THE UNIVERSITY OF WESTERN ONTARIO
BIOHAZARDOUS AGENTS REGISTRY FORM
Approved Biohazards Subcommittee: July 25, 2008
Biosafety Website: www.uwo.ca/humanresources/biosafety/

This form must be completed by each Principal Investigator holding a grant administered by the University of Western Ontario or in charge of a laboratory/facility where the use of Level 1, 2 or 3 biohazardous agents are described in the laboratory or animal work proposed. The form must also be completed if any work is proposed involving animals carrying zoonotic agents infectious to humans or involving plants, fungi, or insects that require Health Canada (HC) or Canadian Food Inspection Agency (CFIA) permits. The form must also be completed if any work is proposed involves plants or insects that require Health Canada (HC) or Canadian Food Inspection Agency (CFIA) permits.

This form must also be updated at least every 3 years or when there are changes to the biohazards being used.

Containment Levels will be required in accordance with Laboratory Biosafety Guidelines, 3rd edition, 2004, Health Canada (HC) or Containment Standards for Veterinary Facilities, 1st edition 1996, Canadian Food Inspection Agency (CFIA).

Completed forms are to be returned to Occupational Health and Safety, OHS (Stevenson-Lawson Building, Room 295) for distribution to the Biohazard Subcommittee. For questions regarding this form, please contact the Biosafety Officer at extension 81135. If there are changes to the information on this form (excluding grant title and funding agencies), modifications must be submitted to Occupational Health and Safety. See website: www.uwo.ca/humanresources/biosafety/

PRINCIPAL INVESTIGATOR SAVITA DHANVANTARI
SIGNATURE S. Dhanvantari
DEPARTMENT LHRI
ADDRESS 268 GROSVENOR ST LONDON ON N6A4V2
PHONE NUMBER X 65738
EMAIL sdhanvan@uwo.ca

Location of experimental work to be carried out: Building(s) SJHC Room(s) F4-127a

*For work being performed at Institutions affiliated with the University of Western Ontario, the Safety Officer for the Institution where experiments will take place must sign the form prior to its being sent to Occupational Health and Safety (See Section 12.0, Approvals). For research being done at Lawson Health Research Institute, London Regional Cancer Program, Child and Parent Research Institute, or Robarts Research Institute, a University Biosafety Committee member can also sign as the Safety Officer for the Institution.

FUNDING AGENCY/AGENCIES: SEE ATTACHED FOR GRANTS, TITLES + SUMMARIES
GRANT TITLE(S):

PLEASE ATTACH A BRIEF DESCRIPTION OF YOUR WORK THAT EXPLAINS THE BIOHAZARDS USED AND HOW THEY WILL BE USED. PROJECTS SUBMITTED WITHOUT A SUMMARY WILL NOT BE REVIEWED.

Names of all personnel working under Principal Investigators supervision in this location:

REBECCA MCGIRR
TUMELO MOLEKO
VANESSA ROTA
SHIRLEY (YUE) HU
MARK MIGUELS

*** DESCRIPTION MUST BE ATTACHED TO THIS FORM OR PROJECT WILL NOT BE REVIEWED***

1.0 Microorganisms

1.1 Does your work involve the use of microorganisms or biological agents of plant or animal origin (including but not limited to viruses, prions, parasites, bacteria)? ⊘YES O NO

If no, please proceed to Section 2.0

Do you use microorganisms that require a permit from the CFIA? ⊘YES O NO

If YES, please give the name of the species. _____

What is the origin of the microorganism(s)? _____

Please describe the risk (if any) of escape and how this will be mitigated:

ALL MATERIAL TO BE HANDLED BY TRAINED STAFF IN LEVEL 2+3 CONTAINMENT

Please attach the CFIA permit.

Please describe any CFIA permit conditions:

IN VITRO CELL WORK ONLY, @ * AS PER ATTACHED PERMIT

1.2 Please complete the table below:

Name of Biological agent(s)*	Is it known to be a human pathogen? YES/NO	Is it known to be an animal pathogen? YES/NO	Is it known to be a zoonotic agent? YES/NO	Maximum quantity to be cultured at one time? (in Litres)	Source/ Supplier	Health Canada or CFIA Containment Level
LENTIVIRUS	⊘Yes O No	⊘Yes O No	⊘Yes O No	0.1	INVITROGEN	O 1 ⊘2 O 3 +3
	O Yes O No	O Yes O No	O Yes O No			O 1 O 2 O 3
	O Yes O No	O Yes O No	O Yes O No			O 1 O 2 O 3
	O Yes O No	O Yes O No	O Yes O No			O 1 O 2 O 3

*Please attach a Material Safety Data Sheet or equivalent from the supplier.

2.0 Cell Culture

2.1 Does your work involve the use of cell cultures? ⊘YES O NO

If no, please proceed to Section 3.0

2.2 Please indicate the type of primary cells (i.e. derived from fresh tissue) that will be grown in culture in the table below

Cell Type	Is this cell type used in your work?	Source of Primary Cell Culture Tissue	AUS Protocol Number
Human	O Yes ⊘No		Not applicable
Rodent	O Yes ⊘No		
Non-human primate	O Yes ⊘No		
Other (specify)	O Yes ⊘No		

*** DESCRIPTION MUST BE ATTACHED TO THIS FORM OR PROJECT WILL NOT BE REVIEWED***

2.3 Please indicate the type of established cells that will be grown in culture in the table below.

Cell Type	Is this cell type used in your work?	Specific cell line(s)*	Supplier / Source
Human	O Yes ⊘ No		
Rodent	⊘ Yes O No	CHO, INS-1, ATCI-6, INRI-G9, GLUTag, N2A, PC12	SEE ATTACHED
Non-human primate	O Yes ⊘ No		
Other (specify)	O Yes ⊘ No		

*Please attach a Material Safety Data Sheet or equivalent from the supplier. (For more information, see www.atcc.org)

2.4 For above named cell types(s) indicate HC or CFIA containment level required O 1 ⊘ 2 O 3

3.0 Use of Human Source Materials

3.1 Does your work involve the use of human source materials? O YES ⊘ NO
If no, please proceed to Section 4.0

3.2 Indicate in the table below the Human Source Material to be used.

Human Source Material	**Source/Supplier /Company Name**	**Is Human Source Material Known to Be Infected With An Infectious Agent? YES/NO**	**Name of Infectious Agent (If applicable)**	**HC or CFIA Containment Level (Select one)**
Human Blood (whole) or other Body Fluid		O Yes O No		O 1 O 2 O 3
Human Blood (fraction) or other Body Fluid		O Yes O No		O 1 O 2 O 3
Human Organs or Tissues (unpreserved)		O Yes O No		O 1 O 2 O 3
Human Organs or Tissues (preserved)		O Yes O No		O 1 O 2 O 3

4.0 Genetically Modified Organisms and Cell lines

4.1 Will genetic modifications be made to the microorganisms, biological agents, or cells described in Sections 1.0 and 2.0? ⊘ YES O NO If no, please proceed to Section 5.0

4.2 Will genetic modification(s) involving plasmids be done? O YES, complete table below O NO

Bacteria Used for Cloning *	Plasmid(s) *	Source of Plasmid	Gene Transfected	Describe the change that results
JM109	pcDNA3.1	INVITROGEN	1. TKGFP 2. FERRITIN 3. MAGA	1. CELL FLUORESCE 2. } IRON ACCUMULATION 3. }

* Please attach a Material Data Sheet or equivalent if available.

4.3 Will genetic modification(s) involving viral vectors be done? ☑ YES, complete table below O NO

Virus Used for Transduction *	Vector(s) *	Source of Vector	Gene Transfected	Describe the change that results
LENTIVIRUS	~~Ultrapower~~ pLenti6	INVITROGEN	Luciferase Red Fluorescent Protein	cells will fluoresce under UV light

* Please attach a Material Safety Data Sheet or equivalent.

4.4 Will genetic sequences from the following be involved?

- HIV O YES, please specify _____ ☑ NO
- HTLV 1 or 2 or genes from any Level 1 or Level 2 pathogens O YES, specify _____ ☑ NO
- SV 40 Large T antigen O YES ☑ NO
- E1A oncogene O YES ☑ NO
- Known oncogenes O YES, please specify _____ ☑ NO
- Other human or animal pathogen and or their toxins O YES, please specify _____ ☑ NO

4.5 Will virus be replication defective? ☑ YES O NO

4.6 Will virus be infectious to humans or animals? O YES ☑ NO

4.7 Will this be expected to increase the containment level required? O YES ☑ NO

5.0 Human Gene Therapy Trials

5.1 Will human clinical trials be conducted using the viral vector in 4.0? O YES ☑ NO
If no, please proceed to Section 6.0 If YES attach a full description of the make-up of the virus.

5.2 Will virus be able to replicate in the host? O YES O NO

5.3 How will the virus be administered? _____

5.4 Please give the Health Care Facility where the clinical trial will be conducted:_____

5.5 Has human ethics approval been obtained? O YES, number: _____ O NO O PENDING

6.0 Animal Experiments

6.1 Will live animals be used? ☑ YES O NO If no, please proceed to section 7.0

6.2 Name of animal species to be used MICE: C57BL6, CD1

6.3 AUS protocol # 2006-071-06 (C57BL6); 2008-117(CD1)

6.4 Will any of the agents listed be used in live animals O YES, specify: _____ ☑ NO

*** DESCRIPTION MUST BE ATTACHED TO THIS FORM OR PROJECT WILL NOT BE REVIEWED***

7.0 Use of Animal species with Zoonotic Hazards

7.1 Will any of the following animals or their organs, tissues, lavages or other body fluids including blood be used?

- Pound source dogs O YES ⊘ NO
- Pound source cats O YES ⊘ NO
- Cattle, sheep or goats O YES ⊘ NO
- Non- Human Primates O YES, please specify species _____ ⊘ NO
- Wild caught animals O YES, please specify species & colony # _____ ⊘ NO
- Birds O YES ⊘ NO
- Others (wild or domestic) O YES, please specify _____ ⊘ NO

8.0 Biological Toxins

8.1 Will toxins of biological origin be used? ⊘ YES ⊘ NO If no, please proceed to Section 9.0

8.2 If YES, please name the toxin(s) ~~STREPTOZOTOCIN, GANCICLOVIR~~ Not biological toxins
Please attach information, such as a Material Safety Data Sheet, for the toxin(s) used.

8.3 What is the LD_{50} (specify species) of the toxin ~~STZ: 3000mg/kg GCV: 2000mg/kg~~ } mouse.

9.0 Insects Requiring CFIA Permits

9.1 Do you use insects that require a permit from the CFIA? O YES ⊘ NO
If no, please proceed to Section 10.0

9.2 If YES, please give the name of the species. _____

9.3 What is the origin of the insect? _____

9.4 What is the lifestage of the insect? _____

9.5 What is your intention? O Initiate and maintain colony, give location: _____
O "One-off" use, give location: _____

9.6 Please describe the risk (if any) of escape and how this will be mitigated:

_____
_____
_____

9.7 Please attach the CFIA permit.

9.8 Please describe any CFIA permit conditions:

_____
_____
_____

10.0 Plants Requiring CFIA Permits

10.1 Do you use plants that require a permit from the CFIA? O YES ☑ NO
If no, please proceed to Section 11.0

10.2 If YES, please give the name of the species. _____

10.3 What is the origin of the plant? _____

10.4 What is the form of the plant (seed, seedling, plant, tree...)? _____

10.5 What is your intention? O Grow and maintain a crop O "One-time" use

10.6 Do you do any modifications to the plant? O YES O NO
If yes, please describe: _____
_____

10.7 Please describe the risk (if any) of loss of the material from the lab and how this will be mitigated:
_____
_____
_____

10.8 Is the CFIA permit attached? O YES O NO

10.9 Please describe any CFIA permit conditions:
_____
_____
_____

11.0 Import Requirements

11.1 Will the agent be imported? ☑ YES, please give country of origin USA O NO
If no, please proceed to Section 10.0

11.2 Has an Import Permit been obtained from HC for human pathogens? ☑ YES O NO

11.3 Has an import permit been obtained from CFIA for animal pathogens? ☑ YES O NO

11.4 Has the import permit been sent to OHS? ☑ YES, please provide permit # A-2007-00178-4 O NO

12.0 Training Requirements for Personnel Named on Form

All personnel named on the above form who will be using any of the above named agents are required to attend the following training courses given by OHS:

- Biosafety
- Laboratory and Environmental/Waste Management Safety
- WHMIS (Western or equivalent)
- Employee Health and Safety Orientation

As the Principal Investigator, I have ensured that all of the personnel named on the form who will be using any of the biohazardous agents in Sections 1.0 to 9.0 have been trained.

SIGNATURE _____

*** DESCRIPTION MUST BE ATTACHED TO THIS FORM OR PROJECT WILL NOT BE REVIEWED***

13.0 Containment Levels

11.1 For the work described in sections 1.0 to 9.0, please indicate the highest HC or CFIA Containment Level required. O 1 ⊘ 2 O 3

13.2 Has the facility been certified by OHS for this level of containment?
⊘ YES, permit # if on-campus B10-LHRI-0048 PHAC CL 2, CL2+ review completed March 23/09 previous review 30/06/2005
O NO
O NOT REQUIRED

14.0 Procedures to be Followed

14.1 As the Principal Investigator, I will ensure that this project will follow the Western Biosafety Guidelines and Procedures Manual for Containment Level 1 & 2 Laboratories (and the Level 3 Facilities Manual for Level 3 projects). I will ensure that UWO faculty, staff and students have an up-to-date Position Hazard Communication Form, found at http://www.wph.uwo.ca/

SIGNATURE S. Mawhinney Date: 02/03/09

15.0 Approvals

UWO Biohazard Subcommittee: SIGNATURE: _____
Date: _____

Safety Officer for Institution where experiments will take place: SIGNATURE: _____
Date: March 23, 2009

Safety Officer for University of Western Ontario (if different from above): SIGNATURE: _____
Date: _____

Approval Number: _____ Expiry Date (3 years from Approval): _____

Special Conditions of Approval:

*** DESCRIPTION MUST BE ATTACHED TO THIS FORM OR PROJECT WILL NOT BE REVIEWED***

Funding Source and Program Name: Canadian Diabetes Association Grant-in-Aid
Project Title: Functional molecular imaging of the pancreatic islet
Principal Investigator: Savita Dhanvantari
Hours/week: 25
Budgetary Overlap: 0
Status: Funded, July 2007-June 2009 Grant # OG-2-07-2424-SD
Renewable: Yes

Concept of the project:
This grant is addressing the following question: Can we image *endogenous* islets non-invasively? To this end, we are transfecting pancreatic cell lines with genes that encode contrast agents for PET, MRI and bioluminescence. Cells will be visualized with the appropriate imaging modality to test the viability of the tranfected genes. For PET and bioluminescence imaging, we will make a transgenic mouse in which both a PET and bioluminescence reporter gene will be expressed in the beta cell via the mouse insulin 1 promoter. This mouse model will be used to study islet regeneration non-invasively. This CDA grant is focusing on using our imaging capabilities to visualize beta cell *regeneration* in vivo, by imaging mechanisms of regeneration and regenerative therapies.

Employees paid: Rebecca McGirr, research technician (full-time)
Tumelo Moleko, MSc student

SUMMARY: Functional Molecular Imaging of the Pancreatic Islet

Rationale: In this proposal, we describe the development of cell and animal models in which the effects of regenerative/anti-apoptotic therapy can be imaged *in vivo*, using imaging modalities that can detect cellular and molecular function. Currently, the only way to visualize the effects of exogenous therapy on the reversal of islet apoptosis is through histological examination of the islets, and measurements of blood glucose and insulin levels. In order to advance our understanding of the mechanisms underlying islet apoptosis and regeneration, and to more precisely assess the effects of regenerative therapy, there is a critical need for real-time readouts of regeneration in the living animal. This proposal will focus on continuing our development of *in vivo* imaging technologies so that we can detect islet apoptosis and regeneration in live cells and small animal models. The outcome of these studies will be the establishment of both an *in vitro* and a pre-clinical model in which islet regenerative therapies can be evaluated over time.

HYPOTHESES: **1.** GLP-1-induced beta cell regeneration can be quantitatively imaged *in vivo* using PET. **2.** Mechanisms of beta cell apoptosis and regeneration can be imaged in live cells. **3.** An endogenous MR contrast agent will be synthesized by cells expressing the bacterial gene *magA*.

EXPERIMENTAL DESIGN

1. Imaging endogenous beta cell regeneration using PET. A transgenic mouse model of beta cell regeneration that can be imaged by PET will be generated. This will involve generating a mouse that expresses a HSV1-thymidine kinase-luciferase fusion gene under the control of the mouse insulin 1 promoter. We will use a known model of inducing beta cell apoptosis using STZ in neonatal mice, and then administer GLP-1 to induce islet regeneration. We will image changes in beta cell mass over time.

2. Developing a high-throughput imaging platform to assess islet regenerative therapies. INS-1 cells expressing firefly luciferase will be treated with agents that induce apoptosis and subsequently with GLP-1 to induce regeneration. Live cells will be imaged using bioluminescence. For imaging mechanisms of apoptosis and regeneration, cells will be transfected with caspase-3- and Akt-sensitive luciferase reporters. Bioluminescence will be correlated to GLP-1-induced changes in these apoptosis signaling molecules.

3. To develop a transgene reporter for imaging islets using MRI. A bacterial magnetosome gene, MagA, will be transfected into INS-1 cells. We will determine if MagA can cause beta cells to retain iron. Cells will be imaged with 3 Tesla MRI to determine if intracellular iron retention caused by MagA expression results in the synthesis of an endogenous MR contrast agent.

RELEVANCE TO DIABETES: Imaging technologies will enable us to directly visualize changes in beta cell mass during the progression of diabetes. Our work will result in the development of preclinical and high-throughput models in which treatments to improve beta cell mass in diabetes can be screened using *in vivo* imaging.

Funding Source and Program Name: NSERC Discovery Grant
Project Title: Role of the prohormone convertases in pancreatic alpha cell function.
Hours/week: 10
Budgetary Overlap: 2%
Status: Funded, June 2005-2010 Grant #R3715A01
Renewable: Yes

Concept of the project:
We wish to develop a research program investigating the sorting of proglucagon to the regulated secretory pathway. We hypothesize that the mechanism of sorting involves a sorting receptor and a sorting signal on proglucagon. Since the processing enzymes CPE and PC1 have been shown to direct some prohormones to the regulated secretory pathway, we will examine the roles of these proteins, as well as that of PC2, in the trafficking of proglucagon. Additionally, we wish to identify a sorting signal on proglucagon, which may take the form of an alpha helix. Mutational analysis followed by pulse-chase and immunoprecipitation, as well as receptor binding experiments, will be used to determine such a sorting signal. Finally, we will conduct microarray experiments on an alpha cell line transfected with PC1 to determine if the expression of PC1 can act as a developmental brake on the maturation and differentiation of the alpha cell. There is no conceptual overlap with the present CDA application.

Budgetary relationship:
There will be an approximately 2% overlap due to the purchase of RIA kits and reagents for Western blots.

Employees paid: ~~none~~ MARK MIGUELS

SUMMARY: Role of the Prohormone Convertases in Pancreatic Alpha Cell Function

OBJECTIVE: To examine the roles of prohormone convertases in the post-translational processing of proglucagon, the targeting of proglucagon to the regulated secretory pathway, and in the maturation and maintenance of the regulated secretory pathway of the pancreatic alpha cell.

RATIONALE FOR THE HYPOTHESIS: Proglucagon is expressed in the alpha cells of the pancreas, the L cells of the intestine and in select neurons of the brainstem. In the alpha cell, proglucagon is cleaved by the processing enzyme PC2 to produce glucagon. In contrast, PC1 processes proglucagon to glucagon-like peptide (GLP)-1 and GLP-2 in the intestine and brain. In addition to processing, PC1 and PC2 may be able to target prohormones to the regulated secretory pathway in endocrine cells. Finally, the expression of PC1 and the loss of expression of PC2 in alpha cells are correlated with a loss of differentiated phenotype. Therefore, PC1 and PC2 may have diverse functions in the pancreatic alpha cell.

HYPOTHESES: 1) Overexpression of PC1 in alphaTC1-6 cells lacking PC2 will result in the production of the intestinal proglucagon-derived peptides. **2)** The sorting of proglucagon to the regulated secretory pathway requires interaction of a sorting signal with the membrane-bound form of PC1 or PC2. **3)** Overexpression of PC1 will result in an immature alpha cell phenotype. Therefore, PC1 may act as a developmental brake during alpha cell maturation.

EXPERIMENTAL PLAN:

1. The processing of proglucagon by PC1 in the alpha cell. We will stably transfect PC1 into a clone of alphaTC1-6 cells that lack PC2. Production of proglucagon-derived peptides will be determined by high pressure liquid chromatography followed by radioimmunoassay, and by Western blot analysis.

2. Identification of a sorting signal on proglucagon. The sequence and structure of glucagon suggests two possible sorting signals: the alpha-helical structure of glucagon, and an internal pair of basic amino acids that does not get cleaved. We will mutate these structures and examine the effect on the sorting of proglucagon in a non-glucagon-expressing cell line.

3A. Interaction of proglucagon with membrane-bound forms of prohormone convertases. Neuro2A and PC12 cells stably expressing proglucagon will be generated. The function of the lipid-binding C-terminal tail of PC1 and PC2 in the sorting of proglucagon will be assessed with mutants that: a) lack the C-terminal tail; or b) contain only the transmembrane domain fused to the rest of the enzyme. Binding assays will determine the binding kinetics of proglucagon to PC1 and PC2.

3B. Role of the PCs in the sorting of proglucagon in alphaTC1-6 and GLUTag cells. The trafficking of proglucagon to the regulated secretory pathway will be determined in: a) alphaTC1-6 cells that lack PC2, and b) GLUTag cells that lack PC1. Cells will then be transfected with the constructs described in 2A, and the sorting of proglucagon will be assessed by metabolic labeling and immunoprecipitation.

4) Determining changes in the secretory protein expression profile in alphaTC1-6 cells after PC1 overexpression. Microarray analysis coupled with SELDI-TOF-MS will be used to compare the regulated secretory protein profile between wild-type alphaTC1-6 cells, cells lacking PC2, and cells overexpressing PC1. We will also engineer a mouse expressing PC1 under the control of the alpha cell-specific region of the proglucagon promoter. Together, the cell and mouse models will demonstrate the importance of the PCs in the development and maintenance of the neuroendocrine phenotype of the alpha cell.

SIGNIFICANCE: From the proposed studies, we will elucidate the roles of PC1 and PC2 in the processing and trafficking of proglucagon, and in the maturation of the regulated secretory pathway of the alpha cell. Identification of role of the PCs in mediating alpha cell function will provide critical insight into the processes regulating islet growth and differentiation.

Funding Source and Program Name: NSERC Collaborative Health Research Program
Project Title: Development of GLP-1 receptor probes for imaging changes in beta cell mass
Hours/week: 20
Budgetary Overlap: 0
Status: Funded, May 2008-April 2011
Renewable: no

Concept of the project:
This Innovation proposal will focus on the development of a novel radiolabelled probe for imaging using SPECT (single photon emission computed tomography) and PET (positron emission tomography) that can bind to GLP-1 receptors on the beta cell. We will develop radiometal chelates from peptides that bind to the GLP-1 receptor: GLP1, exendin-4 and exendin 9-39. All peptide conjugates will be tested for their ability to bind the GLP-1 receptor in INS-1 and CHO-GLP-1R cells. Chelates will be radiolabelled with either ^{111}In (SPECT) or ^{68}Ga (PET) and cells will be scanned with the appropriate imaging modality to test the imaging capability of the probes.

Budgetary relationship:
There is no budgetary overlap.

Employees paid: Vanessa Rota, graduate student

SUMMARY: Development of GLP-1 receptor probes for imaging changes in beta cell mass

Objectives of proposed research program

In order to advance our understanding of the changes in beta cell mass before the onset of diabetes and during the progression of Type 1 and Type 2 diabetes, there is a critical need for imaging probes that can detect beta cells in the living animal. This Collaborative Health Research Project proposes to develop a novel targeted probe for imaging using SPECT (single photon emission computed tomography) and PET (positron emission tomography) that can bind to GLP-1 receptors on the beta cell. The objectives of our proposal are:

1. To design and synthesize glucagon-like peptide-1 (GLP-1) peptide derivatives that can be labeled with a metal radionuclide.
2. To screen peptide-metal chelates for binding to the GLP-1 receptor on beta cells.
3. To apply this technology for the in vitro imaging of isolated pancreatic islets.
4. To apply this technology for the in vivo imaging of islet regeneration using SPECT/CT and PET in a mouse model of diabetes.

This project is a highly inter-disciplinary collaboration between investigators in the natural sciences (Chemistry and Molecular Biology) and the health/medical sciences (Diabetes and Imaging Sciences).

Experimental Plan

1. To design and synthesize GLP-1 (7-36) derivatives for labelling with a metal radionuclide - Derivatives of GLP-1 (7-36) and exendin-4 where a pendant metal chelator is incorporated as part of the peptide structure will be designed. Dr. Luyt is a synthetic chemist and provides a science/engineering component of the project. The following DOTA conjugated peptides will be synthesized using automated solid-phase Fmoc chemistry: GLP-1 (7-36NH2); a degradation-resistant form of GLP-1, GLP-1 (D-Ala2); the GLP-1 receptor agonist, exendin-4; and the antagonist, exendin 9-39. These peptides derivatives are capable of coordinating to the SPECT radionuclide 111In or the PET radionuclide 68Ga. All peptides will be analyzed and purified using HPLC with characterization by ESI or MALDI mass spectrometry.

2. To identify a suitable imaging probe by screening peptide-metal chelates for GLP-1 receptor binding and activation - Peptide-metal chelates will be screened for their ability to bind to the GLP-1 receptor by performing receptor binding assays on the cell line CHO-GLP-1R. Dr. Dhanvantari is an NSERC supported expert in the cell biology of diabetes and is another science/engineering member of the team.

3: To image beta cell lines and isolated islets with our novel probe - Peptide-metal chelates will be complexed to the radioactive isotope of indium, 111In, and incubated with cells and islets prior to planar imaging with a gamma camera to confirm the imaging capabilities of the peptide probe. For PET imaging, cells/islets will be incubated with 68Ga-peptide conjugate and imaged on a preclincal PET scanner.

4. To quantitatively image islet destruction and regeneration in rodent models of Type 1 diabetes - Using dedicated preclinical SPECT/CT and PET scanners, we will evaluate the ability of the most promising GLP-1 probes to non-invasively image changes in beta cell mass in vivo in a mouse model of diabetes. Mice will be rendered diabetic through low-dose streptozotocin treatment, and then treated with growth factors known to play a role in beta cell regeneration. During diabetes and therapy, mice will be systemically administered our GLP-1 probe and imaged using SPECT or PET, with the preferred modality being determined based on appropriate matching of isotope half-life and in vivo behaviour. In this way, we will be able to longitudinally assess the effects of regenerative therapy in diabetes.

Novelty and Expected Significance:

This project describes: 1) a combined synthetic chemistry/cell biology approach that will lead to the discovery of a novel molecular imaging probe targeting the GLP-1 receptor; and 2) the application of the probe to the medical imaging of changes in beta cell mass during the progression of diabetes. A successful probe would be further investigated for clinical usefulness, thereby permitting the clinical evaluation of changes in beta cell mass during the progression of diabetes.

Canadian Food Inspection Agency
Government of Canada

Agence canadienne d'inspection des aliments
Gouvernement du Canada

Permit No./N° de permis:
A-2007-00178-4
ORIGINAL
2007/01/12
year/mo/day
année/mois/jour

IMPORT PERMIT PERMIS D'IMPORTATION

THIS PERMIT IS ISSUED PURSUANT TO:/CE PERMIS EST DÉLIVRÉ CONFORMÉMENT A:

THE HEALTH OF ANIMALS ACT AND REGULATIONS/LOI ET RÈGLEMENT SUR LA SANTÉ DES ANIMAUX

Importer/Importateur
LAWSON RESEARCH INSTITUTE

268 GROSVENOR STREET, ROOM H417
LONDON, ONTARIO
N6A4V2

Contact: Dr. Savita Dhanvantari Applicant Name: DR. SAVITA DHANVANTARI
Phone: (519) 646-6100 ext. 65738 Fax: (519) 646-6110

Exporter/Exportateur
INVITROGEN CORPORATION INC.

1600 FARADAY AVENUE
CARLSBAD CALIFORNIA
UNITED STATES
440190
Contact: Mike Galleno

Phone: (760) 603-7219 Fax: (760) 602-6519

Quarantine/Destination/Quarantaine

Producer/Producteur

Valid/Valide **from/du** 2007/01/12 year/month/day année/mois/jour **to/au** 2008/01/31 year/month/day année/mois/jour

Country of Origin/ Pays d'Origine UNITED STATES

For the entry of/ Pour l'entrée de: ____ Single shipment/Chargement simple XX Multiple shipments/Chargements multiples

Place of entry into Canada/Lieu d'entrée au Canada:
Various Ports of Entry

FOR THE IMPORTATION OF:/POUR L'IMPORTATION DE:
(Description of things(s)/Description de la ou des choses)
1. Product Description: ONE OR MORE OF THE INVITROGEN LENTIVIRAL PRODUCTS LISTED ON THE ATTACHMENT TITLED "ATTACHMENT TO ANIMAL PATHOGEN(S) IMPORT PERMIT # A-2007-00178-4.

(TO BE USED IN ROOM 4-508, CULTURE ROOM F4-127A, LAWSON HEALTH RESEARCH INSTITUTE, LONDON, ON.) Proposed End Use: "In Vitro" Scientific Name: Biocontainment Level: 2

A PERSON WHO IMPORTS A THING UNDER THIS PERMIT SHALL COMPLY WITH ALL THE CONDITIONS SET OUT HEREIN/TOUTE PERSONNE QUI IMPORTE UNE CHOSE EN VERTU DE CE PERMIS DEVRA RESPECTER TOUTES LES CONDITIONS DÉCRITES CI-DESSOUS

Selected Conditions / Conditions Choisies

ONE OR MORE OF THE INVITROGEN LENTIVIRAL PRODUCTS LISTED ON THE ATTACHMENT TITLED "ATTACHMENT TO ANIMAL PATHOGEN(S) IMPORT PERMIT # A-2007-00178-4.

(TO BE USED IN ROOM 4-508, CULTURE ROOM F4-127A, LAWSON HEALTH RESEARCH INSTITUTE, LONDON, ON.)

1. The original or a copy of the signed original of this permit and any other necessary import / export documentation pertaining to the shipment of animal(s) or thing(s) must be provided for inspection at the first port of entry or to a Canadian Food Inspection Agency Import Service Center.

2. The conditions in this permit can only be changed or amended by a CFIA inspector. Any change to the permit by an unauthorized person will render the permit invalid.

Canada

Canadian Food Inspection Agency
Goverment of Canada

Agence canadienne d'inspection des aliments
Gouvernement du Canada

Permit No./N° de permis:
A-2007-00178-4
ORIGINAL
2007/01/12
year/mo/day
année/mois/jour

IMPORT PERMIT PERMIS D'IMPORTATION

THIS PERMIT IS ISSUED PURSUANT TO:/CE PERMIS EST DÉLIVRÉ CONFORMÉMENT A:

THE HEALTH OF ANIMALS ACT AND REGULATIONS/LOI ET RÈGLEMENT SUR LA SANTÉ DES ANIMAUX

Importer/Importateur	Exporter/Exportateur
LAWSON RESEARCH INSTITUTE 268 GROSVENOR STREET, ROOM H417 LONDON, ONTARIO N6A4V2 Contact: Dr. Savita Dhanvantari Applicant Name: DR. SAVITA DHANVANTARI Phone: (519) 646-6100 ext. 65738 Fax: (519) 646-6110	INVITROGEN CORPORATION INC. 1600 FARADAY AVENUE CARLSBAD CALIFORNIA UNITED STATES 440190 Contact: Mike Galleno Phone: (760) 603-7219 Fax: (760) 602-6519

Selected Conditions / Conditions Choisies (Continued/Suite)

3. The imported material must be packaged in appropriate shipping containers to prevent accidental spillage of contents during shipping. Importers should be aware of their obligations under Transport Canada's regulations concerning transportation of dangerous goods.

4. All infectious material must be handled in appropriate animal pathogen containment level 2 facilities as described in Containment Standards for Veterinary Facilities, 1996, AAFC publication no. 1921.

5. The material authorized for importation by this permit is to be used in in vitro studies ONLY and must not to be introduced into laboratory, domestic or wild animals (including birds or fish) unless written authorization is obtained from the Canadian Food Inspection Agency.

6. The animal(s) or thing(s) imported under this permit must not be removed from the premises of destination listed on this permit, unless written authorization is obtained from the Canadian Food Inspection Agency.

7. Upon completion of the tests or experiments, the imported material as described on this permit and any derivatives thereof must be autoclaved, incinerated or alternatively disposed of in a manner approved by an inspector of the Canadian Food Inspection Agency.

8. Records pertaining to the imported product's use, storage and disposal must be maintained for two (2) years following importation. These records must be made available for inspection by the Canadian Food Inspection Agency upon request.

9. The importer is responsible for all costs incurred or associated with any testing or treatment of the animal(s) or thing(s) that may be required under the import permit or under the authority of the Health of Animals Act or the Health of Animals Regulations. The importer shall pay all fees for services required in respect of the importation under the National Animal Health Program Cost Recovery Fees Regulations in place at the time of importation.

10. Consideration of an application necessary for issuance of a permit to import the described animal or thing is subject to Class 1 fees.

11. The issuance of this permit does not relieve the owner or the importer of the obligation to comply with any other relevant federal, provincial or municipal legislation or requirement.

12. Failure to comply with the conditions contained in this permit or with the provisions of the Health of Animals Act and Regulations may result in the cancellation of this permit and will result in the forfeiture to the Crown of the imported thing(s) or in the removal of the thing(s) from Canada, all without compensation to, and at the expense of the importer. The importer(s) are responsible for the imported thing(s), their freedom from extraneous disease, active or latent, and genetic or other defects. The importer, his heirs, executors, successors and assigns release and discharges Her Majesty the Queen in right of Canada and the CFIA of and from all claims and demands, damages, actions or causes of action arising or to arise by reason of the importation of the thing(s) and agrees to indemnify and save harmless Her Majesty the Queen in right of Canada and the CFIA from and against all actions, damages, claims

Canada

Canadian Food Inspection Agency
Goverment of Canada

Agence canadienne d'inspection des aliments
Gouvernement du Canada

Permit No./N° de permis:
A-2007-00178-4
ORIGINAL
2007/01/12
year/mo/day
année/mois/jour

IMPORT PERMIT PERMIS D'IMPORTATION

THIS PERMIT IS ISSUED PURSUANT TO:/CE PERMIS EST DÉLIVRÉ CONFORMÉMENT A:

THE HEALTH OF ANIMALS ACT AND REGULATIONS/LOI ET RÈGLEMENT SUR LA SANTÉ DES ANIMAUX

mporter/Importateur	**Exporter/Exportateur**
.AWSON RESEARCH INSTITUTE .68 GROSVENOR STREET, ROOM H417 .ONDON, ONTARIO N6A4V2 Contact: Dr. Savita Dhanvantari Applicant Name: DR. SAVITA DHANVANTARI Phone: (519) 646-6100 ext. 65738 Fax: (519) 646-6110	INVITROGEN CORPORATION INC. 1600 FARADAY AVENUE CARLSBAD CALIFORNIA UNITED STATES 440190 Contact: Mike Galleno Phone: (760) 603-7219 Fax: (760) 602-6519

Selected Conditions / Conditions Choisies (Continued/Suite)

and demands which may be brought in respect of or arising out of the importation of such thing(s), any contamination with extraneous disease or other defects.

3. This permit is conditional upon a permit being obtained under the Human Pathogens Importation Regulations to import the pathogenic material and upon that import permit being produced and valid when the above pathogenic material is presented to an inspector for inspection at the time of importation.

Additional Conditions Additionnelles

ONE OR MORE OF THE INVITROGEN LENTIVIRAL PRODUCTS LISTED ON THE ATTACHMENT TITLED "ATTACHMENT TO ANIMAL PATHOGEN(S) IMPORT PERMIT # A-2007-00178-4.

TO BE USED IN ROOM 4-508, CULTURE ROOM F4-127A, LAWSON HEALTH RESEARCH INSTITUTE, LONDON, ON.)

1. No culturing of containment level 3 or 4 pathogens shall be done.

Cinthia Labrie

Authorized By:/Approuvé par:
CINTHIA LABRIE

For the Minister of Agriculture and Agri-Food
Pour le ministre d'agriculture et agroalimentaire

The information is required by (for) the Canadian Food Inspection Agency for the purpose of verifying import products. Information may be accessible or protected as required under the provisions of the Access to Information Act.

CFIA / ACIA 5067 (S8/04)

Canada

Canadian Food Inspection Agency
Agence canadienne d'inspection des aliments

Office of Biohazard Containment and Safety
Science Advice and Biohazards Division
Science Strategies Directorate, CFIA
159 Cleopatra Drive, Ottawa, Ontario K1A 0Y9
Tel: (613) 221-7068 Fax: (613) 228-6129
Email: ImportZoopath@inspection.gc.ca

Bureau du confinement des biorisques et sécurité
Division des avis scientifiques et contrôle des biorisques
Direction des stratégies scientifiques, ACIA
159 promenade Cleopatra, Ottawa, Ontario K1A 0Y9
Tél: (613) 221-7068 Téléc: (613) 228-6129
Courriel: ImportZoopath@inspection.gc.ca

ATTACHMENT TO ANIMAL PATHOGEN(S) IMPORTATION PERMIT
ATTACHEMENT AU PERMIS D'IMPORTATION D'AGENTS ZOOPATHOGÈNES
#A-2007-00178-4

Issued to/ Délivré à: Dr. Savita Dhanvantari, Lawson Health Research Institute,
268 Grosvenor Street, London ON N6A 4V2.

Includes the following animal pathogen containment Level 2 microorganisms:
Inclut les agents zoopathogènes de niveau de confinement 2 suivant:

Invitrogen Lentiviral Products / Produits Lentiviral d'Invitrogen:

- PCDNA6.2/C-EMGFP-GW/TOPO (K35920)
- PCDNA6.2/N-EMGFP-GW/TOPO (K36020)
- PCDNA6.2/C-YFP-GW/TOPO (K36120)
- PCDNA6.2/N-YFP-GW/TOPO (K36620)
- Virapower II Lenti GW System (K36720)
- Virapower II Lenti C-Lumio system (K37020)
- Virapower II Lenti N-Lumio system (K37120)
- POL III MIR Rnai Vector (K493500)
- POL II MIR Rnai GFP Vector (K493600)
- Lenti POL II MIRE Rnai Vector (K493700)
- Lenti POL II MIRE Rnai w/GFP (K493800)
- Block it Lenti RNAi Expression system (K494400)
- Virapower Lentiviral directional (K495000)
- Virapower Lentviral Gateway (K496000)
- Lentiviral T Rex Expression system (K496500)
- Virapower packaging mix (K497500)
- Virapower Zeo Lenti Expression (K498000)
- Virapower Zeo Lentiviral Support Kit (K498500)
- Virapower UBC Lenti expression (K499000)
- Virapower Lentiviral support (K497000)
- Plenti6/Block it RNAi vector (K494300)
- Plenti 6/V5 Directional TOPO (K495510)
- VP TR GW Vector kit (K496700)
- PCDNA6.2/EMGFP-BSD/V5 Dest (V36620)
- Plenti6.2/V5-DEST GW vector (V36820)
- Plenti6.2-GW/EMGFP Exp vector (V36920)
- Plenti6/TR vector (V48020)
- Block-iT Lenti RNAi ZW GW Vector (V48820)
- Plenti6/V5 Gtwy vector pack (V49610)
- Plenti4/V5 -Dest Gateway vector (V49810)
- Plenti6/UBC/V5 Dest vector (V49910)
- Block it Lentiviral Inducible RNAi (K492500)
- Promotorless Lenti Exp kit (K591000)

The above products may contain one or more of the following components / Les produits ci-dessus peuvent contenir un ou plusiers des composants suivants:

Plenti6/Block it Dest RNAi , PLP1, PLP2, PLP3/VSVG, Plenti6/V5-Dtopo, Plenti6/V5-GW/LacZ, plenti6/V5 Dest vector, plenti6/TR, plenti4/TO/V5 Dest, plenti4/TO/V5-GW/LacZ, plenti 4/V5 Dest, plenti4/V5 -GW/LacZ vector, plenti4/Blockit Dest, plenti6/UBC/ V5 Dest vector, plenti6/UBC/V5-GW/LacZ vector, plenti6/R4R2/V5-Dest, 293 FT cells, PCDNA6.2-GW/MIR Neg TB, PCDNA6.2-GW-EMGFP-MIR Neg, Plenti6.2/C-Lumio/V5 DEST,Plenti 6.2/C-Lumio-V5-GW/LA, Plenti6.2/N-Lumio/V5 Dest, Plenti6.2/N-Lumio/V5-GW/LA, Plenti6.2-GW/EMGFP Kit, Plenti 6.2.V5 Dest Kit, Plenti6.2/V5-GW LacZ, PCDNA6.2/EMGFP-BSD/V5 Dest, PCDNA6.2/EMGFP-BSD/V5-GW/C, PCDNA6.2/C-EMGFP-GW, PCDNA6.2/C-EMGFP-GW/CAT, PCDNA6.2/N-EMGFP-GW, PCDNA6.2/N-EMGFP-GW, PCDNA6.2/C-YFP-GW, PCDNA6.2 C-YFP-GW/CAT, PCDNA6.2/N-YFP-GW, PCDNA6.2/N-YFP-GW-CAT.

REVISED: May 01, 2006.

Cinthia Labrie

Cinthia Labrie
A/Chief, Animal Pathogen Importation Program/
Chef intérimaire, Programme d'importation des agents zoopathogènes

Jan 12/07
Date

Canada

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Material Safety Data Sheet

Revision Date: 03-Mar-2006

1. IDENTIFICATION OF THE SUBSTANCE/PREPARATION AND THE COMPANY/UNDERTAKING

Product code 351275
Product name VIRAPOWER PKG. MIX 195 UG, LYOPHILIZED

Contact manufacturer
INVITROGEN CORPORATON
1600 FARADAY AVENUE
PO BOX 6482
CARLSBAD, CA 92008
760-603-7200

INVITROGEN CORPORATION
2270 INDUSTRIAL STREET
BURLINGTON, ONT
CANADA L7P 1A1
800-263-6236

GIBCO PRODUCTS
INVITROGEN CORPORATION
3175 STALEY ROAD P.O. BOX 68
GRAND ISLAND, NY 14072
716-774-6700

2. COMPOSITION/INFORMATION ON INGREDIENTS

Hazardous/Non-hazardous Components
The product contains no substances which at their given concentration, are considered to be hazardous to health

3. HAZARDS IDENTIFICATION

Emergency Overview
The product contains no substances which at their given concentration, are considered to be hazardous to health.

Form
Solid

Principle Routes of Exposure/ Potential Health effects

Eyes	No information available
Skin	No information available
Inhalation	No information available
Ingestion	No information available

Specific effects

Carcinogenic effects	No information available
Mutagenic effects	No information available
Reproductive toxicity	No information available

Sensitization	No information available
Target Organ Effects	No information available

4. FIRST AID MEASURES

Skin contact	Wash off immediately with plenty of water
Eye contact	Rinse thoroughly with plenty of water, also under the eyelids.
Ingestion	Never give anything by mouth to an unconscious person
Inhalation	Move to fresh air
Notes to physician	Treat symptomatically

5. FIRE-FIGHTING MEASURES

Suitable extinguishing media	Dry chemical
Special protective equipment for firefighters	Wear self-contained breathing apparatus and protective suit

6. ACCIDENTAL RELEASE MEASURES

Personal precautions	Use personal protective equipment
Methods for cleaning up	Soak up with inert absorbent material

7. HANDLING AND STORAGE

Handling	No special handling advice required
Storage	Keep in properly labelled containers

8. EXPOSURE CONTROLS / PERSONAL PROTECTION

Occupational exposure controls

Exposure limits

Engineering measures	Ensure adequate ventilation, especially in confined areas

Personal protective equipment

Respiratory protection	In case of insufficient ventilation wear suitable respiratory equipment
Hand protection	Protective gloves
Eye protection	Safety glasses with side-shields
Skin and body protection	Lightweight protective clothing
Hygiene measures	Handle in accordance with good industrial hygiene and safety practice
Environmental exposure controls	Prevent product from entering drains

9. PHYSICAL AND CHEMICAL PROPERTIES

General Information

Form	Solid

Important Health Safety and Environmental Information

Boiling point/range	**°C** No data available	**°F** No data available
Melting point/range	**°C** No data available	**°F** No data available
Flash point	**°C** No data available	**°F** No data available
Autoignition temperature	**°C** No data available	**°F** No data available
Oxidizing properties	No information available	

Water solubility	No data available

10. STABILITY AND REACTIVITY

Stability	Stable.
Materials to avoid	No information available
Hazardous decomposition products	No information available
Polymerization	Hazardous polymerisation does not occur

11. TOXICOLOGICAL INFORMATION

Acute toxicity

Principle Routes of Exposure/ Potential Health effects

Eyes	No information available
Skin	No information available
Inhalation	No information available
Ingestion	No information available

Specific effects

Carcinogenic effects	No information available
Mutagenic effects	No information available
Reproductive toxicity	No information available
Sensitization	No information available

Target Organ Effects — No information available

12. ECOLOGICAL INFORMATION

Ecotoxicity effects	No information available.
Mobility	No information available.
Biodegradation	Inherently biodegradable.
Bioaccumulation	Does not bioaccumulate.

13. DISPOSAL CONSIDERATIONS

Dispose of in accordance with local regulations

14. TRANSPORT INFORMATION

IATA

Proper shipping name	Not classified as dangerous in the meaning of transport regulations
Hazard Class	No information available
Subsidiary Class	No information available
Packing group	No information available
UN-No	No information available

15. REGULATORY INFORMATION

International Inventories

U.S. Federal Regulations

SARA 313
Not regulated

Clean Air Act, Section 112 Hazardous Air Pollutants (HAPs) (see 40 CFR 61)
This product contains the following HAPs:

U.S. State Regulations

California Proposition 65
This product contains the following Proposition 65 chemicals:

WHMIS hazard class:
Non-controlled

This product has been classified according to the hazard criteria of the CPR and the MSDS contains all of the information required by the CPR

16. OTHER INFORMATION

This material is sold for research and development purposes only. It is not for any human or animal therapeutic or clinical diagnostic use. It is not intended for food, drug, household, agricultural, or cosmetic use. An individual technically qualified to handle potentially hazardous chemicals must supervise the use of this material.

The above information was acquired by diligent search and/or investigation and the recommendations are based on prudent application of professional judgment. The information shall not be taken as being all inclusive and is to be used only as a guide. All materials and mixtures may be present unknown hazards and should be used with caution. Since Invitrogen Corporation cannot control the actual methods, volumes, or conditions of use, the Company shall not be held liable for any damages or losses resulting from the handling or from contact with the product as described herein. THE INFORMATION IN THIS MSDS DOES NOT CONSTITUTE A WARRANTY, EXPRESS OR IMPLIED, INCLUDING ANY IMPLIED WARRANTY OF MERCHANTABILITY OR FITNESS FOR ANY PARTCULAR PURPOSE.

End of Safety Data Sheet

Description of Cell Lines and Sources.

INS-1 832/13
Dr. Chris Newgard, Duke University Medical Center

INS-1 cells were originally derived from a rat insulinoma (1). These cells were stably transfected with the human proinsulin gene and maintain expression through 66 population doublings (2). They exhibit glucose-stimulated insulin secretion and are a good model to study beta cell physiology.

αTC1-6
Dr. C. Bruce Verchere, University of British Columbia

αTC1 cells were derived from an SV40 large T antigen transgenic mouse (3). Clonal population 6 showed proglucagon gene expression and no proinsulin or prosomatostatin gene expression. These cells represent a pure population of pancreatic alpha cells and are useful for the study of alpha cell physiology.

InR1-G9
Dr. Patricia Brubaker, University of Toronto.

InR1 cells were derived from a BK-induced hamster glucagonoma. Clone G9 expresses proglucagon very strongly, and processes proglucagon to glucagon, similar to normal alpha cells (4).

GLUTag
Dr. Dan Drucker, The Toronto Hospital and University of Toronto

These cells were derived from a transgenic mouse expressing the SV40 large T antigen in the L cells of the instestine (5). They express proglucagon and process it to the intestinal peptides GLP-1 and GLP-2. These cells are a good model for the study of GLP1-synthesis and secretion (6).

Neuro2A
Dr. Y Peng Loh, National Institutes of Health, Bethesda, MD

Neuro 2A cells were derived from a mouse neuroblastoma. They do not express any prohormone processing enzymes and are ideal for the study of hormone processing and sorting (7).

CHO-GLP1R
Dr. Michael Wheeler, University of Toronto.

CHO cells are derived from Chinese hamster ovary, and these cells have been stably transfected with the human GLP-1 receptor (8). They are used for *in vitro* receptor binding assays.

PC12

Dr. Walter Rushlow, The University of Western Ontario

PC12 cells are originally derived from rat pheochromocytoma. We are using them for the study of hormone processing and sorting.

1. **Asfari M, Janjic D, Meda P, Li G, Halban PA, Wollheim CB** 1992 Establishment of 2-mercaptoethanol-dependent differentiated insulin-secreting cell lines. Endocrinology 130:167-178
2. **Hohmeier HE, Mulder H, Chen G, Henkel-Rieger R, Prentki M, Newgard CB** 2000 Isolation of INS-1-derived cell lines with robust ATP-sensitive K+ channel-dependent and -independent glucose-stimulated insulin secretion. Diabetes 49:424-430
3. **Powers AC, Efrat S, Mojsov S, Spector D, Habener JF, Hanahan D** 1990 Proglucagon processing similar to normal islets in pancreatic alpha-like cell line derived from transgenic mouse tumor. Diabetes 39:406-414
4. **Drucker DJ, Philippe J, Mojsov S** 1988 Proglucagon gene expression and posttranslational processing in a hamster islet cell line. Endocrinology 123:1861-1867
5. **Drucker DJ, Jin T, Asa SL, Young TA, Brubaker PL** 1994 Activation of proglucagon gene transcription by protein kinase-A in a novel mouse enteroendocrine cell line. Mol Endocrinol 8:1646-1655
6. **Brubaker PL, Schloos J, Drucker DJ** 1998 Regulation of glucagon-like peptide-1 synthesis and secretion in the GLUTag enteroendocrine cell line. Endocrinology 139:4108-4114.
7. **Zhang CF, Dhanvantari S, Lou H, Loh YP** 2003 Sorting of carboxypeptidase E to the regulated secretory pathway requires interaction of its transmembrane domain with lipid rafts. Biochem J 369:453-460
8. **Xiao Q, Giguere J, Parisien M, Jeng W, St-Pierre SA, Brubaker PL, Wheeler MB** 2001 Biological activities of glucagon-like peptide-1 analogues in vitro and in vivo. Biochemistry 40:2860-2869

Cell lines established in our lab.

aTC1-6 tkgfp clones 1 (low expresser) and 5 (high expresser)

INS-1 832/13 tkgfp clones 2 (low expresser) and 3 (high expresser)

GLUTag tkgfp

These cell lines have been stably transfected with a plasmid containing the coding sequence of the herpes simplex virus 1 thymidine kinase (HSV1 tk) fused in-frame to green fluorescent protein (gfp) (1). The expression of the gene is under the control of the CMV promoter. This construct was obtained from Dr. Y. Gelovani, MD Anderson Cancer Center, TX.

INS-1 832/13-Ferritin
These cells have been stably transfected with the human ferritin heavy chain fragment inserted into pcDNA3.1, which was obtained from Dr. J. Koropatnick, London Regional Cancer Program. This gene will cause iron accumulation in the cells, for the purposes of imaging the cells by MRI (2).

INS-1 832/13-MagA
These cells have been stably transfected with the bacterial gene, MagA, inserted into pcDNA3.1. This plasmid was constructed in our lab. This gene will cause iron accumulation in the cells, for the purposes of imaging by MRI (3).

1. **Jacobs A, Dubrovin M, Hewett J, Sena-Esteves M, Tan CW, Slack M, Sadelain M, Breakefield XO, Tjuvajev JG** 1999 Functional coexpression of HSV-1 thymidine kinase and green fluorescent protein: implications for noninvasive imaging of transgene expression. Neoplasia 1:154-161
2. **Cohen B, Ziv K, Plaks V, Israely T, Kalchenko V, Harmelin A, Benjamin LE, Neeman M** 2007 MRI detection of transcriptional regulation of gene expression in transgenic mice. Nat Med 13:498-503
3. **Goldhawk D, Lemaire C, McCreary C, McGirr R, Dhanvantari S, Thompson R, Figueredo R, Koropatnick J, Foster P, Prato F** 2009 Magnetic Resonance Imaging of Cell Overexpressing MagA, an Endogenous Contrast Agent for Live Cell Imaging. Mol Imaging in press

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Material Safety Data Sheet

Revision Date: 17-Apr-2006

1. IDENTIFICATION OF THE SUBSTANCE/PREPARATION AND THE COMPANY/UNDERTAKING

Product code 350484
Product name pcDNA3.1/(+)

Contact manufacturer
INVITROGEN CORPORATON
1600 FARADAY AVENUE
PO BOX 6482
CARLSBAD, CA 92008
760-603-7200

INVITROGEN CORPORATION
2270 INDUSTRIAL STREET
BURLINGTON, ONT
CANADA L7P 1A1
800-263-6236

GIBCO PRODUCTS
INVITROGEN CORPORATION
3175 STALEY ROAD P.O. BOX 68
GRAND ISLAND, NY 14072
716-774-6700

2. COMPOSITION/INFORMATION ON INGREDIENTS

Hazardous/Non-hazardous Components
The product contains no substances which at their given concentration, are considered to be hazardous to health

3. HAZARDS IDENTIFICATION

Emergency Overview
The product contains no substances which at their given concentration, are considered to be hazardous to health.

Form
Solid

Principle Routes of Exposure/ Potential Health effects

Eyes	No information available
Skin	No information available
Inhalation	No information available
Ingestion	No information available

Specific effects

Carcinogenic effects	No information available
Mutagenic effects	No information available
Reproductive toxicity	No information available

Sensitization	No information available
Target Organ Effects	No information available

4. FIRST AID MEASURES

Skin contact	Wash off immediately with plenty of water
Eye contact	Rinse thoroughly with plenty of water, also under the eyelids.
Ingestion	Never give anything by mouth to an unconscious person
Inhalation	Move to fresh air
Notes to physician	Treat symptomatically

5. FIRE-FIGHTING MEASURES

Suitable extinguishing media	Dry chemical
Special protective equipment for firefighters	Wear self-contained breathing apparatus and protective suit

6. ACCIDENTAL RELEASE MEASURES

Personal precautions	Use personal protective equipment
Methods for cleaning up	Soak up with inert absorbent material

7. HANDLING AND STORAGE

Handling	No special handling advice required
Storage	Keep in properly labelled containers

8. EXPOSURE CONTROLS / PERSONAL PROTECTION

Occupational exposure controls

Exposure limits

Engineering measures	Ensure adequate ventilation, especially in confined areas

Personal protective equipment

Respiratory protection	In case of insufficient ventilation wear suitable respiratory equipment
Hand protection	Protective gloves
Eye protection	Safety glasses with side-shields
Skin and body protection	Lightweight protective clothing
Hygiene measures	Handle in accordance with good industrial hygiene and safety practice
Environmental exposure controls	Prevent product from entering drains

9. PHYSICAL AND CHEMICAL PROPERTIES

General Information

Form	Solid

Important Health Safety and Environmental Information

Boiling point/range	°C No data available	°F No data available
Melting point/range	°C No data available	°F No data available
Flash point	°C No data available	°F No data available
Autoignition temperature	°C No data available	°F No data available
Oxidizing properties	No information available	

Water solubility	No data available

10. STABILITY AND REACTIVITY

Stability	Stable.
Materials to avoid	No information available
Hazardous decomposition products	No information available
Polymerization	Hazardous polymerisation does not occur

11. TOXICOLOGICAL INFORMATION

Acute toxicity

Principle Routes of Exposure/ Potential Health effects

Eyes	No information available
Skin	No information available
Inhalation	No information available
Ingestion	No information available

Specific effects

Carcinogenic effects	No information available
Mutagenic effects	No information available
Reproductive toxicity	No information available
Sensitization	No information available

Target Organ Effects — No information available

12. ECOLOGICAL INFORMATION

Ecotoxicity effects	No information available.
Mobility	No information available.
Biodegradation	Inherently biodegradable.
Bioaccumulation	Does not bioaccumulate.

13. DISPOSAL CONSIDERATIONS

Dispose of in accordance with local regulations

14. TRANSPORT INFORMATION

IATA

Proper shipping name	Not classified as dangerous in the meaning of transport regulations
Hazard Class	No information available
Subsidiary Class	No information available
Packing group	No information available
UN-No	No information available

15. REGULATORY INFORMATION

International Inventories

U.S. Federal Regulations

SARA 313
Not regulated

Clean Air Act, Section 112 Hazardous Air Pollutants (HAPs) (see 40 CFR 61)
This product contains the following HAPs:

U.S. State Regulations

California Proposition 65
This product contains the following Proposition 65 chemicals:

WHMIS hazard class:
Non-controlled

This product has been classified according to the hazard criteria of the CPR and the MSDS contains all of the information required by the CPR

16. OTHER INFORMATION

This material is sold for research and development purposes only. It is not for any human or animal therapeutic or clinical diagnostic use. It is not intended for food, drug, household, agricultural, or cosmetic use. An individual technically qualified to handle potentially hazardous chemicals must supervise the use of this material.

The above information was acquired by diligent search and/or investigation and the recommendations are based on prudent application of professional judgment. The information shall not be taken as being all inclusive and is to be used only as a guide. All materials and mixtures may be present unknown hazards and should be used with caution. Since Invitrogen Corporation cannot control the actual methods, volumes, or conditions of use, the Company shall not be held liable for any damages or losses resulting from the handling or from contact with the product as described herein. THE INFORMATION IN THIS MSDS DOES NOT CONSTITUTE A WARRANTY, EXPRESS OR IMPLIED, INCLUDING ANY IMPLIED WARRANTY OF MERCHANTABILITY OR FITNESS FOR ANY PARTCULAR PURPOSE.

End of Safety Data Sheet

Promega

E. coli Competent Cells

All technical literature is available on the Internet at: www.promega.com/tbs/
Please visit the web site to verify that you are using the most current version of this Technical Bulletin. Please contact Promega Technical Services if you have questions on use of this system. E-mail: techserv@promega.com

I. Description

The *E. coli* Competent Cells are prepared according to a modified procedure of Hanahan (1). The competent cells can be used for many standard molecular biology applications. Competent cells of strains HB101 and JM109 are available for convenient transformation in two efficiencies: High Efficiency at greater than 10^8cfu/µg and Subcloning Efficiency at greater than 10^7cfu/µg. JM109 cells (2) are an ideal host for many molecular biology applications. HB101 cells (3) are useful for cloning in vectors that do not require α-complementation for blue/white screening. The BMH 71-18 *mut*S strain is suitable for use in in vitro mutagenesis procedures where a repair (-) strain is required. BL21(DE3)pLysS cells[a] can be used with protein expression vectors that are under the control of the T7 promoter, such as pET vectors. This strain is lysogenic for lambda-DE3 (4), which contains the T7 bacteriophage gene 1, encoding T7 RNA polymerase (5) under the control of the *lac*UV5 promoter. BL21(DE3)pLysS also contains the pLysS plasmid, which carries the gene encoding T7 lysozyme. T7 lysozyme lowers the background expression level of target genes under the control of the T7 promoter but does not interfere with the level of expression achieved following induction with IPTG. For genotypic information on the *E. coli* Competent Cells, see Table 1.

Table 1. Genotypes of *E. coli* Competent Cells Offered by Promega.

Strain	Genotype
BL21(DE3)pLysS	F-, *omp*T, *hsd*S_B (r_B-, m_B-), *dcm*, *gal*, λ(DE3), pLysS, Cm^r
BMH 71-18 *mut*S	*thi*, *sup*E, Δ(*lac-pro*AB), [*mut*S::Tn10], [F', *pro*AB, *laq*I^qZΔM15]
HB101	F-, *thi*-1, *hsd*S20 (r_B-, m_B-), *sup*E44, *rec*A13, *ara*-14, *leu*B6, *pro*A2, *lac*Y1, *gal*K2, *rps*L20 (str^r), *xyl*-5, *mtl*-1
JM109	*end*A1, *rec*A1, *gyr*A96, *thi*, *hsd*R17 (r_k-, m_k+), *rel*A1, *sup*E44, Δ(*lac-pro*AB), [F', *tra*D36, *pro*AB, *laq*I^qZΔM15]

II. Product Components and Storage Conditions

Product	Size	Cat.#
JM109 Competent Cells, >10^8cfu/µg*	1ml (5 × 200µl)	L2001
JM109 Competent Cells, >10^7cfu/µg	1ml (5 × 200µl)	L1001
HB101 Competent Cells, >10^8cfu/µg	1ml (5 × 200µl)	L2011
HB101 Competent Cells, >10^7cfu/µg	1ml (5 × 200µl)	L1011
BL21(DE3)pLysS Competent Cells, >10^6cfu/µg	1ml (5 × 200µl)	L1191
BMH 71-18 *mut*S Competent Cells, >10^7cfu/µg	1ml (5 × 200µl)	L1201

* For Laboratory Use

Storage Conditions: Always store Competent Cells at -70°C. Thaw on ice when ready for use. **Do not refreeze thawed, unused aliquots.**

All cells are supplied in 200µl aliquots and are provided with 3ng of Competent Cells Control DNA for use as a positive control. Typically, 100µl of Competent Cells are required for standard transformations.

invitrogen™

Material Safety Data Sheet

Revision Date: 12-Feb-2006

1. IDENTIFICATION OF THE SUBSTANCE/PREPARATION AND THE COMPANY/UNDERTAKING

Product code 350857
Product name pLenti6/Ubc/V5-LacZ Vector, 10ug Lyophilized

Contact manufacturer
INVITROGEN CORPORATON
1600 FARADAY AVENUE
PO BOX 6482
CARLSBAD, CA 92008
760-603-7200

INVITROGEN CORPORATION
2270 INDUSTRIAL STREET
BURLINGTON, ONT
CANADA L7P 1A1
800-263-6236

GIBCO PRODUCTS
INVITROGEN CORPORATION
3175 STALEY ROAD P.O. BOX 68
GRAND ISLAND, NY 14072
716-774-6700

2. COMPOSITION/INFORMATION ON INGREDIENTS

Hazardous/Non-hazardous Components
The product contains no substances which at their given concentration, are considered to be hazardous to health

3. HAZARDS IDENTIFICATION

Emergency Overview
The product contains no substances which at their given concentration, are considered to be hazardous to health.

Form
suspension

Principle Routes of Exposure/ Potential Health effects

Eyes	No information available
Skin	No information available
Inhalation	No information available
Ingestion	No information available

Specific effects

Carcinogenic effects	No information available
Mutagenic effects	No information available
Reproductive toxicity	No information available

Sensitization	No information available
Target Organ Effects	No information available

4. FIRST AID MEASURES

Skin contact	Wash off immediately with plenty of water
Eye contact	Rinse thoroughly with plenty of water, also under the eyelids.
Ingestion	Never give anything by mouth to an unconscious person
Inhalation	Move to fresh air
Notes to physician	Treat symptomatically

5. FIRE-FIGHTING MEASURES

Suitable extinguishing media	Dry chemical
Special protective equipment for firefighters	Wear self-contained breathing apparatus and protective suit

6. ACCIDENTAL RELEASE MEASURES

Personal precautions	Use personal protective equipment
Methods for cleaning up	Soak up with inert absorbent material

7. HANDLING AND STORAGE

Handling	No special handling advice required
Storage	Keep in properly labelled containers

8. EXPOSURE CONTROLS / PERSONAL PROTECTION

Occupational exposure controls

Exposure limits

Engineering measures	Ensure adequate ventilation, especially in confined areas

Personal protective equipment

Respiratory protection	In case of insufficient ventilation wear suitable respiratory equipment
Hand protection	Protective gloves
Eye protection	Safety glasses with side-shields
Skin and body protection	Lightweight protective clothing
Hygiene measures	Handle in accordance with good industrial hygiene and safety practice
Environmental exposure controls	Prevent product from entering drains

9. PHYSICAL AND CHEMICAL PROPERTIES

General Information

Form	suspension

Important Health Safety and Environmental Information

Boiling point/range	**°C** No data available	**°F** No data available
Melting point/range	**°C** No data available	**°F** No data available
Flash point	**°C** No data available	**°F** No data available
Autoignition temperature	**°C** No data available	**°F** No data available
Oxidizing properties	No information available	

Water solubility	No data available

10. STABILITY AND REACTIVITY

Stability	Stable.
Materials to avoid	No information available
Hazardous decomposition products	No information available
Polymerization	Hazardous polymerisation does not occur

11. TOXICOLOGICAL INFORMATION

Acute toxicity

Principle Routes of Exposure/ Potential Health effects

Eyes	No information available
Skin	No information available
Inhalation	No information available
Ingestion	No information available

Specific effects

Carcinogenic effects	No information available
Mutagenic effects	No information available
Reproductive toxicity	No information available
Sensitization	No information available

Target Organ Effects — No information available

12. ECOLOGICAL INFORMATION

Ecotoxicity effects	No information available.
Mobility	No information available.
Biodegradation	Inherently biodegradable.
Bioaccumulation	Does not bioaccumulate.

13. DISPOSAL CONSIDERATIONS

Dispose of in accordance with local regulations

14. TRANSPORT INFORMATION

IATA

Proper shipping name	Not classified as dangerous in the meaning of transport regulations
Hazard Class	No information available
Subsidiary Class	No information available
Packing group	No information available
UN-No	No information available

15. REGULATORY INFORMATION

International Inventories

U.S. Federal Regulations

SARA 313
Not regulated

Clean Air Act, Section 112 Hazardous Air Pollutants (HAPs) (see 40 CFR 61)
This product contains the following HAPs:

U.S. State Regulations

California Proposition 65
This product contains the following Proposition 65 chemicals:

WHMIS hazard class:
Non-controlled

This product has been classified according to the hazard criteria of the CPR and the MSDS contains all of the information required by the CPR

16. OTHER INFORMATION

This material is sold for research and development purposes only. It is not for any human or animal therapeutic or clinical diagnostic use. It is not intended for food, drug, household, agricultural, or cosmetic use. An individual technically qualified to handle potentially hazardous chemicals must supervise the use of this material.

The above information was acquired by diligent search and/or investigation and the recommendations are based on prudent application of professional judgment. The information shall not be taken as being all inclusive and is to be used only as a guide. All materials and mixtures may be present unknown hazards and should be used with caution. Since Invitrogen Corporation cannot control the actual methods, volumes, or conditions of use, the Company shall not be held liable for any damages or losses resulting from the handling or from contact with the product as described herein. THE INFORMATION IN THIS MSDS DOES NOT CONSTITUTE A WARRANTY, EXPRESS OR IMPLIED, INCLUDING ANY IMPLIED WARRANTY OF MERCHANTABILITY OR FITNESS FOR ANY PARTCULAR PURPOSE.

End of Safety Data Sheet

SIGMA-ALDRICH

MATERIAL SAFETY DATA SHEET

Date Printed: 12/11/2003
Date Updated: 09/02/2002
Version 1.2

Section 1 - Product and Company Information

Product Name	STREPTOZOTOCIN MIXED ANOMERS
Product Number	S0130
Brand	SIGMA
Company	Sigma-Aldrich Canada, Ltd
Street Address	2149 Winston Park Drive
City, State, Zip, Country	Oakville ON L6H 6J8 CA
Technical Phone:	9058299500
Emergency Phone:	800-424-9300
Fax:	9058299292

Section 2 - Composition/Information on Ingredient

Substance Name	CAS #	SARA 313
STREPTOZOCIN	18883-66-4	No

Formula C8H15N3O7

Synonyms 2-Deoxy-2-(((methylnitrosoamino)carbonyl)amino)-D-glucopyranose * 2-Deoxy-2-(3-methyl-3-nitrosoureido)-D-glucopyranose * 2-Deoxy-2-(3-methyl-3-nitrosoureido)-alpha(and beta)-D-glucopyranose * D-Glucopyranose, 2-deoxy-2-(((methylnitrosoamino)carbonyl)amino)- * D-Glucose, 2-deoxy-2-(((methylnitrosoamino)carbonyl)amino)-(9CI) * D-Glucose, 2-deoxy-2-(3-methyl-3-nitrosoureido)- * N-d-Glucosyl(2)-N'-nitrosomethylharnstoff (German * N-D-Glucosyl-(2)-N'-nitrosomethylurea * NCI-C03167 * NSC-85598 * NSC-85998 * RCRA waste number U206 * Streptozocin * Streptozotocin * STRZ * U-9889 * Zanosar

RTECS Number: LZ5775000

Section 3 - Hazards Identification

EMERGENCY OVERVIEW

Harmful.
Limited evidence of a carcinogenic effect.
Probable Carcinogen (US). Target organ(s): Pancreas. Blood. Calif. Prop. 65 carcinogen & reproductive hazard.

HMIS RATING

HEALTH: 0*
FLAMMABILITY: 0
REACTIVITY: 0

NFPA RATING

HEALTH: 0
FLAMMABILITY: 0

REACTIVITY: 0

*additional chronic hazards present.

For additional information on toxicity, please refer to Section 11.

Section 4 - First Aid Measures

ORAL EXPOSURE
If swallowed, wash out mouth with water provided person is conscious. Call a physician.

INHALATION EXPOSURE
If inhaled, remove to fresh air. If breathing becomes difficult, call a physician.

DERMAL EXPOSURE
In case of contact, immediately wash skin with soap and copious amounts of water.

EYE EXPOSURE
In case of contact with eyes, flush with copious amounts of water for at least 15 minutes. Assure adequate flushing by separating the eyelids with fingers. Call a physician.

Section 5 - Fire Fighting Measures

FLASH POINT
N/A

AUTOIGNITION TEMP
N/A

FLAMMABILITY
N/A

EXTINGUISHING MEDIA
Suitable: Water spray. Carbon dioxide, dry chemical powder, or appropriate foam.

FIREFIGHTING
Protective Equipment: Wear self-contained breathing apparatus and protective clothing to prevent contact with skin and eyes.
Specific Hazard(s): Emits toxic fumes under fire conditions.

Section 6 - Accidental Release Measures

PROCEDURE TO BE FOLLOWED IN CASE OF LEAK OR SPILL
Evacuate area.

PROCEDURE(S) OF PERSONAL PRECAUTION(S)
Wear self-contained breathing apparatus, rubber boots, and heavy rubber gloves. Wear disposable coveralls and discard them after use.

METHODS FOR CLEANING UP
Sweep up, place in a bag and hold for waste disposal. Avoid raising dust. Ventilate area and wash spill site after material pickup is complete.

Section 7 - Handling and Storage

HANDLING
User Exposure: Do not breathe dust. Do not get in eyes, on skin, on clothing. Avoid prolonged or repeated exposure.

STORAGE
Suitable: Keep tightly closed.
Store at -20°C

Section 8 - Exposure Controls / PPE

ENGINEERING CONTROLS
Use only in a chemical fume hood. Safety shower and eye bath.

PERSONAL PROTECTIVE EQUIPMENT
Respiratory: Government approved respirator in nonventilated areas and/or for exposure above the TLV or PEL.
Hand: Compatible chemical-resistant gloves.
Eye: Chemical safety goggles.

GENERAL HYGIENE MEASURES
Wash contaminated clothing before reuse. Wash thoroughly after handling.

Section 9 - Physical/Chemical Properties

Appearance	Physical State: Solid. Color: Slightly yellow Form: Fine crystals

Property	Value	At Temperature or Pressure
Molecular Weight	265.23 AMU	
pH	N/A	
BP/BP Range	N/A	
MP/MP Range	114 °C	
Freezing Point	N/A	
Vapor Pressure	N/A	
Vapor Density	N/A	
Saturated Vapor Conc.	N/A	
Bulk Density	N/A	
Odor Threshold	N/A	
Volatile%	N/A	
VOC Content	N/A	
Water Content	N/A	
Solvent Content	N/A	
Evaporation Rate	N/A	
Viscosity	N/A	
Surface Tension	N/A	
Partition Coefficient	N/A	
Decomposition Temp.	N/A	
Flash Point	N/A	
Explosion Limits	N/A	
Flammability	N/A	
Autoignition Temp	N/A	
Refractive Index	N/A	
Optical Rotation	N/A	
Miscellaneous Data	N/A	
Solubility	Solubility in Water:Soluble.	

N/A = not available

Section 10 - Stability and Reactivity

STABILITY
Stable: Stable.
Materials to Avoid: Strong oxidizing agents, Strong acids, Strong bases.

HAZARDOUS DECOMPOSITION PRODUCTS
Hazardous Decomposition Products: Carbon monoxide, Carbon dioxide, Nitrogen oxides.

HAZARDOUS POLYMERIZATION
Hazardous Polymerization: Will not occur

Section 11 - Toxicological Information

ROUTE OF EXPOSURE
Skin Contact: May cause skin irritation.
Skin Absorption: May be harmful if absorbed through the skin.
Eye Contact: May cause eye irritation.
Inhalation: Material may be irritating to mucous membranes and upper respiratory tract. May be harmful if inhaled.
Ingestion: May be harmful if swallowed.

TARGET ORGAN(S) OR SYSTEM(S)
Pancreas. Liver. Kidneys. Blood. Reproductive system.

SIGNS AND SYMPTOMS OF EXPOSURE
Anemia. To the best of our knowledge, the chemical, physical, and toxicological properties have not been thoroughly investigated. Nausea. Vomiting.

TOXICITY DATA
Oral
Rat
5,150 mg/kg
LD50
Oral
Mouse
3,000 mg/kg
LD50
Intravenous
Dog
25 - 50 mg/kg
LD50
Intravenous
Mouse
275 mg/kg
LD50
Intraperitoneal
Mouse
219 mg/kg
LD50
Subcutaneous
Mouse
335 mg/kg
LD50

Intravenous
Woman
440 MG/KG
LDLO
Remarks: Gastrointestinal:Ulceration or bleeding from duodenum.

Kidney, Ureter, Bladder:Urine volume increased. Nutritional and Gross Metabolic:Changes in:K

Intravenous
Rat
138 MG/KG
LD50

Intraperitoneal
Mouse
360 MG/KG
LD50
Remarks: Tumorigenic:Active as anti-cancer agent.

Subcutaneous
Mouse
335 MG/KG
LD50

Intravenous
Mouse
275 MG/KG
LD50
Remarks: Tumorigenic:Active as anti-cancer agent.

Parenteral
Mouse
264 MG/KG
LD50
Remarks: Tumorigenic:Active as anti-cancer agent.

Intravenous
Dog
50 MG/KG
LD50
Remarks: Tumorigenic:Active as anti-cancer agent.

CHRONIC EXPOSURE - CARCINOGEN
Result: This product is or contains a component that has been reported to be probably carcinogenic based on its IARC, OSHA, ACGIH, NTP, or EPA classification.

Species: Rat
Route of Application: Intraperitoneal
Dose: 470 MG/KG
Exposure Time: 26W
Frequency: I
Result: Tumorigenic:Neoplastic by RTECS criteria. Liver:Tumors. Kidney, Ureter, Bladder:Kidney tumors.

Species: Rat
Route of Application: Intravenous
Dose: 25 MG/KG
Result: Tumorigenic:Neoplastic by RTECS criteria. Kidney, Ureter, Bladder:Kidney tumors.

Species: Mouse
Route of Application: Intraperitoneal
Dose: 470 MG/KG
Exposure Time: 26W
Frequency: I
Result: Tumorigenic:Neoplastic by RTECS criteria. Lungs, Thorax,

or Respiration:Tumors. Kidney, Ureter, Bladder:Kidney tumors.

Species: Mouse
Route of Application: Intravenous
Dose: 250 MG/KG
Result: Tumorigenic:Carcinogenic by RTECS criteria. Kidney, Ureter, Bladder:Kidney tumors.

Species: Hamster
Route of Application: Intraperitoneal
Dose: 100 MG/KG
Exposure Time: I
Result: Tumorigenic:Equivocal tumorigenic agent by RTECS criteria. Liver:Tumors. Kidney, Ureter, Bladder:Kidney tumors.

Species: Rat
Route of Application: Intravenous
Dose: 65 MG/KG
Result: Tumorigenic:Equivocal tumorigenic agent by RTECS criteria. Gastrointestinal:Tumors. Kidney, Ureter, Bladder:Kidney tumors.

Species: Rat
Route of Application: Intravenous
Dose: 50 MG/KG
Result: Tumorigenic:Neoplastic by RTECS criteria. Kidney, Ureter, Bladder:Kidney tumors.

Species: Rat
Route of Application: Intravenous
Dose: 30 MG/KG
Result: Tumorigenic:Neoplastic by RTECS criteria. Gastrointestinal:Tumors. Kidney, Ureter, Bladder:Kidney tumors.

Species: Rat
Route of Application: Intravenous
Dose: 65 MG/KG
Result: Tumorigenic:Equivocal tumorigenic agent by RTECS criteria. Liver:Tumors.

Species: Rat
Route of Application: Intravenous
Dose: 50 MG/KG
Result: Tumorigenic:Equivocal tumorigenic agent by RTECS criteria. Liver:Tumors. Kidney, Ureter, Bladder:Kidney tumors.

Species: Rat
Route of Application: Intravenous
Dose: 50 MG/KG
Result: Tumorigenic:Neoplastic by RTECS criteria. Kidney, Ureter, Bladder:Kidney tumors.

Species: Rat
Route of Application: Intravenous
Dose: 65 MG/KG
Result: Tumorigenic:Equivocal tumorigenic agent by RTECS criteria. Kidney, Ureter, Bladder:Kidney tumors.

Species: Rat
Route of Application: Intravenous
Dose: 30 MG/KG
Result: Tumorigenic:Neoplastic by RTECS criteria.

Gastrointestinal:Tumors.

Species: Rat
Route of Application: Intravenous
Dose: 65 MG/KG
Result: Tumorigenic:Equivocal tumorigenic agent by RTECS criteria. Gastrointestinal:Tumors.

IARC CARCINOGEN LIST

Rating: Group 2B

NTP CARCINOGEN LIST

Rating: Clear evidence.
Species: Mouse/rat
Route: Intraperitoneal

IRIS/EPA CARCINOGEN LIST
Rating: Group B2

CHRONIC EXPOSURE - TERATOGEN
Result: Laboratory experiments have shown teratogenic effects.

Species: Rat
Dose: 50 MG/KG
Route of Application: Intraperitoneal
Exposure Time: (1D PRE)
Result: Effects on Embryo or Fetus: Extra embryonic structures (e.g., placenta, umbilical cord).

Species: Rat
Dose: 50 MG/KG
Route of Application: Intraperitoneal
Exposure Time: (8D PREG)
Result: Effects on Embryo or Fetus: Extra embryonic structures (e.g., placenta, umbilical cord). Effects on Embryo or Fetus: Fetotoxicity (except death, e.g., stunted fetus). Specific Developmental Abnormalities: Cardiovascular (circulatory) system.

Species: Rat
Dose: 50 MG/KG
Route of Application: Intraperitoneal
Exposure Time: (7D PREG)
Result: Effects on Embryo or Fetus: Fetotoxicity (except death, e.g., stunted fetus). Specific Developmental Abnormalities: Craniofacial (including nose and tongue).

Species: Rat
Dose: 50 MG/KG
Route of Application: Intraperitoneal
Exposure Time: (12D PREG)
Result: Specific Developmental Abnormalities: Cardiovascular (circulatory) system.

Species: Rat
Dose: 35 MG/KG
Route of Application: Intravenous
Exposure Time: (8D PREG)
Result: Effects on Embryo or Fetus: Extra embryonic structures (e.g., placenta, umbilical cord). Effects on Embryo or Fetus: Fetotoxicity (except death, e.g., stunted fetus).

Species: Rat
Dose: 30 MG/KG
Route of Application: Intravenous
Exposure Time: (5D PREG)
Result: Effects on Embryo or Fetus: Fetotoxicity (except death, e.g., stunted fetus). Effects on Newborn: Growth statistics (e.g., reduced weight gain).

Species: Rat
Dose: 40 MG/KG
Route of Application: Intravenous
Exposure Time: (5D PREG)
Result: Specific Developmental Abnormalities: Endocrine system. Effects on Newborn: Biochemical and metabolic.

Species: Rat
Dose: 65 MG/KG
Route of Application: Unreported
Exposure Time: (5D PREG)
Result: Specific Developmental Abnormalities: Musculoskeletal system.

CHRONIC EXPOSURE - MUTAGEN
Result: Laboratory experiments have shown mutagenic effects.

Species: Human
Dose: 50 MG/L
Cell Type: Other cell types
Mutation test: DNA damage

Species: Human
Dose: 1 MMOL/L
Cell Type: kidney
Mutation test: DNA damage

Species: Human
Dose: 100 UMOL/L
Cell Type: HeLa cell
Mutation test: DNA inhibition

Species: Human
Dose: 1 GM/L
Exposure Time: 3H
Cell Type: lung
Mutation test: DNA inhibition

Species: Human
Dose: 500 MG/L
Cell Type: lung
Mutation test: Cytogenetic analysis

Species: Human
Dose: 100 UMOL/L
Cell Type: Other cell types
Mutation test: Sister chromatid exchange

Species: Rat
Dose: 2 MMOL/L
Cell Type: Other cell types
Mutation test: Morphological transformation.

Species: Rat
Dose: 1 MMOL/L
Cell Type: kidney
Mutation test: DNA damage

Species: Rat
Dose: 1 MMOL/L
Cell Type: Other cell types
Mutation test: DNA damage

Species: Rat
Route: Oral
Dose: 20 MG/KG
Mutation test: DNA damage

Species: Rat
Dose: 300 UMOL/L
Cell Type: liver
Mutation test: DNA damage

Species: Rat
Route: Intraperitoneal
Dose: 250 MG/KG
Mutation test: Unscheduled DNA synthesis

Species: Rat
Dose: 50 UMOL/L
Cell Type: liver
Mutation test: Unscheduled DNA synthesis

Species: Rat
Dose: 2 MMOL/L
Cell Type: Other cell types
Mutation test: Unscheduled DNA synthesis

Species: Rat
Dose: 200 UMOL/L
Cell Type: Other cell types
Mutation test: Sister chromatid exchange

Species: Rat
Dose: 111 MG/KG
Cell Type: S. typhimurium
Mutation test: Host-mediated assay

Species: Mouse
Route: Intraperitoneal
Dose: 150 MG/KG
Mutation test: DNA

Species: Mouse
Dose: 500 UMOL/L
Cell Type: leukocyte
Mutation test: DNA damage

Species: Mouse
Dose: 200 MG/KG
Cell Type: leukocyte
Mutation test: Other mutation test systems

Species: Mouse
Dose: 200 MG/KG

Cell Type: Other cell types
Mutation test: DNA inhibition

Species: Mouse
Dose: 12 MG/KG
Cell Type: S. typhimurium
Mutation test: Body fluid assay

Species: Mouse
Dose: 10 MG/L
Cell Type: lymphocyte
Mutation test: Mutation in mammalian somatic cells.

Species: Mouse
Dose: 500 UG/KG
Cell Type: S. typhimurium
Mutation test: Host-mediated assay

Species: Mouse
Dose: 1 MG/KG
Cell Type: E. coli
Mutation test: Host-mediated assay

Species: Hamster
Dose: 250 MG/L
Cell Type: lung
Mutation test: DNA damage

Species: Hamster
Dose: 38 UMOL/L
Cell Type: ovary
Mutation test: Cytogenetic analysis

Species: Hamster
Dose: 189 UMOL/L
Cell Type: ovary
Mutation test: Sister chromatid exchange

Species: Hamster
Dose: 189 UMOL/L
Cell Type: lung
Mutation test: Sister chromatid exchange

Species: Hamster
Dose: 100 UMOL/L
Cell Type: ovary
Mutation test: Mutation in mammalian somatic cells.

Species: Hamster
Dose: 500 UMOL/L
Cell Type: lung
Mutation test: Mutation in mammalian somatic cells.

CHRONIC EXPOSURE - REPRODUCTIVE HAZARD

Species: Rat
Dose: 70 MG/KG
Route of Application: Intravenous
Exposure Time: (1D PRE)
Result: Maternal Effects: Ovaries, fallopian tubes.

Species: Rat

Dose: 40 MG/KG
Route of Application: Intravenous
Exposure Time: (1D PREG)
Result: Effects on Fertility: Post-implantation mortality (e.g., dead and/or resorbed implants per total number of implants). Specific Developmental Abnormalities: Central nervous system. Specific Developmental Abnormalities: Cardiovascular (circulatory) system.

Species: Rat
Dose: 60 MG/KG
Route of Application: Intravenous
Exposure Time: (1D PRE)
Result: Effects on Fertility: Other measures of fertility

Species: Rat
Dose: 40 MG/KG
Route of Application: Parenteral
Exposure Time: (1D PRE)
Result: Effects on Fertility: Post-implantation mortality (e.g., dead and/or resorbed implants per total number of implants). Effects on Embryo or Fetus: Fetotoxicity (except death, e.g., stunted fetus). Specific Developmental Abnormalities: Other developmental abnormalities.

Species: Hamster
Dose: 150 MG/KG
Route of Application: Intraperitoneal
Exposure Time: (3D PRE)
Result: Maternal Effects: Menstrual cycle changes or disorders. Effects on Fertility: Pre-implantation mortality (e.g., reduction in number of implants per female; total number of implants per corpora lutea). Effects on Fertility: Post-implantation mortality (e.g., dead and/or resorbed implants per total number of implants).

Species: Domestic Animals
Dose: 100 MG/KG
Route of Application: Intravenous
Exposure Time: (85-95D PREG)
Result: Maternal Effects: Other effects. Endocrine:Diabetes mellitus.

Section 12 - Ecological Information

No data available.

Section 13 - Disposal Considerations

APPROPRIATE METHOD OF DISPOSAL OF SUBSTANCE OR PREPARATION
Contact a licensed professional waste disposal service to dispose of this material. Observe all federal, state, and local environmental regulations.

Section 14 - Transport Information

DOT
Proper Shipping Name: Environmentally hazardous substances, solid, n.o.s.
UN#: 3077
Class: 9
Packing Group: Packing Group III

Hazard Label: Class 9
PIH: Not PIH

IATA
Proper Shipping Name: Environmentally hazardous substance, solid, n.o.s
IATA UN Number: 3077
Hazard Class: 9
Packing Group: III

Section 15 - Regulatory Information

EU ADDITIONAL CLASSIFICATION
Symbol of Danger: Xn
Indication of Danger: Harmful.
R: 40
Risk Statements: Limited evidence of a carcinogenic effect.
S: 36
Safety Statements: Wear suitable protective clothing.

US CLASSIFICATION AND LABEL TEXT
Indication of Danger: Harmful.
Risk Statements: Limited evidence of a carcinogenic effect.
Safety Statements: Wear suitable protective clothing.
US Statements: Probable Carcinogen (US). Target organ(s): Pancreas. Blood. Calif. Prop. 65 carcinogen & reproductive hazard.

UNITED STATES REGULATORY INFORMATION
SARA LISTED: No

UNITED STATES - STATE REGULATORY INFORMATION

CALIFORNIA PROP - 65
California Prop - 65: This product is or contains chemical(s) known to the state of California to cause cancer. This product is or contains chemical(s) known to the state of California to cause male reproductive toxicity. This product is or contains chemical(s) known to the state of California to cause female reproductive toxicity.

CANADA REGULATORY INFORMATION
WHMIS Classification: This product has been classified in accordance with the hazard criteria of the CPR, and the MSDS contains all the information required by the CPR.
DSL: No
NDSL: No

Section 16 - Other Information

DISCLAIMER
For R&D use only. Not for drug, household or other uses.

WARRANTY
The above information is believed to be correct but does not purport to be all inclusive and shall be used only as a guide. Sigma-Aldrich Inc., shall not be held liable for any damage resulting from handling or from contact with the above product. See reverse side of invoice or packing slip for additional terms and conditions of sale.

SIGMA-ALDRICH

Material Safety Data Sheet

Version 3.0
Revision Date 01/02/2009
Print Date 02/03/2009

1. PRODUCT AND COMPANY IDENTIFICATION

Product name : Ganciclovir

Product Number : G2536
Brand : Sigma

Company : Sigma-Aldrich Canada, Ltd
2149 Winston Park Drive
OAKVILLE ON L6H 6J8
CANADA
Telephone : +1 9058299500
Fax : +1 9058299292
Emergency Phone # : 800-424-9300

2. COMPOSITION/INFORMATION ON INGREDIENTS

Formula : $C_9H_{13}N_5O_4$
Molecular Weight : 255.23 g/mol

CAS-No.	EC-No.	Index-No.	Concentration
Ganciclovir			
82410-32-0	-	-	-

3. HAZARDS IDENTIFICATION

Emergency Overview

Target Organs

Bone marrow, Central nervous system, Blood, Eyes, Liver, Kidney, Reproductive system.

WHMIS Classification

Not WHMIS controlled. Not WHMIS controlled.

HMIS Classification

Health Hazard: 1
Chronic Health Hazard: *
Flammability: 0
Physical hazards: 0

Potential Health Effects

Inhalation May be harmful if inhaled. May cause respiratory tract irritation.
Skin May be harmful if absorbed through skin. May cause skin irritation.
Eyes May cause eye irritation.
Ingestion May be harmful if swallowed.

4. FIRST AID MEASURES

General advice
Consult a physician. Show this safety data sheet to the doctor in attendance.

If inhaled
If breathed in, move person into fresh air. If not breathing give artificial respiration Consult a physician.

In case of skin contact
Wash off with soap and plenty of water. Consult a physician.

In case of eye contact
Rinse thoroughly with plenty of water for at least 15 minutes and consult a physician.

If swallowed
Never give anything by mouth to an unconscious person. Rinse mouth with water. Consult a physician.

5. FIRE-FIGHTING MEASURES

Flammable properties

Flash point	no data available
Ignition temperature	no data available

Suitable extinguishing media
Use water spray, alcohol-resistant foam, dry chemical or carbon dioxide.

Special protective equipment for fire-fighters
Wear self contained breathing apparatus for fire fighting if necessary.

6. ACCIDENTAL RELEASE MEASURES

Personal precautions
Use personal protective equipment. Avoid dust formation. Avoid breathing dust. Ensure adequate ventilation. Evacuate personnel to safe areas.

Environmental precautions
Prevent further leakage or spillage if safe to do so. Do not let product enter drains.

Methods for cleaning up
Pick up and arrange disposal without creating dust. Keep in suitable, closed containers for disposal.

7. HANDLING AND STORAGE

Handling
Avoid exposure - obtain special instructions before use. Avoid formation of dust and aerosols.
Provide appropriate exhaust ventilation at places where dust is formed. Normal measures for preventive fire protection.

Storage
Keep container tightly closed in a dry and well-ventilated place.

Keep in a dry place.

8. EXPOSURE CONTROLS/PERSONAL PROTECTION

Contains no substances with occupational exposure limit values.

Personal protective equipment

Respiratory protection
Where risk assessment shows air-purifying respirators are appropriate use a full-face particle respirator type N99 (US) or type P2 (EN 143) respirator cartridges as a backup to engineering controls. If the respirator is the sole means of protection, use a full-face supplied air respirator. Use respirators and components tested and approved under appropriate government standards such as NIOSH (US) or CEN (EU).

Hand protection
For prolonged or repeated contact use protective gloves.

Eye protection
Safety glasses

Skin and body protection
Choose body protection according to the amount and concentration of the dangerous substance at the work place.

Hygiene measures
Avoid contact with skin, eyes and clothing. Wash hands before breaks and immediately after handling the product.

9. PHYSICAL AND CHEMICAL PROPERTIES

Appearance

Form	powder

Safety data

pH	no data available
Melting point	no data available
Boiling point	no data available
Flash point	no data available
Ignition temperature	no data available
Lower explosion limit	no data available
Upper explosion limit	no data available
Water solubility	no data available

10. STABILITY AND REACTIVITY

Storage stability
Stable under recommended storage conditions.

Materials to avoid
Strong oxidizing agents

Hazardous decomposition products
Hazardous decomposition products formed under fire conditions. - Carbon oxides, nitrogen oxides (NOx)

11. TOXICOLOGICAL INFORMATION

Acute toxicity

LD50 Oral - mouse - > 2,000 mg/kg

Irritation and corrosion

no data available

Sensitisation

no data available

Chronic exposure

IARC: No component of this product present at levels greater than or equal to 0.1% is identified as

probable, possible or confirmed human carcinogen by IARC.

Genotoxicity in vitro - Human - lymphocyte
Sister chromatid exchange

Signs and Symptoms of Exposure

Nausea, Vomiting, Weakness, Dizziness, Headache, Sweating

Potential Health Effects

Inhalation	May be harmful if inhaled. May cause respiratory tract irritation.
Skin	May be harmful if absorbed through skin. May cause skin irritation.
Eyes	May cause eye irritation.
Ingestion	May be harmful if swallowed.
Target Organs	Bone marrow, Central nervous system, Blood, Eyes, Liver, Kidney, Reproductive system.,

Additional Information
RTECS: MF8407000

12. ECOLOGICAL INFORMATION

Elimination information (persistence and degradability)

no data available

Ecotoxicity effects

no data available

Further information on ecology

no data available

13. DISPOSAL CONSIDERATIONS

Product
Observe all federal, state, and local environmental regulations.

Contaminated packaging
Dispose of as unused product.

14. TRANSPORT INFORMATION

DOT (US)
Not dangerous goods

IMDG
Not dangerous goods

IATA
Not dangerous goods

15. REGULATORY INFORMATION

DSL Status
This product contains the following components that are not on the Canadian DSL nor NDSL lists.

	CAS-No.
Ganciclovir	82410-32-0

WHMIS Classification

Not WHMIS controlled.	Not WHMIS controlled.

16. OTHER INFORMATION

Further information

The above information is believed to be correct but does not purport to be all inclusive and shall be used only as a guide. The information in this document is based on the present state of our knowledge and is applicable to the product with regard to appropriate safety precautions. It does not represent any guarantee of the properties of the product. Sigma-Aldrich Co., shall not be held liable for any damage resulting from handling or from contact with the above product. See reverse side of invoice or packing slip for additional terms and conditions of sale.