from a defined source. Their main limitation is that the epitope to which they are raised is not conserved in all isolates of the virus, and false negatives result. The two types of antibody should be considered complementary and both have a place in plant virology in which their different qualities can be exploited.

Production of high quality antibodies requires pure virus preparation for immunization. It is important that the isolates used are representative of the virus to be studied. In cases in which serological variation is reported, it may be prudent to use more than one isolate for immunization.

For PAb production, the virus preparation should be as free as possible from contaminating plant proteins; this is not so critical in MAb production, because crossreacting clones can be eliminated in the screening process. However, the purification should avoid severe physical or chemical damage, which may expose hidden epitopes, because it is possible that an antibody could be raised that will not detect native virus.

New Zealand white rabbits are normally used for production of PAbs because they yield high volumes of serum and can be easily bled from the marginal ear veins. High serum levels of specific antibody (approx 10 mg/mL) can be achieved by hyperimmunization. As the immunizations are carried out across several weeks, class-shifting of antibody type occurs; the resulting serum predominantly contains antibodies of class G, rather than the class M, which dominate during the primary response ***(5)***. Plant viruses are relatively immunogenic, but an increased response can be obtained by the use of an adjuvant. Preparations such as Quil-A or the Ribi adjuvant system encourage good titers of antibody, are easy to administer, and have fewer side effects than the Freund's formulations.

For MAb production, it is usual to use Balb-C mice, the strain from which most of the myeloma cell fusion partners were derived. Female mice are preferred, because they do not fight when kept together. Immunization protocols vary, but hyperimmunity is required to produce sufficient primed splenocytes. Generally, the immune status of an animal is assessed by monitoring circulating antibody levels. Adjuvants can be used to improve the immune response, but should not be added to the final boost injection. Usually a batch of six mice are immunized, and the best responders are used for cell fusion (*see* **Note 1**).

Immunization and bleeding of animals is covered by legislation in most countries and must only be carried out by licensed personnel in licensed premises. The killing and removal of organs from mice is also subject to license in some countries, and it is usual that these procedures are carried out by animal technicians.

Although crude antisera preparations are useful for some purposes—e.g., for trapping virus particles on grids for electron microscopy—many serological techniques require the use of purified antibodies and antibodies that have