



For Office Use Only	
Protocol No	GT /

## Notification of an Exempt Dealing

### COVER SHEET

Project Title: Cloning of mitochondrial fragments from invertebrates	
Project Supervisor: A/Professor Mark Dowton	
Faculty/Unit: School of Biological Sciences	
Telephone No: x 5653	Fax No: 02 4221 4135
Starting Date: 1/8/10	Completion Date: 31/7/15
Room(s) where GMO will be used, including containment level: 35.108, 35.110	
Room(s) where GMO is stored: 35.108, 35.G02	
Other Universities and/or facilities to be involved in the research:	

GTRC Declaration
The GTRC has evaluated this dealing and agrees that it is an exempt dealing as specified by Schedule 2 of the <i>Gene Technology Regulations 2001</i> and the <i>Gene Technology Amendment Regulations 2011</i> .

Signature of Project Supervisor: ..... Date: .....

Signature of Head of Unit: ..... Date: .....

Signature of Chair of GTRC:..... Date: .....



## Gene Technology Review Committee

### 1 Project Summary - briefly describe the project, including the aims of the proposed dealing, method of producing GMOs and their use (this should be written in plain English).

Aim: Mitochondrial fragments from a range of invertebrates will be amplified by PCR, then cloned into the pGEM-T Easy (Promega vector). This vector is non-conjugative. Engineered plasmids will then be used to transform *E. coli* (JM 109) cells. The mitochondrial fragments will then be sequenced, using vector-specific primers. Long fragments will be sequenced by primer walking. The mitochondrial fragments are not toxic.

### 2 Exemption Category – circle the item that applies

#### Part 1 Exempt dealings

Item	Description of dealing
2	A dealing with a genetically modified <i>Caenorhabditis elegans</i> , unless: (a) an <i>advantage</i> is conferred on the animal by the genetic modification; or (b) as a result of the genetic modification, the animal is capable of secreting or producing an infectious agent.
3	A dealing with an animal into which genetically modified somatic cells have been introduced, if: (a) the somatic cells are not capable of giving rise to infectious agents as a result of the genetic modification; and (b) the animal is not infected with a virus that is capable of recombining with the genetically modified nucleic acid in the somatic cells.
3A	A dealing with an animal whose somatic cells have been genetically modified <i>in vivo</i> by a replication defective viral vector, if: (a) the <i>in vivo</i> modification occurred as part of a previous dealing; and (b) the replication defective viral vector is no longer in the animal; and (c) no germ line cells have been genetically modified; and (d) the somatic cells cannot give rise to infectious agents as a result of the genetic modification; and (e) the animal is not infected with a virus that can recombine with the genetically modified nucleic acid in the somatic cells of the animal.

Item	Description of dealing
4	<p>(1) Subject to subitem (2), a dealing involving a host/vector system mentioned in Part 2 of this Schedule and producing no more than 25 litres of GMO culture in each vessel containing the resultant culture.</p> <p>(2) The donor nucleic acid:</p> <p>(a) must meet either of the following requirements:</p> <p>(i) it must not be derived from organisms implicated in, or with a history of causing, disease in otherwise healthy:</p> <p>(A) human beings; or</p> <p>(B) animals; or</p> <p>(C) plants; or</p> <p>(D) fungi;</p> <p>(ii) it must be characterised and the information derived from its characterisation show that it is unlikely to increase the capacity of the host or vector to cause harm;</p> <p><i>Example</i></p> <p>Donor nucleic acid would not comply with subparagraph (ii) if its characterisation shows that, in relation to the capacity of the host or vector to cause harm, it:</p> <p>(a) provides an advantage; or</p> <p>(b) adds a potential host species or mode of transmission; or</p> <p>(c) increases its virulence, pathogenicity or transmissibility; and</p> <p>(b) must not code for a toxin with an LD<sub>50</sub> of less than 100 µg/kg; and</p> <p>(c) must not code for a toxin with an LD<sub>50</sub> of 100 µg/kg or more, if the intention is to express the toxin at high levels; and</p> <p>(d) must not be uncharacterised nucleic acid from a toxin-producing organism; and</p> <p>(e) must not include a viral sequence, unless the donor nucleic acid:</p> <p>(i) is missing at least 1 gene essential for viral multiplication that:</p> <p>(A) is not available in the cell into which the nucleic acid is introduced; and</p> <p>(B) will not become available during the dealing; and</p> <p>(ii) cannot restore replication competence to the vector.</p>
5	<p>A dealing involving shot-gun cloning, or the preparation of a cDNA library, in a host/vector system mentioned in item 1 of Part 2 of this Schedule, if the donor nucleic acid is not derived from either:</p> <p>(a) a pathogen; or</p> <p>(b) a toxin-producing organism.</p>

### 3 Circle the host/vector system that will be used

#### Host/vector systems for exempt dealings

Item	Class	Host	Vector
1	Bacteria	<p><i>Escherichia coli</i> K12, <i>E. coli</i> B, <i>E. coli</i> C or <i>E. coli</i> Nissle 1917 — any derivative that does not contain:</p> <p>(a) generalised transducing phages; or</p> <p>(b) genes able to complement the conjugation defect in a non-conjugative plasmid</p>	<p>1. Non-conjugative plasmids</p> <p>2. Bacteriophage</p> <p>(a) lambda</p> <p>(b) lambdoid</p> <p>(c) Fd or F1 (eg M13)</p> <p>3. None (non-vector systems)</p>

Item	Class	Host	Vector
		<i>Bacillus</i> — specified species — asporogenic strains with a reversion frequency of less than $10^{-7}$ :	<ol style="list-style-type: none"> <li>1. Non-conjugative plasmids</li> <li>2. Plasmids and phages whose host range does not include <i>B. cereus</i>, <i>B. anthracis</i> or any other pathogenic strain of <i>Bacillus</i></li> <li>3. None (non-vector systems)</li> </ol>
		(a) <i>B. amyloliquefaciens</i>	
		(b) <i>B. licheniformis</i>	
		(c) <i>B. pumilus</i>	
		(d) <i>B. subtilis</i>	
		(e) <i>B. thuringiensis</i>	
		<i>Pseudomonas putida</i> — strain KT 2440	<ol style="list-style-type: none"> <li>1. Non-conjugative plasmids including certified plasmids: pKT 262, pKT 263, pKT 264</li> <li>2. None (non-vector systems)</li> </ol>
		<i>Streptomyces</i> — specified species:	<ol style="list-style-type: none"> <li>1. Non-conjugative plasmids</li> <li>2. Certified plasmids: SCP2, SLP1, SLP2, PIJ101 and derivatives</li> <li>3. Actinophage phi C31 and derivatives</li> <li>4. None (non-vector systems)</li> </ol>
		(a) <i>S. aureofaciens</i>	
		(b) <i>S. coelicolor</i>	
		(c) <i>S. cyaneus</i>	
		(d) <i>S. griseus</i>	
		(e) <i>S. lividans</i>	
		(f) <i>S. parvulus</i>	
		(g) <i>S. rimosus</i>	
		(h) <i>S. venezuelae</i>	
		<i>Agrobacterium radiobacter</i>	<ol style="list-style-type: none"> <li>1. Non-tumorigenic disarmed Ti plasmid vectors, or Ri plasmid vectors</li> <li>2. None (non-vector systems)</li> </ol>
		<i>Agrobacterium rhizogenes</i> — disarmed strains	
		<i>Agrobacterium tumefaciens</i> — disarmed strains	
		<i>Lactobacillus</i>	<ol style="list-style-type: none"> <li>1. Non-conjugative plasmids</li> <li>2. None (non-vector systems)</li> </ol>
		<i>Lactococcus lactis</i>	
		<i>Oenococcus oeni</i> syn.	
		<i>Leuconostoc oeni</i>	
		<i>Pediococcus</i>	
		<i>Photobacterium angustum</i>	
		<i>Pseudoalteromonas tunicata</i>	
		<i>Rhizobium</i> (including the genus <i>Allorhizobium</i> )	
		<i>Sphingopyxis alaskensis</i> syn.	
		<i>Sphingomonas alaskensis</i>	
		<i>Streptococcus thermophilus</i>	
		<i>Synechococcus</i> — specified strains:	
		(a) PCC 7002	
		(b) PCC 7942	
		(c) WH 8102	
		<i>Synechocystis</i> species — strain PCC 6803	
		<i>Vibrio cholerae</i> CVD103-HgR	

Item	Class	Host	Vector
2	Fungi	<i>Kluyveromyces lactis</i> <i>Neurospora crassa</i> — laboratory strains <i>Pichia pastoris</i> <i>Saccharomyces cerevisiae</i> <i>Schizosaccharomyces pombe</i> <i>Trichoderma reesei</i> <i>Yarrowia lipolytica</i>	1. All vectors 2. None (non-vector systems)
3	Slime moulds	<i>Dictyostelium</i> species	1. <i>Dictyostelium</i> shuttle vectors, including those based on the endogenous plasmids Ddp1 and Ddp2 2. None (non-vector systems)
4	Tissue culture	Any of the following if they cannot spontaneously generate a whole animal: (a) animal or human cell cultures (including packaging cell lines); (b) isolated cells, isolated tissues or isolated organs, whether animal or human; (c) early non-human mammalian embryos cultured <i>in vitro</i>  Either of the following if they are not intended, and are not likely without human intervention, to vegetatively propagate, flower or regenerate into a whole plant: (a) plant cell cultures; (b) isolated plant tissues or organs	1. Non-conjugative plasmids 2. Non-viral vectors, or replication defective viral vectors unable to transduce human cells 3. Baculovirus ( <i>Autographa californica</i> nuclear polyhedrosis virus), polyhedrin minus 4. None (non-vector systems)  1. Non-tumorigenic disarmed Ti plasmid vectors, or Ri plasmid vectors, in <i>Agrobacterium tumefaciens</i> , <i>Agrobacterium radiobacter</i> or <i>Agrobacterium rhizogenes</i> 2. Non-pathogenic viral vectors 3. None (non-vector systems)