Satellite RNAs of Plant Viruses: Structures and Biological Effects

MARILYN J. ROOSSINCK,1* DAVID SLEAT,2 AND PETER PALUKAITIS3

INTRODUCTION .................................................................................................................. 265
NUCLEOTIDE SEQUENCE AND STRUCTURE ............................................................... 266
Potential ORFs .................................................................................................................... 266
Secondary Structure ........................................................................................................... 266
Satellites of CMV .............................................................................................................. 266
RNA C of TCV .................................................................................................................... 267
CyRSV satellite RNA ......................................................................................................... 268
Satellite RNA of TobRV ..................................................................................................... 269
Satellite of LTSV ............................................................................................................... 269
Sequence Variation ........................................................................................................... 269
REPLICATION .................................................................................................................... 270
Specificity and Efficiency .................................................................................................. 270
Enzymology ....................................................................................................................... 271
Self-Processing .................................................................................................................. 272
Nepovirus satellites .......................................................................................................... 272
Sobemovirus satellites ...................................................................................................... 272
Cucumovirus satellites ...................................................................................................... 272
EFFECT ON HELPER VIRUS ............................................................................................ 272
Alteration in Virus Titer ..................................................................................................... 272
Symptom Modulation ........................................................................................................ 272
Strategies for Virus Control ............................................................................................. 273
Satellite inoculation or spraying ....................................................................................... 273
Transgenic plants ............................................................................................................... 273
Potential risks .................................................................................................................... 274
SATELLITELIKE MOLECULES ........................................................................................ 274
ORIGINS ............................................................................................................................. 274
CONCLUSIONS ................................................................................................................ 274
ACKNOWLEDGMENTS ..................................................................................................... 275
REFERENCES ....................................................................................................................... 275

INTRODUCTION

A majority of plant viruses contain single-stranded RNA genomes, many of which are divided (131). These viruses may encapsidate both genomic and subgenomic RNAs. In addition, satellites are often associated with plant RNA viruses. Satellites by definition are completely dependent on their helper viruses for replication. They may encode their own coat protein (the satellite viruses), or they may rely on the helper virus for encapsidation as well as replication (the satellite RNAs) (41, 47, 98, 143, 179). They can be distinguished from defective interfering (DI) RNAs by the absence of significant sequence homology with their helper viruses. Recently, some RNA molecules which have some characteristics of satellite RNAs, but which are not true satellites per the above definition, have been observed. These include RNA molecules with some regions of extensive sequence homology with their helper viruses, such as the DI-like satellite RNA chimera of turnip crinkle carmovirus (TCV) (181), and RNA molecules which are not required by the helper virus for experimental infection, but which appear to be essential for the natural life cycle of the virus, such as the small RNA associated with groundnut rosette virus (GRV) (7, 140, 142). These molecules will not be discussed in the general portion of this review, but will be discussed briefly in a separate section.

The first satellite, described in 1962, was found associated with tobacco necrosis virus. It was serologically unrelated to tobacco necrosis virus and hence was a satellite virus; i.e., it encoded its own coat protein (109). In 1969, a satellite was found associated with tobacco ringspot nepovirus (TobRV), which was encapsidated by the helper virus, and the term "satellite" was broadened to include satellite RNAs (175). In recent years, a large number of satellite RNAs, associated with several groups of plant viruses, have been reported. In addition, several satellite viruses and DI RNAs have been described. Since this field has become increasingly broad, this review will be limited to satellite RNAs. A number of recent reviews on satellite viruses and satellite RNAs have appeared (47, 98, 179); however, with the rapidly growing body of knowledge in this field, considerable new information is available. This review will focus on a functional approach to the satellite RNAs, grouping the satellites by overall structural relatedness. Although a large number of satellites have now been sequenced, much less is known about their biology, with some notable exceptions. The

* Corresponding author.
replication of satellites is also an area in which little is known about most satellites, in part because of a lack of knowledge about replication of RNA plant viruses in general.

Satellite RNAs range in size from just under 200 nucleotides (nt) to approximately 1,500 nt (Table 1). The larger satellites appear to contain functional open reading frames (ORFs), although as yet no function has been assigned to any of their gene products. The small satellites do not appear to encode any functional ORFs, but tend to be highly structured (Fig. 1). Small satellites may be either linear or circular. The circular satellites have been referred to as virusoids in some of the literature because of their structural resemblance to viroids (28, 82, 111). However, since many properties distinguish viroids from satellites, virusoids will be referred to as circular satellite RNAs in this review to avoid confusion.

Despite their small size and the usual absence of any potential gene products, satellites may have a dramatic effect on the symptoms induced by their helper virus, ranging from amelioration to severe exacerbation. These symptom effects vary with the helper virus, host plant, and satellite. It is hoped that a better understanding of the structures of such satellites will lead to testable models for the biological effects of satellites.

**NUCLEOTIDE SEQUENCE AND STRUCTURE**

Satellites can be broadly classified into two groups on the basis of their coding capacity. The larger satellites of the nepoviruses contain ORFs which have been shown to be functional in vitro and in some cases in vivo (Table 1). The smaller satellites do not appear to contain any ORFs which are functional in vivo, although in some cases in vitro translation products have been observed (see below) (3, 79). The smaller satellites can be further divided into groups on the basis of several criteria related to their nucleotide sequence and structure: the small linear satellites, which include the small nepovirus satellites and the cucumovirus satellites; the circular satellites associated with sobemoviruses; and the DI-like satellite RNA chimaera of TCV. The small linear satellites can be further subdivided into those that have a circular replication intermediate (the small nepovirus satellites) and those that do not (the cucumovirus satellites).

**Potential ORFs**

Sequence analyses of a number of satellites have indicated potential ORFs that might be expressed in vivo and might have some potential role in either replication or pathogenesis. In a few cases protein products of these potential ORFs have been detected in vivo, but in general, definitive proof of the function of potential ORFs in vivo has been elusive and has been a source of much controversy.

The large satellites of the nepoviruses unequivocally can be assigned a role as mRNAs in vivo. The best characterized of these satellites is the satellite RNA of tomato black ring nepovirus (TBRV). An Mf 48,000 protein was first observed in a rabbit reticulocyte lysate programmed with TBRV satellite RNA (46, 48, 49). A protein of identical mobility was found in vivo in tobacco protoplasts infected with TBRV containing satellite RNA, but not in uninfected protoplasts (48), suggesting that TBRV satellite RNA can indeed act as an mRNA in vivo. Sequence analysis of several isolates of TBRV satellite RNAs subsequently identified the ORF for the Mf 48,000 protein, which is composed of 419 to 424 amino acids, depending on the isolate (78). The TBRV satellite RNA isolates can be divided into two groups on the basis of their nucleotide and amino acid sequence homology: the S- and L-sat RNAs and the E- and C-sat RNAs. Within each group, proteins are ~90% homologous, whereas there is only 57% homology between the groups (78). However, all of the potential TBRV satellite-encoded proteins are highly basic and hydrophobic, suggesting a functional similarity.

Several other large nepovirus satellites have been shown to direct translation of proteins in vitro (summarized in Table 1). These range in size from Mf 37,000 for the grapevine fanleaf nepovirus (GFVL) satellite RNA to Mf 45,000 for the myrobalan latent ring spot nepovirus (MLRV) satellite RNA. In general, the amino acid sequences of these potential ORFs vary greatly and the extent of sequence similarity is proportional to the extent of serological relatedness of their respective helper viruses (46). However, the proteins encoded by TBRV, GFVL, and chicory yellow mottle nepovirus L2 (CYMV-L2) satellite RNAs all contain a highly basic domain in the first 100 amino acid residues, as well as a hydrophobic N-terminal domain followed by a very hydrophilic region (173). Although no biological functions have been assigned to any of these proteins, their secondary similarity is highly suggestive of a similar function. The highly basic domains are reminiscent of histones, and an RNA-binding role has been suggested for these proteins (78).

Although many of the small satellites contain potential ORFs, there is strong evidence against any in vivo function for these ORFs. The S-sat and the F-sat RNAs of cucumber mosaic cucumovirus (CMV) have both been shown to produce protein products in vitro (3, 79, 150), and the E-sat, Y-sat, and OY2-sat RNAs of CMV have been shown to bind 80S ribosomes in vitro (79, 80); however, the ORFs of the CMV satellite RNAs are not conserved among different isolates (56, 105). Moreover, neither mutagenesis of the 5'-proximal initiation codon in an infectious clone of the D-sat RNA (23) nor a frameshift mutation of the ORFs of Y-sat RNA (33, 89, 130) altered the biological activity of these satellite RNAs.

Although several of the small nepovirus and sobemovirus satellites contain short ORFs, none that have been tested have been able to direct the synthesis of any protein products in vitro (43, 112, 137, 151, 173) (Table 1). In addition, where more than one isolate of these satellites has been sequenced, such as the satellite RNA of tomato bushy stunt virus (TBSV) (15, 17) or lucerne transient streak sobemovirus (LTSV) (1, 110), the ORFs are not always conserved. Moreover, with the sobemoviruses, the circular structure argues against translation in vivo (114).

Of the tombusvirus satellites, only the satellite RNA of cymbidium ringspot tombusvirus (CyRSV) has been sequenced (172). Although several small ORFs were present, CyRSV satellite RNA also failed to direct the synthesis of any detectable protein products in vitro (172).

In summary, with the exception of the large nepovirus satellites, plant virus satellites generally do not appear to encode any functional proteins, and, hence, their biological functions must rely on the nucleotide sequence and the resultant secondary structure.

**Secondary Structure**

The small satellites have highly ordered secondary structures, with the base pairing ranging from 49% (satellite RNAs of CMV) to 73% (satellite RNAs of solanum nodiflorum mottle sobemovirus [SNMV]) (Table 2). The small size
and high degree of secondary structure are probably responsible for the high stability and survivability of satellites in vitro as well as in vivo (88, 138, 139). The latter may in turn account for the highly infectious nature of many satellites (25, 65, 86, 100, 104, 177, 195).

**Satellites of CMV.** In contrast to the secondary structures of the other satellites described below, the secondary structures of a number of satellite RNAs of CMV are based not solely on computer-generated structures, but also on nuclease cleavage sensitivity data. The structures determined for six satellite RNAs of CMV are very similar (56, 62, 80), with base pairing ranging from 49 to 52% (Table 2); one of these is shown in Fig. 1A. The sequences involved in pathogenicity are located in discrete small domains: C is the domain...
TABLE 1. Summary of satellite RNAs

| Helper virus* | Mol mass (kDa) | No. of nt | Mol. mass of translation products (kDa)* | Reference |
|---------------|----------------|-----------|----------------------------------------|-----------|
| Cucumber mosaic cucumovirus (CMV) | 110–125 | 333–342 | 2.0–3.9 | 2, 3, 26, 32, 56, 62, 79, 80, 85, 105, 116, 150, 168, 183 |
| Peanut stunt cucumovirus (PSV) | 127 | 393 | – | 32, 80, 81, 127, 128, 130 |
| Tobacco ringspot nepovirus (TobRV) | 89–120 | 359 | – | 15, 17, 151 |
| Tomato blackring nepovirus (TBRV) | 111, 245 | –340, –750 | NR | 165 |
| Chickory yellow mottle nepovirus (CYMV) | 480 | 1,375* | 48 | 46, 48, 49, 78, 134 |
| Arabis mosaic nepovirus (ArMV) | 75 | 300 | NR | 57, 173 |
| Strawberry latent ringspot nepovirus (SLRV) | 400 | –1,200 | 38 | 132 |
| Myrobalan latent nepovirus (MLRV) | 450 | –1,400 | 45 | 46, 53 |
| Grapevine Bulgarian latent nepovirus (GBLV) | 500 | –1,500 | NR | 54 |
| Grapevine fanleaf nepovirus (GFLV) | 400 | 1,114* | 37 | 50, 158 |
| Pea enation virus (PEV) | 300 | –900 | NR | 31 |
| Velvet tobacco mottle sobemovirus (VTMoV) | 120 | 365, 366 | – | 43, 73 |
| Solanum nodiflorum mottle sobemovirus (SNMV) | 130 | 377 | – | 73, 112 |
| Lucerne transient streak sobemovirus (LTSV) | 120 | 324 | – | 1, 110, 137 |
| Subterranean clover mottle sobemovirus (SCMoV) | 332, 388 | NR | 28 |
| Turnip crinkle carmovirus (TCV) | 130, 170 | 194, 230, 355 | – | 4, 181 |
| Tomato bushy stunt tombusvirus (TBSV) | – | –700 | NR | 51 |
| Artichoke mottled crinkle tombusvirus (AMCV) | – | –700 | NR | 51 |
| Carnation Italian ringspot tombusvirus (CIRV) | – | –700 | NR | 51 |
| Cymbidium ringspot tombusvirus (CyRSV) | – | 621 | – | 12, 172 |
| Petunia asteroid mosaic tombusvirus (PAMV) | – | –700 | NR | 51 |
| Pelargonium leaf curl tombusvirus (PLCV) | – | –700 | NR | 51 |
| Groundnut rosette virus (GRV) | – | – | – | – |
| MC* | 300 | –900 | NR | 145 |
| NG* | 300 | –900 | NR | 142 |
| Beet necrotic yellow vein furovirus (BNYVV) | – | – | – | – |
| 3b* | 650 | 1,777* | 25 | 8, 190 |
| 4b* | 540 | 1,467* | 31 | 8, 190 |
| 5b* | 510 | –1,400 | NR | 190 |
| 6b* | 360 | –1,000 | NR | 190 |

* Abbreviations for helper viruses are given in parentheses.
* Molecular masses of the satellite RNAs, as assessed by gel electrophoresis, are given in kilodaltons. –, not reported as molecular mass.
* Number of nucleotides (nt) is given where sequences are known. An estimation of the number, based on molecular mass or gel electrophoresis mobility, is given where sequence information is lacking (indicated by –).
* Molecular masses of in vitro translation products are given in kilodaltons; –, no translation product observed in in vitro translation.
* Virus strain.
* NR, no report.
* Number of nucleotides indicated does not include the 3' poly(A) tail.
* Satellite RNA designation.

involved in chlorosis induction in tobacco and tomato (89, 130, 185a); and N is the region involved in necrosis induction in tomato (33, 130, 184) (see Effect on Helper Virus, below). The structure of the necrosis domain (not shown in Fig. 1A) has been determined only for the necrosis-inducing 369-nt Y-sat RNA of CMV (80). The lower half of the Y-sat RNA structure is quite different from the structure shown here. Potentially similar tertiary structures are a matter of conjecture.

RNA C of TCV. The chimeric RNA C of TCV, which is part satellite RNA and part DI-like RNA, contains only slightly more base pairing (56%) than the satellite RNAs of CMV do (49 to 52%) (Table 2). However, the proposed structure (Fig. 1B) (181) is quite different from that shown for the CMV satellite RNAs (Fig. 1A). Although sequences involved in replication are scattered throughout the molecule (18, 180), considerably less sequence alteration can be tolerated in the 3' half of the molecule, which contains the DI-like portion of 3' genomic RNA sequences (180). Modification of sequences in one specific region affects monomer accumulation (18, 180) and modulates symptom expression (M) (180). In addition, other sequences of the TCV DI-like component, as well as part of the adjacent satellitelike sequences, are involved in determining virulence (180).
Considerable in vitro manipulation of the sequence of TCV RNA C is possible (18, 25, 180), compared with the CMV satellite RNAs, which tolerate few deletions or insertions (33, 89, 114a, 130). Hence, it would be interesting to know what effects these sequence manipulations have on the secondary structure of TCV RNA C.

CyRSV satellite RNA. The 621-nt satellite RNA of CyRSV also has a forked structure, with adjacent 5' and 3' ends (172). There are also stretches of sequence in common with the helper virus genomic RNA, although these regions are neither as extensive as in TCV (ca. 19% of the satellite RNA of CyRSV) nor as similar to the genomic RNA (60 to 100% homology). In addition, the homology to the genomic RNA is scattered throughout the 5'-two-thirds of the CyRSV satellite RNA (172).

Satellite RNA of TobRV. The 359-nt satellite RNA of TobRV can be folded into a cruciform structure with 69% base pairing (Table 2) and 5' and 3' contiguous ends (Fig. 1C) (111). TobRV satellite RNA also exists in a circular form in infected leaves, but not in isolated virus particles (124). The limited sequence rearrangements seen in the satellite RNAs from strains 62L and NC-87, with respect to the RNA from the budblight strain, are located between nucleotides 100 and 140 (R in Fig. 1C) and do not alter the proposed secondary structure (17).

The TobRV satellite RNA contains two ribozymes (see Self-Processing, below): a hammerhead ribozyme (H) in the positive-sense RNA, involving an alteration in the local secondary structure (39, 72); and a paperclip ribozyme (P) in the negative-sense RNA (72, 195). These are involved in the processing of the multimeric positive- and negative-sense RNA synthesized during replication of the satellite TobRV RNA (13, 113, 160).

Second-structure models have also been computer generated for two other small nepovirus satellites: the 300-nt satellite RNA of arabis mosaic nepovirus (ArMV) (106) and the 457-nt satellite RNA (S1) of CYMV (173). Both of these satellites contain sequences in common with TobRV satellite RNA (106, 173). The sequences of satellite ArMV RNA and satellite CYMV-S1 RNA that are most highly conserved, both with each other and with TobRV satellite RNA, can be folded into the two ribozyme structures described above, with an overall sequence homology of 75% in the hammerhead ribozyme and 80% in the paperclip ribozyme (72, 106, 173).

The above three small nepovirus satellites all contain a free hydroxyl group at the 5' end and a 2',3'-cyclic phosphodiester at the 3' end (15, 106, 156). They also all produce circular forms in planta, which are not encapsidated (106, 124, 173). Thus, these satellites appear to have quite similar genome replication strategies, although they probably cannot interchange helper viruses or be supported in their replication by other nepoviruses (159).

Satellite of LTSV. The proposed structure of a circular satellite of LTSV is shown in Fig. 1D (110). This satellite RNA contains 324 nt, 72% of which are base paired (Table 2). The highly base-paired structure is reminiscent of the structure of viroids, although the structural intermediates formed during denaturation (169) and replication (82, 187) of circular satellites are dissimilar.

During replication, LTSV satellite RNA and the satellites of other sobemoviruses generate multimeric forms of both positive and negative polarity (21, 82, 187). The processing events to produce monomeric molecules of LTSV satellite RNA involve self-cleavage reactions (39). In this case, however, the ribozymes of both the positive and negative LTSV satellite RNA (+H and −H, respectively, in Fig. 1D) are of the hammerhead class (38). By contrast, the satellite RNAs of velvet tobacco mottle sobemovirus (VTMOV), SNMV, and subterranean clover mottle sobemovirus (SCMoV), which have structures very similar to that of LTSV satellite RNA, contain only self-cleaving (hammerhead) ribozymes in their positive-sense RNA and do not produce monomeric negative-sense RNA (21, 28, 39, 82).

The sobemovirus satellites differ from the small nepovirus satellites structurally in two ways: (i) the circular form is preponderant in the sobemovirus satellites (45, 64, 163, 192), and the linear form is preponderant in the nepovirus satellites (106, 124, 173); and (ii) the linear satellites contain branch points, giving rise to cruciform structures (106, 173), whereas the circular satellites consist of rod-shaped, viroid-like molecules (28, 73, 110). The significance of these structures on the biology of the various satellites has yet to be determined.

### Sequence Variation

Isolates of a number of satellites have been sequenced. In most cases the sequence variants do not show biological variation. However, there are some noted exceptions—the satellite RNAs of CMV (56, 80, 85, 105, 127) and peanut stunt cucumovirus (PSV) (146). For most satellites, only one isolate has been found and/or sequenced. For some, such as the satellite RNAs of CMV, PSV, TobRV, and LTSV, there are minor sequence variants that do not affect either the biology or the secondary structure of the satellites (1, 17, 32, 56, 80, 85, 105, 110, 127, 146). These variant satellites show 1 to 20% sequence differences.

### Table 2. Secondary structure of small satellite RNAs

| Helper virus<sup>a</sup> | Total nt | No. (%) of base-paired nt | No. of bp involving: |
|-------------------------|----------|--------------------------|---------------------|
|                         |          |                          | G-C | G-U | A-U |
| CMV                     |          |                          |     |     |     |
| Bi<sup>b</sup>          | 340      | 170 (50)                 | 46  | 16  | 23  |
| Bi<sup>c</sup>          | 337      | 164 (49)                 | 44  | 18  | 20  |
| G<sup>b</sup>           | 333      | 174 (52)                 | 47  | 16  | 24  |
| WL<sup>b</sup>          | 336      | 166 (49)                 | 43  | 15  | 25  |
| Q<sup>b</sup>           | 336      | 174 (52)                 | 47  | 16  | 24  |
| TobRV                   |          |                          |     |     |     |
| S<sup>b</sup>           | 359      | 246 (69)                 | 63  | 23  | 37  |
| CYMV                    |          |                          |     |     |     |
| Si<sup>b</sup>          | 457      | 304 (67)                 | 79  | 29  | 44  |
| ArMV                    | 300      | 202 (67)                 | 55  | 10  | 36  |
| VToMV                   | 365      | 248 (68)                 | 72  | 13  | 39  |
| SNMV                    | 377      | 274 (73)                 | 76  | 20  | 41  |
| LTSV                    |          |                          |     |     |     |
| A<sup>c</sup>           | 324      | 232 (72)                 | 65  | 11  | 40  |
| Ca<sup>c</sup>          | 322      | 220 (68)                 | 61  | 4   | 45  |
| Na<sup>c</sup>          | 324      | 230 (71)                 | 61  | 12  | 42  |
| TCV                     |          |                          |     |     |     |
| C<sup>d</sup>           | 356      | 198 (56)                 | 68  | 3   | 28  |
| CyRSV                   | 621      | 394 (63)                 | 93  | 12  | 92  |

<sup>a</sup> Helper virus abbreviations are as in Table 1.

<sup>b</sup> Virus strain.

<sup>c</sup> Satellite designation.
more extensively studied (2, 56, 105, 116, 127, 168). At least 28 isolates, as well as 15 cloned cDNA variants, have been sequenced (2, 26, 32, 56, 62, 79–81, 85, 99, 105, 115, 116, 118b, 127, 168, 183). Some of these sequence alterations affect the pathogenicity of the satellites in a host-specific manner (33, 89, 130, 153, 184). Other sequence variants contain sequence alterations that affect satellite replication (56, 153). Most sequence alterations, however, do not result in any discernible biological effect (2, 56, 105, 168). The sequence variation in CMV satellites is not random, but is concentrated in certain hot spots (Fig. 2). The sequence variation results in less conservation of the secondary structure in the lower half of the molecule (Fig. 1A) than in the upper half (55a).

Although most CMV satellite RNAs are 333 to 342 nt, some isolates from Japan are 369 to 386 nt (80, 127). The extra 30 to 40 nt in these larger CMV satellite RNAs are not inserted as a single block, relative to the smaller CMV satellite RNAs, but are interspersed between nt 110 and 160 (80, 127). These extra sequences are not related to CMV and therefore may originate from either some host plant or another viral genome.

The CMV satellite RNAs and RNAs C and D of TCV are the only satellite-like RNAs for which sequence variation has been observed in plants, within an evolving population derived from a homogeneous source (i.e., RNA transcripts of a cDNA clone) (19, 23, 25, 89, 118). Sequence variation was observed in one region of several satellites after either one or a few passages. For one CMV satellite RNA, this sequence variation was observed in any of five hosts, although the accumulation of the variant form was greatest in tobacco (118). The rate and nature of variation are determined by the particular satellite and the strain of helper virus (118, 171a). Some satellites do not appear to undergo any detectable variation in sequence (116, 118, 118a), although this may simply reflect a negative selection (see Specificity and Efficiency, below). Some satellites are not as efficiently replicated as others, and different helper virus strains can selectively replicate a particular satellite out of a mixture (89, 100, 153, 154a). Thus, satellite replication can be specifically influenced by minor nucleotide sequence and subtle secondary or tertiary structure changes.

**REPLICATION**

By definition, satellites are dependent on their helper viruses for replication. The helper virus, in turn, is dependent on the host plant to supply some components necessary for replication, and thus a complex three-way interaction between satellite, helper virus, and plant host is required for satellite replication. The details of this interaction have not been clearly elucidated in most instances, but some information is available about several systems.

**Specificity and Efficiency**

The specificity of satellite replication occurs at the level of both the helper virus and the host plant. With the satellite RNAs of TobRV and CMV, the apparent level of satellite replication varies widely with the strain of helper virus (86, 100, 140, 153, 171, 176). In addition, related viruses may or may not replicate the same satellite RNAs. Most CMV
satellite RNAs are replicated efficiently by the related cucumovirus tomato aspermy cucumovirus (TAV) (65, 70, 89, 120, 121, 140). However, PSV, the third member of the cucumovirus group, which often contains a satellite of its own (102, 146), will not support the replication of CMV satellite RNAs (102). Moreover, the PSV satellite RNA replication is not supported by CMV (102). Similarly, the TobRV satellite RNA is not supported by cherry leafroll nepovirus (159).

Among the sobemovirus satellites, the ability to replicate heterologous satellites is even more varied. SNMV does not support the replication of satellite RNA of LTSV (92), even though LTSV does support the replication of both SNMV (92) and SCMMoV (27) satellite RNA. Replication of LTSV satellite RNA also is supported by three unrelated sobemoviruses—sowbane mosaic, turnip rosette, and southern bean mosaic sobemoviruses (1, 42, 152). An early report on the inability of VTMoV and SNMV to support the replication of each other’s satellite RNA (63) must be reexamined, since in that study the helper viruses themselves did not replicate in the absence of the homologous satellite, in contrast to two more recent studies (44, 92, 93).

The tombusvirus satellites are more closely related than are their helper viruses, and tomato bushy stunt tombusvirus (TBSV) may support the replication of satellites isolated from any of the other tombusviruses (51, 52). In addition, TBSV satellite RNA replication can be supported by eggplant mottled crinkle tombusvirus (51).

For some viruses, the helper virus specificity has been partially mapped. Although the different serotypes of TBRV (S [Scottish] and G [German] serotypes) cannot support the replication of the heterologous satellite, pseudorecombinants between the S and G serotypes have been used to map the specificity of satellite replication to RNA 1 (unpublished work of A. F. Murant and J. H. Raschke cited in reference 47). The related nepovirus, grapevine chrome mosaic nepovirus, cannot support the replication of TBRV satellite RNA (36). Pseudorecombinants between grapevine chrome mosaic nepovirus and TBRV are viable, but neither combination will support the replication of the TBRV satellite RNA (36).

In addition to the helper virus specificity, the host plant often plays a significant role in the efficiency of satellite replication. The small satellite RNA of ArMV replicates very efficiently in Chenopodium quinoa (28). By contrast, the large satellite RNA of another nepovirus, TBRV, replicates poorly in C. quinoa but replicates very efficiently in Nicotiana clevelandii and Petunia hybrida (143). The TBSV satellite RNA replicates more efficiently in N. benthamiana than in N. clevelandii (51). With both CYMV satellite RNA (157) and CMV satellite RNAs (100, 153), replication in squash (Cucurbita pepo) is generally poor, whereas replication in tobacco is generally very efficient. In one instance, two closely related strains of CMV had dramatically different efficiencies of satellite replication in squash (171). This difference was mapped to RNA 1 of CMV and appears to involve the 3’ 600 nt of CMV RNA 1 (171a).

**Enzymology**

Since satellites are dependent on their helper virus for replication, the enzymology of satellite replication must depend largely on the enzymology of helper virus replication. Unfortunately, information on the replication of RNA plant viruses is minimal (reviewed in references 40, 68, and 131). Recently, however, significant progress has been made toward understanding viral RNA replication by the purification of a replicase complex from CMV-infected tobacco, which was capable of producing positive-stranded viral RNA from input positive-stranded template (75). This complex consists of two virally encoded proteins of Mr 111,000 and 95,000, which correspond to the gene products of CMV RNAs 1 and 2, respectively, and a single host-encoded protein of Mr 50,000 (75). As yet, however, no analysis of the replication of satellite RNA by this complex has been reported.

A few earlier studies, using partially purified replicase complexes from CMV-infected plants, have examined the replication of satellites in vitro. (i) A particulate fraction of tissue homogenates from CMV-infected cucumber cotyledons was shown to be capable of producing replicative forms of the viral and satellite RNAs in vitro (91). (ii) A cell-free system from CMV-infected tobacco was able to synthesize double-stranded (ds) RNA forms of both CMV and satellite RNA in vitro (199). (iii) A membrane-bound RNA-dependent RNA polymerase (RdRp) complex was isolated from tobacco protoplasts inoculated with RNAs 1 and 2 of CMV; this complex was capable of synthesizing CMV and satellite ds RNAs in vitro (148). Only low levels of this RdRp activity were found in the soluble fraction of either uninoculated protoplasts or those inoculated with any combination that did not include both RNAs 1 and 2 or CMV (148). The association of the CMV replicase with membranes is further supported by the observation of ds RNA forms of CMV and satellite RNA in vesicles of infected tobacco (34). Vesicles containing ds RNA were also found associated with protoplasts of TAV-infected tissue (74).

Replication of the satellite RNAs does not always involve the same mechanism as replication of the viral RNAs, and it seems likely that other factors which are specific for satellite replication are involved. In a template-dependent, cell-free system, RdRp isolated from CMV-infected tobacco utilized CMV satellite RNA as a template with about half the efficiency of the viral RNA (161), even though in tobacco plants infected by CMV and its satellite RNAs, the satellites are the predominant RNA species (100, 140). This suggests that purified RdRp complexes may be lacking a factor required for efficient satellite replication.

Protoplasts inoculated with TobRV and TobRV satellite RNA effectively replicated the satellite even in the presence of actinomycin D, indicating that synthesis of host RNA is not required for satellite replication (10). However, preincubation of the protoplasts with actinomycin D 24 h prior to inoculation effectively inhibited replication of TobRV satellite RNA (10), suggesting host factor involvement. No analysis of helper virus replication was done in this study, so it is not clear whether host RNA synthesis affects the helper virus differentially from how it affects the satellite.

Other lines of evidence suggest some differences in the mechanism of replication of satellite RNA versus helper virus RNA. (i) Biologically active cDNA clones of satellite RNAs of CMV (22, 115, 128) and TBRV (66) and RNA C of TCV (182) can tolerate extra sequences at either the 5′ or the 3′ end, but not both (unless only 1 or 2 nt) and are infectious whether or not the 5′ end is protected by a cap structure. By contrast, transcripts of cDNA clones of plant virus RNA show less tolerance, vis-à-vis biological activity, of extra nucleotides at the 5′ end and often require a cap structure for infectivity (30, 35, 76, 77, 90, 162, 170, 186, 200). (ii) Usually there is very little sequence identity at the 5′ and 3′ termini of satellites and their helper viruses (67, 78, 167), suggesting...
some differences in the mechanism of recognition for replication.

Self-Processing

Since some satellites produce concatemeric forms during replication (5, 11, 21, 82, 106, 113, 125, 199), there must be processing events to produce the “natural” monomeric linear or circular forms. In a few cases, these processing events can occur in vitro in the absence of proteins, i.e., autocatalytic cleavage (28, 39, 154a, 160). In other cases, either the processing events have not been studied, or the evidence suggests a mechanism other than self-cleavage (18). There are basically three types of self-processing satellites: (i) the linear nepovirus satellites that form circular and multimeric intermediates in vitro (106, 113); (ii) the circular sobemovirus satellites that form multimeric intermediates in vitro (21, 82); and (iii) the linear cucumovirus satellites that form multimeric intermediates in vitro (125, 199), but do not appear to have a circular intermediate (124).

Nepovirus satellites. The satellite RNA of TobRV has been cloned. Longer-than-unit-length transcripts of this satellite have been shown to undergo autocatalytic cleavage (15, 58, 60). The cleavage sites and sequences involved in the ribozyme function of both the positive- and negative-sense satellite RNA of TobRV have been mapped (13, 14). ArMV and CYMV-S1 satellite RNAs show similar sequences and possible secondary structures around the cleavage and catalytic sites (72, 173). The sequences on the positive-sense satellite RNAs can be folded into the so-called hammerhead structure common to a number of virus-derived ribozymes (39, 173). The sequences on the negative-sense satellite RNAs can be folded into a different common structure (known in different laboratories as the hairpin or paperclip structure) (72, 173). There appear to be a number of differences in the cleavage mechanisms of these two types of ribozymes (16, 37, 69, 72, 160), and both are being exploited to produce antiviral, catalytic molecules (59, 71).

A satellite with sequences and secondary structure similar to the nepovirus satellites has also been found in association with a luteovirus, barley yellow dwarf virus (135). RNA transcripts of this cloned satellite will also undergo self-processing in vitro (135).

Sobemovirus satellites. Circular satellite RNAs of LTSV, VTMoV, SNMV, and SCMoV have been shown to contain hammerhead ribozyme structures (28, 39). Partial dimeric RNA transcripts synthesized from cDNA clones of some of these satellites have been shown to be capable of autocatalytic cleavage (28, 39). A similar situation exists for the negative-sense RNA transcripts of only LTSV satellite RNA (28, 39). The similarities and differences in sequence and catalytic activities of these satellites are being used as a guide to determine the mechanism of cleavage and the actual structure of the catalytic entities. The somewhat conflicting data from different laboratories suggest that there may be more than one catalytic structure (38, 133, 174, 178).

Cucumovirus satellites. Whereas the multimeric forms of the nepovirus and sobemovirus satellites are believed to be generated from a circular template by a rolling-circle mode of replication (9, 82, 113), this does not appear to be the case with the multimeric forms of the satellites of cucumoviruses, TCV (a carmovirus), or CyRSV (a tombusvirus). For these satellites, no circular intermediates could be detected (11, 18, 125, 154a). In the case of CMV satellite RNAs, we have observed both self-cleavage and self-ligation of the ds RNA dimeric and monomeric forms, respectively, of the satellite RNAs isolated from infected plants, but no self-processing of single-stranded (ss) transcripts or encapsidated satellite RNAs (185a). Since large quantities of the ds RNAs are produced during CMV satellite RNA replication (96, 97, 101), these autocatalytic reactions may account for the multimeric RNAs produced during satellite replication (125, 199). It is also possible that these multimeric forms are not replicative intermediates, but merely represent the products of some atavistic process associated with these satellite RNAs.

EFFECT ON HELPER VIRUS

Alteration in Virus Titer

Although some satellites affect the titer of the helper virus, others do not (Table 3). Moreover, some satellites affect the titer of the helper virus in some host species, but not in others (see references in Table 3). Therefore the reported absence of any effect on the virus titer may be a consequence of an insufficiently broad survey. Nevertheless, the lack of any consistent effect suggests that complex interactions involving the host, the helper virus, and the satellite are involved in alterations of virus titer, as they are in other host-helper-satellite interactions. Hence, competition with the helper virus genome for its replicase may be only one possible mechanism of reducing the virus titer. In no situation described to date, however, has the presence of the satellite increased the titer of the helper virus.

In CMV satellite RNA, 12 conserved nucleotides comprising a loop and a few residues of the stem of a stable stem-loop structure are complementary to sequences in similar 5’-proximal structures of Q-CMV RNAs 1 and 2 (167). In vitro binding studies, however, detected stable complexes between satellite RNA and Q-CMV RNAs 3 and 4 (166). Therefore it has been suggested that CMV satellite RNA may regulate the expression of the coat protein gene from RNA 4 (166). However, using multiple strains of CMV and several CMV satellite RNAs, we were able to detect stable complexes involving CMV RNAs 2, 3, and 4 in various combinations, and none of these complexes inhibited the in vitro translation of coat protein from RNA 4 (40a).

Symptom Modulation

One of the most dramatic effects of satellites is to alter the symptoms induced by the helper virus. These alterations can be either an attenuation or an exacerbation of the virus-induced symptoms (Table 3). In fact, some satellite RNAs of CMV can ameliorate the symptoms induced by the helper virus on one host and intensify them on another host (61, 86, 129, 153, 197). The symptom modulation can also be affected by the particular strain of helper virus (61, 103, 153, 176). Thus, a three-factor interaction involving the particular satellite, the strain of helper virus, and the species of host determines the type of response.

In most cases, satellites attenuate the symptoms induced by different strains of helper virus. Exacerbation of symptoms is much rarer (Table 3). Attenuation of symptoms is usually accompanied by a reduction in virus titer. This has led to the suggestion that the competition for the replicase between the satellite and helper virus genomes results in a reduction in the concentration of the helper virus elicitor of host pathogenesis (96, 98, 100, 140, 176). However, in some satellite-helper virus combinations a reduction in pathogenicity was not accompanied by a reduction in the titer of the
TABLE 3. Effect of satellite RNAs on their helper viruses

| Helper virus* | Effect on virus accumulationb (reference) | Effect on symptomsc (reference) |
|---------------|------------------------------------------|---------------------------------|
| CMV          | –/0 (61, 86, 104, 140, 188)             | –/0+/0 (61, 86, 107, 140, 153, 188, 197) |
| PSV          | –/0 (146)                               | –/0 (98, 146)                   |
| TobRV        | S′ –/0 (15, 176)                         | –/0 (15, 58, 176)              |
|              | F′ – (164)                              | 0 (164)                         |
| TBRV         | 0 (47)                                  | 0 (143, 144)                    |
| CYMV         | L1* NR + (47)                           | 0 (157)                         |
|              | S1* NR –/0 (29)                         | 0 (29)                          |
| ArMV         | L1* NR                                  | 0 (47)                          |
|              | S1* NR                                  | 0 (157)                         |
|              | GBLV NR                                 | NR                             |
|              | GFLV – (49a)                            | – (49a)                         |
| PEV          | NR                                      | 0 (31)                          |
| VTMoV        | NR                                      | + (44)                          |
| SNMV         | 0 (92)                                  | + (92)                          |
| LTSV         | 0 (93)                                  | + (93)                          |
| SCMoV        | NR                                      | NR                             |
| TCV          | C′ 0 (4, 5, 123)                         | + (4, 5, 123)                   |
|              | D′ 0 (4, 5)                             | 0 (4, 5, 25, 181)              |
|              | F′ 0 (4, 5)                             | 0 (4, 5, 181)                  |
| TBSV         | – (51)                                  | – (51)                          |
| AMCV         | – (51)                                  | – (51)                          |
| CIRV         | – (51)                                  | – (51)                          |
| CyRSV        | – (11, 51)                              | – (51)                          |
| PAMV         | – (51)                                  | – (51)                          |
| PLCV         | – (51)                                  | – (51)                          |
| GRV          | C′ NR                                   | + (145)                         |
|              | G′ NR                                   | –/0+/0 (142)                    |
| BNYVV        | 3′ NR                                   | + (94, 95, 119, 190)            |
|              | 4′ NR                                   | 0+/0 (95, 190)                  |
|              | 5′ NR                                   | 0+/0 (190)                      |
|              | 6′ NR                                   | 0 (190)                         |

* Helper virus abbreviations are as in Table 1.

b –, decrease in virus accumulation; 0, no effect; NR, not reported.

c –, amelioration of symptoms; 0, no effect; +, exacerbation; NR, not reported.

d Virus strain.

e Satellite RNA designation.

Helper virus (70, 144). This implies that symptom attenuation by satellites probably involves more than one mechanism and may be an active process. However, it is not due to a generalized inhibition of the ability of the plant to respond to virus infection, since the satellite of one virus (CMV), while attenuating the symptoms of its helper virus, does not attenuate the symptoms of the nonhelper viruses tobacco mosaic tobamovirus, potato virus X potexvirus, or potato virus Y potyvirus in dual infections (194). This suggests that the satellite may interact with a helper component normally involved in virus-host interactions leading to pathogenicity, resulting in either a reduction or a blockage in such helper virus-host interactions.

Few satellites exacerbate the symptoms induced by the helper virus (Table 3). Of these, the best characterized are the satellite RNAs of CMV. Although most CMV satellite RNAs attenuate (to differing extents) the symptoms induced by CMV on all host species tested (86, 129, 140, 197), some satellites attenuate the symptoms of the helper virus on all but one or two hosts, on which the symptoms are exacerbated (85, 129, 140, 153, 197). Examples of symptom intensification include chlorosis on tobacco (and pepper) (129, 153, 183, 188), chlorosis on tomato (61, 153), and necrosis on tomato (107, 188). Sequences controlling chlorosis on either tobacco or tomato have been mapped to specific nucleotide changes between nt 135 and 175 of the CMV satellite RNA sequence (89, 130, 185a), with a single nucleotide (position 149) controlling the host specificity of chlorosis (185a). Similarly, sequences controlling necrosis induction have been mapped to between nt 290 and 310 of the CMV satellite RNA molecule (33, 130, 184). In addition, sequences flanking the above domains affect either the extent of pathogenesis or the range of helper viruses that support the induction of a particular pathogenic response (117, 153). Moreover, both necrosis and chlorosis involve interactions between the CMV satellite RNA and a factor(s) derived from CMV RNA 2, i.e., either the encoded 2a protein or RNA 2 itself (183, 185a). That is, whereas interactions between certain satellite RNAs and either CMV RNA 2 or the 2a protein induce severe pathogenic responses in some host species, on all other host species tested the same combination attenuates viral symptoms (183, 185a). The host component(s) involved in the above interactions is clearly a critical factor and the most refractory to analysis at this point. Nevertheless, determination of the nature of both the host factor(s) and the interaction with the viral components represent the next frontier of satellite research.

**Strategies for Virus Control**

The ability of satellites to attenuate the disease symptoms induced by their helper viruses has led to the suggestion that satellites may be useful as biological control agents of pathogenic molecules (97, 101, 176). Several approaches to test the viral control potential of satellites have been used (6, 55, 60, 70, 83, 87, 136, 191, 193, 194, 198).

**Satellite inoculation or spraying.** The application of mild strains of CMV containing satellite RNA to greenhouse and field crops has been evaluated. In several cases, CMV containing satellite RNA was able to protect plants to various extents against infection by more virulent strains either applied to the plants or introduced by natural infestation via the aphid vectors of CMV and its satellite RNA (55, 83, 87, 136, 191, 194, 198). Moreover, tomato plants so tested were also protected to a great extent, although not completely, against infection by CMV containing a necrogenic satellite RNA (55, 83, 136, 198). Thus, one can obtain variable levels of protection against CMV and pathogenic satellite RNAs in the field by using a CMV-plus-satellite "vaccine."

**Transgenic plants.** Infection by viruses containing satellites still results in some reduction in plant weight and/or crop yield, and such plants are reservoirs of virus for transmission to uninfected plants (136, 193, 194). Therefore an alternative strategy of protection against virus infection that makes use of the attenuation properties of satellites is to engineer plants that express satellites constitutively from the
plant genome (6, 60, 70, 84). Two satellites have been introduced into and expressed from the tobacco genome: the satellite RNAs of CMV (6, 70, 84) and TobRV (60). These plants show no adverse effects of low levels of satellite production. In both cases, inoculation of the transgenic plants with the respective helper virus resulted in an attenuation of virus-induced disease, as well as a reduction in the titer of the helper virus (6, 60, 70, 84). For transgenic tobacco plants expressing the CMV satellite RNA, inoculation with TAV resulted in an attenuation of TAV symptoms, although there was no reduction in the level of replication of TAV (70). Therefore these data indicate that satellites can function as co-biocontrol agents of their helper viruses, whether applied mechanically to the host plants or expressed as a transgene in the host plants.

Potential risks. Experiments involving the application of satellites to control virus disease are not without environmental risks (154). For most satellites it is not known whether there are any hosts that will react adversely to the presence of replicating satellite, since extensive host range studies have not been done. Even for satellites such as those of CMV, some of which are considered nonpathogenic, only about 40 of the 800 possible host species have been tested for potential adverse reactions (129, 140, 194, 197). Moreover, such studies have been done with only a few strains of helper virus. Since the pathology is also determined by sequences in the helper virus (103, 129, 153, 185), the number of combinations (helper-satellite-host species) that would have to be tested is astronomical. Even if one could be certain that the satellite were nonpathogenic on all host species in the presence of all helper virus strains, there is the possibility of mutations arising during satellite replication to generate pathogenic forms (118). In fact, we have observed such mutations (involving only a single nucleotide change) during passage of CMV satellite RNAs derived from cDNA clones (171a). Since the rate of mutation will be determined by the particular strain of helper virus and the host species used for propagation (each supplying components of the viral replicase [75]), the probability of converting a nonpathogenic satellite to a pathogenic one in the field may be higher or lower than that observed in experimental situations. Thus, both the application of a helper virus containing a satellite or the use of transgenic plants expressing the satellite are associated with potential biohazards. To this already identifiable risk, one can add two observations: (i) satellites can become associated with other helper virus strains during (natural) double infections (42, 149, 198); and (ii) once replicated and encapsidated by the helper virus, even genetically expressed satellites can become mobilized to non-target plants (84). Thus, the risk of dissemination of a molecule with a potential for becoming pathogenic is greatly increased. Nevertheless, various laboratories still consider satellites to be potentially useful biocontrol agents.

SATELLITELIKE MOLECULES

Recently, a few RNA molecules which appear to have many satellites-like qualities, but are not in fact true satellites, have been identified. By definition, satellite RNAs are not required by the helper virus for infection. At least two instances have been reported of RNA molecules which are not required for mechanical transmission/infection of the helper virus, but which are required for natural infection of the helper virus. RNAs 3 and 4 of beet necrotic yellow vein furovirus (BNYVV), were proposed to be satellites (47) but more recently have been shown to be required for either

spread in root tissue or transmission by the fungal vector of the virus (122, 189). However, RNAs 5 and 6 of BNYVV (see Table 1) may be true satellites (190). In addition, the small RNA molecule associated with GRV was originally reported to be a satellite (145) but recently was shown to be required for aphid transmission of the helper virus by the assistor virus (141).

In addition to these satellite-like RNA molecules, there is a chimeric molecule, RNA C of TCV, consisting of a DI-like RNA component and a satellite RNA component (181). TCV has two true satellites, RNA D and RNA F, which are virtually identical to the 5′ half of RNA C (181). However, the remaining sequences of RNA C have been derived from the helper virus genome, thus making this a unique RNA molecule with the properties of both a DI-like RNA and a satellite RNA.

ORIGINS

Although much speculation about the origin of satellites has been published (23, 24, 41, 108, 143, 197), there is little concrete evidence to support any theory. Hybridization studies have attempted to locate satellite sequences in the genomes of the helper virus hosts, generally without success (5, 6, 198a). However, the satellite sequences could exist in the plant genome in fragments, which, under appropriate conditions are brought together. Although many laboratories have reported the “spontaneous” appearance of satellites in experimental plants (25, 51, 57, 99, 115, 123, 147, 155, 157, 164, 183, 195, 197), most of these could be accounted for either by very low levels of satellites already existing in virus isolates, which are amplified under the appropriate conditions (i.e., experimental conditions), or by contamination of experimental plants with satellite. In one study of CMV satellite RNAs, the satellite RNA molecules were shown to be stable on tobacco plants for 25 days in the absence of helper virus (88). Since many satellites are highly structured (Fig. 1), they tend to be very stable and highly infectious, making an examination of their origin very difficult.

There have been two reports of the appearance of a satellite in plants inoculated with RNA transcripts of a helper virus derived from a cDNA clone; RNA C of TCV appeared after inoculation of plants with TCV genomic RNA transcripts (25, 123). By contrast, 20 serial passages of RNA transcripts of the For- CMV, RNAs in tobacco did not generate any satellite RNA (154a). Thus, the availability of biologically active cDNA clones of the helper virus genomes should make studies of the origin of satellites more feasible and may indicate different origins for different satellites.

CONCLUSIONS

Since their discovery in 1969, considerable progress has been made in identifying and characterizing new satellites, as well as localizing sequences important in various biological and biochemical functions. The dramatic effects that satellites can have on the symptoms induced by their helper viruses, as well as the limited sequence information which they contain, provide an ideal system for studying the relationship between RNA molecules and biological effects. Moreover, the apparent selection pressure for specific sequences, either by the helper virus or by the host plant, makes satellites an intriguing system for studies in molecular evolution. Satellites have also become a source of new and potentially useful (as well as litigious) ribozymes.

It seems certain that the number of known satellites will
increase as more plant viruses are purified and characterized. It is possible that such molecules will also be found in animal systems. The delta agent of hepatitis B virus has many properties of a satellite RNA (20, 196), although it also has some properties of a satellite virus. In addition, as more knowledge about satellites becomes available, it seems certain that more complex satellite-helper virus-host plant interactions will become apparent. New tools, such as the polymerase chain reaction and in vitro replication (75) systems, should enhance future research on satellites.

ACKNOWLEDGMENTS

We thank various colleagues for supplying unpublished data. Our unpublished work was supported by grant nos. DE-FG02-86ER13505 from the U.S. Department of Energy and 91-37303-6426 from the U.S. Department of Agriculture.

REFERENCES

1. AbouHaidar, M. G., and Y. C. Paliwal. 1988. Comparison of the nucleotide sequences of viroid-like satellite RNAs of the Canadian and Australian strains of lucerne transient streak virus. J. Gen. Virol. 69:2369–2373.
2. Avila-Rincon, M. J., C. W. Colmer, and J. M. Kaper. 1986. In vitro translation of cucumoviral satellites. I. Purification and nucleotide sequence of cucumber mosaic virus-associated RNA 5 from cucumber mosaic virus strain S. Virology 152: 446–454.
3. Avila-Rincon, M. J., C. W. Colmer, and J. M. Kaper. 1986. In vitro translation of cucumoviral satellites. II. CARN 5 from cucumber mosaic virus strain S and SP6 transcript of cloned (S)CARN 5 cDNA produce electrophoretically comigrating protein products. Virology 152:455–458.
4. Altenbach, S. B., and S. H. Howell. 1981. Identification of a satellite RNA associated with turnip crinkle virus. Virology 112:25–33.
5. Altenbach, S. B., and S. H. Howell. 1984. Nucleic acid species related to the satellite RNA of turnip crinkle virus in turnip plants and virus particles. Virology 134:72–77.
6. Baulcombe, D. C., G. R. Saunders, M. W. Bevan, M. A. Mayo, and B. D. Harrison. 1986. Expression of biologically active viral satellite RNA from the nuclear genome of transformed plants. Nature (London) 321:446–449.
7. Bock, K. R., A. F. Murant, and R. Rajeshwari. 1990. The nature of the resistance in groundnut to rossette disease. Ann. Appl. Biol. 117:379–384.
8. Bouzoubaa, S., G. Jonard, K. Richards, and C. Patz. 1985. Nucleotide sequence analysis of RNA-3 and RNA-4 of beet necrotic yellow vein virus, isolates F2 and G1. J. Gen. Virol. 66:1553–1564.
9. Branch, A. D., and H. D. Robertson. 1984. A replication cycle for viroids and other small infectious RNAs. Science 223:450–455.
10. Buckley, B., and G. Brüening. 1990. Effect of actinomycin D on replication of satellite tobacco ringspot virus RNA in plant protoplasts. Virology 177:298–304.
11. Burgyan, J., and M. Russo. 1988. Studies on the replication of satellite RNA associated with cymbidium ringspot virus. J. Gen. Virol. 69:3089–3092.
12. Burgyan, J., M. Russo, and D. Gallitelli. 1986. Translation of cymbidium ringspot virus RNA in cowpea protoplasts and rabbit reticulocyte lysates. J. Gen. Virol. 67:1149–1160.
13. Buzyayan, J. M., W. L. Gerlach, and G. Brüening. 1986. Non-enzymatic cleavage and ligation of RNAs complementary to a plant virus satellite RNA. Nature (London) 323:349–353.
14. Buzyayan, J. M., W. L. Gerlach, and G. Brüening. 1986. Satellite tobacco ringspot virus RNA: a subset of the RNA sequence is sufficient for autolytic processing. Proc. Natl. Acad. Sci. USA 83:8859–8862.
15. Buzyayan, J. M., W. L. Gerlach, G. Brüening, P. Keese, and A. R. Gould. 1986. Nucleotide sequence of satellite tobacco ringspot virus RNA and its relationship to multimeric forms.
16. Buzyayan, J., A. Hampel, and G. Brüening. 1986. Nucleotide sequence and newly formed phosphodiester bond of spontaneously ligated satellite tobacco ringspot virus RNA. Nucleic Acids Res. 14:9729–9743.
17. Buzyayan, J. M., J. S. McNinch, I. R. Schneider, and G. Brüening. 1987. A nucleotide sequence rearrangement distinguishes two isolates of satellite tobacco ringspot virus RNA. Virology 169:95–99.
18. Carpenter, C. D., P. J. Cascone, and A. E. Simon. 1991. Formation of multimers of linear satellite RNAs. Virology 183:586–594.
19. Carpenter, C. D., P. J. Cascone, and A. E. Simon. 1991. Mutations in a satellite RNA of turnip crinkle virus result in addition of poly(U) in vitro. Virology 183:595–601.
20. Chen, P.-J., G. Kalpana, J. Goldberg, W. Mason, J. Werner, J. Gerin, and J. Taylor. 1986. The structure and replication of the genome of hepatitis delta virus. Proc. Natl. Acad. Sci. USA 83:8774–8778.
21. Chu, P. W. G., R. I. B. Franci, and J. W. Randles. 1983. Detection, isolation and characterization of high molecular weight double-stranded RNAs in plants infected with velvet tomato mottle virus. Virology 126:480–492.
22. Collmer, C. W., and J. M. Kaper. 1986. Infectious RNA transcripts from cloned cDNAs of cucumber viral satellite RNAs. Biochim. Biophys. Res. Commun. 138:290–296.
23. Collmer, C. W., and J. M. Kaper. 1988. Site-directed mutagenesis of potential protein-coding regions in expressed cDNA of cucumber viral satellite RNAs. Virology 163:293–298.
24. Collmer, C. W., A. Hadidi, and J. M. Kaper. 1985. Nucleotide sequence of the satellite of peanut stunt virus reveals structural homologies with viroids and certain nuclear and mitochondrial introns. Proc. Natl. Acad. Sci. USA 82:3110–3114.
25. Collmer, C. W., L. Stenzler, N. Fay, and S. H. Howell. 1991. Nonmutant forms of the avirulent satellite D of turnip crinkle virus are produced following inoculation of plants with mutant forms synthesized in vitro. Virology 183:251–259.
26. Collmer, C. W., M. E. Toussaint, and J. M. Kaper. 1983. Cucumber mosaic virus-associated RNA 5 X. The complete nucleotide sequence of a CARN 5 incapable of inducing tomato necrosis. Virology 127:230–234.
27. Dall, D. J., D. J. Graddon, J. W. Randles, and R. I. B. Franci. 1990. Isolation of a subterranean clover mottle virus-like satellite RNA from lucerne infected with lucerne transient streak virus. J. Gen. Virol. 71:1873–1875.
28. Davies, C., J. Hammerschlag, and R. H. Symons. 1990. Structure, self-cleavage, and replication of two viroid-like satellite RNAs (virusoids) of subterranean clover mottle virus. Virology 177: 216–224.
29. Davies, D. L., and M. F. Clark. 1983. A satellite-like nucleic acid of arabis mosaic virus associated with hop netted disease. Ann. Appl. Biol. 103:439–448.
30. Dawson, W. O., D. L. Beck, D. A. Knorr, and G. L. Grantham. 1986. cDNA cloning of the complete genome of tobacco mosaic virus and production of infectious transcripts. Proc. Natl. Acad. Sci. USA 83:3099–3153.
31. Demler, S. A., and G. A. de Zoeten. 1989. Characterization of a satellite RNA associated with pea enation mosaic virus. J. Gen. Virol. 70:1075–1084.
32. Devic, M., M. Jaegle, and D. Baulcombe. 1989. Symptom production on tobacco and tomato is determined by two distinct domains of the satellite RNA of cucumber mosaic virus (strain Y). J. Gen. Virol. 70:2765–2774.
33. Devic, M., M. Jaegle, and D. Baulcombe. 1990. Cucumber mosaic virus satellite RNA (strain Y); analysis of sequences which affect systemic necrosis on tomato. J. Gen. Virol. 71:1443–1449.
34. Diaz-Ruiz, J. R., M. J. Avila-Rincon, and I. Garcia-Luque. 1987. Subcellular localization of cucumovirus-associated satellite double-stranded RNAs. Plant Sci. 50:239–248.
35. Domier, L. L., K. M. Franklin, A. G. Hunt, R. E. Rhoads, and J. G. Shaw. 1989. Infectious in vitro transcripts from cloned
cDNA of a potyvirus, tobacco vein motting virus. Proc. Natl. Acad. Sci. USA 86:3509–3513.
36. Doz, B., G. Macquaire, R. Delbos, and J. Dunez. 1980. Caractéristiques et rôle du RNA 3, RNA satellite du virus des anneaux noirs de la tomate. Ann. Virol. (Inst. Pasteur) Paris 131:489–493.
37. Feldstein, P. A., J. M. Buzayan, and G. Bruning. 1989. Two sequences participating in the autolytic processing of satellite tobacco ringspot virus complementary RNA. Gene 82:53–61.
38. Forster, A. C., C. Davies, C. C. Sheldon, A. C. Jeffries, and R. H. Symons. 1988. Self-cleaving viroid and new RNAs may only be active as dimers. Nature (London) 334:265–267.
39. Forster, A. C., and R. H. Symons. 1987. Self-cleavage of plus and minus RNAs of a viroid and a structural model for the active sites. Cell 49:211–220.
40. Fraenkel-Conrat, H. 1987. RNA-directed RNA polymerases of plants. Crit. Rev. Plant Sci. 4:213–226.
40a.Fraile, A., F. García-Arenal, and P. Palukaitis. Unpublished data.
41. Francki, R. I. B. 1985. Plant virus satellites. Annu. Rev. Microbiol. 39:151–174.
42. Francki, R. I. B., P. W. G. Chu, and P. K. Keese. 1983. The satellite nature of a viroid-like RNA from lucerne transient streak virus, p. 175–180. In H. D. Robertson, S. H. Howell, M. Zaitlin, and R. L. Malmberg (ed.), Current communications in molecular biology. Plant infectious agents. Viruses, viroids, virusoids, and satellites. Cold Spring Harbor Laboratory, Cold Spring Harbor, N.Y.
43. Francki, R. I. B., J. W. Randles, P. W. G. Chu, J. Robozinski, and T. Hatta. 1985. Viroid-like RNAs incorporated in conventional virus capsids, p. 265–297. In K. Maramorosch and J. McKeffer, Jr. (ed.), Subviral pathogens of plants and animals: viroids and prions. Academic Press Ltd., London.
44. Francki, R. I. B., C. J. Grivell, and K. S. Gibb. 1986. Isolation of velvet tobacco mottle virus capable of replicating with and without a viroid-like RNA. Virology 148:381–384.
45. Francki, R. I. B., J. W. Randles, T. Hatta, C. Davies, P. W. G. Chu, and G. D. McLean. 1983. Subterranean clover mottle virus: another virus from Australia with encapsidated viroid-like RNA. Plant Pathol. 32:47–59.
46. Fritsch, C., I. Koenig, A. F. Murant, J. H. Raschke, and M. A. Mayo. 1984. Comparison among satellite RNA species from five isolates of tomato black ring virus and one isolate of myrobalan latent ringspot virus. J. Gen. Virol. 65:289–294.
47. Fritsch, C., and M. A. Mayo. 1989. Satellites of plant viruses, p. 289–321. In C. L. Mandahar (ed.), Plant viruses, vol. I. Structure and replication. CRC Press, Inc., Boca Raton, Fla.
48. Fritsch, C., M. A. Mayo, and A. F. Murant. 1978. Translation of the satellite RNA of tomato black ring virus in vitro and in tobacco protoplasts. J. Gen. Virol. 40:587–593.
49. Fritsch, C., M. A. Mayo, and A. F. Murant. 1980. Translation products of genome and satellite RNAs of tomato black ring virus. J. Gen. Virol. 46:381–389.
49a.Fuchs, P. Personal communication.
50. Fuchs, M., M. Pinch, M. A. Serghini, M. Ravelonandro, B. Walter, and L. Pinch. 1989. The nucleotide sequence of satellite RNA in grapevine fanleaf virus strain F13. J. Gen. Virol. 70:955–962.
51. Gallitelli, D., and R. Hull. 1985. Characterization of satellite RNAs associated with tomato bushy stunt virus and five other definitive tombusviruses. J. Gen. Virol. 66:1533–1543.
52. Gallitelli, D., R. Hull, and R. Koenig. 1985. Relationships among viruses in the tombusvirus group: nucleic acid hybridization studies. J. Gen. Virol. 66:1523–1531.
53. Gallitelli, D., P. Piazolla, V. Savino, A. Quaquarelli, and G. P. Martelli. 1981. A comparison of myrobalan latent ringspot pot virus with other nepoviruses. J. Gen. Virol. 53:57–65.
54. Gallitelli, D., V. Savino, and O. A. deSeraur. 1983. Properties of a distinctive strain of grapevine Bundesliga latent virus. Phytopathology 73:27–32.
55. Gallitelli, D., C. Voylas, G. Martelli, M. S. Montasser, M. E. Tousignant, and J. M. Kapoor. 1991. Satellite-mediated protection of tomato against cucumber mosaic virus. II. Field test under natural epidemic conditions in southern Italy. Plant Dis. 75:93–95.
55a.García-Arenal, F. Personal communication.
56. García-Arenal, F., M. Zaitlin, and P. Palukaitis. 1987. Nucleotide sequence analysis of six satellite RNAs of cucumber mosaic virus: primary sequence and secondary structure alterations do not correlate with differences in pathogenicity. Virology 158:339–347.
57. García-Luque, I., J. M. Kaper, J. R. Díaz-Ruíz, and M. Rubio-Huertas. 1984. Emergence and characterization of satellite RNAs associated with Spanish cucumber mosaic virus isolates. J. Gen. Virol. 65:539–547.
58. Gerlach, W. L., J. M. Buzayan, R. I. Schneider, and G. Bruning. 1986. Satellite tobacco ringspot virus RNA: biological activity of DNA clones and their in vitro transcripts. Virology 151:172–185.
59. Gerlach, W. L., J. P. Haseloff, M. J. Young, and G. Bruning. 1990. Use of plant viral satellite RNA sequences to control gene expression, p. 177–184. In T. P. Pirone and J. G. Shaw (ed.), Viral genes and plant pathogenesis. Springer-Verlag, New York.
60. Gerlach, W. L., D. Llewellyn, and J. Haseloff. 1987. Construction of a plant disease resistance gene from the satellite RNA of tobacco ringspot virus. Nature (London) 328:802–805.
61. Gessenes, D., R. Provvedenti, and M. C. Edwards. 1982. Tomato white leaf: the relation of an apparent satellite RNA and cucumber mosaic virus. Phytopathology 72:1533–1538.
62. Gords, K. H. J., and R. H. Symons. 1983. Satellite RNA of cucumber mosaic virus forms a secondary structure with partial 3′-terminal homology to genomal RNAs. Nucleic Acids Res. 11:947–960.
63. Gould, A. R., R. I. B. Francki, and J. W. Randles. 1981. Studies on encapsidated viroid-like RNA. IV. Requirement for infectivity and specificity of two RNA components from velvet tobacco mottle virus. Virology 116:420–426.
64. Gould, A. R., and T. H. Hatta. 1981. Studies on encapsidated viroid-like RNA. III. Comparative studies on RNAs isolated from velvet tobacco mottle virus and Solanum nodiflorum mottle virus. Virology 109:137–147.
65. Gould, A. R., P. Palukaitis, R. H. Symons, and D. W. Mossop. 1978. Characterization of a satellite RNA associated with cucumber mosaic virus. Virology 84:443–455.
66. Greif, C., O. Hemmer, G. Demangeat, and C. Fritsch. 1990. In vitro synthesis of biologically active transcripts of tomato black ring virus satellite RNA. J. Gen. Virol. 71:907–915.
67. Greif, C., O. Hemmer, and C. Fritsch. 1988. Nucleotide sequence of tomato black ring virus RNA-1. J. Gen. Virol. 69:1517–1529.
68. Hall, T. C., W. A. Miller, and J. J. Bujarski. 1982. Enzymes involved in the replication of plant viral RNAs. Adv. Plant Pathol. 1:179–211.
69. Hampel, A., and R. Tritz. 1989. RNA catalytic properties of the minimum (−)TRSV sequence. Biochemistry 28:4929–4933.
70. Harrison, B. D., M. A. Mayo, and D. C. Baulcombe. 1987. Virus resistance in transgenic plants that express cucumber mosaic virus satellite RNA. Nature (London) 328:799–802.
71. Haseloff, J., and W. L. Gerlach. 1988. Simple RNA enzymes with new and highly specific endonuclease activities. Nature (London) 334:585–591.
72. Haseloff, J., and W. L. Gerlach. 1989. Sequences required for self-catalysed cleavage of the satellite RNA of tobacco ringspot virus. Gene 62:43–52.
73. Haseloff, J., and R. H. Symons. 1982. Comparative sequence and structure of viroid-like RNAs of two plant viruses. Nucleic Acids Res. 10:3681–3691.
74. Hatta, T., and R. I. B. Francki. 1981. Cytopathic structures associated with tonoplasts of plant cells infected with cucumber mosaic virus and tomato aspermy virus. J. Gen. Virol. 63:343–346.
75. Hayes, R. J., and K. W. Buck. 1990. Complete replication of a eukaryotic virus RNA in vitro by a purified RNA-dependent RNA polymerase. Cell 63:363–368.
76. Hayes, R. J., and K. W. Buck. 1990. Infectious cucumber mosaic virus RNA transcribed in vitro from clones obtained from cDNA amplified using the polymerase chain reaction. J. Gen. Virol. 71:2503–2508.

77. Heaton, I. A., J. C. Carrington, and T. J. Morris. 1989. Turnip crinkle virus infection from RNA synthesized in vitro. Virolology 178:214–218.

78. Hemmer, O., M. Meyer, C. Greiff, and C. Fritsche. 1987. Comparison of the nucleotide sequences of five tomato black ring virus satellite RNAs. J. Gen. Virol. 68:1823–1833.

79. Hidaka, S., K. Hanada, and K. Ishikawa. 1990. In vitro messenger properties of a satellite RNA of cucumber mosaic virus. J. Gen. Virol. 71:439–442.

80. Hidaka, S., K. Hanada, K. Ishikawa, and K. Miura. 1988. Complete nucleotide sequence of two new satellite RNAs associated with cucumber mosaic virus. Virolology 164:326–333.

81. Hidaka, S., K. Ishikawa, Y. Takeuchi, S. Kubo, and K. Miura. 1984. Complete nucleotide sequence of RNA 5 from cucumber mosaic virus (strain Y). FEBS Lett. 174:38–42.

82. Hutchins, C. J., P. Keese, J. E. Vissader, P. D. Rathjen, J. L. Mclanes, and R. H. Symons. 1985. Comparison of multimeric plus and minus forms of viroids and viroidoids. Plant Mol. Biol. 4:293–304.

83. Jacquesmond, M. 1982. Phénomènes d'interférences entre les deux types d'ARN satellite du virus de la mosaïque du concombre. Production des tombes vis-à-vis de la nécrose létale. C.R. Acad. Sci. (Paris) 294:991–994.

84. Jacquesmond, M., J. Anselem, and M. Tepfer. 1988. A gene coding for a monomeric form of cucumber mosaic virus satellite RNA confers tolerance to CMV. Mol. Plant Microbe Interact. 1:311–316.

85. Jacquesmond, M., and G. J.-M. Lauquin. 1988. The cDNA of cucumber mosaic virus-associated satellite RNA has in vivo biological properties. Biochem. Biophys. Res. Commun. 151:388–395.

86. Jacquesmond, M., and J.-P. Leroux. 1982. L'ARN satellite du virus de la mosaïque du concombre. II. Etude de la relation virus-ARN satellite chez diers hôtes. Agronomie 2:55–62.

87. Jacquesmond, M., and H. Lot. 1981. L'ARN satellite du virus de la mosaïque du concombre. I. Comparison of the aptitude to induce the nécrose de la tomate d'ARN satellites isolés de plusieurs souches du virus. Agronomie 1:927–932.

88. Jacquesmond, M., and H. Lot. 1982. L'ARN satellite du virus de la mosaïque du concombre. III. La propriété du survie in vivo. Agronomie 2:533–538.

89. Jaege, M., M. Devic, M. Longstaff, and D. Baulcombe. 1990. Cucumber mosaic virus satellite RNA (Y strain): analysis of sequences which affect yellow mosaics symptoms on tobacco. J. Gen. Virol. 71:1905–1912.

90. Janda, M., R. French, and P. Ahlquist. 1987. High efficiency T7 polymerase synthesis of infectious RNA from cloned brome mosaic virus cDNA and effects of 5' extensions on transcript activity. Virolology 188:259–262.

91. Jaspar, E. M. J., D. S. Gill, and R. H. Symons. 1985. Viral RNA synthesis by a particulate fraction from cucumber seedlings infected with cucumber mosaic virus. Virology 144:410–425.

92. Jones, A. T., and M. A. Mayo. 1984. Satellite nature of the viroid-like RNA-2 of Solanum nodiflorum mottle virus and the ability of other plant viruses to support the replication of viroid-like RNA molecules. J. Gen. Virol. 65:1713–1721.

93. Jones, A. T., M. A. Mayo, and G. H. Duncan. 1983. Satellite-like properties of small circular RNA molecules of lucerne transient streak virus. J. Gen. Virol. 64:1167–1173.

94. Jupin, I., S. Bouzoubaa, K. Richards, G. Jonard, and H. Guille. 1990. Multiplication of beet necrotic yellow vein virus RNA 3 lacking a 3' poly(A) tail is accompanied by reappearance of the poly(A) tail and a novel short U-rich tract preceding it. Virology 178:281–284.

95. Jupin, I., L. Quillet, U. Niesbach-Klöggen, S. Bouzoubaa, K. Richards, H. Guille, and G. Jonard. 1990. Infectious synthetic transcripts of beet necrotic yellow vein virus RNAs and their use in investigating structure-function relations, p. 187–203. In T. P. Pirone and J. G. Shaw (ed.), Viral genes and plant pathogenesis. Springer-Verlag, New York.

96. Kaper, J. M. 1982. Rapid synthesis of double-stranded cucumber mosaic virus-associated RNA 5: mechanism controlling viral pathogenesis? Biochem. Biophys. Res. Commun. 105:1014–1022.

97. Kaper, J. M. 1984. Plant disease regulation by virus-dependent satellite-like replicating RNAs, p. 317–343. In E. Kurstak and R. G. Marusyk (ed.), Control of virus diseases. Marcel Dekker, Inc., New York.

98. Kaper, J. M., and C. W. Collmer. 1988. Modulation of viral plant diseases by secondary RNA agents, p. 171–194. In E. Domingo, J. J. Holland, and P. Ahlquist (ed.), RNA genetics, vol. III. Variability in RNA genomes. CRC Press, Inc., Boca Raton, Fla.

99. Kaper, J. M., A. S. Duriat, and M. E. Tousignant. 1986. The 368-nucleotide satellite of cucumber mosaic virus strain Y from Japan does not cause lethal necrosis in tomato. J. Gen. Virol. 67:2241–2246.

100. Kaper, J. M., and M. E. Tousignant. 1977. Cucumber mosaic virus-associated RNA 5. I. Role of host plant and helper strain in determining amount of associated RNA 5 with viroids. Virology 80:186–195.

101. Kaper, J. M., and M. E. Tousignant. 1984. Viral satellites: parasitic nucleic acids capable of modulating disease expression. Endowments 8:184–200.

102. Kaper, J. M., M. E. Tousignant, J. R. Diaz-Ruiz, and S. A. Tolin. 1978. Peanut stunt virus-associated RNA 5: second tripartite genome virus with an associated satellite-like replicating RNA. Virology 88:166–170.

103. Kaper, J. M., M. E. Tousignant, and L. M. Geletka. 1990. Cucumber-mosaic-virus-associated RNA-5. XII. Symptom-modulating effect is codetermined by the helper virus satellite replication support function. Res. Virol. 14:487–503.

104. Kaper, J. M., M. E. Tousignant, and H. Lot. 1976. A low molecular weight replicating RNA associated with a divided genome plant virus: defective or satellite RNA? Biochim. Biophys. Res. Commun. 72:1237–1243.

105. Kaper, J. M., M. E. Tousignant, and M. T. Steen. 1988. Cucumber mosaic virus-associated RNA 5. XI. Comparison of 14 CARDNA 5 variants relates ability to induce tomato necrosis to a conserved nucleotide sequence. Virology 163:284–292.

106. Kaper, J. M., M. E. Tousignant, and G. Steger. 1988. Nucleotide sequence predicts circularity and self-cleavage of 300-ribonucleotide satellite of arabis mosaic virus. Biochim. Biophys. Res. Commun. 154:325–328.

107. Kaper, J. M., and H. E. Waterworth. 1977. Cucumber mosaic virus associated RNA 5: causal agent for tomato necrosis. Science 196:429–431.

108. Kaper, J. M., and H. E. Waterworth. 1981. Cucumoviruses, p. 257–332. In E. Kurstak (ed.), Handbook of plant virus infections and comparative diagnostic. Elsevier/North-Holland Biomedical Press, Amsterdam.

109. Kassanis, B. 1962. Properties and behaviour of a virus depending for its multiplication on another. J. Gen. Microbiol. 27:477–488.

110. Keese, P., G. Bruening, and R. H. Symons. 1983. Comparative sequence and structure of circular RNAs from two isolates of lucerne transient streak virus. FEBS Lett. 190:185–190.

111. Keese, P., and R. H. Symons. 1987. The structure of viroids and viroidoids, p. 1–47. In J. S. Semancik (ed.), Viroids and viroid-like pathogens. CRC Press, Inc., Boca Raton, Fla.

112. Kilbertis, P. A., and D. Zimmer. 1984. Translation strategy of solanum nodiflorum mottle virus RNA: synthesis of a coat protein precursor in vitro and in vivo. Nucleic Acids Res. 12:333–343.

113. Kiefer, M. C., S. D. Daubert, I. R. Schneider, and G. Bruening. 1982. Multimeric forms of satellite of tobacco ringspot virus RNA. Virology 121:262–273.

114. Konarska, M., W. Filipowicz, H. Domdey, and H. J. Gross. 1981. Binding of ribosomes to linear and circular forms of the 5'-terminal leader fragment of tobacco mosaic virus. Eur. J. Biochem. 114:221–227.
114. Kurath, G., P. Aeschleman, and P. Palukaitis. Unpublished observations.
115. Kurath, G., and P. Palukaitis. 1987. Biological activity of T7 transcripts of a prototype clone and a sequence variant clone of a satellite RNA of cucumber mosaic virus. Virology 159:199–208.
116. Kurath, G., and P. Palukaitis. 1989. RNA sequence heterogeneity in natural populations of three satellite RNAs of cucumber mosaic virus. Virology 173:231–240.
117. Kurath, G., and P. Palukaitis. 1989. Satellite RNAs of cucumber mosaic virus: Recombinants constructed in vitro reveal independent functional domains for chlorosis and necrosis in tomato. Mol. Plant Microbe Interact. 2:91–96.
118. Kurath, G., and P. Palukaitis. 1990. Serial passage of infectious transcripts of a cucumber mosaic virus satellite RNA clone results in sequence heterogeneity. Virology 176:8–15.
118a. Kurath, G., M. J. Roossinck, and P. Palukaitis. Unpublished data.
118b. Kurath, G., D. E. Slet, and P. Palukaitis. Unpublished data.
119. Kuzuzalas, M., V. Ziegler, S. Bouzoubaa, K. Richards, C. Putz, H. Guillel, and G. Jonard. 1986. Beet necrotic yellow vein virus: different isolates are serologically similar but differ in RNA composition. Ann. Appl. Biol. 109:155–162.
120. Lee, H. S. 1986. Modification of tomato aspermy virus symptom by cucumber mosaic virus-associated satellite RNA. Korean J. Plant Pathol. 2:145–149.
121. Lee, H. S., and J. Kummert. 1985. Induction of tomato necrosis by cucumoviruses, as related to specific interactions between genomic and satellite RNAs. Parasitica 41:45–55.
122. Lemaire, O., D. Merdinhogiu, P. Valentin, C. Putz, V. Ziegler-Graff, H. Guillel, G. Jonard, and K. Richards. 1988. Effect of beet necrotic yellow vein virus RNA composition on transmission by Polvmyxa betae. Virology 162:232–235.
123. Li, X. H., and A. E. Simon. 1990. Symptom intensification on cruciferous hosts by the virulent satellite RNA of turnip crinkle virus. Phytopathology 80:238–242.
124. Linthorst, H. J. M., and J. M. Kaper. 1984. Circular satellite RNA molecules in satellite of tobacco ringspot virus-infected tissue. Virology 137:206–210.
125. Linthorst, H. J. M., and J. M. Kaper. 1984. Replication of peanut stunt virus and its associated RNA 5 in cowpea protoplasts. Virology 139:317–329.
126. Liu, Y. C., U. T. Helen, J. I. Cooper, D. J. Bertoli, D. Coates, and G. Baur. 1990. The nucleotide sequence of a satellite RNA associated with arabis mosaic nepovirus. J. Gen. Virol. 71:1259–1263.
127. Masuta, C., Y. Hayashi, W. Q. Wang, and Y. Takanami. 1990. Comparison of four satellite RNA isolates of cucumber mosaic virus. Ann. Phytopathol. Soc. Jpn. 56:207–212.
128. Masuta, C., S. Kuwata, and Y. Takanami. 1988. Effects of extra 5' nonstructural bases on the infectivity of transcripts from a cDNA clone of satellite RNA (strain Y) of cucumber mosaic virus. J. Biochem. 104:841–846.
129. Masuta, C., S. Kuwata, and Y. Takanami. 1988. Disease modulation on several plants by cucumber mosaic virus satellite RNA (Y strain). Ann. Phytopathol. Soc. Jpn. 54:332–336.
130. Masuta, C., and Y. Takanami. 1989. Determination of sequence and structural requirements for pathogenicity of a cucumber mosaic virus satellite RNA (Y-sat RNA). Plant Cell 1:1165–1173.
131. Mathews, R. E. F. 1991. Plant virology, 3rd ed. Academic Press, Inc., San Diego, Calif.
132. Mayo, M. A., H. Barker, and D. J. Robinson. 1982. Satellite RNA in particles of strawberry latent ringspot virus. J. Gen. Virol. 63:417–423.
133. Mei, H.-Y., T. W. Kaaret, and T. C. Bruce. 1989. A computational approach to the mechanism of self-cleavage of hammerhead RNA. Proc. Natl. Acad. Sci. USA 86:9727–9731.
134. Meyer, M., O. Hemmer, and C. Fritsch. 1984. Complete nucleotide sequence of a satellite RNA of tomato black ring virus. J. Gen. Virol. 65:1575–1583.
135. Miller, W. A., T. Hercus, P. M. Waterhouse, and W. L. Gerlach. 1991. A satellite RNA of barley yellow dwarf virus contains a novel hammerhead structure in the self-cleavage domain. Virology 183:711–720.
136. Montasser, M. S., M. E. Tousignant, and J. M. Kaper. 1991. Satellite-mediated protection of tomato against cucumber mosaic virus. I. Greenhouse experiments and simulated epidemic conditions in the field. Plant Dis. 75:86–92.
137. Moser, B. A., M., and R. L. S. Forster, 1983. Lucerne transient streak virus RNA and its translation in rabbit reticulocyte lysate and wheat germ extract. Virology 128:176–185.
138. Mossop, D. W., and R. I. B. Francki. 1978. Survival of a satellite RNA in vivo and its dependence on cucumber mosaic virus for replication. Virology 86:562–566.
139. Mossop, D. W., and R. I. B. Francki. 1979. The stability of satellite viral RNAs in vivo and in vitro. Virology 94:243–253.
140. Mossop, D. W., and R. I. B. Francki. 1979. Comparative studies on two satellite RNAs of cucumber mosaic virus. Virology 95:395–404.
141. Murant, A. F. 1990. Dependence of groundnut rosette virus on its satellite RNA as well as on groundnut rosette virus. Virology 162:232–235.
142. Murant, A. F., and J. K. Kumar. 1990. Different variants of the satellite RNA of groundnut rosette virus are responsible for the chlorotic and green forms of groundnut rosette disease. Ann. Appl. Biol. 117:85–92.
143. Murant, A. F., M. A. Mayo, and A. A. Good. 1973. Evidence for two functional RNA species and a “satellite” RNA in tomato black ring virus. J. Gen. Virol. 19:275–278.
144. Murant, A. F., R. Rajeshwari, D. J. Robinson, and J. H. Ruschke. 1988. A satellite RNA of groundnut mosaic virus that is largely responsible for symptoms of groundnut rosette disease. J. Gen. Virol. 69:1479–1486.
145. Naidu, R. A., G. B. Collins, and S. A. Ghabrial. 1991. Symptom-modulating properties of peanut stunt virus satellite RNA sequence variants. Mol. Plant Microbe Interact. 4:268–275.
146. Nakashima, K., and Y. Ehara. 1989. Two satellite RNAs found in cucumber mosaic virus strain Y. Ann. Phytopathol. Soc. Jpn. 55:579–585.
147. Nitta, N., Y. Takanami, S. Kuwata, and S. Kubo. 1988. Inoculation with RNAs 1 and 2 of cucumber mosaic virus induces viral RNA replicase activity in tobacco mesophyll protoplasts. J. Gen. Virol. 69:2695–2700.
148. Okhi, S. T., H. Tanaka, and Y. Takanami. 1989. Cucumber mosaic virus satellite RNA transmissible to plants infected with a different isolate of CMV. Ann. Phytopathol. Soc. Jpn. 55:69–71.
149. Owens, R. A., and J. M. Kaper. 1977. Cucumber mosaic virus-associated RNA S II. In vitro translation in a wheat germ protein-synthesis system. Virology 80:196–203.
150. Owens, R. A., and I. R. Schneider. 1977. Satellite of tobacco ringspot virus RNA lacks detectable mRNA activity. Virology 80:222–224.
151. Paliwal, Y. C. 1984. Interaction of the viroid-like RNA 2 of lucerne transient streak virus with southern bean mosaic virus. Can. J. Plant Pathol. 6:93–97.
152. Palukaitis, P. 1988. Pathogenicity regulation by satellite RNAs of cucumber mosaic virus: minor nucleotide sequence changes alter host responses. Mol. Plant Microbe Interact. 1:175–181.
153. Palukaitis, P. 1991. Virus-mediated genetic transfer in plants, p. 140–162. In M. Levin and H. S. Strauss (ed.), Risk assessment in genetic engineering: environmental release of organisms. McGraw-Hill Book Co., New York.
154. Palukaitis, P. Unpublished data.
155. Piazzolla, P., D. Gallitelli, and V. Savino. 1982. Appearance of satellite RNA (CARN A 5) in six cucumber mosaic virus isolates from the open field. Phytopathol. Medit. 21:32–34.
156. Piazzolla, P., L. Rubinio, M. E. Tousignant, and J. M. Kaper. 1989. Two different types of satellite RNA associated with chiocory yellow mottle virus. J. Gen. Virol. 70:949–954.
157. Piazzolla, P., C. Vovlas, and L. Rubinio. 1986. Symptom regulation induced by chiocory yellow mottle virus satellite-like RNA. J. Phytopathol. 115:124–129.
158. Pinck, L., M. Fuchs, M. Pinck, M. Ravelonandro, and B.
159. Ponz, F., A. Rowhani, S. M. Mircetich, and G. Bruening. 1987. Cherry leafroll virus infections are affected by a satellite RNA that the virus does not support. Virology 160:185-190.

160. Prody, G. A., J. T. Bakos, J. M. Buzayan, I. R. Schneider, and G. Bruening. 1986. Autolytic processing of dimeric plant virus satellite RNA. Science 231:1577-1580.

161. Quadt, R., and E. M. J. Jaspars. 1991. Characterization of cucumber mosaic virus RNA-dependent RNA polymerase. FEBS Lett. 279:273-276.

162. Quillet, L., H. Guilley, G. Jonard, and K. Richards. 1989. *In vitro* synthesis of biologically active beef necrotic yellow vein virus RNA. Virology 172:293-301.

163. Randles, J. W., C. Davies, T. Hatta, A. R. Gould, and R. I. B. Franki. 1981. Studies on encapsidated viroid-like RNA. I. Characterization of velvety tobacco mottle virus. Virology 108:111-122.

164. Rezaian, M. A. 1980. Three low molecular weight RNA species detected in tobacco ringspot virus. Virology 100:400-407.

165. Rezaian, M. A., and A. O. Jackson. 1981. Low-molecular-weight RNAs associated with tobacco ringspot virus are satellites. Virology 114:534-541.

166. Rezaian, M. A., and R. H. Symons. 1986. Anti-sense regions in satellite RNA of cucumber mosaic virus form stable complexes with the viral coat protein gene. Nucleic Acids Res. 14:3229-3239.

167. Rezaian, M. A., R. H. V. Williams, and R. H. Symons. 1985. Nucleotide sequence of cucumber mosaic virus RNA 1. Presence of a sequence complementary to part of the viral satellite RNA and homologies with other viral RNAs. Eur. J. Biochem. 156:331-339.

168. Richards, K. E., G. Jonard, M. Jacquemond, and H. Lot. 1978. Nucleotide sequence of cucumber mosaic virus-associated RNA 5. Virology 89:395-408.

169. Riesner, D., J. M. Kaper, and J. W. Randles. 1982. Stiffness of viroids and viroid-like RNA in solution. Nucleic Acids Res. 10:5587-5598.

170. Rochon, D. M., and J. C. Johnston. 1991. Infectious transcripts from cloned cucumber necrosis virus cDNA: evidence for a bifunctional subgenomic mRNA. Virology 181:656-665.

171. Roossinck, M. J., and P. Palukaitis. 1991. Differential replication in zucchini squash of a cucumber mosaic virus satellite RNA maps to RNA 1 of the helper virus. Virology 181:371-373.

172a. Roossinck, M. J., and P. Palukaitis. Unpublished observations.

172. Rubinio, L., J. Burgyan, F. Greico, and M. Russo. 1990. Sequence analysis of cymbidium ringspot virus satellite and defective interfering RNAs. J. Gen. Virol. 71:165-160.

173. Rubinio, L., M. E. Tousignant, G. Steger, and J. M. Kaper. 1990. Nucleotide sequence and structural analysis of two satellite RNAs associated with chicory yellow mottle virus. J. Gen. Virol. 71:1897-1903.

174. Ruffner, D. E., S. A. C. Dahm, and O. C. Uhlenbeck. 1989. Studies on the hammerhead RNA self-cleaving domain. Gene 82:31-41.

175. Schneider, I. R. 1969. Satellite-like particle of tobacco ringspot virus that resembles tobacco ringspot virus. Science 166:1627-1629.

176. Schneider, I. R. 1977. Defective plant viruses, p. 201-219. In J. A. Romberger, J. D. Anderson, and R. L. Powell (ed.), Beltsville Symposia in Agricultural Research. I. Virology in agriculture. Alanheld, Osmun & Co., Montclair, N.J.

177. Schneider, I. R., R. M. White, and G. V. Gooding. Jr. 1972. Two new isolates of the satellite of tobacco ringspot virus. Virology 50:902-906.

178. Sheldon, C. C., and R. H. Symons. 1989. RNA stem stability in the formation of a self-cleaving hammerhead structure. Nucleic Acids Res. 17:5665-5677.

179. Simon, A. E. 1988. Satellite RNAs of plant viruses. Plant Mol. Biol. Rep. 6:240-252.

180. Simon, A. E., H. Engel, R. P. Johnson, and S. H. Howell. 1988. Identification of regions affecting virulence, RNA processing and infectivity in the virulent satellite of turnip crinkle virus. EMBO J. 7:2645-2651.