3D SUB-DIFFRACTION RESOLUTION IMAGING OF QUANTUM DOT LABELED CELLULAR STRUCTURES AT STANDARD CONFOCAL MICROSCOPES

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KEYWORDS: Quantum dots, sub-diffraction resolution imaging, tri-exciton, confocal microscopy, 2 photon microscopy

ABSTRACT

Quantum dot triexciton imaging (QDTI) utilizes the ability of some CdSe quantum dots (QD) to absorb three photons and the subsequent blue-shifted emission from higher-order excitonic states. By narrowing the point spread function the tri-exciton formation has the potential to increase the optical resolution in the axial and lateral direction by approximately 1.7-fold (1). Due to the relatively long lifetime of the QD excited states, this method requires only standard lasers and can be easily implemented on any conventional confocal microscope (2). We applied QDTI to investigate various cellular structures labeled with QD655 antibodies. By combining with deconvolution we achieve a further improvement in contrast resulting in an about 2-fold resolution improvement (3). Careful selection of fluorophores enables QDTI to be used with color multiplexing for 3D imaging of subcellular structures and multilayered cell clusters with improved axial and lateral resolution. To further extent the potential of QDTI for high resolution imaging deep in biological samples we use NIR light for QD excitation and optimized conditions for the generation of tri-excitons in QDs by the pulsed Ti:Sapph laser. We applied this novel approach to image QD labeled subcellular structures in cells and tissue with improved optical resolution. In summary, this QDTI provides the possibility for 3D sub-diffraction imaging of biological samples labeled with commercially available quantum dots QD655 ranging from subcellular structures to cells clusters and tissue at standard confocal microscopes.

