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Mutations in the CTCF Binding Sites on the MVM Genome Reduces Capsid Protein Expression

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Infection of host cells by the parvovirus Minute Virus of Mice (MVM) induces an ATM-dependent, p21 and CHK1-independent, DNA Damage Response (DDR) leading to a pre-mitotic cell cycle arrest necessary for virus replication. MVM initiates replication within discreet sub-nuclear foci termed APAR bodies, which eventually overrun the entire nuclear compartment. Using high-throughput adaptations of chromosome conformation capture assays, we have mapped the sites on the cellular genome where MVM localizes to initiate replication. These cellular regions preferentially accrue DNA damage in uninfected as well as MVM infected cells, and are also constituent parts of chromosomal substructures called Topologically Associating Domains (TADs). The boundaries of TADs on the cellular genome are susceptible to torsional stress that can induce local DNA breaks, but are architecturally stabilized by cis-interactions of chromosomal DNA bound by the proteins CTCF and Cohesin. CTCF and Cohesin proteins also play essential roles in DDR-signaling. MVM contains two consensus CTCF binding sites, one within the NS1 coding region and the other within the capsid ORF. Strikingly, mutations (that did not alter the amino acid sequence) of both of the CTCF binding sites on the MVM genome, but neither mutation individually, reduced the expression of the MVM capsid proteins, but had no effect on NS1 expression. Surprisingly, the double CTCF mutants generated all MVM mRNAs at near normal levels. These transcripts were found to be full-length, spliced, and efficiently transported to the cytoplasm. Further findings on the role of CTCF and Cohesin in regulating MVM expression will be discussed.