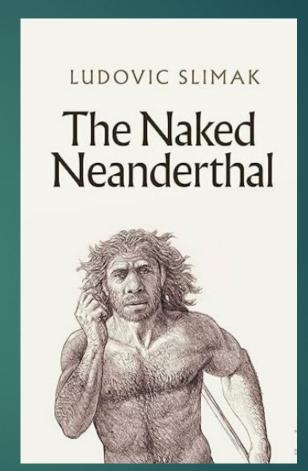
# The Naked Neanderthal by Ludovic Slimak, 2022

Summarized by Charles J Vella, PhD, November 2023

#### The Naked Neanderthal by Ludovic Slimak, 2022

- Ludovic Slimak has been a
   paleoanthropologist for 30 years at
   the University of Toulouse in
   France and director of the Grotte
   Mandrin research project.
- His work focuses on the last Neanderthal societies, and he is the author of several hundred scientific studies on these populations.
- He has excavated at Mandrin since 1998.





# Ludovic Slimak: early biography from long interview he gave

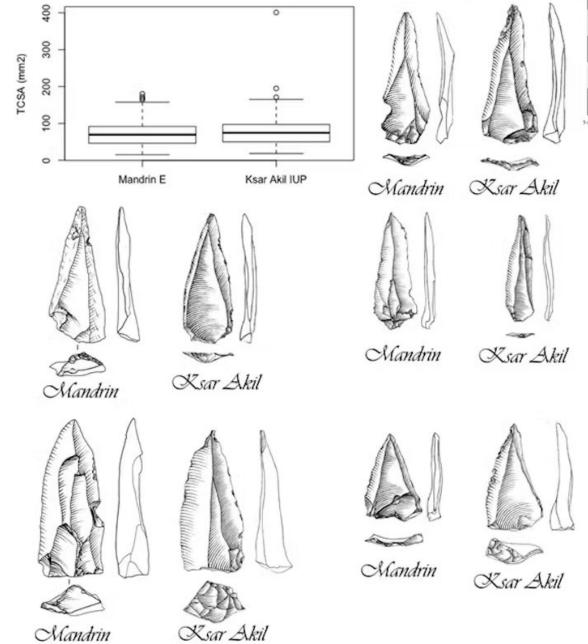
- ➤ At the age of four, Slimak was asked by his father what he'd like to do when he grew up. "I said I wanted to make holes in the ground to find old things. I didn't know it was a job, until he told me about archaeology." He's been at it ever since.
- ➤ Slimak was born in 1973; his father was a forester. His early years were spent surrounded by trees, as the family moved across France. "My grandfather lived in the Pyrenees. He was born in 1918, but really, he was a man from the 19th century. I spent so much time with him that I also feel like a man from another era, lost in the modern world."
- ➤ As creatures, Neanderthals were far more creative than Homo Sapiens.
- ➤ By 10, Slimak had talked his way into various archeological digs he'd come across near their home. At 14, he was already something of an expert. "By 18, I was working on a dig here in the Rhône Valley, at a Neanderthal site maybe 70km north."

#### L. Slimak

- ➤ "From then on, I knew I wanted to dedicate my entire life to these creatures." At first, university didn't feel a fit for this born outdoor explorer. In his 20s, he realized a degree would help him carve out a career, so enrolled in a course at Aix-Marseille University.
- ➤ To help pay his way, Slimak learned to play the bagpipes after writing to Glasgow's College of Piping, and through busking and playing in Marseille's premier late-90s Celtic band earned enough to keep his research afloat. In 2004, he completed his PhD and was soon recruited by Stanford University, before being hired by France's prestigious Centre National de la Recherche Scientifique, where he's worked ever since.
- ► His Neanderthal hunting has seen him direct digs everywhere from the Horn of Africa to the Arctic Circle.

#### L. Slimak

- ▶ In early May, he published a potentially groundbreaking paper claiming that Homo sapiens had not colonized Europe in one, but in three distinct waves between 54,000 and 42,000 years ago. According to this viewpoint, each migratory wave yielded its own archeological culture: the Neronian (54,000 years ago), the Châtelperronian (between 45 and 46,000 years ago) and the Proto-Aurignacian (42,000 years ago)
- ▶ The May 2023 paper explains that what we thought to be the first wave of colonization of Sapiens from the Near East to Europe was in fact the last of three waves. In the process, Homo sapiens interacted intermittently with the Neanderthals over thousands of years. It's a large view of continental Europe till the Eastern Mediterranean coast, which claims that we have missed something huge and what we saw in the Rhône Valley is only the visible tip of misunderstandings on the early Sapiens' presence in the continent.



It is the similar phases in tool technologies from both regions that led me to believe they were spread from the Near East to Europe during three distinct waves of colonization. This precise community of knowledges and traditions induced that the Neronian was in fact the archeological indication of a very early migration of Sapiens in Europe, far before expected and I published these conclusions in 2017.

Flint points from Grotte Mandrin in France and Ksar Akil in Lebanon. Drawings and measurements by Laure Metz and Ludovic Slimak, Fourni par l'auteur

# Mandrin Grotto



🗅 Digging deep: the Grotte Mandrin site. Photograph: Grégoire Bernardi/The Observer

#### Grotte Mandrin

- ▶ These findings could not have been possible without the other papers we have published in the past year and a half. The first one, <u>"The Modern Human Incursion in Neanderthal Territories"</u>, shows that we find Homo sapiens in the Rhône Valley as early as 54,000 years ago, while we thought that for all continental Europe, Homo sapiens would have come by 45 to 42,000 years ago.
- ▶ We published another major paper, <u>"Bow and arrow technology of the first modern humans in Europe"</u>, that gives the technical and cultural context of these societies. Again, we claim the bow and arrow technology emerged 40,000 years earlier in Eurasia than was previously estimated.
- Analyzed and published the 9 hominin teeth we found over 30 years in Mandrin. They come from different phases of occupations of the cave spanning 42,000 to 120,000 years. At this age, all these teeth should have only been from Neanderthals. But this was not the case. 1 tooth was archaic H. sapiens

#### **Grotte Mandrin**

- Mandrin Rock Shelter: It's a vaulted rock shelter that opens to the north that overhangs the Rhône Valley. And what is very important from an archaeological perspective in the Rhône Valley is its very strong, cold, Northern wind the Mistral.
- ► The Mistral was already blowing in the time period I research. Back then, the climate in Europe was Polar, so there were no trees and very little vegetation. When the Mistral blew, it took the sand and the silt from the river in the Rhône Valley and cast it in the rock shelter, depositing it year after year.
- ▶ I like to say it's like Pompeii but instead of a catastrophic event, we have sand and silt. And instead of one event, we have 12 events: 12 major archeological periods in the site that range from a climatically very warm period, the last interglacial, to the extinction of Neanderthal 42,000 years ago.

#### L. Slimak theories

▶ In the first wave dating back to 54,000 years ago, what we see in Grotte Mandrin is that the Sapiens population must have stayed for one generation, something like 40 years. They are in Neanderthal territories, but they won't stay there for another 12,000 years. After that, we will have other Neanderthals. The question of their relation is something fascinating, because when you have a look at the DNA of any early Sapiens in Europe, we see that all these early Sapiens have Neanderthal DNA. But if we focus on the last Neanderthals, we realize that there's not a single Neanderthal with a recent Homo Sapiens DNA.

➤ Slimak: What happened? Why do we have all Sapiens in Europe with Neandertal DNA and not a single Neandertal have Sapiens DNA?

#### CJV: There was MH DNA in Ns

- ► For many years, the only evidence of human-Neanderthal hybridization existed within modern human genes.
- ➤ 2016 researchers published a new set of Neanderthal DNA sequences from Altai Cave in Siberia, as well as from Spain and Croatia, that show evidence of human-Neanderthal interbreeding as far back as 100,000 years ago farther back than many previous estimates of humans' migration out of Africa (Kuhlwilm et al., 2016). Their findings are the first to show human gene flow into the Neanderthal genome as opposed to Neanderthal DNA into the human genome.
- ► Harris, et al., 2023: 6% of N Genome from MH DNA from ~250 KA. Every Neandertal sampled after this time had a mitochondrial genome derived from Africa; every individual with a Y chromosome was likewise part of an African clade.
- ► Evidence that some of the African-to-Neandertal gene flow was deleterious.

# Absence of reciprocity: "I give you my sister, you give me your sister."

Slimak: So we know from Claude Lévi-Strauss' Elementary Structures of Kinship that the question of the reproduction of societies is not a question of love. It's a question of exchanges and alliances between populations. So that means that when two groups meet, it's very important for them to exchange genes. And we know from DNA how they do it, it's universal for both Neanderthal and Sapiens: through female mobility. That means: "My sister will go in your group, but your sister will come in my group". And like that, we will build an alliance - we call this patrilocality. But if your sister comes in my group, my sister will have to come in yours. I can't have your sister in exchange of flint or 10 horses.

# Population encounters

- ▶ One possible instance when we see this is when there's <u>a total war</u> between populations, and one group is going to seek to destroy another group. But in fact, it's not really a genocide, because when that happens, traditionally what they do is that they keep the children and the women, and then they have children with these women.
- ▶ Another scenario could have been that these two populations had very good relations, where you're happy when you see fresh blood coming because you are very tiny group, very isolated, and suddenly you see a new group and say: "Oh, there's fresh blood coming" and that's very good news.

# **Populations**

- ▶ And the two populations certainly tried to exchange genes, but we know from DNA that Sapiens and Neanderthal were separated by 300,000 to 500,000 years of genetic distinction and what we call their inter-fecondity was very partial. This means that if they had children, for example, those children could be boys, sterile or not able to survive. So I would say it's very likely that the two populations met and tried to exchange genes in Europe, but that only worked very partially.
- ▶ I would also like to add that while Sapiens' tools may be more efficient, Neanderthals' are more singular. If you take crafts from Homo Sapiens, for example, 100 tools or 100 flints from 50 to 100,000 years ago, the 10,000 tools or flints after will be exactly the same. The population has a very clear project in their mind and regardless of the natural geologies, the environment, the climate, they reproduce the same thing.

#### N tools

- ▶ But if you take a Neanderthal tool in comparison, and then you analyze a million after that in the same layer, in the same societies, they are all completely different. Each tool is a specific creation. There's an incredible creativity among Neanderthals. And there's also a total absence of standardization that we find in our ancestors and in our contemporary societies.
- ➤ Slimak: we have projected all our fantasies on the Ns, saying: "Look, we have been racist, in fact, Neanderthals are just like us". But the 30 years I have spent in caves and the millions of flints I have seen tell a different story. It's not at all a humanity that is like us.

# Critique of Slimak

➤ Critique in interview: While your scientific colleagues recognise your research as ambitious, not everyone is convinced. You said that there was 100% certainty about the identification of that broken molar, but others will say that it could also be a shaped tooth of a young Neanderthal.

► Likewise, some are skeptical that the sophisticated tools that we found in the Grotte Mandrin, the Châtelperronian tools, were the handicraft of modern humans and not the Neanderthals. What is your answer to them?

# It's not only one tooth, it's millions of flints.

- ► The objection that the research is not clear and only based on one tooth. Well, no, it's not only one tooth, it's millions of flints.
- ▶ And even if we did not have any hominin remains, we would be able to identify these artefacts as Sapiens'. Like, for example, for the Aurignacians (35,000 years ago) or the Proto-Aurignacians (42,000 years ago), we did not have any teeth for years. Now I think we have two or three for all Europe and in the Levant we have two or three very isolated teeth, but before we find these teeth everybody was happy and was saying: "Well, it's clear it's absolutely homo Sapiens because we have this connection with the Near East."

#### The Last Neanderthal

- ➤ Slimak's central argument, a synthesis of decades of his own and others' research, is that Neanderthals possessed a distinctive form of intelligence in some ways superior to that of *Homo sapiens*. The author explains how recent discoveries have informed a reassessment of this species' social and artisanal practices, and he offers adventurous speculations on the dimensions and meaning of its cognitive endowments.
- ▶ Discusses how our own human prejudices have limited our appreciation of the Neanderthals' achievements, a perceptual blindness he convincingly relates to modern forms of racism. Slimak shows how we have much more to learn about ourselves by studying "exotic sensibilities" and more fully acknowledging "our nature not as humanity but as a humanity. A contemplation of human otherness.

# Slimak does not like the "New" Neandertal Imaging

- ► "The Neanderthal no longer exists, except in our minds," writes Ludovic Slimak in *The Naked Neanderthal*
- ► For Slimak, the rehabilitated view is equally problematic, based not on true understanding, but on a construct that says more about us than them. "We have sought projections of ourselves in the Neanderthals," he writes, arguing that their consciousness was fundamentally different from our own.
- ➤ Surprisingly, Slimak is skeptical of the evidence that Neanderthals created cave art and symbolic objects such as necklaces of shell beads. Likewise, while he has no doubt they buried their dead, he argues this won't help us understand them, as we know that empathy and sorrow for the deceased is shared by some other animal species.

#### Mandrin Rock Shelter

- ▶ Instead, he finds unique insights through an exhaustive excavation he conducted of a rock shelter near Mount Ventoux in France a Rosetta Stone of the Neanderthal world. The excavation turned up some astonishing discoveries in a 12-metre-deep patch of soil dating from between 80,000 and 123,000 years ago, which records events in a time of great climatic change.
- ➤ Trapped in the sand were the well-preserved remains of hunts there were 61 different prey species, from lions to turtles. Neanderthals not only hunted dangerous beasts but also ones with more complex behaviors, which entailed very different hunting strategies. This, says Slimak, contradicts the idea that Neanderthals couldn't adapt to target different prey in the face of climate change one proposed explanation of their demise.

# N being in the world

- ▶ Most fascinating was evidence in a layer of soil deep in the cave, where there is no sunlight. Here, the fossil bones came from deer carried in by Neanderthal hunters. But while most hunts include young and old, male and female prey in equal measure, this layer contained only mature male deer, with a large number of skulls with antlers attached possible hunting trophies. For Slimak, this points to a Neanderthal ritual: the systematic targeting of the trickiest specimens to kill, perhaps as a rite of passage.
- ▶ Demise of Ns: having rejected a failure to adapt under climate change, Slimak places the blame firmly on the superior weaponry of *Homo* sapiens. "There is no objective, logical, rational reason why these populations, which evolved independently for hundreds of millennia, should have developed ways of being in the world which are the same as ours."

#### **Grotte Mandrin**

"Finding Homo Sapiens sandwiched between Neanderthal occupants in these caves? It totally reshapes our understanding of our origins and rewrites what we've believed previously. If both species brushed up against each other over this long period of time, far more important than what happened to Neanderthals, we should be asking: what did these two species do together? Did they communicate? And most importantly, how did they interact? Because Neanderthals experienced and existed in the world differently to our ancestors. Not just by culture, but by their very nature."

#### N and MH differences

► He points to the way prehistoric Homo Sapiens and Neanderthal crafts are vastly different. "We might not know much about Neanderthals," he goes on, "but through what they created, we can see something incredible. When you take Homo Sapiens tools made of flint, spanning tens of thousands of years, in different parts of the world, they're always the same. Standardized. It can't be cultural." There was likely little contact between these different settlements. "There's something innate within the behavior of Homo Sapiens – within our behavior – to act and think in a certain way. It's in our nature." Neanderthal crafts, though, don't share this pattern of standardization. "Look carefully at Neanderthal tools and weapons. They're all unique. Study thousands and you'll find each is completely different. My colleagues never realized that. But when I did, I saw there was a deep divergence in the way Homo Sapiens and Neanderthals each understand the world."

# Flints at Grotte Mandrin



# Ns were more unique than MHs

Slimak feels this comparison can and should be made with Neanderthals. "Their tools and weapons are more unique than ours. As creatures, they were far more creative than us. Sapiens are efficient. Collective. We think the same, and don't like divergence. And I don't just mean western culture. Go to any Aboriginal society: there are clear rules and customs, and shared styles of clothing. Expectation to act in a certain manner; to follow regulations." Our ancestors, he says, lived like this instinctively. "You don't see that with Neanderthals." By seeing Neanderthals as a reference point against which we can measure ourselves, Slimak reckons humanity is offered a gift: "We have an opportunity to look in a mirror and see ourselves for what we truly are. To help us redefine, which we must do urgently."

# MH efficiency

- ► The way he sees it, this <u>isn't just an interesting philosophical theory</u>. "Neanderthals vanished, I think, because of high human efficiency. And this <u>efficiency now threatens to destroy us, too</u>. That's what's killing the planet's biodiversity." For Slimak, *The Naked Neanderthal* isn't a history book. "It's about us in the present. Urging humanity to see itself for what it is by comparing us to something else, in the hope of changing the course of our future. Because by understanding our nature and the risk this efficiency poses we can save ourselves from a similar fate."
- Over millennia, humankind has also developed an advanced, impressive technology and culture, of a type Neanderthals could never have imagined. "So while there is something dangerous in our nature, as a collective we can control and reshape it. Understanding this is the key to humanity's future. Because if we don't think carefully, next time it won't be Neanderthals that our efficiency destroys, it'll be humankind itself that's the victim."

#### Charlie Vella's reaction to new book

- ► Ludovic Slimak's new book about his view of Ns is poetry and philosophy, as much as paleontology.
- ► Slimak knows his French poetry, existential phenomenology, Heidegger, Sartre, and Levi-Strauss.
- Emphasis of N's "Being in the world"
- ► He is absolutely definitive in his opinions. He dismisses other scientific opinions in single sentences.
- ► Claims only field researchers (like himself) know N best; not lab, nor genetic, studies.
- ► He dismisses N art, N hunting weapons

# The Naked Neanderthal by Ludovic Slimak, 2022

- ► Chapter 1: A Neanderthal in Our Heart and Soul: Another Intelligence
- ► The Ns were never other versions of us—not brothers, not cousins—when it comes to mental structures, but an utterly different humanity. To approach them is to encounter a fundamentally divergent consciousness.
- "As a bare minimum, you need to have directly extracted evidence from these cave archives for decades in order to say anything worthwhile about this extinct humanity."
- ► Current battle: Ns as another us vs an archaic form of humanity with lower intellectual abilities.
- ▶ Their extinction raises the specter of our own extinction.

#### Ns are extinct

- ▶ N extinction is proven fact, despite 1-2% DNA kin modern humans; Ns did not survive. Genetic leftovers only represent distant encounters between biologically diverse populations which were only partially fertile.
- ► That humanity is totally extinct. Not a genetic dilution in us. They are not dissolved into us. Slimak gives date of N extinction as 42 Ka.
- ➤ Our dating methods are two imprecise to establish any relationship between colonization of Europe and the extinction of Ns. There will be no research to resolve this lack of klg anytime soon.

# Did N go extinct at 40 Ka?

▶ No new N skeleton in France in last 50 years.

- MH art has singularly bridged our history. Picasso, having seen Altamira in Spain, said "They invented everything." Rock frescoes that speak across the ages. Unlike N art.
- ► The Neandertal could not have been human in the way we imagine. Ns are not versions of ourselves

► There are no archeological sites in Europe for which our temporal measurements are sufficiently precise to establish with any certainty that MHs and Ns encountered each other.

#### His criticisms

▶ Key N archives are based on old excavations before 1979. Bones in boxes are decontextualized; often not collected based on modern archeological methods. The Gibraltar data are based on archeology, not N bones. Mousterian tools still appear at 30 Ka in Europe. He critiques C14 datings.

- ► Huge number of conclusions derived from molecular analyses are debatable.
- ► At Mandrin, it has taken, using brushes 2 to 4 months a year for 30 years to reach 60 centimeters of sediments

# Chapter 2: A Boreal Odyssey

- European Artic Circle: While Northern Europe was covered in ice, Northern Russia and boreal Siberia was not, due to lack of Atlantic ocean precipitation; no ice sheets developed during the ice ages. The mammoth steppes. Cold, but markedly dry region.
- Inuits: cold is never a problem for humans, only lack of protein.
- ➤ Slimak does not accept the need for technical inventiveness to adapt to cold regions. He worked in -25°C areas without any difficulty.
- ► The absence of Ns above the 55<sup>th</sup> parallel lead to early theories that Ns could not exist there. And that climate change lead to their extinction.
- ▶ Slimak believes human bodies are highly adaptable to climate change. Colonization at very high altitudes calls into question climate-based theories and the idea that they had limited adaptability. Paleolithic societies did not need special technological or social means to deal with diverse biotropes. Their bodies did most of the work.

# Russian polar steppes

- Russia is world's largest country. Half is boreal forest; the largest on the planet.
- Current melting of permafrost is both revealing and destroying archeological evidence.
- Only 3 recognized Arctic archeological sites older than 20 Ka. All in Russia. Manotovaya Kurya at 40 Ka. Only 7 stone tools and a 1.3 meter mammoth tusk with regular incisions. Unclear if N or MH.
- ▶ Polar hunters at 48 Ka: Taymyr peninsula entire frozen mammoth with human stone cutmarks an unknown human population
- ➤ Yana RHS (Rhino Horn site) deposit, at 30 Ka, 500 Km north of Arctic circle treeless steppe tundra, permafrost: 1000s of carved ivory and flint pieces; 1000s of hares (soft skins removed); a MH group, the Ancient Northern Siberians contained long strands of N DNA (80 to 100 generations prior), but no D DNA

# Byzovaya



# Polar enigma – did Ns live in Arctic

- Permafrost has produced wide variety of artic fauna: mammoths, rhinos, muskox, bison, brown bear, horse, reindeer, polar foxes
- ➤ Komi Republic drawers with collections from Byzovaya in 2007 filled with Mousterian stone tools dated to 28 Ka; definitively dated multiple times (Tom Higham); no animal bone or ivory pendants like at Yana; no interest in tusks, only in flesh
- ▶ Why were there Mousterian tools dated to 28 Ka in Artic circle?
- Slimak thins decorations, jewelry, representations, are the exclusive domain of sapiens

# Nordic Transgressions

- ▶ Byzovaya: Polar refuge of last Ns? 1000 km north of Europe
- ► Lavallois stone tools dated to 28.5 Ka; The Mousterian in Europe has only been found with Ns; but no human fossil bones at Byzovaya; hunted mammoths and wolves
- ► So paleolithic people of Byzovaya remains unknown
- ► MH Yana RHS site is 2000 km to east of Byzovaya; had N ancestry
- ► In 2008, discovery of Ust-"Ishim, oldest MH at 45 Ka; N ancestry from 2 Ky before; no D DNA; no descendants

## Cannibals in the forest: If I eat you, is it love or hunger?

- ▶ 100 Ka ago, Ns in the forest were cannibals.
- Anthropophagy = eating human bodies
- ► Catholic Church: body and blood of Christ
- Cannibalism was universal
- ▶ Néron Cave, France: Ns; burnt faunal and human bones; 120 Ka
- ▶ In 1993, thought it was nutritional cannibalism
- ➤ Spent 2 months a year for 6 years redigging the original hole below the cave; in 1999, published about 78 bones from 6 individuals

#### Cannibals in the forest: cannibalism at Moula

- ▶ 2019 study of Moula: claim of nutritional cannibalism due to population crash and famine; he disagrees
- ▶ There were 80 temperate European sites, not just 5
- ▶ Use of temperate & polar faunal to prove age does not work here
- ▶ Post glaciation, temperature was 3°C warmer than today; warmest in 400 millennia; he thinks it was not nutritional cannibalism
- ▶ Notes that Inuit have never practiced cannibalism; no known acts among hunter gatherers occupying their traditional territories, in areas that they knew well. No horse hunting Ns of the steppes ever found themselves transported into vast temperate forests teeming with deer.
- ► At Moula, only 1 in 4 fauna were stripped of flesh, compared to every N bone found.

#### No answers

▶ "When it comes to Ns, any evidence of rituals, symbols or actions of a spiritual nature is singularly lacking. Did they even bury their dead? That question may never be answered. Given the significant rarity of N remains, the very question as to the existence of burials divides the scientific community into two passionately, but robustly opposed camps.

▶ Did they ritually strip their dead of their flesh? Once again, the Ns slip through our fingers. There is no simple answer. No obvious answer. To the best of our knowledge... Everyone has an opinion. Everyone draws their own conclusions."

## 4. Rituals and Symbols

➤ Symbols = art, adornment, graves, rituals

Slimak believes Ns buried their dead. And that they cared for their elderly and disabled.

▶ Death of chimpanzee Pansy indicate awareness of life and death, csness of self and others, empathy and empathy and filial love in chimpanzees. These behaviors do not distinguish humans from animals. Altruism and empathy are documented in most mammals.

▶ These behaviors do not help us to distinguish between Ns and MHs.

#### Climate

► When it comes to Ns, climate change statistics have very large confidence intervals = large timing variation. "..more often than not, an inexact process."

▶ 130-80 Ka = no archeological sequence which records the whole of reversal of climates during this period

### A Temporal Fault in the Rocks: Mandrin Grotto

- ➤ 2008: A Rosetta Stone of the People of the Forest: Mandrin Grotto, a limestone cave that perfectly preserved all fossils, 100 Ka, highly forested environment
- ► More beaver fossils than anywhere in Europe
- Originally 1.5 feet high; 2 months of excavation for 8 years; 12 square meters of archeological material; 15 distinct layers; 61 different species, one of the most biodiverse finds in Europe
- ▶ 123 Ka at bottom, 80 Ka at top, 12 meters above; most complete sequence of this interglacial period; 4 carnivores indicated large biome to feed them
- ► Flint tools: fur-making, meat cutting; ochre to disinfect & soften leather

## Mandrin Cave today: originally you had to crawl



## Hunting with meaning; rites of passage

- Subterranean area two meters thick with fossilized hyena coprolites; Ns hunted hyenas in the cave
- One area with only fully mature male deer fossils; antler hunting trophies; as well as bears, lynx, wolves.
- N hunts were not just about protein, but ritualized, invested in codes that go beyond feeding. No other carnivore hunts with systematic selection of prey based on gender and age. This is distinctly human.
- ➤ Slimak has several paragraphs where he criticizes scientific positivism, the quantification of information vs human interpretations.

## Symbolism

▶ Symbolism as bad list of ingredients. "Take one pinch of burial, add three pinches of bodily adornments, one pinch of burial, a bit of moveable art, leave to simmer for 100 millennia and you end up with a tasty modern human ready to express deep symbolic thought."

► He thinks N hunting, cannibalism, etc., had ritual components.

#### Neandertal Aesthetics: Illusions turn to dust

- Current theory of Ns as same as MHs, and only our prejudices prevented us from seeing its true humanity.
- ▶ But there is incredibly little N art.
- ► All interpreted as Ns expressed themselves visually and symbolically. Restoring them to full humanity. Our humanity.
- ► Closer analysis turns these illusions to dust. Not a single N aesthetic artifact is manufactured; none bears the slightest trace of a deliberate artisanal modification. No ornament has a drilled hole. We are not dealing with objective facts, but rather interpretations, projections and constructions.

#### No N aesthetics

- Bower birds of New Guinea collect objects to attract females; N never did this behavior. N collecting of unusual objects is shared with Australopithecus.
- Believes all N pierced shells were natural (pierced by crabs). No evidence they were worn as adornments.
- ▶ No evidence that eagle talons were worn as a necklace. No evidence of adornment in Ns. No N necklaces.
- ▶ Believes N use of feathers was for nutrition like the Inuits use them.
- ► The presence of ocher is ubiquitous used as sunscreen, skin work, to grip tools. Use of it as colorant is not evidence of symbolism.

#### Neandertal, where art thou?

- ▶ We imprison Ns in our own way of being in the world.
- ▶ MHs produced art in industrial quantities in the UP.
- ▶ Ns were terrific craftsmen, but did not apply these skills to aesthetics.
- ▶ Ns were not sapiens.
- Châtelperron culture, i.e. Renne Cave excavated 60 years ago: last N chance; in it the Mousterian was replaced by slender blades; like discovering a transistor in a Roman villa. But no human bone or teeth at these sites.
- ▶ No signs of N soul.

#### Carleton Coon's 1939 Neandertal in a suit and hat:



#### Ns in suit and ties

▶ Given ambiguity of N "art", there is currently no robust scientific basis for the claim that Ns and Sapiens followed convergent evolutionary trajectories leading with surprising synchronicity to a similar emergence of symbolic thought. This is at odds with archeological facts.

▶ We know very little about Ns and we need to stop dressing them up as us.

► You would not mistake a N on a subway as one of us.

#### Mandrin Grotto and Lebanon have similar MH tools

▶ His new research of the MH tools from Lebanon and Mandrin Grotto.

► MHs at Mandrin at 54 Ka.

▶ N stone tools are 99.9% of known N archeology; they are the only things that speak to us of their cognition.

### 6. Understanding the Human Creature

- ► The N were never just another version of us, but we are a long way from understanding its own ways of being human.
- ▶ Starting from the postulate that 500 K years of divergence between Ns and us had no impact on neuronal structures of our two populations or that they have evolved independently towards the same purpose (another us) is, strictly speaking, a new form of creationism.
- ► There is no archeological evidence that an encounter between these two populations ever took place. Only genetic evidence.
- ► The scale of our ignorance concerning one of the most significant events in the history of humanity is staggering.

## Fire memory

Use of cave wall soot to date presence of a group in Mandrin Cave: Revealed successive generations of hominins there. And they were able to distinguish MH vs N fires in the cave. Fire memory of 8 millennia.

Soot analysis revealed that at one point MH and Ns inhabited the cave only one year apart.

▶ Best evidence that they were contemporaneous in a specific area.

▶ Ns had occupied Mandrin for 80,000 years. Ns occupied it over 100 times.

### Slimak's 3 Wave Theory

➤ Slimak believes that the reason Ns disappeared was fundamentally connected to the arrival of H. sapiens. Not a slowly progressive replacement, but a conquest. Slimak posits a causal connection between appearance of sapiens and extinction of Ns; not just the principal factor in the disappearance of Ns, but the direct and unique cause of the N extinction. Just like colonization of Native Americans.

▶ He believes there were 3 waves of MH entry into Europe. The first two waves (incl. Mandrin at 54 Ka) failed. The third was a culturally homogenous population wave which rapidly occupied the whole of Europe. Ns did not just merge genetically with MHs. Like what happened to Native Americans by European colonizers.

## Genetic merging?

► Slimak thinks 2% N DNA in current humans occurred via contact between Ns and sapiens at 100 Ka in Asia.

► <u>First sapiens in Europe systematically have N DNA</u>. Sapiens in Romania, Bulgaria, Czech, & Siberia.

▶ But paleogenetics has not established N and MH genetic mixing during colonization period. No creole N population created by hybridization between Ns and MHs at moment of N extinction. Genetic exchange in only one direction: from N to sapiens. There is no sapiens DNA into the aboriginal N population.

### "I take your sister, but I don't give you mine."

- ► Slimak quotes Claude Levi-Straus repeatedly on elementary structures of kinship. Every human society is based on exchange of women. Women enter other families of related men. Ns practiced patrilocality. Usually based on reciprocity. "I give you my sister, you give me yours" = creates alliances.
- ► The absence of genetic signs of sapiens interbreeding in last Ns indicates nature of their relationship. A non-reciprocity: "I take your sister, but I don't live you mine."
- Ns are extinct. There was a radical replacement of populations. When sapiens appears, Ns disappear.
- ▶ But no archeological record of conflict between the two.

#### Call to arms.

- ▶ Only 40 N skeletons covering 400 millennia in 150 years of research.
- ► <u>Absence of evidence is not evidence of absence</u>; We have an archeological void, a lack of visibility. <u>Not proof that there was no conflict</u>.
- Sapiens technology made resource acquisition better, as well as expansion.
- ▶ Slimak thinks weapons were fundamental to understanding N demise. Thinks evidence of N projectile weapons is very rare. Radiometry technology based on production of massive lances or javelins that required close contact with the game. Only close quarter hunting with pole weapons. The rarity of arms in N archive is striking.
- ▶ The <u>UP</u> is about mechanical propulsion which involved standardization.

### Weapons

- ► Mechanical propulsion, esp. archery, allows technological progress toward microlithization, serial production, and standardization.
- ► This was a fundamental difference between sapiens and Ns.
- ➤ Slimak believes sapiens used archery based on nanopoints findings at Mandrin at 54 Ka. Weapons technology was critical in N replacement.
- ► (CJV: Note that prior evidence of archery in Europe was from 10 to 12 Ka.)
- ► There was both a numerical and technological difference between Ns and sapiens, reminiscent of Native American colonization.

#### Fundamental Structures of the Two Humanities

- ► Their biological diversity
- ▶ No N population ever produced weapons in a systematic way, that they were never interested in normalized, standardized form of production that would have enabled them to acquire technical systems specifically geared to acquisition of their prey. Slimak: Ns had structures of understanding the world that were unique and distinct from those of sapiens.
- ▶ They are another humanity. A different way of being in the world.
- ► Modern sapiens society is very normative; little tolerance for major differences.

### N creativity

- ▶ Unlike Ns who never made same tool twice. A <u>culture without</u> <u>normalization</u>, <u>without standardization</u>. Ns had infinite creativity in their tool making. Each tool is defacto a unique object. The artisanal production of objects by Ns reveals a perception of reality that has no structural echo in sapiens society.
- Scientific quantitative analysis of these fossil societies cannot understand their ethological differences from MHs.
- ► Slimak believes N creativity is beyond us; their art is the dialectic with their stone materials. Absolute artisanal freedom.
- ► For sapiens, art is an expression of ego. N creativity is not about ego. Art is not separable from everyday artisanal productions.

#### Ns were not us

- ▶ Ns are not only different, but in many mental aspects they overshadow sapiens – in their total, ongoing creativity, essentially free from the ego which structures so much of the differentiation of group and individual in sapiens populations.
- Ns left millions of remarkable artisanal objects which allow us to come fact to face with the fundamental structures of thought and the true nature of this extinct humanity.
- ▶ Racism is the refusal of difference. By making Ns into another us we reveal this unconscious racism that still fundamentally structures the schemas of our own society, incapable as we are of imaging any form of alterity.

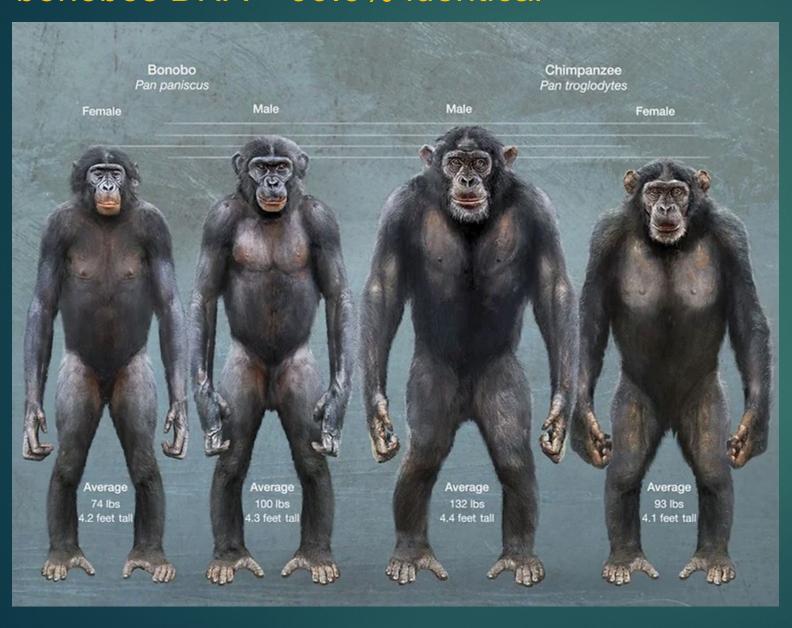
Human Evolution Scientific Update

November 2023 Charles J Vella, PhD

## The world's 1st CRISPR therapy has just been approved.

- Exa-cel, (brand name Casgevy), received its first regulatory approval on Nov. 16, 2023 from the U.K. Medicines and Healthcare products Regulatory Agency (MHRA) to treat two debilitating blood disorders:
  - ▶ sickle cell disease and
  - ▶ transfusion-dependent beta-thalassemia.
- Advisors to the U.S. Food and Drug Administration (FDA) determined that the drug was safe for clinical use in late October, and the FDA itself is expected to rule on whether to approve the treatment by December.
- Questions remain surrounding the <u>treatment's affordability and its long-term</u> <u>safety.</u>

## Humans & bonobos/chimpanzees DNA= 98.7 identical; chimps and bonobos DNA = 99.6% identical



## Bonobos and chimpanzees

 Bonobos and chimpanzees diverged from a common ancestor between one million and two million years ago, after the formation of the Congo River separated 1 population of apes into 2

 Bonobo is less aggressive than the chimp, with a smaller skull and shorter canine teeth. And it spends more time playing and having sex.
Females dominate. Cooperative. Do not participate in lethal inter-group aggression or kill within their own group.

## The self-domestication hypothesis: evolution of bonobo psychology is due to selection against aggression

- ► Experiments indicate that selection against aggression in mammals can have multiple effects on their morphology, physiology, behavior and psychology, and that these results resemble a syndrome of changes observed in domestic animals.
- ▶ We hypothesize that selection against aggression in some wild species can operate in a similar way. Here we consider the bonobo, *Pan paniscus*, as a candidate for having experienced this 'self-domestication' process.
- ▶ We first detail the changes typically seen in domesticated species including shifts in development. We then show that bonobos show less severe forms of aggression than chimpanzees, *Pan troglodytes*, and suggest that this difference evolved because of relaxed feeding competition.
- ► We next review evidence that phenotypic differences in morphology and behavior between bonobos and chimpanzees are analogous to differences between domesticates and their wild ancestors.

#### Self domestication

- ► Again, bonobo traits echo those of domesticates, including juvenilized patterns of development.
- ▶ We conclude that the self-domestication hypothesis provides a plausible account of the origin of numerous differences between bonobos and chimpanzees, and note that many of these appear to have arisen as incidental by-products rather than adaptations. These results raise the possibility that self-domestication has been a widespread process in mammalian evolution.
- ► <u>Similar to dogs from wolves, Russian silver fox experiments</u>: Domesticated animals tend to be smaller and less aggressive than their wild counterparts, they may also have floppy ears, variations to coat color, a smaller brain, and a shorter muzzle

# No scientific evidence that Homo naledi was advanced, new study argues

- ► A new study in the peer reviewed *Journal of Human Evolution* has cast doubt on claims that Homo naledi, a small-brained hominin dating to between 335,000 and 241,000 years ago, deliberately buried their dead and produced rock art in Rising Star Cave, South Africa.
- ▶ 3 eLife preprints claimed the features represented the earliest evidence of deliberate burial by a hominin species, and that Homo naledi lit up dark passageways using fire and intentionally carried the bodies of at least three individuals deep inside the Rising Star Cave system, dug pits, deposited corpses inside the pits, and covered the bodies with sediments.
- ► However, a group of experts with specializations in biological anthropology, archaeology, geochronology, and rock art, have now called for a deeper dig into the science behind the findings in a first, peer-reviewed critique titled "No scientific evidence that Homo naledi buried their dead and produced rock art" published in the Journal of Human Evolution.

### No evidence yet

- ► The evidence presented so far was not compelling enough to support the deliberate burial of the dead by Homo naledi, nor that they made the purported engravings.
- ► "We really need substantial additional documentation and scientific analyses before we can rule out that natural agents and post-depositional processes were responsible for the accumulation of bodies/<u>body parts</u> and to prove the intentional excavation and filling of pits by Homo naledi," Professor Martinón-Torres said.
- ▶ Moreover, Professor Petraglia added, "Unfortunately, there is a distinct possibility that the so-called stone artifact next to the hominin hand is a geofact (that is, a natural rock feature (as opposed to an artifact, which is a stone carved by an intelligent agent)), and not a product of stone tool flaking by Homo naledi."

### No evidence yet

- ▶ Professor Herries said, "There is no evidence that Homo naledi lit fires in the cave, purported burning locations could just be from manganese staining and charcoal within the cave [has yet] to be dated. Charcoal from natural fires is not uncommon in caves."
- ▶ "Detailed analyses are also needed to demonstrate that the so-called 'engravings' are indeed human-made marks, as marks like these can be produced as a product of natural weathering or animal claws," said Dr. Garate.
- ► The JHE commentary also offers a <u>brief insight on the state of the field</u> regarding the importance of responsible social communication and the challenges brought by new models of scientific publication.

### No evidence of fire use or engravings

- ► More importantly, no scientific evidence (e.g., Fourier-transform infrared spectroscopy, micromorphology, archaeomagnetism) has been presented to indicate the occurrence of in situ burnt material, let alone hearths.
- ▶ Previously acquired radiocarbon dates obtained by the site investigators on one of the apparent hearths resulted in very young dates (Lee Berger, unpublished data), questioning its association to *H. naledi*. Moreover, the occurrence of charcoal is also common in caves, including in South African landscapes, where there are frequent wildfires, so finding burned material in a cave setting does not automatically indicate anthropogenic activity.
- Numerous examples of shallow cross hatched and patterned natural erosional lines can be found throughout the Malmani dolomite, the geological formation that hosts Rising Star Cave and all the other Plio-Pleistocene hominin fossils in the region.

# No scientific evidence that *Homo naledi* buried their dead and produced rock art. María Martinón-Torres, et al., JHE, 2023

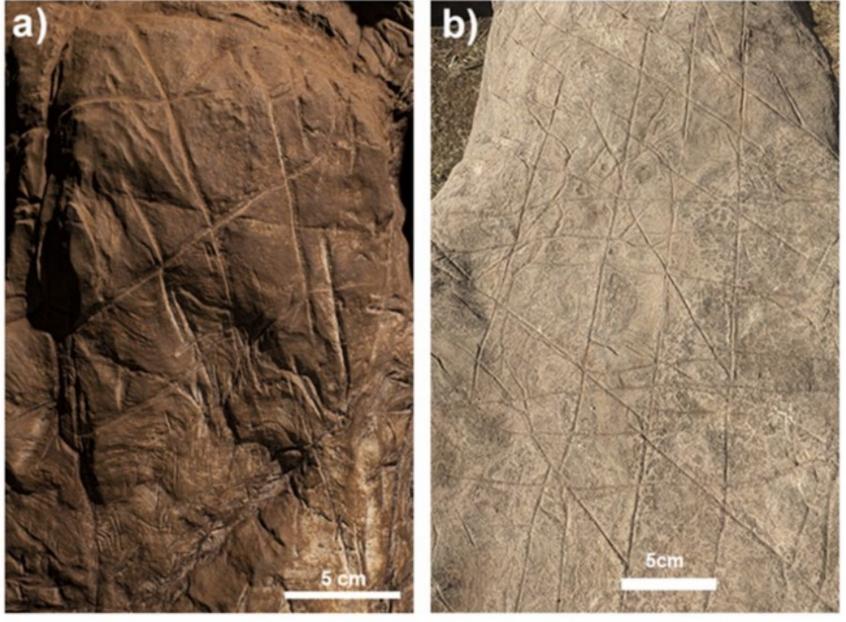
- ▶ Geological, taphonomic and paleontological evidence to suggest that natural formation scenarios may account for skeletal accumulations, such as a natural death trap, water transport of bodies/body parts and carnivore activity (e.g., Val, 2016; Stiner, 2017; Egeland et al., 2018; Pettitt, 2022).
- ▶ Preprints were accompanied by a strong media campaign. The media hype that accompanied both the unreviewed and reviewed, though currently unmodified, pre-prints at the time of this writing, triggered strong public controversy and an immediate debate about 'modern human behavior' but also about the way in which scientific work is communicated and perceived by the public.
- ► The peer reviews were unanimous in considering the evidence inadequate in its present form. Despite this, these versions remain available and communicated to the press and social media without yet integrating any of the referee's comments.

#### No evidence

- ► Here we argue that the evidence presented so far is not compelling enough to support the deliberate burial of the dead by *H. naledi* nor that they made the purported engravings.
- ► Substantial additional documentation and scientific analyses are needed before we can rule out that natural agents and post-depositional processes are responsible for the accumulation of bodies/body parts and to prove the intentional excavation and filling of pits by *H. naledi*.
- Moreover, detailed analyses are needed to demonstrate that the <u>so-called</u> 'engravings' are indeed human-made marks and that, like the purported evidence of fire use, they can be securely linked to *H. naledi*.
- Our commentary also offers a brief insight on the state of the field regarding the importance of responsible social communication and the challenges brought by new models of scientific publication.

#### Flaws

- ► Highlighting the various flaws in Berger's conclusions, the authors explain, "In a deliberate interment, the body is generally articulated... [yet from] the evidence available at Rising Star Cave, we infer that the hominin bones are not articulated but scattered."
- "[T]he <u>scattering of the elements implies they were not</u> <u>covered/protected after placement</u>," continue the researchers.
- ▶ Berger and his colleagues have not made use of any techniques to date these markings. Despite this, they assert in the title of their paper that the engravings were made between 335,000 and 241,000 years ago by *Homo naledi*.



The purported rock engravings in the Rising Star Cave, South Africa (a) and (b) markings produced by natural weathering in a similar cave system. (Martinón-Torres et al., *Journal of Human Evolution*, 2023)

### No convincing scientific evidence

▶ While Berger et al. (2023b) provide a list in their Table 1 of "known humans who have entered the Dinaledi system (in approximate order of entry)", this is not consistent with statements made previously with regard to the cave system and its use by cavers.

#### **▶** Concluding remarks

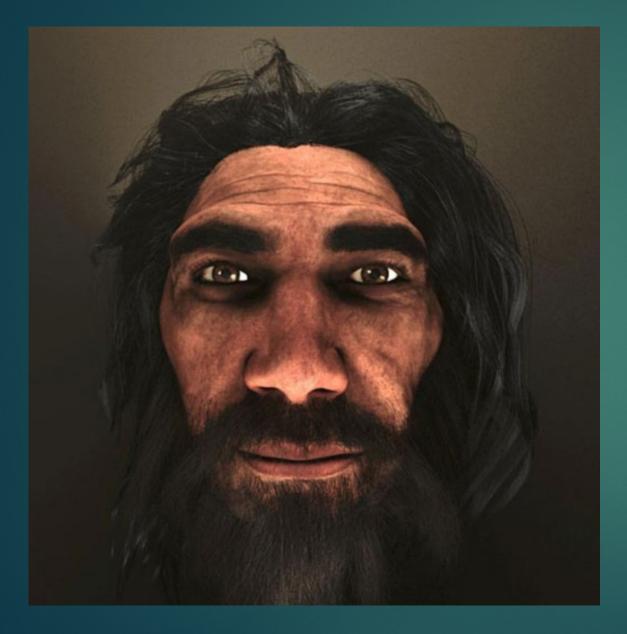
▶ "There is no convincing scientific evidence to indicate that *H. naledi* buried their dead and produced rock art in the Rising Star Cave system based on the information thus far presented. As explained here, the investigators have not employed the wide range of scientific methods (e.g., chronology, taphonomy, sedimentology, micromorphology, geochemistry) designed to answer the questions posed nor applied the basic principles of archeothanatology to identify a deliberate burial."

### Lee Berger's response

▶ "The arguments put forward are predominantly what has already appeared in the eLife reviews and these same authors' public comments, and have already been taken into account in the revised manuscript being resubmitted to eLife," Berger told Live Science in an email.

- ► Another external study by a different team addressing the claim of deliberate *H. naledi* burial is currently undergoing peer review.
- ► Live Science approached Netflix for comment on the accuracy of "Unknown: Cave of Bones" but did not receive one by the time of publication.

# New Greek reconstruction of *H. heidelbergensis*





# Petralona skull: 200-400 Ka: has been identified as *H. erectus*, *H. heidelbergensis*, early *H. sapiens*; often compared to Broken Hill skull

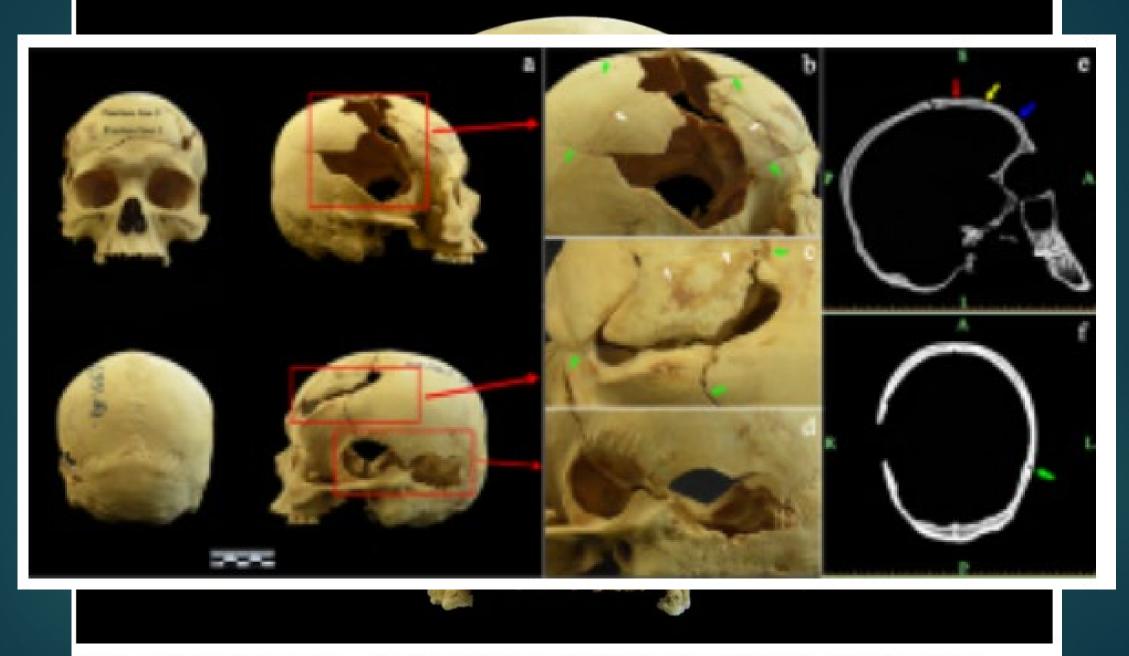




Used Mauer Jaw, 600 Ka

A shaman's surgical art? A neurosurgical and osteoarchaeological study of a therapeutic trepanation from the Yanghai cemetery in Turpan Basin,

- Chiskull found in China shows signs of healing after part of it was removed 2700 years ago, suggesting that a man survived at least eight weeks after surgery to relieve pressure in his head
- ► The Yanghai cemetery in Xinjiang, China, is a large, ancient burial ground containing the graves of a clan that practiced shamanism, generally defined as a belief system using trance to communicate with the supernatural. Qian Wang at Texas A&M University and his colleagues used CT scans to analyze a skull found in the cemetery from a man aged between 30 and 35 years who lived sometime around 750 to 800 BC.
- ► The man had experienced a blunt force injury causing an epidural hematoma, when blood accumulates under the skull. This puts pressure on the <u>brain</u> and can be life-threatening.



This ancient skull shows signs of healing within the walls of fracture lines inflicted by a blunt force injury

### Trepanation

- Signs of healing were seen within the walls of the fracture lines, indicating that the man made a full recovery from the procedure and went on to live for at least eight weeks.
- ▶ In order to successfully remove the hematoma, the <u>doctor was able to design</u> <u>a bone flap</u> according to the injury by using the fracture lines," says Wang. He and his colleagues <u>believe</u> this is the most advanced and skilled craniotomy ever found in the Xinjiang region and in the wider Eurasian steppe.
- ▶ Wang thinks this and similar discoveries make it highly likely that meticulous head surgeries were commonly performed by shamans in what is now China during the Bronze and Iron ages. Advanced procedures like this may have separated medicine from witchcraft and pioneered a more methodical and scientific approach towards healthcare

### Surgery

Cannabis has been found in the cemetery, which may have been used as an anesthetic. Shamans of the Subeixi culture, from another part of China at the same time, used horsehair to suture wounds as early as 1000 BC.

► Trepanation, the removal of part of the skull as a medical procedure, has been performed for thousands of years in many parts of the world, but it is less commonly found in archaeological excavations in Asia than in Europe

# Beads found in Israel are the oldest known example of the use of organic red pigments



Experimental replica of shell beads with Natufian red organic colorant made of roots of Rubiaceae plant. Credit: Laurent Davin, CC-BY 4.0

# Plant-based red coloration of shell beads 15,000 years ago in Kebara Cave, Mount Carmel (Israel)

- Found the oldest-known example of the use of organic red pigments to color an object—in this case, beads.
- Researcher noticed that an array of shell beads in a museum in Israel appeared to have an unusual red tint.
- Prior research has shown that humans have used rocks and minerals to color objects red going back at least 500,000 years.
- But use of plant-based materials to redden objects came much later. Until this latest find, the earliest known example was from 6,000 years ago.

## Color from root of Madder plant

- ► The team <u>first ruled out an ochre source for the coloring</u> and found that it contained a lot of carbon, a sign that it was likely from an organic source. Raman <u>spectroscopy revealed signatures common to all the beads, which came from the same source—they also matched closely with compounds found in roots of plants of the Rubiaceae (madder) family.</u>
- ► They also dated the beads to approximately 15,000 years ago, making them the oldest-known example of organic red pigment use.
- ► The research team suspects the people coloring the beads had crushed the roots of a local madder plant and then boiled the results—after fermenting, the liquid could have been used to dye the <u>beads</u>.

### BACKWARD project -- Paleoproteomics

- ▶ No hominid aDNA older than ~0.4 million years has been retrieved yet.
- ► BACKWARD will use paleoproteomics to address:
  - ▶ (i) the phylogenetic relationships among South African early hominins, and
  - ▶ (ii) the hominid paleobiodiversity in Southeast Asia;
- ▶ BACKWARD will also screen large sets of morphologically non-informative isolated fossil fragments of bones and teeth, to identify the species and sex of the organism from which they originated. Proteomes that are around 2 million years old can be retrieved from samples preserved in tropical conditions.
- ▶ Recent study: mass spectrometric (MS) sequencing of enamel proteomes from four ca. 2 million year (Ma) old dental specimens attributed morphologically to *P. robustus*, from the site of Swartkrans; the oldest genetic data ever collected from a hominin. Enabled determination the biological sex of all the specimens.

# Demographic and hormonal evidence for menopause in wild chimpanzees - Brian M. Wood

- ► Humans and some whales are the only known species in which females live long after they stop being able to reproduce; both develop menopause (end of your menstrual cycles).
- ► Chimpanzees should now be added to the list, and they offer clues about the evolutionary imperatives behind menopause in women.
- ► The vast majority of mammal females produce offspring until the end of their lives, but <u>humans experience a decline in reproductive hormones</u> and the permanent cessation of ovary function around age 50.
- ► <u>Females of five species of toothed whale</u>, including orcas and narwhals, similarly survive well beyond fertile age.
- ► It isn't obvious why natural selection would favor this trait, and only among a handful of species.

# Grandma chimps offer clues for evolution of menopause in humans

- ➤ Some scientists have put forward the "grandmother hypothesis" as a possible explanation: the idea that older females enter a post-reproductive state to consume fewer resources and focus on improving their grandchildren's odds of survival.
- New study: examined the mortality and fertility rates of 185 female chimpanzees in the Ngogo community of wild chimpanzees in Kibale National Park, Uganda, between 1995 and 2016.
- Specifically, the team calculated <u>a metric called the post-reproductive</u> representation (PrR), which is the average proportion of the adult life span that is spent in a post-reproductive state.
- ► Showed Ngogo chimpanzee females—but not other chimpanzees from other populations—lived on average 20 percent of their adult years in a post-reproductive state, just a little under what has been observed in humans.

### But Chimps aren't good grandmas

- ► Hormonal patterns closely mirrored what was seen in human females experiencing menopausal transition.
- In chimpanzee society, daughters leave the community in which they are born, while the males who remain mate promiscuously.
- ► That means <u>males don't know who their offspring are</u>, and by extension, grandmothers don't know which grandoffspring are theirs—so the "grandmother <u>hypothesis</u>" won't apply.
- ► Instead, Wood said that menopause might have evolved to reduce competition for limited breeding opportunities between aging females and their daughters.
- ▶ When a female chimp first enters a new group, she starts out with a low level of relatedness to other members, though this increases over time as she breeds.
- ► Since her genes are by then widespread, she has less to gain in breeding conflict against a younger female.

### Chimps

- ► Found clear evidence for menopause in females living past the age of 50. Unlike the case for humans and toothed whales, however, postreproductive chimps in this population are not involved in the raising of related offspring, suggesting that a different process is driving its development.
- One explanation for this discrepancy is that substantial PrR could be a temporary response to unusually favorable ecological conditions at Ngogo, including low levels of predation, high food availability, and successful between-group competition.
- A second possibility is that substantial PrR is an evolved, species-typical trait in chimpanzees, which has not been observed elsewhere owing to recent negative human impacts, especially disease epidemics.

### Reproductive conflict hypotheses

- Grandmother hypothesis is unlikely to apply to chimpanzees, whose aged females generally live apart from their daughters, as daughters leave their natal groups at adulthood.
- ▶ In the context of female-biased dispersal, a more relevant theory may be the reproductive conflict hypothesis, which highlights the fact that after migrating into a new group, females become increasingly related to other group members as they age and face competition with younger females for limited breeding opportunities. The oldest females might stop reproducing in order to limit the inclusive fitness costs of that competition.
- ► The grandmother and reproductive conflict hypotheses are not mutually exclusive alternatives, and both may be required to explain why all human societies have higher PrR than documented here for chimpanzees.

### Humans caught more diseases after we domesticated animals

- ➤ Analysis of DNA from human remains up to 37,000 years old shows that more infectious diseases jumped from animals to people after the dawn of farming.
- ► This is the <u>first direct evidence that the domestication of animals led to humans acquiring more infectious diseases</u>
- ▶ The study looked at a wide range of microbes in human remains from all over the world and many different times, making it the largest and most comprehensive of its kind so far. The oldest human remains were from around 37,000 years ago, but most were between several thousand and a few hundred years old.
- ▶ Most of the microbial DNA the researchers found was from soil bacteria, suggesting bacteria got into the bones after burial. In teeth, much of the microbial DNA came from bacteria known to live in people's mouths.

#### Bacterial diseases

- The team was also able to identify many disease-causing bacteria and viruses that were in the blood of people before they died and may have caused or contributed to their death.
- ► The most common was the bacterium that causes the plague, *Yersinia pestis*, found in 39 people, which is 3 per cent of the remains. This bacterium mainly infects rodents, but can be spread to people by fleas.
- ▶ The oldest plague cases were in three people who lived in various parts of Asia around 5700 years ago. The bacterium was also found in a person entombed on Orkney in Scotland around 4800 years ago about 800 years before the previous earliest known case of the plague in Britain.

#### Bacterial diseases

- ➤ Overall, the team found lots of plague cases between 6000 and 3000 years ago. Then there was a gap until 2000 years ago, when there was a wave lasting for a few centuries, then another gap until a second wave corresponding with the medieval "Black Death" plague pandemic.
- ► The early form of plague wasn't very transmissible and died out, to be replaced later by more transmissible strains that caused pandemics.
- ► The next most common microbe was *Borrelia recurrentis*, which causes a disease spread by body lice called louse-borne relapsing fever. This disease is now rare, but the team found it in 31 people, 2.3 per cent of the total, suggesting it was widespread in the past.
- ► Other diseases identified include <u>malaria</u>, hepatitis B, leprosy and leptospirosis, also known as Weil's disease.

#### Zoonotic diseases = diseases from animals

- ► The researchers divided the kinds of microbes they found into five broad types, including zoonotic diseases, those that jumped from animals into humans. They found there was an increase in zoonotic diseases from around 6000 years ago, but not of any of the other four types.
- ► The risk and extent of zoonotic pathogen transmission likely increased with the adoption of more widespread husbandry practices and pastoralism

► Limitation: Another issue with the study is that standard DNA sequencing misses RNA viruses, such as flu and coronaviruses, which may have caused major outbreaks in the past.

# The landscape of ancient human pathogens in Eurasia from the Stone Age to historical times

- ▶ Abstract: Infectious diseases have had devastating impacts on human populations throughout history. Still, the origins and past dynamics of human pathogens remain poorly understood¹. To create the first spatiotemporal map of diverse ancient human microorganisms and parasites, we screened shotgun sequencing data from 1,313 ancient human remains covering 35,000 years of Eurasian history for ancient DNA deriving from bacteria, viruses, and parasites.
- ▶ Demonstrate the widespread presence of ancient microbial DNA in human remains, identifying over 2,400 individual species hits in 896 samples. We report a wide range of pathogens detected for the first time in ancient human remains, including the food-borne pathogens *Yersinia enterocolitica* and *Shigella* spp., the animal-borne *Leptospira interrogans*, and the malaria-causing parasite *Plasmodium vivax*.

### Ancient pathogens

- ▶ Our findings extend the spatiotemporal range of previously described ancient pathogens such as Yersinia pestis, the causative agent of plague, Hepatitis B virus, and Borrelia recurrentis, the cause of louse-borne relapsing fever (LBRF).
- ► For LRBF we increase the known distribution from a single medieval genome to 31 cases across Eurasia covering 5,000 years.
- ➤ Grouping the ancient microbial species according to their type of transmission (zoonotic, anthroponotic, sapronotic, opportunistic, and other), we find that most categories are identified throughout the entire sample period, while zoonotic pathogens, which are transmitted from living animals to humans or which have made a host jump into humans from animals in the timeframe of this study, are only detected from ~6,500 years ago.

### Zoonotic pathogens

- ▶ The incidence of zoonotic pathogens increased in our samples some 1,000 years later before reaching the highest detection rates ~5,000 years ago, and was associated with a human genetic ancestry component characteristic of pastoralist populations from the Eurasian Steppe.
- ► Results provide the first direct evidence for an epidemiological transition to an increased burden of zoonotic infectious diseases following the domestication of animals. However, they also reveal that the spread of these pathogens first becomes frequent thousands of years after increased animal-human contact, likely coinciding with the pastoralist migrations from the Eurasian Steppe. This study provides the first spatiotemporal map of past human pathogens using genomic paleoepidemiology, and the first direct evidence for an epidemiological transition of increased zoonotic infectious disease burden after the onset of agriculture, through historical times.

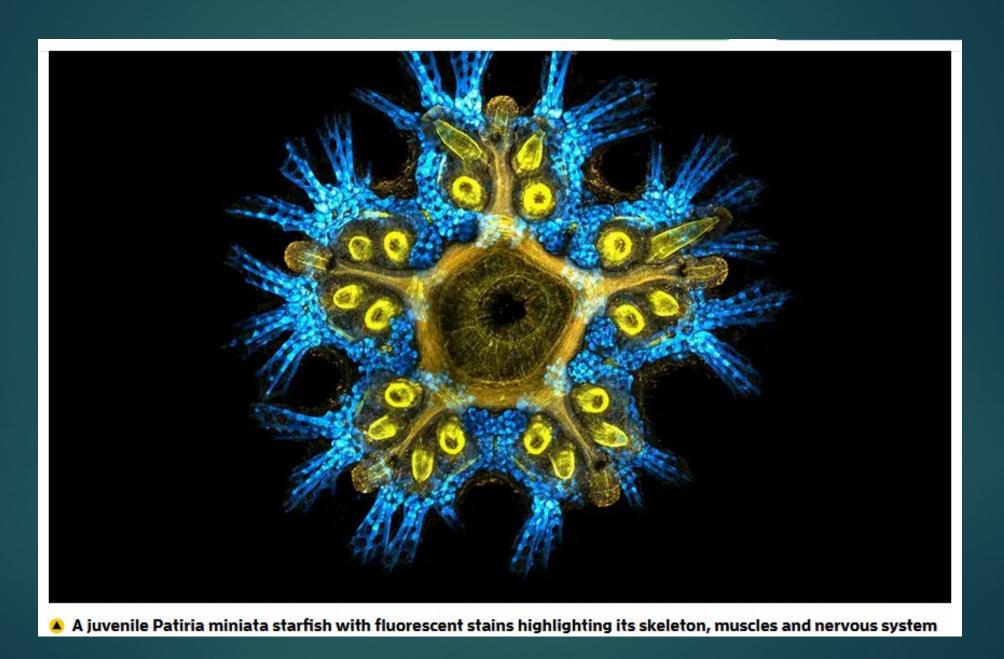


The Tasmanian tiger (one shown in captivity in 1930) went extinct after the last one died in a zoo in 1936. Researchers have now extracted RNA from one museum specimen.

### First decoding of the RNA of an extinct animal

- ► For the first time, researchers have successfully extracted and decoded RNA from an extinct animal. RNA is much less stable than DNA.
- ► The thylacine, also known as the Tasmanian tiger, was a wolflike marsupial that went extinct after the last one died in a zoo in Hobart, Tasmania in 1936.
- Now a roughly 130-year-old museum specimen has yielded bits of RNA, the fragile molecules responsible for turning DNA's genetic instructions into cellular functions. Only RNA can reveal how an organism's cells actually functioned. In recent years, researchers have mapped out the thylacine DNA genome
- ▶ The results shed new light on thylacine biology and may inform efforts to bring the marsupial back from extinction. In the not-too-distant-future, Pask and other researchers are hoping to bring thylacine back to Tasmania. Their plan to deextinct the animal involves modifying the genes of one of the thylacine's closest living relatives, another marsupial called a fat-tailed dunnart (*Sminthopsis crassicaudata*).

#### A stained starfish



# Echinoderms/Sea stars don't have a body - they're just a big squished head

- Scientists trying to work out where a starfish's head is have come to a startling conclusion: it is effectively the whole animal.
- Echinoderms have five lines of symmetry radiating from a central point and no physically obvious head or tail. Yet they are closely related to animals like us and evolved from a bilaterian ancestor.
- ► Formery and his colleagues decided to look at a set of genes known to direct the head-to-tail organization of all bilaterians. In these animals, these genes are turned on, or expressed, in stripes in the outer layer of the developing embryo. The genes that are expressed in each stripe define which point on the head-to-tail axis it will become.
- Strikingly, there is no ectodermal territory in the sea star that expresses the characteristic bilaterian trunk genetic patterning program. This finding suggests that from the perspective of ectoderm patterning, echinoderms are mostly head-like animals

### Head walking on its lips

- ► Echinoderms represent a really extreme experiment in how to use that bilateral network to produce a very, very different body plan
- ▶ Genes that determine the head end in bilaterians were expressed in a line running down the middle of each arm on the underside of the starfish. The next head-most genes were expressed on either side of this line, and so on.
- ► The genes normally expressed in the trunk of bilaterians were missing in the outer layer of the animal. This suggests that <u>starfish have</u> jettisoned their trunk regions and freed up the outer layer to evolve in new directions
- ► The body of an echinoderm, at least in terms of the external body surface, is essentially a head walking about the seafloor on its lips

#### Echinoderms

► Animals like us may have kept their trunks to escape predation by swimming away.

► Echinoderms hunkered down and armored themselves, so they didn't need a trunk

### Elusive origins of Life

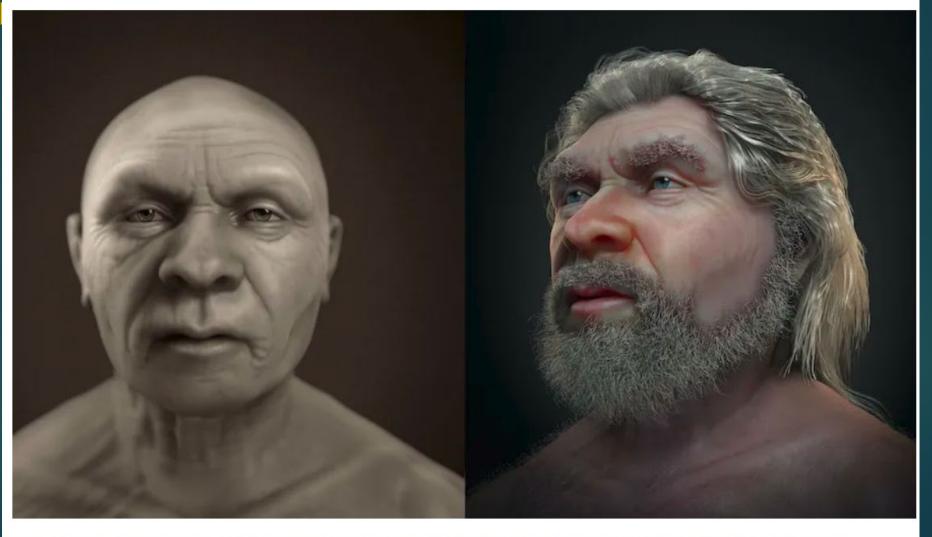
- ► How did life on earth begin?
- ▶ 1953: Structure of DNA; 3 weeks later Miller-Urey experiment chemical cocktail (methane, ammonia, hydrogen) + spark produced amino acids; Sep 1953 calculation of 4.5 B years as age of earth
- ▶ 2022: first 3-D model of bacterial cell; cells are incredibly complicated
- ► DNA is not the only requirement; cells are highly dynamic
- ▶ No current universal definition of life. 12 years ago = 123 definitions
- Current search for mandatory elements for living processes and which appeared first.
- ► LUCA = 3.9 B years?; but water on earth = 4.2 B

### Complex origins of life

- ▶ But meteorites contain amino acids
- ► First life on planet did not have DNA; some now argue RNA was the essential ingredient for duplication of life; can reliably duplicate; study = single RNA strand can have 5 genetic lines
- ► Others think <u>metabolism</u> is key
- ► Theory of autocatalysis: ability of a set of molecules to react together in order to replicate themselves; single celled microbes have 6683 different autocatalysis networks; probably needs metals to work = life depends on its environment
- ► Environments: not just water; also geochemical pools on land exposed to UV light; deep sea hypothermal vents with complex mixtures of chemicals
- ► Life is community. Not 1<sup>st</sup> organism, but 1<sup>st</sup> ecosystem; films rather than individual cells

A new facial reconstruction depicts a Neanderthal whose skeleton was found by priests in a French cave: La Chapelle-

aux-Sai

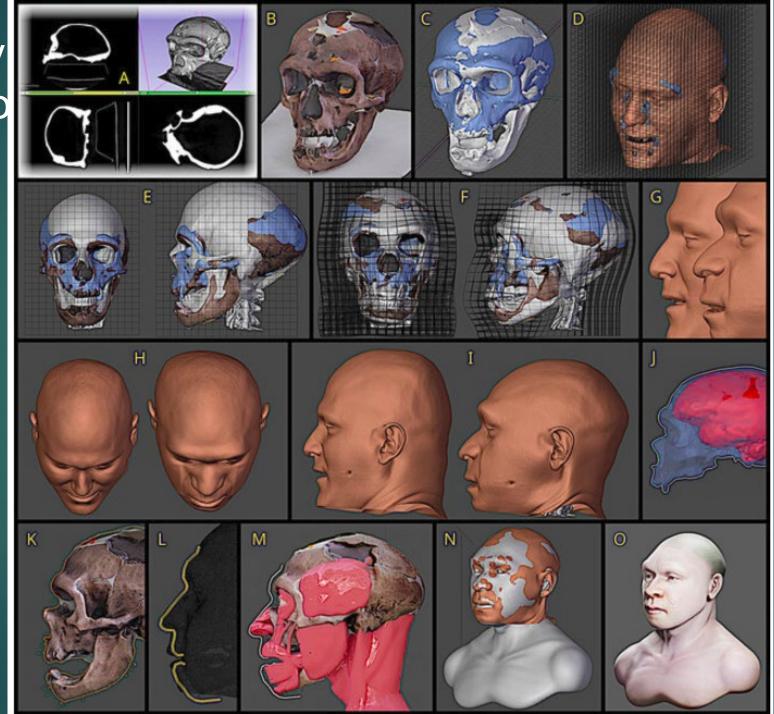


Side-by-side facial reconstructions of the "old man" Neanderthal. (Image credit: Cícero Moraes et al)

### La Chapelle-aux-Saints

- ▶ n 1908, a group of Catholic priests discovered what looked like the skeletal remains of a man buried inside a cave in La Chapelle-aux-Saints, a commune in south-central France. The nearly complete skeleton lacked several teeth, earning him the nickname the "old man."
- ▶ Now, 115 years later, forensic artists have created a digital facial approximation of the Neanderthal, who lived to be about 40 years old, offering a glimpse of what he may have looked like when he lived sometime between 47,000 and 56,000 years ago, according to a new facial approximation

# CT methodology for reconstruction



## Head Louse

## What creature?



## Genetic analysis shows head lice evolution mirrors human migration and colonization in the Americas

- ► The human louse is a wingless, blood-sucking parasite that lives its entire life on its host. It is one of the oldest known parasites to live on humans, and the two species have coevolved for millennia
- ► Head lice came to the Americas twice—once during the first wave of human migration across the Bering Strait, and again during European colonization.
- ► Human lice are more than annoying human parasites, they are 'satellites' of our evolution. Because <u>human lice feed on human blood</u>, they need us to survive, and over millions of years this resulted in a <u>long co-evolutionary history together</u>.

- ► Evidence of nuclear admixture was uncommon (12%) and was predominate in the New World potentially mirroring the history of colonization in the Americas.
- Head louse populations are strongly structured geographically. This current study highlights a genetic relationship between Asia (cla) and Central America (clb) subclusters supporting the idea that the Central America cluster (clb) is probably of Native American origin since a potential source population for the first people of the Americas has been suggested to be East Asia. Thus, microsatellite data suggest that current human louse populations in the Americas retain human louse genetic diversity from louse brought by the first people. This work adds to our previous studies showing that human lice could be used to track aspects of human evolution.

#### Lice: twice

➤ Our results showed the presence of two divergent nuclear louse genetic clusters, with very limited and recent admixture, mostly in the New World. We suggested that this pattern is reflective of human migration to the Americas, with an early wave of louse-human co-migration during the population of the New World followed by the most recent European migration.

We also hypothesized that there is a potential mechanism that is preventing admixture between these two divergent nuclear clusters, most likely epigenetic in origin

#### Lice and *H. erectus*

- Scientists already believed that early ancestors of our species, Homo sapiens, diverged from other archaic humans about 1.2 million years ago.
- ► The new study showed two almost identical-looking but genetically different strains of head lice diverged about the same time. That indicates each of the two kinds of head lice infested a different species of early human as the human species diverged.
- Genes from both types of head lice are found on people today, suggesting that after infesting Homo erectus for 1 million years, one type jumped from that soon-to-be-extinct species onto Homo sapiens

## Genome sequences of 36,000- to 37,000-year-old modern humans at Buran-Kaya III in Crimea: Abstract

- ▶ Populations genetically related to present-day Europeans first appeared in Europe at some point after 38,000–40,000 years ago, following a cold period of severe climatic disruption. These new migrants would eventually replace the pre-existing modern human ancestries in Europe, but initial interactions between these groups are unclear due to the lack of genomic evidence from the earliest periods of the migration.
- ► Here we describe the genomes of two 36,000–37,000-year-old individuals from Buran-Kaya III in Crimea as belonging to this newer migration. Both genomes share the highest similarity to Gravettian-associated individuals found several thousand years later in southwestern Europe.

## Buran-Kaya III in Crimea

► These genomes also revealed that the population turnover in Europe after 40,000 years ago was accompanied by admixture with pre-existing modern human populations.

► European ancestry before 40,000 years ago persisted not only at Buran-Kaya III but is also found in later Gravettian-associated populations of western Europe and Mesolithic Caucasus populations.

## Genomic affinities between Buran-Kaya III and other ancient individuals.



## Genome sequences of 36,000- to 37,000-year-old modern humans at Buran-Kaya III in Crimea

- ► How did our species, Homo sapiens, arrive in Western Europe?
- Published in *Nature Ecology & Evolution*, new study analyzes two skull fragments dating back between 37,000 and 36,000 years to conclude that our ancestors came from Eastern Europe and migrated westwards. These two individuals interbred with Neanderthals and with the very first European Homo sapiens, who arrived around 45,000 years ago and were thought to have become extinct following a major climatic catastrophe.
- ➤ Together with lithic tools and pierced mammoth ivory beads, small skull fragments of the two skulls found in 2009 at an archaeological site in the Crimea, Buran Kaya III, bear witness to the presence of anatomically modern humans in Eastern Europe.

E. Andrew Bennett, et al., 2023

#### Eastern MHs

- ► These individuals are the oldest representatives of Western Europeans to have established themselves permanently in Europe and to have left traces in the genomes of present-day Europeans.
- ▶ It is estimated they <u>settled</u> in the <u>region after the ice age that took place from 40,000 to 38,000 years ago</u>.
- In addition to extremely low temperatures, the <u>latter period was also marked by</u> the eruption of a super-volcano in the Phlegrean Fields region near Naples, which left south-eastern and eastern Europe covered in ash. Ecological disaster: some hypothesized resulting ecological crisis wiped out both the last Neanderthal and the first Homo Sapiens populations of the early Upper Paleolithic.

#### 2<sup>nd</sup> wave of MHs

- ▶ Present-day Europeans bear no trace of the genomes of these first sapiens Europeans, unlike the human populations that lived in Europe after the ecological crisis of 40,000 years ago, some of whose genomes have been sequenced.
- ► Analysis of these two skull fragments, which are thought to be 700 years apart from one another, revealed that these individuals were part of the second wave of European settlement by H. sapiens that occurred after the ecological crisis.
- ▶ Both individuals are descendants of distant interbreeding with Neanderthals. Our study also showed that the more recent individual bore traces of interbreeding with individuals from the first wave of settlement thought to have been exterminated by the -40,000 year ice age, represented by the Zlatý Kůň individual (-45,000 years). We were therefore able to conclude that the first H. sapiens were not completely replaced and some must have survived the ecological crisis.

#### Had descendants

- ► The genomes of individuals from Buran Kaya III also revealed a genetic link with contemporary and much later Caucasian populations, in line with similarities identified by archaeologists between lithic tools found in the southern Caucasus and those found at Buran Kaya III at the same period.
- ► This link indicates the direction of the migration of Buran Kaya III's ancestors in Europe: from the Middle East via the Caucasus to the territory of present-day Ukraine.
- ► The strongest genetic link has been identified between the genomes of individuals from Buran Kaya III and those from south-west France and northeast Spain (Serinyà, -27,000 years BC) and, to a lesser extent, those from Austria (Krems-Wachtberg, -30,500 years BC) and the Czech Republic (Dolní Věstonice, -31,000 years BC) who lived 5000 to 7000 years later.

#### **Ancestors of Gravettians**

► These individuals, close to those from Buran Kaya III, were part of the population associated with the Classical Gravettian period, which produced the female ivory statuettes known as the "Gravettian Venuses" found in France, Germany, Austria and the Czech Republic.

▶ This genetic link between individuals from Buran Kaya III and those hailing from the Gravettian culture suggests the former were ancestors of the latter and were already practicing a culture that can be described as proto-Gravettian. The genetic ties indicate that these populations spread from east to west.

#### Similar lithics

Moreover, the lithic tools produced by Crimean individuals have been attributed to the Gravettian complex, but this attribution has been rejected by other archaeologists, mainly because of their early date and their location to the east, far from the classic "Gravettian" culture that was produced in central and western Europe between -34,000 and -26,000 years ago, i.e., 5,000 to 7,000 years later and 3,000 km further east.

➤ Our genetic results prove the individuals from Buran Kaya III were the ancestors of the Western Europeans, producers of the Gravettian culture and artists of the famous Gravettian Venuses.

Giant 1.5-foot-long Vangunu giant rat (*Uromys vika*) that can crack open coconuts photographed for 1st time on remote Solomon island



### Deep-time paleogenomics and the limits of DNA survival

- ▶ Review: the state-of-the-art in paleogenomics and discuss key challenges, including technical limitations, evolutionary divergence and associated biases, and the need for more precise dating of remains and sediments. We conclude that with improvements in laboratory and computational methods, the emerging field of deep-time paleogenomics will expand the range of questions addressable using ancient DNA.
- ▶ Pleistocene =2.6 Ma to 10 Ka
- ▶ Cold climate favors fossil preservation
- Population turnover and interspecies gene flow: <u>demographic trends in large mammals closely track available habitat: food availability determines population size</u>

#### aDNA preservation

- aDNA from bones and teeth that are several hundreds of thousands of years old and beyond one million years old has now been recovered and analyzed
- But very rare because postmortem processes lead to successive degradation of DNA molecules into increasingly small fragments
- ▶ aDNA recovery is significantly more difficult with age; Most ancient DNA studies fall within the last 50 ka and the most recent glacial cycle.
- ▶ aDNA been recovered from remains and sediments in high-latitude permafrost and lower latitude caves, suggesting that deep-time genomics is feasible in ideal preservation environments.

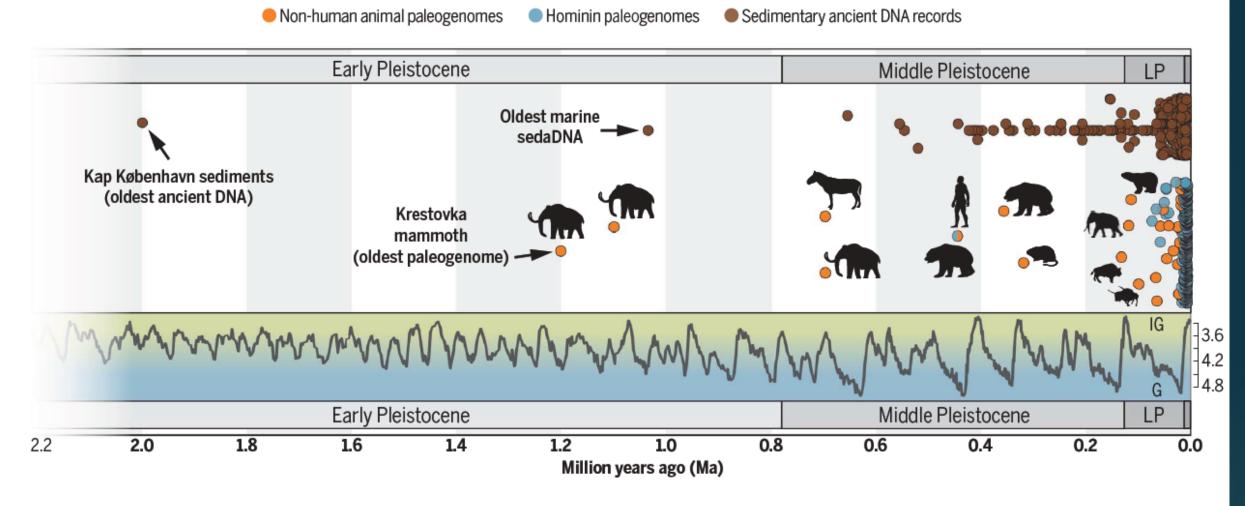


Fig. 1. The temporal distribution of ancient DNA studies to date highlights gaps and opportunities for deep-time paleogenomics and sedimentary ancient DNA. Most ancient DNA studies fall within the last 50 ka and the most recent glacial cycle. The climate curve is based on benthic  $\delta^{18}$ -oxygen measurements [per mil; LRO4 stack from (42)]. Sedimentary ancient DNA data are from the AncientMetagenomeDir (v23.06.0) (57) and (58), with meta-

barcoding records older than one million years excluded. Paleogenomic data are available from (59). Paleogenomes older than 100 ka are annotated with a silhouette of the study taxon, with the deep-time paleogenomes including a 130-ka steppe bison (36); 330-ka collared lemming (40); 360-ka cave bear (9); 430-ka cave bear and hominin (35, 60); 700-ka horse (8); and 700-ka, 1.1-Ma, and 1.2-Ma mammoths (10). LP, Late Pleistocene; IG, Interglacial; G, Glacial.

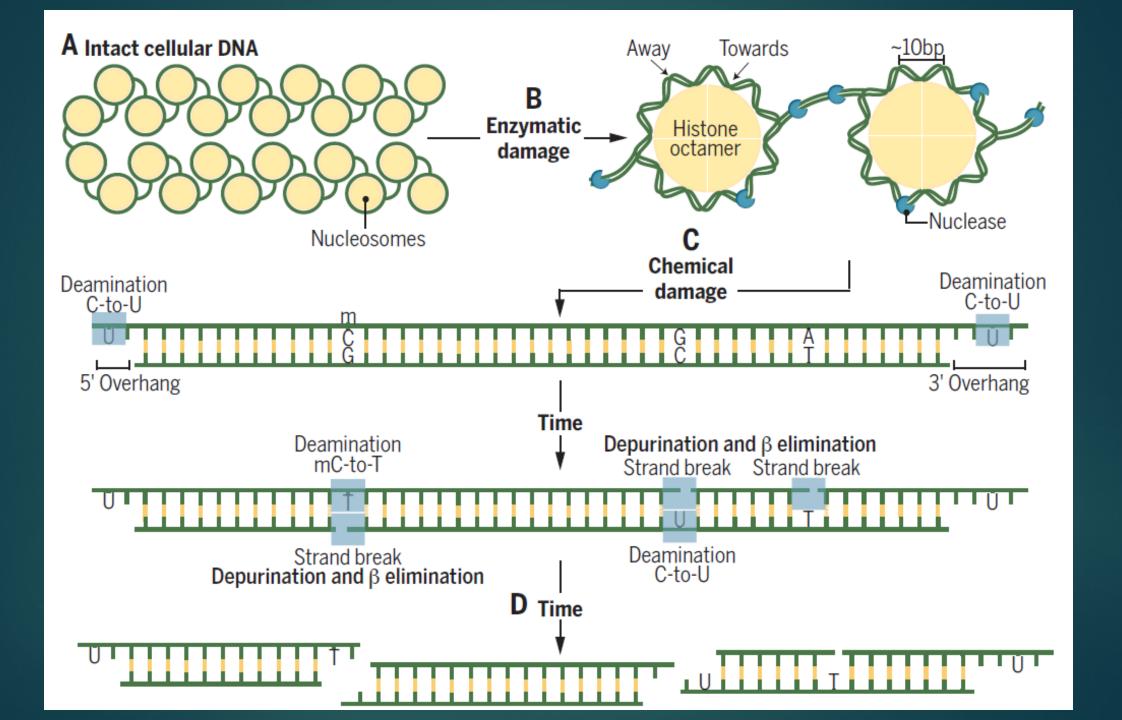
#### DNA recovery length?

▶ DNA is regularly recovered from remains and sediments that date to within the last 100 ka.

- ▶ As of September 2023, the oldest reconstructed paleogenome is from a permafrost-preserved mammoth dating to between 1 and 2 Ma and the oldest isolated DNA is from ~2-Ma sediment from northern Greenland. However, the maximum age of recoverable and useful DNA molecules remains uncertain.
- Deep-time DNA molecules are short—often <35 bases. DNA fragmentation and degradation begins after death and continues until fragments are too short to be useful.

#### Use of damage as correctives

- ▶ Depurination, deamination, DNA crosslinking and oxidative damage are the destructive processes in DNA degradation.
- ► These typical damage patterns can be used to:
  - bioinformatically corroborate the authenticity of recovered ancient sequences and,
  - ▶ to reduce their impact on sequence accuracy,
  - ► can be identified and removed from ancient DNA data sets using standard bioinformatic approaches.



## Degradation processes (prior slide)

- ► (A) The <u>integrity of DNA length is maintained by a cell's enzymatic</u> repair machinery and, in eukaryotic genomes, packaged in histone-DNA complexes.
- ▶ (B) Following death, repair stops and DNA damage begins to accumulate. Nucleases and microorganisms cleave DNA in regions between nucleosomes and when the DNA backbone faces away from histones.
- ▶ (C) Over time, chemical damage also accumulates. Cytosine bases are converted to uracil and methylated cytosines are converted to thymines (by deamination). Cytosines are particularly vulnerable to deamination in single-stranded regions such as in overhanging regions at DNA termini, but deamination is possible in some double-stranded contexts

#### Degradation

▶ Fragmentation occurs after the loss of purine bases (depurination), creating abasic sites that can be cleaved by b elimination. Depurination and b elimination create a region of single-stranded DNA, which leaves cytosines vulnerable to deamination.

▶ (D) Given enough time, DNA molecules will become too short to be identifiable.

▶ (E) Depurination leads to overrepresentation of adenine and guanine bases adjacent to strand breaks. C-to-T mismatches are elevated near read ends and observed throughout damaged reads. Whereas 3' G-to-A mismatches are observed in double-stranded libraries, single-stranded libraries show a C-to-T signal at both ends by retaining the native termini of the molecules.

#### Reference bias: no known species relative

▶ aDNA data sets comprise both endogenous DNA from the target organism(s) and introduced exogenous/foreign DNA.

► These categories of molecules can be separated by identifying each read through taxonomic assignment, which can be problematic if the ancient organism has no close living relative to act as a genomic reference.

▶ Lack of a close taxonomic reference, reference bias, and errors introduced by damage will also impede variant and consensus calling. Bioinformatic approaches mitigate these challenges by directly modeling DNA damage and/or bias as part of genotyping

### Speciation and hybridization

- Speciation is not always a simple process of cladogenesis (formation of a new group of organisms or higher taxon by evolutionary divergence from an ancestral form) followed by reproductive isolation. Instead, modern and paleogenomic data have shown that interspecific hybridization is surprisingly common and perhaps driven in part by repeated habitat redistribution associated with glacial cycles. For example, brown bears and polar bears hybridized during previous glacial and interglacial periods as well as in the modern era.
- ▶ Similarly, a mammoth paleogenome dating to the Early Pleistocene revealed that Columbian mammoths (Mammuthus columbi) originated after hybridization between two distinct ancient mammoth lineages. Taxonomically diverse deep-time paleogenomes could clarify the timing, rate, and extent of genomic introgression episodes and their role in evolution.

### Ghost lineages and molecular evolution

- Deep-time paleogenomes could also identify unknown "ghost" lineages that contributed to species' ancestries, as exemplified in the paleogenomic characterization of the Krestovka mammoth.
- As deep-time paleogenomes tend to occupy basal phylogenetic positions within their clades, they can also provide important calibrations for estimating rates of molecular evolution. For example, paleogenomic data from a Middle Pleistocene hominin from Sima de los Huesos in presentday Spain confirmed hypotheses from Late Pleistocene genomes that Neanderthals and Denisovans diverged during the early Middle Pleistocene, whereas the inclusion of a ~700-ka horse paleogenome in the equid phylogeny pushed the estimated time for the origin of living equids to more than twice that previously hypothesized

#### Who was ancestral?

- ► Paleogenomes from the Early and Middle Pleistocene can also be used to test hypotheses about relationships among species, including how derived forms are related to earlier forms.
- ► An outstanding question in paleontology is whether fossil morphospecies are true species, synchronous ecomorphs, or chronospecies that were direct ancestors of succeeding species.
- ▶ A paleogenomic study of ancient North American bison dating from ~130 to 110 ka, for example, showed that two samples exhibiting extreme size dimorphism and representing supposedly distinct species—the longhorn bison and the steppe bison—actually belong to the same lineage that dispersed into North America only a few tens of thousands of years earlier. Conversely, deep-time paleogenomics can also give context to species for which we have only limited remains, such as Denisovans

## The impact of glacial cycles on biodiversity

➤ Since the change in glacial periodicity (to ~100 ka cycles that occurred at 1.2 to 0.7 Ma), the dominant pattern has been cycles of long glaciations separated by short, warm interglacials. This pattern is believed to have driven the demography and range dynamics of many species.

► Long interglacials, for example, have been correlated with bottlenecks in cold-adapted taxa and expansion and speciation in warm adapted taxa. Of particular interest is the unusually long interglacial that occurred 420 to 370 ka (Marine Isotope Stage 11).

#### Entire deep-time ecological communities

- ► Ability to reconstruct entire deep-time ecological communities.
- ► To date, only five studies have attempted to use sedimentary ancient DNA to reconstruct plant and/or animal communities dating to the Middle Pleistocene or older:
  - Kjær reconstructed components of an Early Pleistocene interglacial ecosystem from sediment extracted from the present day polar desert in northern Greenland;
  - Armbrecht reconstructed an Early to Middle Pleistocene <u>marine ecosystem</u> from Iceberg Alley in the Southern Ocean;
  - Courtin reconstructed a Middle Pleistocene interglacial ecosystem Roma permafrost megaslump in Eastern Siberia; and
  - Willerslev reconstructed Middle Pleistocene <u>plant communities from sediments collected below the Greenland ice sheet and from coastal Siberian permafrost.</u>

#### Minerals and DNA

- ▶ Kjaer found that DNA adsorbed preferentially to clay mineral surfaces compared with nonclay surfaces, particularly to the clay mineral smectite, which can bind 200 times more DNA than quartz and is a common mineral in terrestrial samples. Their best performing extraction protocol recovered 40% of DNA bound to quartz and only 5% of DNA bound to smectite, suggesting that most DNA was inaccessible. ??
- ▶ Although anecdotal, this observation points to several opportunities for improving deeptime DNA research, including using mineralogical characterization to identify the most promising sites for deep-time sedimentary DNA recovery and refining experimental approaches to recover DNA bound to all mineral surfaces. In the absence of improved methods to release bound DNA, microscopic evaluation of sedimentary samples will improve the efficiency of DNA recovery.

### What if there is no reference genome?

- ▶ Many species that are obvious targets for deep-time DNA research are extinct and some such as Xenocyon canids and basal members of the elephant and horse families have no evolutionarily close living relative for which an ideal reference genome can be produced. This presents challenges to ancient DNA authentication and identification as well as to reference-guided genome assembly.
- ► Finally, as reference-based taxonomic assignment is always limited to sequences deposited in public databases, the ongoing population of these databases will continue to improve robust identification of DNA recovered from Early and Middle Pleistocene remains and sediments.

### Sample dating

- One considerable challenge for studies of deep-time DNA is knowing the age of samples so that they can be placed into broader evolutionary and geological contexts.
- ▶ As most ancient DNA to date is from organisms that lived within the last several tens of thousands of years, it is usually possible to estimate their age directly using radiocarbon dating. Up to ~50 ka. Trapped charge dating methods, such as electron spin resonance (ESR) for tooth enamel or luminescence approaches for minerals such as quartz and feldspar, can provide age estimates for samples dating throughout the Pleistocene but require that sediments remain undisturbed since burial.