Case Report

Delayed Recovery of Effector Memory CD4+ T Cells by Highly Active Antiretroviral Therapy in a Patient with HIV-1 Infection

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Department of Infectious and Respiratory Diseases, 1Department of Clinical and Laboratory Medicine, 2Department of Gastro-Intestinal Medicine, Tohoku University Graduate School of Medicine, Sendai 980-8574, and 3First Department of Internal Medicine, Ehime University School of Medicine, Ehime 791-0295

Okada, S., Kiruchi, M., Hasagawa, H., Ishikawa, M., Ohno, I., Kaku, M. and Hattori, T. Delayed Recovery of Effector Memory CD4+ T Cells by Highly Active Antiretroviral Therapy in a Patient with HIV-1 Infection. Tohoku J. Exp. Med., 2002, 196(3), 213-218 —— Effector memory T cells, which are potentially important in the protection against various pathogens, have been shown to be CCR7 negative. We report a case with HIV-1 infection in whom the change in the cell numbers of the subsets of CD4+ T cells, including CCR7 negative memory CD4+ T cells, in peripheral blood was monitored for more than one year after the initiation of highly active antiretroviral therapy. Percentage of each subsets in lymphocytes was measured by triple staining on lymphocyte fraction in flow-cytometry. The numbers of total CD4+ T cell, naive T cells, and CCR7 positive memory T cells successfully increased within half a month after the initiation of the therapy. Instead, the recovery of the cell number in CCR7 negative memory T cells delayed, and continued to increase until 10 months after the initiation of the therapy. It suggests the possibility of delay in recovery of immune systems after the initiation of the therapy in HIV-1-infected patients, despite the prompt recovery of total CD4+ T cells. —— HIV-1 infection; CCR7; HAART; effector memory T cells

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It is well established that CD4+ T cells are the targets of HIV-1 infection, and that the recovery of these cells by highly active antiretroviral therapy (HAART) reduces the possibilities of opportunistic infections. However, it remains to be clarified which subtypes of CD4+ T cells are most affected in HIV infection and how these subtypes are reconstituted.

Received November 12, 2001; revision accepted for publication March 1, 2002.
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by the therapy.

Circulating CD4+ T cells are mixed of populations at different functional status, and can be classified into two groups: CD45RA+ naive and CD45RO+ memory T cells. By determination chemokine receptors, memory T cells are further separated into groups having different destination and different roles. CCR7 on lymphocytes and mature dendritic cells is a receptor for CC chemokines secondary lymphoid tissue chemokine (SLC) and EBI1-ligand chemokine (ELC) constitutively expressed in the secondary lymphoid tissues, and is one of the important lymph node-homing receptors (von Andrian and Mackay 2000). We have recently made a mouse monoclonal antibody specific for CCR7 using synthetic peptides as antigen (Hasegawa et al. 2000). By staining this lymph node-homing receptor, CD45RO+ memory T cells are further divided into two groups, CCR7+CD45RO+ central memory (TCM) and CCR7-CD45RO+ effector memory (TEM) (Sallusto et al. 1999). Both naive and memory T cells have already been reported to be increased with HAART (Fleury et al. 2000; Vigano et al. 2000). In this study, we studied the changes in the numbers of further classified subsets of CD4+ T cells for one year in a patient with HIV infection who had newly started HAART, in order to identify the best parameter of the recovery of the immune systems in HIV-1 infection.

**CASE REPORT**

A 39-year-old, male patient was admitted to hospital in October, 1999, due to a severe Herpes Zoster infection in his left arm. He had fever of about 38°C for two weeks, did not respond to the normal regimen of oral administration of acyclovir on the day of admission. He received additional acyclovir for 9 days. At the diagnosis of HIV-1 infection, CD4+ T cell count was 203 cells/µl and HIV-1 RNA was 25,300 copies/ml, and fever continued more than three weeks. Serum HIV-1 RNA was measured by quantitative RT-PCR (Amplicore HIV-I monitor) at Mitsubishi Bio-chemical Laboratories, Inc. He had no sign of other opportunistic infections. From the middle of November, we started treatment with zidovudine (AZT), lamivudine, and nelfinavir. HIV-1 RNA in serum decreased to below 400 copies one week after the initiation of the therapy, and has been maintained at that level to date. Low grade fever continued for two more months after the initiation of medication. Mild reactivation of Herpes Zoster was observed on his left arm after the initiation of the therapy until April of this year, at which time it was almost cured leaving a small amount of desquamation. Nearly one year after the initiation of the antiretroviral therapy, the patient well tolerated the therapy without any signs of opportunistic infection, despite mild side effects such as macrocytic anemia, palpitation while receiving 600 mg of AZT daily. Most of the side effects improved after the dose of AZT was decreased to 400 mg per day.

At the initiation of therapy, we started monitoring the number of CD4+ T cell subset count in peripheral blood. Cell numbers were calculated by the multiplications of total white blood cell count, percentage of lymphocytes, CD4+ T cells, and percentages of each subset in CD4+ T cells, which were measured by FACS-Calibur (Becton Dickinson, Franklin Lakes, NJ, USA). FITC-conjugated mouse anti-CCR7 mAb2, PerCP-conjugated anti-CD4 (RPA-T4) or anti-CD8 (RPA-T8) mAbs (Bekton Dickinson), and phycoerythrin-conjugated anti-CD45RA(4KB5) or anti-CD45RO (UCHL1) mAbs (DAKO, Glostrup Denmark) were used for staining.

The number of CD4+ T cell counts increased rapidly after the initiation of the therapy, and the number reached a plateau within a few month (Fig. 1). TEM decreased from almost normal to a markedly low number one month after the initiation of the therapy, and
subsequently increased slowly, while the number of other subsets, naive and TCM, increased immediately after the initiation of the therapy (Fig. 2). TCM/naive, TEM/naive and TEM/TCM ratios were calculated based on Fig. 2. TEM/naive and TEM/TCM ratios decreased immediately from 2.43 to 0.05 and 4.80 to 0.05, respectively after the start of therapy, and then, both ratios had been increasing gradually to the last measurement (13 months after the initiation of the therapy). However, there was no significant changes in the TCM/naive ratio throughout the follow-up period.

The subsets in CD4+ T lymphocytes in five HIV-1 negative normal controls and three other HIV-1 infected hemophiliacs that are consisted of one long-term nonprogressor and two with progressive disease who have been infected with HIV-1 for more than fifteen years and have not been given any antiretroviral therapy were also measured (Table 1). In the patients with progressive HIV-1 infection (pt. 3 and 4 in Table 1, open squares in Fig. 2), numbers of TEM were very low, while the numbers of the other subsets were found to be normal. On the other hand, high number of TEM was found in the long-term nonprogressor (pt. 2 in Table 1, closed square in Fig. 2) had a than normal subjects, but the number of naive and
TABLE 1. Backgrounds of the patients with HIV-1 infection and normal controls in whom CD4+ T lymphocyte subset were measured

<table>
<thead>
<tr>
<th>HIV infection</th>
<th>Age</th>
<th>Sex</th>
<th>Background of infection</th>
<th>Duration after diagnosis</th>
<th>CD4 (/μl)</th>
<th>HIV RNA (copies/ml)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Pt. 1*</td>
<td>39</td>
<td>m</td>
<td>homosexual</td>
<td>1 week</td>
<td>203</td>
<td>25 300</td>
</tr>
<tr>
<td>Pt. 2*</td>
<td>30</td>
<td>m</td>
<td>hemophilia</td>
<td>&gt;15 years</td>
<td>618</td>
<td>&lt;400</td>
</tr>
<tr>
<td>Pt. 3</td>
<td>35</td>
<td>m</td>
<td>hemophilia</td>
<td>&gt;15 years</td>
<td>287</td>
<td>17 000</td>
</tr>
<tr>
<td>Pt. 4</td>
<td>31</td>
<td>m</td>
<td>hemophilia</td>
<td>&gt;15 years</td>
<td>339</td>
<td>5000</td>
</tr>
<tr>
<td>Normal Control</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Cont. 1</td>
<td>39</td>
<td>m</td>
<td></td>
<td></td>
<td>330</td>
<td></td>
</tr>
<tr>
<td>Cont. 2</td>
<td>30</td>
<td>m</td>
<td></td>
<td></td>
<td>643</td>
<td></td>
</tr>
<tr>
<td>Cont. 3</td>
<td>32</td>
<td>m</td>
<td></td>
<td></td>
<td>483</td>
<td></td>
</tr>
<tr>
<td>Cont. 4</td>
<td>34</td>
<td>m</td>
<td></td>
<td></td>
<td>307</td>
<td></td>
</tr>
<tr>
<td>Cont. 5</td>
<td>31</td>
<td>m</td>
<td></td>
<td></td>
<td>570</td>
<td></td>
</tr>
</tbody>
</table>

*a patient in the case report. The data is of the day of initiation of the therapy.  
*Long-term nonprogressor.  
Pt., patient; Cont., control subject; m, male.

TCM cell counts were comparable to those of normal subjects (open circles).

DISCUSSION

In our patient, the numbers of total CD4+ T cells, naive T cells and TCM started to increase immediately after the therapy as previously been reported (Fleury et al. 2000; Vigano et al. 2000). However, TEM count decreased promptly followed by a slow increase. The increase of TEM was apparently delayed compared to that in naive T cells or TCM.

The reason for the prompt decrease in TEM count after the initiation of the therapy in our patient is not clear. Recently, it was proposed that the division rate of naive T cells increased in HIV-1 infection, and which is then restored by antiretroviral therapy (Hazenberg et al. 2000). Others reported that the increase in turnover of CD4+ T cells in untreated HIV-1-infected patients is mainly depend on that of CD45RA-effector/memory T cells (McCune et al. 2000). Although it is difficult to speculate how T cell dynamics changed by the therapy in our patient, we calculated the ratios of TEM or TCM counts to naive T cell counts and found that TEM/naive ratio changed remarkably by the therapy and that of TCM/naive did not change. These findings indicate that TEM production probably from TCM or naive T cells was enhanced and/or that TEM may not be utilized in the tissues in untreated HIV-1-infected, and that HAART changed the dynamics followed by a slow increase in TEM count. Accordingly, TEM/naive and TEM/TCM ratios increased gradually throughout the follow-up period, although TCM/naive ratio have not changed. Chemokines SLC and ELC, adhesion molecules and antigen presentation at lymph nodes are involved in the differentiation fromCCR7 positive cells to TEM (von Andrian and Mackay 2000). Therefore, the slow increase of TEM may imply a slow recovery of the function of lymph nodes, which are reported to be one of the major targets of HIV (Pantaleo et al. 1991). Further studies are needed to identify which factor of lymph nodes is critical in this slow recovery.

Because it has been reported that HIV-1-specific activated CD4+ T cells were suppressed during the HIV-1 infection and were not be
recovered after HAART (McMichael and Rowland-Jones 2001), the increased TEM count after HAART may not directly control HIV-1 infection. However, it is clear that TEM is important in the immune responses against various pathogens exposed during HIV-1 infection; the highest TEM cell numbers in one long-term nonprogressor (Fig. 2) indicated the functional aspects of TEM in their protective role against progression of the disease.

We observed the reactivation of the Herpes Zoster infection, which lasted for around four months after the initiation of therapy. Various types of opportunistic infections have been reported to be reactivated after the initiation of HAART (DeSimone et al. 2000). It is generally said that reactivation of opportunistic infection is caused by the restoration of immune systems because the site is rich in inflammatory cells. However, it is not likely that the recurrence of herpetic bullous lesion observed in our patient was due to restored T cell recruitment to the site of inflammation. Some authors also have reported a discrepancy between the restoration in total CD4+ T cell count and pathogen-induced disease (Cinti et al. 2000). The temporary decrease in TEM after HAART could have induced the reactivation of Herpes Zoster infection. Furthermore, the slow recovery of this subset after therapy suggests slow reconstitution of the immune system and a delayed recovery from Herpes Zoster infection.

Finally, our observation suggests the possibility of the CCR7-CD45RO+CD4+T cell count as an indicator of immune status in HIV-1 infection, although further investigation on the function of CCR7-CD45RO+CD4+T cells in untreated and treated patients with HIV-1-infected patients is needed.

Acknowledgments

We thank Professor Kunio Shirato for continuous encouragement on the study. This work was supported by a Grant-in-Aid for Scientific Research Expenses for Health and Welfare Programs from the Ministry of Health and Welfare, Japan.

References


