Outbreak, Surveillance and Investigation Reports
Field Epidemiology Training Program, Bureau of Epidemiology
Department of Disease Control, Ministry of Public Health, Thailand
Tel: +6625901734-5, Fax: +6625918581, Email: osireditor@osirjournal.net, http://www.osirjournal.net

A Large Outbreak of Japanese Encephalitis in Rakhine State, Myanmar: Implication for Vaccine Policy

Pwint Mon Oo[1,*], Thaung Hlaing[1], Saw Lwin[2], Chakarat Pittyawonganon[3], Jeeraphat Sirichaisinthop[4], San Kyawt Khine[5]

1 National Malaria Control Program, Ministry of Health, Myanmar

2 Universal Research Co-operation, Yangon Region, Myanmar

3 Field Epidemiology Training Program, Bureau of Epidemiology, Department of Disease Control, Ministry of Public Health, Thailand

4 Department of Disease Control, Ministry of Public Health, Thailand

5 Vector Borne Disease Control Program, Rakhine State, Myanmar

*Corresponding author, email address: pwintmonoo@gmail.com

Abstract

Japanese Encephalitis is endemic in Rakhine State, western Myanmar. In July 2014, 27 patients were admitted to Sittwe General Hospital, Rakhine State. Few patients died and some patients were confirmed as having Japanese Encephalitis (JE) infection. An outbreak investigation was conducted in forty six villages of nine JE affected townships in Rakhine state. The objectives were to describe the outbreak and to identify potential risk factors, reservoir and vector of JE virus in Rakhine State. Active case findings, environmental and entomological study were performed. We found 49 JE suspected cases with 10 deaths. Among them, 21 cases were confirmed as Japanese encephalitis infection by positive immunoglobulin M (IgM) ELISA. *Culex tritaeniorhynchus* was found in most JE affected townships. Almost all villagers had no awareness about JE transmission and prevention. Our investigation underscored the importance of collaboration between human and animal doctors, the raising of knowledge and awareness about JE transmission and prevention before the rainy season in Rakhine State, and the need for more studies on usefulness of JE vaccination among high risk population.

Keywords: Japanese encephalitis, outbreak investigation, Rakhine State, Myanmar

Introduction

Japanese Encephalitis (JE) is endemic in Rakhine State of Myanmar due to virus circulation among domestic animals through *Culex* mosquito.[1] From 2009 to 2012, an average of 25 JE cases was reported every year in Myanmar. The highest reported areas were Rakhine State and Yangon Region.[2] However, in 2013, there was only one JE case reported in Rakhine State.[1]

Most of JE virus infection in human is asymptomatic as less than 1% of infected people develop symptoms. The incubation period is 5-15 days.[3] Illness usually begins with acute fever, followed by mental change, focal neurologic deficits and movement disorders. The case fatality rate is approximately 20-30%. Among survivors, 30-50% have serious neurologic or psychiatric sequelaes.[4] JE is one of the vaccine preventable diseases. Although one dose could be protective, vaccination with two doses is more likely to achieve 100% seroconversion.[5]

JE virus is transmitted by mosquitoes. In Myanmar, *Cx. tritaeniorhynchus* is the important vector in spreading this virus to humans and domesticated animals such as horses, donkeys, and pigs.[6] *Cx. tritaeniorhynchus* breeds in rice paddy field and is active at twilight. Sporadic human and animal JE cases are usually reported during the monsoon rains.[4]

In July 2014, 27 patients were admitted to Sittwe General Hospital in Rakhine State. Among them, eight died with neurological symptoms. Serum samples of all patients were sent to National Health Laboratory for enzyme-linked immunosorbent assay (ELISA) testing and seven were tested positive for JE. After receiving notification from the vector borne

disease control (VBDC) team in Rakhine State, a central VBDC team was sent to investigate the outbreak. The objectives of this study were to conduct an outbreak investigation and describe magnitude of the outbreak. Potential risk factors, reservoirs and vectors of JE virus were explored as well.

Methods

A descriptive study was conducted in 46 villages of nine JE affected townships in Rakhine State from 3 to 18 Aug 2014. To confirm the diagnosis, data on JE cases from the national VBDC program were reviewed. Information on demographic, clinical manifestations and laboratory results were reported to the national VBDC surveillance system by VBDC team leader. Medical records in Sittwe General Hospital (Reference hospital), including death certificates, were assessed as well (Figure 1).

A suspected JE case was a person who developed acute encephalitis syndrome with three or more of the following symptoms: acute fever, headache, convulsion, muscle spasticity, and alteration in mental and neurological signs[4] between 7 Jun and 30 Sep 2014. A confirmed JE case was a suspected patient with positive immunoglobulin M (IgM) ELISA for JE virus.

To confirm the existence of an outbreak, we compared JE report from VBDC program in the previous five years, 2009-2013. In addition, we also reviewed the clinical and demographic data from medical records in Sittwe General Hospital to describe magnitude of JE outbreak, and active case finding that was conducted in three villages with high morbidity rate from three JE affected townships (Sittwe, Minbya and Kyauktaw Townships) to find out new JE cases. Parents of children aged 5-15 years old who lived within one mile (1.6 km) around the patient house, either with or without fever, were approached. Moreover, we collected blood samples and sent to National Health Laboratory (NHL) in Yangon for laboratory confirmation.

An entomological survey was conducted in six villages with the highest attack rate of five JE affected townships to identify vector of JE virus. We used indoor collection in the morning, light trap, outdoor human landing catch or spray sheet collection[7]. This was conducted by the entomology team of central VBDC.

In order to search for high risk population, potential risk factors and reservoirs for JE, an environmental study was conducted by observing around 11 villages out of seven townships. In each village, one staff in the local authority, one basic health staff and one family member of JE patients were interviewed using unstructured questionnaires about knowledge and prevention behaviors.

Descriptive statistics were employed to display the distribution of suspected JE cases by time of onset, townships, and signs and symptoms. Attack rates and case fatality rates were explored by townships, including entomological and laboratory results. Association between potential risk factors and survival was examined by odds ratio and 95% confidence intervals. All analyses were performed using Epi Info 7[8].

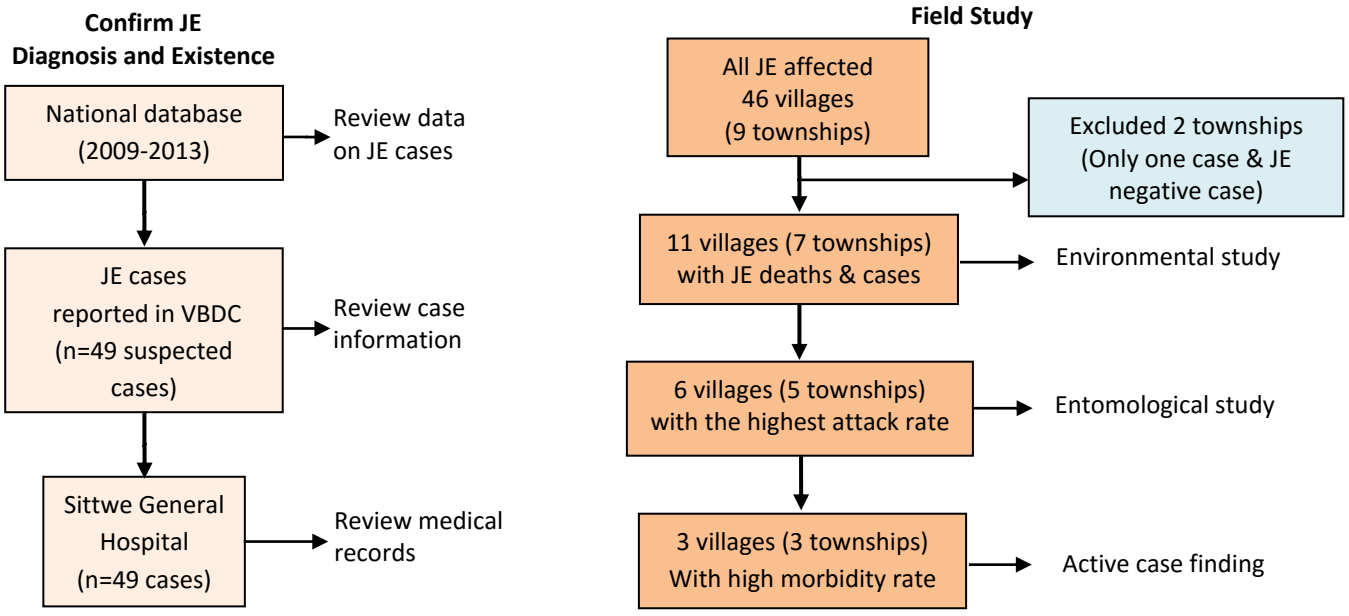


Figure 1. Method of data collection and field investigation in a Japanese encephalitis (JE) outbreak, Rakhine State, Myanmar, 2014

Laboratory Investigation

Blood specimens of suspected JE cases were confirmed for JE infection at NHL using JE IgM ELISA kit. The IgM ELISA consists of one enzymatically amplified 'two-step' sandwich-type immunoassay. In this assay, JE detect negative control (represents non-reactive serum), JE detect IgM positive control (represents reactive serum), and unknown serum samples were diluted with 1/100. The process was performed following the manufacturer's instruction. Sample dilution buffer for JE detect IgM was then incubated in microtitration wells coated with anti-human IgM antibodies. This was followed by incubating with both JE virus (JEV) derived recombinant antigen (JERA) and normal cell antigen (NCA) separately. After incubation and washing, the wells were treated with a JERA-specific antibody labeled with the enzyme horseradish peroxidase. After the third incubation and washing step, the wells were incubated with the tetramethylbenzidine substrate. An acidic stopping solution was added and the degree of enzymatic turnover of the substrate was determined by absorbance measurement at 450 nanometers. Above a certain threshold, the ratio of the absorbencies of the JERA and the control wells was accurately determined for presence of JEV antibodies.[9]

Results

Rakhine State is situated on the western coast of Myanmar. The main economy of the state is agriculture-based and others are forest-related works, animal husbandry, fishery, prawn breeding and some domestic small industries. Majority (75%) of the population is living in the rural area. There are total 17 townships, three sub-townships, 120 wards, 1,040 village tracts and 3,862 villages.[1]

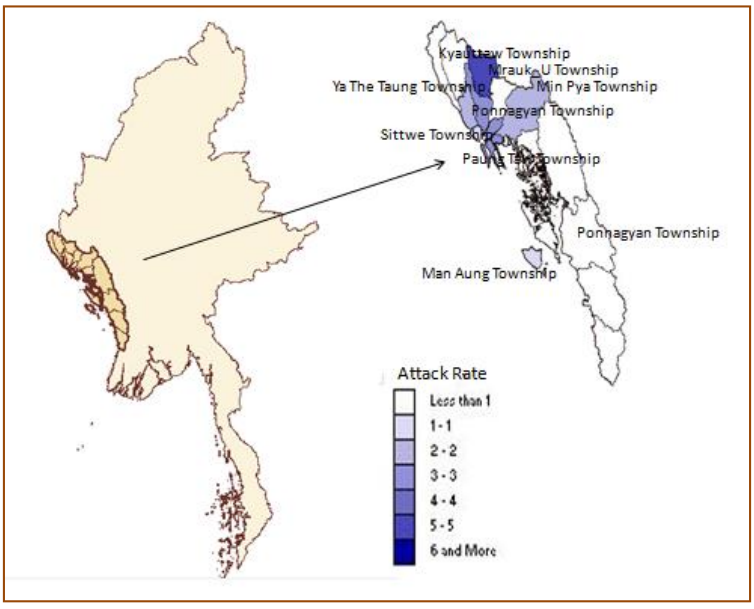
Diagnosis and Magnitude of the Outbreak

During the study period, active case finding and reports from VBDC identified a total of 49 suspected cases, with attack rate of 3.1 per 100,000 population, in 46 villages of nine townships in Rakhine State, including Kyauktaw, Minbya, Pauktaw, Ponnagyun, Sittwe, Rathedaung, Mrauk-U, Munaung and Toungup Townships (Figure 2).

Kyauktaw Township had the highest attack rate (5.4 per 100,000 population) among all the affected townships. The highest mortality rate (2.2 per 100,000 population) was recorded in Ponnagyun Township while the highest case fatality rate (50.0%) was in Sittwe Township (Table 1). From active case finding, 30 blood samples were collected. Nevertheless, all of them were tested negative for JE IgM by ELISA.

Among 49 suspected cases, there were 29 males (59%) and 20 females (41%). Most of cases were school-children and aged range from three months to 36 years, with median age as eight years. The highest attack rate, 5.4 per 100,000 population, was observed among 5-9 years old.

Samples of 35 JE suspected cases were sent to NHL and 21 cases (60%) were confirmed for JE by IgM ELISA. Two cases were with equivocal (uncertain) results. Among the 21 laboratory confirmed cases, four cases died and 10 survived with sequelae such as mental changes, focal neurological deficits while the others recovered completely.



Source: Data from Sittwe and Kyauktaw Hospitals in Rakhine State

Figure 2. Attack rate of suspected Japanese encephalitis cases by townships in Rakhine State, Myanmar, 3-18 Aug 2014 (n = 49)

Table 1. Attack rate and case fatality rate of suspected Japanese encephalitis cases by townships in Rakhine State, Myanmar, 3-18 Aug 2014

Township	Number of population[1]	Total number of cases*	Number of death*	Attack rate per 100,000	Mortality rate per 100,000	Case fatality rate (%)	Distance to	
							Rural health center (min)	Hospital (hour)
Sittwe§, ‡	269,205	8	4	3.0	1.5	50.0	-†	0.5 (car)
Kyauktaw§, ‡	221,644	12	1	5.4	0.5	8.3	20-25 (foot)	1 (car/boat)
Mrauk-U	214,307	2	0	0.9	0	0	20-25 (car/ boat)	1 (boat)
Minbya§, ‡	195,150	4	0	2.0	0	0	20-25 (boat)	1 (boat)
Pauktaw	191,426	8	1	4.2	0.5	12.5	30 (boat)	3 (boat)
Rathedaung‡	169,277	5	1	3.0	0.6	20.0	30 (boat)	2 (boat)
Toungup	148,417	1	0	0.7	0	0	45 (boat)	1 (boat)
Ponnagyun‡	135,942	8	3	5.9	2.2	37.5	30-35 (boat)	2 (boat)
Munaung	61,360	1	0	1.6	0	0	45 (boat)	1 (boat)

* Data sources: Sittwe and Kyauktaw Hospitals

‡ Entomological study was performed in this area.

§ Active case finding was performed in this area.

† As all 8 cases visited the hospital, no information was obtained from the interview.

All suspected JE patients first presented with fever, and followed by convulsion (63%), unconscious (53%), mental change (51%), nausea/vomiting (22%), headache (16%), or encephalitis (90%). Median duration from onset of illness to admission to hospital was four days, with inter-quartile range of 3-5 days. Ten patients died of acute encephalitis syndrome. No cases had history of JE vaccination.

All characteristics such as gender, age group, townships and symptoms of JE did not show any statistical difference when compared 10 deaths and 39 survivals of JE suspected cases (Table 2).

The JE outbreak started on 26 Jun 2014 and ended on 24 Sep 2014. The shape of the epidemic curve resulted gradual increase in nature (Figure 3). Compared with the previous five years data from VBDC program recorded in Rakhine State, less than five JE cases were recorded in each year during 2009-2013 and there was a clear increase in number of JE cases reported in 2014 (Figure 4).

Table 2. Analysis on characteristics and symptoms of suspected Japanese encephalitis cases in Rakhine State, Myanmar, 3-18 Aug 2014

Characteristic	Death (n=10)		Survival (n=39)	
	Number	Percent	Number	Percent
Gender				
Male	4	13.8	25	86.2
Female	6	30.0	14	70.0
Age group (year)				
0-10	8	22.2	28	77.8
>10	2	15.4	11	84.6
Townships				
Big*	6	27.3	16	72.7
Small**	4	14.8	23	85.2
Symptoms				
Fever	10	100.0	39	100.0
Headache	3	30.0	5	12.8
Convulsion	8	80.0	23	59.0
Mental change	6	60.0	19	48.7
Neck stiffness	0	0	8	20.5
Nausea/vomiting	1	10.0	9	23.1
Focal neurological sign	3	30.0	7	17.9

* Kyauktaw, Mrauk-U and Sittwe Townships

** Munaung, Minbya, Pauktaw, Ponnagyun, Toungup and Rathedaung Townships

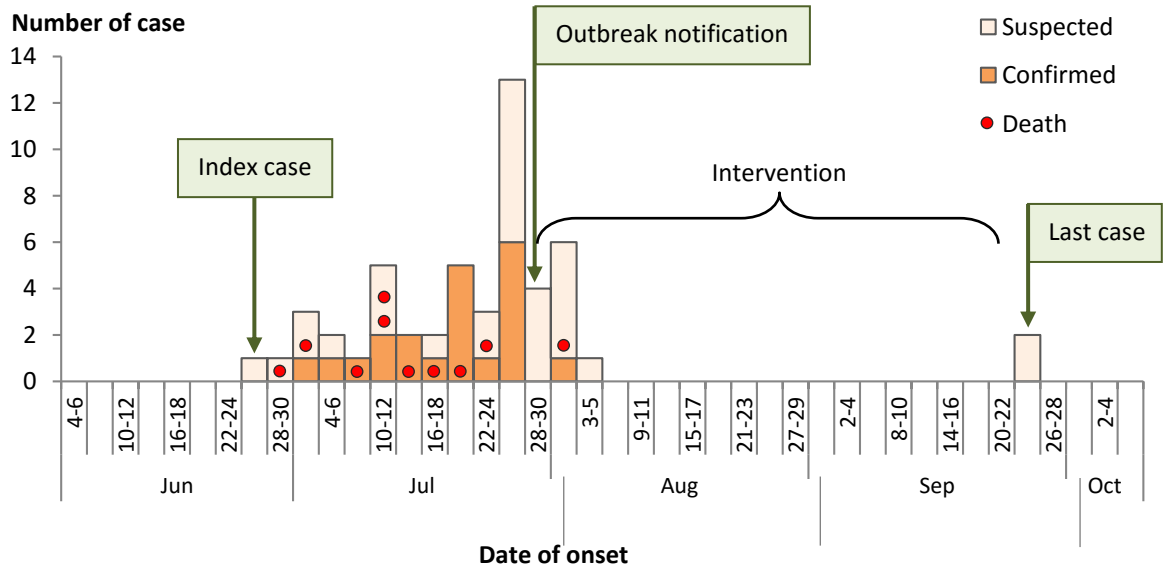


Figure 3. Distribution of Japanese encephalitis cases by date of onset in Rakhine State, Myanmar, 2014 (n=49)

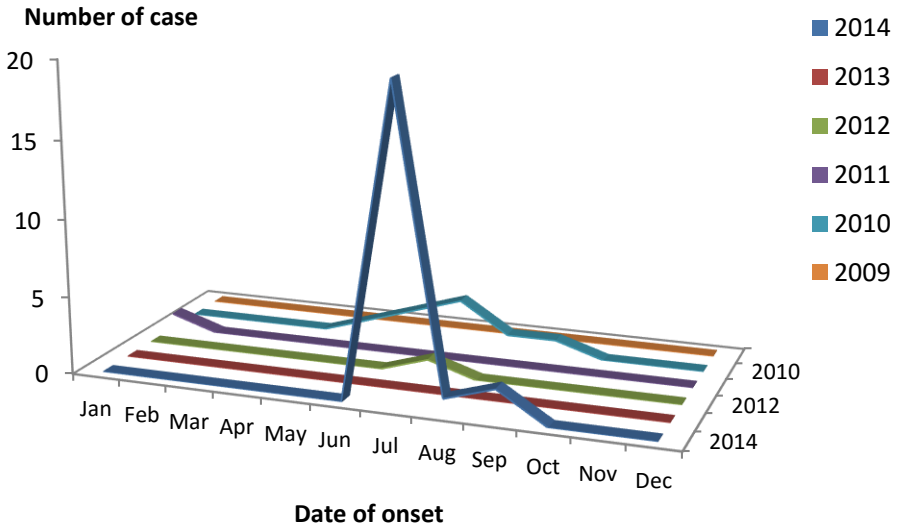


Figure 4. Number of Japanese encephalitis cases in Rakhine State reported to Vector Borne Disease Control Unit, Myanmar, 2009-2014

Potential Risk Factor, Reservoir and Vectors

According to the information from the local authority, basic health staff and families of JE patients, two months prior to this outbreak, there were mass causalities of pigs (more than 20-1000 pigs died), and still-born or mummified fetuses, abortion and infertility of pigs in some villages of Kyauktaw and Minbya Townships. Later, JE cases occurred sporadically in Pauktaw, Ponnagyun, Sittwe, Rathedaung, Mrauk-U, Munaung and Toungup Townships in Rakhine State. Although most villagers suspected that the cause of death in many pigs was due to swine fever, there was no definite evidence or laboratory confirmation.

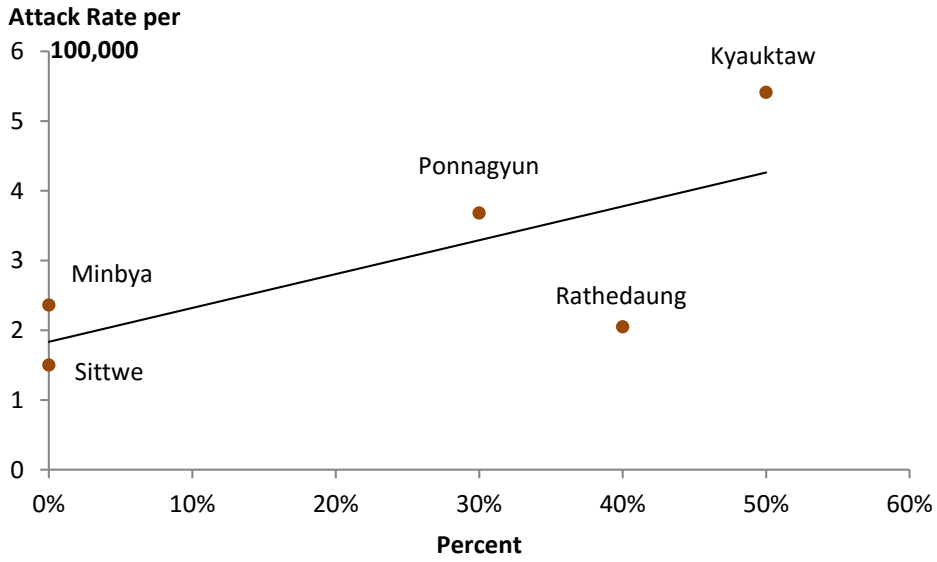
From the observation, there were also domestic or commercial poultry raised in the villages. The domestic pigs and cattle were kept under or in front of most of the houses in JE affected villages. Rice paddies were situated in most of the affected villages.

About 90% of people used ordinary bed net while only 10% of them applied the long-lasting insecticide-treated net. All people and pigs had no prior vaccination for JE. All the villagers did not have awareness about JE or knowledge related to transmission and prevention of the disease.

From the entomological survey, two major species of mosquitoes, *Cx. quinquefasciatus* and *Cx. tritaeniorhynchus*, were identified. The highest proportion of *Cx. tritaeniorhynchus*, the main vector of JE, was found to be 80% in Nagoomay Village, Kyauktaw Township (Table 3). A scatter plot between JE attack rate and proportion of *Cx. tritaeniorhynchus* mosquitoes surveyed from five townships suggested positive association. Despite

Table 3. Results of entomological survey among adult mosquitoes in Rakhine State, Myanmar, 3-18 Aug 2014

Name of village (Township)	Collection method	Number of mosquito	Type of adult *Culex* found	Percent of adult *Cx. tritaeniorhynchus*
Northern Sanpya (Sittwe)	- Indoor in the morning - Light trap - Outdoor human landing catch	48	*Cx. quinquefasciatus*	0
Balepound (Minbya)	Spray sheet	33	*Cx. quinquefasciatus, Cx. tritaeniorhynchus*	40
Athetmyintlet (Ponnagyun)	Spray sheet	28	*Cx. quinquefasciatus, Cx. tritaeniorhynchus*	30
Laydaungkan (Rathedaung)	Spray sheet	42	*Cx. quinquefasciatus*	0
Kathettaw (Kyauktaw)	Spray sheet	8	*Cx. quinquefasciatus, Cx. tritaeniorhynchus*	50
Nagoomay (Kyauktaw)	Light trap	4	*Cx. quinquefasciatus, Cx. tritaeniorhynchus*	80



Remark: Linear regression coefficient = 4.9, p-value = 0.18

Figure 5. Comparison between attack rate of Japanese encephalitis and proportion of adult *Cx. tritaeniorhynchus* in Rakhine State, Myanmar, 3-18 Aug 2014

that, the association was not statistically significant by linear regression (Figure 5).

Discussion

The investigation highlighed that an outbreak of JE occurred in Rakhine State during 2014 as 21 JE confirmed cases, which increased highly compared to those of the previous five years. JE outbreak was observed with case fatality rate 20.4% (10/49) as the JE patients died within a short period after appearance of the symptoms. Almost all JE cases were under 15 years old (90%).

Cx. tritaeniorhynchus which can spread JE was collected from the entomological study in three JE affected townships. Animal husbandry and paddy fields were found to be closely located near the residence of the cases. JE outbreak was most likely caused by mosquito bite, starting around early June 2014. According to interviews, many pigs died before JE outbreak in humans. JE virus can maintain in an enzootic cycle between mosquitoes and amplifying vertebrate hosts, primarily pigs.[4] The domesticated pigs under the patients' houses were most likely to be reservoirs of JE infection. Veterinarians could not find the cause of death of many pigs. Moreover, there was no established information sharing system for zoonotic diseases between veterinarians and public health officers in Rakhine State.

Many villagers in the study areas had no awareness about JE transmission and prevention. If early notification of JE cases could be carried out by the villagers, it would have been helpful for timely control

of the outbreak and saved the lives. Even though JE vaccination for pig is available, it is not the best solution since birds or other animals could also act as JE amplifying hosts. In endemic area, human vaccination could be more effective long-term control measure. On the other hand, this outbreak was difficult to control as most of the villagers could not afford for JE vaccine.

Limitations

Environmental and entomological studies were not be able to conduct in all JE affected villages because of unfavorable weather condition, poor transportation, insecurity and budget constraints. Despite outdoor human landing collection, using animal bait is the most suitable method for catching *Culex* mosquitoes[6]. However, this method could not be applied in all JE affected townships due to heavy rain. In addition, specimens from 14 suspected cases could not send for laboratory confirmation because of transportation barriers, inability to do blood test in the villages and higher expenses. Some patients had died before collection of blood samples.

Public Health Action and Recommendations

Despite the fact that JE vaccination have 90-97% efficacy,[9] it was costly and did not include in the expanded programme on immunization (EPI) in Myanmar. Hence, many people could not be vaccinated.

Advocacy meetings were conducted at the central level as well as in Rakhine State. Utilization of insecticide-impregnated bed nets, and long-sleeved shirts and pants were promoted to avoid mosquito bites[10]. Health education on disease transmission and prevention was provided to all affected villages as well. In addition, searching for more patients, opening of mobile clinic and vector control such as fogging were performed in the affected areas, in collaboration with the central VBDC team, VBDC team in Rakhine State and the local authorities.

We recommended that awareness about JE disease and personal protection should be raised in the communities before JE transmission season commences in the endemic areas. Local health authority should develop an early warning system of JE outbreak in case of abortion and fatalities in pigs through network among villagers, veterinarians and local health authorities.

The VBDC in Rakhine State should provide timely and immediate response such as health education on awareness and JE prevention and control. They should also encourage collaboration between physicians and veterinarians for information sharing as early as possible. Information obtained from this study was shared with the responsible persons and discussed for improvement of strategies and better support.

At the central level, introduction of JE vaccination to high risk group in collaboration with EPI program should be considered as an epidemiological study or a cost-benefit study. Thus, political commitment, financial resources and inter-sectoral collaboration between Ministry of Health and other stakeholders to set up JE vaccination programs for young children as well as rigorous monitoring and surveillance should be considered in prevention and control of JE.

Conclusion

An outbreak of JE was identified in Rakhine State during 2014. Total 21 confirmed cases out of 49 JE suspected cases were reported from 46 villages of nine townships, including 10 fatal cases. *Cx. tritaeniorhynchus* were found in most of the JE affected townships. Collaboration among physicians, veterinarians, local health providers, local health authority and community should be strengthened for better control measure in the future. JE vaccination should be considered for high risk population. Awareness raising about JE disease such as JE transmission, prevention and control should be started before the rainy season in Rakhine State to enhance knowledge and practice among the villagers.

After the study period which ended on 24 Sep 2014, two more JE cases were confirmed by IgM ELISA, with one each from Pauktaw Township and Myebon Township in Rakhine State. No more cases were reported since October 2014.

Acknowledgement

The authors wish to acknowledge the financial contribution from President's Malaria Initiatives through the United States Agency for International Development (USAID). We also would like to express our thanks to the staff from Central Vector Borne Disease Control Program and Vector Borne Disease Control Program of Rakhine State, Ministry of Health, Myanmar and International Field Epidemiology Training Program, Department of Disease Control, Ministry of Health, Thailand.

Specially, we are grateful to Dr. Chuleeporn Jiraphongsa and all advisors for providing guidance. We also appreciate our team members for their enthusiastic support, effort and active involvement in this study. Finally, we would like to express our sincere gratitude to local staff in Rakhine State for data retrieving and interviewing during the study.

Suggested Citation

Oo PM, Hlaing T, Lwin S, Pittyawonganon C, Sirichaisinthop J, Khine SK. A large outbreak of Japanese encephalitis in Rakhine State, Myanmar, 2014: implication for vaccine policy. OSIR. 2016 Jun;9(2):8-15.
<http://www.osirjournal.net/issue.php?id=97>.

References

1. Myanmar. Ministry of Health. Annual Report of Vector Borne Diseases Control Program, Rakhine State. Nay Pyi Taw: Ministry of Health, Myanmar; 2013.
2. Myanmar. Ministry of Health. Annual Report of Vector Borne Diseases Control Program. Nay Pyi Taw: Ministry of Health, Myanmar; 2013.
3. Mori MC, Siegel R. Live attenuated Japanese encephalitis (SA14-14-2) vaccine: The best alternative? The Vaccine Revolution. 2000 June 3 [cited 2014 Nov 10]. <https://web.stanford.edu/~siegelr/mori.html>.
4. Hills SL, Weber IB, Fischer M. Japanese encephalitis. In: Centers for Disease Control and Prevention. CDC Health Information for International Travel 2014. New York: Oxford University Press; 2014 [cited 2014 Nov 10]. <http://wwwnc.cdc.gov/travel/yellowbook/2016/infectious-diseases-related-to-travel/japanese-encephalitis>.
5. The Center for Food Security and Public Health. Japanese encephalitis. 2016 Jan [cited 2014 Nov 10]. <http://www.cfsph.iastate.edu/Factsheets/pdfs/japanese_encephalitis.pdf>.
6. Myanmar. Ministry of Health. Training modules on training course in entomology and vector control in Myanmar. Nay Pyi Taw: Ministry of Health, Myanmar; 2012.
7. Gordis L. Epidemiology. 5th ed. Philadelphia: Elsevier/Saunders; 2013.
8. Centers for Disease Control and Prevention. Epi Info [cited 2014 Nov 10]. <http://wwwn.cdc.gov/epiinfo/html/prevVersion.htm>.
9. InBios International, Inc. JE DetectTM IgM Antibody Capture ELISA (MAC-ELISA). 2011 Oct 28 [cited 2014 Nov 10]. <http://www.filgen.jp/Product/Bioscience4/InBios/JEMS-1%20JE%20Detect%20IgM%20Antibody%20Capture%20ELISA%20Insert.pdf>.
10. Daniel WW. Biostatistics: a foundation for analysis in the health sciences. 8th ed. Georgia: John Wiley & Sons, Inc.; 2005.