THE UNIVERSITY OF WESTERN ONTARIO
BIOLOGICAL AGENTS REGISTRY FORM
Approved Biohazards Subcommittee: October 14, 2010
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 (UWO) or in charge of a laboratory/facility where the use of Level 1, 2 or 3 biological agents is 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 Public Health Agency of Canada (PHAC) or Canadian Food Inspection Agency (CFIA) permits.

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

Containment Levels will be established in accordance with Laboratory Biosafety Guidelines, 3rd edition, 2004, Public Health Agency of Canada (PHAC) 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), (Support Services Building, Room 4190) for distribution to the Biohazards Subcommittee. For questions regarding this form, please contact the Biosafety Officer at extension 81135 or biosafety@uwo.ca. If there are changes to the information on this form (excluding grant title and funding agencies), contact Occupational Health and Safety for a modification form. See website: www.uwo.ca/humanresources/biosafety

PRINCIPAL INVESTIGATOR	Fred Possmayer
DEPARTMENT	OBS/GYN and BIOCHEM
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Location of experimental work to be carried out: Building(s) Dental Sciences Building
Room(s) DSB2019A, DSB5004,DSB5006

*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 the University of Western Ontario Biosafety Officer (See Section 15.0, Approvals).

FUNDING AGENCY/AGENCIES: CIHR
GRANT TITLE(S): Lipid-lipid and lipid-protein interactions in pulmonary surfactant

List all personnel working under Principal Investigators supervision in this location:

Name	UWO E-mail Address	Date of Biosafety Training
Lin Zhao	Lzhoa3@uwo.ca	training? Sept 2002

Please explain the biological agents and/or biohazardous substances used and how they will be stored, used and disposed of. Projects without this description will not be reviewed.

Culture experiments will be conducted in Laboratory DSB2019 which is shared with Drs, Dean Betts, Tim Regnault and Dan Hardy. Experiments are conducted in accordance with UWO's Laboratory Health and Safety Manual. In general, experiments are conducted by my research associate, Dr. Lin Zhao, whom I share with Drs. T. Regnault and D. Hardy.

Our cell lines are stored in a liquid nitrogen depository in the laboratory of Dr. Moshmi Bhattacharya. Culture manipulations are conducted, where possible, in an approved biosafety cabinet in laboratory DSB 5019. This facility is approved and has been inspected recently.

After studies, the cultures, samples and any other potentially contaminated materials are inactivated with 70% isopropanol or 70% ethanol , where appropriate. These reagents are kept in 250 or 500 ml platic squeeze bottle and a few ml are forced into the culture dishes or whatever. The bottles are labeled inflammable``. The dishes are flasks bagged in Biohazard disposable containers and autoclaved prior to disposal.

In general, standard commercial vectors and reagents are employed for genetic manipulation of RNA and DNA. One change over our previous protocols is that we propose to introduce the use of shRNAi to downregulate expression of the three known Lipid Phosphate Phosphohydrolase isoforms (Nanjundan M and Possmayer F., 2003, Pulmonary phosphatidic acid phosphatase and lipid phosphate phosphohydrolase. *Am J Physiol Lung Cell Mol Physiol* 284: L1-23). These are available commercially as lentiviral vector constructs. At present there is no reason to suspect these materials will constitute a biohazard.

These shRNAi reagents will be purchased from OpenBiosystems and stored as glycerol stocks in a freezer.

Cholera toxin and pertussis toxin are stored lyophilized and small aqueous stocks are stored frozen.

Please include a one page research summary or teaching protocol.

Our laboratory's studies have been primarily related to various aspects of pulmonary surfactant, a material which is essential for normal breathing. As part of these studies, we have been studying alveolar epithelial Type II cell lines. Type II cells, which cover <5% of the alveolar surface, synthesize and secrete pulmonary surfactant. During these studies, we examined lung epithelial T_7 cells, a purported Type II cell line derived from mice bearing a temperature-sensitive (TS) Large T antigen (de Mello et al. (2000)) *In Vitro Cell. Dev. Biol.* 36: 374-382). T_7 cells grow well at 37°C but stop dividing and differentiate at 41°C. During our studies, we discovered that this T_7 line had changed its phenotype from Type II to Type I. Type I cells are extremely difficult to isolate from the lung and in fact, this has only been reported a few times and by a single group. Although the number of Type I cell and Type II cells is similar, Type I cells cover >95% of the alveolar surface. These cells function in gas exchange and transport, but their functions are not well understood.

This discovery led us to examine the circumstances involved in establishing and manipulating Type I cell characteristics. In particular, we have found that cAMP affects the levels of lipid phosphate phosphohydrolase (LPP) activity and LPP isoform expression. We are currently attempting to determine the relative contributions of these isoforms to Type I cell LPP activity and the manner in which expression of these LPP isoforms and their activity is controlled. For these studies, we propose to transient infect cells with lentiviral pGFZ and pLKO.I vectors to produce shRNAi to modify LPP isoform levels. These studies will provide information on the factors regulating the alveolar Type 1 cell phenotype and the manner in which LPP isoform expression is controlled. It will alaso establish a new and convenient platrform for studying Type 1 cell function.

1.0 Microorganisms

1.1 Does your work involve the use of biological agents? xO YES O NO
(non-pathogenic and pathogenic biological agents including but not limited to bacteria and other microorganisms, viruses, prions, parasites or pathogens of plant or animal origin)? If no, please proceed to Section 2.0

Do you use microorganisms that require a permit from the CFIA? O YES xO 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:

_____
_____
_____

Please attach the CFIA permit.
Please describe any CFIA permit conditions:

_____
_____
_____

1.2 Please complete the table below:

Name of Biological Agent(s)* (Be specific)	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	PHAC or CFIA Containment Level
E. coli (dH5alpha)	O Yes xO No	O Yes xO No	O Yes xO No	1-2 liters	Commercial or In House	xO 1 O 2 O 2+ O 3
	O Yes O No	O Yes O No	O Yes O No			O 1 O 2 O 2+ O 3
	O Yes O No	O Yes O No	O Yes O No			O 1 O 2 O 2+ O 3
	O Yes O No	O Yes O No	O Yes O No			O 1 O 2 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? xO 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:

Cell Type	Is this cell type used in your work?	Source of Primary Cell Culture Tissue	AUS Protocol Number
Human	O Yes xO No		
Rodent	O Yes xO No		

Non-human primate	O Yes xO No		
Other (specify)	O Yes xO No		

2.3 Please indicate the type of established cells that will be grown in culture in:

Cell Type	Is this cell type used in your work?	Specific cell line(s)*	Containment Level of each cell line	Supplier / Source of cell line(s)
Human	xO Yes O No	A549,H440 (lung epithelial) HEK293	1 or 2?	ATCC,In house,
Rodent	xO Yes O No	MLE-12, T_7 (mouse lung epithelial, SV40 large T antigen)	2	Jeff Whitsett who developed this cell line. It is now available from ATCC Martin Post, Paediatrics, H Sick Children
Non-human primate	O Yes xO No			
Other (specify)	O Yes xO No	HEK 293(E1A oncogene) CHO		

*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 PHAC or CFIA containment level required O 1 xO 2 O 2+ O 3

3.0 Use of Human Source Materials

3.1 Does your work involve the use of human source materials? O YES xO 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 Infected With An Infectious Agent? YES/UNKNOWN**	**Name of Infectious Agent (If applicable)**	**PHAC or CFIA Containment Level (Select one)**
Human Blood (whole) or other Body Fluid		O Yes O Unknown		O 1 O 2 O 2+ O 3
Human Blood (fraction) or other Body Fluid		O Yes O Unknown		O 1 O 2 O 2+ O 3
Human Organs or Tissues (unpreserved)		O Yes O Unknown		O 1 O 2 O 2+ O 3
Human Organs or Tissues (preserved)		Not Applicable		Not Applicable

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? xO 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 from transformation or tranfection

Ecoli (dH5 alpha)	*pCMV-SPORTS*	*Open Biosystems*	*LPP1-3*	*.no changes noted when we have done similer experiments in the past* *We have not used these particular clones yet but do not anticipate any difference*

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

See Appendix added at end for plasmid maps

4.3 Will genetic modification(s) of bacteria and/or cells involving viral vectors be made?
xO YES, complete table below O NO

Virus Used for Vector Construction	Vector(s) *	Source of Vector	Gene(s) Transduced	Describe the change that results from transduction
Not applicable,(we will not use viruses)	pGPZ pKIO.1	OpenBiosystems OpenBiosystems "	Mouse LPP1-3 shRNAi	Unknown Hope specific mRNAs for isoforms will be downregulated

* Please attach a Material Safety Data Sheet or equivalent.

4.4 Will genetic sequences from the following be involved?

- HIV O YES, please specify _____ xO NO
- HTLV 1 or 2 or genes from any Level 1 or Level 2 pathogens O YES, specify _____ xO NO
- SV 40 Large T antigen xO YES O NO

As stated above, the T7 line bears the large T antigen.

HPV ?

- E1A oncogene xO YES
  HEK-293 cells will be used to express the LPPs
- **Other** Known oncogenes xO YES, please specify __E1A in HEK 293 cells_____ xO NO
- Other human or animal pathogen and or their toxins O YES, please specify _____ x O NO

4.5 Will virus be replication defective? O YES O NO
Virus will not be used, only vector constructs as indicated above. These are replication defective.
4.6 Will virus be infectious to humans or animals? O YES O NO

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

5.0 Human Gene Therapy Trials

5.1 Will human clinical trials be conducted involving a biological agent? O YES xO NO
(including but not limited to microorganisms, viruses, prions, parasites or pathogens of plant or animal origin)
If no, please proceed to Section 6.0

5.2 If YES, please specify which biological agent will be used: _____
Please attach a full description of the biological agent.

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

5.3 How will the biological agent 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? O YES xO NO If no, please proceed to section 7.0

6.2 Name of animal species to be used_____

6.3 AUS protocol # _____

6.4 Will any of the agents listed in section 4.0 be used in live animals O YES, specify: _____ O NO

6.5 Will the agent(s) be shed by the animal: O YES O NO, please justify:

7.0 Use of Animal species with Zoonotic Hazards

7.1 Will any animals with zoonotic hazards or their organs, tissues, lavages or other body fluids including blood be used (see list below)? O YES xO No If no, please proceed to section 8.0

7.2 Will live animals be used? O YES xO No

7.3 If yes, please specify the animal(s) used:

- Pound source dogs O YES O NO
- Pound source cats O YES O NO
- Cattle, sheep or goats O YES, please specify species _____ O NO
- Non-human primates O YES, please specify species _____ O NO
- Wild caught animals xO YES, please specify species & colony # __In some cases we may examine extracts from the 13-stripped ground squirrel. These extracts are from a collaboration with Dr. Jim Staples BIOLOGY and Ruud Veldhuizen Lawson Research Institute,UWO.
- The extracts will be in chloroform:methanol and so will be sterile.
- _____ O NO
- Birds O YES, please specify species _____ O NO
- Others (wild or domestic) O YES, please specify ___We may also obtain lungs from investigators using Xenopus __laevis (e.g. Tom Drysdale, VRL, UWO) to extract surfactant_____ O NO

7.4 If no live animals are used, please specify the source of the specimens: See above_____

8.0 Biological Toxins

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

8.2 If YES, please name the toxin(s)__Cholera toxin, pertussis toxin_____
Please attach information, such as a Material Safety Data Sheet, for the toxin(s) used.
Could not find a Materials Sheet for either cholera or pertussis toxin

8.3 What is the LD_{50} (specify species) of the toxin_____Cholera toxin 250 ug for mice. Presume Pertussis is in the same overall range._____

8.4 How much of the toxin is handled at one time*? ____Cholera__10 ug____,Pertussis 5 ug_____

8.5 How much of the toxin is stored*? ___Cholera_____500 ug____Perussis 50 ug_____

8.6 Will any biological toxins be used in live animals? O YES, Please provide details:_____ xO NO

*For information on biosecurity requirements, please see:
http://www.uwo.ca/humanresources/docandform/docs/healthandsafety/biosafety/Biosecurity_Requirements.pdf
Web site does not seem to function

9.0 Insects

9.1 Do you use insects? O YES xO 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 life stage of the insect? _____

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

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

9.7 Do you use insects that require a permit from the CFIA permit? O YES O NO
If YES, Please attach the CFIA permit & describe any CFIA permit conditions:

_____

_____

10.0 Plants

10.1 Do you use plants? O YES xO 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
If YES, Please attach the CFIA permit & describe any CFIA permit conditions:

_____

_____

11.0 Import Requirements

11.1 Will any of the above agents be imported? xO YES, please give country of origin____USA_____ O NO
shRNA constructs are purchased from Thermo Open BioSystems.
If no, please proceed to Section 12.0

11.2 Has an Import Permit been obtained from HC for human pathogens? O YES O NO
No known human pathogens
11.3 Has an import permit been obtained from CFIA for animal or plant pathogens? O YES O NO
No known animal or plant pathogens
11.4 Has the import permit been sent to OHS? O YES, please provide permit # _____ 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 biological agents in Sections 1.0 to 9.0 have been trained.

SIGNATURE _____ Signature

13.0 Containment Levels

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

13.2 Has the facility been certified by OHS for this level of containment?
xO YES, date of most recent biosafety inspection: __Nov 2010_____
O NO, please certify
O NOT REQUIRED for Level 1 containment

13.3 Please indicate permit number (not applicable for first time applicants): BIO-LHRI-0020_____

14.0 Procedures to be Followed

14.1 Please describe additional risk reduction measures will be taken beyond containment level 1, 2, 2+ or 3 measures, that are unique to this agent.

_____
_____
_____

14.2 Please outline what will be done if there is an exposure to the biological agents listed, such as a needlestick injury or an accidental splash:

Will reread the UWO instructions as listed below and comply as indicated.__

3.5 MEDICAL PROCEDURES AND INCIDENT REPORTING

The following emergency response procedures shall be followed when a worker has been potentially exposed to a biohazardous agents via a needlestick, cut, animal bite or scratch, via mucous membrane contact, or via non-intact skin contact.

Worker

1. The exposed site must be washed immediately.
a) In case of a needlestick, cut, animal bite or scratch, wash with soap and water after allowing the wound to bleed freely.
b) If mucous (eyes, nose, mouth) membrane or non-intact (cuts, rash, eczema or dermatitis) skin contact, flush with water at the nearest faucet or eye wash station for a minimum of ten minutes.
2. The worker must immediately inform the Supervisor/Principal Investigator of the exposure incident.
3. The worker must seek prompt medical attention at Workplace Health (during the hours of operation), the nearest hospital emergency department or emergency clinic, or a Medical Practitioner of their choosing. Any information including the Material Safety Data Sheet or equivalent for the biohazardous agent must also be taken to the care provider.
4. The worker must provide information for a Accident/Incident Report (obtained from her/his Supervisor/Principal Investigator), describing the incident in detail, including the route of exposure and the emergency actions taken, and a description of the worker's duties as they relate to the exposure incident.

_____
_____

14.3 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 working in my laboratory have an up-to-date Hazard Communication Form, found at http://www.wph.uwo.ca/

SIGNATURE _____ Date: Signature

15.0 Approvals

1) UWO Biohazards Subcommittee: SIGNATURE: _____
Date: _____

2) Safety Officer for the University of Western Ontario
SIGNATURE:_____
Date:_____

3) Safety Officer for Institution where experiments will take place (if not UWO):
SIGNATURE:_____
Date:_____

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

Special Conditions of Approval:

Appendix

2.2

Biosafety level 2

This level is similar to Biosafety Level 1 and is suitable for work involving agents of moderate potential hazard to personnel and the environment.[7] It includes various bacteria and viruses that cause only mild disease to humans, or are difficult to contract via aerosol in a lab setting, such as *C. difficile*, most Chlamydiae, hepatitis A, B, and C, influenza A, Lyme disease, dengue fever, *Salmonella*, mumps, measles, HIV,[8] scrapie, MRSA, and VRSA. Genetically modified organisms have also been classified as level 2 organisms[citation needed], even if they pose no direct threat to humans. This designation is used to limit the release of modified organisms into the environment. Approval by the FDA is required to release these organisms. An example is genetically modified food crops. BSL-2 differs from BSL-1 in that:

1. laboratory personnel have specific training in handling pathogenic agents and are directed by scientists with advanced training;
2. access to the laboratory is limited when work is being conducted;
3. extreme precautions are taken with contaminated sharp items; and
4. certain procedures in which infectious aerosols or splashes may be created are conducted in biological safety cabinets or other physical containment equipment.

Material Safety Data Sheet

1. PRODUCT AND COMPANY IDENTIFICATION

Product Name Clones

Cat No. Clones
Synonyms No information available.
Recommended Use Laboratory chemicals

2. HAZARDS IDENTIFICATION

Target Organs Liver, Kidney
Potential Health Effects
Acute Effects
Principle Routes of Exposure
Eyes May cause irritation.
Skin May cause irritation. May be harmful in contact with skin.
Inhalation May cause irritation of respiratory tract. May be harmful if inhaled.
Ingestion May be harmful if swallowed. Ingestion may cause gastrointestinal irritation, nausea, vomiting and diarrhea.

Company
Thermo Fisher Scientific
Open Biosystem Products
601 Genone Way # 2100
Huntsville, AL 35806 United States
Tel: (303) 604-9499
Fax:(303) 604-9680
CAUTION!
Emergency Telephone Number
Chemtrec US: (800) 424-9300
Chemtrec EU: (202) 483-7616
Emergency Overview
Revision Number 1
May cause eye, skin, and respiratory tract irritation . Handle in accordance with good industrial hygiene and safety practice. The toxicological properties have not been fully investigated.
Creation Date 23-Apr-2009 **Revision Date** 23-Apr-2009
Appearance Yellow Brown **Physical State** Liquid. **Odor** No information available

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Chronic Effects Tumorigenic effects have been reported in experimental animals.. Experiments have shown reproductive toxicity effects on laboratory animals. May cause adverse liver effects. May cause adverse kidney effects.
See Section 11 for additional Toxicological information.
Aggravated Medical Conditions Preexisting eye disorders. Kidney disorders. Skin disorders.

3. COMPOSITION/INFORMATION ON INGREDIENTS

Haz/Non-haz
Component CAS-No Weight %
Peptones 73049-73-7 1
Water 7732-18-5 80 - 96
Sodium chloride 7647-14-5 1
Agar 9002-18-0 1 - 2
Yeast, ext. 8013-01-2 0.5
Glycerol 56-81-5 8

4. FIRST AID MEASURES

Eye Contact Rinse immediately with plenty of water, also under the eyelids, for at least 15 minutes. Obtain medical attention.
Skin Contact Wash off immediately with plenty of water for at least 15 minutes. Get medical attention immediately if symptoms occur.
Inhalation Move to fresh air. If breathing is difficult, give oxygen. Get medical attention immediately if symptoms occur.
Ingestion Do not induce vomiting. Obtain medical attention.
Notes to Physician Treat symptomatically.

5. FIRE-FIGHTING MEASURES

Flash Point Not applicable
Method No information available.
Autoignition Temperature No information available.
Explosion Limits
Upper No data available
Lower No data available
Suitable Extinguishing Media Substance is nonflammable; use agent most appropriate to extinguish surrounding fire..
Unsuitable Extinguishing Media No information available.

Hazardous Combustion Products No information available.
Sensitivity to mechanical impact No information available.
Sensitivity to static discharge No information available.

Specific Hazards Arising from the Chemical
Thermal decomposition can lead to release of irritating gases and vapors.
Protective Equipment and Precautions for Firefighters
As in any fire, wear self-contained breathing apparatus pressure-demand, MSHA/NIOSH (approved or equivalent) and full protective gear.

6. ACCIDENTAL RELEASE MEASURES

Personal Precautions Ensure adequate ventilation. Use personal protective equipment. Avoid contact with skin, eyes and clothing.
Environmental Precautions Should not be released into the environment.
Methods for Containment and Clean Up
Soak up with inert absorbent material. Keep in suitable and closed containers for disposal.

7. HANDLING AND STORAGE

Handling Ensure adequate ventilation. Wear personal protective equipment. Avoid contact with skin, eyes and clothing. Do not breathe vapors or spray mist.
Storage Keep container tightly closed in a dry and well-ventilated place. Long term. Keep at -80°C.

8. EXPOSURE CONTROLS / PERSONAL PROTECTION

Engineering Measures Ensure adequate ventilation, especially in confined areas. Ensure that eyewash stations and safety showers are close to the workstation location.
Exposure Guidelines
Component ACGIH TLV OSHA PEL NIOSH IDLH
Glycerol = 10 mg/m3 TWA = 10 mg/m3 TWA total dust
= 5 mg/m3 TWA respirable fraction
= 15 mg/m3 TWA total
Component Quebec Mexico OEL (TWA) Ontario TWAEV
Glycerol = 10 mg/m3 TWAEV mist = 10 mg/m3 TWA mist = 10 mg/m3 TWAEV
NIOSH IDLH: *Immediately Dangerous to Life or Health*
Personal Protective Equipment
Eye/face Protection Wear appropriate protective eyeglasses or chemical safety goggles as described by OSHA's eye and face protection regulations in 29 CFR 1910.133 or European Standard EN166
Skin and body protection Wear appropriate protective gloves and clothing to prevent skin exposure
Respiratory Protection Follow the OSHA respirator regulations found in 29 CFR 1910.134 or European Standard EN 149. Use a NIOSH/MSHA or European Standard EN 149 approved respirator if exposure limits are exceeded or if irritation or other symptoms are experienced.

9. PHYSICAL AND CHEMICAL PROPERTIES

Physical State Liquid
Appearance Yellow Brown
Odor No information available
Odor Threshold No information available.
pH Not applicable
Vapor Pressure No information available.
Vapor Density No information available.
Viscosity No information available.
Boiling Point/Range Not applicable
Melting Point/Range No information available.
Decomposition temperature °C No information available.
Flash Point Not applicable
Evaporation Rate No information available.
Specific Gravity No information available.
Solubility No information available.
log Pow No data available

10. STABILITY AND REACTIVITY

Stability Stable under normal conditions.
Conditions to Avoid Excess heat.
Incompatible Materials None known
Hazardous Decomposition Products None known
Hazardous Polymerization Hazardous polymerization does not occur.
Hazardous Reactions . None under normal processing..

11. TOXICOLOGICAL INFORMATION

Acute Toxicity
Component Information
Component LD50 Oral LD50 Dermal LC50 Inhalation
Water > 90 mL/kg Oral LD50 Rat > 90 mL/kg Oral LD50 Rat > 90 mL/kg Oral LD50 Rat
Sodium chloride > 42 g/m3 Inhalation LC50 Rat 1 h
> 10 g/kg Dermal LD50 Rabbit
= 3 g/kg Oral LD50 Rat
> 42 g/m3 Inhalation LC50 Rat 1 h
= 3 g/kg Oral LD50 Rat
> 10 g/kg Dermal LD50 Rabbit
> 42 g/m3 Inhalation LC50 Rat 1 h
> 42 g/m3 Inhalation LC50 Rat 1 h
> 10 g/kg Dermal LD50 Rabbit
= 3 g/kg Oral LD50 Rat
> 42 g/m3 Inhalation LC50 Rat 1 h
Agar 11000 g/kg (Rat) Not listed Not listed
Glycerol > 570 mg/m3 Inhalation LC50 Rat 1 h
> 21900 mg/kg Dermal LD50 Rat
= 12600 mg/kg Oral LD50 Rat
> 570 mg/m3 Inhalation LC50 Rat 1 h
= 12600 mg/kg Oral LD50 Rat
> 21900 mg/kg Dermal LD50 Rat
> 570 mg/m3 Inhalation LC50 Rat 1 h
> 570 mg/m3 Inhalation LC50 Rat 1 h
> 21900 mg/kg Dermal LD50 Rat
= 12600 mg/kg Oral LD50 Rat
> 570 mg/m3 Inhalation LC50 Rat 1 h

Irritation No information available.
Toxicologically Synergistic
Products
No information available.
Chronic Toxicity
Carcinogenicity There are no known carcinogenic chemicals in this product
Sensitization No information available.
Mutagenic Effects Mutagenic effects have occurred in experimental animals.
Reproductive Effects Experiments have shown reproductive toxicity effects on laboratory animals.
Developmental Effects No information available.
Teratogenicity No information available.
Other Adverse Effects Tumorigenic effects have been reported in experimental animals.. See actual entry in RTECS for complete information..
Endocrine Disruptor Information No information available

12. ECOLOGICAL INFORMATION

Ecotoxicity

.
Component Freshwater Algae Freshwater Fish Microtox Water Flea
Sodium chloride Not listed = 12946 mg/L LC50 Lepomis
macrochirus 96 h static
= 7650 mg/L LC50
Pimephales promelas 96 h
static
= 9675 mg/L LC50 Lepomis
macrochirus 96 h flowthrough
Not listed = 1000 mg/L EC50 Daphnia
magna 48 h
Glycerol Not listed 51000 - 57000 mg/L LC50
Oncorhynchus mykiss 96 h
Not listed > 500 mg/L EC50 Daphnia
magna 24 h
Persistence and Degradability No information available

Bioaccumulation/ Accumulation No information available
Mobility .
Component log Pow
Glycerol = -1.76

13. DISPOSAL CONSIDERATIONS

13. DISPOSAL CONSIDERATIONS

Waste Disposal Methods Chemical waste generators must determine whether a discarded chemical is classified as a hazardous waste. Chemical waste generators must also consult local, regional, and national hazardous waste regulations to ensure complete and accurate classification

14. TRANSPORT INFORMATION

DOT Not regulated
TDG Not regulated
IATA Not regulated
IMDG/IMO Not regulated

15. REGULATORY INFORMATION

All of the components in the product are on the following Inventory lists: All of the components in the product are on the following Inventory lists:
International Inventories
Component TSCA DSL NDSL EINECS ELINCS NLP PICCS ENCS AICS CHINA KECL
Peptones Present Present - - - Present - Present Present KE-28131
Water Present Present - - - Present - Present Present KE-35400
Sodium chloride Present Present - 231-598-3 - Present 1-236 Present Present KE-31387
Agar XU X - 232-658-1 - X - X X KE-00275 X
Yeast, ext. XU Present - - - Present - Present Present KE-05-1355
Glycerol Present Present - 200-289-5 - Present 2-242; 7-338 Present Present KE-29297
Legend:
X - Listed
E - Indicates a substance that is the subject of a Section 5(e) Consent order under TSCA.
F - Indicates a substance that is the subject of a Section 5(f) Rule under TSCA.
N - Indicates a polymeric substance containing no free-radical initiator in its inventory name but is considered to cover the designated polymer made with any free-radical initiator regardless of the amount used.

P - Indicates a commenced PMN substance
R - Indicates a substance that is the subject of a Section 6 risk management rule under TSCA.
S - Indicates a substance that is identified in a proposed or final Significant New Use Rule
T - Indicates a substance that is the subject of a Section 4 test rule under TSCA.
XU - Indicates a substance exempt from reporting under the Inventory Update Rule, i.e. Partial Updating of the TSCA Inventory Data Base Production and Site Reports (40 CFR 710(B).
Y1 - Indicates an exempt polymer that has a number-average molecular weight of 1,000 or greater.
Y2 - Indicates an exempt polymer that is a polyester and is made only from reactants included in a specified list of low concern reactants that comprises one of the eligibility criteria for the exemption rule.
U.S. Federal Regulations
TSCA 12(b) Not applicable
SARA 313
Not applicable

SARA 311/312 Hazardous Categorization
Acute Health Hazard No
Chronic Health Hazard No
Fire Hazard No
Sudden Release of Pressure Hazard No
Reactive Hazard No
Clean Water Act
Not applicable
Clean Air Act
Not applicable
OSHA
Not applicable
CERCLA
Not Applicable
California Proposition 65
This product does not contain any Proposition 65 chemicals.
State Right-to-Know
Component Massachusetts New Jersey Pennsylvania Illinois Rhode Island
Glycerol Present (mist) - Present - Toxic (mist); Flammable (mist)
U.S. Department of Transportation
Reportable Quantity (RQ): N
DOT Marine Pollutant N
DOT Severe Marine Pollutant N
U.S. Department of Homeland Security
This product does not contain any DHS chemicals.
Other International Regulations
Mexico - Grade No information available

Canada
This product has been classified in accordance with the hazard criteria of the Controlled Products Regulations (CPR) and the MSDS contains all the information required by the CPR.
WHMIS Hazard Class
Non-controlled

16. OTHER INFORMATION

Prepared By Regulatory Affairs
Thermo Fisher Scientific
Tel: (412) 490-8932
Creation Date 23-Apr-2009
Print Date 23-Apr-2009
Revision Summary "***", and red text indicates revision
Disclaimer
The information provided on this Safety Data Sheet is correct to the best of our knowledge, information and belief at the date of its publication. The information given is designed only as a guide for safe handling, use, processing, storage, transportation, disposal and release and is not to be considered as a warranty or quality specification. The information relates only to the specific material designated and may not be valid for such material used in combination with any other material or in any process, unless specified in the text.
End of MSDS

Thermo Scientific Open Biosystems Freedom ORF Clones and Collection (glycerol)

Catalog #: OHS1164, OHS1166, OHS1169

Introduction

Thermo Scientific Open Biosystems Freedom ORF clones are collections of high quality sequence verified genes cloned into vectors that are free from the IP constraints commonly found with other recombinational cloning systems. Each clone has been amplified and sequenced to ensure the highest quality standard. Open Biosystems offers a set of over 1,600 human Clontech BD Creator Universal Clones, which represents the initial release of the Freedom™ ORF collection.

The Clontech BD Creator™ system is a flexible recombinational cloning system based on Cre-loxP reactions. These ORF clones are provided to you in the pDNR-Dual vector, allowing you to move the inserts directly into any expression vector of choice through a simple Cre recombinase reaction. The resulting expression clone contains the ORF in both proper orientation and reading frame. The BD Creator™ Universal Clones are constructed for immediate 3' tagging with a 6xTN affinity tag. A large portion of the genes in this set are represented by two clones; one with a stop codon and one without, giving you the freedom to produce native or fusion proteins to tag the ORF.

Clone Storage

80°C indefinitely.

Product Description

96-well microtiter plates containing bacterial cultures of *E. coli* in LB broth with an inert growth indicator + 8% glycerol + ampicillin at a concentration of 100 µg/mL.

Making A Stock Culture

Once the clone has been streak isolated and the identity of the strain has been confirmed, we recommend making a stock of the pure culture. Grow the pure culture in LB broth + ampicillin at a concentration of 100 µg/mL. Transfer 920 uL of culture into a polypropylene tube and add 80 uL sterile glycerol to make an 8% glycerol freezing solution. Vortex the culture to evenly mix the glycerol throughout the culture. The culture can be stored indefinitely at -80°C.

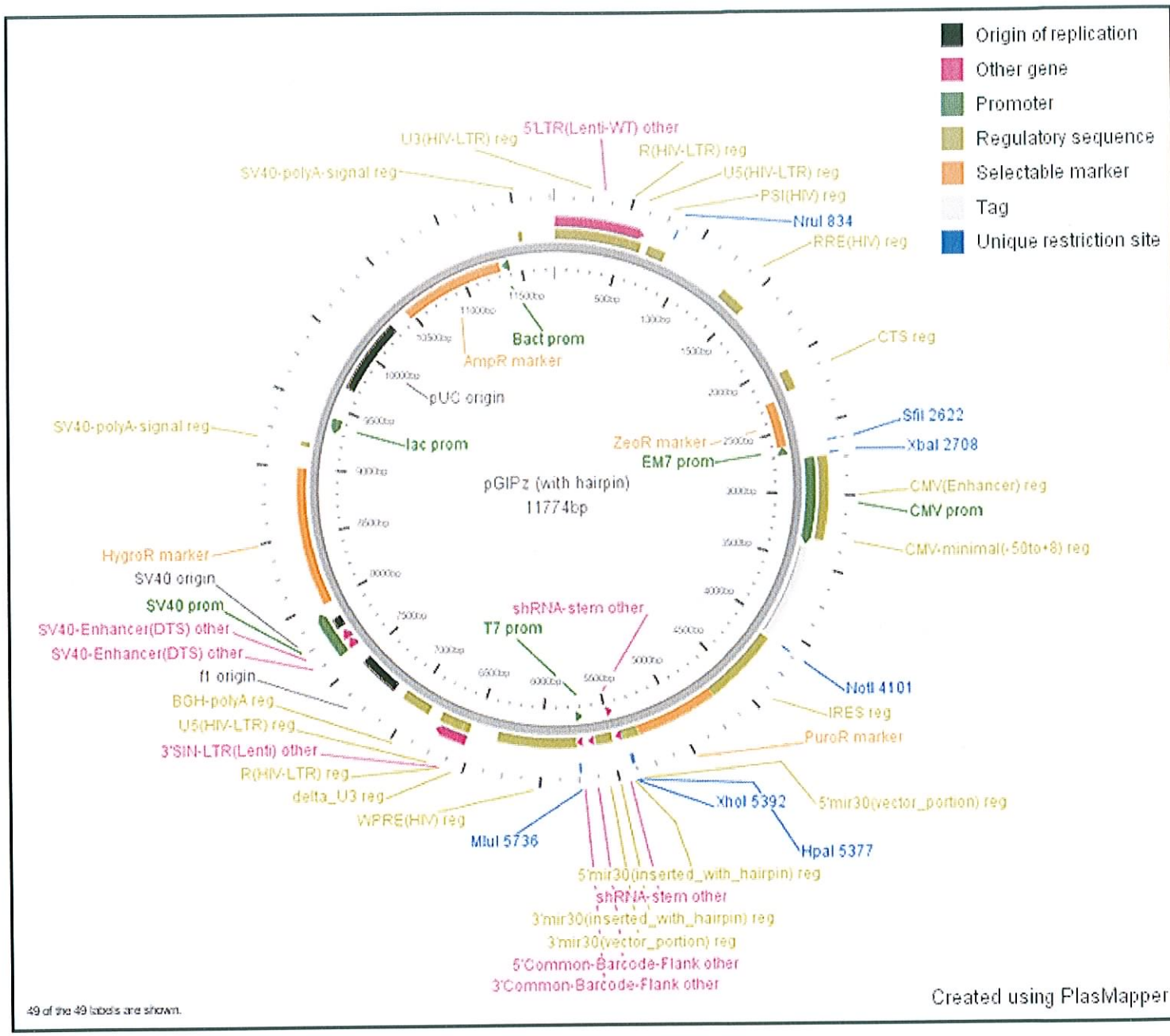
pDNR-Dual Vector map, Multiple Cloning Site, and Sequence Information

Sequence of pDNR-Dual available at:

http://www.clontech.com/techinfo/vectors/vectorsD/text/pDNR-Dual.txt

1

4.3A
pGIPZ



4.3B pLKO.1

Vector Information pLK0.1

The pLKO.1 HIV-based lentiviral vector (Figures 1-2, Table 1) allows for transient and stable transfection of shRNA and also the production of viral particles using lentiviral packaging cell lines. Stable cell lines can be selected using the puromycin selectable marker.

Human U6 Promoter RNA generated with four uridine overhangs at each 3' end

hPGK Human phosphoglycerate kinase promoter

PuroR Puromycin mammalian selectable marker

3' SIN LTR 3' self inactivating long terminal repeat (Shimada, *et al.* 1995)

f1 ori f1 origin of replication

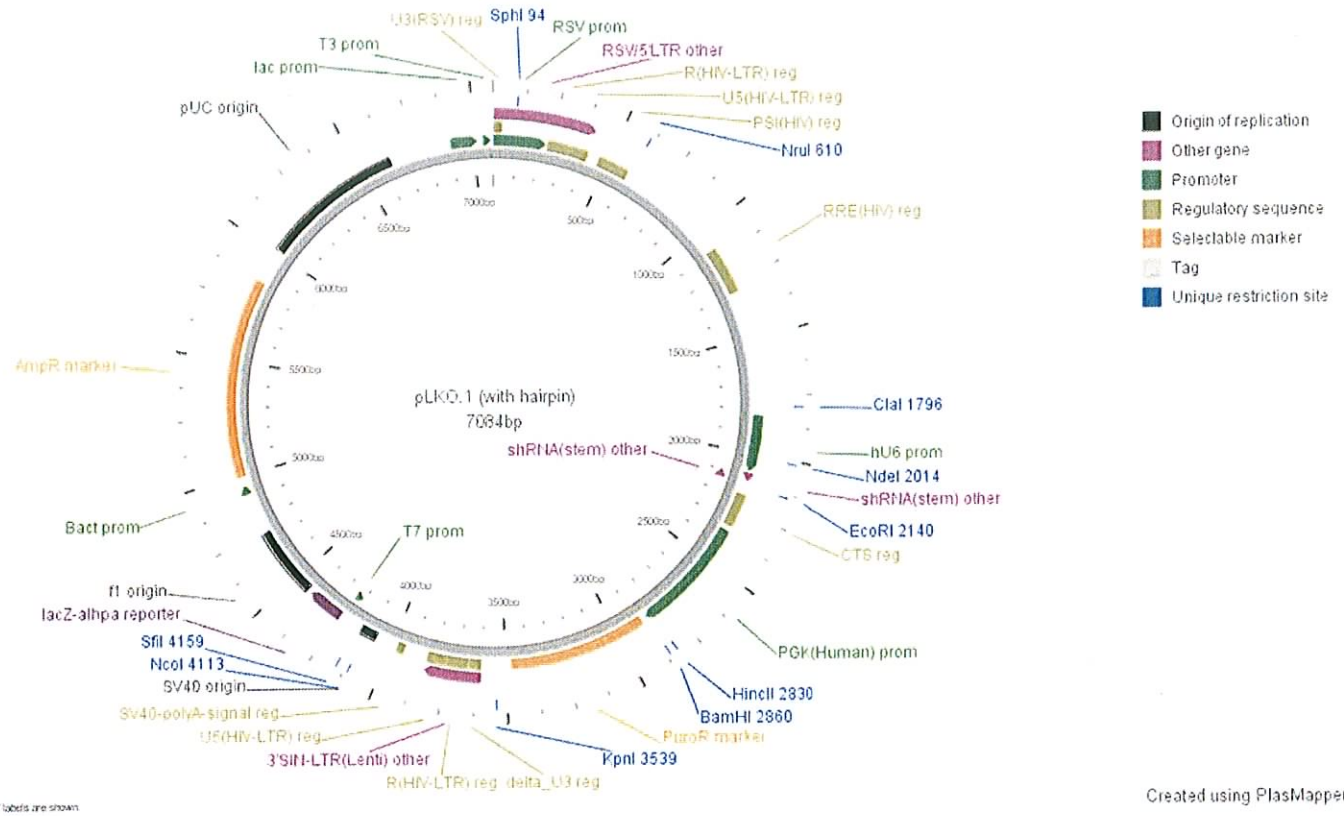
AmpR Ampicillin bacterial selectable marker

5'LTR 5' long terminal repeat

RRE Rev response element

cPPT Central polypurine tract
Table 1. Features pLKO.1 vector
Vector Map
Figure 1. The pLKO.1 vector

Figure 2. Map of the pLKO.1 vector



2

8.2A
Cholera Toxin from *Vibrio cholerae* ~95% (SDS-PAGE), lyophilized powder, 1×10^5-1×10^6 units/mg protein Toxin consisting of an A subunit (27 kDa) surrounded by five B subunits (approximately 12 kDa each), which attach the toxin to ganglioside GM_1 on the cell surface. The A subunit catalyzes ADP-ribosylation of the α-subunit of the stimulatory G protein ($G\alpha_s$), reducing GTPase activity and activating the α-subunit. This activation of $G\alpha_s$ leads to an increase in the activity of adenylyl cyclase, resulting in increased levels of cAMP. Also ADP-ribosylates transducin in the eye rod outer segments, inactivating its GTPase activity. Cholera toxin has also been reported to ADP-ribosylate tubulin. Shown to be a potent mucosal vaccine adjuvant, inducing T helper cell type 2 responses by inhibiting the production of interleukin-12.

8.2B
Pertussis toxin from *Bordetella pertussis* buffered aqueous glycerol solution Pertussis toxin catalyzes the ADP-ribosylation of the α subunits of the heterotrimeric guanine nucleotide regulatory proteins G_i, G_o, and G_t. This prevents the G protein heterotrimers from interacting with receptors, thus blocking their coupling and activation. Since the G_α subunits remain in their GDP-bound, inactive state, they are unable to inactivate adenylyl cyclase or open K^+ channels.

MSDS Request

MSDS Temporarily Unavailable for Cholera Toxin

We were unable to locate the MSDS you were searching for. The MSDS may be temporarily unavailable or you may have encountered an error in the product number. Please see the most common problems below for help in locating the MSDS.

Canadian Food Inspection Agency | Agence canadienne d'inspection des aliments

Office of Biohazard Containment and Safety
Science Branch, CFIA
59 Camelot 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
Direction générale des sciences, ACIA
59 promenade Camelot, Ottawa, Ontario K1A 0Y9
Tél: (613) 221-7068 Téléc: (613) 228-6129
Courriel: ImportZoopath@inspection.gc.ca

October 20th, 2009

Ms. Shamila Survery / Mr. Michael Decosimo
Cedarlane Laboratories Ltd
4410 Paletta Court
Burlington, Ontario L7L 5R2

By Facsimile: (289) 288-0020

SUBJECT: Importation of *Escherichia coli* strains

Dear Ms. Survery / Mr. Decosimo:

Our office received your query about the importation of *Escherichia coli* from the American Type Culture Collection (ATCC) located in Manassas, Virginia, United States. The following *Escherichia coli* strains are consider to be level 1 animal pathogens:

- 5K
- 58
- 58-161
- 679
- 1532
- AB284
- AB311
- AB1157
- AB1206
- AG1
- B
- BB4
- BD792
- BL21
- BL21 (DE3)
- BM25.8
- C
- C-1a
- C-3000
- C25
- C41 (DE3)
- C43 (DE3)
- C600
- Cavalli Hfr
- CIE85
- DH1
- DH10 GOLD
- DH10B
- DH5
- DH5-alpha
- DP50
- DY145
- DY380
- E11
- EJ183
- EL250
- EMG2
- EPI 300
- EZ10
- FDA Seattle 1946
- Fusion-Blue
- H1443
- HF4714
- HB101
- HS(PFAMP)R
- Hfr3000
- Hfr3000 X74
- HMS174
- J52
- J53
- JC3272
- JC7661
- JC9387
- JF1504
- JF1508
- JF1509
- JJ055
- JM83
- JM101
- JM109
- K12
- KC8
- KA802
- KAM32
- KAM33
- KAM43
- LE450
- LE451
- LE452
- MB408
- MBX1928
- MC1061
- MC4100 (MuLac)
- MG1655
- MM294
- MS101
- NC-7
- Nissle 1917
- One Shot STBL3
- OP50
- P678
- PA309
- PK-5
- PMC103
- PR13
- Rri
- RV308
- S17-1λ-PIR
- SCS1
- SMR10
- SOLR
- SuperchargeEZ10
- SURE
- TOP10
- TG1
- U5/41
- W208
- W945
- W1485
- W3104
- W3110
- WA704
- WP2
- X1854
- X2160T
- X2541
- X2547T
- XL1-BLUE
- XL1-BLUE-MRF
- XLOLR
- Y10
- Y1090 (1090)
- YN2980
- W3110
- WG1
- WG439
- WG443
- WG445

The Office of Biohazard Containment and Safety (BCS) of the Canadian Food Inspection Agency (CFIA) only issues import permits for microorganisms that are pathogenic to animals, or parts of microorganisms that are pathogenic to animals. As the products listed above are not considered pathogenic to animals, the Office of BCS does not have any regulatory requirements for their importation.

Please note that other legislation may apply. You may wish to contact the Public Health Agency of Canada's (PHAC) Office of Laboratory Security at (613) 957-1779.

Note: Microorganisms pathogenic to animals and veterinary biologics require an import permit from the CFIA.

Sincerely,

Cinthia Labrie
Head, Animal Pathogen Importation Program
Office of Biohazard Containment & Safety

Canada

Cell Line Info

Cell Biology

ATCC® Number:	**CCL-185™**	Order this Item	Price:	**$279.00**

Designations:	A549
Depositors:	M Lieber
Biosafety Level:	1
Shipped:	frozen
Medium & Serum:	See Propagation
Growth Properties:	adherent
Organism:	*Homo sapiens* (human)
Morphology:	epithelial 
Source:	**Organ:** lung **Disease:** carcinoma
Cellular Products:	keratin
Permits/Forms:	In addition to the MTA mentioned above, other ATCC and/or regulatory permits may be required for the transfer of this ATCC material. Anyone purchasing ATCC material is ultimately responsible for obtaining the permits. Please click here for information regarding the specific requirements for shipment to your location.
Isolation:	**Isolation date:** 1972
Applications:	transfection host (Nucleofection technology from Lonza Roche FuGENE® Transfection Reagents)
DNA Profile (STR):	Amelogenin: X,Y CSF1PO: 10,12 D13S317: 11 D16S539: 11,12 D5S818: 11 D7S820: 8,11 THO1: 8,9.3 TPOX: 8,11 vWA: 14
Cytogenetic Analysis:	This is a hypotriploid human cell line with the modal chromosome number of 66, occurring in 24% of cells. Cells with 64 (22%), 65, and 67 chromosome counts also occurred at relatively high frequencies; the rate with higher ploidies was low at 0.4%. There were 6 markers present in single copies in all cells. They include der(6)t(1;6) (q11;q27); ?del(6) (p23); del (11) (q21), del(2) (q11), M4 and M5. Most cells had two X and two Y chromosomes. However, one or both Y chromosomes were lost in 40% of 50 cells analyzed. Chromosomes N2 and N6 had single copies per cell; and N12 and N17 usually had 4 copies.
Isoenzymes:	G6PD, B
Age:	58 years

Related Links

NCBI Entrez Search

Cell Micrograph

Make a Deposit

Frequently Asked Questions

Material Transfer Agreement

Technical Support

Related Cell Culture Products

Login Required

Product Information Sheet

Cell Biology

ATCC® Number:	**CRL-2110™** [Order this Item]	Price:	**$379.00**

Designations:	MLE 12
Depositors:	JA Whitsett
Biosafety Level:	2 [CELLS CONTAIN PAPOVAVIRUS]
Shipped:	frozen
Medium & Serum:	See Propagation
Growth Properties:	adherent
Organism:	Mus musculus, transgenic (mouse, transgenic)
Morphology:	epithelial
Source:	**Organ:** lung **Strain:** FVB/N **Cell Type:** epithelialSV40 transformed
Cellular Products:	lung surfactant proteins B and C (SP-B, SP-C)
Permits/Forms:	In addition to the MTA mentioned above, other ATCC and/or regulatory permits may be required for the transfer of this ATCC material. Anyone purchasing ATCC material is ultimately responsible for obtaining the permits. Please click here for information regarding the specific requirements for shipment to your location.
Isolation:	**Isolation date:** 1992
Tumorigenic:	Yes
Age:	5 month old
Gender:	female
Comments:	This line was established in 1992 by Kathryn A. Wikenheiser from pulmonary tumors in a mouse transgenic for the SV40 large T antigen under the control of the promoter region of the human surfactant protein C gene. The cells express the mRNA for large T antigen. Lung surfactant proteins B and C were detected. The cells secrete phospholipids in response to phorbol esters and ATP but not in response to forskolin.

Propagation:

Related Links

NCBI Entrez Search
Make a Deposit
Frequently Asked Questions
Material Transfer Agreement
Technical Support
Related Cell Culture Products

BioProducts

Cell, microbial and molecular genomics products for the life sciences

BioServices

Bio-materials management; basic repository to complex partnership-level services

BioStandards

Biological Reference Material and Consensus Standards for the life science community

Cell Biology

ATCC® Number:	**CRL-1573™**	Order this Item	Price:	**$279.00**

Designations:	293 [HEK-293]
Depositors:	FL Graham
Biosafety Level:	2 [CELLS CONTAIN ADENOVIRUS]
Shipped:	frozen
Medium & Serum:	See Propagation
Growth Properties:	adherent
Organism:	*Homo sapiens* (human)
Morphology:	epithelial 
Source:	**Organ:** embryonic kidney **Cell Type:** transformed with adenovirus 5 DNA
Permits/Forms:	In addition to the MTA mentioned above, other ATCC and/or regulatory permits may be required for the transfer of this ATCC material. Anyone purchasing ATCC material is ultimately responsible for obtaining the permits. Please click here for information regarding the specific requirements for shipment to your location.
Restrictions:	These cells are distributed for research purposes only. 293 cells, their products, or their derivatives may not be distributed to third parties.
Applications:	efficacy testing [92587] transfection host (Nucleofection technology from Lonza Roche FuGENE® Transfection Reagents) viruscide testing [92579]
Receptors:	vitronectin, expressed
Tumorigenic:	YES
DNA Profile (STR):	Amelogenin: X CSF1PO: 11,12 D13S317: 12,14 D16S539: 9,13 D5S818: 8,9 D7S820: 11,12 THO1: 7,9.3 TPOX: 11 vWA: 16,19
Cytogenetic Analysis:	

Related Links

NCBI Entrez Search

Cell Micrograph

Make a Deposit

Frequently Asked Questions

Material Transfer Agreement

Technical Support

Related Cell Culture Products

Login Required

Product Information Sheet

Western

Toxin Info

TOXIN USE RISK ASSESSMENT

Name of Toxin:	Cholera toxin
Proposed Use Dose:	10 µg
Proposed Storage Dose:	500 µg
LD_{50} (species):	250 µg

Calculation:

250 µg/kg x 50 kg/person

Dose per person based on LD_{50} in µg = 12500

LD_{50} per person with safety factor of 10 based on LD_{50} in µg = 1250

Comments/Recommendations:

Toxins of Biological Origin

Biological toxins are produced by certain bacteria, fungi, protozoa, plants, reptiles, amphibians, fish, echinoderma (spiny urchins and starfish), mollusks, and insects.

The EH&S Biosafety Office regulates the **possession, use, and transfer of unfractionated mixtures and purified preparations of biological toxins with a mammalian LD_{50} of ≤ 100 ug/kg body weight, as well as the organisms, both natural and recombinant, which produce these biological toxins.** These are called "Acute Toxins". Registration forms can be found at http://www.ehs.ufl.edu/Bio/default.asp

The following table from the UF EH&S Biological Safety Manual lists LD_{50} values for some biological toxins. Toxins not on this list may still require registration. For more information, please contact the Biosafety Office at 392-1591.

Toxin	LD50 (ug/kg)*
Abrin	0.7
Aerolysin	7.0
Botulinin toxin A	0.0012
Botulinin toxin B	0.0012
Botulinin toxin C1	0.0011
Botulinin toxin C2	0.0012
Botulinin toxin D	0.0004
Botulinin toxin E	0.0011
Botulinin toxin F	0.0025
b-bungarotoxin	14.0
Caeruleotoxin	53
Cereolysin	40-80
Cholera toxin	250
Clostridium difficile enterotoxin A	0.5
Clostridium difficile cytotoxin B	220
Clostridium perfringens lecithinase	3
Clostridium perfringens kappa toxin	1500
Clostridium perfringens perfringolysin O	13-16
Clostridium perfringens enterotoxin	81
Clostridium perfringens beta toxin	0.4
Clostridium perfringens delta toxin	5
Clostridium perfringens epsilon toxin	0.1
Conotoxin	12-30
Crotoxin	82
Diphtheria toxin	0.1
Listeriolysin	3-12
Leucocidin	50

Modeccin	1-10
Nematocyst toxins	33-70
Notexin	25
Pertussis toxin	15
Pneumolysin	1.5
Pseudomonas aeruginosa toxin A	3
Ricin	2.7
Saxitoxin	8
Shiga toxin	20
Shigella dysenteriae neurotoxin	1.3
Streptolysin O	8
Staphylococcus enterotoxin B	25
Staphylococcus enterotoxin F	2-10
Streptolysin S	25
Taipoxin	2
Tetanus toxin	0.001
Tetrodotoxin	8
Viscumin	2.4-80
Volkensin	1.4
Yersinia pestis murine toxin	10

*Please note that the LD50 values are from a number of sources (see below). For specifics on route of application (i.v., i.p., s.c.), animal used, and variations on the listed toxins, please go to the references listed below.

Reference:
1. Gill, D. Michael; 1982; Bacterial toxins: a table of lethal amounts; Microbiological Reviews; 46: 86-94
2. Stirpe, F.; Luigi Barbieri; Maria Giulia Battelli, Marco Soria and Douglas A. Lappi; 1992; Ribosome-inactivating proteins from plants: present status and future prospects; Biotechnology; 10: 405-412
3. Registry of toxic effects of chemical substances (RTECS): comprehensive guide to the RTECS. 1997. Doris V. Sweet, ed., U.S. Dept of Health and Human Services, Public Health Service, Centers for Disease Control and Prevention, National Institute for Occupational Safety and Health; Cincinnati, Ohio

More Examples of Biological Toxins Which May Require Registration

Aflatoxins	Leiurotoxins
Amanitin	Lipid A - all types
Amphibian venoms	Lipopolysaccharides from all species
Anatoxin A	Maitotoxin
Anthrax toxin	Medamine
Aspergillus sp toxins	Microcystins
Bacillus sp. toxins - all	Mojave toxin
Bordetella sp. toxins	Mycotoxins - all
Botulinum toxins - all	Myotoxins
Brevetoxins	Neurotoxins - all
Bungarotoxins	Notexin
Cardiotoxin	Nodularin
Charybdotoxin	Ochratoxin

TOXIN USE RISK ASSESSMENT

Name of Toxin:	Pertussis
Proposed Use Dose:	5 µg
Proposed Storage Dose:	50 µg
LD_{50} (species):	15 µg

Calculation:

15 µg/kg x 50 kg/person

Dose per person based on LD_{50} in µg = 750

LD_{50} per person with safety factor of 10 based on LD_{50} in µg = 75

Comments/Recommendations:

Material Safety Data Sheet

Version 4.2
Revision Date 11/05/2010
Print Date 04/01/2011

1. PRODUCT AND COMPANY IDENTIFICATION

Product name	: Cholera Toxin Vibrio cholerae		
Product Number	: C8052		
Brand	: Sigma		
Product Use	: For laboratory research purposes.		
Supplier	: Sigma-Aldrich Canada, Ltd 2149 Winston Park Drive OAKVILLE ON L6H 6J8 CANADA	Manufacturer	: Sigma-Aldrich Corporation 3050 Spruce St. St. Louis, Missouri 63103 USA
Telephone	: +19058299500		
Fax	: +19058299292		
Emergency Phone # (For both supplier and manufacturer)	: 1-800-424-9300		
Preparation Information	: Sigma-Aldrich Corporation Product Safety - Americas Region 1-800-521-8956		

2. HAZARDS IDENTIFICATION

Emergency Overview

Target Organs

Bowel

WHMIS Classification

D2B Toxic Material Causing Other Toxic Effects — Moderate skin irritant; Moderate eye irritant

GHS Classification

Acute toxicity, Oral (Category 5)
Skin irritation (Category 2)
Eye irritation (Category 2A)
Specific target organ toxicity - single exposure (Category 3)

GHS Label elements, including precautionary statements

Pictogram

Signal word: Warning

Hazard statement(s)

H303	May be harmful if swallowed.
H315	Causes skin irritation.
H319	Causes serious eye irritation.
H335	May cause respiratory irritation.

Precautionary statement(s)

P261	Avoid breathing dust/ fume/ gas/ mist/ vapours/ spray.
P305 + P351 + P338	IF IN EYES. Rinse cautiously with water for several minutes. Remove contact lenses, if present and easy to do. Continue rinsing.

HMIS Classification

Health hazard: 2

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

Potential Health Effects

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

3. COMPOSITION/INFORMATION ON INGREDIENTS

Synonyms : Cholera enterotoxin
Cholergen

CAS-No.	EC-No.	Index-No.	Concentration
Tris (hydroxymethyl) aminomethane			
77-86-1	201-064-4	-	>= 5.82 - <= 5.94 %
2-Amino-2-(hydroxymethyl)propane-1,3-diol hydrochloride			
1185-53-1	214-684-5	-	>= 31.3 - <= 31.9 %
Sodium chloride			
7647-14-5	231-598-3	-	>= 57.6 - <= 58.8 %
Exotoxin, vibrio cholerae			
9012-63-9	-	-	>= 0.5 - <= 2.5 %
Edetate disodium dihydrate			
6381-92-6	205-358-3	-	>= 0.96 - <= 0.98 %

4. FIRST AID MEASURES

General advice
Consult a physician. Show this safety data sheet to the doctor in attendance.Move out of dangerous area.

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

Conditions of flammability
Not flammable or combustible.

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.

Hazardous combustion products
Hazardous decomposition products formed under fire conditions. - Nature of decomposition products not known.

Explosion data - sensitivity to mechanical impact
no data available

Explosion data - sensitivity to static discharge
no data available

6. ACCIDENTAL RELEASE MEASURES

Personal precautions
Use personal protective equipment. Avoid dust formation. Avoid breathing vapors, mist or gas. Ensure adequate ventilation. Evacuate personnel to safe areas. Avoid breathing dust.

Environmental precautions
Do not let product enter drains.

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

7. HANDLING AND STORAGE

Precautions for safe handling
Avoid contact with skin and eyes. Avoid formation of dust and aerosols.
Provide appropriate exhaust ventilation at places where dust is formed. Normal measures for preventive fire protection.

Conditions for safe storage
Keep container tightly closed in a dry and well-ventilated place.

8. EXPOSURE CONTROLS/PERSONAL PROTECTION

Contains no substances with occupational exposure limit values.

Personal protective equipment

Respiratory protection
For nuisance exposures use type P95 (US) or type P1 (EU EN 143) particle respirator.For higher level protection use type OV/AG/P99 (US) or type ABEK-P2 (EU EN 143) respirator cartridges. Use respirators and components tested and approved under appropriate government standards such as NIOSH (US) or CEN (EU).

Hand protection
Handle with gloves. Gloves must be inspected prior to use. Use proper glove removal technique (without touching glove's outer surface) to avoid skin contact with this product. Dispose of contaminated gloves after use in accordance with applicable laws and good laboratory practices. Wash and dry hands.

Eye protection
Safety glasses with side-shields conforming to EN166 Use equipment for eye protection tested and approved under appropriate government standards such as NIOSH (US) or EN 166(EU).

Skin and body protection
impervious clothing, The type of protective equipment must be selected according to the concentration and amount of the dangerous substance at the specific workplace.

Hygiene measures
Handle in accordance with good industrial hygiene and safety practice. Wash hands before breaks and at the end of workday.

Specific engineering controls
Use mechanical exhaust or laboratory fumehood to avoid exposure.

9. PHYSICAL AND CHEMICAL PROPERTIES

Appearance

Form	solid
Colour	no data available

Safety data

pH	no data available
Melting/freezing	no data available

point	
Boiling point	no data available
Flash point	no data available
Ignition temperature	no data available
Autoignition temperature	no data available
Lower explosion limit	no data available
Upper explosion limit	no data available
Vapour pressure	no data available
Density	no data available
Water solubility	no data available
Partition coefficient: n-octanol/water	no data available
Relative vapour density	no data available
Odour	no data available
Odour Threshold	no data available
Evaporation rate	no data available

10. STABILITY AND REACTIVITY

Chemical stability
Stable under recommended storage conditions.

Possibility of hazardous reactions
no data available

Conditions to avoid
no data available

Materials to avoid
Dimethyl sulfate, Acid chlorides, Halogenated hydrocarbon, Metals, Acids

Hazardous decomposition products
Hazardous decomposition products formed under fire conditions. - Nature of decomposition products not known.

11. TOXICOLOGICAL INFORMATION

Acute toxicity

Oral LD50
no data available

Inhalation LC50
no data available

Dermal LD50
no data available

Other information on acute toxicity
no data available

Skin corrosion/irritation
no data available

Serious eye damage/eye irritation
Eyes: no data available

Respiratory or skin sensitization
no data available

Germ cell mutagenicity
no data available

Carcinogenicity

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.

ACGIH: No component of this product present at levels greater than or equal to 0.1% is identified as a carcinogen or potential carcinogen by ACGIH.

Reproductive toxicity

no data available

Teratogenicity

no data available

Specific target organ toxicity - single exposure (Globally Harmonized System)
no data available

Specific target organ toxicity - repeated exposure (Globally Harmonized System)
no data available

Aspiration hazard
no data available

Potential health effects

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

Synergistic effects
no data available

Additional Information
RTECS: Not available

12. ECOLOGICAL INFORMATION

Toxicity

no data available

Persistence and degradability
no data available

Bioaccumulative potential
no data available

Mobility in soil
no data available

PBT and vPvB assessment
no data available

Other adverse effects

no data available

13. DISPOSAL CONSIDERATIONS

Product
Offer surplus and non-recyclable solutions to a licensed disposal company. Contact a licensed professional waste disposal service to dispose of this material.

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.
Exotoxin, vibrio cholerae	9012-63-9

WHMIS Classification

D2B	Toxic Material Causing Other Toxic Effects	Moderate skin irritant Moderate eye irritant

This product has been classified in accordance with the hazard criteria of the Controlled Products Regulations and the MSDS contains all the information required by the Controlled Products Regulations.

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.

SIGMA-ALDRICH

MATERIAL SAFETY DATA SHEET

Date Printed: 04/01/2011
Date Updated: 10/03/2008
Version 1.6

Section 1 - Product and Company Information

Product Name	PERTUSSIS TOXIN
Product Number	P0317
Brand	SIGMA
Company	Sigma-Aldrich Canada, Ltd
Address	2149 Winston Park Drive Oakville ON L6H 6J8 CA
Technical Phone:	9058299500
Fax:	9058299292
Emergency Phone:	800-424-9300

Section 2 - Composition/Information on Ingredient

Substance Name	CAS #	SARA 313
PERTUSSIS TOXIN	70323-44-3	No

Synonyms: Histamine-sensitizing factor * IAP * Islet activating protein * Lymphocytosis-promoting factor * Pertussigen

RTECS Number: XW5883750

Section 3 - Hazards Identification

EMERGENCY OVERVIEW

Biohazard. Toxic.
Toxic by inhalation, in contact with skin and if swallowed.
Biomedical material. May cause human disease. Target organ(s): Pancreas.

HMIS RATING

HEALTH: 3*
FLAMMABILITY: 0
REACTIVITY: 0

NFPA RATING

HEALTH: 3
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 immediately.

INHALATION EXPOSURE

If inhaled, remove to fresh air. If not breathing give

artificial respiration. If breathing is difficult, give oxygen.

DERMAL EXPOSURE
In case of skin contact, flush with copious amounts of water for at least 15 minutes. Remove contaminated clothing and shoes. Call a physician.

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.

METHODS FOR CLEANING UP
Inactivate spilled material with 5% sodium hypochlorite. Place in appropriate container. 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: Use respirators and components tested and approved under appropriate government standards such as NIOSH (US) or CEN (EU). 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.
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

Property	Value	At Temperature or Pressure
pH	N/A	
BP/BP Range	N/A	
MP/MP Range	N/A	
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	N/A	

N/A = not available

Section 10 - Stability and Reactivity

STABILITY
Stable: Stable.
Materials to Avoid: Strong oxidizing agents.

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

HAZARDOUS POLYMERIZATION
Hazardous Polymerization: Will not occur

Section 11 - Toxicological Information

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

TARGET ORGAN(S) OR SYSTEM(S)
Pancreas.

CONDITIONS AGGRAVATED BY EXPOSURE
Potentially neurotoxic.

ADDITIONAL INFORMATION
Pertussis toxin is one of the active factors responsible for whooping cough. This toxin causes potentiation of insulin-releasing responses, promotion of lipolysis on adipocytes, inhibition of epinephrine-induced hyperglycemia, ADP-ribosylation of cell-membrane protein, activation of adenylate cyclase, lymphocytosis-promoting activity, histamine-sensitizing activity, hemaggltination activity, and adjuvant activity.

TOXICITY DATA
Oral
Mouse
0.0018 mg/kg
LD50
Remarks: Pertussis toxin is one of the active factors responsible for whooping cough. This toxin causes potentiation of insulin-releasing responses, promotion of lipolysis on adipocytes, inhibition of epinephrine-induced hyperglycemia, ADP-ribosylation of cell-membrane protein, activation of adenylate cyclase, lymphocytosis-promoting activity, histamine-sensitizing activity, hemaggltination activity, and adjuvant activity.

Intravenous
Rat
114 UG/KG
LD50
Remarks: Nutritional and Gross Metabolic:Weight loss or decreased weight gain. Behavioral:Change in motor activity (specific assay). Sense Organs and Special Senses (Nose, Eye, Ear, and Taste):Eye:Lacrimation.

Intraperitoneal
Mouse
17160 NG/KG
LD50

Intravenous
Mouse
127 UG/KG
LD50
Remarks: Sense Organs and Special Senses (Nose, Eye, Ear, and Taste):Eye:Lacrimation. Behavioral:Change in motor activity (specific assay). Nutritional and Gross Metabolic:Weight loss or decreased weight gain.

Section 12 Ecological Information

No data available.

Section 13 Disposal Considerations

APPROPRIATE METHOD OF DISPOSAL OF SUBSTANCE OR PREPARATION
Disposal should be made in accordance with existing disposal practices employed for infectious wastes at your institution. Observe all federal, state, and local environmental regulations.

Section 14 Transport Information

DOT
Proper Shipping Name: Toxins, extracted from living sources, solid, n.o.s.
UN#: 3462
Class: 6.1
Packing Group: Packing Group I
Hazard Label: Toxic substances
PIH: Not PIH

IATA
Proper Shipping Name: Toxins, extracted from living sources, solid, n.o.s.
IATA UN Number: 3462
Hazard Class: 6.1
Packing Group: I

Section 15 Regulatory Information

EU ADDITIONAL CLASSIFICATION
Symbol of Danger: T
Indication of Danger: Toxic
R: 23/24/25
Risk Statements: Toxic by inhalation, in contact with skin and if swallowed.
S: 45-36/37/39
Safety Statements: In case of accident or if you feel unwell, seek medical advice immediately (show the label where possible). Wear suitable protective clothing, gloves, and eye/face protection.

US CLASSIFICATION AND LABEL TEXT
Indication of Danger: Biohazard, Toxic
Risk Statements: Toxic by inhalation, in contact with skin and if swallowed.
Safety Statements: In case of accident or if you feel unwell, seek medical advice immediately (show the label where possible). Wear suitable protective clothing, gloves, and eye/face protection.
US Statements: Biomedical material. May cause human disease.
Target organ(s): Pancreas.

UNITED STATES REGULATORY INFORMATION
SARA LISTED: No

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. 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 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.