Mini Review

Immunogenetics of human T-cell leukemia virus type 1 (HTLV-1)-associated myelopathy/tropical spastic paraparesis (HAM/TSP)

Mineki Saito

Department of Immunology, Graduate School of Medicine, University of the Ryukyus, Okinawa, Japan

Human T-cell leukemia virus type 1 (HTLV-1) is a replication-competent human retrovirus associated with two distinct types of disease: the malignancy known as adult T-cell leukemia (ATL) and a chronic inflammatory central nervous system disease HTLV-1-associated myelopathy/tropical spastic paraparesis (HAM/TSP), whereas the vast majority of infected individuals remain asymptomatic carriers of the virus in lifetime. It is not yet fully understood why do certain individuals develop ATL or HAM/TSP, and how does HTLV-1 persist in spite of host immune response. This review focuses on the complex virus-host interactions and the cellular immune responses to HTLV-1 infection seen in HAM/TSP patients, which are important factors in determining HTLV-1 proviral load and the risk of developing disease.

Rec.1/15/2009, Acc.2/5/2009, pp310-316

Correspondence should be addressed to:
Mineki Saito, Department of Immunology, Graduate School of Medicine, University of the Ryukyus, Uehara 207, Nishihara-cho, Nakagami-gun, Okinawa 903-0215, Japan. Phone: +81-98-895-1200, Fax: +81-98-895-1200, e-mail: mineki@med.u-ryukyu.ac.jp

Key words HTLV-1, HAM/TSP, HLA, immune response, disease susceptibility

Introduction

Human T-cell leukemia virus type 1 (HTLV-1) infection is of particular interest to the field of immunology as well as microbiology because HTLV-1 is never be eliminated from the host in spite of a vigorous cellular and humoral immune responses against the virus, but causes no disease in vast majority of infected subjects (asymptomatic carriers: AC). Since HTLV-1 infection causes two distinct intractable diseases without effective treatment known as adult T-cell leukaemia[1,2] and HTLV-1-associated myelopathy/tropical spastic paraparesis (HAM/TSP)[3,4], evaluation of the individual risk for developing diseases in each AC would certainly be of considerable importance especially in HTLV-1 endemic area such as southern Japan, the Caribbean, Central and South America, the Middle East, Melanesia, and equatorial regions of Africa[5]. HAM/TSP is a chronic progressive myelopathy characterized by spastic paraparesis, sphincter dysfunction and mild sensory disturbance in the lower extremities, and the main pathological features are chronic inflamma-

tion in the spinal cord characterized by perivascular lymphocytic cuffing and parenchymal lymphocytic infiltration. It is therefore widely assumed that the immune response against HTLV-1 causes the inflammatory spinal cord damage seen in HAM/TSP patients[6]. In this review, I shall summarize the recent work for HAM/TSP attempting to resolve outstanding question, i.e. why do some HTLV-1-infected people develop disease whereas the vast majority remains healthy in lifetime.

HTLV-1 infection and clinical features of HAM/TSP

HTLV-1 is classified as a complex retrovirus in the genus *Deltaretrovirus* of the subfamily *Orthoretrovirinae*, and infects 10-20 million people worldwide[7]. HTLV-1 can be transmitted through sexual contact[8], injection drug use[9], and breastfeeding from mother to child[10,11]. Although HTLV-1 infection is associated with a range of non-malignant chronic inflammatory diseases in the eyes, the lungs, or the skeletal muscles[7], HAM/TSP is the best-recognized with chronic progressive myelopathy characterized by spastic paraparesis, sphincter dysfunction and mild sensory disturbance in the lower extremities[12]. Pathological analysis of HAM/TSP autopsy materials indicates that the disease affects the spinal cord, predominantly at the thoracic level[13-15]. Loss of myelin and axons in the lateral, anterior, and posterior columns is associated with perivascular and parenchymal lymphocytic infiltration with the presence of foamy macrophages, proliferation of astrocytes, and fibrillary gliosis. Clinical progression of HAM/TSP is associated with increased proviral load in individual patients, and the ratio of proviral loads in cerebrospinal fluid (CSF) cells/in peripheral blood mononuclear cells (PBMCs) is significantly associated with clinically progressive disease[16]. MHC class I tetramer analysis of lymphocytes isolated from the CSF of HAM/TSP patients showed even higher frequencies of HTLV-1 Tax11-19-specific, HLA-A*02-restricted CD8 lymphocytes compared to PBMCs[17]. Therefore, an increased proliferation or migration of HTLV-1-infected and/or HTLV-1 specific lymphocytes to the central nervous system (CNS) might be closely associated with HAM/TSP pathogenesis[18]. The presence of atypical lymphocytes (so-called "flower-cells") in peripheral blood and CSF, a moderate pleocytosis and raised protein content in CSF are typically found in HAM/TSP patients. Oligoclonal bands, raised concentrations of inflammatory markers such as neopterin, tumor necrosis factor (TNF)-α, interleukin (IL)-6 and interferon (IFN)-γ, and an increased intrathecal antibody synthesis specific for HTLV-1 antigens have also been described[19].

Risk factors for HAM/TSP

Previous population association study of 202 cases of HAM/TSP and 243 AC in Kagoshima, HTLV-1 endemic southern Japan, revealed that one of the major risk factors is the HTLV-1 proviral load. The median proviral load was more than ten times higher in HAM/TSP patients than in AC, and a high proviral load was also associated with an increased risk of progression to disease[20]. It was suggested that genetic factors such as HLA is related to the high proviral load in HAM/TSP patients and genetic relatives. Namely, possession of the HLA-class I genes HLA-A*02 and Cw*08 was associated with a statistically significant reduction in both HTLV-1 proviral load and the risk of HAM/TSP, whereas possession of HLA-class I HLA-B*5401 and class II HLA-DRB1*0101 predispose to HAM/TSP in the same population[21,22]. Since the function of class I HLA proteins is to present antigenic peptides to cytotoxic T lymphocytes (CTL), these results imply that individuals with HLA-A*02 or HLA-Cw*08 mount a particularly efficient CTL response against HTLV-1, which may be an important determinant of HTLV-1 proviral load and the risk of HAM/TSP. Further analysis to look at non-HLA host genetic factors revealed that non-HLA gene polymorphism also affects the risk for developing HAM/TSP. For example, the TNF-α promoter -863 A allele[23] and the longer CA repeat alleles of MMP-9 promoter[24] predisposed to HAM/TSP, whereas IL-10 -592 A[25], SDF-1 +801A[23] and IL-15 +191 C alleles[23] conferred protection against HAM/TSP. The polymorphisms of MMP-9 and IL-10 promoter each linked to the HTLV-1-encoded transactivator Tax mediated transcriptional activity of each gene[24,25].

Meanwhile, although most studies of HTLV-1 genotype have reported no association between variants of HTLV-1 and the risk of HAM/TSP, Furukawa et al reported the association between HTLV-1 *tax* gene variation and the risk of HAM/TSP[26]. The *tax* subgroup A that belongs to cosmopolitan subtype A was more frequently observed in HAM/TSP patients and this effect was independent of protective allele HLA-A*02. HLA-A*02 appeared to give protection against only one of the two prevalent sequence variants of HTLV-1, *tax* subgroup B that belongs to cosmopolitan subtype B, but not against *tax* subgroup A in Japanese population[26]. Interestingly, HLA-A*02 appears not to give protection against infection with cosmopolitan subtype A in a population in Iran[27]. These findings suggest that both host genetic factors and HTLV-1 subgroup play a part in determining the risk of HAM/TSP, although the effect of HTLV-1 genotype is relatively small so the factors that determine the different outcomes of HTLV-1 infection must lie chiefly in the host.

Estimation of the odds for developing HAM/TSP

Based on these observations, a best-fit logistic regression equation that can be used to predict the odds of HAM/TSP has been developed[23]. Using this equation, knowledge of HTLV-1-infected individuals' ages, sex, provirus load, HTLV-1 *tax* subgroup, and genotypes at the loci HLA-A (HLA-A*02), HLA-C (HLA-Cw*08), stromal cell-derived factor (SDF)-1 (+801G/A), and TNF-α (-863A/C) allowed for the correct identification of 88% cases of HAM/TSP in Kagoshima cohort. To validate whether this multivariate logistic equation can be useful to identify HAM/TSP related symptom in AC, the individual odds of 181 consecutive AC were calculated and compared with their clinical parameters and laboratory findings[28]. Interestingly, although no clear difference was seen between the odds of HAM/TSP and either sex, family history of HAM/TSP or ATL, and history of blood transfusion, however, brisk patellar deep tendon reflexes, which suggest latent central nervous system compromise, and flower cell-like abnormal lymphocytes, which is the morphological characteristic of ATL cells, has found to be associated with a higher odds of HAM/TSP. These observations indicated that this best-fit logistic regression equation may be useful for detecting subclinical abnormalities in AC in Kagoshima, where HTLV-1 endemic southern Japan.

The immune response to HTLV-1

1) The humoral immune response to HTLV-1

In HTLV-1 infection, anti-HTLV-1 antibody that often includes IgM is detected in all infected individuals, either AC or patients with HTLV-1-associated diseases. It has been reported that HAM/TSP patients generally had higher anti-HTLV-1 antibody titer than AC with the similar HTLV-1 proviral load[29-31]. These data suggest that there was persistent expression of HTLV-1 proteins *in vivo* and the existence of an augmented humoral immune response to HTLV-1 in HAM/TSP patients. Levin et al reported some intriguing evidence for antigen mimicry in HTLV-1 infection[32]. Namely, antibodies that recognize HTLV-1 Tax protein can cross-react with a host nuclear riboprotein hnRNP-A1. However, since the host protein hnRNP-A1 is not confined to the central nervous system but is widely expressed, and is not normally accessible to antibody attack, it is unlikely that anti-Tax antibody explains the onset or initial tissue damage of HAM/TSP. Rather, anti-Tax antibody might be associated with subsequent inflammation following initial tissue damage, which probably caused by the antiviral immune responses to HTLV-1 and induce the release of auto-antigens.

2) The natural killer (NK) cell response

Previous reports indicated that patients with HAM/TSP had both a lower frequency and a lower activity of NK cells (especially the $CD3^+$ $CD16^+$ subset) than AC, although the results were not normalized with respect to the proviral load[33]. Since an important mechanism of induction of NK cell-mediated killing is recognition by the NK cell of a complex of the non-polymorphic MHC molecule HLA-E bound to a peptide derived from the signal sequence of some other MHC class I molecules, synthetic tetramers of HLA-E with the HLA-G signal sequence peptide was used to identify NK cells in HAM/TSP patients[34]. The results clearly showed a lower frequency of HLA-E tetramer-binding cells in HAM/TSP patients than AC, and as in the earlier studies[33], this reduction in frequency was particularly notable in the $CD3^+$ cells whereas there was no significant difference in the frequency of HLA-E tetramer-binding $CD3^-$ cells between patients with HAM/TSP and AC. These results suggest that the activity of the NK or NK-like cell response was associated with the presence or absence of HAM/TSP. On the other hand, we previously reported that an uncontrolled preliminary trial by oral administration of viable *Lactobacillus casei* strain Shirota containing fermented milk for HAM/TSP patients resulted in significant increase of NK cell activity with improvements in clinical symptoms[35], suggesting that NK cells might be associated with the pathogenesis of HAM/TSP.

3) The regulatory T cells (Tregs)

It has been reported that HTLV-1 preferentially and persistently infects $CD4^+CD25^+$ lymphocytes *in vivo*[36], which contain the majority of the $Foxp3^+$ Tregs[37]. In HAM/TSP patients, the percentage of $Foxp3^+$ Tregs in $CD4^+CD25^+$ cells is lower than that in AC and uninfected healthy controls[38], however, the percentage of $Foxp3^+$ cells in the $CD4^+$ population tended to be higher in the HAM/TSP patients than in the AC[39]. This is probably because $CD25^+$ cells contain both Tregs and activated non-Tregs, and HTLV-1 infected individuals especially HAM/TSP patients increases the number of activated T cells expressing CD25. Interestingly, the percentage of $Foxp3^+$ Tregs positively correlated with the HTLV-1 proviral load and the CTL activity negatively correlated with the frequency of $Foxp3^+$ Tregs[39], suggesting that an increase in Tregs reduces CTL activity, which in turn increases the HTLV-1 proviral load.

4) The $CD4^+$ helper T cell response to HTLV-1

The HTLV-1 antigen most commonly recognized by $CD4^+$ T cells is the Envelope (Env) protein, in contrast with the

immunodominance of Tax in the CD8+ T cell response. Since an HTLV-1 Env gp21 immunodominant epitope was restricted by HLA-DRB1*0101, and HLA-DRB1*0101 was associated with susceptibility to HAM/TSP in independent HTLV-1-infected populations in southern Japan[21,22] and northeastern Iran[27], a synthetic tetramer of DRB1*0101 and the immunodominant HTLV-1 Env380-394 peptide was used to analyze Env-specific CD4+ T cells directly *ex vivo*[40]. The results clearly showed that the frequency of tetramer+ CD4+ T cells was significantly higher in HAM/TSP patients than AC with similar proviral load. Direct *ex vivo* analysis of tetramer+ CD4+ T cells from two unrelated DRB1*0101 positive HAM/TSP patients indicated that certain TCR Vβs were utilized and antigen-specific amino acid motifs were identified in CDR3 regions from both patients. These data suggest that the observed increase in virus-specific CD4+ T cells in HAM/TSP patients, which may contribute to CD4+ T cell-mediated antiviral immune responses and to an increased risk of HAM/TSP, was not simply due to the rapidly growing HTLV-1 infected CD4+ T cells but was the result of *in vivo* selection by specific MHC-peptide complexes, as observed in freshly isolated HLA-A*0201/ Tax11-19 tetramer+ CD8+ T cells[41] and muscle infiltrating cells from HAM/TSP patients and HTLV-1 infected polymyositis patients[42].

The cytotoxic T lymphocyte (CTL) response to HTLV-1

Previous reports indicated that the HTLV-1 specific CD8+ CTL are typically abundant, chronically activated, and mainly targeted to the viral transactivator protein Tax[6]. Also, as already mentioned, the median proviral load in PBMCs of HAM/TSP patients was more than ten times higher than that in AC, and a high proviral load was also associated with an increased risk of progression to disease[20]. Furthermore, HLA-A*02 and HLA-Cw*08 genes were independently and significantly associated with a lower proviral load and a lower risk of HAM/TSP[21,22], and CD8+ T cells efficiently kill autologous Tax-expressing lymphocytes in fresh PBMCs in HTLV-1 infected individuals[43]. These data have raised the hypothesis that the class I-restricted CD8+ CTL response plays a critical part in limiting HTLV-1 replication *in vivo*, and that genetically determined differences in the efficiency of the CTL response to HTLV-1 account for the risk for developing HAM/TSP. However, since the frequency of HTLV-1-specific CD8+ T cells were significantly elevated in HAM/TSP patients than AC[44,45], and these cells have the potential to produce proinflammatory cytokines[46], there is a debate on the role of HTLV-1-specific-CD8+ T cells, i.e. whether these cells contribute to the inflammatory and demyelinating processes of HAM/TSP, or whether the dominant effect of such cells *in vivo* is protective against disease, although these two mechanisms are not mutually exclusive. Recently, we reported that a frequency of CD8+ T cells that were negative for costimulatory molecules such as CD27, CD28, CD80, CD86 and CD152 were significantly higher in patients with HAM/TSP than in age-matched uninfected controls, but there was no such difference between AC and uninfected controls[47]. We also found a significantly lower frequency of perforin+ cells and granzyme B+ cells in the CD8+ T cells in HTLV-1 infected subjects than in uninfected controls, although there was no significant difference between patients with HAM/TSP and AC. Furthermore, the lytic capacity of HTLV-1 specific CTL between HAM/TSP and AC estimated by CD107a mobilization assay showed the significantly lower CD107a staining in HTLV-1 specific CTL in HAM/TSP than AC. Based on these findings, we have suggested that patients with HAM/TSP have a high frequency of HTLV-1 specific CD8+ T cells with poor lytic capacity, whereas AC have a lower frequency of cells with high lytic capacity.

Conclusions

As shown in Figure 1, the evidence summarized in this paper is consistent with the idea that virus-host immunologic interactions play a pivotal role in HAM/TSP pathogenesis. Genetically determined less efficient CTL response against HTLV-1 may cause higher proviral load and antigen expression in infected individuals, which lead to activation and expansion of antigen-specific T cell responses, subsequent induction of large amounts of proinflammatory cytokines and chemokines, and progression of HAM/TSP development.

Acknowledgments

The author thanks the Ministry of Health, Labor and Welfare, Japan, the Ministry of Education, Science, Sports and Culture, Japan (ref 17590886) and Takeda Science Foundation for financial supports.

References

1) Hinuma Y, Nagata K, Hanaoka M, Nakai M, Matsumoto T, Kinoshita KI, Shirakawa S, Miyoshi I: Adult T-cell leukemia: antigen in an ATL cell line and detection of antibodies to the antigen in human sera. Proc Natl Acad Sci USA, 78: 6476-6480, 1981.
2) Yoshida M, Seiki M, Yamaguchi K, Takatsuki K: Monoclonal integration of human T-cell leukemia provirus in all primary tumors of adult T-cell leukemia suggests causative

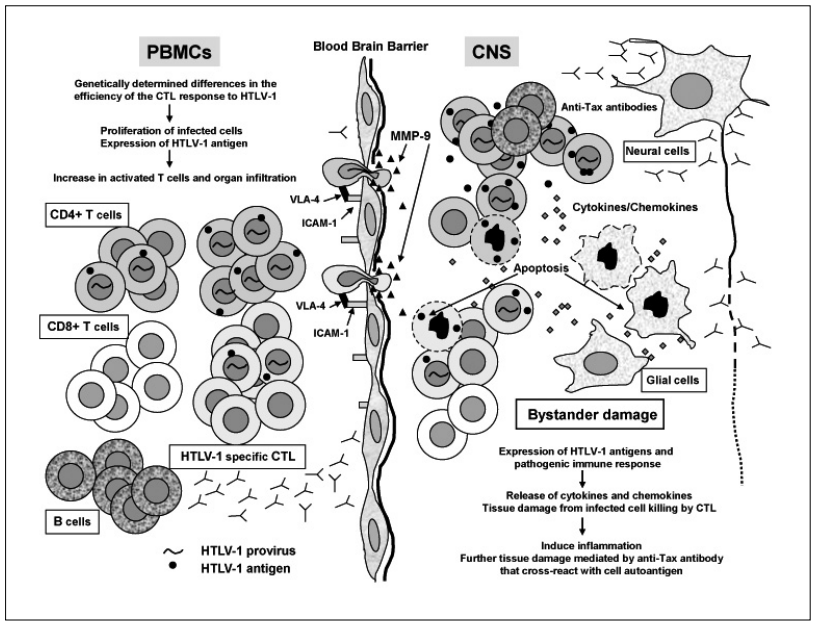

Fig.1 Hypothesis for the pathogenesis of Human T-cell leukemia virus type 1 (HTLV-1)-associated myelopathy/tropical spastic paraparesis (HAM/TSP)

In patients with HAM/TSP, genetically determined less efficient CTL response against HTLV-1 may cause higher proviral load and antigen expression, which lead to activation and expansion of antigen-specific T cell responses, subsequent induction of large amounts of proinflammatory cytokines and chemokines, and progression of HAM/TSP development. It is also possible that the immunoglobulin G specific to HTLV-1-Tax, which cross-react with heterogeneous nuclear ribonuclear protein-A1 (hnRNP-A1), is associated with subsequent inflammation following initial tissue damage.

role of human T-cell leukemia virus in the disease. Proc Natl Acad Sci USA, 81: 2534-2537, 1984.

3) Osame M, Usuku K, Izumo S, Ijichi N, Amitani H, Igata A, Matsumoto M, Tara M: HTLV-I associated myelopathy, a new clinical entity. Lancet, 1: 1031-1032, 1986.
4) Gessain A, Barin F, Vernant JC, Gout O, Maurs L, Calender A, de The G: Antibodies to human T-lymphotropic virus type-I in patients with tropical spastic paraparesis. Lancet, 2: 407-410, 1985.
5) Verdonck K, Gonzalez E, Van Dooren S, Vandamme AM, Vanham G, Gotuzzo E: Human T-lymphotropic virus 1: recent knowledge about an ancient infection. Lancet Infect Dis, 7: 266-281, 2007.
6) Bangham CR: The immune response to HTLV-I. Curr Opin Immunol, 12: 397-402, 2000.
7) Uchiyama, T: Human T cell leukemia virus type I (HTLV-I) and human diseases. Annu Rev Immunol, 15: 15-37, 1997.
8) Roucoux DF, Wang B, Smith D, Nass CC, Smith J, Hutching ST, Newman B, Lee TH, Chafets DM, Murphy EL: A prospective study of sexual transmission of human T lymphotropic virus (HTLV)-I and HTLV-II. J Infect Dis, 191: 1490-1497, 2005.
9) Proietti FA, Carneiro-Proietti AB, Catalan-Soares BC, Murphy EL: Global epidemiology of HTLV-I infection and associated diseases. Oncogene, 24: 6058-6068, 2005.
10) Hino S, Yamaguchi K, Katamine S, Sugiyama H, Amagasaki T, Kinoshita K, Yoshida Y, Doi H, Tsuji Y, Miyamoto T: Mother-to-child transmission of human T-cell leukemia virus type-I. Jpn J Cancer Res, 76: 474-480, 1985.
11) Kinoshita K, Amagasaki T, Hino S, Doi H, Yamanouchi K, Ban N, Momita S, Ikeda S, Kamihira S, Ichimaru M, et al: Milk-borne transmission of HTLV-I from carrier mothers to their children. Jpn J Cancer Res, 78: 674-680, 1987.
12) Nakagawa M, Nakahara K, Maruyama Y, Kawabata M, Higuchi I, Kubota H, Izumo S, Arimura K, Osame M: Therapeutic trials in 200 patients with HTLV-I-associated myelopathy/ tropical spastic paraparesis. J Neurovirol, 2: 345-355, 1996.
13) Iwasaki Y: Pathology of chronic myelopathy associated with HTLV-I infection (HAM/TSP). J Neurol Sci, 96: 103-123, 1990.
14) Yoshioka A, Hirose G, Ueda Y, Nishimura Y, Sakai K:

Neuropathological studies of the spinal cord in early stage HTLV-I-associated myelopathy (HAM). J Neurol Neurosurg Psychiatry, 56: 1004-1007, 1993.

15) Izumo S, Umehara F, Osame M: HTLV-I-associated myelopathy. Neuropathology, 20 Suppl: S65-S68, 2000.

16) Takenouchi N, Yamano Y, Usuku K, Osame M, Izumo S: Usefulness of proviral load measurement for monitoring of disease activity in individual patients with human T-lymphotropic virus type I-associated myelopathy/tropical spastic paraparesis. J Neurovirol, 9: 29-35, 2003.

17) Nagai M, Yamano Y, Brennan MB, Mora CA, Jacobson S: Increased HTLV-I proviral load and preferential expansion of HTLV-I Tax-specific CD8+ T cells in cerebrospinal fluid from patients with HAM/TSP. Ann Neurol, 50: 807-812, 2001.

18) Hayashi D, Kubota R, Takenouchi N, Nakamura T, Umehara F, Arimura K, Izumo S, Osame M: Accumulation of human T-lymphotropic virus type I (HTLV-I)-infected cells in the cerebrospinal fluid during the exacerbation of HTLV-I-associated myelopathy. J Neurovirol, 14: 459-463, 2008.

19) Jacobson S: Immunopathogenesis of human T cell lymphotropic virus type I-associated neurologic disease. J Infect Dis, 186 Suppl 2: S187-S192, 2002.

20) Nagai M, Usuku K, Matsumoto W, Kodama D, Takenouchi N, Moritoyo T, Hashiguchi S, Ichinose M, Bangham CR, Izumo S, et al: Analysis of HTLV-I proviral load in 202 HAM/TSP patients and 243 asymptomatic HTLV-I carriers: high proviral load strongly predisposes to HAM/TSP. J Neurovirol, 4: 586-593, 1998.

21) Jeffery KJ, Siddiqui AA, Bunce M, Lloyd AL, Vine AM, Witkover AD, Izumo S, Usuku K, Welsh KI, Osame M, et al: The influence of HLA class I alleles and heterozygosity on the outcome of human T cell lymphotropic virus type I infection. J Immunol, 165: 7278-7284, 2000.

22) Jeffery KJ, Usuku K, Hall SE, Matsumoto W, Taylor GP, Procter J, Bunce M, Ogg GS, Welsh KI, Weber JN, et al: HLA alleles determine human T-lymphotropic virus-I (HTLV-I) proviral load and the risk of HTLV-I-associated myelopathy. Proc Natl Acad Sci USA, 96: 3848-3853, 1999.

23) Vine AM, Witkover AD, Lloyd AL, Jeffery KJ, Siddiqui A, Marshall SE, Bunce M, Eiraku N, Izumo S, Usuku K, et al: Polygenic control of human T lymphotropic virus type I (HTLV-I) provirus load and the risk of HTLV-I-associated myelopathy/tropical spastic paraparesis. J Infect Dis, 186: 932-939, 2002.

24) Kodama D, Saito M, Matsumoto W, Sabouri AH, Izumo S, Arimura K, Usuku K, Bangham CR, Osame M: Longer dinucleotide repeat polymorphism in matrix metalloproteinase-9 (MMP-9) gene promoter which correlates with higher HTLV-I Tax mediated transcriptional activity influences the risk of HTLV-I associated myelopathy/tropical spastic paraparesis (HAM/TSP). J Neuroimmunol, 156: 188-194, 2004.

25) Sabouri AH, Saito M, Lloyd AL, Vine AM, Witkover AW, Furukawa Y, Izumo S, Arimura K, Marshall SE, Usuku K, et al: Polymorphism in the interleukin-10 promoter affects both provirus load and the risk of human T lymphotropic virus type I-associated myelopathy/tropical spastic paraparesis. J Infect Dis, 190: 1279-1285, 2004.

26) Furukawa Y, Yamashita M, Usuku K, Izumo S, Nakagawa M, Osame M: Phylogenetic subgroups of human T cell lymphotropic virus (HTLV) type I in the tax gene and their association with different risks for HTLV-I-associated myelopathy/tropical spastic paraparesis. J Infect Dis, 182: 1343-1349, 2000.

27) Sabouri AH, Saito M, Usuku K, Bajestan SN, Mahmoudi M, Forughipour M, Sabouri Z, Abbaspour Z, Goharjoo ME, Khayami E, et al: Differences in viral and host genetic risk factors for development of human T-cell lymphotropic virus type 1 (HTLV-1)-associated myelopathy/tropical spastic paraparesis between Iranian and Japanese HTLV-1-infected individuals. J Gen Virol, 86: 773-781, 2005.

28) Nose H, Saito M, Usuku K, Sabouri AH, Matsuzaki T, Kubota R, Eiraku N, Furukawa Y, Izumo S, Arimura K, et al: Clinical symptoms and the odds of human T-cell lymphotropic virus type 1-associated myelopathy/ tropical spastic paraparesis (HAM/TSP) in healthy virus carriers: application of best-fit logistic regression equation based on host genotype, age, and provirus load. J Neurovirol, 12: 171-177, 2006.

29) Ishihara S, Okayama A, Stuver S, Horinouchi H, Shioiri S, Murai K, Kubota T, Yamashita R, Tachibana N, Tsubouchi H, et al: Association of HTLV-I antibody profile of asymptomatic carriers with proviral DNA levels of peripheral blood mononuclear cells. J Acquir Immune Defic Syndr, 7: 199-203, 1994.

30) Kira J, Nakamura M, Sawada T, Koyanagi Y, Ohori N, Itoyama Y, Yamamoto N, Sakaki Y, Goto I: Antibody titers to HTLV-I-p40tax protein and gag-env hybrid protein in HTLV-I-associated myelopathy/tropical spastic paraparesis: correlation with increased HTLV-I proviral DNA load. J Neurol Sci, 107: 98-104, 1992.

31) Nagasato K, Nakamura T, Shirabe S, Shibayama K, Ohishi

K, Ichinose K, Tsujihata M, Nagataki S: Presence of serum anti-human T-lymphotropic virus type I (HTLV-I) IgM antibodies means persistent active replication of HTLV-I in HTLV-I-associated myelopathy. J Neurol Sci, 103: 203-208, 1991.

32) Levin MC, Lee SM, Kalume F, Morcos Y, Dohan FC Jr, Hasty KA, Callaway JC, Zunt J, Desiderio D, Stuart JM: Autoimmunity due to molecular mimicry as a cause of neurological disease. Nat Med, 8: 509-513, 2002.

33) Yu F, Itoyama Y, Fujihara K, Goto I: Natural killer (NK) cells in HTLV-I-associated myelopathy/tropical spastic paraparesis-decrease in NK cell subset populations and activity in HTLV-I seropositive individuals. J Neuroimmunol, 33: 121-128, 1991.

34) Saito M, Braud VM, Goon P, Hanon E, Taylor GP, Saito A, Eiraku N, Tanaka Y, Usuku K, Weber JN, et al: Low frequency of CD94/NKG2A+ T lymphocytes in patients with HTLV-1-associated myelopathy/tropical spastic paraparesis, but not in asymptomatic carriers. Blood, 102: 577-584, 2003.

35) Matsuzaki T, Saito M, Usuku K, Nose H, Izumo S, Arimura K, Osame M: A prospective uncontrolled trial of fermented milk drink containing viable Lactobacillus casei strain Shirota in the treatment of HTLV-1 associated myelopathy/tropical spastic paraparesis. J Neurol Sci, 237: 75-81, 2005.

36) Yamano Y, Cohen CJ, Takenouchi N, Yao K, Tomaru U, Li HC, Reiter Y, Jacobson S: Increased expression of human T lymphocyte virus type I (HTLV-I) Tax11-19 peptide-human histocompatibility leukocyte antigen A*201 complexes on CD4+ CD25+ T Cells detected by peptide-specific, major histocompatibility complex-restricted antibodies in patients with HTLV-I-associated neurologic disease. J Exp Med, 199: 1367-1377, 2004.

37) Sakaguchi S, Ono M, Setoguchi R, Yagi H, Hori S, Fehervari Z, Shimizu J, Takahashi T, Nomura T: Foxp3+ CD25+ CD4+ natural regulatory T cells in dominant self-tolerance and autoimmune disease. Immunol Rev, 212: 8-27, 2006.

38) Oh U, Grant C, Griffith C, Fugo K, Takenouchi N, Jacobson S: Reduced Foxp3 protein expression is associated with inflammatory disease during human t lymphotropic virus type 1 Infection. J Infect Dis, 193: 1557-1566, 2006..

39) Toulza F, Heaps A, Tanaka Y, Taylor GP, Bangham CR: High frequency of CD4+FoxP3+ cells in HTLV-1 infection: inverse correlation with HTLV-1-specific CTL response. Blood, 111: 5047-5053, 2008.

40) Nose H, Kubota R, Seth NP, Goon PK, Tanaka Y, Izumo S, Usuku K, Ohara Y, Wucherpfennig KW, Bangham CR, et al: Ex vivo analysis of human T lymphotropic virus type 1-specific CD4+ cells by use of a major histocompatibility complex class II tetramer composed of a neurological disease-susceptibility allele and its immunodominant peptide. J Infect Dis, 196: 1761-1772, 2007.

41) Saito M, Taylor GP, Saito A, Furukawa Y, Usuku K, Weber JN, Osame M, Bangham CR: In vivo selection of T-cell receptor junctional region sequences by HLA-A2 human T-cell lymphotropic virus type 1 Tax11-19 peptide complexes. J Virol, 75: 1065-1071, 2001.

42) Saito M, Higuchi I, Saito A, Izumo S, Usuku K, Bangham CR, Osame M: Molecular analysis of T cell clonotypes in muscle-infiltrating lymphocytes from patients with human T lymphotropic virus type 1 polymyositis. J Infect Dis, 186: 1231-1241, 2002.

43) Hanon E, Hall S, Taylor GP, Saito M, Davis R, Tanaka Y, Usuku K, Osame M, Weber JN, Bangham CR: Abundant tax protein expression in CD4+ T cells infected with human T-cell lymphotropic virus type I (HTLV-I) is prevented by cytotoxic T lymphocytes. Blood, 95: 1386-1392, 2000.

44) Greten TF, Slansky JE, Kubota R, Soldan SS, Jaffee EM, Leist TP, Pardoll DM, Jacobson S, Schneck JP: Direct visualization of antigen-specific T cells: HTLV-1 Tax11-19-specific CD8(+) T cells are activated in peripheral blood and accumulate in cerebrospinal fluid from HAM/TSP patients. Proc Natl Acad Sci USA, 95: 7568-7573, 1998.

45) Nagai M, Kubota R, Greten TF, Schneck JP, Leist TP, Jacobson S: Increased activated human T cell lymphotropic virus type I (HTLV-I) Tax11-19-specific memory and effector CD8+ cells in patients with HTLV-I-associated myelopathy/tropical spastic paraparesis: correlation with HTLV-I provirus load. J Infect Dis, 183: 197-205, 2001.

46) Kubota R, Kawanishi T, Matsubara H, Manns A, Jacobson S: Demonstration of human T lymphotropic virus type I (HTLV-I) tax-specific CD8+ lymphocytes directly in peripheral blood of HTLV-I-associated myelopathy/tropical spastic paraparesis patients by intracellular cytokine detection. J Immunol, 161: 482-488, 1998.

47) Sabouri AH, Usuku K, Hayashi D, Izumo S, Ohara Y, Osame M, Saito, M: Impaired function of human T-lymphotropic virus type 1 (HTLV-1)-specific CD8+ T cells in HTLV-1-associated neurologic disease. Blood, 112: 2411-2420, 2008.