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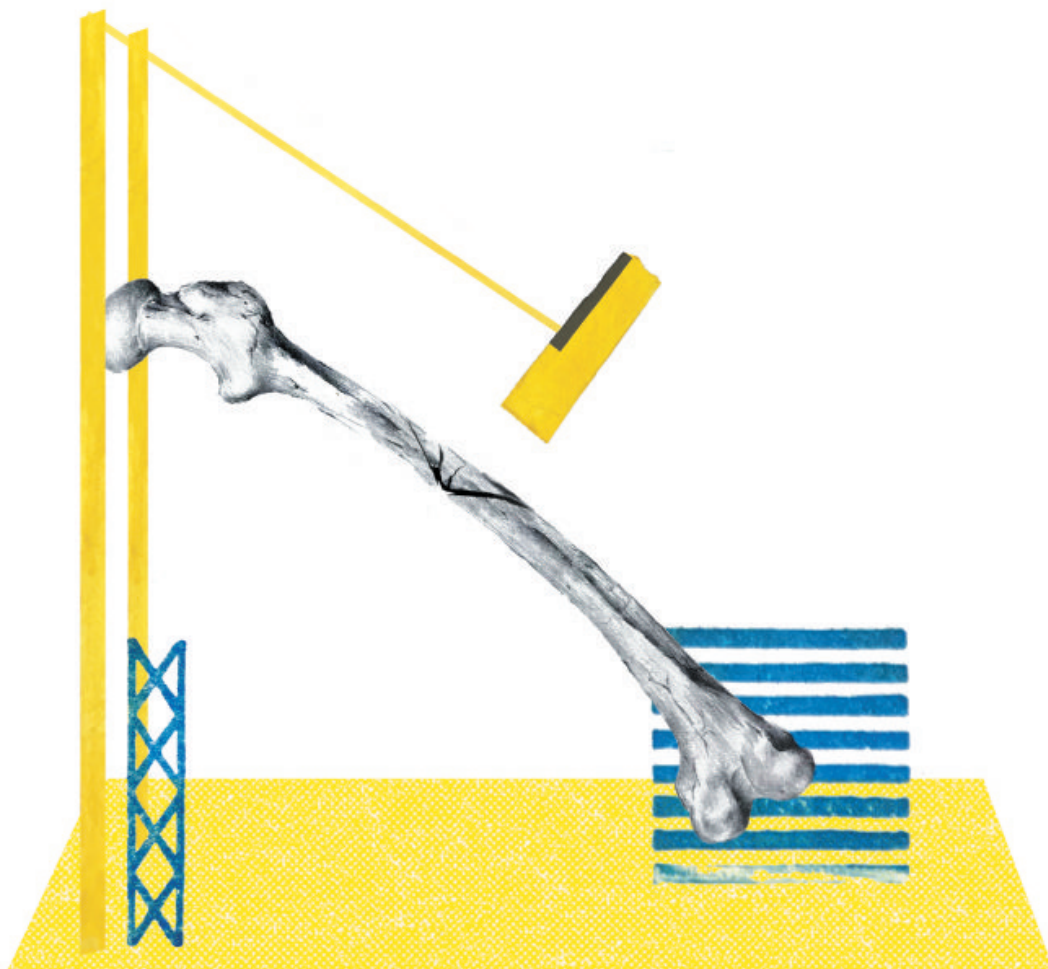
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PhD Dissertation  
by

**Sarah Frédérique Scheirs**

An experimental approach to define  
a new perimortem blunt force trauma  
bone pattern presented in a human model.



Dr. Ignasi Galtés and Dr. Assumpció Malgosa

Department of Animal Biology, Plant Biology and Ecology  
Doctorate in Biodiversity

2019











**Universitat Autònoma de Barcelona**

Departament de Biologia Animal, de Biologia Vegetal i d'Ecologia,  
Unitat d'Antropologia Biològica

**AN EXPERIMENTAL APPROACH TO DEFINE A NEW PERIMORTEM  
BLUNT FORCE TRAUMA BONE PATTERN PRESENTED IN A HUMAN  
MODEL**

Sarah Frédérique Scheirs

PhD Dissertation

Bellaterra  
2019



A dissertation submitted to the faculty at the *Universitat Autònoma de Barcelona*, by Sarah Frédérique Scheirs to obtain the degree of Doctor of Philosophy in Biodiversity at the *Departament de Biologia Animal, de Biologia Vegetal i d'Ecologia, Unitat d'Antropologia Biològica* in collaboration with the *Institut de Medicina Legal i Ciències Forenses de Catalunya*.

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*For my mom,*

*One of the strongest women I know.  
Thank you for always believing in us  
and telling us to chase our dreams.*





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## Abstract

**INTRODUCTION** - Determining the time of injury is an important but remains a challenging task in forensic anthropology. In literature, many descriptions can be found to make a distinction between peri- and post-mortem fractures but they are generally related to the post-mortem time frame. Characteristics that are more related to fractures in fresh conditions however, are not extensively investigated.

**AIM** - With this research we want to improve the current knowledge on perimortem fracture characteristics by defining a perimortem trauma pattern, that may be useful to distinguish peri- from post-mortem bone trauma.

**MATERIALS & METHODS** - This study approached this issue from an experimental point of view by comparing perimortem long bone and rib fractures from human autopsies to both fresh and dry experimentally reproduced bone fractures. A three-point bending setup was used and the bones were fractured by a slow and fast load, with and without added compression in the case of long bones to control the variable of muscle presence. In order to visualise some traits clearer, bones were cooked or directly brought in contact with black Indian ink.

**RESULTS** - As a result of our extensive research we defined new bone patterns in long bone and rib fractures with each their specific characteristics, different for dry and fresh bones. In long bones we defined layered breakage, bone scales, crushed margins, flakes with flake defect, wave lines, and plastic deformation as perimortem specific traits. With layered breakage, wave lines, and flakes as the most valuable traits.

In ribs we observed peels, folds, longitudinal lines, incomplete fractures, differential fracture edges on the internal and external part of the rib fracture, and plastic deformation. Of which incomplete fractures, plastic deformation, peels, and folds are specific perimortem traits.

**CONCLUSION** - The traits make it possible to differentiate perimortem from post-mortem fractures. Furthermore, some of the traits give information about the biomechanics of the fracture due to their location on the fracture. Additionally, in the case of long bones, flakes and plastic deformation may also be an indicator of trauma in *intra vitam* conditions, especially related with muscular reaction to injury, therefore narrowing the perimortem time gap. This might introduce a new timeframe; the *intra vitam* period, changing the anthropological paradigm.

All these new insights presented in this study might be valuable for forensic anthropologists with their perimortem bone trauma analysis and may help them to solve complicated forensic cases.







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# 1. Introduction

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*“We must have perseverance and above all confidence in ourselves. We must believe that we are gifted for something and that this thing must be attained.”*

Marie Curie



The forensic anthropologist has many different tasks, but during trauma analysis one of the most important ones is to determine the time frame in which skeletal trauma was inflicted<sup>1</sup>. Determining whether trauma occurred before, during or after death (ante-, peri-, and post-mortem, respectively) is still one of the main challenges during the interpretation of fractured skeletal remains<sup>2,3</sup>.

Antemortem trauma is defined as trauma that occurs prior to death. This also means that there will be signs of bone remodelling on the margins of the fracture<sup>4,5</sup>. Evidence of the healing process can develop as early as one week after the injury. Between weeks one and three, the edges of the fracture will start to become remoulded and rounded, and shortly after, bony calluses will begin to form<sup>5,6</sup>. While this time frame is not important for the determination of the cause death, antemortem fractures are of great value in forensic anthropology since they can contribute to the identification of skeletal remains<sup>5</sup>. Furthermore, antemortem trauma can be useful to document any history of abuse, accidental, or violent trauma<sup>6</sup>.

Both peri- and post-mortem trauma do not show any of these signs of healing, and the differentiation between perimortem trauma and post-mortem taphonomic factors such as geological, biological or (un)intentional human alterations remains difficult<sup>7-11</sup>. Moreover, the term 'perimortem' has a different meaning in the field of forensic pathology and forensic anthropology<sup>5</sup>. For forensic pathologists, 'perimortem' implies that an event occurred at or around the time of death. However, in forensic anthropology, 'perimortem' implies that trauma was inflicted when the bone still had its organic components, causing it to behave as fresh bone<sup>2,5</sup>. Perimortem injuries are of most interest in forensic cases. It defines the time around death until the bone starts losing its organic components and post-mortem

characteristics can be observed, which may occupy a long period of time in forensic anthropology<sup>5,6</sup>.

Bone consists of both organic and inorganic material. Type 1 collagen comprises 90% of the bone's organic content<sup>4</sup>. The inorganic component of bone is a mineral composed of calcium phosphate called hydroxyapatite. Collagen occurs as long elastic fibres and gives bone its flexibility, whereas the mineral component gives bone its strength and rigidity. The combination of the organic and the mineral components makes the bone stiff and elastic<sup>9</sup>.

Dry bone lacks organic compounds which makes the bone stiff but brittle<sup>12</sup>. When fresh bone is submitted to a mechanical loading, it absorbs forces first through elastic deformation, after which the bone is able to return to its original shape. With the application of greater force, the bone absorbs the inflection through plastic deformation, after which the bone will deform permanently (the yield point), before it ultimately fails and fractures<sup>12</sup>, a property known as the Young modulus (Figure 1). Dry bone lacks viscoelasticity and is unable to withstand as much elastic deformation as fresh bone. This causes dry bone to fracture immediately after the strength threshold is reached, thus no plastic deformation is observed when dry bone is fractured.

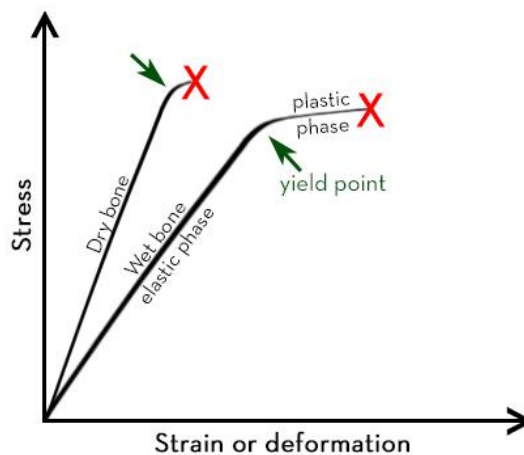


Figure 1: Young's modulus of dry and wet bone.

Due to the differences in fracture biomechanics between fresh and dry bones, the fracture patterns between these two different states are different<sup>1-5,13</sup>.

In the present study, the focus will be on perimortem blunt force trauma in long bones and ribs. Blunt force trauma is a result of a slow load application to bone. These kinds of injuries define those from homicidal assault, to blows by an object to the head or torso; and from transportation accidents, to falls from heights<sup>8,4</sup>. Fracturing a bone will result in different types of fractures, such as transverse, oblique, butterfly, spiral, comminuted, and buckle fractures<sup>4</sup>. Different fracture mechanisms produce these different fracture types, each type with its own characteristics. However, experience has taught us that most fractures are a combination of these fracture types and therefore are of a more complex nature.

For a long time, forensic anthropologists have been using several traits to distinguish fresh bone from dry bone fractures. A well-known trait to distinguish peri- from post-mortem fractures is the general appearance of the fracture. Dry bone is brittle and therefore the fracture lines will run along or perpendicular to the grain of the bone and the edges are jagged (Figure 2). Fractures in fresh bone on the other hand will have more curved or V-shaped fracture lines, and the fracture edges are smooth<sup>1,3,9,14</sup>.

Another common trait used in the distinction between both time frames is colour. If a fracture occurs post-mortem, it results in a colour difference between the exterior of the bone and the centre of the fracture due to differing exposures to soil and other environmental factors. However, when no colour difference is observed, this does not necessarily mean that the fracture can only be perimortem, since the colour could have changed again due to exposure through time (Figure 2)<sup>1,4,9,13,14</sup>.



*Figure 2:* Dry bones with brittle edges and a clear colour difference between the fracture margin and the exterior part of the bone.

Despite the available literature about the differences between wet and dry bone fractures, in practice there can still be doubt about the timing of a bone fracture<sup>1-3,11</sup>. This is especially the case when the bones do not meet the extremely fresh or extremely dry conditions. The existing literature gives primarily information about how to recognize a post-mortem fracture pattern, but there is still very little information about a specific perimortem fracture pattern. From our knowledge, no existing studies have used perimortem human bone samples to optimize the determination of the timing of bone trauma.

It would therefore be ideal if a specific perimortem bone pattern could be defined. A pattern with clear characteristics that are related to the particular biomechanics of wet bone. Furthermore, the occurrence of these traits could be statistically tested to have a higher level of confidence in the analysis. Moreover, we hypothesize that due to the mechanism of muscular contractions, there might be a specific *intra vitam* bone pattern. This may shorten the perimortem timeframe and therefore help forensic professionals to define the time of injury more accurately.

The optimal method would be one according to the Daubert standard that tests whether a method is valid according to five factors, namely that the theory or technique in question: (1) can be and has been tested; (2) has been subjected to peer review and publication; (3) has a known or potential error rate; (4) has standards that control and maintain the operation; and (5) has been accepted within the scientific community<sup>15</sup>. Such a method should be developed by means of an experimental approach.

Literature can be found on studies that use an experimental approach to investigate bone fractures, nevertheless all of them are based on an animal model. In order to develop a valid method according to the Daubert Standard, we believe it is essential to perform the experiments and make observations using human samples.

This research was performed on human material only. Animal material was excluded in order to obtain human specific results. That makes this study original and distinct, but it also means that there are limitations since we rely on victims from accidents presenting our specific variables, and on samples from people that donated their corpse to science.

## **2. Objectives**

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The main purpose of this research is to improve the current knowledge on bone characteristics and find new features that may be useful to distinguish peri- from post-mortem bone trauma. We will try to find answers by approaching this issue in a practical way from an experimental point of view based on the following research questions;

1. What are the macroscopic characteristics that define a distinct perimortem pattern in wet long bone fractures, caused by blunt force trauma?
2. Can certain characteristics be related to certain trauma mechanisms and therefore deduce the biomechanical circumstances?
3. Can the anthropological paradigm be changed by the presence of *intra vitam* traits?
4. Can perimortem traits be visualised with different imaging techniques?

## **2.1. Structure of the dissertation**

After the *Introduction* and *Objectives*, this dissertation will continue with a general chapter on *Materials and methods*. For a more detailed description of the materials and methods in each study, you can consult the corresponding article in the following *Results*. In this chapter, our seven articles are presented, stating the addressed research questions. Five articles are already published in either the *International Journal of Legal Medicine or Forensic Science, Medicine and Pathology*. Both journals are globally recognized for forensic research. Another article is published in a new Spanish forensic journal; *Revista Internacional de Antropología y Odontología Forense* and one article has been submitted to *Forensic Science, Medicine and Pathology*. The *Results* chapter is followed by a general *Discussion* that allows a holistic approach to the main objective of this research and the dissertation finishes with *Conclusions*.





### **3. Materials and methods**

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This research was only performed on human material. The study was therefore approved by the Ethics Commission of Human and Animal Experimentation (CEEAH) of the UAB and by the Ethics Commission of the Hospital Universitari de Bellvitge – Institut de Medicina Legal I Ciències Forenses de Catalunya (CEIC, Generalitat de Catalunya, Departament de Salut), to comply with the ethical requirements.

In this study human fractured long bones and ribs were collected from autopsies for further medicolegal investigations, and unfractured long bones and ribs were used for experimental purposes. Long bones were chosen due to their high incidence in forensic cases. Ribs, on the other hand, were chosen since they have a very different architecture compared to long bones and they are quite problematic since the literature on rib fractures is scarce.

### **3.1. Samples**

This study used two main sample groups: perimortem fractured specimens and healthy unfractured bone samples for experimental analysis.

#### **3.1.1. Autopsy Samples**

Fresh fractured specimens were obtained from forensic autopsies at the Institute for Legal Medicine and Forensic Science of Catalonia (IMLCFC). The trauma circumstances of all the bone fractures were traffic accidents, falls, train collisions, and CPR (in the case of ribs). They were removed for complementary medicolegal investigation with a post-mortem interval (PMI) between 12 and 24 hrs. These samples were defined as perimortem and *intra vitam* fractures since they all occurred close to the time of death without signs of healing.

### **3.1.2. Experimental samples**

Unfractured, healthy dry and fresh bones were obtained from the Medical Anatomy Department of the Universitat Autònoma de Barcelona (UAB) acquired from people that donated their body to science with a PMI of 1-3 days.

### **3.1.3. Long bones**

In total, 43 autopsy long bone samples were used with an age range of 55-91 years old. For the experimental part, 33 fresh bones and 24 dry samples were available from donors with an age range of 56-94 years old.

### **3.1.4. Ribs**

151 anterolateral autopsy rib samples with an age range of 21-87 years old were compared with the pattern exhibited by 30 experimentally fractured ribs; 18 fresh and 12 dry rib samples from donors with an age range of 27-94 years old.

All samples are kept in the private collection at the IMLCFC, registered as a collection at the Instituto de Salud Carlos III (Reference C.0004241).

## **3.2. Bone preparation**

### **3.2.1. Autopsy samples**

In order to analyse the fractures obtained from autopsies, they were defleshed using a soft cleaning method that will have no damaging effects on the bones when applied in a controlled and stable manner<sup>16-18</sup>. Fresh bone samples were placed in a water with detergent solution (one cup of commercial degreasing detergent in 5 L of water) that was heated up to 100 °C. After reaching 100 °C, the temperature was lowered to 80–90 °C and the bones were boiled for 2–5 hrs, depending on the amount of flesh that had to be removed. After cooking, the remaining soft tissue was physically removed, and the bones were placed overnight in a 50% acetone-water solution to degrease the bones. Afterwards, they were cleaned with water and left to dry. If the bones were very greasy, they were boiled again in a 2.5% ammonia solution until all the grease was removed.

### **3.2.2. Experimental samples**

The fresh bones provided by the UAB were surgically defleshed with only the periosteum remaining attached. After the experiments, they were cleaned as described in 3.2.1.

The dry bones were dusted off and ready to be used.

### **3.2.3. Fracture line staining**

To investigate how fracture lines could be made more visible to the naked eye, two non-destructive methods were tested. The first enhancing method involved the addition of 1 ml of ink to the water and detergent solution followed by the rest of the defleshing method described in 3.2.1.

In the second method, ten new bones were defleshed according to the method described in 3.2.1. When the bones were clean and dry, 1–5 drops of ink were applied onto the fracture line and capillary action would cause the ink to diffuse through the fracture lines. In total, three different types of ink were used to make the observations; Talens® Black Indian Ink, Parker® Black Ink, and Pelikan® Blue Ink.

## **3.3. Fracture reproduction**

Fresh and dry human long bones were used to reproduce fractures in order to compare them to the samples received from autopsies.

### **3.3.1. Slow speed fracture reproduction**

A three-point bending was conducted to experimentally reproduce post-mortem fractures. First, this was done with the use of a servo-hydraulic testing machine from research group GRABI (EM2/20 MicroTest, with an U10M/25-kN loading cell) and was controlled through the SCM3000 program to register the applied forces. Furthermore, cellulose blocks were attached to the two supports and loading cell as an attempt to simulate soft tissue and to prevent direct contact on the bone. The supports were adjustable depending on the bone that was broken (long bone or rib) (Figure 3).



*Figure 3: Slow speed experimental set-up.*

The bending tests were performed on six fresh and four dry human bones at a strain rate of 160 mm/min. A three-point bending results in a tension side opposite to a compression side and was chosen since it is a common type of fracture in long bones<sup>19</sup>; furthermore, it is easy to control. In this type of bending the tension side will fail first under the applied force and will continue in two directions until it reaches the compression side, this will usually result in a butterfly fracture<sup>4</sup>. The relation between force and stress during bending can be written as the following equation<sup>20</sup>:

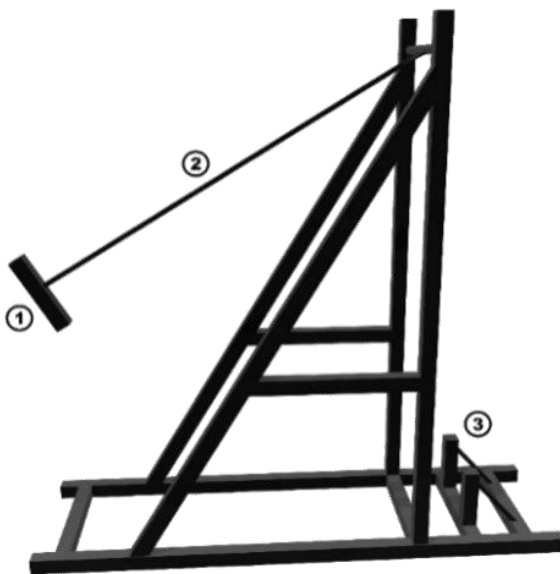
$$\text{Stress} = \frac{\text{AxialForce}}{\text{Area}} - \frac{\text{BendingMoment}}{\text{SecondAreaMoment}} \cdot \text{Distance}$$

The 'AxialForce' is zero when no compression is applied to the bone. When compression was applied (see section 3.3.3), the axial force would be larger than zero. 'Area' is the total area of the central cross section of the bone. 'BendingMoment' is determined by the total applied force and the distance between the two supports. During the experiments, the total force that was applied fluctuated between 1728 N and 3489 N. 'SecondAreaMoment' is a

measure of efficiency of a cross-sectional shape to resist bending when a force is applied. The final variable is the 'Distance' from the centre of gravity of the central cross section of the bone to the point where the stress is measured<sup>21</sup>.

### 3.3.2. High speed fracture reproduction

A pendulum impact test machine, the Blunt Force Trauma Simulator (BFT Simulator), was designed and custom-made to experimentally reproduce fractures in dry and fresh long bones (UAB). The machine consists of a metal frame and pendulum to which hammers of different weights can be attached. The set is loosely based on the Charpy impact test<sup>22-24</sup>. Hammers of 5.6 kg and 8.0 kg were available for inclusion in the study and could be physically switched when appropriate (Figure 4).



*Figure 4:* 3D model of the BFT Simulator with removable hammer (1), 1,51 m long arm (2), and adjustable holders for the sample (3) (reused with permission from Springer)

The bone samples were placed horizontally and attached with tie-wraps to the two movable metal holders, with the anterior side facing the hammer.



The bones were hit in the middle of the anterior side of the bone from a 90° angle. Additionally, a piece of soft rubber was attached to the hammer to avoid direct contact with the bone and to provide soft tissue simulation.

To reproduce fractures caused by a high-speed impact in ribs, a devised drop test machine was designed that was connected to an oscilloscope, which was, in turn, connected to a computer to measure the velocity of the drop (Figure 5). This high-speed bending test was performed on dry and fresh ribs at a strain rate of 3 m/s with the loading cell facing the external side of the rib. In both experiments, a rubber coating was attached to the loading cells as an attempt to simulate soft tissue and to prevent direct contact of the metal loading cell onto the ribs.



*Figure 5:* Drop test machine to fracture rib samples at high-speed.

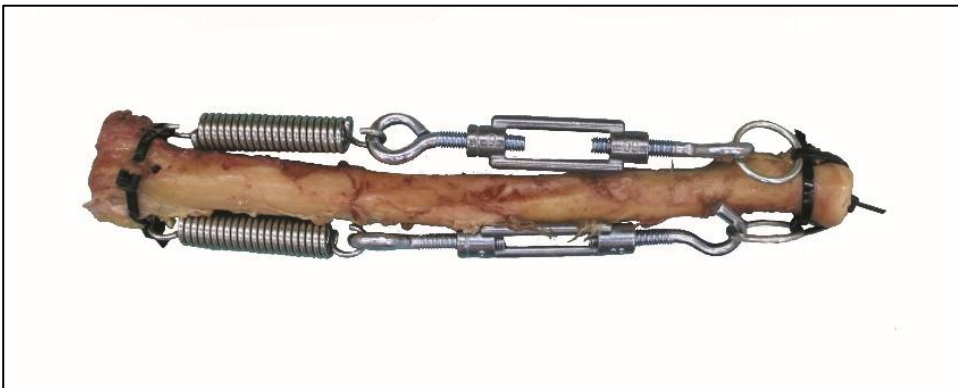
### 3.3.3. Bone compression

We hypothesized that soft tissue and especially muscular contractions may have an influence on the occurrence of the traits when a bone fractures *in vivo*. A way to approach this effect on was to apply axial compression.

First, during the slow-speed experiments we attached metal tensors to the diaphysis of the bone and estimated that the applied compression by the tensors was 7 kg ( $70 \pm 5$  N).

Later for the high-speed experiments, we improved the compression set-up by adding a spring. This way, the effect of muscle contraction could be investigated (Figure 6). The maximum amount of compression exerted to the bone varied between 12 and 19 kg, for upper and lower limbs, respectively, measured with a dynamometer.

Dry bone samples were all broken without compression, as no muscle compression can be expected in dry, post-mortem bone fractures.



*Figure 6:* Experimental set-up with metal tensors and springs to add axial compression.

### **3.3.4. Statistical analyses**

In the study of long bones, a logistic regression was performed to assess the relationship between the occurrence of any long bone trait and the sample type, i.e., autopsy and experimental samples (with and without compression). We hypothesize that if statistical differences were to be found, it means that the analysed trait was related to the tested sample type.

In the study of ribs, another logistic regression was carried out to establish whether there were statistically significant associations between fracture pattern, trauma mechanism, sex, and age. The odds ratios (ORs) were adjusted for cases, where the rib samples came from. Moreover, the percentages of occurrence of the traits were calculated and compared between groups of traumas caused by CPR and accidents using a Mann-Whitney U test.

A confidence interval of 95% was used where results with a p-value  $\leq 0.05$  were considered statistically significant.

All statistical analyses were performed in IBM SPSS statistics software.

### **3.4. CT analysis**

To investigate the detectability of perimortem traits on a CT scan, 15 fractured long bone fragments were analysed. They were collected from 10 forensic cases including; eight males and two females with an age range of 16 to 85 yrs, and mean age of 40 years. The bone samples were scanned with the Aquilion™ PRIME from Toshiba Medical Systems, allowing a multislice computed tomography (MSCT) to be taken, generating up to 160 slices per rotation.

The scan was taken with a slice thickness of 0.50 mm in the axial plane and 0.43 mm in the sagittal and coronal plane, with 120 kVp and 300 mAs as basic parameters. The acquired images were obtained in a Digital Imaging and Communications in Medicine or DICOM format. The individual slices were examined using Starviewer Medical Imaging Software, and Philips IntelliSpace Portal was used to reconstruct a 3D model of the bone samples.



## 4. Results

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#### **4.1. New insights in the analysis of blunt force trauma in human bones. Preliminary results (2016)**

International Journal of Legal Medicine in 2016 (IF 2.382, Q1 in Legal Medicine)

Addressed research questions:

What are the macroscopic characteristics that define a distinct perimortem pattern in wet long bone fractures, caused by blunt force trauma?

Can certain characteristics be related to certain trauma circumstances and therefore deduce the biomechanical circumstances?

Can the anthropological paradigm be changed by the presence of *intra vitam* traits?

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## New insights in the analysis of blunt force trauma in human bones. Preliminary results

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**Abstract** Determining the time of injury is an important but still a challenging task in forensic anthropology. In literature, many descriptions can be found to make a distinction between perimortem and postmortem fractures. Characteristics that are more related to fractures in fresh conditions, however, are not extensively investigated. This study compared 28 perimortem fractures from autopsies and 21 both fresh and dry experimentally reproduced human bone fractures. Preliminary results showed the following five distinct traits that might be related to perimortem conditions: layered breakage, bone scales, crushed margins, wave lines and flakes with matching flake defect. These distinct traits might not only be good estimators of perimortem trauma but also may be an indicator of trauma in *intra vitam* conditions, especially related with muscular reaction to injury. Furthermore, layered

breakage seems to be a good trait to infer the biomechanics of trauma.

**Keywords** Forensic anthropology · Bone trauma · Time of injury · Perimortem trauma

### Introduction

The interpretation of bone trauma is an important task for forensic anthropologists. The mechanical properties of bone allow anthropologists to attempt to reconstruct the biomechanics of trauma and to derive knowledge about possible circumstances related to the manner of injury [1]. This allows professional forensic anthropologists to complement the work of forensic pathologists in the assessment of bone injuries in fresh cadavers [2, 3]. One of the main and most important challenges in trauma analysis however is the assessment of the time of injury [4].

To assess the time an injury occurred, a distinction is made between antemortem, perimortem and postmortem trauma [5]. Antemortem trauma is defined as trauma that occurs prior to death and where signs of bone remodelling can be observed on the edges of the fractures. These antemortem injuries are of great value for forensic anthropologists as they can contribute to the identification of skeletal remains [6, 7]. On both perimortem and postmortem trauma, there are no signs of healing. Although there are some known morphological or possible colour differences, the differentiation between perimortem trauma and postmortem taphonomic factors such as geological, biological or (un)intentional human alterations remains difficult [4, 8–11].

According to Symes et al. (2012), anthropologists use the term perimortem when they imply that trauma was inflicted when the bone still has its organic components causing it to

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behave as fresh bone [12]. Therefore, it is difficult to distinguish an injury that is associated with the death of an individual from an event that occurs in a period of time close after death, when bone is still fresh [7, 13]. Due to varying circumstances in decomposition rates, the length of this period, the perimortem interval, can vary significantly. For this reason, the perimortem stage has an unpredictable time span and may occupy a long period of time. Nawrocki (2009) argues that the perimortem interval lasts until skeletal remains exhibit postmortem, dry bone, characteristics [10, 14, 15]. From a medicolegal point of view, the perimortem stage is important, since any injury directly associated with the manner of death is considered a perimortem injury [4, 5].

In literature, there are already many characteristics known to make the distinction between perimortem and postmortem fractures like colour, smooth and rough edges [8, 16, 17]. In reality, however, this distinction is still a challenge in forensic cases, especially when the bones are still fresh. From our knowledge, there are no studies that shorten the perimortem interval by defining distinct *intra vitam* traits. The present study focused on perimortem blunt force trauma in human long bones. Blunt force trauma is a result of a slow load applied to bone. These kinds of injuries define all injuries from homicidal assault, blows by an object to the head or torso, traffic accidents and falls from heights [8, 18].

The main aim of this study is to obtain bone patterns that might allow us to optimise the distinction between perimortem and postmortem fractures. This will be done by means of comparing fractured specimens from autopsies with experimental reproduced fractures. We hypothesise that there may be specific *intra vitam* traits. In order to test our hypothesis, we will investigate macroscopic characteristics that define a distinct perimortem pattern by searching for evidence that might suggest the presence of musculoskeletal activity and surrounding flesh. In case we would find such evidence, we will try to reproduce them in both dry and fresh bones.

## Materials and methods

This study used both perimortem fractured and healthy unfractured bone samples. In total, 28 fractured specimens from 16 cases were used in this study (Table 1). These specimens came from forensic autopsies at the Institute for Legal Medicine and Forensic Science of Catalonia (IMLCFC), which were removed for complementary medicolegal investigation. The Medical Anatomy Department of the Autonomous University of Barcelona (UAB) provided 21 healthy, unfractured, fresh and dry bones from people that donated their bodies to science (Table 2). This study was approved by the Ethics Commission of Human and Animal Experimentation (CEEAH) work of the UAB, in order to comply with the ethical requirements.

**Table 1** Descriptive table of perimortem bone fractures from autopsies (IMLCFC)

Case no.	Type of bone	Sex	Age	Trauma circumstances
#001	Femur (×2) Tibia (×2) Fibula (×2)	Male	22	Traffic accident
#002	Humerus Ulna	Male	20	Traffic accident
#003	Femur Humerus Ulna	Male	56	Traffic accident
#004	Radius Femur	Male	50	Traffic accident
#005	Humerus	Female	41	Fall
#006	Humerus	Female	30	Fall
#007	Tibia Fibula	Male	73	Traffic accident
#008	Humerus	Male	83	Fall
#009	Femur	Male	77	Fall
#010	Humerus	Male	39	Fall
#011	Femur	Male	34	Fall
#012	Femur (×2)	Male	25	Traffic accident
#013	Femur	Male	49	Fall
#014	Femur	Female	63	Fall
#015	Femur	Male	85	Fall
#016	Humerus Ulna	Male	39	Fall

**Table 2** Fresh and dry bone samples from donors (UAB)

No.	Type of bone	Sex	Age
Exp#001	Fresh humerus	Female	94
Exp#002	Fresh humerus	Male	74
Exp#003	Fresh humerus Fresh ulna Fresh radius	Male	67
Exp#004	Fresh humerus	Female	86
Exp#005	Fresh femur	Female	88
Exp#006	Fresh femur	Male	68
Exp#007	Fresh femur	Male	46
Exp#008	Fresh humerus	Female	94
Exp#009	Fresh humerus	Female	81
Exp#010	Dry humerus	Male	62
Exp#011	Dry fibula	Male	70
Exp#012	Dry humerus	Female	86
Exp#013	Dry humerus	Male	85
Exp#014	Dry tibia	Female	72
Exp#015	Dry femur	Female	64
Exp#016	Dry fibula	Male	83
Exp#017	Dry radius	Male	78
Exp#018	Dry ulna	Male	68
Exp#019	Dry ulna	Female	75

### Fracture reproduction

The fresh bones from the UAB were provided completely defleshed with only the periosteum remaining attached. These fresh and dry human bones (Table 2) were used to reproduce fractures in order to compare them to the samples received from autopsies (Table 1). The samples from autopsies were defined as *intra vitam* fractures; they occurred close to the time of death without signs of healing. Fracturing a long bone will result in different types of fractures depending on how the force is applied to the bone. They can result in transverse fractures, oblique fractures, spiral fractures and butterfly fractures (like our samples in Table 3) [19, 20].

In this study, a three-point bending (referred to as 3-PB in Table 4) was conducted to experimentally reproduce postmortem fractures. This was done with the use of a servo-hydraulic testing machine from research group GRABI (EM2/20 MicroTest, with an U10M/25-kN loading cell) and was controlled through the SCM3000 program (Fig. 1). To break the

bones, cellulose blocks were used on the two supports and the loading cell as an attempt to simulate soft tissue and to prevent direct contact on the bone. The bending tests were performed on six fresh and four dry human bones at a strain rate of 160 mm/min (Table 3). A three-point bending results in a tension side opposite to a compression side and is chosen since it is a common type of fracture in long bones [21]; furthermore, it is easy to control. In this type of bending, the tension side will fail first under the applied force and will continue in two directions until it reaches the compression side; this will usually result in a butterfly fracture [18].

The relation between force and stress during bending can be written as the following Eq. [22]:

$$\text{Stress} = \frac{\text{Axial force}}{\text{Area}} - \frac{\text{Bending moment}}{\text{Second area moment}} \times \text{Distance}$$

where the 'axial force' is zero when no compression is applied to the bone. When compression was applied with the tensors,

**Table 3** Overview of all the available samples and corresponding traits that were present (+) and/or absent (–)

	Case no.	Type of bone	Bone Sample	Type of fracture	Perimortem pattern				
					Layered breakage	Wave lines	Bone scales	Flakes/flake defect	Crushed margins
Fractured specimens (IMLCFC)	#001	Femur (R)	1	Transverse	+	–	–	+	+
			2	Transverse	+	–	+	+	–
			3	Comminuted at epiphysis	+	–	+	+	–
			4	Butterfly	+	–	–	+	–
			5	Epiphysial	–	–	–	–	–
			6	Transverse	+	–	–	–	+
#002	Humerus	Ulna	7	Oblique	+	–	–	–	–
			8	Transverse	+	–	–	–	–
			9	Comminuted spiral	+	+	–	–	–
#003	Humerus	Ulna	10	Comminuted at epiphysis	–	–	–	–	–
			11	Transverse	+	–	–	+	–
			12	Epiphysial	–	–	–	–	–
#004	Femur	13	Comminuted	+	–	–	+	–	
#005	Humerus	14	Comminuted	+	–	–	+	–	
#006	Humerus	15	Spiral	–	+	–	–	–	
#007	Tibia	Fibula	16	Comminuted	+	–	–	+	–
			17	Comminuted transversal	+	–	–	–	–
#008	Humerus	18	Comminuted spiral	–	+	–	–	–	
#009	Femur	19	Comminuted at epiphysis	+	–	–	+	–	
#010	Humerus	20	Comminuted	+	–	+	–	–	
#011	Femur	21	Comminuted	+	–	+	–	+	
#012	Femur (R)	Femur (L)	22	Transverse	+	+	+	–	–
			23	Oblique	+	–	–	+	+
#013	Femur	24	Comminuted	+	+	+	–	–	
#014	Femur	25	Comminuted transversal	+	+	–	–	–	
#015	Femur	26	Butterfly	+	+	+	+	–	
#016	Humerus	Ulna	27	Transverse	+	–	+	+	–
			28	Transverse	+	–	–	+	–

the axial force would be larger than zero. According to Rouvière and Delmas, the biceps creates a compression of 4.8 kg on the bone and the triceps 8.5 kg [23]. We estimated that the applied compression was 7 kg ( $70 \pm 5$  N), which is in consonance with the range mentioned by Rouvière and Delmas (1988). 'Area' is the total area of the central cross section of the bone. 'Bending moment' is determined by the total applied force and the distance between the two supports. During the experiments, the total force that was applied fluctuated between 1728 and 3489 N. 'Second area moment' is a measure of efficiency of a cross-sectional shape to resist bending when a force is applied. The final variable is the 'distance' from the centre of gravity of the central cross section of the bone to the point where the stress is measured [24].

In order to simulate more real conditions, two specimens were fractured under axial compression since it is well known that when a bone fractures, muscular contraction plays a role [25–27]. We hypothesised that some of the traits might be related to this mechanical loading. The axial compression was created with the use of metallic tensors attached around the proximal and distal bone ends. We assumed that when the fracture starts, the tensors would cause a simulation of muscle contraction.

### Bone preparation

In order to analyse the fractures, the fractured bones from the reproduction experiments and autopsies were defleshed using the method currently used at IMLCFC. Fresh bone samples were placed in a water with detergent solution (one cup of commercial degreasing detergent in 5 L of water) that was heated up to 100 °C. After it reached 100 °C, the temperature was lowered to 80–90 °C and the bones were boiled for 2–5 h, depending on the amount of flesh that had to be removed. After cooking, the remaining flesh was physically removed and the bones were placed overnight in a 50% acetone-water solution to degrease the bones. Afterwards, they were cleaned with water and left to dry. If the bones were very greasy, they were boiled again in a 2.5% ammonia solution until all the grease was removed.

### Results

During the macroscopic examination of the non-experimental fractured bones, four distinct characteristics could be observed with the naked eye being layered breakage in the compression side, bone scales in the compression side, flakes with flake defect and crushed margins. One-trait, wave lines were easier, but not exclusively, visualised when using some magnification ( $\times 10$ – $\times 40$ ). These traits are summarised in Table 3 and defined a perimortem pattern. Later, these patterns were compared to the experimentally broken bones (Table 4).

Layered breakage (Fig. 2) could be observed in 82% of the fractured samples from autopsies and in all the experimental postmortem fractures in fresh bone. This trait was always present in the compression side when a fracture takes place in the diaphysis of a long bone. Layered breakage is very distinctive since it is a pattern that occurs in the cortical bone rather than the surface or edge of the bone. The remaining 18% of samples, where layered breakage was not observed, were spiral fractures and fractures at the epiphyses. Furthermore, we were not able to experimentally reproduce this trait in dry bones.

Wave lines appeared on the smooth edges of fractures. It is a smooth undulation that has a gentle slope and a rapid drop, resembling a 'wave' (Fig. 3). This trait could mostly be observed in butterfly fractures, spiral fractures and comminuted fractures with large pieces where the edges were smooth and long.

Bone scales appeared on the compression side and resemble 'fish scales' (Fig. 4). These bone scales are a form of plastic deformation that is tangible. They occur close to the margin of the fracture, but there is still some space left between the margin and the first scales occurring. They were small and very superficial, but there were also cases where they were thicker.

A flake is a result of a broken bone scale or a superficial loss of bone (Fig. 5). When one of the scales breaks off, it is referred to as a flake (Fig. 5c). Flakes are rarely recovered since they tend to get lost in the surrounding flesh in fresh fractures. The imprint that is left on the bone is defined as a flake defect. This may resemble a bevel; however, a bevel goes through the entire cortical bone [8]. A flake defect is only superficial or until the middle of the cortical.

Crushed margins are similar to bone scales, but occur on the margin of the fracture, making this trait very vulnerable for destruction (Fig. 6). Moreover, they were only observed on the compression side.

As seen in Table 4, none of these distinct features could be experimentally reproduced in dry bones, in contrast to the fresh bones where some of them were reproduced. Dry bone fractures had a very different structure than the fresh ones (Fig. 7). In all the fresh bones, without compression, layered breakage could be reproduced. In the fresh bones with compression, observations were made of layered breakage, bone scales and flakes with flake defects. Additionally, only in the fresh bones with compression, plastic deformation could be observed.

### Discussion

When bone is subjected to blunt trauma, it will undergo two mechanisms before a fracture occurs. In the first phase, the bone will undergo elastic deformation. This

**Table 4** Overview of the experimental samples and corresponding traits that were present (+) and/or absent (-)

	Case no.	Type of bone	Bone sample	Type of loading	Type of fracture	Perimortem pattern				
						Layered breakage	Wave lines	Bone scales	Flakes/flake defect	Crushed margins
Fresh sp. (UAB)	Exp#001	Humerus	E1	3-PB	Transverse	+	-	-	-	-
	Exp#002	Humerus	E2	3-PB	Transverse	+	-	-	-	-
	Exp#003	Humerus Ulna Radius	E3	3-PB + axial compression 3-PB	Very small butterfly	+	-	+	+	-
			Very small butterfly		+	-	-	-	-	
			Oblique		+	-	-	-	-	
	Exp#004	Humerus	E6	3-PB + axial compression	Transverse	+	-	+	+	-
	Exp#005	Femur	E7	3-PB	Transverse	+	-	-	-	-
	Exp#006	Femur	E8	3-PB + axial compression	Oblique	+	+	+	+	-
	Exp#007	Femur	E9	3-PB + axial compression	Transverse	+	+	+	-	-
Exp#008	Humerus	E10	3-PB	Transverse	+	-	-	-	-	
Exp#009	Humerus	E11	3-PB	Transverse	+	-	-	-	-	
Dry sp. (UAB)	Exp#010	Humerus	E12	3-PB	Fake butterfly	-	-	-	-	-
	Exp#011	Fibula	E13	3-PB	Comminuted	-	-	-	-	-
	Exp#012	Humerus	E14	3-PB	Oblique	-	-	-	-	-
	Exp#013	Humerus	E15	3-PB	Comminuted	-	-	-	-	-
	Exp#014	Tibia	E16	3-PB	Transverse	-	-	-	-	-
	Exp#015	Femur	E17	3-PB	Transverse	-	-	-	-	-
	Exp#016	Fibula	E18	3-PB	Transverse	-	-	-	-	-
	Exp#017	Radius	E19	3-PB	Comminuted	-	-	-	-	-
	Exp#018	Ulna	E20	3-PB	Comminuted	-	-	-	-	-
	Exp#019	Ulna	E21	3-PB	Fake butterfly	-	-	-	-	-

means that the bone can return to its original dimensions. In the second phase, it will endure plastic deformation, which will deform the bone permanently. The plastic phase will continue until breaking occurs [9, 28]. Fresh bone is moist and contains water and organic components as lipids and collagen, which make the bone stiff and elastic. Dry bone lacks these organic components, which make the bone stiff but brittle [29]. The loss of viscoelasticity in dry bone makes the bone unable to withstand as much strain or elastic deformation as wet bone and is more sensitive to degradation. This will cause dry bone to fracture immediately after the strength threshold is reached, whereas wet bone will first go through a plastic phase where the bone is deformed before fracturing [9,

18]. Therefore, the fracture patterns in dry and fresh fractures will be different [30].

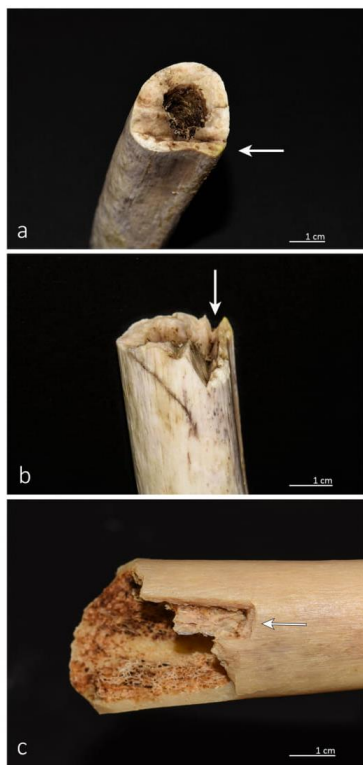
Macroscopic patterns that are used to differentiate between perimortem and postmortem fractures are a consequence of those bone biomechanics and composition characteristics. Macroscopic descriptions are preferred in fracture assessment since this is the fastest and easiest way in the field. For this reason, our study was focused on the macroscopic appearance of fractures. Colour is a reliable and known trait as any fracture after decomposition will give a different colour, which will make it possible to differentiate the old (perimortem) from the newly (postmortem) exposed bone surfaces. However, colours can change again after alteration due to acquired patina through the years. This means that, if there is no colour



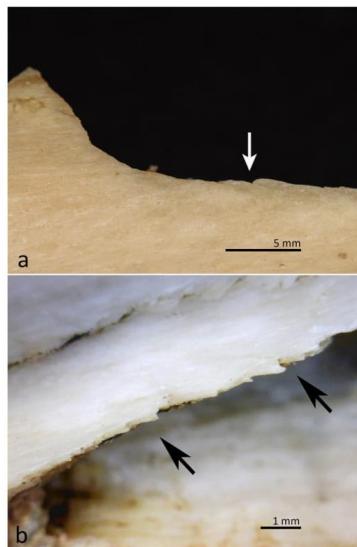


**Fig. 1** Servo-hydraulic testing machine EM2/20 MicroTest, with the U10M/25-kN loading cell and two supports to create a three-point bending, from UPC

difference, it does not necessarily mean that the fracture can only be perimortem [9, 18]. The general appearance of bone is



**Fig. 2** The white arrows indicate the layered breakage in the cortical bone, opposite to a straight tension side (a, b sample 2 and c sample 19)



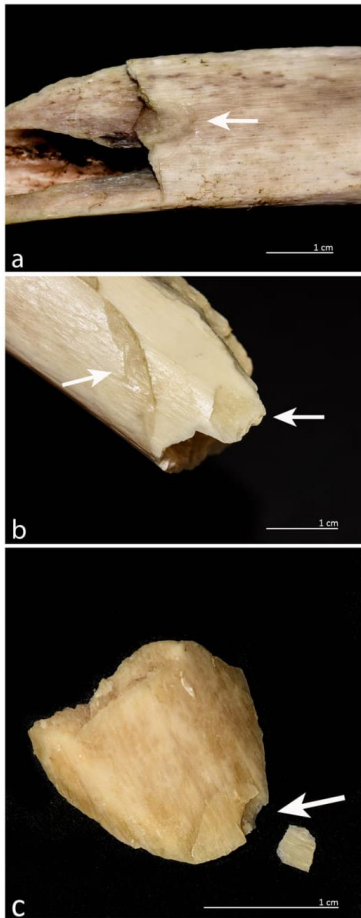
**Fig. 3** Wave lines on the smooth edges of a fracture (a sample 26 and b sample 22)

also an important trait. Dry bone is much more brittle, causing fracture lines to run along or perpendicular to the grain of the bone causing jagged edges [8, 9, 16]. Notwithstanding, the differentiation between very fresh and very dry fractures are easy; in the forensic field, most of the recovered bones are in an intermediate stage between very fresh and extremely dry.

Bones that are in an intermediate stage of drying do not meet these extreme conditions. Therefore, it would be more valuable to obtain additional knowledge about characteristics that are linked more to fresh conditions. With an experimental approach, we were able to reproduce fracture patterns similar to those found in traumatic cases and compare them to find



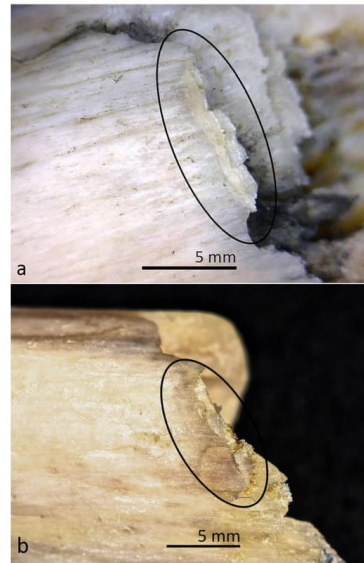
**Fig. 4** Bone scales in a compressed area of a fracture (sample 22)



**Fig. 5** a, b Several flake defects on the bone surface (samples 2 and 14, respectively) and c a piece of bone with a flake defect and matching flake from an experimentally reproduced fracture (sample E3)

specific patterns that may be used in forensic examinations. This study reports the following five distinct characteristics that can be valuable traits to utilise in the distinction between perimortem and postmortem fractures: layered breakage, bone scales, crushed margins, flakes with flake defect and wave lines. These characteristics constitute a specific, unreported perimortem pattern; however, not all characteristics must be involved in all perimortem lesions.

Our results showed that layered breakage was the most common perimortem trait, which occurred in 82% of the samples. This morphological feature occurred in almost all our



**Fig. 6** Crushed margins on the edge of a fracture in the compression side (a sample 23 and b sample 21)

fresh perimortem and postmortem fractures, and it can be considered as a specific trait of the compression side. Only in spiral fractures and fractures at the epiphyses, layered breakage could not be observed. The edges of spiral fractures are very smooth, which was the reason that this feature was not observed in this type of fracture. Nevertheless, the presence of a spiral fracture is already a sign that the fracture is most likely a perimortem fracture since this type of fracture is not likely to appear in dry bone fractures [16]. During bending, the process of fracturing will start at the tension side and



**Fig. 7** Fractured dry bone with clear irregular and brittle edge

will continue towards the compression side. In literature, it is mentioned that the tension side is regular and smooth and the compression edge is irregular [18]. However, we observed a characteristic layered pattern in the cortical area that is unreported in literature. As this trait always appeared in the compression side, it may be possible to reconstruct the biomechanical properties more precisely and deduce the direction of impact. Since layered breakage only could be reproduced in fresh postmortem fractures, it may be an important trait for fractures in fresh bones and not only limited to *intra vitam* circumstances.

The next common trait was the presence of flake defects that could be observed in 46% of the samples. Our findings on flakes are congruent with the findings previously reported by Moraitis et al. [31, 32]. They found signs of flaking on a mandible and a long bone and suggested this as a perimortem trait. In this study, we were able to reproduce flake defects and the other traits under conditions that even might suggest that these traits might be related to *intra vitam* circumstances. The other traits, bone scales, wave lines and crushed margins, were less common and were only observed in 29, 25 and 14% of the samples, respectively. Crushed margins need a very specific mechanism; this is probably the reason why they occurred infrequently. Bone scales, wave lines, flakes and crushed margins were not observed in dry bone and fresh experimental bone fractures without compression. However, we were able to reproduce bone scales, wave lines and flake (defects) when applying axial compression that seems to be necessary in order for them to be present. We therefore suggest that these distinct traits may be related to the unique micro-architecture of the bone, but more importantly, they may be relying on the presence of muscles and flesh.

One of the challenges in forensic anthropology is to find traits that are clearly related to *intra vitam* circumstances in order to shorten the time gap between very fresh and very dry fractures. In literature, there is only one trait that suggests an *intra vitam* fracture. This is when a fractured long bone shows evidence of an axial shortened displacement [25]. We hypothesise that bone scales, flakes with flake defects, crushed margins and wave lines are related to musculoskeletal activity and the presence of surrounding flesh. One of the limitations of this study was the limited sample size. Testing this hypothesis deserves further research, whereby a larger sample size should be obtained. Improving the experimental part could get further knowledge about the occurrence of the traits. Further limitations of this research are the average age of the autopsy samples, being 42 years old, versus the experimental bone samples with an average age of 75 years. However, these limitations might not all be solved as these type of studies depend on the availability of human samples.

In summary, during this early stage study, preliminary results were obtained about a new pattern with five distinct macroscopic characteristics that may be specific for fractures

in a perimortem stage. Layered breakage was the most frequent trait found. It was a specific trait that occurred in fractures when the bones were still wet. Furthermore, it was a great indicator of the direction of impact as it could always be observed in the compression side. Additionally, initial results were obtained to relate bone scales, flakes, crushed margins and wave lines to *intra vitam* circumstances, when muscles and flesh are involved. Future research will potentially improve these criteria. The fact that we possibly could shorten the time gap of time of injury by assessing morphological traits in a fracture that are specific for *intra vitam* circumstances will allow us to interpret blunt force trauma more accurately in the future.

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## **4.2. Black cracks: staining of fracture lines (2017)**

Forensic Science, Medicine and Pathology in 2017 (IF 2.027, Q1 in Legal Medicine)

### Addressed research questions

Can perimortem traits be visualised with different imaging techniques?

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## TECHNICAL REPORT

## Black cracks: staining of fracture lines

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**Abstract** During the investigation of fresh bone fractures, it might be difficult to visualize all the fracture lines that could contribute to the interpretation of the biomechanics behind a fracture. To optimize the examination of the fracture, the bones should first be defleshed to expose the osseous surface. To reveal small fracture lines more clearly, we developed two easy, fast, cheap and non-destructive methods to enhance fracture lines and bone defects by coloring the fracture lines with ink. One method consists of cooking the bone in ink, and the second method uses capillary action for ink penetration. We strongly recommend the use of the latter method with Talens® Black Indian Ink for the clearest results.

**Keywords** Forensic anthropology · Bone fractures · Ink · Biomechanics · Perimortem

### Introduction

Bone can serve as an excellent piece of evidence for forensic analysis due to its general resistance to decomposition and ability to preserve important clues that can be used for crime

reconstruction. Understanding the structure of bone and its material properties allows forensic anthropologists to reconstruct the biomechanics of trauma and gain knowledge regarding specific instruments and mechanisms related to the cause of injury. This knowledge of trauma permits these professionals to aid medical examiners in determining the cause and manner of death [1, 2].

One of the most important elements in forensic anthropology, used to reconstruct the contextual information of the blunt force trauma, is the analysis of the fracture pattern. A fracture always starts on the tension side leading to separation of osseous tissue and the generation of a fracture line, also known as tension lines [3]. Examining these lines may contribute to the understanding of the biomechanics behind a fracture and may help to infer the direction of impact. When skeletal analysis is required in order to examine these traits, the bones must first be defleshed to expose the osseous surfaces. Literature pertaining to these defleshing methods to expose the osseous surface is scarce. Furthermore there are presently no methodologies that enhance the evidentiary features of fractures without damaging the skeletal elements. Therefore, the aim of this paper is to present a method to enhance the visibility of fracture lines in blunt force trauma.

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### Materials and methods

Specimens used in this study came from autopsied bodies at the Catalanian Institute for Forensic Science and Legal Medicine (IMLCFC) that were removed for further medico-legal analysis. To deflesh the bones, the specimens were immersed in water with degreasing detergent and the solution was heated. The temperature was kept at 80–90 °C and the bones were cooked for 2–5 h, depending on the amount of flesh that had to be removed. After cooking, the remaining

flesh was carefully removed with surgical equipment and the bones were placed in a 50% acetone-water solution overnight to degrease. Afterwards, they were cleaned with water and left to dry. If the bones were very greasy, they were cooked again in a 2.5% ammonia solution until all the grease was removed [4]. This cleaning method is a safe choice for bone examination as it will have no damaging effects on the bones when applied in a controlled and stable manner [5–7].

To investigate how fracture lines could be made more visible to the naked eye, two new and non-destructive methods were tested. The first enhancing method involved the addition of 1 ml of ink to the water and detergent solution followed by the rest of the defleshing method described above. In the second method, a further ten bones were defleshed according to the method described above. When the bones were clean and dry, 1–5 drops of ink were applied onto the fracture line and capillary action<sup>1</sup> would cause the ink to diffuse through the fracture lines.

In total, ten samples were cooked with three different types of ink; Talens® Black Indian Ink, Parker® Black Ink, and Pelikan® Blue Ink.

## Results

When cooking the bones in ink, following method 1, the fracture lines were clearly more visible when the bones were cooked with Talens® Black Indian Ink (Fig. 1). Unfortunately, when cooking the bones in Pelikan® Blue Ink or Parker® Black Ink, the ink did not penetrate the fracture lines. Although Parker® Black Ink was used, it was clear when adding the ink to the water, that the base color of the ink was blue, therefore the bone appeared blue after cooking.

The results from the second method were also in favor of using Talens® Black Indian Ink. The ink easily diffused into the fracture lines resulting in the fracture lines becoming more apparent (Fig. 2). The other two types of ink (Pelikan® Blue Ink and Parker® Black Ink) did not penetrate the fracture lines as well as the Talens® Black Indian Ink did and caused permanent spotting on the bone while the Talens® Black Indian Ink could be washed out to a great extent.

## Discussion

The analysis of bone fractures can give important information about the biomechanics of trauma. Direct assessment of tension lines or small fracture lines, that are not visible on x-rays,

<sup>1</sup> Definition 'Capillary action' according to the Environmental Engineering Dictionary: the means by which liquid moves through porous spaces in a solid due to forces of adhesion, cohesion and surface action. [10]



**Fig. 1** Upper bone cooked without ink, bone below cooked with Talens® Black Indian Ink where the fracture lines are more visible

incorporate confidence in trauma evaluation, allowing to reconstruct the impact point and direction of trauma [3]. Nevertheless, even after defleshing the bones, these small fracture lines may not be clear to the naked eye or may not be visible on pictures when they are essential in presentations in the courtroom or teaching classes.

Porta also emphasized the importance of the assessment of fractures to get further knowledge and to complement investigations in traffic accidents. There is mention of a technique which can enhance fractures to allow for clearer examination which consists of immersing a cleaned bone in a bleach solution for some minutes and then let the bone dry in an oven. This treatment would cause the fracture to open [3]. However, during fracture examination, it is crucial that the bone and fractures are not damaged in any way and original features are kept preserved. For this reason, we preferred not to use this method because bleach is a corrosive reagent that could damage the fracture and even after drying it will continue to degrade bone [5, 8, 9].

In this technical report, we present two new, simple, cheap and non-destructive methods to enhance perimortem fracture lines after defleshing the bones without damaging them. This preparation improves the assessment of bone injuries in order to facilitate the assessment of the biomechanics behind a fracture, and to improve their visibility for high quality photographic presentations. Both methods use ink penetration into the lesion and offer both an improvement of visual recognition of the existing fractures. We strongly recommend the method that depends on capillary action for ink diffusion. The fracture lines were better visualized since the color penetrates only the fracture lines. The results of the cooking method were not always satisfactory and were dependent on the condition of the bones. When the bones were very greasy, the ink

**Fig. 2** Visible difference of fracture lines, before (*left*) and after (*right*) adding some drops of Talens® Black Indian Ink to the fracture benefiting capillary action



did not enter the fracture lines, in contrast to the other method that relied on capillary action where this was not an issue. Literature demonstrated that cooking a bone in controlled circumstances does no damage at a histological level. Further research, however, should be done to confirm that ink does not damage the bone on a microscopic level either. Nevertheless, this method could be used during the examination of fracture lines in bone fractures and will aid in the understanding and interpretation of the biomechanics behind a fracture. Furthermore, it can be used during court sessions to present photographic evidence to a jury as well as in class for educational purposes. With this technique we show that efficiency is a by-product of simplicity.

### Key points

1. Examining fracture lines may contribute to the understanding of the biomechanics behind a fracture.
2. There are no methodologies that enhance the evidentiary features of fractures without damaging the skeletal elements.
3. This research presents two new, simple, cheap and non-destructive coloring methods to enhance perimortem fracture lines after defleshing the bones.

4. It can be used during court sessions to present photographic evidence to a jury as well as in class for educational purposes.
5. Best results are obtained using Talens® Black Indian Ink with the method that relies on capillary action.

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### **4.3. Perimortem fracture pattern in ribs by blunt force trauma (2018)**

International Journal of Legal Medicine in 2018 (IF 2.094, Q1 in Legal Medicine)

Addressed research questions:

What are the macroscopic characteristics that define a distinct perimortem pattern in wet long bone fractures, caused by blunt force trauma?

Can certain characteristics be related to certain trauma circumstances and therefore deduce the biomechanical circumstances?

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## Perimortem fracture pattern in ribs by blunt force trauma

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### Abstract

Literature on timing of rib trauma is scarce but remains challenging during forensic cases. This study analysed the macroscopic fracture patterns of perimortem rib fractures and compared them to experimentally reproduced rib fractures on fresh and dry ribs. Six distinctive macroscopic traits were found in ribs that might provide information about the timing of trauma, fracture mechanism and/or trauma circumstances. These traits are peels, folds, differential fracture edges, incomplete fractures, plastic deformation and longitudinal lines. Peels, folds and plastic deformation might provide information about trauma timing. Folds and different fracture edges might provide information about the fracture mechanism. Statistical analyses showed that longitudinal lines, folds and incomplete fractures might provide information about the trauma circumstances and that age might have an influence on the occurrence of complete fractures, longitudinal lines and peels ( $p \leq 0.05$ ). The new insights presented in this study might be valuable for forensic anthropologists in rib trauma analysis.

**Keywords** Forensic anthropology · Rib fractures · Ribs · Biomechanics · Perimortem · Traits

### Introduction

As contributions by forensic anthropologists to medicolegal investigations become more frequent, multidisciplinary

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approaches to forensic investigations are promoted. The mechanical properties of bone allow anthropologists to reconstruct the biomechanics of trauma and to derive knowledge about specific instruments and mechanisms related to the injury. Despite the fact that forensic anthropology has been traditionally associated with skeletonised remains, their biomechanical knowledge allows forensic anthropologists to supplement the work of forensic pathologists when assessing bone injuries in badly decomposed bodies and fresh cadavers [1–3].

In addition to trauma reconstruction, the forensic anthropologist must determine the time frame in which skeletal trauma was inflicted when undertaking trauma analysis. One of the main challenges during the interpretation of skeletal trauma remains the determination of whether trauma occurred during or after death (perimortem or postmortem, respectively). Generally, anthropologists define perimortem trauma as trauma that was inflicted when the bone still has its organic components causing it to behave as fresh bone [4]. Although there are some known morphological or possible colour differences, the differentiation between perimortem trauma and postmortem taphonomic factors remains difficult [5–8].

Literature pertaining to the determination of fracture occurrence as being either antemortem, perimortem, or postmortem is focussed on long bones. There is little to no knowledge regarding other types of bones that are commonly related to

forensic cases, such as ribs. Ribs and long bones have different architecture, resulting in different fracture patterns. It is difficult to apply the known perimortem characteristics due to the structure of the rib and the thin cortical layer. Even though rib fractures are very common in forensic cases, they receive very little attention, and only a few studies have been performed on rib fracture biomechanics [9–14]. Furthermore, studies that have been performed are contradictory and currently no fracture pattern is reported to distinguish perimortem rib fractures from postmortem rib fractures [10, 11]. Further research is required to increase confidence during rib trauma evaluation, primarily to reconstruct the contextual information of the injury and to differentiate them from taphonomical origins.

Based on an empirical demonstration on the variability of rib fracture patterns with respect to loading conditions, this study aims to provide further insight into the characteristics of a perimortem rib injury characterizing morphological traits of rib fractures, defining how these characteristics are related to certain trauma circumstances and whether this pattern allows us to optimize the distinction between perimortem and postmortem fractures. An enhanced understanding of the trauma mechanisms and rib response is potentially valuable for timing and reconstructing traumatic events accounting for rib fractures in forensic contexts.

## Materials and methods

### Sample

This study uses both perimortem fractured and healthy unfractured rib samples. In total, 151 anterolateral fractured rib specimens from 25 cases are being used that come from forensic autopsies at the Institute for Legal Medicine and Forensic Science of Catalonia (IMLCFC), which were removed for complementary medico-legal investigation of the trauma (Table 1). The trauma circumstances of the rib fractures were cardiopulmonary resuscitation (CPR, either manual or mechanical<sup>1</sup>), traffic accidents, falls and train collisions. In this study, they will be divided into two groups: CPR ( $n = 61$ ) and accidents ( $n = 90$ ).

The perimortem fracture patterns of autopsy rib samples were compared with the pattern exhibited by experimentally fractured ribs. The Medical Anatomy Department of the Autonomous University of Barcelona (UAB) provided 30 healthy, unfractured, fresh ( $n = 18$ ) and dry ( $n = 12$ ) anterolateral samples of 4th–6th ribs from 15 adults that donated their

**Table 1** Overview of available perimortem rib fractures obtained from medico-legal autopsies per case

Case	Sex	Age	Trauma circumstances	Fractures
R01	Female	52	Fall	2
R02	Male	62	CPR (mechanical)	4
R03	Male	42	Collision with train	10
R04	Male	49	CPR (manual)	7
R05	Female	70	Fall	8
R06	Male	87	CPR (manual)	3
R07	Male	72	CPR (mechanical)	2
R08	Male	52	CPR (mechanical)	1
R09	Male	75	CPR (mechanical)	5
R10	Male	74	CPR (mechanical)	8
R11	Male	63	CPR (manual)	3
R12	Male	28	CPR (mechanical)	2
R13	Male	59	CPR (manual)	3
R14	Male	75	CPR (mechanical)	2
R15	Male	54	CPR (manual)	5
R16	Male	52	Collision with train	5
R17	Male	62	Collision with train	9
R18	Female	62	CPR (manual)	8
R19	Male	82	CPR (mechanical)	5
R20	Male	40	Collision with train	20
R21	Male	21	Collision with train	12
R22	Male	76	CPR (mechanical)	4
R23	Male	50	Fall	3
R24	Male	40	Collision with train	8
R25	Male	30	Collision with train	12
Total				151

bodies to science (Table 2). This study was approved by the Ethic Commission of Human and Animal Experimental Work (CEEAH) of the UAB, in compliance with the ethical regulations. Before experimental analysis, the fresh ribs from UAB were defleshed in such a way that only the periosteum and a thin layer of flesh were still attached to the bone.

### Fracture reproduction

Ribs provided by the UAB were used to experimentally recreate anterolateral fractures that were compared with samples obtained from autopsies. Three-point bending tests were conducted in an experimental and controlled setting to inflict slow and fast loading trauma.

A servo-hydraulic testing machine (EM2/20 MicroTest) from research group GRABI of the Polytechnic University of Catalunya (UPC) was used with an U10M/25-kN loading cell to simulate a slow loading. The test was controlled through the SCM 3000 program. This low-speed bending test was performed on the external part of six dry and seven fresh

<sup>1</sup> Mechanical CPR is performed by a chest compression system called LUCAS™ [25, 26]. This device consists of a battery-driven suction cup and pressure pad to deliver uninterrupted compressions at a consistent rate and depth to facilitate the return of spontaneous blood circulation.



**Table 2** Overview of available ribs from donors provided by UAB and experimental characteristics

Case	Rib condition	Ribs	Sex	Age	Speed of impact
Exp_R01	Dry	6	Male	63	160 mm/min
Exp_R02	Fresh	1	Male	45	160 mm/min
Exp_R03	Fresh	1	Male	48	160 mm/min
Exp_R04	Fresh	1	Male	57	160 mm/min
Exp_R05	Fresh	2	Female	45	160 mm/min
Exp_R06	Fresh	1	Male	27	160 mm/min
Exp_R07	Fresh	1	Male	35	160 mm/min
Exp_R08	Fresh	4	Male	61	3 m/s
Exp_R09	Fresh	2	Female	94	3 m/s
Exp_R10	Fresh	1	Male	68	3 m/s
Exp_R11	Dry	6	Male	65	3 m/s
Exp_R12	Fresh	1	Male	48	160 mm/min, internal part
Exp_R13	Fresh	1	Female	45	160 mm/min, internal part
Exp_R14	Fresh	1	Male	27	160 mm/min, internal part
Exp_R15	Fresh	1	Male	35	160 mm/min, internal part

human anterolateral rib samples at a strain rate of 160 mm/min. Compression was exerted on the external and tension on the interior side of the rib. Additionally, four fresh rib samples were fractured with the loading cell exerting pressure on the internal side of the rib.

To reproduce fractures caused by a high impact, a devised drop test machine was used that was connected to an oscilloscope, which was, in turn, connected to a computer to measure the velocity of the drop. This high-speed bending test was performed on six dry and seven fresh ribs at a strain rate of 3 m/s with the loading cell facing the external side of the rib.

In both experiments, a rubber coating was attached to the loading cells as an attempt to simulate soft tissue and to prevent direct contact of the metal loading cell onto the ribs.

### Defleshing and macroscopic examination

The fresh rib fractures resulting from the experiments were defleshed by cooking them in a water and detergent solution (one cup of commercial degreasing detergent in 5 L of water). After the water reached 100 °C, the temperature was lowered to 90 °C and the bones were cooked for 2–3 h. After cooking, the remaining flesh was removed, using surgical tools, and the ribs were cleaned with water and left to dry. This method exposes the osseous surfaces to make the ribs suitable for skeletal examination.

### Statistical analyses

Binary logistic regression was carried out to establish whether there were statistically significant associations between fracture pattern, trauma mechanism, sex and age. The odds ratios (ORs) were adjusted for cases. Moreover, the percentages of

the traits for each individual case were calculated and compared between groups of traumas caused by CPR and accidents using a Mann-Whitney test. *p* values  $\leq 0.05$  were considered significant. The statistical analyses were performed in IBM SPSS Statistics software version 23.

## Results

Table 3 gives an overview of the cases and fractures categorised by age, sex and trauma circumstances. The results indicate the highest proportion of fractures within the categories of male, senile age and accidents (traffic accidents, falls and train collisions).

During macroscopic examination of the autopsy specimens, six distinct characteristics were observed which might define a perimortem pattern: incomplete fractures, plastic deformation, peels, folds, longitudinal lines and different

**Table 3** Overview of sample distribution of the rib samples and the amount of cases and fractures within the sample set

		Cases	Fractures
Age	Young adults (20–40 years)	5	54
	Mature (41–60 years)	8	36
	Senile (>60 years)	12	61
Sex	Female	3	14
	Male	22	137
Trauma circumstances	Manual CPR	6	26
	Mechanical CPR	9	32
	Accidents	10	93

appearance of the internal and external fracture edges. However, not all traits are necessarily continuously present.

The first distinct characteristic, a peel (Fig. 1), is a structure of the fracture in which the cortex on one part of the rib fracture is 'peeled off' but still attached. This results in a superficial peel defect on one part of the rib fracture and a thin, peel-like structure on the other part of the rib fracture.

Another remarkable trait is a fold (Fig. 2). Folds are depressed cortical fragments close to the fracture edge, and perpendicular to the axis of the rib.

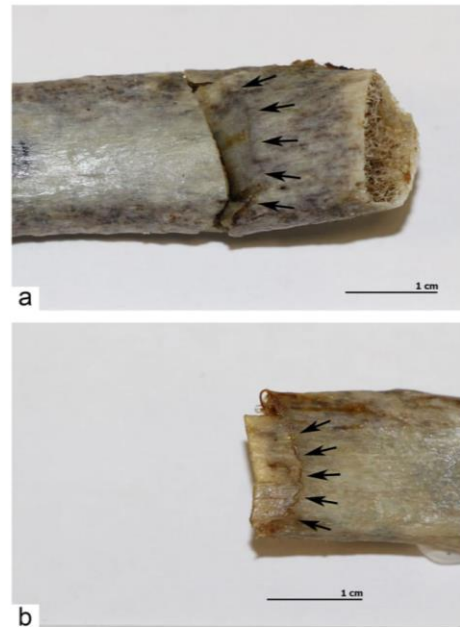
Longitudinal lines are often visualised in fractured ribs (Fig. 3). These are longitudinal cracks that follow the bone axis, frequently located at the internal (pleural) side of the rib.

Among our samples, many incomplete fractures were noted (Fig. 4). Incomplete fractures are partial fractures, which only involve either the outer or inner cortex. Plastic deformation often occurs along with this trait (Figs. 4 and 5) which is characterised by permanent deformation of the bone.

The final trait called 'fracture edges' (Fig. 6) defines that there is a difference between the internal and external sides of complete rib fractures. It applies to fractures that on one side are very irregular and in some cases crushed, while the other side is smooth and straight. This trait is independent of the



**Fig. 1** The right part of the fracture shows the thin peel seen from an anterior (a) and posterior (b) side. The left part of the fracture shows the peel defect

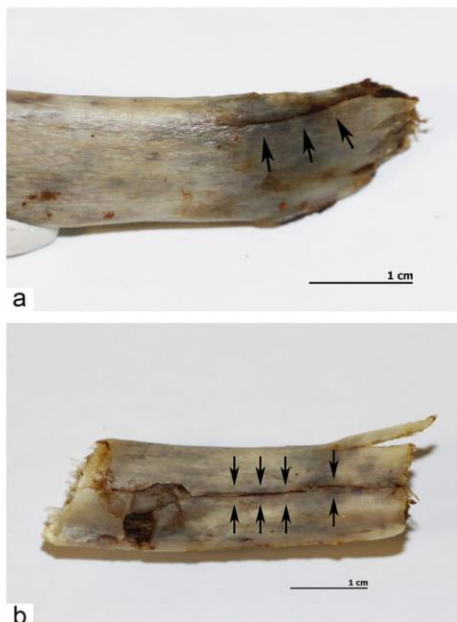


**Fig. 2** Two examples of a fold that is often encountered in rib fractures

morphology of the fracture and only applies to the edges of a fracture. In all experimentally fractured samples in which this trait could be observed, the compression side showed an irregular and crushed edge, while the tension side presented a straighter and smoother edge.

Plastic deformation and different fracture edges on the internal and external sides were found in more than 50% of the perimortem fractures, namely 57.6 and 51.7%, respectively. These traits are followed by peels (33.1%), longitudinal lines (21.2%), folds (16.6%) and incomplete fractures (15.6%).

The results of the binary logistic regression in the analysis of the fracture pattern in ribs show that young adults were statistically significant more likely to get peels compared to mature individuals (OR = 0.09, 95% CI [0.017–0.46],  $p = 0.004$ ) and senile individuals (OR = 0.25, 95% CI [0.074–0.82],  $p = 0.02$ ). Moreover, young adults were statistically significant more likely to get longitudinal lines compared to mature individuals (OR = 0.093, 95% CI [0.013–0.68],  $p = 0.02$ ) and senile individuals (OR = 0.12, 95% CI [0.024–0.56],  $p = 0.007$ ). The risk of having complete fractures depended on both age and trauma circumstances. Thus, senile people were 9.6 times more likely to have complete rib fractures compared to young adults (95% CI [1.4–64.2],  $p = 0.02$ ). Additionally, rib fractures that were caused by accidents were 7.3 times more likely to be complete fractures compared to rib fractures



**Fig. 3** Longitudinal lines on the lateral (a) and internal (b) parts in a fractured rib



**Fig. 4** Two examples of incomplete fractures, both with plastic deformation present

that were caused by manual CPR (95% CI [1.1–48.7],  $p = 0.04$ ). No statistically significant associations could be found between the presence of folds and plastic deformation when comparing groups categorised by trauma circumstances, age and sex ( $p > 0.05$ ). Furthermore, no significant differences were found between the injury pattern of manual and mechanical CPR.

Table 4 shows the results of the Mann-Whitney test comparing trauma caused by CPR and accidents in the autopsy samples. Rib fractures caused by accidents are more likely to result in folds and longitudinal lines compared to rib fractures caused by CPR. No significant differences could be found for the other traits.

The fresh ribs, experimentally fractured under fast loading, were all completely fractured and showed, to a small extent, all the traits except for peels that could not be reproduced. All dry samples showed complete fractures, and no peels, folds, or plastic deformation could be observed at fast or low speed. Furthermore, in both tests, the dry ribs did show longitudinal lines and differences in fracture edge appearance between the internal and external sides of the rib. The fresh ribs fractured under slow loading with the impact on both the internal and

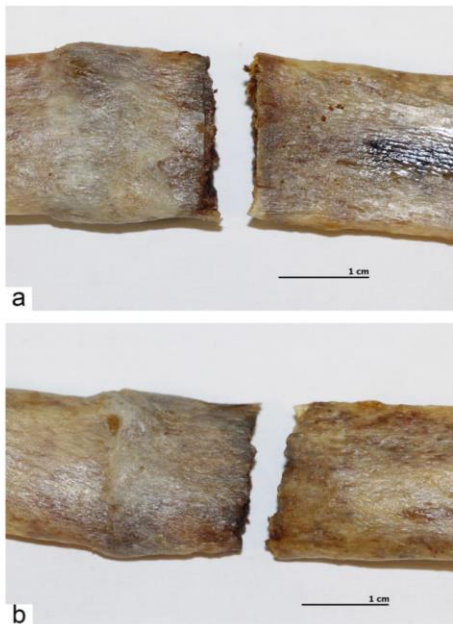
external sides were all incomplete fractures and only showed folds and plastic deformation (Table 5).

The experiments carried out on the internal part of the rib ( $n = 4$ ) are not included in the overview of the experimental samples as they were carried out to observe which side of the rib would break first: the compression or tension side.



**Fig. 5** Plastic deformation on a fractured rib; also, a longitudinal line can be observed





**Fig. 6** a The internal, smooth and straight edge of the rib fracture while the external side of the rib (b) has a more irregular and crushed appearance

## Discussion

Rib fractures are commonly involved in forensic cases such as traffic accidents, falls and cases involving blows to the chest [9, 11, 14]. Nevertheless, biomechanics of rib fractures are poorly understood; therefore, the interpretation of rib fractures remains a challenge for forensic anthropologists [10, 13]. Ribs are part of a closed structural and mechanical system, with a distinct architecture different from long bones. Experimental

research to fractured rib patterns with respect to loading conditions are scarce, probably due to the difficulties in developing a method to systematically analyse the injury patterns, and the difficulty in obtaining human rib samples [11].

This study provides preliminary knowledge on the morphology of the injury pattern of the anterior rib arch. This region, together with the lateral part of the rib, is frequently associated with direct blows to the chest, lateral compaction of the thorax or cardiopulmonary resuscitation (CPR) [15]. This study reports six preliminary macroscopic traits present in autopsy fracture samples of the anterolateral rib arch: peels, folds, longitudinal lines, incomplete fractures, differential fracture edges on the internal and external part of the rib fracture and plastic deformation. These characteristics constitute a specific, unreported perimortem pattern; however, not all characteristics must be present at the same time in all rib fractures.

On the other hand, experimental analysis used in this work might provide further knowledge about the differentiation between perimortem and postmortem trauma. Timing rib fractures still is an uncertain diagnostic in most forensic anthropological reports. In spite of published descriptions to differentiate perimortem from postmortem fractures, they are mainly intended for long bones [3, 5, 16, 17]. Given the differential architecture and structure between long bones and ribs, these known rules are difficult to apply. Therefore, it is more valuable to obtain additional knowledge about perimortem fractured ribs, since it could be the base to approach this challenge. With this purpose, the abovementioned perimortem rib fracture pattern was compared with the fracture pattern of experimentally fractured ribs from UAB donors. Three-point bending trauma tests were simulated on fresh and dry anterolateral individual rib samples. The variability of rib fracture patterns was analysed with respect to two loading conditions, fast speed (3 m/s) and slow speed (160 mm/min).

The results show that incomplete fractures, plastic deformation, peels and folds are not present in dry bones, independent to the loading conditions. It suggests that these traits are related with fresh/wet bone conditions or perimortem conditions. In contrast, longitudinal fractures and differential appearance of the margins appear in both fresh and dry specimens, excluding them as perimortem indicators. The differences may be explained by differences in bone composition between wet and dry bone, which influences the fracture biomechanics and thus the fracture patterns. Considering the nature of incomplete fractures, plastic deformation, peels and folds, it is likely that water and organic material are required to reproduce them in a rib fracture [14, 18, 19].

The results also show that this pattern is related with age and trauma biomechanics, giving clues to reconstruct the trauma circumstances. Evidence concerning the influence of the age in rib fracture pattern seems to be varying in literature [10]. According with our results, longitudinal fracture lines

**Table 4** Results of the Mann-Whitney test

	Mean% CPR	Mean% accidents	<i>p</i> value
Peels	17.5	32.4	0.3
Folds	3.1	23.7	0.05 <sup>a</sup>
Longitudinal lines	2.5	27.8	0.002 <sup>a</sup>
Plastic deformation	40.3	61.7	0.09
Complete fractures	76.9	85.1	0.5
Different fracture edges	52.9	50.1	0.8

<sup>a</sup> Statistically significant results

**Table 5** Overview of experimental samples and their characteristics

Rib condition	Slow		Fast	
	Dry ( <i>n</i> = 6)	Fresh ( <i>n</i> = 7)	Dry ( <i>n</i> = 6)	Fresh ( <i>n</i> = 7)
Incomplete fractures, <i>n</i> (%)	0 (0.0%)	7 (100%)	0 (0.0%)	0 (0.0%)
Plastic deformation, <i>n</i> (%)	0 (0.0%)	5 (71.4%)	0 (0.0%)	1 (14.3%)
Peel, <i>n</i> (%)	0 (0.0%)	0 (0.0%)	0 (0.0%)	0 (0.0%)
Fold, <i>n</i> (%)	0 (0.0%)	6 (85.7%)	0 (0.0%)	1 (14.3%)
Longitudinal lines, <i>n</i> (%)	4 (66.7%)	0 (0.0%)	4 (66.7%)	3 (42.9%)
Internal/external different appearance, <i>n</i> (%)	5 (83.3%)	0 (0.0%)	5 (83.3%)	1 (14.3%)

and peels on the cortical rib surface have been found more probable in the young age group. Additionally, there is a statistically significant increased risk of getting complete rib fractures in older individuals compared to younger individuals. This evidence is in contrast to Love and Symes [10], who did not find major incidence of complete rib fractures between various age categories. Considering our observations, however, chronological age influences the mechanical behaviour of bone as originally described in literature. Therefore, we hypothesize that the relationship between complete fractures, longitudinal fracture lines, peels and age may be explained by aging changes in the architecture and composition of the bone, particularly since bone is more elastic in young people and brittle in the elderly [5, 6, 8, 9, 20, 21].

Moreover, the results suggest that some of the macroscopic traits associated with rib fractures may be indicators of the trauma circumstances. The analysis of these traits shows that complete fractures, folds and longitudinal fracture lines appeared to be more likely to be present in accidents caused by falls, car accidents or train collisions than injuries induced from CPR.

Additionally, folds and differences in fracture edges might provide information about the fracture mechanism. Folds were not reproducible on dry ribs, and in the experimentally fractured fresh ribs folds could only be observed on the compression side. Furthermore, experimental reproductions on dry and fresh ribs under fast loading conditions showed differences in the fracture edges between the compression and tension sides: the compression side appeared irregular, whereas the tension side appeared smooth and regular [22]. This might suggest that these traits could indicate the direction of impact. The smooth fracture edge at the tension side was earlier described by Kieser et al. in a study with 15 pig ribs. However, they did not describe the irregular compression side and could not find this trait in fractured dry bones. Moreover, no implications were provided by them on how this trait could help to interpret the fracture pattern in relation to the fracture mechanism during forensic trauma analysis [12]. This study uses a large sample of human bones instead of animal bone samples which makes this study

innovative and unique. We could find this trait in 78 of our human perimortem rib samples and provide an indication of the forensic implication of this trait, which takes the study of Kieser et al. a step further.

All experimentally fractured fresh ribs that were broken under slow loading conditions were incomplete fractures with only a fracture in the cortex of the compression side. Since the cortex on the tension side was intact in all fresh specimens fractured with slow loading, no distinction could be made between the fracture edges of the compression and tension sides.

Literature regarding the biomechanical properties of bone state that bone is usually stronger under compressive stress compared to tensile stress, which means that bone usually fractures at the tension side prior to the compression side [6, 20, 23]. However, in our study all experimentally fractured fresh rib samples, including the fresh ribs that have been fractured with the loading cell on the internal part of the rib, failed at the compression side prior to the tension side. Previous studies have already concluded this, and incomplete fractures that failed at the compression side prior to the tension side were identified as 'buckle fractures' [12–14]. The finding that ribs also fractured first at compression when the loading cell was put on the interior part of the rib might suggest that ribs always fracture first at compression, regardless of the location of the compressive stress on the rib. In order to obtain further knowledge and to test this hypothesis, future research is recommended with a larger sample size and improved experimental settings in which real trauma conditions can be simulated.

Lastly, peels did not appear in any of the experiments, independently of the rib state and loading conditions. This trait was only observed in autopsy specimens. Intriguingly, the tests were not able to reproduce peels, even in fresh and fast loading conditions. It suggests that its presence may be related to the trauma circumstances, which are difficult to reproduce in a simplified *in vitro* experimental test. In the case of ribs, it is difficult to link the traits with *intra vitam* conditions such as muscular contraction, as hypothesised in some of the traits observed in long bones [9, 10, 24], but the importance of soft tissue and the periosteum should not

be discarded. Analysis of peels deserves further research to obtain more in-depth knowledge about the exact mechanism that is needed for its occurrence.

## Conclusion

Determining the timing and trauma circumstances of skeletal trauma is very important. This study provided further insight into the characteristics of the perimortem fracture pattern of blunt force trauma on ribs.

Six distinct macroscopic traits were observed that might provide information about rib trauma timing, circumstances and/or mechanism: plastic deformation, peels, folds, incomplete fractures, differential fracture edges and longitudinal lines. The presence of peels, folds and plastic deformation suggests that a rib was fractured in perimortem conditions. Moreover, peels, incomplete fractures and longitudinal lines might be related to the age of an individual and might provide further insight into the influence of aging on fracture biomechanics and the fracture pattern. Incomplete fractures and longitudinal lines might also provide information about the impact energy and thereby about the trauma circumstances, and differential fractures edges and folds might indicate the direction of impact and thereby provide information about the fracture mechanism.

The new insights on the fracture patterns of ribs might be of great value for forensic anthropologists in blunt force skeletal trauma analysis. Testing the influences of variables such as age, sex, the presence of flesh, etc., on the fracture pattern accurately and reliably deserves further research. Improving the experimental setting and obtaining a more homogeneous sample might allow us to test the functional implications of these traits in more depth in the future.

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## Compliance with ethical standards

This study was approved by the Ethic Commission of Human and Animal Experimental Work (CEEAH) of the UAB, in compliance with the ethical regulations.

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#### **4.4. *Intra vitam* trauma pattern: changing the paradigm of forensic anthropology? (2018)**

International Journal of Legal Medicine in 2018 (IF 2.094, Q1 in Legal Medicine)

Addressed research questions:

What are the macroscopic characteristics that define a distinct perimortem pattern in wet long bone fractures, caused by blunt force trauma?

Can certain characteristics be related to certain trauma circumstances and therefore deduce the biomechanical circumstances?

Can the anthropological paradigm be changed by the presence of *intra vitam* traits?

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## Intra vitam trauma pattern: changing the paradigm of forensic anthropology?

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### Abstract

This study aims to improve a previous study that reported new traits to characterize a perimortem fracture pattern in human long bones. This second study aims to acquire further knowledge about these perimortem traits, specifically by improving the experimental setting—by using a *Blunt Force Trauma Simulator*—and increasing the sample size with a total of 43 autopsy specimens and 57 reproduced fractures. Additionally, we investigated whether these traits could be related to muscular contractions by adding axial compression in the experimentally fractured specimens. If *intra vitam* traits can be found, it would consequentially be more valuable for forensic anthropologists to shorten the perimortem period. We demonstrate that all traits are perimortem traits. Furthermore, based on our results, we see the tendency that the combination of traits—instead of the presence of each trait individually—may make it possible to distinguish *intra vitam* from perimortem fractures. This study confirms these distinct characteristics that can be valuable to utilize in the distinction between peri- and postmortem fractures.

**Keywords** Forensic anthropology · Bone trauma · Time of injury · Perimortem trauma · Traits

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### Introduction

One of the jobs of the forensic anthropologist is identifying the time of injury, be it antemortem, perimortem, or postmortem trauma [1]. The differentiation between perimortem trauma and postmortem taphonomic factors—such as geological, biological, or (un)intentional human alterations—however, remains difficult during the interpretation of skeletal remains [2–6].

In a recent paper, Scheirs et al. [7] were able to reproduce certain fracture patterns in human bone samples; these were similar to those found in traumatic cases involving blunt force trauma [7]. Consequentially, the authors reported five new traits that characterize a perimortem fracture pattern in human long bones: the presence of layered breakage, crushed margins, bone scales, wave lines, and flakes with corresponding flake defects. The presence of one or several of these traits is described as a perimortem fracture pattern. One of the new findings suggested by these authors is that wave lines, bone scales, flakes, and crushed margins may be related to *intra vitam* circumstances, subsequently shortening the perimortem time gap. The *intra vitam* period is the moment at the time of death, when soft tissue and muscles are involved. Evaluating

these morphological features in a fracture will allow us to interpret blunt force trauma more accurately.

Scheirs' preliminary research was unique, as only human bone samples were used for their experimental analysis. Nevertheless, the research had several limitations, many related to the experimental set-up. One limitation being that the bones were mechanically fractured with a slow servo-hydraulic testing machine (9.6 m/s) may cause a disrupted outcome where in reality, bones would usually fracture rapidly on impact by means of blunt force trauma (i.e., car accident, train collision, fall, etc.). Furthermore, only 9 mechanically fractured bones and 28 fractured autopsy samples were used for comparison and to establish the pattern [7].

This second study aims to acquire further knowledge about these perimortem traits, specifically by improving the experimental setting and increasing the sample size. Additionally, it will be further explored whether wave lines, bone scales, flakes, and crushed margins could be related to the effect of muscular contraction. If *intra vitam* traits can be identified, it would consequently be more valuable for forensic anthropologists to shorten the perimortem period, to then decide whether a fracture occurred closer to the time of death, since current characterization is relational to the postmortem time period.

## Material and methods

### Samples

This study uses both perimortem fractured and healthy unfractured bone samples. In total, 43 fractured specimens from 21 cases were used with a postmortem time interval (PMI) between 12 and 24 h (Table 1). These specimens were obtained from forensic autopsies at the Institute for Legal Medicine and Forensic Science of Catalonia (IMLCFC), which were removed for complementary medico-legal investigation. In order to correctly compare autopsy samples with the experimental samples, only cases with victims of the age of 55 years or older were included in the study.

For experimental purposes, the Medical Anatomy Department of the Autonomous University of Barcelona (UAB) provided 57 healthy, unfractured, fresh ( $n = 33$ ), and dry bones ( $n = 24$ ) from people that donated their bodies to science (Table 1). The fresh bones had a PMI of 1–3 days and were dissected from the donor's limbs, removing all muscular

mass and attachments in such manner that only the periosteum was left on the bones.

This study was approved by the Ethics Commission of Human and Animal Experimental Work (CEEAH) of the UAB, in compliance with the ethical regulations.

### Fracture reproduction

A pendulum impact test machine, the *Blunt Force Trauma Simulator (BFT Simulator)*, was designed and custom-made to experimentally reproduce fractures in the 57 dry and fresh bones (UAB). The machine consists of a metal frame and pendulum to which hammers of different weights can be attached (Fig. 1). The set is loosely based on the Charpy impact test [8–10].

Hammers of 5.6 kg and 8.0 kg were available for inclusion in the study and could be physically switched when appropriate. The bone sample was placed horizontally and attached with tie-wraps to the two movable metal holders, with the anterior side facing the hammer. The bones were hit in the middle of the anterior side of the bone from a 90° angle. Additionally, a piece of soft rubber was attached to the hammer to avoid direct contact with the bone and to provide soft tissue simulation.

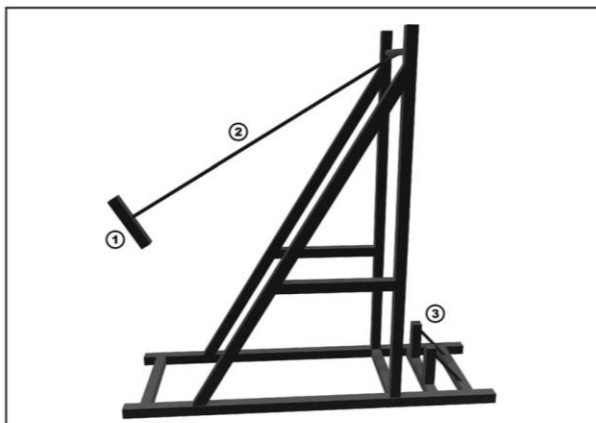
We hypothesize that soft tissue and especially muscular contraction may have an influence on the occurrence of the traits when a bone fractures *in vivo*. A way to approach this effect on our experimental samples is to apply axial compression by a combination of metal tensors and springs attached to the diaphysis of the bone samples. A similar method was already used previously by Scheirs et al. [7]. In the current study, however, the setting was improved by adding a spring between the tensors, and this sought to enhance the effect of muscle contraction, while the bone fractures. From the 33 donor bones, 18 bones were broken without compression and 15 specimens were broken with axial compression. The maximum amount of compression exerted to the bone varied between 12 and 19 kg, for upper and lower limbs (except the fibula), respectively, measured with a dynamometer. Dry bone samples were all broken without added compression, as no muscle compression can be expected in dry, postmortem bone fractures.

The velocity in which the hammer impacts the bones depends on the angle at which it is released. The energy on the other hand depends on the weight of the hammer. The

**Table 1** Descriptive table of all available samples

	Humerus	Ulna	Radius	Femur	Tibia	Fibula	Total	Age range
Autopsy samples	10	5	5	11	6	6	43	55–91
Exp. samples (wet bones)	8	6	6	5	4	4	33	56–90
Exp. samples (dry bones)	3	0	2	9	6	4	24	59–94

**Fig. 1** Schematic drawing of the *BFT Simulator* impact test machine with removable hammer (1), 1.51-m long arm (2), and adjustable holders for the sample (3)



corresponding speed and energy of the impact were calculated as follows:

$$v = \sqrt{2g \cdot L \cdot (1 - \cos\theta)} = \sqrt{2 \cdot 9,81 \frac{m}{s^2} \cdot 1,51m \cdot (1 - \cos 90^\circ)}$$

$$= 5,44 \frac{m}{s} = 19,6 \frac{km}{h}$$

$$E_{5,6 \text{ kg}} = \frac{m \cdot v^2}{2} = \frac{5,6 \text{ kg} \cdot (5,44 \frac{m}{s})^2}{2} = 82,9 \text{ J}$$

$$E_{8,0 \text{ kg}} = \frac{m \cdot v^2}{2} = \frac{8,0 \text{ kg} \cdot (5,44 \frac{m}{s})^2}{2} = 118,4 \text{ J}$$

$v$  velocity at the bottom of the pendulum  
 $L$  length of the pendulum  
 $\theta$  angle from the pendulum to the vertical beam  
 $g$  gravitational acceleration ( $9,81 \text{ m/s}^2$ )  
 $m$  mass of the hammer (5,6 kg and 8,0 kg, respectively)  
 $E$  system energy

### Bone preparation and examination

Fractured experimental specimens and autopsy specimens were defleshed and cleaned for fracture examination. The samples were cooked in a water detergent solution (one cup of commercial degreasing detergent in 5 L of water). When the water reached  $100^\circ\text{C}$ , the temperature was lowered to  $90^\circ\text{C}$  to avoid damage to the bones and their characteristics. After 2–5 h of cooking, the remaining soft tissue was removed using surgical tools and were cleaned with water and left to dry.

Bone examination was conducted to establish both the type of fracture and the presence of any traits. The bone fractures were classified as oblique, butterfly, transverse, spiral, or

comminuted fractures. The examined traits include the five perimortem traits plus the presence of plastic deformation, since this is also a good parameter for fresh bone fractures [11–13]. Most traits can be macroscopically examined by the naked eye. To examine the presence of wave lines, however, a stereomicroscope ( $\times 40$  magnification) was used.

### Statistical analysis

Logistic regression was performed to assess the relationship between the occurrence of any trait and the sample type, i.e., autopsy and experimental samples with and without compression. We hypothesize that if statistical differences were to be found, it means that the analyzed trait was related to the tested sample type. A confidence interval of 95% was used where  $p$  values lower than 0.05 were considered statistically significant. The statistical analyses were performed in IBM SPSS statistics software version 22.

## Results

### Fracture characterization

All types of fractures could be found in the different samples (spiral and epiphyseal fractures excluded due to the experimental set-up). However, fresh experimental fractures are mostly represented by oblique, butterfly, and transverse fractures, while dry bones generally present transverse fractures. Moreover, the percentage of comminuted fractures is much higher in autopsy samples than in the experimental samples. Spiral and epiphyseal fractures are not accounted for during the experimental analysis, even though they are commonly

present in autopsy samples. Both types of fractures need different fracture mechanisms, and all experimental bones were impacted on the anterior side in the middle of the bone causing a three-point bending (Fig. 2).

### Trait characterization

Table 2 gives a brief summary of all the traits and their description previously defined by Scheirs et al. [7].

Figure 3 shows the distribution of the traits in autopsy and fresh experimental samples, distinguishing experiments with and without axial compression. None of the traits were found in dry bone samples; therefore, they will not be included in the following graphs. Bone scales were not found in any of the experimental groups, and crushed margins were scarce in the experimental groups; only two samples where compression was applied presented this trait. The presence of bone scales and crushed margins in autopsy samples is scarce; together, they are one of the most uncommon traits. Wave lines and layered breakage show similar percentages in all groups, the latter being the most common trait in fresh bones. Plastic deformation could be reproduced during the experiments; however, it was clearly more present in those where no axial compression is applied. In contrast, flakes are less frequently present in experimental samples without compression but are equally encountered in autopsy samples and experimental samples with compression.

To establish any significant differences in the occurrence of traits between the autopsy samples and experimental groups, a logistic regression was performed. If significant differences are found, it means that the occurring trait is associated with that specific sample type. No significant differences are found in the occurrence of any trait between autopsy and experimental samples when compression is applied. Only the occurrence of flakes and plastic deformation exhibits significant differences between autopsy and experimental samples without

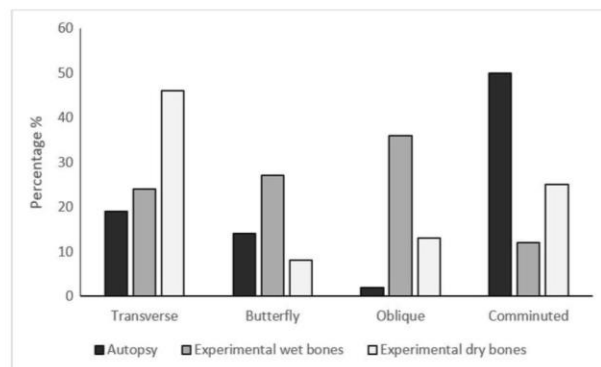
compression ( $p=0.007$  for plastic deformation and  $p=0.035$  for flakes), and between both experimental settings ( $p=0.01$  for plastic deformation and  $p=0.03$  for flakes). Plastic deformation is more common when no compression is applied, whereas flakes are more common when compression is applied.

In dry bones, fracture edges are more jagged and none of the distinct perimortem traits could be experimentally reproduced. Nevertheless, in two specimens, there was something present that may resemble layered breakage, but it should be noted that this observation differed from layered breakage in fresh samples. While the layers in fresh bones tend to be parallel to each other and occupy the entire width of the bone shaft, in dry bone, the layers are not parallel but in different directions to each other; they are very shallow and have a rough surface. Furthermore, close analysis of layered breakage leads us to observe that this trait appeared deeper in the cortical bone in autopsy samples than in the experimental fresh samples (Fig. 4).

### Discussion

By means of an improved experimental setting, the main goal of this research was to gain further knowledge about the fracture mechanism and the conditions for the appearance of the previously reported perimortem traits [7]. In literature, there are already many characteristics known to make a distinction between perimortem and postmortem fractures such as color, and smooth and rough edges [4, 14, 15]. Nevertheless, this distinction is still a challenge in forensic cases, especially when the bones are still wet. In a previous preliminary study, we hypothesized that some of the perimortem traits may be specifically related to *intra vitam* conditions [7]. This would help forensic anthropologists to shorten the time gap of perimortem trauma. In the current study, we are looking for evidence that allows us to suggest that the presence of

**Fig. 2** Overview of the type of fractures in percentages among the experimental and autopsy samples of adults aged 55 years or older





**Table 2** Descriptive table of all the traits

Trait	Description
Layered breakage	A cortical layer pattern in the compression side of the diaphysis of long bones
Wave lines	A smooth undulation, resembling a “wave,” on the smooth long edges of fractures
Bone scales	A form of plastic deformation resembling “fish scales” that occur close to the margin of the fracture at the compression side
Flake (defect)	A flake is a superficial loss of a thin piece of bone. The imprint that is left is defined as a flake defect
Crushed margins	Crushed margins are similar to bone scales but occur directly on the margin of the fracture on the compression side
Plastic deformation	A permanent deformation of the bone making it impossible to return to its original shape

musculoskeletal activity, and surrounding soft tissue, during fracturing plays a role in the occurrence of these traits. To our knowledge, there are no studies so far that shorten the perimortem interval by defining distinct *intra vitam* traits using a human model, which makes this research original.

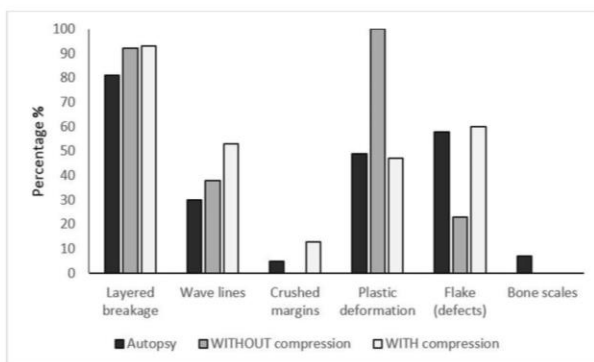
The experimental samples—both fresh and dry—showed transverse, oblique, butterfly, and comminuted fractures. Nonetheless, dry bones mainly represent transverse fractures, autopsy samples are mainly represented by comminuted fractures, and fresh experimental fractures mainly exhibit oblique, butterfly, and transverse fractures. Cohen et al. [16] performed a study on pig bones with a similar experimental setting. However, they could not reproduce butterfly fractures, only oblique fractures, and comminuted fractures [16]. This discrepancy between results clearly supports the importance of the human model to obtain truthful results, which resemble autopsy samples. Our results suggest that the type of fracture does not give any information about whether the fracture has occurred peri- or postmortem since all types of fractures can be found in each experimental group.

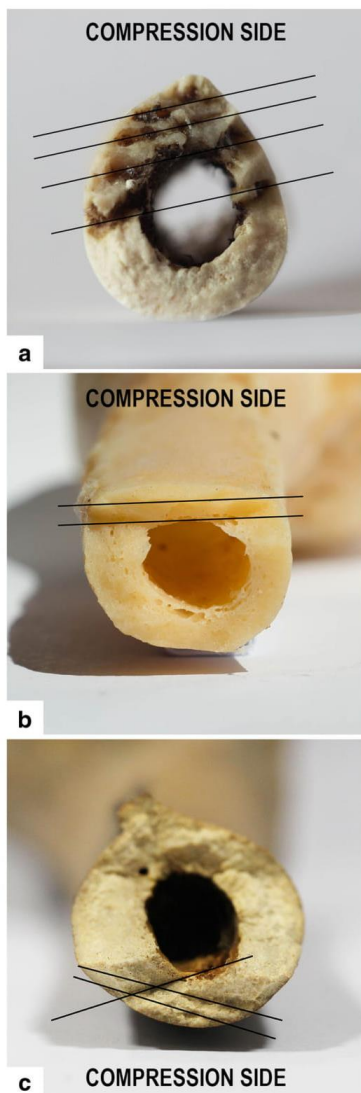
The appearance of the fractures broken by fast impact is in clear contrast with the appearance observed by Scheirs et al. [7] when applying a slow loading to long bone samples. The fractures impacted at high speed are larger and more often

comminuted than those fragmented by a slow loading [7]. The size of the fracture itself seems to be related to the speed of impact; the higher the impact, the larger the fracture. It is already known that high-energy absorption leads to more fractures, therefore a higher chance to obtain a comminuted fracture [4]. In reality, the impact could be even stronger, and therefore, there will likely be more than one point of impact that will act on the bone. That is why autopsy samples still show more comminuted fractures than the experimental ones. Nevertheless, the impact pattern is more comparable to the fractures obtained from autopsies, indicating that the new experimental setting is an improvement to the one in the previous preliminary study. Cohen et al. [16, 17] related comminuted fractures to axial loading and soft tissue in their animal model. Previously, Scheirs et al. [7] already suggested that muscular compression and the presence of soft tissue might influence the manner of fracturing and the presence of traits. The results in this study on the human model suggest that there is a relation between the traits and the *intra vitam*/perimortem pattern involving axial compression and possibly soft tissue [16, 17].

The distribution of the traits in both experimental and autopsy samples is similar to the one obtained in the previous preliminary study [7]. The results confirm that layered

**Fig. 3** Distribution of the traits in percentages over the different experimental groups of wet bones and autopsy samples





**Fig. 4** Differences in layered breakage between a perimortem autopsy (a), experimental fresh (b), and experimental dry (c) sample. The black lines show the layers in the cortical

breakage is the most common perimortem trait, being present in almost all autopsy and fresh experimental samples. The addition of compression did not change the results significantly on this trait, which indicates that axial compression has no

influence on the formation of layered breakage. Although it is not a specific trait for *intra vitam* circumstances, it is a clear perimortem trait. Furthermore, our results allow us to enhance the definition of layered breakage as a perimortem trait. A comparable form of this trait was found in two dry bone fractures; however, the appearance is different. Whereas they are parallel in fresh fractures, they are not in dry fractures as they appear in several directions in one single fracture (Fig. 4). This distinction is important when evaluating the characteristics of bone fractures.

Close analysis of layered breakage leads us to observe that this trait appeared deeper in the cortical bone in autopsy samples than in the experimental fresh samples (Fig. 4). This might suggest that it needs specific *intra vitam* conditions to occur, but during our experiments, it was not possible to reproduce them exactly like autopsy samples. Clearly, this area deserves further research. We hypothesize, however, that it may be related to the age of the available experimental samples. A limitation in this study is the age distribution of the experimental samples. It would be beneficial to also include young people within the study; however, undertaking research with human bone samples presents limitations regarding the number of donors who make their remains available to science, and the subsequent restrictions this places on the breadth of the study. It is well-known that with increasing age and other age-related factors, the cortical layer of the bone becomes thinner [18, 19]. Therefore, young bones have a thicker cortical layer and could therefore form several deep layers when the bone fractures. Since only mature and older bones were used in the experiments, it may have affected the appearance of the layered breakage, as there is no space available to form layers. Layered breakage in the autopsy samples from individuals of 55 years old and more has a similar appearance as the layered breakage from the experimental samples, supporting our theory.

Wave lines are present in experimental fresh bone, but not in dry bone samples. The edges of fractured dry bones are too jagged to show any wave lines. Wave lines can therefore be specified as a perimortem trait. In fresh bone samples, no differences are found between fractures with or without axial loading concerning wave lines. In accordance with our assumption, this indicates that wave lines are a perimortem trait, but not necessarily an *intra vitam* trait as suggested by Scheirs et al. [7].

Furthermore, based on our increased sample size, this study confirms that crushed margins are very rare in autopsy samples. Scheirs et al. [7] suggested that this trait needs a very specific mechanism, probably related with muscular action causing a collision of the fracture edges during fracturing [7]. Sequential to this hypothesis, an attempt was made to reproduce them by adding springs to the experimental setup, so a recoil can take place when the fracture initiates. The results, however, are positive in only two experimental

samples where compression was applied. Clearly, this result is insufficient to test our hypothesis. Furthermore, they are not observed in samples fractured without compression, and differences between both experimental groups are not statistically significant. The mechanism behind the trait suggests a complex fracture process, not only compression, but additionally, the presence of surrounding soft tissue, impact mechanics, bone, and/or individual variables as age may have an influence on their formation. Nevertheless, crushed margins deserve further research to conclude that they could be used as an indicator of trauma under *intra vitam* conditions.

Bone scales did not appear in any of the experimental samples. In autopsy samples, bone scales do not occur often either. Nevertheless, Scheirs et al. [7] found 29% of bone scales in experimental samples applying slow loading and axial compression to break the bone [7]. Differences in impact speed may suggest that these circumstances play an important role in the development of bone scales. It would also offer an explanation as to why this trait does not appear more often in autopsy samples, since bones are usually broken on fast impact.

Flakes, on the other hand, are a very common trait in both autopsy samples and in fresh experimental samples with compression. Flakes are less common in fractures fragmented without compression and are not present in dry bones. Based on statistical analysis, significant differences between the experimental groups are observed. The results clearly show that axial compression is a key factor in flake development. Absence of flakes in dry bone fractures and a similarity in the incidence between autopsy and compression group supports the hypothesis that flakes may be related to musculoskeletal activity and the presence of surrounding tissue. This hypothesis has already been suggested by Scheirs et al. [7] with a smaller sample size [7]. Future research will potentially clear this criterion. The interpretation of blunt force trauma will be more accurate if flakes can be considered as *intra vitam* markers since they are abundantly present in fractures.

The final trait, plastic deformation, appeared in all fresh bone samples; therefore, it is a clear perimortem trait. It is significantly more common in the experimental group without axial compression compared to the other groups, and there are no significant differences between autopsy and compression samples. When fresh living bone is subjected to blunt trauma, it will undergo two mechanisms before a fracture occurs. During the first phase, the bone will undergo elastic deformation. This means that the bone can return to its original dimensions. In the second phase, it will endure plastic deformation, which will deform the bone permanently. The plastic phase will continue until breaking occurs [4, 15]. We hypothesize that axial compression and surrounding soft tissue limit the bending freedom and therefore cause less plastic deformation. During the experiments without compression, there is no compressive force that keeps the bone immobile, causing more bending and subsequently, more plastic deformation.

## Conclusions

With an improved experimental approach, we were able to reproduce fracture patterns in human bone samples similar to those found in traumatic cases by adding axial compression and increasing the impact energy. This way, it is possible to investigate specific patterns that may be helpful in forensic examinations.

This study confirms the six distinct characteristics that can be valuable to utilize in the distinction between peri- and postmortem fractures: layered breakage, bone scales, crushed margins, flakes with flake defect, wave lines, and plastic deformation. The results show that all traits are perimortem traits.

Nevertheless, we see a tendency that the combination of traits—instead of the presence of each trait individually—may make it possible to distinguish *intra vitam* from perimortem fractures.

In the future, we would therefore like to obtain a probabilistic model to assess a pattern that defines a combination of the traits. Increasing the sample size will help to create such a model.

Based on our results in the current and previous study, we highly recommend research with true human bones and avoid research on animal bones. We observe and read many differences in other literature regardless of both being the bone. More research could be done to understand crushed margins and plastic deformation. Nevertheless, the current conclusions may already be helpful to professionals in forensic cases, as the traits may shorten the perimortem time gap resulting in a change of the paradigm to understand the chronological events of a fracture.

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## Compliance with ethical standards

This study was approved by the Ethics Commission of Human and Animal Experimental Work (CEEAH) of the UAB, in compliance with the ethical regulations.

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#### **4.5. Rasgos perimortales de las fracturas de huesos largos (2018)**

Revista Internacional de Antropología y Odontología Forense in 2018

Addressed research questions:

What are the macroscopic characteristics that define a distinct perimortem pattern in wet long bone fractures, caused by blunt force trauma?



## Rasgos perimortales de las fracturas de huesos largos. LONG BONE FRACTURES PERIMORTEM TRAITS.

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**RESUMEN:** Establecer la cronología de una fractura ósea sigue siendo uno de los principales retos a los que se enfrenta el Antropólogo Forense. El objetivo de este trabajo es presentar un patrón morfológico que mejore y facilite el diagnóstico diferencial entre fracturas peri- y post-mortales. Presentamos iconográficamente 5 rasgos relacionados con el periodo perimortem: fractura laminar, márgenes ondulados, superficie escamosa, escamas y defectos en escama y márgenes conminutos.

**PALABRAS CLAVE:** Antropología forense, trauma óseo, data de lesiones, traumatismo perimortem.

**ABSTRACT:** Determining the time of injury is still a challenging task in Forensic Anthropology. The aim of this study is to report a morphological pattern that improves the distinction between peri- and post-mortem fractures. 5 peri-mortem traits are iconographically presented: layered breakage, wave lines, bone scales, flakes with matching flake defect and crushed margins.

**KEY WORDS:** Forensic anthropology. Bone trauma. Time of injury. Perimortem trauma.

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Establecer la cronología de una fractura ósea sigue siendo uno de los principales retos a los que se enfrenta el Antropólogo forense. A partir de un modelo experimental y comparando con ejemplares indubitados de autopsias forenses, hemos encontrado un patrón característico de rasgos macroscópicos asociados a las fracturas *perimortales* de huesos largos (Scheirs et al., 2016). La presencia de uno o más de estos rasgos permite situar la fractura en este periodo cronológico, facilitando el diagnóstico diferencial con fracturas *postmortales*.

En este trabajo presentamos ejemplos fotográficos de cada una de las 5 características morfológicas que pueden encontrarse en fracturas diafisarias. Este patrón está relacionado con las características biomecánicas del

traumatismo, por tanto nos permite reconstruir la dinámica de la lesión (Scheirs et al., 2016).

### 1. FRACTURA LAMINAR.

Rasgo que está presente en la práctica totalidad de fracturas *perimortales*. Muy característico de las fracturas de diáfisis, por el contrario, no se observa en fracturas epifisarias ni espiroideas, donde el grosor cortical es menor al necesario para su aparición. Se observa en el margen que ha experimentado la compresión, en el espesor de la cortical y se distingue por presentar un aspecto laminar, longitudinal, paralelo, con láminas de grosor variable.



Fractura laminar.

## 2. MÁRGENES ONDULADOS.

Rasgo que suele ir asociado a fragmentos o trazos de fractura grandes. Se distingue por márgenes suavemente ondulados,

simulando crestas de olas. Suele aparecer cuando los trazos de fractura son relativamente largos, como en el caso de fracturas conminutas o en mariposa.



Márgenes ondulados.

### 3. SUPERFICIE ESCAMOSA.

Rasgo poco común que suele aparecer asociado a la deformación plástica del lado que ha sufrido compresión. La

cortical aparenta el aspecto escamoso de un pescado, con finas láminas sobrepuestas en la superficie cortical. En ocasiones es útil su distinción mediante lente de aumento o luz rasante.



Superficie escamosa.

#### 4. ESCAMAS Y DEFECTOS EN ESCAMA.

Rasgos relativamente frecuentes. El defecto en escama es el resultado de una pérdida superficial del hueso, situada a

menudo justo en el margen de la fractura. Es útil unir el foco de fractura para poder observar la presencia de estos pequeños defectos que, a modo de finas láminas, se han desprendido de la superficie cortical.



Escamas y defectos.

#### 5. MÁRGENES CONMINUTOS.

Rasgo poco frecuente. Fragmentación del margen de compresión de la fractura, en ocasiones visible mediante el uso de lente de aumento. Aparecen como pequeños fragmentos astillados y adheridos al margen. Es una

característica muy vulnerable a la limpieza del hueso, en especial cuando el margen se somete a una abrasión mecánica, con el riesgo de perder los pequeños fragmentos que permanecen unidos por finas láminas, a modo de bisagra.



Márgenes conminutos.

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 Galtés I, Scheirs S; Ortega M, Rodríguez-Baeza A, Malgosa A

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#### **4.6. The eagle's eye: can perimortem traits be visualised with a CT scan?**

Submitted to Forensic Science, Medicine and Pathology in September 2019  
by

Sarah Scheirs, Mònica Cos, Hannah McGlynn, Assumpció Malgosa, Ignasi Galtés

Addressed research questions:

Can perimortem traits be visualised with different imaging techniques?

## **Abstract**

Perimortem fracture patterns in long bones, defined in previous publications, include layered breakage, bone scales, crushed margins, flakes with flake defect, wave lines, and plastic deformation. The traits help professionals during trauma analysis to differentiate peri- from post-mortem fractures. This study will therefore investigate whether these traits can be recorded with Computed Tomography as CT scans are becoming more popular in forensic science. The individual CT slices were investigated, and a 3D model was reconstructed for each bone sample. Tension lines and severe plastic deformation were visible on the individual slices and the 3D models. Additionally, layered breakage and flake defects were also clearly distinguishable on the 3D reconstructions. Based on the results, CT imaging may be a useful and fast tool to document, visualise, and analyse findings of blunt force trauma.

## **Key words**

Autopsy, computed tomography, post-mortem examination, CT scan, 3D bone reconstruction, perimortem traits

## **Introduction**

During trauma analysis, one of the most important tasks of the forensic anthropologist is to determine the time frame in which skeletal trauma was inflicted [1]. In particular the distinction between peri- and post-mortem trauma is still a challenge since most characteristics described in literature are related to post-mortem trauma features [2,3].

Nevertheless, previously Scheirs et al. described a new blunt force trauma bone pattern, specific for the perimortem period [4,5]. These new traits (Table 1) may help professionals in the field to distinguish perimortem trauma from post-mortem trauma.



Table 1 Descriptive table of the perimortem traits.

Trait	Description
<b>Layered breakage</b>	A layer pattern in the cortical, located on the compression side of the diaphysis of long bones.
<b>Bone scales</b>	A form of plastic deformation resembling 'fish scales' that occur close to the margin of the fracture on the compression side.
<b>Crushed margins</b>	A form of plastic deformation similar to bone scales but occur directly on the margin of the fracture on the compression side.
<b>Flake (defect)</b>	A superficial loss of a thin piece of bone. The imprint that is left, is defined as a flake defect.
<b>Wave lines</b>	A smooth undulation, resembling a 'wave', on the smooth long edges of fractures.
<b>Plastic deformation</b>	A permanent deformation of the bone making it impossible to return to its original shape.

In the current work, we were intrigued whether these traits can be recorded by Computed Tomography (CT). CT scans are becoming more popular in forensic science as a non-invasive medical imaging technique, and have become a frequent addition to medicolegal autopsies [6–8]. This could make it a useful tool in forensic, and even archaeological cases on time determinations of blunt force trauma. A CT scan could especially be useful in cases with only limited access to the bone material.

With this study we would like to show how certain traits can be visualised with CT imaging and its application in forensic investigations.

## Materials and methods

### *Samples*

A total of 15 human fractured long bone fragments with known perimortem traits from 10 forensic cases were included in this study (8 males, 2 females; with age range of 16 to 85 yrs, and mean age of 40 years). The bones were retrieved from autopsies at the Institute of Legal Medicine and Forensic Science of Catalonia for further medicolegal analysis after traffic accidents and falls.

After retrieval from the autopsies, the bones were carefully cleaned by macerating them in a hot water and detergent solution (1 cup of detergent for 5 L of water) for 3–5 hours – depending on the amount of soft tissue that had to be removed – at a temperature of  $\pm 90$  °C. The remaining tissue was removed with surgical tools and the bones were left to dry.



**CT scan**

All bone samples were scanned with the Aquilion™ PRIME from Toshiba Medical Systems, allowing a multislice computed tomography (MSCT) to be taken, generating up to 160 slices per rotation. A total of 2000 slices were obtained after the scan. Since we were interested in the delicate details of the fractures, the scan was taken with a slice thickness of 0.50 mm in the axial plane and 0.43 mm in the sagittal and coronal plane, with 120 kVp and 300 mAs as basic parameters. The acquired images were obtained in a Digital Imaging and Communications in Medicine or DICOM format. The individual slices were examined using Starviewer Medical Imaging Software, and Philips IntelliSpace Portal was used to reconstruct a 3D model of the bone samples.

**Results**

On the individual slices, tension lines were clearly visible and could be easily tracked in the coronal, axial and sagittal view (Fig. 1). Plastic deformation could be visualised in the case of severe deformations and was best observed in the axial plane (Fig. 2). Other traits could not be visualised in the individual slices.

On the reconstructed 3D models, layered breakage could be observed in all samples that featured this trait (Fig. 3, a-f). The 3D model shows very clearly the layered pattern, opposite of the smooth tension side.

Also flake defects were clearly detected on the fracture margins of all the samples presenting this trait (Fig. 4, a-f). Apart from these common characteristics; plastic deformation (Fig. 4f, 3d), tension lines (Fig. 5a), and wave lines (Fig. 5b,c) were also observed on the reconstructed models. Bone scales and crushed margins (except for the one in Fig. 3d) couldn't be observed in any of the reconstructed models.

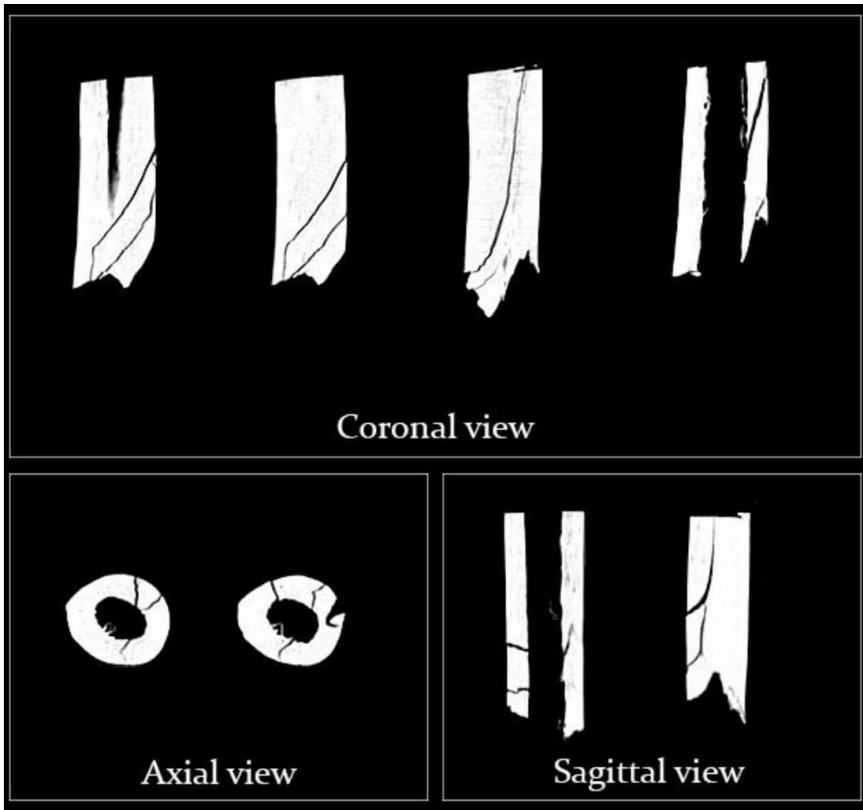


Fig. 1 Individual slices from a bone sample with tension lines in the three viewing planes.

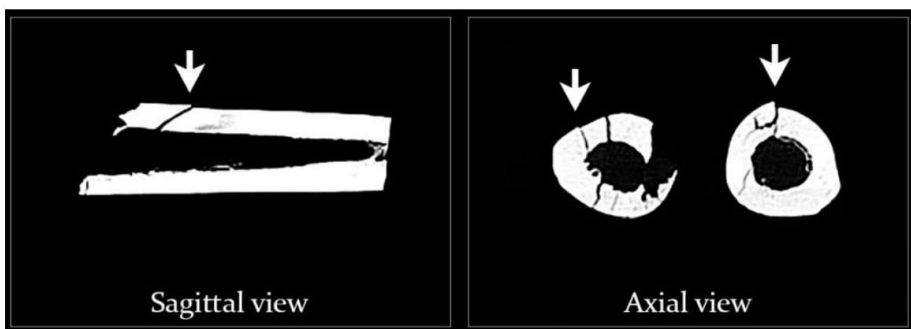


Fig. 2 Individual slices of the sagittal and axial view of a bone sample with severe plastic deformation.

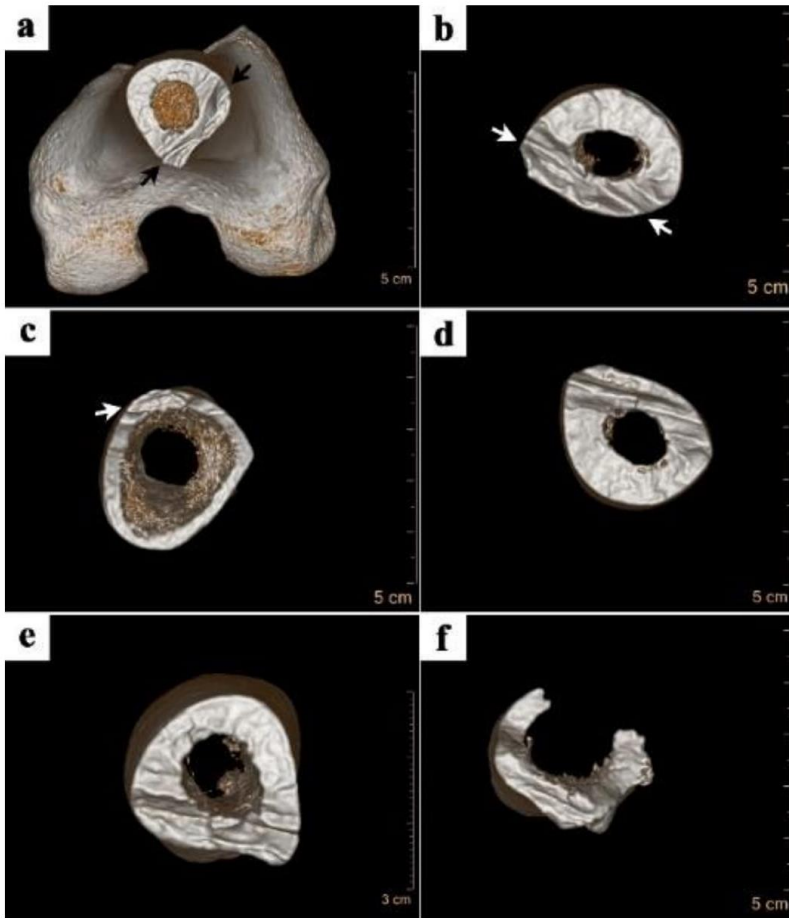


Fig. 3 Layered breakage as seen in the 3D reconstructed CT scan.

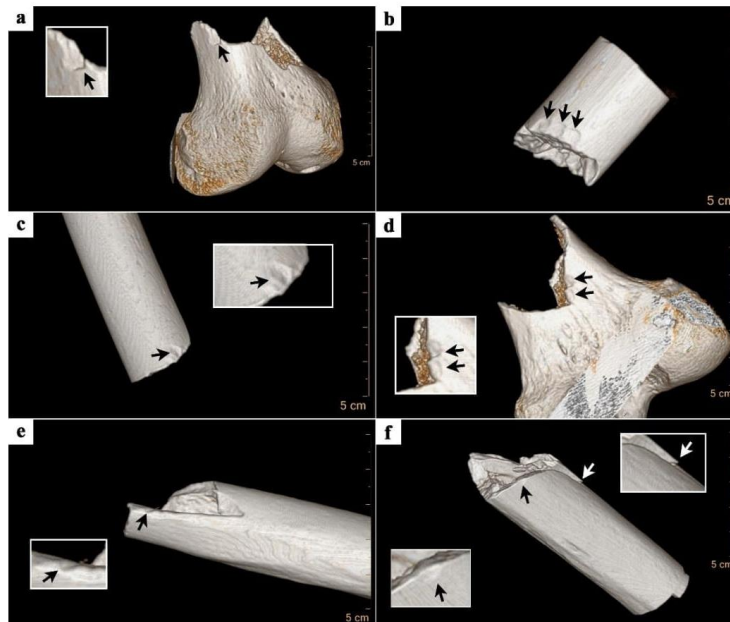


Fig. 4 Flake defects on the margin of different bone samples and plastic deformation (f; white arrow)

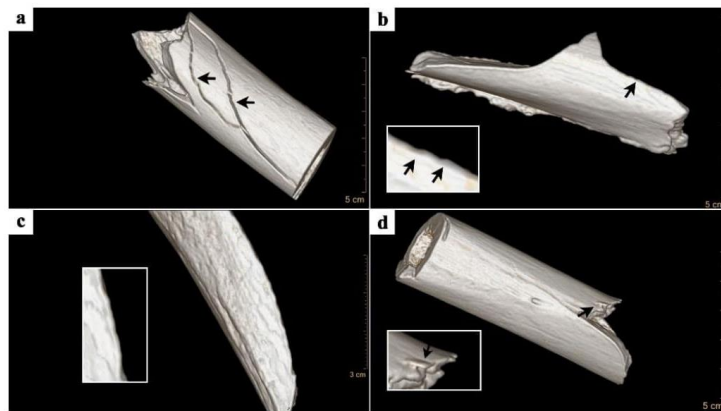


Fig. 5 (a) Tension lines, (b,c) wave lines on the margin, (d) crushed margin/plastic deformation

## Discussion

CT scans are commonly used to document and visualise traumatic findings in forensic settings in a non-invasive way [7,9]. We believe this technique may also be used to gain more information about fracture timing in samples that are difficult to access. For this purpose, we were especially interested in the presence of the previously defined perimortem traits in long bones: layered breakage, wave lines, flake defects, plastic deformation, bones scales, and crushed margins[4,5].

Layered breakage is one of the most common traits found in long bone fractures and as with the naked eye, layered breakage can easily be visualised using a CT scan. It is a specific trait of the compression side with its characteristically parallel layers, very different from the opposite, smooth tension side. On the 3D reconstruction, layered breakage is even clearer than examination with the naked eye since there is no influence of blood or dirt residue. Furthermore, layered breakage is less prominently present in bones with a thin cortical layer (Fig. 3c).

The next most common trait are flakes with their flake defect. Flakes are difficult to retrieve since these superficial bone losses get easily lost in the surrounding tissue. They do however leave an imprint, defined as a flake defect (Fig. 4). They usually occur close to the margin of the fracture [4,5]. In the CT scan they can be visualised as irregularities in the bone surface that catch the eye by the shadows they create. In Fig. 4f, a small flake defect (black arrow) and plastic deformation (white arrow) can be observed.

Albeit not as clear as the other traits, wave lines could also be visualised on the long margins of the fractures (Fig. 5b,c), although the resolution is quite low for this small and detailed trait.

While a crushed margin is an uncommon trait, we could observe it in one explored bone sample (Fig. 5d), in other examined bone samples it was not visible. It is a very sensitive trait and can be easily destroyed if handling the sample without care. It is therefore possible that when the scan can be performed *in situ* – without manipulating the bones itself – the trait will be clearer to observe.

Apart from the abovementioned perimortem traits we also observed tension lines very clearly. They can be observed using a 3D reconstruction (Fig. 5a) and they could be observed in the 2D scan slices, making it possible to follow the path of the lines (Fig. 1).

We are aware of the limitations presented in this report since the CT scan could not be performed *in situ* due to infrastructural limitations. Still, based on the results we may conclude that CT imaging may be a useful and fast tool to document, visualise, and analyse findings of blunt force trauma. It can be a great addition to conventional autopsies but should not be considered as a replacement for them.

It can be a useful non-invasive technique when bones are difficult to access or when there are cultural restrictions. Even though not all traits can be visualised on the CT scan; layered breakage, flake defects, and plastic deformation can themselves aid in perimortem bone trauma analysis.

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#### **4.7. Differentiation between perimortem trauma and heat-induced damage: the use of perimortem traits on burnt long bones (2019)**

Forensic Science, Medicine and Pathology in 2019 (IF (2018) 1.815, Q2 in Legal Medicine)

Addressed research questions:

What are the macroscopic characteristics that define a distinct perimortem pattern in wet long bone fractures, caused by blunt force trauma?

Can certain characteristics be related to certain trauma mechanisms and therefore deduce the biomechanical circumstances?

Can the anthropological paradigm be changed by the presence of *intra vitam* traits?

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## CASE REPORT



## Differentiation between perimortem trauma and heat-induced damage: the use of perimortem traits on burnt long bones

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### Abstract

Burnt human remains present a difficult interpretative dilemma to forensic pathologists and anthropologists. Distinguishing postmortem damage in long bones as a result of fire damage from perimortem fractures is an important challenge in trauma analysis. During our case investigation of a burnt body from a fiery car crash, distinct perimortem traits on long bone fractures were still distinguishable along the charred areas. Next to timing of fractures and shortening the perimortem time gap suggesting that the fractures occurred when soft tissue was still present, the traits make it possible to distinguish blunt trauma caused by the accident from heat-induced bone damage. Applying this specific perimortem pattern could be an additional macroscopic tool to interpret blunt force trauma more accurately in the analysis of burnt remains.

**Keywords** Forensic anthropology · Burnt bones · Bone fractures · Biomechanics · Perimortem

### Introduction

Burnt human remains present a difficult interpretative challenge to forensic pathologists and anthropologists. Color change, shrinkage, and fragmentation, in addition to fracturing and warping are often confounding components of thermally altered remains. Thermally-induced damage causes numerous artefacts that complicate not only the identification of the victim, but also trauma interpretation and reconstruction of the circumstances at time of death [1].

Depending on the temperature and duration of exposure to the fire, bone tissue is affected as a consequence of the destruction of organic and inorganic components of the bone. Known heat-induced alterations on bone tissue include cracking, splitting, and shrinking [2]. Nevertheless, distinguishing

post-mortem damage as a result of fire damage on long bones from perimortem fractures, is one of the main and most important challenges in trauma analysis [3–6].

Recently, Scheirs et al. [7] reported five macroscopic distinct fracture traits that are good estimators of perimortem long bone trauma: layered breakage, bone scales, crushed margins, wave lines and flakes with matching flake defect. During our case investigation of a burnt body from a fiery car crash, the finding of some of these traits on the fractured long bones allowed us to identify perimortem trauma from the accident and differentiate them from heat-induced damage. This work presents the results of this case investigation with the aim to demonstrate the applicability of the perimortem long bone fracture traits on the analysis of burnt remains. Since the traits are specific for long bones, the revision of this case will focus on the burnt fractured long bones of the lower extremities and the perimortem traits observed along the damaged parts.

### Case report

After a fiery car crash a carbonized 45 year old victim arrived at the pathology service for analysis of the cause and circumstances of death.

Examination of the body was carried out by a pathologist and revealed the characteristic pugilistic posture, with adduction of the shoulders and flexion of the elbows and wrists. Bone damage was limited to the lower extremities which were

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severely affected by the fire. Both sides exhibited several fractures with thermal destruction, skeletal loss, and amputation of the feet at the level of the ankles. The long bones revealed a black charred aspect with areas of grey calcined bone, mainly at the distal parts. Given that the decedent had been involved in a high-speed crash, blunt force trauma to the victim was expected, however thermal bone damage caused by the fire masked the injuries. This overlap raises the question of whether the fractures were caused by the vehicle impact or were subsequently caused by fire due to the burning of the vehicle? A forensic anthropologist was added to the investigation team to carefully examine the fractures and to help understand the sequence of events.

### Fracture analysis

Based on previous research by Scheirs et al. [7] on perimortem traits in long bones, only the lower limb fractures in the current case were investigated to check whether traits observed in previous studies could be observed on the fractures in this case. In order to investigate the fractures, the bones were removed from the body and cleaned using a soft macerating method with hot water ( $\pm 90$  °C) and commercial detergent for 2–5 h, depending on the amount of flesh that had to be removed.

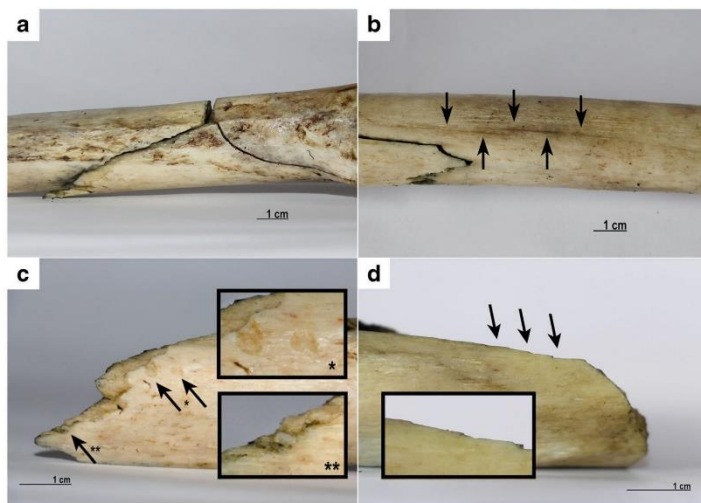
After cleaning and drying, the long bone fractures were macroscopically investigated by the naked eye. The left femur showed a butterfly fracture at the third upper part of the diaphysis (Fig. 1a), exhibiting the breakaway fragment at the internal side of the bone. Longitudinal fractures were present

along the shaft at the anterolateral side (Fig. 1b). Close analysis of the fracture margins revealed numerous flake defects at both compression and tension sides and crushed margins at the compression side (Fig. 1c). Additionally, layered breakage could be observed, and wave lines were visible on the longitudinal margins of the fracture (Fig. 1d).

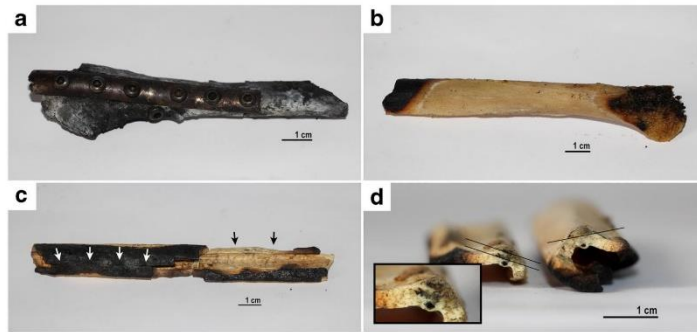
The right fibula had an osteosynthesis plate at the distal end (Fig. 2a). In this area, bone showed a black charred aspect with a transversal fracture and partial bone loss but no perimortem traits were visible. The superior epiphysis was partially burnt, with a more charred and eroded area at the proximal end (Fig. 2b). The diaphysis was also partially charred, and showed a comminuted fracture, with several transversal fractures, regular longitudinal fracture lines (Fig. 2c) and shorter irregular cracks limited to the more charred areas. Additionally, layered breakage with its typical parallel layers was observed in the margins of the comminuted fracture (Fig. 2d).

The right tibia was homogeneously charred. The diaphysis presented comminuted fractures with loss of several fragments (Fig. 3a, b). Longitudinal straight fractures crossing burnt and unburnt areas were observed (Fig. 3c). The proximal and distal end exhibited bone erosion, with cortical loss and large areas of exposed cancellous bone. Analysis of the comminuted fracture margins revealed layered breakage, wave lines in the larger exposed pieces of the fractures (Fig. 3d) and numerous flake defects on the margins in burnt areas (Fig. 3e). The distal end was gray calcined with irregular longitudinal cracks that could overall be observed in the burnt surfaces (Fig. 3f).

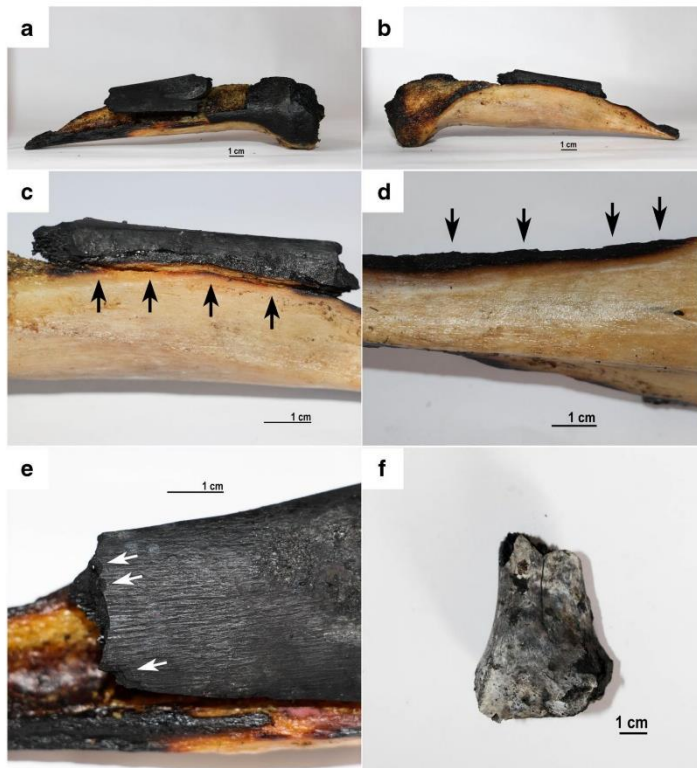
**Fig. 1** Butterfly fracture at the femoral diaphysis (a), longitudinal fractures along the shaft (b), flake defects (\*) and crushed margin (\*\*\*) on the margin (c), wave lines on the longitudinal margin (d). (See Scheirs, et al. (2017, 2019) for a detailed description of the traits [7, 11])



**Fig. 2** Osteosynthesis plate at the distal part of the fibula (a), charred superior epiphysis (b), transversal and longitudinal smooth fractures in the diaphysis (c), layered breakage in the fracture margin (d). (See Scheirs, et al. (2017, 2019) for a detailed description of the traits [7, 11])



**Fig. 3** Medial and lateral view of the right tibia (a, b), longitudinal straight fracture lines (c), wave lines on long margins (d), flake defects in burnt area (e), calcined distal part (f). (See Scheirs, et al. (2017, 2019) [7, 11]) for a detailed description of the traits)



## Discussion

Understanding the biomechanics behind bone fractures is the best interpretative tool to distinguish perimortem trauma from post-mortem fractures as a result of fire. Performing this differential diagnosis is one of the main challenges for the forensic anthropologist during the analysis of burnt remains.

Thermal bone damage is highly variable but predictable across the body because the body's position at the time of death and the shrinking of muscle fibers caused by heating. These actions will determine the areas which will be exposed to fire. According to Shipman et al. [8], ultrastructural changes associated with the dehydration process of the bone tissue exposed to fire are responsible for the bone fractures. They suggest that the fluid content of the bone prior to burning is the major determinant of the heat-induced fracture pattern. Classically, this post-mortem heat-induced bone damage has been characterized by the presence of patina fractures or fine cracks, typically on the surface of flat bones; longitudinal fractures along the long axis of long bones; curvilinear fractures on long bone shafts; transverse fractures perpendicular to the longitudinal axis of a long bone; and delamination or peeling fractures typically of the epiphyseal regions [2]. Furthermore, Symes et al. [1] described the presence of step fractures, often extended from the margin of the longitudinal fracture transversely across the bone shaft, and burn line fractures, following the burn borderline and separating burnt and unburnt bone.

Moreover, perimortem trauma is trauma that was inflicted when the bone still has its water and organic components causing it to behave as fresh bone. When fresh bone is subjected to blunt trauma, it will undergo two mechanisms before a fracture occurs. In the first phase, the bone will undergo elastic deformation. This means that the bone can return to its original dimensions. In the second phase, it will endure plastic deformation, which will deform the bone permanently. The plastic phase will continue until fracturing occurs [9, 10]. Taking this into account, a perimortem (fresh) fracture will have a specific macroscopic pattern, different from post-mortem damage.

Scheirs et al. [7] defined five unreported distinct macroscopic traits of perimortem fractures in long bones: layered breakage, as a very characteristic horizontal cortical layer pattern in the compression side of the diaphysis of long bones; wave lines as smooth undulations that have a gentle slope and a rapid drop, resembling a 'wave' on the smooth long edges of fractures; bone scales as a form of plastic deformation resembling 'fish scales' that occur close to the margin of the fracture at the compression side; crushed margins which are defined as small fractured pieces of bone still attached to the cortical surface at the margin of the compression side of a fracture; and finally, flakes which are superficial losses of thin pieces of cortical bone. The latter has been recently related with the presence of surrounding flesh in fresh fractures [11]. Despite the fact that these characteristics constitute a specific

perimortem pattern, not all characteristics must be involved in all perimortem lesions. Evaluating these morphological features in a fracture will allow us not only to time the injury more accurately but hypothesize if the fracture occurred at the moment of death, when soft tissue and muscles are involved [11].

In the current case, a combination of perimortem blunt trauma and thermal damage is to be expected on the victim because of the car accident and post-crash combustion. The post-mortem thermal damage predominately affected the distal ends of the lower limbs, mainly on the right side, where the proximal epiphysis of the tibia and fibula were also affected. In general, damage followed the characteristically burnt bone fracture biomechanics, defined by Symes et al. [1], including charring and erosion of the cortical surface with some transverse fractures, but mainly longitudinal irregular cracks limited to the burnt surfaces with no presence of perimortem traits. This damage was considered to be related to the conflagration of the car after the accident. On the other hand, perimortem fractures were located at the diaphysis of the left femur, right tibia and fibula. Except for a butterfly fracture on the femur, the other fractures were comminuted. All of them present perimortem traits on the margins, with the presence of layered breakage, crushed margins, wave lines, and numerous flake defects. This damage was considered to be related to the trauma circumstances due to the car accident. No additional damage was considered related to the recovery of the remains.

The present case shows that the recently reported perimortem traits on long bone fractures are still distinguishable in charred bones. In addition to the timing of fractures, the shortening of the perimortem time gap suggests that the fractures occurred when soft tissue was still present. These results might be of interest for other anthropologists who are faced with the challenge of distinguishing blunt trauma from thermally induced bone damage. This new perimortem long bone pattern could be an additional macroscopic tool to interpret blunt force trauma more accurately in the analysis of burnt remains.

## Key points

- 1 This study was carried out to investigate whether specific perimortem traits caused by blunt force trauma could be distinguished from postmortem fire damage on long bones.
- 2 Close analysis of the long bones revealed longitudinal fractures, flake defects, layered breakage, and wave lines, thus fractures could be classified as blunt force trauma.
- 3 The perimortem trait pattern aided in the classification of all conflicted bone trauma in the case.
- 4 The results from this case study might be of great interest for other anthropologists facing similar challenges during the analysis of burnt remains.

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### Compliance with ethical standards

**Conflict of interest** The authors declare that there is no conflict of interest.

**Ethical approval** This study was approved by the Ethics Commission of the Hospital Universitari de Bellvitge – Institut de Medicina Legal i Ciències Forenses de Catalunya (CEIC, Generalitat de Catalunya, Departament de Salut), in order to comply with the ethical requirements.

**Informed consent** Informed consent was not required for this study.

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## **5. Discussion**

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As previously described, bone will undergo an elastic and plastic phase before fracturing, when subjected to stress as happens in blunt force trauma<sup>9,14</sup>. Since fresh bone is moist and contains water and organic components, it will be stiff and elastic. Dry bone lacks these organic components, which make the bone stiff but brittle<sup>12</sup>. Therefore, dry bone is unable to withstand as much strain or elastic deformation as wet bone and is more sensitive to degradation. This will cause dry bone to fracture immediately after the strength threshold is reached, whereas wet bone will first go through a plastic phase where the bone is deformed before fracturing<sup>9,4</sup>. Therefore, the fracture patterns in dry and fresh fractures will be different<sup>25</sup>.

Notwithstanding, the differentiation between very fresh and very dry fractures might seem fairly easy, in the forensic field most of the recovered bones are in an intermediate stage between very fresh and extremely dry.

Bones that are in an intermediate stage of drying, do not meet these extreme conditions. Therefore, it would be more valuable to acquire additional knowledge about characteristics that are more closely linked to fresh conditions. With an experimental approach, we were able to reproduce fracture patterns similar to those found in traumatic cases and compare them to find specific patterns that may be used in forensic examinations.

## **5.1. Long bones**

Blunt force trauma in long bones occurs very often as a result of traffic accidents, falls from heights, and assault. The long bones included in our study are; the humerus, ulna, radius, femur, tibia, and fibula. The experimental long bone samples - both fresh and dry - showed transverse, oblique, butterfly, and comminuted fractures. Nonetheless, dry bones mainly represent transverse fractures, while autopsy samples are mainly represented



by comminuted fractures. Fresh experimental fractures mainly exhibit oblique, butterfly, and transverse fractures. Cohen *et al.* (2017) performed a study on fresh pig bones with a similar experimental setting. However, they were not able to reproduce butterfly fractures, only oblique, and comminuted fractures<sup>26</sup>. This discrepancy between results clearly supports the importance of the human model to obtain truthful results, in order to resemble autopsy samples. Our results suggest that the type of fracture does not give any information about whether the fracture has occurred peri- or post-mortem since all types of fractures can be found in each experimental group after applying a three-point bending. This conclusion is different for spiral fractures, but our equipment did not allow us to reproduce spiral fractures.

Our results show that fractures impacted at high speed are larger and more often comminuted than those fragmented by a slow loading. The size of the fracture itself seems to be related to the speed of impact; the higher the impact, the larger the fracture. It is known that high-energy absorption leads to more fractures, and therefore creates a higher chance to obtain a comminuted fracture<sup>9</sup>. In reality, the impact force can even be stronger, moreover it will be likely that more than one point of impact will act on the bone. That is why autopsy samples still show more comminuted fractures than the experimental ones. Previously, Cohen *et al.* (2017) related comminuted fractures to axial loading and soft tissue in their animal model<sup>26,27</sup>. The results in our study on the human model, support the hypothesis that there is a relation between the traits and the *intra vitam* pattern involving axial compression and possibly soft tissue.

Finding traits that are related to *intra vitam* circumstances may shorten the perimortem time gap between very fresh fractures and the time that no more fresh traits may be observed.



In literature, there is only one trait that suggests an *intra vitam* fracture. This is when a fractured long bone shows evidence of an axial shortened displacement<sup>28</sup>.

### **5.1.1. Long bone fracture traits**

This study defines six distinct characteristics of which five new traits, that can be valuable to utilise in the distinction between peri- and post-mortem long bone fractures: layered breakage, bone scales, crushed margins, flakes with flake defect, wave lines, and plastic deformation.

These characteristics constitute a specific, unreported perimortem pattern; however not all characteristics must be involved in all perimortem lesions.

#### *5.1.1.a Layered breakage*

Layered breakage is a characteristic layered pattern in the cortical area that is unreported in literature. Our results showed that layered breakage was the most common perimortem trait, being present in almost all autopsy and fresh experimental samples. Only in spiral fractures and fractures at the epiphyses, layered breakage could not be observed. Spiral fractures are caused by rotational forces which is the reason that this feature was not observed in this type of fracture caused by bending. Nevertheless, the presence of a spiral fracture is already a sign that the fracture is most likely a perimortem fracture since this type of fracture is not likely to appear in dry bone fractures<sup>1</sup>.

Furthermore, this trait always appears in the compression side, making it an excellent trait to reconstruct the biomechanical circumstances with more precision and deduce the direction of impact.

During the experiments, the addition of axial compression did not change the results significantly on the incidence of layered breakage, and therefore does not influence the formation of layered breakage. Although it is not a specific trait for *intra vitam* circumstances, it is a clear perimortem trait. Nevertheless, close analysis of layered breakage leads us to observe that this trait appeared deeper in the cortical bone in autopsy samples than in the experimental fresh samples. This might suggest it needs specific *intra vitam* conditions to occur, but during our experiments it was not possible to reproduce them exactly like autopsy samples. It is therefore clear to us that this area deserves further research. We hypothesize however, that these results may be related to the age of the available experimental samples.

An undeniable limitation in this study is the age distribution of the experimental samples. It would be beneficial to also include young people, however undertaking research with human bone samples presents limitations regarding the number of donors who make their remains available to science, and the subsequent restrictions this places on the breadth of the study. It is well known that with increasing age and other age related factors, the cortical layer of the bone becomes thinner<sup>29,30</sup>. Therefore, young bones have a thicker cortical layer and could therefore form several deep layers when the bone fractures. Since only mature and older bones were used in the experiments it may have affected the appearance of the layered breakage, as there is no space available to form layers. Layered breakage in the autopsy samples from individuals of 55 years and over have a similar appearance as the layered breakage from the experimental samples, supporting our theory.

As a side note, a comparable form of layered breakage was found in two dry bone fractures, however the appearance is different. Whereas the layers are parallel in fresh fractures, they are not in dry fractures as they appear in

several directions in one single fracture. This distinction is important when evaluating the characteristics of bone fractures.

#### 5.1.1.b *Flakes with flake defect*

Flakes are very common in both autopsy samples and in fresh experimental samples with compression. Flakes itself are not likely to be recovered since they get lost in the surrounding tissue when fracturing occurs, but they leave an imprint, introduced as flake defect.

Flakes are less common in fractures fragmented without compression and are not present in dry bones. Based on statistical analysis, significant differences between the experimental groups (with or without compression) are observed.

The results clearly show that axial compression is a key factor in flake development. Absence of flakes in dry bone fractures and a similarity in the incidence between autopsy and compression group supports the hypothesis that flakes may be related to musculoskeletal activity and the presence of surrounding tissue. The interpretation of blunt force trauma will be more accurate if flakes can be considered as *intra vitam* markers since they are abundantly present in fractures.

#### 5.1.1.c *Wave lines*

Wave lines are less common but could be reproduced in experimental fresh bones with high speed, but not in dry bone samples and fresh samples when fractured under slow speed. The edges of fractured dry bones are too jagged

to show any wave lines. Wave lines can therefore be specified as a perimortem trait. In fresh bone samples, no differences are found between fractures with or without axial loading. In accordance with our assumption, this indicates that wave lines are a perimortem trait, but not necessarily an *intra vitam* trait.

#### 5.1.1.d Crushed margins

Crushed margins are very rare in autopsy samples. We suggest that this trait needs a very specific mechanism, probably related to muscular action causing a collision of the fracture edges during fracturing.

Sequential to this hypothesis, an attempt was made to reproduce them by adding springs to the experimental setup, so a recoil can take place when the fracture initiates.

The results however, were positive in only two experimental samples where compression was applied. Clearly, this result is insufficient to test our hypothesis. Furthermore, they are not observed in samples fractured without compression, and differences between both experimental groups are not statistically significant. The mechanism behind the trait suggests a complex fracture process, not only compression, but also the presence of surrounding soft tissue, different impact mechanics, bone and/or individual variables may have an influence on their formation. Nevertheless, crushed margins deserve further research to conclude that they could be used as an indicator of trauma under *intra vitam* conditions.

#### 5.1.1.e *Bone scales*

Bone scales did not appear in any of the experimental samples. In autopsy samples, they do not occur often either. Bone scales were observed in experimental samples applying slow loading and axial compression to break the bone. Differences in impact speed may suggest that these circumstances play an important role in the development of bone scales. It would also offer an explanation as to why this trait does not appear more often in autopsy samples, since bones are usually broken on fast impact.

#### 5.1.1.f *Plastic deformation*

The final trait, plastic deformation, appeared in all fresh bone samples therefore it is a clear perimortem trait. It is significantly more common in the experimental group without axial compression compared to the other groups and there are no significant differences between autopsy and compression samples. We hypothesize that axial compression and surrounding soft tissue limit the bending freedom and therefore cause less plastic deformation. During the experiments without compression, there is no compressive force that keeps the bone immobile, causing more bending and subsequently, more plastic deformation.

#### 5.1.1.g *CT analysis*

From the individual slides generated by a CT scan, a 3D model could be reconstructed of the bone samples. Layered breakage could be clearly visualised on these 3D models and the layered pattern could easily be identified.

Flake defects could be visualised as irregularities on the bone surface that stand out by the shadows they create. Wave lines could also be visualised on the long margins of the fractures, although the resolution is quite low for this small and detailed trait. Additionally, we could observe crushed margins (plastic deformation) in one explored bone sample, however in other examined bone samples it was not visible. Severe plastic deformation could be visualised in both the individual slices as the 3D reconstruction. Apart from these perimortem traits, we also observed tension lines on both the individual slices and the 3D reconstruction.

Although not all perimortem traits can be visualised with CT imaging, it is still a useful tool for trauma analysis as it is a fast technique to get a rapid overview of the trauma.

#### *5.1.1.h Application in the field: Fractured and burnt remains*

Evaluating the new morphological features in a fracture will allow us not only to time the injury more accurately but hypothesize if the fracture occurred at the moment of time of death, when soft tissue and muscles are involved.

In a real forensic case, a combination of perimortem blunt trauma and thermal damage was observed on the victim because of a car accident and post-crash combustion. Perimortem fractures were located at the diaphysis of the left femur, right tibia, and fibula. Except for a butterfly fracture on the femur, the other fractures were comminuted. All of them presented perimortem traits on the margins, with the presence of layered breakage, crushed margins, wave lines, and numerous flake defects. This damage was considered to be related to the trauma circumstances due to the car accident.

This shows that some of the perimortem traits on long bone fractures can still be distinguishable in charred bones and thus may aid in the timing of fractures, suggesting that the fractures occurred when soft tissue was still present.

### **5.1.2. Fracture staining**

Both methods use ink penetration into the lesion and offer both an improvement of visual recognition of the existing fractures. Nevertheless, the fracture lines were better visualized when relying on capillary forces since the colour penetrates only the fracture lines. The results of the cooking method were not always satisfying, and results depended on the condition of the bone. When the bones were very greasy, the ink did not enter the fracture lines in contrast to the other method that relied on capillary action, where this was not an issue.

This method could be used during the examination of fracture lines in bone fractures and will aid in the understanding and interpretation of the biomechanics behind a fracture. Furthermore, it can be used during court sessions to present photographic evidence to a jury as well as in class for educational purposes.

## **5.2. Ribs**

Rib fractures are commonly involved in forensic cases such as traffic accidents, falls, and cases involving blows to the chest<sup>31-33</sup>. Nevertheless, biomechanics of rib fractures are poorly understood, therefore the interpretation of rib fractures remains a challenge for forensic anthropologists<sup>34,35</sup>. Ribs are part of a closed structural and mechanical system, with a distinct architecture different from long bones. Experimental research to fractured rib patterns with respect to loading conditions are scarce, probably due to the difficulties in developing a method to systematically analyse the injury patterns, and the difficulty in obtaining human rib samples<sup>33</sup>.

### **5.2.1. Rib fracture traits**

This research provides new knowledge on the morphology of the injury pattern of the anterior rib arch. This region, together with the lateral part of the rib, is frequently associated with direct blows to the chest, lateral compaction of the thorax or cardiopulmonary resuscitation (CPR)<sup>36</sup>.

We report six preliminary macroscopic traits that are present in autopsy fracture samples of the anterolateral rib arch: peels, folds, longitudinal lines, incomplete fractures, differential fracture edges on the internal and external part of the rib fracture, and plastic deformation.



### 5.2.1.a *Perimortem traits*

Timing rib fractures still is an uncertain diagnostic in most forensic anthropological reports. In spite of published descriptions to differentiate peri- from post-mortem fractures, they are mainly intended for long bones<sup>7,37-39</sup>. Given the differential architecture and structure between long bones and ribs, these known rules are difficult to apply. It would be more valuable to obtain additional knowledge about perimortem fractured ribs, since it could be the base to approach this challenge.

With this purpose, the abovementioned perimortem rib fracture pattern was compared with the fracture pattern of experimentally fractured ribs from donors. Three-point bending trauma tests were simulated on fresh and dry anterolateral individual rib samples. The variability of rib fracture patterns was analysed with respect to two loading conditions, fast speed (3 m/s) and slow speed (160 mm/min (= 2,7 mm/s)).

The results show that incomplete fractures, plastic deformation, peels, and folds are not present in dry bones, independent of the loading conditions. It suggests that these traits are related to fresh/wet bone conditions, or perimortem conditions. In contrast, longitudinal fractures and differential appearance of the margins appear in both fresh and dry specimens, excluding them as perimortem indicators. The differences may be explained by differences in bone composition between wet and dry bone, which influences the fracture biomechanics and thus the fracture patterns.

Considering the nature of incomplete fractures, plastic deformation, peels and folds, it is likely that water and organic material are required to reproduce them in a rib fracture<sup>12,29,32</sup>.

### 5.2.1.b *Age and bone degradation*

The results also show that this pattern is associated with age and trauma biomechanics, giving clues to reconstruct the trauma circumstances. Evidence concerning the influence of the age in rib fracture pattern seem to be varying in literature<sup>35</sup>. According with our results, longitudinal fracture lines and peels on the cortical rib surface have been found more probable in the young age group. Additionally, there is a statistically significant increased risk of getting complete rib fractures in older individuals compared to younger ones. This evidence is in contrast to Love and Symes<sup>35</sup>, who did not find major incidence of complete rib fractures between various age categories.

Considering our observations however, chronological age influences the mechanical behaviour of bone as originally described in literature. Therefore, we hypothesize that the relationship between complete fractures, longitudinal fracture lines, peels and age may be explained by aging changes in the architecture and composition of the bone, particularly since bone is more elastic in young people and brittle in the elderly<sup>8,9,21,31,39,40</sup>.

### 5.2.1.c *Trauma circumstances*

Some of the macroscopic traits associated with rib fractures may be indicators of the trauma circumstances. The analysis of these traits shows that complete fractures, folds, and longitudinal fracture lines appear to be more likely to be present in accidents caused by falls, car accidents or train collisions than injuries induced from CPR.

Additionally, folds and differences in fracture edges might provide information about the fracture mechanism. Folds could not be reproduced in dry ribs. In the fresh experimentally fractured ribs, folds could only be observed on the compression side. Furthermore, experimental reproductions on dry and fresh ribs under fast loading conditions showed differences in the fracture edges between the compression and tension side: the compression side appeared irregular, whereas the tension side appeared smooth and regular<sup>41</sup>. This might suggest that these traits could indicate the direction of impact. The smooth fracture edge at the tension side was earlier described by Kieser et al. (2013) in a study with 15 pig ribs. However, they did not describe the irregular compression side and could not find this trait in fractured dry bones. Moreover, no implications were provided by them on how this trait could help to interpret the fracture pattern in relation to the fracture mechanism during forensic trauma analysis<sup>42</sup>.

We could find this trait in 51,7% of our human perimortem rib samples and provide an indication of the forensic implication of this trait, which takes the study of Kieser et al. (2013) a step further.

All fresh experimentally fractured ribs that were broken under slow loading conditions resulted in incomplete fractures with only a fracture in the cortex of the compression side. Since the cortex on the tension side was intact in all fresh specimens fractured with slow loading, no distinction could be made between the fracture edges of the compression and tension side.

Literature regarding the biomechanical properties of bone state that bone is usually stronger under compressive stress compared to tensile stress, which means that bone usually fractures at the tension side prior to the compression side<sup>5,8,21</sup>. However, in our study all experimentally fractured fresh rib samples, including the fresh ribs that have been fractured with the

loading cell on the internal part of the rib, failed at the compression side prior to the tension side. Previous studies have already concluded this, and incomplete fractures that failed at the compression side prior to the tension side were identified as ‘buckle fractures’<sup>32,34,42</sup>. The finding that ribs also fractured first at compression when the loading cell was put on the interior part of the rib might suggest that ribs always fracture first at compression, regardless of the location of the compressive stress on the rib. In order to obtain further knowledge and to test this hypothesis, future research is recommended with a larger sample size and improved experimental settings in which real trauma conditions can be simulated.

#### 5.2.1.d *Intra vitam* traits

Lastly, peels did not appear in any of the experiments, independently of the rib state and loading conditions. This trait was only observed in autopsy specimens. Intriguingly, the tests were not able to reproduce peels, even in fresh and fast loading conditions. It suggests that its presence may be related to the trauma circumstances, which are difficult to reproduce in a simplified *in vitro* experimental test. In the case of ribs, it is difficult to link the traits with *intra vitam* conditions such as muscular contraction, as hypothesized in some of the traits observed in long bones but the importance of soft tissue and the periosteum should not be discarded<sup>13,35</sup>. Analysis of peels deserves further research to obtain more in-depth knowledge about the exact mechanism that is needed for its occurrence.



## 6. Conclusions

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*“Don't let anyone rob you of your imagination,  
your creativity, or your curiosity.  
It's your place in the world; it's your life.  
Go on and do all you can with it,  
and make it the life you want to live.”*

Mae Jemison

Based on this extensive research, several conclusions can be drawn which are related to the predetermined research questions that were the backbone of this study.

1. In long bones the perimortem fracture pattern is defined by the presence of: layered breakage, bone scales, crushed margins, flakes with flake defect, wave lines, and plastic deformation.
2. Layered breakage is the most common trait, encountered in all fresh perimortem fractures, except for spiral fractures, and fractures at the epiphyses. Moreover, layered breakage is only observed in the compression side of a fracture and therefore an interesting feature to infer the direction of fracturing, making it easier to deduce biomechanical circumstances.
3. Flakes are the second most common trait and are significantly more common when fracturing occurs while applying a compressive force along the long bone axis. We therefore hypothesize that flakes are an *intra vitam* fracture marker. The presence of *intra vitam* traits can make it possible to shorten the perimortem time gap, allowing us to suggest a change in the fracture timing paradigm. Another possible *intra vitam* trait is plastic deformation since there is less plastic deformation when axial compression is added.

4. A CT scan can be a fast and non-invasive aid in perimortem trauma analysis. Tension lines can easily be visualised and their path can be traced. Furthermore, layered breakage, flake defects and plastic deformation can be observed on the 3D reconstruction of the slices.
5. Layered breakage, flake defects, and wave lines preserve quite well and can even be observed on fractures that are severely damaged by a fire. Making it possible to differentiate perimortem fractures from post-mortem damage and events.
6. The use of Talens® Black Indian ink on a fracture can help to visualise fracture lines, especially when relying on the capillary forces of the fracture lines.
7. In ribs, the perimortem fracture pattern is defined by the presence of: peels, folds, incomplete fractures, and plastic deformation. Additionally, longitudinal lines and differential fracture edges on the internal and external part of the rib fracture were also defined but are not specific perimortem traits.
8. The rib traits are related to different forensic variables. Aging changes in the architecture and composition of the ribs influence the incidence of complete fractures, longitudinal fracture lines, and peels. Complete fractures, folds, and longitudinal fracture lines occur in high impact trauma circumstances while incomplete fractures occur in low impact trauma circumstances. Differential fracture edges and folds indicate the direction of impact and thereby provide information about the fracture mechanism.





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