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PREPAREDNESS FOR NIPAH VIRUS OUTBREAKS

in At-Risk Countries

BACKGROUND

Nipah virus (NiV) is a lethal bat-borne paramyxovirus that first emerged in Malaysia in 1998-9, killing pigs and people. It is now known that Nipah virus is distributed widely across South and southeast Asia, with related henipaviruses found in Australia and Africa. In Malaysia NiV moved from bats into pig farms close to fruit bat habitat, then into people, suggesting that countries with large pig populations where this virus occurs are at-risk of future outbreaks. However, in Bangladesh, NiV has repeatedly been transmitted directly from bats to people as a food-borne infection. Furthermore, it has shown capacity to move person-to-person. The wide distribution of the virus and its reservoir hosts, the large human population in these countries and diverse farming and cultural practices suggest that the risk of future outbreaks is high.

MODERATOR

Jonathan EPSTEIN

Associate Vice President

EcoHealth Alliance
USA

OBJECTIVES

To demonstrate the value of a One Health approach to dealing with the emergence of henipaviruses in 3 different countries, with different impact and involving wildlife, livestock and humans. Each speaker with tell the 'story' of how NiV or HeV emerged in their country, how they mobilized resources to deal with it, and what this means for preventing the risk of emergence in the future. Each talk will touch on:

- how open communication and sharing of reagents and samples among countries helped rapid identification of cause and limited the size of outbreaks
- the public perception of these viruses in their countries

SPEAKERS

- **Emergence of Nipah Virus in Malaysia**
  Ramalan bin Mohamed, Director,
  Veterinary Research Institute, Malaysia
- **Assessing the risk of Nipah virus emergence in Thailand**
  Supaporn Wacharapluesadee, Laboratory Chief,
  Faculty of Medicine,
  Chulalongkorn University, Thailand
- **Repeated emergence of Nipah virus in Bangladesh via novel pathways**
  Stephen Luby, Professor, Stanford University, USA
- **The Application of One Health Approaches to Henipavirus Research**
  Hume Field, Principal Scientist,
  Queensland Centre for Emerging Infectious Diseases, Australia

JONATHAN EPSTEIN

Associate Vice President

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Dr. Jonathan Epstein is a veterinary epidemiologist and Associate Vice President of Conservation Medicine at EcoHealth Alliance (www.ecohealthalliance.org). He is the Asia Regional Coordinator under the USAID-funded Emerging Pandemic Threats: PREDICT program, a global effort to establish an early warning system for emerging viruses with pandemic potential through targeted wildlife disease surveillance. He also directs the One Health Alliance of South Asia (OHASA), a multi-lateral science and policy network with members from India, Pakistan, Bangladesh, Nepal, and Bhutan, focused on the control and prevention of trans-boundary zoonotic diseases. His current research activities are focused on understanding the ecology of emerging zoonotic viruses such as Nipah virus, Ebola and SARSCoV; which spill over through human-animal interfaces such as agriculture and trade. He is currently part of a large international collaboration that is investigating the ecology of Nipah virus in Bangladesh, where outbreaks occur in people almost every year with mortality rates reaching 90%.

Dr. Epstein is also the Executive Director of the Consortium for Conservation Medicine (CCM) based at EcoHealth Alliance. The CCM is a unique collaborative multi-institutional partnership including Johns Hopkins School of Public Health, Tufts School of Veterinary Medicine, University of Pittsburgh School of Public Health, University of Wisconsin-Madison, and the USGS National Wildlife Health Center. The CCM is the first formal inter-institutional partnership to applied conservation medicine - linking ecology, conservation, and health (both human and animal).

He holds adjunct faculty positions at Columbia University's Mailman School of Public Health; Tufts Cummings School of Veterinary Medicine and Tufts School of Medicine; and Mount Sinai School of Medicine. He holds advisory positions on two committees in the International Union for the Conservation of Nature (IUCN): the Wildlife Health Specialist Group and the Bat Species Specialist Group. His work has been published in several leading scientific journals including Science,PNAS, PLoS Pathogens,Emerging Infectious Diseases, The Annalsof the NY Academy of Science, and The Journal of Applied Ecology. Dr. Epstein has been an invited speaker at several US and international meetings including those held by the Institute of Medicine, The American Society of Microbiology, The CSIRO Australian Animal Health Laboratory, The Food and Agriculture Organization, and the World Health Organization.

Dr Hume Field is an internationally recognized authority on emerging diseases associated with bats. He is a veterinary epidemiologist with particular expertise in Hendra virus and Australian bat lyssavirus (the focus of his PhD research), Nipah virus (he worked with US Centres for Disease Control colleagues to control the 1999 Malaysian outbreak and identify the natural reservoir) and SARS coronaviruses (he worked with Australian, Chinese and US colleagues to identify the origins of SARS in 2003-4).

He has been a temporary advisor to the United Nations World Health Organisation (on henipaviruses and SARS), the United Nations Food and Agricultural Organisation (on SARS), and the World Organisation for Animal Health (on rabies and other lyssaviruses). He strongly believes that human, livestock, wildlife and environmental health are inextricably linked, and has long championed a 'One Health' approach to emerging infectious diseases associated with wildlife. He coordinated the Ecology of Emerging Infectious Diseases research program for the Australian Biosecurity Cooperative Research Centre from 2003-2010.

He developed the Wildlife Epidemiology component of the Master of Veterinary Public Health course for the University of Western Sydney in 2004-5. He was visiting Professor of Zoonoses at the University of Malaysia, Sarawak in 2009-10.

His current research is focused on identifying risk factors for Hendra virus spillover. He is Principal Scientist in the Queensland Centre for Emerging Infectious Diseases in Brisbane.

HUME FIELD

Principal Scientist

Queensland Centre
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STEPHEN LUBY

Professor

Stanford University
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Stephen Luby is Professor of Medicine with the Division of Infectious Diseases and Geographic Medicine; Deputy Director for Research at the Center for Global Health Innovation; Senior Fellow at the Woods Institute and Senior Fellow at the Freeman Spogli Institute for International Studies at Stanford University.

Prior to his current appointment, Dr. Luby served for eight years at the International Center for Diarrheal Diseases Research, Bangladesh (ICDDR,B), where he directed the Centre for Communicable Diseases. Dr. Luby was seconded from the US Centers for Disease Control and Prevention (CDC) and was the Country Director for CDC in Bangladesh.

Dr. Luby studied philosophy and earned a Bachelor of Arts summa cum laude from Creighton University in 1981. Dr. Luby earned his medical degree from the University of Texas Southwestern Medical School at Dallas in 1986 and completed his internship and residency in internal medicine at the University of Rochester-Strong Memorial Hospital. He studied epidemiology and public health in the Epidemic Intelligence Service (EIS) and the Preventive Medicine Residency of the Centers for Disease Control and Prevention.

Dr. Luby's career has included an EIS assignment to the South Carolina Department of Health and Environmental Control 1990-91; work with the CDC Malaria Branch in 1992; from 1993-98 Dr. Luby directed the Epidemiology Unit of the Community Health Sciences Department at the Aga Khan University in Karachi, Pakistan; and from 1998-2004 worked as a Medical Epidemiologist in the Foodborne and Diarrheal Diseases Branch of the CDC in Atlanta exploring causes and prevention of diarrheal disease in settings where diarrhea is a leading cause of childhood death.

Dr. Luby's research has addressed a number of public health issues. During his time in Bangladesh he lead a research group that explored the epidemiology of Nipah virus including detailed studies of villager's perspective on and response to the outbreaks and studies of virus circulation in its bat reservoir and spillover into domestic animals and humans. He has published over 200 scientific manuscripts.

Dr. Ramlan earned his Bachelor of Science degree in Biochemistry from University of Malaya, Kuala Lumpur in 1988. He received his Master of Science degree in Clinical Biochemistry in 2000 from the University Putra Malaysia. After 14 years of service in the government he was awarded post-graduate scholarship and obtained his PhD in 2006 from University of Sheffield, United Kingdom in Molecular Biology and Biotechnology.DrRamlan joined the Department of Veterinary Services in 1991 as Research Officer specializes in animal's vaccine development.

During Nipah outbreak in 1998/1999, he was involved directlyand responsible in laboratory diagnosis and testingNipah. He was appointed as the director of Veterinary Research Institute in 2007 and served as reference laboratory for all animal's diseases in the country. He has beenactive in scientific publication and currently serving as Chief Editor of the Malaysian Journal of Veterinary Research.

He also serves as penal reviewer for research proposal under Ministry of Science and Technology.

RAMALAN
BIN MOHAMED

Director

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SUPAPORN
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Laboratory Chief

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Dr. Supaporn Wacharapluesadee is the laboratory chief of Neuroscience Center for Research and Development and associate director of WHO Collaborating Centre for Research and Training on Viral Zoonoses, Faculty of Medicine, Chulalongkorn University, Bangkok Thailand. She received her undergraduate degree in Medical Technology at Chiang Mai University, earned her master's degree in Biochemistry at Mahidol University and her Ph.D. in Biomedical Science at Chulalongkorn University, Bangkok Thailand.

During 1994-1997, she worked in the malaria research laboratory at the Department of Entomology, Armed Forces Research Institute of Medical Sciences. In 1997, she worked at Chulabhorn Research Institute in cancer research areas. During 1998-2000, she worked in HIV research at The HIV/AIDS Collaboration Thai-USA. She started working at Chulalongkorn University in 2000, in the Neurology Division, Department of Medicine.

Dr. Wacharapluesadee's current research focus is in the area of viral encephalitis and zoonoses. She is a molecular biologist and field virologist. Her interests are on diagnostics, pathogenesis, and surveillance researches. She has developed several nucleic acid amplification methods for ante-mortem detection of rabies virus in humans and post-mortem diagnosis in animals. Doctor Wacharapluesadee and her colleagues are interested in the causes and mechanisms that differentiate rabies as furious or paralytic forms. She has developed the PCR protocols for Nipah virus detection from biological samples which is the most sensitive and reliable among others published protocol. Dr. Wacharapluesadee and her colleagues have engaged in wildlife research since 2002. Her group has found that Nipah virus infected bats in Thailand and demonstrated the seasonal prevalence of Nipah virus transmission in Pteropus lylei. This finding correlate with the seasonal outbreaks found in Bangladesh.

Dr. Wacharapluesadee's surveillance team included various scientific disciplines for example medical doctors, veterinarians, wildlife veterinarians, wildlife zoologists and scientists. The one health approach is being used in her current research. The collaboration involved the government sectors, academic sectors and the local community. She believed that the community or village-based initiation is the most powerful tool for the success of disease control and prevention, particularly with zoonoses.

THE APPLICATION OF ONE HEALTH APPROACHES
to Henipavirus Research.

HAYMAN DT, GURLEY ES, PULLIAM JR, FIELD HE.

Abstract

Henipaviruses cause fatal infection in humans and domestic animals. Transmission from fruit bats, the wildlife reservoirs of henipaviruses, is putatively driven (at least in part) by anthropogenic changes that alter host ecology. Human and domestic animal fatalities occur regularly in Asia and Australia, but recent findings suggest henipaviruses are present in bats across the Old World tropics. We review the application of the One Health approach to henipavirus research in three locations: Australia, Malaysia and Bangladesh. We propose that by recognising and addressing the complex interaction among human, domestic animal and wildlife systems, research within the One Health paradigm will be more successful in mitigating future human and domestic animal deaths from henipavirus infection than alternative single-discipline approaches.

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EMERGENCE OF NIPAH VIRUS

In Malaysia

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Abstract

The Nipah outbreak that rocked the swine industry in Malaysia in 1998/1999 has brought about positive changes to the industry. Around 1.1 million pigs were culled as a result of about 105 deaths in humans, mainly affecting swine farmers and their families. The disease was initially considered to be Japanese Encephalitis but subsequently announced to be a novel paramyxovirusnamed Nipah virus.In VRI, the reference laboratory for Nipah was set up to monitor the Nipah disease nationwide and carry out research to update and upgrade information pertaining to the disease in swine and other species. From 2001 to 2011,a total of 28,866 samples involving most animal species includingpigs are tested every year to ensure a national free status. The tests conducted to establish the free status are ELISA or PCR. In pigs, the tests are being done on commercial pig farms whereby 15 percent of the sow population is tested on a yearly basis. The results this far indicate Malaysia has not detected Nipah in swine from 2002 till now indicating the effectiveness of the control methods implemented.

Abstract

A pig-borne virus causing viral encephalitis amongst human beings in Malaysia was detected in 1997 by the Ministry of Health. Initially, the disease was considered to be Japanese encephalitis. Subsequently, it was thought to be a Hendra-like viral encephalitis, but on 10th April, 1999 the Minister of Health announced this mysterious and deadly virus to be a new virus named Nipah virus. The virus was characterized at CDC, Atlanta, Georgia. The gene sequencing of the enveloped virus revealed that one of the genes had 21% difference in the nucleotide sequence with about 8% difference in the amino acid sequence from Hendra virus isolated from horses in Australia in 1994. The virus was named after the village Nipah. In all, the Ministry of Health declared 101 human casualties, and 900,000 pigs were culled by April, 1999. The worst affected area in Malaysia was Negri Sembilan. The symptoms, incubation period in human being and pigs, animal to human transmission, threat of disease to other livestock, and control program adopted in Malaysia is described.

Abstract

Nipah virus, a novel paramyxovirus, closely related to Hendra virus emerged in northern part of Peninsular Malaysia in 1998. The virus caused an outbreak of severe febrile encephalitis in humans with a high mortality rate, whereas, in pigs, encephalitis and respiratory diseases but with a relatively low mortality rate. The outbreak subsequently spread to various regions of the country and Singapore in the south due to the movement of infected pigs. Nipah virus caused systemic infections in humans, pigs and other mammals. Histopathological and radiological findings were characteristic of the disease. Fruitbats of Pteropid species were identified as the natural reservoir hosts. Evidence suggested that climatic and anthropogenic driven ecological changes coupled with the location of piggeries in orchard and the design of pigsties allowed the spill-over of this novel paramyxovirus from its reservoir host into the domestic pigs and ultimately to humans and other animals.

ASSESSING THE RISK OF NIPAH VIRUS EMERGENCE
in Thailand

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Since the initial outbreak of Nipah virus pneumonia/encephalitis in Malaysian abattoir pig workers in 1998 (Chua et al., 2000), there have been more than 25 human outbreaks in India and Bangladesh. Outbreaks in Bangladesh have occurred almost every year since 2001 and tended to be seasonal (Luby et al., 2009; Lo et al., 2012). Pteropid bats are the primary natural reservoirs (Yob et al., 2001; Halpin et al. 2011; Yadav et al. 2012). Their distribution ranges across the tropical regions of Asia and Africa. Viral transmission occurs among bats and spills over to other dead end host species including humans. Humans develop disease from having contact with virus contaminated tissues from infected pigs (Malaysia) (Chua, 2003) or otherwise by drinking bat-contaminated fresh date palm sap (Bangladesh) (Luby et al., 2006). Human-to-human transmission poses a serious threat and has been only documented in Bangladesh (Gurley et al., 2007).

To date there has been no report of pig or human cases of Nipah virus (NiV) infection in Thailand. However, there are several factors which could allow NiV emergence in Thailand. Thailand borders Malaysia where NiV outbreaks have been documented in 1998. Thailand harbors at least 20 million bats with 138 species among which several have been reported as NiV reservoirs. There are approximate 9.7 million pigs in this country and this has led to a preparedness initiative program. It consists of cooperation and joint activities among inter-sectoral organizations concerned with surveillance of wildlife, pigs and active testing for NiV infection in patients with encephalitis. Evaluation of health status and determination of risk behaviors as well as serology screening of residents in NiV prevalent zones are being conducted. This has been done in parallel with studies of bat ecology, behavior, population size and relation to seasons.

The Department of Livestock Development (DLD) under the Ministry of Agriculture and Cooperatives of Thailand has conducted sero-surveillance for Nipah infection in pigs since 1998. Regions 8th and 9th adjacent to Malaysia and regions 2nd and 7th

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with high density of pigs were chosen from a total of 9 regions. Serum samples were collected from pigs at both farms and backyards for seroprevalence study. During 1998-2010, 44,658 pig sera were all negative by ELISA testing. In 2011, 3,352 tested pig sera from twenty (of 77) provinces were also negative. Passive surveillance was performed by postmortem examination of pig lung tissues with interstitial pneumonia. A total of 1,996 lung tissue specimens during 1998-2011 were all negative. This surveillance plan has been continually updated and adjusted, based on the previously obtained information. This included risk-based surveillance, according to the geographical location and seasonal preferences. For effective incident management and coordination of emergency responses, DLD has developed a Manual on Emergency Preparedness and Contingency Plan for NiV outbreaks. This manual has been distributed to the relevant DLD officers.

Further surveys of NiV infection in Thai bats is being conducted since 2002. Three species of flying foxes (genus Pteropus); P. lylei, P.hypomelanus, and P.vampyrus, have now been shown to be major reservoirs of NiV in Thailand (Wacharapluesadee et al., 2005). Although NiV infection has been found mainly in 3 species, P. lylei is dominant reservoir due to its population size and distribution. Furthermore, its habitat is closer to humans compared to the other two Pteropus species. Genetic characterization of NiV found in P. lylei in Thailand, based on 357 base pairs of nucleoprotein gene, indicated 2 existing genotypes: Bangladesh- and Malaysian- like (Wacharapluesadee and Hemachudha, 2007). Seasonal prevalence of NiV transmission in P. lylei has been shown during April-May (Wacharapluesadee et al., 2010). Although both strains are co-circulating, the Bangladesh-like genotype dominates in P.lylei bat populations.

Assessing the risk of Nipah virus emergence at the high risk areas, has been conducted in villagers, pigs, and bats at the areas where NiV infection has been demonstrated in bats. Bats were trapped monthly at two pig farms, using mist nets and harp traps. Blood, saliva and urine specimens were collected from each bat for further identification. Numbers of bats that were captured at the pig farms were highest in May, which is the weaning period that juvenile started to fly. Bats that were captured at pig farms were NiV positive only in May (by serology and/or viral RNA). The findings of seasonal preference in May were similar to our previous findings from pooled urine samples at bat roosts (Wacharapluesadee et al., 2010). The finding of NiV infected bats at the pig farms raises concern of possible NiV transmission to pigs. Although there were no evidence of NiV infection in pigs in the same study, measures to prevent bats from coming into contact with pigs and avoidance of feeding pigs with partially eaten fruits must be actively promoted.

To ensure that there is no human transmission of NiV, a community in Chonburi province located near a temple inhabited by NiV infected bats (P.lylei), was chosen for community-level study. After obtaining written consent, a standard questionnaire which included questions related to potential risk behaviors was administered to 418 local residents. Blood samples were assayed for NiV antibody using an indirect ELISA for IgG antibodies against NiV-infected cell lysate. Positive criteria for NiV infection was a dilution ≥1:400. No NiV-specific IgG antibodies were found. The low NiV prevalence in P.lylei (9.3%) and different culture of not drinking fresh date palm sap may have negative impact upon emergence of infection in Thailand. This is in

contrast with the finding in Bangladesh. People living in risk regions are being educated to avoid risk behavior and monitored continuously concerning symptoms possibly related to NiV infection. Febrile patients and those with respiratory or encephalitis symptoms living in or near NiV zones require particular attention. To date, transmission to human has not been identified in Thailand. Cerebrospinal fluid of 232 patients with encephalitis admitted to King Chulalongkorn Memorial Hospital during 2001-2012, were tested negative for NiV RNA.

Inter-sectoral cooperation among wildlife-animal-human departments in preparedness response to emergence of NiV outbreak in Thailand is a successful example of a transboundary "one health" approach. Establishment of alert system and promotion of awareness from the bottom level (village-based) is also the key of the success for this one health initiative. The ultimate goal of the whole process cannot be achieved without public recognition and willingness, otherwise this cannot be sustainable. Policy makers must value this as an opportunity in strengthening public health infrastructure.

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