Loss of blood CD11c⁺ myeloid and CD11c⁻ plasmacytoid dendritic cells in patients with HIV-1 infection correlates with HIV-1 RNA virus load

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Human blood contains at least 2 subpopulations of antigen-presenting dendritic cells (DCs) that can be differentiated by their expression of CD11c. Myeloid DCs (myDCs), which are CD11c⁺, trap invading pathogens in the tissues and then migrate to lymphoid tissues where they stimulate pathogen-specific T-cell responses. Plasmacytoid DCs (pcDCs), which are CD11c⁻, secrete interferon-α in response to viral infections. This study reports that in HIV-1 infection there is a progressive depletion of both these DC populations and that this correlates with an increasing HIV-1 plasma virus load. The median numbers of myDCs and pcDCs were 6978/mL and 9299/mL, respectively, in healthy male controls and 2298/mL and 1640/mL, respectively, in patients with more than 10⁵ HIV-1 RNA copies/mL. Both DC populations expressed CD4, CCR5, and CXCR4. The findings suggest that loss of DCs in HIV infection may contribute to disease progression.

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were differentiated into the myDC and pDC subpopulations on the basis of their expression of CD11c. The concentration of DCs in blood was estimated by using the percentages generated by the FACS analysis and the original PBMC count.

CD4 and chemokine expression by DCs

Four-color flow cytometric analysis was done on PBMCs from healthy donors. Cells (5 \times 10^5) were labeled with antibodies against the lineage mixture, HLA-DR and CD11c, and allophycocyanin-conjugated anti-CD4, anti-CCR5, or anti-CXCR4 (Pharmingen). The 2 DC populations were identified, and expression of CD4 and chemokine receptors was assessed.

Statistical analysis

The significance of the differences in DC numbers in patients and controls was evaluated with the Mann-Whitney U test. A P value of .05 or less was considered to represent significance. SPSS software (Chicago, IL) was used for the statistical comparisons. The Pearson correlation (r) was used to show regression of DC numbers with increasing virus load and decreasing CD4 count.

Results and discussion

DCs were defined by the absence of labeling with cell-lineage–specific antibodies and expression of HLA-DR (Figure 1A). Labeling with anti-CD11c separated the DCs into 2 populations corresponding to CD11c^+ myDCs and CD11c^- pcDCs (Figure 1A). In the 16 samples from male controls, there was a large variation in the number of blood DCs (range, 7553-44 715 DCs/mL; median, 17 566 DCs/mL). In patients with HIV-1 infection, there was a progressive loss of DCs with an increasing viral load (r = -0.43, P < .01; Figure 2A). In patients with a virus load greater than 1 \times 10^5 copies/mL, the median number of DCs was reduced to 4 102 DCs/mL (P < .01). Despite a marked reduction in the total population of mononuclear cells, the reduced DC number was not a result of a general impairment in hematopoiesis, since the percentage of DCs in the mononuclear cell population also decreased progressively with an increasing virus load (r = -0.43; P < .01; Figure 2B).

The median percentage of total DCs was 0.82% in controls and reduced to 0.37% in patients with viral loads greater than 1 \times 10^5 copies/mL. In contrast, the percentage of monocytes in the patients was unchanged, suggesting that there was not a general loss of myeloid cells. The progressive loss of DCs was not restricted to either cell population: the numbers of both myDCs and pDCs were reduced (r = -0.32 and r = -0.43, respectively; Figure 2C and 2D). The decrease in DC numbers with increasing viral load also correlated with CD4 count, with the median number of DCs reduced to 3904/\text{mL} (P < .01) in patients with CD4 counts lower than 100. There was a weaker correlation with CD4 counts than with virus load for both pDCs and myDCs.

Because myDCs give rise to tissue dermal and Langerhans-type DCs, and these cells function to acquire pathogens and present pathogen-derived peptides to T cells in the secondary lymphoid tissue, the progressive loss of myDCs may be at least partly responsible for the impairment of virus-specific CTL responses in the late stages of HIV-1 disease. CD11c^- pDCs have been reported to be a major source of IFN-\alpha in vivo.\textsuperscript{11,16,17} Loss of these cells, such as in our patients, would reduce levels of IFN-\alpha and thereby contribute to increased virus loads. Some of the loss may represent recruitment to the lymph nodes rather than depletion. Nevertheless, maintenance of pDC numbers in the blood may be important in the successful treatment of HIV-1 infection.

Infection by HIV is one possible mechanism for loss of DCs from the blood. We therefore analyzed DCs for expression of CD4 and the CCR5 and CXCR4 chemokine coreceptors in healthy individuals (Figure 1B). CD4 was expressed by both populations of DCs but at a higher level on pDCs. Low but clearly evident expression of CCR5 and CXCR4 was detected on both pDCs and myDCs. Therefore, DCs may be susceptible to infection with HIV-1.

This study showed a progressive depletion of both myDCs and pDCs in patients with HIV-1 infection. This depletion may impair generation of HIV-1–specific CTL responses and reduce levels of the antiviral cytokine IFN-\alpha, and as a consequence of these changes, virus loads may increase. Restoration of full immunocompetence in patients receiving antiviral therapy may depend on reconstitution of the DC system; thus, treatment with drugs that promote generation of DCs, such as Flt3 ligand,\textsuperscript{19} should perhaps be considered for such patients.

Section 1: Introduction

Section 2: Methods

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References

Figure 1. Identification of blood DCs for quantification and expression of HIV-1 receptor and coreceptor molecules. DCs were identified by FACS analysis on the basis of their lack of labeling for the mononuclear cell markers CD3, CD14, CD16, and CD19 but positive staining for HLA-DR. The myDCs were differentiated from the pDCs by their expression of CD11c (A). (B) Histograms show expression of CD4, CXCR4, and CCR5 on myDCs and pDCs. In each plot, the solid curve depicts labeling with an isotype control antibody and the open curve shows labeling with a specific antireceptor antibody.
References

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