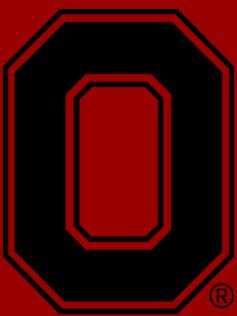


Mapping Spatial Patterns in Cortical Bone Histology from the Femoral Midshaft using Geographic Information Systems Software

Timothy P. Gocha M.Sc., Amanda M. Agnew Ph.D.

The Ohio State University: Department of Anthropology, Division of Anatomy



INTRODUCTION

- When constructing a biological profile for unknown human skeletal remains, forensic anthropologists must estimate the age at death of the decedent. As a complement to traditional macroscopic methods, or when the necessary macroscopic elements are absent or damaged, age estimation can be conducted through a histological examination of remodeling events in cortical bone.
- During the last half century the femoral midshaft has been the most commonly employed skeletal site for obtaining histological age estimates; however, different methods employ various sampling locations that differ in size, number, and location for the collection of histological data across the cortex.
- To effectively determine which area(s) of the femoral cortex are most useful for estimating age at death a thorough understanding of histological remodeling from across the entire cortex is necessary.
- Recently the utility of GIS software for the study of human bone microstructure has been demonstrated (Rose *et al.* 2012). Building upon that, this study examines patterns in the spatial distribution of all histological remodeling events across the entirety of the femoral midshaft.



Figure 1. Composite image of femoral cross-section viewed in polarized light. 2 mm



Figure 2. GIS map of polygons representing bone structures. Light gray = cortical area, dark gray = medullary area, white = trabeculae, black = resorption spaces.

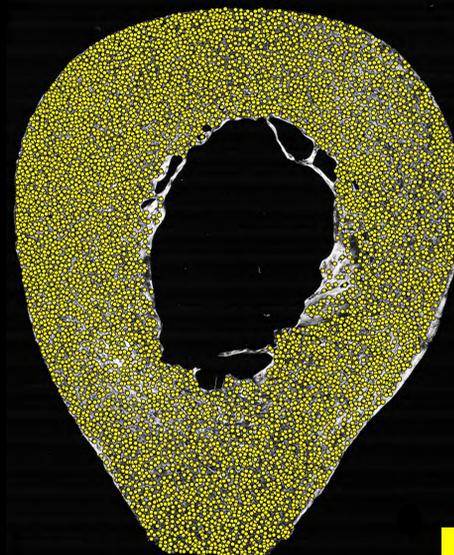


Figure 3. GIS map of remodeling events. Each yellow dot indicates either an intact or fragmentary osteon.

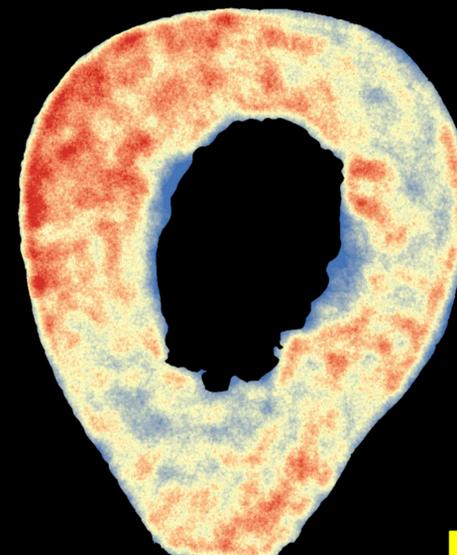


Figure 4. GIS map of remodeling event density. Red indicates areas of highest remodeling density, blue indicates lowest remodeling density.



Figure 5. GIS map of cortical sampling methods. Lines indicate subdivision of the cortex into periosteal, middle, and endosteal thirds, as well as APML and offset quadrants.

MATERIALS AND METHODS

- Thirty complete cross-sections from the femoral midshaft of modern cadaveric donors were used for this study: 15 males and 15 females, ranging in age from 21-97 years (mean = 58.9; SD = 22.1 years). Age distribution was similar for both sexes (Mann-Whitney *U* test, $p=0.486$)
- Each cross-section was photographed under polarized light and micrographs were compiled into seamless cross-sectional images (Figure 1). Images were imported into ArcGIS v10.1 (ESRI®), where polygon features were created to overlay the cortical, medullary, trabecular, and resorption areas (Figure 2), and point features were created to mark all remodeling events (i.e. intact and fragmentary osteons, Figure 3).
- A total of 230, 870 remodeling events were manually notated from all 30 samples, and density maps were generated for each sample to visualize the clustering of remodeling events across the cortex (Figure 4). To assess remodeling across the depth of the cortex, cortical area for each sample was digitally divided into periosteal, middle, and endosteal thirds, as well as offset anterolateral, anteromedial, posteromedial, and posterolateral quadrants. APML and Offset cortical quadrants were also overlaid with cortical third divisions such that the periosteal, middle, and endosteal third of each quadrant could be analyzed (Figure 5).
- The total number of remodeling events contained within the area of each region of interest was used to calculate its osteon population density (OPD; remodeling events/mm²). All OPD values for each cortical sampling method were subjected to stepwise linear regression to determine which area(s) of the femoral cortex are most useful for estimating age at death.

RESULTS AND DISCUSSION

- Density maps of remodeling events revealed that remodeling events are not evenly distributed throughout the femoral cortex. A striking qualitative pattern emerged where the highest density of remodeling events was nearly always in the anterolateral region of the cortex (Figure 4), in males and females and across the age spectrum.
- All stepwise linear regression models produced indicate a robust relationship between OPD and its ability to predict age at death (Table 1). When the total bone OPD is considered the model has an adjusted R^2 of 0.815 ($p = 0.000$, S.E.E. 9.413). However, when different regions of the cortex are considered rather than the entirety of the cortex, the ability to predict age at death from OPD is even stronger.
- Examination of periosteal, middle, and endosteal cortical thirds reveals that OPD in the periosteal third alone is the best predictor of age at death (adjusted $R^2 = 0.815$, $p = 0.000$, S.E.E. 8.313). This is encouraging with regards to previous methods for histological aging from the femur, as it is in this region that most methods require sampling (e.g. Kerley 1965, Ahlqvist & Damsten 1969, Drusini 1987, Ericksen 1991).
- When OPD is considered circumferentially around the cortex by anterior, posterior, medial, and lateral quadrants, the anterior quadrant alone is the best predictor of age (adjusted $R^2 = 0.836$, $p = 0.000$, S.E.E. 8.882), while for offset quadrants the anterolateral quadrant alone is the best predictor of age (adjusted $R^2 = 0.816$, $p = 0.000$, S.E.E. 9.391). However, when OPD from these regions is considered across the depth of the cortex, specifically a combination of the periosteal and middle thirds of the anterior and lateral quadrants, we see the best results for estimating age at death (adjusted $R^2 = 0.905$, $p = 0.000$, S.E.E. 6.747).

Table 1. Stepwise linear regression results for estimating age from osteon population density from various cortical sampling methods. Red indicates regions of the cortex included in the model which yielded the highest adjusted R^2 value. Standard Error of the Estimate is in years.

Cortical Sampling Method	Cortical Regions included in model	Adjusted R^2	p -value	S.E.E.
Whole Bone	Entire Cortex	0.819	0.000	9.40
Periosteal, Middle, & Endosteal Thirds	Periosteal Third	0.857	0.000	8.36
APML Quadrants	Anterior Quadrant	0.838	0.000	8.90
Offset Quadrants	Anterolateral Quadrant	0.814	0.000	9.55
APML Quadrants by Thirds	Anterior Periosteal Lateral Periosteal Lateral Middle Anterior Middle	0.907	0.000	6.73
Offset Quadrants by Thirds	Anterolateral Periosteal Anteromedial Periosteal	0.868	0.000	8.04

CONCLUSIONS

- Overall the results of this study suggest that sampling locations/regions of interest for quantification of remodeling events as they relate to age at death should not be selected at random, or simply by convention. Rather, areas of highest remodeling density, most notably from the anterolateral cortex of the femoral midshaft may explain more than 90% of the variation in estimating age at death. Furthermore, while sampling along the endosteal border appears to be unnecessary, one should look deeper into the cortex than the periosteal third alone.
- This study also demonstrates the overall utility and potential of using GIS software for mapping spatial patterns of histological remodeling events in human cortical bone. Continued research is underway to further subdivide the cortex to examine if smaller regions of interest yield even greater ability to estimate age at death. Applications of this approach to skeletal histology will also allow the determination of when/if different cortical regions become saturated with remodeling events and reach the OPD asymptote.
- While OPD alone demonstrates a robust relationship with age at death, it is acknowledged that other histomorphological variables, such as osteon circularity and area also are strongly related to age at death (Goliath 2010). Further research will examine the possible combination of these variables with OPD from well-informed sampling locations as determined by this research.

REFERENCES CITED

Ahlqvist J, Damsten O. 1969. A modification of Kerley's method for the microscopic determination of age in human bone. *J Forensic Sci* 14:205-212. Drusini A. 1987. Refinements of two methods for the histomorphometric determination of age in human bone. *Z Morphol Anthropol* 77:167-176. Ericksen MF. 1991. Histologic estimation of age at death using the anterior cortex of the femur. *Am J Phys Anthropol* 84: 171-179. Goliath JR. 2010. Variation in Osteon Circularity and Its Impact on Estimating Age at Death. Unpublished M.A. thesis. The Ohio State University. Kerley ER. 1965. The microscopic determination of age in human bone. *Am J Phys Anthropol* 23: 149-164. Rose DC, Agnew AM, Gocha TP, Stout SD, Field JS. 2012. The use of Geographic Information Systems Software for the Spatial Analysis of Bone Microstructure. *Am J Phys Anthropol* 148: 648-654.

ACKNOWLEDGEMENTS

Thanks to all of the donors who shared their generous gifts. Thanks to Drs. Sam Stout, Paul Scullin, and Mark Hubbe for constructive comments on this project. Thanks also to everyone in the Skeletal Biology Research and Injury Biomechanics Research Labs at Ohio State.

