Available online at www.sciencedirect.com

ScienceDirect

Current Opinion in Virology

Imaging lymphoid tissues in nonhuman primates to understand SIV pathogenesis and persistence

Claire Deleage[1], Baris Turkbey[2] and Jacob D Estes[1]

CD4+ T cells are the primary HIV-1 target cell, with the vast majority of these cells residing within lymphoid tissue compartments throughout the body. Predictably, HIV-1 infection, replication, localization, reservoir establishment and persistence, as well as associated host immune and inflammatory responses and disease pathology principally take place within the tissues of the immune system. By virture of the fact that the virus-host struggle is played out within lymphoid and additional tissues compartments in HIV-1 infected individuals it is critical to understand HIV-1 infection and disease within these relevant tissue sites; however, there are obvious limitations to studying these dynamic processes in humans. Nonhuman primate (NHP) research has provided a vital bridge between basic and preclinical research and clinical studies, with experimental SIV infection of NHP models offering unique opportunities to understand key processes of HIV-1 infection and disease that are either not practically feasible or ethical in HIV-1 infected humans. In this review we will discuss current approaches to studying the tissue based immunopathogenesis of AIDS virus infection in NHPs, including both analyses of tissues obtained at biopsy or necropsy and complementary non-invasive imaging approaches that may have practical utility in monitoring HIV-1 disease in the clinical setting.

Addresses
[1] AIDS and Cancer Virus Program, Leidos Biomedical Research, Inc., Frederick National Laboratory for Cancer Research, BG 535, Post Office Box B, Frederick, MD 21702, USA
[2] Molecular Imaging Program, National Cancer Institute, Building 10, Room B3B69F, Bethesda, MD 20814, USA

Corresponding author: Estes, Jacob D (estesj@mail.nih.gov)

Current Opinion in Virology 2016, **19**:77–84

This review comes from a themed issue on **Animal models for viral diseases**

Edited by **Luca Micci** and **Mirko Paiardini**

For a complete overview see the Issue and the Editorial

Available online 1st August 2016

http://dx.doi.org/10.1016/j.coviro.2016.07.002

Introduction

HIV-1 pathogenesis and disease progression result from the cumulative effects of complex and dynamic host–viral interactions that begin during the early acute stage of infection and in the absence of therapeutic intervention continue through end-stage disease. Upon viral transmission to a susceptible host, host innate and adaptive immune responses are induced and amplified within infected tissues. However, some of these responses, intended to limit or clear the infection, can also have deleterious effects, potentially enhancing viral replication and inducing inflammation that can lead to tissue damage and immunopathology. This host–viral interplay is exemplified in the gastrointestinal (GI) tract, where extensive early viral replication results in massive depletion of lamina propria CD4+ T cells, loss of important mucosal immune cell subsets (i.e. CD103+ DCs, TH17 cells, and IL-22+ lymphocytes) [1] and GI tract mucosal damage leading to microbial translocation and resultant systemic inflammation and pathological immune activation [2,3]. Because HIV-1 replication, host responses and resultant disease pathology occurs within tissues (i.e. lymphoid tissues, GI tract, CNS, among others) of infected humans it is critical to understand the disease process within these tissues where the host and virus interact. Nonhuman primate (NHP) research provides a vital bridge between basic and preclinical research and clinical studies. Experimental SIV infection of NHP models offer unique opportunities that are either not practically feasible or ethical in HIV infected humans, such as: (i) examining the critical early stages of infection following a known time of viral transmission, with a defined virus, particularly for mucosal transmission; (ii) pathogenesis studies with longitudinal tissue sampling; and (iii) testing novel preventative, intervention and 'cure' strategies that are conceptually unproven and/or potentially harmful. NHPs provide the most physiologically relevant HIV-1 models, compared to alternative small animal models such as humanized mice, with the additional advantage of longitudinal tissue sampling, up to and including tissues obtained at scheduled necropsy, allowing a level of extensive immunopathologic tissue based analysis that is not possible studying HIV-infected humans [4–7]. In this review we will cover current approaches to studying the tissue based immunopathogenesis of HIV/SIV virus infection in NHPs, including both analyses of tissues obtained at biopsy or necropsy and complementary non-invasive imaging approaches.

SIV pathogenesis: imaging GI tract damage and LT fibrosis

Tissue based analyses in SIV infected NHPs focusing on tissue compartments relevant to immunopathogenesis have been essential in understanding key aspects and

processes that drive lentiviral disease progression. Given that the intestinal immune system is considered the largest single immunologic organ in the body, containing upwards of 40% of all CD4+ T lymphocytes [8,9], the preferred cellular target for the virus [10], it is not surprising that this organ system is impacted early and severely by HIV/SIV infections and plays a key role in disease progression. Tissue based histological imaging studies have demonstrated that damage to the GI tract epithelial barrier shortly after SIV infection leads to local translocation of microbial constituents from the lumen of the intestine into the lamina propria and distal dissemination into systemic tissue compartments [3]. Deep sequencing analysis of bacterial DNA isolated from tissues of infected animals revealed a preference for bacterial translocation of the phylum Proteobacteria, suggesting that these bacteria preferentially translocate into the host [11]. These observations provided compelling direct evidence for GI tact pathology, in particular, alterations in intestinal structural integrity and function leading to translocation of bacteria with putative pathogenic bacterial species that may represent an even greater potential for immune activation [11]. Collectively, these data strongly suggest that GI tract damage plays a key role in contributing to the heightened state of chronic inflammation and pathological immune activation, as well as progressive immune deficiency and immune deregulation during SIV infection.

However, direct evidence that GI tract damage leading to microbial translocation independently causes systemic inflammation and immune activation was still lacking. To this end, we developed a NHP model of GI tract damage *in the absence of SIV infection* to directly determine the connection between GI tract damage, microbial translocation and systemic inflammation and immune activation as an important independent driver of characteristic pathologic features observed in HIV/SIV infections [12••]. We developed a NHP model of chemically induced colitis, utilizing dextran sulfate sodium (DSS), in SIV uninfected rhesus macaques (RMs) and demonstrated that damage to the integrity of the GI tract mucosal barrier, in the absence of SIV infection, caused local and systemic microbial translocation, with corresponding inflammation and immune activation similar in scope and character to that seen in chronically SIV-infected animals [12••]. Importantly, sustained GI tract damage recapitulated hallmark pathological features of the SIV disease, including fibrosis of secondary lymphoid tissues [12••]. This study directly demonstrated that GI tract damage leading to microbial translocation is independently sufficient to drive local and systemic inflammation and immune activation in the absence of SIV infection, and highlights GI tract damage as a key pathological feature of HIV/SIV disease where adjunctive therapy to modulate these processes could provide clinical benefit, independent of any direct impact on viral replication.

While histological imaging was necessary to demonstrate that GI tract damage leading to microbial translocation drives local and systemic inflammation and immune activation and associated tissue pathology; given the limited accessibility of tissue samples in patients, developing novel non-invasive clinically relevant techniques to monitor and image these pathogenic processes in HIV infected individuals is essential. Thus, we explored the utility of using an FDA approved gadolinium-based magnetic resonance imaging (MRI) contrast agent (gadofosveset trisodium; Ablavar®) that reversibly binds to serum albumin and has been used clinically for magnetic resonance angiography to diagnose vascular disease [13] to monitor and quantify GI tract inflammation in our SIV-negative NHP colitis model [12••]. We longitudinally imaged RMs, acquiring fat saturated T1-weighted MRIs following sequential DSS treatment cycles (which induced histologically documented sustained chronic colitis, inflammation, immune activation, and microbial translocation) and performed quantitative MRI scoring, assessing the severity of colitis using clinically relevant MRI criteria for bowel inflammation [14]. In this NHP model of chemically induced colitis we demonstrated the utility of utilizing a novel non-invasive MRI approach for evaluating the severity of colitis longitudinally, with findings corroborated by parallel histopathological analyses, suggesting its potential usefulness in the clinical setting (Figure 1). While these studies have obvious translatability to HIV-1 disease, these results may also have broad applicability in noninvasive monitoring of other diseases like IBD and colon cancer in human patients.

The central feature of HIV-1 disease is the progressive loss of $CD4^+$ T cells. While monitoring CD4+ T cell numbers in the peripheral blood has provided key insights into HIV-1 disease and is a clinically validated surrogate marker for staging HIV-1 infection that allows convenient monitoring of patients, it is clear that the peripheral blood compartment represents only a small fraction of the body's total CD4+ T cell population [15•]. Secondary lymphoid tissues (i.e. lymph nodes, spleen, gut associated lymphoid tissue (GALT), among others) harbor most of the body's CD4+ T cells and play an essential role in their survival and maintenance. CD4+ T cell loss in HIV-1 infection is a dynamic and progressive process of cell death due to infection, activation, and loss of needed survival signals that is played out within lymphoid tissues (LTs) [12••,16–19]. A variety of different mechanisms have been invoked to account for the progressive loss of CD4+ T cells, but progressive inflammation related structural damage to secondary LTs has been demonstrated to play a significant role in this process [2,3,12••,20–23].

Studies in SIV infected NHPs have been essential in understanding this process and demonstrated that multiple pathological changes in the morphology, structure and