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Supplemental information

GS-CA1 and lenacapavir stabilize the HIV-1 core and modulate the core interaction with cellular factors

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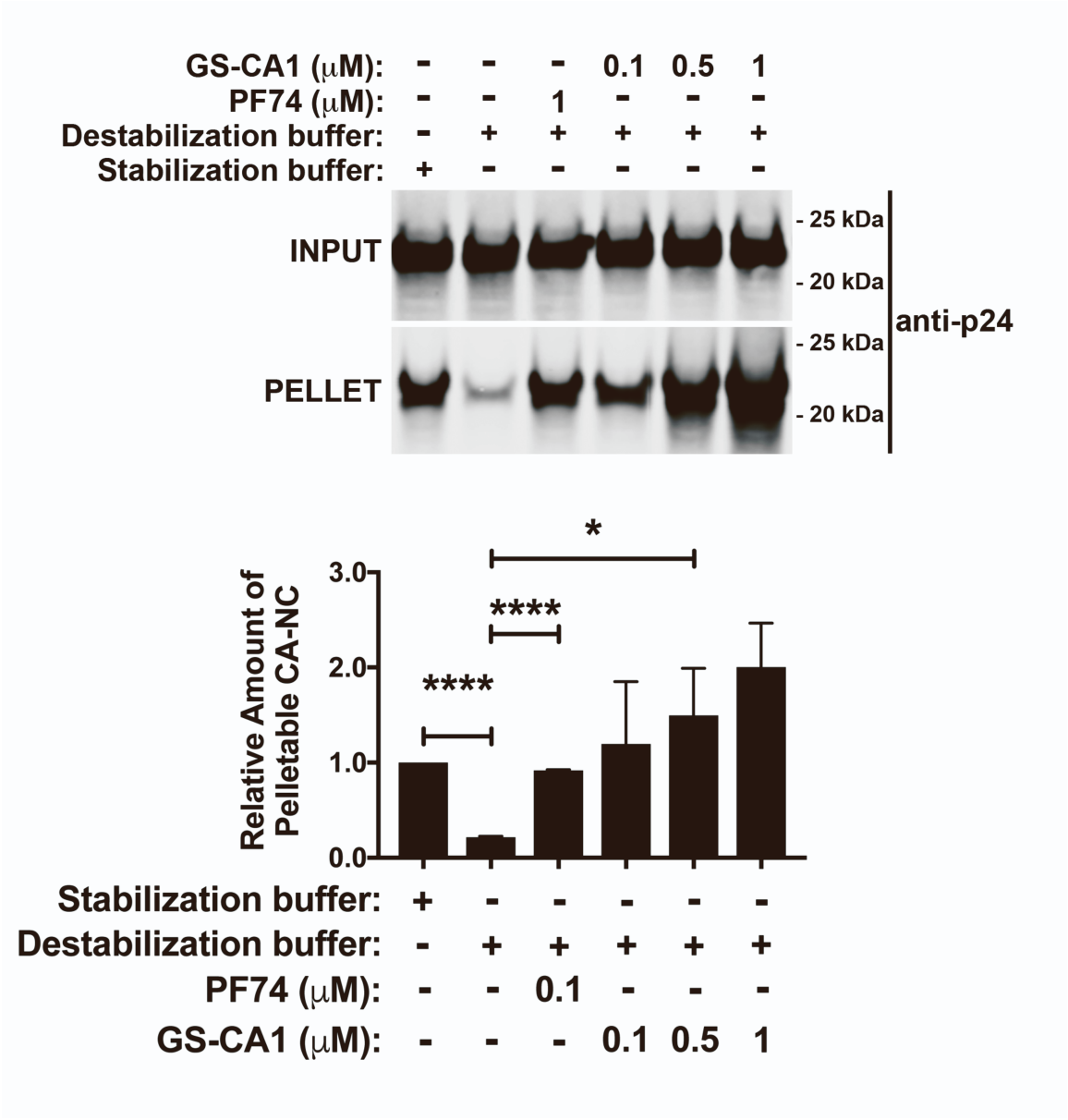

Figure S1. GS-CA1 stabilizes in vitro-assembled HIV CA-NC complexes, Related to Figure 3. HIV-1 CA-NC complexes were added to destabilization buffer with PF74, GS-CA1, or DMSO as a vehicle control. As a control, CA-NC complexes were added to the stabilization buffer. A fraction of each sample was stored as the input. Mixtures were incubated for 1 h at room temperature and pellets were harvested by ultracentrifugation. INPUT and PELLET samples were analyzed by western blotting using anti-p24 antibodies. The relative amounts of pelletable CA-NC protein were determined from western blots for three experiments (a representative western blot is shown).The mean relative amounts of pelletable CA-NC ± SD are shown. * indicates P-value < 0.005, **** indicates P-value < 0.0001 as determined by using the unpaired t-test.

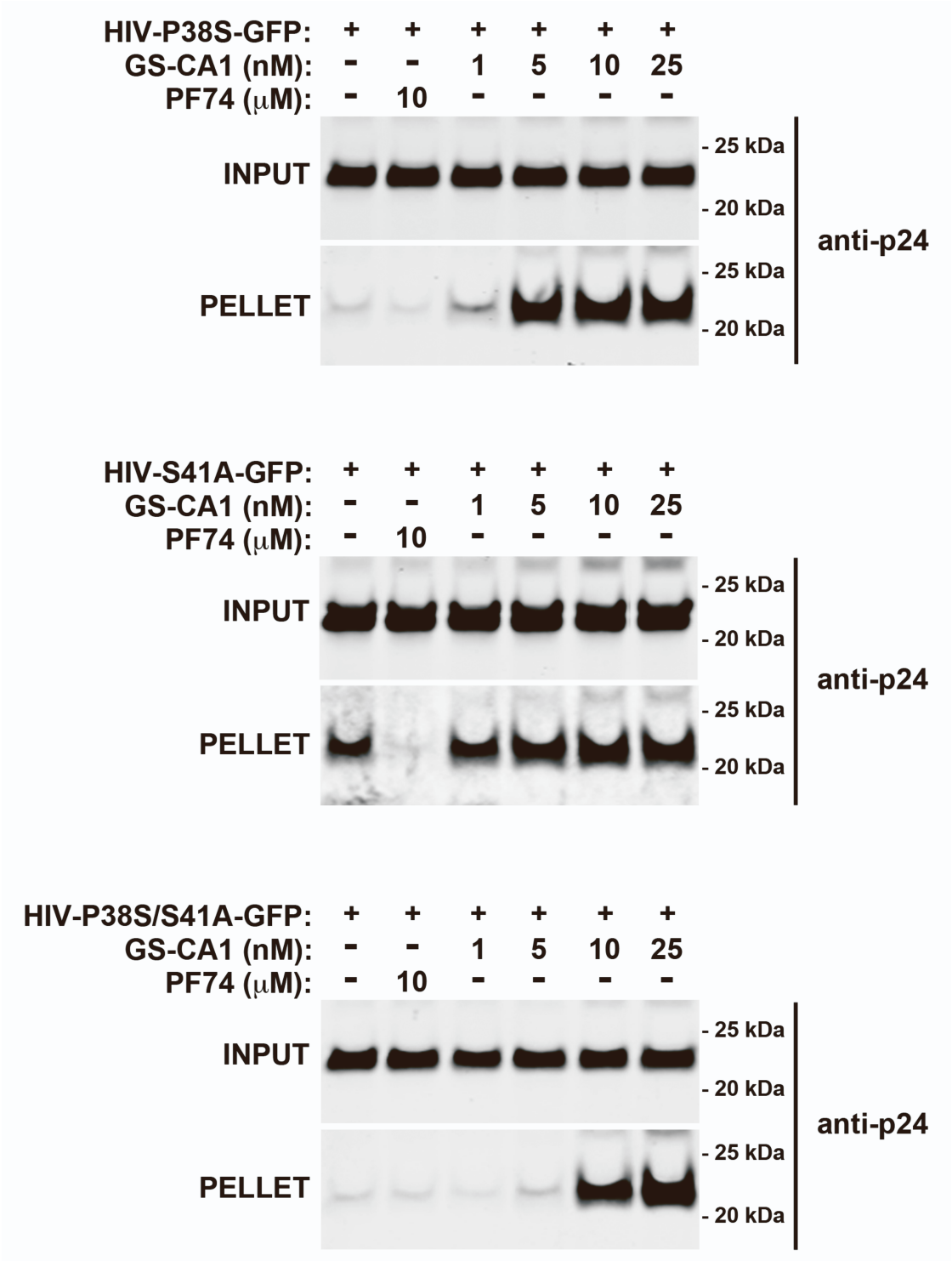

Figure S2. GS-CA1 stabilizes P38S, S41A, and P38S/S41A mutant HIV-1 cores during infection, Related to Figure 3A. Human A549 cells were incubated with HIV-1-P38S-GFP, HIV-1-S41A-GFP, or HIV-1-P38S/S41A-GFP with PF74, GS-CA1, or DMSO as a vehicle control. After 16 h at 37°C cell extracts were fractionated and INPUT and PELLET fractions were analyzed by western blotting using antibodies against the HIV-1 p24 capsid protein. Experiments were repeated at least three times and a representative figure is shown.

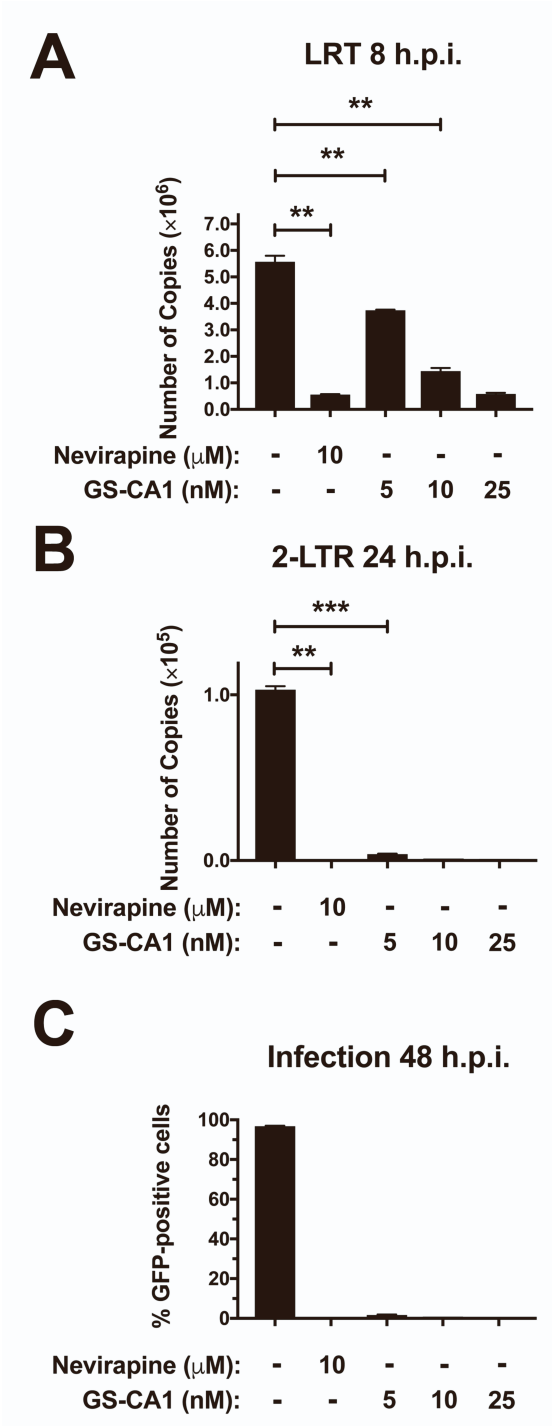

Figure S3. GS-CA1 does not inhibit reverse transcription but inhibits HIV-1 DNA integration into the host genome, Related to Figure 4A. (*A*, *B*, and *C*) A549 cells were infected with wild-type HIV-1-GFP at MOI = 2 with 10 μM nevirapine, GS-CA1, or DMSO as a vehicle control. After incubation for the indicated times, DNA was extracted from infected cells and analyzed for HIV-1 late reverse transcription products (LRTs) (*A*) or 2-LTR circles (*B*) by quantitative PCR. (*C*) Infected GFP-positive cells were determined at 48 h post-infection. Experiments were repeated three times and representative figures are shown. Measurements were performed in triplicates, and the mean ± SD are shown. ** indicates P-value < 0.001, *** indicates P-value < 0.0005, as determined by using the unpaired t-test.