



## The Review: Nipah Virus Infection

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Received: 13-05-2019; Revised: 18-06-2019; Accepted: 27-06-2019.

### ABSTRACT

Nipah virus is an emerging zoonosis with the potential to cause significant morbidity and mortality in humans and major economic and public health impacts. According to World Organisation for Animal Health (Office International des Épizooties: OIE), Nipah virus is a notifiable disease of importance to international trade. Nipah virus (NiV) are emerging zoonotic viruses that cause severe and often lethal respiratory illness and encephalitis in humans. Henipaviruses can infect a wide range of species and human-to-human transmission has been observed for NiV. While the exact route of transmission in humans is not known, experimental infection in different animal species suggests that infection can be efficiently initiated after respiratory challenge. The limited data on histopathological changes in fatal human cases of NiV suggest that endothelial cells are an important target during the terminal stage of infection; however, it is unknown where these viruses initially establish infection and how the virus disseminates from the respiratory tract to the central nervous system and other organs. MRI was useful in characterizing the disease in acute infection, as well as detection of spine abnormalities and subclinical infection. Here we review the current concepts in Nipah pathogenesis, clinical signs and symptoms, diagnosis, treatment, prevention and control in humans.

**Keywords:** Nipah; zoonotic; pathogenesis; respiratory tract; central nervous system.

### INTRODUCTION

Human Nipah virus (NiV) infection, an emerging zoonotic disease, was first recognized in a large outbreak of 276 reported cases in Malaysia and Singapore from September 1998 through May 1999. Almost all patients had contact with sick pigs and presented primarily with encephalitis; 39% died. Large fruit bats of *Pteropus* genus are the natural reservoir of NiV. Presumably, pig became infected after consumption of partially bat eaten fruits that dropped in pigsty. In 1994. In India, during 2001 and 2007 two outbreaks in human were reported from West Bengal, neighboring Bangladesh. Large fruit bats of *Pteropus* genus are the natural reservoir of NiV. Fruit bats of the *Pteropodidae* family are the natural host of NiV without apparent disease. They can shed the virus in their excretions and secretions, such as saliva, urine, semen and excreta. NiV infections have been reported in domestic animals, for example, pigs, which can act as the intermediate hosts for the transmission of the virus to humans.

Respiratory involvement including pneumonia has been found to be considerably more among patients in Bangladesh than Malaysia. This may be due to genetic diversity of the viral strains. The prominent respiratory involvement probably is responsible for human to human transmission.

It is important to develop guidelines for surveillance, diagnosis, case management, prevention and control of Nipah virus encephalitis so that human cases can be detected promptly and further human-to-human transmission can be prevented.

A host to which it seems well adapted. Illnesses caused by Nipah virus were first described in 1998-1999, during widespread outbreaks among pigs and people in Malaysia. The virus had apparently been transmitted from bats to pigs around 1996, and was thereafter maintained in swine populations. It was not detected immediately, as the mortality rate was low and the illness resembled other pig diseases. Nipah virus subsequently spread to pig farmers and abattoir workers in Malaysia and Singapore, causing severe, often fatal, encephalitis in more than 250 people. Some other species, including cats, dogs and goats, were also affected. The Malaysian outbreaks were controlled in both domesticated animals and humans by culling more than one million pigs. In addition, pig farming was permanently banned in some high- risk areas.

While Nipah virus encephalitis has not been documented in Malaysia since that time, human cases have been reported regularly in Bangladesh and a neighboring region of northern India since 2001. Many of these cases seem to be acquired directly from bats by drinking raw date palm sap, a widely consumed local delicacy. The sap is thought to become contaminated when bats visit and drink from unprotected sap collection sites at night. Person- to-person transmission also occurs after close, unprotected contact. How widely Nipah virus circulates in bats is still uncertain; however, viral RNA and seropositive bats have also been identified in areas where no clinical cases have ever been reported. A recent outbreak of neurological disease in horses and humans in the Philippines also appears to have been caused by virus.



## HISTORY OF NIPAH VIRUS<sup>5</sup>

Nipah virus is a previously unknown virus of the family Paramyxoviridae that has been identified primarily in humans and pigs in Malaysia. In humans, the virus causes fever, severe headache, myalgia, and signs of encephalitis or meningitis. The case fatality rate has been about 40%. The first human cases of disease attributed to Nipah virus occurred in late September 1998 in the northern city of Ipoh. The cases were first attributed to the Japanese encephalitis (JE) virus; however, the epidemiology of the disease was not consistent with JE. Most of the cases were in adult males who had direct contact with pigs. In March 1999, Malaysian researchers identified the virus as a previously unknown paramyxovirus. This was confirmed by the CDC. The virus was first called Hendra-like virus because it is similar to virus, first identified in horses in Australia in 1994. The virus is now named after the village near Kuala Lumpur from where it was first isolated. As of late April 1999, 257 cases of febrile encephalitis had been reported in Malaysia, including 100 deaths.

## TRANSMISSION AND RESERVOIR OF THE NIPAH VIRUS<sup>6-9</sup>

In Pteropus bats, Nipah virus has been found repeatedly in urine, and viral RNA has been detected rarely in oropharyngeal swabs and rectal swabs from naturally or experimentally infected bats. It has also been found in fruit that had been partially eaten by bats. Despite high seroprevalence rates, only a few bats in a colony may shed the virus at any given time, and excretion from the colony may be sporadic. Fruit bats (Pteropodidae family) have been identified as the reservoir for HNV

How bats transmit this virus to domesticated animals is uncertain, but ingestion of contaminated fruit, water, or aborted bat fetuses or birth products (e.g., by pigs) is suspected. Nipah virus is highly contagious in swine, which can act as amplifying hosts and shed this virus in respiratory secretions and saliva. Experimental infections suggest that shedding may start as early as 2 days after infection and persist for up to 3 weeks. Nipah virus appeared to be transmitted within a farm by aerosols and direct contact between pigs; virus spread between farms was usually associated with pig movements. Although this virus has not been reported, to date, in the urine of pigs, it can occur in the kidneys, and exposure to pig urine is a risk factor for human infections. Anecdotal evidence suggests that vertical transmission may occur across the placenta. Transmission in semen may be possible, and re-used vaccination needles may have contributed to the spread of the virus between pigs in Malaysia.

Humans can be infected by direct contact with infected swine, probably through the mucous membranes, but possibly also through skin abrasion. Humans can shed Nipah virus in respiratory secretions, saliva, and urine, and contact with respiratory secretions is thought to be the main route of spread. Some people also became ill after unprotected contact with deceased patients. How long Nipah virus can remain viable in the general environment

is uncertain; however, it can survive for up to 3 days in some fruit juices or mango fruit, and for at least 7 days in artificial date palm sap (13% sucrose and 0.21% BSA in water, pH 7.0) held at 22°C. This virus is reported to have a half-life of 18 hours in the urine of fruit bats

### a. The Mode of Transmission

Infected bats shed virus in their excretion and secretion such as saliva, urine, semen and excreta but they are symptomless carriers. The NiV is highly contagious among pigs, spread by coughing. There is strong evidence that emergence of bat-related viral infection communicable to humans and animals has been attributed to the loss of natural habitats of bats. As the flying fox habitat is destroyed by human activity the bats get stressed and hungry, their immune system gets weaker, their virus load goes up and a lot of virus spills out in their urine and saliva. Similar fluctuation of virus shedding may be associated with the stressful physiological conditions or seasons. Evidence of seasonal preference of transmission in *P. lylei* was recently demonstrated in a study in Thailand. The period April-June was the time (highest in May) when viral RNA could be mainly detected in urine which was associated with a fluctuation of population numbers that was observed only in May and correlated with young bats leaving to fly. There were focal outbreaks of NiV in Bangladesh and India in 2001 during winter. Drinking of fresh date palm sap, possibly contaminated by fruit bats (*P. giganteus*) during the winter season, may have been responsible for indirect transmission of Nipah virus to humans.

There is circumstantial evidence of human-to-human transmission in India in 2001.

During the outbreak in Siliguri, 33 health workers and hospital visitors became ill after exposure to patients hospitalized with Nipah virus illness, suggesting Nosocomial infection.

During the Bangladesh outbreak the virus is suggested to have been transmitted either directly or indirectly from infected bats to humans. Strong evidence indicative of human-to-human transmission of NiV was found in Bangladesh in 2004.

### b. Agent

The nucleotide sequences of NiV strains isolated from pigs and persons were remarkably similar and suggest that the entire outbreak was caused by 1 or 2 closely related strains. Indeed, all human cases of NiV infection inoriginated from a single or perhaps 2 introductions of NiV from its bat reservoir into pigs. NiV is a highly pathogenic paramyxovirus belonging to genus Henipavirus. It is an enveloped RNA virus.

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infection in Malaysia and Singapore could have originated from a single or perhaps 2 introduction of NiV from its bat reservoir into pigs.

In Bangladesh, by contrast, recurrent Nipah outbreaks have been recognized since 2001 and the strains of Nipah isolates show substantial heterogeneity in their nucleotide sequences. This heterogeneity suggests repeated introductions of Nipah virus from its host reservoir into the human population in Bangladesh.

Nipah cases tend to occur in a cluster or as an outbreak, although 18% cases in Bangladesh were isolated. There is strong evidence that the emergence of bat-related viral infection communicable to humans and animals has been attributed to loss of natural habitat of bats. It has been speculated that migratory fruit bats were forced away from their natural habitats in 1998 because of forest fires prevalent at that time in the region and attracted by the fruit trees in pig farms. As the flying fox habitat is destroyed by human activity the bats become stressed, their immune system weakens, their viral load increases and more virus is shed in the urine and saliva.

#### c. Incubation Period

The median incubation period of the secondary cases who had a single exposure to Nipah case was nine days (range 6–11 days) but exposure to onset of illness varies from 6–16 days. The median incubation period following single intake of raw date palm sap to onset of illness is 7 days (range: 2-12 days).

The pathologic findings in the brain of Nipah encephalitis cases showed evidence of necrotizing vasculitis. There was widespread central nervous system (CNS) involvement due to severe vasculitis of mainly small blood vessels, which resulted in endothelial damage. Eosinophilic cytoplasmic and nuclear viral inclusions were detected in many neurons adjacent to vasculitic vessels, a finding which is present in infections caused by other paramyxoviruses. The main pathology appeared to be widespread ischemia and infarction caused by vasculitis-induced thrombosis, although direct neuronal invasion may also play a major role in the pathogenesis of the encephalitis.

Alveolar hemorrhage, pulmonary edema and aspiration pneumonia were often encountered in the lung. These may lead to pneumonia and acute respiratory distress syndrome (ARDS) ultimately.

#### d. Surveillance

Beginning in 2006, The Institute of Epidemiology, Disease Control and Research (IEDCR) in collaboration with ICDDR, B established Nipah surveillance in 10 District level Government hospitals of the country where Nipah outbreaks had been identified. Presently surveillance system is functioning in five hospitals. Establishing appropriate surveillance systems are necessary to detect NiV outbreaks quickly and appropriate control measures can be initiated.

#### e. Geographical Distribution of NiV

Outbreaks of NiV in Southeast Asia have a strong seasonal pattern, usually during winter and spring from December to May, and a limited geographical range. This could be related to the breeding season of the bats, increased shedding of virus by the bats and the date palm sap harvesting season. Apart from the four countries with reported human NiV outbreaks (Bangladesh, India, Malaysia and Singapore), other countries in which Pteropus fruit bats live are at potential risk of NiV infection. The distribution of these bats extends from the east coast of Africa, across South and Southeast Asia, east to the Philippines, Pacific islands and Australia. Nipah virus can emerge as a human pathogen anywhere in these distribution areas. Countries/areas with serological evidence or molecular detection in the natural reservoir (Pteropus bats species) and several other bat species include Bangladesh, Cambodia, Mainland China, Ghana, India, Indonesia, Madagascar, Papua New Guinea, the Philippines, Taiwan and Thailand.

#### f. The Latest Outbreak in India

Outbreak of NiV infection was reported in the Southern State of Kerala in India. At the beginning, three deaths in the same family were reported in the Kozhikode District of Kerala. Field investigation revealed that there were bats living in an abandoned water well on the.

In response to the outbreak, a multi-disciplinary team led by the Indian Government's National Centre for Disease Control (NCDC) has been formed in Kerala and the World Health Organization (WHO) has also provided technical support to the Government of India as needed. Contact tracing has been initiated. Infection prevention and control measures have been strengthened in health facilities. Relevant sectors including animal health, wildlife and environment sectors have been involved to establish the origin and spill-over of the disease from animal to human. Risk communication messages were being delivered to the community, public and stakeholders to increase their awareness of the disease. So far, the disease has not spread to new areas. The WHO considered that the outbreak was localised at the moment and the risk was low at the national and regional levels

#### 4. ETIOLOGY<sup>09,10</sup>

Nipah virus (NiV) is a single-stranded RNA virus belonging to the genus Henipavirus, family Paramyxoviridae, which was discovered in Malaysia in 1998– 1999. It is closely related to Hendra virus. There are two lineages: a Malaysian strain (mNiV) and a Bangladesh strain (bNiV). Nipah virus is a member of the genus Henipavirus in the family Paramyxoviridae. This genus also includes Hendra virus. There seem to be multiple strains of Nipah virus. At least two major strains were isolated from pigs in Malaysia, and the strains that cause human cases in Bangladesh and India differ from outbreak strains isolated in Malaysia.





**SPECIES AFFECTED**<sup>9,10</sup>

Fruit bats of the genus *Pteropus* (flying foxes) are the main reservoir hosts for Nipah virus. *P. vampyrus*, the Malayan flying fox, and *P. hypomelanus*, the island flying fox, are known to carry this virus in Malaysia. *P. giganteus* is thought to be an important host in Bangladesh and India and possibly other locations. Although live virus has not yet been isolated from this species, Nipah virus RNA has been detected and many bats are seropositive. Viral RNA and/or antibodies have been found in a few other species of fruit or insectivorous bats, although their significance is unclear.

Many domesticated mammals seem to be susceptible to Nipah virus. This virus can be maintained in pig populations, but other domesticated animals appear to be incidental (spillover) hosts. Sick goats, dogs, cats and horses were observed in the outbreak area in Malaysia, and infections in dogs, a cat, a horse and goats were confirmed by immunohistochemistry. Experimental infections with Nipah virus have established in pigs, cats, ferrets, nonhuman primates, guinea pigs, golden hamsters (*Mesocricetus auratus*) and mice.

**a. Zoonotic Potential**

Nipah virus can cause serious illnesses in people. A number of cases have been linked to drinking raw date palm sap, which had probably been contaminated by bats. Drinking fermented date palm sap (alcohol content approximately 4%) appeared to be a risk factor in a few cases. Zoonotic cases were acquired from pigs in Malaysia (bat to human transmission appears to be uncommon or absent in sick animals of other species (a dog, various livestock), but the evidence in these cases was speculative and/or circumstantial.

**b. Epidemiology**

The morbidity and mortality data of human NiV infection is presented in Table Case fatality rate of NiV ranges from 40-70% although it has been as high as 100% in some outbreaks. Distribution of bat species previously shown to have Nipah virus.

The known geographic distribution of NiV correlates with the range of reservoir species—flying foxes—including Australia and Southeast Asia to India and the Eastern African islands. Annual outbreaks occur in India and Bangladesh. Pigs are an amplifying host for NiV. Dogs and cats are susceptible but are not thought to be involved in transmission. NiV is zoonotic and causes high fatality rates in humans.

**GLOBAL EPIDEMIOLOGY**

There have been epidemics of meningococcal disease in Asia, Europe and the Americas, but the largest and most frequently recurring outbreaks occurred in sub-Saharan Africa. Epidemics occur in seasonal cycles during the dry season from December to June in the African meningitis belt which stretches across the continent from Senegal to Ethiopia. Major epidemics occur every five to 12 years in

this region, during which the attack rates can reach 1 000 cases per 100 000 persons in affected areas. Other regions of the world also have occasional outbreaks but significantly lower incidence of less than three cases per 100 000 population.

**Table 1:** Distribution of species in different geographical areas

Species	Geographic range
<i>Pteropus</i>	Australia; Cambodia; Indonesia; <b>Malaysia*</b> ; Maldives;
<i>hypomelanus</i>	Myanmar; Papua New Guinea; Philippines; Solomon Islands; <b>Thailand</b> ; Viet Nam
<i>Pteropus vampyrus</i>	Brunei Darussalam; <b>Cambodia</b> ; Indonesia; <b>Malaysia</b> ; Myanmar; Philippines; <b>Thailand</b> ; Tonga; Vanuatu
<i>Pteropus lylei</i>	<b>Cambodia*</b> ; <b>Thailand#</b> ; Viet Nam
<i>Pteropus giganteus</i>	<b>Bangladesh</b> ; China; <b>India</b> ; Maldives; Nepal; Pakistan; Sri Lanka
<i>Eonycteris spelaea</i>	China; India (Andaman Is., Andhra Pradesh, Assam, Karnataka, Manipur, Meghalaya, Nagaland, Nicobar Is., Sikkim, Tamil Nadu, Uttaranchal); Indonesia; <b>Malaysia</b> ; Myanmar; Philippines; Thailand
<i>Cynopterus</i>	Cambodia; China; India (Andhra Pradesh, Bihar, Goa, Karnataka, Maharashtra, Nagaland, Tamil Nadu); Indonesia (Sulawesi, Sumatra); Lao People's Democratic Republic; <b>Malaysia</b> ; Myanmar; Nepal; Philippines; Singapore; Sri Lanka; Thailand; Viet Nam
<i>Scotophilus kuhlii</i>	Bangladesh; India; Indonesia; <b>Malaysia</b> ; Pakistan; Philippines; Sri Lanka
<i>Hipposideros larvatus</i>	Bangladesh; Cambodia; China; India; Indonesia (Bali, Jawa, Kalimantan, Sumatra); Lao People's Democratic Republic; Malaysia (Peninsular Malaysia, Sabah, Sarawak); Myanmar; <b>Thailand#</b> ; Viet Nam

According to the World Health Organization (WHO), there are no reliable estimates of global meningococcal disease burden due to inadequate surveillance in several parts of the world. Of the *N. meningitidis* serogroups identified, serogroups A, B, C, X, W, and Y are responsible for the majority of the disease, but serogroup distribution varies by location and time. serogroups C, W and X.

**Table 2:** Morbidity and mortality due to Nipah or Nipah-like virus, Asia-Pacific Region 1998-2008

Year/Month	Location	No. cases	No. deaths	Case Fatality
Sep 1998 - Apr 99	Malaysia (Perak, Selangor and Negeri Sembilan states)	265	105	40%
Mar 1999	Singapore	11	1	9%
Feb 2001	Siliguri (India)	66	45	68%
Apr-May 2001	Meherpur, Bangladesh	13	9	69%
Jan 2003	Naogaon, Bangladesh	12	8	67%
Jan 2004	Goalando, Bangladesh	29	22	76%
Apr 2004	Faridpur, Bangladesh	36	27	75%
Jan-Mar 2005	Tangail, Bangladesh	12	11	92%
Jan-Feb 2007	Thakurgaon, Bangladesh	7	3	43%
Mar-Apr 2007	Kushtia, Bangladesh	8	5	63%
April 2007	Nadia, India	5	5	100%
Feb 2008	Manikgonj and Rajbari,	11	6	55%
Apr 2008	Shatkira and Jessore	2	1	50%
<b>Total</b>		<b>477</b>	<b>248</b>	<b>52%</b>



Dry and dusty conditions during the dry season between December. In the Americas, Australia and Europe, majority of cases were accounted by serogroups B, C and Y, though increasing numbers of serogroup W have been observed in some areas. In Asia, five major serogroups (A, B, C, W and Y) were reported to be variedly present in different countries.

#### c. Species Affected

Flying foxes, also known as fruit bats, are the main reservoir host for NiV. Flying foxes roost in forests and swamps, flying to fruit trees at night to feed. Neutralizing antibodies to NiV have been found in many of the flying foxes in Southeast Asia, including 75% of sampled flying foxes. Neutralizing antibodies to NiV.

Infected pigs can be amplifying hosts. Dogs and cats are also susceptible to NiV infection, but are not believed to be capable of transmitting NiV to humans or other animals.

#### d. Morbidity and Mortality

During the Malaysian outbreak, there was 10–15% mortality in piglets.<sup>3</sup> Mortality occurs in less than 5% of growing pigs.

### 4. CLINICAL SIGNS AND SYMPTOMS<sup>11,12</sup>

Although some Nipah virus infections can be asymptomatic or mild, most recognized clinical cases have been characterized by respiratory disease and/or acute neurological signs. The initial symptoms are flu-like, with fever, headache, sore throat and myalgia. Nausea, vomiting and a nonproductive cough may also be seen. This prodromal syndrome may be followed by encephalitis, with symptoms such as drowsiness, disorientation, signs of brainstem dysfunction, convulsions, coma and other signs. Nipah virus infections in some patients appear as respiratory disease, including atypical pneumonia or acute respiratory distress syndrome. These patients may or may not develop neurological signs. Septicemia, bleeding from the gastrointestinal tract, renal impairment and other complications are possible in severely ill patients. Survivors of encephalitis may have mild to severe residual neurological deficits, or remain in a vegetative state.

Some people infected with Nipah virus develop relapsed encephalitis or late-onset encephalitis, months or years later. The latter syndrome occurs in a person who was initially asymptomatic or had a non-neurological illness. The clinical signs usually develop acutely, with symptoms that may include fever, headache, seizures and focal neurological signs. Some cases are fatal.

Mortality is not common in sows or boars affected by NiV, although adults can die suddenly with no symptoms or within 24 hours after the onset of symptoms. Neurological signs seen in adults include tongue drooping, frothy salivation, head pressing, agitation, titanic spasms and seizures, and pharyngeal muscle paralysis. Bloody nasal discharge can occur post mortem, and abortions have also been observed.

### a. CASE MANAGEMENT OF NIPAH ENCEPHALITIS

#### • Suspected case

A person fulfilling both of the following criteria is defined as a suspected case:

1. Features of acute encephalitis as demonstrated by
    - a. Acute onset of fever AND
    - b. Evidence of acute brain dysfunction as manifested by
      - i. Altered mental status OR
      - ii. New onset of seizure OR
      - iii. Any other neurological deficit
  2. Epidemiological linkage
    - a. Drinking raw date palm sap OR
    - b. Occurring during Nipah season OR
    - c. Patient from Nipah endemic area
- Probable case

A person with features of acute encephalitis

- a. During a Nipah outbreak in the area OR
- b. With history of contact with confirmed Nipah patient

In both suspected and probable cases, the patient might present with respiratory features with or without encephalitis. The respiratory features are, Illness < 7 days duration and Acute onset of fever and Severe shortness of breath, cough and Chest radiograph showing diffuse infiltrates.

• Confirmed case-A suspected or probable case with laboratory confirmation of Nipah virus infection either by:

- a. IgM antibody against Nipah virus by ELISA in serum or cerebrospinal fluid
- b. Nipah virus RNA identified by PCR from respiratory secretions, urine, or cerebrospinal fluid

#### • Definition of Cluster

Two or more suspect cases living within a 30 minute walk of each other who develop symptoms within 21 days of each other.

#### • Clinical features

The following symptoms were observed (in order of frequency in Bangladeshi cases) Fever, Altered mental status, Severe weakness, Headache, Respiratory distress, Cough, Vomiting, Muscle pain, Convulsion, Diarrhoea.

#### • General Signs

Reduced GCS score, Raised temperature, Increased respiratory rate (Adult:  $\geq 25$ /min; children of  $\geq 12$  months:  $\geq 40$ /min), Increased heart rate (Adult:  $\geq 100$ /min; children of  $\geq 12$  months:  $\geq 140$ /min), Crepitations in lung Hypertension/Hypotension.



- Neurological signs

Oculoparesis, Pupillary abnormality, Facial weakness, Bulbar weakness, Limb weakness, Reduced deep tendon reflexes, Plantar-absent/extensor. Infection with Nipah virus is associated with encephalitis (inflammation of the brain). After exposure and an incubation period of 5 to 14 days, illness presents with 3-14 days of fever and headache, followed by drowsiness, disorientation and mental confusion. These signs and symptoms can progress to coma within 24-48 hours. Some patients have a respiratory illness during the early part of their infections, and half of the patients showing severe neurological signs showed also pulmonary signs.

## 5. DIAGNOSIS<sup>12,13</sup>

Laboratory diagnosis of a patient with a clinical history of NiV can be made during the acute and convalescent phases of the disease by using a combination of tests. Virus isolation attempts and real time polymerase chain reaction (RT-PCR) from throat and nasal swabs, cerebrospinal fluid, urine, and blood should be performed in the early stages of disease. Antibody detection by ELISA (IgG and IgM) can be used later on. In fatal cases, immunohistochemistry on tissues collected during autopsy may be the only way to confirm a diagnosis.

Virus isolation is required for definitive diagnosis, but handling NiV requires a BSL4 laboratory.

Quantitative real-time polymerase chain reaction is available, as is immunohistochemistry. Immunofluorescence may be complicated by cross-reactivity with Hendra virus, although several monoclonal antibodies are now available.

An indirect enzyme linked immunosorbent assay (ELISA), virus neutralization tests using pseudotype particles, and multiplexed microsphere assays have all been developed to detect antibodies to NiV at the BSL2 level.

- Differential Diagnosis
  1. Other viral encephalitides
  2. Bacterial meningitis
  3. Cerebral malaria

- **IMMUNITY**

Multiple vaccines are in development but none are commercially available. Vector- based vaccines include those utilizing canarypox virus, Newcastle disease virus, and vesicular stomatitis virus. The Hendra virus vaccine used in horses has shown some cross-protection against NiV in animal models.

- **LABORATORY DIAGNOSIS**

Procedures for the laboratory diagnosis of NiV include serology, histopathology PCR and virus isolation. Serum Neutralization Test, ELISA, RT-PCR are used for laboratory confirmation.

Most countries in the South-East Asia Region do not have adequate facilities for diagnosing the virus or on ways of controlling it. Bangladesh, India and Thailand have developed laboratory capacity for diagnostic and research purposes. Nipah virus is classified internationally as a biosecurity level (BSL) 4 agent. BSL 2 facilities are sufficient if the virus can be first inactivated during specimen collection.

- **CLINICAL HISTORY**

Disease and fatalities in people may be one of the first signs of a NiV outbreak. Symptoms associated with NiV-induced encephalitis in humans include fever, headache, dizziness, vomiting, and progression to impaired consciousness. Swine history will include respiratory and neurological symptoms with relatively low mortality in affected populations.

Nipah virus infections can be diagnosed by virus isolation, the detection of antigens or nucleic acids, and serology. Histopathology also aids diagnosis. In swine, Nipah virus has been detected in respiratory secretions, blood and various tissues including the bronchial and submandibular lymph nodes, lung, spleen, kidney and brain. In experimentally infected cats, this virus has been found in the lung and spleen, and less often, in the kidney, lymph nodes and other organs. It can also be detected in feline blood, urine and respiratory secretions. As Nipah virus is a BSL4 pathogen and must be cultured under high-security conditions. Electron or immunoelectron microscopy may also be helpful. Molecular methods (e.g., RT- PCR), comparative immunostaining or differential neutralization assays can distinguish.

- **TESTS TO DETECT NUCLEIC ACIDS, VIRUS, OR ANTIGENS**

Immunohistochemistry (IHC) can be performed to detect NiV. The N protein antigen is commonly targeted. Detection of phosphoprotein (P) antigen can also be used with IHC, although N protein antigen is expressed in greater quantities than P protein antigen and is therefore of better diagnostic value.

Immunofluorescence can rapidly detect NiV viruse, since mono-specific anti sera to individual proteins of NiV Two monoclonal antibodies with affinity for the N protein or P, V, and W protein of henipaviruses have been developed

- **TESTS TO DETECT ANTIBODY**

Multiplexed microsphere immunoassays have been developed to detect either antibody binding to recombinant soluble protein, or antibody inhibition of receptor binding. Spectrally distinct microspheres allow specific and sensitive quantification and differentiation between HeV and NiV antibodies in a sample. Because of the recombinant subunit proteins used, this assay does not have to be conducted in a BSL4 lab if the samples are treated to inactivate virus.



### ▪ **SAMPLE**

Oro-pharyngeal/nasal swabs, urine, and serum can be used for isolation from live animals, while brain, lung, kidney, and spleen samples can be used post mortem. If possible, urine should also be collected for analysis. Strict biosecurity protocols, including stringent use of personal protective equipment, should be followed when sampling pigs with suspected NiV infection.

### ▪ **POST-EXPOSURE**

Neutralizing antibody titers appear 7–10 days post-experimental infection with maximum titers seen 14–16 days post-infection.

#### • **Incubation Period**

Clinical cases in humans usually become apparent several days to 14 days after exposure; however, incubation periods as short as 2 days or as long as a month or more have been reported. Some people with mild or subclinical infections can develop late-onset encephalitis months or years later. One such case occurred after 11 years.

#### • **Standard case definition**

The clinical case definitions for Nipah are highly context dependent. The particular symptoms of Nipah are non-specific, but in the context of an outbreak they efficiently identify persons at risk for Nipah infection.

#### • **Suspect Nipah patient**

Person from a community affected by an outbreak who has:

- 1) fever with new onset of altered mental status or seizure and/or
- 2) fever with headache and/or
- 3) cough with shortness of breath.

#### • **Probable Nipah patient**

Suspect case-patients who resided in the same village where confirmed case-patients were living during the outbreak period and who died before complete diagnostic specimens could be collected.

#### • **Confirmed Nipah Patient**

Person who has laboratory confirmation of Nipah virus infection either by:

- 1) IgM antibody against Nipah virus identified in serum or cerebrospinal fluid.
- 2) Nipah virus RNA identified by PCR from respiratory secretions, urine, or cerebrospinal fluid.

### **6. TREATMENT IN NIPAH VIRUS INFECTION<sup>14,15,16</sup>**

Treatment is supportive, with some patients requiring measures such as mechanical ventilation. Ribavirin appeared to be promising in some outbreaks, but had little or no effect on the outcome in animal models, and its

efficacy is currently considered to be uncertain. Other potential treatments, such as the administration of antibodies to Nipah virus, are being investigated in preclinical studies.

#### • **MRI Features of Acute Nipah Encephalitis**

All patients had multiple small (less than 1 cm in maximum diameter) bilateral abnormalities within the sub cortical and deep white matter; in some patients, the cortex, brainstem, and corpus callosum were also involved

DW MRI is capable of depicting acute cytotoxic oedema in the clinical assessment of acute cerebral infarction.<sup>16</sup> In Nipah virus patients with acute infection, DW MRI, supported by contrast enhancement, was helpful in confirming that the effects of acute viral infection were responsible for these lesions, and that they were not pre-existing abnormalities caused by ageing or other non-virus related causes.

#### • **Re-Emergence and Threat**

MRI has been helpful in showing that the henipaviruses can cause different patterns including acute vasculitis-associated cerebral infarction and relapsed and late-onset encephalitis, probably representing different pathological processes. Although it has been 10 years since the initial Malaysian outbreak, the continued re-emergent clusters of Bangladeshi Nipah cases

- Cleaning and Disinfection
- NiV survival in the environment is unclear; however, infections seem most common in cool, dry weather in India and Bangladesh.
- NiV can be inactivated by 0.1% formalin and 0.5% household bleach. In general, paramyxoviruses are susceptible to acids, alcohols, aldehydes, alkalis, halogens, and oxidizing agents. NiV has limited susceptibility to biguanides, phenolic compounds, and quaternary ammonium compounds.

Like other paramyxoviruses, Nipah virus is readily inactivated by soaps, detergents and many disinfectants. Routine cleaning and disinfection with sodium hypochlorite or commercially available disinfectants is expected to be effective. Sodium hypochlorite was recommended for the disinfection of pig farms in Malaysia.

#### ▪ **Treatment**

Treatment is limited to supportive care. Because Nipah virus encephalitis can be transmitted person-to-person, standard infection control practices and proper barrier nursing techniques are important in preventing hospital-acquired infections (nosocomial transmission). The drug ribavirin has been shown to be effective against the viruses in vitro, but human investigations to date have been inconclusive and the clinical usefulness of ribavirin remains uncertain.





Passive immunization using a human monoclonal antibody targeting the Nipah G glycoprotein has been evaluated in the post-exposure therapy in the ferret model and found to be of benefit

#### ▪ Entry in the CNS

HNV infection of the CNS and the development of neurological signs are associated with the disruption of the blood-brain barrier (BBB) and expression of TNF- $\alpha$  and IL-1 $\beta$ . These pro-inflammatory cytokines have been shown to play a role in increasing the permeability of the blood-brain barrier. Nipah virus nucleoprotein is detected in bronchial epithelium

(A) and endothelium (B) in the lungs, neurons in the brain (C) and olfactory epithelium (D) in the nasal turbinates of ferrets intranasally infected with the Malaysia strain of Nipah virus. 40X magnification.

Induction of neuronal injury and death While the source of TNF- $\alpha$  and IL-1 $\beta$  expression in the brain is currently unknown, they can be released by microglia, which are also infected by HNV. However, whether disruption of the BBB is a direct cytopathic effect of virus replication in the microvasculature or an indirect effect through expression of TNF- $\alpha$  and IL-1 $\beta$  by bystander cells such as neurons and microglia remains unclear.

#### a. Vaccines

Multiple vaccines have been developed for NiV, though none are commercially available. A HeV soluble G subunit-based vaccine has shown promise in preventing infection in animals exposed to lethal doses of NiV. A vaccination study has shown the HeV soluble G vaccine capable of preventing NiV from causing disease in ferrets

Recombinant Newcastle disease virus-vectored NiV vaccines expressing the G and F proteins of NiV have shown to be experimentally effective in producing long-lasting neutralizing NiV antibodies in pigs. This vaccine requires a booster four weeks after the initial vaccination to produce adequate neutralizing titers. Advantages of this vaccine are the ability to easily culture the vaccine virus in chicken eggs and the amenability to lyophilization, facilitating both production and storage of the vaccine.

A recombinant vesicular stomatitis virus (rVSV) vector vaccine expressing NiV G or F protein has been shown to induce a strong humoral anti-NiV response in hamsters following a single vaccine dose.

#### • Cross-protection

It is likely that novel henipaviruses induce NiV cross-reactive antibodies. There is good cross-protection provided by the G protein of HeV. Passive administration of monoclonal antibodies to HeV G protein have prevented disease in ferrets challenged with a lethal dose of NiV. Vaccinations using HeV soluble G protein have been found to protect against challenge with live NiV.

Nipah virus (NiV) is a member of the family Paramyxoviridae, genus Henipavirus.

NiV was initially isolated and identified in 1999 during an outbreak of encephalitis and respiratory illness among pig farmers and people with close contact with pigs in Malaysia and Singapore.

#### • Risk of Exposure

In the Malaysia and Singapore outbreak, Nipah virus infection was associated with close contact with Nipah virus-infected pigs.

In Bangladesh and India, where Nipah virus infection is more frequent, exposure has been linked to consumption of raw date palm sap and contact with bats. Importantly, human-to-human transmission has been documented and exposure to other Nipah virus infected individuals is also a risk factor.

### 7. PREVENTION AND CONTROL<sup>17,18</sup>

There is no effective treatment for Nipah virus disease, but ribavirin may alleviate the symptoms of nausea, vomiting, and convulsions. Treatment is mostly focused on managing fever and the neurological symptoms. Severely ill individuals need to be hospitalized and may require the use of a ventilator. Human-to-human transmission of NiV has been reported in recent outbreaks demonstrating a risk of transmission of the virus from infected patients to healthcare workers through contact with infected secretions, excretions, blood or tissues.

PREVENTION OF NIV- To reduce the risk of infection when travelling to places affected by NiV, the public should adopt the following measures:

- Avoid contact with farm animals or wild animals, especially bats and pigs;
- Observe good personal hygiene; wash hands frequently with liquid soap and water, especially after contact with animals or their droppings/secretions, and taking care of or visiting sick people;

Infected people initially develop influenza-like symptoms of fever, headaches, myalgia (muscle pain), vomiting and sore throat. This can be followed by dizziness, drowsiness, altered consciousness, and neurological signs that indicate acute encephalitis. Some cases may develop atypical pneumonia and severe respiratory problems, including acute respiratory distress. Most people who survive acute encephalitis make a full recovery, but long term neurologic conditions have been reported in survivors. Approximately 20% of patients are left with residual neurological consequences such as seizure disorder and personality changes. The case fatality rate is estimated to range from 40% to 75%. There are currently no drugs or vaccines specific for NiV infection. The primary treatment for human cases is intensive supportive care.





### ▪ Control

While the general recommendation is to avoid drinking any unpasteurized juices in endemic regions, keeping bats away from sap collection sites with protective coverings (e.g., bamboo sap skirts) may be helpful in areas where people are unlikely to stop drinking raw date palm sap. Smearing lime on the collection area to discourage bats appeared to have little inhibitory effect in one study. Fruit should be washed thoroughly, peeled or cooked before eating. Good personal hygiene, including hand washing, is likely to reduce the risk of infection from the environment. Nipah virus has been classified as a Hazard Group 4/ BSL4

pathogen; infected animals, body fluids and tissue samples must be handled with appropriate biosecurity precautions.

People who come in close contact with potentially infected animals should wear protective clothing, impermeable gloves, masks, goggles and boots. Because Nipah virus can be transmitted from person to person, barrier nursing should be used when caring for infected patients. Patients should be isolated, and personal protective equipment such as protective clothing, gloves and masks should be used.

Good hygiene and sanitation are important; in one study, hand washing helped prevent disease transmission. Vaccines are currently not available for humans. Good biosecurity is important in preventing infections on pig farms; strategies should target routes of contact with other pigs as well as fruit bats. Fruit tree plantations should be removed from areas where pigs are kept. Wire screens can help prevent contact with bats when pigs are raised in open-sided pig sheds Nipah virus infection can be prevented by avoiding exposure to sick pigs and bats in endemic areas and not drinking raw date palm sap Early recognition of infected pigs can help protect other animals and humans. Due to the highly contagious nature of the virus in swine populations, mass culling of seropositive animals may be necessary.

### ▪ Disease reporting

Veterinarians who encounter or suspect a Nipah virus infection should follow their national and/or local guidelines for disease reporting. In the U.S., state or federal veterinary authorities should be informed immediately.

### ▪ Media monitoring

This is an important source of information for detection of unusual health events and outbreak detection, including Nipah virus. A core group of RRT evaluates media news and decides on unusual events. The RRT checks the unusual events on health and communicates with local health authorities at the place of the unusual event for confirmation and to get an update report.

### ▪ Intensification of surveillance during Nipah season

Surveillance is enhanced or intensified during Nipah season from January through March, when most Nipah outbreaks have been identified. This will increase the possibility of identification of Nipah virus infection and to understand the characteristics of the virus. Blood, cerebrospinal fluid and throat swabs are collected from suspected patients and sent for laboratory investigation.

#### ▪ National Rapid Response Team (NRRT)

The NRRT is stationed at IEDCR headed by Director. The team members consist of epidemiologist, clinicians, lab scientists, social scientists, communication experts. Animal science experts, toxicologists, clinicians, local health care providers.

#### ▪ District Rapid Response Team (DRRT)

The DRRT is headed by CS and consists of clinical and laboratory expertise, health educators, and other experts in public health.

#### ▪ Upazilla Rapid Response Team (URRT)

It is headed by UHFPO and consists of of clinical and laboratory expertise, health educators, and other experts in public health.

### CONCLUSION

In conclusion, knowledge and awareness on the disease should be improved and disseminated to health services, veterinarians, farmers and consumers. Nipah virus, as other zoonotic agents, might be included in monitoring plans, in particular for wild animals. Prioritization may drive the attention to other pathogens showing for example higher incidence in the population. However, field investigations may demonstrate radical and unexpected epidemiological changes. For example, the discovery of a novel ebolavirus-like filovirus in Spanish microbats demonstrated that the potential for such spill over events is not limited to Africa or Asia [68]. It is therefore important to enhance our preparedness to counter potential future introduction of exotic pathogens as henipa viruses in non endemic areas by conducting active pre-emergence research. Of utmost importance, monitoring the evolving epidemiology of a dangerous pathogen like the Nipah virus is an essential element to be able to promptly adapt control plans in the case that it might become a new public health priority. Finally, a better understanding of the molecular mechanisms of HNV pathogenesis will be important for developing effective counter measures to prevent and treat infection with these often-lethal viruses.

**Acknowledgement:** The authors gratefully acknowledge in charge Principal Dr. S. A. Tamboli of Appasaheb Birnale College of Pharmacy, Sangli for providing essential facilities and his help in manuscript preparation.



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Source of Support: Nil, Conflict of Interest: None.

