



Guidelines for veterinarians handling potential Hendra virus infection in horses

Version 5.1

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Summary of changes from previous version of this document (Version 5.0 September 2013):

- The line “Advise neighbours that a horse is being investigated for HeV infection” on page 20 has been removed and added as advice for the horse owner/manager to undertake.

This document is subject to regular review. Comments on its content and format are encouraged.

The most recent edition of this document is located on the Biosecurity Queensland website www.biosecurity.qld.gov.au

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1. Purpose

These guidelines are specifically intended to assist veterinarians in the safe investigation of illness in horses where Hendra virus (HeV) is considered as a possible cause.

Whenever this is the case, a professional risk management approach is required. The investigating veterinarian must manage the level of risk assessed for the situation—the serious zoonotic nature of HeV requires taking stringent infection control precautions.

These guidelines recognise that investigation and management of possible HeV cases will continue to be a joint approach between horse owners, private veterinary practitioners and Biosecurity Queensland. It is expected that private veterinary practitioners will investigate and where appropriate sample ill horses. As HeV is a notifiable disease, private veterinary practitioners must notify Biosecurity Queensland about possible HeV cases in Queensland.

There may be circumstances where a private veterinary practitioner may not be in a position to manage the level of associated risk. In these cases, the horse owner, the private veterinary practitioner and Biosecurity Queensland will need to consult.

These guidelines do not address routine work practices and any changes to those practices that the veterinary profession may introduce as a result of HeV incidents and new information.

Biosecurity Queensland encourages private veterinary practitioners to use these guidelines as appropriate in supporting the development of infection control procedures for routine veterinary practice.

All veterinary investigations of horses should be conducted using the *Guidelines for veterinary personal biosecurity* developed by the Australian Veterinary Association (AVA) as necessary. The *Guidelines for veterinary personal biosecurity* provides a practical and comprehensive understanding of zoonotic diseases and information on how to significantly reduce the risk of zoonotic infection and are available on the AVA website at www.ava.com.au

The *Guidelines for veterinarians handling potential Hendra virus infection in horses* will be updated as more knowledge and information becomes available. The current version of these guidelines is available at www.biosecurity.qld.gov.au

2. Scope

This document is intended for use by veterinarians to help them implement appropriate infection control procedures to manage the risk from HeV when investigating illness in horses. It provides information that relates to:

- identification of possible cases
- safe work practices to prevent human infection, including the use of personal protective equipment (PPE) and development of biosecurity procedures
- responsibilities of people involved with the case.

Investigation and management of possible HeV cases is a joint responsibility between horse owners, private veterinary practitioners and Biosecurity Queensland.

The primary role for Biosecurity Queensland is to:

- accept samples and conduct laboratory testing
- manage positive cases in animals
- provide advice and information about HeV.

On confirmation of a positive HeV case in an animal, Biosecurity Queensland will manage the case.

For the purpose of this document, we have assumed that:

- All suspected HeV cases will be notified to Biosecurity Queensland as required under legislation. Notification to a government veterinary officer can be made by contacting 13 25 23 (business hours) or 1800 675 888 (24 hours), or through direct contact with a government veterinary officer.
- Potential cases will be investigated by private veterinary practitioners as part of normal practice using these guidelines and their own procedures as a reference.
- Private veterinary practitioners should make the final decision about whether to collect samples and submit them for laboratory testing with reference to these guidelines and, where appropriate, obtain advice from Biosecurity Queensland.
- Private veterinary practitioners will apply appropriate infection control procedures to protect themselves and others against HeV risk from clinically ill horses or those that may be in the pre-clinical phase and excreting HeV.

3. Reference information

3.1 General

HeV was first isolated during 1994 when it occurred in a stable in the suburb of Hendra, Brisbane. Early names included acute equine respiratory syndrome and equine morbillivirus. However, following characterisation of the virus, it is now termed Hendra virus. HeV and Nipah virus are in the genus Henipavirus in the family Paramyxoviridae.

Flying foxes are a host reservoir of HeV¹. 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.

HeV has the potential to be a serious zoonotic disease for which stringent biosecurity and safety measures are necessary. There are important public health and workplace health and safety issues that require consideration. Careful risk management of the situation, safe work practices and PPE are required to manage potential exposure.

While much is now known about HeV, the scientific information available for HeV is not complete and there may be insufficient scientific knowledge to answer some questions that are posed. The consequences of HeV infection for both horses and humans can be potentially catastrophic. As a result, a conservative ‘precautionary principle’ approach should be taken whenever uncertainty exists—that is, procedures should be put in place to limit possible harm in all cases where HeV is considered as a differential diagnosis.

Given the potential consequences of HeV, it is vital that possible cases be notified and investigated. As is always the case when dealing with a disease situation, it may not be clear from the outset that HeV is involved. However, if HeV is included as a differential diagnosis then, as a precautionary measure, the veterinarian should implement the safety precautions discussed in these guidelines until such time as HeV infection can be excluded.

Veterinarians who treat horses should:

- Review their existing work practices so that individual assessment of horses for zoonotic diseases and prevention of potential transmission of these diseases to humans and other animals is built in to normal procedures.
- Develop plans for responding to a potential HeV case, including minimising risk to themselves, their workers and others (including clients).
- Review their infection control procedures as these are currently the primary defence for managing both horses in the pre-clinical phase of HeV infection (where horses can excrete virus before showing overt clinical signs) and clinically ill horses. This should incorporate procedures such as hand hygiene, PPE, avoiding contamination of premises and equipment, routine cleaning and disinfection of equipment (e.g. stomach tubes, endoscopes, dental equipment etc) between horses.
- Apply their professional judgement in the interpretation of laboratory results and seek advice as appropriate.

3.2 Epidemiology

Epidemiological information about HeV is incomplete and remains the subject of ongoing research. This section will be updated as new information becomes available.

Incidents

Until 2011, there had been 14 incidents of HeV over 17 years from 1994 to 2010. In 2011, there were 10 HeV incidents in Queensland and eight in New South Wales.

A case of HeV was confirmed in a horse at Chinchilla, Queensland, in 2011, which was the first detection west of the Great Dividing Range. Another case was confirmed in a horse at Macksville, New South Wales, which is the southern most case detected to date. Prior to 2011, the known detections had occurred on or east of the Great Dividing Range from Cairns to northern New South Wales.

There were eight incidents in 2012 and HeV infection in horses continues to be identified in 2013. A full list of confirmed HeV cases since its discovery in 1994 are outlined in detail on the Biosecurity Queensland website, www.biosecurity.qld.gov.au

Flying foxes

Flying foxes are a natural reservoir of HeV and the closely related Nipah virus. HeV is present in flying fox populations in Australia and Papua New Guinea. Nipah virus disease is not known to be present in Australia but is present in South East Asia and Indonesia.

Serological surveillance of Australian flying foxes has found antibodies to HeV in all four mainland Pteropus species—the black, grey headed, little red and spectacled flying foxes^{1,2,3,4}. There is no evidence that other animals may act as a natural reservoir for HeV^{5,6}.

Flying foxes are mobile animals and HeV should be considered wherever horses and flying foxes are in proximity to each other. Horses are also transported long distances and could be moved within the incubation period from an area where they may have been in contact with flying foxes or a horse infected with or incubating HeV, to an area where flying foxes do not exist.

A three year longitudinal study in flying foxes found that HeV is not present continuously in flying fox colonies, and that the level of excretion in any particular colony fluctuates over time⁷. The study indicated that flying foxes can be infected and excrete virus at any time of the year and that spill-over of virus to horses requires factors other than just the presence of virus. There is currently no reliable method to predict the high risk periods where flying foxes may be excreting HeV.

The majority of incidents coincide with the period from mid/late pregnancy to early birthing of three of the four Australian flying fox species. This correlation does not necessarily indicate a causal association, but does suggest a biological or ecological basis for ‘spillover’ from flying foxes to horses.

All properties with HeV cases have had some level of flying fox activity identified in the vicinity but not necessarily the presence of a roosting colony.

The exact route of transmission of HeV between flying foxes and from flying foxes to horses has not been positively identified but is thought to occur via contact or droplet transmission. HeV has been isolated from the urine, saliva, faeces and uterine fluids of flying foxes and it is thought that pasture or discarded fruit contaminated with flying fox body fluids and/or excreta are probable routes of viral transmission to horses^{3,8,9,10}. Transmission to horses via the nasal route is also plausible^{11,12,13,28}.

Horses

HeV has been detected in horse blood, oral and nasal secretions. HeV genetic material has been detected in urine, faeces and a wide range of body tissues of infected horses^{2,14,15,16,17,18}. It is possible HeV could be present in any body fluids.

Polymerase chain reaction (PCR) testing of 'natural' cases in horses has identified HeV genetic material in blood, nasal secretions and a wide range of body tissues, indicating that HeV virus is likely to be widespread throughout the body and fluids of an infected horse.

Research conducted by the Australian Animal Health Laboratory (AAHL) in 2009 using the Redlands 2008 HeV isolate, demonstrated that by the time a horse is showing clinical signs, virus is systemically widespread throughout the body and body fluids¹⁶.

The AAHL research has also shown that viral ribonucleic acid (RNA) could be detected continually in nasal swabs from as early as two days post exposure, which was three to five days before the onset of clinical signs indicating that systemic spread of the virus may be preceded by local viral replication in the nasal cavity or oropharynx.

The data indicates that nasal secretions of asymptomatic horses may pose a transmission risk during the early phase of disease that precedes viraemia, fever, or other discernable clinical signs of HeV infection. However, the increasing gene copy number recovered over time also suggests that the risk provided by these animals is relatively low compared with animals in the immediate pre-symptomatic and symptomatic stages of infection.

Early clinical signs seen during the AAHL research were depression, an increase in body temperature and heart rate and a discomfort or restlessness expressed by weight shifting between legs (both fore and hind limbs). A critical part of reducing HeV exposure risk includes early consideration of HeV in the differential diagnosis with the institution of appropriate infection control procedures until a definitive diagnosis is obtained¹⁶.

Duration and type of exposure also contributes to infection risk. Longer contact time and invasive procedures may increase the potential for acquisition of an infectious dose of virus. Certain types of contact or procedures may contribute to human infection, such as nasal intubation or routine dental procedures, where operator risk is increased even in the preclinical stage of infection.

The febrile and then symptomatic horse, particularly those late in the disease process are likely to shed more virus from a variety of excretions and pose a higher risk of disease transmission. Conducting necropsies poses a higher risk of infection transmission because of the potential for gross contamination and the handling of sharp instruments¹⁶.

Aetiology

Hendra virus belongs to the genus Henipavirus of the family Paramyxoviridae, order Mononegavirales. It has an unusually large genome which encodes six major structural proteins:

- Nucleocapsid protein (N)
- Phosphoprotein (P)
- Matrix protein (M)
- Fusion glycoprotein (F)
- Attachment glycoprotein (G)
- Large polymerase protein (L)¹⁹

HeV uses host cell surface membrane-bound ligands ephrin-B2 and ephrin-B3 as cellular receptors. The entry of HeV is initiated by the attachment of the G envelope glycoprotein to the host cell receptors ephrin B2 and/or B3, followed by activation of the F fusion protein, which triggers fusion between the viral envelope and the host cell membrane. Ephrin-B2 has widespread cellular distribution, including neurons, endothelial cells, smooth muscle surrounding arteries, placental tissue, spleen and sinusoidal lining of lymph nodes. Ephrin-B3 expression is mostly restricted to the central nervous tissue however can also be found in lymphoid cells^{19, 20}.

HeV is a fragile enveloped virus and is very susceptible outside the host to elevations of temperature and desiccation and varying susceptibility to pH changes. It usually has a short life (hours) outside the body and is rendered non-infectious by soap and detergents. However, in some experimental circumstances the virus can survive longer. For example in one experiment virus survived more than four days at 22 degrees Celsius in pH neutral bat urine. In other experiments the virus survived on mango flesh for less than two hours to more than two days depending on temperature, pH and desiccation. Viruses incubated in lychee juice showed greater persistence than either papaw or mango juice with two to three fold longer half-lives and survival for more than three days⁴¹. Fomites could therefore pose a HeV transmission risk.

Microscopically, lesions of vascular damage and vasculitis are observed in cases of HeV infection¹⁶.

It is reasonable to assume that the underlying virus-induced damage to vascular endothelium plays a major role in producing the clinical presentation (respiratory, neurological, colic etc.) and relates to the organ system/s sustaining severe and compromising endothelial damage. Consequently, the primary presenting clinical signs in infected horses can vary. As the disease progresses, a predominance of respiratory and/or neurological signs tends to be seen.

Incubation period

In experimentally infected horses the incubation period has ranged between 3–11 days^{16, 22, 23}. In naturally infected horses, the attributed incubation period has ranged between 5–16 days²⁴. The course of illness for fatally infected horses averages a little over two days from first signs to death.

Information from the confirmed cases in horses to date suggests that approximately 20 per cent can survive acute infection.

Index cases (the first confirmed cases) have typically been horses paddocked or kept outside in areas that were attractive to flying foxes. In-contact horses kept in a paddock situation with an index case have been confirmed as being infected on multiple occasions including—Mackay (1994), Proserpine (2008), Bowen (2009), Boonah, Wollongbar, South Ballina, Beachmere (2011) and Rockhampton (July 2012).

Horse-to-horse transmission of HeV appears to be more efficient in a stabled situation compared to a paddock situation. In paddock situations to date the majority (65 per cent) have involved one infected horse that died without any in-contact horses becoming infected. It appears that HeV has the potential to spread to other horses either through direct contact with infectious body fluids or excreta, or through indirect contact via contaminated fomites, including human-assisted transfer.

Two events in stables (Hendra 1994 and Redlands 2008) and one event on a property comprising multiple small paddocks (Cawarral 2009) have resulted in multiple horses becoming infected. It should be noted that all these events appear to have arisen from a horse initially becoming infected in a paddock or outside yard.

Horses experimentally infected with the original virus isolate from Hendra (1994) did not transmit infection to in-contact horses under biosafety level 4 conditions at AAHL²⁵. In this same study, a horse was infected following contact with the urine of an infected cat. It is not known whether urine from an infected cat can transmit infection to other animals or humans or horses in a natural setting.

Genetic analysis of the Redlands 2008 HeV isolate showed 99.6% similarity at the amino acid level to the original HeV isolate (Hendra 1994)¹⁶.

Other species

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^{21, 25, 26, 27, 28, 29, 30, 39}. Conversely rats, rabbits and a dog developed antibodies but not clinical signs when exposed to HeV²¹.

Experimental studies in Canada (2010)³⁰ showed that the response of pigs to inoculation with large doses of virus ranged from no clinical disease to severe interstitial pneumonia. This work has demonstrated that pigs can be infected experimentally, but it does not confirm whether or not pigs can be infected naturally. However, pigs can be infected naturally with the closely related Nipah virus.

Previous surveillance in commercial piggeries in Queensland indicated no evidence of HeV or Nipah virus infection in pigs³¹.

In July 2011, test results confirmed the presence of antibodies to HeV in a dog sampled on a property in Queensland. It was reported that the dog did not show any clinical signs of illness. No HeV genetic material was detected by PCR in samples collected from the dog on three occasions over a three week period. This was the first reported case of HeV antibody detection in a dog outside of an experimental setting.

In July 2013 a dog was confirmed to be infected with HeV on a property in New South Wales.

Both cases of HeV infection in dogs were on properties where HeV infection had been confirmed in horses and where potential for close contact between the dog and horse existed.

The few cases of HeV infection in dogs (both natural and experimental) have shown no overt clinical signs.

Humans

Seven cases of human infection with HeV have been recorded. All cases had a high level of exposure with blood or body fluids from an infected horse. In all cases, HeV had not been considered a diagnosis for the horse at the time of exposure and occurred before the equine case was confirmed as HeV. Four people died as a result of HeV infection—a case fatality rate of 57 per cent. The infectious dose for HeV in humans is unknown. Epidemiological evidence from at least one of the seven known human infections with HeV suggests human infection most plausibly occurred from a horse in the late incubation period, i.e. up to 72 hours before onset of clinical signs in the horse³².

Disease epidemiology in humans to date is consistent with infection being caused by direct contact with respiratory or oral secretions and by other equine tissues and fluids, or by droplet transmission. However, procedures that generate aerosols may pose a risk of infection.

3.3 Hendra virus vaccine

A recombinant subunit vaccine for HeV was released commercially in November 2012 following evaluation and issue of a Minor Use Permit (PER13510) by the Australian Pesticides and Veterinary Medicines Authority (APVMA). Vaccine use should be in accordance with the permit conditions.

The subunit vaccine is based on a recombinant soluble version of the HeV attachment glycoprotein (sG). It was developed subsequent to initial research which demonstrated that subunit sG vaccine constructs could prevent henipavirus clinical disease and virus transmission or shedding in small animal models^{33,34,35}.

The vaccine works by stimulating the production of antibodies that neutralise HeV by binding to the G glycoprotein of the virus, rendering it unavailable for attachment to the cells of the animal and preventing infection³⁶.

Like most paramyxoviruses, HeV infection of host cells involves two viral glycoproteins. The attachment (G) glycoprotein binds to the host cell receptors; ephrin B2 and ephrin B3, important bi-directional cell-cell signalling molecules that are highly conserved and widely expressed particularly within the nervous and vascular systems across all mammalian species. The second viral glycoprotein is the fusion (F) protein, which upon triggering facilitates the fusion between the viral and host cellular membranes³⁷.

In the initial vaccine efficacy trials, there was no evidence of viral shedding by immunised horses after HeV challenge, as reflected by PCR negative test results on all daily clinical samples. Following euthanasia of immunised horses (7 to 9 days post challenge, and 1–3 days after clinical signs first became apparent in control animals), there was no evidence of HeV viral replication in any tissue of immunised horses collected at post mortem examination, after what would be expected to be the period of acute infection³⁶.

To demonstrate the immunogenicity of the vaccine under field conditions two trials were conducted. Horses were given two single doses of the vaccine by intramuscular injection on Days 0 and 21. 100 per cent of vaccinated animals in both trials seroconverted, confirming that two doses of vaccine given three weeks apart are sufficient to generate an antibody response in horses from four months of age. Serum neutralising antibody levels on day 42 (three weeks after the second dose of vaccine) in all vaccinated horses from both trials were equivalent to those seen in the earlier efficacy studies by challenge as described above, in which vaccinated horses were shown to be protected from challenge with HeV³⁶.

At approximately six months post-vaccination, when horses were challenged with HeV via the intranasal route, all were protected from clinical signs of HeV disease. In addition, virus was not reisolated from any clinical samples (collected pre-and post-mortem) from any of the horses, and there was no evidence of virus spreading beyond the site of administration (i.e. the upper respiratory tract). In non-immunised (surrogate control) ferrets, viral infection was detected and all succumbed to acute HeV infection³⁶.

Vaccination is the single most effective way of reducing the risk of HeV infection in horses. Human infection and death have occurred following high-level exposure to body fluids from an infected horse. Vaccinating horses is an important measure to prevent this occurring and provides a public health and workplace health and safety benefit. Widespread uptake of the horse vaccine has the potential to significantly reduce the number and risk of human exposures.

Similar to all vaccines, no one vaccine can be 100 per cent effective in all animals on all occasions, therefore it is critical that people in contact with horses continue to practice good biosecurity and hygiene, even if a horse is vaccinated.

Under the APVMA Minor Use Permit only registered veterinarians who have completed the training module are accredited to administer the vaccine and all horses must be microchipped prior to vaccination.

When requesting laboratory testing for HeV, record whether the horse is vaccinated and if so also note the microchip details on the specimen advice sheet (see section 9 ‘Sampling, dispatch and laboratory testing’).

More information about the HeV vaccine for horses including the process for veterinarians to become accredited to administer the vaccine can be found at www.vetsaustralia.com.au

3.4 Cedar virus – a newly discovered Paramyxovirus

Cedar virus (CedPV) is a newly discovered Paramyxovirus which has been isolated from flying foxes and shares significant features with HeV and Nipah virus. It displays antigenic cross-reactivity with henipaviruses and uses the same receptor molecule (ephrin B2) for entry during infection. Preliminary challenge studies with CedPV in ferrets and guinea pigs, confirmed virus replication and production of neutralising antibodies, although clinical disease was not observed³⁸.

The research noted that the major genetic difference between CedPV and HeV or Nipah virus was within the coding strategy of the P gene, which is known to play an important role in evading the host innate immune system. This discovery is part of ongoing research.

4. Hendra virus investigation planning and preparedness

Warning

Safety of people is the primary consideration when investigating potential HeV cases. If any person is concerned about their health, they should seek medical advice and contact their local GP, their nearest Queensland Health Population Health Unit or the Queensland Health 24-hour hotline on 13 HEALTH (13 43 25 84).

Apply the precautionary principle. An infected horse can excrete HeV in nasal or nasopharyngeal secretions for several days before the onset of clinical signs and while symptomatic. A strongly symptomatic horse poses the greatest transmission risk to other horses and humans through a variety of body fluids and excretions.

Dead horses can be sampled adequately for HeV testing without conducting a complete necropsy. Necropsy on a recently dead HeV infected horse is a very high risk activity because the horse is likely to be maximally infectious at this time, and should only be undertaken by a person who is suitably experienced and knowledgeable about how to manage exposure to HeV and how to use the relevant personal protective equipment.

Veterinary practices should develop and implement infection control procedures to manage the risks associated with investigating and sampling potential HeV cases.

Infection control procedures are currently the primary defence for managing both horses in the pre-clinical phase, (where they may excrete HeV but still appear clinically normal) and clinically ill horses.

These guidelines provide information that should support development of such plans and procedures, but may not contain all the information needed. Individuals should source extra information to suit their own particular situation.

4.1 Plan outline

Good preparation for the investigation of potential HeV cases should be undertaken as part of normal practice management and before a veterinarian conducts a field investigation. Plans, procedures, appropriate equipment and training are all required to assist the smooth, timely handling of investigations.

Biosecurity Queensland recommends that veterinary practices have a HeV case investigation procedure printed and readily accessible in each practice vehicle.

This should act as a checklist to ensure veterinarians adhere to all key elements in the investigation of potential HeV cases.

Laminating the investigation procedure will allow it to be carried onto the property and decontaminated before exiting the property.

4.2 Summary of steps to consider

The following is the minimum number of steps that need to be considered before investigating a horse with potential HeV infection:

1. From the client's initial phone call or enquiry, obtain as much history as possible and make an initial case assessment.
 - Ask if the horse is vaccinated against HeV (you may wish to refer to the HeV Vaccination Registry to confirm vaccination status).
 - Be familiar with the various HeV clinical presentations (see section 5 'Clinical presentation').
 - Always take appropriate precautions based on any suspicion of HeV; do not wait for confirmation. Even with vaccinated horses it is important to take appropriate precautions as no vaccine is completely effective in 100 per cent of animals.
2. Know the notifications and other legal considerations required.
 - There is a legal requirement to notify Biosecurity Queensland on 13 25 23 (business hours) or the Emergency Animal Disease Watch Hotline on 1800 675 888 (any time), or by direct contact with a government veterinary officer (see section 6 'Notifications and other legal considerations'). There may be individual and practice requirements for notification as well.
3. If HeV is suspected before you enter the property, know the precautions and procedures to follow. These include:
 - Workplace health and safety precautions that the veterinary practice agrees must be taken (see section 7 'Workplace health and safety precautions').
 - Duties to ensure the health and safety, so far as is reasonably practicable of yourself, workers and others who are assisting you (see section 7 'Workplace health and safety precautions').
 - The choice of personal protective equipment and disinfectants used (see section 8 'Infection control and biosecurity precautions').
 - Entry and exit procedures (see Appendix 2 'Entry/exit process').
 - Appropriate biosecurity advice for clients.
4. If you do not suspect HeV before the examination of a horse:
 - Know the immediate steps to take to minimise the risk and exposure to you and others if there is unplanned contact with an ill horse that may be infected with HeV (see section 8.7 'Unplanned contact with a suspect horse').
5. Develop a sampling protocol.
 - Prepare a HeV sample collection procedure that includes:
 - samples required and the safest methods of collecting them
 - appropriate equipment needed.

- Prepare a HeV sample dispatch procedure (see section 9 ‘Sampling, dispatch and laboratory testing’). This procedure will explain:
 - how to safely pack samples or which couriers will provide this service
 - where to dispatch samples (Note: dispatch HeV samples to a Queensland Government laboratory, not a private laboratory)
 - which couriers will transport samples and which couriers provide an out-of-hours service.
- 6. Prepare biosecurity advice that can be given to clients (see section 10 ‘Biosecurity advice to clients’). This information will explain what owners and managers involved with managing the case and the site should do.
- 7. Prepare advice about disposal of carcasses (see section 11 ‘Carcass disposal’). This should detail:
 - The disposal method of choice for the area (deep burial on site if possible).
 - Advice for owners/contractors to assist safe disposal and to prevent disease spread.
- 8. Prepare a list of contacts that you will be required to liaise with and record the contact details (see section 13 ‘List of contacts’).
- 9. A response to HeV will be a team effort and a number of different people may be involved, all with different responsibilities.
- 10. Understand your responsibilities regarding media and confidentiality (see section 14 ‘Media and confidentiality’).

4.3 Training

Veterinary practices developing a HeV investigation plan must conduct training on its use and application to ensure that all persons are competent to manage a HeV investigation. Regular training should be conducted and training records should be kept.

The use of recommended personal protective equipment (PPE) will require training if PPE is not routinely used. A number of PPE suppliers provide training in correct fitting and use of PPE. The AVA has produced an online video, *Suit Up*, regarding appropriate PPE including correct donning and doffing. The video can be found on the AVA website at www.ava.com.au

The packaging of samples for laboratory testing may also require training (available from private providers) to ensure diagnostic samples comply with transport requirements. Alternatively, a decision can be made (and recorded in the procedure) to use a courier company that will provide a packing and transport service for such samples.

5. Clinical presentation

HeV infection of horses typically causes an acute illness that is rapidly fatal¹⁶. There are no pathognomonic signs that define HeV infection in horses. Horses infected with HeV have shown variable and sometimes vague clinical signs.

There is, however, a range of clinical signs recorded from confirmed cases, including some signs that have been common to many confirmed cases. This necessitates applying professional veterinary judgement to ill horses to decide whether HeV may be involved. Laboratory testing is required to confirm whether a horse is infected with HeV.

HeV should be considered where there is acute onset of clinical signs that may include increased body temperature (not all cases have been febrile at examination), increased heart rate and rapid progression to death associated with either respiratory and/or neurological signs.

Note: Based on research conducted at AAHL, an elevated temperature and heart rate should be considered as early warning of the possibility of HeV infection¹⁶. Progression to include other clinical signs, as mentioned below, increases the possibility of HeV infection.

The precautionary principle should be applied at the first indication of clinical illness and also when conducting invasive/aerosol-generating procedures of the respiratory tract and other high-risk procedures (e.g. endoscopy of the upper and lower respiratory tract, dentistry using power floats, necropsy, broncho-alveolar lavage and nasal lavage).

The AAHL research indicates that nasal secretions of asymptomatic horses may pose a transmission risk during the early phase of disease that precedes viraemia, fever or other discernable clinical signs¹⁶. Appropriate infection control and biosecurity measures must be applied to the examination, sampling and management of such horses.

In most recorded cases, there has been strong presentation of clinical signs however occasional cases have demonstrated a much milder presentation of clinical signs. Several horses have been confirmed to be infected by laboratory testing in the absence of overt clinical signs.

HeV has an affinity for endothelial cells and causes systemic vasculitis. The organ/system where the greatest damage occurs would appear to contribute directly to the clinical signs seen.

Information from the confirmed cases to date suggests a mortality rate of approximately 80 per cent.

The following have been seen commonly in horses with HeV infection:

- acute onset of illness
- increased body temperature
- increased heart rate
- discomfort/weight shifting between legs (both fore and hind limbs)
- depression
- rapid deterioration, usually with respiratory and/or neurological signs.

Respiratory signs, including:

- pulmonary oedema and congestion
- respiratory distress—increased respiratory rates
- terminal nasal discharge—can be initially clear progressing to stable white froth and/or stable blood-stained froth
- pulmonary involvement leading to terminal weakness, ataxia and collapse.

Neurological signs, including:

- ‘wobbly gait’ progressing to ataxia
- altered consciousness—apparent loss of vision in one or both eyes, aimless walking in a dazed state
- head tilting
- circling
- muscle twitching—myoclonic spasms have been seen in acutely ill and recovered horses
- urinary incontinence
- recumbency with inability to rise +/- thrashing
- facial paralysis and/or a locked jaw
- spasms of the jaw or involuntary chomping
- opisthotonus
- seizures.

Other observations that may be seen in horses with HeV infection:

- previous unexplained horse deaths
- facial oedema and/or swollen lips
- teeth grinding
- muscle trembling
- altered gait or high stepping
- wide based stance
- anorexia
- congestion of oral mucous membranes
- a high case fatality rate within 48 hours where there are multiple cases
- colic-like signs (rolling and thrashing or quiet abdominal sounds on auscultation of the abdomen in pre-terminal cases)
- straining with difficulty passing manure
- stranguria (difficult urination)—seen in several terminal cases in both males and females
- dribbling urine—seen in some terminal cases
- protruding penis
- hot hooves
- bad breath/halitosis
- delayed blood clotting times.

5.1 Hendra virus and other animals

Laboratory studies have shown that species other than horses can develop disease when inoculated with HeV. A study done in 1995 showed that mice, rats, rabbits, chickens and dogs did not develop any clinical disease following subcutaneous inoculation with HeV²¹. In the same study, cats and guinea pigs became infected and succumbed to the disease. Equivocal neutralising antibody titres were detected in three of four rats and one of two dogs in the study, while rabbits developed unequivocal neutralising antibody titres.

Experimental research has found that cats, pigs, hamsters, ferrets, guinea pigs and African green monkeys can be infected with HeV and develop fulminant clinical signs^{21,25,26,27,28,29,30,39}.

AAHL research reassessed the role of mice as an animal model for HeV infection and found that mice are susceptible to HeV infection after intranasal exposure, with aged mice reliably developing encephalitic disease²⁸. This research proposed an anterograde route of neuroinvasion to the brain, possibly along olfactory nerves. This was supported by evidence for the development of encephalitis in the absence of viraemia and the sequential distribution of viral antigen along pathways of olfaction in the brain of intranasally challenged animals.

In July 2011, test results confirmed the presence of antibodies to HeV in a dog sampled on a property in Queensland. It was reported that the dog did not show any clinical signs of illness. No HeV genetic material was detected by PCR in samples collected from the dog on three occasions over a three week period. This was the first reported case of HeV antibody detection in a dog outside of an experimental setting.

In July 2013 a dog was confirmed to be infected with HeV on a property in New South Wales.

Both cases of HeV infection in dogs were on properties where HeV infection had been confirmed in horses and where potential for close contact between the dog and horse existed.

The few cases of HeV infection in dogs (both natural and experimental) have shown no overt clinical signs.

The potential exists for susceptible non-equine domestic species to become infected with HeV and this should be considered as part of the management of HeV incidents.

Biosecurity Queensland samples susceptible species other than horses for HeV testing, if they have been assessed as having had close contact with a confirmed HeV infected horse.

If private veterinarians are considering sampling species other than horses for HeV testing, they should contact Biosecurity Queensland first to discuss the case.

Further research is underway in the area of susceptibility and transmission of HeV in non-equine species, including dogs.

5.2 Differential diagnoses

As there are no pathognomonic signs that define HeV infection in horses and given horses infected with HeV have shown variable and often vague clinical signs, there are a large number of differential diagnoses.

6. Notifications and other legal considerations

6.1 Notifications

HeV is a notifiable disease in Queensland under the *Stock Act 1915* and the *Exotic Diseases in Animals Act 1981*. This places an obligation on a veterinarian who suspects, diagnoses or confirms the presence of a notifiable disease to notify the nearest government veterinary officer as soon as possible and by the quickest means of communication available.

Obligations to notify based on suspicion, diagnosis or confirmation of HeV are also present in the *Veterinary Surgeons Act 1936*.

Equivalent legislative provisions are in place in other states and territories of Australia and veterinarians working in these areas should be aware of the relevant legislation.

If it appears that human exposure may be associated with the case, the veterinarian should also include this information when contacting the government veterinarian. Notification is also an opportunity to seek advice.

Notification in Queensland can be made by contacting one of the following:

- Biosecurity Queensland on 13 25 23 (business hours)
- Emergency Animal Disease Watch Hotline on 1800 675 888 (24 hours).

When a veterinarian makes contact, they should explain that they are a veterinarian calling to notify of a possible case of HeV.

Biosecurity Queensland will contact Queensland Health whenever HeV is confirmed or highly suspected as per an agreed notification protocol. Queensland Health will decide whether any people require monitoring and/or medical assistance. To make this assessment, Queensland Health will work with the veterinarian and the horse owner to identify the people they may need to contact.

Biosecurity Queensland will notify the Australian Veterinary Association, Equine Veterinarians Australia and other bodies as appropriate. Notification to industry bodies is made only after a positive diagnosis but not in the instance of possible cases (unless there are exceptional circumstances).

6.2 Quarantine and other restrictions

Biosecurity Queensland may place a premises in quarantine and implement a disease control program where a suspected or confirmed HeV case occurs in an animal. In Queensland, HeV incidents are managed under the *Exotic Diseases in Animals Act 1981*.

A private veterinary practitioner does not have the legal powers to quarantine a property but should advise the owner of voluntary actions to implement for managing the immediate situation including prevention of disease transmission to humans and other animals.

If a decision is made to quarantine the property, Biosecurity Queensland will manage the animal disease control program. Biosecurity Queensland may retain the services of the private veterinary practitioner as part of the response. If this course of action is chosen, Biosecurity Queensland will enter into an agreement with the veterinary practitioner.

If a property is not quarantined, the investigating veterinarian will continue to manage the animal disease investigation. Where HeV is a differential diagnosis, the aim of an investigation is to:

- maintain safety of all people and animals involved
- clinically assess the situation
- obtain the appropriate samples to help make a definitive diagnosis
- give appropriate biosecurity advice to the owner and report findings.

In addition to any restrictions implemented by Biosecurity Queensland to manage the disease in animals, Queensland Health may implement restrictions or make recommendations to manage the disease or potential disease in people. Queensland Health will work with the veterinarian and the horse or property owner and any other relevant person to identify the people they may need to contact.

If a business or undertaking is involved in a HeV incident, Workplace Health and Safety Queensland may provide advice and/or monitor compliance with workplace health and safety legislation.

If any person has concerns about possible exposure of people to a horse infected with HeV, they should seek medical advice and contact their general practitioner, local hospital emergency department or local public health unit. For general enquiries about HeV infection in humans, call the Queensland Health Hotline on 13 HEALTH (13 43 24 84).

7. Workplace health and safety precautions

The *Work Health and Safety Act 2011* (the Act) places duties on people at workplaces for health and safety. Veterinarians and practice principals who conduct a business or undertaking must ensure, so far as is reasonably practicable, the health and safety of themselves and workers (e.g. employees, students, trainees, contractors, sub-contractors and volunteers). The same duty also applies to any other people who may be at risk from work carried out by the business (e.g. clients).

The Act sets out specific duties which a person conducting a business or undertaking (PCBU) must comply with, so far as is reasonably practicable. These include:

- Providing and maintaining a working environment without risks to health and safety.
- Providing and maintaining safe plant and structures.
- Providing and maintaining safe systems of work.
- The safe use, handling and storage of plant, structures and substances.
- Providing adequate facilities for the welfare of workers at workplaces (e.g. washrooms and dining areas).
- Providing any information, training, instruction or supervision that is necessary to protect all persons from risks to their health and safety arising from work.
- Monitoring the health of workers and the conditions at the workplace to prevent illness or injury.

The Act also requires the PCBU to notify notifiable incidents, including occupational infection with HeV, to Workplace Health and Safety Queensland.

Veterinarians and practice principals should consider the following workplace health and safety precautions:

- Discuss HeV vaccination with horse owners. The HeV vaccine's ability to not only protect horses from infection but also to break the cycle of virus transmission from horses to humans provides a public health and workplace health and safety benefit.
- Conduct a risk assessment for HeV and develop a plan for responding to suspected HeV cases with supporting policies, procedures and training.
- Implement a triage system to identify HeV risk factors when taking bookings for horse consultations.
- Routinely assess HeV risks for all horse contact as a standard work practice
- Provide a dedicated HeV kit with appropriate equipment and PPE, and provide training in the proper use of PPE.

Adopt standard precautions, as developed by the AVA, for all contact with horses. This includes:

- Cover cuts and abrasions with a water-resistant dressing and adopt personal hygiene, including hand hygiene before and after horse contact, between horses and after removing PPE.
- PPE including disposable gloves for contact with blood, body fluids, excretions, non-intact skin and mucous membranes, and protective clothing and facial protection where there is a risk of droplets, splashes and sprays of blood and body fluids.
- Appropriate reprocessing of reusable veterinary equipment and horse gear after use and between horses.

- Safe handling, transport, storage and disposal of clinical waste (including sharps).
- Safe handling, transport, storage and cleaning of contaminated clothing and other laundry.
- Safe handling, transport, storage and disposal of pathology specimens.
- Safe handling and disposal of animal excreta and stable manure.
- Stable hygiene and environmental cleaning using appropriate cleaning agents and disinfectants.
- Management of blood/body fluid spills and accidental blood/body fluid exposures and sharps injuries.

Adopt standard precautions plus airborne precautions for high risk procedures on clinically normal horses (e.g. necropsy, aerosol-generating procedures involving nasopharyngeal secretions) and for contact with potential or confirmed HeV. This includes PPE as detailed in section 8.

If there is a potential case of HeV in a horse on your premises, you should:

- Assess zoonotic risks and take steps to manage these risks.
- Follow the workplace health and safety guidance outlined in your HeV case investigation procedure, ensuring appropriate infection control practices and use of PPE.
- Isolate the sick or dead horse from all people, all other horses and all other domestic animals on the premises and from public access areas where possible.
- Make the necessary notifications and if appropriate discuss the required case plan with Biosecurity Queensland.
- Develop a biosecurity plan to safely manage the situation on your premises, or implement an already agreed site biosecurity plan.
- Establish entry/exit procedures.
- Limit human contact with the sick or dead horse and with all horses on the site that have had contact with the horse until it is resolved that the problem is not HeV or that HeV is no longer present.
- Send required samples to a government veterinary laboratory for HeV testing (see section 9 ‘Sampling, dispatch and laboratory testing’).
- Ensure sharps safety when collecting samples (e.g. do not recap needles and consider using a safety engineered blood collection system).
- Advise any person who is concerned about their health at any time to seek medical advice.
- If necessary seek advice from Biosecurity Queensland about disposal of any dead horses pending sample results. Where carcass disposal occurs on property, this could include ensuring that a disposal option is used that is safe and does not cause environmental harm or contamination. Disposal should be undertaken by people who are aware of the risks and familiar with appropriate disposal methods.

If a potential case of HeV is identified when visiting other premises in a professional capacity, workplace health and safety duties continue to exist because the premises becomes the workplace of the attending veterinarian and their staff while they are on site.

If there is a potential case of HeV in a horse on a premises that you are visiting, in addition to the above, you should:

- Consult and cooperate with other people at the premises who may have a workplace health and safety duty for the matter (e.g. the property owner).
- Inform the owner/manager about the zoonotic risks and how to protect against infection, including personal hygiene and the use of PPE.
- Advise the owner/manager to:
 - isolate the sick or dead horse from all people, all other horses and all other pets on the premises and from public access areas where possible
 - develop a biosecurity plan to manage the situation on the premises
 - limit human contact with the sick or dead horse and all other horses on the property that have had contact with the horse until it is resolved that the problem is not HeV or that HeV is no longer present on the property
 - advise immediate neighbours that a horse is being investigated for HeV infection
 - if necessary seek advice from Biosecurity Queensland about the disposal of any dead horses pending sample results and to ensure that the disposal contractor is aware of the risk.
- Ensure the health and safety of any person at the premises who assists you with a veterinary assessment or procedure (e.g. restraining the horse) and ensure that they understand your instructions for their health and safety
- Consider using a trained veterinary professional to assist you (e.g. veterinary nurse) rather than using the horse owner or carer.

If Biosecurity Queensland places a property under quarantine for HeV, that property becomes a workplace of Biosecurity Queensland. The property owner may also have health and safety duties (e.g. if running a business such as a riding school). Biosecurity Queensland will work closely with the owner/manager to discharge the health and safety duties while the property remains under quarantine and will determine what activities take place in the quarantined area of that property.

For all other non-quarantined properties, health and safety duties remain with the owner/manager/ carer and with the private veterinary practitioner and Biosecurity Queensland if they enter the property in a business capacity.

For more information, contact Workplace Health and Safety Queensland on 1300 369 915 or visit the Workplace Health and Safety Queensland website at www.worksafe.qld.gov.au

8. Infection control and biosecurity precautions

8.1 Personal safety and prevention of zoonotic risk

Human infection with HeV has a high case fatality rate. Human infections have occurred from high levels of exposure to the respiratory secretions and/or blood from a horse infected with HeV (both live horses and dead horses at necropsy examination). Great care is needed to ensure the personal safety of the veterinarian and others who may be assisting.

Human HeV infection has been associated with close contact with an infected horse in the late incubation period while it was asymptomatic³².

A major problem has been that HeV is often diagnosed retrospectively in horses (i.e. after human exposure has occurred). This reinforces both the need for early diagnostic consideration of HeV and the institution of appropriate infection control procedures. It also reinforces the importance of developing appropriate infection control procedures to use in day-to-day situations.

In particular, perform hand hygiene and treat tissues, blood and other body fluids (especially respiratory and nasal secretions and saliva) and excretions as potentially infectious. Take appropriate precautions to prevent any direct contact with, splash back of or accidental inoculation with these body fluids or excretions.

Vaccination of horses will help prevent zoonotic risk. The HeV vaccine's ability to not only protect horses from infection but also to break the cycle of virus transmission from horses to humans provides a public health and workplace health and safety benefit. Widespread uptake of the horse vaccine has the potential to significantly reduce the number and risk of human exposures.

The *Medical Journal of Australia* contains an article titled 'Hendra virus infection in a veterinarian' detailing a case where a veterinarian became infected with HeV while performing a necropsy on a HeV infected horse without taking adequate precautions⁴⁰.

The article 'Human Hendra virus encephalitis associated with equine outbreak, Australia, 2008' was published in *Emerging Infectious Diseases*³². It details the HeV infection of two veterinary clinic staff.

Further information on infection control recommendations can be found in the multi-agency document, *Hendra Virus Infection Prevention Advice* on the Queensland Health website at www.health.qld.gov.au.

8.2 Personal protective equipment

Work health and safety legislation requires the person conducting a business or undertaking to ensure that PPE provided to workers is suitable having regard to the nature of the work and any hazard associated with the work, and is of a suitable size and fit and reasonably comfortable for the worker who is to use or wear it. The PPE must be maintained, repaired or replaced so that it continues to minimise risk, including by ensuring that it is clean and hygienic, and in good working order. The person must also ensure, so far as is reasonably practicable, that the PPE is used or worn by the worker or any other person at the workplace.

It is recommended that veterinarians discuss their specific safety needs with a supplier of safety equipment. Veterinarians will receive expert advice and be provided with a selection of products appropriate for individual situations from the vast array of safety equipment products available. Alternatively PPE can be obtained from safety supply stores and most hardware stores.

PPE forms a part of the risk management approach to personal safety. Other factors, such as vaccination of horses, HeV planning and preparedness, infection control practices and managing exposure to HeV risks, also play their part. PPE should be combined with these to provide the best protection possible.

The PPE suitable for use with HeV may not be used routinely in everyday veterinary practice. You should source PPE ahead of time and receive training in its correct use, storage and maintenance.

Note: Biosecurity Queensland does not have specialist PPE trainers and sources its PPE and training from commercial PPE providers to meet the various demands of emergency animal disease response.

The PPE selected is intended to form a barrier between the person and the virus and should include gloves, particulate respirator, eye protection, overalls and impervious boots.

The following PPE is used by Biosecurity Queensland officers when investigating potential HeV situations to collect samples to test for HeV:

- impervious rubber boots
- splash-proof overalls (long sleeves with a hood) or cotton or disposable overalls with impervious or splash-proof apron
- disposable impermeable gloves (nitrile gloves recommended), double-gloved
- face shield or safety eyewear (to protect against facial splashing)
- a particulate respirator.

Where Biosecurity Queensland officers assess that a higher level of risk exists or a horse is known HeV positive, the standard of PPE is increased accordingly.

Note: Splash-proof recommendation is used for PPE as this usually means the items are lighter and better suited to hotter conditions that may be faced during investigation. Impervious PPE, particularly impervious overalls/suits, need to be used with great care as there is a real risk of rapidly overheating, particularly if used in direct sunlight.

Using PPE can create significant heat stress issues. The risk of heat stress should be managed where ever possible (e.g. by ensuring adequate hydration before commencing work, scheduling work times to avoid the hottest part of the day, using cool clothing and cooling scarves and vests, drinking fluids, using portable shade, taking frequent rest breaks in cool areas, rotating teams or using a buddy system, and knowing the signs of heat stress and stopping work if these signs start to develop).

Ensure you have adequate PPE supplies for repeat visits and for those assisting you. Always remove PPE carefully to avoid contamination and perform hand hygiene after removing PPE. Decontaminate reusable PPE after use and do not reuse disposable PPE.

The AVA has produced an online video, *Suit Up*, regarding appropriate PPE for investigation of a horse suspected of being infected with HeV, including correct donning and doffing. The video can be found on the AVA website at www.ava.com.au

8.2.1 Hendra virus personal protective equipment rebate program

The Queensland Government have implemented a PPE rebate program to assist private veterinary practices offset the cost of eligible PPE used in the testing of suspect HeV cases.

Queensland veterinary practices can apply for a start-up rebate for eligible PPE purchased since 24 March 2012. Private veterinary practices are eligible for a rebate of up to \$250 for each eligible veterinary surgeon employed or engaged in the practice.

In addition, veterinarians who submit a sample for HeV testing via the Biosecurity Queensland Veterinary Laboratory Network System can apply for a replenishment-rebate. This is an additional rebate of \$250 to replenish PPE used in taking samples in the investigation of a horse suspected of being infected with HeV. For more information on this program including eligibility criteria visit www.qraa.qld.gov.au or call 1800 623 946.

8.3 Respiratory protective equipment

It is recommended that veterinarians discuss their specific respiratory protective equipment (RPE) needs with a supplier of safety equipment. Veterinarians will receive expert advice and be provided with a selection of products appropriate for individual situations from the vast array of safety equipment products available.

The minimum level of respiratory protection for investigating potential HeV situations is a P2 disposable facepiece respirator or a P2 filter in a half facepiece respirator. For a higher level of protection a P2 or P3 filter in a full facepiece respirator or a powered air purifying respirator (PAPR) can be used.

Note: A standard surgical facemask is not a respirator and will not provide respiratory protection because it does not have adequate filtering and/or fitting properties. Surgical masks should not be worn for suspected or confirmed HeV cases. A dust mask will not filter bioaerosols and is also unsuitable.

The following comments in this section are drawn from a range of existing safety recommendations and previous experience:

- Disposable facepiece respirators with exhalation valves can reduce moisture build up and breathing resistance and increase user comfort.
- To ensure proper protection from close fitting RPE it is essential that an adequate face seal is achieved. Facial hair including a beard, long moustache, side burns or stubble growth may interfere with the face seal. Hence, people who are not clean-shaven should not take part in any investigations unless different RPE is used (e.g. a PAPR that draws air in through a filter and supplies it to a hood worn over the head). Both P2 and P3 filters can be used with a PAPR. The PAPR device can also be used to provide a higher level of protection for aerosol-generating procedures and a higher level of comfort in hot environments.
- When a P3 filter is used in a half facepiece respirator, a protection factor equivalent to using a P2 filter is achieved. For P3 filter classification a full facepiece respirator or a PAPR with either a full facepiece or head covering is required.
- Fluid resistant brands of disposable P2 facepiece respirators are available for situations where fluid strikethrough may occur.

8.4 Training in the use of personal protective equipment

Work health and safety legislation requires that workers are provided with information, training and instruction in the proper use and wearing of PPE and its storage and maintenance.

It is recommended that veterinarians consult a commercial provider for their PPE training requirements. This should include training in correct donning and doffing procedure (putting on and taking off PPE), respirator fit checking, correct storage, maintenance, cleaning, disinfection and disposal of PPE.

Fit checking allows the selected respirator to be donned correctly and fit checked to ensure it has sealed on the face correctly. A fit check should be performed each time a respirator is donned. Do not handle or touch the respirator once it has been correctly positioned on the face, as this may modify the face seal.

Respirator fit testing is a method of determining the brand and size of respirator that is best suited to an individual's facial characteristics, and can be performed using qualitative or quantitative test methods. Veterinarians should provide respirator fit testing to ensure that they and their workers know which respirator will best protect them.

It is recommended that all records of PPE training and results of fit testing be retained by the practice.

Refer to Australian Standards AS 1715 for information on implementing a respiratory protection program.

8.5 Disinfectants

Specific testing of disinfectant compounds against HeV has not been conducted.

From the *AUSVETPLAN decontamination manual*, HeV is a member of Category A viruses. This category of viruses contains a lipid envelope.

Disinfectants named here have been drawn from the list that is known to have either effect against all viruses or specific action against Category A viruses.

Disinfectants include:

- soaps and detergents
- Virkon®
- hypochlorites
- iodophors/iodine
- biguanidines (e.g. chlorhexidine)
- quaternary ammonium compounds.

Others are named in AUSVETPLAN but these require special precautions for their safe use and are not listed here.

The use of any disinfectants that are hazardous chemicals must be in accordance with the hazardous chemicals provisions of the *Work Health and Safety Regulation 2011*.

8.6 Entry/exit process

Establishing entry/exit procedures, whether for routine property visits or for possible HeV investigations, provides a clear process to apply relevant infection control procedures, including PPE, so that personal safety and disease control are managed.

The broad principles are:

- Define the edge of the hot or dirty area (i.e. the area where HeV contamination may be present).
- Select a site at this edge that has good access from the cold or clean area.
- At this site establish an entry/exit decontamination site. This site allows you to prepare safely in the cold area, including donning of PPE, preparing sampling equipment, preparing disinfectant solutions.
- Only enter the hot area when you are fully prepared.
- The arrangement of this site then allows you to exit through a decontamination process that protects personal safety and prevents disease spread.
- If repeat visits are likely or if other colleagues may visit, mark the site clearly so the same site is used each time. This prevents other people setting up on potentially contaminated ground.
- Recommend this process to other people who may need to access the hot area.

A more detailed outline for the entry/exit process can be found at Appendix 2.

8.7 Unplanned contact with a suspect horse

The risk of unplanned contact with a suspect horse should be addressed when developing a veterinary practice HeV investigation procedure (see section 4 ‘Hendra virus investigation planning and preparedness’).

To minimise exposure risk, it is strongly recommended that a dedicated field kit appropriate for managing possible HeV cases (including PPE, cleaning agents, disinfectants, sampling equipment and waste disposal bags) is compiled and available to veterinary practitioners. This will provide veterinarians with ready access to the equipment needed to adequately protect themselves and others against exposure in situations where there is no prior suspicion or warning.

If HeV is suspected during routine work where no specific precautions have been taken, follow these steps:

- Minimise exposure. Withdraw to a safe area and instruct accompanying people to do the same.
- Assess the degree of exposure and use soap and water to wash off contamination—shower if necessary (where this is possible).
- If exposure has occurred, seek prompt medical advice and also contact the local Queensland Health Population Health Unit.
- Notify Biosecurity Queensland of the case.

If the veterinarian is in a position to proceed with the case, follow these steps:

- Assess whether it is safe (for veterinarians and assistants) to re-enter the ‘dirty/hot’ area to sample the horse/s. Only if it is safe to proceed and the level of PPE is adequate, continue with the investigation as per the veterinary practice’s HeV case investigation procedure.
- Seek advice from Biosecurity Queensland as required.

9. Sampling, dispatch and laboratory testing

Note: It is the horse owner's responsibility to meet the cost of transporting samples to a Biosecurity Queensland veterinary laboratory. Biosecurity Queensland will meet all laboratory testing costs to test samples for HeV in diagnostic cases (charges apply for HeV health testing).

Private veterinary practitioners should make the final decision about whether to collect samples and submit them for laboratory testing with reference to these guidelines and, where required, obtain advice from Biosecurity Queensland.

All HeV testing is conducted at government laboratories. Biosecurity Queensland conducts HeV testing at the Coopers Plains laboratory. Direct submission of samples to the Coopers Plains laboratory will achieve the shortest turnaround times.

If HeV is a differential diagnosis then it should be excluded by laboratory testing before samples are sent to private laboratories for diagnostic testing for other diseases.

Veterinarians should phone ahead to advise the laboratory of the impending submission so appropriate arrangements to facilitate testing can be made, especially outside of business hours (See section 13 'List of contacts' for laboratory contact details).

Horses vaccinated against HeV will influence the serological test selection and the interpretation of serological results. Both vaccination with the sG subunit vaccine and natural infection induce neutralising anti-G antibodies. Ensure you provide the vaccination status of the horse and if vaccinated also provide the microchip details when requesting diagnostic or health testing for HeV. This will allow for the most appropriate serological tests to be undertaken in a timely manner.

9.1 Preferred samples

Take stringent precautions for yourself and anyone assisting you while conducting any sampling on possible HeV cases. Only take samples if your risk of exposure and that of others, such as anyone assisting you, can be adequately managed.

Samples may need to be taken from both live and dead horses, and therefore veterinary assessment of the risks associated with taking samples is required. Veterinary practitioners need to use their professional judgment as to what is safe and appropriate in each situation and proceed accordingly.

Dead horses can be sampled adequately for HeV testing without conducting a complete necropsy. Necropsy on a recently dead HeV infected horse is a very high risk activity because the horse is likely to be maximally infectious at this time, and should only be undertaken by a person who is suitably experienced and knowledgeable about how to manage exposure to HeV and how to use the relevant PPE.

Consider taking duplicate samples to allow further diagnostic work if the samples are HeV negative.

Biosecurity Queensland recommends that multiple sites be sampled from each horse as this will raise the diagnostic sensitivity of the sampling procedure as a whole.

A wide range of relevant samples will:

- Increase the overall diagnostic sensitivity, particularly if virus 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 humans and other susceptible animals.
- Increase the confidence in a negative HeV diagnosis.

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 filled to minimise the risk of a high anticoagulant concentration interfering with testing.
- Swabs—nasal, oral and rectal swabs may be used for PCR testing and virus isolation. Nasal swabs may detect infection at an earlier stage of infection than blood or other clinical samples (e.g. 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) – allows serological testing to be undertaken or additional diagnostic testing.

Swabs should not be transported dry and preferably be transported in liquid virus transport medium (VTM). Saline can be used if VTM is not available.

Lithium–heparin (LiHep) blood samples are no longer preferred. These samples provide no test 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, nasal, oral and rectal swabs should be sufficient for detection of HeV infection in a very high proportion of HeV-infected horses.

Other additional 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 (vaginal, urethral)	✓	✓
	Cut surface of mandibular lymph node		✓
Tissue [^]	Whole or part of mandibular lymph node		✓
	Other fresh or fixed samples		✓
	Blood clot obtained by jugular cut down		✓

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

9.2 Sample packing and dispatch

There are packaging requirements that must be met to transport biological samples by road and rail (surface) or air. Requirements apply to all samples sent by private veterinary practitioners to a laboratory. The principle of safe transport of biological samples by surface or air methods is the same-packaged material should not have any possibility of escaping from the package under normal conditions of transport.

Information regarding classification, packaging, labelling and documentation can be found in the *Requirements for the packaging and transport of pathology specimens and associated materials* produced by the National Pathology Accreditation Advisory Council or other relevant International Air Transport Association (IATA) references.

The latest edition can be found at www.health.gov.au

All people responsible for the packaging and transport of biological samples must be appropriately trained and their competency assessed for the task that they are to perform. Training from an accredited provider is required for the packing of some categories of samples.

Some courier companies will provide a complete service (i.e. they will come to a veterinary practice, pack and dispatch the samples correctly).

It is recommended that veterinary practices establish an account with the courier service of their choice so that samples for HeV testing can be dispatched for testing without delay.

A previously established account is usually required for a courier service to make a collection. The Queensland Government does not pay for the transport of samples to veterinary laboratories for disease investigation.

Steps to consider to correctly transport biological materials include:

- classification
- packaging
- labelling
- documentation.

For transport of all pathology samples and associated material by air or surface transport methods, the packaging must consist of three components (this is known as triple packaging):

- primary receptacle
- secondary packaging
- outer packaging.

Biological hazard classification

It is important to determine the classification of samples collected from horses potentially infected with HeV. The classification needs to be established by the veterinarian collecting the samples, based on their professional assessment of factors such as the known history, clinical presentation, endemic local conditions and professional judgment concerning individual circumstances associated with the case.

Classification of samples collected for HeV testing may include:

- Category A infectious substances
- Category B infectious substances
- Exempt or Category C.

Infectious substances

Infectious substances are those substances which are known to contain, or are reasonably expected to contain, pathogens. Pathogens are defined as micro-organisms (including bacteria, viruses, rickettsiae, parasites, fungi) and other agents such as prions, which can cause disease in humans or animals.

Category A infectious substances

A Category A substance is an infectious substance in a form that, when exposure to it occurs, is capable of causing permanent disability, or a life-threatening or fatal disease in otherwise healthy humans or animals.

Category A infectious substances should be triple packaged and compliant with IATA Packing Instructions 602 (including labelling and documentation). A shipper's declaration for dangerous goods must be completed with Packing Instruction 602. Refer to IATA references for the latest guidance regarding packing, labelling and documentation.

Note: if there is any doubt as to whether or not a substance meets the criteria, it must be included as Category A.

HeV is listed as a Category A infectious substance.

Category B infectious substances

A Category B substance is an infectious substance that does not meet the criteria for inclusion in Category A.

Category B infectious substances must be triple packaged and compliant with IATA Packing Instructions 650 (including labelling and documentation). Refer to IATA references for the latest guidance regarding packing, labelling and documentation.

Exempt or Category C

An Exempt or Category C substance is a patient sample for which there is minimal likelihood that pathogens are present. This category is not subject to IATA Dangerous Goods Regulations if the sample is transported in triple packaging that prevents leakage and is marked with the words 'Exempt animal specimens', as appropriate.

Note: if HeV is known or reasonably expected to be present the specimens should be classified as Category A infectious substances.

The attending veterinarian needs to determine the biological hazard classification of specimens collected (Category A or Category B or other), based on their professional assessment of factors such as the known history, clinical presentation, endemic local conditions and professional judgment concerning individual circumstances associated with the case.

Notes on dispatch of samples:

- The veterinarian collecting the samples needs to determine the biological hazard classification of samples and adhere to the associated packing, labelling and documentation requirements.
- Dispatch the samples to a government veterinary laboratory in the shortest time possible. Private veterinary laboratories do not conduct HeV testing. Direct submission to the Biosecurity Queensland laboratory at Coopers Plains will achieve the shortest turnaround times.
- Call ahead to notify the laboratory that samples for HeV testing are coming.
- The Queensland Government is not responsible for the dispatch and transport of samples to the laboratory. It is recommended that veterinary practices establish an account with the courier service of their choice so that samples for HeV testing can be dispatched for testing without delay.
- Fill out a specimen advice sheet (SAS) with all details, including the horse's **Hendra virus vaccination status**, property identification code (PIC) and a thorough history. Place the SAS outside the sample package so it can be read before the package is opened.
- Ensure a contact number is on the SAS so the duty pathologist can advise you of the test results.
- A copy of the SAS is available from the Biosecurity Queensland website, www.biosecurity.qld.gov.au. The form can be completed electronically by searching for 'GEN-008 Specimen advice sheet'.
- Clearly write HENDRA VIRUS TESTING on the SAS.
- Note that private laboratories will forward samples to a government laboratory if necessary; however, this will add more time until results are available and may incur additional costs.
- If within easy driving distance of a government veterinary laboratory, an option is to pack the samples appropriately, ensure they are appropriately secured in the vehicle and drive them to the laboratory. Before departure, notify the laboratory that these samples will be arriving.
- Before enlisting a client to deliver samples to a laboratory, veterinarians should check that the stress of a suspected HeV case is not affecting their client's capability to drive safely.
- Samples should be kept refrigerated, NOT frozen. It is important to ensure the cold chain is maintained during transport to preserve the integrity of the samples.

Note: A government laboratory will provide diagnostic HeV testing free of charge but this is unlikely to identify a cause if samples are HeV negative. Consider collecting duplicate samples to allow further diagnostic work.

9.3 Laboratory testing

HeV can be detected through laboratory testing by virus isolation or polymerase chain reaction. Seroconversion to HeV in response to infection or vaccination can be detected by serology (ELISA or VNT).

Polymerase chain reaction (PCR) test:

- Conducted on blood, swabs and tissue samples.
- Detects the direct presence of genetic material (virus) in a sample.
- Can detect live and dead virus but cannot differentiate between the two.
- Most useful early in the clinical course of disease.
- Typically one round of testing is run on each weekday at Biosecurity Queensland's laboratory at Coopers Plains. This is usually commenced at 2 pm.
- More frequent testing rounds may be conducted in response to increased demand.
- Results are usually reported to the submitter within one to two working days of receiving the samples at the Coopers Plains laboratory.

Interpretation of PCR results:

- A positive result indicates the presence of viral genome in the sample. It does not indicate whether the virus is viable and infectious.
- A positive PCR 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 (e.g. within the central nervous system).
- A negative PCR 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 HeV (resulting in a low concentration of virus, or absence of virus at sampled sites) or may have recovered from infection.

Enzyme linked immunosorbent assay (ELISA):

- Conducted on serum samples.
- Detects the presence of antibodies.
- Regarded as a screening test.

Interpretation of ELISA results:

- A negative ELISA result is a reliable indicator that a horse has not been previously infected with HeV or have a current vaccination status.
- As a screening test, non-negative results, including nonspecific reactor results require further testing to clarify the result. Use of VNT or re-sampling and ELISA are recommended.
- A positive result should be returned for infected and vaccinated horses as vaccine will induce the production of neutralising anti-G antibodies. Differentiating infected from vaccinated animals (DIVA) testing is required to differentiate infected from vaccinated animals.
- Since the ELISA is a screening test, non-negative results must be followed by a positive VNT before the animal could be considered seropositive to HeV.

Virus neutralisation test (VNT)

- Detects the presence of HeV antibody in a sample.
- The test involves mixing the blood sample with live virus to see if virus is neutralised by antibodies present in the sample. This test must be conducted under high-level biocontainment and is undertaken at AAHL.
- VNT are run on samples that return non-negative results on ELISA. This test may take 7–10 days to complete.
- In unvaccinated animals this test remains the gold standard for detection of an antibody response to HeV infection. It will also return a positive result with vaccinated horse.

Interpretation of VNT results:

- A positive VNT indicates the animal is seropositive to HeV either from infection or vaccination.
- A negative VNT means 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.

9.3.1 Hendra virus testing with vaccinated horses

Both vaccination with sG recombinant subunit and natural infection induce neutralising anti-G antibodies that are detected by all serological assays in use prior to release of the HeV vaccine in November 2012. None of the standard assays differentiates between the anti-G antibodies produced in response to infection and those produced in response to vaccination.

HeV DIVA test

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. AAHL have 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.

Hendra virus is an enveloped virus that contains a range of immunogenic proteins:

- Nucleocapsid protein (N)
- Phosphoprotein (P)
- Matrix protein (M)
- Fusion glycoprotein (F)
- Attachment glycoprotein (G)
- Large polymerase protein (L protein).

The G-glycoprotein is the most highly immunogenic viral protein and induces virus neutralising antibodies. The sG glycoprotein is the only HeV antigen used in the current 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 (N, P, M, F or L) is specific to infected animals.

Since there is no unique antigenic marker in the HeV vaccine, there is no potential for a single vaccine-specific test.

Interpretation of a panel of tests that detect the G glycoprotein or other HeV proteins (HeV DIVA tests) will enable differentiation of:

- serologically negative animals
- serologically positive animals due to vaccination
- serologically positive animals due to infection.

9.3.2 Urgency of Hendra virus testing for horses

Urgent testing for HeV may be undertaken under certain circumstances.

If urgent testing is requested during business hours, veterinarians should contact a Biosecurity Queensland duty pathologist to discuss the situation (see section 13 'List of contacts').

If urgent testing is requested outside of normal business hours, veterinarians should contact the Emergency Animal Disease Watch Hotline 1800 675 888 and discuss the situation with the Biosecurity Queensland veterinary officer.

The decision to conduct urgent HeV testing is made following discussion between the submitter and relevant Biosecurity Queensland staff. This may include the Biosecurity Queensland veterinary officer and the on-duty pathologist.

Requests for urgent testing will be assessed on a case-by-case basis. Reasons that may necessitate urgent testing for HeV include:

- Significant human exposure to body fluids of suspect horse/s.
- The exposure of large numbers of animals to body fluids of suspect horse/s.

10. Biosecurity advice to clients

If HeV is suspected in a client's horse it is important that sound biosecurity advice is provided to ensure the health, safety and welfare of people and other animals associated with the horse under investigation.

It is important to advise the client of the zoonotic potential of HeV and the steps they can take to manage the risk of exposure to themselves and others.

If HeV is highly suspected and potential human exposure has already occurred, clients should contact their general practitioner, local hospital emergency department or local public health unit.

Biosecurity Queensland will contact Queensland Health whenever HeV is confirmed or highly suspected. Queensland Health will make an assessment as to whether any people require monitoring and/or medical assistance. To make this assessment, Queensland Health will work with the veterinarian and the horse owner to identify the people they may need to contact.

In addition to the advice in section 7 'Workplace health and safety precautions' advice to clients can include the following:

- Isolate the sick or dead horse from all people, all other horses and all other domestic animals on the premises and from public access areas where possible.
- Advise neighbours that a horse on your property is being investigated for a HeV infection.
- If clients must have close contact with a sick horse where HeV has not been ruled out, you should provide advice on appropriate infection control (including PPE) such as:
 - People should cover cuts and abrasions with a water-resistant dressing.
 - Put on appropriate PPE before approaching the horse.
 - After handling the horse, remove and dispose of PPE carefully, making sure there is no contact with the facial area, particularly eyes, mouth and nose.
 - Wash hands with soap and water and dry them or use hand wipes and waterless alcohol-based hand rub immediately after removing PPE.
 - Carefully remove any clothing contaminated with a sick horse's body fluids.
- If clients have handled a sick horse, they should follow these steps before having contact with other horses:
 - Wash off any contamination with plenty of soap and water.
 - Shower and wash hair.
 - Change clothes and footwear.
- Arrange activities so that unaffected horses are handled first and have contact with the sick horses last.
- Advise the client to stop or limit horse movements on and off the property.
- Advise the client to stop or limit movements of horse products (such as manure) and equipment (such as tack, dental equipment) off the property.
- Advise the client to stop or limit visiting horse practitioners (such as farriers or equine dentists) and advise visiting horse practitioners who must have close contact with the horse about the potential HeV risk.
- When disposing of a dead horse the disposal contractor should be informed that the horse has been tested for, or is suspected of being infected with HeV. Appropriate precautions should be taken.

- People should seek medical advice if at all concerned about possible exposure to HeV.

Useful references are available at www.biosecurity.qld.gov.au including the fact sheets *Hendra virus information for horse owners, handlers, competitors and event organisers*. Relevant fact sheets are also available from the Workplace Health and Safety Queensland website at www.worksafe.qld.gov.au

11. Carcass disposal

If a carcass is held until results of a HeV investigation are known, the owner or person in charge remains responsible for the disposal. The owner or person in charge can dispose of the carcass using their normal methods if HeV is not diagnosed or not suspected.

Biosecurity Queensland will provide advice on the disposal of a carcass that has been confirmed to be infected with HeV. Biosecurity Queensland consults with the Department of Environment and Heritage Protection for disposal advice on a confirmed HeV carcass. It is important to ensure any potentially infectious animal does not pose a risk of infection to people or other animals and does not cause environmental contamination.

Where HeV is suspected or confirmed, care will be required in the disposal of the carcass. The following information may assist this process:

- Isolate the carcass from all people and other animals until a disposal method is finalised and can be undertaken safely.
- Disposal options include deep burial on property, burning on property or transport off the property (e.g. disposal at a landfill site).
- Deep burial on the property is the option of choice, and preferably where the carcass is lying so the carcass does not have to be moved.
- Burning on property would mainly be by pyre burning (i.e. the carcass is put on a fire built above ground).
- Dispose of the intact carcass (do not dismember).
- Transport off site for disposal will require prior planning and coordination.

Note: Costs associated with the disposal of horses are considered as costs to be covered by the owner of the horse.

The Workplace Health and Safety Queensland fact sheet *Hendra virus – information for businesses that dispose of horse carcasses* is a valuable reference regarding workplace health and safety considerations for the disposal of horses that may be infected with HeV and is available at www.worksafe.qld.gov.au

11.1 Steps common to all methods of carcass disposal

- People physically handling the carcass of a horse suspected or known to have HeV during the disposal process should wear appropriate PPE.
- Machinery operators should wear sufficient PPE to protect themselves against possible exposure (e.g. splashes of body fluids and from aerosols if generated).
- If machinery is used, the machinery operator should outline the workplace health and safety requirements with respect to the use of the machinery to all people on site.
- Treat all body fluids and excreta with caution.

- Any ground area where body fluids have spilled can be disinfected by wetting the soil thoroughly to an area and depth equal to that of the spillage. Alternatively, the layer of contaminated soil can be removed and placed in the burial pit, on the pyre or sent with the carcass off site.
- If the carcass has to be moved, spillage of body fluids or excreta will need to be managed. Enclose the head in a strong plastic bag and tie this off around the neck to help contain fluids. After the carcass is moved, all spillage should be dealt with through disinfection or removed and disposed of as above.
- Any part of machinery and equipment that comes in direct contact with the carcass or with body fluids or excreta should be cleaned and disinfected.
- When cleaning and disinfecting, avoid creating splashes and aerosols for personal safety (do not use high-pressure hoses).
- Suitable disinfectants include soaps and detergents, Virkon®, hypochlorites, iodophors/iodine, biguanidines (e.g. chlorhexidine) and quaternary ammonium compounds (see section 8.5 'Disinfectants'). Note that many disinfectants have reduced activity in the presence of organic matter.
- Perform hand hygiene after carcass disposal.
- All people should disinfect off through the property's entry/exit point.
- If you are unsure about the disposal process, isolate the carcass and contact Biosecurity Queensland on 13 25 23 or, if necessary, call the Emergency Animal Disease Watch Hotline on 1800 675 888.

11.2 Disposal by burial on site

- Ensure burial onsite is consistent with local government (Council) and Department of Environment and Heritage Protection regulations.
- Do not bury next to a water course where flooding could expose the carcass.
- A trench is easier to work with as the carcass can be pushed in over the side, especially if the trench is dug next to the carcass.
- The machinery operator may be able to provide advice on which is preferable for the location—a trench or hole.
- The aim is to bury with a minimum covering of soil over the carcass of two metres. This contains fluids and odours and also prevents animals scavenging on the carcass.

Note: *Take normal precautions for animals destroyed with barbiturate solutions to minimise the risk of animals scavenging on the carcass.*

11.3 Disposal by burning

- Pyre burning may require a permit issued by the local fire authority. A permit will be refused if the fire presents a safety concern to the community.
- Pyre burning is probably unsuitable for smaller properties due to community concerns, lack of fuel and possible fire danger to other properties.

- A large amount of fuel will be needed. The *AUSVETPLAN Operational Procedures Manual— Disposal* states: ‘Carcasses can be completely consumed using dry wood alone at the rate of 1.5 tonnes for a 500 kilogram adult bovine or 1.5 tonnes of coal briquettes or equivalent combinations. For multiple carcasses, the amount of fuel may be reduced to 1.0 tonnes per adult bovine because of economies of scale. Straw and liquid fuels are required to start the burn. The same rule of thumb could be applied to horses.
- For best efficiency when building a pyre, it is essential to build air channels at the bottom of the pyre to let air circulate into the bottom of the fire.

11.4 Transport off site for disposal

- Planning and co-ordination is required.
- Carcasses may be regarded as clinical waste which is a regulated waste as defined in the *Environmental Protection Regulation 2008*. The disposal of regulated waste other than at the place of generation (i.e. off site) is a notifiable activity which must be reported to the Department of Environment and Heritage Protection.
- Make contact with the receiving entity to ensure they agree to receive the carcass and dispose of it.
- Arrange bio-secure transport (transport that will not leak body fluids or expose people or horses to the carcass).
- Arrange machinery to load the carcass into this transport.
- Manage body fluids.
- Ensure management of the carcass at the receiving end can be done safely.
- The most likely destination will be a registered landfill.

See chapter 3 of the *AUSVETPLAN Operational Procedures Manual—Disposal* for further information.

12. Downtime

Personal downtime refers to the time that should be spent away from other susceptible animals to prevent possible spread of HeV. With HeV this refers to the time taken to complete a series of actions more than a definitive period of time. It is recommended that there is no close contact with people or susceptible animals until the following actions are completed:

- Wash exposed areas of skin thoroughly with soap and water.
- Remove any contaminated clothes and double bag. Wash when possible in a separate hot wash cycle with detergent (avoid contamination when doing this). Do not wash potentially contaminated clothing with other household laundry.
- Take a hot shower using shampoo and soap.
- Dress in clean clothes.
- Put on clean footwear or footwear not worn in the dirty area.
- If HeV is confirmed or highly suspected and potential human exposure has already occurred, seek medical advice and also contact the local Queensland Health Population Health Unit.
- Once this has been completed, normal work can be resumed.

If appropriate infection control procedures have been followed during the investigation of a suspected HeV case, including PPE and no breaches/exposure/contamination has occurred no downtime is required.

Equipment downtime refers to the time taken to ensure any equipment used or taken into the dirty area is safe and does not pose a risk of disease transmission. Where possible, use disposable equipment in the dirty area. This equipment can be double-bagged or otherwise sealed safely and held until results are known. If negative, treat as per the practice waste management protocol.

If positive the disposable equipment would be classified as clinical waste and should be managed appropriately. More information on this can be found in the information sheets *Waste management – Defining Clinical Waste* and *Waste management—Clinical or related waste treatment and disposal* on the Department of Environment and Heritage Protection website, www.ehp.qld.gov.au

All sharps should be treated as clinical waste and disposed of appropriately in a container that complies with the relevant Australian Standards.

Equipment that will be reused on other animals must be decontaminated. Circumstantial evidence indicates that equipment can act as a fomite and transfer HeV.

- Most reusable equipment should have been decontaminated during exit from the dirty area. Double check for any remaining organic material and repeat the decontamination process if present.
- Wear PPE when decontaminating equipment, avoid generating aerosols and splashes and wash hands after removing PPE.
- Guidelines from infection control practices in human medicine require equipment to be cleaned with detergent and water prior to soaking in a disinfectant (i.e. the equipment is not to be scrubbed or dipped in the disinfectant without prior cleaning). The rationale is that without prior cleaning, the disinfectant may be unable to penetrate through any organic matter to adequately disinfect the surface. Also, some disinfectants (e.g. sodium hypochlorite) are less effective in the presence of organic matter or ‘fix’ the organic matter onto the surface (e.g. aldehydes).
- Standards for cleaning, disinfecting and sterilising equipment are set down in Australian Standards.
- Use disinfectants safely in accordance with the product’s safety data sheet.
- Equipment that cannot be properly decontaminated on the premises by cleaning and disinfecting should be taken off the premises double-bagged or otherwise sealed. If possible, leave equipment sealed until results are known. If negative, clean as per practice routine.
- If positive, or if equipment cannot be isolated for this long, then the person cleaning should don appropriate PPE, remove equipment from its sealed container, clean with detergent and water and complete the process with a recommended disinfectant.
- Note that long-term contact with some of the recommended disinfectants could cause harm to the equipment. Leave in contact with the disinfectant for the recommended time as per label directions and then wash off.
- Check your vehicle for degree of exposure to contaminated items and decontaminate using a recommended disinfectant. Do not forget the steering wheel, door handles, and floor mat.
- Seek advice from Biosecurity Queensland as required.
- When equipment has completed this process, it can be reused

13. List of contacts

It is recommended that each practice develop a list of key contacts that may be of assistance in the management of potential HeV infection in horses.

Biosecurity Queensland

Phone: 13 25 23 (business hours)
Emergency Animal Disease Watch Hotline
1800 675 888 (anytime)
Website: www.biosecurity.qld.gov.au

Coopers Plains Laboratory

Health and Food Sciences Precinct Specimen Receipt (Loading Block 12)
39 Kessels Road, Coopers Plains Qld 4108
Phone: (07) 3276 6062
Email: bslclo@daff.qld.gov.au

Queensland Health

Phone: 13 HEALTH (13 43 25 84)
Website: www.health.qld.gov.au
Queensland Health public health unit contact details are available at:
<http://www.health.qld.gov.au/cdcg/contacts.asp>

Workplace Health and Safety Queensland

Phone: 1300 369 915
Website: www.worksafe.qld.gov.au

Department of Environment and Heritage Protection

Phone: 13 QGOV (13 74 68)
Website: www.ehp.qld.gov.au

14. Media and confidentiality

14.1 Media

It is important that any cases of HeV infection are openly and accurately communicated to the public.

Media enquiries can be directed to the Department of Agriculture, Fisheries and Forestry's media unit on 13 25 23. This allows a consistent approach to be taken by people who have the best access to all information about the overall situation and relieves owners and veterinary practitioners of the pressure of media attention.

HeV incidents typically generate media interest. Involved veterinarians may be asked for public comment. The investigating veterinarian has the right to refuse to comment. If they do choose to comment, it is recommended that only facts are presented, that they are not drawn into conjecture and that they comment only on the part of the operation they were directly involved in.

The AVA and Biosecurity Queensland are able to provide advice to private practitioners about handling the media.

14.2 Confidentiality

Normal confidentiality provisions apply in the management of HeV incidents. HeV incidents can generate considerable media interest and any statement made should respect the normal confidentiality provisions expected by clients, patients, staff and others involved.

15. Closing the case

Closing any case where a veterinarian has elected to test a horse for HeV will require interpretation of laboratory results (if available) and exercising professional judgement.

Negative laboratory results alone may not be sufficient grounds to close the case. Interpretation of the results, in combination with the history and present situation, will most likely be required when making the assessment to close a case.

Approximately 20 per cent of horses may survive the acute phase of HeV infection. Paired serological testing taken 10–14 days apart may provide evidence of exposure to HeV or rule out infection.

Conclusive diagnosis of another cause of the illness can close the case.

Related reference documents

Legislation www.legislation.qld.gov.au	<i>Exotic Diseases in Animals Act 1981</i>
	<i>Veterinary Surgeons Act 1936</i>
	Work Health and Safety Regulation 2011
	Environmental Protection Regulation 2008
AUSVETPLAN www.animalhealthaustralia.com.au	Decontamination manual
	Hendra virus response policy brief
	Disposal procedures manual
Biosecurity Queensland www.biosecurity.qld.gov.au	Hendra virus information for horse owners
	Hendra virus—veterinary practice pack
Workplace Health and Safety Queensland www.worksafe.qld.gov.au	Hendra virus: information for horse properties and other horse related businesses
	Hendra virus: information for veterinarians
	Hendra virus—information for businesses that dispose of horse carcasses
Queensland Health www.health.qld.gov.au	Hendra virus infection
	Hendra Virus Infection for Healthcare Workers
	Hendra virus infection prevention advice

Appendix 1- Equipment required to collect samples for HeV testing

1. Sampling requirements

- Determine how many animals are to be sampled and what samples should be safe to collect. If you are sampling animals other than horses, contact Biosecurity Queensland for sample requirements
- Determine individual identification of all animals to be sampled.
- Try to collect more than one sample from each animal (see section 9.1 ‘Preferred samples’).
- Label all sample containers uniquely before entry to the hot area.

Equipment required to collect samples from each live horse:

Checklist	Equipment
<input type="checkbox"/>	Shielded vacutainer needle and holder (plus spares)
<input type="checkbox"/>	2 x 10 mL EDTA vacutainer (plus spares)
<input type="checkbox"/>	2 x 10 mL serum vacutainer (plus spares)
<input type="checkbox"/>	4 x liquid virus transport media (plus spares)—if VTM is not available, place swabs in saline
<input type="checkbox"/>	4 x swabs (plus spares)

Additional equipment required to collect samples from each dead horse:

Checklist	Equipment
<input type="checkbox"/>	Scalpel, scissors, forceps if collecting tissue samples
<input type="checkbox"/>	Sample jars for fresh and formalised tissue samples
<input type="checkbox"/>	Cut-resistant gloves (e.g. Kevlar®) may be considered.

Note: AAHL have indicated that the clot from the jugular vein (taken following cut-down onto the vein) and submandibular lymph node tissue can increase overall test sensitivity when combined with swabs. Collection of these samples requires limited necropsy work and should only be taken if the associated risks can be managed—including any blood released during the procedure.

Additional equipment for sampling live and dead animals:

Checklist	Equipment
<input type="checkbox"/>	Small (but adequate) sharps container with built-in needle removal facility
<input type="checkbox"/>	2 x A4 zip lock bags to remove samples off premises
<input type="checkbox"/>	Plastic bucket for carrying equipment

2. Personal protective equipment (PPE) per person

Checklist	Equipment
<input type="checkbox"/>	1 x pair of disposable overalls (at least splash resistant rating) plus spares.
<input type="checkbox"/>	1 x P2 disposable facepiece respirator (plus spares), or reusable negative pressure respirator/s or PAPR/s and filters
<input type="checkbox"/>	Pack of disposable gloves (nitrile gloves recommended)
<input type="checkbox"/>	Safety eyewear and/or a face shield
<input type="checkbox"/>	1 x roll of duct tape
<input type="checkbox"/>	1 x pair of impervious rubber boots or boot covers
<input type="checkbox"/>	1 x disposable or washable hat or cap if overalls do not have a hood
<input type="checkbox"/>	Cut-resistant gloves (e.g. Kevlar®) may be considered.

People in close proximity must wear the same PPE as the veterinarian.

3. Disinfection and waste disposal equipment required (suggested minimum)

Checklist	Equipment
<input type="checkbox"/>	Foot bath and 2–3 buckets
<input type="checkbox"/>	Scrubbing brush
<input type="checkbox"/>	Hoof pick or medium screwdriver
<input type="checkbox"/>	Adequate water (20L minimum)
<input type="checkbox"/>	Hand drying facilities (disposable paper towel etc.)
<input type="checkbox"/>	Soap or detergent for personal decontamination
<input type="checkbox"/>	Virkon® or supplies of other chosen disinfectant (hypochlorite, iodophor/iodine, biguanidine, quaternary ammonium compound)
<input type="checkbox"/>	Small hand-sprayer for chosen disinfectant
	If using reusable respirators a small hand-sprayer with suitable disinfectant (e.g. Trigen®) for the respirator (filters should be disposed of)
<input type="checkbox"/>	1 x heavy duty garbage bag
<input type="checkbox"/>	1 x clinical waste bag
<input type="checkbox"/>	2 x zip/cable ties
<input type="checkbox"/>	Ground sheet or plastic mat (no more than 1 m ²).

4. Communications

- Determine the communication system that will be used on site (e.g. mobile phone in zip lock/clip seal bag, landline).
- Ensure any communication item is able to be decontaminated off the property. For example, seal items in plastic bags or similar so they can be used without having to take them out of the bag, then decontaminate the outside of the bag.

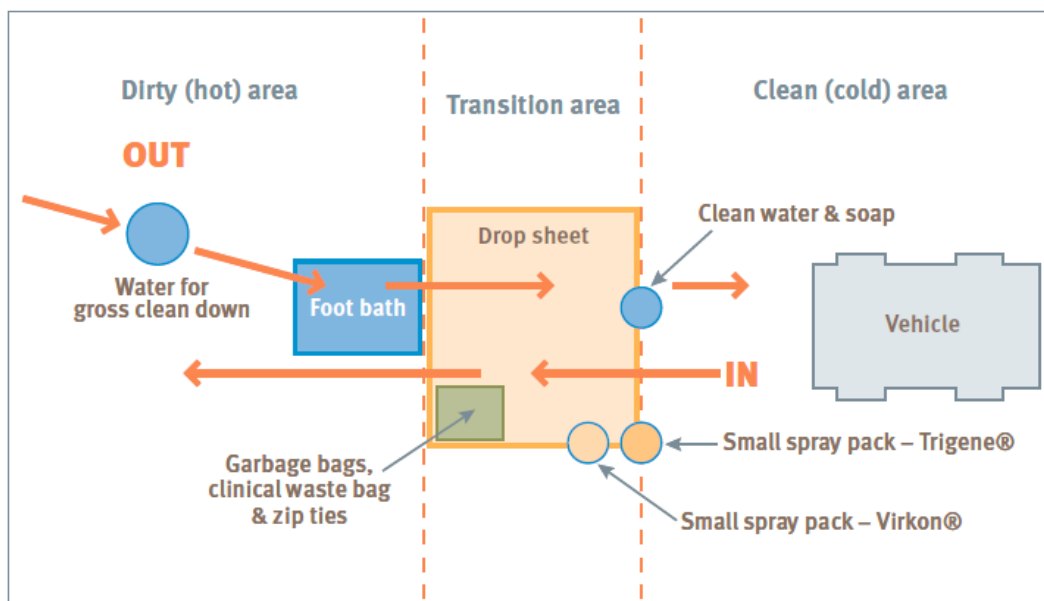
Appendix 2—Entry/exit process

1. Set up an entry/exit site

At the selected entry/exit point, identify a ‘clean’ (cold) area, a ‘dirty’ (hot) area (i.e. the contaminated area where the possible case is situated) and a small transition area between the clean/cold and dirty/hot areas (see Figure 1).

On the clean side, lay out all equipment required for the investigation and, before donning PPE, double check that nothing has been missed and that no unnecessary equipment is being taken into the dirty area. Leave vehicles in the clean area. If vehicles are taken into the dirty area, they will need to be decontaminated.

Figure 1. Suitable entry/exit decontamination site



2. Entering the hot area

Make sure containers of disinfectant, along with soap and clean water, are available and placed at the entry/exit point for use during exit.

Don PPE in the following sequence to assist best personal protection:

1. Wash hands with soap/detergent and dry hands.
2. Don overalls then boots (overall legs go outside boots).
3. Don first pair of gloves*.
4. Don respirator and perform fit check of the respirator.
5. Don safety eye wear.
6. Pull overalls hood up if present and zip to chin.
7. Perform respirator fit check.
8. Double-glove*.

* Secure one set of gloves onto the sleeves of the overalls with tape. It is personal preference as to whether the inner or outer pair of gloves are taped to the overall sleeves.

3. If using a powered air purifying respirator (PAPR):

- Wash hands with soap/detergent and dry hands.
- Don overalls then boots (overall legs go outside boots).
- Pull overalls hood up if present and zip to chin.
- Don PAPR then gloves.
- Double-glove*

**Secure one set of gloves onto the sleeves of the overalls with tape. It is personal preference as to whether the inner or outer pair of gloves are taped to the overall sleeves.*

4. Enter the hot area

- You should enter the hot area only after fully dressed in PPE and with all required equipment.
- Any person assisting or in close proximity must wear the same standard of PPE.

5. Undertake the required sampling

- Make sure that samples are uniquely and clearly labelled.
- Do not place yourself or assistants at risk of injury at any time.
- Use techniques that minimise the chance of contamination of people and their PPE.
- Undertake safe sharps handling and disposal of waste to prevent accidental exposure via needle stick injury (i.e. do not re-cap needles, use sharps container).

6. When sampling is completed

- Place labelled samples in a clip seal bag for removal.

7. Exiting the hot area

- Remove gross contamination from self and equipment. Do this before reaching the entry/exit point to minimise the risk of spreading contamination beyond the designated hot/dirty area.
- Use a brush and soap or detergent and water.
- Clean the treads of the boots (e.g. at a tap on site or a bucket strategically placed back from the entry/exit site).
- Go to the hot/dirty side of the entry/exit point.
- Double-bag the samples in clip seal bags and disinfect them to the clean side. Be careful not to contaminate the samples with disinfectant.
- Spray disinfectant on the outer gloves.

8. Removal (doffing) of PPE

If non-disposable PPE cannot be adequately decontaminated on site, double-bag it and remove it for later attention—this is not a preferred option.

Handle used PPE with care to avoid dispersal of contaminants.

To remove PPE where a P2 disposable facepiece respirator or reusable half-face or full-face respirator is used:

1. Remove the outer pair of gloves to garbage bag.
2. Wash hands, still encased in the inner pair of gloves, in disinfectant.
3. Peel disposable overalls down and over boots
 - Step out of boots and onto plastic drop sheet (transition area)
 - Place disposable overalls in garbage bag.
4. Remove hat/cap to garbage bag or soak in disinfectant, double bag and remove for laundering.
5. Remove and disinfect safety eyewear, carefully avoiding splashes.
6. Remove respirator (disposable respirators to garbage bag or mist/wipe reusable respirators with disinfectant solution). Do not touch the front of the respirator; handle by the straps.
7. Disinfect boots and place in clean/cold area.
8. Tie off the garbage bag and then:
 - Disinfect it.
 - Double bag it in a biological waste bag and tie off and place in clean/cold area.
9. Disinfect yourself:
 - Disinfect and scrub all potentially contaminated areas and exposed skin with an approved disinfectant.
 - Put on clean shoes.
 - Do not walk back over contaminated ground.
10. Carefully, without contaminating your clean clothes, disinfect/rinse the drop sheet and buckets/containers and brushes in the foot bath, ensuring they are free of contamination and then place them in the clean/cold area.
 - Take off the 2nd (inner) pair of gloves and put them in a biological waste bag and tie off. Double bag.
 - Tip out the tub/footbath with the disinfectant and spray undersides with disinfectant spray.
 - Pack buckets, brushes, drop sheet in footbath.
 - Apply a final spray with disinfectant from the spray pack to all bags to leave the site.
11. Wash hands in clean water with disinfectant or use an alcohol-based hand rub.

Where the respirator is a powered air purifying respirator (PAPR):

1. Remove the outer pair of gloves to garbage bag.
2. Wash hands, still encased in the inner pair of gloves, in disinfectant.
3. Remove and disinfect the PAPR.
4. Peel disposable overalls down and over boots:
 - Step out of boots and onto plastic drop sheet (transition area)
 - Place disposable overalls in garbage bag
5. Then follow the above process from step 7 (above).

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