



**Western**  
UNIVERSITY • CANADA

**BIOSAFETY GUIDELINES AND  
PROCEDURES MANUAL**  
For Containment Level 1 & 2 Laboratories



**Occupational Health and Safety, 9<sup>th</sup> Edition**  
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**WESTERN UNIVERSITY**  
**BIOSAFETY POLICIES AND PROCEDURES MANUAL**  
For Containment Level 1 & 2 Laboratories

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## **1. BIOSAFETY PROGRAM FRAMEWORK**

### **1.1 WESTERN UNIVERSITY HEALTH AND SAFETY POLICY**

Under the provisions of the Ontario Occupational Health and Safety Act the University, as an employer, is responsible for ensuring compliance with the Act and regulations and for taking every precaution reasonable in the circumstances for the protection of workers.

In compliance with the requirements of the Ontario Occupational Health and Safety Act, the University has approved the Western University Health, Safety and Wellness policy. Pursuant to this policy, the University has established this formal Biosafety Program to identify, reduce and monitor the risks associated with potentially hazardous biological agents.

This Manual describes the requirements and procedures established by the University for work involving potentially hazardous biological agents. It is based on the Public Health Agency of Canada (PHAC) "Canadian Biosafety Standard for Facilities Handling or Storing Human and Terrestrial Animal Pathogens and Toxins, Second Edition). All work conducted by University members with potentially hazardous biological agents on University premises or under the control of the University is to be performed in accordance with the requirements of this manual.

Questions regarding application or interpretation of this manual should be directed to the Western Biosafety Officer, Occupational Health and Safety, (519) 661-2111 ext. 88730.

#### **1.1.1 HEALTH, SAFETY and WELLNESS**

*"Supporting a healthy and safe community for teaching and research"*

The University strives to foster the development of a safety consciousness in all members of the University community for the purpose of minimizing the risk of injury to persons or the damage to property or facilities. As safety standards change, the University is committed to keeping abreast of these changes, to communicating these standards within the campus, and to ensuring compliance on an ongoing basis.

The University will comply with all applicable Federal, Provincial and Municipal legislation with respect to health and safety. Legislated standards in health and safety are accepted by the University as minimum standards, and the University reserves the right to establish and enforce more stringent standards as may be considered appropriate. Such policies are considered as binding upon all students, staff and faculty.

## 1.2 REGULATION OF BIOLOGICAL AGENTS WITHIN THE UNIVERSITY

Western University (Western) has five Health and Safety Committees, as shown by Figure 1.2 below.

Figure 1.1: Western Health and Safety Committees



The Chair of the University Health and Safety Committee is the Vice President, Administration. Several other committees report to this committee, including the Biosafety Committee and the Biohazards Subcommittee detailed in Sections 1.3, 1.4 and 1.5 below.

### 1.3 THE BIOSAFETY COMMITTEE

Western's Biosafety Officer is responsible for administering the Biosafety program on a day-to-day basis and for providing technical advice on safety procedures, equipment and relevant regulations.

#### Mandate

The Western University Biosafety Committee (WUBC) is a Sub-committee of the University Health and Safety Committee (UHSC) and makes recommendations to the UHSC on all matters pertaining to biosafety at Western. The WUBC is also mandated to fulfil the responsibilities of a Research Institution Biological Safety Committee as described in the most current version of the Public Health Agency of Canada Biosafety Standard. These responsibilities include verifying that all work with biohazardous agents carried out at Western University is in accordance with the safety practices as stated in the Guidelines.

#### Terms of Reference

1. To make recommendations to the University Health and Safety Committee on actions and/or policies related to biosafety at Western University.
2. To monitor and promote compliance with the established policies and procedures as set out in the most current version of the Public Health Agency of Canada's "Canadian Biosafety Standard", and other governmental and University guidelines related to biosafety.

To develop and recommend policies and procedures to meet or exceed the requirements for each containment level as stated in most current version of

the Public Health Agency of Canada's "Canadian Biosafety Standard" .

3. To approve protocols involving the use of potentially biohazardous agents including genetically modified organisms and animals potentially carrying infectious zoonotic agents and to confirm the appropriate containment level for the work; to verify that the appropriate facilities and procedures are in use and to ensure that appropriate procedures for the use, storage and disposal of the named agents are followed.
4. To approve applications for greenhouse or field trials of genetically modified organisms.
5. To review the procurement and use of all biohazards, including animals, human source tissues, blood and body fluids for research and undergraduate teaching, and to recommend and monitor the appropriate safety precautions.
6. In collaboration with the University Biosafety Officer, to review, recommend and act as an expert resource for biosafety education and training programs at Western.
7. To recommend to the University that appropriate institutional occupational health programs be put in place as necessary to achieve the outcomes stated above.
8. To establish Sub-committees to carry out specific tasks as mandated by the UBSC.

### **Membership**

1. Voting: Special Advisor (Research) or designate, Chair  
Director, University Workplace Health (ex-officio)  
Director, Animal Care and Veterinary Services (ex-officio)  
Director, Occupational Health and Safety (ex-officio)  
Schulich School of Medicine & Dentistry (2 with at least one from the Department of Microbiology and Immunology)\*  
Faculty of Science (2 with at least one with plant and fungal expertise)\*  
Faculty of Health Sciences (1)\*  
Faculty of Engineering (1)\*  
Physical Plant Facilities Engineering (1)\*  
Robarts Research Institute (1)\*  
Research Technician (1)\*  
Campus Police Representative (1)\*

\*Appointed by Dean of Faculty, Director/Chair of Department, or Scientific Director of Centre/Institute

2. Non-Voting: University Biosafety Officer  
Society of Graduate Students member (1)\*

### **Term**

A maximum appointment of 3 years - renewable.

### **Frequency of Meetings, Quorum**

The Biosafety Committee will meet at least three times a year. For voting purposes 5 members must be present.

## **1.4 SUB-COMMITTEE FOR THE REVIEW OF BIOHAZARDOUS AGENT AND GENE THERAPY PROTOCOLS:**

### **Mandate**

The Biohazards Sub-committee is a Sub-committee of the University Biosafety Committee and makes recommendations to the WUBC on all matters related to the use of potentially biohazardous materials at Western. The Sub-committee reviews all protocols involving work with potentially biohazardous materials to ensure that such use is in conformity with the most current Canadian Biosafety Standard published by the Public Health Agency of Canada, The Canadian Food Inspection Agency standards, and all relevant governmental and University guidelines.

### **Terms of Reference**

To review and approve protocols involving the use of potentially biohazardous agents including genetically modified organisms and animals potentially carrying infectious zoonotic agents at all biosafety containment levels and to ensure that appropriate procedures are communicated to the research users for the use, storage and disposal of the named agents and that these procedures are followed.

1. To review and approve all Containment Level 3 protocols.
2. To ensure that the Containment Level 3 Facility is operated in accordance with the most current Canadian Biosafety Standard published by the Public Health Agency of Canada, and the Canadian Food Inspection Agency standards.
3. To review the Biological Agents Permit Application annually to ensure that the form meets the needs of the Biosafety Program.
4. To generate an Annual Report to be forwarded to the UBSC by May 1 of each year.

## Membership

1. Voting: Director, Department of Animal Care and Veterinary Services (ex-officio)  
Three other members elected by the UBSC at least one of whole is a virologist and one a bacteriologist  
Director, Workplace Health (ex-officio)  
Special Advisor (Research) or designate, as Chair
  
2. Non-voting: University Biosafety Officer

### Frequency of Meetings, Quorum:

This Sub-committee meets as needed. For voting purposes, 3 voting members must be present.

## 1.5 RESPONSIBILITIES

The Ontario Occupational Health and Safety Act and Regulations outline the rights and responsibilities of all workplace parties. All faculty, staff, and students at Western University are required to follow these general acts and regulations.

For more information, please see the Ontario Ministry of Labour website at: [www.labour.gov.on.ca/english/](http://www.labour.gov.on.ca/english/)

### The Biological Safety Officer (BSO) has the following functions:

1. verifying the accuracy and completeness of licence applications;
2. communicating with the PHAC on behalf of the licence holder;
3. promoting and monitoring compliance, including but not limited to:
  - arranging for and documenting training related to biosafety and biosecurity policies, standards, and practices;
  - notifying the PHAC of any inadvertent possession of human pathogens and toxins or of any Security Sensitive Biological Agents (SSBAs) not received as expected;
  - conducting periodic internal inspections and biosafety audits and reporting findings to the license holder;
  - informing the license holder of any non-compliance by a person conducting activities under the license that is not resolved after that person has been made aware of it;
4. assisting in the development and maintenance of the Biosafety Manual and standard operating procedures (SOPs) related to biosafety and biosecurity; and
5. assisting with internal investigations of incidents.

The BSO may require any person who conducts controlled activities under the licence to provide them with any records that are necessary to assist them in carrying out their functions.



### 1.5.1 RESPONSIBILITIES OF PRINCIPAL INVESTIGATORS WORKING WITH POTENTIALLY HAZARDOUS BIOLOGICAL MATERIALS

The primary responsibility for the safety of staff, students and the public lies with the Principal Investigator in charge of the research. Principal Investigators must be familiar with, follow, and ensure that all individuals working within their laboratories follow the procedures outlined in this manual. In particular,

Principal Investigators are responsible for:

1. Obtaining biosafety permits where required
2. Ensuring that all conditions of the permit are followed
3. Ensuring that the appropriate containment cabinets are functioning properly by having them tested according to the Procedures for the Effective Use of Biological Safety Cabinets. See website:  
[http://www.uwo.ca/hr/form\\_doc/health\\_safety/doc/procedures/biological\\_safety\\_cabinets.pdf](http://www.uwo.ca/hr/form_doc/health_safety/doc/procedures/biological_safety_cabinets.pdf)
4. Ensuring that all persons working under their control have had appropriate training in working safely with potentially hazardous biological materials
5. Providing appropriate personal protective equipment and standard operating procedures
6. Ensuring that all persons working under their control follow applicable University safety manuals, procedures and policies
7. Ensuring that Hazard Communication Forms are up-to-date for all persons working under their control
8. Ensuring agents listed on the permit are up-to-date. Changes must be approved through the modification process found at:  
[http://www.uwo.ca/hr/form\\_doc/health\\_safety/doc/procedures/bio\\_modification\\_info.pdf](http://www.uwo.ca/hr/form_doc/health_safety/doc/procedures/bio_modification_info.pdf)

#### **Notification must be provided to the BSO without delay in the following circumstances:**

- when a person has reason to believe that a human pathogen or toxin has been released inadvertently from a facility;
- when a human pathogen or toxin that a person is not authorized to possess is inadvertently produced or otherwise comes into their possession;
- when an incident involving a human pathogen or toxin has caused, or may have caused, disease in an individual;
- when there is reason to believe that a human pathogen or toxin has been stolen or is otherwise missing;
- where an SSBA is not received within 24 hours of the date and time when it was expected to be received; **and**
- **where the holder of an HPTA security clearance is convicted of a criminal offence**, notification must be provided to the BSO in the following circumstances:
  - before making any of the following changes, if they could affect biocontainment, where controlled activities with Risk Group 3 or 4 human pathogens or SSBA toxins are conducted:
    - changes to the physical structure of their facility;

- changes to any equipment; or
- changes to their SOPs; and
- within a reasonable time of a licence holder making a name change.

**Prompt notification to the BSO is essential as the BSO is required to notify the PHAC on behalf of the Licence Holder.**

### **1.5.2 RESPONSIBILITIES OF PERSONS WORKING WITH POTENTIALLY HAZARDOUS BIOLOGICAL MATERIALS**

1. Follow all safety manuals and standard operating procedures
2. Wear personal protective equipment
3. Participate in medical surveillance programs as deemed necessary by Workplace Health
4. Report hazards or potentially hazardous incidents to Supervisor
5. When required, seek information from their supervisor or other resources

### **1.5.3 CRIMINAL CODE OF CANADA**

In 2004, workplace safety legislation was passed which establishes, for the first time in Canadian history, a duty to ensure workplace health and safety under the Criminal Code. These changes apply to all Canadian workplaces including the administrative, teaching and research areas at Western.

In 2004, the Criminal Code of Canada imposed a legal duty which applies to everyone who undertakes, or has the authority, to direct how work is performed.

If you are diligently following applicable Occupational Health and Safety (OHS) regulations and best practices in your workplace, and are monitoring compliance, then these legislative changes will serve to reinforce the importance of your efforts.

## **1.6 BIOSAFETY PROGRAM COMPONENTS**

The Program includes the following components:

1. Biosafety Database
2. Biosafety Guidelines and Procedures Manual
3. Containment Level 2 and 3 Inspections & Permits
4. Animal Use Sub-committee Safety Forms & Biological Agents Permit Applications
5. Purchasing of Biohazardous Material
6. Biohazardous Waste Management
7. Medical Surveillance
8. Training
9. Biosecurity

## **1.7 LICENCING AND BIOSAFETY PERMITS**

### **1.7.1 REQUIREMENT FOR WESTERN UNIVERSITY BIOLOGICAL AGENTS PERMIT APPLICATION AND OTHER PERMITS**

A Western University Biological Agents Permit Application is required for all laboratory activities (research and teaching) which involve the use or manipulation of potentially hazardous biological agents, and materials containing such agents (including viruses, bacteria, fungi, parasites, recombinant DNA, prions, other microorganisms/genetic systems, and human and animal tissues, cells, blood and body fluids), and which are:

- (i) Supervised or conducted by employees or members of the University, or
- (ii) Supported by funds provided by or through the University.

All such activities are to be conducted and performed in accordance with the Western University Biosafety Policies and Procedures Manual and any relevant guidelines or legislation.

All activities involving potentially hazardous biological agents and meeting any of the above criteria must be identified on the Biological Agents Registry Form. The release of grants and supporting funds by the University is dependent on a completed signed University Biological Agents Permit Application.

Biological Agents Permit Applications are available on the Western Human Resources website at: <http://www.uwo.ca/hr/safety/topics/biosafety/index.html>

After completion, the form is sent to the Western Biosafety Officer in Occupational Health and Safety. The form is reviewed by the Biohazards Sub-committee and approved by the Chair of the Biohazards Sub-committee and the Biosafety Officer.

The submission of an application for a Western University Biological Agents Permit Application implies willingness to allow the Western Biosafety Officer to visit the laboratory sites on campus used by the Principal Investigator in order to determine compliance with the Western University Biosafety Policies and Procedures Manual.

For research requiring containment levels two or three, Western's Biosafety Officer will inspect the worksite to ensure that it meets the operational and physical requirements as per the current Public Health Agency of Canada Biosafety Standard. Western's Biosafety Officer can issue a Biosafety Permit following the inspection.

After this period, the Principal Investigator must submit a new application form every three years even if the activities involving biological agents have not been altered or modified since the previous submission. If the activities involving biological agents have been altered or modified since the previous submission, a biohazard modification form must be completed. This form can be obtained from Western's Biosafety Officer. For more information, see:

[http://www.uwo.ca/hr/form\\_doc/health\\_safety/doc/procedures/bio\\_modification\\_info.pdf](http://www.uwo.ca/hr/form_doc/health_safety/doc/procedures/bio_modification_info.pdf)

## **2.0 LEGISLATION, GUIDELINES AND STANDARDS**

Activities involving the use of biological agents and laboratory animals, the production and disposal of waste, and the use of certain equipment are governed by various legislation, guidelines and standards. Adherence to the requirements of this Manual will ensure that work is performed safely and in compliance with the requirements of external agencies and regulatory bodies.

## **2.1 IMPORTATION, USE AND DISTRIBUTION OF BIOLOGICAL AGENTS**

### **2.1.1 THE HUMAN PATHOGENS AND TOXINS ACT**

The Human Pathogens & Toxins Act was a new law passed June 23, 2009. According to the Public Health Agency of Canada (PHAC) website, this law “contains prohibitions and requirements relating to a full range of laboratory activities. A new program and regulatory framework is based on requirements of the Human Pathogens and Toxins Regulations (HPTR) and the Canadian Biosafety Standard”.

For more information on the Act, please see:

<http://www.phac-aspc.gc.ca/lab-bio/regul/hpta-lapht-eng.php>

For more information on the Regulations, please see:

<http://gazette.gc.ca/rp-pr/p2/2015/2015-03-11/html/sor-dors44-eng.php>

Western University holds licenses for all on-campus activities involving biohazardous biological agents. To maintain this license, Western University Biosafety Officer must report all prescribed biological agents and toxins used on campus to the Public Health Agency of Canada on an annual basis. Thus it is mandatory that all Principal Investigators inform the Biosafety Officer when their research changes.

### **2.1.2 Canadian Food Inspection Agency**

The Canadian Food Inspection Agency (CFIA) works with Agency scientists and technical experts to establish the biocontainment levels, procedures, and protocols that are needed to work safely with animal and zoonotic pathogens, chemical hazards, and plant pests of quarantine significance, and to protect laboratory staff, the Canadian public, and the environment.

In accordance with the [Health of Animals Act](#) and its regulations, the CFIA is responsible for issuing permits for non-indigenous animal pathogens, emerging animal disease pathogens, aquatic and plant pathogens, as well as animals, animal products and by-products, tissue, sera, and blood that are infected with animal pathogens.

The CFIA also provides information on the containment standards for [Facilities Handling Aquatic Animal Pathogens](#), [Foreign Animal Disease Diagnostic Laboratories](#), and [Facilities Handling Plant Pests](#).

**The CFIA also provides a framework for the oversight of [Laboratory Management](#) for facilities conducting testing under the CFIA's mandate.**

As of April 1, 2013, some programs at the CFIA's Office of Biohazard Containment and Safety were transferred to the [Public Health Agency of Canada](#) (PHAC) (refer to Section 2.2).

The CFIA and PHAC have joined forces to update and synchronize three existing Canadian biosafety standards and guidelines by creating the [Canadian Biosafety Standards and Guidelines \(CBSG\)](#). The CBSG has replaced the *Laboratory Biosafety Guidelines*, the *Containment Standards for Veterinary Facilities* and *Containment Standards for Laboratories, Animal Facilities and Post Mortem Rooms Handling Prion Disease Agents*.

### **2.1.3 Public Health Agency of Canada**

To strengthen its ability to promote and protect the health and safety of Canadians, in 2004 the Government of Canada established the [Public Health Agency of Canada](#) (PHAC). PHAC was confirmed as a legal entity in 2006 by the Public Health Agency of Canada Act. The creation of PHAC marked the beginning of a new approach to federal leadership and collaboration with provinces and territories on public health. It responds to a consensus from the provinces, public health experts and concerned citizens on the need for federal leadership on public health to be consolidated in a public agency. PHAC serves as the national authority for the biosafety and biosecurity of human pathogens and toxins in Canada. The Agency's Center for Biosecurity administers and enforces the Human Pathogens Importation Regulations (HPIR) and the [Human Pathogens and Toxins Act](#) (HPTA). The HPTA will be supported by the Human Pathogens and Toxins Regulations (HPTR) when they come into force on December 31, 2015, at which time the HPIR will be repealed.

PHAC is responsible for issuing import permits for human and terrestrial animal pathogens, with the exception of non-indigenous animal pathogens and pathogens causing emerging animal diseases. PHAC is also responsible for issuing Containment Level 2 compliance letters and the certification of High Biocontainment laboratories. PHAC provides tools to individuals who design, operate or work in laboratories in which human pathogens are manipulated for diagnostic, research or development purposes. These laboratories may be located in universities, hospitals, government departments or industrial settings. Tools provided by PHAC include [Pathogen Safety Data Sheets](#) (PSDS) for infectious substances, which provide detailed descriptions of the hazardous properties of specific human pathogens and toxins.

Also available from PHAC are the [Canadian Biosafety Standards and Guidelines](#) (CBSG). Developed jointly by PHAC and CFIA, the CBSG is a technical document which provides details on the physical containment and operational practice requirements for facilities possessing, handling, storing, or using human and terrestrial animal pathogens or toxins. Following the standards and guidelines outlined in this document will help protect personnel from exposure to infectious materials and toxins, as well as aid in the prevention of the inadvertent release of these materials. The CBSG is also used by PHAC and CFIA to verify regulatory compliance of facilities where infectious materials or toxins are handled and stored, as well as for the certification or recertification of containment zones.

## 2.2 EXPORT REQUIREMENTS FOR BIOLOGICAL AGENTS

Many pathogens and associated equipment that are destined for export from Canada require permits. Canada is a signatory to the 1972 Biological and Toxin Weapons Convention. This international Convention stresses the goal of non-proliferation of biological and toxin weapons through the prohibition of the development, production, stockpiling or acquisition of microbiological (biological) and toxin weapons and their destruction. The Department of Foreign Affairs and International Trade Canada currently controls certain toxicological and biological agents as well as their related equipment, components, materials and technology, under item 2007 of the Export Control List of this international Convention. For assistance or advice, contact the Department of Foreign Affairs and International Trade Canada, Export Control Division, telephone 1-800-267-8376 or contact their website at <http://www.international.gc.ca/controls-controles/index.aspx?view=d> (1).

## 2.3 TRANSPORTATION OF HUMAN SPECIMENS

The careful handling, transport and shipment of diagnostic specimens and infectious agents are absolutely essential in Canada. Transportation methods must minimize risks to employees of the carrier, the public and the staff of the receiving laboratory. Hazards are compounded by improper packaging. A broken specimen container may lead to contamination of both laboratory and non-laboratory personnel. An improperly labeled package may be opened inadvertently by untrained or unprotected staff. All shipments must follow the Western Transportation of Dangerous Goods Policy that can be found at:

<http://www.uwo.ca/hr/safety/topics/tdg.html>

The transportation of infectious substances, including blood, is an essential part of routine laboratory procedures in both research and diagnostic settings. Samples must be transported by road and/or air to assist researchers collaborating with other researchers at removed locations, or to carry out primary diagnostic tests on samples obtained from ill patients. Although there has never been a reported case of illness associated with a transportation accident involving an infectious substance, transportation accidents involving infectious substances have occurred. Therefore, it is important that infectious substances be packaged and transported according to tested and approved methods.

The transportation of infectious substances within Canada is regulated by the *Transportation of Dangerous Goods Regulations* (SOR/2001-286), administered by Transport Canada. Transport Canada defines the labeling, packaging and documentation requirements necessary for shipping infectious substances, including diagnostic specimens, within Canada. Their regulation also requires that any individual transporting an infectious substance be trained in the transportation of dangerous goods (infectious substances). In addition, shippers of risk group 4



materials are required to have an emergency response assistance plan to respond to any shipping emergency occurring anywhere in Canada (1).

The air transportation of infectious substances internationally is regulated by the International Civil Aviation Organization (ICAO). As the majority of carriers (both passenger and courier/cargo) around the world are members of this organization, anyone shipping infectious substances internationally is likely subject to ICAO regulations. The ICAO regulations define the labeling, packaging and documentation requirements necessary for international shipping of infectious substances by air. It also requires that any individual transporting an infectious substance be trained in the transportation of dangerous goods (infectious substances). The ICAO requirements are based upon the United Nations Recommendations on the Transportation of Dangerous Goods (1). Shipping infectious substances by air also falls under the *Dangerous Goods Regulations* (DGR) of the International Air Transport Association (IATA). These regulations set out all the ICAO mandates and the airline industry's universal rules on how to safely package and transport infectious substances (1).

Very specific packaging and documentation requirements must be met before such materials may be shipped from Western University. Shippers must be trained and certified. Contact OHS if biohazardous agents are to be shipped from Western University.

### **2.3.1 TRANSPORTATION OF BIOHAZARDOUS MATERIALS WITHIN FACILITIES**

Transportation of biohazardous materials within Western facilities must be done in closed, leak proof containers. A trolley or cart should be used whenever possible.

An acceptable example is the Bio-safe carrier with handle available from Thermo Scientific (catalog numbers 56609-112 and handle 56609-111) as shown here.

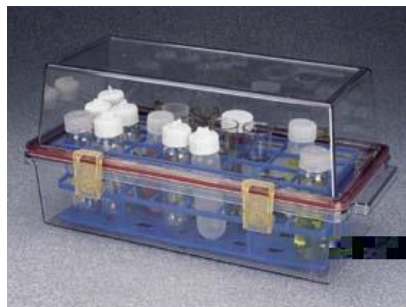


Figure 1.2: Bio-Safe Carrier

## **2.4 LABORATORY ANIMALS**

All aspects of the proposed use of animals in research and the operational procedures for the care and maintenance of animals must satisfy the Guidelines for the Care and Use of Experimental Animals of the Canadian Council on Animal Care,

the Animal Use Sub-committee as well as this manual. This ensures that not only are laboratory personnel and the environment protected, but also that every care is taken to avoid causing the animals unnecessary pain or suffering and to provide the animals with the highest quality care. Under the Ontario Animals for Research Act and its Regulations, it is a requirement that all Principal Investigators who intend to conduct research, testing or teaching projects at Western University that involve the use of animals, must obtain the approval of the Animal Use Sub-committee before starting the project. To obtain such approval, the Principal Investigator must submit the Animal Use Protocol Form that is available from Animal Care and Veterinary Services. The Animal Use Protocol Form contains a section which addresses occupational health and safety issues (including biosafety) and is reviewed by the Safety Officer(s).

The completed protocol form must be signed by the Principal Investigator and is then submitted to the Animal Use Sub-committee at the University for review, approval and signature. Please refer to the practices and procedures in Section 3.2 Working with Laboratory Animals.

## **2.5 WASTE MANAGEMENT**

Western University is a large diverse workplace that generates many kinds of hazardous and nonhazardous waste. The handling, packaging, transport and disposal of waste in Ontario are governed by municipal, provincial and federal government legislation. To enable compliance with these regulations, the University has developed programs, procedures and internal services focussed on specific waste categories. Occupational Health and Safety has prepared a Hazardous Waste Management Handbook containing procedures for the packaging, labelling and disposal of biological, chemical, radioactive, sharp, and other hazardous waste at Western University. The electronic version of this document is available on the Western Human Resources website at:

[http://www.uwo.ca/hr/form\\_doc/health\\_safety/doc/manuals/hazardous\\_handbook.pdf](http://www.uwo.ca/hr/form_doc/health_safety/doc/manuals/hazardous_handbook.pdf)

Laboratory waste contaminated with or containing biological agents must be autoclaved, incinerated or disinfected to inactivate the biological agents prior to disposal. Where on-site functioning autoclaves are not available and the conventional use of chemical disinfectants for the inactivation of hazardous biological agents in laboratory waste is not practicable or not efficacious, other waste handling and disposal methods must be considered.

In the case of autoclaving, representative autoclave loads must be validated using biological indicators as per "[Autoclave Cycle Verification Testing Using Biological Indicator Ampoules](#)" prior to the disposal of waste.

For further information about our Environmental or Waste Management programs please contact Occupational Health and Safety.

## **2.6 AUTOCLAVES/ STEAM STERILIZERS**

### **2.6.1 AUTOCLAVE INSTALLATION AND TESTING**



Every autoclave must be inspected at the time of installation and should have a valid permit from TSSA (Technical Safety and Standards Authority) of Ontario. After the initial installation, this equipment is to be inspected annually. The scope of inspection will include a visual inspection, a review of the conditions of operation and the protective devices such as the pressure relief valves, temperature controls (if any), steam quality control, and the measures being taken by the user for its safe and efficient operation as required by the Boilers and Pressure Vessels Act of Ontario.

Upon satisfactory completion of the inspection, a permit of inspection will be issued which will authorize operation of the equipment. Western Power Plant maintains a list indicating the locations of autoclaves. Annual inspections are performed automatically, according to this list. If you have received a new autoclave, have moved an autoclave or are using one that has not been inspected during the last 12 months, please notify Western Power Plant. You must provide the information necessary to have this equipment added to the equipment list so that the required inspections are scheduled and performed in the future.

## **2.6.2 AUTOCLAVE USE**

Western Autoclaves must be operated as per “[Standard Operating Procedures for Autoclaving](#)”. This procedure must be posted near the autoclave.

Prior to using any autoclave, personnel must be trained on its safe use. For training and autoclave information, contact your Supervisor or the Departmental contact listed on the “Standard Operating Procedures for Autoclaving” sign posted nearby.

## **2.7 BIOLOGICAL SAFETY CABINETS: ASSESSMENT AND TESTING**

Biological safety cabinets, when properly used in research and teaching activities involving the manipulation of hazardous biological agents, are effective in containing and controlling particulates and aerosols. Biological safety cabinets complement good laboratory practices and procedures. Biological safety cabinets used in laboratory activities at Western University must be inspected, tested and approved for use annually (unless otherwise noted) by trained service personnel to ensure that the cabinet is functioning as intended by the manufacturer.

For information on biological safety cabinets, please see the Procedures for the Effective Use of Biological Safety Cabinets available from:

[http://www.uwo.ca/hr/form\\_doc/health\\_safety/doc/procedures/biological\\_safety\\_cabinets.pdf](http://www.uwo.ca/hr/form_doc/health_safety/doc/procedures/biological_safety_cabinets.pdf)

## **2.8 WORKPLACE HAZARDOUS MATERIALS INFORMATION SYSTEM (WHMIS)**

Biohazardous agents are controlled by Workplace Hazardous Materials Information System (WHMIS) Regulations and are classified as Class D, Division 3, “Biohazardous Infectious Material”. These materials are the organisms, or toxins they produce, that can cause disease in people or animals. Examples include

bacteria, viruses, fungi and parasites. Since these organisms can live in body tissues or fluids, tissues and fluids are also treated as biohazardous infectious material (2).

For more information on WHMIS, please visit the Canadian Centre for Occupational Health and Safety website: [www.ccohs.ca](http://www.ccohs.ca)

### **2.8.1 WHMIS TRAINING**

WHMIS regulations require that all people working with or likely to be exposed to biohazards must be educated and trained on biohazards. Workers must be educated in general information such as the classes and symbols of controlled products. Training refers to instruction in site- specific information such as standard operating procedures and emergency procedures. Both education and training are important parts of understanding the risks that may be present at your workplace (2).

For on-line WHMIS education please visit the web at:

<http://uwo.ca/hr/learning/required/index.html>

### **2.8.2 MATERIAL SAFETY DATA SHEETS**

WHMIS regulations require that workers have access to information on all hazards in the workplace, including biohazards.

Material Safety Data Sheets (MSDS) for infectious microorganisms (biological agents) have been prepared by the Office of Laboratory Security, Public Health Agency of Canada. These are available on the Internet via a hyperlink on the Western Human Resources website at:

<http://www.uwo.ca/hr/safety/topics/biosafety/external.html>

These MSDS contain health hazard information, recommended precautions, safe handling methods, decontamination methods and other information that is relevant to the laboratory setting.

In the absence of a Public Health Agency of Canada MSDS, all attempts to get health and safety information on a biohazard must be made. This includes contacting the supplier, distributor, or other source of the biohazard.

For example some suppliers have information sheets available by phone, fax or internet. The American Tissue Culture Collection ([www.atcc.org](http://www.atcc.org)) has information sheets with the organism, source and biosafety of level of the biohazard.

## **2.9 BIOSAFETY LABORATORY VISITORS**

All biosafety laboratory visitors must dress appropriately, as required by the Laboratory Supervisor. They must wear the personal protective equipment required to be worn in the laboratory.

All visitors must be accompanied by the Laboratory Supervisor or designate who is responsible for them in the case of an emergency.

All lab visitors must follow the rules and procedures of the laboratory.

Anyone not complying with the above will not be allowed entry into the lab or will be asked to leave the lab.

### **2.9.1 SERVICE ANIMALS**

If a person wishes to enter a laboratory accompanied by their service animal, the laboratory supervisor is to contact Occupational Health and Safety. Personnel in the Health and Safety Office will review the lab environment and determine if there is a risk for the animal or the research.

## **3.0 SAFETY PRACTICES AND PROCEDURES**

Individuals who work in a laboratory that handles infectious substances are at risk of exposure to the substances they handle. According to the Public Health Agency of Canada, laboratory-acquired infections (LAIs) are not uncommon — over 5,000 cases and 190 deaths had been reported. These figures are believed to be a significant underestimate because of underreporting. In addition, only about 20% of infections can be attributed to any known, single exposure event (1).

There are a number of ways in which infectious substances can enter the body and cause infection. These include ingestion, inhalation, or contact with mucous membranes, including conjunctivae (transfer of microorganisms to the eyes by contaminated hands), or with nonintact skin.

The types of events that can lead to an infection include the following: exposure to infectious aerosols; spills and splashes; accidental needle stick injuries; cuts from sharp objects and broken glass; bites and scratches from animals or ectoparasites; oral pipetting (which is prohibited); centrifuge accidents and; secondary spread of infectious materials to non-laboratory areas (1). In educational settings, needle stick injuries and animal bites are the most common types of accidents involving potential biohazard exposure (3).

Exposure to aerosols may be the greatest biohazard facing laboratory workers. Aerosols can present a risk in terms of inhalation, ingestion, mucous membrane contact, etc. Operational practices and techniques must be used to minimize the creation of aerosols associated with common laboratory procedures (1).

These operational practices and techniques are set by the Biosafety Laboratory Guidelines established by the Public Health Agency of Canada.

### **3.1 GENERAL LABORATORY SAFETY PRACTICES**

The following general practices are required by the Public Health Agency of Canada (1) for all laboratories handling infectious substances.

1. A documented procedural (safety) manual must be available for all staff, and its requirements followed; it must be reviewed and updated regularly.
2. Personnel must receive training on the potential hazards associated with the work involved and the necessary precautions to prevent exposure to infectious agents and release of contained material; personnel must show evidence that they understood the training provided; training must be documented and signed by both the employee and supervisor; retraining programs should also be implemented.
3. Eating, drinking, smoking, storing of food, personal belongings, or utensils, applying cosmetics, and inserting or removing contact lenses are not permitted in any laboratory; the wearing of contact lenses is permitted only when other forms of corrective eyewear are not suitable; wearing jewelry is not recommended in the laboratory.
4. Oral pipetting of any substance is prohibited in any laboratory.
5. Long hair is to be tied back or restrained so that it cannot come into contact with hands, specimens, containers or equipment.
6. Access to laboratory and support areas is limited to authorized personnel.
7. Doors to laboratories must not be left open (this does not apply to an open area within a laboratory).
8. Open wounds, cuts, scratches and grazes should be covered with waterproof dressings.
9. Laboratories are to be kept clean and tidy. Storage of materials that are not pertinent to the work and cannot be easily decontaminated (e.g., journals, books, correspondence) should be minimized; paperwork and report writing should be kept separate from such biohazardous materials work areas.
10. Protective laboratory clothing, properly fastened, must be worn by all personnel, including visitors, trainees and others entering or working in the laboratory; suitable footwear with closed toes and heels must be worn in all laboratory areas.
11. Where there is a known or potential risk of exposure to splashes or flying objects, whether during routine operations or under unusual circumstances (e.g., accidents), eye and face protection must be used. Careful consideration should be given to the identification of procedures requiring eye and face protection, and selection should be appropriate to the hazard.
12. Gloves (e.g., latex, vinyl, co-polymer) must be worn for all procedures that might involve direct skin contact with biohazardous material or infected animals; gloves are to be removed when leaving the laboratory and decontaminated with other laboratory wastes before disposal; metal mesh gloves can be worn underneath the glove.
13. Protective laboratory clothing must not be worn in nonlaboratory areas; laboratory clothing must not be stored in contact with street clothing.
14. If a known or suspected exposure occurs, contaminated clothing must be decontaminated before laundering (unless laundering facilities are within the

containment laboratory and have been proven to be effective in decontamination).

15. The use of needles, syringes and other sharp objects should be strictly limited; needles and syringes should be used only for parenteral injection and aspiration of fluids from laboratory animals and diaphragm bottles; caution should be used when handling needles and syringes to avoid auto-inoculation and the generation of aerosols during use and disposal; where appropriate, procedures should be performed in a BSC; needles should not be bent, sheared, recapped or removed from the syringe; they should be promptly placed in a puncture-resistant sharps container (in accordance with Canadian Standards Association [CSA] standard Z316.6-95(R2000))<sup>(6)</sup> before disposal.
16. Hands must be washed after gloves have been removed, before leaving the laboratory and at any time after handling materials known or suspected to be contaminated.
17. Work surfaces must be cleaned and decontaminated with a suitable disinfectant at the end of the day and after any spill of potentially biohazardous material; work surfaces that have become permeable (i.e., cracked, chipped, loose) to biohazardous material must be replaced or repaired.
18. Contaminated materials and equipment leaving the laboratory for servicing or disposal must be appropriately decontaminated and labelled or tagged out as such.
19. Efficacy monitoring of autoclaves used for decontamination with biological indicators must be done regularly (i.e., consider weekly, depending on the frequency of use of the autoclave), and the records of these results and cycle logs (i.e., time, temperature and pressure) must also be kept on file.
20. All contaminated materials, solid or liquid, must be decontaminated before disposal or reuse; the material must be contained in such a way as to prevent the release of the contaminated contents during removal; centralized autoclaving facilities are to follow the applicable containment level 2 requirements.
21. Disinfectants effective against the agents in use must be available at all times within the areas where the biohazardous material is handled or stored.
22. Leak-proof containers are to be used for the transport of infectious materials within facilities (e.g., between laboratories in the same facility).
23. Spills, accidents or exposures to infectious materials and losses of containment must be reported immediately to the laboratory supervisor; written records of such incidents must be maintained, and the results of incident investigations should be used for continuing education.
24. An effective rodent and insect control program must be maintained.

### **3.2 WORKING WITH LABORATORY ANIMALS**

Animals can harbour infectious organisms, which are acquired naturally. Some infectious agents can give rise to a chronic carrier state, or an agent might be shed intermittently. If the possibility that such an agent may be excreted, secreted, exhaled or shed by an animal during the course of an experiment cannot be

excluded, then all those animals should be kept at the containment level appropriate to the risk. Animals may also be intentionally inoculated with viruses or other organisms in any of the four Risk Groups or with viable materials (e.g., transformed cells) that are suspected of containing these agents. Under these circumstances, the animals should be kept at the containment level appropriate to the risk of the agent. In some cases, *in vivo* work may increase that risk. Naturally occurring or experimentally induced infections in laboratory animals may be transmitted to other laboratory animals, invertebrates and laboratory workers. Laboratory animals and insects may scratch or bite or may be the source of aerosols.

Besides the risk from an infection that the animal or insect may be harbouring, there is also a risk that some of the material being injected may adhere to the fur or exoskeleton and remain as a potential hazard. In all situations, it is the responsibility of the principal investigator, Western's Biosafety Officer and the Biohazards Subcommittee in consultation with Government agencies and the animal care authorities, to determine the risk levels inherent in the proposed activity.

The requirements for the maintenance of animals may differ in scale and degree, but the basic principles for microbiological safety will be similar to those outlined in Section 3.1 and should include the following precautions.

1. Infected animals and insects should be segregated from uninfected animals wherever possible, and it is preferable to separate any handling area from the holding area.
2. Animals or insects in use in an experiment must be maintained at a level of containment which is at least equivalent to the containment level for the biological agent with which it has been infected or treated.
3. Provision must be made to ensure that inoculated animals or insects cannot escape.
4. Dead animals or insects and the refuse from the animal room and cages (e.g. bedding, feces and food) must be placed in a leakproof container and autoclaved or incinerated, if potentially infected.
5. All cages must be properly labelled, and procedures in the holding area must minimize the dispersal of dander and dust from the animals and cage refuse.
6. Gloves and safety glasses should be worn by animal care providers while feeding and watering animals or cleaning cages.
7. Gloves, boots, floors, walls and cage racks should be disinfected frequently.
8. All aspects of the proposed use of animals in research must meet the current veterinary standards and regulations for the care and maintenance of experimental animals as described by the Canadian Council on Animal Care, relevant provincial legislation, the University and the Animal Use Subcommittee.
9. The appropriate species must be selected for the animal experiments.
10. The investigator and/or person(s) responsible for the animal experiment must ensure that all those having contact with the animals and waste materials are familiar with and aware of any special precautions and procedures that may be required. Where possible, personnel should be protected by immunization with appropriate vaccines.

11. All incidents, including animal bites and scratches or cuts from cages or other equipment must be documented and the employee should report to Workplace Health for medical assessment and follow-up.
12. All Animal Care and Veterinary Services (ACVS) procedures and protocols must be followed with respect to the proper handling and care of animals. All staff members that work with animals must have training as required by ACVS.
13. There are animal facilities at Western University which require specific personal protective equipment and operating procedures. Use of these animal facilities requires strict adherence to these procedures.

### **3.3 HUMAN PATHOGENS**

Some microorganisms (viruses, bacteria, fungi, etc.) are species specific, selectively infecting and causing disease in a limited number of, or only one, host species. Unrelated and distantly related species may not be similarly affected by the same infectious microorganism due to differences in physiology, metabolism, biochemistry, and other factors. In general, the risk to a laboratory technician working with a virus that only infects and causes disease in rodents is lower than the risk to a laboratory technician working with tissues and cells from humans or other primates. If the human material contains a viable pathogen, it will likely be a human pathogen, with the potential to infect and cause disease in another human. Although a single mode of transmission may predominate, disease causing micro-organisms can be spread or transmitted from one host to the next, directly or indirectly, by a number of methods. Transmission methods include aerosol generation and inhalation, ingestion of contaminated food and water, skin and mucous membrane contact with contaminated surfaces, contact contamination of an open wound or lesion, and autoinoculation via a cut, and laceration or puncture with a contaminated instrument.

#### **3.3.1 HUMAN BLOODBORNE PATHOGENS**

Human blood is recognized as a potential source of pathogenic microorganisms that may present a risk to workers who are exposed during the performance of their duties. Although the hepatitis B virus (HBV) and the human immunodeficiency virus (HIV) are often cited as examples, a “bloodborne pathogen” is any pathogenic microorganism that is present in human blood or other potentially infectious materials and that can infect and cause disease in persons who are exposed to blood containing this pathogen. “Other potentially infectious materials” means material that has the potential to transmit bloodborne pathogens. This includes infected human tissues and the following body fluids: semen, vaginal secretions, cerebrospinal fluid, synovial fluid, pleural fluid, peritoneal fluid, pericardial fluid, amniotic fluid, saliva in dental procedures, and any other body fluid that is visibly contaminated with blood.

In 1988, the Centers for Disease Control published a series of recommendations and precautions for the protection of workers who have, or are likely to have, contact with human blood and certain body fluids and may be at risk of exposure to bloodborne pathogens such as hepatitis B virus (HBV) and human

immunodeficiency virus (HIV). These recommendations became known as “Universal Blood and Body Fluid Precautions” or simply, “Universal Precautions”.

### **3.3.2 UNIVERSAL BLOOD AND BODILY FLUID PRECAUTIONS**

The possibility of undiagnosed infection combined with the increasing prevalence of HBV and HIV led the Center for Disease Control (Atlanta, Georgia) to recommend that blood and other body fluids from all humans be considered potentially infectious and that precautions be taken to minimize the risk of exposure. This approach, called "Universal Precautions", is a method of infection control, intended to prevent parenteral, mucous membrane, and non-intact skin exposure of workers to bloodborne pathogens.

All human blood, human body fluids, and other materials are considered potentially infectious for hepatitis B virus (HBV), hepatitis C virus (HCV), human immunodeficiency virus (HIV), and other bloodborne pathogens. Precautions must be consistently used. Body fluids to which universal precautions apply include blood, body fluids containing visible blood, semen, vaginal secretions, cerebrospinal fluid, synovial fluid, pleural fluid, peritoneal fluid, pericardial fluid, and amniotic fluid.

It is prudent to minimize non-intact skin and mucous membrane contact with these materials. Hepatitis B immunization is highly recommended as an adjunct to universal precautions for workers who have occupational exposure to human blood or other potentially infectious materials. Western University Workplace Health provides this immunization to employees at risk, free of charge.

#### **General Precautions**

- 1) All workers should routinely use appropriate barrier precautions to prevent skin and mucous membrane exposure when contact with human blood or other body fluids is anticipated.
- 2) Eating, drinking, smoking, applying cosmetics or lip balm, and handling contact lenses are prohibited.
- 3) Gloves should be worn when touching blood and body fluids, mucous membranes, or non-intact skin, for handling items or surfaces soiled with blood or body fluids, and for performing venipuncture and other vascular access procedures. If a glove is torn or damaged during use, it should be removed and a new glove should be used as promptly as safety permits. Disposable gloves should not be washed or disinfected for reuse. Washing with surfactants may enhance penetration of liquids through undetected holes in the glove. Disinfecting agents may cause deterioration of the glove material.
- 4) Masks and protective eyewear or face shields should be worn during procedures that are likely to generate droplets of blood or other body fluids to prevent exposure of mucous membranes of the mouth, nose, and eyes.
- 5) Gowns or aprons should be worn during procedures that are likely to generate splashes of blood or other body fluids. Protective clothing should be removed before leaving the area.



- 6) Hands and other skin surfaces should be washed immediately and thoroughly if contaminated with blood or other body fluids. Hands should be washed immediately after gloves are removed since no barrier is 100% effective.
- 7) Workers should take precautions to prevent injuries caused by needles, scalpels, and other sharp instruments or devices during procedures, when cleaning used instruments, during disposal of used needles, and when handling sharp instruments after procedures. Needles and syringes should be used only in those situations when there is no alternative. To prevent needlestick injuries, needles should not be recapped, purposely bent or broken by hand, removed from disposable syringes, or otherwise manipulated by hand. After they are used, disposable syringes and needles, scalpel blades, and other sharp items should be placed in puncture-resistant containers for disposal. The puncture-resistant container should be located as close to the use area as practical. Contaminated reusable pointed and sharp objects such as large bore needles and scalpels should be placed in a puncture resistant container for transport to the reprocessing area.
- 8) Mouthpieces, resuscitation bags, or other ventilation devices should be available for use in areas in which the need for resuscitation is predictable.
- 9) Workers who have exudative lesions, weeping dermatitis, cuts, open wounds or other breaks in the skin should either refrain from all direct contact with blood and other body fluids until the condition resolves, or utilize protective barriers to reduce the risk of exposure.
- 10) Pregnant workers should be especially familiar with and strictly adhere to precautions to minimize the risk of prenatal transmission of bloodborne pathogens.

### **3.4 MEDICAL SURVEILLANCE AND IMMUNOPROPHYLAXIS**

Western University Workplace Health provides several health surveillance, testing and immunoprophylaxis programs for University employees. Additional programs will be added as needed. Immunoprophylaxis and information pertaining to the availability and the advisability of immunizing agents are available through the Western University Workplace Health. Laboratory personnel should be protected against laboratory-acquired infections by appropriate immunization with relevant, licensed vaccines unless they already have documented protective levels of pre-existing immunity.

Hepatitis B immunization is strongly recommended for all workers who routinely handle or have occupational exposure to human blood, body fluids, organs or tissues. Western University offers and provides hepatitis B immunization free of additional cost to risk employees through Workplace Health.

### **3.5 MEDICAL PROCEDURES AND INCIDENT REPORTING**

The following emergency response procedures shall be followed when a worker has been potentially exposed to a biohazardous agent via a needlestick, cut, animal bite or scratch, via mucous membrane contact, or via non-intact skin contact.

### **Worker**

1. The exposed site must be washed immediately.
  - a) In case of a needle-stick, cut, animal bite or scratch, wash with soap and water after allowing the wound to bleed freely.
  - b) If mucous (eyes, nose, mouth) membrane or non-intact (cuts, rash, eczema or dermatitis) skin contact, flush with water at the nearest faucet or eye wash station for a minimum of ten minutes.
2. The worker must immediately inform the Supervisor/Principal Investigator of the exposure incident.
3. The worker must seek prompt medical attention at Workplace Health (during the hours of operation), the nearest hospital emergency department or emergency clinic, or a Medical Practitioner of their choosing. Any information including the Material Safety Data Sheet or equivalent for the biohazardous agent must also be taken to the care provider.
4. The worker must provide information for a Accident/Incident Report (obtained from her/his Supervisor/Principal Investigator), describing the incident in detail, including the route of exposure and the emergency actions taken, and a description of the worker's duties as they relate to the exposure incident.

### **Supervisor / Principal Investigator**

1. Supervisors/Principal Investigators must complete and sign the University Accident/Incident Report.
2. The supervisor must ensure that exposure incidents are reported within 24 hours to Human Resources, fax (519) 661-2079. The form can be found at:

[http://uwo.ca/hr/form\\_doc/health\\_safety/form/aiir.pdf](http://uwo.ca/hr/form_doc/health_safety/form/aiir.pdf)

3. The supervisor must refer the affected worker(s) to the nearest hospital emergency department or emergency clinic, or preferably, to Workplace Health during hours of operation.

### **Occupational Health & Safety**

1. Occupational Health and Safety will investigate accidents/incidents as appropriate. Accidents/incidents may be used as training tools for faculty, staff and students as long as confidential information is omitted.

### **Workplace Health**

1. Workplace Health Services shall confer with the affected individual(s) and/or attending physician(s)/caregiver(s).
2. Counselling regarding potential exposure and infection, immunoprophylaxis and follow-up testing shall be offered to any worker if her/his exposure is determined to be of a nature that may transmit biohazardous agents.

### **Important Emergency Contact Numbers**

Workplace Health: ext. 82047

Campus Community Police Services: 911 from any campus phone or 519-661-3300 from a cellular or off-campus phone

## **4.0 EMERGENCY PROCEDURES**

### **4.1 SPILLS AND OTHER INCIDENTS RESULTING IN UNCONTROLLED RELEASES**

Emergency response plans required at Containment Levels 2 and 3 must include procedures for dealing with spills or other laboratory incidents that could be expected to result in the release of biological agents. Since the capacity of most commonly used laboratory culture containers is small, it is anticipated that most spills within the laboratory will be limited in size and therefore, of a minor nature. Although the specific response will depend on the type and nature of the incident, decontamination and clean-up procedures incorporating the steps outlined below are recommended. If the spill is large or of a nature that cannot be handled by laboratory personnel, call Campus Police at 911 from any campus phone or 519-661-3300 from a cellular or off-campus phone. Campus Community Police Services will activate the University Emergency Response Team. Effective disinfectants must be available in the laboratory at all times and for immediate use. In the event of a spill or container breakage resulting in the unintentional release of a biological agent:

- (i) Place paper towel or absorbent on the liquid
- (ii) Pour a strong disinfectant solution or granules (i.e. 10% bleach) around, but not on the spill, and mix the disinfectant with the spilled material cautiously;
- (iii) Evacuate the laboratory for a time expected to be sufficient for decontamination of the mixed material, normally 20 minutes;
- (iv) Carefully place paper into a bag for incineration;
- (v) Decontaminate all surfaces exposed to the spill with the disinfectant.

If aerosols may have been created in the spill or unintentional release, evacuate the laboratory for a time sufficient for most aerosols to settle, be dispersed, or removed by the ventilation system, usually 20-30 minutes. The use of respiratory protection should be considered for re-entry. Then proceed with items (i)-(v) above.

During an emergency, the first priority is the protection of the health and safety of personnel, followed by the environment (i.e. sewer drains), followed by equipment or property.

## **5.0 CLASSIFICATION OF BIOLOGICAL AGENTS**

### **5.1 GENERAL**

The standards and practices described in this manual apply to all laboratory research and teaching activities conducted within the University and its affiliated institutions where such activities involve the use of known biological agents or cultures, or when an agent has been recently isolated or is suspected to be present

in the material handled. Judgments of the inherent risks of a pathogen are made on the basis of such factors as the severity of the disease it causes, the routes of infection, its virulence and infectivity. This judgement should take into account the existence of effective therapies, immunization, presence or absence of vectors, quantity of agent and whether the agent is indigenous to Canada. It should consider possible effects on other species, including plants and animals. Due to their unknown characteristics, emerging pathogens and novel agents may require more stringent specialized practices and procedures for their safe handling.

Biological agents are classified according to Risk Groups, which are analogous to the Containment Levels described in Section 6.2. These classifications presume ordinary circumstances in the laboratory, or growth in small volumes for experimental, diagnostic or teaching purposes. The classifications of biological agents reflect the judgements made on their inherent risks. The general Criteria are indicated in Section 5.4. Risk Groups for agents will be identified on request to the Biosafety Officer, Biohazards Sub-committee and Office of Laboratory Security. Large volumes and high concentrations of a biological agent in growth media may pose greater risks than smears of the same agent on a microscope slide. Other unusual manipulations may also increase the hazard.

## **5.2 RECOMBINANT DNA AND GENETIC MANIPULATION**

For the purposes of this document, recombinant DNA includes:

- DNA molecules produced outside living cells by joining natural or synthetic DNA segments to DNA molecules capable of replication in living cells,
- DNA molecules produced in living cells by joining enriched or natural segments to intracellular DNA, and,
- DNA molecules resulting from replication of such recombinant molecules.

Guidance in assessing potential risks in recombinant DNA research can only be very general; each case requires individual assessment. It is unrealistic to define all of the genetically engineered organisms that might be created or used in the laboratory. The majority of this research involves only a very low possibility of creating a hazard because the source of the DNA being transferred, the vector and the host are all innocuous or have low risk characteristics. However, some genetic manipulation does raise a significant possibility of risk.

In general, containment levels for activities involving recombinant DNA will be assigned according to the following criteria and considerations:

- 1) If none of the components of the genetic manipulation (DNA, vector, host) presents any known hazard and none can be reasonably foreseen in their combination, then no restrictions beyond the requirements of Containment Level 1 are necessary.
- 2) If one of the components used in the procedure is hazardous, then, in general, determination of the containment level required will begin at the level appropriate to the known hazard. The level of containment may be increased or decreased depending on the particular gene transferred, the expression of the gene in the

recombinant organism, the envisaged interactions between the transferred gene and the host-vector system, and other relevant factors.

3) In any activity involving genes coding for hazardous products, host-vector systems with limited ability to survive outside of the laboratory (affording biological containment) should be used. Their use may reduce the level of physical containment required.

4) The containment level may be reduced if it is known that the DNA or vectors are mutant and defective in their disease-causing or replication characteristics.

5) In the case of animal virus vectors, including retroviruses, one must consider the nature of the helper cells and the likelihood that replication-competent viruses may be produced.

### **5.3 ANIMAL CELLS, BLOOD AND FLUIDS, AND FIXED TISSUES**

The biological hazards of animal cells, tissues, blood and body fluids arise from the possibility that they might contain or transmit infectious agents. It is prudent to consider all cell lines to be potentially infectious. Cells known or suspected to contain such agents, or primary cultures from animals and humans known or reasonably suspected to be infected, should be assigned to the risk group for the suspected agent. Primate cell lines, all samples of human tissues and fluids, all primate tissues, and all cell lines new to the laboratory should be handled at Containment Level 2. Factors such as the particular source of the material, the volume and concentration of the agent, the extent of culturing and incubation, the types of manipulations to be conducted, and the use of additional precautions could influence the containment level required.

#### **5.3.1 ANIMAL CELLS**

##### **1) Primary cell cultures and animal tissues**

The following containment requirements apply to primary cell cultures and tissues from human, non-human primate and non-primate animal sources when handled in the laboratory. Cells and tissues known or suspected to be contaminated or infected with biohazardous agents must be handled at the containment level appropriate to those agents.

##### **2) Established cell lines**

Human or other animal cell lines known to not be contaminated or infected with biohazardous agents may be handled at Containment Level 1. Cultures known or suspected to be contaminated or infected with any biohazardous agents must be handled at the containment level appropriate to those agents.

For more information on the use of animal cell lines, please see the “Use of Human and Rodent Primary and Established Cells in Rodents: Containment Requirements” on the following website: <http://www.uwo.ca/hr/safety/topics/biosafety/policies.html>

#### **5.3.2 BLOOD AND BODILY FLUIDS**

The need for precautionary measures extends also to situations in which human blood, saliva, urine and other body fluids or feces must be handled. The precautions required may be more stringent when the specimens are used for culturing purposes, but initially, their handling should be consistent with Containment Level 2. Reduction of the containment level may be acceptable if potential hazards associated with the material are expected to be diminished because of dilution, use of chemical or other treatments or additional protective measures and practices.

### **1) Culturing of specimens in research laboratory**

Blood or blood fractions and other body fluid specimens of human or animal origin that are known or suspected to contain any biohazardous agents must be handled at the containment level appropriate to those agents when these specimens are cultured in volumes greater than that which is necessary for routine diagnostic work.

### **2) Clinical diagnostic work in laboratory**

For clinical diagnostic work with specimens of human blood, serum and other body fluids (urine, cerebrospinal fluid, etc.) from the general population, Containment Level 2 and Universal Precautions apply. For routine clinical diagnostic work with specimens that are known to be from infected individuals, the containment level appropriate to the agent must be maintained.

## **5.3.3 FIXED TISSUES AND TISSUE SECTIONS**

Tissues and tissue sections from human and animal sources are routinely fixed by treatment with chemical agents, such as formaldehyde to preserve structures for later examination and study. Generally, these chemical treatments inhibit all biological activity.

In general, fixed tissues and tissue specimens should be handled under at least Containment Level 1 conditions. A higher level of containment may be required depending on the source of the material, the nature of the agent and whether or not it is inactivated. Where a biological agent that usually requires a higher level of containment is present in the tissue, the laboratory Principal Investigator must provide documentation to the Western University Biosafety Committee or the Biohazards Subcommittee which support a request for a lower level of containment.

## **5.4 BIOLOGICAL AGENT RISK GROUP CRITERIA AND CATEGORIES**

Classification of organisms according to risk group has traditionally been used to categorize the relative hazards of infective organisms. The factors used to determine which risk group an organism falls into is based upon the particular characteristics of the organism, such as

- pathogenicity;
- infectious dose;
- mode of transmission (host range);

- availability of effective preventive measures (availability of effective treatment).

These classifications presume ordinary circumstances in the research laboratory or growth in small volumes for diagnostic and experimental purposes. Four levels of risk have been defined by the Public Health Agency of Canada as follows.

#### **5.4.1 Risk Group 1(*low individual and community risk*)**

Any biological agent that is unlikely to cause disease in healthy workers or animals.

#### **5.4.2 Risk Group 2(*moderate individual risk, low community risk*)**

Any pathogen that can cause human disease but under normal circumstances is unlikely to be a serious hazard to laboratory workers, the community, livestock or the environment. Laboratory exposures rarely cause infection leading to serious disease; effective treatment and preventive measures are available, and the risk of spread is limited.

#### **5.4.3 Risk Group 2(*plus level 3 operations*)**

According to the Public Health Agency of Canada, these are any Level 2 agents that require additional requirements, or Level 3 operations, such as Lentiviral vectors for example. Projects that may require these measures are assessed on a case-by-case basis with the Biosafety officer, Biohazards Subcommittee and the Public Health Agency of Canada.

#### **5.4.4 Risk Group 3(*high individual risk, low community risk*)**

Any pathogen that usually causes serious human disease or can result in serious economic consequences but does not ordinarily spread by casual contact from one individual to another, or that causes diseases treatable by antimicrobial or antiparasitic agents.

#### **5.4.5 Risk Group 4(*high individual risk, high community risk*)**

Any pathogen that usually produces very serious human disease that is often untreatable, and may be readily transmitted from one individual to another, or from animal to human (or vice-versa), directly or indirectly, or by casual contact (1).

**NOTE:** Risk Group 4 agents are not approved for use at Western University and shipments including such agents must not be accepted. Contact the Western University Biosafety Officer if you anticipate or contemplate their use.

As a general precaution, agents should be elevated to the next risk group when manipulation may result in the production of infectious droplets and aerosols.

## **6.0 CONTAINMENT OF BIOLOGICAL HAZARDS**

## **6.1 INTRODUCTION**

Bacteria, viruses, fungi and parasites are used in a variety of laboratory settings, in many cases because of their significance as etiological agents, but also because a better understanding of their nature is important to many areas of biology. In addition, there is growing interest in the use of this information and the agents themselves in industrial applications. Hazards may not always be readily apparent. Risks posed by biological agents and other potentially pathogenic materials will vary with the agent or material, and the circumstances under which it is used. Risks can be minimized to acceptable levels by controlling or reducing the hazards, but they may not be entirely eliminated. Some laboratory procedures and processes are more likely than others to contribute to the dissemination of hazardous agents. Among factors that can contribute to the risk involved, the following are generally viewed as particularly significant.

### **6.1.1 AEROSOLS**

Because of their insidious nature, aerosols pose special problems in that the laboratory worker may be unwittingly exposed to the material handled. Procedures which can produce aerosols include grinding, blending, sonicating, resuspending packed cells or viruses, inserting a hot loop into a culture, centrifugation, flaming an inoculation loop so that the material sputters, forceful ejection of fluid from a pipette or syringe and opening a tube within which the air pressure may differ from that of the room. This may occur when the tube is opened at a temperature different from that at which it was sealed. Formation and dispersal of aerosols can be controlled by the use of proper techniques or special equipment. For example, both screw-capped safety cups and sealed centrifuge heads permit use of a centrifuge in an open laboratory with minimum risk of aerosol dispersal, provided that the cup or head is opened inside a suitable safety cabinet. However, while the use of available safety devices is recommended, their use is not a substitute for good technique. Once formed, aerosols can be captured by high efficiency particulate air (HEPA) filters or removed from the laboratory by local and room ventilation methods. A biological safety cabinet provides some operator protection against airborne materials, including aerosols.

### **6.1.2 LARGE VOLUMES AND HIGH CONCENTRATIONS**

The risks to laboratory personnel or the environment may increase as the volume or concentration of the biological agent increases. The procedures described in this manual relate primarily to small scales of operation normally encountered in University laboratories.

### **6.1.3 EFFLUENTS AND WASTE**

Effluents are a major potential means for dissemination of agents to the environment outside of the laboratory. These include air exhausted or escaping to the outside, liquid and solid wastes, and contaminated glassware.



## **Air**

The purpose of an air exhaust system is to remove contaminated air from a work area, to convey it through a decontaminating system if necessary, and to discharge it to the outside. Its design should provide adequate air exchanges, a negative pressure differential between the room and the air source to ensure that contaminated air departs only through the exhaust system, and airflow patterns through the room so that all parts of the room are swept by the airflow. The influence of opening and closing doors on these airflow patterns is of particular importance. Decontamination of air is best achieved with a HEPA filter. HEPA filters are ineffective unless properly installed. Testing of these filters *in situ* with an aerosol at the time of installation and at regular intervals is essential to ensure the integrity of the barrier. Normally, HEPA filters will require replacement only when they offer excessive resistance to airflow due to loading or when irreparable leaks are detected. Vacuum lines also serve as a conduit through which air may leave the laboratory and must also be protected.

## **Liquids**

Some liquid wastes, particularly those in which agents have been cultured, will require sterilization or disinfection to inactivate the agent before disposal to the sewage system. Hazardous chemical and radioactive liquid wastes may require an additional procedure to inactivate viable biological agents before removal from the laboratory. It is dangerous and illegal to dispose of hazardous chemicals and radioactive materials into drains and the sewage system. Autoclaving (steam sterilization) is generally the best method of inactivating biological agents and should be used whenever possible.

Liquid waste containers designed to withstand autoclaving temperatures must be used. Containers of liquid waste must be placed into a tray or pan of sufficient capacity to contain all liquid in the event of vessel failure or breakage inside the autoclave chamber. Although some chemical disinfectants can be used for the inactivation of many biological agents, others may be less effective against particular microorganisms, or may be suitable only for some of the types of disinfection required in the laboratory (disinfection of work surfaces or instruments, clean up after spills or accidents, and disinfection of liquid wastes). Before adoption, it is recommended that a disinfectant be tested against the biological agent to determine the concentration and contact time required to achieve the objective under the conditions employed.

## **Solids**

Reusable items such as glassware should be sterilized by autoclaving whenever possible. Otherwise, a specific chemical disinfection procedure, proven to be effective against the particular biological agent, must be used. Disposable items which are contaminated with biological agents only, should be incinerated or must be autoclaved or chemically disinfected before disposal. Disposable sharp waste must be carefully collected in a puncture-resistant waste container and incinerated. Intact and broken glassware for disposal must be collected in puncture-resistant containers and properly labelled.

Disposable non-sharp items (gloves, empty plastic culture dishes, flasks and tubes, absorbent tissue, etc.) which are contaminated with biological agents must be collected in autoclave bags. After autoclaving, cooling and verification, these bags of waste must be placed into black plastic garbage bags for disposal. Hazardous chemicals and radioactive solid wastes have unique procedures to inactivate viable biological agents which may be present before removal from the laboratory. Autoclaving is generally not recommended in all situations involving such wastes, since the high temperature, steam and pressure may contribute to potentially hazardous reactions. It is dangerous and illegal to dispose of hazardous chemicals and radioactive materials in the regular garbage going to landfill.

#### **6.1.4 PIPETTING**

Mouth (oral) pipetting is prohibited in any laboratory. Using commercially available pipetting devices can reduce pipetting accidents. However, delivery of fluids should be slow, as forceful ejection produces bubbles and spraying which can generate an aerosol. Pipettes, especially glass, must be inserted into pipetting devices carefully and without excessive force, to avoid breakage and potential injuries.

### **6.2 PHYSICAL CONTAINMENT LEVELS**

Four levels of containment (1 - 4), appropriate to the four risk groups for potentially hazardous biological agents, are defined. These levels of containment are regarded as adequate for most laboratory uses of the listed agents. It remains the responsibility of the Principal Investigator and Western University to require a higher level of containment for specific manipulations, if these appreciably increase the possibility of infection. Containment Level Two laboratories are inspected at least annually by Occupational Health and Safety.

Classification of organisms according to risk group is not meant to establish the actual handling of biological hazards in the laboratory setting. For example, the risk group system does not take into account the procedures that are to be employed during the manipulation of a particular organism. Containment levels are selected to provide the end-user with a description of the minimum containment required for handling the organism safely in a laboratory setting. In addition to the inherent characteristics of each organism as described in section 5.4, the containment system includes the engineering, operational, technical and physical requirements for manipulating a particular pathogen. These containment levels are applicable to facilities such as diagnostic, research, clinical, teaching and production facilities that are working at a laboratory scale. Four containment levels are described as follows (1):

#### **6.2.1 CONTAINMENT LEVEL 1 (CL1)**

This applies to the basic laboratory that handles agents requiring containment level 1. CL1 requires no special design features beyond those suitable for a well-designed and functional laboratory. Biological safety cabinets (BSCs) are not

required. Work may be done on an open bench top, and containment is achieved through the use of practices normally employed in a basic microbiology laboratory.

#### **6.2.1.1 PHYSICAL REQUIREMENTS**

1. Separated from public areas by door.
2. Size of door openings to allow passage of all anticipated equipment.
3. Surfaces to be scratch, stain, moisture, chemical and heat resistant in accordance with laboratory function (recommended).
4. Surfaces to provide impact resistance in accordance with laboratory function (recommended).
5. Interior coatings to be gas and chemical resistant in accordance with laboratory function (e.g., will withstand chemical disinfection, fumigation) (recommended).
6. Bench tops to have no open seams (recommended).
7. Bench tops to contain spills of materials (e.g., with marine edges and drip stops) (recommended).
8. Benches, doors, drawers, door handles, etc. to have rounded rims and corners (recommended).
9. Backsplashes, if installed tight to wall, to be sealed at wall-bench junction (recommended).
10. Reagent shelving to be equipped with lip edges (recommended).
11. Drawers to be equipped with catches, i.e., to prevent the drawer from being pulled out of the cabinet (recommended).
12. Cabinet doors not to be self-closing (recommended).
13. Autoclave or other acceptable means of waste treatment/disposal to be provided (recommended).
14. Windows, if they can be opened, to be protected by fly screens.
15. Hooks to be provided for laboratory coats at laboratory exit; street and laboratory clothing areas to be separated.
16. Hand washing sinks to be located near the point of exit from the laboratory or in anteroom (1).

#### **6.2.1.2 OPERATIONAL REQUIREMENTS**

The basic laboratory safety practices described in Section 3.1 must be followed. Where chemical disinfection procedures are employed, effective concentrations and contact times must be used. Chemical disinfectants used to decontaminate materials to be removed from the laboratory must be replaced regularly (1).

#### **6.2.2 CONTAINMENT LEVEL 2 (CL2)**

Containment Level 2 is suitable for work with agents in Risk Group 1 or 2. The primary exposure hazards associated with organisms requiring CL2 are through the

ingestion, inoculation and mucous membrane route. Agents requiring CL2 facilities are not generally transmitted by airborne routes, but care must be taken to avoid the generation of aerosols (aerosols can settle on bench tops and become an ingestion hazard through contamination of the hands) or splashes. Primary containment devices such as BSCs and centrifuges with sealed rotors or safety cups are to be used as well as appropriate personal protective equipment (i.e., gloves, laboratory coats, protective eyewear). As well, environmental contamination must be minimized by the use of handwashing sinks and decontamination facilities (autoclaves) (1). In addition to the requirements of Containment Level 1, the following are required:

### **6.2.2.1 PHYSICAL REQUIREMENTS**

1. Access limited to authorized personnel.
2. Laboratory room doors to have appropriate signage (e.g., biohazard sign, containment level, contact information, entry requirements).
3. Doors to the containment laboratory are lockable (this does not apply to areas within the containment laboratory).
4. Office areas to be located outside of the containment laboratory. Paperwork stations for data collection can be within the containment laboratory provided they are located away from laboratory work areas (recommended).
5. Doors, frames, casework and bench tops are to be non absorptive (i.e. the use of organic materials should be avoided). (recommended).
6. Working surfaces of benchtops are to be non-absorptive.
7. Surfaces are to be scratch, stain, moisture, chemical and heat resistant in accordance with laboratory function.
8. Surfaces are to provide impact resistance in accordance with laboratory function (recommended).
9. Surfaces are to be continuous and compatible with adjacent and overlapping materials (i.e., to maintain adhesion and a continuous perimeter). (recommended).
10. Interior coatings are to be gas and chemical resistant in accordance with laboratory function (e.g., will withstand chemical disinfection, fumigation).
11. Bench tops are to have no open seams (recommended).
12. Bench tops are to contain spills of materials (e.g., with marine edges and drip stops) (recommended).
13. Benches, doors, drawers, door handles, etc. are to have rounded rims and corners (recommended).
14. Backsplashes, if installed tight to wall, are to be sealed at wall-bench junction (recommended).
15. Reagent shelving is to be equipped with lip edges (recommended).
16. Drawers to be equipped with catches, i.e., to prevent the drawer from being pulled out of the cabinet (recommended).
17. Cabinet doors should not to be self-closing (recommended).
18. 100% outside air is to be supplied (recommended).

19. Autoclave or other acceptable means of waste treatment/disposal is to be provided.
20. Windows, if they can be opened, are to be protected by fly screens.
21. Hooks are to be provided for laboratory coats at laboratory exit; street and laboratory clothing areas are to be separated.
22. Handwashing sinks are to be located near the point of exit from the laboratory or in anteroom.
23. Handwashing sinks are to be provided with "hands-free" capability (recommended).
24. Biological Safety Cabinets (BSCs) and other primary containment devices are to be provided. Examples for use include procedures with the potential for producing aerosols and those involving high concentrations, large volumes or particular types of agents (1).

### **6.2.2.2 OPERATIONAL REQUIREMENTS**

In addition to the general practices required for all laboratories handling infectious substances, the following describe the minimum operational practices required for containment level 2.

1. Good microbiological laboratory practices intended to avoid the release of infectious agents are to be employed.
2. BSCs must be used for procedures that may produce infectious aerosols and that involve high concentrations or large volumes of biohazardous material. Laboratory supervisors, in consultation with the Biological Safety Officer/Institutional Biosafety Committee, should perform a risk assessment to determine which procedures and what concentrations and volumes necessitate the use of a BSC.
3. Appropriate signage indicating the nature of the hazard being used (e.g., biohazard sign, containment level) must be posted outside each laboratory; if infectious agents used in the laboratory require special provisions for entry, the relevant information must be included on the sign; the contact information of the laboratory supervisor or other responsible person(s) must also be listed.
4. Entry must be restricted to laboratory staff, animal handlers, maintenance staff and others on official business.
5. All people working in the containmentment area must be trained in and follow the operational protocols for the project in process. Trainees must be accompanied by a trained staff member. Visitors, maintenance staff, janitorial staff and others, as deemed appropriate, must also be provided with training and/or supervision commensurate with their anticipated activities in the containment area.
6. Emergency procedures for spill clean-up, BSC failure, fire, animal escape and other emergencies must be written, easily accessible and followed. A record must be made of other people entering the facility during an emergency (1).

### **6.2.2.3 ADDITIONAL REQUIREMENTS FOR LEVEL 2 PLUS LABORATORIES**

As per the Public Health Agency of Canada, work involving some Level 2 agents requires additional measures.

1. There must be a program in place (with appropriate authority to oversee safety and containment practices) for the management of biological safety issues.
2. General operational protocols must be supplemented with protocols similar to each project in progress.
3. Personnel must have demonstrated proficiency in microbiological practices and techniques.
4. Infectious agents should be stored inside the containment laboratory; agents stored outside of the containment laboratory must be in leak proof containers in a restricted area; emergency response procedures must take into account the existence of infectious agents that are stored outside of the containment area.
5. Personnel entering the containment laboratory must remove street clothing and change into dedicated laboratory clothing and shoes. Dedicated laboratory clothing and shoes must be removed before leaving the containment laboratory in a manner that minimizes any contamination of the skin with the potentially contaminated dedicated laboratory clothing. The use of full coverage protective clothing (i.e., completely covering all street clothing) is an acceptable alternative. When a known or suspected exposure may have occurred, all clothing, including street clothing, requires appropriate decontamination.
6. If an additional layer of protective clothing (e.g. solid-front gowns with tight fitting wrists, gloves, respiratory protection) is worn over laboratory clothing when handling infectious materials, it should be removed after completion of work (e.g. dedicated for use at the BSC).
7. Centrifugation of infectious materials must be carried out in closed containers placed in sealed safety cups or rotors that are unloaded in a BSC.
8. It is recommended that all activities with infectious materials are conducted in a BSC. If this is not possible, other primary containment devices in combination with personal protective clothing and equipment must be used.
9. In the event of an emergency, exit protocols must be established whereby routine procedures might be bypassed; a reporting area must be identified where further steps must be taken (e.g. disinfecting footwear, changing, showering).

### **6.2.3 CONTAINMENT LEVEL 3 (CL3)**

Containment Level 3 is suitable for work with agents in Risk Group 1, 2, or 3. The operational requirements for the Level 3 laboratory are substantially greater than those for Levels 1 and 2 and the laboratory staff must receive specific training in the safe handling and manipulation of the agents used in this laboratory. The Containment Level 3 laboratory is designed to minimize environmental release of hazardous materials and provide enhanced worker protection, and it must undergo annual performance testing and verification. A Containment Level 3 laboratory requires specialized design and construction. Those responsible for biosafety in an

institution should maintain close control and seek expert advice in, and remain in close communication throughout, all phases of design, construction, initial and annual performance testing and verification, operation and maintenance.

The following are additional to the requirements of Containment Levels 1 and 2. This applies to the laboratory that handles agents requiring containment Level 3. These agents may be transmitted by the airborne route, often have a low infectious dose that produces effects and can cause serious or life-threatening disease. The Level 3 facility at Western University is for research with non-aerosol agents, such as HIV.

### **6.2.3.1 PHYSICAL REQUIREMENTS**

1. Doors to provide restricted access by installation of a controlled access system (e.g., card key) or equivalent.
2. Electronic locking systems to be backed up with a physical key-lock system (recommended).
3. Office areas are to be located outside of containment laboratory. Paperwork stations for data collection can be within containment laboratory provided they are located away from laboratory work areas.
4. Entry to laboratory is to be provided via an anteroom.
5. Anteroom door(s) located between the clean and dirty change rooms are not to be opened simultaneously with either the containment laboratory door or the clean change entry door. (Interlock, visual or audible alarms, or protocols are all acceptable means).
6. Interlocked doors, if present, are to have manual overrides for emergency exit.
7. Entry to laboratory zone to be provided with clothing change areas separating personal and laboratory clothing dedicated to that zone (i.e., "clean" change area separated from "dirty" change area).
8. Exit from laboratory is to be provided with a walk-through shower on the containment barrier (i.e., between "dirty" and "clean" change anterooms). (Recommended for HIV labs)
9. Containment laboratories to be located in close proximity to supporting mechanical services to limit the amount of potentially contaminated services (recommended).
10. Containment laboratories are to be located away from external building envelope walls (recommended).
11. A laboratory support area is to be provided adjacent to the containment facility for all supporting laboratory manipulations (recommended).
12. Doors, frames, casework and bench tops are to be nonabsorptive (i.e., the use of organic materials should be avoided).
13. Surfaces are to provide impact resistance in accordance with laboratory function.
14. Surfaces are to be continuous and compatible with adjacent and overlapping materials (i.e., to maintain adhesion and a continuous perimeter); wall and floor welded seams are acceptable in level 3 laboratories.

15. Continuity of seals are to be maintained between the floor and wall (a continuous cove floor finish up the wall is recommended).
16. Interior coatings are to be cleanable.
17. Interior surfaces are to minimize movement of gases and liquid through perimeter membrane.
18. Bench tops are to have no open seams.
19. Bench tops are to contain spills of materials (e.g., with marine edges and drip stops) (recommended).
20. Benches, doors, drawers, door handles, etc. are to have rounded rims and corners (recommended).
21. Backsplashes, if installed tight to wall, are to be sealed at wall-bench junction (recommended).
22. Reagent shelving is to be equipped with lip edges (recommended).
23. Drawers are to be equipped with catches, i.e., to prevent the drawer from being pulled out of the cabinet (recommended).
24. Cabinet doors are not to be self closing (recommended).
25. 100% outside air is to be supplied.
26. Directional inward airflow provided in such a way that air will always flow towards areas of higher containment (e.g.,  $\pm 25$  Pa differential).
27. Visual pressure differential monitoring devices are to be provided at entry to containment laboratory.
28. Alarm (visual or audible) is to be provided in the laboratory and outside laboratory area (i.e., to warn others and maintenance personnel) to signal air handling systems failure).
29. Where determined necessary by a local risk assessment, supply air duct is to be provided with backdraft protection (i.e., HEPA filter; bubble tight backdraft damper).
30. Supply air system is to be independent of other laboratory areas. CL3 supply can be combined with areas of lower containment when provided with backdraft protection (i.e., HEPA filter, bubble tight backdraft damper) downstream from the connection (recommended for HIV labs).
31. Supply air system is to be interlocked (i.e., fans, dampers, electrical) with exhaust air system, to prevent sustained laboratory positive pressurization.
32. Exhaust air is to be HEPA filtered (recommended for HIV labs).
33. HEPA filters installed into the supply and exhaust system is to conform to the requirements of IEST-RP-CC001.3.
34. Where HEPA filters are used for backdraft protection in accordance with local risk assessment, supply HEPA filter housings are to be designed to withstand structural change at applied pressure of 2500 Pa [10 in. w.g.].
35. Exhaust HEPA filter housings are to be designed to withstand structural change at applied pressure of 2500 Pa [10 in. w.g.] and to be provided with a method of isolation and decontamination (recommended for HIV labs).
36. Exhaust air system is to be independent of other laboratory areas. CL3 exhaust can be combined with areas of lower containment when provided with a HEPA filter upstream from the connection (recommended for HIV labs).



37. Supply and exhaust systems located outside of containment are to be accessible for repairs, maintenance, cleaning and inspection (recommended).
38. Where backdraft protection is required in accordance with local risk assessment, supply air duct work that is outside the containment perimeter (e.g., between containment perimeter and HEPA filter or bubble tight backdraft damper) to be sealed airtight in accordance with SMACNA Seal Class A.
39. Exhaust air ductwork that is outside the containment perimeter (e.g., between containment perimeter and HEPA filter or bubble tight backdraft damper) is to be sealed airtight in accordance with SMACNA Seal Class A (recommended for HIV labs).
40. Airflow control devices and duct sensors are to be located downstream of the exhaust HEPA filter and upstream of the supply bubble tight backdraft damper or HEPA filter, or if located upstream, duct penetrations to be sealed in penetrations to be sealed in accordance with SMACNA Seal Class A (recommended for HIV labs).
41. Bubble tight backdraft dampers and HEPA filters are to be located in close proximity to the containment perimeter (recommended for HIV labs).
42. Double-door barrier autoclave with bioseal is to be located on containment barrier; body of autoclave to be preferably located outside of containment for ease of maintenance (recommended for HIV labs).
43. Barrier autoclave is to be equipped with interlocking doors, or visual or audible alarms to prevent both doors from opening at the same time.
44. For materials that cannot be autoclaved (e.g., heat sensitive equipment, samples, film) other proven technologies for treatment (e.g., incineration, chemical, or gas) are to be provided at containment barrier.
45. All penetrations are to be sealed with non shrinking sealant at containment.
46. All conduit and wiring are to be sealed with non shrinking sealant at the containment barrier.
47. Window positioned on containment barrier is to be sealed in place; window glazing material to provide required level of security.
48. Observation windows are to be installed on containment barrier.
49. Hand washing sinks are to be located near the point of exit from the laboratory or in anteroom.
50. Hand washing sinks are to be provided with "hands-free" capability.
51. BSCs and other primary containment devices are to be provided.
52. When it is not possible to limit the quantities of hazardous chemicals within the laboratory, emergency shower equipment is to be provided in accordance with applicable regulations (i.e., ANSI Z358.1-1998).
53. Domestic water branch piping serving laboratory area(s) are to be provided with backflow prevention, in accordance with CAN/CSA-B64.10-01/B64.10.1-01, and isolation valves, are to be located in close proximity to the containment barrier.
54. Drain lines and associated piping (including autoclave condensate) are to be separated from lower containment laboratory areas and to go directly to main building sanitary sewer at point of exit from building (downstream of all

- other connections).
55. Autoclave condensate drain is to have a closed connection. For CL3, open connection is allowable if located within containment barrier.
  56. Drainage traps are to be provided to required deep seal depth in consideration of air pressure differentials.
  57. Floor drains are not to be provided, except when essential (e.g., body shower and animal rooms) (recommended).
  58. Plumbing vent lines are to be independent of lower containment plumbing vent lines, or combined with lines from lower containment when provided with a filter of efficiency equivalent to that of HEPA upstream from the connection (CL3 laboratories manipulating organisms, such as HIV, that are not infectious via inhalation are not required to fulfill this criterion).
  59. Compressed gas cylinder(s) are to be located outside the laboratory (recommended). Portable vacuum pump is to be provided in the laboratory. Internal contamination of vacuum pump to be minimized (e.g., HEPA filtration of vacuum line, use of disinfectant traps).
  60. Emergency lighting is to be provided.
  61. Life safety systems, lighting, HVAC systems, BSCs, security systems and other essential equipment are to be supported with emergency back-up power.
  62. Circuit breakers are to be located outside biocontainment area.
  63. Fluorescent light ballasts and starters to be located outside containment area (recommended).
  64. Laboratory is to be equipped with a communication system between containment area and outside support area.
  65. System (e.g., fax, computer) is to be provided for electronic transfer of information and data from laboratory area to outside laboratory perimeter (Note: paperwork from the containment laboratory may be removed after appropriate decontamination, i.e., autoclaving, irradiation, microwaving; such practices are generally not recommended for use on a routine basis) (1).

### **6.2.3.2 OPERATIONAL REQUIREMENTS**

In addition to the operational practices for all laboratories handling infectious substances and those minimum requirements for containment Level 2, the following describe the minimum operational practices required at containment Level 3.

1. There must be a program for the management of biological safety issues in place with appropriate authority to oversee safety and containment practices (see Chapter 2, Section 2.5).
2. Everyone entering the containment laboratory must have completed a training course in procedures specific to the containment laboratory and must show evidence of having understood the training; training must be documented and signed by the employee and supervisor.
3. Employees working in the containment area must have knowledge of the physical operation and design of the facility (e.g., air pressure gradients between zones, directional airflow patterns, alarm signals for air pressure failure, containment perimeter).

4. A protocol specific to the operation of the laboratory must be developed and read by personnel; employees must certify in writing that they have understood the material in the protocol. This should include entry and exit protocols for people, animals, equipment, samples and waste. General protocols must be supplemented with protocols specific to each project in progress.
5. Personnel must have demonstrated proficiency in microbiological practices and techniques.
6. Smoke testing (i.e., using a smoke pencil held at the door between the anteroom and the containment facility, and other doors as required) should be done periodically by laboratory staff to verify correct airflow; a containment check must be performed before entering the containment laboratory (e.g., verify correct reading on the pressure monitoring device).
7. People entering a containment facility must be well prepared and bring all materials they will need with them. If something has been forgotten, established traffic patterns must still be adhered to (i.e., do not go back to get it; either phone for someone to bring it or exit using proper protocols).
8. Routine laboratory cleaning must be done by personnel using the containment facility or by specific personnel dedicated and trained for this task.
9. The containment laboratory must be kept locked.
10. Infectious agents should be stored inside the containment laboratory. Agents stored outside of the zone must be kept locked, in leakproof containers; emergency response procedures are to take into account the existence of such infectious agents outside of the containment Level 3 laboratory.
11. Personal items such as purses and outdoor clothing must not be brought into the containment laboratory.
12. Drainage traps must be filled with liquid (i.e., through regular sink usage, automatic primers or by filling traps in areas that are not frequently used).
13. Laboratory samples and supplies may be carried into the containment laboratory or passed in through a pass-box. If the barrier autoclave is used to pass materials into the laboratory, the autoclave must have been cycled before the outer "clean side" door is opened.
14. Personnel entering the containment laboratory must remove street clothing and jewelry, and change into dedicated laboratory clothing and shoes. Dedicated laboratory clothing and shoes must be removed before leaving the containment laboratory in a manner that minimizes any contamination of the skin with the potentially contaminated dedicated laboratory clothing. The use of full coverage protective clothing (i.e., completely covering all street clothing) is an acceptable alternative. When a known or suspected exposure may have occurred, all clothing, including street clothing, requires appropriate decontamination. Laboratories manipulating organisms, such as HIV, that are not infectious via inhalation, are not required to remove street clothing.
15. An additional layer of protective clothing (i.e., solid-front gowns with tight-fitting wrists, gloves, respiratory protection) may be worn over laboratory clothing when infectious materials are directly handled and should be

- removed after completion of work (e.g., dedicated for use at the BSC).
16. Centrifugation of infectious materials must be carried out in closed containers placed in sealed safety cups or rotors that are unloaded in a BSC.
  17. Animals or arthropods that have been experimentally infected must remain in the laboratory or appropriate animal containment facility.
  18. When a known or suspected aerosol exposure may have occurred, protocols based on a local risk assessment must be in place to determine whether showering is required on exit from the laboratory.
  19. All activities with infectious materials are conducted in a BSC. If this is not possible, other primary containment devices in combination with personal protective clothing and equipment must be used. No work with open vessels containing infectious materials is conducted on the open bench.
  20. Heat-sensitive materials that cannot be autoclaved out of the containment laboratory must be decontaminated at the containment barrier (e.g., fumigated with formaldehyde, vaporized hydrogen peroxide or a suitable alternative; disinfected using liquid chemicals; or subjected to other technology proven to be effective).
  21. Emergency procedures for failure of air handling systems and other containment emergencies must be written, easily accessible and followed.
  22. In the event of life-threatening emergencies, personal health and safety are a priority. Exit protocols must be established whereby routine procedures might be bypassed; a reporting area must be identified where further steps must be taken (e.g., disinfecting footwear, changing, showering) (1).

### **6.3 ANIMAL BIOHAZARD CONTAINMENT FACILITIES**

Laboratory facilities must provide containment for laboratory animals exposed to or harbouring infectious agents that is appropriate to the risk level of the infectious agents involved. In addition to the physical requirements identified in Section 6.2, special equipment (e.g., filter cages, isolation caging systems) appropriate to the animal species as well as to the level of risk must be used.

Operational procedures for the care and maintenance of the infected animals must satisfy the Guidelines for the Care and Use of Experimental Animals of the Canadian Council on Animal Care and the Animal Use Sub-committee. In order to ensure not only protection for laboratory personnel and the environment, but to ensure that every care is taken to avoid causing the animals unnecessary pain or suffering and to provide the animals with the highest quality care.

#### **6.3.1 ANIMAL ESCAPE**

Rodents kept in micro isolators rarely escape from the biological safety cabinets. If they do escape, they can easily be corralled into a corner. Use the proper personal protective equipment and retrieve the animal with tongs or other suitable equipment.

### **7.0 TRAINING**

Occupational Health and Safety offers several courses including:

- WHMIS
- X-Ray Safety Training
- Radiation Safety
- Laser Safety Training
- Faculty/Supervisors Safety Responsibilities Seminar
- Employee Health and Safety Orientation
- Environmental and Waste Management Training
- Biosafety Training

Biosafety training is mandatory for all new principal investigators, research staff and graduate students who work with biohazards that could include microorganisms, cell cultures and human blood and body fluids.

On completion of Biosafety training, the participant will:

- understand the process of risk assessment for work with microorganisms and cell lines
- understand the concept of containment level as it applies to biohazard laboratories
- understand how a biological safety cabinet works and its role in a biohazard laboratory
- know the procedures for responding to accidental exposure or spills of biohazardous materials
- understand the risks associated with human blood and body fluids
- know how to apply precautions when working with human blood and body fluids.

For comments or information, please contact the Western Biosafety Officer, Occupational Health and Safety, (519) 661-2111 ext. 88730

## **7.1 RETRAINING**

Personnel are required to be re-trained per the training policy on the website:

[http://uwo.ca/hr/form\\_doc/health\\_safety/doc/policies/training\\_policy.pdf](http://uwo.ca/hr/form_doc/health_safety/doc/policies/training_policy.pdf)

## **8.0 BIOHAZARD RECEIPT POLICY**

The purchase of all biohazards must be performed by authorized staff within the Western Purchasing Department, on behalf of the Permit Holder. All purchases are reviewed and approved by the Western Biosafety Officer or authorized OHS personnel prior to receipt by the Permit Holder to ensure that the facilities meet the operational and physical requirements necessary to handle the hazard safely. Biohazards may not be purchased via the University's Low Value Purchase Order procedure or by calling directly to any supplier. Records shall be maintained in the laboratory.

Acquisitions not purchased must be listed on the permit or authorized by calling the

Western Biosafety Officer, at extension 88730 prior to their receipt.

## **8.1 COMPLIANCE ENFORCEMENT POLICY**

Western University assumes the responsibility of ensuring to the Public Health Agency of Canada and Canadian Food Inspection Agency that the use of biohazards will be undertaken in a safe manner and in compliance with their guidelines. To aid in determining the level of risk or immediate danger to safety and health, all compliance violations will be categorized as major or minor offences. This policy is intended as a means to categorize and give guidance for the anticipated response that is needed. When issues of non-compliance are identified by Occupational Health and Safety, all deficiencies must be corrected and reported in writing to the Western Biosafety Officer.

Any offence occurring twice in any 1 year period will be considered as a second offence. A major offence would result from violations which cause immediate risk or danger to safety and health or cause a release of biohazards to the environment or community. For example, a major offence would be one of the following deficiencies:

1. Use or storage of food/drink or smoking in the laboratory
2. Inadequate training of new staff
3. Non-participation in the Level 2 Inspection Program
4. Unauthorized possession/use of biohazards
5. Inadequate or unsafe storage areas for biohazards
6. Non-compliance with a Western Standard Operating Procedure in a Western Animal Facility.

A minor offence would be an infraction which poses no immediate risk or threat to safety, health, the environment or the Licence. Examples of a minor offense would be one of the following deficiencies:

1. Inadequate signage
2. Inadequate posting (i.e. permit)
3. Inappropriate use of biohazard warning labels

Major Offence Actions:

1. First Offense: A written notification will be sent to the Chair, Biosafety Committee, to the Supervisor, copy to Department Chair, Director OHS and Biosafety Officer. Immediate correction action of the violation is required, written reply in 7 days. If the written reply is not received after 7 days, the second notice will be copied to the Dean of Faculty. A meeting will be arranged with the Permit Holder, Department Chair, Director OHS, Biosafety Committee Chair and Western Biosafety Officer if there is no response from the Permit Holder after 7 days of second notice.
2. Second Offense: The Permit Holder will be notified in writing by the Western Biosafety Officer that the permit will be suspended until a meeting with the Biosafety Committee can be held to discuss the

- offence(s).
3. Third Offence: The Western Biosafety Officer will recommend cessation of activity(ies).

Note: For the second and third occurrences, notification of the above actions will be copied to the Dean of Faculty, Department Chair, Director OHS and Biosafety Committee Chair.

Minor Offence Actions:

1. First Offense: A written notification will be sent to the Supervisor, copy to Department Chair, Director OHS and Biosafety Committee Chair. Corrective action of the violation is required, written reply in 21 days. If the written reply is not received until after 21 days, the second notice will be copied to the Dean of Faculty. A meeting will be arranged with the Permit Holder, Department Chair, Director OHS, Biosafety Committee Chair and Western Biosafety Officer if there is no response from the Permit Holder after 14 days of second notice.
2. Second Offence: A meeting will be arranged with the Supervisor, Department Chair, Director OHS, Biosafety Committee Chair and Western Biosafety Officer to review the issues.
3. Third Offence: The Supervisor will be notified in writing by the Western Biosafety Officer that the permit will be suspended until a meeting with the Biosafety Committee can be held.
4. Fourth Offence: The Western Biosafety Officer will recommend permit cancellation to the Biosafety Committee.

Note: For the second, third and fourth occurrences, notification of the above actions will be copied to the Dean of Faculty, Department Chair, Director OHS and Biosafety Committee Chair.

All permit holders, employees and Western facility users must work in accordance with regulations and/or Western policies and procedures. If there is a compliance issue, representative(s) from the affiliated institution, the Dean, Director or other individuals responsible will be informed and corrective measures may be taken.

## REFERENCES

- (1) Canadian Biosafety Standard, Second Edition
- (2) Canadian Centre for Occupational Health and Safety website, 2005, [www.ccohs.ca](http://www.ccohs.ca)
- (3) Canadian Standards for Veterinary Facilities. Ottawa: Canadian Food Inspection Agency, 1996.