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An efficient regeneration and *Agrobacterium tumefaciens*-mediated transformation systems for Mongolian bentgrass (*Agrostis mongolicum*)

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Objectives

The aim of this study was to establish an efficient regeneration and *Agrobacterium*-mediated transformation systems for Mongolian bentgrass (*Agrostis mongolicum*).

Materials and Methods

1. Materials:

- Plant: Mongolian bentgrass (*Agrostis mongolicum*)

- *Agrobacterium tumefaciens* strain EHA 105 harboring a binary vector pCUMB carries a target gene ABA-responsive element binding factor3 (*ABF3*), reporter genes green fluorescence protein - β -glucuronidase (*gfp-gus*), and a selectable marker gene hygromycin phosphotransferase (*hph*).

2. Methods: Callus induction → Callus type selection → Regeneration → Transformation → Regeneration/Selection → PCR → Southern blot analysis

Results and discussion

Selecting a callus-type was a critical factor to develop an efficient regeneration and transformation system, callus type II (whitish yellow color, green-spotted, and compact) was shown higher shoot regeneration and gene transformation ability than the other type of calluses. Based on the number of regenerated shoots per callus and their morphology, 0.05 mg l⁻¹ TDZ or 0.5 mg l⁻¹ BA in combination with 0.1 mg l⁻¹ NAA were chosen as the most suitable for shoot regeneration from calluses of Mongolian bentgrass. Transgenic plants were obtained from selection medium containing 50 mg l⁻¹ hygromycin, and thus transformation efficiency was 6.67%. The results of PCR and Southern blot analysis showed that the *ABF3* transgene was stably integrated into the genome of transgenic plants. These transgenic plants showed normal growth in terms of morphologies of whole plant.