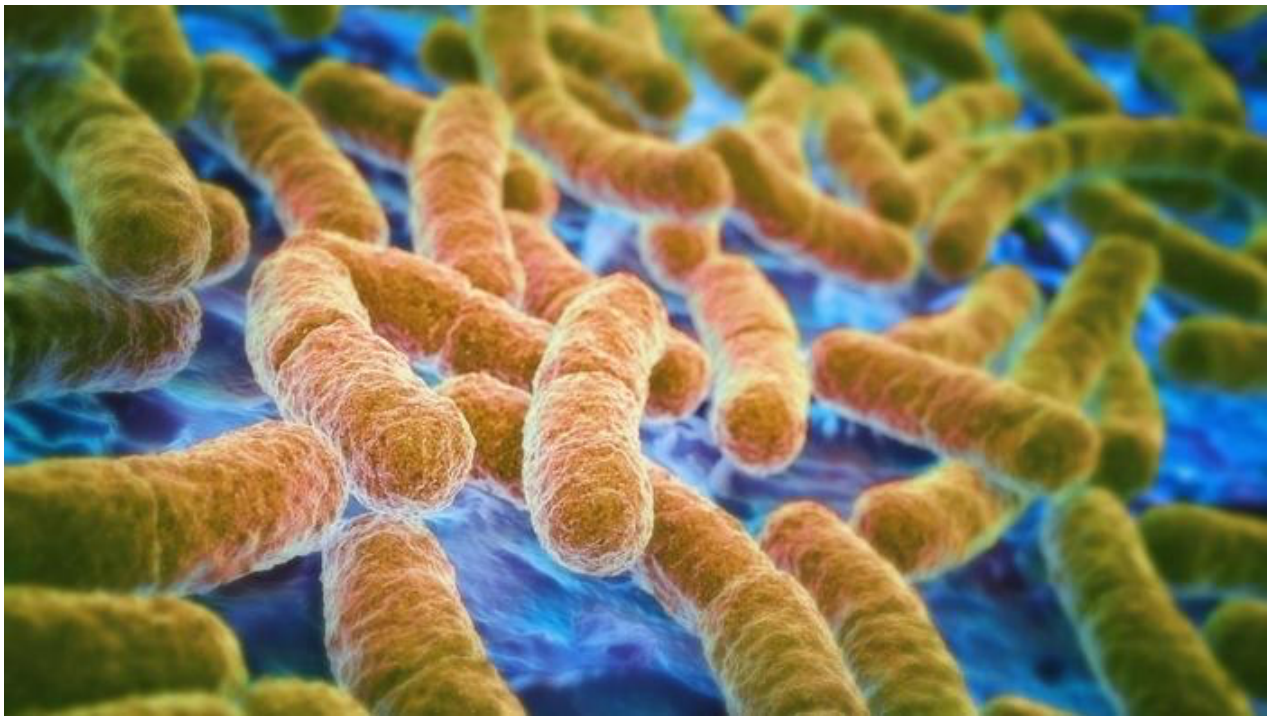
Learning Event 2: Overview of Biological and Chemical Materials in the BSL-3 Laboratory



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Contents

Guidance on Collaborative Review

Dear Subject Matter Experts,

Origin of the Content Outline:

This content outline has been crafted based on our collaborative workshops. The details and structure have been derived from the diverse resources you generously provided, ensuring a foundation that is both comprehensive and reflective of our collective expertise.

Role of the Designer:

As the instructional designer, my responsibility is to organize, structure, and present information in an effective and coherent manner. I take the intricate details of our subject and ensure they are communicated effectively to our intended audience.

Your Expertise:

Your role is crucial. You ensure that the information is accurate, relevant, and up to date. Your deep knowledge helps us maintain the integrity and credibility of our content.

Actionable Feedback Defined:

When reviewing, we kindly ask for "actionable feedback." This means:

1. **Specificity:** Instead of saying "This section isn't clear," it would be helpful to indicate which part is unclear and why.
2. **Suggestions:** If something seems off, please provide an alternative or explain how it can be improved.
3. **References & Resources:** If you believe additional sources or references should be included, please provide the exact details or links.

Your detailed feedback ensures that we create a product that is both authoritative and user-friendly. Thank you for your commitment to excellence.

Modality

1-2 Hour(s) eLearning: Overview of Biological and Chemical Materials in the BSL-3 Laboratory

Asynchronous e-learning modules will provide participants with the foundational knowledge and information they need to understand critical concepts related to Biological and Chemical Materials in BSL-3 laboratories. The modules will include interactive activities, videos, and quizzes to reinforce key concepts and assess participants' understanding of the material. This will allow them to review the material at their own pace and deepen their understanding of the concepts. Existing assets will be utilized to design and develop eLearning modules to provide a comprehensive understanding of biosafety practices in laboratory settings.

Topics to be Covered

- ✓ Characteristics of infectious biological agents
- ✓ Most common pathogens and unique considerations
- ✓ Risk groups for infectious biological agents
- ✓ Biosafety levels and common enhancements
- ✓ Control measures for infectious biological agents
- ✓ Hierarchy of controls used when working with infectious biological agents
- ✓ Risks associated with common hazardous chemicals.

Note: The list of topics included was generated during the content outlining phase of our curriculum design process. This was based on initial discussions and planning sessions aimed at ensuring comprehensive coverage and relevance to our objectives.

Course Learning Objectives

Lesson 1: Characteristics of Infectious Biological Materials

- Describe inherent characteristics of infectious biological agents handled in the BSL-3 laboratory
- Identify routes of transmission, environmental stability, signs and symptoms of infection, incubation times, and susceptibility to disinfectants for infectious biological agents handled in a BSL-3 laboratory
- Categorize infectious biological agents into risk groups

Lesson 2: Control Measures for Infectious Biological Materials

- Describe biosafety levels and common enhancements
- Describe control measures used when working with infectious biological agents

Lesson 3: Hazardous Chemical Characteristics and Control Measures

- Describe the risks associated with common hazardous chemicals handled in the BSL-3 laboratory
- Describe control measures used when working with hazardous chemical materials
- Identify appropriate selection, preparation, and use of chemical disinfectants for work in the BSL-3 laboratory

Note: The list of objectives provided here was formulated during the content mapping and content outline phases of our development process. This approach ensures that our objectives are thoroughly planned and aligned with the overall educational goals.

Overview of Biological and Chemical Materials in the BSL-3 Laboratory

Lesson 1: Characteristics of Infectious Biological Materials

Infectious Biological Agents in BSL-3 Laboratories

Infectious biological agents requiring BSL-3 containment include those that can cause serious or potentially lethal diseases through inhalation. These agents often have low infectious doses and no known treatment or vaccines, necessitating stringent safety measures to prevent laboratory-acquired infections (LAIs).

The following are important considerations for biological agents requiring BSL-3 containment:

Infectivity	Ability to enter and multiply in the host	High
Pathogenicity	Ability of an organism to cause disease in a host.	High
Virulence	Ability to cause severe disease once an infection occurs.	Severe
Toxicity	The extent to which a substance produced by a pathogen is harmful.	Low but can cause severe or chronic disease
Invasiveness	Ability to penetrate and spread throughout the tissue	Usually moderate or high
Antigenicity	Ability to stimulate an immunological response	Usually moderate or high

Characteristics of Infectious Biological Agents

When making safety, handling, and equipment-related decisions about infectious biological agents, personnel in BSL-3 laboratories must consider various characteristics of the agents.

- Prevalence
- Environmental Stability
- Susceptibility to Disinfectants
- Modes of Transmission
- Host Range
- Infectious Dose
- Incubation Time
- Signs and Symptoms of Infection
- Methods of Inactivation
- Preventative Measures

- Treatments

Classification System for Infectious Biological Agents

Hierarchy of Susceptibility

The **hierarchy of susceptibility** of biological infectious agents refers to the varying degrees of risk posed by different pathogens based on their transmissibility, virulence, and potential impact on public health. This hierarchy helps in categorizing infectious agents into different biosafety levels (BSL), with BSL-3 designated for agents that can cause serious or potentially lethal disease through inhalation.

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Risk Groups

Biological agents are classified into four Risk Groups (RGs) according to their relative pathogenicity and risk of infection for healthy adult humans by the following criteria:

Risk Group 1 (RG1)	A **Risk Group 1 (RG1**) agent is not associated with disease in healthy adult humans.
Risk Group 2 (RG2)	A **Risk Group 2 (RG2)** agent can cause human disease and might be a hazard to employees. It is unlikely to spread to the community, but if it does, there is usually effective prophylaxis or treatment available.
Risk Group 3 (RG3)	A **Risk Group 3 (RG3**) agent can cause severe human disease and is a serious hazard to employees. It may present a risk of spread to the community. If it does spread to the community, there is usually effective prophylaxis or treatment available.
Risk Group 4 (RG4)	A **Risk Group 4 (RG4**) agent can cause severe human disease and is a serious hazard to employees. It may present a high risk of spread to the community. There is usually no effective prophylaxis or treatment available.

Applying these criteria, laboratory personnel can categorize pathogens into their proper risk group.

Subjectivity of Risk Groups

Risk assessment is ultimately a subjective process. The investigator must make an initial assessment based on the Risk Group (RG) of an agent.

Schedule 1 of the Code of Practice provides a non-exhaustive classification list of biological agents. Only agents in hazard groups 2 – 4 are listed. If the agent you are working with is not on the list, you will need to determine the categorization based on the criteria previously covered. Biological agents should not be automatically categorized into hazard group 1. The Office of Laboratory Science and Safety (OLSS) takes a much more active role in activities that involve RG3 and RG4 agents than they do for RG2.

Note that if the biological agent to be assessed cannot be classified clearly in one of the aforementioned groups, it shall be classified in the highest risk group among the alternatives.

Biosafety Levels vs. Risk Groups

The four Biosafety Levels (BSL-1, BSL-2, BSL-3, and BSL-4) are different from Agent Risk Groups (RG1, RG2, RG3, and RG4).

An agent in a higher Risk Group should not automatically be handled in a correspondingly high BSL.

Note the following agents in Risk Group 2 and Risk Group 4 that are handled at BSL-3 rather than BSL-2 or BSL-4.

RG4 Agents (severe pathogenicity, easy transmission, lack of effective treatments)	**BSL-3 Containment**
Nipah virus	Nipah virus work can be performed at BSL-3 if the procedures do not generate aerosols and if additional safety measures, such as enhanced personal protective equipment and engineering controls, are in place.
Hendra virus	Some non-aerosolizing procedures involving Hendra virus might be conducted at BSL-3 with stringent biosafety precautions, including the use of respiratory protection and other enhanced safety protocols.

RG2 Agents (moderate individual risk, low community risk)	**BSL-3 Containment**
Coxiella burnetii	Requires BSL-3 containment when handled in the laboratory because it is highly infectious through aerosol transmission and has a very low infectious dose.
Mycobacterium bovis (BCG strain)	Requires BSL-3 containment due to its potential to cause serious disease and the need for enhanced safety measures to prevent aerosol transmission in a laboratory setting.

Pathogens in BSL-3 Laboratories

Certain pathogens require BSL-3 containment measures due to their potential for causing severe diseases, the type of work or research being done, and the risks associated with exposure.

Bacillus Anthracis (Anthrax)

Description	Causative agent of Anthrax
Select Agent	Yes
Work Focus	Pathogenesis; vaccine development, including production and testing of recombinant proteins and live-attenuated strains to induce protective immunity; therapeutic interventions; genetic studies; virulent and attenuated

	strains to evaluate the effectiveness of new antibiotics, antitoxins, and other therapeutic measures.
Prevalence	Occurs globally among wild and domestic animals. Sporadic outbreaks occur in agricultural areas of Africa, Asia, and the Americas. Rare human cases through occupational exposure to infected animals or animal products, or through bioterrorism.
Environmental Stability	Extremely stable in the environment and can survive for decades in soil and animal products. The spores are resistant to heat, desiccation, UV light, and chemical disinfectants, making them a persistent threat in contaminated areas. This high stability is a significant factor in the potential use of anthrax as a biological weapon.
Susceptibility to Disinfectants	Spores are highly resistant and can remain viable on some surfaces for years; effective agents include chlorine dioxide and formaldehyde.
Modes of Transmission	Inhalation of spores; contact with contaminated animal products; ingestion; cutaneous (via broken skin); injection
Host Range	Humans; wild and domestic animals, such as cattle, sheep, and goats
Infectious Dose	Varies by species and route; inhalation LD50 for humans is 2,500-55,000 spores, while cutaneous anthrax can occur with as few as 10 spores
Incubation Time	Varies depending on the route of infection; inhalation anthrax typically 1-7 days; cutaneous anthrax 1-12 days; and gastrointestinal anthrax 1-7 days
Signs & Symptoms of Infection	Cutaneous: ulcerative lesions; Inhalation: flu-like symptoms progressing to severe respiratory distress; Gastrointestinal: severe abdominal pain, vomiting, and diarrhea. May also cause fever, cough, chest discomfort, shortness of breath, and muscle aches
Inactivation Methods	Autoclaving, incineration, chlorine dioxide, peracetic acid, formaldehyde
Preventative Measures	Pre-exposure vaccination (anthrax vaccine adsorbed - AVA) for high-risk individuals; environmental controls to reduce exposure to contaminated animal products.
Treatments	Combination of antimicrobials and vaccines post-exposure; numerous other therapeutic strategies are being explored.

Mycobacterium Tuberculosis complex

Description	Causative agent of tuberculosis
Select Agent	No
Work Focus	Examine genetic makeup to understand mechanisms of infection and resistance; develop and test new antibiotics and vaccines. Investigate the bacterium's interaction with the host immune system to identify potential therapeutic targets. Evaluate advanced diagnostic methods to improve the detection and management of tuberculosis infections.
Prevalence	Global; highest burden in Southeast Asia, Africa, and the Western Pacific. Common in areas with high HIV prevalence and limited access to healthcare (developing countries).
Environmental Stability	Known for high environmental stability. Can survive desiccation and resist disinfectants for weeks to months. Can remain viable on surfaces and in

	aerosols for extended periods, especially under conditions of low humidity and absence of direct sunlight.
Susceptibility to Disinfectants	Resistant to many disinfectants, including bleach (sodium hypochlorite), hydrogen peroxide, phenolics, alcohols, and glutaraldehyde. Requires appropriate concentration and contact time to ensure efficacy.
Modes of Transmission	Inhalation of airborne droplets; direct contact with contaminated surfaces; ingestion of contaminated milk; handling of infected tissues
Host Range	Primarily affects humans but can also affect animals such as cattle, goats, and non-human primates
Infectious Dose	Very low; as few as 1-10 organisms
Incubation Time	Commonly 2 to 12 weeks; active disease can develop within months of exposure, while latent infections can persist for years before reactivating
Signs & Symptoms of Infection	Persistent cough, chest pain, hemoptysis (coughing up blood), fever, night sweats, weight loss, fatigue
Inactivation Methods	Heat (autoclaving), UV radiation, bleach (sodium hypochlorite), alcohol, hydrogen peroxide, phenolic disinfectants
Preventative Measures	Bacille Calmette-Guérin (BCG) vaccine is available and used in many countries, but not generally recommended in the U.S.; preventive measures include TB skin testing and interferon-gamma release assays (IGRAs).
Treatments	Treatment involves prolonged courses of antibiotics, typically isoniazid, rifampin, ethambutol, and pyrazinamide; MDR and XDR strains require second-line drugs and more complex regimens.

For information on other pathogens commonly used at BSL-3, refer to

- Biosafety in Microbiological and Biomedical Laboratories (BMBL) 6th Edition
- American Biological Safety Association (ABSA) Fact Sheets
- Canadian Pathogen Safety Data Sheets

Lesson 2: Control Measures for Infectious Biological Materials

Biosafety and Biosecurity

Biosafety refers to principles, practices, and procedures implemented to prevent unintentional exposure to biological agents and toxins, or their accidental release. Biosafety protects laboratory personnel, the environment, and the public.

Biosecurity involves strategies and practices designed to protect against the theft, misuse, or intentional release of harmful biological agents and toxins. It focuses on preventing the deliberate introduction of pathogens or biotoxins into the environment.

Overview of BSL-3 Laboratories

BSL-2 vs. BSL-3

BSL-2 involves agents that pose moderate hazards to personnel and the environment (e.g., Staphylococcus aureus, Hepatitis B virus). Personnel working in a BSL-2 laboratory wear basic PPE, implement standard practices, and use Class I or II biological safety cabinets.

BSL-3 involves work with indigenous or exotic agents that can cause serious or potentially lethal diseases through inhalation (e.g., Mycobacterium tuberculosis, SARS-CoV-2). Personnel working in a BSL-3 laboratory wear enhanced PPE, implement more stringent practices, and use Class II or III biological safety cabinets.

BSL-3 Enhancements

BSL-2 and BSL-3 laboratories have the same standard microbiological practices. However, in BSL-3, where biological and chemical hazards increase significantly, enhanced measures ensure higher levels of safety and containment.

The following are safety and containment requirements of BSL-3 laboratories beyond those of BSL-2.

Access Controls

- Restricted access: only persons whose presence in the facility or laboratory areas is required for scientific or support purposes are authorized to enter.
- Access to the laboratory is through **two** consecutive self-closing doors.
- The laboratory is separated from areas that are open to unrestricted traffic flow within the building.
- Some laboratories have advanced access control devices, such as biometrics.

Entry/Exit Procedures

- All persons entering the laboratory are advised of the potential hazards and meet specific entry/exit requirements in accordance with institutional policies.
- All persons who enter operational laboratory areas are provided information on signs and symptoms of disease and receive occupational medical services including medical evaluation, surveillance, and treatment, as appropriate, and offered available immunizations for agents handled or potentially present in the laboratory.

Personal Protective Equipment

- Laboratory personnel wear protective clothing with a solid-front (tie-back or wrap-around gowns, scrub suits, or coveralls).
- Protective clothing is not worn outside the laboratory.
- Reusable clothing is decontaminated before being laundered.
- Clothing is changed when contaminated.
- Two pairs of gloves and shoe covers are worn when appropriate.
- Respiratory protection is considered, along with enrollment in a properly constituted respiratory protection program.

Handwashing and Eye-Washing Stations

- Handwashing sinks in BSL-3 laboratories are hands-free and operate automatically.
- If a laboratory suite is segregated into different zones, a sink is available for handwashing in each zone.

Biosafety Cabinets

- Class II and III Biosafety Cabinets are used in BSL-3 laboratories.
- Class II BSCs have HEPA-filtered supply and exhaust air and vertical air flow to minimize contamination.

Air Handling Systems

- Equipment that may produce infectious aerosols is used within primary barrier devices that exhaust air through HEPA filtration or other equivalent technology before being discharged into the laboratory. These HEPA filters are tested annually and replaced as needed.
- A ducted mechanical air ventilation system is required. This system provides sustained directional airflow by drawing air into the laboratory from “clean” areas toward “potentially contaminated” areas. The laboratory is designed such that under failure conditions the airflow will not be reversed at the containment barrier.
- When present, HEPA filter housings have gas-tight isolation dampers, decontamination ports, and/or bag-in/bag-out (with appropriate decontamination procedures) capability.

Specialized Training

- Laboratory personnel receive specific training in handling pathogenic and potentially lethal agents. They are supervised by scientists competent in handling infectious agents and associated procedures.
- Training includes annual updates and additional training when equipment, procedures, or policies change.

Safety Manual & Emergency Response

- The safety manual contains protocols for emergency situations including exposures, medical emergencies, laboratory malfunctions, and other potential emergencies.
- Personnel receive training in emergency response procedures.

Incident Reporting

- A system is established for reporting and documenting near misses, laboratory accidents, exposures, unanticipated absences due to potential LAIs, and for the medical surveillance of potential LAIs.
- Incidents that result in exposure to infectious materials are immediately evaluated per institutional policy. All such incidents are reported to the laboratory supervisor, as with BSL-2, but also institutional management, and appropriate safety, compliance, and security personnel according to institutional policy.

BSL-3 Laboratories at the Roybal Campus

The Roybal Campus of CDC houses many BSL-3 laboratories that each focus on a particular concern, as shown:

- Highly infectious viruses
- Influenza
- Tuberculosis
- Mosquito-borne viruses
- Agents risky for bioterrorism
- Gastrointestinal diseases
- Parasitic infections
- Bacterial pathogens
- Respiratory illnesses
- Environmental microbes
- Fungal infections
- Poxviruses and rabies
- HIV/AIDS
- Vector-borne diseases
- Sexually transmitted infections

Hierarchy of Controls

The **hierarchy of controls** is a systematic approach used in BSL-3 laboratories to minimize the risks associated with handling dangerous pathogens. This hierarchy prioritizes the implementation of control measures based on their effectiveness, starting with the most effective methods and moving to the least effective.

At the top of the hierarchy are elimination and substitution, which are often not feasible in BSL-3 settings due to the necessity of working with the specific pathogen. Therefore, the focus is on engineering controls, administrative controls, and personal protective equipment (PPE). Engineering controls, such as HEPA-filtered ventilation systems, negative air pressure, and biosafety cabinets, provide physical barriers that contain the pathogens and prevent their release into the environment.

Administrative controls are the next layer in the hierarchy and involve policies and procedures designed to reduce the risk of exposure. This includes rigorous training for laboratory personnel, standard operating procedures for safely handling and disposing of infectious agents, and protocols for emergency response in case of accidental exposure or spills.

Personal protective equipment (PPE) forms the final layer of defense, providing individual protection to workers. In a BSL-3 laboratory, this typically includes laboratory coats, gloves, face shields, and respiratory protection.

Lesson 3: Hazardous Chemical Characteristics and Control Measures

Characteristics of Hazardous Chemicals

Hazardous chemicals are substances that pose significant risks to health, safety, and the environment due to their inherent properties. These characteristics include

- **Toxicity:** can cause acute or chronic health effects such as respiratory issues, skin irritation, and organ damage upon exposure
- **Corrosiveness**: can cause severe burns and tissue damage on contact with skin or eyes
- **Flammability or explosiveness:** present fire and explosion hazards if not handled and stored correctly
- R**eactivity**: reactive chemicals can undergo dangerous reactions when exposed to certain conditions, such as heat, light, or other chemicals, leading to potentially violent outcomes
- **Carcinogenicity**: can cause cancer
- **Mutagenicity**: can cause genetic mutations
- **Teratogenicity:** can cause developmental defects

Common Hazardous Chemicals in BSL-3 Laboratories

In BSL-3 laboratories, a number of chemicals are used for research, or for disinfection, decontamination, and inactivation. With exposure, all pose safety risks that laboratory personnel should be aware of.

Chemical	Use for Inactivation	Hazard
Formaldehyde	Inactivates viruses and bacteria by cross-linking their proteins and nucleic acids, killing them and making them unable to replicate. This method is often used in vaccine production.	• Carcinogenic cancer-risk • Respiratory irritation (coughing, wheezing, difficulty breathing) • Skin irritation (redness, itching, dermatitis) • Eye Irritation (redness, tearing, and burning)
Ethanol	Disrupts the lipid membranes of microbes, leading to their rapid denaturation and destruction. Commonly applied to surfaces and equipment through spraying or wiping, ensuring thorough decontamination. Its volatility ensures quick evaporation, reducing the risk of residue and contamination.	• Highly flammable: significant fire hazard • Respiratory irritation (coughing, sore throat, shortness of breath) • Central nervous system effects (dizziness, headache,

		drowsiness, unconsciousness) • Skin and eye irritation (redness, dryness, burning)
Hydrogen Peroxide	Oxidizing properties inactivate a wide range of pathogens, including bacteria, viruses, and spores. Often used in vaporized form for decontaminating rooms and equipment.	• Oxidizing agent: can react violently with combustible or flammable materials, leading to fire or explosions • Skin and eye irritation:(eye/skin burns, redness, pain, tissue damage) • Respiratory irritation: (coughing, sore throat, difficulty breathing) • Gastrointestinal distress: ingestion of hydrogen peroxide can cause nausea, vomiting, and abdominal pain

Although these chemicals are used at BSL-3, they may also be used at other biosafety levels.

Other chemicals to be aware of can be found at:

Robotics

Robotics in BSL-3 laboratories enhance safety in handling hazardous biological agents by minimizing direct human interaction. This reduces the risk of exposure and contamination. These robotic systems are designed to perform a variety of tasks, including sample handling, processing, and decontamination, with high precision and consistency.

To ensure the safety and functionality of these robotic systems, hazardous chemicals are employed for the decontamination and sterilization of both the equipment and the biological agents they handle. Chemicals such as formaldehyde, hydrogen peroxide, and chlorine dioxide are used to effectively inactivate pathogens on robotic surfaces and components, ensuring that any biological agents are neutralized before maintenance or human intervention.

This rigorous chemical decontamination process not only protects laboratory personnel but also maintains the integrity and performance of the robotic systems, ensuring they operate in a safe and

sterile environment. The integration of robotics with stringent chemical decontamination protocols represents a significant advancement in biosafety, enhancing both safety and efficiency in high-containment laboratory settings.

Handling Chemicals in BSL-3 Laboratories

The use of hazardous chemicals in BSL-2 and BSL-3 laboratories differs in the level of containment and safety measures required. This is because of varying risk associated with the biological agents handled.

In BSL-2 laboratories, hazardous chemicals are used for disinfection and sterilization to manage moderate-risk agents that can cause human disease but are typically not severe and have available treatments. Common chemicals include ethanol, sodium hypochlorite (bleach), and phenol, which are used with standard safety protocols such as gloves, lab coats, and basic ventilation systems.

In contrast, BSL-3 laboratories handle high-risk agents that can cause severe or potentially lethal diseases through inhalation, necessitating more stringent chemical usage and safety practices. Hazardous chemicals like formaldehyde, glutaraldehyde, and chlorine dioxide are employed for comprehensive decontamination. This requires specialized equipment such as biosafety cabinets and advanced ventilation systems to prevent aerosolized pathogens from escaping.

Additionally, personnel in BSL-3 labs must use enhanced personal protective equipment (PPE), including respirators and full-body suits, and undergo rigorous training in chemical handling and emergency procedures to ensure a higher level of protection against the more dangerous agents present.

Planning and Preparation

Since fume hoods are not available for containment, BSL-3 laboratory personnel must plan and prepare for handling hazardous solutions and chemicals.

- Ensure certified biosafety cabinets are in place
- Understand properties of chemicals
- Be aware of potential interactions
- Know specific protocols to follow

Autoclaving

In BSL-3 laboratories, hazardous chemical waste cannot be autoclaved for a variety of reasons:

- High temperatures and pressures involved in autoclaving can cause hazardous chemicals to become volatile or react violently, posing significant safety risks.
- Many chemicals can degrade under autoclave conditions, potentially producing toxic fumes or dangerous byproducts that could harm personnel or damage equipment.
- Certain chemicals can corrode or damage the autoclave's materials, leading to costly repairs and potential contamination of future loads.
- Some chemicals may not be effectively neutralized by autoclaving, leaving residual hazardous waste that still requires proper disposal.

Removal

Proper disposal of hazardous chemical waste requires specialized procedures and equipment to ensure safety and compliance with regulatory standards.

BSCs

In BSL-3 laboratories, hazardous chemicals may require use of biosafety cabinets.

BSCs

Hazardous chemicals at BSL-3 that require a biosafety cabinet are those that pose severe health risks through inhalation, contact, or accidental ingestion, and can cause significant harm or death even in small quantities. These chemicals include potent biological toxins, such as botulinum toxin, ricin, and aflatoxins, which can cause paralysis, organ failure, or cancer. Other examples are

Chemical	Reason for BSC use
Acrylamide	While preparing acrylamide solutions, especially in powdered form, there is a risk of inhaling the neurotoxic and potentially carcinogenic particles. A BSC helps contain any dust or aerosolized particles, reducing the risk of exposure.
Formaldehyde	Formaldehyde is used for tissue fixation and as a disinfectant. It releases fumes that are toxic and carcinogenic. A BSC provides proper ventilation to capture and filter these fumes, protecting the user from inhalation.
Phenol-Chloroform	Used in nucleic acid extraction, both phenol and chloroform are highly toxic and can cause severe burns. Phenol is also volatile, and inhaling its vapors is dangerous. A BSC helps in containing vapors and aerosols, preventing inhalation and skin contact.
Beta-mercaptoethanol	It is highly toxic and volatile, emitting harmful vapors. When used in procedures like protein denaturation, a BSC is necessary to protect the user from inhaling these toxic fumes.

Resources

BMBL (Biosafety in Microbiological and Biomedical Labs 6th)

Pathogen Safety Data Sheets - Canada.ca

Defining Hazardous Waste

Biosafety Manual (cdc.gov)

Biological Agents Code of Practice

Hierarchy of Controls

Risk groups | my.ABSA.org - For the Biosafety and Biosecurity Professional

NIH Guidelines for Research Involving Recombinant or Synthetic Nucleic Acid Molecules (NIH Guidelines)

https://www.who.int/csr/resources/publications/biosafety/WHO_CDS_CSR_LYO_2004_11.pdf

CDC Lab Standards

CDC BLOODBORNE PATHOGENS PROGRAM

Biocontainment - eCFR :: 7 CFR 33.12

HHS Occupational Health and Safety Manual

NCIRD BSL-3 Safety Training: Online

Defining Hazardous Waste: Listed, Characteristic and Mixed Radiological Wastes | US EPA

Introduction to Hazardous Waste Identification (epa.gov)

OSHA Regulation 1910.119 from CFR Table 29 - Process safety management of highly hazardous chemicals.

OSHA Regulation 1910.120 from CFR Title 29 - Hazardous waste operations and emergency response

HW Listing Reference Document - Sep 2012(F).pdf (epa.gov)

Occupational Exposure to Hazardous Chemicals in Laboratories

OLSS_ChemicalHygienePlan_CDC-001-00354_rev3_final.pdf

Chemical Safety | Safety Practices | OSSAM (cdc.gov)

Chemical Storage Guidelines (cdc.gov)

Selected EPA-Registered Disinfectants | US EPA