Australian Government
Department of Health, Disability and Ageing
Office of the Gene Technology Regulator

June 2025

Risk Assessment and Risk Management Plan for

DIR 215

Limited and controlled release of canola genetically modified for dairy protein production

Applicant: Miruku Australia Pty Ltd

Summary of the Risk Assessment and Risk Management Plan for Licence Application No. DIR 215

Decision

The Gene Technology Regulator (the Regulator) has decided to issue a licence for this application for the intentional release of a genetically modified organism (GMO) into the environment. A Risk Assessment and Risk Management Plan (RARMP) for this application has been prepared by the Regulator in accordance with the *Gene Technology Act 2000* (the Act) and corresponding state and territory legislation, and finalised following consultation with a wide range of experts, agencies and authorities, and the public. The RARMP concluded that the proposed field trial poses negligible risk to human health and safety and the environment and that any risks posed by the dealings can be managed by imposing conditions on the release.

The application

Applicant	Miruku Australia Pty Ltd (Miruku)
Project title	Limited and controlled release of canola genetically modified for dairy protein production[1]
Parent organism	Canola (*Brassica napus* L.)
Introduced genes	Introduced gene[2] producing dairy protein: • modified β-casein gene based on the gene from cattle (*Bos taurus*) for dairy protein production Introduced marker gene: • *bar* gene from bacterium *Streptomyces hygroscopicus* for tolerance to the herbicide glufosinate
Genetic modification method	*Agrobacterium*-mediated transformation
Number of lines	Up to 50 lines
Previous releases	None in Australia or overseas
Proposed locations	Up to 2 sites per year in 2025, 5 in 2026, 10 in 2027, 15 in 2028 and 20 in 2029. Sites to be selected from 135 possible local government areas in New South Wales, Victoria, Western Australia and South Australia
Proposed release size	Up to 1 ha in 2025, 5 ha in 2026, 25 ha in 2027, 105 ha in 2028, and 300 ha in 2029, with a maximum of 436 ha over the period of release
Proposed period of release	From issue of licence until December 2029
Principal purpose	To produce dairy protein in GM canola under field conditions

[1] The title of the project as supplied by the applicant is "Limited and controlled release of Canola genetically modified for dairy protein and fat composition".

[2] Confidential Commercial Information (CCI): Some details about the introduced genetic elements in GM canola have been declared as CCI under section 185 of the Act. This information will be made available to the prescribed experts and agencies that will be consulted on this application. CCI is not available to the public.

Risk assessment

The risk assessment process considers how the genetic modification and proposed activities conducted with the GMOs 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 current scientific/technical knowledge, information in the application (including proposed limits and controls) and relevant previous approvals. Both the short- and long-term impacts are considered.

Credible pathways to potential harm that were considered included exposure of people or other desirable organisms[3] to the GM plant material, potential for persistence or dispersal of the GMOs, transfer of the introduced genetic material to non-GM canola plants and potential for fitness advantages to pest organisms. Potential harms associated with these pathways included toxicity and allergenicity to people, toxicity to desirable animals, and environmental harms due to weediness.

The risk assessment concludes that risks to the health and safety of people or the environment from the proposed dealings are negligible. No specific risk treatment measures are required to manage these negligible risks. The principal reasons for the conclusion of negligible risks are that the proposed limits and controls, such as not using GM plant material in commercial human food or animal feed, will effectively minimise exposure to the GMOs. In addition, there is currently no evidence to suggest the introduced genetic modifications would lead to harm to 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.

As the level of risk is considered negligible, specific risk treatment is not required. However, since this is a limited and controlled release, the licence includes limits on the size, location and duration of the release, as well as controls to prohibit the use of GM plant material in commercial human food and animal feed, to minimise dispersal of the GMOs or GM pollen from the trial site, to transport GMOs in accordance with the Regulator's guidelines, to destroy GMOs at the end of the trial and to conduct post-harvest monitoring at the trial sites to ensure the GMOs are destroyed.

[3] Desirable organisms are those that are valued and should be protected, while undesirable organisms cause harm and should be controlled (OGTR, 2013). This is determined by legislation, government policies, national and international guidance material, and widely accepted community norms. Undesirable plants that cause economic, social or environmental harm, or harm to human/animal health, are called weeds. Animals that cause harm are known as pests.

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Abbreviations

AICIS	Australian Industrial Chemicals Introduction Scheme
APVMA	Australian Pesticides and Veterinary Medicines Authority
CCI	Confidential Commercial Information
CMA	Cow's milk allergy
Da	Daltons
DAFF	Department of Agriculture, Fisheries and Forestry
DIR	Dealings involving Intentional Release
FSANZ	Food Standards Australia New Zealand
GM(O)	Genetically modified (organism)
ha	Hectare(s)
HGT	Horizontal gene transfer
IBC	Institutional Biosafety Committee
kDa	Kilodaltons
LGA	Local Government Area
m	Metre(s)
NLRD	Notifiable Low Risks Dealings
NSW	New South Wales
OGTR	Office of the Gene Technology Regulator
PAT	Phosphinothricin acetyltransferase
RARMP	Risk Assessment and Risk Management Plan
Regulations	Gene Technology Regulations 2001
Regulator	Gene Technology Regulator
SA	South Australia
TGA	Therapeutic Goods Administration
the Act	The *Gene Technology Act 2000*
UTR	Untranslated region
Vic	Victoria
WA	Western Australia

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.

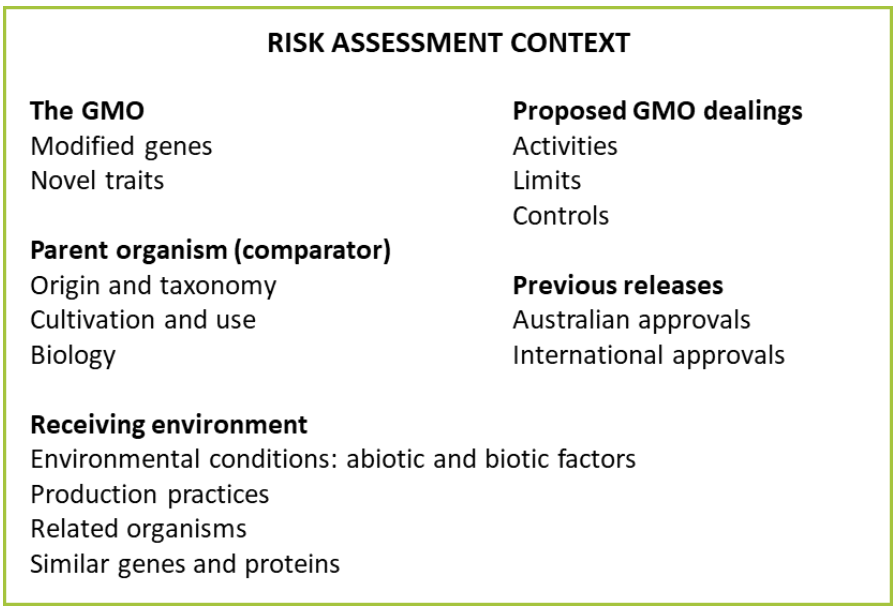

Figure 1. Summary of parameters used to establish the risk assessment context, within the legislative requirements, operational policies and guidelines of the OGTR, and the Risk Analysis Framework

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.

7. Section 52 of the Act requires the Regulator to seek comment on the RARMP from agencies - the Gene Technology Technical Advisory Committee (GTTAC), State and Territory Governments, Australian Government authorities or agencies prescribed in the Regulations, Australian local councils and the Minister for the Environment - and from the public. The advice from the prescribed experts, agencies and authorities and how it was taken into account is summarised in Appendix A. Two public submissions were received and their consideration is summarised in Appendix B.

1.1 Interface with other regulatory schemes

8. 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). These dealings may also be subject to the operation of State legislation recognising an area as designated for the purpose of preserving the identity of GM crops, non-GM crops, or both GM crops and non-GM crops, for marketing purposes.

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

Section 2 The proposed dealings

10. Miruku Australia Pty Ltd (Miruku, the applicant) proposes to release several GM canola lines into the environment under limited and controlled conditions. The GM plants have been genetically modified for dairy protein production.

11. The purpose of the release is to assess dairy protein production in GM canola under field conditions. The applicant will also evaluate agronomic performance of the GM canola lines in the field.

12. The dealings involved in the proposed intentional release are to:

- conduct experiments with the GMOs
- breed the GMOs
- propagate the GMOs
- use the GMOs in the course of manufacture of a thing that is not the GMOs
- grow the GMOs
- transport the GMOs
- dispose of the GMOs

and the possession, supply or use the GMOs in the course of any of these dealings.

13. Initial transformation of the GMOs will occur in Australia under a Notifiable Low Risks Dealings (NLRD) authorisation.

14. The GM plant material would not be used for commercial human food or animal feed.

15. The GM seeds will be processed to release the protein components to use in food products that may only be used in human sensory testing to assess their feel, smell, taste and appearance. They will not be used for commercial food or feed. Sensory testing would result in negligible consumption of the components from the GM seeds as the products are not intended to be swallowed during testing. These trials would only occur if Miruku obtains the appropriate approvals for each trial in accordance with the National Statement on Ethical Conduct in Human Research.

2.1 The proposed limits of the dealings (duration, size, location and people)

16. The release is proposed to take place between May 2025 and December 2029. Planting would occur primarily during the winter cropping season, but occasionally a summer crop cycle may also be used.

17. GM canola is proposed to be grown at up to 52 trial sites over the period of release. The proposed maximum number of sites, planting area per site, combined total planting area for each year, and cumulative maximum total planting area are detailed Table 1.

Table 1. Proposed duration and maximum number of sites and planting area per year

Year	Maximum number of sites per year	Maximum area (ha) per site	Maximum combined area (ha) per year	Cumulative maximum total area (ha)
2025	2	0.5	1	1
2026	5	1	5	6
2027	10	2.5	25	31
2028	15	7	105	136
2029	20	15	300	436

18. Sites for trial release would be selected from 135 possible local government areas (LGAs) in New South Wales (NSW), Victoria (Vic), Western Australia (WA) and South Australia (SA) (Table 2). The field trials would occur on research stations or private land in rural areas where persons other than those conducting dealings would not have access to the field trial sites.

Table 2. LGAs where GM canola trial sites may be located

New South Wales	Victoria	Western Australia	South Australia
Berrigan	Ararat	Albany	Adelaide Plains
Bland	Ballarat	Beverley	Barossa
Blayney	Benalla	Boddington	Light
Cabonne	Buloke	Boyup Brook	Wakefield
Coolamon	Campaspe	Bridgetown-Greenbushes	
Coonamble	Central Goldfields	Brookton	
Cootamundra-Gundagai	Colac Otway	Broomehill-Tambellup	
Cowra	Corangamite	Carnamah	
Dubbo	Gannawarra	Coorow	
Edward River	Glenelg	Corrigin	
Federation	Golden Plains	Cranbrook	
Forbes	Greater Bendigo	Cuballing	
Gilgandra	Greater Geelong	Cunderdin	
Greater Hume	Greater Shepparton	Dalwallinu	
Griffith	Hepburn	Denmark	
Gunnedah	Hindmarsh	Donnybrook-Balingup	
Gwydir	Horsham	Dowerin	
Hay	Indigo	Dumbleyung	
Hilltops	Loddon	Esperance	
Inverell	Macedon Ranges	Gnowangerup	
Junee	Mildura	Goomalling	
Leeton	Mitchell	Greater Geraldton	
Liverpool Plains	Moira	Jerramungup	
Lockhart	Moorabool	Katanning	
Mid-Western	Mount Alexander	Kent	
Moree Plains	Moyne	Kojonup	
Murray River	Northern Grampians	Manjimup	
Murrumbidgee	Pyrenees	Merredin	
Muswellbrook	Southern Grampians	Mingenew	

New South Wales	Victoria	Western Australia	South Australia
Narrabri	Strathbogie	Moora	
Narrandera	Swan Hill	Morawa	
Narromine	Wangaratta	Nannup	
Orange	West Wimmera	Narrogin	
Parkes	Wodonga	Northam	
Snowy Valleys	Wyndham	Perenjori	
Tamworth	Yarriambiack	Pingelly	
Temora		Plantagenet	
Upper Hunter		Quairading	
Wagga Wagga		Ravensthorpe	
Walgett		Tammim	
Warren		Three Springs	
Warrumbungle		Toodyay	
Weddin		Victoria Plains	
		Wagin	
		Wandering	
		West Arthur	
		Wickepin	
		Williams	
		Wongan-Ballidu	
		Woodanilling	
		Wyalkatchem	
		York	

19. Only trained and authorised persons would be permitted to deal with the GM canola.

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

20. The applicant has proposed a number of controls to restrict the spread and persistence of the GM canola and the introduced genetic material in the environment. These include:

- locating each trial site at least 50 m away from the nearest natural waterway
- surrounding each planting area with either a 50 m monitoring zone, a 35 m monitoring zone and 15 m pollen trap, or a 10 m monitoring zone and covering the planting area with an insect-proof tent (see Figure 2)
- inspecting monitoring zones from 14 days prior to flowering until harvest, to identify and destroy volunteer canola or related species
- surrounding each monitoring zone with an isolation zone of 950 m, or 350 m with use of a pollen trap, or 390 m with use of an insect-proof tent, where no canola or sexually compatible species are grown (see Figure 2)
- treating non-GM canola plants grown in planting areas and pollen traps as if they are GMOs
- cleaning equipment and clothing after use on trial sites
- bagging or tenting GM canola
- using seeding or harvesting methods or equipment that minimise dispersal of GM plant material
- controlling rodents on trial sites
- restricting access to trial sites to authorised persons
- cleaning of planting areas post-harvest
- tilling and irrigating each planting area during the post-harvest monitoring period
- post-harvest monitoring of the trial sites for 24 months to identify any volunteer canola, and destroy

volunteers before they reach flowering

- destroying all GMOs not required for further experimentation
- transporting and storing GMOs in accordance with the current Regulator's Guidelines for the Transport, Storage and Disposal of GMOs
- not allowing the GMOs or GM products to be used for commercial human food or animal feed.

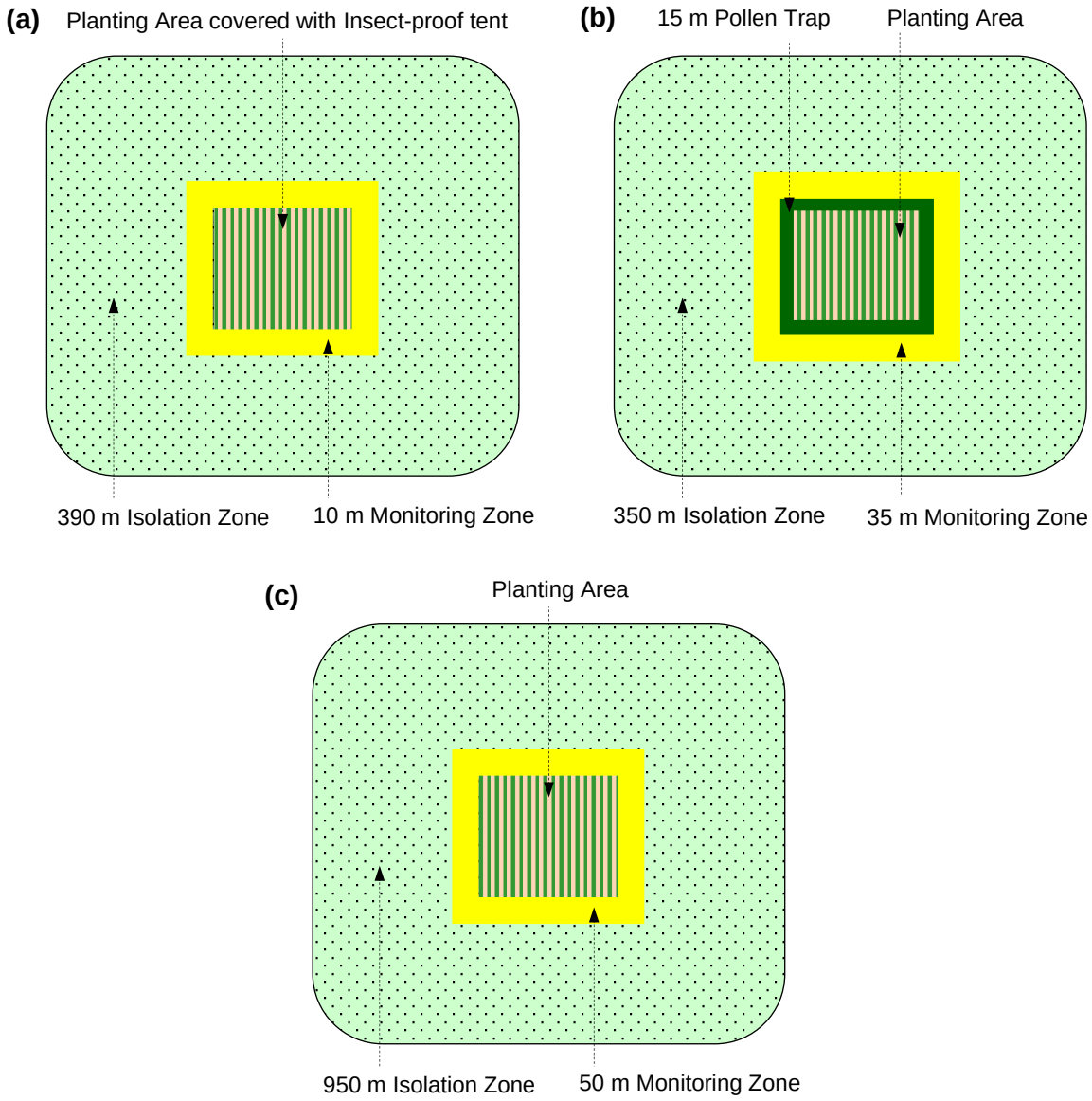

Figure 2. Diagrams (not to scale) showing the relationships between planting area, pollen trap, monitoring zone and isolation zone. Site layout (a) with insect-proof tent, (b) without insect-proof tent and with pollen trap, and (c) without insect-proof tent or pollen trap.

21. The proposed limits and controls are taken into account in the risk assessment (Chapter 2) and their suitability for containing the release will be evaluated in the risk management plan (Chapter 3).

Section 3 The parent organism

22. The parent organism is *Brassica napus* L., commonly known as canola, rapeseed or oilseed rape. *B. napus* is exotic to Australia.

23. Canola is the third-most widely grown crop in Australia. It is grown mainly in WA, NSW, Vic and SA (ABARES, 2024a). Canola oil is used as food and the canola meal remaining after oil extraction is used as animal feed.

24. *B. napus* is naturalised in Australia. In agricultural areas where it is grown, it can be a weed in subsequent crops. There are isolated reports of *B. napus* as an environmental weed in WA and Vic (Randall, 2017). However, the most recent Western Australian state government environmental weed risk assessment gives *B. napus* a weed risk rating of negligible to low (Moore and Nazeri, 2022), and the most recent Victorian state government environmental weed list gives *B. napus* a risk ranking score of zero and classified as 'lower risk' (White et al., 2022).

25. Detailed information about the parent organism is contained in the document *The Biology of* Brassica napus *L. (canola)* and Brassica juncea *(L.) Czern. & Coss. (Indian mustard)* (OGTR, 2024), which was produced to inform the risk analysis process and is available from the Resources page on the OGTR website. Baseline information from this document will be used and referred to throughout the RARMP.

26. While non-GM canola is not generally regarded as allergenic or toxic to humans or animals, it does produce some toxins and anti-nutritional factors such as erucic acid and glucosinolates, and some cases of canola food, pollen and dust allergies have also been reported (OGTR, 2024).

Section 4 The GMOs, nature and effect of the genetic modification

27. The applicant proposes to release 5 groups of up to 10 canola lines each, genetically modified for dairy protein production.

4.1 The genetic modifications in the GMOs proposed for release

28. The specific parental canola variety for the GMOs is 'Oscar'.

29. All GM canola lines contain one of 2 variants (denoted BCN2 and BCN3) of the gene encoding β-casein (*CSN2*) from domestic cattle, fused to a gene derived from common plant species. Each line also contains a sequence to target the expressed β-casein fusion proteins to the cell wall or vacuoles and a glufosinate herbicide tolerance marker gene. Genes introduced into the GM canola lines are summarised in Table 3. Regulatory and localisation sequences are summarised in Table 4. Each GM canola line contains only a subset of the genetic elements listed in Table 3 and Table 4.

30. The identities of some the genetic elements, and the arrangement of genetic elements in the vectors introduced into GM canola have been declared Confidential Commercial Information (CCI). Under section 185 of the Act, the confidential information is made available to the prescribed agencies and experts that are consulted on the RARMP for this application.

Table 3. Introduced genes

Genetic element	Source organism	Encoded protein	Intended function
bar	*Streptomyces hygroscopicus*	Phosphinothricin acetyltransferase	Herbicide resistance
BCN2	*Bos taurus*	Miruku β-casein (CSN2) variant 2	β-casein production
BCN3	*B. taurus*	Miruku β-casein (CSN2) variant 3	β-casein production
CCI gene	CCI (common plant species)	CCI	CCI

4.1.1 BCN2 and BCN3

31. The introduced *BCN2* and *BCN3* constructs encode variants to A2 β-casein from cattle (*Bos taurus*). The purpose of the introduction of *BCN2* and *BCN3* is to produce the β-casein protein variants in seeds of

GM canola. Expression is driven by a seed-specific promoter, either alone, or in combination with organelle localisation sequences.

32. Caseins are the primary proteins in bovine milk, comprising 80% of total protein content (Hassanin et al., 2022). β-casein accounts for approximately 35% of bovine milk protein (Daniloski et al., 2022), and is highly polymorphic, with at least 15 known variants (Sebastiani et al., 2020). The A1 and A2 variants are the most common, which differ by a single amino acid at residue 67 (Farrell et al., 2004). β-casein has a highly charged and hydrophilic N-terminal region and a primarily hydrophobic C-terminus (Dauphas et al., 2005), which is primarily due to the presence of 5 phosphorylated serine residues (Creamer et al., 1981; McCarthy et al., 2013). This amphiphilic nature enables β-casein to act as an emulsifier, and partial dephosphorylation of these serine residues results in reduced ability of β-casein to stabilise emulsions and the accumulation of β-casein in larger globules (Cassiano and Areas, 2003; McCarthy et al., 2013).

33. The *BCN2* and *BCN3* coding sequences are identical to the A2 β-casein sequence, except *BCN2* replaces serine residues at positions 17, 19 and 35 with aspartic acid, while *BCN3* replaces serine residues at positions 15, 18 and 35 with aspartic acid and serine residues at positions 17 and 19 with glutamic acid. These modifications are expected to result in partial dephosphorylation of β-casein and alter its emulsification properties.

34. *BCN2* and *BCN3* coding sequences are fused with a gene derived from a common plant species and are expressed as a chimeric fusion protein using a seed-specific promoter.

4.1.2 bar

35. The *bar* (*bialaphos resistance*) gene is derived from the bacterium *Streptomyces hygroscopicus* (Thompson et al., 1987). The *bar* gene encodes a phosphinothricin acetyltransferase (PAT) enzyme that confers tolerance to glufosinate herbicide. PAT acetylates glufosinate, converting it to *N*-acetyl-L-glufosinate, which is not toxic to plants (OECD, 2002).

36. Expression of *bar* is controlled by a 35S promoter from the Cauliflower mosaic virus (*CaMV35S*), which drives constitutive expression of *bar* in all plant tissues (Kay et al., 1987).

37. The Regulator has previously assessed and approved GM crops containing the *bar* gene for commercial release in Australia, most recently under licence DIR-190, and for field trials, most recently under DIR 204.

4.1.3 Regulatory and localisation sequences

38. The GM canola lines will also contain introduced regulatory sequences and localisation signals to control expression of the inserted genes (Table 4). These include promoters to drive gene expression, terminators, and localisation signals to spatially restrict protein localisation. The identities of some of the promoters and 3'UTR sequences introduced into GM canola have been declared CCI by the Regulator. Under section 185 of the Act, the CCI has been made available to the prescribed agencies and experts that are consulted on the RARMP for this application.

Table 4. Introduced regulatory elements and localisation sequences

Genetic element	Source	Intended function
AtCel1 cell wall signal	Cell wall signal from *Arabidopsis thaliana CEL1* gene	Cell wall targeting sequence
CaMV35S promoter	Cauliflower mosaic virus	Constitutive promoter
CCI 3'UTR	*Glycine max*	Terminator and polyadenylation signal
CCI promoter	*G. max*	Seed-specific promoter
CT-CVS C-terminus	C-terminal beta-conglycinin vacuolar sequence from *Glycine max*	Terminator

Genetic element	Source	Intended function
ER Retention	C-terminal H/KDEL tag sequence from *A. thaliana*	Endoplasmic reticulum retention
GmCVS vacuolar signal	Vacuole signal from *G. max* beta-conglycinin gene	Vacuole targeting sequence
HvAVSP vacuolar signal	Vacuole signal from *Hordeum vulgare* aleurain gene	Vacuole targeting sequence
Prr	*Nicotiana tabacum*	Cell wall targeting sequence
nos 3'UTR	*Agrobacterium tumefaciens*	Terminator and polyadenylation signal

4.2 Method of genetic modification

39. The GM canola lines were generated by *Agrobacterium*–mediated transformation. This method has been widely used in Australia and overseas for introducing genes into plants. Information about this method can be found in the document Methods of plant genetic modification, available from the OGTR Risk Assessment References page.

40. Canola breeding line 'Oscar' was transformed using the methodology described by Zhang et al. (2005). Transformants were selected on media containing the herbicide glufosinate. Selection agents were also used to eliminate *Agrobacterium* during *in vitro* selection of the transformed canola plants. *Agrobacterium* is not normally transmitted from one generation to the next via seed, therefore selected GM canola plants were propagated by single seed descent.

41. Parental canola lines were transformed with one of 5 binary plasmid vectors classified into 2 categories, which are described in Table 5. The arrangement of the genetic elements in the 5 vectors introduced into GM canola have been declared CCI.

Table 5. Categories of binary vector transformed into GM canola

Category	Vector number	Description
1	1	β-casein version 2 fusion (BCN2) fusion protein with glufosinate resistance
2	2-5	β-casein version 3 fusion (BCN3) fusion protein with various cell wall or vacuole targeting sequences and glufosinate resistance

4.3 Toxicity/allergenicity of the proteins associated with the introduced genes

42. As the GMOs are at an early stage of development, no toxicity or allergenicity studies have been conducted on the GM canola plants or purified proteins produced by the introduced genetic elements. The genetic element components of the β-casein fusion protein and their encoded proteins have also not been assessed by authorities in any countries for toxicity and allergenicity.

43. Discussion of the toxicity/allergenicity of some the introduced genes that have been declared CCI is made available to the prescribed agencies and experts that are consulted on the RARMP for this application as required under section 185 of the Act.

4.3.1 β-casein

44. A recent population-based study reported cow's milk allergy (CMA) in 1.3% of 1-year-old Australian infants (Soriano et al., 2023). Allergic reactions to cow's milk proteins are characterised by asthma, atopic dermatitis, urticaria (hives), rhinitis, gastrointestinal disorders, and anaphylaxis (Docena et al., 1996). Tolerance to cow's milk proteins usually develops as children mature, and consequently is present in less than 0.5% of adults (Fiocchi et al., 2010).

45. Caseins are known food allergens. In one study of 80 patients with known CMA, 100% of patients produced immunoglobulin-E (IgE) antibodies against casein proteins, indicating an immune reaction (Docena et al., 1996), while in another study of CMA patients, 75.3% produced IgE antibodies against caseins (Shoormasti et al., 2011).

46. Little information was found regarding potential toxicity or allergenicity of β-casein specifically. Upon digestion, proteolysis of the A1 variant of β-casein produces the peptide β-casomorphin-7 (BCM-7), which is implicated in adverse gastrointestinal effects (Giribaldi et al., 2022). However, the applicant proposes to introduce sequence variations to the A2 variant in GM canola, which does not produce BCM-7 and consequently is not associated with digestive intolerance. Nonetheless, given the high rate of allergenicity to caseins among CMA patients, (Daniloski et al., 2022), allergenicity to A2 β-casein variants is possible.

47. Allergenicity to caseins via dermal contact is rare and often requires existing damage to the skin barrier to cause immune reaction (Jensen et al., 2022). One study describes a single patient who developed dermatitis and rhinitis in response to casein protein present in microbiology laboratory culture media (Nakonechna et al., 2019). In another study, a single patient developed rhinitis and asthma following exposure to casein in a dermatological formulation associated with their occupation (Bonadonna et al., 2003). No information was found in the literature on toxicity or allergenicity associated with dermal contact with β-casein specifically.

4.3.2 PAT protein

48. The *bar* gene and its encoded PAT protein have been extensively assessed in previous RARMPs for commercial release of GM crops including canola (DIR 021/2002, DIR 108, DIR 138, DIR 175), cotton (DIR 062/2005, DIR 143, DIR 145, DIR 173) and a limited and controlled release of GM wheat (DIR 204). The PAT protein has been assessed to lack toxicity to humans or animals, or allergenicity in humans on the following basis:

- the *bar* gene was derived from the common soil bacterium *S. hygroscopicus*, which is not considered a pathogen of humans or other animals;
- no sequence homology has been found between PAT and any known toxic or allergenic proteins;
- the PAT protein does not possess any of the characteristics associated with food allergens;
- the PAT protein is inactivated by heat, e.g. through cooking, and by low pH, e.g. in the human stomach;
- the PAT protein is rapidly degraded in simulated gastric or intestinal fluid; and
- purified PAT protein was not toxic to mice and rats when administered at high doses in acute toxicity studies.

49. FSANZ has approved food derived from a number of GM crops expressing PAT protein as safe for human consumption. This includes GM canola (ANZFA, 2001; FSANZ, 2017), cotton (FSANZ, 2005a, 2010a, b, 2013), corn (FSANZ, 2005b) and rice (FSANZ, 2008).

4.4 Characterisation of the GMOs

50. The applicant has stated that the GM canola lines proposed for release are still in development. Initial observations of the GM canola grown in controlled glasshouse conditions indicated no phenotypic differences compared to non-GM canola. Both GM and non-GM canola exhibited similar growth patterns, morphology, germination rates, time to flowering, plant height and seed count per plant. No quantitative data were provided by the applicant. Further data on the agronomic performance of the GM canola lines will be collected during the field trials.

Section 5 The receiving environment

51. The receiving environment forms part of the context in which the risks associated with dealings involving the GMOs are assessed. Relevant information about the receiving environment includes abiotic and biotic interactions of the crop with the environment where the release would occur; agronomic

practices for the crop; presence of plants that are sexually compatible with the GMOs; and background presence of the gene(s) used in the genetic modification (OGTR, 2013).

52. Detailed information about the commercial cultivation and distribution of canola in Australia is presented in the document *The Biology of* Brassica napus *L. (canola) and* Brassica juncea *(L.) Czern. & Coss. (Indian mustard)* (OGTR, 2024).

5.1 Relevant abiotic factors

53. The geographical distribution of commercial canola cultivation in Australia is limited by several abiotic factors, the most important being water availability. Canola is generally grown as a winter crop in winter-dominant medium and high rainfall environments that receive annual rainfall of more than 350 mm (GRDC, 2009; OGTR, 2024). Germination of seed will only occur if there is sufficient soil moisture, and drought stress after anthesis can significantly reduce yield due to abortion of seed and reduced pod numbers. Canola is also sensitive to waterlogging, whereby waterlogged soil deprives canola seeds of oxygen and impairs germination (GRDC, 2009; OGTR, 2024). Other abiotic stresses that can reduce canola yields include frost, particularly during early pod development, and heat stress (GRDC, 2009).

5.2 Relevant biotic factors

54. A number of diseases have the potential to significantly reduce the yield of canola. Blackleg disease caused by the fungal pathogen *Leptosphaeria maculans* is the most serious disease affecting commercial canola production in Australia (GRDC, 2009; OGTR, 2024). Other damaging diseases of canola include stem rot caused by the fungus *Sclerotinia sclerotiorum* and damping-off, caused mainly by the fungus *Rhizoctonia solani* (GRDC, 2009, 2015).

55. Canola is most susceptible to insect pests during establishment of the crop, particularly from redlegged earth mite (*Halotydeus destructor*), blue oat mites (*Penthaleus major*, *P. falcatus* and *P. tectus* sp. n.), lucerne fleas (*Sminthurus viridis*), cutworms (*Agrotis* spp.) and aphids (*Brevicoryne brassicae*, *Myzus persicae*, *Lipaphis pseudobrassicae* and *Aphis craccivora*, which may also act as viral vectors) (GRDC, 2009). From flowering to crop maturity, severe damage can be caused by aphids, Rutherglen bugs (*Nysius vinitor*), diamondback moth caterpillars (*Plutella xylostella*) and heliothis caterpillars (family *Noctuidae*).

56. Canola is highly susceptible to weed competition during the early stages of growth (GRDC, 2009, 2015). Hybrid canola varieties have greater seedling vigour than open-pollinated canola and so are more competitive with weeds (GRDC, 2015, 2017). Common weeds of Australian canola crops include grassy weeds (such as rigid ryegrass, vulpia and wild oat), volunteer cereals, and weeds from the Brassicaceae family including wild radish (*Raphanus raphanistrum*), Indian hedge mustard (*Sisymbrium orientale*), shepherds purse (*Capsella bursa-pastoris*), Asian mustard (*Brassica tournefortii*), charlock (*Sinapis arvensis*), turnip weed (*Rapistrum rugosum*) and Buchan weed (*Hirschfeldia incana*) (GRDC, 2015, 2017).

5.3 Relevant agricultural practices

57. The applicant specifies that GM canola seeds would be planted in trial sites during the winter cropping season, but a summer cropping cycle may also be used occasionally. Non-GM canola lines planted at the trial sites for comparative purposes would be treated as if it were GM canola.

58. GM and non-GM canola crops would be maintained in a similar manner to commercial canola crops (see Section 2.2). Standard cultivation practices for canola in Australia are discussed in *GRDC Canola GrowNotes* (GRDC, 2015, 2017).

59. The applicant used the glufosinate tolerance conferred by the introduced *bar* gene as a selectable marker during transformation. Glufosinate herbicide is not intended to be applied to plants growing in the field trial. Glufosinate is not routinely used to control volunteer canola (AOF, 2019).

60. The applicant specifies that the GM canola would be grown at field sites either as an irrigated or dryland crop. Seeds would be planted in row plots with typical row spacing for canola, e.g. 30-40 cm, in plots spaced 1-2 m apart, although other configurations may be used. Small areas would be hand-planted or planted with a small plot cone-seeder, while larger areas would be planted with commercial equipment.

61. Nitrogen fertiliser would be deep injected pre-plant or at planting. Land would be cultivated once or twice after planting to control weeds, aerate soil and allow efficient irrigation. Furrow or flood irrigation would be used where necessary, and pre-irrigation may be conducted to store soil moisture and reduce salt levels in the soil.

62. Pest monitoring would be conducted once or twice per week by field technicians.

63. Canola seeds would be harvested by hand for small plantings or with commercial equipment for larger plantings, when seed moisture reaches 5-8%.

64. Planting areas will be left fallow after harvest and site cleaning, to facilitate the germination and monitoring of volunteers. Sites may be replanted with GM canola in subsequent years or would be planted with rotation crops such as cereals or pulses.

65. Additional agricultural practices proposed by the applicant are discussed further in Chapter 3, Section 3.1.

5.4 Presence of related plants in the receiving environment

66. Canola is primarily self-pollinating, but approximately 30% of seeds are produced by cross-pollination (Hüsken and Dietz-Pfeilstetter, 2007). Cross-pollination can be mediated by insects, wind or physical contact (OGTR, 2024).

67. Canola has been reported to outcross in the field with the following species: *Brassica carinata*, *B. napus, B. juncea, B. oleracea, B. rapa, Hirschfeldia incana* (Buchan weed), *R. raphanistrum* (wild radish) and *S. arvensis* (charlock) (Ford et al., 2006; Warwick et al., 2009). All of these species are known to be present in Australia, with the exception of *B. carinata* (Atlas of Living Australia, accessed 17 January 2025).

68. Of the *Brassica* species in Australia, canola may potentially hybridise under natural conditions with sexually compatible species that include: other *B. napus* groups or subspecies (including vegetables such as swedes, rutabaga and kale), *B. juncea, B. rapa* (wild turnip; includes vegetables such as turnip, Chinese cabbage and pak choi) and *B. oleracea* (wild cabbage; includes vegetables such as cauliflower, Brussels sprouts, kale and cabbage) (Salisbury, 2002). However, hybrids between *B. napus* and *B. oleracea* have been shown to be difficult to obtain (Ford et al., 2006).

69. Under open pollination conditions, naturally occurring hybrids between *B. napus* and the related weedy species *R. raphanistrum*, *H. incana* and *S. arvensis* have been reported at very low frequencies (Darmency and Fleury, 2000; Darmency et al., 1998; Salisbury, 2002), and are generally sterile or predominantly sterile (Salisbury, 2002).

70. Canola is widely grown as an oil seed crop in Australia, and the proposed trial sites are located in commercial canola growing regions. Commercial canola in these areas includes non-GM canola and GM canola authorised for commercial release. Most Australian canola crops are herbicide tolerant, with 4 herbicide tolerance traits available for commercial cultivation: triazine tolerance (non-GM), imidazolinone tolerance (non-GM), glyphosate tolerance (GM), or glufosinate tolerance (GM) (Brown, 2021; Matthews et al., 2021). Details of all GM canola varieties approved by the Regulator in Australia under a licence are available from the OGTR website. GM canola authorised by the Regulator as safe for anyone to grow in Australia without a licence is listed on the GMO Register.

5.5 Presence of similar genes and their products in the environment

71. *BCN2* and *BCN3* are modified sequences of the gene *CSN2* derived from cattle (*B. taurus*), which are present in the agricultural environment. The genetic sequence of the other part of the fusion protein constituent is derived from common plant species and closely related sequences naturally occur in all plants.

72. The *bar* gene is from the common soil bacterium *S. hygroscopicus*, which is widespread and prevalent in the environment.

73. Regulatory and localisation sequences are derived from common plants, a plant virus (CaMV) or a soil bacterium (*A. tumefaciens*) that are widespread in the environment. Although some of the regulatory sequences are derived from plant pathogens (*A. tumefaciens* and CaMV), they comprise only small parts of the total genomes and cannot of themselves cause disease.

Section 6 Relevant Australian and international approvals

6.1 Australian approvals

Approvals by the Regulator

74. The GM canola lines included in this application have not been previously approved for release in Australia.

Approvals by other government agencies

75. The GM canola lines included in this application have not been previously approved by any other government agencies in Australia.

6.2 International approvals

76. The GM canola lines included in this application have not received any approvals from authorities in other countries.

Chapter 2 Risk assessment

Section 1 Introduction

77. 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 3). 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.

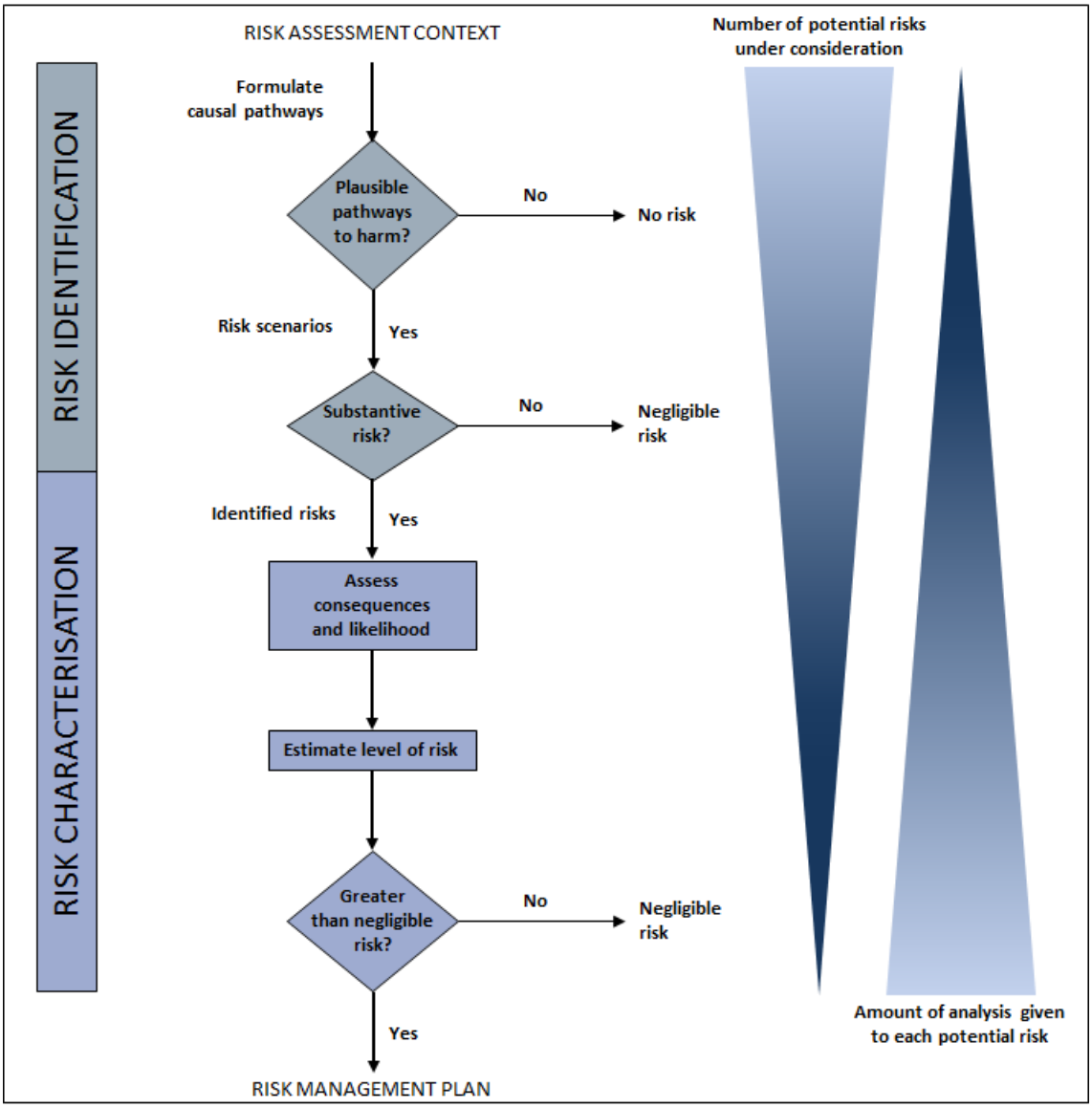

Figure 3. The risk assessment process

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

79. 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.

80. 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 4), i.e. the risk is considered to be no greater than negligible.

81. 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.

82. A weed risk assessment approach is used to identify traits that may contribute to risks from GM plants, as this approach addresses the full range of potential adverse outcomes associated with plants. In particular, novel traits that may increase the potential of the GMO to spread and persist in the environment or increase the level of potential harm compared with the parental plant(s) are used to postulate risk scenarios (Keese et al., 2014). Risk scenarios postulated in previous RARMPs prepared for licence applications for the same or similar GMOs, are also considered.

Section 2 Risk identification

83. Postulated risk scenarios are comprised of three components (Figure 4):

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.

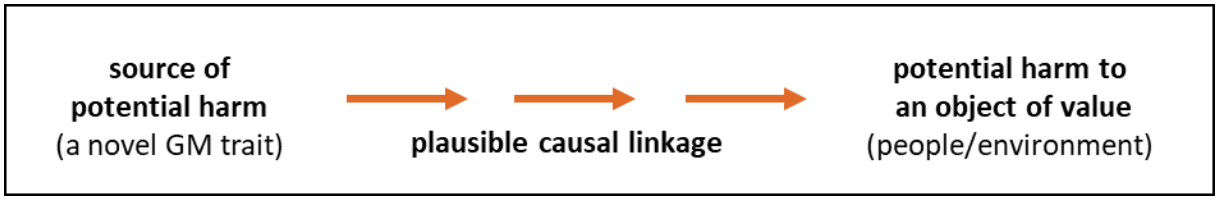

Figure 4. Components of a risk scenario

84. 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 restrict the spread and persistence of the GMOs
- the characteristics of the parent organism(s).

2.1 Risk source

85. The sources of potential harms can be intended novel GM traits associated with one or more introduced genetic elements, or unintended effects/traits arising from the use of gene technology.

86. As discussed in Chapter 1, the GM canola lines have been modified by the introduction of fusion protein sequence comprised of variants of *CSN2* from cattle and a gene derived from common plant species. The introduced genes are intended to produce dairy protein in seed. These introduced genes are considered further as sources of potential harm.

87. The GM canola lines also contain the *bar* gene which confers glufosinate herbicide tolerance, and was used as selectable marker genes. The *bar* and its protein product PAT have been extensively characterised and assessed as posing negligible risk to human or animal health or to the environment by the Regulator, as well as by other regulatory agencies in Australia and overseas. As the *bar* gene has not

been found to pose a substantive risk to either people or the environment, its potential effects will not be further considered for this application.

88. The introduced genes are controlled by introduced regulatory sequences derived from various species (see Table 3). Regulatory sequences and introns are naturally present in all plants, and the introduced sequences are expected to operate in similar ways to endogenous sequences. The regulatory sequences are DNA that is not expressed as a protein, so exposure is to the DNA only and dietary DNA has no toxicity (Delaney et al., 2018). Hence, potential harms from the regulatory sequences and introns will not be considered further. However, seed-specificity of promoters will be discussed in the context of other risk sources.

89. The genetic modifications involving introduction of genes have the potential to cause unintended effects in several ways. These include insertional effects such as interruptions, deletions, duplications or rearrangements of the genome, which can lead to altered expression of endogenous genes. There could also be increased metabolic burden due to expression of the introduced proteins, novel traits arising out of interactions with non-target proteins and secondary effects arising from altered substrate or product levels in biochemical pathways. However, these types of effects also occur spontaneously and in plants generated by conventional breeding. Accepted conventional breeding techniques such as hybridisation, mutagenesis and somaclonal variation can have a much larger impact on the plant genome than genetic engineering (Schnell et al., 2015). Plants generated by conventional breeding have a long history of safe use, and there are no documented cases where conventional breeding has resulted in the production of a novel toxin or allergen in a crop (Steiner et al., 2013). Therefore, the potential for the processes of genetic modification to result in unintended effects will not be considered further.

2.2 Causal pathway

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

- routes of exposure to the GMOs, the introduced gene(s) and gene product(s)
- potential exposure to the introduced gene(s) and gene product(s) from other sources in the environment
- the environment at the site(s) of release
- agronomic management practices for the GMOs
- spread and persistence of the GMOs (e.g. reproductive characteristics, dispersal pathways and establishment potential)
- tolerance to abiotic conditions (e.g. climate, soil and rainfall patterns)
- tolerance to biotic stressors (e.g. pests, pathogens and weeds)
- tolerance to cultivation management practices
- gene transfer to sexually compatible organisms
- gene transfer by horizontal gene transfer (HGT)
- unauthorised activities.

91. Although all of these factors are taken into account, some are not included in risk scenarios because they have been considered in previous RARMPs and are not expected to give rise to substantive risks.

92. The potential for horizontal gene transfer (HGT) from GMOs to species that are not sexually compatible, and any possible adverse outcomes, have been reviewed in the literature (Keese, 2008; Philips et al., 2022) and assessed in previous RARMPs. No risk greater than negligible was identified, due to the rarity of HGT events and because the gene sequences are already present in the environment and available for transfer via demonstrated natural mechanisms. Therefore, HGT will not be assessed further.

93. 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

94. Potential harms from GM plants are based on those used to assess risk from weeds (Keese et al., 2014; Virtue, 2008) including:

- harm to the health of people or desirable organisms, including toxicity/allergenicity
- reduced biodiversity for nature conservation
- reduced establishment or yield of desirable plants
- reduced products or services from the land use
- restricted movement of people, animals, vehicles, machinery and/or water
- reduced quality of the biotic environment (e.g. providing food or shelter for pests or pathogens) or abiotic environment (e.g. negative effects on fire regimes, nutrient levels, soil salinity, soil stability or soil water table).

95. Judgements of what is considered harm depend on the management objectives of the land where the GM plant may be present. A plant species may have different weed risk potential in different land uses such as dryland cropping or nature conservation.

2.4 Postulated risk scenarios

96. Four risk scenarios were postulated and screened to identify any substantive risks. These scenarios are summarised in Table 6 and examined in detail in Sections 2.4.1 – 2.4.4.

97. In the context of the activities proposed by the applicant and considering both the short and long term, none of the 4 risk scenarios gave rise to any substantive risks.

Table 6. Summary of risk scenarios from the proposed dealings with GM canola

Risk scenario	Risk source	Causal pathway	Potential harm	Substantive risk?	Reason
1	Introduced genes for dairy protein production	Cultivation of GM canola at trial sites ⬇ Exposure of people and desirable animals to products of the introduced genes	Increased toxicity or allergenicity for people OR increased toxicity to desirable animals	No	• The GM canola would not be used as commercial human food or animal feed • The short duration and proposed controls for the field trial would restrict exposure of animals to the GM plants through contact or consumption • The limits and controls of the field trial would restrict exposure of people to the GM plants • β-casein fusion proteins are not expected to be toxic but could be allergenic. However, people will not consume GM canola seeds or products (other than a small number of people as part of the sensory tests), and animals are unlikely to consume a dose that would cause toxicity.
2	Introduced genes for dairy protein production	Cultivation of GM canola at trial sites ⬇ Persistence of GM canola seed at trial sites or dispersal of GM seed outside trial limits ⬇ Establishment of populations of volunteer GM plants expressing the introduced genes in the environment	Increased toxicity or allergenicity for people OR increased toxicity to desirable animals OR reduced establishment or yield of desirable plants	No	• The limits and controls of the field trial would minimise dispersal or persistence of GM seeds • GM canola is susceptible to standard weed management measures • As discussed in Risk Scenario 1, no substantive risk was identified for increased adverse effects in people or toxicity to animals • Canola has limited ability to compete with other plants and the genetic modifications are not expected to alter the dispersal characteristics of the GM canola.

Risk scenario	Risk source	Causal pathway	Potential harm	Substantive risk?	Reason
3	Introduced genes for dairy protein production	Cultivation of GM canola at trial sites ⬇ Pollen from GM plants dispersed outside the trial sites ⬇ Outcrossing with sexually compatible plants ⬇ Establishment of populations of hybrid GM plants expressing the introduced genes in the environment	Increased toxicity or allergenicity for people OR increased toxicity to desirable animals OR reduced establishment or yield of desirable plants	No	• The controls of the field trial would minimise pollen flow to sexually compatible plants outside the trial sites • As discussed in Risk Scenario 1, no substantive risk was identified for increased adverse effects in people or toxicity to animals • As discussed in Risk Scenario 2, the genetic modifications are not expected to alter the dispersal characteristics of the GM canola.
4	Introduced genes for dairy protein production	Cultivation of GM canola at trial sites ⬇ Consumption of GM canola seed by pest animals ⬇ Increased fitness of pest animals	Reduced establishment of yield of desirable plants OR Reduced biodiversity	No	• The limited scale and other proposed limits and controls minimise exposure of pests to the GM seeds • GM canola seeds are unlikely to contribute a large proportion of the overall diet for pest species • Consumption of GM canola seed containing β-casein fusion proteins is unlikely to provide a fitness advantage to pest species • Pests are controlled by current pest management practices.

2.4.1 Risk scenario 1

Risk source	Introduced genes for dairy protein production
Causal pathway	⬇ Cultivation of GM canola at trial sites ⬇ Exposure of people and desirable animals to products of the introduced genes ⬇
Potential harm	Increased toxicity or allergenicity for people OR Increased toxicity to desirable animals

Risk source

98. The source of potential harm for this postulated risk scenario is the introduced genes for dairy protein production in GM canola plants.

Causal pathway

99. The GM canola would be grown at the trial sites. As the introduced genes for dairy protein production are controlled by a seed-specific promoter, the encoded fusion proteins would be produced in seeds of the GM plants. A review of the literature corroborates seed-specific expression of the promoter. A study utilising the promoter to express an introduced gene in a common plant species showed expression in seeds but not in leaf tissue. Trial staff would be exposed to the seeds during harvesting. Given the promoter is seed-specific, inhalation of pollen is not expected to lead to exposure of the introduced fusion proteins. The applicant has not tested the seeds or any other tissues for levels of produced fusion proteins.

100. People involved in the breeding, cultivating, harvesting, transporting and processing of the GM canola may be exposed to expressed proteins through contact with the GMOs, including direct contact with GM plant material. This would be expected to primarily occur at the trial site but could also occur anywhere the GM seeds are transported or used. The proposed limits and controls of the trial would minimise the likelihood that people or other organisms would be exposed to GM plant material. The GM canola is not proposed for use in commercial human food, and therefore, people are unlikely to be exposed to the introduced genetic elements or their products as a result of consuming GM canola seed. The applicant proposes that GM canola will only be handled by trained and authorised staff and all GM plant material would be transported in accordance with the Regulator's Guidelines for the Transport, Storage and Disposal of GMOs.

101. The applicant proposes human sensory testing of ingredients isolated from GM canola seed to assess the taste, smell and texture of the oil products and protein concentrates isolated from the seed. Although the products are not intended to be swallowed during testing, ingestion of small amounts may occur. People participating in sensory evaluations could also be exposed to protein concentrates enriched in β-casein fusion proteins by dermal contact, contact with mucous membranes or inhalation.

102. The applicant does not propose use of the GM canola in commercial animal feed, and livestock would not be permitted to graze the trial sites. Therefore, livestock are not expected to be exposed to GM plants grown at the trial sites. Desirable wild animals, such as native mammals and birds, could enter the trial sites and consume GM plants including seeds. The limited size and duration of the field trial would restrict the number of desirable wild animals exposed to GM plants grown at the trial sites.

Potential harm

103. If people or animals were exposed to the GM canola, the potential harms are increased toxicity or allergenicity to people or increased toxicity to desirable animals.

104. Toxicity is the adverse effect(s) of exposure to a dose of a substance as a result of direct cellular or tissue injury, or through the inhibition of normal physiological processes (Felsot, 2000). Allergenicity is the

potential of a substance to elicit an immunological reaction following its ingestion, dermal contact or inhalation, which may lead to tissue inflammation and organ dysfunction (Arts et al., 2006).

105. As mentioned in Chapter 1 Section 3, while non-GM canola is not generally regarded as allergenic or toxic to humans or animals, it does produce some allergens, toxins and anti-nutritional factors. No allergic reactions to processed canola oil have been reported in the literature, though there are reports of food allergies to *B. napus* seed extracts (Poikonen et al., 2006) and flour (Alvarez et al., 2001). As discussed in Chapter 1, Section 4.1, the proteins encoded by the introduced genes result in dairy protein production. There is no reasonable expectation that the introduced genes expressed in the GM canola would affect the pathways producing endogenous toxins or allergens in canola, or lead to the production of novel toxins or allergens.

106. As discussed in Chapter 1, Section 4.3, the introduced fusion proteins and genetic constructs have not been assessed for toxicity and allergenicity by any authorities or in animal feeding studies. However, some caseins and some proteins related to the second fusion protein constituent derived from common plant species are known food allergens. Allergy to milk proteins is most common in young children and decreases rapidly with age (Jensen et al., 2022). Allergenicity to casein proteins occurs via ingestion of the proteins. The GM canola seeds containing β-casein fusion proteins are not intended for human consumption and, as noted in paragraph 101, negligible levels of ingestion are likely during sensory assessment. Furthermore, allergic reactions to casein proteins via dermal contact are rare and often require a damaged skin barrier to cause an immune response (Jensen et al., 2022). In addition, the molecular weight of a compound must be less than 500 Da in size to penetrate intact skin (Bos and Meinardi, 2000). Bioinformatic analysis of the amino acid sequence for the endogenous *CSN2* protein from which the BCN2 and BCN3 sequences are derived indicate a molecular weight (independent of the fusion protein) of 18.62 kDa, which is too large to penetrate the skin. Therefore, the BCN2 or BCN3 β-casein protein fused to the second protein constituent is also expected to be too large to penetrate the skin.

107. There are few studies about toxicity of casein proteins in the literature and no reports related to humans were found. Canola seeds can be eaten by several animal species, including beetles, rodents and birds. Seed predation is greatest when seeds are buried at shallow depths, and mice can climb canola plants and feed on seed pods. Some reports indicate casein tolerance in laboratory animals. An early study suggested it is almost impossible to administer lethal amounts of casein orally to rats (Boyd et al., 1967). A high-casein diet was also shown to attenuate renal injury and inflammation in rats without adverse health effects (Shimada et al., 2020). Conversely, another study demonstrated that consumption of a high-casein diet for a 4-week period resulted in a twofold increase in colonic DNA damage in rats (Toden et al., 2005). However, it is unclear whether β-casein specifically is responsible for these genotoxic effects or whether this is the result of other caseins.

108. While studies have demonstrated allergenicity of milk proteins and caseins more broadly, no scientific evidence was identified to indicate allergenicity or toxicity of β-casein specifically in humans or animals. The genetic constructs proposed for insertion into GM canola are derived from the sequence for the A2 variant of β-casein, which is a common component of cow's milk consumed by humans and is not associated with gastrointestinal intolerance linked to the A1 variant.

109. Further information about the β-casein fusion protein relevant to consideration of this risk scenario has been declared CCI by the Regulator. Under section 185 of the Act, the CCI has been made available to the prescribed agencies and experts that are consulted on the RARMP for this application.

Conclusion

110. Risk scenario 1 is not identified as a substantive risk because the GM plant material would not be used as commercial human food and animal feed, the GM dairy fusion proteins are not expected to be toxic, and the proteins encoded by the introduced genes are too large to penetrate an intact skin barrier and cause allergenicity via dermal contact. Further, the limits and controls of the field trial would restrict exposure of people and desirable animals to the GM plants. Therefore, this risk could not be considered greater than negligible and does not warrant further detailed assessment.

2.4.2 Risk scenario 2

Risk source	Introduced genes for dairy protein production
Causal pathway	⬇ Cultivation of GM canola at trial sites ⬇ Persistence of GM canola seed at trial sites or dispersal of GM seed outside trial limits ⬇ Establishment of populations of volunteer GM plants expressing the introduced genes in the environment ⬇
Potential harm	Increased toxicity or allergenicity for people OR Increased toxicity to desirable animals OR Reduced establishment or yield of desirable plants

Risk source

111. The source of potential harm for this postulated risk scenario is the introduced genes for dairy protein production in GM canola plants.

Causal pathway

112. The GM canola would be grown at the trial sites. GM seeds could be physically dispersed outside the trial sites by human activity, animal activity, wind or water. GM seeds could also persist on trial sites after completion of the trial. These GM seeds could grow in the environment and establish populations of volunteer GM plants.

113. Viable GM canola seeds could be dispersed outside the trial sites by human activity, such as transport of seeds and movement of agricultural machinery. Inadvertent dispersal of seeds by people dealing with the GMOs would be minimised by cleaning of all equipment prior to removal from the trial sites. The applicant also proposes that areas used for equipment cleaning would be inspected for volunteers.

114. GM seeds could be dispersed outside the trial limits by animal activity. Canola seeds have no specific adaptions, such as burrs or hooks, for dispersal by animals (OGTR, 2024). Dispersal of canola seed via endozoochory (consumption and excretion of viable seed) by birds only occurs at very low levels (Twigg et al., 2008; Woodgate et al., 2011). Canola seeds could be transported short distances by hoarding animals, such as ants and mice. The applicant is also proposing to implement rodent control measures at trial sites.

115. Canola seeds lack specialised structures that would assist their dispersal by wind (OGTR, 2024). However, the GM canola may be windrowed prior to harvesting, and under strong wind conditions plant material containing seeds could disperse outside trial sites. The applicant proposes to minimise the likelihood of wind dispersal by implementing measures including ensuring high density of GMO plants prior to windrowing, using a windrow roller, or by appropriate site selection.

116. The GM canola seeds could be dispersed by water during flooding or heavy runoff, although seeds are unlikely to remain viable after prolonged exposure to water (OGTR, 2024). To minimise the potential for seed dispersal during flooding, the applicant proposes to locate the trial at sites that are not prone to flooding.

117. During harvest of the GM canola, a small percentage of the seeds are expected to remain on the trial sites. Persistence of GMOs at the trial sites after the field trial is finished could occur if seeds in the seed bank were dormant. Canola generally does not exhibit primary dormancy, but secondary dormancy has been described (OGTR, 2024). A study in western Canada revealed that secondary seed dormancy

prolonged persistence of volunteer canola plants (Gulden et al., 2003). Persisting canola seed banks have been shown to significantly contribute to the dynamics of feral canola populations (Pivard et al., 2008). A long-term monitoring study in Germany detected GM canola volunteers in arable fields for up to 15 years after the field trial concluded, but did not detect spatial dispersion (Belter, 2016). In Australia, volunteers can be found for up to 3 years after growing canola due to persistence in seed banks, though the majority of volunteer seedlings emerge the year following a canola crop (AOF, 2019).

118. To minimise persistence of GM seeds on the trial sites, the applicant proposes to promote seed germination by light post-harvest tillage and irrigation. During a post-harvest monitoring period, the applicant proposes to inspect the trial sites at least once every 35 days for at least 24 months, and destroy any canola volunteers, until the site is free from volunteers for at least 12 months. The suitability of the proposed controls to manage GM seed dispersal and persistence is discussed in detail in Chapter 3, Section 3.1. These control measures are expected to minimise persistence of viable GM canola seeds on the trial sites.

119. If GM canola seeds were dispersed outside trial limits, it is unlikely that they would establish ongoing volunteer populations. Even in environments without active weed management, volunteer canola populations along transportation routes rely on recurrent spillages to persist (Yoshimura et al., 2006) and volunteer canola dispersed into natural areas was reported to rapidly become extinct (Busi and Powles, 2016).

120. In agricultural areas of Australia where canola is grown, volunteer populations are controlled by a range of weed management measures. Effective methods for control of canola volunteers include grazing, mowing, cultivation and application of a range of knockdown or selective herbicides (AOF, 2019). The introduced genetic modifications are not expected to affect the susceptibility of GM volunteers to standard weed management measures. Some of the canola lines will contain the *bar* gene and be tolerant to glufosinate herbicide. However, as discussed in Chapter 1, Section 5.3, glufosinate herbicide is not routinely used for controlling volunteer canola (AOF, 2019).

121. The applicant states that GM canola grown in controlled glasshouse conditions did not display any differences in growth patterns, morphology and fertility compared to non-GM canola. Both GM and non-GM canola exhibited similar germination rates, time to flowering, height, and seed count per plant. It is not expected that the production of β-casein fusion proteins in canola would affect seed yield, viability or germination, and no information was found in the literature to suggest casein production in plants would have such effects. Though the applicant does not anticipate that the introduced genetic modifications would affect seed dormancy of the GM canola, or the overall ability of GM canola volunteers to survive in the environment, this is an area of uncertainty. In the unlikely event of increased dormancy or dispersal, the imposed control measures, including post-harvest monitoring for volunteers, would restrict GM seed dispersal and persistence.

Potential harm

122. If the GM canola entered the Australian environment, the potential harms are increased toxicity or allergenicity to people, increased toxicity to desirable animals, or reduced establishment or yield of desirable plants.

123. As discussed in Risk scenario 1, no substantive risk was identified for increased toxicity or allergenicity of the GM canola for people or increased toxicity to desirable animals. Although the β-casein fusion protein constituents may be associated with allergenicity, the GM plant material would not be used as commercial human food and animal feed, and the proteins encoded by the introduced genes are too large to penetrate an intact skin barrier to cause allergenicity via dermal contact. The limits and controls of the field trial would further restrict exposure to the GM canola and any associated toxicity or allergenicity.

124. Populations of volunteer GM canola could reduce establishment or yield of desirable plants. GM volunteers could directly compete with agricultural crops, pastures or native vegetation. GM volunteers could also reduce the yield of commercial canola crops by providing a reservoir for pathogens, such as the important fungal diseases blackleg and stem rot (see Chapter 1, Section 5.2). No information could be found to suggest that the introduced genetic modifications are likely to alter the susceptibility of the GM

canola to pathogens. In the unlikely event that the introduced genetic modifications alter the susceptibility of the GM canola to pathogens, it is expected to be minor given expression of the fusion proteins is restricted to seeds.

125. All domesticated crop species are expected to be poor competitors with pasture species or established native vegetation. Canola is considered a less competitive crop species than wheat or barley (GRDC, 2011), which are the main crops grown in NSW, Vic, SA and WA (ABARES, 2024b). Therefore, canola volunteers have limited ability to compete with desirable plants. As discussed in Chapter 1, Section 4.4, the applicant did not identify any differences in morphological or growth characteristics in the GM canola plants grown in the glasshouse, compared to non-GM canola. Therefore, the GM canola plants are not expected to show increased spread or persistence in the environment. However, given the GM plants have not yet been grown in the field, it is uncertain whether the introduced genetic modifications would increase their overall competitiveness. In addition, no information could be found to suggest that the introduced genetic modifications would enable the GM canola to produce allelopathic substances which would negatively affect plant establishment around them. Further, any allelopathic substances produced would be expected to be restricted to seeds by the seed-specific promoter. A standard condition of a licence for a field trial would be that the applicant immediately notify the OGTR of any unintended effects, including changes that would result in increased weediness or seed dormancy.

126. The genetic modifications are not expected to affect the susceptibility of GM volunteers to standard weed management measures. Although some of the GM canola lines will contain the *bar* gene and be tolerant to glufosinate herbicide, as discussed in Section 5.3, glufosinate herbicide will not be applied to the GM canola plants as part of this trial and other methods are available to manage GM canola volunteers.

Conclusion

127. Risk scenario 2 is not identified as a substantive risk because the proposed limits and controls of the field trial would minimise dispersal or persistence of GM seeds, the GM canola is susceptible to standard weed management measures, and the genetic modifications are not expected to increase the overall weediness or competitiveness of the GM canola with other plants. Therefore, this risk could not be considered greater than negligible and does not warrant further detailed assessment.

2.4.3 Risk scenario 3

Risk source	Introduced genes for dairy protein production
Causal pathway	⬇ Cultivation of GM canola at trial sites ⬇ Pollen from GM plants dispersed outside the trial sites ⬇ Outcrossing with sexually compatible plants ⬇ Establishment of populations of hybrid GM plants expressing the introduced genes in the environment ⬇
Potential harm	Increased toxicity or allergenicity for people OR Increased toxicity to desirable animals OR Reduced establishment or yield of desirable plants

Risk source

128. The source of potential harm for this postulated risk scenario is the introduced genes for dairy protein production in GM canola plants.

Causal pathway

129. The GM canola would be grown at the trial sites. Pollen from the GM plants could be transported outside the trial site by wind or insect vectors and could fertilise sexually compatible plants. Hybrid seeds containing the introduced genes could be harvested or could grow as volunteers.

130. It should be noted that vertical gene flow *per se* is not considered an adverse outcome, but may be a link in a chain of events that may lead to an adverse outcome.

131. Canola is primarily self-pollinating, but approximately 30% of seeds are produced by cross pollination. Outcrossing decreases rapidly with distance, with the majority of cross-pollination occurring over distances less than 10 m (OGTR, 2024). The introduced genetic modifications are not expected to affect the pollen dispersal characteristics of the GM canola.

132. The GM canola could outcross with nearby canola crops or volunteers, if there is synchronicity of flowering. As discussed in Chapter 1, Section 5.4, canola can also occasionally hybridise with the related horticultural crops *B. juncea, B. oleracea* and *B. rapa* and the related weeds *H. incana, R. raphanistrum* and *S. arvensis*.

133. The applicant has proposed control measures to minimise pollen flow from GM plants growing on the trial sites to sexually compatible plants outside the trial sites (Chapter 1, Section 2.2). Each planting area would be surrounded by a monitoring zone which would be inspected for sexually compatible plants from before the GM canola flowers and until harvest. The applicant also proposes the planting area will be surrounded by a large isolation zone, or a smaller isolation zone combined with use of either a pollen trap or an insect-proof tent. In addition, any GM volunteers growing on the trial sites after harvest would be destroyed prior to flowering. The suitability of the proposed controls to manage pollen flow is discussed in detail in Chapter 3, Section 3.1. These controls are expected to minimise pollen flow from the GM canola to sexually compatible non-GM plants outside the trial sites.

134. If pollen from GM plants fertilised plants in a commercial canola crop, hybrid GM seeds could be harvested for human food and animal feed, or be replanted in a crop. However, even in the complete absence of measures to restrict pollen flow, outcrossing rates between neighbouring commercial canola fields are less than 0.1% under Australian conditions (Rieger et al., 2002). Therefore, the planting seed described in this risk pathway could only contain a very low proportion of hybrid GM seed, so people and desirable animals could only be exposed to very low levels of the hybrid GMOs.

135. If pollen from GM plants fertilised sexually compatible plants growing as crops, volunteers or weeds, the hybrid GM seeds could grow as volunteers. Populations of hybrid GM volunteers could be consumed by desirable animals or could reduce the establishment or yield of desirable plants.

136. It is not expected that the intended modifications would change the pollination characteristics of the GM canola to increase the likelihood of pollination of non-GM canola or related species.

Potential harm

137. The potential harms from this risk scenario are adverse health effects in people and/or desirable organisms, or reduced establishment or yield of desirable plants.

138. As discussed in Risk scenario 1, no substantive risk was identified for increased toxicity or allergenicity of the GM canola for people or increased toxicity to desirable animals. Similarly, in the rare event of outcrossing between the GM canola and sexually compatible plants, the proteins encoded by the introduced genes are too large to penetrate an intact skin barrier to cause allergenicity via dermal contact.

139. As discussed in Risk scenario 2, the GM canola is not likely to be weedier or more competitive than non-GM canola. Similarly, in hybrids between the GM plants and sexually compatible plants, the genetic modifications are not expected to confer an overall increased ability to compete with other plants.

Conclusion

140. Risk scenario 3 is not identified as a substantive risk because the controls of the field trial would minimise pollen flow to sexually compatible plants outside the trial sites. GM hybrids are not likely to differ from the GM canola, for which Risk scenarios 1 and 2 did not identify adverse health effects in people, toxicity in animals or weediness as substantive risks. Therefore, this risk could not be considered greater than negligible and does not warrant further detailed assessment.

2.4.4 Risk scenario 4

Risk source	Introduced genes for dairy protein production
Causal pathway	⬇ Cultivation of GM canola at trial sites ⬇ Consumption of GM canola seed by pest animals ⬇ Increased fitness of pest animals ⬇
Potential harm	Reduced establishment or yield of desirable plants OR Reduced biodiversity

Risk source

141. The source of potential harm for this postulated risk scenario is the introduced genes for dairy protein production in GM canola plants.

Causal pathway

142. The GM canola would be grown at the trial sites. The GM canola produces seeds with dairy protein production.

143. Pest animals, such as rodents, larger mammalian species, or birds may ingest the GM canola seed at the trial site and may have a fitness advantage as a result of consuming the GM canola seeds. Populations of these pests may then increase as a consequence of this increased fitness.

144. As discussed in Risk scenario 1, insects, rodents and birds may consume canola seeds. The applicant has proposed the use of insect proof tents as one option for the planting area. If this were used, access to GM canola by insects would be limited by the insect-proof tent. The applicant has also proposed use of pesticide treatment to control insects as required. As noted in Risk scenario 3, the applicant has proposed rodent control measures that would limit rodent access.

145. If animals did consume GM canola seeds, it is likely that the seed will only make up a subset of the animal's overall diet, and the GM seed will only be available for a short period of time before harvest and until any seed on the soil surface is buried by tilling.

Potential harm

146. The potential harms from this risk scenario are reduced establishment or yield of desirable plants or reduced biodiversity.

147. If pests consuming the GM canola seeds with expression of β-casein fusion proteins had a fitness advantage over those which did not consume the GM canola seeds, populations could increase to a greater extent than expected. In that case, they may have a greater negative effect on native or other desirable plants, or on desirable animals. This could occur via reduced establishment or yield of native or other desirable plants due to increased consumption by greater pest animal populations. In natural environments, this may result in the loss of biodiversity. In agricultural areas this may result in reduced

crop yields. They might also increase competition with desirable animals for food sources or territory and thereby reduce animal biodiversity.

148. While there is a body of literature supporting caseins as a source of amino acids and their role in muscle growth in humans, there is limited evidence to show similar growth benefits in animals consuming caseins. A study in which dairy cows were provided 300 g casein per day for 2 weeks via infusion found that a casein-infused diet increased amino acid concentrations and milk production compared to cows fed grass silage alone (Vanhatalo et al., 2003). Another study in malnourished rats determined that re-feeding with a casein-infused diet increased bone strength and body weight catch-up compared to a whey-infused diet, but was insufficient to return the rats to a normal body weight (Masarwi et al., 2016). However, no studies were found to suggest a casein-infused diet leads to enhanced fitness or competitiveness of animals.

149. Birds may consume the GM canola seeds containing β-casein fusion proteins, however any increased fitness advantage to birds consuming the seeds is expected to be minor and would not be inherited by subsequent generations.

150. If enhanced fitness occurred in pest animals that consumed the GM canola seed, the degree of the improvement is uncertain, but expected to be minor and transient; would be confined to each individual animal; and is unlikely to change the existing adverse impact of known pest animals if consumed. Additionally, the limits and controls of the trial proposed by the applicant are proposed to limit the consumption GM canola seeds by pest species. The details of those control measures are discussed in Chapter 3. Based on these factors, it is considered that the increased levels of β-casein fusion proteins in the GM canola seeds is unlikely to provide a fitness advantage that would increase the existing impact of known pest animals, though this is an area of uncertainty.

Conclusion

151. Risk scenario 4 is not identified as a substantive risk due to the lack of evidence to suggest any fitness advantage to pest animals as a result of consuming higher levels of β-casein fusion proteins, and the proposed limits and controls to restrict exposure of animals to the GMOs, including pest management practices. Further, GM seed consumption is unlikely to cause a sustained benefit for animals. Therefore, this risk could not be considered greater than negligible and does not warrant further detailed assessment.

Section 3 Uncertainty

152. 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.

153. 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.

154. As field trials of GMOs are designed to gather data, there are generally data gaps when assessing the risks of a field trial application. However, field trial applications are required to be limited and controlled. 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.

155. For DIR 215, uncertainty is noted particularly in relation to:

- potential for increased toxicity to livestock/other desirable organisms or increased allergenicity to people of the GM canola
- potential for the genetic modifications to increase plant competitiveness and survival
- potential for the genetic modifications to increase competitiveness of pest species if consumed.

156. 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 these GMOs.

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

Section 4 Risk evaluation

158. 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.

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

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

160. Four 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 6 and include:

- limits on the size and duration of the proposed release
- suitability of controls proposed by the applicant to restrict the spread and persistence of the GM canola plants and their genetic material
- the products of the introduced genes are not expected to be toxic
- GM canola plant material is not expected to confer increased fitness to pest species
- none of the GM plant material would enter commercial human food or animal feed.

161. Therefore, risks to the health and safety of people, or the environment, from the proposed release of the GM canola 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.

Chapter 3 Risk management plan

Section 1 Background

162. 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.

163. 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.

164. All licences are subject to 3 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.

165. 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

166. The risk assessment of risk scenarios listed in Chapter 2 concluded that there are negligible risks to people or the environment from the proposed field trial of GM canola. These risk scenarios were considered in the context of the scale of the proposed release (Chapter 1, Section 15), the proposed controls (Chapter 1, Section 2.2), and the receiving environment (Chapter 1, Section 5), 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

167. 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 licence.

3.1 Limits and controls on the release

168. Sections 15 and 2.2 of Chapter 1 list the limits and controls proposed by the applicant. Many of these are discussed in the 4 risk scenarios considered in Chapter 2. The appropriateness of these limits and controls is considered further in the following sections.

3.1.1 Consideration of limits proposed by the applicant

169. The applicant proposes that the field trial would take place between May 2025 and December 2029. This would limit the duration of the trial to 4 years and 8 months. The applicant has also proposed that GM canola would be grown at a maximum of 2 sites per year in 2025, 5 in 2026, 10 in 2027, 15 in 2028 and

20 in 2029. Across all sites, GM canola is proposed to be grown on planting areas up to a combined total area of 1 ha in 2025, 5 ha in 2026, 25 ha in 2027, 105 ha in 2028, and 300 ha in 2029 – a total of 436 ha over the duration of the licence. The applicant proposes up to 7 ha per site in 2028 and 15 ha per site in 2029. However, monitoring such a large area could be challenging and, at this stage, the applicant does not have experience monitoring large sites for GM field trials. Therefore, it is considered appropriate to limit the maximum combined planting area for each site to 5 ha in 2028 and 2029 (a maximum of 206 ha over the duration of the licence, if issued). This would ensure effective monitoring for volunteer GM canola on each site. The size and short duration of the trial would restrict the exposure of people and desirable animals to the GMOs (Risk scenario 1).

170. The applicant proposes that only authorised and trained people would be permitted to deal with the GMOs. Standard licence conditions included in the licence state that only people authorised by the licence holder are covered by the licence and permitted to deal with the GMOs. In addition, the licence holder must inform all people dealing with the GMOs of relevant licence conditions. These measures would ensure that the field trial is conducted in accordance with the specified limits and controls (important for all risk scenarios).

3.1.2 Consideration of proposed controls regarding exposure to the GMOs

171. The applicant proposes to grow both GM canola and non-GM canola in the trial sites. The licence limits the plants that can be intentionally grown in the planting area to the GMOs, non-GM canola, and any plants approved in writing by the Regulator. As non-GM canola may be mingled with or fertilised by GM canola, a standard licence condition has been imposed requiring non-GM canola plants grown in a trial site to be treated as if they are GMOs. This measure manages the dispersal or persistence of GM seed (Risk scenario 2).

172. The applicant proposes that GM plants or products from the GM plants would not be used in commercial human food or animal feed, and this requirement has been included in the licence. This condition would maintain the risk context by restricting the exposure of people and desirable animals to the GMOs via consumption (Risk scenario 1) and would also minimise dispersal of the GMOs by livestock or during transport or processing for human food or animal feed use (Risk scenario 2).

173. The GM canola is not expected to be toxic (Chapter 1 Section 4.3), so there are no specific controls proposed to manage increased toxicity to people or animals. General controls included in the licence will limit exposure of people to the GMOs.

174. Any human sensory testing must be approved by a Human Research Ethics Committee (HREC) in accordance with the National Statement on Ethical Conduct in Human Research. This condition would maintain the risk context by ensuring exposure of people to products derived from the GM canola would be conducted under the oversight of a HREC. This ensures (among other considerations) that those conducting sensory testing would be required to consider risks and benefits of the research, to consider appropriate exclusion criteria for participants, and that participants are informed of risks, including potential allergenicity, and informed consent is provided prior to testing.

175. People who are allergic to the constituents of the fusion protein could have an allergic reaction to GM canola as a result of the genetic modification. The applicant has proposed to restrict trial site access to authorised personnel. Due to the limited scale of the proposed trial, a limited number of people would be exposed to the GM canola. Consequences of allergic reactions to these proteins can be severe, including anaphylaxis. Therefore, an additional licence condition is included to not engage personnel with a known allergy to the proteins expressed as a result of modifications in the GM canola to conduct dealings that may expose them to the GM canola. It is considered that the imposed condition is appropriate to protect people with known allergies to these proteins from contact with GM canola seed (Risk scenario 1).

176. The applicant has proposed fencing around trial sites to restrict access by large animals. This would limit exposure to GM canola to large animals through direct contact with plant material (e.g. through grazing). If consumed, potential harm to desirable animals from the introduced genetic elements is expected to be minimal. In addition, the licence requires that the GMOs must not be used in a way that

results in its use as animal feed, which would restrict access to the GMOs by animals. Therefore, the licence does not impose the use of fencing at trial sites.

3.1.3 Consideration of proposed controls regarding pollen flow from the GMOs

177. The applicant has proposed 3 different planting options to control pollen flow from the trial sites while the GMOs are flowering (Chapter 1, Figure 2).

178. The first option is to cover the planting area with an insect-proof tent, and to surround the planting area with a 10 m monitoring zone and a 390 m isolation zone (Chapter 1, Figure 2a). The tents would be in place at least 7 days before flowering until the GMOs complete flowering, and would be inspected for damage fortnightly and after any extreme weather event. The tents are expected to minimise the likelihood of insect-mediated pollen flow. The tents may also reduce wind-mediated pollen flow as it is expected that surrounding the GMOs with a tent would lessen air flow across the GMOs. Therefore, the use of an insect-proof justifies a reduced monitoring zone of 10 m and an isolation zone of 390 m. This option has been included in licences for previous GM canola field trials (e.g. DIR 188) and is considered an effective means of restricting pollen flow from canola.

179. The second option is to surround the planting area with a 15 m pollen trap of non-GM canola plants, a 35 m monitoring zone and a 350 m isolation zone (Chapter 1, Figure 2b). The pollen trap would be comprised of a mix of early, mid and late flowering non-GM canola to ensure synchronised flowering between the pollen trap and the GM canola. Pollen trap plants may provide sufficient forage for incoming pollinating insects so that they do not visit the GM plants, and any insects that reach the GM plants are expected to deposit most GM pollen on pollen trap plants while exiting the trial site. Pollen trap plants may also absorb some pollen dispersed by wind. The applicant has proposed an isolation zone of 390 m. As discussed in previous RARMPs for GM canola field trials (e.g. DIR 164, DIR 188 and DIR 205), the use of a pollen trap justifies a reduced isolation zone of 350 m and thus an overall distance of 400 m between the GMOs and any crops of related species.

180. The third option is to surround the planting area with a 50 m monitoring zone and a 950 m isolation zone (Chapter 1, Figure 2c). A combined isolation distance of 1 km as proposed in this trial setup is considered appropriate where pollen tents or pollen trap crops are not used, or when a pollen trap fails to function (e.g. failure to grow to a required density, or to form a continuous barrier, or to flower at the same time as the GM plants) or the insect-proof tent fails. The Canadian Regulations and Procedures for Pedigreed Seed Crop Production (CSGA, 2022) require that foundation production of male sterile *B. napus* seed must be separated from other *B. napus* plants by an 800 m isolation distance, of which the first 50 m must be practically free from related plants, and the remaining distance must be reasonably free from related plants. Therefore, the proposed 50 m monitoring zone and combined 1 km isolation distance, which are more stringent than these Canadian requirements, are considered effective measures to restrict pollen flow from the GM canola. This option has been included in licences for previous GM canola field trials (e.g. DIR 164 and DIR 188) and is considered an effective means of restricting pollen flow from canola.

181. All 3 planting options are considered appropriate and are included in the licence. For all 3 options, a licence condition requires that the monitoring zone would be inspected at least once every 35 days from 14 days prior to flowering of the GMOs until the GMOs are harvested, to ensure that it is free from any sexually compatible plants. This condition also requires that the isolation zone would be inspected at least once every 35 days from 14 days prior to flowering of the GMOs until the GMOs complete flowering, to ensure that it is free from intentionally planted sexually compatible plants. GM canola plants would not be planted at a trial site if any plants that are sexually compatible with canola were being grown in the monitoring or isolation zones.

182. The imposed measures to control pollen flow would minimise outcrossing between the GMOs grown on the trial sites and sexually compatible plants growing outside the trial sites (Risk scenario 3).

183. After harvest of the trial sites, the applicant proposes to inspect for volunteer canola plants at least once every 35 days for at least 24 months and until the site is free of volunteers for at least 12 months. Identified volunteer plants would be destroyed prior to flowering which would minimise the likelihood of further pollen flow. These post-harvest inspections are required in the licences for other GM canola field

trials and are considered an effective means of restricting pollen flow from GM canola volunteers to plants outside the trial sites (e.g. DIR 164 and DIR 188). These post-harvest inspections are considered appropriate to manage pollen flow and are included in the licence.

3.1.4 Consideration of proposed controls regarding dispersal of the GMOs

184. The applicant proposes that any equipment used with the GMOs would be cleaned as soon as practicable and before use for any other purpose, to avoid movement of viable plant material together with equipment. Personnel and clothing will also undergo physical examination before leaving the trial site to minimise unintentional movement of GM material. The applicant would contain the GM seeds during transport and storage in accordance with the Regulator's Guidelines for the Transport, Storage and Disposal of GMOs. These measures for the handling of GMOs would minimise exposure of people and other desirable organisms to the GMOs (Risk scenario 1), and dispersal of GMOs into the environment (Risk scenario 2) during transport, and have been included in the licence.

185. The applicant proposes to not locate the trial site in a flood-prone area in order to minimise the chance of viable plant material being washed away from the sites. This has been included as a condition in the licence. The licence also requires the trial sites to be located at least 50 m away from waterways as a standard licence condition for canola licences and that any extreme weather events must be reported to the Regulator. These measures would minimise dispersal of GM seeds by flooding (Risk scenario 2).

186. The GM canola seeds could be dispersed short distances from the trial sites during sowing, windrowing or harvest activities; by pod shattering, by seed-hoarding behaviours of animals such as ants or rodents; or by strong winds or runoff after heavy rain. As described in Section 3.1.3, the planting areas would be surrounded by monitoring zones that are inspected while the GMOs are growing, so volunteers growing from dispersed GM seeds during this period would be detected and destroyed. Specific conditions to minimise dispersal of GM plant material from windrowed plants by wind or rain have also been included in the licence. The applicant also proposes to inspect the monitoring zones after harvest to destroy any volunteers growing from dispersed GM seeds. As the short-distance seed dispersal mechanisms listed above are unlikely to transport seeds further than 10 m from the trial sites, the licence only requires post-harvest inspections of the innermost 10 m of the monitoring zone.

187. The applicant proposes to control the number of rodents present at trial sites. This would limit the potential dispersal of GMOs outside the trial sites (Risk scenario 2). As discussed above, rodents are unlikely to transport seeds further than 10 m from the trial sites. Post-harvest inspection of the innermost 10 m of the monitoring zone is considered sufficient to detect volunteers growing from rodent-mediated dispersal. Therefore, additional rodent control measures are not included in the licence.

188. The licence includes additional conditions to manage short-distance dispersal of GM seeds. These include requiring the trial site to be cleaned within 14 days after harvest by a method that removes GM seeds from the soil surface, and requiring post-harvest inspections of any area used to clean equipment or any other area where GMOs are known to have dispersed. This combination of controls would minimise short-distance dispersal of GM seeds leading to establishment of volunteer populations outside the trial sites (Risk scenario 2).

3.1.5 Consideration of proposed controls regarding persistence of the GMOs

189. After harvest, the applicant proposes to destroy GMOs not required for further evaluation or future trials. This would involve both cleaning the trial site within 14 days after harvest in a manner that destroys any surviving GMOs and destroying any harvested GM seed that is not required for experimentation or future planting. The measures are considered appropriate and have been included in the licence.

190. The applicant has proposed that GMOs would be destroyed by destructive analysis (e.g. ground up, hammer milled and/or roller milled), herbicide application, root cutting and shredding/mulching, uprooting, burning/incineration, light tillage to a depth of no more than 5 cm, autoclaving, or seed burial to a depth of at least 1 m. These methods are considered effective for rendering canola plants and/or seed non-viable, and have been included in the licence. To ensure the effectiveness of destruction by seed

burial, a licence condition specifies how this must be carried out, including a requirement that seeds must be wet at time of burial to encourage decomposition.

191. As discussed in Section 3.1.2, the applicant proposes to also grow non-GM canola on the trial sites, which would be treated as if they are GMOs. Non-GM canola in the trial site may be cross-pollinated by the GM canola, resulting in hybrid seeds. Therefore, it is appropriate to require non-GM canola to be destroyed in the same manner as GM canola, to manage persistence of the GMOs. This measure is included in the licence.

192. As detailed in Section 3.1.3, the applicant proposes to inspect the trial sites after harvest and destroy any identified volunteers. In minimum-tillage Australian farms, the canola seedbank is reported to decline rapidly, and no viable seed was recovered from the seedbank by 2.5 years after canola harvest (Baker and Preston, 2008). Similarly, OGTR monitoring data for 9 GM canola trial sites planted in 2015 found that in most sites no canola volunteers emerged more than one year after harvest and no volunteers emerged at any site more than 2.5 years after harvest. Therefore, the proposed monitoring for least 24 months, and until the site is free of volunteer canola plants for at least 12 months, is considered to be appropriate. This monitoring duration was also required for previous GM canola field trials and is considered effective for managing persistence of canola seed (e.g. DIR 164, DIR 188 and DIR 205).

193. To deal with the case of failed crops that are not harvested, licence conditions require that GMOs must be harvested or destroyed within 8 months after planting, and that if all GMOs in a planting area have been destroyed, then the area is considered to have been harvested and cleaned.

194. The applicant also proposes that canola volunteers identified during inspections would be destroyed prior to flowering, which would minimise the likelihood of GM canola seed dispersal (Risk scenario 2) and pollen flow to non-GM plants outside the trial site (Risk scenario 3). This measure is considered appropriate and has been included in the licence conditions.

195. The applicant proposes shallow cultivation of the trial sites to encourage seed germination. The licence conditions require that tillage depth would be no greater than 5 cm, to avoid deep burial of seed that could induce dormancy. The first tillage would occur within 60 days after harvest and the final tillage would occur during the volunteer-free period prior to sign-off. To ensure that the final tillage produces conditions that are conducive to germination of volunteers, the licence requires this tillage at a time when specified levels of rainfall or irrigation occur to provide sufficient moisture to the seedbank (Attachment B of the licence).

196. The combination of control measures described in this section would minimise the persistence of GM seeds leading to establishment of GM volunteer populations in the environment (Risk scenario 2).

3.1.6 Summary of licence conditions to be implemented to limit and control the release

197. A number of licence conditions are imposed to limit and control the release, based on the above considerations. These include requirements to:

- limit the duration of the release to the period from May 2025 to December 2029
- limit the size of the release to a maximum of 2 sites per year in 2025, 5 in 2026, 10 sites in 2027, 15 sites in 2028 and 20 sites in 2029, with planting areas up to a combined total area of 1 ha in 2025, 5 ha in 2026, 25 ha in 2027, 75 ha in 2028, and 100 ha in 2029
- limit the location of the release to nominated local government areas in NSW, Vic, WA and SA
- not allow GM plant material to be used in commercial human food or animal feed
- treat any non-GM canola grown in planting areas like the GMOs
- not permit persons with an allergy to the proteins produced as a result of the modification, or related proteins to conduct dealings that may expose them to GM plant material
- control pollen flow from the trial sites using one of the following options:

a. cover the planting area with an insect proof tent, and surround the planting area with a monitoring zone of 10 m and an isolation zone of a further 390 m, or

b. surround the planting area with a pollen trap of 15 m, a monitoring zone of 35 m and an isolation zone of a further 350 m, or

c. surround the planting area with a monitoring zone of 50 m and an isolation zone of a further 950 m

- locate trial sites at least 50 m from any natural waterways
- transport and store the GMOs in accordance with the Regulator's guidelines
- destroy all GMOs not required for further evaluation or future trials
- clean equipment used with the GMOs before use for any other purpose
- clean the planting areas, monitoring zones, areas where equipment has been cleaned, and other areas where GMOs are known to have dispersed after harvest
- apply any measures to promote the germination of any canola seeds that may be present in the soil after harvest, including watering and shallow tillage
- monitor each trial site at least once every 35 days for at least 24 months after harvest and until no volunteers are identified for at least 12 months, and destroy any canola plants that emerge.

3.2 Other risk management considerations

198. 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

199. 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.

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

201. In addition, Miruku must have access to an Institutional Biosafety Committee (IBC) and be an accredited organisation under the Act.

3.2.2 Contingency plan

202. If a licence were issued, Miruku would be required to submit a contingency plan to the Regulator before planting the GMOs. This plan would detail measures to be undertaken in the event of any unintended presence of the GM canola outside permitted areas.

203. Before planting the GMOs, Miruku would also be required to provide the Regulator with a method to reliably detect the GMOs or the presence of the genetic modifications in a recipient organism.

3.2.3 Identification of the persons or classes of persons covered by the licence

204. If issued, the persons covered by the licence would be the licence holder and employees, agents or contractors of the licence holder and other persons who are, or have been, engaged or otherwise authorised by the licence holder to undertake any activity in connection with the dealings authorised by the licence. Prior to growing the GMOs, Miruku would be required to provide a list of people and organisations that will be covered by the licence, or the function or position where names are not known at the time.

3.2.4 Reporting requirements

205. 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 field trial.

206. A number of written notices would also be 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 and actual dates of planting
- details of areas planted to the GMOs
- expected dates of flowering
- expected and actual dates of harvest and cleaning after harvest
- details of inspection activities.

3.2.5 Monitoring for compliance

207. 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. Post-release monitoring continues until the Regulator is satisfied that all the GMOs resulting from the authorised dealings have been removed from the release sites.

208. 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.

209. 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 of 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

210. Additional information has been identified that may be required to assess an application for a commercial release of the GM canola, or to justify a reduction in limits and controls.

211. This includes:

- additional molecular and biochemical characterisation of the GM canola lines, particularly with respect to expression of the introduced genes and proteins in the seeds

- biochemical characterisation of the GM canola lines, particularly with respect to potential for allergenicity related to the introduced genetic elements
- additional phenotypic characterisation of the GM canola lines leading to potential for increased weediness.

Section 5 Conclusions of the consultation RARMP

212. The risk assessment concludes that the proposed limited and controlled release of GM canola 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.

213. However, 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, as these were important considerations in establishing the context for assessing the risks.

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Appendix A: Summary of submissions from prescribed experts, agencies and authorities on the consultation RARMP

The Regulator received a number of submissions from prescribed experts, agencies and authorities[4] on the consultation RARMP. All issues raised in submissions relating to risks to the health and safety of people and the environment were considered in the context of the currently available scientific evidence and were used in finalising the RARMP that formed the basis of the Regulator's decision to issue the licence. Advice received is summarised below.

Submission	Summary of issues raised	Comment
1	Agrees that the risk assessment identifies all plausible risk scenarios by which the proposed release could give rise to risks relating to the health and safety of people or the environment.	Noted.
	Agrees with the overall conclusion of the RARMP.	Noted.
	Advises that the Regulator should further consider risks associated with the potential for increased dormancy and increased weediness as a result of the fusion protein.	Risk scenario 2 discusses the potential for the fusion protein to increase dormancy or weediness of the GM canola, and preliminary data from the applicant suggest no changes in growth, germination rates, time to flowering or seed count per plant between non-GM canola and the GMOs. Text has been added to state that no reports were found in the literature to suggest production of the β-casein fusion protein could increase dormancy or weediness of the GMOs. In the unlikely event of increased dormancy or dispersal, the imposed control measures, including post-harvest monitoring for volunteers, would restrict GM seed dispersal and persistence.
	Advises that the Regulator should further consider controls in regards to cleaning of equipment and verification of the cleaning process.	The licence requires that equipment used in connection with the GMOs are cleaned after use and before use for any other purpose, which requires removal or destruction of the GMOs. The Regulator has issued many licences for dealings with GM crops, including other canola licences, and the condition requiring cleaning of equipment has been highly effective in preventing spread and persistence of GMOs. It is considered unlikely that verification of the cleaning process would increase the effectiveness of the cleaning condition already imposed by the licence.
	Advises that the Regulator should seek more detail about the sensory testing, and whether further controls around the testing are required.	The licence prohibits the use of GM plant material as human food, except for as part of sensory testing. The licence also requires that sensory testing must be conducted under oversight by a Human Research Ethics Committee (HREC), which is required to review and approve the research proposals in accordance

[4] Prescribed expects, agencies and authorities include GTTAC, State and Territory Governments, Australian Government agencies and the Minister for the Environment.

Submission	Summary of issues raised	Comment
		with the National Statement on Ethical Conduct in Human Research and therefore the risks associated with the GM products considered and managed under that framework. Some additional text about this has been added in Chapter 3 of the RARMP.
	Advises that the Regulator should reconsider the distance of the trial sites from waterways.	The RARMP includes information that indicates that GM canola seeds are unlikely to remain viable after prolonged exposure to water during a flooding event or when waterlogged. Thus, it is considered that GM canola is unlikely to persist if spread in this manner. Text has been added in Chapter 1 to provide further context for this point. Numerous licences have been issued for limited and controlled releases of GM canola and other crops which required trial sites are located 50 m distance from waterways, and this has been an effective at controlling seed dispersal. Therefore, the 50 m distance from waterways (in combination with not planting in flood prone areas) is considered appropriate and is consistent with previous licences issued for limited and controlled release of canola.
	Advises that the Regulator should consider clarifying what is considered flood prone.	When selecting a site for a planting area, a licence holder must consider a number of factors related to potential for flooding. This includes, but is not limited to, site flooding history, distance from waterways and topography. A prescriptive definition of flood prone is not practical for a number of reasons, as is reflected in the range of definitions across different jurisdictions and for different purposes. Additionally, current definitions may not be appropriate for future conditions, including changes resulting from climate change. Guidance is now included as a note to Condition 29 of the licence, to provide clarity for the licence applicant about how 'flood prone' may be considered for a site.
2	No advice or comments on the RARMP.	Noted.
3	Notes that the RARMP should appropriately address biosecurity concerns.	The RARMP does not consider potential biosecurity concerns associated with the proposed dealings with the GM canola. Biosecurity matters are the remit of the Department of Agriculture, Forestry and Fisheries (DAFF) and are not in the remit of the Regulator. The RARMP does, however, address risks associated with weediness and pest species associated with field trials of the GMOs.
	Notes that individual landowner must obtain relevant town planning approvals for use of the land where required.	Noted. The RARMP only lists local government areas proposed for potential release of the GMOs. The licence applicant must seek approval from landowners prior to planting. However, issues related to land use approvals are outside the remit of the Regulator. The licence holder is required to inform the Regulator of the location of all sites planted under this licence and

Submission	Summary of issues raised	Comment
		these sites are listed on the OGTR website once notified.
4	Has no concerns regarding the licence application or the introduction of protein-coding genes in canola seed. Noted the total maximum planting area over the period of the trial.	Noted.
5	Concludes that limited and controlled release of the GMO poses negligible risks to the health and safety of people or the environment under the proposed controls and conditions limiting the scale, location and duration of the release, and measures to restrict the spread and persistence of the GMOs and their genetic material in the environment.	Noted.
6	Agrees with the suitability of the proposed options for control of pollen flow from trial sites while GMOs are flowering. Expresses a preference for use of insect-proof tents, as these would also limit bird interactions with GM seed.	The RARMP discusses suitability of all three options for pollen flow control. All options have been approved and considered effective means of restricting pollen flow in previous GM canola releases (e.g. DIR 164, DIR 188 and DIR 205). Risk scenario 2 discusses bird-mediated dispersal of canola seed. Canola seeds do not have specific adaptations such as burrs or hooks, which would facilitate dispersal following bird interaction. Further, dispersal via consumption and excretion of viable canola seed only occurs at very low levels.
	Notes that the risk scenarios described in the RARMP are appropriate and well measured. Supports the conclusion of the RARMP that the field trial poses negligible risk of harm to human health and safety and the environment.	Noted.
7	Notes marker genes are widely used in transgenesis. Their use has been assessed numerous times and risks found to be negligible. Notes the GMOs would not be used for commercial human food or animal feed, and that volunteer plants will be controlled.	Noted.
	Notes that the RARMP does not identify a significant risk to human health and safety or the environment.	Noted.
	Notes the potential allergenicity of β-casein proteins, and a lack of historical assessments relating to GM products containing β-casein. Notes that additional analyses will be required if the GMOs were to be grown for human consumption commercially.	Noted.

Submission	Summary of issues raised	Comment
	Notes similarity to licence application DIR-211, which also proposes to produce casein in a crop plant. Expresses concern regarding the safety of the GM product to human consumers, and notes a lack of information about such trials in this licence application.	This licence application proposes only field trials of GM canola, and does not permit use of the GMOs in commercial human food or animal feed. Products derived from the GMOs may be used in sensory testing only (as described in Risk scenario 1 and Section 3.1.2 of the RARMP). Permission for the GM canola and its products to be sold as food for human consumption requires a separate regulatory assessment and decision from Food Standards Australia New Zealand (FSANZ). FSANZ is also responsible for setting the requirements for GM food labelling in Australia.
	Notes that the GM canola lines have not been assessed elsewhere, and suggests that the risk assessment and proposed licence conditions appear stricter than expected for other GM canola varieties.	While approval of the GMOs by overseas regulatory bodies is considered as part of the risk context, risks associated with GM canola lines proposed for release as part of this application was assessed through structured and rigorous approach as set out in the Regulator's *Risk Analysis Framework*. Risk scenarios were postulated and characterised by considering scientific evidence and the likelihood of harm, and licence conditions were proposed to appropriately manage these risks.
	Expresses uncertainty regarding the ability to inspect planting areas and pollen traps for related species, noting that related *Brassica* species are morphologically similar, particularly at early developmental stages, and that plants could be densely planted. Also notes that accessing areas for inspection would damage plants in the planting area or pollen trap.	The licence requires that planting areas and associated areas must be inspected by people trained to recognise plants of related species (Condition 26). For each trial site, the licence holder must also notify the Regulator of how inspection activities will be managed and strategies used for detection and destruction of related species (Condition 47(a)x).
	Notes that examination of recent planting site history for related species presence would be a useful means of preventing related species volunteers at trial sites.	Noted. The proposed licence conditions do not prescribe a means by which the licence holder must limit growth of related species volunteers. However, the proposed conditions require that the licence holder effectively monitor for presence of related species, and that identified volunteers are destroyed before flowering or prevented from flowering (Condition 26).

Appendix B: Summary of submissions from the public on the consultation RARMP

The Regulator received 2 submissions from members of the public on the consultation RARMP. The issues raised in the submission are summarised in the table below. All issues that related to risks to the health and safety of people and the environment were considered in the context of currently available scientific evidence in finalising the RARMP that formed the basis of the Regulator's decision to issue the licence.

Submission	Summary of issues raised	Comment
1	Opposes genetic modification of canola.	Noted.
2	Notes no issues relating to the protection of human health or safety and the environment. Notes current availability of casein proteins in the environment and a lack of associated health concerns. Also notes effective management of risks associated in the proposed field trial by restriction of public access to the GMOs, and proposes that an allergen warning would be sufficient for future, broader releases.	Noted. Potential allergenicity related to the GMOs would be assessed if the applicant applied for commercial release of the GM canola in the future. Allergenicity related to food products derived from the GM canola would be assessed by FSANZ.
	Supports production of a plant-based source of casein protein, and highlights its value as a protein source for people who eat a plant-based diet. Also notes the potential for large-scale casein production which is cheaper and more environmentally-friendly compared to casein sourced from dairy.	Noted.