1 Introduction

Watermelon, one of the most important vegetable crops, is eaten chiefly as a fresh fruit. It originated from tropical and subtropical Africa, and now is widely distributed throughout the tropics, Mediterranean region, South Asia, and East Asia including China.

Watermelon is attacked by many fungi, the most serious diseases in the tropics and in the United States being caused by Colletotrichum lagenarium, Phytophthora parasitica, Fusarium oxysporum, and Mycosphaerella melonis, etc. (du Cellier and Duke 1993). In addition, bacterial wilt (spread by cucumber beetles), squash bugs, and pickleworms are also common problems with cultivation of watermelon. Bacterial fruit blotch is a major concern in the US. It is thought to be a seedborne disease and is more serious in humid areas than in dry areas. Excess rain, heat, or drought can affect watermelon production. Excess rain during any stage of growth can reduce watermelon yields. Excessive heat and direct sunlight increase the likelihood of yield losses due to sunburn, which causes yellowing of the rind. Drought may reduce watermelon yields by diminishing plant growth, and limiting the development and size of the melons.

By using pathogen-resistant and stress-resistant varieties, watermelon yields can be increased. Several varieties, including Crimson Sweet, Jubilee, Dixilee, etc., have been developed by conventional breeding to have resistance to Fusarium wilt and anthranose. However, conventional breeding has limitations in improving the quality and productivity of watermelon. Recent advances in Agrobacterium-mediated transformation have made it possible to introduce foreign genes to improve their productivity and quality beyond the limit of conventional breeding. Using genetic transformation, pathogen-resistant and stress-resistant watermelons can be developed in a relatively short time. Furthermore, the sweetness of watermelon can be regulated through the introduction of enzymes which regulate carbohydrate content into the watermelon genome. Genetic transformation of watermelon has been reported by Choi et al. (1994). This chapter describes their transformation system for watermelon by co-culturing cotyledonary explants with Agrobac-
Transgenic Watermelon (*Citrullus lanatus*)

2 Genetic Transformation

Numerous studies have been conducted on various aspects of in vitro culture, i.e., micropropagation, organogenesis, somatic embryogenesis, production of triploids and tetraploids (Anghel and Rosu 1985; Srivastava et al. 1989; Dong and Jia 1991; Compton and Gray 1993a) etc., and the subject has been reviewed (Jelaska 1986; Adelberg et al. 1997). The studies on genetic engineering, however, are recent (Choi et al. 1994).

2.1 Plant Materials and Culture Conditions

Zygotic embryos of F₁ hybrid watermelon (*Citrullus lanatus*; cvs. Sweet Gem and Gold Medal) were dissected out of the mature seeds and surface-disinfected with 70% ethanol for 1 min and 1% sodium hypochlorite for 10 min. They were rinsed three times with sterile deionized-distilled water.

The basal MS medium used throughout the experiments consisted of Murashige and Skoog’s (1962) inorganic salts, 100 mg l⁻¹ myo-inositol, 0.4 mg l⁻¹ thiamine·HCl, 3% sucrose, and 0.4% Gelrite. The pH of all media was adjusted to 5.8 before autoclaving. Twenty-five milliliters of medium was dispensed into 87 x 15-mm plastic Petri dishes. Nine zygotic embryos were placed in each Petri dish containing the basal medium and incubated at 25°C in the dark. After 5 days incubation, seedlings 2- to 3-cm-long germinated, and their cotyledons were excised and transversely cut into proximal and distal halves (cotyledonary explants).

2.2 Induction of Adventitious Shoots

To induce adventitious shoots, cotyledonary explants of Sweet Gem and Gold Medal were placed onto medium supplemented with either 0.02, 0.2, 1, 2, 4 or 9 mg l⁻¹ 6-benzylaminopurine (BA) in Petri dishes. Two to four Petri dishes were cultured per treatment with four explants per dish. Cultures were maintained at 25°C in the light (16-h photoperiod, about 7 Wm⁻² cool-white fluorescent lamps) or in the dark. After 4 weeks of culture, the number of explants with adventitious shoots and the number of shoots formed per explant were counted under a dissecting microscope.

2.3 Transformation of *Agrobacterium*

pBI121, a binary vector carrying the CaMV 35S promoter-GUS gene-NOS terminator fusion and NOS promoter-neomycin phosphotransferase gene-NOS