Probable Post-Influenza Cerebellitis

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The nucleoprotein (NP) gene of type B influenza virus was detected by reverse transcription polymerase chain reaction (RT-PCR) from the cerebrospinal fluid (CSF) of a patient presenting with ataxia due to cerebellitis. The CSF was obtained 7 and 9 weeks after flu syndrome occurred, suggesting persistence of viral genes in the central nervous system (CNS). Although an unusually high serum hemagglutination inhibition (HI) titer against influenza virus B was noted, HI titers of the CSF were not elevated.

(Key words: nucleocapsid protein gene, polymerase chain reaction (PCR), type B influenza virus)

Introduction

Influenza A and B viruses occasionally cause acute encephalopathy and postinfection encephalitis (1-5). In addition, the pandemic of influenza A in 1919 was followed by an increased incidence of von Economo lethargic encephalitis and ensuing parkinsonism (6, 7). However, the association of these central nervous system (CNS) disorders with influenza virus infection has not been precisely clarified because the virus or viral antigen has only rarely been identified in the lesion. From the cerebrospinal fluid (CSF) of a patient with post-influenza cerebellitis, we recently amplified the nucleotide sequence of influenza B virus by reverse transcription polymerase chain reaction (RT-PCR).

Case Report

A 31-year-old woman presented with flu symptoms of fever up to 39°C, sore throat, myalgia and arthralgia at the beginning of September 1996 and gradually developed ataxia and dysarthria which naturally waned and disappeared in three months. Initial physical examination was normal except for cerebellar signs including ataxic gait, scanning speech, poor coordination, truncal titubation, decreased tone of the extremity muscles, and rebound phenomenon. Peripheral blood cell count, chemistry, and urinalysis were normal. CSF was clear and colorless with a normal cell count, protein, and glucose. The serum hemagglutination inhibition (HI) titer to influenza virus B was remarkably elevated, although that of the CSF was negative (Fig. 1). There was no elevation of antibody titer to EB virus or mumps virus. Computed tomography (CT) scan and magnetic resonance imaging of the brain were essentially normal. There was no history of taking aspirin which precipitates Reye encephalopathy (8-10). Cerebellitis due to type B influenza virus was suspected from the laboratory data and clinical course (Fig. 1).

Detection of influenza virus gene from the CSF was performed by the method previously described (11) with modifications using 10 μl of CSF. Reverse transcription was performed using random primers (hexadeoxyribonucleotide mixture, Takara), and reverse transcriptase derived from Molony murine leukemia virus (Wako, Tokyo). The base sequences of the primers LNP-1 (5'-CAGCATTTTCTTGTGAGCTTCG-3'), LNP-2 (5'-TAATCCTCTGCTGTGTCCCTCC-3'), LNP-3 (5'-AGGGACTGAAAAGGGTTGGACT-3'), and LNP-4 (5'-GGCTTCATACCCAACCATAGAG-3') corresponded to 11-32, 1,699-1,721, 695-716, and 1,113-1,134 of the plus sense of the NP gene of influenza virus B/Lee/40 (12). In the second polymerase chain reaction (PCR), 1 μl of the first PCR product was used as the template. Twenty PCR cycles consist of denaturing at 94°C for 1 minute, annealing at 55°C for 30 seconds and elongation at 72°C for 2 minutes; these were performed in both the first PCR and the second PCR. Although in the first PCR using LNP-1 and LNP-2, 1,710 base-pair band was not detected, the second PCR using LNP-3 and LNP-4 revealed 440 base-pair bands (Fig. 2). No band was detected from the CSF of a healthy individual. Influenza virus was not isolated from the CSF in Madin Darby Canine Kidney (MDCK) cells.
Flu syndrome  Cerebellar ataxia

![Graph showing the clinical course of the patient. The rise of serum HI titer against influenza virus B/Mie/1/93 was noted. The CSF and serum were obtained on 10/11/96 and 10/24/96.]

**Figure 1.** Clinical course of the patient. The rise of serum HI titer against influenza virus B/Mie/1/93 was noted. The CSF and serum were obtained on 10/11/96 and 10/24/96.

<table>
<thead>
<tr>
<th>Antibody titer to flu B</th>
<th>Serum</th>
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<tr>
<td></td>
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**Figure 2.** Agarose gel electrophoresis of the nested RT-PCR products of the influenza-infected cells and the patient’s CSF. (A) Lane 1, the second PCR product of influenza B/Lee/40 infected cells (BL); lane 2, the second PCR product of the patient’s CSF of 10/11/96; lane 3, the first PCR product of BL; lane 4, the first PCR product of the patient’s CSF of 10/11/96; lane 5, molecular weight marker. (B) Lane 1, molecular weight marker; lane 2, the first PCR product of the patient’s CSF of 10/24/96; lane 3, the first PCR product of influenza B/Lee/40 infected cells (BL); lane 4, the second PCR product of the patient’s CSF of 10/24/96; lane 5, the second PCR product of BL.

**Discussion**

There are two recognized entities of influenza virus-related CNS disorder; influenza encephalopathy and post-influenzal encephalitis. Influenza encephalopathy occurs in the acute phase of flu syndrome and the viruses are sometimes isolated from the CSF. Post-influenzal encephalitis occurs 2 to 4 weeks after flu syndrome and the viruses have never been isolated; it is thought to be an autoimmune process or angiomyelinopathy (13, 14). The NP gene of influenza virus B, which is thought to be essential for virus replication, was detected in the second PCR from the CSF of 7 weeks and 9 weeks after the onset of flu syndrome. These results suggest not only the viral etiology of this patient’s cerebellitis but also the possible persistence of viral genes in the CNS between initial flu syndrome and cerebellar ataxia. However, etiologies other than influenza virus infection can not be ruled out completely in the absence of virus detection or seroconversion during the initial flu syndrome. Prolonged shedding of influenza viruses in
immunocompromized hosts (15–17) and the persistence of influenza viral genes in cultured cells (18) have been reported. The sharp increase in the antibody titer from 10/11 to 10/24/96 (Fig. 1) seems to be related to severe viremia precipitating the infection of the CNS. Although type A influenza viruses infect humans, swine, avians, and seals, type B viruses infect only humans. Reservoirs of type B viruses between epidemic seasons are yet to be identified. Detection of influenza virus gene in the CSF from this patient and the lack of rise in HI titer of CSF imply that human CNS could be one of the reservoirs of type B influenza virus during the nonepidemic period, where virus genes persistently replicate, evading the attack of immune systems.

References

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