



Australian Government

Department of Health and Aged Care
Office of the Gene Technology Regulator

March 2025

Risk Assessment and Risk Management Plan **(consultation version)**

for

DIR 214 – Trial of a genetically modified (GM) vaccine for the prevention of respiratory disease in horses

Applicant – The University of Queensland

This RARMP is open for consultation until 28 April 2025.

Written comments on the risks to human health and safety and the environment posed by this proposed trial are invited. You may make your submission

Via the consultation hub: <https://consultations.health.gov.au> (search for DIR 214)

via mail to: The Office of the Gene Technology Regulator, MDP 54 GPO Box 9848,
Canberra ACT 2601 or

or via email to: ogtr@health.gov.au.

Please note that issues regarding animal safety and the quality and efficacy of the GM vaccine do **not** fall within the scope of these evaluations as they are the responsibilities of other agencies and authorities.

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Summary of the Risk Assessment and Risk Management Plan (Consultation Version) for Licence Application No. DIR 214

Introduction

The Gene Technology Regulator (the Regulator) has received a licence application for a trial of a genetically modified organism (GMO) into the environment. It qualifies as a limited and controlled release application under the *Gene Technology Act 2000* (the Act).

The University of Queensland (UQ) proposes to conduct a trial using a genetically modified (GM) adenovirus vaccine in horses.

Rhodococcus equi is a soil-borne bacterium that causes a severe respiratory disease in young horses, known as rattles. The GM vaccine has been designed to express the *R. equi* virulence protein VapA and be replication deficient. A maximum of 10 young horses contained within UQ Gatton’s facilities would receive intramuscular injections and intranasal instillations with the GM vaccine, with the aim of evaluating the vaccine’s safety and efficacy.

Supply of veterinary products also requires approval by the Australian Pesticides and Veterinary Medicines Authority (APVMA). UQ will need to apply to the APVMA for a permit to allow the supply and limited use of the GM vaccine for the purpose of conducting research.

The Regulator has prepared a draft Risk Assessment and Risk Management Plan (RARMP) for this application, which concludes that the proposed trial poses negligible risk to the health and safety of people and the environment. Licence conditions have been drafted for the proposed trial. The Regulator invites submissions on the RARMP, including draft licence conditions, to inform the decision on whether or not to issue a licence.

The application

Project Title	Trial of a genetically modified (GM) vaccine for the prevention of respiratory disease in horses.
Parent organism	Human adenovirus 5 (AdV5)
Genetic modifications	<p>Deleted genes:</p> <ul style="list-style-type: none"> • Viral early-transcribed region 1 (E1) - to render virus unable to replicate. • Viral early-transcribed region 3 (E3) - to improve the transgene carrying capacity of the viral vector and increase the host immune response to the vector. <p>Introduced gene:</p> <ul style="list-style-type: none"> • Virulence-associated protein A (VapA) from <i>Rhodococcus equi</i> - Expression of the VapA
Principal purpose	The proposed trial aims to evaluate the immunogenicity, safety and efficacy of a genetically modified (GM) vaccine for the prevention of severe respiratory disease (rattles) in horses
Previous trials	The proposed study would be the first trial to be conducted with the GMO
Proposed limits and controls	

Proposed duration	5 years
Proposed locations	A contained trial site in Gatton, Queensland (UQ Dutton Campus)
Proposed controls	<ul style="list-style-type: none"> • only registered veterinarians would administer the GMO • only trained and authorised personnel would access the animal enclosures • a maximum of 10 horses would be administered with the GMO • personnel would use personal protective equipment (PPE) • transport, storage and disposal of the GMO would be carried out according to the Regulator’s <i>Guidelines for the Transport, Storage and Disposal of GMOs</i>

Risk assessment

The risk assessment process considers how the genetic modifications and proposed activities conducted with the GMO might lead to harm to people or the environment. Risks are characterised in relation to both the seriousness and likelihood of harm, taking into account information in the application (including proposed controls), relevant previous approvals and current scientific/technical knowledge. Both short- and long-term impacts are considered.

Credible pathways to potential harm that were considered included the potential exposure of people or animals to the GMO; and the potential for the GMO to transfer or acquire genetic material from other viruses. The potential for the GMO to be released into the environment and its effects were also considered.

The risk assessment concludes that the trial poses negligible risks to human health and safety and to the environment. No specific risk treatment measures are required to manage these negligible risks. Important factors in reaching the conclusions of the risk assessment included that the GM vaccine is replication incompetent, and unintended exposure to the GMOs would be minimised by the limits and controls.

As risks to the health and safety of people, or the environment, from the proposed trial of the GM vaccine have been assessed as negligible, the Regulator considers that the dealings involved do not pose a significant risk to either people or the environment.

Risk management

The risk management plan describes measures to protect the health and safety of people and to protect the environment by controlling or mitigating risk. The risk management plan is given effect through licence conditions. Draft licence conditions are detailed in Chapter 4 of the RARMP.

As the level of risk is considered negligible, specific risk treatment is not required. However, since this is a trial, the draft licence includes limits on the number of horses vaccinated, types of facilities used, limits on the duration of the trial, as well as a range of controls to minimise the potential for the GMO to spread in the environment. In addition, there are several general conditions relating to ongoing licence holder suitability, auditing and monitoring, and reporting requirements which include an obligation to report any unintended effects.

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Abbreviations

AdV	Adenovirus
AICIS	Australian Industrial Chemicals Introduction Scheme
APVMA	Australian Pesticides and Veterinary Medicines Authority
BSC	Biosafety cabinet
CDC	Centers for Disease Control and Prevention
DIR	Dealings involving Intentional Release
DNA	Deoxyribonucleic acid
E1-2	Early regions 1-4
EU	European Union
FDA	United States Food and Drug Administration
FSANZ	Food Standards Australia New Zealand
GM	Genetically modified
GMO	Genetically modified organism
GTTAC	Gene Technology Technical Advisory Committee
HAdV	Human Adenovirus
i.m.	Intramuscular
IATA	International Air Transport Association
IBC	Institutional Biosafety Committee
L1-5	Late regions 1-5
OGTR	Office of the Gene Technology Regulator
PC2	Physical containment level 2
PCR	Polymerase chain reaction
PCR	Polymerase chain reaction
PPE	Personal protective equipment
PPNA	Polypeptide neoantigen
RAF	<i>Risk Analysis Framework</i>
RARMP	Risk Assessment and Risk Management Plan
SOP	Standard Operating Procedure
TGA	Therapeutic Goods Administration
the Act	<i>The Gene Technology Act 2000</i>
the Regulations	The Gene Technology Regulations 2001
the Regulator	The Gene Technology Regulator
USA	United States of America
UQ	University of Queensland
WHO	World Health Organization

Chapter 1 Risk assessment context

Section 1 Background

1. An application has been made under the *Gene Technology Act 2000* (the Act) for Dealings involving the Intentional Release (DIR) of genetically modified organisms (GMOs) into the Australian environment.
2. The Act and the Gene Technology Regulations 2001 (the Regulations), together with corresponding State and Territory legislation, comprise Australia’s national regulatory system for gene technology. Its objective is to protect the health and safety of people, and to protect the environment, by identifying risks posed by or as a result of gene technology, and by managing those risks through regulating certain dealings with GMOs.
3. Section 50 of the Act requires that the Gene Technology Regulator (the Regulator) must prepare a Risk Assessment and Risk Management Plan (RARMP) in response to an application for release of GMOs into the Australian environment. Sections 50, 50A and 51 of the Act and sections 9 and 10 of the Regulations outline the matters which the Regulator must take into account and who must be consulted when preparing the RARMP.
4. The Risk Analysis Framework (OGTR, 2013) explains the Regulator’s approach to the preparation of RARMPs in accordance with the Act and the Regulations. The Regulator has also developed operational policies and guidelines that are relevant to DIR licences. These documents are available from the Office of the Gene Technology Regulator (OGTR) website.
5. Figure 1 shows the information that is considered, within the regulatory framework, in establishing the risk assessment context. This information is specific for each application. Potential risks to the health and safety of people or the environment posed by the proposed release are assessed within this context. Chapter 1 provides the specific information for establishing the risk assessment context for this application.

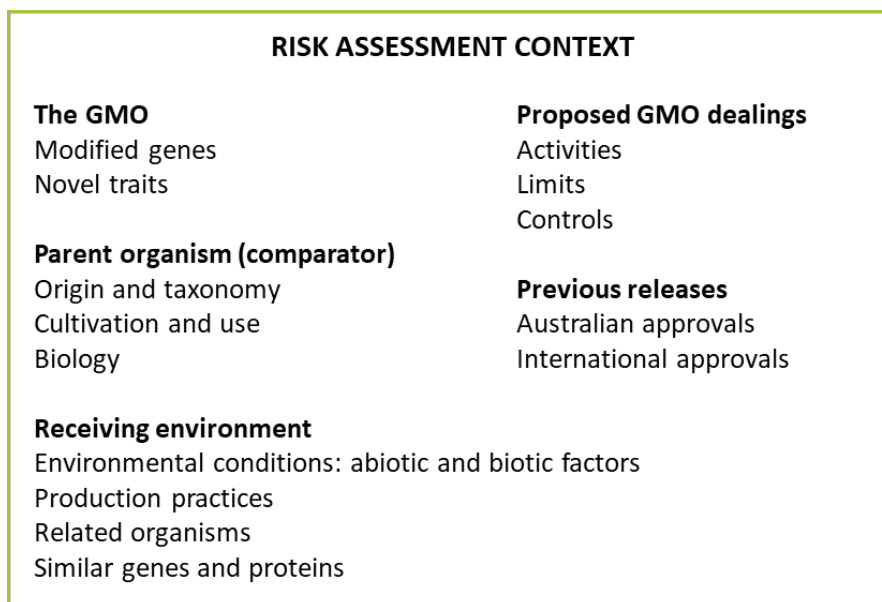


Figure 1. Summary of parameters used to establish the risk assessment context, within the legislative requirements, operational policies and guidelines of the OGTR, and the Risk Analysis Framework

6. In accordance with section 50A of the Act, this application is considered to be a limited and controlled release application, as the Regulator was satisfied that it meets the criteria prescribed

by the Act. Therefore, the Regulator was not required to consult with prescribed experts, agencies and authorities before preparation of the RARMP.

1.1 Interface with other regulatory schemes

7. Gene technology legislation operates in conjunction with other regulatory schemes in Australia. The GMOs and any proposed dealings may also be subject to regulation by other Australian government agencies that regulate GMOs or GM products, including Food Standards Australia New Zealand (FSANZ), the Australian Pesticides and Veterinary Medicines Authority (APVMA), the Therapeutic Goods Administration (TGA), the Australian Industrial Chemicals Introduction Scheme (AICIS) and the Department of Agriculture, Fisheries and Forestry (DAFF).

8. To avoid duplication of regulatory oversight, risks that have been considered by other regulatory agencies will not be re-assessed by the Regulator.

9. The APVMA provides a national registration and permit scheme for agricultural and veterinary chemical products. It administers the provisions of the *Agricultural and Veterinary Chemicals Code Act 1994* (AgVet Code). For registration, the APVMA assesses whether a new veterinary vaccine meets the criteria set out in the AgVet Code before it is registered in the Register of Agricultural and Veterinary Chemical Products. A new veterinary vaccine that is not registered may be legally used, such as in animal trials, by obtaining a permit from the APVMA.

10. As part of the permit process, the APVMA assesses the quality, safety and efficacy of the vaccine. Quality aspects could include batch-to-batch consistency in vaccine composition, purity and potency. The APVMA audits the Good Manufacturing Practice record of the applicant. Safety aspects include the toxicological profile of the vaccine and its residues, including metabolites and degradation products. Associated food safety risks and consumer dietary exposure are also considered by the APVMA. The APVMA approves the label, handling and directions for supply of veterinary vaccines to ensure safe use. The APVMA may also impose conditions on a permit for the supply of veterinary vaccines for research purposes.

11. Research involving animals has to be undertaken under the oversight of the relevant statutory framework within state and territory animal welfare acts. The statutory framework has been supplemented gradually by codes of practice and ethical principles developed by the National Health and Medical Research Council.

1.2 The proposed dealings

12. Rattles, a severe respiratory disease affecting young horses (<12 months of age/foals), is a major cause of illness and death in thoroughbred foals. The disease is caused by the ubiquitous soil bacterium *Rhodococcus equi*. Foals become infected with the bacterium by inhaling contaminated dust particles. Once in the lungs, the bacterium is engulfed by immune cells called macrophages. The bacterium then upregulates the expression of a protein called VapA, which aids its survival by disrupting the macrophages' ability to destroy *R. equi*.

13. The University of Queensland (UQ) is seeking authorisation to carry out a trial with a GM adenovirus-based vaccine (the GMO) in young horses. The GMO will deliver an expression cassette which encodes the *R. equi* VapA protein, with the aim of eliciting an immune response against it. The proposed trial will evaluate the safety and efficacy of the GM adenovirus based vaccine following intramuscular and intranasal instillations.

14. The dealings involved in the proposed trial are to:

(a) conduct experiments with the GMO, which includes:

- i. preparing the GMO for administration;
- ii. administering the GMO to horses by intramuscular (i.m) injection or intranasal instillations;
- iii. collecting samples from horses;

(b) transport the GMO; and

(c) dispose of the GMO;

and the possession (including storage), supply and use the GMO for the purposes of, or in the course of, any of these dealings.

1.3 The proposed limits of the trial (duration, scale, location, people and animals)

15. The trial is proposed to take place over a period of 5 years from the date of issue of the licence. Up to 10 horses would receive both i.m and intranasal instillations of the GMO.

16. The trial would take place in containment facilities at the UQ Gatton campus.

1.4 The proposed controls to restrict the spread and persistence of the GMOs in the environment

17. The applicant has proposed a number of controls to minimise exposure to the GMO, and to restrict the spread and persistence of the GMOs in the environment. These include:

- Staff preparing and administering the GMO would use personal protective equipment (PPE) including gowns, gloves, face masks and safety glasses;
- Staff entering the enclosures would use gloves and waterproof footwear. They would visit animals that have not been exposed to the GMO prior to entering the enclosures housing animals that have received the GMO. After exiting the enclosures, the footwear will be chemically decontaminated and kept on site in closed containers;
- Staff collecting biological samples will wear PPE as described above;
- Disinfectant, either 0.9% (w/v) Virkon™, 60% (v/v) ethanol or 2,500 ppm sodium hypochlorite, would be used to decontaminate equipment and areas immediately adjacent (2m radius) to where the release has occurred.

- Small disposable items (tubes, gloves, needles, syringes) used in connection with the GMO will be disposed of by placing in an appropriate container immediately after use. The container will then be autoclaved and disposed of through the institutional clinical waste streams.
- At the end of the proposed release, the immunised horses will be euthanised and the carcasses disposed of via deep burial.
- All materials that cannot be decontaminated will be disposed of via clinical waste streams using an approved contractor.

1.5 Details of the proposed dealings

1.5.1 *Manufacturing of the GMO*

18. The GMO was manufactured using a commercial system under a NLRD assessed by UQ's Institutional Biosafety Committee (IBC). The GMO was manufactured and stored in PC2 facilities at UQ St Lucia Campus.

1.5.2 *Transport and storage of the GMO*

19. The applicant proposed to transport, store and dispose of the GMO under an NLRD. Briefly, the GMO will be transported from PC2 facilities at the UQ St Lucia campus to the Gatton campus in university vehicles. The GMO would be contained in 25ml screw cap vials and placed in a plastic bag, then contained within a hard plastic, sealable container which will be labelled as containing GMOs.

20. Biological samples containing the GMO will be transported from the trial site to the UQ St Lucia campus and analysed under an NLRD. GMO waste will remain on site for decontamination.

21. A logbook will be kept at each facility to document the transport and receipt of the GMO and biological samples containing GMOs.

1.5.3 *Trial site*

22. The proposed trial site located at the UQ's Gatton campus within the Queensland Animal Science Precinct (QASP). The QASP facility is surrounded by a secure external fence that is regularly checked to ensure its integrity.

23. QASP can only be accessed through locked control points by authorised personnel.

24. The GMO administration would take place within a DAFF approved biosecurity section of the QASP facility. The biosecurity section is an enclosed building which contains holding pens and has its own perimeter fence with access via locked gates that can only be opened by authorised personnel approved by the Facility Manager. This would serve as the primary facility where GMO-administered horses will remain for at least 2 weeks or until the GMO is no longer detected in nasal swab samples. Once the animals have returned a negative nasal swab PCR test for the GMO, they would be transferred to an outdoor section of the facility which is also enclosed by secure fencing. In this RARMP, the outdoor section is described as the secondary facility. The proposed trial site is shown in Figure 2.



Figure 2 The location of the proposed release site (boxed area) (provided by the applicant).

1.5.4 Trial design

25. The applicant has proposed a trial design involving 2 study groups, and up to 10 horses. All animals will be administered a primary dose of the GMO (1 mL containing 1×10^7 infectious units) via intramuscular injection into the muscles of the upper neck. Experimental group 1 (n=5) will be administered a booster dose (1 mL containing 1×10^9 infectious units) using the same procedure, while the booster dose for experimental group 2 (n=5) will be delivered via intranasal instillation (1 mL containing 1×10^9 infectious units). Nasal swabs, blood, and rectal temperature measurements will be collected at regular intervals indicated in Table 1 below.

Table 1. Design of the trial.

Procedure conducted on animals	Timepoints (Days)
Intramuscular injection of GMO	0 (N=10), 14 (N=5)
Intranasal instillation of GMO	14 (N=5)
Blood collected	0,7,14,21,28,35,42
Nasal swabs collected	0-7,14-21
Monitoring (measurement of rectal temperature, inspection of injection sites)	Several time points from day 0 to 42.
End point (All animals euthanised and tissue samples collected)	42

26. Blood samples collected from the animals will be tested for the presence of antibodies against VapA.

27. The nasal swabs collected from the vaccinated animals will be tested for the presence of the GMO vector and transgene via qPCR. If samples collected on Day 21 are negative for both the GMO vector and transgene, the animals will be moved to the secondary large animal facility within QASP.

28. Animals which test positive will be tested at 5 day intervals until they return a negative test. Additionally, samples which test positive for GMO DNA, would be further tested for the presence of viable viral particles, including any potential recombinant viral strains with restored replication ability.

29. The movement of animals that tested negative by PCR to the GMO to the secondary facility may be subject to approval from other agencies.

1.5.5 Preparation and administration

30. The preparation of GMO would take place, on a steel bench in the anteroom of the enclosed facility containing the holding pens immediately prior to vaccinating the animals. During the preparation and administration of the GMOs, trained personnel will be wearing PPE including disposable overalls that include an attached hood, P2 face masks, a full-face shield, and knee-high rubber boots.

31. Horses will be restrained with a tether rope attached to a head halter. The tether will be attached to the fencing components of the holding pens to prevent any sudden/unexpected movements by the animals. The animals will be trained in this process prior to injection so they become accustomed to the process.

32. Needles will be used for i.m. injections. After administration, the needle-syringe will be disposed of directly into an appropriate sharps container.

33. Administration of the GMO via intranasal instillation will be performed using syringes with no needles attached. The 1 mL dose will be delivered dropwise with the head of the animal slightly elevated. The timing of the administration of the drops will be synchronised with the animal breathing in. The nostril will also be gently sealed with a gloved hand around the syringe as drops are dispensed. The second nostril will remain uncovered. The head of the animal will remain elevated for 2 to 3 minutes after the dose is delivered, with the nostril gently covered by the gloved hand.

1.5.6 Sample collection and analysis

34. Sample collection will be performed by trained personnel who are wearing appropriate PPE including knee-high rubber boots, disposable overalls (including hood), two pairs of disposable gloves, P2 mask and a full-face shield.

35. Blood and nasal swab samples will be collected at the time points depicted in Table 1.

36. On day 42 all animals will be euthanised and tissues will be collected. These tissues would include nasal, oropharyngeal, muscle from the immunisation site, muscle distant from the immunisation site, liver and kidney. Samples will be transported to PC2 laboratories at the UQ campus and analysed under a NLRD.

1.5.7 Decontamination and disposal

37. The applicant has proposed that either 0.9% (w/v) Virkon™, 60% (v/v) ethanol or 2,500 ppm sodium hypochlorite, be used to decontaminate equipment and areas immediately adjacent (2m radius) to where administration of the GMO has occurred.

38. The applicant has also proposed to take regular environmental swab samples from areas within the trial site and test these for the presence of GMO nucleic acids. If GMO DNA is detected in any of the environmental samples the area from which the swab was taken and adjacent areas would be decontaminated. The positive sample would be further tested for viable and replicative particles.

39. All hard impermeable surfaces in the areas where the administration of the GMO is taking place would be decontaminated after the administrations have occurred.

40. All faeces, urine and saliva from the vaccinated animals will be washed from the holding pens within the primary facility daily. The wash off material will be treated with 2,500 ppm sodium hypochlorite for 10 minutes, and the material discharged into the facility's settlement ponds. Any large particulate (10cm³) matter will be broken up to ensure contact with the disinfectant.

41. Urine and saliva excretions cannot be treated at the secondary large animal facility, However, faecal material will be regularly collected and stored in a pile within the pens over the course of the proposed trial. At the end of the proposed trial, it will be saturated with 2,500 ppm sodium hypochlorite for 10 minutes and discarded.

42. GMO-contaminated waste generated during the proposed trial and sampling of animals, such as PPE items (disposable overalls, disposable gloves, etc), plasticware (syringes), sharps (needles), scalpel blades, and packaging material for individual items will be disposed of via the approved clinical waste streams in accordance with an approved NLRD.

1.5.8 Training

43. All personnel involved in the proposed trial will be trained by the UQ's biosafety team and the project leader. Training will include biosafety modules and instruction regarding the nature of the GMO.

44. Personnel will also receive specific training on the general requirements of the proposed release and any specific conditions imposed by the regulator. Records of the training and acknowledgement of having understood their obligations will be kept and made available on request.

45. With respect to handling of horses, the proposed trial will take place at a facility dedicated to research on large animal species. Handling of horses will be performed by qualified research and veterinary staff with many years of experience working with horses

1.5.9 Accountability and Monitoring

46. The applicant proposes to produce a manual for the trial. The manual will a copy of the licence, training records of the staff, records of transport and administration of the GMO and records of decontamination and disposal of the GMO.

1.5.10 Contingency plans

47. In the event of the escape of an animal, unintentional release, spill, leak, or loss of GMOs, efforts will be made to locate and/or retrieve the GMOs and return them to containment or render them non-viable as soon as reasonably practicable. The incident will be immediately reported to the UQ Biosafety Advisor, who will advise the Regulator and the UQ IBC as soon as possible. In the event of exposure of a person to the GMO, the applicant has proposed that the person would be instructed to wash the exposed area with excess of water and soap or antiseptic and seek medical attention if required.

Section 2 Parent organism

48. The characteristics of the parent organism provide a baseline for comparing the potential for harm from dealings with the GMO. The GM vaccine is derived from the human adenovirus serotype 5 (HAdV-5).

2.1 Classification and genome characteristics

49. Adenoviruses (AdVs) are non-enveloped, icosahedral virions belonging to the *Adenoviridae* family. The family consists of 6 genera which are delineated based on sequence similarity as well host range. These include Aviadenovirus which are only known to infect birds, Barthadenovirus which predominantly infect scaled reptiles, Ichtadenovirus which infect fish, Siadenovirus which

infect frogs and tortoises, and Testadenovirus which only infect turtles and tortoises (Benkő et al., 2022). The most aetiologically significant genus in the family, Mastadenovirus, exclusively infect mammals. Also belonging to this genus are the 7 currently described species of human adenoviruses which are classified from A-G (Robinson et al., 2011). HAdV-5 is a member of species C (Yang et al., 2019).

50. With linear double stranded DNA genomes ranging from 26 to 45kb long, adenoviruses are among the largest non-enveloped viruses known to date (Gallardo et al., 2021; Lally et al., 1984). HAdV-5 sits in the middle of this range with a genome of approximately 36kb which encodes for over 40 proteins (Akello et al., 2021). The genome contains 6 discrete transcriptional units with 5 of these units (E1a, E1b, E2, E3, E4) being transcribed early in the replication process, and one unit being transcribed later (Afkhami et al., 2016).

51. The early genes are involved in initiating replication of the viral genome and host immune evasion (King et al., 2018). The late transcriptional unit is further segmented into 5 elements (L1, L2, L3, L4, L5) which all encode different structural components of the virus (Afkhami et al., 2016). Flanking both ends of the genome are inverted terminal repeats which harbour the viral origin of replication (Hay, 1985; Lusby et al., 1980).

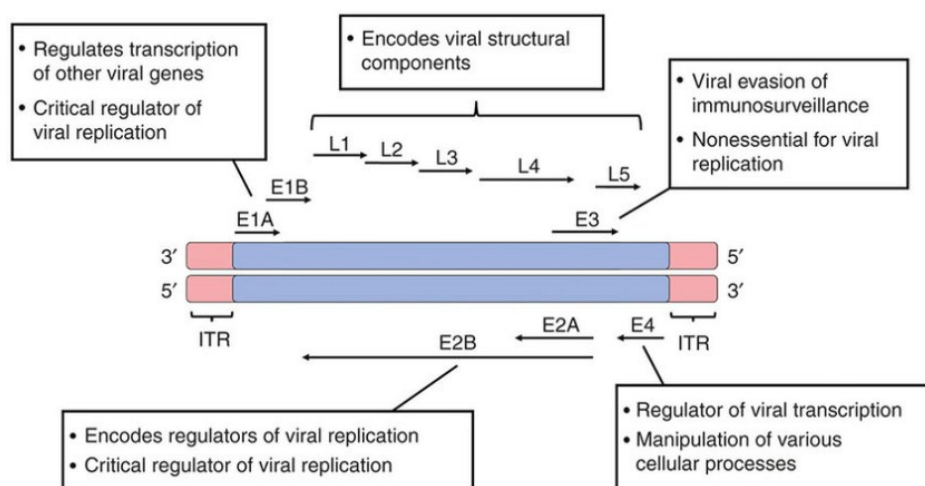


Figure 3. Schematic of general genomic organisation of human and mammalian AdVs, as presented in Afkhami et al. (2016)

52. The E1 and E3 coding regions are exclusive to Mastadenoviruses (human and mammalian AdVs) (ICTV, 2023). The E1 regions are essential for viral gene expression and replication (Roy et al., 2004; Saha and Parks, 2017b). The E2 gene encodes E2 proteins which are mainly involved in viral DNA replication and transcription of late genes (Roy et al., 2004; Saha and Parks, 2017b). The E3 gene encodes viral proteins that aid the virus in evading the host immune response. The E4 gene modulates cellular function and assists with viral DNA replication and RNA processing. The late transcriptional units (L1-5) encode viral structural and non-structural proteins that are involved in the capsid formation and maturation of the new virions (Roy et al., 2004; Saha and Parks, 2017b).

53. The virus is characterised by an icosahedral shaped (pseudo icosahedral) capsid which is comprised of three major proteins, hexons, penton bases, and fibers (Robinson et al., 2011). The fibres are primarily purposed for attachment, while the hexons and penton bases form the structural components of the capsid (Jennings and Parks, 2023). There are several additional proteins which comprise the virus capsid including IIIa, VI, VIII, and IX (Reddy et al., 2010; Russell, 2009). Protein VII is largely responsible for wrapping the viral DNA and IVa2 protein packs it into the immature virions (Jennings and Parks, 2023).

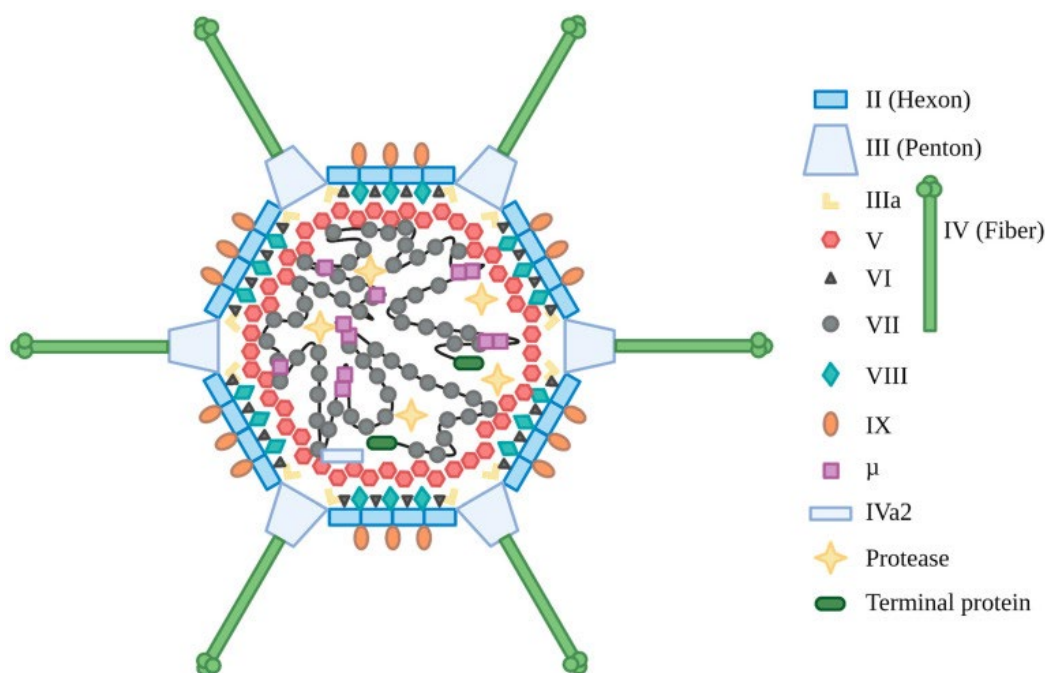


Figure 4. Adapted from (Jennings and Parks, 2023) displays the arrangement of the proteins that comprise the adenoviral particle. The hexons and pentons form the major structural components of the capsid and are coloured blue, whilst the fibres which enable viral attachment to the host receptor are green.

2.2 Infection cycle

54. The life cycle of an adenovirus takes approximately 24–36 hours and involves three major stages (infection, replication and assembling) (Giberson et al., 2012). The route of infection is usually via aerosol into the respiratory or gastrointestinal tracts, the oropharynx or the conjunctiva (Khanal et al., 2018; Usman and Suarez, 2020). For human adenoviruses in the A,C,E, and F species, this is primarily by binding to coxsackie adenovirus receptors (CARs) which is facilitated by the fibre protein (Bergelson et al., 1997; Russell, 2009). B species adenoviruses bind to the CD46 receptor whilst D species members can utilise both (Khanal et al., 2018; Marttila et al., 2005). Upon binding to these receptors, viral particle uptake is facilitated by the clathrin-coated pits of the early endosomal pathway (Wodrich et al., 2006).

55. The now partially uncoated virus must translocate to the nucleus where it can replicate its genome. To do this, the viral particle commandeers cellular microtubules utilising dynein to bind to them (Kelkar et al., 2004). Nuclear localisation signals located on critical components of the remaining capsid ensure that entry into the nucleus occurs (Kelkar et al., 2004; Wodrich et al., 2006; Wodrich et al., 2003). From inside the nucleus, expression of E2 genes produce the 3 primary components of replication machinery which are the precursor terminal protein (pTP), AdV DNA polymerase (AdV DNA pol) and the DNA-binding protein (DBP)(Hoeben and Uil, 2013). These assemble onto the ITRs of viral genomes in order to replicate them (Hoeben and Uil, 2013). Following replication, the mature virions can be assembled in a process that involves the sequential assembly of pentons, hexons and minor structural proteins into empty capsids, and then packaging of the newly replicated viral genomes into them (Ahi and Mittal, 2016). A viral protease then cleaves the precursor proteins creating the mature particles (Ahi and Mittal, 2016).

56. Initially expressed at low levels from the E3 region, Adenovirus Death Protein (ADP) increases its expression triggered by virion assembly via the major late promoter (MLP) and induces the lysis of the host cell (Georgi and Greber, 2020). This in turn allows the egress and dissemination of the virus.

57. Adenoviruses are also known to remain in the nucleus in the form of an episome (Harui et al., 1999). Although the details of persistence are yet to be fully understood, it can last for several years and is thought to be mediated by the host cells ability to suppress early phase gene expression (Ehrhardt et al., 2003; Tatsis et al., 2007; Zheng et al., 2016).

58. Because they lack the appropriate enzymes, adenoviruses cannot actively integrate themselves into their host's genome (Dehghan et al., 2019; Desfarges and Ciuffi, 2012; Harui et al., 1999; Hoppe et al., 2015). Although extremely rare, it is possible for them to randomly become integrated into the genomic DNA (Harui et al., 1999; Stephen et al., 2008).

2.3 Pathology

59. The distribution of human adenoviruses is global and infections occur more frequently in susceptible populations such as young children, the immunocompromised, and those living in crowded spaces including military training camps (Hendrix et al., 1999; Khanal et al., 2018). In these instances, adenovirus is capable of causing disease in a range of tissues leading to conditions including, acute respiratory disease, adenoidal–pharyngeal conjunctivitis, keratoconjunctivitis, hepatitis, acute gastroenteritis, persistent interstitial infection in the kidney, and haemorrhagic cystitis. The species of adenovirus infection is typically predictive of the particular tissue in which the disease state occurs. For example, HAdV species C serotypes 1, 2, and 5 are a common cause of respiratory infection in young children. Outside of vulnerable populations, the vast majority of adenoviral infections are subclinical (Benkő et al., 2022).

60. HAdVs are generally transmitted by aerosol droplets excreted from the respiratory tract of an infected individual. It is estimated that adults lacking specific anti-HAdV antibodies may be infected by inhaling as few as 5 viral particles. However, only 10 % of exposed individuals may become ill (Musher, 2003) although this is likely higher in children.

61. HAdVs can also be transmitted by ocular secretions or by the oral-faecal route with food and water as possible vectors. They can be indirectly spread by towels, handkerchiefs, food, eating utensils and other items that were contaminated by an infected person (Pond, 2005).

62. The incubation period of natural HAdV infection ranges from 2 days to 2 weeks, depending on the viral species and serotype as well as the route of infection (Allard and Vantarakis, 2017; Flinders Womens and Childrens and Flinders Medical Centre, 2023). For respiratory infections, the incubation period is generally 4-8 days, and 3-10 days for intestinal infections (Allard and Vantarakis, 2017). The symptoms of mild infection usually last for a few days to a week but for the severe infections, symptoms may last longer.

2.4 Host range

63. Human and non-human AdVs have a range of vertebrate hosts including people, horses, cattle, pigs, sheep, goats and domestic fowl, wild birds, bats and reptiles (Allard and Vantarakis, 2017). Humans are the natural host for HAdVs (Benkő et al., 2022). Although animal models have been used as tool to study HAdV infections, there is no report of natural HAdV infections of non-human hosts (Bertzbach et al., 2021; Ismail et al., 2019).

64. HAdVs have been found in the faeces of domestic animals living near humans, such as dogs, goats, pigs, and sheep, and in faeces from wild guinea pigs. HAdV and canine AdV were found concomitantly in faeces samples from pampas foxes (*Lycalopex gymnocercus*). The presence of HAdV in the faeces of these animals was not considered a threat to the health of the animals, since it is more likely that the virus was ingested and eliminated in faeces but did not replicate in the gut of those animals (Monteiro et al., 2015; Pauly et al., 2015). However, these animals could act as reservoirs for humans and non-human adenoviruses.

65. The administration of HAdVs to mice, rabbits, rats, guinea pigs and non-human primates resulted in the development of specific antibodies. However, no clinical signs of a systemic disease

have been observed in infected animals, suggesting that HAdVs have restricted ability to replicate and cause disease in non-human hosts (Pereira and Kelly, 1957).

2.4.1 *Shedding and Biodistribution*

66. HAdV shedding is largely dependent on the tissue and infection route. Respiratory infections are expected to generate the highest viral load soon after infection and virus can be detected in respiratory samples for up to 2 months following infection (Huh et al., 2019). It is estimated that sputum or oral secretions of infected adults contain 10^6 to 10^7 viral particles per millilitre (mL) (Musher, 2003).

67. HAdV shedding was also evaluated in faecal and oral swabs after the administration of a live, oral vaccine containing two serotypes of replication competent HAdV (HAdV-4 and HAdV-7). Over half of the vaccine recipients shed viable viral particles in faecal samples between 7-28 days following vaccination. No shedding was detected after 28 days of vaccination or at any time point in throat swabs (Allard and Vantarakis, 2017).

68. The presence of genomic DNA of HAdV species C was observed in human tonsil and adenoid tissues after surgical removal. Viable viruses were isolated following long-term culture of tissue samples with permissive cells, suggesting that low levels of infectious viruses may persist in these tissues in a latent form (Proenca-Modena et al., 2019).

69. In animal models, viable viral particles were recovered in cultures of spleen cells collected from rabbits and guinea pigs 8 weeks after intravenous administration of wild type (WT) HAdV-5. However, the number of viral particles was very low and only observed when cells were cultured for long periods (~70-100 days) (Faucon et al., 1974; Pereira and Kelly, 1957).

70. As discussed in Section 2.2, a small amount of WT AdV DNA can persist as episomal copies in the cytoplasm of infected cells. This latent form of the virus may be reactivated if the host becomes immunosuppressed, leading to a new cycle of infection and viral shedding (Radke and Cook, 2018).

2.5 Prevalence

71. An estimation of the seroprevalence of HAdV-4, 5, -26 and -35 (serotypes commonly tested in the clinics or used in clinical/pre-clinical trials) is shown in Figure 4. These data are analysed based on approximately 30 studies published over the past 20 years (Mennechet et al., 2019). HAdV-5 is the most widely reported and has the highest seroprevalence globally. HAdV-26, appears to have high seroprevalence in Africa and Asia; and low in North America and Europe (Mennechet et al., 2019).

72. In Australia, the Laboratory Virology and Serology (LabVISE) reports from the Department of Health (1991-2000) showed an average of about 1400 reported cases of AdV infection per year over 10 years and only about 18 reported cases of HAdV-D26 infection (Spencer, 2002). It is important to note that the majority of AdV reported infections have not been serotyped and that testing for AdV infections may not be common in Australia. These numbers indicate that HAdVs are

present in the Australian environment.

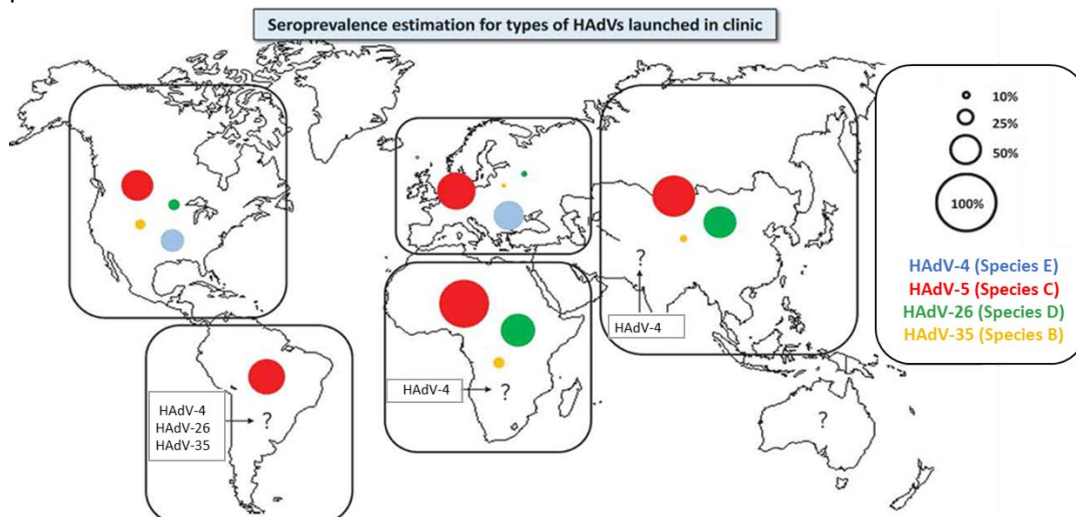


Figure 5: Seroprevalance for AdV types used in the clinic (Adapted from Mennechet, 2019)

2.6 Integration, mutation and recombination of AdV

73. As described in Section 2.2 of this chapter, AdV DNA can be maintained as multiple episomal copies in the cytoplasm of infected cells (Harui et al., 1999). In addition, AdVs do not have the machinery for efficient integration into the host genome and exhibit extremely low levels of integration (Dehghan et al., 2019; Desfarges and Ciuffi, 2012; Harui et al., 1999; Hoppe et al., 2015). Random integration of virus DNA into the host genome has been observed in very rare cases (Harui et al., 1999; Stephen et al., 2008).

74. Mutation and homologous recombination are important source of genetic variation in viruses. The HAdV-5 shows a mutation rate of 0.0046 substitutions per genome replication, this value is similar to other double-stranded DNA viruses (Risso-Ballester et al., 2016). Homologous recombination can occur when a host cell is infected by multiple AdVs at the same time. A recently isolated recombinant HAdV strain belonging to Species C is proposed to have originated through the recombination between HAdV-1 and HAdV-2 (Zhang and Huang, 2019). Homologous recombination appears to be restricted to members of the same species and occurs in the regions of high sequence homology (Lukashev et al., 2008). However, bioinformatic analysis suggested that HAdV-E4, a species E AdV was a result of a recombination event between species B and C (Gruber et al., 1993).

75. Genomic analysis of 51 circulat strains showed that recombination among HAdV Species C is more likely to occur in the regions E4 and E1, while recombination events in the hexon and fibre gene and the E3 region were almost absent. As E1 and E4 regions encode genes that regulate the transcription and other cellular processes, recombination events in these regions do not result in serotype diversity (Dhingra et al., 2019). This is in contrast to that observed in HAdV Species D (HAdV-D), where recombination events between genes encoding the major capsid proteins (hexon, penton base, fibre) resulted in more than 50 HAdV-D serotypes (Robinson et al., 2013).

2.6.1 Antiviral treatments for human AdV

76. There is no specific treatment for AdVs. Most AdV infections are mild and do not require medical care (CDC, 2019). Antiviral drugs such as Cidofovir and Ribavirin may be used as treatment for severe AdVs disease in immunocompromised individuals (Yusuf et al., 2006; Waye and Sing, 2010; Lion, 2019).

2.6.2 Risk group of human AdVs

77. The Australian Standard 2243.3:2022 Safety in Laboratories Part 3: Microbiological safety and containment (Standards Australia/New Zealand, 2022) classifies AdV as a Risk Group 2 organism.

2.7 Environmental stability and decontamination methods for human AdV

78. AdVs are resistant to many common disinfectants and can remain infectious for long periods in the environment. Most serotypes are stable at 36 °C for a week, for several weeks at room temperature, and for several months at 4 °C. They are stable for weeks in tap water, sewage effluent and sea water and for 7 days to 3 months on dry surfaces (Public Health Agency of Canada, 2014; CDC, 2019).

79. AdVs can be effectively inactivated using heat treatment (56 °C for 30 min, 60 °C for 2 min or autoclaving) or chemical agents such as chlorine, formaldehyde or alcohol-based disinfectants (Rutala et al., 2006; Public Health Agency of Canada, 2014; Allard and Vantarakis, 2017; CDC, 2019). For example, surfaces can be decontaminated with 70% (v/v) ethanol, 0.2% (v/v) chlorine or 0.9% (w/v) Virkon S (>5 min contact time) (Rutala et al., 2006). Liquid waste may be treated with bleach with a final concentration of 10% (v/v) (~0.5% chlorine), for 15 minutes (Allard and Vantarakis, 2017).

2.8 Adenovirus as a vaccine vector

80. Adenoviral vectors have been extensively investigated as vaccines, gene and cancer therapies in non-clinical and clinical trials. They induce strong immune responses and have been shown to be effective and safe (Tolcher et al., 2006; Wold and Toth, 2013a; Stebbings et al., 2022). In 2021, the TGA approved COVID-19 vaccines Vaxzevria (AstraZeneca, 2022) and Jcovden (Janssen) which are based on adenoviral vectors derived from ChAdV-Y25 and HAdV-26, respectively (Mendonca et al., 2021; TGA, 2022). Although AstraZeneca has removed Vaxzevria from the Australia market (Therapeutic Goods Administration, 2024), it is estimated that about 14 million doses of the Vaxzevria vaccine have been administered in Australia (TGA, 2023). Common side effects observed include injection site pain or tenderness, tiredness, headache, muscle pain, fever and chills. Severe side effects were observed in rare cases and included severe allergic reaction, blood clots, myocarditis and pericarditis (Australian Government - Department of Health and Aged Care, 2023).

81. A rabies vaccine (ONRAB®) based on replication-competent HAdV-5 encoding rabies glycoprotein has been used to protect wildlife in North America since 2006 (Rosatte et al., 2009). The vaccine consists of a suspension of the recombinant AdV encapsulated in a plastic blister and it is recommended for field vaccination of skunks, raccoons and foxes. It is distributed as baits in the environment by hand placement or dropped from a low flying aircraft. When an animal finds and chews the bait, it releases the vaccine into the animal's mouth, which will orally vaccinate the animal against rabies. Studies conducted in non-target animals (i.e. wood rats, eastern cottontail rabbits, Virginia opossums, eastern wild turkeys and fox squirrels) experimentally vaccinated with high doses of ONRAB®, showed that the vaccine does not induce behaviour changes, superficial or histological lesions. These results suggest that the vaccine offers a low risk of adverse effects to wild populations (Fry et al., 2013; USDA, 2019).

82. Adenoviral vectors have also been used to deliver neoantigen-based cancer therapies. Neoantigens are peptides or proteins expressed in tumour cells as result of random mutations in the cell genome. They are recognised by the host immune system as foreign or non-self, triggering an immune response to eliminate the cancer cell. As neoantigens are unique to each type of cancer, the risk of inducing autoimmunity is very low, qualifying them as safe immunogens with low risk of damage to healthy tissues (D'Alise et al., 2019). Non-clinical and clinical studies have shown that adenoviral vaccines delivering neoantigens, can induce cellular immunity against cancer

cells. (D'Alise et al., 2019; D'Alise et al., 2022; Palmer et al., 2022). Neoantigen-based therapies can be personalised to individual patients (Zhao et al., 2021) and have been considered safe and well tolerated (Palmer et al., 2022).

Section 3 The nature of the GMO and effect of the modifications

83. The two regional deletions and single gene insertion of the GMO are tabled in the application summary above. Further details of the role of these modifications are provided below.

3.1 The genetic modifications

84. The GM vaccine has been produced using a commercially available replication-deficient adenovirus vector system (Takara Bio USA Inc, 2025), (Clontech Laboratories, 2012). The supplied plasmid vector encodes for a HAdV5 genome which has both the E1 and E3 regions deleted and allows for the generation of recombinant adenovirus vectors expressing heterologous antigens.

85. The expression cassette is inserted in the E1 region of the AdV vector. The cassette contains a cytomegalovirus immediate-early (CMVie) promoter driving the expression of the VapA virulence gene from *R. equi*. The CMV promoter can drive the expression of recombinant proteins in a wide range of mammalian cells and is a common choice in the construction of viral vectors for gene therapies and vaccines (Au et al., 2022).

3.2 The effect of the genetic modifications

3.2.1 Deletion of E1 and E3 regions

86. As previously mentioned, the E1 region of HAdV5 is a critical part of the replication cycle and without it, the virus is rendered completely replication incompetent. The E1 region of the genome encodes for 2 separate proteins, named E1A and E1B.

87. E1A potentially transforms host cellular environments to permit HAdV infection by interacting with a gamut of regulatory proteins, despite containing no intrinsic DNA-binding or enzymatic activity (King et al., 2018). Different isoforms of E1A are expressed during different stages of infection (King et al., 2018). In the early part of infection, the expression of this protein drives the host cell into a replication phase, in turn allowing the virus to replicate (Ferrari et al., 2014; Ferrari et al., 2008; Ghosh and Harter, 2003).

88. E1B also plays an important role in HAdV replication and dissemination. This multifunctional protein can repurpose factors from the host cell to promote the transcription of downstream regions of the viral genome. It can also facilitate the degradation of host antiviral proteins and decrease the host ability to block viral replication (Hidalgo et al., 2019). E1B has also been implicating in blocking the translation of p53, a protein with immunological functions (Hidalgo et al., 2019).

89. The deletion of both the E1A and E1B renders the virus replication incompetent and attenuates the HAdV5s ability to evade the host immune system (Saha and Parks, 2017a).

90. The E3 region of the genome is not essential for viral replication *in vitro* but does confer additional immune evasion capabilities *in vivo* (Ginsberg et al., 1989). It achieves this by blocking an immune response that would normally lead to host cell lysis (Lichtenstein et al., 2004). Therefore, deletion of the E3 region of HAdV serves to further attenuate the virus *in vivo*

3.2.2 Insertion of the VapA expression cassette

91. The inserted gene VapA is derived from *R. equi*, a widely distributed soil-borne bacterium which can cause significant, often fatal pulmonary disease when inhaled by foals. Human infection is rare and usually limited to the immunocompromised (Prescott, 1991). The bacterium can exist

both freely in soil, and as a facultative intracellular pathogen. There is even evidence it may colonise amoebae (Price et al., 2024).

92. In the bacterium, VapA is encoded on a pathogenicity island of the roughly 80kb pVAPA plasmid (Okoko et al., 2015). There are many homologues of VapA in closely related bacterial species, but few of them confer virulence to the degree that VapA does (Ganderton et al., 2023; Miranda-CasoLuengo et al., 2024; Okoko et al., 2015). This may be due to the unique N-terminal domain of VapA which allows it to be expressed as surface protein and subsequently released into the phagosome lumen (Miranda-CasoLuengo et al., 2024)

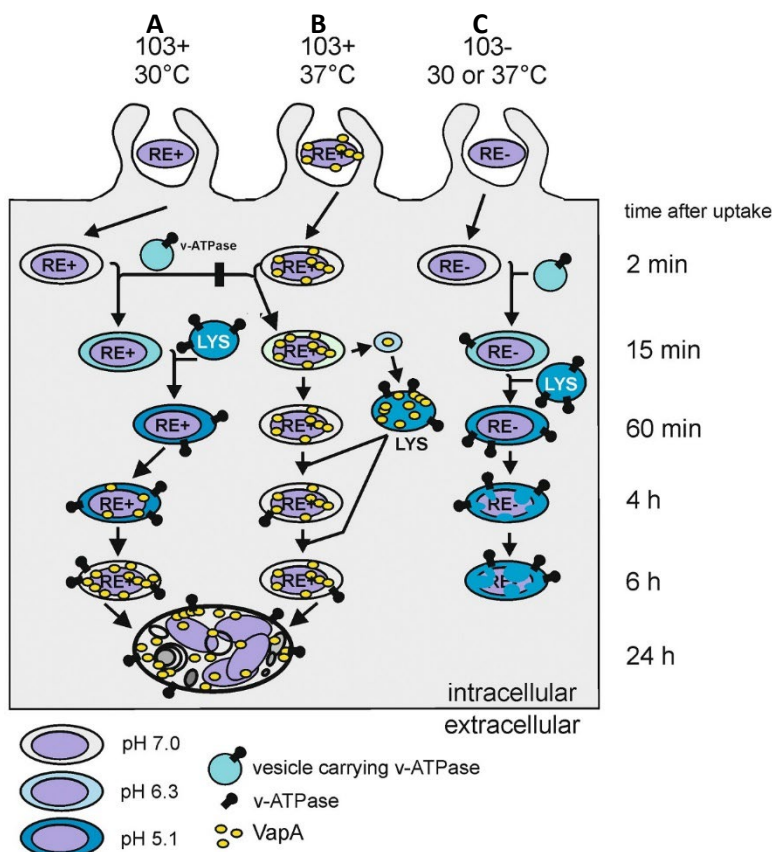


Figure 6. VapA expression facilitates infection of the host macrophage when expressed at high levels. 103+ (A) represents *R. equi* with VapA and 103- (B) is a VapA deficient mutant adapted from (Haubenthal et al., 2023).

93. *R. equi* is a facultative intracellular bacterium that preferentially replicates within macrophages. During infection, the bacterium is engulfed by macrophages and subsequently confined within vacuoles called phagosomes. Its virulence is largely mediated by VapA, which is an immunodominant protein (Jain et al., 2003; Meijer and Prescott, 2004). High levels of VapA expression are triggered in *R. equi* by temperatures around 37°C (Figure 6. B), which is presumably adaptive to mammalian hosts (Haubenthal et al., 2023).

94. Once present in sufficient concentration, VapA facilitates *R. equi* infection of its host by reducing the fusion of *R. equi*-containing phagosome with lysosomes or by accumulating in the host phagolysosome and neutralising its pH (von Bargen et al., 2019). This in turn, allows the bacterium to escape intracellular degradation and persist within macrophages, contributing to the overall disease state (Rofe et al., 2017). The exact mechanisms which mediate the function of VapA are still unclear although it is thought to involve the exclusion of proton-pumping vacuolar-ATP from phagosomes, permeabilising their membranes (von Bargen et al., 2019). It has however been

demonstrated, that the presence of VapA alone does not allow generalist bacteria to overcome phagocytotic immune defences (Haubenthal et al., 2023), with only *R. equi* and very closely related bacteria able to utilise the effects of this protein to gain virulence. However, there has been one report of non-pathogenic *Escherichia coli* being able to persist intracellularly with supplementation of VapA (Wright et al., 2018). In this way, VapA has not been demonstrated to be a toxin in mammals independent of *R. equi* or closely related bacteria (da Silveira et al., 2024).

95. The proposed GMO has been designed to express VapA under the CMV promoter in order to generate an immune response against it in vaccinated foals with the goal of providing immunity against *R. equi*.

3.3 Biodistribution and shedding

96. Biodistribution and shedding studies determine the location of a product after administration to a person or animal. For example, the GMO may travel from the administration site to other tissues or organs (biodistribution) or be excreted in body fluids or faeces (shed). As this is the first trial proposed with this GMO, its biodistribution and shedding has not been evaluated. It can be anticipated that the GM vaccine is likely to have a similar biodistribution and shedding as other HAdV vector-based vaccines and gene therapies. Studies conducted in animal models and humans suggest that biodistribution and shedding of replication defective adenoviral vectors depend on the dose and administration route.

3.3.1 I.M. administration of adenoviral vectors

97. Intramuscular administration of a single dose of a replication defective vaccine, based on the Chimpanzee Adenovirus (ChAd) serotype 68, resulted in the presence of the viral vector DNA in blood of rats from 4 hours to up to 4 days after injection. Blood samples tested negative for the presence of the viral vector at day 8. Similarly, blood samples of rhesus macaques that received 3 doses of the ChAd68-based vaccine tested negative for the viral vector DNA 7 days after the last vaccine administration (Dai et al., 2022). Analysis of biodistribution showed the presence of viral vector DNA at the administration site for up to 15 days, in spleen and stomach for up to 8 days, and in liver, prostate, colon, bone marrow, stomach, mesenteric lymph nodes, brain, kidney, lung, testis at days 1 and 2 following administration (Dai et al., 2022).

98. Viral vector DNA was not detected in blood samples of mice following immunisation with a single i.m. dose of a vaccine based on the replication defective ChAd serotype Y25. Only 1 out of 160 faecal samples tested positive for the viral vector DNA on day 2. Biodistribution analyses showed the presence of the viral vector DNA at the administration sites, in axillary lymph node, bone marrow, heart, inguinal lymph node, liver, lung, mammary gland, mesenteric lymph node, sciatic nerve and spleen samples for up to 9 days (Stebbing et al., 2022).

99. Biodistribution studies of HIV vaccines, based on HAdV-5 and HAdV-35 vectors, failed to detect viral vector DNA in blood of rabbits and mice that received a single i.m. dose of the vaccines containing $0.5 - 1 \times 10^{11}$ vp. The biodistribution of viral vectors varied depending on the animal model but viral vector DNA was primarily detected on day 9 at the administration sites (muscle), spleen, lymph nodes and liver (Sheets et al., 2008; Shimada et al., 2022). The number of viral particles decreased over time and low levels of viral DNA were detected at the administration site, spleen and lymph nodes samples collected from a small number of the rabbits (3-5 out of 10 animals) 3 months after administration.

100. In humans, the i.m. administration of a gene therapy based on a replication-defective HAdV-5 failed to induce viral shedding in blood, faeces, throat swabs or urine samples. Samples were analysed on days 2, 7 and 14 as well as 4, 8 and 12 weeks after treatment (Matyas et al., 2005 reviewed in Brandon, 2008).

3.3.2 *Inhalation and intranasal-bronchial administration of adenoviral vectors*

101. Wild type adenovirus, which presumably enters via the nasal epithelium, can be detected shedding in urine in some instances (Brandon EFA, 2008). Despite this, when GM HAdV doses of 2×10^7 infectious units (I.U.) up to 6×10^9 I.U. were administered in a volume of 100 ml into the nasal cavity of human cystic fibrosis patients, no shedding was detected in urine, sputum or rectal swabs 24 hours after administration, although viral DNA was detected in nasal swabs at the same time point (Zabner et al., 1996).

102. Other studies have demonstrated that direct administration of 1×10^9 to 9.4×10^8 pfu HAdV to the nose and lungs of cystic fibrosis patients does not lead to any detectable virus in blood samples (Brandon EFA, 2008). The presence of viral DNA in saliva up to one week, nasal swabs up to two weeks and bronchial fluid up to three weeks following viral delivery to the nasal and bronchial epithelium has been observed (Brandon EFA, 2008). Direct delivery of 1×10^9 pfu of GM AdV into the trachea of mice lead to detectable viral DNA in their lungs for up to 24 hours post administration (Brandon EFA, 2008).

103. Delivery of HAdV5 via bronchiolar lavage to patients with lung carcinoma resulted in limited detection in sputum over the first two days but no detection in urine (Tiesjema et al., 2010).

3.3.3 *Ocular administration of adenoviral vectors*

104. Ocular administration of virus would presumably share many distribution kinetic characteristics of intranasal administration considering the close proximity and connectedness of the nasal cavity and conjunctiva (Aydin et al., 2021). A study delivering HAdV-5 via ocular administration in monkeys found viral DNA in blood up to 7 days after inoculation, and in bone marrow, eye, heart, kidney, liver and spleen, but not in gonads and lungs at 6 days post inoculation (Brandon EFA, 2008). At day 29, viral DNA was still found in the liver and spleen, but not in blood and bone marrow. Ocular administration of HAdV in humans, did not produce shedding via sputum or urine when tested 3 weeks post administration (Brandon EFA, 2008).

3.3.4 *Oral administration of adenoviral vectors*

105. Oral administration would likely share many distribution kinetics with both ocular and nasal administration due to the connectedness of the nasopharyngeal space. Oral administration of replication competent AdV-based rabies vaccine (see Section 2.8) has been evaluated in target and non-target animals. Viral DNA was detected in oral and anal swabs samples collected from target animals such as raccoons (*Procyon lotor*), striped skunks (*Mephitis mephitis*) and red foxes (*Vulpes vulpes*), from 6 h up to 34 days after administration of the vaccine. Oral and anal samples of most of the Norway rats (*Rattus norvegicus*) and cotton rats (*Sigmodon hispidus*) included in the study tested positive for the presence of the viral DNA for up to 3 days and samples of 8/9 Virginian Opossum were positive for up to 9 days. Low levels of viral DNA were detected in oral swab samples of 1/10 Virginia opossums on day 23, and in 1/9 cotton rats on day 17 and in anal swab sample of 1/10 Norway rats on day 34 (Sobey et al., 2019). Administration of a 10x dose of the same AdV-based rabies vaccine resulted in shedding of viral DNA in faeces of Eastern wild turkey (up to day 3), opossum (up to day 6), eastern cottontail (up to day 5), fox squirrel and wood rat (up to day 14). Viral DNA was detected in oral swab samples collected from wild turkeys, opossums and eastern cottontails on day 4 and in samples collected from fox squirrels on day 7 (Fry et al., 2013).

106. It is important to note that most of the shedding studies reported the presence of viral vector DNA in biological samples but, with one exception, the presence of viable viral particles was not investigated (Brandon, 2008; Clayman et al., 1998). The detection of viral particles and the persistence of AdV vectors is likely to be overestimated in those studies as PCR is likely to detect fragments of viral DNA resulting from its clearance. In addition, shedding studies of the adenoviral rabies vaccine administered to non-target animals used 10 times the recommended dose of the vaccine. Further, the GMO is replication defective and viral shedding, if any, would be limited to the initial number of viral particles administered.

3.4 Host range of the GM vaccine

107. The GM vaccine is expected to infect the same range of hosts as the parent organism. As discussed in Section 2.4 of this chapter adenoviruses are generally very host specific, humans are the natural host for HAdV-5, but animals have been experimentally infected with HAdVs and adenoviral vectors. Some studies have determined that HAdV-5 vectors can successfully infect horse cells with modest efficiency, although they do not cause a high degree of cytopathic effect (Ishihara et al., 2006).

3.5 Stability in the environment and decontamination

108. The stability of GM vaccine in the environment has not been tested. However, AdVs can persist for long periods on surfaces and in water. It is expected that the survival of the GM vaccine in the environment would be similar to the parent organism. Methods of decontamination effective against the parent organism are expected to be equally effective against the GM vaccine (see Section 2.7, this chapter).

Section 4 The receiving environment

109. The receiving environment forms part of the context for assessing risks associated with dealings with GMOs (OGTR, 2013). It informs the consideration of potential exposure pathways, including the likelihood of the GMO spreading or persisting outside the site of release.

4.1 The Trial site

110. The intended primary receiving environment would be the foals which are being directly administered with the GMO via i.m injection or intranasal instillation.

111. The secondary receiving environment would be the Trial site, including the Primary facility and the Secondary facility (an outdoor paddock housing the horses) and areas used to prepare the vaccine doses.

112. The principal route by which the GMO may enter the wider environment is by shedding. As discussed in Section 3 of this Chapter, the GMO is replication defective. In the event of shedding of the GMO at the administration sites, in faeces or body fluids, the number of viral particles excreted would be limited to the initial vaccine dose.

4.2 Related viral species in the environment

113. The presence of related viruses may offer an opportunity for introduced genetic material to transfer between the GMO and other organisms in the receiving environment.

114. Horses are known to be infected by two adenoviruses, equine adenovirus 1 (EAdV-1) and equine adenovirus 2 (EAdV-2), that are associated with respiratory disease and intestinal disease, respectively. The genomes of EAdV-1 and EAdV-2 have been completely sequenced, and both have 69% sequence identity to HAdV-5 (Cavanagh et al., 2012; Giles et al., 2015).

115. AdVs have been reported in Australian bearded dragons (*Pogona* spp) and native birds, including rainbow lorikeets (*Trichoglossus haematodus*), galahs (*Eolophus roseicapilla*), and sulphur-crested cockatoos (*Cacatua galerita*); and in brushtail possums (*Trichosurus vulpecula*) from New Zealand (Hyndman et al., 2019; Vaz et al., 2020). It can therefore be anticipated that non-human AdVs are likely be present in the environment.

116. Insects, ticks and leeches are present in the Queensland environment. These animals are not known to transmit AdVs but can be exposed to the virus while feeding on an infected animal. DNA of AdV Species C has been detected in ticks collected from wildlife in Kenya (Ergunay et al., 2022). Studies conducted in leeches showed that the viral DNA can persist for up to 50 days in animals experimentally fed with human blood containing 1.6×10^6 vp/mL of HAdV. The concentration of

the viral DNA increased from day 1 to day 7 and subsequently decreased (Kampmann et al., 2017). It is important to note that both studies showed the presence of the adenoviral DNA but no viable particles of the virus. The increase of viral DNA concentration observed in leeches could result from viral replication in the remaining human blood or experimental variability. For example, individual leeches could have ingested different amounts of blood during the feeding experiment.

117. The prevalence of HAdVs in Australia based on the reported cases and seroprevalence is low as mentioned in Section 3.4.3.

118. In addition, the COVID-19 vaccine Vaxzevria approved for use in Australia during the pandemic, was based on a Chimpanzee adenoviral vector (MHRA, 2020). Other AdV vector-based gene therapies and vaccines have been evaluated in clinical trials. Therefore, AdV vectors could be present in people or the environment.

Section 5 Previous authorisations

119. The proposed GMO is authorised to be manufactured under a NLRD. The Regulator has not previously assessed or approved any DIR or DNIR licences for dealings with the proposed GM vaccine.

120. However, the Regulator has issued commercial and limited and controlled DIR licences ([DIR-180](#), [DIR-182](#), [DIR-184](#), and [DIR-195](#)) utilising replication defective adenoviral vector-based vaccines for humans and animals.

121. In addition, the Regulator has issued several DNIR licences (DNIR-588, DNIR-599, DNIR-606, DNIR-609, DNIR-636, DNIR-637), authorising clinical trials of replication defective adenoviral vector-based vaccines and gene therapies in Humans.

Chapter 2 Risk assessment

Section 1 Introduction

122. The risk assessment identifies and characterises risks to the health and safety of people or to the environment from dealings with GMOs, posed by or as the result of gene technology (Figure 7). Risks are identified within the established risk assessment context (Chapter 1), taking into account current scientific and technical knowledge. A consideration of uncertainty, in particular knowledge gaps, occurs throughout the risk assessment process.

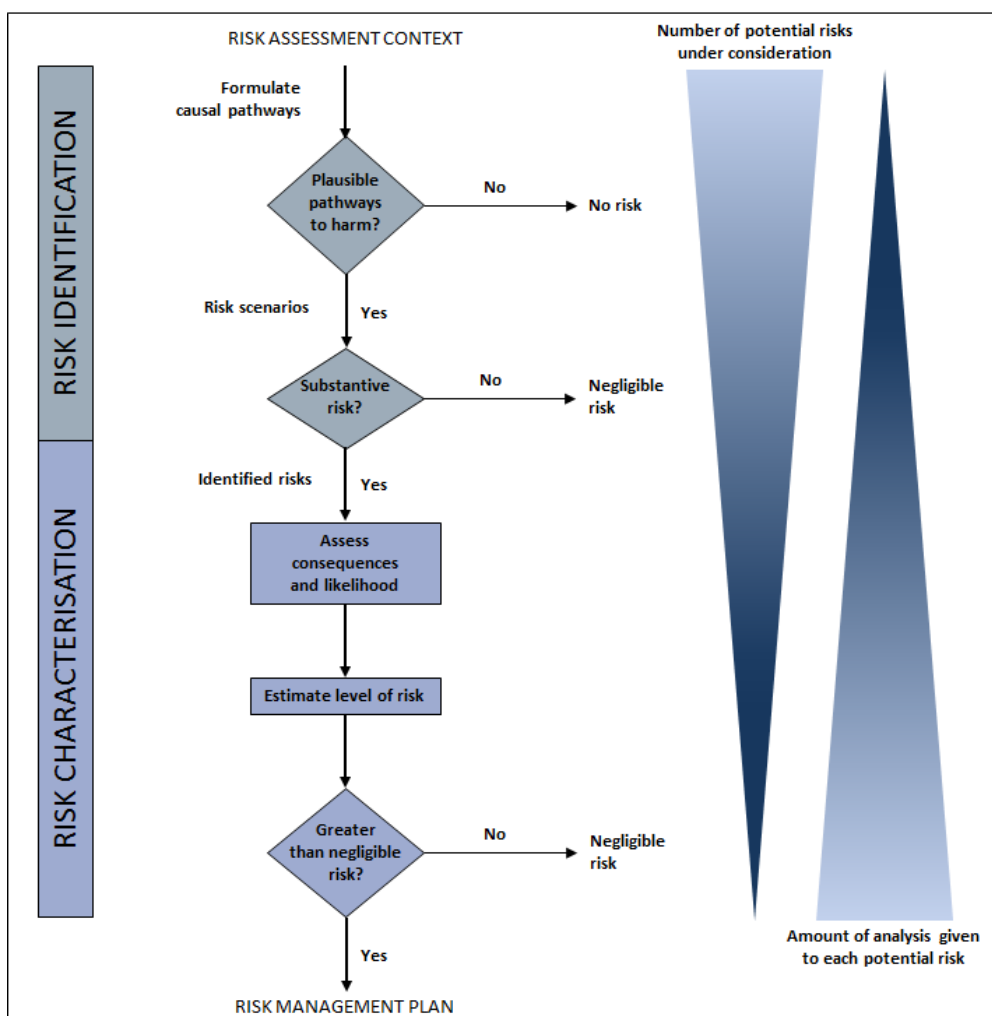


Figure 7. The risk assessment process

123. The Regulator uses a number of techniques to identify risks, including checklists, brainstorming, reported international experience and consultation (OGTR, 2013).

124. Risk scenarios examined in RARMPs prepared for licence applications for the same or similar GMOs, are also considered.

125. Risk identification first considers a wide range of circumstances in which the GMO, or the introduced genetic material, could come into contact with people or the environment. This leads to postulating plausible causal pathways that may give rise to harm for people or the environment from dealings with a GMO. These are called risk scenarios.

126. Risk scenarios are screened to identify substantive risks, which are risk scenarios that are considered to have some reasonable chance of causing harm. Risk scenarios that could not

plausibly occur, or do not lead to harm in the short and long term, do not advance in the risk assessment process (Figure 7), i.e. the risk is considered to be no greater than negligible.

127. Risk scenarios identified as substantive risks are further characterised in terms of the potential seriousness of harm (consequence assessment) and the likelihood of harm (likelihood assessment). The consequence and likelihood assessments are combined to estimate the level of risk and determine whether risk treatment measures are required. The potential for interactions between risks is also considered.

Section 2 Risk identification

128. Postulated risk scenarios are comprised of three components (Figure 8):

- i. the source of potential harm (risk source)
- ii. a plausible causal linkage to potential harm (causal pathway)
- iii. potential harm to people or the environment.

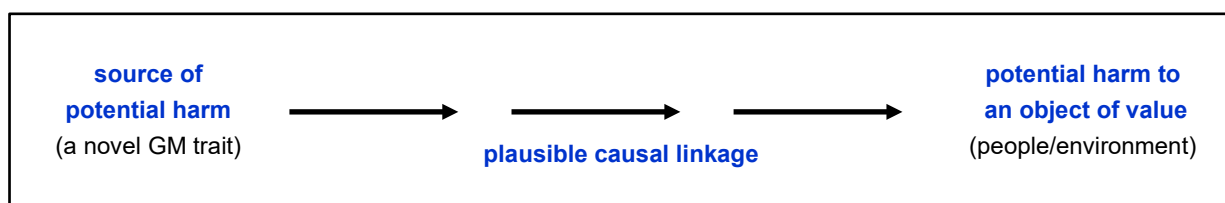


Figure 8. Components of a risk scenario

129. When postulating relevant risk scenarios, the risk context is taken into account, including the following factors detailed in Chapter 1:

- the proposed dealings
- the proposed limits including the extent and scale of the proposed dealings
- the proposed controls to limit the spread and persistence of the GMO and
- the characteristics of the parent organism(s).

2.1 Risk source

130. The parent organism of the GMO is the human AdV serotypes 5 (HAdV-5). Details on the pathogenicity and transmissibility of HAdV-5 are discussed in Chapter 1, Section 2.3. Infection is generally the result of inhalation of aerosolised droplets arising from respiratory, or ocular secretions containing the virus. It can also occur via faecal-oral transmission. When infecting humans, HAdV-5 may cause common cold-like symptoms, as well as eye infections or diarrhoea.

131. Infection with AdV can result in latent infection in lymphoid tissues and increase the period of viral persistence in the body. However, AdV largely remains episomal throughout latent infection and seldom integrates into a host cell genome. The low rate of integration appears to be random and does not appear to cause tumorigenesis in humans and therefore will not be further discussed.

132. As discussed in Chapter 1, the GM adenovirus has been modified by the deletion of E1 and E3 regions which are intended to enhance the safety profile of the GMO and the introduction of an expression cassette containing the VapA transgene from *R. equi*. The inserted transgene is intended to illicit an immune response in young horses and is considered further as a source of potential harm.

133. The expression of the introduced gene is controlled by adenoviral regulatory sequences. Regulatory sequences are naturally present in all organisms and the introduced/endogenous sequences are expected to operate in similar ways to endogenous sequences. The regulatory sequences are DNA that is not expressed as a protein; they are adenovirus specific and do not present a risk in the absence of adenovirus cellular machinery. Hence, potential harms from the regulatory sequences will not be further assessed for this application.

2.2 Causal pathway

134. The following factors are taken into account when postulating plausible causal pathways to potential harm:

- the proposed dealings
- the proposed limits including extent and scale of the proposed dealings
- the proposed controls to limit the spread and persistence of the GMOs
- characteristics of the parent organism
- routes of exposure to the GMOs, the introduced gene(s) and gene product(s)
- potential effects of the introduced gene(s) and gene product(s) on the properties of the organism
- potential exposure of other organisms to the GMOs in the environment
- the environment at the site(s) of release
- spread and persistence of the GMOs (e.g. dispersal pathways and establishment potential)
- environmental stability of the organism (tolerance to temperature, UV irradiation and humidity)
- gene transfer by horizontal gene transfer (HGT)
- unauthorised activities, and
- practices during and after administration of the GMOs.

135. Although all of these factors are taken into account, some are not included in the risk scenarios below as they may have been considered in previous RARMPs and a plausible pathway to harm could not be identified.

136. As discussed in Chapter 1 Section 1.1, the APVMA assesses the quality, safety and efficacy of the vaccine. The APVMA may also impose conditions on a permit for the supply of veterinary vaccines for research purposes.

137. Previous RARMPs have considered the potential for unauthorised activities to lead to an adverse outcome. The Act provides for substantial penalties for non-compliance and unauthorised dealings with GMOs. The Act also requires the Regulator to have regard to the suitability of the applicant to hold a licence prior to the issuing of the licence. These legislative provisions are considered sufficient to minimise risks from unauthorised activities, and no risk greater than negligible was identified in previous RARMPs. Therefore, unauthorised activities will not be considered further.

2.3 Potential harm

138. The following factors are taken into account when postulating relevant risk scenarios for this licence application:

- harm to the health of people or desirable organisms, including disease in humans or other animals or adverse immune response to the GM vaccine
- the potential for establishment of the GMO that could cause harm to people or the environment harm to the health of people or desirable organisms, including toxicity/allergenicity

2.4 Postulated risk scenarios

139. Three risk scenarios were postulated and screened to identify substantive risk. These hypothetical scenarios are summarised in Table 1 and discussed in depth in Section 2 (this chapter).

Table 1. Summary of hypothetical risk scenarios from dealings with GM vaccine

Risk scenario	Risk source	Possible causal pathway	Potential harm	Substantive risk	Reason
1	GMO	<p>Exposure of people undertaking dealings with the GMO via: needle-stick injury, aerosols, fomites, contact with broken skin or mucous membranes during</p> <p>(a) Preparation and administration of the GMO</p> <p>(b) Transporting the GMO</p> <p>(c) Disposing of the GMO including decontamination</p> <p>(d) Collection and analysis of biological samples</p> <p>(e) Animal bites or scratches</p> <p style="text-align: center;">↓</p> <p>Transduction of cells by GMO</p> <p style="text-align: center;">↓</p> <p>Expression of VapA</p>	<p>Local inflammation, flu-like symptoms, or eye infection</p> <p>Illness associated with over-expression of VapA</p>	No	<ul style="list-style-type: none"> • Only veterinarians would administer the GMO. Procedures would not require removing or recapping the needles. • Only trained and experienced personnel would handle the GMO or access the trial site. • Staff handling the GMO or collecting biological samples would wear appropriate PPE. • The GMO is replication defective. • The dose received through accidental exposure would be far smaller than that administered during vaccination/treatment based on adenoviral vectors. • VapA is not an intrinsically toxic protein and has not been demonstrated to cause disease in humans despite being ubiquitous in soil. • Any reactions to VapA would be transient and the GMO would be rapidly cleared by the immune system. • Horses involved in the study would be trained in the procedures beforehand and would be less likely to bite and scratch. • Staff conducting the dealings would be highly trained in animal husbandry.
2	GMO	<p>Administration of the GMO into young horses</p> <p style="text-align: center;">↓</p>	As per Risk Scenario 1.	No	<ul style="list-style-type: none"> • The GMO is replication defective, and shedding would be limited to the

Risk scenario	Risk source	Possible causal pathway	Potential harm	Substantive risk	Reason
		GMO is shed at the injection sites or via oral fluids or faeces ↓ Exposure of people or non-target animals to the GMO through contact with inoculated horses or GMO-contaminated material ↓ Transduction of cells by GMO ↓ Expression of VapA			number of viral particles administered. <ul style="list-style-type: none"> • Following administration, the horses are kept in an enclosed primary facility until a negative nasal swab PCR test for the GMO. • Excretions from the horses would be regularly decontaminated • Humans are the natural hosts for HAdVs. Exposure of other animals to the GMO is unlikely to cause disease. • As per risk scenario 1, if an animal is exposed to the GMO, the expression VapA, and any adverse reaction would be transient.
3	GMO	Administration of the GMO into young horses ↓ a) The horse is coinfecting with another AdV; or b) An animal or person is exposed to the GMO while infected with another AdV ↓ GMO transduces a host cell co-infected with another AdV ↓ (a) Complementation of E1 and E3 by AdV (b) Homologous recombination with AdV ↓ Production of novel recombinant AdVs	Local inflammation, flu-like symptoms or eye infection Disease in people or animals	No	<ul style="list-style-type: none"> • As per RS 1 and 2, human exposure to an infectious dose of the GMO is highly unlikely. • It is highly unlikely that both GMO and AdV would coinfect the same cell at the same time. • Recombination among AdVs is usually restricted to the same species. • Homologous recombination in AdV-C is more likely to occur in E1 and E4 regions, which are not involved in virus tropism. • Multiple recombination events would be required to produce a replication competent AdV with altered tropism and immune evasion properties.

2.4.1 Risk scenario 1

Risk source	GMO
Causal pathway	<p>Exposure of people undertaking dealings with the GMO via needle-stick injury, aerosols, fomites, contact with broken skin or mucous membranes during:</p> <p style="text-align: center;">↓</p> <ul style="list-style-type: none"> (a) Preparation and administration of the GMO (b) Collection and analysis of biological samples (c) Transporting the GMO (d) Disposing of the GMO including decontamination (e) Animal bites or scratches <p style="text-align: center;">↓</p> <p style="text-align: center;">Transduction of cells by GMO</p> <p style="text-align: center;">↓</p> <p style="text-align: center;">Expression of VapA</p>
Potential harm	<p>Local inflammation, flu-like symptoms, or eye infection Illness associated with over-expression of VapA</p>

Risk source

140. The source of potential harm for this postulated risk scenario is the GM vaccine.

Causal Pathway

141. There are a number of ways that people may be exposed to the GMOs while undertaking the dealings as part of this trial.

Exposure during preparation and administration of the GMO

142. The GMO would be prepared at the trial sites and administered via i.m. or intranasal administration. There is potential for exposure of people to the GMO during the preparation and administration via needle stick, sharps injury, aerosol formation, spills or eye splash.

143. Needles would only be used during the administration of the GMO to the foals via i.m. injection; this procedure would not require recapping the needles. Only veterinarians or those highly trained in animal husbandry and veterinary procedure would administer the GMO and needles and syringes would be disposed into sharps bins immediately after use. The foals would be trained in advance of the procedure and any horses which did not acclimatise well would be excluded from the trial. The horses would also be tethered to fencing during the administration procedure, limiting the magnitude of unexpected or sudden movements.

144. These measures would minimise the potential exposure of people to the GMOs via needle stick, sharps injury or horse bites. In the event of exposure, the person would be instructed to wash the exposed area with excess of water and soap or antiseptic and seek medical attention if required. In addition, as AdVs usually cause respiratory, gastrointestinal or eye infections, it is unlikely that exposure via needle stick or sharps injury would result in infection.

145. Dealings generating aerosols or accidental spills during preparation or administration could result in exposure via inhalation of aerosols or exposure through the oral route. However, those administering the GM vaccine would be wearing extensive PPE including masks, face shields, double gloves, disposable overalls with hoods and knee-high rubber boots. This would minimise the potential exposure of people to the GMOs via aerosol, spills or eye splash. Contact with broken skin is not a route of AdV infection, but it could result in exposure to the GMO. The applicant proposed that, in the unlikely event of exposure of skin, eye or mucosa to the GMO, the affected area would be decontaminated where possible, and the person would be instructed to seek medical advice. The attending physician would be informed of the GMO and its characteristics. In addition, any spills on the floor or surfaces would be decontaminated using chemical disinfectants.

Transport and disposal of the GMO

146. Staff involved in conducting the trial would be required to transport the GMO from a PC2 laboratory to the trial site. The GMO would be contained in a screw cap hard plastic vial, the vial would be contained within a plastic bag, and the plastic bag would be placed in an unbreakable hard plastic container. The three redundant levels of containment would prevent exposure of staff handling the container to the GMO in the event of an unexpected mechanical shock to the package. Alternatively, staff who are cleaning or decontaminating the GMO or GMO contaminated areas of the trial site may be exposed to the GMO via splashing. All staff involved in decontamination would be required to wear PPE including waterproof boots, disposable overalls, eye protection, and gloves. These measures would prevent direct exposure of staff to the GMO in the event of splashes.

Collection and analyses of biological samples

147. Personnel entering the animal enclosures would wear waterproof footwear, disposable overalls including a hood, P2 masks, face shields, and double gloves. All non-reusable PPE would be decontaminated after use. Blood, nasal and rectal temperatures would be collected from young horses at several timepoints. Needles would be used for the collection of blood samples by veterinarians or staff experienced in handling horses using standard procedures.

148. Samples would be analysed in PC2 laboratory facilities at UQ. Analysis of samples would be conducted using standard PC2 laboratory under an NLRD (see Chapter 1, Section 2).

Potential harm

149. If people are exposed to the GMO, they could develop flu-like symptoms, eye infections or local inflammation for a short period of time before the virus is cleared by the immune system. They could also develop a strong immune response against VapA which is expressed by the GMO.

150. As the GMO is replication incompetent, it is unable to produce further viral particles which are required to sustain an infection. This would also mean that the VapA protein would be unable to accumulate and only persist until the point of clearance of the GMO, limiting the magnitude of any immune response.

151. *R. equi* and therefore VapA and several homologous bacterial proteins from closely related species, are ubiquitous in soil, with especially high volume in areas containing ruminant manure. Due to their presence in dust and soil, most people would likely have been exposed to VapA continuously throughout their lives. Despite this, there have been no reports of VapA associated hyper immune response in humans. This suggests that despite being an immunodominant protein, VapA is not a potent allergen to humans. Furthermore, VapA does not function as a toxin despite conferring virulence to *R. equi*. This is because its virulence is not derived from cytotoxicity, but rather from facilitating host colonisation/infection by the *R. equi*. In the absence of *R. equi* VapA likely does not cause significant harm to humans or animals. Although there is some evidence that VapA can enable the persistence of non-pathogenic *E. coli*, in order for this effect to occur *in vivo*, the GMO would have to express the protein in macrophages, which subsequently would have to be

involved in engulfing the bacteria in question. These events in combination would be unlikely to occur.

152. VapA is derived from *R. equi* and its ability to induce an immune response against a protein produced by human or animal cells has not been evaluated. However, there is likely a high level of exposure that many people and animals would have to VapA, as well as the VapAs immunodominant characteristics. To date, there is no report of antibodies against VapA cross-reacting with human proteins or with proteins expressed by microorganism that are beneficial to humans (e.g. microorganism present in the human microbiota). Therefore, it is unlikely that an immune response generated against these antigens would result in an adverse effect in humans.

153. Immunocompromised individuals are a high-risk group for development of severe disease following HAdV infection. However, the GMO cannot replicate or cause disease. In the event of exposure via needle stick/sharps injury, horse bites or mucosa/broken skin, the volume and hence the amount of GMO transferred would be far smaller than that administered during vaccination/treatment based on adenoviral vectors. For example, individuals immunised with Vaxzevria, an adenoviral Covid-19 vaccine received of 5×10^{10} viral particles per dose, corresponding to not less than 2.5×10^8 infectious units and serious side effects were rarely observed (Chapter1, Section 4). WHO recommends that immunocompromised individuals should receive an additional dose of Vaxzevria, as these individuals are less likely to produce an efficient immune response against the COVID19 antigen following vaccination (WHO, 2022). It can be anticipated that the exposure of immunocompromised individuals to a small amount of virus would not induce a robust immune response against VapA.

154. Should severe adverse events occur, antiviral drugs such as Cidofovir and Ribavirin may be used as treatment to adenoviral infections. Therefore, it is highly unlikely that the GMO will cause serious harm to either healthy or immunocompromised individuals.

155. Further mitigating factors are the inability of a replication incompetent virus to complete its infection cycle and disseminate to new cells, and the likely small dose of GMO which would be delivered through accidental exposure. Taken together, these factors would also mean that even if some symptoms resulted from transient infection, these would be mild.

156. **Conclusion**

157. The potential for an unintentional exposure of people to the GMO resulting in local inflammation, flu-like symptoms, eye infection or serious adverse immune reaction in humans is not identified as a risk that could be greater than negligible. Therefore, it does not warrant further detailed assessment.

2.4.2 Risk scenario 2

Risk source	GMO
Causal pathway	<p style="text-align: center;">Administration of the GMO into foals</p> <p style="text-align: center;">↓</p> <p style="text-align: center;">GMO is shed at the injection sites, or via oral fluids or faeces</p> <p style="text-align: center;">↓</p> <p style="text-align: center;">Exposure of animals to the GMO through contact with inoculated horses or GMO-contaminated material</p> <p style="text-align: center;">↓</p> <p style="text-align: center;">Transduction of cells by GMO</p> <p style="text-align: center;">↓</p> <p style="text-align: center;">Expression of VapA</p>
Potential harm	Local inflammation, flu-like symptoms, or eye infection; Illness associated with over-expression of VapA

Risk source

158. The source of potential harm for this postulated risk scenario is the GM vaccine.

Causal Pathway

159. The principal route by which the GMO may enter the wider environment following vaccination is via shedding. People or animals could be exposed to the GMO by coming into contact with vaccinated foals or GMO-contaminated material. Risk scenario 1 explores the likelihood and consequences of people being exposed to the GMO. The likelihood and consequence of exposure to animals is the focus of this risk scenario.

Shedding of the GMO within the enclosures

160. Shedding data available for vaccines and gene therapies based on AdV vectors suggest that the i.m. administration of the GMO is unlikely to result in shedding. As highlighted in section 3.3, shedding of adenoviral vectors has not been observed in urine or faecal swabs following nasal administration. Some shedding has been observed following nasal administration in sputum and nasal swabs within the first 2 weeks. Similar results have been observed following ocular administration, with no shedding in urine or sputum 3 weeks following administration. Overall, there are limited data regarding nasal administration of replication-defective adenoviral vectors. Therefore, data available from oral administration of replication competent AdV-based vaccine are used here as a comparative. Shedding data for the replication competent AdV-based rabies vaccine suggest that the direct instillation of the GM vaccine into the oral cavity is likely to result in shedding in oral fluids and faeces for the first two weeks. Oral administration would likely share many distribution kinetics with both ocular and nasal administration due to the connectedness of the nasopharyngeal space. However, as the GMO cannot replicate, shedding would be limited to the initial number of viral particles administered. As mentioned, most shedding studies focused on the presence of viral DNA in biological samples but not viable viral particles. PCR based assays used to detect viral DNA are very sensitive and able to detect small fragments of DNA. The shed GMOs detected in biological samples would consist of viral DNA in incomplete or non-infectious particles. The number of viable viral particles shed would necessarily be smaller than the dose of the GMO initially administered, and their persistence would be limited.

161. Following intranasal instillation, the GMO could be shed in faeces and saliva. The presence of GMO in urine is plausible but unlikely (Brandon, 2008). As discussed in Section 2.7 (Chapter 1), AdVs can persist in the environment for weeks or months and could be shed into the soil within the facility.

Exposure to the GMO in the wider environment

162. The applicant proposed that horses will be kept in the primary contained facility for at least 2 weeks following the last administration of the GMO and contact with other animals is unlikely. Horses would be tested for the presence of the GMO in nasal swab samples and may be relocated to the secondary facility if the GMO is not detected.

163. Faeces, urine and saliva produced within the primary facility will be washed from the holding pens and decontaminated prior to discharged into the facility's settlement ponds. Exposure to the GMO in the wider environment within 2 weeks following administration of the GMO is unlikely.

164. In the secondary facility, as the horses would be housed in open air, small animals such as possums, bats, rodents or birds could enter the enclosure. Alternatively, a vaccinated horse could escape the facility and come into contact with people or animals in the environment. The fencing around the secondary facility and the broader trial site would make this outcome highly unlikely.

165. As mentioned previously, i.m. administration of the GMO is unlikely to result in shedding, and shedding is expected to be minimal 2 weeks after intranasal instillation of the GMO. If shedding occurs, the animals mentioned above could be exposed to the GMO via ingestion of food, soil or drinking water contaminated with small doses of the GMO shed by a vaccinated horse. A wild animal exposed in this way could then act as a vector and spread the GMO into the wider environment by the oral-faecal route. However, any animal entering the enclosure would only be exposed to a small dose of the GMO, which is a human specific virus, this would be highly unlikely to trigger an infection. In addition, as the GMO is replication incompetent it would be unable to disseminate from an infected animal to another. It can be anticipated that the shedding of the GMO by exposed birds or other animals would be far smaller than the dose of the GMO initially administered to the foals. This further reduces the likelihood of exposure of other animals to the GMO.

166. Among the species which may be able come into direct contact with GMO-vaccinated foals are birds. Adenoviral infections in birds are caused by AdV species belonging to the *Aviadenovirus* genus and natural infection of birds by human adenoviruses does not occur. It has been demonstrated in laboratory conditions that intranasal administration of vaccines vectored by HAdV-5 results in poor seroconversion, a result that likely reflects the limited ability of the HAdV-5 to infect the respiratory tract cells of birds (Gao et al., 2006).

167. In the highly unlikely event that the GMO transduces the cells of birds or other animals, it could lead to expression of the VapA protein and an allergic reaction to the VapA protein. Given the current prevalence of this protein in the environment due to the ubiquitous nature of *R. equi*, animals are likely to have been repeatedly exposed to it already. As no allergic reactions to the bacteria itself have been documented, it is unlikely that allergic reactions to the VapA protein would emerge. Further, HAdVs are not known to infect birds or other animals in nature and no adverse effects have been observed in animals exposed to a replication-competent HAdV-5 vaccines. Therefore, it is unlikely that exposure to GMO would result in adverse effects or disease in birds or other animals.

168. As discussed in Section 4 of Chapter 1, the GMO could be present in the blood of foals for up to 4 days following injection. Insects, ticks or leeches feeding on the horses could be exposed to the GMO. Although adenoviral DNA has been isolated from ticks and leeches, there is no evidence that they could transmit the virus while feeding on other animals. Studies conducted in mice showed the HAdV-5 vector has a half-life in blood of less than 2 min when administered intravenously (Alemany et al., 2000). In the event of transfer of the GMO to ticks and leeches via blood, it is unlikely that the GMO persists long enough to infect the next animal that the ticks or leeches feed on. The ingestion of GMO-exposed tick/leech by other animals (e.g. birds), could result in the presence of a small amount of GMO in the gastrointestinal tract. As discussed previously, it is unlikely that this could result in adverse effects or disease in birds or other animals.

Dissemination of the GMO into the soil and contamination of waterways

169. As mentioned, horses will be kept in the primary contained facility for at least 2 weeks after the last administration of the GMO. All faeces, urine and saliva produced within this facility will be decontaminated during this time. Minimum shedding is expected 2 weeks following administration, when animals would potentially be relocated to the secondary facility.

170. If shedding occurs and in the event of rain, the GMO shed within the secondary facility could disseminate into the soil and eventually disperse into waterways. Based on the parent organism's ability to survive in the environment, the GMO could persist in water for weeks (see Section 2.7, Chapter 1). However, due to its replication incompetence, the GMO would be unable to maintain itself in a reservoir host and therefore exposure to the environment for long periods would eventually degrade it. In addition, any GMO contaminating waterways in this way would be enormously diluted, minimising the potential for exposure of animals in the environment through ingestion of contaminated water. As discussed in Chapter 1, Section 2.4., AdVs can infect a broad range of hosts including reptiles and fish, but humans are the natural hosts for HAdV. Furthermore, adenoviral infections in fish are rare and caused by a single species of AdV belonging to the *Ichtadenovirus* genus (Harrach et al., 2019). Therefore, the likelihood of infection of animals following exposure to a GMO contaminated water source is highly unlikely.

Potential harm

171. As the GMO is replication incompetent, it is expected that shedding of viable particles of the GMO would be limited to a dose significantly lower than the initial dose administered. WT HAdV cannot replicate or cause disease in animals. In the event of exposure of an animal to the GMO, it is likely that the presence of the GMO would be transient, and it would be cleared by the host immune system. Similar effects would be expected in the event of further transmission to another animal. It is highly unlikely that the GMO would infect or cause disease via ingestion of contaminated water, in reptiles or fish.

172. HAdV-based vaccines have been shown to be safe in animals. However, the effects of the VapA protein in horses has not been evaluated, although there are a small amount of data for the GMO expressing VapA in mice. As previously mentioned, VapA is abundant in the environment due to the ubiquitous nature of *R. equi*. Therefore, it is unlikely that exposure of animals to VapA via the GMO would result in a hyper immune response.

Conclusion

173. The potential of the GMO to be released into the environment and result in adverse immune reactions or disease in other animals is not identified as a risk that could be greater than negligible. Therefore, this scenario does not warrant further assessment.

2.4.3 Risk scenario 3

Risk source	GMO
Causal pathway	<p style="text-align: center;">Administration of the GMO into horses</p> <p style="text-align: center;">↓</p> <p style="text-align: center;">a) The horse is coinfectd with an EAdV; or</p> <p style="text-align: center;">b) An animal or person is exposed to the GMO while infected with another AdV</p> <p style="text-align: center;">↓</p> <p style="text-align: center;">GMO transduce a host cell co-infected with another AdV</p> <div style="display: flex; justify-content: space-around;"> <div style="text-align: center;"> <p>↙</p> <p>Complementation of E1 and E3 by AdV</p> <p>↓</p> <p>Production of more replication incompetent GMOs</p> </div> <div style="text-align: center;"> <p>↘</p> <p>Homologous recombination with AdV in E1, E3 or other regions of high homology</p> <p>↓</p> <p>(i) Formation of replication defective AdV expressing VapA</p> <p style="text-align: center;">AND</p> <p>Replication competent GMO without VapA expression cassette</p> <p style="text-align: center;">OR</p> <p>(ii) Replication competent AdV with defective immune evasion properties (E3)</p> <p style="text-align: center;">AND</p> <p>Replication incompetent GMO with immune evasion properties (E3)</p> <p style="text-align: center;">OR</p> <p>(iii) AdV or GMO with altered tropism</p> </div> </div>
Potential harm	Novel disease in people or animals

Risk source

The source of potential harm for this postulated risk scenario is the GMO.

Causal Pathway

174. The transmission of the GMO can occur via the pathways mentioned in Risk Scenario 1 and 2 potentially resulting in transduction of host cells. If a horse, animal or a person exposed to the GMO has an existing AdV infection at the time of exposure or acquires an AdV infection while the GMO is present, this co-infection could potentially result in complementation and/or recombination of the GMO with WT AdVs and cause an adverse immune reaction and/or disease in people or animals. However, for the complementation or recombination to occur the GMO and a WT AdV must co-infect the same cell at the same time during the same replication phase. Given the GMO is replication incompetent and cannot initiate its own replication cycle, the propensity for it to recombine would be further reduced.

175. Additionally, the WT adenovirus would have to share a high level of homology between the regions capable of recombining. The most likely pathway for a WT adenovirus coming into contact with the GMO would be in the horses which are being vaccinated.

Complementation of E1 and E3 by AdV in horses and other mammals

176. As discussed in Chapter 1, AdVs can infect a broad range of hosts. However, as E1 and E3 regions are exclusive to mastadenoviruses (Section 2.1, Chapter 1), complementation is only possible if the co-infection involves the GMO and another mammalian AdV. There are two such viruses which are endemic in Australian horses, EAdV-1 and EAdV-2, and are associated with mild respiratory and gastrointestinal disease respectively. Despite both viruses officially belonging to the *Mastadenovirus* genus, they are only distantly related to HAdV-5 with each having sequence homology of ~69% (Cavanagh et al., 2012; Giles et al., 2015). EAdV-2 in particular, belongs to a separate cluster within the *Mastadenovirus* genus. (Mekonnen, 2017).

177. These large genomic differences alongside the gastro-intestinal tropism of EAdV-2 would make a co-infection and subsequent complementation between it and the GMO highly unlikely. As for EAdV-1, its genomic organisation is more typical of mastadenoviruses and thus its E1 and E3 regions may have some limited propensity to complement the GMO despite have a large amount of sequential diversity. This virus also shares the same tropism as the GMO (although in different species), and the GMO planned delivery route in the case of intranasal instillation. It has been suggested that a large proportion of horses who harbour EAdV-1 have a sub-clinical presentation (Lee et al., 2022). A Korean study detected EAdV-1 nucleic acids from nasal swabs in 5 out of 359 horses tested, with only one of those horses displaying detectable symptoms (Lee et al., 2022). Little is known about the exact prevalence of EAdV-1 in Australian horses, but some evidence suggests seroprevalence could be as high as 54%. The potentially high prevalence of EAdV-1 in domestic horses, as well as its upper-respiratory tract tropism, and ability to infect sub-clinically, make the presence of the GMO and a WT EAdV1 in the same environment at the same time a plausible scenario for co-infection.

178. As per Risk Scenario 2, it is unlikely that an animal other than the horses would be exposed to the GMO shed in the faeces or oral fluids of vaccinated animals. If an animal ingests water, food or soil that has been contaminated with the GMO, it could transduce cells of the oral mucosa and/or gastrointestinal tissue. If the animal is infected with another mastadenovirus at the time of exposure or acquires an AdV infection while the GMO is present, it could result in co-infection. In the event of an exposure, the dose of GMO transferred to an animal other than the vaccinated horse is expected to be low. Therefore, it is highly unlikely that both the GMO and the AdV would co-infect the same cell at the same time to allow complementation.

179. In the unlikely event of co-infection occurring in the same cell, the E1 and E3 regions provided in *trans* could lead to replication of the GMO, resulting in subsequent transduction of neighbouring cells. However, as HAdVs have restricted ability to replicate in animals (Section 3.4, Chapter 1), the GMO is not expected to replicate efficiently in animal cells even if the E1 region is provided in *trans*. The GMO is also unlikely to persist for protracted periods of time as it expresses the immunodominant VapA antigen as well as having its immune evasion abilities deleted. This would mean the GMO would be quickly cleared from a host cell and have extremely limited opportunity to be complemented by a wild AdV.

180. Complementation of E1 and E3 by AdV in humans

181. HAdV infects over 80% of the human population (Ismail et al., 2018). Although the prevalence of HAdV infections in Australia is expected to be low (Section 3, Chapter 1), it is plausible that the E1 and E3 genes could be provided in *trans* from a pre-existing or acquired HAdV infection in persons accidentally exposed to the GMO if a co-infection in the same cell occurs. This could result in complementation by the HAdV leading to replication of the GMOs.

182. As discussed in Risk Scenario 1, the exposure of people undertaking dealing with the GMO is unlikely due to work practices that people conducting the dealings would follow. As AdVs are prevalent in respiratory, gastrointestinal or ocular tissue, it is unlikely that viral particles would be present in subcutaneous/skin cells in the case of a needle stick injury or contact with abraded skin. In the event of exposure of people to the GMO via aerosols or contact with mucous membranes, it is possible that a GMO could transduce a cell already infected with a wild HAdV. However, as mentioned above, the GMO is highly vulnerable to clearance by the host immune system and unable to initiate its own replication. Therefore, if a single cell co-infection is established, complementation would only result in the production of replication defective virions during the period of the co-infection. The modifications to the GMO would make them reliant on the wild AdV to initiate their release from the host cell, and following this, they may enter another host cell but would be unable to replicate and proliferate further unless that cell also was infected with a wild type AdV. This reliance on the wild type virus for each round of replication and dissemination in conjunction with the GMOs vulnerability to immune attack would naturally limit the GMOs proliferation through a host even with complementation of another virus.

Homologous recombination with AdV

183. Recombination is common among circulating WT AdVs in nature. It is a key driver for evolution of viruses in general but is particularly so for AdV as the mutation rate of their dsDNA genome is lower than that of other viruses (Risso-Ballester et al., 2016). Similar to complementation, homologous recombination also requires the person or animals exposed to the GMO to be infected with a WT- AdV at the same time.

184. Following administration of the GMO, horses will be kept in the primary contained facility and contact with other animals is unlikely. In the secondary containment facility proposed by the applicant, there may exist a limited opportunity for animals outside of the trial to come into contact with the GMO. The only animals which are known to harbour adenoviruses which could plausibly gain access to the fenced areas containing the GMOs are birds, rodents and bats. Birds are perhaps the most likely to be able to contact the trial animals. There are several barriers to a co-infection of the GMO and WT-AdV in birds. In the highly unlikely event that a co-infection did occur, recombination between the two adenoviruses would be high unlikely as bird infecting AdVs (Aviadenoviruses) are a distantly related genus with little homology.

185. Recombination is more likely to occur between related viruses. HAdVs belonging to Species C, as the parent organism, show up to 99% of DNA homology (Ghebremedhin, 2014). Thus, the GMO is more likely to recombine with a HAdV-C strain than with other species of human or non-human AdV.

186. Recombination between the GMO and a human or non-human AdV strain could potentially result in the generation of different GM recombinants. These GM recombinants are described in Table 2.

Table 2. Plausible theoretical recombinants of GMO and wild-type AdVs

Recombinant region	Resultant recombinant	Outcome	Likelihood
E1	<ul style="list-style-type: none"> Replication competent GMO without E3 gene Replication incompetent AdV with VapA coding region 	<ul style="list-style-type: none"> Replication competent GMO that is still less immuno evasive than WT AdV Replication incompetent AdV expressing VapA 	Unlikely

E3	<ul style="list-style-type: none"> • Replication incompetent GMO with E3 gene • Replication competent AdV without E3 	<ul style="list-style-type: none"> • Replication incompetent GMO with modified immuno-evasive properties • Replication competent AdV without immuno-evasive properties (a WT AdV unable to evade the host immune system) 	Unlikely
Capsid genes (hexon, penton and fibre)	<ul style="list-style-type: none"> • Replication incompetent GMO with different hexon, penton or fibre. • Replication competent AdV without VapA but with different (hexon, penton or fibre) 	<ul style="list-style-type: none"> • Altered tropism and host range of GMO • Altered tropism and host range of AdV 	Highly unlikely

187. As depicted in the table above, there are several possible recombination scenarios which might result from an event including the GMO and a WT-AdV. The GMO could potentially regain its E1 region and become replication competent. It would still however, be unable to evade the host immune system and trigger its dissemination as it would not have the E3 region. It would also still express the VapA protein which would aid in host immune recognition.

188. Alternatively, the GMO could regain its E3 region and therefore its immuno-evasive properties but remain replication incompetent due to still lacking the E1 region. The recombinant virus would not be able to replicate and would eventually be cleared by the immune system of the host. As an HAdV, the recombinant virus is not expected to cause disease in birds and animals.

189. There is also a possibility that a recombination event would result in the WT AdV acquiring the transgene VapA. Should this to occur, the novel AdV would likely be very vulnerable to immune attack from its host as VapA is an immunodominant antigen. This would likely result in rapid clearance of the virus. Additionally, the applicant has indicated that bioinformatic analysis shows that the GMO genomic regions 1kb upstream and downstream of the transgene share no homology with EAdV-1, the virus most likely to co-occur with the GMO.

190. As discussed in Chapter 1, recombination in in HAdV-C, occurs most frequently in the E1 and E4 regions. The likelihood of homologous recombination at the hexon, penton and fibre regions of AdV, resulting in the GMO with an altered cell tropism is very low. In the event of recombination, the resulting AdV would remain replication incompetent.

191. As discussed in Risk Scenario 1, and earlier in this risk scenario, AdVs are prevalent in respiratory, gastrointestinal or ocular tissue and are unlikely to be commonly present in subcutaneous/skin cells in the case of a needle stick injury during administration. Furthermore, viruses typically need to both be in a replicative phase to recombine, the GMO in this case is replication incompetent. This would limit the possibility of a recombination event even if a co-infection of the same cell type did occur. Exposure to the GMO by people via inhalation or contact

with mucus membranes is plausible but the likelihood is limited by the proposed work practices including PPE and staff training.

192. In order for a full reversion into a virus with comparable virulence to the wild type, multiple recombination events would need to occur to the GMO, likely over several distinct events, restoring both the E1 and E3 regions. This is highly unlikely. If this were to occur, the resultant virus is unlikely to be more pathogenic than a wild type, as the expression of VapA would enhance the host's immune response to the virus, and aid in its clearance.

193. Finally, there is a high seroprevalence of AdVs in both humans and horses, and these two species have interacted extensively for centuries. Despite this, there have been no reports of recombinant human and horse adenoviruses to date.

Potential harm

194. If complementation were to occur, the number of replication incompetent GMOs produced in the host cells would increase, resulting in increased expression of the VapA protein in the host. The expression of the vaccine antigens in the host could induce an antibody against VapA. The potential for cross reactivity of these antigens with cells of human, birds or animals has not been investigated but is unlikely to cause harm given the abundant prevalence of VapA in the environment.

195. The temporary increase in the number of adenoviral particles could result in mild symptoms such as local inflammation, flu-like symptoms or eye infection that are expected to resolve within one week (Chapter 1, Section 3).

196. If homologous recombination were to occur, it could result in the formation of replication competent HAdV-5 in humans. The person exposed could potentially experience mild respiratory or eye infections depending on the route of exposure as described in Chapter 1, Section 2.3. These infections are self-limiting and rarely need medical intervention. If needed, antiviral therapies could be used (Chapter 1, Section 2.6.1). Theoretically, if homologous recombination in the major capsid proteins (HAdV-C) or other AdV regions with high homology occurs, it could alter the tropism and host range of the virus. However, the risks of potential increased harm are negligible as AdVs do not typically cause severe disease and the resultant recombinants are unlikely to be more pathogenic than a WT-AdV strain

Conclusion

197. The exposure of people or animals to a GMO which has acquired the E1 gene, an AdV that has acquired VapA or other recombinant viruses resulting in adverse immune response or disease in people or animals is not identified as a risk that could be greater than negligible. Therefore, it does not warrant further assessment.

Section 3 Uncertainty

198. Uncertainty is an intrinsic property of risk analysis and is present in all aspects of risk analysis. This is discussed in detail in the Regulator's [Risk Analysis Framework](#) document.

199. Uncertainty is addressed by approaches such as balance of evidence, conservative assumptions, and applying risk management measures that reduce the potential for risk scenarios involving uncertainty to lead to harm. If there is residual uncertainty that is important to estimating the level of risk, the Regulator will take this uncertainty into account in making decisions.

200. As trials are designed to gather data, there are generally data gaps when assessing the risks of a trial application involving GMOs. However, proposed trials are required to have limits and controls. Even if there is uncertainty about the characteristics of a GMO, limits and controls restrict exposure to the GMO and thus decrease the likelihood of harm.

201. For DIR 214, uncertainty is noted particularly in relation to:

- Biodistribution and shedding of the GMO. Data available for other adenoviral vector-based vaccines/therapies indicate that the GMO is likely to be shed in faeces and saliva following intranasal or i.m. administration. However, there is uncertainty as to whether the data gathered in humans or other animals would be relevant to horses.
- The effect of VapA expression in mammalian cells. As biochemical properties and mechanism of action of VapA are not well understood, there exists some uncertainty as to the effects VapA expression in animals.

202. Overall, the level of uncertainty in this risk assessment is considered low and does not impact on the overall estimate of risk. However, additional data, including information to address these uncertainties, may be required to assess possible future applications with reduced limits and controls, such as a larger scale trial or the commercial release of the GMO.

203. Chapter 3 Section 4, discusses information that may be required for future release.

Section 4 Risk evaluation

204. Risk is evaluated against the objective of protecting the health and safety of people and the environment to determine the level of concern and, subsequently, the need for controls to mitigate or reduce risk. Risk evaluation may also aid consideration of whether the proposed dealings should be authorised, need further assessment, or require collection of additional information.

205. Factors used to determine which risks need treatment may include:

- risk criteria
- level of risk
- uncertainty associated with risk characterisation
- interactions between substantive risks.

206. Three risk scenarios were postulated whereby the proposed dealings might give rise to harm to people or the environment. In the context of the limits and controls proposed by the applicant, and considering both the short and long term, none of these scenarios were identified as substantive risks. The principal reasons for these conclusions are summarised in Table 2 and include:

- the GMO has been modified to render it replication incompetent
- limited ability and opportunity for the GMOs to transfer the introduced genes through recombination
- suitability of limits and controls proposed by the applicant.

207. Therefore, risks to the health and safety of people, or the environment, from the proposed release of the GM [species] plants into the environment are considered to be negligible. The *Risk Analysis Framework* (OGTR 2013), which guides the risk assessment and risk management process, defines negligible risks as risks of no discernible concern with no present need to invoke actions for mitigation. Therefore, no additional controls are required to treat these negligible risks. Hence, the Regulator considers that the dealings involved in this proposed release do not pose a significant risk to either people or the environment.¹

¹ As none of the proposed dealings are considered to pose a significant risk to people or the environment, Section 52(2)(d)(ii) of the Act mandates a minimum period of 30 days for consultation on the RARMP.

Chapter 3 Risk management plan

Section 1 Background

208. Risk management is used to protect the health and safety of people and to protect the environment by controlling or mitigating risk. The risk management plan addresses risks evaluated as requiring treatment and considers limits and controls proposed by the applicant, as well as general risk management measures. The risk management plan informs the Regulator’s decision-making process and is given effect through licence conditions.

209. Under section 56 of the Act, the Regulator must not issue a licence unless satisfied that any risks posed by the dealings proposed to be authorised by the licence can be managed in a way that protects the health and safety of people and the environment.

210. All licences are subject to three conditions prescribed in the Act. Section 63 of the Act requires that each licence holder inform relevant people of their obligations under the licence. The other statutory conditions allow the Regulator to maintain oversight of licensed dealings: section 64 requires the licence holder to provide access to premises to OGTR inspectors and section 65 requires the licence holder to report any information about risks or unintended effects of the dealing to the Regulator on becoming aware of them. Matters related to the ongoing suitability of the licence holder must also be reported to the Regulator.

211. The licence is also subject to any conditions imposed by the Regulator. Examples of the matters to which conditions may relate are listed in section 62 of the Act. Licence conditions can be imposed to limit and control the scope of the dealings and to manage risk to people or the environment. In addition, the Regulator has extensive powers to monitor compliance with licence conditions under section 152 of the Act.

Section 2 Risk treatment measures for substantive risks

212. The risk assessment of risk scenarios listed in Chapter 2 concluded that there are negligible risks to people and the environment from the proposed trial of GM adenovirus. These risk scenarios were considered in the context of the scale of the proposed release (Chapter 1, Section 1, the proposed controls (Chapter 1, Section 1.4), and the receiving environment (Chapter 1, Section 4), and considering both the short and the long term. The risk evaluation concluded that no specific risk treatment measures are required to treat these negligible risks. Limits and controls proposed by the applicant and other general risk management measures are discussed below.

Section 3 General risk management

213. The limits and controls proposed in the application were important in establishing the context for the risk assessment and in reaching the conclusion that the risks posed to people and the environment are negligible. Therefore, to maintain the risk context, licence conditions have been imposed to limit the release to the proposed size, location and duration, and to restrict the spread and persistence of the GMOs and their genetic material in the environment. The conditions are discussed and summarised in this Chapter and listed in detail in the draft licence.

3.1 Limits and controls on the trial

214. Sections 2.1 and 2.2 in Chapter 1 list the limits and controls proposed by University of Queensland. Many of these are discussed in the risk scenarios considered in Chapter 2. The appropriateness of the limits and controls is considered further in the following sections.

3.1.1 Consideration of limits and controls

215. The proposed trial would involve a maximum of 10 young horses. Animals would be kept in contained enclosures including, holding pens and a large animal facility within Queensland Animal

Sciences Precinct at the UQ's Gatton campus. Conditions maintaining the risk context and proposed limits of the trial, such as the maximum number of animals and duration of the study, have been included in the draft licence. Although the applicant proposed that the trial will be completed within 12 months after commencement, as a standard practice, a draft licence condition require completion of the trial within 5 years.

216. Transport of the GMO, biological samples, material and waste reasonably expected to contain GMO, to and from the trial site would be conducted in accordance with the Regulator's [Guidelines for the Transport, Storage and Disposal of GMOs](#) under an NLRD.

217. Conditions are proposed in the draft licence in the event of storage and disposal within the trial site. These are also in accordance conducted in accordance with the Regulator's [Guidelines for the Transport, Storage and Disposal of GMOs](#).

218. The applicant proposed to maintain restraints on the horses during administration of the GMO. This measure would further minimise the potential of exposure via horse bites or needle stick injury during administration of the GMO and are proposed as draft licence conditions.

219. Staff preparing and administering the GMO would wear PPE including, gowns, gloves, masks and eye protection minimising the risk of exposure to the concentrated GMO via spill or aerosol formation. In addition, as discussed in Chapter 1, Section 4, the administration of the GMO via intranasal instillation or i.m. injection could result in shedding of the GMO in saliva for up to 2 weeks. There is a potential for personnel exposure to the GMO via horse bites or sneezes. Therefore, draft licence conditions have been proposed requiring that staff administering the GMO or collecting samples from the animals have experience handling horses. Conditions are also in place to ensure that staff collecting samples or administering the GMO wear PPE including face masks, eye protection, gloves and overalls.

220. Other researchers or technical staff may be present and working in the wider facility. It is therefore proposed as a licence condition, that all horses administered with the GMO be individually identifiable, and all enclosures housing the horses be signed as containing GMOs. This condition would ensure that personnel working in proximity to the site, but not involved in the study, would have the opportunity to avoid potential exposure to the GMO. To this end, it is also a condition of the licence that access to the horses is restricted to trained personnel.

221. The proposed trial design involves keeping horses in the primary facility holding pens for at least 7 days following the final administration. They would also require a negative PCR test for the GMO from nasal swabs prior to relocation to the secondary facility. This measure would limit the exposure of other animals such as rodents and birds which may have access to the secondary large animal facility to the GMO, and as such, this measure has been proposed as a draft licence condition.

222. In addition to controls mentioned above, a condition to require horse-proof fencing has been added to the licence to prevent trial animal escape and limit dissemination of the GMO to the environment via escaped horses.

223. The applicant proposed to remove and decontaminate faeces and animal waste from the holding pens within the primary facility daily in the first 14 days after GMO administration and to decontaminate areas around the administration site immediately after administration. These measures minimise the potential for dispersion of the GMO into the environment and are included in draft licence conditions. They have also proposed to collect and decontaminate the majority of faeces within the secondary facility at the conclusion of the trial. As discussed in Chapter 1, Section 4 i.m. administration of the GMO is unlikely to result in shedding. Although, the administration of the GMO via intranasal instillation could result in shedding of the GMO in saliva, nasal secretions or faeces for up to 2 weeks, minimal or no shedding is expected from animals relocated to the secondary after testing negative for the presence of GMO in nasal swab samples. Therefore, decontamination of faeces within the secondary facility is not proposed as a licence condition.

224. The applicant proposes to collect samples (blood, nasal swabs and rectal temperatures) at various time points after administration of the GMO. As mentioned in Chapter 1, Section 4, the GMO could be present in blood samples in the first four days after i.m. administration of the GMO. Nasal swab samples

are likely to contain GMO for the first two weeks after administration of the GMO via intranasal administration. As discussed in Risk Scenario 2, part of the shed GMOs detected in biological samples are likely to consist of incomplete viral DNA and not viable viral particles. However, as a precaution, the draft licence proposes conditions that apply to blood samples collected from animals up to 7 days after i.m. administration, and to any other sample, materials or waste, that are reasonably expected to contain the GMO.

225. Conditions are included in the draft licence requiring the licence holder to ensure that all GMOs, including material or waste that has been in contact with the GMO be decontaminated by autoclaving, chemical treatment or by high-temperature incineration.

226. The applicant proposed that that all GMO-inoculated horses will be euthanised and the carcasses disposed of via deep burial at the end of the trial. This is part of the trial design and not imposed as a licence condition. However, as horse meat and tissues can be traded as commodities, a licence condition has been added prohibiting the sale or distribution of trial animal tissues for these purposes. The risks to people or animals from exposure to the GMO through ingestion has not been assessed, therefore, this condition maintains the risk context.

227. A standard condition is included in the draft licence requiring the licence holder to ensure that dealings are conducted to ensure containment of the GMO so as not compromise the health and safety of people and minimise unintentional exposure to the GMO.

228. Other conditions included in the draft licence are standard conditions that state that only people authorised by the licence holder are covered by the licence, and that the licence holder must inform all people dealing with the GMOs, of applicable licence conditions.

3.1.2 **Summary of licence conditions to be implemented to limit and control the trial**

229. A number of licence conditions have been drafted to limit and control the proposed trial, based on the above considerations. These include requirements to:

- limit the trial to 10 horses, which is to be conducted in a contained trial site at the Queensland Animal Science Precinct;
- restrict access to the GMO and GMO - inoculated horses;
- ensure personnel involved in the trial are appropriately trained and follow appropriate behavioural requirements;
- ensure appropriate PPE is used;
- restrict personnel permitted to administer the GMO;
- require decontamination of the GMO, biological samples, materials and equipment that have been in contact with the GMO at the trial site using effective disinfectants or disposal methods.
- Store the GMO, samples, or other materials or waste, that are reasonably expected to contain the GMO, within the trial site, in accordance with the Regulator's [Guidelines for the Transport, Storage and Disposal of GMOs](#).

3.2 **Other risk management considerations**

230. All DIR licences issued by the Regulator contain a number of conditions that relate to general risk management. These include conditions relating to:

- applicant suitability
- contingency plans
- identification of the persons or classes of persons covered by the licence
- reporting requirements
- access for the purpose of monitoring for compliance.

3.2.1 **Applicant suitability**

231. In making a decision whether or not to issue a licence, the Regulator must have regard to the suitability of the applicant to hold a licence. Under Section 58 of the Act, matters that the Regulator must take into account include:

- any relevant convictions of the applicant
- any revocation or suspension of a relevant licence or permit held by the applicant under a law of the Commonwealth, a State or a foreign country
- the capacity of the applicant to meet the conditions of the licence.

232. If a licence were issued, the conditions would include a requirement for the licence holder to inform the Regulator of any information that would affect their suitability.

233. In addition, the applicant organisation must have access to an IBC and be an accredited organisation under the Act.

3.2.2 *Reporting requirements*

234. If issued, the licence would require the licence holder to immediately report any of the following to the Regulator:

- any additional information regarding risks to the health and safety of people or the environment associated with the dealings
- any contraventions of the licence by persons covered by the licence
- any unintended effects of the trial.

235. A number of written notices are also required under the licence regarding dealings with the GMO, to assist the Regulator in designing and implementing a monitoring program for all licensed dealings. The notices include:

- expected date of administration with the GMO for each trial site
- cease of administration with the GMO for each trial site.

3.2.3 *Monitoring for compliance*

236. The Act stipulates, as a condition of every licence, that a person who is authorised by the licence to deal with a GMO, and who is required to comply with a condition of the licence, must allow inspectors and other persons authorised by the Regulator to enter premises where a dealing is being undertaken for the purpose of monitoring or auditing the dealing.

237. If monitoring activities identify changes in the risks associated with the authorised dealings, the Regulator may also vary licence conditions, or if necessary, suspend or cancel the licence.

238. In cases of non-compliance with licence conditions, the Regulator may instigate an investigation to determine the nature and extent of non-compliance. The Act provides for criminal sanctions or large fines and/or imprisonment for failing to abide by the legislation, conditions of the licence or directions from the Regulator, especially where significant damage to the health and safety of people or the environment could result.

Section 4 Issues to be addressed for future releases

239. Additional information has been identified that may be required to assess an application for a commercial release of the GMO, or to justify a reduction in limits and controls. This includes:

- data regarding shedding of the GMO by vaccinated horses
- data regarding the safety of the VapA

Section 5 Conclusions of the consultation RARMP

240. The risk assessment concludes that the proposed trial of the GMO poses negligible risks to the health and safety of people or the environment as a result of gene technology. These negligible risks do not require specific risk treatment measures.

241. If a licence is issued, conditions would be imposed to limit the trial to the proposed scale, location and duration, and to restrict the spread and persistence of the GMOs and its genetic material in the environment, as these were important considerations in establishing the context for assessing the risks.



Chapter 4 Draft Licence conditions

Section 1 Interpretations and Definitions

1. In this licence:

- (a) unless defined otherwise in this licence, words and phrases used in this licence have the same meaning as they do in the Act and the Gene Technology Regulations 2001;
- (b) words importing a gender include every other gender;
- (c) words in the singular number include the plural and words in the plural number include the singular;
- (d) expressions used to denote persons generally (such as “person”, “party”, “someone”, “anyone”, “no one”, “one”, “another” and “whoever”), include a body politic or corporate as well as an individual;
- (e) references to any statute or other legislation (whether primary or subordinate) are a reference to a statute or other legislation of the Commonwealth of Australia as amended or replaced from time to time and equivalent provisions, if any, in corresponding State law, unless the contrary intention appears;
- (f) where a word or phrase is given a particular meaning, other grammatical forms of that word or phrase have corresponding meanings;
- (g) specific conditions prevail over general conditions to the extent of any inconsistency.

2. In this licence:

‘Act’ means the *Gene Technology Act 2000* (Commonwealth) or the corresponding State law under which this licence is issued.

‘APVMA’ means the Australian Pesticides and Veterinary Medicines Authority.

‘Decontaminate’ (or **‘Decontamination’**) means, as the case requires, kill the GMOs by one or more of the following methods:

- chemical treatment;
- autoclaving;
- high-temperature incineration; or
- a method approved in writing by the Regulator.

Note: ‘As the case requires’ has the effect that, depending on the circumstances, one or more of these techniques may not be appropriate.

‘External service provider’ means a person engaged by the licence holder solely in relation to transport, storage and/or disposal of the GMOs, and who is not undertaking any dealings with the GMOs that are not for those purposes.

‘GM’ means genetically modified.

‘GMO’ means the genetically modified organisms that are the subject of the dealings authorised by this licence.

‘NLRD’ is a Notifiable low risk dealing. Dealings conducted as an NLRD must be assessed by an institutional biosafety committee (IBC) before commencement and must comply with the requirements of the Gene Technology Regulations 2001.

‘OGTR’ means the Office of the Gene Technology Regulator.

‘Personal information’ has the same meaning as in the *Privacy Act 1988*. Personal information means information or an opinion about an identified individual, or an individual who is reasonably identifiable:

- (a) whether the information or opinion is true or not; and
- (b) whether the information or opinion is recorded in a material form or not.

‘Primary facility’ means an enclosed building within the Trial site which contains animal holding pens.

‘Regulations’ means the Gene Technology Regulations 2001 (Commonwealth) or the corresponding State law under which this licence is issued.

‘Regulator’ means the Gene Technology Regulator.

‘Risk group 2 organism’ means an organism that satisfies the criteria in AS/NZS 2243.3:2010 for classification as Risk Group 2

‘Sample’ means any biological material collected from a treated trial participant for analysis as part of the trial

‘Secondary facility’ means a fenced paddock within the Trial site.

‘Storage facility’ means a third-party facility offering logistical services and distribution of clinical supplies.

‘Trial site’ means the Queensland Animal Biosciences Precinct at The University of Queensland Gatton Campus.

Section 2 General conditions and obligations

Holder of licence

3. The licence holder is The University of Queensland

Remaining an Accredited Organisation

4. The licence holder must, at all times, remain an accredited organisation.

Validity of licence

5. This licence remains in force until it is suspended, cancelled or surrendered. No dealings with the GMO are authorised during any period of suspension, or after the licence has been cancelled or surrendered.

Note: Although this licence has no expiry date, the duration of preparation and administration of the GMOs is restricted in accordance with Condition 23.

Persons covered by this licence

6. The persons covered by this licence are:
 - (a) the licence holder, and any employees, agents or External service providers engaged by the licence holder; and
 - (b) the project supervisor(s); and
 - (c) other persons who are, or have been, engaged or otherwise authorised by the licence holder or the project supervisor to conduct any of the dealings authorised by this licence.
7. The licence holder must keep a record of all persons covered by this licence, and must keep a record of the contact details of the project supervisor(s) for the licence.

Note: Where External service providers are used, it is sufficient to record the company name and the position or job title of the person(s) conducting the dealing.

8. The licence holder must provide information related to the persons covered by the licence when requested to do so in writing by the Regulator and must provide the information within a time period stipulated by the Regulator.

Description of GMOs covered

9. The licence authorises specified dealings in respect of the GMOs identified and described in **Attachment A**.

Dealings authorised by this licence

10. The licence holder and persons covered by this licence may conduct the following dealings with the GMOs:
- (a) conduct the following experiments with the GMOs:
 - i) prepare the GMO for administration to trial horses;
 - ii) administer the GMO to trial horses by intramuscular injection or intranasal instillation;
 - iii) collect Samples from GMO administered horses;
 - iv) prepare Samples described in 10(a)iii) for transport;

(b) transport the GMOs;

(c) dispose of the GMOs;

and may possess, supply, use or store the GMO for the purposes of, or in the course of, any of these dealings.

11. Supply of the GMOs for the purposes of dealings by a person or organisation not covered by this licence is only authorised by this licence if the Regulator provides prior written approval to the licence holder.

Note: For approval to be granted, the receiving person or organisation must have an appropriate authorisation to conduct dealings with the GMOs. This is likely to be a NLRD or a licence issued by the Regulator.

12. This licence does not apply to dealings with the GMOs conducted as an NLRD or pursuant to another authorisation under the Act.

Conditions imposed by the Act

Note: The Act mandates the following 3 conditions.

Informing people of licence conditions (section 63)

13. The licence holder must inform any person covered by the licence, to whom a particular condition of the licence applies, of the following:
- (a) the particular condition, including any variations of it; and
 - (b) the cancellation or suspension of the licence; and
 - (c) the surrender of the licence.

Monitoring and audits (section 64)

14. If a person is authorised by this licence to deal with the GMOs and a particular condition of this licence applies to the dealing by that person, the person must allow the Regulator, or a person

authorised by the Regulator, to enter premises where the dealing is being undertaken, for the purposes of auditing or monitoring the dealing.

Additional information to be given to the Regulator (section 65)

15. The licence holder must immediately inform the Regulator, if they become aware of:
- (a) additional information about any risks to the health and safety of people, or to the environment, associated with the dealings authorised by the licence; or
 - (b) any contraventions of the licence by a person covered by the licence; or
 - (c) any unintended effects of the dealings authorised by the licence.

Note 1: For the purposes of this condition:

- (a) *The licence holder is taken to have become aware of additional information if they were reckless as to whether such information existed; and*
- (b) *The licence holder is taken to have become aware of contraventions, or unintended effects, if they were reckless as to whether such contraventions had occurred, or such unintended effects existed.*

Note 2: Contraventions of the licence may occur through the action or inaction of a person.

Note 3: Additional information includes any changes at a Trial site, which might increase the likelihood of unintentional exposure of people or release of the GMO into the environment.

Note 4: An example of informing immediately is contact made at the time of the incident via the OGTR free call phone number 1800 181 030.

Informing the Regulator of any material changes of circumstance

16. The licence holder must immediately, by notice in writing, inform the Regulator of:
- (a) any relevant conviction of the licence holder occurring after the commencement of this licence;
 - (b) any revocation or suspension after the commencement of this licence, of a licence or permit held by the licence holder under a law of the Commonwealth, a State or a foreign country, being a law relating to the health and safety of people or the environment;
 - (c) any event or circumstances occurring after the commencement of this licence that would affect the capacity of the licence holder to meet the conditions in it.
17. The licence holder must provide information related to the licence holder's ongoing suitability to hold a licence when requested to do so in writing by the Regulator, and must provide the information within a time period stipulated by the Regulator.

Further conditions with respect to informing persons covered by the licence

18. If a particular condition, including any variation of it, applies to an External service provider covered by this licence, the licence holder must not permit that person to conduct any dealings unless the person has been informed of the condition, including any variation of it.

Note: Information required under Condition 18 may be provided to External service providers who are engaged solely for storage and transport of the GMO through labelling of the outermost container of the GMOs in accordance with Condition 41(a).

19. If a particular condition, including any variation of it, applies to a person with respect to any dealing, other than to an External service provider, the licence holder must not permit a person covered by this licence to conduct that dealing unless:
 - (a) the licence holder has obtained from the person a signed and dated statement that the person:
 - i) has been informed by the licence holder of the condition and, when applicable, its variation; and
 - ii) has understood and agreed to be bound by the condition, or its variation; and
 - iii) has been trained in accordance with sub-condition 19(b) below; and
 - (b) the licence holder has trained that person in a manner which enables them to conduct the dealings in accordance with the conditions of this licence.
20. The licence holder must notify all persons covered by the licence, from whom Personal information relevant to the administration and/or enforcement of the licence is collected by the licence holder, that such Personal information may be disclosed to the Regulator.
21. The licence holder must ensure that a copy of the licence is readily available to all persons covered by the licence, other than External service providers, who are conducting dealings with the GMO.

Note: The licence may be made available electronically.

Section 3 Limits and control measures

Limits on trials conducted under this licence

22. The GMO may be administered to a maximum of 10 horses.
23. The preparation and administration of the GMO must be completed within 5 years from the date of issuing of the licence.
24. The licence holder must ensure that:
 - (a) administration of the GMO occurs within the Primary facility;
 - (b) horses remain in the Primary facility from the first administration of the GMO until at least 7 days after the last administration of the GMO; and
 - (c) horses have tested negative for the presence of the GMO via nasal swab PCR test before being moved from the Primary facility to the Secondary facility.
 - (d) A record of testing results must be kept for the duration of the licence and provided to the Regulator upon request.

Note: This licence condition aims to ensure that animals to be relocated do not contain residual episomes from the GMO.

Trial sites

25. Access to Trial sites must be restricted to persons authorised by the Licence holder.
26. Signs indicating the presence of the GMO must be displayed at all entrances to the Trial site.
27. The Trial site must be surrounded by a secure fence
28. The Secondary facility must be surrounded by a fence capable of containing horses.

Preparation and administration of the GMOs

29. Administration of the GMO to horses must not commence prior to approval of an Animal Ethics Committee and the APVMA.

30. Preparation and administration of the GMO must be conducted by suitably qualified and trained staff.
31. Persons administering the GMO and collecting Samples must also be trained in horse handling.
32. The licence holder must ensure that horses are properly restrained (*e.g.* with a tether) during;
 - (a) administration of the GMO
 - (b) Sample collection which requires sharps

Note: Before any of these activities take place, the details of each trial site must have been notified to the Regulator in accordance with Condition 48(a).

Conditions relating to the conduct of the dealings

33. The licence holder must ensure that each horse administered with the GMO is individually identifiable.
34. Following the first administration of the GMO and for at least 7 days after the last administration, animal excreta within the primary facility containing the GMO-inoculated animals must be removed from the holding pens daily and decontaminated.
35. Conditions that apply to dealings with GMOs do not apply to:
 - (a) nasal swabs collected from horses at least 2 weeks after administration of the GMO;
 - (b) blood Samples collected from horses at least 7 days after administration of the GMO; and
 - (c) other Samples, materials and waste, that are reasonably expected not to contain the GMO. Upon request from the Regulator, the licence holder must provide a written justification for this expectation.
36. The licence holder must ensure that dealings are only conducted in a manner which:
 - (a) does not compromise the health and safety of people; and
 - (b) minimises the exposure of persons conducting the dealings to the GMO.
37. The licence holder must ensure that procedures are in place to account for the GMO and GMO inoculated horses from generation to destruction. Records must be made available to the Regulator on request.
38. The licence holder must ensure that GMO-inoculated horses are not used, sold or otherwise disposed of for any purpose which would involve or result in its use as food for humans or feed for animals.

Work practices at Trial sites

39. For the purposes of Condition 36, work practices and behaviours within a Trial site must include, but are not limited to, the following:
 - (a) persons preparing or administering the GMO, or collecting blood, or nasal swab Samples that are reasonably expected to contain the GMO must wear personal protective equipment (PPE), including but not limited to overalls, gloves, boots, eye protection and a surgical face mask
 - (b) all work surfaces must be Decontaminated after they have been used for conducting dealings authorised by this licence;
 - (c) the Primary facility must be Decontaminated following the final removal of GMO-inoculated horses;
 - (d) equipment used for dealings with the GMOs must be Decontaminated after use;

Transport, storage and disposal of the GMOs

40. Unless covered by an NLRD, the licence holder must ensure that transport of the GMOs is conducted only for the purposes of, or in the course of, another dealing permitted by this licence, or for supply in accordance with Condition 11.
41. The licence holder must ensure that transport of the GMO, and storage of the GMO within the trial site, follows these sub-conditions:
- (a) GMOs must be contained within sealed, unbreakable primary and secondary container(s), with the outer packaging labelled to indicate at least:
 - i) that it contains GMOs; and
 - ii) that it contains biohazardous material as designated by a biohazard label; and
 - iii) the contact details for the licence holder; and
 - iv) instructions to notify the licence holder in case of loss or spill of the GMOs; and
 - (b) the external surface of the primary and secondary container must be Decontaminated prior to and after transport; and
 - (c) procedures must be in place to ensure that GMOs can be accounted for and that a loss of GMOs during transport or storage or failure of delivery can be detected; and
 - (d) access to the GMOs is restricted to authorised persons for whom Condition 18 or Condition 19 has been met (i.e. the GMOs are within a locked unit or an area which has restricted access). This includes situations where containers are left for collection in a holding area, or left unattended prior to Decontamination; and

Note: All stored GMOs remain the responsibility of the licence holder.

- (e) if the GMO is being transported or stored with a coolant (e.g. dry ice, liquid nitrogen or any other coolant) which will release a gas, a mechanism to allow the escape of the gas must be included. If water ice is used as a coolant then the outer packaging should be constructed so as to prevent any leakage. All containers must be able to withstand the temperatures to which they will be subjected; and

Note: When transporting and storing with coolants, it is preferable for coolants to be used outside of the secondary container.

- (f) a consolidated record of all GMOs being stored under this condition is maintained and made available to the Regulator upon request; and
- (g) for the purposes of transport entirely within a building, where the GMOs are accompanied by an authorised person for whom Condition 19 has been met, Conditions 41(a)iii), 41(a)iv) and 41(c) do not apply.

Note: This condition does not apply to transport of the GMO to and from the trial site conducted as an NLRD

42. The licence holder must ensure that all GMOs and waste reasonably expected to contain the GMOs are Decontaminated:
- (a) prior to disposal, unless the method of disposal is also a method of Decontamination; and
 - (b) before or upon suspension, cancellation or surrender of the licence, unless covered by another authorisation under the Act, or exported; and
 - (c) by autoclaving, chemical treatment, high-temperature incineration or any other method approved in writing by the Regulator.

Note: This condition does not apply to dealings conducted as an NLRD (see Condition 40)

43. Where transport is conducted by External service providers for the purpose of destruction, the licence holder must ensure that the GMO, or waste reasonably expected to contain the GMO, enters the clinical waste stream for Decontamination via autoclaving or high-temperature incineration.

Note 1: This condition does not apply to dealings conducted as an NLRD (see Condition 40)

Note 2: In the event of a spill during transport by an External service provider, compliance with relevant State or Territory legislation and regulations to manage clinical or biohazardous spills is sufficient.

Contingency plans

44. The licence holder must ensure that any person exposed to the GMOs is offered prompt medical attention. The clinician must be provided with any relevant information about the GMO.
45. If there is a spill or an unintentional release of the GMOs at the Trial site, the following measures must be implemented:
- (a) the GMOs must be contained to prevent further dispersal; and
 - (b) persons cleaning up the GMO must wear appropriate PPE as specified in condition 39(a); and
 - (c) the exposed area must be Decontaminated with an appropriate chemical disinfectant effective against the GMO; and
 - (d) any material used to clean up the spill or PPE worn during clean-up of the spill must be Decontaminated; and
 - (e) the licence holder must be notified as soon as reasonably practicable.
46. If there is an unintentional release of the GMOs, the licence holder must ensure that the following persons are notified as soon as reasonably possible:
- (a) the relevant IBC; and
 - (b) the Regulator.
47. If a GMO-inoculated horse escapes its containment, the animal must be captured and returned to the appropriate facility within the Trial site, and the licence holder must notify the Regulator immediately.

Section 4 Reporting and Documentation

*Note: The following licence conditions are imposed to demonstrate compliance with other conditions and facilitate monitoring of compliance by staff of the OGTR. Notices and reports may be emailed to OGTR.M&C@health.gov.au. A summary of notification and reporting requirements is provided at **Attachment B**.*

48. At least 14 days prior to first administering the GMO at the Trial site, or a timeframe agreed to in writing by the Regulator, the licence holder must notify the Regulator, in writing, of:
- (a) the commencement of the trial; and
 - (b) the proposed reporting structure for the trial at the site and how the reporting structure enables the licence holder to become aware of all reportable events including but not limited to Conditions 15, 16, 49 and 50;
 - (c) details of how the persons covered by the licence (for that type of dealing) will be informed of licence conditions applicable to them and how they will be trained to safely conduct the dealings;

- (d) the expected date of first administration;

Note: For the purpose of finding out whether the Act has been complied with, an OGTR inspector may, if entry is at a reasonable time, enter a facility occupied by the licence holder or a person covered by the licence and exercise monitoring powers.

- 49. The licence holder must notify the Regulator, in writing, of the end of the trial, no later than 30 days after:
 - (a) the final dose being administered; or
 - (b) the decision that no further horses will be administered with the GMO.
- 50. The licence holder must inform the Regulator as soon as reasonably possible:
 - (a) in the event of a loss or spill of the GMO;
 - (b) in the event of the exposure of a person, or animals other than the horses being administered, to the GMO.
- 51. Upon request from the Regulator, the licence holder must provide any signed records or documentation collected under a condition of this licence, within a time period stipulated by the Regulator



ATTACHMENT A

DIR No: 214

Full Title: Trial of a genetically modified (GM) vaccine for the prevention of respiratory disease in horses

Organisation Details

Postal address: The University of Queensland
St Lucia
Queensland, 4072

Accreditation No: Accr-030

GMO Description

GMOs covered by this licence

The GM vaccine contains a replication defective Human Adenovirus serotype 5 modified by the deletion or introduction of the genes or genetic elements listed in Table 1 below.

Parent Organism

Common Name: Human Adenovirus
Scientific Name: Human Adenovirus C, serotype 5

Modified traits

Category: Veterinary
Description: The GMO has deletions of the E1 and E3 regions of its genome which prevent replication and immune evasion respectively. It includes an insertion of the VapA gene from *R. equi* under the CMVie promoter.

Table 1. Nucleic acid responsible for conferring the modified traits

Genetic modifications	
Source, identity, nature of modification	Modified trait description
• Deletion of viral early-transcribed region 1 (E1)	to render virus replication incompetent.
• Deletion of viral early-transcribed region 3 (E3)	to render the virus unable to evade the host immune system.
• Insertion of VapA	to induce an immune response

Purpose of the dealings with the GMO:

The purpose of the proposed trial is to evaluate the immunogenicity, safety and efficacy of a genetically modified vaccine in horses for the prevention respiratory disease (Rattles).

Route of administration of the GMOs

The GMO would be administered via intramuscular injection or via intranasal instillation.



Attachment B – Summary of reporting requirements*

Prior to the commencement of the trial	Condition	Timeframe for reporting
Expected date of first administration at trial site	48(d)	At least 14 days prior to the first administration of the GMO at each trial site.
The proposed reporting structure for the trial	48(b)	
Information to be provided at any time during the trial	Condition	Timeframe for reporting
Any additional information related to the health and safety of people and the environment associated with the dealings covered by the licence, or any unintended effects of the dealings authorised by the licence	15(a),(c)	As soon as the licence holder becomes aware
Information related to any contravention of the licence by a person covered by the licence	15(b)	As soon as the licence holder becomes aware
Any relevant conviction of the licence holder	16(a)	Immediately
Any revocation or suspension of a licence or permit held by the licence holder under a law of the Commonwealth, a State or a foreign country	16(b)	Immediately
Any event or circumstances that would impact the licence holder capacity to meet the licence conditions	16(c)	Immediately
Any unintentional release of the GMO via escaped horses.	46	As soon as reasonably possible after becoming aware of the event
In the event of exposure of a person or animal other than the horses in the trial to the GMO	50(b)	Immediately
In the event of loss or spill of the GMO	50(a)	Immediately
Provide notification to the Regulator, in writing, of the final GMO administration.	49(a)	Within 30 days of the decision to cease GMO administration at that particular trial site.
Information to be provided on request by the Regulator		
Information related to the persons covered by the licence	7	Within a timeframe stipulated by the Regulator
Information related to the licence holder’s ongoing suitability to hold a licence	17	Within a timeframe stipulated by the Regulator
Copies of signed and dated statements and training records	19	Within a timeframe stipulated by the Regulator
A consolidated record of all GMOs being stored	41(f)	Within a timeframe stipulated by the Regulator
Any signed records or documentation collected under a condition of this licence	51	Within a timeframe stipulated by the Regulator

* Notifications and documents to be sent to OGTR.M&C@health.gov



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