Wounded to the bone: digital microscopic analysis of traumas in a Medieval mass grave assemblage (Sandbjerget, Denmark, AD 1300-1350)

Authors’ name and affiliations

Alexandra Boucherie¹,²
Marie Louise S. Jørkov³
Martin Smith²

¹ Centre de Recherche en Archéologie et Patrimoine, Université Libre de Bruxelles, Avenue F. Roosevelt, 1050, Brussels, Belgium.
² Department of Archaeology, Anthropology and Forensic Science, Bournemouth University, Fern Barrow, Poole, BH12 5BB, UK.
³ The Laboratory of Biological Anthropology, Department of Forensic Medicine, University of Copenhagen, Frederik V’s Vej 11, DK-2100 Copenhagen, Denmark.

Corresponding author
Alexandra Boucherie
alexandra.boucherie@hotmail.fr

Permanent address
Department of Archaeology, Anthropology and Forensic Science, Bournemouth University, Fern Barrow, Poole, BH12 5BB, UK.
ABSTRACT:

Battle-related mass burials are considered the most unequivocal evidence of past violence. However, most published studies involve only macroscopic analysis of skeletal remains, commonly arriving only at broad conclusions regarding trauma interpretation. The current study considers a possible avenue for achieving both greater detail and accuracy through digital microscopy.

Patterns of injury were investigated among 45 individuals from a Medieval Danish mass grave (Sandbjerget, AD 1300-1350). Injuries were recorded on every anatomical element, except hand and foot bones. Each was photographed and cast, facilitating remote evaluations. Macroscopic analysis was compared with digital microscopy in order to test the relative utility of the latter in characterizing skeletal injuries (mechanism, weapon class, direction, timing of injury).

The location of 201 observed injuries, mainly sharp force defects, suggested that many lesions were probably not inflicted by face-to-face opponents. Some microscopic features were indicative of a specific lesion type and weapon class. Digital microscopy was therefore demonstrated to be a complementary tool to macroscopic assessment, enhancing feature observation and quantification and serving to compensate for many of the limitations of macroscopic assessment.

KEYWORDS: medieval archaeology, mass grave, paleopathology, traumatology, lesion morphology, digital microscopy.
1. Introduction

As a form of social interaction, violent acts serve as one option among a range of strategies open to human beings for resolving intra- and inter-group tensions (Merry, 2009). Acts of violence appear to have been ubiquitous, although highly variable in scale and frequency throughout human history. Such behaviour is also culturally specific and subject to wide variability in forms of action that are regarded as socially acceptable (Walker, 2001; Schepers-Hughes and Bourgeois, 2004; Judd, 2006; Knüsel and Smith, 2014; Martin and Harrod, 2015). Until recently most attempts to understand the nature of past conflicts, especially for the Middle Ages, have relied upon historical sources (as discussed by Kelly, 2005; Fry, 2007; Livingstone-Smith, 2007). Archaeologists have favored material remains, such as fortifications and weapons, as ostensibly unbiased evidence of offensive and defensive strategies (Kenyon, 1990; Carman and Hardings, 1999; Keeley et al., 2007, Arkush and Tung, 2013). However, such material remains are open to alternative interpretations, such as symbols of wealth or status (Sutherland and Holst, 2005). In this respect, human remains constitute an alternative line of evidence as the most direct and unequivocal indicators for the prevalence (or absence) of violence in the past (Knüsel and Smith, 2014). Evidence generally consists either of single individuals with one or more injuries consistent with violence or multiple injuries detected on groups of skeletonized remains found in association (commonly referred to as ‘mass graves’). The latter offer opportunities to look for patterns that may inform about the nature of past conflicts between groups (Fiorato et al., 2000; Erdal, 2012).

The ‘mass grave’ is in fact a surprisingly ambiguous concept with limited agreement across osteologists and forensic practitioners concerning its definition. In the current article, we adopt Komar’s (2008) definition: a ‘mass’ burial as a single burial context containing the remains of more than ten individuals. Investigations of such assemblages can provide valuable information from both archaeological and anthropological perspectives. Analysing the physical arrangement of skeletons is essential for understanding how people adapted their mortuary practices to an unusual number of individuals killed during a single event (Cunha and Silva, 1997; Kjellström, 2005; Duday, 2008 and 2009; Castex et al., 2014; Constantinescu et al., 2015). Considering age, sex and other distinguishing features among these individuals identifies biased demographic profiles (Cunha and Silva, 1997; Fiorato 2000; Kjellström, 2005; Brødholm and Holck, 2012). Above all, trauma provides unequivocal evidence not only about the cause and manner of death, but also the intensity and nature of
armed conflicts (Knüsel and Smith, 2014). Lesion patterns confirm the intentionality of blows, and they illustrate fighting techniques and equipment involved (Lovell, 1997; Fiorato et al., 2000; Kjellström, 2005; Fibiger et al., 2013). Lastly, placing a mass grave in its broader context leads to nuanced conclusions regarding the nature of conflicts and to reevaluations of historical sources (Knüsel and Smith, 2014).

With regard to the Medieval period, apart from a few notable exceptions, such specific assemblages in Europe are scarce. The small number of identified Medieval war graves is initially surprising, considering the number of battles recorded by historical sources (Keen, 1999). However, as the dead from large battles were usually interred outside regular burial sites, the probability of recovering them is indeed quite low (Margerison and Knüsel, 2002; Dawson et al., 2003; Castex, 2008). Currently, only nine battle-related samples dated from the Medieval and post-Medieval periods, from the 8th to 19th centuries, have been anthropologically examined in Europe. By far the largest and potentially the most informative of these was a series of mass burials discovered over an extended period on the Swedish island of Gotland. Here three graves (from a total of five) excavated during the earlier 20th century and attributed to the battle of Visby (AD 1361) contained an estimated 1185 individuals (Ingelmark, 1939). A further 60 skeletons recovered at Uppsala, Sweden, are alleged to be the victims of the Battle of Good Friday (AD 1520) (Kjellström, 2005). Among other European assemblages (Zoffmann, 1982; Cunha and Silva, 1997; Eickhoff et al., 2012; Dziedzic et al., 2011; Constantinescu et al., 2015; Nicklisch et al., 2017), the most famous remains the mass grave from the Battle of Towton (AD 1461, North Yorkshire, UK), containing at least 61 individuals, 38 of which were analysed osteologically (Fiorato et al., 2000). For comparison these published examples have been aggregated and are referred to as the ‘reference sample’.

The extent to which analyses of such mass burials can offer useful insights into past conflict relies upon a second issue: the identification and interpretation of skeletal injuries. This latter is commonly rendered more difficult by taphonomic processes (Dawson et al., 2003; Kjellström, 2005; Hart, 2005; Erdal, 2012). Distinguishing real injuries from post-mortem damage is a most challenging task (Sauer, 1998). To understand how bone breakage occurred, biomechanical rules must also be considered (Berryman and Haun, 1996; Lovell, 1997; Fibiger et al., 2013; Martin et al., 2015). This diagnostic process is usually based on macroscopic observations of lesion morphology (Novak, 2000; Byers 2005; Judd, 2006).
However, commonly used criteria are subjective and unstandardized (Ubelaker and Adams, 1995; Cappella et al., 2014; Nahkhaeizadeh et al., 2014).

In the last few years, new methods have become available for examining trauma through improved technology. Initially developed in forensic and experimental cases to document the cause and manner of human death, such techniques have potential for improved objectivity. For example, computed tomography (CT) has been adopted for autopsies (Thali et al., 2003; Jacobsen and Lynnerup, 2009; Rutty et al. 2013; Scharf 2015). Scanning Electron Microscopy (SEM) has been widely used on experimental cases discriminating various bladed weapons (Bromage and Boyde, 1984; Bartelink et al., 2001; Lewis, 2008; Alunni-Perret et al., 2008; Ferllini, 2012; Kooi and Fairgrieve; 2013). More recently, analyses of cutmarks using digital microscopy has increased in osteoarchaeology and forensic anthropology (Bello and Soligo, 2008; Shaw et al., 2011; Crowder et al., 2013; Bonney, 2014; Dittmar, 2016). Offering technical capabilities, such as three-dimensional and depth of field enhancements, the utility of such a tool is a subject of on-going refinements and discussions (Bello et al., 2009). This method has been applied to archaeological examples involving considerable numbers of purported weapon-related injuries (Jantzen et al., 2010; Messina et al., 2013; Appleby et al., 2014; Constantinescu et al., 2015), although its effectiveness remains to be confirmed.

The discovery in 1994 of an apparent battle-related mass grave, dated to the 14\textsuperscript{th} century (AD 1300-1350), in Sandbjerget, Denmark, presents opportunities to obtain new data on violent events in Medieval Scandinavia (Bennike, 2006). Radiocarbon dates obtained from the skeletons suggest two historical episodes: rebellions led by noblemen against the Danish Royal Army between 1288 and 1293 and the siege conducted at Naestved in 1344 by the Danish King Valdemar Atterdag.

Two decades on from the initial discovery, the present project set out to apply current technology to further explore this episode of interpersonal conflict through systematic assessment and characterization of possible violence-related injuries. The prevalence, distribution and nature of the traumatic injuries were examined to detect patterns that might provide new information regarding the circumstances in which these injuries were inflicted. The collection was placed in a broader context by comparing it to the previously defined reference sample (Table 1).

Injuries were characterized independently by both macroscopic (from images) and microscopic methods. The principal aim was to test the relative accuracy of macroscopic
versus microscopic examination for characterizing certain features of trauma such as
category, lesion type, weapon class, direction, and timing of injury.

2. Material and Methods

This analysis was conducted on skeletal material recovered from Sandbjerget situated near
Naestved, Denmark (figure 1). A single grave containing multiple individuals was excavated
from an area of 12 m², in 1994, by the archaeology team from the South Zealand Museum.
Radiocarbon results indicated that the individuals buried on this site died around AD 1300-
1350 (Bennike, 2006). The Minimum Number of Individuals (MNI) was estimated as 60,
based on the number of right femora and right ulnae. Despite some problems of preservation,
affecting skulls, ribs, innominates and vertebrae, repeated examples of traumatic injuries were
identified on both cranial and postcranial elements (Bennike, 2006). The hypothesis of a mass
grave related to a conflict episode was strongly supported by archaeological data, i.e. a
simultaneous deposition of multiple individuals in a unique context with a demographic
profile (exclusively adult males) inconsistent with a catastrophic event such as an epidemic
(Komar, 2008, Bennike, 2006; Duday, 2008).

Data collection was performed by the first author at the Institute of Forensic Medicine in
Copenhagen where the collection is curated, with further analyses developed at Bournemouth
University. Photography and casting were essential steps in this analysis since digital
microscopy was only accessible at Bournemouth and it was not possible to transport the
original collection.

The biological profile

A total of 45 individuals were selected for study; 34 were complete, six were represented by a
cranium, and five by postcranial elements (table A.6). We excluded 15 individuals because
their skeletal elements were too fragmentary and showed taphonomic alterations that could
have limited trauma assessment. Biological profiles included sex estimation based on
morphological characteristics of the os coxae (Bruzek, 2002; Murail et al., 2005). When the
innominate was absent, qualitative criteria from the skull and measurements of postcranial
elements were used (Ascádi and Nemeskéri, 1970; Bass, 2005). Age-at-death was estimated
from the auricular surface of the ilium (Buckberry and Chamberlain, 2002). When the ilium
was absent, an age range was obtained using cranial suture closure and dental attrition
(Brothwell, 1981; Meindl and Lovejoy, 1985). The younger adults, between 20 and 30 years
old, were distinguished from those above 30 years of age at the time of death, through the scoring of late fusing epiphyses: the sternal end of the clavicle and the iliac crest (Owings-Webbs, 1985). The individuals were classified into four age groups: adolescents (ADO), from 12 to 20 years old (Shapland and Lewis, 2013), younger adults (YA) between 21 and 30 years, middle-aged adults (MA), between 31 and 40 years and older adults (OA), above 40 years old.

The recording of traumas

Trauma is defined as an injury caused by external factors to an organism (Erdal, 2012). Skeletal trauma is considered as any disruption of the integrity of bone (Merbs, 1989; Ortner, 2003). The overall assemblage was examined for signs of trauma, although assessment of much of the axial skeleton (thorax and vertebral column) was hampered by absence of skeletal elements or taphonomic alterations. Only hand and foot bones were excluded because their size would not have been adequate for cast production. Each apparent injury was initially analysed macroscopically by recording the aspect of the trauma, followed by overall and detailed photographs of injuries and their edges. The exact position of each lesion was registered, including the skeletal region (cranial vs postcranial), element and side. Maximum width and length of the injury were measured with a sliding caliper. Casts were then produced using a dental impression agent (polyvinylsiloxane) to record the trauma morphology in three dimensions (Dittmar et al., 2015). For each injury, a specific area of the edge was selected for casting, comprising an area of cortical bone, free of attached soil and postmortem cracking. This non-destructive method permitted microscopic observation of the traumatic injuries at the first author’s host institution (Bournemouth University).

Attention to Detail: Sampling and Analysis

A sub-sample of lesions was selected for further specific analyses (n=68). These derived from 42 individuals (Table 2). We decided to include any individual showing post-cranial injuries due to their rarity (n=24). Three individuals were excluded since the casts produced were not of sufficiently quality to be used (too much attached soil, presence of cracking). For each remaining individual, we selected a maximum of four traumatic injuries, including at least one injury affecting post-cranial and facial bones, if present. The representativeness of this sample was compared to the total number of injuries in the population through a $\chi^2$ goodness-of-fit test.
Macroscopic analysis

Following data capture, the lead author undertook macroscopic analysis from photographs, twice in a two-year interval and intra-observer errors were calculated through a Cohen’s kappa coefficient (table A.1 and A.2). The overall shape and characteristics of the edges, the walls and the sides of each defect were observed (figure 2) (Alunni-Perret et al., 2008; Lewis, 2008). Injury was noted as superficial or penetrative, depending on the depth of the bone disruption. A traumatic lesion was defined as penetrative if walls were deeper than 0.3 cm. This criterion can provide information on the lethal capacity of the blow, considering that penetrative blows were more likely to damage vital organs (Kimmerle and Baraybar, 2008). Striations and other damage (irregular margins) to the internal walls of lesions were recorded when present (Novak, 2000; Byers, 2005; Kimmerle and Baraybar, 2008). From these morphological traits, diagnostic interpretations were made about the trauma mechanism, the lesion type, the class of weapon, the direction of the blow and the timing of injury (Merbs 1989; Sauer, 1998; Byers 2005; Ortner 2008; Kimmerle and Baraybar, 2008) (table A.2).

Sharp force trauma is produced by pointed and edged instruments. It was identified here from the presence of any linear cut mark showing well-defined edges with flat, smooth and polished cut surfaces on both sides and with a V, U or semi-V shaped cross-section (an intermediate between U and V shape) (Lovell, 1997; Houck, 1998; Boylston, 2000). Within sharp force injuries, lesion types were classified into incision, puncture and scoop defects (Novak, 2000; Byers, 2005; Kimmerle and Baraybar, 2008). A scoop defect is defined as a penetrative concave defect with a piece of bone removed (Kimmerle and Baraybar, 2008), usually under a substantive force. The class of weapon was deduced from lesion features (shape, edges and damage) (Alunni-Perret et al., 2008; Lewis, 2008). Short-light weapons (such as knives, daggers and spears) were distinguished from long-heavy weapons (such as swords, axes and polearms) (Kimmerle and Baraybar, 2008). An attempt to detect blunt force trauma was made according to published diagnostic features (Novak, 2000; Byers, 2005; Arbour, 2008), but none was identified.

The angle at which the weapon passed through the bone indicated the direction of the blow (Symes, 1998; Boylston, 2000; Prieto 2007). We distinguished three directions of force: above, perpendicular and below.
When the lesions evidenced rounded edges and bone remodeling, they were classified as antemortem (Sauer, 1998; Novak, 2000; Ortner, 2003). Perimortem traumas occur at or around the time of death, and their diagnosis was based on characteristics consistent with fresh bone breaks (sharp edges, similar colour on both external and internal edges) with no macroscopic evidence of healing (Boylston, 2000; Novak, 2000; Byers, 2005; Judd, 2006; Ortner, 2008). Postmortem damage, caused by post-depositional phenomena (e.g. soil acidity, biological activity, ground pressure), were identified when the edges showed irregularity and roughness and when the colour of the inner fractured surfaces were lighter than the outer surfaces of the bones (White, 1992; Novak, 2000; Weber and Czarnetzki, 2001; Byers, 2005; Ortner, 2008).

Microscopic analysis

The negative impressions were converted into positive casts using a polyurethane resin Polytek© EasyFlo 60. To analyse lesion morphology, these positive replicas were examined with a Keyence© VHX 5000 digital microscope. General observation of the casts was carried out at x50 and x100 magnification, whereas the analysis of the 3D reconstruction was executed under x200 magnification. This reconstruction was obtained by selecting an area of interest with manual focus (delimitation of an upper and lower limit). Similar settings were constantly used: a set range of 3x3 images, a pitch of 10µm and a number of 25 images. Several measurements were taken on the cross-section of the 3D reconstruction, situated at the mid-point on the longest axis of the apparent traumatic defect (Bello and Soligo, 2008; table A.3). After two weeks, a set of measurements was redone with the digital microscope on 10% of the sample, and a Lin’s concordance correlation coefficient was calculated to confirm the reproducibility of the protocol. In addition, images obtained at x50, x100 and x200 magnifications were captured to rate complementary qualitative criteria (table A.4). From those images, a second set of observations was done two years after by the first author and intra-observer errors were calculated through a Cohen’s kappa coefficient. Finally, to provide additional images of trauma, four samples (from a frontal, parietal, rib and mandible) were observed with a Scanning Electron Microscope, JEOL 6010©, through InTouchScope© software.
Statistical analyses

To establish the injuries distribution pattern, the prevalence of traumatic lesions was recorded by noting the number of affected bones and number of traumatic injuries. These data were then analyzed through χ² tests, t-tests and Kruskal-Wallis tests, by skeletal region, element, side and age category. Differences in the nature of these injuries (category, lesion type, weapon class and direction) were also interrogated by skeletal region, side and age category, through χ² tests.

The resolution of the digital microscope against macroscopy for rating and interpreting injuries was tested by calculating percentage of agreement for qualitative and diagnostic features determined by both methods (number of similar answers / total number of cases). Also, we examined the potential of digital microscopy in characterizing the nature of the trauma by testing whether microscopic features were discriminant of a specific diagnostic interpretation macroscopically rated, through χ² tests and one-way ANOVA. Finally, with similar tests, the utility of the digital microscope in determining the timing of injury was investigated by identifying microscopic features capable of differentiating perimortem and postmortem defects initially identified macroscopically. All statistical analyses were performed on R®, version 3.0.2, with a p-value significance threshold of 0.05.

3. Results

The individuals for whom sex could be estimated were exclusively male (n=45). The middle adult group, (30-40 years old), represented 51% of the sample (table 3, table A.5). Thirty-four individuals were represented by both cranial and postcranial elements (table A.6).

3.1. Gross observations: overall injury prevalence and patterning of traumas

Of the 824 skeletal elements, belonging to 45 individuals that were examined for trauma, 17% were affected (n=134) (table 4). Every individual showed at least one traumatic injury. In total, 201 injuries were observed, i.e. a mean of 4.46 lesions by individual and 11 lesions maximum (individual 31) (table 4).

3.1.1. Analysis by body region

Significantly more injuries were found in cranial elements (23.5%), as opposed to 6.7% postcranial elements (χ²=32.64, df=1, p<0.05) (table 4, table A.10). The number of traumatic injuries was also significantly higher for the crania than the postcranial skeleton, 177 injuries
(3.9 per individual) versus 24 injuries (0.5 per individual) (t=8.11, p<0.05) (table 5, table A.10). Every cranium showed at least one traumatic injury.

3.1.2. Analysis by skeletal element

Among the cranial elements, the parietal bones had significantly more injuries ($\chi^2=91.98$, df=1, p<0.05) (table 4, table A.7), with 79 lesions (table 5, table A.10). The vault bones showed significantly more than the mandible and the facial bones (t=57.27, p<0.05) (table A.7). Injuries were significantly more prevalent on the lateral (parietal and temporal) than on frontal and posterior parts of the cranium ($\chi^2=12.24$, df=1, p<0.05) (table A.10).

Twenty-four postcranial injuries were detected, 25% of which were located both on the humerus and femur but this was not statistically significant among the postcranial bones considered (t=0.23, p>0.05) (Table 5, table A.10). Trauma showed a trend for higher frequencies on the lower limbs compared to the upper, without statistically significance ($\chi^2=1.08$, df=1, p>0.05) (table A.10, also see figure 3).

3.1.3. Analysis by side

Overall, more traumatic injuries were observed on the left side (n=82) than the right, although this was not statistically significant ($\chi^2=0.58$, df=1, p>0.05) (table A.10). On the cranial region, frontal and lateral lesions were significantly more frequent on the right side, whereas the injuries on the posterior cranium were significantly more common on the left ($\chi^2=12.24$, df=1, p<0.05) (table A.7). Postcranial injuries were significantly more frequent on the left than the right side ($\chi^2=5.25$, df=1, p<0.05) (table A.10).

3.1.4 Analysis by age group

There was a trend for the male middle adults (30-40) to present the highest number of trauma (n=98). This was not significant (table A.11).

3.2. Macroscopic and microscopic observations on a sub-sample: intra-observer errors

Sixty-height traumatic injuries from 42 individuals were selected to be both macroscopically and microscopically observed in detail and to be characterized. This selection was representative of the whole sample of present lesions and was confirmed by a $\chi^2$ goodness-of-fit test ($\chi^2=1.7823$, p>0.05).
The macroscopic observations were made twice in two-year interval. Cohen’s kappa coefficients regarding both qualitative criteria rating and diagnostic interpretations were calculated. They are considered as satisfactory (>0.60) and excellent (>0.90) according to Landis and Koch classification (1977) (table A.7). Similar results were found for intra-observer errors between the two sets of microscopic observations (table A.8).

Regarding microscopic measurements, the Lin’s concordance correlation coefficient calculated was above 0.70 for each measurement (table A.9). The agreement between the two sets of measurements taken by digital microscopy was sufficiently satisfactory to include measurements in statistical analyses.

3.3. Macroscopic observations: characterising traumas

Within the sub-sample of 68 traumatic lesions, 58 were identified as perimortem (85%), eight as postmortem damage (figure 4) and two as antemortem injuries. These latter were situated on one left femur and one right clavicle (figure 4). Among the perimortem traumas, 88% occurred on the cranium and 12% on the postcranial skeleton. 22 injuries were right-sided, 29 left-sided and seven parasagittal (table A.12, figure 4).

Every perimortem injury was identified as sharp force trauma (table 6). Twenty-nine were classified as scoop defects, 28 as incisions and one as a puncture defect. In total, 93.1% were penetrating defects and 89.6% were inflicted by a long-heavy weapon. Direction of the blow (assuming an individual standing upright in anatomical position) was different depending on the skeletal region and the side. Cranial injuries were significantly inflicted from above (n=35) whereas postcranial bones were significantly affected from below (n=3) (χ²=4.6, df=1, p<0.05, table A.13). Right, left and parasagittal lesions had significantly come from above (χ²=15.24, df=1, p<0.05, table A.13).

3.4. Macroscopic versus microscopic rating process

The data acquired on this sub-sample by both macroscopic and microscopic observations were confronted. Qualitative traits rated from photographs and with the digital microscope were compared (table 7). The angle of the wall was the feature that entailed the most discrepancies (59% of agreement), followed by the shape, the wall aspect and the edge aspect. The floor aspect and the cross-sections were registered only three times from images whereas the digital microscope was able record it for every case. More striations and damage were observed using the microscope, with bone shards only rated microscopically (figure 5). The
scores of agreements for diagnostic interpretations were mostly good, above 70%, except for the lesion type. Analysis from images allowed identification of puncture defects and a more balanced sample of scoop and incision injuries (table 7).

3.5. Discriminant Function? Exploring digital microscopy for trauma analysis

Data obtained with the digital microscope on this sub-sample were analysed. Few microscopic qualitative features were representative of a diagnostic interpretation obtained through macroscopic observation. Even edges and damages were significantly linked to penetrating injuries (p<0.05, table A.14). Striations were also significantly linked to scoop defects (p<0.05, table A.14) whereas the amount of damage was higher in long-heavy bladed injuries (p<0.05, table A.14). No microscopic feature was discriminant of a direction (p>0.05, table A.14).

Regarding microscopic measurements, none was found to be representative of a specific category, lesion type or weapon class (table A.15). Only one microscopic measurement was representative of the direction of the blow: the length of the trauma was significantly higher in perpendicular injuries (p<0.05, table A.15).

Microscopically, cross-section and floor aspect were not distinct between perimortem and postmortem traumas (p>0.05, table A.16), although significantly more damage on the sides was present in postmortem defects ($\chi^2=3.8556$, df=1, p<0.05; table A.16). No microscopic measurements (depth, angle of the wall, breadth, opening angle) was found to be significant in distinguishing postmortem damage from perimortem traumas (p>0.05; table A.16).

4. Discussion

4.2. Unknown warriors: characterizing the burial group

At first glance, the Sandbjerget collection shares many characteristics with the reference sample of battle-related mass graves (table 1): multiple males deposited in a single burial context, apparently having died during a singular event and showing a high prevalence of violence-related injuries. However, this assemblage also exhibits slight differences regarding the age distribution, which is at variance with the normal expectation for a Medieval army. Most of the individuals were middle-aged adults (30-40 years old) with two older adults also present. This distribution contrasts with those observed from other mass graves such as Visby, Towton and Uppsala where younger adults predominated (Ingelmark, 1939; Fiorato et al.,
2000, Kjellström, 2005). At Sandbjerget, this feature may simply indicate the presence of seasoned veterans on the battlefield. Alternatively, the presence of greater numbers of mature males may also represent the able-bodied males from the local population who had banded together for their collective defense. When the respective conflict event took place, the younger men of the community might have already been away fighting. However, we cannot exclude that this age discrepancy is linked to the fact that we analysed only 75% of the whole assemblage and that age distribution is likely to reflect methods used in age-at-death estimation.

4.3. Verifying Violence: the pattern of injuries

Caution is needed when looking at the results of the prevalence and distribution pattern of traumatic injuries at Sandbjerget since several factors could limit interpretations. Firstly, the level of fragmentation, surface preservation and overall representation of some bones, especially for the thorax, rendered the observation of the traumas difficult. The superposition of bodies within the grave, associated with soil pressure, had certainly damaged skeletons, explaining why some skulls were crushed and block lifted (Bennike, 2006, figure 6). Such damage sustained in the burial environment might also have the effect of masking any blunt force injuries that were present. Therefore, it is possible that some lesions were missed. The selection of 45 individuals out of 60, combined with the exclusion of hand and foot bones from the analysis, has probably further hampered the detection of additional injuries (Judd, 2002). Also, given the number of lesions affecting bone, it is reasonable to assume that there were also other wounds affecting soft tissue that remain inaccessible (Dawson et al., 2003; Gasperetti and Sheridan, 2013). Given these points, it is necessary to stress that the prevalence detected by the present study is likely underestimated.

4.3.1. Injury prevalence and distribution

Two hundred and one traumatic lesions were detected at Sandbjerget, with a mean number per individual for cranial injuries of 3.9. This prevalence is similar to that of Towton, suggesting the respective conflict episodes to have been highly intense (Novak, 2000). Of the total number of injuries, 88% were localized on the skull (n=177) while only 24 injuries were present on the postcranium. Similarly, uneven distributions were observed in the Towton, Uppsala and Bucharest mass graves (table 1). For instance, at Uppsala, 60% of the skulls showed 92 traumatic injuries whereas only 11 lesions were postcranial (Kjellström, 2005). In contrast, at Visby, 60% of the traumas were displayed on the postcranium (Ingelmark, 1939).
We cannot exclude that differences observed are linked to distinct methodological approaches used in trauma examination.

The overall injury distribution can give insights into fighting techniques and equipment that were employed. Even taking the poor postcranial preservation into account, it is clear at Sandbjerget that the head was the principal target of the assailants (Novak, 2000). In this regard, it is interesting to note that the head represents only 12% of overall body surface area, which would therefore appear to confirm the lethal intentionality of the blows (Fibiger et al., 2013). The head may have been the focus for tactical reasons, i.e. to temporarily incapacitate the individual, although there may also be a psychological component as the head is linked to the victim’s identity (Fibiger et al., 2013; Constantinescu et al., 2015). Novak (2000) suggested that the head was mostly affected, at Towton, due to a lack of adequate (or any) head protection. She suggested that the injured men might have been archers since they usually wore limited head protection (Bradbury, 1985; Lindhom and Nicolle, 2003). This explanation may be equally plausible here. Also, the lack of helmets could again indicate that the Sandbjerget group represents a local muster of civilian defenders with correspondingly poor equipment. It is also possible that most of the cranial injuries were given when the victim was already severely wounded and the helmet had been removed or lost (Knüsel and Boylston, 2000; Bennike, 2006).

Of course, these suggestions rest on an assumption of individuals fighting on foot. An equally important possibility is that this distribution might indicate mounted antagonists, with the head simply being the region of the body most easily within reach from horseback (Kjellström, 2005). Most of the cranial injuries were located on the lateral vault, with the parietal bone most affected, and were significantly more frequent on the right side. This low frequency of blows coming from the front, combined with the predominance of right-sided injuries, was also found in Uppsala (Kjellström, 2005). It has been proposed that these former features are inconsistent with face-to-face combat, as opposed to the presence of predominantly left-sided cranial lesions commonly considered to indicate direct hand-to-hand frontal attacks between right-handed assailants (Ingelmark, 1939; Knüsel and Boylston, 2000). Injuries detected at Visby and Towton displayed this latter pattern: 69% of cranial lesions were left-sided at Visby, whilst left-sided lesions with a high proportion of frontal traumas were observed at Towton (Ingelmark, 1939; Novak, 2000). In contrast, at Sandbgerget and Uppsala the absence of such pattern may suggest that other fighting
techniques were involved, i.e. direct or backhand blows given from horseback (Kjellström, 2005).

Conversely, injuries located on the posterior of the cranium were significantly concentrated on the left side, as observed in Uppsala (Kjellström, 2005). These posterior lesions may indicate attacks from the rear, with blows struck at fleeing men, (Ingelmark, 1939; Šlaus, 2010; Constantinescu et al., 2015) or at already fallen men (Brødholt and Holck, 2012).

The direction in which the cranial blows were delivered may further support these suggestions since they were significantly inflicted from above. This feature, together with the lack of frontal and left lesions would appear more consistent with the presence of cavalry, the natural choice of troops for pursuing routed opponents. Although, we cannot exclude that some cranial injuries were delivered from above to already seriously wounded men, lying on the ground.

No significant difference was found in the postcranial distribution of injuries, at Sandbjerget, between the upper and the lower limbs, even if more postcranial traumas were found on the lower limbs. This unclear pattern contrast with the ones observed in both Towton and Visby mass grave. At Towton, most of the injuries were located on the right forearms and were therefore interpreted as defensive injuries (Novak, 2000). Here, the presence of some defensive injuries among the humeral lesions cannot be excluded. In Visby, on the contrary, a higher frequency of lower limb injuries was found. This pattern might be attributable to the styles of armour worn during mid-14th century, when leg protection was undeveloped. In such a circumstance, striking lower limbs could be an efficient means of incapacitating an opponent prior to the coup de grâce (Mays, 1998). In fact, at Sandbjerget, it is noticeable that many postcranial injuries are located around both upper and lower joints (figure 3). This feature may suggest that blows were given to points where armour tends to offer less protection, i.e. where armour pieces intersect to allow motion.

4.3.2. The nature of injuries

In the selection of trauma characterized in detail, 85% were perimortem injuries (n=58/68). Each injury was inflicted by a sharp bladed implement; in 89.6% of cases a long-heavy implement was involved. The predominance of surviving cranial remains, compared to the thoracic skeleton for instance, could explain the result in favor of long-heavy weapons, knowing that short implements are frequently used for stabbing on the thorax (Prieto, 2007).
Unlike Towton, Wittstock, Bucharest or Lützen, no blunt force trauma was detected (Novak, 2000; Eickhoff et al., 2012; Constantinescu et al., 2015; Nicklisch et al., 2017). In 93% of traumas, the injuries were penetrating and were likely to have caused brain damage. Similarly, high frequencies of sharp force trauma, with lethal consequence, are common to other conflict-related mass graves (table 1). The location, morphology and diagnostics traits of injuries present in Sandbjerget seem to support death in battle rather than capture or execution (Boylston, 2000; Judd, 2002, Hougen, 2008).

Only two antemortem traumas were identified. At Aljubarrota, Towton and Lützen, the high percentage of antemortem injuries was linked to prior experiences in battle (Cunha and Silva, 1997; Novak, 2000; Nicklisch et al., 2017) (table 1). Here, their scarcity, as in Uppsala (Kjellström, 2005), may imply that few of them had experienced previous armed conflicts, which would again support the notion that these men were civilians who had become involved in this violent episode. Alternatively, if these men had been archers, this class of soldier was specifically involved with fighting at a distance and so could be less likely to acquire injuries in hand-to-hand fighting than other kinds of infantry. We also concede the possibility that antemortem injuries were missed because of the preservational state of the assemblage and the size of the sample analysed.

4.4. Methodological aspects

Studies which apply digital microscopy to investigate skeletal traumas in archaeological samples remain relatively rare and the current study represents one of the few that does (Messina et al., 2013; Appleby et al., 2014; Constantinescu et al., 2015). The principal question addressed here was to assess whether digital microscopy can provide valuable information on trauma that may compensate for the limitations of macroscopic observation (Ubelaker and Adams, 1995; Cappella et al., 2014; Nahkhaeizadeh et al., 2014). The consistency of both methods in the rating process is highly variable, between 52 and 96%. It was found that the contribution of the microscope is dependent on the qualitative feature under examination. With the digital microscope’s ability to visualize cross-sections in detail, some criteria were more easily scored such as the aspect of the wall, the edges, the floor and the angle of the wall. Previous studies using a Scanning Electronical Microscope (SEM) (Symes, 1998; Alunni-Perret et al., 2008; Lyne and Fairgrieve, 2009; Ferllini, 2009) have formerly raised this point. Additional features of lesion morphology have been frequently observed using digital microscopy, such as striations and damage. The observation of
striations was reported in some cases (Bromage and Boyde, 1984; Symes, 1998, Lyne and Faigrieve, 2009) but not in all (Ferllini, 2009; Kooi and Fairgrieve, 2013). However, the variety of damage easily observed by microscopy has been widely recognized (Alunni-Perret et al., 2008; Lewis, 2008; Capuani et al., 2013; Lyne and Fairgrieve, 2009; Ferllini, 2009; Kooi and Fairgrieve, 2013). Digital microscopy necessitates a small window for observing the injury, mainly focusing on the edges and so does not lend itself to exploration of the overall form of injuries. It is therefore arguably better seen as a complementary technique to more conventional observation, rather than a stand-alone solution to all aspects of trauma analysis. Some further limitations that should be considered include the point that the recognition and interpretation of features using digital microscopy are highly dependent on the observer experience (Crowder et al., 2013). The current examination was performed on casts and not directly on bone lesions, although this non-destructive protocol is widely recognized (Crowder et al., 2013, Donnellan et al., 2013; Constantinescu et al., 2015). Finally, taphonomic conditions and the narrow observation window of the protocol, i.e. we made casts on a portion of the edges and analyzed the cross-section at 50% of an area of interest, could have limited the diagnosis (Bello et al., 2009; Dittmar et al., 2015).

A point that was confirmed by the current study is the discriminant power of microscopic qualitative features in the light of diagnostic interpretation (category, lesion type, weapon class or direction). Scoop defects were significantly linked to numerous striations. This relationship has not been frequently observed (Kimmerle and Baraybar, 2008; Lyne and Fairgrieve, 2008) as striations are considered more discriminant for knife marks and incisions (Houck, 1998; Alunni-Perret et al., 2008; Lewis, 2008; Thompson and Ingris, 2009). Injuries produced by long-heavy weapons showed microscopically significantly more damage, which is a common distinction of such weapons (e.g. swords, hatchets, axes) used in both hacking and chopping motions (Alunni-Perret et al., 2008; Lewis, 2008; Lyne and Fairgrieve, 2009; Capuani et al., 2012). The most frequently used weapon, at Sandbjerget, could be narrowed towards a sword since characteristic features were present: a deep semi-V or U-shaped cross section with smooth, straight walls and intense damage on the sides (Lewis, 2008), which is consistent with weapons in use during this period (table A.17). No qualitative features were indicative of a direction, contrasting with previous results (Bromage and Boyde, 1984; Bello and Soligo, 2008; Kooi and Fairgrieve, 2013).

Only one measurement taken by digital microscopy was indicative of a specific interpretation: the length of the cut mark was variable by the direction. Blows given from above significantly
have a shorter length than blows given from below or perpendicularly, perhaps because a smaller portion of the blade penetrates bone when a blow is given from above. It is possible that other factors had influenced the length of cut marks such as the force and the speed of the blow. This result differs from previous studies that found that the angle of the wall was representative of a direction (Bello and Soligo, 2008). No straight link between a measurement and a weapon class has been found here. Yet, Bartelink and colleagues (2001) noticed that classification of weapon can be inferred from the width of the cutmark. In any case, this question must be cautiously discussed as several intertwined extrinsic and intrinsic factors influence injury morphology, including the implement used, the force and angle of the blow, and the bone density at the impact site (Bartelink et al., 2001; Capuani et al., 2012).

The only microscopic qualitative feature that significantly distinguished postmortem damage from perimortem injuries was the presence of irregular damage at the margins. This confirms common macroscopic observations made on postmortem defects, which are by nature produced by several taphonomic factors (Ubelaker, 1991). However, no microscopic measurements were indicative of the timing of injury.

This main issue has so far been addressed by few studies. Reichs (1998) developed a specific protocol but this was especially made for postmortem dismemberment. Pechnilova and colleagues (2011) elaborated a histological approach to find if an osteon fracturing pattern could differ between fresh and dry bones, without any conclusive results (Pechnikova et al., 2011). Houck (1998) described that the presence of post cut shrinkage of the periosteum, on SEM images, can distinguish perimortem and postmortem injuries. A few years ago, pilot studies based on detecting signs of hemorrhaging and red blood cell modifications appeared to be promising (Bardale and Dixit, 2007; Cattaneo et al., 2010) but the micromorphology of fracture margins produced around death and later after death remains an unsolved issue.

For the moment, timing of injury evaluation (i.e. perimortem vs postmortem) should strictly be based on a combination of macroscopic features, such as the overall shape, the irregularity of the edges, the tactile roughness of the margins and the colour of edges compared to adjacent surfaces (Ubelaker and Adams, 1995; Sauer, 1998; Cappella et al., 2014).

It appears that digital microscopy should be used as a complementary technique to macroscopic evaluation since it allows observing more precise aspects of the cutmark cross-section (edges, wall, floor) and fine details such as striations and damage. Microscopic measurements have the potential to reflect specific lesion type, category or weapon class,
even though other criteria involved in the injury morphology, such as force and rapidity, need to be considered. Casting appears to be a useful technique to remotely analyse trauma when osteological samples cannot be observed with digital microscopy at the curating institution (Dittmar et al., 2015).

**Conclusion**

The osteological collection from the Sandbjerget mass grave provides an important opportunity to collect information on an episode of organized conflict, from the perspective of individuals who did not survive the battle. The distribution pattern of the sharp force traumas prompts suggestions regarding the specific circumstances in which these men were killed. The patterning observed is most consistent with routing civilian troops being attacked from above, and probably behind, possibly by cavalry, whilst apparently lacking adequate head protection. A portion of injuries may have been inflicted when men were already seriously injured. Overall, the Sandbjerget injury distribution shares a lot of similarities with that seen at the battle of Good Friday dating from two centuries later (AD 1520), at Uppsala (Sweden). Though, it should be born in mind that the sample size and methodological approaches used are limiting comparisons.

The microscopic trauma analysis performed on this archaeological collection has produced interesting results. These data show that the digital microscope is an excellent complementary tool to the macroscopic assessment of skeletal traumas. It not only facilitates the documentation of noted features but also enhances the presence of other criteria. Unlike the SEM, observed criteria can also be easily quantified with digital microscopy (Bello and Soligo, 2008). The data recorded here do not yet have the potential to completely replace macroscopic descriptions, especially on the question of the distinction between perimortem and postmortem injuries. But, results suggest that some biomechanical links can be drawn between microscopic features and type of lesion and weapon.

In palaeotraumatology, the relationship between the micromorphology of the injury and the implement, and the force and angle involved in its origin is still complex and unclear. There is a clear need for further investigations applying digital microscopy to lesion inflicted to bone experimentally. Testing different parameters (weapons, force and direction) in a controlled environment will be fundamental in order to obtain a comparative database of microscopic trauma characteristics. This would permit the formulation of standards with sufficient reliability for application to archaeological cases.
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Authors contributions

This research was designed by A.B., M.S., and M.L.J. for a MSc thesis developed at Bournemouth University. The data collection was executed at the Forensic Medicine Institute of Copenhagen (Denmark) by A.B., with the assistance of M.L.J., and the data treatment was performed by A.B. at Bournemouth University, with the assistance of M.S. Each author worked on the final version of the paper.

REFERENCES

Arkush E., Tung T., 2013. Patterns of War in the Andes from the Archaic to the Late Horizon: Insights from Settlement Patterns and Cranial Trauma. J Archaeol Res. 21, 307-369.


