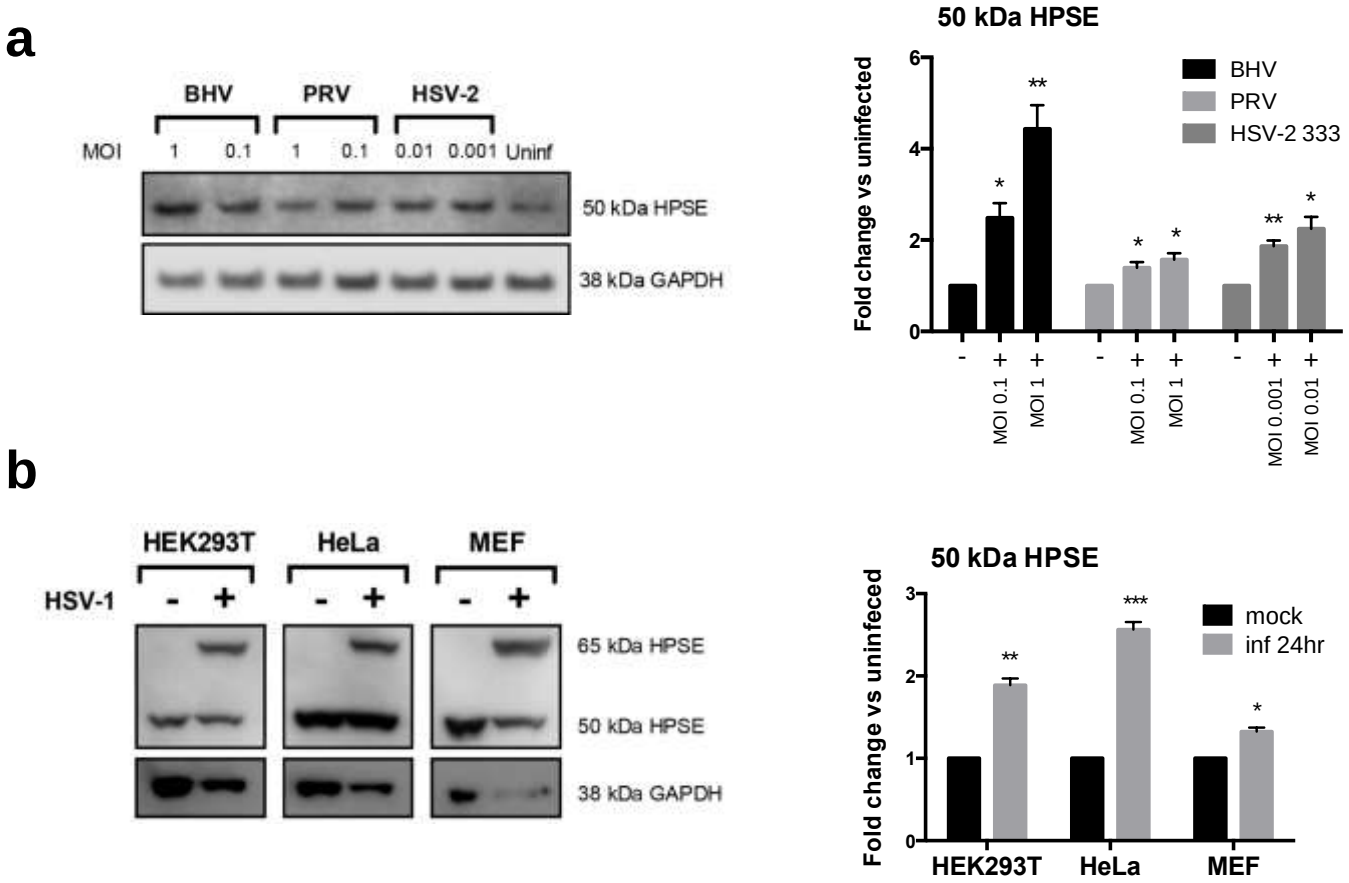
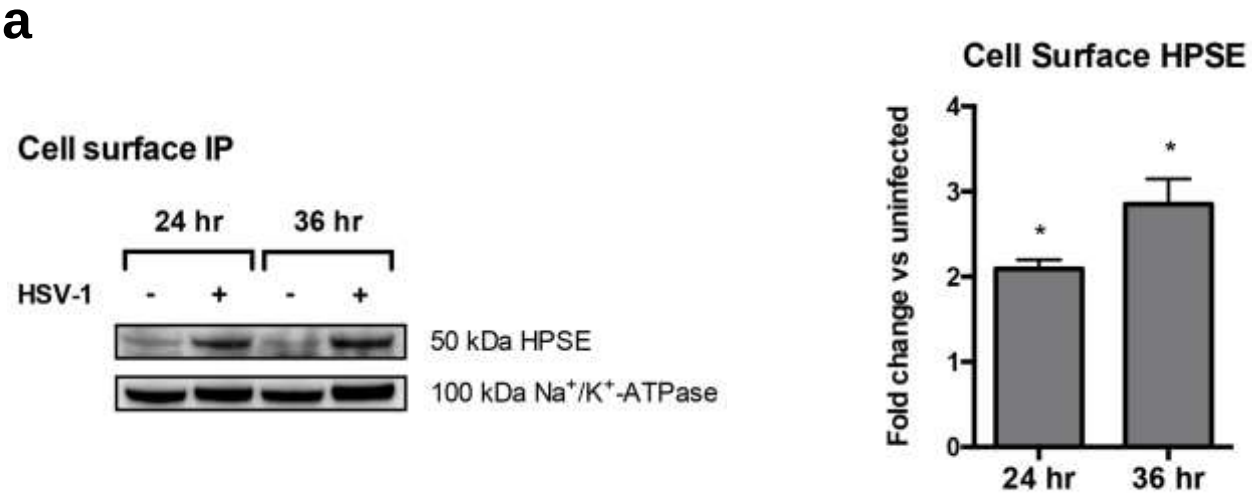


Supplementary Figure 1

HPSE upregulation with other herpesviruses and cell types

a. Increased active HPSE (50 kDa) protein expression upon infection with multiple herpes viruses: bovine herpes virus (BHV), pseudorabies virus (PRV), and herpes simplex virus type-2 (HSV-2). *Left*- HCE cells were infected with the above viruses at the specified MOIs for 24 hrs, and cell lysates were analyzed by western blot. *Right*- Densitometry quantification of 50 kDa active HPSE normalized to GAPDH. Fold change vs uninfected is shown for each virus.

b. Increased HPSE expression upon HSV-1 infection of multiple cell types: HEK293T, HeLa, mouse embryonic fibroblasts (MEF). *Left*- The specified cells were infected with KOS-WT at MOI 0.1 for 24 hr, and cell lysates were analyzed by western blot. *Right*- Densitometry quantification of 50 kDa active HPSE normalized to GAPDH. Fold change of infected over uninfected cells is shown for each cell type. All data are presented as mean ± s.e.m. of three independent experiments (n=3). Asterisks denote a significant difference as determined by Student's *t*-test; **P*<0.05, ***P*<0.01, ****P*<0.001. See Supplementary Figure 9 for full-length images of blots.

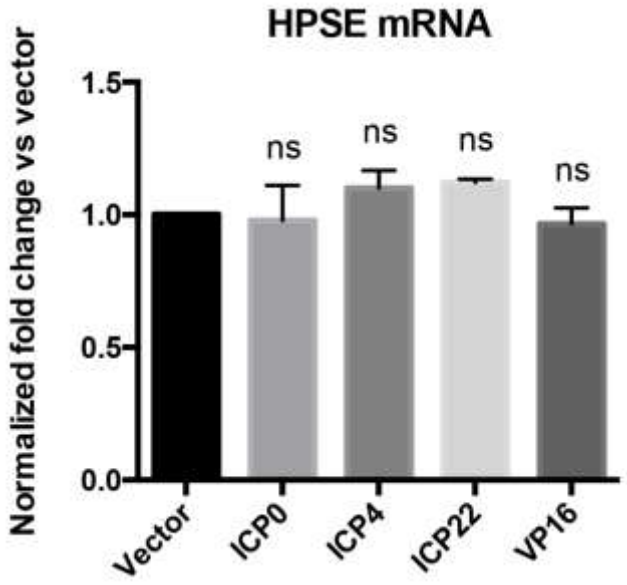


Supplementary Figure 2

Increased expression of HPSE at the cell surface

a. Cell surface proteins were labeled with Sulfo-NHS-SS-Biotin and lysed. Biotinylated proteins from the remaining lysates were isolated using NeutrAvidin agarose column, eluted, and analyzed by western blot. At 24 and 36 hpi, an increase in active HPSE was observed when compared to uninfected samples. Na^+/K^+-ATPase was used as loading control for cell surface extract. Also shown is densitometry quantification of cell surface HPSE based on western blot. Values are normalized to Na^+/K^+-ATPase for each time point. All data are presented as mean ± s.e.m. of three independent experiments (n=3). Asterisks denote a significant difference as determined by Student's *t*-test; **P*<0.05. See Supplementary Figure 9 for full-length images of blots.

a

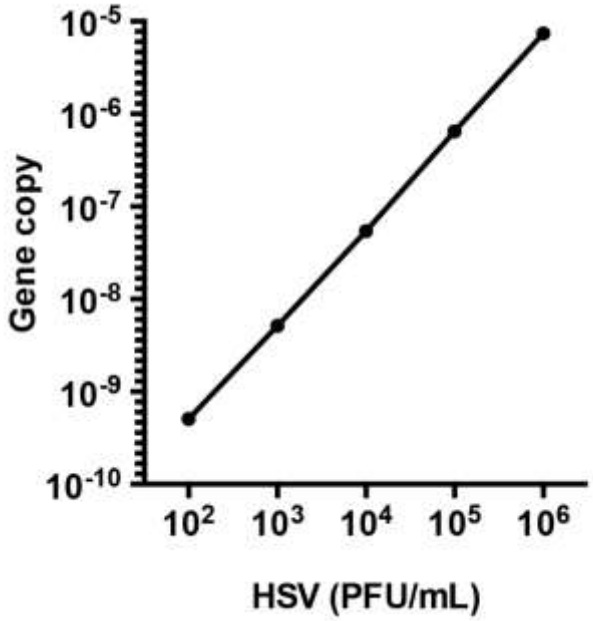


Supplementary Figure 3

HPSE mRNA levels unaffected by herpesviral transcriptional regulators

a. No significant change was observed in HPSE mRNA levels after transfection of HCE cells with expression constructs encoding transcriptional regulators ICP0, ICP4, ICP22, and VP16. Empty vectors of respective plasmids were used as transfection control. All data are presented as mean ± s.e.m. of three independent experiments (n=3). Asterisks denote a significant difference as determined by Student's *t*-test; ns, not significant.

a

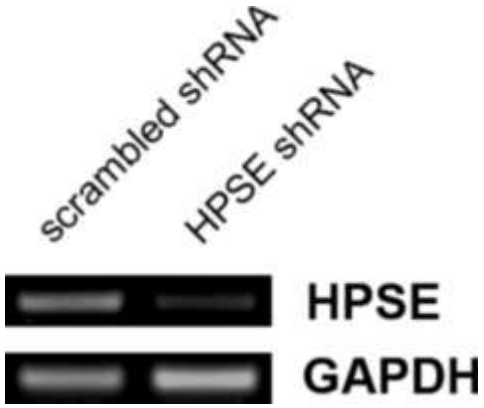


Supplementary Figure 4

Standard curve for viral DNA PCR assay

a. Standard curve of virus gene copy and plaque forming units. Standard curve created using KOS-WT virus of known titer. The coefficient of determination (R^2) of the standard curve is 0.999.

a

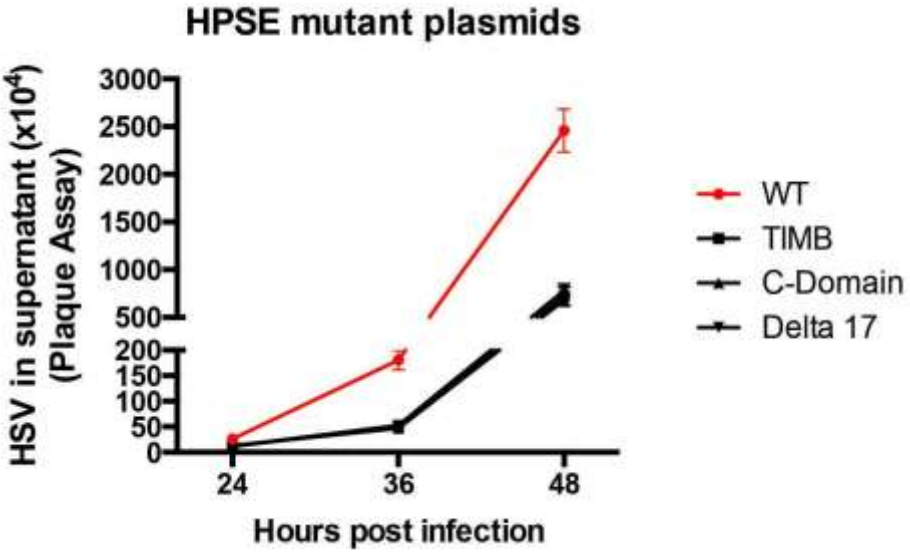


Supplementary Figure 5

HPSE knockdown in HCE cells

a. HPSE knockdown in HCE cells shown by reverse transcriptase PCR using primers against HPSE and GAPDH. Representative gel of three independent experiments is shown (n=3). See Supplementary Figure 9 for full-length images of gels.

a

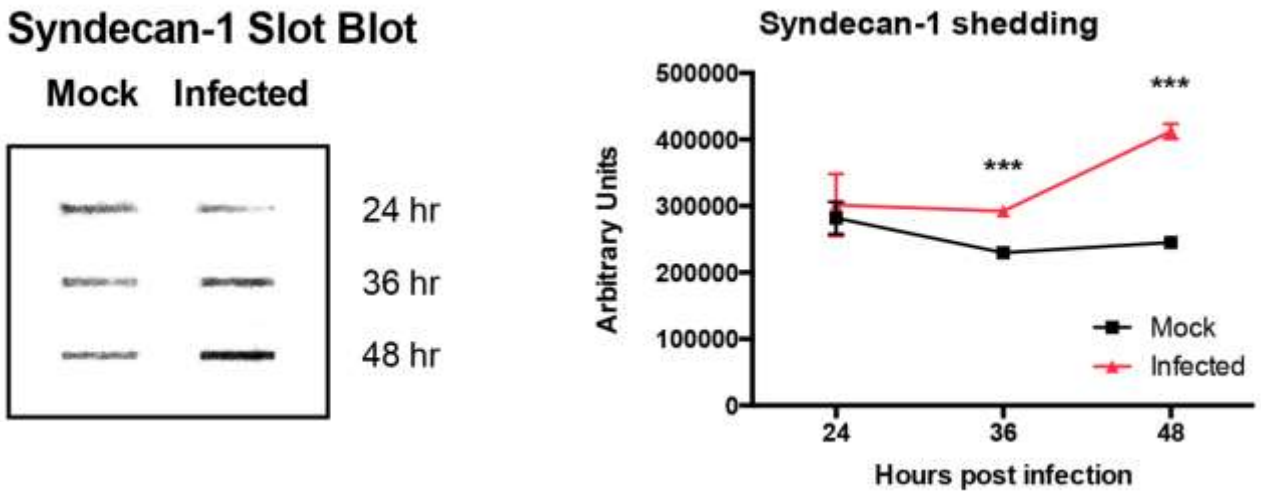


Supplementary Figure 6

Active HPSE is required for increased viral egress

a. Mutant HPSE plasmids lacking TIM barrel, C-Domain, and Delta 17 domains were transfected alongside WT HPSE into HCE cells, and infected with KOS-WT at MOI 0.1. At the specified times post-infection, amount of virus in the culture supernatant was quantified by titering on Vero cells in a plaque assay. All data are presented as mean ± s.e.m. of three independent experiments (n=3).

a



Supplementary Figure 7

Syndecan-1 shedding is increased upon infection.

a. *Left*- HCE cell culture supernatants after infection or mock treatment at 24, 36, and 48 hrs were vacuum filtered through Millipore Immobilon-Ny+ nylon membrane using Bio-Dot SF apparatus (Bio-Rad). Membranes were blotted overnight with antibody specific for Syndecan-1 (Santa Cruz Biotechnology, sc-6532) followed by species-specific secondary antibody and chemiluminescence detection. *Right*- Densitometry quantification of syndecan-1 slot blot. All data are presented as mean ± s.e.m. of three independent experiments (n=3). Asterisks denote a significant difference as determined by Student's *t*-test; ***$P<0.001$.

Fig. 2a

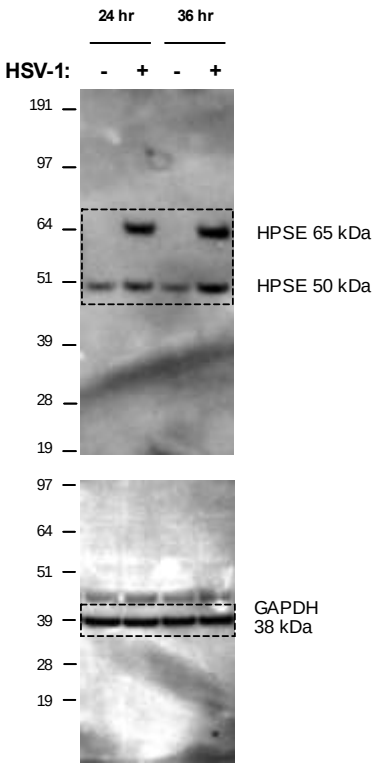


Fig. 3c

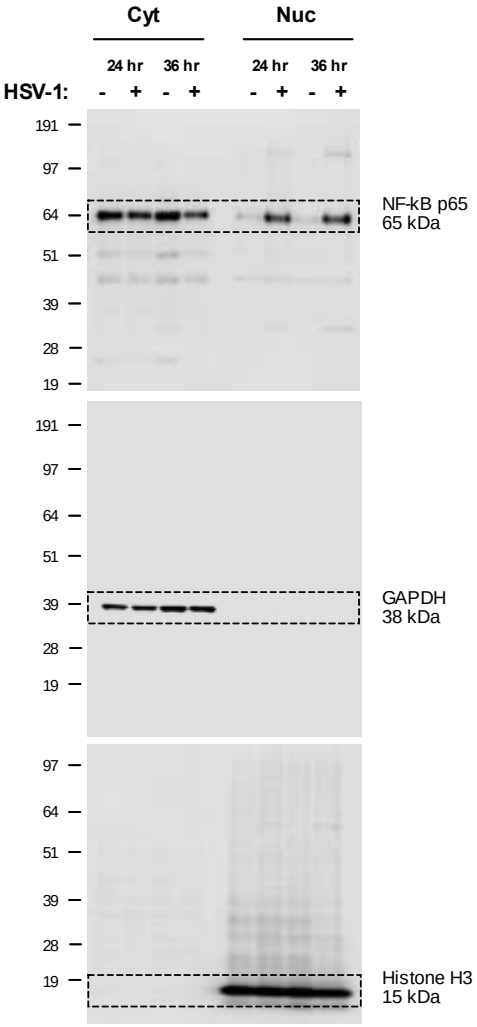


Fig. 3d

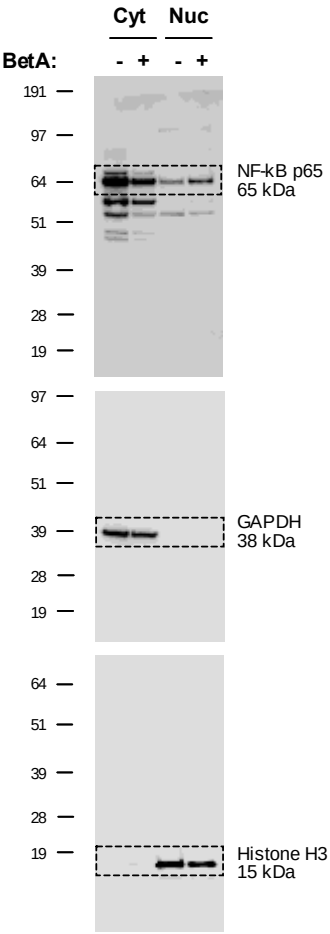


Fig. 5a

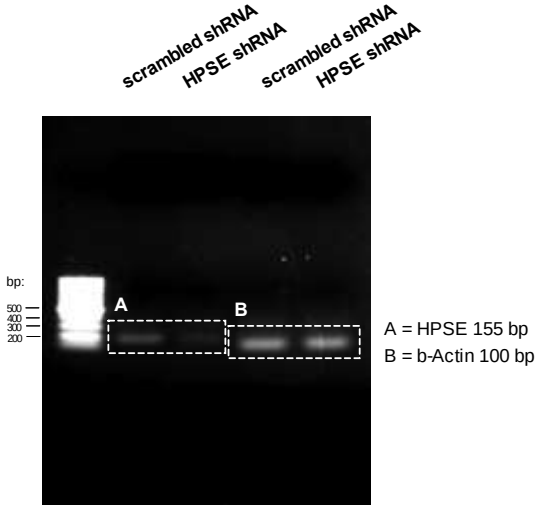
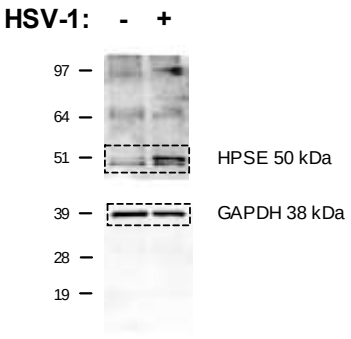


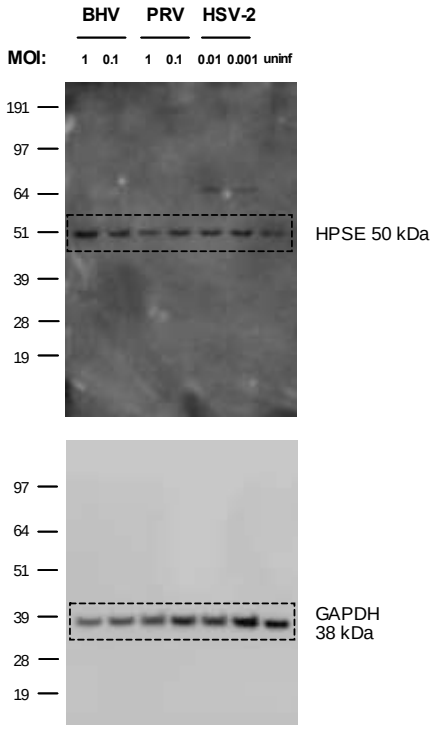
Fig. 5f



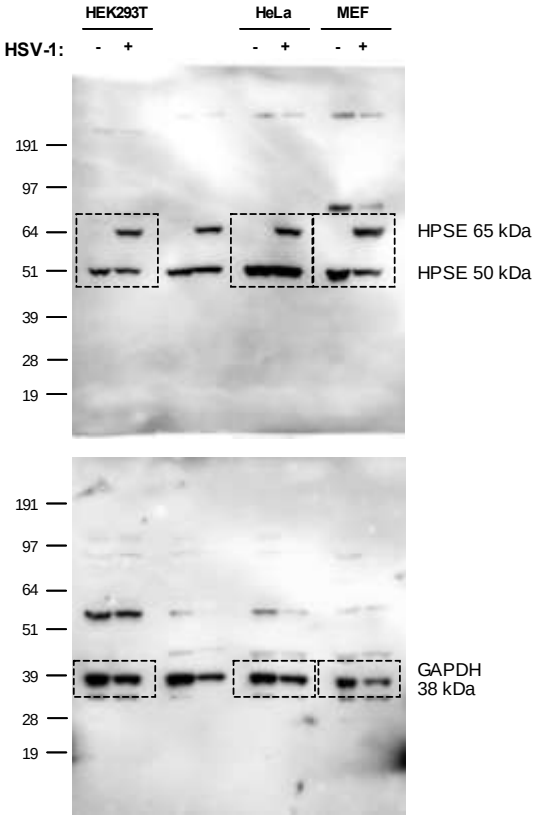
Supplementary Figure 8

Full-length images of blots and gels presented in main figures.

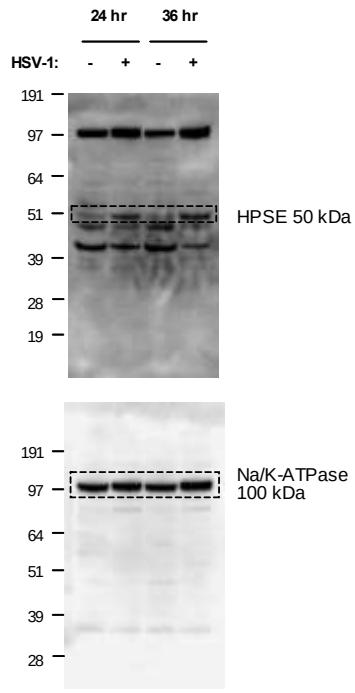
Supplementary Fig. 1a



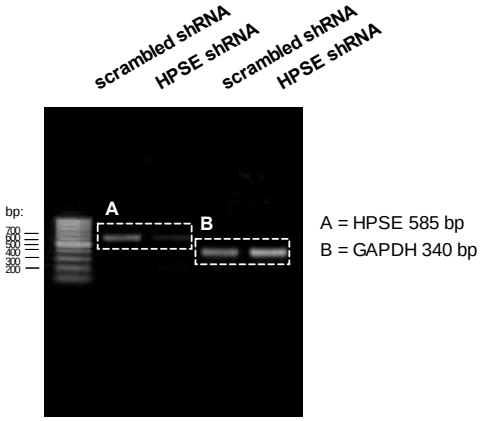
Supplementary Fig. 1b



Supplementary Fig. 2a



Supplementary Fig. 2a



Supplementary Figure 9

Full-length images of blots and gels presented in supplementary figures.