Ancient DNA, Strontium isotopes, and osteological analyses shed light on social and kinship organization of the Later Stone Age

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In 2005 four outstanding multiple burials were discovered near Eulau, Germany. The 4,600-year-old graves contained groups of adults and children buried facing each other. Skeletal and artifactual evidence and the simultaneous interment of the individuals suggest the supposed families fell victim to a violent event. In a multidisciplinary approach, archaeological, anthropological, geochemical (radiogenic isotopes), and molecular genetic (ancient DNA) methods were applied to these unique burials. Using autosomal, mitochondrial, and Y-chromosomal markers, we identified genetic kinship among the individuals. A direct child-parent relationship was detected in one burial, providing the oldest molecular genetic evidence of a nuclear family. Strontium isotope analyses point to different origins for males and children versus females. By this approach, we gain insight into a Late Stone Age society, which appears to have been exogamous and patrilocal, and in which genetic kinship seems to be a focal point of social organization.

burial rites | Neolithic | violence | Corded Ware Culture | exogamy

Toward the end of the Central European Neolithic, in the third millennium B.C., the cultural complex of the Corded Ware Culture (CWC) forms one of the major archaeological entities in Europe (Fig. 1) (1). A characteristic and eponymous feature is the decoration of pots by impressions of a twisted cord. Varying groups can be recognized between the Moskva River and the Swiss lakes, between Southern Scandinavia and the Carpathians (2). Graves and grave goods are the main source of information, whereas settlement structures are rarely encountered (3). A large number of graves, sometimes with burial mounds, indicate a high population density in the area of the rivers Elbe and Saale during this period (4). The CWC elements of the Mittelelbe-Saale group (our region of study) are estimated to occur at 2700 cal B.C. using radiocarbon dates, and the end can be fixed at about 2200 to 2000 cal B.C. (5). In the latest phases, we observe a temporal overlap with the Bell Beaker Culture and the Unetice Culture.

The skeletal remains of our ancestors, their grave goods, and mortuary practices are valuable sources of biological, cultural, and socio-historical information. The detailed analysis of burial grounds, one of the major sources of information, is where most scientific disciplines concerned with prehistory meet and interact best. During the CWC, the dead are placed in a flexed position, with men's heads oriented to the west, while the women's heads are to the east (4). As a consequence, males are resting on their right sides, females on their left, with both sexes facing south. Exceptions to this rule are rare in the CWC. Compared to other Neolithic cultures, double and multiple burials are also common in CWC contexts radiocarbon dated to 2700 to 2400 (2200) B.C. In many cases, however, such multiple interments are because of secondary burials; that is, they represent multiple-phase collective burials rather than single inhumations (6).

Results and Discussion

Archaeological and Anthropological Features. In 2005, excavations at Eulau (Saxony-Anhalt, Germany), close to the discovery site of the world famous Nebra sky disk (7), revealed four closely grouped and well-preserved multiple burials [Fig. 2 and 3, supporting information (SI) Figs. S1 and S2]. The burials were discovered on a tertiary terrace with gravel sediments covered by loess directly above the river Saale. Ring ditches of approximately 6 meters in diameter surrounded three of the four graves (Fig. S3). It is assumed that most of the graves originally possessed small mounds, which are unrecognizable today because of centuries of agricultural activity. Grave goods accompanying the Eulau individuals are somewhat unspectacular: men and boys were given stone axes; women and girls received flint tools or animal tooth pendants. In addition, butchered animal bones give evidence of at least one food offering per grave. The burials are free from later disturbances and comprise the remains of 13 human individuals as part of a larger cemetery. The burial custom, associated artifacts, and radiocarbon dating allow these graves to be attributed to the Central European CWC (Table 1).

Two of the graves contained four individuals: grave 99 contained a female (35–50 years), a male (40–60 years), and two children of 4 to 5 and 8 to 9 years; grave 98 contained a female (30–38 years) and three children of 0.5 to 1, 4 to 5, and 7 to 9 years of age. Grave 93 held three bodies: a male (25–40 years) and two children of 4 to 5 and 5 to 6 years of age. Grave 90 contained the remains of two people: a female of 25 to 35 years and a child of 4 to 5 years of age at death. Intriguingly, the arrangement of the dead seems to mirror their relations in life. The latter is reflected by the face-to-face arrangement of several pairs of individuals and the positioning of their arms and hands, which are interlinked in several cases (see Fig. 2). Single-phase grave pits were recognized for each multiple burial, attesting to the simultaneity of the internments (in contrast to other Neo-

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Data deposition. The sequences in this paper have been deposited in the Genbank database (accession nos. FJ424615–FJ424625).

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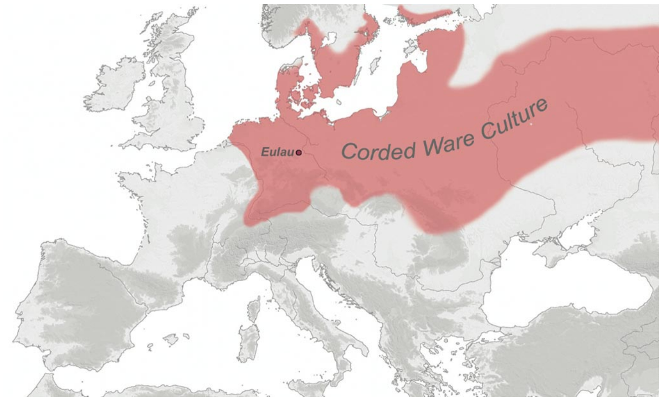

Fig. 1. Geographical distribution of European CWC groups and location of the Eulau site.

lithic multiple burials). Given the over-all similarity of the four burials, their proximity, and the statistically indistinguishable radiocarbon dates for each, we assume that all four multiple burials were laid out contemporaneously.

Morphological aging and sexing according to modern standards (8) revealed all burials to contain children ranging from newborns up to 10 years of age, and adults of ≈30 years or older (see *SI Text*). Interestingly, there are no adolescents or young adults. Another striking aspect of the Eulau burials is the prevalence of interpersonal violence observable in five of the individuals. The most prominent example features a stone projectile point embedded in a vertebra of individual 5 (grave 90) (Fig. 4) and skull fractures in individual 7 (grave 98) (Fig. S4) and individual 4 (grave 99, not shown). Among the observed traumatic lesions are defense injuries, such as those of the forearm and the metacarpals on males from grave 93 and 99, whereas others represent lethal *perimortem* cranial and postcranial injuries. The occurence of both types is a strong indicator of lethal aggression (9, 10).

Overall, the injury patterns point to a violent event, which

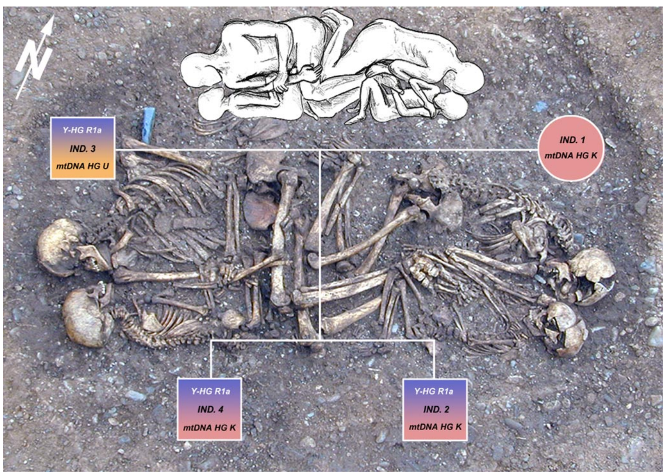

Fig. 2. The genetic relationship of individuals from grave 99. An *in situ* photograph is overlain by a pedigree reconstructed from the genetic results with squares denoting male, circles female, and triangles indifferent individuals. Different genetic lineages are illustrated by color fillings of the symbols with the mitochondrial (maternal) lineages highlighted in red and orange and the Y-chromosomal (paternal) lineage shown in blue (HG, Haplogroup; Ind., individual). When no genetic results could be received, the symbol is shown in gray. A reconstruction of the situation at death was added on top to emphasize the ''intertwining'' and the orientation of the bodies.

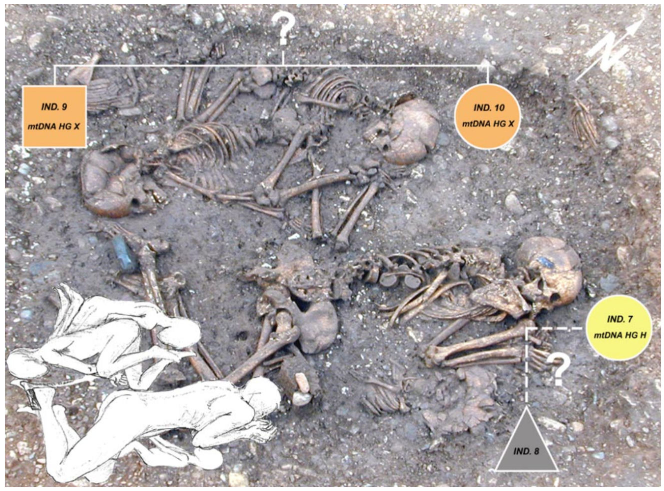

Fig. 3. The genetic relationship of individuals from grave 98. Annotations as in Fig. 2.

most probably resulted in the death of all 13 individuals. The most plausible interpretation is that the graves are the result of a violent raid, with 85% of the dead being subadults and women, and the survivors returning to bury the dead. The great care seen in the treatment of the dead (11) seems to support this scenario. Whereas a demographic profile might be the result of chance, in other examples of suggested raids, in particular the cases of the c. 4900 cal B.C. Linearbandkeramik mass grave of Talheim, Germany (12) and Asparn Schletz, Austria (13), the victims lack any signs of careful mortuary treatment.

Ancient DNA. Analysis of ancient DNA was undertaken to understand the genetic affiliation of the group members of these four multiple burials and to confirm conventional sexing (see *SI Text*) (14, 15). Although ancient DNA analyses ostensibly seem to be the method of choice to answer questions of kinship on a molecular level, so far only a few studies have been able to address kinship within an archaeological context (16–20).

Here, we applied a hierarchical aDNA approach to analyze maternal lineages (mtDNA), potential genetic kinship by biparental loci (autosomal short tandem repeats or STRs) in 12 out of the 13 individuals, and paternal affiliation by indication via Y-STRs and Y-chromosomal single nucleotide polymorphisms (Y-SNPs).

Because of their exceptional preservation, we were able to retrieve ancient DNA and reproduce unambiguous mtDNA profiles of hypervariable segment I (HVS I) for 9 out of 12 individuals. Differing mtDNA haplotypes across all four multiple burials indicate high mtDNA variability among the individuals, except where they are closely related. No close maternal genetic relationship could be observed across the four multiple burials.

Nuclear DNA could also be retrieved for some individuals, albeit with limited success and completion of STR profiles (Table S1 and S2). However, in less successful cases nuclear typing provided at least genetic sex determination. In more successful cases (grave 99) the profiles of autosomal STR and Y-STR results—although fragmentary—affirmatively support the interpretation of a nuclear family, which is identified by mtDNA and Y-SNP results. Thus, the combination of all molecular results allowed us to reconstruct the genetic relationships among the individuals in graves 98 and 99, whereas this was not possible in the less well-preserved graves 90 and 93.

One burial (grave 99) (see Fig. 2), consisting of male and female adult individuals and two children facing each other in

Table 1. Summary of archaeological, anthropological, molecular genetic and radiocarbon data

Burial	Ind.	Preservation	Sample	Age at death	Grave goods/ Dressing	Flexed position	Orientation	Sex Arch.	Sex Morph.	Sex Mol.	Traces of violence	Radiocarbon age*
99	1	Excellent	A: tooth 48 B: femur C: tooth 35 D: tibia	35–50	Two silex blades	Leftside	East-West	Female	Female	Female	n.d.	BP 4073 ± 27 (KIA27850) 2632–2570 (51.2%) cal B.C.
	2	Excellent	A: tooth 64 B: femur C: tooth 75	4–5	—	Rightside	Towards 1	Female	?	Male	n.d.	—
	3	Excellent	A: tooth 37 B: femur C: humerus	40–60	One axe blade, one bone prickle/spur	Rightside	West-East	Male	Male	Male	Yes	BP 4074 ± 24 (KIA27849) 2631–2571 (55.1%) cal B.C.
	4	Excellent	A: tooth 55 B: femur C: tooth 36	8–9	—	Leftside	Towards 3	Male	?	Male	Yes	—
90	5	Good	A: tooth 17 B: tooth 16	25–35	One animal tooth pendant	Leftside	East-West	Female	Female	Female	Yes	BP 3969 ± 29 (KIA27878) 2563–2465 (68.2%) cal B.C.
	6	Very bad	A: tooth 75 B: -	4–5	—	Rightside	Towards 5	Female	?	?	n.d.	—
98	7	Excellent	A: tooth 17 B: tooth 16	30–38	One silex blade	Leftside	East-West	Female	Female	Female	Yes	BP 4049 ± 26 (KIA27851) 2619–2494 (68.2%) cal B.C.
	8	Bad	A: - B: -	0.5–1	—	Rightside	Towards 7	Female	?	n.a.	n.d.	—
	9	Good	A: tooth 74 B: tooth 36	7–9	One axe blade, one silex flake, one pricker	Rightside	West-East	Male	?	Male	n.d.	BP 4053 ± 27 (KIA27852) 2620–2495 (68.2%) cal B.C.
	10	Good	A: tooth 85 B: tooth 65	4–5	—	Leftside	East-West	Female	?	Female	n.d.	—
93	11	Medium	A: tooth 28 B: tooth 36	25–40	One axe blade, one pricker	Rightside	West-East	Male	Male	Male	Yes	BP 4101 ± 27 (KIA27879) 2675–2578 (53.8%) cal B.C.
	12	Very bad	A: tooth 75 B: tooth 65	4,5–5,5	—	Rightside	West-East	Male	?	?	n.d.	—
	13	Very bad	A: tooth 84 B: tooth 64	4–5	—	Leftside?	West-East	?	?	?	n.d.	—

*Sigma values are given in parentheses; ABCD, samples taken for independent DNA extractions; Ind., individuals; n.a., not analyzed; n.d., not detectable; ?, unknown; -, none.

pairs, was shown to contain a nuclear family. The woman (ind. 1) and both children (inds. 2 and 4) share the same mtDNA haplogroup K1b. Additionally, the Y chromosome haplogroup R1a of the boys corresponds with the man's (ind. 3). It is noteworthy that in grave 99 the orientation of the adult individuals follows the established pattern of the CWC, whereas both children clearly deviate from it. It appears that the burial orientation pattern was overruled for each boy to face a parent to express a biological relationship.

The second successful reconstruction (grave 98) (see Fig. 3) revealed that two of the three younger individuals (inds. 9 and 10) were most likely siblings or, according to their mtDNA (haplogroup X2), were at least maternally related. However, the DNA data excluded the adult female (ind. 7, haplogroup H) as their putative mother. This is reflected in the arrangement of the individuals (following the regular burial pattern) because the two children were not facing the adult woman. Although the genetic results rule out any close maternal relationship, they either were buried with just another adult victim or, more likely, were paternally (aunt) or socially (step-mother) related to the woman. Interestingly, the male child in Grave 98 (ind. 9) takes the social position of a male adult, attested by a stone axe as a grave good. This indicates some aspects of rites of passage and might also hint at the importance of social relations inside a household.

So far, archaeological sites with similar findings have most

Fig. 4. A flint arrowhead embedded in a lumbar vertebra of individual 5 (grave 90). (Scale bar, 2 cm.)

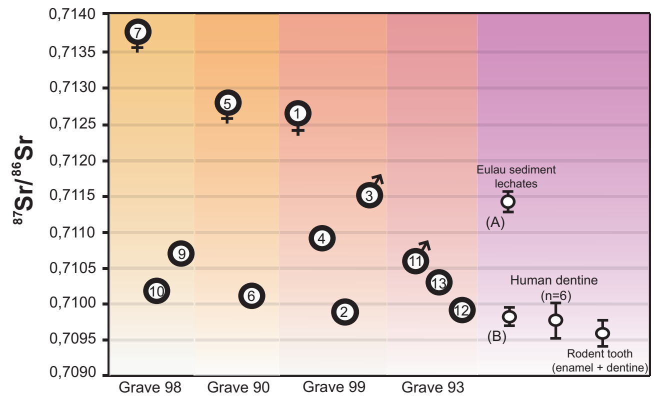

Fig. 5. Strontium isotope results showing the females (female symbol), males (male symbol) and children (circle) by grave. Numbers in symbols correspond to individuals in Table 1. The children's $^{87}Sr/^{86}Sr$ forms a discrete group around 0.7105, whereas the females $^{87}Sr/^{86}Sr$ are all >0.7125, indicating the females spent their childhood at a different geological region to the children. Typical errors are ± 0.00001. Water-leached sediments from immediately above the level of the grave cuts (denoted by "A") and from the layer into which the graves were cut (denoted by "B") provide an estimate of the local range of Sr isotopes. Sr isotopes in human dentine and a whole rodent will be dominated by post depositional Sr uptake, and reflect the Sr isotopes of the immediate burial environment, hence their similarity to layer B.

often led to an *a priori* assumption of close genetic kinship among two or more individuals within one grave. Indeed, our image of past family life and structure is still dominated by 19th century ideals (21) and reconstructions of prehistoric families and social units even now adhere to these orthodox schemes (22). The almost complete lack of archaeological evidence for the structure of basic social units has contributed to the persistence of these ideals, but here we have shown that basic social organization can be revealed from the study of ancient biomolecules. In Eulau, we have established the presence of the classic nuclear family in a prehistoric context in Central Europe, to our knowledge the oldest authentic molecular genetic evidence so far. Their unity in death suggests a unity in life. However, this does not establish the elemental family to be a universal model or the most ancient institution of human communities. For example, polygamous unions are prevalent in ethnographic data and models of household communities have apparently been involving a high degree of complexity from their origins (23, 24).

Strontium Isotopes. Modern geochemical fingerprinting by Strontium and other isotope analyses can yield conclusive data on areas of origin, mobility, and migration, as well as traditions of residence and exogamy of the members of ancient communities. Strontium (Sr) isotopes in tooth enamel reflect dietary strontium derived from soils during childhood and will vary between individuals from distinct geological regions. They can be used to infer the childhood geological location of an individual and hence identify subsequent movement, which may be related to subsistence behavior, traditions of exogamy, or mass migrations (25–27).

The $^{87}Sr/^{86}Sr$ ratio of the children's teeth from Eulau form a coherent group centred on 0.7103 and with a small overall variation (Fig. 5, Table S3). This is in marked contrast with the females' isotopic ratios, which are all greater than 0.7125. It is not clear whether the two males analyzed form a group. Certainly one has an $^{87}Sr/^{86}Sr$ value similar to the children; the other is intermediate between the children and females. The homogeneity of the children suggests the burials represent a single community or a group of communities from a single location. Sr isotope ratios were measured on MilliQ water lechates of sediments representing the layer immediately above the grave cuts (see Fig. 5, layer A) and from the layer into which the graves were cut (layer B) as a proxy for the local range of isotopes. In the absence of significant numbers of faunal teeth, which are often used to define the local strontium isotopic range (28), we also measured Sr isotopes in the dentine of the human teeth and in a whole (dentine and enamel) rodent tooth from the site. It has been proposed that the Sr isotopes in dentine will be dominated by post depositional strontium uptake (29) and represent the isotope ratio of the immediate burial environment. The similarity of the Eulau dentine values with the isotopes from sediment (layer B), and their very narrow range, suggests this to be the case. The children and males' Sr isotopes all fall between those of the two sediment samples, suggesting they are all local. There is variation in Sr isotopes down the sediment column, which is expected where there is differential weathering of mineral components (30), so the local signal is broad. But the females clearly fall outside this range, and must have spent their early lives elsewhere. This indicates the practice of exogamy and patrilocality; the females moved to the location of the males, where they had their offspring. Without detailed mapping of Sr isotopes over a wide area, it is impossible to say where the females grew up. The closest location that is likely to give similar, more radiogenic, strontium isotopes would be the Palaeozoic deposits in the Harz mountains, some 60 km to the northwest of Eulau, although we cannot rule out an origin much further afield. This is the subject of ongoing investigations.

Our Strontium isotope analyses reveal the men and children are most likely to be "locals" who lived and died in the Eulau region. The females, however, were brought up at a different location from where they raised their children, indicating the practice of exogamy and patrilocality for this late Neolithic community. Up to now, exogamy and patrilocality in prehistory are as well most often assumed *a priori* or inferred from exotic grave goods in female graves. Clear evidence for such practices has so far only been shown for the early phases of the Neolithic in Central Europe (31).

Synthesis of Integrative Studies. This multidisciplinary study has revealed detailed information about the daily lives, the social organization, and the death of a Stone Age community in Central Europe 4,600-years ago. By a combination of methods, we are able to gain a detailed insight into parent-offspring relations (nuclear families) and possibly the association of related generations (extended families), as well as the marriage system (exogamy).

Our study of the Eulau individuals shows that their deaths were sudden and violent, apparent in lesions caused by stone axes and arrows, with evidence of attempts of some of the individuals to defend themselves from blows. The demographic structure of the examined individuals from the four multiple burials does not comprise all age-classes and therefore implies that some of the community survived. Undoubtedly these survivors had intimate knowledge of the victims' family and social relationships and buried them in strict accordance to the cultural specifications of the CWC, even to the extent of indicating the proposed supragenetic social connections of the woman and the two children from grave 98. Our genetic study has revealed the subtleties in these burial rites and the importance (and respect) of nuclear familial relationships in the treatment of the dead. Based on our observations, we propose that biological relationship can indeed be inferred from the subtle positioning of the dead within multiple burials. It seems that physical closeness of individuals implies biological kinship and a face-to-face orientation indicates a parent-offspring relationship. The known spectrum of CWC burial rites can therefore be extended accordingly and may also be applied to graves 90 and 93.

Furthermore, it becomes increasingly evident that biological kinship represents the basis for social relationships expressed in the arrangement of the deceased in the Central European Neolithic. As shown here for the CWC, kin were also buried in close contact to each other in earlier cultures, which interred their dead in larger collective graves (32). Careful placement and orientation of a body in a grave containing more than one individual therefore may indeed be seen as a detailed statement about social ties connecting the persons in life. This clearly intended intra-group expression of relationships may also have wider implications for the interpretation of inter-cultural differences in burial rites, which can explicitly differ between contemporaneous and neighboring archaeological groups, like the CWC and Bell Beaker Culture in our region of study (33, 34). Further multidisciplinary results like these from the Eulau multiple graves may provide us with a fine enough resolution of the permeability of cultural boundaries to trace the pathways of individuals and groups, their origins, and their familial (and social) networks.

The beginning and end of the Neolithic Age are characterized by profound economic, social, and cultural change or crises (35). The Middle Elbe-Saale area in which Eulau is located represents a key region of Central Europe. There are fertile soils, a stable continental climate, and natural traffic routes, as well as rich resources favoring long-range cultural contacts. This rendered the region an area coveted and settled by numerous synchronous and successive archaeological cultures (36). Its occupation was, in all likelihood, frequently accompanied by hostile actions and high population dynamics, providing a ready explanation for the evidence of interpersonal violence, both in Eulau and other locations (37, 38). Our investigations of the Eulau multiple burials reveal that modern archaeological investigations are most productive when they are based on the close transdisciplinary collaboration of archaeology, biological anthropology, molecular genetics, and geochemistry. Analyses of ancient DNA and suitable stable isotopes for investigating mobility and migration represent the proper approach to such research issues, which can reach beyond the statement of a community tragically united in fate. This approach, coupled with archaeological and anthropological data, has allowed the reconstruction of a plausible scenario of a tragedy 4,600-years ago, and closes existing gaps in our knowledge about social and kinship organization in prehistoric times.

Methods

The special nature of the multiple burials from Eulau was apparent immediately upon discovery. Therefore, in preparation of genetic and biochemical analyses, tooth samples were taken from all individuals *in situ* and under DNA free conditions.

Ancient DNA Analysis. We analyzed 12 out of 13 individuals from the four multiple burials from Eulau (Naumburg, Saxony-Anhalt, Germany). From each individual, three samples (A, B, and C) were chosen from well-preserved long bones and teeth of each individual (see Table 1). One child (grave 98, ind. 8) was omitted from the analysis because of its state of preservation unsuitable for aDNA analysis. Samples A were taken directly under DNA free conditions *in situ* during the excavation. Samples B and C were collected DNA-free during early stages of sample preparation at the State Museum of Prehistory, Saxony-Anhalt, Germany, wearing face masks and gloves during sampling. It is of importance to mention that samples B and C were treated with a specific consolidant (''Archaeofix,'' State Office for Heritage Management and Archaeology Saxony-Anhalt), when preserving the finds *en bloc* and afterward with acetone, to unhinge bone parts for further sampling purposes (B and C samples). Despite strict precautions against contamination during and after excavation, all persons having being involved in the investigation of the Eulau individuals have also been typed to monitor potential sources of contamination (Table S4 and Table S5).

The samples were further processed (i.e., DNA extracted, amplified, cloned, sequenced, and analyzed) at the ancient DNA facilities of the Institute of Anthropology, Johannes Gutenberg University Mainz, as described previously (39). Additional or alternative steps to our standard protocol are described here. In addition to extensive UV-treatment, we removed the sample surface by shot blasting with aluminum oxide abrasive (Harnisch & Rieth). Additionally, we amplified a 363-bp fragment (np 00034–00397) of HVS II using four overlapping primers for individuals 9 and 10 to reveal a higher resolution on the subhaplogroup level. Primer sequences and all alignments of sequenced clones are given in Fig. S5 and Table S6.

STR analysis were carried out using the AmpF/STR Profiler Plus kit (Applied Biosystems), comprising nine autosomal STR systems (D3S1358, vWA, FGA, D8S1179, D21S11, D18S51, D5S818, D13S317, and D13S317) and the sex-specific marker amelogenin. Reactions were set up in a volume of 25 μl, containing 10-μl Reactionmix (Applied Biosystems), 5-μl Primermix (Applied Biosystems), 2.5 U AmpliTaq Gold (Applied Biosystems), and 3 to 7 μl of DNA extract. PCR was performed according to the manufacturer's protocol (Applied Biosystems), with the exception that 45 cycles were used instead of 30. For specific paternity affiliation we used the AmpF/STR Yfiler kit (Applied Biosystems) comprising 17 Y-chromosome STR systems (DYS393, DYS456, Y-GATA, DYS458, DYS389 I/II, DYS391, DYS19, DYS437, DYS390, DYS439, DYS438, DYS385 a/b, DYS635, DYS448, DYS392). The AmpF/STR Yfiler PCR was performed in a final reaction volume of 12.5 μl, containing 4.5-μl Reactionmix (Applied Biosystems), 2.5-μl Primermix (Applied Biosystems), 0.5 U AmpliTaq Gold (Applied Biosystems), and 3 to 5 μl of DNA extract, and was amplified according to the manufacturer's instructions (Applied Biosystems), except that 45 cycles were used instead of 30. All fragment analyses were carried out on an Applied Biosystems 3130 Genetic Analyzer (Applied Biosystems) and were analyzed using GeneMapper Software v2.5 (Applied Biosystems). Detailed results of STR analyses are shown in Table S1.

After indication of paternal kinship in individuals 2, 3 and 4 of grave 99 via Y-STRs, we directly amplified the characteristic Y-SNP SRY10831.2 region from all extracts of the three individuals to confirm haplogroup status R1a. PCR conditions were as described (39), except for adding 5 to 7 μl of extract, 50 cycles for amplification of nuclear DNA, and primer annealing at 57 °C. PCR products were cloned and sequenced as described above and all aligned sequences are shown in Fig. S5.

Strontium Isotope Analyses. The enamel surface of an intact molar was first cleaned using a dental burr and hand drill. A wedge of enamel and dentine (*ca.* 0.5-mm wide, 1-mm deep) representing the complete growth axis of the enamel was removed using a flexible diamond-impregnated dental disk. Any dentine adhering to the enamel section was then removed using a dental burr, and the remaining enamel sample cleaned in an ultrasonic bath. The whole enamel section was dissolved in 3-ml 7N HNO_3. Thus, the strontium isotopes represent the mean value of the whole enamel growth period, weighted for variations in enamel thickness, and variations in Sr concentration. Any detritus was removed by centrifuging, and the supernatant was dried and redissolved in 3N HNO_3. An aliquot of this solution was removed, representing 3 mg of solid enamel (containing $\approx$100–300 ng of Sr), and made up to 0.5-ml 3N HNO_3 to be loaded onto ion exchange columns.

The strontium was separated using standard ion exchange chromatography using 70 μl of Eichrom Sr spec resin (50–100 μm). Samples were loaded in to 0.5-ml 3N HNO_3 and washed with 4-ml 3N HNO_3. Strontium was eluted in 1.5-ml MilliQ water. The elutant was dried down and loaded using a few μl 10% HNO_3 onto rhenium filaments preconditioned with 1-μl $TaCl_5$ solution and 1-μl 10% H_3PO_4. Isotope ratios were measured on a ThermoFinnegan Triton Thermal Ionization Mass Spectrometer. The data are corrected for mass fractionation using a $^{86}Sr/^{88}Sr$ value of 0.1194 and an exponential fractionation law. ^{87}Rb is subtracted using the measurement of ^{85}Rb and a ^{85}Rb/^{87}Rb value of 2.59265 (40). Data are corrected to NBS 987 using a value of 0.710248. The typical precision for ^{87}Sr/^{88}Sr achieved for a tooth sample using this method is $\pm$ 0.00001.

Sr isotope results for the teeth from Eulau are provided in Table S3. Molars were sampled in all cases. For the adults, the Sr isotopes represent the weighted average of dietary strontium from around 2.5 to 7.5 years for M2 and 9 to 14 years for M3. For the children, the first dentition begins to form *in utero* and is complete in the first year after birth.

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1. Mallory JP (1997) in *Encyclopedia of Indo-European Culture* (Fitzroy Dearborn), pp 127–128.
2. Buchvaldek B, Strahm C eds. (1992). *Die kontinentaleuropäischen Gruppen der Kultur mit Schnurkeramik. Schnurkeramik-Symposium* 1990 (Univerzita Karlova, Prague).
3. Gross E, *et al.* eds. (1987). *Zürich ''Mozartstrasse.'' Neolithische und bronzezeitliche Ufersiedlungen 1*. Ber. ZD Monogr. 4 (Zürich).
4. Fischer U (1956) *Die Gräber der Steinzeit im Saalegebiet* (de Gruyter, Bonn), pp 120–121.
5. Furholt M (2003) *Die absolutchronologische Datierung der Schnurkeramik in Mitteleuropa und Südskandinavien*, (Habelt, Bonn).
6. Dresely V (2004) *Schnurkeramik und Schnurkeramiker im Taubertal*, (Theiss, Stuttgart).
7. Meller H, Garrett K (2004) Star search: Relic thieves, a 3600-year-old disk of the heavens, and an intrepid archaeologist add up to a real-life thriller. *National Geographic* 205.
8. White TD, Folkens PA (2005) *The human bone manual*, (Academic, Amsterdam).
9. Lovell N (1997) Trauma analysis in paleopathology. *Yearb Phys Anthrop* 40:139–170.
10. Rogers J (2004) in *Violent Interactions in the Mesolithic*, ed Roksandic M (Archaeopress, Oxford), pp 9–21.
11. Komar D (2008) Patterns of mortuary practice associated with genocide: Implications for archaeological research. *Curr Anthrop* 49:123–133.
12. Wahl J, König HG (1987) Anthropological-traumatological examinations of human skeletal remains from the Bandkeramik mass grave at Talheim, Kreis Heilbronn. *Fundberichte aus Baden-Württemberg* 12:65–186.
13. Teschler-Nicola M, *et al.* (1999) Evidence of genocide 7000 BP. Neolithic paradigm and geo-climatic reality. *Coll Antropol* 23:437–450.
14. Handt O, Krings M, Ward RH, Pääbo S (1996) The retrieval of ancient human DNA sequences. *Am J Hum Genet* 5:368–376.
15. Kaestle FA, Horsburgh KA (2002) Ancient DNA in anthropology: Methods, applications and ethics. *Yearb Phys Anthrop* 45:92–130.
16. Stone AC, Stoneking M (1998) mtDNA analysis of a prehistoric Oneota population: implications for the peopling of the New World. *Am J Hum Genet* 62:1153–1170.
17. Keyser-Tracqui C, Crubezy E, Ludes B (2003) Nuclear and mitochondrial DNA analysis of a 2,000-year-old necropolis in the Egyin Gol Valley of Mongolia. *Am J Hum Genet* 73:247–260.
18. Schultes T, Hummel S, Herrmann B (2000) Ancient DNA-typing approaches for the determination of kinship in a disturbed collective burial site. *Anthropol Anz* 58:37–44.
19. Dissing J, *et al.* (2007) The last Viking King: A royal maternity case solved by ancient DNA analysis. *Forensic Sci Int* 166:21–27.
20. Alt KW, Vach W, Wahl J (1995) Kinship analysis of skeletal remains from the Bandkeramik mass grave at Talheim, Kreis Heilbronn. *Fundberichte aus Baden-Württemberg* 20:195–217.
21. Nelson C (2007) *Family ties in Victorian England*, (Praeger Publishers, Westport).
22. Strouhal E (1990) *Nubica* I/II:611–629.
23. Murdock GP (1967) *Ethnographic Atlas: A Summary* (Pittsburgh Univ Press, Pittsburgh).
24. Hodder I (1984) in *Ideology, Power, and Prehistory*, eds Miller D, Tilley C (Cambridge Univ Press, Cambridge), pp 51–67.
25. Bentley RA (2006) Strontium isotopes from the earth to the archaeological skeleton: a review. *Archaeol Method Theory* 13:135–187.
26. Price TD, Wahl J, Bentley RA (2006) Isotopic evidence for mobility and group organization among Neolithic farmers at Talheim, Germany, 5000BC. *European Journal of Archaeology* 9:259–284.
27. Bentley RA, Wahl J, Price TD, Atkinson TC (2008) Isotopic signatures and hereditary traits: snapshot of a Neolithic community in Germany. *Antiquity* 82:290–304.
28. Bentley RA, Price TD, Stephan E (2004) Determining the 'local' 87Sr/86Sr range for archaeological skeletons: a case study from Neolithic Europe. *Journal of Archaeological Science* 21:365–375.
29. Budd P, Montgomery J, Barreiro B, Thomas RD (2000) Differential diagenesis of strontium in archaeological human dental tissues. *Appl Geochem* 15:687–694.
30. Blum JD (1997) in *Tectonic Uplift and Climate Change*, ed Ruddiman WF (Plenum Press, London), pp 259–288.
31. Bentley RA, *et al.* (2002) Prehistoric migration in Europe: Strontium isotope analysis of Early Neolithic skeletons. *Curr Anthropol* 43:799–804.
32. Meyer C, *et al.* (2008) The Human Skeletal Remains from the Neolithic Burial Chamber of Benzingerode. Anthropological Analyses of the Inhumations in a Collective Grave of the Bernburg Culture. *Archäologie in Sachsen-Anhalt Sonderband* 7:107–151.
33. Heyd V (2007) Families, treasures, warriors, and complex societies: Beaker groups and the third millennium BC along the Upper and Middle Danube. *Proceedings of the Prehistoric Society* 73:321–370.
34. Harrison R, Heyd V (2007) The transformation of Europe in the third millenium BC: The example of ''Le Petit Chasseur I + III'' (Sion, Valais, Switzerland). *Prähistorische Zeitschrift* 82:129–214.
35. Hayden B (1993) *Archaeology: The Science of Once and Future Things*, (W.H. Freeman, New York).
36. Beier H-J, Einicke R (1994) The Neolithic in the regions Mittelelbe-Saale and Altmark. *Beiträge zur Ur- und Frühgeschichte Mitteleuropas* 4:243–255.
37. Christensen J (2004) Warfare in the European Neolithic. *Acta Archaeologica* 75:129–156.
38. Walker PL (2001) A Bioarchaeological perspective on the history of violence. *Annu Rev Anthropol* 30:573–596.
39. Haak W, *et al.* (2005) Ancient DNA from the first European farmers in 7500-year-old Neolithic sites. *Science* 310:1016–1018.
40. Steiger RH, Jaeger E (1977) Subcommission on geochronology: Convention on the use of decay constants in geo- and cosmochronology. *Earth Planet Sci Lett* 36:359–362.