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DETERMINING THE CORRELATION BETWEEN METHYLATION AND GENE EXPRESSION AT A FLOWERING TIME QTL IN MAIZE

CASTELLETTI S., SALVI S., TUBEROSA R.

Dept. of Agroenvironmental Science and Technology, University of Bologna, Bologna, I-40127 (Italy)

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In maize, the genetic control of flowering time has been investigated in several quantitative trait locus (QTL) studies. One of the major QTLs for flowering time, the *Vegetative to generative transition 1* (*Vgt1*) locus, corresponds to an upstream (70 kb) non-coding regulatory element of *ZmRap2.7*, a repressor of flowering. Among the polymorphisms that distinguish late and early *Vgt1* alleles, the insertion of a MITE transposon was found to be highly associated with flowering time in a number of independent studies. As cytosine methylation is known to influence gene expression, we aimed to determine if methylation might be involved in the relationship existing between *Vgt1* and *Rap2.7*. The methylation state at *Vgt1* was assayed using an approach that combines digestion with McrBc (an endonuclease that acts upon methylated DNA), and quantitative PCR. The analyses were performed on genomic DNA from leaves of six different maize lines at four stages of development. The results showed a trend of reduction of methylation from the first to the last stage with the exception of a short genomic region flanking the MITE insertion, where both alleles display a stable and very dense methylation throughout leaf development. Bisulfite sequencing of a small portion of *Vgt1* revealed differential methylation for a single cytosine between the two alleles. Finally, *ZmRap2.7* expression was determined at four developmental stages for the six genotypes and found to correlate with *Vgt1* methylation.