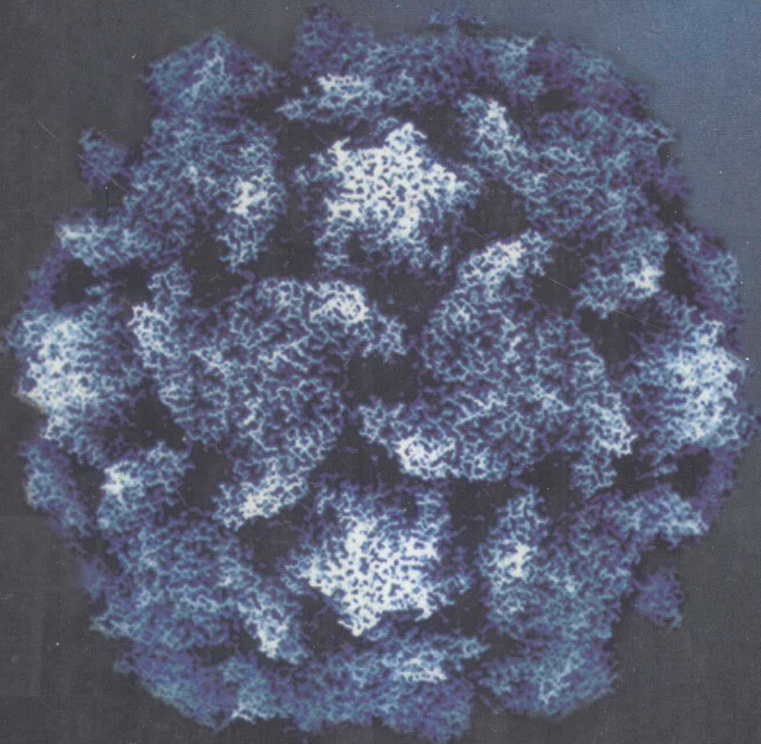

PRINCIPLES OF

VIROLOGY

Molecular Biology,
Pathogenesis, and Control



S. J. FLINT

L. W. ENQUIST

R. M. KRUG

V. R. RACANIELLO

A. M. SKALKA

VIROLOGY

Molecular Biology, Pathogenesis, and Control

S. J. Flint

Department of Molecular Biology
Princeton University
Princeton, New Jersey

L. W. Enquist

Department of Molecular Biology
Princeton University
Princeton, New Jersey

R. M. Krug

Institute for Cellular and
Molecular Biology
University of Texas at Austin
Austin, Texas

V. R. Racaniello

Department of Microbiology
College of Physicians & Surgeons
Columbia University
New York, New York

A. M. Skalka

Institute for Cancer Research
Fox Chase Cancer Center
Philadelphia, Pennsylvania



**ASM
PRESS**

Washington, D.C.

Front cover illustration: A model of the atomic structure of the poliovirus type 1 Mahoney strain. The model has been highlighted by radial depth cuing so that the portions of the model that are farthest from the center are bright. Prominent surface features include a star-shaped mesa at each of the fivefold axes and a propeller-shaped feature at each of the threefold axes. A deep cleft or canyon surrounds the star-shaped feature. This canyon is the receptor-binding site. Courtesy of Robert Grant, Stéphane Crainic, and James Hogle (Harvard Medical School).

Back cover illustration: Progress in the global eradication of poliomyelitis has been striking, as illustrated by maps showing areas of known or probable circulation of wild-type poliovirus in 1988 and 1998. Blue indicates either the presence of virus or insufficient surveillance to exclude the possibility of virus circulation. In 1988, virus was present on all continents except Australia. By 1998, the Americas were free of wild-type poliovirus, and transmission was interrupted in the western Pacific region (including the People's Republic of China) and in the European region (with the exception of southeastern Turkey). There are currently three major foci of transmission: southern Asia (Afghanistan, Pakistan, and India), West Africa (mainly Nigeria), and central Africa (mainly the Democratic Republic of Congo). Source: World Health Organization.

Copyright © 2000 ASM Press
American Society for Microbiology
1752 N Street NW
Washington, DC 20036

Library of Congress Cataloging-in-Publication Data

Principles of virology: molecular biology, pathogenesis, and control / S. J. Flint ... [et al.].

p. cm.

Includes bibliographical references and index.

ISBN 1-55581-127-2 (hardcover)

1. Viruses. 2. Virology. I. Flint, S. Jane.

[DNLM: 1. Viruses. 2. Virology—methods.

QW 160 P957 2000]

QR360.P697 2000

579.2—dc21

DNLM/DLC

for Library of Congress

99-14697

CIP

10 9 8 7 6 5 4 3 2

All Rights Reserved

Printed in the United States of America

Illustrations and illustration concepting: J/B Woolsey Associates

Cover and interior design: Susan Brown Schindler

*We dedicate this book to the students,
current and future scientists, and physicians
for whom it was written.
We kept them ever in mind.*

*We also dedicate it to our families:
Jonn, Gethyn, and Amy Leedham
Kathy and Brian
Marjorie, Dana, Nora, and Stephen
Doris, Aidan, Devin, and Nadia
Rudy, Jeanne, and Chris*

*Oh, be wiser thou!
Instructed that true knowledge leads to love.*

WILLIAM WORDSWORTH
Lines Left upon a Seat in a Yew-Tree
1888

Acknowledgments

This book could not have been completed without help and contributions from many individuals. Indeed, this venture would not have begun were it not for the initial encouragement we received from our colleagues in virology. We are especially indebted to Patrick Fitzgerald (formerly Director, ASM Press) for his enthusiastic support when for the project was little more than a rough plan and for his sound advice during its development. The creative energy and ideas of Priscilla Schaffer (University of Pennsylvania) had a substantial impact on the initial planning. Our sincere thanks also go to our colleagues who took considerable time and effort to review this text in its evolving manifestations. Their expert knowledge and advice on issues ranging from teaching virology to organization of individual chapters and style were invaluable, even when orthogonal to our approach, and are inextricably woven into the final form of the book.

We thank the following individuals for their reviews and comments on multiple chapters or the entire book: Nicholas Acheson (McGill University), Clifford W. Bond (Montana State University), Martha Brown (University of Toronto Medical School), Teresa Compton (University of Wisconsin), Stephen Dewhurst (University of Rochester Medical Center), Mary K. Estes (Baylor College of Medicine), Ronald Javier (Baylor College of Medicine), Richard Kuhn (Purdue University), Muriel Lederman (Virginia Polytechnic Institute and State University), Richard Moyer (University of Florida College of Medicine), Leonard Norkin (University of Massachusetts), Martin Petric (University of Toronto Medical School), Marie Pizzorno (Bucknell University), Nancy Roseman (Williams College), David Sanders (Purdue University), Dorothea Sawicki (Medical College of Ohio), Bert Semler (University of California, Irvine), and Bill Sugden (University of Wisconsin).

We also are very grateful to those who gave so generously of their time to serve as expert reviewers of individual chapters or topics: Edward Arnold (Center for Advanced Biotechnology and Medicine, Rutgers University),

Albert Bendelac (Princeton University), Susan Berget (Baylor College of Medicine), Jim Broach (Princeton University), Michael J. Buchmeier (The Scripps Research Institute), Bruce Chesebro (Rocky Mountain Laboratories, National Institute of Allergy and Infectious Diseases), Marie Chow (University of Arkansas Medical Center), Michael Cordingly (Bio-Mega/Boehringer Ingelheim), Ted Cox (Princeton University), Robert Doms (University of Pennsylvania), Emilio Emini (Merck Sharp & Dohme Research Laboratories), Charles Grose (Iowa University Hospital), Peter Howley (Harvard Medical School), James Hoxie (University of Pennsylvania), Frederick Hughson (Princeton University), Tom Kelly (Johns Hopkins University School of Medicine), Elliott Kieff (Harvard Medical School), Arnold Levine (The Rockefeller University), Michael Malim (University of Pennsylvania), William Mason (Fox Chase Cancer Center), Bernard Moss (National Institutes of Health), Roger Pomerantz (Thomas Jefferson University), Glenn Rall (Fox Chase Cancer Center), Barry Rouse (University of Tennessee College of Veterinary Medicine), Priscilla Schaffer (University of Pennsylvania), Robert Schneider (New York University School of Medicine), Christoph Seeger (Fox Chase Cancer Center), Aaron Shatkin (Center for Advanced Biotechnology and Medicine, Rutgers University), Thomas Shenk (Princeton University), John M. Taylor (Fox Chase Cancer Center), Alice Telesnitsky (University of Michigan Medical School), Peter Vogt (The Scripps Research Institute), M. Gerard Waters (Princeton University), Robin Weiss (Institute of Cancer Research, Chester Beatty Laboratories), Lindsay Whitton (The Scripps Research Institute), and Eckard Wimmer (State University of New York at Stony Brook). Their rapid responses to our requests for details and checks on accuracy, as well as their assistance in simplifying complex concepts, were invaluable. All remaining errors or inconsistencies are entirely ours.

We thank our colleagues who provided expert advice on specific topics or information prior to publication: James Alwine (University of Pennsylvania), Carl Baker (National Institutes of Health), Amiya Banerjee (Cleveland Clinic Foundation), Silvia Barabino (University of Basel), Barklie Clements (University of Glasgow), Don Coen (Harvard Medical School), Ron Desrosiers (Harvard Medical School), Bert Flanagan (University of Florida), Samuel Gunderson (European Molecular Biology Laboratory), Walter Keller (University of Basel), Ihor Lemischka (Princeton University), Michael Malim (University of Pennsylvania), James Manley (Columbia University), William Mason (Fox Chase Cancer Center), Bernard Moss (National Institutes of Health), Peter O'Hare (Marie Curie Research Institute), Radhakris Padmanabhan (University of Kansas Medical Center), Stuart Peltz (University of Medicine and Dentistry of New Jersey—Robert Wood Johnson Medical School), Jack Rose (Yale University), Rozanne Sandri-Goldin (University of California, Irvine), Bert Semler (University of California, Irvine), Thomas Shenk (Princeton University), Joan Steitz (Yale University), Wesley Sundquist (University of Utah), Heinz-Jürgen Thiel (Institut für Virologie, Giessen, Germany), Sandra Weller (University of Connecticut Health Center), and Eckard Wimmer (State University of New York at Stony Brook).

Since the inception of this work, we have believed that the illustrations must complement and enrich the text. Execution of this plan would have not been possible without the support of Jeff Holtmeier (Director, ASM Press) and the technical expertise and craft of our illustrators, J/B Woolsey Associates. The illustrations are an integral part of the exposition of the information and ideas discussed, and credit for their execution goes to the knowledge, insight, and en-

thusiasm of Patrick Lane of J/B Woolsey Associates. Many of the figures could not have been completed without the help and generosity of our many colleagues (mentioned in the figure legends) who provided original images. Special thanks go out to those who crafted figures tailored specifically to our needs or provided multiple pieces: Mark Andrade (Fox Chase Cancer Center), Edward Arnold (Rutgers University), Bruce Banfield (Princeton University), Christopher Basler and Peter Palese (Mount Sinai School of Medicine), Amy Brideau (Princeton University), Roger Burnett (Wistar Institute), Rajiv Chopra and Stephen Harrison (Harvard University), Marie Chow (University of Arkansas Medical Center), Richard Compans (Emory University), Friedrich Frischknecht (European Molecular Biology Laboratory), Wade Gibson (Johns Hopkins University School of Medicine), Ramon Gonzalez (Princeton University), Thomas Leitner (Los Alamos National Laboratory), Paul Masters (New York State Department of Health), Rolf Menzel (Small Molecule Therapeutics), B. V. Venkataram Prasad (Baylor College of Medicine), Wesley Sundquist (University of Utah), Jose Varghese (Commonwealth Scientific and Industrial Research Organisation), Robert Webster (St. Jude Children's Research Hospital), Thomas Wilk (European Molecular Biology Laboratory), and Alexander Wlodawer (National Cancer Institute).

We thank all those who guided and assisted in the preparation and production of the book: Jeff Holtmeier (Director, ASM Press) for steering us through the complexities inherent in a team effort, Ken April (Production Editor, ASM Press) for keeping us on track during production, Greg Payne (Senior Editor, ASM Press) for organizing the reviews of the text, and Susan Schmidler (Susan Schmidler Graphic Design) for her elegant and creative designs for the layout and cover. We are also grateful for expert secretarial and administrative support from Norma Caputo (Princeton University), Helen Pirrello (Rutgers University), Josephina de la Rosa (Columbia University), and Mary Estes and Rose Walsh (Fox Chase Cancer Center) that facilitated preparation of this text, and we thank Amy Leedham for assistance with editing.

We owe a very special debt of gratitude to the Rockefeller Foundation for the award of a residency for our team at the Bellagio Study and Conference Center, Bellagio, Italy, in June 1997. The extraordinary beauty of the Center and the gracious hospitality of its staff made that an unforgettable as well as rewarding experience. The unique opportunity for study and extended consultation with each other and for interaction with resident scholars from many countries and disciplines had a major impact on the delineation of the purpose, approach, and scope of this textbook. ASM Press generously provided the financial support for travel to Bellagio as well as for our many meetings.

This often consuming enterprise was made possible by the emotional, intellectual, and logistical support of our families, to whom the book is dedicated.

Our acknowledgments close with a special tribute to the person who has done more than any other to support the author team, Trisha Barney (Princeton University), who has carried responsibilities too numerous to list since the very beginning of the project. However, we are especially grateful for her patience and good humor and particularly for collecting all the permissions necessary for preparation of the figures. Trisha's dedication, attention to detail, and problem-solving skills were critically important to our progress.

Organization of This Book

The enduring goal of scientific endeavor, as of all human enterprise, I imagine, is to achieve an intelligible view of the universe. One of the great discoveries of modern science is that its goal cannot be achieved piecemeal, certainly not by the accumulation of facts. To understand a phenomenon is to understand a category of phenomena or it is nothing. Understanding is reached through creative acts.

A. D. HERSHEY
Carnegie Institution Yearbook 65

The major goal of this book is to illustrate why and how animal viruses are studied. In so doing, we highlight unifying principles that emerge from these studies that may be used for investigation of questions still outstanding and of viral systems about which our knowledge is still quite limited. We have been selective in our choice of topics, viruses, and examples. The purpose was not to be comprehensive, for it is not possible for one reasonably sized text to cover every facet of animal virology. *Fields Virology* (1996), in two encyclopedic volumes, has made the best attempt to be all-inclusive and is recommended as a resource for detailed reviews of specific virus families.

For purposes of readability, references are generally omitted from the text, but each chapter ends with a list of relevant books, review articles, and selected research papers, for readers who wish to pursue specific topics. If an experiment is featured in a chapter, one or more references will be listed to provide more detailed information. In many cases, the figures and their legends also provide important detail whose inclusion in the text might interrupt the flow of thought. Text boxes include basic background information (boxes with aqua and pale green backgrounds); detailed discussions of specific topics (purple and greenish beige); definitions of terms, explanations of nomenclature, and clarifications of common sources of confusion in terminology (maroon and gray); discussions of specific experiments (blue and yellow); and cautionary information (orange). The text itself follows the general strategy

used by all viruses and covers the principles of critical steps in virus replication in the following sections.

Part I: The Science of Virology

Chapter 1 provides a general introduction to the field with historical perspectives and a discussion of how viruses are classified. It summarizes the unifying principles that are the foundations of virology. In chapter 2, we describe general methods by which animal viruses are studied in the laboratory as an entrée into the molecular biology section.

Part II: Molecular Biology

The next section, consisting of chapters 3 to 13, focuses on molecular processes that take place in an infected host cell. Chapter 3 provides a description of the extracellular virus particle (a virion). Virion architecture is discussed in the context of providing both protection and delivery of the viral genome in a single vehicle. Chapters 4 through 13 describe the broad spectrum of molecular processes that characterize the common steps of the reproductive cycle of viruses in a single cell, from decoding genetic information to genome replication. The intention is to describe how these common steps are accomplished by diverse but representative viruses, while illuminating principles applicable to all.

Part III: Pathogenesis and Control

This section moves away from events in the single cell and addresses issues related to the interplay between viruses and their host organisms. Chapter 14 provides a detailed description of the components of the immune system and strategies that viruses have evolved for coping with it, while chapter 15 describes the different types of relationships (some short and others of long duration) that viruses can establish with their hosts. Chapter 16 describes the roles of viruses in cell transformation and in oncogenesis, the formation of tumors. As viruses are of paramount importance as disease agents, chapters 17 to 19 address issues of pathogenesis and the principles involved in developing methods of treatment and control of infection. An entire chapter (chapter 18) is devoted to the AIDS virus, not only because it is the causative agent of a serious, worldwide epidemic but also because of its unique and informative interactions with the human immune defenses. Chapter 20, the final chapter, is a foray into the future and a consideration of viral evolution, the ways in which new viruses emerge, and the overall implications for virus and host survival.

Appendix

The appendix provides a brief description and a general scheme for the reproductive cycles in single cells of viruses that serve as important examples in several chapters of the text. This section is intended to be a reference resource when one reads individual chapters and a convenient means by which specific topics may be related to the overall infectious cycles of the selected viruses. On the opening page of each chapter, therefore, is a set of icons, which are based on the diagrams of virus structures in the appendix and which represent the selected viruses described in that chapter.

Reference

Fields, B. N., D. M. Knipe, and P. M. Howley (ed. in chief). 1996. *Fields Virology*, 3rd ed. Raven Press, New York, N.Y.

Contents

Acknowledgments xix

Organization of This Book xxiii

I	The Science of Virology	1
I	Foundations of Virology	2
	Luria's Credo	3
	Virus Prehistory	3
	Viral Infections in Antiquity	3
	The First Vaccines	4
	Microorganisms as Pathogenic Agents	6
	The Discovery of Viruses	8
	The First Viruses	8
	Identification of Human Viruses	10
	The Definitive Properties of Viruses	11
	The Structural Simplicity of Viruses	11
	The Intracellular Parasitism of Viruses	11
	Viruses Defined	15
	Cataloging of Diverse Animal Viruses	16
	Many Sizes and Shapes Produced by Evolution	16
	Genetic Content	19
	Capsid Symmetry	19
	Enveloped or Naked Particles	20
	Virion Enzymes	20
	Unifying Principles	20
	A Common Strategy	20
	Highlights of Virus Strategies Outlined in This Book	21
	References	21

2 Virus Cultivation, Detection, and Genetics 24
Introduction 25
Cultivation of Viruses 25
Cell Culture 25
Embryonated Eggs 27
Laboratory Animals 28
Detection of Viruses in the Host 29
Measurement of Infectious Units 29
Measurement of Virus Particles and Their Components 33
The One-Step Growth Cycle 40
Genetic Analysis of Viruses 42
Classical Genetic Methods 42
Engineering Mutations into Viruses 44
Engineering Viral Genomes: Viral Vectors 51
Perspectives 55
References 55

***II* Molecular Biology 57**

3 Virus Structure 58
Introduction 59
Functions of the Virion 59
Nomenclature 60
Methods for Studying Virus Structure 62
Building a Protective Coat 65
Helical Structures 66
Capsids or Nucleocapsids with Icosahedral Symmetry 66
Complex Viruses 84
Packaging the Nucleic Acid Genome 87
Direct Contact of the Genome with Proteins of the Outer Shell 87
Packaging by Specialized Virion Proteins 88
Packaging by Cellular Proteins 88
Viruses with Envelopes 89
Viral Envelope Components 90
Simple Enveloped Viruses: Direct Contact of External Proteins with the Capsid or Nucleocapsid 92
Enveloped Viruses with Additional Protein Layers 92
Other Components of Virions 94
Virion Enzymes 94
Other Viral Proteins 95
Cellular Macromolecules 96
Perspectives 96
References 97

4 Virus Attachment to Host Cells 100

Introduction 101

Protection and Release of Genetic Material 102

Finding the Right Cells To Infect 102

The Architecture of Cell Surfaces 102

Extracellular Matrix: Components and Biological Importance 103

Properties of the Plasma Membrane 103

Cell Membrane Proteins 105

Interaction of Viruses with Cell Receptors 106

General Principles 106

Identification of Viral Receptors on Cells 112

How Virions Attach to Receptors 123

Perspectives 128

References 129

5 Virus Entry into Cells 132

Introduction 133

Uptake of Macromolecules by Cells 134

The Plasma Membrane Is a Semipermeable Barrier 134

Cells Take Up Macromolecules by Phagocytosis and Endocytosis 134

Cell Membranes Fuse during Endocytosis 136

Mechanisms of Virus Entry into Cells 137

Uncoating at the Plasma Membrane 137

Virus Entry via the Endocytic Pathway 142

Entry of Nonenveloped Viruses into Cells 147

Cellular Molecules That Play Roles in Virus Uncoating 151

Transport of the Viral Genome into the Nucleus 152

The Cellular Pathway of Protein Import into the Nucleus 152

Import of DNA Genomes 156

Import of the Retroviral Genome into the Nucleus 157

Import of Influenza Virus RNP into the Cell Nucleus 159

Perspectives 159

References 160

6 Genome Replication and mRNA Production by RNA Viruses 162

Introduction 163

Mechanisms of Viral RNA Synthesis 164

RNA-Dependent RNA Polymerases 164

Accessory Proteins in RNA-Dependent RNA Synthesis 168

The Nature of the RNA Template 172

Initiation and Elongation 174

Cellular Sites of Viral RNA Synthesis 183

Unique Mechanisms of mRNA and Genome Synthesis of the Hepatitis Delta Satellite Virus 184

The Switch from mRNA Production to Genome RNA Synthesis 185

- Regulation of Initiation: Different Polymerase Specificities 187
- Suppression of Intergenic Stop-Start Reactions by Nucleocapsid Protein 188
- Antitermination and Different RNA Polymerases 189
- Suppression of Termination Caused by a Stem-Loop Structure 189
- Different Templates Used for mRNA Synthesis and Genome Replication 189
- Suppression of Polyadenylation 190

Origins of Diversity in RNA Viruses 190

- Misincorporation of Nucleotides 190
- Segment Reassortment and RNA Recombination 191
- RNA Recombination Leading to the Production of Pathogenic Viruses 192
- RNA Recombination Leading to the Production of Defective-Interfering RNAs 192
- RNA Editing 193

Perspectives 194

References 194

7 Reverse Transcription and Integration: Hallmarks of the Retroviral Viruses 198

Retroviral Reverse Transcription 199

- Introduction 199
- The Pathway of Retroviral Reverse Transcription 200
- General Properties and Structure of Retroviral Reverse Transcriptases 207
- Other Examples of Reverse Transcription 212

Retroviral DNA Integration 214

- Introduction 214
- Pathway of Retroviral Integration 214
- Integrase Structure and Mechanism 219

Hepadnaviral Reverse Transcription 224

- Introduction 224
- Pathway of Reverse Transcription 225

Perspectives 232

- Comparison of Systems 232

References 233

8 Transcription Strategies: DNA Templates 236

Introduction 237

Properties of Cellular and Viral Transcription Systems 238

- Cellular Transcription Systems Used by Viruses 238
- Production of Viral DNA Templates Transcribed by Cellular RNA Polymerases 238
- Transcription by RNA Polymerase II 240

Transcription of Viral DNA Templates by the Cellular Machinery Alone 252

Viral Proteins That Regulate Transcription by RNA Polymerase II 253

- Patterns of Regulation by Viral Proteins 253
- Autoregulation by the Human Immunodeficiency Virus Type 1 Tat Protein 253
- The Transcriptional Cascades of DNA Viruses 261

A Viral DNA-Dependent RNA Polymerase 277**Transcription of Viral Genes by RNA Polymerase III 278**

- Transcription of the Adenoviral VA-RNA Genes 279

Virus-Specific Applications of Cellular Transcription Components 280**Inhibition of the Cellular Transcription Machinery in Virus-Infected Cells 281****Perspectives 281****References 282****9 Genome Replication Strategies: DNA Viruses 286****Introduction 287****DNA Synthesis by the Cellular Replication Machinery: Lessons from Simian Virus 40 289**

- Eukaryotic Replicons 289
- Cellular Replication Proteins and Their Functions during Simian Virus 40 DNA Synthesis 292

Mechanisms of Viral DNA Synthesis 299

- Priming and Elongation 299
- Viral Replication Origins and Their Recognition 303
- Viral DNA Synthesis Machinery 311
- Resolution and Processing of Viral Replication Products 313

Mechanisms of Exponential Viral DNA Replication 315

- Induction of Synthesis of Cellular Replication Proteins by Viral Gene Products 315
- Synthesis of Large Sets of Viral Replication Proteins 318
- Viral DNA Replication Independent of Cellular Proteins 318
- Delayed Synthesis of Virion Structural Proteins 318
- Inhibition of Cellular DNA Synthesis 319
- Localization of Replicating Viral DNA to Specialized Intracellular Sites 319

Limited Replication of Viral DNA 320

- Replication as Part of the Cellular Genome: Integrated Parvoviral DNA 320
- Regulation of Replication via Different Viral Origins: Epstein-Barr Virus 321
- Controlled and Exponential Replication from a Single Origin: the Papillomaviruses 322

Origins of Genetic Diversity in DNA Viruses 323

- Fidelity of Replication by Viral DNA Polymerases 323
- Recombination of Viral Genomes 324

Perspectives 326**References 327**

10 Processing of Viral Pre-mRNA 330

Introduction 331

Covalent Modification during Viral Pre-mRNA Processing 333

Capping the 5' Ends of Viral mRNA 333

Synthesis of 3' Poly(A) Segments of Viral mRNA 337

Splicing of Viral Pre-mRNA 339

Alternative Splicing of Viral Pre-mRNA 345

Interactions Between Polyadenylation and Splicing in Viral Gene Expression 348

Editing of Viral mRNAs 351

Export of Viral and Cellular RNAs from the Nucleus 351

The Cellular Export Machinery 352

Export of Viral mRNA 353

Posttranscriptional Regulation of Viral or Cellular Gene Expression by Viral Proteins 358

Temporal Control of Viral Gene Expression 358

Inhibition of Cellular mRNA Production by Viral Proteins 361

Regulation of Turnover of Viral and Cellular mRNAs in the Cytoplasm 363

Regulation of mRNA Stability by a Viral Protein 363

Regulation of mRNA Stability in Transformation 364

Perspectives 365

References 366

11 Translational Control of Viral Gene Expression 370

Introduction 371

Mechanisms of Eukaryotic Protein Synthesis 372

General Structure of Eukaryotic mRNA 372

The Translational Machinery 372

Initiation 373

Elongation and Termination 383

The Diversity of Viral Translation Strategies 384

Polyprotein Synthesis 384

Leaky Scanning 386

Reinitiation 388

Suppression of Termination 388

Ribosomal Frameshifting 390

Regulation of Translation during Viral Infection 391

Inhibition of Translation as a Cellular Defense against Viral Infection 392

Regulation of eIF4F Activity 395

Perspectives 398

References 399

12 Intracellular Transport of Viral Components:

Prelude to Assembly 402

Introduction 403

Assembly within the Nucleus 404

Import of Viral Proteins for Assembly 404

Assembly at the Plasma Membrane 406

The Cellular Pathway by Which Viral Membrane Proteins Are Transported to the Plasma Membrane 406

Viral Proteins in the Secretory Pathway 418

Sorting of Viral Proteins in Polarized Cells 421

Inhibition of Transport of Cellular Proteins by Viral Infection 424

Signal Sequence-Independent Transport of Viral Proteins to the Plasma Membrane 425

Interactions with Internal Cellular Membranes 429

Localization of Viral Proteins to Compartments of the Secretory Pathway 429

Localization of Viral Proteins to the Nuclear Membrane 430

Transport of Viral Genomes to Assembly Sites 431

Genome RNA Transport from the Nucleus to the Cytoplasm 432

RNA Transport from the Cytoplasm to the Plasma Membrane 432

Perspectives 433**References 434****13 Assembly, Exit, and Maturation of Progeny Virions 438****Introduction 439****Methods of Studying Virus Assembly and Egress 440**

Structural Studies of Virus Particles 440

Visualization of Assembly and Exit by Electron Microscopy 441

Biochemical and Genetic Analysis of Assembly Intermediates 441

Methods Based on Recombinant DNA Technology 443

Assembly of Protein Shells 443

Formation of Structural Units 443

Capsid and Nucleocapsid Assembly 446

Self-Assembly and Assisted Assembly Reactions 448

Selective Packaging of the Viral Genome and Other Virion Components 452

Concerted or Sequential Assembly 452

Recognition and Packaging of the Nucleic Acid Genome 452

Incorporation of Virion Enzymes and Other

Nonstructural Proteins 460

Acquisition of an Envelope 460

Sequential Assembly of Internal Components and Budding from a Cellular Membrane 460

Coordination of the Assembly of Internal Structures with the Acquisition of the Envelope 461

Release of Virus Particles 463

Release of Nonenveloped Viruses 463

Assembly at Internal Membranes: the Problem of Exocytosis 464

Maturation of Progeny Virions 467
 Proteolytic Processing of Virion Proteins 467
 Other Maturation Reactions 470
Cell-to-Cell Spread 472
Perspectives 473
References 473

III

Pathogenesis and Control 477

I4 Virus Offense Meets Host Defense 478

The Host Defense Against Viral Infections 479

- Primary Defenses 479
- Immune Defenses 480

The Innate Immune Response 484

- Cytokines 484
- Interferons 485
- Complement 489
- Collectins 492
- NK Cells 492

The Adaptive Immune Response 493

- General Features 493
- Cells of the Adaptive Immune System 496
- Antigen Presentation and Activation of Immune Cells 502
- The Cell-Mediated Adaptive Response 506
- The Antibody Response 508

Perspectives 511

References 515

I5 Patterns of Infection: a Delicate Balance 518

Introduction 519

- Infection Strategies 519
- Life Cycles and Patterns of Viral Infections 519

Initiating an Infection 520

- Basic Requirements 520
- Tropism 521
- Successful Infections Must Evade Host Defenses 521
- Many Other Variables Govern the Result of Infection 531

Acute Infections 531

- Definition and Requirements 531
- Acute Infections Present Common Public Health Problems 533
- Defense against Acute Infections 534
- Multiple Acute Infections in a Single Host 534
- Pathogenic Effects of an Acute Infection 535