

High-resolution U-series dates from the Sima de los Huesos hominids yields 600_{-66}^{66} kyrs: implications for the evolution of the early Neanderthal lineage

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Abstract

The Sima de los Huesos site of the Atapuerca complex near Burgos, Spain contains the skeletal remains of at least 28 individuals in a mud-breccia underlying an accumulation of the Middle Pleistocene cave bear (*Ursus deningeri*). We report here on new high-precision dates on the recently discovered speleothem SRA-3 overlaying human bones within the Sima de los Huesos. Earlier analyses of this speleothem by TIMS (thermal-ionization mass-spectrometry) showed the lower part to be indistinguishable from internal isotopic equilibrium at the precision of the TIMS instrumentation used, yielding minimum age of 350 kyr (kyr $\frac{1}{4}$ 10^3 yr before present). Reanalysis of six samples of SRA-3 by inductively-coupled plasma-multicollector mass-spectrometry (ICP-MS) produced high-precision analytical results allowing calculation of finite dates. The new dates cluster around 600 kyr. A conservative conclusion takes the lower error limit ages as the minimum age of the speleothem, or 530 kyr. This places the SH hominids at the very beginnings of the Neanderthal evolutionary lineage.

Keywords: Atapuerca; Sima de los Huesos; Middle Pleistocene; Neanderthal; Uranium-series

1. Introduction

We report on new high-precision dates on the SRA-3 speleothem that overlies human fossils at the site of the Sima de los Huesos (SH) in the Sierra de Atapuerca near Burgos, Spain (Fig. 1). Deep within the Cueva Mayor system and far removed from any modern entrance, the Sima de los Huesos (SH, "pit of the bones") contains an enigmatic accumulation

of Middle Pleistocene fossil humans. Well preserved remains of at least 28 human individuals (Bermúdez de Castro et al., 2004), fragmented and scattered, are found in a mud-breccia (Fig. 2) laying stratigraphically below a jumble of bones of the Middle Pleistocene cave bear (*Ursus deningeri*). The SH collection now comprises more than 80% of the Middle Pleistocene record world-wide for the genus *Homo* and provides for an unprecedented study of within-population variations (Arsuaga et al., 1997c; Lorenzo et al., 1998). The SH hominids are the evolutionary ancestors to the Neanderthals (Arsuaga et al., 1991, 1993; Arsuaga et al., 1997a,b) and thus, dating of the deposit is clearly of great importance.

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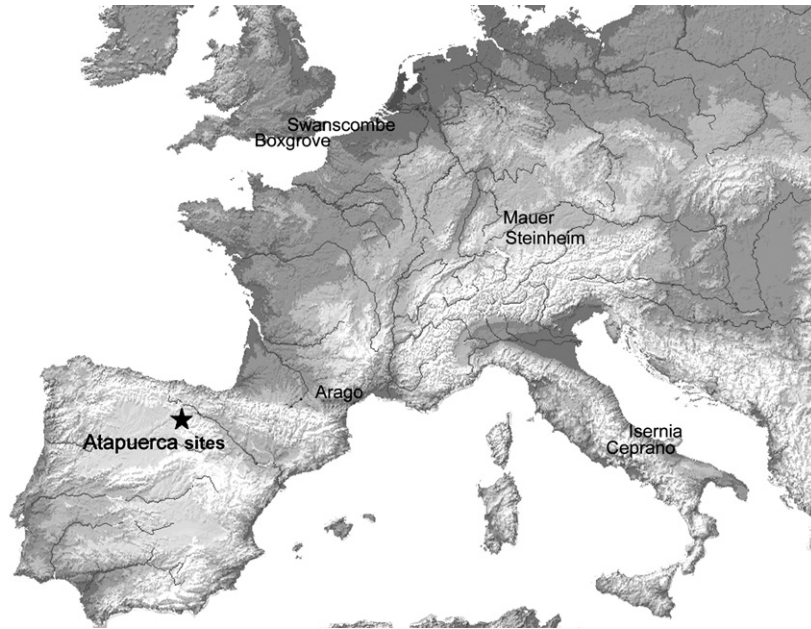


Fig. 1. Location of Atapuerca.

2. Stratigraphy and previous dating

The area of excavation (Figs. 3 and 4) consists of two connected, sediment-filled chambers, the Rampa and the Sima de los Huesos proper. The Rampa is an inclined 9 m long chamber at the foot of which the Sima de Los Huesos proper opens into an oblong 8 m by 4 m chamber. The vast majority of the human fossils were recovered from area B in the Sima proper (Figs. 2e4), in a muddy matrix-supported bonebreccia. The excavation has not yet reached bedrock at B but has extended down into the sterile sandy unit below. Three 1 m² pits were excavated in the Rampa, at the top (SRA), midpoint (SRM) and foot (SRB). Each pit exposes about 1.5 m of section. Since 2003, over 55 human fossils have been recovered from below a recently discovered speleothem at SRA while only a few human bones have been recovered at SRB, and SRM. A jaw fragment (AT 75) with two teeth (left M₂eM₃), designated as



Fig. 2. Breccia of human bones in the Sima de Los Huesos at Area B, showing three crania.

Individual 6 from the site, was originally recovered in the Sima proper from area A (square Q 10), together with an isolated tooth (AT-1760 ¼ left P₃) from the same individual (also found in Q 10). Another tooth from this individual (AT-1763 ¼ left P₄) was subsequently found at SRB, and yet another (AT-1759 ¼ left M₁) from area B (square T-13)(Bermúdez de Castro et al., 2004). One tooth from SRA (AT-4328 ¼ right P₃) has also been assigned to Individual 6, based on the compatibility of the wear stages and anatomical similarity in the morphology of the cusps. Thus, Individual 6 is represented in SRA, SRB, area B and area A of the Sima; a distribution which covers the entire extent of the Sima deposits (Figs. 3 and 4). This distribution suggests that the date for the deposit established in SRA also applies to the rest of the Rampa and SH proper.

The entire sequence of human remains is capped by a sheet of speleothem flowstone (Colada), generally earthy and impure. U-series and radiocarbon dating indicates the Colada formed from about 68 kyr to about 25 kyr. The range of U-series nominal dates for 25 bear bones (88e220 kyr) and for 16 human bones (114e182 kyr) that underlie the Colada are similar and rather broad, but are clearly affected by irregular post-depositional uranium cycling. Nine additional bear bones were analyzed by the combined ESR and U-series method (Bischoff et al., 1997). Dates for six of these yielded 200 ± 4 kyr whereas the other three yielded dates of 320 ± 4 kyr. Thus, the earlier results seemed to provide a firm minimum age of about 200 kyr for the human entry; and suggestive evidence of possible entry prior to 320 kyr.

3. SRA-3 speleothem

During the 2001 field season excavation at SRA, the top of the Rampa was extended up slope. This excavation exposed

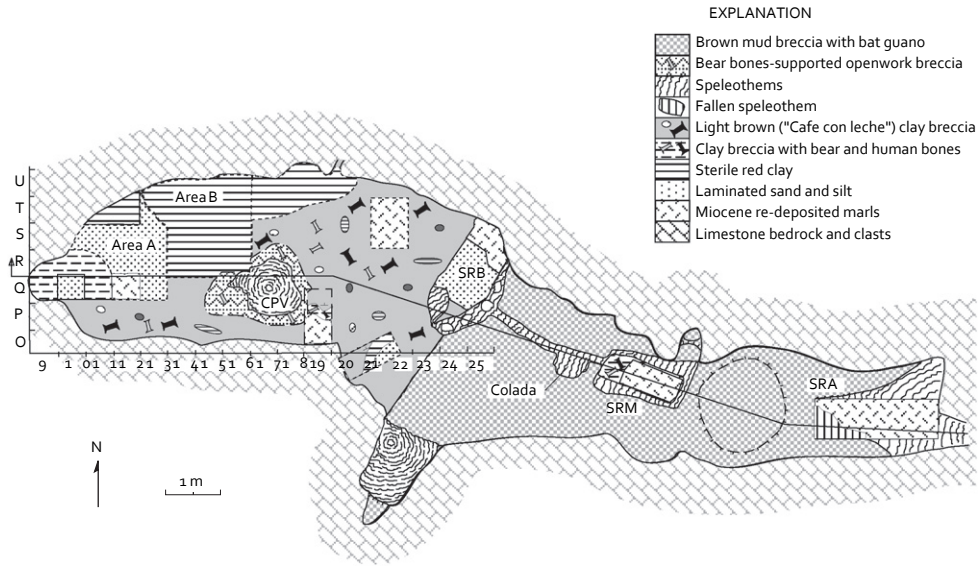


Fig. 3. Plan view of the Sima de los Huesos showing presently exposed sedimentary units and location of excavated areas. SRA, SRM, and SRB are test pits cut into the inclined ramp (Rampa) and Areas A and B are within the Sima proper. Hominid bones are most abundant at Area B but have also been recovered at Area A, SRA, SRM, and SRB. Numbers and letters (key axes) refer to the half-meter intervals of the excavation grid. Projection of entrance shaft is shown between SRA and SRM.

a 14-cm thick in situ speleothem (SRA 3, Figs. 4 and 5) lying stratigraphically beneath the Colada speleothem and, therefore, older than the Colada. Immediately below the SRA 3 speleothem, bones belonging to at least two individuals of the bear species, *U. deningeri*, were recovered, and among the bear bones two human phalanges were found. Subsequent

excavations have recovered over 55 additional human fossils from below the SRA 3 speleothem. The newly exposed speleothem was examined carefully to establish that it is, indeed, in situ, that it formed in place covering the bones. The lowermost 1 cm contains fragments of the underlying sediment and small fragments of bone. Therefore SRA-3 is clearly younger than

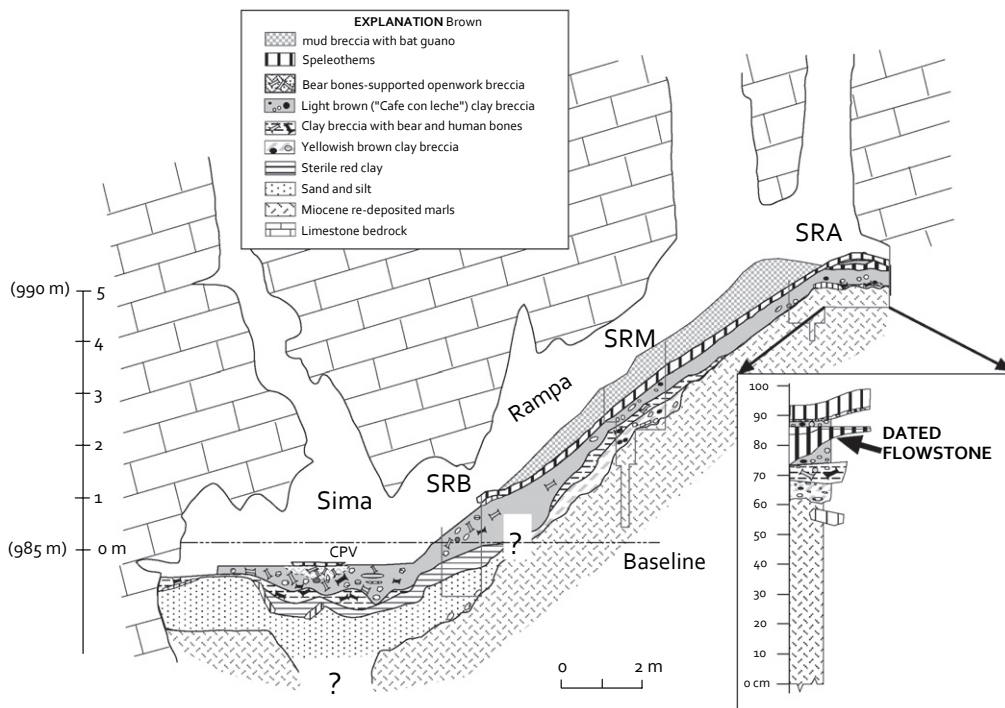


Fig. 4. Interpretive cross-section of the Sima de los Huesos, taken along axis shown in Fig. 3; Vertical exaggeration is 1.14. The largest concentration of human and bear fossils is at B. Dated speleothem (unit 3) and position of newly discovered human bones is at SRA shown in the inset stratigraphic column.

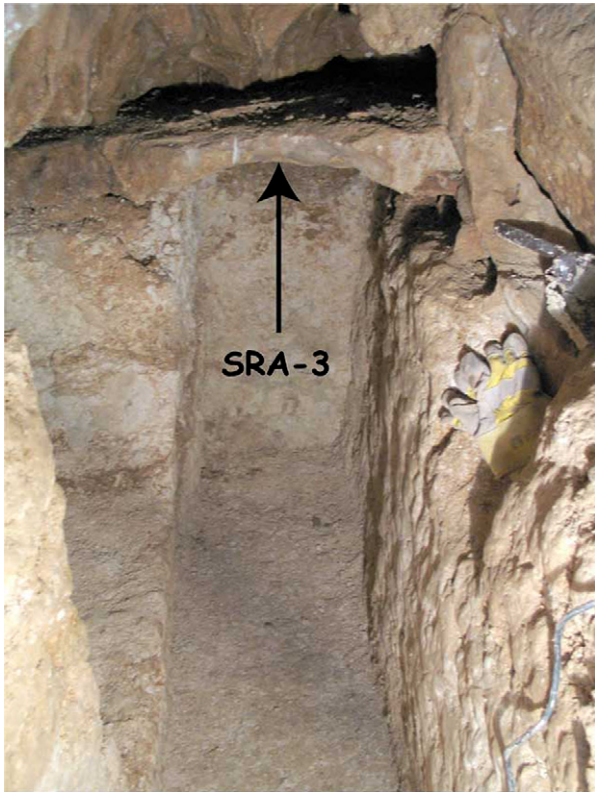


Fig. 5. Photo of SRA-3 speleothem *in situ*. Gloves at side of pit provide scale.

the human bones. The speleothem is of high purity and crystallinity and, therefore, excellent material for U-series dating to provide a minimum age for the human bones. The speleothem is laminated (*ca.* 1 cm laminae, Fig. 6), is pure calcite, containing less than 0.3 wt% organic carbon and containing exceptionally low amounts of detrital contamination. The laminae truncate at about 4 cm below the top, representing a hiatus in speleothem growth.

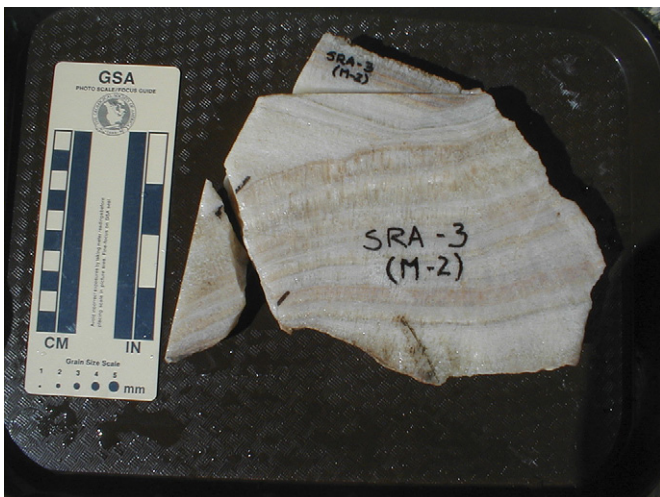


Fig. 6. Photo of slab cut through SRA-3 speleothem showing hiatus at 4 cm below top. Six samples analyzed in present study were taken from 5 through 13 cm down from the top.

U-series analyses by thermal-ionization mass-spectrometry (TIMS) of 14 samples reported in 2003 (Bischoff et al., 2003) were taken about every centimeter spanning the entire 14 cm thickness. Results indicated that the uranium contents are relatively high averaging 0.6 ppm, and the $^{230}\text{Th}/^{232}\text{Th}$ ratios are all well in excess of 20, the limiting value below which extraneous (detrital) Th significantly affects the date (Bischoff and Fitzpatrick, 1991). Three samples above the hiatus yielded finite dates in stratigraphic order from 153 to 281 kyr. Those below the hiatus were indistinguishable from internal isotopic equilibrium at the precision we were able to obtain from the TIMS the instrumentation used at the time, yielding minimum age of 350 kyr.

4. New ICP-MS analyses of SRA-3

Thermal-ionization mass-spectroscopy (TIMS) has revolutionized U-series dating by providing direct analysis of the isotopes on samples as small as a few tens of milligrams with long-term standard reproducibility of around 1% (Shen et al., 2002). Multi-collector inductively-coupled plasma mass-spectrometry (MC ICP-MS) has improved Th ionization, and therefore, improved sensitivity and, for a given sample size, improved analytical precision over TIMS (Luo et al., 1997; Shen et al., 2002). The new analyses reported here were made with an IsoProbe MC ICP-MS (GV Instruments/Thermo Electron Corporation) at Lawrence Livermore National Laboratory. We resampled six points below the hiatus, from 5 to 13 cm across the slabbed flowstone to apply the improved precision, in hopes of extending the minimum age estimate. We also analyzed aliquots of equilibrium standards for comparison, two of a solution of the Schwartzwalder Mine secular-equilibrium standard (Ludwig et al., 1985), and three of a solution of the Congo Pitchblend secular-equilibrium standard. After acid dissolution, Th and U isotopes were isolated by anion exchange. We use ^{233}U and ^{229}Th spikes obtained from the National Institute of Standards and Technology, and calibrated them against an elemental standard of U and the Harwell Uraninite equilibrium standard, HU-1. Samples (0.1–1 g) were dissolved in HNO_3 , with excess H_2O_2 to remove organics. Spikes of ^{233}U and ^{229}Th were added such that $^{233}\text{U}/^{235}\text{U}$ mass ratio is less than 100 and $^{229}\text{Th}/^{230}\text{Th} \geq 100$. After complete dissolution the solution was boiled to remove all H_2O_2 and allowed to sit overnight to allow equilibration between spike and natural isotopes. The dried residue was dissolved and loaded onto an anion exchange resin in HCl-form (Bio-Rad Laboratories AG-1-X8, 100–200 mesh size) for isolation of U isotopes, followed by a second column in HNO_3 -form to isolate Th isotopes.

Analyses of the purified Th and U fractions for ^{230}Th and ^{234}U are made by MC ICP-MS. The small beam currents from these isotopes were measured by pulse counting on a Daly detector, while the spike isotopes ^{229}Th and ^{233}U were measured simultaneously on a Faraday cup. The Daly/Faraday cross-calibration factor for the isotope dilution analyses and the mass bias corrections were determined using a natural uranium standard (NIST 4321C), and confirmed by quality control analyses of NBS U010 isotopic uranium standard.

Table 1

U-series analyses by ICP-MS and derived dates of SRA-3 speleothem overlying hominin fossils from the Sima de Los Huesos, Atapuerca. Half-lives of ^{234}U and ^{230}Th taken from Cheng et al. (2000)

Below top (cm)	USGS Lab ^b	U (ppm)	$^{234}\text{U}/^{238}\text{U}$	$^{230}\text{Th}/^{232}\text{Th}$	$^{230}\text{Th}/^{234}\text{U}$	$^{230}\text{Th}/^{234}\text{U}$ equilibrium ^a	Date (kyr)	Minimum age ^b
5	05-6	0.506 ± 0.0009	1.0144 ± 0.0027	200	1.004 ± 0.0036	1.0063	570 ⁺¹⁰³ ₋₅₃	>517
7	05-7	0.620 ± 0.0010	1.0148 ± 0.0031	730	1.0031 ± 0.0036	1.0065	632 ^{+∞} ₋₈₁	>551
9	05-8	0.542 ± 0.009	1.0155 ± 0.003	87	1.0024 ± 0.0036	1.0068	604 ⁺¹⁸⁹ ₋₆₇	>537
10	05-15	0.560 ± 0.001	1.0160 ± 0.0030	210	1.0007 ± 0.0031	1.007	563 ⁺⁷⁶ ₋₄₅	>518
10.5	05-9	0.6000 ± 0.001	1.0131 ± 0.0028	93	1.0033 ± 0.0034	1.0058	668 ^{+∞} ₋₉₇	>571
13	05-14	0.7424 ± 0.001	1.0145 ± 0.0034	430	1.001 ± 0.0036	1.0064	563 ⁺⁹⁴ ₋₅₀	>513

^a Equilibrium activity ratio calculated from the analyzed $^{234}\text{U}/^{238}\text{U}$ activity ratio and setting time at infinity in the age equation.

^b Minimum age calculated by subtracting the error limit from the measured $^{230}\text{Th}/^{234}\text{U}$ activity ratio.

5. Results

Analytical results indicate optimum conditions for closed system-behavior of the SRA-3 speleothem (Table 1). Uranium contents are relatively high ranging from 0.5 to 0.75 ppm. $^{234}\text{U}/^{238}\text{U}$ activity ratios cluster tightly around 1.015 and $^{230}\text{Th}/^{232}\text{Th}$ ratios range from 87 to 730. The $^{230}\text{Th}/^{232}\text{Th}$ ratios are all well in excess of 20, the limiting value below which extraneous (detrital) Th significantly affects the date (Bischoff and Fitzpatrick, 1991). $^{230}\text{Th}/^{234}\text{U}$ ratios, ranging from 1.001 to 1.004 are all very close to their respective equilibrium values of 1.0058 to 1.0070. However, because of the high precision of the analyses, averaging about 0.35%, finite dates are calculated with remarkable and unprecedented in-group reproducibility so very close to equilibrium. Because the samples plot close to the asymptotic end of the $^{230}\text{Th}/^{234}\text{U}$ decay curve, very small analytical differences result in very large age differences and a large range of error estimates. All the samples yielded finite dates ranging from 563 to 668 kyr (average 600 kyr). Two of the samples, at -5 cm and at 10.5 cm, had upper limits of equilibrium (infinite age). All the others had finite ages for their upper limit. A conservative approach is to consider the lower limit ages as the minimum age of the speleothem, a range from 513 to 571 kyr (average 530 kyr). We show in Table 2 the analytical results of our analyses of solutions of equilibrium standards. The results show that we obtain equilibrium values within analytical error. Therefore, in as-much as the composition of the SRA-3 samples are very close to equilibrium, they are finite, and we deem the calculated dates to be real because of internal

consistency and reproducibility, and because of our results on the equilibrium standards. Results of the two groups are shown on an evolution diagram (Ludwig, 2000) in Fig. 7.

6. Limits on maximum age and biostratigraphic correlations

How does this older age assignment square with other indications of relative chronology? This minimum age is consistent with the normal magnetization of the fossiliferous muds of the SH, giving a maximum lower limit of 780 kyr, the base of the Brunhes Normal Chron (Parés et al., 2000). The SH rodent fauna lacks the Lower Pleistocene vole *Mimomys savini*, present in Gran Dolina levels TD3 to TD8 and it also lacks its successor *Arvicola cantianus*, which is also absent in Gran Dolina (Cuenca-Bescós et al., 1997; Cuenca-Bescós et al., 1999). Therefore, the absence of these two voles in the SH assemblage allows no age estimate. Neither does the presence of wide-ranging rodent species *Allocelestus*, *Pliomys*, and *Apodemus*. A new and rare discovery in the SH, however, of two molars of the rodent *Clethrionomys acrorhiza* does permit a chronological inference. Maul et al. (1998) have shown that some teeth parameter of members of this genus changed rather continuously from the Lower Pleistocene through the present. If this same pattern of change is followed by *C. acrorhiza* from the SH site, they would correspond to about Marine Isotope Stage (MIS) 15/16 (500–600 kyr).

Table 2

U-series analyses by ICP-MS of two solutions deemed to be in secular equilibrium. SM refers to a solution of the Schwartzwalder Mine secular-equilibrium standard (Ludwig et al., 1985). CP refers to a solution of the Congo Pitchblende standard of unknown origin

Sample	USGS Lab	U (ppm)	$^{234}\text{U}/^{238}\text{U}$	$^{230}\text{Th}/^{234}\text{U}$	$^{230}\text{Th}/^{234}\text{U}$ equilibrium ^a	Date (kyr)
SM	06-16	6.31 ± 0.08	1.000 ± 0.0035	0.999 ± 0.0038	1.002	707 ^{+∞} ₋₁₃₇
SM	06-18	6.32 ± 0.08	0.9987 ± 0.0032	0.999 ± 0.0036	0.999	Equilibrium
SM ^b			0.99882 ± 0.0033		0.999	Equilibrium
CP	06-19	0.645 ± 0.008	0.999 ± 0.0033	1.002 ± 0.0038	1.000	Equilibrium
CP	06-20	0.646 ± 0.008	0.999 ± 0.0035	1.000 ± 0.0042	0.999	Equilibrium
CP	06-21	0.645 ± 0.008	1.006 ± 0.0042	1.006 ± 0.0042	1.000	Equilibrium

^a Equilibrium activity ratio calculated from the analyzed $^{234}\text{U}/^{238}\text{U}$ activity ratio and setting time at infinity in the age equation.

^b $^{234}\text{U}/^{238}\text{U}$ mean ratio from ~40 analyses by the Berkeley Geochron Laboratory (Warren Sharp, personal communication). The Berkeley Geochron Laboratory uses SM as their primary standard so an equilibrium value for $^{230}\text{Th}/^{234}\text{U}$ is assumed.

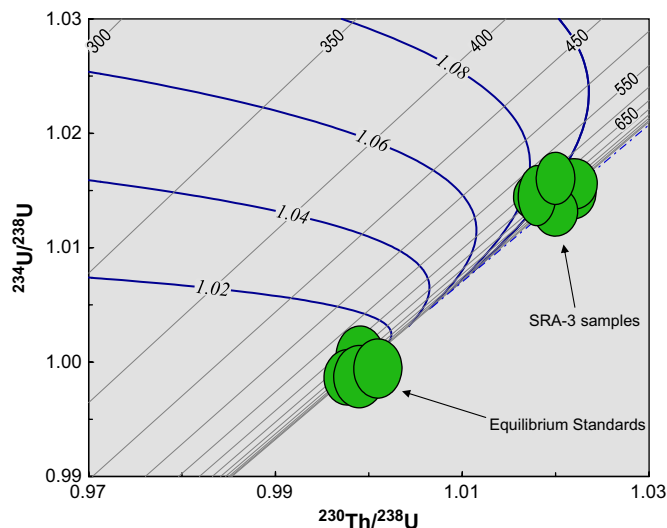


Fig. 7. Evolution diagram (Ludwig, 2000) for U-series isotopic analyses of six SRA-3 speleothem samples and replicates of two equilibrium standards. Scale is greatly expanded close to equilibrium line. Ellipses represent 1 sigma error. Curved lines show evolution of initial $^{234}\text{U}/^{238}\text{U}$ activity as labeled. Inclined straight lines represent isochrons labeled in kyr. SRA-3 samples cluster tightly together, and differ significantly from the equilibrium standards.

Among the larger fauna, *U. deningeri*, the Middle Pleistocene cave bear, is predominantly represented at the site. Although the transition to its descendent, *Ursus spelaeus*, is believed to have occurred toward the end of the Middle Pleistocene, its precise chronology is not yet established (García et al., 1997). However, the SH population of *U. deningeri* does not show any of what might be considered to represent transitional characters, but, rather exhibits a number of primitive features in the dentition and metapodials. Thus, the SH population resembles more closely earlier members of this lineage (García, 2003), such as specimens from Mauer, Mosbach or Arago (MIS 12–16) which approximately date to between 450 and 600 kya based on faunal and radiometric criteria (Faluéres et al., 2004; Koenigswald and Tobien, 1987; Koenigswald, 1992). Remains of *Panthera leo fossilis* have also been recovered from SH (García et al., 1997; Morales et al., 1987). This large-sized lion is found at the site of Isernia la Pineta, one of the few sites with Calibrated Local Faunas, where it has been dated by Ar–Ar at 605 kyr (Coltorti et al., 2005). The ratio of the length and breadth of the M_1 in the SH specimens coincides with that found to characterize this subspecies from the sites of Mauer which correspond approximately to MIS 11–16; 400–600 kyr (Koenigswald and Tobien, 1987; Koenigswald, 1992).

A fragmentary lower first molar attributed to *Canis* sp. was recovered from the SH. The metric dimensions of this tooth fall within the range of the recently published large male sample of *Canis mosbachensis* from the Untermassfeld late Early Pleistocene site (Sotnikova, 2001) (Jaramillo Chron) as well as later *Canis* species (García, 2003).

This compatibility between the new age estimates determined for the SRA-3 speleothem and the biostratigraphic indicators from the site provides strong evidence that the

radiometric results should be considered conclusive for a minimum age of 530 kyr (MIS 14) for the SH sediments. In addition, the presence of several skeletal parts representing the same individual (Individual 6) in different sectors of the Sima complex, including below the SRA-3 speleothem, suggests that the age of the speleothem can be applied to the entire hominid sample from the site.

7. Evolutionary implications

The SH humans are evolutionarily ancestral to Neandertals and show a combination of clear Neandertal-derived features, others which appear in a more incipient fashion and primitive traits commonly found in earlier members of the genus *Homo* (Arsuaga et al., 1991; Arsuaga et al., 1993; Arsuaga et al., 1997a,b,c). Genetic estimates for the timing of the split between the Neandertal and modern human evolutionary lineages have provided a wide range of ages, falling between approximately 300–800 kyr (Ho et al., 2005; Ingman et al., 2000; Krings et al., 1997, 1999; Ovchinnikov et al., 2000). The new dating of the SH site implies that the SH hominids are near the beginning of the Neandertal evolutionary lineage.

Given this early temporal placement of the SH hominids, it might be expected that specimens which postdate the SH population should show more derived Neandertal features. Nevertheless, the Neandertal traits of the SH hominids are somewhat more pronounced than in either the Mauer mandible or the cranial and mandibular specimens from Arago. It should be pointed out that the age of the Mauer site is based on biostratigraphic indicators which provide only an approximate estimation and could be roughly contemporaneous with the new age estimate for SH. In contrast, the cranial and mandibular remains from Arago have been radiometrically dated to >350 kyr (Faluéres et al., 2004). However, the dating of this latter site, U-series dating of a stalagmitic formation which covers the sediments where the cranial and mandibular remains were recovered, and whose most ancient portion shows internal isotopic equilibrium, is also a minimum age, and represents a similar situation to that which characterized the dating of the SH site before the present results. Thus, the potential for TIMS to provide accurate age estimates for other Middle Pleistocene sites in Europe which have been considered to be beyond the limits of U-series analysis may help to refine our understanding of the evolutionary process, which gave rise to the Neandertals. In Fig. 8 we show a summary diagram of what we consider the relative chronological positions of important European faunal and hominid sites.

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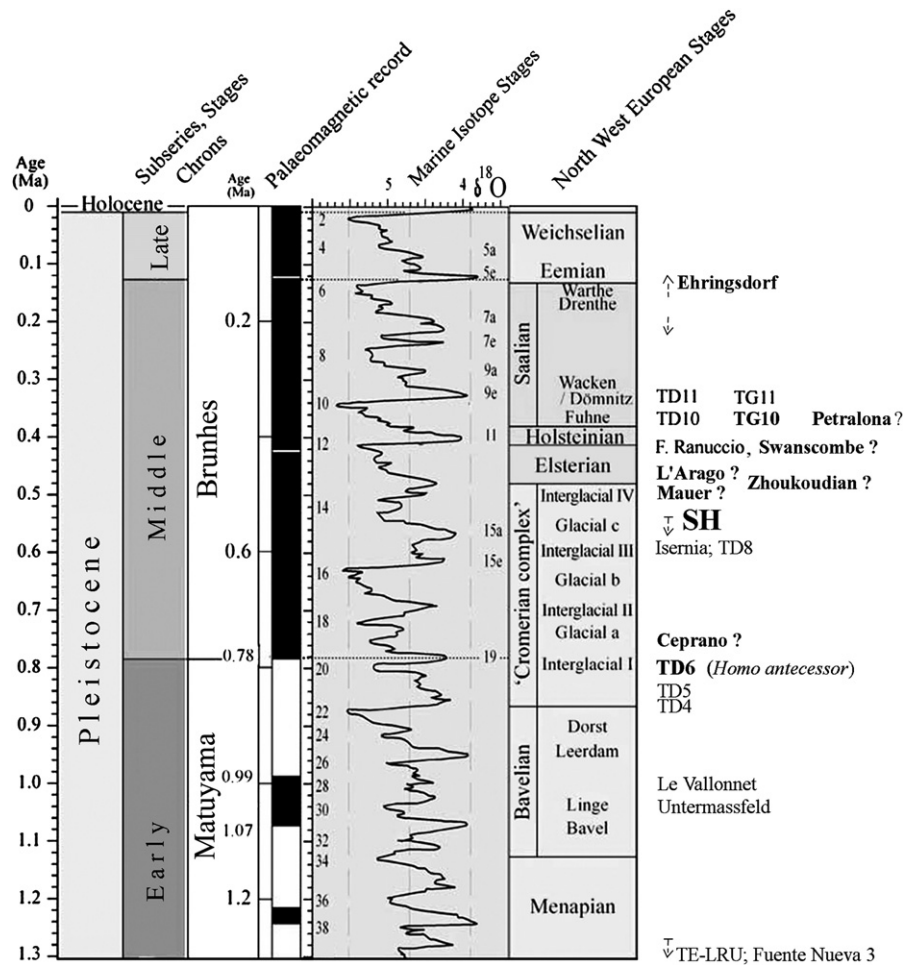


Fig. 8. Summary diagram showing relative chronological positions of important European faunal and hominid (bold) sites. Chronological positions estimated from paleomagnetism, ESR, U-series, K–Ar, Ar–Ar dating and paleontological (micro- and macromammals) biostratigraphy. Sources: Geomagnetic polarity timescale (Cande and Kent, 1995), marine $\delta^{18}\text{O}$ isotope record (Imbrie et al., 1984), NW Europe glacial/interglacial stages (Gibbard and Kolfshoten, 2004), TE Trinchera Elefante, Atapuerca-LRU considered to date off our diagram at 1.5–1.25 myr (Cuenca Bescós and Rofes Chávez, 2004; Rosas et al., 2004), Fuente Nueva 3 (Agustí et al., 2004), Untermassfeld (Kahlke, 2001), Le Vallonnet (d’Lumley et al., 1988), Gran Dolina TD11, TD10, TD8 and TD6 (Falgúeres et al., 1999), Ceprano and Isernia la Pineta (Coltorti et al., 2005; Manzi et al., 2001), Mauer (Koenigswald, 1992) and L’Arago (Falgúeres et al., 2004), Swanscombe (Stringer and Hublin, 1999), Petralona and TG10-TG11 [Falgúeres personal communication, 2006], and Ehringsdorf (Kahlke, 1995).

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