Original Research Report

Elevated Serum Levels of RCAS 1 Are Associated with a Poor Recovery of the CD4+ T Cell Count after ART in HIV-1-infected Patients

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Objective: RCAS1 (receptor-binding cancer antigen expressed on SiSo cells) is an apoptosis-associated protein that induces apoptosis in activated T-cells. The aim of this study is to investigate the role of RCAS1 in HIV-1 infection.

Methods: We examined the serum levels of RCAS1 in HIV-1-infected patients at different clinical and immunological stages.

Results: Although the RCAS1 levels did not correlate with the clinical stage, they did correlate significantly with the CD8+ T cell numbers. Furthermore, the RCAS1 levels were also significantly higher in patients whose CD4+ T cell counts did not respond to antiretroviral therapy (ART) than in those who responded to ART.

Conclusions: The present findings therefore suggest that the RCAS1 level affects the CD4+ T cell counts in HIV-1-infected patients with ART.

Key words: RCAS1, HIV-1 infection, immunological prognosis


Introduction

RCAS1 has been demonstrated to be a type II membrane protein expressed on human tumor cells. RCAS1 can also be secreted, and both the secreted and transmembrane forms act as a ligand thereby inducing apoptosis in receptor-positive cells including T cells, B cells and NK cells1). These results suggest that RCAS1 may assist tumor cells in their survival or escape from immunosurveillance2). Several reports have shown the serum RCAS1 level to be a significant prognostic factor in patients with certain malignancies3-5). In addition, monocytes/macrophages have also been shown to express RCAS1, and soluble RCAS1 molecules have been detected in culture supernatants of lipopolysaccharide-stimulated macrophages6). Furthermore, an association between RCAS1 and Epstein-Barr virus infection has also been reported7). In the present study, to elucidate whether RCAS1 plays a role in HIV-1 infection, we examined the serum levels of RCAS1 in patients during various clinical and immunological stages of HIV-1 infection.

Methods

The subjects consisted of 82 HIV-1-infected patients and 12 HIV-1-seronegative healthy controls. The characteristics of these patients and controls are shown in Table 1. None of the HIV-1-positive patients exhibited any signs of acute HIV-1 infection. Informed consent for blood sampling was obtained. The study was conducted according to the ethical guidelines of our hospital, and was approved by an authorized representative of the hospital. We first measured the levels of sRCAS1, sFasL and sTRAIL in the 82 HIV-infected patients at two different clinical stages (AIDS vs. non-AIDS) and at two different immunological statuses (>800 CD8+ T cells/µl vs. <800 CD8+ T cells/µl). The serum levels of RCAS1, FasL and TRAIL were measured using specific ELISA kits (RCAS1, Medical and Biological Laboratories Co. Nagoya, Japan; sFasL and...
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Table 1 Serum levels of RCAS1 of the subjects

<table>
<thead>
<tr>
<th></th>
<th>No. of cases</th>
<th>Age</th>
<th>sRCAS1</th>
</tr>
</thead>
<tbody>
<tr>
<td>Healthy Control</td>
<td>12</td>
<td>32.5±7.0</td>
<td>6.81±1.87</td>
</tr>
<tr>
<td>HIV infected Pt.</td>
<td>82</td>
<td>38.6±11.1</td>
<td>6.23±3.31</td>
</tr>
<tr>
<td>ART naïve</td>
<td>41</td>
<td>37.0±10.6</td>
<td>5.91±3.11</td>
</tr>
<tr>
<td>CD4-responder</td>
<td>26</td>
<td>36.6±10.8</td>
<td>5.34±2.16</td>
</tr>
<tr>
<td>CD4-non-responder</td>
<td>15</td>
<td>41.8±11.1</td>
<td>8.72±4.29*</td>
</tr>
</tbody>
</table>

*p<0.01 for the difference between CD4-responder and CD4-non-responder

Table 2 Characteristics of patient with ART

<table>
<thead>
<tr>
<th></th>
<th>CD4-responder</th>
<th>Cd4-non-responder</th>
<th>p-value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Age</td>
<td>37.4±10.4</td>
<td>42.9±10.1</td>
<td>0.1106</td>
</tr>
<tr>
<td>CD4+ T cells counts before ART (μl)</td>
<td>189.3±132.6</td>
<td>154.2±125.5</td>
<td>0.4209</td>
</tr>
<tr>
<td>duration of ART (years)</td>
<td>4.88±2.38</td>
<td>4.58±4.52</td>
<td>0.7844</td>
</tr>
</tbody>
</table>

sTRAIL, DIACLONE Research Co. Besancon, France) according to the manufacturer’s recommendations. We used Student’s t test for comparisons between the two groups.

**Results**

The levels of sRCAS1 were not significantly different between the AIDS group and the non-AIDS group (AIDS vs. non-AIDS; 6.42±3.18 vs. 5.72±3.65). Levels of sRCAS1 were significantly higher in the patients with >800/μl CD8+ T cells than in the patients with <800/μl CD8+ T cells (CD6 <800/μl vs. CD8 ≥ 800/μl; 7.33±3.77 vs. 4.96±2.06). There were no significant correlations between the serum levels of sFasL, sTRAIL and sRCAS1 (data not shown). We next measured the levels of sRCAS1 in 41 ART-naïve HIV-1-infected patients. There was no significant difference in the levels of RCAS1 between ART-naïve HIV-1-infected patients and the controls (Table 1). We next examined 41 HIV-infected patients who had undetectable levels of plasma HIV-1 RNA after they were treated with ART. To evaluate the effects of ART on the recovery of the CD4+ T cell count in the peripheral blood, we divided these 41 patients into two groups according to their CD4+ T cell counts: CD4 responders (n=26), CD4+ T cells increased by more than 300/μl after the initiation of ART; CD4 non-responders (n=15), CD4+ T cells increased by less than 300/μl after the initiation ART. There were no statistically significant differences in age, the CD4+ T cell counts before ART, or the duration of ART between the responders and non-responders (Table 2). As shown in Table 1, the serum levels of RCAS1 were significantly higher in the CD4 non-responders.

**Discussion**

In HIV-1 infection, infected CD4+ T cells undergo apoptotic cell death. In addition, a significant number of uninfected CD4+ T cells in HIV-1-infected patients undergo apoptosis, induced either by immunological activation, by the effects of HIV-1 proteins, or by elevated levels of death-inducing ligands. These apoptotic mechanisms contribute to an impairment of the immune system in HIV-1-infected patients. It has recently been shown that FasL and TRAIL contribute to the apoptosis of HIV-1-uninfected CD4+ T cells, and that the serum levels of sFas and sTRAIL correlate with the clinical, immunological and virological status8,9). The present results are thus consistent with these previous findings.

RCAS1 was originally discovered as a tumor-associated antigen, which induces apoptosis in RCAS1 receptor-positive immune-regulating cells, such as activated T cells, thereby helping such tumors escape immune surveillance.

Although we could not examine the molecular mechanisms of RCAS1 and the expression of RCAS1 receptor on HIV-1 infected CD4+ T cells because of the unavailability of RCAS1, the expression of RCAS1 receptor has already been reported to increase when T cells are activated11. Given the fact that an HIV-1 infection is characterized by a state of chronic T cell activation10, it is therefore very possible that the expression of RCAS1 receptor increases in HIV-1 infected CD...
Furthermore, the patients with high levels of RCAS1 showed a low CD4+ T cell recovery in response to ART. This result shows that RCAS1 induces apoptosis in CD4+ T cells, which are either HIV-1-infected cells or uninfected cells, even after ART has sufficiently suppressed HIV-1 production. Although the mechanisms of regulating RCAS1 and RCAS1 receptors in HIV-1 infection also remain unclear, the findings of a lack of any correlation between the serum levels of sFasL, sTRAIL and RCAS1 suggest the expression of RCAS1 to be regulated by a mechanism that is different from the FasL and TRAIL systems. Although our findings were not able to elucidate whether FasL, TRAIL, or RCAS1 plays a more important role in HIV-1 infection, RCAS1 was suggested to play a role as one of the mechanisms not only inducing the apoptosis of CD4+ T cells in HIV-1-infected patients, but also causing a progression of the disease in HIV-1 infection.

In future studies, we plan to examine the precise mechanisms that regulate the RCAS1/RCAS1 receptor system in HIV-1.

Acknowledgment: This study was partly supported by Health Science Research Grants on HIV/AIDS from Ministry of Health, Labor and Welfare of the Japanese Government. The sponsor had no role in the study design, data collection, data analysis, data interpretation, or writing of this report.

References


