Supporting Information

Liu et al. 10.1073/pnas.1316208110

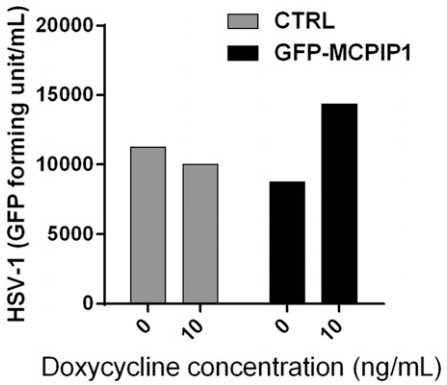

Fig. S1. Monocyte chemotactic protein-induced protein 1 (MCPIP1) induction did not affect HSV-1 infection. Doxycycline-induced CEM-CTRL or GFP-MCPIP1 cells were infected with HSV-1-GFP [multiplicity of infection (MOI) = 0.5] for 2 d. The supernatant of the culture was subjected to virus titration on A549 cells.

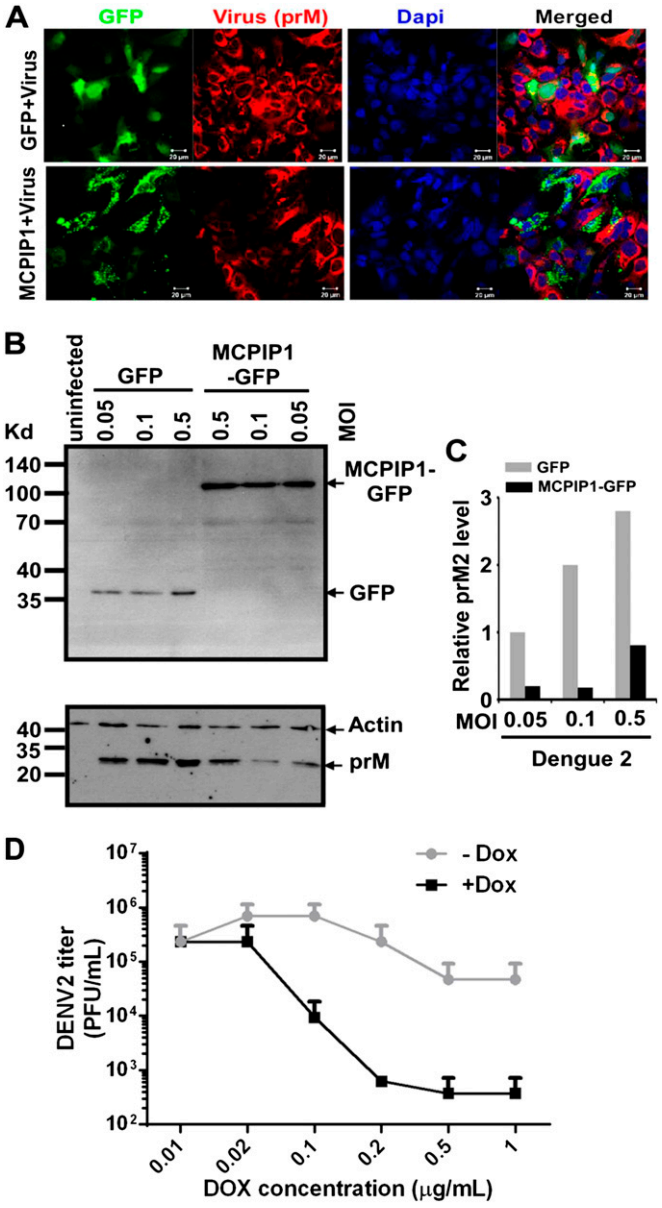

Fig. S2. MCPIP1 expression limited Dengue 2 virus replication. (*A*) GFP- or GFP-MCPIP1–expressing Huh7.5.1 cells were infected with DENV2 at MOI = 1 for 2 h. Cells were fixed and permeabilized after an additional 48 h of incubation and immunostained for DENV prM protein. Nuclei were stained by DAPI. (*B* and *C*) GFP- or GFP-MCPIP1–expressing Huh7.5.1 cells were infected with DENV2 at MOI = 0.05, 0.1, and 0.5 for 2 h. Cells were harvested after an additional 48 h of incubation, and Western blot was performed with DENV prM antibody or GFP antibody. Actin was served as loading control. The intensity of bands for prM was quantified by Gel-Pro Analyzer software and presented as fold changes at the right of the image. (*D*) DENV2-infected HeLa-Dox^{ON}-GFP-MCPIP1 cells were incubated for 2 d. Supernatants were collected and titered for the release of infectious virus. Dox, doxycycline.

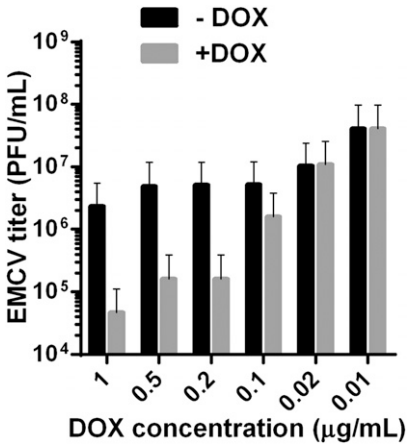

Fig. S3. MCPIP1 restricted encephalomyocarditis virus (EMCV) production. Supernatants collected from EMCV-infected HeLa-Dox^{ON}-GFP-MCPIP1 cells were tiered on BHK-21 cells for the production of EMCV. Dox, doxycycline.

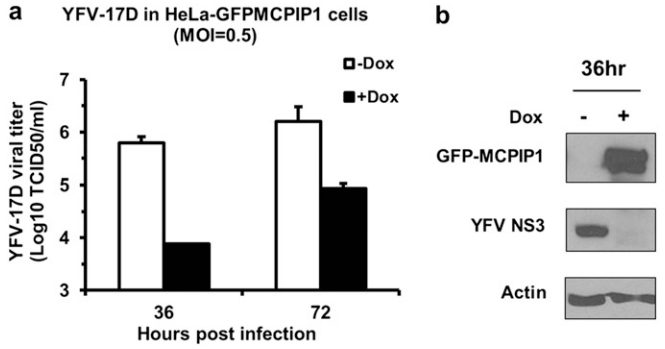

Fig. S4. MCPIP1 restricted Yellow fever virus-vaccine strain 17D (YFV-17D) propagation. HeLa-Dox^{ON}-GFP-MCPIP1 cells were cultured in the presence or absence of 0.2 μg/mL doxycycline (Dox) for 24 h to induce or repress GFP-MCPIP1 expression, respectively, and subsequently infected with YFV-17D virus (MOI = 0.5). (*A*) At indicated time points postinfection, cell-free culture supernatants were harvested, and the infectious progeny virus yields were determined by 50% tissue culture infectious dose ($TCID_{50}$) assay. In *B*, whole-cell lysates harvested at 36 h postinfection were subjected to immunoblot analysis of YFV NS3, GFP-MCPIP1, and actin (loading control) expression. Data shown are representative of two independently performed experiments.

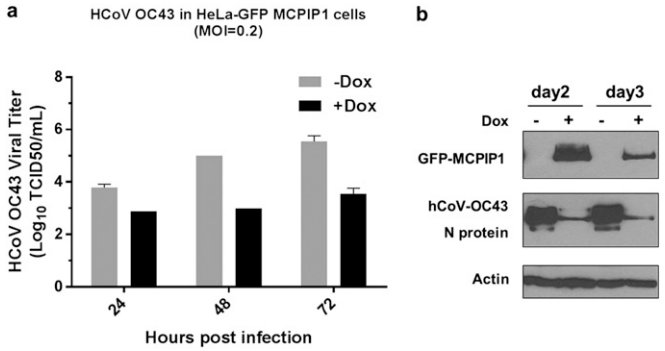

Fig. S5. MCPIP1 restricted human coronavirus OC43 (HCoV-OC43) production. HeLa-Dox^{ON}-GFP-MCPIP1 cells were cultured in the presence or absence of 0.2 μg/mL doxycycline (Dox) for 24 h to induce or repress GFP-MCPIP1 expression, respectively, and subsequently infected with HCoV-OC43 (MOI = 0.2). (*A*) Cell-free culture medium was harvested at the indicated time points postinfection, and the progeny infectious virus titers were determined by $TCID_{50}$ assay. In *B*, whole-cell lysates harvested at 48 and 72 h postinfection were immunoblotted for expression of GFP-MCPIP1 and HCoV-OC43 N protein. The same membrane was reprobed with anti–β-actin to show equal loading. Data shown are representative of two independently conducted experiments.

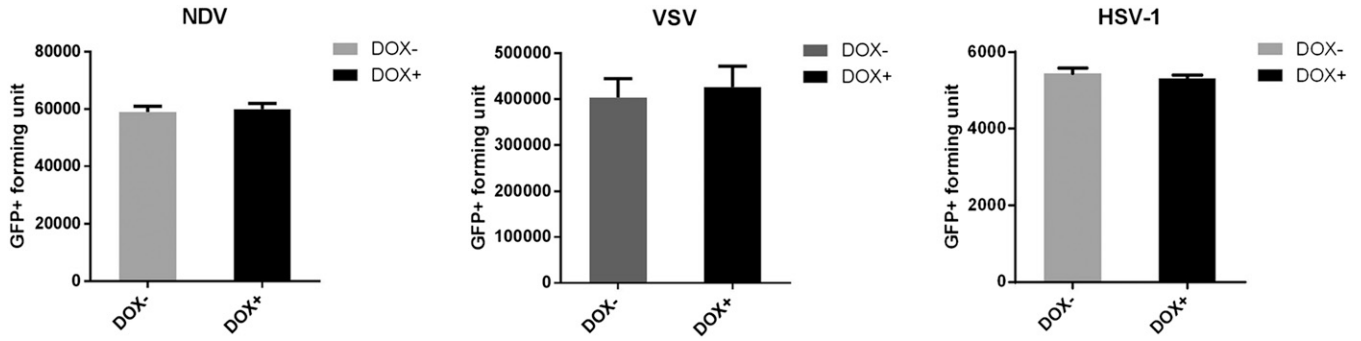

Fig. S6. MCPIP1 did not affect new castle diseases virus (NDV), VSV, and HSV-1 production. HEK293-Dox^{ON}-MCPIP1 cells were infected by GFP expressing NDV, VSV, and HSV-1; 48 h later, the cells were counted for the number of GFP+ cells. Dox, doxycycline.

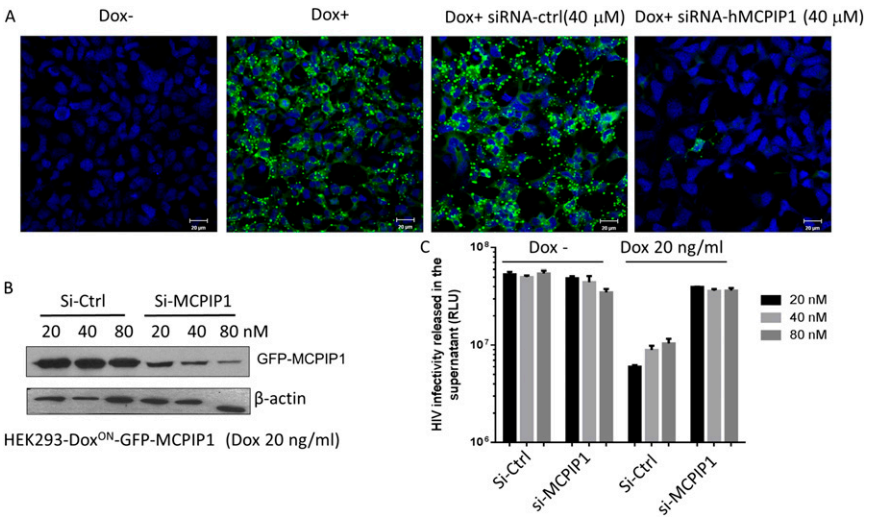

Fig. S7. Knockdown of MCPIP1 in HEK293-Dox^{ON}-MCPIP1 cells increased HIV-1 production. HEK293-Dox^{ON}-GFP-MCPIP1 cells were transfected with the pNL4.3 proviral construct and then treated with Dox (20 ng/mL) to induce the expression of GFP-MCPIP1. Cells were then transfected with siRNA-CTRL or siRNA-MCPIP1 at various concentrations. *A* and *B* show the knockdown of GFP-MCPIP1, whereas in *C*, the production of infectious HIV-1 was measured by TZM-bl assay. Dox, doxycycline; RLU, relative light unit.

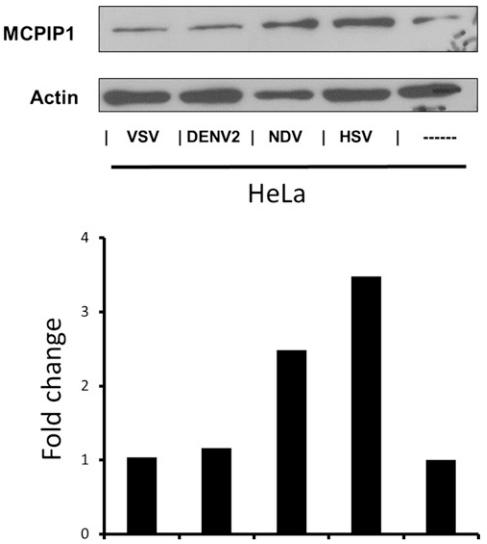

Fig. S8. Induction of MCPIP1 on viral infections. HeLa cells were infected by indicated viruses (MOI ~ 1). Cell lysates were made 24 h thereafter and subjected to Western blotting for the detection of endogenous MCPIP1 and β-actin.

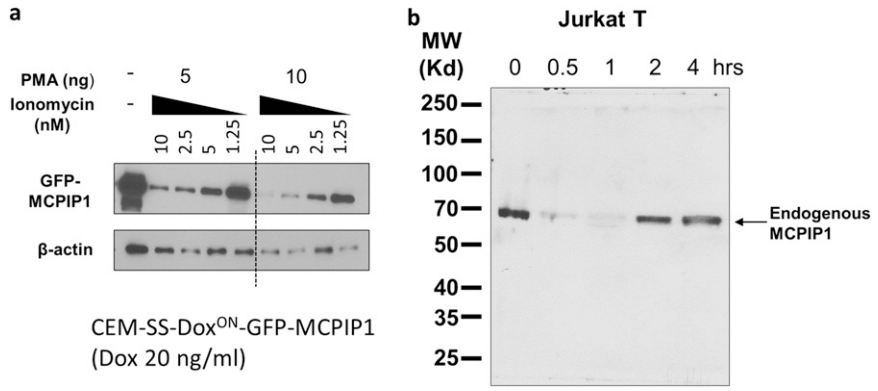

Fig. S9. Phorbol 12-myristate 13-acetate (PMA)/ionomycin treatment reduced MCPIP1 level in human T-cell lines. (*A*) CEM -Dox^{ON}-GFP-MCPIP1 cells were first treated with Dox for 1 d, and then, the cells were stimulated with PMA/ionomycin. (*B*) Jurkat T cells were stimulated by PMA (5 ng/mL) /ionomycin (200 nM), and cell lysates were collected at indicated time points for Western blotting analysis. Dox, doxycycline; MW, molecular mass.

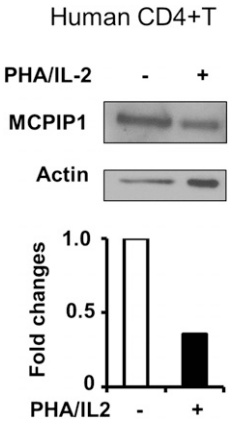

Fig. S10. Phytohemagglutinin (PHA) /IL-2 stimulation decreased MCPIP1 expression. Purified human CD4+ T cells were stimulated with PHA/IL-2 for 3 d. Endogenous MCPIP1 was detected by Western blotting.

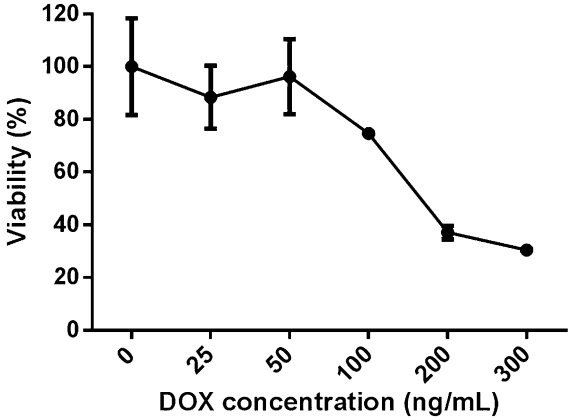

Fig. S11. Cell viability on MCPIP1 induction. Doxycycline (Dox) was added to CEM-Dox^{ON}-GFP- MCPIP1 cells at different concentrations. After 48 h of incubation, cells were lysed and subjected to CellTiter-Glo Luminescent Cell Viability Assay.