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COMBINING GENETIC, MOLECULAR AND BIOINFORMATIC TOOLS FOR THE ISOLATION OF MAIZE *GAMETOPHYTIC FACTOR 1 (Ga1)* GENE

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Ga1 (*Gametophytic Factor 1*) is a gametophytic mutant widely present in many popcorn inbred lines. The presence, of the dominant allele: *Ga1^s*, in the sporophyte prevents fertilization by pollen carrying the recessive allele *gal*. The gene has been roughly mapped on maize chromosome 4, exploiting its loose linkage with another mutant, *sugary 1* (*Su1*) (House and Nelson, 1958), but its position was never investigated in greater detail.

The goal of our research is to create a high-resolution map of the chromosomal region of interest, aiming at the isolation and characterization of the gene. For that purpose large segregating populations have been generated and their phenotypic and molecular characterization allowed to determine a more precise location of *Ga1* on *bin* 4.02 and to pin point the gene in a 6 cM region, flanked by molecular marker *umc1509* and a second marker derived from the genomic sequence AZM4_21836. Currently, we are increasing the size of the segregating populations to identify more recombinant individuals and, at the same time, we are adopting various approaches to generate or identify more markers in the region of interest. The exploitation of the markers most tightly linked to *Ga1*, has led to the identification of two BAC contigs: 149 and 150, of the public maize physical map. Those contigs seem to be partially overlapping and thus spanning the entire region containing *Ga1*. EST and TC sequences annotated on the contigs are going to be tested on Oat-Maize Addition lines (OMA) to quickly verify their actual localization on chromosome 4, and eventually on chromosome 4 Radiation Hybrid lines (RH), to establish their position on the genetic map more precisely. All sequences obtained shall be used to produce new markers, representing potential candidate genes.

A second approach, based on the exploitation of rice-maize synteny has been pursued to identify candidate genes localized in the rice syntenic region. Our preliminary results indicate that at least part of the *Ga1* region is collinear with a portion of rice chromosome 11.

Finally, a Bulk Segregant Analysis (BSA) approach, using AFLP markers, has been used to identify molecular markers tightly linked to *Ga1*. Those markers would eventually allow a chromosome landing strategy, avoiding chromosome walking steps that, in species with complex genomes such as maize, are very difficult and time consuming.

The isolation of genes with gametophytic expression is relevant for the study of pollen in itself and it allows shedding some of light on the genetic and molecular mechanisms involved in plant sexual reproduction. Such results are of extreme interest for species such as maize, whose grain has a huge economical value. Accordingly, those results may have relevant effects on the production of hybrids and on the control of transgene dispersion through pollen, a topic of the highest importance, that has considerable implications concerning acceptance, utilization and diffusion of GMO plants.