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Application of *Agrobacterium*-mediated Transformation of Korean White-grain Wheat (*Triticum aestivum*) Varieties

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[Introduction]

The *Agrobacterium*-mediated transformation method is frequently used in plant gene editing since single-copy transgene integration is relatively high in *Agrobacterium*-mediated transformation compared to particle bombardment. However, there are still obstacles such as low embryogenic callus induction and unoptimized inoculation conditions in applying *Agrobacterium*-mediated transformation to Korean wheat varieties. Therefore, research to improve the efficiency of *Agrobacterium*-mediated transformation of Korean wheat varieties is essential. This study investigated the possibility of applying wheat transformation technology to Korean white-grain wheat varieties. In addition, the *GRF-GIF* chimera gene, which was recently reported to increase the regeneration efficiency, was cloned and applied from cv. Keumgang, the most produced variety in Korea.

[Materials and Methods]

The spikes were collected from 8 Korean white-grain wheat varieties (Yeonbaek, Dahong, Jokyeong, Baekjung, Hanbaek, Baekchal, Jeokjung, and Keumgang) grown in a greenhouse at Kongju University. The immature embryos were isolated from the collected immature grain and used for *Agrobacterium*-mediated transformation. The *ZmUbi* promoter::*GRF-GIF*::*tNOS* cassette was cloned into a pCAMBIA1300-based vector and inserted into *Agrobacterium* strain EHA105. The efficiency of wheat transformation was confirmed by measuring the transgene integration rate through PCR analysis.

[Results and Discussion]

After *Agrobacterium* inoculation with immature embryos, callus induction was performed in a medium containing hygromycin antibiotic for 4 weeks. After the selection for 4 weeks, 1 callus in cv. Baekjung and 4 calli in cv. Jeokjung survived. For other varieties, callus induction was additionally performed for 4 weeks after subculture, but callus did not grow. The calli derived from cv. Baekjung and Jeokjung were transferred to the regeneration medium to induce shoots for 4 weeks. A total of 10 shoots were regenerated in cv. Jeokjung, but no shoot was regenerated in cv. Baekjung. Leaves were collected from the 10 putative transformants and used for PCR to confirm transformants. As a result of PCR analysis, 5 putative transformants derived from 2 calli showed a positive reaction. The transformation efficiency of cv. Jeokjung was 1.16% (2/172). The putative transformants were transplanted into the soil after root induction, and the copy number will be confirmed later through Southern blot. This study can be useful information for the development of the transformation system using Korean wheat varieties.

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