



CULTURE MEDIUM PH AFFECTS *AGROBACTERIUM TUMEFACIENS*-MEDIATED TRANSIENT EXPRESSION OF THE GUS GENE IN WHEAT IMMATURE EMBRYOS

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Agrobacterium tumefaciens-mediated transformation of wheat was the latest to be reported among the major agronomically important crops. As another monocots species, wheat is not naturally infected by *A. tumefaciens*, suggesting that there is a deficiency in one or more important factors for the occurrence of gene transfer. The knowledge of some bacterial requirements for this process has helped to increase the efficiency of genetic transformation in wheat. The acidic pH is reported as favorable to gene transfer to plants by *A. tumefaciens*, so the objective of this work was to analyze the pH effect on inoculation/co-cultivation step in wheat transformation. Immature embryos of Bobwhite SH9826 and PF020037 wheat genotypes were inoculated and co-cultivated in the medium with pH adjusted to 5.0, 5.3, 5.5 and 5.7. The treatments consisted of 100-120 explants inoculated and 30-40 explants not inoculated control. Immature embryos were collected from 13 to 14 days after anthesis (0.8 to 1.5 mm). The explants were inoculated for 15 minutes in a suspension of *A. tumefaciens* AGL1 strain containing plasmids pAL154/pAL156, and subsequently were transferred to the same medium for co-cultivation for three days in the dark at 22°C. Histochemical GUS assay was performed to verify the transient expression of the GUS gene. The variables analyzed were the number of embryos with blue spots (NEBS) and the number of blue spots per embryo (NBSE). The frequency of embryos with blue spots ranged from 0 to 22%. Bobwhite SH9826 showed blue spots only when medium pH was 5.0, being 7.1% NEBS and an average of 1.0 NBSE. PF020037 showed blue spots in medium with pH 5.0, 5.3 and 5.5, with 22.2%, 5.3%, 1.7% NEBS and 2.5, 0.8, 0.3 NBSE. Both genotypes did not show blue spots on pH 5.7. Inoculation/co-cultivation culture medium with pH 5.0 seems to be the best for *A. tumefaciens*-mediated wheat transient transformation. Based on these results, PF020037, which is a Brazilian genotype, is a better host for infection by *A. tumefaciens* than Bobwhite SH9826 under circumstances in which the experiments were performed. The explants that were not destroyed in histochemical assay are being cultured *in vitro* to verify the efficiency of stable transformation.