Thompson Rivers University

BioSafety Manual

REV.	REASON FOR REVISION	ISSUE DATE	ISSUED BY
A	Original Issue	April 2010	TRU Biosafety Officer
B	Revised format	April, 2012	TRU Biosafety Officer

DISCLAIMER

The materials contained in this document have been compiled from sources believed to be reliable and to represent the best opinions on the subject. This document is intended to serve only as a starting point for good practices and does not purport to specify minimal legal standards. No warranty, guarantee, or representation is made by TRU as to the accuracy or sufficiency of information contained herein, and TRU assumes no responsibility in connection therewith. This document is intended to provide basic guidelines for safe practices. Therefore, it cannot be assumed that all necessary warning and precautionary measures are contained in this document and that other or additional information or measures may not be required. Users of this document should consult alternate sources of safety information prior to undertaking specific tasks.

ACKNOWLEDGEMENTS

Thanks to the Occupational Health and Safety Department of Wilfred Laurier University for giving permission to use their Biosafety Manual as a template for this manual.

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1. INTRODUCTION

1.1 Scope

This manual describes requirements and procedures established for work with potentially hazardous biological agents. These are applicable to all laboratory research activities and teaching labs that may involve exposure to these agents. The manual is based upon the Health Canada Laboratory Guidelines (2004 edition) and reflects best practices.

1.2 Regulatory Forces and Guidelines

Guidelines developed by Public Health Agency of Canada (PHAC) form the basis for the biosafety practices included in this manual. These guidelines must be followed to ensure the continuation of granted funds from federal agencies.

The PHAC Laboratory guidelines:

- Mandate the establishment of an Institutional Biosafety Committee (IBC) for the review and oversight of biological research.
- Outline roles and responsibilities for biosafety.
- Establish the practices, procedures, and conditions under which work with biological agents must be conducted.

Obtainment, possession, use, or transfer of any biological agent or toxin is strictly controlled by federal regulations. Importation of human pathogens is regulated by the Importation of Human Pathogens Regulations. The Health of Animals Act and its regulations give the Canadian Food Inspection Agency (CFIA) the authority to control the use of significant animal pathogens associated with reportable animal diseases.

The requirements for packaging and shipment of biomedical materials are governed by the Transportation of Dangerous Goods Regulations (TDGR), which are administered by Transport Canada.

1.3 Definitions

- **Infectious agent (Risk Group 2):** An organism that is capable of producing an infection or infectious disease.
- **Infectiousness:** The relative ease with which a disease is transmitted to other hosts.
- **Human pathogen (Risk Group 2):** An organism infectious to and capable of causing disease in humans.
- **Opportunistic microorganism (Risk Group 1)**: Organisms that take advantage of temporary decreases in a host's immune defences to cause infection. Under normal circumstances do not cause disease.

1.4 Compliance Enforcement Policy

TRU assumes the responsibility of ensuring to PHAC and the Canadian Food Inspection Agency (CFIA) that the use of biohazards will be undertaken in a safe manner and in compliance with Health Canada and CFIA guidelines. The Biosafety Officer (BSO) is responsible for ensuring compliance enforcement. All supervisors are responsible ensuring their students comply with TRU safe operating procedures (SOPs)

All compliance violations are categorized as major or minor offences. The categories aid in determining the

level of risk or immediate danger to safety and health, and suggest the response that may be required when issues of non-compliance are identified by Occupational Health and Safety (OHS).

All deficiencies must be in writing to the AVP Research and Graduate Studies, including the corrective action taken. Any offence occurring twice in any one-year period will be considered a second offence.

A major offence is a violation that causes immediate risk or danger to safety and health, or could cause a release of biohazards into the environment or the community. Examples of major offences include:

1. Use or storage of food/drink or smoking in the laboratory.
2. Inadequate training of new staff.
3. Refusal to participate in the Level 2 Inspection Program.
4. Unauthorized possession/use of biohazards.
5. Inadequate or unsafe storage areas for biohazards.

A minor offence is an infraction that poses no immediate risk or threat to safety, health,or the environment. Examples include:

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

Actions Following a Major Offence:

1. First Offense: A written notification will be sent to the Permit Holder or Supervisor by the Biosafety Officer () with a copy to the Department Chair, the Manager of OH&S and the Biosafety Committee Chair. Immediate correction of the violation is required, and a written reply to the BSO in 7 days. If the written reply i snot received within 7 days, a second notice will be sent, with a copy to the Dean of the Faculty. If there is no response within 7 days to the second notice, a meeting will be arranged with the Permit Holder, Department Chair, and the Manager of OH&S.
2. Second Offense: The Permit Holder will be notified in writing by the Biosafety Officer that the permit will be suspended until a meeting with the Biosafety Committee can be held to discuss the offence(s).
3. Third Offence: The Biosafety Officer will recommend to the Biosafety Committee that the Permit Holder's permit be cancelled. This recommendation will be copied to the Dean of the Faculty of Science, the Department Chair, and the Manager of OH&S.

Actions Following a Minor Offence:

1. First Offense: A written notification will be sent by the BSO to the Permit Holder or Supervisor with a copy to the Department Chair, the Manager of OH&S and the Biosafety Committee Chair. Corrective action of the violation is required, with a written reply in 21 days. If the written reply is not received within 21 days, a second notice will be copied to the Dean of the Faculty. If there is no response within 14 days of the second notice, a meeting will be arranged the BSO with the Permit Holder, the Department Chair, and the Manager of EOHS.
2. Second Offence: A meeting will be arranged by the BSO with the Permit Holder or Supervisor, the Department Chair, and the Manager of OH&S, to review the issues.

3. Third Offence: The Permit Holder will be notified in writing by the Biosafety Officer that the permit will be suspended until a meeting with the Biosafety Committee can be held.
4. Fourth Offence: The Biosafety Officer will recommend permit cancellation to the Biosafety Committee.

Note: For the second, third and fourth occurrences, notification of the actions outlined above will be copied to the Dean of the Faculty, the Department Chair, the Manager of OH&S and the Biosafety Committee Chair.

1.5 Material Safety Data Sheets

Material Safety Data Sheets for infectious microorganisms (biological agents) have been prepared by the Office of Biosafety, Laboratory Centre for Disease Control, Health Canada. These MSDS are available on the Internet via a hyperlink at http://www.phac-aspc.gc.ca/msds-ftss/index.html .

These MSDS contain health hazard information, recommended precautions, spill clean-up procedures, and other information that is relevant specifically to the laboratory setting. They serve as an additional safety resource for laboratory personnel working with biological agents.

MSDSs for chemicals used in TRU laboratories can be found clicking on the MSDS icon on any TRU computer desktop.

2. CLASSIFICATION OF BIOLOGICAL AGENTS

2.1 Risk Groups

Biological agents are categorized into risk groups based on the relative hazards they pose. The factors used to determine an agent's risk group includes its pathogenicity, infective dose, mode of transmission, host range, availability of effective preventive measures and availability of effective treatment.

These classifications presume ordinary circumstances in the research laboratory or growth in small volumes for diagnostic and experimental purposes.

Risk Group 1 (low individual and community risk)

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

Risk Group 2 (moderate individual risk, limited community risk)

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

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

A biological agent that usually causes serious human or animal disease, or can result in serious economic consequences, but does not ordinarily spread by casual contact from one individual to another, or can be treated by antimicrobial or antiparasitic agents.

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

A biological agent that usually produces very serious human or animal disease, often untreatable, and which may be readily transmitted from one individual to another, or from animal to human or vice- versa, directly or indirectly, or by casual contact.

An abbreviated list of organisms by Risk Group can be found in Appendix A. Since these classifications are updated regularly, a complete and current list should be consulted by contacting the Office of Laboratory Security at (613) 957-1779 or accessing the web site at http://www.phac-aspc.gc.ca/ols-bsl/index.html

2.2 Categories of Pathogens

As a general precaution, the risk group for agents should be raised when manipulation may result in the production of infectious droplets or aerosols. Agents with similar pathogenic characteristics that are not included in these lists should be considered in the same risk category. Since many agents may be referred to in the literature by a variety of names, the Supervisor in consultation with the Biosafety Officer must fully verify an unlisted organism's characteristics before determining a classification.

2.3 Inventory

Inventory records are maintained by the Biosafety Officer. The Biosafety Committee will review the list of registered materials annually. Inventory shall include material from commercial, university or private institutions regardless of whether it is a gift or purchased. The Principal Investigator (PI) or Supervisor will be responsible for properly providing the BSO with the biological materials acquired from outside sources.

All new organisms must be registered with the Biosafety Officer before they are introduced to TRU

Registration assists the Biosafety Officer in developing a catalogue/inventory of biohazardous materials on campus, and registration is used by the Biosafety Committee to assist in assigning biological safety levels (Risk Group category) to each agent. It also helps to ensure that research teams are working with these materials in a manner that is safe for everyone's protection and that appropriate permits are obtained.

Importation requirements and forms may be obtained from:

Public Health Agency of Canada at http://www.phac-aspc.gc.ca/ols-bsl/index.html

Canadian Food Inspection Agency at http://www.inspection.gc.ca/english/sci/bio/bioe.shtm

3. BIOSAFETY CONTAINMENT LEVELS

Classification of biological agents into Risk Groups does NOT establish guidelines for handling agents safely in the laboratory setting. Therefore, Containment Levels have been established to provide end users with a description of the minimum containment required. Containment Levels describe the characteristics of the agent and the engineering, operational, technical and physical requirements for manipulating it safely.

Risk assessment is a critical step in the selection of an appropriate Containment Level for any biohazardous work. A detailed assessment should be conducted to determine both containment level facility requirements and operational practices requirements. The risk assessment should include the Risk Group information, the potential for aerosol generation, the quantity and concentration of the agent, the agent's stability and the type of work (e.g., *in vitro*, *in vivo*).

It is the responsibility of the Principal Investigator (PI)/Supervisor in consultation with the BSO to conduct risk assessments and to require the highest appropriate level of containment available for manipulation of specific infectious agents.

3.1 Biosafety Containment Level 1 (CL 1)

Containment Level 1 (CL1) applies to the basic laboratory, which handles agents requiring no special design features beyond those suitable for a well-designed and functional laboratory. Biological safety cabinets (BSCs) are not required. Work may be done on an open bench top, and containment is achieved through the use of practices normally employed in a basic microbiology laboratory.

3.1.1 Standard Microbiological Practices

- Access to the laboratory is limited or restricted at the discretion of the Principal Investigator or Lab Supervisor when experiments are in progress.
- Work surfaces are decontaminated at the beginning and end of the day and after any spill of viable material.
- All contaminated liquid wastes are decontaminated before disposal.
- Persons must wash their hands after handling viable organisms and before leaving the laboratory.
- All procedures are performed carefully to minimize the creation of aerosols.

3.1.2 Special Practices

- Contaminated materials that are to be decontaminated at another location must be placed in a durable, leak-proof container which is closed before being removed from the laboratory (e.g. Biohazardous sharps) Such containers should be wiped with disinfectant solution before leaving the lab.
- An insect and rodent control program must be in effect.

3.1.3 Containment Equipment

- Special containment equipment is generally not required for manipulation of agents assigned to Biosafety Containment Level I.

3.1.4 Laboratory Facilities

- The laboratory is designed so that it can be easily cleaned.

- Bench tops are impervious to water and resistant to acids, alkalis, and organic solvents.

3.2 Biosafety Containment Level 2 (CL 2)

The primary exposure hazards associated with organisms requiring Containment Level 2 are through ingestion, inoculation and mucous membrane routes. Agents requiring CL2 facilities are not generally transmitted by airborne routes, but care must be taken to avoid the generation of aerosols (aerosols can settle on bench tops and become an ingestion hazard through contamination of the hands) or splashes.

Primary containment devices such as BSCs and centrifuges with sealed rotors or safety cups must be used, along with appropriate personal protective equipment (i.e., gloves, laboratory coats, protective eyewear). Environmental contamination must be minimized through the use of handwashing sinks and decontamination facilities (autoclaves).

3.2.1 Standard Microbiological Practices

- Access to the laboratory is limited or restricted by the Principal Investigator or Supervisor.
- Work surfaces are decontaminated at the beginning and end of the day and after any spill of viable material.
- All infectious liquid or solid wastes must be decontaminated before disposal.
- All persons are required to wash their hands after handling infectious materials and before leaving the lab.
- All procedures must be performed carefully to minimize the creation of aerosols. Procedures that create aerosols must be conducted in a biological safety cabinet.
- Lab coats must remain in the laboratory. Lab coats and gloves are to be removed if the worker leaves the laboratory for any reason.

3.2.2 Special Practices

- Contaminated materials to be decontaminated at another location must be placed in durable leak-proof containers that are closed before being removed from the laboratory, e.g. biohazardous sharps
- The PI or Lab Supervisor limits access to the laboratory. In general, anyone at increased risk of acquiring infection is not allowed in the laboratory. The PI or Lab Supervisor is responsible for assessing each circumstance and determining who may enter or work in the laboratory.
- If the use of an infectious agent(s) requires special provisions (e.g. vaccination), a biohazard warning sign must be posted on the access door to the laboratory area. The biohazard warning sign should identify the infectious agent(s), provide the name(s) of the PI and/or other responsible persons, and indicate the special requirements for entering the laboratory.
- An insect and rodent control program must be in effect.
- Special care must be taken to avoid skin contamination with infectious materials; gloves must be worn when skin contact with infectious materials is possible.
- All waste from laboratories must be appropriately decontaminated before disposal.
- Hypodermic needles and syringes may be used only for injection and aspiration of fluids from diaphragm bottles.

- Spills and accidents that result in overt exposure to infectious materials must be reported immediately to the PI and the Biosafety Officer.
- When appropriate (considering the agent handled), baseline serum samples for laboratory personnel should be collected.
- Infectious agents must be stored inside a leak proof container that is clearly labelled.

3.2.3 Containment Equipment

Biological safety cabinets (class I or II) must be used whenever:

- Procedures with a high potential for creating infectious aerosols are conducted. These may include centrifuging, blending, vigorous shaking or mixing, sonic disruption, and opening containers of infectious materials.
- High concentrations or large volumes of infectious agents are used. Such materials must be centrifuged in a biological safety cabinet and opened in a biological safety cabinet.

Air from these cabinets may be recirculated to the room only after passage through a high-efficiency particulate air (HEPA) filter.

3.2.4 Laboratory Facilities

- The laboratory is designed so that it can be easily cleaned.
- Bench tops are impervious to water and resistant to acids, alkalis and organic solvents.
- Each laboratory shall contain a sink for hand washing.
- Coat hooks must be provided near the exit for laboratory coats. Such coats are to be kept separate from streetcoats.
- A biohazard sign with appropriate information must be posted on the entrance of the laboratory.
- A list of all infectious agents being used in laboratory must be posted on the entrance of the laboratory.

All the biological agents used at TRU require Biosafety Containment Level 1 or 2.

NO EMPLOYEE WILL PERFORM BIOSAFETY CONTAINMENT LEVEL 3 OR 4 WORK AT THE UNIVERSITY.

4. GENERAL SAFETY PRACTICES AND PROCEDURES

4.1 General Laboratory Safety Practices

- Employees must be advised of potential hazards before entering the work area. Laboratory doors should be kept closed.
- Safety glasses must be worn in all designated laboratory areas.
- Lab coats must be worn in the laboratory areas by all personnel, including visitors, trainees and others entering or working in the laboratory. (Refer to Section 4.10.2). Contaminated clothing must be disinfected by appropriate means.
- Mouth pipetting is strictly prohibited.
- Eating, drinking, smoking, chewing gum and/or storing food is not permitted in the laboratory areas.
- The lab should be kept clean and free of materials not pertinent to the work.
- Work surfaces shall be decontaminated at least once a day and after any spill.
- Employees must wash their hands after handling infectious materials and before leaving the laboratory.
- Gloves must be worn for all procedures that involve contact with body fluids, infectious materials, or infected animals. Gloves must be autoclaved.
- All spills, accidents and possible exposures to infectious materials must be reported immediately to the PI and the BSO.
- The BSO will ensure that training in laboratory safety for infectious materials is provided.
- All procedures shall be performed carefully to minimize the creation of aerosols.
- All contaminated or infectious liquid or solid materials must be decontaminated before disposal or re-use.
- Where infectious agents are used in a laboratory, a biohazard warning sign incorporating the universal biohazard symbol must be posted on the access door to the work area.

4.2 Cultivation of Bacteria on Laboratory Media

For microbiological investigations it is essential to learn the skills of inoculating specimens onto culture media:

- Always practice aseptic technique
- Clean benchtop with supplied disinfectant before beginning your work and upon completion
- Ensure loops and picks are flamed upon completion of you work
- Discard any waste biohazardous material in the appropriate area, to ensure adequate disinfection is completed

4. 3 Working with Laboratory Animals

By definition, all work involving animals is considered biohazardous. Animals can harbour infectious organisms that can be transmitted to humans.

Laboratory facilities must provide containment for laboratory animals exposed to or harbouring infectious agents that is appropriate to the risk level of the infectious agents involved.

Requirements for the maintenance of animals may differ in scale and degree, but the basic principles for microbiological safety will be similar to those outlined in Section 3 and shall include the following precautions:

1. Infected animals and insects shallbe segregated from uninfected animals and is preferable to separate any handling area from the holding area.
2. Animals or insects in use in an experiment must be maintained at a level of containment that is at least equivalent to the containment level for the biological agent with which it has been infected or treated.
3. Provision must be made to ensure that inoculated animals or insects cannot escape.
4. Dead animals or insects and the refuse from the animal room and cages (e.g. bedding, faeces and food) must be placed in a leak-proof container and autoclaved or incinerated.
5. All cages must be properly labelled, and procedures in the holding area must minimize the dispersal of dander and dust from the animals and cage refuse.
6. Animal care providers while feeding and watering animals or cleaning cages shall wear gloves and eye protection.
7. Gloves, boots, floors, walls and cage racks shall be disinfected as defined by Standard Operating Procedures (SOPs)

In addition to the preceding:

8. All aspects of the proposed use of animals in research must satisfy all the standards and regulations of the Canadian Council on Animal Care (CCAC), and the Animals for Research Act. All work involving animals must receive prior approval from the TRU Animal Care Committee.
9. The appropriate species must be selected for animal experiments.
10. The investigator and/or person(s) responsible for an animal experiment must ensure that all those having contact with the animals and waste materials are aware of and familiar with any special precautions and procedures that may be required. Where possible and warranted, personnel should be protected by immunization with appropriate vaccines.
11. All incidents, including animal bites and scratches or cuts from cages or other equipment must be documented and reported by the employee to the Lab Supervisor.

4.4 Working with Human Pathogens (Infectious Agents)

Some microorganisms (viruses, bacteria, fungi, etc.) are species-specific, selectively infecting and causing disease in one or a limited number of host species. Unrelated and distantly related species may not be similarly affected by the same infectious microorganism due to differences in physiology, metabolism, biochemistry, etc. In general, the risk to laboratory personnel working with a virus that only infects and

causes disease in rodents is lower than the risk to laboratory personnel working with tissues and cells from humans or other primates. If the human material contains a viable pathogen, it will likely be a human pathogen, with the potential to infect and cause disease in another human.

Although a single mode of transmission may predominate, disease-causing microorganisms can be spread or transmitted from one host to the next, directly or indirectly, by a number of methods, including aerosol generation and inhalation, ingestion of contaminated food and water, skin and mucous membrane contact with contaminated surfaces, contact contamination of an open wound or lesion, and autoinoculation via a cut, laceration or puncture with a contaminated instrument.

4.5 Human Bloodborne Pathogens

Human blood is recognized as a potential source of pathogenic microorganisms that may present a risk to workers who are exposed during the performance of their duties. Although the hepatitis B virus (HBV) and the human immunodeficiency virus (HIV) are often cited as examples, a "bloodborne pathogen" is any pathogenic microorganism that is present in human blood or other potentially infectious materials and that can infect and cause disease in persons who are exposed to blood or other potentially infectious materials containing this pathogen.

"Other potentially infectious materials" means material that has the potential to transmit bloodborne pathogens. This includes infected human tissues and the following body fluids: semen, vaginal secretions, cerebrospinal fluid, synovial fluid, pleural fluid, peritoneal fluid, pericardial fluid, amniotic fluid, saliva in dental procedures, and any other body fluid that is visibly contaminated with blood.

4.5.1 Universal Blood and Body Fluid Precautions

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

Body fluids to which universal precautions apply include blood, body fluids containing visible blood, semen, vaginal secretions, cerebrospinal fluid, synovial fluid, pleural fluid, peritoneal fluid, pericardial fluid, and amniotic fluid.

Universal precautions generally do not apply to faeces, breast milk, nasal secretions, sputum and saliva, sweat, tears, urine, and vomitus unless they contain visible blood. Although these materials are not implicated in the transmission of bloodborne pathogens, it is prudent to minimize non-intact skin and mucous membrane contact with these materials.

Hepatitis B immunization is recommended as an adjunct to universal precautions for workers who have occupational exposure to human blood or other potentially infectious materials. This immunization is provided to employees at risk, free of charge.

4.5.2 General Precautions

1. All workers should routinely use appropriate barrier precautions to prevent skin and mucous

membrane exposure when contact with human blood or other body fluids is anticipated.

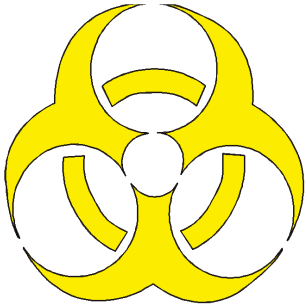
2. Eating, drinking, smoking, applying cosmetics or lip balm, and handling contact lenses are prohibited.
3. Gloves shall be worn when touching blood and body fluids, mucous membranes, or non-intact skin, for handling items or surfaces soiled with blood or body fluids, and for performing venipuncture and other vascular access procedures. If a glove is torn or damaged during use, it shall be removed and a new glove used as promptly as safety permits. Disposable gloves shall not be washed or disinfected for reuse. Washing with surfactants may enhance penetration of liquids through undetected holes in the glove. Disinfecting agents may cause deterioration of the glove material.
4. Masks and protective eyewear or face shields shall be worn during procedures that are likely to generate droplets of blood or other body fluids to prevent exposure of mucous membranes of the mouth, nose, and eyes.
5. Gowns or aprons shall be worn during procedures that are likely to generate splashes of blood or other body fluids. Protective clothing should be removed before leaving the area.
6. Hands and other skin surfaces shall be washed immediately and thoroughly if contaminated with blood or other body fluids. Hands shall be washed immediately after gloves are removed since no barrier is 100% effective.
7. Workers shall take precautions to prevent injuries caused by needles, scalpels, and other sharp instruments or devices during procedures, when cleaning used instruments, during disposal of used needles, and when handling sharp instruments after procedures. Needles and syringes should beused only in those situations when there is no alternative. To prevent needlestick injuries, needles should not be recapped, purposely bent or broken by hand, removed from disposable syringes, or otherwise manipulated by hand. After they are used, disposable syringes and needles, scalpel blades, and other sharp items shall be placed in puncture-resistant containers for disposal. The puncture-resistant container shall be located as close to the use area as practical.
8. Workers who have exudative lesions, weeping dermatitis, cuts, open wounds or other breaks in the skin shall either refrain from all direct contact with blood and other body fluids until the condition resolves, or utilize protective barriers to reduce the risk of exposure.
9. Pregnant workers shall be especially familiar with and strictly adhere to precautions to minimize the risk of perinatal transmission of bloodborne pathogens.

4.6 Biosafety Signs and Labels

The Public Health Agency of Canada and the Medical Research Council (MRC) require that warning signs and/or symbols be used to inform personnel and visitors of the potential of hazards in the workplace. Specifically, with regard to biohazards, the universal biohazard warning sign must be used to "signify the actual or potential presence of a biohazard and to identify equipment, containers, rooms, materials, experimental animals or combinations thereof, which contain and/or are contaminated with, viable hazardous agents."

1. TRU requires that the universal biohazard symbol be used to designate the presence of agents/substances that are believed to be biohazardous.
2. All laboratories and work areas utilizing and/or storing biohazardous substances must have the appropriate biohazardous caution sign posted prominently. If infectious agents are used, a

biohazard sign must be located outside the laboratory door to indicate the nature of the hazard, the biohazard level, special provisions for entry and contact information for the Principal Investigator and/or other responsible person(s).



3. Principal Investigators/Supervisors are responsible for ensuring that all hazard signs are current and accurate. Notify the Biosafety Officer if changes are necessary in laboratory door signage and/or equipment labeling.

4.7 Access/Security Controls

Doors must be locked when laboratories are unoccupied and only authorized persons are permitted to enter laboratory working areas. Children under the age of 14 years must not be permitted to enter laboratory working areas.

4.8 Cell Culture

All new cell lines introduced into TRU must be registered with the Biosafety Officer.

Storage and retrieval of frozen cell cultures from liquid nitrogen require appropriate personal protective equipment. There are three major risks associated with liquid nitrogen (-196 °C): frostbite, asphyxiation and exposure. Gloves shall be worn that are thick enough to provide insulation, but flexible enough to allow manipulation of ampoules.

When ampoules are submerged in liquid nitrogen, a high-pressure differential results between the outside and the inside of the ampoule. If the ampoule is not perfectly sealed, the pressure differential may result in inspiration of liquid nitrogen, which may cause the ampoule to explode violently when thawed. Wear eye protection, a face shield and earplugs are recommended

Biological safety cabinets shall be kept clean and free of unnecessary equipment and material to ensure proper functioning of the cabinet.

Liquid waste shall be decontaminated by chemical disinfectant (e.g., 10% hypochlorite). Vacuum collection flasks for liquid waste shall be kept outside the cabinet in a secure place and should contain an appropriate disinfectant .The collection flask shall also have a back-up trap to protect the central vacuum line. Decanting shall be done to minimize splashing.

All flasks should be properly labelled.

Decontamination of the biological safety cabinet shall be done with a liberal spray with supplied disinfectant followed by a wipe with 70% ethanol at the end of the procedure.

Solid waste shallbe placed in biological waste bags and the bags then sealed for autoclaving. Biohazard

disposal containers with lids shall be used for primary disposal.

Glass pipets shalld be placed in a pipet container with an appropriate disinfectant. Plastic disposable pipets must be disposed of in an appropriate container.

Contaminated sharps shall be placed in the yellow biohazardous sharps containers

4.9 Training Requirements

Biosafety training is mandatory for all new Principal Investigators, supervisors, research staff and students who work with microorganisms, cell cultures and human blood and body fluids. Principal Investigators are responsible for ensuring new employees have received training appropriate to the specific biological materials and/or processes in the lab. This training is to be provided prior to initiation of work and shall be documented by the Biosafety Officer.

On completion of Biosafety training, the participant will:

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

THOSE INVOLVED IN BLOODBORNE PATHOGENS RESEARCH HAVE ADDITIONAL TRAINING REQUIREMENTS.

4.10 Personal Protective Equipment (PPE)

The type and extent of clothing and equipment to be selected for any particular procedure depend on the research operations and levels of risk associated with them. At a minimum, a lab coat, closed-toe shoes, and gloves must be worn in any microbiology laboratory. Lab coats, closed-toe shoes, and gloves prevent biohazardous materials from contact with the skin, including areas where there might be cuts, abrasions, or dermatitis. The legs are a vulnerable area if uncovered, so it is inappropriate to wear skirts or shorts. Closed- toe shoes protect the feet from spills as well as injuries from dropped sharps. Soles must be non slip to avoid slips and falls,

4.10.1 Lab Coats

The lab coat protects street clothing from contamination and prevents possible cross-contamination from any normal flora present on the skin.

Lab coats must be worn by all personnel, including visitors, trainees and others entering or working in the laboratory. Coats must be properly fastened. If contaminated, lab coats shall be decontaminated by autoclaving before being placed in the laundry. If decontamination is not possible, any contaminated coat shall be placed in the biohazard waste container.

4.10.2 Gloves

Appropriate gloves must be worn for all procedures that might involve direct or accidental skin contact

with biohazardous materials. Latex or vinyl gloves offer a high level of dexterity and a higher level of sensitivity; however, they don't offer a great deal of protection from needle sticks, animal bites or sharps. All gloves will eventually permeate and shall therefore be changed periodically. If gloves become contaminated or torn, remove immediately and wash hands with soap.

Some procedures may require double gloving.

Gloves should overwrap the cuff and lower sleeve of the lab coat if double gloving is practiced

Gloves must be removed prior to leaving the laboratory and decontaminated with other laboratory wastes before disposal. The following chart lists preferential gloves for different products**.**

TYPE	ADVANTAGES	DISADVANTAGES	FOR USE WITH:
Natural rubber latex	Low cost, good physical properties, dexterity	Poor against oils, greases, organic solvents, ethidium bromide. May cause allergic reactions.	Bases, acids, alcohols, dilute aqueous solutions. Fair vs. aldehydes, ketones.
Natural rubber blends	Low cost, dexterity, generally better chemical resistance than natural rubber.	Physical properties often inferior to natural rubber. May cause allergic reaction.	Bases, acids, alcohols, dilute aqueous solutions. Fair vs. aldehydes, ketones.
Polyvinyl chloride (PVC)	Low cost, very good physical properties, average chemical resistance.	Plasticizers can be stripped.	Strong acids and bases, salts, aqueous solutions, alcohols, oils, greases and petroleum products.
Neoprene	Average cost, average chemical resistance, average physical properties, high tensile strength, high heat resistance.	Poor vs. chlorinated hydrocarbons	Oxidizing acids, alcohols, anilines, phenol, glycol ethers, solvents, oils, mild corrosives
Nitrile	Low cost, excellent physical properties, dexterity	Poor vs. chlorinated organic solvents	Oils, greases, xylene, aliphatic hydrocarbons, perchloroethylene, trichloroethane, ethidium bromide. Fair vs. toluene.
Butyl	Good resistance to polar organics, high resistance to gas and water vapour	Expensive, poor vs. hydrocarbons, chlorinated solvents	Glycol ethers, ketones, esters, aldehydes, polar organic solvents
Polyvinyl alcohol (PVA)	Resists broad range of organics, good physical properties.	Very expensive. Water sensitive, poor vs. light alcohols, acids and bases.	Aliphatic and aromatic hydrocarbons, chlorinated solvents, ketones (except acetone), esters, ethers
Fluro-elastomer (Vitron®)	Good resistance to organic and aromatic solvents. Flexible.	Extremely expensive. Poor physical properties. Poor vs. some ketones, esters, amines	Aromatics and aliphatic hydrocarbons, chlorinated solvents, oils, lubricants, mineral acids, alcohols.
Norfoil, Silver Shield™, 4H™	Excellent chemical resistance.	Poor fit, stiff, easily punctures, poor grip.	Use for Hazmat work. Good for range of solvents, acids and bases.

5. BIOLOGICAL SAFETY CABINETS (BSCs)

Protection of the respiratory system is of major concern in any biological safety program because infectious organisms can readily enter the human body through the respiratory tract. Engineering controls (biological safety cabinets) are the primary barrier for inhalation of biohazard and should be use whenever possible. Respirators should only be used as secondary means of control. Any individual needing respiratory protection will be required to participate in the Respiratory Protection Program. The BSO will contact the Dean of Science who will ensure respirators will be made to individuals who require such.

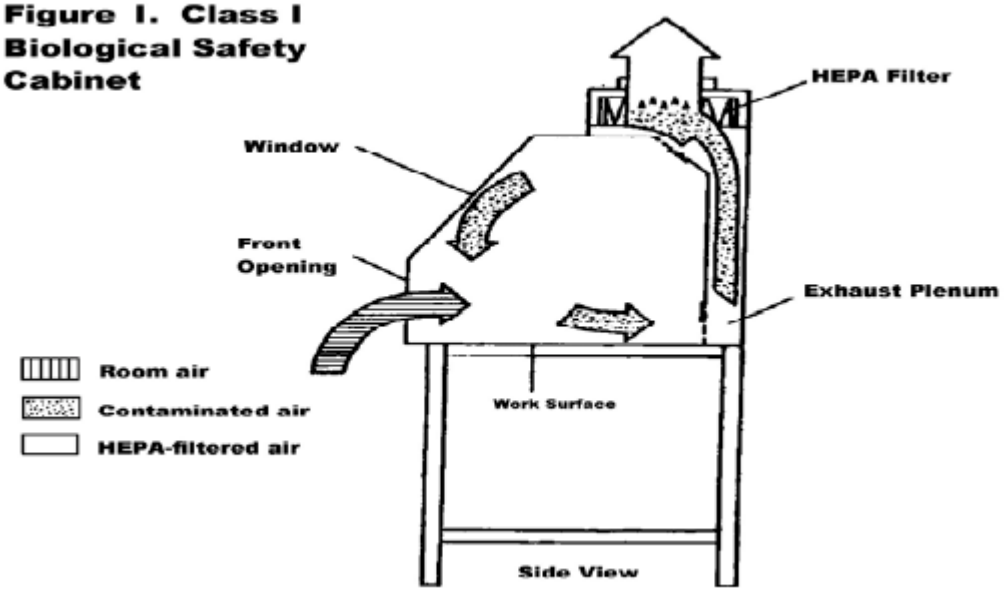
A biological safety cabinet is a ventilated cabinet that uses a combination of HEPA (high efficiency particulate air) filtration, laminar air flow and containment to provide personnel, product and environmental protection from particulates or aerosols involving biohazardous materials. It is distinguished from a chemical fume hood by the presence of HEPA filtration and the laminar nature of the air flow.

The following is a basic description of the two classes of biological safety cabinets, their capabilities and the limitations of each class.

5.1 Class I Biological Safety Cabinet

This is a ventilated cabinet which provides partial protection to the worker and environment but no protection to the work. These cabinets have unrecirculated airflow away from the operator that is discharged to the atmosphere after filtration through a HEPA filter. Chemical carcinogens and low level of radioactive materials and volatile solvents can be used in a Class I safety cabinet.

Figure I. Class I Biological Safety Cabinet



5.2 Class II Biological Safety Cabinet

This is a ventilated cabinet that provides personnel, product and environmental protection. These

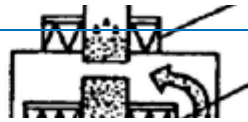


Figure 2 Class II, Type A Biological Safety Cabinet

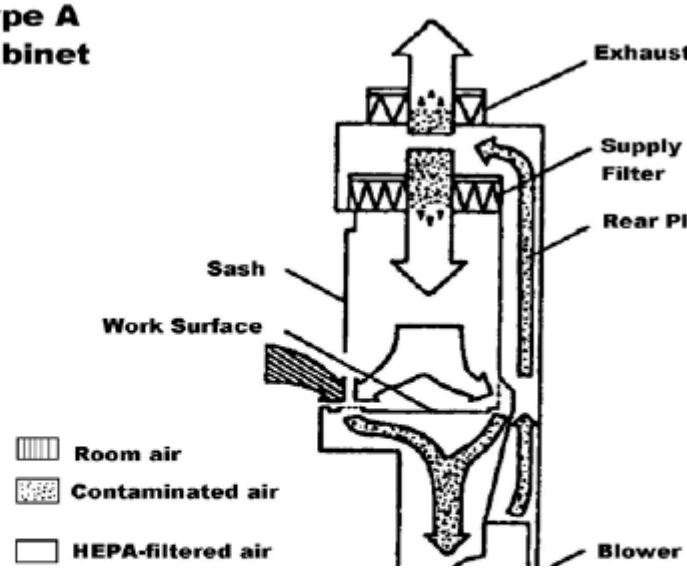


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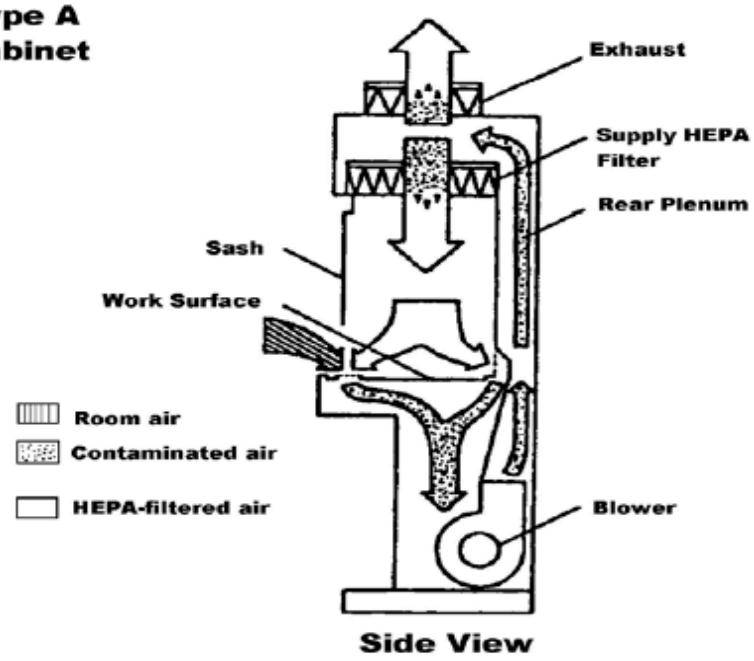
Figure 2 Class II, Type A Biological Safety Cabinet



5.3 Working in a Biological Safety Cabinet

- Turn the cabinet on for at least 10-15 minutes prior to use, if the cabinet is not always operating.
- Disinfect work surface with suitable disinfectant followed by 70% alcohol.
- Consider the materials necessary for the work to be conducted in the cabinet.
- Place all required materials on an absorbent pad to avoid aerosol generation
- Place items into the cabinet so they can be used efficiently without unnecessary disruption of the air flow, working with materials from the clean to the dirty side.

- Wear appropriate personal protective equipment. At a minimum, this will include a buttoned laboratory coat and gloves.
- Delay manipulation of materials for approximately one minute after placing the hands/arms inside the cabinet.
- Minimize the frequency of hand movements in and out of the cabinet.
- If it is required to move out of the BSC pull hands slowly and horizonatally from the cabinet. Allow time for the air flow disruption to diminish before putting hands back into the BSC
- Do not disturb the airflow by covering any grillwork with materials.
- Work at a moderate pace to prevent the air flow disruption that occurs with rapid movements.
- Wipe the bottom and side of the hood surfaces with disinfectant when work is completed.
- Leave the hood running for several minutes following the procedures before turning off the blower

NOTE: BE VERY CAREFUL WHEN USING SMALL PIECES OF MATERIAL SUCH AS KIMWIPES IN THE HOOD. THESE CAN BE BLOWN INTO THE HOOD AND DISRUPT THE MOTOR OPERATIONS.

**** THE BSC IN S365C IS T USE FOR STERILITY ONLY. FOR USE WITH POTENTIAL PATHOGENS USE THE BSC IN S367. NO CHEMICAL USE IS ALLOWED IN EITHER OF THESE BSCs.**

THE BSC IN S363 MAY BE USED WHEN VOLATILE CHEMICALS ARE REQUIRED TO BE USED IN THE BSC

5.4 Certification of BSC

All biological safety cabinets must be certified on installation, when the filters are changed, before and after a move or transfer and annually. Cabinets must not be moved without first undergoing a decontamination process. The Biosafety Officer must approve any modifications to any biological safety cabinets. Cabinets must undergo certification following modification.

The certification process is arranged for annually by the BSO.

If problems are encountered in operating a biological safety cabinet, do not continue to use it; contact the BSO immediately.

The certification of the biological safety cabinets is essential to their safe and effective use.

6. LABORATORY EQUIPMENT SAFE OPERATING PROCEDURES

6.1 Sonicators

When used with infectious agents sonicators can release significant amounts of hazardous aerosols, and shall be operated inside a biological safety cabinet whenever possible.

Sonicators are devices commonly used for disrupting cells and mixing samples. Vortexers are also used for mixing samples. The following safety measures should be used with a sonicator to reduce the chance of aerosol formation.

Safe Operating Procedures include:

- Loosely cap all samples.
- Make sure there is enough water in the sonicator.
- Avoid prolonged sonication.
- Inspect all glassware to be used in the sonicator. Do not use chipped or cracked glassware.
- Routinely replace the sonicator liquid.
- Avoid sonicating volatile compounds.
- When possible, use secondary containment (container within container within the sonicator).
- Perform sonicating in isolated rooms and areas.
- Make sure you have adequate ear protection.
- Allow aerosols to settle for at least one minute before opening containers.

6.2 Centrifuges

Safe use of centrifuges requires proper maintenance and operation. Failed mechanical parts or improper operation can result in release of projectiles, hazardous chemicals and biohazardous aerosols. Maintenance and repairs must be performed only by trained, qualified personnel.

Centrifuges are a source of potential biological contamination due to the rapid speeds and relatively high pressure exerted by such devices. The following safety measures shall be used when using any centrifuge:

Safe Operating Procedure includes:

- Prior to starting, make sure the centrifuge is clean. Do not operate with any material spills in either the body or the rotor.
- Ensure that the interlocking device prevents the lid from being opened when the rotor is in motion and the centrifuge from starting when the lid is open.

- Make sure the centrifuge is level. If a portable model, make sure it is secure on the bench top before starting.
- Inspect all equipment to be placed in centrifuge for cracks or weak areas. Ensure that the tube material provides the necessary chemical resistance and speed rating.
- Use the lowest speed and time setting that will accomplish the job.
- Avoid over-filling tubes.
- Balance all loads.
- Do not open the lid until it comes to a complete stop.
- Wait for at least one minute before opening the lid to remove your sample.
- Should a spill occur, disinfect immediately and dry completely before the next run.
- Periodically inspect centrifuge. Check seal around top, baskets, rotors and wiring.
- Avoid use of volatile materials when possible.
- Plastic centrifuge tubes with seal-forming screw tops should be used whenever possible.
- Centrifuges should not be placed into a biological safety cabinet if the motor produces strong air current because the air turbulence generated may disrupt the laminar airflow.

6.3 Vacuum

If there is a vacuum system serving multiple areas, care should be taken that there are filters in the system, and that there is an overflow trap containing an appropriate disinfectant to prevent entry of contaminated material into the piping system and pumps. It is often best to use either a stand-alone pump-type vacuum system, or to use a water siphon vacuum system that is attached to a faucet (provided that measures are taken to prevent back-siphonage).

6.4 Gas Chromatograph

Gas chromatography (GC) procedures involve the use of compressed gas cylinders and may involve the use of flammable solvents and toxic chemicals. Be familiar with the use and handling of compressed gas cylinders, and with hazardous properties, precautionary measures, and handling instructions for any hazardous materials being used. Refer to MSDSs (found on-line – use the MSDS icon on the desktop) or other reliable reference material. The following guidelines will assist in the safe operation of GCs:

- Wear proper eye protection. GC columns are fragile and breakage could result in small projectiles during handling. As well, samples are prepared in various hazardous solvents that could damage the eyes upon contact.
- When cutting a GC column, be sure that the cut is made away from the body.
- Ensure that GC column cutters are capped or otherwise stored to prevent injury when not in use.

- Discard small pieces of GC columns as sharps waste.
- Ensure that the oven is allowed to cool before installing or removing a column or injector or prior to performing maintenance.
- Ensure that gases are turned off prior to removing or installing a column.
- Test for leaks after the installation of the column and whenever a leak is suspected. Use a technique that will not damage or sacrifice the integrity of the instrument.
- Electron capture detectors (ECD) have a radioactive source and therefore need to be registered as part of the University's Radiation Safety program. Contact Ron Smith ext.5544 for more information about Canadian Nuclear Safety Commission (CNSC) requirements.
- Ensure that the instrument and gases are turned off and the power cord disconnected prior to performing maintenance.

6.5 Ultraviolet lamps

Exposure to ultraviolet light (UV) may result in serious and painful injury to the eyes or skin depending on the wavelength and intensity of the light and the duration of exposure.

- Label all UV light sources conspicuously with the following warning (or equivalent): "Warning – this device produces potentially harmful UV light. Protect eyes and skin from exposure."
- Ensure that the UV light source is shielded.
- Ensure that appropriate PPE is worn and is sufficient to protect the eyes and skin. PPE shall include at least UV resistant face shield, gloves, and lab coat.
- Shielding the equipment or the work area may be warranted.

7. DECONTAMINATION

Decontamination is a process or treatment that renders an instrument or environmental surface safe to handle. A decontamination procedure can be as simple as clean-up with detergent and water or as thorough as sterilization. Sterilization, disinfection, and antisepsis are all forms of decontamination.

This section on disinfection and sterilization deals with the cleaning of work surfaces and the treatment of equipment and biological wastes. The information provided in the following is intended to assist the investigator/supervisor in ensuring that a safe environment is afforded to laboratory personnel, as well as custodial and facilities staff. The initial risk assessment for any project shall include an evaluation of the processes and/or disinfectants to be used to ensure that the biohazardous agents/substances involved in the research are inactivated during spill cleanup, before cleaning equipment for re-use, and before final disposal. Microorganisms vary in their resistance to destruction by physical and chemical means. It is important to distinguish between *sterilization, disinfection and decontamination*.

The objective of any decontamination routine adopted in a biohazard area shall be to kill or inactivate any potential hazardous agent. This is likely to require either treatment with moist heat (autoclaving) or treatment with a chemical agent in liquid form.

7.1 Sterilization

Sterilization is the complete destruction or inactivation of all living organisms. The sterilization process must be validated, and the validation documented. The sterilization process must also be monitored routinely (spore strips, *Geobacillus stearothermophilus* indicator)

7.1.1 Steam Sterilization

To ensure sterilization, steam must reach the material for a prescribed period of time. Containers must be left open to allow for steam penetration.

7.2 Disinfection

Disinfection is defined as the reduction of many or all disease causing microorganisms (refers to the vegetative state, spores survive disinfection in or on a surface or object so that they are no longer considered to be capable of transmitting disease).

7.2.1 Chemical Disinfectants

Chemical agents often provide the only practical means by which effective inactivation of biological material can be achieved. When employed with discretion, and knowledge of its limitations, a chemical disinfectant might be used for any of the following purposes.

1. Cleaning and decontamination of work surfaces.
2. Cleaning and decontamination of equipment.
3. Spill or accident clean-ups.
4. Decontamination of containers and transfer equipment suitable for recycling.
5. Decontamination of experimental wastes.

7.2.2 Decontamination of Biohazardous Materials

a. Do not autoclave 1% sodium hypochlorite. It must be collected as a liquid waste be

decontaminated and disposed to the sewer.

b. Cultures are to be autoclaved before disposal. Ensure caps of containers are loose to ensure steam penetration. Bags of biohazardous waste shall not be tied tightly or knotted.

ALL BAGGED WASTE MUST BE AUTOCLAVED FOR 60 MINUTES AT 121^{O}C Biohazardous waste is to be placed in the red receptacle in front of the autoclave in S365E for processing.

8. DISPOSAL OF BIOHAZARDOUS WASTE

Biohazardous waste is a commonly used term that includes infectious, pathological, microbiological and pharmaceutical actives of unknown toxicity. For specific questions, contact the OH&S office. For the purposes of this manual, biohazardous waste is defined as waste containing material of sufficient quantity that exposure to the waste by a susceptible host could result in an infectious disease. Laboratory waste contaminated with or containing biological agents must be autoclaved or disinfected to inactivate the biological agents prior to disposal or cleaning for reuse.

Biohazardous waste from laboratories includes the following:

1. Human blood and body fluids, including plasma, serum, other blood products, emulsified human tissue, spinal fluids, pleural and peritoneal fluids.
2. Cultures and stock of infectious agents.
3. Items contaminated with infectious agents, such as: disposable culture dishes, devices used to transfer, inoculate and mix cultures, (e.g., pipettes), and disposable bench top covers.
4. Animal blood and materials contaminated with blood from animals are considered biological waste, and although not biohazardous in nature, are to be handled and disposed of in the same manner as biohazardous waste, by being placed in the laboratory's biohazardous waste container. In the case of liquid, waste should be decontaminated with an appropriate disinfectant.

Sharps are also considered biohazardous waste and must be collected in an approved hard-walled sharps container. Disposable needles and syringes must not be replaced in their sheath or guard prior to being deposited into the sharps container. Sharps containers with contents exposed to biological materials must be disposed of by placing the container in the facility's biohazardous waste container.

When sharps containers are full notify the BSO for removal. Do not overfill!

Biohazardous waste resulting from work in Biosafety Levels 1 and 2 must be collected within the laboratory in biohazardous waste bags. These bags must be loosely closed and placed in the biohazardous waste disposal container in the laboratory. Liquid waste must be collected in the laboratory at the point of generation. If possible, all liquid waste should be put in appropriate leakproof autoclavable containers ensuring they are not too full and loosely capped prior to being autoclaved. If it is not possible to autoclave, the liquid waste must be decontaminated and disposed to the sewer.

9. EMERGENCY PROCEDURES

9.1 Spills

Spills of biohazardous substances may constitute a significant and ongoing health hazard if not handled in an appropriate manner. Effective disinfectants must be available in the laboratory at all times and for immediate use. As part of the laboratory safety regimen, each laboratory shall have a spill cleanup plan detailing specific disinfectants and procedures for that area. Clean-up of any spill requires the use of appropriate Personal Protective Equipment.

Since the capacity of most commonly used laboratory culture containers is small, it is anticipated that most spills within the laboratory will be limited in size and therefore minor in nature. Although the specific response will depend on the type and nature of the incident, decontamination and clean-up procedures incorporating the steps outlined below are recommended. If a spill is large or of a nature that cannot be handled by laboratory personnel, call Security at ext. 3333 or 9-911 from any campus phone.

9.1.1 Biohazardous Spill Inside a Biological Safety Cabinet

Spills confined to the interior of a biological safety cabinet should present little hazard provided: a) clean-up is initiated at once, and b) the cabinet ventilation system continues to operate to prevent the escape of contaminants.

Suggested biohazard control procedure:

- Pour a strong disinfectant (sodium hypochlorite) around but not on the spill, and mix the disinfectant with the spilled material cautiously.
- Pour or wipe walls, work surfaces and equipment with a solution of appropriate disinfectant
- Allow to stand for the required contact time for the particular hazard (usually 20-30 min).
- Remove excess decontaminant solution with paper towel.
- Used disinfectant, gloves, clothes, paper towels and contaminated lab coats shall be placed in a biohazard bag and autoclaved.
- Inform the Lab Supervisor and Biosafety Officer of the spill.
- Decontaminate all surfaces exposed to the spill with a suitable disinfectant
- This procedure will not disinfect the cabinet filters, blower etc

9.1.2 Biohazardous Spill Outside a Biological Safety Cabinet

A spill of biohazardous materials outside the containment of a biological safety cabinet could represent significant health risks due to the difficulty in containment and potentially larger volume of the spill.

Suggested biohazard control procedure:

- Pour a strong disinfectant (sodium hypochlorite) around but not on the spill, and mix the disinfectant with the spilled material cautiously.
- Remove contaminated clothing and place into an autoclave bag. Thoroughly wash hands.
- Ensure all laboratory doors are closed.
- Post warning signs on the outside of the laboratory door.

- Inform the BSO
- Wearing personal protective equipment, Pour a strong disinfectant (sodium hypochlorite) around but not on the spill, and mix the disinfectant with the spilled material cautiously.
- Evacuate the laboratory for the time expected to be sufficient for the decontamination of the spilled material (normally 20-30 min).
- Wearing appropriate PPE, carefully place used paper towel into an autoclave bag.
- Decontaminate all surfaces exposed to the spill with a suitable disinfectant.

9.2 Accidents/Incidents

Rapid and accurate reporting of accidents and incidents involving exposure to biohazardous agents is important in identifying potentially hazardous operations and procedures.

- All spills, accidents and overt or possible exposures must be reported in writing to the Principal Investigator/BSO or acting alternate as soon as circumstances permit. The PI/BSO must file the report with the Occupational Health and Safety Office within 24 hours. Actions taken to prevent future occurrences shall be Included in the report.
- Accidents/incidents occurring during transportation of infectious substances are to be reported to the Occupational Health and Safety Office as soon as circumstances permit. **http://www.tru.ca/hsafety/formschecklists.html**
- **Please refer to the following for information re accident reporting . TRU security staff are trained in first aid and can be contacted at ext 5033. The following website contains information on accident reporting:**

http://www.tru.ca/hsafety/incident_reporting.html#How%20do%20I%20report%20an%20incident

9.2.1 Animal Bites and Scratches

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

Worker

The exposed site must be washed immediately.

a) For a needlestick, cut, animal bite or scratch, wash with soap and water after allowing the wound to bleed freely.
b) For a mucous (eyes, nose, mouth) membrane or non-intact skin contact (to cuts, rash, eczema or dermatitis), flush with water at the nearest faucet or eye wash station.

The worker must inform the BSO/Principal Investigator of the exposure incident as soon as circumstances permit.

The worker must seek prompt medical attention at the nearest hospital emergency department or emergency clinic, a medical practitioner of their choosing.

The worker must provide information for a University Accident/Incident/ Occupational Disease Report (obtained from her/his supervisor or the Principal Investigator), describing the incident in detail, including the route of exposure, the emergency actions taken, and a description of the worker's duties

as they relate to the exposure incident.

http://www.tru.ca/hsafety/formschecklists.html

9.2.2 Exposure to Infectious and Communicable Disease Agents

The following emergency response procedures shall be followed when a worker has been exposed to infectious or communicable disease agents via inhalation, a needlestick, cut or puncture wound, via ingestion or mucous membrane contact, or via non-intact skin contact.

Worker

The exposed site must be washed immediately.

a) For a needlestick, cut or puncture wound, wash with soap and water after allowing the wound to bleed freely.
b) For a mucous (eyes, nose, mouth) membrane or non-intact skin contact (cuts, rash, eczema or dermatitis), flush with water at the nearest faucet or eye wash station.

The worker must immediately inform the Lab Supervisor/Principal Investigator of the exposure incident. The worker must seek prompt medical attention at the nearest hospital emergency department or emergency clinic, a medical practitioner of their choosing.

The worker must provide information for a University Accident/Incident / Occupational Disease Report (obtained from her/his supervisor/Principal Investigator), describing the incident in detail, including the route of exposure and the emergency actions taken, and a description of the worker's duties as they relate to the exposure incident. Please see the following URL for details http://www.tru.ca/hsafety/formschecklists.html

Supervisor/PI

Supervisors/Principal Investigators must complete and sign the University Accident /Incident report for employees or contact OH&S re student injuries.

http://www.tru.ca/hsafety/incident_reporting.html#How%20do%20I%20report%20an%20incident

The supervisor must ensure that exposure incidents are reported within 24 hours to the Occupational Health & Safety Office.

The supervisor must refer the affected worker(s) to the nearest hospital emergency department or emergency clinic.

9.2.3 Medical Surveillance/Immunization

- All students working in any area, including field work collecting soil samples, or on projects in the laboratory shall have their vaccinations up to date, including tetanus within 10 years see guide http://www.cdc.gov/vaccines/recs/schedules/adult-schedule.htm#everyone. Contact the **South Shore Public Health Unit** 519 Columbia St., Kamloops BC **Phone:** (250) 851-7300 for information or to arrange for a tetanus shot.
- Please check with BSO if working with sewage samples or body fluids such as blood as further vaccinations may be required.

- Anyone collecting samples from caves must be fit tested with the appropriate respirator to protect against Hanta virus. Once fit tested the respirator must be worn when working in the cave environment.
- Please contact the BSO if you have any underlying condition that may require medical surveillance.

10. Transportation of Biohazardous Material

Transportation of biohazardous materials whether domestically or internationally, is governed by the Transportation of Dangerous Goods Regulations (TDGR) which are administered by Transport Canada. Contained in these regulations are outlines for specific packaging requirements, labeling, documentation, training, and emergency response plans. The TDGR also imparts to Transport Canada the authority to inspect, seize, and administer fines in cases of non-compliance.

10.1 Definitions

10.1.1 Internal Versus External

TRU defines "Internal" as the transportation of biohazardous materials within the university. Internal transportation also refers to local transport by person or by commercial vehicle, i.e. taxi, within a restricted area such as within the city or short distances between cities within the province.

TRU defines "external" as the transportation of biohazardous materials outside the university over long distances nationally or internationally.

10.1.2 Infectious Substances

Under the TDGR a biohazardous material is categorized under Class 6 - Poisonous and Infectious substances. Infectious substances are defined as substances containing viable microorganisms or their toxins, which are known or suspected to cause disease in animals or humans.

There are no regulations governing the internal transportation of biohazards belonging to Risk Groups 1 and 2. There is, however, a restricted list of organisms in Risk Group 2 that are regulated by Transport Canada and Health and Welfare Canada. Such organisms include Hepatitis B virus or Human Immunodeficiency virus (HIV).

10.2 Infectious Substances: Internal Transportation Packaging Requirements

10.2.1 Within the laboratory

The work area and procedure(s) shall be arranged so as to minimize the number of required moves from one place to another within the laboratory and to reduce the possibility of breakage or a spill, or, if a spill should occur, to effectively contain the biohazard.

Unbreakable and/or watertight containers or doubling of containers shall be used.

The use of absorbent material around or underneath the containers shall be also considered.

The degree of protection should depend on the level of risk should the biohazardous contents spill.

10.2.2 From One Laboratory to Another

If the load is light or of a small volume, then use baskets, trays, or a secondary container to carry the

primary watertight container housing the biohazardous material.

Absorbent material shall be used between the two containers or as lining for the basket or tray, or the secondary container may also have a cover.

Heavier loads shall be transported by cart with the material arranged in such a way as to minimize any loss of material should the cart strike an unseen object, wall, door, bump, etc.

Laying absorbent material under and/or around the biohazardous material or placing the material in a secondary container/tray, with absorbent material in between or as a lining, should also be considered.

10.2.3 Outside the Facility

The efficient and safe transfer of infectious substances requires good co-ordination between the sender, carrier, and receiver to ensure safe and prompt transport and arrival in proper condition. It is important that the sender make advance arrangements with the carrier and the receiver to ensure that specimens will be accepted and promptly processed. In addition, the sender must prepare the appropriate dispatch documents according to the Transportation of Dangerous Goods Act and Regulations. The sender shall also forward all transportation data to the receiver. No infectious substances shall be dispatched before advance arrangements have been made between the sender, the carrier and the receiver, or before the receiver has confirmed with national authorities that the substance can be imported legally and that no delay will be incurred in the delivery of the consignment to its destination.

Under the Transportation of Dangerous Goods Act and Regulations, biological agents and microorganisms belonging to Risk Groups 2, 3, and 4 as identified and listed in Laboratory Biosafety Guidelines, are classed as "infectious substances" in Division 6.2. Very specific packaging and documentation requirements must be met before such materials may be shipped from TRU. A certified packaging system (Saf-T-Pak, or equivalent) suitable for the legal transport of an "infectious substance" must be used. Risk Group 1 microorganisms are not subject to these regulations.

Appendix A: TRU Level II Biohazard Material Inventory

Biohazardous Material

List material that is Risk II or material in Risk Group Level I but used quantities of more than 10 Litres. For Example:

a. Sewage Sludge would be Risk Group II
b. *E. coli* pathogenic strains would be Risk Group II
c. *E. coli K12* would not be entered (Risk Group I organism) except if produced in individual quantities of 10 or more Litres
d. Human Embryonic Kidney Cells (HEK) would be would be Risk Group II
e. Microsystin is a microbial toxin and is Risk Group II
f. If you are unsure include the material in the list and the BSOwill assist with hazard classification

Hazard Classification Method

Process used to determine the risk group of the material

Method	Code	Example
Supplier Information	**S**	Enter code "S" if a Hela Cells were purchased from ATCC and listed in their catalogue as Risk Group II Material
Other Researcher	**R**	Enter code "R" if a cell line was received from a researcher where a risk assessment was done and the material was categorized as Risk Group II.
Guides	**G**	Various guides by CDC or Public Health Agency of Canada list organisms in risk groups. **http://www.phac-aspc.gc.ca/publicat/lbg-ldmbl-04/ch2-eng.php#jmp-lan23**
Internal Review	**I**	The researcher has completed their own internal review process using Public Health Agency of Canada guidelines
Needs to be Reviewed	**N**	Enter code "N" if the risk group needs to be determined in conjunction with the Institutional Biosafety Committee
Other	**O**	

Received From / Date

The Company, institution, researcher or collection area that the material originated from as well as the date received.

For Example:
1. Enter "University of Toronto, Dr. J. Smith / January 19/11" if the material was donated, collected, or purchased from U of T by Dr. J. Smith from University of Toronto on January 19, 2011.
2. Enter "South Thompson River, Pritchard / January 19/10" if the material was collected from the South Thompson River near Pritchard on January, 19, 2010.

3. Enter "ATCC / January 19/09" if the material was purchased by ATCC on January 19, 2009.

Immunization/ Medical Surveillance

Mark this box "Y" if medical surveillance or immunization is required.

For Example:

a. Enter "Y' if samples of sewage were collected from the Kamloops sewage treatment plant. Immunization would be required for Polio, Tetanus, Diphtheria and Hepatitis A &B

b. Enter "N" for HELA cells purchased from ATCC

c. IF you are unsure enter "?"

Type of Work

Type of Work	Code	Example
Animal	**A**	Enter code "A" if material is used on animals or risk group II animal are being used
Tissue Culture	**TC**	Enter code "TC" if material is used for tissue culture
Production	**P**	Enter code "P" if large scale production methods are used (greater than 10 Litres)
Bacteriology/Virology	**B**	Enter code "B" if the work involves the collection or manipulation of Risk Group II bacteria or viruses
Manipulation of Genetic Material	**G**	Enter code "G" if the work involves transferring genetic material from one organism to another
Other	**O**	

TRU Biohazardous Organisms Inventory

Environmental Research : The following organisms and products are classified as biosafety level 1

Dr J VanHamme's research organisms **All organisms are located in -70 freezer in S363 (restricted area)**

UAMH 7308 *Bjerkandera abusta*,

UAMH 8258 *Bjerkandera abusta*,

UAMH 7972 *Pleurotus ostreatus*,

UAMH 8260 *Coriolopsis gallica*,

UAMH 8272 *Trametes versicolor*

- NB11 03/26/05 (green)
- NB7 03/26/05 (blue)
- NB13 03/26/05 (green)
- NB9 03/26/05 (green)
- NB4-1Y June 22/07 Jon (yellow)
- NB4-2W June 22/07 Jon (white)
- NB4 03/26/05 (red)
- NB14 Mar 26/05 (red)
- NB12 03/26/05 (yellow)
- NB3 03/26/05 (light blue)
- NB8 03/26/05 (yellow)
- NB1 03/26/05 (light blue)
- NB10 03/26/05 (blue)
- NB6 03/25/05 (red)
- NB2 03/25/05 (light blue)
- NB5 03/25/05 (red)
- NB5 03/25/05 (red)
- RHA1 Sept 21/07 (red)
- Cornbacterium maltoromaticum LV17 producer (yellow)
- Cornobacterium divergens LV13 indicator (yellow)
- Bascillus ATCC 53969 Feb 1/01 (yellow)
- SE 1906-12 SoxBC E.coli (red)
- SE 19-02-11 SoxBC E.coli (red)
- SE 1902-10 SoxB E.coli (red)
- SE1902-9 SoxB E.coli (red)
- Rhodococcus CW25 June 25/03 E. Dabbs (light blue)
- E.coli PKS06-1 P. Lau, BRI June 25/03 (blue)
- DH5 **!** pk5''G1 Peter Lau #2 Dec 4/02 (blue)
- Rhodococcus SQ1 E. Dabbs June 25/03 (red)
- E.coli MM294-4 June 25/03 (yellow)
- JVH1 9473498 (red) x2
- MM294-4 pDA71 E. Dabbs June 25/03 (blue)

- MM294-4 4# May 06 (green) x2
- Feco-46-9 (green)
- Rhodococcus Rhodocorus ATCC 53968 Feb 1/01 (green)
- Rhodococcus sp. ATCC 55310 Feb 1/01 (red)
- Feco4b-64 (blue)
- Feco4b-7 (light blue)
- Rhodococcus sp. ATCC 5309 Feb 1/01 (light blue)
- JVH1 UO7P June 25/03 (yellow)
- JVH1-UO1 June 25/03
- FSA-4* (green)
- Feco-4b-8 (green)
- 4 tubes JVH1 comp
- NB41Y culture Nov 5, Genomic prep and library production
- S phase comp NB41Y x 12
- E phase comp NB41Y x 14

- NB41Y Electrocompetent cells

- Pediococcus acidilactici Nisin 2 indicator (white)
- Pediococcus acidilactici Nisin 2 indicator (white)
- L. lactis 11454 (green) x2
- L. lactis sub sp. Lactis var diacetylactis Nisin 2 producer (blue) x2
- S. Uberis Nisin U producer (yellow) x2
- L. sakei ATCC 15521 N.A Indicator (red) x2

- Inuoue competent E.coli MM294-4
- Electrocompetent E.coli MM294-4
- CaCl competent E.coli MM294-4

- Fosmid clone containing protein spot 293 & 354---(Fosmid 1-5G)
- Fosmid clone containing protein spot 609---(Fosmid 9-5A)
- Fosmid clone containing protein spot 609, 332, & 508---(Fosmid 7-4F)

Organisms Used in Teaching Laboratories .

All stock organisms are kept frozen in -70 degree freezer in S363. When the organisms are used the plates are stored in fridge in S365D or S367. Both of these rooms are restricted areas.

Organisms Used in Teaching Laboratories	Biosafety level	Received from
Bacillus sp	1	
Bordetella bronchiseptica	2	
Candida albicans	2	
Citrobacter diversus	2	
Citrobacter freundii	2	
Clostridium septicum	2	
Clostridium novyi	2	
Clostridium perfringens	2	
Clostridium septicum	2	
Enterobacter sp	2	
Enterococcus sp	2	
Enterococcus sp (VRE)	2	ATCC
Erysipelothrix rhusiopathie	2	
Escherichia coli	2	
Flavobacterium sp	2	
Klebsiella pneumoniae	2	
Listeria monocytogenes	2	
Mannheimia haemolytica	2	ATCC
Microsoccus sp	1	
Nocardia sp	2	
Pasteurella multocida	2	
Proteus mirabilis	2	
Proteus vulgaris	2	
Providencia rettgeri	2	
Psuedomonas aeruginosa	2	
Salmonella enteritidis	2	
Staphylococcus aureus	2	
Staphylococcus aureus (MRSA)	2	ATCC
Staphylococcus epidermidis	1	
Staphylococcus hyicus	1	
Staphylococcus intermedius	1	
Streptococcus pyogenes	2	
Streptococcus agalactiae	2	
Streptococcus pneumoniae	2	
Streptococcus viridans	1	
Stenotrophomonas maltophilia	2	

The teaching organisms have been part of a culture collection started before HTPA so it is not known where they were received from.

Fungal Organisms	**Biosafety level**	**Received from**
Microsporum canis	BSL 2	ATCC
Trichyphyton sp	BSL 2	ATCC

Other

Organisms	**Biosafety level**	**Received from**
Research soil samples	BSL 2*	
Fish cells (for cell culture)	BSL 2	
Drosophila cells (for cell culture)	BSL 2	

* (based on not knowing the identity of microorganisms) Used by Dr N Cheeptham's research in S365D, a restricted area. The samples have been collected from caves in Well's Grey Park. They are collected and tested to identify possible organisms that may show activity against pathogenic bacteria

Currently with HPTA, any biohazardous agent brought onto campus must meet with the approval of the TRU Biosafety Committee. All researchers keep inventories of the contents of their freezers, fridges etc. These inventories are electronic and forwarded to the Biosafety officer. It is BSO's responsibility to maintain the current inventory with any additions or deletions.

Appendix B : Biohazardous Materials Use Application Form

Use of Biohazardous Materials in Research or Teaching

- This form is to be completed prior to use of potentially biohazardous materials in laboratory sessions or research projects.
- The completed form is to be forwarded to the Biohazard Safety Officer.
- Ensure the application is completed before:
  - Starting new projects
  - Renewing existing projects (complete application every 3 years)
  - Changing what biohazardous materials are being used in projects

Principal Investigator: _____

Phone:_____

Department_____

Email _____

Funding Source or agency

Source_____Grant No._____ Start Date_____ End Date_____

Is this a renewal with previously approved procedure without alterations? ☐yes ☐ no

Approval end date:

New Funding Source, project previously reviewed under prior application

Agency _____ Approval End Date_____

New Project, Not previously reviewed ☐yes ☐ no

Approved Project, change in biohazardous material or procedures ☐yes ☐ no

Work/ project involving biohazardous materials in teaching ☐yes ☐ no

The TRU Biosafety Committee approves the experimental procedures proposed and certifies with the applicant that the experiment will be conducted with the principles outlined in the most current edition of the Public Health Agency of Canada's "Laboratory Biosafety Guidelines" and in the TRU" Biosafety Laboratory Safe Work Procedures" **? include the manual?**

Containment Level One _____ Containment Level Two _____ (Check One)

Principal Investigator or Course Instructor

_____ _____ Date_____

(Print name) (signature)

Approved by Biosafety Committee _____ Date_____

Date of Expiry _____ Permit Number _____

Personnel Using Biohazardous Material

Name	Department	Job Title	Date Trained in Use of Biohazardous Materials

Briefly describe

1. Biohazardous materials used and designated biosafety risk (check with biosafety officer if not sure)

2. The procedures being done using biohazardous materials

3. Protocols for decontaminating spills

4. Does the protocol present conditions that could increase the hazards (e.g. handling large volumes of high concentrations of pathogens)?

5. If using genetically engineered organisms in specific procedures, is there a history of safe use?

6. What precautions will be used to reduce the production of aerosols or infectious droplets

7. Will the biohazardous material being used expose personnel to risks that may require special training, vaccination or other specific protective measure? If so please explain.

8. Will the project result in combined waste ie radioactive biohazardous waste, tissue contaminated with toxic chemicals? If yes, explain disposal handling.

Appendix C: Student Working in Restricted Areas Contract

I have attended the safety/security orientation and completed all the required assignments.

I understand and will comply with Thompson Rivers University's biosafety safe working procedures and security protocols as described.

I know that if I have a medical condition, including a suppressed immune system, or if I have a medical concern, I must seek advice from the physicians at the campus Medical Clinic by calling 5126.

I recognize my responsibility to observe these practices and precautions while present in the laboratory and understand their importance for the safety and welfare of myself, all others in the laboratory, and the environment.

Name:_____

Email_____

Date:_____

Signature_____

Supervisor:_____

Signature: _____

Checkout Signature Biosafety Officer_____

Date_____

The following disciplinary actions may be taken for infractions of safety or security in the areas listed above. Such actions may result in an incomplete for your research or Honours project. When working in this area, infractions place yourself, co-workers and possibly the public at risk For that reason, infractions to safety cannot be tolerated.

First Warning:	verbal warning
Second infraction:	suspension of working privileges in the area for 1 week
Third infraction:	working privileges suspended for the area indefinitely

When you have completed your project, arrange for a tour with Carolynne to ensure you have left the area clean and free from hazardous wastes. This form will be left with the Faculty of Science Administrative Coordinator and must be completed before you receive your deposit for your swipe card.

Appendix D: Risk Assessment Tool

The following information is used to determine risks on campus at TRU. This process can easily be adapted to assess the biosafety level of biohazardous agents as well. Once the risk assessment has been done the risk is controlled by:

- Eliminating the hazard
- Substituting with a less hazardous agent that could serve the same purpose
- Engineering controls (biosafety cabinets)
- Administrative controls (education, signage, safe work procedures, training in use)
- Personal Protective Equipment (lab coat, gloves, face shields etc.)

http://www.tru.ca/hsafety/riskmanagementandinspections.html

Risk Management

Risk Management involves having a systematic process for addressing hazards in the workplace. It is the process of:

- **indentifying** any foreseeable hazard – anything in the workplace that has the potential to harm anyone at the workplace, e.g. moving parts of machinery, toxic chemicals, manual handling tasks.
- **assessing** the risk from the hazard – finding out how significant the risk is, e.g. will it cause serious injury, illness or death and how likely is it to occur.
- **eliminating** the hazard or if this is not possible, controlling the risk from the hazard – implementing strategies to eliminate or control the hazard, e.g. design equipment differently, add machine guards, use safer chemicals, provide lifting devices to minimize manual handling or use of personal protective equipment.
- **reviewing** the risk assessment to monitor and improve control measures and find safer ways of doing things.

Risk Assessment

TRU uses a quantitative tool to identify high, medium and low risks. The tool takes into account the severity and probability of the risk as well as taking into account any countermeasures that TRU already has in place. The tool can be found on the TRU website at:
http://www.tru.ca/hsafety/riskmanagementandinspections.html#How%20do%20I%20assess%20the%20risk?