General Biosafety

Working in a biological laboratory

Martha Adams
madams3@mit.edu
X4-0114

General Biosafety Training – What you will learn.

- Intro to the Biosafety Program
- How you can be exposed to biological agents
- Biosafety levels and corresponding work practices and equipment
- Correct response for an incident involving biological materials

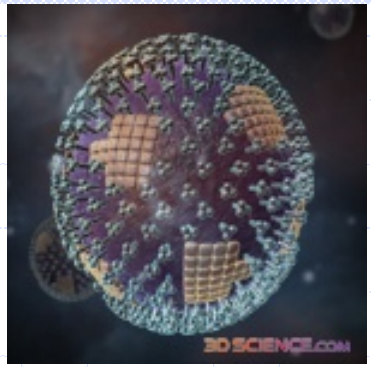
Biosafety Program Services

- Risk assessment and biosafety support
- Registration of biological projects involving:
  - Recombinant DNA
  - Biological Agents
  - Infectious agents
  - Human materials
  - Select agent toxins
- Support to Committee on Assessment of BiohazardsEmbryonic Stem Cell Research Oversight (CAB/ ESCRO)
- Other Institutional Committees
  - Committee on Animal Care (CAC)
  - Committee on the Use of Humans as Experimental Subjects (COUHES)

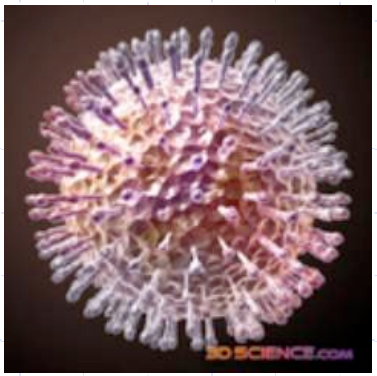
Biosafety Program Services

- Bloodborne Pathogen Program
- Indoor air quality
- Advice on sterilization
- Advice on waste handling, practices, etc.
- Shipping biological materials

What are some risks of work with microorganisms?



- Infections in researcher or others
- Invalidation of the experiment / contamination of lab stocks
- Epidemics in domestic, wild, or agricultural animals

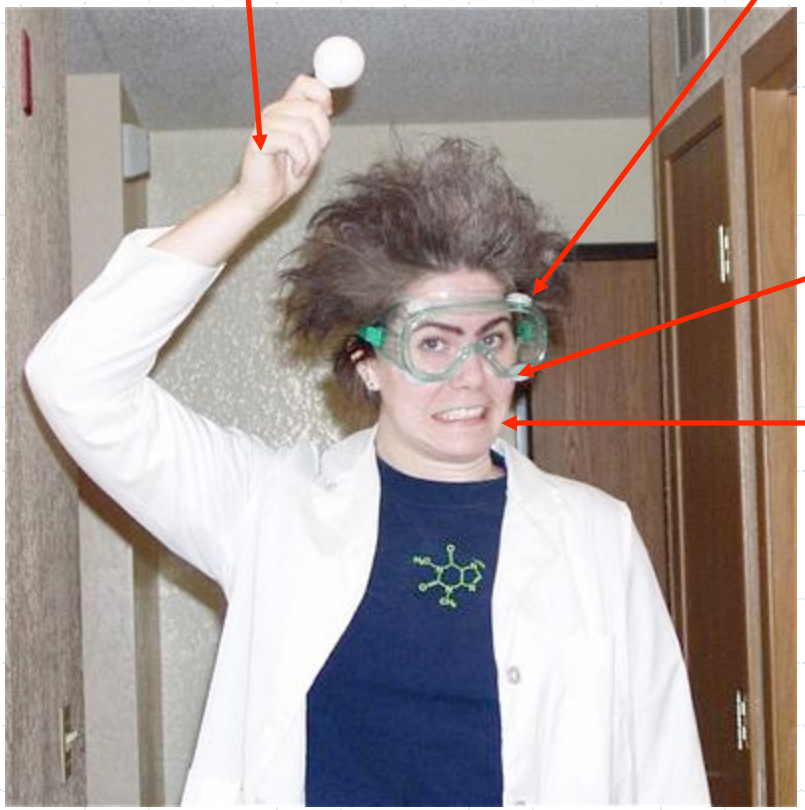


What factors affect how risky it is to work with a biological agent?

- Pathogenicity - health impact
- Infectious dose vs. amount you're using
- Availability of prophylaxis
  - Before exposure
  - After exposure but before infection
- Your health

These and other factors go into choice of biosafety level for research

How can biological materials get into your body when you work with them?



What's a Biosafety Level?

- A combination of
  - lab practice / technique
  - safety equipment
  - facility design
- Based on concept of "containment"
- For protection of
  - personnel
  - lab environment
  - environment outside lab

BL1

Certain experiments require BL1 physical containment and must follow prescribed laboratory practices.

BL1 and above

Biosafety Level 1

Suitable for work involving well-characterized agents not known to cause disease in healthy adult humans and of minimal potential hazard to laboratory personnel and the environment.

- *Bacillus subtilis, E. coli K-12, Saccharomyces cerevisiae*
- *Rodent cells and cell lines*

BL1 and above

Standard Microbiological Practices

- Restrict or limit access when working
- Wash hands
- Use mechanical pipetting devices
- Prohibit eating, drinking or applying cosmetics, i.e. chap stick

BL1 and above

Standard Microbiological Practices

- Open containers away from face.
- Avoid contaminating the outside of the container.
- Disinfect the outside of container before work and before returning it to storage

What sorts of lab activities can make aerosols?

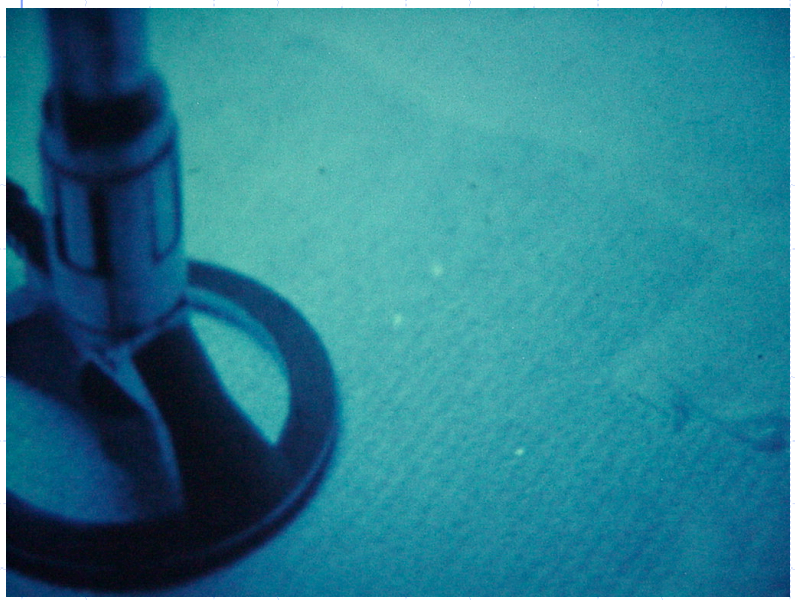
BL1 and above

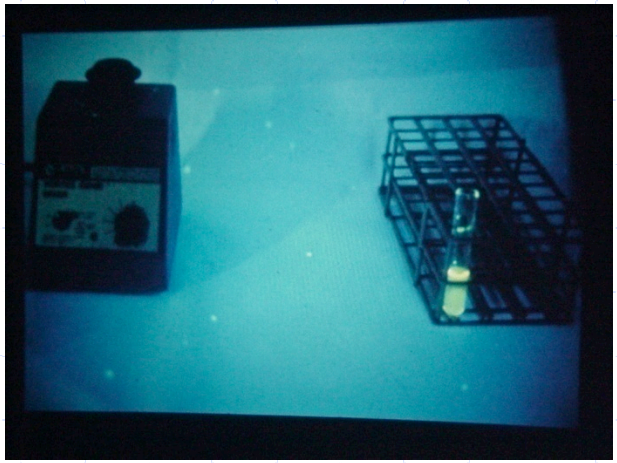
- Pipetting
- Centrifuging
- Grinding
- Blending / Mixing
- Shaking
- Sonicating
- Opening containers
- Inoculating animals intranasally
- Harvesting infected tissues from animals

BL1 and above

Standard Microbiological Practices

- Decontaminate work surfaces daily and after spills
- Minimize splashes and aerosols





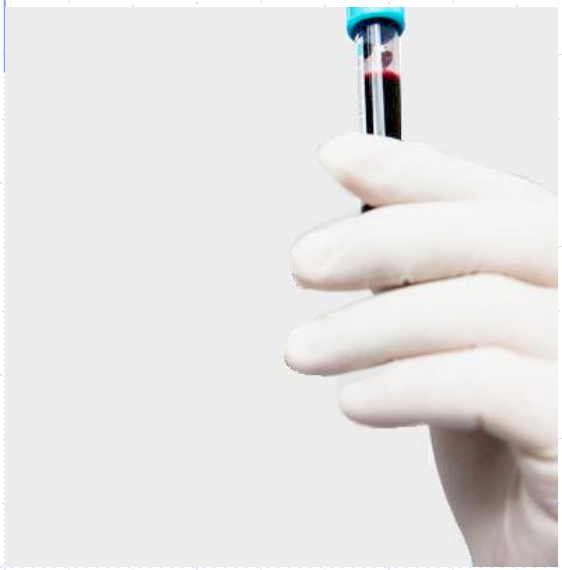
BL1 and above

What to wear

- Lab coat
- Gloves

Your street clothes are part of your PPE!! Wear appropriate clothing while working in a laboratory.

Don’t wear gloves in public areas



Use the one glove technique

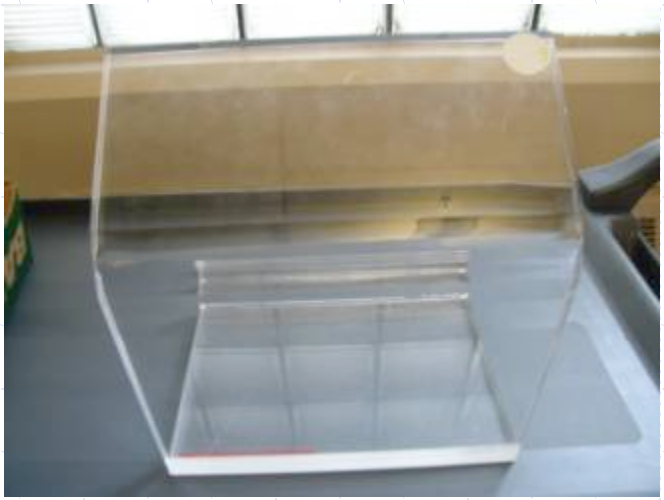
Or

Carry your samples in a secondary container

BL1 and above

Personal Protective Equipment (PPE)

- Additional PPE may be needed
  - Face protection
  - Eye protection
  - Alternatives?



BL1 and above

Signs and Labels

- BL signs at lab entrances
- BL practices signs within labs
- Biohazard stickers on potentially contaminated items such as:
  - Refrigerators/Freezers
  - Incubators
  - Centrifuges
  - Storage Containers
  - Waste Containers

BL1 and above

Biohazardous Waste Disposal

Autoclave Solid Waste

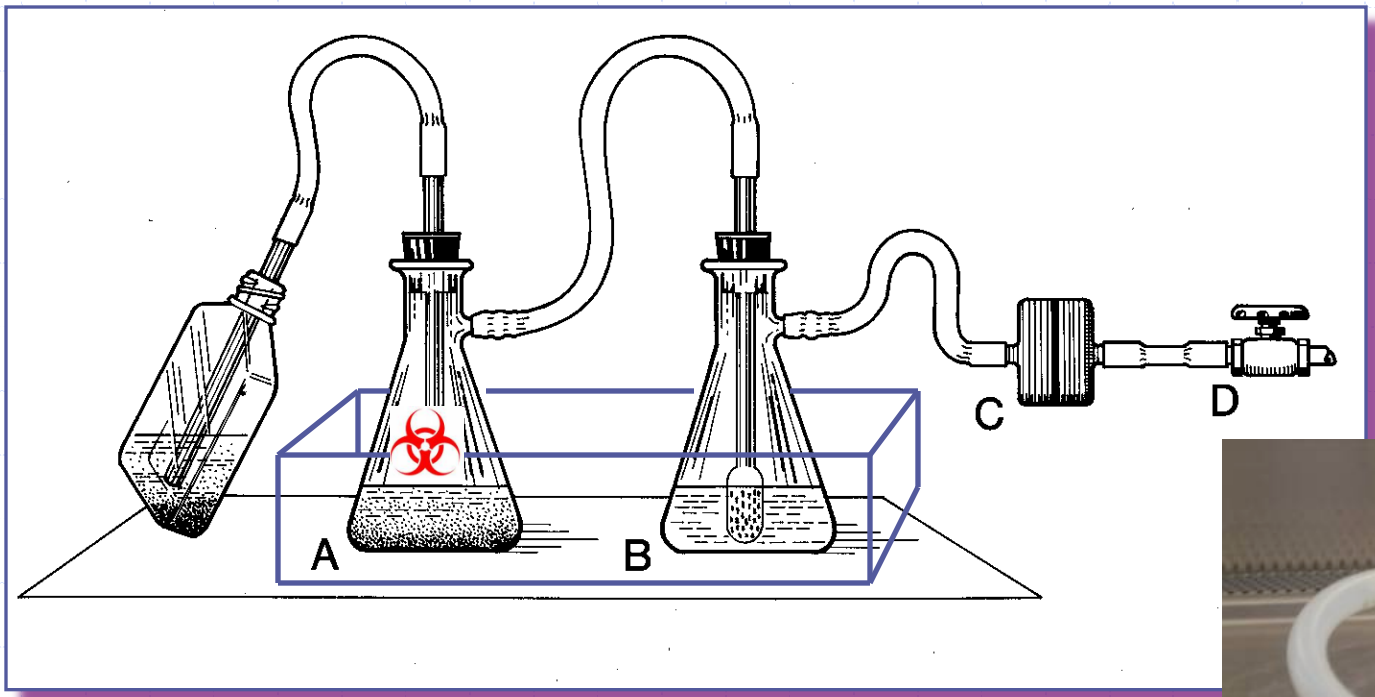
- Place in clear bag in marked bin
- When full, place autoclave tag on bag
- Complete autoclave log book
- Leave bag OPEN for steam penetration
- 121°C, 15 psi, 60 minutes
- PPE: safety glasses, long sleeves and gloves rated for hot items
- Place treated/tagged bags into normal waste bin

Biohazardous Waste Disposal

- Liquid Waste
  - Use chemical disinfectant, such as bleach (1:10) or Wescodyne (1:100)
  - Let sit for twenty minutes
  - Pour down the drain

BL1 and above

Vacuum Line Protection

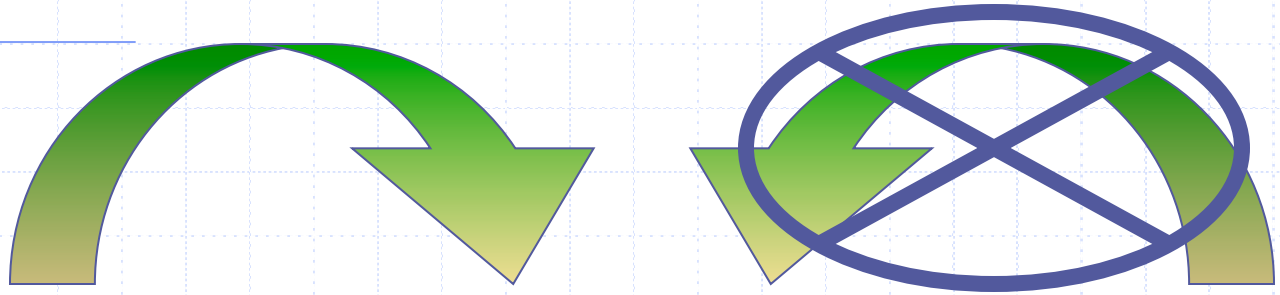


A = primary collection flask with disinfectant
B = overflow flask with disinfectant
C = hydrophobic or HEPA filter
D = to vacuum pump
Bin = secondary containment for flasks

MIT Bio Lab Sharps Disposal

Containers must be sealed and placed in the hallway the night before or morning of pickup.
Containers should be **at least 2/3 full**. Pickup for most areas is Thursday.

YES



NO

Needles & syringes
Razor blades and scalpels
Toothpicks
Pasteur pipettes
Serological pipettes
Pipette tips
Glass vials, slides
Broken glass
Wire
Anything else that can possibly puncture or cut the skin (contaminated or not)

No chemicals, reagents, solutions, or volatile substances
No radioactivity

No liquids

No gloves
No paper towels, kimwipes, wrappers, paper, cardboard, etc
No plastic bottles, plastic tubing, plates, wells and other non-sharp plastic containers*

Please report problems or direct questions to EHS at 2-3477.

** Note: Round bottom falcon tubes, petri dishes, microtiter plates and other non-sharp plastics should be collected with solid lab waste and autoclaved*

BL1 and above

Exercise caution with Sharps

BL2

Experiments at the BL2 physical containment level require prescribed laboratory practices and containment equipment.

BL2 and above

Biosafety Level 2

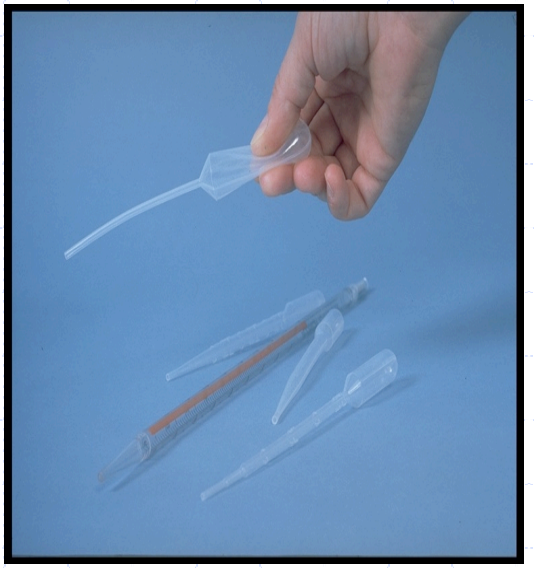
For work involving agents of moderate potential hazard to personnel and the environment.

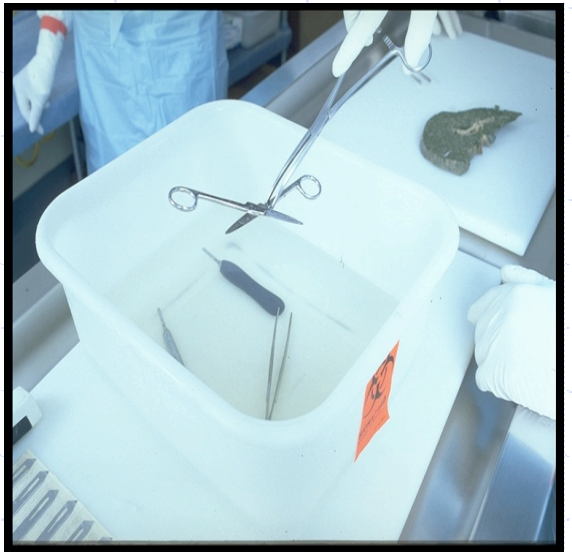
- Work with infectious/pathogenic agents
- Work with human blood, tissue and cell culture.
- Work with non-human primate materials
- Other examples: Hepatitis B virus, *Salmonella typhimurium, Staphylococcus aureus, Streptococcal spp.*

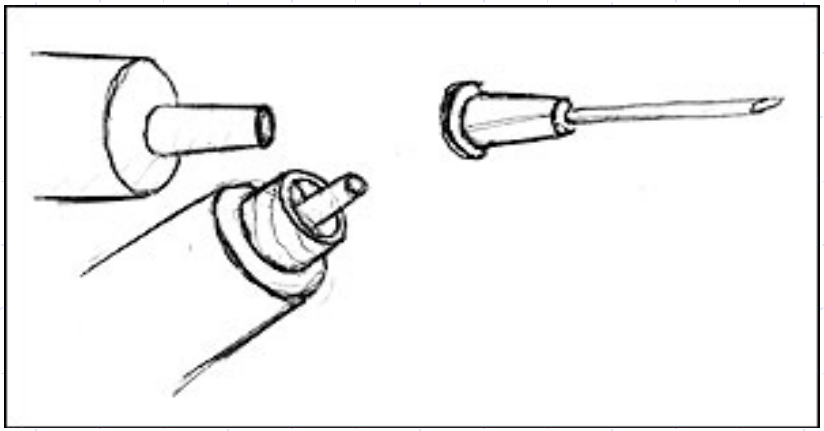
BL2 and above

BL2 Work Practices

- Standard Microbiological Work Practices
- Access to lab is more restricted
- Extra precautions for handling sharps
- Only animals and plants involved in research permitted in lab







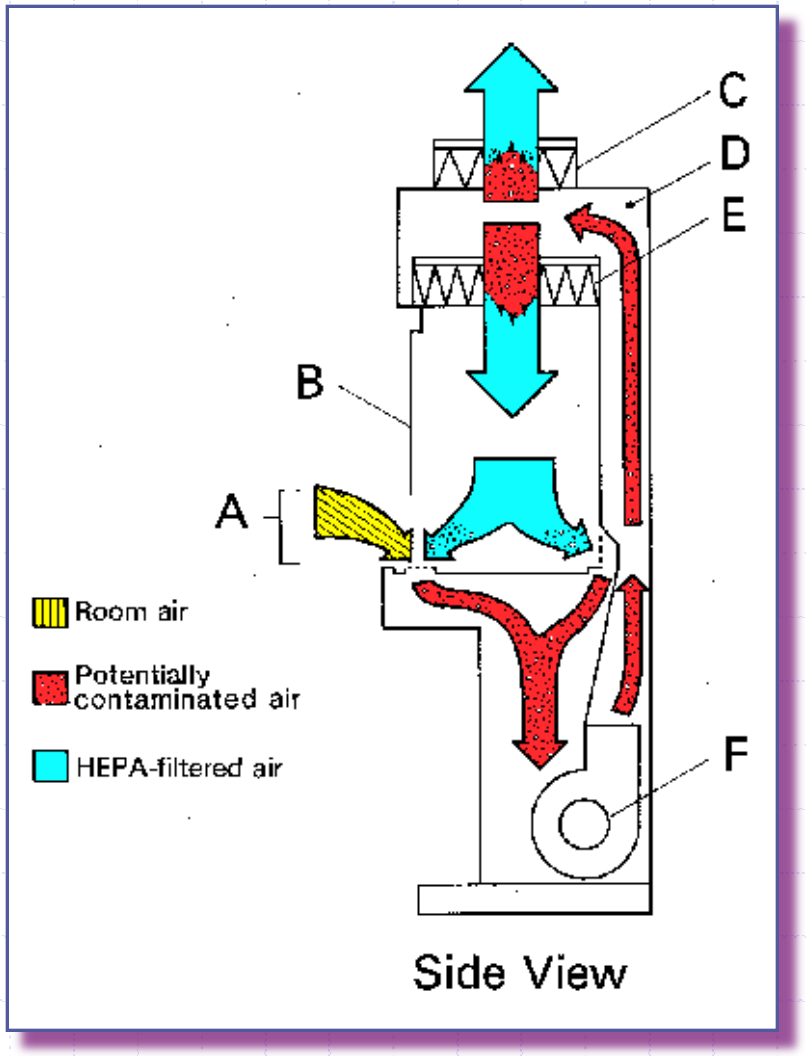
BL2 and above

Engineering Controls

- Avoid bunsens, use
  - Bacticinerator
  - Wrist operated touch-o-matic
  - Disposable loops
- Safety Centrifuges
- BSCs, Fume Hood

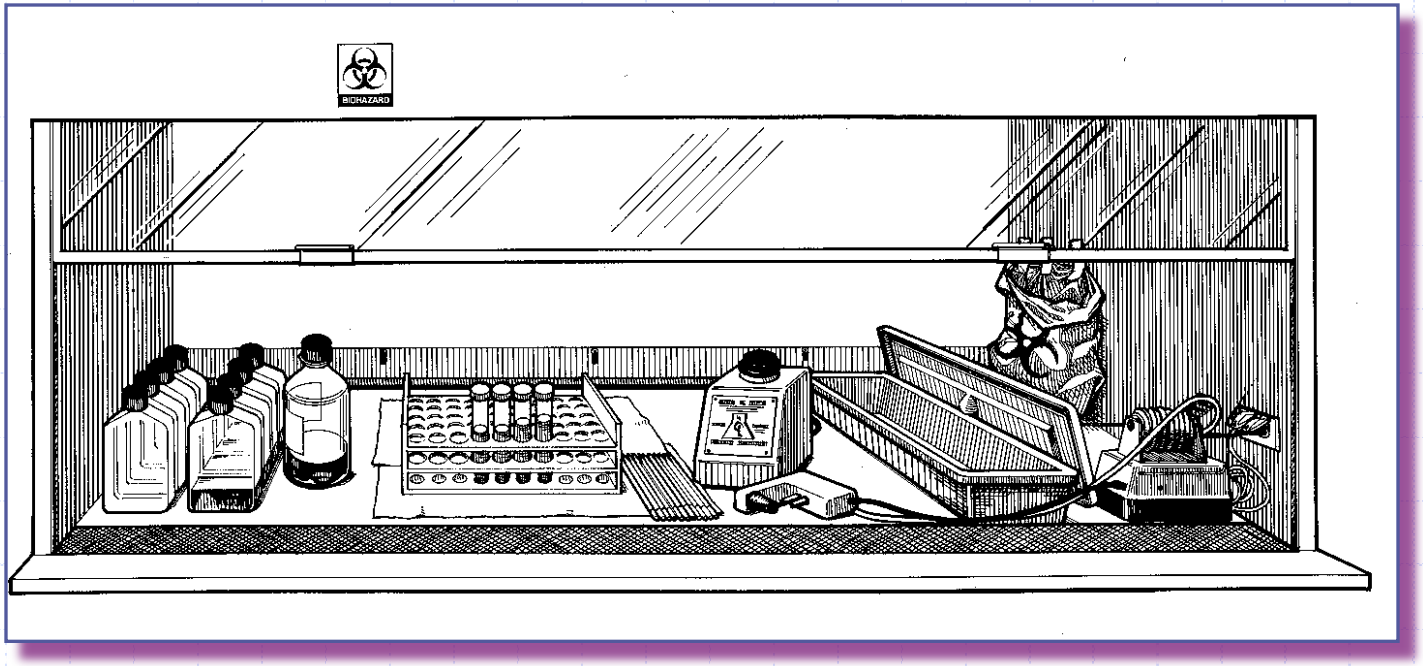
Engineering Controls: Class II Biosafety Cabinet

- Cabinet purge before and after work.
- Wipe down cabinet with 70% ethanol before and after use.
- Minimize hand arm motion and take other measures to reduce disruption of airflow.
- No flammable chemicals.
- Keep glove-sleeve gap closed
- UV lights with caution



Class II Biosafety Cabinet Use

- Layout of Equipment: clean to dirty



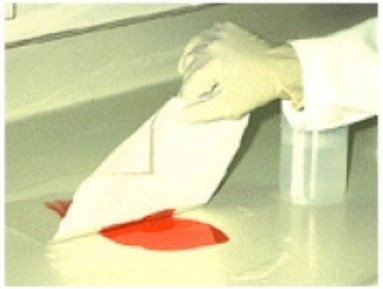
Emergencies - getting help

- Emergency numbers on each phone -- do you know them?
- 100
- 617-253-1212
- Emergency response guide (looks like a flip chart) posted in labs

What to do if you get exposed to a biological material

- If Direct contact to your face or skin
  - Immediately, wash the area with soap & water for 10-15 minutes
  - If it's in your eyes, nose or mouth, flush with water for 15 minutes
  - Tell your supervisor immediately
  - Go to the Medical Department (E23) immediately
- If stuck by a needle or sharp
  - Wash the area with soap and water
  - Tell your supervisor immediately
  - Go to the Medical Department (E23) immediately

How to clean up a spill



- Cover the spill with absorbent or paper towels.

- Disinfect the spill: Circle with disinfectant, saturate, let sit for 20 mins

- Clean up the spill and dispose of spill materials as biowaste
  - Any sharps?
    - Broken glass or sharps, use forceps or a dustpan and broom.
    - Everything goes into sharps container.

  - Otherwise, use autoclave bag
- Report spill to PI
- Clean / discard contaminated clothing

What's right and wrong?

The End

QUESTIONS??

- Biosafety Program
- How you can be exposed
- Biosafety levels - work practices and equipment
- Correct response for emergencies

References

- Guidelines/Regulations
  - Biosafety for Microbiological and Biomedical Laboratories, 5th edition
    - CDC (Center for Disease Control)
    - NIH (National Institute of Health)
    - http://www.cdc.gov/od/ohs/biosfty/bmbl5/bmbl5toc.htm
  - NIH Guidelines for Research Involving Recombinant DNA Molecules, 2002.
    - http://www4.od.nih.gov/oba/rac/guidelines/guidelines.html
  - MA State Biological Waste Regulations.