Characterization of Van Well Sweet Cherry Self-Incompatibility (S) Alleles

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Introduction
The multi-allelic self-incompatibility (S) locus controls self-incompatibility and incompatibility between cultivars of sweet cherry (Prunus avium L.) through a gametophytic self-incompatibility system. If the haploid genome of the pollen and the diploid genome of the style have the same S-allele, pollen tube growth is stopped. Thus, commercially grown cherries are planted with compatible cultivars to ensure adequate fruit set. Van Well nursery sent samples of Sunset Bing, Kimberly, and Glory Cherry sweet cherry cultivars, to determine each of their respective S alleles. Using allele-specific primers, we were able to effectively determine the S genotype of each cultivar. Genetic identification of the S genotype of a specific cultivar is of utmost practical importance to maximize yield for growers and also for breeders to use the genotype to test or modify various fruit characteristics.

Background Information
There are 13 identified S alleles, ranging from S1 to S16 (S8, S11, and S15 are duplicates). Allele specific primers for each allele have been determined and published. By using allele specific primers, one can determine the S allele of a cultivar. The variable cultivars are Sunset Bing (SB), Kimberly (K), and Glory Cherry (GC). We have acquired control samples of cultivars Bing (S3, S4), Chelan (S3, S9), and Skeena (S1, S4), thus having controls for S alleles S1, S3, S4, and S9. We also acquired control primers that anneal to all S alleles to ensure the PCR was successful.

Procedure
- Obtain leaves of sample that has unknown S allele
- Use freezer mill to grind leaves
- Obtain DNA from leaves
- Measure amount of DNA through lambda quantification
- Perform Polymerase Chain Reaction (PCR) with allele-specific primers
- Run Gel Electrophoresis of PCR samples. Positive products indicate that the sample has the allele the primers anneal to. Check samples again next to positive controls.

Conclusions
Through our PCR analysis, we were able to determine that Sunset Bing, Kimberly, and Glory Cherry all have the S3 and S4 alleles. With this knowledge, we can determine which cultivars they are able to be crossed with, which is necessary not only for growers to maximize the yield of the fruit set, but also for the breeders in modifying fruit characteristics.

Results

Lambda Quantification: Increasing amount of lambda DNA and examining the intensity of each amount allows comparison to each sample's intensity to approximate amount of DNA in each sample. Upper gel is a lambda quantification of the variable cultivars. Lower gel is a lambda quantification of the control cultivars.

Gel Electrophoresis of control cultivars, showing the product for alleles S1, S3, S4, and S9.

Gel Electrophoresis of PCR samples testing for S alleles S2, S7, S9, S10, S12, and S16. The gel suggests no PCR product.

Gel Electrophoresis of control cultivar S alleles S3 and S4 compared to results of the sample cultivars with the allele specific primers for S3 and S4. The gel indicates through the presence of PCR products, that Sunset Bing, Kimberly, and Glory Cherry have the S3 and S4 alleles. The last three lanes before the ladder are primers that anneal to every S allele to ensure the PCR was successful.

Future Research
More leaves will be obtained to provide positive controls for the other S alleles. In the future, with these positive controls along with the primers for all the S alleles, we will be able to efficiently determine the S allele of any future sweet cherry leaf sample.

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References