

Molecular Imaging with A Bimodal Fluorescence-Raman Device

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Introduction

It's difficult to deny that live-cell imaging hasn't impacted our perspective on biology. Over the last ten years, there has been a surge in interest in imaging cellular activities at the molecular level. Many innovative approaches are now being used in live cell imaging. Cellular health, on the other hand, is frequently overlooked. For many researchers, all is well if the cell does not go into apoptosis or is blabbed beyond recognition at the end of the experiment. This is completely false. When performing live-cell imaging, numerous aspects must be considered in order to maintain cellular health, including imaging modalities, medium, temperature, humidity, PH, osmolality, and photon dose. Two of the most essential and controllable aspects of live-cell imaging are the wavelength of illuminating light and the total photon dose that the cells are subjected to. The lowest photon dose that yields a metric for the experimental inquiry, rather than the dose that provides cover photo grade photos, should be employed. This is critical to guarantee that the biological processes being studied are in their in vitro condition and have not been switched to a different pathway as a result of environmental stress. The timing of the mitosis is an ideal canary in the gold mine, in that any stress induced from the imaging will result in the increased length of mitosis, thus providing a control model for the current imaging conditions.

Cellular Imaging

Among the most intriguing breakthroughs in cell biology in the last five years has been the implementation of nanoparticles. Because the number of studies documenting the use of nanoparticles is continuously rising, this review will focus solely on applications of nanoparticles on entire cells, either fixed or living, rather than the numerous strictly in vitro or in vivo applications. The review's focus on cells stems from the fact that knowing nanoparticle-cell interactions is the first step toward a

mechanistic understanding of the relationship between animals and nanomaterial's. As a result, cellular investigations serve as a prelude to nanoparticle utilization in in vivo medicinal or imaging applications.

Nanoparticles applied to cells and employed for imaging subcellular components are of special relevance in this paper. Although cytotoxicity and the effects of nanoparticle loading on cells are unimportant in fixed cells, nanoparticle biocompatibility and cellular absorption processes are crucial in live cell research. Nanoparticle toxicity/biocompatibility is most often determined by their concentration, according to studies of their impact on cellular proliferation and viability. Different cellular uptake methods are used by cells depending on the kind of cell treated, the size, and the surface charge of the nanoparticle conjugate (nanoconjugate), with the most common being clathrin-dependent mechanisms, macro pinocytosis, and phagocytosis.

The optically fluorescent semiconductor quantum dots and noble metal nanoparticles with size and shape-dependent optical characteristics are the subject of this review. Furthermore, a distinct type of semiconductor material—TiO₂—is given special attention since it is easily functionalized by both optically fluorescent agents and molecules for subcellular targeting. Optical microscopy and electron microscopy are still two of the most potent tools for detecting nanoparticles in cells. However, complementing novel imaging techniques such as four-photon microscopy, near-infrared surface enhanced Raman scattering, X-ray fluorescence micro- and nano-probe imaging, and coherent X-ray diffraction imaging will greatly improve imaging work with nanoparticles in cells. Some of the future developments with these techniques are expected to allow for 3D imaging with resolution as good as 5 nm³ voxel (coherent X-ray diffraction imaging), permitting imaging of whole frozen cells with the nanoparticles distributed at specific destinations in the cellular interior.

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