Supplementary Contents

Supplemental Figure S1

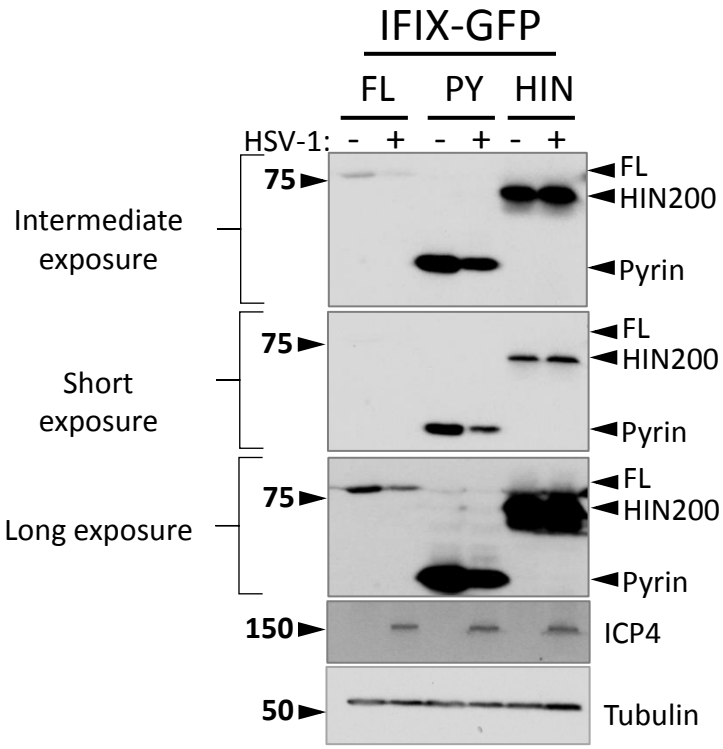

Figure S1. Western blot of 293Ts transfected with IFIX-GFP domain constructs reveals the pyrin domain of IFIX is the target for degradation. FL=full length, PY=pyrin domain, and HIN=HIN200 domain were visualized with an anti-GFP antibody. ICP4 is marker for infection and visualized with an anti-ICP4 antibody. Tubulin is loading control. MOI:10, 6hpi; each anti-GFP blot represents an intact western blotting membrane from each exposure.

Supplemental Figure S2

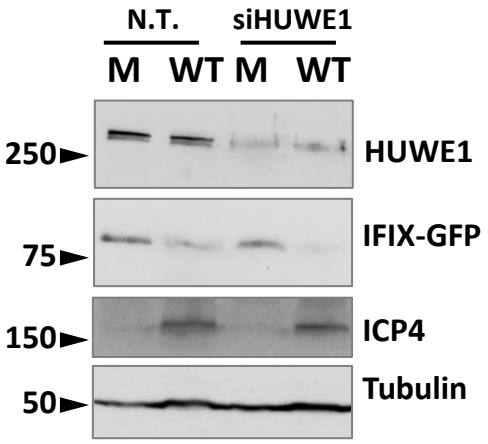

Figure S2. Cellular E3 ubiquitin ligase HUWE1 does not target IFIX for degradation during infection. Inducible IFIX-GFP 293 cells were transfected with the indicated siRNAs and infected with WT HSV-1 24 hours post transfection. MOI:10, 6hpi.

Supplemental Figure S3

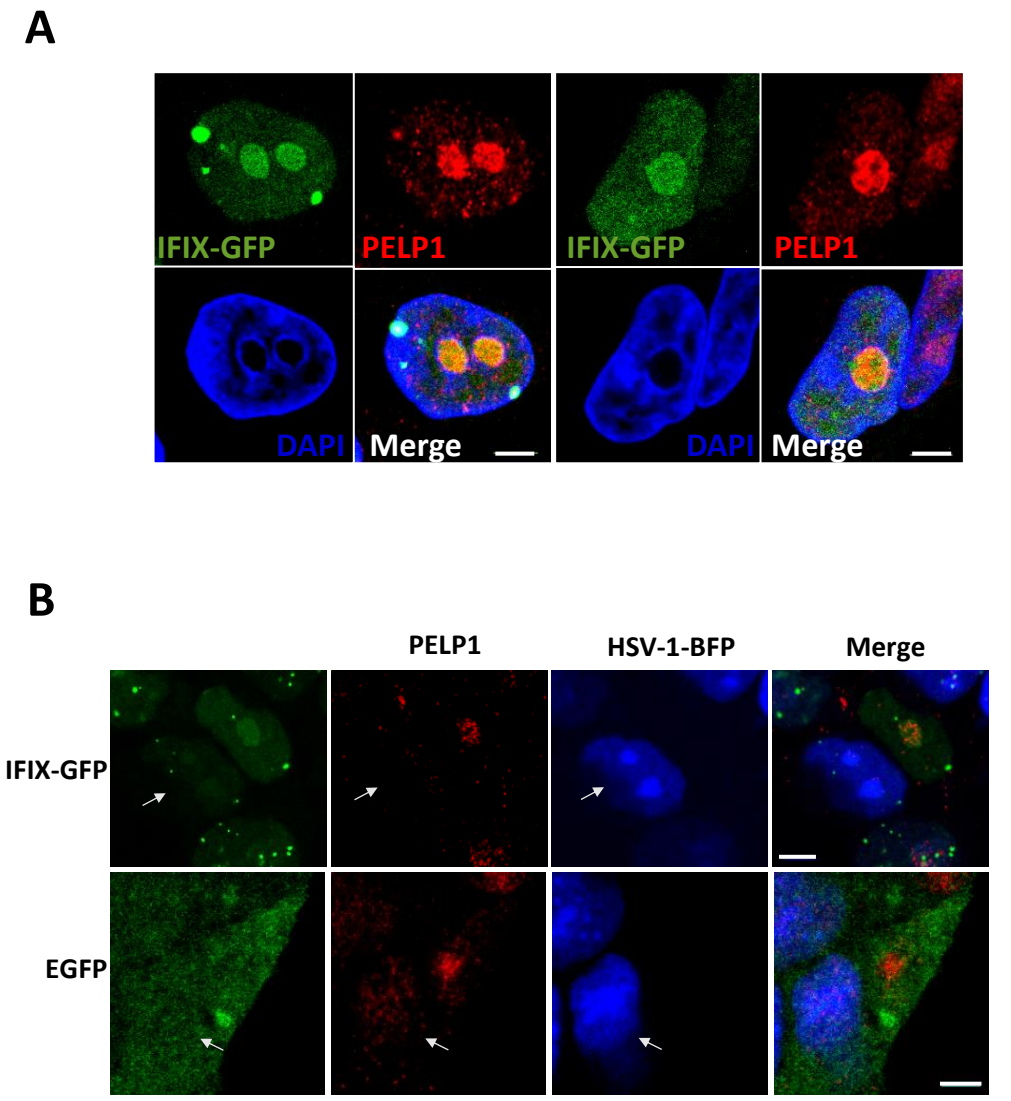

Figure S3. IFIX-GFP and PELP1 localizations in uninfected and HSV-1 infected cells. A) Colocalization of IFIX-GFP and PELP1 within nucleoli of uninfected cells. Microscopy images were taken at 60x oil objective. Bar=5µm. **B)** IF microscopy in IFIX-GFP and EGFP control 293 cells, showing a redistribution of 5FMC protein PELP1 in HSV-1-BFP infected cells (white arrows), MOI:5, 4hpi. Microscopy images were taken at 60x oil objective. Bar=5µm

Supplemental Figure S4

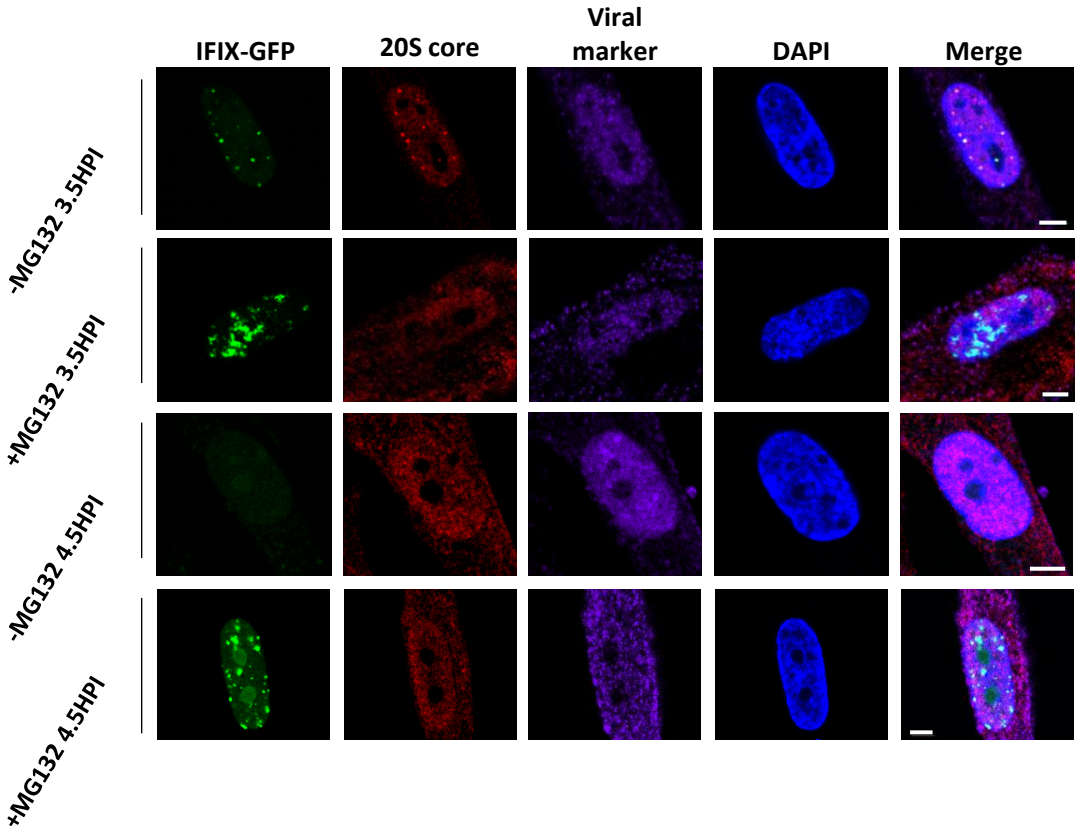

Figure S4. IFIX localization during HSV-1 infection with and without proteasome inhibition. IFIX-GFP stable fibroblasts were treated or not treated with MG132 and imaged at 3.5 and 4.5 hpi. IFIX-GFP, when prevented from degradation, formed aggregates. Microscopy images were taken at 60x oil objective. Bar=5µm

Supplemental Figure S5

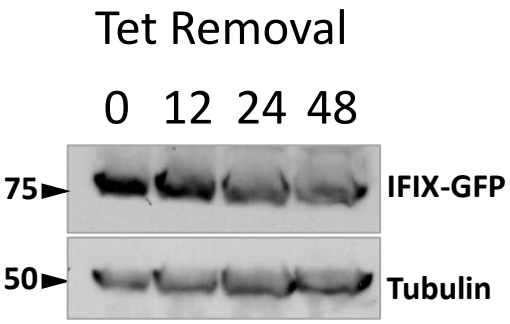

Figure S5. Removal of tetracycline inducer of the IFIX-GFP 293 cell lines for the indicated time points shows IFIX is still present after 48 hours, indicating the turnover rate of IFIX-GFP is longer than the intracellular HSV-1 lifecycle of 24 hours.