Regulation of HIV-1 DNA integration and transcription by nuclear topography

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Efficiency of HIV-1 infection of primary CD4+ T cells is the ultimate result of a complex network of molecular and cellular events, including interaction of viral proteins with cellular factors, metabolic state of the target cell and topography of the infected cell nucleus. By 3D Immuno-DNA FISH (Fluorescence In Situ Hybridization) with DNA probes corresponding to the human genomic regions that are mostly targeted by HIV-1 integration, we discovered that most of these regions are preferentially located in the outer shells of the nucleus. Here, HIV-1 avoids the Lamin Associated Domains (LADs) while it specifically integrates into the DNA within the nuclear pore compartment. By chromatin immunoprecipitation using HIV-1 infected cells, we found that the RNA Pol II-bound, transcriptionally active HIV-1 provirus specifically associates with various nucleoporins of the inner basket of the Nuclear Pore Complex (NPC), some of which were already identified as important factors for integration.
We have previously shown that stability of viral integrase is essential for efficient viral integration and that this process depends on the specific phosphorylation of integrase Ser57 (Manganaro, L, et al. 2010. Nature Med 16, 329). Viruses carrying a mutation at integrase Ser57, or other mutations impairing integrase function, loose connection with the nuclear pore and are found as 2LTR circle episomes all throughout the nuclear space.
We also discovered that silenced, but transcriptionally competent HIV-1 proviruses reside in close proximity to PML nuclear bodies, and that PML occupancy of the latent HIV-1 promoter coincides with the presence of facultative heterochromatic marks at the viral genome. PML degradation and nuclear body disruption results in strong activation of viral transcription; transcriptional activation of HIV-1 requires displacement of the provirus from the PML compartment by active nuclear actin polymerization.
Taken together, these findings underline the importance of nuclear topography and active gene movement in HIV-1 DNA integration and expression and have implications for understanding latency and developing novel strategies aimed at viral eradication.