The complete sequence of the resistance gene used, as well as sequences of all other genetic elements used, should be known prior to integration into the plasmid. On occasion, there are abnormalities associated with *Agrobacterium*-mediated transformation, and abnormalities may be much more common in plants produced via particle-gun transformation.

Initially, new transformants are assayed by Southern blotting to detect inserted DNA and to determine if the coding sequence is intact (correct size of the DNA) (Chapters 42 and 43). Northern assays confirm the presence of RNA, and, therefore, transcription (Chapter 44). Finally, the presence of, and correct size of, the translated protein is determined by Western blot analysis (Chapter 45) (**Note 4**).

In plants grown from seed, it is usually suitable to continue only with plants containing a single insert of the gene of interest. Proper segregation (3:1) of R_1 progeny indicates the presence of one insert. This and the number of copies in the insert can be confirmed molecularly by Southern blotting. DNA analysis (Southern blotting and PCR) allows detection of fragmented gene inserts or backbone sequences from the plasmid (particle gun transformation) or leaking plasmid left border sequences (*Agrobacterium*-mediated transformation) (Chapters 41–43). For breeding purposes, it may be important to determine in which chromosome and where the gene is inserted by various mapping techniques.

Gene stability through successive plant generations is a major concern. Stable expression and performance of the gene need to be confirmed in at least six generations to accept the line for practical use and breeding.

3.2.8. Nutritional Composition, Chemical Analysis, and Quality Assessment of the Final Product

Food safety of genetically modified foods is a critical issue in development of transgenic plants. Governments around the world regulate the commercialization of genetically modified crops and require a wide range of analyses to ensure their food and environmental safety. The World Health Organization (WHO) is playing a leading role in developing internationally accepted principles and procedures for the evaluation of safety of foods produced by biotechnology. Important principles concluded by the WHO in a recent international workshop ***(9)*** include the following: A new food or food component found to be substantially equivalent to an existing food or food component should be treated in the same manner as the existing food with respect to safety; where substantial equivalence could be established for a new food in all aspects except for the inserted gene or its product, the safety assessment should focus on the latter; and a determination that a new food was not substantially equivalent to an existing food does not mean that the new food is unsafe, but