HIV-1 Enhances Sexual Transmission of Hepatitis C virus by Human Langerhans Cells

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Sexual transmission of HCV

Sexual transmission of Hepatitis C virus (HCV), until recently, was thought to be rare. However, there has been a significant rise in the incidence of HCV infection among HIV-infected men-who-have-sex-with-men (MSM) and studies suggest that HCV can be sexually transmitted within this population. The mechanisms facilitating this sexual transmission are unclear. Human Langerhans cells (LCs) reside in the mucosa and have been shown to be involved in limiting dissemination upon sexual contact by degrading HIV-1 and preventing HIV-1 transmission. The activation state of LCs changes susceptibility to HIV-1, leading to LC infection and subsequent HIV-1 transmission. In this study we investigated the role of LCs in HCV infection and transmission. We hypothesize that HIV-1 replication in HIV-1-infected MSM leads to mucosal changes that allow HCV entry and subsequent dissemination to hepatocytes.

Aim of the study

To investigate the role of HIV-1 coinfection on HCV mucosal transmission by Langerhans cells

Results

Primary Langerhans cells are present in HIV-1 positive anal mucosa biopsies

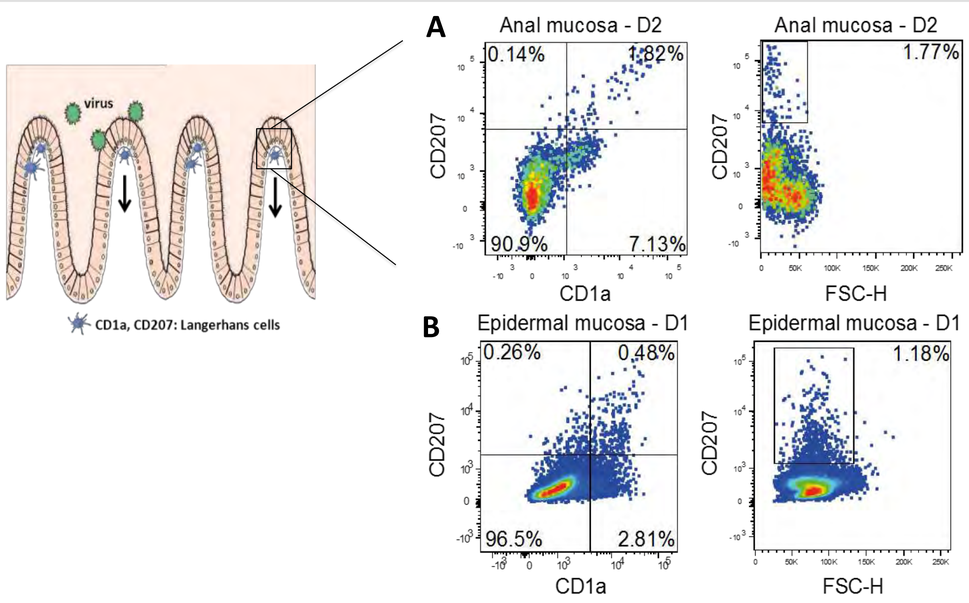


Figure 1. LCs are present in HIV-1 positive anal mucosa biopsies. (A) Single cell suspensions of anal biopsies from HIV-1 infected individuals were stained with antibodies against CD45, CD19, CD20, CD56, CD3, CD1a and CD207 and analyzed by flow cytometry. The percentage of cells present is depicted in the upper-right corner of the dot plots. **(B)** Single cell suspensions of epidermal sheets were stained with antibodies against CD1a and CD207 and analyzed by flow cytometry. One representative donor is depicted.

Immature LCs do not transmit HCV to hepatocytes

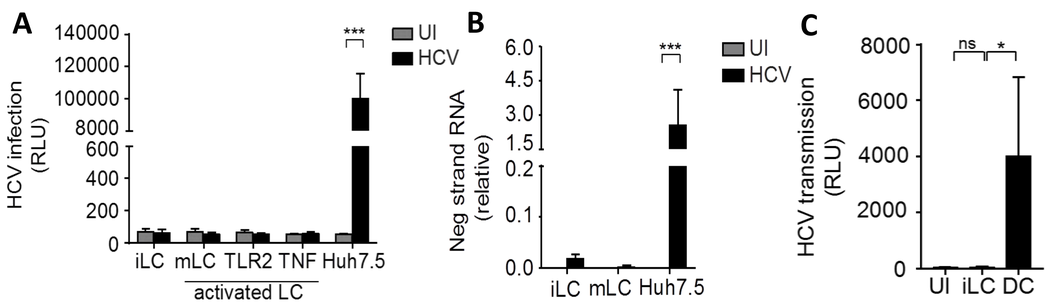


Figure 2 Immature LCs do not transmit HCV to hepatocytes. (A-B) Immature and emigrated LCs were isolated, stimulated with Pam3CSK4 (TLR2) and TNF, for 2 hours, infected with infectious HCV (JFH1-AM120-Rluc) for 6 days and analyzed for luciferase reporter activity. n=2 (immature LCs) and n=4 (emigrated LCs), ***P<0.001, by two-tailed, unpaired Student's t-test. **(B)** Immature and emigrated LCs were isolated and analyzed for negative strand RNA by real-time PCR normalized to beta-globuline. n=3 (immature LCs), n=4 (emigrated LCs) and n=3 (Huh7.5 cell line), ***P< 0.001, by two-tailed, unpaired Student's t-test. **(C)** Immature LCs and DCs were isolated and infected with infectious HCV for 24 hours, washed, co-cultured with huh7.5 cells and analyzed for luciferase reporter activity. n=3 donors, *P< 0.05, ns = not significant, by two-tailed, unpaired Student's t-test.

Activated LCs transmit HCV to hepatocytes

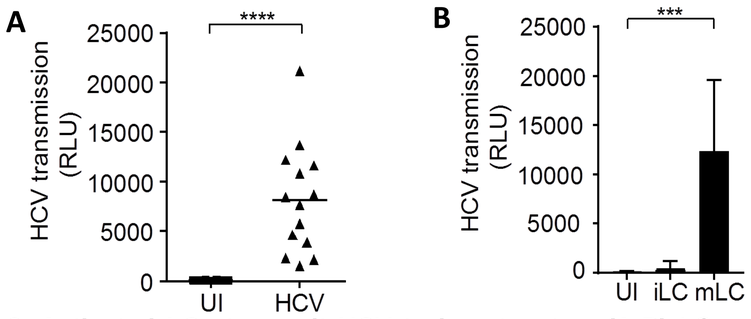
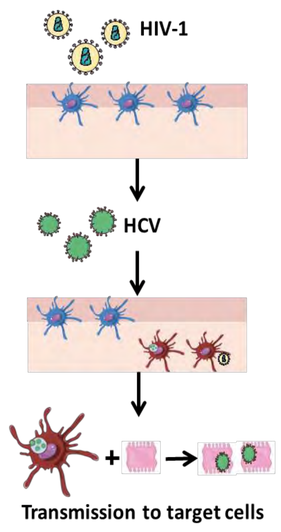


Figure 3. Activated LCs transmit HCV to hepatocytes. (A-B) LCs were exposed to pseudotyped HCV (HIV-1 NL4.3Δenv pseudotyped with HCV env glycoproteins E1 and E2) for 24 hours, extensively washed, co-cultured with huh7.5 cells and analyzed for luciferase reporter activity. **(A)** Emigrated LCs were isolated. n=14 donors, ****P<0.0001, by two-tailed, paired Student's t-test. **(B)** Immature and emigrated LCs from the same donor were isolated,. n=3, ***P<0.001, by two-tailed, paired Student's t-test.

HIV-1 replication is necessary for increased HCV transmission



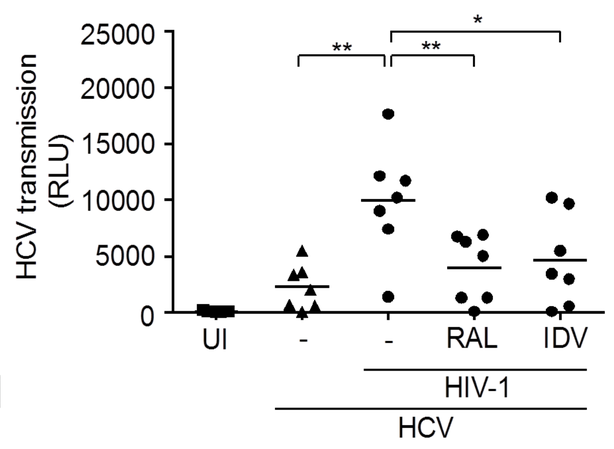


Figure 3. HIV-1 infection enhances HCV transmission ex-vivo but does not affect LC migration. Epidermal sheets were pre-exposed to HIV-1 replication inhibitors Raltegravir or Indinavir for 2 hours, exposed to either HIV-1 (JRCSF) or HIV-1 (SF162) for 48 hours and subsequently exposed to infectious HCV (JFH1-AM120-Rluc) for 24 hours. Cells were harvested, extensively washed, co-cultured with huh7.5 cells and analyzed for luciferase reporter activity. Each dot represent 1 donor. n=7 donors, *P< 0.05, **P<0.01, by two-tailed, paired Student's t-test. RAL: Raltegravir, IDV: Indinavir, HCV: Hepatitis C virus, UI: uninfected.

HIV-1 infection enhances HCV transmission *ex vivo* independent of HCV replication

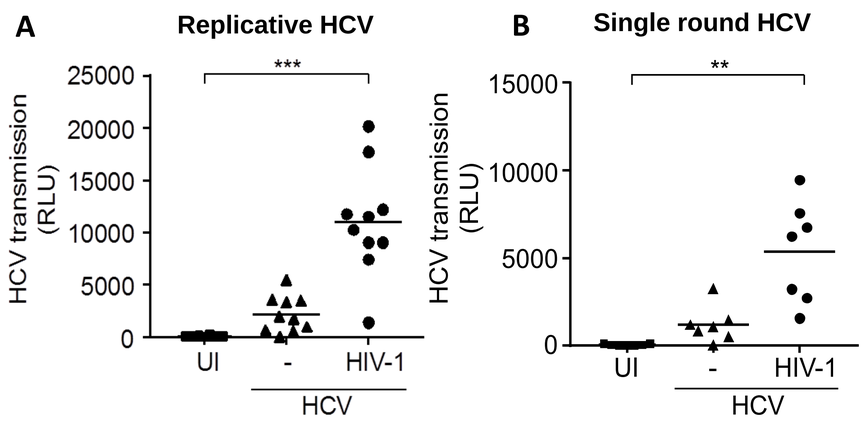


Figure 5. Activated LCs transmit HCV to hepatocytes independent of productive HCV replication. (A-B) Epidermal sheets were exposed to HIV-1 (JRCSF) for 48 hours and subsequently to infectious HCV (JFH1-AM120-Rluc) or pseudotyped HCV (HIV-1 NL4.3Δenv pseudotyped with HCV env glycoproteins E1 and E2) for 24 hours, extensively washed, co-cultured with huh7.5 cells and analyzed for luciferase reporter activity. **(A)** Each dot represent 1 donor. n=10 donors, ***P<0.001 by two-tailed, paired Student's t-test. **(B)** n=7 donors, **P<0.01, by two-tailed, paired Student's t-test.

Conclusion

- Immature LCs do not transmit HCV
- Coinfection with HIV-1 enhances HCV transmission by LCs
- HIV replication is necessary for increased HCV transmission
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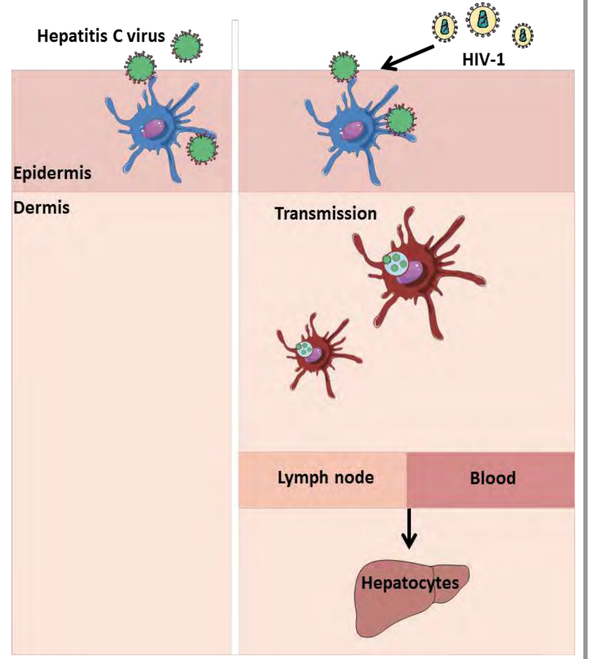
- Activated LCs transmit HCV to hepatocytes independent of productive HCV replication

Our results are important to understand how HIV-1 replication in mucosal tissues in HIV-1 infected MSM, changes LC function, which causes HCV capture and subsequent transmission to hepatocytes.

This novel transmission mechanism implicates an important determinant for HCV susceptibility after sexual contact



Correspondence and funding

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