Harnessing the power of RosBREED: Development, validation, and application of DNA tests for predicting peach flavor and other valuable rosaceous tree fruit traits

**Peach fruit quality DNA tests – Development to Deployment**

DNA tests that predict valuable trait levels are essential for widespread adoption of marker-assisted breeding (MAB) of rosaceous tree fruit. The RosBREED project has facilitated development of DNA tests for important traits in peach, apple, and cherry, including peach sweetness and acidity. Based on a quantitative trait locus discovered on peach chromosome 7 by RosBREED collaborators explaining ~20% of phenotypic variation for titratable acidity (TA) in normal acid peaches and ~10% of variation in soluble solids content (SSC), a DNA test (G7Flav-SSR) was developed at Washington State University (WSU). The G7Flav-SSR DNA test was converted from a SNP array-based marker with six diplotypes to a PCR-based SSR marker with three functional genotypes whose effects were confirmed within RosBREED Peach Demonstration Breeding Programs and validated within the University of Arkansas Peach Breeding Program (UA). Although this DNA test can be used alone, when combined with the D locus DNA test (CPPCT040b) the predictive power of the combined DNA test makes it a valuable tool for MAB. This combined DNA test can be used to enhance breeder confidence in selecting parents and seedlings with specific, valuable TA and SSC levels, thereby increasing overall breeding efficiency.

**Development**

- The G7Flav-SSR DNA test was developed through marker conversion from the RosBREED 9K SNP array with 6 diplotypes to a PCR-based SSR marker with 8 SSR genotypes (1), converted to a high-throughput format (2), and combined with the D locus CPPCT040b DNA test (3).

**1. Marker conversion**

- Although low acidity could be differentiated from high acid, no test alone could differentiate medium acid. Therefore, multiplexing was required.

**2. DNA employed**

- Once multiplexed, all SNP diplotypes were represented by SSR alleles. But, for MAB utility high-throughput conversion was required.

**3. Test for maximum predictability read for validation**

- To maximize MAB utility, G7Flav-SSR was combined with the acidity CPPCT040b DNA test (low acid alleles; high acid alleles).

**Validation**

Validation of G7Flav-SSR was conducted on unselected families of UF germplasm (n= 4). Within UA seedlings grouped as low acid by CPPCT040b, the G7Flav-SSR “LL” genotypes (homozygous for the low acid allele) had a significantly lower SSC than the “HH” genotypes (homozygous for the high acid allele) (Fig. 1). Within the same group the G7Flav-SSR “LL” genotypes had a significantly lower TA than both the “HH” and “HL” genotypes (Fig. 2).

**New tests**

- Additional peach DNA tests developed

  - **Flesh color + blush**
    - MYB10-SSR combined with CCA04b-SSR to predict flesh color (white vs. yellow) and degree of blush

  - **MYB10-SSR allele** (B: high blush; b: low blush) and CCA04b-SSR allele (W-white; Y-yellow) frequencies in UA selected material

  - **Xap-SSR** - currently being validated

- Additional DNA tests are being developed for apple:
  - Lg8a-SSR<sup>2</sup>: acidity
  - Lg1-Fru-SSR: sweetness

- Additional DNA tests are being developed for sweet cherry:
  - G4Mat: maturity date
  - Firmness

- Additional peach DNA tests developed

  - **Flesh type + ethylene evolution**
    - SMF-SSR combined with endoPG<sup>6</sup> to predict flesh type (melting flesh, non-melting flesh, non-softerning flesh) and rate of ethylene evolution

- Cumulative ethylene evolution (day 20–30) in UA melting flesh germplasm

**CONCLUSIONS**

By harnessing the power of collaboration, specifically the integration of pedigree, phenotypic, and genotypic data generated by RosBREED team members for QTL discovery, the development of these DNA tests was possible. Tools such as G7Flav-SSR are now available to make DNA-based predictions a routine part of tree fruit breeding. This advance in DNA-informed breeding represents an example of successful collaboration among institutions across disciplines and sets the stage for the development of more DNA tests. Future work will broaden the adoption of DNA-informed breeding across Rosaceae tree fruit crops through the advancement of these and other DNA tests.

*Acknowledgements: S. Dirlewanger, D. Bendahmane, A. Cardinale, M. Moing, J. Davis, S. Evans, K. Pedersen, J. R. Clark, and M. Davis.

*References:


