BIOSAFETY LEVELS

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Principles of Bio-safety

1. Laboratory Practice and Procedure -

   (Guidelines for working safely in microbiological and biomedical laboratories)

   - **Standard Practices**
   - **Special Practices**

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2. **Bio-safety levels (BSLs) -**

   (Increasing levels of personnel and environmental protection)

   - **Safety equipment** *(Primary barriers)*
   - **Facility design and construction**

     (Secondary barriers)

Principles of Bio-safety

Bio-safety levels (BSL):

- **BSL1** - agents known not to cause any disease
- **BSL2** - Agents associated with disease
- **BSL3** - Indigenous/exotic agents associated with human disease and with potential for aerosol transmission
- **BSL4** - dangerous/exotic agents of life threatening nature

BSL4 Laboratory specifications

	Directional Airflow	Double Door Entry	Autoclave Available	Pass-through Autoclave	Seamless Floors	Monolithic Ceilings	HEPA Filtered Exhaust	HEPA Filtered Supply	supply/exhaust interlock	Personnel Shower	Airlock Entry	Pressure Differential	HEPA Plumbing Vents	Effluent Decontamination	Pressure Decay Testing	Breathing Air System				
BSL-2 Laboratory	●		●																	
BSL-3 Laboratory	●	●		●	●	●														
BSL-3 Laboratory – Q Fever	●	●		●	●	●	●			●										
BSL-3 Animal Facility	●	●		●	●	●	●			●										
BSL-3 AG Lab & Animal	●	●		●	●	●	●	●	●	●	●	●	●	●	●					
BSL-4 Lab & Animal	●	●		●	●	●	●	●	●	●	●	●	●	●	●	●				

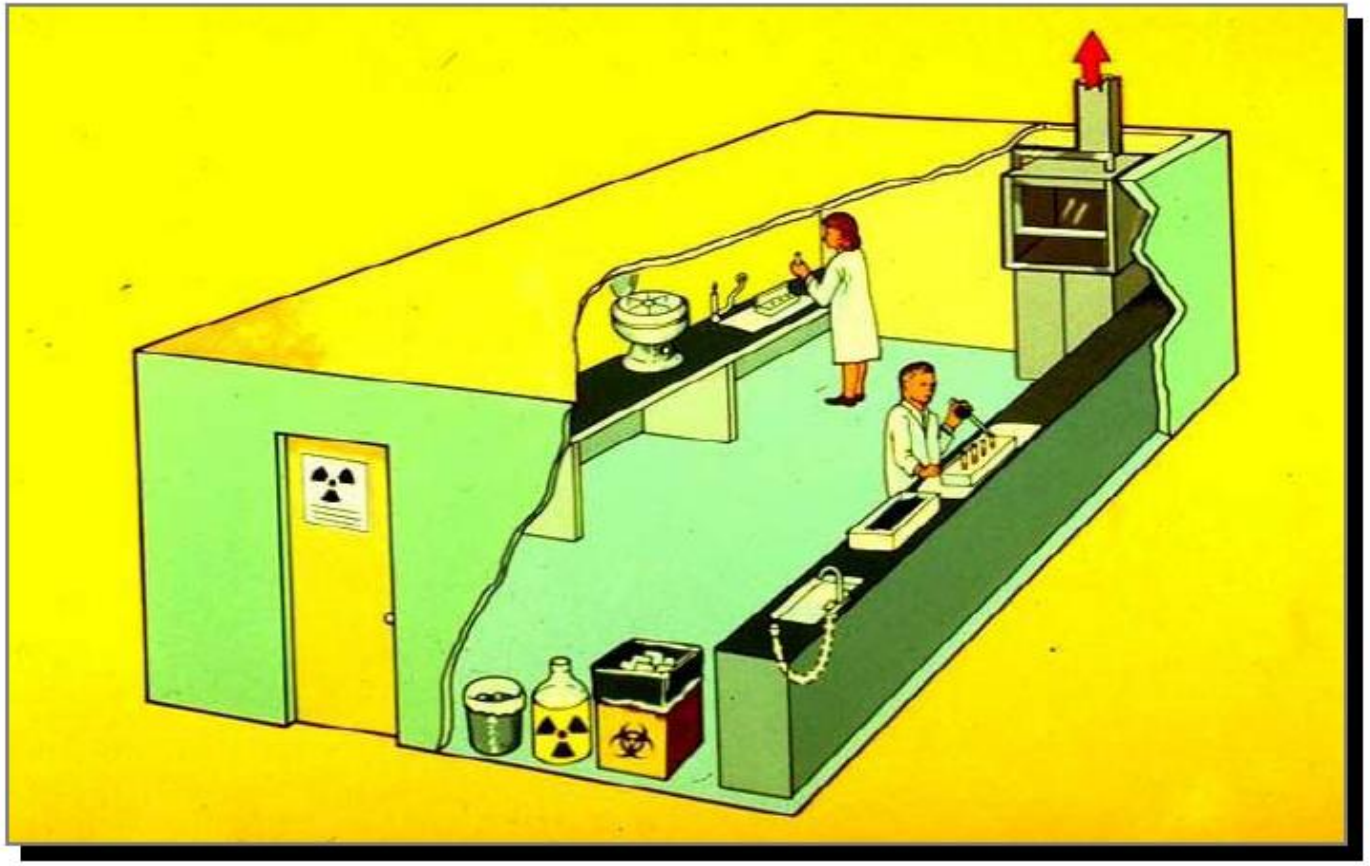
Bio-safety Level 1 (BSL1)

- For microorganisms that don't consistently cause disease in healthy adults

  - *E. coli (K12 & its derivatives)*, *S. cerevisiae*,
  - Basic laboratory
  - Standard Microbiological Practices

BSL1 Laboratory

(Secondary Barriers)



Bio-safety Level 2 (BSL2)

- For microorganisms of moderate potential hazard, transmitted by contact, ingestion, puncture

  - *Salmonella sp.*, herpesvirus, hepatitis B virus, measles virus, toxoplasma, human blood
  - Basic laboratory
  - Standard Practices PLUS

… BSL2

- Standard Microbiological Practices

Plus:

- Training in handling pathogens
- Access to lab limited
- Extreme sharps precautions
- Use of Bio-safety cabinets (BSC) for aerosols

Biosafety Cabinets (BSCs)

Safety Equipment (Primary Barriers)

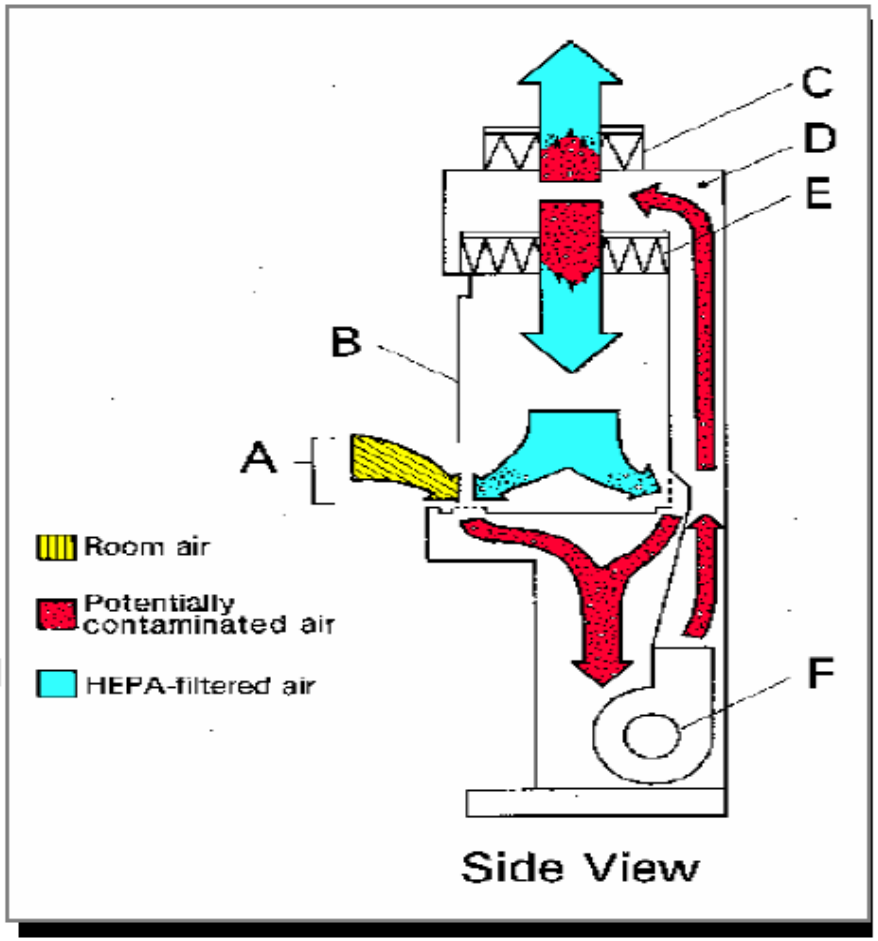
Bio-safety Cabinet (BSC)

Airflow

- -Pressure (120 Pa)
- 99.97% removal of 0.3µm particulates

Use of Biosafety Cabinet

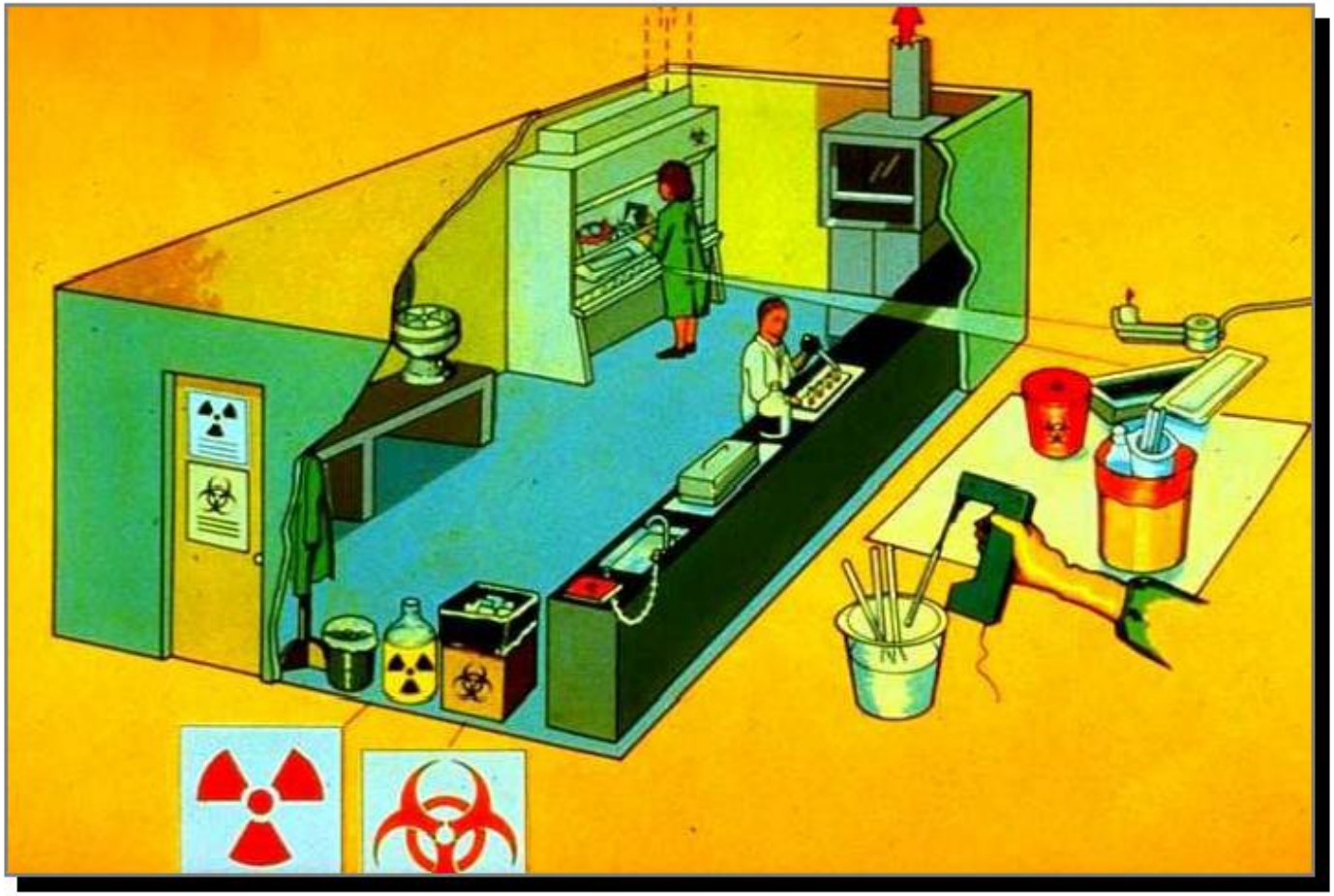
- Turn on fan 15 min before starting
- Don't block grille
- Disinfect work surface with 70% ethanol
- Discard pipets in autoclavable bags
- Minimize movement of hands
- Avoid use of flame unless necessary
- Have cabinet certified annually



HEPA (high efficiency particulate air)

BSL2 Laboratory

(Secondary Barriers)



… BSL2

Laboratory personnel working with human-derived materials should refer to the OSHA *Bloodborne Pathogen Standard for specific required precautions* .

(U.S. Department of Labor, Occupational Safety and Health Administration. 1991. Occupational Exposure to Bloodborne Pathogens, Final Rule. Fed. Register **56**:64175-64182).

Bio-safety Level 3 (BSL3)

For microorganisms that cause serious disease, transmitted by inhalation

- **HIV-1, SARS coronavirus*, H5N1 influenza virus*, yellow fever virus, Hantavirus*, *M. tuberculosis*, *Yersinia pestis* (plague)** ***And Coxiella burnetii (Q fever)***

  Containment lab: double door entry; directional airflow (negative pressure, 12.45 to 25 Pa differential with 6-10 air changes/hr (ACH)

- **BSL2 practices PLUS**

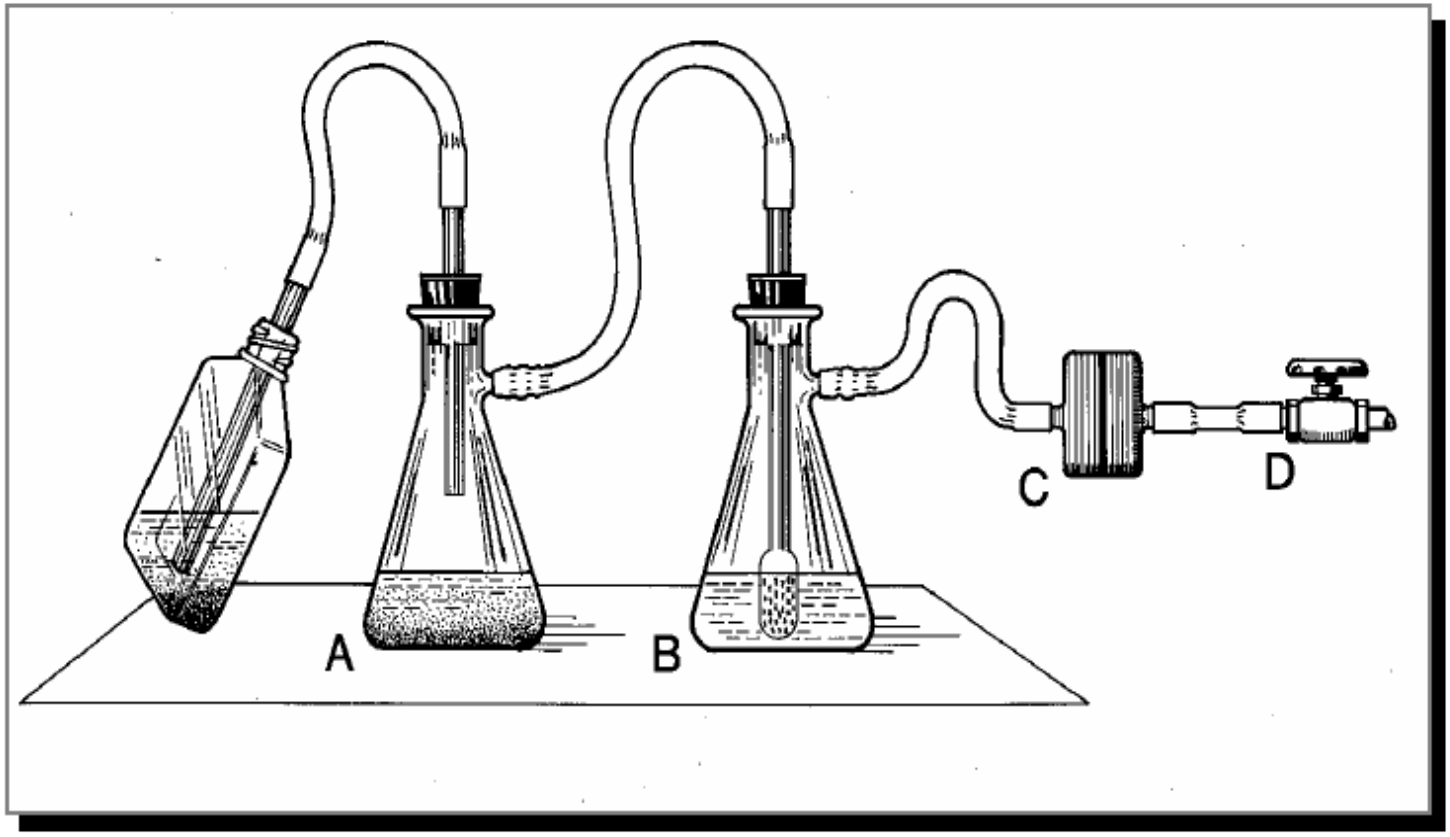
*, preferably in BSL4

BSL3 Special Practices

- Work in certified BSC
- Use bio-aerosol-containing equipment
- Decontaminate spills promptly

BSL3 Special Practices

– Protected Vacuum lines



BSL3 Special Practices

Respiratory protection

BSL3 Special Requirements

Supervision

- Supervisor is a competent scientist experienced working with agents
  - Establishes criteria for entry
  - Restricts access
  - Develops policies/procedures
  - Trains lab personnel

BSL3 Special Requirements

- **Lab Personnel**
  - Strictly follow guidelines
  - Demonstrate proficiency
  - Receive appropriate training
  - Report incidents
  - Participate in medical surveillance

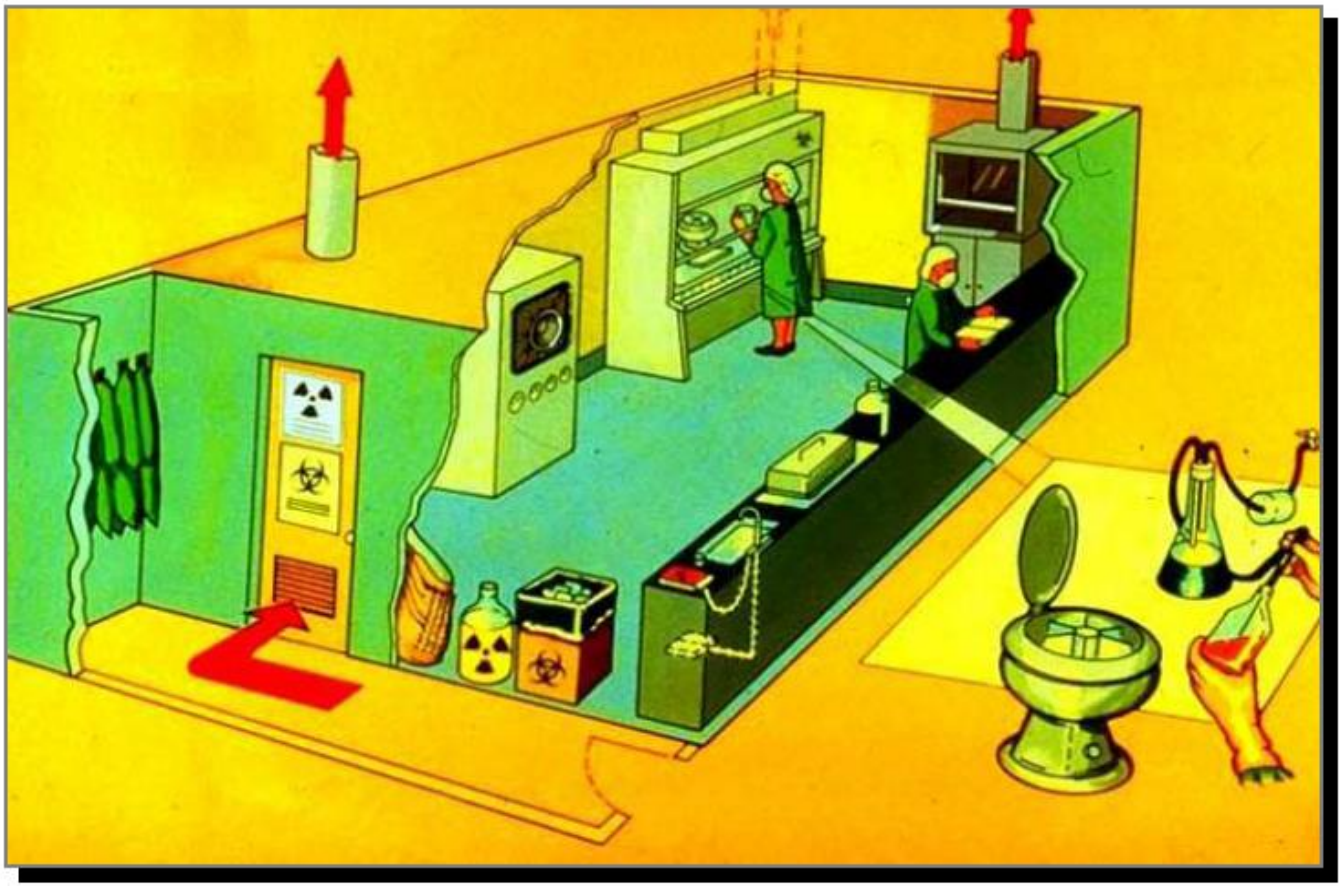
BSL3 Special Requirements

BSL3 lab at ICGEB

- Policies and procedures for entry
- Biohazard warning signs
- Biosafety manual specific to lab
- Training with
  annual updates

BSL3 Laboratory

(Secondary Barriers)



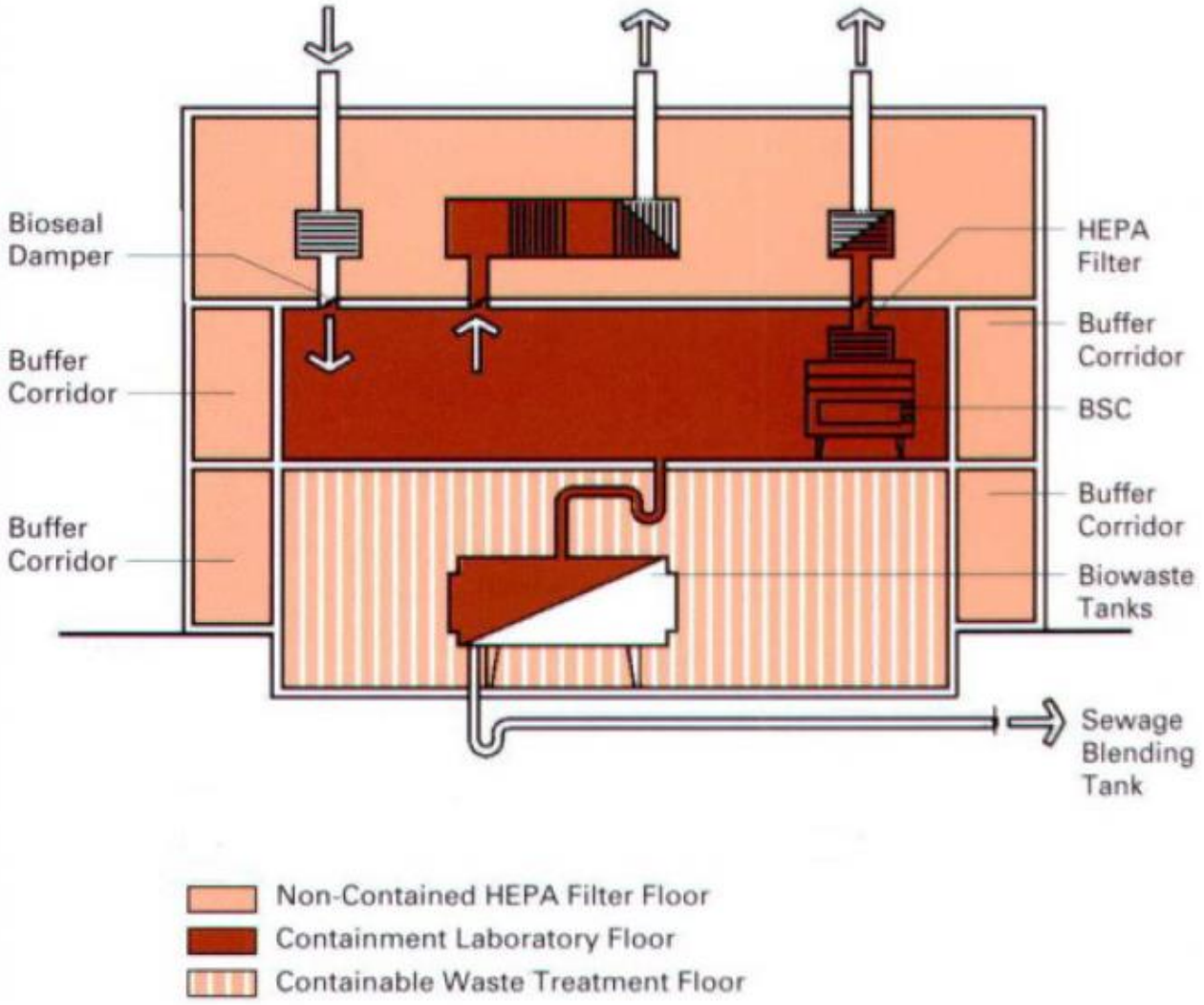
BSL4 Laboratory
(with Tertiary Barriers)

•Dangerous/exotic agents which pose a high risk of life-threatening disease, aerosol-transmitted lab infections or related agents with unknown risk of transmission.

- BSL -3 practices plus:
  - **Clothing change before entering laboratory**
  - **Shower on exit**
  - **All materials decontaminated on exit from facility**

- Safety Equipment:
  - **Class III Biosafety cabinet**
  - **Class I or II biosafety cabinet**
  - **WITH full-body, air supplied,**
    - **positive personnel suit**

BSL4 Laboratory
(Tertiary Barriers)



Risk Assessment

- **Pathogenicity of material – disease incidence and severity**
- **Routes of Transmission – parenteral, airborne or ingestion**
- **Agent Stability – ease of decontamination**
- **Infectious Dose – LD50**
- **Concentration – infectious organisms/vol. & working volume**
- **Origin of material - Wild Type, exotic, primary cells**
- **Availability of effective prophylaxis – Hep. B vaccine**
- **Medical surveillance – exposure management**
- **Skill level of staff**

Accident & Response !!!

Carefully think all the angles of your experiments;

Take precautions ! Remain alert !!

Emergency Responses

Personal contamination

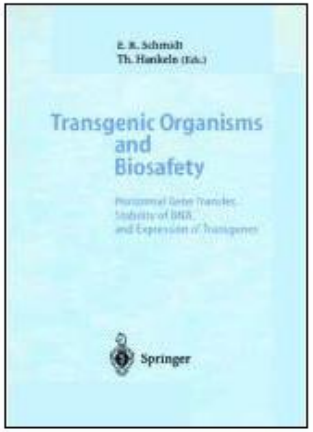
- **Alert** co-workers
- **Clean** exposed surface with soap/water, eyewash (eyes), or saline (mouth)
- **Apply** first aid and treat as an emergency
- **Notify** supervisor or security desk (after hours)
- **Tell** Doctor that you work with infectious agents or toxins

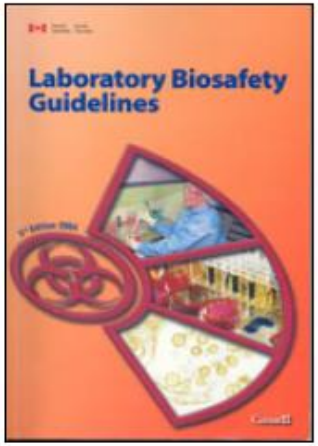
Emergency Responses

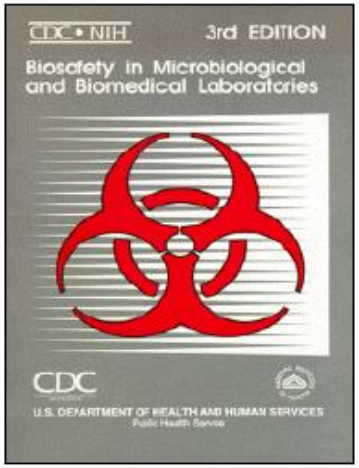
Surface contamination

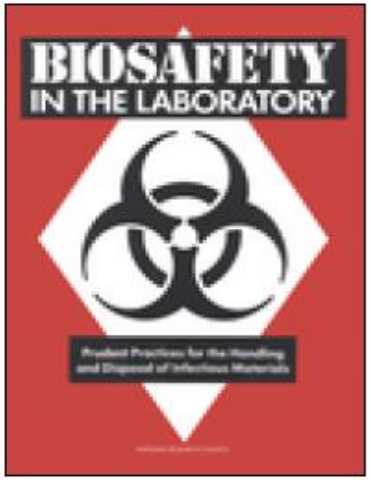
- Alert co-workers
- Define/isolate contaminated area
- Remove glass/lumps with forceps or scoop
- Apply absorbent sheets to spill
- Apply disinfectant to surface and allow adequate contact time (~ 20 min)
- Mop up with sheet and clean with ethanol
- Properly dispose of materials
- Notify supervisor

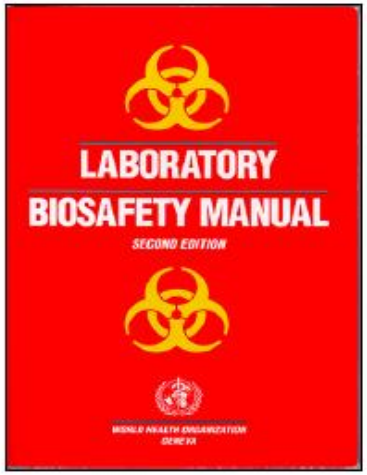
Literature on Bio-safety



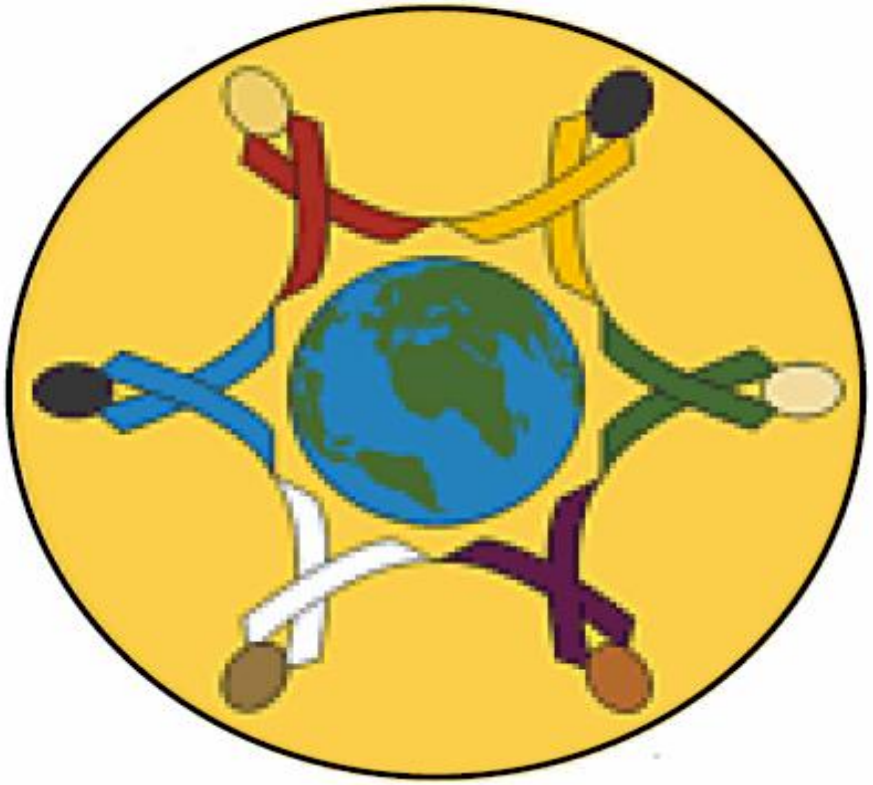








Reading Material



Help maintain BMS
a Bio-safe work place

Acknowledgement

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- Thanks are due to all those original contributors and entities whose pictures used for making this presentation.