Genome-wide Insulator-encoded Nucleosome-Positioning: Regulation of Transcriptional Pausing to Control the Cell Cycle

Magali Hennion [+], Suresh Cuppadrah[^], Artem Barski[^], Adrien Gamot [+], Sophie Queille [+], Emmanuel Käs, Keji Zhao[^], Eldon Emberly[#], and Olivier Cuvier[+@]

[+]Cell Cycle Chromosome Dynamics, LBME-CNRS, Toulouse, France ; [#]Physics Department, SFU-Canada; [@]Laboratory of Molecular Immunology (LMI), National Heart, Lung, and Blood Institute (NHLBI)-NIH, Bethesda, USA

ABSTRACT

Recent breakthrough in the chromatin field was to show that DNA sequences encode the specific positioning of nucleosomes along eukaryotic genomes (Segal et al., Nature, 2006). It is now essential to understand how specific factors may bind DNA motifs to regulate such nucleosome-positioning, in order to regulate chromatin accessibility and/or e.g. gene expression. Of interest are factors that recognize motifs that are specifically distributed in respect to - i.e. 'phased' or 'anti-phased'- nucleosome-positions (Kaplan et al., Nat. Genetics, 2009). Among these, we find using ChIP-seq that the insulator protein BEAF binds to 5,600 genomic loci juxtaposed to promoters, where its motifs are always anti-phased with nucleosomes (Emberly et al., PloS Biol., 2008; unpublished results). We call these BEAF binding sites "Nucleosome-Associated Cis-regulatory Elements", as they might serve to regulate nucleosome-positioning, which may account for how insulators are involved in regulating gene expression through chromatin structure. To test this, we measured the positions of nucleosomes in control and BEAF-depleted cells. Our data combined with novel statistical analyses by clustering approaches highlight a key role of insulators in stabilizing nucleosome-positioning at +1 that mark most of the transcriptional pausing genome-wide. Our Deep-seq analyses confirm that the function of insulators in regulating NELF-mediated pausing regulate the expression of hundreds of genes, particularly for oncogenes whose expression is driven by c-myc, in agreement with recent data (Rahl et al., Cell, 2010). Our genome-wide analyses therefore reveals transcriptional pausing as a key mechanism of insulators function.