



Fig. 1. Western blot analysis of tobacco protoplasts transfected with an expression cassette encoding the PVS 33-kDa CP gene. The PVS CP is detected using commercially available antiserum. Lane 1, protoplasts only (nontransfected); lane 2, protoplasts transfected with recombinant expression cassette, PVS 33-kDa CP detected; lane 3, protoplasts transfected with expression cassette only (not containing the PVS CP gene).

plast viability can be determined by comparing the number of fluorescing protoplasts to the total number of protoplasts.

### 3.3. Detection of CP Expression by Western Blotting

Following PAGE and Western blotting, the protein of interest can be detected using a specific antiserum. The example shown in **Fig. 1** demonstrates the detection of the potato virus S (PVS) 33-kDa CP following its transient gene expression in tobacco protoplasts. The gene encodes the PVS CP and poly(A) sites. Western blots were carried out using specific PVS CP antiserum (many plant virus CP antisera are now commercially available) supplied by Bioreba, Switzerland.

## 4. Notes

1. Peeling of the lower epidermis is less difficult if fine-tipped tweezers are used in conjunction with peeling the epidermis initially from leaf veins, moving out over the leaf surface.
2. It is important not to place peeled sections on top of one another, because this reduces the surface area of cells on which the cell wall degrading enzymes can act.