

**THE UNIVERSITY OF WESTERN ONTARIO
 BIOHAZARDOUS AGENTS REGISTRY FORM**
 Approved Biohazards Subcommittee: September 25, 2009
 Biosafety Website: www.uwo.ca/humanresources/biosafety/

This form must be completed by each Principal Investigator holding a grant administered by the University of Western Ontario or in charge of a laboratory/facility where the use of Level 1, 2 or 3 biohazardous agents 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 biohazards 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 Biohazard 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 Qingping Feng
 SIGNATURE Qingping Feng
 DEPARTMENT Physiology and Pharmacology
 ADDRESS MSB Rm 254, UWO
 PHONE NUMBER x 82989
 EMERGENCY PHONE NUMBER(S) 519-933-9289
 EMAIL Qingping.Feng@schulich.uwo.ca

Location of experimental work to be carried out: Building(s) MSB Room(s) 253

*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 12.0, Approvals).

FUNDING AGENCY/AGENCIES: CIHR, HSFO
 GRANT TITLE(S): 1. Rac1 signaling in myocardial TNF- α expression in sepsis.
2. Heart development in diabetes: Role of NO
3. Cardioprotection by erythropoietin: Role of NO

PLEASE ATTACH A BRIEF DESCRIPTION OF YOUR WORK THAT EXPLAINS THE BIOHAZARDS USED AND HOW THEY WILL BE USED. PROJECTS SUBMITTED WITHOUT A SUMMARY WILL NOT BE REVIEWED. A GRANT SUMMARY PAGE MAYBE ADEQUATE IF IT PROVIDES SUFFICIENT DETAIL ABOUT EACH BIOHAZARD USED.

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

<u>Sharon Lu</u>	<u>Paul Arnold</u>
<u>Lily Xiang</u>	<u>Hoda Moazzen</u>
<u>Jun Liu</u>	<u>Ian Wu</u>
<u>Ting Zhang</u>	<u>Zhi-Xin Shan</u>
<u>Carmen Leung</u>	<u>Murong Liu</u>
	<u>Haixiang Hu</u>

1.0 Microorganisms

1.1 Does your work involve the use of biological agents? YES NO
 (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:

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

1.2 Please complete the table below:

Name of Biological agent(s)*	Is it known to be a human pathogen? YES/NO	Is it known to be an animal pathogen? YES/NO	Is it known to be a zoonotic agent? YES/NO	Maximum quantity to be cultured at one time? (in Litres)	Source/ Supplier	PHAC or CFIA Containment Level
Adenovirus	<input checked="" type="radio"/> Yes <input type="radio"/> No	<input type="radio"/> Yes <input checked="" type="radio"/> No	<input type="radio"/> Yes <input checked="" type="radio"/> No	0.1 L	Applied Biomedical Materials	<input type="radio"/> 1 <input checked="" type="radio"/> 2 <input type="radio"/> 3
Lentivirus	<input checked="" type="radio"/> Yes <input type="radio"/> No	<input type="radio"/> Yes <input checked="" type="radio"/> No	<input type="radio"/> Yes <input checked="" type="radio"/> No	0.1 L	Applied Biomedical Materials	<input type="radio"/> 1 <input checked="" type="radio"/> 2 <input type="radio"/> 3
(O55: B5) E. coli	<input checked="" type="radio"/> Yes <input type="radio"/> No	<input checked="" type="radio"/> Yes <input type="radio"/> No	<input checked="" type="radio"/> Yes <input type="radio"/> No	0.2 L	ATCC	<input type="radio"/> 1 <input checked="" 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"/> 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		Not applicable
Rodent	<input checked="" type="radio"/> Yes <input type="radio"/> No	Mice heart, skin	2007-011-03
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)*	Supplier / Source
Human	<input checked="" type="radio"/> Yes <input checked="" type="radio"/> No		
Rodent	<input checked="" type="radio"/> Yes <input 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		

*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 1 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/NO	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"/> No <input type="radio"/> Unknown		<input type="radio"/> 1 <input type="radio"/> 2 <input type="radio"/> 3
Human Blood (fraction) or other Body Fluid		<input type="radio"/> Yes <input type="radio"/> No <input type="radio"/> Unknown		<input type="radio"/> 1 <input type="radio"/> 2 <input type="radio"/> 3
Human Organs or Tissues (unpreserved)		<input type="radio"/> Yes <input type="radio"/> No <input type="radio"/> Unknown		<input type="radio"/> 1 <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

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

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

Virus Used for Vector Construction	Vector(s) *	Source of Vector	Gene(s) Transduced	Describe the change that results
Adenovirus	-	ABM	-Cre, nNOS	-gene knock down or upregulation
Lentivirus	-Lentivirus Expression System	ABM	-Oct 3/4, Trx5, Gata4, Bcl6, Mx2, Nanog	

* Please attach a Material Safety Data Sheet or equivalent. * Adenovirus previously registered

4.4 Will genetic sequences from the following be involved?

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

4.5 Will virus be replication defective? YES NO

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

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

5.0 Human Gene Therapy Trials

5.1 Will human clinical trials be conducted involving a biological agent? YES 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? YES 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? YES, number: _____ NO PENDING

6.0 Animal Experiments

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

6.2 Name of animal species to be used C57BL/6 mice

6.3 AUS protocol # 2007-011-03

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

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

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

13.0 Containment Levels

11.1 For the work described in sections 1.0 to 9.0, please indicate the highest HC or CFIA Containment Level required. 0 1 2 0 3

13.2 Has the facility been certified by OHS for this level of containment?
 YES, permit # if on-campus BIO - UWO - 0089
 NO, please certify
 NOT REQUIRED for Level 1 containment

14.0 Procedures to be Followed

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

SIGNATURE *Q. Jones* Date: March 29, 2010

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

14.3 Please outline what will be done if there is an exposure to the biohazards listed, such as a needlestick injury:

Contact UWO Health Services for medical evaluation.

15.0 Approvals

UWO Biohazard Subcommittee: SIGNATURE: _____
Date: _____

Safety Officer for Institution where experiments will take place: SIGNATURE: _____
Date: _____

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

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

Special Conditions of Approval:

Biohazard Summary:

Adenovirus

How it is stored:

- Adenovirus' are stored in DMEM media with 2.5% glycerol at -80°C.

How it is used:

- Amplification is performed using HEK 293 cells by infecting the cells with 10µl of the adenovirus in a 60mm dish. The collection of cells are frozen and thawed three times, and the supernatant is separated from cell debris for infection of target cells, which are neonatal cardiomyocytes.

How it is disposed:

- Any solution or containers that handled adenovirus are washed with bleach and autoclaved before disposal.

Lentivirus

How it is stored:

- Lentivirus' are stored in 10mM Tris-HCl, 1mM EDTA, pH8.0 buffer at -80°C.

How it is used:

- Cocktails containing a combination of 6 lentivirus' (Tbx5, Gata4, Baf60C, Nkx2.5, Oct3/4, Nanog) are transfected into isolated fibroblasts from one of four cell sources: mouse neonatal skin, mouse embryos E14.5, mouse neonatal heart, and mouse adult heart. The isolated fibroblasts are grown until confluent and transfected at passage 2 or 3. Cells are incubated with the lentivirus for 24 hours at 37°C and 5% CO₂, and each virus is used at a range of multiplicity of infections (MOIs) of 5-20. After 24 hrs the lentivirus is removed from the cells and inactivated with 10% bleach. Cells are then monitored for viability and characteristic changes of differentiation from fibroblasts into cardiomyocytes.
- Upon approval of the protocol from the UWO animal use subcommittee, we will inject transfected cells that demonstrate characteristic changes into live animals for heart failure therapy. Specifically, the protocol will follow the details used in our *in vitro* work to develop viable cells with characteristic changes. The cells will then be injected into the pericardium cavity of mice and their ability to differentiate in cardiomyocytes will be evaluated using immunohistostaining. The mice will be sacrificed and their hearts will be fixed in paraformaldehyde and imbedded in paraffin. The hearts will then be sectioned onto slides and stained for cardiomyocyte-specific markers like α-actinin or troponin-I.

How it is disposed:

- Any solution or containers that handled lentivirus are washed with bleach and autoclaved before disposal.

Escherichia coli

How it is stored:

- E. coli is stored in glycerol medium at -80°C.

How it is used:

- E. coli is amplified in nutrient agar or broth at 37°C for approximately 18hrs.
- E. coli is injected intravenously or intraperitoneally into mice at a dose of 2.5×10^7 CFU/g of body weight to induce gram-negative sepsis conditions. Mice are then monitored and analyzed as per experiment protocol.

How it is disposed:

- Any container that handled E. coli solution is washed with bleach and autoclaved before disposal.
- All animals treated with E. coli are incinerated after sacrifice or death.

----- Original Message -----

> **Subject:** Re: Biohazardous Agents Registry Form: Feng lab

> **Date:** Fri, 16 Apr 2010 16:23:18 -0400

> **From:** Paul Arnold <parnold4@uwo.ca>

> **To:** Jennifer Stanley <jstanle2@uwo.ca>

>

> Hi Jennifer,

> Dr. Feng has asked me to help address some of the questions you have regarding the Biohazardous Agents Form.

>

>

> - I have attached an updated version of the biohazard summary sheet which explains in more detail the in vitro and in vivo work for lentivirus'.

> - The source of the vectors is Applied Biomedical Materials (ABM). They are the same source mentioned in Table 1.2, but we should have been more clear in Table 4.3. Our apologies.

> - The HEK 293 cells are used for amplification of adenovirus. It is not obvious where in the form that information should be noted.

> - I assume the Level2+ checklist mandatory items which were marked as 'No' were Q.20 and Q.25 on the PHAC additional items checklist. Q.25 is a mistake and should be marked as 'Yes', dedicated laboratory clothing is certainly removed when leaving the contaminated laboratory area. And in regards to Q.20, the note underneath makes allowances for full coverage without the removal of street clothing when working with agents that are not infectious via inhalation. Full coverage is worn by lab individuals, so I assume the appropriate answer to this question should be 'N/A' and not 'No'.

>

>

> Hope that covers what you were looking for,

> Paul

----- Original Message -----

Subject:Re: containment level request - lentiviral project

Date:Fri, 23 Apr 2010 09:34:58 -0400

From:Permit-Permis <permitpermis@phac-aspc.gc.ca>

To:Jennifer Stanley <jstanle2@uwo.ca>

FENG

Dear Jennifer Stanley

Thank you for contacting our Directorate with your questions.

I regret that the Office of Laboratory Security has a policy of not undertaking risk assessments of the human pathogens in facility's inventories, as many of the factors that come into play are specific to a particular location and application. The determination of what risk group to which your pathogens belong is your responsibility.

As a first means of assisting you in determining your inventory's risk groups(s), you may want to consult the schedules for Risk Groups 2, 3, and 4 that are appended to The Human Pathogens and Toxins Act. Schedules 2 to 4 of the Act provide, respectively, non-exhaustive lists or examples of the kinds of human pathogens that are included in each of risk groups 2, 3, and 4. See link below:

<http://www2.parl.gc.ca/HousePublications/Publication.aspx?Docid=4015133&file=4>

If you possess human pathogens that are not included in these schedules, then you would have to determine the risk groups of those pathogens yourself. First, you could consult the definitions of the Risk Groups that are provided in section 3 of the Act. Further, there is greater detail on the criteria for determining the risk group of a pathogen in sections 2.1, 2.3 and 7.2 of the Laboratory Biosafety Guidelines . See attachment below:

For more information you can visit our website;

<http://www.phac-aspc.gc.ca/ols-bsl/pathogen/index-eng.php>

Regards

Josee Davies

Regulatory Technologist/ technologiste en réglementation

Pathogen Regulation Directorate (formerly Office of Laboratory Security) /

Direction de la réglementation des agents pathogènes (anciennement le

Bureau de la sécurité des laboratoires)

Public Health Agency of Canada/ Agence de santé publique du Canada

100 ch. Colonnade Rd. AL: 6201A Ottawa, Ontario, Canada K1A 0K9

Tel: (613) 957-1779

Fax: (613) 941-0596

Jennifer Stanley <jstanle2@uwo.ca> 2010-04-22 11:51 AM

To Permit-Permis <permitpermis@phac-aspc.gc.ca>

cc

Subject containment level request - lentiviral project

Hello there

We have a researcher who proposes to do a lentiviral project - please see details below, what containment do you suggest?

Regards

Jennifer



Containment Level 2+ 3 Operational – Additional Items Checklist

Q.4.
(Chapter 3.1.3)

Is a program in place (with appropriate authority to oversee safety and containment practices) for the management of biological safety issues?

MANDATORY	
<input checked="" type="checkbox"/>	Yes
<input type="checkbox"/>	No
<input type="checkbox"/>	N/A

Q.19.
(Chapter 3.1.3)

Is a containment check performed before entering the containment laboratory (e.g. verify correct readings on the pressure monitoring device)?

MANDATORY	
<input checked="" type="checkbox"/>	Yes
<input type="checkbox"/>	No
<input type="checkbox"/>	N/A

Q.20.
(Chapter 3.1.3)

Does personnel remove street clothing and change into dedicated laboratory clothing and shoes when entering the laboratory?

MANDATORY	
<input type="checkbox"/>	Yes
<input checked="" type="checkbox"/>	No
<input type="checkbox"/>	N/A

Note: the use of full coverage protective clothing is an acceptable alternative. Laboratories manipulating organisms, such as HIV, that are not infectious via inhalation are not required to remove street clothing.

Q.24.
(Chapter 3.1.3)

If an additional layer of protective clothing (e.g. solid-front gowns with tight-fitting wrists, gloves, respiratory protection) is worn over laboratory clothing when handling infectious materials, is it removed after completion of work (e.g. dedicated for use at the BSC)?

RECOMMENDED	
<input type="checkbox"/>	Yes
<input checked="" type="checkbox"/>	No
<input type="checkbox"/>	N/A

Q.25.
(Chapter 3.1.3)

Are dedicated laboratory clothing and shoes removed before leaving the laboratory in a manner that minimizes any contamination of the skin with the potentially contaminated dedicated laboratory clothing?

MANDATORY	
<input type="checkbox"/>	Yes
<input checked="" type="checkbox"/>	No
<input type="checkbox"/>	N/A

Q.34.
(Chapter 3.1.3)

Are infectious agents stored inside the containment laboratory?

RECOMMENDED	
<input checked="" type="checkbox"/>	Yes
<input type="checkbox"/>	No
<input type="checkbox"/>	N/A

Q.35.
(Chapter 3.1.3)

If agents are stored outside of the containment laboratory are they kept in leakproof containers in a restricted area?

MANDATORY	
<input type="checkbox"/>	Yes
<input type="checkbox"/>	No
<input checked="" type="checkbox"/>	N/A

Note: emergency response procedures must take into account the existence of infectious agents that are stored outside of the containment area.



Containment Level 2+ 3 Operational – Additional Items Checklist

Q.36. (Chapter 3.1.3)	Are all activities with infectious materials conducted in a BSC?	RECOMMENDED <input checked="" type="checkbox"/> Yes <input type="checkbox"/> No <input type="checkbox"/> N/A
Q.37. (Chapter 3.1.3)	Are other primary containment devices in combination with personal protective clothing and equipment used if it is not possible to conduct all activities with infectious materials in a BSC?	MANDATORY <input checked="" type="checkbox"/> Yes <input type="checkbox"/> No <input type="checkbox"/> N/A
Q.38. (Chapter 3.1.3)	Is the centrifugation of infectious materials carried out in closed containers placed in sealed safety cups or rotors that are unloaded in a BSC?	MANDATORY <input type="checkbox"/> Yes <input type="checkbox"/> No <input checked="" type="checkbox"/> N/A
Q.129. (Chapter 3.1.3)	In the event of an emergency, is an exit protocol in place whereby routine procedures might be bypassed? Note: in the event of life-threatening emergencies, personal health and safety are always a priority.	MANDATORY <input checked="" type="checkbox"/> Yes <input type="checkbox"/> No <input type="checkbox"/> N/A
Q.130. (Chapter 3.1.3)	Has a reporting area been identified where further steps might be taken (e.g. disinfecting footwear, changing, showering)?	MANDATORY <input checked="" type="checkbox"/> Yes <input type="checkbox"/> No <input type="checkbox"/> N/A
Q.138. (Chapter 3.1.3)	Are the general operational protocols supplemented with protocols specific to each project in progress?	MANDATORY <input checked="" type="checkbox"/> Yes <input type="checkbox"/> No <input type="checkbox"/> N/A
Q.140. (Chapter 3.1.3)	Have personnel demonstrated proficiency in microbiological practices and techniques?	MANDATORY <input checked="" type="checkbox"/> Yes <input type="checkbox"/> No <input type="checkbox"/> N/A



Workplace Health

Policy and Procedure for persons working with Lentivirus and Lentivirus-based vectors

The potential for human inoculation and uncertainties concerning the potential infectivity of agents transduced by the Lentivirus system dictate the need for a simple medical surveillance program.

All persons working with Lentivirus and Lentivirus-based vectors will be seen at Workplace Health prior to the commencement of work with the virus.

A reference serum specimen will be collected and banked.

No annual follow up will be required.

In the event of a needle stick injury in which self-inoculation with Lentivirus or Lentivirus-based vectors is a possibility, the individual will report to Workplace Health and a serum sample for HIV and / or other pathogens will be obtained and tested. All serum specimens will be coded and the test result will be known only to the Workplace Health physician and the donor individual.

Approved Biohazards Committee December 6, 2007

Applied Biological Materials Inc.
MATERIAL SAFETY DATA SHEET - BIOLOGICAL SUBSTANCES

Date Updated: 09/03/2008
Version 1.0

SECTION I - BIOLOGICAL AGENT

NAME: Lentivirus Expression System

SOURCE NAME: *Human Immunodeficiency Virus*

SYNONYM OR CROSS REFERENCE: HIV, AIDS, Acquired Immune Deficiency Syndrome, HTLV III LAV

GENERAL CHARACTERISTICS: Retroviridae (Lentivirus); ss RNA, enveloped icosahedral nucleocapsid, glycoprotein envelope, reverse transcriptase

RECOMBINANT CHARACTERISTICS: 3rd-generation of self-inactivating recombinant lentiviral vectors with enhanced biosafety and minimal relation to the wild-type, human HIV-1 virus. The lentiviral particles produced with this system are replication-incompetent and designed with a number of safety features to enhance its biosafety.

An enhancer deletion in the U3 region of 3' ΔLTR ensures self-inactivation of the lentiviral vector following transduction and integration into genomic DNA of the target cells. Utilization of an RSV promoter upstream of 5' LTR allows efficient Tat-independent production of viral RNA.

The number of lentiviral genes necessary for packaging, replication and transduction is limited to three (*gag*, *pol*, *rev*), and their expression is derived from different plasmids, which all lack packaging signals. These plasmids share no significant homology to the expression vector, preventing the generation of replication-competent virus. None of the *gag*, *pol*, or *rev* genes will be present in the packaged viral genome, thus making the mature virus replication-incompetent.

SECTION II - HEALTH HAZARD

PATHOGENICITY: Insidious onset with non-specific symptoms such as lymphadenopathy, anorexia, chronic diarrhea, weight loss, fever, and fatigue; opportunistic infections and malignant diseases without a known cause for immune deficiency

EPIDEMIOLOGY: First reported in 1981; cases recorded in Americas, Europe, Africa and many other areas; patient categories - homosexually or bisexually active men, drug abusers, Haitian/African emigrants, hemophiliacs, sexual partners of men and women in these categories, infants born to parents in this category

HOST RANGE: Humans

INFECTIOUS DOSE: Unknown

MODE OF TRANSMISSION: Transmitted from person to person through direct exposure to infected body fluids (blood, semen) sexual contact, sharing unclean needles etc.; transplacental transfer can occur

INCUBATION PERIOD: Epidemiologic evidence suggests that duration from exposure to onset of symptoms has a minimum range from 6 months to more than 7 years

COMMUNICABILITY: Period of communicability extends from asymptomatic period through appearance of opportunistic diseases

RESERVOIR: Humans

ZOONOSIS: None

VECTORS: None

SECTION IV - VIABILITY

DRUG SUSCEPTIBILITY: Several reverse transcriptase and protease inhibitors now licensed

SUSCEPTIBILITY TO DISINFECTANTS: Susceptible to many disinfectants - 1% sodium hypochlorite, 2% glutaraldehyde, formaldehyde, ethanol

PHYSICAL INACTIVATION: Effectiveness of 56°C - 60°C heat in destroying HIV in serum not certain, however, heating small volumes of serum for 30 min at 56°C before serologic testing reduces residual infectivity to below detectable levels

SURVIVAL OUTSIDE HOST: Drying in environment causes rapid (within several hours) 90-99% reduction in HIV concentration

SECTION V - MEDICAL

SURVEILLANCE: Serological monitoring for evidence of HIV infection

FIRST AID/TREATMENT: Specific measures for the opportunistic diseases that result from AIDS; "Cocktail" multidrug treatment for HIV

IMMUNIZATION: None available

PROPHYLAXIS: Experimental prophylaxis with AZT/DDI or other appropriate drug

SECTION VI - LABORATORY HAZARDS

LABORATORY-ACQUIRED INFECTIONS: 5 reported laboratory-acquired infections with HIV (splashing of infected materials, inapparent skin exposure, puncture wounds); 18 reported cases in health care workers worldwide

SOURCES/SPECIMENS: Blood, semen, vaginal secretions, CSF, other specimens containing visible blood, unscreened or inadequately treated blood products

PRIMARY HAZARDS: Direct contact with skin and mucous membranes of the eye, nose and mouth; accidental parenteral inoculation; ingestion; hazard of aerosols exposure unknown

SPECIAL HAZARDS: Extreme care must be taken to avoid spilling and splashing infected materials - virus should be presumed in/on all equipment and devices coming in direct contact with infected materials

SECTION VII - RECOMMENDED PRECAUTIONS

CONTAINMENT REQUIREMENTS: Biosafety level 2 practices, containment equipment and facilities for activities involving clinical specimens and non-cultured procedures (primary containment devices may be indicated eg. biological safety cabinets) and for activities involving non-human primates and any animals experimentally infected or inoculated with HIV; Biosafety level 3 practices, containment equipment and facilities for all work culturing HIV

PROTECTIVE CLOTHING: Gloves should be worn when handling potentially infectious specimens, cultures or tissues; laboratory coats, gowns or suitable protective clothing should be worn

OTHER PRECAUTIONS: Keep hands away from the eyes, nose and mouth in order to avoid potential exposure of the mucous membranes; eye goggles or face shields may assist in accomplishing this objective

SECTION VIII -- HANDLING INFORMATION

SPILLS: Allow aerosols to settle; wearing protective clothing, gently cover spill with paper towels and apply 1% sodium hypochlorite, starting at perimeter and working towards the centre; allow sufficient contact time (30 min) before clean up

DISPOSAL: Decontaminate before disposal - steam sterilization, incineration, chemical disinfection

STORAGE: In sealed containers that are appropriately labeled

SECTION IX -- MISCELLANEOUS INFORMATION

The information contained in this Material Safety Datasheet is believed to be accurate but it is the responsibility of the user or supplier to determine the applicability of these data to the formulation of necessary safety precautions.

Applied Biological Materials Inc. shall not be held responsible for any damage resulting from the use of the above product or the information contained in this Material Safety Datasheet.

Viral Vector Information:

pLenti-III-m-Gata4

Accession Number	NM_008092
Vector	pLenti-III-HA
Backbone Size	8183bp

Description	This gene encodes a member of the GATA family of zinc-finger transcription factors. Members of this family recognize the GATA motif which is present in the promoters of many genes. This protein is thought to regulate genes involved in embryogenesis and in myocardial differentiation and function. Mutations in this gene have been associated with cardiac septal defects.
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Species of Genes	Mouse
Host Strain	DH5alpha
Cloning Sites	EcoRI
Insert Size	1.9kb
Promoter	CMV
Tags	HA
Selection Marker	Kanamycin
Sequence Primer	CGCAAATGGGCGGATGGCGTG
Format	Plasmid
Appearance	Clear Liquid
Storage Buffer	10mM Tris-HCl, 1mM EDTA, pH8.0
StorageConditions	1 year when stored at -20°C or lower in a non-frost free freezer

pLenti-III-m-Nanog

Accession Number	NM_008092
Vector	pLenti-III-HA
Backbone Size	8183bp

Description	This gene encodes a member of the GATA family of zinc-finger transcription factors. Members of this family recognize the GATA motif which is present in the promoters of many genes. This protein is thought to regulate genes involved in embryogenesis and in myocardial differentiation and function. Mutations in this gene have been associated with cardiac septal defects.
Species of Genes	Mouse
Host Strain	DH5alpha
Cloning Sites	EcoRI
Insert Size	1.9kb
Promoter	CMV
Tags	HA
Selection Marker	Kanamycin
Sequence Primer	CGCAAATGGGCGGATGGCGTG
Format	Plasmid
Appearance	Clear Liquid
Storage Buffer	10mM Tris-HCl, 1mM EDTA, pH8.0
StorageConditions	1 year when stored at -20°C or lower in a non-frost free freezer

pLenti-III-m-NKX2.5

Accession Number	BC139303
Vector Backbone Name	Please inquire
Vector Backbone Size (bp)	8119bp
Species of Gene(s)	Mouse
Host Strain	DH5a
Cloning Sites	BamHI/XhoI
Insert Size (bp)	2kb
Promoter	CMV
Fusion Proteins or Tags	HA

Selection Marker	Kan
Sequencing Primer	CGCAAATGGGCGGATGGCGTG
Format	Plasmid
Appearance	Clear Liquid
Storage Buffer	10mM Tris-HCl, 1mM EDTA, pH8.0
Storage Conditions	1 year when stored at -20°C or lower in a non-frost free freezer

pLenti-III-m-Oct 3/4

Accession Number	NM_013633
Vector Backbone Name	Please inquire
Vector Backbone Size (bp)	8183bp
Species of Gene(s)	Mouse
Host Strain	DH5a
Cloning Sites	EcoRI
Insert Size (bp)	1324bp
Promoter	CMV
Fusion Proteins or Tags	HA
Selection Marker	Kan
Sequencing Primer	CGCAAATGGGCGGATGGCGTG
Format	Plasmid
Appearance	Clear Liquid
Storage Buffer	10mM Tris-HCl, 1mM EDTA, pH8.0
Storage Conditions	1 year when stored at -20°C or lower in a non-frost free freezer

pLenti-III-m-Tbx5

Accession Number	BC090639
Vector Backbone Name	Please inquire

Vector Backbone Size (bp)	8183bp
Species of Gene(s)	Mouse
Host Strain	DH5a
Cloning Sites	NotI/SalI
Insert Size (bp)	3.2kb
Promoter	CMV
Fusion Proteins or Tags	HA
Selection Marker	Kan
Sequencing Primer	CGCAAATGGGCGGATGGCGTG
Format	Plasmid
Appearance	Clear Liquid
Storage Buffer	10mM Tris-HCl, 1mM EDTA, pH8.0
Storage Conditions	1 year when stored at -20°C or lower in a non-frost free freezer

pLenti-III-m-Baf60c

Accession Number	BC060525
Vector Backbone Name	Please inquire
Vector Backbone Size (bp)	8183bp
Species of Gene(s)	Mouse
Host Strain	DH5a
Cloning Sites	SalI/NotI
Insert Size (bp)	1.6kb
Promoter	CMV
Fusion Proteins or Tags	HA
Selection Marker	Kan
Sequencing Primer	CGCAAATGGGCGGATGGCGTG
Format	Plasmid

Appearance	Clear Liquid
Storage Buffer	10mM Tris-HCl, 1mM EDTA, pH8.0
Storage Conditions	1 year when stored at -20°C or lower in a non-frost free freezer