**Male sterility, a strategy to favor plant breeding and produce stronger crops**

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Breeding between different plants (cross-pollination) is used in agriculture to produce hybrid plants that are more resistant than their parents due to their increased genetic diversity, a phenomenon called ‘hybrid vigor’ that was first described by Charles Darwin. However, many crops such as rice, tomato, and wheat bear flowers that have both male (stamen) and female (pistil) organs within the same individual plant and thus are able to fertilize themselves. Indeed, these species favour self-pollination as their main reproductive strategy since it is fast and efficient. This is a major problem for breeders, who have to find strategies to prevent these plants from self-fertilizing. One of the easiest strategies to circumvent this problem adopted by plant breeders is to produce ‘male sterile’ plants, where the male part does not function. These plants cannot self-pollinate anymore and will produce seeds only by cross-pollination.

Evidently, the more we know about the mechanisms of self-pollination, the easiest it would be for scientists and breeders to exploit these mechanisms to generate male sterile plants.

The male reproductive organ, stamens, are composed of the anther, where pollen grains are produced, and the filament that supports the anther. Self-pollination requires that mature pollen grains are released from the anther (anther opening) and come into contact with the female reproductive organ. This is achieved by a coordinated growth of the two organs, which is mainly controlled by plant hormones.

Stamen growth and pollen release in *Arabidopsis thaliana* that is an easy to grow, self-pollinating species, are coordinated by the plant hormone auxin. This hormone acts through proteins called Auxin Response Factors (ARFs), that can bind the DNA and regulate specific genes involved in stamen development. Among the different ARFs, ARF8 has a major role in stamen growth as mutants flowers lacking ARF8 (*arf8*) are almost completely male sterile.

The authors of a recent paper published in Plant Cell have found that a complex molecular mechanism, called alternative splicing (AS), plays an unforeseen role in stamen growth. AS is conserved in plants and animals and contributes to the production of different protein in the cells.

Since eukaryotic genes contain a coded message (exons) interrupted by meaningless words (introns), a ‘cut and reseal’ mechanism, called splicing, joins together exons to obtain a mature messenger RNA that is then translated into a protein. AS produces multiple messenger RNAs from a single gene, by differentially joining exons and introns. As a result, some small introns could be retained in the mature messenger. The splice variant can be non-functional or, more interestingly, can give rise to another protein with different characteristics from the ‘regular’ protein. This is for example the case of many genes involved in flower development.

While studying the function of two splice variants of the gene *ARF8*, *ARF8.1* and *ARF8.2*, the authors came across a third splice variant, that they called *ARF8.4*, similar to *ARF8.2* but with an additional intron. After having proved that *ARF8.4* is translated into a protein, they assessed the role in stamen growth of *ARF8.1*, *ARF8.2* and *ARF8.4* by analyzing stamen development in *arf8* mutant flowers and in mutant flowers expressing each of the *ARF8* splice variants or combination of them.

The results showed that the expression of *ARF8.4* alone is able to restore proper stamen growth and fertility in the *arf8* mutant. This is due to the ability of ARF8.4 to directly regulate a gene, called *AUX/IAA19,* that is required for stamen growth. What’s more, ARF8.4 acts together with ARF8.2 to control anther opening: ARF8.4 regulates -through a gene called *MYB26*- lignin deposition in specific anther cells, an early event that is necessary for anther opening, while ARF8.2 regulates the production of jasmonic acid, the hormone required for the final anther breaking.

In conclusion, auxin controls male fertility in Arabidopsis by acting through different *ARF8* splice variants, with the intron-retaining splice variant *ARF8.4* playing crucial roles in stamen development. Since auxin controls stamen growth also in commercially important plants such as rice and tomato, the production of different (ARF) splice variants may represents a general mechanism of controlling male fertility. This could help plant breeders to produce male sterile plants and obtain more resistant and vigorous hybrids.

**Original article:**

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Rinaldi G, Palumbo A, Costantino P and Cardarelli M (2018)

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