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Review Articles

The Forensic Value of Long-Bone Diaphysis: A Scoping Review Across Identification, Pathology, Trauma Analysis, and Biomolecular Evidence

Nasser F AlSunbul*

Trauma Surgery Department, Trauma Center, King Saud Medical City, Riyadh 12746, P.O. Box 7794, Saudi Arabia

***Corresponding author:** Nasser F AlSunbul, Trauma Surgery Department, Trauma Center, King Saud Medical City, Riyadh 12746, P.O. Box 7794, Saudi Arabia **mail ID:** NasserAlSunbul@outlook.com

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Abstract

Long-bone diaphyses are central to forensic work given their cortical shafts which endure when other bones degrade. Macro and microstructural features preserve metric, histological, and biomechanical parameters for identification, including stature, sex, and population affinity.

Imaging advances, including CT, micro-CT, and 3D model reconstructions as well as the rise of machine-learning models, have even improved precision and enabled analysis of fragmentary remains. Diaphyseal pathology and pathological changes (osteomyelitis, neoplasms, metabolic and hematologic disease) leave cortical signatures further correlating with antemortem records. Trauma to shafts shows diagnostic patterns across blunt, sharp, and ballistic mechanisms, distinguishing perimortem injury from postmortem damage. Beyond morphology, cortex preserves DNA, proteins, and isotopic profiles under taphonomic stress. Differential remodeling confers complementary value: femoral cortices archive long-term diet and mobility, whereas tibial cortices capture recent exposures. This review synthesizes and underscores the evidence on diaphyseal value across identification, pathology, trauma, and biomolecular applications, highlighting advances, limitations, and femur-tibia comparisons in forensics settings.

Keywords: Forensics, Bone Diaphysis; Postmortem Exam; Trauma Analysis

Introduction

Long human bones, particularly the humerus, femur, and tibia, play a central role in forensic anthropological investigations. These elements are among the most commonly preserved skeletal remains due to their dense cortical diaphyseal structure, thus in such circumstances where other bony remains have degraded, they become indispensable [1].

Long bones contain the diaphysis, which is defined as the elongated shaft of a long bone, it is biomechanically specialized to withstand axial loading and bending forces; however, in a forensic aspect of study. It represents a durable archive of metric, histological, pathological, and biomolecular information critical for personal identification, trauma analytics, and reconstructing anthropology [2].

The forensic value of diaphyseal bone lies in its multi-dimensional applications ranging from biological profile perspectives, the dimensions, and the microstructural features of the diaphyses can provide multiple insights into age, sex, stature, and even ancestry. While diaphyseal cross-sectional geometry informs sex estimation and biomechanical adaptation, Histomorphometric studies can reveal the age-related remodeling processes [3,4].

Recently, current advances in imaging modalities, including radiographs, CT-based, and 3D surface reconstructions, have enhanced precision of the morphometric analyses and enabled the development of multiple machine learning approaches to diaphyseal identification [5].

Diaphyseal bone abnormal alterations, such like changes either pathological or traumatic, can also further extend its utility. Diaphyseal bone abnormal alterations, such as changes that are either pathological or traumatic, further extend its

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utility in identification. Diseases affecting the diaphysis, such as osteomyelitis, neoplasms, and other metabolic disorders, can have an identifiable cortical signatures, which may aid in both the correlation of clinicopathological elements and forensic casework. Moreover, diaphyseal fractures bear a distinctive biomechanical fingerprints that assist in distinguishing between perimortem trauma, postmortem damage, and pathological weakness, hence enabling the interpretation of both the cause and manner of injury [6,7].

Exceeding morphology, the diaphyseal cortical bones are an important biomolecular reservoir, as they offer a high-density preservation for DNA, proteins, and isotopic signatures, even under challenging taphonomic conditions such as hot or arid environments [8].

Comparative studies exhibit the actual differences between the femoral and tibial cortices in terms of how the biomolecule works and survives, underlining the relative strengths in use for forensic sampling strategies [9].

Diaphyseal analyses are like any other elements of surgical or forensic analysis, not without limitations. With events like diagenesis, environmental degradation, and taphonomic overprinting may lead to an obscure true biological signals, complicating the ability of an accurate interpret [10].

Thus, while long-bone diaphyses still play a role as a valuable forensic substrate, their use demands an integrated approach that includes osteological, pathological, biomolecular, and environmental angles.

This scoping review tries to synthesize the current evidence on the forensic value of long-bone diaphyses across four principal domains: identification, pathology, trauma, and biomolecular analysis. An emphasis is also placed on recent advances, challenges, and comparative evaluations of femoral and tibial diaphyses.

Anatomy and Biomechanics of Long-Bone Diaphysis:

The diaphysis of long bones, including the femur, tibia, and humerus, is a tubular structure designed to support and transmit the mechanical loads it encounters. Its morphology mirrors an evolutionary balance between lightness for mobility purposes and strength for the capacity to bear load [11].

Macrostructure

The diaphysis is cylindrical structure, composed of dense cortical (compact) bone enveloping a medullary cavity, providing a high strength-to-weight ratio, but cross-sectional profiles vary by bone and anatomical region. For instance, the femoral shaft demonstrates a circular-to-oval cross-section that is optimized for multidirectional stress resistance, whereas the tibial shaft exhibits a triangular cross-section, enhancing resistance against bending in the sagittal plane. in contrast to the humerus, which have a comparatively less robust, that reflects adaptations to upper limb mobility rather than locomotor stress [12].

Microstructure and Remodeling

On a microstructural and microscopic scale, cortical bone consists of osteons, which are cylindrical units of lamellar bone organized around vascular canals. These osteons can undergo a continuous remodeling, balancing the resorption and deposition, which both strengthen bone and encode the life history markers such as age and activity levels. with osteon histological morphology providing critical parameters for age estimation [11].

Biomechanical Function

The diaphysis parts of long bones acts as a mechanical hinge during movement, which distributes muscle-induced forces, while withstanding the axial compression, bending, torsion, and also shear forces. The femur absorbs a high compressive and also torsional loads during locomotion, likewise, the tibia supports the body load and distributes forces from the knees to the ankles, while the humerus facilitates mobility and range of motion in the upper extremity. These diverse biomechanical roles illustrate how the cross-sectional geometry (CSG) analysis of the diaphyseal shafts are an influential indicator for sex, conditioning, and specific population and demographic adaptations [5].

Forensic Implications

An understanding of the biomechanics of the diaphyseal is valuable to the forensic anthropology. Primarily, its cortical thickness and relative robustness affect its preservation potential, with the femur often surviving better than the tibia in burial contexts. Second, cortical bone distribution determines the reliability of biomolecular recovery, since denser diaphyseal regions yield higher DNA and collagen content than other trabecular bone [13].

Another Implication lays in trauma interpretation which require an awareness of mechanical loading mainly in the fields of bending-induced fractures, spiral fractures, and ballistic disruptions which all may interact with diaphyseal anatomy in foreseeable methods.

Finally, the biomechanical uniqueness of each shaft contributes to its unique value in identification. For instance, morphometric properties of the tibial shaft can inform sex estimation in specific populations, while femoral shaft length and curvature are robust correlates of stature. All together, these features make the diaphysis both a resilient and informative substrate, a Fundamental value in forensic reconstructions [14-16].

Forensic Value in Identification

The identification of an unknown human remains is the cornerstone of forensic investigation and anthropology, and long-bone diaphyses serve among one of the most reliable substrates for this objective. Their dense cortical structure, comparative resistance to diagenetic processes, and their relative abundance both in archaeological and forensic contexts make them indispensable [5,8,9]. Many complementary approaches are used for the identification; those previously reported are metric and morphometric analysis, radiographic and 3D imaging, biomolecular

applications, and the emerging use of machine learning models.

Stature Estimation

Stature estimation is a well-established application since the oldest of times to use in diaphyseal metrics. Long-bone length, specifically in describing the femoral and tibial shafts, shows a strong correlation with body height across populations. Regression equations derived from diaphyseal dimensions allows the forensic practitioners to reliably predict stature with a reasonable accuracy [11].

Furthermore, population-specific assessments remain necessary, as historical patterns and environmental variables influence bone generation. Virtual morphometrics, including CT-derived reconstructions, now provide a way of a non-invasive means of measuring fragmentary shafts and incorporating geometric morphometric approaches into stature estimation [17].

Studies now emerge on a comparative analysis use of tibial osteometry to refine stature estimation methods, available evidence on approaches recently developed for more efficient identification using whole bone study and inclusion of age-correction factors [18].

population-specific regression formula also remains a cornerstone in accurately identifying stature across different populations, a study correlated the length of humerus bones, have strong correlations with stature and produced reliable regression equations applicable in a forensic context in the form of disaster victim identification. further emplying the need of a tailored estimation methods to every population [19,20].

Sex Estimation

Sex estimation from diaphyseal bone uses the cross-sectional geometry (CSG), cortical thickness, and morphometric indices. The femoral shaft offers high discriminatory value given its robusticity and curvature, while the tibial shaft exhibits dimorphism in dimensions such as midshaft diameter and robusticity index. Histomorphometry can provide further resolution, as osteon size and density exhibit more sex-related trends. Emerging studies that uses machine learning models especially while also integrating 3D morphometric data, have demonstrated accuracies exceeding the traditional discriminant function analyses [11,14,21].

Literature now finds that the epiphyseal diameters of both femur and tibia achieve classification rates that exceed 90% [22].

Histomorphometric measurements further indicates the predictive value of secondary osteon counts and canal widths, with stepwise discriminating functions providing accuracies of over 70%, even when applied to fragmented cortical bone [23].

Ancestry and Population Affinity

Advancements in geometric morphometrics, coupled with shape statistical analysis, enhance the discriminatory value of the diaphyseal morphology in differentiating between populations. Although an ancestral estimation using bone diaphyseal is less accurate than cranial methods, the literature indicates that tibial and femoral shaft morphology encodes biomechanical adaptations reflective of ancestry. For instance, cross-sectional shape variation correlates with locomotor and climatic adaptations, which may indirectly inform population affinity [5].

Radiographs and the Use of 3D Technology

Radiographic imaging of diaphyseal shafts provides an essential morphometric data. As Postmortem (CT) became a central and essential method in forensic profiling, enabling the assessment of trabecular distribution, assessment of cortical thickness, and morphology. Micro-CT further enhances this by capturing more fine details such as cut marks, osteon structures, and fracture patterns at voxel resolutions below 126 μm, providing morphometric precision and a courtroom-ready visualizations to be used. CT and MRI both allow the reconstruction of the internal canal morphology, cortical thickness, and trabecular distribution. Three-dimensional (3D) surface scanning has expanded the possibilities for fragmentary remains, enabling the virtual reconstruction and measurement. Radiographic analytics, particularly in the field of trabecular patterning in the metaphyseal-diaphyseal transition zones, can also assist in age estimation. These imaging approaches preserve bone integrity while generating datasets suitable for computer-aided remodeling and AI applications [12,24-26].

Artificial Intelligence and Machine Learning

Machine learning algorithms trained on large morphometric datasets with the integration of AI in forensic anthropology has revolutionized the use of diaphyseal data for a more accurate sex and stature than classical regression or discriminant functions. Convolutional neural networks implemented on CT and 3D scan images automate landmarking and feature extraction, minimizing interobserver error [13].

Fragmentary and Commingled Remains

Diaphyseal cortical bone is also a reliable source for biomolecular analyses, which can supplement morphometric approaches in fragmentary cases. In events such as mass disasters, archaeological sites, or secondary burials, morphometric variables or histological parameters from even the smallest cortical fragments can yield sufficient measures for partial identification [10].

Applications from the Literature

Real-world applications illustrate that even when cranial or pelvic bones are deficient, diaphyseal shafts remain viable guides for reconstructing biological profiles. Emphasizing the reliability of diaphyseal analysis in forensic settings. For instance, femoral shaft dimensions have resolved identification in confusing remains following mass disasters, while tibial fragments have been essential in war crime investigations due to their preservation in challenging depositional environments [21].

As cross-sectional geometry (CSG) evaluations emerge and highlights the strengths and limitations of diaphyseal methods as

it can yield biomechanical and sex-related insights it is of a limited use in the estimation in age or stature estimation given inconsistent results Particularly in the context of diagenetic changes [27].

Likewise, inclusive regression models for diaphyseal lengths in children increase utility but still maintaining large prediction ranges across various groups, both occur with the developments of long bone-based sex determination, that shows flexibility of diaphyseal data, with tibial and fibular measures assessed by logistic regression, obtaining an accuarcy as high as of 91%, making them valuable alternatives when dimorphic skeletal features are deficient [28,29].

which complements CT-based measurements of humeral and femoral head diameters, that have achieved classification accuracies exceeding 92% [30].

Although diaphyseal shows strengths in identification methods that mainly lie in their resilience, broad application, and compatibility with traditional osteological and the new emerging technological methods, limitations remain a major setback with wide error margins in stature estimation, population differences and diversity in sex estimation, and being inferior in the setting of ancestry estimation compared to pelvic or cranial approaches.

Additionally, environmental degradation, diagenesis, and pathological remodeling may distort morphometric and histological metrics [12].

Disease Markers in Diaphyseal Bone

The diaphyseal of long bones importace in forensic settings does not only imply as it being the structural support for the skeleton, even while pathological processes affecting the cortex and medullary canal has its own morphological, radiological, and histological signatures [6].

Infectious Lesions and Osteomyelitis

One of the most frequently encountered infections in the diaphyseal bone is Osteomyelitis. Its chronic forms manifest as a combination of cortical thickening, involucrum formation, and cloacae, which also often accompanied by an irregular periosteal reactions. A Radiological manifestation of the infection is also apperant on the form of sequestra that appears as dense fragments surrounded by granulation tissue that is radiolucent. Being aware of these changes can be helpful in forensic settings and assist in recreating the individual's medical history and even link remains to hospital records or surgical records [6,10].

Tumors and Neoplasms

Although primary bone tumors and metastatic disease usually affect the metaphyseal it can have their extension to the diaphyses region, which typically appears in the form of an aggressive periosteal reactions such as the "onion-skin" or "sunburst" patterns in Osteosarcoma, and as lytic or mixed lytic-sclerotic lesions in metastatic tumors appearing on the femoral or humeral shaft. Forensic significance of primary and metastatic bone tumors in diaphysis resides in the correlation of the lesions with the antemortem clinical records and radiographs or the appearance of any antemortem Pathological fractures [5, 14].

Systemic, Hematologic, and Metabolic Disorders

Metabolic and Hematological conditions produce their own distinctive features in the diaphysis, complicating but also enhancing recognition in a forensic setting. rickets, osteomalacia, and osteoporosis manifesting as conditions of metabolic diseases in a deformed cortex, thinning, or fragility. with Rickets and osteomalacia, mainly characterized in bowing of long-bone shafts, a widened metaphyseal-diaphyseal junction, and an irregular mineralization of the cortex. On the other hand, osteoporosis lowers the cortical thickness and alters its biomechanical profile, potentially influencing age or sex estimation if not accounted for [12,21].

Hematological disorders, including sickle cell disease and thalassemia, likewise leave their recognizable diaphyseal markers. With Cortical thinning, marrow hyperplasia, and the appearance of endosteal scalloping are among the most commonly observed manifestations [11].

Such signatures, when matched with medical records, aid in the personal identification and also carry a population-specific value given the distribution of these conditions.

Disease Markers and Forensic Implications

Previously mentioned pathological lesions in the diaphysis serve as potential markers of individualization when being correlated with antemortem radiographs, surgical documentation, and medical records. Not only limited to that, they may also inform the cause of death or even contribute to lifestyle reconstruction, as the appearance of systemic conditions such as metastatic carcinoma or chronic osteomyelitis provides a valuable insights into the overall health status and survivability. Nonetheless, to avoid misinterpretation of genuine pathological signatures from postmortem changes. to avoid inaccurate conclusions of personal identification [9].

Diaphyseal Implants and its role in Tracing Identity

The Use of various Orthopedic implants used in management of cases either with intramedullary (IM) rods or fixation plates have an extended role upon their therapeutic use and could be of a forensic significance

Serial Codes and Manufacture Identification

Implants have manufacturer-imprinted engravings, containing batch numbers and serial codes that are still legible long after implantation. In the field of forensic pathology, such identifiers provide a clear pathway to accurately individualize cases by enabling investigators to trace implants back to the suppliers, distributors, and hospital or surgical records. Such capacity is

invaluable in settings where the DNA is degraded or in events that the skeletal morphometrics alone cannot yield a definitive identity [10,31,32].

Metallurgical and Radiographic Signatures

Implants can have a distinctive and unique radiographic signature. Like for instance, the metallurgy of IM nails being (stainless steel or titanium alloys) can be matched against antemortem radiographs with either the locking mechanisms or screw configurations used, as well as the chosen surgical approach, and even the surgeon's technique can be compared with Operative Records. Although these measures require a presupposed identity ("closed population" scenarios) it can help in scenarios such as a mass disaster or conflict areas, where remains are found fragmented, given that the implants can survive water and fire immersions and other extreme conditions, making them a valuable component in identification [10,32].

Applications from the Literature

Reported literature further demonstrates this utilization, as a one case the extraction of an intermedullary nail that maintained a lot number that traced back and gave confirmation of victim's identity in an absence of soft tissue or a recoverable DNA [6].

Another utilization of Orthopedic Implants in identification was done on the aftermath of the 2011 East-Japan earthquake and tsunami mass-disaster where retained implants within cremated remains helped in the positive identification of two cases [32].

Furthermore, batch numbers engraved on an implant which was traced back to hospital records and yield an accurate identification without the need of DNA Analysis [33].

Differentiating Pathological Changes from Postmortem Alterations

Long-bone diaphyses and their dense cortical structure often overcome alterations from changes produced by environmental, biological, or chemical agents after death, but they also remain highly susceptible to taphonomic processes [6].

Importance of Accurate Differentiation

Failing to distinguish postmortem alteration from a pathology or the contrary can alter innumerable forensic conclusions, particularly in age estimation, trauma analysis, or identification. For instance, root etching a femoral diaphysis may mimic the periosteal reaction that results from chronic osteomyelitis, while soil erosion might simulate metastatic lytic lesions [10].

Postmortem Microbial Bioerosion vs. Osteomyelitis

Cloacae or involucrum that are associated with osteomyelitis, is it often being confused with the tunneling patterns within cortical bone, resembling microbial bioerosion under microscopy [6].

Histopathological examination, mainly the study of osteon morphology and the presence of inflammatory infiltration, remains a key marker for differentiation. Advancements in imaging, including micro-CT, further distinguish biologically induced remodeling from abiotic diagenesis [11].

Recent work highlights that forensic timescales in regard to bone diagenesis can occur much shorter than previously speculated.

Bioerosion, which was traditionally associated with archaeological material, has been observed within weeks to months after deposition, even before full skeletonisation, and is now recognized as a major pathway for cortical degradation [34].

Weathering produces surface cracking, exfoliation, and cortical flaking, may be misinterpreted as indicators of osteoporosis or other metabolic bone diseases. In contrary, true osteoporosis leads to thinning and uniform cortical porosity, often with systemic distribution across many skeletal elements [21].

Experimental taphonomic studies have found that the moisture loss and the occurrence of collagen degradation drive most of the transition from ductile "wet" bone to a brittle "dry" bone. As bone goes and further dehydrates, it stiffen and declines in resilience, resulting in a stepped or jagged fracture typical of a weathered bone. Moreover, burial environments may preserve the moisture longer, delaying such transition, while exposed or aquatic settings that further accelerate collagen breakdown and bioerosion [35].

Distinguishing Trauma from Postmortem Damage

Fracture morphology in long-bone shafts provides another diagnostic challenge.

Perimortem fractures typically display oblique or spiral morphologies with plastic deformation, hinging, and sharp fracture edges. Different from the appearance of postmortem fractures, which tend to appear as a transverse, brittle, and also lack plastic deformation due to loss of the collagen elasticity and bone resilience mentioned before [12].

Taphonomic effects such as soil compaction, freeze-thaw cycles, and carnivore gnawing further processes to a complicate interpretation. Making recognition of differential coloration along fracture margins an additional diagnostic clue to work on. Features such as clot residues or early woven bone deposition confirm vital (antemortem/perimortem) trauma. Rather than the stable isotope and histomorphometric studies that further reveals remodeling is not uniform across cortical bone: endosteal regions remodel faster and may reflect more recent life history, whereas periosteal bone retains older signatures. Such intra-bone variability reinforces the need to consider both biomechanical and microstructural evidence in trauma analysis in a forensic setting [5,34-36].

Root Etching, Insect, and Animal Activity

Root etching is characterized by meandering, irregular grooves on cortical surfaces, which may mimic the periosteal reaction occurring with inflammation. Insect activity can maifiest as linear

striations, while rodent gnawing marks a parallel, and chiseled grooves. The absence of remodeling or woven bone responses distinguishes these findings from pathological processes [21].

Applications from the Literature

Failure to differentiate pathology from postmortem alteration can mislead both the clinical and legal interpretations. For instance, a diaphyseal lesion that was initially interpreted as an osteolytic metastasis was later identified as an extensive soil erosion, altering conclusions regarding the cause of death. Similarly, misclassification of perimortem trauma and dry bone fractures may affect the distinction between homicide and postmortem disturbance [13].

Trauma Analysis in Long-Bone Shafts

The long-bone diaphysis, with its robust cortical structure and apparent biomechanical responses, serves as one of the most informative skeletal regions for trauma analysis. The interpretation of diaphyseal fractures plays a pivotal role in distinguishing perimortem events linked to cause of death from postmortem alterations and in reconstructing mechanisms of injury. Understanding fracture morphology across blunt, sharp, and ballistic trauma is central to forensic casework [37].

Forensic Importance of Diaphyseal Trauma

Contradiction to cranial or pelvic bones, the shafts of the femur, tibia, and humerus resist postmortem degradation for extended periods, preserving fracture morphologies even when exposed to adverse taphonomic conditions. The dense cortical composition enables the accurate assessment of trauma mechanics, including loading direction, energy transfer, and fracture timing. This makes the diaphysis a crucial archive of traumatic signatures in both forensic anthropology and clinical forensic medicine [12].

Experiments in Fracture Studies showcased that diaphyseal shafts are especially valuable for assessing energy thresholds of trauma. Impact research has also concluded that high-energy bending forces constantly generate butterfly along with comminuted wedge fractures, illustrating the predictable biomechanical behavior of diaphyseal bone [7].

Differentiating Perimortem from Postmortem Fractures

A foundational step in trauma analysis involves establishing whether a fracture occurred during life or around the time of death (perimortem) in opposed to after death (postmortem) changes.

Perimortem fractures typically appears as a spiral or oblique fracture, plastic deformation, and hinging. The bone's alive moisture and collagen content impart elasticity, producing smooth fracture edges and radiating fracture lines. In addition to bone tears, delamination, and break-away notches, can further confirm a trauma of perimortem origin. These changes rarely occur in postmortem dehydrated bone that is characterized by jagged, right-angled cracks, brittle fragmentation, and the lack of plasticity [5,7].

Advancements also highlight the role of micro-CT and high-resolution imaging use in differentiating trauma signatures. As micro-CT permits 3D reconstruction of bone saw marks and cut features, enabling the measurements of kerf width, wall angles, and striation regularity even in remains that are partially burned or fragmented. Moreover, experiments on heat-altered bone show that pre-existing saw marks and kerf traits can persist after exposure to temperatures reaching up to 800 °C. Complementary research on heat-induced bone color changes provides additional contextual data, as specific hues (brown, black, grey, calcined white) correlate to temperature ranges and can assist in restoring burning conditions [38-40].

The diagnostic challenge arises in partially dried bone, where perimortem and postmortem features might overlap. Histopathological examination of the microcracks and scanning electron microscopy (SEM) can improve resolution, while chemical and elemental techniques, for instance, Laser-Induced Breakdown Spectroscopy (LIBS) with neural networks, provide additional assistance by differentiating biogenic from diagenetic signals. Though originally developed and mainly used in archaeology, these approaches holds a promise for forensic trauma analysis, particularly in commingled or mass disaster occasions [12,41].

Blunt Force Trauma in Long Shaft Bones

Blunt force trauma (BFT) produces a distinctive fracture morphology depending on the load of direction and magnitude. Compression loading (e.g., vehicular impact) yields and produces transverse or short oblique fractures, while bending tension forces from falls or blows outcomes in spiral or butterfly fractures, especially when the femoral shaft is involved. Torsional loading, however, produces spiral fractures with helical propagation. Perimortem blunt fractures are distinguished by smooth surfaces and sometimes differential staining due to exposure of fresh cortical tissue. In forensic contexts, these patterns provide insight into cause, force vector, and number of blows. Recent work further confirms that butterfly and wedge fractures in diaphyseal shafts are highly consistent under controlled high-energy bending loads, pointing out their diagnostic value in reconstructing impact dynamics [7,12,42].

Sharp Force Trauma

Sharp force trauma occurs in a less common mean in diaphyseal shafts compared to flat bones but still remains crucial in homicide investigations. Machetes, axes, or heavy blades produces a V-shaped notches, kerf walls, and striations. Unlike cortical surface erosion caused by taphonomic processes, sharp force lesions often show a consistent kerf morphology and a localized impact zones. The microscopic evaluation may identify false start kerfs or secondary chipping, helping reconstruct the weapon type and number of strikes. Moreover, differentiation from postmortem changes (e.g., excavation tool marks) is critical, requiring comparison to experimental reference collections. Micro-CT and high-resolution imaging are allowing 3D visualization of kerf wall morphology,

distinguishing blade angle, motion direction, and kerf width even in burned or fragmented bone. Studies also emphasize that sharp force patterns can be modified but not omitted by thermal alteration, meaning that diagnostic striations may even persist after exposure to high temperatures [7,13,21,38].

Ballistic Trauma in Diaphyseal Shafts

Ballistic trauma presents a highly diagnostic feature due to its high-energy interaction between the projectile force and cortical bone. Entry wounds are often rounded or oval-shaped, with circumferential microfracturing and internal grinding, while exit wounds are more larger, irregular in shape, and is characterized by external bevelling. Keyhole fractures occur when projectiles strike directly, combining blunt and penetrating mechanisms. The radiating fractures may extend along the diaphysis, providing an information on the projectile trajectory and velocity. Distinguishing true ballistic trauma from postmortem cracking requires recognition of secondary bone spall and associated lead or copper particles detected via SEM–EDS analysis [6,11].

The literature has reported that comminuted stellate patterns are the predominant fracture morphology in long bones that are subjected to direct gunshot, with traits such as wing pieces, ring defects, and concentric fractures frequently detected in both femur and humerus. emphasizing that fracture characteristics are influenced by the bone properties, mainly cortical thickness and shaft diameter. Consistent features such as plug-and-spall bone fragments, radiating cracks, and concentric heaving fractures demonstrate that "false butterfly fractures" may arise in tubular bones under ballistic loading, mimicking patterns of blunt-force while in reality it reflects the radiating fractures generated at the impact site [43,44].

Hence, the concept of "sequence of trauma" determining which fractures occurred first is of a high significance in where cases of torture or multi-phase violence occured. Cross-cutting fracture lines and overlap analysis provides further evidence for the chronological reconstruction [9,37].

Applications from The Literature

The evidentiary value of diaphyseal shafts in forensic analysis is well documented. A tibial spiral fracture that was initially interpreted as an accidental fall was later reclassified as an inflicted twisting injury based on its fracture propagation, underscoring the diagnostic detail of perimortem trauma. In another event, femoral diaphyseal gunshot fractures were central to reconstructing the firearm type and shooting distance, findings that were subsequently confirmed by the analysis of ballistic residue. Likewise, sharp force kerfs were identified on humeral shafts and revealed evidence of postmortem dismemberment rather than a perimortem assault, a distinction that has critically altered judicial conclusions. These examples demonstrate how diaphyseal trauma analysis provides an interpretive value that extends beyond simple classification, directly informing forensic reconstructions and legal outcomes [5,6,13].

Comparative Value of Tibia vs. Femur in Forensic Practice

Among the long bones of the human skeleton, the femur and tibia hold superb forensic value. Both show high resistance to taphonomic forces and are frequently recovered in archaeological and forensic contexts. Yet their comparative value differs depending on depositional environment, preservation of biomolecules, and evidentiary potential for application. This section outlines recent evidence on the relative forensic advantages of femoral and tibial diaphyses, considering preservation rates, DNA and isotope yields, and their application in forensic investigations [10,12].

Preservation in Different Depositional Environments

The femur, being the largest and most robust bone, and its dense cortical shaft tend to preserve exceptionally well. Its thick cortical bone shields trabecular tissue and marrow cavity from microbial invasion. Even in acidic soils, femora often retain cortical structure longer than ribs or vertebrae. Archaeological reports demonstrate intact femoral shafts in burials several centuries old. femoral powder consistently produce high quality DNA samples with both organic and silica-bead methods, reinforcing femoral priority. Even more, in a routine forensic practice, the femoral midshafts are still prioritized for STR typing, but tibial samples still provide s robust supplementary mtDNA and are especially valuable when femora are absent or too degraded. A case study compared femur and clavicle yields and confirmed. Still, forensic casework demonstrates tibia can sometimes outperform femur bone in certain mitochondrial DNA assays due to superior initial cellular density [5,6,45,46].

Isotope and Trace Element Analysis

Bone turnover variation strongly influences its isotopic and elemental evidence. The femoral cortex remodels relatively slower, meaning its isotope profiles archive more long-term diet, migration histories, and population-level signatures such as strontium, oxygen, and lead. Contrary to the tibial cortex that remodels at a more rapidly rate, capturing more recent life history. Forensic isotope studies highlight this distinction: famine-related dietary shifts were detectable in tibiae but absent in femora, underlining the tibia's value for reconstructing the last years of life [11,12].

More recent biochemical studies have further emphasized these differences. Tibial remodeling makes it more responsive to toxins, toxicology, and trace element accumulation, particularly for substances build up like lead, fluoride, and heavy metals absorbed during later life. Even so, femoral cortical samples yet remains superior for long-term stable isotope signatures, offering more insights into the diet and mobility habits over decades of life [17,47-49].

Taken this all together, the femur provides a long-term archive of life history, while the tibia serves as a short-term recorder, and using both in combination offers a more complete reconstruction of the individual's biography in forensic and archaeological investigations. (**Figure1**)

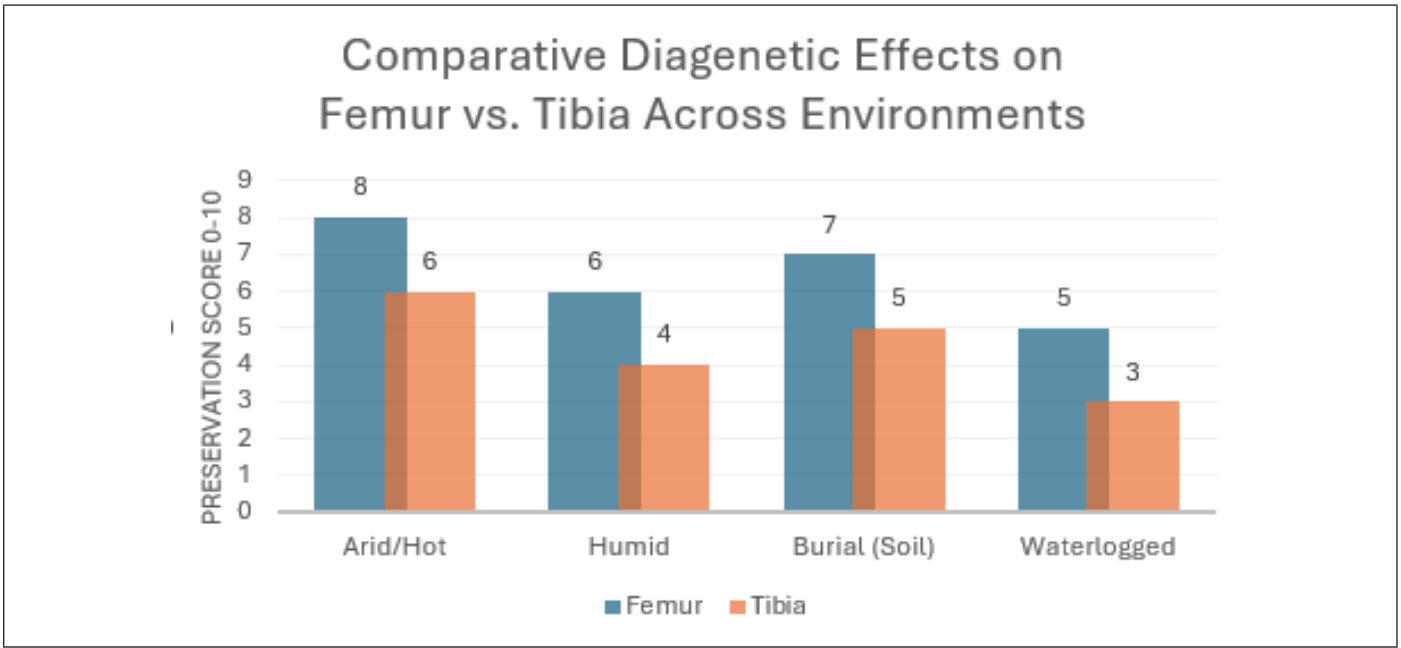

Figure 1: Comparative Preservation of Femoral and Tibial Diaphyses under Different Environments.

Applications from the Literature

A. *Mass Disaster*

In the events of Mass causality events, intact femora are prioritized to use in DNA-based identification, given their characteristics mentioned earlier in dense cortex and reliable nuclear DNA recovery. Also, reported environmental diagenesis plays a decisive role in cases where there was an unstable groundwater or acidic soils that accelerated tibial degradation, femoral shafts resisted longer, thus enhancing their value in disaster victim identification (DVI) [10,50,51].

B. *War Graves*

An archaeological study on a WWII mass grave have further demonstrated DNA typing success varied by skeletal element: femur and tibia bones provided partial STR profiles when environmental conditions permitted. Notably, femora recovered from older graves mainly contained low nuclear DNA, while tibia and other cancellous bones occasionally retained residual collagen and soft-tissue portions that further improved the DNA yields, highlighting the importance of a multi element sampling approaches in commingled or degraded contexts [3,51].

C. *Clinical Forensics*

In a modern hospital-based forensic practice, tibial samples are usually favored for toxicology (fluoride, lead, heavy metals), which clearly reflects their faster remodeling and exposure to recent life metabolic uptake. However, femoral midshaft samples still serves as the gold standard for DNA extraction due to their dense and slow-remodeling cortex [14].

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