

Detection of Transgenes in Cotton (*Gossypium hirsutum* L.) by using Biotechnology/Molecular Biological Techniques

Authors : Ahmad Ali Shahid, M Shakil Shaukat

Abstract : Agriculture is the backbone of economy of Pakistan and Cotton is the major agricultural export and supreme source of raw fiber for our textile industry. To combat against the developing resistance in the target insects and combating these challenges wholesomely, a novel combination of pyramided/stacked genes was conceptualized and later realized, through the means of biotechnology i.e., transformation of three genes namely, Cry1Ac, Cry2A, and EPSP synthase (glyphosate tolerant) genes in the locally cultivated cotton variety. The progenies of the transformed plants were successfully raised and screened under the tunnel conditions for two generations and the present study focused on the screening of plants which were confirmed for containing all of these three genes and their expressions. Initially, the screening was done through glyphosate spray assay and the plants which were healthy and showed no damage on leaves were selected after 07 days of spray. In the laboratory, the DNA of these plants were isolated and subjected to amplification of the three genes. Thus, seventeen out of twenty were confirmed positive for Cry1Ac gene and ten out of twenty were positive for Cry2A gene and all twenty were positive for presence of EPSP synthase gene. Then, the ten plant samples which were confirmed with presence of all three genes were subjected to expression analysis of these proteins through ELISA. The results showed that eight out of ten plants were actively expressing the three transgenes. Real-time PCR was also done to quantify the expression levels of the EPSP synthase gene. Finally, eight plants were confirmed for the presence and active expression of all three genes in T3 generation of the triple gene transformed cotton. These plants may be subjected to T4 generation to develop a new stable variety in due course of time.

Keywords : agriculture, cotton, transformation, cry genes, ELISA, PCR

Conference Title : ICAB 2015 : International Conference on Agriculture and Biotechnology

Conference Location : Jeddah, Saudi Arabia

Conference Dates : January 26-27, 2015