Influx of Th1 Lymphocytes in the Intestine Mucosa Contributes to the Impaired Homing of Th17 Cells to the Gut in Treated HIV-1-Infected Individuals

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Background

During HIV-1 infection, the integrity of the intestinal immune barrier is disrupted due to a deep depletion of $CD4^+$ T cells in the gut, including Th17 cells. Gut homing of Th17 cells mainly depends on the CCR6-CCL20 chemotactic axis. We previously established that despite effective cART, Th17 cells homing to the small intestine mucosa is altered due to a reduced CCL20 production by enterocytes [**],[***].

[**]*Loiseau et al., CROI 2015 Seattle-poster 274, T Regulatory Cells Disrupt the CCL20-CCR6 Axis Driving Th17 Homing to the Gut*

[***]*Loiseau et al., Mucosal Immunology 2016, CCR6- regulatory T cells blunt the restoration of gut Th17 cells along the CCR6-CCL20 axis in treated HIV-1-infected individuals*

Aims

The regulating loop occurring between IL-17 secretion, CCL20 production, and Th17 cells gut homing was shown to be altered in treated HIV-1-infected individuals [**],[***]. Despite cART, persistence of various inflammatory stimuli in the gut mucosa could contribute to a disequilibrium in the intestinal immune homeostasis. We thus aimed to assess how Th1 cells could influence the homing of Th17 to the gut along the CCR6-CCL20 axis in this setting.

Methods

Small bowel biopsies were obtained by endoscopy in 10 $HIV\text{-}1^+$ and 10 $HIV\text{-}1^-$ individuals. Gut $CXCR3^+$ and Th1 cells frequency was evaluated by flow cytometry and immunohistochemistry. The effect of IL-2 and IFN-γ on CCL20 expression was assessed in an *ex-vivo* model of human primary enterocytes cultured as a tight monolayer on transwells. The impact of Th1 cells on CCL20 production was explored on cocultures done between the enterocytes and Th1 and/or Th17 cells to mimic lamina propria-epithelium interactions. The expression of CCL20 by the enterocytes was evaluated by qRT-PCR and ELISA.

Results

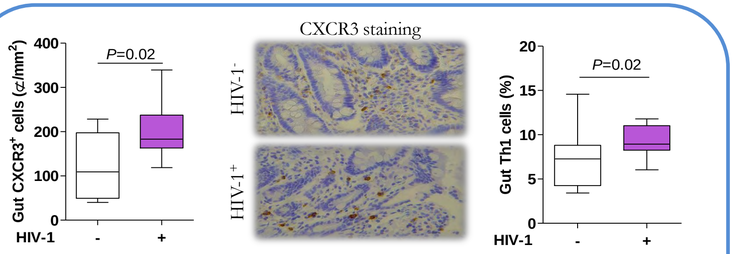


Figure 1 : CXCR3⁺ and Th1 cells homing is increased in the small intestinal mucosa of HIV-1-infected individuals. *CXCR3⁺ cells were quantified by immunohistochemistry and Th1 cells frequency was determined by flow cytometry.*

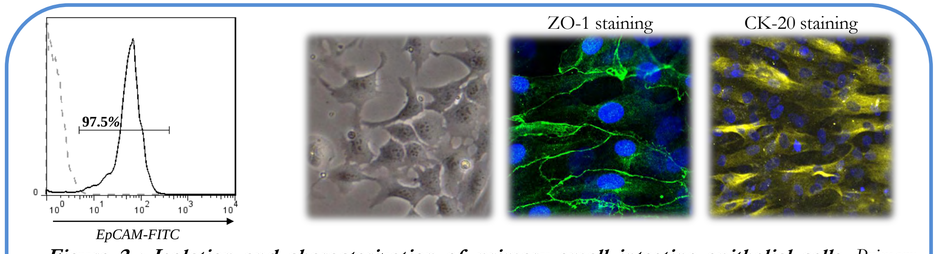


Figure 2 : Isolation and characterization of primary small intestine epithelial cells. *Primary small intestine epithelial cells were isolated from surgical specimens. Flow cytometric assessment of the purity of magnetically sorted EpCAM⁺ small intestine epithelial cells is shown on the left panel. Cells differentiation was revealed by a continuous ZO-1 staining (green) and expression of CK-20 (yellow). Microscopal imaging on an inverted confocal laser-scanning microscope (LSM 710, Zeiss), original magnification ×630.*

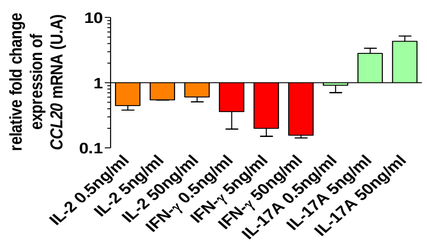


Figure 3 : Pro-inflammatory cytokines reduce expression of the CCL20 chemokine by the small intestine epithelial cells, in contrary to Th17 cytokine. *Primary intestinal epithelial cells were stimulated overnight with IL-2, IFN-γ or IL-17.*

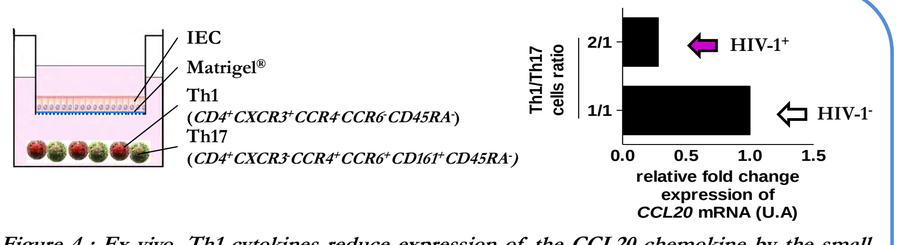
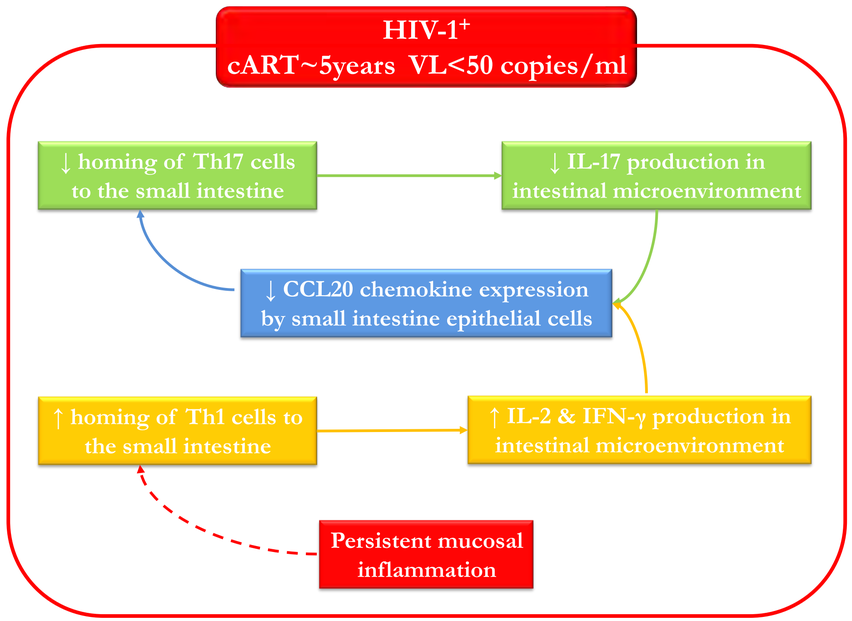


Figure 4 : Ex vivo, Th1 cytokines reduce expression of the CCL20 chemokine by the small intestine epithelial cells in contrary to Th17 cytokines. *Lymphocytes were isolated by flow cytometry and co-cultured overnight with primary intestinal epithelial cells as described on the left panel. Th1/Th17 ratio measured in the small intestine mucosa of uninfected and treated HIV-1-infected individuals are indicated by white and purple arrows respectively. IEC : Intestinal Epithelial Cells.*

Conclusion



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