

# Strawberry DNA Testing to Improve Fruit Quality and Disease Resistance

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## Summary

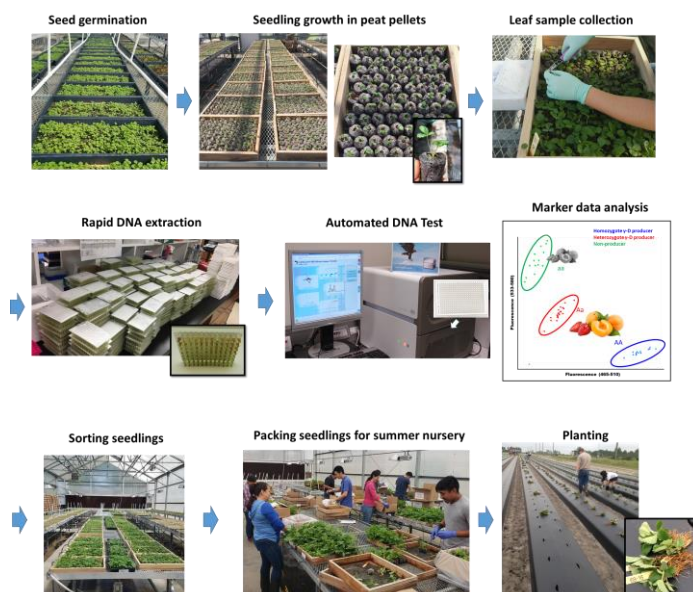
Over the last years, we have identified several important genes controlling strawberry flavor, flowering and disease resistance, and DNA tests are developed to precisely track genes during the breeding process. Using these tests, we screen tens of thousands of seedlings to combine many desirable characteristics into a single variety.

## DNA tests for cultivated strawberry

Currently, we have developed DNA tests for fruity aroma (peach-like flavor), day-neutral flowering and multiple disease resistances (Phytophthora crown/root rot, Colletotrichum crown rot, charcoal rot, anthracnose fruit rot, bacterial angular leaf spot). Using high-throughput DNA tests we ‘stack the deck’ for desirable traits in future strawberry varieties.

## Methods

For the effectiveness of molecular breeding in strawberry, DNA testing tools must be high-throughput, accurate, low-cost and user friendly. The accuracy of our current DNA tests are approximately 95%. To maintain and upgrade marker accuracy, we identified candidate genes for fruit quality and disease resistance using whole genome DNA sequencing approaches. Gene-specific DNA markers were developed and combined with a rapid DNA extraction method to screen large number of breeding samples (Fig. 1). Small leaf punches are collected from seedlings, DNA rapidly extracted, and DNA tests run and scored. Only the seedlings with desirable trait combinations are retained, and the rest are discarded.

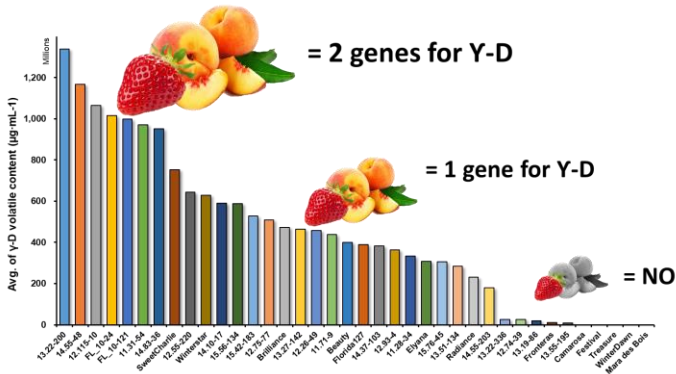


**Figure 1.** The procedure of high-throughput marker-assisted seedling selection (MASS) in UF strawberry breeding program.

## Results

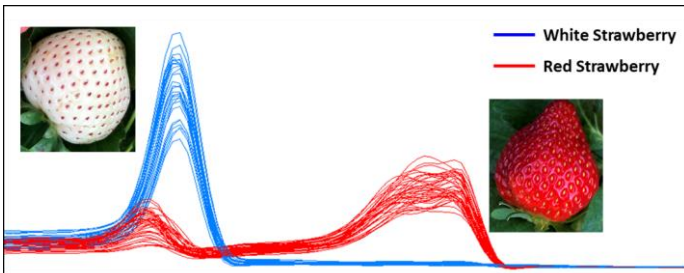
### DNA tests for fruit quality

Gamma-decalactone ( $\gamma$ -D) is an important aroma compound contributing ‘fruity’, ‘sweet’ or ‘peach-like’ flavor in strawberry fruit. A single gene controls the volatile compound,  $\gamma$ -D, and our current DNA marker perfectly predicts the presence and absence of the  $\gamma$ -D gene. However, the production of this aroma varies among Florida varieties and elite breeding germplasm. Remarkably, we discovered a gene dosage effect on the regulation of  $\gamma$ -D contents in cultivated strawberry fruits, and developed a gene-specific DNA marker that predicts both the presence and the amount of  $\gamma$ -D (Fig. 2).



**Figure 2.** Accumulation of  $\gamma$ -D volatile aroma in UF breeding selections and varieties. The level of Y-D content correlates with gene dosage.

Our breeding program has been developing a new strawberry variety with white color when fully ripe. We recently identified a white fruit skin color gene and developed a gene-specific DNA test. As shown in Fig. 3, the newly developed test perfectly predicts red or white fruit skin color from seedlings leaf tissues before any fruits are produced.



**Figure 3.** Development of a DNA test for a strawberry gene that controls fruit skin color. Two different types of DNA sequence patterns explain either white fruit color (blue) or red fruit color (red).

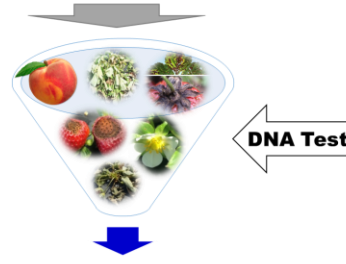
### Stacking multiple disease resistance genes with fruit quality genes using DNA markers

DNA tests are currently available for resistance to the following diseases: Phytophthora crown and root rot, Colletotrichum crown rot, anthracnose fruit rot, bacterial angular leaf spot, and fusarium wilt. Powdery mildew resistance is controlled by multiple genes, and we are currently identifying markers linked to many chromosome regions for PM resistance. The breeding program has identified two genes conferring resistance to charcoal rot. The development of DNA test is currently underway. In 2019, a total of 48,900 seedlings were screened by DNA tests for a combination of characteristics (multiple disease resistances, flavor, and day-neutral

flowering). About 15,000 seedlings were retained for field evaluation in the 2020-2021 season. This effectively increases the size of the breeding program, since the screening process started with more than 50,000 seedlings, before the process of “stacking the deck” began.

## 2020 MASS

54,000 Seedlings



15,000 seedlings

- ✓ Fruity aroma
- ✓ Day neutrality
- ✓ Phytophthora crown rot
- ✓ Colletotrichum crown rot
- ✓ Anthracnose fruit rot
- ✓ Angular leaf spot
- ✓ Fusarium wilt

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