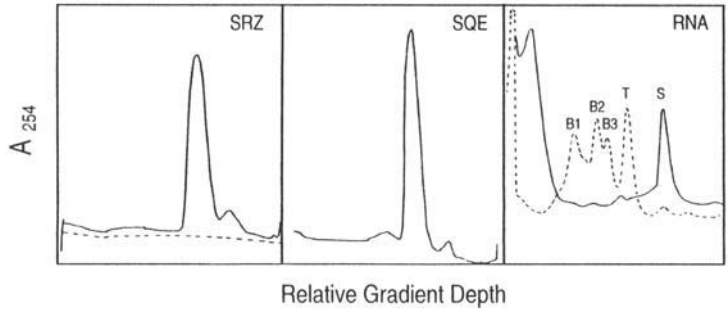

Fig. 2. Sedimentation of purified SYNV and viral RNA. SRZ panel: Patterns from SYNV-infected tobacco (solid line) and uninfected tobacco (dashed line) on rate-zonal sucrose gradients. The preparations were separated from tobacco and centrifuged in the gradients after clarification by Celite filtration and concentration by high-speed centrifugation. The particles have a sedimentation coefficient estimated at 1044 S. SQE panel: Banding of the particle recovered from SRZ in quasiequilibration sucrose gradients. The particles have a density of 1.18 g/mL in sucrose. RNA panel: Comparison of the sedimentation rates of SYNV RNA (S) with the three RNAs of brome mosaic virus (B-1, B-2, and B-3), and tobacco mosaic virus RNA (T). Note that the purity of the preparation can be determined from the amounts of the ribosomal RNAs sedimenting near BMV RNAs 2 and 3. Modified from **ref. *8***.

11. Centrifuge at 110,000*g* in a Beckman SW28 rotor for 30 min at 4°C.
12. Recover the major light-scattering band slightly more than halfway down the gradient (**Fig. 2**, SRZ) by use of a density gradient fractionator. Layer the recovered band over quasiequilibrium sucrose gradients made from 5.6-mL layers of 300, 400, 500, and 600 mg/mL sucrose in maintenance buffer.
13. Centrifuge for 1 h at 110,000*g* in a SW28 rotor at 4°C, and recover the light-scattering band (**Fig. 2**, SQE). Dilute the virus with an equal volume of maintenance buffer. Pellet at 90,000*g* in a Type 30 rotor for 30 min at 4°C. Quickly aspirate the supernatant from the pellet and resuspend in 0.5–1 mL of maintenance buffer with a Pasteur pipet.
14. The pellets should resuspend easily, and highly purified preparations will have a milky appearance. Optimum virus recovery is 2–5 A_{260} U/100 g of tissue, but the purified virus preparations exhibit considerable light scattering because of the membrane and size of the virions. Also, because the virions contain a low proportion of RNA (<2%), a prominent peak is not observed at 260 nm. However, the yields may vary, depending on the age of the plants, the environmental conditions under which the plants were grown, and the variables introduced during preparation.
15. Recovery of SYNV RNA is relatively straightforward by dissociation of virus by 1% sodium dodecyl sulfate (SDS) followed by sucrose density gradient centrifu-