

## DEVELOPMENT AND MAPPING OF FUNCTIONAL MOLECULAR MARKERS FOR FRUIT QUALITY TRAITS IN *MALUS X DOMESTICA*

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The understanding of the genetic mechanism underlying fruit quality traits in apple is one of the main steps to improve marker-assisted selection and breeding of apple cultivars. Therefore, molecular markers derived from fruit modulated EST and from other candidate genes controlling fruit traits are a useful tool to map genes and QTL involved in fruit development and quality. In order to develop functional markers for fruit quality traits we followed two main approaches. On the one hand, the public sequence database was screened for transcription factors and 425 *Malus* EST, putatively involved in DNA binding or transcript regulation, were identified. These genes are supposed to play a key role in fruit quality related metabolic pathways: 28 microsatellite containing sequences were tested and 13 were placed on 'Fiesta' x 'Discovery' reference map. On the other hand, in a previous microarray analysis performed in our lab, 300 genes, differentially expressed during different fruit developmental stages, were identified. The corresponding EST sequences provided the second source of functional markers and were assembled to identify unique transcripts: 153 sequences resulted to be unique while 147 redundant sequences were assembled in 36 contigs. In total 189 sequences were screened for the presence of microsatellites (SSR), the sequences not containing repeated elements were BLAST searched against PlantGDB-assembled Unique Transcript database (PUT) and the resulting PUTs were analyzed to identify repeated motif. Furthermore, insertion/deletion (INDEL) and single nucleotide polymorphism (SNP) were searched in the assembled sequences. Finally, more than 50 PCR primer pairs were designed flanking SSR, INDEL or SNP containing regions or including most of the differentially expressed sequences and, in case of such differences were not identified, sequence reactions on parental genotypes were performed. SNP detection was performed digesting PCR products with different restriction enzymes while SSR and INDEL were scored as length polymorphism. Twenty-seven polymorphic markers were placed on 'Fiesta' x 'Discovery' or 'Fiesta x Prima' reference maps, increasing the total number of genes mapped in *Malus x domestica* to 40.

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