



INTERACTIONS BETWEEN GENOMIC RNAs AND A SATELLITE  
RNA OF CUCUMOVIRUSES

by

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CONTENTS

|  | Page |
|--|------|
| SUMMARY  | v    |
| STATEMENT  | viii |
| ACKNOWLEDGEMENTS   | ix   |
| ABBREVIATIONS  | x    |
| <br>   |      |
| <i>CHAPTER 1. INTRODUCTION</i>                                     | 1    |
| The Cucumovirus Group  | 1    |
| Properties of the Cucumovirus Capsid                               | 2    |
| Properties of the Cucumovirus Genome                               | 3    |
| Encapsidation of the Cucumovirus Genome                            | 4    |
| Genetic Analyses of Cucumoviruses                                  | 5    |
| Scope of this Thesis   | 7    |
| <br>   |      |
| <i>CHAPTER 2. GENERAL MATERIALS AND METHODS</i>                    | 9    |
| <i>MATERIALS</i>   | 9    |
| Virus Isolates   | 9    |
| Chemicals  | 9    |
| Buffers and Solutions  | 9    |
| Instruments  | 13   |
| <i>METHODS</i>   | 13   |
| Plants, Inoculation and Virus Propagation                          | 13   |
| Virus Purification   | 14   |
| Isolation of Viral and <i>E. coli</i> RNA                          | 19   |
| Isolation of Total Leaf RNAs and Virus-specific ds-RNA             | 21   |
| RNA Base Ratio Analyses  | 23   |
| Analytical Gel Electrophoresis of Viral RNAs                       | 23   |
| Preparative Electrophoretic Separation of Viral RNA Components     | 25   |
| Construction, Isolation and Characterization of Pseudorecombinants | 26   |
| Aphid Transmission Studies   | 27   |

|   | Page   |
|---|--------|
| Stabilization of Viruses with Formaldehyde and<br>Glutaraldehyde  | 27     |
| Serological Techniques  | 27     |
| Isolation of Viral Protein  | 29     |
| Polyacrylamide Gel Electrophoresis of Viral Protein   | 30     |
| Amino Acid Analyses   | 30     |
| Preparation of Complementary DNA Transcripts  | 31     |
| <i>Sat</i> -RNA   | 32     |
| <i>QCMV</i> -RNA 3 and <i>STNV</i> -RNA   | 34     |
| Radiiodination of <i>Sat</i> -RNA   | 35     |
| RNA-cDNA Hybridization Procedures   | 36     |
| RNA-RNA Hybridization Techniques  | 38     |
| Spectrophotometry   | 39     |
| Scintillation Spectrophotometry   | 39     |
| Precautions Against RNase   | 41     |
| <br><i>CHAPTER 3. COMPARATIVE PROPERTIES OF FOUR CUCUMOVIRUS<br/>ISOLATES</i>   | <br>42 |
| Purification and Properties of GCMV and MCMV  | 42     |
| Properties of the Four Cucumovirus Isolates   | 46     |
| Conclusions   | 53     |
| <br><i>CHAPTER 4. GENETIC ANALYSES OF PSEUDORECOMBINANTS<br/>CONTAINING RNA 3 FROM DIFFERENT CUCUMOVIRUS<br/>ISOLATES</i> | <br>54 |
| Development of a Procedure for the Preparative<br>Fractionation of Cucumovirus Genomic RNAs                               | 54     |
| Recovery and Specific Infectivity of Fractionated<br>Genomic RNA of <i>QCMV</i>   | 57     |
| Infectivity of Heterologous Mixtures of Genomic<br>RNAs from Different Cucumoviruses                                      | 59     |
| Construction and Characterization of Pseudorecombinants   | 59     |
| Association of <i>Sat</i> -RNA with Certain Pseudorecombinants  | 62     |
| Biological Properties of Pseudorecombinants   | 64     |
| Association of RNA 3 with the Aphid Transmission of <i>CMV</i>  | 69     |
| Conclusions   | 72     |

|  | Page    |
|--|---------|
| <i>CHAPTER 5.</i> BIOLOGICAL PROPERTIES OF Sat-RNA AND ITS<br>INCIDENCE IN CUCUMOVIRUSES ISOLATED IN AUSTRALIA | 74      |
| Screening of Cucumovirus Isolates for the Presence of<br>Sat-RNA   | 74      |
| Some Properties of the Cucumovirus Isolates  | 75      |
| Comparison of the Base Sequences of Sat-RNA from Four<br>Cucumovirus Isolates                                  | 79      |
| Transmission of Sat-RNA between Cucumoviruses  | 80      |
| Interference of Cucumovirus Replication by Sat-RNA   | 81      |
| Effect of Plant Host on the Replication of Sat-RNA   | 84      |
| Dilution End-Point of Sat-RNA  | 85      |
| Modification of CMV-Host Interactions by Sat-RNA   | 86      |
| Attempts to Detect Sat-RNA Sequences in Nucleic Acids<br>Isolated from Healthy Plants                          | 90      |
| Conclusions  | 91      |
| <br><i>CHAPTER 6.</i> COMPARATIVE BASE SEQUENCE HOMOLOGY BETWEEN<br>Sat-RNA AND CARNA 5                        | <br>92  |
| Experimental   | 92      |
| Conclusions  | 96      |
| <br><i>CHAPTER 7.</i> REPLICATION AND <i>IN VIVO</i> SURVIVAL OF Sat-RNA                                       | <br>97  |
| Dependence of Sat-RNA Replication on Helper Viruses  | 97      |
| Dependence of Sat-RNA Replication on Cucumoviruses   | 98      |
| Detection of ds-RNA Associated with the Replication<br>of Sat-RNA  | 102     |
| <i>In vivo</i> Survival of Sat-RNA in the Absence of Helper<br>Virus   | 103     |
| Ability of Other Plant Viral RNAs to Survive <i>in vivo</i>  | 104     |
| QCMV-RNA 3   | 106     |
| STNV-RNA   | 110     |
| Conclusions  | 113     |
| <br><i>CHAPTER 8.</i> STUDIES ON THE <i>IN VITRO</i> STABILITY OF Sat-RNA<br>AND STNV-RNA                      | <br>114 |
| The Possible <i>in vivo</i> Survival of Sat-RNA as a ds-RNA  | 114     |
| Kinetics of Thermal Denaturation of Sat-RNA and STNV-RNA   | 117     |

|  | Page    |
|--|---------|
| Kinetics of Nuclease Digestion of Sat-RNA and STNV-RNA   | 120     |
| Comparison of the Stability of Sat-RNA and STNV-RNA with that of the RNA of their Helper Viruses in Sap Extracts | 122     |
| Conclusions  | 124     |
| <br><i>CHAPTER 9. GENERAL DISCUSSION</i>   | <br>125 |
| Properties of the Cucumovirus Isolates   | 125     |
| Genetics of Cucumoviruses  | 127     |
| Properties of Sat-RNA, and its Interaction with Helper Viruses   | 134     |
| Satellites: Their <i>in vivo</i> Survival and Possible Origins   | 141     |
| <br><i>APPENDIX : Papers Published</i>   | <br>147 |
| <br><i>REFERENCES</i>  | <br>148 |

SUMMARY

1. Fourteen cucumovirus isolates were examined with respect to their RNA profiles, antigenic properties and amino acid compositions. All isolates contained four major RNA components, designated RNAs 1-4 in order of decreasing molecular weight, the three largest of which are genomic. However, some isolates had an additional RNA with a molecular weight of approximately  $1.05 \times 10^5$  daltons. Antigenic properties of the isolates divided them into two serologically unrelated groups; 11 strains of CMV (two of the CMV isolates were considered to be the same) and 2 strains of TMV. Amino acid composition data confirmed this division, and in addition separated the CMV strains into 2 sub-groups.

2. Heterologous mixtures of genomic RNAs 1+2 and RNA 3 from three strains of CMV and a strain of TAV were used to investigate the genetic function of RNA 3. It was confirmed that RNA 3 specifies coat protein and it was also demonstrated that it is associated with aphid transmission. In addition it was shown that symptom expression on host plants can be determined by genetic information on different RNA components. Some host reactions appear to be associated with gene(s) located on RNAs 1 and/or 2 and others on RNA 3 alone. However, in some instances, the symptom expression appears to involve interactions between genetic information on both RNA 3 and RNAs 1 and/or 2.

3. Two types of RNA, each with a molecular weight of approximately  $1.05 \times 10^5$  daltons, designated RNA 5 and Sat-RNA, have been found in purified preparations of CMV and have been characterized by molecular

hybridization analysis using  $^{32}\text{P}$ -labelled complementary DNA (cDNA) probes. RNA 5 was shown to consist of specific cleavage products of RNAs 1-4. In contrast, Sat-RNA has a unique nucleotide sequence with no detectable homology with CMV-RNAs.

4. Sat-RNA was compared with a similar low molecular weight RNA (CARNA 5) isolated in the U.S.A. Whereas CARNA 5 is known to induce severe disease symptoms in tomato plants in the presence of its CMV helper, no such reactions could be induced by all strains of CMV examined in the presence of Sat-RNA. A comparison of the base sequences of these two satellite RNAs showed that they have approximately 70% of their nucleotide sequences in common. It would appear that this difference in their primary structures is reflected in differences in their biological properties.

5. Sat-RNA was readily transmitted from one cucumovirus strain to another, and in all strains of CMV examined, it was replicated (and encapsidated) to high levels. In contrast, Sat-RNA was produced in low amounts in the presence of TAV. As a consequence of Sat-RNA replication, the yield of its associated CMV and the proportion of CMV-RNAs 1 and 2 were both markedly reduced. Sat-RNA is unable to replicate autonomously and is hence dependent on cucumoviruses for both its replication and encapsidation. This helper function could not be fulfilled by either alfalfa mosaic virus or tobacco ringspot virus. Using cDNA transcribed to Sat-RNA as a probe, it was shown that Sat-RNA is able to survive *in vivo* for prolonged periods in the absence of its helper virus. This capacity for *in vivo* survival was also shared by the RNA of satellite tobacco

necrosis virus (STNV-RNA), but not by the genomic RNA 3 of CMV.

6. Both Sat-RNA and STNV-RNA was shown to be more resistant to nuclease digestion and inactivation in crude plant extracts than the RNAs of their respective helper viruses. It is possible that the *in vivo* survival of Sat-RNA and STNV-RNA may be related to features of their molecular structure.