Australian Government
Department of Health
Office of the Gene Technology Regulator

Risk Assessment and Risk Management Plan for

DIR 129

Limited and controlled release of sugarcane genetically modified for herbicide tolerance

Applicant: Sugar Research Australia Ltd

October 2014

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Summary of the Risk Assessment and Risk Management Plan

for

Licence Application DIR 129

Decision

The Gene Technology Regulator (the Regulator) has decided to issue a licence for this application for a limited and controlled release of a genetically modified organism (GMO) into the environment. A Risk Assessment and Risk Management Plan (RARMP) for this application was prepared by the Regulator in accordance with requirements of 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 concludes that this field trial poses negligible risks 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

Application number	DIR 129
Applicant:	Sugar Research Australia Limited (SRA)
Project Title:	Limited and controlled release of sugarcane genetically modified for herbicide tolerance
Parent organism:	Sugarcane (*Saccharum spp.* Hybrid)
Introduced genes[1] and modified traits:	• one gene from a plant species (herbicide tolerance) and • three variants of a gene from a bacterium (herbicide tolerance)
Proposed release dates:	November 2015 – November 2021
Proposed locations:	Seven[2] sites in the local government areas of Bundaberg, Mackay, Burdekin, Moreton Bay and Cairns (Queensland)
Proposed release size:	30 hectares per season
Primary purpose:	To evaluate the field performance of GM herbicide tolerant sugarcane and to conduct breeding to develop commercially useful GM herbicide tolerant sugarcane clones.

Risk assessment

The risk assessment concludes that risks to the health and safety of people, or the environment, from the proposed release are negligible.

The risk assessment process considers how the genetic modification and 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 information in the application (including proposed limits and controls), relevant previous approvals and current scientific/technical knowledge, and advice received from a wide range of experts, agencies and authorities consulted on the RARMP. Both the short and long term potential harms are considered.

[1] The identities of these genes have been declared Confidential Commercial Information (CCI) under section 185 of the Act.

[2] The applicant requested an additional site in the Bundaberg LGA during consultation.

Credible pathways to potential harm that were considered included: unintended exposure to the GM plant material; increased spread and persistence of the GM sugarcane relative to unmodified plants; and transfer of the introduced genetic material to non GM sugarcane, or other sexually compatible plants. Potential harms associated with these pathways included toxicity to people and other animals, allergic reactions in people and environmental harms associated with weediness.

The principal reasons for the conclusion of negligible risks are that the introduced genetic modifications are unlikely to cause harm to human health or safety or to the environment, the introduced genes are similar to those already existing in the environment, and furthermore, the imposed limits and controls effectively contain the GMOs and their genetic material and minimise exposure.

Risk management plan

The risk management plan concludes that risks posed by the proposed dealings can be managed so as to protect people and the environment by imposing conditions on the release.

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 evaluates and treats identified risks, evaluates controls and limits proposed by the applicant, and considers general risk management measures. The risk management plan is given effect through licence conditions.

As the level of risk is assessed as negligible, specific risk treatment is not required. However, as this is a limited and controlled release, the licence includes limits on the size, locations and duration of the release, as well as controls including containment provisions at the trial sites; prohibiting the use of GM plant materials in human food or animal feed; destroying GM plant materials not required for further studies; transporting GM plant materials in accordance with the Regulator's guidelines; and conducting post-harvest monitoring at the trial sites to ensure all GMOs are destroyed.

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Abbreviations

APVMA	Australian Pesticides and Veterinary Medicines Authority
CaMV	Cauliflower mosaic virus
CCI	Confidential Commercial Information as declared under section 185 of the *Gene Technology Act 2000*
DIR	Dealings involving Intentional Release
FSANZ	Food Standards Australia New Zealand
GM	Genetically modified
GMO	Genetically modified organism
ha	Hectare
HT	Herbicide tolerance
LGA	Local government area
m	Metres
NLRD	Notifiable low risk dealings
OGTR	Office of the Gene Technology Regulator
PC2	Physical Containment level 2
PCR	Polymerase chain reaction
RARMP	Risk Assessment and Risk Management Plan
Regulations	Gene Technology Regulations 2001
Regulator	Gene Technology Regulator
SRA	Sugar Research Australia Limited
TGA	Therapeutic Goods Administration
the Act	The *Gene Technology Act 2000*

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 in conjunction with the Gene Technology Regulations 2001 (the Regulations), an inter-governmental agreement and corresponding legislation that is being enacted in each State and Territory, 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 genetically modified organisms (GMOs).

3. This chapter describes the parameters within which potential risks to the health and safety of people or the environment posed by the proposed release are assessed. The risk assessment context is established within the regulatory framework and considers application-specific parameters (Figure 1).

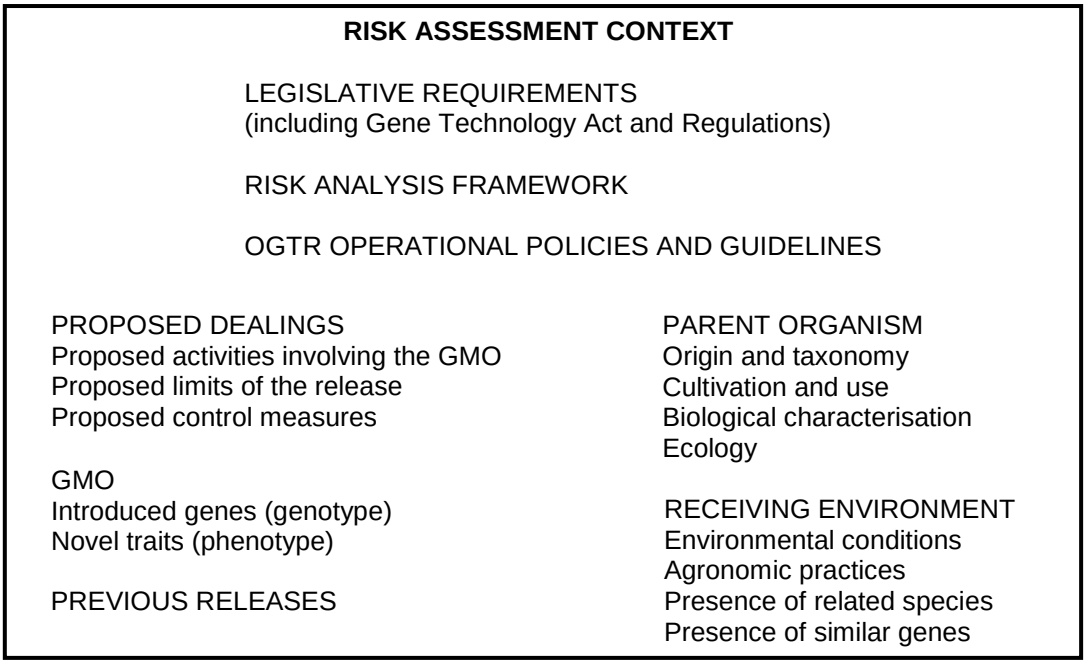

Figure 1 Summary of parameters used to establish the risk assessment context

Section 2 Regulatory framework

4. Sections 50, 50A and 51 of the Act outline the matters which the Gene Technology Regulator (the Regulator) must take into account, and the consultation required when preparing the Risk Assessment and Risk Management Plans (RARMPs) that inform the decisions on licence applications. In addition, the Regulations outline further matters the Regulator must consider when preparing a RARMP. In accordance with section 50A of the Act, this application is considered to be a limited and controlled release application, as its principal purpose is to enable the applicant to conduct experiments and the applicant has proposed limits on the size, locations and duration of the release, as well as controls to restrict the spread and persistence of the GMOs and their genetic material in the environment. Therefore, the Regulator was not required to consult with prescribed experts, agencies and authorities before preparation of the Risk Assessment and Risk Management Plan (RARMP; see section 50 of the Act).

5. Section 52 of the Act requires the Regulator to seek comment on the RARMP from the States and Territories, the Gene Technology Technical Advisory Committee, Commonwealth authorities or agencies prescribed in the Regulations, the Minister for the Environment, relevant local council(s), and 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 considerations are summarised in Appendix B.

6. The Risk Analysis Framework (OGTR 2013) explains the Regulator's approach to the preparation of RARMPs in accordance with the legislative requirements. Additionally, there are a number of operational policies and guidelines developed by the Office of the Gene Technology Regulator (OGTR) that are relevant to DIR licences. These documents are available from the OGTR website.

7. Any dealings conducted under a licence issued by the Regulator may also be subject to regulation by other Australian government agencies that regulate GMOs or GM products, including Food Standards Australia New Zealand (FSANZ), Australian Pesticides and Veterinary Medicines Authority (APVMA), Therapeutic Goods Administration (TGA), National Industrial Chemicals Notification and Assessment Scheme and Department of Agriculture. These dealings may also be subject to the operation of State legislation declaring areas to be GM, GM free, or both, for marketing purposes.

Section 3 The proposed dealings

8. Sugar Research Australia Limited (SRA) proposes to release up to 1800 lines[3] of genetically modified (GM) sugarcane into the environment under limited and controlled conditions.

9. The purpose of the trial is to evaluate the GM sugarcane lines grown under field conditions for key changes to agronomic characteristics such as sugar and cane yield, and herbicide tolerance.

10. The proposed dealing will continue the work done in the last five years under licence DIR 096 with an aim of selecting promising GM plants and evaluating their suitability for commercial production. The dealings involved in the proposed intentional release include:

- conducting experiments with the GMOs
- breeding the GMOs
- propagating the GMOs
- growing or culturing the GMOs
- transporting the GMOs
- disposing of the GMOs

and the possession, supply or use of the GMOs for the purposes of, or in the course of, any of the above. These dealings are detailed further below.

3.1 The proposed limits of the dealings (size, locations, duration and people)

11. The applicant proposes to grow GM sugarcane plants between November 2015 and November 2021.

12. The GMOs are proposed to be planted at seven sites[4] at SRA Woodford, SRA Southern and ISIS Central Sugar Mill Co. Ltd in Bundaberg, SRA Central located in Mackay, SRA Burdekin and SRA Durre in Burdekin (two sites) and SRA Meringa located south of Cairns (Table 1). The facility

[3]The term 'line' is used to denote plants derived from a single plant containing a specific genetic modification resulting from a single transformation event.
[4] The applicant has proposed an additional site in the Bundaberg LGA during consultation.

at SRA Meringa would only be used for flower production, crossing of commercial cultivars into GM sugarcane, seed collection, germination and storage, with GM sugarcane only grown in pots and no field planting.

Table 1 Proposed localities for GM sugarcane field trials

Trial sites	Bundaberg	Bundaberg	Mackay	Burdekin 1	Burdekin 2	Meringa	Woodford
LGA	Bundaberg Regional Council	Bundaberg Regional Council	Mackay Regional Council	Burdekin Shire Council	Burdekin Shire Council	Cairns City Council	Moreton Bay Regional Council
Geographical location	SRA Southern Ashfield Rd Bundaberg	ISIS Central Sugar Mill Co. Ltd Private Mail Bag 1 Childers, Qld 4660	SRA Central Peaks Downs Highway Te Kowai	SRA Burdekin Bruce Highway Brandon	SRA Durre farm, 35 Sayers Rd Barratta	SRA Meringa Bruce Highway Gordonvale	SRA Woodford, 90 Old Cove Rd, Woodford
Nursery area (m^2)	200	NA	200	200	NA	200	200
Planting area (ha)	5	5	5	5	5	NA	5

13. The collective area of the field trials (over 7 sites) would be up to 30 hectares (ha) per growing season. Up to 1000 m^2 of nursery area will also be used for growing potted GM sugarcane plants.

14. The applicant is proposing that only trained and authorised staff would be permitted to deal with the GM sugarcane. Any other visitors to the sites would be accompanied by an authorised SRA representative and would not deal with the GMOs.

15. The applicant proposes to use the hot water treatment facilities at Burdekin, Mackay and Bundaberg (see para 53) as a prophylactic measure against disease development to treat sugarcane stalks before planting at the nursery.

3.2 The proposed controls to restrict the spread and persistence of the GMOs and their genetic material in the environment

16. The applicant has proposed a number of controls to restrict the spread and persistence of the GM sugarcane lines and the introduced genetic material in the environment, including:

- surrounding the field trial sites by one guard row of non-GM sugarcane outside each planting of GM cane
- an isolation zone of 6 m separating the guard row from non-GM sugarcane
- separating GM sugarcane from commercial sugarcane by at least 10 m
- locating the trial sites at least 50 m away from natural waterways
- separating GM sugarcane material from non-GM material when propagating seedlings or setts, and clearly identifying GM material
- separating GM from non-GM sugarcane in crossing facilities by at least 1 m (glasshouses, pot holding areas, photoperiod facility and a crossing shed)
- monitoring GM sugarcane in photoperiod facilities for spikelet opening, and enclosing inflorescences in pollen lanterns prior to spikelet opening

- harvesting and processing GM sugarcane from the trial separately from any other sugarcane
- monitoring the hot water treatment facility regularly during operations with GM sugarcane, and cleaning it after use including filtering the effluent water
- analysing GM plant material at the trial sites, in certified PC2 laboratories or other approved facilities
- destroying all plant material not required for experimentation or propagation
- following cleaning of sites, monitoring for and destroying any GM sugarcane that may grow for at least 12 months and until the site is free of volunteers for a continuous 6 month period
- transporting GM plant material in accordance with Regulator's *Guidelines for the Transport, Storage and Disposal of GMOs (2011)* and specific licence conditions
- not allowing the GM plant material or products to be used for human food or animal feed.

17. Figure 2 and Figure 3 show the two proposed planting layouts including some of these controls. These controls, and the limits outlined above, have been taken into account in establishing the risk assessment context (this Chapter), and their suitability for containing the proposed release is evaluated in Chapter 3, Section 3.1.1.

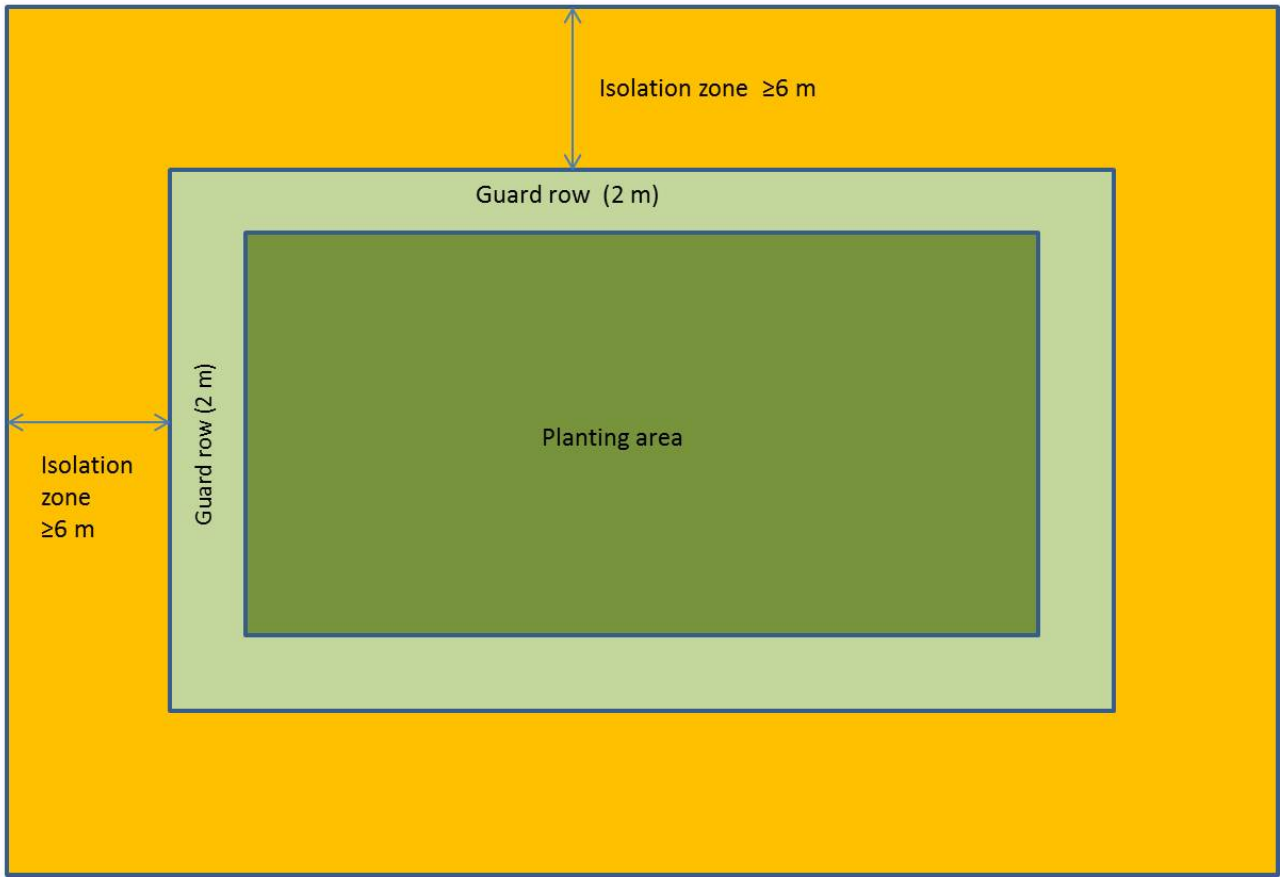

Figure 2 Proposed trial layout 1, including some of the controls (not drawn to scale)

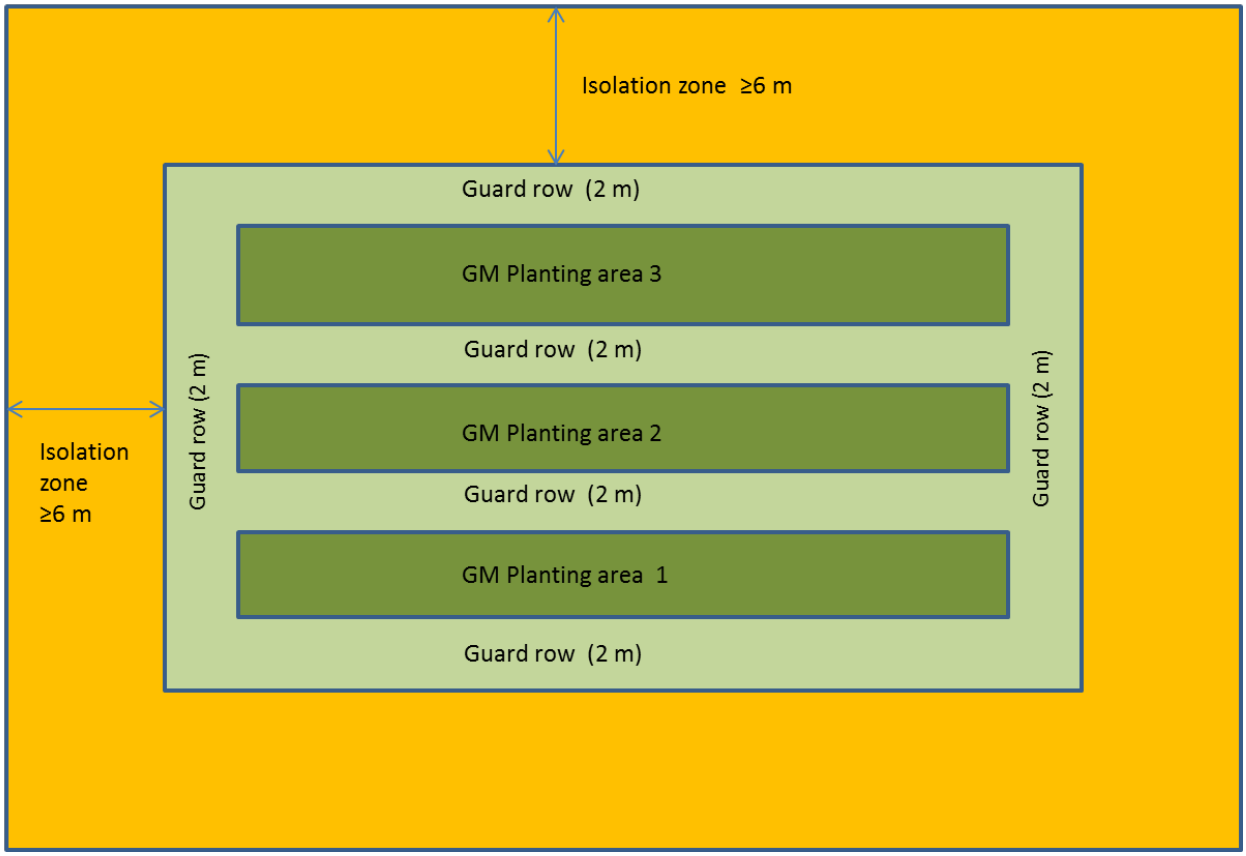

Figure 3 Proposed trial layout 2, including some of the controls (not drawn to scale)

Section 4 The parent organisms

18. The parent organism is cultivated sugarcane, *Saccharum spp.* which is an interspecific hybrid of *S. spontaneum* and *S. officinarum*. Sugarcane is exotic to Australia and is commercially cultivated on the east coast of Australia from northern New South Wales (NSW) to far north Queensland (Qld).

19. Approximately 80% of the chromosomes in commercial cultivars are derived from *S. officinarum*, 10% from *S. spontaneum* with the remainder being hybrid chromosomes from the two species (D'Hont et al. 1996; Piperidis et al. 2000). Repeated back-crossing of initial hybrids to a female *S. officinarum* parent produced the current commercial cultivars with higher sugar accumulation, cane yield and many desirable agronomic and disease-resistance traits.

20. Detailed information about the parent organism is contained in the reference document *The Biology of the* Saccharum *spp. (Sugarcane)(OGTR 2011)*, which was produced to inform the risk assessment process for licence applications involving GM sugarcane plants. This document is available from the OGTR website.

Section 5 The GMOs, nature and effect of the genetic modification

5.1 Introduction to the GMOs

21. The applicant proposes to release up to 1800 lines of GM sugarcane. The lines were produced using biolistics mediated plant transformation. Information about this transformation method can be found in the risk assessment reference document *Methods of plant genetic modification* available from the Risk Assessment References page on the OGTR website.

22. The purpose of these modifications is to introduce herbicide tolerance into sugarcane.

5.1.1 Genes expected to confer herbicide tolerance and the encoded proteins

23. Two herbicide tolerance (HT) genes are included in each GM sugarcane line. HT1, HT3 and HT4 are variants of a gene from a common soil bacterium, and HT2 is a gene from a common plant species. The identity of the introduced genes and the source organisms have been declared CCI and were made available to the prescribed experts and agencies that were consulted on the RARMP for this application.

24. Short regulatory elements (promoters, enhancers, introns and terminators) are used to control expression of the introduced genes. Regulatory elements used in GM sugarcane come from maize (*Zea mays*), Cauliflower mosaic virus, *Agrobacterium tumefaciens* and potato (*Solanum tuberosum*) (Table 2).

Table 2 Promoters and other introduced genetic elements used in the GM sugarcane

Name of genetic element	Function	Source gene	Source organism
Ubi1	Promoter	*Polyubiquitin gene*	*Zea mays*
H2B	Promoter	*Histone 2B*	*Zea mays*
H2B-E	Promoter	*Synthetic promoter comprised of Ubi1, H2B and CaMV 35S*	*Zea mays* and *Cauliflower mosaic virus*
CaMV 35S	*Enhancer*	*CaMV 35S gene*	*Cauliflower mosaic virus*
Ubi1	*Intron*	*Polyubiquitin gene*	*Zea mays*
Nos	*Terminator*	*Nopaline synthase gene*	*Agrobacterium tumefaciens*
PinII	*Terminator*	*Proteinase inhibitor II gene*	*Solanum tuberosum*

25. Promoters are DNA sequences that are required in order to allow RNA polymerase to bind and initiate correct transcription. The promoters proposed to be used in the GM sugarcane lines are listed in Table 2. The *Ubiquitin1 (Ubi1)* promoter was obtained from maize; it is a constitutive promoter which has been used in plant genetic modification (Christensen et al. 1992). The *H2B-E* promoter is a synthetic promoter; it is an enhanced *H2B* promoter (originally from maize) and contains three copies of the *CaMV 35S* enhancer at the 5' of the promoter and the maize *Ubi1* intron 3' of the promoter.

26. Also required for gene expression in plants are messenger ribonucleic acid (mRNA) terminators, including a poly-adenylation signal. The mRNA terminators proposed to be used in the GM sugarcane lines were derived from the *nopaline synthase (nos)* gene from *A. tumefaciens* and *proteinase inhibitor II (pinII)* gene from potato. The *nos* terminator has been used in a wide variety of constructs for plant genetic modification (Reiting et al. 2007). Although *CaMV* and *A. tumefaciens* are plant pathogens, the regulatory sequences comprise only a small part of its total genome, and are not capable of causing disease.

5.2 Toxicity/allergenicity associated with the introduced genes, their encoded proteins and associated products

27. The genes conferring herbicide tolerance were obtained from a common plant species and a bacterium, and homologues of their encoded proteins occur naturally in a range of organisms, including plants widely consumed by people and animals. On this basis, humans and other organisms have a long history of exposure to these genes, or highly homologous variants, and their

expressed proteins. Information about the identity of the introduced genes has been declared CCI and was made available to the prescribed experts and agencies.

28. Bioinformatic analyses on the HT1 protein sequence did not identify any similarities to known toxins or allergens. The HT1 protein has also been studied for properties associated with toxicity and allergenicity. Based on these studies it was concluded that there is no evidence to indicate that the protein encoded by the HT1 gene is toxic and it has a low likelihood to be allergenic.

29. A protein highly similar to HT1 has also been extensively assessed for allergenicity and toxicity. The assessment indicates that the protein will not have adverse effects in humans. HT3 and HT4 are variants of HT1, and perform the same function in the GM sugarcane lines. They are not expected to differ in respect to toxicity or allergenicity.

30. Potential toxicity of end products formed as a result of the introduction of HT1 has also been assessed. Based on data from toxicity studies, it was concluded that these end products were of very low acute toxicity and were not genotoxic (ie capable of causing damage to DNA), and that food derived from a specific GM crop in which this gene is expressed as safe and wholesome as food derived from the corresponding non-GM crop.

31. The HT2 gene was isolated from a common food crop plant safely consumed by humans for centuries. No new by-products are anticipated to be produced as a result of the genetic modification in the GM sugarcane.

32. Bioinformatic analyses conducted with the HT2 protein sequence did not identify any similarities to known toxins or allergens. Studies based on HT2 protein concluded that there is no evidence for the protein to be toxic, and that it has a low likelihood to be allergenic.

33. A comprehensive search of the scientific literature yielded no information to suggest that any of the encoded proteins are toxic or allergenic to people, or toxic to other organisms.

34. No studies on the toxicity or allergenicity of the GM sugarcane lines and their products have been undertaken by the applicant to date as the proposed trial is at an early stage. Such studies may have to be conducted if approval was sought for the GMOs or their products to be considered for human consumption in Australia.

5.3 Characterisation of the GMOs

5.3.1 Stability and molecular characterisation

35. The applicant states that in previous trials of some of the GM sugarcane lines[5], conducted under licence DIR 096, no loss of transgene functionality was found. Twenty vegetatively propagated plants were tested over two crop cycles for each of 130 GM lines. Further, two events were crossed with 22 commercial cultivars and other non-GM sugarcanes. Progenies from all these crosses segregated in a Mendelian fashion. Under the proposed trial, crossing experiments will be continued to test transgene stability in diverse genetic backgrounds.

36. It is established that genetic transformation of plants results in deoxyribonucleic acid (DNA) integration into plant nuclear DNA. Studies in sugarcane and other species show that the introduced genes and traits remain stable across generations and transmit through progenies (Butterfield et al. 2002; Perlak et al. 1990; Rae et al. 2005; Sebastian et al. 1989; Umbeck et al. 1989).

37. The insertion of genes into sugarcane via microprojectile bombardment has been shown to be stable (Bower et al. 1996; Hansom et al. 1999). However, it is possible that expression of genetic changes may alter during successive cycles of vegetative propagation through various epigenetic effects such as change in methylation patterns and gene silencing phenomena. It is also possible that

[5] The term 'line' is used to denote plants derived from a single plant containing a specific genetic modification resulting from a single transformation event.

some GM sugarcane plants are chimeric and vegetative reproduction may result in loss of the genetic change. The applicant anticipates that such trait loss would in a very small proportion of GM lines, if any, and screening with herbicides during tissue culture will eliminate these plants before the next generation.

5.3.2 Phenotypic characterisation

38. GM sugarcane plants would be subjected to the herbicide treatments at three stages during the process of selection before being released into the field, ensuring that they express the intended herbicide tolerance trait.

39. Vickers et al. (2005) showed that both the transformation and tissue culture process had a significant negative impact on the agronomic performance (both yield and commercial cane sugar content) of sugarcane. This is usually the case for most of the lines in the initial plant crop and the negative effects gradually disappear in the subsequent generations (Joyce et al. 2013). However, the applicant states that in the trials conducted under licence DIR 096 and in earlier studies (Joyce et al. 2013), GM lines identical to non-GM parental lines for growth and yield have been identified. This suggests that GM sugarcane without adverse genetic effects can be produced.

40. The applicant states that significant agronomic impacts from expression of the introduced herbicide tolerance genes are not anticipated. This expectation is supported by the observation of GM sugarcane lines grown under licence DIR 096. The proposed field trial is designed to determine whether the herbicide tolerance genes have any phenotypic effects other than imparting herbicide tolerance to the GM sugarcane plants.

Section 6 The receiving environment

41. The receiving environment includes: any relevant biotic/abiotic properties of the geographic regions where the release would occur; intended agricultural practices, including those that may be altered in relation to normal practices; other relevant GMOs already released; and any particularly vulnerable or susceptible entities that may be specifically affected by the proposed release (OGTR 2013).

42. The factors relevant to the growth, distribution and cultivation of commercial sugarcane can be found in *The Biology of the* Saccharum *spp. (Sugarcane)* (OGTR 2011).

43. The proposed dealings involve planting of GM sugarcane at the following seven sites including SRA Woodford (northwest of Brisbane), SRA Southern and ISIS Central Sugar Mill Co. Ltd located in Bundaberg, SRA Central located in Mackay, SRA Burdekin and SRA Durre (near Ayr) in Burdekin and SRA Meringa located south of Cairns (Figure 4).

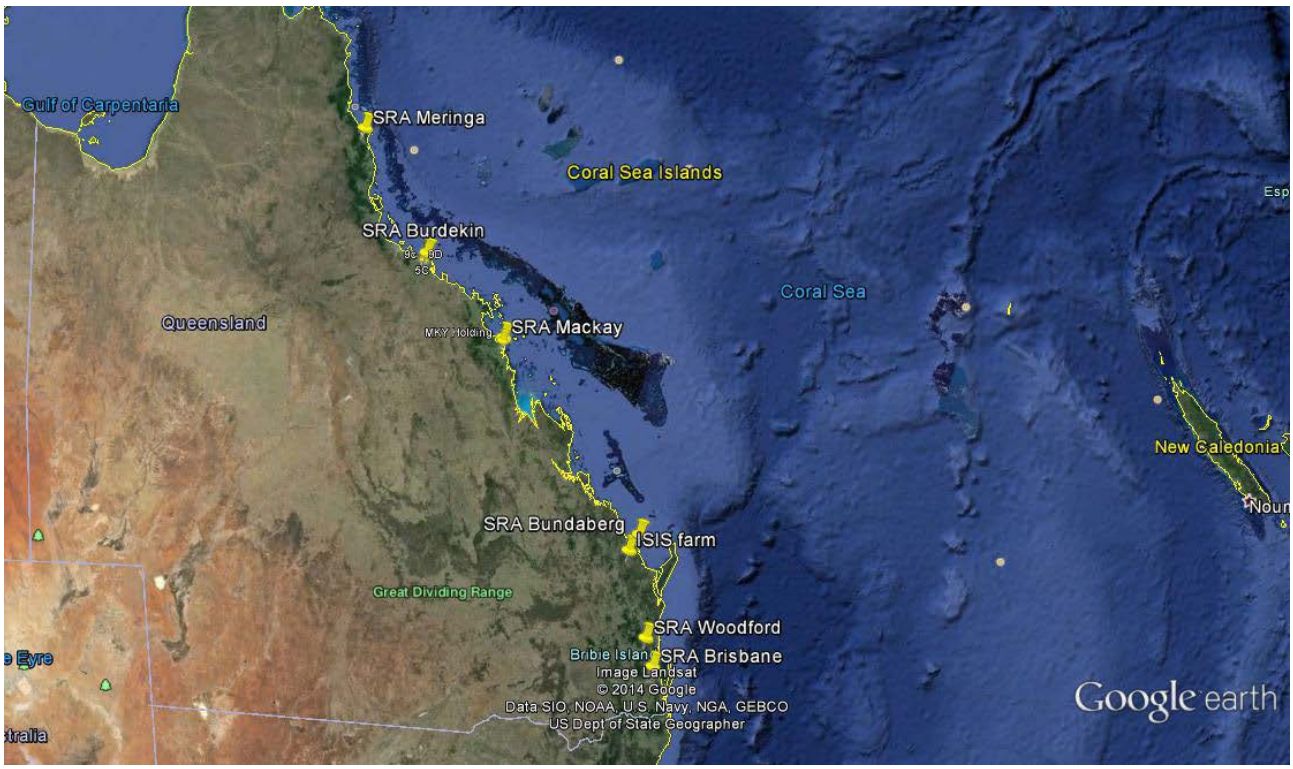

Figure 4 Geographic distribution of the trial sites (Supplied by SRA)

44. The SRA Meringa site will only be used for flower production, crossing, seed collection germination, and storage and the GM sugarcane material will be grown only in seedling trays or pots.

45. All the SRA sites are in agricultural areas on the outskirts of rural towns in Qld. Commercial sugarcane is grown in the immediate vicinity of each of the proposed sites on both commercial sugarcane farms and within the SRA stations, except at SRA Woodford which is not close to commercial sugarcane farms. The sites can only be accessed via private roads.

6.1 Relevant abiotic factors

46. The abiotic factors relevant to the growth and distribution of commercial sugarcane in Australia are discussed in *The Biology of the* Saccharum *spp. (sugarcane)* (OGTR 2011).

47. The release is proposed to take place in the Qld local government areas of Moreton, Bundaberg, Mackay, Burdekin and Cairns. The Cairns and Burdekin regions have a tropical climatic type, while the Mackay, Bundaberg and Moreton Bay regions have a sub-tropical climatic type (as defined by the Koppen Classification system used by the Australian Bureau of Meteorology).

48. With the exception of SRA Woodford, the proposed field trial sites are on flat arable land not subject to flooding (information supplied by the applicant). SRA Woodford is located on an undulating site which has no history of flooding. All of the GM sugarcane field trial sites would be located at least 50 m from the nearest waterway. The SRA Meringa photoperiod facility is within approximately 20 m of a small natural waterway, but the GM sugarcane at this location would be large plants in pots held within a large trolley. All other parts of the proposed crossing facility at SRA Meringa are more than 50 m from natural waterways. SRA Meringa has no history of flooding. To date, no incidents of dispersal of GM sugarcane plant material from field trials or crossing facilities as a result of cyclones or storms have been reported to the OGTR.

6.2 Relevant agricultural practices

49. The size, locations and duration of the proposed limited and controlled release of the GM sugarcane lines are outlined in Section 3.1 and Table 1 of this Chapter.

50. In Qld, commercial sugarcane is planted from autumn to spring and harvested after 12-18 months. The GM sugarcane is proposed to be grown in the field (at SRA Woodford, Southern, Central, Durre, Burdekin and ISIS Central Sugar Mill Co. Ltd in Bundaberg) and data collection and harvest would occur at the end of each growing season from one plant crop and two or more ratoons of the selected lines. At SRA Meringa, plants would only be grown in pots. The Meringa facility would only be used for flower induction, crossing, seed collection, seed germination and storage.

51. GM sugarcane plants proposed for release have been or will be produced in the Physical Containment 2 (PC2) laboratories in SRA Indooroopilly. In the majority of the cases, test plants would be transported to the SRA stations in the trial locations to be hardened on seedling benches for 8-10 weeks prior to planting or may be hardened in the PC2 glasshouses at SRA Indooroopilly and then transported to the trial sites. Field preparation and planting of GM plants will be similar to non-GM plants and as practised under licence DIR 096. Hot water treatment (see below) may also be used to treat the GM sugarcane before planting.

52. Tissue-cultured plants from selected GM events with herbicide tolerance are proposed to be planted in pots and further assessed for herbicide tolerance. Tolerant GM lines would be used for the next stage of development. True seedlings generated through crossing would be assessed on seedling benches for herbicide tolerance. The SRA breeding program entails progressive selection of GM sugarcane through multiple stages including nursery screening, progeny assessment trial, clonal assessment trial, final assessment trial, agronomy trial, pathology trial and plant breeder trial.

Hot Water Treatment

53. Hot water treatment is a best management practice in commercial sugarcane cropping. The applicant proposes to use hot water treatment facilities at Burdekin, Mackay and Bundaberg. In Burdekin, the applicant proposes to use the facility at Burdekin Productivity Services (BPS), the only provider for this service in that region. In Mackay and Bundaberg, SRA proposes to use their own facilities for this treatment prior to planting in the field. Secure access facilities containing tanks are used to treat the GM sugarcane. The procedure involves using cold water treatment for 48 hours, followed by hot water treatment for 3 hours. Barcoded cane stalks will be double-contained with shade cloth wrapping as the primary container and the enclosed tank as the secondary container. The tank will be filled well below the rim to avoid over flow of water. A filter is placed to the outlet as a precautionary measure. Tanks will be cleaned after each use. The facility will be monitored by SRA staff authorised under the DIR 129 licence.

Crossing and seed germination

54. Starting from 2016, the applicant proposes to perform crossing of GM lines with commercial varieties or advanced lines each year, with crossing only occurring at the Meringa facility. The total number of crosses per year would not exceed 50. The number of crosses would depend on the number of promising clones selected in final assessment trials and on their flowering propensity. Setts from these clones would be germinated in a non-PC2 growth cabinet (1-2 weeks) or glasshouse and then transferred to pots on trolleys maintained in the Photoperiod house at SRA Meringa. Before spikelets begin to open flowers would be cut and transferred to pollen-impermeable bags (lanterns) which are enclosed on all sides and the top, and the bottom would be secured at the base of the inflorescence. After seed setting, seed-bearing flowers would be collected in seed impermeable muslin bags and dried in a dehydrator for long-term storage. Seeds would be stored in the SRA Meringa Seed Bank in a locked box. Lanterns and muslin bags would be thoroughly cleaned to destroy any pollen or seed material.

55. All seeds produced from each cross would be germinated in non-PC2 growth cabinets or glasshouses. GM seeds would be germinated at a different time to wild-type seed and would occur in covered containers to prevent seed loss. After germination the cabinet would be thoroughly cleaned to destroy all viable material. Seedlings would be transported in double-sealed containers to a glasshouse where they will be grown until large enough for transfer into pots (8-10 weeks). Unused seedlings at the glasshouse or seedling bench stage would be destroyed. When no growth is observed, the material would be discarded into the GM waste disposal area. Seedlings grown in pots would be tested in PATs at trial sites after selection for herbicide tolerance.

56. The applicant proposes that all harvested plant materials would be stored in the holding area and destroyed as soon as practical. After the final harvest, each GM trial site would be burnt, deep-ploughed and then inspected for sugarcane volunteers. Any volunteers would be destroyed by herbicide treatment. All field sites would be inspected for at least 12 months and until volunteer-free for at least 6 months continuously.

6.3 Presence of related plants in the receiving environment

57. The SRA stations are located within commercial sugarcane growing regions. Commercial sugarcane is grown adjacent to some of the proposed field trial sites. The SRA stations are used for breeding of commercial cultivars and include field trials of non-GM sugarcane; these would be at least 6 m from the GM sugarcane trial sites. Within the crossing facility at SRA Meringa, the applicant has proposed to keep the GM and non-GM sugarcane plants separate; plants would be separated by a distance of at least 1 m, unless plants were used for crossing under a pollen lantern.

58. Sugarcane is known to cross with other species within the Saccharum genus, however of these species, only *S. spontaneum* and *S. officinarum* are known to occur as isolated populations in Australia (OGTR 2011). Other members of the genus are kept in various Australian germplasm collections, including a clone garden near the proposed trial site at SRA Meringa, where *S. officinarum, S. spontaneum* and *S. robustum* plants are maintained. These species are all sexually compatible with cultivated sugarcane. *S. robustum* is thought to be the ancestral species from which *S. officinarum* is derived (Brown et al. 2007; D'Hont et al. 1998) and it has been proposed that it should be classified as *S. officinarum* (Irvine 1999). Naturalised populations of *S. spontaneum* have been recorded at several locations in northern Qld, including in sugarcane growing areas along a significant part of the Mulgrave and Herbert rivers (Bonnett et al. 2007; Bonnett et al. 2010; Bonnett et al. 2008). Preliminary molecular analysis of this population suggested the plants have reproduced vegetatively (Bonnett et al. 2008).

59. Sugarcane has been reported to produce hybrids with a number of species of closely related genera in a group known as the *Saccharum* complex. However, these hybrids have usually occurred under controlled experimental conditions, often with *Saccharum*, in particular *S. officinarum*, as the female (Aitken et al. 2007; Nair et al. 2005; Wang et al. 2009). Genera for which hybridisation has been established are *Erianthus* and *Miscanthus*, but these exotic species do not occur in Qld (Bonnett et al. 2008). Possible hybridisations to the genera *Narenga, Imperata, Schlerostachya* and *Miscanthidium* have been reported, but these events have not been verified by molecular methods (Bonnett et al. 2008). Blady grass (*Imperata cylindrica*) is common throughout Qld coastal areas; however the applicant has stated that it has not been observed in the vicinity of the SRA stations proposed to be used for the trial.

60. Sugarcane has been reported to cross with a number of species not considered to be close relatives. A small number of hybrids between sugarcane and sorghum (*Sorghum bicolor*) have been generated from experiments in which large amounts of *S. officinarum* pollen were used to pollinate male sterile sorghum flowers (Nair 1999). Crosses of sorghum as the pollen donor with commercial *Saccharum* hybrids have been reported, but also under controlled conditions. The few hybrids produced by the above crosses lacked vigour and showed slow growth (Nair 1999). Hybrids between *Saccharum* and sorghum have not been observed under natural conditions (Bonnett et al. 2008). Similarly, following hand pollination of thousands of *S. officinarum* florets with maize (*Z.*

mays) pollen, only a single hybrid plant was produced (Bonnett et al. 2008). Wild Sorghum species occur as widespread weeds of cultivated areas in Qld. According to the applicant, maize and sorghum are not cultivated in close proximity to the proposed trial sites. Although hybridisation between *Saccharum* and *Bambusa* (bamboo) has been reported (Rao et al. 1967), subsequent analysis has suggested the hybridisation was not genuine (Grassl 1980).

6.4 Presence of similar genes and encoded proteins in the environment

61. The introduced gene sequences were originally isolated from naturally occurring organisms, which are already widespread and prevalent in the environment. HT1, HT3 and HT4 are modifications of the same gene derived from a common soil bacterium.

62. The HT2 gene was obtained from a common food crop plant and therefore humans routinely encounter this gene, its gene products, or homologues, through consumption of this plant.

63. Short regulatory sequences are derived from plants (including maize and potato), a soil bacterium (*A. tumefaciens*) and a plant virus (Cauliflower mosaic virus; CaMV). Although some of these sequences are derived from plant pathogens (*A. tumefaciens* and CaMV), the regulatory sequences comprise a small part of the pathogen's total genome, and are not known to be capable of causing disease in themselves.

Section 7 Relevant Australian and international approvals

7.1 Australian approvals

7.1.1 Approval by the Regulator

64. As indicated earlier, the proposed trial will be a continuation of dealings currently conducted under DIR 096 and will involve some of the same lines.

65. The Regulator has previously issued licences for the limited and controlled release of GM sugarcane with different introduced traits. Information on previous DIR licences can be found on the GMO record on the OGTR website. There have been no reports of adverse effects on human health or the environment resulting from any of these releases.

7.1.2 Approval by other government agencies

66. The Regulator is responsible for assessing risks to the health and safety of people and the environment as a result of gene technology. Other government regulatory requirements may also have to be met in respect of release of GMOs, including those of the Department of Agriculture - Biosecurity, Food Standards Australia New Zealand (FSANZ), and Australian Pesticides and Veterinary Medicines Authority (APVMA). This is discussed further in Chapter 3.

67. APVMA has regulatory responsibility for the use of agricultural chemicals, including herbicides and insecticidal products, in Australia. The GM sugarcane has been modified to be herbicide tolerant and the applicant intends to apply herbicides during the trial. The application of herbicides is subject to regulation by the APVMA.

68. FSANZ is responsible for human food safety assessment and food labelling, including GM food. The applicant does not intend to use materials from the GM sugarcane lines in human food; accordingly no application has been submitted to FSANZ. FSANZ approval would need to be obtained before materials from these GM sugarcane lines could be sold as food or food ingredients.

7.2 International approvals of GM sugarcane

69. None of the herbicide tolerant GM sugarcane lines have been released overseas.

Chapter 2 Risk assessment

Section 1 Introduction

70. 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 5). Risks are identified within the context established for the risk assessment (see Chapter 1), taking into account current scientific and technical knowledge. A consideration of uncertainty, in particular knowledge gaps, occurs throughout the risk assessment process.

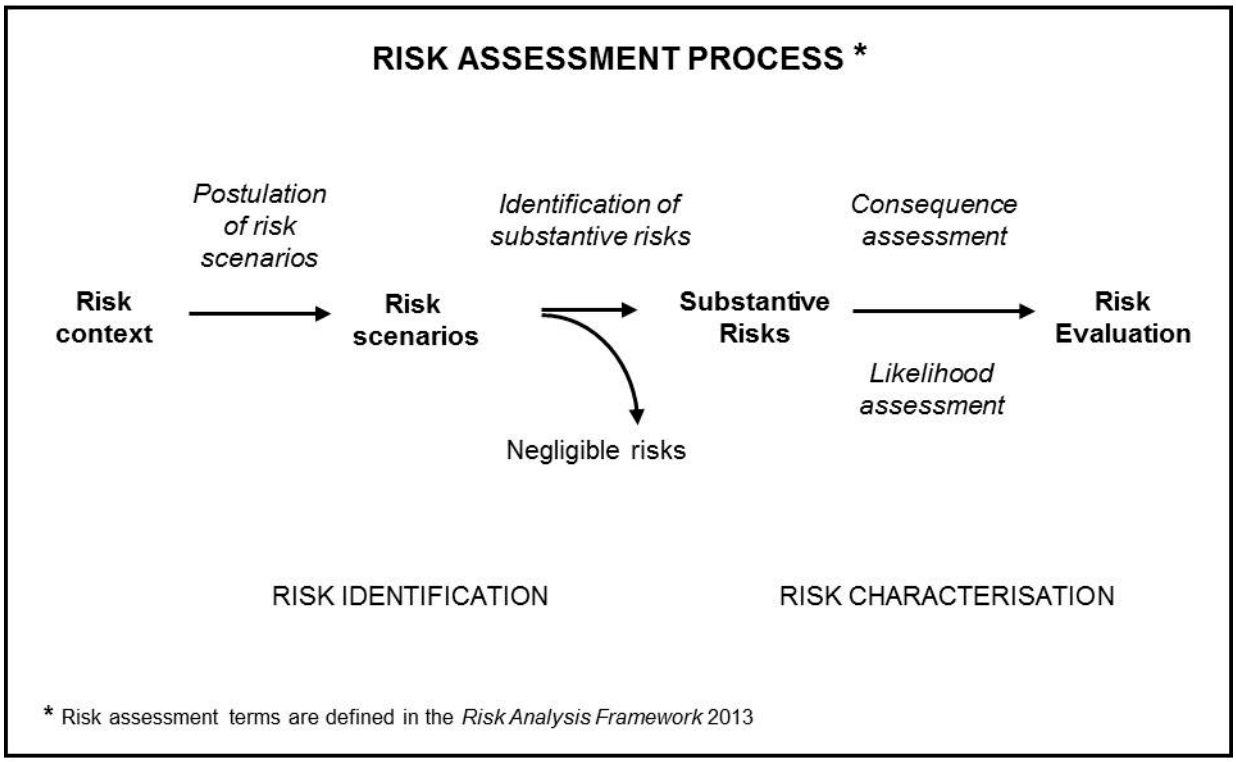

Figure 5 The risk assessment process

71. Initially, risk identification considers a wide range of circumstances whereby the GMO, or the introduced genetic material, could come into contact with people or the environment. Consideration of these circumstances leads to postulating plausible causal or exposure pathways that may give rise to harm for people or the environment from dealings with a GMO (risk scenarios) in the short and long term.

72. Postulated risk scenarios are screened to identify substantive risks that warrant detailed characterisation. A substantive risk is only identified for further assessment when a risk scenario is considered to have some reasonable chance of causing harm. Pathways that do not lead to harm, or could not plausibly occur, do not advance in the risk assessment process.

73. A number of risk identification techniques are used by the Regulator and staff of the OGTR, including checklists, brainstorming, reported international experience and consultation (OGTR 2013). 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. 2013). In addition, risk scenarios postulated in previous RARMPs prepared for licence applications of the same and similar GMOs are also considered.

74. Substantive risks (*i.e.* those identified for further assessment) are characterised in terms of the potential seriousness of harm (Consequence assessment) and the likelihood of harm (Likelihood assessment). The level of risk is then estimated from a combination of the Consequence and Likelihood assessments. Risk evaluation then combines the Consequence and Likelihood assessments to determine level of risk and whether risk treatment measures are required. The potential for interactions between risks is also considered.

Section 2 Risk Identification

75. Postulated risk scenarios are comprised of three components (Figure 6):

i. The source of potential harm (risk source)

ii. A plausible causal linkage to potential harm (causal pathway)

iii. Potential harm to an object of value, people or the environment.

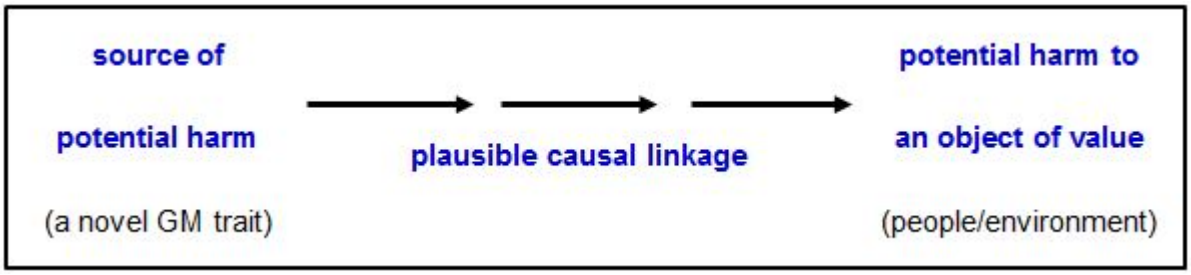

Figure 6 Risk scenario

76. In addition, the following factors are taken into account when postulating relevant risk scenarios:

- the proposed dealings, which may be to conduct experiments, develop, produce, breed, propagate, grow, import, transport or dispose of the GMOs, use the GMOs in the course of manufacture of a thing that is not the GMO, and the possession, supply and use of the GMOs in the course of any of these dealings
- the proposed limits including the extent and scale of the proposed dealings
- the proposed controls to limit the spread and persistence of the GMO
- characteristics of the parent organism(s).

77. Additional information relevant to the risk assessment has been declared CCI by the Regulator. The CCI was made available to the prescribed experts and agencies that were consulted on the RARMP for this application.

2.1 Risk source

78. The source 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.

79. As discussed in Chapter 1, the proposed GM sugarcane will be modified by the introduction herbicide tolerance genes. These introduced genes are considered further as potential sources of risk.

80. All of the introduced genes include regulatory sequences, which are derived from potato (*Solanum tuberosum*), a plant virus (Cauliflower mosaic virus), and a common soil bacterium (*Agrobacterium tumefaciens*). There is no evidence that regulatory sequences themselves have toxic or allergenic effects (EPA 1996); such effects for these sequences will not be further assessed for this application. Although the viral sequence is derived from a plant pathogen, it only constitutes a

small part of the genome and cannot itself cause disease. However, regulatory sequences, especially the promoters, control the levels of gene expression and hence the levels of the derived proteins in the GM plants. The effects of these protein levels, in particular, the toxicity and allergenicity of these plants (or at least materials derived from them), will be discussed below.

2.2 Causal pathway

81. 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 effects of the introduced gene(s) and gene product(s) on the properties of the organism
- 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 (invasiveness) of the GM plant, including
    - establishment
    - reproduction
    - dispersal by natural means and by people
- tolerance to abiotic conditions (eg climate, soil and rainfall patterns)
- tolerance to biotic stressors (eg pest, pathogens and weeds)
- tolerance to cultivation management practices
- gene transfer to sexually compatible organism
- gene transfer by horizontal gene transfer (HGT)
- unauthorised activities.

82. Although all of these factors are taken into account, some have been considered in previous RARMPs or are not expected to give rise to substantive risks.

83. The potential for horizontal gene transfer (HGT) and any possible adverse outcomes has been reviewed in the literature (Keese 2008) as well as assessed in many previous RARMPs. These RARMPs are available from the GMO Record on the OGTR website **Error! Hyperlink reference not valid.**or by contacting the OGTR. No risk greater than negligible was identified due to the rarity of these events and because the wild-type gene sequences are already present in the environment and available for transfer via demonstrated natural mechanisms. Therefore, HGT will not be assessed further.

84. The potential for unauthorised activities to lead to an adverse outcome has been considered in previous RARMPs. 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 a 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 *Herbicide resistance management*

85. Herbicide resistance comes under the regulatory oversight of the APVMA. The APVMA has primary regulatory responsibility for agricultural chemicals in Australia. The APVMA operates the national system that evaluates, registers and regulates agricultural and veterinary chemical products.

Any changes to a product that is already on the market must also be referred to the APVMA. The development of resistance to a herbicide would have implications for the choice of herbicide(s) available for weed control operations in agriculture and elsewhere. The APVMA assesses all herbicides used in Australia and sets their conditions of use.

86. In the event of commercial release, the applicant proposes to develop a herbicide resistance management plan as required by APVMA for herbicide registration purpose.

87. Herbicide resistance is primarily a risk to agricultural production, rather than a risk to the health of people or the environment. Therefore, this issue will not be discussed further.

2.4 Potential harm

88. Potential harms from GM plants include:

- harm to the health of people or desirable organisms, including toxicity/allergenicity
- reduced establishment of desirable plants, including having an advantage in comparison to related plants
- reduced yield of desirable vegetation
- reduced products or services from the land use
- restricted movement of people, animals, vehicles, machinery and/or water
- reduced quality of the biotic environment (eg providing food or shelter for pests or pathogens) or abiotic environment (eg negative effects on fire regimes, nutrient levels, soil salinity, soil stability or soil water table)
- reduced biodiversity through harm to other organisms or ecosystems.

89. These harms are based on those used to assess risk from weeds (Standards Australia New Zealand & CRC for Australian Weed Management 2006). Judgements of what is considered harm depend on the management objectives of the land where the GM plant is expected to spread to and persist. A plant species may have different weed risk potential in different land uses such as dryland cropping or nature conservation.

2.5 Postulated risk scenarios

90. Six risk scenarios were postulated and screened to identify substantive risk. These scenarios are summarised in Table 3 and more detail of these scenarios is provided later in this Section. Postulation of risk scenarios considers impacts of the GM sugarcane or its products on people undertaking the dealings, as well as impacts on people and the environment if the GM plants or genetic material were to spread and/or persist.

91. In the context of the activities proposed by the applicant and considering both the short and long term, none of the six risk scenarios gave rise to any substantive risks that could be greater than negligible.

Table 3 Summary of risk scenarios from dealings with GM sugarcane genetically modified for herbicide tolerance

Risk scenario	Risk source	Causal pathway	Potential harm	Substantive risk?	Reason
1	Introduced herbicide tolerance genes	Growing GM plants at the sites ⬇ Expression of genes in GM plants ⬇ Exposure of people who specifically deal with the GM plant material or other organisms that come into contact with the GM plant material in the trial sites	Allergic reactions in people or toxicity in people and other organisms	No	• The limited scale, short duration and other proposed limits and controls minimise exposure of people and other organisms to the GM plant material. • Plant material from the GMOs would not be used for human food or animal feed. • Introduced proteins occur naturally in the environment and are not known to be toxic or allergenic to people or toxic to other organisms (the proteins have a history of safe use).
2	Introduced herbicide tolerance genes	Dispersal of GM seed outside trial limits ⬇ Growth of GM plants ⬇ Expression of genes in GM plants ⬇ Spread and persistence of populations of GM plants outside trial limits ⬇ Exposure of people or other organisms to GM plant material	Allergic reactions in people or toxicity in people and other organisms, and reduced establishment and yield of desirable plants, reduced biodiversity	No	• The limited scale, short duration and other proposed limits and controls minimise the likelihood that GM plant material would leave a trial site. • Many environmental factors are expected to limit the spread and persistence of sugarcane in the areas proposed for release. • Risk scenario 1 considered that there is no substantive risk of toxicity or allergenicity from the GM sugarcane.
3	Introduced herbicide tolerance genes	Dispersal of GM pollen outside trial limits ⬇ Vertical transfer of introduced genes to other sexually compatible plants ⬇ Expression of genes in plants ⬇ Exposure of people or other organisms to GM plant material	Allergic reactions in people or toxicity in people and other organisms	No	• Pollen viability is very low under natural conditions. • Pollen lanterns would reduce pollen dispersal in glasshouses and crossing facilities. • The limited scale, short duration and other proposed limits and controls minimise the likelihood that GM plant material would leave a trial site. • Introduced proteins occur naturally in the environment and are not known to be toxic or allergenic to people or toxic to other organisms (the proteins have a history of safe use).

Risk scenario	Risk source	Causal pathway	Potential harm	Substantive risk?	Reason
4	Introduced herbicide tolerance genes	Dispersal of GM pollen outside trial limits ⬇ Vertical transfer of introduced genes to other sexually compatible plants ⬇ Expression of genes in plants ⬇ Spread and persistence of populations of GM plants outside a trial site	Reduced establishment and yield of desirable plants, reduced biodiversity	No	• There is limited sexual compatibility with relatives of sugarcane. • The limited scale, short duration and other proposed limits and controls minimise the likelihood that GM plant material would leave a trial site.
5	Introduced herbicide tolerance genes	Dispersal of GM pollen within a site ⬇ Hybridisation of GM plants of this trial with GM plants (including volunteers) of another trial ⬇ Expression of genes in stacked GM plants ⬇ Exposure of people or other organisms to GM plant material	Allergic reactions in people or toxicity in people and other organisms	No	• The limited scale, short duration and other proposed limits and controls minimise exposure of people and other organisms to the GM plant material. • Most introduced proteins occur naturally in the environment and are not known to be toxic or allergenic to people or toxic to other organisms (the proteins have a history of safe use). • The stacking of genes from different GM plants is unlikely to increase the toxicity or allergenicity of plants.
6	Introduced herbicide tolerance genes	Dispersal of GM pollen within a site ⬇ Hybridisation of GM plants of this trial with GM plants (including volunteers) of another trial ⬇ Dispersal of plants or viable plant material containing stacked genes outside a site ⬇ Expression of genes in stacked GM plants ⬇ Spread and persistence of populations of GM plants outside a trial site	Reduced establishment and yield of desirable plants, reduced biodiversity	No	• The stacking of the herbicide tolerance with genes from different GM plants is unlikely to increase the weediness of plants, especially if herbicide is applied. • The limited scale, short duration and other proposed limits and controls minimise the likelihood that GM plant material would leave a trial site.

2.5.1 Risk scenario 1

Risk source	*Causal pathway*	*Potential harm*
Introduced herbicide tolerance genes	Growing GM plants at the sites ⬇ Expression of genes in GM plants ⬇ Exposure of people who specifically deal with the GM plant material or other organisms that come into contact with the GM plant material in the trial sites	Allergic reactions in people or toxicity in people and other organisms

Risk source

92. The source of potential harm for this postulated risk scenario is the introduced herbicide tolerance genes.

Causal pathway

93. The herbicide tolerance genes are expressed in the plant tissues. People who are involved in the breeding, cultivating, harvesting, transporting and processing of the GM sugarcane may be exposed to their products through contact (including inhalation of pollen). This would be expected to mainly occur in the trial sites, but could also occur anywhere the GM plant material is transported or used for experimental analysis. Organisms that may be present in the trial sites, including rodents and invertebrates, may be exposed to the GM plant material.

94. The proposed limits and controls of the trial would minimise the likelihood that people or other organisms would be exposed to GM plant material. Although people may directly handle the GM plant material, it is not to be used as human food. Further, as the field trial is limited to seven[6] sites totalling a maximum of 30 ha per year, only a small number of people would deal with the GM plant material and a small number of organisms are likely to be exposed to it. GM plant material is not to be used as animal feed.

95. Human contact with, or inhalation of, GM plant materials would be limited to people with access to the sites. The proposed trial sites are located on SRA stations accessible only by private road, therefore access to the general public would be minimised.

96. Sugarcane plants possess leaves with sharp edges and irritating hairs, as such, workers typically wear protective clothing which would reduce dermal contact.

97. After harvest, the applicant proposes to destroy GM sugarcane materials produced, apart from retaining some plant materials for research purposes and for new plants within the trial.

98. The applicant has also proposed a series of measures, such as the monitoring and inspecting of sites, together with the cleaning of equipment, the storing of seed and the disposal of waste material that will together help reduce exposure of people to GM plant material in sites and the adjacent areas.

Potential harm

99. People exposed to the proteins expressed from the introduced genes or their associated products may show toxic or allergenic reactions, while other organisms may show toxic reactions.

100. Proteins are not generally associated with toxic effects. As opposed to small molecular weight chemicals (eg pesticides), proteins have a number of properties that limit their ability to produce toxic effects upon ingestion, these include their likely digestion in the gastrointestinal tract and difficulties they encounter in traversing plasma membranes (Hammond et al. 2013). However, a small number of proteins have been shown to be toxic to humans and mammals, these originating mainly from animals (*eg* snakes, scorpions) and bacteria (Henkel et al. 2010; Karalliedde 1995).

[6] The applicant proposed an additional site in the Bundaberg LGA during consultation.

Lectins and protease inhibitors are plant proteins that have toxic properties, but for most people the level of exposure and response to the majority of these compounds is such that they are often classified as anti-nutrients (Delaney et al. 2008). The most well-known plant proteins that are definitely toxic to humans are the lectins that consist of a ribosome-inactivating peptide (RIP) linked to a carbohydrate binding peptide, examples being ricin, abrin and modeccin (de Virgilio et al. 2010; Stirpe 2005).

101. All known food allergens are proteins, those derived from plants coming chiefly from peanut, tree nuts, wheat and soybean (Delaney et al. 2008; Herman & Ladics 2011). The structural and functional properties of plant food allergens can be used to classify them into approximately 30 families, these then being grouped into a small number of superfamilies (Hauser et al. 2008; Radauer & Breiteneder 2007; Salcedo et al. 2008). The major superfamilies are the prolamins, cupins, pathogenesis-related (PR) proteins, profilins and protease inhibitors.

102. Chapter 1, Section 5.2 presents a review of the potential toxic and allergenic properties of the proteins encoded by the introduced genes. It was concluded that none of the introduced proteins were likely to be toxic to people or other organisms, or allergenic to people. In respect of the general information on toxic and allergenic plant proteins outlined above, none of the introduced plant proteins can be classified as a lectin or protease inhibitor (*i.e.* a likely toxin), or a prolamin, cupin, PR protein *etc* (*i.e.* a likely allergen).

103. For the evaluation of protein safety, the ILSI International Food Biotechnology Committee has collaborated with a group of experts to produce a two tiered method to assess the safety to health of proteins (Delaney et al. 2008; Hammond et al. 2013). The first tier examines five issues: (i) history of safe use of the protein; (ii) bioinformatics analysis; (iii) mode of action; (iv) *in vitro* digestibility and stability; and (v) expression level and dietary intake. Only if potential safety issues were identified in this evaluation would a second tier assessment be recommended, a prime example of such an assessment being a dose toxicology study.

104. The introduced proteins were examined against the first tier criteria:

(i) History of safe use. Non-GM sugarcane is not known to be toxic to humans or other organisms (OGTR 2011). Although no toxicity studies have been performed on the GM sugarcane plant material, the introduced genes were isolated from naturally occurring organisms that are widespread and prevalent in the environment, such as common food plants or naturally occurring bacteria (see Chapter 1, section 5.2 and 6.4). People and animals are exposed to most of the proteins produced by these genes through their diet and the environment. No information was found to suggest that any of the proteins encoded by the introduced genes are toxic or allergenic to people or other organisms (Chapter 1, Section 5.2).

(ii) Bioinformatic analysis. Although the HT1 gene has not been specifically studied, a highly similar protein has been assessed for allergenicity and toxicity. Bioinformatic comparison of the amino acid sequence of this gene did not identify similarities to known allergenic or toxic proteins. None of the introduced proteins are members of protein classes that are known to have members with toxic or allergenic properties. The HT2 gene is present in a GM food crop and is not expected to have any toxic or allergenic properties.

(iii) Mode of action. The known cellular roles of the proteins do not present any noteworthy concern.

(iv) *In vitro* digestibility to the introduced proteins. *In vitro* studies conducted with heterologously produced similar protein demonstrated that it was rapidly degraded in intestinal fluid and completely inactivated at temperatures above 56°C. No evidence of adverse effects was observed in mice following acute oral exposure or in a repeated dose dietary exposure study. This comprehensive assessment demonstrated that the protein does not present a risk for adverse effects in humans.

(v) Expression level and dietary intake. No data is available with regard to protein expression. The GM sugarcane and material obtained would not be used for human/animal consumption.

105. All the proteins successfully pass criteria (i), (ii), (iii) and (iv). Issues relating to points (v) may need to be addressed prior to a commercial release of any of the GM sugarcane lines.

106. Sugarcane pollen may be an allergen (Chakraborty et al. 2001), although allergic responses to the commercial hybrid cultivars of sugarcane have not been reported in Australia. Due to the limited quantities of pollen produced by sugarcane, it is expected that people would be exposed to very small quantities of pollen, if any. As discussed above, the introduced proteins in the GM sugarcane are not considered to be toxic or allergenic and the GM sugarcane lines are unlikely to be any more toxic or allergenic than non-GM sugarcane.

107. Organisms exposed to the proteins expressed from the introduced genes or their associated products may show toxic reactions. The information presented above regarding the potential toxicity of the proteins to humans may be applicable to other mammals; in the case of other organisms, there is no direct information. However, it is likely that only a small number of organisms will feed on the GM plant material. Moreover, as the introduced genes were isolated from naturally occurring organisms that are widespread and prevalent in the environment, such as common food plants or naturally occurring bacteria, it is likely that most organisms that may enter the trial sites and feed on the GM plants, have had prior to exposure to these proteins or their homologues.

108. Gene technology has the potential to cause unintended effects in several ways, including altered expression of an endogenous gene by random insertion of an introduced DNA in the genome, increased metabolic burden due to higher expression of the introduced protein, novel traits arising out of interactions with non-target proteins and secondary effects arising from altered substrate or product levels in biochemical pathways. Such an effect could lead to elevation of the concentration of a naturally occurring sugarcane compound to a level where it induces a toxic or allergenic reaction if consumed in an average diet. It is also possible that an entirely novel compound could be produced. Unintended changes such as an increase in the level of an endogenous toxin, can be induced in plants by conventional methods of plant breeding (Haslberger 2003). However, even though conventional breeding can involve the introduction of hundreds and even thousands of genes into a plant, there has never been a report of a novel toxin or allergen appearing in a new line of a plant produced by such techniques (Steiner et al. 2013; Weber et al. 2012). The implication is that the introduction into sugarcane of any of the genes that are the subject of this application is unlikely to result in the production of a novel toxin or allergen. This includes the production of such a compound via the site of insertion or the production of a fusion protein.

109. ***Conclusion***: Risk scenario 1 is considered to be a negligible risk due to the likely limited exposure of humans to the expressed proteins, and the predicted lack of significant toxicity or allergenicity of the introduced proteins to humans and other organisms. Therefore, this risk is not considered to be a substantive risk that warrants further detailed assessment.

2.5.2 Risk scenario 2

Risk source	*Causal pathway*	*Potential harm*
Introduced herbicide tolerance genes	Dispersal of GM plant material and seed outside trial limits ⬇ Growth of GM plants ⬇ Expression of genes in GM plants ⬇ Spread and persistence of populations of GM plants outside trial limits ⬇ Exposure of people or other organisms to GM plant material	Allergic reactions in people or toxicity in people and other organisms, and reduced establishment and yield of desirable plants, reduced biodiversity

Risk source

110. The source of potential harm for this postulated risk scenario is the introduced herbicide tolerance genes.

Causal pathway

111. The herbicide tolerance genes are present or remain present at a trial site after the trial has finished in plant tissues. If reproductive plant material and seed was dispersed outside any of the trial sites, these could give rise to plants expressing the introduced genes. These plants could spread and persist in the environment outside the trial limits, and people and other organisms may be exposed to GM plant materials.

112. Dispersal of GM plant material outside the limits of any trial site could occur through the activity of people (including the use of agricultural equipment), the activity of animals such as rodents, herbivores and birds, or through extremes of weather such as flooding or high winds.

113. Baseline information on the characteristics of weeds in general, and the factors limiting the spread and persistence of non-GM sugarcane plants in particular, is given in *The Biology of the* Saccharum *spp. (sugarcane)* (OGTR 2011). In summary, the document concludes that modern cultivars of non-GM sugarcane are not recognised as weeds in Australia where sugarcane occurs almost exclusively as a managed agricultural crop.

114. Modern sugarcane cultivars (*Saccharum spp.)* are not invasive in natural undisturbed environments and do not appear to be a problem as volunteer weeds in Australia. The establishment, spread and persistence of sugarcane populations is likely to be limited by complex interactions involving weed competition, pest infestation, disease infection, moisture stress, soil fertility and other environmental conditions (Bakker 1999; Hogarth & Allsopp 2000; OGTR 2011).

115. Although sugarcane can produce seeds, commercial sugarcane is propagated vegetatively, with setts (stem pieces) being planted in the field. Usually one bud per cutting develops into a primary stalk which then gives rise to tillers, for more details see *The Biology of the* Saccharum *spp. (sugarcane)* (OGTR 2011).

116. As discussed in the RARMPs for DIR 051/2004, DIR 070/2006, DIR 078/2007, DIR 095 and more recently DIR 096, viable sugarcane stems could be unintentionally dispersed from the sites during transportation. Sugarcane volunteers have been found growing along roadsides or railways in sugarcane cultivation areas. These volunteers are believed to have originated from stem cuttings fallen from vehicles during transportation and generally consist of only a few stools (a group of stems growing from a single original plant base) and normally do not become self-perpetuating or result in further spread.

117. In the course of the dealings the applicant proposes to transport GM sugarcane setts to the release sites, cultivate GM sugarcane plants and collect GM plant material for research purposes or

new plantings. Accidental spillage or dispersal of GM plant material, especially setts, in the course of these dealings could allow the GM sugarcane plants to establish and persist in the environment.

118. Transport of GM sugarcane plants and setts to and between SRA stations is proposed to be conducted according to transport guidelines issued by the Regulator. The applicant also proposes to transport harvested material between planting areas within SRA stations in trailers, with the GM sugarcane plant material being tied down.

119. Transport of GM sugarcane plants and setts according to the Regulator's transport guidelines would reduce the risk of dispersal. These procedures also require cleaning and monitoring of any areas where spills have occurred. In addition, appropriate environmental conditions are necessary for survival and persistence of any dispersed setts. For example, soil-borne fungal infections are known to reduce sett germination, leading to the common practice of treating the setts with fungicide prior to planting (FAO 2004).

120. During harvesting of GM sugarcane accidental mixing with non-GM sugarcane could occur through, for example, harvester driver error. This could lead to the GM sugarcane being inadvertently treated as non-GM sugarcane, thus allowing dispersal from the site as a component of a non-GM sugarcane harvest, or mixed with non-GM sugarcane that may be used for replanting at other locations. At the field sites, the applicant proposes to separate GM sugarcane from any adjacent sugarcane plots by a guard row of non-GM sugarcane and a 6 m isolation zone. This would clearly separate the GM sugarcane from neighbouring non-GM sugarcane and minimise the likelihood of accidental mixing at harvest.

121. In the crossing facilities, the applicant proposes to separate the GM sugarcane from non-GM sugarcane by at least 1 m and clearly label the GM sugarcane. These measures would minimise any inadvertent mixing of non-GM sugarcane with GM sugarcane. On the seedling benches at the field sites (SRA Woodford, Southern, Central and Burdekin), GM sugarcane and non-GM sugarcane plants would be clearly identifiable, including the use of barcoding of the seedling trays. Thus clear identification of GM plants would reduce the potential for accidental mixing through human error.

122. Persistence of GM sugarcane at the site could occur if GM plant material, such as stem segments, were to remain on site after harvest. All plant materials other than that collected for future research or for new plantings within the proposed release would be destroyed and the sites would be monitored for volunteers after the final harvest and any volunteer sugarcane plants would be destroyed. These measures would minimise the potential for persistence of GM sugarcane after the trial and the potential for its dispersal.

123. On completion of the trial the sugarcane plant material would be destroyed by a combination of harvesting, mulching, herbicide treatment and burning. For small trials that may be hand harvested, it has been proposed that stalks may be harvested and left to rot in the field. This could lead to dispersal of potentially viable stem pieces by animals. Small stem pieces are not expected to remain viable over long periods in the field, as they are degraded rapidly by soil micro-organisms under tropical conditions. Normally, stem cuttings need to be dipped in fungicide to enable them to survive until germination (FAO 2001; FAO 2004).

124. Severe weather conditions such as flooding may cause dispersal of GM plant material. However, control measures have been proposed by the applicant to minimise dispersal by flooding. These include locating the proposed release field sites at least 50 m away (with the exception of the SRA Meringa photoperiod facility) from natural waterways on land that is not subject to flooding. The applicant states that SRA Meringa, Burdekin, Durre, ISIS Central and Southern have no history of flooding. SRA Woodford is located on an elevated part of a flood-prone area and the applicant plans to use the highest part of the site for the trial, which has no history of flooding. The applicant states that flooding is rare at SRA Central and it is located 5 km from the nearest major waterway. The applicant also states that sugarcane plants are generally not uprooted by floodwaters, unless significant water flow occurs. Although the SRA Meringa photoperiod facility is within

approximately 20 m of a small natural waterway, the risk of dispersal of sugarcane from this facility is limited because the GM sugarcane grown at this location would be large plants in pots held within a large trolley (which would restrict movement of pots). In addition SRA Meringa has no history of flooding. All other parts of the crossing facility at SRA Meringa are more than 50 m from natural waterways.

125. As sugarcane can reproduce through vegetative cuttings, any viable sugarcane stem pieces left on the ground could be dispersed by pests such as feral pigs or other large animals. Dispersal of cane material through these means has not been reported to date.

126. The sexual reproduction of sugarcane has been little studied in the field, as seed production is not important for sugarcane cropping. Conditions for seed production have been studied in breeding facilities, however, these are conducted under optimal environmental conditions, and the relevance of these observations to field-grown sugarcane is limited. Sugarcane plants can flower and produce seed in the field where appropriate conditions exist for floral induction and production of viable pollen. These factors are discussed further in Risk scenario 3. In this application, the production of viable seed is most likely at SRA Meringa, and this station is not proposed to be used for field trials. The applicant proposes to monitor GM sugarcane at SRA Meringa for flowering, and contain inflorescences in pollen lanterns prior to spikelets opening to control dispersal of pollen.

127. Sugarcane seed is short-lived, losing 90% of viability within 80 days at 28ºC unless it is desiccated (Rao 1980). More recent data suggests that seed can remain viable for at least 2-3 months when stored at room temperature and that viability is somewhat cultivar dependent (Powell et al. 2008). Sugarcane seed needs appropriate environmental conditions for germination (eg. temperature and humidity) and these are seldom present without human intervention. Furthermore, seedlings require particular favourable environmental conditions (eg. temperature and humidity) to survive for the first three to four weeks after germination (Breaux & Miller 1987) and normally human intervention is required to successfully establish sugarcane seedlings. In a recent study on abiotic limits for germination of sugarcane seed, Pierre et al (2014) concluded that water availability is the main factor limiting seed germination in Australia rather than temperature. Fertile sugarcane seed production in Australia occurs during the driest time of the year when monthly rainfall is between 40 – 70 mm, even though the mean maximal temperature is around 26^{o}C, which is below the optimal range of 27^{o}C to 36^{o}C arrived at by the study. It is therefore highly likely that environmental factors, such as water availability and fluctuations in temperature, would affect seed viability and limit the survival and establishment of any dispersed seeds.

128. Observations over a number of decades indicate a lack of seed germination in regions south of the Burdekin region (the Burdekin region is referred to as the region around Ayr, south of Townsville) (Sugarcane Researchers 2008), and only low numbers of seedlings north of the Burdekin region (see Figure 4). For example, as discussed in DIR 096, sugarcane growers and researchers occasionally observe sugarcane seedlings in the field in the Herbert district near Ingham, and further north. It is unknown whether any field-germinated seedlings survive to maturity. Of the SRA stations proposed to be used for the trial, only SRA Meringa is located in northern Qld, an area where environmental conditions, including temperature, are thought to be conducive to sugarcane seed germination in the field. At this facility, the applicant proposes to grow the sugarcane in pots in a photoperiod facility and contain open inflorescences in pollen lanterns or in bags for drying seed, and so the dispersal of seed into the environment is considered highly unlikely. The applicant states that any pollen would only be able to be transferred to other sugarcane flowers present in the crossing facility, as flowering of field grown sugarcane at Meringa is usually finished by the time the sugarcane in the photoperiod facility commences flowering; the initiation of flowering in the photoperiod facility is delayed to optimise synchronised flowering of the sugarcane plants within the facility. At the Locations in the Burdekin region (SRA Durre and Burdekin) and south (SRA Central, Woodford and Southern) environmental conditions are not considered suitable for fertile seed production, seed germination and establishment.

129. There is no data to suggest that seed viability or dispersal would be altered in the GM sugarcane plants compared to the non-GM parental sugarcane plants (Risk scenario 2). Survival of any GM sugarcane seedlings would be limited by factors such as humidity, temperature, low intrinsic competitive ability, nutrient availability, pests and diseases and other environmental factors that normally limit the spread and persistence of sugarcane plants in Australia. In addition, the genetic modification is unlikely to increase the ability of the GM sugarcane to survive outside the area of cultivation.

130. Currently there is no evidence to indicate that GM herbicide tolerant crops are more invasive or persistent than their conventional counterparts (Crawley et al. 2001). Although GM herbicide tolerant sugarcane has not been specifically evaluated for its invasiveness, other herbicide tolerant crops, including oilseed-rape, sugar beet and maize were found to be no more invasive than their non-GM counterpart (Crawley et al. 2001). The impact of the genetic modifications on survival of the GM sugarcane plants is uncharacterised under field conditions. However, a number of predictions can be made based on knowledge of the gene functions and their effects when expressed in the GM plants. Predictions can also be made based on the observed phenotypes of other GM plants expressing the same genes.

131. The GM sugarcane plants contain introduced genes for herbicide tolerance (Chapter 1, Section 5.1.1). These GM sugarcane lines could have a selective advantage in an environment where the application of herbicides is a standard practice. Such a scenario could occur within the cultivation setting at the SRA stations, where the GM sugarcane could establish and persist if herbicides were used exclusively as part of the agricultural practices to manage the cultivation of sugarcane, eg. when removing unwanted volunteers.

132. In the highly unlikely event that the GM sugarcane was dispersed into an area where the specific herbicides were applied, it may have an advantage over non-GM sugarcane. However, the herbicide tolerance is unlikely to significantly increase the weediness potential of the GM sugarcane because the sugarcane would still be susceptible to the application of other herbicides, and control could still be achieved through herbicide application and/or by non-chemical methods. The spread and persistence of the GM sugarcane plants would also be limited by multiple factors that normally limit the spread and persistence of sugarcane in Australia such as its low fertility and seed viability, poor ability of seedlings to establish and compete without human intervention, nutrient requirements, and susceptibility to pests and diseases (Bakker 1999; Hogarth & Allsopp 2000; OGTR 2011).

133. Studies conducted with these genes introduced in another food crop did not indicate any change in factors that may improve survival such as seed dormancy, improved germination and seedling vigour as compared to the non-GM comparator. Equivalent data is not available for the GM sugarcane lines.

134. In addition, a reduction in plant vigour is routinely observed in sugarcane plants which have undergone tissue culture. Data provided by the applicant from release DIR 070/2006 show that plant height, stalk number, stem diameter and cane yield were reduced in GM sugarcane expressing *nptII* compared to untransformed sugarcane. These effects are expected to generally decrease the competitiveness of GM sugarcane.

Potential Harm

135. If the GM sugarcane plants were to establish or persist better in the environment than non-GM sugarcane, then this could lead to one or more harms. The potential for increased allergenicity in people or toxicity in people and other organisms as a result of contact with GM plant materials has been considered in Risk Scenario 1 and was not considered to be a risk that would be greater than negligible.

136. If the introduced genes provide the GM sugarcane plants with a significant selective advantage over non-GM sugarcane plants and if they were able to establish and persist in favourable non-agricultural environments, this may give rise to lower abundance of desirable species, reduced species richness, or undesirable changes in species composition. Similarly, the GM sugarcane plants could adversely affect agricultural environments if they exhibited a greater ability to establish and persist than non-GM sugarcane[7].

137. ***Conclusion:*** Risk scenario 2 is considered to be a negligible risk as none of the intoduced traits are associated with weediness, it is unlikely that any of the characteristics associated with weeds will occur in the GM plants, and because of the limits and controls proposed for the field trial. Therefore, this risk is not considered to be a substantive risk that warrants further detailed assessment.

2.5.3 Risk scenario 3

Risk source	*Causal pathway*	*Potential harm*
Introduced herbicide tolerance genes	Dispersal of GM pollen outside trial limits ⬇ Vertical transfer of introduced genes to other sexually compatible plants. ⬇ Expression of genes in plants ⬇ Exposure of people or other organisms to GM plant material	Allergic reactions in people or toxicity in people and other organisms

Risk source

138. The source of potential harm for this postulated risk scenario is the introduced herbicide tolerance genes.

Causal pathway

139. Pollen from the GM sugarcane could be dispersed outside the limits of the trial. Pathways include:

(a) Dispersal of pollen from GM sugarcane at the trial to sexually compatible plants outside the trial sites during the trial, or

(b) Dispersal of GM seed outside the trial sites during the trial leading to persistence of GM sugarcane outside and/or after the trial has finished leading to pollen dispersal to sexually compatible plants.

Sexually compatible plants include commercial non-GM sugarcane as well as GM sugarcane trialled under other DIR licences. Other plant species are also somewhat compatible under natural conditions (see Chapter 1, section 6.3). Any hybrids resulting from fertilisation with GM sugarcane pollen could pass the genetic modification on to their offspring.

140. People and other organisms could be exposed to the proteins introduced through contact with (including inhalation of pollen) or consumption of GM plant material.

141. Vertical gene flow is the transfer of genetic information from an individual organism to its progeny by conventional heredity mechanisms, both asexual and sexual. In flowering plants, pollen dispersal is the main mode of gene flow (Waines & Hegde 2003). For GM crops, vertical gene flow could therefore occur via successful cross-pollination between the crop and neighbouring crops, related weeds or native plants (Glover 2002).

142. Baseline information on vertical gene transfer associated with non-GM sugarcane plants can be found in *The Biology of the* Saccharum *spp. (sugarcane)* (OGTR 2011). In summary, sugarcane

[7] The potential of these harms can be evaluated against the experience of other non-GM and GM herbicide tolerant crops. No commercially released herbicide tolerant crop has been recorded to have negatively impacted on the environment (or the health of humans or animals) beyond that normally associated with the crop.

pollen viability is low under natural conditions, commercial sugarcane varieties show low fertility and crossing to plants outside of the *Saccharum* genus has rarely been observed. Thus, it is highly unlikely that crossing with sexually compatible plants would occur.

143. 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.

144. The dispersal of seed was considered in Risk scenario 2, and concluded to be unlikely due to the limits and controls of the trial.

145. Sugarcane is cultivated within close proximity to all of the proposed trial sites. At SRA Woodford, ISIS Central, Southern, Central, Durre and Burdekin, non-GM sugarcane is cultivated within the SRA stations at a minimum distance of 6 m from the proposed trial sites. At SRA Southern, Central and Burdekin, commercial sugarcane is propagated in adjacent properties at a minimum distance of 10 m from the proposed trial sites. Within the crossing facility at SRA Meringa, the applicant proposes to enclose open GM sugarcane inflorescences prior to spikelet opening, in pollen-impermeable lanterns in order to separate them from non-GM sugarcane inflorescences.

146. As described in the RARMP for DIR 078/2007, DIR 095, DIR 096 and *The Biology of the* Saccharum *spp. (sugarcane) (OGTR 2011)*, sugarcane does not flower with great uniformity, and cultivars differ in their propensity to flower (Bonnett et al. 2007). Shortening day length appears to control the initiation of flowering (Moore & Nuss 1987) and environmental factors impact on the extent of flowering. The frequency of flowering in the Burdekin region (Figure 4) and further south, is generally lower than north of the Burdekin region, however this has not been systematically recorded.

147. Sugarcane is principally a wind pollinated out-crosser with a low frequency of self-pollination. Environmental conditions including temperature, relative humidity and wind intensity have a great influence on pollen viability and pollen movement (discussed below). There is little information available on outcrossing rates and distances for sugarcane.

148. Sugarcane pollen desiccates rapidly and is not viable beyond 35 minutes at 26.5ºC at 65% relative humidity (Moore 1976; Venkatraman 1922). Sugarcane pollen viability is variable and is strongly influenced by temperature, being greatly reduced under night-time temperatures below 21^{o}C (Berding 1981). In Qld, this leads to a general reduction in levels of pollen viability the further south sugarcane is cultivated. The applicant has provided data showing that pollen viability on inflorescences sampled from commercial crops in the Mulgrave region (north of the Burdekin region) was significantly higher than in samples from the Burdekin region, which showed very low pollen viability (Bonnett et al. 2007). These reports indicate that the frequency of flowering and production of viable pollen is very low in most parts of Qld. In addition, commercial sugarcane is often harvested prior to flowering, which would further limit the opportunity for pollen transfer to nearby commercial crops.

149. North of the Burdekin region (i.e. at SRA Meringa, Figure 4), environmental conditions are more suitable for viable pollen production, viable seed production, seed germination and seedling survival. At SRA Meringa the GM sugarcane plants would be grown in the photoperiod facility in pots and monitored for flowering. Flowering plants would have their inflorescence removed shortly before spikelet opening and transferred to a crossing shed, where they would be enclosed in a pollen lantern. This would minimise the dispersal of pollen, thus limiting gene flow to other sugarcane plants. In the event that spikelets open prior to inflorescences being enclosed in pollen lanterns, the open spikelets would be removed and destroyed, similarly any open spikelets on nearby non-GM inflorescences which are not enclosed in pollen lanterns would also be removed and destroyed, thus preventing set of seed. These measures would minimise the dispersal of pollen and the potential for cross-pollination to lead to seed set, thus limiting potential gene flow to other sugarcane plants in crossing facilities. The applicant states that flowering of field grown sugarcane is usually finished

by the time the sugarcane in the photoperiod facility commences flowering, as the initiation of flowering in the photoperiod facility is delayed to optimise synchronised flowering of the sugarcane plants within the facility. As such any pollen would only be able to be transferred to other sugarcane flowers present at the photoperiod facility.

150. The proposed limits and controls of the trial (Chapter 1, Sections 3.1 and 3.2) would restrict the potential for pollen flow and gene transfer to sexually compatible plants. The applicant also proposes to perform post-harvest monitoring of the sites for at least twelve months and until no volunteers are observed for a continuous six month period and to destroy any volunteer plants found at the sites. This would ensure that any remaining GM sugarcane seeds or plants that were potentially the product of gene flow in these areas would be destroyed.

Potential harm

151. People who are exposed to the proteins expressed from the introduced genes or their associated products through contact or consumption of GM plant material may show toxic or allergenic reactions, while organisms may show toxic reactions from consumption of GM plant material.

152. In the rare event of vertical transfer of the introduced genetic material from the GM plants to sexually compatible plants, the genetic material is expected to behave in similar ways as in the GM sugarcane. As discussed in Risk scenario 1, the introduced gene products are not expected to be toxic or allergenic to people, or toxic to other organisms and there is no reason to expect an increased level of toxicity/allergenicity in any of the offspring of the GM sugarcane.

153. ***Conclusion***: Risk scenario 3 is considered to be a negligible risk due to the lack of toxicity or allergenicity of the introduced proteins to humans or other organisms, there being no reasonable expectation that toxicity or allergenicity would be increased in any offspring of the GM plants. Therefore, this risk is not considered to be a substantive risk that warrants further details assessment.

2.5.4 Risk scenario 4

Risk source	*Causal pathway*	*Potential harm*
Introduced herbicide tolerance genes	Dispersal of GM pollen outside trial limits ⬇ Vertical transfer of introduced genes to other sexually compatible plants. ⬇ Expression of genes in plants ⬇ Spread and persistence of populations of GM plants outside a trial site	Reduced establishment and yield of desirable plants, reduced biodiversity

Risk source

154. The source of potential harm for this postulated risk scenario is the introduced herbicide tolerance genes.

Causal pathway

155. The herbicide tolerance genes are present in plant tissues including pollen. Pollen from the GM plants could be transferred outside of a trial site (*eg* via wind) and fertilise sexually compatible plants, whether they be non-GM sugarcane, or plants from another species. Alternatively, if seed was dispersed outside any of the trial sites, plants expressing the introduced genes may grow and disperse pollen. Hybrid plants possessing the introduced genes may form the basis of the spread of these genes in other (initially non-GM) varieties of sugarcane, or other plant species. People and other organisms could be exposed to the proteins expressed from the introduced genes through contact with (including inhalation of pollen) or consumption of GM plant material derived from the plants to which the genes have been transferred.

156. The dispersal of seed was considered in Risk scenario 2, and concluded to be unlikely due to the limits and controls of the trial. Vertical gene transfer was reviewed in Risk scenario 3. The proposed limits and controls of the trial would minimise the possibility that seed or pollen would be dispersed outside the trial limits.

Potential harm

157. If the vertical gene transfer of the introduced genes from the GM plants causes the recipient species to spread and persist in the environment to a degree greater than the non-GM recipient species, then this may lead to a greater level of harm from these species. These potential harms were summarised in Risk scenario 2. In particular, the GM plants may act to reduce the establishment and yield of desired plants and subsequently reduce biodiversity.

158. Risk scenario 2 summarises the reasons that the introduced genes are unlikely to make the GM sugarcane lines more weedy and leading to harm, these reasons being applicable to any non-GM plants to which the genes are transferred.

159. ***Conclusion***: Risk scenario 4 is considered to be a negligible risk, as none of the introduced traits have been associated with weediness. It is unlikely that any of the characteristics associated with weeds will occur in the GM plants themselves or any plants that are the products of the hybridisation of the GM plants and other non-GM plants. Therefore, this risk is not considered to be a substantive risk that warrants further detailed assessment.

2.5.5 Risk scenario 5

Risk source	*Causal pathway*	*Potential harm*
Introduced herbicide tolerance genes	Dispersal of GM pollen within a site ⬇ Hybridisation of GM plants of this trial with GM plants (including volunteers) of another trial ⬇ Expression of genes in stacked GM plants ⬇ Exposure of people or other organisms to GM plant material	Allergic reactions in people or toxicity in people and other organisms

Risk source

160. The source of potential harm for this postulated risk scenario is the introduced herbicide tolerance genes.

Causal pathway

161. The applicant has indicated that non-GM commercial and GM or non-GM research crops of sugarcane will be present at or near the proposed release sites. GM sugarcane developed under DIR 095 may be planted at the same stations. These GM sugarcane plants contain introduced genes for drought tolerance, enhanced nitrogen use efficiency, altered plant growth, altered sucrose production, altered sucrose accumulation and increased efficiency of post-harvest processing for cellulosic ethanol.

162. Low levels of gene flow both within and between the sugarcane trials at the SRA stations may lead to stacking of GM traits, thus producing a GM sugarcane plant with genes for herbicide tolerance as well as traits in DIR 095. If hybrids were to occur and persist, people working in such trials could be exposed to GM hybrid plants. This may give rise to an increased level of toxicity in people compared to any individual GM sugarcane plant in the proposed release. However, at the field trial sites, environmental conditions are not favourable for the production of significant amounts of viable pollen, making it unlikely that stacking will occur.

163. If cross-pollination were to occur, combination of traits through stacking is likely to contribute only incrementally to the ability of the GM hybrid plants to spread and persist. Spread and persistence would be limited by factors such as low fertility and seed viability, poor ability to

establish and thrive without human intervention, competition with other plants, soil type and fertility, and pests and diseases that limit the spread and persistence of non-GM sugarcane in Australia (see Risk scenario 2). Environmental conditions required for seed germination and establishments are only found in northern Queensland. At the crossing facilities at SRA Meringa, GM sugarcane plants will not be grown in the field and measures are proposed to minimise pollen transfer to other sugarcane plants (see Risk scenario 3).

164. Therefore, the general prevailing environmental conditions at the sites and the proposed limits and controls of the trial would restrict the potential for pollen flow and successful gene transfer to other GM plants at the proposed sites.

165. The applicant has also proposed limits and controls to minimise the dispersal of pollen in the glasshouse and crossing facilities. GM sugarcane plants would be monitored for spikelet opening and the inflorescences enclosed in pollen lanterns prior to spikelet opening, and to destroy any open spikelets not enclosed in pollen lanterns.

166. Vertical gene transfer associated with sugarcane plants was discussed in risk scenario 3. In summary, sugarcane pollen viability is extremely low under natural conditions and commercial sugarcane varieties show very low fertility. Thus, it is highly unlikely that crossing with other GM plants would occur.

167. The proposed limits and controls of the trial would minimise the possibility that seed or other plant material would leave any trial site. These are discussed in risk scenario 2. All GM seed and plant material will be transported in accordance with the Regulator's transport guidelines, which will minimise the opportunity for its dispersal.

Potential harm

168. People who are exposed to the proteins expressed from the introduced genes or their associated products through contact or consumption of GM plant material may show toxic or allergenic reactions, while organisms may show toxic reactions from consumption of GM plant material.

169. As discussed in Risk scenario 1, the introduced gene products of this application are not expected to be toxic or allergenic to people, or toxic to other organisms and there is no reason to expect the production of an associated compound with a toxic or allergenic property. The toxicity and allergenicity associated with the introduced genes of other sexually compatible GM plants growing at SRA trial sites have been assessed earlier. According to the RARMPs for DIR 078/2007 and DIR 095, there is no information to suggest that the proteins encoded by the introduced genes of those applications are likely to be toxic to people or other organisms, or allergenic to people. As discussed above (Risk scenario 4), plants that are the product of hybridisation between two GM varieties are unlikely to exhibit a level of toxicity or allergenicity greater than that of either parent (*i.e.* the stacking of genes from different GM plants will be unlikely to generate a plant with a higher level of toxicity or allergenicity than the individual GM parents).

170. ***Conclusion***: Risk scenario 5 is considered to be a negligible risk, due to the predicted lack of significant toxicity or allergenicity of the introduced proteins to humans or other organisms, and the nature of the limits and controls imposed on the field trial. Therefore, this risk is not considered to be a substantive risk that warrants further detailed assessment.

2.5.6 Risk scenario 6

Risk source	*Causal pathway*	*Potential harm*
Introduced herbicide tolerance genes	Dispersal of GM pollen within a site ⬇ Hybridisation of GM plants of this trial with GM plants (including volunteers) of another trial ⬇ Dispersal of plants or viable plant material containing stacked genes outside a site ⬇ Expression of genes in stacked GM plants ⬇ Spread and persistence of populations of GM plants outside a trial site	Reduced establishment and yield of desirable plants, reduced biodiversity

Risk source

171. The source of potential harm for this postulated risk scenario is the introduced herbicide tolerance genes.

Causal pathway

172. As discussed in Risk Scenario 5, the applicant has indicated the presence of commercial or research crops of sugarcane at or near the proposed release sites. Pollen from the GM plants of one trial could inadvertently fertilise other sexually compatible GM plants, including volunteers, inside a site. GM sugarcane with drought tolerance, enhanced nitrogen use efficiency, altered plant growth, altered sucrose production, altered sucrose accumulation and increased efficiency of post-harvest processing for cellulosic ethanol from DIR 095 could occur at the proposed locations.

173. If hybridisation occurred between the GM plants of this application and those of DIR 095, the progeny would have the traits of herbicide tolerance stacked with traits in DIR 095.

174. If plant material, which unknowingly was the product of hybridisation between the GM plants of this trial and other GM plants in any site, was purposely taken outside of a trial site, or was inadvertently dispersed outside of a trial site, GM hybrid plants could arise from the germination of the seed. These plants then could spread and persist in the environment. However, it is unlikely that the stacking of the herbicide tolerant trait of this application with those of the traits of the GM plants of DIR 095 (drought tolerance, enhanced nitrogen use efficiency, altered plant growth, altered sucrose production, altered sucrose accumulation and increased efficiency of post-harvest processing for cellulosic ethanol) will significantly influence the invasiveness of the plants, but this is an area of uncertainty. As discussed above (Risk scenario 5), the combination of such traits is not likely to produce a plant that is more invasive than the individual parents.

175. Risk scenario 5 reviews the growing of GM plants from different trials adjacent to each other, such as possibly may occur at the SRA trial sites, noting the applicant proposes using guard rows and isolation zones to restrict pollen flow.

176. The proposed limits and controls of the trial would minimise the possibility that seed would leave any trial site. These are discussed under Risk scenario 2. All GM seed will be transported in accordance with the Regulator's transport guidelines, which will minimise the opportunity for its dispersal.

Potential harm

177. If the vertical transfer of genes from the GM plants causes the recipient species to spread and persist in the environment to a degree greater than non-GM species, they may lead to a higher level of one or more harms. These harms were summarised in Risk scenario 2. In particular, the GM plants may act to reduce the establishment and yield of desired plants and biodiversity.

178. As discussed in Risk scenario 2, the introduced gene products of this application are unlikely to cause the GM sugarcane lines to have a greater negative impact on the environment than non-GM sugarcane. The weediness associated with the introduced genes of other sexually compatible GM plants growing at the SRA trial sites or at the other field trial sites were assessed in the RARMP for DIR 095. There is no information to suggest that the proteins encoded by the introduced genes of those applications are likely to increase weediness in GM sugarcane.

179. ***Conclusion***: Risk scenario 6 is considered to be a negligible risk, as none of the introduced traits are associated with weediness, it is unlikely that the stacking of genes from different GM trials will lead to a weedy plant. Limits and controls are proposed on the field trial to limit cross-pollination. Therefore, this risk is not considered to be a substantive risk that warrants further detailed assessment.

Section 3 Uncertainty

180. Uncertainty is an intrinsic part of risk analysis[8]. There can be uncertainty about identifying the risk source, the causal linkage to harm, the type and degree of harm, the chance of harm occurring or the level of risk. In relation to risk management, there can be uncertainty about the effectiveness, efficiency and practicality of controls.

181. Risk analysis can be considered as part of a first tier uncertainty analysis, namely a structured, transparent process to analyse and address uncertainty when identifying, characterising and evaluating risk. However, there is always some residual uncertainty that remains. If the residual uncertainty is important and critical to decision making, then this residual uncertainty may be subjected to further analysis (=second tier uncertainty analysis), such as building 'worst case' scenarios, or by using meta-analysis where results from several studies are combined.

182. There are several types of uncertainty in risk analysis (Bammer & SmithsonBammer & Smithson 2008; Clark & Brinkley 2001; Hayes 2004). These include:

- uncertainty about facts:
  - knowledge – data gaps, errors, small sample size, use of surrogate data
  - variability – inherent fluctuations or differences over time, space or group, associated with diversity and heterogeneity
- uncertainty about ideas:
  - description – expression of ideas with symbols, language or models can be subject to vagueness, ambiguity, context dependence, indeterminacy or under-specificity
  - perception – processing and interpreting risk is shaped by our mental processes and social/cultural circumstances, which vary between individuals and over time.

183. For DIR 129, uncertainty is noted particularly in relation to the characterisation of:

- Potential increases in toxicity or allergenicity as a result of the genetic modification
- Potential for increased survival of the GMOs due to unintended effects, including in land uses outside of agriculture.

184. Additional data, including information to address these uncertainties, may be required to assess possible future applications for a larger scale trial, reduced containment conditions, or the commercial release of these GM sugarcane lines if they are selected for further development.

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

[8] A more detailed discussion of uncertainty is contained in the Regulator's *Risk Analysis Framework* available from the Risk Assessment References page on the OGTR website or via Free call 1800 181 030.

Section 4 Risk evaluation

186. 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.

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

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

188. Six risk scenarios were postulated whereby the proposed dealings might give rise to harm to people or the environment. The level of risk for each scenario was considered negligible in relation to both seriousness and likelihood of harm, in the context of the control measures proposed by the applicant, and considering both the short and long term. The principal reasons for these conclusions are summarised in Table 3 and include:

- limits on the size, locations and duration of the release proposed by SRA
- controls proposed by SRA to restrict the spread and persistence of the GM sugarcane plants and their genetic material
- the genetic modifications are unlikely to give rise to adverse effects on human health and safety or the environment
- widespread presence of the same and similar genes, proteins and associated products in the environment and lack of evidence of harm from them
- limited ability and opportunity for the GM sugarcane plants to transfer the introduced genes to commercial sugarcane crops or other sexually related species
- none of the GM plant materials or products will enter human food or animal feed supply chains.

189. The *Risk Analysis Framework (OGTR 2013)*, which guides the risk assessment and risk management process, defines negligible risks as insubstantial with no present need to invoke actions for their mitigation. Therefore, no controls are required to treat these negligible risks. Therefore, 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

Section 1 Background

190. 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, evaluates controls and limits proposed by the applicant, and considers general risk management measures. The risk management plan informs the Regulator's decision-making process and is given effect through licence conditions.

191. 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 are able to be managed in a way that protects the health and safety of people and the environment.

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

193. 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. 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 identified risks

194. The risk assessment of risk scenarios listed in Chapter 2 concluded that there are negligible risks to people and the environment from the proposed field trial of GM sugarcane. These risk scenarios were considered in the context of the scale of the proposed release (Chapter 1, Section 3.1), the proposed containment measures (Chapter 1, Section 3.2), and the receiving environment (Chapter 1Section 6), and considering both the short and the long term. The risk evaluation concluded that no controls are required to treat these negligible risks.

Section 3 General risk management

195. 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 drafted to limit the release to the proposed size, locations and duration, and to restrict the spread and persistence of the GMOs and their genetic material in the environment. The conditions are detailed in the licence and summarised in this Chapter.

3.1 Licence conditions to limit and control the release

3.1.1 Consideration of limits and controls proposed by Sugar Research Australia

196. Sections 3.1 and 3.2 of Chapter 1 provide details of the limits and controls proposed by SRA in their application. These are discussed in the risk scenarios postulated for the release in Chapter 2. Many of these control measures are considered standard for GM crop trials and have been imposed by the Regulator in previous DIR licences. The appropriateness of these controls is considered further below.

197. The duration of the field trial is confined to six years. The field trial is limited to seven sites, with a collective field trial area of 30 ha per season and 1000 m^2 of nursery area. The small size and relatively short duration of the trial limits the potential exposure of humans and other organisms to the GMOs (Risk scenario 1).

198. Only authorised personnel with appropriate training are permitted to deal with the GMOs. This measure limits the potential exposure of humans to the GMOs (Risk scenario 1). A standard licence condition requires all people dealing with the GMOs to be informed of relevant licence conditions.

199. The field trial sites are located more than 50 m from the nearest natural waterways on land which has minimal risk of flooding, which reduces the likelihood of plant material being washed away from the sites (Risk Scenario 2). Although the SRA Meringa photoperiod facility is within approximately 20 m of a small natural waterway, the risk of dispersal of sugarcane from this facility is limited because GM sugarcane at this location would be large plants in pots held within a large trolley (which would restrict movement of pots). In addition, SRA Meringa has no history of flooding which reduces the likelihood of plant material being washed away from the site.

200. At the field sites, the applicant proposes to surround the GM sugarcane with a guard row of non-GM sugarcane and a further 6 m isolation zone. This physical separation helps prevent inadvertent mixing of cane in adjacent plots, through for example harvester driver error at the time of harvest. The applicant has stated that the nearest non-GM sugarcane is at least 6 m from any GM sugarcane and commercially grown sugarcane at a distance of at least 10 m. Separation of other non-GM sugarcane by at least 6 m from the proposed GM sugarcane reduces the likelihood of inadvertent mixing with the GM sugarcane through harvester driver error at the time of harvest. The guard row and 6 m isolation zone as a method for physical separation are therefore included in the licence. The applicant has also proposed to mark GM sugarcane field plantings with star pickets, and display signs indicating GM planting. As separation of GM from non-GM sugarcane with a guard row and 6 m isolation zone is considered an effective measure to reduce the likelihood of inadvertent mixing at harvest, these further measures proposed by the applicant have not been included as a licence condition.

201. At the SRA Durre, Burdekin, Central, Southern, Woodford stations and ISIS Central Sugar Mill Co. Ltd, where planting in the field is to occur, the applicant has proposed to cultivate GM sugarcane plantlets on seedling benches prior to planting in the field, using areas set aside for this purpose. Non-GM sugarcane plants are also cultivated in these facilities. The applicant proposes to keep GM and non-GM plants on separate benches, and label the GM material, including the implementation of a barcoding system to allow for identification of the GM sugarcane plants and plant material during the various stages of assessment. In order to further reduce the potential of unintended dispersal of potted GM sugarcane by, for example, workers unaware of restrictions on their movement, the licence conditions include a requirement that the GM plants kept be clearly identifiable, and to have signs indicating which benches contain GM material.

202. In crossing facilities, the applicant has similarly proposed to barcode label GM sugarcane and maintain the GM sugarcane in clearly signed areas separated by at least 1 m from non-GM sugarcane with the exception of those used for crossing. Clear labelling and 1 m separation to reduce the potential for inadvertent mixture of GM and non-GM material have been included as licence requirements.

203. Sugarcane pollen is thought to be produced in limited quantities and have low viability. Sugarcane has day length and temperature requirements for flowering and viable pollen production, which effectively limits the production of significant amounts of viable pollen to areas of northern Qld (north of the Burdekin region, Figure 4). In addition, the environmental conditions

required for seed germination and establishment are only found in northern Qld. Seed germination and establishment is also limited by factors such as low intrinsic competitive ability, nutrient availability, pests and diseases, factors that normally limit the spread and persistence of sugarcane plants in Australia (see Risk scenario 3).

204. The applicant intends to conduct crossing at SRA Meringa where environmental conditions are suitable for flowering, viable pollen production, seed germination and seedling survival. The applicant has proposed to inspect GM sugarcane at the crossing facilities for signs of spikelet opening three times a week in the period when the plants are likely to flower and enclose GM sugarcane inflorescences in pollen impermeable bags with the openings tied up prior to spikelet opening. Based on results by Skinner (1959), such measures have shown no evidence of pollen escape. The frequency of inspection is considered appropriate for experienced sugarcane breeders in order to predict the onset of spikelet opening. Before spikelets begin to flower, inflorescences to be crossed would be cut and transferred to the crossing shed. These inflorescences would be covered by pollen impermeable lanterns which would be secured at the base of the inflorescence. After seed setting, seed-bearing flowers would be collected in seed impermeable muslin bags and dried in a dehydrator for long-term storage. Sugarcane plants in the crossing facility would be separated by at least 1 m from other sugarcane plants and pollen would be contained within the pollen lanterns, limiting the potential for gene flow (Risk scenario 3 - 6). The applicant has also proposed to remove and destroy any spikelets that may open prior to inflorescences being enclosed in pollen lanterns, similarly any open spikelets on nearby non-GM inflorescences which are not enclosed in pollen lanterns would also be removed and destroyed, thus preventing seed set. These measures would minimise the dispersal of pollen and the potential for cross-pollination to lead to seed set, thus limiting potential gene flow to other sugarcane plants in crossing facilities. These control measures have been included in the licence. At SRA Meringa, non-GM sugarcane is cultivated in the field, where environmental conditions are more suitable for flowering, viable pollen production, seed germination and seedling survival. The applicant states that any pollen dispersal in the crossing facility at SRA Meringa would only be able to be transferred to other sugarcane flowers present at the crossing facility as flowering of field grown sugarcane is usually finished by the time sugarcane in the photoperiod facility commences flowering (Risk scenario 4 and 5).

205. The applicant has stated that any plant material taken off-site for experimental analysis will be transported according to the *Regulator's Guidelines for the transport, storage and disposal of GMOs*. These are standard protocols for the handling of GMOs to minimise exposure of people and other organisms to the GMOs (Risk scenario 1), dispersal into the environment and gene flow/transfer (Risk scenarios 2, 3 and 4). This is included as a licence condition. As discussed in Risk scenario 2, the applicant has proposed to transport sugarcane stems within SRA stations in trailers with the GM sugarcane plant material being tied down. Sugarcane stems are considered propagative plant material, and can potentially give rise to new sugarcane plants. This proposed method of transportation is effectively a lower level of containment than specified by the Regulator's transport guidelines, which require any propagative material to be contained within primary and secondary unbreakable containers, to minimise the risk of dispersal of GMOs. Displacement of stem pieces from trailers during transport could potentially lead to unintended establishment of GM sugarcane plants, thus increasing human and animal exposure to the GMOs (risk scenario 1), allowing the GM sugarcane to disperse (Risk scenario 2), and potentially enabling gene flow (Risk scenario 3 - 6). Therefore, the transport of all GM sugarcane material within SRA stations, is required to occur according to the specified conditions on transport and packaging included in the licence conditions.

206. The applicant has proposed to destroy any viable GM plant material after final harvest by either burning, mulching or by herbicide treatment (Risk scenario 2). Similar disposal and/or

destruction methods have been used previously for GM sugarcane under the licences for DIR 019/2002, DIR 051/2004, DIR 070/2006, DIR 078/2007, DIR 095 and DIR 096, and these are also used in the industry as standard destruction methods (OGTR 2011). These methods of destruction would minimise exposure to the GM plant material (Risk scenario 1) and limit the likelihood of spread and persistence (Risk scenario 2). For small trials that may be hand harvested, it has been proposed that stalks may be harvested and left to rot in the field. As discussed in Risk scenario 3, there is the potential for dispersal of viable cane pieces by animals and extreme weather conditions. Harvesting and leaving to rot in the field is not considered an acceptable method of destruction, and is therefore not listed as an appropriate destruction method in the draft licence and acceptable methods of destruction, including herbicide treatment, mulching and burning have been specified in the licence.

207. The applicant has proposed to use herbicides for the destruction of GM sugarcane with herbicides from groups different to which the GM sugarcane are expected to be tolerant (Risk scenario 2). The availability of herbicides to which the GM sugarcane is susceptible is a necessity, as it will allow for the destruction of GM sugarcane by this method. Therefore, the use of such alternative herbicides for this purpose will be specified in the licence.

208. The applicant has proposed to monitor the field release sites for 12 months post-harvest and destroy any volunteer GM sugarcane, until no volunteers are observed for a continuous six month period. Sugarcane seed has little dormancy (Simpson 1990), and stem pieces capable of giving rise to new plants are not expected to remain viable over long periods in the field, as they are degraded rapidly by soil micro-organisms under tropical conditions. Normally, stem cuttings need to be dipped in fungicide to enable them to survive until germination (FAO 2001; FAO 2004). The proposed time period is therefore considered appropriate for minimising the persistence of the GMOs in the environment, potential for dispersal (Risk scenario 2) and gene flow (Risk scenarios 3 - 6). Therefore, the requirement to monitor the field release sites monthly for at least 12 months and until no volunteers are observed for a continuous six month period, is included as a condition. Furthermore, no plants may be grown in an area of land following its cleaning unless the Regulator has issued a sign-off or the plants are approved by the Regulator.

209. The applicant does not propose using any of the plant material for human or animal consumption. FSANZ conducts mandatory premarket assessments of GM products in human foods. As the GM sugarcane has not been assessed by FSANZ, a condition in the licence prohibits material from the trial from being used for human food or animal feed.

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

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

- limit the release to a total area of 30 ha at seven SRA stations between November 2015 and November 2021
- locate the field trial sites at least 50 m away from natural waterways
- surround the field trial locations by one guard row of non-GM sugarcane and a further isolation zone of at least 6 m
- separate GM sugarcane material from non-GM material when propagating seedlings or setts on seedling benches, and clearly identifying GM material
- separate GM from non-GM sugarcane in crossing facilities by at least 1 m (glasshouses, pot holding areas, photoperiod facility and crossing shed)

- monitor GM sugarcane in photoperiod facilities for spikelet opening and enclose inflorescences in pollen lanterns prior to spikelet opening and destroy any open spikelets not enclosed in pollen lanterns
- harvest and process the GM sugarcane separately from any other sugarcane
- carry out analysis of plant materials at the SRA stations, in PC2 laboratories or in other approved facilities
- destroy all plant materials not required for experimentation or propagation (through methods such as mulching, burning and herbicide treatment)
- after cleaning of sites, monitor for and destroy any GM sugarcane that may grow for at least 12 months, and until no volunteers are observed for a continuous six month period
- transport the GM plant materials in accordance with Regulator's transportation guidelines
- not allow the GM plant material or products to be used for human food or animal feed.

3.2 Other risk management considerations

211. 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 structures
- a requirement that the applicant allows access to the trial sites and other places for the purpose of monitoring or auditing.

3.2.1 Applicant suitability

212. 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 (both individuals and the body corporate)
- 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.

213. On the basis of information submitted by the applicant and records held by the OGTR, the Regulator considers Sugar Research Australia Ltd suitable to hold a licence.

214. The licence includes a requirement for the licence holder to inform the Regulator of any circumstances that would affect their suitability.

215. In addition, any applicant organisation must have access to a properly constituted Institutional Biosafety Committee and be an accredited organisation under the Act.

3.2.2 Contingency plan

216. SRA is 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 sugarcane outside of the permitted areas.

217. SRA is also required to provide a method to the Regulator for the reliable detection of the presence of the GMOs or the introduced genetic materials in a recipient organism. This instrument would be required before conducting any of the licenced dealings with the GMOs.

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

218. The persons covered by the licence are 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, SRA is also required to provide a list of people and organisations who will be covered by the licence, or the function or position where names are not known at the time.

3.2.4 Reporting requirements

219. The licence obliges 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 trial
- any contraventions of the licence by persons covered by the licence
- any unintended effects of the trial.

220. A number of written notices would also be required under the licence that would assist the Regulator in designing and implementing a monitoring program for all licensed dealings. The notices would 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

221. 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 site.

222. 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.

223. 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 health and safety of people or the environment could result.

Section 4 Issues to be addressed for future releases

224. Additional information has been identified that may be required to assess an application for a large scale or commercial release of these GM sugarcane lines, or to justify a reduction in containment conditions. This includes:

- additional molecular and biochemical characterisation of the GM sugarcane lines, particularly with respect to production of potential toxins or allergens
- additional phenotypic characterisation of the GM sugarcane lines, particularly with respect to traits that may contribute to weediness.

Section 5 Conclusions of the RARMP

225. The risk assessment concludes that this proposed limited and controlled release of GM sugarcane poses negligible risks to the health and safety of people or the environment as a result of gene technology, and that these negligible risks do not require specific risk treatment measures.

226. However, conditions have been imposed to limit the release to the proposed size, locations 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.

References

Aitken, K., Li, J., Wang, L., Qing, C., Fan, Y., Jackson, P. (2007). Characterization of intergeneric hybrids of *Erianthus rockii* and *Saccharum* using molecular markers. *Genetic Resources and Crop Evolution* **54**: 1395-1405

Bakker, M. (1999). *Sugarcane cultivation and management.* Kluwer Academic/Plenum Publishers New York.

Bammer, G., Smithson, M. (2008). *Uncertainty and risk: Multidisciplinary perspectives.*, Bammer, G., Smithson, M. (eds). Earthscan London.

Berding, N. (1981). Improved flowering and pollen fertility in sugarcane under increased night temperatures. *Crop Science* **21**: 863-867

Bonnett, G. D, Berding, N, Morgan, T., Fitzgerald, P. (2007) Implementation of genetically modified sugarcane - the need for a better understanding of sexual reproduction. In "*Proceedings of the Australian Society of Sugarcane Technologists"*, **29** pp. 258-266.

Bonnett, G. D, Olivares-Villegas, J. J., Berding, N, Morgan, T. (2010) Sugarcane sexual reproduction in a commercial environment: research to underpin regulatory decisions for genetically modified sugarcane. In "*Proceedings of the Australian Society of Sugarcane Technologists"*, **32** pp. 1-9.

Bonnett, G.D., Nowak, E., Olivares-Villegas, J.J., Berding, N., Morgan, T., Aitken, K.S. (2008). Identifying the risk of transgene escape from sugarcane crops to related species, with particular reference to *Saccharum spontaneum* in Australia. *Tropical Plant Biology* **1**: 58-71

Bower, R., Elliott, A.R., Potier, B.A.M., Birch, R.G. (1996). High-efficiency, microprojectile-mediated cotransformation of sugarcane, using visible or selectable markers. *Molecular Breeding* **2**: 239-249

Breaux, R.D., Miller, J.D. (1987). Seed handling, germination and seedling propagation. Chapter 10. In: DJ Heinz, ed. *Sugarcane improvement through breeding*, Volume 11. Elsevier Amsterdam. pp 385-407.

Brown, J.S., Schnell, R.J., Power, EJ., Douglas, S.L., Kuhn, D.N. (2007). Analysis of clonal germplasm from five *Saccharum* species: *S. barberi , S. robustum, S. officinarum, S. sinense* and *S. spontaneum* . A study of inter- and intra species relationships using microsatellite markers. *Genetic Resources and Crop Evolution* **54**: 627-648

Butterfield, M.K., Irvine, J.E., Garza, M.V., Mirkov, T.E. (2002). Inheritance and segregation of virus and herbicide resistance transgenes in sugarcane. *Theoretical and Applied Genetics* **104**: 797-803

Chakraborty, P., Gupta-Bhattacharya, S., Chowdhury, I., Majumdar, M.R., Chanda, S. (2001). Differences in concentrations of allergenic pollens and spores at different heights on an agricultural farm in West Bengal, India. *Annals of Agricultural and Environmental Medicine* **8**: 123-130

Christensen, A.H., Sharrock, R.A., Quail, P.H. (1992). Maize polyubiquitin genes: structure, thermal perturbation of expression and transcript splicing, and promoter activity following transfer to protoplasts by electroporation. *Plant Molecular Biology* **18**: 675-689

Clark, A.J. and Brinkley, T. (2001). Risk management: for climate, agriculture and policy. Commonwealth of Australia Canberra.

Crawley, M.J., Brown, S.L., Hails, R.S., Kohn, D.D., Rees, M. (2001). Transgenic crops in natural habitats. *Nature* **409**: 682-683

D'Hont, A., Grivet, L., Feldmann, P., Rao, S., Berding, N., Glaszmann, J.C. (1996). Characterisation of the double genome structure of modern sugarcane cultivars (*Saccharum spp*.) by molecular cytogenetics. *Molecular and General Genetics* **250**: 405-413

D'Hont, A., Ison, D., Alix, K., Roux, C., Glaszmann, J.C. (1998). Determination of basic chromosome numbers in the genus *Saccharum* by physical mapping of ribosomal RNA genes. *Genome* **41**: 221-225

de Virgilio, M., Lombardi, A., Caliandro, R., Fabbrini, M.S. (2010). Ribosome-inactivating proteins: from plant defense to tumor attack. *Toxin* **2**: 2699-2737

Delaney, B., Astwood, J.D., Cunny, H., Conn, R.E., Herouet-Guicheney, C., MacIntosh, S., Meyer, L.S., Privalle, L., Gao, Y., Mattsson, J., Levine, M., ILSI International Food Biotechnology Committee Task Force on Protein Safety (2008). Evaluation of protein safety in the context of agricultural biotechnology. *Food and Chemical Toxicology* **46**: S71-S97

EPA (1996). Plant pesticide inert ingredient CP4 Enolpyruvylshikimate-3-D and the genetic material necessary for its production in all plants. Report No. 61, US Environmental Protection Agency (EPA) available online at Federalregister

FAO (2001). *Safe movement of sugarcane germplasm.*, Autrey, L., Frison, E., Putter, C. (eds). The International Board for Plant Genetic Resources FAO in collaboration with The International Society of Sugarcane Technologists

FAO (2004). *Saccharum officinarum*. Available online at FAO.

Glover, J. (2002). Gene flow study: Implications for the release of genetically modified crops in Australia. Bureau of Rural Sciences, Australia, available online at Bureau of Rural Sciences

Grassl, C.O. (1980). Breeding *Andropoganeae* at the generic level for biomass. *Sugarcane Breeders' Newsletter* **43**: 41-57

Hammond, B., Kough, J., Herouet-Guicheney, C., Jez, J.M., ILSI International Food Biotechnology Committe Task Force on the Use of Mammalian Toxicology Studies in the Safety Assessment of GM Foods (2013). Toxicological evaluation of proteins introduced into food crops. *Critical review in Toxicology* **43 (S2)**: 25-42

Hansom, S., Bower, R., Zhang, L., Potier, B., Elliott, A., Basnayake, S., Cordeiro, G., Hogarth, D. M., Cox, M., Berding, N., Birch, R. G. (1999) Regulation of transgene expression in sugarcane. In "*Proceedings of the XXIII International Society of Sugar Cane Technologists"*, STAI, New Delhi, India. pp. 278-290.

Haslberger, A.G. (2003). GM food: the risk-assessment of immune hypersensitivity reactions covers more than allergenicity. *Journal of Food, Agriculture and Environment* **1**: 42-45

Hauser, M., Egger, M., Wallner, M., Wopfner, N., Schmidt, G., Ferreira, F. (2008). Molecular properties of plant food allergens: a current classification into protein families. *The Open Immunology Journal* **1**: 1-12

Hayes, K.R. (2004). Ecological implications of GMOs: robust methodologies for ecological risk assessment. Best practice and current practice in ecological risk assessment for genetically modified organisms.1-73. Department of Environment. Accessed on 11 August 2004.

Henkel, J.S., Baldwin, M.R., Barbieri, J.T. (2010). Toxins from bacteria. *Experientia Supplementum Volume* **100**: 1-29

Herman, R.A., Ladics, G.S. (2011). Endogenous allergen upregulation: transgenic vs. tradtionally bred crops. *Food and Chemical Toxicology* **49**: 2667-2669

Hogarth, D.M., Allsopp, P.G. (2000). *Manual of Canegrowing.* Bureau of Sugar Experiment Stations Brisbane.

Irvine, J.E. (1999). *Saccharum* species as horticultural classes. *TAG Theoretical and Applied Genetics* **98**: 186-194

Joyce, P., Hermann, S., O'Connel, A., Dinh, Q., Shumbe, L., Lakshamanan, P. (2013). Field performance of transgenic sugarcane produced using Agrobacterium and biolistics methods. *Plant BiotechnologyJournal* **pp1-14 doi: 10.1111/pbi.12148**:

Karalliedde, L. (1995). Animal toxins. *British Journal of Anaesthesia* **74**: 319-327

Keese, P. (2008). Risks from GMOs due to horizontal gene transfer. *Environmental Biosafety Research* **7**: 123-149

Keese, P.K., Robold, A.V., Myers, R.C., Weisman, S., Smith, J. (2013). Applying a weed risk assessment approach to GM crops. *Transgenic Research*

Moore, P.H. (1976). Studies on sugarcane pollen. II. Pollen storage. *Phyton, Argentina* **34**: 71-80

Moore, P.H., Nuss, K.J. (1987). Flowering and flower synchronization. Chapter 7. In: DJ Heinz, ed. *Sugarcane improvement through breeding*. Elsevier Amsterdam. pp 273-311.

Nair, N., Selvi, A., Sreenivasan, T., Pushpalatha, K., Mary, S. (2005). Molecular diversity among *Saccharum, Erianthus, Sorghum, Zea* and their hybrids. *Sugar Tech* **7**: 55-59

Nair, N.V. (1999). Production and cyto-morphological analysis of intergeneric hybrids of *Sorghum* x *Saccharum*. *Euphytica* **108**: 187-191

OGTR (2011). The Biology of the *Saccharum spp.* (sugarcane). The Office of the Gene Technology Regulator, Canberra, Australia, available online at OGTR

OGTR (2013). *Risk Analysis Framework.* The Office of the Gene Technology Regulator, Canberra, Australia, available online at OGTR

Perlak, F.J., Deaton, R.W., Armstrong, T.A., Fuchs, R.L., Sims, S.R., Greenplate, J.T., Fischhoff, D.A. (1990). Insect resistant cotton plants. *Bio/Technology* **8**: 939-943

Pierre, J.S., Rae, A.L., Bonnett, G.D. (2014). Abiotic Limits for Germination of Sugarcane Seed in Relation to Environmental Spread. *Tropical Plant Biol* 1-11

Piperidis, G., Christopher, M.J., Carroll, B.J., Berding, N., D'Hont, A. (2000). Molecular contribution to selection of intergeneric hybrids between sugarcane and the wild species *Erianthus arundinaceus*. *Genome* **43**: 1033-1037

Powell, R., Oliveras-Villegas, J. J., Berding, N., Schnmidt, S., Bonnett, G. (2008) The effect of temperature on germination of *Saccharum* seed (*poster paper*). In "*Proceedings of the Australian Society of Sugar Cane Technologists",* **30** pp. 588.

Radauer, C., Breiteneder, H. (2007). Evolutionary biology of plant food allergens. *Journal of Allergy and Clinical Immunology* **120**: 518-525

Rae, A.L., Perroux, J.M., Grof, C.P.L. (2005). Sucrose partitioning between vascular bundles and storage parenchyma in the sugarcane stem: a potential role for the ShSUT1 sucrose transporter. *Planta* **220**: 817-825

Rao, J.T., Alexander, M.P., Kandaswami, P.A. (1967). Intergeneric hybridisation studies between *Saccharum* (sugarcane) and *Bambusa* (bamboo). *Journal of the Indian Botanical Society* **46**: 199-208

Rao, P. S. (1980) Fertility, seed storage and seed viability in sugarcane. In "*Proceedings of the International Society of Sugar Cane Technologists",* **17** pp. 1236-1240.

Reiting, R., Broll, H., Waiblinger, H.-U., Grohmann, L. (2007). Collaborative study of a T-nos real-time PCR method for screening of genetically modified organisms in food products. *Journal of Consumer Protection and Food Safety* **2**: 116-121

Salcedo, G., Sanchez-Monge, R., Diaz-Perales, A., Pacios, L.F. (2008). Review. Plant food allergens: peach non-specific lipid transfer protein Pru p 3 as a model. *Spanish Journal of Agricultural Research* **6**: 30-37

Sebastian, S.A., Fader, G.M., Ulrich, J.F., Forney, D.R., Chaleff, R.S. (1989). Semidominant Soybean Mutation for Resistance to Sulfonylurea Herbicides. *Crop Science* **29**: 1403-1408

Simpson, G.M. (1990). *Seed dormancy in grasses.* Cambridge University Press

Skinner, J.C. (1959). Controlled pollination of sugarcane. Report No. 1, Bureau of Sugar Experiment Stations: technical communications

Standards Australia New Zealand, CRC for Australian Weed Management (2006). *HB 294:2006 National Post-Border Weed Risk Management Protocol.* Standards Australia; Standards New Zealand; pp 1-68.

Steiner, H.Y., Halpin, C., Jez, J.M., Kough, J., Parrott, W., Underhill, L., Weber, N., Hannah, L.C. (2013). Evaluating the potential for adverse interactions within genetically engineered breeding stacks. *Plant Physiology* **161**: 1587-1594

Stirpe, F. (2005). Ribosome-inactivating proteins. Chapter 2. In: RG Wiley, DA Lappi, eds. *Molecular Neurosurgery with Targeted Toxins*. Humana Press pp 9-29.

Sugarcane Researchers (2008). The production of seedlings in commercial sugarcane growing regions in Queensland, Australia.

Umbeck, P., Swain, W., Yang, N.-S. (1989). Inheritance and expression of genes for kanamycin and chloramphenicol resistance in transgenic cotton plants. *Crop Science* **29**: 196-201

Venkatraman, R.S.T.S. (1922). Germination and preservation of sugarcane pollen. *Agricultural Journal of India* **17**: 127-132

Vickers, J.E., Grof, C.P.L., Bonnett, G.D., Jackson, P.A., Morgan, T.E. (2005). Effects of tissue culture, biolistic transformation, and introduction of PPO and SPS gene constructs on performance of sugarcane clones in the field. *Australian Journal of Agricultural Research* **56**: 57-68

Waines, J.G., Hegde, S.G. (2003). Intraspecific gene flow in bread wheat as affected by reproductive biology and pollination ecology of wheat flowers. *Crop Science* **43**: 451-463

Wang, X.-H., Yang, Q.-H., Li, F.-S., He, L.-L., He, S.-C. (2009). Molecular identification of *Saccharum* spp. x *Erianthus fulvus* hybrids using sequence-characterized amplified region markers. *Crop Science* **49**: 864-870

Weber, N., Halpin, C., Hannah, L.C., Jez, J.M., Kough, J., Parrott, W. (2012). Editor's choice: crop genome plasticity and its relevance to food and feed safety of genetically engineered breeding stacks. *Plant Physiology* **160**: 1842-1853

Appendix A Summary of submissions from prescribed experts, agencies and authorities[9]

Abbreviations: C: control measures; **Ch:** Chapter; **GF:** Gene flow; **GM:** Genetically Modified; **I**: inconsistencies; **LC:** Licence conditions; **N:** Neutral; **RARMP**: Risk Assessment and Risk Management Plan; **Y:** Support

Sub.No:	Summary of issues raised	Comment
1	Notes that the licence prohibits the use of material from the trials for human or animal consumption and has no further comments on the RARMP.	Noted.
2	No concerns about the safety of the proposed dealings. Little GM material is likely to enter the food chain as the food product is so purified.	No material will enter the food supply from this field trial.
3	It is not clear why the applicant proposes to use non-PC2 growth cabinets or glasshouses for germination of the GM seed.	The Regulator evaluates the risk of the proposed dealings within the scope of the Act. In the context of this field trial, risks from the use of non-PC2 facilities were considered to be negligible..
	What is the basis for the requirement for post-trial monitoring for at least 12 months and until the site is free of volunteers for a continuous 6 months period?	The length of post-harvest monitoring and requirement for volunteer free time depends on the plant species in question. Based on the relevant information available for sugarcane biology, the Regulator has imposed these controls for GM sugarcane in previous releases and has received no reports that these measures have been ineffective. Therefore, the post-harvest monitoring requirements were considered sufficient as a control.
	Clarity on 6m isolation zone and 10m from commercial sugarcane.	This issue was addressed in Chapter 3 of the RARMP and the licence by requiring a minimum of 6 m separation from non-GM sugarcane, for both trials and commercial crops.
4	Is satisfied with the conclusions of the draft RARMP and has no technical comments.	Noted.
5	The licence application was circulated to a number of agencies and no comments were received which raised concerns regarding the potential for harm to humans or the environment. Accordingly, has no objection to the granting of the licence.	Noted.
6	Agrees with the overall conclusions of the RARMP	Noted.
	Agrees that all plausible risk scenarios have been identified	Noted.
	Generally agrees with the limits and controls proposed in the RARMP	Noted.

[9] Prescribed agencies include GTTAC, State and Territory Governments, relevant local governments, Australian Government agencies and the Minister for the Environment.

Sub.No:	Summary of issues raised	Comment
	Recommends consideration of adequacy of the temporal and physical separation of flowering GM and non-GM sugarcane at Meringa crossing facility	Further information was acquired from the applicant on pollen lanterns. Pollen lanterns are enclosed on all sides and the top, and the bottom part is tied around the stalk of the plant. Additional text was added in Chapter 3, Section 3.1.1 to reflect that pollen escape from the lanterns is highly unlikely. Therefore, additional temporal or spatial measures are not considered necessary.
	Clarify and ensure consistency in the RARMP of descriptions of hot water treatments	The text in Chapter 1, Section 6.2 has been amended to clearly define the locations of the hot water treatment facilities and their operational procedure.
	Clarify the requirements and rationale for separation between GM and commercial non-GM sugarcane in the field trial	The applicant had proposed 6 m isolation from non-GM trials and 10 m isolation from GM sugarcane crop, but the licence only requires at least 6 m isolation distance from any non-GM sugarcane. Text has been amended in Chapter 3, Section 3.1.1 to explain the rationale for inclusion of 6 m isolation zone as an effective measure to reduce the likelihood of inadvertent mixing of cane in adjacent plots.
	Clarify the proposed number and locations of trial sites.	Text has been amended in Chapter 1, Section 3.1 to include the newly proposed site by the applicant and its location.
	Clarify the post-harvest requirements that will apply for trial sites	Further text has been added in Chapter 3, Section 3.1.1 that no plants may be grown in an area of land following its cleaning unless the Regulator has issued a sign-off or the plants are plants agreed to in writing by the Regulator.
7	Is supportive of the application as the consultation RARMP indicates that the proposed release poses negligible risks to people or the environment.	Noted.

Appendix B Summary of submissions from the public

The Regulator received 2 submissions from the public on the consultation RARMP. The issues raised in these submissions are summarised in the table below. All issues raised in the submissions 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.

Abbreviations:

View (general tone): **n** = neutral; **x** = do not support; **y** = support.

Issues raised: **C**: containment; **H**: Health; **LC**: Licence conditions; **M**: Marketing; **R**: Residues; **S**: Segregation

Other abbreviations: **APVMA**: Australian Pesticides and Veterinary Medicines Authority; **Ch**: Chapter; **FSANZ**: Food Standards Australia New Zealand; **GM**: Genetically modified; **GMO**: Genetically modified organism.

Sub. No:	Issue	Summary of issues raised	Comment
1	R	Is concerned about the residue of the herbicides in potential food stuffs when the crops are harvested.	As this application is for a limited and controlled release (field trial), strict licence conditions have been imposed to prohibit GM plant material or products being used for human food or animal feed. If this GM sugarcane was approved for commercial production in the future, the Australian Pesticides and Veterinary Medicines Authority (APVMA) would assess the herbicide for use on the GM sugarcane and set maximum residue levels (MRLs). MRLs are then listed in the Food Standards Code.
2	H	States that recent studies of GM food indicate there is sufficient cause for concern to call for a freeze on approvals of the release of GMOs to the environment. Calls for further scientific testing, including clinical trials to quantify the risk of identified side-effects to human health and the environment. Lists some websites and articles that raise health concerns.	As this application is for a limited and controlled release (field trial), strict licence conditions have been imposed to prohibit GM plant material or products being used for human food or animal feed. FSANZ is responsible for human food safety assessment.
	C	States that, until analysis of known escapes of unapproved GM crops is completed, the causes identified and preventive actions specified and implemented in the RARMP, risks remain unquantified and negligible risk cannot be assumed. Claims that GMO escapes include GM wheat in Oregan, US, GM rice in Chinese supermarkets, and GM canola in Tasmania	Strict licence conditions have been imposed on this field trial to restrict the spread and persistence of the GMOs and their genetic material in the environment. Based on current information and experience the control measures imposed are considered to be effective for restricting spread of the GM sugarcane.

	S, M	States that escapes of GM crops have occurred and continue to occur. It is evident from the Marsh v Baxter court case and subsequent response from the GM authorities that GM escapes are to be expected as a normal part of agriculture which is strongly disagreed.	The GM canola on which the Marsh v Baxter court case is based was assessed by the Regulator to be as safe as conventional canola and therefore it was approved for commercial growing without a need for limits and controls that would be normally be applied to field trials. Marketing and trade issues, including matters relating to segregation and coexistence of different farming systems, are the responsibility of the States and industry, not the Regulator. The RARMP concluded that this limited and controlled field trial poses negligible risks to the health and safety of people and the environment.