

Biocontainment Guidelines, Regulations, and Important Considerations

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Larry J. Shelton, Jr., DVM, MPH
DACLAM, DACVPM

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Objectives

- Rules and Regulations related to biocontainment
- Important Considerations

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Acknowledgements and Disclaimer

- Drs. Calvin Carpenter, Alec Hail, and Jim Swarengen
- The views expressed in this presentation are those of the speaker and do not necessarily reflect the official policy or position of the Department of the Navy, Department of the Army, or any agency of the U.S. government.

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Regulations and Guidance

- Biosafety in Microbiological and Biomedical Laboratories (BMBL)
- Biological Select Agent and Toxin Regulations:
 - DHHS
 - USDA
- NIH Guide for the Care and Use of Laboratory Animals
- USDA Animal Welfare Regulations
- Good Laboratory Practice Regulations
- Other Federal, State, and Local Laws and Requirements
- NIH Design Guidelines
- USDA Design Guidelines
- Planning and Designing Research Animal Facilities, ACLAM

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Major Guidance

- Biosafety in Microbiological and Biomedical Laboratories
 - Also known as the "BMBL"
- Biological Select Agent and Toxin Regulations
 - Also known as "BSAT"

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BMBL

- Biosafety in Microbiological and Biomedical Laboratories (BMBL), 5th edition, DEC 2009
- In 1984, the CDC and NIH, a broad collaborative initiative involving scientists, laboratory directors, occupational physicians, epidemiologists, public health officials and health and safety professionals developed the first edition of BMBL
- <http://www.cdc.gov/biosafety/publications/bmb15/index.htm>

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Sections

- Introduction
- Biological Risk Assessment
- Principles of Biosafety
- Laboratory Biosafety Levels (BSLs)
- Animal Biosafety Levels (ABSLs)
- Principles of Laboratory Biosecurity
- Occupational Health and Immunoprophylaxis
- Agent Summary Statements
- Appendices

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Appendices

- Appendix A: Primary Containment for Biohazards: Selection, Installation and Use of Biological Safety Cabinets
- Appendix B: Decontamination and Disinfection
- Appendix C: Transportation of Infectious Substances
- Appendix D: Agriculture Pathogen Biosafety
- Appendix E: Arthropod Containment Guidelines

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Appendices

- Appendix F: Select Agents and Toxins
- Appendix G: Integrated Pest Management (IPM)
- Appendix H: Working with Human, NHP and Other Mammalian Cells and Tissues
- Appendix I: Guidelines for Work with Toxins of Biological Origin
- Appendix J: NIH Oversight of Research Involving Recombinant Biosafety Issues

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Biological Risk Assessment

- Risk groups result of a classification of microbiological agents based on their association with and severity of disease in humans- Risk Groups 1, 2, 3, 4
- Biosafety level represent those conditions under which the agent ordinarily can be safely handled
- Related to the facility, procedures, etc.
 - Laboratories- biosafety Level 1, 2, 3, 4
 - Animal-ABSL 1-4 (remainder of talk limited to ABSL)
 - USDA also has BSL/ABSL-3 enhanced or BSL-3 Ag

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Risk Groups

- Based on WHO and NIH criteria
 - Risk Group 1: Agents not associated with disease in healthy adult humans
 - Risk Group 2: Agents associated with human disease that is rarely serious and preventive or therapeutic interventions are *often available*
 - Risk Group 3: Agents associated with serious or lethal human disease and preventive or therapeutic interventions *may be available (high individual risk but low community risk)*
 - Risk Group 4: Agents that are likely to cause serious or lethal human disease and preventive or therapeutic interventions are *not usually available (high individual risk and high community risk)*

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Biosafety Levels

- Laboratory (1- 4) and Animal Biosafety Levels (1 – 4 + Ag)
- Agent (Classification based on Risk Group)
- Practices (Personnel Controls)
 - *Standard Microbiological Practices*
 - *Special Practices*
- Safety Equipment
 - Primary Barriers/Containment (Biosafety Cabinet)
 - Personnel Protective Equipment
- Laboratory Facilities
 - Secondary Containment (Engineering Controls)

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Examples

- *Bacillus anthracis*
 - Sterne strain: Risk group 2, BSL/ABSL-2
 - Ames strain: Risk group 3, BSL/ABSL-3
- ▶ Sin Nombre Virus Risk Group 3
 - BSL-2 for Diagnostic/Lab Work
 - BSL-2 with BSL-3 practices for handling infected tissue
 - BSL-3 for viral isolation
 - ABSL-2 for animal studies with animals that do excrete the virus
 - ABSL-3 for animal studies with permissive animals

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ABSL Standard Microbiological Practices

- Facility director establishes and enforces policies
- Safety addressed in animal protocol
- Animal protocols reviewed by IACUC and by Safety
- Safety manual specific to animal facility available
- The laboratory supervisor must ensure that laboratory personnel receive:
 - Appropriate training regarding their duties
 - Necessary precautions to prevent exposures
 - Exposure evaluation procedures.
 - Annual updates or additional training when procedural or policy changes occur

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ABSL Standard Microbiological Practices

- An appropriate medical surveillance program in place
 - Determined by risk assessment
 - Include animal allergy considerations
- Medical staff informed on animal specific occupational hazards
- Personal health status may impact an individual's susceptibility to infection, ability to receive immunizations, or prophylactic interventions
- All laboratory personnel and particularly women of child-bearing age should be provided with information regarding immune competence and conditions that predispose them to infection
- Individuals having these conditions encouraged to self-identify for appropriate counseling and guidance
- Respirator users enrolled in respiratory protection program

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ABSL Standard Microbiological Practices

- A sign with the universal biohazard symbol must be posted at the laboratory entrance where infectious agents/animals are present
- The sign must include
 - The animal biosafety level
 - General occupational health requirements
 - PPE requirements
 - Name and phone number of supervisor or other responsible personnel
 - Required entry/exit procedures
 - Identification of the agent is recommended

Security –sensitive agent information should be posted in accordance with the institutional policy.

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ABSL Standard Microbiological Practices

- Control access to the animal room
- All persons with access advised of hazards and safeguards
- PPE
 - Lab coats, gowns or uniforms recommended
 - Gloves worn when handling animals
 - Wash hands when remove after and before leaving area
 - Eye, face, and respiratory protection used per risk assessment
 - Eating, drinking, smoking, handling contact lenses, applying cosmetics, etc. not be permitted
 - Perform all procedures to minimize the creation of splashes and/or aerosols
 - Mouth pipetting is prohibited; mechanical pipetting devices must be used

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ABSL Standard Microbiological Practices

- Incorporate policies for the safe handling of sharps
 - Use sharps only if no alternative
 - Needles must not be bent, sheared, recapped, removed or otherwise manipulated by hand before disposal
 - Plastic ware should be substituted for glassware whenever possible
 - Broken glassware must not be handled directly-must be removed using a brush and dustpan, tongs, or forceps
 - Equipment with sharp edges or corners avoided
- Equipment and surfaces routinely decontaminated
- Animals and plants not associated with research not permitted in animal areas
- An effective integrated pest management program is required.

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ABSL Standard Microbiological Practices

- All wastes (tissues, carcasses, bedding) transported from the room in a leak proof covered container and disposed in compliance with applicable standards
- Decontaminate all potentially infectious materials before disposal

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ABSL-1 Containment

- ABSL-1: In general, a ABSL-1 facility represents a basic level of containment that relies on standard microbiological practices with no special or secondary barriers recommended, other than a sink for hand washing, and self closing and lockable external doors



Animal Biosafety Level 1

- Agent: Not known to consistently cause disease in healthy humans and minimal potential hazard to environment
- Practices: Standard Microbiological Practices
- Primary Barriers and Safety Equipment:
 - Special devices or equipment (BSC's)- none required
 - PPE as indicated by risk assessment
 - Gloves required if using hazardous materials
- Facilities (secondary barriers):
 - Animal facility separate from unrestricted areas
 - External facility-self closing, self locking doors
 - Facility must have a hand washing sink
 - Designed to facilitate cleaning
 - Animal room -self closing inward opening doors

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Animal Biosafety Level 2

- Agent: Associated with human disease that pose moderate hazards to personnel and the environment
- Routes of transmission include percutaneous injury, ingestion, and mucous membrane exposure
- Animal Biosafety Level 2 builds upon ABSL-1. It differs from ABSL-1 in that:
 - Laboratory personnel have specific training in handling infected animals and pathogenic agents
 - Personnel supervised by scientists competent in handling infectious agents and associated procedures;
 - Primary Barriers: **HEPA Filters**- all procedures conducted in BSCs (Class I or II) or other physical containment equipment when infectious aerosols or splashes may be created
 - Secondary barriers – **inward airflow** into animal facility

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ABSL-2

- When a procedure cannot be performed within a biosafety cabinet, a combination of personal protective equipment and other containment devices must be used- risk assess
- Consider the use of restraint devices and practices that reduce risk of exposure during animal manipulations (physical restraint devices, chemical restraint, medications)

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ABSL-2

- When indicated by risk assessment, animals are housed in primary biosafety containment equipment, such as solid wall and bottom cages covered with filter bonnets for rodents, or in inward flow ventilated caging
- Decontamination recommended for all potentially infectious materials and animal waste before movement outside (e.g. autoclave, chemical disinfection)
- Autoclave, or other appropriate type of biohazardous waste treatment available to process infectious wastes
- If vacuum service is provided, requires **HEPA filter**

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Animal Biosafety Level 3

- Indigenous or **exotic agents** with potential for **aerosol transmission**
- Disease may have serious or lethal consequences
- **Preventative or therapeutic interventions may be available**

At ABSL-3, emphasis is placed on primary and secondary barriers to protect personnel, the community, and the environment from exposure to potentially **infectious aerosols**

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Animal Biosafety Level 3

- Primary Barriers: Physical Containment (Biosafety Cabinets)
- Facilities: BSL-2 plus:
 - Physical separation for access corridors, greater controlled access
 - Self-closing, double door access
 - Exhaust air not recirculated
 - **Directional negative airflow into lab/hallway/animal room**
 - **Sealed finishes and penetrations**

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Animal Biosafety Level 3

- Facilities: BSL-2 plus:
 - Primary ventilated enclosure (BSC's, ventilated racks) exhaust must be HEPA filtered
 - Safety mechanisms should be in place that prevent the cages and exhaust plenums from becoming positive to the surrounding area should the exhaust fan fail
 - Exhaust plenums for these systems should be sealed to prevent escape of microorganisms if the ventilation system becomes static
 - The system should also be alarmed to indicate when operational malfunctions occur

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Animal Biosafety Level 3

- Practices: Standard Microbiological Practices, plus:
 - Decontamination of all waste
 - Decontamination of laboratory clothing
 - Occupational Health programs must be implemented
 - Safety Equipment: Personnel Protective Equipment (protective clothing, gloves, **respiratory protection**)

- N95 Respirator
 - Half faced
 - Full faced
- Powered Air Purifying Respirator
 - Positive Air Pressure Respirator



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Animal Biosafety Level 3

- A method for decontaminating all infectious materials must be available within the facility, preferably within the areas where infectious materials are manipulated (e.g. autoclave, chemical disinfection, or other approved decontamination methods)
- All manipulation of infectious materials/animals or the generation of aerosols must be conducted within BSCs or other physical containment devices
- When a procedure cannot be performed within a biosafety cabinet, a combination of personal protective equipment and other containment devices must be used
- Restraint devices and practices are used to reduce the risk of exposure during animal manipulations (e.g., physical restraint devices, chemical restraint medications)

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Animal BSL-3 Ag

- Animal BSL-3 Ag: requires a special type of facility, where the facility barriers, usually considered secondary barriers, now act as primary barriers.

You can't fit a cow in a BSC!

- Examples are sealed interior surfaces, ventilation systems, pathological incinerators, effluent sterilization systems, HEPA filters, etc.

Dedicated Laboratory

Dedicated Shower

Dedicated Autoclave

Anteroom with Controlled Access

Pathological incinerators for the safe disposal of large carcasses

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Animal Biosafety Level 4

- Agent: Dangerous/exotic agents that
 - Pose high risk of life threatening disease
 - Are aerosol-transmitted infections
 - For which no vaccines or treatments are available
 - or related agents with unknown risk of transmission
- Practices: ABSL-3 plus,
 - Clothing change before entering,
 - Shower on exit, and
 - Material decontaminated on exit from facility
- Primary Barriers and Safety Equipment: All procedures conducted in a Class III hood or with a full-body, positive pressure suit and a Class I or II BSC

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Animal Biosafety Level 4

- Facilities: ABSL-3 plus,
 - Separate building or isolated zone
 - Sealed finishes and penetrations
 - Dedicated supply and exhaust, vacuum, and decontamination systems
 - **Redundant Systems**- exhaust HEPA and vacuum, backflow preventers, fans
 - Sequential passage through an inner (dirty) changing area, a personal shower and an outer (clean) change room prior to exiting
 - Drains directly connected to decontamination system
 - Automatic emergency power source
 - Monitoring and control systems essential systems
 - A double-door autoclave, dunk tank, fumigation chamber, or ventilated anteroom/airlock must be provided at the containment barrier for the passage of materials, supplies, or equipment

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ABSL4 Special Practices

- Personnel must enter and exit the laboratory through the clothing change and shower rooms except during emergencies
- All personal clothing must be removed prior to entry
- Laboratory clothing, including undergarments, pants, shirts, jumpsuits, shoes, and gloves, must be used by all personnel entering the laboratory
- Used laboratory clothing must be treated as contaminated materials and decontaminated before removal from suite for laundering or disposal
- All personnel leaving ABSL-4 are required to take a shower

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ABSL4 Special Practices

- Based on site-specific risk assessment, personnel assigned to work with infected animals **may be required to work in pairs**
- Procedures to reduce possible worker exposure must be instituted, such as use of squeeze cages, working only with anesthetized animals, or other appropriate practices
- Infected animals must/should be housed in ventilated enclosures, with inward directional airflow and HEPA filtered exhaust
- Removal of any viable infectious material - in double sealed non-breakable externally decontaminated containers
- Documentation of personnel entry/exit and all facility decontamination procedures required

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Biosafety Level 4

- There are two models for BSL-4 laboratories:
 - A Cabinet Laboratory where all handling of agents must be performed in a Class III BSC
 - A Suit Laboratory where personnel must wear a positive pressure protective suit



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ABSL-4 Cabinet Laboratory

- The ABSL-4 cabinet laboratory consists of either a separate building or a clearly demarcated and isolated zone within a building
- All manipulations of infectious animals and materials must be conducted in the Class III BSC
- Autoclaves must be provided for decontaminating materials passing out of the Class III BSC
- Autoclave doors interlocked so that only one can be opened at any time
- Autoclave doors automatically controlled so that can only be opened after the decontamination has been completed
- The Class III cabinet must also have a pass-through dunk tank, fumigation chamber, or equivalent decontamination method so that materials and equipment that cannot be autoclaved can be safely removed from the cabinet

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ABSL-4 Suit Laboratory

- Entry must be through an airlock fitted with airtight doors
- Personnel must wear a positive pressure suit with HEPA filtered breathing air
- Inner gloves must be worn to protect against break or tears in the outer suit gloves
- Breathing air systems must have redundant compressors, failure alarms and emergency backup
- Chemical shower provided to decontaminate the surface of the positive pressure suit before exit-capable of operating without power

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BMBL Biosecurity

- New Section added in the 5th edition
- The term "biosecurity" refers to the protection of microbial agents from loss, theft, diversion or intentional misuse
- Not to be confused with "animal biosecurity" used in the Guide. "*Animal biosecurity* includes all measures to control known or unknown infections in laboratory animals."
- The recommendations in this section are advisory. Excluding the Select Agent Regulations, there is no current federal requirement for the development of a biosecurity program
 - Select Agent regulations- discussed later in this talk

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BMBL Biosecurity

- Risk Assessment and Management Process
 1. Identify and prioritize biological materials
 - Prioritize based on consequences of misuse
 2. Identify and Prioritize the threat
 3. Analyze the risk of specific security scenarios
 4. Develop and overall risk management plan
 5. Re-evaluate the Institutions risk posture and protection objectives

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BMBL Biosecurity

- Elements of a Biosecurity Program
 - Program Management
 - Physical Security-Access Control and Monitoring
 - Personnel management
 - Integrity of the individuals
 - Inventory and Accountability
 - Information security
 - Policies and procedures to handle sensitive information
 - Transport of Biological Agents
 - Accident Injury and Incident response Plans
 - Reporting and Communication
 - Training and Practice Drills
 - Security Updates and re-evaluations
 - Select Agents- comply with National Select Agent Program

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BMBL Occupational Health

- Laboratory personnel and support staff must be provided appropriate occupational medical service
 - Medical surveillance
 - Available immunizations for agents
 - Periodic medical evaluations
 - Medical support for injuries/illnesses
- Goal is to
 - Limit opportunities for exposure
 - Promptly detect and treat exposures
 - Use information gained to enhance safety procedures
- Provided for all personnel regardless of employment status
- Special Attention for potential spread to outside human and animal populations
 - Address unexplained worker absences
 - Monitoring, evaluation and follow-up of unexplained/non-specific illnesses

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BMBL Agent Summary Statements

- Background
- Occupational Infections
- Natural Modes of Infection
- Laboratory Safety & Containment Recommendations
 - What BSL under what conditions?
- Special Issues:
 - Vaccines available
 - Select agent status
 - Transfer of agent specifics if applicable

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BMBL Appendices

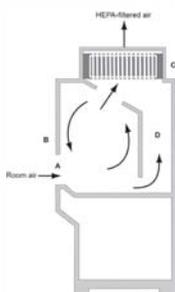
- A- Primary containment- Installation and use of BSCs
- B- Decontamination and Disinfection
- C- Transportation of Infectious Substances
- D- Agriculture Pathogen Biosafety
- E- Arthropod Containment Guidelines
- F- Select Agents and Toxins
- G- Integrated Pest Management
- H-Working with Human, NHP, & Other Mammalian Cells and Tissues
- I- Guidelines for work with Toxins of Biological Origin
- J- NIH Oversight of Research Involving Recombinant Biosafety Issues
- K- Resources
- L- Acronyms

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Biosafety Cabinets (Appendix A)

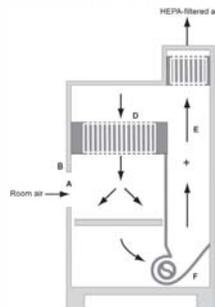
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Class I BSC

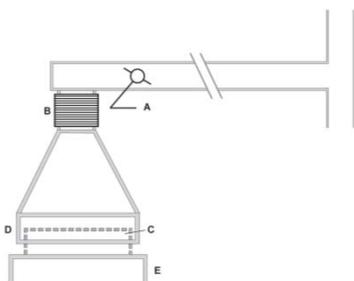


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Class II, Type A1 BSC

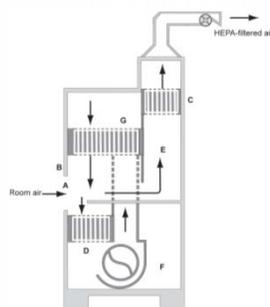


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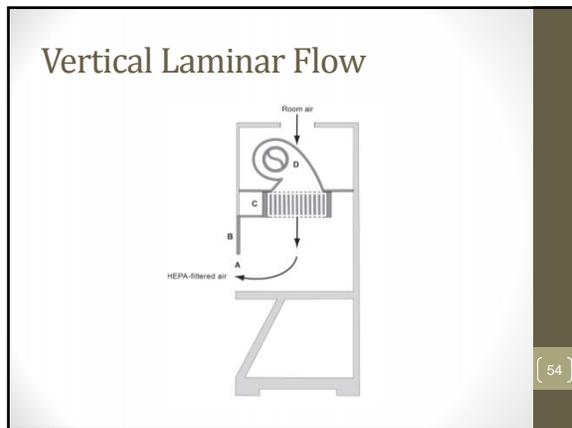
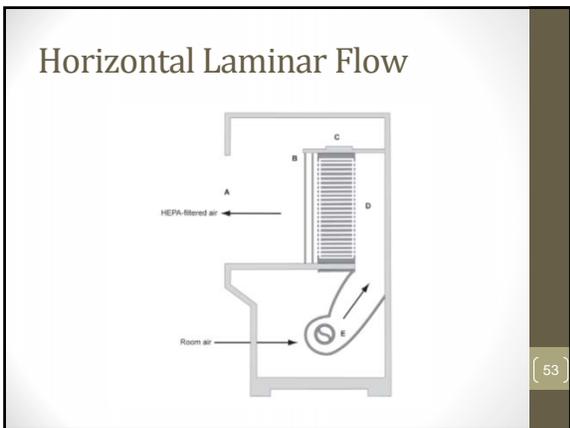
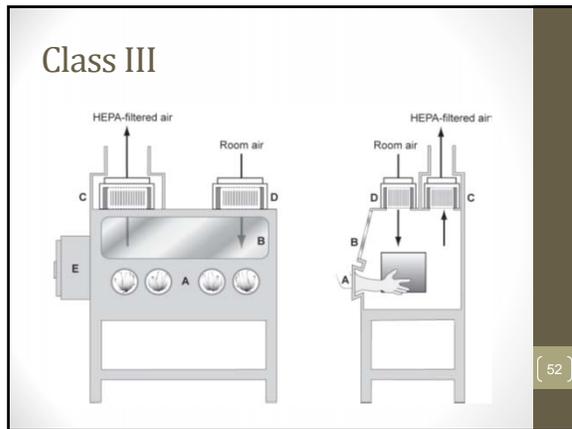
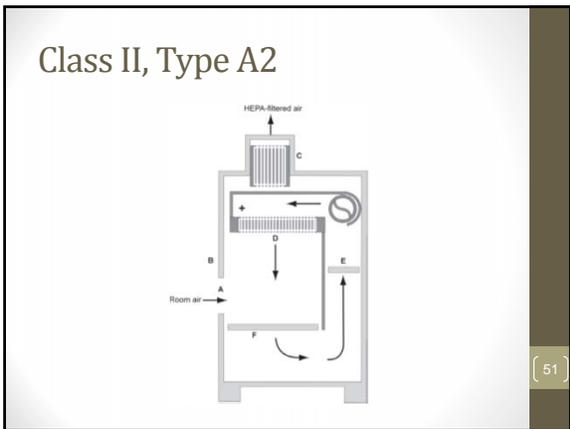
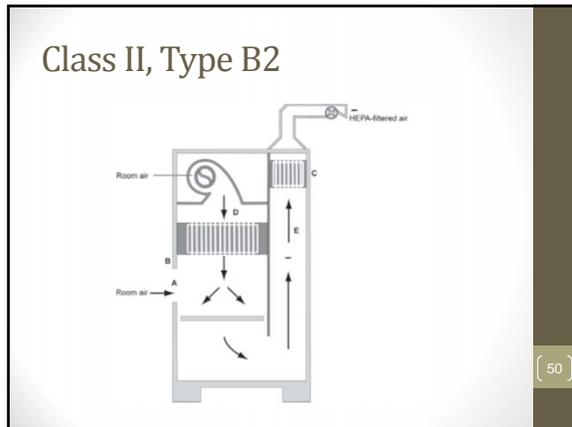
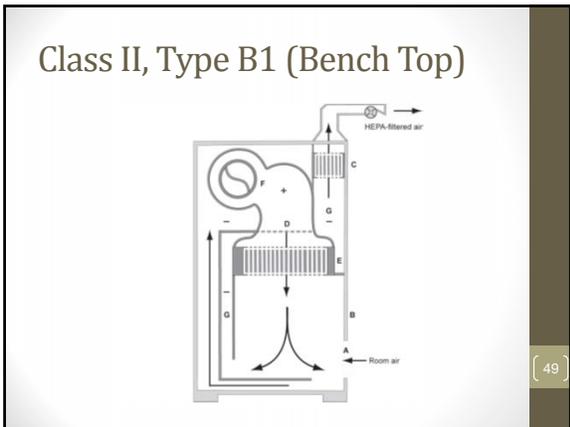


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Class II, Type B1 (Classic)BSC



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A Laminar Flow Bench is NOT a BSC

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Biological Select Agents and Toxins (BSAT)

- Select Agent Program
 - Applies to specific listed biological select agents and toxins
 - <http://www.selectagents.gov/>
- In the wake of the Oklahoma City bombing of the Alfred E. Murrah Building in April 1995, Congress passed the Antiterrorism and Effective Death Penalty Act of 1996. The provisions of this act were implemented by 42 CFR 72.6.
- After the terrorist events of September and October 2001, Congress passed the Uniting and Strengthening America by Providing Appropriate Tools Required to Intercept and Obstruct Terrorism Act of 2001 (USA PATRIOT Act)

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Select Agent Program

- This was followed in June 2002, by passage of Public Health Security and Bioterrorism Preparedness and Response Act of 2002 (Public Law 107-188)
- HHS delegated authority for operating the Laboratory Registration and Select Agents Tracking Program to CDC
- USDA delegated authority for their responsibilities to APHIS
- APHIS and CDC implemented the provisions through a series of regulations that were published in the Federal Register in Dec 2002

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Select Agent Program

- Culminated with the publication of the Select Agents Regulations in the Federal Register in 2005
- This is LAW (not guidelines)
 - 42 CFR Part 73- DHHS, CDC – People protection
 - 7 CFR Part 331- USDA, APHIS- Plant protection
 - 9 CFR Part 121- USDA, APHIS- Animal/Animal Product protection

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Select Agent Program

- The Select Agent List- lists specific bad agents/toxins that could be potentially used by bad people for bad purposes
- HHS (CDC): Biological agents and toxins listed have the potential to pose a severe threat to public health and safety
- USDA (APHIS): Biological agents and toxins listed have the potential to pose a severe threat to animal/plant health or animal/plant products.
- Overlap: Threat to public and animal health

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Select Agent Program

- The Select Agent List is a working document that can be amended as warranted
 - <http://www.selectagents.gov/>
 - HHS, CDC- 34
 - USDA, APHIS
 - Animal -14
 - Plant -7
 - Overlap-11
- Does not include all Risk Group 3 or 4 agents
- Includes some Risk Group 2 agents

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Tier 1 Agents

- Tier 1 agents and toxins present the greatest risk of deliberate misuse with significant potential for mass casualties or devastating effect to the economy, critical infrastructure, or public confidence, and pose a severe threat to public health and safety
- HHS – Eight Agents
Botulinum neurotoxins, Botulinum neurotoxin producing species of Clostridium, Ebola virus, Francisella tularensis, Marburg virus, Variola major virus (Smallpox virus), Variola minor virus (Alastrim), Yersinia pestis
- USDA – Two Agents
Foot-And-Mouth Disease virus and Rinderpest virus
- Overlap – Three Agents-
Bacillus anthracis, Burkholderia mallei, Burkholderia pseudomallei

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Select Agent Program

- Registration
 - Organization
 - Individual labs
- Responsible Official
- Restricted Access/Security Risk Assessment
- Security
- Biosafety
- Exemptions (i.e. agent quantity)
- Restricted Experiments
- Incident/Emergency Response

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Select Agent Program

- Training
- Transfers
- Records
- Inspections
- Notification of Theft, Loss, or Release
- Administrative Review
- Civil and Criminal Penalties

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Personnel Requirements

- Training
 - Insider Threat Awareness
 - Personnel Suitability
- Background Checks
 - Security Risk Assessment
- Occupational Health
- Clearances- “background check”
- Personnel Suitability
 - Pre-access
 - On-going

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Questions?

Contact Information:
Dr. Larry Shelton
Phone: 301-619-9241
Email: larry.j.shelton.mil@mail.mil

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