
Contents

Part I Ultrafast Lasers in Medicine

1 Ultrahigh-Resolution Optical Coherence Tomography Using Femtosecond Lasers

<i>J.G. Fujimoto, A.D. Aguirre, Y. Chen, P.R. Herz, P.-L. Hsiung, T.H. Ko, N. Nishizawa, and F.X. Kärtner</i>	3
1.1 Introduction	3
1.2 Measuring Ultrafast Optical Echoes	6
1.3 Low-Coherence Interferometry	7
1.4 Resolution of OCT	8
1.5 Ultrahigh-Resolution OCT Using Femtosecond Lasers	10
1.6 Ultrahigh-Resolution OCT Imaging Using Ti:Al ₂ O ₃ Femtosecond Lasers	11
1.7 Ultrahigh-Resolution Imaging Using Cr:Forsterite Femtosecond Lasers	15
1.8 Ultrahigh-Resolution OCT Imaging Using Femtosecond Nd:Glass Lasers	20
1.9 Three-Dimensional OCT (3D-OCT) Imaging	22
1.10 Summary	23
References	24

2 Two-Photon Laser Scanning Microscopy

<i>A. Nimmerjahn, P. Theer, and F. Helmchen</i>	29
2.1 Introduction	29
2.2 Theory and Technology	29
2.2.1 Two-Photon Fluorescence Excitation	29
2.2.2 Fluorescence Detection	33
2.2.3 Instrumentation	35
2.2.4 Fluorescence Labeling Techniques	36

VIII Contents

2.3	Applications	39
2.3.1	Functional Fluorescence Imaging	40
2.3.2	Photomanipulation	43
2.4	Limitations	44
2.4.1	Spatial and Temporal Resolution	44
2.4.2	Tissue Damage	45
2.5	Future Perspectives	46
	References	48

3 Femtosecond Lasers in Ophthalmology:

Surgery and Imaging

<i>J.F. Bille</i>	53
3.1 Introduction	53
3.2 Surgical Applications of Femtosecond Lasers in Ophthalmology...	54
3.2.1 Laser–Tissue Interaction	54
3.2.2 All-Solid-State Femtosecond Laser Technology	57
3.2.3 Clinical Instrumentation	60
3.2.4 Experimental Results	61
3.3 Imaging Applications of Femtosecond Lasers in Ophthalmology	63
3.3.1 Principles of Nonlinear Microscopic Imaging	63
3.3.2 Second Harmonic Generation Imaging of Collagen Fibrils in Cornea, Sclera, and Optic Nerve Head.....	64
3.3.3 Two Photon Excited Autofluorescence Imaging of Lipofuscin Granules in RPE	67
3.3.4 Aberration Free Retina Imaging with Closed-Loop Adaptive Optics	70
3.4 Conclusion and Outlook.....	71
References	72

Part II Ultrafast Lasers in Biology

4 Ultrafast Peptide and Protein Dynamics

by Vibrational Spectroscopy

<i>P. Hamm</i>	77
4.1 Introduction	77
4.2 The Challenge of Using IR Spectroscopy as Structure-Sensitive Method	78
4.3 Experimental Methods	80
4.4 Vibrational Spectroscopy of Equilibrium Dynamics of Peptides and Proteins	80
4.4.1 Photon Echo Spectroscopy	81
4.4.2 2D-IR Spectroscopy	83

4.5	Vibrational Spectroscopy of Nonequilibrium Dynamics of Peptides and Proteins	86
4.6	Conclusion and Outlook	90
	References	91

5 Photosynthetic Light-Harvesting

	<i>T. Pullerits, T. Polivka, and V. Sundström</i>	95
5.1	Introduction	95
5.2	Light-Harvesting in Photosynthetic Purple Bacteria: Energy Transfer and Trapping	96
5.2.1	B800	97
5.2.2	Excitons and Polarons in B850	98
5.2.3	Inter-Complex Excitation Transfer	100
5.3	Carotenoid Light-Harvesting in the Peridinin–Chlorophyll Protein (PCP)	104
5.3.1	Steady-State Spectroscopy	104
5.3.2	Energy Transfer Pathways	108
5.4	Carbonyl Carotenoids in Other Light-Harvesting Systems	111
	References	112

6 Primary Photosynthetic Energy Conversion in Bacterial Reaction Centers

	<i>W. Zinth and J. Wachtveitl</i>	117
6.1	Introduction	117
6.2	Structure and Absorption Spectra of Photosynthetic Reaction Centers	120
6.3	Ultrafast Reaction Steps	122
6.4	Some Remarks on Superexchange Electron Transfer	124
6.5	Superexchange vs. Stepwise Electron Transfer	126
6.6	Theoretical Description of the Picosecond ET	130
6.7	Experiments on Modified Reaction Centers	131
6.8	Optimization of Photosynthesis	132
6.9	Conclusion	135
	References	135

7 Ultrafast Primary Reactions in the Photosystems of Oxygen-Evolving Organisms

	<i>A.R. Holzwarth</i>	141
7.1	Structural Basis of Primary Photosynthetic Reactions	141
7.2	Photosystem I Structure	142
7.3	Photosystem II Structure	145
7.4	Energy Transfer Processes	147
7.4.1	Energy Transfer in Core Antenna/RC Particles	147
7.4.2	Is Energy Transfer from the Core to the RC Rate-Limiting?	147

7.4.3	Energy Transfer in PS I Cores	148
7.4.4	Energy Exchange with Red Chlorophylls in PS I Cores	149
7.4.5	Energy Transfer in PS II Cores	151
7.5	Electron Transfer Processes	152
7.5.1	Photosystem I Cores	152
7.5.2	Electron Transfer in PS II RCs	154
7.6	Conclusions	158
	References	158

8 Primary Photochemistry in the Photoactive Yellow Protein: The Prototype Xanthopsin

	<i>D.S. Larsen, R. van Grondelle, and K.J. Hellingwerf</i>	165
8.1	Introduction	165
8.1.1	Biological Function	166
8.1.2	PYP Structure	167
8.1.3	PYP Photocycle	169
8.2	Biophysical Techniques	171
8.3	Time-Resolved Fluorescence Signals	174
8.4	Electronically Resonant Transient Absorption Signals	176
8.4.1	Pump-Probe Measurements	177
8.4.2	Pump-Dump-Probe Measurements	183
8.5	Vibrationally Resonant Ultrafast Signals	186
8.6	Time-Resolved X-Ray Diffraction Measurements	189
8.7	Isolated PYP Chromophores	190
8.8	Quantum Calculations and Molecular Dynamics	192
8.9	Concluding Remarks	194
	References	195

9 Structure Based Kinetics by Time-Resolved X-ray Crystallography

	<i>M. Schmidt</i>	201
9.1	Introduction	201
9.1.1	Structure and Function of Proteins	201
9.1.2	Structure Determination of Intermediate States by Stabilization (Trapping) of their Occupation	203
9.2	Crystallography Meets Chemical Kinetics	206
9.2.1	Chemical Kinetics	206
9.2.2	Time-Resolved X-Ray Structure Analysis	208
9.3	From the Reaction Initiation to Difference Electron Density Maps	213
9.3.1	Reaction Initiation	213
9.3.2	Detectors	214
9.3.3	Data Reduction	215
9.3.4	Difference Maps	215

9.4	Experiments	216
9.4.1	Myoglobin	216
9.4.2	The Photoactive Yellow Protein	218
9.5	A New Method for the Analysis of Time-Resolved X-ray Data ...	220
9.5.1	The Singular Value Decomposition	220
9.5.2	The Noise Filter	222
9.5.3	Transient Kinetics and Kinetic Mechanisms from the SVD	224
9.5.4	Determination of the Structures of the Intermediates	226
9.5.5	Posterior Analysis	227
9.5.6	Verification of the Functionality of the SVD-Driven Analysis by Mock Data	229
9.6	The SVD Analysis of Experimental Time-Resolved Data	230
9.6.1	SVD-Flattening	230
9.6.2	The Mechanistic Analysis of the PYP data	230
9.6.3	The Structures of the Intermediates in the Late Photocycle Between 5 μ s and 100ms	232
9.6.4	Plausible Kinetic Mechanisms	232
9.6.5	The Entire Photocycles of the Wild-Type PYP and its E46Q-Mutant	234
9.7	Picosecond Time Resolution and Beyond	235
9.8	More Applications	236
	References	237
 10 Primary Reactions in Retinal Proteins		
	<i>R. Diller</i>	243
10.1	Introduction	243
10.2	Systems	246
10.3	A First Glance at the Primary Reaction Dynamics	249
10.3.1	11- <i>Cis</i> \rightarrow All- <i>Trans</i> Isomerization	250
10.3.2	All- <i>Trans</i> \rightarrow 13- <i>Cis</i> Isomerization	252
10.4	Discussion	256
10.4.1	When Does Isomerization Occur?	256
10.4.2	Ultrafast Electronic Surface Crossing	263
10.4.3	Reaction Models	264
10.4.4	Wavepacket Dynamics after Electronic Excitation	266
10.4.5	Chromophore-Protein Interaction	268
	References	271
 11 Ultrashort Laser Pulses in Single Molecule Spectroscopy		
	<i>E. Haustein and P. Schwille</i>	279
11.1	Introduction	279
11.2	Basic Concepts of Fluorescence	279
11.2.1	Fluorescence	279
11.2.2	Fluorescence Lifetime	281

XII Contents

11.2.3	Fluorescent Dyes	284
11.2.4	Autofluorescent Proteins	284
11.2.5	Organic Chromophores	284
11.2.6	Quantum Dots	285
11.3	Instrumentation and Set-up	286
11.3.1	Confocal Set-up: Continuous-Wave (cw-) Excitation	286
11.3.2	Confocal Set-up: Pulsed Excitation	287
11.4	Time-Related Single Photon Counting (TCSPC)	289
11.4.1	Fluorescence Lifetime	289
11.4.2	Instrument Response Function (IRF)	291
11.4.3	Analysis of Fluorescence Decays	292
11.5	Fluorescence Correlation Spectroscopy (FCS)	294
11.5.1	One-Photon Excitation	294
11.6	Two-Photon Excitation	297
11.6.1	Correlation of Photon Arrival Times	298
11.7	Gated Detection	300
11.7.1	Time-Resolved Fluorescence Correlation Spectroscopy	301
11.8	Lifetime-Assisted Crosstalk-Suppression for Cross-Correlation Spectroscopy	302
11.9	Anisotropy	302
11.9.1	Theory	302
11.9.2	Time-Resolved Fluorescence Anisotropy	303
11.9.3	Static Anisotropy	303
11.9.4	Time-Resolved Anisotropy	304
11.10	“Burst”-Analysis	304
11.11	Conclusions	306
	References	306
	Index	311

Ultrashort Laser Pulses in Biology and Medicine

Braun, M.; Gilch, P.; Zinth, W. (Eds.)

2008, XV, 320 p., Hardcover

ISBN: 978-3-540-73565-6