Detecting Signals at the Single Molecule Level: Pioneering Achievements in Microscopy

Fluorescence lifetime imaging microscopy (FLIM) is an established tool for a variety of applications in biology and biomedical research. However, recent advances have led to such remarkable improvements in its capacity for contrast and sensitivity that researchers can now employ it to detect signals at the single molecule level. FLIM also offers the additional benefit of independence from fluorophore concentration and excitation intensity. Moreover, its unique sensitivity makes it an excellent reporter of conformational changes and of variations in the molecular surroundings of biological molecules.

Most of this improvement and discovery has occurred during the past decade and to date, information that would benefit a broad range of researchers remains scattered in the literature. Edited by two of the top pioneers in the field, FLIM Microscopy in Biology and Medicine presents the fundamentals of FLIM along with a number of advanced considerations so that a wider audience can appreciate recent and potential improvements that make it such a valuable tool.

In addition to reviewing the latest developments, applications, and approaches to data analysis, the book also takes measure of the current state of the field, presenting the pros and cons of different methods and suggesting where improvements are required. The book also describes ancillary techniques related to the direct determination of lifetimes, including imaging fluorescence anisotropy for the study of molecular rotations.

New Opportunities for Biomedical Researchers...New Challenges for Microscopy Researchers

Discussion sections in all the chapters clearly show the challenges for implementing FLIM for various applications. Certain chapters discuss limits on the number of photons required for highly accurate lifetime determinations as well as the accuracy with which multiple, closely associated lifetime components can reliably be determined. Such considerations are important for users when selecting the most advantageous method of FLIM to use for a particular application.

While this book provides an introduction for those new to FLIM, it gathers a wealth of material to enhance the work of experts involved with pioneering technological improvements or research opportunities in this unique and promising area of microscopy.
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