CRYSTALLINITY DIAGNOSTICS AND 3D-ORIENTATION MEASUREMENTS OF SINGLE NANOCRYSTALS BY NONLINEAR MICROSCOPY

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Ultra-sensitive, super-resolving and non invasive tools are currently developed for studying the organization of molecular assemblies within complex structures down to the nanometric scale in polymers, crystals or confined bio-membranes at room temperature. These structural characterisations are essential for the understanding of local biological or chemical mechanisms. Microscopy imaging can be combined with orientation and molecular structuration studies, relying on polarization properties of optical processes. While the ultimate detection scale of a single molecule has been reached ten years ago using fluorescence, second harmonic generation (SHG) from an isolated nano-crystal containing a few thousands of coherent emitters has been detected recently [1, 2].

Within this context, we have developed at LPQM [3] a high resolution multi-photon excitation microscope sensitive to both molecular orientation and organization, combining two- and three-photon fluorescence (2PF & 3PF) and second (or even third) harmonic generation (SHG & THG). 3D information has been achieved by associating a defocused imaging system [4] to a polarization resolved detection scheme [1, 3]. While this set-up has been applied to probe the local molecular organization in organic monolayers (2PF & SHG) [5] and to elucidate the crystalline nature of protein crystals (3PF), 3D-orientation of isolated nonlinear nanocrystals has been determined for the first time by coherent defocused orientation imaging (SHG).

In addition to know the organization of nano-structures, this method can be dedicated to protein crystallisation process monitoring, in order to understand relations between their structures and their biological functions at molecular scale.