	Experiment title: Characterization of bone tissue at the micron level by 3D SR microtomography and its relation to mechanical properties	Experiment number: LS/1833
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Preliminary report

Introduction The objective of this experiment was to acquire the 3D geometry of the vascular and lacunar porosity of a set of bone specimens that were previously tested in our laboratory using a microtensile loading system.

Materials and methods A total of 3 bovine and 3 human microspecimens were successfully scanned with the 3D SR microtomography system at 12KeV using the multilayer device and the 2000 Frelon camera. The axial resolution was 0.95 micron per pixel. The gauge length of the specimens being 4mm, five reconstructions were needed to obtain the complete 3D attenuation map of each specimen.



Fig. 1 Human bone specimens in the microtensile testing system.

The resulting images were compressed to 1 byte per pixel and a threshold was applied to extract both the vascular and lacunar porosities of the bone tissue. Quantitative stereology of the porosities is currently under way and the average mineralization of each specimen will be estimated using the graylevel calibrated with an aluminium sample of kwown absorption spectrum.

Results First estimations of the porosities provided 2.9 and 1.7% for the vascular and lacunar compartments respectively. The orientation of the lacunae were found to follow the concentric lamellae of the mineralized extracellular matrix. The fine canalicula networking the lacunae remain invisible at this resolution.



Fig. 2 Reconstructed transverse sections of homogeneous bovine bone specimens.

The longitudinal modulus of the first bovine microspecimen measured by nanoindentation and assuming a Poisson ratio of 0.33 was 28.2 ± 2.9 GPa and has to be confronted with the macroscopic longitudinal elastic modulus of 25.7 GPa measured on our microtensile testing system. The account of the porosity obtained with 3D SR μ CT to explain the latter difference is currently under way.



Fig. 3 Lateral and axial views of the vascular (thick Haversian channel) and lacunar (ellipsoidal cavities containing the osteocytes) porosities of human bone tissue.

Discussion The 3D SR microtomography system at ESRF proved to be an efficient method to assess the vascular and lacunar porosities of human bone tissue. Unfortunately, the intensity of the X-ray beam and the required exposure time were found to destroy numerous collagen cross-links of the extracellular matrix and compromise the mechanical properties of the tissue in the exposed part of the specimens. This finding may exclude the possibility of quantifying morphology and mechanical properties in the same human BSUs using 3D SR microtomography. A future study will be needed to establish the X-ray dosis-mechanical degradation relationship in the manufactured bone specimens.