

## ***Raman and Nano-analytical Electron Microscopy Characterisation of Human Cardiovascular Tissue Calcification in Disease***

Amanda Y.F. You<sup>1</sup>, Sergio Bertazzo<sup>1</sup>, Jean-Philippe St-Pierre<sup>1</sup>, Joseph Steele<sup>1</sup>,  
Hainan Xie<sup>1</sup>, Martin Hedegaard<sup>4</sup> & Molly M. Stevens<sup>\*1, 2, 3</sup>

<sup>1</sup>Department of Materials, Imperial College London, London SW7 2AZ, UK.

<sup>2</sup>Department of Bioengineering, Imperial College London, London SW7 2AZ, UK.

<sup>3</sup>Institute of Biomedical Engineering, Imperial College London, London SW7 2AZ, UK

<sup>4</sup>Institute of Sensors, University of Southern Denmark, Odense, Denmark.

\**m.stevens@imperial.ac.uk*

Tissue calcification occurs naturally in bone and the deep zone of articular cartilage as well as in a number of soft tissues as part of disease processes. A better understanding of the structures and nano-scale architecture of ectopic calcification, as well as their surrounding extracellular matrix composition in tissues such as aortae during atherosclerosis and other calcific diseases can reveal similarities and differences between biomineralisation mechanisms that occur in different diseases. Healthy and diseased human aortae, valves, and coronary arteries were analysed using Raman spectroscopy and nano-analytical electron microscopy techniques. Scanning electron microscopy with energy-dispersive X-ray spectroscopy (SEM-EDS), transmission electron microscopy (TEM) with EDS, and selected-area electron diffraction (SAED) with a focused ion beam (FIB) showed the presence of spherical calcium phosphate deposits across the different diseased and even healthy tissues. These spherical deposits were a highly crystalline form of calcium phosphate, which differed from bone mineral in both its crystallinity and structure<sup>1, 2</sup>. Raman mapping of the aorta wall tissue supported these results, based on intensity heat maps of the 960 cm<sup>-1</sup> calcium phosphate and the 1080 cm<sup>-1</sup> phosphate bands that showed discreet regions of calcium phosphate deposits along the tunica media of the aorta wall tissue. Besides that, differences in the chemical composition of the surrounding tissue matrix were also analysed. The results raise interesting questions about the calcification process in atherosclerosis and other ectopic calcification related diseases. They indicate that a different calcification mechanism is present compared to bone. However interestingly it appears that different calcific diseases might undergo the same calcification pathway. The characterisation and study of these tissues can thus contribute to a better understanding of pathophysiological tissue calcification processes and can have important implications in the field of cardiovascular disease.

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- [2] E. Gentleman, R. J. Swain, N. D. Evans, S. Boonrungsiman, G. Jell, M. D. Ball, T. A. Shean, M. L. Oyen, A. Porter and M. M. Stevens, *Nat Mater.* **8**, 763-770 (2009).