

8100–8400 nucleotides), while the smaller of the two, RNA-2, is much more variable in size between members of the genus, ranging in size from approx 1.4 to  $2.4 \times 10^6$  (3400–7200 nucleotides). The genus can be divided into two subgroups, according to the size of the RNA-2 component, those in Subgroups I and II having RNA-2 smaller or greater than 5400 nucleotides, respectively.

Virus particles, isolated from infected tissue, are usually, though not always, of three types: top (T), middle (M), and bottom (B), named originally according to their position following separation by sucrose gradient ultracentrifugation. Those viruses with RNA-2 molecules in the larger size range have T particles consisting of capsid protein only, M containing RNA-2, and B particles containing RNA-1. Those with RNA-2 components in the smaller size range have particles containing two molecules of RNA-2, in addition to those containing a single copy. In these cases, the sedimentation coefficient of the particles containing two copies overlaps that of particles containing RNA-1, so that the B component particles are of two types (4). Often, T particles are not present. In addition, multiple copies of a satellite RNA, if present, may also be encapsidated by viral coat protein.

Each RNA has a small virus-encoded protein (VPg) attached covalently to the 5'-terminus (the presence of which is essential for infectivity), and a poly(A) sequence at the 3'-terminus. Each has a single open reading frame (ORF), encoding a polyprotein that is proteolytically cleaved to produce a series of smaller functional proteins.

The genome organization of a representative nepovirus, tomato black ring (TBRV), is indicated in **Fig. 1A** and **C**. Nucleotide sequence data (5), and comparison with the genome sequences of the related como- and picornaviruses, suggests that RNA-1 ( $M_r 2.69 \times 10^6$ ) encodes proteins of  $M_r$  92, 72, 60, 23, and 2.3 kDa, which would be expected to be processed from a primary transcript of 254 kDa. Proteins identified from either in vitro translation experiments (6,7), or from the investigation of proteins produced in vivo (8), are also indicated in **Fig. 1B** and **D**. The sizes of the proteins in **Fig. 1B** and **D** are those derived from electrophoretic data, which may differ from the values predicted from the nucleotide sequence.

The capsid protein is represented by amino acids 838–1348 at the 3'-terminus of the primary translation product of RNA-2 (ref. 9;  $1.66 \times 10^6$ ), a polyprotein of 1357 amino acids,  $M_r$  150 kDa. It is produced from the 150-kDa polyprotein by cleavage to a 59-kDa protein, which then loses nine C-terminal amino acids, to become the 57-kDa capsid protein,  $M_r$  55,888 (10).

The function of some, but not all, the other proteins has been established. The 92-kDa protein is a component of the viral replicase, the 23-kDa protein is a protease, and VPg (2.3 kDa) becomes attached to the genomic RNAs. VPg is cleaved from the 120-kDa protein, although neither this protein nor the 92- and