Modification Form for Permit BIO-RRI-0057

Permit Holder: Vincent Morris

Approved Personnel
(Please stroke out any personnel to be removed)

Maryam Shirmohammad

Sarah Detombe

Ian MacDonald

Nicole Hague

Additional Personnel
(Please list additional personnel here)

	Please stroke out any approved Biohazards to be removed below	Write additional Biohazards for approval below. Give the full name - do not abbreviate.
Approved Microorganisms	E.coli - dh5 Alpha, Influenza C	Influenza B Virus
Approved Primary and Established Cells	rodent (C57B/6), rodent (mmpq), rodent (C57B1/6), mouse melanoma, B16F1, B16F10, MDAMB231, Cloudman S91 mouse melanoma cells, HMVII Established human melanoma cell line (vaginal)	
Approved Use of Human Source Material		
Approved Genetic Modifications (Plasmids/Vectors)	pcDNA 3.1 Hygro, pcDNA 3.1 neo	
Approved Use of Animals	mice	
Approved Biological Toxin(s)		

* *PLEASE ATTACH A MATERIAL SAFETY DATA SHEET OR EQUIVALENT FOR NEW BIOHAZARDS.*
** *PLEASE ATTACH A BRIEF DESCRIPTION OF THE WORK THAT EXPLAINS THE BIOHAZARDS USED AND HOW THEY WILL BE STORED, USED AND DISPOSED OF..*

As the principal investigator, I have ensured that all of the personnel named on the form have been trained. 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 of Permit Holder: Vincent LaMorie

Current Classification: 2 **Containment Level for Added Biohazards:**

Date of Last Biohazardous Agents Registry Form: Jan 5, 2010

Date of Last Modification (if applicable): Mar 2, 2010

BioSafety Officer(s):

Chair, Biohazards Subcommittee: **Date:**

Animal Viruses and Antisera

ATCC® Number:	**VR-1735™** Order this Item	Price:	**$240.00**
Classification:	Orthomyxoviridae, Influenzavirus B		
Agent:	Influenza B virus		
Strain:	B/Taiwan/2/62 (TC-adapted)		
Original Source:	Patient in Taiwan, 1962.		
Depositors:	A.P. Kendal and ATCC		
Biosafety Level:	2		
Shipped:	frozen		
Permits/Forms:	In addition to the MTA mentioned above, other ATCC and/or regulatory permits may be required for the transfer of this ATCC material. Anyone purchasing ATCC material is ultimately responsible for obtaining the permits. Please click here for information regarding the specific requirements for shipment to your location.		
Host Organism :	**Production Host:** MDCK (ATCCCCL-34)		
Incubation :	For best results, plate cells 24-48 hours in advance. Infect at 80%-90% confluent (not 100% confluent) using a multiplicity of infection (MOI) of 0.01 to 1.0. Incubate at 33.0°C in a humidified, 5% CO2 atmosphere for 2-4 days, until CPE are well advanced in 100% of the culture. In TC, ATCCVR-1735 should be grown in serum free media that contains 1.0 µg/mL TPCK-treated trypsin, 0.125% BSA and 1% HEPES (1 molar stock).		
Effect :	CPE (cell rounding, degeneration, sloughing)		
Store at :	-70.0°C or colder		
Comments :	Derived by adaptation of egg-passage B/Taiwan/2/62 (ATCCVR-295) to MDCK (ATCCCCL-34) cells at ATCC. (Note: ATCCVR-295 and ATCCVR-1735 have not been compared with respect to sequence or infectivity in chick embryo and tissue culture).		
Related Products:	Virus: ATCC VR-295 Host cells: ATCC CCL-34		

Related Links ▸

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BioStandards

Biological Reference Material and Consensus Standards for the life science community

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Animal Viruses and Antisera

ATCC® Number:	**VR-1535™** Order this Item	Price:	**$240.00**

Classification:	Orthomyxoviridae, Influenzavirus B
Agent:	Influenza B virus
Strain:	B/Lee/40
Original Source:	derived from existing strain (derived by tissue culture-adaptation of egg passage influenza B B/lee/40 (VR-101) to MDCK cells (ATCCCCL-34))
Depositors:	MT Coleman, ATCC
Biosafety Level:	2
Shipped:	frozen
Permits/Forms:	In addition to the MTA mentioned above, other ATCC and/or regulatory permits may be required for the transfer of this ATCC material. Anyone purchasing ATCC material is ultimately responsible for obtaining the permits. Please click here for information regarding the specific requirements for shipment to your location.
Host Organism :	*Homo sapiens* *Mus musculus*
Incubation :	**Atmosphere:** 5% CO2 in air recommended **Temperature:** 37.0°C **Duration:**1-3 days **Protocol:** In tissue culture VR-1535 should be grown in serum free media that contains 1 mg/ml TPCK-treated trypsin, 0.125% BSA and 1% HEPES (1M stock).
Effect :	Yes, in vitro effects: cytopathic effects (rounding and sloughing)
Store at :	-70°C (or colder)
Related Products:	source culture:ATCC VR-101
References :	32426: Francis T Jr.. A new type of virus from epidemic influenza. Science 92: 405-408, 1940. 32429: Andrewes CH, et al. A short description of the Myxovirus group (Influenza and related viruses). Virology 1: 176-184, 1955. PubMed: 13267985

Related Links ▸

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Cell, microbial and molecular genomics products for the life sciences

BioServices

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

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Biological Reference Material and Consensus Standards for the life science community

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Western

Schulich
MEDICINE & DENTISTRY

ABSTRACT OF RESEARCH INVOLVING INFLUENZA B VIRUS

Background: Skin melanoma is the 6th most common cancer among white men and women in the United States, and its incidence has been increasing for the last 3 decades. Melanoma patients have increased serum levels of sialic acid proportional to their tumor burden. Also human melanomas have an increased in O-acetylation of their scalic acids compared with normal skin. Influenza B attached to O-acetyl N-acetylneuramic acid (sialic acid). Thus influenza B virus can infect and replicate in melanoma cells in vitro. The purpose of our research is to determine if influenza B virus can infect and destroy melanoma tumors and metastases in mice in vivo.

Hypothesis: Influenza B virus can infect and destroy melanoma cell primary tumors and metastases in vivo.

Objective: Use Influenza B virus to destroy melanoma cells in vivo.

Experimental Approach:

1. Inject melanoma cells intradermally into a syngeneic mouse.

2. After the tumor appears, inject influenza B virus into the tumor.

3. Compare the growth and metastasis of the tumor in the influenza B virus injected mouse with the uninfected but tumor injected mouse.

Expected Results: We expect that the virus will infect and kill the melanoma cells in the infected mouse while the melanoma tumor will continue to grow and metastasize in the mouse not injected with influenza B. Since the virus is being injected intradermally into the tumor, we expect to find a dose that will kill the melanoma tumor cells but not cause illness in the mouse, since influenza B is transmitted through the respiratory tract.

Significance: These experiments will demonstrate the principal that viruses can be used to kill cancer cell in tumors and metastases in vivo.

Vincent L Morris

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Dental Sciences Building • London, Ontario • N6A 5C1 • Canada
PH: 519-661-3427 • F: 519-661-3499 • www.uwo.ca/mni/

Modification Form for Permit BIO-RRI-0057

Permit Holder: Vincent Morris

Approved Personnel
(Please stroke out any personnel to be removed)

Nicole Hague

Ian MacDonald

Additional Personnel
(Please list additional personnel here)

	Please stroke out any approved Biohazards to be removed below	Write additional Biohazards for approval below. *
Approved Microorganisms	E.coli - dh5 Alpha, Influenza C	
Approved Cells	rodent (C57B/6), rodent (mmpq), rodent (C57B1/6), mouse melanoma, B16F1, B16F10, MDAMB231, Cloudman S91 mouse melanoma cells	HMVII established human melanoma cell line, uninfected (vaginal)
Approved Use of Human Source Material		
Approved GMO	pcDNA 3.1 Hygro, pcDNA 3.1 neo	
Approved use of Animals	mice	
Approved Toxin(s)		

* *PLEASE ATTACH A MATERIAL SAFETY DATA SHEET OR EQUIVALENT FOR NEW BIOHAZARDS.*
** *PLEASE ATTACH A BRIEF DESCRIPTION OF THE WORK THAT EXPLAINS THE BIOHAZARDS USED AND HOW THEY WILL BE USED.*

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Signature of Permit Holder: Vincent D'Mari

Classification: 2

Date of Last Biohazardous Agents Registry Form: Jan 5, 2010

Date of Last Modification (if applicable):

BioSafety Officer(s): Stanley Feb 26/10

Chair, Biohazards Subcommittee: Gr Kidder

Health Protection Agency

Protecting people
Preventing harm
Preparing for threats

You are not logged on.

General Cell Collection Detail

General Cell Collection: HMVII

Catalogue No.:	92042701
Cell Line Name:	HMVII
Keywords:	Human vaginal maligant melanoma
Cell Line Description:	Established from a vaginal malignant melanoma of a 65 year-old female with blood type B. Cells produce melanin and can support the replication of influenza C virus.
Species:	Human
Tissue:	Vagina
Morphology:	Epithelial
Growth Mode:	Adherent
Passage No:	145
Subculture Routine:	Split sub-confluent cultures (70-80%) 1:3 to 1:6 i.e. seeding at 2-4x10,000 cells/cm² using 0.25% trypsin or trypsin/EDTA; 5% CO2; 37°C.
Culture Medium:	Ham's F10 or RPMI 1640 + 2mM Glutamine + 10-20% Foetal Bovine Serum (FBS).
Karyotype:	Not specified
Products:	Melanin
Depositor:	Prof T Kasuga, Tokyo Medical and Dental University, Bunkyo-ku
Originator:	Yes
Country:	JAPAN
References:	J Gen Virol 1989;70:1653
Additional Bibliography:	Not specified
Patents:	None specified by Depositor
Research Council Deposit:	No
Release Conditions:	No

The HPA Culture Collections represent deposits of cultures from world-wide sources. While every effort is made to ensure details distributed by HPA Culture Collections are accurate, HPA Culture Collections cannot be held responsible for any inaccuracies in the data supplied. References where quoted are mainly attributed to the establishment of the cell culture and not for any specific property of the cell line, therefore further references should be obtained regarding cell culture characteristics. Passage numbers where given act only as a guide and HPA Culture Collections does not guarantee the passage number stated will be the passage number received by the customer.

Cultures supplied by HPA Culture Collections are for research purposes only. Enquiries regarding the commercial use of a cell line are referred to the depositor of the cell line. Some cell lines have additional special release conditions such as the requirement for a material transfer agreement to be completed by the potential recipient prior to the supply of the cell line. Please view the Terms & Conditions of Supply for more information.

Delivery State:

○ **Frozen**	£375.00
○ **Growing**	£525.00
○ **DNA-5µg (100ng/µl)**	£50.00

Quantity Required:

Add to Shopping Cart

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ABSTRACT OF RESEARCH INVOLVING INFLUENZA C VIRUS

Background: Skin melanoma is the 6th most common cancer among white men and women in the United States, and its incidence has been increasing for the last 3 decades. Melanoma patients have increased serum levels of sialic acid proportional to their tumor burden. Also human melanomas have an increased in O-acetylation of their scalic acids compared with normal skin. Influenza C attached to O-acetyl N-acetylneuramic acid (sialic acid). Thus influenza C virus can infect and replicate in melanoma cells in vitro. The purpose of our research is to determine if influenza C virus can infect and destroy melanoma tumors and metastases in mice in vivo.

Hypothesis: Influenza C virus can infect and destroy melanoma cell primary tumors and metastases in vivo.

Objective: Use Influenza C virus to destroy melanoma cells in vivo.

Experimental Approach:

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Significance: These experiments will demonstrate the principal that viruses can be used to kill cancer cell in tumors and metastases in vivo.

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PH: 519-661-3427 • F: 519-661-3499 • www.uwo.ca/mni/

RRI-0057

Modification Form for Permit BIO-UWO-0174

Permit Holder: Vincent Morris

Approved Personnel
(Please stroke out any personnel to be removed)

Nicole Hague

Ian MacDonald

Additional Personnel
(Please list additional personnel here)

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Approved Use of Human Source Material		
Approved GMO	pcDNA 3.1 Hygro, pcDNA 3.1 neo	
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Signature of Permit Holder:

* **Classification:** 1 2 2l

Date of Last Biohazardous Agents Registry Form: Aug 24, 2007

Date of Last Modification (if applicable): Dec 21, 2009

BioSafety Officer(s): J Stanley January 5, 2010 RD

Chair, Biohazards Subcommittee: S McEldowney

Proposed locations

2220 RD

* In vitro work: Dr Madrena's Level 2 tissue culture (Robarts)

* In vivo work: Level 2 animal room (ACUS)

beiresources
BIODEFENSE & EMERGING INFECTIONS RESEARCH RESOURCES REPOSITORY

MATERIAL SAFETY DATA SHEET

Section 1 – Product and Company Information

Product Name: Influenza C Virus, C/Taylor/1233/47

Catalog Number: NR-3183

Company:
American Type Culture Collection
10801 University Boulevard
Manassas, Virginia, 20110-2204, USA

Telephone: 800-638-6597 or 703-365-2700
Fax: 703-365-2745
24 hour Emergency Number: 703-365-2710 or 800-424-9300 (Chemtrec - transport only)

Product Use: This material is authorized for research, non-commercial purposes only. It is not intended for use on human subjects.

Section 2 – Composition/Information on Ingredient

Substance Name: Influenza C Virus, C/Taylor/1233/47 in allantoic fluid.
CAS #: Not applicable
Alternate CAS #: Not applicable
UN #: 3373
RTECS #: Not available

Synonym or Cross Reference: None known

Section 3 – Hazards Identification

Health Hazards: Influenza C Virus, C/Taylor/1233/47 contains no hazardous chemicals as defined by the Occupational Safety and Health Standards, 29 CFR Part 1910.1200. The OSHA Hazcom Standard is available online at http://www.osha.gov/pls/oshaweb/owadisp.show_document?p_table=STANDARDS&p_id=10099.

Biosafety: Handle as potentially biohazardous material under at least Biosafety Level 2 containment.

Section 4 – First Aid Measures

In case of any exposure, seek medical attention.

Eye Exposure: With clean hands remove any contact lenses. Immediately flush with copious amounts of water, including under the eyelids, for at least 15 minutes. Then report to your safety officer and seek medical attention.

Dermal Exposure: Remove contaminated clothes. Rinse skin with plenty of water and soap if available. Then report to your safety officer and seek medical attention.

Inhalation Exposure: If victim is having trouble breathing or stops breathing seek emergency medical help. Do not perform CPR unless it can be done safely and you are a certified individual.

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MEDICINE & DENTISTRY

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Oral Exposure: If a person is unconscious never give them anything to drink and seek emergency medical help. Do not induce vomiting unless directed by medical personnel. If victim begins to vomit, turn on side or stomach to prevent backflow into respiratory tract.

Section 5 – Fire Fighting Measures

Flammability: Not Flammable

Suitable Extinguishing Media: Water spray, carbon dioxide, dry chemical powder, Halon (where regulations permit), or appropriate foam.

Firefighting

Protective Equipment: Wear self-contained breathing apparatus and protective clothing to prevent inhalation, ingestion, skin and eye contact.

Specific Hazard(s): Material is not flammable. Responders should take into consideration the biohazard risk associated with responding to a fire in the area where the material may be stored or handled.

Section 6 – Accidental Release Measures

Procedure(s) of Personal Precaution(s): At a minimum use PPE listed in Section 8. Wear laboratory coat, gloves and eye protection. Avoid all contact.

Methods for Cleaning Up

Patient/Victim: Wash with soap and water. Work clothes should be laundered separately. Launder contaminated clothing before re-use. Do not take clothing home.

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

Note: The use of additional PPE may be necessary for cleaning solutions.

Section 7 – Handling and Storage

Handling/User Exposure: Handle in accordance with at least Biosafety Level 2 practices. Keep closed or covered when not in use. Wear appropriate PPE. Avoid direct contact. Avoid tasks that may create aerosols.

Suitable Storage: The product should be stored at -60°C or colder immediately upon arrival.

Special Requirements: Follow established laboratory procedures when handling material.

Section 8 – Exposure Controls/PPE

AVOID DIRECT CONTACT

Work should be conducted under at least Biosafety Level 2. Appropriate safety procedures should always be used with this material. Laboratory safety is discussed in the following publication: U.S. Department of Health and Human Services, Public Health Service, Centers for Disease Control and Prevention, and National Institutes of Health. Biosafety in Microbiological and Biomedical Laboratories. 5th ed. Washington, DC: U.S. Government Printing Office, 2007. This text is available online at http://www.cdc.gov/od/ohs/biosfty/bmbl5/bmbl5toc.htm.

Engineering Controls: If possible, use in a containment device such as Class I or II biological safety cabinet with easy access to a safety shower and eyewash. Avoid handling the material in a manner likely to create aerosols. Dispose all sharps in an approved sharps container.

Respiratory Protection: Consult your organization's Respiratory Protection Program and OSHA 1910.134 for respiratory protection guidance.

Personal Protective Equipment: Laboratory coat, gloves and safety glasses with side-shields should be worn.

General Hygiene Measures: Avoid direct contact with skin, eyes, nose or mouth. Do not swallow or inhale. Wash hands at completion of work.

Section 9 – Physical/Chemical Properties

Appearance/Physical State:	Liquid (shipped as frozen solid)
Molecular Weight:	Not available
Boiling Point/Range:	Not available
Melting Point/Range:	Not available
Specific Gravity:	Not available
Density:	Not available
Solubility:	Not available

Section 10 – Stability and Reactivity

Stability: Chemically stable when dried and under ambient conditions. Refer to Section 7 for storage and handling information.
Conditions to Avoid: Not available
Materials to Avoid: Not available

Section 11 – Toxicological Information

Route of Exposure
Eye Contact: Possible route of infection. Avoid eye contact.
Skin Contact: Data not available. Avoid skin contact.
Skin Absorption: Data not available. Avoid skin absorption.
Inhalation: Inhalation of aerosolized virus is a likely route of infection. Fomites may play a role in transmission. Avoid inhalation.
Ingestion: The fecal oral route is a route of infection in birds and a possibility in humans. Fomites may play a role in transmission. Avoid ingestion.
Parenteral Exposure: Data not available. Avoid parenteral exposure.

Sensitization
Skin: Not known to occur.
Respiratory: Not known to occur.

Target Organ(s) or System(s)
See below

Signs and Symptoms of Exposure

Influenza C Virus does infect humans. However, it is a milder infection than Influenza A and B Viruses. Either asymptomatic infections or mild upper respiratory infections with symptoms similar to the common cold. Lower respiratory tract complications are rare. Influenza C Virus has never been associated with a large human epidemic.

Toxicity Data: Not available

Effects of Long Term or Repeated Exposure: Data not available

Chronic Exposure–Teratogen: Data not available

Chronic Exposure–Mutagen: Data not available

Chronic Exposure–Reproductive Hazard: Data not available

Section 12 – Ecological Information

Ecotoxicological Information: No ecological information available

Section 13 – Disposal Considerations

Decontaminate all wastes before disposal (steam sterilization, chemical disinfection, and/or incineration).

Dispose of in accordance with applicable regulations.

Section 14 – Transport Information

Contact BEI Resources for transport information.

Section 15 – Regulatory Information

Influenza C Virus, C/Taylor/1233/47 is not a Select Agent and hence is not required to be registered with the CDC.

Section 16 – Other Information

Disclaimer of expressed and implied warranties
The above information was compiled from sources believed to be reliable and 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. ATCC®, BEI Resources and the U.S. Government shall not be held liable for any damage resulting from handling or from contact with the above product. See the appropriate Product Information Sheet, Certificate of Analysis and invoice for additional terms and conditions of sale.

Biodefense and Emerging Infections Research Resources Repository
P.O. Box 4137
Manassas, VA 20108-4137 USA
www.beiresources.org

800-359-7370
Fax: 703-365-2898
E-mail: contact@beiresources.org

Abbreviations

ATCC – American Type Culture Collection
BEI Resources – Biodefense and Emerging Infections Research Resources Repository
CAS # – Chemical Abstract System Registry Number
CDC – Center for Disease Control
CFR – Code of Federal Regulations
CPR – CardioPulmonary Resuscitation
EPA – Environmental Protection Agency
HEPA – High Efficiency Particulate Air (Filter)
HHS – Health and Human Services
OSHA – Occupational Safety & Health Administration
PPE – Personal Protective Equipment
RTECS # – Registry of Toxic Effects of Chemical Substances Number
UN # – United Nations Committee of Experts on the Transport of Dangerous Goods Number
USDA – United States Department of Agriculture

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800-359-7370
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E-mail: contact@beiresources.org

MORRIS

Subject: Fwd: Re: Influenza C project - Madrenas & Morris
From: Jennifer Stanley <jstanle2@uwo.ca>
Date: Tue, 22 Dec 2009 11:42:38 -0500
To: rsn@uwo.ca

-------- Original Message --------
Subject: Re: Influenza C project - Madrenas & Morris
Date: Tue, 22 Dec 2009 11:37:23 -0500
From: Vincent L Morris <vmorris@uwo.ca>
To: Jennifer Stanley <jstanle2@uwo.ca>

To Jennifer:

Tissue culture will be done in room 2220 RRI.

Thanks again for all your help.

Vince

----- Original Message -----
From: Jennifer Stanley <jstanle2@uwo.ca>
Date: Monday, December 21, 2009 1:44 pm
Subject: Influenza C project - Madrenas & Morris
To: Joaquin Madrenas <madrenas@robarts.ca>
Cc: Vincent L Morris <vmorris@uwo.ca>, rsn@uwo.ca

> Hello there Dr. Madrenas:
>
> I understand that you and Dr. Morris are collaborating on a
> project
> involving Influenza C, and that the in vitro work will be done
> at
> Robarts. Can you let me know where in Robarts? The two
> locations I have
> under your name (inspected in July of this year) are Rooms 2278
> and
> 2276. Is the work done in one of these rooms or is it
> going to be done
> in another location...perhaps a shared tissue culture space?
>
> Regards.
>
> Jennifer

MORRIS

Subject: Re: Fwd: Re: Biohazards Subcommittee Protocols
From: Gerald M Kidder <gmk@uwo.ca>
Date: Thu, 24 Dec 2009 12:22:24 -0500
To: Jennifer Stanley <jstanle2@uwo.ca>

Good- consider it approved at CL2.

----- Original Message -----
From: Jennifer Stanley <jstanle2@uwo.ca>
Date: Thursday, December 24, 2009 11:25 am
Subject: Fwd: Re: Biohazards Subcommittee Protocols
To: "avpres@uwo.ca" <avpres@uwo.ca>

>
> Hi Jerry
> It sounds like Dr. Siu is okay with the Morris protocol at Level 2.. if so then this is the third "okay" vote
> Jennifer
>
> -------- Original Message --------
> **Subject:**Re: Biohazards Subcommittee Protocols
> **Date:**Thu, 24 Dec 2009 11:24:11 -0500
> **From:**Jennifer Stanley <jstanle2@uwo.ca>
> **To:**S Siu <srscons@on.aibn.com>
> **CC:**Tyrrel de Langley <tyrreld@uwo.ca>, Stephen Barr <stephen.barr@uwo.ca>, SRS Consultants <srsadmin@on.aibn.com>, Susan Koval <skoval@uwo.ca>, "Gregory

>
> Hi Dr. Siu > Morris will be doing this project at Level 2. > There is no restriction on the number of deviations at this point, but we

Gerald M. Kidder, Ph.D.
Associate Vice-President (Research)
Support Services Building, room 5183
The University of Western Ontario
London, ON N6A 3K7

Modification Form for Permit BIO- UWO-01 74

Permit Holder: Vincent Morris

Approved Personnel (Please stroke out any personnel to be removed)	Additional Personnel (Please list additional personnel here)
Nicole Hague	Dr. Joaquin Madrenas
Ian MacDonald	

	Please stroke out any approved Biohazards to be removed below	Write additional Biohazards for approval below. *
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Approved Cells	rodent (C57B/6), rodent (mmpq), rodent (C57B1/6), mouse melanoma, B1 6F1, B16F10, MDAMB 231	Cloudman S91 mouse melanoma cells purchased from American Type Culture Collection. Noninfectious.
Approved Use of Human Source Material		
Approved GMO	pcDNA 3.1 Hygro, pcDNA 3.1 neo	
Approved use of Animals	mice	
Approved Toxin(s)		

** PLEASE ATTACH A MATERIAL SAFETY DATA SHEET OR EQUIVALENT FOR NEW BIOHAZARDS.*
*** PLEASE ATTACH A BRIEF DESCRIPTION OF THE WORK THAT EXPLAINS THE BIOHAZARDS USED AND HOW THEY WILL BE USED.*

As the principal investigator, I have ensured that all of the personnel named on the form have been trained. 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 of Permit Holder: Vincent Lamoni

Classification: 1

Date of Last Biohazardous Agents Registry Form: Aug 24, 2007

Date of Last Modification (if applicable): Jul 10, 2009

BioSafety Officer(s): G Stanley Dec. 18/09

Chair, Biohazards Subcommittee: E.M. Kidder

Cloudman mouse melanoma cells are used as allogenic cells to investigate immunosuppression by melanoma cells.

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Cell Biology

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Price: $323.00

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- Technical Support
- Related Cell Culture Products

Designations:	Clone M-3 [Cloudman S91 melanoma]
Depositors:	G Sato
Biosafety Level:	1
Shipped:	frozen
Medium & Serum:	See Propagation
Growth Properties:	adherent
Organism:	*Mus musculus* (mouse)
Morphology:	epithelial
Source:	**Organ:** skin **Strain:** DBA **Disease:** melanoma **Cell Type:** melanocyte;
Cellular Products:	melanin
Permits/Forms:	In addition to the MTA mentioned above, other ATCC and/or regulatory permits may be required for the transfer of this ATCC material. Anyone purchasing ATCC material is ultimately responsible for obtaining the permits. Please click here for information regarding the specific requirements for shipment to your location.
Virus Susceptibility:	herpes simplex; vaccinia; pseudorabies; vesicular stomatitis (Indiana)
Virus Resistance:	poliovirus 1
Tumorigenic:	Yes
Reverse Transcript:	positive
Cytogenetic Analysis:	Stemline number is hypertetraploid. Karyotype is stable within stemline number. Marker chromosomes: A medium size chromosome with a submedian centromere and a smaller chromosome with a median centromere., The remaining 81 chromosomes have terminal centromeres, the first one is larger than normal. A minute chromosome was noted in 20% of the cells.
Gender:	male
Comments:	Clone M-3, a melanin-producing cell line was adapted to cell culture by Y. Yasumura, A.H. Tashjian and G. Sato from a Cloudman S91 melanoma in a (C X DBA) F1 male mouse obtained from the Jackson Memorial Laboratory, Bar Harbor, Maine. Tested and found negative for ectromelia virus (mousepox).
Propagation:	

We will also be including the mouse cell line Cloudman S91 melanoma cells. These cells will be purchased from American Type Culture Collection and are pathogen free. We are using these cells because they are allogenic to the mice listed above and allow use to determine the immunosuppressive ability of the melanoma cells in an allogenic host.

Modification Form for Permit BIO-UWO-0174

Permit Holder: Vincent Morris

Approved Personnel
(Please stroke out any personnel to be removed)

Nicole Hague

Ian MacDonald

Additional Personnel
(Please list additional personnel here)

	Please stroke out any approved Biohazards to be removed below	Write additional Biohazards for approval below. *
Approved Microorganisms		E.coli - dH5alpha
Approved Cells	rodent (C57B/6), rodent (mmpq), rodent (C57B1/6), mouse melanoma, B16F1, B16F10	MDAMB 231
Approved Use of Human Source Material		
Approved GMO		pcDNA3.1 Hygro, pcDNA3.1 nco
Approved use of Animals	mice	

see e-mail attached

* *PLEASE ATTACH A MATERIAL SAFETY DATA SHEET OR EQUIVALENT FOR NEW BIOHAZARDS.*
** *PLEASE ATTACH A BRIEF DESCRIPTION OF THE WORK THAT EXPLAINS THE BIOHAZARDS USED AND HOW THEY WILL BE USED.*

Classification: 1

Date of last Biohazardous Agents Registry Form: Aug 24, 2007

Signature of Permit Holder: Vincent Morris

BioSafety Officer(s): JHanley July 10/09

Chair, Biohazards Subcommittee: G M. Kidder

Modification Form for Permit BIO-UWO-0174

Permit Holder: Vincent Morris

Approved Toxin(s)

* *PLEASE ATTACH A MATERIAL SAFETY DATA SHEET OR EQUIVALENT FOR NEW BIOHAZARDS.*
** *PLEASE ATTACH A BRIEF DESCRIPTION OF THE WORK THAT EXPLAINS THE BIOHAZARDS USED AND HOW THEY WILL BE USED.*

Classification: 1

Date of last Biohazardous Agents Registry Form: Aug 24, 2007

Signature of Permit Holder: Vincent Morris

BioSafety Officer(s): [illegible] July 10/09

Chair,Biohazards Subcommittee: G.M. Kidder

Subject: Re: [Fwd: Re: MOdification form: MOrris]
From: Vincent L Morris <vmorris@uwo.ca>
Date: Thu, 25 Jun 2009 20:11:02 -0400
To: Jennifer Stanley <jstanle2@uwo.ca>

To Jennifer Stanley:

The MDAMB 231 cell line is an established breast tumor cell line. We are using it to determine how breast tumor cells spread to and grow in lymph nodes.

Thanks,
Vince

----- Original Message -----
From: Jennifer Stanley <jstanle2@uwo.ca>
Date: Thursday, June 25, 2009 3:10 pm
Subject: [Fwd: Re: MOdification form: MOrris]
To: Vincent L Morris <vmorris@uwo.ca>

> Hi Dr. Morris
> OK - cloning strains of E. coli (ie dH5 alpha)
> Can you provide some details on the use of the MDAMB 231 cell line?
> Thanks
> Jennifer
>
> -------- Original Message --------
> Subject: Re: MOdification form: MOrris
> Date: Wed, 27 May 2009 20:25:12 -0400
> From: Vincent L Morris <vmorris@uwo.ca>
> To: Jennifer Stanley <jstanle2@uwo.ca>
> References: <4A159F0C.1050800@uwo.ca>
> <fc24e515711e.4a1a5723@uwo.ca>
> <4A1A99FF.5030208@uwo.ca> <4A1BFDD1.7080800@uwo.ca>
>
>
>
> Thanks,
>
> Vince Morris
>
> ----- Original Message -----
> From: Jennifer Stanley <jstanle2@uwo.ca>
> Date: Tuesday, May 26, 2009 10:33 am
> Subject: Re: MOdification form: MOrris
> To: Vincent L Morris <vmorris@uwo.ca>
>
> > Dr. Morris
> > I will also put on it E.coli and the plasmids you use in the

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Product Description

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Cell Biology

ATCC® Number: HTB-26™ Order this Item

Designations: MDA-MB-231

Biosafety Level: 1

Medium & Serum: See Propagation

Organism: *Homo sapiens* (human)

Price: $256.00

Depositors: R Cailleau

Shipped: frozen

Growth Properties: adherent

Morphology: epithelial

Source:
Organ: mammary gland; breast
Disease: adenocarcinoma
Derived from metastatic site: pleural effusion
Cell Type: epithelial

Permits/Forms: In addition to the MTA mentioned above, other ATCC and/or regulatory permits may be required for the transfer of this ATCC material. Anyone purchasing ATCC material is ultimately responsible for obtaining the permits. Please click here for information regarding the specific requirements for shipment to your location.

Related Cell Culture Products

Applications: transfection host (Nucleofection technology from Lonza
Roche FuGENE® Transfection Reagents)

Receptors: epidermal growth factor (EGF), expressed
transforming growth factor alpha (TGF alpha), expressed

Tumorigenic: Yes

DNA Profile (STR):
Amelogenin: X
CSF1PO: 12,13
D13S317: 13
D16S539: 12
D5S818: 12
D7S820: 8,9
THO1: 7,9.3
TPOX: 8,9
vWA: 15,18

Cytogenetic Analysis: The cell line is aneuploid female (modal number = 64, range = 52 to 68), with chromosome counts in the near-triploid range. Normal chromosomes N8 and N15 were absent. Eleven stable rearranged marker chromosomes are noted as well as unassignable chromosomes in addition to the majority of autosomes that are trisomic. Many of the marker chromosomes are identical to those shown in the karyotype reported by K.L. Satya-Prakash, et al.

Isoenzymes:
AK-1, 1
ES-D, 1
G6PD, B
GLO-I, 2
Me-2, 1-2
PGM1, 1-2
PGM3, 1

Age: 51 years adult

Use of MDAMB 231 cells by Vincent L. Morris

We are investigating the spread of mammary tumor cells to lymph nodes and other tissues and how these cells can colonize lymph nodes and other organs. Metastasis of mammary tumor cells and their ability to evade the body's immune system is the main reason mammary tumor cells are so deadly. If we can determine the factors affecting the spread of mammary tumor cells and how they evade the bodies immune defenses, we can reduce the number of deaths from this form of cancer. We wil also compare the metastasis of mammary tumor and melanoma cells to assist in determining how both types of tumor cells spread.

Vincent Morris

1.15. Will genetic modifications be made to the microorganisms or biological agents described in 1.1.?	☒ Yes ☐ No	☒ Yes ☐ No	☒ Yes ☐ No	☐ Yes ☐ No	☐ Yes ☐ No
If 'Yes' to 1.15. Complete Table 1.15.1. for Plasmids and/or 1.15.2. for Viral Vectors					
1.15.1. PLASMIDS					
Bacteria Used for Cloning	E coli				
Plasmid(s)	pcDNA3.1Hygro; pcDNA3.1 neo				
Plasmid Source	Invitrogen				
Gene Transfected	Tdtomato;beta galactosidase;mko2hcdf1;mAGhGem				
Describe Resulting Change	marks cells with a staining or fluoresecent tag				
Is this expected to increase the invasiveness, toxicity, or tumorgenicity of the agent in the animal?	☐ Yes ☒ No	☐ Yes ☐ No	☐ Yes ☐ No	☐ Yes ☐ No	☐ Yes ☐ No
1.15.2. VIRAL VECTORS					
Virus Used for Transfection					
Vector(s)					
Vector Source					
Gene Transfected					
Describe Resulting Change					
Is this expected to increase the invasiveness, toxicity, or tumorgenicity of the agent in the animal?	☐ Yes ☐ No	☐ Yes ☐ No	☐ Yes ☐ No	☐ Yes ☐ No	☐ Yes ☐ No

9.1.2. Risk Control Measures – List the control measures required to eliminate or further minimize the risk.

ACTIVITY/HAZARD	RISK CONTROL MEASURES/ACTION
1.2.1. What will you do it someone is bitten or scratched?	injected cells or microspheres pose no added threat to humans, Follow workplace health procedure for bites and scratches (see attached)
1.2.2. How are contaminated materials to be treated prior to disposal?	decontamination by incineration, chemical disinfection or autoclaving
1.2.3. How are the contaminated carcasses to be disposed of?	incineration
1.2.4 List all preventative measures to be taken to minimize the risk of exposure to RESEARCH STAFF handling the material	☒ Safety Glasses ☒ Gloves ☒ Lab coat or equivalent ☒ Mix solutions and handle agent(s) in the fumehood ☐ Contact Workplace Health regarding the medical surveillance required to handle these agents. ☒ N95 Fit-tested Respirator, *specify type*: ☐ Other, *please specify*:
1.2.5. List all preventative	☒ Safety Glasses

Subject: Re: MOdification form: MOrris
From: Jennifer Stanley <jstanle2@uwo.ca>
Date: Tue, 26 May 2009 10:33:53 -0400
To: Vincent L Morris <vmorris@uwo.ca>

Dr. Morris
I will also put on it E.coli and the plasmids you use in the animal protocol, just for the sake of completion (no safety concerns). I assume that you use cloning E.coli (ie dH5 alpha or something like that)?
Jennifer

Jennifer Stanley wrote:
> Got it - thanks!
>
> Vincent L Morris wrote:
> > To Jennifer:
> >
> > I have faxed a copy of the modification to you. I hope you received it.
> >
> > Thanks,
> > Vince
> >
> > ----- Original Message -----
> > From: Jennifer Stanley <jstanle2@uwo.ca>
> > Date: Thursday, May 21, 2009 2:35 pm
> > Subject: MOdification form: MOrris
> > To: Vincent L Morris <vmorris@uwo.ca>
> >
> > > Hi Dr. Morris
> > > Please update this paperwork as soon as possible - your AUS
> > > protocol can
> > > not be approved without it.
> > > Thanks
> > > Jennifer
> > >
> > > -------- Original Message --------
> > > Subject: MOdification form: MOrris
> > > Date: Tue, 12 May 2009 13:57:46 -0400
> > > From: Jennifer Stanley <jstanle2@uwo.ca>
> > > To: Vincent L Morris <vmorris@uwo.ca>
> > >
> > >
> > >
> > > Dr. Morris:
> > > I received an AUS protocol for you which uses the cell line MDA
> > > MB231 cell line. Please complete the attached Modification
> > > form to add this to your existing permit. For more
> > > information, please see:
> > >
> > http://www.uwo.ca/humanresources/docandform/forms/ohs/bio_modification_info.pdf
> > > Thanks!
> > > Jennifer
> > >
> > >
> > >
> > >
> > >

Summary of Approvals for Permit BIO-UWO-0174

Permit Holder: Vincent Morris

Approved Personnel (Please stroke out any personnel to be removed	Additional Personnel
	Dr. Ian McDonald Nicole Hague

	Please stroke out any approved Biohazards* to be removed below	Write additional Biohazards for approval below.
Approved Microorganisms*		
Approved Cells*	rodent (C57B/6), rodent (mmpq)	Rodent (C57Bl/6) mouse melanoma B16F1 and B16F10
Approved Use of Human Source Material*		
Approved GMO*		
Approved use of Animals*	mice	
Approved Toxin(s)*		

Date of last Biohazardous Agents Registry Form Aug 24, 2007

Signature of Permit Holder: Vincent Morris

BioSafety Officer(s): [signature] March 28/07

Chair, Biohazards Subcommittee

G.M. Kidder 28 March 2008

BIO-UWO-0174

THE UNIVERSITY OF WESTERN ONTARIO
BIOHAZARDOUS AGENTS REGISTRY FORM
Revised Biohazards Subcommittee: January, 2007

This form must be completed by each Principal Investigator holding a grant administered by the University of Western Ontario where the use of biohazardous infectious agents are described in the experimental work proposed. The form must also be completed if animal work is proposed involving the use of biohazardous agents or animal carrying zoonotic agents infectious to humans. Containment Levels will be required in accordance with Laboratory Biosafety Guidelines, 3rd edition, 2004, Health Canada (HC) or Containment Standards for Veterinary Facilities, 1st edition 1996, Canadian Food Inspection Agency (CFIA).

Completed forms are to be returned to Occupational Health and Safety (Stevenson-Lawson Building, Room 60) for forward to the Biohazard Subcommittee. For questions regarding this form, please contact the Biosafety Coordinator at extension 81135. If there are changes to the information on this form (excluding grant title and funding agencies) modifications must be completed and sent to Occupational Health and Safety. See website: www.uwo.ca/humanresources

PRINCIPAL INVESTIGATOR Vincent L. Morris
SIGNATURE
DEPARTMENT Microbiology & Immunology
ADDRESS 301 4DSB
PHONE NUMBER 83452
EMAIL vmorris@uwo.ca

Location of experimental work to be carried out: Building(s) MSA Room(s) 312A
*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 it being sent to Occupational Health and Safety (See Section 12.0, Approvals). For research being done at Lawson Health Research Institute, London Regional Cancer Centre, Child and Parent Research Institute or Robarts Research Institute, University Biosafety Committee members can also sign as the Safety Officer.

TITLE OF GRANT(S):
Role of Matrix Metalloproteinases in Cell Movement
(coordinated regulation of epidermal growth factor- and integrin-stimulated migration of primary keratinocytes)a

PLEASE ATTACH A BRIEF DESCRIPTION OF YOUR WORK, SUCH A THE RESEARCH GRANT SUMMARY(S) THAT EXPLAINS THE BIOHAZARDS USED. PROJECTS SUBMITTED WITHOUT A SUMMARY WILL NOT BE REVIEWED.

FUNDING AGENCY/AGENCIES NSERC

Names of all personnel working under Principal Investigators supervision in this location:
i) None
ii)
iii)
iv)
v)

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

1.0 Microorganisms

1.1 Does your work involve the use of microorganisms or biological agents of plant or animal origin (including but not limited to viruses, prions, parasites, bacteria)? ☐ YES ☒ NO
If no, please proceed to Section 2.0

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?
	☐ Yes ☐ No	☐ Yes ☐ No	☐ Yes ☐ No	
	☐ Yes ☐ No	☐ Yes ☐ No	☐ Yes ☐ No	
	☐ Yes ☐ No	☐ Yes ☐ No	☐ Yes ☐ No	
	☐ Yes ☐ No	☐ Yes ☐ No	☐ Yes ☐ No	

1.3 For above named organism(s) or biological agent(s) circle HC or CFIA Containment Level required. 1 2 3

1.4 Source of microrganism(s) or biological agent(s)? _____

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 (ie. derived from fresh tissue) that will be grown in culture in the table below

Cell Type	Is this cell type used in your work?	Source of Primary Cell Culture Tissue
Human	☐ Yes ☐ No	
Rodent	☒ Yes ☐ No	C57Bl6 of MMP9- 2day old pups
Non-human primate	☐ Yes ☐ No	
Other (specify)		

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

Cell Type	Is this cell type used in your work?	Specific cell line(s)	Supplier / Source
Human	☐ Yes ☐ No		
Rodent	☐ Yes ☐ No		
Non-human primate	☐ Yes ☐ No		
Other (specify)	☐ Yes ☐ No		

2.4 If cell lines [illegible] 2.3

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

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 if the following will be used in the laboratory

- Human blood (whole) or other bodily fluids ☐ YES ☐ NO If YES, Specify _____
- Human blood (fraction) or other bodily fluids ☐ YES ☐ NO If YES, Specify _____
- Human organs (unpreserved) ☐ YES ☐ NO If YES, Specify_____
- Human tissues (unpreserved) ☐ YES ☐ NO If YES, Specify_____

3.3 Is human source known to be infected with and infectious agent ☐ YES ☐ NO
If YES , please name infectious agent _____

3.4 For above named materials circle HC or CFIA containment level required 1 2 3

4.0 Genetically Modified Organisms and Cell lines

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

4.2 Will genetic sequences from the following be involved:

- HIV ☐ YES ☐ NO
if YES specify_____
- HTLV 1 or 2 or genes from any CDC class 1 pathogens ☐ YES ☐ NO
if YES specify_____
- Other human or animal pathogen and or their toxins ☐ YES ☐ NO
if YES specify_____

4.3 Will intact genetic sequences be used from

- SV 40 Large T antigen ☐ YES ☐ NO If YES specify_____
- Known oncogenes ☐ YES ☐ NO If YES specify_____

4.4 Will a live vector(s) (viral or bacterial) be used for gene transduction ☐ YES ☐ NO
If YES name virus_____

4.5 List specific vector(s) to be used:_____

4.6 Will virus be replication defective ☐ YES ☐ NO

4.7 Will virus be infectious to humans or animals ☐ YES ☐ NO

4.8 Will this be expected to increase the Containment Level required ☐ YES ☐ NO

5.0 Human Gene Therapy Trials

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

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

5.3 How will the virus be administered? _____

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

5.5 Has human ethics approval been obtained? ☐ YES ☐ NO

6.0 Animal Experiments

6.1 Will any of the agents listed be used in live animals? ☐ YES ☒ NO
If no, please proceed to section 7.0

6.2 Name of animal species to be used mice qj

6.3 AUS protocol # 2005-017-04 renewed May 1/07 qj.

6.4 If using murine cell lines, have they been tested for murine pathogens? ☐ YES ☐ NO

7.0 Use of Animal species with Zoonotic Hazards

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

- Pound source dogs ☐ YES ☒ NO
- Pound source cats ☐ YES ☒ NO
- Sheep or goats ☐ YES ☒ NO
- Non- Human Primates ☐ YES ☒ NO If YES specify species _____
- Wild caught animals ☐ YES ☒ NO If YES specify species _____
  colony # _____

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 _____

8.3 What is the LD_{50} (specify species) of the toxin _____

9.0 Import Requirements

9.1 Will the agent be imported? ☐ YES ☒ NO
If no, please proceed to Section 10.0
If yes, country of origin_____

9.2 Has an Import Permit been obtained from HC for human pathogens? ☐ YES ☐ NO

9.3 Has an import permit been obtained from CFIA for animal pathogens? ☐ YES ☐ NO

9.4 Has the import permit been sent to OHS? ☐ YES ☐ NO
If yes, Permit # _____

10.0 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

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

SIGNATURE Vincent L'Abbé

11.0 Containment Levels

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

11.2 Has the facility been certified by OHS for this level of containment? ☒ YES ☐ NO
via letter

11.3 If yes, please give the date and permit number. N/A

12.0 Approvals

UWO Biohazard Subcommittee:

Signature [signature] Date 24 Aug. '07

Safety Officer for Institution where experiments will take place

Signature [signature] Date Aug. 24/07

Safety Officer for University of Western Ontario (if different than above)

Signature _____ Date _____

1. EXPERIMENTATION MUST BE APPROVED TO THIS FORM OR PROJECT WILL NOT BE APPROVED