Modification Form for Permit BIO-LHRI-0053

Permit Holder: John Lewis

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

Additional Personnel
(Please list additional personnel here)

Amy Robertson
Hon Leong

	Please stroke out any approved Biohazards to be removed below	Write additional Biohazards for approval below. *
Approved Microorganisms	E. coli	
Approved Cells	Human (established), HT-1080, PC-3, DU145	
Approved Use of Human Source Material		
Approved GMO	pSM2	PL3-TRE-LucGFP-2L
Approved use of Animals		
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: _____

Classification: 1

Date of Last Biohazardous Agents Registry Form: Sep 13, 2007

Date of Last Modification (if applicable): _____

BioSafety Officer(s): Pete Ferguson, Dave Ryder Dec. 9/09

Chair, Biohazards Subcommittee: _____

Description of work to be performed with biohazard:

The new material to be added to the biohazard approval form is a plasmid from Addgene (plasmid 11685). This agent is named **pL3-TRE-LucGFP-2L**, and is a 7117 base pair DNA plasmid that can be activated within the cell upon addition of a tetracycline derivative, doxycycline. Doxycycline converts the tetracycline repressor protein (already present in the cell) to its active form, initiating translation of the plasmid via actively binding to the tetracycline responsive element (TRE) located within the plasmid. Translation of the plasmid will allow detection of this tetracycline repressor protein within the cell by reporting both luciferase and EGPF proteins, both located downstream of the TRE. **This plasmid does not contain any human genes.**

Identification of the tetracycline repressor protein within the cell will aid in the confirmation of a cell line containing this inducible protein that has been created in our lab. The plasmid, contained in a previously approved organism (E.coli DH5a), will be propagated and isolated in the laboratory. The plasmid will also be subjected to digestion using restriction enzymes, transfected into HT-1080 cells using a commercial agent, and will be identified within the cell by fluorescence microscopy and the luciferase assay.

BIO-LHRI-0053

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 Jim Lewis
SIGNATURE _____
DEPARTMENT Oncology
ADDRESS LRCP Rm FH-323
PHONE NUMBER x52194
EMAIL jim.lewis@lhsc.on.ca

Location of experimental work to be carried out: Building(s) LRCP Room(s) FH-823
*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):
Hyaluron-mediated intravasation and tumour cell metastasis in the dissemination of cancer

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 NCIC

Names of all personnel working under Principal Investigators supervision in this location:
i) Heather Pilecki
ii) Qian Liu
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?
E. coli	☐ Yes ☐ No	☐ Yes ☐ No	☐ Yes ☐ No	1L culture.
	☐ 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)? Invitrogen (ONE SHOT).

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	
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	HT-1080, PC-3, DU145	ATCC
Rodent	☐ Yes ☐ No		
Non-human primate	☐ Yes ☐ No		
Other (specify)	☐ Yes ☐ No		

2.4 For above named cell types(s) circle HC 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 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 N/A
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: N/A * transfection using pSM2 vector

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

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

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? N/A

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 N/A

6.3 AUS protocol # _____

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 N/A

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 _____

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

11.3 If yes, please give the date and permit number: Dec. 20/06 · 802-5787

12.0 Approvals

UWO Biohazard Subcommittee

Signature C.M. Kidder Date 13 Sept. '07

Safety Officer for Institution where experiments will take place

Signature Simon Fraser Date June 29, 2007.

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

Signature J. Stanley Date Sept 12/07

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