Fluorescent Nanodiamonds for Specifically Targeted Bioimaging: Application to the Interaction of Transferrin with Transferrin Receptor

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Fluorescent nanodiamonds (FND) as optical probes to image biological systems have attracted considerable attention because of their special optoelectronic properties – bright fluorescence, protracted photostability and a broad spectral range, as well as because of their low cytotoxicity and ease of conjugation with biomolecules after surface functionalization

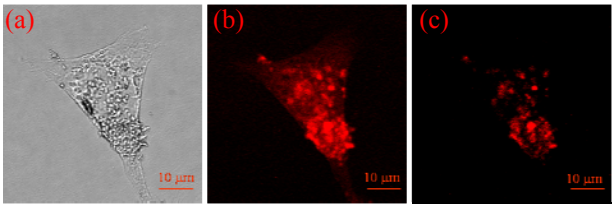

Fig. 1: Images of a fixed HeLa cell after treatment with FND-Tf bioconjugates (10 μg/mL) for 1 h in an incubator; (a) bright field image, (b) confocal scanning image obtained on collecting fluorescence of FND at >550 nm upon excitation at 514.5 nm; (b) confocal scanning image with fluorescence collected at 663–738 nm. The 100-nm diamonds were irradiated with a 2.5-MeV ion beam and thermally annealed to emit bright fluorescence in the region 550–800 nm upon excitation at 514.5 nm; accordingly, the interference of cellular autofluorescence at 520–650 nm was eliminated significantly on detecting fluorescence at >660 nm.

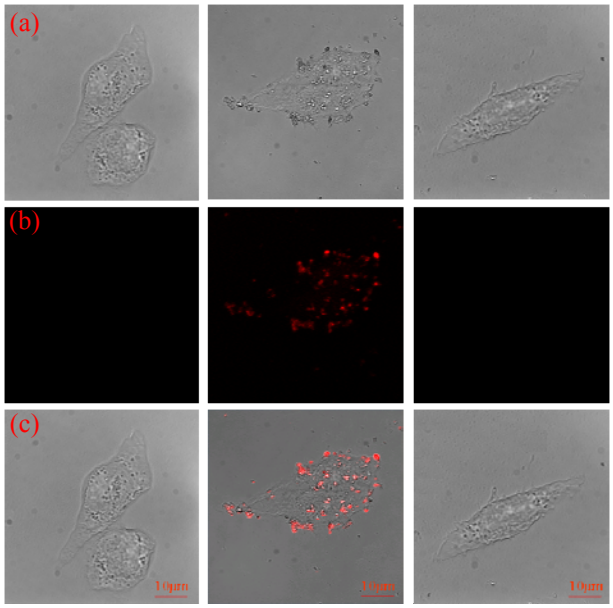

Fig. 2: Comparison of confocal fluorescence images of HeLa cells treated with carboxylated FND and FND-Tf bioconjugates; (a) the left two bright-field images are HeLa cells treated respectively with carboxylated FNDs and FND-Tf bioconjugates, and the right one image is a HeLa cell presatured with free transferrin for 1 h before being treated with FND-Tf bioconjugates; (b) corresponding confocal fluorescence images; (c) overlays of bright and fluorescence images. The receptor-mediated uptake of FND-Tf bioconjugates into HeLa cells was confirmed from confocal fluorescence images.

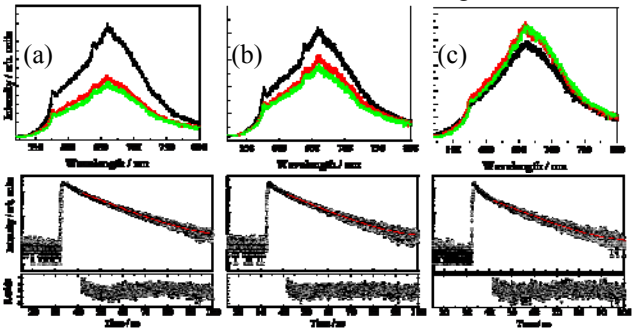

Fig. 3: Comparison of fluorescence spectra and lifetime of (a) carboxylated FND, lifetime is 11.8±0.1 ns (b) FND-Tf bioconjugates, lifetime is 11.6±0.1 ns (c) FND-Tf-TfR complexes on a fixed HeLa cell, lifetime is 12.1±0.1 ns. These results of the nearly identical spectra and lifetime indicate that the surface effects of chemical interactions on the emission center of FND are negligible.

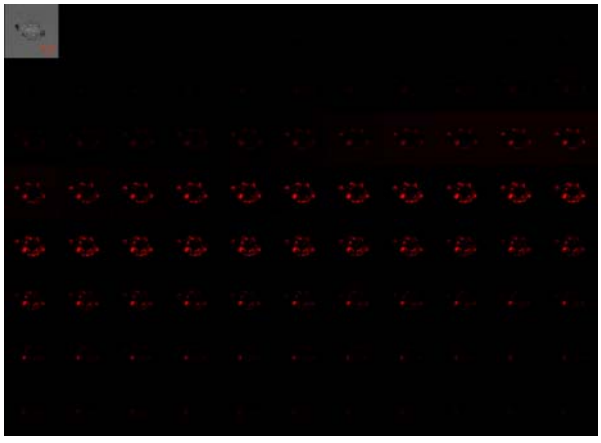

Fig. 4: Confocal fluorescence images of a HeLa cell treated with FND-Tf bioconjugates, recorded with a step 200 nm in the vertical direction. Fluorescence signals were observed within a thickness ~10 μm, indicative of translocation of FND through the cell membrane.