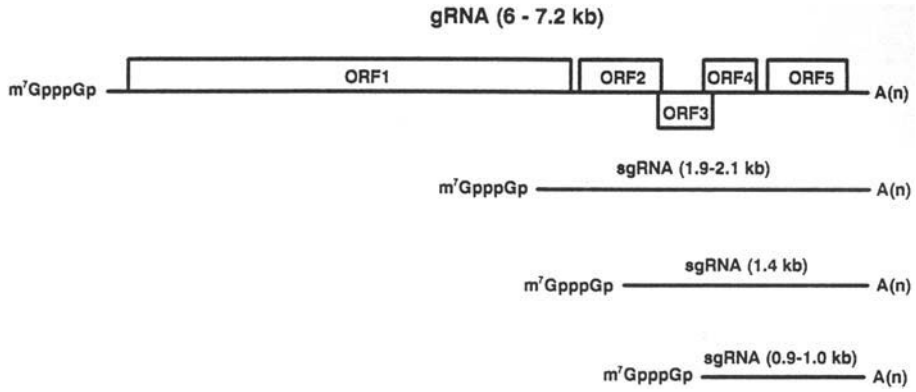

Fig. 1. General organization of a potexvirus genome and its subgenomic RNAs. Genomic organization of PVX, the type member of the potexviruses. Three subgenomic RNAs (sgRNAs) are shown below genomic RNA (gRNA). A(n) indicates the poly(A) tails at the 3' end of gRNA and sgRNAs. $m^7GpppGp$ represents the cap structure at the 5' end of both genomic and subgenomic RNAs. ORF1–ORF5 correspond to the five ORFs. ORF5 encodes the coat protein. Sizes of RNA in kb are indicated in parentheses.

However, recent information indicates that the CP of PVX could be expressed in vivo from a dicistronic message ***(51)***. In addition to the gRNA, several subgenomic RNAs (sgRNAs) of 0.9, 1.4, and 2.1 kb in length have been detected from plants infected with potexviruses (***12–15***; **Fig. 1**). These sgRNAs are capped and polyadenylated like the gRNA ***(13,16,17)***. The 5' ends of potexviral sgRNAs correspond to internal genomic regions; the 3' ends are coterminal with the genomic RNAs ***(13)***. The results of in vitro translation of sgRNAs suggest that the 25-kDa protein (ORF2) of PVX is expressed as a single translation product of the 2.1 kb sgRNA; both 12-kDa (ORF3) and 8-kDa (ORF4) proteins are expressed from the same 1.4-kb sgRNA, which appears to be functionally bicistronic ***(18)***. The CP can be efficiently translated from the 0.9 kb sgRNA ***(19)***.

2. Materials

2.1. Virus Purification

1. 0.1*M* Phosphate buffer, pH 7.2.
2. 0.1*M* Tris-borate acid buffer, pH 7.5.
3. *n*-Butanol.
4. Polyethylene glycol (PEG), mol wt 8000.
5. Sodium chloride.
6. Sucrose cushion: 30% Sucrose in 0.1*M* Tris-borate acid buffer, pH 7.5 (w/v).