BISHOP’S UNIVERSITY

Biosafety Manual

Adapted from McGill University
Office of Environmental Health and Safety *Biosafety Manual and the St. FX Biosafety Protocol*

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**1 Introduction**

The ***Bishop’s Biosafety Manual*** has been prepared for the benefit of those who handle or work in proximity to potentially infectious biological agents. This manual attempts to address the concerns most frequently encountered personnel by those who work in areas where biohazardous materials are used or stored.

This document applies to all members of the University community including (but not necessarily confined to) faculty members (continuing, contract, visiting and adjunct), students, staff, research associates, postdoctoral fellows, and so on. It also applies to all administrative personnel who are responsible for the oversight of research/teaching or the delivery of research/teaching support involving potential biosafety issues at Bishop's University. This manual applies whether research and teaching activities are funded or not as long as they involve biohazards; whatever the funding source is, and wherever the research/teaching activities are taking place.

This biosafety manual applies to:

* pathogenic organisms and parasites infectious to humans, animals or plants;
* human and non-human primate cell cultures, tissues and body fluids (e.g., blood, urine);
* potentially infectious cell cultures, tissues and body fluids (e.g., sheep amniotic fluids);
* genetically-modified micro-organisms which may be hazardous to humans, animals or plants;
* plasmids, phage or other vectors which may be hazardous to humans, animals, or plants;
* recombinant DNA which may be hazardous to humans, animals or plants;
* biological toxins and venoms.

This Biosafety manual does not apply to:

* transgenic organisms which are not micro-organisms, i.e., plants, animals;
* human bodily fluids and other potentially infectious materials as might be encountered in normal clinical practice in Student Health Services, Occupational Health Services, and the rendering of first aid;
* consumer products for testing which have been obtained from retail outlets;
* procedures associated with the control of zoonotic infections;
* activities associated with licensed abattoirs;
* Activities associated with human anatomy or physiology laboratories (unless blood, body fluids…etc are used as described above)

The Vice-Principal Academic is primarily responsible for ensuring that appropriate standards and biohazard use, as defined by the *2022 Canadian Biosafety Standard 3rd Ed.*, are met, the implemented, and disseminated.

Specifically, Animal Care Committee (ACC) or Biosafety Officer are responsible for the review and approval of research and teaching protocols to ensure that all biosafety issues within Bishop’s jurisdiction are in compliance with institutional, municipal, federal and provincial regulations, and the guidelines of the CBSS.

The ACC-BSO have the mandate to develop, implement and maintain Bishop’s University animal care and use program, in accordance with its Terms of Reference. It may also have the mandate to develop, implement and maintain Bishop’s University biosafety program, in accordance with this Biosafety Manual and the support of the Biosafety Officer.

The University Biosafety Officer shall:

* assist Principal Investigators to assess facilities and prepare the biosafety certificate application;
* co-sign approved biosafety certificates;
* provide advice on biohazardous materials and work procedures;
* provide general biosafety training;
* provide support and sign documentation for import permit applications;
* ensure that steam sterilization cycles are verified using biological indicators and that records of users and cycles are maintained;
* in cases of non-compliance with this protocol or any federal, provincial or municipal legislation,
	+ inform the Principal Investigator, Department Chair and appropriate Dean of the non-compliance, specify actions to be taken to deal with the non-compliance and set deadlines for such actions;
	+ refer continuing issues of non-compliance to the ACC or Vice-Principal Academic.

It is understood that the responsibilities of the Principal Investigator of any project involving biohazardous material include:

* applying to and receiving approval from the ACC-BSO before obtaining and/or commencing work with biohazardous material;
* complying with and enforcing the guidelines and standards set by regulatory and granting agencies, University policies and all certificate and permit terms and conditions;
* ensuring that amendments to the certificate including addition of new personnel, changes in organism or termination of projects are submitted in a timely manner;
* maintaining a current inventory of biohazardous materials including the source;
* providing competent supervision and ensuring that all persons working under his/her control have received appropriate training in working with biohazardous materials;
* inspecting the work area routinely;
* taking appropriate action to remedy unsafe acts and conditions;
* ensuring the safety of any service personnel (e.g., Facilities Management), contractors or visitors and advise them of any potential hazards in the work area;
* ensuring all visitors are supervised;
* ensuring that all containment facilities are functioning and personal protective equipment is available;
* developing and continually reviewing site-specific emergency response plans for the work areas and ensuring that appropriate spill response supplies are available;
* ensuring that the work area is secured against unauthorized access at all times and that all biosecurity measures are followed.

As this manual does not address the chemical and physical hazards commonly encountered in the workplace, it is to be regarded as an addendum to existing Bishop’s University Health and Safety Policy or laboratory procedures.

**1.1 Definition of Biohazard/Biosafety**

A biohazard can be defined as any biological material, organism, or material produced by such an organism that is known or suspected to cause human or animal disease. Biohazardous/infectious material falls under Class D, Division 3 of the Workplace Hazardous Materials Information System (WHMIS), and includes:

* pathogenic organisms and parasites infectious to humans, animals or plants;
* human and non-human primate cell cultures, tissues and body fluids (e.g., blood, urine);
* potentially infectious cell cultures, tissues and body fluids (e.g., sheep amniotic fluids);
* genetically-modified micro-organisms which may be hazardous to humans, animals or plants;
* plasmids, phage or other vectors which may be hazardous to humans, animals, or plants;
* recombinant DNA which may be hazardous to humans, animals or plants;
* biological toxins and venoms.

Exposure to biohazardous agents may occur via puncture wounds or as a result of absorption through the respiratory tract, digestive system, skin and mucous membranes: such exposures may result while handling microorganisms, animals, cell cultures and tissues or diagnostic specimens. Investigators who are uncertain as to whether a material is biohazardous or not should consult the Animal Care Committee (ACC) or the University Health and Safety Coordinator (Biosafety Officer).

Biosafety can be defined as the measures employed when handling biohazardous materials to avoid infecting oneself, others or the environment. These measures include containment principals, technologies and practices that are implemented to prevent unintentional exposure to infectious material.

**2. Laboratory Safety Protocols**

Basic requirements in a laboratory using infectious materials are:

* Ensure that all laboratory personnel, including service and custodial staff and visitors, understand the chemical and biological dangers associated with the lab. Affix biohazard signs to Laboratory Information Cards on doors outside laboratories where biohazardous material is handled or stored. Post the spill response protocol in a visible location in the laboratory.
* Restrict laboratory access and keep doors locked when the laboratory is unattended.
* Keep the facility clean and free of clutter. Ensure that emergency safety devices (e.g., fire extinguishers, eyewashes, etc.) are easily accessible and in working order.
* Ensure that all personnel, students and visitors adhere to University policies for eye and face protection and for protective clothing. Remove lab coats or gowns and gloves before leaving the laboratory; never wear lab clothing in eating facilities.
* Avoid eating, drinking, smoking, storage of food and food utensils, application of cosmetics or lip balm and insertion or removal of contact lenses in the laboratory.
* Restrain long hair. Avoid wearing loose clothing or jewelry, shorts and open-toed shoes or sandals.
* Observe "Universal Precautions" when collecting, processing, storing, shipping or transporting human blood and body fluids; i.e., handle such specimens as if infected with a bloodborne pathogen such as hepatitis B or C or human immunodeficiency virus (HIV).
* Carry out procedures so as to minimize risks of splashes, spills and generation of aerosols.
* Avoid mouth pipetting by mouth.
* Use hypodermic needles only when absolutely necessary. Do not bend, break, shear or recap used needles.
* Wash hands after handling infectious material (even when gloves have been worn) and before leaving the laboratory.
* Decontaminate all contaminated materials before disposal or reuse.
* Decontaminate laboratory surfaces following any spill of biohazardous materials and at the end of each workday.
* Report all spills and accidents/incidents.

**3. Classification of Pathogens**

**3.1 Conventional Pathogens**

Criteria for classification of infectious agents are outlined in the document Canadian Biosafety Standard published by the Public Health Agency of Canada. Essentially, microbiological pathogens are classified according to their impact upon the individuals who manipulate them, upon their colleagues, and upon the surrounding community. Agents that pose little or no risk are assigned to Risk Group 1, while those with the greatest hazardous potential are in Risk Group 4. Risk assessment is based upon several factors, including:

* severity of induced disease
* route(s) of infection
* virulence and infectivity of the microorganism
* antibiotic resistance patterns
* availability of effective medical treatment (e.g., antibiotic therapy) or vaccine
* presence of vectors (e.g., arthropods)
* whether the pathogen is indigenous to Canada
* possible effects on other animals and plants

Before setting up experiments involving new biohazards, consideration should also be given to conditions under which the infectious agent is used. For example, manipulation of large volumes and high concentrations of an infectious microorganism in culture media presents a greater risk than smearing the same pathogen on a slide. Work involving release of microbial aerosols, passage in animals and infection of arthropod vectors also increase the hazard. In these cases, pathogens should be handled as if they were in the next highest risk group: i.e., if the experimental procedure is likely to generate large amounts of aerosolized Risk Group 2 agent, the physical and operational requirements applicable to Risk Group 3 agents should be observed.

**3.2 Genetically Engineered Organisms**

The term "biotechnology" describes a variety of techniques for manipulation of cells; biotechnology has long been used for purposes such as selective breeding of animals and food production (bread, yogurt, beer).

More recently, 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. Genetically engineered organisms are used for treatment of waste and spills, and plants can be made resistant to cold, disease, pests and drought.

However, biotechnology carries with it the potential for harm. 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.

**3.3 Tissue Cultures**

Cell cultures derived from humans or animals known to be infected with a pathogen, as well as cultures known or suspected to contain infectious microorganisms (e.g., herpesvirus or EBV-transformed cultures) should be assigned to the risk group appropriate for the suspected or known pathogen and handled using the relevant containment level and work practices. Risk groups and containment levels for specific pathogens can be obtained from the federal [Laboratory Biosafety Guidelines](http://www.phac-aspc.gc.ca/publicat/lbg-ldmbl-04/index-eng.php).

In addition, cell cultures may carry unsuspected oncogenic, allergenic or infectious particles. It is impractical, if not impossible, to screen such cultures for all potentially harmful microorganisms: even well characterized lines with a history of safe use can become contaminated by adventitious, possibly infectious, microorganisms. For this reason, it is prudent to treat all eukaryotic cultures as moderate risk agents (i.e., Risk Group 2) and to use containment level 2 facilities and work practices whenever working with them.

**4. Containment**

The term "containment" is used in describing measures used to provide a barrier between the infectious organism(s) being handled and the worker (and, ultimately, the community at large). Containment is achieved through the use of appropriate safety equipment, facility design and lab procedures and practices.

**4.1 Containment Levels: Facility Design and Work Practices**

Careful consideration must be given to both facility design and work practices to ensure protection of laboratory personnel, their colleagues and the community as a whole. Four containment levels are outlined in the Health Canada guidelines: of the four containment levels, the highest safety standards (Level 4) are reserved for the most hazardous pathogens (Risk Group 4), and the least stringent (Level 1) for those which have minimal impact on health (Risk Group 1).

**4.1.1 Level 1**

Level 1 containment is used when working with agents (Risk Group 1) that pose no risk to healthy adults:

* The laboratory may be near a public area but doors should be kept closed.
* Work may be carried out on an open bench top.
* Lab surfaces (walls, ceilings, furniture and floors) should be cleanable.
* Open windows should have insect screens.
* Eyewash stations and hand-washing facilities should be available.
* Street clothes and lab coats should not be kept together.
* Disinfection should be carried out as required, using effective concentrations and contact times; solutions should be replaced regularly.

**4.1.2 Level 2**

Level 2 containment is appropriate for work with Risk Group 2 agents. The following precautions, in addition to those for containment Level 1, are recommended:

* The facility should be away from public areas and should have self-closing doors;
* a biohazard sign with relevant risk level and information should be posted at the entrance;
* service and custodial staff should be informed of hazards and procedures: the latter should be expected to clean floors only and only remove black bag garbage;
* laboratory furnishings should be constructed with impervious and readily cleanable work surfaces;
* coat hooks must be provide for laboratory coats near the exit;
* lab coats may be front-closing, but should not be worn outside the lab
* hand washing facilities much be located near the exit;
* autoclave must be available in or near the laboratory;
* use Class II biological safety cabinets for procedures that generate infectious particles and have been tested and certified annually or after repairs according to accepted standards;
* procedures should be carried out such that aerosol generation is minimized;
* an emergency spill response plan should be in place;
* wear gloves to prevent skin contamination.

**4.1.3 Level 3**

Level 3 containment is recommended for work with Risk Group 3 agents. Measures should include the recommendations outlined for levels 1 and 2, plus the following:

* The lab should be away from general work areas, with controlled access.
* There should be a change and shower area within the containment facility perimeter.
* The area should be kept at negative pressure relative to surrounding areas.
* Supply and exhaust air should be HEPA-filtered or provided by dedicated systems.
* A hands-free handwashing sink should be located near the exit.
* Lab windows should be unbreakable and sealed shut.
* Lab personnel should be trained in handling, disposal, and emergency procedures. Written protocols for these procedures should be developed and posted in a visible location.
* Personnel should wear solid-front lab clothing, which should be autoclaved before laundering or disposal.
* A medical surveillance program is recommended.

**4.2 Biological Safety Cabinets**

Biological safety cabinets reduce the risk of airborne infection by reducing the escape of aerosolized infectious agents into the laboratory environment. In addition to protecting workers, some biological safety cabinets protect the work inside the cabinet from airborne contamination (product protection). Biological safety cabinets minimize contact between the operator and pathogens through the use of directional airflow, HEPA filtration of supply and/or exhaust air, and, in some cases, a physical barrier such as a plastic or glass shield.

**4.2.1 Hepa Filters**

HEPA (High Efficiency Particulate Air) filters are an essential component of the biological safety cabinet, and have particle removal efficiencies of 99.97% or better for 0.3 micron diameter particles. This size particle is used as the basis for filter definition because it is considered the most difficult to remove. Thus, a filter that can trap 0.3-micron diameter particles can easily eliminate other sizes.

HEPA filters consist of continuous sheets of glass fiber paper pleated over rigid corrugated separators and mounted in a wooden or metal frame. The filter medium is delicate and should never be touched. As well, the gaskets used to seal the filter frame to the cabinet must not be disturbed; thus the biological safety cabinet should not be moved without subsequently being tested and certified.

While HEPA filters remove particulates from an airstream, they are not effective at collecting chemical gases or vapours. Thus, it is inadvisable to use recirculating Class II cabinets with agents which have significant amounts of hazardous volatile components. Although 100% exhaust Class II cabinets can be used in experiments which involve use of chemicals of moderate toxicity, it should be remembered that these cabinets are not explosion-proof. Use of flammable or explosive products is to be avoided unless the cabinet has been specifically designed for their use.

**4.2.2 Classes of Biological Safety Cabinets**

Horizontal and vertical clean benches are not biological safety cabinets: HEPA- filtered air is directed over the work surface and then discharged directly into the room. Thus, these units provide product protection, but do not protect the operator from exposure to the materials being handled; they must not be used for work with potentially infectious or toxic materials.

There are three basic types of biological safety cabinet, each providing different levels of containment:

**4.2.2.1 Class II**

* open-fronted
* protects operator, product and environment from particulate contamination
* for work with low to moderate risk agents (Risk Groups 2 and 3)

*General principle of operation*: Escape of pathogens into the worker's environment is prevented by an inward flow of room air which enters the front opening without crossing the work area and by HEPA filtration of exhaust air (this provides environmental protection), while downward flow of HEPA-filtered air through the work area removes work zone contaminants and protects the product. The amounts of room air drawn into the intake grille and the amount of air exhausted through the exhaust filter are equal. This balance is critical: positive pressure will allow the outflow of pathogens, while negative pressure will result in inflow of room contaminants.

The different types of Class II cabinets (e.g., Type A, Type B or 100% exhaust) vary in:

* airflow velocities
* amount of cabinet air recirculated (from 0 to 70%)
* amount of cabinet air exhausted (from 30 to 100%)
* destination of exhaust air (back to lab or outside)
* exhaust ducting (building system versus dedicated ducts)

It should be kept in mind that toxic or radiolabelled chemicals must not be handled in cabinets that recirculate air within the cabinet or exhaust into the laboratory.

**4.2.2.2 Class III**

* totally enclosed, gas tight, with glove ports for manipulation of pathogens
* provides the greatest level of operator and product protection
* for work with high risk pathogens (Risk Group 4)

*General principle of operation*: These cabinets form a physical barrier between the operator and microbiological agent. Internal negative pressure confines any leaks to the inside of the cabinet. Supply and exhaust air is HEPA-filtered; a dedicated exhaust fan, separate from that of the facility ventilation system, discharges directly to the outdoors. There is no recirculation of air within the cabinet.

A Class III cabinet system must be designed to allow for safe introduction, handling and removal of all materials throughout the procedure. Equipment such as the incubator, refrigerator, centrifuge, autoclave and chemical dunk tank are connected to the cabinet system.

**4.2.3 Placement of the Biological Safety Cabinet in the Lab**

Since an uninterrupted curtain of inward flowing air at the front is critical to cabinet performance, the biological safety cabinet should be situated in an area where there will be no interference with this air barrier. Interfering room air currents may be caused by:

* pedestrian traffic
* room ventilation such as overhead supply diffusers, fans, fume hoods, heating and air conditioning registers
* drafts from open windows
* operation of doors

The ideal location would be a "dead end" corner of the lab, away from doorways, throughways, windows, room air supply diffusers, fume hoods and heating equipment.

**4.2.4 Working Safely in a Biological Safety Cabinet**

Biological safety cabinets must be combined with good work practices for optimum safety and contamination control. Recommended practices when using a biological safety cabinet include the following:

* Movement of arms into and out of the cabinet can disrupt airflow, adversely affecting cabinet performance. Whenever possible, place all materials needed for a procedure inside the cabinet before starting. Avoid bringing non-essential equipment and supplies into the cabinet.
* Place supplies, equipment and absorbent towels so that air intake or exhaust grilles are not obstructed.
* Keep opening and closing of lab doors and other personnel activity to a minimum.
* Open flames contribute to the heat load, generate convection currents that disrupt airflow patterns and may damage the HEPA filter. Gas can escape from loose connections or damaged tubing and may be ignited by sparks or heat from cabinet motors and switches. The use of an open flame in the presence of flammable alcohol-based disinfectants further increases the risk of fire or explosion. Pre-sterilized loops, needles, etc. or a micro-incinerator (e.g. Bacti-Cinerator) should be used instead of a flame. Installation of new natural gas lines into biological safety cabinets will only be considered under exceptional circumstances and upon providing written justification to Environmental Health & Safety demonstrating that there are no other viable methods available and the use of an open flame cannot be avoided.
* Attach a HEPA filter cartridge between the vacuum trap and the source valve.
* Work at least 4-6 inches inside the cabinet window.
* Carry out work on an absorbent pad to contain small spills. Clean up spills as soon as they occur; remove and disinfect the grille if contaminated.
* Designate separate areas within the cabinet for contaminated and clean materials; place contaminated material at the rear of the work area.

**4.2.5 Cabinet Start Up and Shut Down Procedures**

Before using the cabinet:

* Turn off the UV lamp; turn on the fluorescent lights.
* Disinfect the work surface (refer to Section 6).
* Place essential items inside the cabinet.
* Allow the blower to run for at least five minutes before starting work.

After completion of work:

* Leave blower on for at least five minutes to purge the cabinet.
* Remove and decontaminate equipment and materials, and disinfect cabinet surfaces.
* Turn off the blower and fluorescent lamp, and turn on the UV light.

**4.2.6 Maintenance/Certification of Biological Safety Cabinets**

Biological safety cabinets must be tested and certified annually. Cabinet performance must also be evaluated:

* upon initial installation in the laboratory
* when moved from one building or laboratory to another
* when moved from one area to another within the same room
* whenever maintenance is carried out on internal parts, and whenever filters are changed

**5. Emergency Spill Response — Safe Handling of Biological Spills**

All individuals who work in a lab where pathogens are used must know how to handle these agents safely and what to do in case of a spill. An emergency spill response protocol specific for the microorganisms in use should be prepared and posted in a visible location within the laboratory.

**5.1 Prevention**

An accident prevention plan should be the first priority. General safety precautions include:

* Limit access to rooms where microbiological agents are used.
* Wear appropriate protective clothing.
* Use the appropriate biological safety cabinet.
* Use plastics rather than breakable glassware to reduce likelihood of puncture wounds, cuts and generation of aerosols in the event of an accident.
* Transport materials on carts that have lipped shelves, using secondary containers (i.e. tubs) to catch spills.
* Disinfect waste.

**5.2 The spill Response Plan**

Response procedures should be established before a spill occurs. Assessment of the hazards presented by the pathogen(s) in use should be based upon:

* virulence and infectivity of the agent
* viability - e.g., does the organism become inactive when dried?
* route of entry - e.g., can the organism enter the body via aerosols or splash to the eye?
* quantity and location of possible spill
* immune status of the individuals at risk

The necessary clean-up materials should be available on site. In preparing a spill response kit, ascertain that it contains the appropriate clean-up materials, protective clothing and equipment. The kit should be stored in a visible and accessible location immediately outside the facility and should include:

* disposable protective clothing (e.g., long-sleeved coat or gown, mask, gloves)
* absorbent paper
* autoclavable container and bags
* disinfectant appropriate for the pathogen(s) handled: be sure to replace the disinfectant before it expires
* autoclavable squeegee or forceps and dustpan

**5.3 Spill Response Procedures**

The appropriate spill response depends on the nature of the spilled organism and on the size of the spill. Sections 5.3.1 and 5.3.2 outline suitable approaches to handling minor and major spills.

**5.3.1 Minor Spills**

Small spills can be cleaned up immediately by lab personnel, provided that the organism does not pose a health risk (i.e., if the spill consists of low to moderate risk agents). Cover with a disinfectant-soaked towel (using a spray bottle for distributing the disinfectant generates aerosols and is to be avoided). Autoclave or discard contaminated material in a biomedical waste container.

**5.3.2 Major Spills**

For spills of large volumes of moderate risk agents (greater than 500ml), proceed as follows:

* Treat serious injuries before attempting to contain the spill.
* Evacuate the area immediately if exposure to an aerosolized microorganism presents a potential health hazard; close the facility door(s) and allow aerosols to settle for 30 minutes. Remove contaminated clothing and place it in an autoclave bag or other sealed container; disinfect and wash exposed skin. Report the spill by dialing 2711.
* Don the appropriate protective clothing and cover the spill with absorbent material such as paper towels to reduce splashing. Pour disinfectant around the perimeter of the spill rather than directly onto it to minimize creation of aerosols. Work the disinfectant toward the centre and let it sit for at least 20 minutes.
* If the spilled material has leaked through the grilles of a biological safety cabinet, leave the cabinet running and pour in enough disinfectant (avoid alcohol due to explosion hazard) to dilute the spill tenfold. Drain the catch tray after the time interval appropriate for the disinfectant.
* Wipe down any adjacent walls, cabinets, furniture and equipment that may have been splashed.
* Use forceps/squeegee and dustpan to pick up and transfer the contaminated material into an autoclavable bag or biomedical waste container.
* Decontaminate the waste and cleaning utensils.

**6. Sterilization and Disinfection in the Laboratory**

There is an important distinction between sterilization and disinfection. Whereas sterilization results in destruction of all forms of microbial life, disinfection results in destruction of specific pathogenic microorganisms.

**6.1 Microbial Resistance to Physical and Chemical Agents**

Microorganisms vary in their resistance to destruction by physical or chemical means. A disinfectant that destroys bacteria may be ineffective against viruses or fungi. There are differences in susceptibility between gram-negative and gram-positive bacteria, and sometimes even between strains of the same species. Bacterial spores are more resistant than vegetative forms, and non-enveloped, non-lipid-containing viruses respond differently than do viruses which have a lipid coating.

Information on the susceptibility of a particular microorganism to disinfectants and physical inactivation procedures can be found in the material safety data sheet (MSDS) for that agent. MSDSs provide additional details such as health hazards associated with the microorganism, mode of transmission, containment requirements and spill response procedures. The departmental technician has available, and can provide to individuals, MSDS’s on a number of infectious microorganisms.

**6.2 Physical Sterilants and Disinfectants**

**6.2.1 Heat Sterilization and Decontamination**

Generally, sterilization is best achieved by physical methods such as steam or dry heat, which are less time-consuming and more reliable than chemical germicides. Of these physical procedures, steam autoclaving is the most practical option for the majority of laboratories for both sterilization and decontamination purposes.

|  |  |  |  |
| --- | --- | --- | --- |
| **Method** | **Advantages** | **Disadvantages** | **Uses** |
| Steam Autoclaving:under pressure 121oC/15 psi for 15-90 mins | * minimal time required
* most dependable sterilant for lab use
 | * loading and packing critical to performance
* maintenance and quality control essential
* damages heat-sensitive items
 | * sterile glassware
* media
* instruments
* decontamination of reusable supplies and equipment
* decontamination of infectious waste
 |

**6.2.2 Other Physical Agents of Sterilization and Disinfection**

**6.2.2.1 Ultraviolet Light (Germicidal Lamps)**

The light (approximately 260 nm wavelength) emitted by UV lamps is germicidal, and can reduce the number of pathogenic microorganisms on exposed surfaces and in air. However, UV light has poor penetrating power; accumulations of dust, dirt, grease or clumps of microorganisms may shield microorganisms from the direct exposure required for destruction. ***UV light can cause burns to skin and eyes***, and factors such as lamp age and poor maintenance can reduce performance. For safe and reliable use of germicidal lamps:

* Clean the bulb at least every 2 weeks; turn off power and wipe with an alcohol-moistened cloth.
* Blue light output is not an indication of the lamp's effectiveness; measure radiation output at least twice yearly with a UV meter or replace the bulb when emission declines to 70% of its rated output.
* Post warning signs to discourage personnel from entering areas where there is risk of exposure to UV light.
* Wear UV protective goggles, caps, gowns and gloves in rooms with UV installations.

**6.2.2.2 Miscellaneous Physical Methods**

The procedures listed below are included for the reader's interest:

* Infrared radiation: used for heat treatment of small metal and glass items.
* Microwaves: used for treatment of liquids, nonmetallic objects, and biohazardous waste.
* Gamma irradiation: disrupts DNA and RNA in living organisms, and is used by hospital and laboratory suppliers for materials that do not tolerate heat and pressure (i.e., autoclaving) or chemical treatments.
* Membrane filtration: physically removes particulates (e.g., microorganisms) from heat-sensitive pharmaceutical and biological fluids. The size of the particles removed is determined by the pore size of the filter membrane.

**6.3 Chemical Sterilants and Disinfectants**

Instruments or materials that cannot withstand sterilization in a steam autoclave or dry-air oven can be sterilized with a gas such as ethylene oxide or a broad spectrum liquid chemical germicide. Chemical decontamination of surfaces may also be necessary for very large or fixed items. Since liquid chemical germicides generally require high concentrations and several hours of exposure time for sterilization purposes, they are usually used for disinfection rather than for sterilization purposes. The majority of chemical disinfectants have toxic properties: follow the manufacturer's directions for use and wear the appropriate personal protective equipment (e.g., gloves, eye protection, apron), especially when handling stock solutions.

Choice of a chemical germicide for use on contaminated equipment, supplies, laboratory surfaces or biohazardous waste depends upon a number of factors, including:

* number and nature of microbes to be destroyed (e.g., spores *vs* vegetative cells, bacteria *vs* viruses)
* type and configuration of item to be disinfected (fissures, crevices and enclosures may shield organisms)
* purpose of treatment (e.g., disinfection *vs* sterilization)
* interaction with other active chemicals
* whether the item is covered with soil which might inactivate the disinfectant
* contact time required for disinfection
* toxicity to individuals, culture systems, environment, residual toxicity on items
* pH, temperature, hardness of available dilution water
* cost

Direct contact between germicide and microorganism is essential for disinfection. Microorganisms can be shielded within air bubbles or under dirt, grease, oil, rust or clumps of microorganisms. Agar or proteinaceous nutrients and other cellular material can, either directly (through inactivation of the germicide) or indirectly (via physical shielding of microorganisms) reduce the efficacy of some liquid germicides.

No one chemical germicide is effective for all disinfection or sterilization purposes. A summary of chemical germicides, their use, effective concentrations**,** advantages and disadvantages are outlined below.

|  |
| --- |
| **CHLORINE COMPOUNDS:** |
| **Sodium hypochlorite solution**1 (liquid bleach) |
| Effective concentrations, contact times | 100-10,000 ppm (.01-1%) free chlorine10-60 min (3,000 ppm for broad spectrum)**1 a 1/10 dilution of 5.25% bleach provides 5,250 ppm available chlorine** |
| Advantages | Broad spectrum; inexpensive; widely available; bactericidal at low temperature |
| Disdvantages | Toxic, corrosive to skin and metals; efficacy decreases as pH increases; inactivated by organic matter; deteriorates under light and heat: shelf life of dilutions is less than 1 week. **DO** **NOT USE ON MOLD OR FUNGUS.** |
| Some uses | General disinfectant; waste liquids; surface decontamination; emergency spill clean-up; instrument disinfection |
| **IODINE PREPARATIONS:** |
| **Iodophors**6 |
| Effective concentrations, contact times | 30-1,000 ppm (.003-.1%) free iodine10-30 min |
| Advantages | Broad spectrum; germicidal over a wide pH range; generally nonstaining, less toxic and less irritating than aqueous or alcoholic iodine solutions |
| Disdvantages | Not consistently sporicidal; efficacy reduced by organic matter; some iodophor solutions support growth of *Pseudomonas* 7 |
| Some uses | Germicidal soaps and antiseptics; surface decontamination; work surface wipedown; instrument disinfection |
| **ALCOHOLS** |
| Effective concentrations, contact times | 70-80% ethanol60-95% isopropanol10-30 min |
| Advantages | Low toxicity; rapid action; low residue; non-corrosive |
| Disadvantages | Rapid evaporation limits contact time; flammable, eye irritant; may damage rubber, plastic, shellac; ineffective against bacterial spores |
| Some uses | Skin disinfectant (antiseptic); surface decontamination; benchtop, cabinet wipedown |
| **HYDROGEN PEROXIDE** |
| Effective concentrations, contact times | 3-30% aqueous solution10-60 min 6% for 30 min may kill spores |
| Advantages | Rapid action; no residue; low toxicity; environmentally safe |
| Disadvantages | Limited sporicidal activity; corrosive to some metals; potentially explosive at high concentrations; stock solutions irritating to skin and eyes |
| Some uses | Surface decontamination; instruments and equipment |

**7. Biohazards Associated with Exposure to Animals**

**7.1 Zoonoses**

Individuals whose work involves exposure to or handling of animals and animal tissues, body fluids and cell cultures should be aware of the possibility of acquiring, and take measures to avoid contracting, a zoonosis. Zoonoses are diseases that can be transmitted from animals to humans and may be acquired through:

* animal bites and scratches
* contact with animal tissues and cultures, body fluids and excreta
* exposure to aerosols produced as a result of activities such as cleaning of cages

Over 150 diseases have been classified as zoonoses, some of which are listed in **Table 4** below. A more complete listing can be found in the "Guide to the Care and Use of Experimental Animals", published by the Canadian Council on Animal Care.

**TABLE 4 -**

Examples of laboratory-acquired zoonoses, causative microorganisms, and animals most commonly associated with transmission to humans.

|  |  |  |  |
| --- | --- | --- | --- |
| **Disease** | **Agent** | **Means of spread** | **Host animal** |
| **Bacterial** |
| Anthrax | *Bacillus anthracis* | Contact, inhalation, ingestion | Farm animals |
| Brucellosis | *Brucella* spp. | Contact, ingestion | Swine, dogs, cattle, sheep, goats |
| Q Fever | *Coxiella burnetii* | Contact, inhalation, ingestion | Cattle, sheep, goats |
| Tuberculosis | *Mycobacterium* spp. | Contact, inhalation, ingestion | Primates |
| Salmonellosis | *Salmonella* spp. | Contact, inhalation, ingestion | Farm animals, rodents, reptiles, amphibia |
| Tetanus | *Clostridium tetani* | Bite and soil-contaminated puncture wounds | Horses, other equinae (also carried by other mammals, and present in soil) |
| **Viral** |
| Rabies | *Rabies virus* | Bites, saliva contact | Dogs, bats, other feral animals |
| Monkey B Virus | *Herpesvirus simiae* | Bite wounds, contact | Old World monkeys |
| Lymphocytic choriomeningitis (LCM) | *Lymphocytic choriomeningitis virus* | Contact, inhalation | Mice, guinea pigs, hamsters, monkeys |
| **Fungal, Protozoan** |
| Toxoplasmosis | *Toxoplasma gondii* | Ingestion of oocytes, inhalation | Cats |
| Ringworm | Dermatophytes | Contact | Dogs, cats, guinea pigs, cattle |
| Histoplasmosis | *Histoplasma capsulatum* | Inhalation of fungi | Dogs, other domestic and wild species |

**7.2 Theory and Practical Training Requirements for Animal Users**

The Bishop’s University Animal Care Committee (ACC), in conjunction with the Research Office, provides online theory training in animal use for research and teaching. This training is mandatory for all individuals who intend to work with animals at Bishop’s and its affiliated hospitals.

**8. Safe Handling of Laboratory Equipment**

**8.0.1 Aerosols**

In addition to avoiding obvious sources of infection such as splashes, cuts, accidental inoculation or ingestion, laboratory workers should also be aware that some pathogens, when airborne, may cause infection if inhaled.

Aerosolized microorganisms are generated during most routine laboratory procedures involving manipulation of liquid suspensions. Not all pathogens can infect via the aerosol route, but for those that do, the risk of infection depends on the type and concentration of agent and on the health status of the exposed individual.

The degree of penetration and retention of airborne pathogens in the respiratory tract is determined primarily by size: particles which are 10 mm in diameter or smaller are most efficiently inhaled, deposited and retained in the upper respiratory tract or in lung alveoli. Larger particles (140 mm or greater diameter) are also of concern because they can settle and contaminate work surfaces, equipment and personnel.

**8.1 Centrifuges**

Improperly used or maintained centrifuges can present significant hazards to users. Failed mechanical parts can result in release of flying objects, hazardous chemicals and biohazardous aerosols. The high-speed spins generated by centrifuges can create large amounts of aerosol if a spill, leak or tube breakage occurs.

To avoid contaminating your centrifuge:

* Check glass and plastic centrifuge tubes for stresslines, hairline cracks and chipped rims before use. Use unbreakable tubes whenever possible.
* Avoid filling tubes to the rim.
* Use caps or stoppers on centrifuge tubes. Avoid using lightweight materials such as aluminum foil as caps.
* Use sealed centrifuge buckets (safety cups) or rotors which can be loaded and unloaded in a biological safety cabinet. Decontaminate the outside of the cups or buckets before and after centrifugation. Inspect o-rings regularly and replace if cracked or dry.
* Ensure that the centrifuge is properly balanced.
* Do not open the 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.
* Decant supernatants carefully and avoid vigorous shaking when resuspending packed cells.
* Clean spills promptly.

**8.2 Freezing Apparatus**

Spills inside freezing equipment may place laboratory and maintenance personnel at risk; for safe use of such equipment:

* Periodically check freezers, liquid nitrogen tanks and dry ice chests for broken ampoules, tubes etc.
* To minimize breakage and leaks, place primary containers such as test tubes inside secondary containers prior to storage in freezing units.
* For electrical safety, remember to shut down units before proceeding with decontamination.

**8.3 Vacuum/Aspirating Equipment**

Glass vacuum vessels may rupture and shower laboratory personnel with glass fragments and flask contents. To reduce these risks:

* Use metal flasks and vacuum traps whenever possible.
* Tape glass containers with duct or adhesive tape to contain glass shards in case of rupture or, use a secondary metal container that is at least as tall as the vacuum flask.

To prevent exposure of lab personnel or maintenance employees who may be required to repair the central vacuum system, vacuum line connections that draw biohazardous aerosols or fluids should be fitted with:

* a HEPA filter in the line leading into the vacuum line: cartridge-type in-line filters provide an effective barrier to escape of aerosols into vacuum systems, and are commercially available for this purpose (discard used filters as biomedical waste)
* an overflow flask in case of accidental aspiration of liquids out of the collection vessel. This flask should:
	+ be of sufficient capacity
	+ be placed between the collection flask and the air filter
	+ contain the appropriate disinfectant
	+ contain an antifoam agent whenever air bubbling generates excessive foam

**8.4 Needles and Syringes**

When working with syringes and needles, the following precautions are recommended:

* Perform all operations with infectious material in a biological safety cabinet.
* Do not bend, shear by hand, or recap needles.
* Place used needles and syringes in puncture-resistant containers and decontaminate before disposal.

**8.5 Pipettes**

**8.5.1 Selection of a Mechanical Pipetting Aid**

Improper handling of pipettes can lead to contamination of the user and/or to generation of hazardous aerosols. Mechanical pipetting aids should be used for all pipetting procedures: never pipette by mouth.

Selection of a pipetting device should be based upon:

* intended use
* ease of handling
* delivery accuracy
* user preference
* quality of seal formed with pipettes to be used; liquid should not leak from the pipette tip
* whether the pipetting aid can be sterilized

**8.5.2 Safe Use of Pipettes**

If infectious aerosols are likely to be generated, perform pipetting operations in a biological safety cabinet. Handling pipettes as described below will reduce splashing and aerosolization:

* Plug pipettes with cotton.
* Check pipettes before using; cracked or chipped suction ends may damage the seals of the pipetting aid.
* Keep pipettes upright while in use and between steps of a procedure to prevent contamination of the mechanical aid.
* Gently expel contents close to the surface of a liquid or allow to flow down the side of the container.
* Avoid mixing fluids by alternate suction and blowing, or by bubbling air from the pipette.
* Avoid forceful ejection of the contents; use TD (short for "to deliver", also referred to as "mark-to-mark") rather than TC ("to contain") pipettes, as the last drop of fluid does not have to be expelled with TD pipettes.
* Use easier-to-handle shorter pipettes when working inside a biological safety cabinet.
* Submerge used non-disposable pipettes horizontally in disinfectant solution; dropping them in vertically may force out any liquid remaining in the pipette.

**8.6 Autoclave**

Autoclaves are ideal for decontaminating biohazardous waste and for sterilizing surgical dressings, glassware and microbiological media and liquids. They must be loaded carefully to allow for steam penetration, since steam must contact pathogens in order to destroy them. Longer times are needed for larger loads, large volumes of liquid and denser materials. Proper loading and packing procedures include the following precautions:

* Wrap packages to allow for steam penetration; aluminum foil does not allow steam penetration, and should not be used for wrapping.
* Do not overload the chamber.
* Avoid overpacking of autoclave bags.
* Do not seal bags or close bottles and other containers tightly.
* Do not stack containers.

The changes that are seen on autoclave indicator tapes following an autoclave cycle do not guarantee that the contents of containers are sterile: they indicate only that the tape on the outside of the packages has been exposed to a certain amount of heat or steam. The time required for effective sterilization depends on the size of the load, volumes of liquid and density of materials to be autoclaved. Regular use (at least monthly) of either a heat-resistant biological indicator such as *Bacillus stearothermophilus* should be used to ensure that the cycle in use really achieves sterilization. The indicator is placed in the area least likely to reach sterilizing conditions, such as in the middle of the largest or densest package. A subsequent colour change indicates that the load has been exposed to the required conditions for a sufficient length of time.

Safe work practices when using an autoclave include the following:

* Read the operating manual and post proper work procedures near the autoclave.
* Never autoclave hazardous chemicals.
* Open the door slightly to allow escape of steam before unloading.
* Wear insulated gloves or mitts when unloading.

**8.7 Miscellaneous Equipment**

* **Microscopes**: disinfect the stage, eyepieces, knobs and any other contaminated parts. Select a disinfectant that will be effective on the pathogens and non-corrosive to the microscope.
* **Microtomes**: disinfect knives and anti-roll plates after use.
* **Water baths**:
— Clean regularly; add disinfectant, such as a phenolic detergent, to the water. Avoid using sodium azide to prevent growth of microorganisms (sodium azide forms explosive compounds with some metals).
— Raise the temperature to 90oC or higher for 30 minutes once a week for decontamination purposes.
— To prevent electrical shocks, unplug the unit before filling or emptying and have the continuity-to-ground checked on a regular basis.
* **Tissue grinders**: use in a biological safety cabinet; wrap glass grinders in a wad of absorbent paper and wear gloves. Polytetrafluoroethylene (PTFE, "Teflon") grinders are safer, as they will not break.
* **Microbiological transfer loops**: to eliminate the spattering and aerosolization associated with flaming of loops, char the material before fully inserting the loop into the flame: i.e., before flaming, hold the loop close to (but not into) the flame. Alternatively, use disposable loops or a microincinerator.

**9. Reporting of Accidents/Incidents**

All accidents, dangerous incidents, workplace exposures to infectious material, or suspected occupational diseases should be reported using the incident report form available from area personnel officers, human resources or security. Forms should be completed and submitted to HR or Security within 24 hours of the accident: these accident reports aid in determining the cause of the accident and in developing measures for preventing recurrence. Any near-accidents or incidents which could result in an accident should also be reported, as these reports are useful in evaluating hazards for prevention of future accidents. All information provided is held confidential within the Joint Health and safety committee.

**10. Waste Disposal**

To protect individuals and the community from unnecessary exposure to biohazardous agents, biomedical waste must not be disposed of with regular waste. Disposal of biomedical waste is governed by the Regulation Respecting Biomedical Waste (Quebec), and encompasses the following categories:

* human anatomical waste (body parts or organs)
* animal anatomical waste (carcasses, body parts, organs)
* non-anatomical waste, which includes:
	+ sharps which have contacted animal or human blood, biological fluids or tissues
	+ tissue or microbial cultures, and material contaminated by such cultures
	+ containers or materials saturated with blood products

Biomedical waste should be disposed of frequently to reduce accumulation of these materials in work areas.

* Line boxes with two biohazard plastic bags.
* Affix a biohazard warning sign and user identification to the outside.
* Ensure that liquids are in leakproof unbreakable containers.
* Place sharps in a plastic puncture-proof container prior to disposal in the biomedical waste box.
* Store the box at 4oC or lower in a locked refrigerator.
* Use separate boxes for each category of waste, e.g., human anatomical should not be mixed with animal anatomical or non-anatomical waste.

**11. Transport of Containment Levels 1 and 2 Material**

Whenever biohazardous materials are moved, whether it be within the lab, between labs or buildings or by public carrier, precautions must be taken to control the risks associated with a spill or leak. Arrangements should be made to:

* Limit the number of moves & use secondary containment;
* Reduce the possibility of breakage;
* Contain the material in the event of a leak or spill.

**11.1 Transport Within or Between Labs**

When transporting within or between laboratories:

* Place specimens in leakproof and breakage-resistant receptacles. Close with screwcaps rather than snap caps whenever possible.
* Use unbreakable leakproof secondary containers; for light loads that are to be carried, ensure that the secondary containers have solid handgrips. Small tubes can be sealed inside zipper-lock freezer bags, which are inexpensive, leakproof and will not break if dropped.
* For heavier items, use a cart with guard rails or raised edges. Load so that the contents will not dislodge if the cart should bump into a wall or door.

**11.2 National and International Transportation Regulations**

Within Canada, transport of biohazardous material is regulated by federal and provincial laws and acts. Use of regular mail for shipment of material that is known to be infectious is prohibited by Canada Post.

**11.2.1 Importation of Biohazardous Materials**

The Public Health Agency of Canada (PHAC) Human Pathogens Importation Regulations govern the importation of all material that could potentially contain human pathogens. An Application for Permit to Import Human Pathogens form and Containment Level 2 Checklist must be completed and sent to PHAC for approval before bringing human pathogens into Canada.

Permits to import animal pathogens into Canada are issued through the Canadian Food Inspection Agency (CFIA), Before importing animal pathogens, complete and submit the following forms to the CFIA:

* [Application for Permit to Import Animal Pathogens](http://www.inspection.gc.ca/english/for/pdf/c5083perimpe.pdf), signed by the applicant.

The procedure to follow when importing biohazardous substances is summarized below:

* Obtain the importation permit(s) from Health Canada and/or the CFIA.
* Provide copies of the permit to the sender, who must make sure that it accompanies the shipment into Canada.
* Ensure that the sender packs and labels the infectious materials according to regulations.
* Arrange to have someone available on the delivery day to accept and examine the package.
* Have the necessary supplies and equipment on hand for decontamination and disposal in case of leakage during transport.
* Acknowledge receipt to the sender.

Note that both sender and receiver are required to keep copies of shipping documents for at least 2 years.

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**13. Glossary**

**Aerosol:** A suspension in air of liquid or solid microscopic particles.

**Antiseptic:** Acting against sepsis. An antiseptic agent is one that has been formulated for use on living tissue such as mucous membranes or skin to prevent or inhibit growth or action of organisms. Antiseptics should not be used to decontaminate inanimate objects.

**Aseptic procedure:** A procedure carried out in a manner that prevents contamination of material.

**Autoclave:** An apparatus which employs physical means (moist heat under pressure) to sterilize or decontaminate. Two types of autoclave are:

* *Gravity displacement autoclave*: this type of autoclave operates at 121oC. Steam enters at the top of the loaded inner chamber, displacing the air below through a discharge outlet.
* *Vacuum autoclave*: this type of autoclave can operate at 134oC, allowing for reduced sterilization cycle time. The air is pumped out of the loaded chamber before it is filled with steam.

**Bacterial spore:** See "Spore, bacterial".

**Bactericide:** An agent that kills vegetative bacteria but not mycobacteria or spores.

**Bacteriostatic:** Inhibiting growth of bacterial organisms without necessarily killing them or their spores.

**Bacterium:** A single-celled microorganism, ranging in size from .4 to 2.0 microns, which multiplies by subdivision.

**Biocide:** An agent that can kill all pathogenic and non-pathogenic living organisms, including spores.

**Bloodborne pathogens:** Infectious microorganisms that are carried in the blood of infected humans or animals and that can be transmitted through contact with infected blood, body fluids, tissues or organs. Bloodborne pathogens are implicated in diseases such as malaria, syphilis, brucellosis, tuberculosis, hepatitis B and acquired immunodeficiency syndrome (AIDS). Workplace transmission of a bloodborne pathogen can occur via:

* accidental inoculation with a contaminated "sharp"
* exposure through open cuts, skin abrasions, and mucous membranes of eyes and mouth
* indirect transmission (e.g., touching mouth, eyes, nose or open cuts with contaminated hands)

**Broad spectrum:** A wide range. A broad spectrum disinfectant is effective against a wide range of microorganisms, including bacterial spores, mycobacteria, non-lipid and lipid viruses, fungi and vegetative bacteria.

**Decontamination:** Removal of microorganisms to a lower level, such that there is no danger of infection to unprotected individuals. Sterilization and disinfection are decontamination procedures.

**Disinfectant:** An agent used to kill microorganisms on inanimate objects such as instruments or surfaces.

**Disinfection:** Use of physical or chemical agents to destroy pathogens and potential pathogens on inanimate objects. Disinfection does not necessarily result in sterilization.

* "*High level disinfection*" inactivates fungi, viruses and bacteria. High level chemical disinfectants may be ineffective against bacterial spores if they are present in large numbers. Extended exposure times may be required.
* "*Intermediate level disinfection*" destroys fungi, some viruses (lipid and most non-lipid medium-size and small viruses), mycobacteria and bacteria.
* "*Low level disinfection*" kills vegetative forms of bacteria, some fungi, and some medium-size and lipid-containing viruses. Low level disinfectants do not reliably kill bacterial spores, mycobacteria or small or non-lipid viruses.

**Etiologic agent:** A disease-causing organism or toxin.

**Fungicide:** An agent that destroys fungi.

**Germicide:** An agent which destroys microorganisms, especially pathogenic microorganisms ("germs"). Sterilants, disinfectants and antiseptics are germicides.

**Gravity displacement autoclave:** See Autoclave

**Infectious:** Able to cause disease in a susceptible host.

**Infectious agent:** A biological organism that can establish a process of infection.

**Iodophor:** Literally, an "iodine-carrying" compound. An iodophor is a combination of iodine and a solubilizing surface-active agent, or carrier.

**Microorganism:** A microscopic organism, such as a bacterium, protozoan, yeast, virus or alga.

**Pathogenic organisms:** Organisms capable of causing disease, either directly (by infecting) or indirectly (by producing a toxin that causes illness).

**ppm:** Abbreviation for parts per million, used to describe concentrations in liquids or gases, e.g., 10,000 ppm is approximately equivalent to 10 g/liter or a 1% W/V solution.

**Prions:** Virus-like proteinaceous infectious agents. Prions differ from viruses in that they are not known to contain either DNA or RNA.

**Protozoa:** Nucleated microorganisms, some of which are large enough to be detected with the naked eye. Sizes range from .01 to 200 microns.

**psi:** Abbreviation for pounds per square inch, a unit of pressure equal to the pressure exerted on an area of one square inch. 1 psi = 7.03 x 10-2 kilograms per square centimeter.

**Recombinant DNA techniques:** Procedures which transfer genetic material between organisms or species.

**Sanitization:** Reduction of microbiological load on objects and surfaces to a safe level.

**Sharps:** Sharp objects such as needles, scalpel blades, broken glass, pasteur pipettes or any other object that can penetrate skin.

**Spore, bacterial:** A bacterial spore is a resistant body formed as part of the life cycle of some bacteria. Bacterial spores are able to withstand severe environmental conditions (e.g., heat, drying, chemicals) for many years. When conditions are favourable, spores germinate into vegetative bacterial cells capable of replication.

**Sporicide:** An agent that destroys bacterial and fungal spores.

**Sterilization:** Use of physical or chemical means to bring about the total destruction of all viable microbes, including resistant bacterial spores.

**Tincture of iodine:** A germicidal solution of iodine in aqueous alcohol, used primarily as antiseptics on skin and tissue.

**Universal Precautions:** Precautions taken when handling, storing, transporting or shipping items or specimens containing or contaminated with human blood and body fluids: all such materials are treated as if infectious.

**Vector:** An agent, such as an insect, that can carry a disease-producing organism from one host to another.

**Vegetative form:** In bacteria, a stage of active growth, as opposed to a resting state or spore formation.

**Viable:** Able to grow and multiply.

**Virucide:** An agent that destroys or inactivates viruses.

**Virus:** A microorganism, ranging in size from .01 to .25 microns (10 - 250 nanometers), that can reproduce only within living cells.

**Virulence:** The disease-producing power of a microorganism.

**Zoonosis:** A disease that can be transmitted from animals to humans.