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Clinical Image Open Access

Gus A Gene Expression in Transgenic Tomato Plants Mediated *Agrobacterium tumefaciens*

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Clinical Image

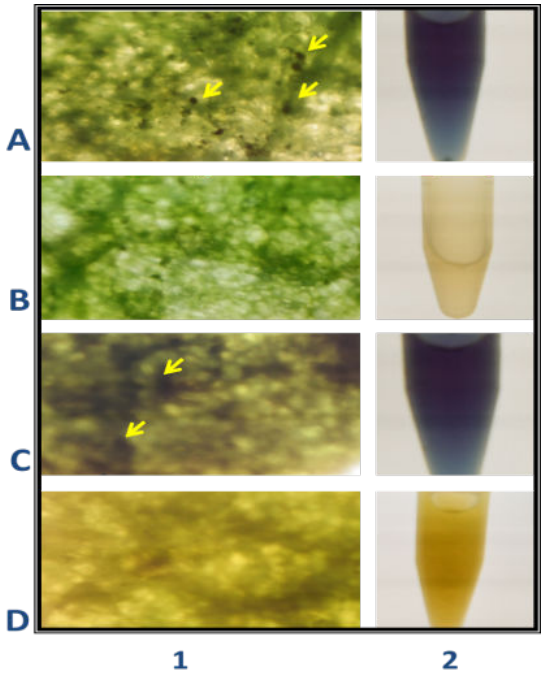

Figure 1: Expression of gus A gene in transgenic tomato and tobacco plants. Gus-assay showing β-glucuronidase activity in tissues (1) and their total protein extraction solutions (2) which showed blue color with transformed samples with X-Gluc assay while, control did not show a positive reaction. Transformed tomato. (B) Untransformed tomato (control). (C) Transformed tobacco. (D) Untransformed tobacco (control).

Technology of transgenic plants production will have an important and powerful impact on some of the immediate problems of vegetable crops, such as abiotic stresses and phytopathogens attack, and could reduce dependence on chemical pesticides and fungicides [1] (Figure 1).

Disarmed *Agrobacterium tumefaciens* bacterial strain LBA4404 harboring binary plasmid pROK–HAL1 (14.1 kb) was used with 6.5 kb T-DNA region. This plasmid contains the gus A (β-glucuronidase) reporter gene which is interrupted with an intron, the neomycin phosphotransferase gene (NPTII) conferring the kanamycin resistance under the control of nopaline synthesis gene promoter and transcription terminator site of cauliflower mosaic virus (CaMV) and hal I gene fused with the CaMV 35 S promoter, plus the termination site of CaMV.

Tomato and tobacco samples under study were assayed histochemically for Gus expression to confirm the identity of transgenic plants in the presence of X-GLUC A (5-bromo-4-chloro-3-indolyl-B-D-glucuronic acid) solution as described by [2]. To assess transient Gus expression, tissue samples of each treatment was placed in separate Eppndorf tubes containing small volumes of X-gluc solution (1 mM in 50 mM $NaHPO_4$. pH - 7.0 and 0.1% Triton x-100) in addition to Eppendorf tubes containing extractions of total soluble protein. Tubes were incubated for overnight at room temperature. Tissues were cleaned in 70% ethanol before observing the development of the blue color. The formed blue foci were observed under light microscope using normal light, also Gus reaction was detected in tissue extraction solutions under the same conditions used in explant treatments. These results proved that both Gus assay methods can be used in identification of the Gus gene.

References

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