LETTER

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The genome of the offspring of a Neanderthal mother and a Denisovan father

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Neanderthals and Denisovans are extinct groups of hominins that separated from each other more than 390,000 years ago[1,2]. Here we present the genome of 'Denisova 11', a bone fragment from Denisova Cave (Russia)[3] and show that it comes from an individual who had a Neanderthal mother and a Denisovan father. The father, whose genome bears traces of Neanderthal ancestry, came from a population related to a later Denisovan found in the cave[4–6]. The mother came from a population more closely related to Neanderthals who lived later in Europe[2,7] than to an earlier Neanderthal found in Denisova Cave[8], suggesting that migrations of Neanderthals between eastern and western Eurasia occurred sometime after 120,000 years ago. The finding of a first-generation Neanderthal–Denisovan offspring among the small number of archaic specimens sequenced to date suggests that mixing between Late Pleistocene hominin groups was common when they met.

Neanderthals and Denisovans inhabited Eurasia until they were replaced by modern humans around 40,000 years ago (40 ka)[9]. Neanderthal remains have been found in western Eurasia[10], whereas physical remains of Denisovans have thus far been found only in Denisova Cave[4–6,11,12], where Neanderthal remains have also been recovered[8]. Although little is known about the morphology of Denisovans, their molars lack the derived traits that are typical of Neanderthals[5,11].

DNA recovered from individuals of both groups suggests that they diverged from each other more than 390 ka[1,2]. The presence of small amounts of Neanderthal DNA in the genome of 'Denisova 3', the first Denisovan individual to be identified[4–6], indicates that the two groups mixed with each other at least once[8]. It has also been shown that Neanderthals mixed with the ancestors of present-day non-Africans around 60 ka[2,8,13], and possibly with earlier ancestors of modern humans[1,14,15]; and that Denisovans mixed with the ancestors of present-day Oceanians and Asians[5,16,17]. Denisovans may furthermore have received gene flow from an archaic hominin that diverged more than a million years ago from the ancestors of modern humans[8].

A fragment of a long bone, 'Denisova 11' (Fig. 1), was identified among over 2,000 undiagnostic bone fragments excavated in Denisova Cave as being of hominin origin using collagen peptide mass fingerprinting[3]. Its mitochondrial (mt)DNA was found to be of the Neanderthal type and direct radiocarbon dating showed it to be more than 50,000 years old[3]. From its cortical thickness, we infer that Denisova 11 was at least 13 years old at death (Extended Data Fig. 1 and Supplementary Information 1). We performed six DNA extractions[18,19] from bone powder collected from the specimen, produced ten DNA libraries[20] from the extracts (Extended Data Table 1 and Supplementary Information 2, 3) and sequenced the Denisova 11 genome to an average coverage of 2.6-fold. The coverage of the X chromosome was similar to that of the autosomes, indicating that Denisova 11 was a female. Using three different methods, we estimate that contaminating present-day human DNA fragments constitute at most 1.7% of the data (Supplementary Information 2).

To determine from which hominin group Denisova 11 originated, we compared the proportions of DNA fragments that match derived alleles from a Neanderthal genome ('Altai Neanderthal', also known as 'Denisova 5') or a Denisovan genome (Denisova 3), both determined from bones discovered in Denisova Cave[6,8], as well as from a present-day African genome (Mbuti)[6] (Supplementary Information 4). At informative sites[1], 38.6% of fragments from Denisova 11 carried alleles matching the Neanderthal genome and 42.3% carried alleles matching the Denisovan genome (Fig. 2a), suggesting that both archaic groups contributed to the ancestry of Denisova 11 to approximately equal extents (Supplementary Information 4). Approximately equal proportions of Neanderthal-like and Denisovan-like alleles are found in each of the ten DNA libraries originating from Denisova 11 but not in libraries from other projects that were prepared, sequenced and processed in parallel, which excludes an accidental mixing of DNA in the laboratory or a systematic error in data processing (Supplementary Information 3).

To estimate the heterozygosity of Denisova 11, we restrict the analysis to transversion polymorphisms to prevent deamination-derived substitutions from inflating the estimates, and find 3.7 transversions per 10,000 autosomal base pairs. This is over four times higher than the heterozygosity of the two Neanderthal (Altai Neanderthal and 'Vindija 33.19') and one Denisovan (Denisova 3) genomes sequenced to date, and similar to the heterozygosity seen in present-day Africans. In fact, the heterozygosity of Denisova 11 is similar to what would be expected if this individual carried one set of chromosomes of Neanderthal origin and one of Denisovan origin, as estimated from the number of differences between randomly sampled DNA fragments from either the Vindija 33.19 or the Altai Neanderthal genome and the Denisova 3 genome (Fig. 2b and Supplementary Information 5).

Denisova 11 could have had approximately equal amounts of Neanderthal and Denisovan ancestry because she belonged to a population with mixed Neanderthal and Denisovan ancestry, or because her parents were each from one of these two groups. To determine which of these two scenarios fits the data best, we considered sites at which the genomes of the Altai Neanderthal and Denisova 3 carry a transversion difference in a homozygous form. At each of these sites, we recorded the alleles carried by two randomly drawn DNA fragments from Denisova 11. Note that in 50% of cases, both fragments will come from the same chromosome, making 50% of heterozygous sites appear homozygous. As a consequence, the expected proportion of apparent heterozygous sites is 50% for a first-generation (F_1) offspring, whereas it is 25% in a population at Hardy–Weinberg equilibrium with mixed ancestry in equal proportions (Supplementary Information 6). We find that in 43.5% of cases, one fragment from Denisova 11 matches the Neanderthal genome and the other matches the Denisovan genome, whereas in 27.3% and 29.2% of cases both fragments match the state

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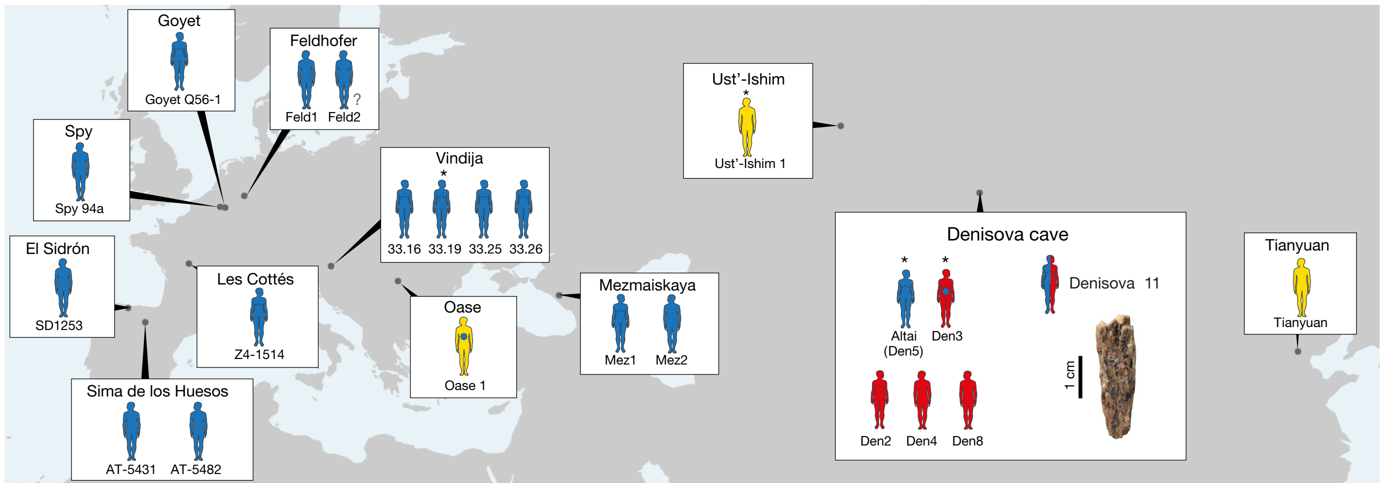

Fig. 1 | Location of Neanderthals, Denisovans and ancient modern humans dated to approximately 40 ka or earlier. Only individuals from whom sufficient nuclear DNA fragments have been recovered to enable their attribution to a hominin group are shown. Full or abbreviated names of specimens are shown near each individual. Blue, Neanderthals; red, Denisovans; yellow, ancient modern humans. Asterisks indicate that the genome was sequenced to high coverage; individuals with an unknown sex are marked with a question mark. Note that Oase 1 has recent Neanderthal ancestry (blue dot) that is higher than the amount seen in non-Africans. Denisova 3 has also been found to carry a small percentage of Neanderthal ancestry. Data were obtained from previous publications[1,2,5–8,11–13,21–24].

seen in the Neanderthal or the Denisovan genome, respectively (Fig. 2c). For comparison, when a low-coverage Neanderthal genome ('Goyet Q56-1')[7] is analysed in the same way, the two fragments match different states in 2.1% of cases, while they both match the Neanderthal state in 90.3% of cases and the Denisovan state in 7.5% of cases (Fig. 2c).

Obviously, the Altai Neanderthal and Denisova 3 are unlikely to be identical to the genomes of the individuals that contributed ancestry to Denisova 11. To take this into account, we used coalescent simulations to estimate the expected proportions of DNA fragments matching a Neanderthal or a Denisovan genome in populations with demographic histories similar to those of the Altai Neanderthal and Denisova 3 (Supplementary Information 6). The proportion of cases in which one of the two DNA fragments sampled from Denisova 11 matches the Neanderthal state and the other the Denisovan state fits the expectation for an F_1 Neanderthal–Denisovan offspring, but not an offspring of two F_1 individuals, an offspring of an F_1 parent and a Neanderthal or a Denisovan parent, nor an individual from a population of mixed ancestry at Hardy–Weinberg equilibrium (Extended Data Fig. 2 and Supplementary Information 6). We conclude that Denisova 11 did not originate from a population carrying equal proportions of Neanderthal and Denisovan ancestry. Rather, she was the offspring of a Neanderthal mother, who contributed her mtDNA, and a Denisovan father.

We next plotted the distribution of sites across the genome, for which Denisova 11 carries an allele matching the Altai Neanderthal genome and a different allele matching the Denisova 3 genome. Such sites are distributed largely uniformly (Fig. 3), as would be expected for an F_1 offspring of Neanderthal and Denisovan parents. To explore the ancestry of the parents of Denisova 11, we looked for regions in the genome that deviate from a pattern consistent with Denisova 11 being an F_1 offspring (Extended Data Fig. 3). Using four tests for enrichment of Denisovan or Neanderthal ancestry, we identify at least five approximately 1-Mb long (0.72–0.95 Mb) regions, all of

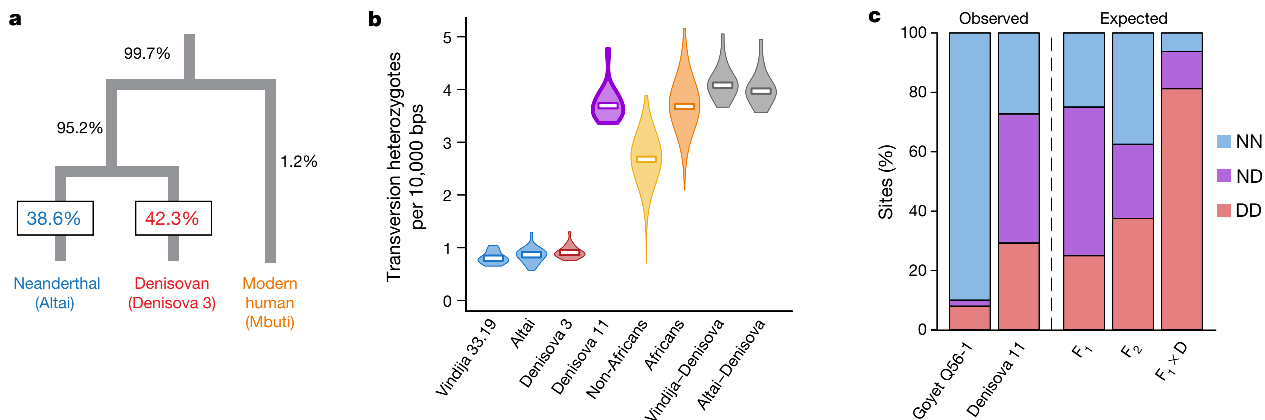

Fig. 2 | Denisova 11 has both Neanderthal and Denisovan ancestry. **a**, Percentage of DNA fragments from Denisova 11 matching derived alleles found on each branch of a tree relating a Neanderthal, a Denisovan and a present-day human genome. **b**, Distribution of heterozygosity per chromosome in two Neanderthals (blue), a Denisovan (red), Denisova 11 (purple) and present-day humans ($n = 235$ non-African individuals (yellow) and $n = 44$ African individuals (orange) from a previous publication[28]), and the expectation for a Neanderthal–Denisovan F_1 offspring (grey). The violins represent the distribution from the minimum and maximum heterozygosity values for the autosomes of each archaic hominin and of present-day humans ($n = 5,170$ pairs of chromosomes for non-Africans and $n = 968$ for Africans). White squares represent autosome-wide estimates for the archaic hominins, and the average of estimates across individuals for present-day humans. **c**, Percentage of sites at which two sampled DNA fragments both carry Neanderthal-like alleles (NN, blue), Denisovan-like alleles (DD, red), or one allele of each type (ND, purple); and the expectations for an offspring of a Neanderthal and a Denisovan (F_1), of two F_1 parents (F_2), and of an F_1 and a Denisovan ($F_1 \times D$). The expected proportions for simulated Neanderthal and Denisovan genomes are shown in Extended Data Fig. 2.

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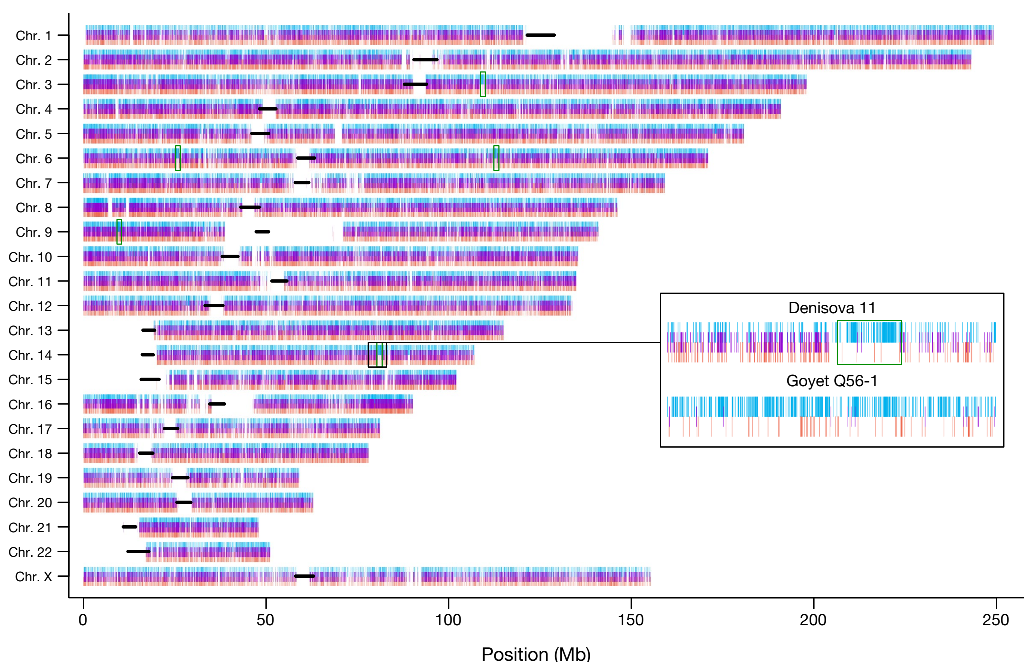

Fig. 3 | Distribution of Neanderthal-like and Denisovan-like alleles across the Denisova 11 genome. Positions for which one randomly drawn DNA fragment matches the Neanderthal genome and another matches the Denisovan genome are marked in purple. Positions are marked in blue if both DNA fragments match the Neanderthal genome and in red if both match the Denisovan genome. Black lines indicate centromeres. The inset shows one region out of five (green boxes) for which both chromosomes carry predominantly Neanderthal-like alleles. For comparison, the distribution of alleles in this region is shown for a Neanderthal genome (Goyet Q56-1).

which are homozygous for Neanderthal ancestry. This suggests that the Denisovan father of Denisova 11 had some Neanderthal ancestry. Given conservative estimates of the size and number of these regions, it is likely that there was more than one Neanderthal ancestor in his genealogy, possibly as far back as 300–600 generations before his lifetime (Supplementary Information 7). Notably, the heterozygosity in the regions of Neanderthal ancestry in Denisova 11 is higher than in the same regions in the genomes of Vindija 33.19 or the Altai Neanderthal, suggesting that the Neanderthals that contributed to the ancestry of Denisova 11's father were from a different population than her mother (Supplementary Information 5).

To explore how the mother of Denisova 11 was related to the two Neanderthals that have been sequenced to high coverage to date, we evaluated the proportions of fragments from Denisova 11 that match derived alleles from either of these two Neanderthal genomes. Denisova 11 shares derived alleles seen in the Altai Neanderthal genome in 12.4% of cases and those present in the Vindija 33.19 genome in 19.6% of cases, showing that the Neanderthal mother of Denisova 11 came from a population that was more closely related to Vindija 33.19 than to the Altai Neanderthal (Supplementary Information 8). We estimate the population split times of Denisova 11's Neanderthal mother from the ancestors of the Altai Neanderthal to approximately 20,000 years (20 kyr) before the time when the Altai Neanderthal lived, and her split time from the ancestors of Vindija 33.19 to around 40 kyr before Vindija 33.19. The population split between the Denisovan father of Denisova 11 and Denisova 3 is estimated to approximately 7 kyr before the latter individual (Supplementary Information 8). In Fig. 4, we present a population scenario that is compatible with these observations as well as with the population split times and molecular estimates of the ages of the three high-coverage archaic genomes[2]. We caution that the age estimates are associated with uncertainties, for example, regarding demography, mutation rates and generation times, and note that additional gene flow events are likely to have affected the population split times. Nevertheless, that a Neanderthal in Siberia who lived approximately 90 ka shared more alleles with Neanderthals who lived at least 20 kyr later in Europe[2,7] than with an earlier Neanderthal from the same cave[8] suggests that eastern Neanderthals spread into Western Europe

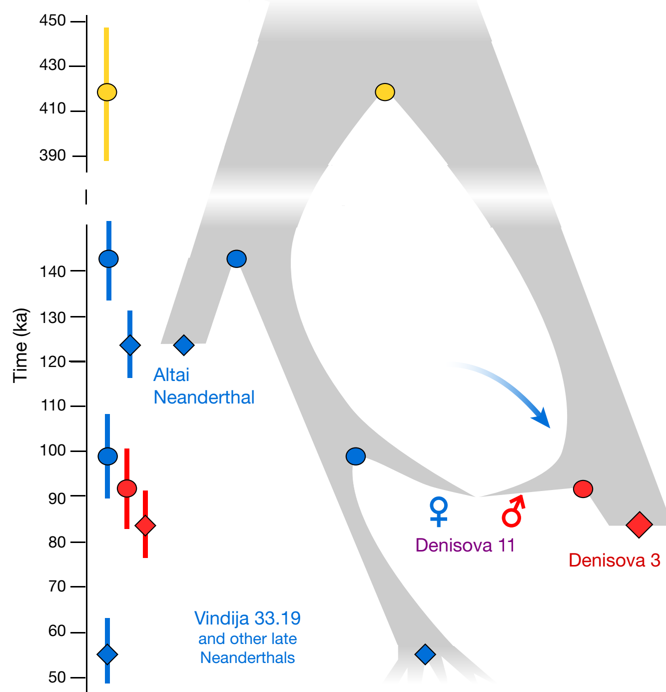

Fig. 4 | Relationships and gene flow events between Neanderthal and Denisovan populations inferred from genome sequences. Diamonds indicate ages of specimens estimated via branch shortening[2]; circles indicate population split times estimated from allele sharing between Denisova 11 and the high-coverage Neanderthal and Denisovan genomes (blue and red) and among the three high-coverage genomes (yellow, from a previous publication[2]). Markers indicate the means of these estimates, error bars indicate 95% confidence intervals based on block jackknife resampling across the genome ($n = 523$ blocks). Note that the confidence intervals do not take the uncertainty with respect to population size, mutation rates or generation times into account. Ages before present are based on a human–chimpanzee divergence of 13 million years[22,29]. The arrow indicates Neanderthal gene flow into Denisovans.

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sometime after 90 ka or that western Neanderthals spread to Siberia before that time and partially replaced the local population. These two non-mutually exclusive hypotheses could be tested by sequencing the genomes of early Neanderthals from Western Europe.

In conclusion, the genome of Denisova 11 provides direct evidence for genetic mixture between Neanderthals and Denisovans on at least two occasions: once between her Neanderthal mother and her Denisovan father, and at least once in the ancestry of her Denisovan father. Therefore, of the six individuals from Denisova Cave from whom nuclear DNA is available[5,6,8,11,12], two (Denisova 3 and Denisova 11) show evidence of gene flow between Neanderthals and Denisovans. We note that of the three genomes[21–24] retrieved from modern humans who lived at a time when Neanderthals were present in Eurasia (that is, approximately 40 ka or earlier)[9], one individual—'Oase 1'—had a Neanderthal ancestor four to six generations back in his family tree[23].

It is notable that one direct offspring of a Neanderthal and a Denisovan (Denisova 11) and one modern human with a close Neanderthal relative (Oase 1) have been identified among the few individuals from whom DNA has been retrieved and who lived at the time of overlap of these groups (Fig. 1). In conjunction with the presence of Neanderthal and Denisovan DNA in ancient and present-day people[2,5,8,13,16,17,25–27], this suggests that mixing among archaic and modern hominin groups may have been frequent when they met. However, Neanderthals inhabited western Eurasia[10] whereas Denisovans inhabited yet unknown parts of eastern Eurasia[5,17]. Thus, their zones of overlap may have been restricted in space and time. This, as well as possibly reduced fitness of individuals of mixed ancestry, may explain why Neanderthals and Denisovans remained genetically distinct. By contrast, the spread of modern humans across Eurasia after around 60,000 years ago may have allowed repeated interactions with archaic groups over a wider spatial range. Admixture between them may have resulted in archaic populations becoming partly absorbed into what were probably larger modern human populations[6,8].

Online content

Any methods, additional references, Nature Research reporting summaries, source data, statements of data availability and associated accession codes are available at https://doi.org/10.1038/s41586-018-0455-x.

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METHODS

Sampling and pre-treatment of bone powder. An overview of the laboratory experiments is shown in Extended Data Table 1. Bone powder was removed from the specimen using disposable sterile dentistry drills after the removal of a thin layer of surface material. Six samples were collected, each consisting of approximately 30 mg of bone powder. Because a previous analysis of the bone revealed that it is contaminated with present-day human DNA[3], each sample of bone powder was incubated with 1 ml 0.5% sodium hypochlorite solution as previously described[19] and as indicated in Extended Data Table 1, to reduce the amounts of present-day human and microbial DNA[7,19]. Residual sodium hypochlorite was removed by three consecutive 3-min washes with 1 ml water[19]. One extraction negative control (no powder) was included in each set of extractions.

DNA extraction and DNA library preparation. DNA was extracted using silica columns[18] as previously described[19], and eluted in 50 µl 10 mM Tris-HCl, 1 mM EDTA, 0.05% Tween-20, pH 8.0. Subsequently, 10 µl of each DNA extract (including the extraction negative controls) were used to prepare single-stranded DNA libraries as previously described[19,20]. A library preparation negative control was included in every experiment. Two additional 5-µl aliquots from extracts E3652 and E3655 were used to generate additional libraries (library preparation setup C in Extended Data Table 1), resulting in a total of 10 DNA libraries. The number of DNA molecules in the libraries was estimated by digital droplet PCR[30] or quantitative PCR[20]. Each library was amplified to the plateau while incorporating a pair of unique indexes[31] using 1 µM primers[19,31] and AccuPrime Pfx DNA polymerase (Life Technologies)[32]. Amplification products were purified using the MinElute PCR purification kit (Qiagen) or SPRI technology[33] on a Bravo NGS workstation (Agilent Technologies) as previously described[34]. Indexed DNA libraries were pooled with libraries from other projects. Heteroduplices, which confound DNA separation and concentration measurements in chromatography, were removed from the pools by single cycle amplification using Herculase II Fusion DNA polymerase (Agilent Technologies)[32] with primers IS5 and IS6[35]. Prior to deeper sequencing of libraries R5507, R5509, R9880, R9881, R9882, R9883 and R9873, heteroduplices were removed from each library separately. The concentration of DNA in each pool or each individual library, respectively, was determined using the electrophoresis system implemented on the DNA-1000 chip (Agilent Technologies).

Sequencing and data processing. Sequencing was performed on Illumina platforms (MiSeq or HiSeq 2500) using 76-cycle paired-end runs adapted to double-indexed libraries[31]. Bases were called using Bustard (Illumina). Adaptor sequences were trimmed and overlapping paired-end reads were merged into single sequences using leeHom[36]. Demultiplexing was carried out using jivebunny[7]. Sequences generated from a given library were merged using SAMtools[37] and aligned to the human reference genome (hg19/GRCh37) with the decoy sequences as previously described[2] using BWA[38] with parameters adjusted to ancient DNA[6]. PCR duplicates were collapsed using bam-rmdup (https://bitbucket.org/ustenzel/biohazard) and DNA fragments of length ≥35 bases that mapped within regions of unique mappability (Map35_100% from a previous publication[8]) with a mapping quality of 25 or higher[7] were used for analyses. Further filtering criteria used for certain analyses are described in the Supplementary Information.

Reporting summary. Further information on research design is available in the Nature Research Reporting Summary linked to this paper.

Code availability. The computer code used for simulations is included in Supplementary Information 6.

Data availability. Sequences generated from Denisova 11 have been deposited in the European Nucleotide Archive under study accession number PRJEB24663.

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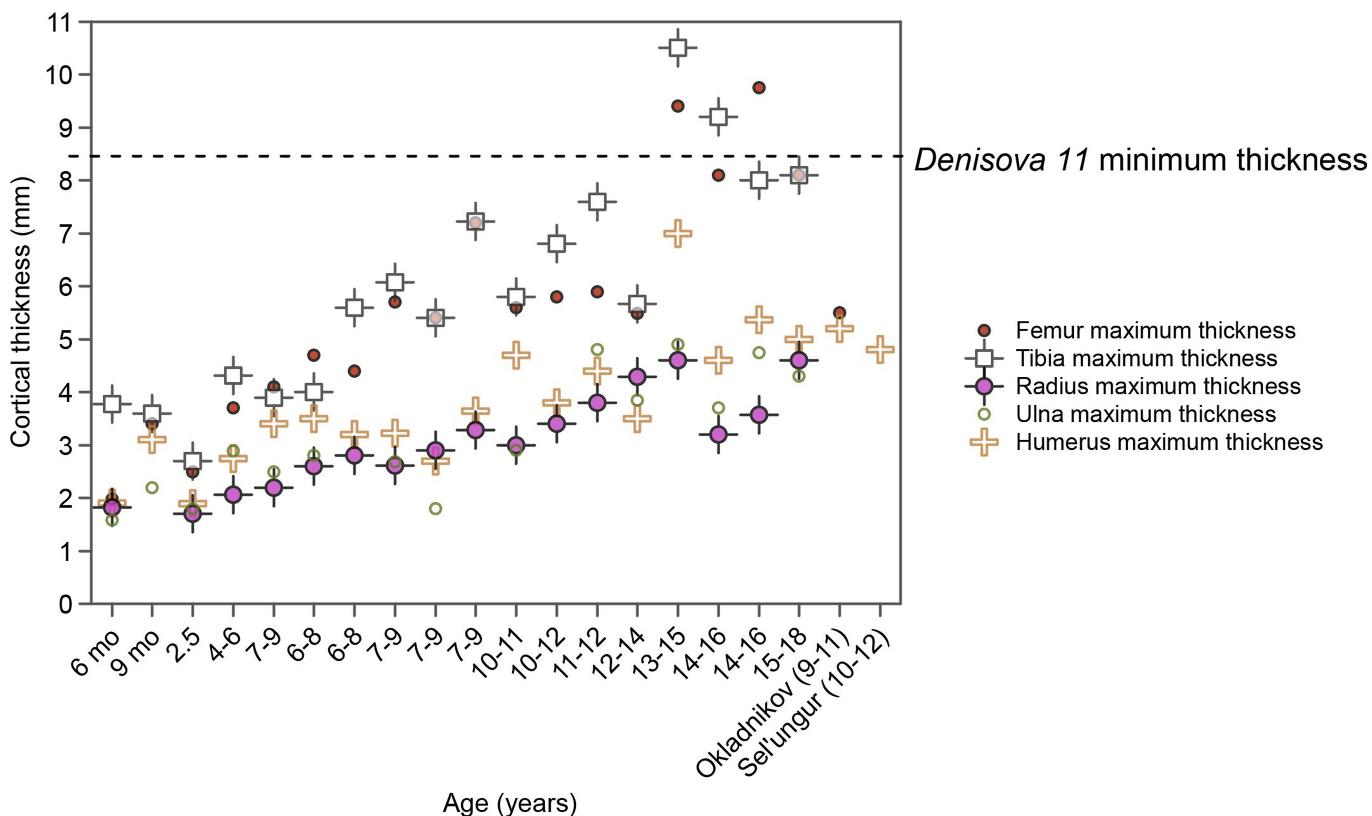

Extended Data Fig. 1 | Comparison between cortical thickness of long bones from modern humans, Neanderthals and Denisova 11. Maximum cortical thickness of femora, tibiae, humeri, radii and ulnae from humans from the Bronze Age and two Neanderthals compared to the minimum thickness of Denisova 11 (dashed line).

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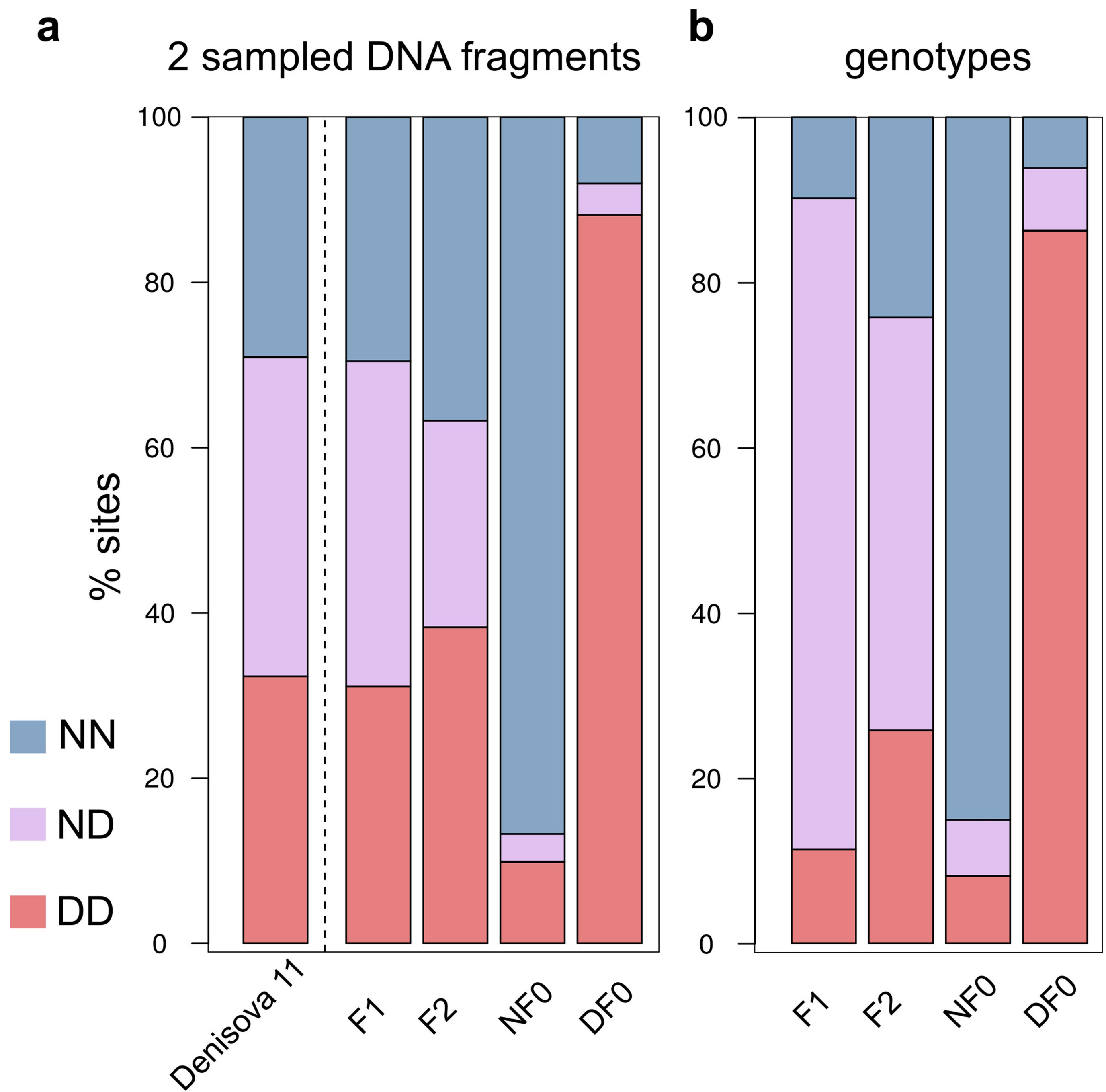

Extended Data Fig. 2 | Comparison of the genome of Denisova 11 and simulated genomes. Percentage of sites at which Denisova 11 and genomes simulated under the demographic model described in Supplementary Information 6 carry two Neanderthal alleles (NN, blue), two Denisovan alleles (DD, red) or one allele of each type (ND, purple). **a**, Percentages calculated for two random DNA fragments from Denisova 11 and from simulated F_1, F_2, Neanderthal (NF_0) or Denisovan (DF_0) genomes. **b**, Proportions of sites for the simulated genotypes, before sampling two fragments.

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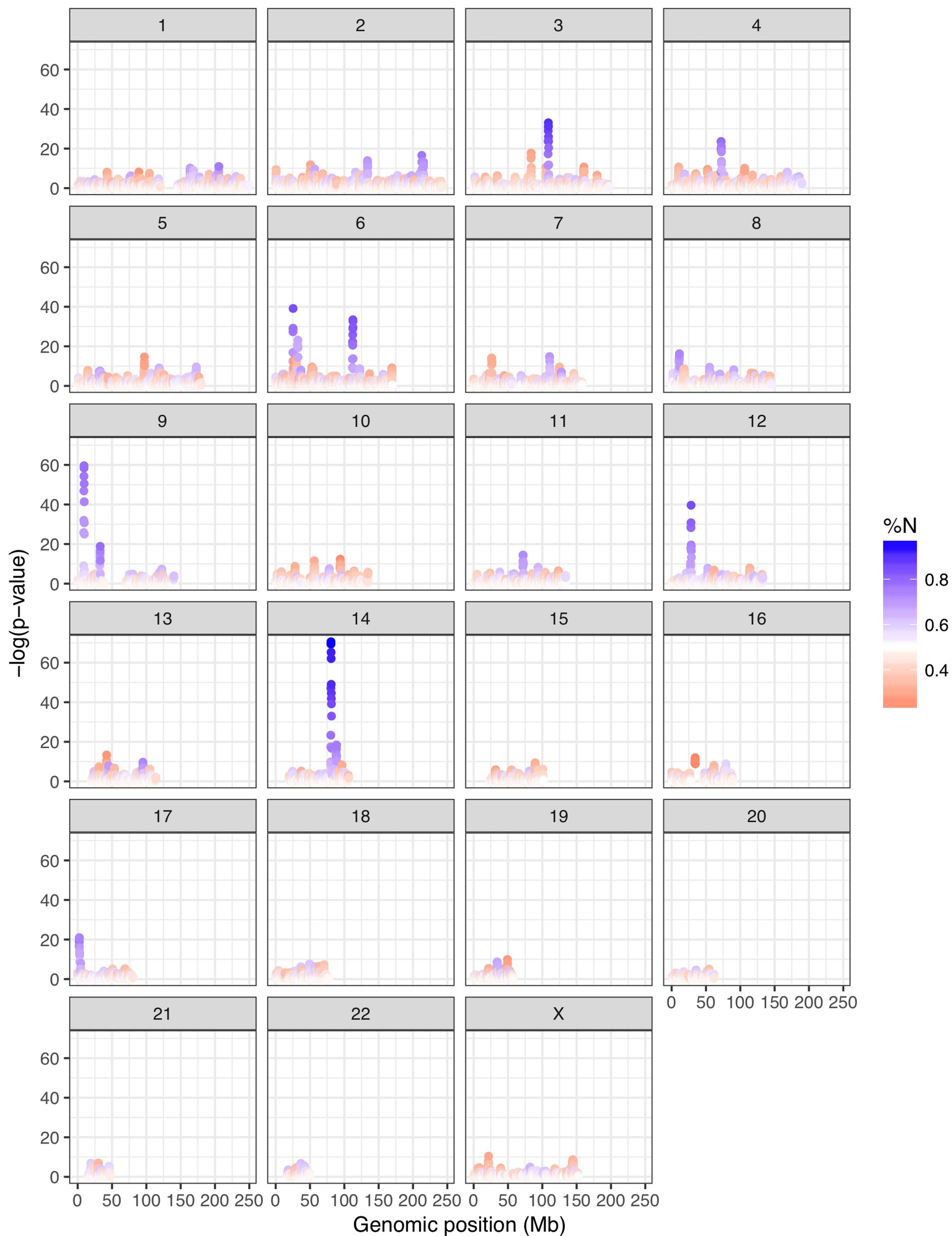

Extended Data Fig. 3 | Neanderthal and Denisovan allele proportions from Denisova 11 in 1-Mb windows. The *y* axis shows −log(*P*) of the deviation of Neanderthal and Denisovan allele counts from the genome-wide average (χ^2 test of goodness-of-fit; see Supplementary Information 7). The colour shows the proportion of alleles matching the Neanderthal state (%N) within each 1-Mb window (100-kb steps, $n = 26{,}414$ windows).

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Extended Data Table 1 | DNA extracts and DNA libraries prepared from the Denisova 11 specimen

	Extr. set	Bone powder [mg]	Pre-treatment [minutes]	Extract ID	Library prep. setup	Input in library [µl]	Molecules in library	Indexed library ID	DNA fragments sequenced	DNA fragments sequenced (L ≥35)	Mapped fragments (L ≥35, MQ ≥25)	Mapped fragments (%)	Unique fragments (L ≥35, MQ ≥25, Map35_100%)	Average fragment length [bp]	Fragments with C to T substitution
Denisova 11	1	27.4	15	E3259	A	10	2.27E+08	R5507	133,898,498	89,793,496	2,139,377	2.4	1,656,500	52.7	404,188
		27.8	15	E3261	A	10	2.00E+08	R5509	145,234,847	94,543,170	1,712,750	1.8	1,201,280	48.8	329,411
	2	29.0	15	E3652	B	10	4.33E+08 *	R5780	2,391,986	1,565,197	171,150	10.9	152,336	56.4	31,758
					C	5	4.63E+08	R9880	379,368,999	228,704,750	22,501,299	9.8	14,767,988	56.5	3,028,792
					C	5	4.03E+08	R9881	333,009,774	203,041,282	20,747,195	10.2	13,805,425	56.6	2,850,253
		29.7	15	E3654	B	10	4.16E+08 *	R5782	2,671,910	1,669,048	81,750	4.9	72,730	54.5	15,618
		33.5	15	E3655	B	10	3.49E+08 *	R5783	2,348,997	1,510,249	199,762	13.2	177,860	59.3	31,952
					C	5	4.19E+08	R9882	368,237,790	225,412,495	27,395,573	12.2	17,849,890	59.8	3,173,678
					C	5	3.69E+08	R9883	343,471,978	224,455,462	28,522,051	12.7	18,048,369	60.4	3,215,383
	3	27.1	30	E3922	C	10	7.43E+07	R9873	348,156,224	222,947,600	62,160,161	27.9	17,009,638	53.3	4,282,134
Controls	1	ENC	15	E3262	A	10	2.55E+07	R5510	12,220	4,123	38	0.9	35	55.2	2
		LNC	-	-	A	-	7.60E+06	R5521	12,444	2,170	11	0.5	10	47.2	1
	2	ENC	15	E3663	B	10	1.83E+06 *	R5791	32,008	4,183	473	11.3	412	51.2	8
		LNC	-	-	B	-	2.54E+06 *	R5792	31,455	2,908	70	2.4	58	49.0	3
	3	ENC	30	E3926	C	10	2.73E+07	R9877	61,825	13,861	2,472	17.8	2,145	57.0	9
		LNC	-	-	C	-	1.30E+07	R9888	68,130	5,275	100	1.9	67	46.2	6

Data are shown by DNA extraction set, and libraries prepared in the same setup are denoted with the same letter (A, B or C). Relevant negative controls are marked in grey. The number of molecules in each library was quantified by digital droplet PCR or quantitative PCR (denoted by asterisk). The numbers of DNA fragments sequenced per library are indicated for the combined data from all sequencing runs. Mapped fragments were counted if they were at least 35 bases long and mapped to the human reference genome with a mapping quality of 25 or higher; and their percentage was calculated out of sequenced fragments of length 35 bases or more. Following the removal of PCR duplicates, unique DNA fragments were retained if they mapped to the reference genome within the used mappability track. Such fragments were considered to contain a terminal cytosine (C) to thymine (T) substitution relative to the human reference genome if a putative cytosine deamination was within the first three or last three bases of the strand. bp, base pairs; ENC, extraction negative control; Extr., extraction; L, length; LNC, library preparation negative control; Map35_100%, previously published mappability track[8]; MQ, mapping quality; Prep., preparation.

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For final submission: please carefully check your responses for accuracy; you will not be able to make changes later.

Experimental design

1. Sample size

Describe how sample size was determined.

The sample size is 1, as we are determining the genome of a single individual. For comparisons with other archaic genomes, all available high-coverage Neandertal and Denisovan genomes (n=3) were used.

2. Data exclusions

Describe any data exclusions.

Sequencing data excluded based on pre-established criteria: sequences that did not map to the human genome, sequences that were shorter than 35 bases, sequences mapping with a low mapping quality or within regions of low mappability - all of which were excluded to avoid using sequences that were not endogenous to the individual sequenced.

3. Replication

Describe the measures taken to verify the reproducibility of the experimental findings.

The bone fragment was sampled on three different occasions, on different areas of the specimen (total of six samples of bone powder). Six DNA extracts were prepared on three occasions, and ten DNA libraries were generated in three different experiments. We show in SI 3 that the results are stable across all different libraries.
To allow the reproducibility of the analyses, all filtering steps and the comparative data used are detailed in the Methods section and the supplementary information; and the sequencing data generated here has been deposited in the European Nucleotide Archive.

4. Randomization

Describe how samples/organisms/participants were allocated into experimental groups.

This is not relevant for our study as we determined the genome of a single individual, therefore there were no experimental groups.

5. Blinding

Describe whether the investigators were blinded to group allocation during data collection and/or analysis.

Blinding was not relevant for our study, as this is the genome of a single individual.

Note: all in vivo studies must report how sample size was determined and whether blinding and randomization were used.

6. Statistical parameters

For all figures and tables that use statistical methods, confirm that the following items are present in relevant figure legends (or in the Methods section if additional space is needed).

n/a	Confirmed	
☐	☒	The exact sample size (*n*) for each experimental group/condition, given as a discrete number and unit of measurement (animals, litters, cultures, etc.)
☒	☐	A description of how samples were collected, noting whether measurements were taken from distinct samples or whether the same sample was measured repeatedly
☒	☐	A statement indicating how many times each experiment was replicated
☐	☒	The statistical test(s) used and whether they are one- or two-sided *Only common tests should be described solely by name; describe more complex techniques in the Methods section.*
☐	☒	A description of any assumptions or corrections, such as an adjustment for multiple comparisons
☒	☐	Test values indicating whether an effect is present *Provide confidence intervals or give results of significance tests (e.g. P values) as exact values whenever appropriate and with effect sizes noted.*
☐	☒	A clear description of statistics including central tendency (e.g. median, mean) and variation (e.g. standard deviation, interquartile range)
☐	☒	Clearly defined error bars in all relevant figure captions (with explicit mention of central tendency and variation)

See the web collection on statistics for biologists for further resources and guidance.

Software

Policy information about availability of computer code

7. Software

Describe the software used to analyze the data in this study.	Basecalling: Bustard; Adapter trimming: leeHom; Demultiplexing: jivebunny; Mapping: BWA; Handling bam files: SAMtools; PCR duplicates removal: bam-rmdup; F3- and F4-statistics: Admixtools; Identification of variable sites: heffalump; Heterozygosity estimates: snpAD; Demography simulations: scrm; HMM: pomegranate; Statistics: R.

For manuscripts utilizing custom algorithms or software that are central to the paper but not yet described in the published literature, software must be made available to editors and reviewers upon request. We strongly encourage code deposition in a community repository (e.g. GitHub). *Nature Methods* guidance for providing algorithms and software for publication provides further information on this topic.

Materials and reagents

Policy information about availability of materials

8. Materials availability

Indicate whether there are restrictions on availability of unique materials or if these materials are only available for distribution by a third party.	The sequencing data generated from the Denisova 11 bone fragment and the CT scans of the specimen are publicly available. Requests for further sampling of the bone fragment should be addressed to the Institute of Archaeology and Ethnography of the Russian Academy of Sciences.

9. Antibodies

Describe the antibodies used and how they were validated for use in the system under study (i.e. assay and species).	No antibodies were used.

10. Eukaryotic cell lines

a. State the source of each eukaryotic cell line used.	No eukaryotic cell lines were used.
b. Describe the method of cell line authentication used.	No eukaryotic cell lines were used.
c. Report whether the cell lines were tested for mycoplasma contamination.	No eukaryotic cell lines were used.
d. If any of the cell lines used are listed in the database of commonly misidentified cell lines maintained by ICLAC, provide a scientific rationale for their use.	No eukaryotic cell lines were used.

Animals and human research participants

Policy information about studies involving animals; when reporting animal research, follow the ARRIVE guidelines

11. Description of research animals

Provide all relevant details on animals and/or animal-derived materials used in the study.

No animals were used.

Policy information about studies involving human research participants

12. Description of human research participants

Describe the covariate-relevant population characteristics of the human research participants.

The research did not involve human participants.