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Transformation of tobacco plants with OTSA gene.

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Objectives

Transformation of tobacco with Escherichia coli K12 trehalose-6-phosphate synthase gene OTSA.

Materials and Methods

1. Material:

Plant - Tobacco(Nicotiana tabacum).

Agrobacterium strain - LBA4404/ pCAMBIA 2300 containing otsA

2. Methods:

Callus induction, Co-cultured with *Agrobacterium tumefaciens* (LBA4404/pCAMBIA-2300-otsA), Regeneration, PCR.

Results and Discussion

Trehalose is the major disaccharide of microorganisms, fungi, and insects. Trehalose is synthesized from glucose-6-phosphate(G6P) and uridine-5-diphosphoglucose(UDPG) in a reaction catalyzed by trehalose phosphate synthase(TPS). Here we performed transfer of *otsA* gene encoding TPS in tobacco by *Agrobacterium tumefaciens* LBA4404 carrying pCAMBIA 2300 with *otsA* gene designated HHB1. Tobacco leaf callus were induced on MS medium containing 2 mg/ ℓ NAA, 0.5 mg/ ℓ Kinetin, 1g/ ℓ Casein, and 20g/ ℓ sucrose. Callus transformed *otsA* gene were regenerated whole plants on MS medium containing 2 mg/ ℓ NAA, 0.5 mg/ ℓ BA, 200 mg/ ℓ kanamycin, 250 mg/ ℓ cefotaxime, and 30g/ ℓ sucrose. In genomic DNA PCR, NPTII and *otsA* specific DNA fragments were amplified by their corresponding DNA primers, indicating that the transgenic tobacco plants were confirmed as otsA transformants. Further characterizations of *otsA*-transformed tobacco are undertaking.

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