



Standard Operating Procedure

RAB011: Bait delivery of Rabbit Haemorrhagic Disease Virus (RHDV1) K5 strain

Prepared by Trudy Sharp and Glen Saunders

Background

Rabbit haemorrhagic disease (RHD) is used in Australia to minimise the impacts of the introduced European rabbit (*Oryctolagus cuniculus*) on agricultural production and the environment. Other available rabbit control methods include poisoning, warren destruction, surface harbour removal, shooting, trapping and exclusion fencing. Biological control with myxomatosis also occurs naturally but the disease is no longer deliberately released.

RHD is an acute, highly contagious disease that only infects wild and domestic European rabbits (*Oryctolagus cuniculus*). In most adult rabbits the disease progresses rapidly from fever and lethargy to sudden death within 48-72 hours of infection. The disease causes acute liver damage with resultant blood clotting abnormalities. Death occurs due to obstruction of blood supply in vital organs and/or internal haemorrhages. RHD has a high mortality rate, killing 70 to 90% of susceptible rabbits.

The disease is caused by the rabbit haemorrhagic disease virus 1 (RHDV1) (Czech 351 strain) and its release in Australia has controlled rabbit populations since 1996. Over time, rabbits have developed some genetic resistance to the Czech strain (as they did for myxomatosis) and its effectiveness as a biological control agent has been reduced. A new strain - RHDV1 (K5 strain) from Korea—has been evaluated and could improve the biological control of rabbits in Australia. In adult rabbits, infection with the K5 strain resulted in slightly increased mortality rates and requires over 30 times less virus to infect rabbits compared to the Czech strain. In addition, K5 appears able to overcome the partial protection offered by previous infection with another virus - the benign rabbit calicivirus RCV-A1, which particularly occurs in more temperate areas of Australia. More recently another variant of the disease - RHDV2 - was found to be circulating in wild rabbits. RHDV2

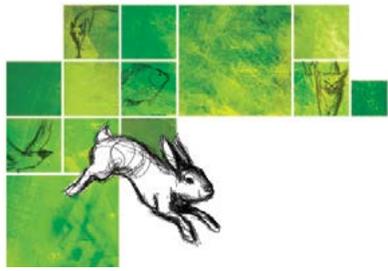
unlike RHDV1 also infects the European brown hare (*Lepus europaeus*). At this stage it is unknown how RHDV1 and RHDV2 will interact in wild rabbit populations. To maximise the impact of the disease, the deliberate release of RHDV1 into wild rabbit populations is used to initiate outbreaks. The virus can be delivered via small quantities of treated oat or carrot bait or by direct injection of the virus into captured rabbits. Procedures for the direct inoculation of the virus into rabbits are described in [RAB012 Inoculation of rabbits with rabbit haemorrhagic disease virus \(RHDV1\) K5 strain](#).

This standard operating procedure (SOP) is a guide only; it does not replace or override the legislation that applies in the relevant state or territory jurisdiction. The SOP should only be used subject to the applicable legal requirements (including OH&S) operating in the relevant jurisdiction.

Application

- Introduction or 'seeding' of RHD into rabbit populations using the K5 strain is used as part of an integrated approach to rabbit management and is not meant to be used as a stand-alone method.
- Where RHDV1 K5 is used as a control agent, follow-up control of remaining rabbits should be undertaken to ensure long-term effects.
- Release of RHDV1 K5 should take into account level of existing immunity, rabbit breeding patterns including presence of young rabbits, natural spread of the virus and the activity of insects, which can act as vectors.
- Do not release RHDV1 K5 into breeding populations i.e. when young rabbits are observed. Younger rabbits can become infected but are less likely to die because of RHD, particularly in the presence of maternal antibodies. These rabbits can become immune to future outbreaks of RHD.





RAB011: Bait delivery of RHDV1 K5

- Populations of rabbits visibly affected by myxomatosis should not be baited because they are less likely to succumb to RHD.
- Bait delivery of RHDV1 has been shown to be more effective in reducing rabbit numbers compared to the inoculation technique. This is thought to be due to the larger numbers of rabbits that are initially exposed to the virus. Also, bait delivery is not as labour intensive or costly as inoculation and allows managers to more effectively target populations when they are most susceptible.
- Bait delivery of RHDV1 K5 is also considered more humane than the inoculation method as rabbits do not need to be caught. Capture, handling and restraint are significant stressors for wild rabbits, which can lead to disruption of social structure, debilitation and sometimes death.
- Baiting of rabbits with RHDV1 K5 can only be carried out under conditions set down in a specific permit issued by the Australian Pesticides & Veterinary Medicines Authority (APVMA) under Commonwealth legislation (Agricultural and Veterinary Chemicals Code Act 1994). RHDV1 must also be used in accordance with relevant State, Territory and other Commonwealth legislation.
- Freeze-dried (also called lyophilised) RHDV1 K5 is a restricted chemical product (under Regulation 45 of the *Agricultural and Veterinary Chemicals Code Regulations 1995*). This listing requires special precautions in the preparation, handling, storage and use of RHDV1 treated baits, along with specific regulations regarding labelling or availability.
- Handling of RHDV1 K5 and preparation of bait must only be performed by authorised persons who have the appropriate training.
- Prepared bait can only be obtained through authorised government agencies.

Animal Welfare Considerations

Impact on target animals

- RHD is an acute, highly infectious usually fatal condition that affects domestic, farmed and wild rabbits of the species *Oryctolagus cuniculus*.
- In most rabbits, death from RHD is sudden. Some animals show no signs of illness prior to death whilst others will have elevated temperature, anorexia, apathy, dullness, prostration and reddened eyes. Respiratory signs (e.g. rapid respiration, bloody nasal discharge) and occasionally nervous signs (e.g. convulsions, paralysis, squealing) may be seen in the later stages. Five to 10% of rabbits may show a chronic or subclinical course of disease. These animals may have jaundice, weight loss and lethargy for up to 1 to 2 weeks before either dying or surviving with immunity.

Impact on non-target animals

- All rabbits in Australia are derived from the European rabbit (*Oryctolagus cuniculus*) and are therefore potentially susceptible to infection. Farmed and pet rabbits should be vaccinated against RHDV1 (which should provide protection against both the Czech and Korean K5 strains but not RHDV2). Vaccinations are available from veterinary practitioners. The Australian Veterinary Association (AVA) recommends that rabbits are vaccinated against RHDV1 at 10-12 weeks of age and then given an annual booster. During an outbreak, rabbits may be vaccinated earlier than 10 weeks, with a booster recommended 4 weeks later. Further detailed information on vaccination can be found on the AVA website: <http://www.ava.com.au/rabbit-calicivirus>
- There is no evidence that RHDV1 K5 causes infection in other species of native and domestic mammals and birds.

Health and Safety Considerations

- Operators using RHDV1 K5 must strictly follow the directions on the approved label when preparing for use, using, storing, transporting or disposing of the virus.
- RHDV1 K5 only affects European rabbits. However, due to the presence of rabbit and viral proteins in the product, it is possible that accidental administration of the product to an operator could be accompanied by an adverse allergic reaction. When mixing the virus in feed material it is advisable to wear gloves and a face shield to prevent contact of the virus with skin, eyes and mucous membranes.
- Appropriate personal protective equipment, including cotton overalls, water impermeable gloves and a face mask or safety glasses, should be worn when preparing and handling freeze dried RHDV1 K5 and treated bait.
- Thoroughly wash exposed skin with soap and water. Wash contaminated clothing and gloves.
- If poisoning occurs, contact a doctor or the Poisons Information Centre (Ph 13 11 26).
- For further information refer to the product insert available from the supplier.

Equipment Required

Treated baits

- Treated bait is prepared by the application of reconstituted freeze dried RHDV1 K5 to carrots or oats in an enclosed mixing device that meets the standard requirements for 1080 bait mixing such as a 'Moree Mixer'.
- Oats should be standard intact oats with husks attached and carrots should be good quality and freshly diced. One vial reconstituted product (10 mL) diluted with an additional 90 mL of water (100 mL total volume) is sufficient to treat 10kg of freshly diced carrots OR 5kg of oats.
- Prepare the viral solution as per the product label.
- Place the feed material in the mixer, add the prepared viral solution via a fine spray from the spray nozzles and then

- gently mix while tumbling in the sealed mixing device.
- Once mixed, transfer the treated bait material to appropriately labelled, sealable plastic containers or heavy duty plastic bags which have significant strength to prevent damage to bait or leakage.
- Store treated baits in a cool, shaded area until ready for use.
- Treated baits should be used within 24 hours of preparation.
- Equipment used to prepare treated baits should be decontaminated after use by rinsing with 0.5% sodium hypochlorite. After rinsing, wash thoroughly with excess water and allow equipment to dry.

Other Equipment

- Personal protective equipment (including water impermeable gloves, face shield, overalls, boots etc.)
- Towel, soap, dish or bucket
- First aid kit
- Plastic 'sharps' disposal container for disposal of needles and other sharps.
- Appropriately labelled and leakproof containers for storing treated bait - refer to permit for specifications.
- 0.5% sodium hypochlorite for decontamination of equipment etc. (e.g. a 1 in 20 dilution in water of household bleach solution containing 10% hypochlorite).
- Bait mixer
- Carrot cutter (if required)

Suppliers of RHDV1 K5:

Vials of freeze-dried RHDV1 K5 are manufactured and supplied by NSW Department of Primary Industries' (NSW DPI) Elizabeth Macarthur Agricultural Institute (EMAI).

Procedures

Always read relevant permit for conditions and directions for use.

Assessment of site and estimation of rabbit numbers

- Warrens, rabbit harbour and scratching and feeding areas should be located to ensure accurate placement of bait.
- The density of rabbits on the site should be estimated using spotlight counts and warren monitoring. The location and numbers of rabbits on neighbouring properties should also be approximated.
- Contact your local vertebrate pest control authority for more information and advice on site assessment and monitoring of rabbit numbers.

Free-feeding

- Perform free-feeding of at least 2 feeds with carrots or 3 feeds for oats. Free-feeding of non-treated bait is an essential step to allow rabbits to become accustomed

to eating bait material. It also enables an estimation of amount of treated bait required and assessment of any non-target uptake.

- The rate of bait for free-feeding should be appropriate to the local rabbit density, typically 2 to 5kg/km for oats and 15 to 20kg/km for carrots
- Distribute free-feeds by broadcast (carrots only) or by trailing (oats and carrots).

Placement of treated baits

- Broadcast the treated bait at approximately 10% of the rate applied in the final night of free-feeding.
- Lay the treated bait in or around a small number of warrens in the area targeted for a RHD outbreak. As with free-feeding, treated oats must be applied in trails but treated chopped carrot may be applied by trails or broadcast.
- If treating individual warrens, select 3-4 within targeted area and apply treated feed by hand around warren entrances. Apply a maximum of 5kg treated carrot or 2kg treated oats per warren.
- Do not apply treated bait to crops or in situations where livestock may have access to the bait.
- The treated baits should be laid as soon as possible after preparation and on the day prepared. It is preferable to lay baits in the evening as rabbits are active between dusk and dawn.
- Any treated bait that has not been consumed by the following morning should be recovered and destroyed by deep burial.
- Equipment used to prepare and distribute the treated feed should be decontaminated at the end of each day by rinsing with 0.5% sodium hypochlorite. After rinsing, wash thoroughly with excess water and allow equipment to dry.

Procedural Notes

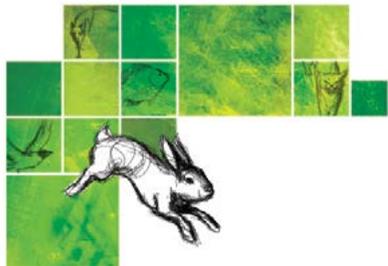
Read product label for more detailed information

Preparation and storage of RHVD1 K5 inoculum

- The viral suspension should be prepared and stored according to the directions on the label.
- The contents of the vial should be used within 24 hours of initial opening. Store open vial at 4-8°C.
- Unused vials of virus should be discarded into contaminated waste containers for appropriate disposal.

Disposal of contaminated waste

- Any needles /sharps should be immediately place in a designated and appropriately labelled 'sharps' container.
- Used vials and syringes should be soaked in 0.5% sodium hypochlorite prior to disposal by burial in a local authority landfill.



RAB011: Bait delivery of RHDV1 K5

- All bags used for storing bait must be destroyed by deep burial.
- Sharps containers should be conveyed to a disposal facility equipped for the disposal of biomedical waste. Contact your local waste reduction and disposal services for more information.

Further information

Contact the relevant federal, state or territory government agency from the following list of websites:

- Australian Department of the Environment and Energy
<http://www.environment.gov.au/>
- Australian Department of Agriculture and Water Resources
<http://www.agriculture.gov.au/>
- ACT Transport Canberra and City Services
<http://www.tccs.act.gov.au/city-living>
- NSW Department of Primary Industries
<http://www.dpi.nsw.gov.au>
- NT Department of Land Resource Management
<https://landresources.nt.gov.au/>
- QLD Department of Agriculture and Fisheries
<https://www.daf.qld.gov.au/>
- SA Department of Primary Industries and Regions
<http://www.pir.sa.gov.au/biosecurity>
- TAS Department of Primary Industries, Parks, Water and Environment
<http://dpiuwe.tas.gov.au/>
- VIC Department of Economic Development, Jobs, Transport and Resources
<http://economicdevelopment.vic.gov.au/>
- WA Department of Agriculture and Food
<https://www.agric.wa.gov.au/>

Also refer to:

The Centre for Invasive Species Solutions

<https://invasives.com.au/>

or <http://www.pestsmart.org.au>

References

1. Abrantes, J., Van Der Loo, W., Le Pendu, J., & Esteves, P. J. (2012). Rabbit haemorrhagic disease (RHD) and rabbit haemorrhagic disease virus (RHDV): a review. *Veterinary Research*, 43, 12.
2. Australian Government (2008). *Threat abatement plan for competition and land degradation by rabbits*. Department of the Environment, Water, Heritage and the Arts (DEWHA), Canberra.
3. Cox, T.E., Strive, T., Mutze, G., West, P. and Saunders, G. (2013). *Benefits of rabbit biocontrol in Australia*. PestSmart Toolkit publication, Invasive Animals Cooperative Research Centre, Canberra, Australia.
4. Elsworth, P., Cooke, B. D., Kovaliski, J., Sinclair, R., Holmes, E. C., & Strive, T. (2014). Increased virulence of rabbit haemorrhagic disease virus associated with genetic resistance in wild Australian rabbits (*Oryctolagus cuniculus*). *Virology*, 464, 415-423.
5. Invasive Animals Cooperative Research Centre (2015). *RHDV K5: Frequently asked questions*. Invasive Animals Cooperative Research Centre, Canberra.
6. OIE (2010) *Manual of Diagnostic Tests and Vaccines for Terrestrial Animals 2015*, Chapter 2.6.2 Rabbit Haemorrhagic Disease.
7. RHD-Boost (2014). *Import and evaluate new rabbit haemorrhagic disease virus (RHDV) variants to strengthen rabbit biocontrol. Report to the Vertebrate Pests Committee*. PestSmart Toolkit publication, Invasive Animals Cooperative Research Centre, Canberra, Australia.
8. Saunders, G., and Tracey, J. (2003). *Methods for introducing rabbit haemorrhagic disease virus (RHDV) into rabbit populations: Assessment of the oral delivery method*. Vertebrate Pest Research Unit, NSW Agriculture, Orange.
9. Sharp, T. (2012) RAB010: *Bait delivery of Rabbit Haemorrhagic Disease Virus (RHDV)*. PestSmart Toolkit Publication. Invasive Animals Cooperative Research Centre. Canberra.
10. Williams, K., Parer, I., Coman, B., Burley, J. and Braysher, M. (1995). *Managing vertebrate pests: rabbits*. Australian Government Publishing Service, Canberra.



CENTRE FOR
INVASIVE SPECIES SOLUTIONS

The Centre for Invasive Species Solutions manages these documents on behalf of the Invasive Plants and Animals Committee (IPAC), and has reformatted these in accordance with IPAC meeting no 9, agenda item 3.5. The authors of these documents have taken care to validate the accuracy of the information at the time of writing [July, 2016]. This information has been prepared with care but it is provided “as is”, without warranty of any kind, to the extent permitted by law.