home ranges of ca. 5000–70,000 km^2 (Kaczensky et al., 2008, 2011). Not much is known about water dependence of kiang, but they rarely are seen utilizing fresh water from rivers and streams (Schaller, 1998; St-Louis & Côté, 2009). Water may, however, be important for kiang as evidenced by radial trails originating from a small spring

Fig. 8.8 Kiang in summer congregate on moist sedge meadows in the lake and river basins in late summer that offer relatively higher green forage biomass (top panel photo—courtesy of Karma Sonam; bottom panel photo—courtesy of Abhishek Ghoshal)

and spreading over a small plateau frequented by kiang in eastern Ladakh (Bhatnagar, personal observation). In winter, low to medium snow cover can satisfy their need for water and allow them to use foraging habitat with greater flexibility (Payne et al., 2020). Heavy snowstorms or *dzuds* can, however, cause starvation related mortality as has been observed in all the three species (Kaczensky et al., 2011; Schaller, 1998; Schoenecker et al., 2016). The Przewalski's horse and kulan

Fig. 8.9 The frequency of extreme snow fall in winters (dzuds) is on the rise in many parts of Central Asia causing starvation related losses. Equids tend to move to areas with less snow such as windswept crests or areas with browse. Low to moderate amounts of snow can help them reduce dependence on watering points and allow foraging with greater flexibility (panel **a**, Przewalski's horse, photo—courtesy of Tara Buk, Smithsonian National Zoo and Conservation Biology Institute; panel **b**, Persian onager, photo—courtesy of Amanda Carberry, Columbus Zoo and Aquarium; panel **c**, Kiang, photo—courtesy of Rigzen Dorjay)

do avoid deep snow by shifting latitudinally or vertically to areas with lower snow cover (Cao, unpubl. data). In windswept, open habitats it is extremely difficult for equids to find shelter; however, perhaps they may behaviourally avoid hypothermia by facing against the wind and tucking their bushy tail under their hind legs (Mejdell et al., 2020). Other morphological traits including hair coat, skin thickness, and subcutaneous fat also have been attributed to cold adaptation in various breeds of domestic horse (Mejdell et al., 2020). Strangely enough, clustering behaviour has not evolved nor has lying down on a hairy underside to avoid wind despite the fact that even on a steppe, wind shear reduces wind speed dramatically close to ground level. In resource rich areas kulan aggregates in large groups, a strategy that possibly helped with exposing forage through thin snow (Cao, unpubl. data), but even in these circumstances inter-animal distances are much larger than they are in musk oxen or wild bovids. This may mean that the thermoneutral zone of these cold-adapted animals is much wider than that of bovines (bison, yak, Asian buffalo, etc.) which evolved under tropical conditions (see Prins et al., in press). To our knowledge, except for the study on the Przewalski's horse (Arnold et al., 2006), there is no information on thermo-physiological adaptations under field conditions or the role of hormones and adipose tissue mobilization in other cold-adapted equids (i.e., kiang, kulan, and the Yakut and Alaskan horse), but we believe that this could be a fertile field of research.

The three cold-adapted equids have adapted to utilize the relatively low amounts of seasonally available and dispersed forage, under conditions of extreme cold and aridity, thus perhaps filling among the most harsh and extreme niches inhabited by terrestrial large mammals (Fig. 8.9).

Physiological Basis of Cold Adaptation

Equids are homeothermic (warm-blooded) animals that maintain their core body temperature within a narrow limit even when the environmental temperature varies extensively (Sneddon, Chap. 9). They achieve this via endothermy (heat produced by the body). Endothermic animals maintain body temperature (Tb) constant by altering their metabolic heat production. When the ambient temperature (Ta) falls below Tb, the temperature gradient ($\Delta T = Tb - Ta$) triggers mechanisms to increase heat production, decrease the rate of heat loss, or permit the core body temperature to drop below "normal" (Tattersall et al., 2012).

When animals can maintain a stable core body temperature, using only physiological regulatory mechanisms, this is described as their thermoneutral zone (Autio, 2008; Rubner, 1982; Sjaastad et al., 2016a, 2016b). The lower end of the thermoneutral zone is referred to as the lower critical temperature (LCT; Autio et al., 2008; Mejdell et al., 2020). In ad libitum fed and cold-adapted yearling quarter and quarter horse crosses the LCT is –15 °C (Cymbaluk & Christison, 1989). When the ambient temperature drops below LCT, animals need to generate more body heat and conserve it via a reduction in body temperature and respiratory rate. In domestic horses, managed in cold environments, animals are provided with supplemental feed to meet the increased caloric needs. However, wild equids, living in extreme cold environments, may experience limited food availability further decreasing their ability to increase body heat and/or metabolic rate. Interestingly, Yakutian horses withstand temperature fluctuations between +38 and –70 °C, do not require additional shelter, and can graze on vegetation deep under snow cover (Librado et al., 2015). In domestic European breeds of horses and other mammals, under these circumstances, other physiological and anatomical characteristics may play a role in temperature regulation. A transient increase in heat production is achieved by inducing shivering (defined as uncoordinated or nonsynchronous contraction of skeletal muscles). Skeletal muscles generate heat via aerobic mechanisms which results in rapid heat production. Equids also increase body (surface) heat via piloerection (erection of the hair of the skin due to contraction of the tiny arrectores pilorum muscles that elevate the hair follicles above the rest of the skin and move the hair vertically). Furthermore, they allow the temperatures in the extremities (lower legs), ears, and muzzle to decrease to minimize surface heat loss. Increases in body heat is also accomplished via an increase in metabolism. Amongst wild equids, most information is available in the Przewalski's horse (Arnold et al., 2006). Studies suggest that animals adapt to changing environmental/seasonal condition by altering their metabolic rate. Specifically, during winter months, Przewalski's horse lower their heart rate, decrease locomotion, and exhibit significantly lower peripheral body temperatures leading to an overall reduction in energy requirements. However, there is a knowledge gap as to the physiological mechanisms involved in cold adaptation in kulans, kiangs, and Yakutian horses that warrants additional research.

Alternatively, heat production can be increased by breakdown of fat reserves (converting triglycerides to free fatty acids) also referred to as the non-shivering

thermogenesis (Jastroch et al., 2018). These mechanisms are controlled by catecholamines and the sympathetic nervous system. The primary target of this mechanism is the brown adipose tissue. Heat production occurs within fat cells and these deposits are highly vascularized with high concentrations of mitochondria. The heat generated is rapidly distributed throughout the body, via the blood stream, to increase core body temperature. Utilization of brown adipose tissue is catalyzed by the mitochondrial uncoupling protein, UCP 1 (Jastroch et al., 2018). UCP1 is almost exclusively expressed in the brown/beige fat tissues. Although brown adipose tissue is present in equids and is considered to play a role in non-shivering thermogenesis, the UCP1 gene is inactivated via conversion into a pseudogene even in cold-adapted domestic equids (Gaudry & Campbell, 2017; Jastroch et al., 2018). Further this inactivation in ancestral equids is estimated to have occurred *ca*. 20–25 million years ago. However, other UCP proteins (UCP 2, UCP 3) have been described and their role in equid brown adipose tissue utilization remains poorly understood. In the mouse and humans, UCP2 is expressed in brown adipose tissue in addition to several other tissue types and is considered to be involved in fatty acid metabolism and in turn, thermogenesis (Ricquier, 1999). Likewise, although little is known about the expression patterns of UCP 3 in equids, this mitochondrial protein is mostly expressed in skeletal muscles of humans and rodents as well as in the heart and brown fat, at least in rodents (Ricquier, 1999). These findings warrant additional studies in equids.

Equids Also Utilize Additional Metabolic Mechanisms to Regulate Body Heat

Specifically, animals can reduce their metabolic rate during winter, lower the heart rate, and lower thyroid hormone T3 levels with a concomitant increase in T4 (Brinkmann et al., 2016). Studies conducted in the Przewalski's horse examined various factors influencing the metabolic rate in the context of changes in heart rate including thermoregulation, locomotor activity (LA), and heat increment of feeding (HI) (Arnold et al., 2006). During winter months, heart rate decreased to almost half the value (*ca*. 44 beats per min) of that during summer months (*ca*. 89 beats per min). Locomotor activity also decreased in winter, and the mean subcutaneous temperature declined to reach the lowest values in April. Results suggest that regulation of energy expenditure is not influenced by energy intake, but that animals appear to respond to diurnal changes which influence circulating hormone levels (e.g., T3 and other hormones that regulate metabolism) resulting in lower energy demands during winter. Although, in most species, thermoregulation is facilitated by peripheral vasoconstriction which in turn lowers the surface temperature and causes a decrease in heat loss from the body, Przewalski's horses reduce their endogenous heat production by lowering the core body temperature with a concomitant decrease in skin surface temperature (Arnold et al., 2006). Furthermore, Przewalski's horses decrease their food intake during winter and mobilize body fat for energy and heat

production. Like Shetland ponies, the Przewalski's horse also exhibits hypometabolism resulting in reduced energy needs (Brinkmann et al., 2012).

Morphological Adaptation to Cold

Cold-adapted species often have numerous morphological characteristics that appear to be adaptations to cold environments including large body size, increased hair length, height, and higher subcutaneous body fat deposition/distribution. Extensive studies on cold adaptation have been conducted on Arctic species of mammals and birds, but studies are limited in wild equids (Feist & White, 1989; Scholander et al., 1950). Early studies showed an almost linear relationship between the basal heat production and body surface area, estimated using the Meeh's formula (Scholander et al., 1950). Although several arguments have been presented in favour of the role of surface to volume ratio in thermoregulation, Steegmann (2007) stated that mass and not surface area influences cold adaptation in humans. Interestingly, analysis of inter-breed variation in the horse suggests that larger animals have an advantage over smaller animals since heat production is proportional to volume and heat loss is proportional to surface area (Langlois, 1994). This phenomenon is called the Bergmann's rule. However, Meiri and Dayan (2003) reported that only 65% of mammal species evaluated adhered to this rule and more research is warranted in cold-adapted mammals, especially wild equids. Alternatively, animals may limit the ratio of surface area to volume by assuming a more spherical form, also referred to as Allen's rule which hypothesizes that in response to cold species will evolve shorter limbs, tail, ears, and/or wings. In the context of equids, most cold-adapted equids tend to be stocky with shorter limbs (e.g., Yakutian horse, Icelandic pony, Przewalski's horse). The latter adaptation also limits heat loss from the leg by minimizing the exposure of blood vessels to ambient temperature (Fig. 8.10).

Thermal Insulation

Animals also combat cold induced heat loss by thermal insulation. This involves vasoconstriction of the peripheral blood vessels, resulting in the diversion of warm blood from the cooler surface to the warmer core. This results in equilibration of surface temperature with ambient temperature to complement the insulation provided by the subcutaneous adipose tissue, skin, and hair (Langlois, 1994). In the Plateau pika (*Ochotona curzoniae*), chronic exposure to cold leads to browning of subcutaneous white adipose tissue fat and a concomitant increase in the amount of brown adipose tissue in order to increase thermogenesis (Li et al., 2018). It is plausible that the cold-adapted equids also may express similar changes and warrants additional research. Most wild equids also grow a thicker undercoat and longer outer coat during the autumn. For example, the Przewalski's horse grows a winter coat

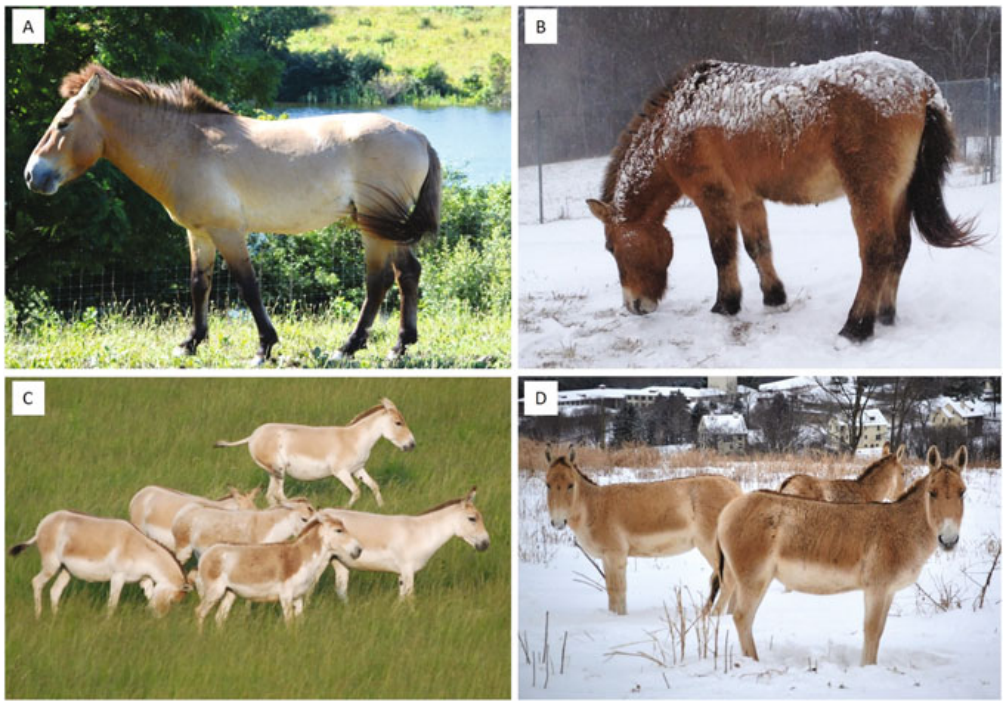

Fig. 8.10 Przewalski's horse (**a**, **b**) and Persian onager (**c**, **d**) exhibit pronounced morphological adaptation (thicker coat) during summer (**a**, **c**) and winter (**b**, **d**) (Przewalski's horse, photos (**a**, **b**)—courtesy of Smithsonian National Zoological Park and Conservation Biology Institute; Persian onager, photos—courtesy of Columbus Zoo and Aquarium (**c**) and Smithsonian National Zoological Park and Conservation Biology Institute (**d**))

annually to combat extreme cold. Interestingly, the Persian onager also grows a dense coat and does not exhibit extensive hair shedding with warming temperatures. Brinkmann et al. (2018) reported that, to support this hair growth, animals may require additional dietary protein. When a herd of Shetland ponies were maintained on a restricted diet, those in the treatment group exhibited limited hair growth compared to animals on a more nutritious control diet. The latter were able to grow their hair longer which increased their ability to protect from extreme cold. Under food restricted conditions, animals also metabolize more subcutaneous fat to generate the necessary body heat. A comparison of coats of horses, donkey, and mules demonstrates that while donkeys' hair coat shows no seasonal changes, horses grow thicker hair during winter (Osthaus et al., 2018); the donkey is, of course, a North African species that is adapted to very hot circumstances. However, when hair width was analysed, mules had the thickest hair followed by the horse and then the donkey, suggesting that horses and mules have a greater ability to withstand extreme cold. It is not known yet how the genetics of hybridization of horse with donkey would lead to thick hairs.

Horses with thicker coats can tolerate extreme cold. A thicker coat traps the body warmth and helps keep the skin temperature warmer than the environment. However, when exposed to rain, it leads to matting of the coat, and as a result the coat is

no longer able to trap the warmth. Jørgensen et al. (2016) studied the shelter preference of horses exposed to Nordic winter conditions. Interestingly, although cold-adapted horses prefer to spend time outdoors, when exposed to wet conditions, these horses choose to seek shelter and spend more time indoor. This may support our conclusion that caballoid horses are dry-cold adapted species (c.f., Fig. 8.2).

Heat Loss in Wild Equids

Heat loss is correlated with body size. A larger horse innately has a smaller surface area to dissipate heat compared to a similarly built smaller horse (Mejdall et al., 2020). Equids typically dissipate heat via their skin but there is limited information on what parts of the body are involved in this heat loss. Thermal imaging is currently used in animal health diagnostics, especially in animals afflicted with joint disorders and for evaluating animal performance during exercise (Cilulko et al., 2013). Further, the radiant energy measured is a manifestation of both the internal body heat and the thermal properties of the skin, coat, and the temperature gradient between the skin surface and the environment. Interestingly, these measurements also permit the analysis of muscle groups typically involved in heat production or dissipation. Recently, Domino et al. (2020) used infrared thermal imaging to compare the surface thermal patterns of horses and donkeys exposed to identical environmental conditions. This approach allows the detection of radiant heat which is converted into a thermogram wherein the colour gradient corresponds to the temperature gradient (Fig. 8.11). These authors report that, under similar environmental conditions (20.2 °C), horses exhibit higher surface temperature (*ca*. 22.7 °C) as compared with donkeys (*ca*. 18.8 °C) and attributed the differences in the thermal properties of the skin and the coat between the horse and donkeys. Specifically, donkeys had a thicker skin, more subcutaneous fat, and a longer hair coat leading to better thermal insulation than in the horse.

Thermal imaging of Przewalski's horse and Persian onagers exposed to extreme cold environments reveals that both species have thermal signatures around the eyes and head, and lower surface temperatures on the other parts of their body (Fig. 8.11). Surface temperatures along the side of the muzzle, face, withers, and posterior gluteal muscles were generally warmer, overall, for the Przewalski's horse than for the Persian onager. For Persian onagers, the limbs appeared cooler, in these extreme cold conditions, when compared with Przewalski's horses. These findings suggest that, similar to donkeys, the Persian onager may have better insulation, and possibly a more efficient counter current exchange system in the limbs, than do horses, making them more cold tolerant.

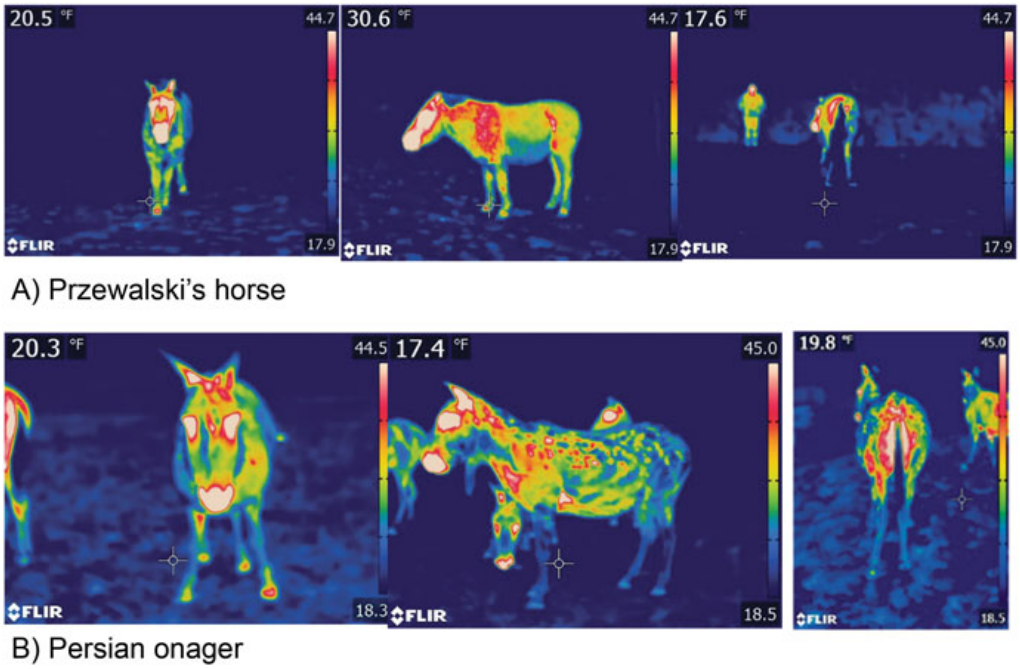

Fig. 8.11 Infrared thermal imaging of the Przewalski's horse (panel **a**) and the Persian onager (panel **b**). Thermal images are global emissivity patterns of a target object displayed as a temperature gradient (colour spectrum). Hotter areas appear red or white (when the temperature exceeds the set threshold high limit). Colder areas appear purple or blue (lowest temperature measured). All images were captured when the ambient temperature (value shown on the bottom right corner of the image) was between –7.8 and –7.5 °C. The temperature indicated on the top left corner of the image corresponds to the temperature recorded at the location of the cursor. The colour scale depicted on the right side of all images represents a heat map corresponding to the temperature gradient depicted in the image. As shown in the images, both Przewalski's horse and Persian onagers lose the most body heat in the region of their eyes and muzzle (white) while the limbs appear to lose the least amount of heat (as depicted by the blue colour) (photos—courtesy of Priya Bapodra)

Genetics of Cold Adaptation

The Przewalski's horse, Persian onager, and kiang are well adapted to cold environments, as are several breeds of domesticated equids occurring in such environments. As mentioned earlier in the chapter, all three species occur in latitudes/altitudes that experience extreme climatic conditions. All three species exhibit morphological adaptations critical for survival in extreme cold, including being exceptionally hairy (grow a dense winter coat), are compact, and have short limbs (Librado et al., 2015). Although some genetic data resources are available for the Przewalski's horse, Persian onager, and the kiangs, no systematic studies have been conducted to reveal the genetic basis underlying cold adaptation. Most information available today is gleaned from studies conducted on the Yakutian horse (Librado et al., 2015). They can withstand ambient temperatures below –70 °C and prefer to stay outdoor even under extreme weather conditions. Since a significant portion of the

year the ground is covered in several feet of snow, Yakutian horses often forage by digging for vegetation buried under snow.

In a comparative analysis involving Yakutian horses, two ancient specimens (from early nineteenth century), two Late Pleistocene specimens, several domestic horses, and three wild Przewalski's horses, genetic adaptation to cold appeared to be controlled by modification of non-coding and protein coding sites as well as gene duplication, suggesting the Yakutian horses are well suited for fast adaptive processes (Librado et al., 2015). Specifically, regions upstream of the translation start sites (TSS) revealed significant enrichment of adaptive candidates. Modifications were within the most proximal kilobase upstream from the TSS where regulatory gene elements are often localized. Candidate genes involved those that regulate hair density, subcutaneous fat accumulation, and relatively small surface area relative to body mass, all of which also minimize heat loss. Other genes involved include transglutaminase 3 (*TGM3*) whose loss of function in mice results in impaired and thinner hair (John et al., 2012), thyroid hormone receptor-associated gene (*THRAP3*) which interacts with adrenoreceptor α1B that regulates the vasoconstriction/vasodilatation reflex following cold exposure. Enrichment for hypothyroidism, endocrine system diseases, and type 1 diabetes mellitus also were observed. The latter was interpreted to reflect an adaptation to regulating glycaemia (glucose production to exert anti-freezing effects). Yakutian horses also exhibit adaptation in the protein coding sites; about 130 genes, enriched for number of pathways, including galactose, starch, and sucrose metabolism and diseases like Nelson's syndrome which leads to an excess of adrenocorticotropic hormone. Furthermore, Yakutian horses also exhibit gene duplication that support cold adaptation; genes affected were enriched for several biological pathways including steroid hormone biosynthesis, fatty acid metabolism, metabolic pathways, and olfactory transduction. Genes associated with abnormality of temperature regulation (*ACADSB*, *ATP1A2*, *CYP11B2*, and *HSPG2*) also were enriched suggesting the Yakutian horses have evolved several mechanisms that support survival in subarctic climate. A recent study evaluated genes related to cold adaptation in Arctic or Antarctic environments in the human, Arctic fox, Yakut horse, mammoth, polar bear, and minke whale (Yudin et al., 2017). Although no cold-adaptive genes were conserved in all six species, several general mechanisms and biochemical pathways involved in reorganization of the cardiovascular function, thickness and strength of the skin, increased heat production, improved immunity, and behavioural changes were identified (Yudin et al., 2017). However, additional studies are warranted in wild equids to identify specific genetic signatures for cold adaptation.

Conservation Implications

The three extant, cold-adapted equids face several threats across their wide range (Moehlman et al., 2016), however, here, we focus primarily on their possible responses to the changing climate, which is likely to result in global warming,

climatic uncertainties, and increased extreme weather events (IPCC, 2021). Climate change can impact the very basis of survival—that is, food (Wu et al., 2017), water, and shelter, and could be very challenging for these equids. In fact, the vast declines in the historical range of equids (Orlando, 2015) may be due to a combination of climatic and anthropogenic causes (Bouman & Bouman, 1994; Buuveibaatar et al., 2016). It is, therefore, a matter of significant conservation concern as to what could happen as a result of more rapid climatic and land use changes.

Overall, equids inhabiting extreme cold environments, appear to be well adapted to survive under these conditions. They also appear to tolerate a fairly large range of temperature and precipitation (Fig. 8.4), that may enable them to cushion the largely unknown changes in resources brought about by a changing climate. Despite this evolutionary advantage, it is likely that only species that are able either to, maintain core body temperatures near normal, or migrate to warmer areas, could still survive. In the absence of a similar adaptive behaviour, many species of wild equids could perish, as was previously reported in the reintroduced populations of Przewalski's horse in Mongolia (Kaczensky et al., 2011). It remains unclear how the projected global warming trends (IPCC, 2021), with the related uncertainties, changes in habitats and increased extreme events, will impact the distribution and survival of wild equids. It is also possible that climate extremes may challenge the ability of animals to adapt quickly. Therefore, it will be important that large-scale conservation efforts designate large areas of the extant habitats, including areas that might become future refuges for conservation of equid species.

The seasonally abundant forage in the equid range in Central Asia/Tibetan Plateau regions also attracts pastoralists who have livestock that often intensively graze the rangelands, resulting in competitive exclusion of many wild herbivores (Berger et al., 2013). This impact of humans and their livestock contributed to the decimation and extinction of the Przewalski's horse in the nineteenth century (Bouman & Bouman, 1994). Competition between livestock and wild equids is already a growing issue of concern and may impact the survival of the latter by both interference and exploitative competition, as can be seen for the Przewalski's and domestic horses in the Dzungarian Gobi during winter (Šturm et al., 2017), and reduced kiang abundance in high livestock density areas in Ladakh (Bhatnagar & Wangchuk, 2002). Using stable isotope studies, Kaczensky et al. (2017) showed that pre-extinction populations of Przewalski's horses in Mongolia that were sympatric with domestic horses relied on browsing in winter but shifted to more suitable grazing in the reintroduced populations that were free of competition from domestic horses. Even though kiangs consumed barely 10% of available forage in eastern Ladakh (most of the rest being consumed by livestock) there is increasing consternation among nomads and officials that the kiang is competing for forage with the valuable cashmere goats (Bhatnagar et al., 2006). The combined effects of shifting climate niche and food resources (under the Representative Concentration Pathway 2.6) are likely to reduce suitable habitat for kiang on the Tibetan plateau by about 30% while also reducing preferred forage by over 7% (Wu et al., 2017). In much of their range, because of being outcompeted by livestock, kulan will need to increase the amount of browse in the diet (Šturm et al., 2017, 2021; Xu et al., 2012), which

Fig. 8.12 Most people inhabiting equid range are pastoralists and their flocks may compete with wild equids (photo—courtesy of Yash Veer Bhatnagar).

can be a challenge for equids due to the higher presence of phenolic compounds in this food source (Duncan, 1992) (Fig. 8.12).

The availability of water impacts pastures that can be utilized by equids (Nandintsetseg et al., 2016; Schoenecker et al., 2016). In fact, the most water tolerant of the three species, kulan, can use pastures about 20 km from water sources, and their long-range movements were governed by water availability rather than by the distribution of preferred habitats (Nandintsetseg et al., 2016). With light snow as a source of water in winter, kulan (and possibly other equids) can use pastures farther from water sources (Payne et al., 2020). However, heavy snow can make it difficult to access forage and prolonged deep snow can lead to heavy mortality of these ungulates (Schaller, 1998). Climate change may result in higher unpredictability in precipitation in some areas and years, that may cause corresponding uncertainties in habitat availability and the need for long-distance and frequent movements.

Since Przewalski's horses are often restricted to the small ranges where they were introduced, changing climate could limit their ability to migrate to ideal environments, hence they may not be able to adapt quickly to rapid changes (Kaczensky et al., 2011). This could affect the survival probability of this species. There is also a need to evaluate potential translocation and reintroduction sites for availability of food especially during extreme winters, and for water year-round.

In some areas, such as the Gobi, Przewalski's horses could thrive due to a greater tolerance by the herders who pride their lifestyle coexisting with the high-profile reintroduced species (Kaczensky et al., 2017). This underscores the importance of working closely with stakeholders to improve their perceptions as a means of conservation of the equids. With reference to the Przewalski's horse, Kaczensky et al. (2017) also highlight the need for disaster planning by local herders, multiple reintroduction sites with spatially dispersed populations for reintroduced Przewalski's horses, and a landscape-level approach beyond protected area boundaries to allow for migratory movements of Asiatic wild equids. This will need greater political will, resources, and integrated planning and implementation at landscape scales.

The monodactyl equids have faced millions of years of competition with tridactyl equids and other herbivores to finally establish in the newly formed open habitats that expanded during the Pleistocene. However, climate and anthropogenic pressures, over the past 2 million years, have caused severe declines in their range, diversity, and abundance, but the recent declines have been the most dramatic. Without evidence-informed interventions, increasing pressure on the mountainous grasslands, deserts, and steppes from, e.g., livestock, mining, infrastructure, compounded by uncertainties of forage, water, and other resource availability due to the accelerated climate warming, could result in further declines of this amazing taxa of cold-adapted equids.

Perspectives

As described in this chapter, our current understanding of cold adaptation in wild equids is derived from a small number of studies on the domestic horse and donkey. Most information on physiological adaptation to cold is extrapolated from systematic studies on domestic equids that have been studied in temperate conditions. It is, therefore, imperative to address this knowledge gap in all species of cold-adapted wild equids. Specifically, questions need to be addressed, such as are physiological mechanisms reported in domestic equids that permit adaptation to extreme cold environments conserved in their wild counterparts? Furthermore, with the availability of high-quality genome assemblies for several equids, studies also are warranted to identify the genomic/genetic basis of cold adaptation.

References

Arnold, W., Ruf, T., & Kuntz, R. (2006). Seasonal adjustment of energy budget in a large wild mammal, the Przewalski horse (*Equus ferus przewalskii*) II. Energy expenditure. *Journal of Experimental Biology, 209*, 4566–4573.

Autio, E. (2008). Loose housing of horses in a cold climate. Kuopion Yliopiston Julkaisuja C. Luonnontieteet Ja Ympäristötieteet 245.

Autio, E., Sihto, U., Mononen, J., & Heiskanen, M.-L. (2008). Energy intake and growth of weanling horses in a cold loose housing system. *Agricultural and Food Science, 17*(4), 338–350. https://jukuri.luke.fi/handle/10024/474660

Azzaroli, A. (1983). Quaternary mammals and the "End-Villafranchian" dispersal event—A turning point in the history of Eurasia. *Palaeogeography, Palaeoclimatology, Palaeoecology, 44*, 117–139. https://doi.org/10.1016/0031-0182(83)90008-1

Azzaroli, A. (1992). Ascent and decline of monodactyl equids: A case for prehistoric overkill. In *Annales Zoologici Fennici*. Finnish Zoological Publishing Board, formed by the Finnish Academy of Sciences, Societas Biologica Fennica Vanamo, Societas pro Fauna et Flora Fennica, and Societas Scientiarum Fennica, pp. 151–163

Azzaroli, A. (2000). On *Equus livenzovensis* Baigusheva 1978 and the "stenonid" lineage of equids. *Palaeontographia Italica, 87*, 1–17.

Barnosky, A. D., Koch, P. L., Feranec, R. S., et al. (2004). Assessing the causes of late Pleistocene extinctions on the continents. *Science, 306*, 70–75.

Bendrey, R. (2012). From wild horses to domestic horses: A European perspective. *World Archaeology, 44*, 135–157.

Bennett, D., & Hoffmann, R. S. (1999). *Equus caballus* Linnaeus, 1758 Horse. *Mammalian Species, 628*, 1–14.

Bennett, E. A., Champlot, S., Peters, J., Arbuckle, B. S., et al. (2017). Taming the late quaternary phylogeography of the Eurasiatic wild ass through ancient and modern DNA. *PLoS One, 12*, e0174216. https://doi.org/10.1371/journal.pone.0174216

Berger, J., Buuveibaatar, B., & Mishra, C. (2013). Globalization of the cashmere market and the decline of large mammals in Central Asia. *Conservation Biology, 27*, 679–689.

Bernor, R. L., Cirilli, O., Jukar, A. M., et al. (2019). Evolution of early Equus in Italy, Georgia, the Indian subcontinent, East Africa, and the origins of African Zebras. *Frontiers in Ecology and Evolution, 7*.

Bhatnagar, Y. V., & Wangchuk, R. (2002). Status and conservation of the Tibetan Gazelle (*Procapra picticaudata*) in Ladakh. In *Anon. 2002. Conserving Biodiversity in the Trans-Himalaya: Initiatives of field conservation in Ladakh*. Report Submitted to the Wildlife Institute of India, the International Snow Leopard Trust, and US Fish and Wildlife Service.

Bhatnagar, Y. V., Wangchuk, R., Prins, H. H., Van Wieren, S. E., & Mishra, C. (2006). Perceived conflicts between pastoralism and conservation of the kiang *Equus kiang* in the Ladakh Trans-Himalaya, India. *Environmental Management, 38*(6), 934–941.

Boeskorov, G. G., Potapova, O. R., Protopopov, A. V., Plotnikov, V. V., Maschenko, E. N., Shchelchkova, M. V., Petrova, E. A., Kowalczyk, R., van der Plicht, J., & Tikhonov, A. N. (2018). A study of a frozen mummy of a wild horse from the Holocene of Yakutia, East Siberia, Russia. *Mammal Research, 63*(3), 307–314.

Bouman, I., & Bouman, J. (1994). Przewalski's horse. In L. Boyd & K. A. Houpt (Eds.), *The history of Przewalski's horse* (pp. 5–38). State University of New York Press.

Brikiatis, L. (2014). The De Geer, Thulean and Beringia routes: key concepts for understanding early Cenozoic biogeography. *Journal of Biogeography, 41*(6), 1036–1054.

Brinkmann, L., Gerken, M., Hambly, C., Speakman, J. R., & Riek, A. (2016). Thyroid hormones correlate with field metabolic rate in ponies, *Equus ferus caballus*. *The Journal of Experimental Biology, 219*, 2559–2566.

Brinkmann, L., Gerken, M., & Riek, A. (2012). Adaptation strategies to seasonal changes in environmental conditions of a domesticated horse breed, the Shetland pony (*Equus ferus caballus*). *The Journal of Experimental Biology, 215*(7), 1061–1068. https://doi.org/10.1242/jeb.064832

Brinkmann, L., Riek, A., & Gerken, M. (2018). Long-term adaptation capacity of ponies: effects of season and feed restriction on blood and physiological parameters. *Animals, 12*, 88–97.

Buuveibaatar, B., Mueller, T., Strindberg, S., Leimgruber, P., Kaczensky, P., & Fuller, T. K. (2016). Human activities negatively impact distribution of ungulates in the Mongolian Gobi. *Biological Conservation, 203*, 168–175.

Ceballos, G., Ehrlich, P. R., Barnosky, A. D., et al. (2015). Accelerated modern human–induced species losses: Entering the sixth mass extinction. *Science Advances, 1*, e1400253. https://doi.org/10.1126/sciadv.1400253

Cilulko, J., Janiszewski, P., Bogdaszewski, M., & Szczygielska, E. (2013). Infrared thermal imaging in studies of wild animals. *European Journal of Wildlife Research, 59*, 17–23.

Cirilli, O., Pandolfi, L., Rook, L., & Bernor, R. L. (2021). Evolution of old world *Equus* and origin of the Zebra-Ass clade. *Scientific Reports, 11*, 10156. https://doi.org/10.1038/s41598-021-89440-9

Clark, P. U., Dyke, A. S., Shakun, J. D., Carlson, A. E., Clark, J., Wohlfarth, B., Mitrovica, J. X., Hostetler, S. W., & McCabe, A. M. (2009). The last glacial maximum. *Science, 325*(5941), 710–714.

Crees, J. J., & Turvey, S. T. (2014). Holocene extinction dynamics of *Equus hydruntinus*, a late-surviving European megafaunal mammal. *Quaternary Science Reviews, 91*, 16–29. https://doi.org/10.1016/j.quascirev.2014.03.003

Cymbaluk, N., & Christison, G. I. (1989). Effect of diet and climate on growing horses. *Journal of Animal Science, 67*, 48–59.

Domino, M., Romaszewski, M., Jasiński, T., & Maśko, M. (2020). Comparison of the surface thermal patterns of horses and donkeys in infrared thermography images. *Animals, 10*(12), 2201. https://doi.org/10.3390/ani10122201

Duncan, P. (Ed.). (1992). *Zebras, asses, and horses: An action plan for the conservation of wild equids* (1st ed.). Island Pr.

Ehlers, J., Gibbard, P. L., & Hughes, P. D. (Eds.). (2004). *Quaternary glaciations: extent and chronology* (Vol. 15). Elsevier.

Feh, C., Munkhtuya, B., Enkhbold, S., & Sukhbaatar, T. (2001). Ecology and social structure of the Gobi Khulan *Equus Hemionus* Subsp. in the Gobi B National Park, Mongolia. *Biological Conservation, 101*(1), 51–61. https://doi.org/10.1016/S0006-3207(01)00051-9

Feh, C., Shah, N., Rowen, M., Reading, R., & Goyal, S. P. (2002). Status and action plan for the Asiatic wild ass (*Equus hemionus*). In P. D. Moehlman (Ed.), *Equids: zebras, asses and horses—Status survey and conservation action plan* (pp. 62–71). World Conservation Union.

Feist, D. D., & White, R. G. (1989). Terrestrial mammals in cold. In L. C. H. Wang (Ed.), *Animal adaptation to cold. Advances in comparative and environmental physiology* (Vol. 4). Springer. https://doi.org/10.1007/978-3-642-74078-7_9

Forsten, A. (1988). Middle Pleistocene replacement of stenonid horses by caballoid horses—Ecological implications. *Palaeogeography, Palaeoclimatology, Palaeoecology, 65*, 23–33.

Forsten, A. (1991). Size trends in holarctic anchitherines (Mammalia, Equidae). *Journal of Paleontology, 65*, 147–159.

Forsten, A. (1999). A review of *Equus stenonis cocchi* (Perissodactyla, Equidae) and related forms. *Quaternary Science Reviews, 18*, 1373–1408.

Fox-Dobbs, K., Leonard, J. A., & Koch, P. L. (2008). Pleistocene megafauna from eastern Beringia: Paleoecological and paleoenvironmental interpretations of stable carbon and nitrogen isotope and radiocarbon records. *Palaeogeography Palaeoclimatology Palaeoecology, 261*(1–2), 30–46.

Garcés, M., Cabrera, L., Agustí, J., & Parés, J. M. (1997). Old World first appearance datum of "*Hipparion*" horses: Late Miocene large-mammal dispersal and global events. *Geology, 25*(1), 19–22.

Gaudry, M. J., & Campbell, K. L. (2017). Evolution of UCP1 transcriptional regulatory elements across the mammalian phylogeny. *Frontiers in Physiology, 8*, 670. https://doi.org/10.3389/fphys.2017.00670

Guthrie, R. D. (1982). Mammals of the Mammoth Steppe as paleoenvironmental indicators. In D. M. Hopkins, J. V. Matthews, C. E. Schweger, & S. B. Young (Eds.), *Paleoecology of Beringia* (pp. 307–326). Academic Press. ISBN 978-0-12-355860-2.

Guthrie, D. R. (2001). Origin and causes of the Mammoth Steppe: A story of cloud cover, Woolly Mammal tooth pits, buckles, and inside-out Beringia. Beringian Paleoenviron. - Festschr. Honour DM Hopkins, *20*, 549–574. https://doi.org/10.1016/S0277-3791(00)00099-8

Guthrie, R. D. (2003). Rapid body size decline in Alaskan Pleistocene horses before extinction. *Nature, 426*(6963), 169–171.

Guthrie, R. D. (2013). *Frozen fauna of the mammoth steppe*. University of Chicago Press.

Guthrie, R. D., & Stoker, S. (1990). Paleoecological significance of mummified remains of Pleistocene horses from the North Slope of the Brooks Range, Alaska. *Arctic, 43*, 267–274.

Hack, M. A., East, R., Rubenstein, D. I., et al. (2002). Status and action plan for the plains Zebra (*Equus burchellii*). In *Equids zebras asses horses status survey and conservation action plan* (pp. 43–60).

Halsey, L. G., & Stroud, M. A. (2012). 100 years since Scott reached the pole: a century of learning about the physiological demands of Antarctica. *Physiological Reviews, 92*(2), 521–536.

Hansen, J., Sato, M., Russell, G., & Kharecha, P. (2013). Climate sensitivity, sea level and atmospheric carbon dioxide. *Philosophical Transactions of the Royal Society A: Mathematical, Physical and Engineering Sciences, 371*(2001), 20120294.

Harris, R. B., & Miller, D. J. (1995). Overlap in summer habitats and diets of Tibetan Plateau Ungulates. *Mammalia, 59*(2), 197–212. https://doi.org/10.1515/mamm.1995.59.2.197

Henley, S. R., Ward, D., & Schmidt, I. (2007). Habitat selection by two desert adapted ungulates. *Journal of Arid Environments, 70*(1), 39–48. https://doi.org/10.1016/j.jaridenv.2006

Hoffecker, J. F., Powers, W. R., & Goebel, T. (1993). The colonization of Beringia and the peopling of the New World. *Science, 259*(5091), 46–53.

Huntford, R. (2010). *Race for the South Pole: The expedition diaries of Scott and Amundsen*. Bloomsbury.

Hutchinson, G. E. (1957). Concluding remarks. *Cold Spring Harbor Symposia, 22*, 415–427.

IPCC. (2021). Climate Change 2021: The physical science basis. In V. Masson-Delmotte, P. Zhai, A. Pirani, S. L. Connors, C. Péan, S. Berger, N. Caud, Y. Chen, L. Goldfarb, M. I. Gomis, M. Huang, K. Leitzell, E. Lonnoy, J. B. R. Matthews, T. K. Maycock, T. Waterfield, O. Yelekçi, R. Yu, & B. Zhou (Eds.), *Contribution of Working Group I to the sixth assessment report of the intergovernmental panel on climate change*. Cambridge University Press.

Janis, C. (1976). The evolutionary strategy of the Equidae and the origins of rumen and cecal digestion. *Evolution, 30*, 757–774.

Janis, C. M., & Bernor, R. L. (2019). The evolution of equid monodactyly: A review including a new hypothesis. *Frontiers in Ecology and Evolution, 7*, 119.

Jastroch, M., Oelkrug, R., & Keipert, S. (2018). Insights into brown adipose tissue evolution and function from non-model organisms. *The Journal of Experimental Biology, 221*, jeb169425. https://doi.org/10.1242/jeb.169425

Jiang, D., Klaus, S., Zhang, Y. P., Hillis, D. M., & Li, J. T. (2019). Asymmetric biotic interchange across the Bering land bridge between Eurasia and North America. *National Science Review, 6*, 739–745.

John, S., Thiebach, L., Frie, C., Mokkapati, S., Bechtel, M., Nischt, R., Rosser-Davies, S., Paulsson, M., & Smyth, N. (2012). Epidermal Transglutaminase (TGase 3) is required for proper hair development, but not the formation of the epidermal barrier. *PLoS One, 7*(4), e34252. https://doi.org/10.1371/journal.pone.0034252

Jónsson, H., Schubert, M., Seguin-Orlando, A., et al. (2014). Speciation with gene flow in Equids despite extensive chromosomal plasticity. *Proceedings of the National Academy of Sciences, 111*, 18655. https://doi.org/10.1073/pnas.1412627111

Jørgensen, G. H. M., Aanensen, L., Mejdell, C. M., & Bøe, K. E. (2016). Preference for shelter and additional heat when exposed to Nordic winter conditions. *Equine Veterinary Journal, 48*(6), 720–726.

Kaczensky, P., Ganbaatar, O., Von Wehrden, H., & Walzer, C. (2008). Resource selection by sympatric wild equids in the Mongolian Gobi. *Journal of Applied Ecology, 45*(6), 1762–1769. https://doi.org/10.1111/j.1365-2664.2008.01565.x

Kaczensky, P., Ganbataar, O., Altansukh, N., Enkhsaikhan, N., Stauffer, C., & Walzer, C. (2011). The danger of having all your eggs in one basket—winter crash of the re-introduced Przewalski's horses in the Mongolian Gobi. *PLoS One, 6*, e28057.

Kaczensky, P., Lkhagvasuren, B., Pereladova, O., Hemami, M., & Bouskila, A. (2020). *Equus hemionus* ssp. *hemionus* (amended version of 2015 assessment). The IUCN Red List of Threatened Species 2020: e.T7952A176245867. https://doi.org/10.2305/IUCN.UK.2020-3.RLTS.T7952A176245867.en. Downloaded on 1 June 2021.

Kaczensky, P., Šturm, M. B., Sablin, M. V., Voigt, C. C., Smith, S., Ganbaatar, O., Balint, B., Walzer, C., & Spasskaya, N. N. (2017). Stable isotopes reveal diet shift from pre-extinction to reintroduced Przewalski's horses. *Scientific Reports, 7*, 5950.

Klingel, H. (1977). Observations on social organization and behaviour of African and Asiatic wild asses (*Equus africanus* and *E. hemionus*). *Zeitschrift für Tierpsychologie, 44*, 323–331.

King, S. R. B. (2002). Home range and habitat use of free-ranging Przewalski horses at Hustai National Park, Mongolia. *Applied Animal Behaviour Science, 78*, 103–113.

King, S. R. B., & Gurnell, J. (2005). Habitat use and spatial dynamics of takhi introduced to Hustai National Park, Mongolia. *Biological Conservation, 124*(2), 277–290.

Koch, P. L., & Barnosky, A. D. (2006). Late quaternary extinctions: State of the debate. *Annual Review of Ecology, Evolution, and Systematics, 37*, 215–250. https://doi.org/10.1146/annurev.ecolsys.34.011802.132415

Kuntz, R., Kubalek, C., & Ruf, T. (2006). Seasonal adjustment of energy budget in a large wild mammal, the Przewalski horse (*Equus ferus przewalskii*) I. Energy intake. *Journal of Experimental Biology, 209*, 4557–4565.

Lambert, F., Delmonte, B., Petit, J. R., et al. (2008). Dust-climate couplings over the past 800,000 years from the EPICA Dome C ice core. *Nature, 452*(7187), 616–619.

Langlois, B. (1994). Inter-breed variation in the horse with regard to cold adaptation: a review. *Livestock Production Science, 40*(1), 1–7. https://doi.org/10.1016/0301-6226(94)90260-7

Larson, E. J. (2011). Poles apart: Scott, Amundsen and science. *Endeavour, 35*(4), 129–136. https://doi.org/10.1016/j.endeavour.2011.08.001

Li, J., Yang, Q., Bai, Z., et al. (2018). Chronic cold exposure results in subcutaneous adipose tissue browning and altered global metabolism in Qinghai-Tibetan plateau pika (*Ochotona curzoniae*). *Biochemical and Biophysical Research Communications, 500*(2), 117–123. https://doi.org/10.1016/j.bbrc.2018.03.147. Epub 2018 Apr 13. PMID: 29626477.

Librado, P., Der Sarkissian, C., Ermini, L., Schubert, M., Jónsson, H., Albrechtsen, A., Fumagalli, M., Yang, M. A., Gamba, C., Seguin-Orlando, A., Mortensen, C. D., Petersen, B., Hoove, C. A., Lorente-Galdos, B., Nedoluzhko, A., Boulygina, E., Tsygankova, S., Neuditschko, M., Jagannathan, V., . . . Orlando, L. (2015). Tracking the origins of Yakutian horses and the genetic basis for their fast adaptation to subarctic environments. *Proceedings of the National Academy of Sciences, 112*(50), E6889–E6897. https://doi.org/10.1073/pnas.1513696112

Lindsay, E. H., Opdyke, N. D., & Johnson, N. M. (1980). Pliocene dispersal of the horse *Equus* and late Cenozoic mammalian dispersal events. *Nature, 287*, 135–138.

Lisiecki, L. E., & Raymo, M. E. (2005). A Pliocene-Pleistocene stack of 57 globally distributed benthic $\delta^{18}O$ records. *Paleoceanography, 20*(1), 1–17.

Liu, X., & Dong, B. (2013). Influence of the Tibetan plateau uplift on the Asian monsoon-arid environment evolution. *Chinese Science Bulletin, 58*, 4277–4291.

Lorenzen, E. D., Nogués-Bravo, D., Orlando, L., et al. (2011). Species-specific responses of Late Quaternary megafauna to climate and humans. *Nature, 479*, 359–364.

Louys, J., Wilkinson, D. M., & Bishop, L. C. (2012). Ecology needs a paleontological perspective. In J. Louys (Ed.), *Paleontology in ecology and conservation* (pp. 23–38). Springer.

Lugauer, B. (2010). *Differences of movement pattern between Asiatic wild ass (Equus hemionus) and Przewalski's horse (Equus ferus przewalskii).* Diploma thesis, University of Vienna. Faculty of Life Sciences. https://doi.org/10.25365/thesis.8685

MacFadden, B. J. (1984). Systematics and phylogeny of *Hipparion*, *Neohipparion*, *Nannippus*, and *Cormohipparion* (Mammalia, Equidae) from the miocene and pliocene of the new world. *Bulletin of the American Museum of Natural History, 179*(1), 189–195.

MacFadden, B. J. (1986). Fossil horses from "*Eohippus*" (*Hyracotherium*) to *Equus*: Scaling, Cope's Law, and the evolution of body size. *Paleobiology, 12*, 355–369.

MacFadden, B. J. (2001). Three-toed browsing horse *Anchitherium clarencei* from the early Miocene (Hemingfordian) Thomas Farm, Florida. *Florida Museum of Natural History Bulletin, 43*, 79–109.

Martin, P. S. (1984). Prehistoric overkill: The global model. In P. S. Martin & R. G. Klein (Eds.), *Quaternary extinctions: A prehistoric revolution* (pp. 354–403). The University of Arizona Press.

McGrew, P. O. (1944). An early Pleistocene (Blancan) fauna from Nebraska. *Field Museum of Natural History, Geology Series, 9*, 33–66.

McHorse, B. K., Biewener, A. A., & Pierce, S. E. (2019). The evolution of a single toe in horses: Causes, consequences, and the way forward. *Integrative and Comparative Biology, 59*, 638–655.

McNab, B. K. (1971). On the ecological significance of Bergmann's rule. *Ecology, 52*, 845–854. https://doi.org/10.2307/1936032

Meiri, S., & Dayan, T. (2003). On the validity of Bergmann's rule. *Journal of Biogeography, 30*, 331–351.

Mejdell, C. M., Bøe, K. E., & Jørgensen, G. H. M. (2020). Caring for the horse in a cold climate—Reviewing principles for thermoregulation and horse preferences. *Applied Animal Behaviour Science, 231*, 105071. https://doi.org/10.1016/j.applanim.2020.105071

Moehlman, P. D. (2002). Status and action plan for the African Wild Ass (*Equus Africanus*). In *Equids zebras asses horses status survey and conservation action plan* (pp. 2–10).

Moehlman, P. D., King, S. R. B., & Kebede, F. (2016). Status and conservation of threatened equids. In J. I. Ransom & P. Kaczensky (Eds.), *Wild equids ecology, management, and conservation* (pp. 167–186). Johns Hopkins University Press.

Murchie, T. J., Monteath, A. J., Mahony, M. E., et al. (2021). Collapse of the Mammoth-Steppe in Central Yukon as revealed by ancient environmental DNA. *Nature Communications, 12*, 1–18. https://doi.org/10.1038/s41467-021-27439-6

Namgail, T., Rawat, G. S., Mishra, C., van Wieren, S. E., & Prins, H. H. T. (2012). Biomass and diversity of dry alpine plant communities along altitudinal gradients in the Himalayas. *Journal of Plant Research, 125*(1), 93–101. https://doi.org/10.1007/s10265-011-0430-1

Nandintsetseg, D., Kaczensky, P., & Ganbaatar, O. (2016). Spatiotemporal habitat dynamics of ungulates in unpredictable environments: The khulan (*Equus hemionus*) in the Mongolian Gobi desert as a case study. *Biological Conservation, 204*, 313–321.

Nazarova, L., Lüpfert, H., Subetto, D., et al. (2013). Holocene climate conditions in Central Yakutia (Eastern Siberia) inferred from sediment composition and fossil Chironomids of Lake Temje. Baikal-Hokkaido Archaeol. Proj. Environ. Arch. Proxies Reconstr. Approaches*, 290–291*, 264–274. https://doi.org/10.1016/j.quaint.2012.11.006

Novellie, P., Lindeque, M., Lindeque, P., et al. (2002). Status and action plan for the mountain zebra (*Equus Zebra*). In *Equids zebras asses horses status survey and conservation action plan* (pp. 28–42).

Orlando, L. (2015). Equids. *Current Biology, 25*(20), R973–R978. https://doi.org/10.1016/j.cub.2015.09.005

Orlando, L., Ginolhac, A., Zhang, G., Froese, D., Albrechtsen, A., Stiller, M., Schubert, M., Cappellini, E., Petersen, B., Moltke, I., Johnson, P. L. F., Fumagalli, M., Vilstrup, J. T., Raghavan, M., Korneliussen, T., Malaspinas, A. -S., Vogt, J., Szklarczyk, D., Kelstrup, C. D.,

. . . Willerslev, E. (2013). Recalibrating *Equus* evolution using the genome sequence of an early Middle Pleistocene horse. *Nature, 499*(7456), 74–78.
Osthaus, B., Proops, L., Long, S., Bell, N., Hayday, K., & Burden, F. (2018). Hair coat properties of donkeys, mules and horses in a temperate climate. *Equine Veterinary Journal, 50*(3), 339–342. https://doi.org/10.1111/evj.12775
Outram, A. K., Stear, N. A., Bendrey, R., Olsen, S., Kasparov, A., Zaibert, V., Thorpe, N., & Evershed, R. P. (2009). The earliest horse harnessing and milking. *Science, 323*(5919), 1332–1335.
Owen-Smith, N. (1987). Pleistocene extinctions: The pivotal role of megaherbivores. *Paleobiology, 13*, 351–362. https://doi.org/10.1017/S0094837300008927
Parrish, J. T., & Spicer, R. A. (1988). Late Cretaceous terrestrial vegetation: A near-polar temperature curve. *Geology, 16*(1), 22–25.
Payne, J. C., Buuveibaatar, B., Bowler, D. E., Olson, K. A., Walzer, C., & Kaczensky, P. (2020). Hidden treasure of the Gobi: understanding how water limits range use of khulan in the Mongolian Gobi. *Scientific Reports, 10*, 2989.
Petrov, K. A., Dudareva, L. V., Nokhsorov, V. V., Stoyanov, K. N., & Makhutova, O. N. (2016). The role of plant fatty acids in regulation of the adaptation of organisms to the cold climate in Cryolithic zone of Yakutia. *Journal of Life Sciences, 26*, 519–530.
Prescott, G. W., Williams, D. R., Balmford, A., et al. (2012). Quantitative global analysis of the role of climate and people in explaining late quaternary megafaunal extinctions. *Proceedings of the National Academy of Sciences, 109*, 4527. https://doi.org/10.1073/pnas.1113875109
Prins, H. H. T. (1998). The origins of grassland communities in northwestern Europe. In M. F. Wallis de Vries, J. P. Bakker, & S. E. van Wieren (Eds.), *Grazing and conservation management* (pp. 55–105). Kluwer Academic Publishers.
Prins, H. H. T., de Jong, J. F., & Geraeds, D. (in press). The evolutionary history of the African buffalo: Is it truly a Bovine? In A. Charon, D. Cornélis, P. Chardonnet, & H. H. T. Prins (Eds.), *African Buffalo: Ecology and management*. Cambridge University Press.
Rao, M. P., Davi, N. K., D'Arrigo, R., et al. (2015). Dzuds, droughts, and livestock mortality in Mongolia. *Environmental Research Letters, 10*(7), 074012.
Raymo, M. E. (1994). The initiation of northern hemisphere glaciation. *Annual Review of Earth and Planetary Sciences, 22*, 353–383. https://doi.org/10.1146/annurev.ea.22.050194.002033
Ricquier, D. (1999). Uncoupling protein-2 (UCP2): Molecular and genetic studies. *International Journal of Obesity, 23*(Suppl 6), S38–S42.
Rubenstein, D. I. (1989). Life history and social organization in arid adapted ungulates. *Journal of Arid Environments, 17*, 145–156.
Rubenstein, D. I., Cao, Q. S., & Chiu, J. (2016). Equids and ecological niches: behavioral and life history variations on a common theme. In J. I. Ransom & P. Kaczensky (Eds.), *Wild equids: ecology, management and conservation* (pp. 58–68). Johns Hopkins University Press.
Rubner, M. (1982). *The laws of energy conservation in nutrition*. Academic Press, Inc (London) Ltd.
Saarinen, J., Cirilli, O., Strani, F., Meshida, K., & Bernor, R. L. (2021). Testing equid body mass estimate equations on modern zebras—with implications to understanding the relationship of body size, diet, and habitats of Equus in the Pleistocene of Europe. *Frontiers in Ecology and Evolution, 9*, 622412. https://doi.org/10.3389/fevo.2021.622412
Sandom, C., Faurby, S., Sandel, B., et al. (2014). Global late quaternary megafauna extinctions linked to humans, not climate change. *Proceedings of the Royal Society B: Biological Sciences, 281*, 20133254. https://doi.org/10.1098/rspb.2013.3254
Schaller, G. B. (1998). *Wildlife of the Tibetan Steppe*. University of Chicago Press. https://press.uchicago.edu/ucp/books/book/chicago/W/bo3646086.html
Schoenecker, K. A., King, S. R. B., Nordquist, M. K., Nandintsetseg, D., & Cao, Q. (2016). Habitat and diet of equids. In J. I. Ransom & P. Kaczensky (Eds.), *Wild equids ecology, management, and conservation* (pp. 41–55). Johns Hopkins University Press.

Scholander, P., Hock, R., Walters, V., & Irving, L. (1950). Adaptation to cold in arctic and tropical mammals and birds in relation to body temperature, insulation, and basal metabolic rate. *The Biological Bulletin, 99*(2), 259–271. https://www.jstor.org/stable/1538742

Schulz, E., & Kaiser, T. M. (2013). Historical distribution, habitat requirements and feeding ecology of the genus *Equus* (Perissodactyla). *Mammal Review, 43*, 111–123.

Shackleton, N. (1967). Oxygen isotope analyses and Pleistocene temperatures re-assessed. *Nature, 215*(5096), 15–17.

Shah, N. (2002). Status and action plan for the Kiang (*Equus Kiang*). In P. D. Moehlman (Ed.), *Equids: Zebras, asses and horses. Status survey and conservation action plan* (pp. 72–81). International Union for Conservation of Nature and Natural Resources/Species Survival Commission, Equid Specialist Group.

Shah, N., & Qureshi, Q. (2007). Social organization and determinants of spatial distribution of Khur (*Equus Hemionus* Khur).

Shah N., St. Louis, A., & Qureshi, Q. (2015). *Equus kiang*. The IUCN Red List of Threatened Species 2015: e.T7953A45171635. https://doi.org/10.2305/IUCN.UK.2015-4.RLTS.T7953A45171635.en. Downloaded on 1 June 2021.

Shotwell, J. A. (1961). Late tertiary biogeography of horses in the Northern Great Basin. *Journal of Paleontology, 35*, 203–217.

Sjaastad, Ø. V., Sand, O., & Hove, K. (2016a). Aging in animals. In O. V. Sjaastad, O. Sand, & K. Hove (Eds.), *Physiology of domestic animals* (3rd ed., p. 891). Scandinavian Veterinary Press.

Sjaastad, Ø. V., Sand, O., & Hove, K. (2016b). Regulation of body temperature. In O. V. Sjaastad, O. Sand, & K. Hove (Eds.), *Physiology of domestic animals* (3rd ed., pp. 767–794). Scandinavian Veterinary Press.

Soest, V., & Peter, J. (1994). *Nutritional ecology of the Ruminant*. Cornell University Press.

Steegmann, A. T. (2007). Human cold adaptation: An unfinished agenda. *American Journal of Human Biology, 19*(2), 218–227. https://doi.org/10.1002/ajhb.20614

Sternberg, T. (2018). Investigating the presumed causal links between drought and dzud in Mongolia. *Natural Hazards, 92*(1), 27–43.

St-Louis, A., & Côté, S. D. (2009). *Equus kiang* (Perissodactyla: Equidae). *Mammalian Species, 835*, 1–11.

St-Louis, A., & Côté, S. D. (2014). Resource selection in a high-altitude rangeland equid, the kiang (*Equus kiang*): influence of forage abundance and quality at multiple spatial scales. *Canadian Journal of Zoology, 92*, 239–249.

Šturm, M. B., Ganbaatar, O., Voigt, C. C., & Kaczensky, P. (2017). Sequential stable isotope analysis reveals differences in dietary history of three sympatric equid species in the Mongolian Gobi. *Journal of Applied Ecology, 54*, 1110–1119.

Šturm, M. B., Smith, S., Ganbaatar, O., Buuveibaatar, B., Balint, B., Payne, J. C., Voigt, C. C., & Kaczensky, P. (2021). Isotope analysis combined with DNA barcoding provide new insights into the dietary niche of khulan in the Mongolian Gobi. *PLoS One, 16*, e0248294.

Tattersall, G. J., Sinclair, B. J., Withers, P. C., Fields, P. A., Seebacher, F., Cooper, C. E., & Maloney, S. K. (2012). Coping with thermal challenges: physiological adaptations to environmental temperatures. *Comprehensive Physiology, 2*, 2151–2202.

Uniyal, S., Awasthi, A., & Rawat, G. R. (2005). Biomass availability and forage quality of *Eurotia ceratoides* Mey in the range lands of Changthang, eastern Ladakh. *Current Science, 89*, 201–205.

Van Dierendonck, M. C., & Wallis de Vries, M. F. (1996). Ungulate reintroductions: Experiences with the takhi or Przewalski horse (*Equus ferus przewalskii*) in Mongolia. *Conservation Biology, 10*(3), 728–740.

Vilstrup, J. T., Seguin-Orlando, A., Stiller, M., et al. (2013). Mitochondrial phylogenomics of modern and ancient equids. *PLoS One, 8*, e55950. https://doi.org/10.1371/journal.pone.0055950

Vincent, E., & Berger, W. H. (1985). Carbon dioxide and polar cooling in the Miocene: the Monterey hypothesis. In E. T. Sundquist & W. S. Broecker (Eds.), *The carbon cycle and atmospheric CO2: Natural variations Archean to present* (Vol. 32, pp. 455–468). American Geophysical Union.

Wakefield, S., Knowles, J., Zimmermann, W., et al. (2002). Status and action plan for the Przewalski's horse (*Equus ferus przewalskii*). In *Equids: Zebras, asses and horses* (pp. 82–92).

Wang, H., He, Z. Q., Wang, H., & Niu, Y. X. (2012). Study on survival status of reintroduced *Equus przewalskii* in Dunhuang West Lake National Nature Reserve. *Journal of Gansu Forestry Science and Technolgy, 37*, 45–46, 65.

Weinstock, J., Willerslev, E., Sher, A., Tong, W., Ho, S. Y. W., Rubenstein, D., Storer, J., Burns, J., Martin, L., Bravi, C., Prieto, A., Froese, D., Scott, E., Xulong, L., & Cooper, A. (2005). Evolution, systematics, and phylogeography of Pleistocene horses in the New World: a molecular perspective. *PLoS Biology, 3*(8), e241.

White, M. A., Thornton, P. E., & Running, S. W. (1997). A continental phenology model for monitoring vegetation. *Global Biogeochemical Cycles, 11*(2), 217–234. https://doi.org/10.1029/97GB00330

Williams, S. D. (2002). Status and action plan for the Grevyís Zebra (*Equus Grevyi*). In *Equids zebras asses horses status survey and conservation action plan* (pp. 11–27).

Wu, X., Dong, S., Liu, S., Su, X., Han, Y., Shi, J., Zhang, Y., Zhao, Z., Sha, W., Zhang, X., Gao, F., & Xu, D. (2017). Predicting the shift of threatened ungulates' habitats with climate change in Altun Mountain National Nature Reserve of the Northwestern Qinghai-Tibetan Plateau. *Climatic Change, 142*, 331–344.

Xu, W., Xia, C., Yang, W., Blank, D. A., Qiao, J., & Liu, W. (2012). Seasonal diet of Khulan (Equidae) in Northern Xinjiang, China. *Italian Journal Of Zoology, 79*, 92–99.

Yang, S. X., Deng, C. L., Zhu, R. X., et al. (2020). The Paleolithic in the Nihewan Basin, China: Evolutionary history of an early to late Pleistocene record in Eastern Asia. *Evolutionary Anthropology Issues, News, and Reviews, 29*, 125–142. https://doi.org/10.1002/evan.21813

Yokoyama, Y., Lambeck, K., De Deckker, P., Johnston, P., & Fifield, L. K. (2000). Timing of the Last Glacial Maximum from observed sea-level minima. *Nature, 406*(6797), 713–716.

Yudin, N. S., Larkin, D. M., & Ignatieva, E. V. (2017). A compendium and functional characterization of mammalian genes involved in adaptation to Arctic or Antarctic environments. *BMC Genetics, 18*, 33–43.

Zhang, Y., Cao, Q. S., Rubenstein, D. I., Zang, S., Songer, M., Leimgruber, P., Chu, H., Cao, J., Li, K., & Hu, D. (2015). Water use patterns of sympatric Przewalski's horse and khulan: Interspecific comparison reveals niche differences. *PLoS One, 10*, e0132094.

Zhang, R., Zhang, T., Kelgenbayev, N., et al. (2017). A 189-year tree-ring record of drought for the Dzungarian Alatau, Arid Central Asia. *Journal of Asian Earth Sciences, 148*, 305–314.

Zhang, X. C., Shao, C., Ge, Y., Chen, C., Xu, W.-X., & Yang, W.-K. (2020). Suitable summer habitat of the khulan in the Mt. Kalamaili Ungulate Nature Reserve and estimation of its population. *Ying Yong Sheng Tai Xue Bao, 31*, 2993–3004. https://doi.org/10.13287/j.1001-9332.202009.032

Zimov, S. A., Zimov, N. S., Tikhonov, A. N., & Chapin, F. S. I. I. (2012). Mammoth steppe: a high-productivity phenomenon. *Quaternary Science Reviews, 57*, 26–45.