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Identification of plant competent cells for highly efficient transformation system

Lim Youg Suk, Dong Ill Shin, Nam Jun Kang, Ill-Whan Sul *

Department of Biotechnology, Daegu University of Foreign Studies, HyupSeuk-Li, namCheun-Myun, KyungSan City, KyungPook 128-1, South Korea

Objectives

Plant cells or tissues must become competent prior to undergoing organogenesis or somatic embryogenesis. This can be accomplished under specific conditions such as growing cells and tissues on defined media supplemented with plant growth regulators (Christianson and Warnick, 1983 and 1987). During this period, they undergo active cell metabolism and division, and are amenable to incorporating delivered DNA into the host plant genome (Potrykus, 1990). It is likely that the frequency of transformation may be closely related to competence of cells for transformation events. There are no studies at the molecular level on cell competence and its relationship to regeneration and transformation in plant.

Materials and Methods

Leaf explants from tomato were cultured on an optimal regeneration medium (1/2 MS medium supplemented with Staba vitamins (Staba, 1969), sucrose (20 g.l⁻¹), myo-inositol (10 mg.l⁻¹), and BA (2 mg.l⁻¹), for different time periods of 3-day intervals up to 30 days. Following incubation in a medium containing BA, explants were transferred to a fresh basal medium without any BA. Each plate contained ~ 40 to 50 leaf segments derived cultured. Three plates were used for each treatment.

Results and Discussion

Leaf segments subjected to 3 days of culture on the regeneration medium developed adventitious buds; however, explants exposed to 6, 9, 12, and 15 days of culture on the regeneration medium showed an increase in shoot regeneration frequency over time. Explants grown for 15 days on the regeneration medium produced > 90 % regeneration frequency. Interestingly, maximum frequency of regeneration after 15 days of culture was appeared. Plant cells undergo mitotic division following cell division and differentiate into organized organs, and these developmental events are manipulated in vitro primarily by exogenous growth regulator composition and concentration (Christianson and Warnick, 1983; Coleman and Ernst, 1989). A three-day incubation period in a BA-medium induced adventitious bud formation; the shoot regeneration potential of leaf explants was optimal following a 15 day incubation with BA. Thus, cells during competent periods could be transformed maximally either abiotic and/or biotic transformation systems because of highly amenable to regeneration (Kouider et al., 1984; Patel et al., 1986).

* Corresponding author : Ill-Whan Sul, TEL: 053-810-7029, E-mail: iwsul@dufs.ac.kr