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Factors affecting direct somatic embryogenesis for pepper transformation

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Objectives

Current transformation protocols for pepper (*Capsicum annuum* L.) remain inefficient and limited to a few cultivar. This study was aim to develop of transformation system via somatic embryogenesis in pepper (*Capsicum annuum* L.)

Materials and Methods

1. Materials

Plants - Two inbred lines of *Capsicum annuum*.L. (Subi and Karac)

Explant -Immature zygotic embryo from seed in green fruit

Agrobacterium strain - EHA105/pIG121Hm

2. Methods

Hygromycin-resistant somatic embryos were induced directly from zygotic immature embryo explant without callus phase in MS solid medium containing 2mg/l 2,4-D, 20mg/l hygromycin, 400mg/l carbencillin, 100mg/l cefotaxime.

Results and Discussion

Pepper somatic embryos were induced directly from the apical region of immature zygotic embryo without involving intermediated callus in MS medium containing 2mg/l 2,4-D and regenerated into plantlets successfully in hormone-free MS medium. We established direct somatic embryogenesis using immature zygotic embryo.

In order to establish an efficient transformation system via direct somatic embryogenesis, several factors related to transformation were investigated. The induction rate of hygromycin-resistant somatic embryo was significantly influenced by immature zygotic embryo stage. When zygotic embryos at topedo stage (1-2mm) were used as explant, somatic embryo induction rate is approximately 55.4%. However, it is 12.5-19.5% in use of zygotic embryo at early or late cotyledon stage (4-5 mm or 7-8 mm).

The optimal concentration of *Agrobacterium* and co-cultivation time in pepper transformation via somatic embryogenesis using immature zygotic embryo was OD_{600} 0.8 and 3 days, respectively. The addition of L-cystein to the co-cultivation medium increased hygromycin- resistant somatic embryo induction from 25% in no L-cystein medium to 80% in 400mg/l L-cystein containing medium.

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