

Correspondences

Mitochondrial DNA of an Iberian Neandertal suggests a population affinity with other European Neandertals

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Mitochondrial DNA (mtDNA) sequences have been retrieved so far from twelve Neandertal (*Homo neanderthalensis*) specimens: Feldhofer 1 and 2 in Germany [1,2], Mezmaiskaya in Russia [3], Monte Lessini in Italy [4], Vindija 75, 77 and 80 in Croatia [5,6], Engis 2 and Scladina in Belgium [6,7], La Chapelle-aux-Saints and Rochers de Villeneuve in France [6,8] and Sidron 441 in Spain [9]. There are only six Neandertal sequences (two from Feldhofer, two from Vindija, one from Mezmaiskaya and one from Monte Lessini) that cover a significant section (>300 bp) of the hypervariable region 1 (HVR1) of the mtDNA. Here we report the seventh extensive HVR1 mtDNA sequence of a Neandertal. The specimen stems from El Sidrón cave (Asturias, North of Spain) and was radiocarbon dated to around 43,000 years of age [9].

The retrieval and the analysis of Neandertal mtDNA sequences have allowed the exclusion of the possibility of a mitochondrial contribution of Neandertals to the modern human gene

pool [1,6]. In addition, a low genetic diversity was observed among Neandertals, similar to that observed among modern humans [5]. With the gradual accumulation of more Neandertal sequences, it should be possible to start exploring the phylogeography of Neandertals.

El Sidrón cave is a karstic system in Asturias (Northern Spain), where some Neandertal bones were accidentally discovered by cavers in 1994. The site has been under systematic excavation since 2000 [9] and has yielded several hundred skeletal remains, belonging to at least eight hominids. Most remains were found at the Galería del Osario (43° 23' 01" N, 5° 19' 44" W), which consists of a small lateral gallery leading around 220 meters deep into the karst system. Previously, one tooth was subjected to ancient DNA analysis and yielded a short 47 bp long Neandertal mtDNA sequence [10]. Although this suggested that the cave environment was favourable to ancient DNA preservation, the level of DNA fragmentation and the low ratio of endogenous versus contaminant sequences (around 5%) prevented the retrieval of more genetic data. To lower the rate of contamination with human DNA, an adult femur fragment designated for ancient DNA analysis was excavated in September 2005 under strict clean conditions and kept frozen at -20°C. To our knowledge, this is the first time that a Neandertal sample has been excavated with precautions specifically designed to prevent contamination and further DNA damage.

The sequence obtained by amplification, cloning and sequencing of overlapping fragments (Supplemental data) ranges from positions 16,076 to 16,378 (Figure 1) of the mitochondrial genome and is similar, but not identical, to previously published Neandertal HVR1 sequences. Compared to the Cambridge reference sequence for modern humans (CRS), it carries one substitution (C in 16,178) that was not

previously described for other Neandertals.

Genealogical analysis of the Neandertal mtDNA sequences with a reduced median network has shown two groups of sequences within the Neandertal mtDNA variation [4]. Some haplotypes, those of Mezmaiskaya and Monte Lessini, are clearly more diverse and branch near the root in the genealogy. Both have an A at position 16,078 and T at position 16,154 of the HVR1, while the rest of the sequences are more similar to each other and share a G in position 16,078 and a C in position 16,154; these positions are very stable in modern humans and, therefore, not prone to recurrent mutations. The described El Sidrón sequence belongs to the latter group, along with Feldhofer 1 and 2 and Vindija 75–80. Placing these data within the framework for the post-glacial recolonisation of Europe [11] suggests that Central European Neandertals (including those from Croatia) along with the Iberian Peninsula Neandertals might represent a relatively homogenous genetic group with a common

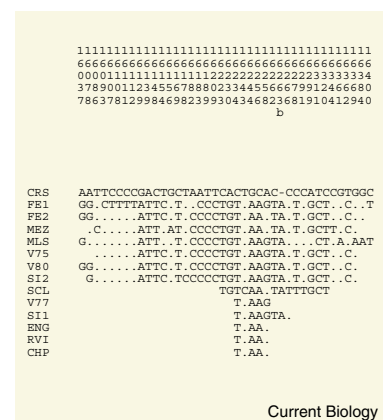


Figure 1. Genetic diversity of known Neandertals.

Variable sites in the comparison of the Cambridge reference sequence (CRS) and 12 Neandertal sequences, five of them represented by short fragments. Abbreviations: CHP = La Chapelle aux Saints; ENG = Engis 2, FE1 = Feldhofer 1, FE2 = Feldhofer 2, MEZ = Mezmaiskaya, MLS = Monti Lessini, RVI = Rochers de Villeneuve, SCL=Scladina, SI2= El Sidrón bone (present study), SI1 = El Sidrón 441, V75 = Vindija 75, V77 = Vindija 77, V80 = Vindija 80.

demographic history that is probably different to that of other Neandertal groups placed in Eastern Europe and in the Italian Peninsula.

The most recent common ancestor of the seven Neandertal sequences has been estimated to be around 250,000 ± 65,000 years of age (assuming an age of around 40,000 years for these specimens) by coalescent methods (Supplemental data). The clade containing the four most similar sequences (Feldhofer 1 and 2, Vindija 75-80 and El Sidrón, between positions 16,076-16,378) sharing the G16,078-C16,154 haplotype, has been estimated to be 130,000 ± 30,000 years of age. Interestingly, this date is roughly concordant with the end of a dramatic glacial maximum around 135,000 years ago [12]. Therefore, it seems that, despite the broad geographic range, the El Sidrón, Feldhofer and Vindija Neandertals may belong to the same group of Neandertals that expanded from a hypothetical southern glacial refugium after a demographic collapse associated with this glacial maximum.

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Supplemental data

Supplemental data are available at <http://www.current-biology.com/cgi/content/full/16/16/R629/DC1/>

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A highly divergent mtDNA sequence in a Neandertal individual from Italy

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Neandertals are documented in Europe and Western Asia from about 230,000 to 29,000 years ago. Analyses of mitochondrial DNA (mtDNA) from Neandertal samples [1,2] and other analyses [3–5] appear incompatible with the hypothesis that Neandertals are direct ancestors of modern Europeans [6,7]. However, there are broad geographic gaps in the sampling of Neandertal DNA diversity. Here, we describe the sequence of the first mitochondrial hypervariable region

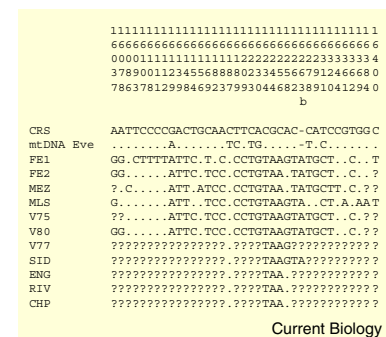


Figure 1. Neandertal diversity. Comparison of the Cambridge reference sequence (CRS) and the putatively ancestral sequence to all extant human variation (mtDNA Eve) with 11 Neandertal sequences, five of them represented by short fragments. CHP = La Chapelle aux Saints, CRS = Cambridge reference sequence, ENG = Engis 2, FE1 = Feldhofer 1, FE2 = Feldhofer 2, MEZ = Mezmaiskaya, MLS = Monti Lessini, RIV = Rochers de Villeneuve, SID = El Sidrón 441, V75 = Vindija 75, V77 = Vindija 77, V80 = Vindija 80.