EPSTEIN-BARR VIRUS REWIRES HOST GENOME THROUGH CTCF AND PARP1 RELOCATION

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Poly [ADP-ribose] Polymerase 1 (PARP1) has the catalytic activity of adding ADP-ribose polymers to the aminoacidic residues of proteins, determining their activity regulation. It has been shown that, following Epstein-Barr Virus (EBV) infection, the activity of PARP1 is modulated by the viral oncoprotein LMP1, which results in a deregulation of gene expression [1]. Furthermore, given the ability of this protein to bind DNA, some of its targets are DNA-binding proteins such as CTCF [2], SMC1A and SMC3 [3][4] which are part of the Cohesin complex that interacts with CTCF in regulating the three-dimensional structure of the human and viral genome. Moreover, it is well known how EBV reprograms gene expression of the host cell [5], but little is known about the reorganization of the three-dimensional structure and the relocation of transcription factors and co-factors upon viral infection. Considering the importance of PARP1 and CTCF in the regulation of both cell and viral gene expression, we evaluated the binding of both proteins on the host B-cell genome before and three weeks after EBV infection by CUT&RUN assay. From the peak annotation it emerges that PARP1 binds equally to both the TSS region and the intergenic regions before EBV infection, while three weeks after infection it appears to relocate to the promoter region. It is also interesting to note that many CTCF and PARP1 binding sites are shared under both conditions, while a large portion of them is specific. Moreover, both PARP1 and CTCF share the same position on the genome at the level of gene promoters that from a GO pathway enrichment analysis were found to be involved in the signaling of AMPK and HIF-1, both targets of LMP1 [6] and PARP1 [7]. Interestingly, there seem to be a switch between CTCF and PARP1 occupancy on genes involved in the signaling pathway of Herpesvirus infection.

By mass spectrometry analysis of B cells pre- and post-EBV infection, we were able to identify changes in chromatin-binding protein complexes that correlate with chromatin three-dimensional structure alterations highlighted by HiC experiments and histone modifications.

Overall, these data suggest that, after EBV infection of the B cells, CTCF and PARP1 undergo a relocation on the host genome which reflects the deregulation of gene expression as well as a reorganization of its the three-dimensional structure. All these changes are functional to proliferation maintenance and could be possible targets for the treatment of EBV-driven malignances.

[1] Martin KA, Lupey LN, Tempera I., 2016, J Virol., Sep 12;90(19):8520-30
[2] Morgan, S.M., Tanizawa, H., Caruso, L.B. *et al.*, 2022, *Nat Commun* 13, 187 (2022)
[3] Krastev, D.B., Li, S., Sun, Y. *et al., 2022, Nat Cell Biol* 24, 62–73
[4] Kliza KW, Liu Q, Roosenboom LWM, Jansen PWTC, Filippov DV, Vermeulen M., 2021, Mol Cell., Nov 4;81(21):4552-4567.e8
[5] Mrozek-Gorska P, Buschle A, Pich D, et al., 2019, *Proc Natl Acad Sci U S A*., 116(32):16046-16055
[6] Lo AK, Lo KW, Ko CW, Young LS, Dawson CW., 2013, J Pathol., 230 (3): 336-46
[7] Hulse M, Caruso LB, Madzo J, Tan Y, Johnson S, Tempera I., 2018, PLoS Pathog., Nov 5;14(11):e1007394