



Biosafety Laboratory Level



(BSL1-4)

Assoc. Prof. Chonlaphat Sukasem, B.Pharm., Ph..D.

Secretariat, Biosafety Association of Thailand, BAT

Member, IBC for Mahidol University

Member, IBC for Ramathibodi Hospital

Chair, IBC for Department of Pathology

chonlaphat_suk@hotmail.com

chonlaphat.suk@mahidol.ac.th

081 90 23457

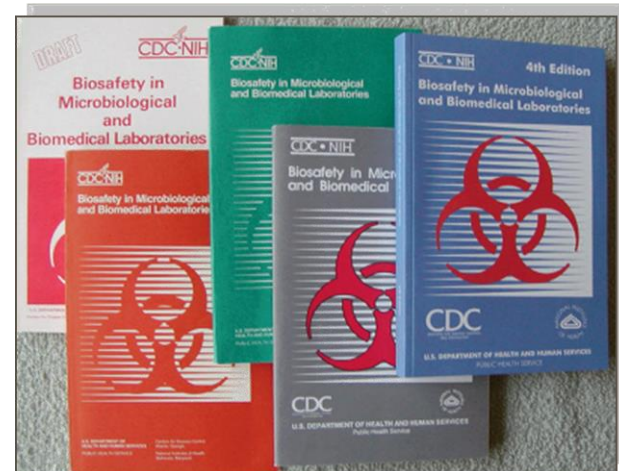


Goals of today's talking

BIOSAFETY :

- knows the BSL 1-4
- Choose the right one

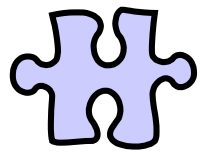
<http://www.cdc.gov/od/ohs/biosfty/bmbl5/bmbl5toc.htm>



4 Primary controls

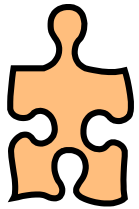


Biosafety: prevent Lab workers, experimental contamination and the environment



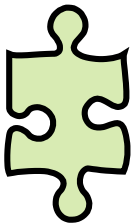
I. Engineering Controls

ข้อกำหนดทางวิศวกรรม



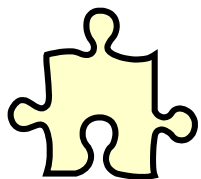
II. Administrative Controls

การบริหารจัดการที่ดี



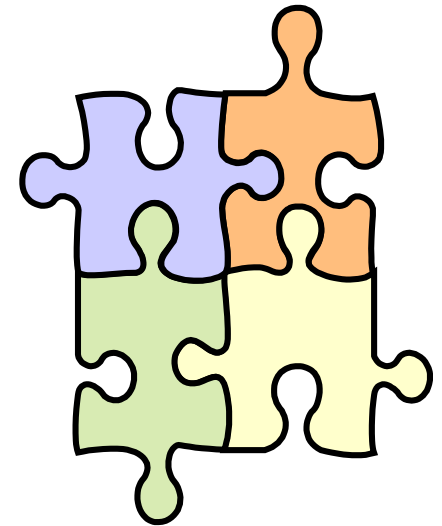
III. Practices and Procedures

วิธีปฏิบัติที่เป็นมาตรฐาน



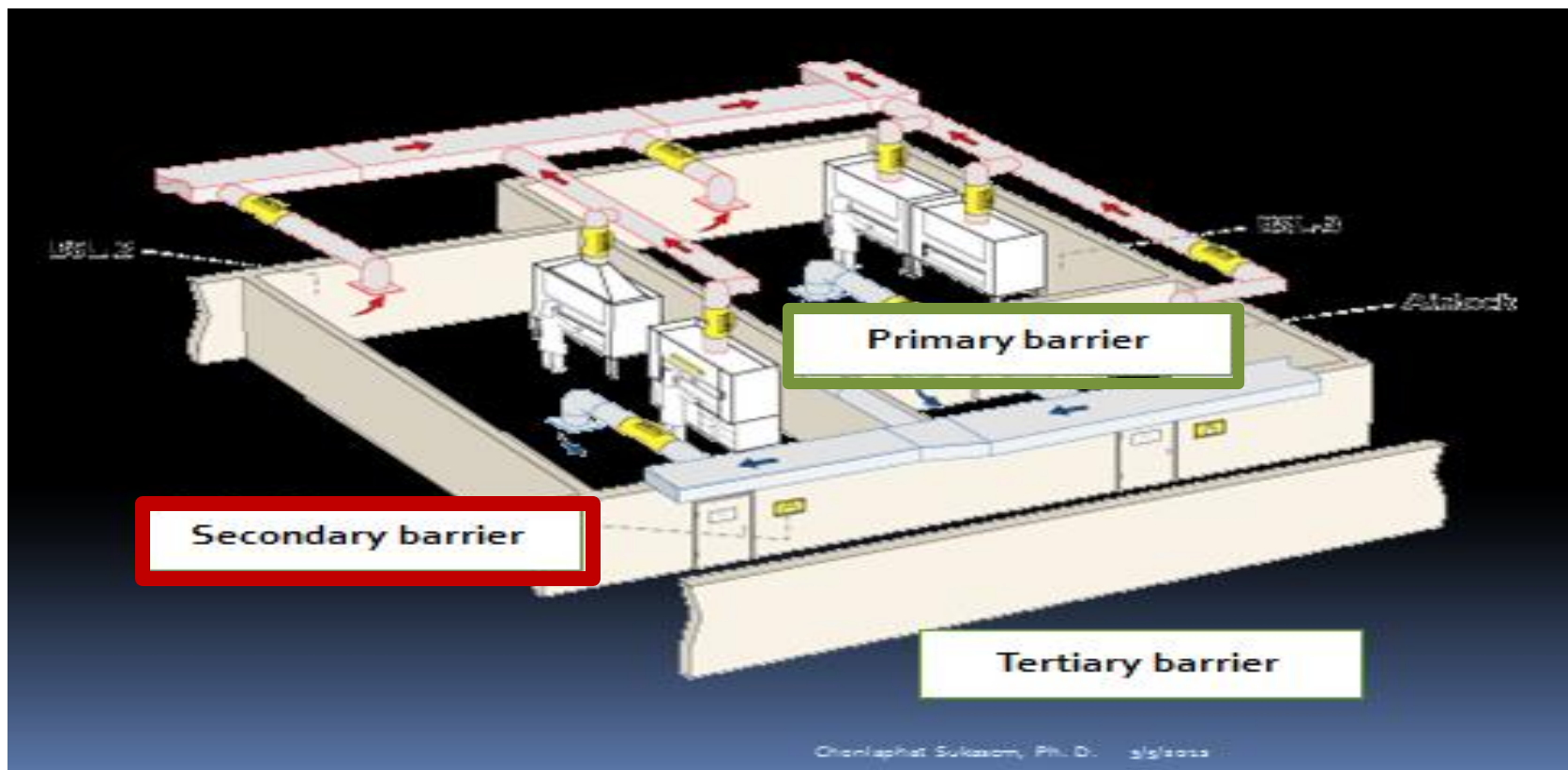
IV. Personal Protective Equipment

อุปกรณ์ป้องกันอันตรายเฉพาะบุคคล



Biosafety Lab

- Technology based, reduce or eliminate exposure to hazards by changes at the source of the hazard.
- Containment: **Primary vs Secondary**, Containment levels





Biosafety Lab Levels

- **BSL-1** - basic lab
- **BSL2** - basic lab + aerosol confinement
 - biosafety cabinet
- **BSL3** - containment laboratory
 - 2 door separation from building occupants
- **BSL4** - maximum containment laboratory
 - separate building from general research population

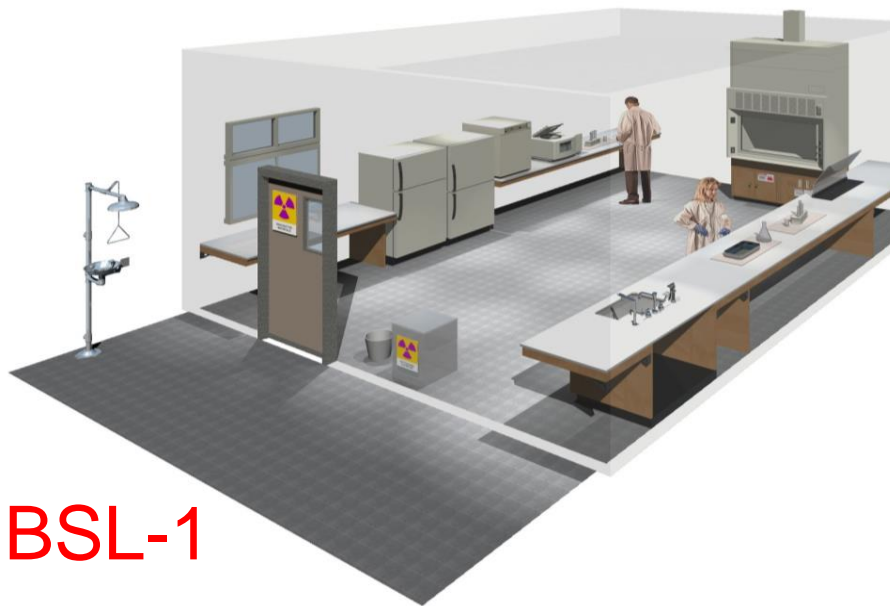
Secondary Barriers

Biosafety containment levels

- Each level associated with appropriate
 - Equipment (engineering control)
 - Practices (SOP)
 - work procedures (SOP)
- Diagnostic and health-care laboratories (public health, clinical or hospital-based) must be **biosafety level 2** or above

World Health Organization. Laboratory biosafety manual. – 3rd ed.

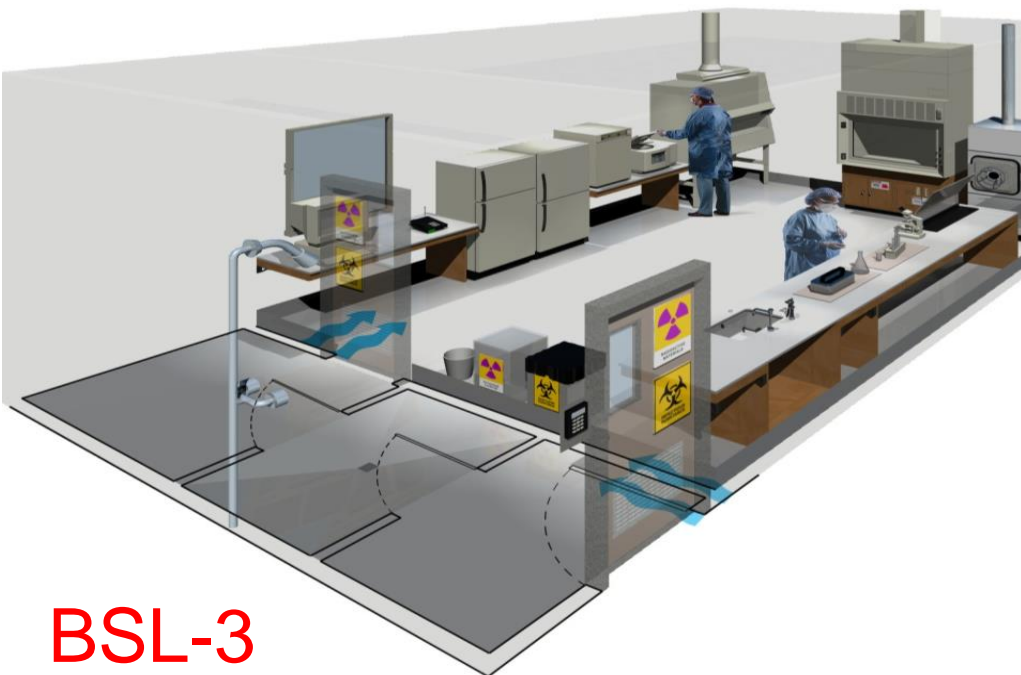
<http://www.who.int/csr/resources/publications/biosafety/Biosafety7.pdf>



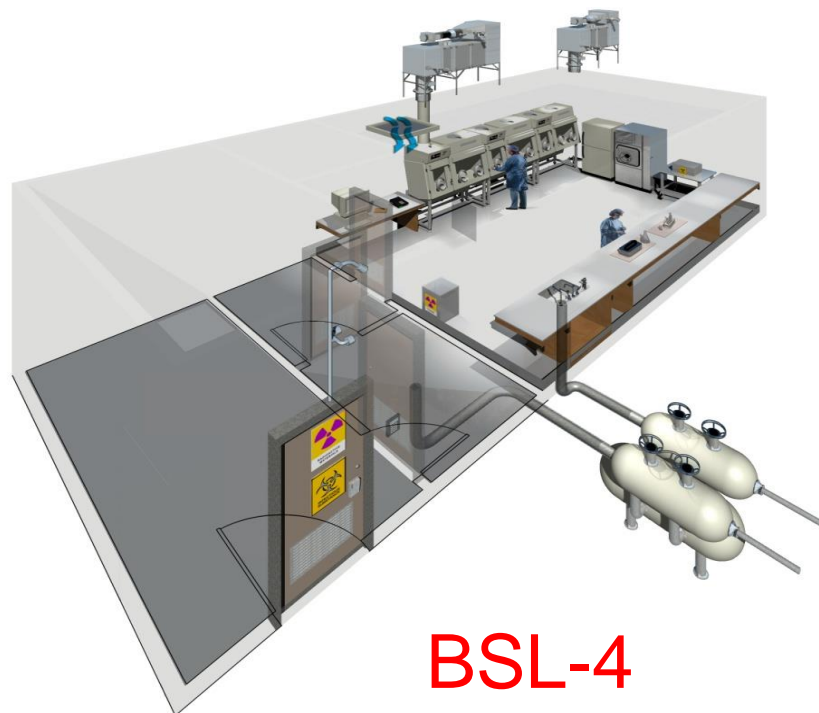
BSL-1



BSL-2



BSL-3

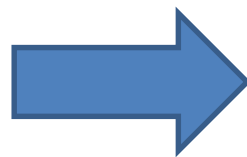


BSL-4

Laboratory biosafety level (BSL)

รศ. ดร.ชลภัทร์ สุขเกษม

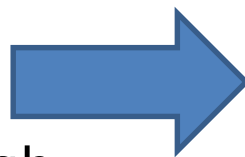
BSL-1: low risk



**Basic BSL-
Laboratory**

BSL-2: moderate

BSL-3: high risk



**Containment BSL-
Laboratory**

BSL-4: extreme high

***Public space and area: Access control and safety

หลีกเลี่ยงการจัดตั้งบริเวณ main corridor



BSL-1 laboratories are not necessarily separated from the general traffic patterns in the building.

Engineering controls

Fire precaution

Construction: primary and secondary exit routes

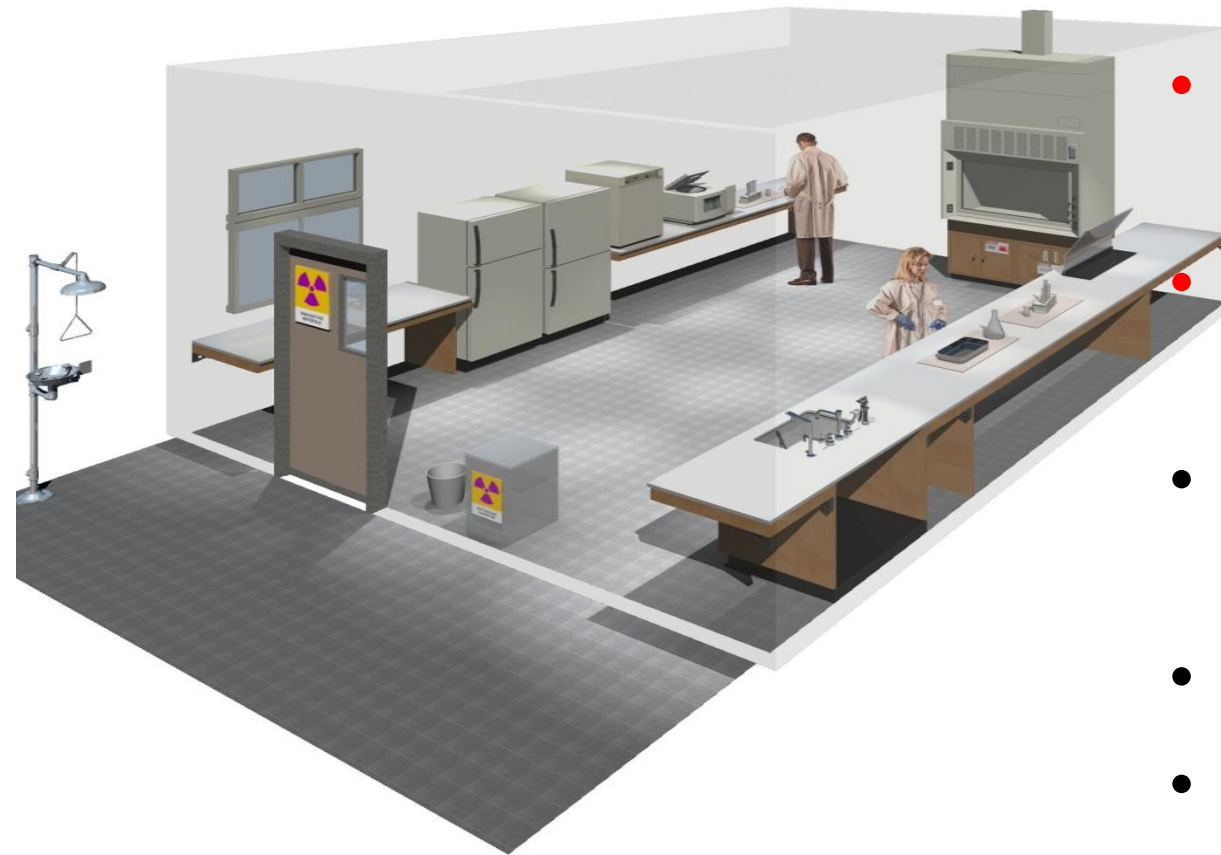
- 3^{ชั้น} ต้องมีบันไดหนีไฟ
- ทางหนีไฟ > 2 ม.
- ทางออก > 2 ทาง
- ประตู > 110 ซม.



BSL-1 laboratory

- Door
- **a sink for hand washing**
- **Easily cleaned work surfaces**
- Impervious bench tops
- Sturdy furniture
- Windows fitted with fly screens

***Emergency shower and eyewash available



BSL-1 Practices

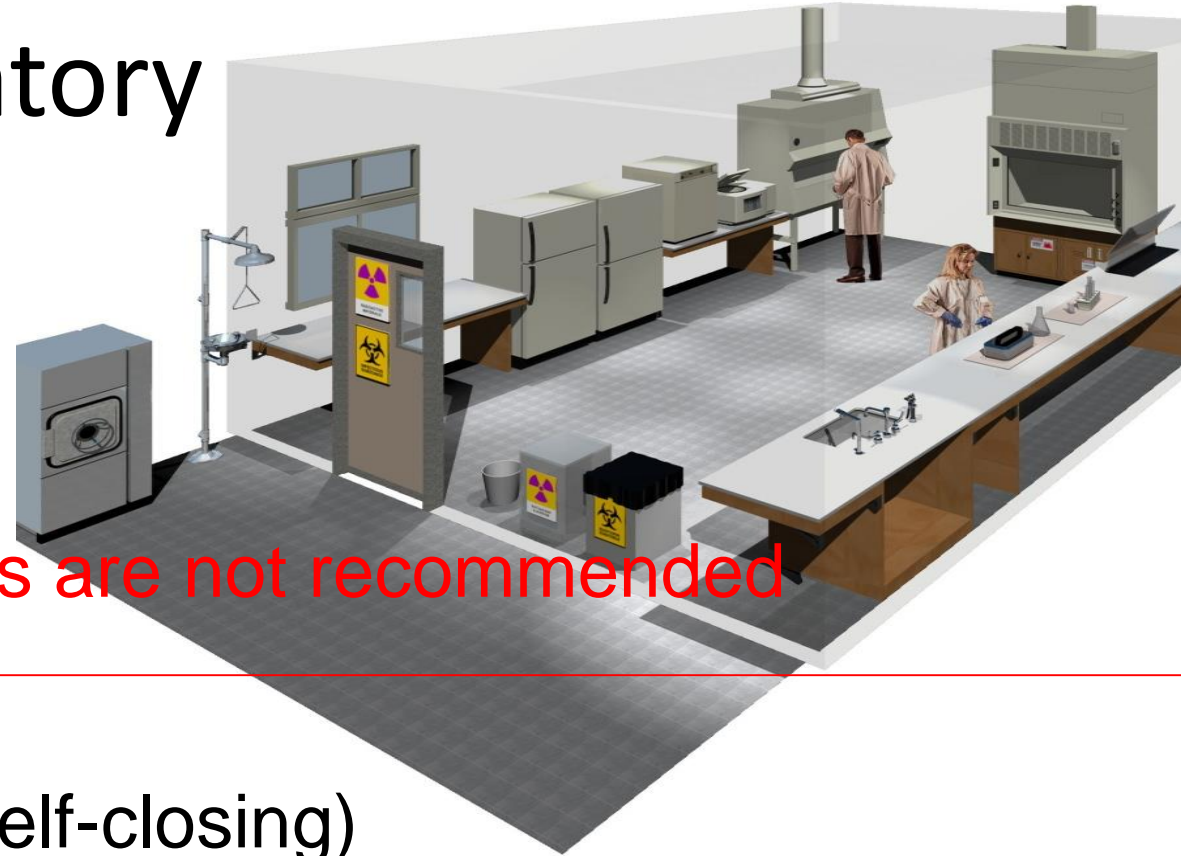
They follow **basic safety procedures** and require **no special equipment or design features**.

- **Bench-top work allowed**
- Standard Microbiological Practices
- **Required Hand washing**
- Red bag waste
- 2° containment (autoclave)

- E.coli K-12
- Transgenic Plants
- Plasmids
- Fungi
- Mold
- Yeast

BSL-2 laboratory

BSL-1++



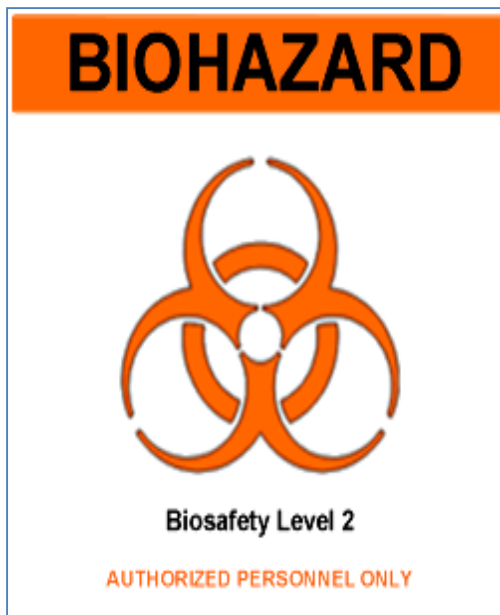
windows are not recommended

BSL1 Lab plus:

- lockable doors (self-closing)
- chairs covered with non-fabric material
- **Biosafety cabinets should be installed**
- airflow, non-recirculation recommended
- door closed when work is in progress
- autoclave (accessible)

BSL-1>>>BSL-2

- 1) Specific training (handling pathogenic agents)
- 2) access to the laboratory is restricted
- 3) all procedures in which infectious aerosols or splashes may be created are conducted in BSCs.



Biohaz. Sign posted (entrance)
Label all equipment

BSL-2 Laboratory



BIOHAZARD

All Personal Protective Equipment shall be removed prior to leaving this work area.

Eating, drinking, smoking, applying cosmetics or lip balm, and handling contact lenses are prohibited in this work area.

Name of infectious agent(s):

Transmission route(s):

Special requirements for entering this area:

Emergency contacts

Name: _____ Title: _____ Phone: _____

EH&S office

(M-F 8-5) 646-3327 (after hours) 911

*Biosafety Level 2 is similar to BSL-1 and is suitable for work involving agents of moderate potential hazard to personnel and the environment. It differs from BSL-1 in that:

1. Lab personnel have specific training in handling pathogenic agents and directed to competent scientist;
2. Access to the lab is limited when work is being conducted;
3. Extreme precautions are taken with contaminated sharp items and
4. Certain procedures in which infectious aerosols or splashes may be created are conducted in biological safety cabinets or other physical containment equipment.

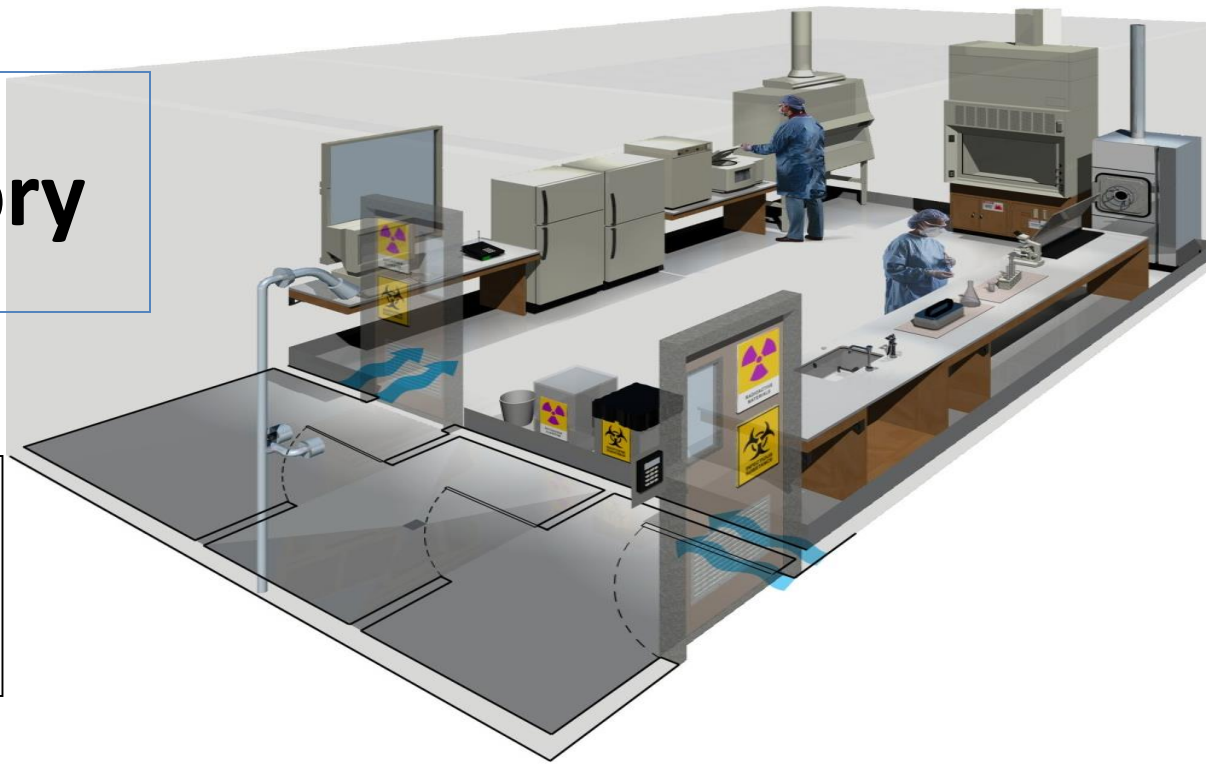
BSL-2 Practices

- **Limited access to lab** when work in progress
- Waste decontamination facilities such as an autoclave.
- Lab coat, safety glasses and gloves required
- Red bag & sharps containers required

- Human or Primate Cells
- HSV
- HBV
- Patient specimens

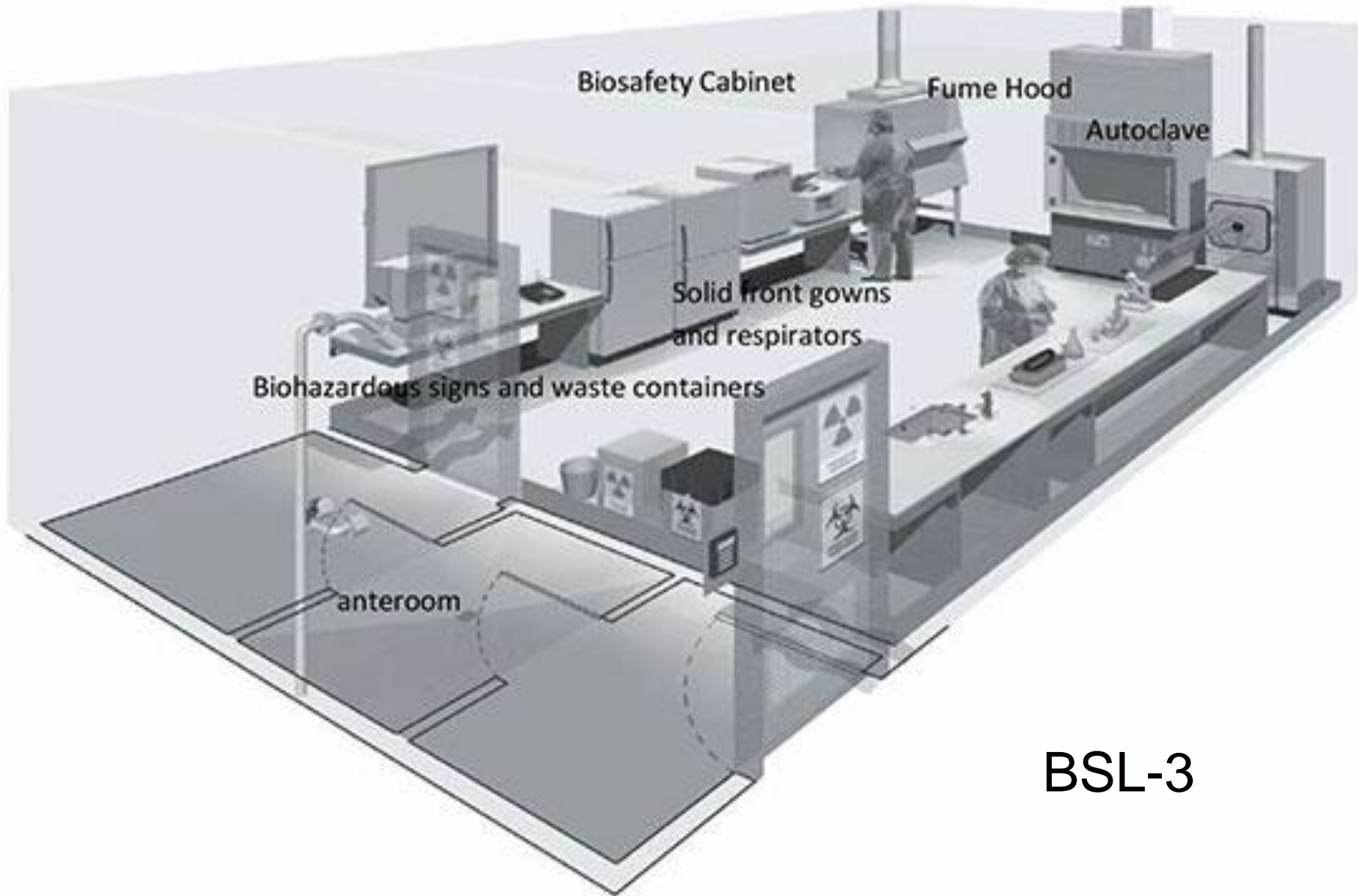
BSL-3 laboratory

BSL-1+2++



Lab Design & facilities

- 2 doors in a series to access lab and anteroom
- Special exhaust ventilation
(Not re-circulated, airflow (HVAC))
- negative pressure
- sinks, doors, floor, airlock...
- Able to wash entire lab



Biosafety Cabinet

Fume Hood

Autoclave

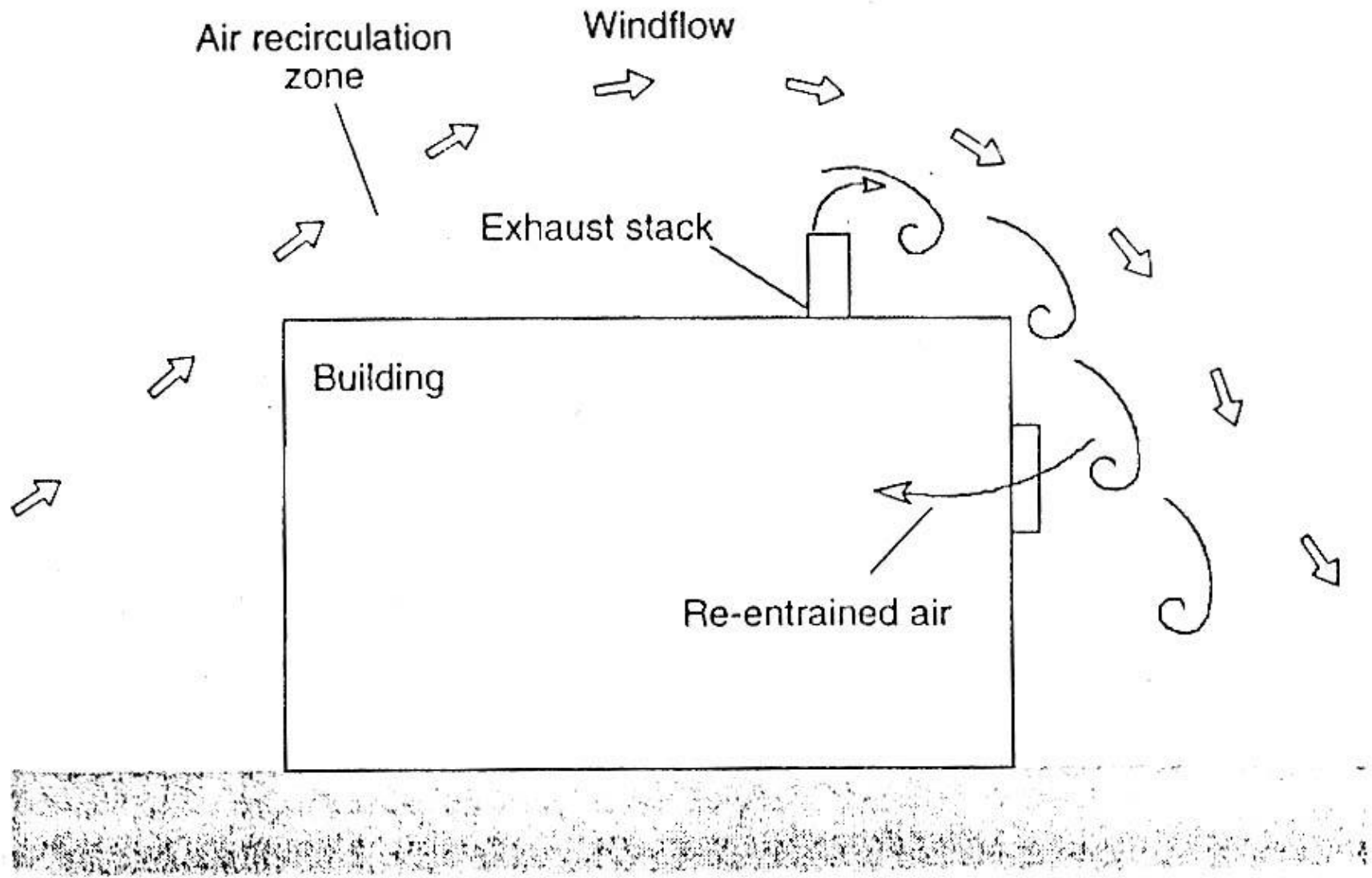
Solid front gowns
and respirators

Biohazardous signs and waste containers

anteroom

BSL-3

FIGURE S3-7. Air recirculation zone* created by wind blowing over a building



***Height of air recirculation zone may be variable. Air should be exhausted above this zone to prevent re-entrainment.**

BSL-3 Practices

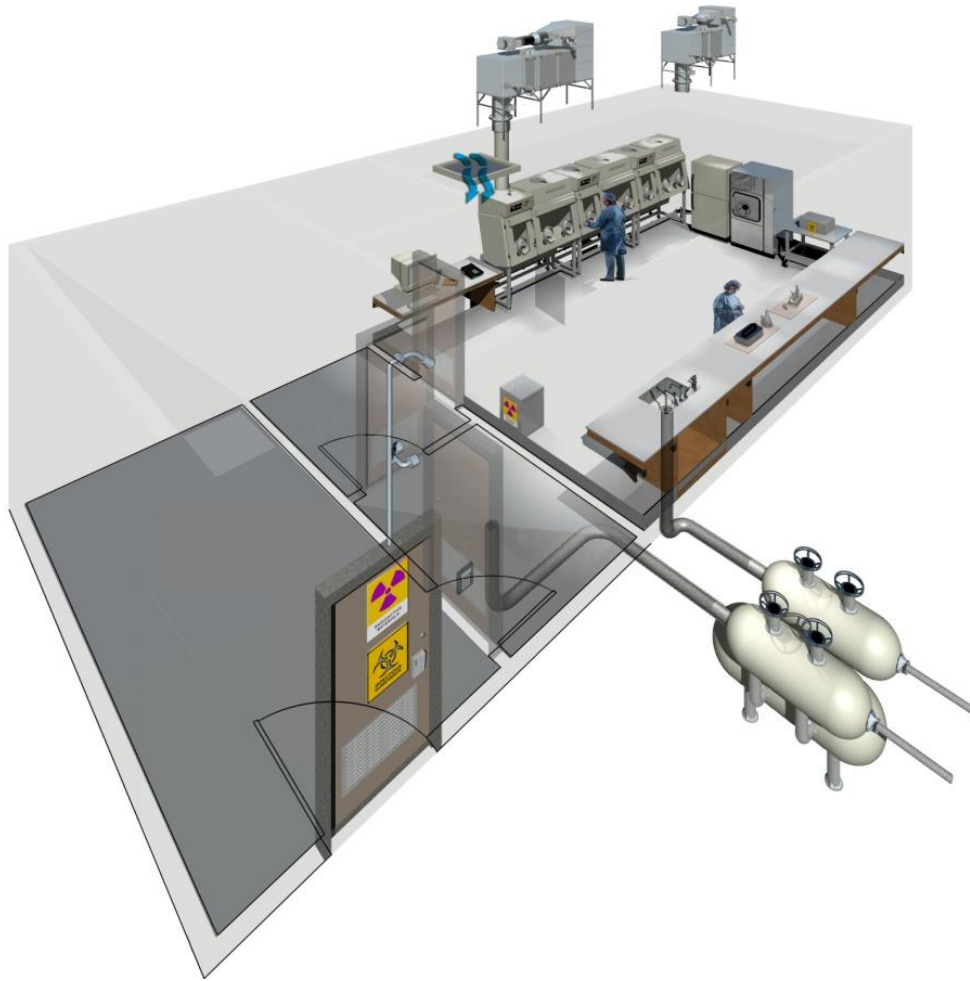
- **Public access NOT permitted**
- Autoclave required and waste is disposed at the end of day
- Required foot activated hand washing sink and controls
- **No sharps unless absolutely necessary**

*****Biosecurity: Storage of serum samples from at-risk personnel.**

BSL-3 Practices

- **Bench top work not permitted**
- **All work in biosafety cabinet**
- Documented training and personnel competency certification (for BSL-3 procedures)
- **Baseline serology and medical surveillance**
- **Offered appropriate immunizations**
- Spills – report immediately and treat accordingly

BSL-4 laboratory

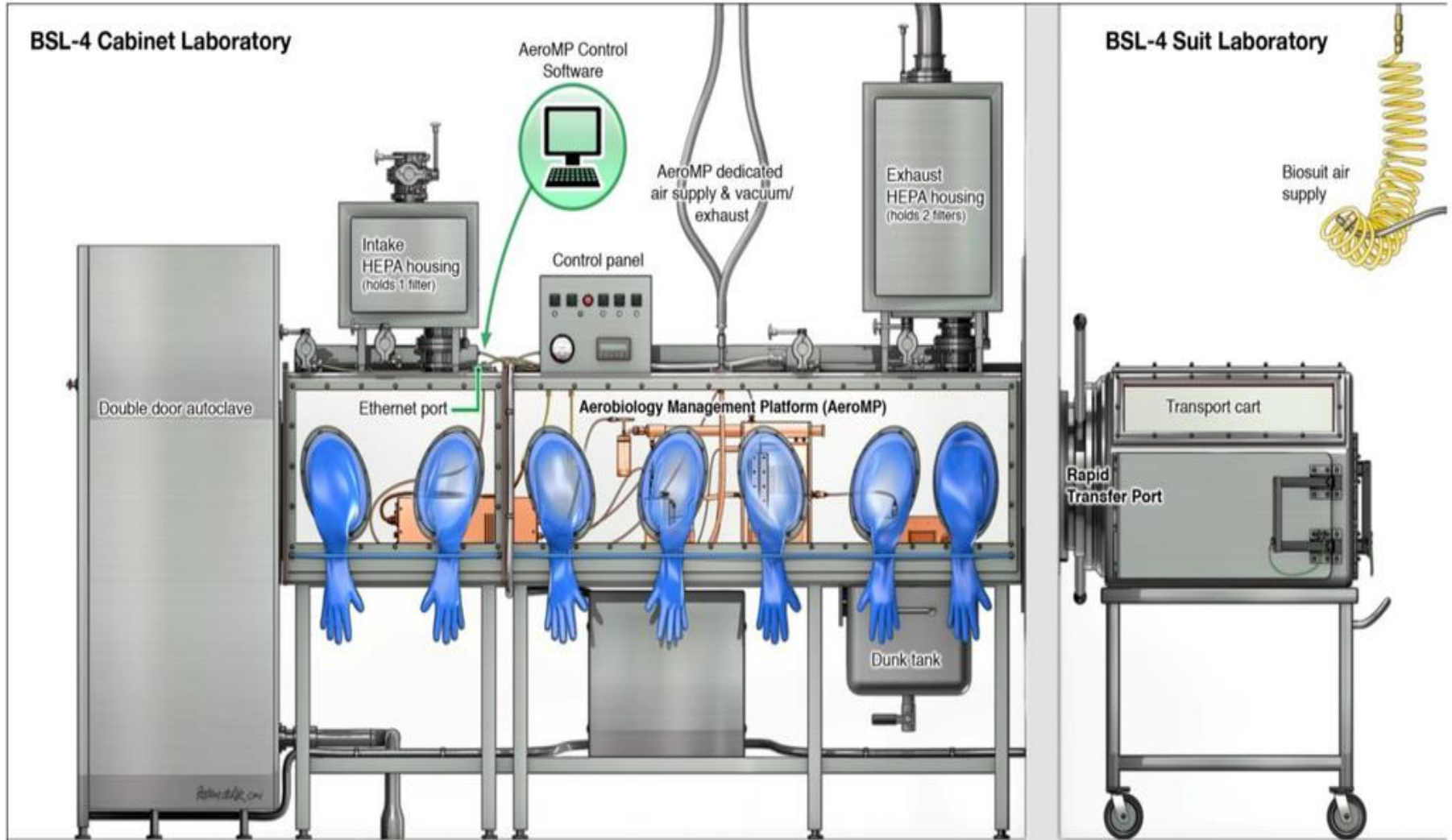


BSL-1+2+3++

- Supply air: HEPA
- Exhaust air: 2 HEPA
- A double-door, pass through autoclave

- Lassa Fever Virus
- Ebola Hemorrhagic Fever Virus
- Marburg Virus

BSL-4: Cabinet and Suit Laboratory





A positive pressure supplied air protective suit.

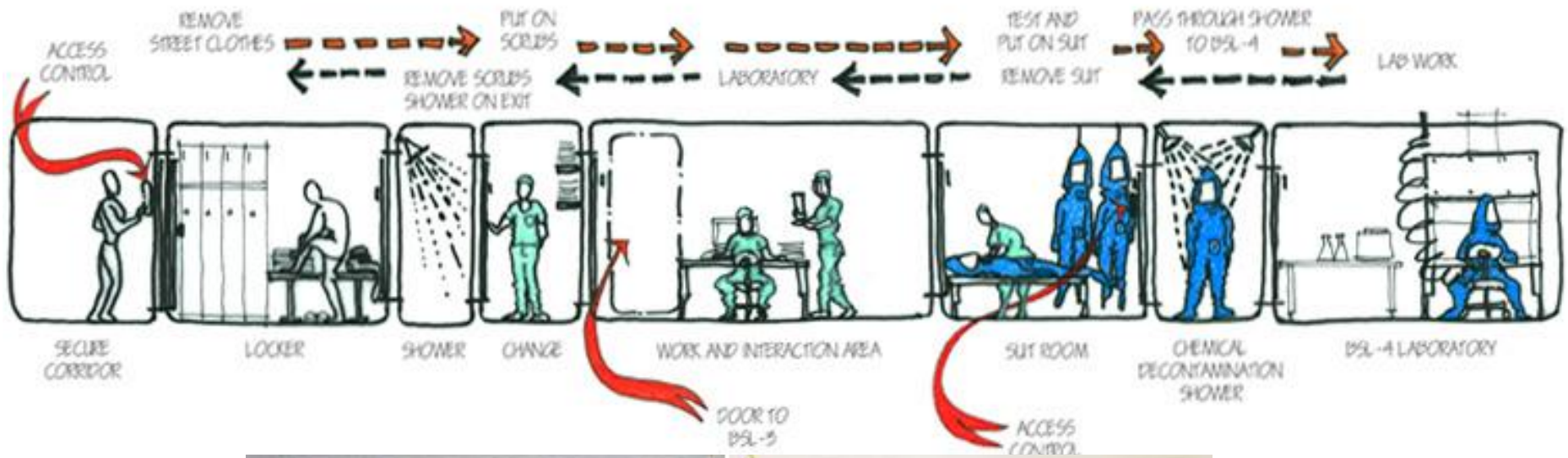


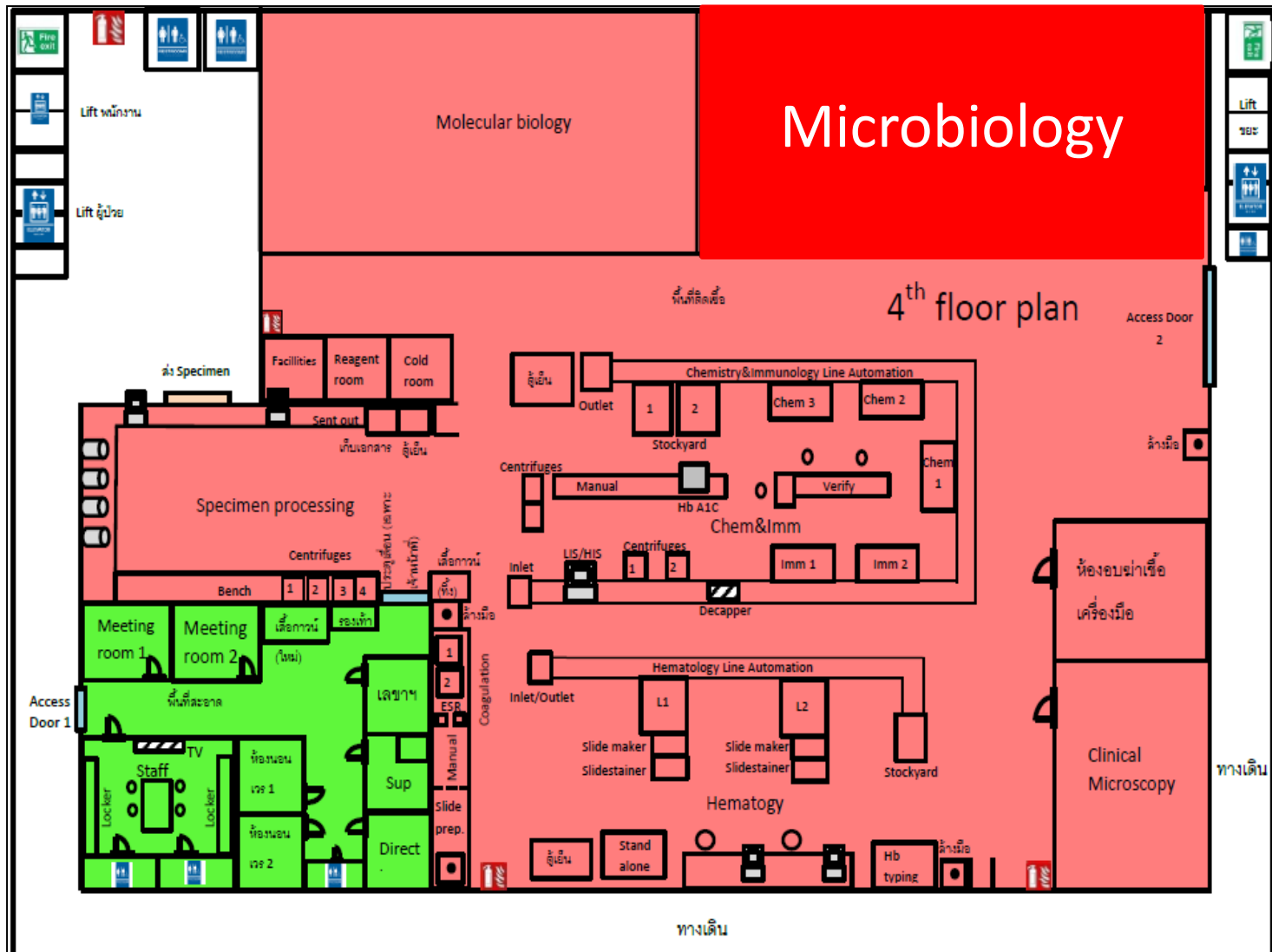
Table 5. Relation of risk groups to biosafety levels, practices and equipment

Risk Group	Biosafety Level	Laboratory type	Laboratory practices	Safety equipment
1	Basic - Biosafety Level 1	Basic teaching, research	GMT	None; open bench work
2	Containment - Biosafety Level 2	Primary health services; diagnostic, research	GMT plus protective clothing, biohazard sign	Open bench plus BSC for potential aerosols
3	High Containment - Biosafety Level 3	Special diagnostic, research	As Level 2 plus special clothing, controlled access, directional air flow	BSC and/or other primary devices for all activities
4	Maximum containment - Biosafety Level 4	Dangerous pathogen units	As Level 3 plus airlock entry, shower exit, special waste disposal	Class III BSC, or positive pressure suits in conjunction with Class II BSCs, double ended autoclave (through the wall), filtered air

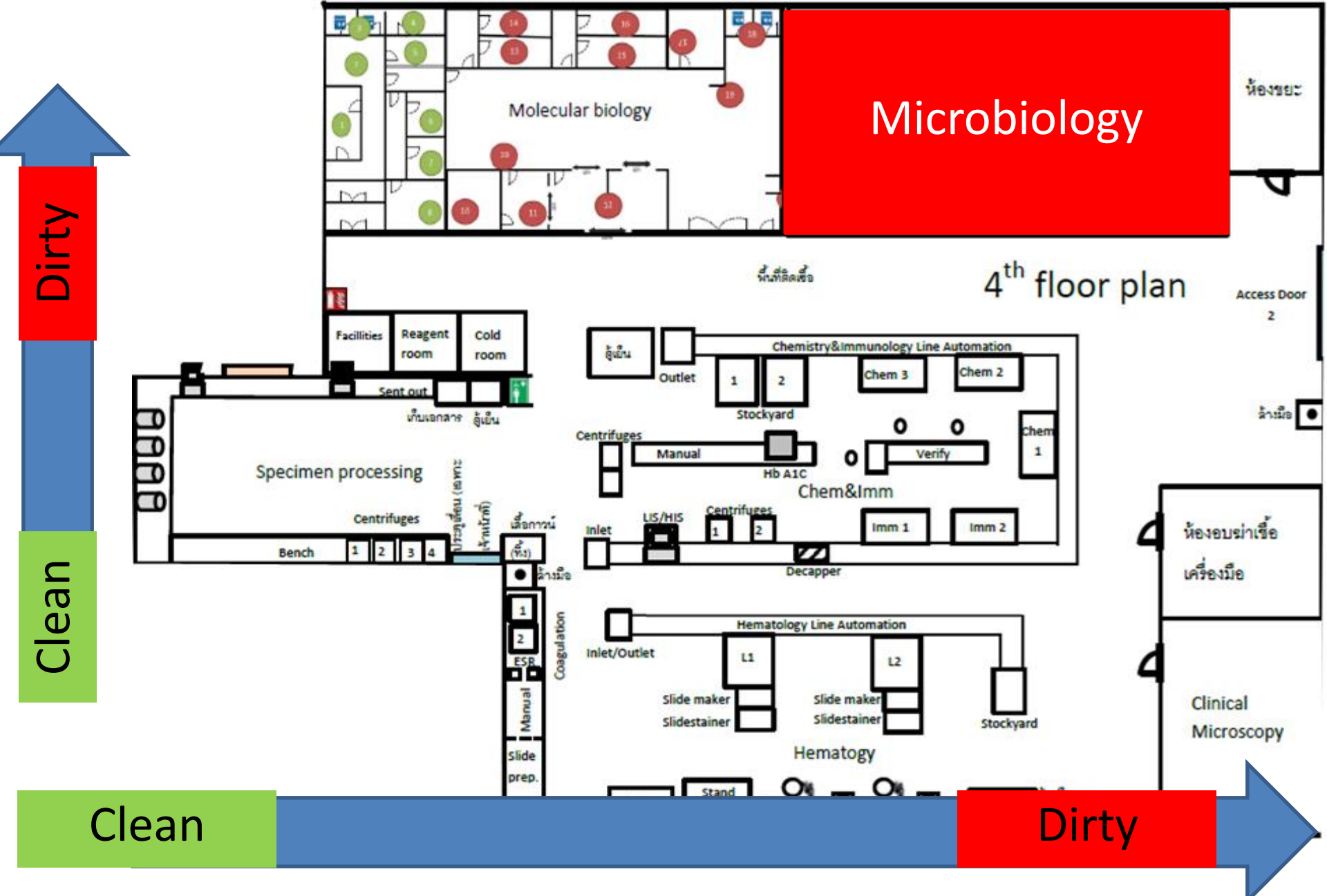
GMT: Good Microbiological Techniques (Chapter 5)

BSC: Biological Safety Cabinet

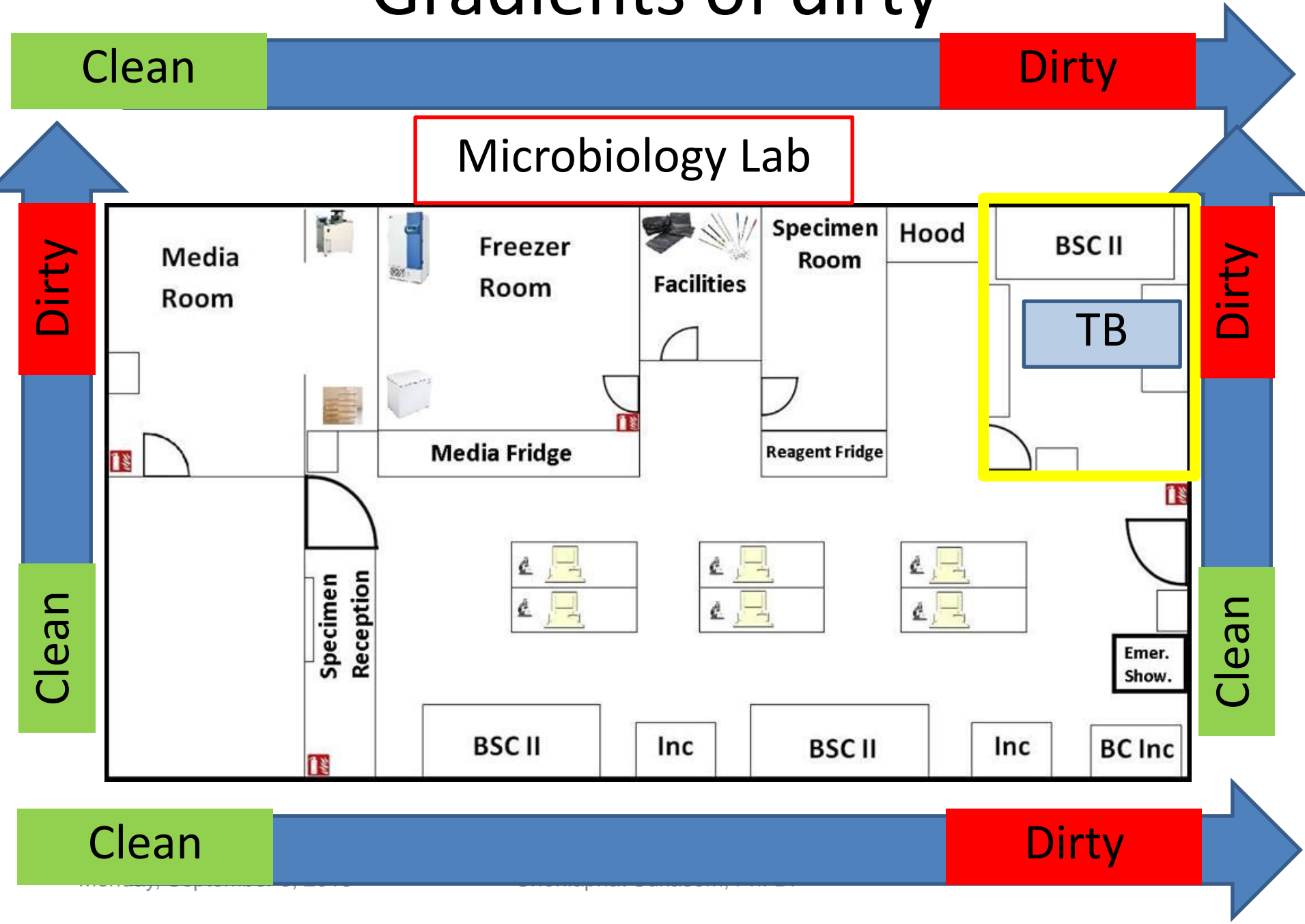
Divide into Zone

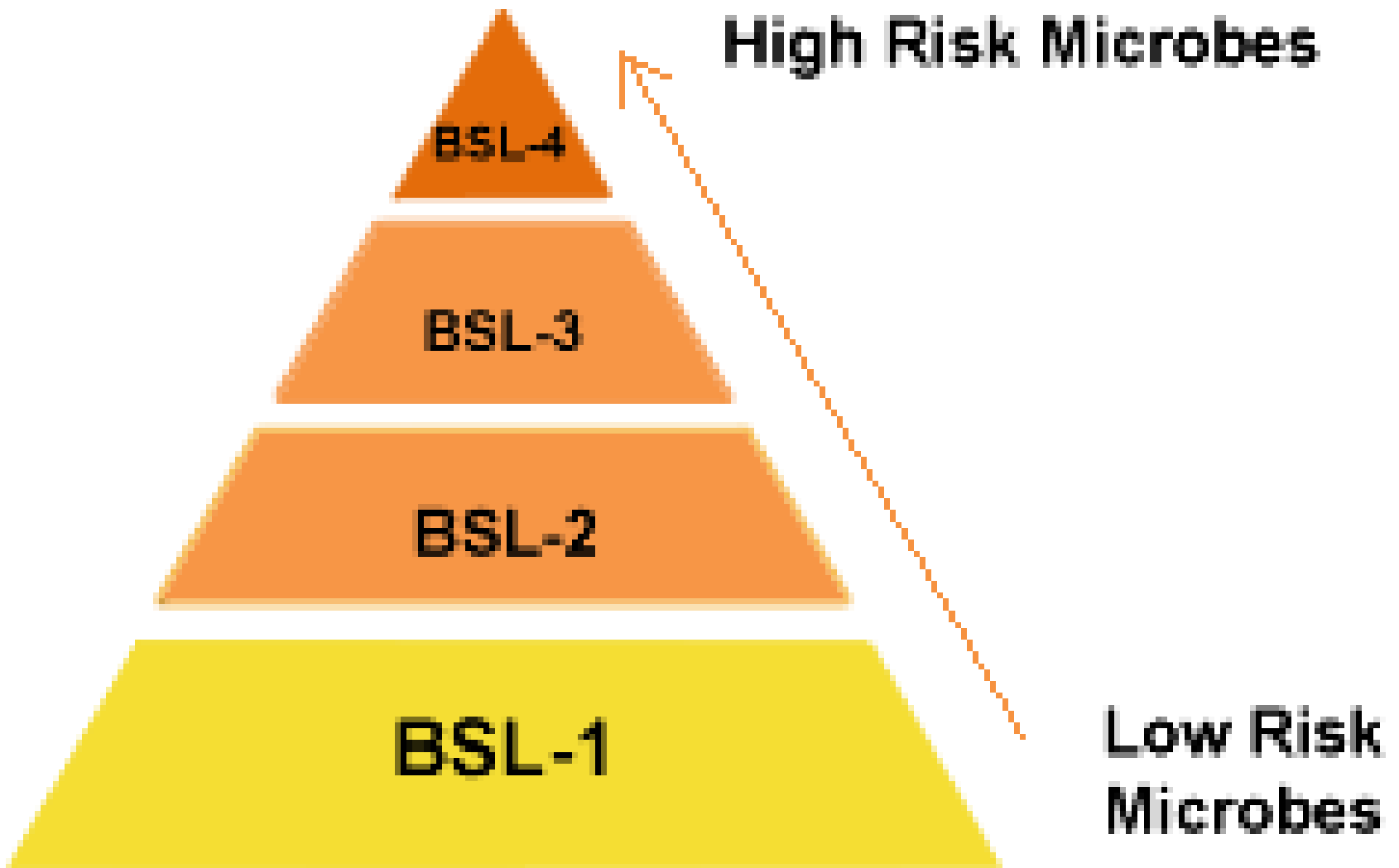


Gradients of dirty



Gradients of dirty





Engineering controls



Appropriate level of assessed risk

Risk Group	Individual risk	Community risk
1	low	no
2	moderate	Limited
3	high	Moderate
4	High	High



BSL Vs RG

There are 4 levels of Laboratory:

Biosafety Laboratory level 1 (BSL 1)

Biosafety Laboratory level 2 (BSL 2)

Biosafety Laboratory level 3 (BSL 3)

Biosafety Laboratory level 4 (BSL 4)

Risk Groups (RG) \neq Biosafety Levels (BSL)

Principles in Biosafety



Each level is appropriate for:

- ❖ Operations performed
(aerosol, solid, liquid, volume)
- ❖ Routes of transmission
- ❖ Lab function or activity
(clinical, Research, isolation and culture)

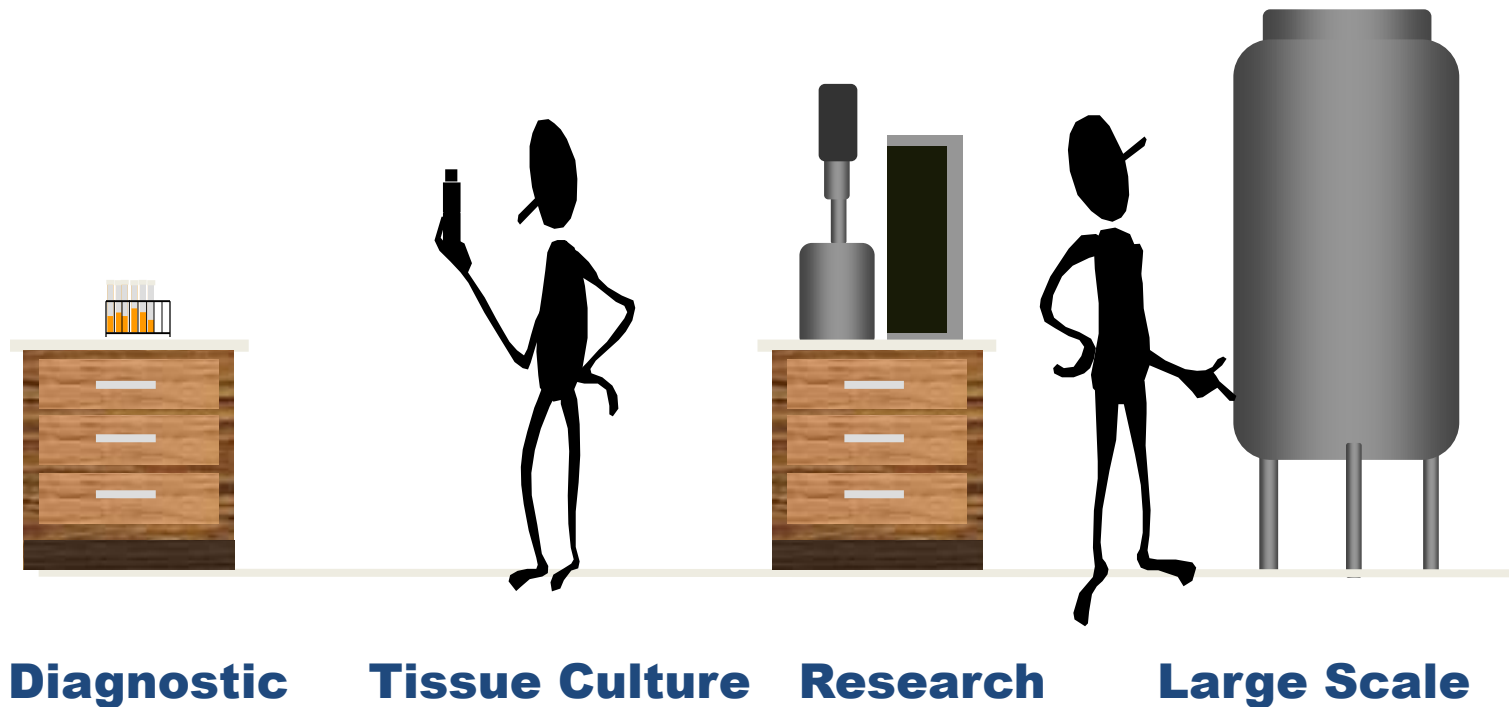


The higher the biosafety level, the more restrictive the laboratory environment will be increasing levels of personnel and environmental protection increasing levels of complexity in SOPs



Scale Can Make a Difference

Larger volumes (10 L) have higher risk



TB Laboratory Risk Assessment

- Handling solid medium vs liquid cultures??
 - Solid media – dry colonies; colony mass not readily dispersed as droplet nuclei



Summary

- 4 BSLs: BSL-1, BSL-2, BSL-3, BSL-4
- Risk Groups (RG) \neq Biosafety Levels (BSL)
- Risk assessment is the key