

Nipah Virus Infection- Malaysia Experience

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The emergence of Nipah virus in Malaysia

- Approximately 1.1 million pigs were culled in Peninsular Malaysia between September 1998 and May 1999.
- Of 257 reported and attributed human cases in Malaysia, 105 were fatal.



- The disease in pigs was highly contagious, and characterized by acute fever with respiratory involvement and sometimes nervous signs in all age classes.
- Sows and boars sometimes died peracutely (Nor *et al.* 2000).

The predominant clinical syndrome in humans

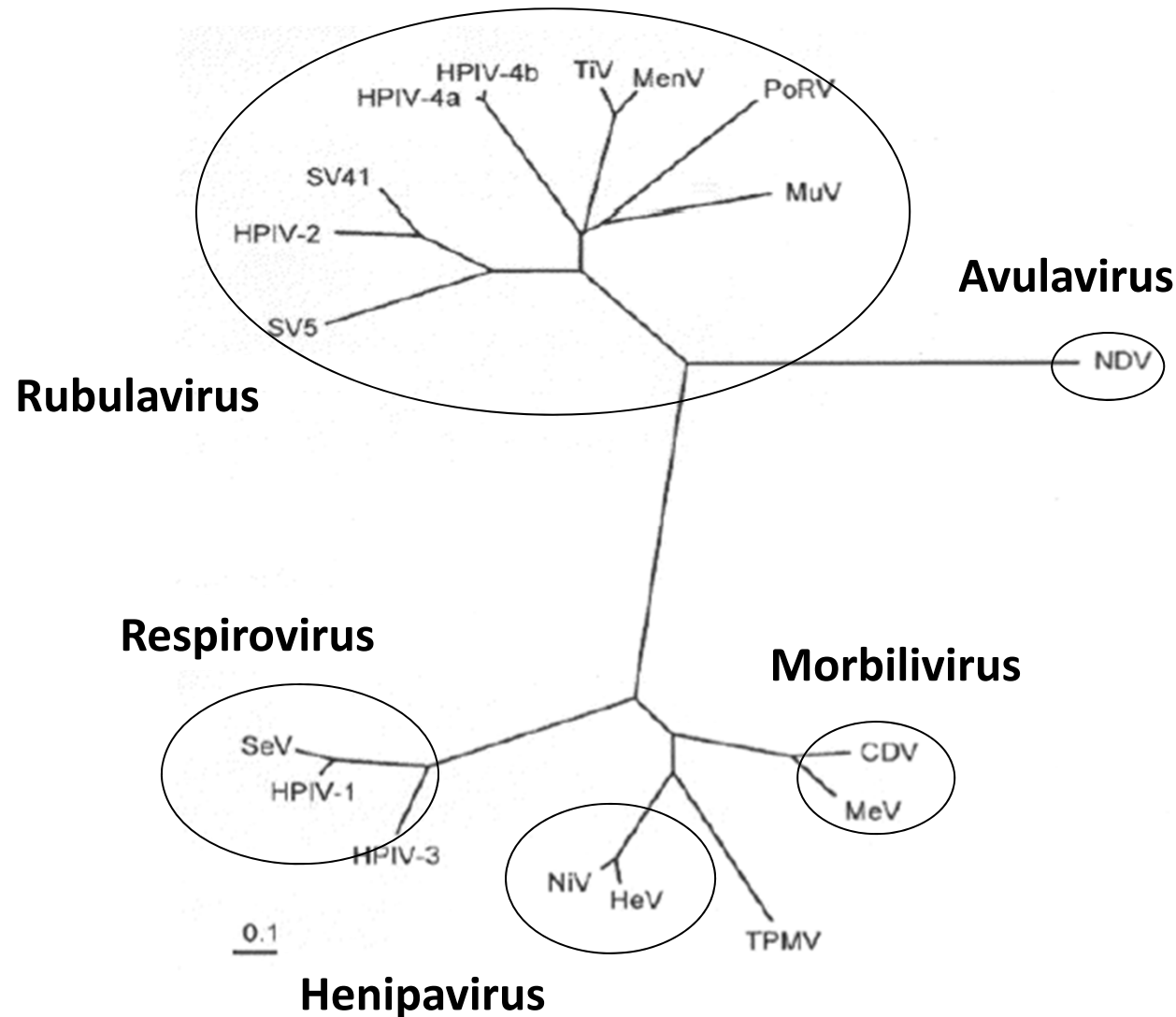
- Encephalitic rather than respiratory, with clinical signs including fever, headache, myalgia, drowsiness, and disorientation sometimes proceeding to coma within 48 hours.
- many of the surviving encephalitis cases suffer nervous sequel.
- The majority of human cases were employed in the pig industry and had a history of direct contact with live pigs.

The causative agent

- Nipah virus of the family *Paramyxoviridae*.
- Closely related to Hendra virus than to other paramyxoviruses. Genus Henipahvirus.
- Hendra virus is a recently emerged and zoonotic virus first described in horses, humans and bats in Australia in 1994 .
- Nipah virus has been shown through serology to infect people, pigs, horses, dogs, cats, chickens, and bats.
- Clinical disease in humans, pigs, dogs, and cats.
- Nipah virus is classified as a biosecurity level 4 (BSL-4); poses a risk to people during examination or treatment of infected animals.



A Phylogenetic Tree Based on the Deduced Amino Acid Sequences of the Matrix Protein of Member of the Family *Paramyxoviridae*. Showing the Close Relationship between HeV and HeV (Source: Chua et al., 2002a)



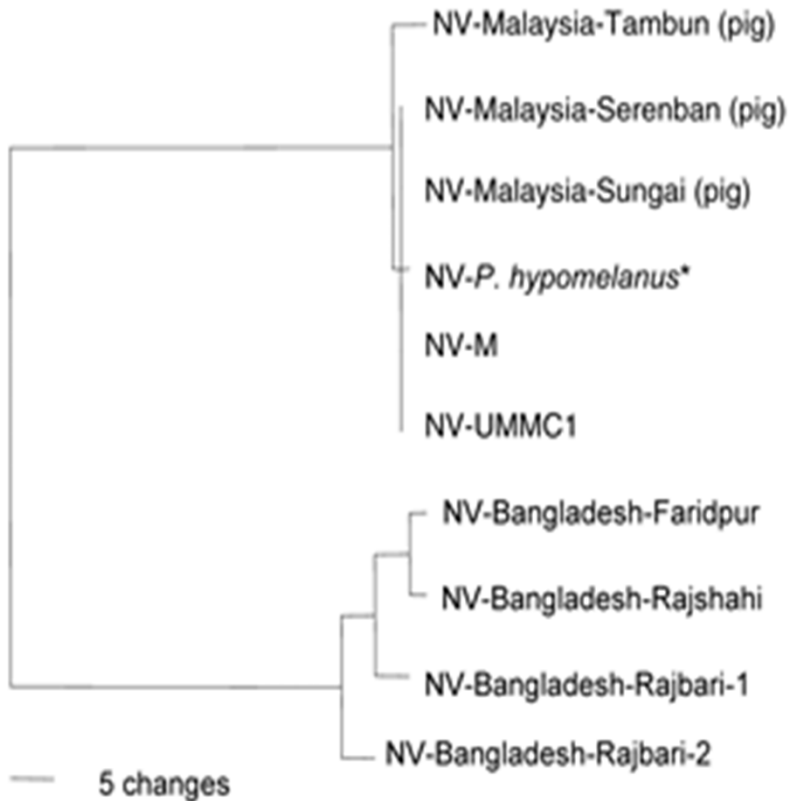


Figure 1. The Phylogenetic Relationship between the N Gene Sequences of the 4 Human NiV Isolates from the Bangladesh Outbreak in 2004 and the N Gene Sequences from Pig and Human NiV Isolates from Malaysia (Source: Harcourt et al., 2005)

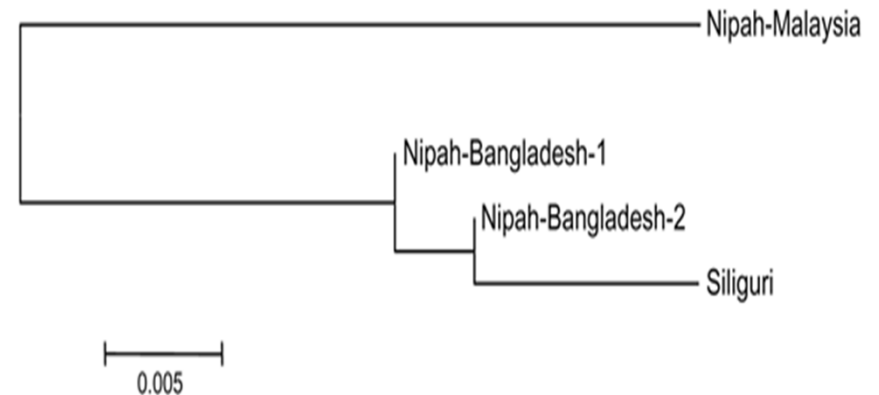
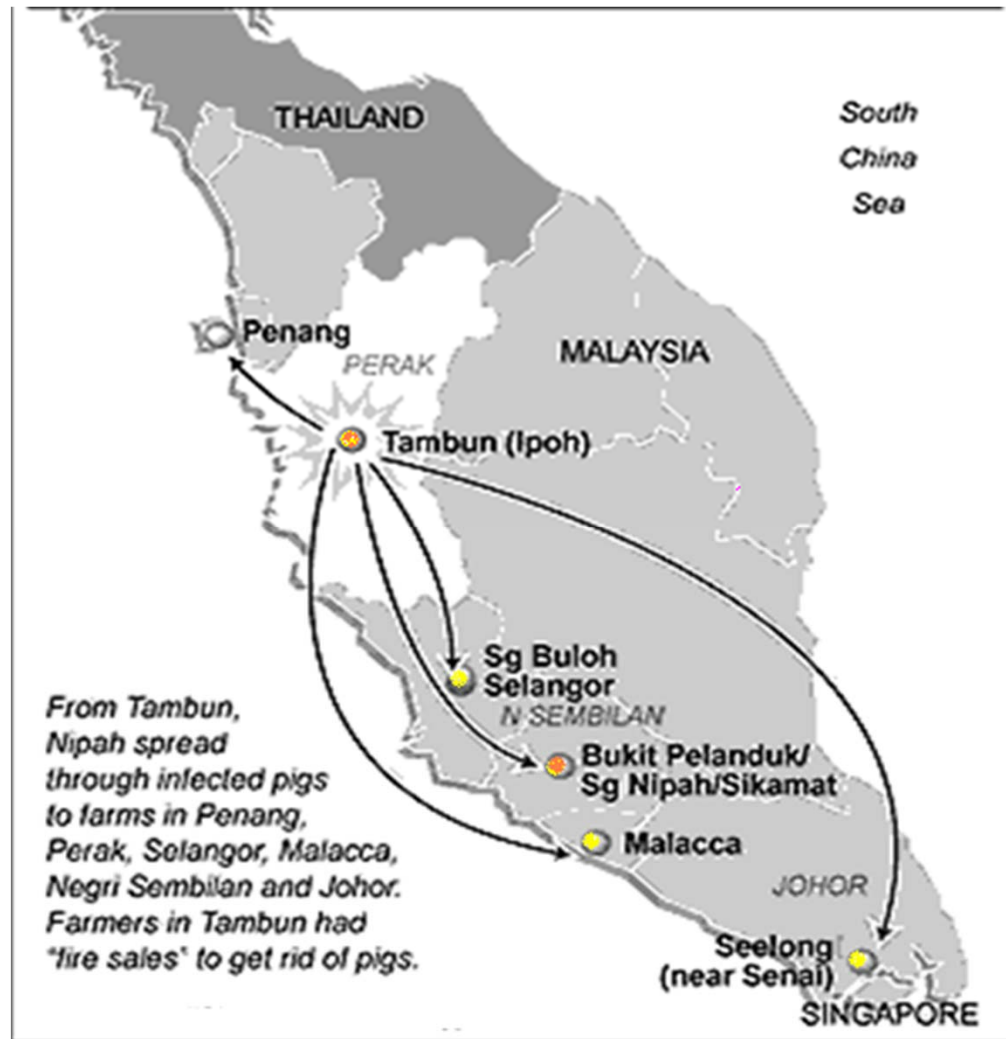


Figure 2. The Phylogenetic Tree of Partial M-gene Nucleotide Sequences of Siliguri (India) Nipah virus Isolates from the Bangladesh and Malaysia isolates (Source: Chadha et al., 2006)

Epidemiology of Nipah Virus

- The primary means of spread between farms and between regions was the movement of pigs.
- The primary mode of transmission on pig farms was via the oro-nasal route.
- Secondary modes of transmission between farms within localized farming communities - roaming infected dogs and cats, and sharing of boar semen, and lorries transporting pigs.
- The early epidemiology of the disease in Perak, and the spillover mechanism that first introduced the infection to pigs was thought to be from bats that eating fruits in the farm.
- Retrospective investigations suggest that Nipah virus has been responsible for sporadic disease in pigs in Peninsular Malaysia since late 1996, but was not recognized as a new syndrome because the clinical signs were not markedly different from those of several endemic pig diseases, and because morbidity and mortality were not remarkable .

The epidemic started in the northern Malaysian State of Perak, from where 'fire sales' dispersed pigs across the country



(Source: Cobey, S., 2005)



T A M B U N , I P O H P E R A K



DIAGNOSIS ON-FARM

- There are no pathognomonic features ascribable to Nipah virus disease in pigs.
- Differential diagnoses should include the following:
 - Classical swine fever
 - Porcine reproductive and respiratory syndrome
 - Aujeszky's disease (Pseudorabies)
 - Swine enzootic pneumonia (*Mycoplasma hyopneumoniae*)
 - Porcine pleuropneumonia (*Actinobacillus pleuropneumonia*)
 - Pasteurellosis

The clinical disease in pigs

- Infects pigs of all ages.
- Different clinical picture in different classes of animals. For example, sows were noted to present primarily a neurologic syndrome and porkers a respiratory syndrome.
- Proportion of pigs in a farm may not exhibit any clinical signs. Clinical disease in pigs can be very subtle and a large proportion of pigs in a farm may not exhibit any clinical signs.
- The incubation period was estimated to be 7 to 14 days.
- Neutralizing antibodies were detectable 10-14 days post-infection.

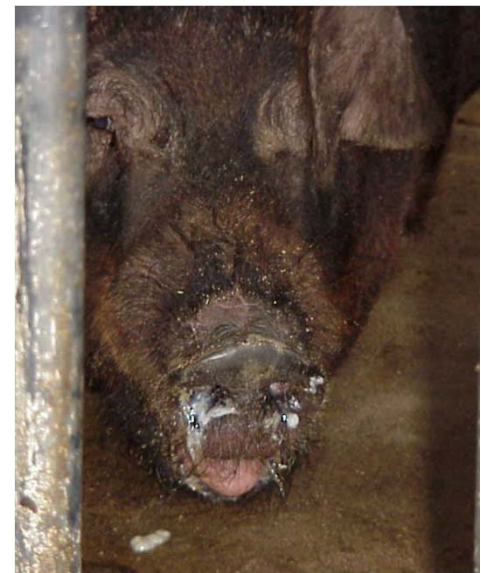


Clinical observations in weaners and porkers

- Acute febrile illness with respiratory signs ranging from rapid and labored breathing to harsh non-productive coughing.
- In severe cases, there was blood-tinged mucous discharge from the nostrils. In less severe cases, open mouth breathing was a feature.
- Neurological signs included trembling, twitching, muscular spasms, rear leg weakness and varying degree of lameness or spastic paresis.

Clinical observations in sows and boars

- Found dead overnight,
- Acute febrile illness with laboured breathing (panting), increased salivation and serous, mucopurulent or blood-tinged nasal discharge.
- Neurological signs including agitation and head pressing, tetanus like spasms and seizures, nystagmus, champing of mouth, and apparent pharyngeal muscle paralysis were observed.
- Abortions in affected sows.

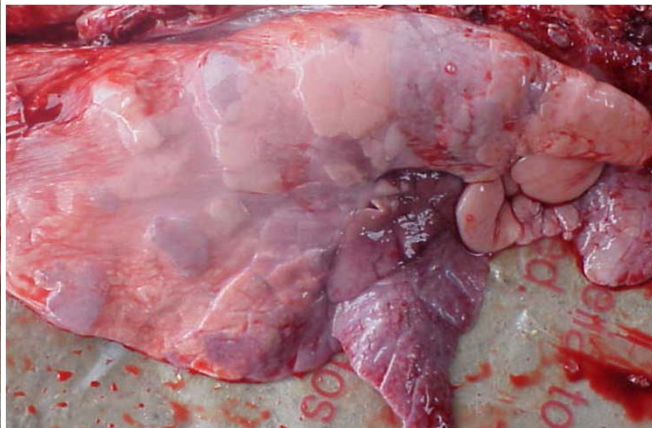
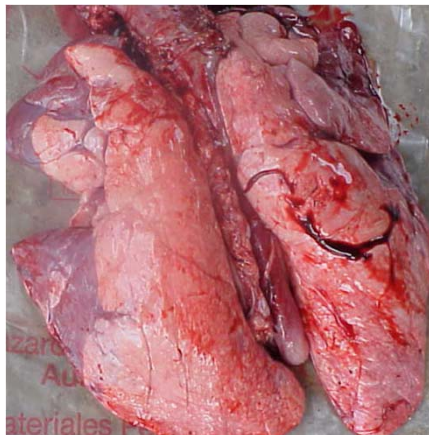


Clinical observations in suckling pigs

- Mortality of suckling pigs was estimated to be 40 percent.
- Most of the infected piglets showed symptoms of open mouth breathing, leg weakness with muscle tremors and neurologic twitches.

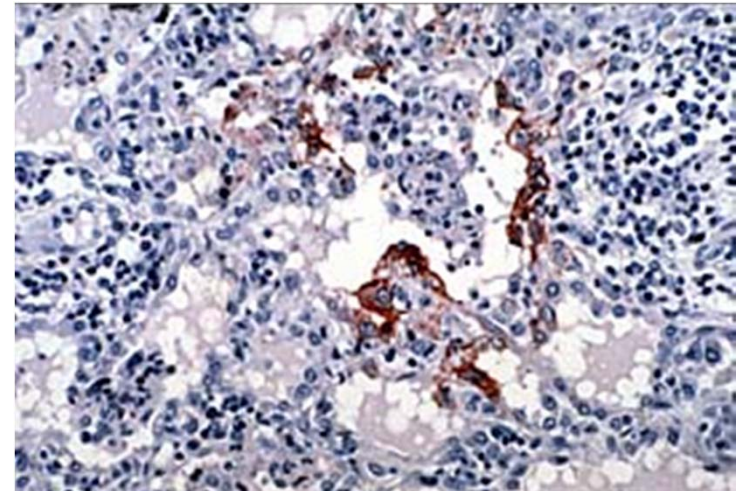
Necropsy findings in pigs

- The post-mortem findings in pigs are relatively non-specific. The lung and the meninges were the key organs affected.
- Mild to severe lung lesions with varying degrees of consolidation, emphysema and petechial-to-ecchymotic haemorrhages, and blood-tinged exudates in the airways.
- The interlobular septa were distended.
- Generalized congestion and oedema in the meninges.



CONFIRMATORY LABORATORY DIAGNOSIS

- Serology; SNT and ELISA
- Histopathology
 - formation of syncytia is a feature of Nipah virus (*Hooper et al. 2001*),
- Immunohistochemistry,
- Electron microscopy,
- Polymerase chain reaction
- Virus isolation.



Putative host

- Fruit bats of the genus *Pteropus* have been identified as a natural reservoir host of Nipah virus (*Johara et al. 2001*).
- Of 324 bats from 14 species surveyed, neutralizing antibodies to Nipah virus were found in 21 bats from five species (four species of fruit bats, including two flying fox species, and one insectivorous species). Only two flying fox species showed a substantial seroprevalence.
- Nipah virus was isolated from the urine of a free living colony of *Pteropus hyomelanus* in Malaysia (*Chua et al. 2001*).

Other species

- Peridomestic rodents, insectivores, and birds in Malaysia (*Asiah et al., unpublished data*) were negative serology for NiV indicate that these animals did not play a role as secondary reservoirs for Nipah virus.
- Dogs readily acquired infection following close association with infected pigs, targeted surveillance indicated that Nipah virus did not spread horizontally within dog populations.

The economic and social impacts of the outbreak

- The outbreak had a devastating impact on the pig industry in Malaysia. Most of the 257 human encephalitis cases and the 105 fatalities were pig industry people.
- Major economic costs have been incurred in controlling the outbreak, in lost domestic and export markets, and in allied businesses.

The economic and social impacts

	USD million
Compensation for the 1.1 million pigs destroyed	97
The control programme	136
Tax revenue estimated lost	105
Local pork consumption during the peak of the outbreak dropped by 80 percent	120
Exporting pigs to Singapore and Hong Kong.	124

The economic and social impacts

	Impacts
Approximately 618 homes and 111 shops, schools and banks, were evacuated.	It was estimated that 36 000 people involving farm workers and primary supporting services like drug and vaccine sales, feed and transport had suffered from the loss of employment due to closure of farms.
Prior to the outbreak, Malaysia had a standing pig population of 2.4 million.	During the stamping out operation an estimated 901,228 pigs from 896 farms were destroyed in the infected areas (Phase 1). A further 50 farms were culled under the national testing and surveillance programme (Phase 2).
Drastic change in the direction of the future of the pig industry in Malaysia.	Pig farming is now allowed only in “identified pig farming areas (PFA + better biosecurity) and with farmers in other areas encouraged to undertake other agricultural and livestock activities.

Outbreak CONTROL AND ERADICATION

- Operations require a high level of organization --- legislative, managerial, logistical, technical, and procedural activities.
- Encompassing plans for:
 - management activities (control centres, high level coordination, information management, laboratory preparedness);
 - control procedures (destruction and disposal of animals, valuation and compensation, decontamination of premises);
 - various livestock enterprises;



Movement controls on pigs

- After Nipah was declared as a disease under the legislation, all movement of pigs or pig meat (local, intrastate and interstate) was banned.
- Media releases and public notices were used to advise of the restrictions.
- The ban was enforced by increased DVS and police patrols on roads from infected areas.
- Movement of pigs outside declared zones to Government abattoirs was allowed, with each consignment transported under permit and escorted by DVS officers.

Mass culling of active disease farms

- Infected zones of 2 km radius and buffer zones of 10 km radius were imposed around infected premises. All pigs within the buffer zone were culled over a 2-month period (a total of 901, 228 pigs from 896 farms).
- The Department of Veterinary Services, the Department of Transport, the Army, other related government agencies and NGOs were involved in the culling operation.
- The pigs were culled by shooting, and disposed of by burying in deep pits within the infected area, either on-farm or off-farm.
- Chlorinated lime and detergents were used to disinfect premises and burial sites.

A national testing and surveillance programme

- A national testing and surveillance programme was implemented to determine the Nipah status of all pig farms in Peninsular Malaysia.



- The programme resulted in the culling of a further 50 seropositive farms (*Ong et al. 2000*),
- Freedom from Nipah virus infection in the swine population of Peninsular Malaysia in mid 2001.

A national abattoir monitoring and testing programme

- The third phase Nipah virus control and eradication programme involved ongoing monitoring of pigs sent to abattoirs.
- The programme incorporated a trace-back system based on ear-notch identification to allow pigs to be traced back to farms of origin.
- The porker class of pig was targeted for screening, as the presence of antibodies in pigs of this age denoted infection on the farm of origin within the last four months.
- The programme aimed to demonstrate that Nipah virus was not circulating on pig farms, and thus to restore public confidence in pork consumption.

Financial assistance

- The Malaysian Government approved establishments of two funds:
 1. *the Humanitarian Fund* - to relieve hardship caused by the loss of family members, and
 2. *the Nipah Trust Fund* - to provide financial assistance for the pigs culled.
- A committee headed by the Secretary General of Ministry of Agriculture operated the trust funds.
- The day to day management entrusted to the DVS Director General.

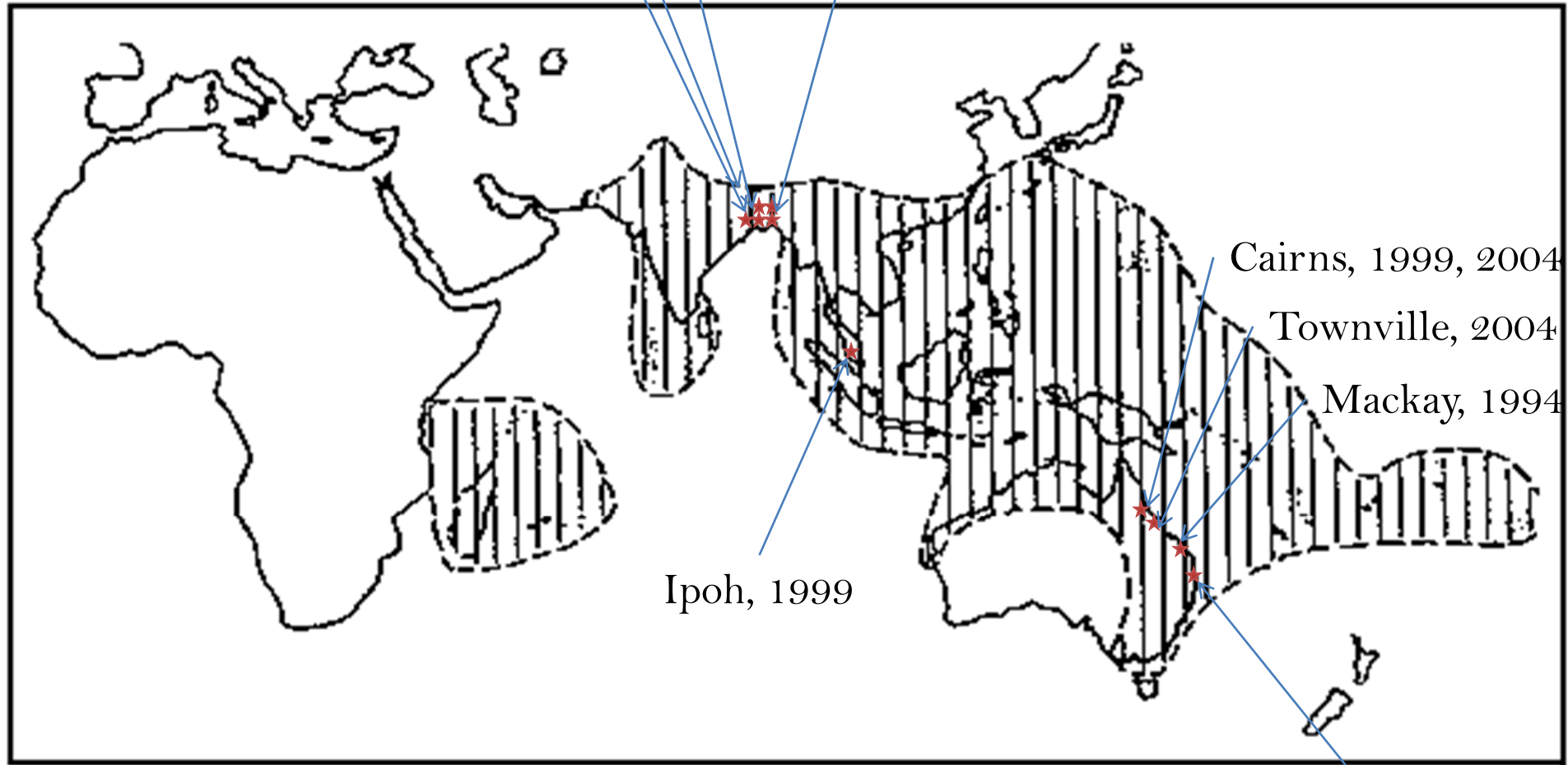


THE ROLE OF BATS IN THE EPIDEMIOLOGY OF NIPAH VIRUS INFECTION

- The world distribution of flying foxes (genus *Pteropus*) extends from the west Indian Ocean islands of Mauritius, Madagascar and Comoro, along the sub- Himalayan region of Pakistan and India, through South-east Asia, the Philippines, Indonesia, New Guinea, the South-west Pacific Islands (to the Cook Islands), and Australia .
- There are about 60 species of flying foxes in total. All flying fox species eat fruits, flowers or pollen, and roost communally in trees (*Hall & Richards 2000*).

Nadia, 2007
Siliguri, 2001
Naogaon, 2003
Meherpur, 2001

Manikganj, 2004
Faridpur, 2004, 2010, 2011
Tangail, 2005
Rajabri, 2011



Adapted from Hall and Richards, 2000

Brisbane, 1994
Sunshine Coast, 2006

NiV reservoirs 1

- In Malaysia, *Pteropus*, *P. vampyrus* (17%) and *P. hypomelanus* (31%), Small fruit bats, *Cynopterus brachyotis* (3.6%), and *Eonycteris spelaea* (5.3%), and one insectivorous species, *Scotophilus kulhi* (3%) (Johara et al., 2001).
- *P. hypomelanus* isolated NiV from pooled urine and a partially eaten fruit of a seropositive colony in Pulau Tioman (Chua et al. 2002a) .
- Serological and viral evidences of NiV in *Pteropus* spp. in Bangladesh, Cambodia, Indonesia, and Thailand (Olson et al., 2002, Harcourt et al., 2005; Wacharapluesadee et al., 2005; Chadha et al., 2006).

NiV reservoirs 2

- In Cambodia, 11.5% of *Pteropus lylei*'s tested were seropositive to NiV (Olson et al., 2002). NiV isolated from the urine of bats from the same colony has an isolation rate of $\approx 0.2\%$ (Reynes et al., 2005). NiV RNAs were also detected from the pooled saliva of *Hipposideros larvatus*.
- Molecular characterisation of the isolate demonstrated it was closely related (98%) to the Malaysian isolates (Reynes et al., 2005).



NiV reservoirs 3

- In Thailand, viral RNA was also found in saliva and urine of *P. lylei*, and saliva of the insectivorous Horsefield's roundleaf bat (*H. larvatus*).
- Seropositive bats to NiV were detected in *P. hypomelanus* (15.4%), *P. lylei* (9.3%), *P. vampyrus* (2.6%) and *H. larvatus* (1.3%).
- The isolate obtained from the saliva of *H. larvatus* was identical to that of the Malaysian, and the isolate obtained from urine and saliva of *P. lylei* was identical to that of the Bangladeshi (Wacharapluesadee et al., 2006).



NiV reservoirs 4



- In Indonesia, 35.7% serum samples of *P. vampyrus* neutralised NiV and 2.8% of that was also positive for HeV (Sendow et al., 2006).
- *P. giganteus* of India, (J. Epstein, Senior Epidemiology of the Consortium of Conservation Medicine, USA, unpublished data, 2007),
- In *P. giganteus* of Bangladesh, (Health and Science Bulletin, 2005) and in *Eidolon* sp. and *P. rufus* of Madagascar (lehle et al., 2007)

Factors that contribute to the emergence of NiV

- Establishment of pig farms within the range of natural host led to the initial introduction into the pig population;
- The maintenance of high densities of pigs led to the rapid dissemination of the infection within local pig populations; and
- the transport of pigs to other geographic areas for commerce led to the rapid spread of disease in pigs in southern Malaysia and Singapore.
- The presence of high density, amplifying host population facilitated transmission of the virus to human.

THANK YOU

Malaysia was declared free from
Nipah virus infection in mid 2001