

ASSOCIATION OF SPORADIC AMYOTROPHIC LATERAL SCLEROSIS WITH HUMAN T LYMPHOTROPIC VIRUS 1, HUMAN IMMUNODEFICIENCY VIRUS AND HUMAN HERPES VIRUS 8

A. Tarazi¹, S. Nafissi^{1*}, A. A. Amirzargar², P. Ayatollahi¹ and A. Soltanzadeh¹

1) Department of Neurology, Shariati Hospital, School of Medicine, Tehran University of Medical Sciences, Tehran, Iran

2) Department of Immunology, School of Medicine, Tehran University of Medical Sciences, Tehran, Iran

Abstract- Amyotrophic lateral sclerosis is a progressive neurodegenerative disease with uncertain etiology. For many years, viruses have been suspected as causative agents. There are conflicting reports about the possible role of viruses such as human herpes virus 8 (HHV8) and retroviruses in the pathogenesis of the sporadic amyotrophic lateral sclerosis. We conducted a prospective case-control study to investigate the association of seropositivity of HHV8, human T lymphotropic virus 1 (HTLV1) and human immunodeficiency virus (HIV) with risk of amyotrophic lateral sclerosis. Thirty patients with confirmed amyotrophic lateral sclerosis and 30 controls matched by sex and age were included in this study. Enzyme immunoassays for the determination of antibodies to HHV8, HTLV1 and HIV were performed on the serum samples of cases and controls. For HHV8, one in case group and one in control group were considered reactive for anti HHV8 IgG. For HTLV1 and HIV, the results were negative in both groups. This study does not support any association between seropositivity of HHV8, HIV and HTLV1 with amyotrophic lateral sclerosis.

Acta Medica Iranica, 44(4): 256-258; 2006

© 2006 Tehran University of Medical Sciences. All rights reserved

Key words: Amyotrophic lateral sclerosis, viral infections, human herpesvirus 8, human T lymphotropic virus 1, human immunodeficiency virus

INTRODUCTION

Amyotrophic lateral sclerosis (ALS) is characterized by degeneration of upper and lower motor neurons resulting in progressive weakness and death. Annual incidence rate is 0.4 to 1.76 per 100000 and men are affected twice as often as women. The pathophysiology of motor neuron degeneration has not been fully determined.

Received: 18 Jun. 2005, Revised: 16 Nov. 2005, Accepted: 20 Nov. 2005

*** Corresponding Author:**

S. Nafissi, Department of Neurology, Shariati Hospital, School of Medicine, Tehran University of Medical Sciences, Tehran, Iran
Tel: +98 21 8633039
Fax: +98 21 8633039
E-mail: nafisi@sina.tums.ac.ir

Numerous hypotheses about the role of toxic, immunologic, genetic and infectious factors have been suggested but none of them has been exactly proven. Viruses have been suspected as causative agents since many years (1). There are several reports about the etiologic role of enteroviruses (2), human herpesvirus 8 (HHV8) (2, 3), retroviruses such as human T lymphotropic virus (HTLV) (4) and human immunodeficiency virus (HIV) (5) in ALS; however, these findings were not supported by other studies (6, 7).

The purpose of the present study was to evaluate the association between ALS risk and seropositivity of HTLV1, HIV and HHV8 in a case control study.

MATERIALS AND METHODS

Patients and controls

Thirty consecutive patients admitted to the Shariati Hospital between June 2003 and September 2004 with definite or probable sporadic ALS according to the El Escorial criteria were selected (8). Twenty six patients (86.7%) were male and four patients (13.3%) were female. The mean age at the time of first presentation was 48.3 years (range 21-69 years). The overall mean duration of the disease from onset was 1.53 years (SD 1.02, range 0.3-4 years). The presentation of the disease in 20 patients (66.7%) was spinal and in 10 patients (33.3%) was bulbar. Necessary investigations were undertaken in all patients to exclude other disease processes. For evaluation of seropositivity of HHV8, HTLV1 and HIV, a blood sample was obtained from patients at the time of diagnosis.

For each patient, an age and sex matched control from patients with stroke or epilepsy was selected. Informed consent was obtained from all participants.

Serological studies

The serological examinations were performed at the Immunogenetic Laboratory of Tehran University of Medical Sciences. All sera were tested for HTLV1, HIV and HHV8 antibodies by using ELISA test provided by DRG for HTLV1, Biomerieux for HIV and Biotrin for HHV8. The Biotrin HHV8 ELISA is a direct EIA based on the binding of HHV8 specific antibodies to lytic peptide antigens coupled to microlitre test strips. Specifically bound antibodies are detected by an anti human IgG peroxidase conjugate and a subsequent substrate reaction. The presence or absence of anti HHV8 IgG is determined in relation to the cut-off calibrator (COC). Samples with an absorbance reading greater than $COC \times 1.2$ were considered reactive and those less than $COC \times 0.8$ are considered non reactive for anti-HHV8 IgG. In this study, the COC value was 0.218. DRG HTLV1 EIA employs microplates coated with synthetic antigens derived from the most immunogenic epitopes. Results are evaluated against a cut off value as negative or positive. Ratio of the sample OD450nm and the cut-off value (S/Co) is measured. $S/Co < 1.0$ was considered negative and more than 1.2 was considered positive. The

Biomerieux HIV EIA is based on a one step principle. A mixture of HIV antigens and HIV antibodies coupled to horseradish peroxidase serves as the conjugate with tetramethylbenzidine and peroxide as the substrate. The development of color indicates the presence of HIV antibody or HIV antigen, while no or low color development suggests the absence of HIV antibodies or antigens. A test sample is reactive if sample absorbance is $>$ cut off value and non reactive if $<$ cut off. The cut off is calculated by the absorbance of the negative control (NC) + 0.100.

RESULTS

All sera from ALS patients and control group were negative for HTLV1 and HIV antibodies when examined by ELISA. The serum sample of one ALS patient and one from control group were positive for HHV8 antibody. The only patient positive for HHV8 antibody was a 60 year-old man with a 3 year history of bulbar symptoms with gradual progression of weakness and atrophy to involve the limbs. He had clinically probable ALS according to El Escorial criteria without any atypical feature.

DISCUSSION

A viral etiology for ALS has long been considered because of the selective vulnerability of motor neurons to certain viruses. Detection of enterovirus nucleic acid in the neuronal cell bodies within the gray matter of the spinal cord of patients with ALS supports the association between persistent enterovirus RNA and ALS (9). In this study, we decided to search for three neurotropic viruses in ALS patients: HTLV1, HIV and HHV8.

The potential role of retroviruses as causative factors in ALS is hypothesized by several studies, although their results are conflicting. In one study, the HTLV proviral DNA was found in peripheral blood cells of ALS patients (4). They suggested the involvement of HTLV related viruses in ALS. In contrast, this hypothesis was not supported by another similar study (6). A retrospective study described six patients of HIV1 associated ALS like disorder (5) but no clear relationship between HIV

infection and ALS has been confirmed. An ALS like syndrome with new HIV infection recovered completely after treatment with antiretroviral drugs (10). In our study, all sera of the ALS patients and the control group were negative for HTLV1 and HIV antibody. Our results do not support the association between these retroviruses and ALS.

In recent studies, it has been shown that HHV8 besides being associated with Kaposi sarcoma and lymphoproliferative disease is strongly neurotropic. A few studies detected the evidence of HHV8 infection in ALS patients (2, 3) and suggested a possible relationship between these two; however, another effort failed to prove it (7). In our study, an ALS patient and a control subject were considered seropositive for anti HHV8 IgG. Therefore, there was no significant evidence of HHV8 infection in our ALS group. We found no causal association between retroviruses as well as HHV8 and ALS. The discrepancy between our results and other reports is probably due to difference in methodological strategy. It should be noted that our sample may not be truly representative of the Iranian ALS population. Thus, the results of our study might not be generalized to the whole ALS patients in Iran. Further research effort is required to determine the association of retroviruses and HHV8 with ALS in Iranian population.

Acknowledgement

We wish to thank the patients who freely participated in this study. This investigation was supported by a grant from the Tehran University of Medical Sciences.

Conflict of Interests

We have no conflict of interests.

REFERENCES

1. Weiner LP, Stohlman SA, Davis RL. Attempts to demonstrate virus in amyotrophic lateral sclerosis. *Neurology*. 1980 Dec; 30(12):1319-1322.
2. Cermelli C, Vinceti M, Beretti F, Pietrini V, Nacci G, Pietrosevoli P, Bartoletti A, Guidetti D, Sola P, Bergomi M, Vivoli G, Portolani M. Risk of sporadic amyotrophic lateral sclerosis associated with seropositivity for herpesviruses and echovirus-7. *Eur J Epidemiol*. 2003; 18(2):123-127.
3. Cone LA, Nazemi R, Cone MO. Reversible ALS-like disorder in HIV infection. An ALS-like syndrome with new HIV infection and complete response to antiretroviral therapy. *Neurology*. 2002 Aug 13; 59(3):474
4. Ferrante P, Westarp ME, Mancuso R, Puricelli S, Westarp MP, Mini M, Caputo D, Zuffolato MR. HTLV tax-rex DNA and antibodies in idiopathic amyotrophic lateral sclerosis. *J Neurol Sci*. 1995 May; 129 Suppl:140-144.
5. Moulignier A, Moulounguet A, Pialoux G, Rozenbaum W. Reversible ALS-like disorder in HIV infection. *Neurology*. 2001 Sep 25; 57(6):995-1001.
6. Andrews WD, Al-Chalabi A, Garson JA. Lack of evidence for HTLV tax-rex DNA in motor neurone disease. *J Neurol Sci*. 1997 Dec 9; 153(1):86-90.
7. Sola P, Bedin R, Casoni F, Barozzi P, Mandrioli J, Merelli E. New insights into the viral theory of amyotrophic lateral sclerosis: study on the possible role of Kaposi's sarcoma-associated virus/human herpesvirus 8. *Eur Neurol*. 2002; 47(2):108-112.
8. Brooks BR. El Escorial World Federation of Neurology criteria for the diagnosis of amyotrophic lateral sclerosis. Subcommittee on Motor Neuron Diseases/Amyotrophic Lateral Sclerosis of the World Federation of Neurology Research Group on Neuromuscular Diseases and the El Escorial "Clinical limits of amyotrophic lateral sclerosis" workshop contributors. *J Neurol Sci*. 1994 Jul; 124 Suppl:96-107.
9. Berger MM, Kopp N, Vital C, Redl B, Aymard M, Lina B. Detection and cellular localization of enterovirus RNA sequences in spinal cord of patients with ALS. *Neurology*. 2000 Jan 11; 54(1):20-25.
10. MacGowan DJ, Scelsa SN, Waldron M. An ALS-like syndrome with new HIV infection and complete response to antiretroviral therapy. *Neurology*. 2001 Sep 25; 57(6):1094-1097.