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Original Article

Determining the clinical characteristics and prognostic factors for the outcomes of Japanese encephalitis in adults: A multicenter study from southern Taiwan

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KEYWORDS
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Abstract *Background:* In Southeast Asia, Japanese encephalitis (JE) is an important cause of viral encephalitis which may cause severe neurological sequelae. JE affects mostly children; therefore, clinical presentations and prognosis of adult JE patients are seldom addressed. This

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Japanese encephalitis;
Outcome;
Prognostic factor

study aimed to describe the clinical characteristics and prognostic factors for the outcome of adult JE patients.
Methods: Medical records of adult JE patients with acute encephalitis syndrome during 2001—2018 from five medical centers in southern Taiwan were reviewed. Clinical characteristics, brain images, and prognostic factors for outcomes were analyzed. Patients were divided into the good outcome (GO) group and poor outcome (PO) group according to their Glasgow Coma Scale (GCS) scores (GCS >8 vs. ≤ 8) at discharge.
Results: Sixty-eight patients (men, 61.8%; median age, 50 years) were included. Summer is the epidemic season, and the number of cases peaked in June. The most common symptoms at initial presentation were altered consciousness and fever (both 94.1%), followed by headache (51.4%). The most commonly involved brain regions were thalamus (55.7%) and basal ganglion (37.7%). The median GCS score at nadir was 8, and the median time from onset to nadir was five days. Fifty-two patients were included in the GO group, while 16 were included in the PO group. On multivariate analysis, flaccidity, rigidity, and elevated CSF protein level were identified as independent prognostic factors for PO.
Conclusion: Initial clinical presentations of abnormal muscle tone including flaccidity, rigidity and high CSF protein levels are independent prognostic factors for PO in adult JE patients.

Introduction

Japanese encephalitis virus (JEV), along with other mosquito-borne flaviviruses such as Zika virus and dengue virus, belong to the family *Flaviviridae*.[1,2] JEV is commonly associated with transmission between amplifying vertebrate hosts, such as pigs and ardeid birds via *Culex tritaeniorhynchus*, the main transmission vector of JEV. Symptomatic Japanese encephalitis (JE) is an uncommon condition and occurs in approximately 1 in 250 patients with subclinical infections.[3] The initial symptoms of JE are mostly nonspecific flu-like prodrome, with some evolving to fever, altered sensorium, headache, and seizures.[3,4] The mortality rate in symptomatic patients is 20—30%, and severe neurological deficits occur in 30—50% of the survivors.[5] Therefore, early identification of the disease and prompt medical treatment are of utmost importance.[6]

JE is a major cause of viral encephalitis in South and East Asian countries, including Taiwan.[7,8] The global incidence rate of JE is 1.8 per 100,000 people. Approximately 67,900 cases of JE are reported annually and 75% of which occur in children under 14 years of age.[7] In Taiwan, JE vaccination has been included in the national immunization program since 1968. Since then, the JE-affected population in Taiwan shifted gradually from children to adults, and up to 94.8% of the JE patients are over 20 years old during 2000—2014.[8,9] Moreover, about 40% of all JE cases occur in southern Taiwan because of its tropical climate and thriving agricultural activity.[10]

Only a few studies have evaluated the prognostic factors of JE, and most were conducted among children.[11—13] Compared with adults, children who survive have poorer outcomes and frequently develop neurological deficits like dystonia.[14] Most studies focused on the outcomes of adults affected by JE are case reports or case series, and only a limited number of studies report the detailed clinical presentations of patients with JE or prognostic factors for mortality and neurological sequelae.[2,4,15] Herein, we aimed to conduct a retrospective study in five medical centers in southern Taiwan to describe the clinical features of adult JE patients and determine the prognostic factors for different outcomes.

Methods

Identification and definition of JE cases

JE has been a reportable disease in Taiwan since 1955. It is mandatory for clinicians to report all cases that meet the clinical criteria of JE to the Centers of Disease Control of Taiwan (Taiwan CDC).[16] Taiwan CDC developed an E/M-specific capture immunoglobulin M (IgM) and immunoglobulin G (IgG) enzyme-linked immunosorbent assay (E/M-specific IgM/IgG ELISA) for differentiation of JE and dengue fever in 1998. The detection method and its high sensitivity and specificity has been described previously.[8,17] Since 2001, acute-phase serum obtained within 7 days after onset of symptoms and all cerebrospinal fluid (CSF) samples collected from reported JE cases are subjected to real-time polymerase chain reaction for diagnosis, in addition to E/M-specific IgM/IgG ELISA and the virus isolation method.[17] This study implemented the World Health Organization (WHO) recommendation[18] and defined JE based on the following criteria: presence of clinical criteria of acute encephalitis syndrome[2] and (1) detectable JE specific IgM in CSF or serum, or (2) evidence of seroconversion or a 4-fold increase of IgM or IgG in the convalescence phase by the ELISA method, or (3) isolation of virus from blood, CSF fluid or tissue, or (4) detection of JE-virus genome in serum, plasma, blood, CSF or tissue.

Study population and data collection

This retrospective study included patients with confirmed diagnosis of JE between January 1, 2001 and June 30, 2018,

who were admitted in five medical centers in southern Taiwan, including Kaohsiung Medical University Chung-Ho Memorial Hospital (and its affiliated branch hospital of Kaohsiung Municipal Siaogang Hospital), Kaohsiung Veterans General Hospital, Kaohsiung Chang Gung Memorial Hospital, National Cheng Kung University Hospital, and Chi-Mei Medical Center (Yongkang and Liouying branch). We only included patients who were diagnosed with JE during hospitalization for acute stage of encephalitis. Patients aged <20 years, with incomplete medical data, and with evidence of central nervous system co-infections were excluded. A detailed review of medical charts during this period was performed, including initial clinical features, neurological examinations, laboratory data of blood tests and CSF analysis, imaging study, and treatments with antiviral agents or steroids. Steroid treatment was only included in the analysis when the dose administered was higher than dexamethasone 12 mg per day or its equivalence for >7 days.[4] We also classify the living areas into urban, suburban and rural area, for which might affect the access to medical source. The classification method was described in a previous study.[19]

In order to evaluate the influence of JE vaccination upon outcomes, we divided the patients into pre- and post-vaccination-era groups using the year 1965 as a cut-off point. Taiwan implemented national JE vaccination program since 1968 for children younger than 3 years.[17,20] According to one epidemiological study from Taiwan, the proportion of target population vaccinated with any dose of JE vaccine between 1970 and 2000 was above 85%.[21] Since we could not acquire the accurate vaccination records from the patients, we classify the patient groups into the pre- and post-vaccination eras. Patients born after 1965 were generally included in the mass JE vaccination program with high vaccination rates as mentioned above, therefore defined as the "post-vaccination-era" group.

The Glasgow Coma Scale (GCS) scores of the patients were recorded at initial encounter, nadir, and discharge, respectively. The GCS score change between initial encounter and nadir state were calculated. The neurologic manifestations at initial encounter were recorded: hyporeflexia (decrease or loss of deep tendon reflex), limb rigidity, and flaccidity. Patients were divided into different outcome groups for further analyses and comparisons. Patients with GCS scores >8 at the time of discharge were included in the good outcome (GO) group and those with GCS score ≤8 or with ventilator dependence were included in the poor outcome (PO) group.[2]

Statistical analysis

The data were expressed as median (range) or numbers (percentage) for different parameters. Statistical differences between the two outcome groups were analyzed by the Pearson chi-square test, Fisher's exact test or univariate logistic regression according to the nature of the variables. In multivariate analysis, binary logistic regression using backward strategies was used for independent factor analysis. All statistical analyses were performed using IBM SPSS Statistics for Windows version 22.0 (Armonk, NY); the significance level was set at a two-tailed p value < 0.05.

Results

Demographic characteristics and clinical features

A total of 111 patients were diagnosed of JE during the 18-year study period. Forty patients were excluded due to unavailability of medical records. Two patients were excluded due to early discharge against medical advice. One patient was excluded due to co-infection with herpes simplex virus. A total of 68 patients were included for further analysis (Fig. 1).

The demographics and clinical features of the 68 patients are shown in the first two columns of Table 1. Among the 68 enrolled participants, 42 (61.8%) were men, and the median age was 50 years. The majority of patients belonged to the 40-to-59-year age group (69.1%). Twenty-six (39.4%) patients belonged to the post-vaccination era. Most of the cases occurred in summer, especially in June (Fig. 2), which was consistent with the reports of previous studies in Taiwan.[8,10,22] The majority of patients dwelt in the suburban areas (54.5%).

The median hospitalization duration was 17 (range: 4–90) days. The most common symptoms at initial presentation were altered consciousness and fever (both 94.1%), followed by headache (51.4%). The median GCS score at initial encounter was 12 (range, 3–15). Moreover, seizure occurred in 22 patients (32.4%), of whom 17 had generalized seizures and five had focal seizures. Hyporeflexia was observed in 16 patients (25.4%). Twenty patients had abnormal muscle tone, 15 (23.8%) presented with flaccidity, and five (7.9%) presented with rigidity. The median Sequential Organ Failure Assessment (SOFA) score was 1.5 (range: 0–8) and the Charlson Comorbidity Index (CCI) was 1 (range: 0–7) (Table 1).

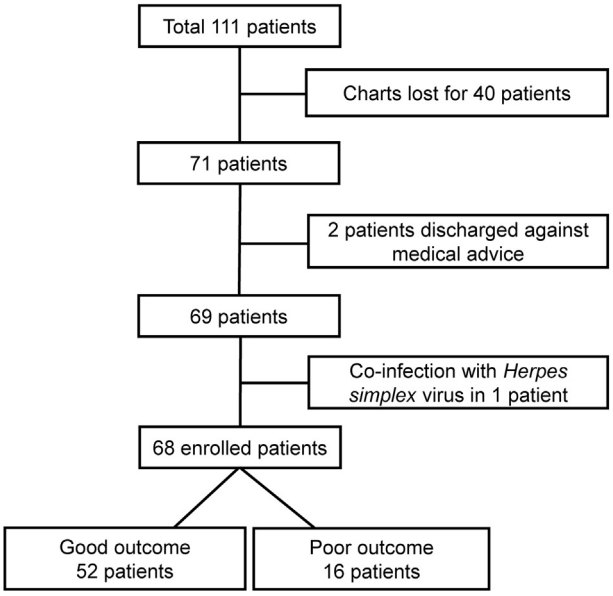

Figure 1. Flow chart of the process of patient enrollment.

Table 1 Demographic data and clinical features of the study population with a comparison between the good and poor outcome groups.

Characteristics (n = 68)	n (%) or median (range)	Good outcome (n = 52)	Poor outcome (n = 16)	p-value
Basic profile				
Male	42 (61.8)	32 (61.5)	10 (62.5)	0.945
Median age	50 (22–74)	50.5 (22–74)	45.5 (27–61)	0.173
Age group				0.760
>60	8 (11.8)	7 (13.4)	1 (6.3)	–
50-59	27 (39.7)	22 (42.3)	5 (31.3)	–
40-49	20 (29.4)	14 (26.9)	6 (37.5)	–
30-39	9 (13.2)	6 (11.5)	3 (18.8)	–
20-29	4 (5.9)	3 (5.8)	1 (6.3)	–
[a]Living area				0.380
Urban	21/66 (31.8)	17/50 (34)	4 (25)	–
Suburban	36/66 (54.5)	25/50 (50)	11 (68.8)	–
Rural area	9/66 (13.6)	8/50 (16)	1 (6.3)	–
[a]Post-vaccination era	26/66 (39.4)	20/51 (39.2)	6/15 (40.0)	0.956
Co-morbidity				
Hypertension	24 (35.3)	17 (32.7)	7 (43.8)	0.551
Diabetes mellitus	12 (17.6)	8 (15.4)	4 (25)	0.456
Stroke or other CNS disease	6 (8.8)	4 (7.7)	2 (12.5)	0.620
Malignancy	4 (5.9)	3 (5.8)	1 (6.3)	0.999
Renal disease	3 (4.4)	2 (3.8)	1 (6.3)	0.559
CCI	1 (0–7)	1 (0–7)	1 (0–5)	0.464
Clinical syndromes				
Fever	64 (94.1)	48 (92.3)	16 (100.0)	0.566
Headache	35 (51.4)	29 (55.8)	6 (37.5)	0.201
Myalgia	11 (16.2)	9 (17.3)	2 (12.5)	0.492
Dizziness	23 (33.8)	18 (34.6)	5 (31.3)	0.804
Nausea/vomiting	23 (33.8)	16 (30.8)	7 (43.8)	0.337
Weakness	26 (38.2)	21 (40.4)	5 (31.3)	0.511
[a]Neurological findings				
Seizure overall	22 (32.4)	14 (26.9)	8 (50)	0.084
Generalized seizure	17 (25.0)	10 (19.2)	7 (43.6)	0.049*
Focal seizure	5 (7.4)	4 (7.7)	1 (1.9)	0.999
Hyporeflexia	16/63 (25.4)	8/49 (16.3)	8/14 (57.1)	0.002*
Muscle tone				
Flaccidity	15/63 (23.8)	6/49 (12.2)	9/14 (64.3)	<0.001*
Rigidity	5/63 (7.9)	3/49 (6.1)	2/14 (14.3)	0.029*
Clinical course and severity				
Consciousness level				
Drowsy/confused	51 (75)	40 (76.9)	11 (68.8)	0.573
Stupor	9 (13.2)	7 (13.5)	2 (12.5)	0.999
Coma	4 (5.9)	1 (1.9)	3 (18.8)	0.143
GCS at admission	12 (3–15)	13 (5–15)	10 (3–15)	0.092
GCS at nadir	8 (3–15)	9 (3–15)	4.5 (3–7)	<0.001*
GCS change between admission and nadir	2 (0–12)	1 (0–11)	6 (0–12)	0.002*
Days from symptom onset to admission	3 (0–17)	3 (0–17)	3 (0–7)	0.641
Days from symptom onset to diagnosis	7 (0–34)	6.5 (0–34)	7.5 (0–18)	0.865
Days from symptom onset to GCS nadir	5 (1–20)	5 (1–19)	5.5 (2–20)	0.131
Hospital duration (days)	17 (4–90)	15 (4–90)	27.5 (12–59)	0.111
SOFA	1.5 (0–8)	1 (0–8)	3 (0–8)	0.009*
ICU admission	42 (61.8)	26 (50)	16 (100)	<0.001*
ICU duration (days)	13 (4–31)	9.5 (4–31)	20.5 (12–31)	<0.001*
[a,b]Inotropic agent use	5/67 (7.4)	2/51 (3.9)	3 (18.8)	0.084
Death	3 (4.4)	0 (0)	3 (18.8)	<0.011*

Table 1 (*continued*)

Characteristics (n = 68)	n (%) or median (range)	Good outcome (n = 52)	Poor outcome (n = 16)	p-value
[a]**Management**				
Steroid use	25/66 (37.9)	18/51 (35.3)	7/15 (46.7)	0.425
Acyclovir use	56/66 (84.8)	42/51 (82.4)	14/15 (93.3)	0.433
Acyclovir duration (days)	8 (0–15)	8 (0–15)	9 (0–15)	0.560

[a] Numbers were expressed by case number/available total cases under circumstances of data loss.
[b] Mainly norepinephrine or dopamine with short course less than 5 days.
SOFA = Sequential Organ Failure Assessment; CCI = Charlson Comorbidity Index; ICU = intensive care unit; GCS = Glasgow Coma Scale; CNS = central nervous system.
*$p < 0.05$.

Laboratory and image findings

The laboratory findings are shown in Table 2. Unlike the commonly expected hemogram in patients affected by vector-borne viral diseases with leukopenia and thrombocytopenia,[23,24] 40 patients (58.8%) were found to have leukocytosis and only two (2.9%) had leukopenia. Only 13 (19.1%) patients had thrombocytopenia, while 45 (75%) had elevated C-reactive protein (CRP).

Fifty-two (76.5%) patients were positive for anti-JEV IgM with the first collected serum or CSF samples, while the remaining 16 (23.5%) patients were diagnosed during seroconversion. CSF samples were obtained in 66 patients. Nearly all patients (64, 97%) had CSF pleocytosis with mostly lymphocyte predominance. Interestingly, 12 (18.2%) patients had predominant polymorphonuclear (PMN) pleocytosis. Sixty-five (98.5%) patients had elevated CSF protein. As anticipated, CSF to serum glucose ratios were normal in most patients except eight (12.1%) with CSF/serum glucose ratio <0.4. The CSF opening pressure was measured in 56 patients, and half of them were normal (<180 mmH_2O).

Among all patients, the brain computed tomography (CT) and magnetic resonance imaging (MRI) results of 61 patients were available. By CT, only 11 patients (18.0%) had thalamus, hippocampus, or basal ganglion involvement (data not shown). According to MRI results, 34 patients (55.7%) showed thalamic involvement, while 23 (37.7%) showed basal ganglion involvement. Other areas involved included leptomeningeal enhancement, hippocampus, and midbrain, which were found in <30% of all patients, respectively (Table 3).

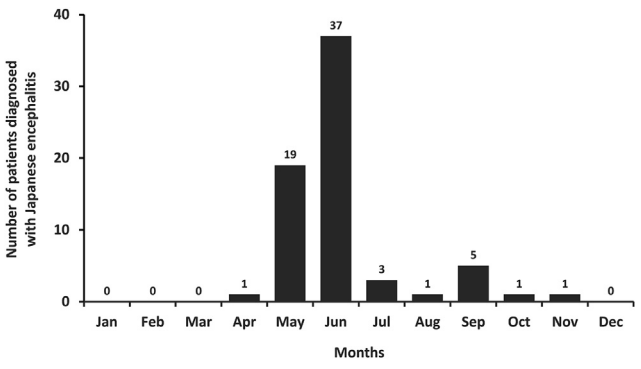

Figure 2. Number of Japanese encephalitis cases per month.

Antiviral agent and corticosteroid use

Among 66 patients with traceable medications records, antiviral therapy with acyclovir was administered in 58 (87.9%). Six patients (9.1%) were initially suspected of having tuberculosis meningitis and underwent anti-tuberculous treatment. Twenty-five patients (36.8%) were treated with steroids, mostly dexamethasone (Table 1).

Clinical course, outcomes, and prognostic factors

Forty-two (61.8%) patients were admitted to the intensive care unit (ICU), primarily due to rapid progression of the disease or subsequent respiratory failure, with a median ICU stay of 13 (range, 4–31) days. The median time from symptom onset to the nadir of GCS score was 5 (range, 1–20) days, and the median GCS score at the nadir was 8 (range, 3–15) (Table 1). Fifty-two patients (76.4%) were classified as the GO group, while 16 patients (23.5%) were of the PO group. About 35 (51.4%) patients were discharged with full GCS score, while 13 (19.1%) were ventilator-dependent.

All clinical features, laboratory data, and imaging findings were compared between the GO group and PO group (Tables 1 and 2). On univariate analysis, the significant factors associated with the PO group were higher SOFA score at presentation ($p = 0.009$), flaccidity ($p < 0.001$), rigidity ($p = 0.029$), hyporeflexia ($p = 0.002$), generalized seizure ($p = 0.049$), lower nadir GCS score ($p < 0.001$), greater GCS score change between admission and nadir ($p = 0.002$), ICU admission ($p < 0.001$), longer ICU stay ($p < 0.001$), higher CSF sugar level ($p = 0.031$); and higher CSF protein level ($p = 0.008$). Furthermore, we used CSF protein levels of 75, 100, 125 and 150 mg/dL for subgroup analysis and found that initial CSF protein level >125 mg/dL ($p = 0.021$) were significantly associated with PO (Table 2). Treatments with antiviral agents or steroids were not significantly associated with outcomes. With regard to the imaging findings, midbrain involvement was found in 28.6% and 8.5% of patients from the PO and GO groups, respectively; however, the difference was not statistically significant (Table 3). With multivariate analysis, the prognostic

Table 2 Laboratory data of the study population with a comparison between the good and poor outcome groups.

Blood exams (n = 68)	n (%) or median (range)	Good outcome (n = 52)	Poor outcome (n = 16)	p-value
WBC (*$10^3/\mu l$)	12.1 (2.6–65.3)	12.1 (2.6–23.4)	12 (4.6–65.3)	0.250
[a]Leukocytosis	40 (58.8)	30 (57.7)	10 (6.5)	0.859
[a]Leukopenia	2 (22.9)	2 (3.8)	0 (0)	0.999
Platelet (*$10^3/\mu l$)	185 (69–358)	186.5 (97–358)	175 (69–274)	0.113
[a]Thrombocytopenia	13 (19.1)	10 (19.2)	3 (18.8)	0.999
GFR (ml/min)	77.25 (5–167)	73.15 (0–167)	85 (0–164)	0.431
GPT (IU/ml)	22 (6–243)	20 (6–243)	26.5 (7–139)	0.560
Serum Sugar (mg/dL)	131.5 (64–297)	127 (89–241)	145 (64–297)	0.080
CRP mg/L	28.6 (0.2–297.8)	25.95 (0.2–298)	37.75 (0.2–210)	0.660
[a,b]CRP elevation	45/60 (75)	36/46 (78.3)	9/14 (64.3)	0.290
CSF analysis (n = 66)	**n (%) or median (range)**	**Good outcome (n = 51)**	**Poor outcome (n = 15)**	**p-value**
[c]Open pressure (mmH_2O)	182.5 (50–500)	185 (50–500)	170 (60–370)	0.640
CSF WBC (/μl)	176.5 (0–1105)	176 (0–663)	180 (30–1105)	0.250
[a]Pleocytosis	64 (97)	49 (96.1)	15 (9.8)	0.999
Lymphocyte (%)	65 (0–100)	64 (0–100)	64 (15–98)	0.687
PMN predominant (%)	12 (18.2)	10 (19.6)	2 (13.3)	0.999
CSF protein (mg/dL)	105.4 (54–299)	99 (0–198)	127 (78–299)	0.008*
CSF protein >75 (mg/dL)	54 (81.8)	39 (76.4)	15 (100.0)	0.054
CSF protein >100 (mg/dL)	36 (54.5)	25 (49.0)	11 (73.3)	0.141
CSF protein >125 (mg/dL)	16 (24.2)	9 (17.6)	7 (46.7)	0.021*
CSF protein >150 (mg/dL)	11 (16.7)	5 (9.8)	6 (40.0)	0.006*
CSF sugar (mg/dL)	66 (27–140)	62 (27–137)	83 (45–140)	0.031*
CSF/Serum glucose ratio	0.49 (0.18–1.80)	0.49 (0.18–1.17)	0.48 (0.40–1.80)	0.316
CSF/Serum glucose ratio <0.4	8 (12.1)	7 (13.7)	1 (6.7)	0.671

[a] Leukocytosis: WBC >11 × $10^3/\mu l$; leukopenia: WBC <4 × $10^3/\mu l$; thrombocytopenia: platelet <150 × $10^3/\mu l$; CRP elevation: >5 mg/L; CSF protein elevation: >45 mg/dL; pleocytosis: CSF WBC count >5 cells/mL.
[b] Data were expressed by case number/available total cases.
[c] n = 56, 43 in good outcome group and 13 in poor outcome group.
CSF = cerebrospinal fluid; WBC = white blood cell; CRP = C-reactive protein; GFR = glomerular filtration rate; GPT = glutamate pyruvate transaminase; PMN = polymorphonuclear cell.
*$p < 0.05$.

Table 3 Involved anatomical areas found on magnetic resonance imaging for the overall population with a comparison between good and poor outcome groups.

Area (n = 61)	n (%)	Good outcome (n = 47)	Poor outcome (n = 14)	OR (95% CI)	p-value
Thalamus	34 (55.7)	24 (51.0)	10 (71.4)	2.396 (0.658–8.229)	0.185
Hippocampus	11 (18.0)	7 (14.9)	4 (28.6)	2.286 (0.558–9.366)	0.251
Midbrain	8 (13.1)	4 (8.5)	4 (28.6)	4.300 (0.915–20.205)	0.065
Pons	4 (6.6)	3 (6.3)	1 (7.1)	1.128 (0.108–11.785)	0.920
Basal Ganglion	23 (37.7)	1 (38.3)	5 (35.7)	1.180 (0.382–3.42)	0.774
Meningeal enhance	14 (23.0)	10 (21.3)	4 (28.6)	1.480 (0.382–5.730)	0.570

OR = odds ratio; CI = confidence interval.

factors associated with PO were found to be flaccidity ($p = 0.001$), rigidity ($p = 0.043$), and elevated CSF protein ($p = 0.015$) (Table 4).

Discussion

JE is a vector-borne disease that causes severe neurological sequelae. Although several studies have been conducted to evaluate patients with JE, most of them mainly included children and only a few manuscripts reported the clinical manifestations and prognostic factors of JE in adults.[2,22,25] According to the notifiable infectious disease statistical system of Taiwan CDC, there were 481 confirmed JE patients during 2001–2018, and 92.5% of them were adults.[8,16] With regard to the association between outcomes and prognostic factors in adult JE, to our knowledge, this is the first study to report a large number of JE cases in

Table 4 Prognostic factors associated with poor outcome.

Variables	Univariate analysis		Multivariate analysis	
	OR (95% CI)	p-value	OR (95% CI)	P-value
SOFA	1.420 (1.093–1.844)	0.009*	1.509 (0.974–2.340)	0.066
Hyporeflexia	6.833 (1.859–25.115)	0.004*	–	–
Seizure				
Generalized seizure	3.325 (0.971–11.382)	0.056	–	–
Focal seizure	1.187 (0.117–12.085)	0.885	–	–
Muscle tone				
Flaccid	20.000 (4.189–95.481)	<0.001*	65.35 3 (5.022–850.540)	0.001*
Rigidity	8.889 (1.046–75.518)	0.043*	36.497 (1.128–1180.420)	0.043*
GCS nadir	0.496 (0.342–0.720)	<0.001*	–	–
GCS change between admission and nadir	1.329 (1.110–1.591)	0.002*	–	–
CSF protein	1.019 (1.005–1.034)	0.008*	1.031 (1.006–1.056)	0.015*
CSF sugar	1.028 (1.003–1.055)	0.031*	–	–

OR = odds ratio; CI = confidence interval; SOFA = Sequential Organ Failure Assessment, CSF = cerebrospinal fluid.
*$p < 0.05$.

adults. Our univariate analysis found that initial high SOFA score, high CSF protein level, and more severe neurological presentations including generalized seizure, greater change of GCS score between admission to nadir, flaccidity, rigidity, and hyporeflexia were associated with PO (Tables 1 and 2). In addition, when divided with a 25 mg/dL interval, we found CSF protein level higher than 125 mg/dL to be a cut-off value for PO prediction (Table 2). Further multivariate analysis determined that elevated CSF protein, flaccidity, and rigidity were three independent prognostic factors associated with PO.

The classic description of JE includes a parkinsonian syndrome, which is characterized by masklike facies, tremor, generalized hypertonia, cogwheel rigidity, and other movement abnormalities. Acute flaccid paralysis has been reported in 5–20% of JE patients and is attributed to anterior horn cell involvement.[23] Reflex changes are also common manifestations of JE, and hyporeflexia during acute stage may be caused by extensive anterior horn cell damage and/or cerebral shock.[25–27] In the present study, about one-fourth of the patients presented with hyporeflexia; 25.4% of the patients had flaccidity and 11.1% had rigidity. The neurologic manifestations of rigidity, flaccidity, hyporeflexia and generalized seizure were found to be associated with PO by univariate analysis, whereas only flaccidity and rigidity were found to be significant with multivariate analysis. Ooi et al. analyzed 118 children with JE in Malaysia to determine the long-term outcome of survivors and reported that poor perfusion, GCS score $\leq$8, and two witnessed seizure episodes were significant predictors for PO. In the study, abnormal muscle tone or reflexes were prominent factors for PO by univariate analysis but were not significant in multivariate analysis.[25] Another study conducted by Misra et al. prospectively evaluated 28 patients (17 adults and 11 children) to determine their prognosis and concluded that hyporeflexia predicted PO in bedridden patients.[27] Several neurologic abnormalities have been mentioned to predict PO in JE patients, however, the results of multivariate analysis varied.[25,27] Possible explanations include heterogeneous populations of adults and children and the various sample sizes. In addition, abnormal muscle tone may also correspond to reflex changes, for example, flaccidity with hyporeflexia and spasticity with hyperreflexia.[27] This particular relationship might contribute to the diverse results in multivariate analysis. Of note, seizure, an important indicator of central nervous system discharge abnormality and a significant prognostic factor for PO in children,[11,25,28] failed to show significance in adult population in the present study and previous reports.[2,4] Compared with patients without seizure, those presented with generalized seizure were observed more frequently in the PO group in our study (43.6% and 19.2%, respectively) (Table 1). The numerical predominance was found to have statistical significance by the chi-square test ($p = 0.049$), but not concurred by multivariate analysis. In our opinion, seizure could potentially be a predictor for poor outcome, under the circumstance of a larger case number. In summary, the study result suggests that muscle tone abnormalities at initial presentation predicts poor prognosis in adult JE patients, as seizures do in children.

CSF studies in JE patients often discloses elevated opening pressure, CSF pleocytosis, and elevated protein level with normal glucose levels.[29] Previous case series found that CSF protein levels correlated with clinical neurological symptoms.[22,30] This may imply that an abnormality in intrathecal protein synthesis or blood–brain barrier properties would lead to PO.[31] In this study, pleocytosis and elevated CSF protein levels were noted in almost all patients. More than half of the patients had high opening pressure, CSF lymphocyte predominance, and normal CSF/serum sugar ratio. However, some of the patients had PMN predominance (18.2%) in CSF and low CSF/serum sugar ratio <0.4 (12.1%), which could be confused with bacterial infection. For that reason, JE should still be considered as a differential diagnosis during peak seasons of JE in endemic regions, regardless of the CSF parameters. It is worth mentioning that six patients (9.1%) were initially suspected of having tuberculosis meningitis according to CSF data and anti-tuberculous drugs were administered. In regions with both tuberculosis and JE endemics like Taiwan, it is possible for these two disease entities to present similar clinical or laboratory features and cause difficulties in diagnosis.[32]

The classic imaging findings of JE are hypodense lesions in the thalamus and basal ganglia. MRI is more sensitive than CT scan and revealed prominent changes in the thalamus, basal ganglia, substantia nigra, cerebellum, pons, cerebral cortex, and spinal cord.[33] The thalamus and basal ganglion are critical functional regions for motor control, which explains the impaired motor functions observed in patients with more severe JE.[22] In addition, midbrain involvement was found to correlate with PO in a case series.[2] In our study, 68.9% of patients had notable abnormalities on MRI. The most frequently involved areas were the thalamus (55.7%) and basal ganglion (39.3%). However, no significant differences were observed between the PO and GO group for the various involved areas.

No significant differences were observed between the PO group and GO group in terms of steroid or antiviral treatments. The effect of steroid use on outcome improvement remained inconsistent among studies.[4,34–36] There is no available therapy that can effectively treat JE, and supportive care remains the mainstay of treatment. Hence, vaccination plays an important role in disease prevention and alleviation of disease burden in endemic areas. In Taiwan, routine JE vaccination with at least two inactivate vaccines derived from the Nakayama-NIH strain of JE virus-infected mouse brain or the inactivated freeze-dried Beijing strain has been implemented for all children under the age of three since 1968. The incidence of JE dramatically decreased from 2.0 per 100,000 people in the 1960s to 0.59–1.61 per 1,000,000 people after 2000.[8,17] We found no outcome differences between the pre- and post-vaccination-era groups, which may imply absence of long-term protective effect of the vaccine. According to previous studies, although the vaccine provided 96.8% protection after receiving at least two doses, the protective antibody could only be detected in 32% of the patients 3 years after the final vaccination.[37,38] Other studies showed that positive antibody rate was 63% after 16–20 years since the last booster dose for those who received four doses, and the positive antibody rates decreased as people aged.[17,39] The decline in antibody levels could partially explain the similar outcomes despite the reception of the JE vaccine. Therefore, adults at high risk of JE virus exposure such as those working in rural agricultural areas or pig rearing businesses are recommended to receive the JE vaccine booster dose according to the Taiwan CDC and Advisory Committee on Immunization Practices recommendations.[40]

This study has several limitations. First, owing to its retrospective nature, certain data loss was inevitable. Second, although we collected cases from five medical centers for 18 years in southern Taiwan, the region with the most JE cases in this country,[16] the case number from the post-vaccination era was relatively small (26 persons, 39.4%). Third, we did not have access to the accurate vaccination record of the study population. In this regard, our observations may not fully represent the current post-vaccinated generation in Taiwan.

In conclusion, CSF protein level and abnormal muscle tones are independent prognostic factors for PO. There are no outcome differences between the pre- and post-vaccination-era groups or in patients treated with steroid or antiviral agents. Our study results imply that a careful evaluation of the muscle tone and CSF protein level in adult JE patients are essential for outcome prediction.

Declaration of Competing Interest

None.

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