GENE TECHNOLOGY TECHNICAL ADVISORY COMMITTEE

COMMUNIQUE
No. 9

This is the ninth communique of the Gene Technology Technical Advisory Committee (GTTAC). It covers matters considered at the fourteenth and fifteenth meetings of GTTAC held on 22 May 2003 and 24 July 2003 respectively, as well as matters considered by GTTAC out-of-session in the period from 10 April 2003 to 24 July 2003.

GTTAC is a statutory advisory committee to the Gene Technology Regulator (the Regulator) and the Gene Technology Ministerial Council. All committee members and expert advisers hold office on a part-time basis.

The Regulator receives input from GTTAC on applications for licences to conduct dealings with genetically modified organisms (GMOs), as well as comments on the Risk Assessment and Risk Management Plan (RARMP) that is prepared for each of these applications.

The purpose of this Communique is to provide a brief overview of the applications and RARMPs considered by GTTAC and the advice the Committee has provided to the Regulator with regard to those applications and RARMPs.

The Communique also provides an overview of any other major issues discussed by GTTAC.

Dealings Not Involving the Intentional Release of Genetically Modified Organisms

Dealings Not Involving the Intentional Release of GMOs (DNIRs) are dealings that are usually undertaken within a certified facility (so that the organism is physically contained) and where the personnel involved in the dealing have been assessed as having adequate training and experience for the task. These are typically laboratory-based projects.

Applications and RARMPs for the following DNIRs were assessed:

Application Number and Title	Project Description	GTTAC Comments
DNIR 163/2003 The development of glycine mosaic comovirus (GMV) as a vector for heterologous gene expression in plants.	The aim of this dealing is to develop GMV based vectors to express genes in plants.	GTTAC agreed that the risk assessment identified all the risks associated with the proposed dealings and that the measures proposed in the risk management plan are adequate to deal with the identified risks.
DNIR 178/2003 Functional and molecular analysis of defects of the mitochondrial electron transport chain.	The aim of this dealing is to study human cells that have a metabolic defect of the mitochondrial energy production pathways to determine on which chromosome the disease causing gene is located. These cells will be transformed with a gene to immortalise them prior to study.	GTTAC agreed that the risk assessment identified all the risks associated with the proposed dealings and that the measures proposed in the risk management plan are adequate to deal with the identified risks. GTTAC advised that laboratory guidelines must be followed and that the use of sharp instruments should be avoided where possible. GTTAC requested that the applicant should be asked why they have chosen this method of immortalising cells.

Application Number and Title	Project Description	GTTAC Comments
DNIR 186/2003 Molecular virology of HIV-1 and SIV.	The aim of this dealing is to analyse the structure/function relationship between wild type and mutant viral genes and elements in HIV-1 and SIV to understand their role in viral gene expression, replication, particle assembly and pathogenesis.	GTTAC agreed that the risk assessment identified all the risks associated with the proposed dealings and that the measures proposed in the risk management plan are adequate to deal with the identified risks. GTTAC advised that laboratory guidelines must be followed and that the use of sharp instruments should be avoided where possible. GTTAC also advised that laboratory workers should be warned of an increased risk to people who are immunosuppressed.
DNIR 187/2003 Viral Assembly of Moloney murine leukaemia virus (MoMLV), Mason-Pfizer monkey virus (M-PMV), human foamy virus (HFV) and avian sarcoma / eukosis virus (ASLV).	The aim of this dealing is to understand the role of various MoMLV, M-PMV, HFV or ASLV genes by transfecting mammalian cells with mutated or wild type clones of these retroviruses.	As for DNIR 186/2003.
DNIR 188/2003 Pathogenesis of macrophage-tropic HIV-1.	The aim of this dealing is to examine the ability of HIV-1 strains to induce cell killing by transfecting mammalian cell lines with HIV-1 DNA.	GTTAC agreed that the risk assessment identified all the risks associated with the proposed dealings and that the measures proposed in the risk management plan are adequate to deal with the identified risks. GTTAC advised that laboratory guidelines must be followed and that the use of sharp instruments should be avoided where possible.

Application Number and Title	Project Description	GTTAC Comments
DNIR 194/2003 Evaluation of cellular immunological function with recombinant virus.	The aim of this dealing is to evaluate if a treatment can augment or sustain HIV positive patients' cellular immune response to HIV and help further define the mechanisms involved.	As for DNIR 188/2003.
DNIR 203/2003 Construction and use of herpes simplex virus mutants	The aim of this dealing is to determine how minor changes to the HSV viral protein gB will alter the response of cytotoxic T lymphocytes (immune cells) by infecting mice with HSV-1 gB mutants.	As for DNIR 188/2003. GTTAC suggested that the applicants consider using a cannula to deliver GMOs to mice.
DNIR 207/2003 Molecular aspects of plant-pathogen interactions - Thielaviopsis	The aim of this dealing is to identify genes in *T. basicola* (a pathogen causing black root disease in plants) which may be involved in virulence.	As for DNIR 163/2003. GTTAC advised that extra care should be taken to ensure that waste and equipment potentially contaminated with fungal spores is successfully decontaminated.
DNIR 208/2003 Recombinant murine cytomegalovirus (MCMV) encoding hepatitis C virus (HCV) proteins.	The aim of this dealing is to insert genes encoding HCV proteins into MCMV. The recombinant MCMV will be used as a delivery system to express HCV proteins in murine liver.	As for DNIR 188/2003.
DNIR 216/2003 Development of *Trichoderma harzianum* for biocontrol of plant pathogens	The aim of this dealing is to improve the biocontrol efficacy of *Trichoderma harzianum* by inserting the chitinase gene into its genome.	As for DNIR 163/2003.
DNIR 217/2003 Structure/activity studies of novel toxins from native venomous organisms (jellyfish)	The aim of this dealing is to produce milligram quantities of toxic jellyfish venom proteins by expressing them in a bacterial host. The structure and activity of these proteins will be assessed.	As for DNIR 163/2003.

Application Number and Title	Project Description	GTTAC Comments
DNIR 218/2003 Generation of recombinant canine herpesviruses (CHV)	The aim of this dealing is to develop recombinant CHVs that express heterologous antigens derived from genomic, viral or bacterial genes. These viruses will be used as experimental vaccines to immunise foxes, dogs and ferrets against infectious diseases and/or to reduce their fertility.	As for DNIR 163/2003.
DNIR 219/2003 Recombinant mycobacteria as new anti-tuberculosis vaccines	The aim of this dealing is to express mycobacterium tuberculosis antigens in the vaccine strain *mycobacterium bovis* bacillus Calmette-Geurin. The recombinant bacteria will be tested as a vaccine against tuberculosis.	As for DNIR 163/2003.
DNIR 222/2003 Expression of virus encoded antigens using vaccinia expression systems	The aim of this dealing is to construct recombinant vaccinia viruses containing genes encoding Epstein-Barr virus (EBV) and cytomegalovirus (CMV) antigens. The viruses will be used to infect cell lines, which will be used as targets in T cell assays.	As for DNIR 186/2003. GTTAC advised that there is also a risk to people undertaking the dealing from exposure to aerosols and splashes containing GM vaccinia viruses. GTTAC recommended that these people be vaccinated against vaccinia virus.
DNIR 225/2003 Mouse models of colorectal cancer using a TVA based retroviral gene transfer system	The aim of this dealing is to investigate the role of various genes in colorectal cancer by transferring candidate oncogenes and a tumour suppressor gene directly into the intestinal epithelium of mice using an avian retrovirus.	As for DNIR 188/2003.
DNIR 227/2003 Structure/activity studies of novel toxins from native venomous organisms (brown snake)	The aim of this dealing is to introduce genes encoding brown snake venom proteins into bacterial and/or eukaryotic hosts to produce milligram quantities of these proteins for biophysical and functional studies.	As for DNIR 163/2003.

Dealings Involving the Intentional Release of Genetically Modified Organisms

Dealings Involving the Intentional Release of GMOs (DIRs) are dealings that result in the introduction of a GMO into the environment. DIRs may involve a limited and controlled release (field trial) where measures are imposed in the licence conditions to control the movement of the GMO or its genetic material, or a general (commercial) release of a GMO where minimal oversight conditions have been required, to date.

RARMPs for licence applications for DIRs are released for public comment as part of the consultation process for these applications. Information on how to obtain copies of applications and RARMPs for DIRs is provided at the end of this document.

Advice on Applications

GTTAC considered the following applications concerning the release of transgenic cotton in Australia and provided advice on issues to be considered in the preparation of the associated RARMPs.

- **Field trial of genetically modified cotton (*Gossypium hirsutum*) expressing an insecticidal gene (*vip3A*) (DIR 034/2003)**

  The OGTR has received a licence application from Syngenta Seeds Pty Ltd (Syngenta) for the limited and controlled release of genetically modified (GM) insecticidal cotton into the environment. Syngenta proposes to conduct trials on 30 sites covering a total area of 10 hectares, over three years, in the cotton growing regions of Queensland (QLD), New South Wales (NSW) and Western Australia (WA).

  The main aim of the proposed release is to assess the agronomic performance and efficacy of the insecticidal activity of the new lines in all the major cotton growing areas of Australia.

  The GM cotton proposed for release is a backcross of an insecticidal cotton, described by Syngenta as COT102, into three elite Australian cotton cultivars. Limited and controlled field trials of COT102 have been previously approved in Australia under PR-151, DIR 017/2002 and DIR 025/2002.

  The GM cotton contains an insecticidal gene (*vip3A*), derived from a common soil bacterium which encodes an insecticidal protein (VIP3A) that is toxic to lepidopteran caterpillar pests of cotton. It also contains a bacterial gene *hph*, conferring resistance to hygromycin, an antibiotic that was used as a selectable marker in the initial laboratory stages of developing the GM cotton.

  The insecticidal VIP3A protein produced by this GM cotton is different from insecticidal (Cry) proteins that are present in most other types of GM cotton that are currently being trialled or grown commercially in Australia.

  None of the cotton plants from the proposed release, or their by-products, will be used for human food or animal feed. The applicant proposes to sell the lint for use in clothing and upholstery. Lint does not contain genetic material or protein.

  Details of the plasmid map, including the gene construct containing the insecticidal *vip3A* gene, and the regulatory sequences (promoters) have been declared Confidential Commercial Information (CCI) under section 185 of the Act. However, this information

has been made available to GTTAC and other prescribed expert authorities that are being consulted on the preparation of the RARMP.

GTTAC discussed this application and advised the Regulator that the following issues should be considered in the preparation of the RARMP:

- The risks posed by DIR 034/2003 are similar to those posed by previous cotton applications;
- The advice provided in relation to previously assessed GM cottons (DIR 017/2002 and DIR 025/2002) should be considered in the preparation of the RARMP for DIR 034/2003; and
- At the completion of the three years of field trials, the applicant should be requested to provide data on the levels of expression of the introduced proteins under Australian field conditions.

- **Breeding and pre-commercial evaluation of transgenic cotton expressing an insecticidal protein gene and a herbicide tolerance gene (DIR 036/2003)**

The OGTR has received an application from the Commonwealth Scientific and Industrial Research Organisation (CSIRO) for a licence for the limited and controlled release of GM cotton into the environment. CSIRO proposes to release the transgenic cotton on 16 sites within existing cotton growing regions in QLD and NSW covering a total area of less than 45 hectares per growing season for 3 seasons.

Three insecticidal GM cottons are proposed for release which express vegetative insecticidal protein (VIP) from the *vip* gene. This protein is different to the other insecticidal proteins present in the GM insecticidal cottons being trialed or grown commercially in Australia. The applicant expects the new GM cottons to provide alternative options to manage the risk of resistance development in insect populations. The COT102 cotton line contains insecticidal and antibiotic resistance genes. COT102 was previously approved under the voluntary system as PR-151 and more recently under the current regulatory system in licences DIR 017/2002 and DIR 025/2002. The COT200 series cotton lines express only the insect resistance gene under a different promoter to that of COT102. The COT200 series lines do not contain the antibiotic resistance gene. The third type of insecticidal GM cotton proposed for release will be derived by conventional crosses between insecticidal cotton (COT102) and another GM cotton tolerant to the herbicide Liberty® (glufosinate ammonium), generated previously as part of a separate dealing (DIR 015/2002). It is expected that addition of the Liberty® cotton trait will allow more effective weed control in insect resistant cotton crops by allowing the crop to be sprayed with glufosinate-ammonium to kill problem weeds without damaging the crop itself.

The proposed trial is part of an ongoing breeding program to develop lines suitable for commercial development. The main aim of the proposed release is to evaluate the agronomic performance of cotton lines modified to express a new insecticidal protein that is toxic to lepidopteran caterpillar pests of cotton. The release would also allow the assessment of the efficacy of the insecticidal protein, the combining of the insecticidal and herbicide tolerance traits by crossing different GM lines (containing insecticidal and herbicide tolerant traits), and production of seed for future releases, subject to future approvals.

None of the cotton plants from the release, or their by-products, would be used for animal feed or human food. However, the applicant proposes to sell lint from the release. Lint does not contain genetic material or protein.

Details of the gene construct including the plasmid map and regulatory sequences for the COT102 event have previously been declared as CCI. CSIRO has sought approval for details of the gene construct including the plasmid map and regulatory sequences of the COT200 series lines to be declared as CCI. However, this information has been made available to GTTAC and other prescribed expert authorities that are being consulted on the preparation of the RARMP.

GTTAC discussed this application and advised the Regulator that the following issues should be considered in the preparation of the RARMP:

- The risks posed by DIR 036/2003 are similar to those posed by previous cotton applications such as DIR 015/2002, DIR 017/2002 and DIR 025/2002;
- Advice provided in relation to previously assessed GM cottons should be considered in the preparation of the RARMP for DIR 036/2003; and
- The applicant should be requested to provide data on the levels of expression of the introduced proteins under Australian field conditions at the completion of the three year field trial.

- **Field trial for breeding and pre-commercial evaluation of GM cotton expressing tolerance to the herbicide glufosinate ammonium (DIR 038/2003)**

The OGTR has received an application from CSIRO for a licence for the limited and controlled release of GM cotton into the environment. CSIRO is proposing to release this transgenic cotton on 16 sites, in NSW and QLD, over a total area of 45 hectares per year over three growing seasons.

The aim of the proposed release is breeding and pre-commercial field evaluation of GM herbicide tolerant Liberty® cotton. The release would also be used for demonstration purposes.

Liberty® cotton is tolerant to the herbicide glufosinate ammonium (also called phosphinothricin), the active constituent of herbicides Basta® and Liberty® (hence the name Liberty® cotton). It is expected that use of Liberty® cotton plants will allow more effective weed control in cotton crops by allowing the crop to be sprayed with glufosinate ammonium to kill problem weeds without damaging the crop itself.

None of the cotton plants from the release, or their by-products, would be used for animal feed or human food. However, the applicant is proposing to sell lint from the conventional cotton plants in the surrounding buffer rows as well as from the GM cotton plants.

CSIRO has requested that details of the gene construct including the plasmid map and regulatory sequences be declared as CCI under section 185 of the Act. However, this information has been made available to GTTAC and other prescribed expert authorities that are being consulted on the preparation of the RARMP.

GTTAC discussed this application and advised the Regulator that the following issues should be considered in the preparation of the RARMP:

- The risks posed by DIR 038/2003 are similar to those posed by previous Liberty® cotton applications such as DIR 015/2002, PR-82, PR-82X, PR 124, PR 124-X and PR 124X(2);
- Advice provided in relation to previously assessed GM cottons (Liberty® cotton) should be considered in the preparation of the RARMP for DIR 038/2003;

- At the completion of the three years of field trials, the applicant should be requested to provide data on levels of expression of the introduced protein under Australian field conditions; and
- Herbicide application practices for commercial release of the same GMO should incorporate rotation of herbicides.

- **Field evaluation of high-oleic (HO) cotton (DIR 039/2003)**

  The OGTR has received an application from CSIRO for a licence for the limited and controlled release of GM cotton into the environment. CSIRO is proposing to release two transgenic cotton lines on two hectares at one site at the Australian Cotton Research Centre Narrabri, NSW.

  The main aims of the proposed release are to conduct agronomic evaluation of the GM cotton lines and to test for maintenance of the HO phenotype under field conditions.

  Oil derived from conventional cottonseed is used in food applications around the world, following processing to remove gossypol and other toxic or anti-nutritional compounds such as cyclopropenoid fatty acids. The high levels of polyunsaturated fatty acids present in most non-GM cotton seed often necessitates additional processing through partial hydrogenation to obtain oil with higher stability and more resistant to oxidation (ie to avoid becoming rancid). However, hydrogenation results in fatty acid structural forms (trans, rather than the cis arrangement of hydrogen atoms more commonly found in nature) that may increase cholesterol levels upon consumption. HO cotton has an altered ratio of fatty acids in cottonseed oil, with increased oleic acid levels (monounsaturated fatty acid) and decreased levels of linoleic (polyunsaturated fatty acid with low stability) and palmitic acids (saturated fatty acid associated with blood cholesterol-raising properties). Oil from GM HO cottonseed is expected to have a greater stability than other seed oils. This may enable direct use in frying or for margarine hard stock, without the need for hydrogenation that current oils require.

  None of the cotton plants from the release, or their by-products, will be used for animal or human consumption. The applicant is proposing to sell lint from non-GM cotton used as pollen trap rows surrounding the release site, but not from the release. Lint does not contain genetic material or protein.

  GTTAC discussed this application and advised the Regulator that the following issues should be considered in the preparation of the RARMP:

  - The risks posed by DIR 039/2003 are low and manageable;
  - Advice provided in relation to previously assessed cottons should be considered in the preparation of the RARMP for DIR 039/2003;
  - The applicant should be requested to provide complete fatty acid, gossypol and protein profiles of cottonseed at the completion of the field trial; and
  - Post-harvest monitoring should be conducted for twelve months, although non-cotton crops could be planted six months post-harvest.

- **Agronomic assessment and seed increase of GM cotton expressing insecticidal genes from *Bacillus thuringiensis* (DIR 040/2003)**

  The OGTR has received an application from Dow AgroSciences Australia Limited (Dow AgroSciences) for a licence for the intentional release of GM insect resistant / herbicide tolerant cotton into the environment, on a limited scale and under controlled conditions on two sites covering a total area of 0.04 hectares in NSW.

There have been no previous releases of this GM cotton in Australia. The main aim of the proposed release is to evaluate the agronomic performance and insecticidal efficacy of a cotton line modified to express two insecticidal proteins (Cry1Ac and Cry1Fa) that are toxic to lepidopteran caterpillar pests of cotton. This line also contains a marker gene (*pat*) which confers tolerance to the herbicide glufosinate ammonium. Seed would also be retained for potential future releases, which would require further licence applications and separate assessment processes.

None of the cotton plants from the release, or their by-products, would be used for animal or human food.

Some specific Dow AgroSciences documents, which contain some details of the gene construction, gene sequence information and molecular characterisation of the GMO, have been declared as CCI under section 185 of the Act. However, this information has been made available to GTTAC and other prescribed expert authorities that are being consulted on the preparation of the RARMP.

GTTAC discussed this application and advised the Regulator that the following issues should be considered in the preparation of the RARMP:

- The risks posed by DIR 040/2003 are similar to those posed by previous GM cotton applications;
- The advice provided in relation to previously assessed DIRs 005/2001, 006/2001, 008/2001, 009/2001, 017/2002 and 023/2002 should be considered in the preparation of the RARMP for DIR 040/2003; and
- At the completion of the field trial, the applicant should be requested to provide data on the levels of expression of the introduced proteins under Australian field conditions.

Advice on RARMPs

Advice on cotton

GTTAC considered the RARMP prepared in response to the following application concerning the release of transgenic cotton in Australia.

- **Commercial release of herbicide tolerant (Roundup ReadyÒ) and herbicide tolerant/insect resistant (Roundup Readyâ/INGARDâ) cotton (DIR 023/2002)**

  The OGTR has received a licence application from Monsanto for the intentional release of Roundup Ready® and Roundup Ready®/INGARD® cotton into the environment in the cotton growing regions of NSW and QLD, south of latitude 22º South. Approval would enable the continued commercial release of the GM cotton. Monsanto also proposes the phasing-out of Roundup Ready®/INGARD® cotton over the next two years while Roundup Ready®/Bollgard II® cotton (which was approved for commercial release in September 2002; DIR 012/2002) is phased-in over the same period.

  Roundup Ready® cotton contains a gene that provides tolerance to glyphosate, the active ingredient of the herbicide Roundup®. Conventional cotton is susceptible to glyphosate. The use of Roundup Ready® cotton allows the application of Roundup® for the control of weeds that emerge in the crop. Roundup Ready®/INGARD® cotton was produced by conventional breeding of Roundup Ready® cotton with INGARD® cotton. The Roundup Ready®/INGARD® cotton inherits an insecticidal gene from INGARD® cotton that produces a protein toxic to lepidopteran caterpillar pests.

It is intended that GM cotton plants and their by-products, including cottonseed, be used in the same manner as conventional cotton, including for human food and stockfeed. Cottonseed is processed for oil that is used in a variety of food products and for cotton linters (a type of fibre that does not contain any genetic material) that are used as a cellulose base for several consumer food products. FSANZ has approved the use of oil and linters from Roundup Ready®, INGARD® and Bollgard II® cotton in human food.

GTTAC discussed the RARMP for this application and advised the Regulator as follows:

- The Committee agrees with the assessment made by the OGTR on risk of toxicity, allergenicity, weediness and gene transfer; and
- The Committee agrees with the proposed licence conditions.

Advice on Papaya

GTTAC considered the RARMP prepared in response to the following application concerning the release of transgenic papaya in Australia.

- **Field trial for evaluation of genetically modified papaya to delay fruit ripening and to test the expression of the introduced genes (DIR 026/2002)**

  An application has been received from the University of Queensland for a licence for the intentional release of GM papaya (*Carica papaya* L, cultivar 'Solo') plants into the environment. Approval would enable the continued limited and controlled release (field trial) of GM papaya approved under the former voluntary system (PR-128), as well as for several new lines of GM papaya. The applicant proposes to study up to 300 individual papaya plants at one site, over a total area of 1.7 hectares in QLD.

  Papaya fruit has poor storage qualities. If fruit ripening is delayed over several days to weeks it may be possible to decrease spoilage due to over-ripening during transportation and storage. The release involves growing several lines of papaya plants that have been modified to delay fruit ripening by down-regulation of a plant hormone, ethylene, or by modifying the ethylene receptor molecule. Some plants have also been modified to express a reporter gene that can be used to identify the plants with genetic modifications.

  The applicant aims to assess the rate of fruit ripening on the tree for a limited number of fruits but proposes to harvest most fruits before full ripening has occurred. Additionally, reporter gene expression will be evaluated to assess the effectiveness of the same promoter that drives expression of the fruit ripening genes. Other physiological, nutritional and quality attributes of the fruit will also be evaluated.

  None of the fruits that are produced during the trial will be used for human or animal consumption.

  GTTAC discussed the RARMP for this application and advised the Regulator as follows:

  - The Committee agrees with the assessment made by the OGTR on the risk of toxicity, allergenicity, weediness and gene transfer;
  - The condition requiring bagging of all flowers, including male, female and hermaphrodite flowers, could be removed;
  - Reference to gene containment due to 'geographical isolation' should be removed from the RARMP;
  - The integrity of the insect-proof cage should be ensured by appropriate construction and frequent monitoring; and

- The GMO's susceptibility to disease compared to that of wild-type papaya should be an area of future study.

Advice on Pineapple

GTTAC considered the RARMPs prepared in response to the following applications concerning the release of transgenic pineapple in Australia.

- **Field test of pineapple plants modified to control flowering (DIR 027/2002)**

  An application has been received from the University of Queensland for a licence to continue the limited and controlled release (field trial) of GM pineapple (*Ananas comosus*, now called *Ananas comosus* var. *comosus*) first planted in 1999 under the former voluntary system (PR-95). The University of Queensland is proposing to continue the trial on one site in QLD, over a total area of 0.1 hectares.

  The aim of the proposed release is to test pineapple plants that have been modified to control flowering. Another aim of the release is to assess the activity of different regulatory sequences under field conditions.

  The GM pineapple plants have been modified by insertion of a truncated copy of the pineapple ACC (1-aminocyclopropane-1-carboxylate) synthase (*ACACS3*) gene to 'silence' the existing gene in the pineapple. ACC synthase is a key enzyme in the pathway that leads to formation of ethylene in plants and has a role in natural flowering. Other GM pineapple plants have been modified by insertion of a reporter gene (*uidA*), which allows assessment of the activity of different regulatory sequences under field conditions. All of the GM pineapples also contain a selectable marker gene (*SuRB*) conferring resistance to ALS inhibitors, including sulfonylurea herbicide, which is used to select transgenic plants in the laboratory.

  None of the pineapple plants from the trial, or their by-products, will be used for human food, animal feed or therapeutics.

  GTTAC discussed the RARMP for this application and advised the Regulator as follows:

  - The Committee agrees with the assessment made by the OGTR on risk of toxicity, allergenicity, weediness and gene transfer; and
  - The Committee agrees with the proposed licence conditions.

  GTTAC provided the following general advice on DIR RARMPs:

  - Future RARMPs should indicate where the applicant proposes containment measures that are incorporated into the licence conditions.

- **Field trial of pineapple plants modified for blackheart reduction and to delay flowering (DIR 028/2002)**

  An application has been received from the Queensland Department of Primary Industries for a licence to continue the limited and controlled release (field trial) of GM pineapple (*Ananas comosus*, now called *Ananas comosus* var. *comosus*) first planted in 2000 under the former voluntary system (PR-137 and PR-152). The applicant is proposing to continue the trial on two sites in QLD covering a total area of 0.22 hectares.

The aims of the proposed release are to conduct a field evaluation of pineapple plants that have been modified for blackheart reduction and/or to control flowering, as well as to assess the activity of different regulatory sequences under field conditions.

The QDPI proposes the release of four types of genetically modified pineapples. One type of GM pineapple proposed for release contains an additional copy of the pineapple polyphenol oxidase (*PINPPO2*) gene that controls the occurrence of blackheart disorder and an additional copy of the pineapple ACC (1-aminocyclopropane-1-carboxylate) synthase (*AC-ACS2*) gene that controls natural flowering. Two other types of GM pineapple contain an additional copy of either the polyphenol oxidase gene or ACC synthase gene. The fourth type of pineapple has been genetically modified by introduction of ß-glucuronidase (*uidA*) gene responsible for ß-glucuronidase expression to test for activity of regulatory sequences under field conditions. All the GMOs also contain the neomycin phosphotransferase (*nptII*) gene, which confers antibiotic resistance and some associated regulatory sequences. In addition, some of the GMOs also contain non-expressed bacterial genes, ie. β-galactosidase (*Lac Z*), ampicillin (*bla*) and streptomycin/spectinomycin (*aad*) genes. These genes are under the control of bacterial promoters and will not be expressed in the GM pineapple plants.

None of the pineapple plants from the trial, or their by-products, will be used for human food, animal feed or therapeutics.

Details of the gene constructs and the gene silencing strategy have been declared CCI under section 185 of the Act. However, this information has been made available to GTTAC and other prescribed expert authorities that are being consulted on the RARMP.

GTTAC discussed the RARMP for this application and advised the Regulator as follows:

- The Committee agrees with the assessment made by the OGTR on the risk of toxicity, allergenicity, weediness and gene transfer; and
- The Committee agrees with the proposed licence conditions.

GTTAC provided the following general advice to the Regulator:

- GTTAC recommends that a paper on the pleiotropic effects of polyphenol oxidase (PPO) should be commissioned to aid the Committee in assessing future applications involving silencing PPO genes.

Advice on Grapevine

GTTAC considered the RARMP prepared in response to the following application concerning the release of transgenic grapevine in Australia.

- **Field trial of genetically modified grapevines – evaluation of berry colour, sugar composition, flower and fruit development and gene flow study (DIR 031/2002)**

  An application has been received from CSIRO for a licence for the intentional release of GM grapevines (*Vitis vinifera* L) into the environment. Approval would enable the continued limited and controlled release (field trial) of GM grapevines, approved under the former voluntary system, on one site in Victoria, over a total area of 0.38 hectares.

  The aims of the proposal are to evaluate the field performance of GM grapevines containing additional copies of various grapevine genes which are modified to improve berry colour, sugar composition, flowering and fruit quality. The applicant also proposes to monitor pollen flow using GM grapevine containing green fluorescent protein (GFP).

Several types of GM grapevines are proposed for release. The GMO's will contain one of the following modifications:

- Additional copies of the *ppo* gene, derived from grapevine, coding for the enzyme polyphenol oxidase. The modified *ppo* gene derived from grapevine has been introduced to test silencing of the natural copy of the *ppo* gene, thereby reducing browning in GM sultanas.
- Modified *sh4* gene from grapevine, designed to improve flower and fruit characters, which are beneficial to the grape industry.
- Additional copies of a *ufgt* gene encoding UDP glucose flavonoid 3-O-glucosyl transferase from grapevine which is involved in the anthocyanin pathway, designed to improve berry colour through enhanced expression of the *ufgt* gene.
- The modified *dfr* gene from grapevine which is designed to down-regulate the expression of the enzyme dihydroflavonol reductase, to alter the production of anthocyanin or tannin which are important for the stabilisation of colour during wine making and for wine mouthfeel.
- The modified *inv* gene designed to down-regulate the invertase enzyme, which breaks down sucrose into fructose and glucose. This is expected to maintain sucrose levels in the grape berries.
- The *gfp* reporter gene isolated from jellyfish (*Aequorea victoria*), encoding for GFP that enables the visual identification of plant tissues expressing the transgene.
- All of the GM grapevines contain antibiotic resistance marker genes, either *nptII* (confers resistance to kanamycin & neomycin) or *hph* (confers resistance to hygromycin).

None of the grapevine plants from the release, or their by-products, would be used as animal or human food.

GTTAC discussed the RARMP for this application and advised the Regulator as follows:

- The Committee agrees with the assessment made by the OGTR on risk of toxicity, allergenicity, weediness and gene transfer;
- The Committee agrees with the licence conditions; and
- The RARMP should be amended to clarify that taste testing would not be allowed until further information on toxicity and allergenicity of the GM fruit became available.

Advice on Cholera Vaccine

GTTAC considered the RARMP prepared in response to the following application concerning the release of genetically modified cholera vaccine in Australia.

- **Orochol© vaccine (DIR 033/2002)**

  An application has been received from CSL Limited for a licence for the continued commercial release of live GM cholera vaccine (Orochol®). The proposal was previously approved under the former voluntary system.

  Cholera is a disease with an extremely low incidence in Australia. In the last ten years, an average of four cases have been reported annually. The majority of these cases involved people who had entered or returned to Australia from other countries.

  Orochol® is a self-administered prescription medicine to immunise people intending to travel against cholera. Following extensive evaluation of its safety, quality and efficacy, the vaccine was registered as a prescription medicine under the *Therapeutic Goods Act 1989* in April 2000. Since this time, over 60,000 doses have been distributed nationally.

Orochol® vaccine contains the live bacterium *Vibrio cholerae*. Native cholera bacteria produce a toxin containing 2 subunits, A and B. The GM vaccine strain has been produced by deleting most of the toxic A-subunit gene (*ctxA*) and inserting a mercury resistance operon (*mer*) into the haemolysin gene (*hlyA*). The non-active B-subunit of the cholera molecule is still synthesised but it does not cause disease.

GTTAC discussed the RARMP for this application and advised the Regulator as follows:

- The Committee endorsed the RARMP and the proposed licence conditions for DIR 033/2002; and
- The GMO should be considered for entry on the GMO Register.

New certification guidelines for contained facilities

OGTR representatives provided the Committee with a copy of the proposed revised *Guidelines for the Certification of Physical Containment Facilities* (the Guidelines) that will be issued in August 2003. The Committee discussed the draft document, provided advice to the Regulator on waste disposal in plant containment facilities and endorsed these revised Guidelines.

Draft policy principle

OGTR representatives provided the Committee with an overview and copy of the draft Gene Technology (Recognition of Designated Areas) Policy Principle 2003. The Committee was asked to provide advice to the Gene Technology Ministerial Council (GTMC) on the content of the draft policy principle.

The Committee discussed the draft Gene Technology (Recognition of Designated Areas) Policy Principle 2003.

GTTAC advised the GTMC that:

- No amendments are required to the content of the draft policy principle and related documents; however
- The Committee seeks the GTMC's assurance that the policy principle could not be used to restrict contained dealings involving GMOs (ie DNIRs and NLRDs).

Presentations

At the May meeting of GTTAC the Committee received and discussed a presentation on *Gene Containment*.

Enquiries and Risk Assessment and Risk Management Plans

For all enquiries and to obtain copies of applications or RARMPs for dealings involving the intentional release of GMOs into the environment, please phone the OGTR Free-call hotline on 1800 181 030. The RARMPs are also available electronically from our website at http://www.ogtr.gov.au/publications/riskassessments.htm

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