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fluorescence resonance energy transfer; 2.6 Two-photon fluorescence lifetime imaging; References; 3 Two-photon fluorescence microscopy through turbid media; 3.1 Two-photon fluorescence microscopy of microspheres embedded in turbid media; 3.1.1 Measurement of two-photon fluorescence images; 3.1.2 Comparison with Monte-Carlo simulation; 3.2 Limiting factors on image quality in imaging through turbid media

3.3 Quantitative comparison of penetration depth between two-photon excitation and single-photon excitation

References; 4 Fibre-optical nonlinear microscopy; 4.1 Fibre-optical confocal microscopy; 4.1.1 Image formation; 4.1.2 Milestones in fibre-optical confocal microscopy; 4.2 Two-photon fluorescence imaging systems using a single-mode optical fibre coupler; 4.2.1 Fibre-optical two-photon fluorescence microscopy; 4.2.2 Coupling efficiency and splitting ratio; 4.2.3 Spectral and temporal broadening; 4.2.4 Fluorescence axial response; 4.2.5 Three-dimensional optical transfer function analysis

4.2.6 Discussion

4.3 Fibre-optical second harmonic generation microscopy; 4.3.1 Coupling efficiency and splitting ratio; 4.3.2 Second-harmonic generated axial response; 4.3.3 Three-dimensional coherent transfer function analysis; 4.3.4 Polarisation anisotropy; 4.4 Towards nonlinear endoscopic imaging; 4.5 Summary; References; 5 Nonlinear optical endoscopy; 5.1 An introduction to nonlinear optical endoscopy; 5.1.1 Optical fibres and ultrashort pulse delivery; 5.1.2 Scanning mechanisms; 5.1.3 Geometries of fibre-optical nonlinear optical microscopy

5.2 Nonlinear optical microscopy using double-clad PCFs

5.2.1 Characterisation of double-clad PCFs; 5.2.2 Experimental arrangement; 5.2.3 Axial resolution; 5.2.4 Improvement of signal level; 5.2.5 Nonlinear optical imaging; 5.2.6 SHG polarisation anisotropy measurement; 5.3 A nonlinear optical endoscope based on a double-clad PCF and a MEMS mirror; 5.3.1 Endoscope design; 5.3.2 Axial resolution and signal level; 5.3.3 Endoscopic imaging; 5.3.4 3D tissue imaging; 5.4 Nonlinear optical microscopy using a double-clad PCF coupler; 5.4.1 A double-clad PCF coupler; 5.4.2 Experimental arrangement

5.4.3 System performance

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#### Sommario/riassunto

The introduction of femtosecond pulse lasers has provided numerous new methods for non-destructive diagnostic analysis of biological samples. This book is the first to provide a focused and systematic treatment of femtosecond biophotonic methods. Each chapter combines theory, practice and applications, walking the reader through imaging, manipulation and fabrication techniques. Beginning with an explanation of nonlinear and multiphoton microscopy, subsequent chapters address the techniques for optical trapping and the development of laser tweezers. In a conclusion that brings together the various topics of the book, the authors discuss the growing field of femtosecond micro-engineering. The wide range of applications for femtosecond biophotonics means this book will appeal to researchers and practitioners in the fields of biomedical engineering, biophysics, life sciences and medicine.

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