

gating progeny can be used in initial screening. This will save time, but it must be understood that, in lines with a single gene insertion, 25% of progeny will not contain the transgene, 50% will be heterozygous for the gene (contain only a single copy), and 25% will be homozygous for the gene. Heterozygous plants may not be as resistant as homozygous plants among segregating progeny. If the species requires outcrossing for seed production, there may be a distinct advantage to eliminating susceptible and off-type lines at the vegetative stage, prior to selecting parents for homozygous seed production. This is particularly important if resistant phenotypes are relatively rare among the lines.

### 3.1.3. Growing and Transplanting In Vitro-Produced Plantlets

In vitro-produced plantlets normally cannot be transplanted directly in the field. They first must be transferred to sterile soil or artificial planting medium in small pots and then transplanted to the field after a period of growth and soil adaptation. It is convenient to use plastic or styrofoam flats for this purpose. The young plantlets in flats should be shaded from direct sunlight in the first few days after transplanting. Sanitation is very important. Flats should be placed on a pathogen-free surface, since *Pythium* and other damping-off fungi can invade the sterile planting media and kill the young plantlets rapidly, if the flats are placed on a contaminated surface. This can result in a complete loss of the planting. A balanced formulation of major and minor elements must be added, especially when artificial planting media are used, and the amount applied should increase as the plantlets grow and require more nutrients. Development of a strong root system is especially important, not only for a fast start once the plantlets are transplanted to the field, but also to prevent the planting medium from falling away from the roots as the plantlets are lifted from their pots and transplanted to the field (**Note 1**).

Plantlets should be transplanted from the flats to the field while they are growing vigorously. Transplanting machinery may be used for this purpose, but the process requires meticulous organization to prevent errors, when hundreds of small, replicated plots are involved (**Note 2**).

### 3.1.4. Plant Care During Growing Season

Most cultural practices, including irrigation, fertilization, and weed control, should be those used in commercial production of the crop.

### 3.1.5. Harvest and Storage

Small-plot harvesting without mixing between plots and without labeling mistakes is essential, but difficult to achieve. Careful planning and close supervision is essential. Appropriate storage is especially important in cases in which