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细胞生物学中的激光 成像和操控

Laser Imaging and Manipulation in Cell Biology

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Edited by Francesco S. Pavone

Laser Imaging and Manipulation in Cell Biology

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Laser Imaging and Manipulation in Cell Biology



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Two-photon imaging of hippocampal pyramidal neurons (YFP labelled).

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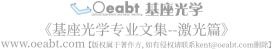
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Introduction

Francesco S. Pavone

Since the development of nonlinear laser imaging tools, such as the two-photon technique [1] for example, many technological advancements have been made in the field of microscopy and, more generally, imaging. It took more than 60 years to move from the discovery of the two-photon interaction [2] to its exploitation in microscopy. Since the 1990s, an exponential growth of publications in the field of microscopy (Figure 1) has led to the introduction of the two-photon technique in the laboratories of many researchers worldwide.

1

Since the first interaction schemes, where all photons were accumulated and collected on the detector after the laser irradiation (integration mode), other kinds of investigation modes have been developed, based, for example, on the lifetime response of the fluorescent molecule (fluorescent lifetime microscopy), on the spectral behavior of fluorescence emission (multispectral two-photon emission), or on the ability of the illuminated molecule to double the frequency of the coherent excitation due to its nonlinear susceptibility (second- and third-harmonic generation microscopy).

Further developments in microscopy have led to other nonlinear interaction schemes such as coherent anti-Stoke Raman spectroscopy (CARS) [3] (Figure 2) and resonant Raman scattering [4].

The nonlinear characteristic of the interaction of pulsed light with a molecule has also led to applications that are useful in increasing the resolution below the diffraction limited barrier [5].

All these imaging tools, together with well-developed photon based technology, such as confocal microscopy, have enlarged the field of applications in biological imaging of molecules, cells, and tissues.

Consequently, the new frontier of cell biology imaging has moved from a fixed cell to a living cell with the advent of the laser and more sensitive wide-field fluorescent microscopes. The advent of confocal microscope has improved the axial resolution, while the application of multiphoton processes has finally permitted the study of cell biology in tissues and, consequently, in living organisms, as well as allowing optical manipulation [6].

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