



Beyond the bones: What the dire wolf can teach us about evolution and conservation narratives

Anish Ganguly

Independent Author, Nottingham, United Kingdom

Abstract

Dire wolves (*Aenocyon dirus*) were a distinct species of large carnivores that thrived during the Late Pleistocene epoch. Their evolutionary trajectory reflects adaptation to megafaunal prey, with morphological features such as robust jaws and dentition suited for processing bone. However, their specialization may have contributed to their eventual extinction, as climatic shifts and the disappearance of large herbivores reduced available food sources.

The dire wolf's deep evolutionary divergence makes true de-extinction biologically unfeasible and ethically questionable. Creating genetic proxies risks ecological disruption and diverts resources from endangered living wolves. Conservation should prioritize extant canids, where proven strategies, genetic tools, and urgent threats make real biodiversity gains achievable, responsible, and immediately impactful.

Keywords: Dire wolf, quaternary, pleistocene rewilding, dextinction, conservation, genetic engineering

Introduction

The dire wolf (*Aenocyon dirus*) was one of the apex predators of the Late Pleistocene epoch, a period characterized by pronounced climatic fluctuations and the coexistence of diverse megafaunal assemblages (Dundas 1999; Barnosky *et al.* 2004; Perri *et al.* 2021) ^[1, 15, 42]. With an estimated shoulder height of approximately 90 cm and a body mass ranging between 60 and 70 kilograms, the dire wolf exceeded the size and robustness of the modern gray wolf (*Canis lupus*). Its heavily built frame and powerful jaws indicate adaptations for subduing large-bodied prey (Wang, Tedford and Barnes, 2009) ^[59]. The species occupied an extensive geographic range across both North and South America (Caro *et al.*, 2023), where it fulfilled a crucial ecological function within Pleistocene predator-prey systems. Stable isotope analyses and comparative functional morphology suggest that *A. dirus* primarily hunted large herbivores, including horses (*Equus* spp.), bison (*Bison* spp.), New World camels (*Camelops* spp.), and possibly juvenile mammoths (*Mammuthus* spp.) (Van Valkenburgh and Hertel, 1993) ^[57].

The extinction of the dire wolf approximately 13,000 years ago coincided with the broader terminal Pleistocene megafaunal collapse (Svenning *et al.* 2024) ^[56]. This event is widely interpreted as the result of interacting drivers such as rapid climatic shifts, significant reductions in megaherbivore populations, and increased ecological competition—particularly from more generalist canids and other carnivores capable of adjusting to changing prey communities (Yeakel *et al.*, 2024) ^[60]. For much of the twentieth century, the dire wolf's skeletal resemblance to the gray wolf led researchers to classify it within the genus *Canis*. (Nagaoka *et al.*, 2016) ^[39] Its robust cranial morphology, dentition, and overall body plan were considered sufficiently consistent with extant canids—

wolves, coyotes, and jackals—to justify this taxonomic placement (Nowak, 1979) ^[41]. More recent research has challenged this long-standing assumption, revealing that morphological similarity does not necessarily reflect close evolutionary relatedness (Gedman *et al.*, 2025)

Recent advancements in ancient DNA analysis have profoundly transformed our understanding of this enigmatic carnivore (Ray *et al.*, 2013) ^[44]. In a landmark study, Perri *et al.* (2021) ^[42] successfully extracted and sequenced both nuclear and mitochondrial genomes from dire wolf remains and compared them with genomic data from a wide array of extant and extinct canids. Their findings were striking: *Aenocyon dirus* diverged from other canid lineages approximately 5.7 million years ago, far earlier than previously assumed (Perri *et al.* 2021) ^[42]. Rather than being a close relative of the gray wolf (*Canis lupus*), as traditional morphological classifications implied, the dire wolf represented a deeply divergent lineage that had evolved in parallel with *Canis* species (Perri *et al.* 2021) ^[42]. The study also illuminated key ecological distinctions (Perri *et al.* 2021) ^[42]. Whereas gray wolves exhibit considerable dietary flexibility and occupy a wide range of habitats, dire wolves appear to have been more specialized predators that relied heavily on large-bodied Pleistocene herbivores (Perri *et al.* 2021) ^[42]. Their robust cranial and postcranial morphology, combined with inferred pack-hunting strategies, positioned them as effective exploiters of megafaunal resources (Perri *et al.* 2021) ^[42]. Yet this specialization likely limited their ecological resilience (Perri *et al.* 2021) ^[42]. Following the Late Quaternary megafaunal extinctions, dire wolves may have lacked the dietary and behavioral plasticity needed to adapt, contributing to their ultimate disappearance while gray wolves persisted (Perri *et al.* 2021) ^[42].

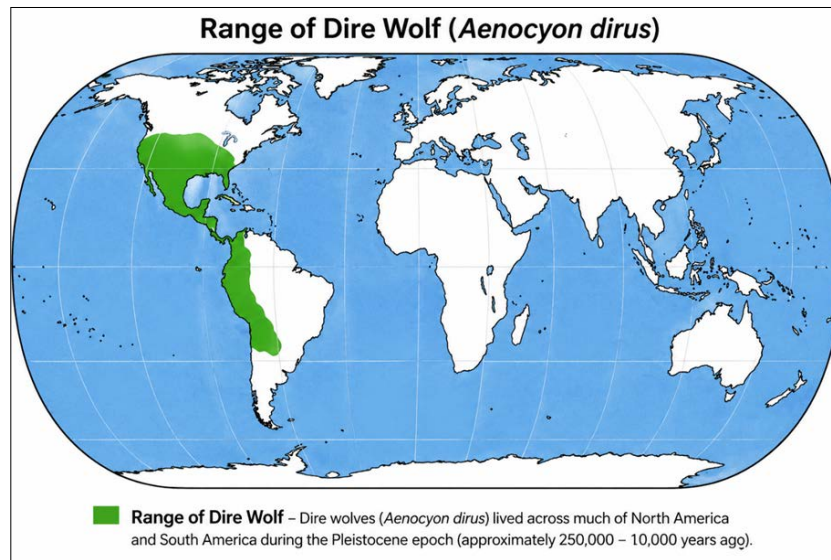


Fig 1: Range of Dire Wolf

The genetic evidence presented by Perri *et al.* (2021) ^[42] also supports the taxonomic resurrection of the genus *Aenocyon*—a name originally proposed in the 19th century—to reflect the dire wolf’s substantial evolutionary distance from *Canis*. The depth of genetic divergence indicates that interbreeding between dire wolves and modern wolves, coyotes (*Canis latrans*), or domestic dogs (*Canis lupus familiaris*) was highly unlikely (Jindrichova *et al.*, 2022). Consequently, the close morphological resemblance among these species is best interpreted as a compelling example of convergent evolution rather than shared recent ancestry (Losos, 2011) ^[34]. This genetic revelation carries significant implications for interpreting evolutionary relationships inferred solely from morphological traits (Losos 2011) ^[34]. Stable isotope analyses indicate that dire wolves relied heavily on large Pleistocene herbivores—including bison (*Bison* spp.), horses (*Equus* spp.), and camels (*Camelops* spp.)—whereas contemporaneous canids such as the gray wolf (*Canis lupus*) maintained more flexible and opportunistic feeding strategies (Coltrain *et al.*, 2004; Fox-Dobbs *et al.*, 2007) ^[10, 19]. Consequently, the terminal Pleistocene megafaunal extinctions, driven by rapid climatic change and increasing anthropogenic impacts, disproportionately affected dire wolves by eliminating the prey resources on which they were most dependent (Barnosky *et al.*, 2004) ^[1]. Morphological analyses further suggest that dire wolves were biomechanically optimized for subduing large, struggling prey but less adapted for pursuing smaller, more agile species (Perri *et al.*, 2021) ^[42]. This specialization placed them at a competitive disadvantage relative to more generalist predators—including gray wolves, cougars (*Puma concolor*), and expanding human hunter-gatherer populations—during a period of ecological upheaval (Van Valkenburgh & Hertel, 1993) ^[57]. Genomic data complement this picture: dire wolves diverged from other canid lineages roughly 5.7 million years ago and remained genetically isolated, lacking the introgressive hybridization that enhanced the adaptive flexibility of wolves, coyotes (*Canis latrans*), and domestic dogs (*Canis lupus familiaris*) (Perri *et al.*, 2021) ^[42]. Together, these lines of evidence suggest that the dire wolf’s ecological specialization, compounded by low genetic resilience and intensified

interspecific competition, ultimately limited its capacity to adjust to rapidly transforming Late Quaternary ecosystems (Neufeld, 2018). This case underscores a central lesson in paleobiology: morphological convergence can obscure deep evolutionary divergences, and anatomical similarities must be evaluated in conjunction with genetic evidence (Neufeld, 2018). It also reframes the extinction of the dire wolf as the terminal loss of a uniquely adapted but evolutionarily isolated lineage that lacked the flexibility exhibited by other canids as they transitioned into post-Pleistocene environments (Neufeld, 2018). This extinction pattern resonates strongly with modern biodiversity crises, in which ecological specialization and reduced genetic variability are consistently associated with heightened extinction risk (Margeay, 2012). Numerous contemporary species exemplify this vulnerability. The giant panda (*Ailuropoda melanoleuca*), for instance, is heavily reliant on highly specific habitat and dietary requirements, rendering it particularly sensitive to environmental disturbances and anthropogenic pressures (Cardillo *et al.*, 2005) ^[5]. Similarly, fragmented populations and declining genetic diversity in many modern taxa constrain their evolutionary adaptability—paralleling the dire wolf’s limited capacity to respond to rapid Late Quaternary ecological change (Hendry *et al.*, 2010). These observations contribute to ongoing debate surrounding the controversial concept of Pleistocene rewilding (Hendry *et al.*, 2010) ^[46]. Proponents of this strategy argue for the introduction of ecological analogues—or, more speculatively, genetically engineered reconstructions—of extinct megafauna to restore lost ecological processes and re-establish trophic interactions in degraded ecosystems (Donlan *et al.*, 2006) ^[14]. Within this framework, surrogate species such as gray wolves (*Canis lupus*) or bioengineered hybrids have been proposed as stand-ins for the dire wolf, with the aim of reinstating top-down regulation of herbivore populations and revitalizing ecosystem functions that have been absent since the terminal Pleistocene megafaunal extinctions (du Toit, 2019) ^[17]. This argument rests on the premise that many ecosystems in the Americas have remained functionally “empty” of their historical assemblages of large predators and herbivores since the collapse of these once-dominant species (du Toit, 2019) ^[17].

Table 1: Social and Behavioral similarity between Dire wolf and Grey Wolf

Trait	Dire Wolf	Grey Wolf	Comments
Pack Size	5-12 (avg-8)	4-12 (avg-6)	Similar Range
Social System	Family Based Pack	Family Based Pack	Likely Similar
Breeding	Monogamous Pair	Monogamous Pair	Similar
Cooperative Hunting	Yes(Megafauna Specialists)	Yes (varied prey)	Core trait similar
Territory Size	Large (hundred of km ²)	Large (varies with pray)	Dependent on pray density
Pup Rearing	Cooperative	Cooperative	Similar
Communications	Full canid repertoire	Full canid repertoire	Similar
Social Hierarchy	Present	Present	Similar

The concept of Pleistocene rewilding raises substantial ecological, ethical, and scientific concerns (Richmond *et. al.*, 2010) ^[46]. A central issue is the assumption that we can reliably reconstruct the ecological functions of extinct species. Despite significant advances in paleoecology, our understanding of Pleistocene predator–prey dynamics remains incomplete, and it is unlikely that any extant canid fully repli-cates the ecological role once played by the dire wolf (Richmond *et. al.*, 2010) ^[46]. Thus, substituting *Aenocyon dirus* with a modern analog such as the gray wolf (*Canis lupus*) may not restore historical trophic functions and could yield fundamentally different ecological outcomes (Middleton, 2022) ^[37]. Equally problematic is the possibility that introducing apex predators into ecosystems that have developed in their absence for millennia may lead to ecological disruption (Jorgensen, 2015). Many contemporary landscapes now sustain species assemblages, land-use patterns, and human infrastructures that differ profoundly from their Pleistocene counterparts (Richmond *et. al.*,2010) ^[46]. Introducing large carnivores could trigger unforeseen trophic cascades, intensify interspecific competition, and increase conflict with human activities (Jorgensen, 2015). Such interventions may ultimately create new ecological imbalances rather than restore past ones (Richmond *et. al.*,2010) ^[46].

From an evolutionary standpoint, the extinction of the dire wolf represents a unique and irreversible loss (Middleton, 2022) ^[37]. With its deep genetic divergence—estimated at more than five million years—from other canids, *A. dirus* occupied a distinct evolutionary and ecological niche that cannot be meaningfully replicated by living species (Perri *et al.*, 2021) ^[42]. Efforts to replace, mimic, or “simulate” the dire wolf run the risk of oversimplifying extinction and diverting attention from the anthropogenic drivers that continue to threaten global biodiversity (Middleton, 2022) ^[37]. These concerns highlight the broader ethical tension between technological intervention and ecological humility(Middleton, 2022) ^[37]. In recent years, however, rapid advances in genomics and synthetic biology have reignited debate about the feasibility of de-extinction technologies(Perri *et al.*,2021) ^[42]. The dire wolf has emerged as a prominent candidate for such efforts due to its iconic status and the recent sequencing of its nuclear and mitochondrial genomes (Perri *et al.*, 2021) ^[42]. Proponents suggest that, with CRISPR-mediated gene editing and cloning techniques, it may be theoretically possible to engineer an organism exhibiting some phenotypic and ecological resemblance to *A. dirus*, using a closely related canid—such as the gray wolf or the dingo (*Canis dingo*)—as a genomic template(Shapiro *et. al.*,2025) ^[54]. Yet this proposition remains highly controversial and fraught with scientific limitations (Shapiro *et. al.*,2025) ^[54].

Extant wolf species are confronting a complex conservation crisis characterized by habitat fragmentation, human-induced mortality, disease, and genetic erosion (Murray,2015) ^[38]. The Ethiopian wolf (*Canis simensis*), confined to isolated Afro-alpine regions, exists in only a few small subpopulations, severely limiting dispersal and gene flow (Lai,2025) ^[32]. These populations are extremely vulnerable to stochastic events, with disease outbreaks such as rabies and canine distemper causing sudden, high mortality (lai,2025) ^[32]. In addition, habitat conversion for agriculture, road mortality, and deliberate persecution further reduce population stability (Hindrikson,2015). The red wolf (*Canis rufus*) faces a different but equally severe challenge: hybridization with the coyote (*Canis latrans*) driven by low population densities and human-caused disruption of breeding pairs(Hindrikson,2015). This introgression threatens the genetic integrity of the species, while continued anthropogenic mortality, including shootings and collisions, exacerbates demographic instability(Hindrikson,2015). The gray wolf (*Canis lupus*), despite being more widespread, is under pressure from human-wildlife conflict, legal culling in response to livestock predation, and ongoing habitat fragmentation, particularly in regions where protections have been weakened (Hindrikson,2015). The Indian wolf (*Canis lupus pallipes*) and Himalayan wolf lineages (*Canis lupus chanco* or *Canis himalayensis*) similarly face habitat loss, prey depletion, and conflict with livestock owners, placing their populations at risk (Habib,2013) ^[22]. Collectively, these pressures create a convergence of ecological, genetic, and demographic threats that jeopardize the persistence of extant wolf species (Hindrikson,2015) ^[38]. Immediate, coordinated conservation measures that integrate habitat protection, population monitoring, genetic management, and conflict mitigation are essential to prevent further declines and preserve the ecological and evolutionary roles of these apex predators (Hindrik-son,2015).

In light of recent advances in the genetic resurrection of the dire wolf (*Aenocyon dirus*), it becomes increasingly important to critically evaluate the allocation of scientific resources. Biologically and ethically, prioritizing the conservation of extant canid species is imperative, as living wolves continue to fulfill essential ecological roles that cannot be replicated by resurrected species. The ongoing debate surrounding de-extinction raises fundamental questions: should scientific efforts focus on bringing back the dire wolf through advanced genetic technologies, or should they be directed toward the preservation and management of existing wolf populations that are currently threatened and remain integral to eco-system stability? Allocating resources to living species ensures tangible conservation outcomes, maintains biodiversity, and addresses urgent threats such as habitat loss, disease, and

human-wildlife conflict, whereas de-extinction projects, while technologically innovative, remain speculative and may divert attention from species that require immediate protection.

Colossal Biosciences Dire Wolf Project and its criticism:

The Colossal Biosciences Dire Wolf Project represents one of the most ambitious applications of synthetic biology and de-extinction research to date, aiming to approximate select phenotypic and behavioral traits of the extinct dire wolf (*Aenocyon dirus*) by genetically modifying modern gray wolves (*Canis lupus*) (Craw, 2025) ^[13]. Ancient DNA recovered from dire wolf remains dated between approximately 13,000 and 72,000 years old enabled researchers to assemble partial genomic profiles despite the substantial post-mortem degradation typical of Late Pleistocene material (Colossal, 2025a; CNN, 2025) ^[4]. Through comparative genomic analyses across multiple canid species, investigators identified twenty key loci within fourteen genes associated with traits such as increased body size, robust cranial structure, enhanced musculature, pale coat coloration, and distinctive vocalization tendencies (Colossal, 2025a; CRISPR Medicine News, 2025) ^[4, 11].

Using multiplex CRISPR–Cas9 editing, Colossal researchers introduced these variants into cultured fibroblast-derived cells from gray wolves (Colossal, 2025a) ^[4]. Fifteen of the engineered edits corresponded to allelic states inferred for ancestral dire wolf genotypes, while the remaining five targeted pigmentation pathways—including loss-of-function mutations in *MC1R* and *MFSD12*—to recreate the lighter coat phenotype hypothesized for some dire wolf populations (Wikipedia, 2025; Colossal, 2025a) ^[4]. Importantly, the company emphasized that no ancient DNA was directly transplanted; rather, modern wolf genomes were modified to approximate reconstructed dire wolf alleles (Colossal, 2025a) ^[4]. As chief scientist Beth Shapiro described, the resulting organisms constitute “gray wolves with 20 edits” (LiveScience, 2025), underscoring that these animals are engineered analogues rather than biological resurrections.

Following successful *in vitro* genome editing, nuclei from the modified cells were transferred into enucleated oocytes via somatic cell nuclear transfer (SCNT), producing approximately forty-five embryos that were subsequently implanted into domestic dog (*Canis lupus familiaris*) surrogates (Colossal, 2025b) ^[4]. This process resulted in the birth of three viable pups—Romulus and Remus in October 2024, and Khaleesi in January 2025 (BiopharmaTrend, 2025 ^[26, 56]; Wikipedia, 2025). Early phenotypic assessments indicate that these animals possess broader skulls, reinforced shoulder morphology, pale pelage, and projected adult body masses approaching ~68 kg, with body lengths of up to six feet—substantially larger than most gray wolves (People, 2025) ^[43]. Behavioral observations further report early-onset howling, heightened stalking instincts, and pronounced wariness toward humans, suggesting partial restoration of wild-type canid behavioral repertoires (BiopharmaTrend, 2025; People, 2025) ^[43].

Although the Colossal Biosciences initiative has been widely publicized as achieving the first successful “de-extinction birth” of dire wolves, the scientific community has emphasized several critical caveats. Genomic analyses demonstrate that dire wolves diverged from gray wolves more than five million years ago and are as genetically

distinct from modern canids as humans are from chimpanzees (Perri *et al.*, 2021) ^[42]. This deep phylogenetic separation underscores that the animals produced by Colossal are not true dire wolves but genome-edited gray wolves that incorporate a limited number of inferred ancestral alleles (LiveScience, 2025; Vox, 2025) ^[58]. Critics note that over twelve million nucleotide differences separate the two species, rendering a full genomic reconstruction of *Aenocyon dirus* impossible with current technologies (Vox, 2025) ^[58]. These scientific limitations are complemented by broader ethical concerns regarding animal welfare, ecological relevance, and opportunity costs—namely, whether significant financial and research investment would be better directed toward the conservation of threatened extant species (Business Insider, 2025; Vox, 2025) ^[4, 58].

A rigorous assessment by Jensen-Seaman *et al.* (2025) ^[26] further challenges claims of dire wolf de-extinction. Their analysis demonstrates that the Colossal project has produced genome-edited gray wolves rather than individuals of *A. dirus* (Colossal, 2025a) ^[4]. The authors critically examine the molecular and morphological evidence presented by Colossal, noting key limitations in the quality of ancient DNA, the reliability of phylogenetic inferences drawn from fragmented genomic sequences, and the functional interpretation of a small subset of phenotypic loci (Jensen-Seaman *et al.* 2025) ^[26]. They argue that editing a restricted number of putative trait-associated genes is insufficient to recapitulate the extensive genomic divergence characteristic of the dire wolf lineage (Jensen-Seaman *et al.* 2025) ^[26]. The paper also identifies methodological pitfalls—including uncertainties in specimen provenance, reference genome bias, and the difficulty of assigning complex traits to limited genetic edits—and explores the ethical implications of presenting phenotypic proxies as resurrected species (Jensen-Seaman *et al.* 2025). Jensen-Seaman *et al.* (2025) ^[26] ultimately conclude that, despite technological achievements, the animals produced by Colossal represent engineered analogues rather than a genuine re-establishment of *Aenocyon dirus* (Jensen-Seaman *et al.* 2025) ^[26].

In summary, the Colossal Biosciences Dire Wolf Project represents an extraordinary technical milestone in multiplex gene editing and mammalian cloning, successfully producing living canids that re-capitulate selected morphological and behavioral traits inferred for *Aenocyon dirus* (Jensen-Seaman *et al.* 2025) ^[26]. Yet the project simultaneously illustrates the conceptual constraints inherent to de-extinction efforts. While synthetic biology can generate “proxy organisms” that approximate certain phenotypic attributes of extinct species, it cannot recreate an evolutionary lineage that diverged millions of years ago (Jensen-Seaman *et al.* 2025) ^[26]. The dire wolf’s profound genetic distinctiveness underscores that true restoration of long-extinct taxa remains beyond the reach of current biotechnology, reinforcing the need for careful scientific and ethical consideration in future de-extinction initiatives (Craw, 2025) ^[13].

Current conservation trends of existing wolf species and its importance:

Extant wolf species, encompassing the gray wolf (*Canis lupus*), Ethiopian wolf (*Canis simensis*), red wolf (*Canis rufus*), Indian wolf (*Canis lupus pallipes*), and Himalayan wolf lineages (*Canis lupus chanco* or *Canis himalayensis*), represent a diverse group of apex and mesopredators that

continue to play vital ecological roles across the globe (Krofel, 2022) ^[31]. Their conservation status, however, varies considerably, and many populations face significant threats from habitat fragmentation, human-wildlife conflict, disease outbreaks, and genetic bottlenecks (Krofel, 2022) ^[31]. The Ethiopian wolf (*C. simensis*), for example, is considered one of the most endangered canids worldwide, with fewer than 450 mature individuals remaining in isolated Afro-alpine regions (Krofel, 2022) ^[31]. Populations in the Bale Mountains and surrounding plateaus are particularly susceptible to rabies and canine distemper outbreaks, which have historically caused dramatic mortality events, underscoring the fragility of these small, geographically restricted populations (Krofel, 2022) ^[31]. The Ethiopian Wolf Conservation Programme (EWCP), a collaborative initiative involving the Ethiopian Wildlife Conservation Authority, WildCRU, and local communities, has implemented a multifaceted approach to mitigate these threats (Lai, 2025) ^[32]. Key interventions include strategic translocations to form metapopulations, vaccination campaigns to control disease spread, habitat protection, and intensive community engagement to reduce human-wolf conflict (Lai, 2025) ^[32]. These efforts exemplify a holistic conservation model that combines biological management with socio-ecological considerations, demonstrating that focused, evidence-based interventions can maintain and potentially recover critically endangered wolf populations (Lai, 2025) ^[32]. Similarly, the red wolf (*C. rufus*), native to the southeastern United States, has been the focus of intensive captive breeding and reintroduction programs due to historical extirpation from most of its former range (Murray, 2015) ^[38]. The Red Wolf Recovery Program, coordinated by the U.S. Fish & Wildlife Service, maintains a genetically managed captive population to maximize diversity while preparing

individuals for reintroduction into suitable habitats (Murray, 2015) ^[38]. Wild populations, though limited and vulnerable, are monitored continuously for reproductive success, survival, and genetic health (Murray, 2015). This meticulous management is essential to prevent inbreeding depression and maintain population viability, particularly given the small number of remaining founders (Murray, 2015) ^[38]. Gray wolves (*C. lupus*), with their broader geographic distribution, have also benefited from concerted conservation initiatives (Firlein, 2018) ^[18]. In North America, the reintroduction of gray wolves (*C. lupus*) to Yellowstone National Park and central Idaho in the 1990s is a prime example of restoration ecology: wolves have since regulated ungulate populations, facilitated vegetation recovery, and influenced the behavior and abundance of other species, demonstrating cascading ecosystem effects (Firlein, 2018) ^[18]. European gray wolf populations, particularly in countries such as Germany, Italy, and France, are increasingly subject to legal protections, habitat corridors, and non-invasive monitoring, reflecting a growing commitment to maintaining viable populations and promoting coexistence with human communities (Firlein, 2018) ^[18]. Across Asia, the Indian wolf (*C. l. pallipes*) and Himalayan wolf lineages (*C. l. chanco* or *C. himalayensis*) are gaining attention from researchers who emphasize habitat conservation, genetic monitoring, and human-wildlife coexistence strategies to mitigate livestock predation and prevent retaliatory killing (Habib, 2013) ^[22]. Collectively, these examples illustrate that extant wolves, though challenged by anthropogenic pressures, benefit from a combination of scientific research, policy support, and community-based conservation, providing tangible evidence that coordinated action can prevent extinction and sustain ecological function (Habib, 2013) ^[22].

Table 2: Project outcome for species revival projects via biotechnological approaches

Species / Project	Technique	Results So Far	Status
Woolly Mammoth	CRISPR gene editing of Asian elephant genome	Mammoth traits (cold-adapted genes) inserted into elephant cell lines	No mammoth yet; target 2028+
Thylacine	Genome reconstruction + marsupial surrogate	High-quality genome assembled; gene-editing phase ongoing	No live animal yet
Dodo	Gene editing of Nicobar pigeon (closest relative)	Genome sequencing completed	No living dodo recreated
Pyrenean ibex	Somatic cell nuclear transfer (cloning)	One cloned kid born in 2003; died within minutes due to lung defects	First extinct animal briefly revived; project discontinued
Passenger pigeon	Gene editing of band-tailed pigeon	Genome mapping done; editing research ongoing	No living passenger pigeon yet
Northern white rhinoceros	IVF + stem cell research	Multiple viable embryos created; stored for implantation	No live birth yet
Pleistocene horse (attempted cloning)	Nuclear transfer from preserved remains	No viable clone produced	Project unsuccessful

From a biological perspective, prioritizing the conservation of living wolf populations is more compelling than allocating significant resources to de-extinction projects such as the resurrection of the dire wolf (*Aenocyon dirus*). Extant wolves, including the gray wolf (*C. lupus*), Ethiopian wolf (*C. simensis*), red wolf (*C. rufus*), Indian wolf (*C. l. pallipes*), and Himalayan wolf lineages (*C. l. chanco* or *C. himalayensis*), fulfill crucial ecological roles as apex predators and regulators of prey populations, influencing trophic dynamics and ecosystem stability (Firlein, 2018) ^[18]. By contrast, the dire wolf (*A. dirus*) inhabited Late

Pleistocene ecosystems that no longer exist, and its resurrected form—currently being pursued through synthetic biology by initiatives such as Colossal Biosciences—may not perfectly replicate the genetic, behavioral, or ecological traits of the extinct species (Jensen-Seaman *et al.* 2025) ^[26]. Ecological unpredictability is a substantial concern: reintroduced or genetically engineered dire wolves (*A. dirus*) could compete with existing predators, disrupt prey populations, or introduce unforeseen disease dynamics (Jensen-Seaman *et al.* 2025) ^[26]. In addition, many extant wolves face genetic challenges,

including inbreeding and loss of heterozygosity, which threaten long-term viability (Jensen-Seaman *et al.* 2025) ^[26]. Conservation interventions such as captive breeding, translocations, habitat protection, and genetic management are well-established strategies to address these risks, with clear evidence of effectiveness, whereas de-extinction remains experimental and highly uncertain (Jensen-Seaman

et al. 2025) ^[26]. Financially, the cost of de-extinction is prohibitive relative to conventional conservation programs; synthetic biology efforts require immense investment in genome editing, cloning, and animal husbandry, re-sources that could otherwise support critical habitat preservation, community engagement, and monitoring of living wolf populations (Jensen-Seaman *et al.* 2025) ^[26].

Table 3: Budget allocation and project outcome for species revival projects via traditional approaches

Species / Project	Conservation Method	Results So Far
California condor	Captive breeding, release, monitoring, lead mitigation	Population increased from 27 captured in 1987 to ~600+ birds with many birds flying free again; still ongoing management challenges.
Black-footed ferret	Ex-situ breeding & reintroductions	Recovered from near-extinction in wild; hundreds in captivity and hundreds back in wild sites across US & Canada.
Arabian oryx	Captive breeding (Operation Oryx) + reintroductions	From extinct in the wild by 1972 to established wild populations in several Middle-Eastern reserves and IUCN status improved.
Lear's macaw	In-situ protection, research, monitoring	Population grew enough to downlist from critical levels, habitat protection increased; economic benefits uneven.
Whooping crane	Captive breeding, migratory training, habitat management	Population recovered from ~15 birds mid-20th century to several hundred today (ongoing).
Mountain gorilla	In-situ habitat protection, anti-poaching, veterinary interventions	Population increased in protected parks; often cited as field success (exact totals varies by programme).

Ethically, focusing on extant wolves aligns with conservation priorities and moral responsibility (Murray, 2015, Firle, 2018) ^[18, 38]. Humans are largely responsible for the decline of many wolf populations through habitat destruction, persecution, and fragmentation, creating a direct ethical imperative to prevent their extinction. Extant wolf conservation programs, such as the EWCP (for Ethiopian wolves, *C. simensis*) or the Red Wolf Recovery Program (for red wolves, *C. rufus*), are grounded in social-ecological ethics (Murray, 2015) ^[38]: they actively involve local communities, respect indigenous knowledge, provide economic and educational benefits, and seek to reduce conflict between humans and wildlife. By contrast, de-extinction initiatives often operate within private or commercial frameworks, raising questions about equity, resource allocation, and accountability (Jensen-Seaman *et al.* 2025) ^[26]. While de-extinction may capture public imagination and offer potential technological spillovers—such as the development of tools for genetic rescue—its immediate ecological and ethical benefits are speculative (Jensen-Seaman *et al.* 2025) ^[26]. Resources allocated to resurrect a species that has been extinct for thousands of years carry an opportunity cost: funds, scientific expertise, and policy attention could instead strengthen the conservation of species currently facing imminent extinction (Jensen-Seaman *et al.* 2025) ^[26]. Furthermore, resurrected dire wolves (*A. dirus*) would not be identical to their Pleistocene ancestors, raising questions about authenticity, ecological appropriateness, and the fundamental goals of conservation (Jensen-Seaman *et al.* 2025) ^[26]. In contrast, extant wolves offer clear, measurable contributions to biodiversity, ecosystem function, and human-wildlife coexistence, fulfilling both ecological and ethical imperatives in ways that de-extinction cannot guarantee (Firle, 2018) ^[18].

Discussion

The dire wolf's divergence from all living canids is estimated at more than 5.7 million years, placing it not merely as a separate species but as a deep, ancient lineage with no close modern relatives (Perri *et al.*, 2021) ^[42]. Unlike the often-invoked mammoth–elephant model, there

is no extant canid sufficiently close to serve as a meaningful surrogate for genomic reconstruction (Jensen-Seaman *et al.* 2025) ^[26]. Any engineered organism would therefore be, at best, a functional approximation or genetic chimera rather than a true representative of *Aenocyon dirus* (Jensen-Seaman *et al.* 2025) ^[26]. This underscores a core conceptual limitation of de-extinction: while advanced synthetic biology can generate “proxy organisms,” it cannot reconstitute evolutionary identities lost millions of years ago (Jensen-Seaman *et al.* 2025) ^[26].

Beyond questions of feasibility, such efforts raise significant ethical and ecological concerns (Richmond *et al.*, 2010) ^[46]. Introducing reconstructed apex predators into modern ecosystems—ecosystems that have functionally reorganized in their absence for millennia—entails profound trophic uncertainty. Moreover, diverting resources from the protection of endangered extant species to speculative resurrection projects risks exacerbating ongoing biodiversity loss (Sandler, 2017) ^[50]. There is also a broader societal hazard: the perception that technology can reverse extinction may erode public understanding of the finality and gravity of species loss (Jorgensen, 2015) ^[38].

While ancient DNA research offers transformative insights into past ecosystems, the notion of rebuilding the dire wolf in a laboratory remains largely a symbolic provocation rather than a viable conservation strategy (Jensen-Seaman *et al.* 2025) ^[26]. A more constructive path lies in applying lessons from the dire wolf's extinction to present-day conservation. Protecting genetic diversity, maintaining habitat connectivity, and fostering ecological flexibility in threatened species are crucial for enhancing resilience in the face of accelerating climate change (Jensen-Seaman *et al.* 2025) ^[26]. The dire wolf's disappearance illustrates that even dominant predators can be rapidly lost when environmental pressures intensify and adaptive options are limited (Perri *et al.*, 2021) ^[42].

The genetic recreation of the extinct dire wolf (*Aenocyon dirus*) presents both potential opportunities and substantial challenges from a species conservation perspective (Jensen-Seaman *et al.* 2025) ^[26]. On one hand, advances in ancient DNA sequencing and CRISPR-based gene editing make it theoretically possible to reconstruct aspects of the dire wolf

genome, providing novel tools for studying evolutionary processes and exploring lost genetic diversity (Perri *et al.*, 2021; Shapiro, 2015) ^[42, 53]. Such technologies could also indirectly support conservation through “genetic rescue” efforts, by introducing adaptive alleles into endangered canid populations—such as the Ethiopian wolf (*Canis simensis*) or red wolf (*Canis rufus*)—to enhance resilience and reduce inbreeding depression (Ryder, 2019 ^[48]; Johnson *et al.*, 2010). Additionally, reintroducing a recreated apex predator might theoretically contribute to the restoration of trophic dynamics and ecosystem function, as demonstrated by the ecological effects following the gray wolf (*Canis lupus*) reintroduction in Yellowstone National Park (Ripple and Beschta, 2004) ^[47].

The potential drawbacks are substantial. Dire wolves were not close relatives of extant *Canis* species, having diverged millions of years ago; any recreated form would thus constitute an artificial hybrid rather than a true resurrection (Perri *et al.*, 2021) ^[42]. This raises critical questions regarding authenticity, ecological compatibility, and ethical justification (Jensen-Seaman *et al.* 2025) ^[26]. Introducing such organisms into contemporary ecosystems—already dramatically altered since the Pleistocene—could destabilize existing predator-prey relationships or competitively exclude native carnivores (Seddon *et al.*, 2014) ^[52]. Furthermore, critics emphasize that de-extinction initiatives may divert finite financial and scientific resources from the urgent protection of endangered species and habitats (Shapiro, 2015; Bennett *et al.*, 2017) ^[2, 53]. In this context, while the genetic recreation of the dire wolf illustrates the transformative potential of conservation genomics, it simultaneously highlights the ethical and ecological risks of creating novel organisms with uncertain roles in modern ecosystems (Revive & Restore, 2020; Sherkow and Greely, 2013) ^[45, 55].

The strategic focus on living wolves is reinforced by the practical successes and replicable methodologies developed over decades (Murray, 2015) ^[38]. Translocation programs, captive breeding initiatives, non-invasive genetic monitoring, habitat corridor establishment, and community-based coexistence strategies have demonstrably increased population stability, reproductive success, and survival rates among multiple species (Firlein, 2018) ^[18]. Moreover, cross-border collaborations, such as the Wolf Working Group and the Large Carnivore Initiative for Europe, provide frameworks for international cooperation, ensuring that conservation strategies are applied at ecologically meaningful scales rather than limited by political boundaries (Firlein, 2018) ^[18]. By contrast, de-extinction remains technologically experimental, ecologically untested, and socially contested, suggesting that investment in living species is both a scientifically responsible and ethically defensible priority (Jensen-Seaman *et al.* 2025) ^[26]. Importantly, emerging genetic technologies developed for de-extinction, such as genome editing and cloning, could still be leveraged to support conservation of extant species—for example, by introducing genetic diversity into small or inbred populations (Jensen-Seaman *et al.* 2025) ^[26]. In this sense, scientific innovation does not need to be abandoned; rather, it should be strategically applied to enhance the survival of species that are currently threatened and whose ecological roles are irreplaceable.

In conclusion, the global conservation of extant wolves, including the gray wolf (*C. lupus*), Ethiopian wolf (*C.*

simensis), red wolf (*C. rufus*), Indian wolf (*C. l. pallipes*), and Himalayan wolf lineages (*C. l. chanco* or *C. himalayensis*), represents a biologically urgent, ethically sound, and ecologically responsible priority (Firlein, 2018) ^[18]. Populations continue to face imminent threats, yet their preservation is achievable through established conservation strategies that integrate science, policy, and community engagement (Firlein, 2018) ^[18]. While the de-extinction of dire wolves (*A. dirus*) captures public imagination and demonstrates technological ingenuity, it remains speculative, costly, and fraught with ecological and ethical uncertainties (Jensen-Seaman *et al.* 2025) ^[26]. Prioritizing extant wolf conservation ensures immediate biodiversity protection, maintains essential ecological functions, and fulfills a moral obligation to species imperiled by human activity. Moreover, advances in genetic technologies can be applied judiciously to support endangered wolves without diverting attention from the pressing, tangible work of conserving species that are alive today (Shapiro, 2015) ^[53]. In this context, the protection and management of living wolf populations offer the greatest return on investment for global biodiversity conservation and provide a concrete pathway to preserving ecosystem resilience, human-wildlife coexistence, and the integrity of Earth’s natural heritage.

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