Characterization of the Interaction Between Staufen-1 With HIV-1 Rev and HERV-K(HML-2) Rec
Katharina Fiddeke, Alexander Rohde, Kirsten Hanke, Jula Wamara, Oliver Hohn, Norbert Bannert
HIV and Other Retroviruses, Robert Koch Institute, Berlin, Germany

Background: The Rev protein of HIV-1 is an RNA transport factor that enhances nuclear export of intron-containing retroviral transcripts and appears to be critical for various aspects of subsequent RNA utilization pathways. The cellular protein Staufen-1 is in several ways functionally related to retroviral RNA transport proteins. It shuttles RNA out of the nucleus and has previously been shown to become incorporated into HIV-1 particles by direct binding to the genomic RNA and to interact with the Gag protein of HIV-1, promoting Gag oligomerization and RNA encapsidation. Moreover, we have recently demonstrated that it interacts with Rev of HIV-1 and the Rec and Gag proteins of HERV-K(HML-2).

Methodology: Using pull-down and coimmunoprecipitation experiments along with deletion mutants we have mapped the interaction sites of Staufen-1 with Rev and Rec. The effect of Staufen-1 on nucleo-cytoplasmic export and translation was determined with reporter constructs in HEK 293T cells. Stress granules were induced with arsenite and the localization of the proteins in granules was investigated by immunofluorescence.

Results: Having demonstrated a direct interaction between Staufen-1 and HIV-1 Rev we found that the NLS of Rev is the region required for binding. In the Staufen-1 protein, the RBD3 domain is the main mediator of the interaction with Rev. This is in contrast to the situation with Rec, where binding is mediated primarily through the RBD4 domain. While Staufen-1 strongly enhances nucleocytoplasmic export and/or translation of unspliced HERV-K(HML-2) transcripts in the presence of Rec, this seems not to be the case for unspliced HIV-1 transcripts in the presence of Rev. Moreover, we demonstrate that under cellular stress conditions Rec and Rev associate in Staufen-1-positive stress granules. However, Staufen-1 appears to attract only Rec, but not Rev, into stress granules.

Conclusions: Although Staufen-1 interacts with both HIV-Rev and HERV-K(HML-2) Rec, only the nucleocytoplasmic transport and translation of HERV-K(HML-2) is significantly upregulated by Staufen-1. Our data also indicate that Rev and Rec are recruited into stress granules by different mechanisms.