DEVELOPING SELF-COMPATIBILITY IN 'NONPAREIL' ALMOND

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This project is aimed at developing technologies that will be used to reduce/eliminate self-incompatibility, a trait that would improve the efficiency of almond production. Unlike peaches, almonds are self-incompatible and require pollen from a heterologous source as well as an insect vector (bees) for its delivery. The development of self-compatible cultivars that are self-fruitful will mean that no pollinator trees or bees would be required to set fruit. This will be accomplished by identifying the genes involved in self-incompatibility in almond and altering their expression using plant transformation technologies that will be used to inactivate the self-incompatible mechanism. The development of plant transformation technologies will also provide the opportunity to introduce other useful genes into almond. Plant biotechnology offers powerful new approaches that will enhance the ongoing breeding program as it would permit very precise genetic improvement of a commercially important variety such as 'Nonpareil' with potentially minimal impact on other overall productivity and quality characteristics.

Identifying and distinguishing the genes in almond responsible for self-incompatibility: Self-incompatibility (SI) is a widespread mechanism in flowering plants which prevents self-fertilization and promotes out-crossing. In almond this trait is controlled by a single locus with multiple codominant alleles (similar but not identical family of genes that perform the same function) referred to as S-alleles. When an almond pistil is pollinated by its own pollen, SI is triggered which results in abortion of that pollen growth within the stylar tissues. Thus, unless the plant is cross pollinated there is greatly reduced fruit set. The S-alleles encode SRNase proteins (an enzyme that destroys RNA) that are expressed in stylar tissue and that are responsible for the inactivation of "self" pollen growth. Our major objective has been to develop DNA tools to identify the cross-incompatibility group (CIG) that would clearly distinguish among the different S-allele family members, with each almond cultivar having a specific set of S-alleles. Almond has been shown to have 4 predominant S-alleles in California designated, Sa, Sb, Sc and Sd that correspond to 6 CIG groups (Kester et al., 1994). We have successfully developed a DNA test that can clearly distinguish almond cultivars belonging to any of these 6 CIG groups. We have authenticated our DNA test by confirming the previous CIG assignments. This test is so precise

that we have expanded the list of almond cultivars that fall into these 6 CIG to include some cultivars and selections that are of current interest. In addition we have discovered 7 new S-alleles that can be categorized into 11 additional CIG groups that include cultivars and selections that did not fall into the original 6 CIG groups.

Transformation of Nonpareil: The focus of the transformation experiments is still on the tissue culture phase involving callus induction and regeneration of shoots from the callus. In addition we have initiated somatic embryos from almond zygotic embryos. Leaf explants from established shoot cultures of Jeffery and Nonpareil are being examined for adventitious bud induction and shoot regeneration. This work is ongoing with various factors being tested, including concentration of plant growth regulators, modification of basal media types, and different gelling agents. Immature zygotic embryos were isolated from Nonpareil nuts collected in June (2000) when the endosperm was in the jelly phase and the embryo was not fully expanded. Somatic embryo cultures were initiated and these cultures are being examined for repetitive embryogenesis, good multiplication rate *in vitro,* and the formation of normal embryos.

Conclusion and future directions: We can conclude from these experiments that we need to keep trying different approaches and strategies to increase regeneration and transformation in almond. A major success has been the isolation of somatic embryos from almond. We plan to continue this effort this year. Another major success has been the development of a DNA test to identify all of the S-alleles of almond varieties and selections in CA. We have identified 7 new S-alleles that have been confirmed by cloning and DNA sequence analysis. Our next task is to further confirm CIG's with the new S-alleles for almond cultivars and selections relevant to almond production in California.

Publications:

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