
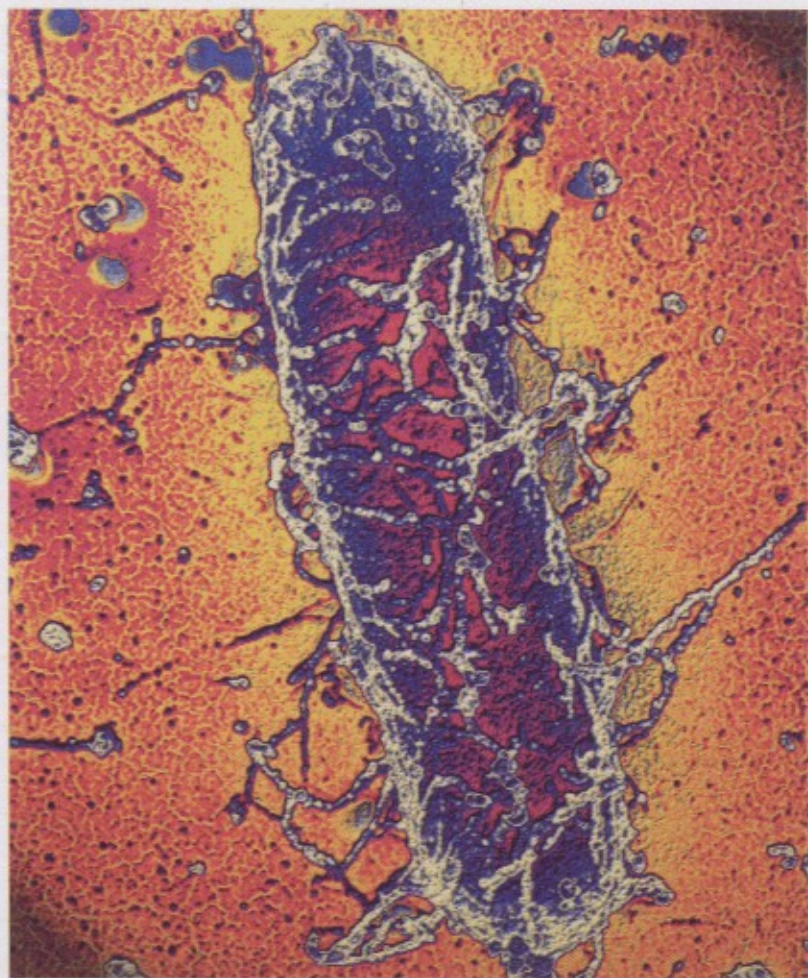


Nanotechnologies for the Life Sciences

Edited by Challa Kumar

 WILEY-VCH

Biofunctionalization of Nanomaterials



ntLS 

Contents

Preface XIII

List of Contributors XVII

1	Biofunctionalization of Fluorescent Nanoparticles	1
	<i>Michael J. Murcia and Christoph A. Naumann</i>	
1.1	Introduction	1
1.2	Fluorescent Nanoparticle Probes	2
1.2.1	Dye-doped Nanoparticles	3
1.2.2	Quantum Dots (QDs)	5
1.2.3	Metal Nanoparticles	7
1.2.4	Hybrid Architectures Involving Fluorescent Nanoparticles	9
1.2.4.1	Metal–Dye	9
1.2.4.2	Dye-doped Silica Shells	9
1.2.4.3	Quantum Dot-containing Microspheres	10
1.3	Bioconjugation of Fluorescent Nanoparticles	11
1.3.1	General Considerations	11
1.3.1.1	Overview	11
1.3.1.2	Common Coupling Reactions	13
1.3.2	Bioconjugation of Polymeric Nanoparticles	13
1.3.2.1	Noncovalent Approaches	13
1.3.2.2	Covalent Approaches	15
1.3.3	Bioconjugation of Quantum Dots	15
1.3.3.1	Noncovalent Approaches	16
1.3.3.2	Covalent Approaches	16
1.3.4	Bioconjugation of Metallic Nanoparticles	16
1.3.4.1	Noncovalent Approaches	17
1.3.4.2	Covalent Approaches	17
1.4	Design of Biocompatible Coatings	17
1.4.1	General Considerations	17
1.4.1.1	Overview	17
1.4.1.2	Colloidal Stability	18
1.4.1.3	Biocompatible Surfaces	19

1.4.1.4	Cytotoxicity	20
1.4.2	Nanoparticle-stabilizing Coatings	21
1.4.3	Low Cytotoxicity Coatings	23
1.5	Applications	23
1.5.1	Biosensing	24
1.5.1.1	Polymeric Sensors	25
1.5.1.2	Quantum Dot Sensors	25
1.5.1.3	Metallic Sensors	26
1.5.2	Fluorescent Nanoparticles as Labels in Biological Imaging	27
1.5.2.1	Dye-doped Nanoparticles	27
1.5.2.2	Quantum Dots	27
	References	29

2 Biofunctionalization of Carbon Nanotubes 41

Elena Bekyarova, Robert C. Haddon, and Vladimir Parpura

2.1	Introduction	41
2.2	Carbon Nanotubes – Types, Structures and Properties	42
2.3	Synthesis of Carbon Nanotubes	43
2.4	Approaches to Aqueous Solubilization of Carbon Nanotubes	47
2.4.1	Chemical Modifications	47
2.4.2	Use of Water-compatible Surfactants	47
2.4.3	Functionalization with Water-soluble Polymers	48
2.4.4	Interaction and Functionalization with Biological Molecules	49
2.4.4.1	Noncovalent Biofunctionalization	50
2.4.4.2	Covalent Biofunctionalization	52
2.5	Applications of Biofunctionalized Carbon Nanotubes	54
2.5.1	Assembly of Electronic Devices	54
2.5.2	Biosensing	58
2.5.3	Substrates for Neuronal Growth	63
2.6	Concluding Remarks	65
	Acknowledgments	65
	References	65

3 Biofunctionalization of Magnetic Nanoparticles 72

Yong Gao

3.1	Introduction	72
3.2	Functionalization of Magnetic Nanoparticles for <i>In Vitro</i> Protein/Cell Separation	74
3.3	Functionalization of Magnetic Nanoparticles for Biochemical/Chemical Synthesis of Therapeutic Drugs and Their Intermediates	80
3.4	Functionalization of Magnetic Nanoparticles for <i>In Vivo</i> Bio-imaging, Drug Targeting and Tumor Hyperthermia Treatments	82
3.4.1	MR Imaging	83
3.4.2	Targeted Drug Delivery	86
3.4.3	Magnetic Hyperthermia	87

3.5	Conclusions	88
	Acknowledgments	89
	References	89
4	Biofunctionalization of Gold Nanoparticles	99
	<i>Ming Zheng and Xueming Huang</i>	
4.1	Introduction	99
4.2	General Synthetic Routes	99
4.2.1	Direct Synthesis of Ligand-protected Au NPs	100
4.2.1.1	Strongly Ionic Ligand-protected Au NPs	102
4.2.1.2	Weakly Ionic Ligand-protected Au NPs	102
4.2.1.3	Au NPs Protected with Neutral Ligands	102
4.2.2	Ligand Exchange Reaction	103
4.3	Preparative-scale Synthesis and Solution-phase Characterization of DNA-directed Nanoparticle Assemblies	103
4.4	Bifunctional Proteins for Programmable Assembly of Nanoparticles	111
4.5	Strategies for Eliminating Nonspecific Interactions and Enabling Specific Binding with Biomolecules	113
4.6	Biological Applications	118
4.6.1	Nucleic Acids	118
4.6.2	Proteins	118
4.6.3	Cells and Virus	119
	Acknowledgments	120
	References	120
5	Biofunctionalization of Phospholipid Polymer Nanoparticles	125
	<i>Junji Watanabe, Jongwon Park, Tomomi Ito, Madoka Takai, and Kazuhiko Ishihara</i>	
5.1	Introduction	125
5.2	Nanofabrication for Biomedical Applications	126
5.2.1	Nano-scaled Processing	126
5.2.2	Key Materials for Nanofabrication	127
5.3	Design of Bioconjugate Nanoparticles	129
5.3.1	Bioconjugate Phospholipid Polymer	129
5.3.2	Solution Properties by Fluorescence Probe	129
5.3.3	Bioconjugate Nanoparticles	131
5.3.4	Surface Elemental Analysis by X-ray Photoelectron Spectroscopy	132
5.3.5	Surface ζ -Potential on Nanoparticles	132
5.3.6	Particle Size by Dynamic Light Scattering and Morphology by Scanning Electron Microscope	134
5.3.7	Determination of Active Ester Groups on Nanoparticles	135
5.4	Biofunction on Nanoparticles	137
5.4.1	Design of Sequential Enzymatic Reaction	137
5.4.2	Amplified Signal on Nanoparticles	138

5.5	Application for Molecular Diagnosis	139
5.5.1	Example of C-reactive Protein Detection Using Nanoparticles	139
5.5.2	High-performance Diagnosis in Serum	143
5.6	Conclusions	145
	Acknowledgments	145
	References	145

6 Biofunctionalization of Metallic Nanoparticles and Microarrays for Biomolecular Detection 150

Grit Festag, Uwe Klenz, Thomas Henkel, Andrea Csáki, and Wolfgang Fritzsche

6.1	Introduction	150
6.1.1	Applications	151
6.1.2	Array Fabrication	151
6.1.3	Detection Methods	153
6.1.3.1	Optical Absorbance	153
6.1.3.2	SPR Imaging	155
6.1.3.3	Raman Scattering	155
6.1.3.4	Electrical Detection	156
6.1.3.5	Electrochemical Detection	156
6.1.3.6	Gravimetric	158
6.2	Nanoparticles and their Biofunctionalization	158
6.2.1	Types of Nanoparticles used for Biomolecular Detection	159
6.2.1.1	Metal Nanoparticles	159
6.2.1.2	Core/Shell Particles	159
6.2.1.3	Magnetic Nanoparticles	160
6.2.1.4	Quantum Dots	160
6.2.2	Synthesis of Gold (Silver) Nanoparticles	161
6.2.3	Biofunctionalization	161
6.2.3.1	Modification of Gold Nanoparticles with Oligonucleotides/DNA	162
6.2.3.2	Modification of Gold Nanoparticles with Proteins	165
6.2.3.3	Biofunctionalization of other Metal Nanoparticles	167
6.2.4	Biological Applications of Gold Nanoparticles	167
6.3	Substrates and their Biofunctionalization	168
6.3.1	Molecular Thin Films	169
6.3.1.1	Self-assembly Monolayers	169
6.3.1.2	Optimization of Gold Nanoparticle-based Microarrays for DNA Detection	171
6.3.2	Nanoporous Gels	172
6.4	Outlook	175
	References	176

7 Conjugation of Nanomaterials with Proteins 183

Mohammed J. Meziani, Yi Lin, and Ya-Ping Sun

7.1	Introduction	183
7.2	Coupling of Inorganic Nanoparticles with Proteins	184

7.2.1	Chemical Functionalization Methods	184
7.2.2	Protein-assisted Assemblies of Inorganic Nanoparticles	188
7.2.2.1	Crosslinking Route through Protein Recognition	188
7.2.2.2	Template-directed Approach	191
7.2.3	Supercritical Fluid Methods	195
7.2.3.1	BSA-conjugated Silver Nanoparticles	196
7.2.3.2	BSA-conjugated Semiconductor Nanoparticles	197
7.2.3.3	Assembly and Disassembly of Nanoparticles through Protein Isomeric Conversion	202
7.3	Coupling of Carbon Nanotubes and Proteins	204
7.3.1	Non-specific Adsorption	206
7.3.2	Specific Conjugation and Biorecognition	211
7.4	Conclusions and Perspectives	221
	Acknowledgment	221
	References	222

8 Stabilization and Functionalization of Metallic Nanoparticles: the Peptide Route 235

Raphaël Lévy and R. Christopher Doty

8.1	Introduction	235
8.2	Metallic Nanoparticles – An Overview	236
8.2.1	Metallic Nanoparticles – Preparation	236
8.2.2	Metallic Nanoparticles – Optical Properties	238
8.2.3	Metallic Nanoparticles – Applications	242
8.3	Stabilization and Functionalization of Metallic Nanoparticles – The Peptide Route	248
8.3.1	Peptides, Proteins and Nanoscale Science	248
8.3.2	Peptide Toolbox for Bionanotechnology	249
8.3.3	Peptides as Capping Ligands	250
8.3.3.1	Interactions of Amino Acids with Noble Metals	250
8.3.3.2	Peptides as Reducing Agent and Template in Metallic Nanoparticle Synthesis	250
8.3.3.3	Rational Design of a Peptide Capping Ligands for Gold Nanoparticles: CALNN	251
8.3.3.4	Combinatorial Exploration of Peptides as Capping Ligands: the CALNN Family	252
8.3.3.5	Peptide-capped Silver Nanoparticles	252
8.3.3.6	Peptides as Capping Ligands for Fluorescent and Magnetic Nanoparticles	252
8.3.4	Peptide Extensions to Introduce Functionalities	253
8.3.4.1	Biotin and Strep-tag II	253
8.3.4.2	Peptide-DNA Hybrids	254
8.3.4.3	His-tag and Nickel Nitrilotriacetic Acid (Ni-NTA)	254
8.3.5	Chromatography of Peptide-capped Nanoparticles	255
8.3.5.1	Size-exclusion Chromatography	256

8.3.5.2	Affinity Chromatography	256
8.3.6	Recognition of Materials	256
8.3.7	Peptide-based Linkers	258
8.3.7.1	A Peptide-Peptide Linker Based on Leucine-zipper Sequences	259
8.3.7.2	A Peptide-DNA Linker Based on Metallopeptides	259
8.3.7.3	A Peptide-Texas Red Linker Obtained by Phage Display	259
8.3.8	Biologically Active Peptides	259
8.3.9	Self-assembling Peptides	260
8.3.9.1	Fibers and Nanotubes	260
8.3.9.2	Peptide-based Amphiphiles	262
8.4	Concluding Remarks	262
	References	263

9 Folate-linked Lipid-based Nanoparticles for Tumor-targeted Gene Therapy 270

Yoshiyuki Hattori and Yoshie Maitani

9.1	Introduction	270
9.2	Gene Delivery and Expression System	270
9.3	Nanoparticles for Gene Delivery System	271
9.4	Folate-linked Vectors	272
9.4.1	Folate Receptors	273
9.4.2	Folate Receptor-targeting Liposomes	273
9.5	Folate-linked Lipid-based Nanoparticles	277
9.5.1	Formulations	277
9.5.2	Nanoplex and Transfection Activity <i>In Vitro</i>	280
9.5.3	Selectivity of Folate-linked Nanoparticle	282
9.5.4	Transfection Activity <i>In Vivo</i>	285
9.6	Application of Suicide Gene Therapy	287
9.7	Conclusions	291
	List of Abbreviations	292
	References	293

10 Magnetic Core Conducting Polymer Shell Nanocomposites for DNA Attachment and Hybridization 299

Jean-Paul Lellouche

10.1	Introduction	299
10.2	Chemical Design of DPyr- and DCbz-containing Monomers: Introduction of Molecular Diversity	301
10.3	Synthetic Approaches for Mono- and Dicarboxylated DPyr-/DCbz-based Monomers	302
10.4	Oxidative Polymerization of DPyr-/DCbz-based Monomers around Magnetite Nanoparticles	305
10.4.1	General Considerations	305
10.4.2	Characterization of Magnetically Responsive PolyDPyr- and PolyDCbz-Magnetite Nanocomposites	307

10.5	Development of a DNA-based Biological System for Nanocomposite Parallel Screening	311
10.5.1	Covalent Attachment of an NH_2 -5'-modified 20-mer DNA Probe onto NCs towards DNA-Biofunctionalized NCs. Covalent Amide Bond Chemistry and Resulting NC-Supported DNA Hybridizations	313
10.5.2	Attachment of a Biotin-5'-modified 20-mer DNA Probe to DNA-biofunctionalized NCs. Quasi-covalent Linkage Using the Streptavidin-Biotin System and the Resulting NC-supported DNA Hybridizations	318
10.5.3	Storage: Medium-term Stability of Some PolyDPyr-/PolyDCbz-Magnetite NCs	320
10.6	Typical Experimental Procedures for NC Fabrication and NC-Supported DNA Hybridizations	320
10.6.1	Typical Optimized Procedures for NC Fabrication Including Magnetite Preparation	320
10.6.1.1	Magnetite Preparation Using the Oxidative Hydrolysis of Iron(II) Sulfate in an Alkaline KOH Medium	320
10.6.1.2	PolyDPyr-Magnetite Nanocomposites	322
10.6.1.3	PolyDCbz-Magnetite Nanocomposites	322
10.6.2	Covalent Attachment of an Aminated NH_2 -5'-modified DNA Probe. Hybridization Experiments onto PolyDPyr-/PolyDCbz-Magnetite NCs. Typical Experimental Procedures	323
10.6.2.1	Specific Reagents, Buffers and Washing/Assay Solutions	324
10.6.3	Quasi-covalent Attachment of a Biotin-5'-modified DNA Probe and DNA Hybridization Experiments onto Streptavidin-modified PolyDPyr-(5a)/PolyDCbz(5b)-magnetite NCs. Typical Experimental Procedures	324
10.7	Conclusions and Research Outlook	325
	Acknowledgments	325
	References	326
11	Gelatin Nanoparticles and Their Biofunctionalization	330
	<i>Sushma Kommareddy, Dinesh B. Shenoy, and Mansoor M. Arjiji</i>	
11.1	Introduction	330
11.2	Gelatin and Gelatin Derivatives	331
11.2.1	Gelatin	331
11.2.2	Chemical Modification of Gelatin	332
11.2.2.1	PEGylation	333
11.2.2.2	Thiolation	335
11.2.2.3	Other Conjugates of Gelatin	335
11.3	Nanoparticulate Carriers of Gelatin and Gelatin Derivatives	337
11.3.1	Desolvation	337
11.3.1.1	Desolvation Using Ethanol	338
11.3.1.2	Two-step Desolvation	338
11.3.2	Coacervation	338

XII | *Contents*

11.3.3	Nano-encapsulation by Water-in-oil Emulsion Method	339
11.4	Characterization of Gelatin and Modified Gelatin Nanoparticles	340
11.5	Loading and Release of Payload from Gelatin Nanoparticles	342
11.6	Biocompatibility Studies	343
11.7	Applications of Gelatin and Modified Gelatin Nanoparticles	344
11.8	Conclusions	347
	References	348
	Index	353