

Fig S1

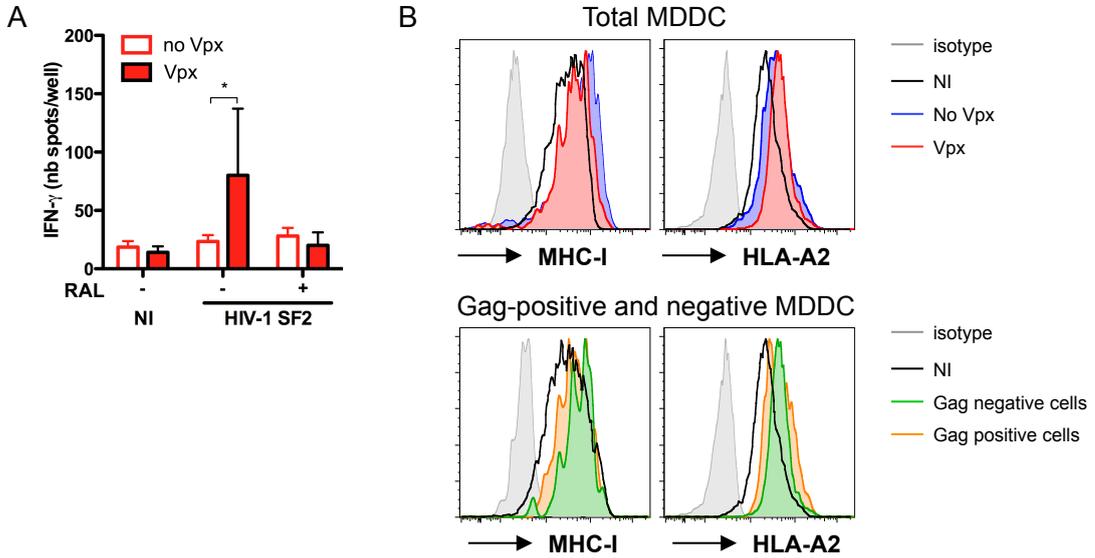


Fig S1. Vpx-treated MDDC infected with wild-type HIV-1 efficiently stimulate HIV-1-specific CTL (A) 10^6 Monocyte-derived dendritic cells (MDDC) were pre-treated for 2h with SIVmac VLP, containing or not Vpx, and infected with wild-type HIV-1 SF2 or NL4-3 strain (100ng p24 and 300ng p24 respectively). After 48h of infection, cells were cultured with EM40F21 for 18h in an IFN- γ ELISPOT assay and the number of IFN- γ spots were determined per well. Background IFN- γ secretion by CTL cultured with non-infected (NI) MDDC is represented. Data show mean of duplicates. (B) Vpx-treated MDDC infected with VSV-G-pseudotyped SF2 for 72h were analyzed by flow cytometry. Histograms show expression of MHC-I and HLA-A2 in total cells (top panels) and in Gag-positive and Gag-negative cells (bottom panels).

Fig S2

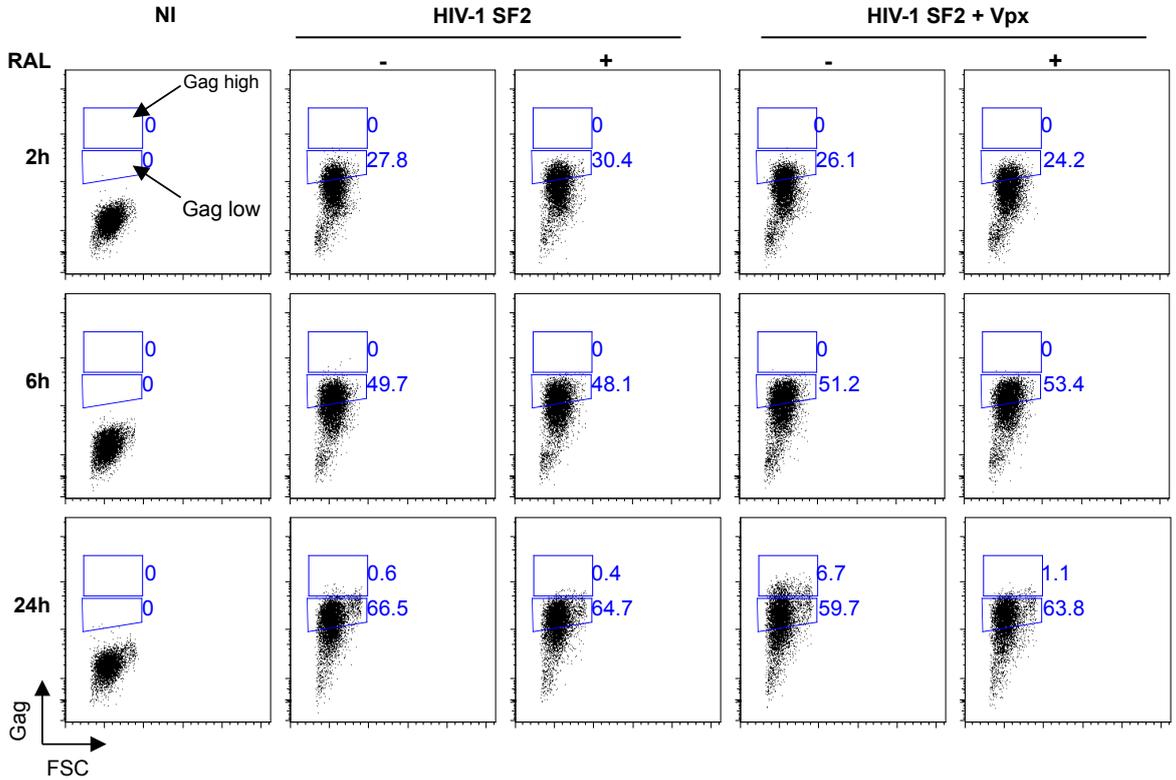
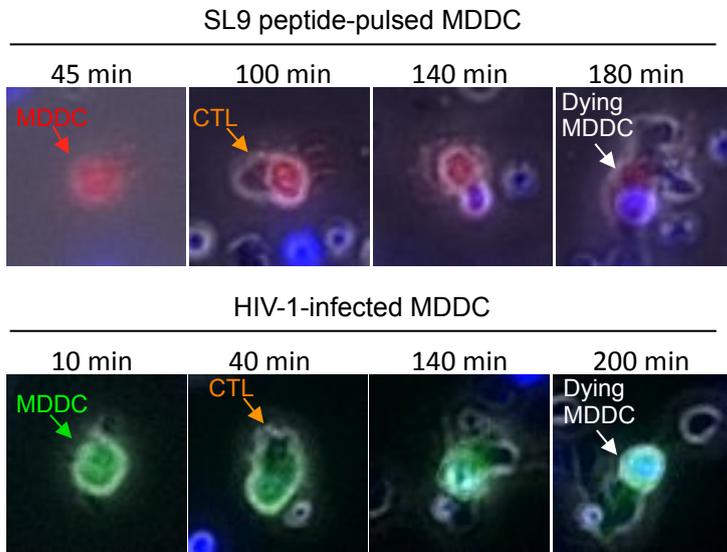


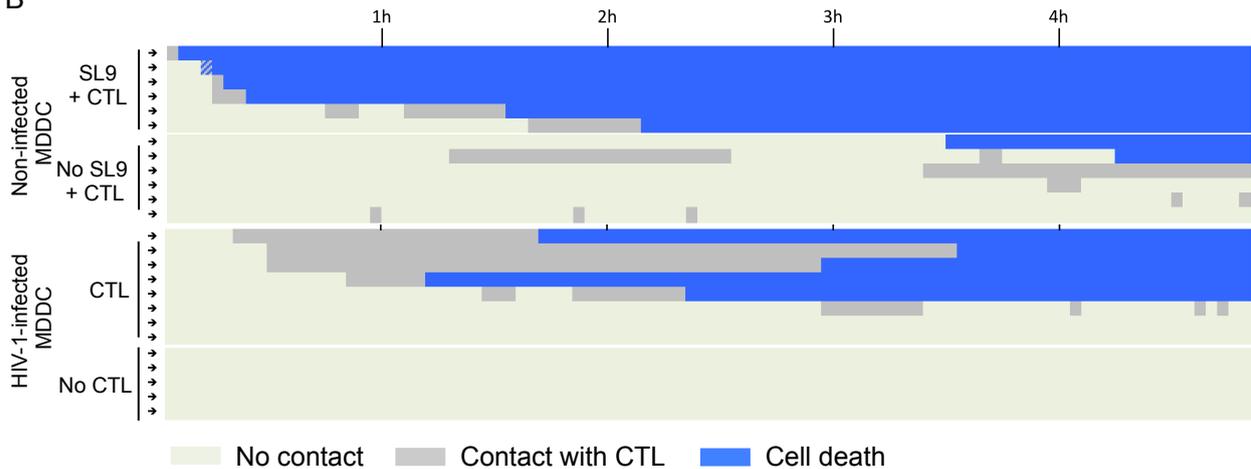
Fig S2. Capture of high viral loads by MDDC at early time points. 10^6 VLP-treated MDDC were infected with 100ng of VSV-G-pseudotyped SF2 for 2h. Cells were extensively washed and incubated in fresh medium. Flow cytometry analysis was performed on cells collected 2h post-infection (directly after washing), 6h (4h after incubation with fresh medium) and 24h post-infection. Dot plots show percentage of cells expressing Gag in one representative experiment.

Fig S3.

A



B



C

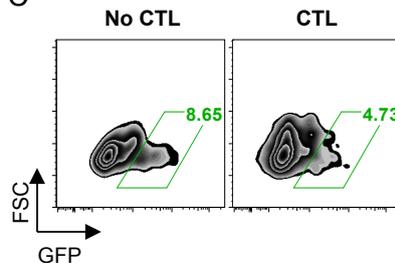


Fig S3. Live imaging of MDDC and CTL co-cultures (A) MDDC from donor #2 were infected as in figure 4 and imaged by time-lapse microscopy in the presence of DAPI dye. Green cells represent live Vpx-treated MDDC infected with NL4-3-IRES-GFP and blue cells represent dead cells. As a control, uninfected CMRA-labelled MDDC (red cells) exposed to the SL9 peptide were co-cultured with CTL (top panels). (B) Individual GFP⁺ or CMRA⁺ cells were tracked and time of contact with CTL (in grey) and cell death (in blue) were quantified for each cell. Each line/arrow corresponds to one MDDC tracked over 5 hours (h). (C) Total cells from (A) were recovered from U-dishes after 5 hours of filming, fixed and monitored for GFP by flow cytometry. Dot plots show percentage of GFP⁺ cells gated on MDDC from donor #2.

Fig S4

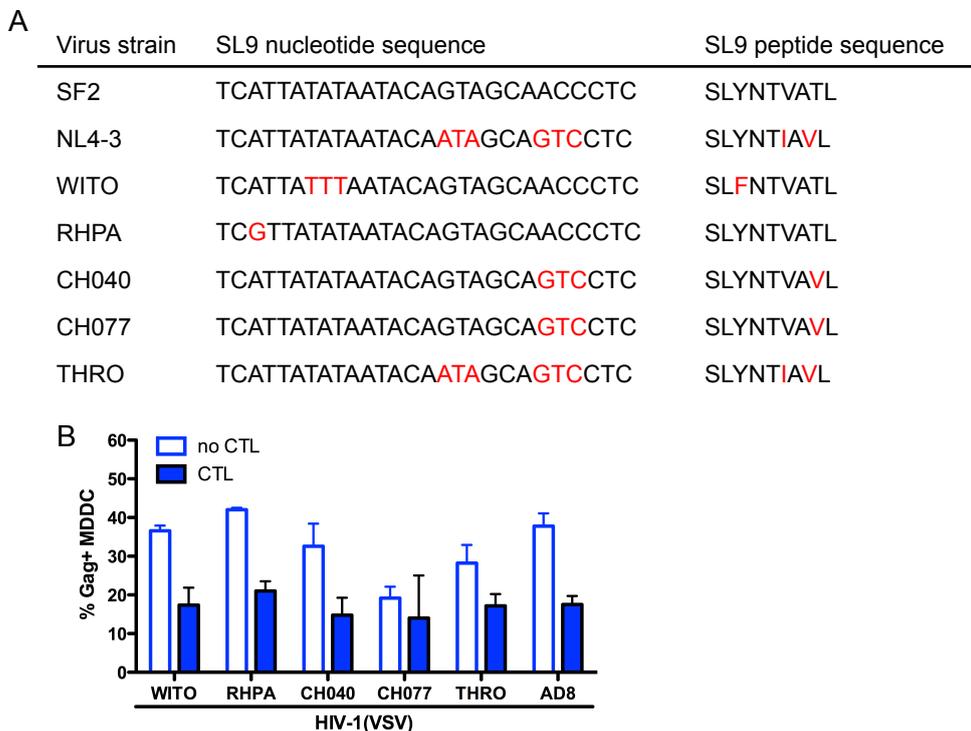


Fig S4. CTL decrease Vpx-mediated infection of MDDC by VSV-G-pseudotyped transmitted/founder HIV-1. (A) Sequence alignment of the SL9 epitope region in Gag p17 of T/F HIV-1 strains. (B) Vpx-treated MDDC were infected with equal amounts of the indicated VSV-G-pseudotyped HIV-1 strains (25ng p24/10⁶ cells) for 48h and cultured with EM40F21 CTL for 24h. Cells were fixed and stained for DC-Sign and Gag and analyzed by flow cytometry. Data represent mean (\pm SD) percentage of Gag from three independent experiments.

Supplementary movies

Figure 4A represents still images of the movies. Untreated MDDC were exposed to control SL9 peptide (S1 movie) and Vpx-treated MDDC (donor #1) were infected with NL4-3-IRES-GFP (S2 movie). Cells were plated with EM40F21 CTL at a 1:3 ratio on U-dishes coated with fibronectin and imaged by time-lapse microscopy. Red cells (S1 movie) represent CMRA-labelled MDDC exposed to SL9 peptide and green cells (S2 movie) represent infected MDDC. In both experiments, DAPI dye was added into the medium to identify dead cells (blue cells). The green arrow designates live MDDC. The white arrow indicates contact with CTL. The blue arrow shows dying cells following contact with CTL.