

Marker-assisted selection (MAS) is one of the main applications of genomic research to breeding. We describe the initial steps taken by the IRTA-ASF peach breeding program towards its routine selection for fruit shape (round, flat and aborting) and fruit acidity (acid vs. subacid).

Integration of MAS into a breeding program requires: a) the identification of alleles tightly linked to target traits, b) the integration of the genotyping step to the work pipeline of the breeding program and c) the adaptation of high-throughput protocols for DNA extraction and genotyping.

We decided to start MAS with fruit shape and fruit acidity, two simply inherited traits already mapped in the *Prunus* reference map and that are key breeding targets. Linked alleles to these genes were found based on single-family analysis and later validated using wide germplasm collections. In very few cultivars the identified molecular markers were not useful. Once the appropriated alleles were identified, molecular markers were genotyped during two years in different progenies of the breeding program. We started with plants that were already in the field producing fruits to estimate selection efficiency. After genotyping and phenotyping more than 600 individuals for each trait, the prediction was more than 95% accurate. The second year MAS was applied at the seedling stage with 192 individuals from different crosses. All plants were transferred to the field ordered by phenotype predictions. Also in this plant case prediction was correct more than 95% of the times. We are currently starting to use MAS for discarding unwanted genotypes without any further testing.

### **Functional characterization of fire blight resistance in *malus fusca* - fine mapping, cloning and characterization of Fire Blight resistance genes from *Malus fusca***

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Fire blight (FB), caused by the gram-negative bacterium, *Erwinia amylovora* (Ea), is the most important bacterial disease on pome fruit worldwide including apple. Although the use of antibiotics as a control measure has been successful to some degree, their application is unwanted, forbidden or strictly regulated in many countries due to the threats they pose to the environment. Hence, there is the need for an alternative means of control. Recently, research has shown that genetic resistance can play a vital role in the management of fire blight disease. In different wild species of *Malus*, several potential sources to FB resistance have been identified and as a result, some wild apple accessions have been utilised as sources of resistance. Consequently, different quantitative trait loci (QTLs) for FB resistance have been identified by Peil et al. 2007;

Durel et al. 2009, and Parravicini et al. 2011 and also by Calenge et al. 2005; Khan et al. 2006; in wild apple accessions and cultivated apple (*Malus x domestica*) respectively. These QTLs exhibit different levels of resistance to the disease. With the aim of identifying, isolating and characterizing resistance to fire blight from the wild species *M. fusca*, we report here the first results of QTL analysis and marker development for fine mapping. F1 progenies of *Malus fusca* x Idared were grafted and inoculated in the greenhouse with *E. amylovora* strain Ea222 in two consecutive years. DArT, SSR and SNP markers were developed and used to establish a genetic map and to perform QTL analysis. A major QTL could be localised on LG10. Two DArT markers were closely linked to the QTL. Four SSR markers linked to resistance were developed using the reference sequence of Golden Delicious for fine mapping the QTL. This research is in progress and additionally a bacterial artificial chromosome (BAC) library of *M. fusca* will be screened with SSRs flanking the QTL.

### **Analysis of flavonoid regulation in Rosaceae**

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The flavonoid pathway provides phenolic compounds that play major roles in Rosaceous fruit colour. These compounds also determine many of the dietary health-related properties in both the peel and cortex. Regulation is primarily conducted by a complex of transcription factors (TFs), central to which are the MYB TFs. Using examples from peach and apple, we provide functional analysis for the control of flavonols, proanthocyanidins (PAs) and anthocyanins by the MYB family. In peach, the pattern of UFGT gene expression correlates with TFs which up-regulate anthocyanin biosynthesis (MYB10 and bHLH3), or repress (MYB111 and MYB16) transcription of biosynthetic genes. The expression of a potential PA-regulating transcription factor, MYBPA1, corresponds with PA levels. Functional assays show that MYB10 positively regulates the promoters of UFGT and DFR but not LAR. In contrast, MYBPA1 trans-activates the promoters of DFR and LAR, but not UFGT, suggesting exclusive roles of anthocyanin and PA regulation. In apple the flavonoid biosynthetic pathway is most active in the skin, with the flavan-3-ols, catechin and epicatechin, acting as the initiating units for the synthesis of PA polymers. We have examined the genes involved in PA production in different apple cultivars and show that flavan-3-ol biosynthesis is under the control of biosynthetic enzymes ANR and LAR1. These steps are under the control of developmental and environmental stimuli, such as temperature and light. Heating fruit rapidly reduces expression of the transcriptional activation complex responsible for red skin colour, while a single night of low temperatures is sufficient to elicit a large increase in transcription of MYB10 and consequently the biosynthetic pathway.

### **Analysis of a TFL1 homologous gene and its relationship to the inflorescence variation in Japanese flowering cherries**

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