UNBC UNIVERSITY OF NORTHERN BRITISH COLUMBIA

Manual

This manual does not address chemical, physical and/or radiation hazards that are commonly encountered in a laboratory setting and is to be regarded as an addendum to the UNBC Chemical Laboratory Safety and Methodology Manual.

FOREWORD

This reference manual outlines the safe use, storage, handling, waste disposal and emergency management of bio-hazardous materials for the University of Northern British Columbia (UNBC) campuses and for its students and workers in the field. This information supplements and reiterates information provided in the Workplace Hazardous Materials Information System (WHIMIS) and the Occupational Health and Safety (OHS) Regulations. If there is a difference between this manual and current WHIMIS and OHS policy, those government policies take precedence.

ACKNOWLEDGEMENTS

The materials used to develop this program were provided by a variety of reliable sources. We gratefully acknowledge the contributions of other universities.

DISCLAIMER

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Emergency Numbers

UNBC Prince George Campus

Security and First Aid 3333

(Security will contact additional personnel as required)

Non – Emergency Numbers

UNBC Prince George Campus

Biological Safety	25530 or 25279
Chemical Safety	26472
Chemstores	26472
Radiation Safety	25530 or 26472
Health and Safety Manager	25530
Security	27058

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CHAPTER 1 - INTRODUCTION

The purpose of this manual is to clarify policies and procedures for safe handling of biohazardous material within the UNBC campus in a manner that will encourage research and further student interest in the field of science. In addition to protection of UNBC personnel, these policies and procedures are in compliance with applicable public regulations to protect the health of the general community and to prevent environmental contamination.
This must be done in compliance with applicable regulations and standards of the Public Health Agency of Canada, the Canadian Food and Inspection Agency, the Workplace Hazardous Material Information System and involved funding agencies.

1.1 Definitions

AUTOCLAVE- Is an apparatus for sterilization by steam pressure. This is the most commonly used method for decontaminating biohazardous material at UNBC. Autoclaving provides heat and moisture as physical factors used to destroy organisms. Most organisms can be destroyed using a steam pressure of 121^0 C for a minimum of 15 minutes. Such laboratory wastes as petri dishes, pipettes, culture tubes, glassware, etc. can be effectively decontaminated this way.

BIOHAZARD- Refers to materials of a biological nature that pose a hazard, including biologically active agents (e.g., venoms, allergens) and pathogens that can cause disease/death/illness in humans, animals or plants. It also refers to materials of a biological nature that cause damage to the environment (e.g., certain recombinant and synthetic DNA sequences). Any substance or material, including human/animal tissues, bodily fluids, cell cultures etc. that are infected, synthesized or manipulated with a pathogen, is considered a biohazard.

BIOHAZARDOUS MATERIAL – Any substance which contains or potentially contains: a biohazardous agent.

BIOLOGICAL- Refers to anything that relates to life, living and/or biology. They can be natural processes or manmade preparations, such as drugs, vaccines, or antitoxins that are synthesized from living organisms or their products and used medically as a diagnostic, preventive, or therapeutic agents. Not all biological substances are considered pathogens. A biological becomes a biohazard only when it poses a threat to the health of organisms or the environment.

BIOLOGICAL SAFETY CABINETS- A biological safety cabinet (hood) is an enclosure designed for the containment of biological hazards. Special filters that remove potentially dangerous particles from the cabinet air provide various degrees of safety and sterility for users and cabinet contents.

BIOETHICS - The study of the ethical and moral implications of new biological discoveries and biomedical advances, as in the fields of genetic engineering, drug research, medicine etc.

BIOSAFETY- Containment principles, technologies and practices implemented to prevent unintentional exposure to pathogens and toxins, or their unintentional release.

BIOSAFETY OFFICER (BSO) – Ensures compliance of the program. The backup Biosafety Officer may do any required tasks as delegated by the primary BSO, and has full authority to complete any tasks as required when the primary BSO is available or not.

BIOSECURITY- Biosecurity is the application of risk mitigation strategies designed to reduce the risk of deliberate or negligent biosecurity events from occurring. Risk mitigation strategies are designed and implemented to prepare for, rapidly detect, respond to, and recover from biosecurity events. It is important to note that biosecurity goes beyond protecting pathogens and toxins, it includes the security of biological research and knowledge. Biosecurity events include theft, accidental loss, misuse, diversion, and intentional release of pathogens, toxins, and related assets.

BIOHAZARD- Any pathogen (micro-organism, parasite, primate body fluids, toxins, animal tissue/dander, cell cultures with infectious agents or prions) that is capable of causing a disease in humans and animals. See 'Pathogen.'

CHEMICAL DISINFECTION- This is an effective alternative to autoclaving for large spaces, surfaces and equipment as well as temperature sensitive containers. The initial choice of chemical disinfectant depends upon the resistance of the microorganism of concern and can be a hypochlorite, phenolic, iodine, alcohol, acid or base. A common disinfectant is 1:9 dilution of household bleach or 70% ethyl or isopropyl alcohol.

CONTAINMENT - Is the control of Biohazard material by isolation and separation of an organism from the worker. It is used to describe the measures used to provide a barrier between the infectious organism(s) and the worker.

DECONTAMINATION- The use of physical or chemical means to remove or inactivate biohazardous materials on a surface or item, rendering it safe to be handled. The most common agents of decontamination are liquid chemical disinfectants (bleach, Virkon, Clydox) or physical methods (radiation, filtration). <u>These items are not to be considered sterile.</u>

DISINFECTION- The removal of <u>all or almost all</u> pathogens from a surface. <u>These items are not to be considered sterile.</u>

DUAL USE – Public Health Agency of Canada (PHAC) defines dual-use potential as the qualities of a pathogen or toxin that allow it to be either used for legitimate scientific research purposes or intentionally misused as a biological weapon to cause harm.

DRY HEAT- Dry heat is used for sterilization of anhydrous oils, greases, powders, etc. that cannot be permeated by steam. Because it is less efficient than steam, it requires a temperature of 160 – 170 ^{0}C for a period of 2 - 4 hours

FUME HOODS- A chemical fume hood is a well-ventilated, enclosed chemical and fire resistant work area that provides user access from one side. It isolates and removes toxic and noxious vapors from a working area and protects the user, it may contain liquids that are splashed or sprayed.

HAZARD- Is a source that has a potential for causing harm. A Hazard is not a risk without a specific environment or situation.

HEPA FILTER- (High Efficiency Particulate Air Filter) an air filter that must remove 99.97% of all particles 0.3 micrometer from the air that passes through it.

LARGE SCALE – Activities that involve working with volumes of toxins or the in vitro culture of infectious material on a scale of 10 litres or greater. This could be a single vessel with a volume of 10 litres or greater, or based on the processes and pathogen used, could be multiple vessels with a total volume of 10 litres or greater. Large scale production areas may require increased containment measures to be put in place.

LOCAL RISK ASSESSMENT (LRA) – Site-specific risk assessment that identifies hazards based on the toxins or infectious material in use and the procedures being performed.

PATHOGEN - Refers to the actual disease producing/infectious agent. It can include such microorganisms as: virus, bacterium, prion, parasite or fungus that causes disease in its host. The host may be an animal, human, plant, or even another microorganism.

PATHOGEN SAFETY DATA SHEETS (PSDS) - Pathogens used to be classified under MSDS (Material Safety Data Sheets) to be more accurate they are now classified with the Pathogen Safety Data Sheets. Each pathogen will be classified within one of the 4 risk groups.

RISK- The likelihood of an event with a hazard that has consequences.

RISK ASSESSMENT- A management process which assesses the quantitative and qualitative value or risk related to a concrete situation and a recognized hazard. There are 3 key steps involved to ensure a quality assessment which include, *assessment* (identifying all the hazards you will be working with and the environment with which you are working in), *mitigation* (providing strategies to decrease the likelihood of an accident happening) and *performance* (documentation and follow up of implemented mitigations to be sure they are continuously effective, monitored and modified when necessary).

RISK GROUPS- Organisms are classified into 4 levels based on how hazardous or infective an organism is/or maybe.

RISK TOLERANCE – The level of risk that an organization is willing to accept

PERSONAL CONTAINMENT - Refers to the standard worker practices used to reduce the spread of microorganisms.

PHYSICAL CONTAINMENT - Includes the laboratory design features and the physical barriers that workers use to isolate biohazards.

PRIMARY CONTAINMENT - The protection of personnel and the immediate laboratory environment from exposure to infectious agents by provision of a physical barrier

SECURITY SENSITIVE BIOLOGICAL AGENTS (SSBAs) –A subset of human pathogens and toxins that have been determined to pose an increased biosecurity risk due to their potential for use as a biological weapon.

SECONDARY CONTAINMENT – Special facility building designs and the use of strict operational practices to protect the external environment from exposure to infectious materials used in a laboratory

STANDARD OPERATING PROCEDURES (SOP) – A set of step by step written instructions for performing a variety of routine operations.

STERILE - An item that is free from living germs or microorganisms; aseptic.

STERILIZATION- A procedure that destroys virtually all viable organisms on a surface. The most common agents of sterilization are heat (autoclaving) or gas (ethylene oxide). When properly done – these items are sterile

TOXINS – potent, biologically produced poisons that may have specific host targets. Symptoms of toxin exposure is similar to infections with the host organism. However, toxins are generally more hazardous than handling its parent organism.

1.2 Human Pathogens and Toxins Act (HPTA)

The Public Health Agency of Canada (PHAC) is the national authority on biosafety for human pathogens and toxins.

PHAC introduced the *Human Pathogens and Toxins Act (HPTA)* in June 2009. The Act establishes legal prohibitions and authorities to govern human pathogens and toxins in Canada. It is designated to protect the health and safety of the public against the risks posed by human pathogens and toxins, while allowing science and research to progress.

- Mandatory registration of institutions that have any Risk Group 2, 3, or 4 or Toxin from Schedule 1.
- Teaching institutions must comply
- Can be audited at any time by federal authorities or funding agencies
- Always do a Risk Assessment before working with any pathogen
- Non-pathogenic variety of E. coli (K-12) is exempt as a Risk Group 2
- Natural occurring pathogens and toxins are exempt
- Pathogen Safety Data Sheets (PSDS) replace Material Safety Data Sheets (MSDS)

To see the whole Act, refer to Public Health Agency of Canada's website:
http://lois-laws.justice.gc.ca/eng/acts/H-5.67/page-1.html

1.3 Governmental Policies and Regulations

In order to protect against accidents and occupational hazards, employees and students must comply with all relevant governmental regulations while conducting their various work and

research tasks whether they are working on or off campus. These regulations include, but are not limited to:

- Public Health Agency of Canada, Laboratory Bio-safety Guidelines
  http://www.phac-aspc.gc.ca/publicat/lbg-ldmbl-04/index-eng.php

- Canadian Food Inspection Agency (CFIA), Biohazard Containment and Safety
  http://www.inspection.gc.ca/animals/biohazard-containment-and-afety/eng/1300121579431/1315776600051

- Global Affairs Canada
  https://www.international.gc.ca/gac-amc/index.aspx?lang=eng

- Canada Border Services Agency
  https://www.cbsa-asfc.gc.ca/menu-eng.html

- The Occupational Health and Safety (OHS) Regulations
  http://www.worksafebc.com

- Transportation of Dangerous Goods Regulations
  http://www.tc.gc.ca

- Workplace Hazardous Materials Information System
  http://www.hc-sc.gc.ca

- The Nuclear Safety and Control Act and Regulations
  http://www.cnsc-ccsn.gc.ca

- Canadian Environmental Protection Act and Regulations
  http://www.ec.gc.ca

- British Columbia Environmental Management Act and Regulations
  http://www.bclaws.ca

- The National Fire Code of Canada
  https://nrc.canada.ca/en/certifications-evaluations-standards/codes-canada/codes-canada-publications/national-fire-code-canada-2015

- British Columbia Fire Code
  http://www.housing.gov.bc.ca

1.4 UNBC Policies

In addition to government regulations, UNBC laboratory personnel are required to comply with UNBC policies such as:

- Occupational Health and Safety Policy

- Hazardous Waste Identification, Reporting, and Disposal Policy
- Radionuclides and Radiation Hazard Policy
- Respectful Workplace and Learning Environmental Policy
- Protective Clothing and Equipment Policy
- Safety Training Policy
- Fire Safety Policy
- Fieldwork Safety Policy
- Peroxide-Forming Chemicals Policy
- Cryogenic Liquids Storage, Handling, and Transport Policy

These policies can be viewed at https://www.unbc.ca/policy

1.5 UNBC Safety Committees

- University Joint Occupational Health and Safety Committee
- Research Ethics Board
- Laboratory Safety Committee (Biosafety)
- Radiation Safety Committee
- Field Safety Committee
- Green University Committee
- Animal Care and Use Committee (ACUC)

Additional information and these committees can be found at:
https://www.unbc.ca/provost/university-committees

1.6 Compliance and Enforcement

The University of Northern British Columbia shall comply with the various terms and conditions of all licenses and permits issued to the Institution. This also includes following all applicable Federal and Provincial statutes, and funding agencies agreements pertaining to the use, handling, storage and disposal of biohazardous materials.

Non-compliance with legislated and University requirements can result in significant penalties and fines for the University and its employees as individuals as well as severely impacting funding for research.

1.6.1 Non Compliance

1. On the **first** occurrence of non-compliance, the UNBC Laboratory Safety Committee will send a written notification to the Principal Investigator or Instructor with copies to the CSAM Dean and Program Chair, and the UNBC Joint Health and Safety Committee outlining the nature of the infraction. Immediate response to and correction of the violation is required within the time frame specified on the notification.

2. On the **second** occurrence of the non-compliance within a twelve-month period or when there is no response to the first infraction with in the specified time, the UNBC Laboratory Safety Committee will notify the UNBC Joint Health and Safety Committee who will suspend privileges to obtain and use biohazardous material. The Principal Investigator or Instructor may have this

privilege restored upon written verification from the CSAM Dean indicating rectification of the infraction. A copy will be forwarded to the appropriate Program Chair and the President's Council.

3. On the **third** occurrence of non-compliance within a twelve- month period, the permit or license (if applicable) will be revoked and research activity suspended by the UNBC Joint Health and Safety Committee. The Principal Investigator or Instructor may appeal by conducting a meeting with the representatives of the UNBC Joint Health and Safety Committee and the President's Council. Written notification of the above actions will be sent to the CSAM Dean and the Program Chair.

1.6.2 Unacceptable Risk

When, in the opinion of the Biosafety Officer, there is unacceptable risk to employees, the public, the environment, or University property, the Lab Safety Committee and the UNBC Joint Health and Safety Committee shall be notified. Immediate appropriate action shall be taken, which may include the suspension of research or teaching activity, prohibited entry into the laboratory and /or removal of hazardous material from the premises.

CHAPTER 2 - RESPONSIBILITES AND DUTIES

2.1 Employer

An employer must:

- Take every reasonable precaution to ensure the workplace is safe
- Train employees about any potential hazards and in how to safely use, handle, store and dispose of hazardous substances and how to handle emergencies
- Supply personal protective equipment and ensure workers know how to use the equipment safely and properly
- Immediately report all critical injuries to the government department responsible for Occupational Health and Safety Biosafety. At The University of Northern British Columbia is under the direction of the *Laboratory Safety Committee and the Joint Health and Safety Committee.* The committee has representatives from various parts of the University community including students and staff.

2.2 Supervisor or Principal Investigator

- Ensure laboratory staff and students are familiar with and follow the procedures in the University Biosafety Manual. These individuals must also ensure that all staff and students working under their supervision and within their laboratories are aware of these procedures.
- Are responsible for training laboratory personnel in their laboratory on all emergency procedures and how these procedures should be followed in their specific area.
- Must know, comply, inform and enforce safety regulations to their lab personnel that are approved by the UNBC Laboratory Safety Committee.
- Shall develop spill clean-up procedures appropriate for the materials used in the laboratory.
- Must supply appropriate spill kit and contain items useful in containing and cleaning up a typical spill in the laboratory area. This kit should be outside the working area.
- Must ensure that all those using biohazards materials receive biosafety training. New laboratory workers are required to take the basic biosafety training before they begin their work in the laboratory.
- Immediately report all injuries to the appropriate Safety Officer and the Manager of Health and Safety

2.3 Laboratory Users

Laboratory users will:

- Use personal protective equipment and clothing as directed by the employer
- Report workplace biohazards and dangers
- Work in a manner as required by the employer/Supervisor/Principal Investigator and use the prescribed safety equipment

2.4 Biosafety Officer

The Biosafety Officer (BSO) and/or Backup BSO is the individual assigned to manage biological safety issues and is a member of the UNBC Laboratory Safety Committee and the Joint Health and

Safety Committee. The BSO is responsible for the day-to-day operations of the Biosafety Program, acts as a resource to the University community in assisting any UNBC personnel using biohazardous materials to meet regulatory compliance and University safety policies. Developing and coordinating a biological safety program for the facility.

- Assessing risk pertaining to the use of infectious agents and biological material.
- Ensuring facilities compliance with regulatory requirements and best practices, including the CFIA Containment Standards for Veterinary Facilities.
- Inspecting laboratory facilities for suitability of biological work, and documenting all certifications.
- Developing and implementing emergency procedures for biohazardous incidents (spills, equipment failure, animal escape, etc.)
- Being the first point of contact in case of biosafety emergencies (accidental spills, personnel contamination).
- Planning, developing and training personnel on biosafety practices and procedures.
- Informing staff regarding biosafety issues.
- Providing technical advice regarding safe handling, storage and disposal of infectious agents and biological materials.
- Maintains contact and information updates from PHAC, and other regulatory bodies. Prepares and submits reports, applications and other documentation as required by PHAC.
- Maintains required records, documents and permits and submits them to the UNBC Office of Research and other regulatory bodies as required by PHAC.
- Investigates and reports all Biohazardous incidents to the appropriate authorities.
- Liaise with the UNBC Laboratory Safety Committee and with users of Biohazardous material within the University community.
- Maintains current knowledge and training and credentials in preventative measures, regulatory standards and transportation of Biohazardous materials.
- The Biosafety Officer is vested with the authority to stop immediately any use of Biohazardous material which deviates from the approval outlined in the Biosafety license or is deemed to be in non-compliance with the UNBC application standards, or is deemed to be creating an immediate threat to health and safety prior to an investigation by the UNBC Laboratory Safety Committee. The BSO must file an annual report to the UNBC Board of Governors annually.
- The Biosafety Officer reports to the Director, Safety and Security and collaborates with others in the Research Office on biosafety matters, concerns and issues.
- Enforcement of the institutional regulations, under the jurisdiction of the UNBC Safety Committee and the UNBC Joint Health and Safety Committee.
- The Biosafety Officer is the Responsible Official (RO) for the development, training, and implementation of Biosecurity and Emergency Response Plans.
- The RO is involved in the risk assessment process and the biosecurity measures taken such as inventory control, back ground checks and transfers of biological material.
- The RO is contacted as soon as possible in the event of any theft, loss or release of bio-hazardous material.
- Ensures the UNBC Biohazard License is maintained.

2.5 Laboratory Safety Committee

- It is the responsibility of the *UNBC Laboratory Safety Committee* to develop policies and approval procedures governing the use of biohazards on campus, to verify that all work is carried out in accordance with applicable legislation, guidelines and recognized codes and standards of compliance as required by Public Health Agency of Canada, the Canadian Food Inspection Agency and involved funding agencies such as the Tri-council.
- Establish strategies to ensure ongoing and adequate surveillance, hazard identification, and risk evaluation of laboratory activities.
- Advise and assist all members of the university who have a role in promoting and communicating laboratory safety awareness.
- Receive and review reports concerning services, activities, incidents, and interventions involving laboratory activities and/or field work and to recommend corrective strategies where appropriate.

CHAPTER 3 – BIOSECURITY PLAN AND RISK ASSESSMENTS

UNBC has developed and implement the following biosecurity plan and risk assessments to ensure that the potential risks to cause harm to people, the community, livestock, agriculture and the environment are mitigated. Anyone using biohazardous materials must follow the institutional safety procedures and emergency plan for reporting any incidents involving biohazardous material. Any suspicious activity or the presence of unfamiliar individuals in the biohazardous labs must be immediately reported to Campus Security by calling 3333.

3.1 Biosecurity Plan

The goal of the UNBC biosecurity plan is to prevent the loss, theft, misuse, diversion, or intentional release of pathogens and/or toxins. The elements of the UNBC biosecurity plan consist of: physical protection, personnel suitability/reliability, pathogen accountability and incident/emergency response.

3.1.1 Physical Protection

This deals with the actual physical location of the laboratory and all the measures in place to ensure the access to the laboratory is secure, and minimizes opportunities for unauthorized entry. All UNBC biohazardous labs:

- Access is restricted to labs by use of signage
- Room doors are locked when unoccupied
- Have separate key/card access which is only received once training has been completed
- Provided with necessary personal protective equipment and biosafety cabinets
- Intrusion alarms on laboratory doors/entrances, which are monitored by campus security 24/7

3.1.2 Personnel Suitability/Reliability

- All personnel will be provided appropriate training prior to obtaining access to any biohazardous lab
- Copies of the completed training records are available in the appropriate lab
- Names of employees permitted to work in the labs are maintained and controlled by the BSO.
- Campus security are licensed and are certified in Level II Occupational First Aid

3.1.3 Pathogen Accountability

- Inventories of all pathogens and locations are kept in a central database that is maintained by the BSOs and updated tri-annuallyonce a year
- Discrepancies in inventories, breaches of security and the release of agents must be reported to the BSO

- Approved Internal Biosafety Permits must be completed and reviewed by the Laboratory Safety Committee prior to commencing work
- All samples are labeled with content information
- All biological storage locations are marked with biohazardous signage
- All contaminated waste materials are to be decontaminated by autoclaving prior to disposable, by the individual who generated the waste
- If necessary biological waste may be contracted to be disposed via registered contractor

A printed copy of the PSDS used in the space should be maintained and easily accessible in the lab

3.1.4 Biosecurity Incident and Emergency Response Plan

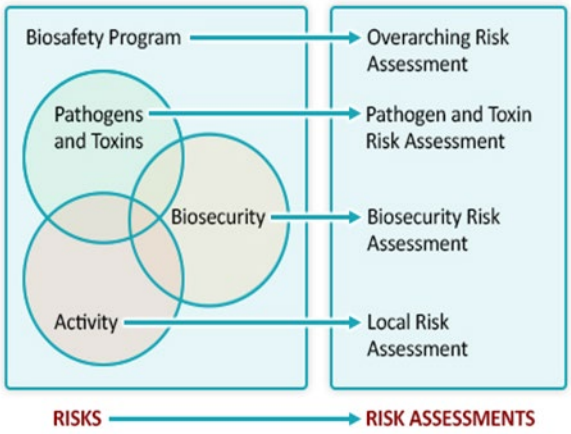
- Loss, theft, accidental release, unauthorized access or suspected tampering is to be immediately reported to the BSO and Principal Investigator. Depending on the concerns the local RCMP may be notified
- Unexplained, recurrent or extended illness of any laboratory personnel using or in the vicinity of biohazardous material should be reported to the Biosafety Officer
- Spill response procedures and equipment are available in each biohazardous lab
- Appropriate biological disinfectants are available in the biohazardous labs

The following elements are incorporated into the incident and emergency response plan:

1. Protects human life over property
2. Personnel are trained and can respond quickly and effectively to an incident
3. Based on collaboration between facility personnel and first responders
4. It addresses the immediate danger and the secondary effects on the people who work in the facility
5. All incidents will be documented and investigated to prevent similar events in the future
6. Reviewed minimally once a year and updated as required.

3.2 Assessments

Four independent but interacting risk assessments have been completed to help identify the potential consequences and the likelihood of exposure to infectious material. The four elements included in UNBCs risk assessment process include, the overarching risk assessment, pathogen and toxic risk assessment, biosecurity risk assessment and local risk assessment.



Assessments will utilize a 5 x 5 risk matrix tool in help to evaluate various risks and workplace hazards.

PROBABILITY/LIKELYHOOD	CONSEQUENCES				
	NEGLIGENT	MINOR	MODERATE	MAJOR	SEVERE
ALMOST CERTAIN >90% chance	Low Risk	Moderate Risk	High Risk	Extreme Risk	Extreme Risk
LIKELY 50-90% chance	Minimum Risk	Low Risk	Moderate Risk	High Risk	Extreme Risk
MODERATE 10-50% chance	Minimum Risk	Low Risk	Moderate Risk	High Risk	High Risk
UNLIKELY 3-10% chance	Minimum Risk	Low Risk	Low Risk	Moderate Risk	High Risk
RARE <3% chance	Minimum Risk	Minimum Risk	Low Risk	Moderate Risk	High Risk

3.2.1 Overarching Risk Assessment (ORA)

The UNBC Biosafety Program (comprised of the LSC and its terms of reference, the Administrative Oversight Plan and the UNBC Biosafety Manual) is under continual review for improvements to maintain or exceed legislative requirements, and will be reviewed minimally once a year, if not more frequently. Program updates and outcomes are communicated to senior level management by the BSO. The LSC, senior management, departments and personnel understand that the potential risks involved in working with biohazardous materials can have either positive or adverse results. As some of these events may contribute to a significant financial loss, property damage, disease and/or liability UNBC is committed to developing, fostering, implementing and monitoring an effective Biosafety Program.

3.2.2 Pathogen and Toxin Risk Assessment (PTRA)

Pathogen and toxin risk assessments determine the likelihood of a pathogen or toxin harming people when used in a controlled laboratory setting. Identifying potential pathogen or toxin risks enables the implementation of the most appropriate safety and administrate controls to mitigate these risks. Due to the characteristics of the pathogens/toxins used at UNBC the risks are generally considered to be of low to moderate risk.

PTRA's will include the following steps:

- Determine the agents risk group, by reviewing the appropriate PSDS or by evaluating the characteristics of the pathogen/toxin in order to determine the risk group (Refer to "SOP BSL-14 Pathogens without a PSDS" to help determine the risk group)
- Determine the appropriate containment levels required for the procedures to be performed
- Assess laboratory safety and establish appropriate control measures

Based on the pathogen/toxin risks a variety of safety controls will be implanted to ensure the risks are kept low. Recommendations may include, one, some or all of the following:

- Ensure that UNBC meets the appropriate physical engineering and operational requirements to work with the pathogen/toxin
- Use of Personal Protective Equipment
- Ensure appropriate training
- Ensure compliance with legislative requirements
- Quantify UNBC liability
- Ensure the prevention of laboratory incidents and infections
- Maintaining valid and appropriate licensing from PHAC or CFIA

3.2.3 Biosecurity Assessment

The biosecurity risks at UNBC are generally considered low to moderate as researchers do not obtain or use pathogens/toxics that are classified higher than a risk group 2. Currently there are no researchers who require, obtain, make or use large volumes (>10 Litres) of any biohazardous materials. All biohazardous materials are stored in locked and restricted access laboratories.

Most risk group level 1 pathogens such as, bacteria, virus, protozoa, fungi, non-infectious plant material, genetic material, protein or non-infectious rodents or animals are considered low biosecurity risk agents. There is a slight increased biosecurity risk with level 2 agents such as bacteria, virus, protozoa, fungi, biological toxins, prions or human source materials.

3.2.4 Local Risk Assessment (LRA)

Prior to the commencement of any research projects or laboratory activities utilizing biohazardous infectious materials requires the approval of the LSC. Completing and submitting an Internal Biohazardous Permit (IBP) application to the LSC will initiate the LRA. IBP applications will be assessed for dual-use, specific pathogen and biosecurity risks by the LSC along with the Primary Investigator, other employees experienced in the procedures and BSO to ensure the health and safety of people, the environment and community. Post approval monitoring will be completed tri-annually, once per each semester, fall, winter and spring, by either the BSO,

alternate BSO or designate. PI and staff working with biohazardous materials need to be able to recognize and notify the LSC and BSO immediately if there has been an unintentional development of the project to be of dual use.

3.3 Dual Use

UNBC undertakes legitimate and well-intentioned research which aims to increase and advance knowledge (i.e. data, models, and information), methodologies (i.e. tools and techniques) and results (i.e. intended or unintended products). Unfortunately, there is a growing concern worldwide regarding the potential negative use of technological advances. The concern is that certain types of research may have the potential to be intentionally used or applied for malicious purposes with detrimental consequences to community health and safety, the environment, or national security. While new knowledge can produce radical or abrupt changes that challenge and transform larger social, economic, environmental and/or governance systems, the potential consideration of the negative consequences of these discoveries must be recognized and addressed.

Bacillus anthracis, the causative agent of anthrax, is an example of the potential consequences of dual use in a laboratory setting. Scientists have been studying the pathogenicity, epidemiology and clinical features of *B. anthracis* for decades in order to develop new vaccines and treatments. However, anthrax also causes terrible diseases in domesticated animals and has a significant impact in the farming economy (Spencer, 2003). In 2001 letters containing *B. anthracis* spores were mailed to two U.S. Senators and several medial offices, killing five people and infecting 17 others. These incidents happened one week after the World Trade Centre and Pentagon were attacked in September. This led to the heightened awareness of the potential theft and misuse of laboratory pathogens to be use in biological warfare (Warrick, 2010).

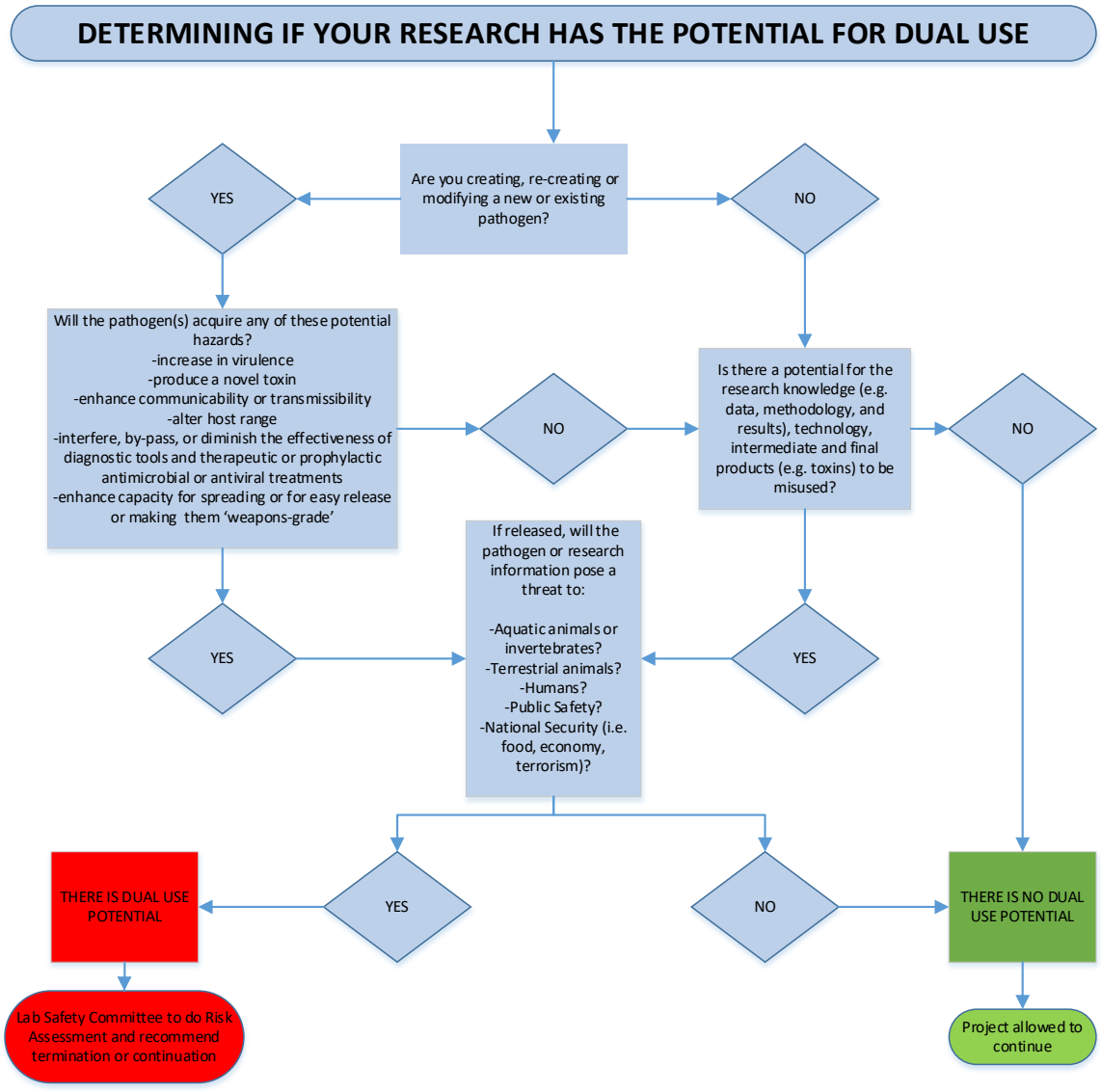
UNBC fosters an environment of collaboration, openness and accessible information; life sciences information is even sometimes public domain. This environment can also present a risk, however unlikely, of theft, misuse and even terrorism. All projects involving biohazards at UNBC will be assessed for dual use potential during the initial permit application, annual renewals, throughout the project until it is completed and the data, analyzed. This is an ongoing review process that is a shared responsibility between those responsible for designing and carrying out the project and those responsible for overseeing the UNBC biosafety program. As UNBC is only equipped to work with risk group level II pathogens, it is unlikely to have high dual risk potential.

3.3.1 Responsibility

The VPR, as the license holder, is ultimately responsible for overseeing the biosafety program. Dual use potential is assessed by the LSC and BSO at time of internal permit submission and renewal. The PI and all personnel involved in the project are responsible for the ongoing assessment of any developments that lead to unanticipated dual use. The BSO is responsible for conducting post approval monitoring, inventory monitoring and user training, where the concept of dual use will be introduced to new users.

3.3.2 During Project Planning

At the time of project planning and completion of the internal permit, the PI will consider the potential for dual use risk in the project. A project cannot commence until the internal permit has been approved by the LSC. To help determine if a project has the potential for dual use at UNBC the following flowsheet will be used:



3.3.3 Ongoing Projects

Ongoing projects are required to be reviewed and re-approved annually by submitting a renewal permit to the Safety Office.

3.3.4 Development of New or Unexpected Dual Use

The PI and research staff involved in the project are responsible for identifying and reporting any dual use that develops that was not previously identified.

3.3.5 Projects Identified as Dual Use

The BSO, LSC and/or VPR can stop a project at any point in time if it is determined that the work has become unsafe, the agent is being misused or mishandled or if there is an unexpected dual use that cannot be mitigated. Identification of dual use in a project will trigger a new risk assessment being completed on the project.

The LSC will notify the VPR of any dual use projects that have been identified or developed. The LSC, VPR and Safety Office will collaborate to determine if the risk is acceptable and if it can be appropriately mitigated. As not to limit or restrict valuable research projects decisions to continue or stop a project, will be determined on a case by case basis.

CHAPTER 4 - BIOSAFETY TRAINING

4.1 Training

All laboratory personnel must attend the UNBC Biosafety training session before working in a laboratory where Level 2 material may be used. A certificate will be issued.

In compliance with the new *Human Pathogen and Toxins Act* which has been in effect since 2009, all persons working with biohazardous materials must have appropriate training and a refresher course every 3 years. Training records will be kept indefinitely.

Custodial and maintenance workers are given a separate course.

General Biosafety training is given by the UNBC Biosafety Officer and follows the necessary requirements given by the Public Health Agency of Canada, *Canadian Biosafety Standard, 2nd Edition.*

All persons working with or around biohazardous material must be instructed in:

- the laboratory specific exposure control plan, including entry control procedures,
- the meanings of various signs, signals or other controls used,
- applicable emergency procedures,
- the recognition and prevention of dangerous situations and exposure routes,
- the symptoms of possible acute/chronic exposures.

As well, these persons must receive documented training in:

- basic biosafety controls and techniques,
- waste disinfection, autoclaving and disposal,
- biosafety cabinets and spills,
- chemical and radioactive biohazards,
- applicable directives which may not be covered in the regular biosafety training or not understood by the individual,
- Specific preventive control methods and requirements of their work and the work area.

All training must be documented and records available to the UNBC Laboratory Safety Committee and the UNBC Joint Health and Safety Committee.

4.2 Special consideration when working with toxins

All laboratory workers handling toxins must be trained in the theory, practice and nature of the hazards of the material to be used.

A risk assessment for safe operation practices must be done before any work is performed, and it is recommended that practice runs with supervision and no toxin be undertaken.

Risk and containment levels must be considered carefully when toxins and infectious agents are used together. If animals are use, there are also other safety issues to be considered.

Routine operations with dilute toxin solutions can be conducted under Biosafety Level 2 conditions with the aid of personal protection equipment and certified biosafety cabinet or fume hood.

4.3 Standard Operating Procedures

The BSO and LSC have developed and implemented various standard operating procedures (SOPs) to help standardize practices and procedures in order to maintain compliance. Users need to review all applicable SOPs for the work that they will be doing. In order to receive access to any biohazardous laboratory on campus, workers and students must not show that they understand the SOP and can demonstrate that they are proficient in all procedures that they are required to complete.

Standard Operating Procedures can be found at:
https://www.unbc.ca/labs/biosafety-standard-operating-procedures

CHAPTER 5 - PATHOGENS

Various microorganisms are responsible for causing materials to become biohazardous or infectious, such as viruses, fungi, parasites, bacteria and their toxic metabolites, human and primate tissues/fluids or cells/cell cultures infected with an agent or prion.

The Canadian Centre for Occupational Health and Safety requires that the employer has the responsibility to implement an occupational health and safety (OHS) program to prevent workplace injury and disease.

The Workplace Hazardous Materials Information System (WHMIS) legislation ensures that workers are provided with adequate health and safety information. It is Canada's national information standard for hazardous materials. WHIMIS covers some risk group level 1 biohazardous infectious material but for any risk group 2 (or higher) biohazardous infectious materials are regulated by the *Public Health Agency of Canada.* Pathogen Safety Data Sheets (PSDS) are similar to WHMIS Safety Data Sheets (SDS) in regards to labeling, storage, disposal and emergency procedures, but they include additional information that is needed to know in regards to the unique nature of pathogens. For example, a PSDS will include how many persons were infected or died, how the pathogen is spread, provides a risk group category, includes the disease signs and symptoms and recommends prophylactic treatments or vaccinations

5.1 Pathogen Safety Data Sheets

- These are produced for personnel working in the life sciences as quick safety reference material relating to infectious micro-organisms and are excellent references. They can be accessed through Health Canada, and contain health hazard information such as infectious dose, viability, decontamination, medical information, laboratory hazards, recommended precautions and handling information.
- The Public Health Agency of Canada (PHAC) has approximately 200 *Pathogen Safety Data Sheets* (PSDS) for infectious substances available.
- You can find those here: https://www.canada.ca/en/public-health/services/laboratory-biosafety-biosecurity/pathogen-safety-data-sheets-risk-assessment.html
- These technical documents provide detailed descriptions of the hazardous properties of specific human pathogens and toxins and recommendations for work practices involving these agents. Updated information on material is posted as it becomes available.

Please note that although the information, opinions and recommendations contained in these Pathogen Safety Data Sheets are compiled from sources believed to be reliable, UNBC accepts no responsibility for the accuracy, sufficiency, or reliability or for any loss or injury resulting from the use of this information. Newly discovered hazards are frequent and this information may not always be completely up to date.

Principle Investigators are responsible for and must insure they have the required PSDA for each controlled product used or stored in the laboratory. They must ensure that it is no more than ***3 years old*** *and be readily available at the workplace as a reference for the workers.*

More information may be found on the PHAC website,under Biosafety Programs and Resources.

The following is an example of a PSDS for *Rabies virus*

Pathogen Safety Data Sheets: Infectious Substances – Rabies virus

PATHOGEN SAFETY DATA SHEET - INFECTIOUS SUBSTANCES

SECTION I - INFECTIOUS AGENT

NAME: Rabies virus.

SYNONYM OR CROSS REFERENCE: Rabies, hydrophobia, lyssavirus[1-9].

CHARACTERISTICS: As a member of the *Lyssavirus* genus, in the family Rhabdoviridae[1,3-9], rabies virus is a bullet-shaped, enveloped virus of approximately 75 nm in diameter by 180 nm in length, and has a single-stranded, negative-sense RNA genome[3]. The *Lyssavirus* genus has 7 members, of which only serotype 1 commonly infects humans, while the other 6 are rare causes of human disease[4].

SECTION II - HAZARD IDENTIFICATION

PATHOGENICITY/TOXICITY: Rabies virus can cause an acute infection, marked by progressive encephalomyelitis, and is usually fatal[10]. The initial symptoms of rabies resemble those of other systemic viral infections, including fever, headache, malaise, and upper respiratory and gastrointestinal tract disorders[1,4,7]. This prodromal phase typically lasts about 4 days, but can last as long as 10 days before specific symptoms develop[1-4]. Almost all cases of clinical rabies are fatal[1,2]. Human rabies is typically seen in 2 forms: furious and paralytic (or dumb)[3].

Furious rabies : Accounts for 80% of rabies cases, is dominated by encephalitis, and presents with hydrophobia, delirium, and agitation(1,3). Hydrophobia is the symptom most identified with rabies; patients have severe difficulty in swallowing and can become fearful at the sight of water despite an intense thirst. Other manifestations of furious rabies include hyperactivity, seizures, and aerophobia(4). Hyperventilation is frequently present, presumably reflecting brain stem infection. Patients then fall into a coma and typically die within 1 to 2 weeks, despite maximal intensive care(3).

Paralytic (dumb) rabies: In contrast to furious rabies, paralytic rabies patients lack signs of cortical irritation, instead presenting with ascending paralysis or symmetrical tetraparalysis[3]. As the condition progresses, the patient becomes confused and death preceded by a coma may ensue[3].

EPIDEMIOLOGY: Rabies occurs throughout the world except in Antarctica, and a few island nations[2,3,5]. The vast majority of cases occur in areas of uncontrolled domestic dog rabies[3]. Rabies is divided into two types for epidemiological purposes: urban and sylvan[1,4].

Urban rabies : Found predominately in developing countries in Asia and Africa[4].

Sylvan rabies : Mostly seen in developed countries in the northern hemisphere[4].

Rabies is estimated to cause 55,000 worldwide human deaths per year, the vast majority of which are in Africa and Asia[6,10]. Several countries, most of which are islands, are rabies-free, including the British Isles, New Zealand, Japan, Taiwan, many of the Caribbean islands, Sweden, Norway, and Spain. These countries remain rabies-free due to the stringency of their quarantine laws for imported animals. Australia was at one time believed to be rabies free, but bat-transmitted rabies is now endemic there[2]. In Canada, a total of 23 people have died of rabies since 1924, and two fatal cases were observed in 2000 and 2003, which were the first cases of rabies in the country since 1985[11].

HOST RANGE: Humans, and many mammals, most commonly wild and domestic canids (e.g. dogs, foxes, coyotes), mustelids (e.g. skunks, badgers, martens), viverids (e.g. mongooses, civets, genets), procyonids (e.g. racoons), and insectivorous and haematophagous bats[1,3,4,8,9].

INFECTIOUS DOSE: Unknown.

MODE OF TRANSMISSION: Rabies is most commonly transmitted to humans via the bite of a rabies-infected animal(2-4,7). Bites to the head, neck, and arms are the most likely to lead to transmission(1). The amount of virus reaching the lesion is also a factor in transmission; for example, when a bite has to penetrate clothing, the saliva may be retained in the fabric and be prevented from entering the wound(2-4). Potential non-bite modes of transmission include contamination of a pre-existing wound, contact of mucous membrane or respiratory tract with the saliva of an infected animal, exposure to aerosolised rabies virus in the laboratory (or from bats), or via organ transplantation from an infected donor(1-4,7).

INCUBATION PERIOD: Varies from days to more than 7 years, with 75% of patients becoming ill within 90 days of exposure[1,3-5].

COMMUNICABILITY: Direct human-to-human transmission is theoretically possible but rare and has only been documented in cases of transplants (corneal, kidney, liver, blood vessel)[1,4-7, 9,10].

SECTION III - DISSEMINATION

RESERVOIR: Urban rabies: stray dogs(1,4). Sylvan rabies; dogs, foxes, coyotes, wolves, jackals, skunks, racoons, mongooses, and other biting mammals such as bats(1,5).

ZOONOSIS: Yes, from the bite of an infected animal[1-9].

VECTORS: None known.

SECTION IV - STABILITY AND VIABILITY

DRUG SUSCEPTIBILITY: Ribavirin (virazole) has shown some efficacy against rabies virus *in vitro*, and interferon-γ was shown to be modestly effective in treating rabies-infected cynomolgus monkeys[12,13].

SUSCEPTIBILITY TO DISINFECTANTS: Rabies virus is inactivated by exposure to 70% ethanol, phenol, formalin, ether, trypsin, β-propiolactone, and some other detergents[3].

PHYSICAL INACTIVATION: Rabies virus does not tolerate pH below 3 or above 11, and is inactivated by ultraviolet light[3].

SURVIVAL OUTSIDE HOST: This virus does not survive well outside its host (in dried blood and secretions) as it is susceptible to sunlight and desiccation[3,9].

SECTION V - FIRST AID / MEDICAL

SURVEILLANCE: Monitoring for symptoms is inadequate since, by the time symptoms are apparent, rabies is invariably fatal. No diagnostic methods are available during the incubation period[3]. Following the incubation period, methods of detection include viral isolation, RT- PCR, and direct immunofluorescence of clinical specimens[1,4-7].

Note: All diagnostic methods are not necessarily available in all countries.

FIRST AID/TREATMENT: First aid for rabies begins with good wound care, which can reduce the risk of rabies by up to 90%. Wash the wound with a soap solution, followed by 70% ethanol or an iodine containing solution[1,3,8]. Following wound care, the clinician must decide whether to institute passive and/or active immunization[3].

There is no established treatment for rabies once symptoms have begun; almost all patients succumb to the disease or its complications within a few weeks of onset(1,3). Supportive therapy includes intubation, sedation, mechanical ventilation, fluid and electrolyte management, nutrition, and management of intercurrent illnesses and complications(3).

IMMUNISATION: Pre-exposure immunization of individuals at high risk for exposure (e.g. laboratory workers, veterinarians, and animal handlers) can be done using Imovax Rabies, a human diploid cell vaccine (HDCV), or RabAvert, a purified chick embryo cell vaccine (PCECV)[2,8]. Currently, both have been approved for use in Canada, and may be used as a pre- and post-exposure prophylaxis[14].

PROPHYLAXIS: Post-exposure rabies prophylaxis with HDCV or PCECV together with the administration of rabies immunoglobulin (RIG) is highly effective[14], although this should not be used in persons who have previously received complete vaccine regimens (pre-exposure vaccination) who require vaccination only[8].

SECTION VI - LABORATORY HAZARDS

LABORATORY-ACQUIRED INFECTIONS: Two cases of laboratory-acquired rabies infections have been reported and are thought to have been acquired via aerosolized virus across mucous membranes[7,15]. No cases of laboratory-acquired infections have been reported in the last several decades. Pre-exposure vaccination is necessary for any individuals working in the laboratory with live virus or diagnostic specimens.

SOURCES/SPECIMENS: Saliva, cerebrospinal fluid, brain tissue, conjunctival or corneal imprints, throat washings, urine, blood, skin biopsies of infected individuals or animals[1,4,6,7].

PRIMARY HAZARDS: Infectious droplets and aerosols containing rabies virus[1,6].

SPECIAL HAZARDS: Fixed tissue preparations can still be infectious so extreme care is needed when handling them[4].

SECTION VII - EXPOSURE CONTROLS / PERSONAL PROTECTI ON

RISK GROUP CLASSIFICATION: Risk Group 3[16].

CONTAINMENT REQUIREMENTS: Containment Level 3 facilities, equipment, and operational practices for work involving infected or potentially infected materials, animals, or cultures.

PROTECTIVE CLOTHING: Personnel entering the laboratory should remove street clothing and jewelry, and change into dedicated laboratory clothing and shoes, or don full coverage protective clothing (i.e., completely covering all street clothing). Additional protection may be worn over laboratory clothing when infectious materials are directly handled, such as solid-front gowns with tight fitting wrists, gloves, and respiratory protection. Eye protection must be used where there is a known or potential risk of exposure to splashes[1,7].

OTHER PRECAUTIONS: All activities with infectious material should be conducted in a biological safety cabinet (BSC) or other appropriate primary containment device in combination with personal protective equipment. Centrifugation of infected materials must be carried out in closed containers placed in sealed safety cups, or in rotors that are loaded or unloaded in a biological safety cabinet. The use of needles, syringes, and other sharp objects should be strictly limited. Open wounds, cuts, scratches, and grazes should be covered with waterproof dressings. Additional precautions should be considered with work involving animals or large scale activities[1,7].

SECTION VIII - HANDLING AND STORAGE

SPILLS: Allow aerosols to settle and, while wearing protective clothing, gently cover the spill with paper towels and apply appropriate disinfectant starting at the perimeter, working inwards towards the centre. Allow sufficient contact time before clean up[1,7].

DISPOSAL: Decontaminate all materials for disposal by steam sterilisation, chemical disinfection, and/or incineration[1,7].

STORAGE: In sealed, leak-proof containers that are appropriately labelled and locked in a Containment Level 3 laboratory[1,7].

SECTION IX - REGULATORY AND OTHER INFORMATION

REGULATORY INFORMATION: The import, transport, and use of pathogens in Canada is regulated under many regulatory bodies, including the Public Health Agency of Canada, Health Canada, Canadian Food Inspection Agency, Environment Canada, and Transport Canada. Users are responsible for ensuring they are compliant with all relevant acts, regulations, guidelines, and standards.

UPDATED: November 2010

PREPARED BY: Pathogen Regulation Directorate, Public Health Agency of Canada.

REFERENCES:

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3. Block, T. P. (2006). Rabies. In R. L. Guerrant, D. H. Walker & P. F. Weller (Eds.), *Tropical Infectious Diseases: Principles, Pathogens, and Practice* (2nd ed., pp. 839-851). Philadelphia, PA: Elsevier Churchill Livingston.

5.2 Recombinant DNA

Recombinant DNA molecules or genetically engineered organisms are defined as;

- Molecules which are constructed outside living cells by joining natural or synthetic DNA segments to DNA molecules that can replicate in living cells or
- DNA molecules that result from the replications of those described above

In recent years "*in vitro*" incorporation of segments of genetic material from one cell into another (recombinant DNA technology) has resulted in altered organisms that can manufacture products such as vaccines, hormones, interferons, and enzymes. However, this biotechnology can also carry a potential for harm and a genetically altered organism may be directly pathogenic or toxic, or if released into the environment, crowd out beneficial organisms, transfer undesirable genetic traits to wild species or mutate into a pathogenic form.

Synthetic DNA segments which are likely to yield a potentially harmful agent are considered as equivalent to their natural DNA counterpart. If the synthetic DNA segment is not expressed *in vivo* as a biologically active polynucleotide or polypeptide, it is exempt from biohazard restrictions.

Because some genetic manipulation does raise some significant possibility of risk, each case needs to have a risk assessment defined in advance, to cover all the possible genetically engineered organisms that might not be known, be created or used in the laboratory.

The following factors should be considered when determining the containment level of a recombinant organism.

- The containment level of the recipient organism
- The containment level of the donor organism
- The replication competency of the recombinant organism
- The property of the donor protein to become incorporated into the recombinant particle
- The potential pathogenic factors associated with the donor protein

If one of the components of the reaction is hazardous, the containment level required should start at the level appropriate to the known hazard, and its containment level increased or decreased accordingly.

Subsequent modifications depend on factors such as;

- Expression of the transferred gene in the recombinant organism
- Ability of the vector to survive outside the laboratory
- Expected interactions between transferred gene, host and other factors

In any research with genes coding for hazardous products, host vector systems with limited ability to survive outside the laboratory should be used; their use will reduce the containment level required.

5.3 Biological toxins

Biological toxins are becoming increasingly accessible, whether from natural or synthetic sources. Some of these toxins can cause death or severe incapacitation even at low exposures.

As toxins do not replicate, are not infectious and usually difficult to transmit manually, they can be handled safely with the proper precautions. Many have low volatility and are relatively unstable in the environment.

The main risk with handling them is accidental exposure through mucus membrane contamination, aerosol generation, needle sticks, or accidents that compromise the skin barrier.

5.4 Cell lines, tissue and cell cultures

The potential laboratory hazards associated with human cells and tissues include blood borne pathogens such as bacteria, fungi, mycoplasma, viruses, parasites and prions as well as agents such as *Mycobacterium tuberculosis* that may be present in lung tissue.

Fixed tissues and tissue sections from human and animal sources that are fixed with chemicals may be handled under Containment Level 1, as generally these chemicals inhibit biological activity.

Although cell lines do not inherently pose a risk to the individuals manipulating them in the laboratory, because of their potential to contain pathogenic organisms, an assessment must be made as to the level of hazard associated with a particular line and for every new cell line. This is required for non-recombinant as well a recombinant cell lines, and the cell line is to be handled at the containment level appropriate to the level of risk determined by the assessment.

Risk groups and containment levels for specific pathogens can be obtained from the *Pathogen Safety Data Sheets* through the Public Health Agency of Canada website.

Cell lines can be grown in an altered manner and with manipulations that may change the behavior of the cell line to a more hazardous state, and a higher level of containment is then required.

Cell cultures may also carry unsuspected oncogenic, allergenic or latent infectious particles so it is prudent to treat all eukaryotic cultures as Risk Group 2 and to use Containment Level 2 facilities and work practices whenever working with them.

Cell lines which are known to contain or be contaminated with a biohazardous agent (e.g., bacteria or virus) must be assigned the same containment level as the agent. The following list contains cells that should be considered Containment Level 2.

- Cell lines from blood, lymphoid, and neural tissue from primates
- All primary cell lines, human or primate
- Secondary (immortalized) cell lines originating from lymphoid or neural tissue
- Cell lines exposed to or transformed by a human or primate oncogenic virus.
- Pathogen deliberately introduced or known endogenous contaminant.
- Fresh or frozen tissue and all cultured cells new to the laboratory until proven to be free of infectious agents.

5.5 Human tissues and fluid

The purpose of this exposure control plan is to eliminate or minimize employee, student and first aid attendants' occupational exposure to blood or other potentially body fluids.

Preventing Exposure

All samples of human blood, blood components or products and other bodily fluids, as well as unfixed tissue, cell or organ cultures of human origin, are to be regarded as potentially hazardous and treated as such due to the possible presence of HIV-1, HIV-11, Hepatitis B and C as well as other viruses and pathogens. These practices apply to all samples, fresh or frozen, from the time they are brought into the laboratory to the time they are denatured by chemical or heat treatment thereby rendering them as non-toxic. Thus, samples treated with protein or DNA denaturing enzymes can be regarded as safe once they are thus treated.

All persons working with human blood and bodily fluids must be vaccinated for Hepatitis B.

CHAPTER 6 - CONTAINMENT AND CLASSIFICATION

6.1 Identification of Biohazardous Material

All users of biohazardous material at UNBC must be accountable for any biohazardous material that they possess. Biohazardous material encompasses, but is not limited to, biological agents, toxins, or products of these materials. The material must include and updated inventory and stored in an area with restricted access to authorized personnel.

The institution must know the following:

- Who is working with and storing biohazardous material
- Where the material is worked with and where it is stored
- When and to whom the materials are transferred
- When and how the materials are destroyed

6.2 Access

Access to secure areas and biohazardous material is the responsibility of the area supervisor or the Principal Investigator.

- There must be controlled access to areas where biohazardous materials are used and stored.
- Laboratories and animal facilities are to be separated from the public and locked at all times.
- Storage rooms, cabinets, freezers and refrigerators containing biohazardous material are to be locked at all times when located in unattended storage areas.
- Access is restricted to the laboratory area and the supervisors must know who is in this area.
- Visitors must be escorted and follow all facility policies and procedures.
- Laboratory keys are not to be shared with unauthorized personnel.
- Lost keys are to be reported to UNBC Facilities and Security.
- Access for routine cleaning, maintenance and repairs in restricted areas should be limited to hours when regular employees are present.

6.3 Risk Groups

Risk groups are recognized internationally by the WHO and usually match up with containment levels. A complete list can be found on the Health Canada website under infectious diseases. Pathogens and their associated Risk groups will no longer be a static part of the hardcopy guidelines. Instead a dynamic list is maintained on the Public Health Agency of Canada website in order to update and accommodate on going risk assessments of risk or current pathogens and addition of new pathogens to the list.

In the existing guidelines biological agents are classified according to risk. The judgment of the inherent risks of pathogens, are based on such factors as:

- The existence (or not) of effective therapies
- The availability of immunization against the specified pathogen
- The presence or absence of vectors
- The quantity of the biological agent being used
- Whether the pathogen is indigenous or exotic
- Is it an emerging pathogen with as yet unknown characteristics?

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

- Pathogenicity
- Infective dose
- Mode of transmission
- Host range
- Availability of effective preventative measures
- Availability of effective treatment

4 RISK CLASSIFICATIONS OF INFECTIOUS MATERIAL			
Risk Group	**Individual**	**Community**	**Examples**
1 **(lowest)** Basic Laboratory, clean open bench, no BSC needed (unlikely to cause disease in healthy workers/animals/plants)	Low	Low	*-non-infectious bacteria* *-E. coli* *-Lactobacillus spp.*
2 Biological safety cabinet needed Pathogens spread via ingestion, inoculation and mucous membrane routes	Moderate	Low	*-Influenza virus* *-Herpes simplex* *-Hepatitis (A, B, C, D, E)* *-Tetanus*
3 Pathogen transmitted by aerosols HEPA filtration required, respiratory protection	High	Low	*-Hepatitis (some C's)* *-West Nile* *-Anthrax* *-TB*
4 **(highest)** serious human disease that may not be treatable, easily transmitted self-contained lab	High	High	*-Ebola virus* *-Herpes B*

**complete list of pathogens/level see Public Safety Data Sheets (PSDS) at www.publichealth.gc.ca*

The table above summarizes how agents are classified by taking into consideration how much risk there is on the individual and the community as a whole. For example, Risk Group 2 has *Moderate individual risk, low community risk.* This includes any pathogen that can cause human or animal disease, but unlikely to be a serious hazard to laboratory workers, the community, livestock or the environment. Lab exposures may cause serious infection, but effective treatment and preventive measures are available and the risk of spread of infection is limited. The use of standard microbiological practices must be used in the laboratory with appropriate personal protective equipment such as gloves, lab coats, and protective eye wear.

Currently no material designated as Risk Group 3 or 4 may be brought into the University for research or teaching purposes. Please contact the Biosafety Officer and/or Laboratory Safety Committee for approval, if you are considering working with any of these agents.

6.4 Containment levels

There are four physical containment levels which are outlined in the *Health Canada Guidelines*. Microbiological agents are typically, but not always, handled in containment facilities at the same level as their risk group. **(Risk Group ≠ Containment Level).** The containment level may be changed if large volumes or aerosols are involved, but the Risk Group is never changed.

6.4.1 Containment Level 1 (CL1):

This refers to the basic laboratory that handles low risk agents. CL1 does not require any special design features beyond those suitable for a well-designed and functional laboratory. Work may be done on open bench tops. Containment is achieved by following basic microbiology laboratory procedures (refer to procedures section in this manual for a review of basic lab skills).

6.4.2 Containment Level 2 (CL2):

CL2 labs will use 'primary containment' procedures, such as using appropriate personal protective equipment (i.e., gloves, lab coats, protective eyewear) and using containment devices such as Biological Safety Cabinets (BSC's)

UNBC does have several level 2 labs on campus. If you will be working with any of the following in your lab must be registered as a level 2 prior to receiving these agents.

If you are using the following you must register as a level 2 lab:

- Microorganisms of low biohazard potential such as those in Risk Group 2, (RG 2), or Biosafety Level 2, (BL 2).
- Recombinant DNA activity requiring BL2 physical containment including animal studies that involve the construction of transgenic animals including human cells.
- Non-recombinant cell and/or tissue culture systems that require this level of containment.
- Oncogenic viral systems classified as low risk.
- Introduction of Risk Group 1 materials into experimental animals.
- Production activities with Risk Group 1 organisms.

Minimal physical requirements include the following:

- Biosafety cabinets – Class II, either Type A1 or A2 cabinet, properly maintained and certified annually or when moved or repaired
- Clean, uncluttered benches and work spaces and floors
- Sink for hand washing
- Eyewash station readily accessible
- Waste handling – autoclave, incineration, or chemical disinfection available nearby
- Centrifuges with sealed rotors or safety caps
- UNBC has specific labs for any biohazard work and is only approved for use in these areas

6.4.3 Containment Level 3 (CL3):

CL3 labs use 'primary and secondary' containment barriers to minimize the release of infectious organisms. These secondary measures include using appropriate respiratory protection, HEPA filtration of exhausted lab air and strictly controlled laboratory access.

There are currently no Level 3 laboratories at UNBC at this time.

6.4.4 Containment Level 4 (CL4):

CL4 labs are isolated units, structurally independent of other areas. Maximum containment of the agent is achieved by complete sealing of the facility perimeter with confirmation by pressure decay testing; isolation of the researcher from the agent by using a positive pressure suit or the use of a Class III Biological Safety Cabinet.

There are currently no level 4 labs at UNBC. In Canada, Winnipeg is the only facility that is classified and set up as a *Containment Level 4.*

6.5 Risk Assessment

Risk Assessment will help identify which Containment levels should be used with an agent and the minimum safety measures and protocols needed to work with an agent safely. Risk Assessments are best done as a committee involving individuals with varying expertise and responsibilities such as, the lab supervisor, facility director, Principle Investigator, microbiologist, and the Biosafety Officer.

Containment Levels and Risk Groups do not necessarily equate with each other.

- The Risk Group does not take into account the manipulation of a particular organism or the quantity of it.
- Containment is achieved through the use of appropriate safety equipment, facility design and laboratory procedures and practice, and is dependent on the both the Risk Group of the materials being used and the manipulations and procedures that are being performed.

The control of biohazardous material at UNBC is maintained by a permit system and must be reviewed and approved by the UNBC Lab Safety Committee prior to work with such material.

The UNBC Lab Safety Committee determines the biosecurity requirements based on the results of the risk assessments of a biohazardous material. The risk assessment includes infectious disease risk, weaponization, consequences of release, and level of threat.

Before any work is started with any Risk Group 2 or higher agent, a risk assessment should be performed. Risk Group 2 agents are no longer able to be ordered or purchased without proper inspections and permits.

CHAPTER 7 - BIOSAFTEY LABORATORY PROCEDURES

7.1 INSPECTIONS AND PERMITS

7.1.1 Requirements for Laboratories using any Risk Group 2 Agents

- The laboratory must be separated from public areas by a door which must be lockable.
- Laboratory access must be limited to authorized personnel.
- Laboratory doors must have appropriate signs for biohazard level, containment level, contact information and entry requirements.
- Laboratory doors must be large enough for passage of anticipated equipment.
- Laboratory doors must be kept closed at all times.
- Office and paperwork areas must be located outside the containment laboratory.
- Working surfaces, doors and frames, and casework should be non-absorptive and any damages surfaces repaired as soon as possible.
- Interior coatings must be resistant to gas and chemicals for disinfection and fumigation purposes.
- Windows must be screened.
- Hooks must be available for laboratory coats/coveralls and street clothes separated from laboratory clothing.
- Hand washing sinks must be located near the laboratory exit, preferably "hands-free" capability.
- An emergency eyewash station and shower must be easily available nearby.
- Biological Safety Cabinets (Class II, either Type A1 or A2), must be located inside the laboratory away from high traffic areas and must be certified annually or when moved or filters changed.
- Waste handling; autoclaving, incinerating, or chemical disinfection must be available nearby.
- The UNBC License must be displayed on the entrance door to any biohazardous lab.

7.1.2 Inspections

All laboratories working with Risk Group 2 must be inspected annually by the Biosafety Officer

7.1.3 Internal permits

Are issued by the Laboratory Safety Committee. An internal permit is required before any work with level 2, or higher, materials is conducted. All forms can be found on the UNBC, Office of Research website at https://www.unbc.ca/labs/biosafety-forms-and-resources

7.2 Level 2 Biosafety laboratory Procedures

Containment Level 2 requirements are as follows as per Canadian Biosafety Standard*, 3rd Edition,* Health Canada, 2015. These apply to laboratories that handle Risk Group 2 agents which are those pathogens that can cause human disease, but under normal circumstances, are unlikely to be a serious hazard to lab workers, the community, livestock, or the environment.

Standard Microbiological Laboratory Practices

- The Laboratory supervisor must enforce the institutional policies that control access to the lab, and doors to the lab must not be left open.
- The institutional Biosafety Manual must be available nearby.
- A procedure manual must be available and followed by all Staff and students.

- Persons must wash their hands after working with potentially biohazardous material and before leaving the lab.
- Open wounds of any kind must be covered with appropriate dressing.
- Eating, drinking, smoking, handling contact lenses, applying cosmetics are not permitted in lab areas.
- Food must be stored outside the laboratory area in designated places.
- Mouth pipetting is prohibited.
- Personal protective clothing and suitable footwear must be worn by all personnel, including visitors. The institution is responsible for laundering after decontamination if necessary.
- In special circumstances lab personnel may need appropriate immunizations for agents handled and require medical surveillance and monitoring and records kept.
- Biological Safety Cabinets must be used for Level 2 procedures.
- Centrifugation is to be performed using screw cap or pressure seal tubes. If there is any possibility of leakage, the centrifuge chamber and rotor must be decontaminated immediately.
- Growth chambers and shakers for bacteria should be covered and mold and microorganism inhibitors used in water baths.
- Plastic containers should be used whenever possible.
- Laboratory clothing must be separated from street clothing and not worn or stored in non- laboratory areas.
- All personnel must receive training on the potential hazards associated with the work involved and the necessary precautions to be taken to avoid exposure to biohazards and to prevent the release of such material in the environment.
- Use of needles, syringes and other sharp objects should be limited and disposed of in appropriate puncture proof containers.
- Appropriate signage indicating the nature of the biohazard and any relevant information must be posted outside the laboratory door, and contact information of the laboratory supervisor listed.
- Laboratories are to be kept clean and tidy with no extraneous materials stored there that is not pertinent to the work and cannot be easily decontaminated.
- Work surfaces are to be cleaned and decontaminated after the work day or after any spill of potentially biohazardous material.
- Effective disinfectants against materials being used and stored must be available and accessible at all times.
- All contaminated materials and equipment must be decontaminated before leaving the laboratory area for servicing or disposal.
- Efficacy monitoring of autoclaves must be done on a regular basis with biological indicators and records of such kept for one year as well as cycle logs.
- Leak proof containers are to be used for transporting biohazardous material within the facilities.
- Spills, accidents and exposure to biohazardous material must be reported immediately to the laboratory supervisor, records kept of such incidents and any resulting investigations.
- Emergency procedures for spill clean ups, BSC failure, and other emergencies must be written and easily accessible and followed.

7.3 Inventory Control Procedures

- Written records of biohazardous materials must be updated each time they are used, received and destroyed.
- Inventories of biohazardous material must be current and available for review by the Biosafety Officer, UNBC Lab Safety Committee or other inspection agency.
- The BSO is responsible for having secure duplicate inventory records of biohazardous material and keeping it indefinitely.
- Prior to shipment, the BSO must be contacted to complete any necessary paperwork for PHAC and CFIA.
- There must be adequate training of those who are working with biohazardous materials.
- Anyone working with biohazardous material must have a laboratory emergency plan.

The inventory shall keep track of the following:

- Name of material
- Catalogue number and lot number if known
- Quantity
- Source, commercial or private
- Date material received
- Date material used
- Name of person using
- Record of transfers within and outside of the institution
- Labeling of samples
- Date material destroyed and decontamination method
- Where material may be stored

7.4 Importing Pathogens into Canada

Human, animal and plant pathogens imported into Canada that are classified as greater than Risk Group 1, require importation permits from either the Public Health Agency of Canada (PHAC), the Canadian Food Inspection Agency (CFIA), or both, if the pathogen affects animals as well as humans.

The Public Health Agency of Canada has regulations to control the importation of human pathogens into Canada and to ensure that adequate facilities exist for proper handling and containing of these pathogens. These regulations allow PHAC to assess, control and manage the risk of inadvertent transmission of communicable disease caused by imported pathogens.

Every person importing a human pathogen in Risk Group 2, 3 or 4 must obtain an importation permit.

Consult the UNBC Biosafety Officer and Public Health Agency of Canada website; www.publichealth.gc.ca for more information.

The *Canadian Food Inspection Agency* is dedicated to safeguarding food, animals and plants which enhances the wellbeing of Canada's peoples, environment and economy. It also shares the responsibility with Health Canada for regulating products from biotechnology.

The *Health of Animal Act* and its regulations provide the legislative authority to control the use of imported animal pathogens as well as pathogens associated with reportable animal diseases. A permit is required for the importation of all animal pathogens into Canada.

The Public Health Agency of Canada (PHAC) and the Canadian Food and Inspection Agency (CFIA)reviews applications for the importation of animal and zoonotic pathogens, assess containment facilities for import purposes, and issues the import permits. The division also maintains a database of animal and zoonotic pathogens and their corresponding containment levels.

For more information on this, consult the UNBC Biosafety Officer and the Canadian Food Inspection Agency website; www.inspection.gc.ca .

7.5 Material Transfer Agreements (MTA's)

Material transfer agreements are legal agreements used for UNBC to assume responsibility for the use of the material that is being transferred to the campus from another research institution or supply house. These legal documents can only be signed by the Vice President of Research on behalf of the university.

For more information, contact the UNBC Office of Research and the Biosafety Officer.

7.6 Transportation of Biohazardous Materials

The Biosafety Officer must be notified for permission to acquire or transport specific biohazards.

Because of the potential of exposing other individuals to the biohazards of infectious material, the transport is strictly regulated for both land and air. Use of regular mail for shipment of material that is known to be infectious is strictly prohibited by Canada Post.

It is the responsibility of the sender/ recipient of the biohazardous material to ensure that all regulations are followed and that permit requirements have been met. For more information, contact the UNBC Biosafety Officer to register for the mandatory training before shipping infectious material either by land or by air.

International agencies and associations such as the World Health Organization, the International Civil Aviation Organization, and the International Air Transport Association have developed standards for the safe international shipment of infectious substances.

7.6.1 Out of the Country or out of Prince George city limits:

The transportation of infectious substances within Canada is administered through Transport Canada and regulated by the Transportation of Dangerous Goods Regulations (TDG) (SOR/85-77). The regulation defines the labeling, packaging and documentation requirements. For more information see the website www.tc.gc.ca/

Transport Canada not only states that the package must meet specific requirements, but that the consignor must also have an Emergency Response Assistance Plan (ERAP) registered with them prior to shipping the package.

Shipping infectious material by air is also regulated by the Dangerous Goods Regulations of the International Air Transport Association, (IATA) For more information see the website http://www.iata.org /

7.6.2 Local within University or Prince George city limits:

The transportation of biohazardous material off the UNBC campus to or from a local laboratory, clinic or other facility must conform to the Transport of Dangerous Goods Act and Regulations. Agents classified as Level 1 or Level 2 may be moved within the local UNBC campus between buildings and within buildings without prior authorization, provided that adequate precautions are taken. This requires sealed containers that ensure adequate protection from escape of the material within, even if the container is dropped on concrete. The exterior of the container must be free of any biohazardous material.

When transporting Level 2 material within or between laboratories, precautions must be taken to control the risks associated with a spill or a leak.

- Limit the number of moves
- Reduce the possibility of breakage and leakage by using strong resistant containers, with screw caps, not snap caps. Use unbreakable, leak proof secondary containers such as zipper locked freezer bags for small tubes.
- Use a cart with guard rails or raised edge for heavier items, and load so that a bump or knock will not dislodge the contents.
- Contain the material in the event of a leak or spill with a tray or pan with raised sides.

When moving infectious substances from one building to another, the following precautions must also be taken.

- Place cushioning absorbent material around the primary breakage resistant, leak proof container.
- Use a secondary leak proof container that will withstand dropping or crushing while in transit
- If the material must be kept refrigerated or frozen during transport, place the coolant such as ice or dry ice, inside an insulated, tertiary vessel. To prevent rupture of the package, ensure that dry ice is able to vent to release carbon dioxide gas.

7.7 Prevention of Biohazardous Spills

For all non-biohazardous and ethidium bromide spills please refer to the *Chemical Laboratory Safety and Methodology Manual.*

Prevention of spills should be the first priority and general precautions should be in place. An emergency spill response protocol specific for the microorganisms in use should be posted in a visible location with the laboratory. Prevention of spills, include but are not limited to the following:

- Use plastics rather than breakable glassware whenever possible.
- Transport materials on carts that have lipped shelves, using secondary containers if possible.
- Do not rush through procedures or when moving goods and materials.

- Be aware of how heavy or awkward some items may be to carry by hand and use carts or extra help.

SUGGESTED BIOHAZARDOUS SPILL KIT

The spill kit should be stored in a visible or marked accessible location within or immediately outside the laboratory. The following items are suggested for a basic spill kit which should be assembled in a single container (e.g. a large bucket). An appropriate chemical decontaminate for the agent being used

- Materials to absorb liquids after decontamination, i.e. paper towels, absorbent lab pads.
- Appropriate personal protective equipment to wear during the clean-up procedure. Gloves, long-sleeved lab coat, and eye/facial protection will be adequate in most cases.
- A mechanical means of handling broken glass, i.e. tongs, forceps, autoclavable dust pans or any other method that prevents direct contact with the broken glass.
- Biohazard bags, sharps containers and/or other containers to place the material in for further treatment or disposal.

7.8 Responsibility for Biohazardous Spill Clean Up

- Responsibility for spill cleanup is shared between several people, depending on the degree of hazard. The person who causes the spill is always responsible for either the cleanup or ensuring that someone with the necessary equipment and expertise has been informed of the spill.
- Before attempting to clean up any spill, ensure that you have appropriate protective protection such as lab coats and gloves before retrieving the spill kit from its storage.
- All spills must be cleaned up as soon as possible and must be reported by the student to the Principal Investigator or the lab supervisor.
- Complete all sections of the report form and deliver to the laboratory supervisor to deliver to the UNBC Manager of Health and Safety.
- Anyone working with biological materials must receive training in spill clean-up appropriate for materials routinely used.
- Always label contents of all containers and waste buckets.

7.8.1 Risk Group 1 or basic Spill Clean-up

- Don lab coats and gloves before proceeding.
- Pick up any broken glass with forceps and dispose of this in a puncture proof container.
- Cover spill with absorbent material such as paper towels.
- Starting from outside edge of the spill and working inwards, saturate the spill with the appropriate disinfectant. Bleach is usually acceptable for most lever 2 pathogens. Refer to the PSDS to ensure you are using an appropriate disinfectant for your biological material.
- Allow to sit for a minimum of 20 minutes to ensure sufficient contact time.
- Pick up used absorbent material and place in plastic garbage bag.

- Wipe spill area again with detergent and hot water, rinse with clean water
- Follow proper regulations for the disposal of biohazard bag and sharps container.
- Remove your gloves.
- Wash your hands well.
- Notify your Supervisor and/or Biosafety Officer

7.8.2 Risk Group 2 Spill Clean Up

This information outlines basic procedures for dealing with small spills, as well as spills of 250mls or larger in an open area.

- Small spills can be cleaned up immediately by lab personnel, provided that the organism does not pose a health risk.
- Evacuate all personnel in the area for 30 minutes. This allows any aerosols to settle.
- Remove any contaminated clothing and gloves and place in biohazard bags to be decontaminated later.
- Close doors to area.
- Post "Do Not Enter" signs on doors.
- Wash hands and any exposed skin
- Don clean protective clothing and gloves.
- Assemble spill kit and prepare disinfectant.
- Pick up any broken glass with forceps and save for decontamination.
- Pour disinfectant around spill, working from the outside in, and gently mix to avoid creating aerosols.
- Allow disinfectant 20 minutes of contact time with spill.
- Clean up liquid with absorbent material provided. Working from the outside in and place in leak proof bag or container.
- Collect any contaminated materials with forceps or squeegee and place in biohazard bag or biohazard container for later decontamination.
- Autoclave any items that have not been in contact with bleach or disinfectant.
- Wipe area again with disinfectant.
- Wipe down any contaminated adjacent equipment, surface areas or furniture with disinfectant.
- Remove personal protective clothing, gloves and goggles and place in biohazard bag for autoclaving.
- Wash hands and any exposed skin again.
- Notify your Supervisor and/or Biosafety Officer

7.8.3 Spill Clean Up of Biohazardous Toxins

In the event of a spill, avoid splashes or generating aerosols during cleanup by covering the spill with paper towels or other disposable absorbent material.

Depending on the toxin, contaminated materials and toxin waste solutions can be inactivated by incineration, extensive autoclaving or by soaking in suitable decontamination solutions.

- Apply an appropriate decontamination solution to the spill (Refer to PSDS).
- Beginning at the perimeter and working towards the center carefully avoiding any splashing, pour decontamination solution slowly.

- Allow sufficient contact time to completely inactivate the toxin (Refer to PSDS).
- Place all disposable cleaning material in secondary containers and autoclave and dispose of as toxic waste.
- General guidelines for laboratory decontamination of toxins can be misleading due to variations in experimental conditions, matrix composition, and criteria for assessing toxin activity.
- Many toxins are inactivated with dilute sodium hydroxide (NaOH) at concentrations of 0.1 – 0.25 N and/or sodium hypochlorite (NaOCl) bleach solutions of 0.1 – 0.5%, freshly prepared.

When working with toxins the following extra pre-cautions should be followed:

- Laboratory work with toxins should be done only in designated, clearly posted rooms with controlled access and designated benches.
- A specific chemical hygiene program should be in place, with no unrelated and nonessential work being performed in the area.
- Inventory must be controlled to account for toxin use, storage, and disposition.
- Toxins must be stored in a restricted area in sealed and labeled containers with contact information readily available.
- Containers holding toxins should be opened in a biosafety cabinet or fume hood.
- Consideration must be given in all experiments to avoid the generation of aerosols.
- Centrifugation of toxins should be performed using thick walled tubes in safety centrifuge cups or sealed rotors and opened only in a Biological Safety Cabinet.
- The outside of primary toxin containers must be decontaminated and placed in a secondary container after work is completed and before removing from the Biological Safety Cabinet.
- Toxin solutions should be transported in leak/spill proof secondary containers
- Select and use correction respiratory protection devices when working with dry toxins
- The Biological Safety cabinets used for toxins should be clearly labeled and use restricted.

For more information on toxin handling and decontamination, refer to the U.S. Department of Health and Human Services Manual, 5th Ed. www.cdc.gov/biosafety/publications/bmbl5/

7.8.4 Spill Clean Up of human blood/fluids

- Isolate the area with a sign or tape.
- Wear gloves, lab coat, goggles and mask.
- Use Spill Kit provided.
- Avoid contact of substances with footwear or clothes.
- Use a dust pan and broom or brush to pick up trash, not your hands.
- Use tongs to pick up any broken glass and place in hard sided container.
- Hold garbage bags away from your body and do not push down on the contents.
- Always tie off garbage bags securely.
- Place garbage bag in red biomedical waste bucket provided.

When working with human blood, tissues or fluids the following extra pre-cautions should be followed:

- Universal precautions must be followed at all times, as human blood, tissue and bodily fluids should be considered infectious. Serological screening of specimens does not guarantee the specimen is free of other infectious agents.

- All samples must be received in sealed containers which do not show evidence of spillage or breakage.
- Appropriate signage indicating the nature of the hazard being used must be posted outside the laboratory door.
- Blood or bodily fluids stored in containers must be opened and handled in such a way as to prevent aerosol formation.
- Centrifuge material with a cap on and load and unload centrifuge rotors in a biosafety cabinet.
- Ensure caps are on containers securely before mixing.
- Whenever possible, all samples are to be opened and worked with in a Biological Safety Cabinet, (CL-2) especially if there is a risk of creating aerosols.
- All surfaces in hoods and lab benches are to be covered in bench paper, and paper removed after usage.
- Laboratory benches and equipment are to be cleaned and disinfected with appropriate disinfectant after each use.
- Do not recap needles, but dispose both the syringe and needle into a puncture resistant sharps container.
- Always use gloves when handling human blood, tissue and bodily fluids.
- Gloves are disposed of when contaminated, removed when work with infectious materials is completed, changed between subjects and are not worn outside the laboratory.
- All liquid wastes generated in the course of the experiments are to be collected in plastic containers marked Biohazard and treated with 10% household bleach, disposing after 24 hours of exposure.
- Samples of solid waste generated in the course of experiments are to be collected in plastic bags marked Biohazard and then to be autoclaved for 30 minutes before disposal.
- Samples must not be frozen in glass containers, but in plastic ones and identified with an appropriate Biohazard warning label.
- Protective lab coats must be worn by all personnel who are working with human blood, tissue, and bodily fluids.
- Lab coats must be removed prior to leaving the area, and soiled coats must not be worn, but placed in an appropriate container for commercial laundering.
- Eye and/or face protection must be worn at all times for anticipated splashes or sprays of infectious materials to the face.
- Wash your hands when finishing the work and before you leave the laboratory.
- If you are exposed overtly to blood, wash the area or wound thoroughly with soap and water and seek medical attention within 1-2 hours. If there is an eye/mucosa splash, flush well with water and seek medical attention as well.
- All accidents, including puncture wounds and spills, which occur during the course of an experiment must be reported to the Principle Investigator and the UNBC Biosafety Officer.

7.8.5 Spill Clean Up of Biohazards in Biological Safety Cabinets

Any spills in a Biosafety Cabinet must be cleaned up immediately with the same procedures as with spills in an open area, but with some extra steps.

- If a spill occurs in a biosafety cabinet, clean up immediately with the cabinet still operating.
- Use a suitable solution of decontaminant to spray or wash cabinet walls and bench. A solution of 10% household bleach or 70% alcohol is usually effective.
- If there is broken glass involved, pick up the glass with forceps and deposit in appropriate container in the cabinet.
- Any items in the cabinet at the time of the spill must be thoroughly cleaned with a disinfectant if they cannot be autoclaved prior to removal from the cabinet.
- Allow the cabinet to run for 10 minutes prior to resuming work.

7.8.6 Biohazardous Decontamination in the Laboratory

Decontamination includes both sterilization and disinfection, and is the reduction of microorganisms to an acceptable level. It is a process or treatment that renders a device, instrument or work surface safe to handle.

All infectious materials must be decontaminated before disposal. Information on the susceptibility of a particular microorganism to disinfectants and physical inactivation procedures can be found in the Pathogen Safety Data Sheet for that agent. Each individual working with biohazardous material is responsible for its proper handling.

Each laboratory is responsibility for ensuring materials, equipment, surfaces, rooms, and samples from containment zones are properly decontaminated.

Every laboratory where biohazardous material is being handled must have effective decontamination procedures. Work areas are to be cleaned and disinfected prior to leaving the laboratory for the day.

General Procedures:

- All infections materials and all contaminated equipment or apparatus must be decontaminated before being washed, stored, or discarded.
- Biohazardous material should be decontaminated at the end of the day or placed in a confined, secure place until this is done.
- Biohazardous materials should not be placed in autoclaves overnight in anticipation of autoclaving the next day.
- All laboratories containing Biohazardous material should designate two separate areas or containers labeled “Biohazardous, to be decontaminated” and “Non- Infectious, to be cleaned”.
- Standard Operating Procedures for Decontaminating Biohazards must be available in the laboratory.
- Autoclaves must be used with biological/chemical indicators on a regular basis to test for efficacy.
- The printed record of an autoclave’s individual run must be saved in a secure place for at least a year for maintenance, servicing, and history.
- All floors, benches, and other surfaces where biohazardous material has been handled should be disinfected as often as required with the appropriate disinfectant.
- Stock solutions of suitable disinfectants should be maintained in the laboratory.

- Autoclave waste containers should be provided for easy disposal of decontaminated waste.

CHAPTER 8 – BIOHAZARDOUS WASTE MANAGEMENT

For disposal of all non-biohazardous and ethidium bromide waste please refer to the *Chemical Laboratory Safety and Methodology Manual.*

A biohazard is a biological agent or substance present or arising from the work environment which presents or may present a hazard to the health or well-being of the worker or community.

UNBC follows the Health Canada *Canadian Biosafety Standard* which require that any biohazardous material must not be disposed of with regular waste and must be decontaminated before disposal or cleaning for reuse. This is to protect individuals and the community from unnecessary exposure to such material. The laboratory using this material is responsible for proper treatment and disposal.

Biohazardous waste encompasses the following material:

- Animal anatomical waste such as body parts, carcasses, organs.
- Non-anatomical waste such as sharps and disposable instruments and equipment which have contacted animal or human blood, biological fluids, or tissues.
- Tissue or microbial cultures and material contaminated by such cultures.
- Live vaccines.
- Containers or materials saturated with blood products.
- Please see the UNBC Biosafety Officer regarding the disposal of any human anatomical waste.

8.1 Biohazardous Waste Handling

- Biohazard waste must not be stored for any length of time, and should be disposed of frequently to reduce accumulation of this material in work areas. Hazardous biological waste must be segregated from non-hazardous waste in appropriate containment and is essential in reducing pollution.
- If waste material must be stored before decontamination, it must be in a locked area at 4^{o}C or lower and treated as soon as possible with no longer than 30 days storage.
- All stored biohazard waste must be in sturdy leak proof and puncture proof containers and identified as to the contents, the name of the user and date of storage. A Biohazard symbol must be displayed on the outer container.

8.2 Biohazardous Waste Containment

Unlike chemicals, infectious material has the ability to replicate, thus there is no “safe” level for release into the environment.

Each lab must have well defined procedures for waste disposal to protect workers, maintenance and cleaning staff from exposure to biohazardous materials. At UNBC, all decontaminated material is collected and stored for regular removal by the Dispensing Chemist.

Principal Investigators and Supervisors are responsible for ensuring that all employees and students are trained and familiar with proper waste disposal procedures and that all lab procedures are in conformance with these requirements.

- There are approved, specified containers in each lab for segregating waste at UNBC.
- The designated sharps container on each lab bench. Sharps include needles and syringes, scalpel and razor blades, and clinical items that may puncture such as Pasteur pipettes.
- Waste containing liquids must be in leak-proof, unbreakable containers prior to disposal.

Biohazard Contaminated Waste NON-TISSUE	Debris Contaminated with Biohazardous or Nuisance Biological Material, but No Tissue	Yellow Biomedical Waste Bucket
Biohazardous Waste TISSUE	Biohazardous and Nuisance Biological Waste Associated Debris	Red Biomedical Waste Bucket

8.3 Decontamination of Biohazardous Waste

Decontamination is considered a procedure in the containment barrier to prevent occupational exposure to infectious agents and/or the accidental release of agents. Approved decontamination methods include autoclaving (heat sterilization), dry heat or gas sterilization, incineration or disinfection with the appropriate chemical. Each laboratory is responsible for their decontamination procedures, which should be part of the laboratory SOP's, and lab workers must be trained in these procedures.

Mixed waste such as infectious waste that is radioactive should first be chemically inactivated with 10% chlorine bleach before being labeled and disposed of as radioactive waste. Do not autoclave radioactive waste.

For decontaminated biohazard waste pick up, please contact the UNBC dispensing Chemist.

8.4 Transport of Biohazardous Waste

Precautions must be taken whenever biohazardous waste is moved from one area to another to control the risks of leakage or spillage and to contain any leaks or spills. Limit the number of moves, and use leak-proof and break resistant containers that are closed securely. Use carts that have guard rails or raised edges for secure transport.

For transport beyond the local laboratory or building, the regulations from Transport Canada are followed. See the Canadian Transport of Dangerous Goods Regulations. Shippers of dangerous goods, including biohazardous material are required to be certified with the proper training. See the UNBC Safety Office for more information regarding this matter.

CHAPTER 9 - EQUIPMENT

9.1 Biological Safety Cabinets (BSC)

Types and Classes of BSC's

There are 3 classes of BSCs Class I, Class II (with many subtypes within) and Class III. Please refer to the Biosafety Officer to ensure you are working with the correct Class of BSC. More detailed information can be found in the Public Health Canada *Canadian Biosafety Standard*.

Class I cabinets have non-recirculated airflow away from the operator that is discharged to the atmosphere after filtration through a HEPA filter. They provide good operator protection but do not protect the material within the cabinet (the product) from contamination.

Class II cabinets are designed for personnel, product and environmental protection. They are designed for work involving microorganisms in containment levels 2, 3 and 4 laboratories and are divided into two types (A and B) on the basis of construction type, airflow velocities and patterns, and exhaust systems (4).

Class III cabinets are totally enclosed and gas-tight with HEPA filtered supply and exhaust air. Work is performed with attached long-sleeved gloves. The cabinet is kept under negative pressure of at least 120 Pa (0.5 in. w.g.), and airflow is maintained by a dedicated exterior exhaust system.

Containment

Biological safety cabinets provide containment for aerosols and separate the work material from the worker and the environment while providing clean air within the enclosed area.

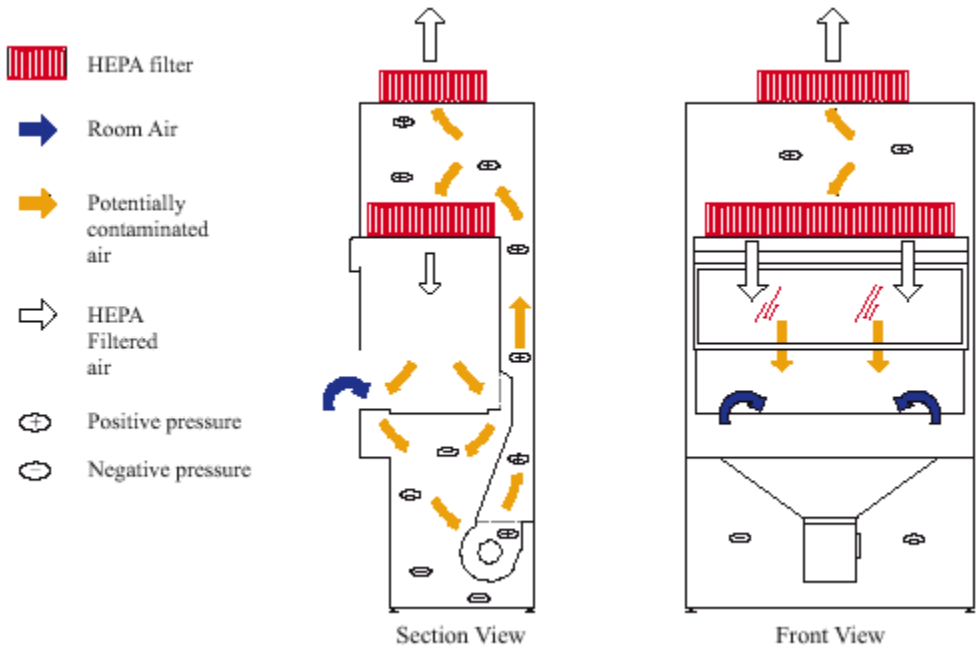
How it works

These cabinets contain fans that direct air into a non-turbulent curtain which is called a laminar air flow.
They provide effective containment and are designed to provide protection for workers, the material and the environment when users observe basic guidelines for their use.

Filtered air is drawn into the cabinet across the work surface and the air is re-filtered before it is vented out. Thus it will maintain a sterile work area by providing a curtain of air between the worker and the material. Most of these cabinets have a UV light for extra protection which is turned off when the cabinet is being used.

Some BSC's are designed to provide only a sterile work surface and do not protect the worker. One must understand what level cabinet is being used and how to use and operate it correctly for maximum protection.

Within type (A), there are two subtypes, A1 (formerly designated type A) and A2 (formerly designated type B3). Within type (B), there are two subtypes, B1 and B2. Class II cabinets are most commonly used in biomedical research laboratories because of their characteristics.



Use of a Biological Cabinet

Class II cabinets are appropriate for both microbiology and tissue culture work. They provide effective containment and a sterile air supply inside.

- Understand how the BSc works and know what level cabinet is being used.
- Minimize the storage of materials inside and around the BSC.
- Plan the work to be done inside
- Assemble all materials that will be needed before starting work to avoid disrupting the air flow
- Do not over load the cabinet
- Avoid disrupting the air flow inside the BSC with items such as a micro centrifuge
- Be sure that all vents and airfoils are clean and unblocked
- Segregate contaminated items from clean items
- Use a horizontal pan with disinfectant inside the BSC for pipette discard
- Do **not** use a Bunsen burner in the cabinet; alternatively use a micro-incinerator for sterilizing inoculation loops.
- The cabinet must be turned on at least 5 minutes before starting work in order to purge the air and remove any particulates
- The operator should wear a closed front lab coat or surgical gown and gloves which should overlap the cuffs.
- All materials needed for the manipulations should be placed in the cabinet before work is initiated to minimize in- and- out motions.
- Do not cover the air intake grill in front or the side vents.
- The operator should work well into the cabinet and at least 4 inches from the front grill.
- When in use, the lab entry door (especially in small rooms) must be kept closed and traffic minimized.
- Do not have electric fans blowing in the room when the BSC is in use.

- Develop appropriate procedures for the collection and decontamination of waste material to avoid clutter.
- The cabinet must be decontaminated with appropriate disinfectant after use.
- Periodic use of 1-10% household bleach in water is acceptable, but it is corrosive, so 70% ethanol may be used more frequently if effective against agent.

Validation

Biological safety cabinets must be inspected and certified annually or when they are moved or repaired. This must be done by a qualified technician and the certification notice displayed on the cabinet.

NOTE: BIOLOGICAL SAFETY CABINETS CANNOT BE USED AS FUME HOODS AS THE HEPA FILTERS WILL BE DESTROYED.

9.2 Laboratory Fume Hoods

Containment

This is the fume hood's ability to contain gases and vapors for effective exhaust. It is usually measured in parts per million. Consistent, uniform airflow across the fume hood opening is a critical performance requirement to deliver safety to laboratory workers.

How It Works

The most common type of fume hood is known as a ducted fume hood, and draws air into the partially contained space through the use of a blower or fan, dilutes the contaminated air with the ambient air and exhausts it to the exterior. The hood usually sits on a storage cabinet and the opening is covered by a transparent sash which is movable vertically across the opening. Raising and lowering the sash changes the air flow into the hood.

Use of a Fume Hood

Fume hoods must be used properly for optimum protection. The fan may be checked with a piece of tissue paper dangled at the hood opening. It is vital to ensure that the type of fume hood being used is appropriate for the hazards being controlled. All supplies and equipment needed in the hood should be placed there at the beginning of the work to avoid moving the sash while chemicals are being used. Do not use the hood as a storage area.

Validation

It must not be assumed that because the fan is running that the fume hood is functioning properly. Fume hoods must be certified annually by a qualified inspector to ensure that there is maximum protection for the worker and this certification notice must be displayed on the cabinet. They must be recertified if they are moved or repaired in any way.

NOTE: FUME HOODS CANNOT BE USED AS BIOLOGICAL CABINETS!

9.3 Centrifuges

Centrifuges pose an additional potential hazard and must be monitored and used appropriately to ensure the containment of biohazardous materials and to prevent aerosol contaminants.

Users must work to minimize the risk of aerosols when centrifuging biohazardous material. Aerosols can be generated when filling centrifuge tubes, removing plugs or caps from tubes

after centrifugation, removing supernatant, re-suspending sedimented pellets and by the very process of centrifugation.

The high speed spins generated by centrifuges can create large amounts of aerosols if a spill, leak or tube breakage occurs. The greatest aerosol hazard occurs when a biohazardous tube breaks during centrifugation. Risk introduced by mechanical failure is avoided by maintaining and inspecting the centrifuge before use.

As ultimately safe centrifuge use is the responsibility of the end users, all users of centrifuges must be adequately trained and familiarized with the manufacturer's instructions prior to handling the instrument.

Safety Considerations

Catastrophic failures of centrifuges are extremely rare, even with high speed ultracentrifuges, but when an event does occur, it generally means the loss of the sample, bottles, potentially the rotor, and in the worst case scenario, the centrifuge. There can also be great danger to nearby equipment and risk to personnel. For this reason, the following steps should be taken to reduce risk of damage. Extra precautions must be taken with the handling of any biohazardous material that may contaminate the rotor and/or centrifuge in the event of a bottle breakage.

- Before use, inspect tubes, O- rings, and buckets for cracks, chips, erosions, bits of broken glass, etc.
- Use unbreakable tubes whenever possible
- The use of aluminum foil to cap biohazardous tubes is forbidden.
- Avoid the use of celluloid (cellulose nitrate) tubes with biohazardous materials. They are flammable and prone to shrinkage with age, distort on boiling and are highly explosive in an autoclave.
- Centrifuge tubes, rotors, and accessories must be prepped and handled in the Biosafety cabinet before and after centrifugation.
- Avoid filling tubes to the rim.
- Once filled, centrifuge tubes must be firmly capped appropriately and wiped with disinfectant before loading in the rotor.
- Always balance buckets, tubes and rotors properly before centrifugation
- Never walk away from a centrifuge until it has reached its maximum set run speed.
- Do not open the centrifuge lid during or immediately after operation, attempt to stop a spinning rotor by hand or with an object or interfere with the interlock safety device.
- To avoid producing aerosols, do not decant supernatants by hand. If you must, do so very carefully. Use a vacuum system with appropriate in-line reservoirs and filters.
- Avoid vigorous shaking of tubes when resuspending packed cells.
- When centrifugation is completed, wipe tubes down with disinfectant before removing cap.
- Clean up any spills promptly.

When using high-speed or ultra- centrifuges, additional practices should include the following:

- Connect the vacuum pump exhaust to a disinfectant trap.
- Record each run in a log book to keep a record of run time and speed for each rotor.

- Install a HEPA filter between the centrifuge and the vacuum pump.
- Never exceed the specified speed limitations of the rotor.

9.4 Autoclaves

Autoclaving is the most reliable method of destroying micro-organisms. The items to be sterilized are heated in a chamber (the autoclave) with saturated steam at a pressure of 15 psi or higher for at least 15 minutes or longer after the temperature reaches 121^{o} C. The material must be in contact with the steam for the required length of time to be effective.

Biohazard material must be labeled as such, contained securely and processed as soon as possible. All users must have successfully completed an authorized training session on the safe operating procedures of the autoclave.

Monitoring

The sterilization process must be monitored to ensure the integrity of the process. This is done with Biological Indicators which are used regularly to ensure that the process has been effective and whether the sterilization has been successful or not.

Biological Indicators are strips that have a set amount of bacterial spores packaged individually in a glassine pouch and are placed in the autoclave with the material to be sterilized. The strips are then incubated and the color indicator in them changes color if the results of autoclaving are unsatisfactory. A positive control is incubated at the same time to ensure that the incubator is working properly.

The monitoring is done weekly, monthly or quarterly depending on how often the autoclave is used. Monitoring is also done after installation of a new autoclave, repair, relocation, malfunction, or suspected failure and a positive control indication.

Chemical indicators, which are usually on autoclave tape or strips are useful for distinguishing between processed and unprocessed items only, and do not guarantee an item is sterile.

Risks

Associated risks with using an autoclave can be heat burns, steam burns, hot fluid scalds, hand and arm injuries from the autoclave door and body injury if there is an explosion. There is also some risk if there is exposure to human pathogens.

Autoclaves are inspected regularly and certified with biological indicators. The UNBC Safety office has copies of the inspection, service and repair records. The name of the person responsible for the autoclave shall be posted near the autoclave. PPE such as heat insulating gloves, lab coats and closed toed footwear must be worn when loading and unloading the autoclave.

Material Preparation

- Ensure that the material is able to be autoclaved.
- Do not autoclave material that contains solvents or substances that may emit toxic fumes.
- Do not autoclave any strong oxidizing agent (bleach) or material with organic material (paper, cloth, etc.) as there is risk of explosion.

- Always consult the Radiation Safety Officer before treating radioactive biological waste for disposal, as the material must be inactivated first before any sort of treatment for disposal.
- Do NOT overload autoclave with too many materials to allow steam circulation.
- Loosely close autoclave bags to allow steam penetration, and leave caps or covers on containers loose.
- Do not mix incompatible materials in the autoclave.
- Glassware must be heat resistant borosilicate and inspected for cracks prior to autoclaving.
- Plastics must be heat-resistant, e.g., polycarbonate (PC), PTFE and most polypropylene (PP) items.
- Sharps must be in a designated "Sharps" container.
- Containers of liquid must be no more than 2/3 full with lids loosened to allow steam to penetrate.
- Place material in low sided autoclavable containers. Place containers of liquid and other materials that may boil over or leak in a secondary pan in the autoclave. The pan must be large enough to handle any leaks or spills.
- Follow autoclave unit procedures carefully to ensure proper and safe sterilization.
- Use chemical indicators (i.e. autoclave tape) with every load to verify autoclaving.
- Add water to containers as appropriate.

Operation

- Every autoclave must have a Standard Operating Procedure in place with the correct operating procedures, safety concerns and a contingency plan.
- Wear appropriate heat-insulating gloves, lab coats and closed toed shoes when loading and unloading the autoclave.
- Allow10 minutes after the run is completed before opening the door carefully and then allow any trapped steam to escape from hot liquids.
- Let liquid loads stand for a full hour before touching with ungloved hands.
- Check autoclave log paper after each run to ensure that the program was successful.
- Record use of the autoclave in log book provided, which should contain operators name, date, time, and duration of run.
- Do not attempt to operate the autoclave if there is a malfunction.
- Report any issues to the autoclave supervisor.

Efficacy Monitoring

Autoclaves must be monitored on a regular basis to ensure that they are decontaminating infective material properly. This is a typical procedure to check the efficacy.

- Biological indicators containing 10^4 to 10^6 cfu/ml of *B. stearothermophilus* spores are placed in the center of a typical load.
- A control indicator is left outside the autoclave
- The load is processed according to standard operating procedures, usually at a temperature of 121^o C for 15 minutes.
- After cycle completion, the biological indicators in the autoclave are retrieved and incubated with the control at 56^o C for a minimum of 24 hours.

- A color change of the autoclaved indicator after incubation means there is growth and sterilization failure. Absence of color change indicates that sterilization of the load was complete. There should be a change in colour of the control that was not autoclaved.
- Failure of sterilization may be due to overloading or improper loading of the autoclave, insufficient sterilization time or malfunction of the autoclave.

9.5 Microwave Ovens

Microwave ovens are commonly used in laboratories for heating and dissolving materials. There are some safety issues with their use in this environment and one should be aware of them.

Safety concerns

- Do not use microwave ovens intended for food items to heat lab materials. Designated ovens for food must not be in the laboratory.
- Superheated liquids can be dangerous. Be aware that all liquids can reach temperatures above their boiling point and can suddenly spill dangerously over the container. Always wear insulating gloves when handling these hot liquids.
- Remember to loosen lids before microwaving as explosions can occur.
- Never place metals or items containing metal components in the microwave oven.
- Allow hot liquids to cool slightly before handling or adding any components to a solution.

CHAPTER 10 - REFERENCES

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