

## H91 The Evaluation of Bone Area as a Histomorphometric Variable for Estimating Age at Death

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The goal of this presentation is to determine if measuring bone area rather than the traditionally used cortical area has the potential to improve histological age estimation methods for the sixth rib.

This presentation will impact the forensic science community by exploring improvements in sixth rib histomorphometry. Measurement techniques used in histological age estimation have previously excluded trabecular bone area. This preliminary study is designed to investigate the relationship of bone area (cortical area + trabecular area) to individuals of known age.

Age-related bone loss has been extensively documented in the medical literature. The fragility of the human skeleton increases with age owing to changes in bone quality, bone remodeling properties, and microarchitecture. While age-related changes in cortical and trabecular

bone microarchitecture have been reported in the anthropological literature, the evaluation of trabecular bone area has not been included in histological age-estimation methods.

With advancing age cortical bone is architecturally modified through the process of trabecularization. This conversion increases the endosteal area and displays a network of interconnecting trabecular struts that are not included in the measurement of cortical bone area. The traditional measurement of cortical area requires the observer to subjectively determine where cortical bone ends and cancellous (trabecular) bone begins, producing measurement variability between observers. By including trabecular bone in area measurements, this subjectivity is removed. Furthermore, it is believed that incorporating both cortical and trabecular bone in area measurements to produce a bone area variable will provide more information for histological analysis of age-related bone loss.

It has been demonstrated that histological age estimation methods based on osteon counts (OPD) are limited when applied to older individuals. This is attributed to the nature of bone remodeling and the osteon population density (OPD) variable used for histological age estimation. Osteon counts from the rib eventually reach an asymptotic value in individuals of advanced age. Two factors exist that determine the age at which osteon counts will reach an asymptote value: (1) cortical area; and, (2) osteon area (size). Including trabecular bone area in histological measurements may improve future histological age estimation methods for the ribs, especially in older individuals where the OPD value becomes less reliable. The objectives of this study are: (1) to remove the subjectivity caused by measuring cortical area; and, (2) to determine if bone area has a stronger correlation with age than cortical area alone.

The sample consisted of midshaft sixth rib cross-sections from 31 known age individuals, ranging from 16-87 years of age (mean age 46.4

+/- 3.2 SD years). These included 26 men and 5 women. All data was collected from a pre-existing collection of sixth rib bone samples at the Office of Chief Medical Examiner in New York City. The following histomorphometric variables were collected: (1) total subperiosteal area (TA); (2) cortical area (CA); (3) bone area (BA); (4) relative bone area (rBA); (5) relative cortical area (rCA); and, (6) endosteal area (EA).

The rib cross-sections were prepared following standard protocols for histological sample preparation. The cross-sectional area of two rib samples per individual was evaluated. Only thin sections with suffice microstructural preservation and an intact cortex were utilized. Thin sections were photographed using a transmitted light microscope and a mounted digital camera attachment (x40 magnification). A series of sectional photographs were merged using digital imaging software. Cortical and trabecular bone areas were measured using a modular imaging software and a digitizing tablet.

Statistical analyses were completed using statistical software. A one-way analysis of variance (ANOVA) with age as the predictor variable was fit for rBA, rCA, CA, BA, EA, and TA to determine the strongest correlation with known age. When significant effects were detected, paired t-tests were utilized to identify significant differences between age categories. Relationships between known age and the respective variables were evaluated by linear regression analysis.

Results from this preliminary research indicate that cross-sectional area measurements were found to decrease with advancing age. Although the sample size is small, individuals greater than 40 years of age were found to have significantly smaller mean BA than those who were less than 40 years of age. Overall, a negative correlation was observed between known age and sixth rib BA (R=-0.34), and CA (R=- 0.35). rBA and rCA displayed highly significant negative correlations with increasing age. The correlation strength was slightly higher between known age and rBA (R=-0.65) compared to known age and rCA (R=-0.59). Known age and TA

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displayed a positive correlation (R=.0.25). With one-way ANOVA, both rBA and rCA show significant difference among age groups at P=.039 and P=.025, respectively.

Previous studies have demonstrated that sixth rib cortical bone area significantly decreases with age and therefore, the variable has been applied to more recent age estimation equations. This overall age-related decrease in bone mass can be attributed to trabecularization of the cortical bone which results in endosteal expansion. The results of this study suggest that evaluating bone area may improve the accuracy of histological methods. Including rBA in future studies will provide a more relevant measure of bone area as it is independent of bone size. Finally, subjectivity and interobserver error may be reduced if the BA and rBA variables are included in future methods.

Histology, Histomorphometry, Bone Area