INTERACTIONS BETWEEN GENOMIC RNAS AND A SATELLITE RNA OF CUCUMOVIRUSES

by

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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 1.05×10^5 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 1.05×10^5 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}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.