The Presence of HTLV-I Proviral DNA in the Central Nervous System of Patients with HTLV-I-Associated Myelopathy/Tropical Spastic Paraparesis

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Abstract

Human T-lymphotropic virus type 1 (HTLV-I) is a pathogenic retrovirus associated with a chronic progressive myelopathy, termed HTLV-I-associated myelopathy (HAM)/tropical spastic paraparesis (TSP), as well as adult T-cell leukemia (ATL). A chronic inflammatory process has been implicated in HAM/TSP by a pathological study, but the exact mechanism still remains unknown. To understand better the complex mechanism of disease induction by HTLV-I, I studied the spreading pattern of HTLV-I in both peripheral blood mononuclear cells (PBMNCs) and central nervous system (CNS) tissues in patients with HAM/TSP using a quantitative polymerase chain reaction (PCR) method. My results indicated the primary event to be the efficient replication of HTLV-I in vivo, whereas HTLV-I is likely to be present in the constituent cells of the CNS in addition to the infiltrating mononuclear cells.

Index Entries: Human T-lymphotropic virus type 1 (HTLV-I); HTLV-I-associated myelopathy; tropical spastic paraparesis; polymerase chain reaction; central nervous system.

Introduction

HTLV-I is an etiologic agent of HAM/TSP (1,2), and its mechanism is presently undergoing intensive research. Pathological studies on the CNS tissue of HAM/TSP patients has demonstrated perivascular lymphocyte infiltration as well as the loss of myelin and axon (3). Thus, it appears that a chronic inflammatory process is operative. However, the exact mechanism of CNS damage remains unclear. The following mechanisms are thought to be possible:

1. The presence of an immunologic aberrance induced by the HTLV-I gene products may cause an autoimmune response against some CNS antigens;
2. The existence of cytotoxic T-lymphocytes (CTL) specific for HTLV-I may destroy the constituent cells of the CNS infected with HTLV-I; or
3. HTLV-I invading into the CNS may directly exert a destructive effect on the CNS cells.

The immunologic abnormalities frequently found in HAM/TSP patients, i.e., spontaneous lymphoproliferation (4), increased activated T-lymphocytes in both the peripheral blood and cerebrospinal fluid (CSF) (5), high levels of interleukin 2 (IL-2) and IL-6 in the sera and CSF (6,7), and the presence of oligoclonal bands in the CSF (2), lend support to the autoimmune hypothesis. However, these features are not specific for HAM/TSP alone and some of them are also present in asymptomatic carriers to a lesser extent.

On the other hand, HTLV-I can also infect the wide spectrum of target cells in vitro, including microglia, oligodendrocytes, and astrocytes (8,9). Therefore, a high level of CTL against HTLV-I in CSF (10,11) may be directed to these CNS cells infected with HTLV-I. Alternatively, HTLV-I may directly destroy these CNS cells in vivo, as observed in the human immunodeficiency virus (HIV) (12).

In either case, it is of critical importance to determine whether or not the HTLV-I genome is present in the constituent cells of the CNS. Up to now, neither conventional immunohistochemical methods nor in situ hybridization unambiguously showed the presence of HTLV-I antigens or genome in the HAM/TSP CNS. This indicates that the amount of the HTLV-I genome in HAM/TSP CNS, if any, is extremely low. Therefore, I used a quantitative PCR method to evaluate the amount of HTLV-I proviral DNA present in the CNS tissue as well as in the PBMNCs of HAM/TSP patients (13,14). The following sections summarize the results of my quantitative PCR studies in order to provide new insights into the mechanism of HAM/TSP.

Analysis of HTLV-I Proviral DNA Amounts in Peripheral Blood Mononuclear Cells

Using the quantitative PCR method, I compared the amounts of HTLV-I proviral DNA in the PBMNCs from patients with HAM/TSP, HTLV-I carriers without HAM/TSP, and HTLV-I seronegative control subjects (13). All subjects were residents of the southwestern part of Japan, which is an endemic area for HTLV-I infection (15). At the same time, antibody titers to recombinant HTLV-I p40\textsuperscript{tax} protein and the gag-env hybrid protein in the sera were measured by an enzyme-linked immunosorbent assay (ELISA) (16). The following results were obtained.

1. The amounts of HTLV-I proviral DNA in the PBMNCs were 10- to 100-fold higher in the HAM/TSP patients than in the carriers without HAM/TSP and without any inflammatory diseases (Fig. 1).
2. The patients with an early onset (15–39 yr, mean = 32.8 yr, eight females and four males), as compared to those with a late onset of the disease (44–61 yr, mean = 53.5 yr, six females), had significantly higher amounts of HTLV-I proviral DNA, particularly in the early stages of the disease (Fig. 2).
3. The carriers without HAM/TSP, but with other inflammatory diseases, i.e., polymyositis, optic neuritis, or bronchoalveolitis, also had a high level of HTLV-I proviral DNA (Fig. 1).
4. The HTLV-I proviral DNA amounts correlated significantly with both the IgG and IgA antibody titers to the recombinant HTLV-I proteins. The HAM/TSP patients had significantly higher titers of the IgG and IgA antibodies to the HTLV-I proteins than did the HTLV-I carriers without HAM/TSP.
5. HAM/TSP patients, having both a high HTLV-I proviral DNA load and high titers of the IgG and IgA antibodies, frequently demonstrated IgM antibodies to the HTLV-I proteins. The IgM antibodies to the HTLV-I proteins were detected more frequently in the HAM/TSP patients than in the carriers without HAM/TSP (40 vs 6%).

These results strongly suggest that the large increase in the HTLV-I proviral DNA is associated with the development of HAM/TSP. I and others estimated that approx 5-50% of the PBMNCs harbor the HTLV-I genome in HAM/TSP (13,18). The provirus was shown to be integrated polyclonally into host DNA in PBMNCs (19). The vigorous replication of HTLV-I may well explain the extremely high titer antibodies as well as the strong CTL against the virus in HAM/TSP patients. Moreover, the spreading of HTLV-I can induce extensive alteration in T-cell function, since the viral tax protein enhances the expression of specific cellular genes, such as IL-2, IL-2 receptor, and c-fos protooncogene (20,21). Therefore, such a large proviral DNA load is likely to predispose to inflammatory conditions through the widespread T-cell activation. Alternatively, it is possible that the activation...