Investigation of effects of opioids on HIV replication and pathogenesis

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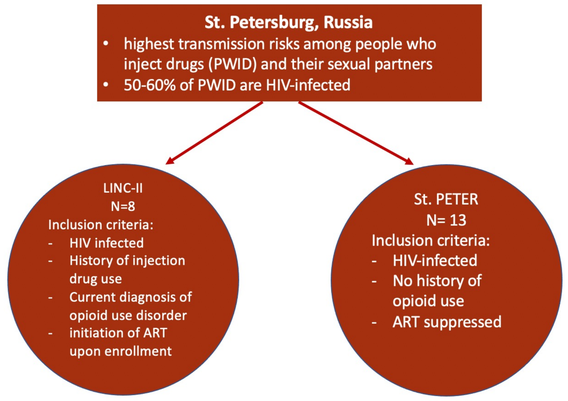
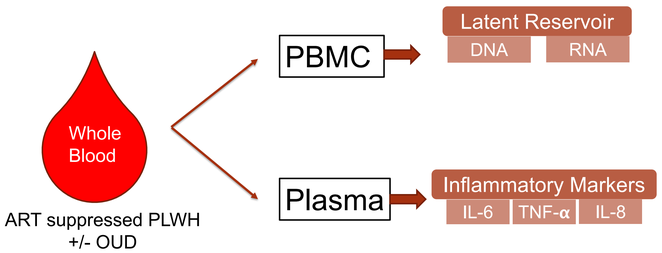
INTRODUCTION

- 13% of intravenous drug users(IDUs) are also HIV positive
- Studies over the past 30 years show that opioid abuse correlates with increased susceptibility to various viral and bacterial infections including HIV.
- How these drugs affect HIV pathogenesis is understudied.
- Opioids are a class of potent analgesics used in clinical setting and are also commonly abused class of drugs. Opioid derivatives, particularly morphine, have immunosuppressive effects on a wide range of immune cells including CD4 T cell, macrophages and dendritic cells.
- Opioids increase HIV infection in monocytes by upregulating expression of anti-HIV microRNA[3] and in neonatal monocyte derived macrophages by upregulating expression of CCR5[4], a coreceptor necessary for HIV entry.
- Impact of opioid use on HIV-1 latency is not well understood.

STUDY DESIGN



METHODS



Latent Reservoir is comparable between PLWH with or without Opioid use Disorder

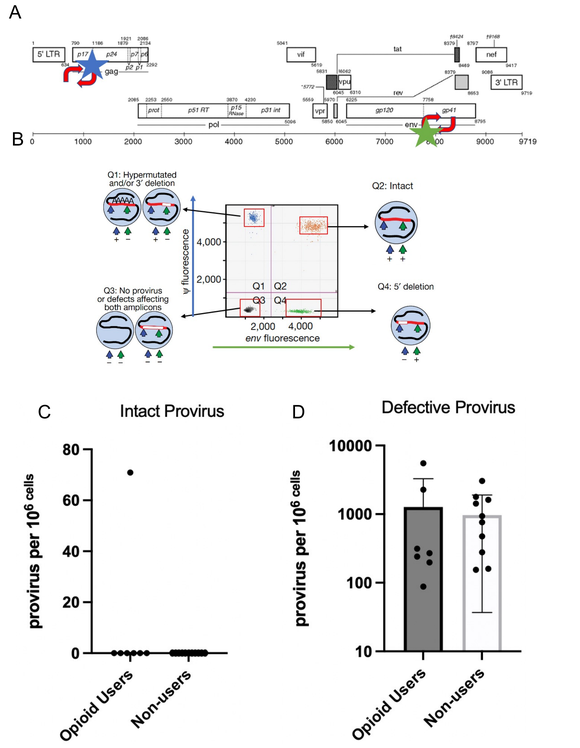
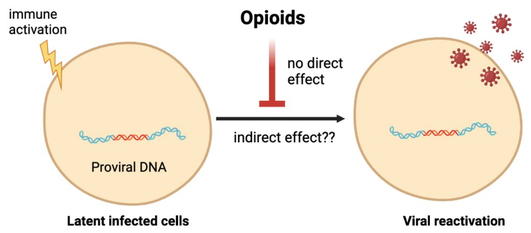


Figure 1. (A) Position of amplicons used in IPDA. (B) Representative IPDA[1] result. (C) Quantification of intact proviruses in PBMCs from ART treated PLWH with (n=8) and without opioid use disorder (n=13). (D) Quantification of defective proviruses in PBMCs from ART treated PLWH with opioid use disorder (n=8) or without any history of opioid use (n=13).

CONCLUSION

- We found no apparent effect of chronic opioid use on HIV proviral landscape in vivo.
- Chronic opioid use correlates with reduction in HIV reactivation in vivo.
- But in vitro, morphine has no effect on HIV reactivation from latently infected cells.



Reactivation of HIV-1 is limited in PLWH with Opioid use disorder.

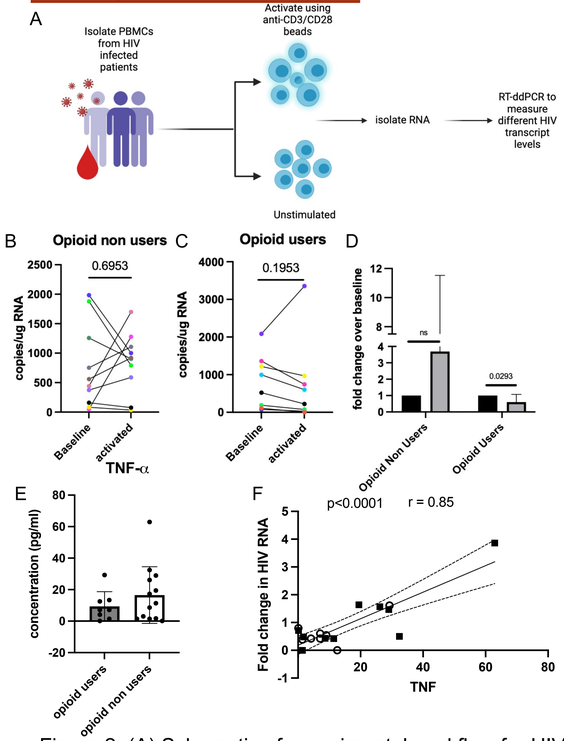


Figure 2. (A) Schematic of experimental workflow for HIV transcript measurement on PBMCs from HIV infected patients. Using RT-ddPCR, HIV transcripts were measured in unstimulated PBMCs and ex vivo activated PBMCs from ART treated PLWH (B) without opioid use disorder (n=13) and (C) without opioid use disorder (n=8). (D) HIV transcript levels in activated PBMCs were normalized to baseline HIV transcript levels and average fold change is plotted as bar graph (E) TNF-**α** level was measured in frozen plasma from PLWH with and without opioid use disorder. (F) Fold change in HIV transcripts upon activation correlation with TNF-**α** level in plasma.

Morphine does not affect HIV infection and latency reactivation in vitro

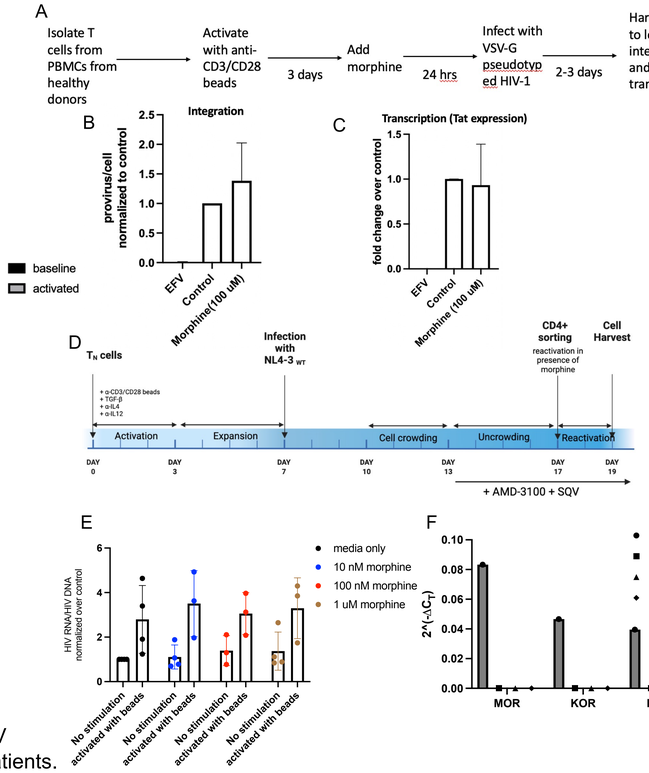


Figure 3. (A) Schematic of experiment design to investigate effect of morphine on HIV replication in primary CD4 T cells in vitro. (B) Activated primary CD4 T cells were treated with different concentrations of morphine 24 hrs before infection with NL4-3-Δenv-GFP and 3 days post infection cells were collected, and HIV integration was measured by Alu-gag PCR. (C) HIV tat expression as measured by qRT-PCR.

(D) Schematic of procedure used for the generation of human primary memory T cells and subsequent establishment of latent infections[5]. (E) Latently infected cells generated from a primary cell model of HIV latency was either left unstimulated with or without morphine or activated with PHA with or without morphine. Following 48 hrs of PHA stimulation, cells were collected for HIV DNA and HIV RNA measurement by qPCR. (F) Opioid receptor expression in brain, PBMCs, CD4 T cells and monocyte derived macrophage (MDMs). Y axis represents 2^(-**Δ**CT) where **Δ**CT = CT(Opioid receptor) – CT (RPL13A). MOR represents Mu opioid receptor, KOR represents Kappa opioid receptor and DOR represents Delta opioid receptor.

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