

Development of *Agrobacterium*-mediated Transformation of Gerbera (*Gerbera hybrida* Hort.)

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Objectives

To develop high frequency of transformation system for Gerbera, various conditions for transformation were probed. The probed conditions were 1) two different preparation of plant materials 2) three different kanamycin concentration, and 3) two different period of placement on the callus induction media.

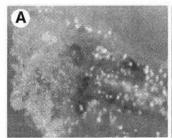
Materials and Methods

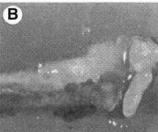
The plant materials were cut or naturally-torn 4 weeks old petioles from the plant grown *in vitro* on the media with half strength of MS salts. To find out the better condition for callus induction two different period of placement on the callus induction media, 7 or 10 days, were tested. And for best selection conditions three different concentration of kanamycin, 10, 20, 30ppm, also tested for. For transformation, *agrobacterium* strain LBA4404 harboring super virulent vector, pTOK233, were used. Actively growing *agrobacterium* were co-cultivated for 3 days with the prepared

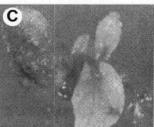
petioles in two different ways, cut or naturally-torn, and washed out throughly. The plant materials were transferred to callus induction media (MS salts and vitamins, 30 g of sucrose, 0.5 ppm of TDZ, 0.1 ppm of NAA, and 8 g of agar) for 7 days or 10 days, respectively. After induction of callus, the petioles were placed on the regeneration media (MS salts and vitamins, 30 g of sucrose, 1.0 ppm of BA, 0.1 ppm of IAA, 1.0ppm of zeatin, 300 ppm of cefotaxime, and 8 g of agar) with three different concentrations of kanamycin (10, 20, 30 ppm). GUS analysis was carried out to scrutinize the efficiency of transformation by different conditions.

Results and Discussions

Preparation of plant materials by cutting petiole showed better callus induction and plant regeneration than by naturally-tearing ones. Seven days of placement on callus induction media gave better callus induction and shoot development. The results from the experiment with three different kanamycin concentration, are currently investigating.







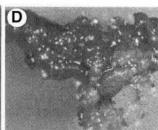


Figure 1. Transformation and regeneration from petiole segment of gerbera. A,Callus induction from lower cut surface of petiole; B, Shoot formation; C, Shoot development; D, GUS expression from transformed callus.