Trans. Natl. Acad. Sci. Tech. Philippines 21: 244-252 (1999).
ISSN 0115-8848

UPDATE: JAPANESE ENCEPHALITIS VIRUS ACTIVITY IN THE PHILIPPINES

VERONICA F. CHAN
Academician, National Academy of Science and Technology Philippines
Professor Emeritus, College of Public Health
University of the Philippines Manila

The virus, its transmission in nature, and the disease

Japanese encephalitis virus (JEV) causes acute encephalitis in humans and is principally rural in distribution. JEV is one of two members of the Flavivirus family that is active in the Philippines, second to the dengue viruses in prevalance. Recovery from JEV infection endows a lifelong immunity against JEV.

Transmission of JEV in nature is by means of a vector, the Culicine mosquitoes, that breed in the vast expanse of rice fields throughout the Philippines. This is a biologic transmission, meaning that the virus must first replicate in the gut of the mosquito before it can transmit the JEV while taking a blood meal. In nature, JEV is maintained in the amplifying host, hogs and domestic birds. Humans and vertebrates are incidental hosts. JEV is transovaially passed in the Culex mosquitoes.

The clinical features of encephilitis due to JEV are: 5 to 15 to 21 days of incubation period, 2 to 4 days of prodromal phase, 5 to 9 days of neurologic symptoms, that os prolonged seizures, respiratory dysfunction and finally death. The phatogenesis includes: transplacental infection leading to abortion, children, 3 to 15 years old, are primarily affected, and the ratio of apparent to inapparent infections ranges from 1:25 to 1000. In the brain, pathological features include neuronal degeneration, small hemorrhages with perivascular cuffing and monocytic infiltration. The disease is severe and life threathening with a case fatality rate of 25% and a rate of neurologic sequelae among surviving patients of 32 to 45%.

Japanese Encephalitis: a disease burden in Asia

Morbidity rates in China ranges from 5.5 to 24/10,000 population while in Taiwan and Thailand, it ranges from 1.8 to 2.5/10,000 population. In Indonesia,

Malaysia and the Philippines, 17 to 50% of hospitalized cases of viral encephilitis are due to JEV indicating high endemicity.

In Thailand, the case fatality rate among cases of Japanese encephalitis is 25%.

Japanese encephalitis among surviving patients suffer severe neurological sequelae. In Guam (1947) where the best documented follow-up of cases after ten years was reported, 40% continue to suffer disabilities, in Shanghai, China (1973-1997) 32% and in Thailand 45%.

Two-thirds of Asia's population live in rural areas, therefore 3 billion are at risk, chiefly children who are less than 15 years old. Based on the 1994 population estimates, 700 million are less than 15 years old and are at high risk. Thus, it is predicted that the annual incidence is 175,000 cases out of which 43,750 deaths and 78,750 disability cases occur as a result of Japanese encephalitis infections (assuming 2.5/25,000 children at risk, 25% of cases are fatal and 45% of surviving patients retain neurologic deficit).

Underreporting outweight overestimation of cases even if all cases of clinical encephilitis are reported without laboratory confirmation. Furthermore, atypical disease presentation contribute to underreporting (such as Guillain Barre syndrome, milder febrile illnesses without signs of enccphilitis, acute psychosis and deaths outside hospitals).

As early as 1958, Dr. William McD. Hammon, Professor of Epidemiology, Graduate School of Public Health, University of Pittsburgh, Pittsburgh, Pennyslvania, USA and Commisioner of the US Army Epidemiological Board, reported serological evidence of JEV presence in the Philippines (Table 1 from Hammon et al, 1980). Neutralizing antibodies against JEV were significantly higher among rural (Negritos around the former Clark AIrbase and Sapang Bato in Pampanga) than among urban children consulting at the Philippine General Hospital representing urban areas. Dr. Hammon argued that these JEV neutralizing antibodies can not be cross-reacting antibodies due to dengue virus because the percentages of seropositively between two viruses in the same area have values counter to each other be it in the rural or urban areas. Others confirmed these findings (Basaca-Sevilla and Halstead, 1966, Macasaet et al, 1970, and Venson et al, 1970).

In 1977, JEV was first isolated in the Philippines from *Culex tritaneurynchus* caught by CDC traps from four barrios in Tagudin, Ilocos Sur (Table 2 Trosper et al, 1980) and from San Jose, Nueva Ecija (Table 3 from Ksiazek et al, 1980). *Culex vishnui, Culex bitaneurynchus* and *Anopheles annularis* yielded also JEV isolates, but their role in the transmission of Jev in nature has to be established. Table 4 shows the identification of two isolates: Ph Ar 281 and Ph Ar 382 by micro-neutralization test. Only the JEV antiserum gave a clear and specific neutralization of the presence of JEV in the Philippines.

Table 1. Group B viruses. Positive results of neutralization test

		JBE		Dengue*		MVE		WN		Ntaya		Zika		SLE		Ilheus		Uganda S	
Locality	Age (yrs)	Ratio	%	Ratio	%	Ratio	%	Ratio	%	Ratio	%	Ratio	%	Ratio	%	Ratio	%	Ratio	%
Negritos	6/12-4	7/18	39	4/14	29	1/14	7	2/12	17	1/11	9	0/11	0	0/10	0	1/10	10	0/4	0
	5-9	18/23	78	7/19	37	2/19	10	2/15	13	4/20	20	0/19	0	0/10	0	0/20	0	0/17	0
	10-14	5/6	83	4/6	67	3/6	50	2/4	50	2/6	33	1/6	17	1/6	17	1/6	17	0/4	0
	15-19	15/15	100	6/12	50	7/15	47	6/15	40	4/11	36	3/11	27	3/15	20	2/11	18	0/3	0
	20+	61/62	98	38/54	70	34/62	55	36/62	58	28/53	53	8/51	16	21/61	34	11/53	19	0/13	0
Sapang	10-14	12/52	23	22/40	55	8/52	15	6/52	11	8/29	28	3/24	12	5/31	16	0/26	0	0/16	0
Bato Fields	15+†	25/51	49	–		–		–		6/12	50	3/12	25	–		1/12	8	0/12	0
Manila	6/12-4	1/32	3	12/33	36	1/26	4	3/27	11	5/22	23	1/13	8	1/23	4	0/18	0	0/2	0
Urban	5-9	4/23	17	13/21	62	4/21	19	6/12	29	7/13	54	0/6	0	5/20	25	0/7	0	–	0
Total		148/282	52	106/199	53	60/215	28	63/208	30	65/177	37	19/153	12	36/176	20	16/163	10	0/71	0

*Combined results of HAI and neutralization tests with viruses of both types.

†Food handlers principally form this general area.

(From: Hammon et al, 1980)

Table 2. Mosquitoes collected near Tagudin, Ilocos Sur Province, Luzon, Republic of Philippines, July 1977

Species	No. of mosquitoes (%)	No. of pools	No. of positive pools
*Culex tritaeniorhynchus**	19,677 (59.1)	210	1
*C. vishnui**	4,895 (14.7)	63	1
C. fuscocephala	981 (2.9)	19	0
C. bitaeniorhynchus	109 (0.3)	5	0
C. gelidus	180 (0.5)	8	0
C. whitmorei	15 (<0.1)	1	0
C. fuscanus	16 (<0.1)	3	0
C. fatigans	16 (<0.1)	2	0
C. pseudovishnui	14 (<0.1)	1	0
Anopheles anuularis	2,851 (8.6)	35	0
A. vagus	1,210 (3.6)	17	0
A. peditaeniatus	108 (0.3)	4	0
A. indefinitus	431 (1.2)	5	0
A. tessellatus	20 (0.1)	1	0
Mansonia uniformis	142 (0.4)	6	0
Aedeomyia catastica	1,244 (3.7)	21	0
Aedes vexans	1,134 (3.4)	24	0
A. lineatopennis	215 (0.6)	8	0
A. albopictus	9 (<0.1)	1	0
Mimomyia luzonensis	7(<0.1)	1	0
Total	33,274(100.0	435	2

*Isolation: Ph Ar 281 C. tritaeniorhynchus pool of 100.
Ph Ar 384 C. vishnui pool of 70.
(From: Trosper et al, 1980)

To completely show on firm grounds that JEV present and actively cause encephalitis among humans in the Philippines, studies by V.F. Chan et al at the College of Public Health, UP Manila Virus Laboratory showed that viral encephalitis cases were due to several viruses, one of them JEV (Table 5 ; note Case No. 14130, female, 2 years and 10 months old with a sustained highly significant antibodies titer of 1:1280 in both the first and second blood samples (Chan et al, 1983). Based on the initial findings, we went on to test all cases of viral encephalitis cases who were referred to the Virus Laboratory during the period of 1981-1982. Indeed, more viral encephalitis cases were found to be due to JEV infection (Table 6, Chan et al, 1983). It was nessecary, however, to demonstrate that the antibodies against JEV were monotypic and specific and cross-reacting antidengue

Table 3. Mosquitoes collected near San Jose, Nueva Ecija Province, Luzon, Republic of the Philippines during August 1977 from which virus isolations were attempted

Species	No. of mosquitoes (%)	No. of pools	No. of positive pools
Culex vishnui	50,652 (63.8)	525	1*
Cx. tritaeniorhynchus	11,396 (14.4)	139	0
Cx, fuscocephala	4,183 (5.3)	57	0
Cx, annulirostris	2,801 (3.5)	33	0
Cx, bitaeniorhynchus	1,744 (2.2	29	0
Cx, whitmorei	1,734 (2.2)	28	0
Cx, gelidus	820 (1.0)	14	0
Cx, fuscanus	117 (0.2)	4	0
Cx, fatigans	36 (0.0)	1	0
Anopheles annularis	2,491 (3.2)	32	0
An. peditaeniatus	1,936 (2.5)	27	0
An. indefinitis	300 (0.4)	3	0
An. tessellatus	200 (0.3)	4	0
Aedes vexans	247 (0.4)	7	0
Ae. lineatopennis	164 (0.2)	3	0
Mansonia uniformis	307 (0.4)	7	0
Total	79,157 (100.0)	913	1

**GET virus isolate, pool of 80 Cx. vishnui.*
(From: Ksiazek et al, 1980)

antibodies because the serologic test used was hemmaglutination-inhabitation (HI) test. To our delight, the HI antibodies were found only against JEV and not against the dengue viruses. (Table 7).

Does JEV feature in outbreaks in the Philippines? YES, and we recall the 1982 epidemic in Nueve Ecija that we documented by showing significant antibody rise in HI antibodies against JEV in paired blood samples of cases.

Prevention and control

Vaccination of the population at risk, children 15 years old and younger, is only the effective option to prevent occurence of Japanese encephalitis in the population. The successful reduction of morbidity due to anti-JEV vaccination is clearly shown by the experience in Japan where inactivated vaccine (mouse brain-derived) has been used since many years back, and also that of China where atttenuated vaccine is being used. The only commercially available vaccine, how-

Table 4. Identification of isolates Ph Ar 281 and Ph Ar 384 by micro-neutralization tests using 1.5-2.5 Log10 virus dose against varying serum dilutions

Sera	JEV (NAK)	281	384	Virus WN (Eg 101)	MVE (Orig.)	SLE (Parton)
JEV*	160	120	320	<10	<10	<10
281**	12	24	12	<4	ND	ND
384**	4	8	12	<4	ND	ND
WN*	<10	<10	<10	640	ND	ND
MVE*	<10	<10	<10	ND***	160	ND
SLE*	<10	<10	<10	ND	ND	160

* Hyperimmune mouse ascitic fluid (HMAF).

** 2 dose mouse antisera.
In addition, the following HMAF were tested against Ph Ar 281 and Ph Ar 384 viruses and found to react at <1:10; CHIK (S-27), GET (MM2021), BEB (MM2354), SIN (AR339, WHA (M78), SAG (Orig.) EEE (Ten Broeck), WEE (Fleming), DEN-1 (Hawaii), DEN-2 (New Guinea-C), DEN-3 (H-87), Den-4 (H-241), TMU (MM1775), LGT (TP21, ZIKA (MR8766), YF (17D), SEP (Aus MK7148), KUN (MRM16), ING (India 633970), BUN (Orig.), BAT (MM2222), BAK (MM2325), UMB Ig 1424) and Normal Mouse Ascitic Fluid.

*** ND = Not Done

ever, is inactivated vaccine which is given three doses on days 0, 7, and 30 by intramuscular or subcutaneous injection and boosted every three years. Travelers to hyperendemic areas are also devised to have their complete vaccination (3 doses) prior to departure.

Vaccination of the amplifying hosts, hogs and domestic birds, is not cost-effective and is not practical, and can not be enforced because when the hogs are JEV infected, they recover. It is only when the pregnant sows that get infected wahen the disease manifests as abortion. Consequently, in countries where central hog farming is practiced the economic undertone of JEV infection is tremendously devastating.

Vector control is not a practical either, because of the bionomics of the Culicines. It will very costly to reduce the source of the Culicines considering their distribution in nature particularly in rice fields. Around one's immediate environment though, eliminate all possible receptacles of water, drain canals of stagnant water, and empty the garbage cans regularly to prevent them from breeding which is the best option to be JEV infection-free. Remember, the Culicines are the mosquitoes that bite you night-time and you hear them buzzing around your ears. Kill the mosquitoes before they kill you.

Table 5. Cytomegalo, Herpes simplex, and Japanese encephalitis viruses as etiologies of viral encephalitis among Filipinos, 1982

Case no.		Reciprocal antibody titers against viruses				Viral etiology
		Cytome-galo v.	Herpes simplex	Dengue 2	Japanese Encephalitis	
14075 Female	First	16	8	<20	<20	Cytomegalovirus
18/12 yr.	Second	16	8	<20	<20	
14076 Male	First	<8	64	<20	<20	Herpes simplex virus
15 yr.	Second	<8	128	<20	<20	
14117 Female	First	64	16	80	40	Cytomegalovirus
7/12 months	Second	64	<8	160	80	
14130 Female	First	8	8	<20	<1280	Japanese encepha-litis virus
2 10/12 yr.	Second	8	8	<20	<1280	
14134 Male	First	<8	<8	<20	<20	Negative; un-confirmed
6 6/12 yr.	Second	<8	8	<20		
14158 male	First	8	16	20	40	Herpes simplex virus
Age?	Second	8	16	20	40	
14517 Sex & Age	First	<8	<8	20	20	negative; un-confirmed
unknown	Second	<8	<8	20	40	
14708 Male	First	<8	16	40	160	Herpes simplex virus
19 yr.	Second	<8	16	40	160	
14727 Male	First	<8	<8	20	20	negative; un-confirmed
10 yr.	Second	<8	M8	20	20	

From: Chan et al, 1983.

Table 6. Serological responses of human viral encephalitis cases against japanese encephalitis virus, Metro Manila, 1982

A. Cases with paired blood samples (6)

Case No.	Age years	Reciprocal of HI antibody titer versus Japanese encephalitis virus		Interpretation
		First Blood	Second Blood	
14045	7	20	80	Jap. enceph. infection
14068	30	40	40	Not Jap. enceph. inf.
14075	18/12	<20	<20	Not Jap. enceph. inf.
14130	2	>1280	>1280	Jap. enceph. infection
14134	6 1/12	<20	20	Not Jap. enceph. inf.
14158	4	<20	<20	Not Jap. enceph. inf.

B. Cases with single blood sample (22)

Reciprocal of serum dilution	Number of viral encephalitis cases with HI titers	Percentage
< 20	9	
20	4	
40	4	81.8
80	1	
160	1	
320	0	
640	1	
1280	2	18.2
2560		
Total	22	100

From: Chan et al, 1983.

Table 7. Monotypic HI Antibody Titers Against Japanese Encephalitis Virus in cases of Human Viral Encephalitis, Metro Manila, 1982

Case No.	Serum	Reciprocal of HI Antibody Titers Against Viruses	
		Dengue 2	Japanese encephalitis
14130	First*	< 20	≥ 1280
	Second**	< 20	≥ 1290
14094	First	< 20	≥ 1280
14096	First	< 20	≥ 1280
14316	First	ND	640
14554	First	ND	≥ 2560

*CFT titers versys cytomegalovirus and Herpes simplex virus were consistently. 1:8.
From: Chan et al, 1993.

REFERENCES

Chan et al. 1993. Phil. J. Microbiol. Inf. Dis. 12: 77-82.
Hammon et al. 1980. J. Trop. Med. Hyg. 7: 323-328.
Ksiazek et al. 1980. SEA J. Trop. Med. Pub. Health 11: 507-509.
Trosper et al. 1980. Trans Royal Soc. Trop. Med. Hyg. 74: 292-295.