

## Contents

### List of Contributors XIII

### Part I Imaging and Diagnostics 1

|          |  |          |
|----------|--|----------|
| <b>1</b> | <b>Quantum Dots for Cancer Imaging</b>   | <b>3</b> |
|          | <i>Yan Xiao and Xiugong Gao</i>  |          |
| 1.1      | Introduction   | 3        |
| 1.2      | Cancer   | 5        |
| 1.2.1    | A Primer on Cancer Biology   | 5        |
| 1.2.2    | The Importance of Early Cancer Detection and Diagnosis                         | 6        |
| 1.2.3    | The Role of Biomarkers in Cancer Early Detection and Diagnosis                 | 7        |
| 1.3      | Quantum Dots: Physics and Chemistry  | 8        |
| 1.3.1    | Photophysical Properties of QDs  | 8        |
| 1.3.2    | Quantum Dot Chemistry  | 11       |
| 1.3.2.1  | Synthesis  | 11       |
| 1.3.2.2  | Surface Passivation  | 12       |
| 1.3.2.3  | Water Solubilization   | 12       |
| 1.3.2.4  | Bioconjugation   | 14       |
| 1.4      | Cancer Imaging with QDs  | 15       |
| 1.4.1    | <i>In Vitro</i> Screening and Detection of Cancer Biomarkers Using Microarrays | 15       |
| 1.4.2    | <i>In Vitro</i> Cellular Labeling of Cancer Biomarkers                         | 19       |
| 1.4.2.1  | Labeling of Fixed Cells and Tissues  | 19       |
| 1.4.2.2  | Live Cell Imaging in Cancer Cells  | 22       |
| 1.4.3    | <i>In Vivo</i> Cancer Imaging  | 28       |
| 1.4.3.1  | <i>In Vivo</i> Tracking of Cancer Cells  | 29       |
| 1.4.3.2  | Tumor Vasculature Imaging  | 31       |
| 1.4.3.3  | Sentinel Lymph Node Mapping and Fluorescence Lymphangiography                  | 33       |
| 1.4.3.4  | <i>In Vivo</i> Whole-Body Tumor Imaging in Animals                             | 35       |
| 1.4.4    | Multimodality Tumor Imaging  | 39       |
| 1.4.5    | Dual-Functionality QDs for Cancer Imaging and Therapy                          | 41       |
| 1.5      | Quantum Dot Cytotoxicity and Potential Safety Concerns                         | 45       |

|          |   |            |
|----------|---|------------|
| 1.6      | Concluding Remarks and Future Perspectives                                  | 47         |
|          | Abbreviations   | 48         |
|          | Acknowledgments   | 50         |
|          | References  | 50         |
| <b>2</b> | <b>Quantum Dots for Targeted Tumor Imaging</b>                              | <b>63</b>  |
|          | <i>Eue-Soon Jang and Xiaoyuan Chen</i>                                      |            |
| 2.1      | Introduction  | 63         |
| 2.2      | Types of Quantum Dots (QDs)   | 64         |
| 2.2.1    | Type I Core–Shell QDs   | 73         |
| 2.2.1.1  | Group II–VI Semiconductor Core–Shell QDs                                    | 74         |
| 2.2.1.2  | Group III–V Semiconductor Core–Shell QDs                                    | 76         |
| 2.2.1.3  | Group IV–VI Semiconductor Core–Shell QDs                                    | 78         |
| 2.2.2    | Reverse Type I and Type II Core–Shell QDs                                   | 78         |
| 2.2.3    | Core–Shell–Shell (CSS) QDs  | 81         |
| 2.3      | Surface Modifications of QDs for Bioapplications                            | 83         |
| 2.3.1    | Preparation of Water-Soluble QDs  | 83         |
| 2.3.2    | Bioconjugation of QDs   | 88         |
| 2.4      | Methods of Delivering QDs into Tumors                                       | 91         |
| 2.4.1    | Passive Targeting   | 91         |
| 2.4.2    | Active Targeting  | 95         |
| 2.4.2.1  | Peptide-Mediated Tumor Uptake   | 95         |
| 2.4.2.2  | Antibody-Mediated Uptake  | 97         |
| 2.5      | Recent Advances in QDs Technology   | 99         |
| 2.5.1    | Bioluminescence Resonance Energy Transfer (BRET)                            | 99         |
| 2.5.2    | Non-Cd-Based QDs  | 100        |
| 2.5.3    | Moving Towards Smaller QDs  | 102        |
| 2.5.4    | Multifunctional Probes  | 102        |
| 2.6      | Conclusion and Perspectives   | 104        |
|          | Acknowledgments   | 105        |
|          | References  | 105        |
| <b>3</b> | <b>Multiplexed Bioimaging Using Quantum Dots</b>                            | <b>115</b> |
|          | <i>Richard Byers and Eleni Tholouli</i>                                     |            |
| 3.1      | Introduction  | 115        |
| 3.2      | The Need for Novel Multiplex Imaging Systems                                | 115        |
| 3.3      | Quantum Dots  | 117        |
| 3.3.1    | Optical Properties  | 117        |
| 3.3.2    | Manufacture   | 119        |
| 3.4      | Bioimaging Applications of Quantum Dots                                     | 120        |
| 3.4.1    | Quantum Dot Use for Immunohistochemistry                                    | 120        |
| 3.4.2    | Quantum Dot Use for <i>In Situ</i> Hybridization                            | 125        |
| 3.4.3    | Quantum Dot Use in Solid- and Liquid-State Multiplex<br>Detection Platforms | 129        |
| 3.5      | Translation to Clinical Biomarker Measurement                               | 131        |
| 3.5.1    | Quantitation  | 131        |

- 3.5.2 Imaging Analysis 132
- 3.5.3 Combinational Microscopy Methods 134
- 3.5.4 Clinical and Mechanistic Biological Applications 134
- 3.5.5 Cancer Molecular Profiling 137
- 3.6 Summary and Future Perspectives 139
- References 142

#### **4 Multiplexed Detection Using Quantum Dots 147**

*Young-Pil Kim, Zuyong Xia and Jianghong Rao*

- 4.1 Introduction 147
- 4.2 *In Vitro* Multiplexed Analysis Using QDs 150
  - 4.2.1 DNA Hybridization 150
  - 4.2.2 Immunoassay 153
  - 4.2.3 Assaying the Activity of Enzymes 157
    - 4.2.3.1 FRET-Based Protease Detection with QDs as the Donor 158
    - 4.2.3.2 BRET-Based Protease Detection with QDs as Acceptor 161
  - 4.2.4 Other *In Vitro* Multiplexed Detection Systems 163
- 4.3 Multiplexed Imaging Using QDs 165
  - 4.3.1 Multiplexed Cellular Imaging 165
  - 4.3.2 Multiplexed Imaging in Small Animals 168
- 4.4 Summary 171
- References 171

### **Part II Therapy 177**

#### **5 Medical Diagnostics of Quantum Dot-Based Protein Micro- and Nanoarrays 179**

*Anisha Gokarna and Yong-Hoon Cho*

- 5.1 Introduction 179
  - 5.1.1 Invention of Protein Microarrays 180
  - 5.1.2 Optical Readout of Protein Arrays: Traditional versus New Labels 182
- 5.2 Protein Arrays 185
  - 5.2.1 The Various Types of Protein Array 185
  - 5.2.2 Methods of Optical Detection in Protein Arrays Using Labeled Probes 186
- 5.3 From Microarrays to Nanoarrays: Why Nanoarrays? 188
- 5.4 Functionalization of QDs for Attachment to Proteins or Other Biomolecules 189
- 5.5 Fabrication of Protein Biochips 193
  - 5.5.1 Printing of QDs-Conjugated Protein Microarray Biochips 193
  - 5.5.2 Nanoarray Protein Chips Using QDs 194
- 5.6 QDs Probes in Clinical Applications 197
  - 5.6.1 Detection of Disease (Cancer) Biomarkers Using QD Labels 197
  - 5.6.2 Certain Limitations in QDs Labeled Microarrays: Nonspecific Binding Effects 201
- 5.6.3 QDs versus Organic Dyes in Protein Microarrays 203

|          |   |            |
|----------|---|------------|
| 5.6.4    | Multiplexing in Protein Microarrays Using QDs   | 206        |
| 5.7      | QDs-Labeled Protein Nanoarrays  | 207        |
| 5.8      | Summary and Future Perspectives   | 212        |
|          | Acknowledgments   | 212        |
|          | References  | 212        |
| <b>6</b> | <b>Imaging and Tracking of Viruses Using Quantum Dots</b>   | <b>219</b> |
|          | <i>Kye-Il Joo, April Tai and Pin Wang</i>   | 219        |
| 6.1      | Introduction  | 219        |
| 6.2      | Quantum Dot–Biomolecule Hybrids   | 220        |
| 6.2.1    | Optical Properties of QDs   | 220        |
| 6.2.2    | Functionalized QDs Conjugated with Biomolecules   | 220        |
| 6.2.3    | Formation of Virus–QD Networks  | 222        |
| 6.2.4    | Decoration of Discretely Immobilized Virus with QDs   | 222        |
| 6.2.5    | QD Encapsulation in Viral Capsids   | 224        |
| 6.3      | Quantum Dots for Single Virus Tracking in Living Cells  | 225        |
| 6.3.1    | Conventional Labeling for Single-Virus Tracking in Live Cells   | 228        |
| 6.3.2    | Problems Encountered in Single-Virus Tracking   | 229        |
| 6.3.3    | Use of QDs for Labeling Enveloped Viruses   | 230        |
| 6.3.4    | Use of QDs for Labeling Nonenveloped Viruses  | 231        |
| 6.4      | Quantum Dots for the Sensitive Detection of Virus and Infection   | 239        |
| 6.4.1    | Monitoring the Progression of Viral Infection with Fluorescent QD Probes  | 239        |
| 6.4.2    | The Use of Two-Color QDs for the Real-Time Detection of Viral Particles and Viral Protein Expression                | 241        |
| 6.4.3    | Viral Detection with pH-Sensitive QDs in a Biological Motor   | 242        |
| 6.5      | Conclusions   | 245        |
|          | Acknowledgments   | 245        |
|          | References  | 246        |
| <b>7</b> | <b>Nanomaterials for Radiation Therapy</b>  | <b>251</b> |
|          | <i>Ke Sheng and Wensha Yang</i>   |            |
| 7.1      | Introduction  | 251        |
| 7.2      | A Brief Introduction to Radiation Therapy, and its Limitations  | 252        |
| 7.3      | Physical Radiosensitizers   | 255        |
| 7.3.1    | Enhanced Radiation Therapy Using High Z but Non-Nanoscaled Materials  | 255        |
| 7.3.2    | Enhanced Radiation Therapy by Gold Nanoparticles  | 257        |
| 7.3.3    | Dose Enhancement of Physical Radiosensitizers   | 259        |
| 7.3.4    | Radiation Therapy Enhancement Using Nonionizing Radiation   | 260        |
| 7.4      | Radiation Therapy in Combination with Photodynamic Therapy Using Semiconductor Nanoparticles as the Energy Mediator | 261        |
| 7.4.1    | Photodynamic Therapy  | 261        |
| 7.4.2    | Semiconductor Nanoparticles as the Energy Mediator for Photodynamic Therapy   | 262        |

|       |  |     |
|-------|--|-----|
| 7.4.3 | Quantum Dots   | 263 |
| 7.4.4 | Photoluminescent Nanoparticles in Radiation Therapy    | 264 |
| 7.5   | Nanobrachytherapy                                      | 271 |
| 7.5.1 | Liposomes  | 273 |
| 7.5.2 | Nanoparticles  | 274 |
| 7.5.3 | Dendrimers   | 275 |
| 7.6   | Nanoparticles as Radioprotectors                       | 277 |
| 7.7   | Radiation Dosimeters Using Semiconductor Nanomaterials | 280 |
| 7.8   | Conclusions  | 282 |
|       | References   | 283 |

## **8 Prospects of Semiconductor Quantum Dots for Imaging and Photodynamic Therapy of Cancer** 291

*Vasudevanpillai Biju, Sathish Mundayoor, Abdulaziz Anas and Mitsuru Ishikawa*

|         |  |     |
|---------|--|-----|
| 8.1     | Introduction   | 291 |
| 8.2     | Basic Principles and Challenges in PDT                                 | 292 |
| 8.3     | Advantages of Quantum Dots for PDT                                     | 294 |
| 8.4     | Synthesis of Quantum Dots  | 295 |
| 8.4.1   | Synthesis of Visible QDs   | 295 |
| 8.4.1.1 | Synthesis of Cadmium-Based QDs   | 295 |
| 8.4.1.2 | Synthesis of InP QDs   | 296 |
| 8.4.2   | Synthesis of NIR QDs   | 296 |
| 8.4.2.1 | Synthesis of Core-Only NIR QDs   | 297 |
| 8.4.2.2 | Synthesis of Core-Shell QDs  | 297 |
| 8.4.2.3 | Synthesis of CdTe/CdSe QDs   | 298 |
| 8.5     | Optical Properties of Quantum Dots                                     | 299 |
| 8.5.1   | Absorption and Fluorescence Properties                                 | 299 |
| 8.5.2   | Photostability of QDs  | 301 |
| 8.5.3   | Two-Photon Absorption by QDs   | 301 |
| 8.6     | Preparation of Biocompatible Quantum Dots                              | 303 |
| 8.7     | Nontargeted Intracellular Delivery of QDs                              | 304 |
| 8.7.1   | Physical Methods of Intracellular Delivery                             | 305 |
| 8.7.2   | Biochemical Techniques   | 307 |
| 8.7.2.1 | Nonspecific Intracellular Delivery of QDs                              | 307 |
| 8.7.2.2 | Cell-Penetrating Peptide-Mediated Delivery                             | 307 |
| 8.7.2.3 | Chitosan- and Liposome-Mediated Delivery                               | 309 |
| 8.8     | Targeting Cancer Cells with QDs  | 309 |
| 8.8.1   | <i>In Vitro</i> Targeting of Cancer Cells with QDs                     | 310 |
| 8.8.1.1 | Targeting Cancer Cells with QD-Antibody Conjugates                     | 310 |
| 8.8.1.2 | Targeting Cancer Cells with QD-Ligand Conjugates                       | 312 |
| 8.8.2   | <i>In Vivo</i> Targeting and Imaging Cancer with QDs                   | 313 |
| 8.8.3   | <i>In Vivo</i> Targeting of Tumor Vasculature and Lymph Nodes with QDs | 314 |
| 8.9     | Quantum Dots for Photodynamic Therapy of Cancer                        | 316 |
| 8.9.1   | Quantum Dot Alone for PDT  | 316 |

- 8.9.2 Potentials of QD–Photosensitizer Conjugates for PDT 318
- 8.10 Toxicity of QDs 320
- 8.11 Conclusions 321
- Abbreviations 322
- References 323

### Part III Synthesis, Characterization, and Toxicology 329

#### 9 Type-I and Type-II Core–Shell Quantum Dots: Synthesis and Characterization 331

*Dirk Dorfs, Stephen Hickey and Alexander Eychmüller*

- 9.1 Introduction 331
- 9.2 Core–Shell UV-Vis Nanoparticulate Materials 332
  - 9.2.1 Type-I Core–Shell Structures 332
  - 9.2.2 Type-II Core–Shell Structures 334
  - 9.2.3 Multiple Shell Structures 337
  - 9.2.4 Nonspherical Nanoheterostructures 349
- 9.3 Characterization of Nanoheterostructures 350
- 9.4 Core-Shell Infrared Nanoparticulate Materials 352
- 9.5 Type I Core–Shell Infrared Structures 353
- 9.6 Type II Core–Shell Infrared Structures 355
- 9.7 Summary and Conclusions 361
- References 362

#### 10 Nanowire Quantum Dots 367

*Thomas Aichele, Adrien Tribu, Gregory Sallen, Catherine Bougerol, Régis André, Jean-Philippe Poizat, Kuntheak Kheng and Serge Tatarenko* 367

- 10.1 Introduction 367
- 10.2 Quantum Dots 368
- 10.3 Growth of Quantum Dots, Nanowires and Nanowire Heterostructures 371
- 10.4 Applications for Nanowires and Nanowire Quantum Dots 378
  - 10.4.1 Nanowires and Quantum Dots in the Life Sciences 378
  - 10.4.2 Nanowire Electronic Devices 379
  - 10.4.3 Single-Photon Sources 380
    - 10.4.3.1 Single-Photon Generation 380
    - 10.4.3.2 High-Temperature Single-Photon Emission from Nanowire QDs 381
- 10.5 Conclusions 386
- References 387

#### 11 Quantum Dot–Core Silica Glass–Shell Nanomaterials: Synthesis, Characterization, and Potential Biomedical Applications 393

*Norio Murase*

- 11.1 Introduction 393
- 11.2 Historical Overview 394

|           |  |            |
|-----------|--|------------|
| 11.2.1    | Quantum Dots and their Incorporation into Glass Matrices                               | 394        |
| 11.2.2    | Single-Particle Spectroscopy and its Application to Biological Systems                 | 396        |
| 11.3      | Incorporation of Quantum Dots in Glass Beads for Bioapplications                       | 397        |
| 11.3.1    | The Stöber Method  | 398        |
| 11.3.2    | Reverse Micelle Method   | 398        |
| 11.3.3    | Bifunctional Glass Beads Derived from the Reverse Micelle Method                       | 404        |
| 11.3.4    | Complex Structures Created in Small Glass Beads with Novel Photoluminescent Properties | 409        |
| 11.3.5    | Emitting Glass Fibers Created by Self-Assembly of the Complex Structured Beads         | 414        |
| 11.4      | Preparation of Cadmium-Free Quantum Dots With Water Dispersibility                     | 414        |
| 11.5      | Summary and Future Perspective   | 422        |
|           | Acknowledgments  | 423        |
|           | References   | 423        |
| <b>12</b> | <b>Toxicology and Biosafety Evaluations of Quantum Dots</b>                            | <b>429</b> |
|           | <i>Pinpin Lin, Raymond H.S. Yang, Chung-Shi Yang, Chia-Hua Lin and Louis W. Chang</i>  |            |
| 12.1      | Introduction   | 429        |
| 12.2      | The Physico-Chemical Properties of Quantum Dots  | 432        |
| 12.2.1    | Surface Chemistry and Coatings   | 433        |
| 12.2.2    | Charge and Size of QDs in Relationship to Potential Toxicity                           | 435        |
| 12.2.3    | Core Metals and Biodegradation of QDs  | 437        |
| 12.3      | Toxicity and Role of Oxidative Stress  | 441        |
| 12.4      | ADME and Biosafety Evaluation  | 445        |
| 12.5      | The Mitochondrion as a Prime Target for QD Toxicity                                    | 450        |
| 12.6      | Environmental and Ecological Concerns  | 456        |
| 12.7      | Concluding Comments and Future Perspectives  | 456        |
|           | References   | 458        |
|           | <b>Index</b>   | <b>465</b> |

