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ANTHROPOLOGY

A ritual murder shaped the Early and Middle Neolithic across Central and Southern Europe

Bertrand Ludes^{1†}, Ameline Alcouffe^{2†}, Irina Tupikova³, Patrice Gérard², Yaramila Tchérémissinoff^{4,5}, Alexandre Ribéron⁶, Jean Guilaïne⁷, Alain Beeching^{8‡}, Eric Crubézy^{2,9*‡}

In the Rhône Valley's Middle Neolithic gathering site of Saint-Paul-Trois-Châteaux (France), the positioning of two females within a structure aligned with the solstices is atypical. Their placement (back and prone) under the overhang of a silo in front of a third in a central position suggests a ritualized form of homicidal ligature strangulation. The first occurrence dates back to the Mesolithic, and it is from the Early Neolithic of Central Europe that the practice expands, becoming a sacrificial rite associated with an agricultural context in the Middle Neolithic. Examining 20 cases from 14 sites spanning nearly two millennia from Eastern Europe to Catalonia reveals the evolution of this ritual murder practice.

INTRODUCTION

The topic of human sacrifice, its significance in understanding human societies, and its archaeological study are subjects of active debate and interest (1–9). The debate around this issue has sparked lively discussions across various fields, including the humanities, as well as social and ecological sciences (10, 11). In the context of the Neolithic period in Europe, scholars have been particularly intrigued by the concept of human sacrifice (12, 13). The prevailing archaeological interpretation of human sacrifice during this era, influenced by the social control hypothesis, often sees it as a form of retainer sacrifice, where officiants killed enslaved people, servants, relatives, wives, concubines, or others to accompany their masters, social superiors, or relatives into the afterlife. (14–19). An alternative viewpoint suggests that human sacrifice might have played a role in ideological integration within agrarian societies (20) rather than being solely a feature of hierarchical societies (11). Moreover, there has long been suspicion of agricultural rituals predominantly involving female participants during the European Neolithic (21). One of the earliest signs of agrarian rites could be in the ritual destruction of grindstones—a symbol of agriculture and harvest—which is a tradition that may have been especially widespread in the Mediterranean region (22); in other sites, the remains of fauna are notable, with notable sacrifices of dogs and bovinds (23). We should note that human sacrifice to obtain abundant harvests (20) is not an exceptional occurrence in farming societies and is particularly well documented for specific historical periods (24). In some well-documented cases, the breakdown by sex and age of the individuals sacrificed shows that, depending on the case, they could have been children, young women, or even adults (25). In Europe, particularly in Central Europe,

there were still abundant traces of such sacrifices (especially of young women) in folklore in the 19th century, and this was the source of one of the most famous writings by J. G. Frazer, one of the fathers of religious anthropology (24).

The principal challenge in archeology, especially in prehistory where written records are absent, is distinguishing ritual sacrifice from other forms of ritualized violence (4, 6). To investigate formal sacrifice, defined as the killing of humans for ritual purposes (3), researchers seek recurrent patterns of behavior that deviate from the norm and that archeologists can hypothesize as sacrificial. The examination of “deviant burials” (26), i.e., those that differ from conventional burial practices for a specific population (27), along with methods of execution becomes essential. Several criteria for exploring the hypothesis of human sacrifices have been established, including indicators of violent death, unusual body positions or burial patterns, multiple concurrent burials, hierarchically related body placements, the inclusion of individuals with or instead of offerings, the distinctive arrangement of individuals, and demographic irregularities (6, 28, 29). The challenge lies in determining the threshold for classifying a burial as atypical, similar to identifying instances of violent death when no obvious signs of lethal trauma are present on skeletal remains. The postulation of human sacrifice emerges when human remains exhibit indicators of a violent demise and appear within contexts deviating from established patterns typical of interred bodies. These contexts encompass scenarios in which an individual subjected to violence is not laid to rest within the confines of a conventional burial site, and their treatment differs from the customary rites accorded to the deceased. Archaeological records most robustly support sacrificial practices when a substantial dataset exhibits a recurring constellation of distinct characteristics (4, 30). Moreover, when we observe a recurring pattern of several diagnostic traits over centuries, and when there is an absence of individuals accompanied by prestige objects, the hypotheses developed favor ritual sacrifices more than retainer sacrifices (4).

Within the Rhône Valley at the end of the Middle Neolithic period—in this region, the Middle Neolithic is between 4250 and 3600/3500 Before Common Era (BCE)—expansive sites spanning several hectares are arranged in a distinct pattern of land use and management. These sites exhibit a wealth of features, including numerous silos, numerous broken grindstones, ceramics sourced from distances

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spanning several tens of kilometers, animal remains indicative of communal meals, instances of animal sacrifices, and graves containing individuals found in configurations reminiscent of silos or resembling such structures (31). While the notion of abandoned villages has been proposed for similar sites in proximity to the Mediterranean (32), the discovery at Saint-Paul-Trois-Châteaux, where two women were found in an unconventional placement beneath the overhang of a storage pit, positioned in front of a third body (a woman) in a central location, suggests a ritualized form of asphyxiation that may even imply a method of homicidal ligature strangulation (33, 34). Given that the silo containing these bodies is part of an architectural orientation toward solstices, we lean toward the hypothesis that these sites, at a certain juncture in their history, functioned as collective gathering places where food security and the agricultural cycle were venerated, particularly through the practice of human sacrifices.

Homicidal ligature strangulation (33, 34) involves a ritualized form of ligature strangulation (33), characterized by its cruelty, in which, in its classical way, the victim, in a prone position, is bound at the throat and ankles with a rope. Self-strangulation becomes inevitable due to the forced position of the legs (33). Currently, this torture, known as *incaprettamento*, is associated with the Italian Mafia and is sometimes used to punish persons perceived as traitors (34). In various circumstances, killing people with homicidal ligature strangulation has been interpreted as a form of symbolic suicide, as it is the individual who, by strangling themselves, causes their death (35). The earliest recorded instance of homicidal ligature strangulation (35–37) dates back to the Italian Mesolithic era, possibly suggesting a highly ancient origin within ceremonial sites (Fig. 1) (36).

Considering the observation of the Rhône Valley and the Mesolithic case, we have investigated, as a basis for research and insights into the socio-religious structuring of a segment of the European Neolithic, similar cases on the Ancient (5500 to 4900 BCE) and Middle Neolithic periods (4250 to 3600/3550 BCE) in Central and Western Europe. This study examines 20 cases spanning nearly 2000 years from Eastern Europe to Catalonia. These cases originated from archaeological sites on alluvial plains, either on major rivers (Danube, Oder, Rhine, Pô) or on coastal rivers on the Mediterranean coast with a distribution quite different from that of the megalithic sites of the same period (38). In these regions, funeral sites are mostly represented by repurposed storage pits or pits dug like silos (39, 40) or ritual silos (39), where archeologists found one or more individuals or isolated bones. The deposit of human remains in circular pits was widespread throughout the Carpathian Basin (41), the Rhine Valley (13, 42–45), the Rhône Valley (46), southern France (14, 47–49), southwestern France (50), Emilia, Italy (51), and the coast of Catalonia (52, 53). In these sites, while some skeletons are in a flexed position—a standard position for this period—others are placed in atypical positions or buried unconventionally, which does not conform to the overall pattern (17). If, in some cases, deaths by stabbing or arrowheads have sometimes been described (15, 16, 40, 54) and if sometimes, researchers interpret these atypical positions as if the individuals had been unceremoniously thrown into the pits (17, 42, 55), in most cases, the cause of death is unknown, even if one hypothesizes that these individuals in atypical positions are cases of retainer sacrifice (14–16). These documented cases underscore the possible development of sacrificial practices in various regions and contexts. Particularly noteworthy are instances of homicidal ligature strangulation found in ritual sites during the Neolithic period (39, 46). These sites often included



Fig. 1. Mesolithic rock art scene from the Addaura Cave. According to J. Guilaine (36), this scene features eleven humans and a deer, which, given its position, is most likely deceased (sacrificed?). Nine of the humans are standing (in gray); several of them are adorned with bird-like beak faces, resembling masks, and they all appear highly animated. The artist aimed to convey a sense of general excitement. They encircle two central humans (highlighted by us in black), in a prone position. They lie on their abdomens with their legs folded beneath them; one has their arms hanging, while the other has them folded behind their neck. There is a rope stretched between their ankles and neck. Male genitalia in the two figures are very clearly depicted, as if erect, and the figure underneath is shown with their tongue hanging out; these two signs are found in cases of strangulation or hanging.

storage pits used for burials, occasionally accompanied by sacrificial offerings (23), broken grindstones, and isolated human remains (39). This investigation contributes valuable insights into the intricate nature of human sacrifice and ritualized violence during the European Neolithic, questioning established interpretations and highlighting the importance of thorough archaeological analysis for a nuanced understanding of these practices.

RESULTS

Saint-Paul-Trois-Châteaux

We have identified numerous sites dating back to the Middle Neolithic period (4250 to 3600/3550 BCE) in the central Rhône Valley. They are distributed according to a distinctive pattern of land use and management, with cave-sheepfolds in the hinterland of the mid-mountain region, villages in the intermediate zone, and large sites (including Saint-Paul-Trois-Châteaux) in the Rhône plain. Within these sites from the Rhône plain, numerous silos and pits are present, containing unique artifacts compared to residential sites, including dozens of deliberately broken grindstones (as this material is highly resistant, intentional breakage is suspected), animal sacrifices, notably of dogs, sometimes whole, gatherings of ceramics from long-distance communities (tens of kilometers away), and particular pit fillings, including carefully selected pebble deposits. Human remains are located in these sites, always in specific pits, some of which, shaped like silos, were constructed specifically to receive them (31).

Saint-Paul-Trois-Châteaux has revealed sepulchral pits excavated by archeologists and anthropologists with appropriate excavation protocol that allows the recording of all skeletal parts and precise observations in situ (56). The bodies of two individuals were found in an

unorthodox position and situation. The site comprises pits and silos arranged in varying proximity, some of which contain human remains. These pits are also located close to others containing fragments of grinding wheels and fauna, as documented by Beeching in 2003 (57). Notably, certain pits, including pits 69 and 70 situated at one extremity of the site, have yielded human remains. These two specific pits are situated within an area delineated by a trench that was originally part of a perishable ovoid structure (no remnants of a wall were recovered during excavation), measuring 19 m in length and 9 m in width at its widest point, open at both ends, and intentionally aligned with the summer and winter solstices (Fig. 2).

Pit 69 resembles a storage pit, but its exceptional dimensions and the fact that no seeds were found inside, particularly on the bottom, nor that the bottom shows any traces of cremation, as is often the case in storage pits to sanitize them, have led us to consider the hypothesis that this “tomb” was built to resemble a storage pit. It was lined internally with a structure made of straw, a perishable material. It yielded the skeletons of three women in anatomical connection (Fig. 3). The eldest among them [more than 50 years, (58)], designated as woman 1, is centrally positioned within the silo, reclining on her left side, with an accompanying vase placed near her head. The second and third

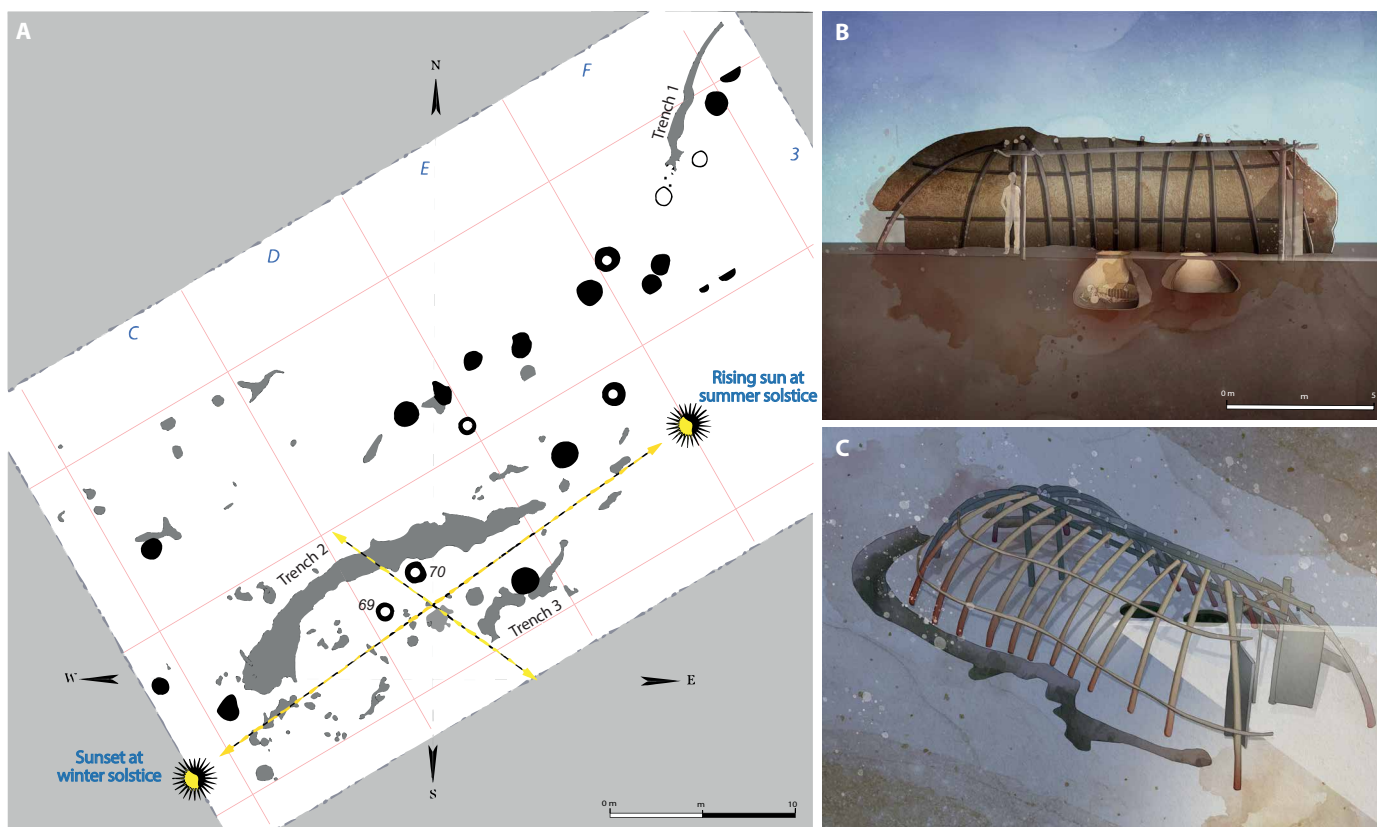


Fig. 2. Saint-Paul-Trois-Châteaux and the area surrounding pits 69 and 70. (A) Map of the western end of the Saint-Paul-Trois-Châteaux site. The black circles represent pits or silos (not all of them were contemporaneous chronologically). Pits 69 and 70 are within an area surrounded by trenches 2 and 3 (incomplete, with its southwestern part leveled). This area is perfectly aligned with the sunrise at summer solstice and sunset at winter solstice. (B) Reconstruction of the area surrounding pits 69 and 70. (C) Reconstruction of the space at sunrise during the summer solstice with pit 69 intentionally off-centered, possibly to allow the passage of sunlight during the solstice, enabling an officiant to be illuminated. Sunrise and sunset directions during the summer and winter solstices vary based on latitude and date due to elliptic changes. For dates between 3800 and 3600 BCE, these changes are minimal and not discernible on a diagram. These directions are calculated with 15-min refraction, with the sun rising approximately 4200 m away, 75 to 80 m higher during the summer solstice (1- to 2-degree angle). During the winter solstice, the sun rises on Montagne de Saint Restitut at 1550 m, 150 m higher (5- to 6-degree angle). The area's orientations are remarkable, perfectly aligning eastward for summer solstice sunrise and westward for winter solstice sunset.

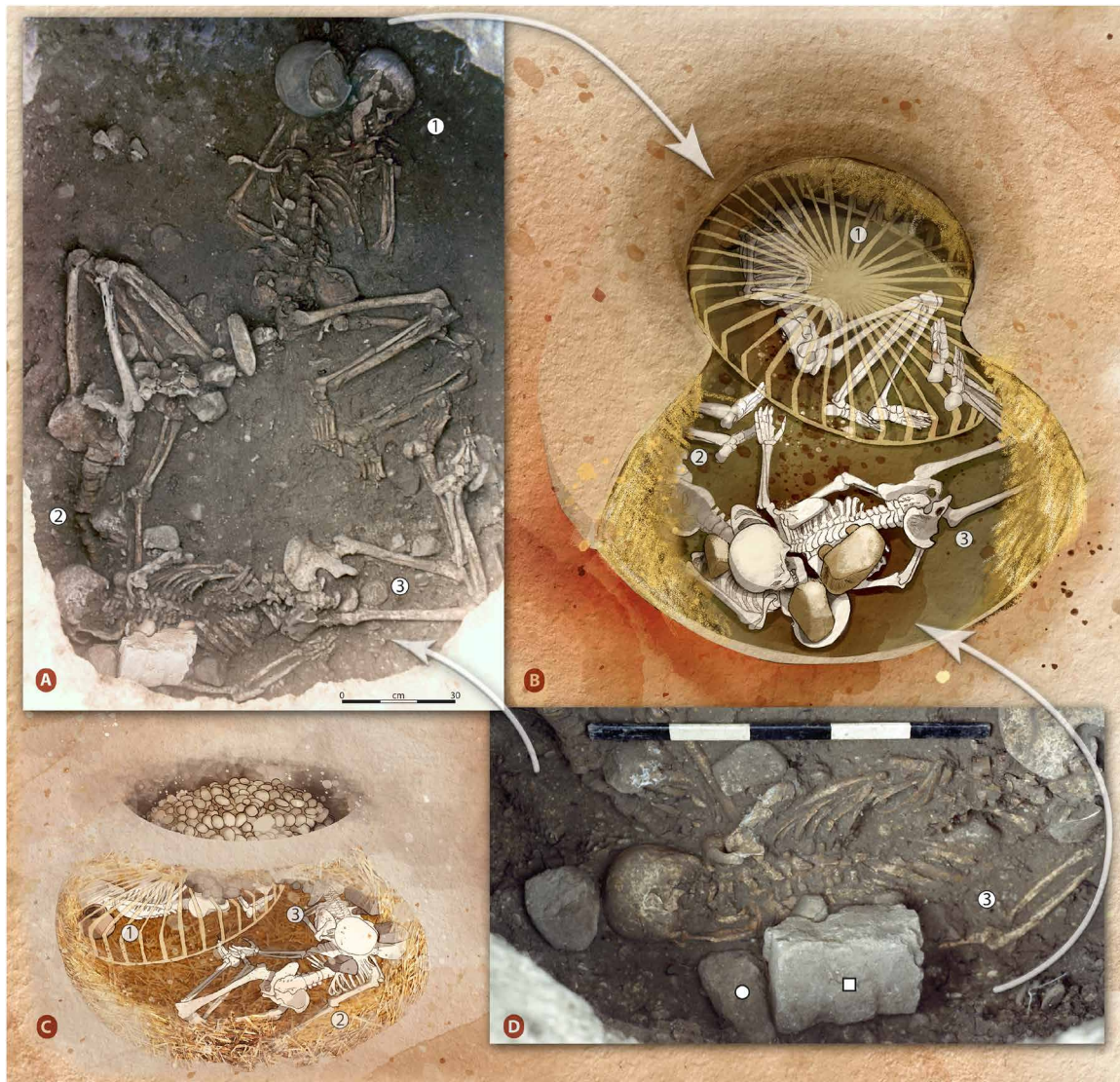


Fig. 3. Saint-Paul-Trois-Châteaux pit 69. (A) View taken from the upper part of the storage pit showing the three skeletons, with one individual in a central position (woman 1) and the other two placed under the overhang of the wall (woman 2 and woman 3). The photos were taken with a wide-angle lens; otherwise, all three individuals cannot be captured in a single shot (credit: A. Beeching). (B) Reconstruction of the remains, blocked under the overhang of the wall of the storage pit lined with straw (typical in storage pits). The woman in the central position was separated from the other two by a feature made of perishable materials. (C) Last, the storage pit was filled with stones, most likely at a later point. (D) Detailed view of the individual (woman 3) in a prone position with a box-shaped stone on the left half of her remains (white square). The upside-down grindstone fragment next to the box-shaped stone covers the head of the individual lying underneath (white circle). The scale displays 50 cm (credit: A. Beeching).

individuals (hereafter woman 2 and woman 3, respectively) are situated beneath an overhang. Woman 2 is on her back with her lower limbs bent and a fragment of grindstone positioned on her skull. Woman 3 is in a prone position with her neck resting on the thorax of woman 2. The knees are bent, and we were able to establish that it was during the decomposition of the body that her legs (woman 3) fell to the bottom of the pit, with her feet sliding along a structure—now gone—which covered the central individual, i.e., woman 1 (59). There is no indication that the wrists were bound, and woman 3 had two pieces of a grindstone placed horizontally on her back. An observer stationed outside the pit would have only been able to see woman 1, positioned conventionally (lying on the side with the knees slightly bent) for the Middle Neolithic. In contrast, only the lower limbs and

feet of woman 2 and woman 3, positioned in a “deviant” manner, were visible. Initially, we conjectured that the bodies of woman 2 and woman 3 were concealed from external observers, possibly to emphasize woman 1’s central placement (59). When we attempted to reconstruct the initial position of the bodies, it became apparent that during the decomposition of the corpses, the thoraxes of the women had collapsed, and some ribs were postmortem fractures or no longer in absolute connection. For this reconstruction, we recreated the thoracic volumes. Upon completion, we highlighted almost no space left between the thorax of woman 3 and the overhang of the storage pit. Consequently, these fragments of grindstone were forcefully inserted during the positioning of the women, thus blocking the two bodies. If they were still alive, in conjunction with their positioning beneath the

pit's overhang, then they could no longer move, and breathing became very difficult. Furthermore, since the initial descriptions, numerous forensic studies have been conducted on individuals in similar positions with pressure applied to them, resulting in their deaths (60, 61). In such a position, death occurs relatively quickly, even if the victims were not drugged or beaten. The prone position induces inadequate ventilation and a decrease in the blood volume pumped by the heart, which can lead to pulseless electrical activity arrest and/or cardiac arrest by asystole (60). This diagnosis, formerly known as positional asphyxia (62), could now be better defined as "prone restraint cardiac arrest." Some individuals are more sensitive than others, but cervical compression is an aggravating factor, as is obstruction of the nose and mouth (62). The intriguing position of the lower limbs of woman 3 is also noteworthy. Her legs collapsed to the side as the body decomposed, and from their placement on the corpse, it appears that the knees would have been bent at slightly over 90° with the legs held more or less vertically. Given the woman's prone position, this suggests a potential case of homicidal ligature strangulation. In this scenario, the woman would have been on her abdomen with a ligature attached to her ankles and neck. The fact that the woman was obstructed by grindstones and the overhang of the storage pit, coupled with the possibility of a tie connecting her ankles to her neck, supports the hypothesis of a deposit while she was still alive. Otherwise, the physical constraints could have been less severe, especially considering that the grindstones were not visible from the outside.

Since the Mesolithic era, homicidal ligature strangulation appears to have been a cruel practice associated with ceremonies and ritual sites. Rock art scenes from the Addaura cave and pit 69 from Saint-Paul-Trois-Châteaux are fascinating because of their atypical position, characteristic of homicidal ligature strangulation. Including the two individuals from Saint-Paul-Trois-Châteaux, we describe 20 individuals (nine men, seven women, and four children) from 16 tombs or pits at 14 archaeological sites (Fig. 4). The oldest sites (5400 to 4800 BCE) are from the Brno-Bohunice from Linear Pottery culture or linear-bandkeramik (LBK) culture in the Czech Republic. The most recent (4000 to 3500 BCE) are the three individuals found at Saint-Paul-Trois-Châteaux in the Rhône Valley and one in Catalonia.

On the basis of typical homicidal ligature strangulation body position (see Materials and Methods), we developed a decision tree for the 16 adult individuals (Fig. 5) that we were able to excavate or locate in the Ancient and Middle European Neolithic, where explicit photos or drawings evoked either homicidal ligature strangulation. As reference points on this decision tree, we have placed the typical cases of homicidal ligature strangulation described in the forensic literature and the two Mesolithic cases identified in parietal representations in the Addaura (18, 36). The decision tree provides a score from 0 to 3, with 3 and 2 associated with contemporary cases and the explicit Addaura cases where the individuals' hands and arms were not tied. The decision tree provides the possibility of a score of 0, corresponding to cases where an individual is placed on their back with lower limbs bent, without compression, or their hands bound. In such an instance, the individual could have moved and struggled, which explains why we did not find any cases with a score of 0. This diagram does not include children because descriptions for these cases were incomplete. However, two of them are indicative of classic homicidal ligature strangulation (Bobila Madurell sud Ms. 70 and Pontetaro T.4), while in the case of two others, aged between 6 and 9 in a prone position, the data are insufficient to specify whether it is a case of homicidal ligature strangulation or positional asphyxia, to which they are particularly susceptible (63).

For 12 individuals, the decision tree indicates a prone position with bending knees compatible with homicidal ligature strangulation. For four individuals, who are not in a prone position but on their back or side, one individual's neck (Hluboké Mašůvky, score 2) is hyperextended, which suggests homicidal ligature strangulation, while for three individuals, Bobila Madurell Sud Ms. 16, and Saint-Paul's woman 3, restraints that block the body and the over flexed cervical support a diagnosis of positional asphyxia, as it most commonly occurs when the head is markedly flexed to the extent that the chin touches the upper chest. Because of upper body compression, it is also the case for adult no. 2 at the Pujolet De Moja E13 site.

Then, to understand the structuring of the 16 tombs in relation to each other, we performed a branch-and-bound cladistic analysis and applied a Bayesian analysis to test whether chronological, geographical, or cultural models—specifically silo burials—influence the evolution of homicidal ligature strangulation to a greater extent than a model with no hypothesis. The Bayesian cultural model (Bayes factor higher than 150 in favor of the cultural model) represented by the silo pits explains the evolution of homicidal ligature strangulation over time better than the geographical model (second) or the chronological model (Table 1) (see Materials and Methods). On the basis of the high retention index (RI) value (RI = 0.72), the cladistic branch-and-bound cladogram (Fig. 6) illustrates the vertical transmission of cultural characteristics (table S3). Therefore, the ritual and its associated traits propagate conservatively, indicating a cultural structure over time. The distinctive feature which influences the cladistic analysis is for individuals with homicidal ligature strangulation, the presence or absence of pits or silos, and the presence or absence of grindstones.

DISCUSSION

The unique circumstances and positioning of two individuals in a pit resembling a silo at Saint-Paul-Trois-Châteaux during the Middle Neolithic period in the Rhône Valley led to the hypothesis that they may have died from positional asphyxia or "prone restraint cardiac arrest" (62). This hypothesis is reinforced by the possibility of similar cases elsewhere, suggesting that more of the cases discussed in this study may also reflect positional asphyxia. However, our excavations and thorough literature review have uncovered 20 probable instances of homicidal ligature strangulation or positional asphyxia spanning nearly 2000 years across various European regions. Unfortunately, insufficient data on the skeletons' positions or the lack of detailed illustrations in a lot of other publications makes an accurate diagnosis challenging and this number (20) is certainly underestimated.

In Saint-Paul-Trois-Châteaux, the possibility that these individuals perished within the grave warrants consideration. If their placement in the grave occurred postmortem and the sole objective was concealment, then a simple act of pushing them under the overhang would have sufficed. However, the specific arrangement—stacked atop each other and entwined with fragments of grindstones—implies a more forceful and deliberate placement, strongly suggesting that their demise likely occurred within the burial context (as discussed earlier). It can be inferred that the woman on her abdomen may have succumbed to prone restraint cardiac arrest due to metabolic acidosis, exacerbated by insufficient ventilation and decreased cardiac output (CO) (60). Conversely, the woman on her back, struggling to breathe under the weight of the other individual on her neck, likely died from positional asphyxia (61). Proving that all the other individuals described have

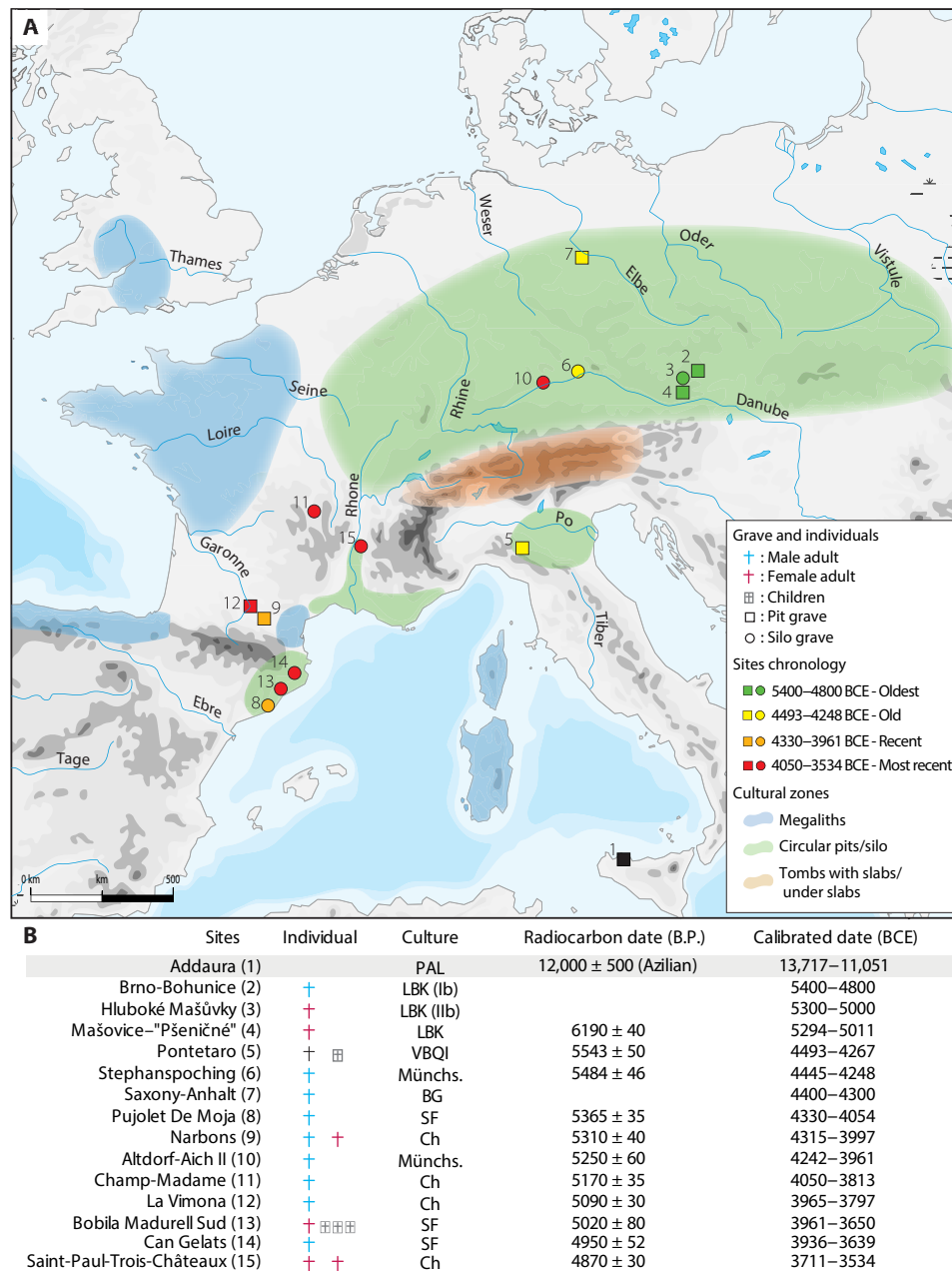


Fig. 4. Location and description for the studied sites. (A) Map of Europe presenting the archaeological sites, numbered from 1 to 15. Number 1 (black square) represents the Mesolithic Addaura site (Sicily). The green zones represent sites with circular pits or silos containing individuals in classic but atypical positions, between 4500 and 3500 BCE; the blue zones are those at the period of extension of megaliths; the brown area, centered on the Alps and the north of Italy, represents tombs created with stone slabs and tombs under slabs. The Addaura site (no. 1) is not included in the analyses. It allows for the comparison of Mesolithic and Neolithic sites. (B) Table of 14 sites described in this study. Radiocarbon dates are actualized with the 2020 Oxcal reference (78). The numbers in brackets refer to the site numbers on the map. Blue dagger, male; red dagger, female; black dagger, undetermined; dagger in box, immature. PAL, Italian Paleolithic culture; LBK, Linearbandkeramik or linear pottery culture; VBQ I, square-mouthed vase culture (first phase); Münchs., Münchshofen culture; BG, Bischeim-Gatersleben transition; SF, Sepulcros de Fosa; Ch, Chassey culture. All references associated with the sites are in the Supplementary Materials. B.P., before the present.

died at the place where they were found is often impossible, either due to insufficient archaeological data or because the archaeological data does not lend itself to such conclusions (for example, when an individual is in a pit with the body not restrained). Moreover, as with contemporary forensic studies (33, 34) or human sacrifices in an archaeological context (25), it is impossible to determine whether some

of these individuals were drugged or beaten or even, as is sometimes the case in mafia scenarios, killed before being placed in that position (33). Similar to current cases, what appears crucial—and this is the only thing we can formally assert—is the position, which points to self-strangulation and the cruelty of the torment, even if the details of this torment cannot always be documented.

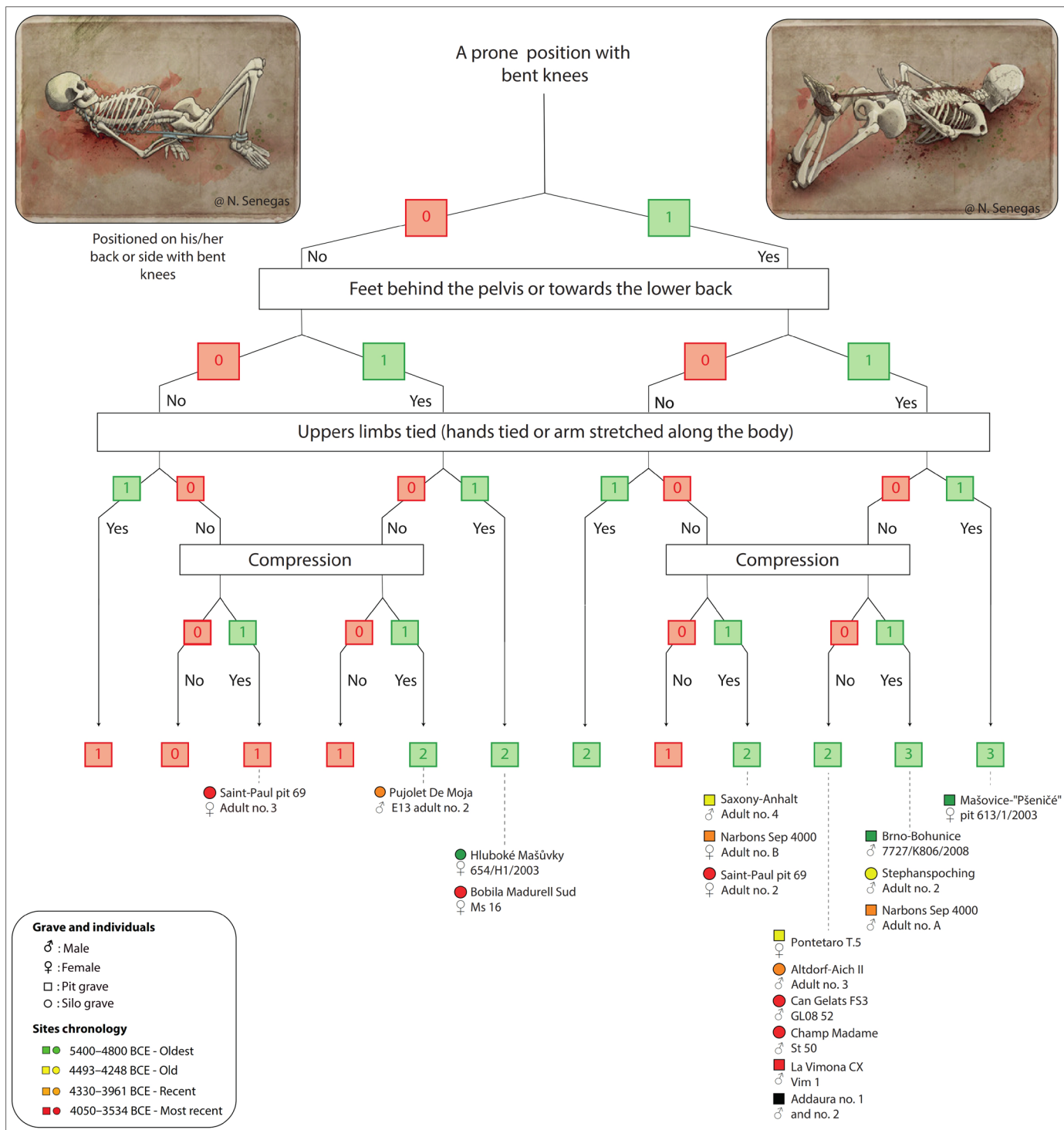


Fig. 5. Decision tree. When dealing with an individual in a prone position with bent knees, a decision tree that takes into account the positioning of the feet and upper limbs, as well as the presence or absence of compression on the head or thorax, allows us to contemplate a scenario resembling homicidal ligature strangulation. The two reconstructions at the top of the figure correspond to the position of the skeleton before decomposition in two models of homicidal ligature strangulation based on recent forensic cases. Depending on taphonomic factors, the bones of the lower limbs may fall more or less to one side.

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Table 1. Bayesian results. Geographical (G_0), chronological (Ch_0), or cultural (Cu_0) model with no hypothesis, (G_1) geographical model considering the site of Saint-Paul-Trois-Châteaux as a French site, (G_2) geographical model considering the site of Saint-Paul-Trois-Châteaux as a Mediterranean site, (Ch_1) chronological model considering the four defined chronological clusters (see Materials and Methods), (Cu_1) cultural model with typological hypothesis considering silo burials as a “monophyletic” group. Better models are those with the higher harmonic mean. The site of Saint-Paul-Trois-Châteaux is closer to the Mediterranean sites (G_1) than the French sites (G_2). However, a chronological scenario is not favorable to explain the burial rites data (Ch_1 has the worst harmonic mean). Last, an evolutionary hypothesis based on cultural factors, specifically silo burials, is very strongly supported by the Bayes factor ($B_{12} > 150$, harmonic mean = -98.69), followed by a geographical hypothesis (harmonic mean = -103.11) and a chronological hypothesis (harmonic mean = -107.46). Hm Minit, initial model harmonic mean; Hm Mi, harmonic mean for each tested model (i); Δ Hm Minit – Hm Mi, result of difference between the two harmonic means (absolute value).

	G_0	G_1	G_2	Ch_0	Ch_1	Cu_0	Cu_1
Avg std dev*	0.0095	0.009019	0.009614	0.01	0.008	0.01	0.0099
Harmonic mean	-104.18	-103.11	-104.21	-103.98	-107.46	-104.38	-98.69
ΔHm Minit – Hm Mi		1.07	0.03		3.91		5.69
2 log B_{12}		2.14	0.06		7.82		11.38
Bayes Factor B_{12}		3 to 20	1 to 3		20 to 150		>150
Best scenario		G_1		Ch_0		Cu_1	
G_0 against G_1 or G_2							
Ch_0 against Ch_1							
Cu_0 against Cu_1							
Best scenario between G_1, Ch_1 or Cu_1				Cu_1			

*Average SD of split frequencies: Should tend to 0.

A transcultural phenomenon emerges in the context of the phylogenetic tree and Bayesian analyses. By transcultural, we mean a phenomenon that spans cultures defined thus far in European Neolithic contexts based on ceramic typology. This phenomenon associates homicidal ligature strangulation with specific Neolithic sites. Among the 20 individuals in homicidal ligature strangulation or positional asphyxia induced by ligature strangulation, 12 were found in former silos or, as seen in the case of Saint-Paul-Trois-Châteaux, in pits designed to resemble silos. In 10 sites where we can accurately define the broader archaeological context of homicidal ligature strangulation, it becomes evident that these occurrences are not associated with conventional funerary sites in the classical sense of the term. These sites do not exhibit the typical arrangement of tombs clustered together, resembling traditional burial grounds or cemeteries. This observation reinforces a cultural specificity in contrast to chronological or regional distinctions. However, cultural evolution is not linear; it appears to have progressed in a mosaic pattern with ancestral traits, such as the absence of deposits in pit-like silo structures, which may have persisted for an extended period in specific recent sites. Another primitive characteristic, the presence of two sacrificial individuals, as seen in the Mesolithic of Addaura, can resurface very late, as evidenced in Saint-Paul-Trois-Châteaux. Establishing a direct correlation between Mesolithic and Neolithic instances or determining whether the practice experienced resurgence during the Neolithic remains presently infeasible, given the nearly six millennia that separate these periods. It is noteworthy, however, that in the Mesolithic, this practice involved two individuals sacrificing wild animals, such as deer (36). This duality of individuals is often associated with animals, particularly domestic ones during the Neolithic period, such as cattle or dogs (23).

In comparison to the Mesolithic site of Addaura, considered as an outpost, derived characteristics emerge from the Early Neolithic

onward, such as sites featuring multiple pits in Brno-Bohunice and the sacrificial practices involving female individuals in the nearby Neolithic site of Hluboké Mašůvky. This transcultural phenomenon spanning almost two millennia does not entirely rule out chronological or regional variations, as suggested by the distinct clusters in the phylogenetic tree. Thus, when considering the two ends of the phylogenetic tree, on one end, we have recent sites with a South European context (Saint-Paul-Trois-Châteaux in the Rhône Valley and Bobila Madurell Sud in Catalonia, to which the Pujole de Moja pit in Catalonia is added). On the other end, there is a polytomy with sites, none recent, and which exhibit a varied geographic distribution (Brno-Bohunice in the Czech Republic, Narbons in France, and Saxony in Germany). Between these two ends, until approximately 3500 BCE, various sites from different regions in Europe, including graves or pits, displayed diverse gender distributions and age groups with increasingly derived characteristics. The significance of these sites remains enigmatic due to their rarity and atypical characteristics. Conversely, the presence of homicidal ligature strangulation in local housing circular pits or silos suggests an association with supplementary rites that possibly involved the demise of other individuals through diverse means. Nevertheless, the “ancient rite of homicidal ligature strangulation” was explicitly confined to pits deemed “special and/or privileged.” However, note that the oldest sites are all in Central Europe in the LBK culture, and several derived characteristics may appear among them. Therefore, this cultural phenomenon could have diversified in Central Europe and structured itself at different rates for almost two millennia before culminating in the late Middle Neolithic in the Rhône Valley and Catalonia. These sites share many standard features huge archaeological sites, silos, ceramics from sites several tens of kilometers away; the presence of grindstones on skeletons; the presence of dogs, *Bos Taurus* (ancestor of cattle), and caprine (goat or

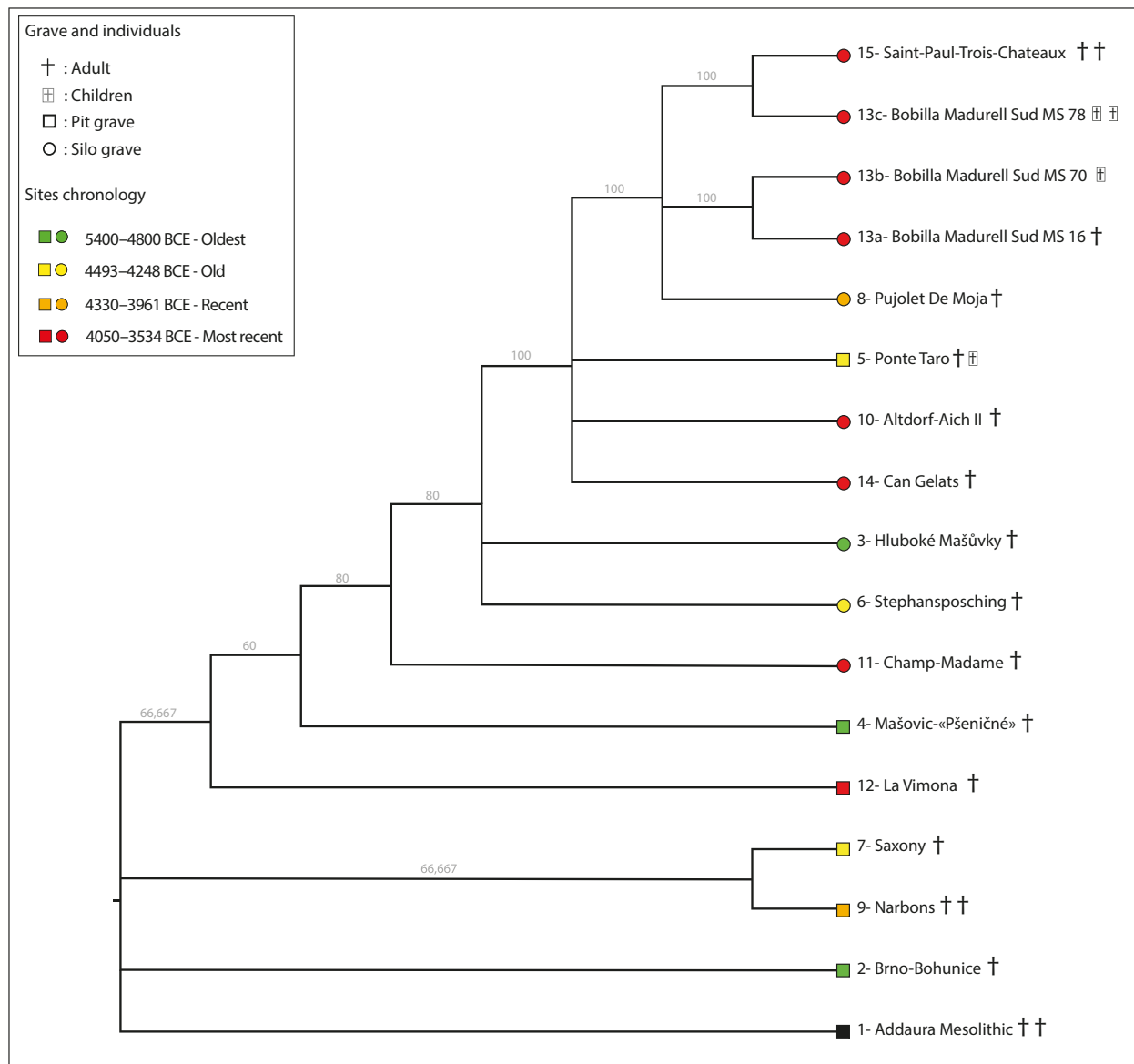


Fig. 6. Fifty percent majority-rule consensus phylogenetic tree (branch-and-bound analysis). Round symbols (○) represent silo burial; square symbols (□) represent pits. The chronological period of each site is defined according to the four clusters used for the Bayesian analyses: oldest sites in green, old sites in yellow, recent sites in orange, and most recent sites in red. The number of individuals in the grave is specified, as well as their age (dagger, adult; dagger in box, child). Consensus tree scores: length = 21, retention index (RI) = 0.72; consistency index (CI) = 0.57. The decision to place the Addaura site at the root of the phylogenetic tree stems from its position as the oldest site, a decision motivated by the hypothesis that the characters observed at Addaura are potentially ancestral. This analysis does not show a perfect chronological or geographical evolution. Admittedly, the most recent sites are positioned at the end of the tree, in a southern Mediterranean context, and show evolved characteristics. However, certain sites from three different chronological clusters are grouped at the ancestral end of the tree in a varied geographical context (France, Germany, and the Czech Republic). Last, it should be noted that the appearance of evolved characters seems to occur mainly in European sites, before spreading to southwestern France and Catalonia.

sheep); and where, from the excavation, archeologists have considered that they were dealing with a ritual site. Moreover, at Saint-Paul-Trois-Châteaux, the monumental structure accessible to only a select few with the alignment of structures concerning summer and winter solstices, symbolizing the agricultural cycle, the abundance of silos representing food security, and the arrangement of two women facing the central figure suggest a profound interconnection between religious systems and power structure in an agricultural society. The

structuring of rituals over time is linked to the convergence of several phenomena: the practice of ritualistic self-strangulation, which may have been an ancient phenomenon dating back to the Mesolithic, the deposits of grindstones that appear in the Early Neolithic Mediterranean (22) and are found quite rapidly in Central Europe (64), and the presence of gathering centers, the oldest of which seem to be the ditched roundels, circular enclosures often concentric with openings sometimes oriented toward the rising sun. These are occasionally

associated with a possible function as ritual gathering places and appeared in Central Europe at the beginning of the fifth millennium, related to the Lengyel culture (65). During the period when sites often associated with ritual practices started emerging from Central Europe to Southern Europe, and when individuals met their demise through a homicidal ligature strangulation ritual, the phenomenon of megaliths took root along the Atlantic coast. The distinction between Atlantic megaliths, Mediterranean megaliths, and the regions of interest undoubtedly exhibited permeability. Nonetheless, it was not until around 3500 BCE that we genuinely witnessed the proliferation of megaliths in Mediterranean territories (66). This expansion included the spread of monumental megalithic structures to the British Isles and the widespread adoption of collective burial practices. Before this era, this ritual sacrifice could be pivotal in ideological integration across a vast expanse stretching from Catalonia to Central Europe and Northern Italy. Subsequent studies will be necessary to corroborate either or challenge this hypothesis.

The identification of a ritual murder that shaped ancient and Middle Neolithic ritual sites across central and southern Europe raises questions about transcultural ritual and religious phenomena. These may have developed across these regions, possibly on a Mesolithic substrate, from the Early Neolithic to the end of the Middle Neolithic before being replaced in certain areas by the megalithic phenomenon. Only continuing research in a transdisciplinary approach involving Neolithic scholars, bioanthropologists, anthropologists, and forensic experts will enable a deeper exploration of these issues.

MATERIALS AND METHODS

Materials

In the investigation of Tomb 69 in Saint-Paul-Trois-Châteaux, we identified a potential instance of sacrifice linked to a diagnosis case of homicidal ligature strangulation. Building upon this finding, the authors reevaluated evidence from specific graves previously excavated by some of them, along with a comprehensive review of literature about comparable or more ancient sites. To identify specific instances indicative of homicidal ligature strangulation, the authors conducted a meticulous examination of various factors, including the condition of the skeletons, the precision of the excavations, and the details provided in relevant publications, including descriptions, drawings, and photographs. They uncovered several occurrences where individuals were lying on their backs or sides, with their lower limbs flexed to the point where their feet aligned with their pelvis, indicating an extension of the hips. Notably, these cases, akin to those observed in Saint-Paul-Trois-Châteaux, were associated with individuals exhibiting characteristics typically associated with homicidal ligature strangulation. These instances could either indicate homicidal ligature strangulation with the body being turned over postmortem or positions resembling homicidal ligature strangulation where the individual is lying on their back. In the latter scenario, if the person is immobilized in that position, there is the potential for death either due to self-strangulation or positional asphyxia (61–63). (Note that the descriptions, references, and provenance associated with each site presented are detailed in sections S1 and S2.)

In East-Central Europe

Brno-Bohunice (Czech Republic). LBK culture, 5400 to 4800 BCE (fig. S1D).

Description: In the bottom of a narrow pit oriented north/south, occupying only the posterior two-thirds with a substantial

gap between the top of his skull and the northern edge of the pit lies a man over fifty years of age in a prone position. His knees are hyper-flexed, nearly in contact with the pit's southern wall. While his lower limbs align with the axis of the pit, his torso has shifted to the left. His face rests against the western wall of the pit, along with his left upper limb; his wrists are not bound.

Suspicion of space around the body: An open space around the corpse can be suspected due to the collapse of various parts of the skeleton, particularly the inferior extremities of the legs and feet on the innominate.

Diagnosis: With a score of three in the decisional tree, this position is a classic case of ligature strangulation, where we may even suspect death in the pit. He was placed on his stomach in the posterior two-thirds of a pit initially designed for a lying down body. If he had been placed dead, with ankles tied to the neck, then we would expect, as in current forensic cases, a straight alignment of the individual. His position favors a scenario where the individual struggles while restrained.

Mašovice-“Pšeničné” (Czech Republic). End of LBK culture, 5294 to 5011 BCE.

Description: A fortuitously found pit revealed an isolated skeleton of a young female (20 to 29 years old). She is in a prone position with a slight extension of the neck and the face against the ground slightly turned to the left. The humeri are in abduction, the elbows are flexed, and the hands are brought under the thorax. The left elbow is against the pit's wall, and the right elbow is against a virtual wall. From the skull to the greater trochanter of the right femur, the bones are aligned as if there were a now-vanished virtual wall. The femora are in adduction but with a rotation to the left, causing the knees to be in contact and also in contact with the wall on the left side of the skeleton. The knees are flexed at more than 90°, but the tibiae collapsed to the right during the body's decomposition, with the feet extending well beyond the limits of the virtual wall.

Suspicion of space around the body: The collapse observed on the right side of the legs indicates an open area around the body.

Diagnosis: The position involves a classic case of ligature strangulation. The virtual wall on the right side does not align with the pit's actual wall, as the extremities of the lower limbs substantially exceeded this virtual boundary. The individual was positioned face down with hands underneath. His left elbow was confined against the left wall of the pit. This virtual boundary might correspond to a form of restraint, perhaps more likely that the individual was tied. If placed alive in the pit, then the humeri's abduction and the skull's slight rotation to the left are understandable, signifying an attempt to breathe.

Hluboké Mašůvky (Czech Republic). LBK IIB culture, 5300 to 5000 BCE (fig. S1A).

Description: In the middle of the storage pit at a depth of 60 cm lies the skeleton of a young female (25 to 30 years old). She is resting on her left side, with the neck in hyperextension; the humeri are in anteversion, with the left elbow in hyperflexion and the right elbow slightly less flexed; the left wrist is tucked under the right forearm. The femora are extended, and the knees are bent, with the heels in contact with the pelvis.

Suspicion of space around the body: As far as can be discerned, all the bones appear connected, and none seem to have been affected by the force of gravity. The decomposition of the corpse did not occur in an empty space.

Diagnosis: The position indicates a classic case of ligature strangulation. The positioning of the forearms suggests that the wrists were

bound. There is no evidence to determine whether the individual died in the pit or was brought already bound and subjected to ligature strangulation.

In West-Central Europe

Stephansposching (Germany). Münchshofen culture, 4445 to 4248 BCE.

Description: The skeleton is prone, with hyper-flexed knees and feet at the pelvis level. The individual is wedged at the pelvic level against the pit wall and in the upper body by the first individual deposited. According to the drawing (44), the skull appears to be in hyperextension, and at least the left upper limb is in antepulsion with the elbow slightly flexed and the hand brought toward the face. It is probable that initially, both individuals, including the one in the prone position, were under the overhang of the wall; it is not impossible that stones partially trapped them.

Suspicion of space around the body: The drawing is not precise enough to assert a space, but the arrangement of some bones (especially the mandible) favors such a space.

Diagnosis: The general position of the body on the abdomen with hyperflexed knees, the individual “wedged” against the wall is indicative of homicidal ligature strangulation.

Altdorf-Aich II (Germany). Münchshofen culture, 4242 to 3961 BCE.

Description: The skeleton is in a prone position with hyperflexed knees in contact with the walls of the pit, and the feet are joined at the level of the pelvis. The skull is turned to the left. The left upper limb is in extension and abduction at 90°. The right upper limb is in adduction of slightly more than 90° with a slight elbow flexion.

Suspicion of space around the body: The drawing is not precise enough to assert a space, but the arrangement of some bones (the mandible and fingers seem to be in anatomical connection) does not favor such a space during decomposition.

Diagnosis: The general position on the abdomen with flexed knees and feet resting on the pelvis is typical of homicidal ligature strangulation. The position of the upper limbs with “arms outstretched” does not rule out the possibility that the body may have been thrown into the pit rather than gently placed.

Hefta, Saxony-Anhalt, (Germany). Bischheim-Gatersleben transition, 4400 to 4300 BCE.

Description: A male individual, aged 15 to 20, is positioned in the center of a gently sloping pit containing six individuals (fig. S2, no. 4). He is in a prone position with his head resting on a child’s chest, his left upper limb positioned over the legs of an adult, and the right side of his chest resting on the pelvis of another adult. Another adult in a prone position is on his lower limbs. His humeri are along his body, the forearms are slightly flexed, and the hands are under the pelvis. Both femora are parallel, extended, and rotated to the left, while the right tibia is flexed at 135° and tilted to the right. The left femur (fractured in the middle of the diaphysis) is flexed at 90° and tilted to the right.

Suspicion of a spatial context around the body: The body decomposed within a space, and during its decomposition, it underwent collapsing effects related to the following facts: (i) It was resting in a sloping pit, causing certain bones, such as the left femur, to be cantilevered between the pit’s bottom and its incline. (ii) It partially rested on other corpses, also decomposing, creating fresh empty spaces. (iii) A corpse on its lower limbs was exerting pressure. The various empty spaces resulted in (i) the head detached from the torso and rolling forward, (ii) the thorax completely collapsing into the slight depression at the center of the pit, (iii) when reconstructing the initial position of the legs, appearing they were vertical, but the placement of a

cadaver on these vertical legs caused during decomposition their downfall.

Diagnosis: The reconstruction of the overall body position, initially lying on the stomach with the knees flexed at more than 90°, supports a diagnosis of homicidal ligature strangulation.

In the central Mediterranean area

Pontetaro T5 (Italy). Vasi a Bocca Quadrata culture (VBQI) phase 1, 4493 to 4267 BCE.

Description: In a large circular structure, there is a 30- to 40-year-old woman, on her stomach with hips bent at 90°, knees bent at 135°, one arm bent under the trunk, and the other extended along the body.

Suspicion of a spatial context around the body: Standardized archaeological data precisely specify the position of the bodies but do not provide information on the presence of any potential space.

Diagnosis: This position is a classic case of ligature strangulation with the upper limbs not tied. Seven non-synchronous burials have been studied, with dog remains also found. These burials are dated between. At the edge of this pit, an 11-year-old child (T.3) is in hypercontraction. T.4 corresponds to a child in a prone position with the hips flexed at 90°, the knees bent to the maximum, and the legs to the left.

In Western Mediterranean

Pujolet De Moja structure E 13 (Spain). Post-Cardial Neolithic of the Molinot type, 4330 to 4054 BCE.

Description: A silo dated to the second half of the fourth millennium at one end of the site yielded the remains of three individuals, represented by a skull in the center between two skeletons, end to end. On one side, a female is on her left side, a typical position for this period. On the other side, there is a sub-adult male on his back. The mandible has dropped, showing a void during decomposition; the clavicles oriented up and down are parallel, indicating upper body compression. Her hips are in extension and knees in flexion, with the ankles at pelvis level. The right hand is on the neck, and the left hand is on the upper part of the thorax. Once deposited, in a similar action for both individuals, two granite grindstones were carefully placed vertically between the upper part of the thorax and the head. In the case of the sub-adult male, the grindstone likely prevented him from moving if he was deposited alive, as his left side is very close to the wall. The fragments of ceramics, *B. taurus* (ancestor of cattle), and the many pieces of grindstones found in the pit are reminiscent of Saint-Paul-Trois-Châteaux.

Suspicion of a spatial context around the body: According to archeologists, the sediment arrived very quickly after the deposition of the individuals. The bone displacements observed by the archeologists would be consistent with small spaces that would have been maintained between the walls and the corpses (67).

Diagnosis: The position of the sub-adult male would be observed if the individual had been tied up, subjected to ligature strangulation, and placed on his back.

Can Gelats structure EF 3 (Spain). Sepulcros de fosa (FS) Middle Neolithic, 3936 to 3639 BCE (fig. S1B).

Description: An adult in a prone position with bent knees collapsed during decomposition, with the upper limbs against the body and the elbows bent to the maximum.

Suspicion of a spatial context around the body: The tibiae collapsed to the side during the body’s decomposition, implying the existence of a space in that location.

Diagnosis: It seems to be a classic case of homicidal ligature strangulation. We cannot rule out the possibility that the body was bound before or after ligature strangulation.

Bobila Madurell SUD structure Ms. 16 (Spain). Sepulcros de fosa (FS) Middle Neolithic, 3951 to 3650 BCE.

Description: In Ms. 16, there is an adult female on her back with knees bent to the maximum: “The legs, in parallel, were in maximum flexion (femora in parallel with tibiae and fibulae), and the feet were in contact with the pelvis. One of the grindstones was under the femorotibial joint, and another one was at the level of the right innominate both vertical and delimiting the skeleton” [(39), pp. 177–179]. The individual is highly compressed, with flexion of the cervical spine, and the hands are not joined. The faunal remains are those of *B. taurus* (ancestor of cattle) and *Caprine* (goat or sheep).

Suspicion of a spatial context around the body: Based on the photos, no evidence suggests the presence of a space; the skeleton is highly compressed.

Diagnosis: The individual is in an extremely confined space with maximum flexion of the knees and neck. If the individual was alive when lowered into the pit, then they died from homicidal ligature strangulation or positional asphyxia. However, we cannot rule out the hypothesis of a body tied up in a manner resembling ligature strangulation to facilitate transport.

Bobila Madurell SUD structure Ms. 70 (Spain). Sepulcros de fosa (FS) Middle Neolithic, 3951 to 3650 BCE.

Description: A silo, Ms. 70, contains a child of undetermined age in a prone position. The photo of the skeleton displays the femora in extension and the tibiae and fibulae perpendicular to the femora that have fallen to the right. Next to it are found remains of *B. taurus*, *Canis familiaris* (dog), about 30 fragmented vases, some of which are from the Chassey culture in the north, lithic remains, six grindstones, two axes, and two smoothers.

Suspicion of a spatial context around the body: The arrangement of the skeleton, along with archaeological observations, supports the presence of a space around the corpse.

Diagnosis: The overall position of the body favors homicidal ligature strangulation. The authors [(39), p. 223] also note that the position suggests a form of punishment. These same authors consider the pit to have a ritualistic significance

Bobila Madurell SUD structure Ms. 78 child II (Spain). Sepulcros de fosa (FS) Middle Neolithic, 3951 to 3650 BCE.

Description: Ms. 78 is a large silo with four individuals. First, two children aged 6 to 9 years old were deposited in the prone position, and in the central area, there was a 5-year-old child on his back with a dog next to him. According to the authors [(39), p. 231] although there are movements in the bones, the overall position is maintained, indicating that individual II had the head raised or inclined with knees bent at a right angle. In contrast, individual I held the head to the left side with the lower limbs extended. Sediment was deposited on part of the pit area, with the child in the center slightly covered (according to the photos), and a very large grindstone was also deposited in the pit. Last, a male adult was placed on his back on top of the added sediment. His head is against the wall with the lower limbs on either side of the grindstone, with his feet reaching but not touching the upper level of the child's skull (perpendicular to it). There are numerous remains of *B. taurus* (an adult and a young animal), those of two ovicaprids of which parts are apparently still connected, hundreds of stones, a minimum of ten grindstones and a pestle, bone tools, etc.

Suspicion of a spatial context around the body: The bone movements support the decomposition of the corpse in a space.

Diagnosis: For child II, considering the context and the overall position of the body, one can reasonably consider homicidal ligature strangulation.

In France

Champ Madame St 50. Chassey culture, 4050 to 3813 BCE (fig. S1F).

Description: A shallow circular pit is located in a grain storage area. In its center, on a hearth, there is a male skeleton in a prone position with knees initially bent at 90° that collapsed to the side during decomposition; it displays torsion of the neck that can only be explained, according to the anthropobiologist who carried out the field study, by “the presence of a device that maintained the observed torsion” (68), which must have been the tie that encircled the neck, the other end of which was attached to the ankles.

Suspicion of a spatial context around the body: The anthropology suggests decomposition within the sediment, with occasional empty spaces that may have persisted in certain areas.

Diagnosis: The hands were not tied and the hypothesis of ligature strangulation in situ is highly probable.

Narbons pit 400. Chassey culture (ancient phase), 4315 to 3997 BCE (fig. S1E).

Description: The double burial we excavated is that of a male and a female deposited simultaneously on a hearth (first the male and then the female) at a depth of more than a meter where both individuals are blocked by the edges of the pit. They are in a prone position with substantially bent knees, and during excavation, we identified that the intertwined lower limbs of the two individuals resulted from their falling during the decomposition of the bodies.

Suspicion of a spatial context around the body: Initially, a gap separated the bodies from the overlying sediment; we suggested that it could be attributed to hides, branches, or planks (69).

Diagnosis: The hands were not tied, and the hypothesis of double ligature strangulation in situ is highly probable.

La Vimona. Chassey culture (ancient phase), 3965 to 3797 BCE (fig. S1C).

Description: A pit with poorly defined contours yielded an adult male in a prone position with knees bent and heels brought back to about twenty centimeters from the pelvis, and the neck initially hyperextended. The right upper limb is flexed to the maximum and against the thorax; on the left, the humerus is against the body; the elbow is flexed; and the forearm is under the abdomen.

Suspicion of a spatial context around the body: According to archaeologists, the extent of the tilting movement of the skull suggests the presence of a void around the body (envelope or protection made of relatively durable organic material) (49).

Diagnosis: The hypothesis of ligature strangulation in situ is highly probable.

Methods

Age and sex determination

In the case of the adult skeletons from Saint-Paul-Trois-Châteaux, the sex estimation was conducted based on the analysis of the innominate (70), while age estimation relied on the assessment of the sacro-pelvic surface (58). We noted that the central individual was likely older than the other two, a notion seemingly substantiated by early signs of osteoarthritis, which were not observed in the other two individuals. The adolescent's age was estimated by examining epiphyseal fusion (71, 72). For other archaeological sites, age and sex

estimations provided are based on those reported in site publications. We specify in the supplementary material which methods physical anthropologists used for each site.

Body position in homicidal ligature strangulation

The position or location of a skeleton can suggest the cause of death (17) and some forms of torture involve specific positions. This is the case for homicidal ligature strangulation. To prevent the individual from moving or freeing themselves from the ties connecting their ankles to their neck, the upper limbs or body must be held in place, or “the individuals must be restrained” (13). Except for the specific position, self-strangulation will not leave traces on the skeleton; fracture of the hyoid bone, for example, is only found in a minority of all fatal strangulations (73). Ligature strangulation, due to the manner of death, could be included in “prone restraint cardiac arrest” (60, 62). The prone position induces inadequate ventilation and a decrease in the blood volume pumped by the heart (CO), which can lead to pulse arrest and/or cardiac arrest by asystole (60). Some individuals are more sensitive than others, but cervical compression is an aggravating factor, as is obstruction of the nose and mouth (34). Death from positional asphyxia by physical restraint occurs when the abnormal position of the victim’s body compromises the breathing process. A supine position with the limbs bound together behind the back can lead to death (34) especially if the mouth is obstructed or the neck is overflexed (74). In homicides committed by the Italian Mafia, the method used and the body’s positioning follow a macabre ritual, laden with significance and intended to be an admonition to others. Sometimes, the murderers bind the body postmortem after the victim has been strangled to facilitate the transportation and disposal of their remains (13). Apart from the ties, the position of the belly and the bent knees are now the criteria for identifying cases of ligature strangulation performed on living victims (13, 15, 16). To our knowledge, no case of homicidal ligature strangulation, classic or atypical, has been described in forensic medicine on skeletons once the bonds have disappeared. In analyzing each case, we meticulously considered taphonomic factors, encompassing the degradation of soft tissue in the corpse and the existence of even the slightest space, which could result in the shifting of bones—specifically, the leg and foot skeleton. This displacement might occur onto the femora if the individual was initially positioned precisely on their stomach or toward one side if there was a subtle tilt in the legs to either direction.

Phylogenetic analysis

To evaluate the evolution of the meaning of homicidal ligature strangulation over time, we examined the aspects associated with this practice in the 16 Neolithic “graves” and the Mesolithic site where representations of it were found. We do not retain the animal sacrifice association observed in several sites in the phylogenetic analysis due to the absence of specific descriptions in some papers. We were able to classify them as ancestral or derived and perform a cladistic phylogeny according to their emergence over time. Eight cultural and biological factors were considered, each with one to four derived factors depending on the type of site, the grave, the number of individuals associated with those in ligature strangulation, the sex and age, the presence of grindstones or fragments of grindstones, and the remains of hearth, sometimes encountered (tables S1 and S2). Because of the 17 analyzed tombs (16 Neolithic and 1 Mesolithic), we opted for a branch-and-bound approach using PAUP software (75, 76). To evaluate the concordance between data and trees, three quality indices are generally used in phylogeny: homoplasy index (HI), RI, and consistency index (CI). Homoplasy is when two (or more)

sites acquire the same characteristics independently (i.e., not from a common ancestor). The lower the HI index, the less homoplasy there is. The RI measures the proportion of characters shared between a group and their common ancestor (75, 76). A high RI indicates that most of the characters of tombs studied are inherited from their common ancestor. This suggests that the characters are conserved throughout evolution. Its cultural interpretation uses a threshold of 0.60. Above this threshold, we can define a vertical transmission of characters (without excluding the fraction of horizontal transmission) (77). Last, CI assesses the coherence of a morphological character with the phylogenetic tree. It is directly dependent on sample size and HI. A high CI means that the characters are relatively stable and are not subject to frequent convergent evolution.

As the Mesolithic case is the oldest found to date and represents a potential scene of homicidal ligature strangulation, the characters it contains are considered ancestral. We have therefore taken the Mesolithic representations in the Addaura with the ligature strangulation of these two adult males, as an outgroup (Fig. 6 and tables S1 and S2). The Mesolithic site roots therefore the cladistic tree as the oldest Neolithic site, Brno-Bohunice (LBK, 5400 to 4800 BCE) from LBK culture (Fig. 6).

Bayesian Modeling

We determined geographic clusters using the sites’ geographical proximity (Fig. 4). From east to west, Hluboké Mašůvky (no. 3), Mašovice-“Pšeničné” (no. 4), and Brno-Bohunice (no. 2) are in east-central Europe; Altdorf-Aich II (no. 10), Stephansposching (no. 6), and Hefta, Saxony-Anhalt (no. 7) are in west-central Europe; Pontetaro (no. 5) is in the eastern Mediterranean; and the sites of Pujolet De Moja (no. 8), Bobila Madurell Sud (no. 13), and Can Gelats (no. 14) are in the western Mediterranean. The French sites are divided between central France, Champ Madame (no. 11) and south-west France, Narbons (no. 9) and La Vimona (no. 12). The European sites were therefore grouped (nos. 2, 3, 4, 6, 7, and 10), those in the Mediterranean (nos. 8, 13a, 13b, 13c, 14, and 5), and the French sites (nos. 9, 11, and 12).

We reviewed the literature and recalibrated the radiocarbon dates to the 2020 time reference (38, 78). Bayesian analyses of two sites with two radiocarbon dates (Narbons and Stephansposching) allowed us to define the most probable dating of the site (phase model with two overlapping dates): (i) Narbons is between 4315 and 3997 BCE and (ii) the Stephansposching site covers the period from 4445 to 4248 BCE. Archaeological evidence refuted the dating of the Pujolet De Moja site for individual E13 suggested by Mestres *et al.* (79) (too recent) by placing the site toward the end of the fifth millennium. The dating of structure E26, dated to 5365 ± 35 BP, which corresponds more closely to the archaeological evidence, will therefore be retained for this site. The Bobila Madurell Sud site is dated on the basis of charcoal found in the settlement. For Can Gelats, the bones of the burial structure FS3 were too degraded to provide sufficient collagen for radiocarbon dating. We assumed a similar dating to the available burial structure FS1. As Vimona 1 belongs to the same burial phase as Cx Ag 18, the dating of Cx Ag 18 will be retained. The Czech sites of Hluboké Mašůvky and Brno-Bohunice and the German site Hefta Saxony-Anhalt do not have exact radiocarbon dates. Estimates are used in all three cases (Fig. 4). Following the updated calibration of radiocarbon dates, we defined four chronological clusters: (a) the oldest cluster with the Czech sites: Mašovice-“Pšeničné”, Hluboké Mašůvky, and Brno-Bohunice; (b) an old cluster: Pontetaro, Stephansposching, and Saxony-Anhalt; (c) a recent cluster: Pujolet de Moja, Narbons, and Altdorf-Aich II; and (d) the most recent cluster

with Spanish and French sites: Bobila Madurell Sud, Can Gelats, La Vimona, Champ Madame, and Saint-Paul-Trois-Châteaux.

We then used Bayesian analyses (80) to test whether geographical (G), chronological (Ch), or cultural (Cu) models influence the evolution of ligature strangulation more than a model with no hypothesis. Bayesian analyses offer the possibility of determining the posterior probabilities of a tree appearing according to a priori conditions or hypotheses. In our study, we postulate hypotheses (models G, Ch, and Cu). We investigate whether a model with hypothesis is more likely to occur than a model with no hypothesis. Three different models were then tested, with three different assumptions: geographical proximity (G), chronological proximity (Ch), or cultural proximity (Cu). Each model tested provides a harmonic mean value necessary to calculate a Bayes factor B_{12} (Eq. 1) and depending on its value, it is possible to define the best-suited model.

$$2\text{Log}B_{12} = 2\Delta | \text{Harmonic mean (initial model)} - \text{Harmonic mean (tested model)} |$$

$0 < 2\text{log}B_{12} < 2 = 1 < B_{12} < 3 \rightarrow$ Weak evidence in favor of a better score model

$2 < 2\text{log}B_{12} < 6 = 3 < B_{12} < 20 \rightarrow$ Positive evidence in favor of a better score model

$6 < 2\text{log}B_{12} < 10 = 20 < B_{12} < 150 \rightarrow$ Strong evidence in favor of a better score model

$2\text{log}B_{12} > 10 = B_{12} > 150 \rightarrow$ Very Strong evidence in favor of a better score model

(1)

(G): In an evolutionary model in which we assume the development of funerary practices according to geographical factors, we tried two sets of simulations with Saint-Paul-Trois-Châteaux associated within the cluster of French sites—G1—then within the Mediterranean cluster—G2. It is at the border between the French sites—nos. 11, 9, and 12—and Mediterranean sites—nos. 8, 13, 14, and 5.

(Ch): In an evolutionary model in which it is assumed that funerary practices develop according to chronological factors, the previously established clusters—a, b, c, and d—will constitute a tree typology, Ch_1 , which will be tested and confronted with a model with no hypothesis, Ch_0 .

(Cu): In an evolutionary model, we assumed that burial practices developed according to cultural factors. We hypothesized that sites containing silos—Cu1—are culturally closer than sites without silos.

We first programmed the Bayesian analyses for four independent sampling runs and one million simulations to obtain convergence of the runs with a stationary state and an average SD of split frequencies lower than or equal to 0.01 (threshold). Because of the excessive heterogeneity between the independent runs of the Monte Carlo Markov Chain (MCMC), we do not retain the stepping-stone method. According to these three hypotheses, the MCMC starts a first simulation cycle with a randomly chosen tree and assigns a score to this tree. A second tree with another configuration is then implemented (second simulation) and a score is newly assigned to it in the same way. If the score of this second tree is better than the previous one, then the second tree

is kept; otherwise, it is rejected. The software repeats steps 3 and 4 one million times. Last, if the final score of a model with a hypothesis (G, Ch, or Cu) outperforms that of a model with no hypothesis, then the model we postulated at the beginning has a greater probability of occurring. We compared the different models against a model without hypotheses as a reference— G_0 , Ch_0 , or Cu_0 . Last, we confronted the three tested scenarios to define which hypothesis is most substantiated by the Bayes factor—no hypothesis, geographical factors, chronological factors, or cultural factors.

Supplementary Materials

This PDF file includes:

Sections S1 and S2
Figs. S1 and S2
Tables S1 to S3
Data S1
References

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