

## **Sub-micron resolution diffraction and fluorescence tomography reveals bone microstructure**

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The structures of many materials including biominerals are highly complex. Understanding their structure is challenging and calls for methods with sensitivity across several length scales in 3D. Multimodal X-ray tomographies are such techniques [1]. Here, we apply diffraction scattering computed tomography (DSCT) to reveal the crystalline properties and fluorescence tomography (FT) to probe element distributions in bone using a 400 nm X-ray beam.

Bone is a complex hierarchical material with essential structural features ranging from the nanoscale to the macroscopic. The link between bone structure and function, and especially the contribution of the nanoscale structural elements to the macroscopic properties remains poorly understood. The osteon is an essential building block in human long bones and contributes to highly anisotropic mechanical properties. In this motif, mineralized collagen fibrils are arranged in a twisted plywood structure surrounding a Haversian canal. While it has been shown that the indentation modulus varies periodically with the osteon lamellae and that this is positively correlated with the mineral content [2], it remains unclear how the mineral phase's properties varies across the osteon. The same holds for oligo elements such as Sr. To unravel the structure of osteons, we combine DSCT and FT with sub-micron resolution. This experiment combines the capabilities of diffraction and fluorescence with those of computed tomography to allow for reconstruction of a diffractogram and a fluorescence spectrum in each volume element within the sample [1]. The resulting >1.5 million diffractograms were Rietveld refined using MultiRef [3] to obtain typical crystallographic parameters, including unit cell parameters, profile parameters etc. This revealed distinct variations in mineral properties and element distributions with distance from the osteon center

[1] Birkbak *et al.* (2015) *Nanoscale* **7**, 18402-18410. [2] Gupta *et al.* (2006) *J. Mater. Res.* **21**, 1913-1921. [3] Frölich & Birkedal (2015) *J. Appl. Cryst.* **48**, 2019-2025.