COMMENTARY

The last Neanderthal

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The mechanism of the Neanderthal extinction and their replacement by modern humans of African origin is one of the most discussed issues in paleoanthropology. Central to this discussion are the questions of the chronological overlap between Neanderthal populations and modern humans in Western Eurasia and the precise geographical circumstances of this overlap. For a long time, the Vindija (Croatia) site was considered to provide solid evidence for a long survival of Neanderthals in Central/Southern Europe. Not only did directly dated Neanderthal remains from layer G1 of the site provide radiocarbon ages postdating the most widely accepted transition time of 40–35,000 radiocarbon years ago (1), but the same layer also yielded a type of split-based bone points commonly assigned to the Aurignacian (2), a stone artefact industry of the early Upper Paleolithic that, to date, only yielded human remains of a modern nature (3). For some, this situation implied the possibility of a long and complex interaction between the two groups of hominins in this region and also falsified the notion of a systematic association between defined archaeological assemblages and specific biological populations at the time of the replacement. In PNAS, Devièse et al. (4) provide new radiocarbon dates for the same Vindija Neanderthal samples, dating them to before 40,000 ^{14}C B.P., significantly older than previous efforts dating this material to 29–28,000 and 33–32,000 radiocarbon years (1). The bone points of layer G1 could not be dated, but the range of ages obtained from faunal and human samples in this layer suggests taphonomic mixing as a likely mechanism to explain their stratigraphic association in this part of the Vindija stratigraphic sequence. The situation in Vindija is therefore not at all exceptional, and previous results can be explained by the effect of sample contamination and layer admixture.

The first message delivered by the new study is one of prudence. Radiocarbon is the most precise method of directly dating human fossil remains. However, at the limit of its application range around 45–40,000 calendar years ago, which unfortunately corresponds to the period of replacement of the last Neanderthals, it is highly sensitive to contamination. For a radiocarbon date of 25,000 y, 1% contamination by modern carbon will produce an age that is 1,628 y too young, but for a radiocarbon date of 42,500 y, the apparent age shifts 8,455 y toward the present (Fig. 1). This length of time is in the higher range of estimates for possible overlap between Neanderthals and modern humans at the scale of Western Eurasia (3, 5). It is therefore critical when dating organic material extracted from fossil bones to authenticate it as composed of degraded proteins, essentially collagen, of the bone itself. From this perspective, the development of extraction techniques implementing "ultrafiltration" in order to eliminate small contaminants represented a major step forward (6). Ever since, sample pretreatments have witnessed several important improvements. In practical terms, these successive advances mean that, for the transition period, the large number of radiocarbon dates on bone samples produced before 2004 should be used with great caution, if not simply forgotten. To overcome contamination issues in an even more thorough way, Devièse et al. (4) used a method based on the extraction of hydroxyproline, an amino acid specific to collagen. Hydroxyproline dating relies on significant bone sample sizes, as the dated carbon atoms are restricted to those deriving from a single amino acid making up roughly 10% of all amino acid positions of mature collagen type I. Hence, bone sample sizes for hydroxyproline dating are larger than

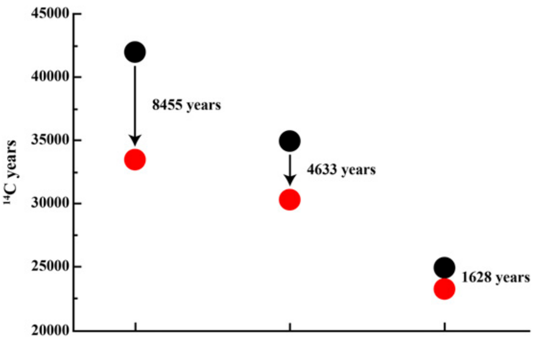

Fig. 1. Contamination effect on radiocarbon dates. The black dots represent three examples of radiocarbon dates without contamination, and the red dots represent the effect of only 1% contamination by modern carbon on the same samples.

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commonly used for ultrafiltration pretreatment and result in greater SEs than with the ultrafiltration technique (table 2 of ref. 4). In the present state of the art, this approach can only be envisioned when large fragments bearing little anatomical information are available. New methods, such as collagen peptide mass fingerprinting (ZooMS), might provide access to such human bone specimens by large-scale screening projects (7).

Since other cases of Neanderthal late survival, such as Ripparo Mezzena in Northern Italy (8), have now been falsified, the latest occurrences of Neanderthal remains in the European fossil record can now be documented by specimens discovered in association with so-called "transitional industries." These industries display features inherited from local Middle Paleolithic assemblages produced by Neanderthals, combined with Upper Paleolithic innovations similar to those encountered in the Aurignacian. At Saint-Césaire and at the Grotte du Renne, France, Neanderthal remains associated with a transitional industry called Châtelperronian, which already displays many Upper Paleolithic features, were directly dated to 36,200 ± 750 and 36,840 ± 660 ^{14}C B.P., respectively (7, 9). Another series of Neanderthal remains from the cave of Spy, Belgium, has also been directly dated to *ca.* 36,000 ^{14}C B.P. (10). This age falls in the time range of the Lincombian-Ranisian-Jerzmanowician (LRJ), a European transitional industry identified in this site. If one assumes that the entire Châtelperronian and LRJ were produced by late Neanderthals, this would push the last occurrence date for these populations to *ca.* 35,000 ^{14}C B.P. Converting this radiocarbon date into a calendar age corresponds to ∼40,000 y ago. By that time, modern humans producing Aurignacian industries had already occupied the neighboring regions of Austria (11), Germany (12), and Northern Italy (13) for some time. Interactions with modern groups may explain the cultural evolution of the last Neanderthals, but a puzzling issue remains regarding possible biological interactions between the two groups. Introgression of Neanderthal DNA into the modern genome has been widely demonstrated and analyzed through the endless availability of extant genetic data. It is quite likely that gene flow also occurred in the opposite direction. However, to date not a single detailed nuclear DNA sequence has been reconstructed from one of these postcontact Neanderthal populations and it is therefore impossible to assess the magnitude and possible effect of modern DNA introgression into late Neanderthal populations.

As far as archaeological assemblages of Middle Paleolithic type can be considered a good proxy to identify the occurrence of Neanderthal populations in different parts of Western Europe, the south of the Iberian Peninsula has sometimes been proposed as an area of their late survival. Specifically, south of the Ebro river, Neanderthals may have survived several millennia after their extinction in the rest of Western Europe (14). This idea is heavily disputed, as the dating of some of the sites on which this model was based have been revised to older ages (15). Still, sites in the Mula basin of Murcia, Spain, are claimed to document a modern replacement of Neanderthals taking place as late as around 37,000 y ago in calibrated chronology (16). The explanations provided for this delay are primarily geographical and environmental, and partly relate to the expansion of forested environments during this time period in Iberian regions south of 40°N.

If one takes a broader view away from the European continent, a series of recent studies have supported early modern human expansion in tropical Asia, as far as China (17), Laos (18), and Indonesia (19), and ultimately in Australia (20) more than 60,000 y ago. If substantiated by further discoveries, this situation would set the far-west Eurasia as a region of delayed replacement for archaic local populations. If modern humans were then able to settle in Asian tropical forests and cross large bodies of water, it is difficult to conceive that their expansion in the Iberian Peninsula would have been stalled by a river and deciduous temperate forests. Regardless of the "Ebro frontier" issue, it is striking to consider that the places where the latest survival of Neanderthals is best documented by the direct dating of diagnostic human remains actually correspond to areas where Neanderthal populations displayed the highest density and a continuous occupation for the tens of millennia preceding their final demise. This suggests that although environmental conditions might have influenced the tempo of modern human penetration in Europe, the decisive factor that slowed their settlement could have been the very presence of relatively dense Neanderthal populations well adapted to the Pleistocene environments of the middle latitudes.

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The last chapter of the Vindija saga reminds us once more that the interpretation of material from older excavations is quite challenging, as the precise archaeological context of discoveries that took place several decades ago generally remains a topic of continuous debate. The resolution of pending issues regarding the European expansion of modern humans will come from the study of new sites or the reassessment of earlier discovered ones with entirely new approaches. Without relying too much on the hope of discoveries, such as spectacular human remains, major progress can be accomplished in the near future through the implementation of new methodological approaches ranging from the screening of anatomically nondiagnostic human bone remains out of faunal assemblages by peptide mass fingerprinting of collagen to improvements to come in the reduction of sample sizes for direct radiocarbon dating.

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