



**APPLICATION FOR LICENCE FOR INTENTIONAL RELEASE OF GMOs INTO THE ENVIRONMENT: Application No. DIR 086/2008**

**SUMMARY INFORMATION**

Project Title:	Limited and controlled release of maize genetically modified to investigate gene function <sup>1</sup>
Applicant:	CSIRO
Common name of the parent organism:	Maize
Scientific name of the parent organism:	<i>Zea mays</i> L.
Modified trait(s):	<ul style="list-style-type: none"><li>• Presence of a modified maize transposable genetic element to ‘tag’ genes for investigation of their function</li><li>• Antibiotic resistance</li><li>• Herbicide tolerance</li><li>• Marker gene expression</li></ul>
Identity of the gene(s) responsible for the modified trait(s):	<ul style="list-style-type: none"><li>• <b><i>UbiDs</i></b> (a modified transposable genetic element and regulatory sequences) from <i>Zea mays</i> L.</li><li>• <b><i>hyg</i></b> (encoding hygromycin phosphotransferase) from the bacterium <i>Escherichia coli</i> (antibiotic resistance selectable marker)</li><li>• <b><i>bar</i></b> (encoding phosphinothricin acetyltransferase) from the bacterium <i>Streptomyces hygrosopicus</i></li><li>• <b><i>uidA</i></b> (encoding GUS) from the bacterium <i>Escherichia coli</i> (reporter gene)</li></ul>
Proposed Location(s):	One site in the ACT
Proposed Release Size:	0.15 ha
Proposed Release Dates:	November 2008 – May 2011

**Introduction**

The *Gene Technology Act 2000* (the Act) in conjunction with the *Gene Technology Regulations 2001*, an inter-governmental agreement and corresponding legislation that is being enacted in each State and Territory, comprise Australia’s nationally consistent regulatory system for gene technology. Its objective is to protect the health and safety of people, and the environment, by identifying risks posed by or as a result of gene technology, and managing those risks by regulating certain dealings with genetically modified organisms (GMOs).

The Act establishes a statutory officer, the Gene Technology Regulator (the Regulator), to administer the legislation and make decisions under the legislation. The Regulator is supported by the Office of the Gene Technology Regulator (OGTR), an Australian Government regulatory agency located within the Health and Ageing portfolio.

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<sup>1</sup> The title of the licence application submitted by CSIRO is ‘A field-based seed increase of transgenic maize activation tagging lines’.

The legislation sets out the requirements for considering applications for licences for dealings with GMOs and the matters that the Regulator must take into account before deciding whether, or not, to issue a licence. The Regulator's *Risk Analysis Framework*<sup>2</sup> outlines the assessment process that will be followed.

## **The application and the proposed dealings**

The Acting Regulator has received an application from CSIRO for a licence for dealings involving the intentional release of genetically modified (GM) maize (*Zea mays* L.) into the Australian environment on a limited scale under controlled conditions.

Eleven lines<sup>3</sup> of GM maize are proposed for release. Each line contains a modified version of a genetic element known as a transposable genetic element that can move within the maize genome. If the transposable genetic element moves into a region controlling the expression of a particular gene, that gene may be over-expressed, effectively 'tagging' that gene.

If this process occurred in maize cells that produce seeds, the offspring could have traits not observed in the parent plants. Seed produced during the proposed release would subsequently be grown in the glasshouse to determine whether those traits are of biological or agronomic interest.

The applicant proposes to limit the release to one site at a CSIRO research facility in the ACT on a maximum area of 0.15 ha between November 2008 and May 2011.

The applicant has also proposed a number of control measures to restrict the dissemination or persistence of the GM plants and their introduced genetic material that will be considered in the assessment of this application including:

- not using the GMOs in human food or animal feed
- restricting access to the site to CSIRO staff only
- conducting the trial within an enclosure that restricts animal access
- detasseling of all GM maize plants (ie removal of the male flowers) to assure no pollen is produced by the GMOs
- pollinating the GMOs under controlled conditions with non-GM maize pollen
- harvesting maize seed by hand
- cleaning the site after harvest by slashing and incorporating all plant material into the soil, except for materials required for future research or release
- monitoring the trial site for at least 8 months and destroying any maize volunteers
- transporting GM seed and plant materials in accordance with OGTR transportation guidelines

## **Parent organism**

The parent organism is maize (*Zea mays* L.), which is exotic to Australia and can be grown as an agricultural crop in all States and Territories in Australia. The female parent of the GM maize lines proposed for release was cultivar Hi II. This cultivar has been adapted to tissue culture conditions and can be modified by standard genetic techniques. The male parent of the GMOs proposed for release was the conventional maize cultivar, Pvv.

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<sup>2</sup> More information on the assessment of licence applications is available from the Office of the Gene Technology Regulator (OGTR). Free call 1800 181 030 or at <<http://www.ogtr.gov.au/ir/process.htm>>.

<sup>3</sup> The term 'line' is used to denote plants derived from a single plant containing a specific genetic modification made by one transformation event.

## **The genetic modifications and their effect**

The GM maize lines contain a modified transposable genetic element, *Ds*, originally derived from maize. Two copies of the regulatory and other non-coding sequences from the maize polyubiquitin gene have also been introduced and together are termed *Ubi*. The *bar* gene (see below) and its regulatory sequences are also present within the modified transposable genetic element. The *Ds* element with its associated *bar* and *Ubi* sequences has been named *UbiDs*. The intended effect of *UbiDs* is to move into new regions of the genome to induce gene over-expression in the affected cells.

The GM maize lines also contain the *hyg*, *bar* and *uidA* genes, encoding hygromycin phosphotransferase, phosphinothricin acetyltransferase and  $\beta$ -glucuronidase.

The *hyg* gene was derived from the common gut bacterium *Escherichia coli* and confers resistance to the antibiotic hygromycin. It was used to select for modified maize cells in the laboratory and also contains the intron from the castor bean (*Ricinus communis*) catalase gene to prevent bacterial expression of this gene. Hygromycin would not be applied to the GM plants during the proposed field trial.

The *bar* gene was derived from *Streptomyces hygroscopicus* and confers tolerance to phosphinothricin based herbicides. It may be used to identify GM maize plants containing the *UbiDs* element in future glasshouse experiments. Phosphinothricin based herbicides would not be used on the GM plants during the proposed field trial.

The *uidA* gene, a reporter gene, was derived from *Escherichia coli* and encodes the enzyme  $\beta$ -glucuronidase. Its expression depends on the immediately adjacent *UbiDs* element. After *UbiDs* has moved into a new region in the genome, the reporter gene will not be expressed. It can, therefore, be used to monitor for movement of the *UbiDs* element in the GMOs and their offspring.

The GM maize lines also contain additional short regulatory sequences that control expression of the introduced genes. These are derived from a soil bacterium (*Agrobacterium tumefaciens*) and a plant virus (Cauliflower mosaic virus). The regulatory sequences of the plant pathogens comprise only a small part of their total respective genome.

## **Method of genetic modification**

The gene construct was introduced into maize on a plasmid vector carried by *A. tumefaciens*. The vector is 'disarmed' since it lacks the genes that encode the tumorigenic functions of *A. tumefaciens*. This method has been widely used in Australia and overseas for introducing new genes into plants.

Transformed plant tissues were identified using the expression of the antibiotic resistance marker gene and grown into plants in the laboratory.

## **Previous releases of the same or similar GMOs**

There has been no previous release of these GM maize lines or any other GM maize in Australia.

## **Suitability of Applicant**

Section 43(2)(f) of the Act requires the Regulator to be satisfied regarding the suitability of the applicant to hold a licence as a pre-requisite for considering DIR applications. The matters to be considered are outlined in Section 58 of the Act and include relevant convictions, revocation of a licence or permit relating to the health and safety of people, and capacity to meet the conditions of the licence.

The Acting Regulator has determined that CSIRO currently meets the suitability requirements and will verify this continues to be the case prior to making any decision regarding the issuing of a licence.

## **Consultation process for this DIR application**

The Regulator has made an assessment of whether the application should be considered as a limited and controlled release, under with Section 50A of the Act. As its principal purpose is to enable the conduct of experiments, and the applicant has proposed limits on the size and duration of the release and controls to restrict the dissemination and persistence of both the GMO and its genetic material in the environment, **the Acting Regulator has decided that the application qualifies as a limited and controlled release.**

This means that the Acting Regulator is not required to consult on the assessment of this application until after a Risk Assessment and Risk Management Plan (RARMP) has been prepared in accordance with section 51 of the Act. In the interim, copies of the application are available on request from the OGTR. Please quote application number DIR 086/2008.

The Acting Regulator will seek comment on the consultation RARMP from the public as well as a wide range of experts, agencies and authorities including the Gene Technology Technical Advisory Committee, State and Territory Governments, Australian Government agencies and the Minister for the Environment, Heritage and the Arts. The RARMP will then be finalised, taking into account matters raised relating to risks to human health and safety and the environment, and form the basis of her decision whether or not to issue a licence.

At this stage, **the RARMP is expected to be released for comment in October 2008.** The public will be invited to provide submissions on the RARMP via advertisements in the media and direct mail to anyone registered on the OGTR mailing list. The RARMP and other related documents will be available on the OGTR website, or in hard copy from the OGTR.

If you have any questions about the application or the assessment process, or wish to register on the mailing list, please contact the OGTR at:

**The Office of the Gene Technology Regulator, MDP54 GPO Box 9848 Canberra ACT 2601**

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