Targeted Tat mRNA latency reversal augmented by IL-7 signaling

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Although considered an essential component of an HIV cure strategy, efforts to reactivate latent HIV have largely failed due to barriers attributable to both the host and virus. These obstacles include heterogeneity in cell state, such as reservoir cell quiescence, and virus-specific blocks to latency reversal, including inefficient transcriptional elongation. We recently showed that ibalizumab (αCD4)-targeted delivery of mRNA encoding Tat overcomes a viral barrier to gene expression with minimal perturbation to host transcriptome and cell state. Whether the effect of transient Tat expression could be enhanced with alterations to cell state remains unexplored. Several cytokines including IL-2, IL-7, and IL-15 have been explored for latency reversal potential due to their combined effect on cell state and viral transcription. Of particular interest is IL-7, which drives homeostatic T cell proliferation and increases viral replication via phosphoSTAT5 (pSTAT5) interaction with the HIV promoter. However, IL-7 administration alone in antiretroviral therapy treated people with HIV (ART-PWH) failed to reactivate viral replication in two clinical trials. To test the hypothesis that simultaneously addressing cellular and viral barriers to latency reversal could enhance viral reactivation, we developed a novel intervention consisting of IL-7-conjugated HIV Tat-mRNA-lipid nanoparticles (IL7-Tat mRNA-LNP). We first assessed delivery of IL-7 conjugated mRNA-LNP to CD4+ T cells using a reporter GFP mRNA. In human PBMC, IL-7 receptor (CD127) expressing T cells treated with IL-7-GFP mRNA-LNPs showed robust GFP expression after 48 hours. Targeted CD4+ T cells also exhibited IL-7 associated biological outputs including pSTAT5 signaling 15 minutes post treatment and subsequent homeostatic proliferation with sustained GFP expression over 7 days. We next assessed whether we could deliver Tat mRNA with IL-7 conjugated mRNA LNPs. In addition to specific targeting and activation of CD127+ cells, IL7-Tat-mRNA-LNPs demonstrated potent viral reactivation within memory CD4+ T cells from fully suppressed ART-PWH (n=4). These cells treated with IL7-Tat-mRNA-LNPs showed intracellular and supernatant p24 expression superior to IL-7 alone, IL-7 GFP mRNA LNP, Ibalizumab-Tat-mRNA-LNP, and anti-CD3/CD28. We have demonstrated that targeted cytokine-mediated activation and expression of HIV Tat overcome surmountable blockades to HIV reservoir reactivation, yielding a potent and specific new approach to HIV latency reversal.