

Towards Routine Rapid Identification of Almond Self-incompatibility and Self-compatibility Groups Using Advanced DNA Fingerprinting Technology



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Abstract

California almond varieties are self-sterile, requiring growers to interplant of cross-compatible pollenizer varieties among the main commercial variety. A single, the S-gene, controls cross-compatibility in most commercial varieties and the self-compatibility being developed in new varieties. The cross-compatibility, or incompatibility, of two almond varieties depends on the forms (alleles) of the S-gene present in each variety. Precise knowledge of the S-allele identities of current and future almond varieties is critical to ensure that orchard plantings are cross-compatible and so fully productive. DNA-based methods have been developed to determine the S-allele identities of almond varieties. However, currently available procedures are tedious and prone to error.



New Size Standard Doubles the Range of Detectable S-alleles

The Plant Identification Lab at Foundation Plant Services is conducting research to improve the accuracy and throughput of S-allele identification in almond using cutting-edge DNA fingerprinting technology. This technology will be a valuable resource to researchers and breeders of new almond cultivars. The technology will also be made available to the California almond industry at large as a service offered by the Plant Identification Lab. Though our research is in progress, we can currently identify many S-alleles. Let us know if you have an S-allele related question. We may be able to help. We also offer almond variety identification and pedigree analysis.

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Old School

Existing DNA technology has allowed initial characterization of California almond cross-incompatibility groups. The method defines specific S-alleles based on the length of the DNA, measured in base pairs (bp). Allele sizes for many common S-alleles as generated by one protocol are shown (Table 1). Allele lengths are compared to one another and to size standards using gel electrophoresis; smaller alleles move faster than larger ones through the agarose gel. This method is slow and inefficient, and allele size can only be measured to within roughly 20 bp.

The New Millennium

The Plant Identification Lab is now using a Genetic Analyzer to identify S-alleles in almond varieties. In a process called capillary electrophoresis, the S-allele fragments are passed through a very small-diameter capillary filled with a polymer. As with the gel, smaller Salleles move through the capillary faster than larger ones. A laser detects the S-alleles as they move past a window in the capillary. Software determines and records the size of the S-allele. Automation makes the system fast, less prone to error, and accurate to within a single base pair. By integrating a new size standard and associated software into our protocol, we have more than doubled the size range of S-alleles that can be detected.





S-alleles for Several Almond Varieties Using Agarose Gel Electrophoresis

Table 1. Almond varieties and corresponding self incompatiblity alleles with lengths in base-pairs.

Genetic Analyzer ABI Prism 3130xI, Applied Biosystems

| S-allele | | Published size ^a | | |
|----------|---|--|--|--|
| 6 | 1 | 850 | 1080 | |
| 11 | 12 | 700 | 1600 | |
| 18 | 1 | 650 | 1080 | |
| 5 | 1 | 600 | 1080 | |
| f | 1 | 470 | 1080 | |
| 16 | 8 | 1050 | nd ^b | |
| 7 | 8 | 2000 | nd | |
| 6 | 14 | 850 | 1370 | |
| 2 | 9 | 800 | 1430 | |
| 14 | 7 | 1370 | 2300 | |
| 7 | 8 | 2000 | nd | |
| | S-al 6 11 18 5 f 16 7 6 2 14 7 | S-allele 6 1 11 12 18 1 5 1 f 1 16 8 7 8 6 14 2 9 14 7 7 8 | S-allelePublish61850111270018165051600f14701681050782000614850298001471370782000 | |

^a Results obtained by various researchers using the protocol of Tamura et al. 2000. Theor Appl Genet 101:344-349 ^b Not detected with Tamura et al. 2000 protocol. ^c Mislabeled on the gel.

Variety Identification and Pedigree are Important **Clues to Identifying Self-incompatibility Groups** The incompatibility groups for the almond varieties that are currently important in California are well established. For these varieties, correct identification of a tree's variety will clarify its incompatibility group.

Since self-incompatibility in almonds is an inherited trait, knowing the pedigree of a new almond variety can help determine its incompatibility group. For example all pollen from Mission is compatible with Nonpareil. However, all trees resulting from a cross between these varieties will have either S7 or S8 from the Nonpareil parent. As a result only half the pollen from any Nonpareil **x** Mission selection would successfully pollinate Nonpareil.

Four possible S-allele combinations from Nonpareil (S7 S8) x Mission (S1 S5)

Only half the pollen from each possible progeny will pollinate Nonpareil

| Progeny 1 | | Progeny 2 | | Progeny 3 | | Progeny 4 | |
|-----------|----|-----------|----|------------|----|------------|----|
| S1 | S7 | S5 | S7 | S 1 | S8 | S 5 | S8 |

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