

**UNIVERSITY OF ONTARIO
INSTITUTE OF TECHNOLOGY**

BIOSAFETY MANUAL

Revision 3

June 2014

Emergency Procedure and Contact Information

UOIT CAMPUS

| | |
|---|-------------------|
| In the event of a medical emergency: | 2400 (911) |
| First Aid: | 2400 |
| Campus Safety and Security: | 2400 |
| Biosafety Officer: | 6521 |
| Office of Research Services | 3692 |
| | |
| Off Hours | |
| Campus Safety and Security | 2400 |

OFF CAMPUS

| | |
|-------------------------------|-----|
| In the event of an emergency: | 911 |
|-------------------------------|-----|

Emergency procedure:

In the event of a medical emergency, where injuries are life-threatening, seek medical attention immediately. Contact Campus Safety and Security who will coordinate the response with emergency personnel.

The following procedure is to be followed when any **worker is exposed to Biohazards:**

1. For any break in the skin or laceration, allow wound to freely bleed and wash the exposed area immediately with mild soap and water.
2. For mucous membrane or skin contact, flush the area with water for 15 minutes (of uninterrupted flow) using the nearest eye wash station.
3. Report the incident to your supervisor immediately to initiate Incident Reporting protocols.
4. Prompt medical attention should be sought at the Campus Health Centre, nearest emergency clinic or hospital or medical practitioner of the workers choosing.

The following procedure is to be followed in the event of a **Biohazardous Spill:**

1. Evacuate the laboratory to allow aerosolized agents to settle or be removed through the ventilation system (~30 mins); leave BSC or fume hoods running.
2. Wearing personal protective clothing, pour the appropriate disinfectant around the spill and mix into the spill cautiously to minimize aerosol generation.
3. Cover the area with absorbent material and let soak for 20-30 minutes.
4. Place all absorbent and contaminated materials including clothing into clearly marked biohazard disposal bags.
5. Wash hands carefully with soap and water.
6. Report the incident to your supervisor to initiate Incident Reporting protocols.

For additional information and the complete details on Emergency Procedures and Incident Reporting, please refer to Chapter 10.

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PREAMBLE

The Laboratory Biosafety Guidelines, 3rd Edition, 2004, published by Health Canada, was the key document governing the use of biohazardous materials in Canada until 2013. This manual was developed based on the containment standards and operational guidelines described in that document. The guidelines were adopted in 2004 and were established for research and teaching laboratories in which biological agents or biohazardous materials were used. In June 2013, the Public Health Agency of Canada's Pathogen Regulation Directorate (PHAC) and the Office of Biohazard Containment & Safety, Canadian Food Inspection Agency (CFIA), released the new jointly developed Canadian Biosafety Standards and Guidelines (CBSG), 1st Edition. This new document amalgamated and updated three previously used biosafety standards on facility design and construction and guidelines for facility operation where human and terrestrial animal pathogens or toxins are handled or stored:

- Laboratory Biosafety Guidelines, 3rd Edition, 2004 (PHAC)
- Containment Standards for Veterinary Facilities, 1st Edition, 1996 (CFIA)
- Prions: Containment Standards for Laboratories, Animal Facilities and Post Mortem Rooms handling Prion Disease Agents, 1st Edition, 2005 (CFIA)

This manual was created and updated using widely accepted national and international reference materials, including legislation directing the importation, transportation, handling, storage and disposal of biological and biohazardous materials, and reflects current best practices. The University of Ontario Institute of Technology (UOIT) Biosafety manual has been reviewed and updated to reflect any new physical containment and operational practice requirements based on recent changes with respects to the CBSG, which will form the bases of regulations created under the Human Pathogens and Toxins Act. The manual is endorsed by the Institutional Biosafety Committee (IBC) and outlines the components of the UOIT Biosafety Program.

In 2015, the second edition of the Canadian Biosafety Standards (CBS) was published to replace the CBSG, 1st Edition. Any changes under this new edition have been updated in the manual.

1.0 INTRODUCTION

This manual is intended to provide information on safe work practices pertaining to biological materials that can be hazardous to the health of individuals and animals. The information and accompanying procedures describe the minimum requirements for working safely with potentially biohazardous materials to prevent laboratory acquired infections (LAIs) in laboratory workers and to prevent the release of biohazardous materials outside the laboratory that can lead to deleterious effects in an exposed population.

The main purpose of this manual is to provide guidance for understanding how to identify, evaluate, handle, and control biological agents and recombinant DNA in research activities and academic teaching laboratories. Handling of such materials requires the use of universal precautionary measures, and is dependent on the risk analysis of the agents involved and the procedures being conducted. The information serves to protect and promote research by controlling the unwanted spread of biohazardous contamination. The goal of the Biosafety Program is to identify, reduce and monitor the risks associated with potentially hazardous biological agents, and to mitigate those risks whenever possible.

1.1 UNIVERSITY HEALTH AND SAFETY POLICY

Under the provisions of the Ontario *Occupational Health and Safety Act*, the University has established a [Health and Safety Policy](#) committed to making every reasonable effort to provide and maintain a safe and healthy work and learning environment for its faculty, staff, students and visitors and to ensure the protection of the external community and the environment. The policy describes prevention of occupational illness or injury as a major continuing objective of the University. As such, the policy institutes the University as responsible for establishing specific safety policies and procedures which will ensure compliance with legislation and facilitate the maintenance of safe working conditions.

The University has a duty to its faculty, staff, students and visitors to ensure a safe and healthy research and learning environment exists for all. Implementation of these biosafety practices and procedures in research and learning is the responsibility of the Principle Investigator. The success of the Biosafety Program rests on the committed efforts of all laboratory supervisors and their staff. To assist in this implementation, services are available through the Office of Research Services and the Biosafety Officer.

1.2 BIOHAZARDOUS MATERIALS

Bacteria, viruses, fungi or other infectious agents are often studied because they cause disease. Since many of these agents are pathogenic (capable of causing disease) to humans, animals, or other forms of life, their use poses risks which vary with each agent and the way in which it is used. Accordingly, additional requirements for working with biohazardous materials are specified in the *Human Pathogens and Toxins Act*, the WHMIS regulations, requirements of the Canadian Food Inspection Agency and the Memorandum of Understanding between the Universities and Federal Granting Agencies, the Research Tri-Council. The Public Health Agency of Canada has previously published the Laboratory Biosafety Guidelines which classify microorganisms into different risk groups based on a number of factors including the severity of the disease that the microorganism causes, the routes of its infection, its virulence and infectivity, the existence of effective therapies, quantity of agent being used and whether the agent is indigenous to Canada.

This Biosafety Manual is based on the Canadian Biosafety Standards and Guidelines (CBSG), 1st Edition, 2013, the Canadian Biosafety Standards (CBS), 2nd Edition, 2015, as well as the Acts and regulations cited above and is designed to:

- 1) protect employees, students, the community and the environment from potential hazards which may arise in the handling of biological agents, and
- 2) ensure that the University complies with the requirements and guidelines of the Public Health Agency of Canada (PHAC), the National Science and Engineering Research Council (NSERC), the Canadian Institutes for Health Research (CIHR), Agriculture and Agri-Food Canada, Canadian Food Inspection Agency (CFIA) and other research sponsors and all applicable federal and provincial regulations respecting biohazardous materials.

The Controlled Products regulation defines biohazardous infectious material as an organism that has been shown to cause disease or is reasonably believed to cause disease in persons or animals and the toxins of such an organism. WHMIS assigns these materials to controlled product Class D, Division 3. Employers are required to have an inventory of all such organisms and must maintain material safety data sheets for each corresponding hazardous material.

Importation and containment standards for facilities handling **Aquatic Animal Pathogens and Plant Pests** are legislated by the *Health of Animals Act*, and the *Plant Protection Act* and their supporting regulations. CFIA has published containment standards and operational guidelines for facilities handling aquatic animal pathogens and plant pest, however this material is outside the scope of this Biosafety Manual. Information on such material can be obtained by contacting the Biosafety Officer.

This manual applies to all research and teaching laboratory activities of the University which involve the use or manipulation of potentially hazardous biological agents and materials containing such agents (including viruses, bacteria, fungi, parasites, recombinant DNA, prions and other micro-organisms/genetic systems, and human and animal tissues, cells, blood and body fluids).

All users of biohazardous materials must be familiar with and comply fully with the policies and procedures outlined in this manual.

Any questions regarding the application or interpretation of this manual should be directed to the Biosafety Officer or the Chair of the Biosafety Committee (Appendix 1).

1.3 Definitions

Authorized personnel/individual

An individual who has been granted access to the containment zone by the facility/laboratory supervisor, BSO, and/or another individual to whom this responsibility has been assigned. This is dependent on completing training requirements and demonstrating proficiency in the SOPs, as determined to be necessary by the facility.

Containment Zone

A physical area that meets the requirements for a specified containment level. Dedicated support areas, including anterooms, showers and dirty change rooms, may be part of the containment zone.

Human Pathogen

A human pathogen is a micro-organism, nucleic acid or protein capable of causing disease in humans and/or animals and falls into biohazard Risk Group 2, 3, or 4 or is listed in Schedules 2, to 4 or Part 2 of Schedule 5 of the *Human Pathogens and Toxins Act* (HPTA). (See Appendix 3 of this Manual).

This includes a substance that contains a human pathogen and any synthetic form of the human pathogen.

Toxin (Biological)

A poisonous substance that is produced or derived from a microorganism and can lead to adverse health effects in human and/or animals. Human toxins are listed in Schedule 1 or Part 1 of Schedule 5 of the *Human Pathogens and Toxins Act*. (See Appendix 3 of this Manual).

2.0 BIOSAFETY PROGRAM FRAMEWORK

2.1 Responsibilities

2.1.1 Faculty/Laboratory Supervisors

University faculty members and laboratory supervisors who are working with biohazardous materials are responsible for complying with the procedures outlined in this manual. In particular, they are responsible for applying for and complying with the conditions of a University Biosafety Certificate (see Section 2.2).

The faculty member/laboratory supervisor in charge of the laboratory is responsible for identifying known and potential biohazards specific to the laboratory and providing additional training and procedures to eliminate or minimize the risks. Personnel must be required to know, understand and follow standard practices and procedures. Training in laboratory safety must be provided and followed and competence in safe technique demonstrated before work is allowed with hazardous agents or toxins.

2.1.2 Vice-President Research, Innovation and International

Responsibility for the administration of the biosafety program lies with the Vice-President Research, Innovation and International. The Vice-President Research, Innovation and International is responsible for signing grant applications attesting to the commitment of the University to comply with the Public Health Agency of Canada's Canadian Biosafety Standards and Guidelines. The Vice-President Research, Innovation and International

shall ensure that grant funds are not released without a current and valid Biosafety Certificate.

2.1.3 The Biosafety Committee

The University Biosafety Committee is responsible for setting appropriate standards of safety for work with potentially hazardous biological agents within University workplaces, or undertaken on grants administered by the University.

All work with potentially biohazardous materials such as viruses, bacteria, fungi, parasites, recombinant DNA, prions and other micro-organisms/genetic systems, and human and animal tissues, cells, blood and body fluids must be submitted to the Biosafety Committee for review. The Committee will determine the appropriate level of containment and issue a Biosafety Certificate which details the conditions under which the work must be undertaken.

No work with biohazardous materials may be undertaken without a valid biosafety certificate issued by the Committee.

The terms of reference of the Biosafety Committee are given in Figure 2.1.

FIGURE 2.1

UNIVERSITY OF ONTARIO INSTITUTE OF TECHNOLOGY BIOSAFETY COMMITTEE TERMS OF REFERENCE

The UOIT Biosafety Committee is responsible for establishing and maintaining a system to ensure that all activities within the University involving infectious biological agents are conducted in a safe manner and in conformity with generally accepted standards. The term “infectious biological agents” includes viruses, bacteria, fungi, parasites, prions, and other micro-organisms/genetic systems that, by virtue of their replicative properties, are potentially harmful to humans and/or other living systems.

The University affirms that the primary responsibility for the safety of staff and the public lies with the faculty member/principal investigator using or authorizing the use of such agents. The University acknowledges its responsibility to provide a policy and procedural framework designed to ensure that those undertaking work under its auspices conduct the work in a safe manner and in conformance with all relevant legislation and regulations.

The UOIT Biosafety Committee is appointed by and reports to the Vice-President Research, Innovation and International. A majority of the members of the Committee shall be selected based on their knowledge and experience in working with infectious biological agents. The committee may have a minority of non-specialist members in order to provide a broader perspective in decision-making.

Duties and Functions of the Committee

1. To develop and promulgate safety standards for the conduct of research and teaching involving infectious biological agents by members of the University;
2. To specify training requirements for all personnel working with infectious biological agents;

3. To take all reasonable care to ensure that research and teaching activities of members of the University involving infectious biological agents are performed in compliance with relevant legislation and guidelines;
4. To establish and maintain a system for authorization of the use of biohazardous materials on University premises or by University faculty and staff;
5. To review all applications for a biosafety certificate, assess the risks of the work, set the appropriate level of containment and specify any additional precautions to be taken including appropriate waste disposal;
6. To review periodically the Biosafety program and procedures and revise or add new procedures as appropriate;
7. To advise the Vice-President Research, Innovation and International on any matters relating to Biosafety including the need for biosafety facilities, policies and programs.

The Chair of the Committee

1. The Chair of the Committee shall be a member of the faculty with expertise in the hazards of biological agents and containment practices and procedures.
2. The Chair is appointed by the Vice-President Research, Innovation and International on the advice of the Committee.
3. The Chair is appointed for a three year term which is renewable.
4. The Chair shall carry out the executive functions of the Committee on a day-to-day basis working closely with the Biosafety Officer.
5. The Chair will review and approve Biosafety Certificates requiring more than Level 1 containment.

Committee Membership and Operating Procedures

1. Committee members are appointed by the Vice-President Research, Innovation and International for a nominal term of three years. Members may be reappointed for additional terms.
2. The following are ex-officio members of the Committee:
 - the Director, Office of Research Services
 - the Biosafety Officer
 - the University Health and Safety Officer
 - the Ethics and Compliance Officer, Office of Research Services

Ex-officio members have no fixed term as they serve by virtue of their position within the University.

3. The Committee shall meet as often as necessary to fulfill its mandate, but not less than once every six months.
4. The Committee shall maintain minutes of its meetings with copies to the joint health and safety committee and the Vice-President Research, Innovation and International.
5. The Committee shall endeavour to reach decisions by consensus. Where no consensus is reached the Chair shall bring the matter to the attention of the Vice-President Research, Innovation and International who shall render a decision in consultation with the Chair.

2.1.4 The Biosafety Officer

The Biosafety Officer shall be a member of the Biosafety Committee, designated by the Committee to carry out the day-to-day functions of the Committee. The Biosafety Officer will review all applications for Biosafety Certificates, verify the proposed procedures and level of containment, and prepare and issue Biosafety Certificates following the appropriate level of review and approval by the Committee. The Biosafety Officer will also conduct regular compliance inspections of all biohazard laboratories.

2.1.5 Laboratory Workers

All persons working in laboratories are responsible for conducting themselves in an appropriate and safe manner, for attending required training courses, and for following the procedures set out in this manual and by the laboratory supervisor.

2.2 Biosafety Certificates

A UOIT Biosafety Certificate is required for all **research and teaching** laboratory activities which involve the use or manipulation of potentially hazardous biological agents and materials containing such agents (e.g. viruses, bacteria, fungi, parasites, recombinant DNA, prions and other microorganisms/genetic systems and human and animal tissues, cells, blood and body fluids), and which are:

- (1) supervised or conducted by employees or members of the University, or
- (2) conducted on University premises, or in a building or location administered by or under the control of the University, or
- (3) supported by funds provided by or through the University. This includes work by University researchers conducted at other institutions.

The major purpose of the Biosafety Certificate is to ensure that a risk assessment is performed for work with the particular agent, that an appropriate level of containment is selected, and that appropriate safety and security controls are in place as required by the applicable legislation and guidelines.

All faculty members/laboratory supervisors proposing to conduct research using biological agents must complete an application for a Biosafety Certificate and submit it to the Office of Research Services.

Biosafety Certificates are issued by the Biosafety Committee to individual faculty members or laboratory supervisors for work with a specific agent in a specific location. Any changes to the agent or the location require an amendment to the Certificate or the issuance of a new Certificate.

A Biosafety Certificate is normally valid for a period of one year from the date of issuance at which time it must be renewed. Renewal will be initiated by the Biosafety Officer and if there are no changes from the previous certificate it will be renewed. If there are changes proposed, then, depending on the nature of the changes, a new application may be required.

The Biosafety Committee may revoke or suspend a Certificate at any time where such action is warranted due to a serious contravention of:

- The Public Health Agency of Canada's CBS (2nd Edition),
- An applicable federal or provincial law,
- The University Biosafety Manual, or
- Any condition of approval specified by the Biosafety Committee.

The Office of Research Services will be notified when a certificate is suspended, and this may result in grant funds being frozen.

A copy of the procedures and application form are provided in Appendix 2. Forms are also available for download from the Biosafety website at:

<http://healthandsafety.uoit.ca/programs/>

If the organism being used requires Containment Level 2, the application must be accompanied by a certificate verifying that any biological safety cabinet to be used has been tested for compliance with the standards in Appendix 4 of this manual. This testing is arranged through Facilities Management. Note that containment levels 3 and 4 are not available at UOIT.

Following the review and approval process a copy of the validated Biosafety Certificate will be returned to the applicant. The certificate must be posted in the workplace covered by the Certificate. A copy of the Certificate will be placed on file in the Office of Research Services.

Application for and the holding of a Biosafety Certificate implies authorization for the Biosafety Officer or other member of the Biosafety Committee to inspect the laboratory or other area covered by the certificate at any reasonable time for purposes of verifying compliance with the terms and conditions of the certificate.

For renewal of a Certificate at Level 2, verification must be provided that any biological safety cabinet has been tested within the past 12 months.

Note that a valid Biosafety Certificate must be on file with the Office of Research Services before any grant funds will be released by the University.

Where research involving biohazards will not be undertaken immediately, but where release of funding is required to purchase equipment or for other purposes, the approval process can proceed in two stages. Application should be made for a Biosafety Certificate using the standard process but indicating that approval "in principle" is only required at this stage. Upon review of the application, the Biosafety Committee may issue a provisional Biosafety Certificate which will allow release of some funding but which will not provide specific approval for the research to begin.

In circumstances where "approval in principle" only has been given, a re-application must be made before a final certification will be made by the Committee.

2.3 Research Conducted at other Institutions

Where faculty members conduct research at another institution and funding is administered by UOIT, the research protocols must be reviewed and approved by the UOIT Biosafety Committee before funds can be released.

The purpose of this review is to ensure that the work is conducted in compliance with PHAC, CFIA and other regulatory requirements in the same manner as if it were conducted on UOIT premises.

In such cases the researcher should submit an application for a UOIT Biosafety Certificate and indicate the institution at which the research is to be carried out. A valid biosafety certificate issued by the institution at which the research is to be carried out must also be submitted with the application.

2.4 Control of Pathogens

Facilities handling infectious agents need not only a Biosafety program but also a Biosecurity plan in place. Biosafety deals with all aspects of containment to prevent any exposure to and accidental release of pathogens; biosecurity is implemented to prevent the loss, theft, misuse or intentional release of pathogens. The specific controls required for individual pathogens will depend on the risk group and the outcome of a risk assessment (see Section 6), but **common to all pathogens is the necessity to maintain accurate inventory controls from purchase through storage and use to final disposal and the need to restrict access to these organisms.**

1. A Biosafety certificate is required to purchase and use any biohazardous agent (all risk groups). All biohazardous agents must be listed on the certificate and agents can only be stored and used in rooms listed on the certificate.
2. In order to acquire any pathogen, either by purchase from a commercial supplier or by transfer from another institution the acquirer must complete and submit for approval the UOIT "[Authorization to Acquire Biohazardous Material](#)" form.
3. Certificate holders must maintain an inventory of all agents in their possession. This inventory must be maintained current and account for additions and removal of agents through use, deactivation and disposal.
4. Access to all laboratories containing biohazardous materials must be restricted to authorized individuals.
5. Access to the containment zone to be limited to authorized personnel and authorized visitors.

2.5 Training

All persons working in a biohazard laboratory shall receive basic instructions in both chemical and biological safety. This training is coordinated through the Biosafety Officer and records of training maintained in the Biosafety Office. Self-study modules in basic biosafety and laboratory chemical safety are available through faculty supervisors who will administer written tests. Researchers must provide additional training specific to the agents and manipulations being conducted in their laboratory, as well as relevant components of the Biosafety Manual/standard operating procedures that are being

followed. Records of this training should be maintained for inspection by the Biosafety Officer.

Trainees must be supervised by authorized personnel when engaging in activities with infectious material until they have fulfilled the training requirements. Visitors, maintenance/janitorial staff, contractors or others who require temporary access to the containment zone must be trained and/or accompanied in accordance with their anticipated activities inside the laboratory.

2.6 Inspections

The Biosafety Officer shall conduct regular inspections of all biohazard laboratories to verify that the conditions of the Biosafety Certificate continue to be met. Any items of non-compliance will be referred to the faculty member/laboratory supervisor for resolution and, if required, brought to the attention of the Biosafety Committee. Records of inspections shall be maintained in the Biosafety Office.

2.7 Material Safety Data Sheets

Biohazardous Infectious Materials are controlled products which fall into Class D Division 3 under the WHMIS regulation. The WHMIS regulation requires that an inventory of such materials be maintained and that a material safety data sheet (MSDS) be available for the organisms and toxins being used. MSDSs are a useful resource in selecting the appropriate containment level based on risk group classification, exposure control and personal protective equipment requirements, as well as first aid and medical surveillance information. Sources for these MSDS and other information useful for conducting a laboratory risk assessment are:

- Public Health Agency of Canada – Office of Laboratory Security
<http://www.phac-aspc.gc.ca/msds-ftss/>
- US Centres for Disease Control – National Institutes for Health
<http://www.cdc.gov/biosafety/publications/bmb15/>
- The American Type Culture Collection (ATCC) – provides data sheets and assigns risk groups or biosafety levels for the material it supplies. ATCC biosafety levels must be compared to risk groups as assigned by PHAC under the *HPTA* (see section 3).

Copies of material safety data sheets should be available in the laboratory for all organisms used within the containment zone.

3.0 LEGISLATION, STANDARDS AND GUIDELINES

3.1 Human Pathogens and Toxins Act (2009)

The [Human Pathogens and Toxins Act](#) (HPTA, Bill C-11) became law on June 23, 2009. The purpose of this Act is to establish a safety and security regime to protect the health and safety of the public against the risks posed by human pathogens and toxins. The Act is administered by the Federal Minister of Health and applies to all micro-organisms, nucleic acids or proteins that are listed in Schedules 1 to 5 to the Act or are classified as belonging to Risk Group 2, 3, or 4.

The HPTA requires that any person possessing, handling or using a human pathogen or toxin covered by the Act register with and obtain a license from the Minister of Health. The University is registered as an institution under this Act through the Office of Research Services.

3.2 Canadian Biosafety Standards and Guidelines, Public Health Agency of Canada

The Office of [Laboratory Biosafety and Security](#) of the Public Health Agency of Canada has issued the Canadian Biosafety Standards and Guidelines (CBSG), renamed the Canadian Biosafety Standards (CBS). The purpose of this document is to provide a national standard for the handling and storing of human and terrestrial animal pathogens and toxins in Canada. The CBS provides the foundation for the development of the regulatory framework to support the full implementation of the HPTA. As with the previous Laboratory Biosafety Guidelines, they have become the *de facto* standard in Canada because adherence to them is required by the Canadian Institute for Health Research (CIHR) and the Natural Science and Engineering Research Council (NSERC) as a requirement in their granting process. The guidelines also represent a standard of “due diligence” for employers.

The CBSG 1st Edition was divided into two distinct parts, Part I – The Standards and Part II – The Guidelines. The standards in Part I provided the physical containment requirements (i.e., structure and design components) and the operational practice requirements (i.e., practices to be followed by personnel). The guidelines in Part II provided guidance on how to achieve the physical and operational biosafety and biosecurity requirements outlined in Part I, and addressed the concepts required for the development and maintenance of a comprehensive risk-based biosafety management program.

Adherence to these Standards and Guidelines is also required by the Canadian Food Inspection Agency as a condition of certifying Biohazard Containment Laboratories.

3.3 Certification of Containment Laboratories

The [Public Health Agency of Canada](#) and the [Canadian Food Inspection Agency](#) require certification of all Containment Level 2, 3 and 4 laboratories. Suppliers of regulated biohazardous agents will require a copy of the certification letter before shipping the material.

The University has registered all containment level 2 laboratories with these two agencies and certification letters are available from the Biosafety Officer and the Office of Research Services. Since these certification letters are for specified rooms only, anyone requiring this documentation should contact the Biosafety Officer or Ethics and

Compliance Officer in Research Services to verify that the appropriate containment certification for the room in question is available.

3.4 Importation and Transfer of Human Pathogens

The [Human Pathogens Importation Regulations](#) (HPIR) are the regulatory authority for facilities wishing to import human pathogens into and transfer specimens within Canada. The purpose of these regulations is to ensure that facilities have appropriate containment for the pathogens they wish to handle. Any facility wishing to import human pathogens requiring containment levels 2, 3, or 4 must have a valid permit issued by the Public Health Agency of Canada prior to importation. Pathogens requiring containment level 1 facilities are not regulated by the HPIR, and therefore a permit is not required for their importation.

Regulations are currently being developed to support the HPTA which will incorporate the oversight of importation and transfer of RG2, RG3 or RG4 human pathogens and toxins as well as the activities involving both imported and domestically acquired human pathogens and toxins.

A human pathogen is any microorganism or parasite that causes disease in humans. This includes zoonotics. Human pathogens may be contained in cultures, diagnostic specimens, or tissue. Many human pathogens are also pathogenic to animals as well.

Importation permits are issued by the Office of Laboratory Security, Public Health Agency of Canada. Applications for permits to import human pathogens can be obtained by either calling directly at (613) 957-1779 or by downloading the form from their website at <http://www.phac-aspc.gc.ca/lab-bio/index-eng.php>.

The completed application form must be forwarded to the Biosafety Committee for review along with a Biosafety Certificate application form, if one has not previously been obtained.

3.5 Importation, Transfer and Containment of Animal Pathogens

The Canadian Food Inspection Agency (CFIA) continues to issue permits for animal pathogens that are not indigenous to Canada (pathogens causing foreign animal and emerging animal diseases), aquatic and plant pathogens as well as for animals, animal products and by-products, tissue, sera and blood that are infected with animal pathogens.

The *Health of Animals Act, 1990* (HAA) and the *Health of Animal Regulation* (HAR) gives the Canadian Food Inspection Agency (CFIA) the legislative authority to control the use of imported animal pathogens and pathogens associated with reportable animal diseases. These include materials of animal origin that contain potential pathogens.

The HAA and regulations apply to persons importing animal pathogens into Canada and transferring those pathogens within Canada. The HAR requires any person importing or transferring an animal pathogen to hold a valid permit for those purposes. Authority for the importation and transfer of terrestrial animal pathogens under the HAA and HAR, with the exception of non-indigenous animal pathogens and emerging animal pathogens, has been transferred to the PHAC as of April 1, 2013.

For an agent brought into Canada under an import permit which restricts its distribution, further approval must be obtained before transferring the agent to another location.

The CFIA also establishes the conditions under which animal pathogens will be maintained and work will be carried out. It is necessary to consider not only the risk to human health but also the level of containment needed to prevent escape of an animal pathogen into the environment, where it may constitute a risk to any indigenous animal species. The CFIA publication *Containment Standards for Veterinary Facilities* (<http://www.inspection.gc.ca/english/sci/bio/anima/convet/convet1-3e.shtml>) has been replaced by the *Canadian Biosafety Standards and Guidelines*, which outlines the minimum design, and physical and operational requirements for Canadian Laboratories and animal facilities that import and work with animal or zoonotic pathogens. Laboratories that apply to import animal or zoonotic pathogens must demonstrate that they meet these requirements before the CFIA can issue an import permit.

Animal pathogens, including pathogens that affect both humans and animals under the control of CFIA are listed in a database (<http://www.inspection.gc.ca/english/sci/bio/anima/disemala/disemalae.shtml>) maintained by the Biohazard Containment and Safety Division, CFIA. This is a dynamic list that is continuously amended to include emerging pathogens that may require restriction. Animal pathogens that are nonindigenous to Canada form a portion of this database and are severely restricted.

Information on the status of animal pathogens as well as the appropriate forms may be obtained from CFIA at their website <http://www.inspection.gc.ca/english/sci/bio/bioe.shtml>. In addition to the *Application for Permit to Import*, applicants must also complete the form *Facility Certification for the Importation of Animal Pathogens*. This form will require the signature of the Biosafety Officer and both forms must be sent to the Office of Research Services before submission to CFIA.

3.6 Export of Pathogens

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 and toxin weapons and their destruction. For assistance and advice on export contact the Department of Foreign Affairs and International Trade Canada, Export Control Division. Their website is http://www.international.gc.ca/controls-controles/about-a_propos/index.aspx.

3.7 Transportation of Biological Agents

Infectious substances fall under Class 6.2 of the Transportation of Dangerous Goods Act and regulations. Very specific packaging and documentation requirements must be met before such materials may be shipped from the University.

An infectious substance is defined in Part 1 of the TDG Regulations as “a substance known or reasonably believed to contain viable micro-organisms such as bacteria,

viruses, rickettsia, parasites, fungi and other agents such as prions that are known or reasonably believed to cause disease in humans or animals and that are listed in Appendix 3 to Part 2, Classification". Infectious substances are divided into two categories for shipping purposes defined by UN numbers and shipping names based on whether the infectious substance affects human only, or both humans and animals.

Information respecting the Transport of Dangerous Goods regulation can be obtained from the following websites:

<http://www.tc.gc.ca/eng/acts-regulations/menu.htm>

<http://www.tc.gc.ca/eng/tdg/clear-part2-339.htm#app3>

http://www.tc.gc.ca/media/documents/tdg-eng/RDIMS-8210418-SHIPPING_INFECTIOUS_SUBSTANCES_-_TDG_BULLETIN_FINAL.pdf

Researchers wishing to ship biological agents should contact the Biosafety Officer for advice on the proper procedures.

3.8 Laboratory Animals

Use of laboratory animals is overseen by the University Animal Care Committee and an Animal Use Protocol must be submitted to and be approved by the Committee for all uses of laboratory animals.

The use of animals for research is covered by a number of federal and provincial regulations and guidelines among which are:

- (1) Ontario Animals for Research Act (RSO 1990, c.A22)
Research Facilities and Supply Facilities Regulation (RRO 1990, ref 24)\
Transportation of Animals Regulation (RRO 1990, reg.25)
General Regulations (RRO 1990, reg 22)

Copies are available at <http://www.e-laws.gov.on.ca/>

- (2) Canadian Council for Animal Care Guides

Copies of these guides are available at <http://www.ccac.ca/>

3.9 General Occupational Health and Safety

Workplace health and safety is governed by the Ontario Occupational Health and Safety Act. Specific regulations under this act which are relevant to biosafety are:

- (1) Regulations for Industrial Establishments (O. Reg 851)
- (2) WHMS Regulation (O. Reg 860)
- (3) Control of Exposure to Biological or Chemical Agents (O. Reg 833)
- (4) Needle Safety (O. Reg 474/07)
- (5) Health Care and Residential Facilities (O. Reg 67/93)

It is important to note that infectious biological agents fall under WHMIS Class D3 and hence the requirements for worker training, labeling and material safety data sheets apply to biological agents.

3.10 Other Relevant Guidelines

There are a number of relevant guidelines published which may be consulted when determining the appropriate containment and procedures for working with specific biohazardous agents. Among these are the following:

- (1) Biosafety in Microbiological and Biomedical Laboratories (5th Edition), US Centres for Disease Control:
<http://www.cdc.gov/biosafety/publications/bmbI5/index.htm>
- (2) Laboratory Biosafety Manual (2nd Edition), World Health Organization
<http://www.who.int/csr/resources/publications/biosafety/Labbiosafety.pdf>
- (3) NIH Guidelines for Research Involving Recombinant DNA Molecules, US National Institutes of Health,
http://oba.od.nih.gov/oba/rac/Guidelines/NIH_Guidelines_prn.pdf

4.0 RISK GROUPS

The Canadian Biosafety Standards and Guidelines classify biological agents into four risk groups based on factors such as the severity of the disease they cause, the routes of infection, their virulence and infectivity. These classifications presume ordinary circumstances in the research laboratory, or growth in small volumes for diagnostic or research purposes.

Factors considered in a risk assessment to determine the risk group include:

- *Pathogenicity/Virulence*: Is the pathogen able to infect and cause disease in humans or animals (i.e., pathogenicity)? What is the degree of disease severity (i.e., virulence)?
- *Route of Infection*: How does the pathogen gain entry into the host (i.e., ingestion, inhalation, mucous membranes, subcutaneous or injection)?
- *Mode of Transmission*: How does the pathogen travel to the host (e.g., direct contact, indirect contact, casual contact, aerosolized droplet or airborne transmission, vectors, zoonosis, intermediate host)?
- *Survival in the Environment*: How stable is the pathogen outside the host? Under what environmental conditions can it survive and for how long?
- *Infectious Dose*: What amount of pathogen is required to cause an infection in the host?
- *Availability of Effective Preventative and Therapeutic Treatments*: Are effective preventative measures available (e.g., vaccines)? Are effective treatments available (e.g., antibiotics, antivirals)?
- *Host Range*: What are the primary, intermediate, and dead-end hosts? Does the pathogen cause infection in a wide range of species, or is the host range more restricted?

The CBSG does not provide lists of pathogens categorized according to risk group. This has been done in order to allow for the addition of new and emerging pathogens and for an ongoing assessment of risk. The current list of human pathogens presented by Risk Group can be found in Schedules 1 to 5 of the HPTA or by calling them at (613) 957-1779.

RISK GROUP 2 HUMAN PATHOGENS applicable to UOIT laboratories: <http://lois-laws.justice.gc.ca/eng/acts/H-5.67/page-22.html>

In order to facilitate local risk assessments for users within the University, this manual contains, in the Appendices, lists of common agents categorized according to risk group. For agents not on these lists, the researcher should contact the Health Canada Office of Laboratory Security directly to obtain the classification.

4.1 Risk Group 1 (low individual and community risk)

Any biological agent that is unlikely to cause disease in healthy workers or animals. See Table A3.1. RG1 pathogens can be opportunistic and may pose a threat to immunocompromised individuals. RG1 pathogens are not regulated by the PHAC or the CFIA due to the low risk to public health, livestock or poultry. Nonetheless, due care should be exercised and safe work practices (e.g., good microbiological techniques) should be followed when handling these materials.

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, public health, livestock, or the

environment. Laboratory exposures rarely cause infection leading to serious disease, but effective treatment and preventive measures are available, and the risk of spread is limited. See Appendix 3.

4.3 Risk Group 3 (high individual risk, low community risk)

Any pathogen that is likely to cause serious disease in humans or animals. Effective treatment and preventive measures are usually available and the risk of spread of disease is low for the public. See Appendix 3.

4.4 Risk Group 4 (high individual risk, high community risk)

Any pathogen that usually produces very serious human and/or animal disease, and may be readily transmitted from one individual to another, or from animal to human or vice-versa directly or indirectly, or by causal contact. Effective treatment and preventive measures are not usually available. See Appendix 3.

In previous versions of the Laboratory Biosafety Guidelines, four levels of containment were specified corresponding to each of the four risk groups. In the current guidelines, four levels of containment are described, however the assignment of a containment level to any particular organism is based on a risk assessment, of which the Risk Group forms only one component. The requirements of the four containment levels are described in the next section.

5.0 CONTAINMENT LEVELS

There are four levels of containment described in the Canadian Biosafety Standards and Guidelines (2013). Containment levels are selected to provide the end-user with a description of the minimum physical containment and operational practices required for handling infectious materials or toxins safely in a laboratory and animal work environments. The containment system includes the engineering, physical, operational and technical requirements for manipulating a particular pathogen.

In previous editions of the Guidelines, the four containment levels directly corresponded to the four risk groups. This association is still in most cases appropriate, however the current guidelines provide for altering the containment level as the result of a local risk assessment. Although the risk group is the major factor in determining the containment level, other factors such as the manipulations and procedures that are being performed may provide justification for either raising or lowering the level of containment.

As a starting point, or default, the containment level should correspond to the risk group. That is, a risk group 1 agent would be assigned a containment level of 1 and a risk group 2 agent would be assigned a containment level of 2, etc. Where a researcher wishes to propose a level of containment different from that corresponding to the risk group, this must be justified on the basis of a local risk assessment (see Section 6) and appropriate documentation must accompany the request for a Biosafety Certificate.

One of the purposes of the Biosafety Certificate is to review the organism and the specific procedures to verify the containment level and to ensure that the appropriate level is being used. The Biosafety Committee may approve an increase or decrease in the level of containment based on their assessment of the risks involved.

The four containment levels are as follows:

5.1 Containment Level 1 (CL1)

Containment Level 1 requires no special design features beyond those suitable for a well-designed and functional laboratory. Biological safety cabinets 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 microbiological laboratory such as good microbiological technique, proper training, appropriate use of personal protective equipment and keeping work surfaces clean.

5.2 Containment Level 2 (CL2)

The primary exposure hazards associated with organisms requiring containment level 2 are through the ingestion, inoculation and mucous membrane route. Agents requiring level 2 containment are not generally transmitted by airborne routes, but care must be taken to avoid the generation of aerosols or splashes. Aerosols can settle on bench tops and become an ingestion hazard through contamination of the hands.

Primary physical containment devices such as biological safety cabinets and centrifuges with sealed rotors or safety cups are to be used, as well, environmental contamination must be minimized by the use of handwashing sinks and decontamination facilities (autoclaves). Operational practices for CL2 include administrative controls (e.g., biosafety program management, training) and procedures (e.g., work practices, PPE use, and

decontamination) that mitigate the risks associated with the activities conducted within the containment zone.

5.3 Containment Level 3 (CL3)

Agents requiring containment level 3 may be transmitted by the airborne route, often have a low infectious dose to produce effects and cause serious life-threatening disease.

Containment level 3 emphasizes additional primary and secondary barriers to minimize the release of infectious organisms into the immediate laboratory and the environment. Additional features to prevent transmission of organisms are appropriate respiratory protection, HEPA filtration of exhausted laboratory air and strictly controlled laboratory access.

5.4 Containment Level 4 (CL4)

Agents requiring level 4 containment have the potential for aerosol transmission, often have a low infectious dose and produce very serious and often fatal disease; there is generally no treatment or vaccine available.

Level 4 is the maximum containment available and represents an isolated unit, functionally, and when necessary, structurally independent of other areas. Containment level 4 emphasizes maximum containment of the infectious agent by complete sealing of the facility perimeter with confirmation by pressure decay testing, isolation of the researcher from the pathogen by his or her containment in a positive pressure suit or containment of the pathogen in a Class III biological safety cabinet line, and decontamination of air and other effluents produced by the facility.

A complete description of the requirements for the four levels of containment is given in the [CBSG](#). The main physical requirements for containment Levels 1 and 2 are summarized in Table 5.1. UOIT does not have Level 3 or 4 facilities.

TABLE 5.1

PHYSICAL CONTAINMENT REQUIREMENTS FOR LEVELS 1 AND 2

| Matrix | Requirement [†] | Level 1* | CL2 |
|--------|---|-------------|-----------|
| | <u>Structure and Location</u> | | |
| 3.1.1 | Containment zones, animal rooms/cubicles, PM rooms, and associated corridors to be separated from public and administrative areas by a door. | Recommended | Required |
| 3.1.2 | Dedicated paper/computer work stations within the containment zone to be separated from laboratory work stations and animal rooms/cubicles. | Recommended | Required |
| 3.1.6 | Cage washing areas to be provided for SA zone. | N/A | Required |
| | <u>Containment Barrier</u> | | |
| 3.2.1 | Openable windows positioned on the containment barrier are to include effective pest control and security. | Required | Required |
| 3.2.4 | Windows on the containment barrier to be positioned to prevent viewing into animal rooms/cubicles from the public. | N/A | Required |
| | | | |
| | <u>Access</u> | | |
| 3.3.1 | Doors to the containment zone to be lockable. | Recommended | Required |
| 3.3.2 | Biohazard warning signage (including the international biohazard warning symbol, containment level, name and telephone numbers of contact person, and entry requirements) to be posted at the containment zone point(s) of entry. | N/A | Required |
| 3.3.3 | Where unique hazards exist, project-specific signage to be posted at the animal room/cubicle and PM room point(s) of entry. | N/A | Required |
| 3.3.11 | Space to be provided at the containment zone point(s) of entry for the storage of personal protective equipment (PPE). | N/A | Required |
| | Access limited to authorized persons | N/A | Required* |
| | Size of door openings to allow passage of all anticipated equipment | Required | Required* |
| | (this does not apply to areas within the containment laboratory) | | Required |

| <u>Surface Finishes and Casework</u> | | | |
|---|--|-------------|----------|
| 3.4.1 | Doors, frames, casework bench tops and laboratory furniture (e.g., stools, chairs) to be constructed from non-absorbent materials. Wood surfaces are permitted in CL2 laboratory work areas if sealed to be non-absorbent. | N/A | Required |
| 3.4.2 | Surfaces and interior coatings to be cleanable and resistance to scratches, stains, moisture, chemicals, heat, impact, repeated decontamination, and high pressure washing, in accordance with function. | Recommended | Required |
| 3.4.6 | Floors to be slip-resistant in accordance with function. | N/A | Required |
| 3.4.7 | Floors in animal rooms/cubicles, PM rooms, and corridors to withstand loading consistent with use. | N/A | Required |
| <u>Air Handling</u> | | | |
| 3.5.1 | HVAC system to provide sufficient air changes per hour (AC/hr) under normal operation to maintain airflow, based on facility function. | N/A | Required |
| <u>Facility Services</u> | | | |
| 3.6.4 | Handwashing sinks to be provided and located as close as possible to the point(s) of exit of the containment zone, animal room/cubicle and PM room. | Required | Required |
| 3.6.6 | Emergency eyewash and shower equipment to be provided in accordance with containment zone activities. | Recommended | Required |
| 3.6.7 | Containment zone to be designed to control the release of large scale process fluids into sanitary sewers. | Recommended | Required |
| <u>Essential Biosafety Equipment</u> | | | |
| 3.7.1 | Certified Biosafety Cabinets (BSCs) and other primary containment devices to be provided, as determined by a Lab Risk Assessment (LRA). | N/A | Required |
| 3.7.2 | Class II B2 Cabinets to be installed and set-up in a manner to minimize reversal of airflow from the face of the BSC (i.e., puff-back) during HVAS system failure. | N/A | Required |
| 3.7.3 | Process equipment, closed systems, and other primary containment devices to be designed to prevent the release of infectious material or toxins. | N/A | Required |
| 3.7.4 | Process equipment for large scale activities with infectious material or toxins to be equipped with | N/A | Required |

| | | | |
|--------|--|-----|----------|
| | sensing devices to monitor containment integrity during operations and alarm to signal failure. | | |
| 3.7.5 | BSCs, when present, to be located away from high traffic areas, doors, windows, and air supply/exhaust diffusers. | N/A | Required |
| 3.7.7 | Large reusable equipment for large scale activities with infectious material or toxins to be designed and constructed to be effectively cleaned, decontaminated, and/or sterilized in situ or in a manner that reduces personnel exposure. | N/A | Required |
| 3.7.8 | Primary containment caging to be provided for animal work in SA zones. | N/A | Required |
| 3.7.10 | Animal cages and cubicles to be designed to prevent animal escape. | N/A | Required |
| 3.7.11 | Technologies for the decontamination of contaminated materials to be provided within the containment zone, unless procedures are in place to transport waste securely out of the containment zone to an appropriate decontamination area. | N/A | Required |
| 3.7.13 | Decontamination technologies to be provided with monitoring and recording devices to capture operational parameters. | N/A | Required |
| 3.7.16 | Vacuum systems to be equipped with a device to prevent internal contamination. | N/A | Required |
| 3.7.18 | A communication system to be provided between the laboratory work areas/animal rooms/cubicles/large scale production areas and outside the containment zone. | N/A | Required |

†Abbreviations available in the CBSG, 1st Edition.

* Based on The Laboratory Biosafety Guidelines, 3rd Edition, 2004 (PHAC).

6.0 RISK ASSESSMENT

Risk assessments provide a top-down review of the biohazards used in laboratories by analyzing possible exposure scenarios to identify important biosafety issues when developing a biosafety program and risk mitigation strategies. An overarching risk assessment process is a broad assessment that supports the biosafety program and may be supported by local risk assessments (LRAs), which examine specific elements of the program. Risk assessments are conducted to ensure that the mitigation strategies are commensurate with the level of risk which may include the use of engineering and administrative controls, practices and procedures, and training. A comparison should be made with requirements outlined by regulatory authorities and prescribed documents as well as existing best practices to clearly identify gaps that need to be addressed. The assessments also consider both the characteristics of the microorganisms and the work that will be conducted to determine the containment facilities and practices needed for the work.

6.1 General

The Biosafety Certificate holder (faculty member or laboratory supervisor) is responsible for assessing the risks of an agent and proposing a biocontainment level for the work. Note that the Biosafety Committee will make the final determination of the appropriate containment level.

In addition to the risk group classification of the organism, the following factors should be taken into account in setting the containment level:

- Potential for aerosol generation
- Quantity of material
- Concentration
- Stability of the agent in the environment
- Type of work proposed (e.g. *in vitro*, *in vivo*, aerosol challenge studies)
- Use of recombinant organisms (e.g. gene coding for virulence factors or toxins, host range alteration, oncogenicity, replication capacity, capability to revert to wild type)

Since the containment level includes consideration of both physical containment and operational procedures, it is quite possible that a risk assessment could lead to using a laboratory meeting the physical requirements of level 2 but with level 3 operational procedures. The risk assessment would be used to justify such a change.

In addition to consulting with the Biosafety Officer and the Biosafety Committee, information to assist in conducting a risk assessment can be found in the following:

- Canadian Biosafety Standards and Guidelines, Public Health Agency of Canada, First Edition, 2014
<http://canadianbiosafetystandards.collaboration.gc.ca/cbsg-nldcb/index-eng.php?page=1>
- Biosafety in Microbiological and Biomedical Laboratories, U.S Department of Health and Human Services, Centers for Disease Control and Prevention and National Institutes of Health, Fourth Edition, 1999.
<http://www.cdc.gov/biosafety/publications/bmbl5/index.htm>

Any proposed change from the containment level which corresponds to the risk group, will require the approval of the Biosafety Committee.

6.2 Large Scale Operations

Large scale operations generally refer to volumes of biological agents manipulated in a single volume in excess of 10 L. Because of the significant quantity of infectious material being handled, the CBSG specifies special containment and operational requirements for such operations. In this case, a detailed risk assessment is required and the special requirements of the CBSG must be followed (consult the Biosafety Officer). The 10 L quantity cut-off between laboratory and large scale is a guideline only and in some cases, the additional large-scale precautions should be taken at volumes below 10 L.

6.3 Medical Surveillance and Immunization

A risk assessment should also review the need for any medical surveillance and immunization of laboratory workers. This assessment should be performed in conjunction with the Biosafety Officer and health professionals. A pre-placement medical surveillance may be necessary for new workers prior to commencing activities with human or animal pathogens or toxins. The primary purpose of medical surveillance is to assess the initial health status of the individual and identify any underlying medical conditions that may increase the risk of harm associated with the anticipated work. Immunocompromised individuals (e.g., through radiation therapy or chemotherapy, pregnancy, diabetes, or other conditions) may be susceptible to infections, or experience more severe illness if they contract an infection following exposure to a pathogen. Before commencing work, these individuals should be informed of any preventative measures available against the infectious material or toxins, the risks and benefits of treatments, as well as the early signs and symptoms of a possible infection. They should also be informed of the post-exposure response plan which should include the procedure to follow in the event of potential exposure, including first aid measures, treatment and incident reporting. The following immunizations are commonly available:

- Diphtheria
- Hepatitis A
- Hepatitis B
- Influenza
- Measles
- Meningococcus
- Mumps
- Pertussis
- Pneumococcus
- Polio
- Rubella
- Tetanus
- Tuberculosis
- Varicella

Other agents for which vaccinations may be appropriate are:

- Anthrax
- Botulism

- Cholera
- Japanese encephalitis
- Lyme disease

- Plague
- Pneumococcus
- Rabies
- Typhoid
- Vaccinia
- Yellow Fever

The Biosafety Committee will, as part of the review of any application for a biosafety certificate, review the need for or advisability of medical surveillance and/or immunization.

6.4 Biosecurity

In today's world facilities handling infectious agents need not only a Biosafety program but also a biosecurity plan in place. While Biosafety deals with all aspects of containment to prevent exposure to and accidental release of pathogens, biosecurity is implemented to prevent the theft, misuse or intentional release of pathogens.

The risk assessment is a primary component of a biosecurity plan. The risk assessment should, based on the organism, review the potential threats, outline the steps to maintain security of biological agents, and determine countermeasures or mitigation strategies.

The Biosafety Officer and Biosafety Committee will review the biosafety security measures as part of the approval and issuance of a biosafety certificate.

A comprehensive biosecurity plan addresses the following elements:

- *Physical Security* – adequate physical security should be in place to minimize opportunities for the unauthorized entry of individuals into containment zones and the unauthorized removal of infectious material or toxins from the facility. Access to the containment zone is limited to authorized personnel. Entry by trainees, visitors, maintenance staff and emergency response personnel is addressed on a case-by-case basis.
- *Personal Suitability and Reliability* – personal suitability and reliability policies and procedures should define and document the training experience, competency, and suitability requirements for personnel who handle or have access to infectious materials or toxins. Personal suitability requirements for working in a biohazard lab include completion of the biosafety and WMHIS training, and completion of any work specific training that outlines the lab operating procedures specific to the hazard, pathogen or equipment used. Personal reliability requirements must be satisfied by initial pre-screening interviews conducted by the lab supervisor or biosafety certificate holder.
- *Infectious Material and Toxin Accountability* – infectious materials and toxin accountability procedures should be established in order to track and document infectious material and toxins within the institution/organization by way of an inventory system, so that material can be located when necessary and missing items can be identified more readily.
- *Incident and Emergency Response* – elements of a biosecurity plan should be integrated into the overall biosafety program and emergency response plan (ERP). All incidents and accidents involving biohazardous agents should be

- reported. Incidents, such as missing infectious materials or toxins or unauthorized entry, should be reported, documented and investigated.
- *Information Security* – information security policies should be created to protect sensitive information from unauthorized access and ensure the appropriate level of confidentiality. Sensitive information may include facility security plans, access codes, passwords, infectious material and toxin inventories and storage locations.

7.0 LABORATORY SAFETY PRACTICES AND PROCEDURES

7.1 Laboratory Acquired Infections

Individuals who work in a laboratory that handles infectious substances are at risk of exposure to these substances. Laboratory-acquired infections are not uncommon – over 5000 cases and 204 deaths had been reported from 1930-2004 (Canadian Biosafety Standards and Guidelines, 2013).

Infection can occur in a number of ways, the most common of which are detailed below.

Accidental Inoculation

Infection may arise as the result of pricking or cutting the skin with infected instruments or objects such as hypodermic needles, scalpels and broken, contaminate glassware. Care must always be exercised when working with needles and other sharps. Bites and scratches from laboratory animals are another potential route of exposure.

Inhalation

Aerosols, fine droplets of a liquid, can remain suspended in air for some time and be transferred from one room to another through air currents and the ventilation system. They can be inhaled or settle out on surfaces where they can contaminate hands and clothing.

Ingestion

Microorganisms can be ingested as a result of mouth pipetting, eating, drinking or smoking. Such practices are therefore forbidden in a laboratory.

Skin, Face and Eyes

Material on contaminated surfaces can be transferred to the hands and from there to the mouth, face and eyes. There is also the potential for spread to non-laboratory areas.

7.2 General Laboratory Safety Practices

The following requirements, taken from the Laboratory Biosafety Guidelines, 2004, the Canadian Biosafety Standards and Guidelines, 2013, and other best practice guidelines referenced in section 11, are required for all laboratories handling infectious or toxic agents:

- (1) Each laboratory must prepare and maintain a detailed procedures manual which is made available to all staff. This manual should reference the University Biosafety manual, but contain procedures specific to the laboratory. The manual must be reviewed and updated at least annually.
- (2) All laboratory personnel and others whose work requires them to enter the laboratory must understand the biological and other hazards with which they will come in contact through their normal work in the laboratory and be trained in appropriate safety precautions and procedures. The University Biosafety Manual

laboratory personnel must be familiar with its contents and follow the procedures outlined in it. The faculty member/principal investigator in charge of the laboratory is responsible for identifying known and potential biohazards specific to the laboratory and providing additional training and procedures to eliminate or minimize the risks. Personnel must be required to know, understand and follow standard practices and procedures. Training in laboratory safety must be provided and followed and competence in safe technique demonstrated before work is allowed with hazardous agents or toxins.

- (3) Eating, drinking, smoking, storing food, personal belongings or utensils, applying cosmetics, and inserting or removing contact lenses are not permitted in any active laboratory work areas within the containment zone. Contact lenses should be used only when other forms of corrective eyewear are not suitable. The wearing of jewelry should be discouraged in the laboratory.
- (4) Oral pipetting of any substance is prohibited in any laboratory.
- (5) Long hair must be tied back or restrained so that it cannot come into contact with hands, specimens, containers or equipment.
- (6) Access to the containment zone and support areas must be limited to authorized personnel.
- (7) Doors to laboratories must not be left open.
- (8) Open wounds, cuts, scratches and grazes should be covered with waterproof dressings.
- (9) The laboratory must be kept neat, orderly and clean. 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 biohazardous materials and active work areas in the containment zone.
- (10) Protective laboratory clothing, properly fastened, must be worn by all personnel while 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 appropriate to the hazard must be used.
- (12) Gloves must be worn for all procedures that might involve direct skin contact with biohazardous material or infected animals. Gloves should be properly removed before leaving the laboratory and decontaminated with other laboratory wastes before disposal. When working inside the biological safety cabinet, gloves should be removed while inside the cabinet. Latex gloves, particularly with powder, have been known to cause or exacerbate latex allergies. Non-powdered gloves are recommended and staff should be advised to report any skin rashes or other symptoms which could be associated with the glove material.

- (13) Protective laboratory clothing must not be worn in non-laboratory 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 available 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 biological safety cabinet. Needles should not be bent, sheared, recapped, or removed from the syringe, but should be promptly placed in a puncture-resistant sharps container (in accordance with CSA Standard Z316.6-95(R2000) 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 labeled or tagged-out as such.
- (19) Efficacy monitoring of autoclaves used for decontamination with biological indicators must be done regularly and the records of these results and cycle logs (i.e. time, temperature and pressure) must 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.
- (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). A cart should be used when transporting biohazardous waste to the autoclave.
- (23) Spills, accidents, or exposures to infectious materials and losses of containment must be reported immediately to the laboratory supervisor and to Research Services. 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.

7.3 Additional Requirements for Level 2 Containment Facilities

In addition to the general practices in Section 7.2, Level 2 containment facilities require the following.

- (1) All procedures must be performed in a manner that minimizes the creation of aerosols and avoids the release of infectious agents.
- (2) Biological Safety Cabinets must be used for procedures that may produce infectious aerosols and that involve high concentrations or large volumes of biohazardous material. Laboratory supervisors should perform a risk assessment to determine which procedures and what concentrations and volumes necessitate the use of a biological safety cabinet.
- (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 are 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 or other approved personnel.
- (5) All persons working in the containment zone must be trained in and follow the operational protocols for the project in progress. Trainees must be accompanied by a trained staff member. Visitors, maintenance staff, custodial 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) When drawing a vacuum using water aspirators or other such equipment, a HEPA filter should be placed in the exhaust air line to trap any aerosolized organisms.
- (7) Emergency procedures for spill clean-up, biological safety cabinet 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.

7.4 Additional Requirements for Level 3 and 4 Containment Facilities

There are no Level 3 and 4 containment facilities at the UOIT. For information on additional requirements for these facilities consult the Canadian Biosafety Standards and Guidelines, 1st Edition (Heath Canada, 2013).

7.5 Human Pathogens

Human body fluids and tissues can be a potential source of pathogenic micro-organisms that may present a risk to workers who are exposed during the performance of their duties. The pathogens of primary concern are the human immunodeficiency virus (HIV), hepatitis B virus (HBV) and hepatitis C virus (HCV). The latter two are designated as Risk Group 2 pathogens and can be handled at Level 2 containment. HIV is a Risk Group 3 pathogen and samples known to contain HIV should not be handled without a detailed risk assessment and the specific approval of the Biosafety Committee.

Laboratories dealing with human body fluids that have not been screened for infectious agents should adopt Level 2 containment practices and consult the appropriate [Public Health Agency of Canada](#) guidelines on infection control in health care facilities.

7.6 Laboratory Animals

Working with laboratory animals poses a variety of unique hazards including exposure to infectious agents, animal bites and scratches, allergies, and physical hazards such as noise and temperature. Animal facilities have specific design requirements set by the Canadian Council on Animal Care and the Canadian Food Inspection Agency.

At the present time the University does not have separate animal facilities and specific requirements for work with animals will be developed on a case-by-case basis. Researchers wishing to conduct work with animals should consult the Compliance Office, Research Services, and the Animal Care Committee.

7.7 Recombinant DNA and Genetic Manipulation

All work with recombinant DNA and Genetic Manipulation requires a documented risk assessment in order to set the appropriate level of containment and operational controls. Guidance in performing this assessment is provided in the *Canadian Biosafety Standards and Guidelines*.

7.8 Cell Lines

All work with cell lines requires a documented risk assessment as to the level of hazard associated with the particular line in order to set the appropriate level of containment and operational controls.

There have been cases of laboratory acquired infections reported as a result of manipulation of primary cell cultures. Although cell lines do not inherently pose a risk to individuals manipulating them in laboratories, they do have the potential to become contaminated with adventitious pathogenic organisms such as bacteria, fungi, mycoplasma, viruses, prions or recombinant virions.

Cell lines that are known or potentially contaminated should be manipulated at the containment level appropriate for the contaminating organism of the highest risk.

Guidance for performing the risk assessment is provided in the [Canadian Biosafety Standards and Guidelines](#) and in the [Swiss Expert Committee Recommendations on the Safe Handling of Human and Animal Cells and Cell Cultures](#).

7.9 Shipping and Handling of Infectious Materials and Toxins

Materials known to contain or suspected of containing infectious material or toxins must be packaged and labelled in accordance with national and international regulations when being transported, or being offered to a commercial carrier for transport. These regulations provide details on the packaging, documentation and certification requirements of each package and shipment designed to ensure the safe shipment of such materials in order to protect the public, shipping and receiving personnel, transportation workers, commercial carriers, and emergency responders. Infectious substances are classified as D3 under the WHMIS regulation and as Class 6, Toxic and Infectious substances under the Transportation of Dangerous Goods Regulations (TDGR).

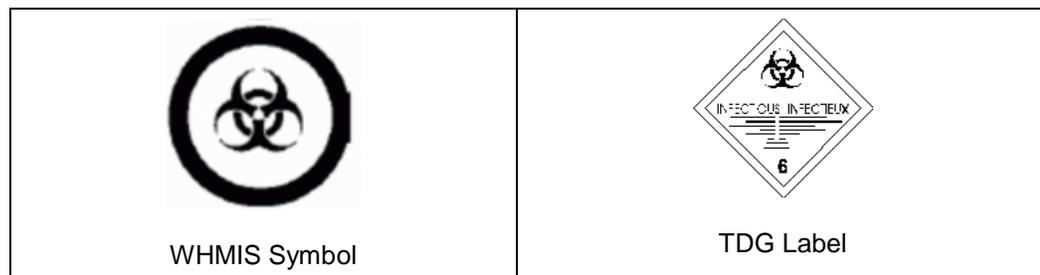
Anyone shipping infectious substances must have received training and hold a valid TDG certificate covering transport of biohazardous materials.

Transportation Between Buildings

Organizations such as universities and colleges that have several buildings containing a multitude of containment zones may need to transport infectious material or toxins between buildings or between rooms. In addition to the inventory and transfer document requirements, infectious material and toxins must be packaged in an appropriate manner to protect against their release during movement or transport, and in accordance with TDGR. Materials must be transferred via containers or buckets and within autoclavable trays or pans of sufficient capacity to contain all the material in the event of a spill en route. Containers must be placed on a trolley or cart while in transit between buildings or rooms and kept under the supervision of an authorized users at all times.

Receiving Biohazardous Materials

Packages containing such materials will bear the following labels bearing the biohazard symbol.



- (1) Packages bearing these markings must only be opened in the appropriate biohazard containment laboratory and only by appropriately trained personnel.
- (2) Receiving Department should immediately notify the consignee that a package has arrived and set the package aside in a secure area awaiting pickup by the consignee. The package must not be opened in the receiving area.

8.0 DECONTAMINATION

A basic principle of biosafety is that all contaminated materials be decontaminated prior to disposal. Decontamination includes both sterilization, the complete destruction of all microorganisms including bacterial spores, and disinfection, the destruction of specific types of microorganisms. These are discussed below.

8.1 Autoclaving

Sterilization is complete destruction or elimination of the pathogenic, reproductive or infective potential of a biological agent. Sterilization is the most effective method of eliminating the risks associated with biohazardous materials.

The use of saturated steam under pressure (autoclaving) is the sterilization method most often used in hospitals and research laboratories. Autoclaving is the most dependable procedure for ensuring the complete destruction of microorganisms. It generally involves heating in a chamber employing saturated steam under a pressure of 103kPa (15 psi) to achieve a chamber temperature of at least 121 C for a minimum of 15 minutes. The time is measured after the temperature of the material being sterilized reaches 121 C.

The most critical factor involved in steam sterilization, other than reaching the desired temperature for the correct time, is the prevention of the entrapment of air that is not displaced by steam. The materials being sterilized must come into contact with steam and heat for actual sterilization to result.

There are two types of autoclave – prevacuum autoclaves where air is removed from the chamber by pulling a vacuum before the saturated steam enters the chamber, and gravity displacement where there is no initial vacuum and the air is removed by displacement by the steam. The former is preferable as it eliminates the problem of entrapment of air in the chamber

The effectiveness of steam autoclaving depends upon various loading factors that influence the temperature to which the material is subjected and the contact time. Particular attention must be paid to packaging, including the size of the containers and their distribution in the autoclave. Containers must have good steam permeability and must be arranged in the autoclave in a manner that permits free circulation of steam. Tight fitting containers do not permit steam penetration. Piling containers above one another and overloading can result in decontamination failure.

Effective operating parameters for autoclaves should be established by developing standard loads and their processing times through the use of thermocouples and biological indicators placed at the centre of the load which will be the most difficult to reach by the steam and heat. A biological indicator is a standardized population of bacterial spores intended to demonstrate favourable sterilization conditions in the load. A typical indicator is *Bacillus stearothermophilus* spores. After the sterilization cycle has occurred, a non-growth of the spore strip indicates the complete destruction of all organisms in the materials.

There are other forms of indicators which can also be used; these involve chemical indicators. “Autoclave Tape” is an example where the white tape changes colour and displays the word “AUTOCLAVED”. It is important to ensure that the change in colour only occurs when the desired temperature has been maintained for the minimum time of 30 minutes.

All three types of indicators should be employed according to the following regime:

- (1) On initial commissioning of the autoclave the load characteristics, cycle time and temperature should be verified using *Bacillus stearothermophilus* spores and a chemical indicator such as "Autoclave Tape".
- (2) Written operating procedures should document the loading procedure, the load characteristics, and the temperature and cycle times which provide complete sterilization.
- (3) A chemical indicator should be used each time the autoclave is used to verify that sterilization has occurred.
- (4) The temperature, cycle time and results of the chemical indicator should be recorded in a log book each time the autoclave is used.
- (5) The autoclave should be verified using *Bacillus stearothermophilus* spores weekly. This should be performed on the first waste run each week and the results recorded in the log book.

8.2 Chemical Disinfection

Disinfection is the reduction of many or all disease causing microorganism and the destruction of pathogenic microorganisms in or on a surface or object so that they are no longer considered to be capable of transmitting disease. Many chemical disinfectants are available on the market. When choosing a particular product it is important to consider a number of factors that influence the effectiveness of any decontamination. These factors are:

- (1) the concentration of the chemical being used,
- (2) the presence of organic material,
- (3) the amount of time required for the chemical to be effective,
- (4) the temperature and pH at which the chemical is effective,
- (5) the level of contamination involved,
- (6) the type of contamination involved,
- (7) the physical characteristics of the contaminated surface/object.

Microorganisms vary in their susceptibility to the action of chemical agents. Generally, vegetative bacteria are the most susceptible. Then in order of increasing resistance are: lipid containing viruses, fungi, non-lipid viruses, *Mycobacterium tuberculosis* and bacterial spores. The latter tend to be the most difficult to inactivate. There are some exceptions to this general order. These include the spongiform encephalopathy viruses which may be more resistant than bacterial spores and some vegetative bacteria such as *Pseudomonas*, which are very resistant to some decontaminants.

There is very little definitive information available on the ability of chemical agents to "inactivate" nucleic acids. It is known that RNA and DNA preparations often can withstand exposure to adverse conditions better than the intact cells or microorganisms from which they were derived.

The ideal situation would be to have a broad spectrum chemical agent able to act effectively against all biohazards. Unfortunately there is no such ideal agent and one must choose among a number of products. It is therefore very important to know both the characteristics of both the chemical agent and the biological agent involved to ensure an effective decontamination procedure. A list of disinfectants for common biological agents can be found in Appendix 5.

9.0 WASTE DISPOSAL PROCEDURES

9.1 General

Waste disposal requires well defined procedures to prevent exposure to hazardous materials. Improper disposal of sharps and needles, glass and biohazardous waste puts waste handlers at risk and jeopardizes the University's access to municipal waste transfer facilities.

Materials contaminated with hazardous biological agents must be collected in the appropriate containers labeled as "biohazardous waste" and sterilized or disinfected prior to disposal. After sterilization or disinfection all biohazard labels must be removed or defaced.

The following general principles must be followed with respect to the generation and disposal of hazardous waste:

- (1) Minimize the generation of hazardous waste by reducing, reusing and recycling where possible.
- (2) Segregate biohazardous and non-biohazardous waste at the point of generation.
- (3) All biohazardous waste must be rendered harmless by either chemical decontamination or heat sterilization before disposal.
- (4) Steam sterilization is generally not recommended for laboratory waste contaminated with or containing a combination of viable **biological** agents and significant amounts of hazardous **chemical** or **radioactive** materials.

Biological waste includes:

- liquids such as used cell culturing media, supernatant, blood or blood fractions (serum), etc., which contain viable biological agents;
- materials considered pathological, including any part of the human body, tissues and bodily fluids, but excluding fluids, extracted teeth, hair, nail clippings and the like that are not infectious;
- any part of an animal infected [or suspected to be infected] with a communicable disease;
- non-sharp, solid laboratory waste (empty plastic cell culture flasks and petri dishes, empty plastic tubes, gloves, wrappers, absorbent tissues, etc.) which may be, or is known to be, contaminated with viable biological agents;
- all sharp and pointed items used in medical care, diagnosis, and research, including the manipulation and care of laboratory animals, which should be considered potentially infectious;
- laboratory glassware which is known or suspected to be contaminated with hazardous biological agents.

9.2 Liquids containing Biohazardous Agents

- Collect liquids in leak-proof containers such as flasks or bottles.
- Liquid waste containers designed to withstand autoclaving temperatures must be used when steam sterilization is utilized. To allow pressure equalization, they **should not be** sealed.

- Containers of liquid waste must be placed into an autoclavable tray or pan of sufficient capacity to contain all liquid in the event of vessel failure or breakage inside the autoclave chamber. Use extreme caution when handling autoclaved liquids since they are hot and may boil over.
- Following steam sterilization or chemical disinfection, innocuous liquids may be disposed of via the laboratory drainage system. Flush with sufficient clean water to purge the drain immediately after disposal of all liquids.

NOTE: If the liquid contains hazardous chemicals or radioactive material, it may not be disposed of via the laboratory drainage system. In this case the liquid should be disposed of as either radioactive or chemical waste as the case may be.

9.3 Solids Containing Biohazardous Agents

- Non-sharp, solid laboratory waste (empty plastic cell culture flasks and petri dishes, empty plastic tubes, gloves, wrappers, absorbent tissues, etc.) which may be, or is known to be, contaminated with viable biological agents should be collected in autoclavable bags. These plastic bags display the biohazard warning symbol and are available from Fisher Scientific.
- Autoclavable bags of solid waste should be closed but not sealed airtight to allow steam penetration before they are placed into the autoclave chamber. After autoclaving and cooling, the biohazard symbols should be removed or defaced and the autoclaved waste placed into a black plastic garbage bag.
- Agar plates may be autoclaved in double bags placed in an autoclave bin to catch any leakage. Any leaked agar should be allowed to solidify and then be disposed of as solid waste.
- After autoclaving, the sterilized waste can be placed into appropriate containers such as 20L pails or cardboard boxes for disposal.

Note: Autoclavable bags should be used for solid, non-sharp, hazardous biological waste only and disposed of appropriately. They should not be used for the collection of other solid hazardous or nonhazardous waste that may require other treatment or disposal methods.

9.4 Sharps

The term "*sharp*" is often used as a catch-all expression for any and all sharp or pointed items such as broken glassware, scalpel and razor blades, lancets, hypodermic syringes with needles, etc., which can cause cuts or puncture injuries. In this manual, sharp waste is subdivided into two categories:

- Needles and blades
- Glass and other sharp or pointed waste

Needle and blade waste is hypodermic, surgical, suture, or IV needles, syringes with needles, lancets, scalpels, blades and similar metallic sharp or pointed items for disposal that are capable of causing punctures, cuts, or tears in skin or membranes

All needles and blades used in medical care, diagnosis, and research, including the manipulation and care of laboratory animals, should be considered potentially infectious. Needles and blades pose a risk to those who use them and needle and blade waste may pose a health risk to those involved in its handling, transportation, and disposal.

- All needle and blade waste for disposal must be carefully collected in an autoclavable needle and blade waste container which meets the requirements of CSA Standard Z316.6-95.
- If the needle and blade waste is contaminated with or contains viable biological agents, it must be treated to inactivate the biological agents. The designated yellow containers for needle and blade waste are autoclavable. The filled container may be steam sterilized along with other laboratory waste.
- After autoclaving the needles and blades can be placed in a sturdy cardboard box and labeled "sharps, handle with care"

Broken glassware, intact small glass containers and tubes, and glass and plastic pipettes must be regarded as potentially sharp and pointed objects and placed into sturdy cardboard boxes or 20 litre pails which are available through Central Stores. Glassware must not protrude such that the lid cannot be closed.

- Glassware waste must not be placed into regular office garbage containers or plastic bags of solid waste.
- Do not put laboratory glassware into the general recycling bins. Its composition may differ from that of recyclable glass containers.
- The container must be closed, taped shut and labeled "**GLASS for DISPOSAL-CAUTION**".
- The sealed labeled container may be placed beside other waste awaiting removal by building service workers.
- The glassware must be free of biological, chemical or radioactive contaminants and liquids.

9.5 Labeling

- Once the waste has been decontaminated all biohazard markers must be removed or defaced. No special labeling is required for non-sharp waste.
- Containers containing sharps must be labeled "Sharps, Handle with Care".
- Containers containing glassware must be labeled "Glass for Disposal - Caution".

9.6 Special Pick up and Disposal of Untreated Biological Laboratory Waste

- Where on-site functioning autoclaves (steam sterilizers) 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.
- To provide another alternative, the University has negotiated a contract with a commercial firm which is licensed to remove and transport biologically contaminated laboratory waste to a designated disposal site. The cost of this service is passed on to the principal investigator of the laboratory generating this waste. To arrange a special pick up, contact the Biosafety Officer.

10.0 EMERGENCY RESPONSE PLAN AND SPILL CONTROL PROCEDURES

Every laboratory working with infectious biological agents must have written procedures to deal with emergencies and spills or other laboratory incidents that could be expected to result in the release of biological agents. An Emergency Response Plan (ERP) for laboratory and/or animal containment zones should be developed in collaboration with experienced facility staff, administrators and emergency responders to ensure that the plan is comprehensive and integrated with facility-wide plans emergency plans where appropriate.

The ERP should include, but is not limited to, the following:

- Emergency procedures based on a LRA outlining how to deal with the release of biological agents or biohazardous materials, and the exposure of individuals
- Protocols for the safe removal, transport and treatment of contaminated personnel and equipment
- Risk assessment tools allowing the identification of emergency scenarios and mitigation strategies
- Consultation plan for coordination with local emergency responders
- Emergency exit/evacuation routes avoiding movement through containment zones
- Consideration of emergencies that may take place within and outside of regular working hours
- Emergency access procedures overriding existing access controls when appropriate
- Record keeping of emergency response personnel who enter the containment zone
- Contingency plans to be implemented to ensure essential operations continue safely and securely
- Emergency training programs, including education on the safe and effective use of emergency equipment
- Emergency drill exercises
- Accident/injury reporting and investigation procedures
- A description of the type of emergency equipment available in the containment zone (e.g., first aid kits, spill kits, eyewash and shower stations) and directions for proper use
- Notification procedures of key personnel and the appropriate federal regulatory agencies

For common emergencies, the emergency response plan or all biosafety laboratories follow the overarching university [Emergency Preparedness Plan](#). This plan outlines the procedures for situations that constitute an emergency. Examples include fire, flooding, major health event, power failure, campus lockdown and others.

Since the capacity of most commonly used laboratory biosafety agents and protocols is small, the most common biological emergency at the university would include spills of culture preparations limited in size and therefore would be of a minor nature.

Spills may also involve other hazards such as radioisotopes and chemicals and these hazards must also be considered when developing emergency procedures. It is important to identify all risks, both potential and actual, before spill cleanup begins.

The emergency procedures for spills and exposures to biohazards is outlined on the second page of the biosafety manual as well as section 10.2 and 10.3 below.

10.1 Basic Biological Spill Cleanup Kit

Laboratories handling biohazardous agents should prepare and have available a basic biological spill cleanup kit before any work is undertaken. The contents of this kit are as follows:

Basic Biological Spill Cleanup Kit

- Written spill cleanup procedure,
- Latex gloves, protective clothing and safety goggles [Note to minimize the potential for latex allergies, non-powdered gloves are recommended],
- Chemical disinfectants appropriate to the agents being handled in the laboratory (5% Wescodyne or 5-10 % hypochlorite (bleach) are the most common),
- Tape or marking pencil to mark off the spill area,
- Biohazard spill notice sign,
- Absorbent material (absorbent paper, paper towels)
- Disposal bags – leak proof, autoclavable, labeled (biohazard),
- Sharps collector and forceps for picking up broken glass or sharps,
- Paper, incident/accident report form and pen to document the spill and any possible personnel exposure.

10.2 Spill Cleanup Protocol

The following procedure applies to small spills which can safely be handled by laboratory personnel.

- (1) Immediately notify other individuals in the area that there has been a biohazard spill.
- (2) If there is a hazard of an aerosol release, evacuate the laboratory for a time sufficient for most aerosols to settle, be dispersed or removed by the ventilation system, usually 20 to 30 minutes.
- (3) If necessary, block access to the area and mark with a biohazard spill warning sign.
- (4) Individuals involved in the spill should check for possible contamination of clothing, footwear and skin. Any potentially contaminated clothing should be left in the laboratory.
- (5) Before beginning cleanup of the spill put on appropriate protective clothing (gloves, lab coats, respiratory protection, etc.)
- (6) Identify the area requiring clean-up and decontamination.
- (7) Set up a disposal bag to allow easy discarding of contaminated cleanup materials.
- (8) Pour a strong disinfectant solution (sodium hypochlorite or Wescodyne) around, but not on the spill and mix the disinfectant with the spilled material cautiously taking care not to create an aerosol.
- (9) Use absorbent materials (paper or cloth towels) to work the decontaminant into the area of the spill.
- (10) Cover the entire area of the spill with absorbent material soaked in the decontaminant and allow it to remain in contact with the spill for 20-30 minutes.
- (11) Place the absorbent material into the disposal bag and repeat the decontamination procedure.
- (12) Remove gloves and place them in a clearly marked biohazard disposal bag with the other contaminated materials. Seal the bag and place it inside another marked biohazard bag for disposal.

- (13) Wash hands carefully with soap and water.
- (14) Complete an accident/incident report form. A copy will be automatically sent to the Health and Safety Officer and an accident investigation and review will be conducted by the Biosafety Officer.

10.3 Emergency Medical Procedures

The following procedure is to be followed for workers exposed to:

- blood or body fluids
- human pathogens or toxins
- infectious or communicable disease
- zoonotic agents

Exposure may be via a needlestick, cut or puncture wound, animal bite or scratch, mucous membrane contact or non-intact skin contact.

- (1) Wash the exposed site immediately
- (2) If a needlestick, cut, puncture wound, animal bite or scratch, wash with soap and water after allowing the wound to bleed freely;
- (3) If mucous membrane (eyes, nose, mouth) contact or non-intact skin contact (cuts, rash or dermatitis), flush with water at the nearest faucet or eye wash station for a minimum of 15 minutes.
- (4) Immediately inform the laboratory supervisor.
- (5) Seek prompt medical attention at the University/College Health and Wellness Centre.
- (6) Complete an [accident/incident report](#). A copy will be automatically sent to the Health and Safety Officer and an accident investigation and review will be conducted by the Biosafety Officer.

10.4 Incident Reporting

For purposes of incident reporting within the University a reportable incident is one which:

- results in personal injury (including injuries requiring first aid or an occupational illness), lost time from work or property damage;
- has the potential to result in personal injury or property damage even though no injury or damage actually occurred;
- involves a fire or explosion;
- occurs to any person on university premises;
- occurs to a university employee during the course of his/her work either on or off university premises;
- occurs to a student during the course of his/her classroom, laboratory or field work;
- occurs to a student during the course of a work placement (either paid or unpaid) which forms part his/her university curriculum;

Incidents involving biohazardous material must be reported as referenced in 10.3.

11.0 REFERENCES

- (1) Laboratory Biosafety Guidelines, 3rd Edition, 2004, Health Canada, Office of Laboratory Security <http://www.phac-aspc.gc.ca/publicat/lbg-ldmbl-04/index.html> .
- (2) Biosafety in Microbiological and Biomedical Laboratories, 5th Edition, US Department of Health and Human Services, Centers for Disease Control and Prevention and National Institutes of Health.
<http://www.cdc.gov/biosafety/publications/bmbl5/index.htm>
- (3) NIH Guidelines for Research Involving Recombinant DNA Molecules, April 2002, US Department of Health and Human Services, Centers for Disease Control and Prevention and National Institutes of Health.
http://oba.od.nih.gov/rdna/nih_guidelines_oba.html
- (4) World Health Organization Laboratory Biosafety Manual, Second edition (revised) http://www.who.int/csr/resources/publications/biosafety/who_cds_csr_lyo_20034/en/
- (5) Primary Containment for Biohazards: Selection, Installation and Use of Biological Safety Cabinets, 2nd edition, September 2000, US Department of Health and Human Services, Centers for Disease Control and Prevention and National Institutes of Health
http://www.cdc.gov/biosafety/publications/bmbl5/BMML5_appendixA.pdf
- (6) Guidelines for the Safe Transport of Infectious Substances and Diagnostic Specimens World Health Organization , 1997, WHO/EMC/97.3,
<http://www.absa.org/pdf/who97.pdf>
- (7) Canadian Food Inspection Agency <http://www.inspection.gc.ca/english/toce.shtml>
- (8) National Microbiological Laboratory
<http://www.nml-lnm.gc.ca/english/index.html>
- (9) Swiss Expert Committee for Biosafety, Recommendation on the Safe Handling of Human and Animal Cells and Cell Cultures.
http://www.efbs.admin.ch/fileadmin/efbs-dateien/dokumentation/empfehlungen/10_Zellkulturen_EFBS_E.pdf
- (10) Public Health Agency of Canada. Infection Control Guidelines, Routine Practices and Additional Precautions for Preventing the Transmission of Infection in Health Care, 1999.
<http://www.phac-aspc.gc.ca/nois-sinp/guide/pubs-eng.php>
- (11) Canadian Biosafety Standards and Guidelines, 1st Edition, 2013, Health Canada, Office of Laboratory Biosafety and Security,
<http://canadianbiosafetystandards.collaboration.gc.ca/cbsg-nldcb/index-eng.php?page=1>

- (12) Guideline C-4: The Management of Biomedical Waste in Ontario, 2009, Ontario Ministry of the Environment.
<https://dr6j45jk9xcmk.cloudfront.net/documents/1761/197-biomedical-waste-en.pdf>

APPENDIX 1

MEMBERSHIP OF THE UOIT BIOSAFETY COMMITTEE

Committee Chair and Co-chair are faculty members of the Faculty of Science and/or Faculty of Health Sciences. Members also comprise of laboratory managers, instructors and lab staff from the Departments of Biology, Faculty of Science and/or Department of Medical Sciences, Faculty of Health Science.

Ex Officio Members

Tanya Neretljak
Biosafety and Radiation Safety Officer
UOIT

David Roger
Health and Safety Officer
UOIT

Jennifer Freeman
Director
Office of Research Services

Sascha Tuuha
Ethics and Compliance Officer
Office of Research Services

APPENDIX 2

PROCEDURE FOR REGISTRATION AND AUTHORIZATION OF BIOHAZARDOUS PROJECTS

1. Authorization must be obtained from UOIT's Biosafety Committee for all biohazardous substances, including toxins, Risk Group 1 and Risk Group 2 agents before acquiring such agents and commencing any research or teaching activities. Please note that Risk Group 3 and 4 agents are not permitted for use at this institution.
2. An application must be submitted to the Compliance Office (compliance@uoit.ca) as far in advance of the proposed use as possible. Approval times may vary depending on the agent, the containment level and factors such as shared laboratories. Please contact the Compliance Office if you are uncertain regarding the appropriate containment level. Forms are available on the Research Services website at http://research.uoit.ca/EN/main/231307/Research_Forms.html
3. If you plan to work with any animal material or live animals, a separate animal use protocol must be submitted to the Compliance Office. Please contact the Compliance Officer in advance of these activities and review the "Requirements for Working with Animals Guide" [http://research.uoit.ca/assets/Default/documents/Animal/REQUIREMENTS FOR WORKING WITH ANIMALS R0 SEPT 14 09.doc](http://research.uoit.ca/assets/Default/documents/Animal/REQUIREMENTS_FOR_WORKING_WITH_ANIMALS_R0_SEPT_14_09.doc).
4. Researchers applying for grants which require submission of an external agency's biohazard containment certificate form shall complete both the agency's form and a UOIT Biosafety Certificate application and forward both to the Office of the Associate Provost, Research (compliance@uoit.ca).
5. When research activities involving biohazardous materials do not require advance approval by a granting agency, the researcher upon receiving notice of the grant approval shall complete a UOIT Biohazard Certificate application and forward it to the Compliance Officer in the Office of the Vice President, Research, Innovation and International (compliance@uoit.ca).
6. It is the responsibility of the researcher applying for the Biosafety Certificate to propose an appropriate level of containment and standard operating procedures for working with the particular agent. The Biosafety Committee

will review the application and determine the appropriate level of containment as per the [Human Pathogens Toxins Act \(HPTA\)](#).

Where work is to be done in Level 2 containment, proof must be submitted with the application that a biosafety cabinet is available in the lab and has been certified within the last 12 months.

Please note that a written risk assessment will be required as part of the application in the following circumstances:

- where the proposed level of containment differs from the risk group of the agent,
 - where the work involves animals,
 - where the work involves recombinant DNA or genetic manipulation,
 - where the work involves cell lines.
7. If approval is granted, a copy of the Biosafety Certificate will be forwarded to the researcher stating the conditions necessary for approval. The original is retained by the Office of the Vice President, Research. Note: Research funds will not be released until a Biosafety Certificate is issued.
 8. Each facility where biohazardous materials are to be handled, stored or transported must be certified for use by the Biosafety Committee. Prior to approval, the Biosafety Officer will inspect the proposed facilities to ensure that appropriate containment standards can be met.
 9. Biosafety Certificates are usually issued for a particular project using a particular biohazardous agent in a specified location, using specified containment and procedures with specified personnel. **Changes to any of these parameters, will require either an amendment to the certificate or the issuance of a new certificate.** A revised permit signifying approval must be obtained from the Compliance Office before any new substances or activities may be implemented. Depending on the nature of the request, the approval process may require the full review and approval of the Biosafety Committee.

The Compliance Officer (compliance@uoit.ca) must be notified immediately of any proposed changes to authorized permits, including: 1) change in location or addition of rooms; 2) addition of new agents (toxins, agents or pathogens); 3) changes in personnel, and 4) the cessation of work with these substances.

10. The researcher must also contact the Compliance Officer upon termination of a permit and will be required to demonstrate that any remaining agents will be properly disposed of. Termination of a permit will normally require that any remaining agents be either destroyed or transferred to another Biosafety Permit holder.

11. Please note that each Biosafety Certificate must be renewed annually prior to the anniversary date of the original approval. Renewals will be issued by the Biosafety Officer pending satisfactory inspection of the lab(s). Please note that the Biosafety Officer will be conducting routine spot inspections throughout the year to ensure that approved containment practices are being upheld. Issues of non-compliance will be documented within the Biosafety file and the researcher will be notified accordingly.



UNIVERSITY OF ONTARIO INSTITUTE OF
TECHNOLOGY
APPLICATION FOR BIOSAFETY CERTIFICATE

1. CERTIFICATE HOLDER INFORMATION

Principal Investigator: _____ Title/Position: _____
Faculty or School: _____
Office Building/Room Number: _____ Telephone Extension: _____
Email: _____
Laboratory Building/Room Number: _____

2. PROJECT INFORMATION

Project Title/Description: _____

Project Sponsor/Agency: _____ Fund/Grant Number: _____
Proposed Level of Containment: Level 1 Level 2
(circle the appropriate level)
Attach a copy of the procedures/protocols for the work
If Level 2 containment required, attach a copy of reports on testing and certification of
biological safety cabinets performed during the previous 12 months.

3. BIOLOGICAL AGENT USAGE

Indicate usage by checking the relevant boxes.

- human tissues and cells human blood and blood fractions human body fluids
 primary human cell cultures established human cell lines

| | | |
|---|---|---|
| <input type="checkbox"/> animal tissues and cells | <input type="checkbox"/> animal blood and blood fractions | <input type="checkbox"/> animal body fluids |
| <input type="checkbox"/> primary animal cell cultures | <input type="checkbox"/> established animal cell lines | |
| <input type="checkbox"/> bacteria | <input type="checkbox"/> viruses | <input type="checkbox"/> fungi <input type="checkbox"/> parasites <input type="checkbox"/> microbial toxins |
| <input type="checkbox"/> recombinant DNA/RNA | <input type="checkbox"/> other (specify) _____ | |

4. BIOLOGICAL AGENTS USED

Identify the biological agents used in the project:

| <u>Common Name(s)</u> | <u>Scientific Name/Species</u> | <u>Risk Group</u> |
|-----------------------|--------------------------------|-------------------|
| _____ | _____ | _____ |
| _____ | _____ | _____ |
| _____ | _____ | _____ |
| _____ | _____ | _____ |
| _____ | _____ | _____ |

5. HUMAN AND/OR ANIMAL USAGE

If human subjects or animals are being used, indicate by marking the appropriate boxes. Provide an attachment briefly outlining the procedures to be used.

- no animals will be used non-primate mammals non-human primates
- other animals (specify) _____
- Animal use protocol approved (attach a copy)
- Animal use protocol pending
- No human subjects will be used Human Subjects to be used
- Human use protocol approved (attach a copy)
- Research Ethics Board approval pending

6. OTHER AGENTS

Is ionizing radiation being used in conjunction with biological agents? Yes No

If yes, provide Radioisotope Permit Number: _____

If yes, list the isotopes _____

Is any other type of radiation used? No Yes (specify) _____

Are any designated substances used: No Yes (specify) _____

NOTES:

1. The University's radioisotope license prohibits the use of radioactive materials in or on humans. Special approval from the Canadian Nuclear Safety Commission will be required. Contact the Chair of the Radiation Safety Committee.

2 Designated Substances are: *Acrylonitrile, Arsenic, Asbestos, Benzene, Ethylene Oxide, Isocyanates, Lead, Mercury, Silica, Vinyl Chloride*

7. PERSONNEL WORKING ON PROJECT

List the names of all individuals who will be working with the agents identified on this application.

8. SIGNATURES

Principal Investigator (please print clearly)

Signature

Date

9. THIS SECTION FOR COMMITTEE USE

Conditions

Approvals

Biosafety Officer

Date

Chair, Biosafety Committee

Date

APPENDIX 3

SCHEDULES OF HUMAN PATHOGENS AND TOXINS

The purpose of the [Human Pathogens and Toxins Act](#) is to establish a safety and security regime to protect the health and safety of the public against the risks posed by human pathogens and toxins. Under this Act every person who knowingly conducts an activity involving a human pathogen or toxin shall take all reasonable precautions to protect the health and safety of the public against the risks posed by that activity.

There are 5 Schedules to the Act which list the agents regulated under the Act. These schedules are reproduced here as a quick reference to researchers in identifying those agents regulated under the Act. The Schedules are constantly updated and reference should be made to the current listings on the [Department of Justice website](#).

The use of any agent listed in the schedules requires registration with the Public Health Agency of Canada. The University has registered specific laboratory rooms and further information on this registration can be obtained from the Office of Research Services, [Ethics and Compliance Officer](#).

The HPTA is not an exhaustive list of risk group 1-4 pathogens. Some non-pathogenic strains of agents (for example, *E. Coli*) require a pathogen risk assessment by the principal investigator and the Biosafety Committee. The risk assessment is to satisfy the containment level requirements for these non-pathogenic strains as well as any biohazardous materials (blood products, cell lines, recombinant DNA, etc.) which may be currently unclassified under the HPTA or through The Public Health Agency of Canada. Non-pathogenic strains of agents should be listed under individual principal investigators' pathogen inventory records.

The Schedules to the Act are:

- Schedule 1: Toxins
- Schedule 2: Risk Group 1 Human Pathogens
- Schedule 3: Risk Group 3 Human Pathogens
- Schedule 4: Risk Group 4 Human Pathogens
- Schedule 5: Prohibited human Pathogens and Toxins

Note that Risk Group 3, 4, and 5 pathogens are prohibited for use at UOIT.

SCHEDULE 1 - TOXINS

Aerolysin
Alpha toxin
Anthrax toxins: Lethal Toxin and Oedema Toxin
Bordetella pertussis Adenylate cyclase toxin
Botulinum neurotoxin
Cholera toxin
Clostridium botulinum C2 and C3 toxins
Clostridium difficile toxins A and B
Clostridium perfringens Epsilon toxin
Dermonecrotic toxin
Diphtheria toxin
Escherichia coli toxins: *E. coli* Cytotoxic Necrotizing Factor (CNF), Heat-labile *E. coli* enterotoxin (LT), Heat-stable *E. coli* enterotoxin (ST), Cytolethal distending toxin (CLDT) and Enteraggregative Shiga-like toxin 1 (EAST)
Exfoliative toxin (also called Exfoliatin)
Exotoxin A
Hemolysin
Listeriolysin O
Pasteurella multocida toxin
Perfringolysin O
Pertussis toxin
Pneumolysin
Pyrogenic exotoxin
Shiga-like toxin (verotoxin)
Shigatoxin
Staphylococcal enterotoxins
Staphylococcus aureus Toxic shock syndrome toxin
Streptolysin O
Tetanolysin
Tetanospasmin (Tetanus toxin)

SCHEDULE 2 - RISK GROUP 2 HUMAN PATHOGENS

| Bacteria | |
|--|-------------------------------------|
| <i>Actinobacillus pleuropneumoniae</i> | <i>Moraxella catarrhalis</i> |
| <i>Actinobacillus ureae</i> | <i>Mycobacterium avium</i> |
| <i>Actinomyces israelii</i> | <i>Mycobacterium leprae</i> |
| <i>Aerococcus ureinae</i> | <i>Mycobacterium smegmatis</i> |
| <i>Aeromonas hydrophila</i> | <i>Mycoplasma genitalium</i> |
| <i>Aggregatibacter actinomycetemcomitans</i> | <i>Mycoplasma pneumoniae</i> |
| <i>Arcanobacterium bernardiae</i> | <i>Neisseria gonorrhoeae</i> |
| <i>Bordetella bronchiseptica</i> | <i>Neisseria meningitidis</i> |
| <i>Bordetella parapertussis</i> | <i>Pasteurella multocida</i> |
| <i>Bordetella pertussis</i> | <i>Porphyromonas gingivalis</i> |
| <i>Borrelia burgdorferi</i> | <i>Proteus mirabilis</i> |
| <i>Campylobacter jejuni</i> | <i>Proteus vulgaris</i> |
| <i>Chlamydia trachomatis</i> | <i>Pseudomonas aeruginosa</i> |
| <i>Chlamydophila pneumoniae</i> | <i>Salmonella</i> |
| <i>Citrobacter freundii</i> | <i>Serratia marcescens</i> |
| <i>Clostridium botulinum</i> | <i>Shigella dysenteriae</i> |
| <i>Clostridium difficile</i> | <i>Shigella flexneri</i> |
| <i>Clostridium perfringens</i> | <i>Shigella sonnei</i> |
| <i>Clostridium tetani</i> | <i>Sphingobacterium faecium</i> |
| <i>Corynebacterium diphtheriae</i> | <i>Staphylococcus aureus</i> |
| <i>Enterococcus faecium</i> | <i>Staphylococcus saprophyticus</i> |
| <i>Escherichia coli</i> | <i>Streptococcus agalactiae</i> |
| <i>Francisella novicida</i> | <i>Streptococcus pyogenes</i> |
| <i>Haemophilus influenzae</i> | <i>Streptococcus salivarius</i> |
| <i>Haemophilus parainfluenzae</i> | <i>Treponema pallidum</i> |
| <i>Helicobacter pylori</i> | <i>Ureaplasma urealyticum</i> |
| <i>Klebsiella pneumoniae</i> | <i>Vibrio cholerae</i> |
| <i>Legionella pneumophila</i> | <i>Yersinia pseudotuberculosis</i> |
| <i>Leptospira interrogans</i> | |
| <i>Listeria monocytogenes</i> | |

| Viruses | |
|---|--|
| <p><i>Adenovirus</i> <i>Avian influenza virus (excluding highly pathogenic strains)</i> <i>Colorado tick fever viruses</i> <i>Cowpox virus</i> <i>Coxsackievirus</i> <i>Epstein Barr virus</i> <i>Hepatitis A virus</i> <i>Hepatitis B virus</i> <i>Hepatitis C virus</i> <i>Hepatitis D virus</i> <i>Hepatitis E virus</i> <i>Herpes simplex viruses</i> <i>Human coronavirus (excluding SARS-CoV)</i> <i>Human herpesvirus 5 (cytomegalovirus)</i> <i>Human herpesvirus 6 (roseolovirus)</i> <i>Human herpesvirus 8 (Kaposi's sarcoma-associated herpesvirus)</i> <i>Human parvovirus</i> <i>Human rotavirus</i> <i>Influenza virus, types A-C (excluding Type A 1918 Spanish Flu and H2N2 strains)</i></p> | <p><i>Measles virus</i> <i>Molluscum contagiosum virus</i> <i>Mumps virus</i> <i>Newcastle disease virus</i> <i>Norwalk virus</i> <i>Papillomaviruses</i> <i>Parainfluenza virus (types 1-4)</i> <i>Reoviruses</i> <i>Respiratory syncytial virus</i> <i>Rhinovirus</i> <i>Semliki Forest virus</i> <i>Sendai virus</i> <i>Simian virus 40</i> <i>Vaccinia virus</i></p> |

| Fungi | |
|--|--|
| <p><i>Aspergillus fumigatus</i> <i>Aspergillus niger</i> <i>Aspergillus oryzae</i> <i>Candidia albicans</i> <i>Cryptococcus neoformans</i> <i>Microsporium audouinii</i> <i>Microsporium ferrugineum</i></p> | <p><i>Sporothrix schenkii</i> <i>Trichophyton concentricum</i> <i>Trichophyton rubrum</i> <i>Trichophyton schoenleinii</i> <i>Trichophyton tonsurans</i></p> |

| Protozoa | |
|--|--|
| <p><i>Acanthamoeba castellanii</i> <i>Leishmania aethiopica</i> <i>Leishmania braziliensis</i> <i>Leishmania chagasi</i> <i>Leishmania donovani</i> <i>Leishmania guyanensis</i></p> | <p><i>Leishmania infantum</i> <i>Leishmania panamensis</i> <i>Plasmodium falciparum</i> <i>Trypanosoma brucei gambiense</i> <i>Trypanosoma brucei rhodiense</i> <i>Trypanosoma cruzi</i></p> |

| Prions | |
|--------------------------------------|--|
| <i>Chronic wasting disease agent</i> | |

SCHEDULE 3 - RISK GROUP 3 HUMAN PATHOGENS

| Bacteria | |
|----------------------------------|-----------------------------------|
| <i>Bacillus anthracis</i> | <i>Mycobacterium microti</i> |
| <i>Brucella abortus</i> | <i>Mycobacterium tuberculosis</i> |
| <i>Brucella canis</i> | <i>Neorickettsia sennetsu</i> |
| <i>Brucella melitensis</i> | <i>Rickettsia akari</i> |
| <i>Brucella ovis</i> | <i>Rickettsia australis</i> |
| <i>Brucella suis</i> | <i>Rickettsia conorii</i> |
| <i>Burkholderia mallei</i> | <i>Rickettsia japonicum</i> |
| <i>Burkholderia pseudomallei</i> | <i>Rickettsia prowazekii</i> |
| <i>Chlamydia psittaci</i> | <i>Rickettsia rickettsii</i> |
| <i>Coxiella burnetii</i> | <i>Rickettsia siberica</i> |
| <i>Francisella tularensis</i> | <i>Rickettsia typhi</i> |
| <i>Mycobacterium africanum</i> | <i>Yersinia pestis</i> |
| <i>Mycobacterium bovis</i> | |
| <i>Mycobacterium canettii</i> | |

| Viruses | |
|--|---|
| <i>African Horse Sickness virus</i> | <i>Mapuera virus</i> |
| <i>Água Preta virus</i> | <i>Mayaro virus</i> |
| <i>Akabane virus</i> | <i>Mobala virus</i> |
| <i>Allpahuayo virus</i> | <i>Monkeypox virus</i> |
| <i>Andes virus</i> | <i>Monongahela virus</i> |
| <i>Araguari virus</i> | <i>Mopeia virus</i> |
| <i>Batken virus</i> | <i>Mucambo virus</i> |
| <i>Bayou virus</i> | <i>Murray Valley encephalitis virus</i> |
| <i>Bear Canyon virus</i> | <i>Negishi virus</i> |
| <i>Bermejo virus</i> | <i>New York virus</i> |
| <i>Bhanja virus</i> | <i>Ngari virus</i> |
| <i>Bijou Bridge virus</i> | <i>Oliveros virus</i> |
| <i>Black Creek Canal virus</i> | <i>O'Nyong-nyong virus</i> |
| <i>Cabassou virus</i> | <i>Oran virus</i> |
| <i>Cano Delgadito virus</i> | <i>Oropouche virus</i> |
| <i>Chikungunya virus</i> | <i>Pergamino virus</i> |
| <i>Dhori virus</i> | <i>Pirital virus</i> |
| <i>Dobrava-Belgrade virus</i> | <i>Piry virus</i> |
| <i>Douglas virus</i> | <i>Powassan virus</i> |
| <i>Dugbe virus</i> | <i>Puumala virus</i> |
| <i>Duvenhage virus</i> | <i>Rabies virus</i> |
| <i>Eastern equine encephalitis virus</i> | <i>Rift Valley fever virus</i> |
| <i>Enseada virus</i> | <i>Rocio virus</i> |
| <i>Everglades virus</i> | <i>Saaremaa virus</i> |
| <i>Flexal virus</i> | <i>Sakpa virus</i> |
| <i>Garissa virus</i> | <i>SARS coronavirus (SARS-CoV)</i> |
| <i>Germiston virus</i> | <i>Seoul virus</i> |
| <i>Hantaan virus</i> | <i>Sin nombre virus</i> |
| <i>Herpesvirus ateles</i> | <i>Slovakia virus</i> |
| <i>Herpesvirus saimiri</i> | <i>Somone virus</i> |
| <i>Highly pathogenic avian influenza virus</i> | <i>Sripur virus</i> |
| <i>Human immunodeficiency virus</i> | <i>St. Louis encephalitis virus</i> |
| <i>Human T-cell lymphotropic virus</i> | <i>Thogoto virus</i> |
| <i>Influenza A H2N2</i> | <i>Tonate virus</i> |
| <i>Israel Turkey meningoencephalitis virus</i> | <i>Topografov virus</i> |
| <i>Issyk-Kul virus</i> | <i>Venezuelan equine encephalitis virus</i> |
| <i>Japanese encephalitis virus</i> | <i>Vesicular stomatitis virus</i> |
| <i>Juquitiba virus</i> | <i>Wesselsbron virus</i> |
| <i>Khabarovsk virus</i> | <i>Western equine encephalitis virus</i> |
| <i>Koutango virus</i> | <i>West Nile fever virus</i> |
| <i>Kunjin virus</i> | <i>Whitewater Arroyo virus</i> |
| <i>Laguna Negra virus</i> | <i>Xingu virus</i> |
| <i>Lechiguanas virus</i> | <i>Yellow fever virus</i> |
| <i>Louping ill virus</i> | |
| <i>Lymphocytic choriomeningitis virus</i> | |
| <i>Maporal virus</i> | |

| Fungi | |
|--|--|
| <i>Blasomyces dermatitidis</i> <i>Cladophialophora bantiana</i> <i>Coccidioides immitis</i> <i>Coccidioides posadasii</i> | <i>Histoplasma capsulatum</i> <i>Paracoccidioides brasiliensis</i> <i>Penicillium marneffe</i> |

| Protozoa - Prions | |
|---|---|
| <i>Bovine spongiform encephalopathy agent and other related animal transmissible spongiform encephalopathies agents</i> <i>Creutzfeldt-Jakob disease agent</i> <i>Fatal Familial Insomnia agent</i> | <i>Gerstmann-Sträussler-Scheinker syndrome agent</i> <i>Kuru agent</i> <i>Variant Creutzfeldt-Jakob disease agent</i> |

SCHEDULE 4 - RISK GROUP 4 HUMAN PATHOGENS

| Viruses | |
|---|---|
| <i>Absettarov virus</i> | <i>Kyasanur Forest virus</i> |
| <i>Alkhumra virus</i> | <i>Lassa fever virus</i> |
| <i>Crimean Congo haemorrhagic fever virus</i> | <i>Machupo virus</i> |
| <i>Ebola virus</i> | <i>Marburg virus</i> |
| <i>Guanarito virus</i> | <i>Nipah virus</i> |
| <i>Hanzalova virus</i> | <i>Omsk haemorrhagic fever virus</i> |
| <i>Hendra virus</i> | <i>Russian spring-summer encephalitis virus</i> |
| <i>Herpes B virus</i> | <i>Sabia virus</i> |
| <i>Hypr virus</i> | |
| <i>Junin virus</i> | |
| <i>Kumlinge virus</i> | |

SCHEDULE 5 - PROHIBITED HUMAN PATHOGENS AND TOXINS

Human Pathogens

Variola virus

APPENDIX 4

BIOLOGICAL SAFETY CABINETS

A properly maintained biological safety cabinet, when used in conjunction with good laboratory techniques, can provide effective primary containment for work with human pathogens. In containment level 2 facilities biological safety cabinets are used for procedures with the potential to produce infectious aerosols and for high concentrations or large volumes of infectious material.

There are three classes of biological safety cabinets, Class I, Class II and Class III. Only cabinets which meet the requirements of the National Sanitation Foundation (NSF) Standard No. 49-2002 and bear an NSF 49 seal should be purchases.

A4.1 Types of Biological Safety Cabinet

Class I Cabinets

Class I cabinets are negative pressure ventilated cabinets with an unrecirculated airflow away from the operator that is discharged through a HEPA (High Efficiency Particle Air) filter either to the laboratory or to the outside. They provide good operator protection, but do not protect the material within the cabinet from contamination since the inward flow of air from the laboratory is unfiltered.

The Class I cabinet is designed for general microbiological research with low and moderate risk agents.

Class II Cabinets

Class II cabinets are designed for personnel, product and environmental protection in that both the incoming and outgoing air are HEPA filtered. Class II cabinets are subdivided into types A1, A2 and B1 and B2 on the basis of construction type, airflow velocities and patterns and exhaust systems. The basic characteristics of these four types are:

Class II, Type A1

- Air is recirculated within the cabinet
- Cabinet air may be exhausted back into the laboratory or to the outdoors via the building exhaust system
- Maintains a minimum average face velocity of 0.38 m/s
- May have positive pressure contaminated ducts and plenums
- Are not suitable for work with low levels of volatile toxic chemicals and volatile radionuclides

Class II, Type A2

- Air is recirculated within the cabinet
- Cabinet air may be exhausted back into the laboratory or to the outdoors via the building exhaust system
- Maintains a minimum average face velocity of 0.5 m/s
- Has ducts and plenums under negative pressure
- Is suitable for work with minute quantities of volatile toxic chemicals and trace amounts of radionuclides.

Class II, Type B1

- Hard-ducted through a dedicated duct exhausted to the atmosphere after passage through a HEPA filter
- Contains negative pressure plenum
- Maintains a minimum average face velocity of 0.5 m/s
- Recirculates 30% of the air within the cabinet
- Suitable for work with low levels of volatile toxic chemicals and trace amounts of radionuclides

Class II, Type B2

- Hard-ducted through a dedicated duct exhausted to the atmosphere after passage through a HEPA filter
- Contains negative pressure plenum
- Does not recirculate air within the cabinet – 100% of cabinet air is exhausted
- Maintains a minimum average face velocity of 0.5 m/s
- Suitable for work with volatile toxic chemicals and radionuclides.

Class III Cabinets

Class III cabinets are totally enclosed and gas-tight with HEPA filtered supply and exhaust air. The cabinet is kept under negative pressure of at least 120 Pa and airflow is maintained by a dedicated exterior exhaust system. The exhaust air is passed through two HEPA filters in series or through a single HEPA filter followed by incineration before discharge from the facility. Removal of materials from the cabinet is through a double door autoclave or air-lock pass-through for decontamination.

Class III cabinets are designed for work with level 4 pathogens and provide an alternative to the positive-pressure suit made for maximum containment laboratories.

A4.2 Installation and Certification

Biological Safety Cabinets should be installed in accordance with the requirements outlined in the Canadian Standards Association Standard Z316.3-95 – *Biological containment cabinets (class I and II): installation and field testing*. They should be located away from high traffic areas, doors and air supply/exhaust grilles that may interrupt airflow patterns. Wherever possible, a 30 cm clearance should be provided on each side of the cabinet to allow for maintenance access.

The correct operation of Biological Safety Cabinets must be verified before they are used, annually thereafter and after any repairs or relocation. Note that they must be decontaminated before any servicing or relocation.

A copy of the certification report must be provided to the user, and kept on file. A copy must be sent to the Office of Research Services for inclusion in the Biosafety Certificate file. A label indicating the date of certification, to what standard the tests were performed, the date of the next certification and the name of the certifier must be affixed to the exterior of the cabinet.

A4.3 Certification Testing Criteria

The routine decontamination and testing of used Class II biological safety cabinets shall include the following required procedures and tests which shall be conducted in accordance with, and in the manner described below.

Decontamination

Cabinet decontamination with paraformaldehyde vapour shall be conducted prior to the testing of biological safety cabinets which have been used for activities involving biological agents assigned to the Risk Groups identified by Health Canada. The biological safety cabinet shall be sealed and decontaminated using the paraformaldehyde vapour technique which is described in NSF Standard 49, and cited in CSA Z316.3-95, or an equivalent procedure acceptable to the Biosafety Committee of the University. The paraformaldehyde holding / contact time shall be a minimum of 2 hours, after which the paraformaldehyde vapour shall be neutralized or vented to the exterior of the building.

Containment System Integrity

Containment system integrity (pressure) testing shall be performed on all biological safety cabinets having air plenums which convey potentially contaminated air at positive pressure and where any portion of these plenums also forms part of the containment shell of the cabinet. The cabinet interior shall be pressurized with air to a differential pressure of 2" w.g. A liquid leak detector shall be applied along all welds, gaskets, penetrations, and seals on the exterior surfaces of the cabinet air plenums. Leakage will be indicated by the presence of bubbles or by the feel or sound of escaping air. Detected leakage shall be corrected using acceptable methods and materials and the repaired area shall be retested to confirm the success of the corrective action.

Note: The performance of this test is required at the time of initial cabinet installation, following cabinet relocation, and at least once in every three year period.

Air Velocities and Volumes

Air velocities shall be measured at multiple points on a grid, across the face of the HEPA filters. The location and spacing of the co-ordinates shall be according to the manufacturer's recommendations and / or applicable standards. Additional air velocity measurements may be required by the manufacturer of the cabinet. The blower speed and air dampers shall be adjusted as required so that the final measured and calculated values are within the acceptable ranges indicated by the manufacturer of the biological safety cabinet.

HEPA Filter Integrity

HEPA filter leak testing shall be performed using sufficient dioctylphthalate (DOP) aerosol (or equivalent) to challenge the air filtration system. The aerosol concentration upstream of the HEPA filters shall be sampled and used as the 100% reference for photometer adjustment prior to testing. All air diffusers and protective grilles downstream of HEPA filters shall be removed to allow direct access to the entire filter surface and perimeter (bond area, gasket, filter frame, and mounting frame) which shall be scanned in overlapping strokes at a traverse rate of not more than 2" per second. Aerosol penetration exceeding 0.01% of the upstream concentration shall be

sealed or corrected using generally accepted methods and the repaired area shall be retested to confirm the success of the corrective action.

Airflow Smoke Patterns

These tests shall be performed using a source of visible smoke to demonstrate the acceptability of airflows associated with the biological safety cabinet:

(a) Downflow Supply Air Distribution

The source of visible smoke shall be passed along the ('smoke split') centreline of the work surface, from one side of the workspace to the other. The smoke shall show smooth flow with no dead spots or upward flow. No smoke shall escape from the cabinet.

b) Supply Air Entrainment / View Screen Retention

The source of visible smoke shall be passed from one side of the cabinet to the other, behind the view screen, and 6" above the top of the front access opening. The smoke shall show smooth downward flow with no dead spots or upward flow. No smoke shall escape from the cabinet.

c) Intake Air Entrainment / Work Access Opening Retention

The source of visible smoke shall be passed along the entire work access perimeter, about 1.5" outside of the workspace of the cabinet. No smoke shall escape from the cabinet once it is drawn in. For Class II cabinets, no smoke shall pass over the work surface or penetrate the work zone.

d) Window Seal

The source of visible smoke shall be passed along the perimeter of the view screen, inside the workspace of the cabinet. No smoke shall escape from the cabinet.

Other Procedures and Tests

Other procedures and tests (electrical safety, fluorescent and UV lighting intensity, vibration, noise level, etc.) may be recommended or performed, depending on the cabinet design and the circumstances of its installation and usage, but their performance is not required on a routine basis.

A4.4 Testing Services

Facilities Management arranges for annual testing of the Biosafety Cabinets (usually in August) and all information regarding the testing is available in the Facilities Office (UA B441). The cabinets should have a sticker indicating the date of the last testing and users should ensure that the testing has been completed within the last 12 months.

If laboratory personnel require testing over and above the annual test, they can contact the helpdesk@dc-uoit.ca or call extension 2326. Extra testing will be charged back.

APPENDIX 5

Disinfectants for Common Agents

| | |
|---|---|
| <p>Vegetative bacteria</p> <ul style="list-style-type: none"> - E. Coli - Staphylococci - Streptococci <p>Enveloped viruses</p> <ul style="list-style-type: none"> - HIV - Herpes | <p>1% domestic bleach</p> <p>75% Ethanol</p> <p>6% formulated Hydrogen peroxide</p> <p>Quaternary ammonia</p> |
| | |
| <p>Mycobacteria and fungi</p> | <p>1% domestic bleach</p> <p>75% Ethanol</p> <p>6% formulated Hydrogen peroxide</p> <p>Phenolic compounds</p> |
| | |
| <p>Spore forming bacteria</p> <ul style="list-style-type: none"> - Bacillus <p>Non-Enveloped Viruses</p> <ul style="list-style-type: none"> - Adenovirus - Hepatitis | <p>10% domestic bleach</p> <p>6% formulated Hydrogen peroxide</p> <p>Formaldehyde</p> <p>Gluteraldehyde</p> |