

Contents of Volumes 228, 230-262

VOLUME 228

AQUEOUS TWO-PHASE SYSTEMS

Harry Walter and Göte Johansson

Section I. General Methodology and Apparatus

1. Phase Diagrams	PER-ÅKE ALBERTSSON AND FOLKE TIERNELD	3
2. Preparation and Analysis of Two-Phase Systems	DONALD E. BROOKS AND RAYMOND NORRIS-JONES	14
3. Partitioning Procedures and Technique: Small Molecules and Macromolecules	GÖTE JOHANSSON	28
4. Partitioning Procedures and Techniques: Cells, Organelles, and Membranes	HARRY WALTER AND CHRISTER LARSSON	42
5. Uses of Poly(ethylene Glycol) with Charged or Hydrophobic Groups	GÖTE JOHANSSON	64
6. Systems Containing Organic Solvents	GÖTE JOHANSSON AND MONICA JOELSSON	74
7. Chiral Resolution	PER-ÅKE ALBERTSSON	84
8. Thin-Layer Countercurrent Distribution and Centrifugal Countercurrent Distribution Apparatus	HANS-ERIK ÅKERLUND AND PER-ÅKE ALBERTSSON	87
9. Columns Using Aqueous Two-Phase Systems	WERNER MÜLLER	100
10. Magnetically Enhanced Phase Separation	PER-OLOF LARSSON	112

Section II. Partitioning and Affinity Partitioning of Macromolecules

A. Proteins

11. Affinity Extraction with Dye Ligands	GERHARD KOPPERSCHLÄGER	121
12. Purification of Lactate Dehydrogenase from Pig Muscle by Affinity Partitioning	MONICA JOELSSON AND FOLKE TIERNELD	136
13. Phosphofructokinase from Baker's Yeast	GERHARD KOPPERSCHLÄGER	144
14. Partitioning of Blood Proteins Using Immobilized Dyes	GERD BIRKENMEIER	154
15. Metal Affinity Partitioning	BONG H. CHUNG, DARWIN BAILEY, AND FRANCES H. ARNOLD	167
16. Starch Synthase from Potato Tubers	ANDREAS BLENNOW	179
17. Isolation of Integral Membrane Proteins by Phase Partitioning with Triton X-114	JOHN S. BRUSCA AND JUSTIN D. RADOLF	182

B. Nucleic Acids

18. Separation of Proteins and Nucleic Acids	WERNER MÜLLER	193
19. Purification of Nucleic Acid-Protein Complexes	GILLIAN E. FRANCIS AND DEREK FISHER	206

Section III. Studies of Protein Interactions, Molecular Structure, Charge, Hydrophobicity, and Conformational Changes

- | | | |
|---|---|-----|
| 20. Cross-Partitioning: Determination of Isoelectric Point by Partitioning | HARRY WALTER AND DANIEL FORCINITI | 223 |
| 21. Charge Determination by Partitioning | GÖTE JOHANSSON | 234 |
| 22. Protein-Protein and Protein-Ligand Interactions | SUSANNE LUNDBERG AND LARS BACKMAN | 241 |
| 23. Estimation of Surface Hydrophobicity of Proteins by Partitioning | VITHALDAS P. SHANBHAG | 254 |
| 24. Detection of Conformational Changes in Proteins by Probing with Poly(ethylene Glycol)-Bound Ligands | GERD BIRKENMEIER AND GERHARD KOPPERSCHLÄGER | 264 |
| 25. Analysis of Structural Changes in Steroid Receptors by Partitioning | JEFFREY C. HANSEN | 276 |
| 26. Use of Partitioning in Predicting Mild Hydrophobic Interaction Chromatography Behavior | PATRICK HUBERT | 287 |

Section IV. Partitioning and Affinity Partitioning of Particulates

A. Cells

Separation and Fractionation of Cell Populations and Obtaining Information on Their Surface Properties

- | | | |
|--|-------------------------------------|-----|
| 27. Analytical Applications of Partitioning: Detection of Differences or Changes in Surface Properties of Mammalian Cell Populations | HARRY WALTER | 299 |
| 28. Cell-Cell Affinity | HARRY WALTER | 320 |
| 29. Testing for Charge and Hydrophobicity Correlates in Cell-Cell Adhesion | KARL-ERIC I. MAGNUSSON | 326 |
| 30. Surface Changes during Growth and Development of Slime Molds | PAUL T. SHARPE | 334 |
| 31. Separation of Pollen from <i>Nicotiana tabacum</i> | BOEL MÄRTENSSON AND SUSANNE WIDELL | 344 |
| 32. Classification of <i>Penicillium</i> Fungi by Cross-Partition Analysis | GÖRAN BLOMQUIST AND GUNNAR B. STRÖM | 354 |

Affinity Partitioning

- | | | |
|---|---|-----|
| 33. Charge-Directed Affinity Partitioning of Cells | JESUS MENDIETA | 363 |
| 34. Factors in the Affinity Extraction of Red Blood Cells Using Poly(ethylene Glycol)-Metal Chelate | GERD BIRKENMEIER, HARRY WALTER, AND KIM E. WIDEN | 368 |
| 35. Use of Poly(ethylene Glycol)-Modified Antibody in Cell Extraction | LAUREL J. KARR, DONNA L. DONNELLY, ANTONI KOZLOWSKI, AND J. MILTON HARRIS | 377 |
| 36. Use of Polyacrylamide-Derivatized Antibody in Dextran-Poly(ethylene Glycol) Systems | DONALD E. BROOKS AND S. JILL STOCKS | 390 |
| 37. Bioextraction of Low Abundance Cells by Affinity Partitioning | CRISTINA DELGADO, GILLIAN E. FRANCIS, AND DEREK FISHER | 395 |

B. Organelles: Separation and Subfractionation

38. Preparation of Synaptosomes and Mitochondria from Mammalian Brain
M. J. LÓPEZ-PÉREZ 403
39. Isolation of All Major Organelles and Membranous Cell Components from Single
Homogenate of Green Leaves D. JAMES MORRÉ AND BERTIL ANDERSSON 412
40. Isolation of Highly Purified Intact Chloroplasts and of Multiorganelle Complexes
Containing Chloroplasts CHRISTER LARSSON 419
41. Purification and Characterization of Plant Mitochondria and Submitochondrial
Particles PER GARDESTRÖM, PATRICE X. PETIT, AND IAN M. MØLLER 424

C. Membranes

Separation and Subfractionation

42. Isolation and Identification of Plasma Membrane Populations
MICHAEL E. BRADLEY, ROSS W. LAMBERT, AND AUSTIN K. MIRCHEFF 432
43. Plasma and Internal Membranes from Cultured Mammalian Cells
D. JAMES MORRÉ, TIMOTHY REUST, AND DOROTHY M. MORRÉ 448
44. Isolation of Highly Purified Plant Plasma Membranes and Separation of Inside-
Out and Right-Side-Out Vesicles
CHRISTER LARSSON, MARIANNE SOMMARIN, AND SUSANNE WIDELL 451
45. Fractionation of Thylakoid Membrane
PER-ÅKE ALBERTSSON, EVA ANDREASSON, HREINN STEFÁNSSON,
AND LOUIE WOLLENBERGER 469
46. Isolation of Smooth Endoplasmic Reticulum and Tonoplast from Etiolated Mung
Bean Hypocotyls SHIZUO YOSHIDA 482

Affinity Partitioning

47. Rat Liver Plasma Membranes ANDERS PERSSON AND BENGT JERGIL 489
48. Synaptic Membranes GÖTE JOHANSSON 496

Membrane Domain Analysis

49. Domain Structure of Biological Membranes Obtained by Fragmentation and Sepa-
ration Analysis PER-ÅKE ALBERTSSON 503
50. Relative Proximity of Domains in Plasma Membrane and Smooth Endoplasmic
Reticulum from Rat Liver J. PETER GIEROW 512

Section V. Aqueous Phase Separations in Biological Systems

51. Lens Cytoplasmic Protein Solutions: Analysis of a Biologically Occurring Aqueous
Phase Separation JOHN I. CLARK 525

Section VI. Aqueous Two-Phase Systems in Large-Scale Processes:
Biotechnology

A. Extractive Bioconversions

52. Cultivation of *Trichoderma reesei* and Production of Cellulolytic Enzymes
FOLKE TIERNELD AND INGRID PERSSON 541

53. Enzyme-Catalyzed Hydrolysis and Recycling in Cellulose Bioconversion
FOLKE TIERNELD 549
54. Biotransformation of Hydrocortisone into Prednisolone
RAJNI KAUL AND BO MATTIASSON 559

B. Proteins: Downstream Processing

55. Recovery of Proteins and Phase-Forming Chemicals
GÖTE JOHANSSON 569
56. Enzyme Recovery by Continuous Crosscurrent Extraction
NEOPHYTOS PAPAMICHAEL AND HELMUT HUSTEDT 573
57. Purification of Oxynitrilases from Plants
RAINER WOKER, JÖRG VERNAU, AND MARIA-REGINA KULA 584
58. Extractive Purification of Aspartase from *Escherichia coli* K12
JENS PAULSEN AND HELMUT HUSTEDT 590
59. Large-Scale Purification of Formate Dehydrogenase
ARNO CORDES AND MARIA-REGINA KULA 600
60. Extraction and Purification of Leucine Dehydrogenase from *Bacillus cereus*
HORST SCHÜTTE, DIETER SCHUSTOLLA, AND HELMUT HUSTEDT 608
61. Purification of Proteins from Cell Culture Supernatant Solutions
ULRICH MENGE 617

C. Design of Proteins for Enhanced Extraction

62. Uses of Fusions of β -Galactosidase and Peptides to Proteins
KRISTINA KÖHLER AND ANDRES VEIDE 627

D. Other Applications of Aqueous Phases in Biotechnology

63. Concentration of Biomaterials: Virus Concentration and Viral Protein Isolation
LENA HAMMAR 640
64. Extraction of Chromatophores
ANNA-LISA SMEDS AND SVEN-OLOF ENFORS 658
65. Extraction of Proteins from Sources Containing Tannins and Anionic Mucilages
HANS VILTER 665
66. Purification of Water-Based Cutting Fluids
GUNNAR B. STRÖM, BJÖRN MÄLARSTIG, AND GÖRAN BLOMQUIST 672

VOLUME 230

GUIDE TO TECHNIQUES IN GLYCOBIOLOGY

William J. Lennarz and Gerald W. Hart

1. Carbohydrate Composition Analysis of Glycoconjugates by Gas-Liquid Chromatography/Mass Spectrometry
ROBERTA K. MERKLE AND IZABELLA POPPE 1
2. Metabolic Radiolabeling of Glycoconjugates
AJIT VARKI 16

3. Nonmetabolic Radiolabeling and Tagging of Glycoconjugates	CARL G. GAHMBERG AND MARTTI TOLVANEN	32
4. Enzymatic Deglycosylation of Asparagine-Linked Glycans: Purification, Properties, and Specificity of Oligosaccharide-Cleaving Enzymes from <i>Flavobacterium meningosepticum</i>	ANTHONY L. TARENTINO AND THOMAS H. PLUMMER, JR.	44
5. Release of Oligosaccharides from Glycoproteins by Hydrazinolysis	T. P. PATEL AND R. B. PAREKH	57
6. Use of Lectins in Analysis of Glycoconjugates	RICHARD D. CUMMINGS	66
7. Saccharide Linkage Analysis Using Methylation and Other Techniques	RUDOLF GEYER AND HILDEGARD GEYER	86
8. Mass Spectrometry of Carbohydrate-Containing Bipolymers	ANNE DELL, ANDREW J. REASON, KAY-HOOI KHOO, MARIA PANICO, ROY A. McDOWELL, AND HOWARD R. MORRIS	108
9. ^1H Nuclear Magnetic Resonance Spectroscopy of Carbohydrate Chains of Glycoproteins	HERMAN VAN HALBEEK	132
10. Determination of Sialic Acids	GERD REUTER AND ROLAND SCHAUER	168
11. Size Fractionation of Oligosaccharides	AKIRA KOBATA	200
12. High-pH Anion-Exchange Chromatography of Glycoprotein-Derived Carbohydrates	MARK R. HARDY AND R. REID TOWNSEND	208
13. High-Performance Liquid Chromatography of Pyridylaminated Saccharides	SUMIHIRO HASE	225
14. High-Performance Liquid Chromatography of Oligosaccharides	JACQUES U. BAENZIGER	237
15. High-Resolution Polyacrylamide Gel Electrophoresis of Fluorophore-Labeled Reducing Saccharides	PETER JACKSON	250
16. Capillary Electrophoresis of Oligosaccharides	ROBERT J. LINHARDT	265
17. Glycosidases in Structural Analysis	GARY S. JACOB AND PETER SCUDDER	280
18. Glycosyltransferases in Glycobiology	MONICA M. PALCIC	300
19. Glycosidase Inhibitors in Study of Glycoconjugates	GUR P. KAUSHAL AND ALAN D. ELBEIN	316
20. Synthesis and Uses of Azido-Substituted Nucleoside Diphosphate Sugar Photoaffinity Analogs	ANNA RADOMINSKA AND RICHARD R. DRAKE	330
21. Glycoform Analysis of Glycoproteins	R. B. PAREKH	340
22. Isolation of Glycosphingolipids	RONALD L. SCHNAAR	348
23. Thin-Layer Chromatography of Glycosphingolipids	RONALD L. SCHNAAR AND LEILA K. NEEDHAM	371
24. Isolation and Characterization of Proteoglycans	VINCENT C. HASCALL, ANTHONY CALABRO, RONALD J. MIDURA, AND MASAKI YANAGISHITA	390
25. Structural Analysis of Glycosylphosphatidylinositol Anchors	ANANT K. MENON	418
26. Detection of O-Linked <i>N</i> -Acetylglucosamine (<i>O</i> -GlcNAc) on Cytoplasmic and Nuclear Proteins	ELIZABETH P. ROQUEMORE, TEH-YING CHOU, AND GERALD W. HART	443
27. Identification of Polysialic Acids in Glycoconjugates	JEAN YE, KEN KITAJIMA, YASUO INOUE, SADAKO INOUE, AND FREDERIC A. TROY II	460
28. Neoglycolipids: Probes of Oligosaccharide Structure, Antigenicity, and Function	TEN FEIZI, MARK S. STOLL, CHUN-TING YUEN, WENGANG CHAI, AND ALEXANDER M. LAWSON	484

VOLUME 231

HEMOGLOBINS (PART B: BIOCHEMICAL AND ANALYTICAL METHODS)

Johannes Everse, Kim D. Vandegriff, and Robert M. Winslow

Section I. Preparation, Purification, and Analysis of Human Hemoglobin

1. Pilot-Scale Preparation of Hemoglobin Solutions
ROBERT M. WINSLOW AND KEITH W. CHAPMAN 3
2. Detection of Red Cell Membrane Components in Human Hemoglobin Preparations
BERNARD J.-M. THEVENIN AND STEPHEN B. SHOHEET 17
3. Detection of Globin Chains by Reversed-Phase High-Performance Liquid Chromatography
BRUNO MASALA AND LAURA MANCA 21
4. Structural Characterization of Hemoglobin Variants Using Capillary Electrophoresis and Fast Atom Bombardment Mass Spectrometry
PASQUALE FERRANTI, ANTONIO MALORNI, AND PIETRO PUCCI 45
5. Purification of Glycated Hemoglobin MARGO PANUSH COHEN AND VAN-YU WU 65
6. Measurement of Endotoxin Levels in Hemoglobin Preparations
ROBERT I. ROTH AND JACK LEVIN 75

Section II. Preparation and Characterization of Nonhuman Hemoglobins

7. Preparation and Characterization of Insect Hemoglobins from *Chironomus thummi thummi*
HANS HEINRICH RUF, ANDREAS G. ALTEMÜLLER, AND KLAUS GERSONDE 95
8. Preparation and Characterization of Invertebrate Globin Complexes
SERGE N. VINOGRADOV AND PAWAN K. SHARMA 112
9. Fish Hemoglobins
ALICE DEYOUNG, LAURA D. KWIATKOWSKI, AND ROBERT W. NOBLE 124
10. Conformational and Functional Characteristics of Bovine Hemoglobin
CLARA FRONTICELLI AND ENRICO BUCCI 150

Section III. Hemoglobin Hybrids and Model Hemes

11. Synthetic Hemes
EISHUN TSUCHIDA AND TERUYUKI KOMATSU 167
12. Semisynthesis of Hemoglobin
RAJENDRA PRASAD ROY AND A. SEETHARAMA ACHARYA 194
13. Preparation of Mixed Metal Hybrids
ANTONIO TSUNESHIGE AND TAKASHI YONETANI 215

Section IV. Chemically Modified Hemoglobins

14. Preparation of Hemoglobin Derivatives Selectively or Randomly Modified at Amino Groups
JAMES M. MANNING 225
15. Amidation of Basic Carboxyl Groups of Hemoglobin
M. JANARDHAN RAO AND A. SEETHARAMA ACHARYA 246

16. Bis(pyridoxal) Polyphosphates as Specific Intramolecular Cross-Linking Agents for Hemoglobin	RUTH E. BENESCH	267
17. Preparation of Intramolecularly Cross-Linked Hemoglobins	ROXANNE Y. WALDER, MARK E. ANDRACKI, AND JOSEPH A. WALDER	274
18. Derivatives of Hemoglobin Prepared by Reaction with Aryl or Alkyl Isothiocyanates	DOUGLAS L. CURRELL	281
19. Hemoglobin Polymerization	SHIRLEY L. MACDONALD AND DUNCAN S. PEPPER	287
20. Preparation of Conjugated Hemoglobins	HONG XUE AND J. TZE-FEI WONG	308
21. Structural Characterization of Modified Hemoglobins	RICHARD T. JONES	322

Section V. Recombinant Hemoglobin

22. Production of Human Hemoglobin in <i>Escherichia coli</i> Using Cleavable Fusion Protein Expression Vector	TIMM-H. JESSEN, NOBORU H. KOMIYAMA, JEREMY TAME, JOSÉE PAGNIER, DANIEL SHIH, BEN LUISI, GIULIO FERMI, AND KIYOSHI NAGAI	347
23. Expression of Recombinant Human Hemoglobin in <i>Escherichia coli</i>	DOUGLAS LOOKER, ANTONY J. MATHEWS, JUSTIN O. NEWAY, AND GARY L. STETLER	364
24. Production of Recombinant Human Hemoglobin A in <i>Saccharomyces cerevisiae</i>	JILL E. OGDEN, ROY HARRIS, AND MICHAEL T. WILSON	374
25. Purification and Characterization of Recombinant Human Sickle Hemoglobin Expressed in Yeast	JOSE JAVIER MARTIN DE LLANO, OLAF SCHNEEWIND, GARY L. STETLER, AND JAMES M. MANNING	390
26. Preparation of Recombinant Hemoglobin in Transgenic Mice	MICHAEL P. REILLY, STEVEN L. MCCUNE, THOMAS M. RYAN, TIM M. TOWNES, MAKOTO KATSUMATA, AND TOSHIO ASAKURA	403
27. Transgenic Swine as Recombinant Production System for Human Hemoglobin	JOHN S. LOGAN AND MICHAEL J. MARTIN	435

Section VI. Hemoglobin Stability and Degradation

28. Detection, Formation, and Relevance of Hemichromes and Hemochromes	JOSEPH M. RIFKIND, OMOTFE ABUGO, ABRAHAM LEVY, AND JANE HEIM	449
29. Measuring Relative Rates of Hemoglobin Oxidation and Denaturation	VICTOR W. MACDONALD	480
30. Hydrogen Peroxide-Mediated Ferrylhemoglobin Generation <i>in Vitro</i> and in Red Blood Cells	CECILIA GIULIVI AND KELVIN J. A. DAVIES	490
31. The Stability of the Heme-Globin Linkage: Measurement of Heme Exchange	RUTH E. BENESCH	496
32. Release of Iron from Hemoglobin	S. SCOTT PANTER	502
33. Thermal Denaturation Procedures for Hemoglobin	KENNETH W. OLSEN	514
34. Photochemical Reduction of Methemoglobin and Methemoglobin Derivatives	JOHANNES EVERSE	524
35. Regeneration of Functional Hemoglobin from Partially Oxidized Hemoglobin in the Presence of Molecular Hydrogen and a Multicomponent Redox Catalyst	EVELYN L. MCGOWN, MAZHAR KHAN, AND KILIAN DILL	537

Section VII. Enzymatic Reactions Catalyzed by Hemoglobin

36. Peroxidative Activities of Hemoglobin and Hemoglobin Derivatives
JOHANNES EVERSE, MARIA C. JOHNSON, AND MARIO A. MARINI 547
37. Cyclooxygenase Activity of Hemoglobin
LUCILLA ZILLETI, MARIO CIUFFI, SERGIO FRANCHI-MICHELI, FABIO FUSI,
GRAZIA GENTILINI, GLORIANO MONETI, MASSIMO VALOTI,
AND GIAN PIETRO SGARAGLI 562
38. Hydroxylation and Dealkylation Reactions Catalyzed by Hemoglobin
J. J. MIEYAL AND D. W. STARKE 573
39. Oxidation of Olefins Catalyzed by Hemoglobin
GIORGIO BELVEDERE AND MICHELE SAMAJA 598

Section VIII. Xenobiotic Adducts of Human Hemoglobin

40. Quantitative Analysis of Hemoglobin-Xenobiotic Adducts
STEVEN R. TANNENBAUM AND PAUL L. SKIPPER 625
41. Quantitative Mass Spectrometry of Hemoglobin Adducts JOHN S. WISHNOK 632
42. Aromatic Amine-Hemoglobin Adducts PAUL L. SKIPPER AND W. G. STILLWELL 643
43. Epoxide Adducts to N-Terminal Valine of Hemoglobin
MARGARETA TÖRNOVIST 650
44. Tobacco-Specific Nitrosamine-Hemoglobin Adducts
STEPHEN S. HECHT, STEVEN G. CARMELLA, AND SHARON E. MURPHY 657
45. Benzo[a]pyrene Diol Epoxide and Related Polynuclear Aromatic Hydrocarbon
Adducts of Hemoglobin KOLI TAGHIZADEH AND PAUL L. SKIPPER 668
46. Analysis of Hemoglobin Adducts by Fluorescence Spectroscopy
BILLY W. DAY AND KULDIP SINGH 674

Addendum

- Addendum to Article [11] EISHUN TSUCHIDA AND TERUYUKI KOMATSU 685

VOLUME 232

HEMOGLOBINS (PART C: BIOPHYSICAL METHODS)

Johannes Everse, Kim D. Vandegriff, and Robert M. Winslow

Section I. Molecular Structure and Dynamics

1. Acid-Induced Folding of Heme Proteins YUJI GOTO AND ANTHONY L. FINK 3
2. X-Ray Crystallography of Partially Ligated Structures ROBERT LIDDINGTON 15
3. Structure and Energy Change in Hemoglobin by Hydrogen Exchange Labeling
S. WALTER ENGLANDER AND JOAN J. ENGLANDER 26
4. High-Pressure Fluorescence Methods for Observing Subunit Dissociation in Hemoglobin
SERGE PIN AND CATHERINE A. ROYER 42
5. Optical Measurements of Quaternary Structural Changes in Hemoglobin
ANDREA BELLELLI AND MAURIZIO BRUNORI 56

- | | |
|---|----|
| 6. Allosteric Equilibrium Measurements with Hemoglobin Valency Hybrids | |
| MICHAEL C. MARDEN, JEAN KISTER, AND CLAUDE POYART | 71 |
| 7. Electron-Transfer Reactions of Hemoglobin with Small Molecules: A Potential Probe of Conformational Dynamics | |
| GEORGE MCLENDON AND JEHUDAH FEITELSON | 86 |

Section II. Spectroscopy

- | | |
|--|-------------------------|
| 8. Proton Nuclear Magnetic Resonance Studies of Hemoglobin | |
| CHIEN HO AND JANICE R. PERUSSI | 97 |
| 9. Infrared Methods for Study of Hemoglobin Reactions and Structures | |
| AICHUN DONG AND WINSLOW S. CAUGHEY | 139 |
| 10. Picosecond Infrared Spectroscopy of Hemoglobin and Myoglobin | |
| ROBERT H. AUSTIN AND LEWIS J. ROTHBERG | 176 |
| 11. Time-Resolved Resonance Raman Spectroscopy as Probe of Structure, Dynamics, and Reactivity in Hemoglobin | |
| JOEL M. FRIEDMAN | 205 |
| 12. Front-Face Fluorescence Spectroscopy of Hemoglobins | RHODA ELISON HIRSCH 231 |
| 13. Stationary and Time-Resolved Circular Dichroism of Hemoglobins | |
| CHRISTIAN ZENTZ, SERGE PIN, AND BERNARD ALPERT | 247 |
| 14. X-Ray Absorption Spectroscopy of Hemoglobin | |
| SERGE PIN, BERNARD ALPERT, AGOSTINA CONGIU-CASTELLANO, STEFANO DELLA LONGA, AND ANTONIO BIANCONI | 266 |
| 15. Modulated Excitation Spectroscopy in Hemoglobin | FRANK A. FERRONE 292 |
| 16. Picosecond Phase Grating Spectroscopy: Applications to Bioenergetics and Protein Dynamics | |
| JOHN DEAK, LYNN RICHARD, MARCO PEREIRA, HUI-LING CHUI, AND R. J. DWAYNE MILLER | 322 |

Section III. Ligand Binding

- | | |
|--|-----|
| 17. Assignment of Rate Constants for O ₂ and CO Binding to α and β Subunits within R- and T-State Human Hemoglobin | |
| ANTHONY J. MATHEWS AND JOHN S. OLSON | 363 |
| 18. Ligand Binding and Conformational Changes Measured by Time-Resolved Absorption Spectroscopy | |
| JAMES HOFRICHTER, ANJUM ANSARI, COLLEEN M. JONES, ROBERT M. DEUTSCH, JOSEPH H. SOMMER, AND ERIC R. HENRY | 387 |
| 19. Femtosecond Measurements of Geminate Recombination in Heme Proteins | |
| JEAN-LOUIS MARTIN AND MARTEN H. VOS | 416 |
| 20. Double Mixing Methods for Kinetic Studies of Ligand Binding in Partially Liganded Intermediates of Hemoglobin | |
| VIJAY S. SHARMA | 430 |
| 21. Hemoglobin-Liganded Intermediates | |
| MICHELE PERRELLA AND LUIGI ROSSI-BERNARDI | 445 |
| 22. Hemoglobin-Oxygen Equilibrium Binding: Rapid-Scanning Spectrophotometry and Singular Value Decomposition | |
| KIM D. VANDEGRIFF AND RICHARD I. SHRAGER | 460 |
| 23. Oxygen Equilibrium Curve of Concentrated Hemoglobin | |
| ROBERT M. WINSLOW, ALISON MURRAY, AND CARTER C. GIBSON | 486 |
| 24. Bezafibrate Derivatives as Potent Effectors of Hemoglobin | |
| CLAUDE POYART, MICHAEL C. MARDEN, AND JEAN KISTER | 496 |

Section IV. Mathematical Analysis and Modeling

25. Simulation of Hemoglobin Kinetics Using Finite Element Numerical Methods	ROBERT L. BERGER, NORMAN DAVIDS, AND MICHELE PERRELLA	517
26. Adair Fitting to Oxygen Equilibrium Curves of Hemoglobin	KIYOHITO IMAI	559
27. Weighted Nonlinear Regression Analysis of Highly Cooperative Oxygen Equilibrium Curves	MICHAEL L. DOYLE, DAVID W. MYERS, GARY K. ACKERS, AND RICHARD I. SHRAGER	576
28. Dimer-Tetramer Equilibrium in Adair Fitting	MICHAEL L. JOHNSON	597
29. Effects of Wavelength on Fitting Adair Constants for Binding of Oxygen to Human Hemoglobin	LAWRENCE J. PARKHURST, TODD M. LARSEN, AND HORNG-YUH LEE	606
30. Adair Equation: Rederiving Oxygenation Parameters	EDWARD C. DELAND	632
31. Linkage Thermodynamics	ENRICO DI CERA	655

VOLUME 233

OXYGEN RADICALS IN BIOLOGICAL SYSTEMS (PART C)

Lester Packer

Section I. Generation, Detection, and Characterization of Oxygen Radicals: Chemistry, Biochemistry, and Intermediate States of Reduction

1. Pulse Radiolysis	CLEMENS VON SONNTAG AND HEINZ-PETER SCHUCHMANN	3
2. Pulse Radiolysis for Investigation of Nitric Oxide-Related Reactions	MANFRED SARAN AND WOLF BORS	20
3. Sulfhydryl Free Radical Formation Enzymatically by Sonolysis, by Radiolysis, and Thermally: Vitamin A, Curcumin, Muconic Acid, and Related Conjugated Olefins as References	MASSIMO D'AQUINO, CHRISTOPHER BULLION, MRIDULA CHOPRA, DAMAYANTI DEVI, SUNITIBALA DEVI, CHRISTINA DUNSTER, GARY JAMES, ERIKA KOMURO, SUBHAS KUNDU, ETSUO NIKI, FARAH RAZA, FORBES ROBERTSON, JITEN SHARMA, AND ROBIN WILLSON	34
4. Suppression of Hydroxyl Radical Reactions in Biological Systems: Considerations Based on Competition Kinetics	CLEMENS VON SONNTAG AND HEINZ-PETER SCHUCHMANN	47
5. Deoxyribose Assay for Detecting Hydroxyl Radicals	OKEZIE I. ARUOMA	57
6. Detection of Hydroxyl Radicals by Aromatic Hydroxylation	HARPARKASH KAUR AND BARRY HALLIWELL	67
7. Measurement of Iron and Copper in Biological Systems: Bleomycin and Copper-Phenanthroline Assays	P. J. EVANS AND B. HALLIWELL	82
8. Oxygen Radicals Generation and DNA Scission by Anticancer and Synthetic Quinones	LEV M. WEINER	92
9. Spin Trapping of Hydroxyl Radicals in Biological Systems	GERALD M. ROSEN, SOVITI POU, BRADLEY E. BRITIGAN, AND MYRON S. COHEN	105
10. <i>In Vivo</i> Detection of Radical Adducts by Electron Spin Resonance	RONALD P. MASON AND KATHRYN T. KNECHT	112

11. Spin Trapping of Alcohol-Derived Radicals in Microsomes and Reconstituted Systems by Electron Spin Resonance
EMANUELE ALBANO, ALDO TOMASI, AND MAGNUS INGELMAN-SUNDBERG 117
12. *In Vivo* Visualization of Oxygen Radical-Dependent Photoemission
MASAHARU TSUCHIYA, MAKOTO SUEMATSU, AND HIDEKAZU SUZUKI 128
13. *In Vivo* Electron Paramagnetic Resonance Imaging of Skin
JURGEN FÜCHS, NORBERT GROTH, THOMAS HERRLING, AND LESTER PACKER 140
14. Electron Spin Resonance Assay of Ascorbate Free Radicals *in Vivo*
AKITANE MORI, XIAOYAN WANG, AND JIANKANG LIU 149
15. Measurement of Superoxide Reaction by Chemiluminescence
NAOHIRO GOTOH AND ETSUO NIKI 154

Section II. Isolation, Characterization, and Assay of Enzymes or Substances Involved in Formation or Removal of Oxygen Radicals

16. Mass Spectrometry of Prostanoids: F₂-Isoprostanes Produced by Non-Cyclooxygenase Free Radical-Catalyzed Mechanism
JASON D. MORROW AND L. JACKSON ROBERTS II 163
17. Quantifying Aldehydes and Distinguishing Aldehydic Product Profiles from Autoxidation and Ozonation of Unsaturated Fatty Acids
RAFAEL CUETO, GIUSEPPE L. SQUADRITO, AND WILLIAM A. PRYOR 174
18. Ferrous Ion Oxidation in Presence of Ferric Ion Indicator Xylenol Orange for Measurement of Hydroperoxides
SIMON P. WOLFF 182
19. Ferrylmyoglobin: Formation and Chemical Reactivity toward Electron-Donating Compounds
CECILIA GIULIVI AND ENRIQUE CADENAS 189
20. Enzymatic and Immunological Measurements of Soluble and Membrane-Bound Phospholipid-Hydroperoxide Glutathione Peroxidase
ANTONELLA ROVERI, MATILDE MAIORINO, AND FULVIO URSINI 202
21. Targeting Superoxide Dismutase by Gene and Protein Engineering
MASAYASU INOUE 212
22. Assays of Plasma and Membrane NADPH Oxidase
OWEN T. G. JONES AND JOHN T. HANCOCK 222
23. Oxidative Chemistry of Peroxynitrite
JOSEPH S. BECKMAN, JUN CHEN, HARRY ISCHIROPOULOS, AND JOHN P. CROW 229
24. Nitric Oxide Assay Using Hemoglobin Method
MICHAEL E. MURPHY AND EIKE NOACK 240
25. Nitric-Oxide Synthase Assays
JOAN M. HEVEL AND MICHAEL A. MARLETTA 250
26. Isoforms of Nitric-Oxide Synthase: Purification and Regulation
ULRICH FÖRSTERMANN, JENNIFER S. POLLOCK, W. ROSS TRACEY, AND MASAKI NAKANE 258
27. Purification, Cloning, and Expression of Nitric-Oxide Synthase
CHARLES J. LOWENSTEIN AND SOLOMON H. SNYDER 264

Section III. Methods for Assessing Molecular, Cell, and Tissue Damage: Assays and Repair of Oxidative Damage

28. Lipid Peroxidation-Induced Membrane Structural Alterations
GUSBERT VAN GINKEL AND ALEX SEVANI 273

29. Iodometric Determination of Hydroperoxides in Lipids and Proteins	WENDY JESSUP, ROGER T. DEAN, AND JANUSZ M. GEBICKI	289
30. Detection of Conjugated Dienes by Second Derivative Ultraviolet Spectrophotometry	FRANCESCO P. CORONGIU AND SEBASTIANO BANNI	303
31. Ultraviolet and Infrared Methods for Analysis of Fatty Acyl Esters in Cellular Systems	ODILE SERGENT, PIERRE CILLARD, AND JOSIANE CILLARD	310
32. High-Performance Liquid Chromatography-Based Thiobarbituric Acid Tests	SUSANNA CHIRICO	314
33. Chemiluminescence-Based High-Performance Liquid Chromatography Assay of Lipid Hydroperoxides	YORIIHIRO YAMAMOTO	319
34. Determination of Phospholipid Hydroperoxides Using Luminol Chemiluminescence-High-Performance Liquid Chromatography	TERUO MIYAZAWA, KENSHIRO FUJIMOTO, TOSHIHIDE SUZUKI, AND KEIICHI YASUDA	324
35. Assay of Cholesterol Autoxidation	G. A. S. ANSARI AND LELAND L. SMITH	332
36. Assay of Fluorescent Lipid Peroxidation Products	HIROYUKI SHIMASAKI	338
37. Carbonyl Assays for Determination of Oxidatively Modified Proteins	RODNEY L. LEVINE, JOY A. WILLIAMS, EARL R. STADTMAN, AND EMILY SHACTER	346
38. Oxidative Damage to Proteins: Spectrophotometric Method for Carbonyl Assay	ABRAHAM Z. REZNICK AND LESTER PACKER	357
39. Dityrosine: A Marker for Oxidatively Modified Proteins and Selective Proteolysis	CECILIA GIULIVI AND KELVIN J. A. DAVIES	363
40. Quantitation of 4-Hydroxynonenal Protein Adducts	KOJI UCHIDA AND EARL R. STADTMAN	371
41. Measurement of Protein Thiol Groups and Glutathione in Plasma	MIAO-LIN HU	380
42. Protein S-Thiolation and Dethiolation	JAMES A. THOMAS, YUH-CHERNG CHAI, AND CHE-HUN JUNG	385
43. Purification of Vicinal Dithiol-Containing Proteins by Arsenical-Based Affinity Chromatography	EDNA KALEF AND CARLOS GITLER	395
44. Labeling of Protein Vicinal Dithiols: Role of Protein-S ₂ to Protein-(SH) ₂ Conversion in Metabolic Regulation and Oxidative Stress	CARLOS GITLER, MYRIAM MOGYOROS, AND E. KALEF	403
45. Transport of Glutathione Disulfide and Glutathione S-Conjugates in Hepatocyte Plasma Membrane Vesicles	THEODORUS P. M. AKERBOOM AND HELMUT SIES	416
46. Methods to Determine Oxidation of Low-Density Lipoproteins	HERBERT PUHL, GEORG WAEG, AND HERMANN ESTERBAUER	425
47. Quantitative Low-Density Lipoprotein Oxidation by Gamma Radiolysis	D. BONNEFONT-ROUSSELOT, M. GARDÈS-ALBERT, J. DELATTRE, AND C. FERRADINI	441
48. Assaying Low-Density Lipoprotein Oxidation by Laser Light Scattering	BERNARD ARRIO, MATHIEU ARRIO, DOMINIQUE BONNEFONT-ROUSSELOT, JUDITH D. CATUDILOC, AND LESTER PACKER	453
49. Fluorescence Analysis of Lipoprotein Peroxidation	NICOLE DOUSSET, GIANNA FERRETTI, MARINA TAUS, PIERRE VALDIGUIÈ, AND GIOVANNA CURATOLA	459
50. Rapid Isolation of Lipoproteins and Assessment of Their Peroxidation by High-Performance Liquid Chromatography Postcolumn Chemiluminescence	WOLFGANG SATTLER, DETLEF MOHR, AND ROLAND STOCKER	469

51. Apolipoprotein B Protein Oxidation in Low-Density Lipoproteins	
NORIKO NOGUCHI AND ETSUO NIKI	490
52. Luciferin Derivative for Assay of Myeloperoxidase and Dopamine Metabolism	
MINORU NAKANO AND SATOSHI KOGA	495
53. Assays for the Chlorination Activity of Myeloperoxidase	
ANTHONY J. KETTLE AND CHRISTINE C. WINTERBOURN	502
54. Oxidative Stress to Lens Crystallins	
JESSICA JAHNGEN-HODGE, ALLEN TAYLOR, FU SHANG, LI LI HUANG, AND CASILDA MURA	512
55. High-Performance Liquid Chromatography-Electrochemical Detection of Antioxidants in Vertebrate Lens: Glutathione, Tocopherol, and Ascorbate	
KENNETH P. MITTON AND JOHN R. TREVITHICK	523
56. Flow Cytometric Assays of Oxidative Burst Activity in Phagocytes	
GREGOR ROTHE AND GÜNTER VALET	539
57. Adult Respiratory Distress Syndrome: Model Systems Using Isolated Perfused Rabbit Lungs	
WERNER SEEGER, DIETER WALMRATH, FRIEDRICH GRIMMINGER, SIMONE ROSSEAU, HARTWIG SCHÜTTE, HANS-JOACHIM KRÄMER, LEANDER ERMERT, AND LADISLAV KISS	549
58. Animal Models of Chronic Ethanol Toxicity	
CHARLES S. LIEBER AND LEONORE M. DECARLI	585
59. Oxidant-Antioxidant Status in Human Immunodeficiency Virus Infection	
WULF DRÖGE, HANS-PETER ECK, AND SABINE MIHM	594
60. Antioxidant Effectiveness in Ischemia-Reperfusion Tissue Injury	
DIPAK K. DAS AND NILANJANA MAULIK	601
61. Animal Models and Molecular Markers for Cerebral Ischemia-Reperfusion Injury in Brain	
VIJAYALAKSHMI RAVINDRANATH	610
62. Histochemical Methods for Localization of Endothelial Superoxide and Hydrogen Peroxide Generation in Perfused Organs	
CHARLES F. BABBS	619
63. Assessment of Oxidative Stress to Eye in Animal Model for Cataract	
DURGA K. BHUYAN AND KAILASH C. BHUYAN	630
64. Measurement of Reaction Products from Hypochlorous Acid and Unsaturated Lipids	
JEROEN VAN DEN BERG AND CHRISTINE WINTERBOURN	639

VOLUME 234

OXYGEN RADICALS IN BIOLOGICAL SYSTEMS (PART D)

Lester Packer

Section I. Oxidative Damage to DNA and DNA Repair

1. Chemical Determination of Oxidative DNA Damage by Gas Chromatography-Mass Spectrometry	
MIRAL DIZDAROGLU	3
2. Assays of Oxidative DNA Damage Biomarker 8-Oxo-2'-deoxyguanosine and 8-Oxoguanine in Nuclear DNA and Biological Fluids by High-Performance Liquid Chromatography with Electrochemical Detection	
MARK K. SHIGENAGA, ELIAS N. ABOUJAOUDE, QIN CHEN, AND BRUCE N. AMES	16

3. Detection and Characterization of Eukaryotic Enzymes That Recognize Oxidative DNA Damage	KRISTA K. HAMILTON, KEUNMYOUNG LEE, AND PAUL W. DOETSCH	33
4. Localization of Strand Breaks in Plasmid DNA Treated with Reactive Oxygen Species	WOLFGANG A. SCHULZ, MAIK S. W. OBENDORF, AND HELMUT SIES	45
5. Detecting DNA Damage Caused by Iron and Hydrogen Peroxide	YONGZHANG LUO, ERNST S. HENLE, RAJAGOPAL CHATTOPADHYAYA, RUCHENG JIN, AND STUART LINN	51
6. Photochemical Synthesis of 8-Hydroxyguanine Nucleosides	PETER K. WONG AND ROBERT A. FLOYD	59
7. Copper-DNA Adducts	MARK J. BURKITT	66
8. Singlet Oxygen DNA Damage: Chromatographic and Mass Spectrometric Analysis of Damage Products	JEAN CADET, JEAN-LUC RAVANAT, GARRY W. BUCHKO, HELEN C. YEO, AND BRUCE N. AMES	79
9. Alkaline Elution versus Fluorescence Analysis of DNA Unwinding	CHRISTA BAUMSTARK-KHAN	88
10. Purification and Properties of Yeast Redoxendonuclease	LAURA AUGERI, KRISTA K. HAMILTON, AMY M. MARTIN, PAULOS YOHANNES, AND PAUL W. DOETSCH	102
11. Shuttle Vector between Prokaryotes and Eukaryotes for Assaying Singlet Oxygen-Induced DNA Damage and Mutagenicity	CARLOS FREDERICO MARTINS MENCK	115
12. Oxidative DNA Damage: Endonuclease Fingerprinting	BERND EPE AND JUTTA HEGLER	122

Section II. Assay of Stress Genes/Proteins, Effects of Reactive Oxygen Species and Antioxidants on Cell Regulatory Processes

13. Effect of Reactive Oxygen Intermediates and Antioxidants on Proliferation and Function of T Lymphocytes	WULF DRÖGE, SABINE MIHM, MICHAEL BOCKSTETTE, AND STEFFEN ROTH	135
14. Assessing Oxygen Radicals as Mediators in Activation of Inducible Eukaryotic Transcription Factor NF- κ B	RALF SCHRECK AND PATRICK A. BAEUERLE	151
15. Analysis of c-Fos and c-Jun Redox-Dependent DNA Binding Activity	STEVEN XANTHOUDAKIS AND TOM CURRAN	163
16. Assessing Gene Expression during Oxidative Stress	DANA R. CRAWFORD, CHERYL A. EDBAUER-NECHAMEN, CHARLES V. LOWRY, SHARON L. SALMON, YONG K. KIM, JOANNA M. S. DAVIES, AND KELVIN J. A. DAVIES	175
17. OxyR Regulon	GISELA STORZ AND SHOSHY ALTUVIA	217
18. Transient Enhancement of Heme Oxygenase 1 mRNA Accumulation: A Marker of Oxidative Stress to Eukaryotic Cells	REX M. TYRRELL AND SHARMILA BASU-MODAK	224
19. Assays for Regulation of Gap Junctional Communication and Connexin Expression by Carotenoids	JOHN S. BERTRAM AND LI-XIN ZHANG	235
20. One-Day Northern Blotting for Detection of mRNA: NDGA Inhibits the Induction of Mn-SOD mRNA by Agonists of Type 1 TNF Receptor	GRACE H. W. WONG AND DAVID V. GOEDDEL	244

21. Evaluation of Biomolecular Damage by Ozone
CARROLL E. CROSS AND BARRY HALLIWELL 252
22. Ozone Exposure of Cultured Cells and Tissues
BRIAN K. TARKINGTON, TIMOTHY R. DUVALL, AND JEROLD A. LAST 257

Section III. Antioxidant Identification, Characterization, and Assay

A. Total Antioxidant Status

23. Measurement of Antioxidants in Human Blood Plasma
PAUL A. MOTCHNIK, BALZ FREI, AND BRUCE N. AMES 269
24. Total Antioxidant Status in Plasma and Body Fluids
CATHERINE RICE-EVANS AND NICHOLAS J. MILLER 279

B. Natural Source Antioxidants

Vitamin E

25. Analysis of Vitamin E Homologs in Plasma and Tissue: High-Performance Liquid Chromatography
WILLY SCHÜEP AND ROSEMARIE RETTENMAIER 294
26. Separation of the Eight Stereoisomers of all-*rac*- α -Tocopherol from Tissues and Plasma: Chiral Phase High-Performance Liquid Chromatography and Capillary Gas Chromatography
GEORGES RISS, ALFRED W. KORMANN, ERNST GLINZ, WILLI WALTHER, AND URS B. RANALDER 302
27. Tocopherone and Epoxytocopherone Products of Vitamin E Oxidation
DANIEL C. LIEBLER 310
28. Light-Induced Generation of Vitamin E Radicals: Assessing Vitamin E Regeneration
VALERIAN E. KAGAN AND LESTER PACKER 316
29. Antioxidative Activity of Tocotrienol in Heterogeneous System: Indication of Restriction within Membrane by Fluorescence Measurement
MASAKAZU YAMAOKA AND KANKI KOMIYAMA 320
30. Determination of Tocopherols and Tocopherolquinone in Human Red Blood Cell and Platelet Samples
GOVIND T. VATASSERY 327

Vitamin C

31. Vitamin C, Dehydroascorbate, and Uric Acid in Tissues and Serum: High Performance Liquid Chromatography
G. BARJA AND A. HERNANZ 331
32. *In Vivo* Determination of Superoxide and Vitamin C Radicals Using Cytochrome *c* and Superoxide Dismutase Derivatives
MASAYASU INOUE AND KEIKO KOYAMA 338

Quinones

33. Assay of Ubiquinones and Ubiquinol as Antioxidants
VALERIAN E. KAGAN, ELENA A. SERBINOVA, D. A. STOYANOVSKY, S. KHWAJA, AND LESTER PACKER 343
34. Antioxidant Properties of α -Tocopherol and α -Tocotrienol
ELENA A. SERBINOVA AND LESTER PACKER 354

Glutathione

35. Determination of Oxidized Glutathione in Blood: High-Performance Liquid Chromatography
MIGUEL ASEÑI, JUAN SASTRE, FEDERICO V. PALLARDO, JOSE M. ESTRELA,
AND JOSE VIÑA 367

Carotenoids and Vitamin A

36. Antioxidant Activity of α -Tocopherol, β -Carotene, and Ubiquinol in Membranes: *cis*-Parinaric Acid-Incorporated Liposomes
MASAHIKO TSUCHIYA, VALERIAN E. KAGAN, HANS-JOACHIM FREISLEBEN,
MASANOBU MANABE, AND LESTER PACKER 371
37. Singlet Oxygen Quenching by Carotenoids
ALFRED R. SUNDQUIST, KARLIS BRIVIBA, AND HELMUT SIES 384
38. Separation of Geometrical Isomers of β -Carotene and Lycopene
WILHELM STAHL AND HELMUT SIES 388
39. Lipoperoxyl Radical-Scavenging Activity of Vitamin A and Analogs in Homogeneous Solution
MARIA A. LIVREA AND LUISA TESORIERE 401

Flavonoids

40. Naturally Occurring Flavonoids: Structure, Chemistry, and High-Performance Liquid Chromatography Methods for Separation and Characterization
DIPAK K. DAS 410
41. Flavonoid Antioxidants: Rate Constants for Reactions with Oxygen Radicals
WOLF BORS, CHRISTA MICHEL, AND MANFRED SARAN 420
42. Assay of Condensed Tannins or Flavonoid Oligomers and Related Flavonoids in Plants
ANN E. HAGERMAN AND LARRY G. BUTLER 429
43. Role of Flavonoids and Iron Chelation in Antioxidant Action
ISABELLE MOREL, GÉRARD LESCOAT, PIERRE CILLARD, AND JOSIANE CILLARD 437
44. Actions of Flavonoids and Coumarins on Lipooxygenase and Cyclooxygenase
J. R. S. HOULT, MICHELE A. MORONEY, AND MIGUEL PAYÁ 443

 α -Lipoic Acid

45. Determination of Structure-Antioxidant Activity Relationships of Dihydrolipoic Acid
YUICHIRO JUSTIN SUZUKI, MASAHIKO TSUCHIYA, AND LESTER PACKER 454

Extracts from Natural Products

46. Antioxidant Action of *Ginkgo biloba* Extract EGb 761
LUCIA MARCOCCI, LESTER PACKER, MARIE-THERESE DROY-LEFAIX,
ABDELHAFID SEKAKI, AND MONIQUE GARDÈS-ALBERT 462

C. Synthetic Antioxidants: Drugs

47. Ebselen: A Glutathione Peroxidase Mimic
HELMUT SIES 476
48. *N*-Acetylcysteine
PETER MOLDEÚS AND IAN A. COTGREAVE 482
49. Preparation and Use of Glutathione Monoesters
MARY E. ANDERSON, ELLEN J. LEVY, AND ALTON MEISTER 492

50. Preparation and Properties of Glutathione Diethyl Ester and Related Derivatives ELLEN J. LEVY, MARY E. ANDERSON, AND ALTON MEISTER	499
51. Antioxidant Activity and Serum Levels of Probucol and Probucol Metabolites SIMON J. T. MAO, MARK T. YATES, AND RICHARD L. JACKSON	505
52. α -Phenyl <i>N</i> -tert-Butylnitron as Antioxidant for Low Density Lipoproteins DIANA M. LEE	513
53. Brain Antioxidant Activity of Spin Traps in Mongolian Gerbils JOHN M. CARNEY AND ROBERT A. FLOYD	523
54. Antioxidant Activity of Nitecapone and Its Analog OR-1246: Effect of Structural Modification on Antioxidant Action LUCIA MARCOCCI, YUICHIRO JUSTIN SUZUKI, MASAHIKO TSUCHIYA, AND LESTER PACKER	526
55. Penicillamine as Antioxidant MASAYUKI MIKI	542
56. Antioxidant Action of Lazaroids EDWARD D. HALL AND JOHN M. MCCALL	548
57. Antioxidant Properties of Aminosalicylates ALLEN M. MILES AND MATTHEW B. GRISHAM	555
58. Antioxidant Action of Stobadine LUBICA HORÁKOVÁ, HELMUT SIES, AND STEEN STEENKEN	572
59. Nitroxides as Antioxidants MURALI C. KRISHNA AND AMRAM SAMUNI	580
60. Tamoxifen and Estrogens as Membrane Antioxidants: Comparison with Cholesterol HELEN WISEMAN	590
61. Reactivity of Plasmalogens to Singlet Oxygen and Radicals OLIVIER H. MORAND	603
62. Antioxidant Activity of Calcium Channel Blocking Drugs I. TONG MAK AND WILLIAM B. WEGGLICKI	620
63. Interactions of Phenoxy Radical of Antitumor Drug, Etoposide, with Reductants in Solution and in Cell and Nuclear Homogenates: Electron Spin Resonance and High-Performance Liquid Chromatography T. G. GANTCHEV, J. E. VAN LIER, D. A. STOYANOVSKY, J. C. YALOWICH, AND VALERIAN E. KAGAN	631

VOLUME 235

BACTERIAL PATHOGENESIS (PART A: IDENTIFICATION AND REGULATION OF VIRULENCE FACTORS)

Virginia L. Clark and Patrik M. Bavoil

1. Recommendations for Working with Pathogenic Bacteria STEPHEN A. MORSE AND JOSEPH E. MCDADE	1
--	---

Section I. Evaluations of Virulence in Animal Models

2. Determination of Median Lethal and Infectious Doses in Animal Model Systems SUSAN WELKOS AND ALISON O'BRIEN	29
3. Experimental Keratoconjunctivitis (Sereny) Assay DENNIS J. KOPECKO	39

4. Mouse Respiratory Infection Models for Pertussis	ROBERTA D. SHAHIN AND JAMES L. COWELL	47
5. Chinchilla Model of Experimental Otitis Media for Study of Nontypable <i>Haemophilus influenzae</i> Vaccine Efficacy	BRUCE A. GREEN, WILLIAM J. DOYLE, AND JAMES L. COWELL	59
6. Animal Models for Ocular Infections	ROGER G. RANK AND JUDITH A. WHITTUM-HUDSON	69
7. Animal Models for Urogenital Infections	ROGER G. RANK	83
8. Animal Models for Meningitis	MARTIN G. TAUBER AND ANDRÉ ZWAHLÉN	93
9. Animal Models for Periodontal Disease	THERESA E. MADDEN AND JACK G. CATON	106
10. Animal Chamber Models for Study of Host-Parasite Interactions	CAROLINE ATTARDO GENCO AND ROBERT J. ARKO	120
11. Animal Models for Immunoglobulin A Secretion	DAVID F. KEREN AND MARK A. SUCKOW	140

Section II. Species and Strain Identification

12. Serogroup and Serotype Classification of Bacterial Pathogens	CARL E. FRASCH	159
13. Analysis of Genetic Variation by Polymerase Chain Reaction-Based Nucleotide Sequencing	KIMBERLYN NELSON AND ROBERT K. SELANDER	174
14. Determinations of Restriction Fragment Length Polymorphism in Bacteria Using Ribosomal RNA Genes	RIVKA RUDNER, BARBARA STUDAMIRE, AND ERICH D. JARVIS	184
15. DNA Fingerprinting of <i>Mycobacterium tuberculosis</i>	DICK VAN SOOLINGEN, PETRA E. W. DE HAAS, PETER W. M. HERMANS, AND JAN D. A. VAN EMBDEN	196
16. Phylogenetic Identification of Uncultured Pathogens Using Ribosomal RNA Sequences	THOMAS M. SCHMIDT AND DAVID A. RELMAN	205

Section III. Bacterial Cell Fractionation

17. Isolation of Outer Membranes	HIROSHI NIKAIIDO	225
18. Isolation and Purification of Periplasmic Binding Proteins	GIOVANNA FERRO-LUZZI AMES	234
19. Isolation and Characterization of Lipopolysaccharides, Lipooligosaccharides, and Lipid A	MICHAEL A. APICELLA, J. McLEOD GRIFFISS, AND HERMAN SCHNEIDER	242
20. Isolation of Peptidoglycan and Soluble Peptidoglycan Fragments	RAOUL S. ROSENTHAL AND ROMAN DZIARSKI	253
21. Purification of Streptococcal M Protein	VINCENT A. FISCHETTI	286
22. Isolation and Assay of <i>Pseudomonas aeruginosa</i> Alginate	THOMAS B. MAY AND A. M. CHAKRABARTY	295

23. Purification of *Escherichia coli* K Antigens
WILLIE F. VANN AND STEPHEN J. FREESE 304

Section IV. Microbial Acquisition of Iron

24. Deferration of Laboratory Media and Assays for Ferric and Ferrous Ions
CHARLES D. COX 315
25. Detection, Isolation, and Characterization of Siderophores
SHELLEY M. PAYNE 329
26. Effects of Iron Deprivation on Outer Membrane Protein Expression
J. B. NEILANDS 344
27. Identification and Isolation of Mutants Defective in Iron Acquisition
J. B. NEILANDS 352
28. Identification of Receptor-Mediated Transferrin-Iron Uptake Mechanism in *Neisseria gonorrhoeae*
CYNTHIA NAU CORNELISSEN AND P. FREDERICK SPARLING 356
29. Isolation of Genes Involved in Iron Acquisition by Cloning and Complementation of *Escherichia coli* Mutants
SUSAN E. H. WEST 363

Section V. Genetics and Regulation

30. Bacterial Transformation by Electroporation
JEFF F. MILLER 375
31. Analysis and Construction of Stable Phenotypes in Gram-Negative Bacteria with Tn5- and Tn10-Derived Minitransposons
VICTOR DE LORENZO AND KENNETH N. TIMMIS 386
32. Use of Transposons to Dissect Pathogenic Strategies of Gram-Positive Bacteria
ROBERT A. BURNE AND ROBERT G. QUTVEY, JR. 405
33. Identification of Bacterial Cell-Surface Virulence Determinants with *TnphoA*
MELISSA R. KAUFMAN AND RONALD K. TAYLOR 426
34. Temperature-Sensitive Mutants of Bacterial Pathogens: Isolation and Use to Determine Host Clearance and *in Vivo* Replication Rates
ANNE MORRIS HOOKE 448
35. Use of Conditionally Counterselectable Suicide Vectors for Allelic Exchange
SCOTT STIBITZ 458
36. Gene Replacement in *Pseudomonas aeruginosa*
DEBBIE S. TODER 466
37. Systems of Experimental Genetics for *Campylobacter* Species
PATRICIA GUERRY, RUIJIN YAO, RICHARD A. ALM, DONALD H. BURR, AND TREVOR J. TRUST 474
38. *In Vivo* Expression Technology for Selection of Bacterial Genes Specifically Induced in Host Tissues
JAMES M. SLAUCH, MICHAEL J. MAHAN, AND JOHN J. MEKALANOS 481
39. Regulation of Alginate Gene Expression in *Pseudomonas aeruginosa*
NICOLETTE A. ZIELINSKI, SIDDHARTHA ROYCHOUDHURY, AND A. M. CHAKRABARTY 493
40. Regulation of Expression of *Pseudomonas* Exotoxin A by Iron
DARA W. FRANK AND DOUGLAS G. STOREY 502
41. Regulation of Cholera Toxin by Temperature, pH, and Osmolarity
CLAUDETTE L. GARDEL AND JOHN J. MEKALANOS 517
42. Posttranslational Processing of Type IV Prepilin and Homologs by PilD of *Pseudomonas aeruginosa*
MARK S. STROM, DAVID N. NUNN, AND STEPHEN LORY 527

Section VI. Enzyme and Toxin Assays

43. Bacterial Immunoglobulin A ₁ Proteases	MARTHA H. MULKS AND RUSSELL J. SHOBERG	543
44. Elastase Assays	LYNN RUST, CALVIN R. MESSING, AND BARBARA H. IGLEWSKI	554
45. Zymographic Techniques for Detection and Characterization of Microbial Proteases	MARILYN S. LANTZ AND PAWEŁ CIBOROWSKI	563
46. Assays for Bacterial Type I Collagenases	JOHN D. GRUBB	594
47. Purification and Assays of Bacterial Gelatinases	JOHN D. GRUBB	602
48. Assays for Hyaluronidase Activity	WAYNE L. HYNES AND JOSEPH J. FERRETTI	606
49. ADP-Ribosylating Toxins	LUCIANO PASSADOR AND WALLACE IGLEWSKI	617
50. Photoaffinity Labeling of Active Site Residues in ADP-Ribosylating Toxins	STEPHEN F. CARROLL AND R. JOHN COLLIER	631
51. Activation of Cholera Toxin by ADP-Ribosylation Factors	JOEL MOSS, S.-C. TSAI, AND MARTHA VAUGHAN	640
52. Toxins That Inhibit Host Protein Synthesis	TOM G. OBRIG	647
53. Assays of Hemolytic Toxins	GAIL E. ROWE AND RODNEY A. WELCH	657
54. Identification and Assay of RTX Family of Cytolysins	ANTHONY L. LOBO AND RODNEY A. WELCH	667
55. Assay of Cytopathogenic Toxins in Cultured Cells	MONICA THELESTAM AND INGER FLORIN	679
56. Use of Lipid Bilayer Membranes to Detect Pore Formation by Toxins	BRUCE L. KAGAN AND YURI SOKOLOV	691
57. Uptake and Processing of Toxins by Mammalian Cells	CATHARINE B. SAELINGER AND RANDAL E. MORRIS	705

VOLUME 236

BACTERIAL PATHOGENESIS (PART B: INTERACTION OF PATHOGENIC BACTERIA WITH HOST CELLS)

Virginia L. Clark and Patrik M. Bavoil

Section I. Bacterial Effects on Immune Cells

1. Induction of Cytokines in Mononuclear and Vascular Cells by Endotoxin and Other Bacterial Products	HARALD LOPPNOW, HELMUT BRADE, ERNST THEODOR RIETSCHEL, AND HANS-DIETER FLAD	3
2. Effect of Bacterial Products on Colony-Stimulating Factor Production	MELINDA S. DETRICK, ROBERT KREISBERG, AND ROBERT N. MOORE	11
3. Effect of Bacterial Products on Tumor Necrosis Factor Production: Quantitation in Biologic Fluids or Tissues	ZELL A. MCGEE AND CHRISTOPHER M. CLEMENS	23
4. Induction of Prostaglandin Release from Macrophages by Bacterial Endotoxin	KEN-ICHI TANAMOTO	31
5. Effect of Bacterial Products on Interferon Production	GERALD SONNENFELD	42

6. Regulation of Osteoclastic Activity in Infection
J. EDWARD PUZAS, DAVID G. HICKS, SUSAN D. REYNOLDS,
AND REGIS J. O'KEEFE 47
7. Effect of Bacterial Products on Neutrophil Chemotaxis
JOHN R. KALMAR AND THOMAS E. VAN DYKE 58

Section II. Phagocytosis and Killing Assays

8. Measurement of Nonopsonic Phagocytic Killing by Human and Mouse Phagocytes
RICHARD F. REST AND DAVID P. SPEERT 91
9. Measurement of Opsonic Phagocytosis by Human Polymorphonuclear Neutrophils
JOHN R. KALMAR 108
10. Measurement of Human Neutrophil Respiratory Burst Activity during Phagocytosis of Bacteria
RICHARD F. REST 119
11. Complement-Mediated Bacterial Killing Assays
DANIEL P. MCQUILLEN, SUNITA GULATI, AND PETER A. RICE 137
12. Measurement of Phagosome-Lysosome Fusion and Phagosomal pH
THOMAS H. STEINBERG AND JOEL A. SWANSON 147
13. Neutrophil Defensins: Purification, Characterization, and Antimicrobial Testing
SYLVIA S. L. HARWIG, TOMAS GANZ, AND ROBERT I. LEHRER 160
14. Purification and Assay of Bactericidal/Permeability-Increasing Protein
JERROLD WEISS 173
15. Regulation of Bacterial Gene Expression in Response to Oxidative Stress
GISELA STORZ AND MICHEL B. TOLEDANO 196

Section III. Adherence

16. Bacterial Lectinlike Adhesins: Determination and Specificity JANINA GOLDBAR 211
17. Possible Interaction between Animal Lectins and Bacterial Carbohydrates
ROBERT E. MANDRELL, MICHAEL A. APICELLA, R. LINDSTEDT,
AND HACKEN LEFFLER 231
18. Coaggregation between Bacterial Species
SUSAN A. KINDER AND STANLEY C. HOLT 254
19. Purification of Somatic Pili
GARY K. SCHOOLNIK 271
20. Genetic, Biochemical, and Structural Studies of Biogenesis of Adhesive Pili in Bacteria
META J. KUEHN, FRANÇOISE JACOB-DUBUISSON, KAREN DODSON,
LYNN SLONIM, ROBERT STRIKER, AND SCOTT J. HULTGREN 282
21. Isolation and Identification of Eukaryotic Receptors Promoting Bacterial Internalization
GUY TRAN VAN NHIEU AND RALPH R. ISBERG 307
22. Identification of Fibronectin as a Receptor for Bacterial Cytoadherence
JOHN F. ALDERETE, ROSSANA ARROYO, AND MICHAEL W. LEHKER 318
23. Interactions of Bacteria with Leukocyte Integrins
EVA ROZDZINSKI AND ELAINE TUOMANEN 333
24. Solid-Phase Binding of Microorganisms to Glycolipids and Phospholipids
CAROLYN D. DEAL AND HOWARD C. KRIVAN 346
25. Characterization of Microbial Host Receptors
PER FALK, THOMAS BORÉN, AND STAFFAN NORMARK 353

Section IV. Invasion and Intracellular Survival in Eukaryotic Cells

26. Culture and Isolation of *Chlamydia trachomatis*
JULIUS SCHACHTER AND PRISCILLA B. WYRICK 377
27. Culture of *Treponema pallidum*
DAVID L. COX 390
28. Measurement of Invasion by Gentamicin Resistance
ERIC A. ELSINGHORST 405
29. Measurements of Invasion by Antibody Labeling and Electron Microscopy
JOSEPH P. M. VAN PUTTEN, JAN F. L. WEEL, AND HEIKE U. C. GRASSMÉ 420
30. Polarized Epithelial Monolayers: Model Systems to Study Bacterial Interactions
with Host Epithelial Cells M. GRACIELA PUCCIARELLI AND B. BRETT FINLAY 438
31. Interjunctional Invasion of Endothelial Cell Monolayers
DAVID A. HAAKE AND MICHAEL A. LOVETT 447
32. Intracellular Growth of Bacteria
SIÂN JONES AND DANIEL A. PORTNOY 463
33. Inhibitors of Cytoskeletal Function and Signal Transduction to Study Bacterial
Invasion ILAN ROSENSHINE, SHARON RUSCHKOWSKI, AND B. BRETT FINLAY 467
34. Methods to Visualize Actin Polymerization Associated with Bacterial Invasion
LEWIS G. TILNEY AND MARY S. TILNEY 476
35. Measurement of Free Intracellular Calcium Levels in Epithelial Cells as Conse-
quence of Bacterial Invasion JOHN L. PACE AND JORGE E. GALÁN 482

Section V. Identification of Genes Involved in Invasion

36. *Shigella flexneri*: Isolation of Noninvasive Mutants of Gram-Negative Pathogens
ROBERT MÉNARD AND PHILIPPE J. SANSONETTI 493
37. Isolation of *Salmonella* Mutants Defective for Intracellular Survival
FRANCES BOWE AND FRED HEFFRON 509
38. Small Plaque Mutants
SIÂN JONES AND DANIEL A. PORTNOY 526
39. Isolation of Hyperinvasive Mutants of *Salmonella*
CATHERINE A. LEE AND STANLEY FALKOW 531
40. Molecular Cloning of Invasion Genes from *Yersinia* and *Salmonella*
VIRGINIA L. MILLER AND BARBARA J. STONE 546
41. Molecular Cloning and Expression of Internalin in *Listeria*
JEAN-LOUIS GAILLARD, S. DRAMSI, PATRICK BERCHE, AND PASCALE COSSART 551
42. Use of *Staphylococcus aureus* Coated with Invasin Derivatives to Assay Invasin
Function SUSANNAH RANKIN, GUY TRAN VAN NHIEU, AND RALPH R. ISBERG 566

VOLUME 237

HETEROTRIMERIC G PROTEINS

Ravi Iyengar

Section I. G_{α} Subunits

1. Measurement of Receptor-Stimulated Guanosine 5'-O-(γ -Thio)triphosphate Bind-
ing by G Proteins
THOMAS WIELAND AND KARL H. JAKOBS 1
2. Receptor-Stimulated Hydrolysis of Guanosine 5'-Triphosphate in Membrane Prep-
arations
PETER GIERSECHIK, THOMAS BOUILLON, AND KARL H. JAKOBS 13

3. Regulation of G-Protein Activation by Mastoparans and Other Cationic Peptides ELLIOTT M. ROSS AND TSUTOMU HIGASHIJIMA	26
4. Guanosine 5'-O-(γ -Thio)triphosphate Binding Assay for Solubilized G Proteins DONNA J. CARTY AND RAVI IYENGAR	38
5. Activation of Cholera Toxin by ADP-Ribosylation Factors: 20-kDa Guanine Nucleotide-Binding Proteins JOEL MOSS, RANDY S. HAUN, SU-CHEN TSAI, CATHERINE F. WELSH, FANG-JEN SCOTT LEE, S. RUSS PRICE, AND MARTHA VAUGHAN	44
6. Pertussis Toxin-Catalyzed ADP-Ribosylation of G Proteins DONNA J. CARTY	63
7. Synthesis and Use of Radioactive Photoactivable NAD ⁺ Derivatives as Probes for G-Protein Structure RICHARD R. VAILLANCOURT, N. DHANASEKARAN, AND ARNOLD E. RUOHO	70
8. Photoaffinity Guanosine 5'-Triphosphate Analogs as a Tool for the Study of GTP-Binding Proteins MARK M. RASENICK, MADHAVI TALLURI, AND WILLIAM J. DUNN III	100
9. Preparation of Activated α Subunits of G _s and G _i s: From Erythrocyte to Activated Subunit LUTZ BIRNBAUMER, DAGOBERTO GRENET, FERNANDO RIBEIRO-NETO, AND JUAN CODINA	110
10. Purification and Separation of Closely Related Members of Pertussis Toxin-Substrate G Proteins TOSHIAKI KATADA, KENJI KONTANI, ATSUSHI INANOBE, ICHIRO KOBAYASHI, YOSHIHARU OHOKA, HIROSHI NISHINA, AND KATSUNOBU TAKAHASHI	131
11. Purification of Transducin JOËLLE BIGAY AND MARC CHABRE	139
12. Expression of G-Protein α Subunits in <i>Escherichia coli</i> ETHAN LEE, MAURINE E. LINDER, AND ALFRED G. GILMAN	146
13. Synthesis and Applications of Affinity Matrix Containing Immobilized $\beta\gamma$ Subunits of G Proteins IOK-HOU PANG, ALAN V. SMRCKA, AND PAUL C. STERNWEIS	164
14. Purification of Activated and Heterotrimeric Forms of G _q Proteins JONATHAN L. BLANK AND JOHN H. EXTON	174
15. Purification of Phospholipase C-Activating G Protein, G ₁₁ , from Turkey Erythrocytes GARY L. WALDO, JOSÉ L. BOYER, AND T. KENDALL HARDEN	182
16. Purification of Recombinant G ₉ α , G ₁₁ α , and G ₁₆ α from Sf9 Cells JOHN R. HEPLER, TOHRU KOZASA, AND ALFRED G. GILMAN	191
17. Expression and Purification of G-Protein α Subunits Using Baculovirus Expression System STEPHEN G. GRABER, ROBERT A. FIGLER, AND JAMES C. GARRISON	212
18. Analysis of G-Protein α and $\beta\gamma$ Subunits by <i>in Vitro</i> Translation EVA J. NEER, BRADLEY M. DENKER, THOMAS C. THOMAS, AND CARL J. SCHMIDT	226
19. Assays for Studying Functional Properties of <i>in Vitro</i> Translated G ₃ α Subunit YVES AUDIGIER	239
20. Myristoylation of G-Protein α Subunits SUSANNE M. MUMBY AND MAURINE E. LINDER	254
21. Specificity and Functional Applications of Anti-peptide Antisera Which Identify G-Protein α Subunits GRAEME MILLIGAN	268
22. Identification of Receptor-Activated G Proteins: Selective Immunoprecipitation of Photolabeled G-Protein α Subunits KARL-LUDWIG LAUGWITZ, KARSTEN SPICHER, GÜNTER SCHULTZ, AND STEFAN OFFERMANN	283
23. Identification of Mutant Forms of G-Protein α Subunits in Human Neoplasia by Polymerase Chain Reaction-Based Techniques JOHN LYONS	295

24. Detection of Mutations and Polymorphisms of $G_{i\alpha}$ Subunit Gene by Denaturing Gradient Gel Electrophoresis PABLO V. GEJMAN AND LEE S. WEINSTEIN 308
25. Construction of Mutant and Chimeric G-Protein α Subunits
SIM WINITZ, MARIJANE RUSSELL, AND GARY L. JOHNSON 321
26. Design of Degenerate Oligonucleotide Primers for Cloning of G-Protein α Subunits
THOMAS M. WILKIE, ANNA M. ARAGAY, A. JOHN WATSON,
AND MELVIN I. SIMON 327
27. Microinjection of Antisense Oligonucleotides to Assess G-Protein Subunit Function
CHRISTIANE KLEUSS, GÜNTER SCHULTZ, AND BURGHARDT WITTIG 345
28. Inactivation of G-Protein Genes: Double Knockout in Cell Lines
RICHARD M. MORTENSEN AND J. G. SEIDMAN 356
29. Targeted Inactivation of the $G_{12\alpha}$ Gene with Replacement and Insertion Vectors:
Analysis in a 96-Well Plate Format
UWE RUDOLPH, ALLAN BRADLEY, AND LUTZ BIRNBAUMER 366
30. G-Protein Assays in *Dictyostelium*
B. EWA SNAAR-JAGALSKA AND PETER J. M. VAN HAASTERT 387
31. Fluorescence Assays for G-Protein Interactions RICHARD A. CERIONE 409
32. Specific Peptide Probes for G-Protein Interactions with Receptors
HEIDI E. HAMM AND HELEN M. RARICK 423
33. Vaccinia Virus Systems for Expression of $G\alpha$ Genes in S49 Cells
FRANKLIN QUAN AND MICHAEL FORTE 436

Section II. $G\beta\gamma$ Subunits

34. Purification of $T\beta\gamma$ Subunit of Transducin JOËLLE BIGAY AND MARC CHABRE 449
35. Adenylyl Cyclase Assay for $\beta\gamma$ Subunits of G Proteins
JIANQIANG CHEN, DONNA J. CARTY, AND RAVI IYENGAR 451
36. Synthesis and Use of Biotinylated $\beta\gamma$ Complexes Prepared from Bovine Brain G
Proteins JANE DINGUS, MICHAEL D. WILCOX, RUSSELL KOHNEN,
AND JOHN D. HILDEBRANDT 457
37. Design of Oligonucleotide Probes for Molecular Cloning of β and γ Subunits
CHRISTINE GALLAGHER AND NARASIMHAN GAUTAM 471
38. Characterization of Antibodies for Various G-Protein β and γ Subunits
ALEXEY N. PRONIN AND NARASIMHAN GAUTAM 482
39. Preparation, Characterization, and Use of Antibodies with Specificity for G-Protein
 γ Subunits JANET D. ROBISHAW AND ERIC A. BALCUEVA 498
40. Isoprenylation of γ Subunits and G-Protein Effectors
BERNARD K.-K. FUNG, JANMEET S. ANANT, WUN-CHEN LIN, OLIVIA C. ONG,
AND HARVEY K. YAMANE 509

VOLUME 238

HETEROTRIMERIC G PROTEIN EFFECTORS Ravi Iyengar

Section I. cGMP Phosphodiesterase

1. Purification of Bovine Retinal cGMP Phosphodiesterase
ANDREA TAR, TUOW D. TING, AND YEE-KIN HO 3

2. Specific Peptide Probes for G-Protein Interaction with Effectors

HELEN M. RARICK, NIKOLAI O. ARTEMYEV, JOHN S. MILLS,
NIKOLAI P. SKIBA, AND HEIDI E. HAMM

13

Section II. Adenylyl Cyclases

3. Determination of Adenylyl Cyclase Catalytic Activity Using Single and Double Column Procedures

ROGER A. JOHNSON, ROBERT ALVAREZ, AND YORAM SALOMON 31

4. Preparation and Use of "P"-Site-Targeted Affinity Ligands for Adenylyl Cyclases

ROGER A. JOHNSON AND ILANA SHOSHANI 56

5. Regulation of *Ca²⁺*-Sensitive Adenylyl Cyclases by Calcium *in Vitro* and *in Vivo*

DERMOT M. F. COOPER 71

6. *G_i* Assays in Transfected Cells

YUNG H. WONG 81

7. Expression and Purification of Recombinant Adenylyl Cyclases in S19 Cells

RONALD TAUSSIG, WEI-JEN TANG, AND ALFRED G. GILMAN 95

8. Transient Expression Assays for Mammalian Adenylyl Cyclases

OFER JACOBOWITZ, JIANQIANG CHEN, AND RAVI IYENGAR 108

9. Identification of Adenylyl Cyclases by Amplification Using Degenerate Primers

RICHARD T. PREMONT 116

Section III. Phospholipases C

10. Assays for G-Protein Regulation of Phospholipase C Activity

MICHAEL DE VIVO 131

11. Chloride Current Assay for Phospholipase C in *Xenopus* Oocytes

EMMANUEL M. LANDAU AND ROBERT D. BLITZER 140

12. Use of Cytosol-Depleted HL60 Cells for Reconstitution Studies of G-Protein-Regulated Phosphoinositide-Specific Phospholipase C- β IsozymesSHAMSHAD COCKCROFT, GERAINT M. H. THOMAS, EMER CUNNINGHAM,
AND ANDREW BALL 15413. Purification of Phosphatidylinositol Transfer Protein from Brain Cytosol for Reconstituting G-Protein-Regulated Phosphoinositide-Specific Phospholipase C- β Isozymes

GERAINT M. H. THOMAS, EMER CUNNINGHAM, AND SHAMSHAD COCKCROFT 168

14. Stimulation of Phospholipase C by G-Protein $\beta\gamma$ Subunits

PETER GIERSCHIK AND MONTSERRAT CAMPS 181

15. Purification of G-Protein-Regulated Phospholipase C from Turkey Erythrocytes

GARY L. WALDO, ANDREW J. MORRIS, AND T. KENDALL HARDEN 195

16. Phospholipase C Activity in *Dictyostelium discoideum* Using Endogenous Nonradioactive Phosphatidylinositol 4,5-Bisphosphate as Substrate

ANTHONY A. BOMINAAR AND PETER J. M. VAN HAASTERT 207

17. Purification of Phospholipase C- β_3 from Rat Brain

DEOK-YOUNG JHON, DONGEUN PARK, AND SUE GOO RHEE 219

18. Characterization of Phospholipase C Isozymes in Bovine Retina: Purification of Phospholipase C- β

CHANG-WON LEE, KWEON-HAENG LEE, AND SUE GOO RHEE 227

19. Purification of 110 kDa Phosphoinositide Phospholipase C Activated by G-Protein $\beta \gamma$ Subunits J. L. BLANK AND J. H. EXTON 237
20. Amplification of Phosphatidylinositol-Specific Phospholipase C- β Isoforms Using Degenerate Primers HAI-WEN MA, RAVI IYENGAR, AND RICHARD T. PREMONT 244

Section IV. Growth and Transformation

21. Analysis of Guanine Nucleotides Associated with Protooncogene Ras RICHARD R. VAILLANCOURT, ANNE E. HARWOOD, AND SIM WINITZ 255
22. Measuring Activation of Kinases in Mitogen-Activated Protein Kinase Regulatory Network ANNE M. GARDNER, CAROL A. LANGE-CARTER, RICHARD R. VAILLANCOURT, AND GARY L. JOHNSON 259
23. Transcriptional Activation Analysis of Oncogene Function CRAIG A. HAUSER, CHANNING J. DER, AND ADRIENNE D. COX 271
24. Biological Assays for Cellular Transformation ADRIENNE D. COX AND CHANNING J. DER 277

Section V. Ions and Channels

25. Monitoring of Receptor-Mediated Changes in Intracellular Calcium at the Cellular and Subcellular Levels by Microfluorimetry and Imaging STEPHEN R. RAWLINGS, JEAN-MARC THELER, AND WERNER SCHLEGEL 297
26. Combination of Microfluorimetric Monitoring of Cytosolic Calcium and pH with Patch Clamp Electrophysiological Recordings in Neutrophil Granulocytes NICOLAS DEMAUREX, STEPHEN R. RAWLINGS, KARL-HEINZ KRAUSE, MARISA E. E. JACONI, P. DANIEL LEW, AND WERNER SCHLEGEL 308
27. G-Protein-Mediated Pathways Assayed by Electrophysiology and Confocal Microscopy LISA STEHNO-BITTEL, JEFF AMUNDSON, AND DAVID E. CLAPHAM 321
28. Purification and Reconstitution of N-Type Calcium Channel Complex from Rabbit Brain DERRICK R. WITCHER, MICHEL DE WAARD, STEVEN D. KAHL, AND KEVIN P. CAMPBELL 335
29. Infusion of Guanine Nucleotides through Recording Electrodes for Studies on G-Protein Regulation of Ion Currents and Channels RODRIGO ANDRADE 348
30. Injection of Antisera into Cells to Study G-Protein Regulation of Channel Function I. MCFADZEAN, M. P. CAULFIELD, Y. VALLIS, AND D. A. BROWN 357
31. Whole-Cell Clamp Analysis for G-Protein Regulation of Channels J. HESCHELER 365
32. Whole-Cell Patch Recording in Brain Slices ROBERT D. BLITZER AND EMMANUEL M. LANDAU 375
33. Reconstitution of Receptor-Regulated Ion Channels in Isolated Patch Membrane ATSUKO YATANI 385
34. Regulation of Potassium Channels by G-Protein Subunits and Arachidonic Acid Metabolites MITSUHIKO YAMADA, ANDRE TERZIC, AND YOSHIHISA KURACHI 394

VOLUME 239

NUCLEAR MAGNETIC RESONANCE (PART C)

Thomas L. James and Norman J. Oppenheimer

Section I. Techniques

A. Experimentation

1. Practical Introduction to Theory and Implementation of Multinuclear, Multidimensional Nuclear Magnetic Resonance Experiments
ARTHUR S. EDISON, FRITS ABILDGAARD, WILLIAM M. WESTLER,
ED S. MOOBERRY, AND JOHN L. MARKLEY 3
2. Measurement of Homo- and Heteronuclear *J* Couplings from Quantitative *J* Correlation
GEERTEN W. VUISTER, STEPHAN GRZESIEK, FRANK DELAGLIO,
ANDY C. WANG, ROLF TSCHUDIN, GUANG ZHU, AND AD BAX 79
3. Two-Dimensional Exchange Spectroscopy of Proteins
SLOBODAN I. MACURA, WILLIAM M. WESTLER, AND JOHN L. MARKLEY 106
4. Pulsed-Field Gradients: Theory and Practice
JAMES KEELER, ROBIN T. CLOWES, ADRIAN L. DAVIS, AND ERNEST D. LAUE 145
5. Selective Pulses and Their Applications to Assignment and Structure Determination in Nuclear Magnetic Resonance
LYNDON EMSLEY 207
6. Modifications of Older Model Nuclear Magnetic Resonance Console for Collection of Multinuclear, Multidimensional Spectral Data
ED S. MOOBERRY, FRITS ABILDGAARD, AND JOHN L. MARKLEY 247

B. Data Processing

7. Using Maximum Likelihood Spectral Deconvolution in Multidimensional Nuclear Magnetic Resonance
PHILIP N. BORER AND GEORGE C. LEVY 257
8. Automated and Semiautomated Analysis of Homo- and Heteronuclear Multidimensional Nuclear Magnetic Resonance Spectra of Proteins: The Program Pronto
MOGENS KJÆR, KIM VILBOUR ANDERSEN, AND FLEMMING M. POULSEN 288
9. Automated Assignment of Multidimensional Nuclear Magnetic Resonance Spectra
HARTMUT OSCHKINAT AND DAVID CROFT 308
10. Quantitative Information from Complicated Nuclear Magnetic Resonance Spectra of Biological Macromolecules
JENS J. LED AND HENRIK GESMAR 318

Section II. Protein Structure

A. General

11. Multidimensional Heteronuclear Nuclear Magnetic Resonance of Proteins
G. MARIUS CLORE AND ANGELA M. GRONENBORN 349
12. Chemical Shifts as a Tool for Structure Determination
DAVID S. WISHART AND BRIAN D. SYKES 363
13. Use of Chemical Shifts and Coupling Constants in Nuclear Magnetic Resonance Structural Studies on Peptides and Proteins
DAVID A. CASE, H. JANE DYSON, AND PETER E. WRIGHT 392

14. Assessment of Quality of Derived Macromolecular Structures

THOMAS L. JAMES 416

B. Classes of Proteins

15. Nuclear Magnetic Resonance Study of Antibodies: A Multinuclear Approach
YOJI ARATA, KOICHI KATO, HIDEO TAKAHASHI, AND ICHIO SHIMADA 440
16. Strategy for Studying Modular Proteins: Application to Complement Modules
PAUL N. BARLOW AND IAIN D. CAMPBELL 464
17. Two-Dimensional Nuclear Magnetic Resonance Spectra of Paramagnetic Systems
LUCIA BANCIL, IVANO BERTINI, AND CLAUDIO LUCHINAT 485
18. Methods to Study Membrane Protein Structure in Solution
GILLIAN D. HENRY AND BRIAN D. SYKES 515
19. Experimental Nuclear Magnetic Resonance Studies of Membrane Proteins
S. J. OPELLA, Y. KIM, AND P. McDONNELL 536

Section III. Dynamics and Disorder

20. Investigation of Protein Motions via Relaxation Measurements
JEFFREY W. PENG AND GERHARD WAGNER 563
21. Nuclear Magnetic Resonance Measurements of Slow Conformational Dynamics
in Macromolecules ANDREW N. LANE AND JEAN-FRANÇOIS LEFÈVRE 596
22. Accounting for Molecular Mobility in Structure Determination Based on Nuclear
Magnetic Resonance Spectroscopic and X-Ray Diffraction Data
WILFRED F. VAN GUNSTEREN, ROGER M. BRUNNE, PIET GROS,
RENÉ C. VAN SCHAIK, CELIA A. SCHIFFER, AND ANDREW E. TORDA 619

Section IV. Protein-Ligand Interactions

23. Protein-Ligand Interactions: Exchange Processes and Determination of Ligand
Conformation and Protein-Ligand Contacts
L. Y. LIAN, I. L. BARSUKOV, M. J. SUTCLIFFE, K. H. SZE,
AND G. C. K. ROBERTS 657
24. Nuclear Magnetic Studies of Protein-Peptide Complexes
A. JOSHUA WAND AND JAMES H. SHORT 700
25. Nuclear Magnetic Resonance Methods for Studying Protein-Ligand Complexes
ANDREW M. PETROS AND STEPHEN W. FESIK 717
26. Use of Nuclear Magnetic Resonance in Probing Ligand-Macromolecule Interac-
tions DAVID E. WEMMER AND PHILIP G. WILLIAMS 739

VOLUME 240

NUMERICAL COMPUTER METHODS (PART B)

Michael L. Johnson and Ludwig Brand

1. Use of Least-Squares Techniques in Biochemistry MICHAEL L. JOHNSON 1
2. Parameter Estimates from Nonlinear Models DONALD G. WATTS 23

3. Effects of Heteroscedasticity and Skewness on Prediction in Regression: Modeling Growth of the Human Heart	ROBERT D. ABBOTT AND HOWARD P. GUTGESELL	37
4. Artifacts of Fourier Series Analysis	MICHAEL L. JOHNSON AND MICHELLE LAMPL	51
5. Quantification of Evolution from Order to Randomness in Practical Time Series Analysis	STEVEN M. PINCUS	68
6. Sequential versus Simultaneous Analysis of Data: Differences in Reliability of Derived Quantitative Conclusions	MARTIN STRAUME	89
7. Model-Independent Quantification of Measurement Error: Empirical Estimation of Discrete Variance Function Profiles Based on Standard Curves	MARTIN STRAUME, JOHANNES D. VELDHIJS, AND MICHAEL L. JOHNSON	121
8. Impact of Variance Function Estimation to Regression and Calibration	LIE-JU HWANG	150
9. Application of Kalman Filter to Computational Problems in Statistics	EMERY N. BROWN AND CHRISTOPHER H. SCHMID	171
10. Modeling Chemical Reactions: Jacobian Paradigm and Related Issues	RICHARD I. SHRAGER	181
11. The Mathematics of Biological Oscillators	G. BARD ERMENROUT	198
12. Diffusion-Modulated Energy Transfer and Quenching: Analysis by Numerical Integration of Diffusion Equation in Laplace Space	JÓZEF KUŠBA AND JOSEPH R. LAKOWICZ	216
13. Maximum Entropy Method of Data Analysis in Time-Resolved Spectroscopy	JEAN-CLAUDE BROCHON	262
14. Analysis of Kinetic Data: Practical Applications of Computer Simulation and Fitting Programs	CARL FRIEDEN	311
15. Determination of Rate and Equilibrium Binding Constants for Macromolecular Interactions by Surface Plasmon Resonance	DANIEL J. O'SHANNESY, MICHAEL BRIGHAM-BURKE, K. KARL SONESON, PRESTON HENSLEY, AND IAN BROOKS	323
16. Analysis of Nonequilibrium Dynamics of Bound, Free, and Total Plasma Ligand Concentrations over Time Following Nonlinear Secretory Inputs: Kinetics of Two or More Hormones Pulsed into Compartments Containing Multiple Variable-Affinity Binding Proteins	JOHANNES D. VELDHIJS, LINDSAY M. FAUNT, AND MICHAEL L. JOHNSON	349
17. Testing Pulse Detection Algorithms with Simulations of Episodically Pulsatile Substrate, Metabolite, or Hormone Release	JOHANNES D. VELDHIJS AND MICHAEL L. JOHNSON	377
18. Error Analysis of Macromolecular Structures Determined with Nuclear Magnetic Resonance Data	JOHN W. SHRIVER AND STEPHEN EDMONDSON	415
19. Characterization of Enzyme-Complex Formation by Analysis of Nuclear Magnetic Resonance Line Shapes	CAROL BETH POST	438
20. Computer Simulations of Nuclear Overhauser Effect Spectra of Complex Oligosaccharides	C. ALLEN BUSH	446
21. Determining Confidence Intervals for Parameters Derived from Analysis of Equilibrium Analytical Ultracentrifugation Data	IAN BROOKS, DONALD G. WATTS, K. KARL SONESON, AND PRESTON HENSLEY	459
22. Boundary Analysis in Sedimentation Velocity Experiments	WALTER F. STAFFORD III	478

23. Statistical Thermodynamic Analysis of Differential Scanning Calorimetry Data: Structural Deconvolution of Heat Capacity Function of Proteins	ERNESTO FREIRE	502
24. Analysis of Two-Dimensional Differential Scanning Calorimetry Data: Elucidation of Complex Biomolecular Energetics	MARTIN STRAUME	530
25. Monte Carlo Simulations of Membranes: Phase Transition of Small Unilamellar Dipalmitoylphosphatidylcholine Vesicles	ISTVÁN P. SUGÁR, RODNEY L. BILTONEN, AND NEIL MITCHARD	569
26. Analysis of Drug-DNA Binding Isotherms: A Monte Carlo Approach	JOHN J. CORREIA AND JONATHAN B. CHAIRES	593
27. Analysis of Multidimensional Spectroscopic Data to Monitor Unfolding of Proteins	GLEN RAMSAY AND MAURICE R. EFTINK	615
28. Estimating Binding Constants for Site-Specific Interactions between Monovalent Ions and Proteins	BERTRAND GARCÍA-MORENO E.	645
29. Pattern Recognition Metric for Comparison of Protein Structures Based on Amino Acid Sequences	JOHN A. SCHETZ	667
30. Fluorescence Lifetime Imaging Microscopy: Homodyne Technique Using High-Speed Gated Image Intensifier	HENRYK SZMACINSKI, JOSEPH R. LAKOWICZ, AND MICHAEL L. JOHNSON	723
31. Computations of Time-Dependent Photon Migration for Biomedical Optical Imaging	EVA M. SEVICK-MURACA	748
32. Applying Bifurcation Theory to Enzyme Kinetics	CRAIG G. HOCKER	781

VOLUME 241

RETROVIRAL PROTEASES

Lawrence C. Kuo and Jules A. Shafer

Section I. Bacterial Expression and Enzymatic Assays

1. Expression Systems for Retroviral Proteases	JEFFREY STEBBINS AND CHRISTINE DEBOUCK	3
2. Genetic Approaches Designed to Minimize Cytotoxicity of Retroviral Protease	C. J. RIZZO AND B. D. KORANT	16
3. Host Strain Selection for Bacterial Expression of Toxic Proteins	ELIZABETH CHIEN	29
4. Assay Methods for Retroviral Proteases	CHRISTOPHER U. T. HELLEN	46
5. Analysis of Human Immunodeficiency Virus Type 1 Protease Activity in Eukaryotic and Bacterial Cells	ANDREW H. KAPLAN, MARIANNE MANCHESTER, LORRAINE EVERITT, AND RONALD SWANSTROM	58
6. Synthetic Approaches to Continuous Assays of Retroviral Proteases	GRANT A. KRAFT AND GARY T. WANG	70

Section II. Purification and Characterization

7. Identification, Purification, and Cell Culture Assays of Retroviral Proteases	KLAUS VON DER HELM, SIGRIG SEELMEIER, ALEXEI KISSELEV, AND HANS NITSCHKO	89
--	--	----

8. Stability of Dimeric Retroviral Proteases	PAUL L. DARKE	104
9. Use of Steady State Kinetic Methods to Elucidate Kinetic and Chemical Mechanisms of Retroviral Proteases	THOMAS D. MEEK, EVELYN J. RODRIGUEZ, AND THELMA S. ANGELES	127
10. X-Ray Structures of Retroviral Proteases and Their Inhibitor-Bound Complexes	DAGMAR RINGE	157
11. Probing Structure-Function Relationships in Human Immunodeficiency Virus Type 1 Protease via Molecular Dynamics Simulation	WILLIAM E. HARTE, JR., AND DAVID L. BEVERIDGE	178
12. Relationships of Human Immunodeficiency Virus Protease with Eukaryotic Aspartic Proteases	XIN-LI LIN, YING-ZHANG LIN, AND JORDAN TANG	195
 Section III. Substrate Specificity and Inhibitor Design		
13. Processing of Retroviral Gag Polyproteins: An <i>in Vitro</i> Approach	CAROL CARTER AND GABRIELE ZYBARTH	227
14. Subsite Preferences of Retroviral Proteinases	BEN M. DUNN, ALLA GUSTCHINA, ALEXANDER WLODAWER, AND JOHN KAY	254
15. Specificity of Retroviral Proteases: An Analysis of Viral and Nonviral Protein Substrates	ALFREDO G. TOMASSELLI AND ROBERT L. HEINRIKSON	279
16. Rapid Determination of Endoprotease Specificity Using Peptide Mixtures and Edman Degradation Analysis	FRANK R. MASIAZ AND BRUCE A. MALCOLM	302
17. Design of Tight-Binding Human Immunodeficiency Virus Type 1 Protease Inhibitors	JOSEPH P. VACCA	311
18. Design of Symmetry-Based, Peptidomimetic Inhibitors of Human Immunodeficiency Virus Protease	DALE J. KEMPF	334
19. Advances in Automated Docking Applied to Human Immunodeficiency Virus Type 1 Protease	MICHAEL D. MILLER, ROBERT P. SHERIDAN, SIMON K. KEARSLEY, AND DENNIS J. UNDERWOOD	354
20. Use of Molecular Dynamics and Free Energy Perturbation Calculations in Anti-Human Immunodeficiency Virus Drug Design	MARGARET A. MCCARRICK AND PETER KOLLMAN	370
21. Qualitative Study of Drug Resistance in Retroviral Protease Using Structural Modeling and Site-Directed Mutagenesis	J. CHRIS CULBERSON, BRUCE L. BUSH, AND VINOD V. SARDANA	385

VOLUME 242

NEOGLYCOCONJUGATES (PART A: SYNTHESIS)

Y.C. Lee and Reiko T. Lee

Section I. Neoglycoproteins

1. Residualizing Glyconjugates: Biologically Inert Tracers for Studies on Protein Endocytosis and Catabolism	SUZANNE R. THORPE AND JOHN W. BAYNES	3
2. Ganglioside-Based Neoglycoproteins	JAMES A. MAHONEY AND RONALD L. SCHNAAR	17

3. Leprosy-Specific Neoglycoconjugates: Synthesis and Application to Serodiagnosis of Leprosy	PATRICK J. BRENNAN, DELPHI CHATTERJEE, TSUYOSHI FUJIWARA, AND SANG-NAE CHO	27
4. Detection and Quantification of Carbohydrate-Binding Sites on Cell Surfaces and in Tissue Sections by Neoglycoproteins	HANS-JOACHIM GABIUS, SABINE ANDRÉ, ANDRÉ DANGUY, KLAUS KAYSER, AND SIGRUN GABIUS	37
5. Chemical Glycosylation of Recombinant Interleukin 2	SUBRAMANIAN SABESAN AND T. JUHANI LINNA	46
6. Neoglycoprotein-Liposome and Lectin-Liposome Conjugates as Tools for Carbohydrate Recognition Research	NOBORU YAMAZAKI, MAKOTO KODAMA, AND HANS-JOACHIM GABIUS	56
7. Modification of Proteins with Polyethylene Glycol Derivatives	YUJI INADA, AYAKO MATSUSHIMA, MISAO HIROTO, HIROYUKI NISHIMURA, AND YOH KODERA	65
8. Michael Additions for Synthesis of Neoglycoproteins	A. ROMANOWSKA, S. J. MEUNIER, F. D. TROPPER, C. A. LAFERRIÈRE, AND R. ROY	90
9. Isolation, Modification, and Conjugation of Sialyl $\alpha(2\rightarrow3)$ -Lactose	C. A. LAFERRIÈRE AND R. ROY	102
10. Coupling of Carbohydrates to Proteins by Diazonium and Phenylisothiocyanate Reactions	CHERYL M. REICHERT, COLLEEN HAYES, AND IRWIN J. GOLDSTEIN	108
11. Coupling of Aldobionic Acids to Proteins Using Water-Soluble Carbodiimide	JÖRGEN LÖNNGREN AND IRWIN J. GOLDSTEIN	116
12. Coupling of Oligosaccharides to Proteins Using <i>p</i> -Trifluoroacetamidoaniline	ELISABET KALLIN	119

Section II. Neoglycolipids

13. Neoglycolipids of 1-Deoxy-1-phosphatidylethanolaminolactitol Type: Synthesis, Structure Analysis, and Use as Probes for Characterization of Glycosyltransferases	GOTTFRIED POHLENTZ AND HEINZ EGGE	127
14. Ceramide Glycanase from the Leech <i>Macrobdella decora</i> and Oligosaccharide-Transferring Activity	YU-TEH LI AND SU-CHEN LI	146
15. Synthesis of Sialyl Lewis X Ganglioside and Analogs	AKIRA HASEGAWA AND MAKOTO KISO	158
16. Synthesis of Ganglioside G_{M3} and Analogs Containing Modified Sialic Acids and Ceramides	MAKOTO KISO AND AKIRA HASEGAWA	173
17. Synthesis of Ganglioside Analogs Containing Sulfur in Place of Oxygen at the Linkage Positions	HIDEHARU ISHIDA, MAKOTO KISO, AND AKIRA HASEGAWA	183
18. Replacement of Glycosphingolipid Ceramide Residues by Glycerolipid for Microtiter Plate Assays	RENÉ ROY, ANNA ROMANOWSKA, AND FREDERIK O. ANDERSSON	198
19. Neoglycolipids: Probes in Structure/Function Assignments to Oligosaccharides	T. FEIZI AND R. A. CHILDS	205

Section III. Synthetic Polymers

20. Use of Glycosylamines in Preparation of Oligosaccharide Polyacrylamide Copolymers	ELISABET KALLIN	221
---	-----------------	-----

21. Synthesis of Poly(<i>N</i> -acetyl- β -lactosaminide-carrying Acrylamide); Chemical-Enzymatic Hybrid Process	KAZUKIYO KOBAYASHI, TOSHIHIRO AKAIKE, AND TAICHI USUI	226
22. Preparation of Glycoprotein Models: Pendant-Type Oligosaccharide Polymers	SHIN-ICHIRO NISHIMURA, TETSUYA FURUIKE, AND KOJI MATSUOKA	235
23. Synthesis of Branched Polysaccharide by Chemical and Enzymatic Reactions and Its Hypoglycemic Activity	KENICHI HATANAKA	247
24. Tailor-made Glycopolymer Syntheses	F. D. TROPPER, A. ROMANOWSKA, AND R. ROY	257
25. Syntheses of Water-Soluble Polyacrylamide-Containing Sialic Acid	C. A. LAFERRIÈRE, F. O. ANDERSSON, AND R. ROY	271
26. Polymer-Supported Solution Synthesis of Oligosaccharides	JIRI J. KREPINSKY, STEPHEN P. DOUGLAS, AND DENNIS M. WHITFIELD	280
27. Syntheses of Clustered Lactosides by Telomerization	WILLIAM K. C. PARK, SIVASUBRAMANIAN ARAVIND, ANNA ROMANOWSKA, JOCELYN RENAUD, AND RENÉ ROY	294

VOLUME 243

INORGANIC MICROBIAL SULFUR METABOLISM

Harry D. Peck, Jr., and Jean LeGall

Section I. Sulfate Transport

1. Sulfate Transport	HERIBERT CYPIONKA	3
----------------------	-------------------	---

Section II. Dissimilatory Sulfate Reduction

A. Dehydrogenases: Hydrogenases from Sulfate-Reducing Bacteria

2. NAD-Dependent Alcohol Dehydrogenases from <i>Desulfovibrio gigas</i>	THEO A. HANSEN AND CHARLES M. H. HENSGENS	17
3. NAD(P)-Independent Lactate Dehydrogenase from Sulfate-Reducing Prokaryotes	THEO A. HANSEN	21
4. Aldehyde Oxidoreductases and Other Molybdenum-Containing Enzymes	JOSÉ J. G. MOURA AND BELARMINO A. S. BARATA	24
5. Nickel-Iron Hydrogenase	RICHARD CAMMACK, VICTOR M. FERNANDEZ LOPEZ, AND E. CLAUDE HATCHIKIAN	43
6. Nickel-Iron-Selenium Hydrogenase	DAULAT S. PATIL	68
7. The Pyruvic Acid Phosphoroclastic Reaction	LARRY L. BARTON	94

B. Electron Carrier Proteins from Sulfate-Reducing Bacteria

8. Monoheme Cytochromes	TATSUHIKO YAGI	104
9. Tetraheme Cytochromes	ISABEL B. COUTINHO AND ANTÓNIO V. XAVIER	119
10. Cytochrome c_3 (M_r 26,000) Isolated from Sulfate-Reducing Bacteria and Its Relationships to Other Polyhemic Cytochromes from <i>Desulfovibrio</i>	MIREILLE BRUSCHI	140

- | | | |
|--|---|-----|
| 11. Hexadecaheme Cytochrome <i>c</i> | YOSHIKI HIGUCHI, TATSUHIKO YAGI, AND GERRIT VOORDOUW | 155 |
| 12. Ferredoxins | JOSÉ J. G. MOURA, ANJOS L. MACEDO, AND P. NUNO PALMA | 165 |
| 13. Flavodoxins | JACQUES VERVOORT, DIRK HEERING, SJAAK PEELLEN,
AND WILLEM VAN BERKEL | 188 |
| 14. Rubredoxin in Crystalline State | LARRY C. SIEKER, RONALD E. STENKAMP, AND JEAN LE GALL | 203 |
| 15. Characterization of Three Proteins Containing Multiple Iron Sites: Rubrerythrin,
Desulfoferrodoxin, and a Protein Containing a Six-Iron Cluster | ISABEL MOURA, PEDRO TAVARES, AND NATARAJAN RAVI | 216 |

C. Terminal Reductases of Sulfate-Reducing Bacteria

- | | | |
|--|---|-----|
| 16. Adenylylsulfate Reductases from Sulfate-Reducing Bacteria | JORGE LAMPREIA, ALICE S. PEREIRA, AND JOSÉ J. G. MOURA | 241 |
| 17. Thiosulfate and Trithionate Reductases | J. M. AKAGI, H. L. DRAKE, JAE-HO KIM, AND DIANE GEVERTZ | 260 |
| 18. Desulforubidin: Dissimilatory, High-Spin Sulfite Reductase of <i>Desulfomicrobium</i>
Species | DANIEL V. DERVARTANIAN | 270 |
| 19. Desulfofusicidin: Dissimilatory, High-Spin Sulfite Reductase of Thermophilic, Sul-
fate-Reducing Bacteria | E. CLAUDE HATCHIKIAN | 276 |
| 20. Low-Spin Sulfite Reductases | ISABEL MOURA AND ANA ROSA LINO | 296 |
| 21. Hexaheme Nitrite Reductase from <i>Desulfovibrio desulfuricans</i> (ATCC 27774) | MING-CHEH LIU, CRISTINA COSTA, AND ISABEL MOURA | 303 |

D. Molecular Biology

- | | | |
|--|--|-----|
| 22. Genetic Manipulation of <i>Desulfovibrio</i> | WALTER M. A. M. VAN DONGEN, JACK P. W. G. STOKKERMANS,
AND WILLY A. M. VAN DEN BERG | 319 |
| 23. Enzymology and Molecular Biology of Sulfate Reduction in the Extremely Thermo-
philic Archeon <i>Archaeoglobus fulgidus</i> | CHRISTIANE DAHL, NORBERT SPEICH, AND HANS G. TRÜPER | 331 |

Section III. Dissimilatory Sulfur Reduction

- | | | |
|---|---|-----|
| 24. Sulfur Reductase from Thiophilic Sulfate-Reducing Bacteria | GUY D. FAUQUE | 353 |
| 25. Sulfur Reductases from Spirilloid Mesophilic Sulfur-Reducing Eubacteria | GUY D. FAUQUE, OLIVER KLIMMEK, AND ACHIM KRÖGER | 367 |

Section IV. Oxidation of Reduced Sulfur Compounds

- | | | |
|--|------------------------------------|-----|
| 26. Purification of Rusticyanin, a Blue Copper Protein from <i>Thiobacillus ferrooxidans</i> | JOHN W. INGLEDEW AND D. H. BOXER | 387 |
| 27. Adenylylsulfate Reductases from Thiobacilli | BARRIE F. TAYLOR | 393 |
| 28. Enzymes of Dissimilatory Sulfide Oxidation in Phototrophic Sulfur Bacteria | CHRISTIANE DAHL AND HANS G. TRÜPER | 400 |
| 29. Reverse Siroheme Sulfite Reductase from <i>Thiobacillus denitrificans</i> | HANS G. TRÜPER | 422 |

30. Purification and Properties of Cytochrome <i>c</i> -555 from Phototrophic Green Sulfur Bacteria	T. E. MEYER	426
31. Purification and Properties of High-Potential Iron-Sulfur Proteins	T. E. MEYER	435
32. Sulfite: Cytochrome <i>c</i> Oxidoreductase of Thiobacilli	ISAMU SUZUKI	447
33. Sulfur-Oxidizing Enzymes	ISAMU SUZUKI	455
34. Sulfide-Cytochrome <i>c</i> Reductase (Flavocytochrome <i>c</i>)	TATEO YAMANAKA	463

Section V. Metabolism of Polythionates

35. Synthesis and Determination of Thiosulfate and Polythionates	DON P. KELLY AND ANN WOOD	475
36. Enzymes Involved in the Microbiological Oxidation of Thiosulfate and Polythionates	DON P. KELLY AND ANN WOOD	501
37. Whole-Organism Methods for Inorganic Sulfur Oxidation by Chemolithotrophs and Photolithotrophs	DON P. KELLY AND ANN WOOD	510

Section VI. Special Techniques

38. Mössbauer Spectroscopy in Study of Cytochrome <i>cd</i> ₁ from <i>Thiobacillus denitrificans</i> , Desulfoviridin, and Iron Hydrogenase	BOI HANH HUYNH	523
39. <i>In Vivo</i> Nuclear Magnetic Resonance in Study of Physiology of Sulfate-Reducing Bacteria	HELENA SANTOS, PAULA FARELEIRA, JEAN LE GALL, AND ANTÔNIO V. XAVIER	543
40. Computational Chemistry and Molecular Modeling of Electron-Transfer Proteins	JOHN E. WAMPLER	559
41. Immunoassay of Sulfate-Reducing Bacteria in Environmental Samples	J. MARTIN ODOM AND RICHARD C. EBERSOLE	607

VOLUME 244

PROTEOLYTIC ENZYMES: SERINE AND CYSTEINE PEPTIDASES

Alan J. Barrett

1. Classification of Peptidases	ALAN J. BARRETT	1
---------------------------------	-----------------	---

Section I. Serine Peptidases

2. Families of Serine Peptidases	NEIL D. RAWLINGS AND ALAN J. BARRETT	19
3. Myeloblastin: Leukocyte Proteinase 3	JOHN R. HOIDAL, N. V. RAO, AND BEULAH GRAY	61
4. Granzyme A	MARKUS M. SIMON AND MICHAEL D. KRAMER	68
5. Granzyme B	MANUEL C. PEITSCH AND JUERG TSCHOPP	80

6. Trypsin: A Mast Cell Serine Protease	LAWRENCE B. SCHWARTZ	88
7. Hepsin	KOTOKU KURACHI, ADRIAN TORRES-ROSADO, AND AKIHIKO TSUII	100
8. Glutamyl Endopeptidases	JENS J. BIRKTOFT AND KLAUS BREDDAM	114
9. Lysyl Endopeptidase of <i>Achromobacter lyticus</i>	FUMIO SAKIYAMA AND TAKEHARU MASAKI	126
10. IgA-Specific Prolyl Endopeptidases: Serine Type	ANDREW G. PLAUT AND WILLIAM W. BACHOVCHIN	137
11. Biochemical and Genetic Methods for Analyzing Specificity and Activity of Precursor-Processing Enzyme: Yeast Kex2 Protease, Kexin	CHARLES BRENNER, ALISON BEVAN, AND ROBERT S. FULLER	152
12. Purification of Recombinant Soluble Forms of Furin Produced by Chinese Hamster Ovary Cells	KAZUHISA NAKAYAMA	167
13. Pro-Protein Convertases of Subtilisin/Kexin Family	NABIL G. SEIDAH AND MICHEL CHRÉTIEU	175
14. Prolyl Oligopeptidases	LÁSZLÓ POLGÁR	188
15. Oligopeptidase B: Protease II from <i>Escherichia coli</i>	DAISUKE TSURU AND TADASHI YOSHIMOTO	201
16. Dipeptidyl-peptidase IV from Rat Liver	YUKIO Ikehara, SHIGENORI OGATA, AND YOSHIO MISUMI	215
17. Acylaminoacyl-peptidase	WANDA M. JONES, ANDREA SCALONI, AND JAMES M. MANNING	227
18. Carboxypeptidases C and D	S. JAMES REMINGTON AND KLAUS BREDDAM	231
19. Serine-Type D-Ala-D-Ala Peptidases and Penicillin-binding Proteins	BENOÎT GRANIER, MARC JAMIN, MAGGY ADAM, MORENO GALLINI, BERNARD LAKAYE, WILLY ZORZI, JACQUELINE GRANDCHAMPS, JEAN-MARC WILKIN, CLAUDINE FRAIPONT, BERNARD JORIS, COLETTE DUEZ, MARTINE NGUYEN-DISTÈCHE, JACQUES COYETTE, MÉLINA LEYH-BOUILLE, JEAN DUSART, LÉON CHRISTIAENS, JEAN-MARIE FRÈRE, AND JEAN-MARIE GHUYSEN	249
20. Cleavage of LexA Repressor	JOHN W. LITTLE, BAEK KIM, KENNETH L. ROLAND, MARGARET H. SMITH, LIH-LING LIN, AND STEVE N. SILATY	266
21. Bacterial Leader Peptidase 1	WILLIAM R. TSCHANTZ AND ROSS E. DALBEY	285
22. Eukaryote Microsomal Signal Peptidases	MARK O. LIVELY, ANN L. NEWSOME, AND MOHAMAD NUSIER	301
23. Endopeptidase Clp: ATP-Dependent Clp Protease from <i>Escherichia coli</i>	MICHAEL R. MAURIZI, MARK W. THOMPSON, SATYENDRA K. SINGH, AND SEUNG-HO KIM	314
24. Multicatalytic Endopeptidase Complex: Proteasome	A. JENNIFER RIVETT, PETER J. SAVORY, AND HAKIM DJABALLAH	331
25. ATP-Dependent Protease La(Lon) from <i>Escherichia coli</i>	ALFRED L. GOLDBERG, RICHARD P. MOERSCHHELL, CHIN HA CHUNG, AND MICHAEL R. MAURIZI	350
26. Mitochondrial ATP-Dependent Protease from Rat Liver and Yeast	STEFAN KUZELA AND ALFRED L. GOLDBERG	376
27. OmpT: An <i>Escherichia coli</i> Outer Membrane Proteinase that Activates Plasminogen	WALTER F. MANGEL, DIANA L. TOLEDO, MARK T. BROWN, KIMBERLY WORZALLA, MIJIN LEE, AND JOHN J. DUNN	384
28. Transient Transfection Assay of the Herpesvirus Maturational Proteinase, Assemblin	WADE GIBSON, ANTHONY R. WELCH, AND JENNIFER LUDFORD	399

29. Purification and Kinetic Characterization of Human Cytomegalovirus Assemblin	MICHELE C. SMITH, JOANNA GIORDANO, JAMES A. COOK, MARK WAKULCHIK, ELCIRA C. VILLARREAL, GERALD W. BECKER, KERRY BEMIS, JEAN LABUS, AND JOSEPH S. MANETTA	412
30. Amino Acid and Peptide Phosphonate Derivatives as Specific Inhibitors of Serine Peptidases	JOZEF OLEKSYSYN AND JAMES C. POWERS	423
31. Isocoumarin Inhibitors of Serine Peptidases	JAMES C. POWERS AND CHIH-MIN KAM	442

Section II. Cysteine Peptidases

32. Families of Cysteine Peptidases	NEIL D. RAWLINGS AND ALAN J. BARRETT	461
33. Catalytic Mechanism in Papain Family of Cysteine Peptidases	ANDREW C. STORER AND ROBERT MÉNARD	486
34. Cathepsin S and Related Lysosomal Endopeptidases	HEIDRUN KIRSCHKE AND BERND WIEDERANDERS	500
35. Cysteine Endopeptidases of <i>Entamoeba histolytica</i>	HENNING SCHOLZE AND EGBERT TANNICH	512
36. Cysteine Endopeptidases of Parasitic Protozoa	MICHAEL J. NORTH	523
37. Glycyl Endopeptidase	DAVID J. BUTTLE	539
38. Pineapple Cysteine Endopeptidases	ANDREW D. ROWAN AND DAVID J. BUTTLE	555
39. Cancer Procoagulant	STUART G. GORDON	568
40. Picornains 2A and 3C	TIM SKERN AND HANS-DIETER LIEBIG	583
41. Adenovirus Endopeptidases	JOSEPH M. WEBER AND KAROLY TIHANYI	595
42. Legumain: Asparaginyl Endopeptidase	SHIN-ICHI ISHII	604
43. Interleukin-1 β Converting Enzyme	NANCY A. THORNBERRY	615
44. Isoprenylated Protein Endopeptidase	ROBERT R. RANDO AND YU-TING MA	632
45. Affinity Chromatography of Cysteine Peptidases	DAVID J. BUTTLE	639
46. Peptidyl Diazomethanes as Inhibitors of Cysteine and Serine Proteinases	ELLIOTT SHAW	649
47. Peptidyl (Acyloxy)methanes as Quiescent Affinity Labels for Cysteine Proteinases	ALLEN KRANTZ	656
48. N,O-Diacyl Hydroxamates as Selective and Irreversible Inhibitors of Cysteine Proteinases	DIETER BRÖMME AND HANS-ULRICH DEMUTH	671
49. Cystatins	MAGNUS ABRAHAMSON	685

VOLUME 245

EXTRACELLULAR MATRIX COMPONENTS

Erkki Ruoslahti and Eva Engvall

Section I. Extracellular Matrix Proteins

1. Nonfibrillar Collagens	NAOMI FUKAI, SUNEEL S. APTE, AND BJORN R. OLSEN	1
2. Fibrillin: Monomers and Microfibrils	LYNN Y. SAKAI AND DOUGLAS R. KEENE	29
3. Tenascins	SUSANNE SCHENK AND RUTH CHIQUET-EHRISMANN	52