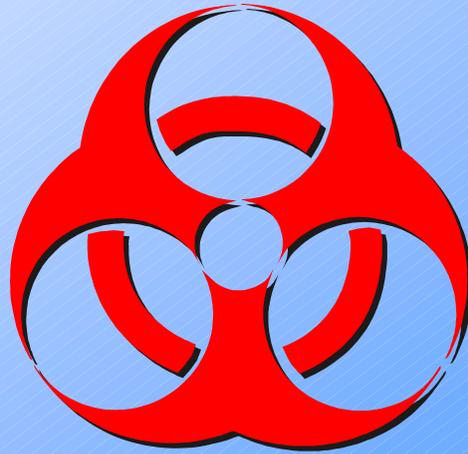
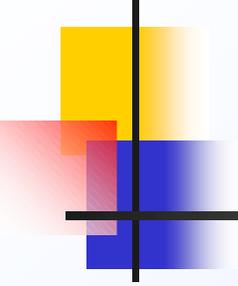


Module 2

Biosafety



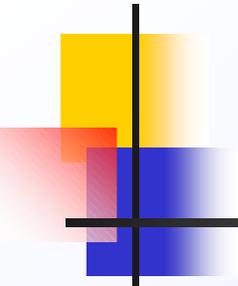


Principles

Definition

Biohazard

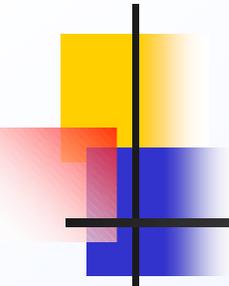
An agent of biological origin that has the capacity to produce deleterious effects on humans, i.e. microorganisms, toxins and allergens derived from those organisms; and allergens and toxins derived from higher plants and animals.



Introduction

Development of Biosafety Practices

- **1941 - Meyer and Eddie**
 - *74 lab associated brucellosis infections in US*
- **1949 - Sulkin and Pike**
 - *222 viral infections (21 fatal)*
 - *Only 27% related to known accidents*



Introduction

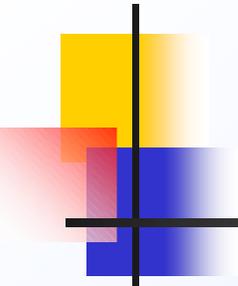
Development of Biosafety Practices

- **1951, 1965, 1976 - Sulkin and Pike**

- *Surveys for lab-associated infections*
- *More than 5,000 labs*
- *Cumulative total of 3,921 cases cited*
- *Most commonly reported:*

- Hepatitis
- Tuberculosis
- Typhoid

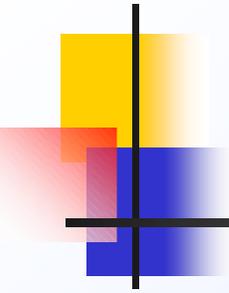
- Venezuelan Equine Encephalitis
- Brucellosis
- Tularemia



Introduction

Development of Biosafety Practices

- **1951, 1965, 1976 - Sulkin and Pike**
 - *Fewer than 20% associated with known accidents*
 - *Exposure to infectious aerosols plausible (but unconfirmed) for >80% of reported cases*



Introduction

Why Biosafety Practices?

Protection:

- workers
- “products”
- co-workers
- lab support personnel
- environment

Introduction

Chain of Infection

Reservoir of pathogen

Portal of escape

Transmission

Route of entry/infectious dose

Susceptible host

Incubation period

*Practices/
Equipment*

PPE

Immunization

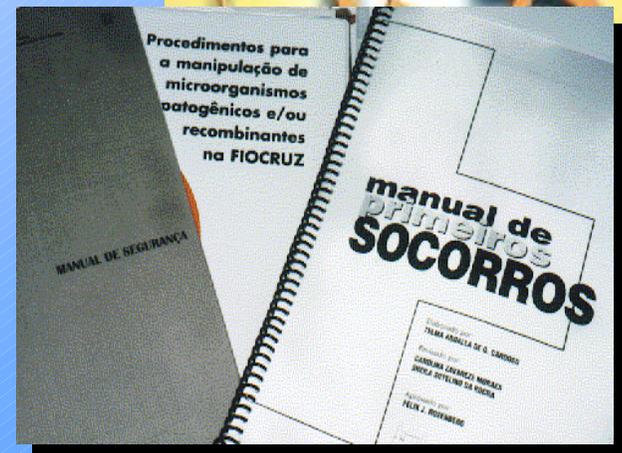
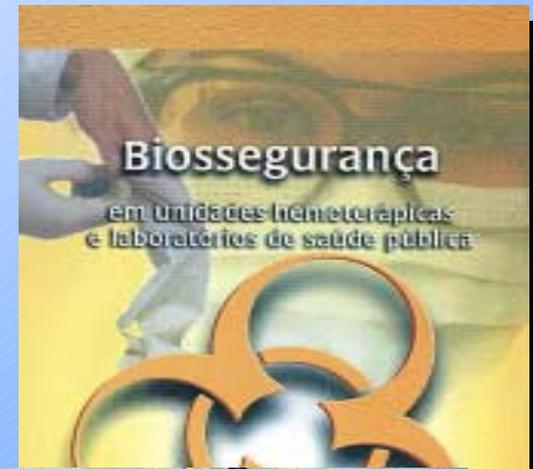
Surveillance

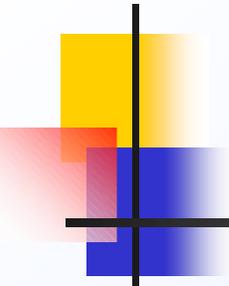
Risk Assessment

Principles

General Lab Requirements

- Knowledgeable supervisor
- Knowledgeable personnel
 - *Aware of potential hazards*
 - *Proficient in practices & techniques*
- Lab specific biosafety manual

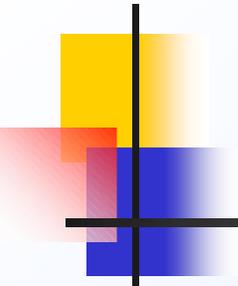




Principles

General Lab Requirements

- **Biosafety Levels (BSLs)**
- **Laboratory Practice and Technique**
 - *Standard Practices*
 - *Special Practices*
- **Safety Equipment (Primary Barriers)**
- **Facility Design and Construction (Secondary Barriers)**



Principles

General Lab Requirements

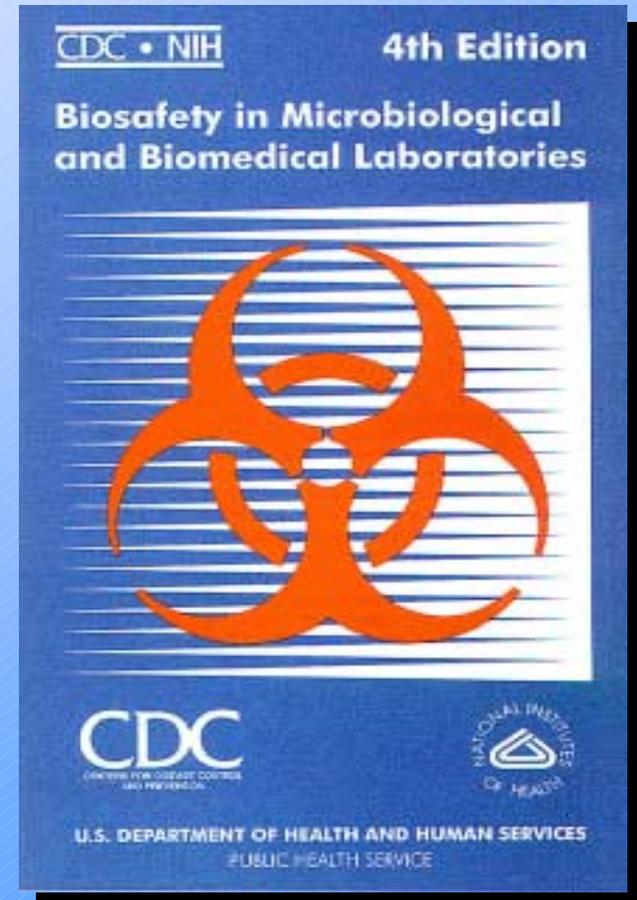
- **Biosafety cabinets (BSCs) - BSL 2/3**
- **Personal protective clothing**
 - *Gloves*
 - *Gowns*
 - *Eye and face protection*
- **Pipetting Devices**
- **Safety centrifuge cups and rotors**

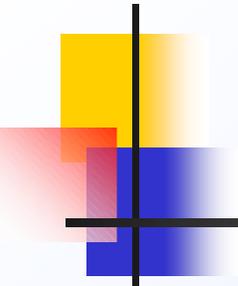
Principles

Definition

Biosafety

The application of combinations of laboratory practice and procedure, laboratory facilities, and safety equipment when working with potentially infectious microorganisms.

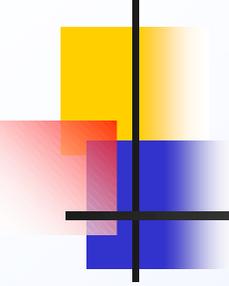




Principles

Biosafety Levels

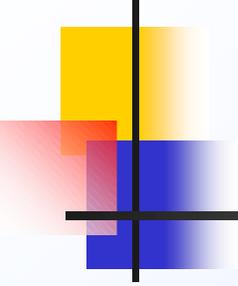
- ***BSL1*** - agents not known to cause disease.
- ***BSL2*** - agents associated with human disease.
- ***BSL3*** - indigenous/exotic agents associated with human disease and with potential for aerosol transmission.
- ***BSL4*** - dangerous/exotic agents of life threatening nature.



Biosafety Level 1

Introduction

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.



Biosafety Level 1

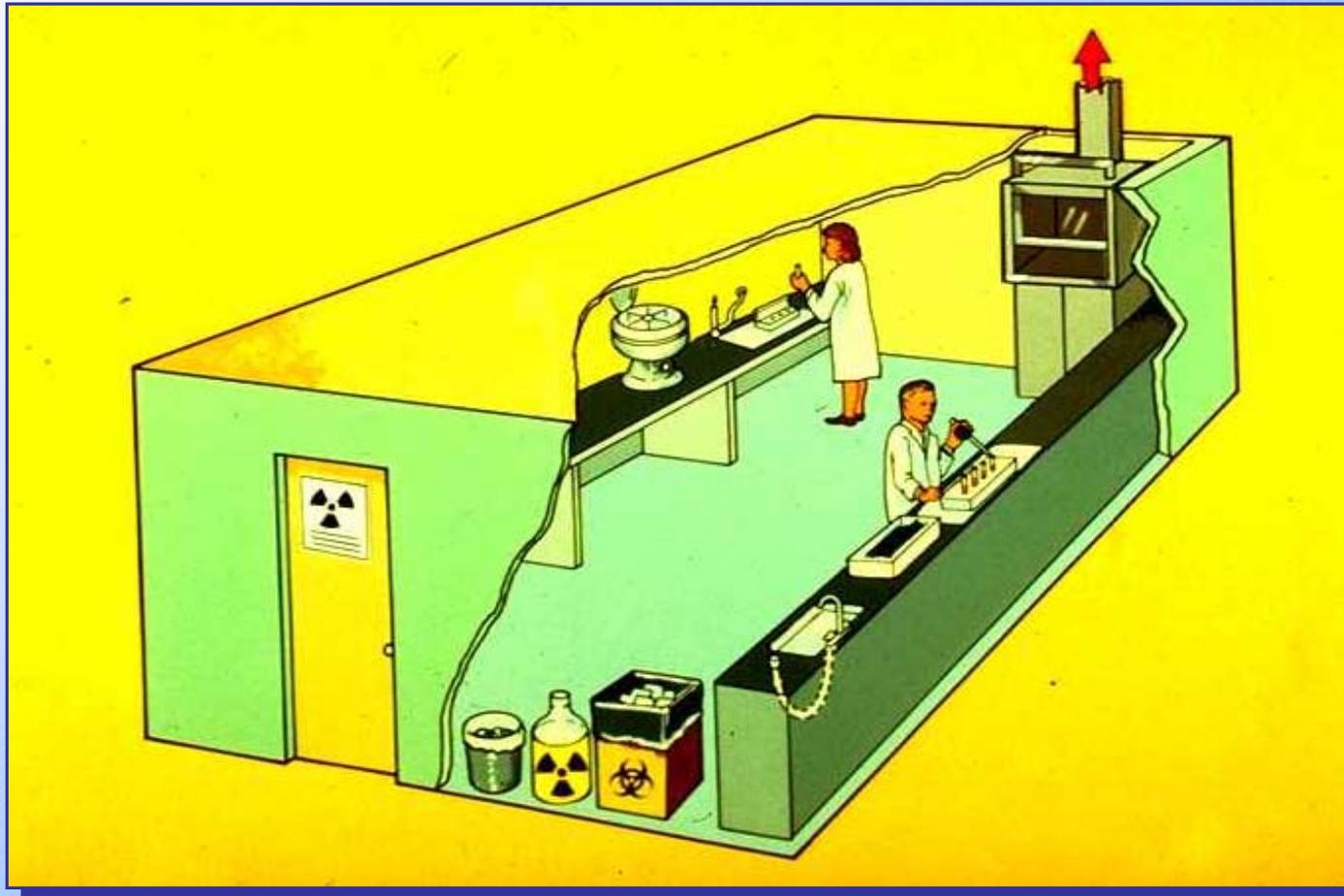
Introduction

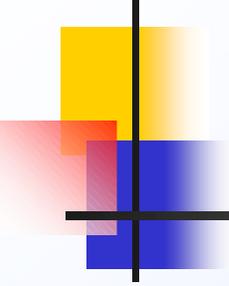
Examples:

- *Bacillus subtilis*
- *Naegleria gruberi*
- *Infectious canine hepatitis virus*
- *E. coli*

Biosafety Level 1

Facility Design (Secondary Barrier)





Biosafety Level 1

Facility Design (Secondary Barrier)

Requirements:

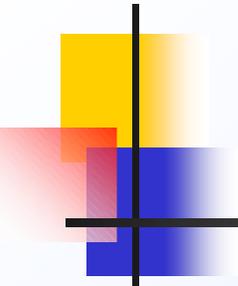
- *Laboratories have doors*
- *Sink for hand washing*
- *Work surfaces easily cleaned*
- *Bench tops are impervious to water*
- *Sturdy furniture*
- *Windows fitted with flyscreens*

Biosafety Level 1

Facility Design (Secondary Barrier)



**Easily cleaned
and
decontaminated**



Biosafety Level 1

Facility Construction (Secondary Barrier)

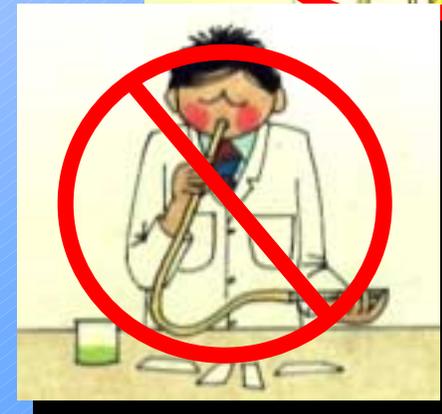
Requirements:

- *Location - not separated*
- *Structure - normal construction*
- *Ventilation - none*

Biosafety Level 1

Standard Microbiological Practices

- **Restrict or limit access when working**
- **Prohibit eating, drinking and smoking**
- **Prohibit mouth pipetting**



Biosafety Level 1

Standard Microbiological Practices



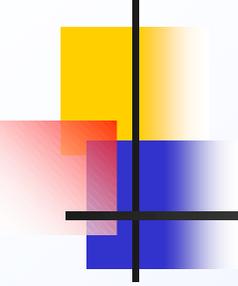
***Use
mechanical
pipetting
devices***

Biosafety Level 1

Standard Microbiological Practices



Wash hands



Biosafety Level 1

Standard Microbiological Practices

- **Minimize splashes and aerosols**
- **Decontaminate work surfaces daily**
- **Decontaminate wastes**
- **Maintain insect & rodent control program**

Biosafety Level 1

Safety Equipment (Primary Barriers)

Protective clothing

- *Lab coat*
- *Gloves*



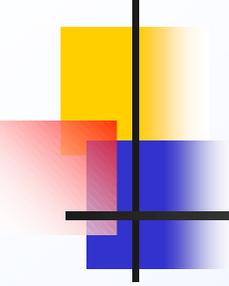
Biosafety Level 1

Safety Equipment (Primary Barriers)

Personal protective equipment

- *Face protection*
- *Eye protection*

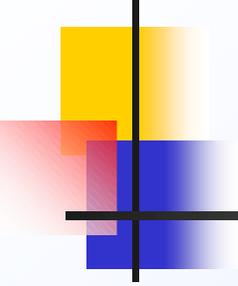




Biosafety Level 1

Special Practices

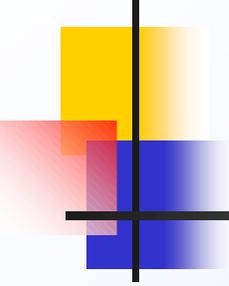
None required



Biosafety Level 1

Training Requirements

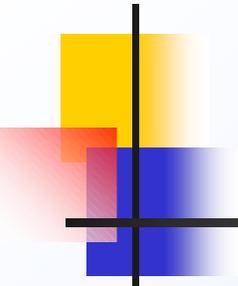
- **Supervisor**
 - *Scientist with general training in microbiology or related science*
- **Lab Personnel**
 - *Specific training in lab procedures*



Biosafety Level 2

Introduction

Suitable for work involving *agents of moderate potential hazard* to personnel and the environment.



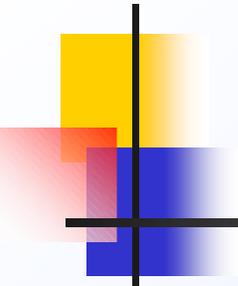
Biosafety Level 2

Introduction

Examples:

- *Measles virus*
- *Salmonellae*
- *Toxoplasma spp.*
- *Hepatitis B virus*

* *Immunization or antibiotic treatment is available*



Biosafety Level 2

Introduction

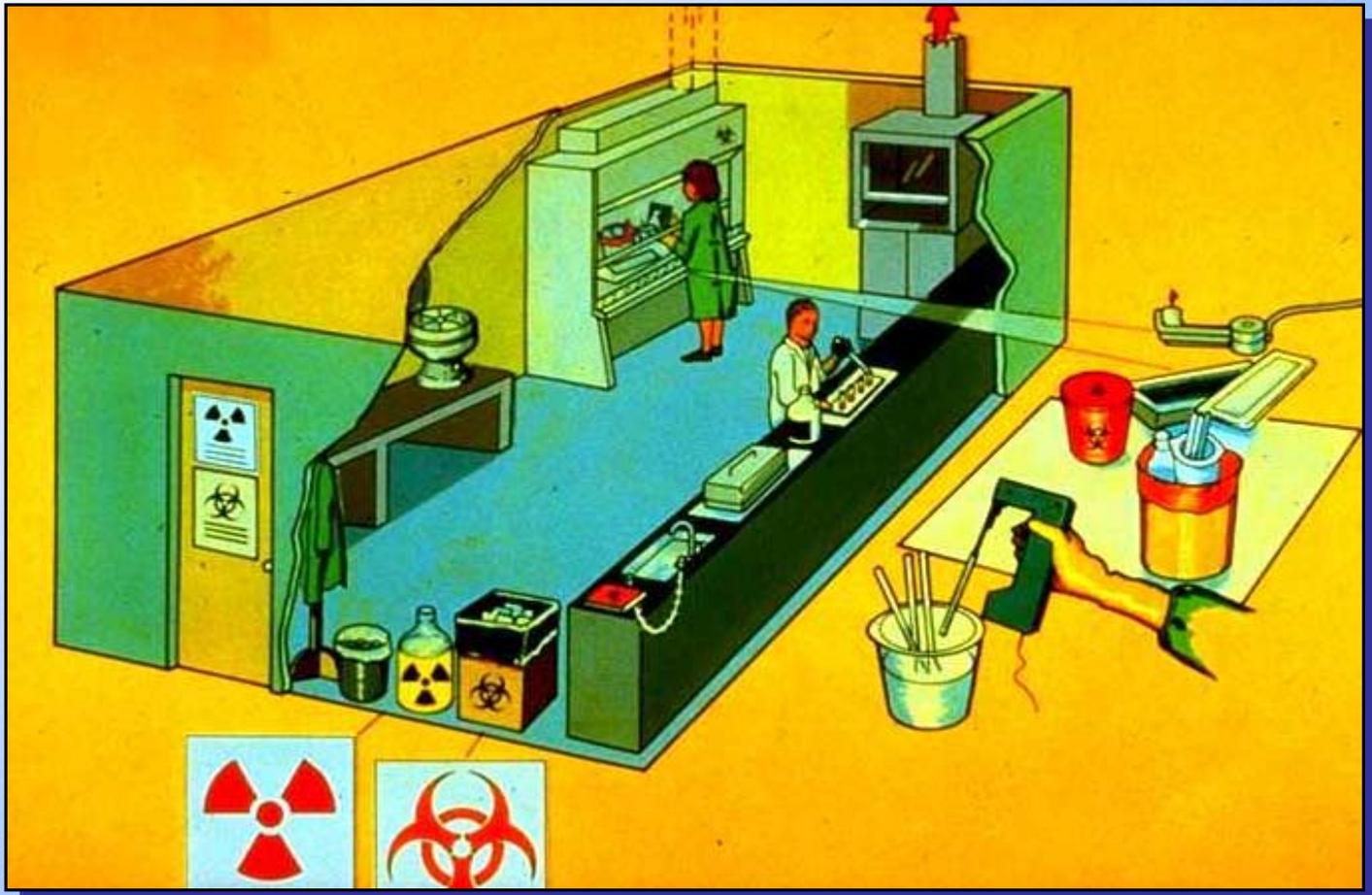
Examples:

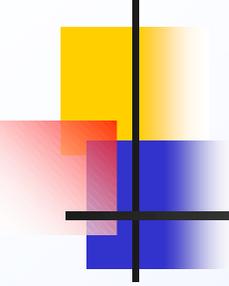
- ***Bloodborne pathogens***
- ***Human body fluids/particularly when visibly contaminated with blood***

**** Extreme precaution with contaminated needles or sharp instruments***

Biosafety Level 2

Facility Design (Secondary Barriers)



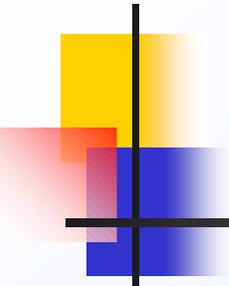


Biosafety Level 2

Facility Design (Secondary Barriers)

Requirements:

- *Laboratories have lockable doors*
- *Sink for hand washing*
- *Work surfaces easily cleaned*
- *Bench tops are impervious to water*
- *Sturdy furniture*



Biosafety Level 2

Facility Design (Secondary Barriers)

Requirements (cont.):

- *Biological safety cabinets installed as needed*
- *Adequate illumination*
- *Eyewash readily available*
- *Air flows into lab without re-circulation to non-lab areas*
- *Windows fitted with flyscreens*

Biosafety Level 2

Facility Design (Secondary Barrier)



**Restricted
access when
work in
progress**

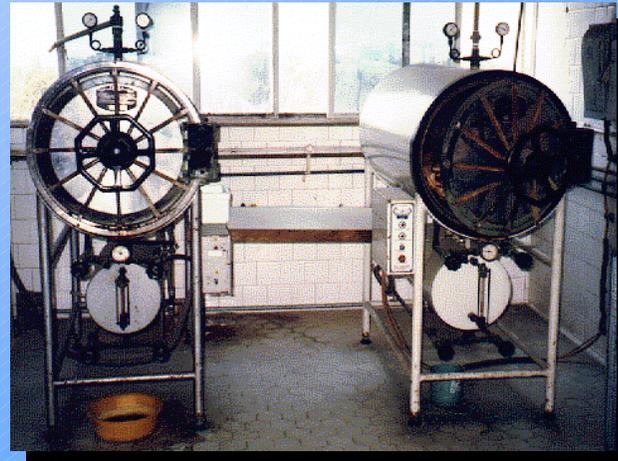
Biosafety Level 2

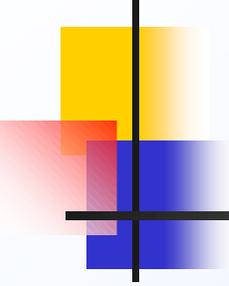
Laboratory Facilities (Secondary Barriers)

- **BSL-1 Facilities PLUS:**



- ***Autoclave available***
- ***Eyewash station available***



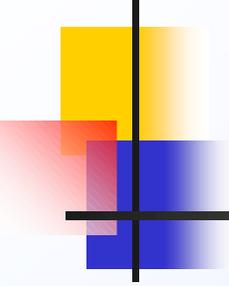


Biosafety Level 2

Facility Construction (Secondary Barrier)

Requirements:

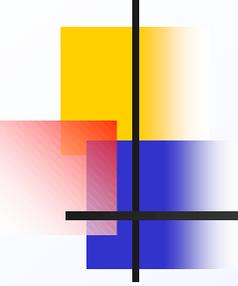
- *Location - separated from public areas*
- *Structure - normal construction*
- *Ventilation - directional*



Biosafety Level 2

Standard Microbiological Practices

As in BSL-1



Biosafety Level 2

Safety Equipment (Primary Barriers)

In addition to BSL-1:

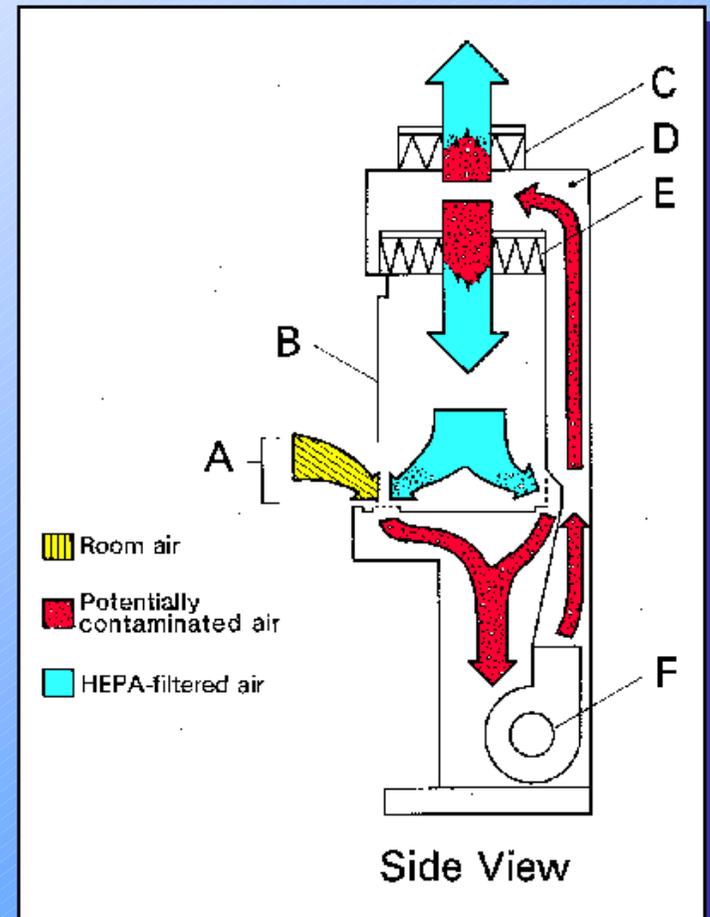
- *Use biosafety cabinets (class II) for work with infectious agents involving:*
 - **Aerosols and splashes**
 - **Large volumes**
 - **High concentrations**

Biosafety Level 2

Safety Equipment (Primary Barriers)

■ Class II Biosafety Cabinet

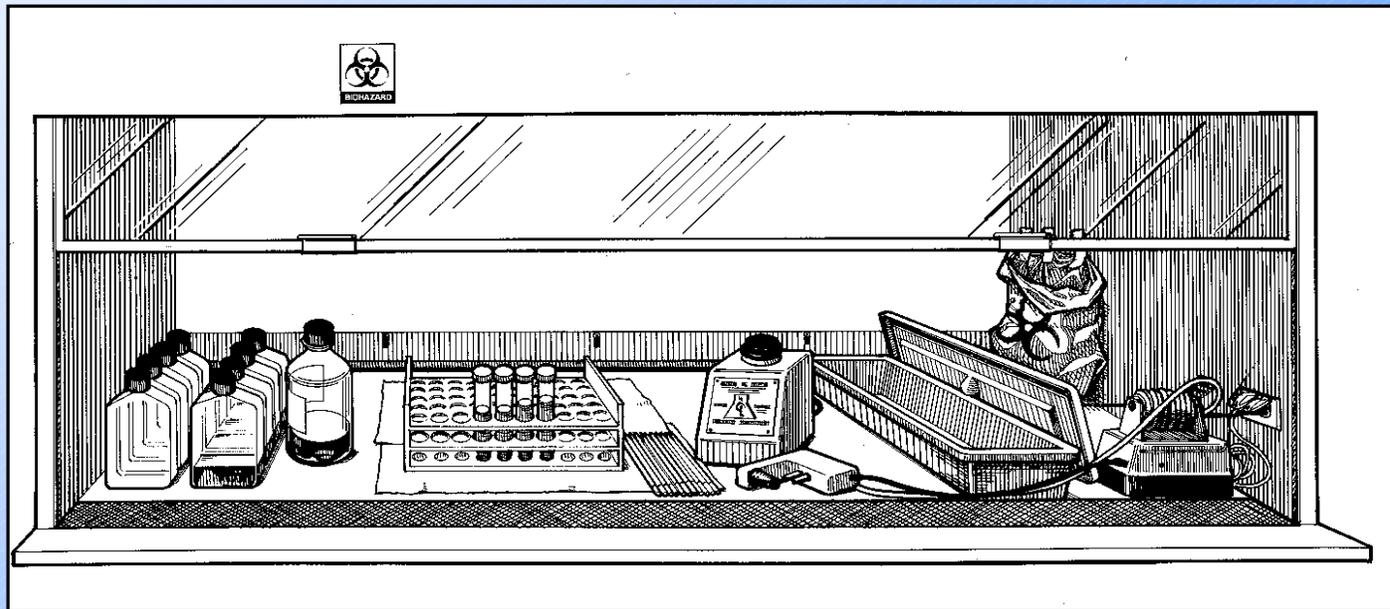
■ *Airflow*



Biosafety Level 2

Safety Equipment (Primary Barriers)

- **Class II Biosafety Cabinet**
 - *Equipment layout*

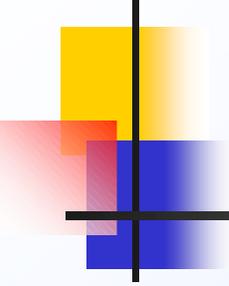


Biosafety Level 2

Safety Equipment (Primary Barriers)

- **Class II Biosafety Cabinet**
 - *Technique*





Biosafety Level 2

Special Practices

Supervision

- *Supervisor is a competent scientist with increased responsibilities*
 - **Limits access if immunocompromised**
 - **Restricts access to immunized**

Lab Personnel

- *Aware of potential hazards*
- *Proficient in practices/techniques*

Biosafety Level 2

Special Practices

Needles & Sharps Precautions

- *Use sharps containers*
- *DON'T break, bend, re-sheath or reuse syringes or needles*



Biosafety Level 2

Special Practices

Needles & Sharps Precautions (cont.)

- ***DON'T place needles or sharps in office waste containers***



Biosafety Level 2

Special Practices

Needles and Sharps Precautions (cont.)

- ***DON'T touch broken glass with hands***



Biosafety Level 2

Special Practices

Needles and Sharps Precautions (cont.)

- *Use plasticware*



Biosafety Level 2

Special Practices

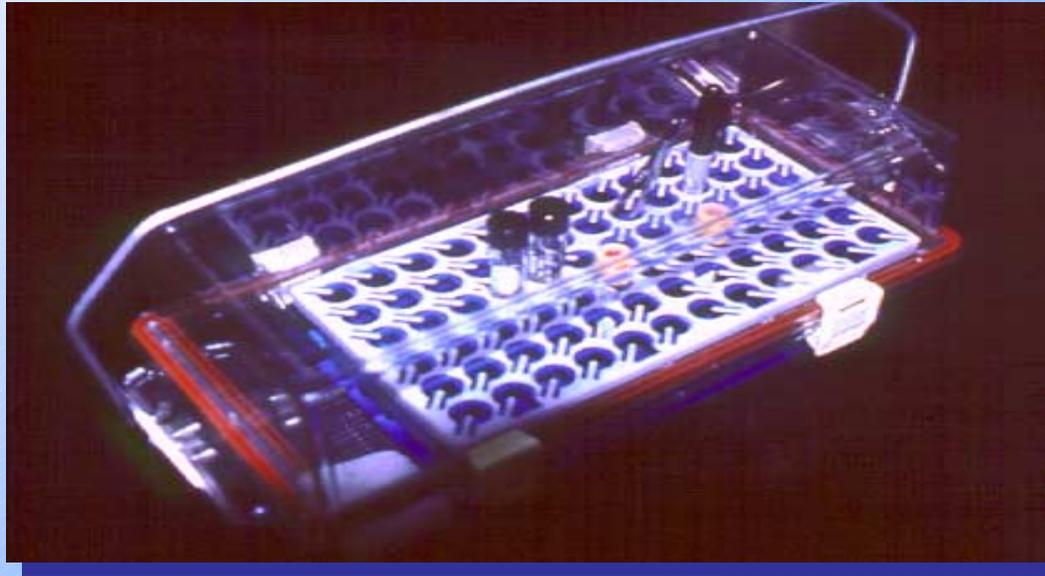
- **Policies and procedures for entry**
- **Biohazard warning signs**
- **Biosafety manual specific to lab**
- **Training with annual updates**



Biosafety Level 2

Special Practices

- **Use leak-proof transport containers**

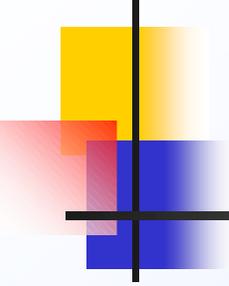


Biosafety Level 2

Special Practices

- **Immunizations**
- **Baseline serum samples**

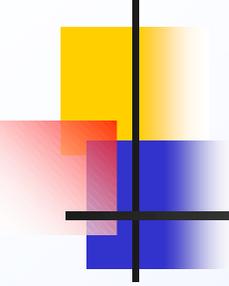




Biosafety Level 2

Special Practices

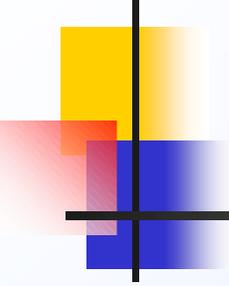
- **Decontaminate work surfaces**
- **Report spills and accidents**
- **No animals in laboratories**



Biosafety Level 3

Introduction

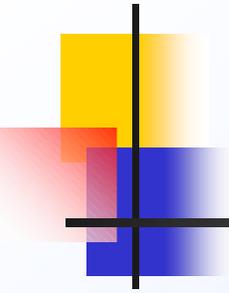
Suitable for work with infectious agents which *may cause serious or potentially lethal disease* as a result of exposure by the inhalation route.



Biosafety Level 3

Introduction

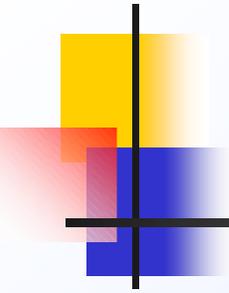
- **Exposure potential to pathogens spread by aerosol**
- **Infection serious, possibly lethal**
- **Examples:**
 - *M. tuberculosis*
 - *St. Louis encephalitis virus*
 - *Coxiella burnetii*



Biosafety Level 3

Laboratory Facilities (Secondary Barriers)

- **BSL-1 and 2 Facilities PLUS:**
 - *Separate building or isolated zone*
 - *Double door entry*
 - *Directional inward airflow*
 - *Single-pass air; 10-12 air changes/hour*



Biosafety Level 3

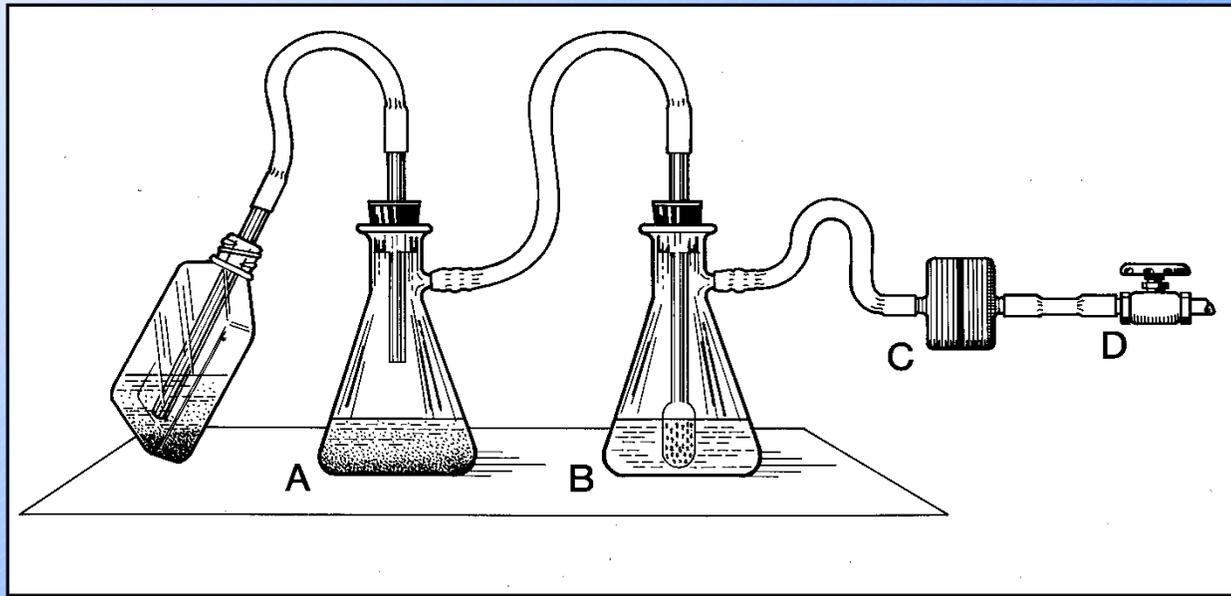
Laboratory Facilities (Secondary Barriers)

- **BSL-1 and 2 Facilities PLUS (cont.):**
 - *Enclosures for aerosol generating equipment*
 - *Room penetrations sealed*
 - *Walls, floors and ceilings are water resistant for easy cleaning*

Biosafety Level 3

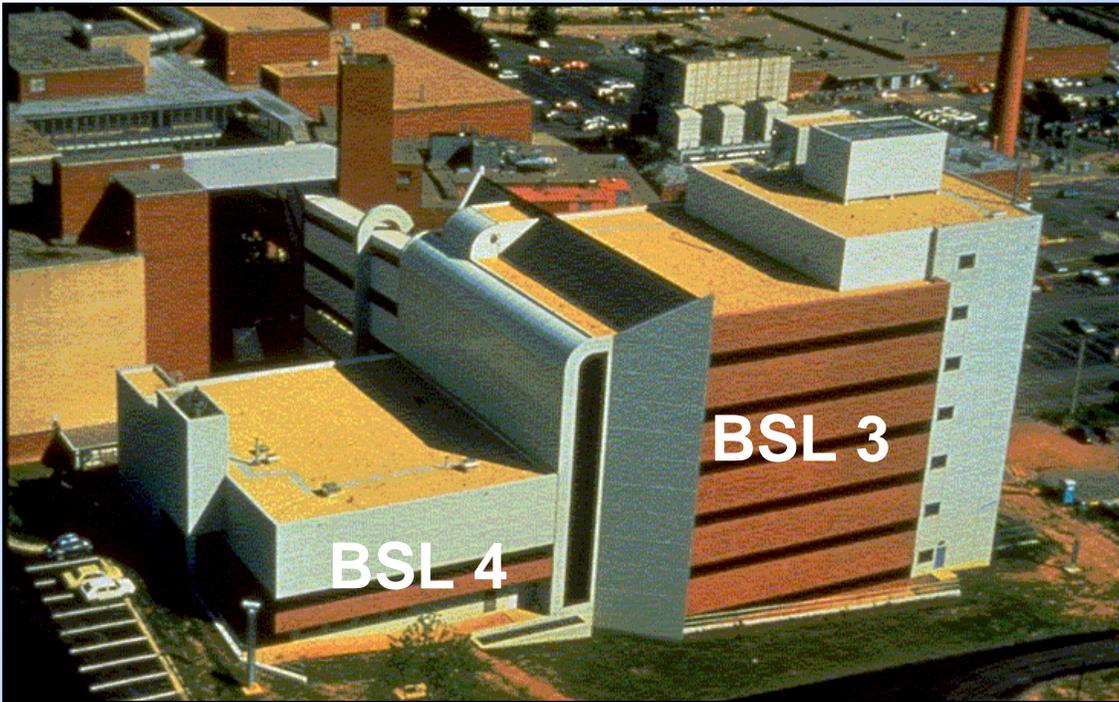
Laboratory Facilities (Secondary Barriers)

- **BSL-1 and 2 Facilities PLUS:**
 - *Vacuum lines protected with liquid disinfectant traps or HEPA filters*



Facility Design

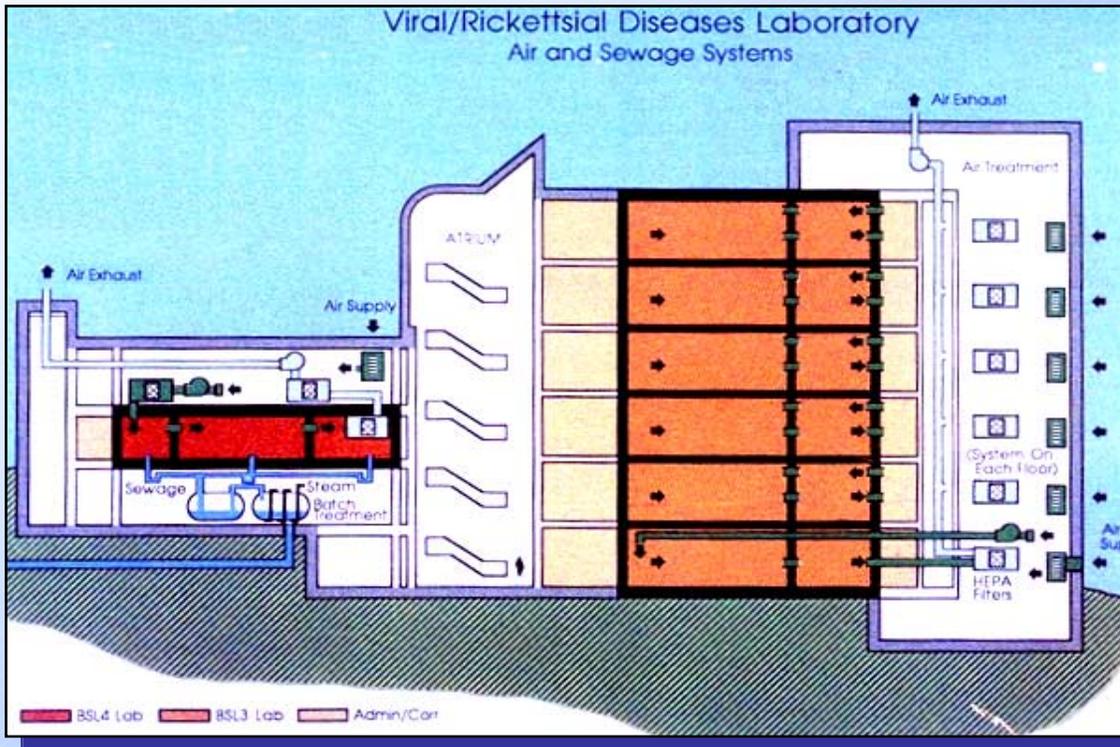
(Tertiary Barriers)



**CDC
Containment
Laboratory**

Facility Design

(Tertiary Barriers)



- Lab structure
- Lab ventilation

Biosafety Level 3

Standard Microbiological Practices

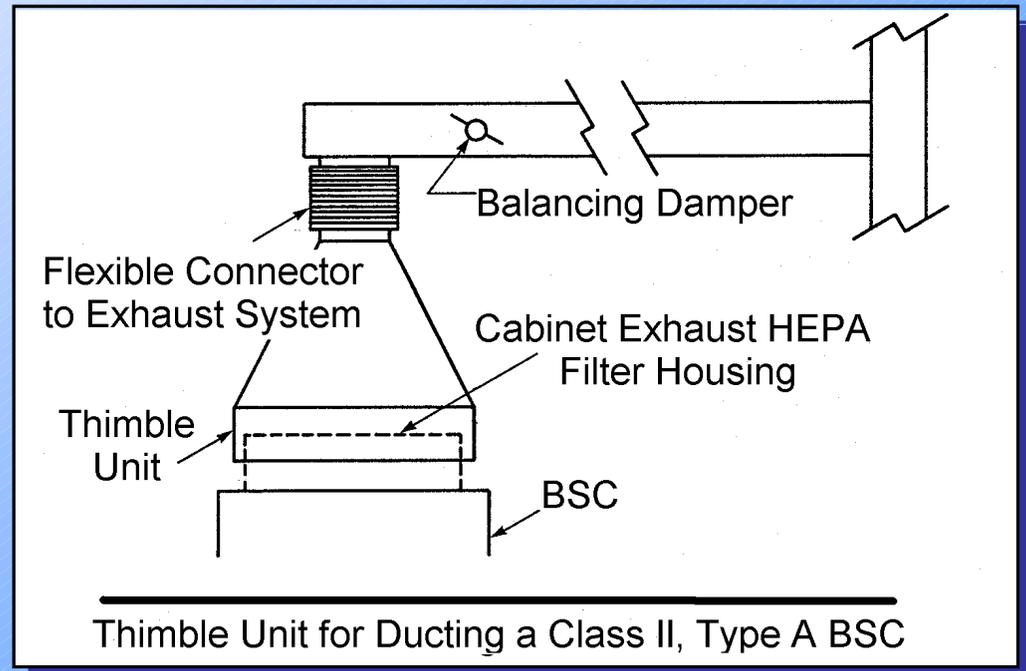
**As in BSL - 1
and - 2**



Biosafety Level 3

Safety Equipment (Primary Barriers)

- **BSL-1 and 2 Safety Equipment PLUS:**
 - *BSC class II or III to manipulate infectious material*



Biosafety Level 3

Safety Equipment (Primary Barriers)

- **BSL-1 and 2 Safety Equipment PLUS:**
 - *Respiratory protection may be indicated*

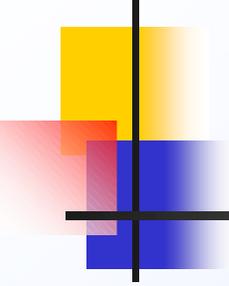


Biosafety Level 3

Special Practices

- **BSL-2 Special Practices PLUS:**
 - *Work in certified BSC*
 - *Use bioaerosol-containing equipment*
 - *Decontaminate spills promptly*



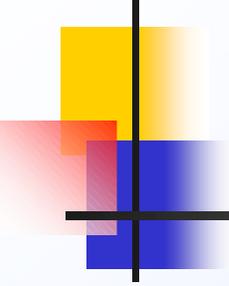


Biosafety Level 3

Special Practices

Supervision

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

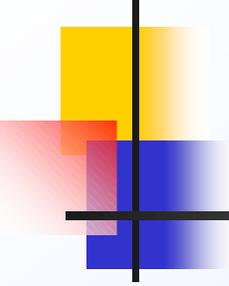


Biosafety Level 3

Special Practices

Lab Personnel

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



Biosafety Level 4

Introduction

Suitable for work with dangerous and exotic agents that *pose a high individual risk of aerosol-transmitted laboratory infections and life-threatening disease.*

Biosafety Level 4

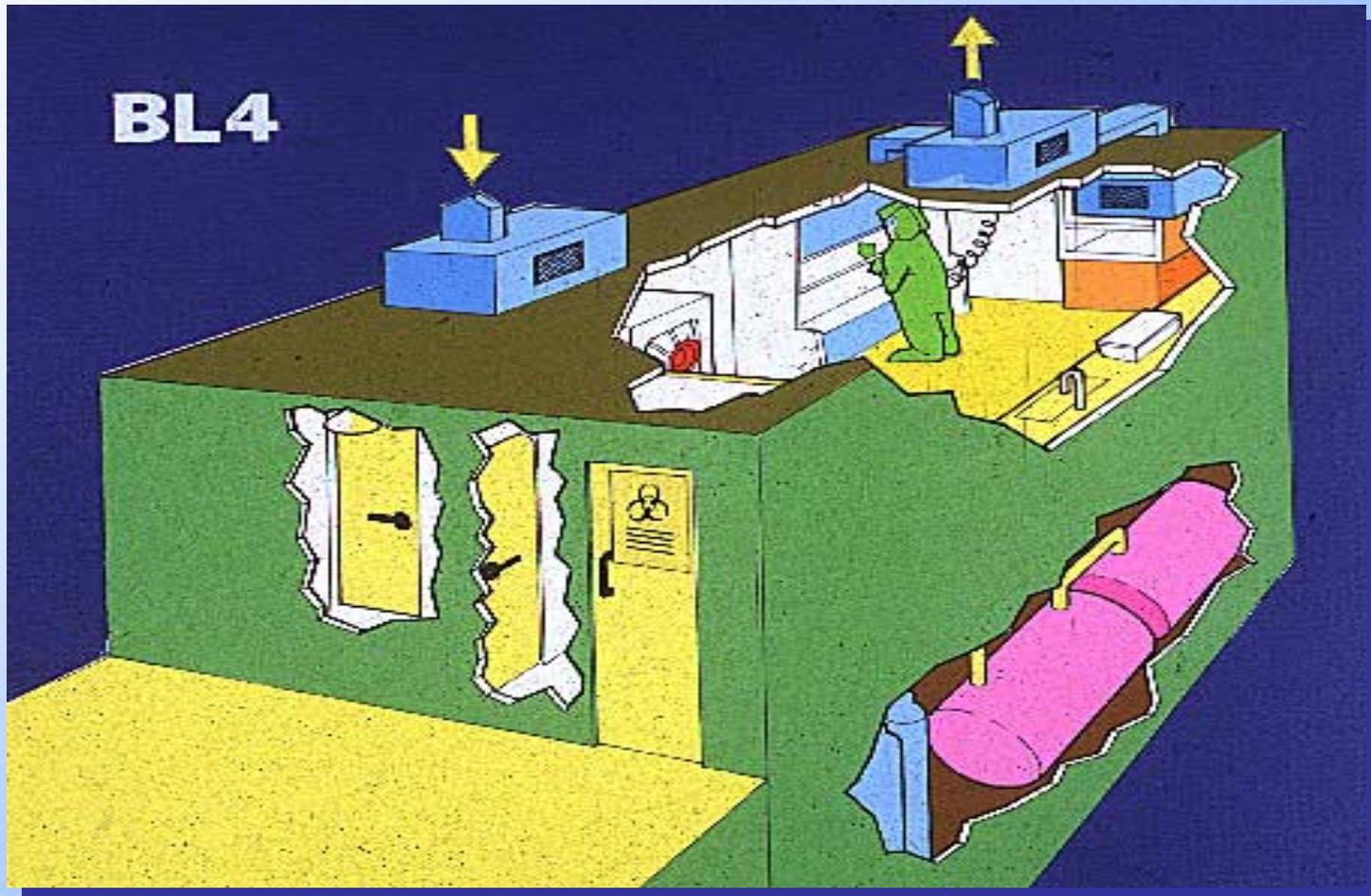
Introduction

- **Exposure potential to pathogens spread by aerosol or with unknown risk of transmission**
- **Infection possibly lethal**
- **Examples:**
 - *Ebola Zaire*
 - *Sin Nombre virus*
 - *Rift Valley Fever*



Biosafety Level 4

Laboratory Facilities (Secondary Barriers)

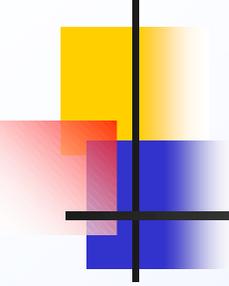


Biosafety Level 4

Laboratory Facilities (Secondary Barriers)

- **BSL-1, 2, and 3 Facilities PLUS:**
 - *Separate building or isolated zone*
 - *Double door entry*
 - *Directional inward airflow*
 - *Single-pass air*
 - *Dedicated supply and exhaust, vacuum, and decon systems*

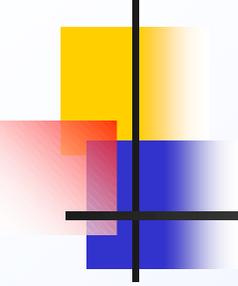




Biosafety Level 4

Laboratory Facilities (Secondary Barriers)

- **BSL-1, 2 and 3 Facilities PLUS (cont.):**
 - *Enclosures for aerosol generating equipment*
 - *Double door autoclaves*
 - *Room penetrations sealed*
 - *Walls, floors and ceilings are sealed to form an internal seal*



Biosafety Level 4

Laboratory Facilities (Secondary Barriers)

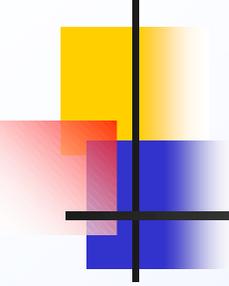
- **BSL-1, 2 and 3 Facilities PLUS (cont.):**
 - *Connecting inner and outer doors - interlocked to prevent simultaneous opening*
 - *Liquid effluents are decontaminated by an approved method and certified before discharge*
 - *Communication system between inside and outside of the lab*

Biosafety Level 4

Laboratory Facilities (Secondary Barriers)

- **BSL 1, 2, and 3 Facilities PLUS:**
 - *Emergency breathing air*
 - *Emergency generator*
 - *Emergency exit*





Biosafety Level 4

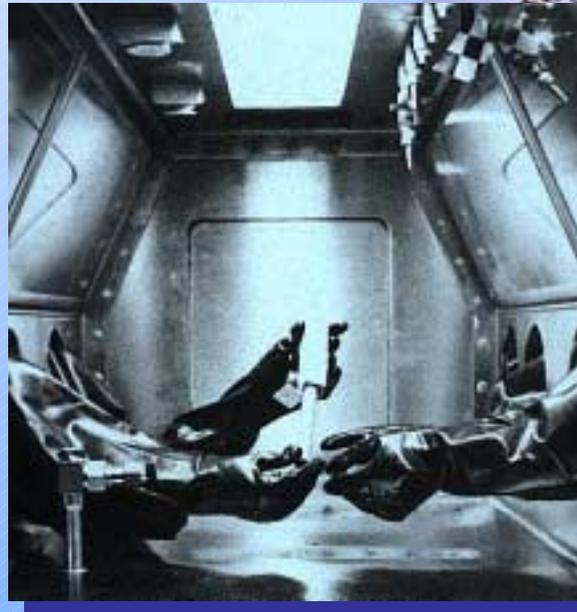
Standard Microbiological Practices

As in BSL 1, 2, and 3

Biosafety Level 4

Safety Equipment (Primary Barriers)

- **BSL 1, 2, and 3 Safety Equipment PLUS:**
 - *Class II (B2) or III biological safety cabinets to manipulate infectious material*



Biosafety Level 4

Safety Equipment (Primary Barriers)

- **BSL 1, 2, and 3 Safety Equipment PLUS:**
 - *Positive pressure personnel suit*



Biosafety Level 4

Special Practices

- **BSL 3 Special Practices PLUS:**
 - *Decontaminate all liquid effluent*
 - *Decontaminate all solid wastes*

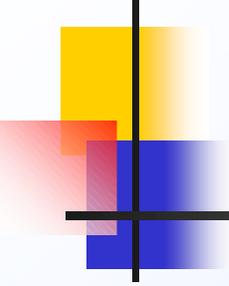


Biosafety Level 4

Special Practices

- **Controlled access**
- **Personnel enter facility through changing room where they are required to change into laboratory clothing**
- **Showers are required upon exit from the laboratory**
- **Supplies enter lab through double-door autoclave or fumigation chamber**



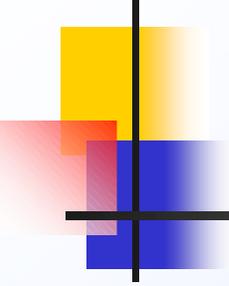


Biosafety Level 4

Special Practices

Supervision

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

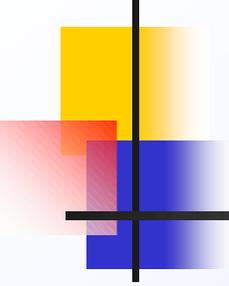


Biosafety Level 4

Special Practices

Lab Personnel

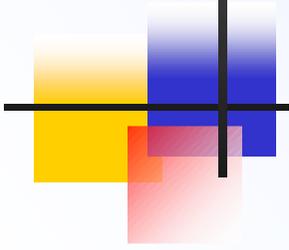
- ***Strictly follow guidelines***
- ***Demonstrate proficiency***
- ***Receive appropriate training***
- ***Report incidents***
- ***Receive available immunizations***
- ***Participate in medical surveillance***



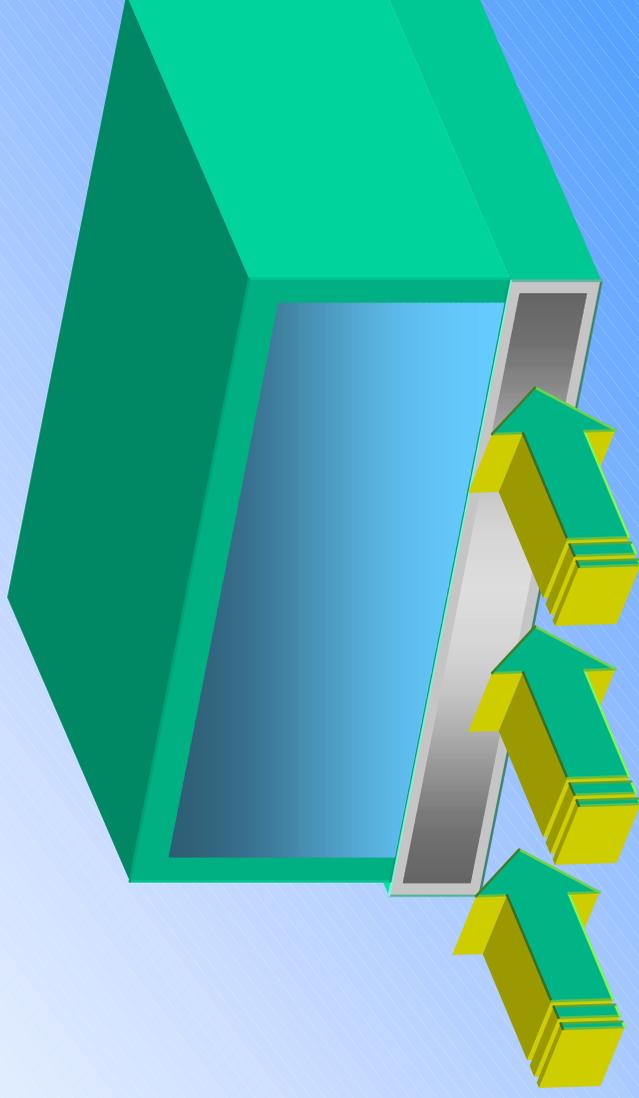
Principles of Biosafety

Summary

- **BSL 1 - 4**
 - *Standard Practices*
 - *Special Practices*
 - *Safety Equipment (Primary Barriers)*
 - *Laboratory Facilities (Secondary Barriers)*
 - *Building (Tertiary Barriers)*



Biological Safety Cabinets

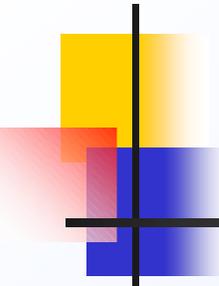


Biological Safety Cabinets

Purpose

- **Product protection**
- **Personal protection**
- **Environmental protection**





Biological Safety Cabinets

Types

A. Class I

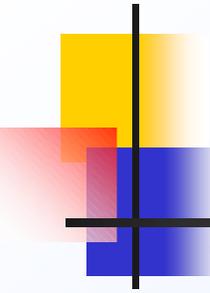
- *inward airflow protects worker*
- *exhaust to outside (w/wo HEPA filter)*

B. Class II

- *worker, product, environmental protection*
- *“sterile” work area*
- *use for work with aerosol-transmissible micro-organisms*
- *use also for tissue culture/ virology*

C. Class III

- *totally enclosed, ventilated, air-tight*
- *suitable for work with BSL3/4 agents*

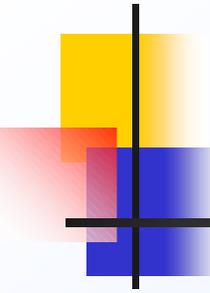


Biological Safety Cabinets

Types

Class II

- *Type A* *30% exhausted to room*
- *Type B3* *30% exhausted to outside*
- *Type B1* *70% exhausted to outside*
- *Type B2* *100% exhausted to outside*



Biological Safety Cabinets

Component

HEPA Filter

- *“High efficiency particulate air” filter*
- *Traps particulates **only**; chemicals, fumes, vapors pass through*
- *Traps particulates 0.3u*

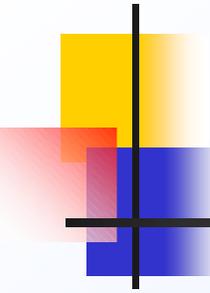
Biological Safety Cabinets

Component

HEPA Filter

- *Metal or wood framed*
- *Continuous sheet of flat filter medium with aluminum separators*
- *Gasket sealed*
- *Adhesive bond between filter pack and frame*





Biological Safety Cabinets

Operating Location

- **Isolated from other work areas**
- **Removed from high traffic areas**
- **Away from airflow ducts**
- **Away from laboratory entry doors**

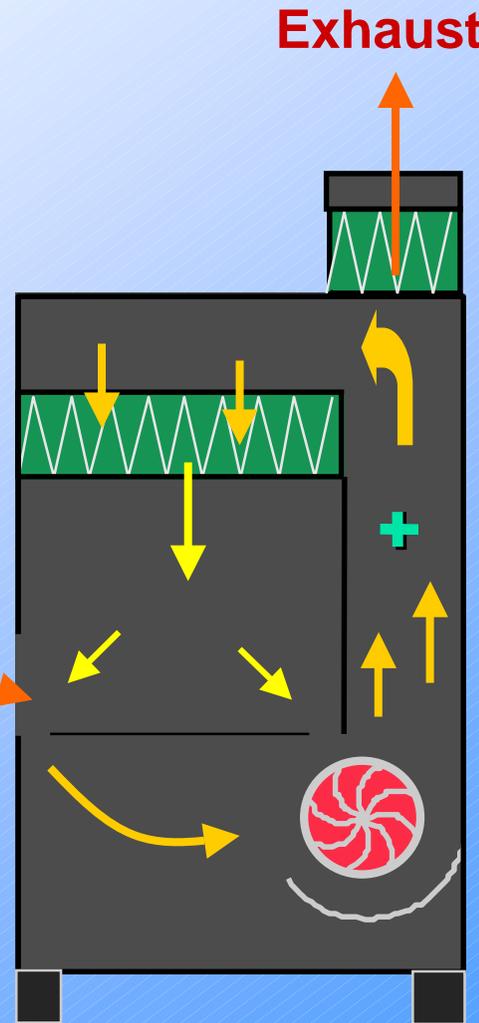
Biological Safety Cabinets

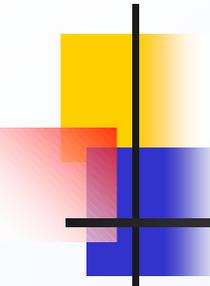
Airflow

Typical Class II



Intake
100
ft/min

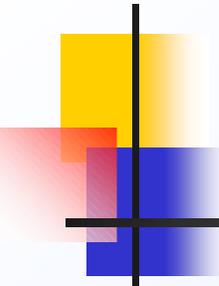




Biological Safety Cabinets

Operating Procedure

- 1. Load BSC with all needed supplies.**
- 2. Turn BSC on and allow to run for 10-15 minutes.**
- 3. Check inward airflow with a piece of tissue.**
- 4. Enter straight into cabinet and perform work in a slow, methodical manner.**
- 5. At end of work, decontaminate all items to be taken out of cabinet.**
- 6. Decontaminate interior of BSC.**
- 7. Allow cabinet to run for 10-15 minutes.**
- 8. Shut off.**



Biological Safety Cabinets

Safe Operation

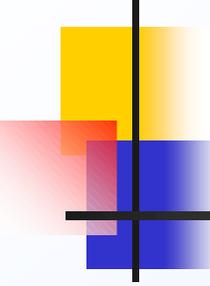
- **Always enter straight into cabinet - no sweeping motions**
- **Place materials well within the cabinet - not on front grill**
- **Place discard pan within cabinet**
- **Watch for disruptions of laminar air flow**
- **Decontaminate materials before removal from cabinet**



Biological Safety Cabinets

Safe Operation

- **Not designed for chemical use**
- **May use for non-volatile toxic chemicals or low-level radioactive materials**
- **May use for “minute” amounts of volatile chemicals**
- **Ensure annual certification**
- **Place all work materials into cabinet before starting**

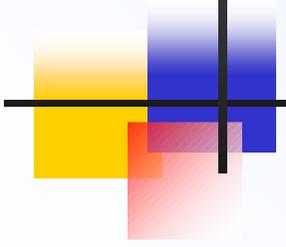


Biological Safety Cabinets

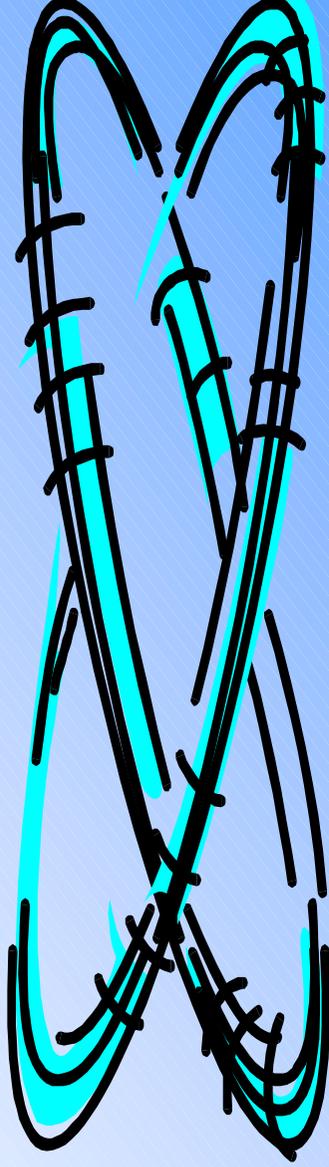
Safe Operation

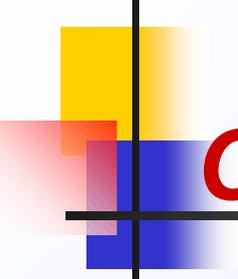
CAUTIONS

- ***Chemicals may damage HEPA filter***
 - **Exposure risk - chemical/infectious agents**
- ***Volatile chemicals NOT retained by HEPA filter***
 - **Exposes personnel if not exhausted**
- ***BSC fans NOT spark proof***
 - **Chemical use may result in fire/ explosion**
 - **Never use NFPA 4 flammables**

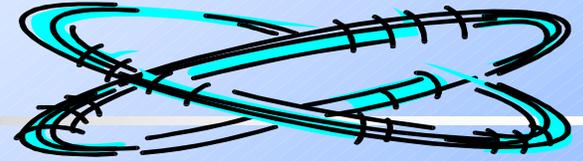


Centrifuges





Centrifuges



Types

Speeds (rpm)

Microcentrifuges

~15,000

Low/high speed

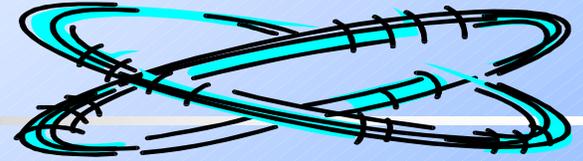
2,000 – 20,000

Ultracentrifuges

~ 120,000

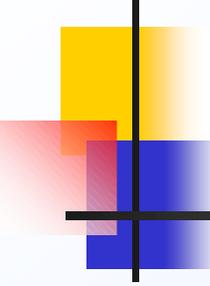
Centrifuges

Hazards



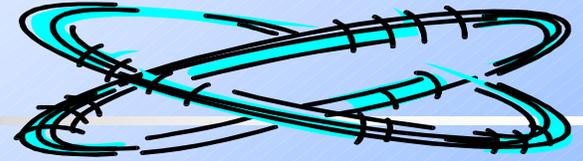
- **Mechanical failure of machine**
- **Lab equipment failure (tubes etc.)**
- **Aerosol generation**
- **Operator error**



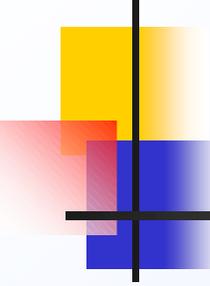


Centrifuges

Operating Procedure

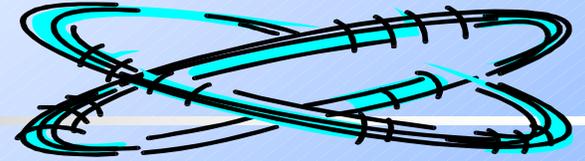


- 1. Check tubes for cracks/chips.**
- 2. Use matched sets of tubes, buckets etc.**
- 3. Tightly seal all tubes and safety cups.**
- 4. Ensure that rotor is locked to spindle and bucket seated.**
- 5. Close lid during operation.**
- 6. Allow to come to complete stop before opening.**



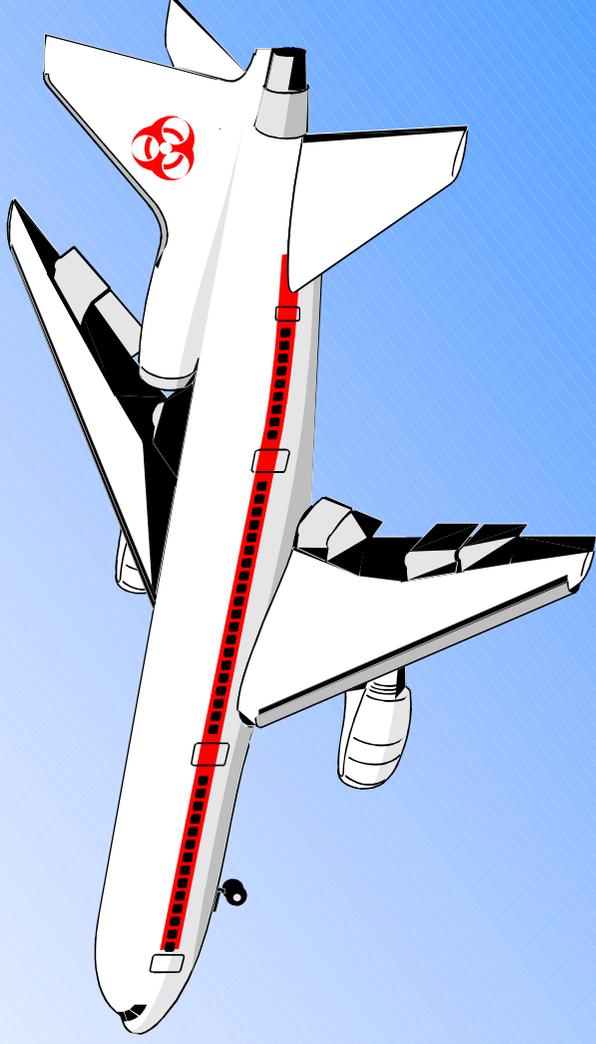
Centrifuges

Safe Operation



- Use safety cups whenever possible
- Disinfect weekly and after all spills or breakage's
- Lubricate O-rings and rotor threads weekly
- Do not use rotors that have been dropped
- Contact your centrifuge rep for specific information

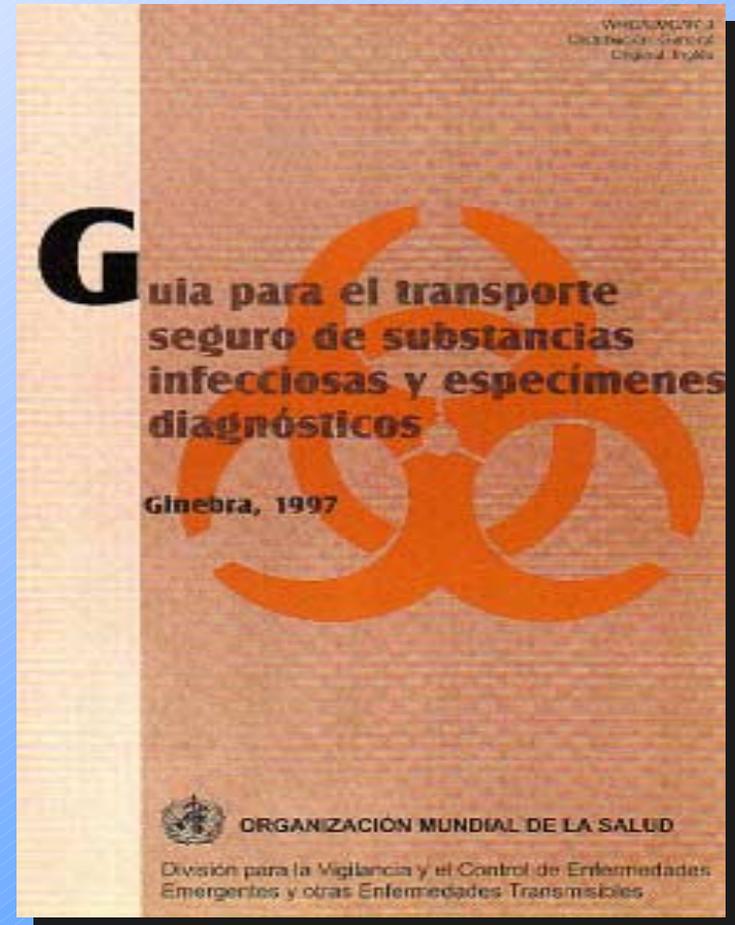
Shipping Biological Specimens



Shipping Biological Specimens

Guideline Documents

- **Recommendations of the United Nations Committee on Dangerous Goods**

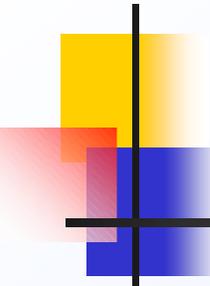


Shipping Biological Specimens

Regulations

- **PHS:** 42 CFR Part 72.
- **DOT:** 49 CFR Part 171-178
- **USPS:** Domestic Mail Manual
- **IATA:** International Air Transport Association
- **ICAO:** International Civil Aviation Organization





Shipping Biological Specimens

Infectious Substance

Definition

- **Contains or has high probability of containing an infectious material...known or reasonably believed to cause disease in humans or animals**
 - *virus, prion, genetic elements*
 - *bacterium, rickettsia, parasite, fungus*
- **Contains a microbial toxin known to be pathogenic**

Shipping Biological Specimens

Infectious Substance

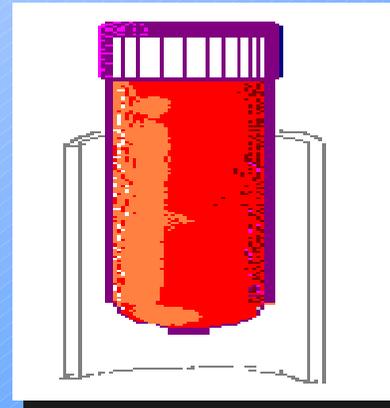
Packaging

- **Primary Container**

- *Positive seal*



- **Absorbent material**

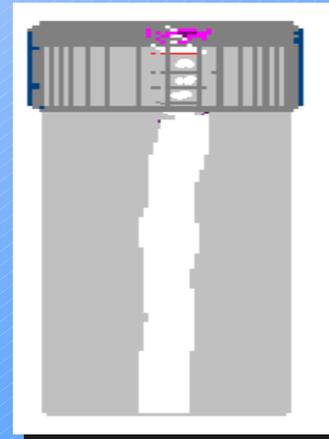
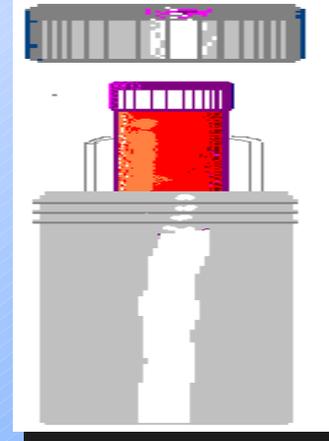


Shipping Biological Specimens

Infectious Substance

Packaging

- Secondary packaging
- Watertight/leakproof

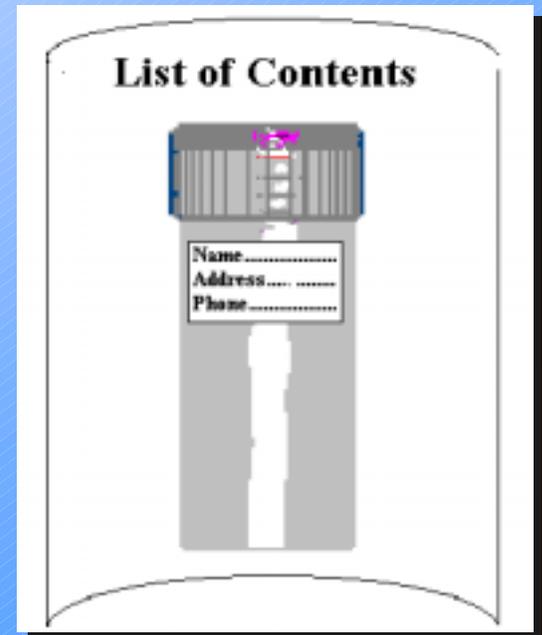


Shipping Biological Specimens

Infectious Substance

Packaging

- **Between Secondary and Outer Container**
 - *List of Contents*
 - *Shippers label*
 - Name
 - Address
 - Phone number

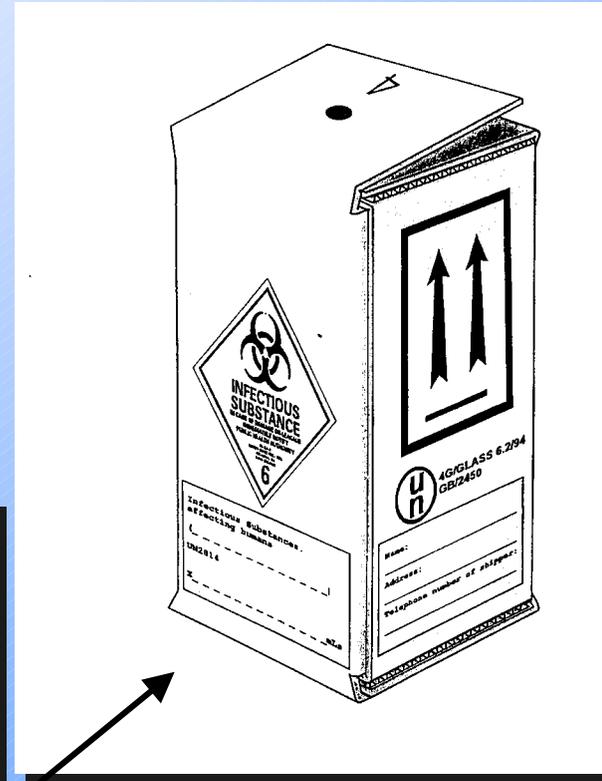


Shipping Biological Specimens

Infectious Substance

Packaging

■ Outer container



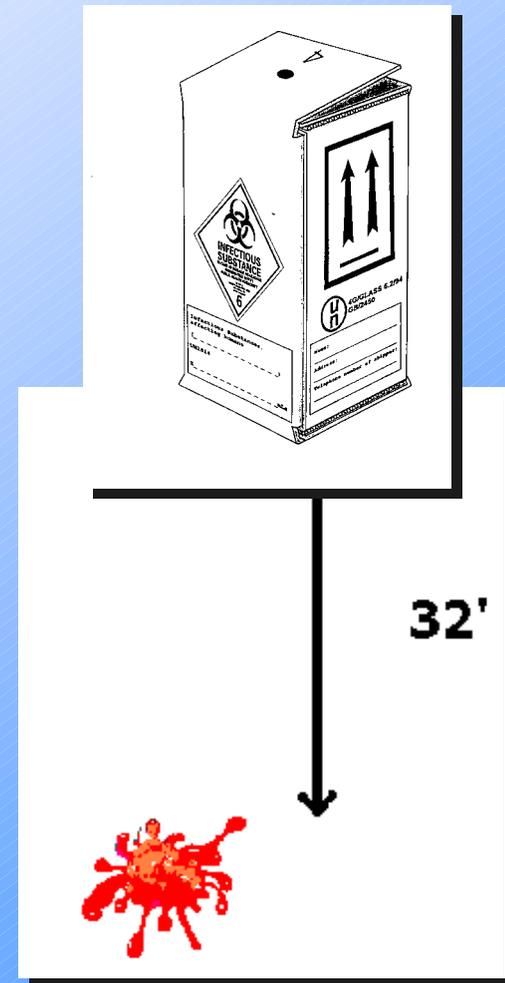
Shipping Biological Specimens

Infectious Substance

Packaging

- Performance tests
 - 49 CFR 178.609

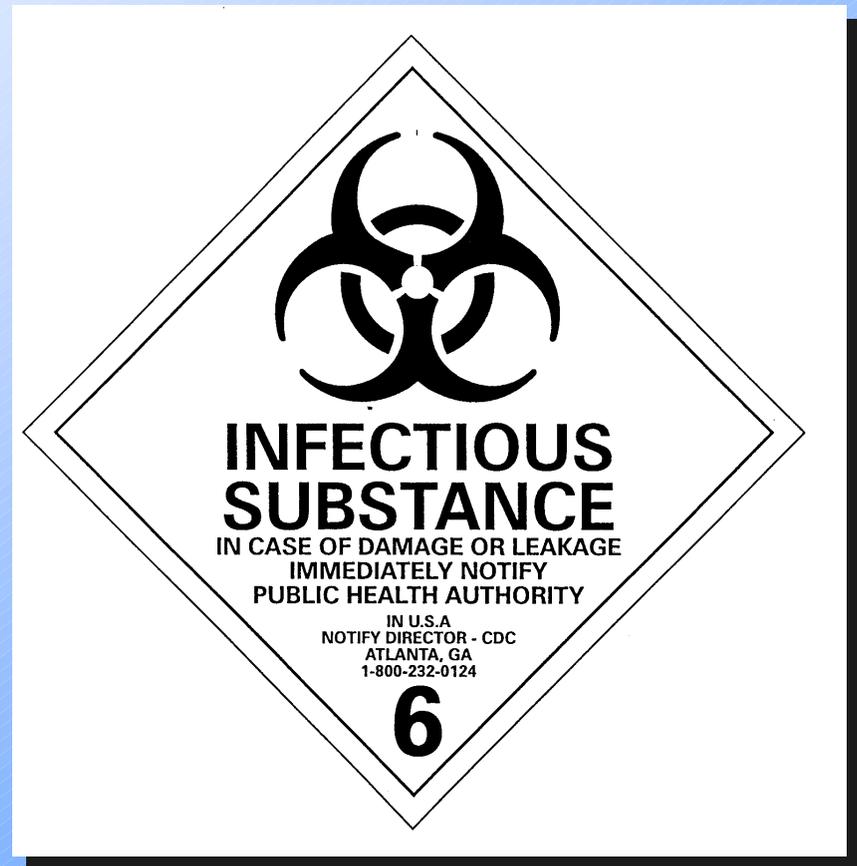
Package shall not leak



Shipping Biological Specimens

Infectious Substance

Packaging label



Shipping Biological Specimens

Infectious Substance



Shipping Biological Specimens

Clinical Specimen

Definition

Human or animal material...collected for the purpose of diagnosis or research....not known to contain viable infectious agents

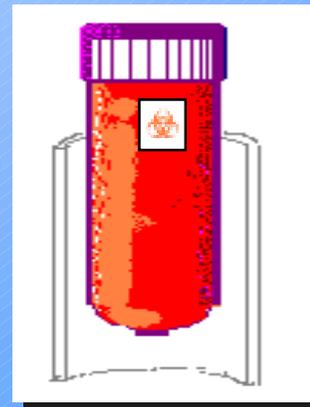
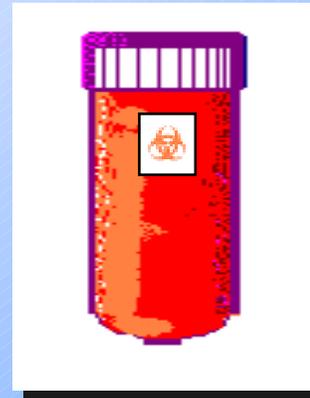


Shipping Biological Specimens

Clinical Specimen

Packaging

- Primary receptacle
- Positive seal
- Biohazard label
- Absorbent material



Shipping Biological Specimens

Clinical Specimen

Packaging

- **Between the secondary and outer packaging**
 - *List of contents*



Shipping Biological Specimens

Clinical Specimen

Packaging

- Outer packaging



Shipping Biological Specimens

Clinical Specimen

Package Label



CLINICAL SPECIMENS

BIOHAZARD

Packaged in Compliance with 42 CFR Part 72

**IN CASE OF DAMAGE
OR LEAKAGE, NOTIFY
CENTERS FOR DISEASE CONTROL
(404) 633-5313**

Standard Form 420 A-1 [8-89]
Prescribed by Dept. HHS [42 CFR 72]
420-301

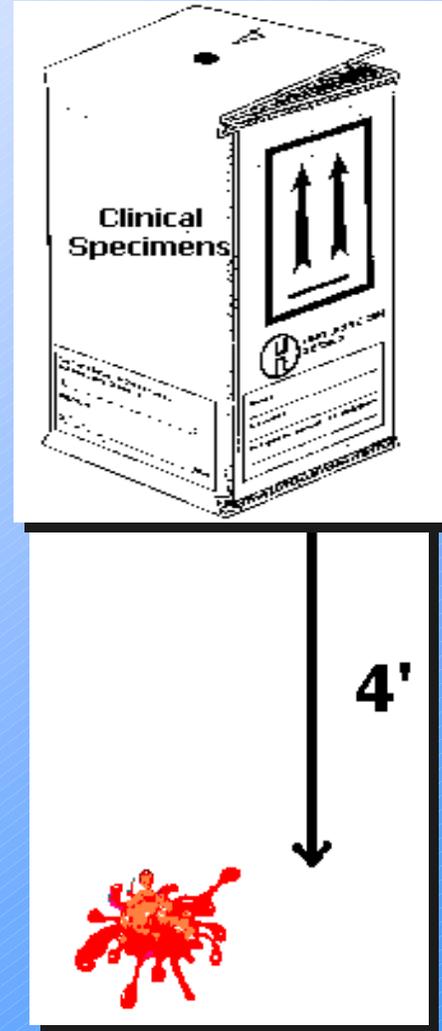
Shipping Biological Specimens

Clinical Specimen

Packaging

- Performance test

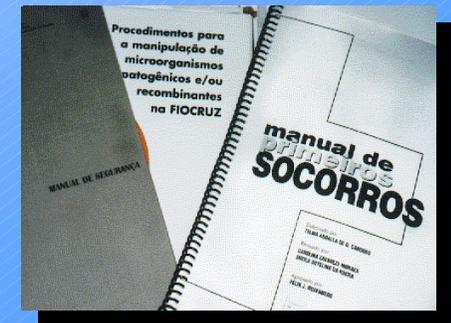
Package shall not leak



Biosafety Manuals

Components

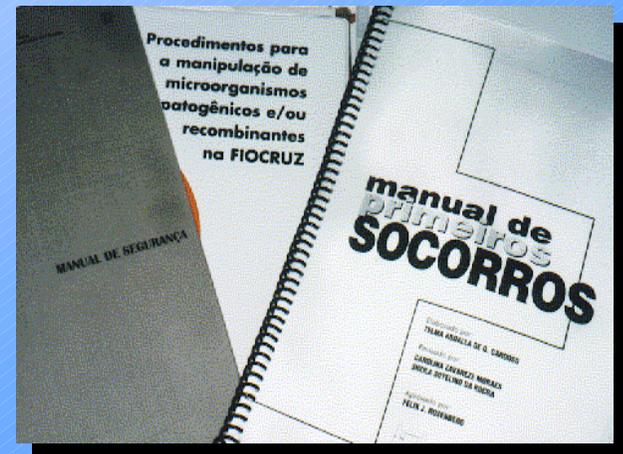
- **Biosafety Level Descriptions**
 - *Standard Practices & Principles*
 - *Special Practices & Procedures*
 - *Containment Devices*
 - *Facility Design*
- **Animal Safety Practices**
- **Agent Summary Statements**



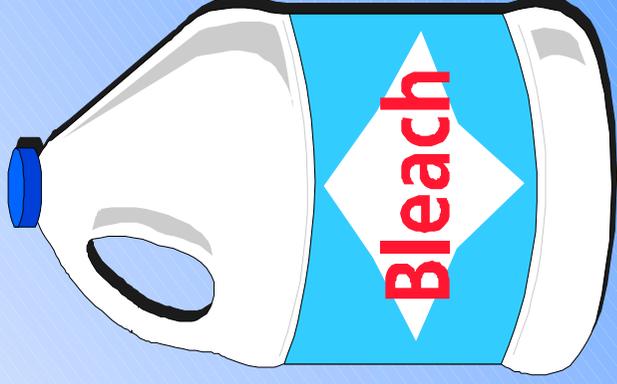
Biosafety Manuals

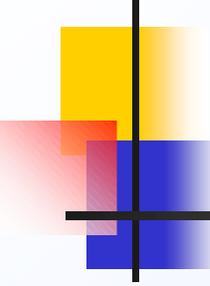
Components

- **Equipment Descriptions**
- **Specimen Handling**
- **Security**
- **Waste**
- **Special Lab Practices**
 - *Tissue culture*
 - *Toxins*



Decontamination



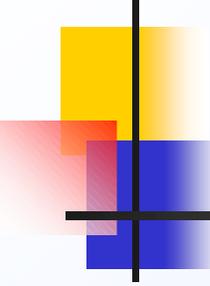


Decontamination

Definition

■ Sterilization

The use of a physical or chemical procedure to destroy all microbial life, including large numbers of highly resistant bacterial spores.

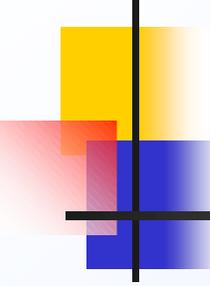


Decontamination

Definition

■ Disinfection

The use of a physical or chemical procedure to virtually eliminate all recognized pathogenic microorganisms but not all microbial forms (bacterial endospores) on inanimate objects.

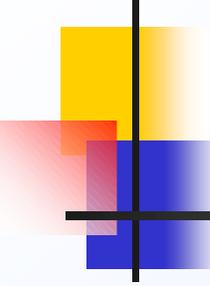


Decontamination

Definition

■ Antisepsis

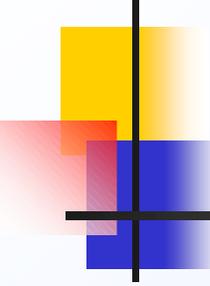
A germicide that is used on skin or living tissue for the purpose of inhibiting or destroying microorganisms.



Decontamination

Agent Selection

- Degree of microbial killing required
- Nature of item/surface to be treated
- Ease of use
- Safety
- Cost



Decontamination

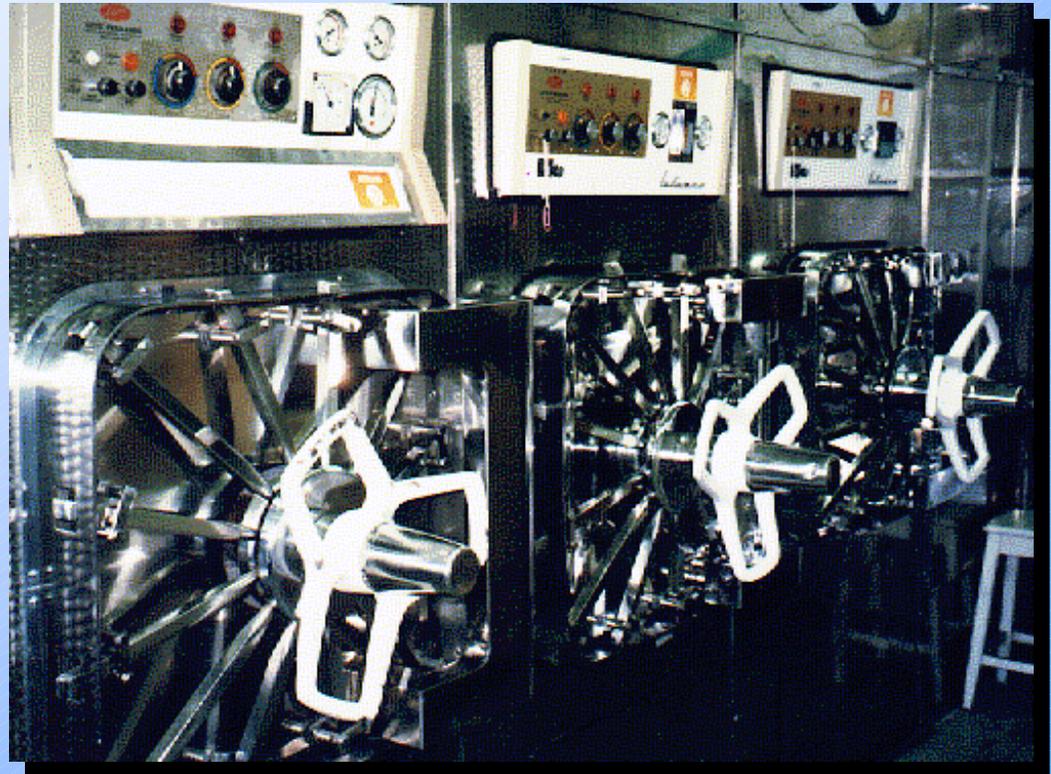
Agent Efficacy

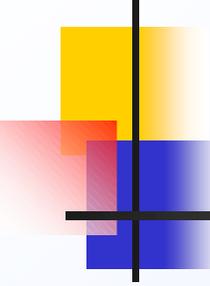
- **Type of organism**
- **Number of organisms**
- **Amount of organic material present**
- **Type & configuration of material to be treated**
- **Type & concentration of germicide**
- **Time and temperature or exposure**
- **pH**
- **Humidity**

Decontamination

Methods

- Heat
- Chemical
- Radiation





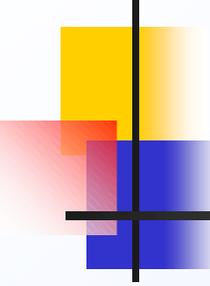
Decontamination

Heat

■ Types

- *Moist – steam*
- *Dry*
- *Incineration*

***The most effective method of sterilization**



Decontamination

Heat

■ Steam sterilization practices

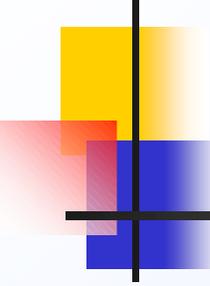
- *Ensure proper functioning of autoclave*
- *Vessels should not be capped or plugged*
- *Large loads require longer contact time*
- *Excessive amounts of liquid should not be added to load*

Decontamination

Heat

- **Steam sterilization verification**
 - *Direct assay*
 - *Thermocouples*
 - *Chemical indicators*
 - *Biological indicators*
*(Bacillus
stearothermophilis)*





Decontamination

Heat

■ Dry heat sterilization

- *Denaturation of proteins: 160⁰ – 170⁰ C/2-4 hours*
- *Effective on impervious non-organic materials like glass*

Decontamination

Heat

■ Incineration

- *Method of choice for animal carcasses*
- *Requires certified incinerator*

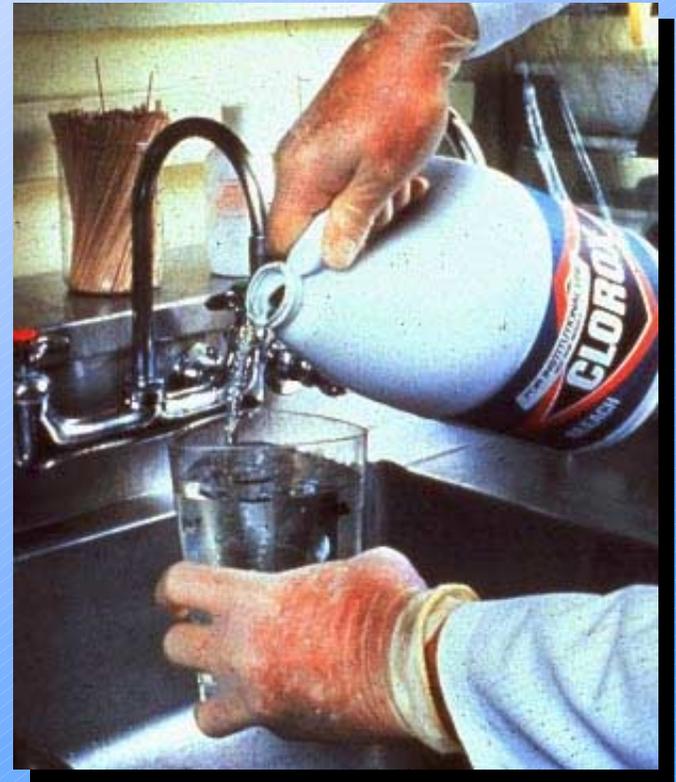


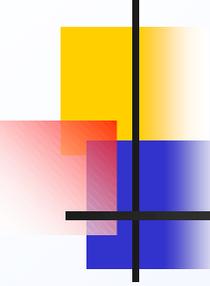
Decontamination

Chemical

■ **Types**

- *Liquids, i.e. chlorox, hydrogen peroxide*
- *Gases, i.e. ethylene oxide*

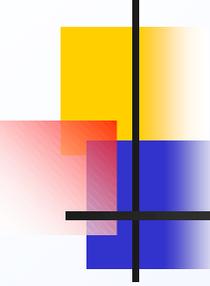




Decontamination

Chemical

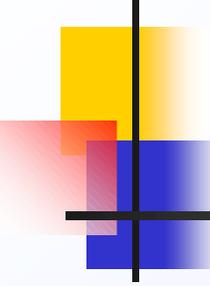
- **Agent selection - complexity**
 - *Over 14,000 registered products*
 - *Over 300 active ingredients*
 - *14 ingredients present in 92% of products*



Decontamination

Chemical

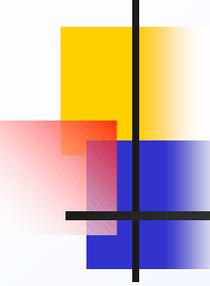
- **Agent selection - activity**
 - *HLD – high level disinfection*
 - *ILD – intermediate level disinfection*
 - *LLD – low level disinfection*



Decontamination

Chemical

- **High level disinfection - sporocides**
 - *Kills all microorganisms except high numbers of bacterial spores*
 - *Require 5-10 min. exposure*
 - *Examples: aldehydes, hydrogen peroxide, paracetic acid*

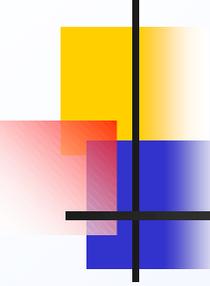


Decontamination

Chemical

■ Intermediate level disinfection - tuberculocides

- *Kills M. tuberculosis var. bovis and all vegetative bacteria, fungi, and most viruses*
- *Require minimum 20 min. exposure*
- *Examples: phenolics, iodophores, chlorine compounds, alcohols*



Decontamination

Chemical

- **Low level disinfection – hospital germicides used for housekeeping**
 - *Kills most vegetative bacteria and some fungi, but not M. tuberculosis var. bovis*
 - *Require minimum 20 min. exposure*
 - *Examples: quaternary ammonium compounds*

Decontamination

Summary

Bacterial Spores

B. subtilis

Mycobacterium

MTB var. bovis

Non-lipid Viruses

Polio-

Rhino-

Fungi

Cryptococcus sp,

Candida sp.

Vegatative Bacteria

Pseudomonas sp.

Staphylococcus sp.

Salmonella sp.

Lipid Viruses

Herpes

CMV

HBV

HIV

Sterilization

HLD

ILD

LLD



Decontamination

Chemical

- **General Lab Use - *Hypochlorite Solutions***
 - *Large Spills/Large Organic Load*
 - undiluted from bottle
 - *Small Spills/Virus Inactivation*
 - 10% - 1:9
 - *General Surface Disinfection*
 - 1% - 1:99





Decontamination

**Disinfectants do not replace
standard microbiological
practices or good hygiene!**

Biological Waste





Biological Waste

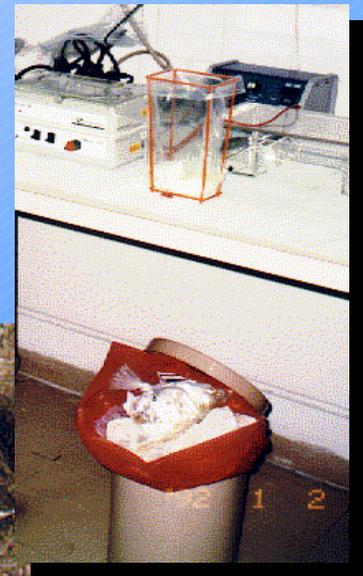
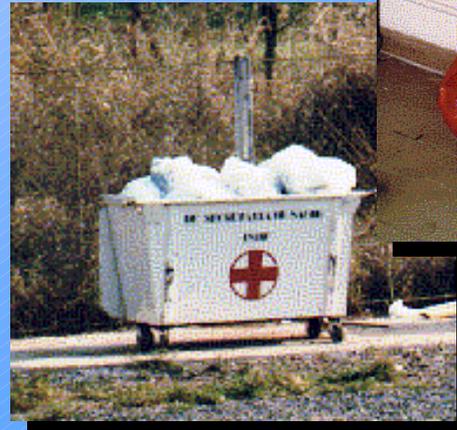
■ Types

- *cultures, stocks, isolates*
- *materials containing or contaminated with blood*
- *sharps*
- *pipettes, wrappers, tips*
- *All materials used in the lab*

Biological Waste

■ Disposal

- *puncture-proof, leak-proof, sealable receptacles*
- *avoid over-filling*
- *dispose properly*



Biological Waste

■ Disposal

- **Never** place lab waste into office waste containers
- Place sharps into “sharps” container
- Line discard containers with autoclave bag
- Decontaminate discard pans before they leave the lab:

1. Disinfect outside
2. Label
3. Tape ends with autoclave
4. Tape
5. Secure for transport to autoclave



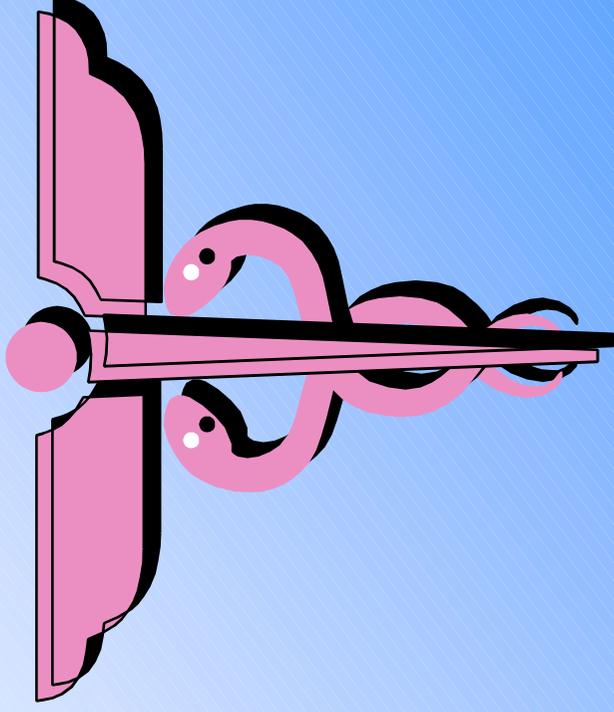


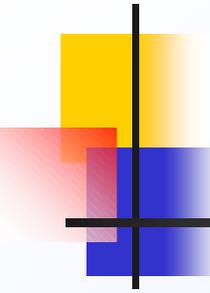
Biological Waste

Decontamination

- **To render the object/material safe by reducing or removing the bioburden**
- **Methods**
 - *chemical ... match, contact time*
 - *physical ... Heat, steam and pressure*
 - *incineration*
 - *other choices, i.e. shredding + chemical*

Medical Surveillance

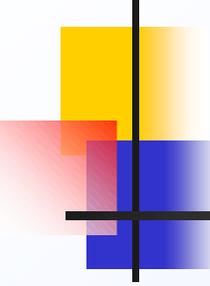




Medical Surveillance

Criteria

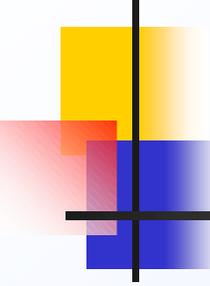
- **Based on risk assessment**
- **Pre-placement**
 - *evaluate physical requirements*
- **Periodic review**



Medical Surveillance

Risk Assessment

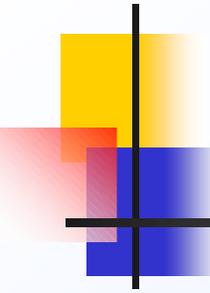
- **The probability of infection**
- **Implies an estimate of numbers exists**
- **Predict an outcome given similar events**



Medical Surveillance

Risk Assessment

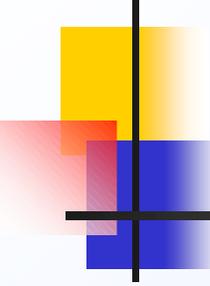
- **What is the natural host?**
- **Does agent cross species barriers?**
- **Wild-type agent or attenuated?**
- **Infectious for normal healthy adult?**
- **What if adult is immunocompromised?**



Medical Surveillance

Risk Assessment

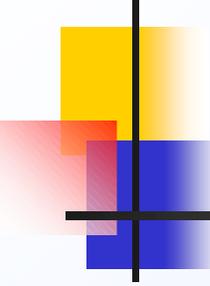
- **Mode of transmission?**
 - *contact*
 - *fomites*
 - *mucous membrane exposure*
 - *ingestion*
 - *inoculation or insect bites*
 - *inhalation*
 - *sex*



Medical Surveillance

Risk Assessment

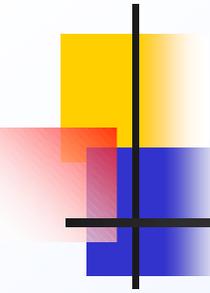
- Volume being manipulated?
- Concentration of agent?
- Infectious dose?
- Past history of lab-associated infection?
- Secondary spread in community?



Medical Surveillance

Risk Assessment

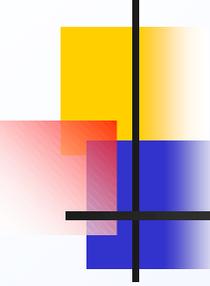
- **Prophylaxis**
 - *Immunizations available?*
 - *Pharmaceuticals?*
 - *Effectiveness?*
- **Post-Exposure**
 - *Anti-microbial agents?*
 - *Pharmaceuticals?*
 - *Effectiveness?*



Medical Surveillance

Risk Assessment

- **Dealing with an unknown agent?**
 - *epidemiological data*
 - *patterns parallel to other agents*
 - *data from animal studies*
 - *route of infection*



Medical Surveillance

Risk Management

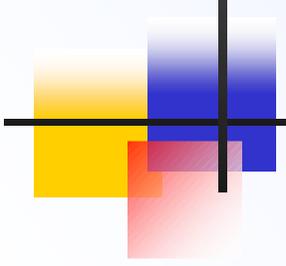
- **Top management**
 - *overall safety policy*
 - *resource allocation*
- **Supervisor**
 - *implement policies*
 - *training, practices & procedures, access*
- **Workers**
 - *strict & rigorous attention to details of practices and procedures*
 - *report incidents and exposures*

Medical Surveillance

Risk Management

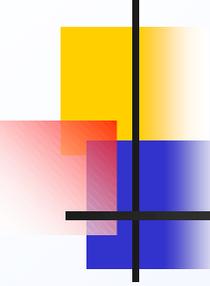
- Occupational Health Clinic
- Immunizations, chemotherapy
- Medical surveillance programs
- Incident (emergency) response
- Incident investigation





Emergency Response





Emergency Response

Personal Contamination

- 1. Alert co-workers**
- 2. Clean exposed surface with soap/water, eyewash (eyes), or saline (mouth)**
- 3. Apply first aid and treat as an emergency**
- 4. Notify supervisor or security desk (after hours)**
- 5. Report to medical clinic for treatment/counseling**



Emergency Response

Surface Contamination

1. Alert co-workers
2. Define/isolate contaminated area
3. Put on appropriate PPE
4. Remove glass/lumps with forceps or scoop
5. Apply absorbent towel(s) to spill; remove bulk & reapply if needed
6. Apply disinfectant to towel surface
- 6. *Allow adequate contact time (20")***
8. Remove towel, mop up; clean with alcohol or soap/water
9. Properly dispose of materials
10. Notify supervisor

