Histone Chaperone CAF1 is essential for the survival of EBV-infected primary B cells and the establishment of viral latency

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EBV preferentially infects nonproliferating, quiescent human B-lymphocytes, activates them, and establishes a latent infection in them [1]. Upon infection, the virus delivers its epigenetically naïve genomic virion DNA (e.g., unmethylated DNA free of histones), which turns into extrachromosomal, fully chromatinized plasmid copies in the nucleus of the latently infected cells later. We studied these early events in primary human B cells and found that nucleosome acquisition on EBV DNA is completed within 48 h post infection and hence prior to the first round of EBV-induced cellular DNA replication. However, close to nothing is known how viral DNA acquires cellular chromatin and which cellular factors might drive this process.

We approached this apparent gap and focused on the role of replication-dependent and -independent histone chaperones, which might load cellular histones on the incoming viral DNA to initiate its chromatinization. Toward this aim, we developed a very efficient genetic technology to knock-out individual cellular genes in primary resting human B cells. We delivered CRISPR-Cas9 ribonucleoprotein complexes by nucleofection to the cells, which were subsequently infected with EBV or cultured on CD40 ligand feeder cells in the presence of IL-4 to drive *in vitro* B cell survival [2].

With this technology at hand, we knocked out single or multiple histone chaperones and studied the EBV infected primary human B cells with respect to cell proliferation, metabolic activities and signs of apoptosis. We found that EBV infected CHAF1B-depleted B cells are severely compromised, are blocked in the first S phase, and do not survive infection suggesting that CHAF1B is an essential factor during EBV induced cellular activation. We also found that CHAF1B is involved in regulating replication of EBV DNA and preventing detrimental genotoxic stress in the first days of EBV infection. Other histone chaperones such as HIRA, DAXX or ATRX alone or in combination seem to be of minor importance during early infection of B cells with EBV.
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[1] P. Mrozek-Gorska *et al.*, 2019, *Proc. Natl. Acad. Sci. U. S. A.*, vol. 116, no. 32, pp. 16046–16055.
[2] E. Akidil *et al.*, 2021, *PLoS Pathogens*, vol. 17, no. 4.