ORIGINAL ARTICLE

Managing atypical and typical herpetic central nervous system infections: results of a multinational study

Y. Cag[1], H. Erdem[2], S. Leib[3], S. Defres[4,5], S. Kaya[6], L. Larsen[7], M. Poljak[8], D. Ozturk-Engin[9], B. Barsic[10], X. Argemi[11], S. M. Sørensen[12], A. L. Bohr[13], P. Tattevin[14], J. D. Gunst[15], L. Baštáková[16], M. Jereb[17], I. S. Johansen[7], O. Karabay[18], A. U. Pekok[19], O. R. Sipahi[20], M. Chehri[21], G. Beraud[22], G. Shehata[23], R. Fontana[24], M. Maresca[24], H. Karsen[25], G. Sengoz[26], M. Sunbul[27], G. Yilmaz[28], H. Yilmaz[27], A. Sharif-Yakan[29], S. Kanj[29], E. Parlak[30], F. Pehlivanoglu[26], F. Korkmaz[31], S. Komur[32], S. Kose[33], M. Ulug[34], S. Bolukcu[9], S. A. Coskuner[35], J. P. Stahl[36], N. Ince[37], Y. Akkoyunlu[38], G. Halac[39], E. Sahin-Horasan[40], H. Tireli[41], G. Kilicoglu[42], A. Al-Mahdawi[43], S. A. Nemli[44], A. Inan[9], S. Senbayrak[9], H. Vahaboglu[45] and N. Elaldi[46]

1) *Dr Lütfi Kirdar Training and Research Hospital, Department of Infectious Diseases and Clinical Microbiology, Istanbul, Turkey,* 2) *Gulhane Medical Academy, Department of Infectious Diseases and Clinical Microbiology, Ankara, Turkey,* 3) *Institute for Infectious Diseases, University of Bern, Switzerland,* 4) *Institute of Infection and Global Health, University of Liverpool, United Kingdom,* 5) *Tropical Infections Diseases Unit In Royal Liverpool and Broadgreen University Hospitals NHS Trust, United Kingdom,* 6) *Karadeniz Technical University School of Medicine, Department of Infectious Diseases and Clinical Microbiology, Trabzon, Turkey,* 7) *Odense University Hospital, Department of Infectious Diseases Q, Odense, Denmark,* 8) *Institute of Microbiology and Immunology, Faculty of Medicine, University of Ljubljana, Ljubljana, Slovenia,* 9) *Haydarpasa Numune Training and Research Hospital, Department of Infectious Diseases and Clinical Microbiology, Istanbul, Turkey,* 10) *Dr. Fran Mihaljevic University Hospital for Infectious Diseases, Department of Infectious Diseases, University of Zagreb School of Medicine, Zagreb, Croatia,* 11) *Nouvel Hôpital Civil, Department of Infectious Diseases, Strasbourg, France,* 12) *Aalborg University Hospital, Department of Infectious Diseases, Denmark,* 13) *Copenhagen University Hospital, Institute of Inflammation Research, Department of Infectious Diseases and Rheumatology, Rigshospitalet, Denmark,* 14) *University Hospital of Pontchaillou, Department of Infectious and Tropical Diseases, Rennes, France,* 15) *Aarhus University Hospital, Department of Infectious Diseases, Aarhus, Denmark,* 16) *Faculty Hospital Brno, Department of Infectious Diseases, Masaryk University, Faculty of Medicine, Brno, Czech Republic,* 17) *University Medical Centre, Department of Infectious Diseases, Ljubljana, Slovenia,* 18) *Sakarya University School of Medicine, Department of Infectious Diseases and Clinical Microbiology, Sakarya, Turkey,* 19) *Private Erzurum Sifa Hospital, Department of Infectious Diseases and Clinical Microbiology, Erzurum, Turkey,* 20) *Ege University School of Medicine, Department of Infectious Diseases and Clinical Microbiology, Izmir, Turkey,* 21) *Hvidovre Hospital, Department of Infectious Diseases, Copenhagen, Denmark,* 22) *Poitiers University Hospital, Department of Infectious Diseases, France,* 23) *Assiut University Hospital, Department of Neurology and Psychiatry, Assiut, Egypt,* 24) *University of Catania, Section of Infectious Diseases, Department of Clinical and Molecular Biomedicine, Catania, Italy,* 25) *Harran University, School of Medicine, Department of Infectious Diseases and Clinical Microbiology, Sanliurfa, Turkey,* 26) *Haseki Training and Research Hospital, Department of Infectious Diseases and Clinical Microbiology, Istanbul, Turkey,* 27) *Ondokuz Mayis University School of Medicine, Department of Infectious Diseases and Clinical Microbiology, Samsun, Turkey,* 28) *Ankara University School of Medicine, Department of Infectious Diseases and Clinical Microbiology, Ankara, Turkey,* 29) *American University of Beirut Medical Center, Beirut, Lebanon,* 30) *Ataturk University School of Medicine, Department of Infectious Diseases and Clinical Microbiology, Erzurum, Turkey,* 31) *Konya Training and Research Hospital, Department of Infectious Diseases and Clinical Microbiology, Konya, Turkey,* 32) *Cukurova University School of Medicine, Department of Infectious Diseases and Clinical Microbiology, Adana, Turkey,* 33) *Tepecik Training and Research Hospital, Department of Infectious Diseases and Clinical Microbiology, Izmir, Turkey,* 34) *Private Umit Hospital, Department of Infectious Diseases and Clinical Microbiology, Eskisehir, Turkey,* 35) *Izmir Bozyaka Training and Research Hospital, Department of Infectious Diseases and Clinical Microbiology, Izmir, Turkey,* 36) *Joseph Fourier University and University Hospital of Grenoble, Department of Infectious Diseases, Grenoble, France,* 37) *Duzce University School of Medicine, Department of Infectious Diseases and Clinical Microbiology, Konuralp, Duzce, Turkey,* 38) *Bezmi Alem Vakif University, School of Medicine, Department of Infectious Diseases and Clinical Microbiology, Istanbul, Turkey,* 39) *Bezmi Alem Vakif University, School of Medicine, Department of Neurology, Istanbul, Turkey,* 40) *Mersin University School of Medicine, Department of Infectious Diseases and Clinical Microbiology, Mersin, Turkey,* 41) *Haydarpasa Numune Training and Research Hospital, Department of Neurology, Turkey,* 42) *Haydarpasa Numune Training and Research Hospital, Department of Radiology, Turkey,* 43) *Department of Neurology, Baghdad Teaching Hospital, Iraq,* 44) *Katip Celebi University School of Medicine, Department of Infectious Diseases and Clinical Microbiology, Izmir, Turkey,* 45) *Medeniyet University, Goztepe Training and Research Hospital, Department of Infectious Diseases and Clinical Microbiology, Istanbul, Turkey and* 46) *Cumhuriyet University School of Medicine, Department of Infectious Diseases and Clinical Microbiology, Sivas, Turkey*

Abstract

There have been many studies pertaining to the management of herpetic meningoencephalitis (HME), but the majority of them have focussed on virologically unconfirmed cases or included only small sample sizes. We have conducted a multicentre study aimed at providing

Clin Microbiol Infect 2016; **22:** 568.e9–568.e17

http://dx.doi.org/10.1016/j.cmi.2016.03.027

management strategies for HME. Overall, 501 adult patients with PCR-proven HME were included retrospectively from 35 referral centres in 10 countries; 496 patients were found to be eligible for the analysis. Cerebrospinal fluid (CSF) analysis using a PCR assay yielded herpes simplex virus (HSV)-1 DNA in 351 patients (70.8%), HSV-2 DNA in 83 patients (16.7%) and undefined HSV DNA type in 62 patients (12.5%). A total of 379 patients (76.4%) had at least one of the specified characteristics of encephalitis, and we placed these patients into the encephalitis presentation group. The remaining 117 patients (23.6%) had none of these findings, and these patients were placed in the nonencephalitis presentation group. Abnormalities suggestive of encephalitis were detected in magnetic resonance imaging (MRI) in 83.9% of the patients and in electroencephalography (EEG) in 91.0% of patients in the encephalitis presentation group. In the nonencephalitis presentation group, MRI and EEG data were suggestive of encephalitis in 33.3 and 61.9% of patients, respectively. However, the concomitant use of MRI and EEG indicated encephalitis in 96.3 and 87.5% of the cases with and without encephalitic clinical presentation, respectively. Considering the subtle nature of HME, CSF HSV PCR, EEG and MRI data should be collected for all patients with a central nervous system infection.

Keywords: Atypical presentation, encephalitis, herpes simplex virus, managing, meningitis
Original Submission: 30 September 2015; **Revised Submission:** 20 March 2016; **Accepted:** 26 March 2016
Editor: M. Paul
Article published online: 13 April 2016

Corresponding author: H. Erdem, GATA Enfeksiyon Hastalıkları ve Klinik Mikrobiyoloji A.D., Etlik, Ankara, Turkey
E-mail: hakanerdem1969@yahoo.com

Introduction

Herpetic meningoencephalitis (HME) is a rare but devastating infectious disease with a mortality rate of up to 70% in the absence of appropriate treatment [1]. Despite rapid diagnostic tests and antiviral therapies, HME is still associated with high rates of mortality and serious sequelae [2–6]. The most important parameters influencing a favourable clinical course are rapid diagnosis and early antiviral therapy initiated within 2 days of the onset of symptoms [6]. Current guidelines recommend the use of acyclovir in proven or suspected cases of encephalitis [7]. However, the question of what constitutes suspected encephalitis is unclear for the majority of cases in routine practice because the symptoms of meningitis and encephalitis generally overlap at the initial stages of both diseases [8,9]. As a result, a significant fraction of cases cannot be classified as suspected meningitis or suspected encephalitis by the examining clinician. Accordingly, the clinician may not predict a herpetic central nervous system (CNS) infection and may delay antiviral therapy. There are a large number of studies devoted to the management of HME, but most of them involve virologically unconfirmed cases, have small sample sizes or are literature reviews [4,10–17].

The goals of this retrospective, multicentre, multinational study included identifying the characteristic features of HME, determining the performance of diagnostic tests for the disease and developing an algorithm for an optimal clinical approach to reach the diagnosis of HME.

Materials and methods

Study design

This retrospective multicentre study was approved by the review board of the Dr Lütfi Kirdar Training and Research Hospital, Istanbul, Turkey.

The predictors of unfavourable outcome in HME cases have been previously published elsewhere [6].

Setting

Patients were drawn from 35 referral centres in ten countries, including Croatia, the Czech Republic, Denmark, Egypt, France, Iraq, Italy, Lebanon, Slovenia and Turkey.

Participants

This study included all consecutive hospitalized patients with HME between 2000 and 2013. The inclusion criteria comprised the presence of all of the following: only adult patients (>15 years of age); patients with positive cerebrospinal fluid (CSF) PCR results for herpes simplex virus (HSV)-1, HSV-2 or both in a patient with a CNS infection; and the unlikely presence of any other infectious disease of the brain or any neurologic disorder other than HSV infection.

The exclusion criteria comprised the presence of all of the following: paediatric patients and the presence of any other infectious or noninfectious disease of the brain.

Definitions

The definitions used in this study according to hospital admission clinical data were as follows.

Encephalitis presentation. Patients with at least one clinical finding compatible with encephalitis at hospital admission such as changes in conscious, disorientation, convulsions, amnesia, personality changes, speech disorders, hallucinations, abulia, history of unconsciousness or syncope, hemiparesis, dizziness, facial and hypoglossal cranial nerve palsies were classified in this category.

Nonencephalitis presentation. Patients without one of the clinical findings compatible with encephalitis at hospital admission noted above were classified in this category.

Unfavourable outcome. Unfavourable outcome was defined as patients who died of HME or survived with sequelae [6].

A questionnaire and a complementary Microsoft for Windows Excel file were distributed to the participating centres. Data on demographics, clinical and routine laboratory parameters, cranial radiologic imaging findings including magnetic resonance imaging (MRI), computed tomography (CT), brain electroencephalography (EEG), routine CSF analysis, CSF PCR for HSV, CSF serologic analysis for HSV, length of hospital stay, treatment and outcomes were collected. At the end of the study period, the centres submitted their data as an Excel document. These data were then merged to form the final database.

Statistical analysis

All the patients diagnosed as HME were classified into two groups: encephalitis presentation and nonencephalitis presentation. The data analysis was conducted in SPSS 16.0 software (IBM SPSS). We present descriptive statistics as frequencies, as percentages for categorical variables and as means ± standard deviations and medians (interquartile range (IQR)) for continuous variables according to the results of a normality test (the one-sample Kolmogorov-Smirnov test). For group comparisons, we used the chi-square test and Fisher's exact test for the categorical variables; for numerical variables, we used Student's *t* test for parametric data and the Mann-Whitney *U* test for nonparametric data. All of the tests were two tailed, and we assigned statistical significance to p values less than 0.05.

Results

A total of 501 HME patients were included retrospectively. Of these patients, five patients were excluded as a result of missing CSF PCR data; 496 patients were enrolled onto the study. The median (IQR) age of the patients was 50.5 (33.3–63.0) years. Of the study group, 266 patients (53.6%) were women. The patients were initially admitted to the departments of infectious diseases (*n* = 326, 65.7%), neurology (*n* = 80, 16.1%), internal medicine (*n* = 62, 12.5%), intensive care unit (*n* = 24, 4.8%) and other departments (*n* = 4, 0.8%).

Clinical presentation

A total of 379 patients (76.4%) presented at least one of the symptoms associated with an initial diagnosis of encephalitis presentation (Table 1). The remaining 117 patients (23.6%) exhibited none of these findings. The primary symptoms that led the clinician to perform lumbar puncture (LP) in these 117 patients are presented in Table 1. In this subgroup of cases, headache, neck stiffness, and Kernig and Brudziński signs were significantly more frequent than the encephalitis presentation group (p <0.05 for all comparisons). In contrast, fever was significantly less frequent compared to the number observed in the encephalitis presentation group (p <0.0001; Table 1). Only two of 117 patients had immunosuppressive conditions, which

TABLE 1. Initial clinical, radiologic imaging and EEG findings at presentation

Variable	Encephalitis presentation (*n* = 379)	Nonencephalitis presentation (*n* = 117)	p
Encephalitis symptoms		NA	NA
Changes in consciousness	304 (80.2)		
Disorientation	221 (58.3)		
Personality changes	123 (32.4)		
Speech disorders	109 (28.8)		
Convulsion	98 (25.9)		
Amnesia	98 (25.9)		
Hallucinations	23 (6.1)		
Abulia	16 (4.2)		
History of unconsciousness[a]	13 (3.4)		
Hemiparesis	6 (1.6)		
History of syncope	3 (0.8)		
Dizziness	1 (0.3)		
Facial and hypoglossal cranial nerve palsy	1 (0.3)		
Nonspecific CNS infection symptoms and signs			
Fever (temperature ≥38°C)	316 (83.4)	73 (62.4)	<0.0001
Headache	227 (59.9)	109 (93.2)	<0.0001
Neck stiffness	106 (27.9)	59 (50.4)	<0.0001
Kernig sign	25 (6.6)	18 (15.4)	0.003
Brudziński sign	20 (5.3)	12 (10.3)	0.055
Abnormal radiologic imaging and EEG findings (*n* = 496)			
MRI	225/268 (83.9)	21/63 (33.3)	<0.0001
CT	135/312 (43.3)	18/81 (22.2)	0.0005
EEG	223/245 (91.0)	13/21 (61.9)	0.0008
MRI or EEG[b]	293/326 (89.9)	29/69 (42.0)	<0.0001
MRI and EEG[b]	182/189 (96.3)	14/16 (87.5)	0.149

Data are presented as *n* (%) or *n*/*N* (%).
CNS, central nervous system; CT, computed tomography; EEG, electroencephalogram; MRI, magnetic resonance imaging; NA, not applicable.
[a]History of unconsciousness not perceivable at hospital admission.
[b]Findings indicating encephalitis by either MRI or EEG.

may explain the absence of encephalitis findings (data not shown).

The comparisons of initial abnormal radiologic imaging and EEG findings between the two groups are presented in Table 1. The number of abnormal findings on MRI, EEG, CT and one of MRI or EEG was found to be statistically more common in the encephalitis group than the nonencephalitis group (p <0.05 for all comparisons). However, the number of abnormal findings on the concomitant use of EEG and MRI was found to be the identical between the two groups (p 0.149).

LP, CSF and routine laboratory analyses

The median time to performing an initial LP after hospitalization was 6.0 (2.0–24.0) hours for 440 patients. The results of initial CSF analyses at various times after hospitalization are presented in Table 2. The comparisons of routine laboratory characteristics at admission between the groups are presented in Table 3. In the encephalitis presentation group, the median percentage of blood neutrophils, blood C-reactive protein level, erythrocyte sedimentation rate, serum aspartate aminotransferase level, blood urea nitrogen and creatinine levels were found to be significantly higher than the nonencephalitis presentation group (p <0.05 for all comparisons). In contrast, both the median serum sodium concentration and the number of patients with hypernatraemia were significantly lower in this group of the patients (p <0.0001 and 0.009, respectively). The mean ± standard deviation serum albumin concentration was also found to be significantly lower in this group of the patients (3.7 ± 0.8 vs. 4.1 ± 0.6 g/dL; p 0.001).

Molecular analysis

Real-time PCR was performed in 365 patients (73.6%), and two patients underwent nested PCR. All of the PCR data derived from samples from the initial LPs used as an inclusion criterion for this study. We were unable to retrieve the PCR method used from the files of 129 patients. The median (IQR) duration between admission and obtaining a positive CSF PCR result for HSV was 72.0 (42.0–136.5) hours. In 112 patients (12.5%), CSF PCR for HSV of an undefined type was positive; 50 of these patients were later found to be positive for HSV-1. Therefore, 351 patients (70.8%) were positive for HSV-1 and 83 patients (16.7%) were positive for HSV-2; positivity with CSF PCR for HSV of an undefined type was established in 62 cases. Further, the number of patients with positive CSF PCR results for HSV-1 was significantly higher in the encephalitis presentation group than the nonencephalitis presentation group (298 (78.6%) vs.

TABLE 2. Results of CSF analyses obtained from initial lumbar puncture of 440 patients after hospitalization

Variable	0 to 24 hours (*n* = 359)	25 to 72 hours (*n* = 59)	73 hours to 10 days (*n* = 16)	11 to 22 days (*n* = 6)
High opening pressure	65/228 (28.6)	3/19 (15.8)	2/6 (33.3)	0/3 (0)
Appearance				
Clear	289/329 (87.8)	42/46 (91.3)	10/13 (76.9)	5/5 (100.0)
Turbid	24/329 (7.3)	2/46 (4.3)	1/13 (7.7)	0
Bloody	13/329 (3.9)	1/46 (2.2)	1/13 (7.7)	0
Xanthochromia	2/329 (0.6)	1/46 (2.2)	1/13 (7.7)	0
Leucocyte count (1/mm^3)				
0–4	14/352 (3.9)	2/58 (3.4)	5/14 (35.7)	1/6 (16.7)
5–49	66/352 (18.8)	19/58 (32.8)	2/14 (14.3)	2/6 (33.3)
50–99	40/352 (11.4)	13/58 (22.4)	2/14 (14.3)	3/6 (50.0)
100–499	176/352 (50.0)	17/58 (29.3)	5/14 (35.7)	0
500–999	31/352 (8.8)	5/58 (8.6)	0	0
≥1000	25/352 (7.1)	2/58 (3.4)	0	0
Neutrophil count (1/mm^3)				
0–4	76/299 (25.4)	21/41 (51.1)	7/11 (63.6)	1/3 (33.3)
5–49	128/299 (42.8)	18/41 (43.9)	4/11 (36.4)	2/3 (66.7)
50–99	57/299 (19.1)	2/41 (4.9)	0	0
100–499	30/299 (10.0)	0	0	0
500–999	5/299 (1.7)	0	0	0
≥1000	3/299 (1.0)	0	0	0
Lymphocyte count (1/mm^3)				
0–4	10/324 (3.1)	1/48 (2.1)	3/12 (25.0)	0
5–49	64/324 (19.8)	18/48 (37.5)	2/12 (16.7)	3/4 (75.0)
50–99	45/324 (13.9)	24/48 (50.0)	2/12 (16.7)	1/4 (25.0)
100–499	160/324 (49.4)	4/48 (8.3)	5/12 (41.7)	0
500–999	29/324 (8.9)	1/48 (2.1)	0	0
≥1000	16/324 (4.9)	0	0	0
Erythrocyte count (1/mm^3)				
0–50	215/293 (73.4)	27/46 (58.7)	9/13 (69.2)	3/4 (75.0)
51–250	32/293 (10.9)	8/46 (17.4)	1/13 (7.7)	1/4 (25.0)
251–500	14/293 (4.8)	3/46 (6.5)	0	0
>500	20/293 (6.8)	7/46 (15.2)	2/13 (15.4)	0
Hemorrhagic appearance	12/293 (4.1)	1/46 (2.2)	1/13 (7.7)	0
Hypoglycorrhachia (CSF/blood glucose ratio <0.60)	132/263 (50.2)	14/23 (60.9)	9/23 (39.1)	3/3 (100.0)
High protein level (>45 mg/dL)	263/339 (77.6)	48/55 (87.3)	13/15 (86.7)	5/5 (100.0)
Lactate (mmol/L), median (IQR)	2.8 (2.3–3.6) (*n* = 18)	2.7 (2.0–3.4) (*n* = 9)	6.3 (3.1–7.4) (*n* = 3)	0
LDH (U/mL), median (IQR)	50 (24.8–59.3) (*n* = 10)	ND (*n* = 2)	0	0

Data are presented as *n* (%) or *n*/*N* (%) unless otherwise indicated.
CSF, cerebrospinal fluid; IQR, interquartile range; LDH, lactate dehydrogenase; ND, not determined.

TABLE 3. Comparison of admission routine laboratory characteristics

Variable	Total (n = 496)	Encephalitis presentation (n = 379)	Nonencephalitis presentation (n = 117)	p	Normal
Hemoglobin (mg/dL) (mean ± SD)	13.1 ± 1.7	13.2 ± 1.7	13.1 ± 1.6	0.560	14–18 (male), 12–16 (female)
Leucocyte (× 103/mm^3), median (IQR)	9.7 (7.5–13.0)	10.0 (7.6–13.4)	8.8 (7.1–11.3)	0.627	4–11
Neutrophil (%), median (IQR)	71 (60–82)	75 (63–83)	65 (55–75)	<0.0001	40–75
Platelet (× 103/mm^3), median (IQR)	209 (171–261)	205 (168–253)	223 (176–274)	0.067	150–450
CRP (mg/L), median (IQR)	0.8 (0.2–2.0)	0.8 (0.3–3.0)	0.5 (0.2–1.0)	0.015	0–8.0
ESR (mm/hour), median (IQR)	19.0 (11.8–30.0)	20.0 (12.0–34.5)	16.0 (11.0–20.0)	0.014	≤15 (male), ≤20 (female)
Glucose (mg/dL), median (IQR)	105.0 (90.0–129.0)	112.5 (91.3–138.8)	95.0 (87.0–104.5)	<0.0001	70.0–110.0
AST (IU/L), median (IQR)	27 (19–37)	27 (19–40)	24 (17–33)	0.013	15–41
ALT (IU/L), median (IQR)	23 (16–32)	22 (16–31)	25 (16–34)	0.404	17–63
Serum sodium (mEq/L)					
Median (IQR) value	136 (131–139)	135 (130–138)	139 (136–141)	<0.0001	135–150
Hyponatremia, n (%)	178 (44.2)	144 (37.9)	34 (29.1)	0.078	
Hypernatraemia, n (%)	7 (1.7)	2 (0.5)	5 (4.3)	0.009	
BUN (mg/dL), median (IQR)	17.0 (11.8–26.0)	19.6 (12.7–33.5)	13.0 (10.6–17.0)	<0.0001	7.0–20.0
Creatinine (mg/dL), median (IQR)	0.8 (0.7–1.0)	0.9 (0.7–1.0)	0.8 (0.6–0.9)	0.001	0.7–1.2
Albumin (g/dL), mean ± SD	3.8 ± 0.7	3.7 ± 0.8	4.1 ± 0.6	0.001	3.5–5.0

ALT, alanine aminotransferase; AST, aspartate aminotransferase; BUN, blood urea nitrogen; CRP, C-reactive protein; ESR, erythrocyte sedimentation rate; IQR, interquartile range.

53 (45.3%); p <0.0001). Conversely, the number of patients with positive CSF PCR results for HSV-2 was significantly lower in this group of patients than the nonencephalitis presentation group (32 (8.5%) vs. 51 (43.6%); p <0.0001) (data not shown). A quantitative analysis of HSV DNA in CSF was available for 41 patients (8.3%). The median (IQR) HSV DNA load in those CSF samples was 1.8×10^4 (1.9×10^3–1.2×10^5) copies/mL (data not shown). A follow-up LP was performed in 167 patients after the initial LP a median (IQR) of 7.0 (3.0–14.0) days later. Of the 167 patients with a follow-up LP, HSV DNA was repeated in 108 of them, and it was found to be as positive in 57 (52.7%). When we considered the second LP, we had treatment data for 54 of 57 PCR-positive cases and 49 of 51 PCR-negative cases. The acyclovir treatment duration did not differ between these two groups (PCR positive (n = 54), median (IQR) 21.0 (19.5–21.5) days; PCR negative (n = 49), median (IQR) 21.0 (14.0–21.0) days; p 0.255) (data not shown).

Serology

Blood HSV IgM was positive in 76.5% of the patients tested (52/68), and blood HSV IgG was positive in 13.2% of the patients tested (7/53). For the CSF analysis, HSV IgM was positive in 39.6% of patients tested (19/48), and HSV IgG was positive in 57.4% of patients tested (27/47). A primary CNS HSV infection (negative CSF IgG and positive CSF IgM) was not observed in any of the 20 (41.7%) of 48 CSF IgG negative patients (data not shown).

Cranial MRI and CT

MRI was performed in 331 (66.7%) of 496 patients. The median (IQR) time between performing MRI and hospital admission was 65.0 (24.0–96.0) hours. Parenchymal involvement was reported in a cohort of 202 patients in 42 patients (20.1%) with cortical changes, 36 patients (17.8%) with white matter changes and involvement of both areas in 124 patients (61.4%). Seventy-two patients had repeated MRI scans after a median (IQR) of 16.0 (10.0–30.8) days after the first scan, and we could provide radiologic review reports in 57 cases with repeated MRI. Repeated MRI revealed regression in 56.1% (32/57) patients and progression in 44.0% (25/57) patients. The sites of involvement in the MRI are presented in Table 4. Cranial CT was performed in 393 patients. The median (IQR) time between CT and admission was 25.0 (9.0–72.0) hours. The abnormal findings that we detected in the CT and MRI data are presented in Table 5.

EEG testing

EEG was performed in 266 patients (53.6%) after a median (IQR) of 3.0 (1.5–5.5) days of hospitalization. EEG abnormalities related to encephalitis were detected in 236 patients (88.7%). The MRI and EEG findings of the patients are presented in Table 6. These findings included nonspecific, diffuse, high-amplitude slow waves in 106 (44.9%) of 236 patients, lateralized/localized slow waves in 65 (27.5%) of 236 patients, temporal lobe spike-and-wave activity in 64 (27.1%) of 236 patients, periodic lateralized epileptiform discharge (PLED) in 55 (23.3%) of 236 patients (right PLED in 27/55 (49.1%) patients, left PLED in 17/55 (30.1%) patients and bilateral PLED in 11/55 (20.0%) patients) and other abnormalities in 40 (16.9%) of 236 patients (data not shown).

Follow-up EEG was obtained in 80 (30.0%) of 266 patients after a median (IQR) of 10.5 (2.8–21.0) days of hospitalization. EEG abnormalities persisted in 62 (77.5%) of 80 patients. These abnormalities included nonspecific, diffuse, high-amplitude slow waves in 26 (41.9%) of 62 patients, lateralized/localized slow waves in 21 (33.9%) of 62 patients, temporal lobe spike-and-wave activity in 14 (22.6%) of 62 patients, PLED in 9 (14.5%) of 62 patients (right PLED, left PLED and bilateral PLED in 2/9 (22.2%), 4/9 (44.4%) and 3/9 (33.3%) patients, respectively) and

TABLE 4. Cerebral involvement sites in initial and follow-up cranial MRI scans

Characteristic	Temporal	Frontal	Parietal	Occipital	Cerebellum	Other	No involvement
Initial MRI (n = 331)[a]							
Left hemisphere	82 (24.8)	26 (7.9)	17 (5.1)	6 (1.8)		29 (8.8)[b]	85 (25.7)
Right hemisphere	75 (22.7)	20 (6.0)	7 (2.1)	1 (0.3)			
Bilateral	73 (22.1)	21 (6.3)	6 (1.8)	4 (1.2)	3 (0.9)		
Follow-up MRI (n = 64)							
Left hemisphere	16 (25.0)	4 (6.3)	1 (1.6)		1 (1.6)	8 (12.5)[c]	15 (23.4)
Right hemisphere	19 (29.7)	7 (10.9)		1 (1.6)			
Bilateral	18 (28.1)	4 (6.2)	2 (3.1)	1 (1.6)			

Data are presented as n (%).
MRI, magnetic resonance imaging.
[a]Number of MRI reports with comments in the hospital records.
[b]Insula 6, diffuse 5, limbic system 4, basal ganglia 3, thalamus 2, hippocampus 2, pons 1, mesencephalon 1, corpus callosum 1, uncus 1, centrum semiovale 1, corona radiata 1, lacunar 1.
[c]Insula 2, diffuse 2, limbic system 1, basal ganglia 1, pons 1, limbic system 1.

TABLE 5. Abnormal findings on cranial CT and MRI scans

Characteristic	CT (n = 393)	MRI (n = 331)	Concomitant use of CT and MRI (n = 270)
Total abnormal findings	153/393 (38.9)	246/331 (74.3)	97/270 (35.9)
Density changes	75/153 (49.0)	167/246 (67.9)	42/97 (43.3)
Edoema	49/153 (32.0)	93/246 (37.8)	21/97 (21.6)
Haemorrhage	17/153 (11.1)	28/246 (11.4)	5/97 (5.2)
Infarction	18/153 (11.8)	24/246 (9.8)	4/97 (4.1)
Cerebral thrombosis	3/153 (1.9)	0	0
Other[a]	0	2/246 (0.8)	0

Data are presented as n/N (%).
CT, computed tomography; MRI, magnetic resonance imaging.
[a]Right facial nerve inflammation 1, basal ganglion infarction 1.

other abnormalities in 17 (27.4%) of 62 patients (data not shown).

Outcome analysis and management algorithm

The median (IQR) length of hospital stay was 19 (11.5–26.0) days among the patients; the median (IQR) length of hospital stay among the encephalitis presentation group was significantly longer than that of the nonencephalitis group (21.0 (15.0–30.0) vs. 10.0 (5.0–15.0) days; p <0.0001). Furthermore, in this group of patients, the number of experienced unfavourable outcomes (deaths and survival with sequelae) were significantly more frequent than that of the nonencephalitis presentation group at the end of the antiviral treatment (41 (10.8) vs. 3 (2.6); p 0.003 and 182 (48.0) vs. 25 (21.4) p <0.0001, respectively) (data not shown). Considering these findings, an algorithmic flowchart is presented in Fig. 1 to aid clinicians in deciding when to consider encephalitis or what kind of algorithm should be followed at the start of antiviral therapy targeting HME. We note that the contraindications of LP should be taken into consideration according to the general concepts reported elsewhere [5].

Discussion

Changes in consciousness, disorientation, language and behavioural abnormalities, cognition and memory impairment, focal neurologic signs and seizures have thus far been the primary clinical indicators of HSV encephalitis. However, in addition to the encephalitic component, many patients may exhibit meningeal inflammation. As a result, 'meningoencephalitis' is a frequently used term for the coexistence of two conditions [6]. In our study, three-fourths of patients exhibited at least one of the specified characteristics of encephalitis. However, none of these manifestations was reported or noted in a quarter of our patients. The patients in the nonencephalitis presentation group were evaluated for headache, fever and neck stiffness indicating the potential presence of dominating meningitis. Accordingly, one-third of the cases in the nonencephalitis presentation

TABLE 6. Initial magnetic resonance MRI and EEG findings

	MRI		EEG	
	Normal	Abnormal	Normal	Abnormal
MRI (n = 331)				
Normal	85/331 (25.7%)	—	9/26 (34.6%)	17/26 (65.4%)
Abnormal	—	246/331 (74.3%)	17/178 (9.6%)	161/178 (90.4%)
EEG (n = 266)				
Normal	16/25 (64.0%)	9/25 (36.0%)	30/266 (11.3%)	—
Abnormal	18/179 (10.1%)	161/179 (89.9%)	—	236/266 (88.7%)

Data are presented as n/N (%).
EEG, electroencephalogram; MRI, magnetic resonance imaging.

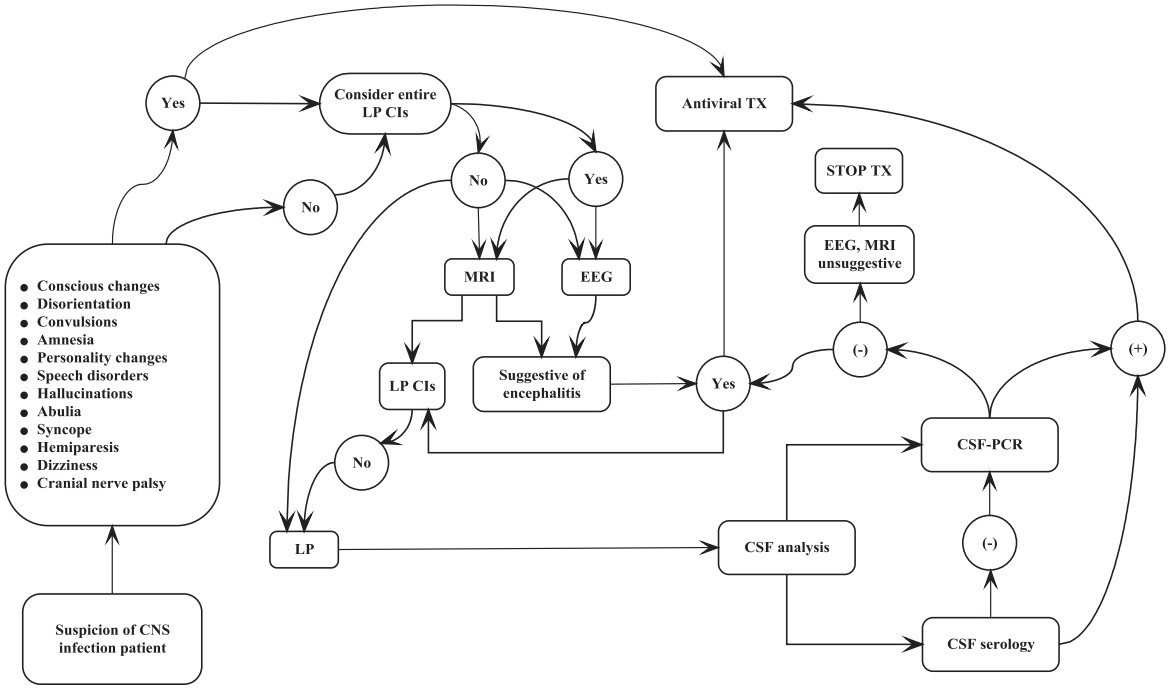

FIG. 1. Diagnostic algorithm. CI, contraindication; CNS, central nervous system; CSF, cerebrospinal fluid; EEG, electroencephalogram; MRI, magnetic resonance imaging; PCR, polymerase chain reaction; Tx, treatment.

group were categorized as meningitis; 17.5% of these patients were found to be infected with HSV-2, while HSV-1 dominated. Atypical presentation of herpetic encephalitis has been reported in mild cases and in immunocompromised patients, including cases of pregnancy [18–20]. However, only two patients without signs of encephalitis demonstrated potential causes of immunosuppression in this study. Because the encephalitis patients may present in early clinical stages, and because of the silent nature of HME, clinicians must not overlook this tricky clinical presentation at diagnosis [21].

In HME, xanthochromia caused by red blood cell breakdown, pleocytosis, mildly or moderately increased protein levels, normal or slightly altered glucose or lactate concentrations have been reported in CSF analyses [7,22]. Our data revealed similar findings. However, the confirmation of the disease relies on molecular studies in which HSV-1 has been reported to account for 90% of HME cases [5]. Although HSV-2 has been reported to cause meningeal infections, many studies have suggested that HSV-2 not only is a major cause of aseptic meningitis but may also cause encephalitis [5,23]. Along these lines, we found that CSF PCR results were positive for HSV-2 in 32 patients (8.4%) in the encephalitis presentation group. The detection of HSV DNA in CSF has a sensitivity of >95% and a specificity of >99% [7,22,24,25]. Predictors resulting in false negativity such as analysing CSF samples obtained early in the progression of the disease or bloody specimens have been known to exist. Hence, PCR reanalysis is recommended from CSF samples obtained after 3 to 7 days in probable cases [7,26]. On the other hand, our data revealed that HSV DNA analysed in the follow-up CSF samples obtained a median of 7 days after the initial LP was undetectable in about half of the cases and seemingly cleared from the CSF with treatment. We were unable to detect a difference in the duration of therapy between PCR-positive and -negative findings according to our analysis of CSF samples obtained from the second LP. Therefore, the initiation of empirical antivirals may obscure a molecular diagnosis when LP is delayed. Although the detection of specific IgG antibodies in the CSF has a diagnostic value similar to positive PCR, these antibodies may be unavailable in the first CSF sample and typically show enhancement after 10 to 12 days. HSV-specific IgM antibody detection is typically reported less often in HSV encephalitis [22]. According to the data obtained from the entire study, 40 and 57% of the cases were positive for IgM and IgG from the CSF, respectively, which indicates serology as a nonnegligible diagnostic modality. Serology can accordingly be used as a complement to molecular testing. In addition, blood HSV IgM, which was positive in three quarters of our cases, may be a surrogate marker in the diagnosis of a CNS infection due to HSV.

MRI is the most valuable radiologic technique for encephalitis because it enables earlier detection of the disease [7,27]. Two previous study suggested that MRI scan revealed brain

involvement in 90 and 95% of HSV encephalitis patients diagnosed by CSF PCR, respectively [28,29]. In this study, MRI data indicated encephalitis in three quarters of all patients; signal changes and oedema were the most frequent findings. MRI obtained within 2 days of hospital admission was reported to be abnormal in approximately 90% of patients in previous studies [30–32]; the median time until the first MRI scan was 65 hours in our cohort of patients. In the initial MRI testing, temporal lobe involvement was observed in 70% of cases; frontal lesions were documented in 20%. Parietal, occipital and cerebellar involvements were infrequently seen in our patients. The extent of MRI abnormality in HSV encephalitis is not correlated with the course of the disease [5]; repeat MRI testing may not be trustworthy for evaluating therapeutic efficacy. According to results of this study, 25 (4%) of 57 patients with follow-up MRI scans obtained after a median of 16 days of treatment showed progression. However, one limitation of this study is that we do not know exactly whether this progression was due to clinical deterioration or due to the lack of a correlation between the neuroimaging and the clinical presentation. Furthermore, because the study period spanned a 14-year period over which the infrastructure at the participating hospitals improved significantly, some of the centres experienced temporary problems in MRI or EEG collection, particularly in the early stages of the study period. As a result, these centres were unable to provide these data. CT is another radiodiagnostic procedure that is inferior to MRI; it can be used when MRI testing is unavailable. In relatively small case series, initial testing with CT was reported to be normal in 21–33% of HSV encephalitis patients [28,33]. However, we found that 60% of our patients had normal CT findings.

EEG has been reported to be more sensitive at the acute stage of encephalitis. Abnormalities such as unilateral or bilateral periodic sharp waves or attenuation of amplitude, focal or generalized slow waves or epileptiform discharges, or electrical seizures can be observed. Focal or lateralized EEG abnormalities are highly indicative of herpetic encephalitis in particular, although EEG has been reported to be less specific than radiologic assessments [34–36]. In this study, nonspecific, diffuse, high-amplitude and lateralized/localized slow waves were the most frequent EEG findings, followed by temporal lobe slow waves and PLEDs. Further, EEG data also suggested encephalitis in 91% of all cases and 62% of nonencephalitis presentation patients. In the follow-up EEG testing obtained a median of 10 days after hospitalization, four fifths of patients still exhibited abnormal EEG findings related to encephalitis. Therefore, although we cannot provide specific data, EEG seems to be a valuable technique for diagnosing HSV encephalitis.

In this study, MRI and EEG data revealed the presence of encephalitis in 33 and 62% of cases without encephalitic clinical presentation, respectively. Concomitant use of MRI and EEG indicated encephalitis in the majority of cases, and there was no significant difference between the encephalitis presentation and the nonencephalitis presentation groups, which motivates the urgent use of both tests in a patient with CNS infection. The patients without positive findings with the concomitant use of these techniques may have gone undetected as a result of testing sensitivity issues or relatively local and insignificant involvements. A delay in establishing an effective antiviral treatment of more than 2 days significantly increases the risk of unfavourable outcome [6].

Our study is the largest case series ever reported with CSF PCR-positive patients for HSV. Although a major limitation of this study is its retrospective design, it is very difficult to provide such a cohort prospectively. After a careful physical examination, encephalitis presentation patients with compatible findings should receive antiviral treatment in a timely manner. LP and molecular analysis should accordingly be performed whenever contraindications are eliminated in patient with suspected CNS infection. Considering the significantly benign nature of the clinical table for patients in the nonencephalitis presentation group, and owing to the improved laboratory infrastructures of the hospitals with easy access to these tests in many parts of the world, suspending antiviral therapy until positive radiologic, EEG or molecular results are obtained seems rational for this subgroup of patients. This approach will likely prevent excessive antiviral use for the entire cohort of CNS infections other than HSV disease.

In conclusion, CSF PCR analysis for HSV should be conducted for all patients with a CNS infection, considering the subtle nature of HME. Furthermore, the combined use of MRI and EEG (or CT if MRI is not feasible) appears to be advantageous. If a patient has compatible clinical, MRI and EEG findings with HME, a negative CSF PCR test should be repeated later. In addition, a negative CSF serology alone cannot rule out HME, and blood HSV IgM may provide clues about the disease.

Acknowledgements

The present study is an European Society of Clinical Microbiology and Infectious Diseases (ESCMID) Study Group for Infectious Diseases of the Brain (ESGIB) and Infectious Diseases International Research Initiative (ID-IRI) cooperative study.

Transparency declaration

All authors report no conflicts of interest relevant to this article.

References

[1] Whitley RJ, Soong SJ, Dolin R, Galasso GJ, Ch'ien LT, Alford CA. Adenine arabinoside therapy of biopsy-proved herpes simplex encephalitis. National Institute of Allergy and Infectious Diseases collaborative antiviral study. N Engl J Med 1977;297:289–94.

[2] Stahl JP, Mailles A, De Broucker T, Steering C, Investigators G. Herpes simplex encephalitis and management of acyclovir in encephalitis patients in France. Epidemiol Infect 2012;140:372–81.

[3] Pewter SM, Williams WH, Haslam C, Kay JM. Neuropsychological and psychiatric profiles in acute encephalitis in adults. Neuropsychol Rehabil 2007;17:478–505.

[4] Dagsdóttir HM, Sigurðardóttir B, Gottfreðsson M, Kristjánsson M, Löve A, Baldvinsdóttir GE, et al. Herpes simplex encephalitis in Iceland, 1987–2011. Springerplus 2014;3:524.

[5] Solomon T, Michael BD, Smith PE, Sanderson F, Davies NW, Hart IJ, et al. Management of suspected viral encephalitis in adults—Association of British Neurologists and British Infection Association national guidelines. J Infect 2012;64:347–73.

[6] Erdem H, Cag Y, Ozturk-Engin D, Defres S, Kaya S, Larsen L, et al. Results of a multinational study suggest the need for rapid diagnosis and early antiviral treatment at the onset of herpetic meningoencephalitis. Antimicrob Agents Chemother 2015;59:3084–9.

[7] Tunkel AR, Glaser CA, Bloch KC, Sejvar JJ, Marra CM, Roos KL, et al. The management of encephalitis: clinical practice guidelines by the Infectious Diseases Society of America. Clin Infect Dis 2008;47:303–27.

[8] Beckham JD, Tyler KL. Encephalitis. In: Bennett JE, Dolin R, Blaser MJ, editors. Mandell, Douglass, and Bennett's principles and practice of infectious diseases. 8th ed. Philadelphia: Elsevier; 2015. p. 1144–63.

[9] Tunkel AR, van de Beek D, Scheld MW. Acute meningitis. In: Mandell, Douglass, and Bennett's principles and practice of infectious diseases. 8th ed. Philadelphia: Elsevier; 2015. p. 1097–137.

[10] Child N, Croxson MC, Rahnama F, Anderson NE. A retrospective review of acute encephalitis in adults in Auckland over a five-year period (2005–2009). J Clin Neurosci 2012;19:1483–5.

[11] Modi A, Atam V, Jain N, Gutch M, Verma R. The etiological diagnosis and outcome in patients of acute febrile encephalopathy: a prospective observational study at tertiary care center. Neurol India 2012;60:168–73.

[12] Mailles A, Stahl JP, Steering Committee and Investigators Group. Infectious encephalitis in France in 2007: a national prospective study. Clin Infect Dis 2009;49:1838–47.

[13] Bell DJ, Suckling R, Rothburn MM, Blanchard T, Stoeter D, Michael B, et al. Management of suspected herpes simplex virus encephalitis in adults in a UK teaching hospital. Clin Med 2009;9:231–5.

[14] Cinque P, Cleator GM, Weber T, Monteyne P, Sindic CJ, van Loon AM. The role of laboratory investigation in the diagnosis and management of patients with suspected herpes simplex encephalitis: a consensus report. The EU Concerted Action on Virus Meningitis and Encephalitis. J Neurol Neurosurg Psychiatry 1996;61:339–45.

[15] Whitley RJ, Alford CA, Hirsch MS, Schooley RT, Luby JP, Aoki FY, et al. Vidarabine versus acyclovir therapy in herpes simplex encephalitis. N Engl J Med 1986;314:144–9.

[16] Riera-Mestre A, Gubieras L, Martinez-Yelamos S, Cabellos C, Fernandez-Viladrich P. Adult herpes simplex encephalitis: fifteen years' experience. Enferm Infecc Microbiol Clin 2009;27:143–7.

[17] Poissy J, Champenois K, Dewilde A, Melliez H, Georges H, Senneville E, et al. Impact of herpes simplex virus load and red blood cells in cerebrospinal fluid upon herpes simplex meningo-encephalitis outcome. BMC Infect Dis 2012;12:356.

[18] Tan IL, McArthur JC, Venkatesan A, Nath A. Atypical manifestations and poor outcome of herpes simplex encephalitis in the immunocompromised. Neurology 2012;79:2125–32.

[19] Klapper PE, Cleator GM, Longson M. Mild forms of herpes encephalitis. J Neurol Neurosurg Psychiatry 1984;47:1247–50.

[20] Sellner J, Buonomano R, Nedeltchev K, Findling O, Schroth G, Surbek DV, et al. A case of maternal herpes simplex virus encephalitis during late pregnancy. Nat Clin Pract Neurol 2009;5:51–6.

[21] Robinson R. Large international study assesses factors associated with outcomes for herpetic meningoencephalitis confirms the importance of rapid treatment. In: Ringel SP, editor. Neurology today. New York: American Academy of Neurology; 2015. p. 11.

[22] Machado Ldos R, Livramento JA, Vianna LS. Cerebrospinal fluid analysis in infectious diseases of the nervous system: when to ask, what to ask, what to expect. Arq Neuropsiquiatr 2013;71:693–8.

[23] Whitley RJ, Gnann JW. Viral encephalitis: familiar infections and emerging pathogens. Lancet 2002;359:507–13.

[24] Sauerbrei A, Wutzler P. Laboratory diagnosis of central nervous system infections caused by herpesviruses. J Clin Virol 2002;25(Suppl. 1):S45–51.

[25] Bhullar SS, Chandak NH, Purohit HJ, Taori GM, Daginawala HF, Kashyap RS. Determination of viral load by quantitative real-time PCR in herpes simplex encephalitis patients. Intervirology 2014;57:1–7.

[26] Aurelius E, Johansson B, Skoldenberg B, Staland A, Forsgren M. Rapid diagnosis of herpes simplex encephalitis by nested polymerase chain reaction assay of cerebrospinal fluid. Lancet 1991;337:189–92.

[27] Schroth G, Kretzschmar K, Gawehn J, Voigt K. Advantage of magnetic resonance imaging in the diagnosis of cerebral infections. Neuroradiology 1987;29:120–6.

[28] Domingues RB, Tsanaclis AM, Pannuti CS, Mayo MS, Lakeman FD. Evaluation of the range of clinical presentations of herpes simplex encephalitis by using polymerase chain reaction assay of cerebrospinal fluid samples. Clin Infect Dis 1997;25:86–91.

[29] Sili U, Kaya A, Mert A, HSV Encephalitis Study Group. Herpes simplex virus encephalitis: clinical manifestations, diagnosis and outcome in 106 adult patients. J Clin Virol 2014;60:112–8.

[30] Hollinger P, Matter L, Sturzenegger M. Normal MRI findings in herpes simplex virus encephalitis. J Neurol 2000;247:799–801.

[31] Tien RD, Felsberg GJ, Osumi AK. Herpesvirus infections of the CNS: MR findings. AJR Am J Roentgenol 1993;161:167–76.

[32] Domingues RB, Fink MC, Tsanaclis AM, de Castro CC, Cerri GG, Mayo MS, et al. Diagnosis of herpes simplex encephalitis by magnetic resonance imaging and polymerase chain reaction assay of cerebrospinal fluid. J Neurol Sci 1998;157:148–53.

[33] Raschilas F, Wolff M, Delatour F, Chaffaut C, De Broucker T, Chevret S, et al. Outcome of and prognostic factors for herpes simplex encephalitis in adult patients: results of a multicenter study. Clin Infect Dis 2002;35:254–60.

[34] Steiner I, Budka H, Chaudhuri A, Koskiniemi M, Sainio K, Salonen O, et al. Viral meningoencephalitis: a review of diagnostic methods and guidelines for management. Eur J Neurol 2010;17. 999–e57.

[35] Lai CW, Gragasin ME. Electroencephalography in herpes simplex encephalitis. J Clin Neurophysiol 1988;5:87–103.

[36] Sutter R, Barnes B, Leyva A, Kaplan PW, Geocadin RG. Electroencephalographic sleep elements and outcome in acute encephalopathic patients: a 4-year cohort study. Eur J Neurol 2014;21:1268–75.