Characterization of calcifications in human kidney by spectromicroscopy at the nanometer scale

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Randall’s plaques are calcium phosphate deposits, at the origin of kidney stones. To date, little is known about the mechanisms involved in their formation. µFTIR (µFourier Transformed InfraRed spectroscopy) on samples from different kidneys indicate the presence of carbo-apatite, whitlockite and their co-existence with amorphous calcium phosphate phases. In the present study, our aim was to localize Randall’s plaques at the early stages of their formation and to characterize their composition and crystallinity as a function of their localization, at the nanometer scale. Small pieces of papilla tip from healthy papillae of human kidneys were chemically fixed and embedded in epoxy resin. Ultrathin sections were analyzed by Transmission Electron Microscopy and Electron Energy-Loss Spectroscopy (EELS). Nano-calciifications were identified within vesicles, in many cases in close contact with collagen bundles (figure 1). These vesicles whose role and nature is to be determine could be the first step toward plaque formation. Selected Area Electron Diffraction (SAED) evidence that microvesicles contained a few nanocrystals whose diffraction pattern is compatible with the presence of crystalline apatite or whitlockite. Basic maps of light elements of biological interest (Ca, P, N, O) confirm that dense deposits are mainly composed of CaP (figure 1). To go further, data were decomposed using the Hyperspy open source software for principal components analysis (PCA). The fine structure of the different EELS characteristic signals allows to investigate the composition of the nano-calciifications in order to try to discriminate between the various phases identified by µFTIR (carbo-apatite, whitlockite, amorphous calcium phosphate).
Figure 1: High angle Annular Dark Field (HAADF) images and EELS analysis revealed the presence of nanocalcifications made of CaP aggregated inside vesicles which "membranes" are nitrogen-rich and contain low amounts of CaP.