Step 1. One g of tissue was macerated in 2 ml of 0.1 M potassium phosphate buffer, pH 7.5 (SP buffer). The resultant pulp was expressed through cheesecloth and the exudate was collected.

Step. 2. The exudate was clarified by stirring it (vol:vol) with a solvent mixture made from equal volumes of chloroform and *n*-butanol.

Step 3. The mixture was centrifuged at 12,200 × *g* for 5 min. The supernatant was retained.

Step 4. Polyethylene glycol (PEG) (Sigma, MW = 8000) and sodium chloride were stirred into the supernatant to give final concentrations of 6% PEG and 0.125 M NaCl, respectively. The suspension was allowed to stand for 15 min. (Note: as CVC is essentially a rapid, qualitative procedure, we have found it imperative to use drop counting throughout, eliminating many tedious measurements, to preserve the necessary simplicity. Thus, in the case of PEG/sodium chloride, we prepared in advance a stock solution of 30% PEG with 0.6 M NaCl. This was then added to the supernatant at the rate of 1 drop PEG/NaCl to 4 drops supernatant, giving a final concentration of 6% PEG and 0.125 M NaCl.)

Step 5. The suspension was centrifuged at 12,200 × *g* for 5 min, and the pellet was retained.

Step 6. The pellet was resuspended in SP buffer using frequent agitation for a minimum of 30 min. The resuspension of the pellet must be done with considerable care to obtain maximum virus yields.

(*a*) Following Step 5, the centrifuge tubes were first drained, and then the sides were wiped dry of supernatant with the aid of a cotton swab. Even small amounts of residual PEG may cause reprecipitation of some of the virus, which would then be subsequently lost at Step 7 (PEG traces may also interfere with the electron microscopy of the virus under study).

(*b*) It is important to allow sufficient time for resuspension and it is also important to provide agitation to aid in the disintegration of the pellet. A Vortex Mixer can normally be used, but resuspension may also be accomplished by teasing the pellet with the pointed end of a bamboo splint. Normally we have used a combination of these two procedures, and sometimes have resorted to using a Pasteur pipette as a slush pump. If the latter device is used, particular care must be exercised or the pellet(s) may become lodged in the pipette. Whatever procedure is used, allow at least 30 min with frequent agitation to ensure thorough resuspension of the virions.

(*c*) The amount of resuspension liquid gives a rough estimate of final concentration. As an example, if buffer is added to the pellets in Step 6 at a rate of 6 drops for each gram of starting tissue, then the concentration would be five to six times the original sample. For