

AGROBACTERIUM-MEDIATED TRANSFORMATION IN DURUM WHEAT

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Durum wheat (*Triticum turgidum* L.) is an important cereal widely used for making pasta, typical bread and semolina. In recent years for many cereals, including durum wheat, biolistic transformation and direct gene transfer methods have allowed the introduction of useful agronomical traits. For the most important cereals, such as rice, maize, bread wheat, sorghum and barley is now possible to provide of an alternative system for genetic transformation based on the activity of *Agrobacterium tumefaciens* (Smith and Townsend) Conn. This mediated transformation has the great advantages over biolistic method to obtain an exact integration of few gene copies into the host genome, and a better stability of the transgene. Many variables can affect the *Agrobacterium*-mediated system, like *Agrobacterium* strain, the natural attitude of plant genotype, the co-cultivation conditions, and the selectable markers.

In this work we report our results concerning the establishment and optimization of a protocol for *Agrobacterium* mediated transformation of durum wheat. We tested different *in vitro* conditions on eight cultivars: Ancomarzio, Bronte, Duetto, Karalis, Neolatino, Ghibli, Sorrento and Vesuvio. The *Agrobacterium* strain AGL1, in different conditions of co-cultivation on 150 immature embryos for each cultivar, was used.

The activity of *gusA* gene was observed to assess the susceptibility of durum wheat cultivars to *Agrobacterium* infection. The qualitative histochemical assay of transient *gusA* gene expression, provides useful information on the efficiency of *Agrobacterium* mediated transformation.

The cultivars Vesuvio, Ghibli and Duetto showed many dark blue spots after ten day of *in vitro* culture. The cultivars Ancomarzio, Karalis Neolatino and Sorrento showed different reactions to the histochemical assay, depending on the auxin source used in the experiments, while the cultivar Bronte has always showed low level of coloration.

The cultivars Karalis and Neolatino showed a good level of regeneration and some T₀ plants were obtained. Four Karalis and two Neolatino T₀ plants were fertile and produced T₁ seeds. The T₂ generation was obtained from T₁ seeds. From T₂ plants the DNA was extracted and tested to verify the presence of Gus gene by qualitative PCR and Southern Blot analysis.