ATYPICAL HSV ENCEPHALITIS WITH INITIAL NEGATIVE POLYMERASE CHAIN REACTION FOR HSV DNA

Şua Sümer, Onur Ural
Selcuk University, Faculty of Medicine, Department of Infectious Disease and Clinical Microbiology, Konya, Turkey

ABSTRACT

HSV-DNA in cerebrospinal fluid by polymerase chain reaction is considered as the gold standard in the diagnosis of Herpes Simplex Virus (HSV) encephalitis. Sometimes the test may be negative in the initial stage of the disease. HSV-DNA quantitation by polymerase chain reaction should be repeated in 3-7 days if a patient is thought to have HSV encephalitis, as indicated by electroencephalography / magnetic resonance imaging of brain findings even though the initial HSV-DNA quantitation by polymerase chain reaction is negative. We presented two cases with a tentative diagnosis of HSV encephalitis with atypical course, whose polymerase chain reaction results were negative for HSV-DNA initially.

Key Words: *HSV-DNA, Herpes Simplex Virus encephalitis, polymerase chain reaction* ***Nobel Med 2014; 10(1): 85-87***

POLİMERAZ ZİNCİR REAKSİYONU İLE BAŞLANGIÇ HSV-DNA'SI NEGATİF OLAN ATİPİK HSV ENSEFALİTİ

ÖZET

Polimeraz zincir reaksiyonu ile beyin omurilik sıvısında HSV-DNA araştırılması HSV ensefaliti tanısında kullanılan altın standart yöntemdir. Bazen hastalığın başlangıç döneminde test negatif olabilir. Herpes Simpleks Virüs (HSV) ensefaliti düşünülen ve beyin elektroensefalografi/manyetik rezonans bulguları uyumlu hastalarda ilk gönderilen polimeraz zincir reaksiyonu ile HSV-DNA negatif saptanırsa, polimeraz zincir reaksiyonu tetkikinin 3-7 gün sonra tekrarlanması önerilir. Bu yazıda, HSV ensefaliti ön tanısıyla tedavi başlanan fakat polimeraz zincir reaksiyonu ile HSV-DNA sonucu negatif olmasına rağmen tekrarlayan polimeraz zincir reaksiyonu tetkikleri sonucu HSV-DNA pozitifliği izlenen atipik seyirli iki olgu sunulmuştur.

Anahtar Kelimeler: HSV DNA, Herpes Simpleks virüs ensefaliti, polimeraz zincir reaksiyonu **Nobel Med 2014; 10(1): 85-87**

INTRODUCTION

Herpes simplex virus (HSV) encephalitis is the most common form of encephalitis acute sporadic encephalitis, with an annual incidence of 1/250,000-500,000.[1-4] Significant improvements in mortality and morbidity of HSV encephalitis have been achieved, due to early antiviral treatment.[3-5] Mortality rate in untreated HSV encephalitis can be as high as 70%,[1-4,6] while recovery without sequelae is less than 3%.[3,4] Therefore, establishing a rapid and accurate diagnosis is critical in the management of the disease.[5]

Magnetic resonance imaging (MRI) of the brain, electroencephalography (EEG) and identification of HSV DNA by polymerase chain reaction (PCR) are used for the diagnosis of the HSV encephalitis. To date, isolation of HSV DNA in cerebrospinal fluid (CSF) by PCR is considered as the gold standard in the diagnosis of HSV encephalitis.[4-7] Furthermore, since the HSV DNA level measured by PCR correlates significantly with prognosis, this technique is also recommended in patients receiving acyclovir treatment as well.[4] In the majority of patients with HSV encephalitis, PCR depicts that HSV DNA is positive though cases initially reported as negative HSV DNA.[4,7] Therefore, guidelines and publications recommend that antiviral treatment be started if encephalitis patients with initial negative PCR result for HSV DNA have temporal involvement, either suspected clinically or as depicted by imaging techniques, and that PCR be repeated in 3-7 days.[4,7,8]

We presented two cases with a tentative diagnosis of HSV encephalitis with atypical course, whose PCR results were negative for HSV DNA initially but became positive during the course of treatment. →

CASE 1

A 37 year-old female patient presented to the emergency room with severe headache and confusion for one day. The patient had a temperature of 38.4°C and was lethargic. Patient had neck stiffness while Burdzinski's sign and Babinski's sign bilaterally were positive. Physical examination was otherwise normal. Lumbar puncture (LP) showed clear CSF with 319 cells/mm^3 (95% lymphocytes). CSF biochemistry revealed protein: 131 mg/dL, glucose: 66 mg/dL (simultaneous blood glucose: 107 mg/dL). Cranial MRI and EEG were ordered. Specimen from the CSF was sent for Gram staining, culture and HSV DNA quantitation. Patient was admitted to the Infectious Diseases and Clinical Microbiology Clinic with a tentative diagnosis of encephalitis and placed on acyclovir at a dose of 10 mg/kg three times a day. Gram staining and culture did not reveal any pathogen. Although cranial MRI and EEG findings were consistent with HSV encephalitis, HSV DNA was negative. Treatment with acyclovir was continued and patient's fever and neurological signs resolved while her general status became normal on day 3. On fifth day of her hospitalization, LP was repeated and sample was sent for HSV DNA quantitation. Results of the second LP showed clear CSF with 143 cells/mm^3 (80% lymphocyte). CSF biochemistry then revealed protein: 125 mg/dL, glucose: 54 mg/dl (simultaneous blood glucose: 98 mg/dL). HSV DNA level measured in the second sample was 24,900 copies/mL. Once the diagnosis was established and appropriate treatment was continued, CSF findings resolved and HSV DNA became negative and treatment was terminated on day 21. Patient healed without any sequelae.

CASE 2

A 41-year-old male patient presented to the emergency room complaining of severe headache, nausea and vomiting for 2 days. During the physical examination, patient was found lethargic with a temperature of 37.6 °C. Neck stiffness was present whereas there was no response to Babinski test bilaterally. LP showed hemorrhagic CSF with 396 cells/mm^3 (55% lymphocyte) while CSF biochemistry indicated protein: 183 mg/dL, glucose: 86 mg/dL (simultaneous blood glucose: 166 mg/dL). Sample from the CSF was sent for Gram staining, culture and HSV DNA quantitation in addition to the cranial MRI and EEG. Patient was admitted to the Infectious Diseases and Clinical Microbiology Clinic with a tentative diagnosis of encephalitis and treatment with acyclovir at a dose of 10 mg/kg three times a day and ceftriaxone 2 g twice a day was started. Gram staining and culture did not reveal any pathogen. Cranial MRI and EEG findings were consistent with HSV encephalitis even though HSV DNA was negative on the second day of hospitalization. Treatment was continued and spinal tap was repeated on the third day of treatment. Despite the fact that CSF findings improved with treatment, HSV DNA in the second sample was negative again. On the fifth day of his admittance, LP was repeated again and showed clear CSF with 121 cells/mm^3 (90% lymphocyte). CSF biochemistry revealed protein: 102 mg/dL, glucose: 62 mg/dL (simultaneous blood glucose: 105 mg/dL). HSV DNA in the third sample was 12,300 copy/ml. Ceftriaxone was terminated and treatment with acyclovir was continued. Treatment was terminated on day 24 when the CSF findings resolved and HSV DNA became negative. Patient healed without any sequelae.

DISCUSSION

HSV encephalitis, which is the most common form of sporadic encephalitis, can be seen in all age groups and in both sexes.[1,5] The pathogen is usually type 1 HSV and infection develops due to reactivation of a latent virus. Initially, majority of the patients have fever, headache and diminished level of consciousness while pleocytosis of CSF is almost always present.[1,2] Further, CSF may be hemorrhagic if and when it causes hemorrhagic and necrotizing encephalitis.[1,9] When encephalitis is suspected in a patient, guidelines recommend that acyclovir treatment must be initiated, and antimicrobial drugs must be added, when appropriate, until the results of diagnostic tests are available. If the time between the onset of symptoms and acyclovir treatment is longer than 4 days, it is considered as a poor prognostic criterion.[8] By that way, it will be possible to prevent delays in treatment and, consequently, morbidity and mortality rates would be reduced. Presenting symptoms, physical examination findings and CSF findings of both of our cases were consistent with HSV encephalitis. Moreover, CSF was hemorrhagic in one case. However, there were no findings that could be attributed to trauma or subarachnoid hemorrhage during spinal tap. Acyclovir treatment was started empirically while waiting for the results of diagnostic tests. Moreover, we could not rule out bacterial meningitis initially and, therefore, put the patient on both acyclovir and ceftriaxone treatment.

Characteristic leptomeningeal involvement in the frontal-temporal lobe, as depicted by cranial MRI and periodic lateralized epileptiform changes in the temporal region on EEG lend support to the confirmation of diagnosis in patients with consistent clinical findings.[1,2,8,10,11] Among the laboratory methods, identification of HSV DNA by PCR has a sensitivity between 91-98%, and specificity between 92-99%.[1,3,5] Even though HSV DNA quantitation by PCR is a very sensitive and reliable method, →

sometimes the test may be negative in the initial stage of the disease. Literature and guidelines recommend that HSV DNA quantitation by PCR should be repeated in 3-7 days if a patient is thought to have HSV encephalitis, as indicated by EEG/MRI findings even though the initial HSV DNA quantitation by PCR is negative. During this time period, the detectable viral load increases.[3-5,8] In patients with suspected HSV encephalitis, appropriate treatment should be initiated without waiting for test results. Even if the PCR result is negative, the treatment should not be discontinued unless a different diagnosis is proved.[4,5] It has been reported that patients with a negative PCR result for HSV DNA initially were especially the ones with symptoms of short duration (1-3 days) and this was attributed to low level of target DNA.[5] In another study on this matter, the authors presented 3 patients with initial negative PCR result for HSV DNA whose cranial MRI findings were consistent with HSV encephalitis and reported that HSV DNA became positive on day 4 in two patients and on day 7 in one patient.[5]

Among the causes of negative initial HSV DNA by PCR are insufficient target DNA in CSF, sensitivity of the PCR method and presence of erythrocyte in CSF, thereby reducing the sensitivity of the test. The likelihood of quantitation by PCR, despite its high sensitivity, decreases if there is insufficient target DNA. Nested PCR, which utilizes a second amplification, is more sensitive than conventional PCR. Moreover, presence of erythrocyte in CSF, even at small amounts, exhibits inhibitory effect and may result in an inaccurate result.[5] In addition, although rare, HSV DNA positivity may not be detected at all in some cases. In such a circumstance, appropriate treatment should be administered if the clinical picture is consistent with HSV encephalitis and other laboratory findings also support the existence of an infection.[4] HSV DNA became positive on day 5 in both cases. The method employed in the present study was conventional PCR and CSF was initially hemorrhagic in one case. Hence, we attributed the negative initial HSV DNA to a number of factors.

In patients with established HSV encephalitis, while under acyclovir treatment, monitoring the HSV DNA level by PCR is recommended. Antiviral treatment should be continued until PCR becomes negative. During treatment of HSV encephalitis, a slow response can sometimes be observed in patients. In a study, HSV DNA became negative in 8 out of 10 patients with established HSV encephalitis while a decrease in the viral load was observed in two cases.[4] Acyclovir resistance has not been defined in immune competent patients.[3] Acyclovir resistance can be observed in the presence of immune suppression and concomitant long-term acyclovir use (such as in AIDS patients and recipients of bone marrow transplant).[3,12] HSV DNA quantitation was carried out during the treatment of the patients. Persistence of HSV DNA positivity on 14th day of the treatment necessitated extending the treatment in both cases. Since the patients did not have a secondary disease that might cause immune suppression, acyclovir resistance was not considered.

Suspicion of HSV encephalitis is sufficient for the initiation of antiviral treatment. Even if the initial PCR is negative for HSV DNA, treatment should not be interrupted in case that clinical and other findings support the infection and PCR should be repeated.

C **CORRESPONDING AUTHOR:** Şua Sümer Selcuk Univ, Faculty of Med, Dept. of Infectious Disease and Clinical Microbiology Alaeddin Keykubat Kampüsü 42250 Konya suasumer@gmail.com

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