



APPLICATION FOR LICENCE FOR INTENTIONAL RELEASE OF GMOs INTO THE ENVIRONMENT: Application No. DIR 070/2006

SUMMARY INFORMATION

Project Title:	Limited and controlled release of GM sugarcane with altered plant architecture, enhanced water or improved nitrogen use efficiency. ¹
Applicant:	Bureau of Sugar Experiment Stations Limited
Common name of the parent organism:	Sugarcane
Scientific name of the parent organism:	<i>Saccharum spp.</i> hybrid
Modified trait(s):	Shoot architecture (shoot number, stalk size and height), water use efficiency, nitrogen use efficiency, and marker gene expression (antibiotic resistance and reporter genes).
Identity of the gene(s) responsible for the modified trait(s):	<ul style="list-style-type: none">• 14 genes for altered plant architecture from rice (<i>Oryza sativa</i>), sugarcane (<i>Saccharum Spp.</i>), barley (<i>Hordeum vulgare subsp. vulgare</i>), bean (<i>Phaseolus coccineus</i>)• 3 genes for enhanced water use efficiency from the bacterium <i>Escherichia coli</i>, thale cress (<i>Arabidopsis thaliana</i>), apple (<i>Malus x domestica</i>)• 1 gene for improved nitrogen use efficiency from corn (<i>Zea mays</i>)• <i>uidA</i> (beta-glucuronidase or GUS) from the bacterium <i>E. coli</i> (reporter gene)• <i>nptII</i> (neomycin phosphotransferase type II) from the bacterium <i>E. coli</i> (antibiotic resistance selectable marker)• <i>bla</i> gene from the bacterium <i>E. coli</i> (antibiotic resistance selectable marker)
Proposed Location(s)	Three sites per season over three seasons in the shires of Caboolture, Bundaberg City Council, and Cairns City Council (Qld)
Proposed Release Size:	Up to 6 hectares per season over three growing seasons
Proposed Release Dates:	February 2007 to November 2010

¹Applicant title is "GM sugarcane field trial – testing the effect on sugar yield of transformation methods".

Introduction

The *Gene Technology Act 2000* (the Act) took effect on 21 June 2001. The Act, supported by the *Gene Technology Regulations 2001*, an inter-governmental agreement and corresponding legislation that is being enacted in each State and Territory, underpins 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.

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 licence².

The application and the proposed dealings

The OGTR has received an application from Bureau of Sugar Experiment Stations Limited (BSES Ltd) for a licence for the intentional release of up to 2500 lines of genetically modified (GM) sugarcane (*Saccharum spp.* hybrid) into the environment on a limited scale and under controlled conditions.

The aim of the study is to conduct early stage research to examine the effect of using different genetic modification methods used to transform sugarcane and the influence of the introduced genes for altered plant architecture, enhanced water or improved nitrogen utilisation on key altered agronomic characters, including sugarcane yield.

The release is proposed to take place at three sites; Caboolture Shire, Bundaberg City Council, and Cairns City Council in Queensland (Qld) on a maximum area of 6 ha during each of the three growing seasons between February 2007 and November 2010.

The applicant has proposed a number of containment measures for the conduct of the field trial that will be considered during the assessment of the application including:

- Surrounding the trial sites by one buffer row of non-GM sugarcane acting as wind barriers and an isolation zone of at least 6 metres
- Regular monitoring of the proposed trial site to prevent GM sugarcane lines from flowering to limit the dispersal of pollen
- None of the material from the proposed trial will be used in human food, animal feed, or other commercial purposes
- Destructive analysis of GM plant materials for research purposes
- Destruction of all GM plant materials not required for further research or propagation at the end of the proposed trial by ploughing out and herbicide treatment and either mulching or burning
- Post harvest monitoring of trial sites regularly for 12 months and destroying any volunteer sugarcane plants with herbicide treatment
- Transportation of GM sugarcane and plant materials in accordance with OGTR transportation guidelines.

² More information on the assessment of licence applications and copies of the *Risk Analysis Framework* are 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> and <http://www.ogtr.gov.au/pdf/public/raffinal2.2.pdf> respectively.

Previous releases of the same or similar GMOs

The GM sugarcane lines proposed for release have not been previously released in Australia or elsewhere.

Under the former voluntary system overseen by the Genetic Manipulation Advisory Committee (GMAC), there have been seven field trials of different types of GM sugarcane in Queensland covering areas ranging from 0.1 – 1.0 ha.

In addition, the Regulator issued licences for limited and controlled release of two other types of GM sugarcane lines: DIR 019/2002 to BSES Ltd in 2002 on an area of 0.7 ha in the Cairns district of Queensland and DIR 051/2004 was issued to the University of Queensland for another type of GM sugarcane lines in 2005 on two sites covering an area of 3.55 hectares in the Burdekin district of Queensland.

There have been no reports of adverse effects on human health or the environment resulting from these releases.

Parent organism

The parent organism is a commercial sugarcane hybrid, Q117, one of Australia's main commercial varieties derived from an interspecific cross between *Saccharum officinarum* L. and *Saccharum spontaneum* L. Sugarcane is exotic to Australia but has been grown commercially as an agricultural crop in Queensland and New South Wales since 1823 and more recently in the Ord district of Western Australia.

Genetic modification and its effect

The introduced gene constructs, with the exception of constructs containing antibiotic resistance selectable marker genes (*bla* and *nptII*) and reporter gene (*uidA*), may affect shoot architecture, water or nitrogen use efficiency.

Fourteen of the 22 gene constructs contain partial and/or complete gene sequences and are expected to alter plant architecture of the GM sugarcane lines such as decreased or increased height, decreased or increased tillering and thicker or narrower stalks. These gene sequences are derived from rice, sugarcane, barley and beans.

One of the 22 gene constructs is expected to confer improved nitrogen use efficiency under low nitrogen conditions. This may lead to reduced suckering. This construct contains a gene derived from corn.

Three of the 22 gene constructs are intended to confer enhanced water use efficiency, either through the expression of a gene that controls other genes or the production of different types of sugars. Genes introduced into these three gene constructs are derived from *Escherichia coli*, thale cress and apple.

The reporter gene (*uidA*), present in one gene construct, encodes an enzyme (β -glucuronidase (GUS)) that enables visual identification of plant tissues in which this gene is being expressed. The tissue containing the GUS enzyme will turn a dark blue colour after adding a staining solution. This staining also provides an indication of the level of activity of the promoter (regulatory sequence) that is being used to control the expression of the reporter gene. The reporter gene was originally derived from the common gut bacterium *E. coli*.

Nineteen of the 22 gene constructs contain the marker gene, *bla*, from the bacterium *E. coli*, which encodes ampicillin resistance. It is linked to a bacterial promoter that does not function in plants, so the gene is not expressed in the GM sugarcane lines. The gene was used to select

for bacteria containing the desired genes, in the laboratory, prior to the production of the genetically modified plants.

Additionally, all the gene constructs contain an antibiotic resistance selectable marker gene, *neomycin phosphotransferase type II (nptII)*. The *nptII* gene encoding for the enzyme neomycin phosphotransferase was originally derived from the common gut bacterium *E. coli*, and confers kanamycin or neomycin resistance on the GM plant. The *nptII* gene was used only as a selective marker during early stages of development of the GM plants in the laboratory.

Short regulatory sequences that control expression of the genes are also present in all the gene constructs. These are derived from plants (*Flaveria trinervia* and maize), a soil bacterium (*Agrobacterium tumefaciens*) and *E. coli*. Although *A. tumefaciens* is a plant pathogen and *E. coli* is a facultative human pathogen, the regulatory sequences comprise only a small part of their respective total genomes, and are not capable of causing disease.

Method of genetic modification

Of the 22 gene constructs in GM sugarcane lines proposed for release 21 of these gene constructs were introduced into sugarcane cultivar, Q117 by microprojectile bombardment. This technique involves coating very small tungsten particles with the transformation vector containing the introduced genes. The particles were then 'shot' into small pieces of sugarcane tissue, which were then regenerated into whole plants. Particle bombardment has been widely used in Australia and overseas for introducing new genes into plants without causing any biosafety problems.

One gene construct containing the *uidA* and *nptII* genes along with the regulatory sequences was introduced into cells from plants of the sugarcane cultivar Q117 by four different species of *Agrobacterium*. These constructs are 'disarmed' since they lack the genes that encode the tumour-inducing functions of *A. tumefaciens*.

A total of 2500 GM sugarcane lines, derived from 22 gene constructs, proposed for release are the result of independent genetic modification events.

Consultation on preparation of the Risk Assessment and Risk Management Plan

The Regulator has made an initial assessment as to whether the proposed release may pose significant risks to human health and safety or the environment, in accordance with section 49 of the Act. Due to the characteristics of the introduced genes, the control measures that have been proposed, and the limited scale and scope of the dealings, **the Regulator has decided that the proposed controlled release does not pose a significant risk to human health and safety or the environment.**

This means that the Regulator is **not required to seek public comment** on the assessment of this proposal until after a risk assessment and risk management plan (RARMP) has been prepared for consultation. In the interim, copies of the application are available on request from the OGTR. Please quote application number DIR 070/2006.

In preparing the RARMP, the Regulator will seek input from a wide range of key stakeholders and expert groups including State and Territory Governments, Australian Government agencies, the Minister for the Environment and Heritage, the Gene Technology Technical Advisory Committee and the relevant local councils. The Regulator will consult again with these prescribed agencies and authorities, as well as the public, in finalising the RARMP, which then forms the basis of her decision whether or not to issue a licence.

At this stage, the consultation version of the RARMP is expected to be released for a six week consultation period in **late November 2006**. 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 from the OGTR, or on the OGTR website.

If you have any questions about the application or the assessment process, please contact the OGTR at:

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