TRANSFER OF THE ANTISENSE ACC-SYNTHASE GENE TO *Malus x domestica* cv. ROYAL GALA VIA *Agrobacterium tumefaciens* CO-CULTIVATION

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Genetic engineering is a powerful tool to generate artificial mutants of the genes of interest for physiological studies and plant breeding purposes. Ethylene is involved in several aspects of post-harvest plant metabolism and physiological disorders, and the biochemical steps of its biosynthesis have been extensively characterized. The enzyme 1-aminocyclopropane-1-carboxylate oxidase (ACCO, EC 1.14.17.4) is a cytosolic enzyme that catalyzes the last committed step in ethylene biosynthesis in higher plants. It is a key regulatory enzyme that catalyzes the precursor ACC from ethylene. In order to investigate the molecular mechanisms underlying fruit harvesting and conservation, we have introduced the antisense sequence of the *Malus x domestica* ACCO gene driven by a constitutive promoter in a high-ethylene producing apple cultivar, Royal Gala. We have co-cultivated leaf discs of *in vitro* plants of Royal Gala with the supervirulent *Agrobacterium tumefaciens* strain EHA105 carrying the transformation vector pGA643 with the antisense ACCO gene and the neomycin phosphotransferase (*neo*) gene that confers kanamycin resistance. The co-cultured explants were transferred to regeneration selective medium containing 22.7 μM thidiazuron (TDZ), 20 $\mu g.mL^{-1}$ of kanamycin and 500 $\mu g.mL^{-1}$ of cefotaxime. Approximately 90 days after co-cultivation, the regeneration of kanamycin resistant shoots was observed from the leaf discs. Seventeen shoots were obtained from 500 co-cultured explants, providing a transformation efficiency of approximately 3.4%, which is similar to previously reported results. The shoots were transferred to rooting medium and will be further molecularly and physiologically characterized. The plants exhibiting reduced expression of endogenous ACCO will consist in important tools to investigate the role of ethylene in apple fruit physiology and metabolism.