**Immediate-early protein 2 (IE2) of HCMV recruits Polycomb repressive complex 2 into viral replication compartments for efficient viral DNA synthesis**

**Adriana Svrlanska, Heike Hofmann-Winkler, Thomas Stamminger, Nina Reuter**

**Institute of Clinical and Molecular Virology, Friedrich Alexander Universität Erlangen-Nürnberg, Erlangen, Germany**

Host cell multiprotein complexes like the Polycomb repressive complex 2 (PRC2) have been identified as epigenetic regulators of herpesviral latency. Here, we provide evidence for a novel function of PRC2 during lytic replication of human cytomegalovirus, suggesting a requirement of PRC2 components for efficient viral DNA replication. By yeast two-hybrid screening and co-immunoprecipitation analysis, we discovered EED, a core component of PRC2, as an interaction partner of the viral transactivator protein IE2. In order to elucidate the function of this interaction, we investigated the PRC2 core proteins during lytic replication. This revealed a virus-induced upregulation of the major factors EZH2, SUZ12 and EED both on the mRNA as well as on the protein level. Depletion of PRC2 components by shRNAs or the destabilization of PRC2 by its inhibitor DZNep resulted in impaired viral late gene expression, which is dependent on efficient viral genome amplification. Consistently, this resulted in a reduced release of viral particles. However, inhibition of the methyltransferase activity of PRC2 by GSK126 had no effect on HCMV replication. By immunofluorescence staining, we found that all major PRC2 components relocalize into viral replication compartments (VRCs) as infection progresses. Interestingly, H3K27me3, normally established by PRC2, was specifically excluded from these sites suggesting a differential role of PRC2 during lytic infection independent of its repressor activity. To elucidate the role of IE2 as a regulator of PRC2 activity, we generated a recombinant virus lacking the EED interaction interface within IE2. Indeed, this virus showed an incomplete relocalization of EED into VRCs. Furthermore, in accordance with our hypothesis, we observed an impaired intracellular accumulation of viral DNA as well as a growth defect compared to wildtype virus. In summary, we identified a novel interaction between IE2 and EED, which contributes to the recruitment of PRC2 into VRCs for efficient viral DNA replication.