ANALYSIS OF COTTON α–GLOBULIN PROMOTER IN TRANSGENIC COTTON, TOBACCO, AND *ARABIDOPSIS*

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Abstract

Globulins are the most abundant seed storage proteins in cotton and therefore, the regulatory sequences controlling their expression represent a good source of seed-specific promoters. We isolated the promoter of cotton α-globulin B gene (Chlan *et al*. 1987) by gene walking using the primers designed from a cotton staged embryo cDNA clone. Using differentially digested Coker 312 genomic DNA fragments, it was possible to reconstruct 1150 bp of 5' flanking sequence of α-globulin B gene. The PCR-amplified α-globulin promoter (AGP) was fused to *gus*A in the binary vector pBI101.3 to create the test construct, pBIAGPGUS. Further, the expression pattern of AGP during seed development and seed germination was studied in the seeds from transgenic plants of *Arabidopsis* and tobacco that were obtained by *Agrobacterium*-mediated transformation. Histochemical analysis of GUS expression in the embryos isolated from seeds at various developmental stages revealed that α-globulin is expressed during seed maturation. Intense GUS staining was observed in the embryos isolated from dry seeds. In the germinating seeds, the GUS activity gradually declined until it disappeared after 7 days of seed germination. These results indicate that the α-globulin promoter is active in the late stages of embryo development.

We have regenerated transgenic cotton plants obtained by *Agrobacterium*-mediated transformation and grown these to maturity in the greenhouse. Developing embryos showed increasing levels of GUS activity as these progressed toward maturity. Fluorometric GUS assay in leaves, stem, floral buds and embryos isolated from dry seeds of the transgenic cotton plant revealed that the promoter expression is restricted specifically to the embryos. Our results on transient and stable expression of AGP>*gus*A indicate that the α-globulin promoter will be useful in controlling expression of transgenes in dicot seeds.

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