Biosafety Training

Given by: Jay Majithia

Biosafety Officer/Health and Safety Advisor

Health, Safety and Employee Well-Being

416 736 2100 ext. 44745

jmajithi@yorku.ca

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Objectives

Research is now interdisciplinary and it is important to be up to date with current knowledge and new skills

By the end of this training session, you should be able to:

- Understand the various elements that ensure proper Biosafety in labs
- Be able to perform a risk assessment on your experiments
- Understand, establish and practice good microbiological practices to prevent contamination of your work and yourself
- Become a competent, qualified and safe lab personnel

Course Overview

1. Overview of the Biosafety Program
2. Classification of Biological Agents
3. Biosafety & Biosecurity
4. Lab-acquired Infections
5. Risk Assessment
6. Good Microbiological Practices
7. Biological Waste Management
8. Emergency Response
9. Internal Processes and Documentation

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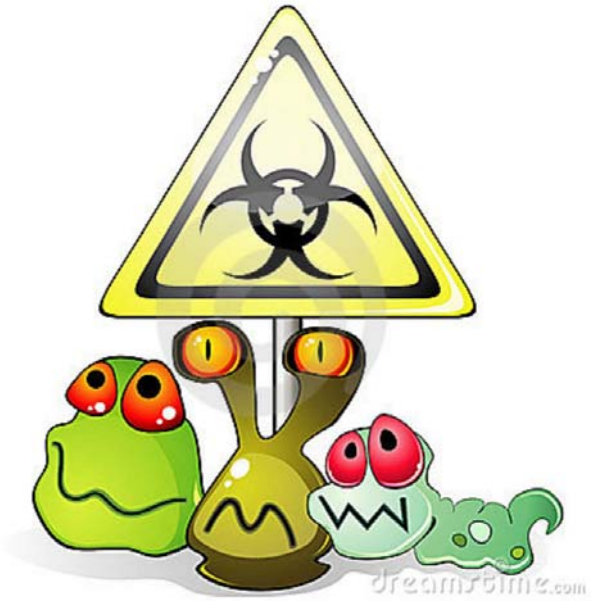
Module 1:

York University

Biosafety Program



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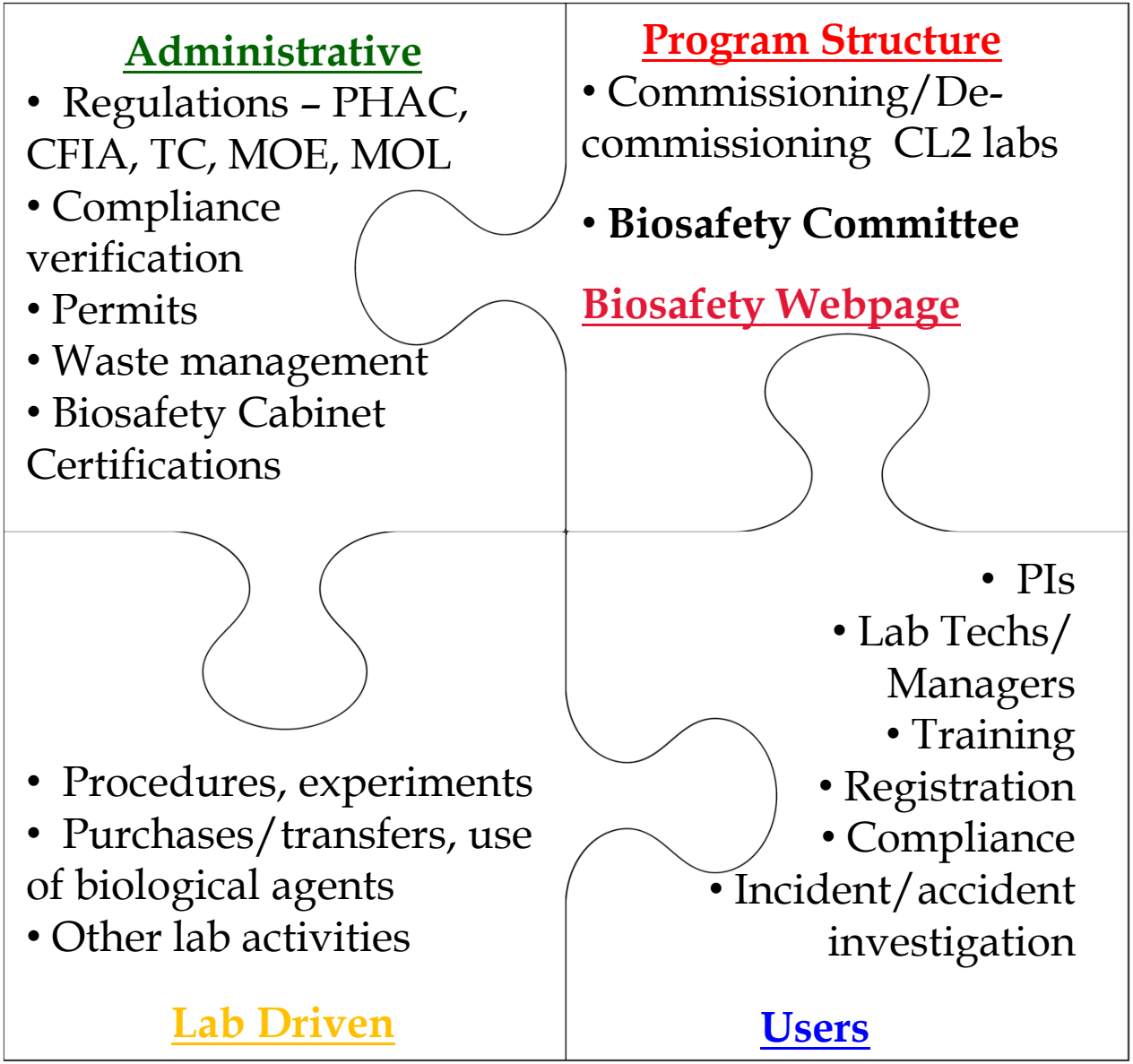
York University Biosafety Program



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Biosafety Regulations

PATHOGENS

Human Pathogens & Toxins

Human Pathogens and Toxins Act & Regulations (2009)

Public Health Agency of Canada / Agence de santé publique du Canada

Animal Pathogens & Toxins

Health of Animals Act & Regulations (1990)

Canadian Food Inspection Agency

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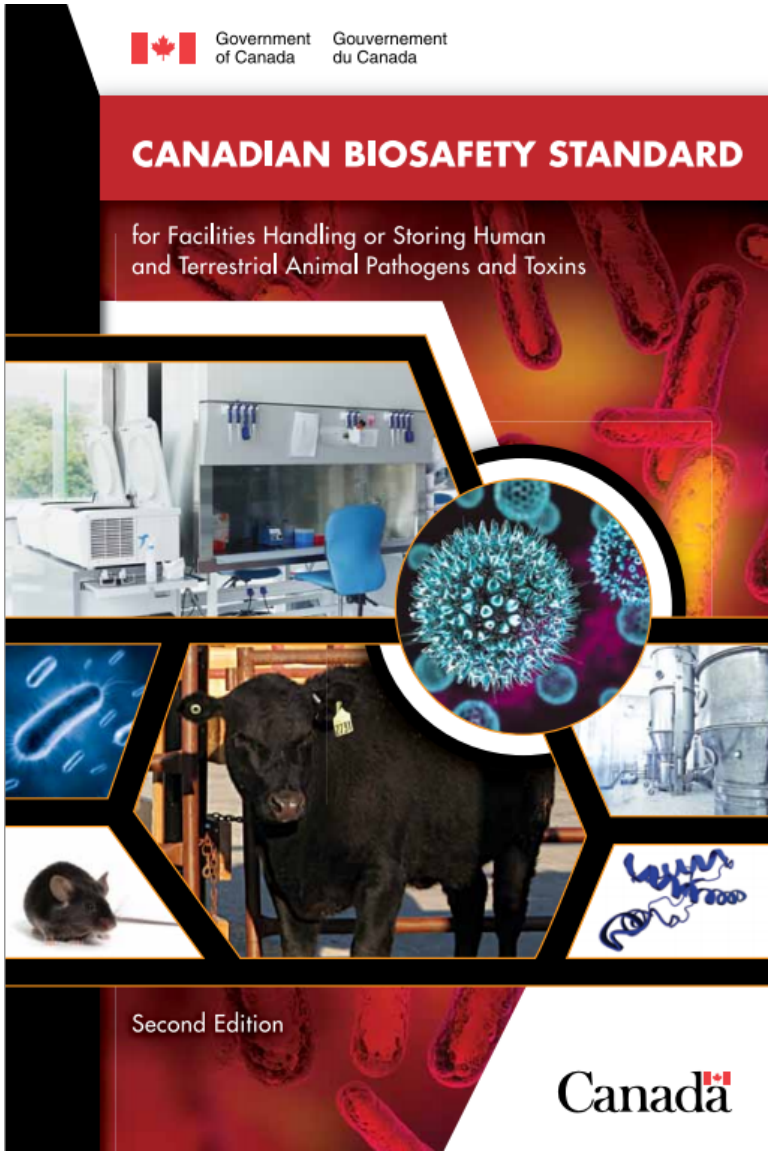
As of December 1st, 2015



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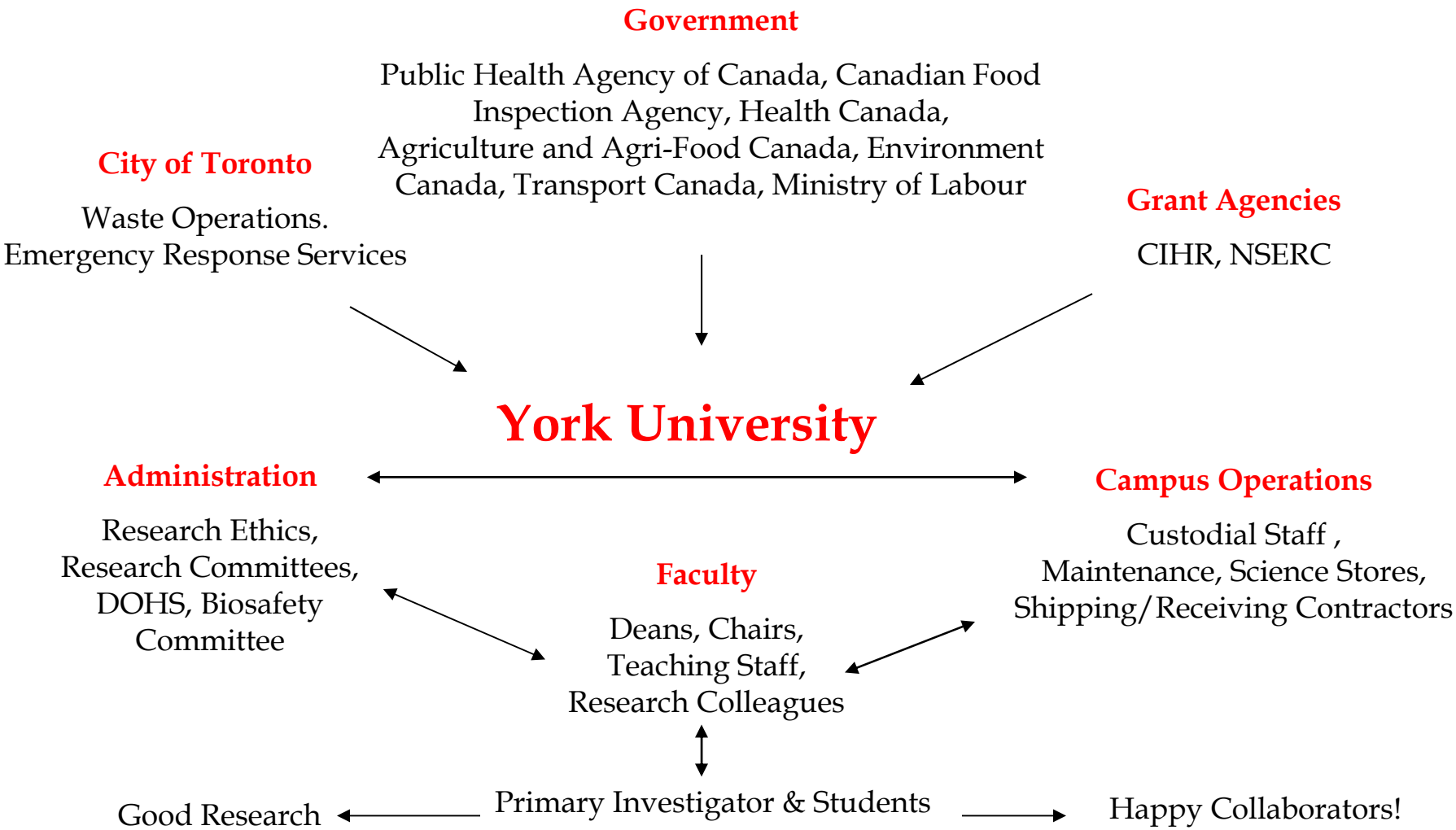
Biosafety @ York University



Biosafety Management at York University

How does York University address government regulation?

- Department of Occupational Health and Safety
- York University Occupational Health and Safety Policy (since 1992)
- York University Biosafety Program (since 2000)
- York University Biosafety Committee

Students: Under the YU Occupational Health and Safety Policy:

"Students are responsible for conducting themselves in a manner which is consistent with their health and safety and that of others. Failure to do so may be considered a breach of the Student Code of Conduct"

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York U Biosafety Program

Program components:

(1) Management

Institutional Biosafety Committee composed of various stakeholders and experts in biological, health sciences and engineering:

- Biosafety Officer
  - Not an expert in all topics
  - Recognizes what evaluations are needed and how to use resources
- Biosafety Chair & Biosafety Committee Members
  - Experts in specific areas
- Office of Research Ethics
  - Ensures researcher is in compliance for funding
- Technical Staff
- Community Members

(2) Hazard Analysis (biological materials)

Risk Assessment and Permit Application Form (Researchers)

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York U Biosafety Program

(3) Support Documents

- Biosafety Program Manual
- Biosafety SOPs (e.g. Safe use of autoclaves, waste disposal)
- Emergency preparedness (e.g. emergency procedures, spill kits…)

(4) Documented Training

- Biosafety Awareness training (DOHS)
- Lab-specific training (Lab Supervisor), Orientation Checklist

(5) Compliance and monitoring

- Lab commissioning
- Lab inspections
- Annual review of biosafety certificates

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Other Related Safety Programs at York

- Department of Occupational Health and Safety
  - Medical Surveillance Program
  - Lab Safety Program
  - Radiation Safety Program

- Office of Research Ethics
  - Involving human subjects
  - Involving animals

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Biosafety Information Sources

- Biosafety Officer/Biosafety Committee
  - Ext. 44745
  - jmajithi@yorku.ca

- Biosafety webpage

  http://www.yorku.ca/dohs/prog-biosafety.html

- Biosafety Program Manual

- Policies, procedures, guidelines

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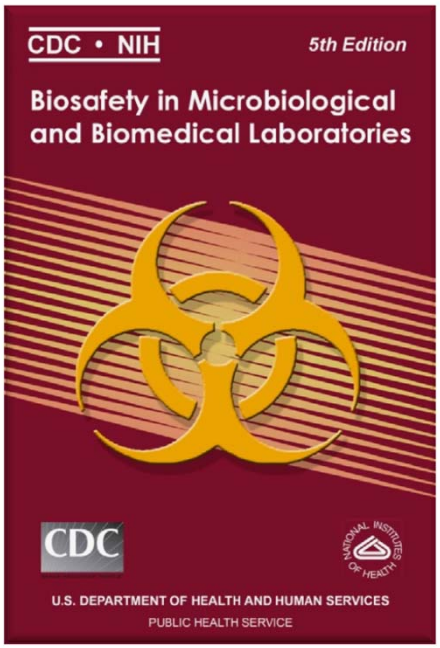
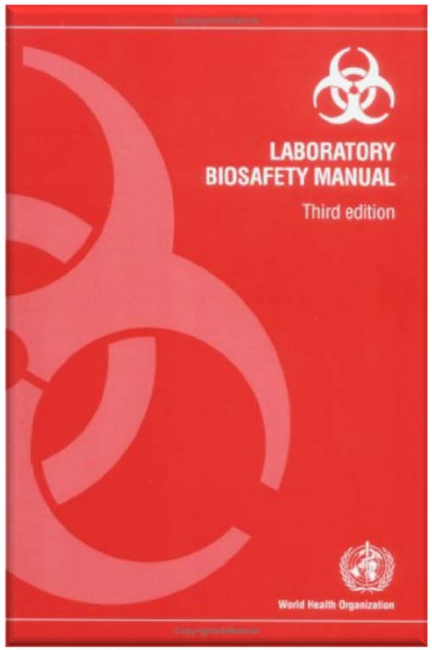
Other Information Resources

- World Health Organization Laboratory Biosafety Manual (3rd Edition, 2004)
  - Provide Biosafety Guidelines at an International Level.

- National Institutes of Health (US) Biosafety in Microbiological and Biomedical Laboratories (5th Edition, 2007)





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Module 2:

Classification

of

Biological Agents

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Terminology

- Biohazard / Biohazardous
- Biosafety
- Biosecurity

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What is a Biological hazard?

A biological hazard (Biohazard) is a potentially infectious agent or hazardous biological material that presents a risk or a potential risk to the health of humans, animals, plants and/or the environment

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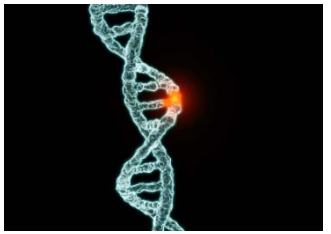
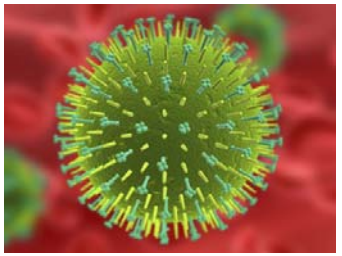
Biological Hazards can include…

- Certain types of recombinant DNA/ RNA
- Microorganisms infectious to humans, animals or plants:
  - bacteria, viruses, fungi, parasites, prions, viral vectors



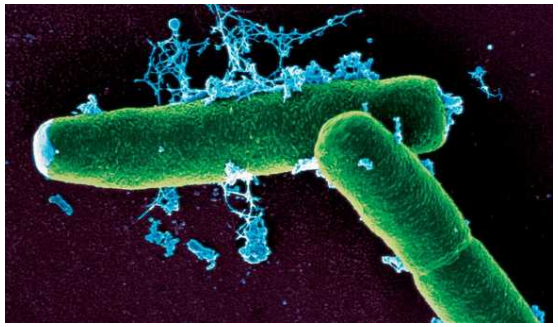


- Biological active agents:
  - toxins, allergens, venom

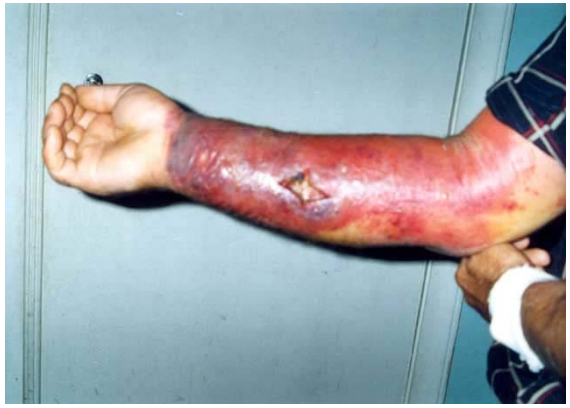
- Other potential biohazardous agents:
  - Blood, bodily fluids, tissue samples (human or animal), diagnostic samples, live vaccines
  - Various cell lines
  - GMOs

Bacteria

- Single celled prokaryotes, lacking a nucleus and other membrane-enclosed organelles
- Typically between 0.5 and 5.0µm in size
- Can induce an immune response (inflammation), secrete endotoxins, produce surface-associated endotoxins (LPS), or form spores that enhance survival
- Many can colonize the body without causing disease, unless there is a disturbance in the immune system, or exposure in high doses in lab activities
- Infections with pathogenic bacteria almost always result in illness



Bacillus anthracis



E. coli

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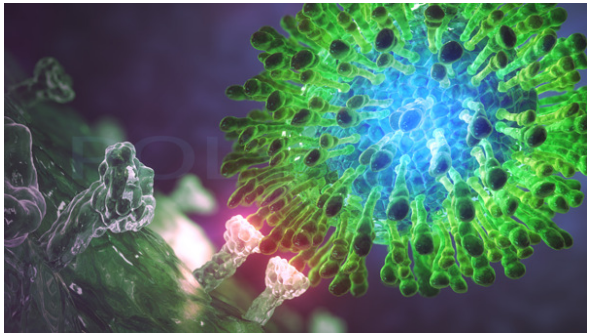
Viruses

- Smallest of replicating microorganisms that can infect human and animal hosts
- Typically between 20 - 300nm
- Various mechanisms to redirect existing host machinery and metabolic functions to replicate
- Easily mutated, so conversion/adaptation to new host is quick
- Some viruses can produce:
  - Latent infections (e.g. HIV)
  - Persistent infections (e.g. HCV)
  - Carcinogenic (e.g. HPV, HBV, HCV, HTLV, EBV)



Hepatitis C Virus entry

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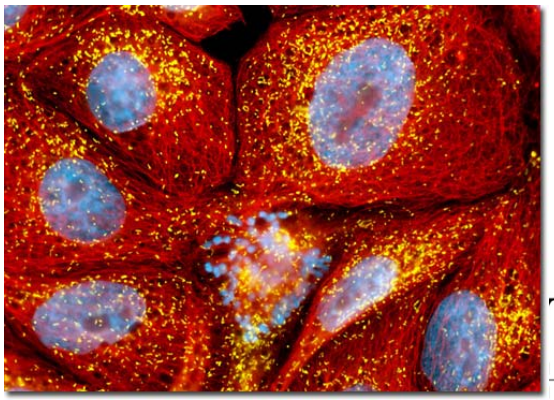
Cell Lines

- Cell lines do not generally pose a risk to the individuals but they have the potential to contain pathogenic organisms – bacteria, fungi, viruses, prions, recombinant virions. These can exist:
  - Naturally
  - Intentionally (via transformation or recombination)

- Cell lines may be infectious without your knowledge, or intentionally injected with a pathogen

- Primary cultures can be at risk of contamination with infectious agents

- Cell lines should be handled and manipulated at the containment level required for the contaminating agent of higher risk, e.g. a risk group 2 agent (e.g. virus) cultured in a risk group 1 cell line should be handled in a CL2 facility



Recombinant DNA

Genetic material from more than one source (natural or synthetic) can be combined to construct novel rDNA

Widely used practice today:

- Production of transgenic animals
- Cloning in expression vectors
- Production of viruses

Level of risk depends on:

- the source of DNA being transferred
- the vector
- the host
- Encoded gene/gene product

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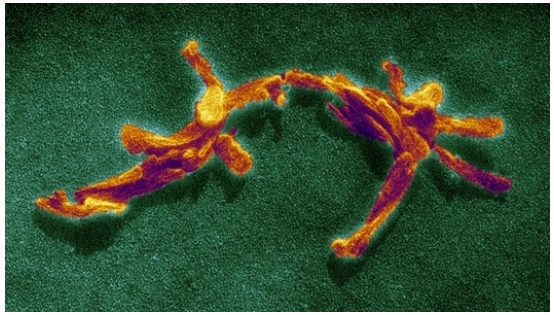
Prions

• Prions are small, proteinaceous infectious particles that are the cause of progressive neurodegenerative diseases in humans and animals known as Transmissible Spongiform Encephalitis

• Prions can induce normally folded proteins to convert to disease-associated misfolded isoforms

• Resistant to destruction

• Accumulation of large amounts of the stable misfolded proteins can cause tissue damage and cell death



e.g. CJD, vCJD, CWD, BSE, Scrapie, Kuru

Precautions:

- Handle tissues as in a CL3 facility
- Handle formalin-fixed tissues and paraffin-embedded blocks as infectious
- Follow up-to-date disinfection protocols (**NOT** regular bio-waste stream)
- Appropriate PPE, avoiding cuts/punctures

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Biological Toxins

- Biological toxins are naturally produced poisonous substances by the metabolic activities of microorganisms, plants and animals.
- Can cause adverse effects, health effects (in low levels), severe incapacitation, possibly death of humans/animals
- Can also be artificially produced (chemical synthesis, genetic engineering)
- Compared to pathogens, toxins are easy to control – they do not replicate, not infectious, and difficult to transmit person to person

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Classification of Biological Agents

Risk groups associated with a pathogen is based on the following risk factors:

- Pathogenicity/Virulence
- Route of infection
- Mode of transmission
- Survival in the environment
- Infectious dose
- Availability of effective preventative/therapeutic treatments
- Host range
- Natural Distribution
- Impact of introduction and/or release into the environment

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Classification of Biological Agents

Risk Group	Individual	Community	Implications	Example
1	Low	Low	Unlikely to cause disease in healthy workers or animals	• Earthworms • *E. Coli K-12* • Asporogenic *Bacillus subtilis*
2	Moderate	Limited	Rarely cause serious human or animal disease	• *E. coli* O157:H7 • Influenza viruses types A, B, C
3	High	Low	May cause serious disease	• HCV, HIV, SIV • *Bacillus anthracis* • Prions
4	High	High	Likely to cause very serious disease	• Ebola virus

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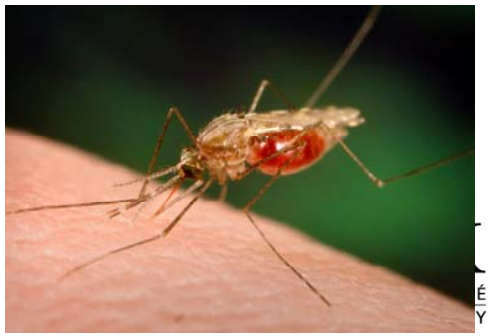
Naturally Occurring Biological Agents

Biological agents can adversely affect human health in many ways

Biohazardous agents are found in the natural environment – in soil, water, plants, animals, humans

Handling these in the lab, or genetically modifying them may enhance their lethal properties, or make them resistant to available treatments or preventives

Since many biological agents reproduce rapidly and require minimal resources for propagation, they can pose a potential danger in a wide variety of occupational settings, and are therefore handled under proper containment



Module 3:

Biosafety

&

Biosecurity

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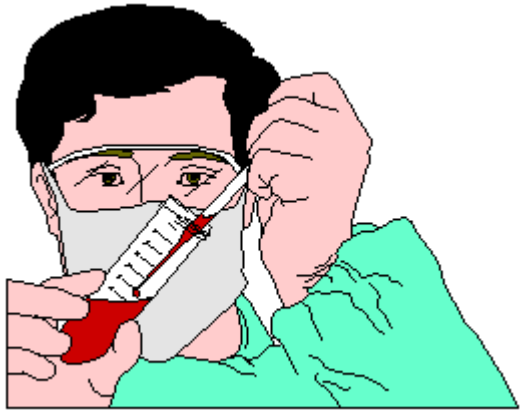
Terminology

- Biohazard
- Biosafety
- Biosecurity



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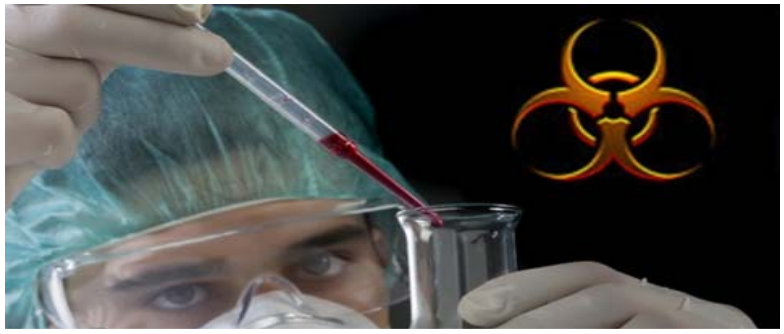
Biosafety

The **combination of principles**, technologies, practices and measures that are implemented when handling Biohazardous materials to:

- **Protect personnel** from unintentional exposure,
- **Contain** the biological agent(s) during manipulation
- **Prevent** environmental **release**



How is Biosafety Achieved?

Administrative controls

- Training
- Issuing certificates
- Authorizations of work
- Compliance verification
- Controlling import/transfers of Biohazardous material
- Inventory control
- Proper disposal and waste management

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How is Biosafety Achieved?

Engineering controls

- Lab design
- Commissioning and decommissioning of labs
- Annual Certification of Biological Safety Cabinets
- Autoclave safety
- Centrifuge and other equipment safety

How is Biosafety Achieved?

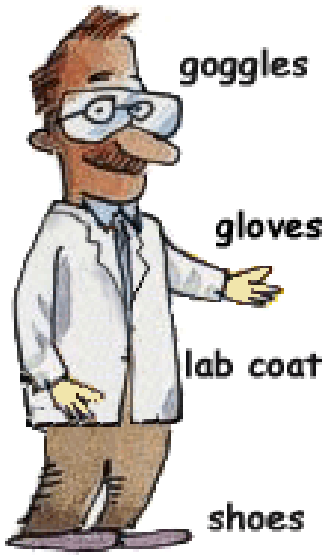
Practices and procedures

- Conducting Risk Assessments
- Appropriate practices in appropriate containment
- Participating in Medical Surveillance Program
- Proper Inventory maintenance

How is Biosafety Achieved?

Proper Use of Personal Protective Equipment

- Gloves
- Safety glasses
- Lab coat
- Closed-toe shoes
- Double gloves when working with blood



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Terminology

- Biohazard
- Biosafety
- Biosecurity

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Brown Student Poisons Ex-Girlfriend with Iodine-125

FROM UNIVERSITY WIRE

Providence police arrested a Brown University graduate student on Friday and charged him with poisoning two fellow students — one of whom is his ex-girlfriend — with a radioactive chemical he allegedly stole from a Brown laboratory.

Short Takes

According to Brown News Bureau Director Mark Nickel, Cheng Gu, a student in molecular pharmacology, prepared a chicken and vegetable dish laced with iodine-125, which he then served Yuanyuan Xiao and her roommate James O'Brien at their home.

Geiger counter would have registered the increased amounts of radiation on him.

Nickel said that iodine-125 is kept locked up at all times, except when it is being used in an experiment.

"As far as the University's procedures for holding and storing the material," everything was done correctly, Nickel said. "There is no indication of any problems with security."

However, when the substance is being used, he said that it is necessary to rely on the experimenter's judgment.

Marie Stoeckel, chief of the occupational and radiological health office of the Department of Health, confirmed that this degree of human woman may have been contaminated," Nickel said, had it not been for Morin and Jacob. "This is an example of things working the way they're supposed to work."

Executive Vice President for Public Affairs and University Relations Laura Freid echoed Nickel's approval of the handling of the situation, by both Brown Risk Management officials and police officers.

[Brown Daily Herald, Nov. 16]

Princeton appointment protested

A handful of Princeton University students joined about 30 other people assembled outside the University's main gate Saturday to protest the appointment of Peter

their lives are more valuable than those of animals. His views on euthanasia and animal rights come from a belief that life necessitates rationality, autonomy and self-consciousness.

Marie Tasy, director of Public and Legislative Affairs for New Jersey Right to Life noted that Singer's presence at other universities has caused protest by students and faculty.

At the protest, Tasy said she felt Singer was "trying to establish a new system of ethics" for society. "He's saying we should be able to judge the quality of life for another human being," she said. "That's a very dangerous philosophy.

Director of Communications

Jennifer Hotz, president of the Mercer Country chapter of New Jersey Right to Life, said she feels the University acted inappropriately by hiring Singer.

"Princeton University had an opportunity to hire someone who advocates life-affirming values. Instead, they chose someone who advocates the killing of disabled infants," she said.

Eric Wang, one of the few students present at the protest, said he attributed the lack of student involvement at the event to a "general sense of apathy."

Patti Staley, president of the Mercy County chapter of Citizens Concerned for Life, said she is very disturbed by Singer's appointment to

Biosecurity

Measures that are employed to protect Biohazardous materials or critical relevant information against loss and/or theft by those who intend to pursue intentional misuse and/or release of infectious material and toxins

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How is Biosecurity Achieved?

Physical barriers

- Buildings access controlled, locks, key card access, self-locking doors
- Structural design to increase the level of security to reflect the transition from general public to laboratory zones
- Animal Care Facility, Life Science Building,

  - departmental design to control traffic flow patterns and access

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How is Biosecurity Achieved?

Psychological barriers

- Security personnel, cameras
- Obvious presence of identifiable security personnel
- Use of cameras, mirrors, mirrored domes and other monitoring tools
- Overall awareness of personnel, people and behaviors

How is Biosecurity Achieved?

Monitoring activities

- Patrols
- Support staff (DOHS personnell, Area HSOs, Biosafety Officer)
- Lab Technicians
- Key/access controls

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How is Biosecurity Achieved?

Personnel clearance

- Keys
- Cards
- Access given to authorized personnel

Biosecurity

- There is growing concern on the use of biological agents as weapons

- Be aware of what is going on in your lab, surroundings

- Report any strange persons, missing material – to your supervisor, Security, Biosafety Officer

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Who is Responsible for Safety?

SUPERVISORS

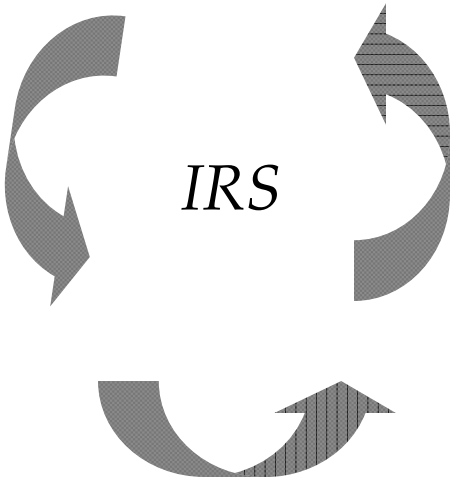
President

Executive

Deans

Chairs

Principal Investigators



WORKERS/STUDENTS

Researchers

Staff

Students

EMPLOYER

York University

IRS - Internal Responsibility System

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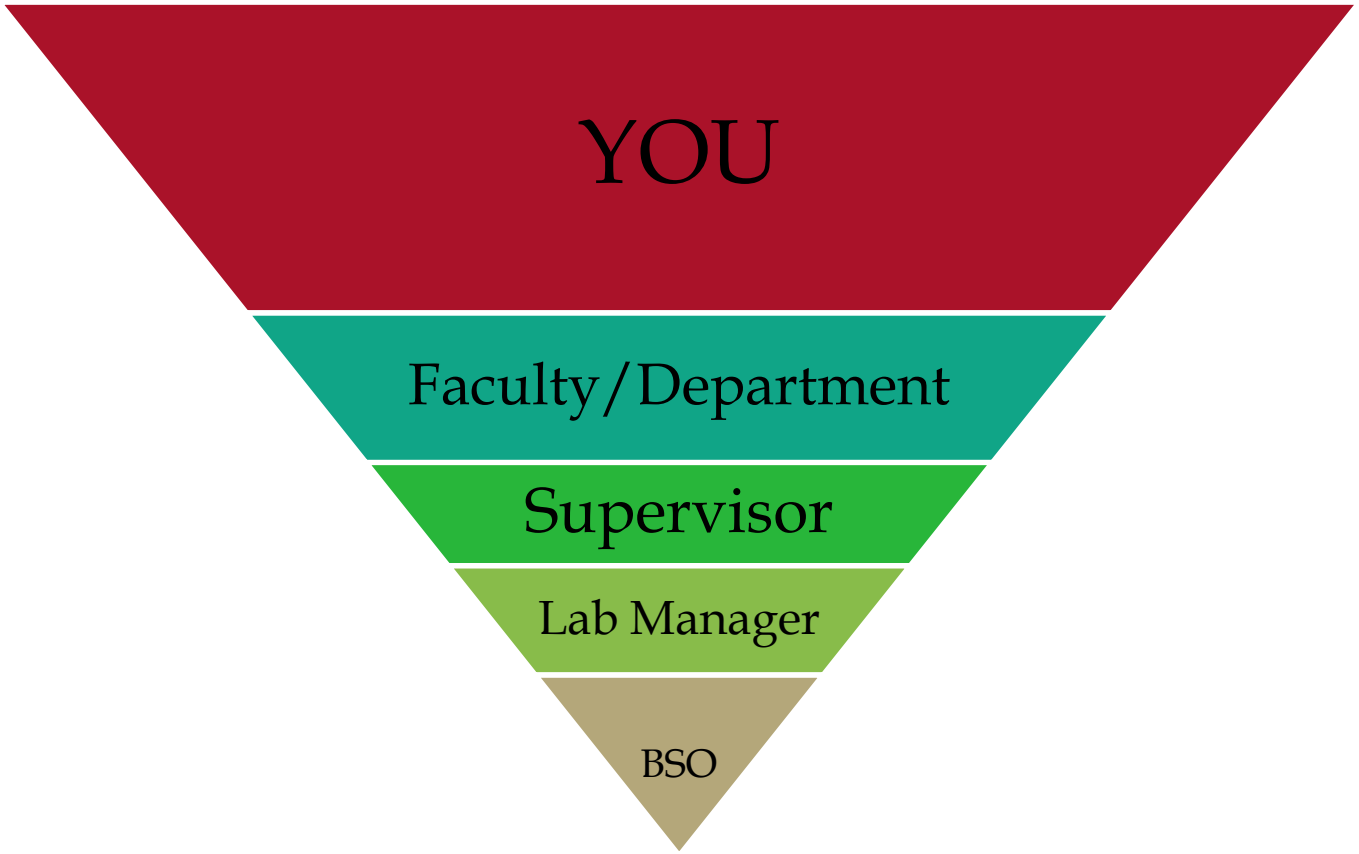
Accountability



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Roles and Responsibilities

You:
Ultimately responsible for all your work and results, compliance to safety policies, adherence to program requirements

Lab/PI:
Informs you of specific hazards, ensures lab is compliant with Biosafety program

Faculty:
Resources, services, guidance, support

Department of Health and Safety, Biosafety Officer/Biosafety Committee:
Administration, training, management, compliance verification, guidance, developing and maintaining program according to regulations and conveying/translating information to you

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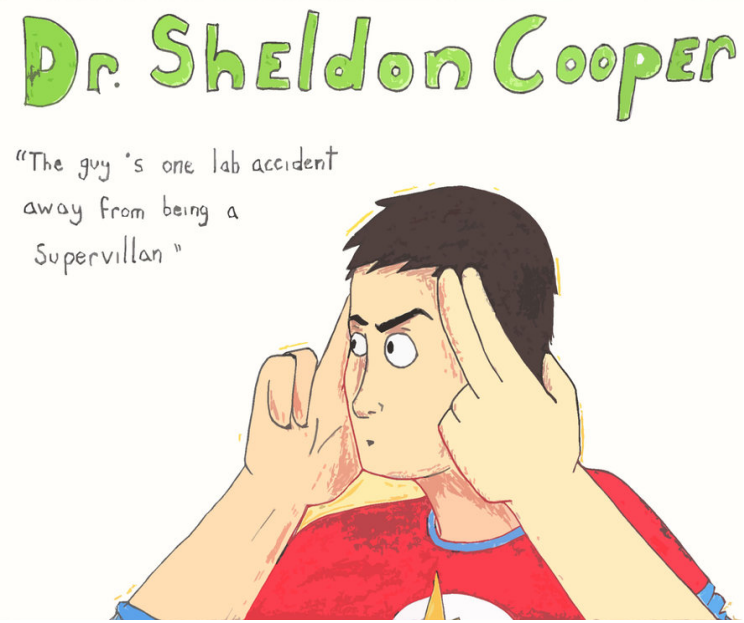
Module 4:

Lab-Acquired

Infections



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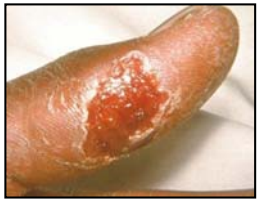
Lab-Acquired Infections

- **Laboratory Acquired Infections** (LAIs) are any infections, whether symptomatic or asymptomatic, acquired through **laboratory or laboratory-related activities**

  - Top 5 laboratory incidents/accidents resulting in LAIs:

    - ✓ Spills and splashes
    - ✓ Infected needle-stick injuries
    - ✓ Aspiration through pipette
    - ✓ Laceration with a sharp object (e.g. broken glass)
    - ✓ Bites or scratches from an infected animal

Pseudomonas



Cut from sharp object

Needlestick injury

Mouse bite

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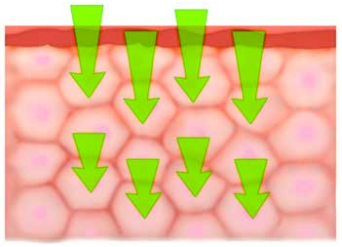
Biological Hazards Routes of Entry

- Inhalation

- Absorption



- Injection

- Ingestion

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Lab Acquired Infections

Working with biological agents involves a certain level of risk

→ Case study: Attenuated Lab-adapted strains – *Yersinia pestis* strain

CDC Home

Centers for Disease Control and Prevention
CDC 24/7: Saving Lives. Protecting People.™

MMWR
All CDC Topics
Choose a topic above
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A-Z Index A B C D E F G H I J K L M N O P Q R S T U V W X Y Z #

Morbidity and Mortality Weekly Report (*MMWR*)

MMWR

Recommend 25 Tweet 2 Share

Fatal Laboratory-Acquired Infection with an Attenuated *Yersinia pestis* Strain --- Chicago, Illinois, 2009

Weekly
February 25, 2011 / 60(07);201-205

- **Lab-adapted strain**
- Symptoms for 3 days
- Died due to cardiac arrest

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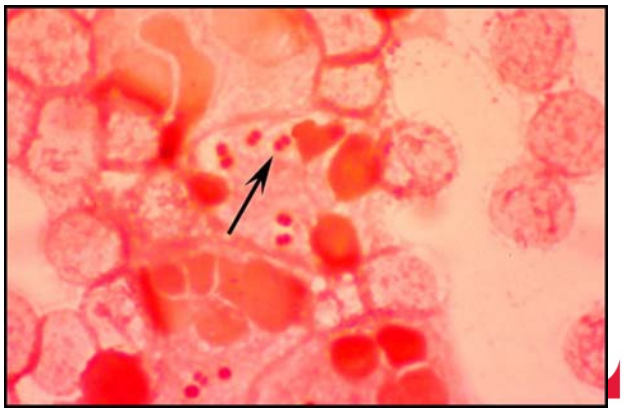
Lab Acquired Infections

Death of 25 year old Research Associate due to exposure to *Neisseria meningitidis* (risk group 2) – May 2012

- **Died 17 hours** after exhibiting the first symptoms
- He had been inoculating live *Neisseria meningitidis*, outside of a Biosafety cabinet
- Lab had **over 20 years experience** working with this bacteria

Neisseria meningitidis, a bacterium that causes roughly 1000 cases of meningococcal disease and 75 fatalities in the United States each year



Lab Acquired Infections

The findings of the Federal Investigation cited 3 serious violations:

1. Failure to work with safety enclosure while working with viable microbial cultures

2. Failure to provide training on the signs and symptoms of illness associated with the microorganisms in use

3. Failure to provide the appropriate vaccines to workers

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Factors Contributing to Lab Associated Infections

- 20% are caused by equipment failure
- 80% are caused by human errors:
  - **Lack of knowledge of biological agent**
  - **Lack of proper emergency responses**
  - **Lack of training, passing down of bad habits**
  - **Carelessness, ignorance in work practices**

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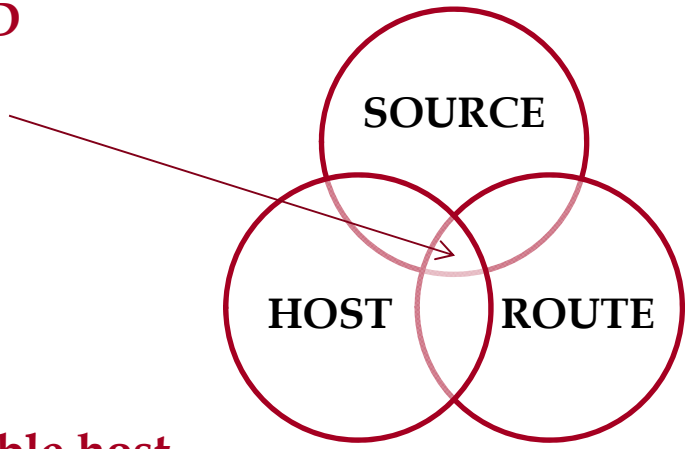
Lab Acquired Infections

Source of infection

- Microorganisms
- Cells and tissues
- Blood and body fluids
- Any items contaminated with the above

LAB ACQUIRED INFECTION



Susceptible host

- Immune system
- Vaccination status
- Age
- Pregnancy

Route of transmission

- Percutaneous inoculations
- Inhalation of aerosols
- Contact of mucous membranes
- Ingestion

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Where do I find this information?

ONLY IN CANADA

Internationally Recognized Resource.
Designed, Researched and Maintained by PHAC & CFIA

PATHOGEN SAFETY DATA SHEETS

http://www.phac-aspc.gc.ca/lab-bio/res/psds-ftss/index-eng.php

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Module 5:

Risk Assessment

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Risk Assessment

Why Bother?

A risk assessment is a very important part of biosafety

- It helps determine to which risk group a biological agent belongs to and under what containment level it should be manipulated

Determining how much risk allows labs to implement operational practices that will reduce or eliminate the risk to lab workers and to the community

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Risk Assessment

How Do You Perform A Risk Assessment?

1. Identify biohazardous material

2. Assign a risk group (1 - 4) to each biohazardous material

3. Choose appropriate measures to protect researchers:
   - Containment level (1-4)
   - Equipment used (protect environment), PPE used (protect worker)
   - Experimental procedures and practices
   - Emergency procedures
   - Waste

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BioRisk Assessment

- Agent Characteristics & Biological Material
- Personnel Supervising & Personnel Using the Material
- Environment: Laboratory, Facility, Government Regulations
- Experimental Protocols
- Lab Practices & Equipment

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Agent Characteristics & Biological Material

- Have you used this agent before?
- Do you know the source and whether it has been proved it is non-replicating
- What are the characteristics of the material upon receipt?
- What are the implications of the manipulations you are planning?
- Is this a material for which Lab Associated Infections/exposure concerns have been reported?

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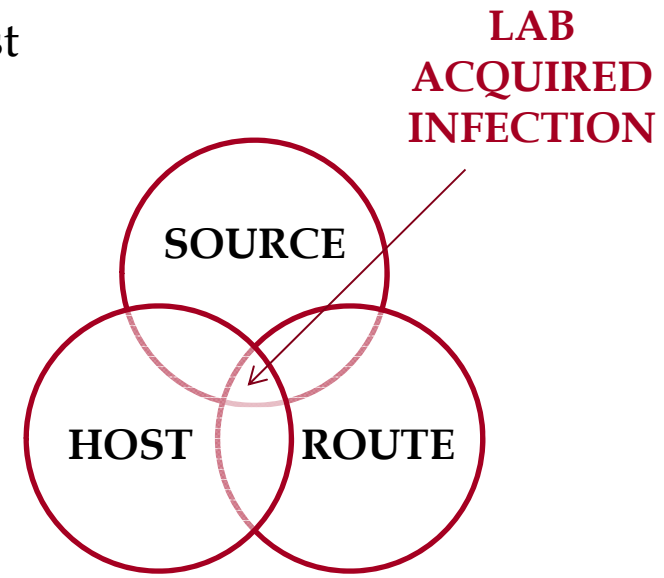
Agent Characteristics & Biological Material

Lab Associated Infection/exposure to biohazardous agents depends on:

- Pathogenicity/virulence
- Route of infection
- Mode of transmission
- Survival in environment/outside of host
- Infectious dose
- Prophylaxis/treatment methods
- Host range
- Ability to release into the environment



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Escherichia coli, enterohemorrhagic - Material Safety Data Sheets (MSDS)

MATERIAL SAFETY DATA SHEET - INFECTIOUS SUBSTANCES

SECTION I - INFECTIOUS AGENT

NAME: ***Escherichia coli,*** **enterohemorrhagic**

SYNONYM OR CROSS REFERENCE: Enterohemorrhagic *Escherichia coli* (EHEC), Verotoxin producing *Escherichia coli* (VTEC), Shiga toxin producing *Escherichia coli* (STEC)

CHARACTERISTICS: Gram negative rod; motile, aerobic; produce Vero / Shiga toxins (VT/STx), 2 types, VT1/Stx1 and VT2/Stx2; serotyping to determine somatic and flagellar antigens

SECTION II - HEALTH HAZARD

PATHOGENICITY: Hemorrhagic colitis, intestinal disease accompanied by cramps and abdominal pain; initially watery, followed by bloody diarrhea; low grade fever; last about 8 days; 5-10% of hemorrhagic colitis victims may develop hemolytic uremic syndrome (HUS); affects all ages, higher death rates occur in elderly and young; can cause thrombocytopenic purpura (TTP) in elderly

EPIDEMIOLOGY: Sporadic and in outbreaks of bloody diarrhea; associated with 15-30% of patients where no other pathogen has been identified; main EHEC serotype in North America from infections is *E. coli* 0157:H7

HOST RANGE: Humans; animals (O157:H7 - piglets, calves and cattle)

INFECTIOUS DOSE: Appears to have low infectious dose, may be similar to that of *Shigella* spp.,10 organisms by ingestion

MODE OF TRANSMISSION: Ingestion of contaminated food (undercooked hamburger meat, unpasteurized milk); fecal-oral transmission; person-to-person transmission (extremely high)

INCUBATION PERIOD: 2-8 days (median of 3-4 days)

COMMUNICABILITY: Communicable for duration of fecal excretion (7-9 days); 3 weeks in one third of children

SECTION III - DISSEMINATION

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PATHOGEN SAFETY DATA SHEET - INFECTIOUS SUBSTANCES

SECTION I - INFECTIOUS AGENT

NAME: *Escherichia coli*, enteroinvasive

SYNONYM OR CROSS REFERENCE: EIEC [1], [ ], intestinal pathogenic *E. coli* [ ], bacillary dysentery [1].

CHARACTERISTICS: Enteroinvasive *Escherichia coli* (EIEC) are in the family Enterobacteriaceae [2]. They are Gram negative, rod shaped, non-spore forming, motile with peritrichous flagella or nonmotile, grow on MacConkey agar (colonies are 2 to 3 mm in diameter and red or colorless), and are capable of aerobic or anaerobic growth [3]. Strains belonging to EIEC are biochemically, genetically, and pathogenically closely related to *Shigella* spp. [1].

SECTION II - HAZARD IDENTIFICATION

PATHOGENICITY/TOXICITY: EIEC causes bacillary dysentery [1], an acute ulcerative infection of the large intestine [1], [4]. EIEC invade cells of the colon and causes watery diarrhea (might be bloody), fever, and abdominal cramps [2], [ ]. In severe cases, the bacteria may attack the colonic mucosa, invading epithelial cells, multiplying, and causing ulceration of the bowel [1].

EPIDEMIOLOGY: EIEC is endemic in most developing countries and may cause occasional outbreaks in industrialized countries [4]. Species of *Shigella* are the major cause of bacillary dysentery, although up to 10% of cases are caused by enteroinvasive *E. coli*. EIEC are rare in United States and Canada, and are less common than ETEC and EPEC strains in the developing world [2]. Three large outbreaks in the United States have been reported. EIEC infections primarily affect children under 5 years living in developing countries [6].

HOST RANGE:Humans [2].

INFECTIOUS DOSE: 10^6-10^{10} organisms [4].

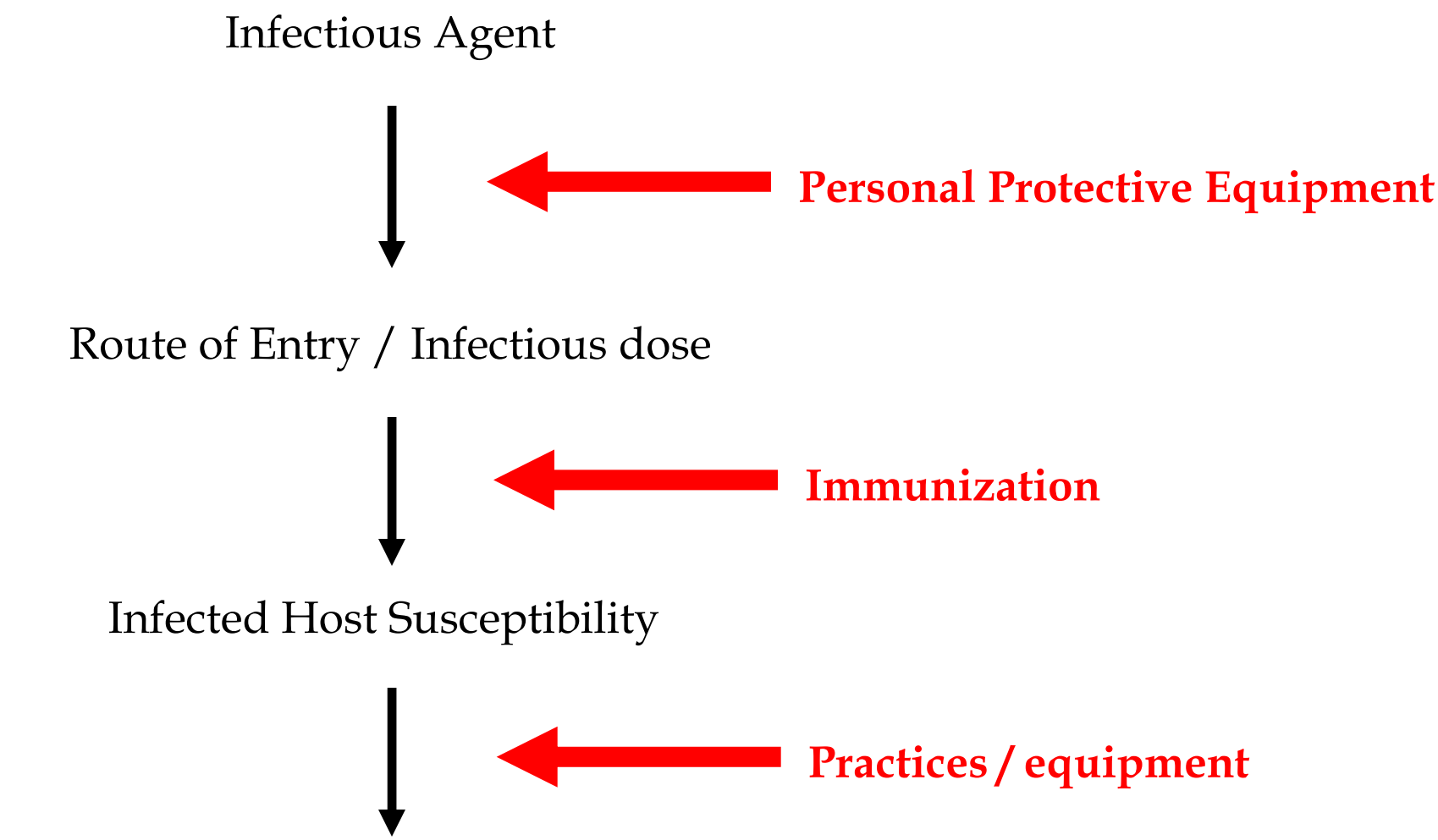
MODE OF TRANSMISSION: EIEC are spread by the fecal/oral route [1], [7]. Contaminated food and water are the usual vehicles for the spread [5], [7]. Food-borne outbreaks have occurred [7]. Person-to person transmission can also occur [1].

INCUBATION PERIOD: The incubation period is between 2-48 hours with an average of about 18 hours [8].

COMMUNICABILITY: Yes. Person-to-person transmission is possible but is uncommon [1], [6], [7].

SECTION III - DISSEMINATION

Risk Assessment / Hazard Analysis



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Blood Borne Pathogens

Include infectious microorganisms that can be found in:

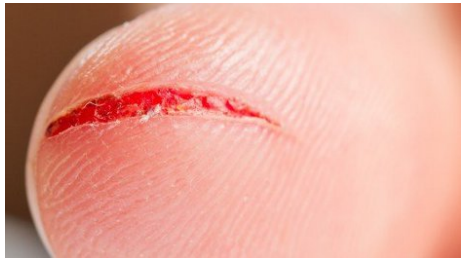
- **Human blood products** (whole blood, plasma, serum, platelets, white blood cells)
- Other **human bodily fluids** (semen, cerebrospinal fluid, vaginal secretions, urine, synovial fluid, pleural fluid, peritoneal fluid, amniotic fluid)

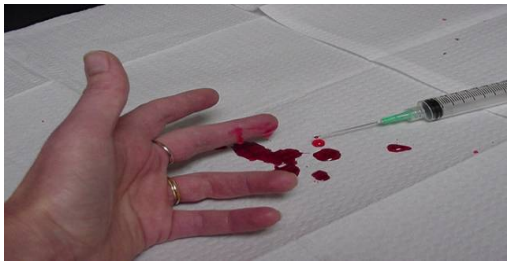
- **Human tissue and/or organs**
- **Primary cell cultures**

Blood Borne Pathogens

Occupational exposure to bloodborne pathogens can occur:

- by infection of mucous membranes (e.g. splash to eye, nose, mouth)
- directly into the bloodstream through skin that is damaged (scraped, cut, abraded)
- by puncture wound (through needle-stick injury)





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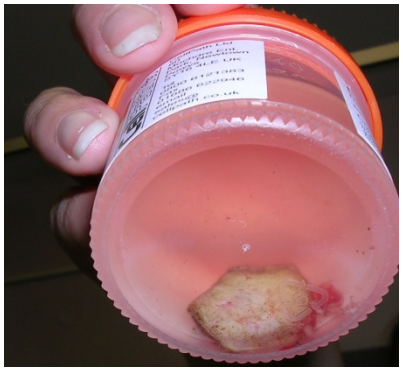
Patient/Clinical Samples

Unless otherwise stated, patient samples and clinical specimens **should be considered** as infectious due to the **potential presence** of bloodborne pathogens

Examples of patient/clinical samples can include:

- Blood
- Semen
- Urine
- Body fluids
- Tissues/biopsies



Blood Borne Pathogens

Risk of Exposure depends on:

- Pathogen/biological agent involved
- Type of body fluid
- Route of exposure
- Duration of exposure
- Volume of blood involved in exposure
- Concentration of virus at time of exposure
- If appropriate PPE worn

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Universal Precautions

A set of strategies developed to prevent of transmission of blood borne pathogens (from blood, bodily fluids and secretions). There are 5 major components:

1. **Risk Assessment**
2. **Hand Hygiene**
3. **PPE and safe work practices**
4. **Environmental Controls**
5. **Administrative Controls**

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BioRisk Assessment

- Agent Characteristics & Biological Material
- Personnel Supervising & Personnel Using the Material
- Environment: Laboratory, Facility, Government Regulations
- Experimental Protocols
- Lab Practices & Equipment

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Personnel Supervising & Personnel Using the Material

- Your knowledge and experience
- Level of mentorship available
- What you work with
- How you work with it
- Health status (Medical Surveillance)
- Due diligence:
  - PPE
  - reporting infractions and near misses
  - Proper inventory maintenance

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Personnel Supervising & Personnel Using the Material

Keeping an **updated** inventory of your biological agents is important:

- Ensures good Biosecurity measures
- Any time an agent is added (imported/created) or removed from a lab, **its movement should be tracked**

What material is presently being used and/or stored in your lab

- ✓ Location
- ✓ Expiry date
- ✓ Use log book
- ✓ MSDS/supplier information

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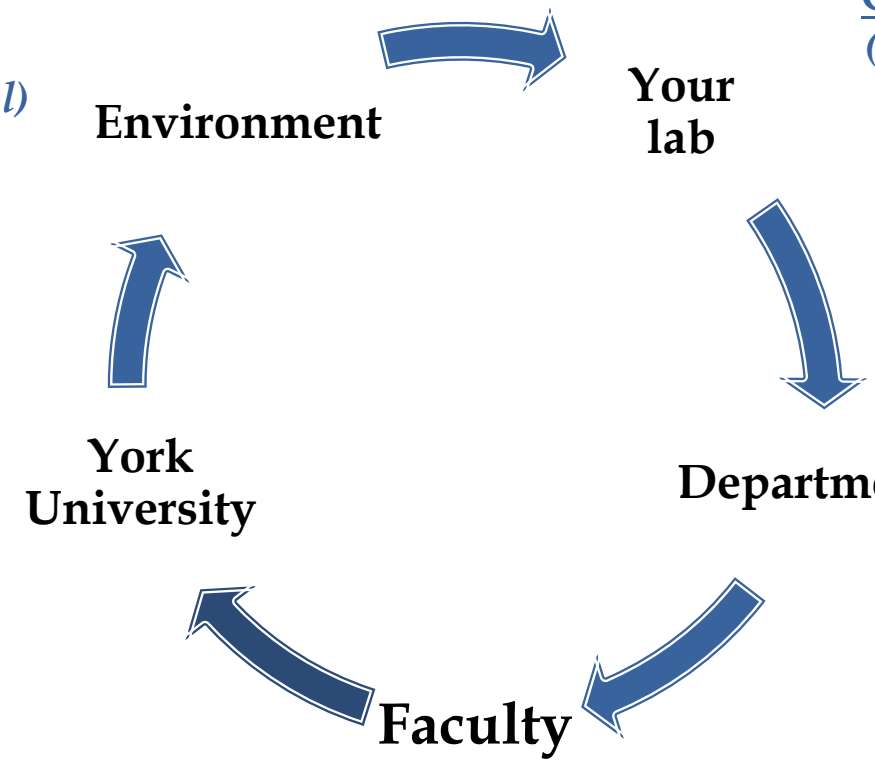
Environment: Laboratory, Facility, Regulations

Regulatory Requirements
PHAC, CFIA, EC, Fed/Prov/Municipal)



Contain your biohazard
(Primary and secondary containment)

Lab Design
(conception, construction, renovations, maintenance)

Control Access
(Physical controls: lab access and inventory)

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Containment of Biological Agents

Primary Containment

- **First** line of defence
- Ensures protection of personnel and immediate environment from exposure
- 'Protective envelope' that encapsulates the infectious agent or animal.
  - Petri dish, vials
  - Biological Safety Cabinets
  - Animal caging equipment

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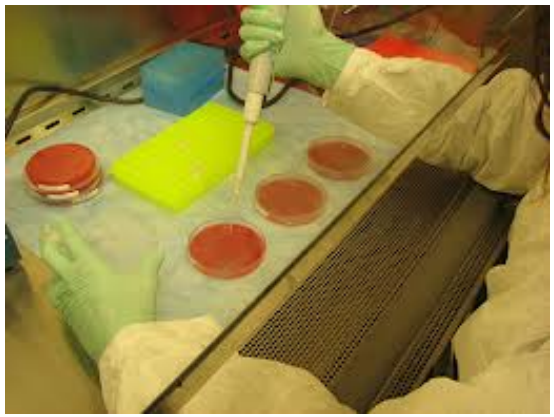
Containment of Biological Agents

Secondary Containment

- Protects the environment external to the laboratory from exposure
- Includes facility design and operational practices, that employs:
  - Directional airflow
  - Air drain and filtration
  - HEPA filtration of lab air
  - Pressure differentials
  - Laboratory design
  - Operational practices



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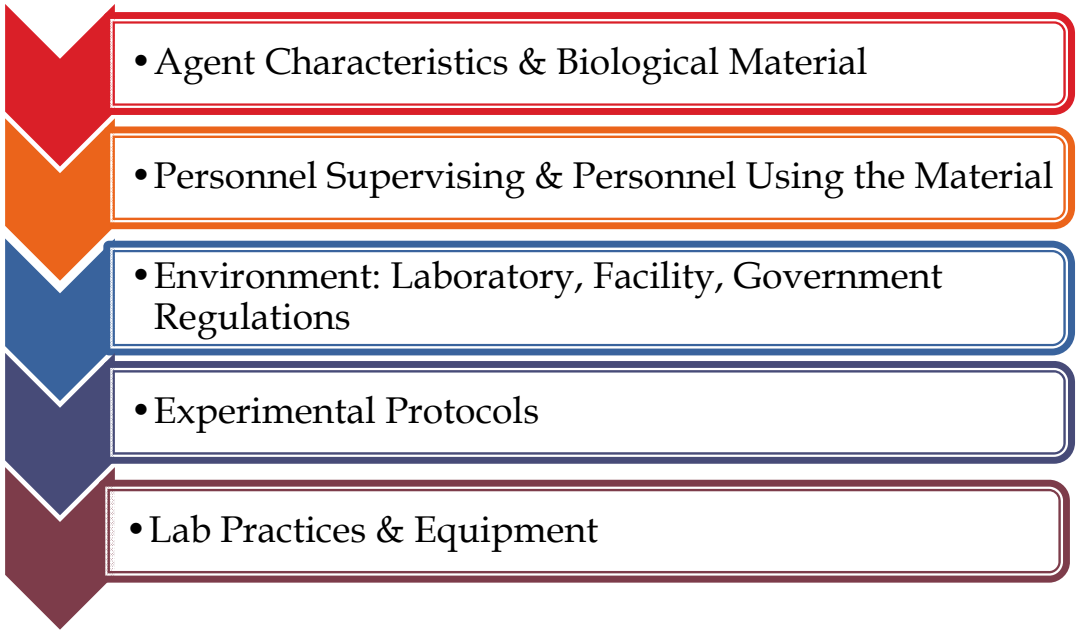
Biocontainment

Classification of organisms into risk groups do not establish the actual handling of biological hazards in the lab

A risk assessment determines the biocontainment level (BCL) of the work – it takes into account:



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Biocontainment Levels

- Biocontainment levels incorporate the **engineering, operational, technical and physical requirements** for handling and manipulating a biological agent

- There are four Biocontainment levels – BCL-1, BCL-2, BCL-3 and BCL-4 – and these need to be certified/inspected regularly

- All biological agents at York University are RG1, RG2 or RG3 pathogens and depending on the agent and risk assessment, they can be used and manipulated in a BCL-1, BCL-2 or BCL-3 facility

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Biocontainment Levels

As the containment level increases so does:

- The potential risk to humans, animals, plants or the environment
- The procedural and facility requirements
- The level of operational containment required
- The degree of protection for personnel, the community and the environment

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Biocontainment Level 1

- Basic laboratory
- Requires no special design features
- Work may be carried out on an open bench
- Biological Safety Cabinet may be present
- Containment is achieved through the use of practices normally employed in a basic microbiology laboratory
- Emergency plan in place

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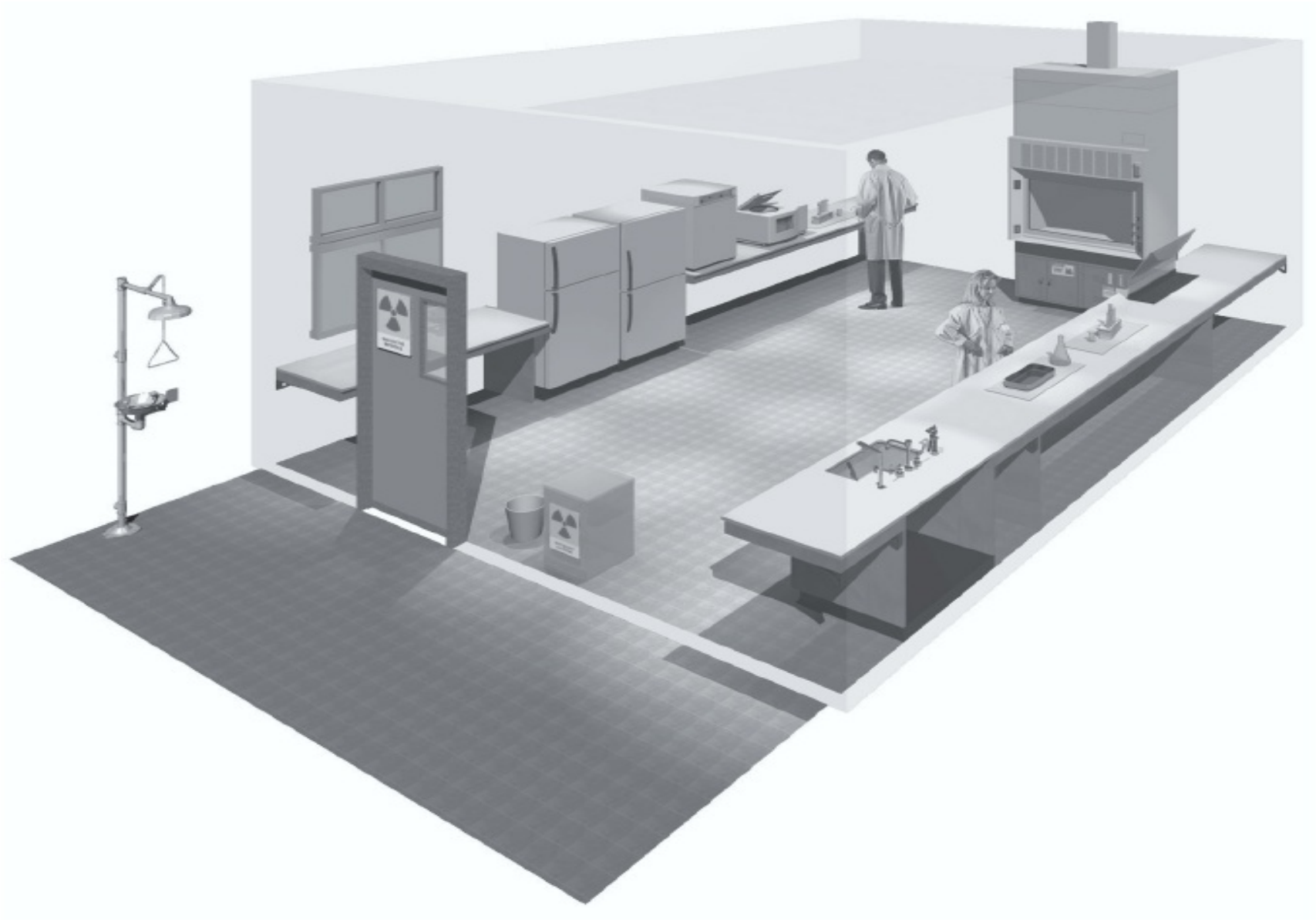
Biocontainment Level 1



Biocontainment Level 2

- Clinical, diagnostic, research & teaching facilities working with Risk Group 2 agents
- Work must be done using primary containment devices such as Class I or Class II Biological Safety Cabinets (with HEPA filters), centrifuges with sealed rotors or safety cups
- Environmental contamination must be minimized by the use of biological waste containers, hand-washing sinks, decontamination facilities (autoclaves)
- Emergency and medical surveillance plan in place
- Access controlled – Biosecurity plan in place

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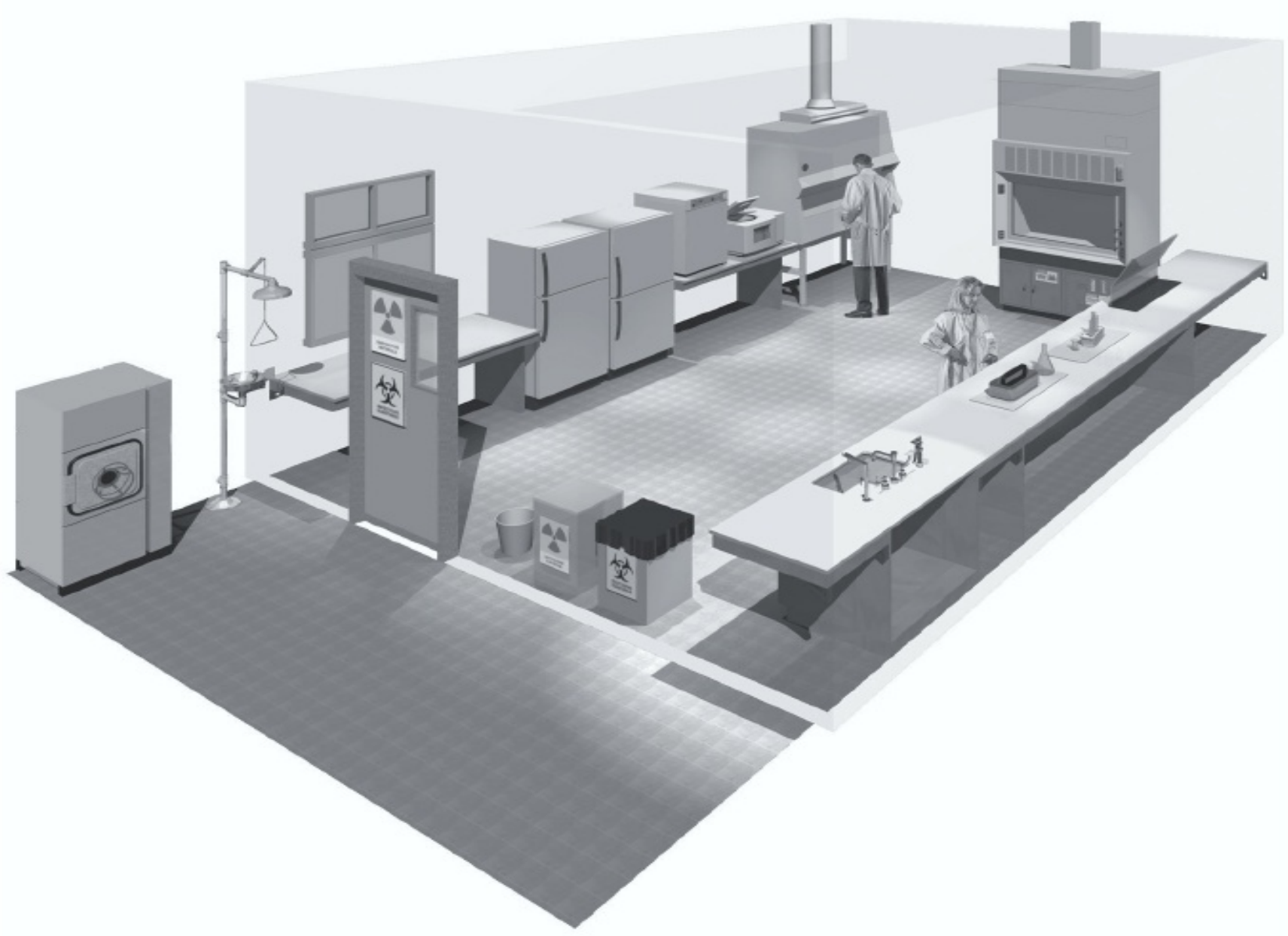
Biocontainment Level 2



Biocontainment Level 2 Labs

- Lots of Biocontainment Level 2 Labs in Canada
  - Biological agents used can cause disease in humans and/or animals, but are unlikely to cause serious disease under normal circumstances.
- Some pathogens handled in a BCL-2 lab may cause serious illness, and even death, in immunocompromised persons:
  - Cold/Flu patients
  - Pregnant
  - Weak immune system

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Biocontainment Level 2 Labs

- Designated BCL-2 lab practices allow the people working in it to safely handle infectious agents that can be transmitted by ingestion, inoculation and through mucous membranes.
    - In most cases the agents handled in a BCL-2 lab are **not** transmitted by the airborne route

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Biocontainment Level 3

- Specialized design and construction
- Operational practices and procedures are very specific
- Standard Operating Procedures (including entry and exit) are strictly enforced
- All staff must undergo specific training on the agents used
- Requires type II or type III biosafety cabinets.
- PPE is very specific, including solid front clothing and dedicated footwear, for use only within the facility
- A medical surveillance program must be in effect

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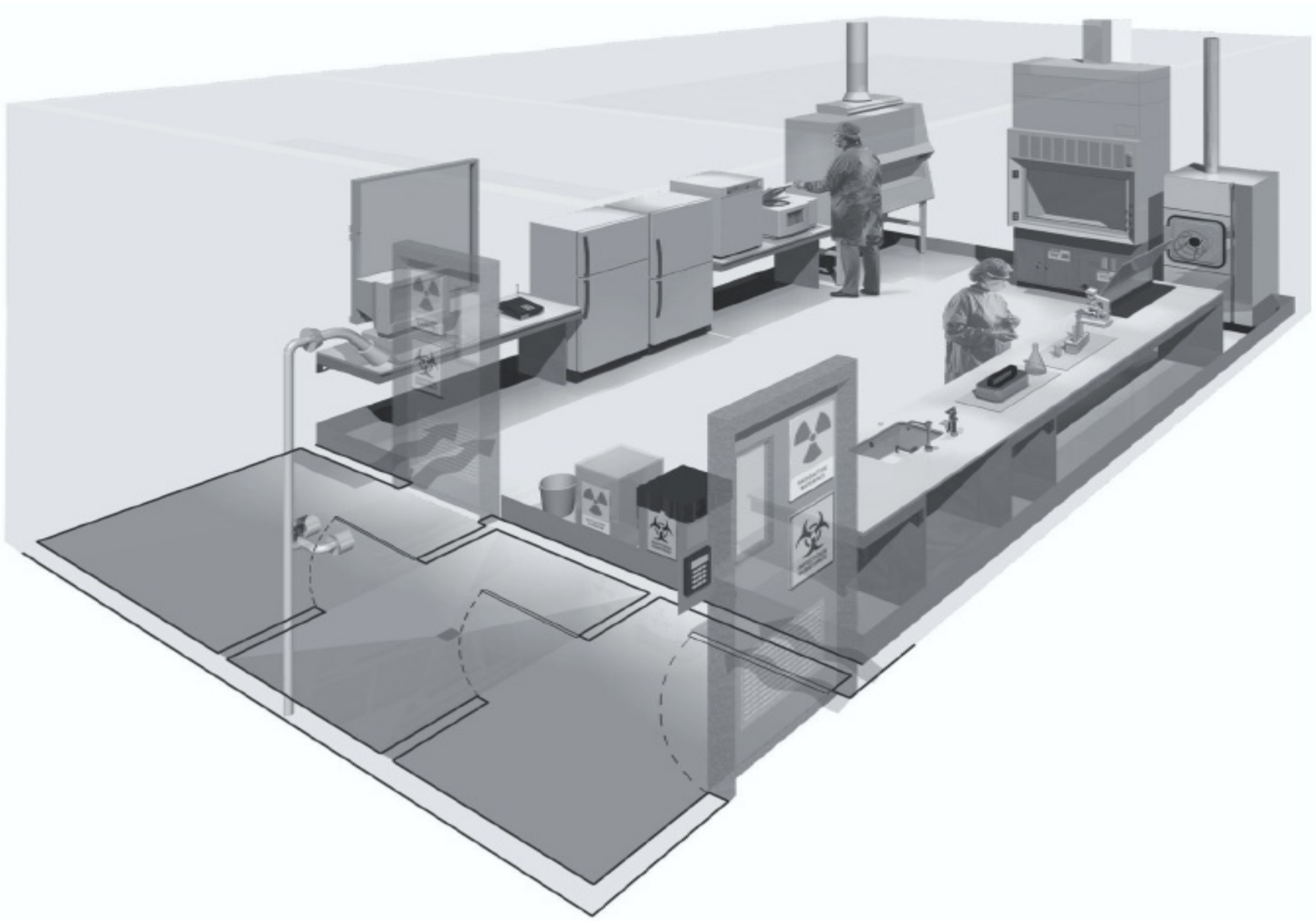
Biocontainment Level 3



Biocontainment Level 4

- The highest level of containment, where **all manipulations** pose a high risk of exposure and infection
- Design specifications are extremely stringent
- The worker is completely isolated from infectious material
- All work is performed in BSC with a positive pressure suit
- Personnel security clearance and qualifications scrutinized
- Entry and exits are through airlocks
- Showers are mandatory before and after entering

There is only one Biocontainment Level 4 facility in Canada (Canadian Centre for Human and Animal Health) in Winnipeg, Manitoba

Risk Assessment Resources

- YU Biosafety Officer
- PHAC Laboratory Biosafety Guidelines
- Supplier information Sheets
  (e.g.) ATCC (Biosafety Level, MSDSs)
- PHAC Pathogen Safety Data Sheets:
  www.phac-aspc.gc.ca/msds-ftss/index.html

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What can

YOU

do to ensure

Biosafety?

BioRisk Assessment

- Agent Characteristics & Biological Material
- Personnel Supervising & Personnel Using the Material
- Environment: Laboratory, Facility, Government Regulations
- Experimental Protocols
- Lab Practices & Equipment

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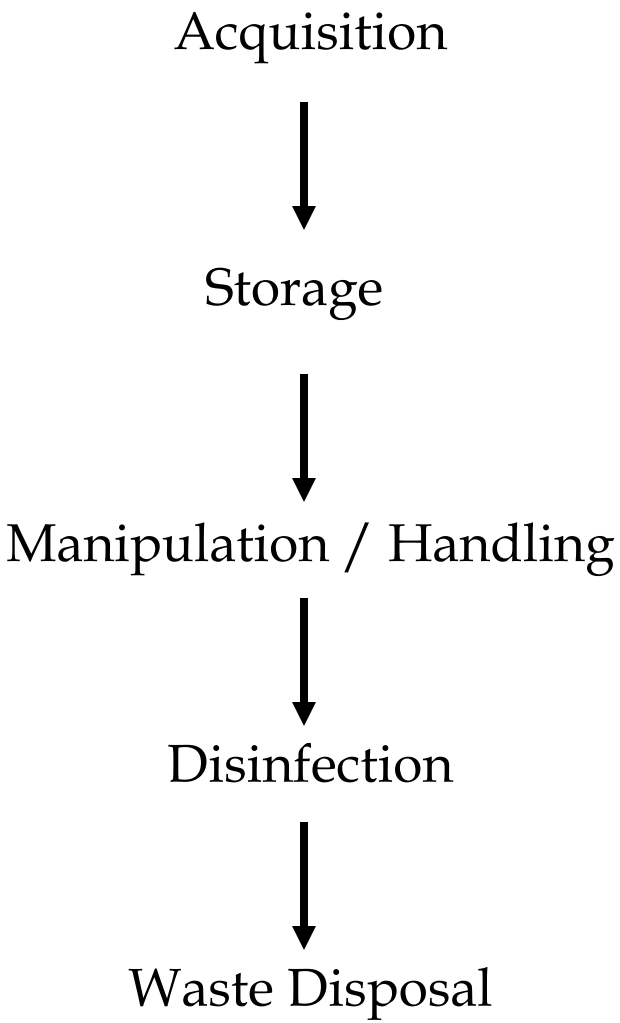
Biosafety in the Lab

Steps to handling biohazardous materials:



- Are the necessary measures in place to handle the sample?
- Where and how is the biohazardous material being stored?
- How is the biohazardous material being manipulated?
- Is the biohazardous material properly transported between labs?
- Is there adequate engineering practices? (BSCs, room design)
- Is the biohazardous material being decontaminated correctly?
- Is the biohazardous material being disposed of properly?

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Module 6:

Good Microbiological Practices

- Good Lab Practices & Equipment

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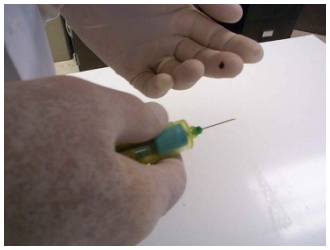
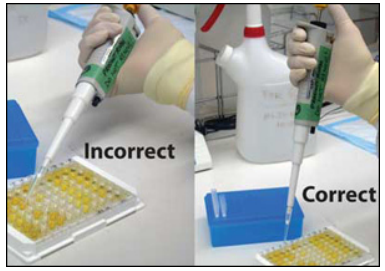
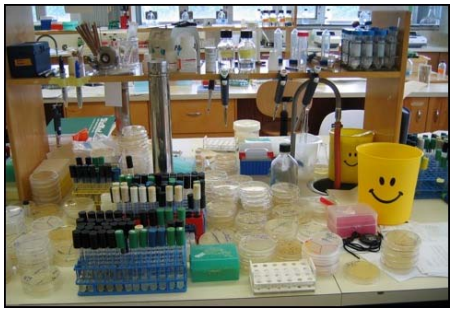
Work practices that may present a risk







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Good Microbiological Practices

Good Microbiological Practice is a basic code of practice that should be applied to all types of work involving microorganisms **irrespective** of containment level

Good Microbiological Practice prevents:

- Contamination of laboratory workers and the environment
- Contamination of the experiment/samples

Good Microbiological Practices

These include, but are not limited to:

- Experimental design, steps
- Uncluttered bench spaces and disinfection of work surfaces
- Use of Personal Protective Equipment
- Using the aseptic technique
- Minimizing use of aerosols
- Proper use of BSCs
- Spills
- Handwashing
- Identification and preparation of biological waste

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Experimental Protocols

Experimental protocols have to be:

- Researched thoroughly
- Designed with safety in mind (along with research)
- Engage the supervisor
- Consider use of alternative biological agents (if applicable) or other tools (e.g. blunt needles instead of sharp needles) to minimize risks of accidents/exposures

Remember, once you begin your experiment, you are in research mode – aspects of biosafety should have been thought of during your design phase

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Experimental Protocols

Some factors to consider when designing experiments:

- Type of agent, mode of transmission, knowledge of signs and symptoms
- Manipulation – what you are doing
- Equipment used (centrifuge, Biosafety Cabinet, needles, other)
- PPE available
- Use of animals (if applicable)
- Vaccinations (if available)
- Training (practical)
- First Aid kit (should be readily available)

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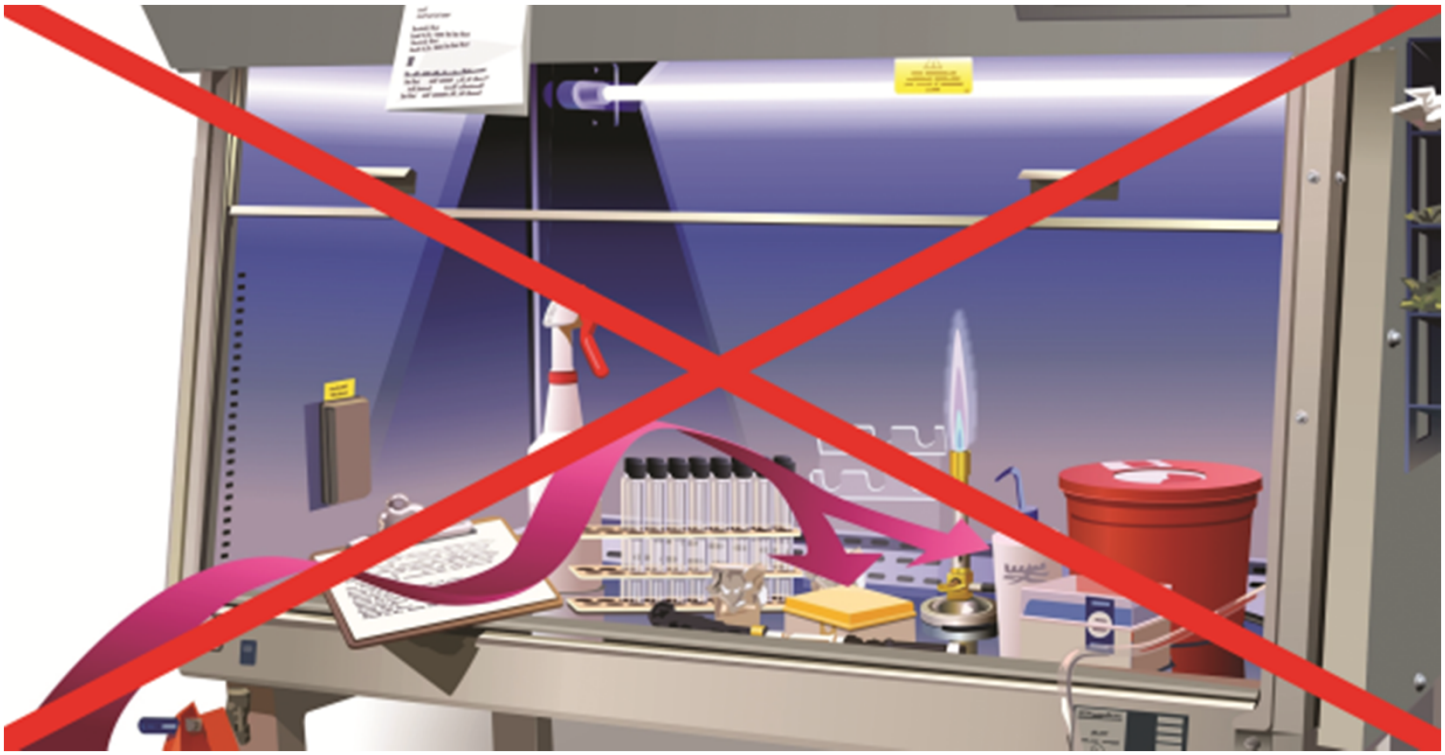
Work Surfaces

Work surfaces have to be kept organised and clutter-free

Over crowding your lab workspace can increase the risk of spills



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Work Surfaces

- Work surfaces (bench tops, fume hoods, BSCs) have to be regularly cleaned with a suitable disinfectant:
    - 10% Bleach
    - 70% Ethanol
    - Virox/Accel (bactericidal and virucidal)

Some factors affecting proper surface decontamination:

- Contact time
- Preparation date/freshness of decontamination solution
- Amount of biological agent

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Personal Protective Equipment

- PPE – creates a barrier between the worker and the source of biological agents – blocking routes of exposure

- Only effective if correctly selected, fitted and cared for
  - Lab Coats and Gowns
  - Gloves
  - Eye Protection
  - Masks and Respirators
  - Footwear

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Personal Protective Equipment

- **Last line of defence against hazards in the lab** (only offers a certain degree of protection)

- Users must be trained on the proper use of PPE

- Wearing PPE should be avoided in public areas - remember to remove your PPE before leaving the lab

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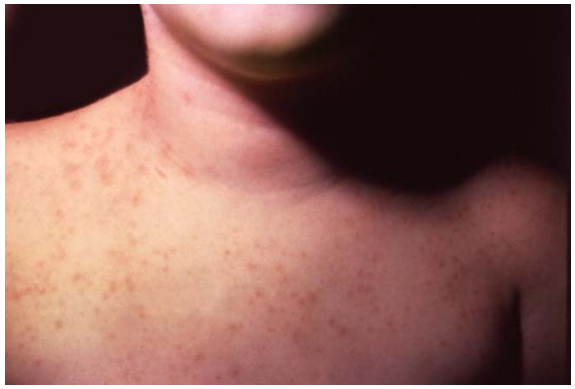
Lab Coats and Gowns

- Lab coats are long-sleeved, knee-length with snaps and fitted cuffs
- Gowns are back-closing
- Must always be worn closed
- Never wash with regular clothing
- Never wear your lab coat in offices, conference rooms, lunch rooms or outside



Echovirus type 9

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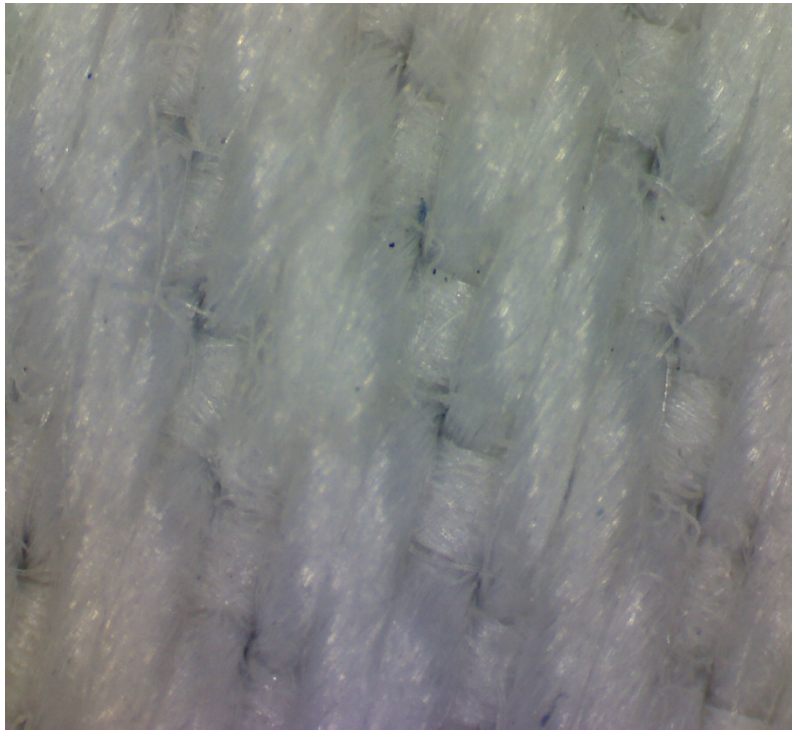
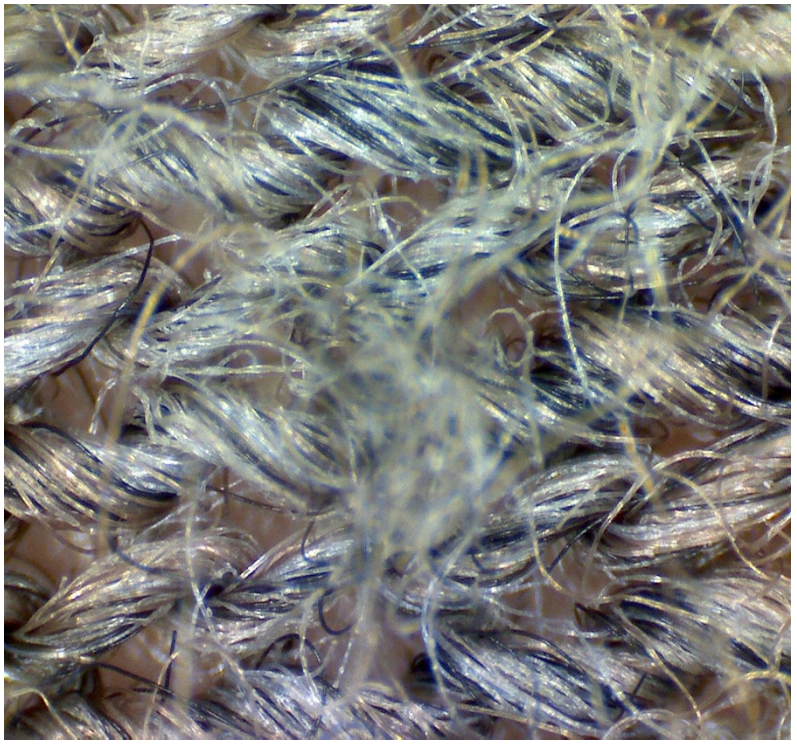
Why you should wear a lab coat?

Lab Coat (200x) **vs.** **T-shirt (200x)**





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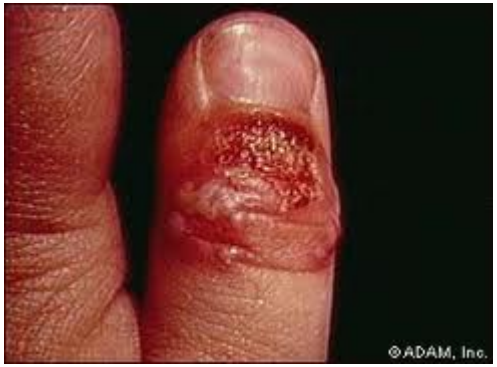
Office of Risk Management

Gloves

…they **ARE** mandatory



Herpes simplex virus (HSV)

- Use gloves appropriate for the task
- Glove selection: latex, nitrile, rubber & vinyl
- Avoid wearing them for longer than 2 hours
- Wash hands once gloves have been removed
- Disposable gloves must be discarded once removed - Do not save for future use
- Dispose of gloves into the proper container – **NOT** in regular garbage
- Do not wear gloves out of the lab (one hand policy), or when touching personal items
- If for any reason a glove fails, resulting in skin contact, consider it an exposure and wash hands immediately - seek medical attention as required

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Proper Glove Removal

(1) Pinch glove near your wrist and pull slowly towards your fingers. Turn the glove inside out while pulling.

(2) Continue holding glove with one hand while removing the other hand from the glove.

(3) Slide finger from glove-free hand under other glove. Slide approximately half of your finger under the glove.

(4) Rotate your finger ~180°, and pull glove outwards towards your fingertips. Turn the glove inside out while pulling.

(5) Holding the glove by the uncontaminated surface, transfer to biohazard waste bin.

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Eye Protection

- Safety glasses are adequate when the potential for splashing is minimal
  - They do not seal to the face, and have gaps at the top, bottom and sides
- Safety goggles provide protection from significant chemical splashes.
- Face shields are required when working with large volumes of hazardous materials -
  - Protection from splashes and flying particles.
  - Face shields must be used in conjunction with safety glasses or goggles.
  - A UV-coated face shield, not safety goggles, is required when working with UV light.

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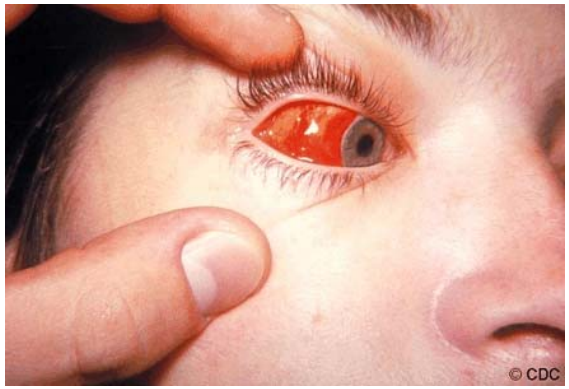
Eye Protection

Avoid the use of contact lenses.



Epstein-Barr Virus Infection

Proper hand washing is important

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Masks and Respirators

Dust masks may be used for some laboratory applications to prevent the inhalation of aerosol- generating experimental procedures

Any other special mask/respirator requirements have to be addressed to Biosafety Officer

Appropriate Footwear

Its simple – shoes worn in the lab must be:

- Closed toe
- Closed heel
- No exceptions

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Procedures to Minimize Potential of Exposure

Opening Tubes

- Manipulate infectious materials within a BSC
- Upon opening, unscrew the cap slightly and wait a few seconds before opening

Pipetting

- Never mix material by suction and expulsion
- Pipettes are calibrated to retain the last drop
- Use plugged pipettes
- Discharge pipettes close to the fluid level and let the contents run down the wall of the container
- Never forcefully expel materials from the pipettes

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Procedures to Minimize Potential of Exposure

Mixing/Homgenizing/Vortexing

- Ensure when blending you are using a leak-proof bearings and a gasket lid
- After mixing, wait a few seconds before opening a lid
- Use a vortex instead of inverting tubes

Centrifugation

- Centrifuge in closed containers.
- Placed in sealed safety cups or rotors
- Open samples in BSC
- Ensure that the centrifuges are well maintained and O-rings are not compromised

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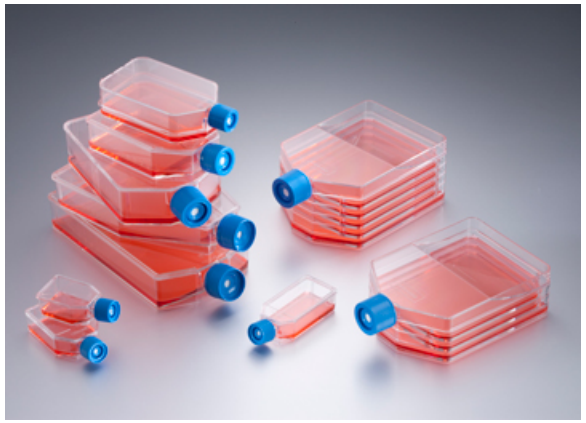
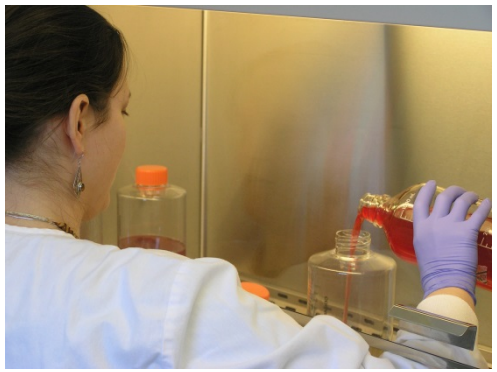
Procedures to Minimize Potential of Exposure

Use of flasks

- Avoid the use of glassware where possible - use plastic tubes, flasks and bottles
- Use screw-capped tubes and bottles, rather than plugs or snap caps





Pouring infectious material

- Perform work over plastic-backed absorbent pads
- Wipe rim of the tube with disinfectant-soaked absorbent paper to remove outer contamination

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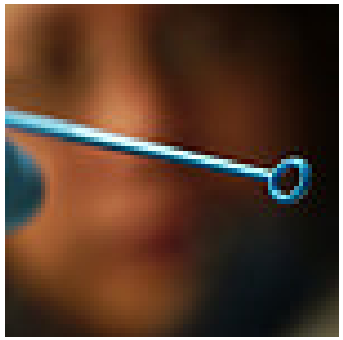
Procedures to Minimize Potential of Exposure

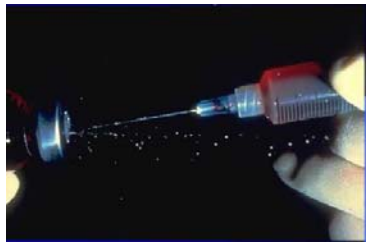


Inoculating loops

- Use a disposable loop instead of a bunsen burner
  - Allow the loop to cool before any procedures
- Use shorter loops to minimize excessive vibrations

Syringes/Needles

- Avoid use whenever possible – use blunt needles instead of sharp needles when applicable
- Withdraw needles from bottles using disinfectant-soaked absorbent pads wrapped around bottle cap
- Fill syringes carefully; use locking syringes if available
- Use a BSC for all operations with infectious material (point needle away from you)



Do not bend, shear or recap needles

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Safe Handling of Sharps/Needles

- **Do not remove cap from needle until ready to inject**
- **Fill syringe carefully**
  - minimize or avoid air bubbles and frothing of the inoculum
- **Take care not to contaminate the hub of the needle**
  - may result in the transfer of infectious material to the fingers.
- **Do not flick syringes containing biohazardous materials**
  - Expel excess air, liquid and bubbles from the syringe vertically into a absorbent material – wool or cotton (if possible, moistened with an appropriate disinfectant)
  - Discard absorbent material appropriately
- **Discard syringes immediately into a sharps container**
  - No need to detach needle from plastic tube
  - DO NOT bend, shear, recap the needle

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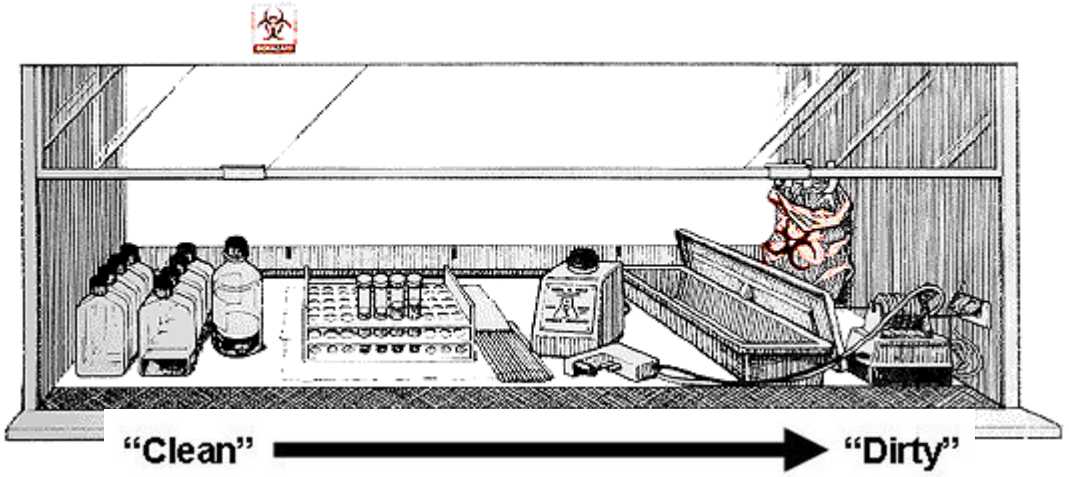
Aseptic Technique

Aseptic technique is a set of specific practices and procedures performed under carefully controlled conditions with the goal of minimizing contamination by pathogens



Video http://www.youtube.com/watch?v=4mKhULnxqcw

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Equipment inside a Lab

Use it
&
maintain it
properly -
your life
depends on it !

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Safe use of Centrifuges

- **Before use**
  - ✓ Check centrifuge rotors & tubes for cracks
  - ✓ Avoid Overfilling
  - ✓ Place caps or stoppers properly
  - ✓ **Balance loads**
  - ✓ Use sealed buckets (safety cups) or sealed rotors
- **Before stepping away:** ensure centrifuge achieves run conditions
- **After run**
  - ✓ Centrifuge has to be completely stopped before opening the lid
  - ✓ Check for spills or leaks **before** removing samples.
  - ✓ Allow aerosols to settle and open samples inside a BSC

- **Maintenance**
  - ✓ Regular cleaning/decontamination
  - ✓ Maintenance/use log

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Cryostats, N_2 Storage vessels, - 80°C Freezers

- Wear gloves during preparation of frozen sections
  - **Freezing tissues does not necessarily inactivate infectious agents**
  - Wear safety shields and heavy gloves when accessing/handling vials from liquid nitrogen
  - Decontaminate frequently

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Biological Safety Cabinets

- **It is <u>the most</u> important safety device for working with biological material.**
  - **Protects the product** (sterile working area)
  - **Protects the operator** (from biological agents using directional airflows)
  - **Protects the environment** (HEPA filters)

- **Provides effective primary containment of biological agents**

Biological Safety Cabinets

Protection is provided through:

- **Creating a sterile working area** - all air within the working area is filtered through HEPA filters
- **A continuous stream of inward air** (inflow), preventing aerosolized particles from escaping through the front opening - protects the operator from exposure
- **Exhaust air is filtered** through HEPA filters, which filter particles that are up to 0.3µm with 99.97% efficiency

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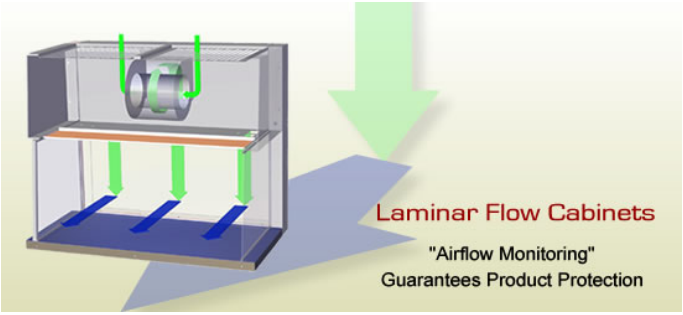
Differences between Laminar Flow and Biological Safety Cabinet

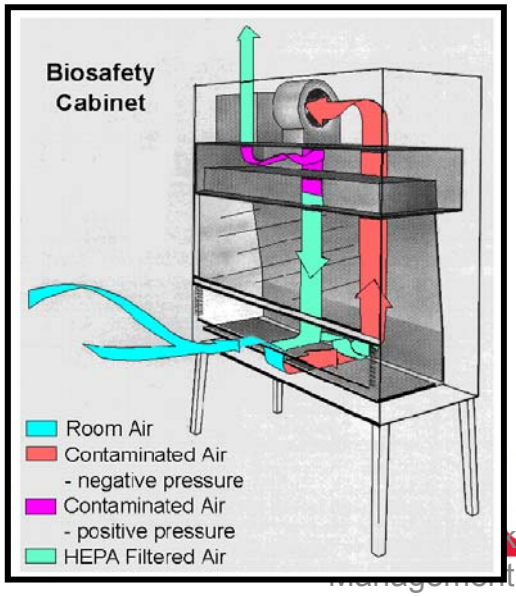
Laminar Flow Hood

- Product protection only
- Potentially infectious aerosols can lead to exposure of the operator and the environment
- Aerosols exposure by blowing air out, into the room:
  - Horizontal-flow
  - Vertical-flow



Biological Safety Cabinet

- Personnel, product and environment protection
- Potential aerosols can be contained within the BSC
- Aerosol exposure is minimal, if BSC is used properly
- Air circulated through HEPA filters



Laminar Flow Hoods and Biological Safety Cabinets

The effectiveness of a LFH/BSC is dependent upon:

- The integrity of the cabinet (certified to national/international standards after installation, any move, annually)
- Proper use practices within the cabinet
    - Prevent turbulence or breaches in the air curtain
- Placement of the cabinet in a room

Video http://www.youtube.com/watch?v=Wg61LdngWlQ

LFHs/BSCs **do not** protect the user from chemical fumes or vapours

- Must use a chemical fume hood for working with chemicals

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Proper Use of Biological Safety Cabinets

Video: Proper use of Biological Safety Cabinets -

http://vimeo.com/7642083

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Hand Hygiene

The act of removing or destroying microorganisms on the hands while maintaining good hand integrity (keeping the skin healthy)

Handwashing is an essential component to reducing the risk of LAIs.

- Before starting any manipulations
- Before leaving the lab
- Whenever the integrity of your gloves is questioned or your hands are obviously soiled
- Before and after completing any task in a BSC
- Every time gloves are removed
- Before contact with face or mouth
- At the end of the day

Proper Handwashing Technique

- Wet your hands with warm running water
- Add soap, and then rub your hands together, making a soapy lather
  - Do this away from the running water for at least 15 seconds, being careful not to wash the lather away.
- Wash the front and back of your hands, as well as between your fingers and under your nails
- Rinse your hands well under warm running water
- Pat hands dry completely with a paper towel.
- Turn off water using same paper towel and dispose in a proper receptacle

Antiseptic handwashing solutions are required in a BCL-2 laboratory

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Proper Handwashing

1. Wet hands

2. Use liquid soap

3. Lather, rub and count to 20

4. Rinse

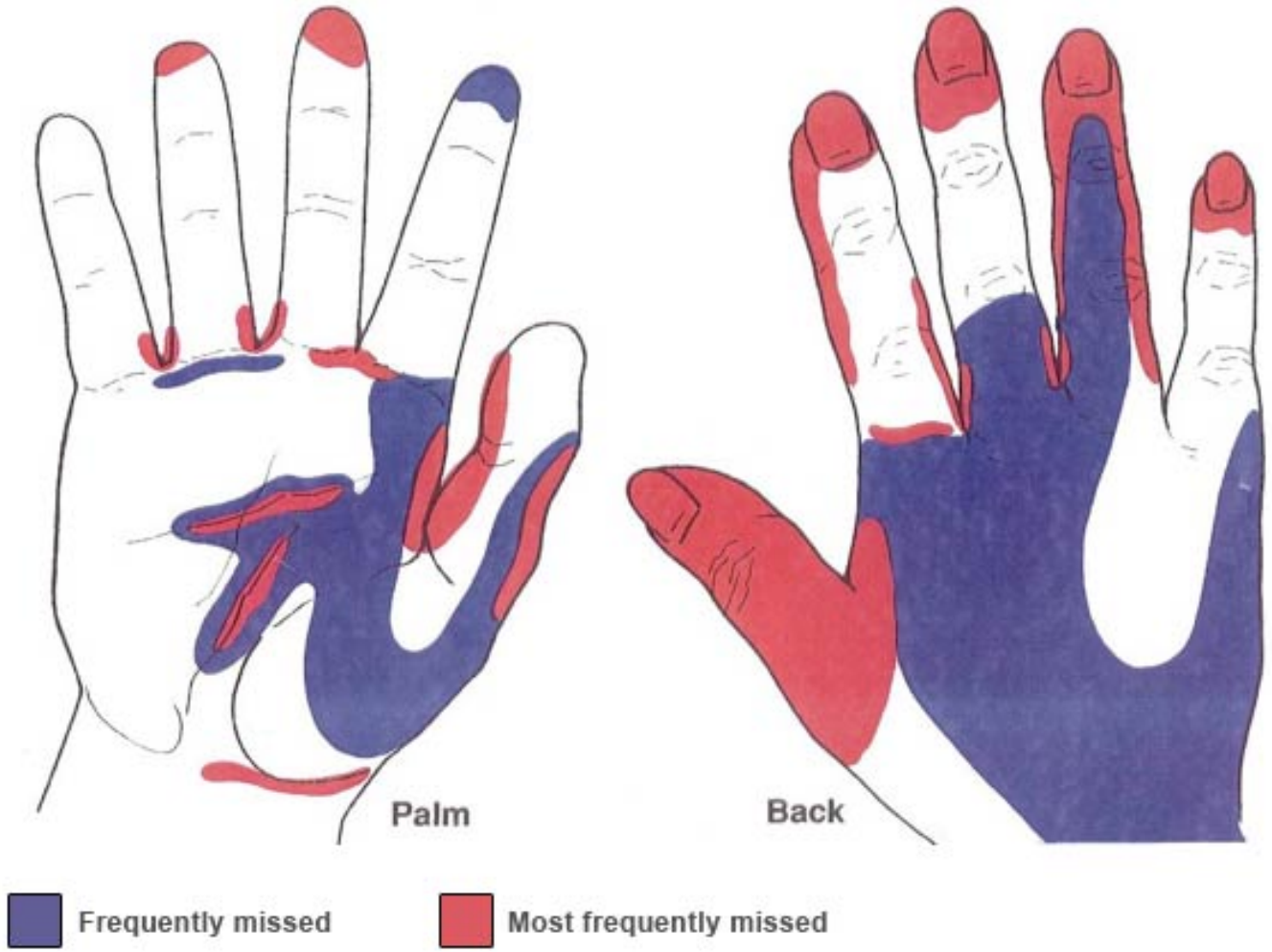
5. Towel or air dry hands

6. Turn off taps with towel or your sleeve

Adapted with permission from Ottawa Public Health, City of Ottawa.

Canada

Areas missed during hand-washing



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Module 7:

Biological Waste Management

Biomedical Waste

Discarded biological material from teaching, clinical and research laboratories and operations is considered as **biomedical waste**

Biomedical Waste can include:

- **Microbiological Waste** *(only stream we can 'disinfect' at York)*
- Human blood and anatomical waste
- Animal blood and anatomical waste
- Sharps Waste
- Cytotoxic Waste
- Waste in contact with human blood waste that is infected or suspected of being infected with any infectious substance (human), or
- Waste containing or derived from one or more wastes described above

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Biomedical Waste

- Treatment of biological waste depends upon the characteristics of the waste (i.e., solid, liquid, mixed) and the associated risk of the biological agent(s)

- The means of treatment for decontamination may involve chemical agents (bleach, ethanol), or physical means (autoclaving, incineration)

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Biomedical Waste

- Diligence must be applied to ensure your own personal safety and that of others, as once treated this material will be directed into the public domain

- Consequently, there are many agencies which are concerned and regulate or set requirements regarding biological waste:
  - City of Toronto
  - Health Canada
  - Canadian Food Inspection Agency
  - Ontario Ministry of the Environment
  - Ministry of Transportation

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Terminology

There are three terms that are often referenced when discussing waste and it is important to understand the difference between them:

Decontamination:

- The process of removing and/or inactivating infectious material/toxins/microorganisms to a lower level such that it removes danger of infection to individuals. This can be achieved by:

  - **Disinfection:**

    Process that eliminates most forms of living microorganisms; can be by the use of agents (physical or chemical) to eliminate harmful microorganisms on inanimate objects (applies to most bacteria and viruses, not to spores)

    Sterilization:

  - The complete destruction of all viable microorganisms (including bacterial spores)

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Chemical Disinfection

- Generally for **disinfection** rather than **sterilization**
- Most common are chlorine compounds and alcohols (broad range)
  - Choice depends on:
    - ✓ Type of material to be disinfected
    - ✓ Organic load
    - ✓ Chemical characteristics

Contact time is crucial

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Preparation of Biomedical Waste

All biological waste should be decontaminated prior to disposal

This includes, but is not limited to:

- **Solid waste** – gloves, pipettes, gowns, tubes, tips, flasks, petri dishes, agar, etc.
- **Liquid** – cell culture media, bacterial cultures, reagents, samples
- **Anatomical waste**:
  - Animal waste – carcasses, faeces, food
  - Human anatomical waste – organs, tissues
- **Human blood and products**, body fluids
- **Sharps**

Preparation of Biological Waste

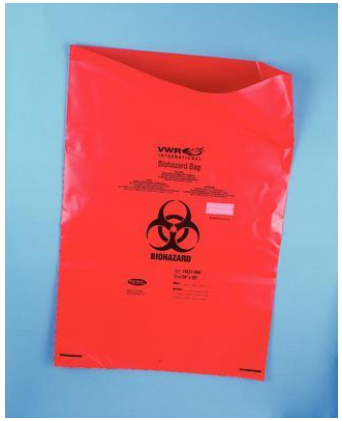
- There are procedures in place to ensure waste is treated appropriately

- **You can take the Hands-On Autoclave Training to be able to safely use autoclaves to decontaminate lab-generated biowaste**.

- **All** containers for biomedical waste must display the biohazard symbol and the words 'Biohazard' in a colour contrasting the container.

Preparing Lab Biohazardous Waste

1. Separate waste
    - Re-usable items (e.g. glass pipettes) from that which will be disposed
    - Dry from liquid material



2. Use only approved autoclave bags (red/orange)
3. Add absorbent material around tube/flask if liquid exists
4. If outside of bag is contaminated, then double bag and well-sealed
5. Do not overfill autoclave bags – ¾ full

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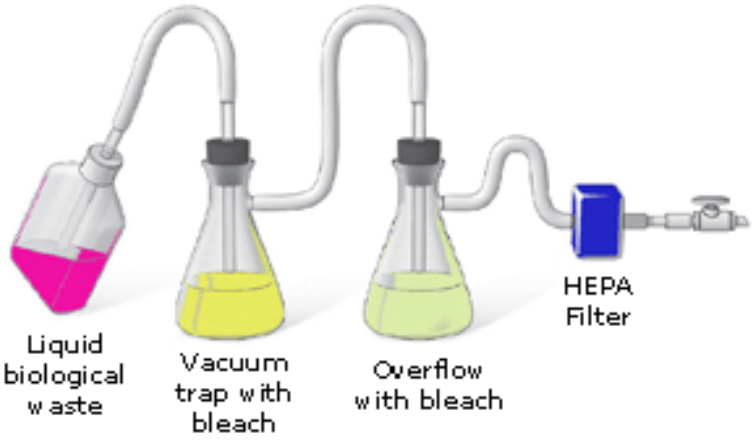
Liquid Waste

- Containers should be leak-proof
- Should contain maximum half its volume of liquid waste
- All liquid for autoclaving should be capped with aluminum foil and marked with autoclave tape
- Undiluted bleach (or appropriate disinfectant) should comprise 10% of final waste volume ***
- Waste should be discarded/treated on a **weekly** basis (in compliance with sewer by-laws)

***** Note: Bleach <u>cannot</u> be autoclaved, so plan your decontamination and waste preparation accordingly**

Liquid Aspiration

- Aspiration through vacuum-traps should be serially set-up with 10% volume of bleach in **both** containers
  - In-line HEPA filters should be used to avoid contamination of vacuum lines
  - Vacuum-grade flasks should be fixed and be used inside secondary containers
  - Secondary containers should be high to contain the liquid in the case of a spill



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Liquid Aspiration

- Both flasks have to be vacuum grade
- Secondary container and clamped
- Common labs - responsibility
- Degradation of bleach – smaller volumes
- Date when it was made and who made it

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Preparation of Biological Waste

Anatomical waste

- Placed in a red biohazard bag
  - Other garbage bags can be used if the waste will immediately be stored in a barrel lined with red biohazard bags.

- Anatomical waste is discarded in Animal Care carcass disposal fridge

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Infected Sharps waste

Include items that are capable of causing punctures or cuts (e.g. scalpels, broken glass, pipettes, test tubes, microscope slides, blood vials or any other material **that has come in contact with biohazardous material**

- Must be placed in a rigid, leak proof, puncture resistant and sealable container
- University currently uses 4.5L yellow containers
- These are available at Science Stores
- Sharps waste is **NOT** treated at York University

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Autoclaving

Autoclaving uses high pressure and steam to **sterilize** biomedical waste

Proper sterilization depends on various factors:

- Nature of waste (solid/liquid)
- Material load size/volume
- Moisture versus dry load cycle
- Sterilization time

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Autoclaving

Autoclaves are run by dedicated staff, however, if **you** are operating an autoclave:

- Learn how to use it
- Ensure proper PPE is worn
- Recognize and prepare acceptable material and packaging
- Proper loading and unloading

All users/operators must take the Hands-on Autoclave Training

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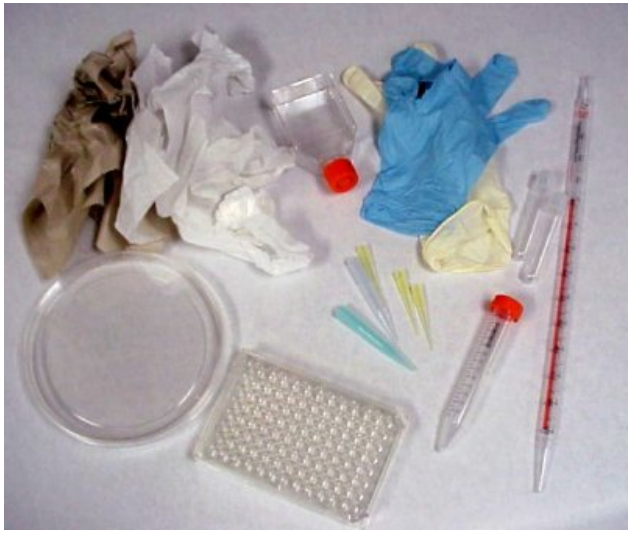
Items that **CAN** be autoclaved

- Culture dishes, plates and related lab supplies
- Cultures and stocks of infectious material
- Discarded live and attenuated vaccines
- Contaminated solid items (petri dishes, eppendorf tubes, tips, pipettes, gloves, paper towels)



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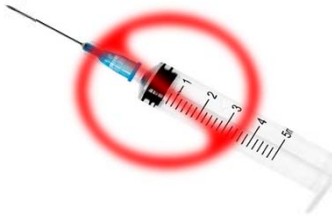
Items that **CANNOT** be autoclaved

- Chemicals (flammables, oxidizers, phenols, acids, alkalides)
- Chemotherapeutic or radioactive waste
- Bleach (or other chlorinated products)
- Certain kinds of plastics
- Sharps



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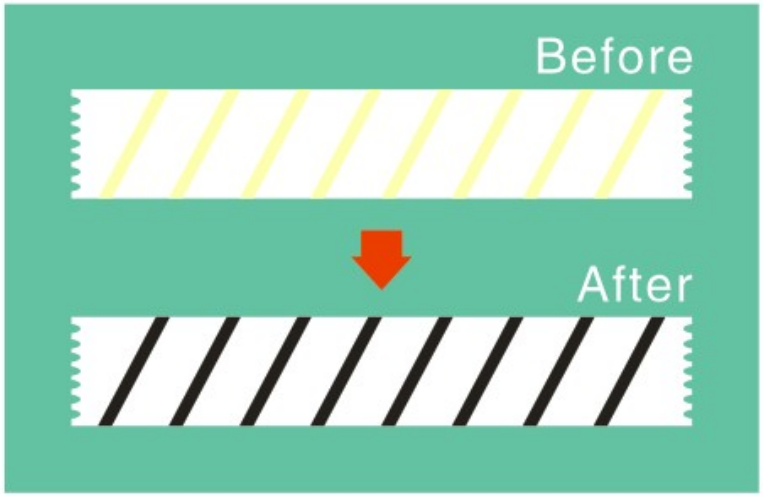
Treated/Untreated Waste

- Treated waste is no longer considered 'biomedical' (i.e. microbiological waste, blood and bodily fluid waste) and can be disposed of in the regular waste stream.



- Any waste that cannot be treated (i.e. sharps, carcasses, tissues and body parts) remains biomedical waste and must be treated off-site.

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Module 8:

Emergency Response

(spills & accidental releases)

Planning your experiment – fail to plan, plan to fail

- Risk assessment can foresee and mitigate the chance of an emergency
- Follow pre-planned work procedures (experimental protocols and SOPs)
- Modifications should be reviewed by supervisor
- Be familiar with emergency spill procedures and location of spill stations, absorbents, and disinfectants
- Be familiar with emergency escape routes, locations of fire extinguishers, eye wash stations, safety showers and first-aid kits
- Carefully planning your experiments, following laboratory safety guidelines and understanding emergency procedures could save your life!

In cases of Emergency, contact:
HSEWB Main Extension - Ext. 55491

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Emergency Response

Emergencies can happen. Situations you may encounter:

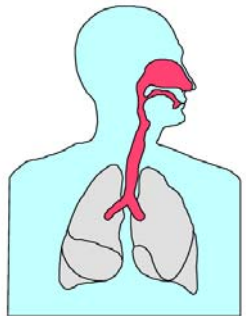
- Splashes/Aerosols generated
- Spills: inside a centrifuge, Biosafety Cabinet

Was anyone exposed?

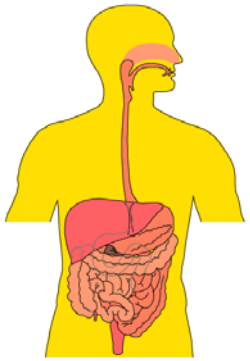
- What is the route of exposure?
- Are aerosols still suspended?
- Is the biological agent contained?

Biological Hazards Routes of Entry

Skin Absorption



Inhalation



Ingestion

Injection

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Emergency Response

- Emergency Response Plans
  - Spills:
    - cell culture, blood, bacteria cultures, virus infected samples, body fluid samples…
  - Sharps/Needlestick exposure:
    - Exploding cryogenic vials, hypodermic needles, scapels, glass slides
  - General Institutional Emergency Plans
    - FSE training manual, York U Security Website

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Spill Procedures

- Spill responses will vary according to what, where, how much and when
- Initiation of spill clean-up should occur immediately to ensure proper decontamination:
    - Biological material
    - Chemicals

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Spill Response

Spill response depends on:

- What is spilled? – characteristics and hazards
- How much is spilled? – volume and concentration
- Where it is spilled? – BSC, lab, centrifuge, outside lab

- Knowing how to properly clean up spills will ensure that you are safe and that your exposure is minimized.
- All spills should be reported to the area Health and Safety Officer

Items inside a spill kit

- Concentrated disinfectant (e.g. bleach or other appropriate disinfectant)
- Spray bottle
- Paper towels or another suitable absorbent
- Biohazard / autoclave bags / appropriate waste container
- Forceps to pick up broken glass
- Sharps container to dispose off broken glass
- PPE: gloves, goggles, lab coat / gown, face shield

Biological Spills

- Contain and cover the spill with absorbent material
- Soak the spill with an appropriate disinfectant (10% Bleach, Virox)
- Work inwards from outside of spill
- Leave on for 20 – 30 minutes
- Mark area so there is no traffic around the spill

Biological Spills inside a BSC

- **Keep the ventilation on**
- Cover the spill area with paper towels or absorbent material
- Soak the spill area with an appropriate disinfectant (10% bleach, Virox)
  - Pour the disinfectant from the outside surface of the absorbent material towards the inside.
- Leave ventilation on for 20 to 30 minutes.
- All items within the cabinet should be disinfected before removal
  - Walls and surfaces wiped down, equipment wiped down and/or autoclaved
- Run ventilation for 10-15 minutes after clean up.
- All waste must be autoclaved (unless bleach was used)
- Report incident

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Biological Spills inside a centrifuge

- Keep closed and allow aerosols to settle for at least 30 minutes.
  - Ensure centrifuge is off / unplugged. If possible move rotors/buckets to a BSC
- Notify others in the lab not to use the centrifuge (include signage)
- Disinfect the centrifuge, rotors and buckets in an appropriate disinfectant
  - Allow at least 20 to 30 minutes of contact time
- Carefully retrieve any broken glass or plastic from inside the centrifuge.
  - Use forceps and place broken glass in a sharps container
- Drain the disinfectant and thoroughly wipe down the inside of centrifuge and all parts including the lid with paper towels soaked in disinfectant
- Rinse the rotors, buckets and the inside of the centrifuge with water
- All waste must be autoclaved – except items that have come in contact with bleach
- Report incident

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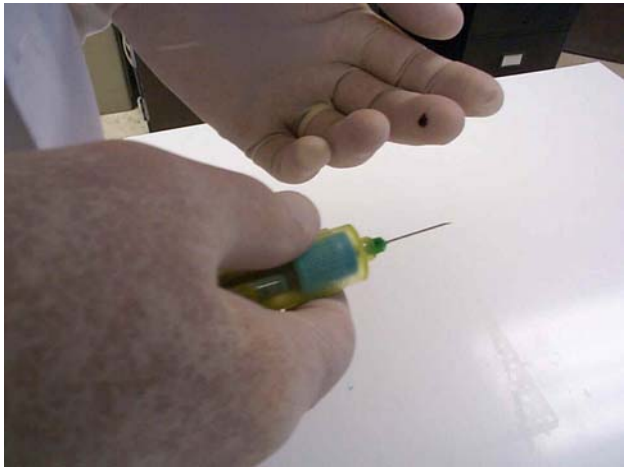
Needle-Stick Injury

- If bleeding, allow blood to flow
- For small punctures, squeeze to encourage blood to flow out of wound
- Wash skin with soap and water
- Apply antiseptic
- Administer First Aid, cover with dressing/band aid
- If known/suspected exposure to human pathogen, seek medical attention immediately
- Notify your Supervisor
  - They will notify:
    - Area Health and Safety Officer
    - YU Biosafety Officer



Emergency Showers and Eyewash Stations

- Know location of emergency showers and eyewash stations
  - Become familiar with their operation. You need to be able to find them and know how to use them with your eyes closed

- Emergency showers should be tested by physical resources
- Eyewash stations should be tested weekly and logged by your

Laboratory Safety Representative

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Aerosols

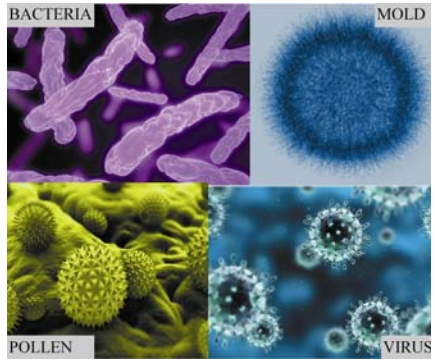
Airborne biological materials can be generated through many laboratory practices

Potential exposure to aerosols can occur in three ways:

- Breathable infectious airborne particles
- Aerosols can settle on surfaces and become an ingestion hazard through contamination of the hands
- Spills or splashes can infect mucous membranes

Precautions should be taken to minimize the production of aerosols.

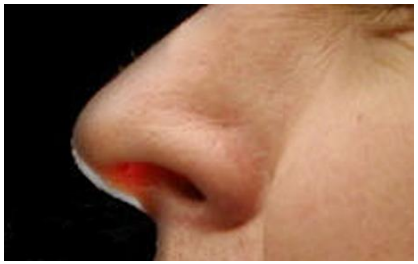
- In order to prevent LAIs through aerosols, all manipulations involving biohazards should be performed in a biological safety cabinet (BSC)
- Decontaminate surfaces and equipment after use



Aerosol Risk

REMEMBER – if the risk was inhalation, there may not be any evidence of an exposure having occurred

- Inform all those in the vicinity
- Restrict access and resuspending or relocation of particles
- Vacate area for 30 minutes before re-entering.
- Report, sign area, seek medical assistance.



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No Excuses!

Reporting

All potential exposures should be reported **immediately** to:

- Your supervisor /PI
- Area Health and Safety Officer
- Security, or
- HSWEB (ext. 55491)

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Accident/Incident Reporting

- **Employees (Faculty, Staff, TAs, postdocs)**
  - Supervisors to complete **S**upervisor **A**ccident **I**nvestigation **R**eport (SAIR)
  - May be contacted by JHSC, EWO, DOHS as part of follow up
  - Health care provided, Loss of time from work (WSIB- Form 7)

- **Students**
  - Student Incident reporting: report to your Supervisor
  - Follow faculty protocol

- **Concerned about exposure to a biohazard?**
  - SEE YOUR DOCTOR
  - CONTACT THE BIOSAFETY OFFICER
    - On Call Occupational Health Doctor

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Module 9:

Internal Processes

&

Documentation

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Material Acquisition

Funding agencies (Tricouncil, CFIA, PHAC, EC) require labs to be compliant to the established federal, provincial, municipal, and international Biosafety programs **before purchasing and receiving of Biological agents**:

- Proper inventory
- Proper adherence to institutional Biosafety program
- Biosafety Permits

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Material Transfer Agreement

Material Transfer Agreements (contracts)

- Between labs - intra-campus
- Between labs - inter-campus
- International Transfers

Inform Biosafety Officer of ALL material shipments and receipts!

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Transportation of Dangerous Goods

Shipping and receiving of:

- Infectious substances
- Diagnostic samples

  - Pre-approved
  - Authorized Individuals
  - Lead time (International Regulations)
  - Appropriate Scheduling (Holidays, Weekends)
  - Transportation within the building
  - Between lab to lab
  - Colleague to Colleague
  - Between Institutions

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Biosafety Permit

Each lab (CL1, CL2 or CL3) handling Biological Agents is required to have a York University Biosafety Permit that details:

- Principal Investigator information
- Active Grants and projects
- Current users in the lab
- Inventory of biological agents used and stored in the lab

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Extra training you may require

- ❑ Transportation of Dangerous Goods Training
- ❑ Radiation Safety Training
- ❑ Animal Care and Veterinary Services Training
- ❑ WHMIS
- ❑ Lab Safety Training
- ❑ Hands-on Autoclave Training

ALL training records for all personnel must be documented in your lab safety binder

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Summary

- Follow general lab rules
- Learn proper lab techniques: get trained!!!
- Keep records of your training and risk assessments
- Know how to properly use lab safety equipment
- Wear PPE appropriate for the work you are doing
- Clean and disinfect work area (right disinfectant, right amount of time)
- Be prepared! Know how to respond in case of an emergency

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THINK SAFE
ACT SAFE
BE SAFE

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York University
Biosafety

http://www.yorku.ca/dohs/prog-biosafety.html

Jay D. Majithia
Biosafety Officer/
Health & Safety Advisor
Tel: 416-736-2100 ext. 44745
E-mail : jmajithi@yorku.ca

Health, Safety and Employee Well-Being
York University
Kinsmen Building
Toronto ON M3J 1P3

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