



**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 isopropanol where appropriate, 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<sub>7</sub> 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<sub>7</sub> cells grow well at 37°C but stop dividing and differentiate at 41°C. During our studies, we discovered that this T<sub>7</sub> 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 transiently infect cells with lentiviral pGFZ and pLKO.1 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 also establish a new and convenient platform for studying Type 1 cell function.

## 1.0 Microorganisms

1.1 Does your work involve the use of biological agents?  YES  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?  YES  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:

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Please attach the CFIA permit.

Please describe any CFIA permit conditions:

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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
<u>E. coli</u> ( <u>dH5alpha</u> )	<input type="radio"/> Yes <input checked="" type="radio"/> No	<input type="radio"/> Yes <input checked="" type="radio"/> No	<input type="radio"/> Yes <input checked="" type="radio"/> No	<u>1-2 liters</u>	Commercial or <u>In House</u>	<input checked="" type="radio"/> 1 <input type="radio"/> 2 <input type="radio"/> 2+ <input type="radio"/> 3
	<input type="radio"/> Yes <input type="radio"/> No	<input type="radio"/> Yes <input type="radio"/> No	<input type="radio"/> Yes <input type="radio"/> No			<input type="radio"/> 1 <input type="radio"/> 2 <input type="radio"/> 2+ <input type="radio"/> 3
	<input type="radio"/> Yes <input type="radio"/> No	<input type="radio"/> Yes <input type="radio"/> No	<input type="radio"/> Yes <input type="radio"/> No			<input type="radio"/> 1 <input type="radio"/> 2 <input type="radio"/> 2+ <input type="radio"/> 3
	<input type="radio"/> Yes <input type="radio"/> No	<input type="radio"/> Yes <input type="radio"/> No	<input type="radio"/> Yes <input type="radio"/> No			<input type="radio"/> 1 <input type="radio"/> 2 <input type="radio"/> 2+ <input type="radio"/> 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  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	<input type="radio"/> Yes <input checked="" type="radio"/> No		
Rodent	<input type="radio"/> Yes <input checked="" type="radio"/> No		
Non-human primate	<input type="radio"/> Yes <input checked="" type="radio"/> No		
Other (specify)	<input type="radio"/> Yes <input checked="" type="radio"/> 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	<input checked="" type="radio"/> Yes <input type="radio"/> No	<u>A549,H440</u> (lung epithelial)	<u>1 or 2?</u>	<u>ATCC,In house,</u>
Rodent	<input checked="" type="radio"/> Yes <input type="radio"/> No	<u>MLE-12,</u>  T <sub>7</sub> (mouse lung epithelial, SV40 large T antigen)	<u>2</u>	<u>Jeff Whitsett who developed this cell line. It is now available from ATCC</u> Martin Post, Paediatrics, H Sick Children
Non-human primate	<input type="radio"/> Yes <input checked="" type="radio"/> No			
Other (specify)	<input type="radio"/> Yes <input checked="" type="radio"/> 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](http://www.atcc.org))

2.4 For above named cell type(s) indicate PHAC or CFIA containment level required     1     2     2+     3

### 3.0 Use of Human Source Materials

3.1 Does your work involve the use of human source materials?     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 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		<input type="radio"/> Yes <input type="radio"/> Unknown		<input type="radio"/> 1 <input type="radio"/> 2 <input type="radio"/> 2+ <input type="radio"/> 3
Human Blood (fraction) or other Body Fluid		<input type="radio"/> Yes <input type="radio"/> Unknown		<input type="radio"/> 1 <input type="radio"/> 2 <input type="radio"/> 2+ <input type="radio"/> 3
Human Organs or Tissues (unpreserved)		<input type="radio"/> Yes <input type="radio"/> Unknown		<input type="radio"/> 1 <input type="radio"/> 2 <input type="radio"/> 2+ <input type="radio"/> 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?     YES     NO    If no, please proceed to Section 5.0

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

Bacteria Used for Cloning *	Plasmid(s) **	Source of Plasmid	Gene Transfected	Describe the change that results from transformation or tranfection
<u>Ecoli (dH5 alpha)</u>		<u>In house</u>	<u>LPP1-3</u>	<u>.no changes noted</u>

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

\*\* Please attach a plasmid map.

See Appendix added at end for plasmid maps



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

6.5 Will the agent(s) be shed by the animal:  YES  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)?  YES  No If no, please proceed to section 8.0

7.2 Will live animals be used?  YES  No

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

- ◆ Pound source dogs  YES  NO
- ◆ Pound source cats  YES  NO
- ◆ Cattle, sheep or goats  YES, please specify species \_\_\_\_\_  NO
- ◆ Non-human primates  YES, please specify species \_\_\_\_\_  NO
- ◆ Wild caught animals  YES, please specify species & colony # \_\_\_In some cases we may examine extracts from the 13-striped ground squirrel. These extracts are from a collaboration with Dr. Jim Staples BIOLOGY and Ruud Veldhuizen Lawson Research Institute, UWO. \_\_\_\_\_  NO
- ◆ Birds  YES, please specify species \_\_\_\_\_  NO
- ◆ Others (wild or domestic)  YES, please specify \_\_\_We may also obtain lungs from investigators using Xenopus \_\_\_laevis (e.g. Tom Drysdale, VRL, UWO) to extract surfactant \_\_\_\_\_  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?  YES  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

8.3 What is the LD<sub>50</sub> (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?  YES, Please provide details: \_\_\_\_\_  NO

\*For information on biosecurity requirements, please see:

[http://www.uwo.ca/humanresources/docandform/docs/healthandsafety/biosafety/Biosecurity\\_Requirements.pdf](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?  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 life stage of the insect? \_\_\_\_\_

9.5 What is your intention?  Initiate and maintain colony, give location: \_\_\_\_\_

"One-time" use, give location: \_\_\_\_\_

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

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9.7 Do you use insects that require a permit from the CFIA permit?  YES  NO

If YES, Please attach the CFIA permit & describe any CFIA permit conditions:

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## 10.0 Plants

10.1 Do you use plants?  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?  Grow and maintain a crop  "One-time" use

10.6 Do you do any modifications to the plant?  YES  NO

If yes, please describe: \_\_\_\_\_

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

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10.8 Is the CFIA permit attached?  YES  NO

If YES, Please attach the CFIA permit & describe any CFIA permit conditions:

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## 11.0 Import Requirements

11.1 Will any of the above agents be imported?  YES, please give country of origin USA  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?  YES  NO

No known human pathogens

11.3 Has an import permit been obtained from CFIA for animal or plant pathogens?  YES  NO

No known animal or plant pathogens

11.4 Has the import permit been sent to OHS?  YES, please provide permit # \_\_\_\_\_  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(s)

**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.  1  2  2+  3

13.2 Has the facility been certified by OHS for this level of containment?  
 YES, date of most recent biosafety inspection: Nov 2010  
 NO, please certify  
 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.<sup>[7]</sup> 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,<sup>[8]</sup> scrapie, MRSA, and VRSA. Genetically modified organisms have also been classified as level 2 organisms<sup>[citation needed]</sup>, 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.

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### 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.

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#### 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/m<sup>3</sup> TWA = 10 mg/m<sup>3</sup> TWA total dust

= 5 mg/m<sup>3</sup> TWA respirable fraction

= 15 mg/m<sup>3</sup> TWA total

**Component Quebec Mexico OEL (TWA) Ontario TWAEV**

Glycerol = 10 mg/m<sup>3</sup> TWAEV mist = 10 mg/m<sup>3</sup> TWA mist = 10 mg/m<sup>3</sup> 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.

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Physical hazards N/A

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NFPA

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Health 1 Flammability 0 Instability 0

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### 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/m<sup>3</sup> Inhalation LC50 Rat 1 h

> 10 g/kg Dermal LD50 Rabbit

= 3 g/kg Oral LD50 Rat

> 42 g/m<sup>3</sup> Inhalation LC50 Rat 1 h

= 3 g/kg Oral LD50 Rat

> 10 g/kg Dermal LD50 Rabbit

> 42 g/m<sup>3</sup> Inhalation LC50 Rat 1 h

> 42 g/m<sup>3</sup> Inhalation LC50 Rat 1 h

> 10 g/kg Dermal LD50 Rabbit

= 3 g/kg Oral LD50 Rat

> 42 g/m<sup>3</sup> Inhalation LC50 Rat 1 h

Agar 11000 g/kg ( Rat ) Not listed Not listed

Glycerol > 570 mg/m<sup>3</sup> Inhalation LC50 Rat 1 h

> 21900 mg/kg Dermal LD50 Rat

= 12600 mg/kg Oral LD50 Rat

> 570 mg/m<sup>3</sup> Inhalation LC50 Rat 1 h

= 12600 mg/kg Oral LD50 Rat

> 21900 mg/kg Dermal LD50 Rat

> 570 mg/m<sup>3</sup> Inhalation LC50 Rat 1 h

> 570 mg/m<sup>3</sup> Inhalation LC50 Rat 1 h

> 21900 mg/kg Dermal LD50 Rat

= 12600 mg/kg Oral LD50 Rat

> 570 mg/m<sup>3</sup> Inhalation LC50 Rat 1 h

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**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

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### 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.

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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

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#### 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

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# 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 µL of culture into a polypropylene tube and add 80 µL 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.

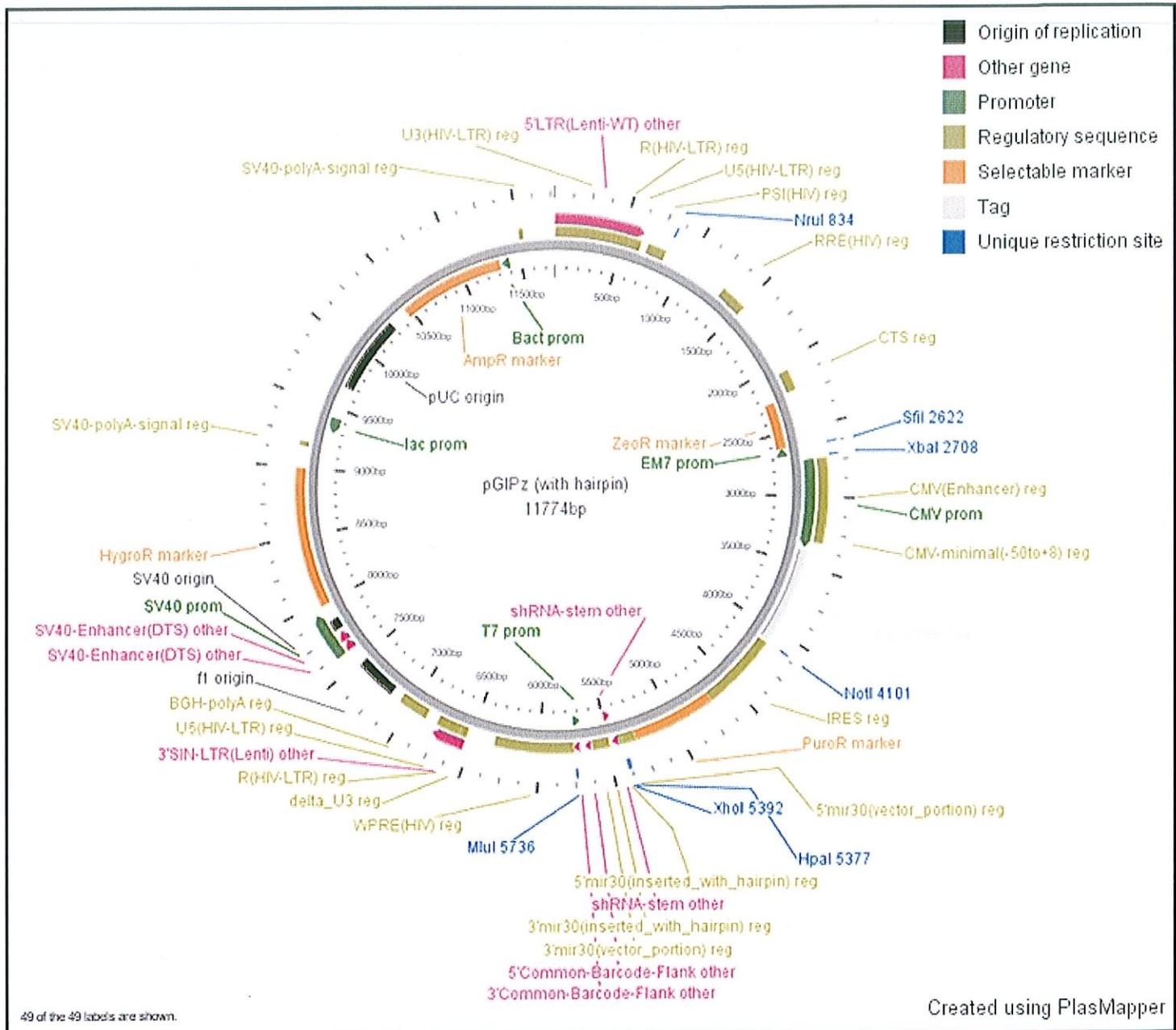
#### **pDNR-Dual Vector map, Multiple Cloning Site, and Sequence Information**

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 pLKO.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

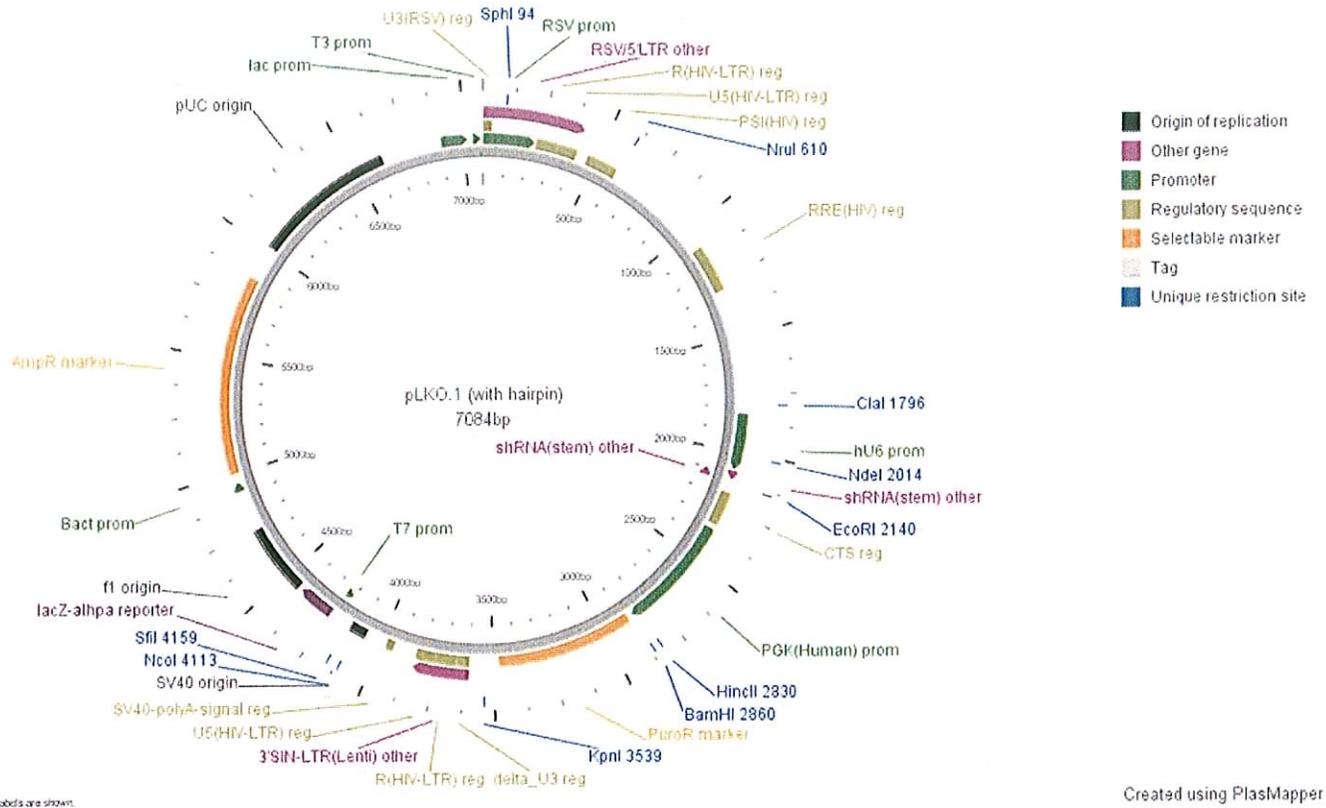
cPPT Central polypurine tract

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,  $1 \times 10^5$ - $1 \times 10^6$  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<sub>1</sub> on the cell surface. The A subunit catalyzes ADP-ribosylation of the  $\alpha$ -subunit of the stimulatory G protein ( $G_{\alpha_s}$ ), reducing GTPase activity and activating the  $\alpha$ -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  $\alpha$  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  $G_{\alpha}$  subunits remain in their GDP-bound, inactive state, they are unable to inactivate adenylyl cyclase or open  $K^+$  channels.



# Toxin Info

## TOXIN USE RISK ASSESSMENT

Name of Toxin:	Cholera toxin
Proposed Use Dose:	10 µg
Proposed Storage Dose:	500 µg
LD <sub>50</sub> (species):	250 µg

<b>Calculation:</b>		
250 µg/kg	x	50 kg/person
Dose per person based on LD <sub>50</sub> in µg = 12500		
LD <sub>50</sub> per person with safety factor of 10 based on LD <sub>50</sub> 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<sub>50</sub> 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<sub>50</sub> 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	Leurotoxins
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**

<b>Name of Toxin:</b>	Pertussis
<b>Proposed Use Dose:</b>	5 µg
<b>Proposed Storage Dose:</b>	50 µg
<b>LD<sub>50</sub> (species):</b>	15 µg

<b>Calculation:</b>				
	15 µg/kg	x	50 kg/person	
	Dose per person based on LD <sub>50</sub> in µg = 750			
	<b>LD<sub>50</sub> per person with safety factor of 10 based on LD<sub>50</sub> in µg =</b>			<b>75</b>

**Comments/Recommendations:**

### 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  
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

Manufacturer : Sigma-Aldrich Corporation  
3050 Spruce St.  
St. Louis, Missouri 63103  
USA

### 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
<b>Tris (hydroxymethyl) aminomethane</b>			
77-86-1	201-064-4	-	>= 5.82 - <= 5.94 %
<b>2-Amino-2-(hydroxymethyl)propane-1,3-diol hydrochloride</b>			
1185-53-1	214-684-5	-	>= 31.3 - <= 31.9 %
<b>Sodium chloride</b>			
7647-14-5	231-598-3	-	>= 57.6 - <= 58.8 %
<b>Exotoxin, vibrio cholerae</b>			
9012-63-9	-	-	>= 0.5 - <= 2.5 %
<b>Edetate disodium dihydrate</b>			
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**

<b>Inhalation</b>	May be harmful if inhaled. Causes respiratory tract irritation.
<b>Ingestion</b>	Harmful if swallowed.
<b>Skin</b>	Harmful if absorbed through skin. Causes skin irritation.
<b>Eyes</b>	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.

Exotoxin, vibrio cholerae

CAS-No.  
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**

Copyright 2010 Sigma-Aldrich Co. License granted to make unlimited paper copies for internal use only. 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

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Section 1 - Product and Company Information

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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

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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

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#### 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

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#### Section 10 - Stability and Reactivity

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##### 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

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#### Section 11 - Toxicological Information

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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, hemagglutination 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, hemagglutination 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.

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Section 12 Ecological Information

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No data available.

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Section 13 Disposal Considerations

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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.

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Section 14 Transport Information

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DOT DOT

Proper Shipping Name: AmToxins, extracted from diving sources solid, in, on, s.s.  
UN# UN3462462  
Class: 6.1  
Packing Group: Packing Group I  
Hazard Label: Toxic substances.  
PIHP: Not PIHP

IATA IATA

Proper Shipping Name: AmToxins, extracted from diving sources solid, in, on, s.s.  
IATA UN Number: 3462462  
Hazard Class: 6.1  
Packing Group: I

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Section 15 Regulatory Information

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EU ADDITIONAL CLASSIFICATION

Symbol of Danger: T  
Indication of Danger: Toxic.  
R: 23/24/25/25  
Risk Statements: Toxic by inhalation, in contact with skin and if swallowed.  
S: 45-36/37/39/39  
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 MSDSSDS contains all the information required by the CPR.  
DSL: No

Section 16 Other Information

DISCLAIMER

For Research 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 packaging slip for additional terms and conditions of sale. Copyright 2010 Sigma-Aldrich Co. License granted to make unlimited paper copies for internal use only.