

# MINERAL CONTENT AND MECHANICAL PROPERTIES OF CEMENT LINES IN HUMAN OSTEOCAL BONE

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## Introduction

Bone toughness is a complex property depending on the arrangement of the basic constituents (mineral and collagen), higher order structural motifs, and their numerous interfaces. Among these, cement lines (CLs) are thin layers separating secondary osteons from each other or from the interstitial bone. Based on the experimental evidence that microcracks are more frequently observed outside osteons than inside, CLs are believed to protect the osteons by deflecting cracks. However, CLs are still a poorly understood structure. Specifically, their composition, mineral properties, and biomechanical behavior are understudied. Here, we characterize CL mineral content and mechanical properties by combining high resolution backscattered electron imaging (hrBEI), including quantitative analysis (qBEI), with nanoindentation (nIND) on the same locations.

## Methods

Cortical bone samples from two human femurs (male individuals, 40 and 81 years old) were harvested. Samples surface was smoothed with standard polishing using sandpaper, including a final step with ion polishing. A combination of qBEI (~0.5  $\mu\text{m}$  pixel size) and hrBEI (~0.1  $\mu\text{m}$  pixel size) was performed on selected osteons ( $n_{\text{tot}} = 62$ ) to quantify mineral content and to highlight nanostructural features. Reduced modulus ( $E_r$ ) and hardness were measured across the CL in selected regions using nIND (Berkovich tip, 1–2.5  $\mu\text{m}$  lateral spacing, 500–1000  $\mu\text{N}$  load, 5500 total indents). For the evaluation, CLs were manually segmented from the corresponding osteons and the bone mineral density distributions (BMDDs) of osteons and CLs were computed (Fig. 1). The correlation between mineral content and  $E_r$  was obtained by superimposing nIND and qBEI measurements.

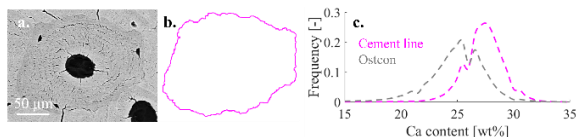


Fig. 1: (a) qBEI of one representative osteon and (b) corresponding CL mask. (c) Global BMDDs for all the analyzed osteons and CLs ( $n_{\text{tot}} = 62$ ).

## Results

CLs are tiny interphases with an average thickness of about 1–3  $\mu\text{m}$  and a higher mineral content than the corresponding osteons (Fig. 1c). A spatial mapping of  $E_r$  across two adjacent osteons exhibits a periodic

alteration of stiffer and softer regions corresponding to bone lamellae, with a distinctive peak characterizing the CL (Fig. 2a and c). This increase in stiffness is spatially correlated with the mineral content (Fig. 2b and d). hrBEI allows to discriminate different osteonal features by their texture and the average  $E_r$  could be computed inside the CL ( $29.34 \pm 1.28$  GPa), the stiffer lamellae ( $23.09 \pm 1.36$  GPa) and the more compliant lamellae ( $18.5 \pm 0.69$  GPa).

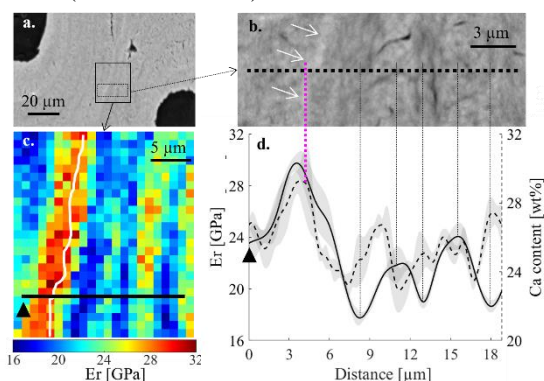


Fig. 2: (a) qBEI overview and (b) hrBEI inset with the CL highlighted by the arrows. (c) Map of  $E_r$  with the CL highlighted by the white line. (d) Spatial evolution of  $E_r$  (solid line) and mineral content (dashed line), measured along the dotted line in (b).

## Discussion

We highlight a contrast in mineral content and elastic properties between the CL and corresponding osteon, which is higher than spatial variations of those properties within the osteon. This stiffness contrast may allow CLs to act as effective interfaces hampering crack propagation. Recent findings that CLs have reduced nanoscale porosity in comparison to adjacent lamellar bone explain the measured higher mineral content [1]. In contrast to previous literature [2,3], we report that CLs are stiffer than surrounding bone, which is in line with a reduced nanoporosity and higher mineral content. However, mineral properties of the CLs like crystal arrangement, dimensions and composition also contribute to the mechanical behavior, but these are unknown. Ongoing work aims at unraveling additional aspects of the mechanical behavior of CLs as well as the mineral properties using X-ray scattering.

## References

1. Tang et al., J Bone Miner Res, in press, 2023.
2. Montalbano et al., J Mater Res, 26(08):1036–1041, 2011.
3. Zhou et al., EML, 41:101031, 2020.

