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Case Report

Accumulation of human T-lymphotropic virus type I (HTLV-I)–infected cells in the cerebrospinal fluid during the exacerbation of HTLV-I–associated myelopathy

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Human T-lymphotropic virus type I (HTLV-I)-associated myelopathy/tropical spastic paraparesis (HAM/TSP) is a slowly progressive, inflammatory disease of the central nervous system (CNS). We report a patient with transverse myelitis, who exhibited acute onset and rapid progression of the disease and whose symptoms resembled those observed in multiple sclerosis with spinal cord presentation. During neurological exacerbation of the condition, the HTLV-I proviral load in the cerebrospinal fluid (CSF) increased to 10 times that in the peripheral blood. This suggests that the accumulation of HTLV-I-infected cells in the CNS contributes to neurological exacerbation. Based on the increased proviral load in the CSF, we diagnosed the disease as acute progressive HAM/TSP. The measurement of the HTLV-I proviral load in the CSF is useful for the diagnosis of HAM/TSP and for monitoring its progression. *Journal of NeuroVirology* (2008) **14,** 459–463.

Keywords: cerebrospinal fluid; HTLV-I–associated myelopathy; human T-lymphotropic virus type I; multiple sclerosis; viral load

Introduction

Human T-lymphotropic virus type I (HTLV-I)–associated myelopathy/tropical spastic paraparesis (HAM/TSP) is a chronic inflammatory disease of the central nervous system (CNS) caused by HTLV-I infection (Osame *et al*, 1987). The incidence of HAM/TSP in HTLV-I–infected individuals is less than 1% (Kaplan *et al*, 1990), and the male-female ratio of affected individuals is approximately 1:2 (Nakagawa *et al*, 1995). These patients exhibit spastic gait, bilateral hypesthesia of the lower limbs, and sphincter dysfunction. Further, HAM/TSP patients exhibit high HTLV-I proviral loads and increased HTLV-I–specific cytotoxic T-lymphocyte (CTL) responses as compared to asymptomatic HTLV-I carriers (Jacobson *et al*, 1990; Nagai *et al*, 1998). Perivascular lymphocytic infiltration of the spinal cord is a histopathological hallmark of this disease (Umehara *et al*, 1993). HTLV-I preferentially infects CD4+ lymphocytes, and these infected CD4+ lymphocytes can be detected in the CNS (Kubota *et al*, 1994; Moritoyo *et al*, 1996). This suggests that the migration of HTLV-I–infected cells into the CNS is an important event that characterizes the development of HAM/TSP. Here, we report a case of acute onset, rapid progression, and repeated

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exacerbation of the disease. These features made it difficult to distinguish this disease from possible relapsing-remitting multiple sclerosis (MS) with spinal cord presentation. We finally diagnosed the condition as HAM/TSP because we detected the accumulation of HTLV-I–infected cells in the CNS during neurological exacerbation of the symptoms.

Case report

In March 2000 a 56-year-old woman experienced a cold sensation in both feet. This sensation gradually developed into numbness and spread upward to her thigh and abdomen. Symptoms of sphincter dysfunction and spastic gait appeared within a few days. The subject experienced difficulties in standing and walking 1 week later, and the use of a walking cane was warranted 2 weeks later. Her serum samples tested positive for anti-HTLV-I antibody, and she was admitted in our hospital in June. The patient had never undergone blood transfusion. The neurological findings were as follows: moderate weakness of the lower limb muscles, bilateral spasticity of the lower limbs, increased deep-tendon reflexes in the upper and lower limbs, bilaterally positive Babinski and Chaddock reflexes, reduced superficial sensation below the midthoracic level, and absence of vibratory sensation in the lower limbs. Urinary dysfunction and constipation were evident. The white blood cell count and the findings of general biochemical examinations were normal, except for the presence of hypercholesterolemia. Anti-nuclear, ribonucleoprotein, SS-A, and SS-B antibodies and *Treponema pallidum* hemagglutination were negative. The serum levels of folic acid and angiotensin-converting enzyme were normal, and the serum tested positive for anti-HTLV-I antibody. The HTLV-I proviral load in the peripheral blood mononuclear cells (PBMCs) was 47 copies/10^4 cells, as was determined by performing a quantitative polymerase chain reaction (Nagai *et al*, 1998). In the cerebrospinal fluid (CSF), the protein content was 73.2 mg/dl; the cell count, 46 cells/mm^3 (mononuclear cells, 96%; polynuclear cells, 4%); the immunoglobulin G (IgG) index, 1.71 (Table 1). The titer of anti-HTLV-I antibody in the CSF was 1024 ×, as determined by the particle agglutination method, and the neopterin concentration was 134 ng/ml. Oligoclonal band was positive, and myelin basic protein was negative in the CSF. The patient's condition was characterized by acute onset, rapid progression, and symptoms that were suggestive of transverse myelitis; these features are uncommon in HAM/TSP. However, both the peripheral blood and CSF tested positive for anti-HTLV-I antibodies. Although HAM/TSP was likely to be the accurate diagnosis, we could not entirely exclude the possibility of MS with spinal cord presentation complicated with HTLV-I infection (McDonald *et al*, 2001). The patient was intravenously administered a high dose of methylprednisolone, followed by oral prednisolone. With this treatment, the laboratory findings improved in the following manner: the HTLV-I proviral load in the PBMCs reduced to 16 copies/10^4 cells; the cell count in the CSF, to 7 cells/mm^3 (mononuclear cells, 100%); the IgG index, to 1.48 (Table 1). However, the patient's neurological symptoms showed no improvement.

In April 2001, the patient developed weakness of the lower limbs and was unable to stand without support. Causalgia of the lower limbs worsened, and the patient experienced a feeling of tightness in the thoracic region. The subject was admitted to our hospital in May for these symptoms. Electrophysiological examination revealed that the auditory brainstem response and visual evoked potential were normal. The somatosensory evoked potential was abnormal in the lower limbs, wherein P40 was not detected. Magnetic resonance imaging (MRI) revealed no abnormalities in the brain and lumbar spinal cord. T2-weighted images of the cervical MRI showed a longitudinal high-intensity area in the sagittal section, and increased intensity in the posterior spinal cord in the axial section (Figure 1). Methylprednisolone pulse therapy was initiated, followed by oral prednisolone and cyclophosphamide pulse therapy; however, the

Table 1 HTLV-I proviral loads in the peripheral blood and cerebrospinal fluid.

	Peripheral blood		Cerebrospinal fluid					
Date	HTLV-I proviral load (copies/10^4 cells)	Anti-HTLV-I Ab titer* (×)	HTLV-I proviral load (copies/10^4 cells)	Cell count (cells/mm^3)	Protein content (mg/ml)	Neopterin concentration (ng/ml)	Anti-HTLV-I Ab titer* (×)	Ratio of HTLV-I proviral load**
Jun/30/2000	47	Positive	NE	46	73.2	134	1024	NE
Sep/9/2000	16	NE	NE	7	NE	NE	NE	NE
Aug/1/2001	21	16384	123	4	42.8	NE	1024	5.9
Jun/4/2004	129	>131072	1355	5	58.8	31	1024	10.5
Jun/30/2004	54	65536	1399	7	50.7	15	512	25.9
Apr/5/2005	127	32768	666	2	53.9	35	512	5.2

*Anti-HTLV-I antibody (Ab) titer as determined by the particle agglutination method. **Ratio of the HTLV-I proviral load in the cerebrospinal fluid to that in the peripheral blood. NE: not examined.

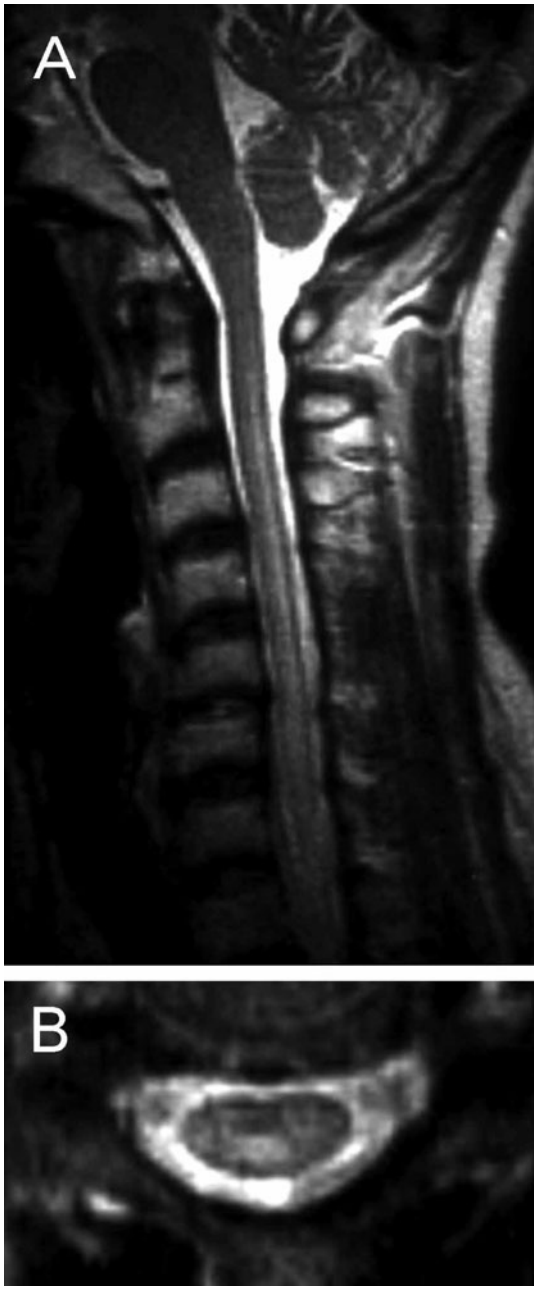

Figure 1 MRI of the cervical cord showing increased intensity in the posterior region. (**A**) T2-weighted image, sagittal section; (**B**) T2-weighted image, transverse section.

neurological findings did not improve. The HTLV-I proviral loads in the PBMCs and CSF were 21 and 123 copies/10^4 cells, respectively (Table 1), with the load in the CSF being 5.9 times that in the PBMCs. We diagnosed the patient's condition as acute progressive HAM/TSP because we detected the accumulation of HTLV-I–infected cells in the CNS.

In June 2004, the patient once again experienced exacerbation of the tight feeling in the thoracic region. The HTLV-I proviral load increased to 129 copies/10^4 cells in the PBMCs and 1355 copies/10^4 cells in the CSF; these values were 6.1 and 11 times the corresponding values prior to the exacerbation, respectively. Further, the HTLV-I proviral load in the CSF was 10.5 times higher than that in the PBMCs (Table 1). Methylprednisolone pulse therapy followed by oral prednisolone therapy ameliorated the feeling of tightness in the thoracic region. At 7 days after the therapy, the neopterin concentration in the CSF decreased. Further, the HTLV-I proviral load in the PBMCs decreased to 54 copies/10^4 cells; i.e., it became 50% less than its value before the therapy (Table 1). In contrast, the load in the CSF remained almost constant at 1399 copies/10^4 cells. Consequently, the ratio of the proviral load in the CSF to that in the peripheral blood increased to 25.9.

In April 2005, during follow-up, the HTLV-I proviral loads in the PBMCs and CSF were 127 and 666 copies/10^4 cells, respectively (Table 1). Thus, the ratio of the load in the CSF to that in the PBMCs reduced to 5.2.

Discussion

An increase in the HTLV-I proviral load is related to an increased risk for the development HAM/TSP and the progression of motor disabilities (Matsuzaki *et al*, 2001). In particular, when the HTLV-I proviral load in the PBMCs increases to more than 100 copies/10^4 cells, the risk for developing HAM/TSP increases exponentially (Nagai *et al*, 1998). In addition, an increase in the proviral load in the peripheral blood parallels neurological deterioration in HAM/TSP patients (Takenouchi *et al*, 2003). In our case, the exacerbation of neurological symptoms was associated with a marked increase in the HTLV-I proviral load not only in the PBMCs but also in the CSF. Furthermore, the proviral load in the CSF attained a value that was 10.5 times that in the PBMCs. These findings suggest that the accumulation of HTLV-I–infected cells in the CNS is related to neurological deterioration of the symptoms. With regard to this accumulation of infected cells, two possibilities can be considered—increased recruitment of cells from the peripheral blood and expansion in the area of the CNS. In our case, the increase in the proviral load in the CSF paralleled that in the peripheral blood, suggesting that the accumulation of HTLV-I–infected cells in the CNS might have been due to increased recruitment of peripheral cells. However, the two above mentioned possibilities are not mutually exclusive.

It is important to bring about a reduction in the HTLV-I proviral load in order to prevent the onset and progression of HAM/TSP. Here, we demonstrated that the HTLV-I proviral load in peripheral blood reduced shortly after corticosteroid therapy

was initiated (within 1 week). However, the load in the CSF remained unchanged after the therapy was initiated (Table 1). These results suggest that the corticosteroid therapy was effective for reducing the HTLV-I proviral load in the peripheral blood but not in the CNS. The observed decrease in the CSF proviral load (666 copies/10^4 cells) 9 months after the therapy may have been caused by reduced recruitment of infected cells from the peripheral blood.

We have previously reported cases of HAM/TSP patients wherein the HTLV-I proviral load decreased following corticosteroid therapy (Takenouchi *et al*, 2003). The reason for this effect of corticosteroid therapy is unclear. HAM/TSP patients exhibit an increased number of activated cells among the PBMCs (Itoyama *et al*, 1989), and these may include HTLV-I–infected cells. Corticosteroids induce apoptosis in activated T cells (Lanza *et al*, 1996). Thus, it is possible that therapy with these drugs contributes to reducing the HTLV-I proviral load via the induction of apoptosis in activated HTLV-I–infected cells. However, further investigations are required to test this possibility.

Our patient's condition was characterized by acute onset, rapid progression, and repeated exacerbation of the neurological symptoms. HAM/TSP is usually a chronic disease that exhibits slow progression (Osame *et al*, 1987). Therefore, it was difficult to distinguish this disease from possible MS with spinal cord presentation in our patient. Although both the PBMCs and CSF tested positive for anti-HTLV-I antibodies, which is one of the most important criteria for the diagnosis of HAM/TSP (Osame *et al*, 1987), we could not entirely exclude the possibility of MS with coincidental HTLV-I infection at the first administration. Recently, Puccioni-Sohler *et al* reported that, in cases of HTLV-I infection wherein the diagnostic criteria of both HAM/TSP and MS are fulfilled, it is possible to differentiate the two diseases based on whether the HTLV-I proviral loads are high in both the PBMCs and the CSF (Puccioni-Sohler *et al*, 2007). In our patient, the proviral load in the PBMCs was low. Although most HAM/TSP patients exhibit high proviral loads, some exhibit low loads (Nagai *et al*, 1998). In addition, the ratio of the HTLV-I proviral load in the CSF to that in the PBMCs has been reported as a useful parameter to be considered for the diagnosis of HAM/TSP (Lezin *et al*, 2005; Puccioni-Sohler *et al*, 2003; Takenouchi *et al*, 2003). In our case, the ratio was high at a value of 5.2 to 25.9, and it increased with neurological deterioration, i.e., when the proviral load in the CNS increased from 5.9 to 10.5 times that in the PBMCs (Table 1). These results suggest that HTLV-I infection induced inflammation in the CNS, resulting in the appearance of important features that enabled us to diagnose the condition as acute progressive HAM/TSP and not MS.

Thus, it is important to measure the HTLV-I proviral load not only in the PBMCs but also in the CSF in order to distinguish HAM/TSP from other myelopathies such as MS. Further, the measurement of these proviral loads is useful for monitoring the progression of HAM/TSP.

Declaration of interest: The authors report no conflicts of interest. The authors alone are responsible for the content and writing of the paper.

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