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GENETIC CONTROL OF POLLEN FUNCTION AND POLLEN-STYLE INTERACTION IN MAIZE

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In higher plant life cycle, sexual reproduction is a complex developmental phase that is submitted to a very fine regulation. In particular, pollen tube growth is an important component of pollen fitness and represents an interesting model for the study of gene expression during tube growth and of pollen-pistil interaction: from the release of pollen from anthers to the penetration of the micropyle by the pollen tube tip, a strict interaction between pollen and the pistil is established, largely based on cell surface interactions between the male gametophyte and the female sporophytic tissues.

In order to understand the molecular basis of pollen pistil interaction, by the identification of the involved genes and the discovery of their function, we are studying a maize mutant responsible of the phenomenon called “cross sterility”: *Gametophytic factor 1* (*Ga1*). The ears carrying the *strong* allele (*Ga1-s*) can be fertilized only by pollen grains carrying the *Ga1-s* allele, while the *ga1* ears can be fertilized by both pollen types, *Ga1-s* and *ga1*.

To this purpose, transcripts differentially expressed in two near-isogenic maize lines characterized by the presence of the dominant (*Ga1-s*) or recessive (*ga1*) allele of the gene, have been detected by AFLP-TP technique. The tissues analyzed were non pollinated silks from the two isogenic lines, and Ga_S/Ga_S silks pollinated by ga_S pollen or by Ga_S pollen, ga_S/ga_S silks pollinated by Ga_S pollen. Our analysis allowed the identification of 170 differentially expressed transcripts showing various degrees of modulation between the genotypes and pollinations considered. These transcripts have been cloned, sequenced and classified in different functional categories. Particular attention is given to those genes which appear to be induced or repressed by incompatible pollination; their expression profile is further checked by real time PCR.

Now we are assigning all these differentially expressed genes to the ten maize chromosomes using a collection of Oat-Maize Addition lines: oat lines each containing one maize chromosome. Since the *ga1* gene has already been mapped on chromosome 4, the differentially expressed genes localized on chromosome 4 will be mapped further on using radiation hybrids: oat lines possessing different fragments of a specific maize chromosome.

Moreover, we are analysing the *in vivo* pollen tube growth by fluorescence microscopy of silks *Ga1-s* and *ga*, pollinated by the two pollen genotypes and collected 4 and 24 hours after pollination. The samples have been stored in a fixing solution; after washing in distilled water, silk tissues have been treated with NaOH in order to make them soft, and then stained with aniline blue. We have observed our samples by fluorescence microscopy, using both GFP and DAPI microscopy filters.