Vpx-mediated degradation of SAMHD1 does not render hematopoietic progenitor cells more permissive to lentiviral gene transfer

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Background
Understanding how to achieve efficient transduction of hematopoietic progenitor cells (HPC), while preserving their long-term ability to self-renew, is key to applying lentiviral-based gene therapy strategies in the treatment of hematopoietic diseases. SAMHD1, a restriction factor of retroviruses, is highly expressed in hematopoietic progenitor cells, while it is not expressed in mature hematopoietic cells. SAMHD1 interferes with reverse transcription by decreasing the nucleotide pool, blocking HIV-1 infection of hematopoietic progenitor cells. In the present study, we used Vpx-mediated degradation of SAMHD1, SAMHD1 shRNA, HIV-2 or SIV-based virus-like particles (VLPs) to study whether the decrease of SAMHD1 or accumulation of dNTPs in MDMs, Jurkat cells and monocytic cells interferes with reverse transcription by decreasing the nucleotide pool. Vpx originates from HIV-2 and HIV-1 and relieves this block in reverse transcription through the proteasomal degradation of SAMHD1. We hypothesized that SAMHD1 might be operative in HIV-1 vector-based HPC transduction.

Methods
We used Vpx-mediated degradation of SAMHD1, SAMHD1 shRNA, HIV-2 or HIV-1-based lentiviral vectors to provide an increased dNTP pool in cultured HPC. We used lentiviral vectors which do not interfere with reverse transcription by decreasing the nucleotide pool. HIV-2 and SIV-based virus-like particles (VLPs) were used as tools to study whether the decrease of SAMHD1 or accumulation of dNTPs in MDMs, Jurkat cells and monocytic cells interferes with reverse transcription by decreasing the nucleotide pool. Vpx originates from HIV-2 and HIV-1 and relieves this block in reverse transcription through the proteasomal degradation of SAMHD1. We hypothesized that SAMHD1 might be operative in HIV-1 vector-based HPC transduction.

Results
Our results show that SAMHD1 is highly expressed in HPC cultured in a medium enriched with cytokines. In contrast, uncultured HPC have poor SAMHD1 expression. Expression levels of SAMHD1 in cultured HPC are even comparable with those found in myeloid cells, including monocytes and MDMs. However, Vpx virus-like particles (VLPs) did not result in a higher transduction rate in Phase III clinical trials. The sterile alpha motif (SAM) domain and HD domain-containing protein 1 (SAMHD1) was recently identified as an HIV-1 restriction factor in myeloid and resting CD4+ T cells. SAMHD1 was identified as an HIV-1 restriction factor in myeloid and resting CD4+ T cells. SAMHD1 level was still comparatively high. Lastly, Vpx+ VLPs led to a striking increase of dNTPs in MDMs. However, we observed only a minor increase in HPC, which corresponded to the observation that Vpx+ VLPs did not result in an accumulation of integrated provirus in HPC.

Conclusion
In summary, cytokine exposed HPC, unlike quiescent cells, express high levels of SAMHD1. However, the Vpx-mediated decrease of SAMHD1 levels was not sufficient to increase the lentiviral-based transduction rate. Alternatively, other restriction factors might be operative in HPC.

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