

***Animal Biosafety:
Biosafety Officer's
Experiences and
Perspectives***

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Overview

- Risk Assessment Challenges
- Training Strategies
- Augmenting SOP's

Framework for RA/RM

Risk Assessment	Risk Management (SOP's & Training)
Pathogen	Practices (good work practices)
Procedures (SOP's) (Training)	Protective equipment (clothing and equipment)
Personnel (Training)	Place (facility design)

Risk Assessment Challenges

- Diverse research in shared facilities
- Limited BSL3 facilities for animal studies
 - Aged, older designs
- Different agents = different SOP's
 - West Nile virus, Creutzfeld Jacob agent, Rickettsia conorii, LCMV, HIV, VSV lab strain
- Animal technicians prefer standardized protocol

Exit
(contaminated)
corridor

Shower

Contaminated and Clean Wash Rooms

Pass through
autoclave

7
BSL-3
Rooms

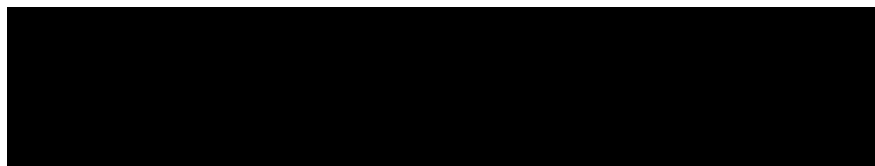
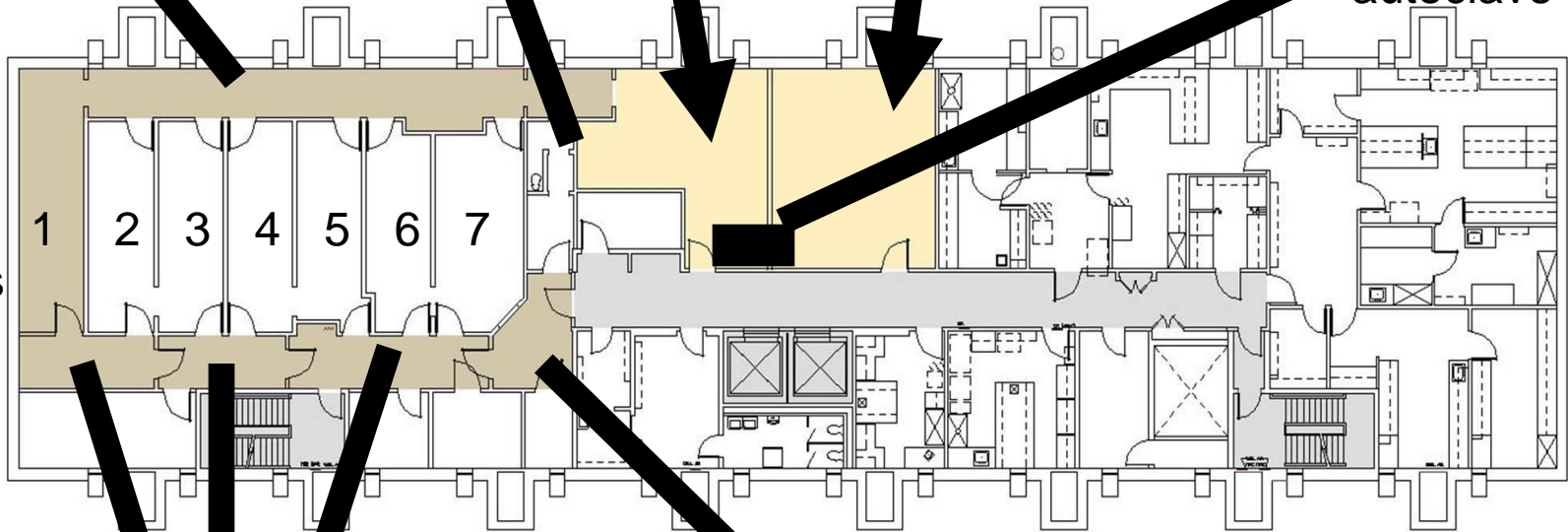
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EXISTING 8th FLOOR PLAN

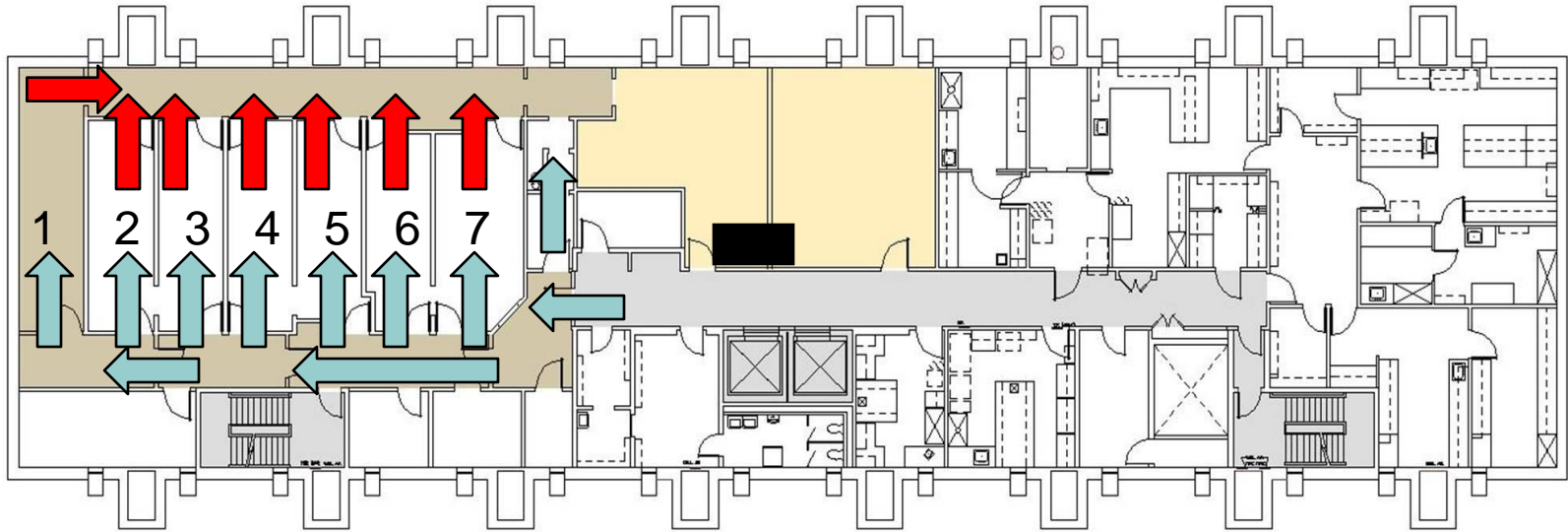
Clean entry corridor

Anterooms

Two Corridor
Containment Lab
Design Scheme

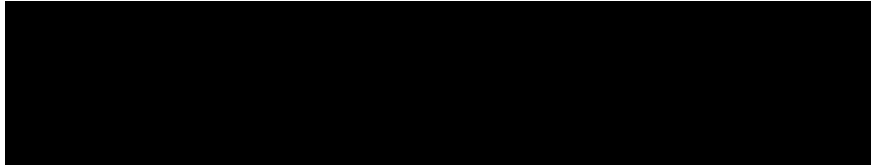


Lab Airflow



EXISTING 8th FLOOR PLAN

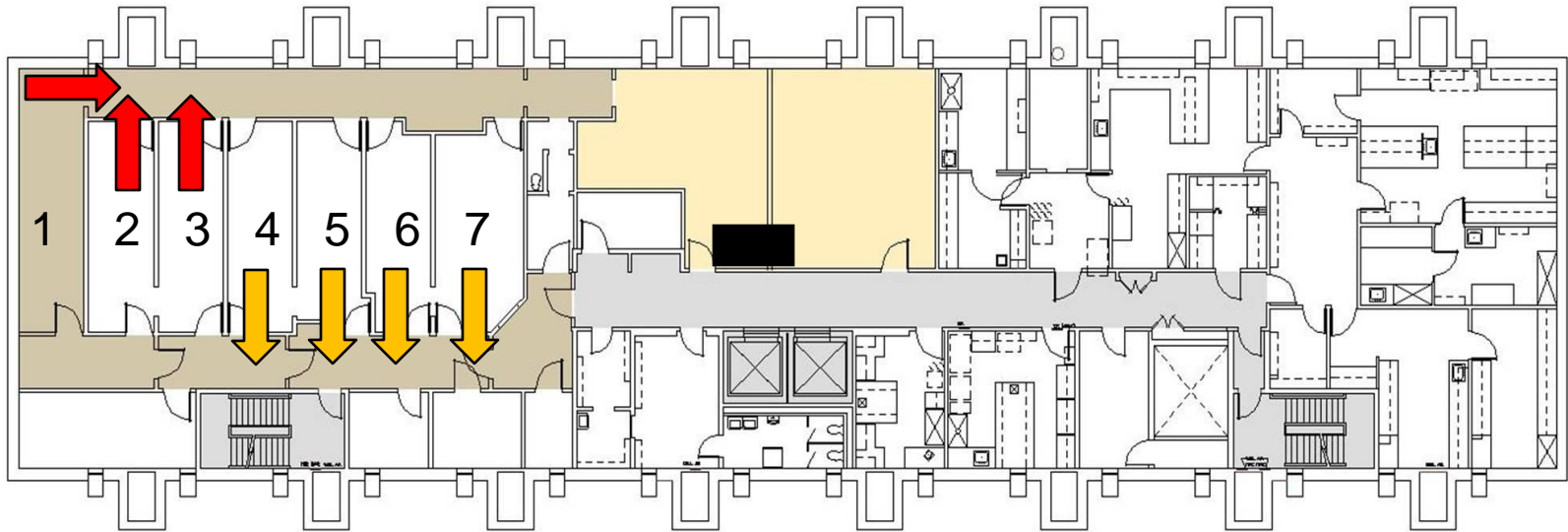
Two Corridor
Containment Lab
Design Scheme



Risk Assessment Challenges

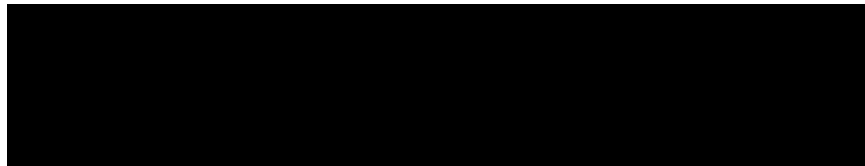
- All animal BSL3 experiments in one facility
- Two “exit” SOP’s agreed upon
 - Exit via contaminated corridor
 - Respiratory protection required
 - Exit back to clean corridor
- “Accepted” by all

Laboratory Exit



EXISTING 8th FLOOR PLAN

Two Corridor
Containment Lab
Design Scheme



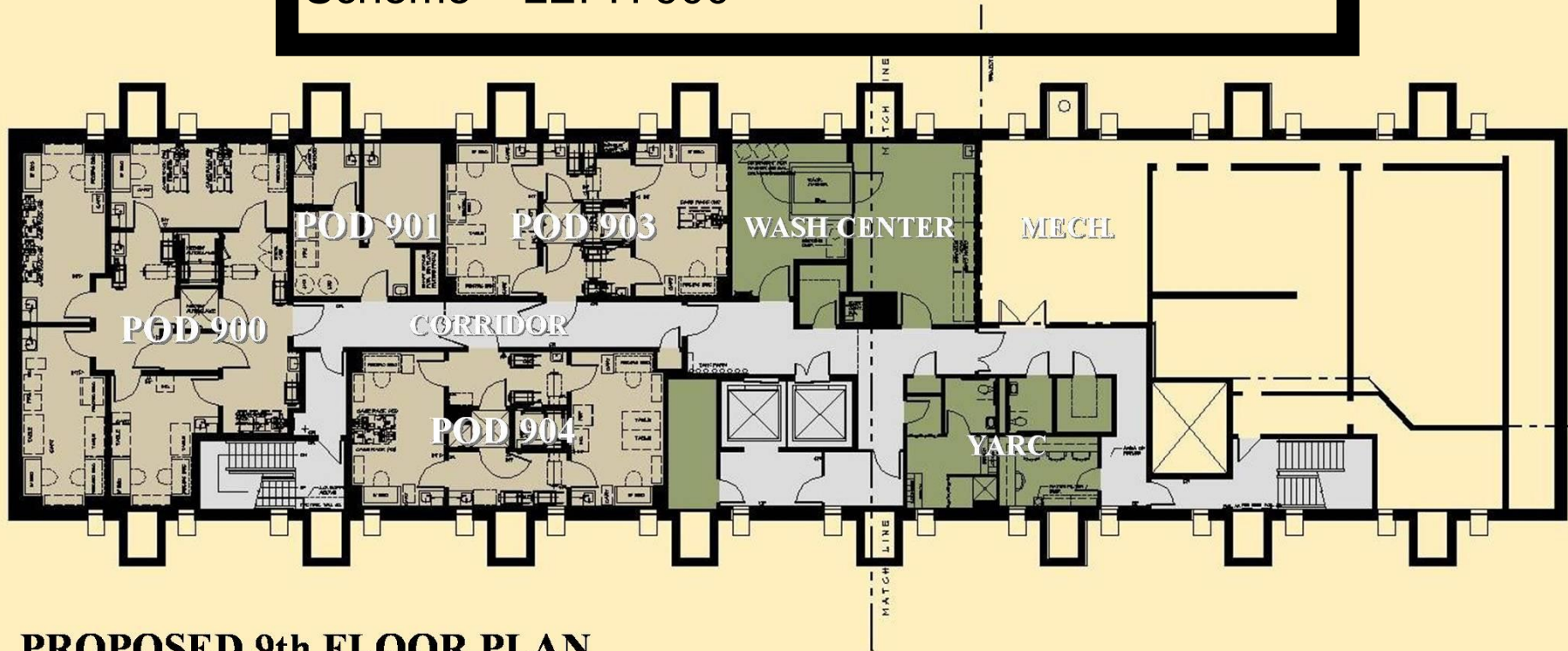
Risk Assessment Challenges

- New biohazard protocols
 - Mycobacterium tuberculosis
 - Highly Pathogenic Avian Influenza virus (H5N1)
- Projected additional requirements
 - Shower out may impact ALL in facility
- Not “accepted”
- Pod concept hatched

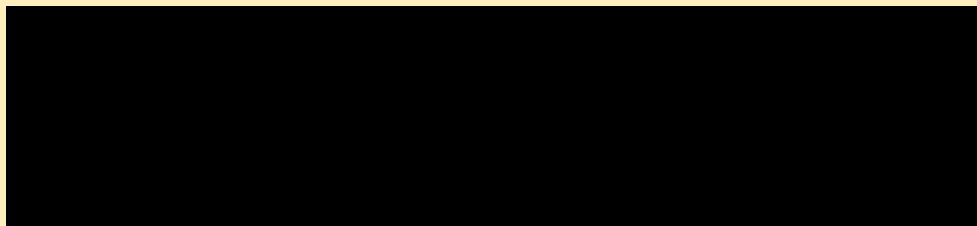
Road to the Pod

- Renovate old animal facility
- Create segregated BSL3/ABSL3 laboratories
 - Each self contained
- Separate high risk and regulated agents
- Group similar risk agents in super pod

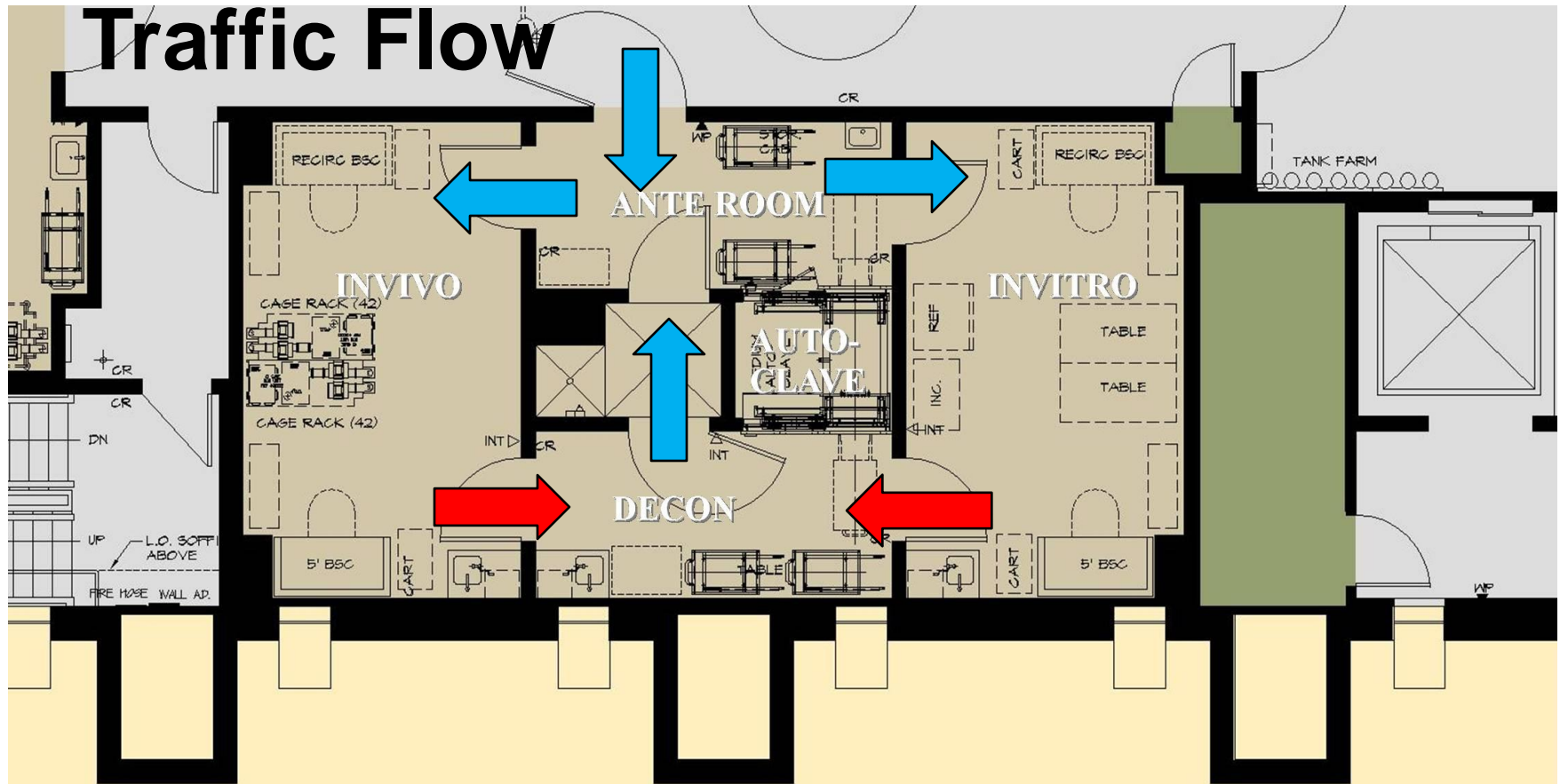
Single Corridor “Segregated” Facility Design Scheme – LEPH 900



PROPOSED 9th FLOOR PLAN

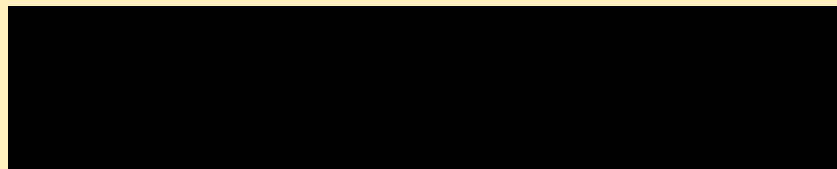


Traffic Flow



Clean
(Anteroom)
and Dirty
(DECON)
Passageways

PROPOSED 9th FLOOR PLAN - Pod 904



One Segregated
BSL3 Suite w/in
BSL3 Facility: Lab
and Animal
Experiments
Combined

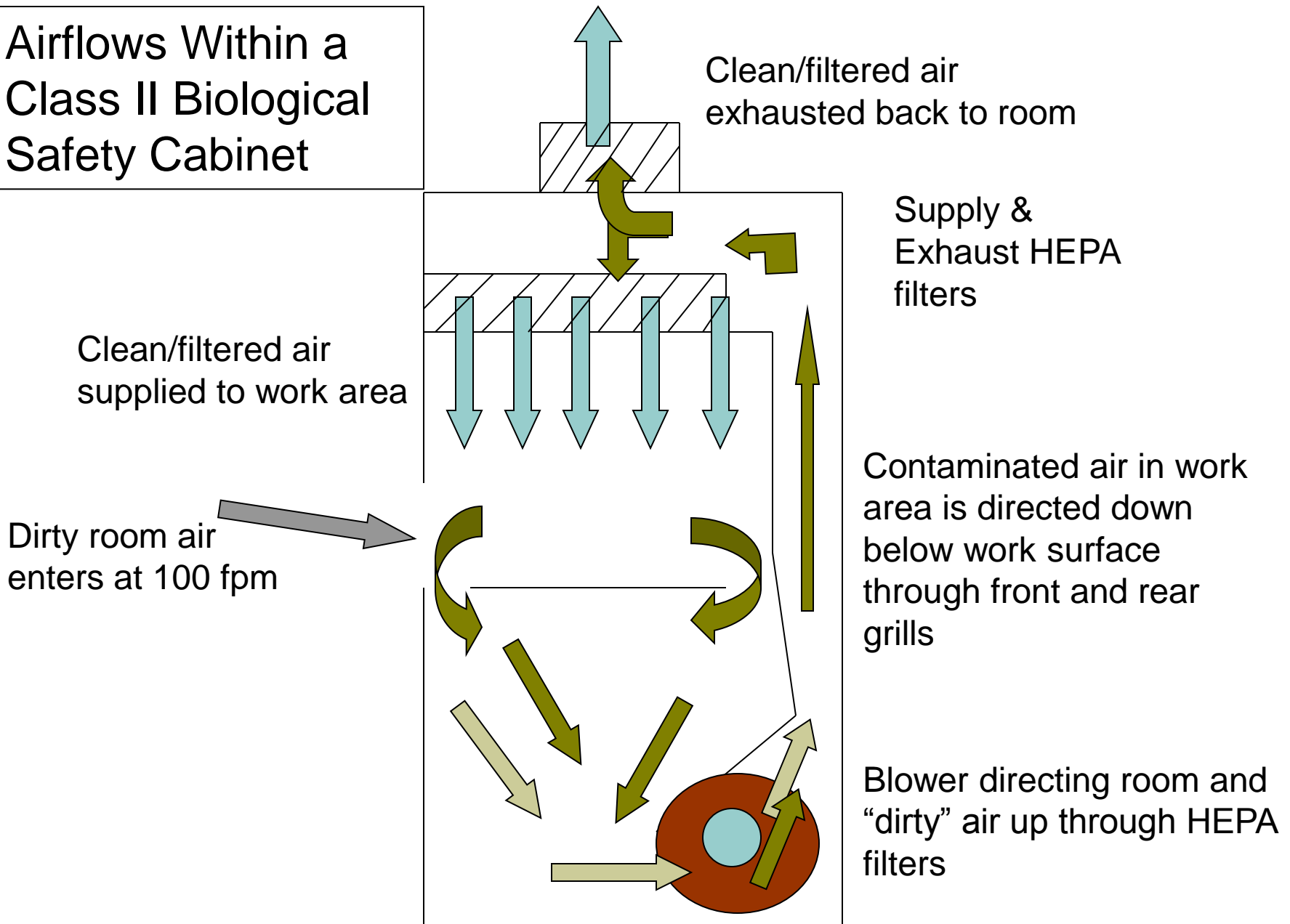
Evolution of Training

- Adult learning techniques incorporated
- More frequent delivery & shorter segments
- Modern techniques required
- Multiple trainers with shared responsibilities

Evolution of BSL3 Training

- Life cycle of BSL3 trainee
 - Discussion of risk → Initial training
 - Facility orientation → SOP walkthrough
 - Shadow authorized BSL3 researchers
 - Observe BSL3 work
 - Perform BSL3 procedures with mock materials

Airflows Within a Class II Biological Safety Cabinet



Getting used to “all work inside the biosafety cabinet”



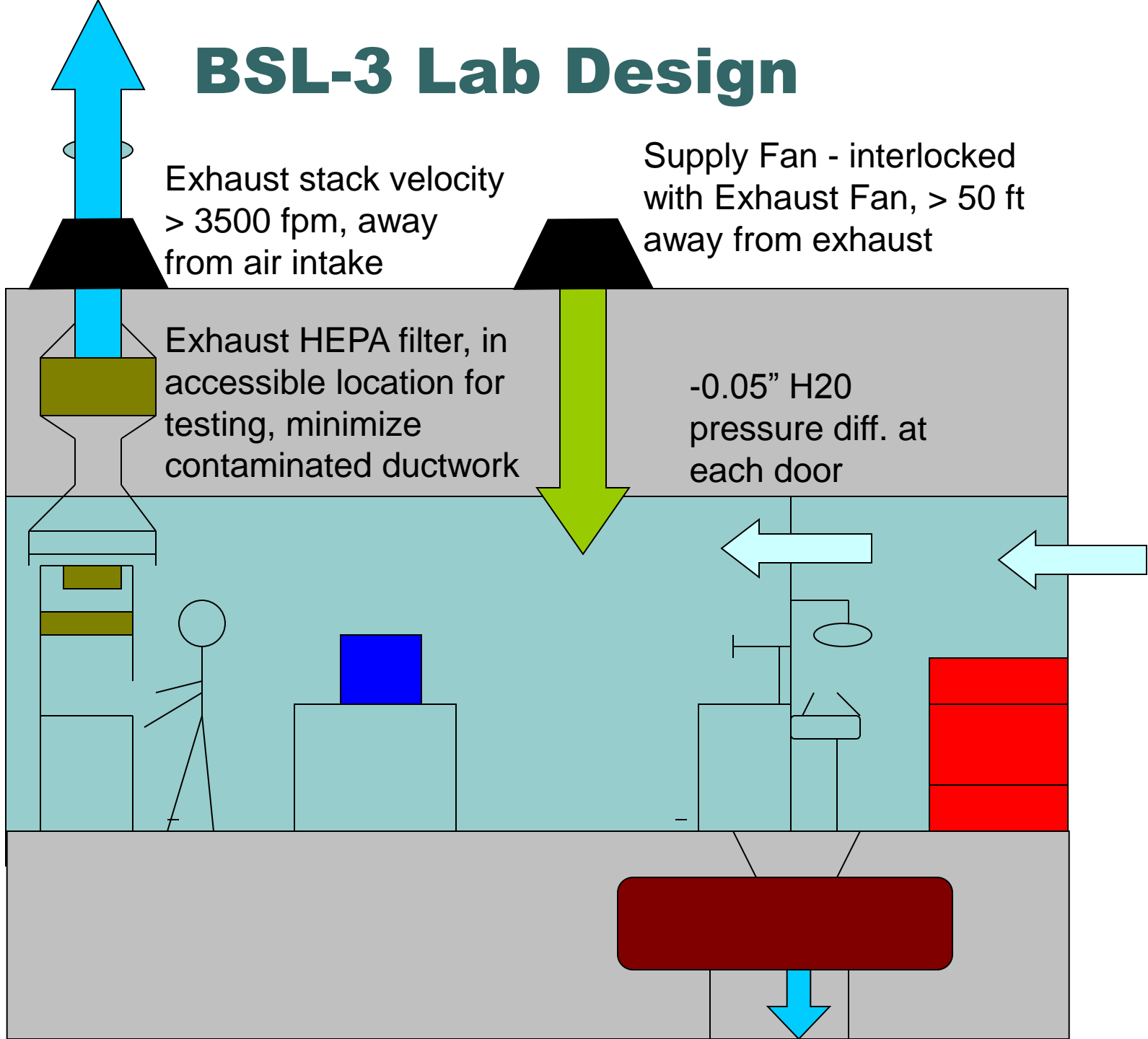
Pre-Occupancy Training

- Risk Awareness
- Understanding containment
 - Facility
 - Equipment
 - Work practices
- Knowledge regulations and site-specific policies
- Incident response
- Tours, classroom and hands-on

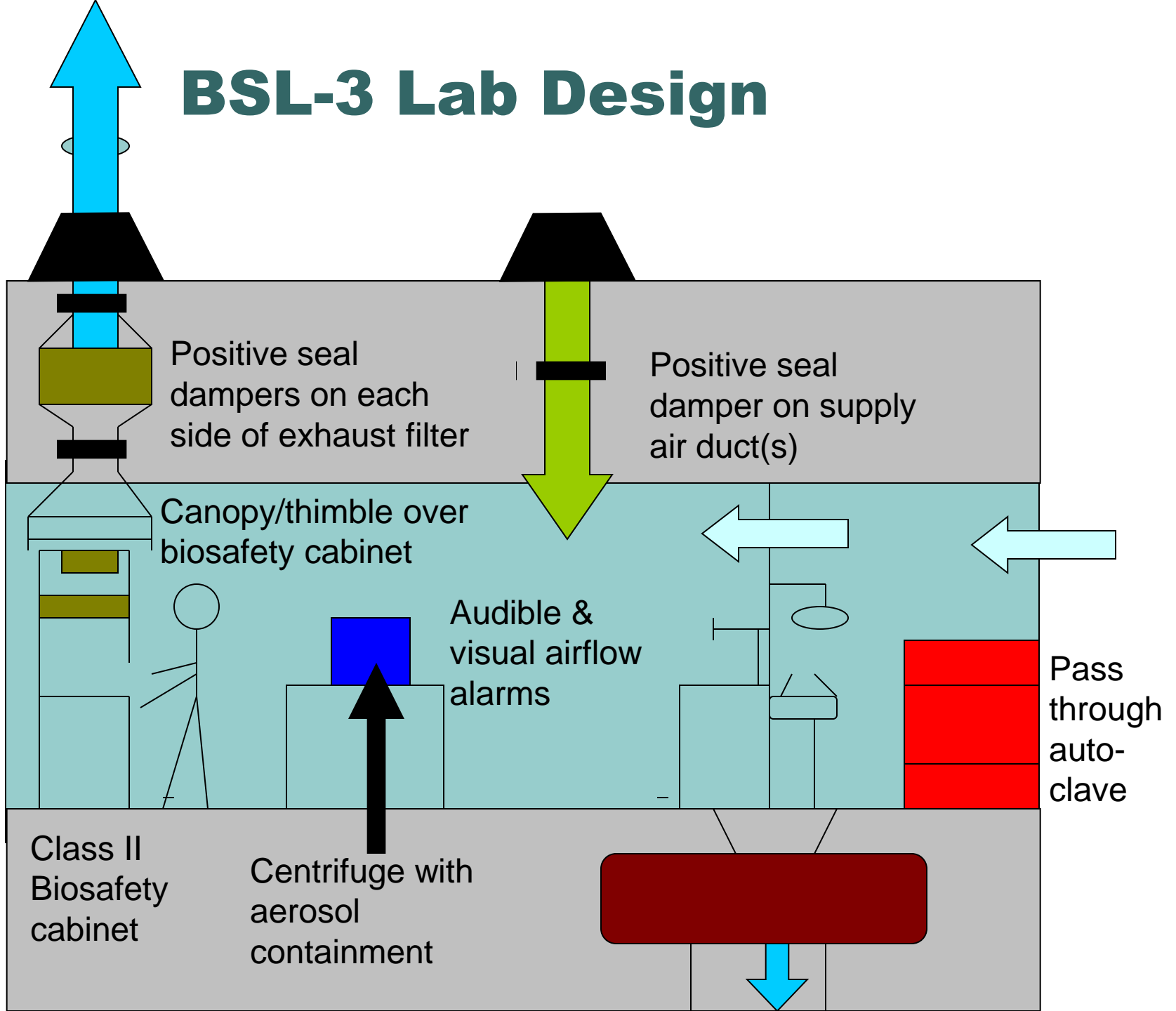
Personnel Training

- Classroom
 - Biosafety, Bloodborne Pathogens, Lab Chemical Safety, BSL-3 Training, Shipping/Transport, Biosecurity, Emergency Response (Drills)
- Tours
 - Orientation covering entry, facility equipment and use, and exit
 - Review of monitors, alarms and contingency plans

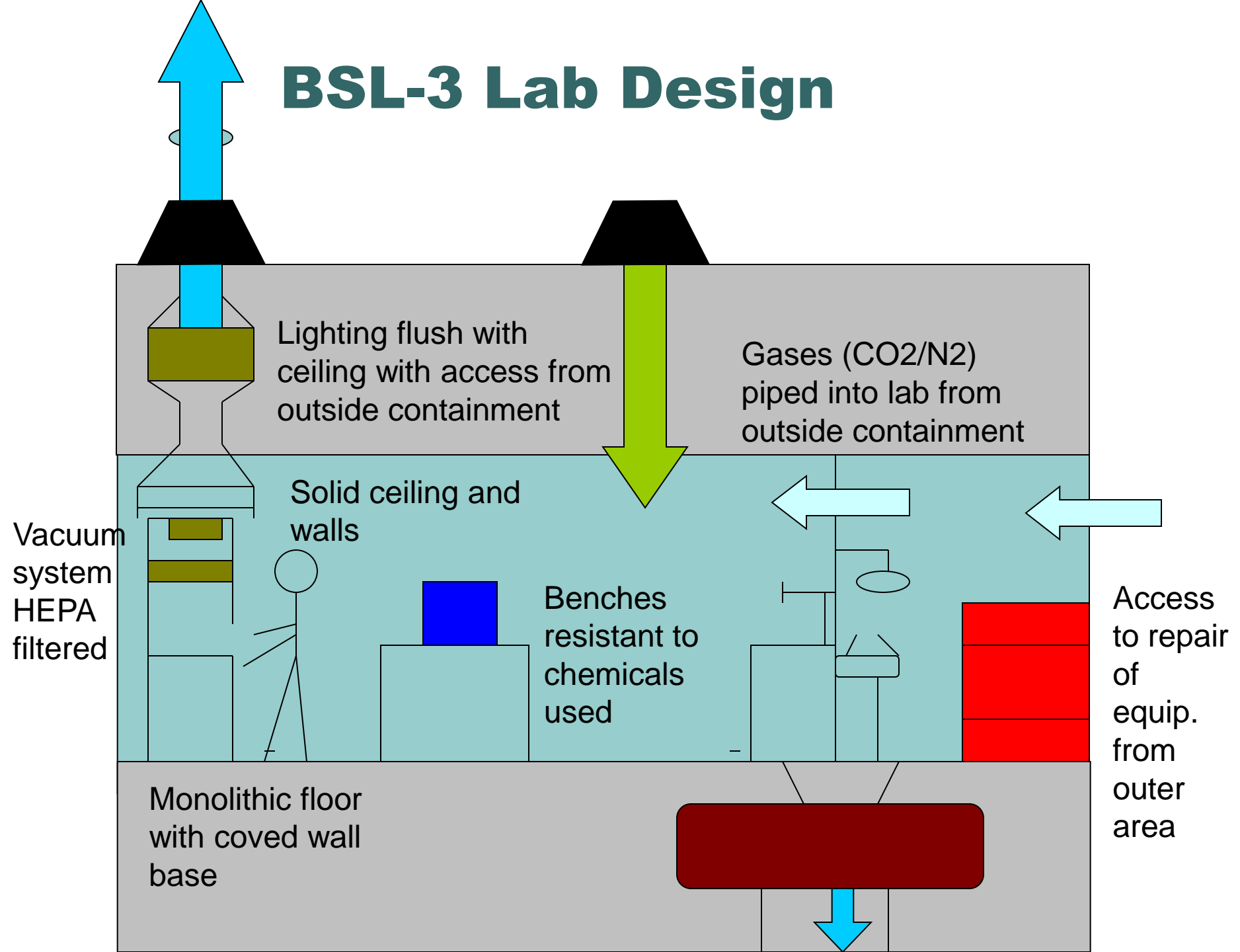
BSL-3 Lab Design



BSL-3 Lab Design



BSL-3 Lab Design



Personnel Training

- Prior work experience
 - 2 months with BSL-2 pathogens
 - 15 hours (15 supervised visits) at BSL-3
 - Separate BSL-3 requirements for lab and animal authorization
 - Relevant education
- Accept training from other institutions, but...
 - Require onsite training/orientation &
 - Successful completion of observation

Training and use of personal protective equipment



Repetition with entry/exit protocols



Mentoring



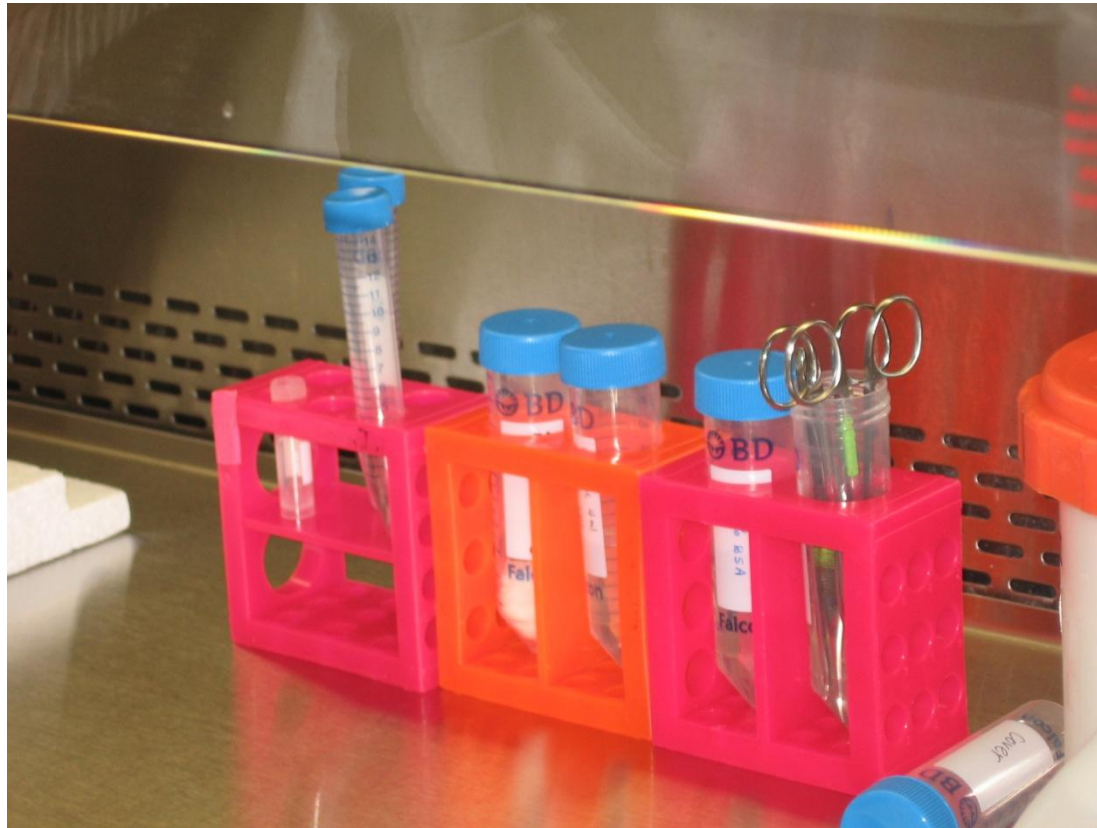
BSL3 Training Life Cycle

- Life cycle (cont.)
 - Perform BSL3 procedures under supervision
 - Gain independent authorization through direct proficiency observation evaluation
 - Transition to becoming a trainer
- Retraining
 - Don't tell trainees what they already know
 - Focus on critical steps that must be addressed

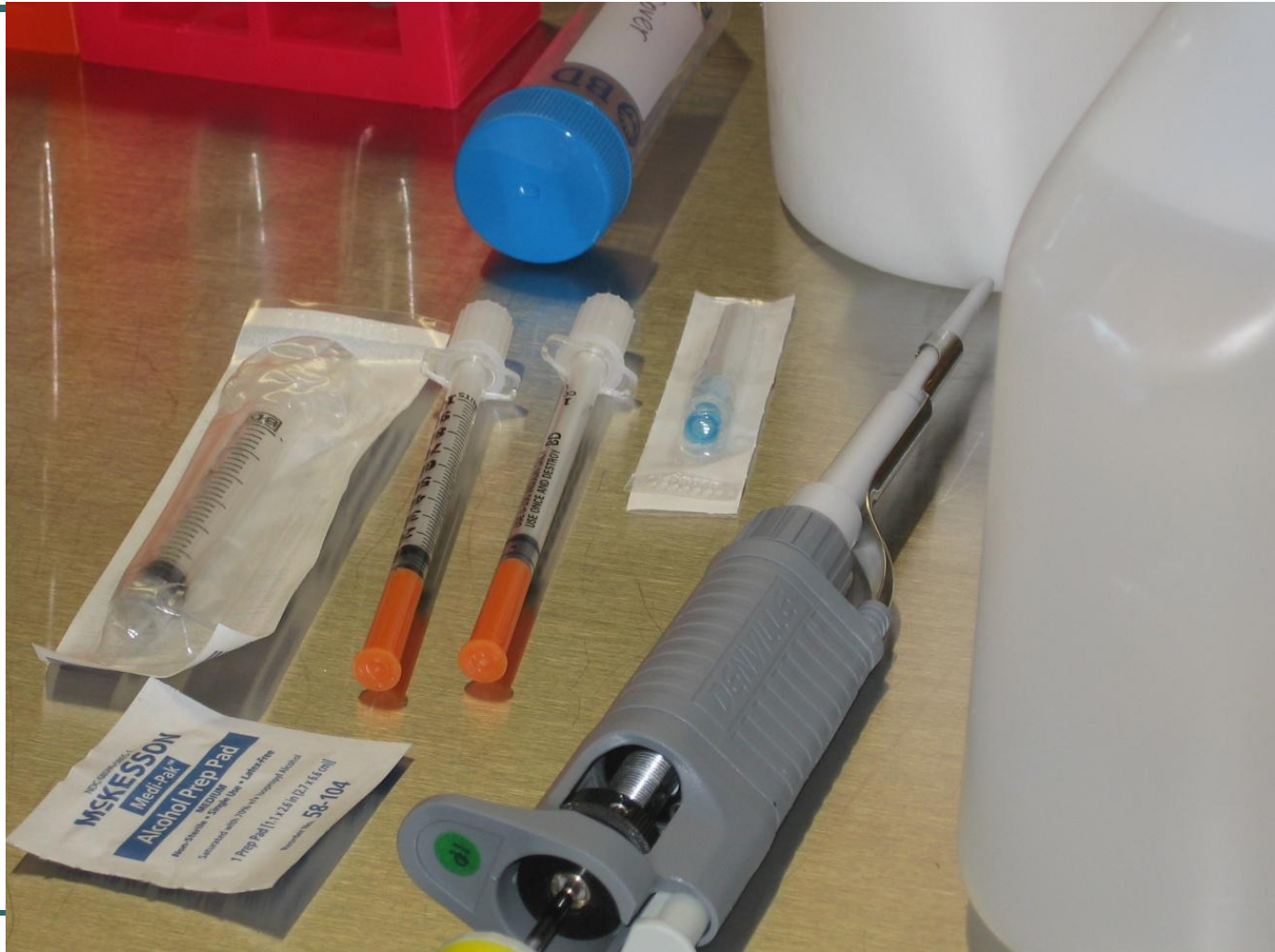
Emergency Intercom to Yale 24/7 Security Dispatch (in each lab)



Position supplies in the work area



Keep items in the biosafety cabinet to a minimum



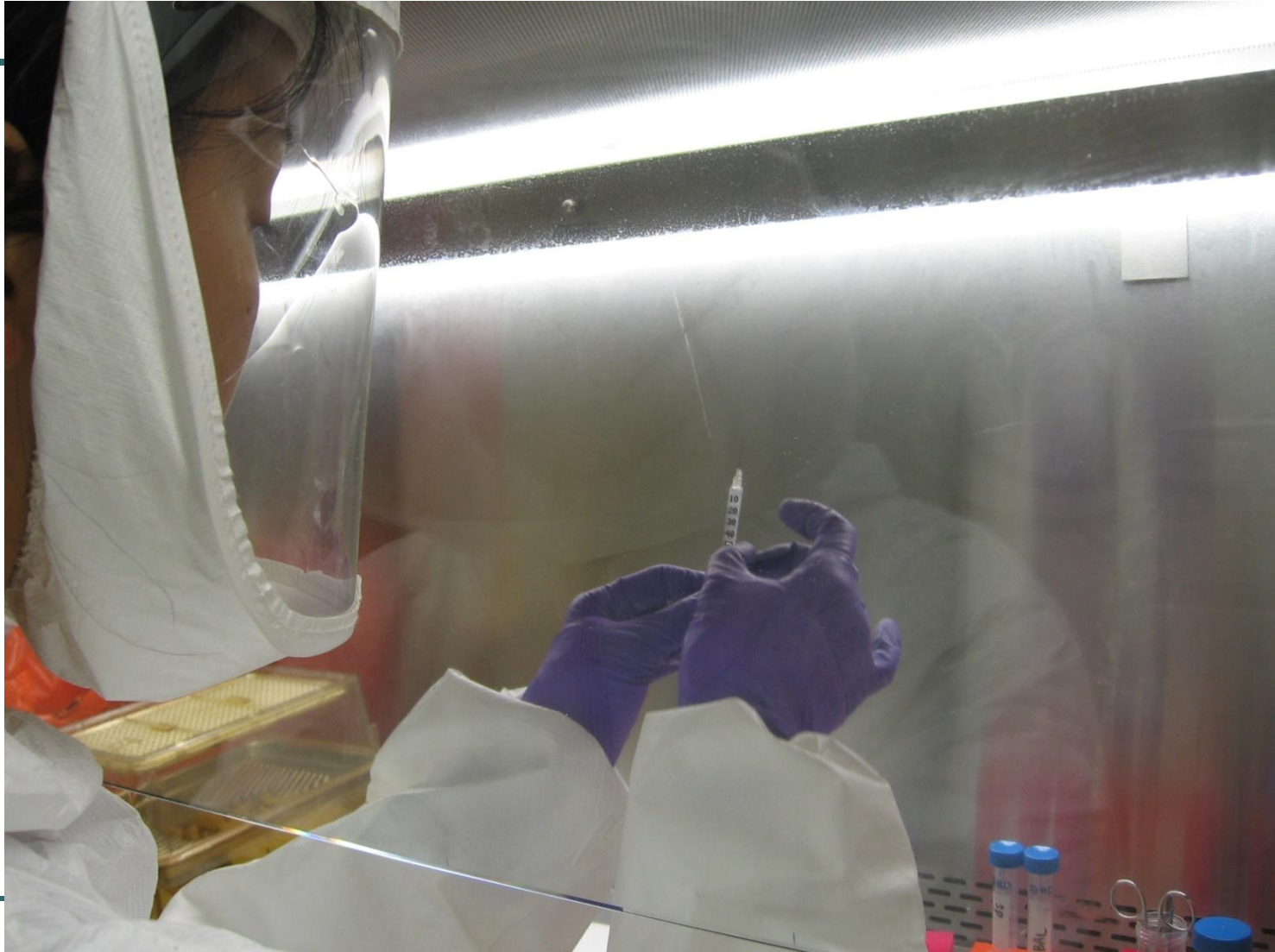
Use of video clips



Sharps Safety



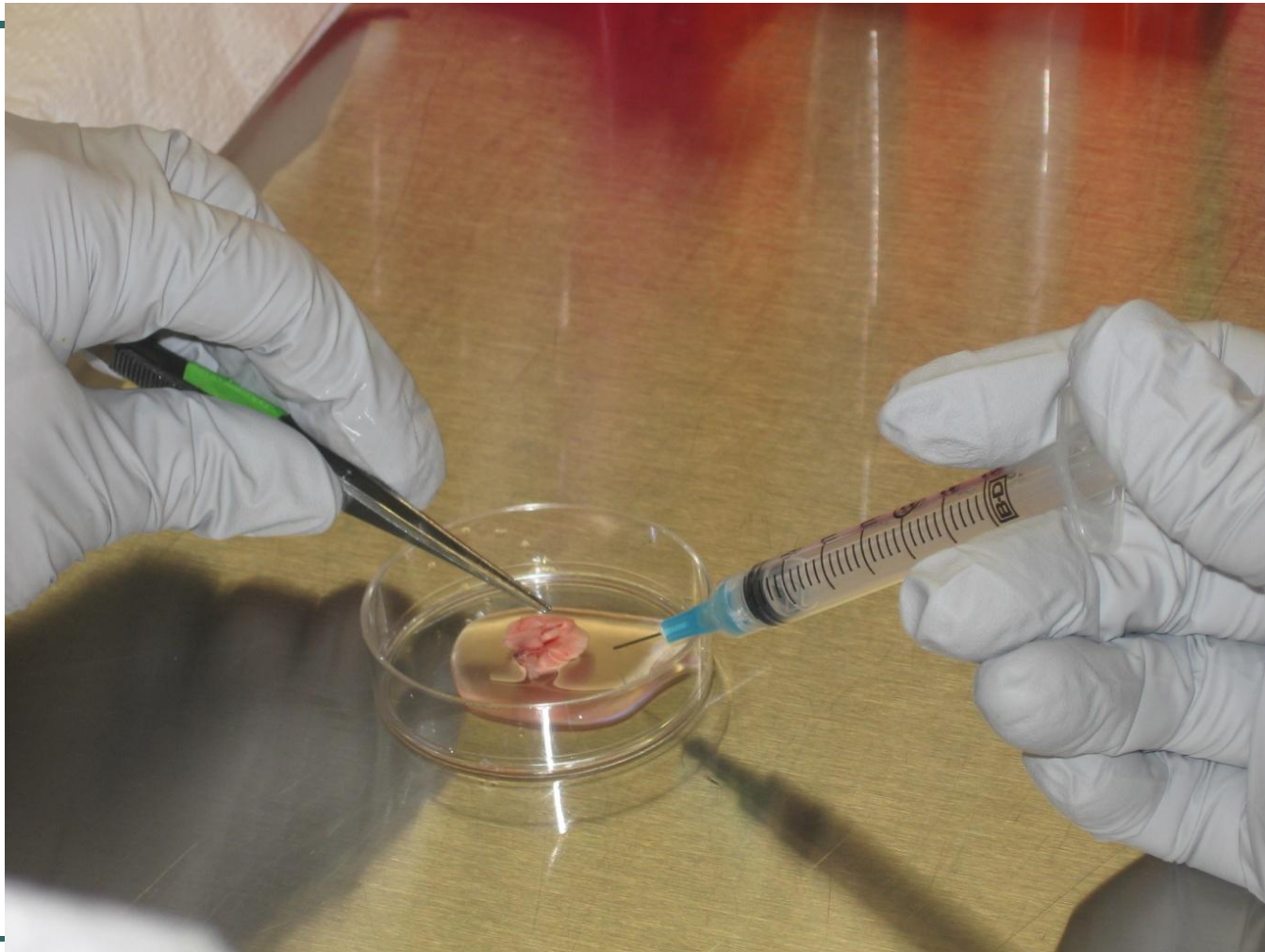
Recognition of aerosol generating procedures



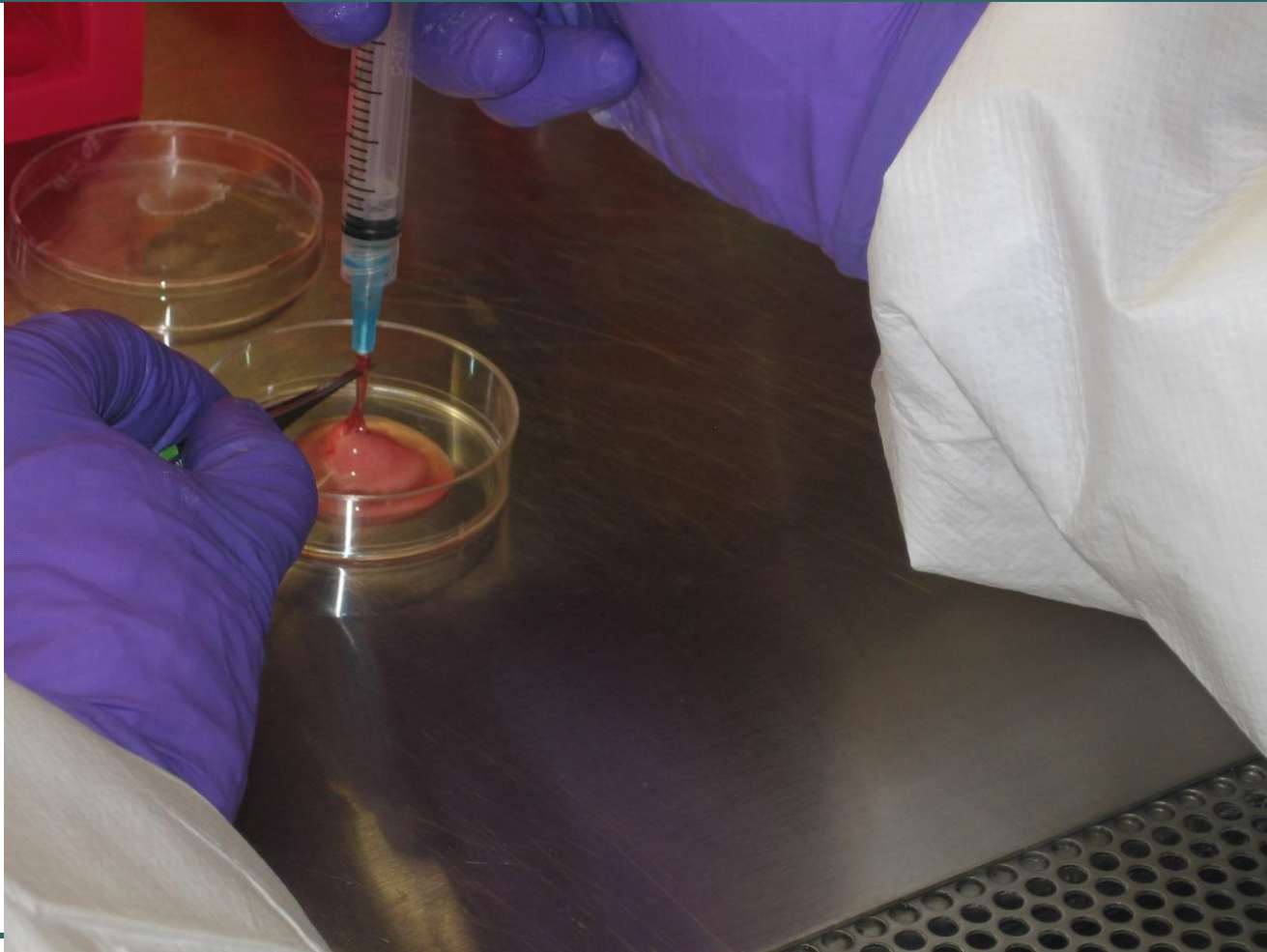
Use of video clips



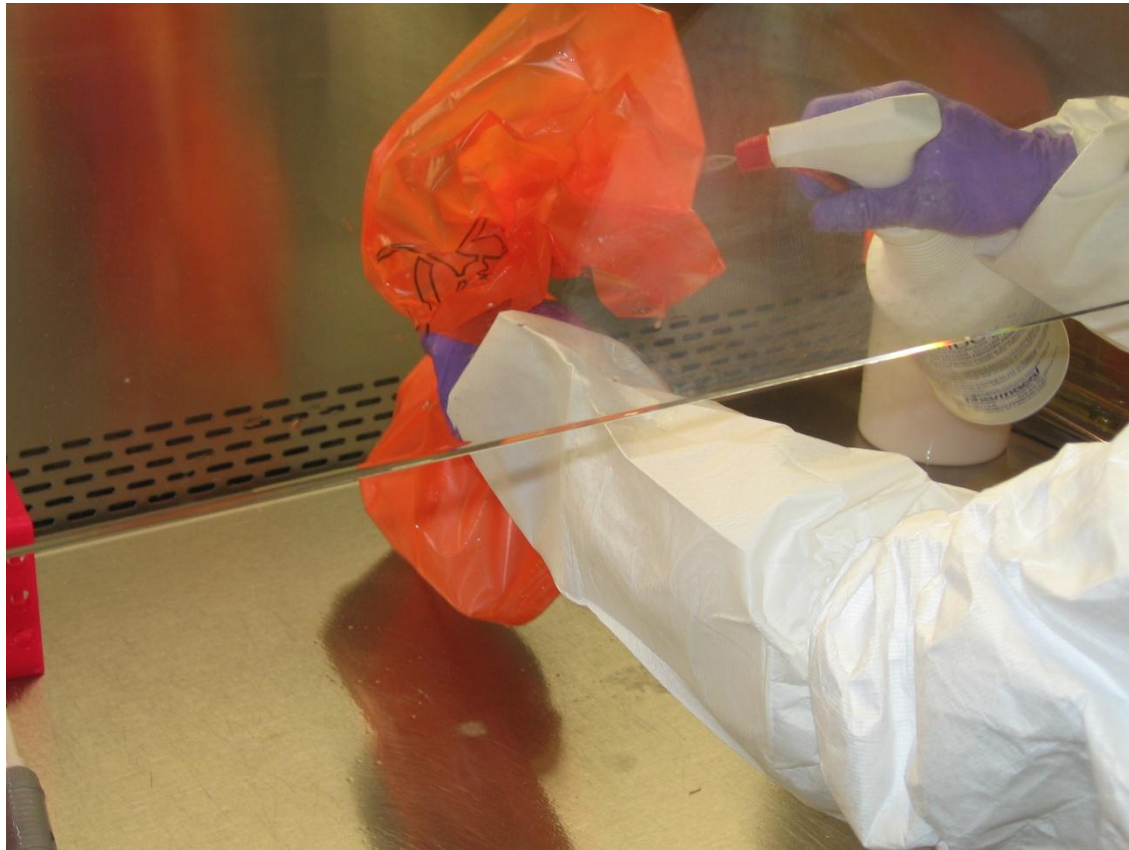
Observe All Procedures Post Necropsy



Keeping hands/fingers away from sharps



Termination of Work



Termination of Work



BSL-3 Work Practice Observation

- 2 Reviewers from BSL-3 Subcommittee
- Monitor:
 - Entry/exit
 - Compliance with PI's approved SOP
 - Aerosol containment
 - Safe handling of sharps (animal BSL-3)
 - Awareness of emergency procedures
 - Comfortable? Safe? Proficient?

Animal protocol SOP start-up meeting

- Researchers, animal technicians, veterinary care, biosafety, IACUC representative
 - **Non-EHS safety representatives on staff in animal care and IACUC**
- Begin with baseline template
- Discuss, review and develop final protocol

Researcher SOP Development

- Best written by those performing work
 - (Beati, Jacoby/Smith, Iwasaki)
- Clear/concise w/ sufficient detail to describe the process
- Reviewed, approved and updated
- Augment SOP's w/ posters (prompts, reminders)

Before Use

- Cabinet on?
- Drain valve closed?
- All waste containers and supplies loaded inside?
- Disinfectant prepared and in cabinet?

During Use

- Perform all work inside the cabinet
- Keep grilles clear
- Discard waste inside the cabinet
- Disinfect all items prior to removal
- Remove gloves or disinfect them before removal from the cabinet

Work Completion

- Seal all waste containers before removal from the cabinet
- Disinfect all items before removal
- Disinfect all work surfaces upon completion
- Back, sides, inside front view screen
- Rear grill, work surface, front grill

**Focus on
simple
essential
practices**

Donning PPE Poster Located in Entry Anteroom

Required PPE for Entering BSL3 Laboratory



BEFORE

exiting area
follow the
steps below
for proper
removal of
PPE



STEP #1: Remove outer gloves



STEP #2: Remove sleeves



STEP #3: Remove shoe covers



STEP: #4 Continue to Exit Anteroom

Instructions for PPE Removal Before Lab Exit to Rear Anteroom

Removal of Remaining PPE, Hand Washing and Lab Exit

PPE Removal Exiting BSL3 Laboratory Station #2



BEFORE
exiting area
follow the
steps below
for proper
removal of
PPE

STEP #1: Remove gown



STEP #2: Remove inner gloves



STEP #3: Remove faceshield

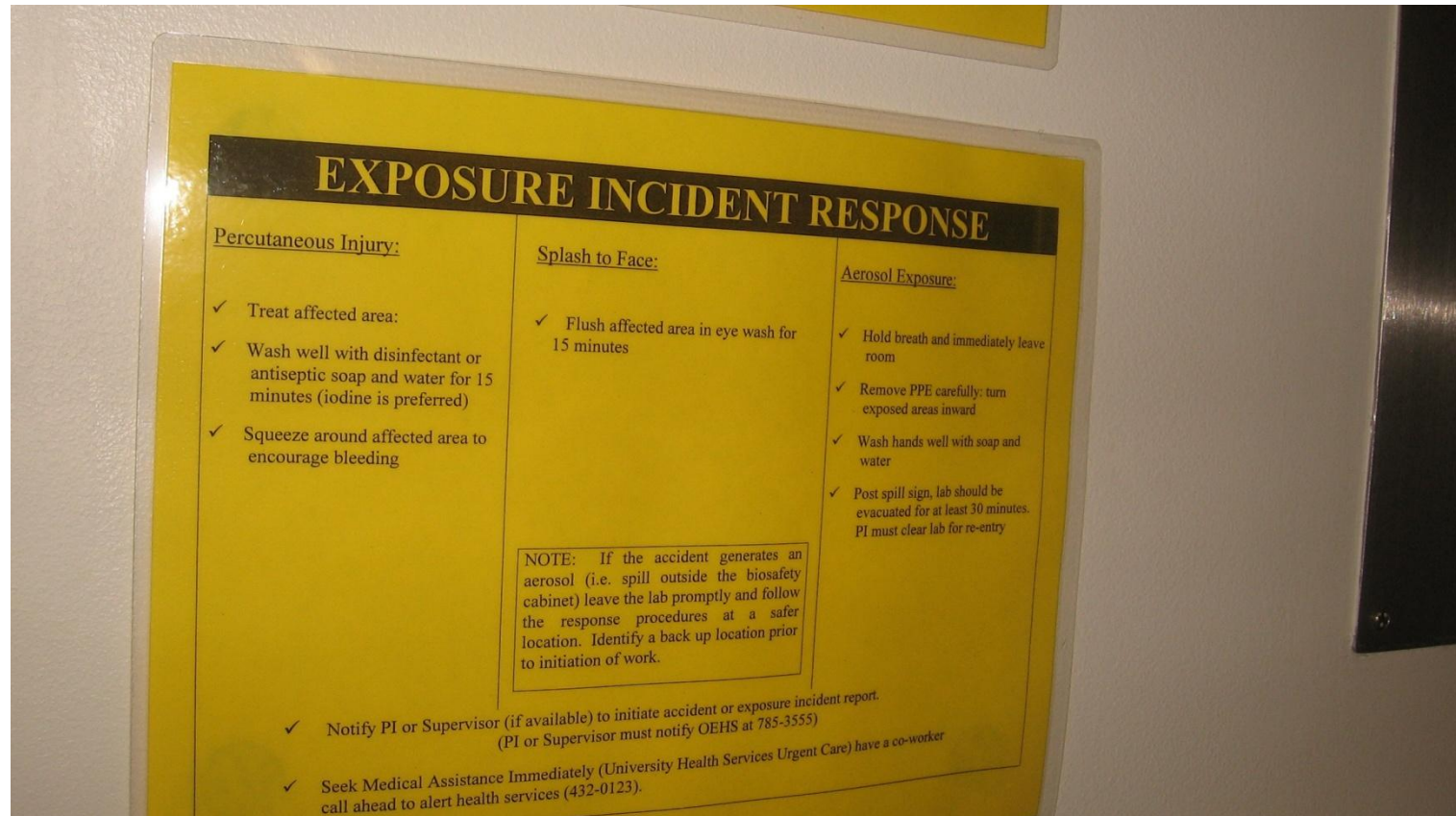


STEP #4: Remove respirator



Step #5: After washing your hands, you may leave the area

Exposure Response Poster



Centrifuge Spill Response SOP

CENTRIFUGE SAFETY

- ◆ Each operator must be trained on the proper operating procedures
- ◆ Keep a log book detailing operation records for centrifuges and rotors
- ◆ Do not exceed safe rotor speed
- ◆ Place a biohazard label on the centrifuge if used for infectious agents
- ◆ Always use sealed safety buckets or sealed rotors with O-rings
- ◆ Check tubes and bottles for cracks and deformities before each use
- ◆ Examine O-ring and replace if worn, cracking or missing
- ◆ Never overfill primary containers; do not exceed $\frac{3}{4}$ full
- ◆ Wipe exterior of tubes or bottles with disinfectant prior to loading into safety buckets or rotor
- ◆ Wipe the exterior of safety buckets or rotors with disinfectant before removing from biosafety cabinet
- ◆ Stop the centrifuge immediately if an unusual condition, such as noise or vibration, begins
- ◆ Wait five minutes after the run before opening the centrifuge to allow aerosols to settle in the event of a breakdown in containment
- ◆ Decontaminate safety buckets or rotors and centrifuge interior after each use
- ◆ Wash hands after removing gloves

CENTRIFUGE SPILL

If you notice that there has been a leak outside the safety bucket or rotor when opening centrifuge:

First:

- ◆ Hold Breath
- ◆ Close centrifuge lid
- ◆ Notify others to evacuate the lab

Then:

- ◆ Immediately leave the lab
- ◆ Post biohazard spill sign

Notify PI or Supervisor:

- ◆ DO NOT re-enter lab until PI and OEHS have given clearance (at least 30 minutes)
- ◆ Follow centrifuge spill instructions in the Biosafety Manual or Spill Response Guide

Decontaminate:

- ◆ Remove PPE turning exposed areas inward
- ◆ Place disposable PPE in biomedical waste (autoclave reusable PPE)
- ◆ Wash any exposed areas with antiseptic soap and water
- ◆ Wash hands thoroughly

For Centrifuge Explosion:
Evacuate room immediately; notify PI and OEHS

Spill Response Protocol

BL3 SPILL RESPONSE PROCEDURES

Use the following guidelines below for response to spills of BL3 material outside of the biosafety cabinet or any other incident that may have generated an aerosol in the containment laboratory, such as failure of physical containment devices during centrifugation.

IMMEDIATE ACTION:

- **Hold breath and leave room immediately; notify others in the room to evacuate immediately.**
- Remove personal protective equipment (PPE) in the airlock or access zone; turn potentially contaminated clothing inward; remove gloves last, and wash any exposed skin areas with antiseptic soap and warm water.
 - *In the event of an exposure incident:
 - needlesticks/puncture wounds: wash the affected area with antiseptic soap and warm water for 15 minutes. Squeeze around the area to encourage the flow of blood out of wound.
 - mucous membrane exposure: Use an eyewash for 15 minutes to flush the affected area.
- **Post a BIOHAZARD SPILL SIGN at entry to BL3 lab door.**
- Notify your Principal Investigator and EHS
- **Do not reenter laboratory until it has been cleared for reentry by the P.I. or EHS (785-3555).** In general a period of at least 30 minutes should be allowed before clean-up is attempted, but the time is contingent upon the supply and exhaust features of the lab.

BSL3 Work Practices Poster

BSL3 Laboratory Practices

1. Keep laboratory doors closed, and locked when unoccupied.
2. Post BSL3 biohazard sign on lab door, include agent, entry requirements, and an emergency contact number.
3. Verify proper air direction before each entry into BSL3.
4. Contact University Health Services for enrollment in the BSL3 medical surveillance program (2-0871).
5. Develop and maintain a lab BSL3 manual.
6. Do not permit visitors inside until lab is closed by P.I. and OEHHS (785-1199).
7. Wear solid front or wrap-around goggles, gloves that can fit over gown cuffs, and safety glasses inside but not outside the laboratory.
8. Perform all work with BSL3 agents inside a biological safety cabinet. Collect all BSL3 waste inside the biological safety cabinet.
9. Use physical containment devices, such as sealed rotors or safety buckets for centrifugation.
10. Load and unload sealed centrifuge rotors and safety buckets inside a biological safety cabinet.
11. Use a biological safety cabinet to contain aerosol-producing equipment.
12. Wear respiratory protection devices when aerosols cannot be contained.
13. Protect the vacuum system from contamination by a hepa or hepa-like filter in between the vacuum system and the collection flask.
14. Avoid using hypodermic needles and other sharps.
15. Wash hands after completing experimental procedures and before leaving the laboratory.
16. Use leakproof, labeled, unbreakable containers for the transport of BSL3 materials between laboratories. Consult OEHHS for interstate shipment requirements.
17. Verify autoclave procedures periodically.
18. Report spills and potential exposures to P.I. and OEHHS. An eyewash should be readily available.
19. Maintain a biological spill kit in a location outside the laboratory to facilitate spill response and decontamination.
20. Report any facility or equipment problems to OEHHS and Physical Plant.

IN CASE OF EMERGENCY

**In the event of fire,
alarm or emergency**

**C... Contain All
Materials**

A... Abandon Work

**L... Lock Select
Agent Away**

**L... Leave the
Laboratory**

Effluent Decontamination SOP



Effluent Decontamination Sink SOP

Step 1: Close drain valve below sink

Step 2: Pour ½ Gallon of Household Bleach into sink drain

Step 3: Start using sink (Green light will come on in first few uses)

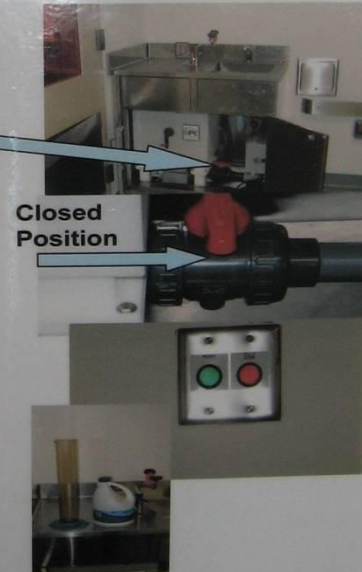
Step 4: Stop using sink when Red Light Comes on or "fill-line" reached

Step 5: Add ½ Gallon of Household Bleach into sink drain

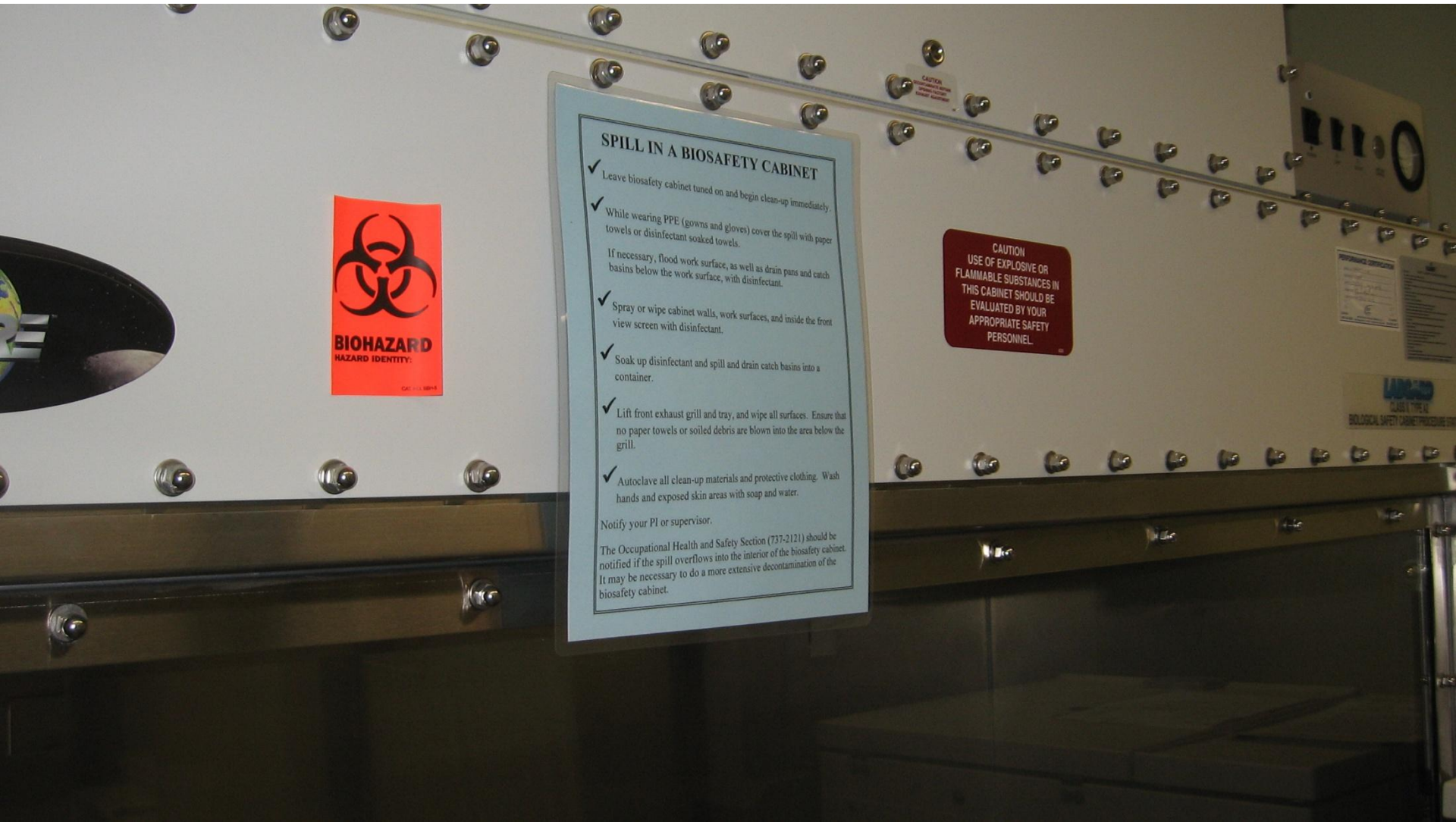
Step 6: Wait 15 – 30 minutes

Step 7: After 30 minutes, open valve and allow liquid in tank to drain

Step 8: Close valve and add ½ Gallon of Household bleach to sink drain to reset the tank



Spill Inside a Biosafety Cabinet



SPILL IN A BIOSAFETY CABINET

- ✓ Leave biosafety cabinet tuned on and begin clean-up immediately.
- ✓ While wearing PPE (gowns and gloves) cover the spill with paper towels or disinfectant soaked towels.
If necessary, flood work surface, as well as drain pans and catch basins below the work surface, with disinfectant.
- ✓ Spray or wipe cabinet walls, work surfaces, and inside the front view screen with disinfectant.
- ✓ Soak up disinfectant and spill and drain catch basins into a container.
- ✓ Lift front exhaust grill and tray, and wipe all surfaces. Ensure that no paper towels or soiled debris are blown into the area below the grill.
- ✓ Autoclave all clean-up materials and protective clothing. Wash hands and exposed skin areas with soap and water.

Notify your PI or supervisor.
The Occupational Health and Safety Section (737-2121) should be notified if the spill overflows into the interior of the biosafety cabinet. It may be necessary to do a more extensive decontamination of the biosafety cabinet.

CAUTION
USE OF EXPLOSIVE OR
FLAMMABLE SUBSTANCES IN
THIS CABINET SHOULD BE
EVALUATED BY YOUR
APPROPRIATE SAFETY
PERSONNEL


BIOHAZARD
HAZARD IDENTITY

RHO

PERFORMANCE CERTIFICATE

LABOR
CLASS 1 TYPE A
BIOLOGICAL SAFETY CABINET PROCEEDURE FOR

Conclusion

- It takes a village
- Best practices are developed together
 - “None of us is as smart as all of us.” Satchel Paige
- Share your knowledge, but listen and learn
- Evolve with your programs