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One-for-all: a monoclonal antibody specific to different recombinant proteins in transgenic citrus plants

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The easy and rapid identification of a recombinant protein in transgenic plants is becoming increasingly relevant as more transgenic plants are used for research and commercial applications. Tagging recombinant proteins with a small peptide (epitope) can perform such a task using a variety of immunological methods. Epitope tags are short, hydrophilic peptide sequences recognized by specific antibodies. Compared with larger protein fusions, the small size of epitope tags makes them less likely to interfere with protein folding and function. We describe herein the detection of the c-myc epitope using different immunological methods in citrus transgenic plants. A c-myc tag sequence (N-EQKLISEEDL-C, corresponding to the C-terminal amino acids (410-419) of human c-myc protein) was added to the DNA sequence by PCR and the resulting proteins are being tested at the CREC. Our experiments with a genetically altered endogenous citrus gene modified to produce a protein with the c-myc tag demonstrate the utility of this technique for detection of trans-proteins in Citrus. Since this tag can be incorporated in the C terminal end of any protein, this technology simplifies different assays that require recognition by protein specific antibodies. We could detect different trans-proteins using the same antibody against the Myc epitope by ELISA or Western blotting. Moreover, expression of recombinant proteins bearing epitope tags can also eliminate the need of isolating proteins and producing antibodies for each new recombinant protein to be studied, which requires more cost and time, and can be problematic as a result of low antigenicity or high background cross-reaction with other proteins.

Our protocol would also accelerate the characterization of the transgenic plants in a timely fashion. This would help to isolate transgenic lines that produce optimum levels of the foreign trans-protein thereby improving and enhancing methods to evaluate and screen new priority transgenic commercial citrus scions and rootstock cultivars for resistance and/or tolerance to Huanglongbing, citrus canker and the Asian citrus psyllid.