AXIAL SUPER-RESOLUTION IN CONFOCAL MICROSCOPY WITH THE THIRD-HARMONIC-DETECTION MODE

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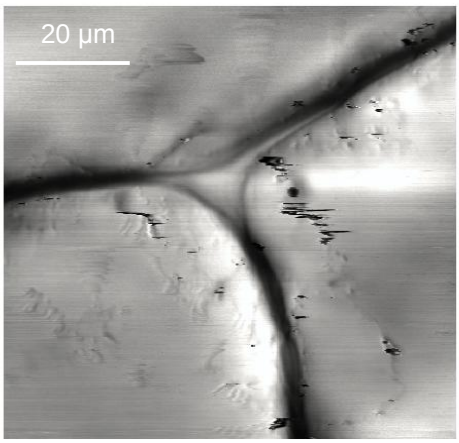
1. DETECTING THE THIRD-HARMONIC WITH SiOx NANOPARTICLES

A second harmonic generation scheme that acts as a virtual pinhole, gating out non-ballistic photons has been recently proposed and demonstrated [1]. To further improve this approach we propose and demonstrate a novel scheme, the third-harmonic detection (THD) mode for increased gating effect and improved axial resolution. Here the THG detector is a silicon-oxide nanoparticle deposited in vacuum by femtosecond laser ablation [2]. Their nanometric dimension bestow both the advantages of pinholed detection and increased axial resolution.

2. IMAGING PROPERTIES OF THE THD MODE

The set-up was a bright-field transmission confocal microscope (illuminator and collector: 60×, water-immersion, 1.2 NA) except for the detection pathway which consisted of a 20× focusing objective to concentrate the transmitted light onto a single nanoparticle. When excited by a femtosecond forsterite laser beam at 1.26 µm, 84 MHz, 50 fs, the THG yield scaled by the expected power of three. This scheme is well adapted to low absorption, low contrast objects since it delivers the cubic analogue of the signal in the classical confocal mode. In the axial direction the aggregate can be considered in the first approximation to behave as a thin layer and provides a sectioning power as $0.64b$, with b the confocal parameter, that is an axial gain 57% over a simple interface [3].



THD transverse cut of an onion peel

3-RESULTS

A 80×80 µm, 420×420 pixels transverse cut of three adjacent cells of an onion epithelium is shown above. Excitation average power was 70 mw. Tight focus on a single nanoparticle was made by a previous computer-controlled scan of the substrate. Superior contrast and details that cannot be seen in the classical mode are evidenced, because of the sharp sectioning.
In conclusion, we have demonstrated a promising imaging scheme for low contrast samples with improved sectioning power capability.

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