

**Recognizing and Resolving Inconsistencies and Inaccuracies in Determining Osteon Circularity: Can Methods be Standardized?**

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Osteon circularity (On.Cr) might help in distinguishing species, interpreting load history, and estimating age. When studying On.Cr using archived backscattered electron(BSE) and circularly polarized images of various primate/non-primate bones, we recognized that inaccuracies can occur when there are seemingly inconsequential differences in scale of actual/physical images used in osteon tracing (although all taken at 50-62.5x). For example, errors might occur if non-digitized Polaroid images (500microns=26mm) from 1980s-1990s are compared to modern digitized images in larger format (500microns=53mm). How closely must image 'sizes' be to achieve <2% error(arbitrary cutoff)? Additionally, do manual and semi-automated computer-mouse-based tracing provide similar data, and how do these compare with tracing using pen on plastic transparencies vs. a digitizing tablet/stylus? Ten osteons (50x BSE images; human femur, 60years) were manually pen-traced on plastic sheets and scanned for analysis(ImageJ). Quantifying On.Cr this way is highly accurate when using ImageJ<sup>1</sup>(Skedros 2000, J.Bone&Mineral Research). The images were reduced ~25% (500micron=39mm) and ~50% (500micron=26mm) to resemble our archived/unadjusted images, and the osteons were manually re-traced/re-scanned. Differences ("errors" vs. 100% size) included: (1)25% reduction 3±2%(max6%); (2)50% reduction 2±2%(max7%). Using the "Quick Select" tool in Photoshop (instead of pen tracing/scanning) resulted in even greater error (average 14±9%). Additional analysis showed <2% error when reduction is <15%. Because achieving accuracy when quantifying scanned pen-tracings in ImageJ can be challenging (as shown when using computer-generated perfect circles), additional studies will be conducted to determine if a digitizing pad and stylus, as used by Crescimanno and Stout (2012, J. Forensic Science), increases accuracy and efficiency.