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**IT-9-P-2328 ACOM-TEM analysis of mineral particles ultrastructural organization in bone tissue**

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Bone tissue has a complex hierarchical architecture that is self-assembled in order to perform diverse mechanical, biological and chemical functions. At the nanoscale it can be viewed as a composite material made up of two principal components: collagen fibrils of ~ 100 nm in diameter and platelet-shaped calcium phosphate mineral crystals of the 5 x 50 x 100 nm dimensions. The size, shape, organization, orientation and internal structure of mineral crystals has been a matter of disputes since bone sections were first studied by electron microscopy in the 1950's [1].
Transmission electron microscopy (TEM) shed new light on this problem by allowing the direct visualization of bone structure. However, a lot of difficulties were faced related to image interpretation and to the choice of samples preparation technique. In collaboration with a medical team, we are now able to produce bone sections as thin as 70 nm. We are also currently exploring new bone sample preparation methods than ultramicrotomy as, e.g. tripod polishing and ion milling.
The novel use of the Automated Crystal Orientation Mapping with a TEM method (ACOM-TEM, also known as ASTAR™ tool from NanoMEGAS) [2] to study the mineral particles ultrastructural organization in bone tissue with the spatial resolution of 20 nm is reported. The ACOM-TEM method operated in scanning mode and relied on the comparison between the high quality electron diffraction patterns collected at every scan position and the simulated patterns calculated for a given crystal in all possible orientations. This method, therefore, allows crystallographic indexing, high-resolution nanocrystal orientation (~ 1°) and crystal phase mapping.
The mineral particles in bone orientation 2-D mapping was, for the first time, analyzed and the presence of disorder, discontinuity and crystallinity degree variations is discussed. Current results are part of larger project aiming to understand the nanostructural characteristics of bone tissue and to identify key structural markers of pathological human bone [3], providing possible development of new diagnostic and pharmaceutical tools.

References:
1. Robinson R. A., Watson M. L. (1952). Collagen-crystal relationships in bone as seen in the electron microscope. Anatom Rec 114: 383–409.
2. Portillo J., Rauch E.F., Nicolopoulos S., Gemmi M., Bultreys D. (2010). Precession Electron Diffraction assisted Orientation Mapping in the Transmission Electron Microscope. Mater Sci Forum Vol. 644 pp 1-7.
3. Gourrier A., Li C., Siegel S., Paris O., Roschger P., Klaushofer K. and Fratzl P. (2010). Scanning small-angle X-ray scattering analysis of the size and organization of the mineral nanoparticles in fluorotic bone using a stack of cards model. J Appl Crystallogr 43, 1385-1392.

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