Protocol for Raman Imaging at ICMCB

Raman imaging measurements were performed on an Invia Reflex spectrometer (Renishaw, GB), equipped with a Leica 2500 microscope. The excitation laser line was the 514nm line of a solid-state Cobolt laser. The configuration spectrometer is a single spectrometer one (one grating, 1800 grooves/mm). The spectral resolution is closed to 2 cm^{-1}. All maps were acquired in the Renishaw Streamline mode (line mapping by use of a cylindrical lens, and synchronization between the motion of the microscope table and the CCD reading). This line mode allows high laser powers (50 to 70 mW) upon keeping reasonable local energy densities. Two types of images were recorded: (i) large-scale ones, with low spatial resolution, as rectangles of some mm side, with a x20 objective and 20µm of separation between adjacent points (ii) detailed zones of some hundreds of µm side, with high spatial resolution, with a x100 objective and 2.6 µm between adjacent points. In both cases the acquisition time for each point is 10 to 15 s, and the total number of spectra in each image is between 50000 and 100000. All data treatments were performed with the Renishaw Wire3.4 software and its Chemometrics module, including PCA (principal component analysis) module.