



Detection of Japanese Encephalitis Virus by Igm Elisa Among Acute Viral Encephalitis Suspected Patients at Jaipur

KEYWORDS

Japanese encephalitis virus, acute viral encephalitis, ELISA.

Mr. Jitendra Kumar Tiwari

Research Scientist-I Department of Microbiology & Immunology, Sawai Man Singh Medical College, Jaipur

Dr. Aradhana Chauhan

Research Scientist-II Department of Microbiology & Immunology, Sawai Man Singh Medical College, Jaipur

Ms. Reshmi T Nair

Junior Research Fellow Department of Microbiology & Immunology, Sawai Man Singh Medical College, Jaipur

Dr. Bharti Malhotra

Sr. Professor & Nodal Officer Department of Microbiology & Immunology, Sawai Man Singh Medical College, Jaipur

ABSTRACT

Background: Amongst the flaviviruses, Japanese encephalitis virus (JEV) is one of the major causes of encephalitis in India and Southeast Asia. The incidence of JE in recent times is showing an increasing trend. It appears that JE may become one of the major public health problems in India,

Aim: To detect JEV among acute viral encephalitis suspected patients by using IgM ELISA at Jaipur.

Materials and Methods: A total of 50 CSF and blood samples were tested for JEV IgM antibody using ELISA among acute viral encephalitis suspected patients.

Results: All samples were negative for JEV IgM antibody ELISA. Among the 50 suspected patients 28 (56%) were males and 22 (44%) females.

Conclusion: The all cases that were negative for JEV infection in the present study may have been due to other common etiological agents. Bacterial and tuberculous meningitis also fall into the differential diagnoses when biochemical findings and the cellular changes in the CSF are not conclusive.

INTRODUCTION:

Amongst the flaviviruses, Japanese encephalitis virus (JEV) is one of the major causes of encephalitis in India and South-east Asia. ⁽¹⁾ JEV is endemic in many parts of Asia, and a higher incidence of cases has been reported in children than in adults. Estimated 35,000– 50,000 clinical cases and 10,000 deaths caused by JE encephalitis are reported annually in Asia. ⁽²⁾ In India, flavi viruses including JEV are detected in many parts of the country. Thus far, JE activity has been reported from 24 states/Union Territories in India. ⁽³⁻⁵⁾

JE virus generates high mortality in pediatric group and today it is a major public health problem in South East Asia. Due to demographic, and environmental reasons vector transmission of JEV infection is very high and its outbreak commonly occurs every year among children in JE endemic and in JE-prone areas. ⁽⁶⁾

In India, the actual JE burden cannot be easily estimated because of scattered occurrence of JE in different states and regions. It could be only possible by strengthening diagnostic facilities for JE confirmation in hospitals situated in rural areas and by establishing national surveillance system for JE. ⁽⁷⁾

In India every year 30,000 to 50,000 cases of JE are reported out of which 10,000 patients died due to unavailability of treatment. Starting from April to November millions of JE cases are reported as vulnerable pediatric threat in many countries like China, India, Nepal, Bangladesh, Bhutan, Tibet, Korea, Japan, Thailand, Malaysia, Vietnam, Philippines. ⁽⁸⁾

Unfortunately, most of the Southeast Asian countries do not have appropriate immunodiagnostic tools for timely detec-

tion of JE. Besides this, due to economic reasons they cannot afford an efficient JE vaccine for immunization due to large number of JE patients. Due to lack of detection and extra delay in treatment very high mortality occurs almost every year in these countries. ⁽⁹⁾ Clinical presentation and patient history is suggestive of diagnosis but it still remains unreliable method of determining the specific etiology. A definite diagnosis improves the accuracy in treatment, as well as in better management of the patient. This prospective study was conducted to know the prevalence of Japanese encephalitis virus infection among hospitalized patients.

MATERIAL AND METHODS:

Study design: This was a prospective study.

Setting: This study was carried out on samples received at the Grade 1 DHR/ICMR Viral Research Diagnostic Laboratory, Advanced Basic Science and clinical research laboratory, Department of Microbiology and Immunology, Sawai Man Singh Medical College, Jaipur, Rajasthan.

Selection of patients:

Inclusion criteria:

The patients, presenting with progressive alteration of sensorium, behavioural abnormality, focal or generalized seizures with or without focal neurological deficit, preceded by a history of prodromal phase of headache, fever and other constitutional symptoms were included.

Exclusion criteria:

Samples from patients suffering from head injury, simple febrile seizures, seizure disorder, heat stroke, metabolic disorders, papilloedema were not included in the present study.

Sample collection and transportation:

A total of 50 CSF and blood samples were collected from patients with AES by a trained clinical staff, labeled and transported on ice at the earliest to the laboratory. Informed consent was obtained from the parents/guardians of the patients.

Sample Processing and Storage in Laboratory:

On receiving the CSF and blood samples in laboratory

they registered and given a unique identification number. The samples were separated and aliquoted in 2 vials. One vial stored at -80 °C for back up. Second vial was used for serology and stored at -20 °C till further use.

Serological Detection:

JEV IgM antibody estimation in CSF and blood was done using ELISA kits (InBios) as per manufacturer's instructions.

RESULTS:**Table-1: Distribution of JEV in acute viral encephalitis suspected patients:**

	Males			Females			Total Samples		
	Samples	Positive	%	Samples	Positive	%	Samples	Positive	%
Children	12	0	0	9	0	0	21	0	0
Adults	16	0	0	13	0	0	29	0	0
Total	28	0	0	22	0	0	50	0	0

In this prospective present study CSF and blood samples from viral encephalitis suspected patients from various departments are tested for detection of JEV IgM antibodies by ELISA. All samples were negative for JEV IgM antibody ELISA.

Among the 50 acute viral encephalitis suspected patients enrolled in the present study 28 (56%) were males and 22 (44%) were females.

In the present study among the 50 viral encephalitis suspected patients 29 (58%) were adults and 21 (42%) were children.

All patients presented with complaints of fever along with altered sensorium (100%). Other important commonly observed symptoms were seizures (80%), headache (25%) and vomiting (18%).

DISCUSSION:

Japanese Encephalitis is one of the leading causes of Acute Encephalopathy, affecting children and adolescents in Tropical and Sub tropical Asia. Epidemic outbreaks of Japanese Encephalitis continue to pose a significant public health problem in most parts of India.

The present study was carried out to diagnose Japanese Encephalitis cases among patients who were clinically suspected as viral Encephalitis.

In this prospective present study CSF and blood samples from viral encephalitis suspected patients from various departments are tested for detection of JEV IgM antibodies by ELISA. Among 50 samples tested all are negative for JEV IgM antibodies. Similarly all samples were negative for JEV IgM ELISA in study of Ramana et al from Andhra Pradesh.

Among the clinical manifestations, reported fever and altered sensorium was present in 100 % of the cases and seizures and headache accounted for 80% and 50% respectively of the cases, Male preponderance, which were noticed in our study is also well documented by chatterjee et al from West Bengal.⁽¹⁰⁾

The available information on the proportion of JEV infections among undifferentiated fever cases varies greatly in different populations. In a study in Thailand, 14% (22/156) of adult patients presenting with acute undifferentiated

fever were due to JEV. In a study carried out in Penang, Malaysia, among pediatric patients with non-specific febrile illness, 0.4% (2/482) were due to JEV infection during 1990 to 1992.⁽¹¹⁾ In another study conducted in 1967 in South Vietnam among American servicemen presenting with acute pyrexia of unknown origin, 6.8% (54/793) of cases were attributed to JEV.⁽¹²⁾

The viral encephalitis cases that were negative for JEV infection in the present study may have been due to other common etiological agents such as mumps, measles, enteroviruses such as coxsackie and echo, herpes simplex, and adenoviruses. Bacterial and tuberculous meningitis also fall into the differential diagnoses when biochemical findings and the cellular changes in the CSF are not conclusive. Cerebral malaria and Reye's syndrome also present with a similar clinical picture.

Japanese Encephalitis was sporadic in nature affecting all age groups, but predominantly, children. No specific antiviral therapy is available for Japanese encephalitis. The specific etiological diagnosis of Japanese Encephalitis cases helps the patient management protocols and avoids unnecessary use of antiviral therapy. Acyclovir therapy which is of no proven advantage in the cases of Encephalitis which were caused due to the Japanese Encephalitis Virus needs supportive and symptomatic treatment.

Thus, the management protocol was restricted to temperature control, seizure control, and the control of aggravating intracranial pressure and fluid and electrolyte management. Current JE vaccines are safe, effective and cost-effective. Effective JE vaccination programs provide the only solution to the ongoing threat of JE to endemic populations.

REFERENCE

1. Gajanan N. Sapkal, Nitin S. Wairagkar, Vijay M. Ayachit, Vijay P. Bondre, and Milind M. Gore. Detection and Isolation of Japanese Encephalitis Virus From Blood Clots Collected During the Acute Phase of Infection. *Am. J. Trop. Med. Hyg.*, 77(6), 2007, pp. 1139–1145
2. Tsai TF, 2000. New initiatives for the control of Japanese encephalitis by vaccination: minutes of a WHO/CVI meeting, Bangkok, Thailand, 13-15 October 1998. *Vaccine* 26 (Suppl 2): 1-3.
3. Jan LR, Yueh YY, Wu YC, Horng CB, Wang GR, 2000. Genetic variation of Japanese encephalitis virus in Taiwan. *Am J Trop Med Hyg* 62: 446–452.
4. Kumar R, Mathur A, Kumar A, Sethi G, Sharma S, Chaturvedi UC, 1990. Virological investigation of acute encephalopathy in India. *Arch Dis Child* 65: 1227–1230.
5. Suvarna Devi P, Behara PL, Swain A, 1996. Japanese encephalitis in Orissa. *Indian Pediatr* 33: 702–703.
6. Kabilan L, Rajendran R, Arunachalam N, Ramesh S, Srinivasan S, Samuel PP and Dash AP: Japanese encephalitis in India, an overview, *Indian J. Pediatric*. 2004; 171(7): 609-615.
7. Ramana R V, pavani P, Chaudhury A. Serological study for Japanese encephalitis virus among hospitalized patients. *International Journal of Pharmaceutical research and Bio Science*, 2012; volume 1 (15): 359-363.
8. Chew-Lim M and Ng CY: Recurrent viruses in a Singapore intensive pig farming estate. *Ann. Acad. Med. Singapore*, 1987; 16(4): 651-654.
9. Gunakasem P, Chantrasri C, Simasathien P, Chaiyanun S, Jatanasen S and Pariyanonth A. "Surveillance of Japanese encephalitis cases in Thailand", *Southeast Asian J. Trop. Med. Public Health*, 1981, 12(3), 333-337.
10. Chatterjee S, Chattopadhyay D, Bhattacharya MK and Mukherjee B: Serosurveillance for Japanese encephalitis in children in several districts of West Bengal, India. *Acta Pediatric*. 2004; 93(3): 390-3.
11. Cardosa MJ, Choo BH and Zuraini I: A serological study of Japanese encephalitis virus infections in northern Peninsular Malaysia. *Southeast Asian J. Trop. Med. Public Health*, 1991; 22(3): 341-346.