

Genotype and procedure dependence of *Agrobacterium*-mediated transformation of *Brassica napus*

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Various experiments to optimize shoot regeneration and transformation efficiency were carried out with the oilseed rape cvs. 'Drakkar' and 'RS306', varying phytohormone combination and concentration and *Agrobacterium* strain. Compared to the slightly modified standard protocol of De Block et al. (1989) for hypocotyl segments was achieved no higher regeneration or transformation efficiency with alternative procedures or phytohormone combinations. The highest regeneration frequency, with up to 13 % regenerating hypocotyls, was obtained when De Block-A5 media were supplied with AgNO₃. Explants on medium without AgNO₃ turned brown or yellow and finally died. The addition of silver nitrate to the nutrient medium for control of ethylene levels was a pre-requisite for efficient shoot regeneration in all transformation experiments.

It is a well-documented phenomenon that the genotype has a strong influence on the regeneration frequencies of various tissue-culture techniques. This is also true for transformation. Following the modified protocol we have co-cultivated hypocotyl segments of the *B. napus* genotypes cv. 'Drakkar' and the resynthesised line 'RS306' with the *A. tumefaciens* strain GV3101 pMP90RK. The results of the experiments revealed a difference between the two genotypes regarding their regeneration response after co-cultivation and the efficiency of transgenic shoots. In 'RS306', a mean regeneration rate of 14.1 % was obtained in comparison to cv. 'Drakkar' with 10.1 %. The highest frequency of transformation in 'RS306' was 20.4 % to 1.2 % in cv. 'Drakkar'. These results show once again, that for each genotype favourable conditions must be found.

Large differences in regeneration and transformation efficiency was found within the genotype 'RS306'. The regeneration efficiency ranging from 8.0 % to 22.8 % in three experiments and transformation efficiency ranged from 6.0 % up to 20.4 %.

The use of *Agrobacterium* strain was another important factor in increasing of the transformation efficiency. Explants co-cultivated with the virulent *A. tumefaciens* strain ATHV C58C1 of nopaline type yielded higher efficiencies than those co-cultivated with GV3101 pMP90RK.

The results indicate the importance of the media supplements, the genotype and the virulence of different *A. tumefaciens* strains. Consequently, every given transformation protocol must be adapted to the genotype to allow the regeneration of a sufficient number of transgenic plants.