Proceedings of the 52nd Italian Society of Agricultural Genetics Annual Congress
Padova, Italy – 14/17 September, 2008
ISBN **978-88-900622-8-5**

Poster Abstract – D.59

A MOLECULAR MODEL FOR EPIGENETIC MECHANISMS IN THE OPAQUE2-MEDIATED REGULATION OF GENE TRANSCRIPTION IN MAIZE ENDOSPERM

LOCATELLI S., ROSSI V.

Unità di Ricerca per la Maiscoltura, CRA, Bergamo, Via Stezzano 24, 24126 Bergamo (Italy)

epigenetic mechanisms, gene transcription, Zea mays

Several evidences in the past ten years have clearly demonstrated that epigenetic mechanisms, such as chromatin structure, histone modification, and cytosine methylation play a pivotal role in regulating gene transcription. These mechanisms are conserved in eukaryotes; however the peculiarities of plant development and of response to environmental cues result in a more flexible epigenetic regulation of the genome activity. This implies that plant-specificities in the epigenetic-mediated regulation exist. In addition, species-specific peculiarities, due to differences in genome organization, have also been reported. The precise mechanisms of the epigenetic-mediated regulation of gene transcription are beginning to be clarified in the Arabidopsis model plant. However, these mechanisms are again poorly understood in crop plants.

In this study we have employed the Opaque2 (O2) mediated control of gene transcription during endosperm development as a model system to investigate the epigenetic regulation in maize. O2 is a transcriptional activator belonging to the b-zip class, which has been extensively characterized in the past twenty years. O2 is specifically expressed throughout endosperm development and activates transcription of several target genes (e. g. 22 kDa α-zein, pyruvate orthophosphate dikinase1: PPDK1, lysine ketoglutarate reductase: LKR, etc.). Usually, O2 acts as a homodimer and binds a conserved O2-box within the promoter of its targets. Previous findings indicate that epigenetic mechanisms are involved in the O2-mediated transcriptional regulation, because cytosine methylation impairs the binding of O2 to O2-box and is related to uni-parental expression of 22 kDa α-zein in specific genetic backgrounds. To clarify the role of epigenetic mechanisms in the O2-mediated transcriptional regulation, we have characterized the epigenetic modifications of various O2 target genes in different developmental stages of endosperms and in sporophytic tissues, such as leaves, where O2 and most of its target are not expressed. First we have analyzed the chromatin structure using DNAse I accessibility assays. Subsequently, the cytosine methylation level and profile was assessed by means of restriction with the methylation sensitive enzyme McrBC and bisulfite sequencing, respectively. Chromatin immunoprecipitation (ChIP) technique with antibodies against specific post-translationally modified histones was employed to analyze the histone modification pattern and to investigate the timing of O2 and RNA Polymerase II binding to their target promoters. The results obtained in this study allow the formulation of a molecular model, which describes the role of epigenetic mechanisms in the regulation of transcription for the O2 target genes. This model implies three different transcriptionally states for the O2 target genes: silenced, poised, and activated state, each with a specific profile of epigenetic marks. The detailed results from this study will be illustrated and discussed.