

# Modification Form for Permit BIO-RRI-0050

Permit Holder: David Holdsworth

## Approved Personnel

(Please stroke out any personnel to be removed)

Hristo Nikolov

Chris Norley

## Additional Personnel

(Please list additional personnel here)

Craig Tschirhart

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

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Approved Primary and Established Cells

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Approved Use of Human Source Material

tissues (unpreserved), bone specimens

procedure with unpreserved tissue within biological safety cabinet

Approved Genetic Modifications (Plasmids/Vectors)

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(see attached)

Approved Use of Animals

rats

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Approved Biological Toxin(s)

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\* 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: 

Current Classification: 2 Containment Level for Added Biohazards: 2

Date of Last Biohazardous Agents Registry Form: Jul 10, 2008

Date of Last Modification (if applicable): \_\_\_\_\_

BioSafety Officer(s):  April 08/11

Chair, Biohazards Subcommittee: \_\_\_\_\_ Date: \_\_\_\_\_

## ***Meniscal Injury Mechanics Experimental Protocol***

***Craig Tschirhart, PhD Candidate, University of Guelph***

### ***Note***

All researchers in contact with cadaveric tissue, must be within a Medical Surveillance program and have up to date tetanus and Hepatitis B vaccinations.

### ***Materials***

1. Cadaveric Knee Joint
2. Scalpal
3. Ultrafix Suture Device
4. Potting Materials
5. 6 DOF Mechanical Loading Apparatus
6. Muscle Angle Eyelet Screws
7. Bone Screws
8. Drill
9. 8 0.7mm Stainless steel beads
10. 4 hypodermic needles (gauge?)
11. 4 spinal needles
12. Bead tetrahedral injection device
13. Butcher knife
14. Surgical gloves
15. Fume Hood
16. 20 dixie cups
17. 10 popsicle sticks
18. Suturing material
19. Radiostereometric Analysis (RSA) Image Acquisition System (Robarts Research Institute)

### ***University of Guelph Procedure***

1. Thaw fresh frozen joint for 12 hours
2. Remove soft tissue down to ligaments and tendon insertions with joint capsule intact using a butcher knife
3. Remove and save a portion of tendon tissue
4. Thread tendon cable around bone screws
5. Drill bone screws into bone at patellar tendon and hamstring tendon insertion sites
6. Drill muscle angle eyelet guides into the femur along the midpoint of the respective muscle actuation angles
7. Load the metallic beads and a small amount of tendon tissue into each of the 4 hypodermic needles using a scalpel
8. Load the hypodermic needles into the bead tetrahedral injection device

9. Place the bead tetrahedral injection device along the posterior medial peripheral surface of the meniscus (ensure that each of four holes line up with the accessible meniscus surface)
10. Insert the spinal needles into the back of the bead injection device to push the bead and tendon material into the meniscus
11. Repeat the bead injection procedure for the anterior medial peripheral surface of the meniscus
12. Remove the proximal femur and distal shank until the joint sits in between the bone pots with all of the femoral condyles and tibial plateau exposed between the pots
13. In the fume hood, fill 9 dixie cups  $\frac{3}{4}$  full of powder bone cement
14. For each Dixie cup, fill the remaining contents with the liquid bone cement. Stir until reaching a fairly viscous consistency. Pour the contents into the tibia pot.
15. Once completed all cups, insert the tibia joint such that the tibial plateau faces vertically upwards in alignment with the pot
16. Hold the joint in position for approximately 20 minutes to allow the bone cement to settle
17. Once the cement has hardened, repeat the potting procedure for the femur joint. When inserting the joint, ensure that it is at an anatomic joint angle of zero. Ensure that the direction of the femur is parallel to the sides of the pot
18. Hold the joint in position for approximately 20 minutes to allow the bone cement to settle
19. Seal the joint in plastic wrap, then place on ice in a cooler and transport to London facility with remaining testing materials

#### **Robarts Research Institute Procedure**

1. Place a fit-tested N95 respirator over nasal cavities and mouth (OPTIONAL).
2. Remove the joint from the cooler
3. Attach the joint to the mechanical testing apparatus
4. Loop the tendon cables around the upper turnbuckle hooks
5. Connect the load cells to the power source and data acquisition system
6. Turn on the laptop and open the labview DAQ software
7. Run the DAQ program.
8. Apply small loads to the joint and tendon cables. Ensure that all loads are registering with the program.
9. Stop the program.
10. Operator 1 maintains the joint vertically by holding the top portion of the loading apparatus.
11. Operator 2 applies forces by loading the weight stacks with weight plates in accordance with the predetermined load. At each weight increment, adjust the turnbuckle hooks until the joint becomes stable without assistance from Operator 1.
12. Run the DAQ program
13. Clear the testing area and capture RSA images while collecting load information to the DAQ system.
14. Remove the joint from the apparatus.
15. Transport to Biohazard-approved Level 2 location with a Biological Safety Cabinet (Rm. 2245E)
16. Place the specimen in the Biological Safety Cabinet

is this still in a plastic wrap or container?

See E-mail

17. Remove the plastic sealant
18. Induce a meniscal beak tear in the transitional zone between corpus and posterior horn
19. Reseal the joint and remove from Biological Safety Cabinet
20. Repeating the loading and data acquisition procedure for the torn meniscus scenario (steps 6 through 16)
21. Repair the meniscus tear using the Ultrafix repair implant
22. Reseal the joint
23. Repeating the loading and data acquisition procedure for the repaired meniscus scenario (steps 6 through 16)
24. Sever the ACL
25. Reseal the joint
26. Repeating the loading and data acquisition procedure for the torn ACL scenario (steps 6 through 16)
27. Remove joint from loading device and wrap in leak proof plastic and place in a cooler
28. Wipe all surfaces with 70% ethanol and use enough 70% ethanol to give a contact time of a minimum of 10 minutes
29. Dispose of all waste, such as gloves and plastic, in a Biohazard Waste container
30. Seal surgical equipment in a leak proof container and return to Guelph for Autoclaving
31. Transport joint back to University of Guelph in a leak-proof container for storage and disposal

### ***Emergency Procedures***

In the event of a spill or container breakage resulting in the unintentional release of a biological agent:

- (i) a paper towel or absorbent will be placed on the liquid
- (ii) a strong disinfectant solution or granules (i.e. Quatricide PV germicidal detergent) will be poured around, but not on the spill, and the disinfectant will be mixed with the spilled material cautiously;
- (iii) the laboratory will be evacuated for 20 minutes to allow for decontamination;
- (iv) paper will be carefully placed into a bag for incineration;
- (v) all surfaces exposed to the spill will be decontaminated with the disinfectant.

## Ron Noseworthy

**From:** Craig Tschirhart [ctschirh@uoguelph.ca]  
**Sent:** Thursday, April 07, 2011 5:09 PM  
**To:** 'David Holdsworth'; Ron Noseworthy  
**Cc:** Greg Dekaban; 'Mark Hurtig'; 'Xunhua Yuan'  
**Subject:** RE: Human Cadaveric Specimen Biohazard Protocol

Yes, the specimens will be sealed in plastic during mechanical testing in the RSA lab.

**From:** David Holdsworth [mailto:david.holdsworth@imaging.robarts.ca]  
**Sent:** Thursday, April 07, 2011 5:04 PM  
**To:** Ron Noseworthy  
**Cc:** Greg Dekaban; Mark Hurtig; Craig Tschirhart; Xunhua Yuan  
**Subject:** Re: Human Cadaveric Specimen Biohazard Protocol

Hi Ron,

Yes, as far as I know, the intent is that the specimen is sealed (wrapped or contained) in plastic during the experiments in the RSA room, and then unsealed in the biohazard hood for the required manipulations.

I hope that Mark, Craig, or Xunhua can confirm if this is correct. (steps 1 -16 are attached)

Regards,

David W. Holdsworth, Ph.D.  
Dr. Sandy Kirkley Chair in Musculoskeletal Research  
Schulich School of Medicine and Dentistry  
University of Western Ontario  
London, Ontario CANADA  
(519) 931-5777 x24154

E-mail

Place a fit-tested N95 respirator over nasal cavities and mouth

Remove the joint from the cooler

Attach the joint to the mechanical testing apparatus

Loop the tendon cables around the upper turnbuckle hooks

Connect the load cells to the power source and data acquisition system

Turn on the laptop and open the labview DAQ software

Run the DAQ program.

Apply small loads to the joint and tendon cables. Ensure that all loads are registering with the program.

Stop the program.

Operator 1 maintains the joint vertically by holding the top portion of the loading apparatus.

Operator 2 applies forces by loading the weight stacks with weight plates in accordance with the predetermined load.

04/08/2011

ach weight increment, adjust the turnbuckle hooks until the joint becomes stable without assistance from Operator 1.

Run the DAQ program

Clear the testing area and capture RSA images while collecting load information to the DAQ system.

Remove the joint from the apparatus.

Transport to Biohazard-approved Level 2 location with a Biological Safety Cabinet (Rm. 2245E)

Place the specimen in the Biological Safety Cabinet

On Apr 7, 2011, at 4:11 PM, Ron Noseworthy wrote:

Hi Dr. Holdsworth,

Dr. Dekaban had a look at the modification and has a question regarding the procedure. I dropped down to your office but you were not there.

He wants to know if in steps 1 – 14 the joint is still in plastic wrap or a plastic container.

Thanks

Ron

**From:** David Holdsworth [mailto:david.holdsworth@imaging.robarts.ca]

**Sent:** Thursday, April 07, 2011 3:43 PM

**To:** Ron Noseworthy

**Cc:** Craig Tschirhart; david.holdsworth; [mhurtig@uoguelph.ca](mailto:mhurtig@uoguelph.ca)

**Subject:** Re: Human Cadaveric Specimen Biohazard Protocol

Hi Ron,

Here is the modified protocol, which identifies the location of the biosafety cabinet (Rm 2245E), and includes the use of Quatricide PV as the preferred disinfectant, rather than bleach. Please attach this to the protocol modification form that I have signed.

I understand that the committee will meet tomorrow, so I hope that there is some way that they can include this quick review. There is very little change to our existing protocol for unpreserved human tissue, expect for the component that will be performed within the hood.

I have already held up Dr. Hurtig's program by delaying this, I'm afraid.

Let me know if there is anything else that I can do to expedite this. I'm sorry for the delay.

Regards,

David W. Holdsworth, Ph.D.  
Dr. Sandy Kirkley Chair in Musculoskeletal Research  
Schulich School of Medicine and Dentistry  
University of Western Ontario

04/08/2011

London, Ontario CANADA  
(519) 931-5777 x24154



## USE OF ROBARTS IMAGING SUITES: BIOSAFETY REQUIREMENTS FOR *IN VIVO* AND *IN VITRO* WORK

Approved: February, 2009  
University of Western Ontario Biosafety Committee  
(Original Approved February 13, 2008)

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### 1.0 Introduction and Scope:

Imaging Facilities at Robarts are used for *in vitro* and *in vivo* work by researchers throughout London affiliated with the University of Western Ontario. The objective of this document is to ensure that this research meets the standards set by the latest versions of the Health Canada Laboratory Biosafety Guidelines, the Containment Standards for Veterinary Facilities by the Canadian Food Inspection Agency and, where animals are involved, the Canadian Council for Animal Care (CCAC). This work must also follow the Biosafety Guidelines and Procedures Manual found at: [www.uwo.ca/humanresources/biosafety](http://www.uwo.ca/humanresources/biosafety).

The goal of this document is to ensure that *in vitro* and *in vivo* experiments meet all applicable guidelines and regulations and are done within the proper containment to protect the work, the animals, the facilities, and the faculty, staff, and students who perform the work.

- This document applies to the 3T MRI, 9.4T Imaging Suite (MRI), Human High Field MRI Laboratory (3T & 7T), and Preclinical Imaging Suite (MicroCT, Ultrasound, SPECT CT) imaging facilities and includes procedures for transport to the facility. With respect to the primate facilities (including the 9.4T MRI suite), upon arrival at the facility the approved facility SOPs take effect.
- This document applies only to containment level 1 (CL1), level 2 (CL2) or level 2 with level 3 operations. Research requiring level 3 containment must contact the Biosafety Officer at [biosafety@uwo.ca](mailto:biosafety@uwo.ca). Level 2 research involving live non-human primates must follow the Standard Operating Procedures (SOPs) for the Center for the Brain and Mind.

### 1.1 General Safety Precautions for *In vivo* and *In vitro* Imaging

- All personnel operating the imaging equipment (9.4T MRI, 3T MRI, 7T MRI, MicroCT, SPECT CT, and Ultrasound) must be trained by Facility Manager or designate.
- All personnel handling animals must have the required Animal Care and Veterinary Services training.

- All animal work must be outlined in an approved animal use protocol.
- All personnel using the Imaging facilities must be trained and follow the Standard Operating Procedures (SOPs) in place for each facility.
- Supervisors must ensure that people using the Imaging facilities have the appropriate health and safety training for the work being performed, per the Health and Safety Training found at:  
[http://www.uwo.ca/humanresources/facultystaff/h\\_and\\_s/training/training\\_idx.htm](http://www.uwo.ca/humanresources/facultystaff/h_and_s/training/training_idx.htm)
- Personnel using each Imaging facility must wear the appropriate personal protective equipment. For more information, see the Laboratory Safety Manual, [www.uwo.ca/humanresources](http://www.uwo.ca/humanresources) or contact the Lab Safety Coordinator.
- Disposal of waste, including hazardous chemical waste, biomedical waste, animal waste and carcasses, must follow the Hazardous Material Management Handbook: [http://www.uwo.ca/humanresources/facultystaff/h\\_and\\_s/enviromental\\_prog/enviromental\\_idx.htm](http://www.uwo.ca/humanresources/facultystaff/h_and_s/enviromental_prog/enviromental_idx.htm)
- Work carried out must meet the requirements of the Biosafety Guidelines and Procedures Manual found at: [www.uwo.ca/humanresources/biosafety](http://www.uwo.ca/humanresources/biosafety)
- Personnel should complete their Hazard Communication Form and have the appropriate medical surveillance. For information, please see: <http://www.wph.uwo.ca/newposition.htm>.
- In case of an emergency, such as medical or fire, personnel follow the SOPs in place for the facility accessible on-line or in the Robarts Health and Safety Office.

Preclinical Imaging Suite: SOP 900 – Emergency Procedures

9.4T MRI Facility: SOP 300 – Standard Operating Procedure:  
Emergency Fire Procedures

3T MRI Facility: SOP 3T 215, 210, and 205 – Standard Operating  
Procedures for Emergency Quench, Fire Code Blue

Human High Field MRI Lab: SOP 220, 230, and 210 – Emergency Fire,  
Emergency Quench, Emergency Code Blue

Where there is an emergency involving human and animal wellbeing, human health and safety is the priority.

- The Principal Investigator must have an approved, current Biohazardous Agents Registry Form on file with the Biosafety Office which reflects the research being done. For more information, see: [www.uwo.ca/humanresources/biosafety](http://www.uwo.ca/humanresources/biosafety).
- The Biosafety Officer(s) in association with the Director, Animal Care and Veterinary Services and the Biohazard Subcommittee determine the containment level required for the work being performed.

## 1.2 Transportation of Animals

### 1.2.1 Transportation of Level 1 Rodents

Level 1 rodents are those not exposed to a CL2 (or higher CL) agent via ingestion, inhalation, injection, or absorption and are not known to carry a level 2 zoonotic agents. Level 1 rodents may be transported to the Robarts imaging facilities and within the Robarts building using standard cages. Level 1 rodents may be transported to the University or within the University buildings in standard cages.

### 1.2.2 Transportation of Level 2 Rodents

Level 2 rodents are those which have been exposed to a CL2 agent. Level 2 rodents must be transported in a HEPA-filtered cages or an apparatus. The cages or apparatus must be approved by the Director, ACVS and the Biosafety Officer(s) for Robarts. The transportation of level 2 animals by road, rail, water or air must also follow the appropriate transportation of dangerous goods regulations.

### 1.2.3 Transportation of Non Human Primates (NHP)

Transportation of non human primates is governed by a separate set of SOPs that have been approved by ACVS, members of the Centre for Brain and Mind, and the Biosafety Officers for Robarts. These SOPs are available in the Brain and Mind Facility or the Robarts Health and Safety office and are to be followed for the transportation of primates (NHP) to and from the primate (NHP) quarters and the MRI suites.

## 2.0 Introduction to Rodent and Non Human Primate (NHP) Imaging Research

Animal projects must be approved by the Animal Use Subcommittee of the University Council on Animal Care. Animals are housed in areas approved by Animal Care and Veterinary Services (ACVS) and the Canadian Council on Animal Care (CCAC). Animals are transported to the facility in cages on carts.

### 2.1 Imaging Involving Level 1 Rodents

- Level 1 rodent work involves rodents that have not been exposed to a level 2 (or higher CL) agent via ingestion, inhalation, injection or absorption and that are not known to carry a level 2 (or higher CL) zoonotic agent. An example of a level 1 rodent is an animal procured from a commercial supplier or one injected with a murine pathogen free cell line approved by Biosafety at level 1.

#### 2.1.1 Safety Precautions

- Follow the Standard Operating Procedure (SOPs) for the decontamination of samples entering the facility and the clean-up of animal excrement, including surface disinfection. Disinfectants

must be approved by the Biosafety Officer or in the SOP and must be effective and safe to use on the equipment. The SOPs are available on-line or in the Robarts Health and Safety Office.

Third Floor Preclinical Imaging Suite: SOP 500 – Cleaning and Decontamination  
First Floor 9.4T MRI Facility: SOP 415 – Cleaning and Disinfection – Level 1 & 2 Experiments  
Second Floor 3T MRI Facility: SOP 400 – Standard Operating Procedure for MRI Decontamination  
First Floor Human High Field MRI Lab: SOP 415 – Cleaning and Decontamination – Level 1 & 2 Experiments

- Gloves and other personal protective equipment must be changed if they have been in contact with animal wastes.
- Procedures such as injections, surgery, anesthesia, and euthanasia can be done on the open bench. Scavenging devices must be used in association with anesthesia or euthanasia with a gaseous agent. If a hazardous chemical or radioactive material is involved, this may require the use of a fume hood elsewhere and additional precautions/approvals.
- The animal may be placed in the coil or bed on the open bench.
- In case of a veterinary emergency, life-saving procedures can be done on the open bench.

## 2.2 Imaging Involving Level 2 Rodents

- Level 2 Rodent work involves animals that have been exposed to a level 2 agent via ingestion, inhalation, injection or absorption or carry a level 2 zoonotic agent. Examples of level 2 pathogens include:
  - ◆ Viral vectors such as adenovirus and retroviruses
  - ◆ Human cell lines such as HEK293, which carries an activated human oncogene, or non-human primate cell lines such as cos-7, because they may carry viruses capable of infecting humans
  - ◆ Microorganisms such as Salmonella sp. or Pseudomonas sp.
  - ◆ Biological toxins such as pertussis and cholera toxin.

Contact the Biosafety Officer at [biosafety@uwo.ca](mailto:biosafety@uwo.ca) for the containment level of the project. For more information, please see [www.uwo.ca/humanresources/biosafety](http://www.uwo.ca/humanresources/biosafety)

## 2.2.1 Safety Precautions

For level 2 projects, there are additional Safety Precautions to those in Section 2.1.1.

- Level 2 agents must be handled in a Class 2 biological safety cabinet. Animals that have been exposed to a level 2 agent must be kept in an approved HEPA-filtered cage or apparatus during the duration of the experiment, including housing, transportation, imaging and during veterinary life saving measures.
- Personnel using an approved HEPA-filtered cage or apparatus must have a plastic container with them. In case of failure or leakage of the cage or apparatus, the cage or apparatus (with the animal inside) is put in the plastic container. The container can only be opened in a biological safety cabinet.
- Animals exposed to a level 2 agent must be housed in a certified ACVS approved level 2 housing facility.

### 2.2.1.1 Preclinical Imaging Suite and Second Floor 3T MRI Facilities

Personnel can transport the animals in a HEPA-filtered cage to the imaging facility. The cage must be opened in the biological safety cabinet to perform procedures such as injections, anesthesia and veterinary life saving measures. The animal is placed in a HEPA-filtered apparatus for imaging in the biological safety cabinet. After imaging, the rodent is transported to a biological safety cabinet in an approved level 2 housing facility. The apparatus is never opened except in a biological safety cabinet.

The apparatus must be certified by a certified contractor such as HEPA Filters Inc. The apparatus must be approved by the Biosafety Officers for Robarts and Animal Care and Veterinary Services. The apparatus must maintain level 2 containment, and requires safety features such as HEPA filtration, O-rings, threaded ends.

HEPA-filtered cages must be approved by the Biosafety Officers for Robarts and by Animal Care and Veterinary Services.

Waste is collected from the biological safety cabinet in bags. The bag is closed in the biological safety cabinet and disposed of by the research personnel. Carcasses are disposed of by research personnel. Waste is disposed of per the Hazardous Materials Management Handbook.

## 2.2.1.2 9.4T MRI Facility and Human High Field MRI Laboratory (3T & 7T)

### 2.2.1.2.1 Approach #1

This facility does not contain a biological safety cabinet. Procedures must be done in a biological safety cabinet in an approved level 2 facility elsewhere.

Animals must be placed in an approved HEPA-filtered imaging apparatus in a biological safety cabinet in an approved level 2 laboratory. Animals are transported to the facility and imaged in this apparatus. The apparatus is never opened except in a biological safety cabinet.

Waste is collected in autoclaveable bags and disposed of by the research personnel. Carcasses are also disposed of by research personnel. Waste is disposed of per the Hazardous Materials Management Handbook.

The apparatus must be certified by a certified contractor such as HEPA Filters Inc. The apparatus must be approved by the Biosafety Officers for Robarts and Animal Care and Veterinary Services. The apparatus must maintain level 2 containment, and requires safety features such as HEPA filtration, O-rings, threaded ends.

### 2.2.1.2.2 Approach #2

In some cases, approach #1 is impractical; approach #2 can then be used for level 2 rodents. This is based on a case-by-case risk assessment and is approved by the Biosafety Officers for Robarts and Animal Care and Veterinary Services.

When the rodents have been previously exposed to a level 2 agent, they are brought to the MRI facilities for imaging using an approved HEPA-filtered transport cage on a cart and placed in the appropriate imaging insert coils.

Approach #2 for MRI and fiber optic imaging of level 2 animals in the MRI suites is based on designing and constructing the whole lab to be under level 2 containment. This means that the air entering and leaving the MRI suites is HEPA-filtered. Entrance is through a controlled air lock and the room is under negative air pressure to the adjacent corridor. Personnel must wear the appropriate personal protective equipment as mandated by the MRI Facility's SOP 210-01. This includes the wearing of a fit-tested N95 respirator when working with level 2 animals as a biological safety cabinet is not available. Protective clothing must be removed before leaving the MRI facilities

as stated in SOP 210. Decontamination procedures for the suites are outlined in the Facility's SOP 415 and the MRI Suite Decontamination Procedures: SOP 3900 for the Center for Brain and Mind. Researchers must follow the Use of MRI Suite for NHP Imaging: SOP 4600 for the Center for Brain and Mind. Personnel must be specially trained to work in the MRI level 2 containment suites.

Waste is collected in autoclaveable bags and disposed of by the research personnel. Carcasses are also disposed of by research personnel. Waste is disposed of per the Hazardous Materials Management Handbook.

## 2.2 Imaging Involving Non-Human Primates

Approach #2 for MRI and fiber optic imaging of level 2 animals in the MRI suites is based on designing and constructing the whole lab to be under level 2 containment. This means that the air coming in and leaving the MRI suites is HEPA-filtered. Entrance is through a controlled air lock and the room is under negative air pressure to the adjacent corridor. Personnel must wear the appropriate personnel protective equipment as mandated by the MRI Facility's SOP 210-01. This includes the wearing of a fit-tested N95 respirator when working with level 2 animals as a biological safety cabinet is not available. Protective clothing must be removed before leaving the MRI facilities as stated in SOP 210. Decontamination procedures for the MRI suites are outlined in the Facility SOP 415 and the MRI Suite Decontamination procedures for the suites are outlined in the Facility's SOP 415 and the MRI Suite Decontamination Procedures: SOP 3900 for the Center for Brain and Mind. Researchers must follow the Use of MRI Suite for NHP Imaging: SOP 4600 and other Center for Brain and Mind Rhesus Facility Standard Operating Procedures. Personnel must be specially trained to work in the MRI level 2 containment suites.

## 3.0 Introduction to *In vitro* Research Involving Imaging

Samples are prepared for imaging in an approved biosafety laboratory. Samples are brought to the imaging facility in sealed leak- and shatter-proof containers. Samples are put in a coil or a bed and/or HEPA-filtered apparatus for imaging purposes.

### 3.1 Imaging Involving Fixed Samples

Level 2 or level 2+3 samples fixed with chemicals such as formalin or comparable agent are no longer considered biohazardous. These samples can be imaged as level 1 samples. If samples need to be opened, they should be opened in a chemical fume hood.

### 3.2 Imaging Involving Level 1 *In Vitro* Work

Samples must be transported to the facility in sealed leak- and shatter-proof containers. Containers must be wiped off with a disinfectant before they leave the laboratory and per the SOPs for the facility. Work with these samples can be done on the open bench, providing that no hazardous chemicals are involved. If hazardous chemicals or radioactive materials are involved, work must be done in a fume hood elsewhere and additional precautions/approvals are required.

### 3.3 Imaging Involving Level 2 *In vitro* Work

For level 2 projects, there are additional safety precautions to those in 3.1. Samples must be worked with using a biological safety cabinet.

#### 3.3.1 Preclinical Imaging Suite and 3T MRI Facilities

If required, samples can be opened under the biological safety cabinet provided.

#### 3.3.2 9.4T MRI Facility and Human High Field MRI Laboratory (3T & 7T)

There are no biological safety cabinets in these facilities. Samples must be prepared in a biological safety cabinet in an approved level 2 laboratory elsewhere. Sealed leak- and shatter-proof containers are not to be opened in the facilities. The sample is kept closed during transportation and imaging of the samples.

### 4.0 Imaging Involving Work at Level 2 plus Level 3 Operations

The researcher must have an approved, current Biohazardous Agents Registry Form on file with the Biosafety Office which reflects the research being done. For more information, see: [www.uwo.ca/humanresources/biosafety](http://www.uwo.ca/humanresources/biosafety)

Certain projects, such as some research involving lentiviral-based vectors, require level 2 plus level 3 operations. For level 2 plus level 3 projects there are additional safety precautions. All work must be carried out in a biological safety cabinet.

#### 4.1 Imaging

- Use portable autoclave to decontaminate waste prior to leaving the imaging facility. Follow the “SOP for the Sanyo Portable Autoclave”.
- Injections must be done in the approved level 2 plus 3 laboratory or the level 3 facility on DSB, 6<sup>th</sup> floor.
- Animals transported on a cart to or within Robarts for imaging must be in a HEPA-filtered cage unit approved by Biosafety and ACVS.
- The cages can be removed from the transport cart and placed in a biological safety cabinet. Animals must be placed in an approved HEPA-filtered imaging apparatus (see section 2.2.1.1) in a biological safety cabinet in an approved level 2 plus 3 laboratory. Animals are transported to the facility and imaged in this apparatus. The apparatus is never opened except in a biological safety cabinet.
- After scanning, all reusable material (i.e. forceps) must be decontaminated in a Wescodyne solution in a biological safety cabinet. The Wescodyne working solution has: 40% H<sub>2</sub>O, 40% ethanol and 20% Wescodyne. It can be prepared in advance.
- Submerge all the reusable instruments (surgical) in the labelled Wescodyne solution for 2 hours.
- Rinse the instruments after 2 hours with H<sub>2</sub>O and let dry.

- After drying, pack in autoclave bags and sterilize in the portable autoclave (this is done to ensure successful sterilization).
- The procedures for disinfection of contaminated animal cages and bedding must be completed. Bedding must be emptied into a biohazard bag inside of the biosafety cabinet. The bedding must be then double bagged and sealed inside a biological safety cabinet. The bag must be wiped with a disinfectant before it is removed from the biological safety cabinet for disposal per the Hazardous Materials Management Handbook.
- Inside the hood, to the empty cage add Wescodyne solution and swirl to ensure contact of all surfaces. Wipe the cage lid with Wescodyne as well and ensure contact for 2 hours (either leave the cage in a dunk tank for 2 hours or put the wet cage into an autoclave bag and leave in the hood for 2 hours). Drain the Wescodyne and return the cages and lids for washing and packing to be autoclaved. Follow the procedures for the facility where the cages came from (ACVS or Robarts barrier facility).
- All sharps must be disposed of in a sharps container within the biosafety cabinet. The container must be wiped on the outside with the Wescodyne solution. The containers are then sent to the incinerator.
- All waste must be labelled appropriately before it is taken for disposal.
- After the scan the rodent/animal must be returned to the biological safety cabinet before it is removed from the HEPA-filtered apparatus and then it can be returned to its cage.
- Disposable personal protective equipment, such as gloves, must be put in an autoclaveable biohazard bag leaving the room.
- Wescodyne solution can be treated as hazardous waste after use per the Hazardous Waste Management Handbook:  
[http://www.uwo.ca/humanresources/docandform/docs/ohs1/manuals/hazardous\\_handbook.pdf](http://www.uwo.ca/humanresources/docandform/docs/ohs1/manuals/hazardous_handbook.pdf).

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, 1<sup>st</sup> 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](http://www.uwo.ca/humanresources)

PRINCIPAL INVESTIGATOR David Holdsworth  
 SIGNATURE [Signature]  
 DEPARTMENT Imaging Research  
 ADDRESS Robarts Research Institute  
 PHONE NUMBER 663 5777 x 34154  
 EMAIL David.Holdsworth@imaging.robarts.ca

Location of experimental work to be carried out: Building(s) Robarts Room(s) Pre-clinical Imaging  
 \*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):  
Advanced pre-clinical anatomical and dynamic micro-computed tomography.

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 C.H.R.

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

- i) Chris Norley
- ii) Hristo Nikolov
- iii) \_\_\_\_\_
- iv) \_\_\_\_\_
- v) \_\_\_\_\_

## 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?                | Is it known to be an animal pathogen?              | Is it known to be a zoonotic agent?                | Maximum quantity to be cultured at one time? |
|-----------------------------|--|--|--|--|
|                             | YES/NO   | YES/NO   | YES/NO   |  |
|                             | <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"/> Yes <input type="radio"/> No | <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"/> Yes <input type="radio"/> No | <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"/> Yes <input type="radio"/> 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 microorganism(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             | <input type="radio"/> Yes <input type="radio"/> No |                                       |
| Rodent            | <input type="radio"/> Yes <input type="radio"/> No |                                       |
| Non-human primate | <input type="radio"/> Yes <input type="radio"/> 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             | <input type="radio"/> Yes <input type="radio"/> No |                       |                   |
| Rodent            | <input type="radio"/> Yes <input type="radio"/> No |                       |                   |
| Non-human primate | <input type="radio"/> Yes <input type="radio"/> No |                       |                   |
| Other (specify)   | <input type="radio"/> Yes <input type="radio"/> No |                       |                   |

2.4 For above named cell types(s) circle HC or CFIA containment level required 1 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 human bone specimens

3.3 Is human source known to be infected with and infectious agent  YES  NO  
If YES, please name infectious agent Call specimens have been tested for infectious agents

3.4 For above named materials circle HC or CFIA containment level required. 1  2  3  
(specimens remain in double-sealed containers as per SOP)

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 rat

6.3 AUS protocol # 2007-003-02

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

**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<sub>50</sub> (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

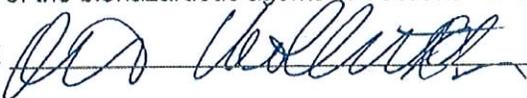
9.4 Has the import permit been sent to OHS? YES  NO   
If yes, Permit # \_\_\_\_\_

**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**

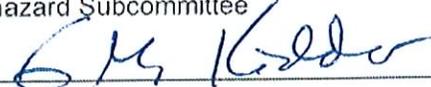
11.1 For the work described in sections 1.0 to 9.0, please circle the highest HC or CFIA Containment Level required. 1  2 3

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: \_\_\_\_\_

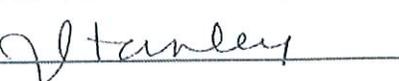
**12.0 Approvals**

UWO Biohazard Subcommittee

Signature  Date 10 July '08

expiry date  
10 July 2010 *gl.*

Safety Officer for Institution where experiments will take place

Signature  Date July 9/08

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

Signature \_\_\_\_\_ Date \_\_\_\_\_

\* Follow Section 3.3 of "Use of Robarts Imaging Suites..."  
(Approved Feb, 2008) - see attached.



## USE OF ROBARTS IMAGING SUITES: BIOSAFETY REQUIREMENTS FOR *IN VIVO* AND *IN VITRO* WORK

Approved: February 13, 2008 (University of Western Ontario Biosafety Committee)

### 1.0 Introduction and Scope:

- Imaging Facilities at Robarts are used for *in vitro* and *in vivo* work by researchers throughout London affiliated with the University of Western Ontario. The objective of this document is to ensure that this research meets the standards set by the latest versions of the Health Canada Laboratory Biosafety Guidelines, the Containment Standards for Veterinary Facilities by the Canadian Food Inspection Agency and, where animals are involved, the Canadian Council for Animal Care (CCAC). This work must also follow the Biosafety Guidelines and Procedures Manual found at: [www.uwo.ca/humanresources/biosafety](http://www.uwo.ca/humanresources/biosafety)

This document applies to the 3T MRI, 9.4T Imaging Suite (MRI), and Preclinical Imaging Suite (MicroCT, Ultrasound, SPECT CT) imaging facilities.

- This document applies to Level 1, Level 2 or Level 2 (plus Level 3 operations) only. Research requiring Level 3 containment must contact the biosafety office at [biosafety@uwo.ca](mailto:biosafety@uwo.ca). Level 2 research involving live non-human primates must follow the Standard Operating Procedures (SOP's) for the Center for the Brain and Mind.

### 1.1 General Safety Precautions for *In vivo* and *In vitro* Imaging

- All personnel operating the imaging equipment (9.4T MRI, 3T MRI, MicroCT, SPECT CT, and Ultrasound) must be trained by Facility Manager or designate. All personnel handling animals must have the required Animal Care and Veterinary Services training.
- All personnel using the facility must be trained and follow the Standard Operating Procedures (SOP's) in place for the facility.
- Supervisors must ensure that people using the facility have the appropriate health and safety training for the work being performed, per the Health and Safety Training found at: [http://www.uwo.ca/humanresources/facultystaff/h\\_and\\_s/training/training\\_idx.htm](http://www.uwo.ca/humanresources/facultystaff/h_and_s/training/training_idx.htm)

- Personnel using this facility must wear the appropriate personal protective equipment. For more information, see the Laboratory Safety Manual, [www.uwo.ca/humanresources](http://www.uwo.ca/humanresources) or contact the Lab Safety Coordinator.
- Disposal of waste, including hazardous chemical waste, biomedical waste and carcasses, must follow the Hazardous Material Management Handbook: [http://www.uwo.ca/humanresources/facultystaff/h\\_and\\_s/enviromental\\_prog/enviromental\\_idx.htm](http://www.uwo.ca/humanresources/facultystaff/h_and_s/enviromental_prog/enviromental_idx.htm)
- Work carried out must meet the requirements of the Biosafety Guidelines and Procedures Manual found at: [www.uwo.ca/humanresources/biosafety](http://www.uwo.ca/humanresources/biosafety)
- Personnel should complete their Position Hazard Communication Form and have the appropriate medical surveillance. For information, please see: <http://www.wph.uwo.ca/newposition.htm>.
- In case of an emergency, such as medical or fire, personnel follow the SOP's in place for the facility accessible on-line or in the Robarts Health and Safety Office.

Preclinical Imaging Suite: SOP 900 – Emergency Procedures  
 9.4T MRI Facility: SOP 300 – Standard Operating Procedure:  
 Emergency Fire Procedures  
 3T MRI Facility: SOP 3T 215, 210, and 205 – Standard Operating  
 Procedures for Emergency Quench, Fire Code Blue

Where there is an emergency involving human and animal wellbeing, human health and safety is the priority.

- The researcher must have an approved, current Biohazardous Agents Registry Form on file with the biosafety office which reflects the research being done. For more information, see: [www.uwo.ca/humanresources/biosafety](http://www.uwo.ca/humanresources/biosafety).
- The biosafety officer(s) in association with the Director, Animal Care and Veterinary Services and the Biohazard Subcommittee determine the containment level required for the work being performed.

## 1.2 Transportation of Animals

### 1.2.1 Transportation of Level 1 Rodents

Level 1 rodents are rodents not exposed to a Level 2 agent via ingestion, inhalation, injection, or absorption and are not known to carry a Level 2 zoonotic agents. Level 1 rodents may be transported to the Robarts imaging facilities and within the Robarts building using standard cages. Level 1 rodents may be transported to the University or within the University buildings in standard cages.

### 1.2.2 Transportation of Level 2 Rodents

Level 2 rodents must be transported in a HEPA-filtered cages or an apparatus. The cages or apparatus must be approved by the Director, ACVS and the Biosafety officer(s) for Robarts. The transportation of Level 2 animals by road, rail, water or air must also follow the appropriate transportation of dangerous goods regulations.

## 2.0 Introduction to Rodent Imaging Research

Rodent projects must be approved by the Animal Use Subcommittee. Rodents are housed in areas approved by Animal Care and Veterinary Services (ACVS) and CCAC. Rodents are transported to the facility in cages on carts. Rodents may undergo procedures, such as injections or anesthesia. Animals are placed inside a suitable coil, bed and/or approved HEPA-filtered apparatus. It is then scanned by a trained operator. Animals may be euthanized if necessary after the imaging is complete.

### 2.1 Imaging Involving Level 1 Rodents

- Level 1 Rodent work involves rodents that have not been exposed to a Level 2 agent via ingestion, inhalation, injection or absorption and that are not known to carry a Level 2 zoonotic agent. An example of a Level 1 rodent is an animal procured from a commercial supplier or rodents injected with a cell line classified by biosafety as Level 1.

#### 2.1.1 Safety Precautions

- Follow the Standard Operating Procedure (SOP's) for the decontamination of samples entering the facility and the clean-up of animal excrement and surface disinfection. Disinfectants must be effective and safe to use on the equipment. The SOP's are available on-line or in the Robarts Health and Safety Office.

Preclinical Imaging Suite: SOP 500 – Cleaning and Decontamination

9.4T MRI Facility: SOP 415 – Cleaning and Disinfection

3T MRI Facility: SOP 400 – Standard Operating Procedure for MRI Decontamination

- Gloves and other personal protective equipment must be changed if they have been in contact with animal wastes.
- Procedures such as injections, anesthetizing and euthanization can be done on the open bench. Scavenging devices must be used on equipment used for anesthetizing or euthanization with a gaseous agent. If a hazardous chemical or radioactive material is involved, this may require the use of a fume hood elsewhere and additional precautions/approvals.

- The animal may be placed in the coil or bed on the open bench.
- In case of a veterinary emergency, procedures such as life-saving measures can be done on the open bench.

## 2.2 Imaging Involving Level 2 Rodents

- Level 2 Rodent work involves animals that have been exposed to a Level 2 agent via ingestion, inhalation, injection or absorption or carry a Level 2 zoonotic agent. Examples of Level 2 pathogens include:
  - ◆ Viral vectors such as adenoviral vectors
  - ◆ Human cell lines such as HEK 293 or non-human primate cell lines such as cos-7 because they carry viral genes capable of cell transformation
  - ◆ Microorganisms such as Salmonella sp. or Pseudomonas sp.
  - ◆ Biological toxins such as pertussis and cholera toxin.
  - ◆ Contact the biosafety officer at [biosafety@uwo.ca](mailto:biosafety@uwo.ca) for the containment level of the project. For more information, please see [www.uwo.ca/humanresources/biosafety](http://www.uwo.ca/humanresources/biosafety)

### 2.2.1 Safety Precautions

For Level 2 projects, there are additional Safety Precautions to those in Section 2.1.1.

- Level 2 agents must be handled in a Class 2 biological safety cabinet. Animals that have been exposed to a Level 2 agent must be kept in an approved HEPA filtered cage or apparatus during the duration of the experiment, including housing, transportation, imaging and veterinary life saving measures.
- Personnel using an approved HEPA filtered cage or apparatus must have a plastic container with them. In case of failure or leakage of the cage or apparatus, the cage or apparatus (with the animal inside) is put in the plastic container. The container can only be opened in a biological safety cabinet.
- Animals exposed to a Level 2 agent must be housed in a certified Level 2 housing facility.

#### 2.2.1.1 Preclinical Imaging Suite and 3T MRI Facilities

Personnel can transport the animals in a HEPA filtered cage to the imaging facility. The cage must be opened in the biological safety cabinet to perform procedures such as injections, anesthesia and veterinary life saving measures. The animal is placed in a HEPA filtered apparatus for imaging in the biological safety cabinet. After imaging, the rodent is transported to a biological safety cabinet in an approved Level 2 housing facility. The apparatus is never opened except in a biological safety cabinet.

The apparatus must be certified by a certified contractor such as HEPA Filters Inc. The apparatus must be approved by the Biosafety Officers for Roberts and Animal Care and Veterinary Services. The apparatus must maintain Level 2 containment, and requires safety features such as HEPA filtration, O-rings, threaded ends.

HEPA cages must be approved by the Biosafety Officers for Roberts and Animal Care and Veterinary Services.

Waste is collected in the biological safety cabinet in bags. The bag is closed in the biological safety cabinet and disposed of by the research personnel. Carcasses are disposed of by research personnel.

#### 2.2.1.2 9.4T MRI Facility

This facility does not contain a biological safety cabinet. Procedures must be done in a biological safety cabinet in an approved Level 2 facility elsewhere.

Animals must be placed in an approved HEPA filtered imaging apparatus in a biological safety cabinet in an approved Level 2 laboratory. Animals are transported to the facility and imaged in this apparatus. The apparatus is never opened except in a biological safety cabinet.

Waste is collected in the biological safety cabinet in bags. The bag is closed in the biological safety cabinet and disposed of by the research personnel. Carcasses are disposed of by research personnel.

The apparatus must be certified by a certified contractor such as HEPA Filters Inc. The apparatus must be approved by the Biosafety Officers for Roberts and Animal Care and Veterinary Services. The apparatus must maintain Level 2 containment, and requires safety features such as HEPA filtration, O-rings, threaded ends.

### 3.0 Introduction to In vitro Research Involving Imaging

Samples are prepared for imaging in a biosafety laboratory. Samples are brought in sealed, leak and shatter proof tubes or other containers to the imaging facility. Samples are put in a coil or a bed and/or HEPA-filtered apparatus for imaging purposes.

#### 3.1 Imaging Involving Fixed Samples

Level 2 or Level 2+3 samples fixed with chemicals such as formalin or comparable agent are no longer considered biohazardous. These samples can be imaged as Level 1 samples. If samples need to be opened, they should be opened in a chemical fume hood.

### 3.2 Imaging Involving Level 1 In Vitro Work

Samples must be transported to the facility in sealed, leak and shatter proof containers. Containers must be wiped off with a disinfectant before they leave the laboratory and per the SOP's for the facility. Work with these samples can be done on the open bench, providing that no hazardous chemicals are involved. If hazardous chemicals or radioactive materials are involved, work must be done in a fume hood elsewhere and additional precautions / approvals are required.

### 3.3 Imaging Involving Level 2 In vitro Work

For Level 2 projects, there are additional Safety Precautions to those in 3.1. Samples must be worked with using a biological safety cabinet.

#### 3.3.1 Preclinical Imaging Suite and 3T MRI Facilities

If required, samples can be opened under the biological safety cabinet provided.

#### 3.3.2 9.4T MRI Facility

There is no biological safety cabinet in this facility. Samples must be prepared in a biological safety cabinet in an approved Level 2 laboratory elsewhere. Sealed, leak and shatter proof containers are not to be opened in the facility. The sample is kept closed during transportation and imaging of the samples.

### 4.0 Imaging Involving Level 2 plus Level 3 Work

The researcher must have an approved, current Biohazardous Agents Registry Form on file with the biosafety office which reflects the research being done. For more information, see: [www.uwo.ca/humanresources/biosafety](http://www.uwo.ca/humanresources/biosafety)

For Level 2 plus Level 3 projects there are additional Safety Precautions. All work must be carried out in a biological safety cabinet.

#### 4.1 Imaging

- Use portable autoclave to decontaminate waste prior to leaving the imaging facility. Follow the “SOP for the Sanyo Portable Autoclave”.
- Injections must be done in the approved Level 2 plus 3 laboratory or the Level 3 facility on DSB, 6<sup>th</sup> floor.
- Animals transported on a cart to or within Robarts for imaging must be in a HEPA filtered cage unit approved by biosafety and ACVS.
- The cages can be removed from the transport cart and placed in a biological safety cabinet. Animals must be placed in an approved HEPA filtered imaging apparatus (see section 2.2.1.1) in a biological safety cabinet in an approved Level 2 plus 3 laboratory. Animals are transported to the facility and imaged in this apparatus. The apparatus is never opened except in a biological safety cabinet.
- After scanning all reusable material (i.e. forceps), they must be decontaminated in a Wescodyne solution in a biological safety cabinet. The Wescodyne working solution has: 40% H<sub>2</sub>O, 40% ethanol and 20% Wescodyne. (It can be prepared in advance)
- Submerge all the reusable instruments (surgical) in the labelled Wescodyne solution for 2 hours.
- Rinse the instruments after 2 hours with H<sub>2</sub>O and let dry.
- After drying, pack in autoclave bags and autoclave in the portable autoclave. (This is done to ensure successful sterilization)
- The procedures for disinfection of contaminated animal cages and bedding must be completed. Bedding must be emptied into a biohazard bag inside of the biosafety cabinet. The bedding must be then double bagged and sealed inside a biological safety cabinet. The bag must be wiped with a disinfectant before it is removed from the biological safety cabinet.
- Inside the hood, to the empty cage add Wescodyne solution and swirl to ensure contact of all surfaces. Wipe the cage lid with Wescodyne as well and ensure contact for 2 hours (either leave the cage in a dunk tank for 2 hours or put the wet cage into an autoclave bag and leave in the hood for 2 hours). Drain the Wescodyne and return the cages and lids for washing and packing to be autoclaved. Follow the procedures for the facility where the cages came from. (ACVS or Robarts barrier facility)
- All sharps must be disposed of in a sharps container within the biosafety cabinet. The container must be wiped on the outside with the Wescodyne solution. The containers are then sent to the incinerator.
- All waste must be labelled appropriately before it is taken for disposal.
- After the scan the rodent/animal must be returned to the biological safety cabinet before it is removed from the HEPA filtered apparatus and then it can be returned to its cage.
- Disposable personal protective equipment, such as gloves, must be put in an autoclaveable biohazard bag leaving the room.
- Wescodyne solution can be treated as hazardous waste after use per the Hazardous Waste Management Handbook:  
[http://www.uwo.ca/humanresources/docandform/docs/ohs1/manuals/hazardous\\_handbook.pdf](http://www.uwo.ca/humanresources/docandform/docs/ohs1/manuals/hazardous_handbook.pdf).