AUSTRALIAN VETERINARY EMERGENCY PLAN

AUSVETPLAN

Response policy brief

Hendra virus infection

Version 3.5, 2013

AUSVETPLAN is a series of technical response plans that describe the proposed Australian approach to an emergency animal disease incident. The documents provide guidance based on sound analysis, linking policy, strategies, implementation, coordination and emergency-management plans.

Standing Council on Primary Industries

This disease strategy forms part of:

AUSVETPLAN Edition 3

This document will be reviewed regularly. Suggestions and recommendations for amendments should be forwarded to:

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DISEASE WATCH HOTLINE

1800 675 888

The Disease Watch Hotline is a toll-free telephone number that connects callers to the relevant state or territory officer to report concerns about any potential emergency disease situation. Anyone suspecting an emergency disease outbreak should use this number to get immediate advice and assistance.

Hendra virus infection

Introduction

This response policy brief for the management of an outbreak of Hendra virus infection in Australia is an integral part of the **Australian Veterinary Emergency Plan**, or **AUSVETPLAN (Edition 3)**. AUSVETPLAN structures and functions are described in the AUSVETPLAN **Summary Document**.

This manual has been produced in accordance with the procedures described in the AUSVETPLAN **Summary Document** and in consultation with Australian national, state and territory governments, and the relevant livestock industries.

In Australia, Hendra virus infection is included as a Category 2 emergency animal disease in the *Government and Livestock Industry Cost Sharing Deed in Respect of Emergency Animal Disease Responses* (EAD Response Agreement).[1]

In this manual, text placed in square brackets [xxx] indicates that that aspect of the manual remains contentious or is under development; such text is not part of the official manual. The issues will be worked on by experts and relevant text included at a future date.

Guidelines for the field implementation of AUSVETPLAN are contained in the disease strategies, response policy briefs, operational manuals, management manuals and wild animal manual. Industry-specific information is given in the relevant enterprise manuals. The full list of AUSVETPLAN manuals that may need to be accessed in an emergency is shown below.

AUSVETPLAN manuals[2]

Disease strategies
- Individual strategies for most of the diseases listed in the EAD Response Agreement
- Bee diseases and pests
- Response policy briefs (for diseases not covered by individual manuals)

Operational manuals
- Decontamination
- Destruction of animals
- Disposal
- Livestock welfare and management
- Public relations
- Valuation and compensation

Enterprise manuals
- Artificial breeding centres
- Feedlots
- Meat processing
- Pig industry
- Poultry industry
- Saleyards and transport
- Zoos

Management manuals
- Control centres management (Parts 1 and 2)
- Laboratory preparedness

Wild animal response strategy

Summary document

The national public health response to Hendra virus is detailed in the *Hendra Virus: National Guidelines for Public Health Units*.[3]

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1 Information about the EAD Response Agreement can be found at www.animalhealthaustralia.com.au/programs/emergency-animal-disease-preparedness/ead-response-agreement

2 The complete series of AUSVETPLAN documents is available on the internet at: www.animalhealthaustralia.com.au/programs/emergency-animal-disease-preparedness/ausvetplan

Nature of the disease

Hendra virus (HeV) is a zoonotic pathogen that has caused natural infection and disease in horses and humans. Humans have been infected as 'spillover' events from infected horses. HeV antibodies were confirmed in a dog in a natural setting for the first time in July 2011. It is unknown how the dog became infected, and it did not show any detectable clinical signs of illness.

There is no specific treatment for infected animals and no specific treatment for HeV infection in humans. Although antiviral medications have been used to treat human HeV infections, their efficacy is unknown. Three people (out of seven) have survived infections with general medical support. Research is being undertaken into the use of specific monoclonal antibodies in intervening and preventing HeV infection from developing in people with high-level exposures.

Aetiology

HeV is in the *Henipavirus* genus of the *Paramyxoviridae* family. This genus contains Hendra and Nipah viruses. A new paramyxovirus, Cedar virus, has recently been detected from pteropid bats. Full genome sequencing of this virus suggests a close relationship with the henipaviruses (Marsh et al 2012).

Susceptible species

Pteropid bats (flying foxes) are the known natural reservoir host of HeV infection (seroprevalence varies between 20% and 50%). Sporadic 'spillover' of HeV from flying foxes to horses occurs; however, the factors associated with spillover events are not yet fully understood, and research is ongoing. Neutralising antibodies to the virus are found in flying fox populations in Australia and Papua New Guinea.

Horses are the only species known to have been infected naturally from flying foxes. In July 2011, antibodies to HeV were confirmed in a dog sampled on a property where HeV had been confirmed in horses. The dog potentially had multiple opportunities for contact with an infected horse, and this is the most plausible route of infection.

Humans have become infected only after close contact with respiratory and/or oral secretions, body fluids and/or blood from an infected horse. No human is known to have been infected through direct contact with flying foxes. To January 2013, seven people have become infected after close contact with HeV-infected horses. Four of these people subsequently died of their infection.

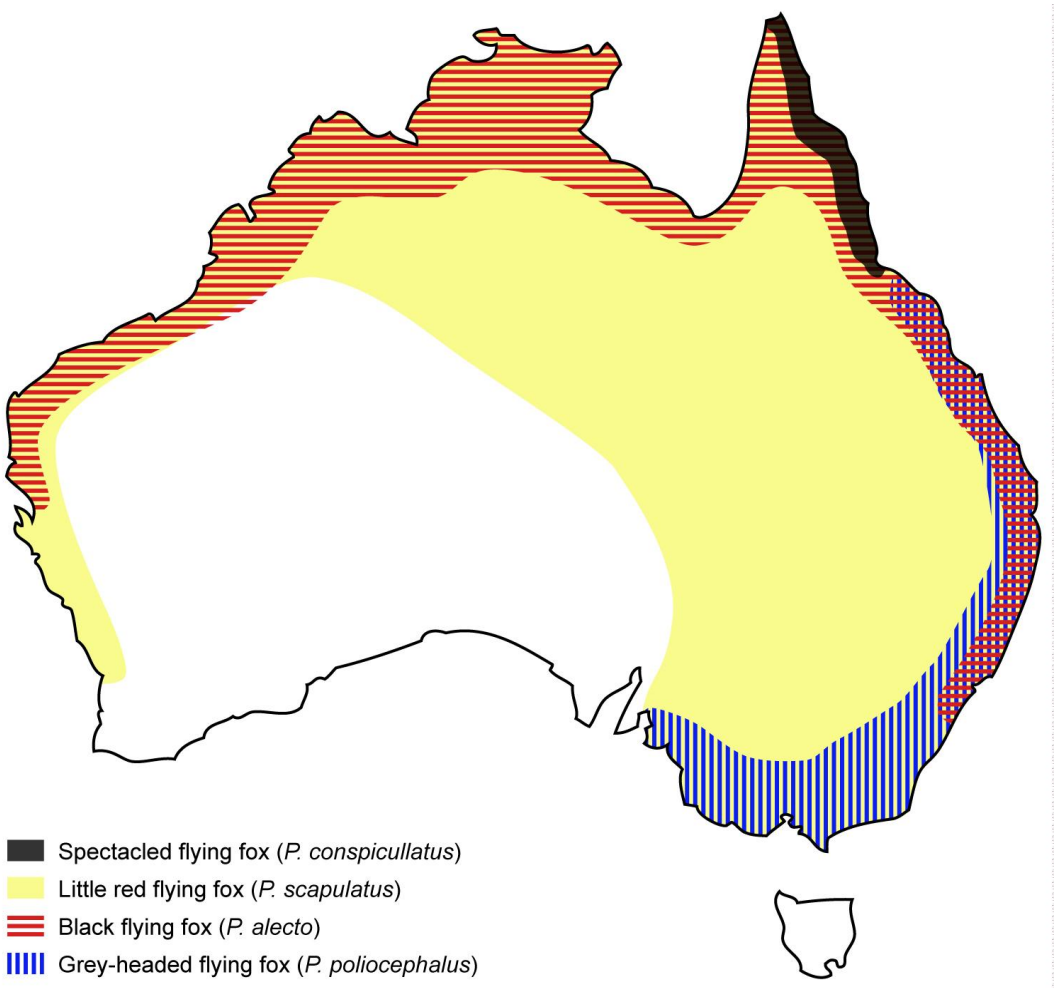
Experimental research on other animal species has found that cats, pigs, hamsters, ferrets, African green monkeys, guinea pigs and mice can be infected with HeV and develop clinical signs (Westbury et al 1995, Bossart et al 2011, Pallister et al 2011, Dups et al 2012). Rats, rabbits and a dog developed antibodies but not clinical signs when exposed to HeV (Westbury et al 1995).

Experimental studies in Canada (Li et al 2010) showed that the response of pigs to inoculation with high doses of HeV ranged from no clinical disease to severe interstitial pneumonia. Although this work has demonstrated that pigs can be infected experimentally, no natural infections have been reported in pigs.

[3] www.health.gov.au/internet/main/publishing.nsf/Content/cdnasongs.htm

Distribution

All confirmed HeV infections in horses have overlapped with the spatial distribution of pteropid bats (see Figure 1). Since the spatial distribution of pteropid bats may change from year to year and from season to season (Roberts et al 2012), Figure 1 is indicative only. Pteropid bats are highly mobile species, and there are occasional incursions into new areas during periods of severe food shortage; these have not been included in Figure 1.



Note: Two individual black flying foxes were sighted in Melbourne in 2010, associated with a severe food shortage (K Cox-Witton, Australian Wildlife Health Network, pers comm, 2013).

Source: Adapted from Richards and Hall (2012)

Figure 1 Spatial distribution of pteropid bats in Australia, 2012

Preliminary findings indicate that little red flying foxes and grey-headed flying foxes typically have a low HeV excretion prevalence, which is consistent with the occurrence of reported equine cases (H Field, Queensland Centre for Emerging Infectious Diseases, pers comm, 2013).

Updated statistics on HeV incidents, including locations, dates, and confirmed cases in humans, horses and a single seropositive dog, can be found on the website of the Queensland Department of Agriculture, Fisheries and Forestry.[4]

[4] www.daff.qld.gov.au/4790_13371.htm

Diagnostic criteria

Case definition

The case definition of an HeV-infected animal[5] is:

- an animal that has been sampled following a suspicious clinical history and tests positive to HeV using one or more of the following tests:
  - polymerase chain reaction (PCR)
  - virus isolation
  - immunohistochemistry
  - demonstration of an antibody response by virus neutralisation test (VNT), or soluble glycoprotein (sG) enzyme-linked immunosorbent assay (ELISA) confirmed by VNT or another confirmatory test, with an antibody profile consistent with infection, but not with vaccination[6]

or

- a nonclinical animal that has been sampled and tests positive to HeV using one or more of the following tests:
  - PCR on blood, with PCR positive results either:
    a. on samples collected on separate occasions and consistent with virus replication, or
    b. of sufficient magnitude to suggest that contamination of samples is improbable
  - virus isolation
  - immunohistochemistry
  - demonstration of an antibody response by VNT, or sG ELISA confirmed by VNT or another confirmatory test, with an antibody profile consistent with infection, but not with vaccination.

Where positive results are obtained from nonclinical animals sampled at anatomical sites that are susceptible to environmental contamination (eg nasal cavity, oral cavity, rectum), or from samples where contamination cannot reasonably be excluded, confirmation of infection by demonstration of an antibody response consistent with infection is necessary.

An animal for which testing has not been possible or for which testing is inconclusive, but there is compelling epidemiological evidence that the animal is/was infected (eg confirmed human infection following contact with an animal with clinical signs and history suggestive of HeV infection), would also meet the case definition.

Clinical signs

HeV infection of horses typically causes an acute illness that is rapidly fatal. There are no pathognomonic clinical signs. Horses infected with HeV have shown variable and often vague clinical signs, including respiratory and/or neurological signs, frequently accompanied by pyrexia.

In most of the recorded cases of infection, there has been strong presentation of neurological or respiratory clinical signs; however, occasional cases have had a much milder

[5] Terrestrial animals known to be susceptible to infection with HeV under experimental or natural conditions. Flying foxes are considered to be arboreal.

[6] The antibody profile refers to sequential antibody levels over time and/or outcomes from DIVA testing (differentiating infected from vaccinated animals).

presentation. From the disease pathogenesis perspective, it is reasonable to assume that virus-induced damage to vascular endothelium and the subsequent vasculitis play a major role in producing clinical signs, and that the clinical presentation relates to the organ system(s) sustaining severe and compromising endothelial damage.

Information from the confirmed cases to date suggests that approximately 20% of horses can survive acute infection.

In experimentally infected cats, guinea pigs, pigs, hamsters, African green monkeys and ferrets, infection was comparable to that seen in naturally and experimentally infected horses. Cats demonstrated inappetence and increased respiratory rate, followed by death within 1–2 days (Westbury et al 1995), whereas ferrets demonstrated severe respiratory and neurological disease, as well as generalised vasculitis (Pallister et al 2011).

No clinical signs were reported for the only known naturally infected dog or for two experimentally challenged dogs.

Laboratory tests

Samples required

A wide range of relevant samples will:

- increase the overall diagnostic sensitivity, particularly if viral genome is at or near the limits of detection
- provide more information about the state of infection and the potential for virus excretion and transmission to others from an individual HeV-infected animal
- increase the confidence in a negative HeV diagnosis (specificity), if a wider range of negative samples are obtained from the same animal.

Note that no one sample type can be considered ideal for agent detection. For this reason, multiple samples should always be submitted, where possible.

Preferred samples (in order of most to least preferred) for diagnosis of an acute case are as follows:

- Ethylenediaminetetraacetic acid (EDTA) blood. Since liquid EDTA blood samples contain both cells and plasma, PCR testing on EDTA samples may detect virus in cells when virus or viral genome is not present in serum. Virus isolation is also possible. Note that the tube should be properly filled to minimise the risk of a high anticoagulant concentration interfering with testing.
- Swabs. Nasal, oral or rectal swabs may be used for PCR testing and virus isolation, and may detect infection at an earlier stage of infection than blood or other clinical samples (eg body fluids and secretions). A urine-soaked swab taken from the ground immediately after urination may also be used for PCR and virus isolation.
- Serum (plain/clotted whole blood). As a single sample, serum allows both PCR and serology. Note, however, that confidence in a negative PCR result based on a single serum sample is limited, because serum can provide false negative PCR results.

Lithium–heparin (LiHep) blood samples are not preferred. These samples provide no detection possibilities that are not already available from clotted and EDTA samples. LiHep blood is more likely to be inhibitory to PCR, which may give false negative results.

Submitting a combination of EDTA blood, serum, and nasal or other swabs should be sufficient for detection of HeV infection.

Other recommended samples that could be taken if it is safe to do so include those listed in Table 1.

Table 1 Additional samples for testing for Hendra virus

Sample type	Sample	Live horse	Dead horse
Swab	Conjunctiva	✓	✓
	Orifice (rectal, vaginal, urethral, buccal)	✓	✓
	Cut surface of submandibular lymph node		✓
Tissue[a]	Whole or part of submandibular lymph node		✓
	Other fresh or fixed samples (eg central nervous system tissue, lungs, spleen, liver, kidney, heart, diverse lymph nodes)		✓

a Tissues should only be collected if appropriate workplace health and safety controls are in place. They should not be collected routinely.

Samples are to be packaged, transported and submitted to the laboratory in accordance with the regulations of the International Air Transport Association (IATA).[7] Details are available in the *Requirements for the Packaging and Transport of Pathology Specimens and Associated Materials*[8] produced by the National Pathology Accreditation Advisory Council, and relevant IATA references.

Laboratory diagnosis

HeV can be detected through laboratory testing by virus isolation, PCR or serology (sG ELISA[9] or VNT). All HeV exclusion testing should use a combination of PCR and serological assay, whenever suitable sample types and volumes are available.

Molecular biology

PCR tests detect sections of the HeV genome. A positive result only indicates the presence of viral genome in the sample; it does not indicate whether the virus is viable and infectious. PCR testing results are generally available within 24 hours after samples are received at the laboratory.

A positive PCR test accompanied by relevant clinical signs is interpreted to mean that an animal has an existing HeV infection.

A positive PCR test on blood samples collected on separate occasions and consistent with virus replication, or of sufficient magnitude to suggest that contamination of samples is improbable, without relevant clinical signs is interpreted to mean that the animal has been, or is, infected and may still have viable virus within its body (eg within the central nervous system).

A negative PCR test needs to be interpreted in relation to the health of the animal and the broader epidemiological context. The animal could be in the early stages of infection with

7 www.iata.org

8 www.health.gov.au/internet/main/publishing.nsf/Content/health-npaac-docs-PackTransPathSpecimens.htm

9 The sG ELISA is the new standard and replaces the sds ELISA. All references to ELISA testing in this document refer to sG ELISA, unless otherwise indicated.

HeV (resulting in a low concentration of virus, or absence of virus at sampled sites) or may have recovered from infection.

Serology

Serological tests, including sG ELISA and VNT, are conducted on serum samples and detect the presence of antibodies to HeV. The ELISA is a screening test, whereas the VNT is a confirmatory test. The ELISA has only been validated in horses, whereas VNTs have been validated for use in dogs and cats.

A negative ELISA result is a reliable indicator that a horse has not been previously infected with HeV or vaccinated. However, antibodies take time to be produced in response to infection or vaccination, which should be considered when interpreting negative results. Since it may take an animal 5–10 days to produce detectable serum antibodies after infection, a single serological test might not detect antibodies if the sampled animal has had insufficient time to develop detectable antibodies.

Non-negative, including nonspecific reactor, results on sds ELISA testing require further testing to clarify the result. Use of the VNT or resampling and use of the sG ELISA are recommended.

A positive ELISA result should be returned for infected and vaccinated horses, because vaccine will induce the production of neutralising anti–G protein antibodies. Differentiating infected from vaccinated animals (DIVA) requires specific testing. Since the ELISA is a screening test, non-negative results to ELISA testing must be followed by a positive VNT before the animal could be considered seropositive to HeV.

A positive VNT result indicates that the animal is seropositive to HeV, from either infection or vaccination. A negative VNT result means that the animal is not seropositive to HeV. Antibodies take time to be produced in response to infection or vaccination, and this should be considered when interpreting negative results.

In unvaccinated animals, the VNT is the 'gold standard' and most specific test for anti-HeV antibodies. Its use is limited by the need to use live, infectious HeV in the test. This restricts conduct of the VNT for animal submissions to the CSIRO Australian Animal Health Laboratory (CSIRO-AAHL).

The VNT may take 7–10 days to complete, excluding the time it takes for the sample to reach a laboratory.

HeV serological testing of vaccinated horses

Standard HeV serological tests (those used for HeV diagnosis or exclusion before use of the HeV vaccine) are based on either whole live virus or viral sG. Both vaccination with the sG recombinant subunit and natural infection induce anti-G antibodies.

Development and assessment of tests to differentiate infected from vaccinated animals (DIVA testing) is an area of active investigation; a project is being funded by the National Hendra Virus Research Program (completion is expected in 2015–16). CSIRO-AAHL has available an experimental assay that can provide DIVA results. The current lack of validation of the DIVA tests, and limited data on test sensitivity and specificity must be considered in the interpretation of results.

The sG protein is the only HeV antigen used in the HeV vaccine. The absence of other HeV proteins in the vaccine means that detection of antibodies to any (one or more) of the other HeV proteins is specific to infected animals.

Table 2 outlines interpretation of the HeV serology profile using DIVA tests.

Table 2 Interpretation of Hendra virus serology profiles, including DIVA tests

sG ELISA and/or VNT	Non–G protein DIVA assays	Profile interpretation
Negative	Negative	Seronegative — no response to either vaccination or infection
Positive	Negative	Seropositive, with a response consistent with vaccination but not infection
Positive	Positive	Seropositive, with a response consistent with infection. Does not exclude a concurrent vaccination response
Negative	Positive	Should not occur — inconsistent results, suggesting that resampling and retesting should be considered

DIVA = differentiating infected from vaccinated animals; ELISA = enzyme-linked immunosorbent assay; sG = soluble glycoprotein; VNT = virus neutralisation test

Virus isolation

Hendra virus isolation can be undertaken at CSIRO-AAHL and is frequently requested on all index cases. Virus isolation confirms the presence of the virus; however, negative results do not indicate that the animal is, or was, not infected with HeV because the virus can be difficult to isolate. Clinical findings and other laboratory tests that are more sensitive for the detection of genetic material (PCR) and an immunologic response (serology) must be considered in interpreting negative virus isolation results.

Laboratory diagnostic capacity

Table 3 shows testing capacity available at different laboratories. The VNT and virus isolation can only be undertaken at CSIRO-AAHL, which is the World Organisation for Animal Health (OIE) and national reference laboratory for HeV disease.

Table 3 Laboratory testing capacity for Hendra virus

Laboratory	ELISA	VNT	DIVA	PCR	Virus isolation
CSIRO-AAHL	X	X	X (experimental assay)	X	X
Biosecurity Science Laboratory, Qld	X			X	
Elizabeth Macarthur Agricultural Institute, NSW	X			X	
Berrimah Veterinary Laboratory, NT				X	
Animal Health Laboratory, WA	X			X	
AgriBioscience Laboratory, Vic	X				

CSIRO-AAHL = CSIRO Australian Animal Health Laboratory; DIVA = differentiating infected from vaccinated animals; ELISA = enzyme-linked immunosorbent assay; PCR = polymerase chain reaction; VNT = virus neutralisation test

Epidemiology

Incubation period

As of January 2013, 80% of known equine cases have had an incubation period of 12 days or less, and 95% have had an incubation period of 15 days or less. No known HeV-positive equine cases have had an incubation period greater than 16 days, and three studies (Murray et al 1995, Baldock et al 1996, Field et al 2010) reported a minimum 4-day incubation period for both experimental and natural infections. One paper reported an incubation period of 3–10 days (Baldock et al 1995). For the purpose of determining a minimum quarantine period, 20 days has been determined to be appropriate, based on the maximum known incubation period of 16 days plus an additional 4 days as a precautionary measure. This quarantine period may be extended, as determined appropriate by the jurisdiction, to include additional time to receive final negative laboratory test results.

Information on the incubation period for cats and dogs is limited. One experimental study showed a 12-day incubation period in cats, and this has, in the past, also been applied to exposed dogs (Westbury et al 1996). However, because of the limited number of cases in dogs and cats, an incubation period of 16 days should be applied for both. Similarly to horses, a safety margin of 4 additional days is used for the purposes of tracing, surveillance (ie daily observations) and quarantine periods.

Persistence of agent

HeV is a lipid-enveloped virus and is susceptible outside the host to desiccation and changes in temperature (Fogarty et al 2008). Experimental evidence indicates that HeV survival in the environment varies from several hours to several days, depending on environmental conditions (Fogarty et al 2008), and spread by fomites should be considered during any incident. Fogarty et al reported the following:

- HeV in flying fox urine (pH ~7) survived for more than 4 days at 22 °C, with a half-life of 19 hours; at 37 °C, it was mostly inactivated in less than 1 day, with a half-life of 3 hours.
- HeV survival on mango fruit flesh at 22 °C decreases with increasing acidity. At pH 5, half-life is 22 hours, whereas at pH 3 it is 0.3 hours.
- HeV survival on fruit pulp and in fruit juice varies depending on the type and pH of the fruit. Viruses incubated in lychee juice showed greater persistence than in either

pawpaw or mango juice, with 2–3-fold longer half-lives and survival for more than 3 days.

- HeV is rapidly inactivated by desiccation at both 22 °C and 37 °C. Virus survival after desiccation in the laboratory is reported as less than 15 minutes at 37 °C. At 22 °C, HeV levels decreased by more than 3 logs within 30 minutes (half-life of 1.2 minutes).

For the purposes of disease control, 5 days is presumed to be the maximum survival time for HeV under optimal environmental conditions — that is, neutral pH, moist air and moderate temperatures. This survival time is doubled to 10 days as a precautionary measure.

Since a small number of people who recovered from initial infection with HeV or Nipah virus subsequently relapsed or developed delayed-onset encephalitis (WHO 2009ab, Taylor et al 2012), it is plausible that recovered, confirmed-infected terrestrial animals may experience recrudescence or relapsing disease. To date, there are no published data on relapsing HeV encephalitis in animals.

There is no evidence that animals or humans experiencing recrudescent disease following infection with other paramyxoviruses are infectious to in-contact individuals.

There are also no published studies showing any evidence of persistence of shedding of infectious virus in humans who have recovered from acute infection and later relapsed, or suffered late-onset encephalitis from either HeV (one case) or Nipah virus infections (O'Sullivan et al 1997, Goh et al 2000, Tan et al 2002, Wong et al 2009, Abdullah et al 2012, Wong and Tan 2012). Taylor et al (2012) reported that virus was not detected in blood samples from two humans infected with HeV 2 and 6 years after acute infection, and neither patient showed evidence of virus shedding after the acute phase of influenza-like illness or encephalitis.

Similar studies have not been undertaken in horses, because current practice is to euthanase recovered horses. It is unknown whether horses pose a risk of further HeV transmission to other susceptible terrestrial animals, including humans, during the period after recovery from the acute phase.

Modes of transmission

Although the exact route of transmission is not known, it is thought that horses become infected with HeV by ingesting material contaminated with infected flying fox body fluids and/or excretions. It is also plausible that horses may become infected directly via droplet inhalation via the nasal route (Mori et al 1995, Rudd et al 2006, Dups et al 2012, Munster et al 2012). Once a horse becomes infected, there is the potential for HeV to be transmitted to other horses, humans and other susceptible species.[10]

Current field and experimental knowledge indicates that HeV is most likely transmitted from horses to other horses or humans by contact with infected body fluids or tissues, or through droplet transmission. Undertaking certain procedures on horses (eg endotracheal intubation, nasal lavage, necropsy) may increase the risk of infection for attending personnel by promoting droplet or aerosol generation. It is not definitively known how the sole natural infection in a dog occurred; close contact with infected horses is suspected.

No definitive studies have been found describing the distance over which respiratory droplets can spread from horses. A maximum distance of 5 metres is assumed, based on the

[10] Susceptible species are those terrestrial domestic animals shown to be experimentally or naturally susceptible to HeV.

absence of transmission to horses beyond this distance in field scenarios, qualitative extrapolation of droplet studies in humans, and observations of exhaling horses after exercise. This 5-metre distance can be extended if circumstances indicate that additional precautions are appropriate (El Saadi et al 2011).

Current evidence suggests that, although there is some risk of transmission in the preclinical phase, transmission risk increases with disease progression, and is highest when the horse is near death and immediately postmortem.

Viral genetic material was identified by PCR in three experimentally infected horses 3–5 days before the appearance of clinical signs (Marsh et al 2011). Further, the same authors reported that most tissues and organs contained HeV genome at the time of death (euthanasia). The report indicates that nasal secretions of asymptomatic horses may pose a transmission risk during the early phase of disease that precedes viraemia, fever or other discernible clinical signs of HeV infection. In light of this finding, people coming into contact with sick horses should give early consideration to HeV in the differential diagnosis and apply infection control procedures relating to blood, and oral and nasopharyngeal secretions. Personal protective equipment (PPE) and other safety procedures are currently the primary defence in the preclinical phase when horses may excrete HeV.[11]

Laboratory analysis has shown that an infected horse showing clinical signs will have virus in blood and tissues, and may be excreting HeV in body fluids and excretions, including urine, oral and nasopharyngeal secretions, and faeces.

Disease prevention

An HeV recombinant subunit vaccine for horses was commercially released on 1 November 2012, following evaluation and the approval of a minor use permit (PER13510)[12] by the Australian Pesticides and Veterinary Medicines Authority. Its use must be in accordance with the permit conditions.

Preclinical trials demonstrated that subunit sG vaccine constructs could prevent henipavirus clinical disease, virus transmission or virus shedding in laboratory animal models (Mungall et al 2006, McEachern et al 2008, Pallister et al 2011, Broder 2012).

Research by CSIRO-AAHL demonstrated that the adjuvanted vaccine protected horses from direct oronasal challenge with a lethal dose of HeV for up to 6 months following the initial course of two vaccinations, administered 21 days apart. Vaccinated horses demonstrated no clinical signs of disease and did not shed any detectable infectious virus (D Middleton, CSIRO-AAHL, pers comm, 2013).

Vaccination can break the cycle of virus transmission from horses to humans, and provide a public health and workplace health and safety benefit. As well as adopting good personal hygiene and biosecurity practices, and using PPE appropriate to the situation when dealing with horses, workplace health and safety planning should consider the public health benefit of horse vaccination. Widespread uptake of the horse vaccine has the potential to significantly reduce the risk of human exposure.

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11 Refer to the *Guidelines for Veterinarians Handling Potential Hendra Virus Infection in Horses* produced by the Queensland Department of Agriculture, Fisheries and Forestry for details of appropriate PPE (www.daff.qld.gov.au/4790_13371.htm).

12 http://permits.apvma.gov.au/PER13510.PDF

The permit and product leaflet specifies that only registered veterinary surgeons can use the vaccine in horses, as an aid in the prevention of clinical disease caused by HeV. Studies on the use of HeV vaccine in breeding animals are currently incomplete; therefore, use in these animals must be weighed against the risk of serious illness or death from disease caused by HeV infection. There are no data to support the use of HeV vaccine in sick horses.

Disease control and eradication

HeV is a zoonotic disease agent for which stringent biosecurity and safety measures (relating to both public health and workplace health and safety) are necessary. Careful risk management of the situation, safe work practices, infection control practices and PPE are required to manage potential human exposure. The website of the Queensland Department of Agriculture, Fisheries and Forestry[13] provides comprehensive guidelines for disease management by private veterinarians.

Within a standard risk management matrix, the consequences of HeV infection are classified as being potentially severe for humans and some susceptible animal species (other than pteropid bats). As a result, a conservative precautionary approach should be taken whenever uncertainty exists — that is, procedures should be put in place to limit possible harm in all cases where HeV infection is considered as a differential diagnosis.

After due investigation and suspicion by the private veterinarian, cases that include HeV infection as a differential diagnosis must be notified to the relevant jurisdictional animal health authority, even though it may not be certain from the outset that HeV infection is involved. As a precautionary measure, the private veterinarian should also implement strict, recommended biosecurity and infection control procedures until HeV infection can be excluded.

Once HeV infection is confirmed, the relevant jurisdictional animal health authority will work with the private veterinary practitioner(s) to manage the disease. Advice will also be provided to public health authorities. The initial response to detection of the disease will include undertaking an epidemiological assessment of the susceptible animals and premises. Management most commonly includes preventing spread of the disease between properties by quarantining infected and susceptible animals; segregating susceptible terrestrial animals from one another within the affected property, where possible, to mitigate the risk of horizontal transmission; considering vaccination of close contact at-risk horses; preventing susceptible terrestrial species (horses) on the property from sheltering under trees attractive to flying foxes; and instituting effective biosecurity and infection control procedures, to minimise the risk of human infection. Movement controls have been effective in containing HeV to an infected premises, and limiting spread between horses and other susceptible species.

Sampling and testing of close contact and suspect susceptible animals (defined below, under 'Epidemiological assessment') is undertaken at prescribed times to determine the disease status of individual animals. After sufficient time has elapsed without detection of disease, quarantine and movement controls are rescinded. All animal owners are encouraged to continue to practise appropriate biosecurity precautions.

The HeV response should focus on management of terrestrial animals. Culling or dispersal of flying fox colonies is not an effective strategy for controlling HeV, as flying foxes are highly mobile, and individuals move between colonies.

[13] www.daff.qld.gov.au

Research on physiological stresses that may be associated with dispersal of flying fox colonies and the impact (if any) on the risk of HeV transmission has recently been completed by the Queensland Centre for Emerging Infectious Diseases (D Edson, L McMichael and H Field, QCEID, pers comm, 2013). This research was commissioned by the National Hendra Virus Research Program, and supported by the Queensland, New South Wales and Australian governments. The key findings are as follows:

- There was no evidence of an association between dispersal or disturbance of flying foxes and HeV excretion.
- There was some evidence that dispersal or disturbance can cause stress that is similar in magnitude and duration to 'natural' stress.
- There was no evidence that dispersal or disturbance-associated stress results in HeV excretion.
- There was no evidence that dispersal or disturbance changes the frequency or scale of flying fox movements between colonies.
- There was evidence that little red flying foxes (*Pteropus scapulatus*) and grey-headed flying foxes (*P. poliocephalus*) have a low HeV excretion prevalence.

Epidemiological assessment

As part of the epidemiological assessment, animals, animal products and fomites are risk assessed for close contact with the infected animal. The following definitions are used:

- A terrestrial animal known to be susceptible to HeV (eg horse, dog, cat) with close contact is one that:
  - has come within 5 metres of a clinical HeV-positive horse, or a suspect horse, or
  - has potentially had direct contact with presumed-contaminated body fluids or substances from a clinical HeV-positive animal, a suspect animal or a recently deceased HeV-positive animal in the 10 days following excretion or secretion of the body fluids or substances, or
  - has potentially had contact with blood or nasopharyngeal secretions shed by preclinical animals in the 10 days before the onset of clinical signs — for example, through dental procedures, stomach tubing or very close nose-to-nose contact, or
  - has had direct contact with a contaminated fomite.
- A contaminated fomite is one that has had direct contact with body fluids (including nasopharyngeal secretions, urine, blood) or faeces from an HeV-positive animal or a suspect susceptible animal (this may be by contacting the infected animal or carcass) in the 10 days following secretion or excretion. This includes contact with blood or nasopharyngeal secretions from an HeV-positive animal that were secreted or excreted in the 10 days before the onset of clinical signs (ie preclinical).
- A suspect horse is any equid on an infected or dangerous contact premises showing any sign of illness (including increased temperature, respiratory signs, colic, neurological signs) consistent with HeV infection.
- A suspect susceptible animal is any susceptible terrestrial domestic animal that is known to be experimentally or naturally susceptible to HeV, other than a horse, on an infected or dangerous contact premises, showing any sign of illness consistent with the current knowledge of HeV infection. Current knowledge of clinical signs of HeV infection in animals other than horses is mainly extrapolated from knowledge of HeV infection in horses and limited experimental work.

Quarantine and movement controls

Management of different classes of animals reflects the risk the animals pose for disease spread. Each class of animal should be segregated to mitigate the risk of horizontal transmission and to allow different management practices for each group. Management procedures include the following:

- HeV-infected animals, as per the case definition, must be euthanased as soon as possible to limit the opportunity for animal-to-animal and animal-to-human transmission. Clinical observations before euthanasia may improve the knowledge of the behaviour of infected animals. Because of the potential risks to the observer (especially with stabled horses), a risk assessment must be undertaken to determine appropriate infection control procedures before personnel make close contact with the animal.
- Horses and other susceptible terrestrial animals having close contact with a suspect or HeV-positive horse need not be euthanased. However, they must be observed for development of clinical signs consistent with HeV infection for 20 days after the date of the last possibility of the animal having infectious contact, and tested for the virus at prescribed minimum intervals (see 'Sampling', below). This is based on the maximum known incubation period of 16 days, plus an additional 4 days as a precautionary measure.
- The remaining susceptible terrestrial animals on an infected, dangerous contact or trace premises that are not HeV-positive, close contact or suspect animals are of low interest. They need not be sampled or tested, but they must be observed as for any other animal under the owner's control and kept segregated from HeV-infected or close contact animals.

Human access to contaminated areas and animals that have been assessed as having had close contact with HeV-infected animals should be avoided as far as possible. Appropriate infection control procedures, including the use of PPE, should be used if access to contaminated areas and close contact animals is necessary.[14]

Appropriate workplace health and safety precautions (eg use of appropriate animal restraint and PPE) must be in place before animals are examined or samples are taken. Human health and safety must be considered when fluid and tissue samples are taken from HeV-positive, close contact or suspect animals. Where necessary, to mitigate the human health risk, CSIRO-AAHL may be commissioned to assist with high-risk procedures, such as necropsy of HeV-positive animals.

Tracing

The incubation period (and safety margin), virus survivability and preclinical shedding are important factors to consider when conducting tracing. Tracing should be undertaken in the following priority order:

1. Identification of susceptible animals in close contact (see above) with the infected horse.
2. Tracing of movements of horses, dogs and cats to and from the infected premises for 16 days before the first observation of unusual morbidity or mortality, to identify any animal that may have been infected at or around the same time as the infected animal. (Note: dogs have not shown clinical signs under experimental or natural conditions.)

[14] Refer to the *Guidelines for Veterinarians Handling Potential Hendra Virus Infection in Horses* produced by the Queensland Department of Agriculture, Fisheries and Forestry for details of appropriate PPE (www.daff.qld.gov.au/4790_13371.htm).

3. Tracing of movements of close contact fomites (see above). Tracing for the 10 days before unusual morbidity or mortality was observed on the infected premises will primarily relate to items contaminated by moderately invasive procedures involving the nasopharyngeal area of horses (eg stomach tubing, endoscopy or dental work).
4. Tracing of movements of people involved with the infected premises (eg veterinarians, farriers, feed delivery drivers, tradespeople, company service personnel) who potentially had contact with oronasal secretions or blood in the 10 days before the animal showed clinical signs and in the period until the animal died or was euthanased. The national public health response is detailed in the *Hendra Virus: National Guidelines for Public Health Units*.[15] Jurisdictions may also have similar documents.[16]

Sampling

Susceptible animals that have had close contact with a known infected animal are sampled three times, specifically:

- as soon as possible after contact, to establish a baseline and identify other animals that may have been infected at the same time as the index case
- 12 days from the date of the last possibility of the animal having infectious contact (eg with a clinical case or potentially infectious fluids), based on the premise that 80% of known infected horses have demonstrated positive PCR results at this time; given the small sample size of naturally infected dogs and cats, the same time frame may be applied to these species
- 20 days from the date of the last possibility of the animal having infectious contact, to identify any remaining susceptible animals that may have been infected by HeV-positive animal(s).

Samples required are listed above (under 'Diagnostic criteria').

Release from quarantine and movement controls

Release from quarantine and movement controls can occur:

- 10 days after the last potential contamination of the premises by fluids or wastes from an infected animal, where no infected, suspect or close contact animals remain on the premises (based on maximum virus survival time in the environment), or
- after negative test results are obtained 20 days from the date of confirmation of disease in a single animal case or from the date of last infectious contact in a multi-animal case, and the disease control authority managing the incident considers that any residual risk of HeV on the premises can be appropriately managed through biosecurity controls.

Destruction

The national policy for dealing with terrestrial animals[17] that are confirmed infected is humane destruction. 'Confirmed infected' means that the animal meets the case definition. Recovered seropositive horses may pose an infection risk to humans and other animals, since it is unclear whether horses that have recovered from HeV infection remain infectious or are capable of permanently clearing the virus. Scientific studies are in progress to help clarify this issue. Evidence of relapsing encephalitis has been observed with both HeV and Nipah virus infection in humans.

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15 www.health.gov.au/internet/main/publishing.nsf/Content/cdnasongs.htm

16 For example, www.health.qld.gov.au/cdcg/index/hendra.asp

17 Terrestrial animals (does not include bats) known to be susceptible to infection with HeV under experimental or natural conditions

Given the limited knowledge about HeV infection in dogs, the risk that seropositive dogs pose to humans and other susceptible animals is unclear.

The national policy on destruction should be reviewed if credible scientific evidence becomes available.

Collection of tissues (by people such as CSIRO-AAHL pathologists with appropriate technical expertise and the use of PPE) from recovered seropositive but PCR-negative animals may significantly increase current scientific understanding. Euthanasia of these animals may be delayed, following a risk assessment, to facilitate this sampling. However, such a delay should not be at the expense of animal welfare.

Disposal

Animals should be disposed of by an appropriate means, as described in the AUSVETPLAN **Disposal Manual**. Consideration must be given to on-site or off-site disposal and the advantages and disadvantages associated with each. In most circumstances, disposal on-site by deep burial or composting is the preferred option. Disposal must be in accordance with jurisdictional, local government and environmental protection legislation and guidelines.

Decontamination

Wherever possible, it is preferable that environmental decontamination be allowed to occur naturally through time and environmental processes. Under natural conditions and after application of a conservative precautionary approach, a contaminated area(s) and fomites will be considered decontaminated 10 days after the last known exposure to HeV (based on a doubling of the maximum survival time of 5 days).

If an area, such as a laboratory postmortem room, or object requires decontamination, the AUSVETPLAN **Decontamination Manual** should be consulted; decontaminants that are active against Category A viruses are appropriate for HeV.

Australia's policy for Hendra virus infection

Hendra virus (HeV) infection is a notifiable terrestrial animal disease in all states and territories of Australia. The detection of HeV infection in animals in Australia results in minor animal and public health impacts, and moderate adverse social effects.

HeV infection is not an OIE-listed disease.

The exact route of transmission of HeV is not known; however, it is known that close contact with flying fox fluids, or infected horses and/or their fluids is required for transmission.

Current scientific information is insufficient to determine whether recrudescence occurs in animals. However, relapsing encephalitis has been observed in both HeV and Nipah virus infection in humans. It is unknown whether recovered animals pose a risk of further HeV transmission to other susceptible animals, including humans.

HeV is currently included as a Category 2 disease in the *Government and Livestock Industry Cost Sharing Deed in Respect of Emergency Animal Disease Responses*. The costs of disease control can be shared 80% by governments and 20% by the relevant industries.

The policy is to eradicate HeV infection in terrestrial animals using:

- ***humane destruction* and *sanitary disposal* of all terrestrial animals that are confirmed infected through laboratory testing and positive findings of HeV antigen, genetic material or an antibody profile specific to infection consistent with the case definition, or compelling epidemiological evidence**
- ***decontamination* of the contaminated environment by natural means (preferred) or application of decontaminants; disposal or decontamination of contaminated fomites**
- ***quarantine, monitoring* and testing of close contact susceptible animals until the risk of HeV spread from the infected animal(s) has been eliminated.**

These strategies will be supported by:

- **stringent *biosecurity* and *workplace health and safety* measures to prevent humans and other susceptible animals from becoming infected**
- ***an epidemiological assessment*, including a risk assessment of susceptible animals and premises, to provide information for management of the situation**
- ***tracing* and *surveillance* to determine the source and extent of infection**
- **communications and a *community engagement campaign* to inform, and address the concerns and needs of, industries and the community.**

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