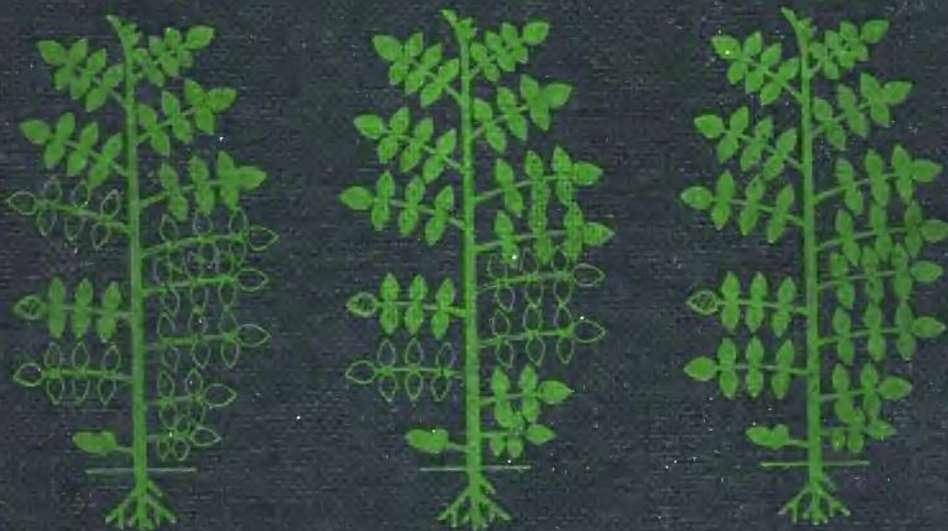


# Plant Virology

---

Third Edition



R. E. F. Matthews

# PLANT VIROLOGY

*Third Edition*

**R. E. F. Matthews**


*Department of Cellular and Molecular Biology  
The University of Auckland  
Auckland, New Zealand*



ACADEMIC PRESS, INC.

*Harcourt Brace Jovanovich, Publishers*

San Diego New York Boston London  
Sydney Tokyo Toronto

This book is printed on acid-free paper. 

Copyright © 1991, 1981, 1970 by Academic Press, Inc.

All Rights Reserved.

No part of this publication may be reproduced or transmitted in any form or by any means, electronic or mechanical, including photocopy, recording, or any information storage and retrieval system, without permission in writing from the publisher.

Academic Press, Inc.

San Diego, California 92101

United Kingdom Edition published by

Academic Press Limited

24-28 Oval Road, London NW1 7DX

Library of Congress Cataloging-in-Publication Data

Matthews, R. E. F. (Richard Ellis Ford), Date.

Plant virology / R.E.F. Matthews. -- 3rd ed.

p. cm.

Includes index.

ISBN 0-12-480553-1 (alk. paper)

1. Plant viruses. 2. Virus diseases of plants. 3. Plant viruses--  
-Control. I. Title.

3B736.M37 1991

632'.8--dc20

90-40409

CIP

Printed in the United States of America

91 92 93 94 9 8 7 6 5 4 3 2 1

---

## Preface

In the 10 years since the second edition was written there have been major developments in the field of plant virology. Most of these developments stem from the application of gene manipulation technology. Two groups of plant viruses have DNA genomes, but the great majority contain RNA. Thus the major influence in the 1980s has been the development of procedures whereby DNA copies of RNA genomes can be made and DNA sequencing techniques applied to them. Complete nucleotide sequences are now known for representatives of over half the groups of plant viruses with RNA genomes. The ability to prepare *in vitro* infectious RNA transcripts of RNA viral genomes from cloned viral cDNA has been of particular importance. For example it has allowed techniques such as site-directed mutagenesis to be applied to the study of genome function. Nucleotide sequence information has had, and continues to have a profound effect on our understanding of many aspects of plant virology. We now know the number, location, size and functions of the genes in many representative viruses.

The ability to introduce DNA copies of single viral genes into the genome of host plants has opened up new possibilities for the control of some important viral diseases. Methods for the assay and detection of viruses and the diagnosis of diseases have improved greatly in sensitivity, specificity and convenience. These advances have been due mainly to advances in serological procedures and in the application of nucleic acid hybridization technology.

The deluge of nucleotide sequence information becoming available opens up the possibility that we can develop a classification system for viruses based on their evolutionary relationships. This sequence information emphasizes more than ever the essential unity that exists between viruses infecting all groups of organisms.

These various developments have led to a very substantial rewriting of most chapters in the text. Some are entirely new. The extent of the changes can be gauged from the following data. Approximately 52% of the written text is new. Much of the remainder has been revised; of the 251 illustrations, 126 are new; of the 21 tables, 20 are new; and approximately 2,000 of the 3,000 references in the bibliography were not in the second edition.

While much of the older material has been deleted for reasons of space, I have

retained many older references to interesting but unexplained biological phenomena, because the technologies for exploring and understanding many of these at the molecular level are now becoming available.

As did the first edition, this volume covers all aspects of the subject from molecular to ecological. The bases for experimental procedures are discussed, but detailed protocols are not provided. The volume is intended primarily for graduate students in plant virology, plant pathology, general virology and microbiology, and for teachers and research workers in these fields. It should also prove useful to some people in related fields such as molecular biologists, biochemists, plant physiologists and entomologists.

I am much indebted to the following colleagues who critically read and commented on sections of the manuscript: P. J. G. Butler, T. W. Dreher, the late R. I. B. Francki, R. L. S. Forster, R. C. Gardner, J. Marbrook, E. Mayr, B. A. Morris, L. R. Nault, D. L. Nuss, Y. Okada, R. A. Owens, D. Penny, A. W. Robards, D. D. Shukla, R. H. Symons, J. M. Thresh, and M. Zaitlin. I thank the following for most useful discussions: J. Berriman, R. I. B. Francki, R. C. Gardner, J. M. Kaper, D. Lane, D. L. Nuss, and J. W. Randles. I also thank the many colleagues in various countries who provided information by correspondence, who sent manuscripts prior to publication, and who provided photographs for illustrations. Figures are acknowledged individually in the text. I also thank editors and publishers for permission to reproduce figures and photographs. I thank M. Gibbs for preparing computer-generated figures; P. R. Fry for help with the bibliography, and last, but not least, Jean Parrott for typing the manuscript.

---

## *Preface to the Second Edition*

There have been substantial developments in many areas of plant virology since the first edition was published.

Advances have been made in all branches of the subject, but these have been most far reaching with respect to the structure of viruses and of their components, and in our understanding of how viral genomes are organized and how viruses replicate in cells. Significant developments have also occurred in our understanding of how viruses are transmitted by invertebrates and in the application of control measures for specific diseases. The taxonomy of viruses has advanced significantly, and there are now 25 internationally approved families and groups of plant viruses. All these developments have required that most sections be entirely rewritten. The extent of the changes can be gauged from the fact that 1881 of the 2667 references in the bibliography did not appear in the first edition.

As did the first edition, this volume is written to cover all aspects of the field, and is intended primarily for graduate students in plant pathology, plant virology, general virology, and microbiology, and for teachers and research workers in these fields. It should also prove useful to some people in related disciplines—molecular biologists, biochemists, plant physiologists, and entomologists.

---

## *Preface to the First Edition*

As in many other areas of biology, there has been rapid growth over the past few years in our knowledge of plant viruses and the diseases they cause. Thus there was a substantial need for a new text covering all aspects of the subject.

This book was written primarily for graduate students in plant pathology, plant virology, general virology, and microbiology and for teachers and research workers in these fields. I hope that it will also prove useful as a reference work for those in disciplines related to plant virology—molecular biologists, biochemists, plant physiologists, and entomologists.

I have attempted to cover, to some degree at least, all aspects of the subject, a difficult task in view of the wide range of disciplines involved. There is a brief historical account of the development of plant virology in the first chapter, but the general approach is not a historical one. Those interested will find this aspect well covered in earlier texts.

Topics dealt with include the structure of viruses and viral components; the replication of viruses; their macroscopic, cytological, and biochemical effects on the host plant; the nature of virus mutation; relationships with invertebrate vectors; and a discussion of ecology and control. Throughout I have attempted to indicate how progress in any particular area has been dependent on the development and application of appropriate experimental methods. Specific details of methodology have not been given since these are available elsewhere.

The subject has grown to the extent that it would be impossible to quote all papers on any given topic in a book of this size. In general I have referred to important early papers and to the most important or most suitably illustrative recent papers. From these the reader should be able to gain rapid access to the literature on any relevant topic.

In a text on a subject that draws a wide range of scientific disciplines, I believe that illustrative material is most important, particularly for students or newcomers to the field. For this reason I have gone to some pains, and have had the support of many colleagues, in selecting graphs and photographs to highlight and supplement the text.

In certain areas, particularly the molecular biology of viral replication, our knowledge of plant viruses lags behind that of animal and bacterial viruses. I have

therefore drawn on information about these viruses where it seemed appropriate to set the stage for considering more fragmentary facts about plant viruses.

One recent development that created problems was the discovery that many diseases previously thought to be caused by unstable viruses are very probably caused by mycoplasma-like organisms. Although, in general, I have not included diseases in which the probability of a mycoplasma-like organism being involved is high, one chapter on agents causing virus-like diseases is devoted mainly to a consideration of such organisms in plant disease. Other recent work of considerable general interest has resulted in the discovery that several plant viruses have their genetic material divided up between two or more particles. Thus I have devoted a chapter to the consideration of defective virus particles, dependent viruses, and multiparticle viruses.

I have followed the Commonwealth Mycological Institute list of "Plant Virus Names" (Martyn, 1968). I have not attempted to deal with individual viruses or virus diseases in any systematic or comprehensive way, so that the list of "Plant Virus Names" should be regarded as a valuable companion book for the present text, especially for those interested in the tremendous amount of literature on the plant pathological aspects of virus diseases.

In the last chapter I have outlined the various viewpoints regarding nomenclature and classification. Since, from the long-term point of view, at least, classification of viruses must take origins into consideration, some space is given to speculation on the origins of viruses.



---

# Contents

<i>Preface</i> .....	xvii
<i>Preface to the Second Edition</i> .....	xix
<i>Preface to the First Edition</i> .....	xxi
<i>1 Introduction</i>	
I. HISTORICAL .....	1
II. DEFINITION OF A VIRUS .....	8
<i>2 Methods For Assay, Detection, and Diagnosis</i>	
I. METHODS INVOLVING BIOLOGICAL ACTIVITIES OF THE VIRUS .....	12
A. Infectivity Assays .....	12
B. Indicator Hosts for Diagnosis .....	17
C. Host Range in Diagnosis .....	18
D. Methods of Transmission in Diagnosis .....	19
E. Cytological Effects for Diagnosis .....	19
F. Mixed Infections .....	20
G. Preservation of Virus Inoculum .....	20
II. METHODS DEPENDING ON PHYSICAL PROPERTIES OF THE VIRUS PARTICLE .....	21
A. Stability and Physicochemical Properties .....	21
B. Ultracentrifugation .....	22
C. Electron Microscopy .....	24

D. Chemical Assays for Purified Viruses .....	26
E. Assay Using Radioisotopes .....	26
III. METHODS DEPENDING ON PROPERTIES OF VIRAL PROTEINS ..	26
A. Serological Procedures .....	27
B. Electrophoretic Procedures .....	44
IV. METHODS INVOLVING PROPERTIES OF THE VIRAL NUCLEIC	
ACID .....	45
A. The Basis for Hybridization Procedures .....	45
B. Dot Blot Hybridization .....	47
C. Preparation and Labeling of Probes .....	48
D. Double-Stranded RNAs in Diagnosis .....	49
V. DISCUSSION AND SUMMARY .....	50

### 3 Isolation

I. CHOICE OF PLANT MATERIAL .....	54
A. Assay Host .....	54
B. Starting Material .....	54
II. EXTRACTION MEDIUM .....	55
A. pH and Buffer System .....	55
B. Metal Ions and Ionic Strength .....	56
C. Reducing Agents and Substances Protecting against Phenolic	
Compounds .....	56
D. Additives That Remove Plant Proteins and Ribosomes .....	56
E. Enzymes .....	57
F. Detergents .....	57
III. EXTRACTION PROCEDURE .....	57
IV. PRELIMINARY ISOLATION OF THE VIRUS .....	57
A. Clarification of the Extract .....	57
B. Concentration of the Virus and Removal of Low-Molecular-Weight	
Materials .....	58
V. FURTHER PURIFICATION OF THE VIRUS PREPARATION .....	60
A. Density Gradient Centrifugation .....	61
B. Gel Filtration .....	61
C. Immunoaffinity Columns .....	62
D. Chromatography .....	62
E. Concentration of the Virus and Removal of Low-Molecular-Weight	
Materials .....	62
VI. STORAGE OF PURIFIED VIRUSES .....	62
VII. IDENTIFICATION OF THE INFECTIVE PARTICLES AND CRITERIA	
OF PURITY .....	63
A. Identification of the Characteristic Virus Particle or Particles .....	63
B. Criteria of Purity .....	63

VIII. VIRUS CONCENTRATION IN PLANTS AND YIELDS OF PURIFIED VIRUS .....	65
A. Measurement of Yield .....	65
B. Reported Yields of Virus .....	66
IX. DISCUSSION AND SUMMARY .....	66

#### 4 *Components*

I. NUCLEIC ACIDS .....	68
A. Isolation .....	69
B. Methods for Determining Size .....	70
C. ssRNA Genomes .....	71
D. dsRNA Genomes .....	77
E. dsDNA Genomes .....	78
F. ssDNA Genomes .....	79
G. Nucleotide Sequences .....	80
II. PROTEINS .....	81
A. Isolation from Virus Preparations .....	81
B. Nature of the Protein Product .....	82
C. Size Determination .....	83
D. Amino Acid Sequences .....	84
E. Secondary and Tertiary Structure .....	85
F. Enzymes and Other Noncoat Proteins in Virus Particles .....	86
III. OTHER COMPONENTS IN VIRUSES .....	87
A. Polyamines .....	87
B. Lipids .....	88
C. Metals .....	88
D. Water .....	89
IV. DISCUSSION AND SUMMARY .....	90

#### 5 *Architecture*

I. METHODS .....	91
A. Chemical and Biochemical Studies .....	91
B. Sizes of Viruses .....	93
C. Electron Microscopy to Determine Fine Structure .....	94
D. X-Ray Crystallographic Analysis .....	96
E. Neutron Small-Angle Scattering .....	96
F. Serological Methods .....	97
G. Methods for Studying Stabilizing Bonds .....	99
II. PHYSICAL PRINCIPLES IN THE ARCHITECTURE OF VIRUSES ...	101
A. Quasiequivalence .....	102
B. Possible Icosahedra .....	103

C. Clustering of Subunits .....	101
D. "True" and "Quasi" Symmetries .....	106
III. GEOMETRIC VIRUSES WITH ssRNA .....	106
A. Helical Rods .....	106
B. Small Icosahedral Viruses .....	116
IV. GEOMETRIC VIRUSES WITH dsRNA .....	130
A. Reoviridae .....	131
V. GEOMETRIC VIRUSES WITH DNA .....	132
A. <i>Caulimovirus</i> Group .....	132
B. <i>Geminivirus</i> Group .....	132
VI. ENVELOPED VIRUSES .....	132
A. Rhabdoviridae .....	132
B. Tomato Spotted Wilt <i>Tospovirus</i> .....	132
VII. THE ARRANGEMENT OF NUCLEIC ACID WITHIN ICOSAHEDRAL VIRUSES .....	136
A. Order in the Nucleic Acid .....	136
B. Interactions between RNA and Protein in Small Isometric Viruses ..	139
VIII. DISCUSSION AND SUMMARY .....	141

## 6 *Replication I: Introduction to the Study of Virus Replication*

I. GENERAL PROPERTIES OF PLANT VIRAL GENOMES .....	141
A. Information Content .....	141
B. Economy in the Use of Genomic Nucleic Acids .....	144
C. The Functions of Viral Gene Products .....	145
II. HOST FUNCTIONS USED BY PLANT VIRUSES .....	147
A. Components for Virus Synthesis .....	147
B. Energy .....	148
C. Protein Synthesis .....	148
D. Nucleic Acid Synthesis .....	148
E. Structural Components of the Cell .....	148
F. Movement within the Plant .....	148
III. GENERALIZED OUTLINE FOR THE REPLICATION OF A SMALL ssRNA VIRUS .....	148
IV. THE STRATEGIES OF PLANT VIRAL GENOMES .....	149
A. The Eukaryotic Protein-Synthesizing System .....	150
B. Other Selection Pressures .....	152
V. METHODS FOR DETERMINING GENOME STRATEGY .....	153
A. Structure of the Genome .....	153
B. Defining Functional ORFs .....	156
C. Recognizing Activities of Viral Genes .....	160
D. Matching Gene Activities with Functional ORFs .....	162
VI. THE REGULATION OF VIRUS PRODUCTION .....	167
A. Regulatory and Recognition Signals in RNA Viral Genomes .....	167
B. Regulatory and Recognition Roles for Viral Proteins .....	181

VII. EXPERIMENTAL SYSTEMS FOR STUDYING REPLICATION <i>IN VIVO</i> .....	183
A. The Intact Plant .....	183
B. Surviving Tissue Samples .....	184
C. Tissue Culture .....	185
D. Cell Suspensions and Tissue Minces .....	185
E. Protoplasts .....	186
F. Radioisotopes .....	188
G. Metabolic Inhibitors .....	189
H. Metabolic Compartmentation .....	189
I. Sites of Synthesis and Assembly .....	190
VIII. ERRORS IN VIRUS SYNTHESIS .....	190
A. Mixed Virus Assembly .....	190
B. Defective Interfering Particles .....	192
IX. FUTURE STUDIES ON VIRUS REPLICATION .....	194
A. <i>In Vitro</i> Mutagenesis and Recombinant Viruses .....	194
B. Transgenic Organisms .....	194
C. <i>In Situ</i> Transcription .....	194
D. The Polymerase Chain Reaction .....	195

## 7 *Replication II: Viruses with Single-Stranded Positive Sense RNA Genomes*

I. ONE STRATEGY: POLYPROTEIN .....	197
A. <i>Potyvirus</i> Group .....	197
II. ONE STRATEGY: SUBGENOMIC RNA .....	203
A. <i>Potexvirus</i> Group .....	203
B. <i>Tombusvirus</i> Group .....	206
III. TWO STRATEGIES: SUBGENOMIC RNAs PLUS READ-THROUGH PROTEIN .....	207
A. <i>Tobamovirus</i> Group .....	207
B. <i>Luteovirus</i> Group .....	226
C. <i>Carmovirus</i> Group .....	229
IV. TWO STRATEGIES: SUBGENOMIC RNA AND POLYPROTEIN ....	231
A. <i>Tymovirus</i> Group .....	231
B. <i>Sobemovirus</i> Group .....	239
V. TWO STRATEGIES: MULTIPARTITE GENOME AND POLYPROTEIN .....	241
A. <i>Comovirus</i> Group .....	241
B. <i>Nepovirus</i> Group .....	245
VI. TWO STRATEGIES: SUBGENOMIC RNA AND MULTIPARTITE GENOME .....	248
A. <i>Bromovirus</i> Group .....	248
B. <i>Cucumovirus</i> Group .....	252
C. Alfalfa Mosaic Virus .....	252

D. <i>Ilarvirus</i> Group .....	258
E. <i>Hordeivirus</i> Group .....	258
VII. THREE STRATEGIES: SUBGENOMIC RNAs, MULTIPARTITE GENOME, AND READ-THROUGH PROTEIN .....	262
A. <i>Tobravirus</i> Group .....	262
B. <i>Furovirus</i> Group .....	265
C. <i>Dianthovirus</i> Group .....	267
VIII. PEA ENATION MOSAIC VIRUS GROUP .....	269
A. Genome Structure .....	269
B. <i>In Vitro</i> Translation .....	269
C. <i>In Vivo</i> Studies .....	269

## 8 *Replication III: Other Virus Groups and Families*

I. CAULIMOVIRUS GROUP .....	271
A. Genome Structure .....	271
B. Proteins Encoded and Their Functions .....	272
C. RNA and DNA Synthesis .....	275
D. Recombination in CaMV DNA .....	277
E. Replication and Movement <i>in Vivo</i> .....	278
II. GEMINIVIRUS GROUP .....	279
A. Genome Structure .....	280
B. Functions of the ORF Products .....	283
C. mRNAs .....	284
D. Control of Transcription .....	286
E. DNA Replication .....	286
F. Agroinfection with Geminiviruses .....	287
III. PLANT REOVIRIDAE .....	288
A. Genome Structure .....	288
B. RNA Transcription and Translation .....	290
C. Intracellular Site of Replication .....	290
D. RNA Selection during Virus Assembly .....	291
IV. PLANT RHABDOVIRIDAE .....	293
A. Genome Structure .....	294
B. Proteins Encoded .....	294
C. The Viral Transcriptase .....	295
D. Genome Replication .....	295
E. mRNA Synthesis .....	295
F. Protein Synthesis .....	296
G. Cytological Observations on Replication .....	296
V. A PLANT MEMBER OF THE BUNYAVIRIDAE .....	296
VI. POSSIBLE USES OF VIRUSES FOR GENE TRANSFER .....	299
A. Caulimoviruses .....	300
B. Geminiviruses .....	300

C. RNA Viruses .....	301
D. Viruses as Sources of Control Elements for Transgenic Plants .....	303

## 9 *Viroids, Satellite Viruses, and Satellite RNAs*

I. VIROIDS .....	306
A. Structure of Viroids .....	306
B. Replication of Viroids .....	311
C. Biological Aspects of Viroids .....	316
D. Molecular Basis for Biological Activity .....	318
E. Diagnostic Procedures for Viroids .....	320
II. SATELLITE VIRUSES AND SATELLITE RNAs .....	321
A. Satellite Plant Viruses .....	322
B. Satellite RNAs .....	325

## 10 *Transmission, Movement, and Host Range*

I. DIRECT PASSAGE IN LIVING HIGHER PLANT MATERIAL .....	339
A. Through the Seed .....	339
B. By Vegetative Propagation .....	343
C. By Grafting .....	343
D. By Dodder .....	345
II. TRANSMISSION BY ORGANISMS OTHER THAN HIGHER PLANTS .....	345
A. Invertebrates .....	345
B. Fungi .....	346
C. <i>Agrobacterium tumefaciens</i> .....	348
III. MECHANICAL TRANSMISSION .....	348
A. Applying the Inoculum .....	349
B. Nature and Number of Infectible Sites .....	351
C. Number of Particles Required to Give an Infection .....	355
D. Mechanical Transmission in the Field .....	357
E. Abiotic Transmission in Soil .....	357
IV. MOVEMENT AND FINAL DISTRIBUTION IN THE PLANT .....	358
A. Methods .....	358
B. The Form in Which Virus Moves .....	359
C. The Role of Plasmodesmata .....	361
D. Time of Movement from First Infected Cells .....	361
E. Rate of Cell-to-Cell Movement .....	362
F. Long-Distance Movement .....	362
G. Role of Viral Gene Products in Virus Spread within the Plant .....	364
H. Final Distribution in the Plant .....	366
V. THE HOST RANGE OF VIRUSES .....	371
A. Limitations in Host Range Studies .....	371
B. Patterns of Host Range .....	373

C. The Molecular Basis for Host Range .....	373
VI. DISCUSSION AND SUMMARY .....	377

## 11 *Disease Symptoms and Effects on Metabolism*

I. MACROSCOPIC SYMPTOMS .....	380
A. Local Symptoms .....	380
B. Systemic Symptoms .....	381
C. Agents Inducing Viruslike Symptoms .....	392
D. The <i>Cryptovirus</i> Group .....	397
II. HISTOLOGICAL CHANGES .....	397
A. Necrosis .....	397
B. Hypoplasia .....	397
C. Hyperplasia .....	398
III. CYTOLOGICAL EFFECTS .....	402
A. Methods .....	402
B. Effects on Cell Structures .....	403
C. Virus-Induced Structures in the Cytoplasm .....	407
D. Cytological Structures Resembling Those Induced by Viruses .....	411
IV. EFFECTS ON PLANT METABOLISM .....	411
A. Experimental Variables .....	411
B. Nucleic Acids and Proteins .....	414
C. Lipids .....	416
D. Carbohydrates .....	416
E. Cell Wall Compounds .....	417
F. Respiration .....	417
G. Photosynthesis .....	417
H. Transpiration .....	418
I. Activities of Specific Enzymes .....	418
J. Hormones .....	420
K. Low-Molecular-Weight Compounds .....	420
L. Summary .....	420

## 12 *Induction of Disease*

I. THE KINDS OF HOST RESPONSE TO INOCULATION WITH A VIRUS .....	42
II. THE ROLE OF VIRAL GENES IN DISEASE INDUCTION .....	42
A. Some General Considerations .....	42
B. Specific Viral Gene Products .....	42
C. Defective Interfering Particles .....	42
D. Future Studies on the Role of Viral Genes in Disease Induction .....	42
III. HOST PROTEINS INDUCED BY VIRUS INFECTION .....	42
A. Biological Aspects of Local and Systemic Acquired Resistance .....	43
B. The Host Proteins Induced in the Hypersensitive Response .....	43



C. Lack of Specificity in PR Protein Induction .....	434
D. Induction of mRNAs for PR Proteins .....	434
E. Synthesis of PR Proteins <i>in Vitro</i> and <i>in Vivo</i> .....	435
F. Other Host Proteins Induced during the Hypersensitive Response ...	435
G. Steps in the Induction of Host Proteins .....	436
H. Other Biochemical Changes during the Hypersensitive Response ...	436
I. Roles of Host-Coded and Viral-Coded Proteins .....	436
J. Systemic Acquired Resistance .....	437
K. Wound Healing Responses .....	438
L. Antiviral Factors .....	439
IV. PROCESSES INVOLVED IN DISEASE INDUCTION .....	439
A. Sequestration of Raw Materials .....	439
B. Effects on Growth .....	440
C. Effects on Chloroplasts .....	444
D. Leaf Ontogeny and Mosaic Disease .....	445
E. Role of Membranes .....	450
V. FACTORS INFLUENCING THE COURSE OF INFECTION AND DISEASE .....	450
A. Virus Concentration in the Inoculum .....	451
B. Plant Factors .....	451
C. Environmental Factors .....	460
D. Interaction with Other Agents .....	464
VI. DISCUSSION AND SUMMARY .....	468

### 13 Variability

I. ISOLATION OF STRAINS .....	470
A. Strains Occurring Naturally in Particular Hosts .....	471
B. Isolation from Systemically Infected Plants .....	471
C. Selection by Particular Hosts or Conditions of Growth .....	471
D. Isolation by Means of Vectors .....	472
E. Isolation of Artificially Induced Mutants .....	472
II. THE MOLECULAR BASIS OF VARIATION .....	474
A. Mutation (Nucleotide Changes) .....	474
B. Recombination .....	475
C. Deletions .....	478
D. Additions .....	480
E. Nucleotide Sequence Rearrangement .....	480
F. Reassortment of Multiparticle Genomes .....	481
G. The Origin of Strains in Nature .....	481
III. CRITERIA FOR THE RECOGNITION OF STRAINS .....	482
A. Structural Criteria .....	482
B. Serological Criteria .....	489
C. Biological Criteria .....	497