

WRIGHT STATE UNIVERSITY

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Department of Environmental Health and Safety

**Laboratory Safety Manual**

**Chemical Hygiene Plan**

Laboratory Safety

## Laboratory Safety Manual

### Chemical Hygiene Plan

*Changing Lives -- Safely*

© Wright State University  
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**Environmental Health and Safety**

**047 Biological Sciences II**

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## 1 Plan Authority

**Table 1. Plan Authority**

AUTHORITY	DATE
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## 2 Version History

**Table 2. Version History**

VERSION HISTORY			
Version	Description of Change	Author	Effective Date
Basic	Original	Original Version	1993
1	Comprehensive revision and format updates	M. Markopoulos	2015

## 3 Reference Documents

Document	Title
	Access to Employee Records
	Ohio Revised Code Chapter 4167, "Public Employee Risk Reduction Program (PERRP)."
	Ohio Revised Code Chapter 3701, "Radiation Control"

**If any process in this document conflicts with any document in ORC or OSHA this document shall be superseded by the ORC or OSHA document. Any reference document external to ORC and/or OSHA shall be monitored by the Plan Owner for current versioning.**

# Laboratory Safety Introduction

## 4 Laboratory Safety Introduction

### 4.1 Preface

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*“First and foremost, the protection of health and safety is a moral obligation. An expanding array of federal, state and local laws and regulations makes it a legal requirement and an economic necessity as well. In the final analysis, laboratory safety can be achieved only by the exercise of judgment by informed, responsible individuals. It is an essential part of the development of scientists that they learn to work with and to accept the responsibilities for the appropriate of hazardous substances.”<sup>1</sup>*

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Our university community is responsible for ensuring that all research and related activities are conducted with minimal hazard to employees, students and the community. The procedures described in this manual are elements essential to our program. Anyone using equipment and facilities of this institution is expected to follow accepted procedures, to report all accidents promptly and bring to their supervisor’s attention any unsafe conditions or practices.

This manual provides members of the Wright State University community with information on the inherent risks associated with laboratory work and suitable safeguards. The policies described have been designed to assist department heads and supervisors in meeting their responsibilities for controlling hazardous situations. Placing these policies and procedures into practice is the responsibility of those not only in administrative positions, but in all positions throughout our organization. It is essential that everyone be thoroughly familiar with this manual and knows who to ask for additional advice and training.

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<sup>1</sup> National Research Council, 1981. Prudent Practices for Handling Hazardous Chemicals in Laboratories, Washington, D.C., National Academy Press, p. 6



Members of the Department of Environmental Health & Safety will assist in developing procedures for the safe handling, containment, and disposal of biological, chemical and radiological agents, as well as in designing safe working environments, selecting and using personal protective equipment and interpreting safety standards. Introduction

A variety of hazards are endemic to laboratories and exposure to them has resulted in illness and injury. In some cases, the sources of these exposures have been identified; but in most cases, they have not. Assume that you may be exposed to certain hazards and act to minimize the risks. Wright State University's Laboratory Safety Manual provides direction and some of the supportive materials you will need to reduce the risk from laboratory hazards and to comply with the many regulations and recommendations that guide laboratory activities.

Over the years, OSHA promulgated rules and published guidance to make laboratories increasingly safe for personnel. Several primary OSHA standards apply to laboratories and these are discussed within. Other OSHA standards also apply to various aspects of laboratories and these are referred to in this document.

The Occupational Exposure to Hazardous Chemical in Laboratories Standard (29 CFR 1450) was created specifically for non-production laboratories. Additional OSHA standards provide rules that protect workers, including those that work in laboratories, from chemical hazards as well as biological, physical and safety hazards. For those hazards that are not covered by a specific OSHA standard, OSHA often provides guidance on protecting workers from these hazards. This manual is designed to provide awareness of the OSHA requirements and provide guidance to protect workers from the diverse hazards encountered in laboratories.

The regulatory requirements and administrative intent to inform and to train employees may explain the need for a safety manual, but why should you, a laboratory worker, read it? The answer is that it is you who may be at risk. Although responsibility for your safety may rest with your supervisor, laboratory or department head, and ultimately the university president, YOU are in the best position to protect yourself. The policies and practices defined in this manual set a baseline for the conduct of laboratory operations. Not following these recommendations puts not only you at greater risk, but also your co-workers and the environment.

### **What can you gain from reading this manual?**

Your awareness of the hazards in your workplace should be heightened and your abilities to recognize them improved. You may discover that common practices are not necessarily sound and safe. On the other hand, years of experience have shown that the practices described in this manual are functional and productive. Developing your base of safety knowledge helps to instill work habits that are

beneficial to science, safety, and health. This manual is only a primer and should be supplemented by formal and informal training by your supervisor, as well as the other laboratory staff. Ask your supervisor to identify and discuss the hazards in your workplace and those specific to protocols you will follow or duties you will be expected to perform. This manual should provide background for formulating the questions that you will need to pose to protect yourself in an environment that offers diverse and changing hazards.

### **What are the hazards?**

Physical hazards, many of which are not unique to the laboratory, are those usually identified with maintenance and housekeeping activities. The amounts of electricity, ranges of light, noise and temperature, esoteric equipment, and materials create special physical hazards. For example, hazards associated with power, plumbing and mechanical systems can result in cuts, bruises, sprains, skeletal injuries, burns and shocks.

Hazards are also associated with the use, storage, and disposal of biological agents, radioactive materials, toxic chemicals and carcinogens. More than one of these types of hazards are likely to be present with interdisciplinary laboratory operations. The effects of exposure to hazardous materials will vary, may be additive and/or synergistic, and may be immediate (acute) or manifest themselves later (chronic), sometimes years later.

While safety ends to focus on avoiding the immediate threats, chronic effects are more difficult to identify and for many substances the risks are unknown. Even for those that are known, it may be difficult to link present illnesses with past exposures.

Laboratory specific hazards can be classified according to exposure routes. Except for irradiation from sources of non-ionizing and ionizing radiation, such as ultraviolet or infrared lamps, lasers, nuclear magnetic resonance and irradiators, e.g., cesium and cobalt, most exposures result from the inadvertent inhalation, absorption, ingestion, or inoculation of hazardous materials. This usually implies that substances were either volatilized or aerosolized in open laboratory space, were deposited or spilled onto laboratory surfaces from which they could be transferred by fingers to mouth, or transferred by cuts or punctures. To reduce the health risk from these materials, it is essential that you identify the possible routes of exposure.

### **Who is at risk?**

Different groups of individuals are subject to different levels of risk:

- those who are in contact with hazards regularly or only occasionally;
- those who are in proximity to hazards daily or less frequently; and
- those removed from hazards but on the same floor, in the same building, or at the same site.

Individuals are assigned to risk levels based on frequency of exposure and proximity to risk. Assignment to an appropriate risk group can be used by employers and supervisors to determine the required training and protective measures. Your awareness of the hazards surrounding you can help assure that you are adequately trained and protected.

### **How can risks be reduced?**

Risk is the probability of your experiencing the consequences of a hazard. Your knowledge of a given laboratory and your ability to recognize the hazards and appropriate responses may diminish your risks. Response may entail avoidance, reduction or minimization and may call for different actions under different circumstances. For example, avoidance might entail not entering a laboratory or entering but keeping your hands in your pockets. However, avoidance might also mean using an alternative non-hazardous material or procedure. The proper use of primary barriers, such as shielding, fume hoods and biological safety cabinets, and designated storage locations can limit the number of areas with potential hazards while the use of gloves, goggles, personal hygiene, *e.g.*, washing your hands regularly, and good laboratory practices can further reduce the chances of exposure. Neither the barrier nor the protective equipment provides absolute protection, and consideration must always be given to minimizing exposures. Just because a risk is small does not imply that it is acceptable, especially if it is controllable and without accompaniment of some clear benefit.

Although exposures to certain chemicals, radioactive and biological materials are known to be potentially hazardous, the degree to which the hazards have been recognized and regulated varies. The amounts and types of radioactive materials used in laboratories are small and generally only present a health risk if they are internalized. With proper laboratory procedures, internal exposure can be easily prevented. However, radioactive substances are the most highly regulated hazardous materials.

Even though the health hazards presented by biological agents are generally well defined and very consequential, the reported incidence of disease resulting from laboratory exposure to infectious agents is low. Most of those reported are associated with hepatitis virus from the handling of blood and blood products. Pioneers in working with hazardous pathogens developed techniques that protected both themselves and their work. The introduction of the biological safety cabinet has been important to both aspects. Spearheaded by research involving recombinant DNA and oncogenic viruses, official guidelines for work with biological materials have been issued, which are thought to be effective by most professionals.

Although knowledge of the toxicity of certain chemicals has been utilized since the early Egyptians, only recently have acute, and more importantly chronic exposures to chemicals been convincingly linked to mutagens and carcinogens. Nonetheless, it has been difficult to regulate exposure to the vast array of laboratory chemicals. The identification of chemicals selected based on their

toxic and carcinogenic potential by OSHA is one of the first attempts to control the exposure of laboratory workers to chemicals.

### **How should you read this manual?**

Scan it fully to appreciate the breadth of safety information available. Although much of it may be familiar, read **GENERAL SAFETY PRACTICES** and ask yourself whether you follow these principles. Perhaps you or your co-workers consider general safety policies, such as not eating or drinking in the laboratory, unreasonable, but drinking tea or coffee in the laboratory has been the known source of ingestion of radioactive contaminant. How many other laboratory chemicals, which have been similarly ingested, go undetected, perhaps daily? Take nothing for granted. In safety, as in science, no problem is too small to be significant. Behavior modification and good work habits are the ultimate means to reduce risks.

Since recognizing hazards is not always possible, your awareness of past experiences, especially emergencies, can be invaluable. Be self-protective. Review the various sections that apply to new and specific job assignments. Do not make assumptions about the background, understanding, and training you and those around you have received. Encourage others to consult this manual and consider its recommendations. Seeking advice, discussing protocols, being aware of what others are doing, especially in unfamiliar or large, shared laboratories, all contribute to a safer and more productive work environment.

This manual is not intended as a substitute for formal education and training in the sciences, and was written with the assumption that the laboratory worker has had a formal introduction to the physical sciences. However, because most college graduates have little knowledge of ionizing radiation, additional background information on that topic appears in Wright State University's **RADIATION SAFETY MANUAL**.

Given that safety information continues to expand along with and because of the laboratory work it supports, no one manual can be completely original, encompassing, or applicable. The health and safety guidelines in this safety manual were distilled from the collective wisdom of many leading research institutions. Many of the standards herein have been set forth by various federal, state, and local agencies. Major sources include:

- National Institutes of Health Biohazards Safety Guide;
- NIH Guidelines for Recombinant DNA Research;
- National Cancer Institute Biological Safety Manual for Research Involving Oncogenic Viruses and Chemical Carcinogens;
- Centers for Disease Control Biosafety Guidelines for Microbiological and Biomedical Laboratories; Occupational Safety and Health Administration Standards; and
- National Research Council's Prudent Practices for Handling Hazardous Chemicals in the Laboratory.

As a useful guide for safety in the laboratory research, The Laboratory Safety Manual is neither the “only approach” nor a replacement for common sense. The presence or absence of information is not meant to assign importance to hazards or to ways of mitigating the, nor is it meant to reduce personal responsibility for one’s own safety.

## **4.2 Purpose**

To ensure employees and students are informed about the hazards of chemicals in their workplace and laboratories and are protected from chemical exposures exceeding the allowable levels (*i.e.*, OSHA permissible exposure limits (PELs)) as specified in Table Z of the Air Contaminants standard (29 CFR 1910.1000) and as specified in other substance-specific health standards. The Laboratory standard (20 CR 1910.1450) achieves this protection by establishing safe work practices in laboratories by implementing a Chemical Hygiene Plan.

## **4.3 Scope and Application**

The information contained in this Laboratory Safety Manual applies to all individuals engaged in laboratory use of hazardous chemicals. Work with hazardous chemicals outside of laboratories is covered by the Hazard Communication standard (20 CFR 1910.1200). Laboratory uses of chemicals which provide no potential for exposure (e.g., chemically impregnated test media or prepared kits for pregnancy testing) are not covered by the Laboratory standard.

## **4.4 Roles and Responsibilities**

Persons responsible for Chemical Hygiene and Laboratory Safety include, but are not limited for the following:

### **4.4.1 Deans, Department Chairs, and Program Managers**

- Bears ultimate responsibility for chemical hygiene and lab safety within the their laboratories and/or facilities.
- Provides continuing support for institutional chemical hygiene and lab safety.

### **4.4.2 Chemical Hygiene Officer**

- Develops and implements appropriate chemical policies and practices.
- Monitors procurement, uses, and disposal of chemicals use in the laboratory.
- Ensures that appropriate audits are maintained.
- Helps project directors and principle investigators develop precautions and adequate facilities.
- Knows the current legal requirements concerning regulated substances.
- Seeks ways to improve the chemical hygiene program.

#### **4.4.3 Laboratory Supervisors**

- Have overall responsibility for chemical hygiene in the laboratory.
- Ensure that laboratory workers know and follow the chemical hygiene rules.
- Ensure that protective equipment is available and in working order.
- Ensure that appropriate training has been provided.
- Provide regular, formal chemical hygiene and housekeeping inspections, including routine inspections of emergency equipment.
- Know the current legal requirements concerning regulated substances.
- Determine the required levels of personal protective equipment (PPE) and equipment.
- Ensure that facilities and training for use of any material being ordered are adequate.

#### **4.4.4 Laboratory Workers**

- Read, understand, and follow all safety rules and regulations that apply to the work area.
- Plan and conduct each operation in accord with the facility's chemical hygiene procedures, including use of PPE and engineering controls, as appropriate.
- Develop good personal chemical hygiene habits.
- Report all accidents and potential chemical exposures immediately.

#### **4.4.5 Physical Plant**

- Prepares a maintenance schedule for all engineering controls required by the Laboratory Safety Manual
- Provides regular maintenance of laboratory equipment, such as safety showers, eyewashes, filters, and fume hood, in accordance with the approved maintenance schedules.
- Promptly repairs improperly functioning equipment or notifies the proper authorities if they are unable to perform the repairs.
- Notifies affected laboratory personnel prior to removal or shutdown of utilities or laboratory safety equipment.
- When necessary, works with the Deans, Department Chairs, and Program Managers to develop cost-effective techniques and systems to manage chemical hygiene challenges.

## **4.5 Regulatory Requirements**

### **4.5.1 Section 5(a)(1) of the Occupational Safety and Health Act of 1970 (OSH Act)**

#### **4.5.1.1 The General Duty Clause**

Section 5(a)(1) of the Occupational Safety and Health Act of 1970 (OSH Act), the General Duty Clause, requires that employers “shall furnish to each of his employees’ employment and a place of employment which are free from recognized hazards that are causing or likely to cause death or serious physical harm to his employees.” Therefore, even if an OSHA standard has not been promulgated that deals with a specific hazard or hazardous operation, protection of workers from all hazards or hazardous operations may be enforceable under section 5(a)(1) of the OSH Act. For example, best practices that are issued by non-regulatory organizations such as the National Institute for Occupational Safety and Health (NIOSH), the Centers for Disease Control and Prevention (CDC), the National Research Council (NRC), and the National Institutes of Health (NIH), can be enforce- able under section 5(a)(1).

#### **4.5.2 The Occupational Exposure to Hazardous Chemicals in Laboratories standard (29 CFR 1910.1450)**

The Occupational Exposure to Hazardous Chemicals in Laboratories standard (29 CFR 1910.1450), commonly referred to as the Laboratory standard, requires that the employer designate a Chemical Hygiene Officer and have a written Chemical Hygiene Plan (CHP), and actively verify that it remains effective. The CHP must include provisions for worker training, chemical exposure monitoring where appropriate, medical consultation when exposure occurs, criteria for the use of personal protective equipment (PPE) and engineering controls, special precautions for particularly hazardous substances, and a requirement for a Chemical Hygiene Officer responsible for implementation of the CHP. The CHP must be tailored to reflect the specific chemical hazards present in the laboratory where it is to be used. Laboratory personnel must receive training regarding the Laboratory standard, the CHP, and other laboratory safety practices, including exposure detection, physical and health hazards associated with chemicals, and protective measure.

#### **4.5.3 Hazard Communication standard (29 CFR 1910.1200)**

The Hazard Communication standard (29 CFR 1910.1200), sometimes called the HazCom standard, is a set of requirements first issued in 1983 by OSHA. The standard requires evaluating the potential hazards of chemicals, and communicating information concerning those hazards and appropriate protective measures to employees. The standard includes provisions for: developing and maintaining a written hazard communication program for the workplace, including lists of hazardous chemicals present; labeling of containers of chemicals in the workplace, as well as of containers of chemicals being shipped to other workplaces; preparation and distribution of material safety data sheets (SDSs) to

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workers and downstream employers; and development and implementation of worker training programs regarding hazards of chemicals and protective measures. This OSHA standard requires manufacturers and importers of hazardous chemicals to provide material safety data sheets to users of the chemicals describing potential hazards and other information. They must also attach hazard warning labels to containers of the chemicals. Employers must make MSDSs available to workers. They must also train their workers in the hazards caused by the chemicals workers are exposed to and the appropriate protective measures that must be used when handling the chemicals.

#### **4.5.4 Air Contaminants standard (29 CFR 1910.1000)**

The Air Contaminants standard provides rules for protecting workers from airborne exposure to over 400 chemicals. Several of these chemicals are commonly used in laboratories and include: toluene, xylene, and acrylamide. Toluene and xylene are solvents used to fix tissue specimens and rinse stains. They are primarily found in histology, hematology, microbiology and cytology laboratories.

#### **4.5.5 Bloodborne Pathogens standard (29 CFR 1910.1030)**

The Bloodborne Pathogens standard (29 CFR 1910.1030), including changes mandated by the Needlestick Safety and Prevention Act of 2001, requires employers to protect workers from infection with human bloodborne pathogens in the workplace. The standard covers all workers with “reasonably anticipated” exposure to blood or other potentially infectious materials (OPIM). It requires that information and training be provided before the worker begins work that may involve occupational exposure to bloodborne pathogens, annually thereafter, and before a worker is offered hepatitis B vaccination. The Bloodborne Pathogens standard also requires advance information and training for all workers in research laboratories who handle human immunodeficiency virus (HIV) or hepatitis B virus (HBV). The standard was issued as a performance standard, which means that the employer must develop a written exposure control plan (ECP) to provide a safe and healthy work environment, but is allowed some flexibility in accomplishing this goal. Among other things, the ECP requires employers to make an exposure determination, establish procedures for evaluating incidents, and determine a schedule for implementing the standard’s requirements, including engineering and work practice controls. The standard also requires employers to provide and pay for appropriate PPE for workers with occupational exposures. Although this standard only applies to bloodborne pathogens, the protective measures in this standard (e.g., ECP, engineering and work practice controls, administrative controls, PPE, housekeeping, training, post-exposure medical follow-up) are the same measures for effectively controlling exposure to other biological agents.



#### **4.5.6 Personal Protective Equipment (PPE) standard (29 CFR 1910.132)**

The Personal Protective Equipment (PPE) standard (29 CFR 1910.132) requires that employers provide and pay for PPE and ensure that it is used wherever “hazards of processes or environment, chemical hazards, radiological hazards, or mechanical irritants are encountered in a manner capable of causing injury or impairment in the function of any part of the body through absorption, inhalation or physical contact.” [29 CFR 1910.132(a) and 1910.132(h)]. To determine whether and what PPE is needed, the employer must “assess the workplace to determine if hazards are present, or are likely to be present, which necessitate the use of [PPE],” 29 CFR 1910.132(d)(1). Based on that assessment, the employer must select appropriate PPE (e.g., protection for eyes, face, head, extremities; protective clothing; respiratory protection; shields and barriers) that will protect the affected worker from the hazard, 29 CFR 1910.132 (d)(1)(i), communicate selection decisions to each affected worker, 29 CFR 1910.132 (d)(1)(ii), and select PPE that properly fits each affected employee, 29 CFR 1910.132(d)(1)(iii). Employers must provide training for workers who are required to use PPE that addresses when and what PPE is necessary, how to wear and care for PPE properly, and the limitations of PPE, 29 CFR 1910.132(f).

#### **4.5.7 Eye and Face Protection standard (29 CFR 1910.133)**

The Eye and Face Protection standard (29 CFR 1910.133) requires employers to ensure that each affected worker uses appropriate eye or face protection when exposed to eye or face hazards from flying particles, molten metal, liquid chemicals, acids or caustic liquids, chemical gases or vapors, or potentially injurious light radiation, 29 CFR 1910.133(a).

#### **4.5.8 Respiratory Protection standard (29 CFR 1910.134)**

The Respiratory Protection standard (29 CFR 1910.134) requires that a respirator be provided to each worker when such equipment is necessary to protect the health of such individual. The employer must provide respirators that are appropriate and suitable for the purpose intended, as described in 29 CFR 1910.134(d)(1). The employer is responsible for establishing and maintaining a respiratory protection program, as required by 29 CFR 1910.134(c), that includes, but is not limited to, the following:

- selection of respirators for use in the workplace;
- medical evaluations of workers required to use respirators;
- fit testing for tight-fitting respirators;
- proper use of respirators during routine and emergency situations;
- procedures and schedules for cleaning,
- disinfecting, storing, inspecting, repairing and discarding of respirators;
- procedures to ensure adequate air quality, quantity, and flow of breathing air for atmosphere-supplying respirators;

- training of workers in respiratory hazards that they may be exposed to during routine and emergency situations;
- training of workers in the proper donning and doffing of respirators, and any limitations on their use and maintenance; and
- regular evaluation of the effectiveness of the program.

#### **4.5.9 Hand Protection standard (29 CFR 1910.138)**

The Hand Protection standard (29 CFR 1910.138), requires employers to select and ensure that workers use appropriate hand protection when their hands are exposed to hazards such as those from skin absorption of harmful substances; severe cuts or lacerations; severe abrasions; punctures; chemical burns; thermal burns; and harmful temperature extremes, 29 CFR 1910.138(a). Further, employers must base the selection of the appropriate hand protection on an evaluation of the performance characteristics of the hand protection relative to the task(s) to be performed, conditions present, duration of use, and the hazards and potential hazards identified, 29 CFR 1910.138(b).

#### **4.5.10 Control of Hazardous Energy standard (29 CFR 1910.147)**

The Control of Hazardous Energy standard (29 CFR 1910.147), often called the “Lockout/Tagout” standard, establishes basic requirements for locking and/or tagging out equipment while installation, maintenance, testing, repair, or construction operations are in progress. The primary purpose of the standard is to protect workers from the unexpected energization or start-up of machines or equipment, or release of stored energy. The procedures apply to the shutdown of all potential energy sources associated with machines or equipment, including pressures, flows of fluids and gases, electrical power, and radiation.

#### **4.5.11 Medical and First Aid standard (29 CFR 1910.151)**

Medical and First Aid standard (29 CFR 1910.151) requires employers to provide medical and first-aid personnel and supplies commensurate with the hazards of the workplace. The details of a workplace medical and first-aid program are dependent on the circumstances of each workplace and employer.

#### **4.5.12 Recordkeeping standard (29 CFR 1904)**

Recordkeeping standard (29 CFR 1904) requires Wright State to keep records of workplace injuries and illnesses.

#### **4.5.13 Access to Worker Exposure and Medical Records standard (29 CFR 1910.1020)**

Access to Worker Exposure and Medical Records standard (29 CFR 1910.1020) requires all employers, regardless of size or industry, to report the work-related death of any worker or hospitalizations of three or more workers. It also requires employers to provide workers, their designated representatives, and OSHA with access to worker exposure and medical records. Employers generally should maintain worker exposure records for 30 years and medical records for the

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duration of the worker's employment plus 30 years, unless one of the exemptions listed in 29 CFR 1910.1020(d)(1)(i) (A)-(C) applies.

All employers, including Wright State, covered by OSHA recordkeeping requirements must post the OSHA Poster in a prominent location in the workplace.

#### **4.5.14 Other OSHA standards**

In addition to the standards listed above, other OSHA standards that pertain to electrical safety (29 CFR 1910 Subpart S-Electrical); fire safety (Portable Fire Extinguishers standard, 29 CFR 1910.157); and slips, trips and falls (29 CFR 1910 Subpart D – Walking-Working Surfaces, Subpart E - Means of Egress, and Subpart J - General Environmental Controls), which pertain to general industry, as well as laboratories. When laboratory workers are using large analyzers and other equipment, their potential exposure to electrical hazards associated with this equipment must be assessed by employers and appropriate precautions taken. Similarly, worker exposure to wet floors or spills and clutter can lead to slips/trips/falls and other possible injuries and employers must assure that these hazards are minimized. While large laboratory fires are rare, there is the potential for small bench-top fires, especially in laboratories using flammable solvents. It is the responsibility of employers to implement appropriate protective measures to assure the safety of workers

## **5 Chemical and Compressed Gas Safety**

### **5.1 Risk and Exposure to Chemicals**

As stated in the introduction, hazards in the laboratory exist and chemical hazards contribute significantly.

Reactive chemicals traditionally have been the focus of life safety in the laboratory. However, their toxicity is of recent and growing concern. The toxic properties of certain chemicals have been known for thousands of years, the significance of risks associated with toxic chemicals in the laboratory on the health of laboratory workers is continuing to expand. While exposures to highly toxic or acutely toxic substances are, given their short-term effects, easy to identify, the long-term effects of exposure to certain chemicals are much more difficult to predict.

The list of compounds for which there is sufficient evidence of carcinogenicity is growing. Many of these chemicals are commonly found in laboratories. The OSHA Laboratory Standard<sup>1</sup> cites five studies on the long-term effects of exposure to toxic substances in the laboratory. While the results are not conclusive, they suggest an increased incidence of pancreatic (and possibly brain tumors) and lymphohaematopoietic malignancies among laboratory chemists. Although it is simple to say that at some level all chemicals are toxic and direct contact should be avoided, special attention must be given to limiting

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exposure to those that are acutely toxic, present reproductive hazards, and to the known and suspected carcinogens.

Unlike the regulation of radioactive materials or infectious agents for which precise standard laboratory guidelines exist, the regulation of chemicals has been made the responsibility of those in the laboratory. The OSHA Laboratory Standard requires laboratories develop a Chemical Hygiene Plan that is available to all laboratory workers. The plan includes standard operating protocols (SOPs) for the use, storage, and disposal of particularly hazardous substances using the best knowledge and techniques available. The SOPs must include the use of engineering controls and personal protective equipment within the boundaries of “designated areas,” which in many instances may mean the entire laboratory. This places much of the burden on laboratory supervisors and all laboratory workers, who must learn to familiarize themselves with the physical and health hazards associated with chemicals in their laboratory and to implement standard operating protocols which will minimize their exposure to them. A basic understanding of exposure, dose and toxicity is essential to this process.

It should be noted that the nature of laboratory work may also necessitate addressing biological safety, radiation safety, and security issues.

#### **5.1.1 Employee Exposure Determination**

OSHA has established permissible exposure limits (PELs), as specified in 29 CFR 1910, subpart Z, for hundreds of chemical substances. A PEL is the chemical-specific concentration in inhaled air that is intended to represent what the average, healthy worker may be exposed to daily for a lifetime of work without significant adverse health effects. The employer must ensure that workers’ exposures to OSHA-regulated substances do not exceed the PEL. However, most of the OSHA PELs were adopted soon after the Agency was first created in 1970 and were based upon scientific studies available at that time. Since science has continued to move forward, in some cases, there may be health data that suggests a hazard to workers below the levels permitted by the OSHA PELs. Other agencies and organizations have developed and updated recommended occupational exposure limits (OELs) for chemicals regulated by OSHA, as well as other chemicals not currently regulated by OSHA. Employers should consult other OELs, in addition to the OSHA PEL, to make a fully informed decision about the potential health risks to workers associated with chemical exposures. The American Conference of Governmental Industrial Hygienists (ACGIH), the American Industrial Hygiene Association (AIHA), the National Institute for Occupational Safety and Health (NIOSH), as well as some chemical manufacturers have established OELs to assess safe exposure limits for various chemicals.

Employers must conduct exposure monitoring  
Employers must conduct exposure monitoring, through air sampling, if there is reason to believe that workers may be exposed to chemicals above the action level or, in the absence of an action level, the PEL. Periodic exposure monitoring should be conducted in accord with

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the provisions of the relevant standard. The employer should notify workers of the results of any monitoring within 15 working days of receiving the results. Some OSHA chemical standards have specific provisions regarding exposure monitoring and worker notification. The relevant standards to must be reviewed to comply with the applicable provisions apply to their laboratory.

#### **5.1.1.1 Exposure**

The nature and quality of chemical, as well as the mode and duration of the exposure, determine the risk inherent in contacting the chemical. Threshold Limit Values (TLV) issued by the American Conference of Governmental Industrial Hygienists (ACGIH) may be used as guidelines for assessing the severity of an exposure. Note that through the adoption of the TLVs by OSHA as Permissible Exposure Levels (PELs), these PELs now carry the weight of law for determining safe exposure as well as levels at which actions must be taken to reduce exposure.

Time Weighted Averages (TWA) refer to the average airborne concentration of substances to which it is believed nearly all workers may be repeatedly exposed during a normal 8-hour workday and 40-hour week, day after day without adverse effect. Because of wide variation in susceptibility, individuals may experience discomfort from some substance at concentrations at or below the threshold limit; a smaller percentage may be affected more seriously by aggravation of a pre-existing condition or by development of an occupational illness.

Short Term Exposure Limit (STEL) is a 15-minute time-weighted average exposure which should not be exceeded at any time even if the eight-hour time-weighted average is within the PEL. If a STEL is not specified, short term exposures should not exceed three times the TWA for no more than a total of 30 minutes per day. Exposure above the TLV up to the STEL should not be longer than 15 minutes and should not occur more than four times a day. There should be at least 60 minutes between successive exposures in this range. These levels are not necessarily conservative when applied to the research setting, where exposures to and synergistic effects from chemicals must also be considered. Likewise, individual experiences and sensitivities should be evaluated. For example, pregnant women and particularly their fetuses may be susceptible to levels lower than anticipated for most adults.

A Ceiling Limit (CL) is the concentration that should not be exceeded during any part of the working day.

The term IDTL means “Immediately Dangerous to Life.” This term refers to concentrations of materials that can cause death in a very short time, usually by causing loss of consciousness followed by death. Obviously, these levels are not encountered during normal operations.

Although the repeated use of some hazardous chemicals may justify the use of specific monitors, if available, for the most part it is your vigilance upon which you

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must rely. This includes the appearance of vapors, moist surfaces, mixing patterns, color changes, skin, eye, or respiratory reactions, and odors. Do not ignore any of these signs and take steps to minimize your contact.

Some chemicals have characteristic odors. A list of odor thresholds has been compiled by the American Industrial Hygiene Association (AIHA). While you should not use your nose to estimate chemical concentration because of the potential for overexposure, it can be of great practical value in identifying the source of an odor and alerting you to possible hazardous levels. Individual olfactory responses, fatigue, and acclimation are important factors. Remember that not all hazardous chemicals have odors and for some the level for olfactory detection may be too high to be of protective value. See the comparison of odor thresholds and TLVs for some hazardous chemicals the following table. (Table 3: Exposure Limits and Odor Threshold Values)

**Table 3: Exposure Limits and Odor Threshold Values**

CHEMICAL	PEL (ppm)	TLV (ppm)	STEL (ppm)	Odor Threshold (ppm)	IDLH (ppm)
ACETIC ACID	10	10	15	2	50
ACETONE	1000	500	750	2-100	2,500
ACETONITRILE	40	2	-	20	500
BENZENE	1	0.5	2.5	30	500
N BUTYL ALCOHOL	100	50	50	0.1-3	1,400
CARBON TETRACHLORIDE	10	5	10	75	200
CHLOROFORM	50c	10c	-	100	500
DIETHYL AMINE	10	5	15	0.02-25	200
FORMALDEHYDE	0.75	-	0.3	1	20
HEXANE	500	50	-	-	1,100
METHYL ALCOHOL	200	200	250	10	6,000
PHOSGENE	0.1	0.1	-	0.5	2
PYRIDINE	5	5	-	0.01	1,000
TOLUENE	200	20	-	0.2	500
TRIETHYLAMINE	25	5	15	1	200
XYLENE	100	100	150	0.5	900

PEL: Permissible Exposure Limit, OSHA Standard 29 CFR 1910.1000, Tables Z-1, Z-2, or Z-3

TWA-TLV: Time Weighted Average-Threshold Limit Values, 1999-2000 ACGIH

TWA-STEL: Time Weighted Average-Short Term Exposure Limit, 1999-2000 ACGIH

IDLH: Immediately Dangerous to Life or Health, NIOSH-MSHA

PPM: Parts of Chemical Per Million Parts of Air

C: Maximum Allowable Exposure Dose

### 5.1.2 Dose

All chemicals may be toxic at some level. However, the dose absorbed is the critical factor impacting the health of the individual. Individuals may be tolerant or susceptible to chemical exposures and the precise dose at which toxic effects will be manifested varies over a range. Dosages of some chemicals with known toxic

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properties may elicit no response or may even have a beneficial effect. For example, reproductive hormones are essential to our health, yet at high concentrations, e.g., those initially used in oral contraceptives, may be carcinogenic. As most toxicological data is based upon experimental work with other species, it is helpful to be able to compare dosage by weight and surface area to evaluate the data. For example, mutagenicity screening for a chemical with Salmonella (Ames test), the dosage of a chemical required to produce death in 50% of the treated animals (LD<sub>50</sub>) is usually determined and used.

The dosages required to produce harmful health effects vary 10 billion-fold between different chemicals (Table 4: Comparison of Dosage by Weight and Surface Area (100 m/kg) Dose). The acutely toxic chemicals, e.g., mold toxins (of which aflatoxin is the most familiar) are at the low end of this range, where single doses of less than 10 mg/kg body weight can be lethal.

**Table 4: Comparison of Dosage by Weight and Surface Area (100 m/kg) Dose**

Species	Weight (g)	Surface (cm <sup>3</sup> )	Dose Weight (mg)	Dose Surface (mg)	Dose Ratio
Mouse	20	46	2	2	1
Rat	200	325	20	14	1.43
Guinea pig	400	565	40	24	1.67
Rabbit	1500	1270	150	55	2.73
Cat	2000	1380	200	60	3.33
Monkey	4000	2980	400	128	3.12
Dog	12000	5770	1200	248	4.84
Man	70000	18000	7000	776	9.02

From Casarett and Doull's Toxicology, Doull, Klassen, Amdur (eds.), pg.22, 1986.

Some chemicals include the LD<sub>50</sub>s with the information that manufacturers are required to provide to purchasers. The EPA and OSHA's Appendices A and B to the Hazard Communication Standard (29 CFR 1910.1200) consider the following "acutely toxic":

- An oral LD<sub>50</sub> (rat) of less than 50 mg/kg
- An inhalation LC<sub>50</sub> (rat) of less than 2 mg/L
- Dermal LD<sub>50</sub> (rabbit) of less than 200 mg/kg

A chemical which "is otherwise capable of causing or significantly contributing to an increase in serious irreversible, or incapacitating reversible, illness." The accompanying table (Table 5: Acute toxicity hazard categories and acute toxicity estimate (ATE) values defining the respective categories) of relative hazard levels based on animal data provides a fuller perspective of the dose range.

**Table 5: Acute toxicity hazard categories and acute toxicity estimate (ATE) values defining the respective categories**

Exposure route	Category 1	Category 2	Category 3	Category 4
Oral (mg/kg body weight) <sup>a,b</sup>	≤ 5	>5 and ≤ 50	>50 and ≤ 300	>300 and ≤ 2000
Dermal (mg/dg body weight) <sup>a,b</sup>	≤ 50	>50 and ≤ 200	>200 and ≤ 1000	>1000 and ≤ 2000
Inhalation – Gases (ppmV) <sup>a,b,c</sup>	≤ 100	>100 and ≤ 500	>500 and ≤ 2500	>2500 and ≤ 20000
Inhalation – Vapors (mg/l) <sup>a,b,c,d</sup>	≤ 0.5	>0.5 and ≤ 2.0	>2.0 and ≤ 10.0	>10.0 and ≤ 20.0
Inhalation – Dusts and Mists (mg/L) <sup>a,b,c</sup>	≤ 0.05	>0.05 and ≤ 0.5	>0.5 and ≤ 1.0	>1 and ≤ 5.0

Source: 29 CFR 1910.1200 App A

Note: Gas concentrations are expressed in parts per million per volume (ppmV).

Notes:

- a) The acute toxicity estimate (ATE) for the classification of a substance is derived using the LD<sub>50</sub>/LC<sub>50</sub> where available;
- b) The acute toxicity estimate (ATE) for the classification of a substance or ingredient in a mixture is derived using:
  - (i) the LD<sub>50</sub>/LC<sub>50</sub> where available. Otherwise,
  - (ii) the appropriate conversion value that relates to the results of a range test, or
  - (iii) the appropriate conversion value from that relates to a classification category;
- c) Inhalation cut-off values in the table are based on 4-hour testing exposures. Conversion of existing inhalation toxicity data which has been generated according to 1 hour exposure is achieved by dividing by a factor of 2 for gases and vapors and 4 for dusts and mists;
- d) For some substances, the test atmosphere will be a vapor which consists of a combination of liquid and gaseous phases. For other substances, the test atmosphere may consist of a vapor which is nearly all the gaseous phase. In these latter cases, classification is based on ppmV as follows: Category 1 (100 ppmV), Category 2 (500 ppmV), Category 3 (2500 ppmV), Category 4 (20000 ppmV). The terms "dust", "mist" and "vapor" are defined as follows:
  - (i) Dust: solid particles of a substance or mixture suspended in a gas (usually air);
  - (ii) Mist: liquid droplets of a substance or mixture suspended in a gas (usually air);
  - (iii) Vapor: the gaseous form of a substance or mixture released from its liquid or solid state.

Understanding the concepts of toxicity, exposure, and dose can help effectively minimize the risk associated with working with hazardous chemicals. Check the chemicals in your laboratory against the PELs and the RELs. If you think that an exposure exceeding these values is likely, check with your laboratory supervisor for steps to minimize exposure, e.g., work in a fume hood and wear gloves. Arrange with Environmental Health and Safety to review the data and monitor your exposure, if necessary. For quick identification of chemicals that may require special handling, in addition to the chemicals for which PELs and RELs exist, the EPA's Acutely Hazardous and Extremely Hazardous Substances are

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included in the appendix along with a list of substances regulated by OSHA as carcinogens. The EPA's Extremely Hazardous list is used with Title III of SARA (community right-to-know) and was developed using the above-mentioned criteria for acutely toxic chemicals and their dispersal potential. A short list of selected chemicals with known reproductive hazards follows, but a more complete list is given by Shepherd T.H. 1983, (Catalog of Teratogenic Agents, 4<sup>th</sup> ed):

#### 5.1.2.1 Examples of Common Chemicals with Known Reproductive Hazards

Acrylonitrile	Formamides
Aniline	Lead (Organic)
Arsenic and its Compounds	Manganese and its Compounds
Benzene Benzo(a)pyrene	Mercury and its Compounds
Beryllium	(Inorganic) Methyl n-Butyl Ketone
Boric Acid (Boron)	Methyl Chloroform
Cadmium and its Compounds	Methyl Ethyl Ketone (MEK) Nitrogen
Carbon Monoxide	Dioxide
Carbon Tetrachloride	Ozone
Chlorodecone (Kepone)	Platinum and its compounds
Chloroform	Polybrominated Biphenyls (PBB)
Chloroprene	Polychlorinated Biphenyls (PCB)
Dibromochloropropane (DBCP)	Selenium and its Compounds
Dichlorobenzene	Styrene
2-4-Diisocyanate	Tellurium and its Compounds
1,1-Dichloroethane	Tetrachloroethylene
Dichloromethane	Thallium and its Compounds
Dioxane	Toluene
Epichlorohydrin	0-Toluidine
Ethylene Dibromide (Dibromoethane)	Trichloroethylene
Ethylene Dichloride	Vinyl Chloride
Ethylene Oxide	Vinylidene Chloride
Fluorocarbons	Xylene
Formaldehyde	

Additional information about the hazardous characteristics of a chemical is available with rapid access to several computerized databases at the National Library of Medicine is available through the Medical Literature Analysis & Retrieval System (MEDLARS). These include:

- CCRIS, Chemical Carcinogenesis Research Information System
- DART, Developmental & Reproductive Toxicology
- EMIC, Environmental Mutagen Information Center
- HSDB, Hazardous Substances Data Bank
- IRIS, Integrated Risk Information System
- RTECS, Registry of Toxic Effects of Chemical Substances

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- TOXLINE & TOXLIT, Toxicology Information
- Online and Toxicology Literature from Special Sources
- TRI, Toxic Release Inventory

### 5.1.3 Chemical Labels and Safety Data Sheets (SDS)

Read the labels on reagent bottles so that you know beforehand what hazards are involved. If sufficient information is not given, as part of compliance with Hazard Communication and Right-to-Understand laws, Environmental Health and Safety can provide information about the various Safety Data Sheets (SDS) for most common chemicals that are on hand.

All containers of chemicals must be labeled clearly. Do not use any substance in an unlabeled or improperly labeled container. Chemicals with printed labels which have been partly obliterated, scratched over, or crudely labeled by hand should not be trusted and, together with unlabeled containers, should be disposed of promptly to avoid adverse reactions. If a material must be transferred to another container, careful attention must be paid to relabeling. The new label must contain all cautions from the original label. Do not use initials or abbreviated names. Carefully remove the label before removing a reagent from its container. Read it again as you promptly recap the container and return it to its proper location. Names of distinctly different substances are sometimes nearly alike and using the wrong substances can lead to accidents.

## 5.2 Chemical Safety

The risk of laboratory injuries can be reduced through adequate training, improved engineering, good housekeeping, safe work practice and personal behavior.

All the precautions listed in the section on **GENERAL SAFETY PRACTICES** should be followed. To avoid direct contact with chemicals, attention must be given to use of fume hoods and selection of personal protective equipment appropriate for the chemicals handled. Be sure to select gloves that are not readily degraded and/or permeated by the specific chemicals used. A table in the appendix to the **GENERAL SAFETY PRACTICES** provides information on the resistance of different glove material to some common chemicals.

### 5.2.1 General Rules for Laboratory Work with Chemicals

- Assigned work schedules should be followed unless a deviation is authorized by the laboratory supervisor.
- Unauthorized experiments should not be performed.
- Plan safety procedures before beginning any operation.
- Follow standard operating procedures at all times.
- Always read the SDS and label before using a chemical.
- Wear appropriate PPE at all times.

- To protect your skin from splashes, spill and drips, always wear long pants and closed-toe shoes.
- Use appropriate ventilation when working with hazardous chemicals.
- Never pipet by mouth.
- Always wash hands with soap and water immediately after working with any laboratory chemicals, even if gloves have been worn.
- Eating, drinking, smoking, gum-chewing, applying cosmetics, and taking medicine in laboratories where hazardous chemicals are used or stored is strictly prohibited.
- Food, beverages, cups, and other drinking and eating utensils should not be stored in areas where hazardous chemicals are handled or stored.
- Laboratory refrigerators, ice chests, cold room, and ovens must not be used for food storage or preparation.
- Contact the laboratory supervisor, Principal Investigator, CHO or EHS office with all safety questions or concerns.
- Know the location and proper use of safety equipment.
- Maintain situational awareness.
- Make others aware of special hazards associated with your work.
- Notify supervisors or chemical sensitivities or allergies.
- Report all injuries, accidents, incidents, and near-misses.
- Unauthorized persons should not be allowed in the laboratory.
- Report unsafe conditions to the laboratory supervisor or C
- Properly dispose of chemical wastes.

#### **5.2.1.1 Working Alone**

Working alone in a laboratory is dangerous and should be strictly avoided. Many tragic accidents illustrate this danger. Accidents are unexpected by definition, which is why coworkers should always be present. Workers should coordinate schedules to avoid working alone.

#### **5.2.1.2 Housekeeping**

Housekeeping can help reduce or eliminate many laboratory hazards. Proper housekeeping includes appropriate labeling and storage of chemicals, safe and regular cleaning of the facility, and proper arrangement of laboratory equipment.

#### **5.2.1.3 Standard Operating Procedures (SOP)**

Standard Operating Procedures (SOPs) outline prudent laboratory practices which must be followed when working with chemicals in a laboratory. These include general and laboratory-specific procedures for work with hazardous chemicals.

## **5.2.2 Nanoparticles and Nanomaterials**

Nanoparticles and nanomaterials have different reactivities and interactions with biological systems than bulk materials, and understanding and exploiting these differences is an active area of research. However, these differences also mean that the risks and hazards associated with exposure to engineered nanomaterials are not well known. Because this is an area of ongoing research, consult trusted sources for the most up to date information available. Note that the higher reactivity of many nanoscale materials suggests that they should be treated as potential sources of ignition, accelerants, and fuel that could result in fire or explosion. Easily dispersed dry nanomaterials may pose the greatest health hazard because of the risk of inhalation. Operations involving these nanomaterials deserve more attention and more stringent controls than those where the nanomaterials are embedded in solid or suspended in liquid matrixes.

Consideration should be given to all possible routes of exposure to nanomaterials including inhalation, ingestion, injection, and dermal contact (including eye and mucous membranes). Avoid handling nanomaterials in the open air in a free particle state. Whenever possible, handle and store dispersible nanomaterials, whether suspended in liquids or in a dry particle form, in closed (tightly-sealed) containers. Unless cutting or grinding occurs, nanomaterials that are not in a free form (encapsulated in a solid or a nanocomposite) typically will not require engineering controls. If a synthesis is being performed to create nanomaterials, it is not enough to only consider the final material in the risk assessment, but consider the hazardous properties of the precursor materials as well.

To minimize laboratory personnel exposure, conduct any work that could generate engineered nanoparticles in an enclosure that operates at a negative pressure differential compared to the laboratory personnel breathing zone. Limited data exist regarding the efficacy of PPE and ventilation systems against exposure to nanoparticles. However, until further information is available, it is prudent to follow standard chemical hygiene practices. Conduct a hazard evaluation to determine PPE appropriate for the level of hazard according to the requirements set forth in OSHA's Personal Protective Equipment standard (29 CFR 1910.132).

## **5.2.3 Highly Toxic and Explosive/Reactive Chemicals/Mixtures**

### **5.2.3.1 Prevention**

The use of highly toxic and explosive/ reactive chemicals and materials has been an area of growing concern. The frequency of academic laboratory incidents in the U.S. is an area of significant concern for the Chemical Safety Board (CSB). The CSB issued a case study on an explosion at Texas Tech University in Lubbock, Texas, which severely injured a graduate student handling a high-energy metal compound. Since 2001, the CSB has gathered preliminary

information on 120 different university laboratory incidents that resulted in 87 evacuations, 96 injuries, and three deaths.

Each facility or laboratory must keep a detailed inventory of highly toxic chemicals and explosive/reactive materials. The date of receipt, amount, location, and responsible individual for all acquisitions, syntheses, and disposal of these chemicals should be documented. A physical inventory should be performed annually to verify active inventory records. Be sure to report security breaches, inventory discrepancies, losses, diversions, or suspected thefts to your laboratory supervisor, EHS, and possibly campus security.

Procedures for disposal of highly toxic materials must be established before any experiments begin, possibly even before the chemicals are ordered. The procedures must address methods for decontamination of any laboratory equipment that comes into contact with highly toxic chemicals. All waste must be accumulated in clearly labeled impervious containers that are stored in unbreakable secondary containment.

Highly reactive and explosive materials that may be used in the laboratory require appropriate procedures and training. An explosion can occur when a material undergoes a rapid reaction that results in a violent release of energy. Such reactions can happen spontaneously and can produce pressures, gases, and fumes that are hazardous. Some reagents pose a risk on contact with the atmosphere. It is prudent laboratory practice to use a safer alternative whenever possible.

If possible, substitutes for highly acute, chronic, explosive, or reactive chemicals should be considered prior to beginning work and used whenever possible.

#### **5.2.3.2 Reactive Chemicals**

Reactive chemicals are substances which, under certain ambient or induced conditions, enter into violent reactions. Some examples: nitroglycerin, nitrocellulose, and organic peroxides. Many substances, when mixed, are potentially explosive (such as hydrazines and nitric acid).

##### **5.2.3.2.1 Peroxide Formers**

Note that the following compounds readily form peroxides upon:

###### **5.2.3.2.1.1 Storage (3 Months)**

- Isopropyl ether
- Divinyl acetylene
- Vinylidene chloride
- Potassium metal
- Sodium amide

#### 5.2.3.2.1.2 Concentration (12 Months)

Ethyl ether<sup>2</sup>  
Tetrahydrofuran  
Dioxane  
Acetal  
Methyl i-butyl ketone  
Ethylene glycol  
Methyl ether (glyme)  
Vinyl ethers  
Dicyclopentadiene  
Diacetylene

#### 5.2.3.2.1.3 Initiation of Polymerization (12 Months)

Styrene  
Butadiene  
Tetrafluoroethylene  
Cumene  
Vinyl acetylene  
di-Vinyl acetate  
Vinyl chloride  
Chlorotrifluoroethylene  
Chlorobutadiene  
(Chloroprene)  
Methyl acetylene  
Vinyl pyridine  
Tetrahydronaphthalene  
Cyclohexene  
Methylcyclopentane

#### 5.2.3.2.2 Oxidizing and Reducing Substances

In many oxidizing and reducing reactions, both agents must be present. In some cases, one or the other substance may create a hazard by coming into contact with a normally innocuous substance. These reactions tend to generate heat and are often explosive, *e.g.*, glycerol and potassium permanganate blended at room temperature for a few minutes react violently producing fire. The following examples of typical oxidizers may:

##### 5.2.3.2.2.1 Increase Rate of Combustion:

Aluminum nitrate

Ammonium persulfate

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<sup>2</sup> Storage in metal containers slows peroxide formation.

Barium chlorate  
Barium peroxide  
Calcium chlorate  
Calcium nitrate  
Calcium peroxide  
Cupric nitrate  
Hydrogen peroxide  
Lead nitrate  
Lithium hypochlorite  
Lithium peroxide  
Magnesium nitrate  
Magnesium perchlorate  
Magnesium peroxide  
Nickel nitrate  
Nitric acid 70% or less  
Perchloric acid 60% or less  
Potassium chlorate

Potassium dichromate  
Potassium nitrate  
Potassium persulfate  
Potassium persulfate  
Silver nitrate  
Silver nitrite  
Sodium perborate  
Sodium perborate  
Sodium perchlorate  
Sodium persulfate  
Strontium chlorate  
Strontium nitrate  
Strontium nitrite  
Thorium nitrite  
Uranium nitrate  
Zinc chlorate  
Zinc peroxide

**5.2.3.2.2 Cause Spontaneous Ignition:**

Calcium hypochlorite  
Chromic acid  
Hydrogen peroxide (27.5-52%)  
Nitric acid  
Potassium bromate  
Potassium permanganate  
Sodium chlorite (more than 40%)  
Sodium peroxide  
Sodium permanganate  
Trichloroisocyanuric acid  
Sodium dichloroisocyanurate

**5.2.3.2.3 Decompose with Catalyst or Heat:**

Ammonium dichromate  
Hydrogen peroxide (52-91%)  
Calcium hypochlorite (over 50%)  
Perchloric acid (60-72.5%)  
Potassium dichloroisocyanurate  
Sodium dichloroisocyanurate

**5.2.3.2.4 Cause Explosive Reaction When Exposed to Catalyst, Heat, Shock or Friction:**

Ammonium perchlorate  
Ammonium permanganate  
Perchloric acid

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## Potassium superoxide

### 5.2.3.2.3 Water Sensitive Substances

Water sensitive chemicals react with water, steam, and moisture in the air to evolve heat and/or flammable or explosive gases. Isolate water-sensitive substances from other reactive compounds, and store in a cool, waterproof area. Some substances that liberate only heat are: strong acids and bases, acid anhydrides and sulfides. Substances that liberate flammable gases when exposed to water include alkali metals, hydrides, nitrites, carbides, and anhydrous metallic salts.

### 5.2.3.2.4 Air Reactive Substances

Air reactive materials are capable of rapid release of energy by themselves, as by self-reaction or polymerization, for example with phosphorus. This category includes substances that can be easily ignited by common sources of heat when mixed with air. Examples of these substances include the following: alkali metals, ammonium nitrate, ammonium perchlorate, ammonium permanganate, benzoyl peroxide, boron hydrides, dinitrobenzene, lithium hydride, and sulfur.

### 5.2.3.2.5 Acid Reactive Substances

Acid reactive chemicals combine with acid to evolve heat, flammable and/or explosive gases, and toxicants. Examples include alkali metals, hydroxides, carbides, nitrites, arsenic and related elements, cyanides, sulfides, and structural alloys (most metals).

### 5.2.3.2.6 Special Organic Compounds

Special organic compounds are unstable and may decompose spontaneously or through contact with the immediate environment (*e.g.*, air, water, and other reactants). Examples include diazonium compounds, diazomethane, chlorination intermediates, butadiene, nitration intermediates, organic sulfates, polymerization reactions, and highly nitrated compounds.

### 5.2.3.2.7 Pyrophoric Agents

Pyrophoric agents burn when exposed to air. In general, they require absolute protection against air. Examples include phosphorous and activated zinc.

## 5.2.4 Specific Chemical Hazards

The Air Contaminants standard (29 CFR 1910.1000) standard provides rules for protecting workers from airborne exposure to over 400 chemicals. Several of these chemicals are commonly used in laboratories and include toluene, xylene, and acrylamide. Toluene and xylene are solvents used to fix tissue specimens and rinse stains. They are primarily found in histology, hematology, microbiology and cytology laboratories.



#### 5.2.4.1 Toluene

Toluene		
Exposure Routes	Symptoms	Target Organs
Inhalation; Ingestion Skin and/or eye contact; Skin absorption	Irritation of eyes, skin, nose, throat; Weakness exhaustion, confusion, euphoria, headache; Dilated pupils, tearing; Anxiety; Muscle fatigue; Insomnia; Tingling, pricking, or numbness of skin; Dermatitis; Liver, kidney damage	Eyes; Skin; Respiratory System; Central nervous system; Liver; Kidneys.

#### 5.2.4.2 Xylene

Xylenes		
Exposure Routes	Symptoms	Target Organs
Inhalation; Ingestion Skin and/or eye contact; Skin absorption	Irritation of eyes, skin, nose, throat; Dizziness, excitement, drowsiness, incoherence, staggering gait; Corneal vacuolization (cell debris); Anorexia, nausea, vomiting, abdominal pain; Dermatitis	Eyes; Skin; Respiratory System; Central nervous system; GI tract; Blood; Liver; Kidneys.

#### 5.2.4.3 Acrylamide

Acrylamide is usually found in research laboratories and is used to make polyacrylamide gels for separations of macromolecules (e.g., DNA, proteins).

Acrylamide		
Exposure Routes	Symptoms	Target Organs
Inhalation; Ingestion Skin and/or eye contact; Skin absorption	Irritation of eyes, skin; Ataxia (staggering gait), numb limbs, tingling, pricking, or numbness of skin; Muscle weakness; Absence of deep tendon reflex; Hand sweating; Tearing, Drowsiness; Reproductive effects; Potential occupational	Eyes; Skin; Central nervous system; Peripheral nervous system; Reproductive system (in animals: tumors of the lungs, testes, thyroid and adrenal glands).

carcinogen.

To prevent worker exposure:

- Review the Hazard Communication Policy for chemicals This policy contains provisions for worker training, warning labels and access to Safety Data Sheets (SDSs).

#### 5.2.4.4 Formaldehyde standard (29 CFR 1910.1048)

Formaldehyde is used as a fixative and is commonly found in most laboratories. No worker must be exposed to an airborne concentration of formaldehyde which exceeds 0.75 parts formaldehyde per million parts of air (0.75 ppm) as an 8-hour time weighted average (TWA), 29 CFR 1910.1048(c)(1).

The Hazard Communication standard requires the maintenance of an SDS, which manufacturers or distributors of formaldehyde are required to provide. The SDS must be kept in an area that is accessible to workers that may be exposed to formaldehyde.

Formaldehyde		
Exposure Routes	Symptoms	Target Organs
Inhalation; Ingestion Skin and/or eye contact	Irritation of eyes, skin, nose, throat, respiratory system; Tearing; Coughing; Wheezing; Dermatitis; Potential occupational nasal carcinogen	Eyes; Skin; Respiratory System

Employers must provide the following to workers to prevent exposure:

- Appropriate PPE, as required per 29 CFR 1910.132, 29 CFR 1910.133, and 29 CFR 1910.1048(h).
- Acceptable eyewash facilities within the immediate work area for emergency use, if there is any possibility that a worker's eyes may be splashed with solutions containing 0.1 percent or greater formaldehyde, 29 CFR 1910.1048(i)(3).

#### 5.2.4.5 Latex

One of the most common chemicals that laboratory workers are exposed to is latex, a plant protein. The most common cause of latex allergy is direct contact with latex, a natural plant derivative used in making certain disposable gloves and other products. Some healthcare workers have been determined to be latex sensitive, with reactions ranging from localized dermatitis (skin irritation) to immediate, possibly life-threatening reactions. Under OSHA's Personal Protective Equipment standard, 29 CFR 1910.132, the employer must ensure that appropriate personal protective equipment (PPE) is accessible at the worksite or issued to workers. Latex-free gloves, glove liners, powder-free

gloves, or other similar alternatives are obtainable and must be readily accessible to those workers who are allergic to latex gloves or other latex-containing PPE, 29 CFR 1910.1030(c)(3)(iii). Latex allergy should be suspected in workers who develop certain symptoms after latex exposure, including:

- nasal, eye, or sinus irritation
- hives or rash
- difficulty breathing
- coughing
- wheezing
- nausea
- vomiting
- diarrhea

An exposed worker who exhibits these symptoms should be evaluated by a physician or other licensed healthcare professional because further exposure could cause a serious allergic reaction. Once a worker becomes allergic to latex, special precautions are needed to prevent exposures. Certain medications may reduce the allergic symptoms, but complete latex avoidance is the most effective approach.

Appropriate work practices should be used to reduce the chance of reactions to latex. If a worker must wear latex gloves, oil-based hand creams or lotions (which can cause glove deterioration) should not be used unless they have been shown to reduce latex-related problems and maintain glove barrier protection. After removing latex gloves, workers should wash their hands with a mild soap and dry them thoroughly.

Powdered latex gloves are prohibited from use.

#### **5.2.5 Compressed Gases**

Compressed gases expose laboratory personnel to both chemical and physical hazards. It is essential that these are monitored for leaks and have the proper labeling. By monitoring compressed gas inventories and disposing of or returning gases for which there is no immediate need, the laboratory can substantially reduce these risks. Leaking gas cylinders can cause serious hazards that may require an immediate evacuation of the area and activation of the emergency response system. Only appropriately trained hazmat responders may respond to stop a leaking gas cylinder under this situation.

When ordering hazardous gases, consider factors such as handling and storage, compatibility of gas regulators, eye and skin absorption, and chemical properties. Remember that some gases are corrosive (e.g., ammonia, chlorine, hydrogen chloride, hydrogen fluoride), flammable (e.g., acetylene, butane, hydrogen, methane, propane), oxidizers (e.g., oxygen, chlorine), toxic (e.g., carbon monoxide, ethylene oxide) or cryogenic (e.g., nitrogen, carbon dioxide, oxygen).

### 5.2.5.1 General Precautions

Cylinders of compressed gases should be handled as high energy sources and therefore as potential explosives. The following rules apply:

- When storing or moving a cylinder, have the cap securely in place to protect the valve stem.
- When moving large cylinders, they must be strapped to a properly designed wheeled cart to ensure stability.
- Cylinders of all sizes must be restrained by straps, chains, or a suitable stand to prevent them from falling.
- Cylinders of toxic, flammable, or reactive gases should be used in fume hoods and, when possible, stored in fume hoods.
- Do not expose cylinders to temperatures higher than 50° C. Some rupture devices on cylinders will release at about 65° C. Some small cylinders including lecture bottles are not fitted with rupture devices and may explode if exposed to high temperatures.
- Never use a cylinder that cannot be positively identified. Do not rely on the color of the cylinder to identify its contents.
- Use the appropriate regulator on each gas cylinder. Adapters or homemade modifications can be dangerous.
- Use only correct, pressure-rated tubing.
- Never lubricate, modify, force or tamper with a cylinder valve. Do not loosen or remove the safety plug or rupture disc.
- Leaks can be monitored by pressurizing the system, turning off the cylinder stem valve and looking for a drop in the discharge pressure. The location of leaks can be identified by painting all fittings and joints with soapy water and watching for bubble formation. When using toxic gases, it is advisable to use a toxic gas detector or indicator for detection and warning. Wrapping the thread with Teflon tape may be necessary to stop the leaks.
- When corrosive gases are being used, the cylinder stem valve should be worked frequently to prevent its freezing.
- Keep cylinders containing liquefied gases upright. Note that is often difficult to determine the contents of a cylinder containing liquefied gas, except by weighing. As long as a liquid is present, the cylinder or vapor pressure will remain constant. The cylinder pressure for liquefied carbon dioxide does provide an indication of cylinder content.
- Do not put oil or grease on the high-pressure side of an oxygen cylinder. Oil or grease on the high-pressure side of an oxygen cylinder can lead to an explosion.
- Do not allow a rapid release of a compressed gas. Rapid release of a compressed gas will cause an unsecured gas hose to whip dangerously and may build up a static charge which could ignite combustible gas.
- Do not extinguish a flame involving a highly combustible gas until the source of gas has been shut off as it can re-ignite causing an explosion.

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- Never bleed a cylinder completely empty. Leave a slight pressure to keep contaminants out. In the case of nitrogen cylinders, leave approximately 10 psi. This prevents contamination of the cylinder.
- When not in use, cylinder and bench valves should be closed tightly.
- Remove the regulators from the empty cylinders and replace the protective caps. Mark the cylinder “Empty” or “MT” and return to the distributor.
- Do not keep cylinders filled with corrosive, explosive, or highly toxic gases more than 6 months; do not keep cylinders with oxygen or liquids or flammable gases more than 3 years.
- If a cylinder begins to leak, if possible, move it outdoors. Contact Environmental Health and Safety/Wright State Police Department as soon as feasible.
- Damaged or corroded cylinders and cylinders with a test date more than 5 years old stamped on the shoulder should be returned to the vendor.
- Do not order a surplus of cylinders. Besides presenting a safety hazard, there usually is a daily rental fee.

#### **5.2.5.2 Cylinder Features**

The valve outlet connection connects to pressure and/or flow-regulating equipment. Specific connections are provided to prevent interchange of equipment for incompatible gases. A CGA number identifies them; for example, CGA 350 is used for hydrogen, carbon monoxide, methane, and some other flammable gases. For information on valve and regulator fittings, consult the manufacturer.

A pressure relief device prevents a fully charged cylinder from bursting in case of exposure to high heat.

The cylinder collar holds the cylinder cap that protects the cylinder valve from mechanical or weather damage. It should be removed from the cylinder only when the cylinder is supported and ready to be attached to pressure-reducing and/or flow control equipment for use.

The DOT number signifies that the cylinder conforms to DOT specifications and that the service pressure for which the cylinder is designed is 2265 psi and 21°C with an exception indicated by the + sign following the last test date, which allows a 10% overfilling.

The cylinder serial number is registered with the DOT, and can be used to verify the contents of the cylinder by querying the manufacturer.

The cylinder test date indicates the month and year of initial hydrostatic test. Thereafter, hydrostatic tests are performed on a cylinder at intervals specified by the DOT (usually every 5 years), or when the supplier feels they are necessary, to determine whether the cylinder is fit for further use. For each hydrostatic test, the new test date is stamped into the cylinder shoulder.

The encircled insignia is that of the original inspector.

Cylinder size is important to consider when purchasing compressed gas, especially flammable gases. The National Fire Protection Association has recommended that all laboratories using flammable gas contain no more than three tanks (maximum size equals approximately 10" x 50") in a nonsprinkled area as long as the presence of other combustible items in the room is minimal.

#### 5.2.5.3 Precautions for Cryogenic Gases

- Avoid contact; both the liquid and the gases can cause frostbite. Do not touch uninsulated piping.
- Wear loose-fitting thermal gloves, goggles and/or face shield, closed shoes.
- Work in a well-ventilated area. Liquefied gas can rapidly expand, e.g., nitrogen expands almost 700-fold.
- Never attempt to prevent vapors from escaping from cylinders of liquefied, cryogenic gases. Since they are not at thermal equilibrium, vapor is produced as the liquid boils and, if not vented to the atmosphere, could produce excessive pressures.
- Use only the special (usually metal) tubing designed for use with these gases. Do not improvise with plastic or rubber tubing.
- Be aware that oxygen enrichment and a fire hazard can result from the condensation of oxygen (boiling point  $-183^{\circ}\text{C}$ ) from the air onto piping cooled by liquid nitrogen (boiling point  $-196^{\circ}\text{C}$ ).
- If skin contacts liquefied cryogenic gases, thaw burned area slowly in cold water. Do not rub.

#### 5.2.5.4 Regulators

The proper choice of a regulator depends on the delivery-pressure range required, the degree of accuracy of delivery pressure to be maintained, and the flow rate required. There are two basic types of pressure regulators, single-stage and two-stage. The single-stage type will show a slight variation in delivery pressure as the cylinder pressure drops. It will also show a greater drop in delivery pressure than a two-stage regulator as the flow rate is increased. In addition, it will show a higher "lock-up" pressure (pressure above the delivery set-point necessary to stop flow) than the two-stage regulator. In general, the two-stage regulator will deliver a more constant pressure under more stringent operating conditions than will the single-stage regulator.

A regulator should be attached to a cylinder without forcing the threads. If the inlet of a regulator does not fit the cylinder outlet, no effort should be made to try to force the fitting. A poor fit may indicate that the regulator is not intended for use on the gas chosen. The following steps should be taken for delivery of gas:

1. After the regulator has been attached to the cylinder valve outlet, turn the delivery pressure-adjusting screw counterclockwise until it turns freely.

- a) Open the cylinder valve slowly until the tank gauge on the regulator registers the cylinder pressure. At this point, the cylinder pressure should be checked to see if it is at the expected value. A large error may indicate that the cylinder valve is leaking.
- b) With the flow-control valve at the regulator outlet closed, turn the delivery pressure-adjusting screw clockwise until the required delivery pressure is reached. Control of flow can be regulated by means of a valve supplied in the regulator outlet or by a supplementary valve installed in a pipeline downstream from the regulator. The regulator itself should not be used as a flow control by adjusting the pressure and in some cases where higher flows are obtained in this manner, the pressure setting may be more than the design pressure of the system.

### 5.2.5.5 Information on Some Common Gases

Become familiar with the threshold limit values lists threshold limit values (TLV), flammability limits (LEL and UEL), and major hazards associated with commonly used gases (Table 6: Data for Common Gases).

**Table 6: Data for Common Gases**

Threshold Limit Vales				
Gas	TWA, ppm <sup>a</sup>	STEL, ppm <sup>a</sup>	Flammability Limits <sup>b</sup> Air, % (Volume)	Major Hazard
Acetylene	None	None	2.5-81.0	Flammable, asphyxiant
Ammonia	25	25	15.0-28	Toxic
Argon	None	None	None	Asphyxiant, cryogenic
Boron trifluoride	1 (Ceiling)	None	None	Toxic, causes burns
1,3-Butadiene	10	None	2.0-11.5	Flammable, skin irritant, carcinogen
Butane	800	None	1.8-8.4	Flammable, anesthetic
Carbon dioxide	5000	None	None	Asphyxiant, cryogenic
Carbon monoxide	50	400	12.5-74.2	Flammable, toxic
Chlorine	0.5	1	None	Toxic, corrosive,

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				irritant, burns
Ethane	None	None	3.0-12.5	Flammable, asphyxiant
Ethylene	None	None	3.1-32.0	Flammable, asphyxiant
Ethylene oxide	1	None	3.0-80.0	Flammable, burns if trapped by clothing, carcinogen
Helium	None	None	None	Asphyxiant, cryogenic
Hydrogen	None	None	4.0-75.0	Flammable, asphyxiant, cryogenic
Hydrogen bromide	3 (Ceiling)	None	None	Toxic, burns, corrosive
Hydrogen chloride	5 (Ceiling)	None	None	Toxic, burns, corrosive
Hydrogen fluoride	3 (Ceiling)	None	None	Toxic, severe slow healing burns, corrosive
Hydrogen sulfide	10	15	4.3-46.0	Toxic, flammable, irritant
Methane	None	None	5.0-15.4	Flammable, asphyxiant, cryogenic
Methyl bromide	5	None	10.0-16.0	Toxic, burns, skin permeable
Methyl chloride	50	100	10.7-17.4	Toxic, flammable
Methyl mercaptan	0.5	None	Unknown	Toxic, flammable
Nitrogen	None	None	None	Asphyxiant, cryogenic
Nitrogen dioxide	3	5	None	Toxic Corrosive
Oxygen	None	None	None	Highly reactive, cryogenic
Phosgene	0.1	None	None	Highly toxic
Propane	None	None	2.2-9.5	Flammable, asphyxiant
Sulfur dioxide	2	5	None	Toxic, causes burns
Vinyl chloride	5	None	4.0-22.0	Flammable, carcinogen, burns

<sup>a</sup> American Conference of Governmental Industrial Hygienists, 1989.

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<sup>b</sup> Matheson Gas Data Bank, 6th Edition, 1980.

#### **5.2.5.5.1 Acetylene**

Acetylene is the most thermodynamically unstable common gas, has a very wide explosive range (from 2% to 80% in air), and under pressure and certain conditions can decompose with explosive force.

To allow safe handling of acetylene in cylinders, suppliers use a porous packing material saturated with a solvent in which the acetylene dissolves. The combination of porous filling and solvent markedly enhances the stability of acetylene.

Acetylene is authorized for shipment only as a dissolved gas in cylinders marked DOT-8 or -8AL, and cylinders so designated may be used only for acetylene.

Never use or store in a prone position.

#### **5.2.5.5.2 Argon, Carbon Dioxide, Helium and Nitrogen**

These gases are inert, colorless, odorless, and tasteless but can cause asphyxiation and death in confined, poorly ventilated areas.

Do not lean into or place your head into a freezer.

In addition, these gases can cause severe frostbite to the eyes or skin.

Some carbon dioxide cylinders contain an educator tube and are intended for liquid withdrawal. These cylinders are specially marked; be sure you are using equipment appropriate to the application.

Air will condense on exposed helium liquid or cold-gas surfaces, such as vaporizers and piping. Nitrogen, having a lower boiling point than oxygen, will evaporate first, leaving an oxygen-enriched condensation on the surface.

To prevent possible ignition of grease, oil, or other combustible materials, care must be taken that equipment is free of these materials.

#### **5.2.5.5.3 Hydrogen**

Hydrogen is a flammable gas. A mixture of hydrogen and oxygen or air in a confined area will explode if ignited by a spark, flame or another similar source. Escaping hydrogen cannot be detected by a sight, smell or taste and, because of its lightness, it tends to accumulate in the upper portions of confined areas.

#### **5.2.5.5.4 Oxygen**

Oxygen supports and can greatly accelerate combustion; keep combustibles away from oxygen and eliminate ignition sources. Oxygen is colorless, odorless, and tasteless and as a liquid or cold gas may cause severe frostbite to the eyes or skin.

Many materials, especially some non-metallic gaskets and seals, constitute a combustion hazard when in oxygen service, although they may be acceptable for use with other gases. Before attaching regulator to cylinder, be certain that the regulator and inlet filter are free of oil grease, or other contaminants, and crack the cylinder valve momentarily to blow out any dust or dirt that might have accumulate in the cylinder outlet.

When using an oxygen torch remember to turn on the natural gas (in sufficient quantity) first and off last and wear UV absorbing eye protection

#### 5.2.5.6 Gas Cylinder Disposal

When compressed gas tanks are empty, label the cylinders with the letters “MT”.

When compressed gas tanks are empty or no longer needed, contact those responsible for chemical waste disposal to return the cylinders to the distributor. If the distributor is no longer available, call Environmental Health and Safety.

**NEVER** dispose of gas cylinders, even small propane cylinders, lecture bottles, or chemical aerosol cans, in the general trash.

#### 5.2.6 Chemical Procurement, Distribution, and Storage

Prudent chemical management includes the following processes:

- Chemical Procurement
- Chemical Storage
- Chemical Handling
- Chemical Inventory
- Transporting Chemicals
- Transferring Chemical

### 5.3 Safety Recommendations – Physical Hazards

Physical hazards in the laboratory include combustible liquids, compressed gases, reactives, explosives and flammable chemicals, as well as high pressure/energy procedures, sharp objects and moving equipment. Injuries can result from bodily contact with rotating or moving objects, including mechanical equipment, parts, and devices.

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*Personnel should not wear loose-fitting clothing, jewelry, or unrestrained long hair around machinery with moving parts.*

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The Chemical Safety Board has identified the following key lessons for laboratories that address both physical and other hazards:

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- Ensure that research-specific hazards are evaluated and then controlled by developing specific written protocols and training.
- Expand existing laboratory safety plans to ensure that all safety hazards, including physical hazards of chemicals, are addressed.
- Ensure that the organization's EHS office reports directly to an identified individual/office with organizational authority to implement safety improvements.
- Develop a verification program that ensures that the safety provisions of the CHP are communicated, followed, and enforced at all levels within the organization.
- Document and communicate all laboratory near-misses and previous incidents to track safety, provide opportunities for education and improvement to drive safety changes at the university.
- Manage the hazards unique to laboratory chemical research in the academic environment. Utilize available practice guidance that identifies and describes methodologies to assess and control hazards.
- Written safety protocols and training are necessary to manage laboratory risk.

#### 5.3.1.1 Procurement of Chemicals

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*“The decision to procure a specific quantity of a specific chemical is a commitment to handle it responsibly from receipt to ultimate disposal. Each operation in which it is handled and each period between operations presents opportunities for misadventure.” – National Research Council. 1980. Prudent Practices for Handling Hazardous Chemicals in Laboratories. Washington, DC. National Academy Press. pp. 10*

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- Information on proper handling, storage, and disposal should be known to those before a substance is received.
- Only containers with adequate identifying labels should be accepted.
- Ideally, a central location should be used for receiving all chemical shipments.
- Shipments with breakage or leakage should be refused or opened in a chemical hood.
- Only the minimum amount of the chemical needed to perform the planned work should be ordered.
- Purchases of high-risk chemicals should be reviewed and approved by the CHO.

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- Proper protective equipment and handling and storage procedures should be in place before receiving a shipment.

### 5.3.2 Chemical Stocks and Storage

Although storing chemicals in alphabetical order may seem convenient, it increases the chances that incompatible materials will mix in the event of leaks, spills, breakage, floods or fires. Physical hazards can be reduced by purchasing the minimal amounts of chemicals required and requesting that they be supplied in shatter-proof containers. Storing heavier items on lower shelves, but not on the floor, will further reduce these hazards. While separating chemicals into mutually exclusive compatible groups for separate storage is ideal, it is difficult to reach a consensus of what those groups should be. Moreover, for these groups to be truly exclusive requires many sub-divisions with appropriate separate and well-maintained storage locations.

Unlike dedicated chemical storage rooms within which partitioned areas or separate storage cabinets or drums can be allocated to specific groups of chemicals, laboratory space must also accommodate personnel, fixtures and equipment. Inevitably this means there will be only a few possible distinct locations for storing chemicals. For convenience, these locations are typically under sinks for corrosives, under fume hoods for flammable and volatile chemicals, and on shelves near a set of balances for general chemicals. An explosion-proof refrigerator may be needed to store flammable chemicals that tend to decompose at room temperature. The explosion-proof refrigerator seals all possible sources of ignition inside and outside the refrigerator. Thus, it can be used for storage of flammable liquids when there is a possibility of accumulation of flammable vapors outside the refrigerator. Due to the reactivity of oxidizers, it is important to segregate them from other chemicals at all storage locations.

Ordering the minimal amounts of the chemicals required is especially important for highly hazardous materials, e.g., explosives, carcinogens, acutely toxic chemicals. These materials should not be purchased in excess so that storing them will not be necessary. For this reason, no storage category for explosives is listed below. Given that fewer separate storage locations will be available than ideal, secondary containers should be used to separate incompatible chemicals within storage groups. For example, chemically resistant plastic trays of adequate size should be used to both separate and contain corrosive liquids such as acids and bases. Also, containers may be useful for keeping track of small amounts of extremely toxic and controlled substances.

The use of a basic color code affixed upon receipt will greatly aid in identifying the correct chemical group and facilitate proper storage and inspection, especially by laboratory staff without backgrounds in chemistry. Chemicals, particularly those known to decompose with time, should also be marked with the date of receipt. In addition to checking the physical condition of primary and/or secondary containers, chemicals should be inspected regularly for signs of

decomposition, such as discoloration, turbidity, caking, moisture in dry chemicals, particulates in liquids, and the buildup of pressure in the vessel. Any of these conditions is adequate cause for disposing of the material as soon as possible.

The following storage scheme was developed by Stanford University and was referenced by Prudent Practices in the Laboratory (2011) is a practical starting approach for a working laboratory and should be further tuned to specific requirements.

Stanford University Compatible Storage Group Classification System  
Should be used in conjunction with specific storage conditions taken from the manufacturer's label and MSDS.

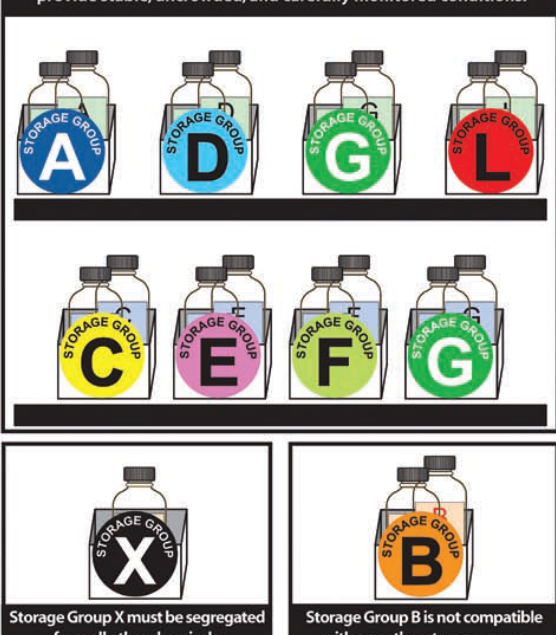
## STORAGE GROUPS

Store chemicals in separate secondary containment and cabinets

- A** Compatible Organic Bases
- B** Compatible Pyrophoric & Water Reactive Materials
- C** Compatible Inorganic Bases
- D** Compatible Organic Acids
- E** Compatible Oxidizers including Peroxides
- F** Compatible Inorganic Acids not including Oxidizers or Combustible
- G** Not Intrinsicly Reactive or Flammable or Combustible
- J\*** Poison Compressed Gases
- K\*** Compatible Explosive or other highly Unstable Material
- L** Non-Reactive Flammable and Combustible, including solvents
- X\*** Incompatible with ALL other storage groups

\*Storage Groups J, K and X: Consult EHS Department For specific storage - consult manufacturer's MSDS

If space does not allow Storage Groups to be kept in separate cabinets the following scheme can be used with extra care taken to provide stable, uncrowded, and carefully monitored conditions.



Storage Group X must be segregated from all other chemicals.

Storage Group B is not compatible with any other storage group.

Last updated: 04/17/09

**Figure 1: Compatible Storage Group Classification System (Prudent Practices, 2011)**

**Table 7: Examples of Compatible Storage Groups**

Group	Examples
<b>A: Compatible Organic Bases</b>	Dithylamine Piperidine Triethanolamine Benzylamine Benzyltrimethylammonium hydroxide
<b>B: Compatible Pyrophoric &amp; Water Reactive Materials</b>	Sodium borohydride Benzoyl chloride Zinc Dust Alkyl lithium solutions such as methyl lithium in tetrahydrofuran

<b>C: Compatible Inorganic Bases</b>	Methanesulfonyl chloride Lithium aluminum hydride Sodium hydroxide Ammonium hydroxide Lithium hydroxide Cesium hydroxide
<b>D: Compatible Organic Acids</b>	Acetic acid Citric acid Maleic acid Propionic acid Benzoic acid
<b>E: Compatible Oxidizers Including Peroxides</b>	Nitric Acid Perchloric acid Sodium hypochlorite Hydrogen peroxide 3-Chloroperoxybenzoic acid
<b>F: Compatible Inorganic Acids and not Including Oxidizers or Combustibles</b>	Hydrochloric acid Sulfuric Acid Phosphoric acid Hydrogen fluoride solution
<b>J: Poison Compressed Gases</b>	Sulfur dioxide Hexafluoropropylene
<b>K: Compatible Explosives or Other Highly Unstable Materials</b>	Picric acid dry (<10% H <sub>2</sub> O) Nitroguanidine Tetrazole Urea nitrate
<b>L: Nonreactive Flammables and Combustibles Including Solvents</b>	Benzene Methanol Toluene Tetrahydrofuran
<b>X: Incompatible with ALL Other Storage Groups</b>	Picric acid moist (10-40% H <sub>2</sub> O) Phosphorus Benzyl azide Sodium hydrogen sulfide

Additional general storage safety precautions include (Prudent Practices, 2011):

- Avoid storing materials and equipment on top of cabinets. With all stored items, maintain a clearance of at least 18 inches from the sprinkler heads to allow proper functioning of the sprinkler system
- To make chemicals readily accessible and to reduce accidents caused by overreaching, do not store materials on shelves higher than 5 ft. If retrieving materials stored above head level, use a step stool.
- Store heavy materials on lower shelves.
- Keep exits, passageways, areas under tables, benches, and emergency equipment areas free of stored equipment and materials to allow for ease of egress and access in case of emergency.

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Chemical specific storage guidelines include the following (Prudent Practices, 2011):

- Label all chemical containers to ensure that chemical will be stored safely.
- Place the user's name and the date received on all purchased materials to facilitate inventory control.
- To assist in maintaining a clean work environment and to ensure that segregation of incompatible chemicals is maintained, provide a definite storage place for each chemical and return the chemical to that location after each use.
- To avoid clutter, avoid storing chemicals on benchtops, except for those chemical being used currently.
- To avoid clutter and to maintain adequate airflow, avoid storing chemical in chemical hoods, except for those chemical in current use.
- Store volatile toxic or odoriferous chemicals in a ventilated cabinet.
- Provide ventilated storage near laboratory chemical hoods.
- Separate and store chemicals according to hazard category and compatibility.
- SDS and label information should be followed for storage requirements.
- Maintain existing labels on incoming containers of chemicals and other materials.
- Labels on containers used for storing hazardous chemicals must include the chemical identification and appropriate hazard warnings.
- The contents of all other chemical containers and transfer vessels, including, but not limited to, beakers, flasks, reaction vessels, and process equipment, should be properly identified.
- Chemical shipments should be dated upon receipt and stock rotated.
- Peroxide former should be dated upon receipt, again dated upon opening, and stored away from heat and light with tightfitting, nonmetal lids.
- Open shelved used for chemical storage should be secured to the wall and contain  $\frac{3}{4}$ -inch lips. Secondary containment devices should be used as necessary.
- Consult the SDS and keep incompatibles separate during transport, storage, use, and disposal.
- Oxidizers, reducing agents, and fuels should be stored separately to prevent contact in the event of an accident.
- Chemicals should not be stored in the chemical hood, on the floor, in areas of egress, on the benchtop, or in areas near heat or in direct sunlight.
- Laboratory-grade, flammable-rated refrigerators and freezers should be used to store sealed chemical containers of flammable liquids that require cool storage. Do not store food or beverages in the laboratory refrigerator.
- Highly hazardous chemicals should be stored in a well-ventilated and secure area designated for that purpose.

- Flammable chemicals should be stored in a spark-free environment and in approved flammable-liquid containers and storage cabinets. Grounding and bonding should be used to prevent static charge buildup when dispersing solvents.
- Chemical storage and handling rooms should be controlled-access areas. They should have proper ventilation, appropriate signage, diked floors, and fire suppression systems.

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*When transporting chemicals from one area to another, place the chemical bottle into a plastic bucket as a secondary container in case of breakage.*

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#### **5.3.2.1.1 Storage Limits**

It is recommended that laboratories have no more than 5 gallons of flammable liquid (15 for organic chemistry laboratories), 1 pound flammable solids, 5 pounds oxidizable materials, 0.59 cubic feet water volume flammable gas, 1 pound unstable (reactive) materials, and no explosives except under special circumstances and then only with the explicit approval of Environmental Health and Safety.

#### **5.3.2.2 Materials in the Laboratory**

When acquiring toxic or hazardous chemicals, obtain the smallest quantity sufficient for your work since their storage may constitute a hazard and disposal costs negate most volume discounts.

In 1990 disposal costs in the U.S. northeast for lab packed chemicals averaged \$10.00 per pound of chemical waste.

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*Purchase chemicals in shatter-proof containers when available.*

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#### **5.3.2.2.1 Flammable Liquids**

A flammable liquid is any liquid with a flashpoint below 100° F. The flash point is the lowest temperature at which a flammable liquid gives off vapor sufficient to form an ignitable mixture with air near the surface of the liquid or within the vessel used. Flammable liquids and solids must be separated from oxidizing materials. Flammable solvents requiring refrigeration should only be stored in flammable storage refrigerators. All domestic type refrigerators must have signs



warning of the danger of storing volatile or flammable chemicals, such as alcohol, acetone, and ether within them.

#### **5.3.2.2.2 Carcinogens and Highly Toxic Chemicals**

Carcinogens and highly toxic chemicals should be stored inside of marked containers in a central laboratory location.

#### **5.3.2.3 Chemical Handling**

- As previously described, a risk assessment should be conducted prior to beginning work with any hazardous chemical for the first time.
- All SDS and label information should be read before using a chemical for the first time.
- Trained laboratory workers should ensure that proper engineering controls (ventilation and PPE are in place).

#### **5.3.2.4 Chemical Inventory**

- Prudent management of chemical in any laboratory is greatly facilitated by keeping an accurate inventory of the chemicals stored.
- Unneeded items should be discarded or returned to the storeroom.

#### **5.3.2.5 Transporting Chemicals**

- Secondary containment devices should be used when transporting chemicals.
- When transporting chemicals outside of the laboratory or between stockrooms and laboratories, the transport container should be break-resistant.
- High-traffic areas should be avoided.

#### **5.3.2.6 Transferring Chemicals**

- Use adequate ventilation (such as a fume hood) when transferring even a small amount of a particularly hazardous substance (PHS).
- While drum storage is not appropriate for laboratories, chemical stockrooms may purchase drum quantities of solvents used in high volumes. Ground and bond the drum and receiving vessel when transferring flammable liquids from a drum to prevent static charge buildup.
- If chemicals from commercial sources are repackaged into transfer vessels, the new containers should be labeled with all essential information on the original container.
- Do not pipet by mouth. Use an aspirator bulb, a pipetting device or a loose-fitting hose attached to a water aspirator.
- When pouring chemicals, hold the bottle with its label toward your palm to protect the label in case some reagent drains down the outside of the bottle.

Do not pour towards yourself when adding liquids or powders. Use a funnel if the opening is small. Use a glass rod between the outside of the funnel and the neck of the receiving bottle so that air can be displaced.

- If a stopper or lid is stuck, use extreme caution in opening the bottle. Friction caused by removing tops can cause an explosion of sensitive substances.
- When a flammable liquid is withdrawn from a drum or when a drum is filled, the drum and the other equipment must be electronically grounded.
- Remove from the container only approximately what is needed, discarding any excess. Never return a chemical to its original container.
- Always add a reagent slowly; never “dump” it in. Observe what takes place when the first small amount is added and wait a few moments before adding more; some reactions take time to start. With a gloved hand, feel the outside of the receiver vessel. If it is hot, cease the additions and seek advice on whether this is part of the reaction profile. If so, the receiver vessel should be placed on ice. If an expected reaction does not initiate, seek advice before adding more reagent.
- To avoid violent reaction and splattering while diluting solutions, always pour concentrated solutions slowly into water or into less concentrated solutions while mixing, preferably on a mechanical stirrer. The more concentrated solution is usually heavier and any heat evolved is better distributed. This procedure is particularly applicable in diluting acids. Always wear goggles and gloves. Use the hood when diluting concentrated solutions.
- Beakers should be supported by holding them around the side with one hand. If the beaker is 500 mL or larger, support it from the bottom with the other hand and consider using heavy-duty beakers. When setting the beaker down, deposit it slowly on the clean surface of the bench. If the beaker is hot, use gloves and place the beaker on a protective pad. Flasks should be grasped by the neck, not by a side arm. Large flasks (3-liter) should be supported at the base when lifted. A round bottomed flask should rest on a properly sized cork ring when not assembled for reaction.
- Never look down the opening of a vessel unless it is empty.

#### 5.3.2.6.1 Shipping Chemicals

- Outgoing chemical shipments must meet all applicable Department of Transportation (DOT) regulations and should be authorized and handles

#### 5.3.3 Fume Hoods

The fume hood is the most important piece of protective equipment in a laboratory. See **GENERAL SAFETY PRACTICES** for guidelines and discussion of their uses and limitations.

### 5.3.4 Signs

Prominent signs of the following types are posted:

- Emergency telephone numbers of emergency personnel/facilities, supervisors, and laboratory workers.
- Location signs for safety showers, eyewash stations, other safety and first aid equipment, and exits
- Warnings at areas or equipment where special or unusual hazards exist.

### 5.3.5 Spills

#### 5.3.5.1 Spill Prevention

Before beginning an experiment, know your facility's policies and procedures for how to handle an accidental release of a hazardous substance, a spill or a fire. Emergency response planning and training are especially important when working with highly toxic compounds. Emergency telephone numbers should be posted in a prominent area. Know the location of all safety equipment and the nearest fire alarm and telephone.

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*Know who to notify in the event of an emergency*

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- Be prepared to provide basic emergency treatment.
- Keep your co-workers informed of your activities so they can respond appropriately.

#### 5.3.5.1.1 Safety Equipment

Safety equipment, including spill control kits, safety shields, fire safety equipment, PPE, safety showers and eyewash units, and emergency equipment should be available in well-marked highly visible locations in all chemical laboratories.

The laboratory supervisor or CHO is responsible for ensuring that all personnel are aware of the locations of fire extinguishers and are trained in their use. After an extinguisher has been used, Physical Plant must promptly recharge or replace it (29 CFR 1910.157(c)(4)). The laboratory supervisor or CHO is also responsible for ensuring proper training and providing supplementary equipment as needed.

#### 5.3.5.1.2 Chemical Solutions in Syringes

Special care must be used when handling solutions of chemicals in syringes with needles. Do not recap needles, especially when they have been in contact with

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chemicals. Remove the needle and discard it immediately after use in the appropriate sharps containers. Blunt-tip needles are available from many commercial sources and should be used unless a sharp needle is required to puncture rubber septa or for subcutaneous injection.

#### **5.3.5.1.3 Unattended Operations**

For unattended operations, laboratory lights should be left on, and signs should be posted to identify the nature of the experiment and the hazardous substances in use. Arrangements should be made, if possible, for other workers to periodically inspect the operation. Information should be clearly posted indicating who to contact in the event of an emergency. Depending on the nature of the hazard, special rules, precautions, and alert systems may be necessary.

#### **5.3.5.2 Spill Response**

Most spills in the laboratory involve comparatively small quantities of chemicals which can readily be cleaned up by laboratory personnel. It is recommended that the laboratory supervisor be notified and that spill control procedures be performed under their supervision. Arrange for disposal of chemicals and clean up materials with Environmental Health and Safety.

If the spill involves hazardous material(s) (i.e., toxic, flammable, corrosive, volatile, reactive or infectious materials) so that additional assistance or equipment is required, contact Environmental Health and Safety immediately; after hours, dial Wright State Police Department's emergency contact number, ext. 2111. Give the following information:

- Name of person calling.
- Type of spill, name of material spilled and approximate quantity.
- Location: building, floor, and room number.

Measures to be taken while waiting for assistance:

- Use absorbent pads to soak up liquid and to act as a vapor barrier.
- Clear laboratory of all personnel.
- Close all doors to corridor or adjacent rooms. Hang an appropriate warning sign on the door.

Other measures to be take while waiting for assistance:

- If a flammable liquid spills, extinguish all flames.
- If volatile chemicals are involved, open windows for ventilation (if possible) but close doors. Call Physical Plant at ext. 4444 (between 7:00 am and 3:30 pm, at all other times call Wright State Police Department at ext. 2111) to have maintenance put the building on total exhaust and total mixed air. Leave the fume hoods running.

- If an infectious or particulate agent is involved, close all windows and have maintenance turn off the air handling units in the building. Be sure to shut off all the fume hoods in the room of the spill. (Wait 30 minutes for aerosol to settle before reentering room).

If the spill occurs in public or common areas, you must notify Wright State Police Department (ext. 2111) and Environmental Health and Safety (ext. 2215) immediately.

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*In all cases immediately alert neighbors, laboratory supervisors, and/or department head.*

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#### **5.3.5.3 Personal Decontamination**

If chemicals are spilled on the body, quickly remove all contaminated clothing while using the safety shower or sink. If a large area is affected or if chemical is highly toxic and skin permeable, call Wright State Police Department's contact number, etc. 2111, to summon medical assistance. Seconds count and no time should be wasted because of modesty. Immediately flood the affected body area in cold water for at least 15 minutes. Do not use neutralizing chemicals, unguents, creams, lotions or salves. Resume rinsing if the pain returns. Report to the head of your department and the Environmental Health and Safety Department as soon as possible. Delayed reactions, often the next day, may occur and should be reported.

Alkali solutions spilled onto the skin may not be as painful as acid burn; in fact, they may not be noticeable until some time later. The reason for this is that acids precipitate a protein barrier on contact with skin, and this both prevents the acid from penetrating further and causes pain. Alkali solutions do not precipitate a protein barrier; the tissue may become thoroughly soaked and deeply damaged with relatively little discomfort, resulting in an insidious wound. For this reason, the skin that has been splashed with alkali should be continuously flushed to reach the alkali that has soaked into the tissue.

#### **5.3.5.4 Laboratory or Area Decontamination**

If chemicals are spilled on the floor or work area, seek the advice of your supervisor and the Environmental Health and Safety Department.

When cleaning up spills, work from the perimeter of the liquid spill inward and then call Environmental Health and Safety to dispose of the materials properly. If the spill is on the floor, use absorbent pads to soak up the liquid and to act as a vapor barrier. If water or some other agent is used as a diluent, be sure it is compatible with the spilled material and other chemicals in the area. The laboratory supervisor will be responsible for designating the proper cleanup

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procedure. If a flammable or toxic chemical is spilled, call Environmental Health and Safety for assistance. Warn everyone to extinguish flames and turn off spark-producing equipment such as brush-type motors and Bunsen burners. Shut down all equipment, close the doors and windows, and vacate the room until it is decontaminated.

Spill control stations containing agents for absorbing and neutralizing spills such as acids and alkali materials are available. Buy replacement kits when necessary.

#### **5.3.5.5 Mercury**

Mercury spills, commonly from broken thermometers, result in many very small particles that are difficult to clean up. Small particles of mercury have an increased rate of vaporization, due to the higher ratio of surface area to volume, and this can cause greater contamination of the air than can safely be handled by normal ventilation. The safe exposure limit can be exceeded by a single broken thermometer if not cleaned up properly. This can be further aggravated by higher temperatures, such as a broken thermometer in an oven. As a precaution, place a container underneath all mercury sources, such as manometers and barometers, and use “unbreakable” (Teflon-coated) or non-mercury thermometers.

If a mercury spill occurs, call Environmental Health and Safety. The department uses a mercury vacuum to clean the spills. Also, all mercury waste must be handled separately from other chemical waste disposal procedures.

#### **5.3.6 Waste Management**

A waste management plan should be in place before work begins on any laboratory activity. The plan should utilize the following hierarchy of practices:

- Reduce waste sources. The best approach to minimize waste generation is by reducing the scale of operations, reducing its formation during operations, and, if possible, substituting less hazardous chemicals for an operation.
- Reuse surplus materials. Only the amount of material necessary for an experiment should be purchased, and if possible, materials should be reused.
- Recycle waste. If waste cannot be prevented or minimized, consider recycling chemicals that can be safely recovered or used as fuel.
- Dispose of waste properly. Sink disposal may not be appropriate. Proper waste disposal methods include incineration, treatment, and land disposal. The Department of Environmental Health and Safety (EHS) determines which methods are appropriate for different types of waste.

##### **5.3.6.1 Collection and Storage of Waste**

- Chemical waste should be accumulated at or near the point of generation, under the control of laboratory workers.

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- Each waste type should be stored in a compatible container pending transfer or disposal. Waste containers should be clearly labeled and kept sealed when not in use.
- Incompatible waste types should be kept separate to ensure that heat generation, gas evolution, or another reaction does not occur.
- Waste containers should be segregated by how they will be managed. Waste containers should be stored in designated location that does not interfere with normal laboratory operations. Ventilated storage and secondary containment may be appropriate for certain waste types.
- Waste containers should be clearly labeled and kept sealed when not in use. Labels should include the accumulation start date and hazard warnings as appropriate.
- Non-explosive electrical systems, grounding and bonding between floors and containers, and non-sparking conductive floors and containers should be used in the central waste accumulation area to minimize fire and explosion hazards. Fire suppression systems, specialized ventilation systems, and kikes should be installed in the central waste accumulation area. Waste management workers should be trained in proper waste handling procedures as well as contingency planning and emergency response. Trained laboratory workers most familiar with the waste should be actively involved in waste management decisions to ensure that the waste is managed safely and efficiently. Engineering controls should be implemented as necessary, and personal protective equipment should be worn by workers involved in waste management.

Additional waste management practices include:

- Non-flammable, non-corrosive, non-metallic, non-toxic, odorless, water soluble liquids should be flushed down the drain in the laboratory sink, followed by large amounts of water.
- Do not pour acids, alkalis, organic solvents, reactives, flammable liquids, toxic material, material not miscible with water, corrosives, compounds that give off strong or toxic vapors, explosive agents, or other substances that are potentially harmful to the environment down the laboratory drains, slop sinks, scullery sinks, or toilet bowls. Such material will be disposed of by Environmental Health and Safety.
- Do not dispose of volatile chemicals by allowing them to evaporate in fume hoods.
- Whenever possible, non-oxidizing acids should be neutralized and flushed down the drain in the laboratory sink followed by large amounts of water. Each laboratory generating halogenated or non-halogenated solvents must supply safety cans for accumulation of both types of solvents. Each laboratory generating a large quantity of compatible solid chemical waste shall supply a five-gallon metal can for accumulation of such waste. Other liquid waste and small quantities of solid chemical waste require

accumulation in separate bottles or cans. It is the responsibility of the laboratory to supply proper containers for accumulation of wastes. Information for purchasing safety cans or five gallon buckets can be obtained from Environmental Health and Safety.

- Never package or dispose of chemical waste unless you are sure you're following proper procedures.

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*Any uncertainties regarding chemical waste disposal shall be addressed by Environmental Health and Safety at ext. 2215.*

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### **5.3.6.2 Chemical Waste Preparation and Labeling**

#### **5.3.6.2.1 Safety Cans**

Safety cans should be used for the accumulation of waste solvents only.

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***NO CORROSIVE OR HEAVY METAL WASTE SOLUTIONS  
ARE TO BE ACCUMULATED IN THESE CANS***

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Waste solvents must be accumulated in respect to their halogen content. A separate can should be used for halogenated solvents and for non-halogenated solvents. Cans must be marked accordingly.

Each safety can is supplied with an inventory sheet or a notebook which shall be used to record all waste accumulated. At a minimum, the following must be recorded each time waste is transferred into a can:

- Chemical name of waste (not chemical or molecular formula).
- Percentage of each chemical if waste is a mixture.
- Total amount of waste transferred (in liters or milliliters).
- Date.

Safety cans are emptied weekly or upon request by the generator.

#### **5.3.6.2.2 Plastic Five Gallon Buckets**

Plastic five gallon buckets are for the accumulation of solid waste only.



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*No liquid waste is to be accumulated in these buckets*

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Each bucket is supplied with an inventory sheet or a

notebook which shall be used to record all waste accumulated. At a minimum, the following must be recorded each time waste is transferred into a can:

- Chemical name of waste including any contaminants (not chemical or molecular formulas).
- Total amount of waste (grams or kilograms).
- Date.

Buckets are picked up for disposal by Environmental Health and Safety when needed or upon request by the generator.

#### **5.3.6.2.3 Old, Outdated or Unwanted Chemicals**

Old, outdated or unwanted chemicals should remain in their original containers if the container is in good condition (i.e. sealed and not leaking). If the label is not clear, then a new label must be affixed.

Old, outdated or unwanted chemicals are picked up for disposal or redistribution by Environmental Health and Safety upon request by the generator.

#### **5.3.6.2.4 Other Chemical Waste**

Other chemical waste not meeting the specification for accumulation in safety cans or metal five gallon cans or are not in their original containers must be accumulated in separate non-leaking sealed containers supplied by the lab.

Use a waste container with a volume as close to that of the waste as possible.

Other chemical waste includes, but is not limited to, acid/base waste, heavy metal waste, aqueous based non-solvent waste, used vacuum pump oil, broken mercury thermometers and contaminated lab ware.

At a minimum, the container must be labeled with the following information:

- Chemical constituents.
- Percentage or amount of each constituent.
- The word "Waste."

This type of chemical waste is picked up for disposal by Environmental Health and Safety upon request by the generator.

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*When in doubt about the disposal of chemicals, consult your supervisor or Environmental Health and Safety.*

**NOTE:** The presence of any radioactivity must be indicated on the hazardous waste labels. These materials must be treated as radioactive waste (Refer to waste handling procedures in **RADIATION SAFETY MANUAL**).

### **5.3.7 Inspection Program**

Maintenance and regular inspection of laboratory equipment are essential parts of the laboratory safety program. Management should participate in the design of a laboratory inspection program to ensure that the facility is safety and healthy, workers are adequately trained, and proper procedures are being followed.

The program includes a combination of routine inspections, self-audits, program audits, peer inspections, EHS inspections, and inspections by external entities.

#### **5.3.7.1 Inspection Elements**

- A checklist is used to ensure all issues are covered and a camera is used to document issues that require correction.
- Conversations with workers should occur during the inspection, as they can provide valuable information and allow inspectors an opportunity to show workers how to fix problems.
- Issues resolved during the inspection should be noted.
- An inspection report containing all findings and recommendations should be prepared for management and other appropriate workers.
- Management should follow-up on the inspection to ensure that all corrections are implemented.

### **5.3.8 Emergency Planning**

In addition to laboratory safety issues, laboratory personnel should be familiar with established facility policies and procedures regarding emergency situations. Topics may include, but are not limited to:

- Evacuation procedures—when it is appropriate and alternate routes;
- Emergency shutdown procedures—equipment shutdown and materials that should be stored safely;
- Communications during an emergency—what to expect, how to report, where to call or look for information;
- How and when to use a fire extinguisher;
- Security issues—preventing tailgating and unauthorized access;
- Protocol for absences due to travel restrictions or illness;

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- Safe practices for power outage;
- Shelter in place—when it is appropriate;
- Handling suspicious mail or phone calls;
- Laboratory-specific protocols relating to emergency planning and response;
- Handling violent behavior in the workplace; and
- First-aid and CPR training, including automated external defibrillator training if available.

It is prudent that laboratory personnel are also trained in how to respond to short-term, long-term and large-scale emergencies. Laboratory security can play a role in reducing the likelihood of some emergencies and assisting in preparation and response for others. Every institution, department, and individual laboratory should consider having an emergency preparedness plan. The level of detail of the plan will vary depending on the function of the group and institutional planning efforts already in place.

Emergency planning is a dynamic process. As personnel, operations, and events change, plans will need to be updated and modified. To determine the type and level of emergency planning needed, laboratory personnel need to perform a vulnerability assessment. Periodic drills to assist in training and evaluation of the emergency plan are recommended as part of the training program.

### 5.3.9 Emergency Procedures

- *Fire alarm policy.* Most organizations use fire alarms whenever a building needs to be evacuated—for any reason. When a fire alarm sounds in the facility, evacuate immediately after extinguishing all equipment flames. Check on and assist others who may require help evacuating.
- *Emergency safety equipment.* The following safety elements should be met:
  - A written emergency action plan has been provided to workers;
  - Fire extinguishers, eyewash units, and safety showers are available and tested on a regular basis; and
  - Fire blankets, first-aid equipment, fire alarms, and telephones are available and accessible.
- *Chemical spills.* Workers should contact the CHO or EHS office for instructions before cleaning up a chemical spill. All SDS and label instructions should be followed, and appropriate PPE should be worn during spill cleanup.
- *Accident procedures.* In the event of an accident, immediately notify appropriate personnel and local emergency responders. Provide an SDS of any chemical involved to the attending physician. Complete an accident report and submit it to the appropriate office or individual within 24 hours.

- *Employee safety training program.* New workers should attend safety training before they begin any activities. Additional training should be provided when they advance in their duties or are required to perform a task for the first time. Training documents should be recorded and maintained. Training should include hands-on instruction of how to use safety equipment appropriately.
- *Conduct drills.* Practice building evacuations, including the use of alternate routes. Practice shelter-in-place, including plans for extended stays. Walk the fastest route from your work area to the nearest fire alarm, emergency eye wash and emergency shower. Learn how each is activated. In the excitement of an actual emergency, people rely on what they learned from drills, practice and training.
- *Contingency plans.* All laboratories should have long-term contingency plans in place (e.g., for pandemics). Scheduling, workload, utilities and alternate work sites may need to be considered.

### 5.3.10 Laboratory Security

Laboratory security has evolved in the past decade, reducing the likelihood of some emergencies and assisting in preparation and response for others. Most security measures are based on the laboratory's vulnerability. Risks to laboratory security include, but are not limited to:

- Theft or diversion of chemicals, biologicals, and radioactive or proprietary materials, mission-critical or high-value equipment;
- Threats from activist groups;
- Intentional release of, or exposure to, hazardous materials;
- Sabotage or vandalism of chemicals or high-value equipment;
- Loss or release of sensitive information; and
- Rogue work or unauthorized laboratory experimentation.

Security systems in the laboratory are used to detect and respond to a security breach, or a potential security breach, as well as to delay criminal activity by imposing multiple layered barriers of increasing stringency.

A good laboratory security system will increase overall safety for laboratory personnel and the public, improve emergency preparedness by assisting with preplanning, and lower the organization's liability by incorporating more rigorous planning, staffing, training, and command systems and implementing emergency communications protocols, drills, background checks, card access systems, video surveillance, and other measures. The security plan should clearly delineate response to security issues, including the coordination of institution and laboratory personnel with both internal and external responders.

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## 6 Criteria for Reducing Employees' Exposure to Hazardous Chemicals

Wright State University's Department of Environmental Health and Safety will follow the procedures listed below, in the order presented, to determine employee's exposure to hazardous chemicals in laboratories covered by OSHA's Laboratory Standard.

### 6.1 Chemical Grouping

Using the chemical inventory for each laboratory, group chemicals according to their Threshold Limit Value (TLV) or Permissible Exposure Limit (PEL), as shown below. Use the level (either the TLV or PEL) that is lowest. If a TLV or PEL does not exist for a particular chemical, use NIOSH's Recommended Exposure Limits (REL). In the absence of a TLV, PEL, or REL, consult with the Director of Environmental Health and Safety. For commercial products having more than 1 chemical component, use the PEL/TLV listed on the Material Safety Data Sheet (MSDS) if one is provided. Should a compound PEL/TLV not be listed, the responsible Industrial Hygienist will consult with the Director of Environmental Health and Safety for guidance.

#### 6.1.1 Chemical Groups

Group 1	Chemicals listed as a known human carcinogen by IARC, NTP and OSHA, and for whom the exposure criteria should be as close to zero as possible.
Group 2	Chemicals with exposure limits of less than 1.0 ppm (parts per million).
Group 3	Chemicals with exposure limits of greater than 1.0 ppm but equal or less than 10.0 ppm.
Group 4	Chemicals with exposure limits of greater than 10.0 ppm.

#### 6.1.2 Evaluating the Use of Chemicals in the Laboratory

The following information is required for the industrial hygienist to determine if exposures are more than the established exposure limit and the action level for a specific chemical could occur in the workplace. The action level, unless specifically listed, is taken to be one-half the value of the TLV, REL or PEL.

1. Room Volume (cubic feet).
2. Quantity of supply air (cubic feet/ hour) provided to the room.
3. Quantity of exhaust air (cubic feet/ hour) from the room. This should involve two calculations; one with general room exhaust only and the second using both the general room exhaust and the chemical fume hood exhaust capacities.
4. Quantity (ml) of chemical(s) used per procedure.
5. Frequency of use.

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6. Exposure limit of the chemical or mixture.
7. How the chemical is used; general room area, in the fume hood, an enclosed process, cold/heated process, etc.
8. Specific gravity of the chemical or compound.
9. Molecular weight of the chemical or compound.

### 6.1.3 Methodology

The following methodology is used to determine if exposures exceeds the permissible exposure limit and the action level for a specific chemical or chemical mixture.

#### 6.1.3.1 Method 1 – Application of Dilution Ventilation

Two formulas for dilution ventilation can be used. The second formula is preferred because it uses less assumptions and its simplicity.

Using either method, the industrial hygienist will make the determination for the action level first. If the 8-hour time-weighted average (TWA) is not exceeded for the action level, there is no need to calculate the TLV/REL/PEL dilution requirements, as it is obvious they cannot be exceeded.

**THE PRINCIPLES OF DILUTION VENTILATION IS TO BE USED ONLY FOR EXPOSURE DETERMINATIONS FOR PROCEDURES CONDUCTED IN THE GENERAL ROOM AREA AND NOT WITHIN THE CONFINES OF A CHEMICAL FUME HOOD OR ENCLOSED SYSTEM.**

**Note: Use of the following formulas assumes that all the chemical evaporates into the room atmosphere under uniform conditions. Knowing that all the chemical does NOT evaporate into the room, the worst-case scenario has been evaluated. For those cases where the existing and required air volumes are close to one another, a closer determination of the quantity actually evaporated may negate the need for general room or personal sampling. The Director of Environmental Health and Safety should be consulted under these conditions.**

#### 6.1.3.1.1 Formula No. 1(ACGIH Manual of Industrial Ventilation 20th Edition)

$$\text{Required Cubic Feet of dilution air /hour} = \frac{0.85 * S.G. * 10^6 * \frac{mL}{hr} * A}{M.W. * \text{Threshold Limit Value}}$$

Where:

S.G = Specific Gravity of the chemical

A = 10 (a conservative protection for poor air mixing and design)

M.W. = Molecular Weight of Chemical

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**REMINDER: USE THE ACTION LEVEL INSTEAD OF THE TLV/REL/PEL FOR THE INITIAL DETERMINATION.**

Compare Required Cubic Feet of Air per hour with the cubic feet of air being exhausted from the laboratory. If the exhausted air EXCEEDS the required air, assume the action level will not be exceeded. If not, general room and personal air samples may be required. Consult with the Director of Environmental Health and Safety. Compare the supply air and exhaust quantities to determine if airflow is positive or negative to the laboratory.

**6.1.3.1.2 Formula No. 2 (Fundamentals of Industrial Hygiene 3<sup>rd</sup> Edition):**

This formula calculates requirements for an 8-hour day. Therefore, one must calculate the amount of air supplied for an 8-hour day. Also, the vapor volume and the amount of the chemical used must be adjusted for an 8-hour day.

$$\text{Volume of Air Required per 8-hour day} = \frac{\text{Total Vapor Volume} * 10^6}{\text{TLV or REL or PEL}}$$

The Total Vapor Volume is determined by:

$$\text{Vapor Volume (ft}^3\text{/mL) * chemical (mL, quantity used/ 8-hour day)}$$

Vapor volumes for chemicals can be found in Appendix C (Fundamentals of Industrial Hygiene), on Safety Data Sheets (SDS) or in any good text on the physical properties of chemicals.

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**REMINDER: USE THE ACTION LEVEL INSTEAD OF THE TLV/REL/PEL FOR THE INITIAL DETERMINATION.**

**6.1.3.2 Method 2 – Operations Conducted in Ventilated Cabinets**

With operations conducted in ventilated cabinets (fume hoods or other exhaust hoods) or in enclosed systems, employees should not be subject to airborne concentrations approaching the action level or permissible exposure limit of a chemical if the hood is located away from major traffic patterns or significant airflows at the face of the hood, operating satisfactorily, and being used according to accepted procedures. The quickest and simplest method to determine if the employee is subject to airborne contaminant outside the hood is to conduct a smoke field or dry ice evaluation while the hood is being used. If no smoke/vapor is observed exiting at the face of the hood under normal operating conditions, it can be assumed that no contaminants are reaching the employee's

breathing zone. Consult with the Director of Environmental Health and Safety if the smoke field evaluation is being considered.

### **6.1.3.3 Method 3 – Air Sampling**

General air and personal air samples at the breathing zone of the employee will be conducted when:

1. The action level or Permissible Exposure Limit (PEL) appears to be exceeded by the dilution ventilation calculations.
2. The smoke field evaluation is questionable or unacceptable.
3. Anytime, when in the professional judgment of the attending industrial hygienist or the Director of Environmental Health and Safety such sampling is warranted.

Air sampling strategy will be as follows:

1. The initial sampling will be accomplished by use of either detector tubes (length of stain) or other monitoring instrumentation. Determinations more than fifty percent (50%) of the action level will give cause to sample by use of personal sampling pumps with the appropriate sample media or any other methodology approved by OSHA for an exposure determination of record.

## **7 Biological Safety**

### **7.1 Introduction**

The hazards that biological agents pose in the laboratory are associated with laboratory-acquired infections. In many cases, the sources of potential infections can be readily identified. Also, in many cases the specific etiological agents are known or there is awareness that the materials with which one is working, e.g., blood or blood products, may contain certain pathogens. Sources of laboratory-acquired infections include in declining order of frequency: bacterial, viral, rickettsial, fungal, chlamydia, parasitic, and unknown. Several deaths have resulted from infections caused by each of these groups. According to Pike<sup>1,2,3</sup>, in the 50 years following 1924, 4079 cases were reported with which 168 deaths were associated. The most prevalent agents identified were, in order of decreasing frequency: brucellosis, Q fever, hepatitis, typhoid fever, tularemia, tuberculosis, Venezuelan equine encephalitis, psittacosis, and coccidioidomycosis. Most disturbing is the fact that Pike could associate a specific accident event with only 18% of these infections. Accidents involving cuts, bites and scratches, spills, sprays, and needlesticks each accounted for approximately one fourth of the incidents. Mouth pipetting accounted for half as many. Other causes of infection cited were from working with the infectious agent (21%), working with animals or ectoparasites infected with the agent (17%), or exposure to aerosols (13%), twenty percent of infections having no known source. Not all laboratory-acquired infections are reported. Few large-scale studies have been conducted, and the data are skewed by clusters of incidences.

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However, it is clear that the acquisition of infections in the laboratory have and does occur.

In recent years, the prevalence of bacterial infection has dropped while viral (60%) and fungal (20%) have risen – hepatitis B, tuberculosis and Shigella being the front runners. Effectively controlling exposure to these sources depends upon your understanding of the factors involved in disease transmission in the laboratory. Methods of transmission include contact (direct and indirect) and vector-borne, but, as with chemical exposure, the routes of infection include ingestion, inhalation, and inoculation. Whether an infection will result depends upon the pathogenicity of the organism, the size of the dose, and your susceptibility. Although we are exposed to various infectious organisms daily outside the laboratory and do not usually succumb to infection, the titers to which we may potentially be exposed in the laboratory can be many fold higher - capable of overwhelming our immune systems.

Containment is the key word in controlling laboratory pathogens. Although engineering design, especially ventilation, is important, your choice of procedures and equipment, and your laboratory and personal hygiene practices, are more important.

## **7.2 Biological Hazards**

Many laboratory workers encounter daily exposure to biological hazards. These hazards are present in various sources throughout the laboratory such as blood and body fluids, culture specimens, body tissue and cadavers, and laboratory animals, as well as other workers. OSHA has additional information on select agents and toxins. These are federally regulated biological agents (e.g., viruses, bacteria, fungi, and prions) and toxins that have the potential to pose a severe threat to public health and safety, to animal or plant health, or to animal or plant products.

The agents and toxins that affect animal and plant health are also referred to as high-consequence livestock pathogens and toxins, non-overlap agents and toxins, and listed plant pathogens.

Select agents and toxins are defined by lists that appear in:

- sections 73.3 of Title 42 of the Code of Federal Regulations (HHS/CDC Select Agent Regulations),
  - sections 121.3 and 121.4 of Title 9 of the Code of Federal Regulations (USDA/APHIS/VS Select Agent Regulations), and
  - section 331.3 of Title 7 of the Code of Federal Regulations (plants - USDA/APHIS/PPQ Select Agent Regulations) and
  - Part 121, Title 9, Code of Federal Regulations (animals – USDA/APHIS).
- Select agents and toxins that are regulated by both HHS/CDC and USDA/APHIS are referred to as “overlap” select agents and toxins (refer to 42 CFR section 73.4 and 9 CFR 121.4). The information below is a starting point

for technical and regulatory information about some of the most virulent and prevalent biological agents and toxins. The OSHA Safety and Health Topics Page entitled Biological Agents can be accessed at: <http://www.osha.gov/SLTC/biologicalagents/index.html>.

### **7.2.1 Anthrax**

Anthrax is an acute infectious disease caused by a spore-forming bacterium called *Bacillus anthracis*. It is generally acquired following contact with anthrax-infected animals or anthrax-contaminated animal products. *Bacillus anthracis* is an HHS and USDA select agent.

### **7.2.2 Avian Flu**

Avian influenza is caused by Influenza A viruses. These viruses normally reside in the intestinal tracts of water fowl and shore birds, where they cause little, if any, disease. However, when they are passed on to domestic birds, such as chickens, they can cause deadly contagious disease, highly pathogenic avian influenza (HPAI). HPAI viruses are considered USDA/APHIS select agents.

### **7.2.3 Botulism**

Cases of botulism are usually associated with consumption of preserved foods. However, botulinum toxins are currently among the most common compounds explored by terrorists for use as biological weapons. Botulinum neurotoxins, the causative agents of botulism, are HHS/CDC select agents.

### **7.2.4 Foodborne Disease**

Foodborne illnesses are caused by viruses, bacteria, parasites, toxins, metals, and prions (microscopic protein particles). Symptoms range from mild gastroenteritis to life-threatening neurologic, hepatic and renal syndromes.

### **7.2.5 Hantavirus**

Hantaviruses are transmitted to humans from the dried droppings, urine, or saliva of mice and rats. Animal laboratory workers and persons working in infested buildings are at increased risk to this disease.

### **7.2.6 Legionnaires' Disease**

Legionnaires' disease is a bacterial disease commonly associated with water based aerosols. It is often the result of poorly maintained air conditioning cooling towers and potable water systems.

### **7.2.7 Molds and Fungi**

Molds and fungi produce and release millions of spores small enough to be air-, water-, or insect-borne which may have negative effects on human health including, allergic reactions, asthma, and other respiratory problems.

### **7.2.8 Plague**

The World Health Organization reports 1,000 to 3,000 cases of plague every year. A bioterrorist release of plague could result in a rapid spread of the pneumonic form of the disease, which could have devastating consequences. *Yersinia pestis*, the causative agent of plague, is an HHS/CDC select agent.

### **7.2.9 Ricin**

Ricin is one of the most toxic and easily produced plant toxins. It has been used in the past as a bioterrorist weapon and remains a serious threat. Ricin is an HHS/CDC select toxin.

### **7.2.10 Severe Acute Respiratory Syndrome (SARS)**

SARS is an emerging, sometimes fatal, respiratory illness. According to the Centers for Disease Control and Prevention (CDC), the most recent human cases of SARS were reported in China in April 2004 and there is currently no known transmission anywhere in the world.

### **7.2.11 Smallpox**

Smallpox is a highly contagious disease unique to humans. It is estimated that no more than 20 percent of the population has any immunity from previous vaccination. *Variola major* virus, the causative agent for smallpox, is an HHS/CDC select agent.

### **7.2.12 Tularemia**

Tularemia is also known as “rabbit fever” or “deer fly fever” and is extremely infectious. Relatively few bacteria are required to cause the disease, which is why it is an attractive weapon for use in bioterrorism. *Francisella tularensis*, the causative agent for tularemia, is an HHS/CDC select agent.

### **7.2.13 Viral Hemorrhagic Fevers (VHFs)**

Hemorrhagic fever viruses are among the agents identified by the Centers for Disease Control and Prevention (CDC) as the most likely to be used as biological weapons. Many VHFs can cause severe, life-threatening disease with high fatality rates. Many VHFs are HHS/CDC select agents; for example, Marburg virus, Ebola viruses, and the Crimean-Congo hemorrhagic fever virus.

### **7.2.14 Pandemic Influenza**

A pandemic is a global disease outbreak. An influenza pandemic occurs when a new influenza virus emerges for which there is little or no immunity in the human population; begins to cause serious illness; and then spreads easily person-to-person worldwide. The list above does not include all the biological agents and toxins that may be hazardous to laboratory workers. New agents will be added over time. For agents that may pose a hazard to laboratory workers but are not

listed above, consult the CDC web page at: <http://www.cdc.gov>. See Table 7 for more information on BSL levels.

An additional OSHA Safety and Health Topics page on Pandemic Influenza has been added in response to the 2009 H1N1 influenza pandemic. It can be accessed at: [www.osha.gov/dsg/topics/pandemicflu/index.html](http://www.osha.gov/dsg/topics/pandemicflu/index.html)

### **7.3 Pathogen Safety Data Sheets (PSDSs) on Infectious Agents**

Although SDSs for chemical products have been available to workers for many years in the U.S. and other countries, Canada is the only country that has developed SDSs for infectious agents. These PSDSs were produced by the Canadian Public Health Agency for personnel working in the life sciences as quick safety reference material relating to infectious microorganisms. These PSDSs on Infectious Agents are organized to contain health hazard information such as infectious dose, viability (including decontamination), medical information, laboratory hazard, recommended precautions, handling information and spill procedures. These PSDSs are available at: <http://www.phac-aspc.gc.ca/msds-ftss>.

### **7.4 Bloodborne Pathogens**

The OSHA Bloodborne Pathogens (BBP) standard (29 CFR 1910.1030) is designed to protect workers from the health hazards of exposure to bloodborne pathogens. Wright State is subject to the BBP standard since some workers jobs put them at reasonable risk of coming into contact with blood or other potentially infectious materials (OPIM).

Wright State has a written Exposure Control Plan, provides training to exposed workers, and complies with other requirements of the standard, including use of Standard Precautions when dealing with blood and OPIM. In 2001, in response to the Needlestick Safety and Prevention Act, OSHA revised the Bloodborne Pathogens standard. The revised standard clarifies the need for employers to select safer needle devices and to involve workers in identifying and choosing these devices. The updated standard also requires employers to maintain a log of injuries from contaminated sharps.

OSHA estimates that 5.6 million workers in the healthcare industry and related occupations are at risk of occupational exposure to bloodborne pathogens, including HIV, HBV, HCV, and others.

All occupational exposure to blood or OPIM places workers at risk for infection with bloodborne pathogens.

OSHA defines blood to mean human blood, human blood components, and products made from human blood.

OPIM means:

- The following human body fluids: semen, vaginal secretions, cerebrospinal fluid, synovial fluid, pleural fluid, pericardial fluid, peritoneal fluid, amniotic fluid, saliva in dental procedures, any body fluid that is visibly contaminated with blood, and all body fluids in situations where it is difficult or impossible to differentiate between body fluids;
- Any unfixed tissue or organ (other than intact skin) from a human (living or dead); and
- HIV- or HBV-containing cell or tissue cultures, organ cultures, and HIV or HBV-containing culture medium or other solutions; and
- blood, organs, or other tissues from experimental animals infected with HIV or HBV.

The Centers for Disease Control and Prevention (CDC) notes that although more than 200 different diseases can be transmitted from exposure to blood, the most serious infections are hepatitis B virus (HBV), hepatitis C virus (HCV), and human immunodeficiency virus (HIV). Fortunately, the risk of acquiring any of these infections is low. HBV is the most infectious virus of the three viruses listed above. For an unvaccinated healthcare worker, the risk of developing an infection from a single needlestick or a cut exposed to HBV-infected blood ranges from 6-30%. The risk for infection from HCV- and HIV-infected blood under the same circumstances is 1.8 and 0.3 percent, respectively. This means that after a needlestick/cut exposure to HCV-contaminated blood, 98.2% of individuals do not become infected, while after a similar exposure to HIV-contaminated blood, 99.7% of individuals do not become infected.

([http://www.cdc.gov/OralHealth/infectioncontrol/faq/bloodborne\\_exposures.htm](http://www.cdc.gov/OralHealth/infectioncontrol/faq/bloodborne_exposures.htm)).

Many factors influence the risk of becoming infected after a needlestick or cut exposure to HBV-, HCV- or HIV-contaminated blood. These factors include the health status of the individual, the volume of the blood exchanged, the concentration of the virus in the blood, the extent of the cut or the depth of penetration of the needlestick, etc.

#### **7.4.1 Required Training and Practices**

Workers are trained and prohibited from engaging in the following activities:

- Mouth pipetting/suctioning of blood or OPIM, 29 CFR 1910.1030(d)(2)(xii);
- Eating, drinking, smoking, applying cosmetics or lip balm, or handling contact lenses in work areas where there is a reasonable likelihood of occupational exposure to blood or OPIM, 29 CFR 1910.1030(d)(2)(ix); and
- Storage of food or drink in refrigerators, freezers, shelves, cabinets or on countertops or benchtops where blood or OPIM are present, 29 CFR 1910.1030(d)(2)(x).



#### 7.4.2 Employer Responsibilities

Wright State as the employers must ensure that the following are provided:

- Appropriate PPE for workers if blood or OPIM exposure is anticipated, 29 CFR 1910.1030(d)(3); The type and amount of PPE depends on the anticipated exposure.
- Gloves must be worn when hand contact with blood, mucous membranes, OPIM, or non-intact skin is anticipated, or when handling contaminated items or surfaces, 29 CFR 1910.1030(d)(3)(ix).
- Surgical caps or hoods and/or shoe covers or boots must be worn in instances when gross contamination can reasonably be anticipated such as during autopsies or orthopedic surgery, 29 CFR 1910.1030(d) (3)(xii).
- Effective engineering and work practice controls to help remove or isolate exposures to blood and bloodborne pathogens, 29 CFR 1910.1030(d)(2)(i), CPL 02-02-069 (CPL 2-2.69); and
- Hepatitis B vaccination (if not declined by a worker) under the supervision of a physician or other licensed healthcare professional to all workers who have occupational exposure to blood or OPIM, 29 CFR 1910.1030(f)(1)(ii)(A)-(C).

#### 7.4.3 Labels

When any blood, OPIM or infected animals are present in the work area, a hazard warning sign (see graphic) incorporating the universal biohazard symbol, 29 CFR 1910.1030(g)(1)(ii)(A), must be posted on all access doors, 29 CFR 1910.1030(e) (2)(ii)(D).



**Figure 2: Biohazard Symbol**

#### 7.4.4 HIV/HBV Laboratories

##### 7.4.4.1 Engineering Controls and Work Practices for All HIV/HBV Laboratories

Employers must ensure that:

- All activities involving OPIM are conducted in Biological Safety Cabinets (BSCs) or other physical-containment devices; work with OPIM must not be conducted on the open bench, 29 CFR 1910.1030(e)(2)(ii)(E);
- Certified BSCs or other appropriate combinations of personal protection or physical containment devices, such as special protective clothing, respirators, centrifuge safety cups, sealed centrifuge rotors, and containment caging for animals, be used for all activities with OPIM that pose a threat of exposure to droplets, splashes, spills, or aerosols, 29 CFR 1910.1030(e)(2)(iii)(A);
- Each laboratory contains a facility for hand washing and an eyewash facility which is readily available within the work area, 29 CFR 1910.1030(e)(3)(i); and
- Each work area contains a sink for washing hands and a readily available eyewash facility. The sink must be foot, elbow, or automatically operated and must be located near the exit door of the work area, 29 CFR 1910.1030(e)(4)(iii).

#### 7.4.5 HIV and HBV Research Laboratories

Additional BBP Standard Requirements Apply to HIV and HBV Research Laboratories

Requirements include:

##### 7.4.5.1 Waste materials:

- All regulated waste must either be incinerated or decontaminated by a method such as autoclaving known to effectively destroy bloodborne pathogens, 29 CFR 1910.1030(e)(2)(i); and
- Contaminated materials that are to be decontaminated at a site away from the work area must be placed in a durable, leak-proof, labeled or color-coded container that is closed before being removed from the work area, 29 CFR 1910.1030(e)(2)(ii)(B).

##### 7.4.5.2 Access

- Laboratory doors must be kept closed when work involving HIV or HBV is in progress, 29 CFR 1910.1030(e)(2)(ii)(A);
- Access to the production facilities' work area must be limited to authorized persons. Written policies and procedures must be established whereby only persons who have been advised of the potential biohazard, who meet any specific entry requirements, and who comply with all entry and exit

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procedures must be allowed to enter the work areas and animal rooms, 29 CFR 1910.1030(e)(2)(ii)(C);

- Access doors to the production facilities' work area or containment module must be self-closing, 29 CFR 1910.1030(e)(4)(iv);
- Work areas must be separated from areas that are open to unrestricted traffic flow within the building. Passage through two sets of doors must be the basic requirement for entry into the work area from access corridors or other contiguous areas. Physical separation of the high-containment work area from access corridors or other areas or activities may also be provided by a double-doored clothes change room (showers may be included), airlock, or other access facility that requires passing through two sets of doors before entering the work area, 29 CFR 1910.1030(e)(4)(i); and
- The surfaces of doors, walls, floors and ceilings in the work area must be water-resistant so that they can be easily cleaned. Penetrations in these surfaces must be sealed or capable of being sealed to facilitate decontamination, 29 CFR 1910.1030(e)(4)(ii).

(These requirements **do not apply** to clinical or diagnostic laboratories engaged solely in the analysis of blood, tissue, or organs, 29 CFR 1910.1030(e)(1).)

### **7.5 General Precautions for Biological Work**

If you are or will be at risk of infection from an agent for which there is a vaccine, e.g., hepatitis, you should consult a supervisor or the Environmental Health and Safety Department about immunization.

Inform Environmental Health and Safety of receipt of any biohazardous agent or materials containing such agents, include information on the storage location and handling and use precautions, and emergency procedures. Use a biohazard warning symbol to designate the storage location of human blood, blood products and any pathogenic agents. If work is conducted at a Biosafety Level 2 (BL2) or above, a warning sign identifying the agent, emergency contact person, and any special precautions must be posted on the laboratory door as well. (See the appendix at the end of this section for a summary of biosafety levels and a description of BL2 and BL3 criteria.)



**Figure 3: Biohazard Symbol**

### 7.6 Standard Microbiological Practices

Standard microbiological practices are common to all laboratories. Special microbiological practices enhance worker safety, environmental protection, and address the risk of handling agents requiring increasing levels of containment.

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*Do not eat, drink, store food, apply cosmetics or smoke in the laboratory.*

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Never mouth pipet, it is unsafe. Numerous types of pipetting aids are available.

Wear disposable high-cuffed disposable gloves, but note that latex gloves are permeable to organic solvents, including ethanol. Given that thin gloves offer little protection against cuts, bites, and self-inoculation, wear the thickest gloves the dexterity required by your work permits; wearing two pairs of thinner gloves allows for safe removal of contaminated outer gloves. See **GENERAL SAFETY PRACTICES** for more information on selection and use of gloves.

Wear lab coats. The primary reason for wearing a lab coat is to protect yourself from contact with hazardous materials. However, the liberation of microorganisms from human skin plays an important role in transmission of airborne infection to humans or experimental materials, wearing a lab coat can help minimize this transmission. To prevent clothing from acting as a bellows, the front of the lab coat should be closed and sleeves should be tucked inside gloves

or taped at the wrists. Disposable, Tyvek™ lab coats are also available and recommended for work in biological safety cabinets.

**Do Not** wear lab coats outside the laboratory environment.

In general, a Class II biological safety cabinet should be used for work with biohazards.

Aerosol generating procedures should be performed in an appropriate enclosure, e.g., the rear third of a biosafety cabinet, (see “Aerosol-Generating Processes” later in this manual). Remember that even a drop falling onto a hard surface can generate an aerosol. Using fluorescein and black light, one can test for aerosol escape.

Avoid the use of needles, scalpels, and other sharp implements. If needles and syringes must be used, cover the tip with absorbent material when adjusting the volume or withdrawing the tip from a septum or injection site. Dispose of sharps in a puncture resistant, leak resistant container. Do not resheath or remove used needles; insert the whole assembly into the container. These containers must be tightly closed to prevent loss of contents, must be labeled “**SHARPS**” and be marked with the international biohazard symbol. All punctures should be washed with soap and water and reported to a supervisor or Environmental Health and Safety.

If experimentation requires the use of pathogens, first develop and test all procedures using non-pathogenic agents.

Use disposable glass or plastic ware. If non-disposable glassware must be used, disinfect contaminated items before cleaning.

Clean up spills immediately with a fresh solution of chlorine bleach solution at a strength of at least fifteen percent.

All waste materials must be accumulated in red or other colored plastic bags labeled with the international biohazard symbol.

Discard non-sharp disposable materials, e.g., gloves, pipets, pipet tips, plastic tubes, that come in contact with blood or potentially infectious materials in red or other colored plastic bags. Treat blood and other potentially infectious fluids with a 15% chlorine bleach solution and decant down the drain. Do not dispose of blood or sharps with the normal laboratory trash.

## **7.7 Guidelines for Specific Subjects of Study**

### **7.7.1 Experiments Using Blood, Blood Products or Human Secretions**

Persons who work with blood or blood products are at increased risk of hepatitis in proportion to the degree of their exposure. Hepatitis B vaccination is recommended for all individuals working with blood or blood products. The most important way for personnel handling blood products to protect themselves from hepatitis B infection (as well as from other blood-borne infections) is to follow the

general precautions outlined above. Handle all blood, blood products and human secretions as if infective (see Wright State University's **EXPOSURE CONTROL PLAN, BLOODBORNE PATHOGENS**).

If an exposure to blood or blood products occurs, report immediately to the Environmental Health and Safety Department for evaluation and possible treatment with hepatitis immune globulin, which, if administered soon after exposure, may prevent acquisition of hepatitis.

### 7.7.2 Recombinant DNA Experiments

The vast majority of laboratory experiments are exempt from the NIH guidelines if the recombinant DNA molecules:

- are not in organisms or viruses
- consist entirely of DNA segments from a single nonchromosomal or viral DNA source
- consist entirely of DNA from a prokaryotic or eukaryotic host
- consist entirely of plasmids (excluding viruses) when propagated in that host (or a closely related strain of the same species)
- were transferred to another host by well-established physiological means (prokaryotic DNA only)
- contains less than one-half of any eukaryotic genome that is propagated and maintained in cells in tissue culture
- use *Escherichia coli* K-12 host-vector systems (some exceptions apply)
- use *Saccharomyces cerevisiae* host-vector systems (some exceptions apply)
- use any asporogenic *Bacillus subtilis* strain that does not revert to a spore former with a frequency greater than  $10^{-7}$  (some exceptions apply)
- derived entirely from extrachromosomal elements of certain organisms.

### 7.7.3 Work with Potentially Infectious Agents

In "*The Transforming Principle*"<sup>4</sup>, Dr. McCarty, who along with MacLeod and Avery discovered that DNA was the genetic material, describes the standards set by Professor Avery: "He would then review the protocol...in this manner I was introduced to Avery's extraordinarily rigorous bacteriological technique...he...had agreed that they would treat all bacterial cultures as though they contained the plague bacillus...it was a common failing to become sloppy in handling nonpathogenic organisms which in turn led to some relaxation of acceptable techniques when dealing with more infectious agents." Although the advent of the biological safety cabinet has obviated the need for a flame, in fact made its use undesirable, the rigors of Avery's protocol review and careful approach are still highly recommended even when handling "normal" cell lines, some of which may present risks to investigators as well as compromise their experiments. For most biohazardous agents, the routes of potential infection are inoculation, ingestion

and inhalation. The general laboratory procedures detailed in **GENERAL SAFETY PRACTICES** should be used to reduce exposure to biological agents. This includes wearing gloves, a lab coat and safety glasses if the organism can infect the eye, using a biological safety cabinet if appropriate as outlined below, and decontaminating all biological wastes before disposal. Decontamination of wastes can be accomplished for all these agents by autoclaving, which may require 60 minutes for a full load, or 20-minute exposure to fresh 15% chlorine bleach. Note that the color change associated with oxidation of media is not a good indicator of inactivation.

The Biosafety Levels (BL) cited below represent a set of standards, special practices, containment equipment, etc., assigned by the CDC (Table 8: CDC Summary of Recommended Biosafety Levels for Infectious Agents). The BL number increases with increasing hazard. Often work at a higher BL is recommended when large volumes, highly concentrated stocks, or aerosol generating procedures are employed. All human specimens should be regarded and handled as infective. The risk from human specimens is not restricted to hepatitis or AIDS but includes many other agents, including some of those listed below, which may be found in blood, blood products, urine, feces, amniotic fluid, etc. Researchers frequently receive blood which has been designated “not for transfusion” and other fresh specimens which have not been screened for these agents. Research with human specimens is BL 2/3 (see the appendix at the end of this section for the definition of BL conditions), and the procedures outlined for these levels should be followed. Personnel who will be exposed to blood or blood products should first be immunized with the Hepatitis B vaccine.

**Table 8: CDC Summary of Recommended Biosafety Levels for Infectious Agents**

Biosafety Level	Agent Characteristics	Practices	Safety Equipment	Facilities (secondary barriers)
BSL-1	Not known to consistently cause disease in health adults	Standard microbiological practices	None	Open bench top sink
BSL-2	Associated with human disease, hazard from percutaneous injury, ingestion, mucous membrane exposure	Standard microbiological practices Limited access Biohazard warning signs Sharps precautions Biosafety manual defining any needed waste decontamination	Class I or II biosafety cabinets (BSCs) or other containment devices used for all agents that cause splashes or aerosols of infectious	Open bench top sink Autoclave

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		or medical surveillance policies.	materials Laboratory coats and gloves Face protection as needed	
BSL-2	Indigenous or exotic agents with potential for aerosol transmission; disease may have serious or lethal consequences	All BSL-2 practices Controlled access Decontamination of all waste Decontamination of laboratory clothing before laundering Baseline serum	Class I or II BSCs or other physical containment devices used for all open manipulations of agents Protective lab clothing and gloves Respiratory protection as needed	Open bench top sink Autoclave Physical separation from access corridors Self-closing, double-door access Exhaust air not recirculated Negative airflow in laboratory
BSL-4	Dangerous/exotic agents which pose high risk of life-threatening disease; aerosol-transmitted lab infections; or related agents with unknown risk of transmission	All BSL-3 practices Clothing change before entering Shower on exit All material decontaminated on exit from facility	All procedures conducted in Class III BSCs, or Class I or II BSCs in combination with full-body, air-supplied, positive pressure personnel suit.	BSL-3 plus: Separate building or isolated zone Dedicated supply and exhaust, vacuum, and decontamination systems Other requirements outline the text

#### 7.7.4 Cell Lines

While data on specific cell lines have been omitted, it is important to recognize that there is no “normal” cell line; many reputedly “normal” lines harbor viruses and potentially hazardous gene sequences. Handle these materials as if infectious and decontaminate culture wastes prior to disposal. All cells should be fixed before subjecting them to an aerosol generating process, e.g., flow cytometers.

#### 7.7.5 Viruses

Fluids, tissues, isolates and cell cultures containing infectious viruses pose a risk following exposure by ingestion, percutaneous or parenteral inoculation, and droplet or aerosol contamination of the mucous membranes of the eyes, nose or mouth or of broken skin. The aerosol risk from handling large volumes and

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concentrated stocks is great since some viruses are stable at ambient temperatures and withstand drying. Variation in viral structures results in differential susceptibility to “germicidal” agents and detergents; however, autoclaving and chlorine bleach treatment are usually effective. See the appendix at the end of this section for a chart showing the relative risk from oncogenic viruses.

#### 7.7.6 Bacteria

Many bacteria are ubiquitous, but some of these such as *Staphylococcus aureus* and group A streptococci are responsible for serious infections in man. The potential routes of exposure are as discussed above for viruses. Aerosols are of major concern when working with large volumes or concentrated stocks, and with pathogenic spore forming species since spores resist adverse or extreme conditions. Safety glasses should be worn when handling bacteria which infect the conjunctiva, e.g., *N. gonorrhoea*. Work at a higher biosafety level is recommended when large volumes, highly concentrated stocks, or aerosol generating procedures are employed with infectious bacteria. All wastes must be decontaminated prior to disposal; chlorine bleach treatment is effective.

#### 7.7.7 Parasites

Infective stages of protozoal parasites of humans may be present in blood, feces, lesion exudates, and infected arthropods. Depending on the parasite, accidental parenteral inoculation, transmission by arthropod vectors, skin penetration including bites from infected animals, and ingestion are the primary laboratory hazards. Aerosol or droplet exposure of the mucous membranes of the eyes, nose, or mouth with trophozoites are potential hazards when working with cultures of *Leishmania* and *Trypanosoma* species. All exposure should be reported to a supervisor and Environmental Health and Safety and treated immediately, e.g., wipe bite with 70% ethanol or irrigate eye with distilled water. In general, protozoa are very fragile, sensitive to drying, and, with notable exceptions such as *T. cruzi*, lysed even by water; however, all spills and waste must be actively treated. BL2 containment and procedures are recommended for work with all parasites except *Babesia*.

#### 7.7.8 Fungi

Fungi are in general not significant causes of human disease. Transmission of fungal diseases from person to person is extremely rare. Fungal spores, however, are generally very allergenic and some of the fungal constituents and by-products can be highly toxic, such as the well-known aflatoxin B. The more common hazardous fungi used in laboratories include: *Blastomyces dermatitides*, *Coccidioides immitis*, *Cryptococcus neoformans*, *Histoplasma capsulatum*, *Sporothrix schenckii*. These agents should be handled at BL2 levels.

A classification of microorganisms according to hazards is presented in Appendix B of the Guidelines for Recombinant DNA Research. Note that

agents of class 1-4 should be handled according to biosafety containment levels 1-4 and that there are restrictions against importation of class 5 agents. For annotation of this list see recent National Research Council publications.<sup>5</sup> Additional information can be quickly retrieved from the report of the American Public Health Association.<sup>6</sup>

## **7.8 Research Animals**

All vertebrate animal experimentation requires the approval of the Institutional Animal Care and Use Committee (IACUC). Animals are to be housed only in accredited animal facilities. Users of laboratory animals must recognize that virtually all laboratory animal species can carry pathogens which are infectious to humans. Inoculated animals readily transmit viruses to cage mates by inhalation and contact with urine, feces, sputum, etc. Caution should be taken when working with any animal. Concern for the health of others who do not work directly with animals should be paramount when laboratory animals are transported or used in general laboratory areas outside of an animal facility.

All procedures on animals should be performed by properly trained personnel. By using safe work practices and appropriate PPE, 29 CFR 1910.132(a), workers can minimize the likelihood that they will be bitten, scratched, and/or exposed to animal body fluids and tissues.

### **7.8.1 Possible Injuries/Illnesses**

The most common work-related health complaints reported by individuals working with small animals

are the following:

- Sprains;
- Strains;
- Bites; and
- Allergies.

Of these injuries, allergies (i.e., exaggerated reactions by the body's immune system) to proteins in small animals' urine, saliva, and dander are the greatest potential health risk. An allergic response may evolve into life-long asthma. Because mice and rats are the animals most frequently used in research studies, there are more reports of allergies to rodents than other laboratory animals. Most workers who develop allergies to laboratory animals will do so within the first twelve months of working with them. Sometimes reactions only occur in workers after they have been handling animals for several years. Initially, the symptoms are present within minutes of the worker's exposure to the animals.

Approximately half of allergic workers will have their initial symptoms subside and then recur three or four hours following the exposure.

### **7.8.1.1 Prevention Practices**

Employers should adopt the following best practices to reduce allergic responses of workers:

- Eliminate or minimize exposure to the proteins found in animal urine, saliva and dander.
- Limit the chances that workers will inhale or have skin contact with animal proteins by using well-designed air handling and waste management systems.
- Have workers use appropriate PPE (e.g., gloves, gowns, hair covers, respirators) to further minimize their risk of exposure.

### **7.8.2 Requirements**

- Protocols involving the acute and/or chronic use of hazardous chemicals, radioisotopes, or biohazards in animals must be reviewed with the Laboratory Animal Utilization Committee and Environmental Health and Safety prior to initiation.
- Anyone planning to work with live vertebrates must receive documented training in their handling. Proper handling and restraint techniques reduce the chances for bites and scratches, and training is required by law.
- Dosed animals may be transported to or from an animal facility, containment area, or laboratory only in a cage with a cover.
- Experimental materials and specimens must be transported in closed containers inside unbreakable canisters.

### **7.8.3 Additional Information**

#### **7.8.3.1 Allergic responses**

- Allergic responses to laboratory animals are the most common cause of human disease related to the use of animals in research. Allergies result from the direct or indirect exposure to allergens such as skin contact or inhalation of fur, dander, saliva, urine, serum, etc. Symptoms can vary from wheezing, sneezing and rhinitis to itching eyes and skin, obvious rashes and asthma. Do not ignore the symptoms. Continued exposure can lead to anaphylaxis and can be life-threatening.

#### **7.8.3.2 Immunizations**

- All users of laboratory animals should have an active tetanus immunization and others as appropriate, e.g., rabies.

### 7.8.3.3 Bites or scratches

- Bites or scratches that break the skin should be washed thoroughly with soap and water and be reported to a supervisor and Environmental Health and Safety.

### 7.8.3.4 Personal protective equipment

- Wearing a face mask or respirator, gloves and a lab coat is strongly encouraged for users of animals to reduce aerosol, direct contact, or inadvertent oral and nasal contact with contaminated hands.
- A full-face respirator is recommended for those at high risk.
- Lab coats should be changed and hands thoroughly washed if an animal, its fluids, or feces is touched. **Do Not** wear lab coats outside the lab environment.

### 7.8.3.5 Pregnant employees

- Pregnant employees should not expose themselves to feces, dander or biohazard areas, and should suspend work involving the handling of cats and monkeys. Likewise, pregnant women without immunity to toxoplasmosis should avoid cat contact to avoid the possibility of congenital disease and fetal death.

## 7.8.4 Zoonotic Diseases

A host of possible infectious agents can be transferred from animals to humans. These infections are referred to as zoonotic diseases. The common routes of exposure to infectious agents are inhalation, inoculation, ingestion and contamination of skin and mucous membranes. Inhalation hazards may arise during work practices that can generate aerosols. These include the following: centrifugation, mixing (e.g., blending, vortexing, and sonication), pouring/decanting and spilling/splashing of culture fluids. Inoculation hazards include needlesticks and lacerations from sharp objects. Ingestion hazards include the following: splashes to the mouth, placing contaminated articles/fingers in mouth, consumption of food in the laboratory, and mouth pipetting. Contamination of skin and mucous membranes can occur via splashes or contact with contaminated fomites (e.g., towels, bedclothes, cups, money). Some of the zoonotic diseases that can be acquired from animals are listed below.

### 7.8.4.1 Zoonotic Diseases – Wild and Domesticated Animals

Wild rodents and other wild animals may inflict an injury such as a bite or scratch. Workers need to receive training on the correct way to capture and handle any wild animals. While they may carry or shed organisms that may be potentially infectious to humans, the primary health risk to individuals working with captured

animals is the development of an allergy. The development of disease in the human host often requires a preexisting state that compromises the immune system. Workers who have an immune compromising medical condition or who are taking medications that impair the immune system (e.g., steroids, immunosuppressive drugs, or chemotherapy) are at higher risk for contracting a rodent disease.

Wild rodents may act as carriers for viruses such as Hantavirus and lymphocytic choriomeningitis virus (LCMV) depending on where they were captured. Additionally, each rodent species may harbor their own range of bacterial diseases, such as tularemia and plague. These animals may also have biting insect vectors that can act as a potential carrier of disease (mouse to human transmission).

#### 7.8.4.1.1 Dogs and Cats

Hazards while working with dogs and cats include bite wound infections, cat scratch disease, toxoplasmosis, visceral larval migrans and sarcoptic mange from dogs and fungus such as ringworm from cats are common.

#### 7.8.4.1.2 Rodents

When working with rodents, precautions should be taken against toxoplasmosis, lymphocytic choriomeningitis, *Salmonella*, *Shigella* and ringworm. Toxoplasmosis is one of the most commonly acquired parasitic diseases in the laboratory.

#### 7.8.4.1.3 Rabbits, Sheep, Swine, and Birds

Rabbits, Sheep, Swine, and Birds can be the source of tularemia, Q fever, *Erysipelas* and *Chlamydia* (psithcosis), respectively.

Examples of zoonotic diseases that can be transmitted from wild and domesticated animals to humans are listed in the following table. ( Table 9: Most common zoonotic diseases in workers)

**Table 9: Most common zoonotic diseases in workers**

Disease	Disease Agent	Cats	Dogs	Birds	Farm Animals	Wild Animals
Brucellosis	<i>Brucella canis</i>		X			
Campylobacteriosis	<i>Campylobacter jejuni</i>	X	X		X	
Cat Scratch Fever	<i>Bartonella henselae</i>	X				
Cryptococcosis	<i>Cryptococcus neoformans</i> and other species			X		

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Hemorrhagic fever with renal syndrome (HFRS) and hantavirus pulmonary syndrome (HPS)	<i>Hantavirus</i>					X
Lymphocytic choriomeningitis	<i>Lymphocytic choriomeningitis virus (LCMV)</i>			X		
<i>Pasteurella pneumonia</i>	<i>Pasteurella haemolytica</i>				X	
Histoplasmosis	<i>Histoplasma capsulatum</i>			X		
Orf	Poxvirus				X	
Plague	<i>Yersinia pestis</i>					X
Q-fever	<i>Coxiella burnetti</i>				X	
Rabies	Rabies virus	X				
Salmonellosis	<i>Salmonella enterica</i> serovar Typhi				X	
Toxoplasmosis	<i>Toxoplasma gondii</i>	X				
Tularemia	<i>Tularemia francisella</i>					X

#### 7.8.4.2 Zoonotic Diseases – Non-human Primates

It should not be surprising that, given our many similarities, humans and non-human primates are susceptible to similar infectious agents. Because of our differences, the consequences of infection with the same agent often vary considerably. Infection may cause few if any symptoms in one group and may be lethal to the other. Exposures to body fluids from non-human primates should be treated immediately.

In 2003, a report entitled, Occupational Safety and Health in the Care and Use of Non-Human Primates was published. This report covers topics relevant to facilities in which non-human primates are housed or where non-human primate blood or tissues are handled. The report describes the hazards associated with work involving nonhuman primates and discusses the components of a successful occupational health and safety program, including hazard identification, risk assessment and management, institutional management of workers after a suspected occupational exposure, applicable safety regulations, and personnel training.

Diseases such as tuberculosis, shigella, campylobacter and salmonella can be a serious threat. Herpesvirus B carried by rhesus, cynomolgus, and other old world monkeys can cause fatal encephalitis in man.

### 7.8.4.3 Prevention practices

Laboratory workers need training to adhere to the following good practices to prevent exposure to zoonotic diseases when working with research animals:

- Avoid use of sharps whenever possible. Take extreme care when using a needle and syringe to inject research animals or when using sharps during necropsy procedures. Never remove, recap, bend, break, or clip used needles from disposable syringes. Use safety engineered needles when practical.
- Take extra precautions when handling hooved animals. Due to the physical hazards of weight and strength of the animal, large hooved mammals pose additional concerns for workers. Hooved mammals may resist handling and may require multiple workers to administer medication or perform other functions.
- Keep hands away from mouth, nose and eyes.
- Wear appropriate PPE (i.e., gloves, gowns, face protection) in all areas within the animal facility.
- A safety specialist may recommend additional precautions, based upon a risk assessment of the work performed.
- Wear tear-resistant gloves to prevent exposure by animal bites. Micro-tears in the gloves may compromise the protection they offer.
- Remove gloves and wash hands after handling animals or tissues derived from them and before leaving areas where animals are kept.
- Use mechanical pipetting devices (no mouth pipetting).
- Never eat, drink, smoke, handle contact lenses, apply cosmetics, or take or apply medicine in areas where research animals are kept.
- Perform procedures carefully to reduce the possibility of creating splashes or aerosols.
- Contain operations that generate hazardous aerosols in BSCs or other ventilated enclosures, such as animal bedding dump stations.
- Wear eye protection.
- Wear head/hair covering to protect against sprays or splashes of potentially infectious fluids.
- Keep doors closed to rooms where research animals are kept.
- Clean all spills immediately.
- Report all incidents and equipment malfunctions to the supervisor.
- Promptly decontaminate work surfaces when procedures are completed and after surfaces are soiled by spills of animal material or waste.
- Properly dispose of animal waste and bedding.
- Workers should report all work-related injuries and illnesses to their supervisor immediately.
- Following a bite by an animal or other injury in which the wound may be contaminated, first aid should be initiated at the work site.

- Contaminated skin and wounds should be washed thoroughly with soap and water for 15 minutes.
- Contaminated eyes and mucous membranes should be irrigated for 15 minutes using normal saline or water.
- Consult an occupational health physician concerning wound care standard operating procedures (SOPs) for particular animal bites/scratches.

#### **7.8.5 Anesthetic Agents**

The choice of anesthesia should be made with care and after consultation with the animal facility staff. Neither diethyl ether nor chloroform should be used routinely for anesthesia or euthanasia of laboratory animals. Chloroform is a potent hepatotoxin and a suspected human carcinogen. The introduction of ether to cold rooms, refrigerators and freezers and to an incinerator via animal carcasses presents very real hazards due to its explosive characteristics.

Volatile chemicals for anesthesia or euthanasia should be used only in the presence of adequate ventilation, i.e., a fume hood or closed system with scavenger designed for this purpose. This requirement is especially noteworthy when working with halothane derivatives since they have been shown to have very adverse effects on some individuals. If you must use inhalants for anesthesia or euthanasia, be advised that enflurane or isoflurane are less toxic to humans than other halothanes, including methoxyflurane, and they provide excellent control of narcosis. However, frequency of exposure is critical following sensitization; the idiosyncratic response of individuals is difficult to predict and can be fatal.

##### **7.8.5.1 Recommendations**

- Ether and chloroform should not be used for anesthesia because of flammability of the former and toxicity and carcinogenicity of the latter.
- Methoxyflurane is the recommended agent for most brief, bench-top surgeries on rodents. Its low vapor pressure and high lipid solubility permit safe induction in a closed jar and intermittent application of a nose cone for maintenance. Careful attention to safe work practices is required to control exposure of the investigators.
- Isoflurane rather than halothane is recommended for anesthesia when delivered with a precision vaporizer. Isoflurane has an excellent margin of patient safety as well as minimal adverse side effects and occupational health risks.
- While some injectable drug combinations may be appropriate for specific physiological studies involving extended bench-top surgical procedures, avoidance of injectables and the associated risk of needlesticks is recommended.
- Nitrous oxide use with volatile anesthetics should be avoided since it is not essential for animal surgery and is toxic to humans.

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### 7.8.6 Perfusion

Perfusion of animals should be conducted in a fume hood over a waste collection table/vessel. The waste should be handled as hazardous chemical waste.

## 7.9 Processes and Equipment

### 7.9.1 Aerosol-Generating Processes

Aerosols (dispersions of particles in air) can result from the use of blenders, mixers, sonicators, cell disrupters, centrifuges, syringes, pipets, aspirators, test and centrifuge tube caps. (The hazards associated with the use of centrifuges is discussed under precautions for laboratory equipment and devices in **GENERAL SAFETY PRACTICES**). Several well-documented studies have made it clear that great attention must be given to prevent contamination of room air with the suspension of liquid or solid particles containing hazardous materials including radioisotopes, infectious agents (viruses and mycoplasma from “normal” cells), as well as toxic chemicals and carcinogens.

The containment of aerosols and aerosol-generating processes is of prime importance. The hazard of an aerosol depends upon the concentration of the material in the suspension, the amount of energy imparted by the equipment creating the aerosol, the degree to which the suspending medium is protective of the material, the degree of danger associated with the material itself, and the susceptibility of the individual to danger from the agent.

Particle size is a factor in determining the path the aerosol will follow. Particles in the range of 1 to 5 microns present the greatest hazard to the laboratory worker since they more readily penetrate the respiratory tract than larger particles and are more readily retained than smaller or larger particles. Many laboratory procedures produce aerosols with particles in this range. Particles larger than 10 microns fall out on surfaces or are impinged on materials with an opposite electrostatic charge. In the respiratory tract, larger particles do not penetrate the lower spaces but are removed by interception and impaction in the upper respiratory tract and subsequently expelled or swallowed. Large droplets that fall out on surfaces dry quickly, and secondary aerosols of the dry particles can be created by air currents or laboratory activity. Significant settling of larger particles from an aerosol can occur in five minutes; however, most of the remaining small particles require 30 minutes to an hour to settle, if fresh currents of air do not prevent their settling. Therefore, it is best to wait before cleaning up a spill of infectious virus, etc. Besides the direct effects of aerosols, they may contaminate surfaces of the skin or equipment and subsequently enter the body because of hand-to-mouth contact and ingestion or through abrasions of the skin.

### 7.9.1.1 Aerosol minimization

In addition to avoiding the creation of an aerosol, three general approaches are recommended to decrease the hazards of aerosols associated with research on tumor specimens, cell and virus cultures and concentrates, and toxic chemical materials:

- Reduce the extent or concentration of the aerosol.
- Contain the aerosol in a primary barrier system.
- Use personal respiratory protection and protective laboratory clothing.

A summary from the National Cancer Institute<sup>7</sup> appears in the following table (Table 10: Summary of Aerosol-Generating Processes).

**Table 10: Summary of Aerosol-Generating Processes**

Potential Aerosol Creating Operation	Measures to Decrease Hazards from Aerosols
Forced expulsion of the last drop of liquid by alternatively sucking and blowing with the pipet, creating splashes and aerosols.	Use gravity flow of liquid with pipet calibrated for mark-to-mark drain-to-tip delivery and with pipet tip in contact with container wall.
Removing the cap or stopper from bottle after vigorous shaking to mix, wash or resuspend material.	Use swirling motion rather than shaking, allow aerosol to settle for a few minutes after bubbles disappear before removing cap or stopper.
Blending materials to disrupt cells, release enzymes or viruses, to homogenize suspensions, etc. without aerosol-tight cover seals or leak-proof rotor bearings.	Use special safety containers with seals to prevent escape of aerosols; use drain/siphon system to remove contents without removing cover.
Sonic disruption of cells or organelles	Use cup or chamber that is aerosol tight; allow aerosol to settle before opening cup. Place sonicator in fume hood or laminar flow cabinet.
Grinding tissue with mortar and pestle, glass tissue grinder, or ball mill.	Use slow spreads, use a clear plastic or inflatable glove bag to further contain the operation within the safety cabinet; allow aerosol to settle before removing cover.
Pouring hazardous materials from one container to another; e.g., decanting supernatants.	Use transfer pipets or closed siphon or vacuum technique.
Sterilizing a wire loop or needle in a flame, creating splatter.	Gradually dry loop or needle near flame, or use specially designed incinerator for loops or pipets.
Withdrawing a syringe needle as from a vaccine bottle or following inoculation of experimental animals	Use sterile cotton gauze to enclose needle; if experiment permits, use disinfectant with cotton or gauze.

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Weighing dry hazardous materials	Use draft-free, low-humidity enclosure for balance; discharge static electricity; use tared weighing containers not open weighing dishes or pipets.
Opening a freeze-dried preparation	If material is in an ampule, nick the ampule with a file, cover its neck with sterile gauze. If material is in a rubber stoppered bottle, first relieve vacuum with a hypodermic needle. If material is to be dissolved or suspended in liquid, introduce the liquid with a syringe and cover needle with gauze wetted with disinfectant.
Removing plugs from flasks and tubes.	Avoid wetting plug; remove plug slowly.
Handling cages that held infected animals or large animals in open areas or unventilated cages.	Avoid disturbing cage contents; if animals are held in open areas; use liquid disinfectants during cage cleaning; keep area clean; use personal respirator.

### 7.9.2 Biological Safety Cabinets

Biological safety cabinets (BSCs) are the primary means of containment for working safely with infectious microorganisms. BSCs are designed to provide personnel, environmental and product protection when appropriate practices and procedures are followed.

Biological safety cabinets are divided into three classes based upon the type of protection provided. The selection of a BSC is determined through a risk assessment.

**Table 11: Selection of a Safety Cabinet through Risk Assessment**

Biological Risk Assessed	Protection Provided			BSC Class
	Personnel	Product	Environmental	
<b>BSL 1 – 3</b>	Yes	No	Yes	I
<b>BSL 1 – 3</b>	Yes	Yes	Yes	II (A1, A2, B1, B2)
<b>BSL – 4</b>	Yes	Yes	Yes	III; II – When used in suit room with suit

Source: BMBL Appendix A

Class I and II cabinets use an air curtain and Class III uses a physical barrier to protect the investigator. Class II and III cabinets filter the air before it is blown onto the work surface, and all three cabinets have filtered exhaust. HEPA (high efficiency particulate air) filters are used since they are efficient in removing at least 99.97% of particles 0.3 microns in diameter. Because of the mechanics of particle filtration, particles of larger and smaller sizes are removed by HEPA filters with

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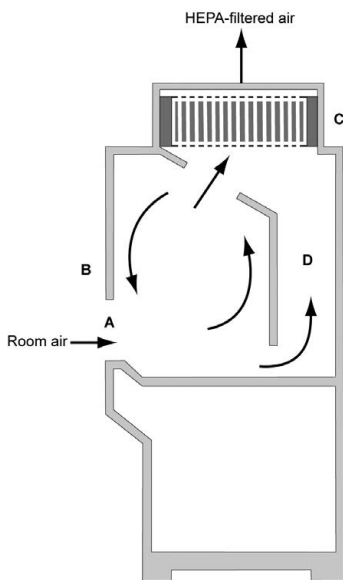
even greater efficiency and the efficiency increases slightly as the filter medium becomes loaded with contaminants. As the filter becomes loaded, the resistance to air movement through the filter increases, with the result that the rate of airflow now will decrease. Therefore, airflows must be adjusted periodically to assure proper performance. Also, these cabinets are subject to the same requirements with regard to location as fume hoods are (see **GENERAL SAFETY PRACTICES**). Annual certification of performance is required for these cabinets. Proper maintenance of cabinets used for work at all biosafety levels cannot be over emphasized since they are a primary containment device.

HEPA filters do not remove gaseous contaminants; instead, wet collectors or adsorptive systems are required, e.g., TEDA impregnated charcoal for radioiodine. The performance characteristics of these filters are not as well-defined as those of particulate filters, since their performance can be affected by ambient temperature, relative humidity, chemical concentration, flow rate, dwell time, chemical composition of the filtered air, and available capacity of the filter.

### 7.9.2.1 Class I Biological Safety Cabinet

The Class I BSC provides personnel and environment protection, but no product protection. The Class I cabinet is the simplest form of biological safety cabinet and consists of an enclosure with a front view panel and a full-width work opening. Room air, drawn into the cabinet through the work opening and into the back-wall baffle, prevents airborne contaminants inside the cabinet from escaping into the room, as in a fume hood. Unlike a fume hood, however, the exhaust is HEPA-filtered before entering the duct. Minimum face velocity for a Class I cabinet is 75 ft/min. Since unfiltered room air is drawn across the work area, the Class I cabinet does not protect experimental materials from ubiquitous airborne contamination.

Class I BSCs are used specifically to enclose equipment (e.g., centrifuges), or procedures with potential to generate aerosols (e.g., cage dumping, culture aeration or tissue homogenation).



**Figure 4: Class I BSC**

Optional modes of operation include a front closure panel with access ports which can be placed over the work opening thus reducing the amount of open area and raising the face velocity. Another option is to attach arm-length gloves to the access ports of the closure panel. In this mode, the cabinet serves as a glove box but does not provide containment equivalent to a Class III system.

Since the operator's hands and arms are not protected from contamination, control of contact contamination is dependent upon the use of gloves and other protective clothing. With the caveats cited for fume hoods, Class I cabinets accommodate many routine laboratory operations such as pipetting, blending, and sonicating. Because they lack a sterile work surface, they are not generally recommended. They do, however, provide personal protection during specific applications with low risk oncogenic viruses, and recombinant DNA at Biosafety

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Level 2 (BL2) containment level, as well as for chemical carcinogens, low-level radioactive materials and volatile solvents, provided:

- The face velocity is 100 ft/min.
- Concentrations of the materials being contained will not reach dangerous levels or contaminate the cabinet or associated exhaust system.
- Exhaust air is ducted to the outdoors.
- Quality of the effluent meets emission regulations.

**7.9.2.2 Class II Biological Safety Cabinets**

In the Class II cabinet, commonly known as laminar flow or biosafety hood, room air is drawn into the grille at the front edge of the work surface, passed through a HEPA filter, and recirculated into the cabinet work space through the overhead grille

Concurrently, the cabinet air is drawn from the work space through the grilles at the front and back edge of the work surface, and a portion of the air is exhausted after passing through a HEPA filter. An air barrier prevents airborne contaminants generated in the cabinet from escaping through the work opening. This air barrier is formed from the room air and downward flowing, HEPA-filtered air drawn into the front grille. The HEPA-filtered air flows downward with uniform velocity and minimum turbulence, minimizing lateral movement of aerosolized contamination within the cabinet and purging the work space.

Effective Class II biological safety cabinets have standards developed by the National Sanitation Foundation (NSF) and certified by the NIH. These cabinets may have fixed or variable vertical work openings. The Class II (Type A1, A2, B1, and B2) BSCs provide personnel, environmental, and product protection. Airflow is drawn into the front of the grille of the cabinet to provide personnel protection. The downward flow of HEPA-filtered air provides product protection by minimizing the change of cross-contamination across the work surface of the cabinet. Because cabinet exhaust air is passed through a certified HEPA filter, it is particulate-free for environmental protection. Type A1 and A2 BSCs the HEPA-filtered exhaust is recirculated to the laboratory or exhausted from the building via a canopy or “thimble” connection; Type B1 and B2 BSC must be discharged directly to the outdoors via a hard connection.

HEPA filters trap particulates and thus infectious agents, but do not capture volatile chemicals or gases. Only Type A2-exhausted or Type B1 and B2 BSCs that exhaust to the outside should be used when working with volatile, toxic chemicals, but amounts must be limited.

**Table 12: Comparison of Biosafety Cabinet characteristics**

BSC Class	Face Velocity	Airflow Pattern	Applications		
			Nonvolatile Toxic	Volatile Chemicals	Toxic and

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			Chemicals and Radionuclides	Radionuclides
<b>I</b>	75	In at front through HEPA to the outside or into the room through HEPA	Yes	When exhausted outdoors
<b>II, A1</b>	75	70% recirculated to the cabinet work area through HEPA; 30% balance can be exhausted through HEPA back into the room or to outside through a canopy unit	Yes (minute amounts)	No
<b>II, B1</b>	100	30% recirculated, 70% exhausted. Exhaust cabinet air must pass through a dedicated duct to the outside through a HEPA filter	Yes	Yes (minute amounts)
<b>I, B2</b>	100	No recirculation, total exhaust to the outside through a HEPA filter	Yes	Yes (small amounts)
<b>II, A2</b>	100	Similar to II, A1, but has 100 lfm intake air velocity and plenums are under negative pressure to room; exhaust air can be ducted to the outside through a canopy unit	Yes	When exhausted outdoors (FORMALLY "B3") (minute amounts)
<b>III</b>	N/A	Supply air is HEPA filtered. HEPA filters in series and is exhausted to the outside via a hard connection	Yes	Yes (small amounts)

All Class II cabinets are designed for work involving microorganisms assigned to biosafety levels 1, 2, 3, and 4. Class II BSCs provide the microbe-free work environment necessary for cell culture propagation and may be used for the formulation of non-volatile antineoplastic or chemotherapeutic drugs.

#### 7.9.2.2.1 Class II, Type A1 BSC

An internal fan draws sufficient room air through the front grille to maintain a minimum calculated or measured average inflow velocity of at least 75 lfm at the face opening of the cabinet. The supply air flows through a HEPA filter and provides particulate-free air to the work surface. Airflow provided in this manner reduces turbulence in the work zone and minimizes the potential for cross-contamination.

The downward moving air "splits" as it approaches the work surface; the fan draws part of the air to the front grille and the remainder to the rear grille. Although there are variations among different cabinets, this split generally occurs

about halfway between the front and rear grilles and two to six inches above the work surface.

The air is drawn through the front and rear grilles by a fan pushed into the space between the supply and exhaust filters. Due to the relative size of these two filters, approximately 30% of the air passes through the exhaust HEPA filter and 70% recirculates through the supply HEPA filter back into the work zone of the cabinet. Most Class II, Type A1 and A2 cabinets have dampers to modulate this division of airflow.

A Class II Type A1 BSC is not to be used for work involving volatile toxic chemicals. The buildup of chemical vapors in the cabinet (by recirculated air) and in the laboratory (from exhaust air) could create health and safety hazards.

It is possible to exhaust the air from a Type A1 or A2 cabinet outside of the building. However, it must be done in a manner that does not alter the balance of the cabinet exhaust system, thereby disturbing the internal cabinet airflow. The proper method of connecting a Type A1 or A2 cabinet to the building exhaust system is through use of a canopy hood,<sup>8,10</sup> which provides a small opening or air gap (usually 1 inch) around the cabinet exhaust filter housing. The airflow of the building exhaust must be sufficient to maintain the flow of room air into the gap between the canopy unit and the filter housing. The canopy must be removable or be designed to allow for operational testing of the cabinet. Class II Type A1 or A2 cabinets should never be hard-ducted to the building exhaust system<sup>8</sup>. Fluctuations in air volume and pressure that are common to all building exhaust systems sometimes make it difficult to match the airflow requirements of the cabinet.

#### **7.9.2.2.2 The Class II, Type B1 BSC:**

Some biomedical research requires the use of small quantities of hazardous chemicals, such as organic solvents or carcinogens. Carcinogens used in cell culture or microbial systems require both biological and chemical containment<sup>11</sup>.

The Class II, Type B cabinet originated with the National Cancer Institute (NCI)-designed Type 212 (later called Type B) BSC, and was designed for manipulations of minute quantities of hazardous chemicals with *in vitro* biological systems. The NSF International NSF/ANSI Standard 49—2007 definition of Type B1 cabinets<sup>8</sup> includes this classic NCI design Type B, and cabinets without supply HEPA filters located immediately below the work surface, and/or those with exhaust/recirculation down flow splits other than exactly 70/30%.

The cabinet supply blowers draw room air (plus a portion of the cabinet's recirculated air) through the front grille and through the supply HEPA filters located immediately below the work surface. This particulate-free air flows upward through a plenum at each side of the cabinet and then downward to the work area through a backpressure plate. In some cabinets, there is an additional supply HEPA filter to remove particulates that may be generated by the blower-motor system.

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Room air is drawn through the face opening of the cabinet at a minimum measured inflow velocity of 100 lfm. As with the Type A1 and A2 cabinets, there is a split in the down-flowing air stream just above the work surface. In the Type B1 cabinet, approximately 70 percent of the down flow air exits through the rear grille, passes through the exhaust HEPA filter, and is discharged from the building. The remaining 30 percent of the down flow air is drawn through the front grille. Since the air that flows to the rear grille is discharged into the exhaust system, activities that may generate hazardous chemical vapors or particulates should be conducted toward the rear of the cabinetwork area<sup>13</sup>.

Type B1 cabinets must be hard-ducted, preferably to a dedicated, independent exhaust system. As indicated earlier, fans for laboratory exhaust systems should be located at the terminal end of the ductwork to avoid pressuring the exhaust ducts. A failure in the building exhaust system may not be apparent to the user, as the supply blowers in the cabinet will continue to operate. A pressure-independent monitor and alarm should be installed to provide warning and shut off the BSC supply fan, should failure in exhaust airflow occur. Since this feature is not supplied by all cabinet manufacturers, it is prudent to install a sensor such as a flow monitor and alarm in the exhaust system as necessary. To maintain critical operations, laboratories using Type B1 BSCs should connect the exhaust blower to the emergency power supply.

#### **7.9.2.2.3 The Class II, Type B2 BSC:**

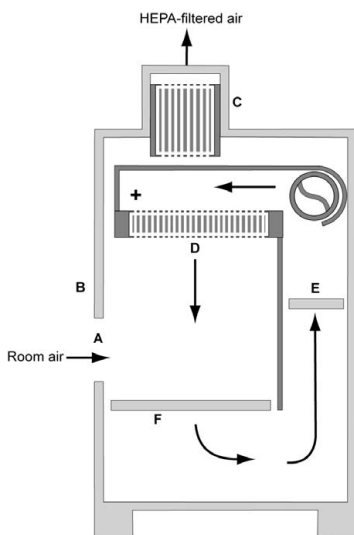
This BSC is a total-exhaust cabinet; no air is recirculated within it. This cabinet provides simultaneous primary biological and chemical (small quantity) containment. Consideration must be given to the chemicals used in BSCs as some chemicals can destroy the filter medium, housings and/or gaskets causing loss of containment. The supply blower draws either room or outside air in at the top of the cabinet, passes it through a HEPA filter and down into the work area of the cabinet. The building exhaust system draws air through both the rear and front grills, capturing the supply air plus the additional amount of room air needed to produce a minimum calculated or measured inflow face velocity of 100 lfm. All air entering this cabinet is exhausted, and passes through a HEPA filter (and perhaps some other air-cleaning device such as a carbon filter if required for the work being performed) prior to discharge to the outside. This cabinet exhausts as much as 1200 cubic feet per minute of conditioned room air making this cabinet expensive to operate. The higher static air pressure required to operate this cabinet also results in additional costs associated with heavier gauge ductwork and higher capacity exhaust fan. Therefore, the need for the Class II, Type B2 should be justified by the research to be conducted.

Should the building exhaust system fail, the cabinet will be pressurized, resulting in a flow of air from the work area back into the laboratory. Cabinets built since the early 1980's usually have an interlock system, installed by the manufacturer, to prevent the supply blower from operating whenever the exhaust flow is

insufficient; systems can be retrofitted if necessary. Exhaust air movement should be monitored by a pressure-independent device, such as a flow monitor.

#### 7.9.2.2.4 The Class II, Type A2 BSC (Formerly called A/B3):

Only when this BSC is ducted to the outdoors does it meet the requirements of the former Class II Type B3.<sup>8</sup> The Type A2 cabinet has a minimum calculated or measured inflow velocity of 100 lfm. All positive pressure contaminated plenums within the cabinet are surrounded by a negative air pressure plenum thus ensuring that any leakage from a contaminated plenum will be drawn into the cabinet and not released to the environment. Minute quantities of volatile toxic chemicals or radionuclides can be used in a Type A2 cabinet only if it exhausts to the outside via a properly functioning canopy connection.



**Figure 5: Model of a Class II, Type A2 BSC**

#### 7.9.2.2.5 Special Applications:

Class II BSCs can be modified to accommodate special tasks. For example, the front sash can be modified by the manufacturer to accommodate the eyepieces of a microscope. The work surface can be designed to accept a carboy, a centrifuge or other equipment that may require containment. A rigid plate with openings for the arms can be added if needed. Good cabinet design, microbiological aerosol tracer testing of the modification and appropriate certification are required to ensure that the basic systems operate properly after modification. Maximum containment potential is achieved only through strict adherence to proper practices and procedures.

#### 7.9.2.2.6 The Class III BSC

The Class III BSC was designed for work with highly infectious microbiological agents and for the conduct of hazardous operations and provides maximum protection for the environment and the worker. It is a gas-tight (no leak greater

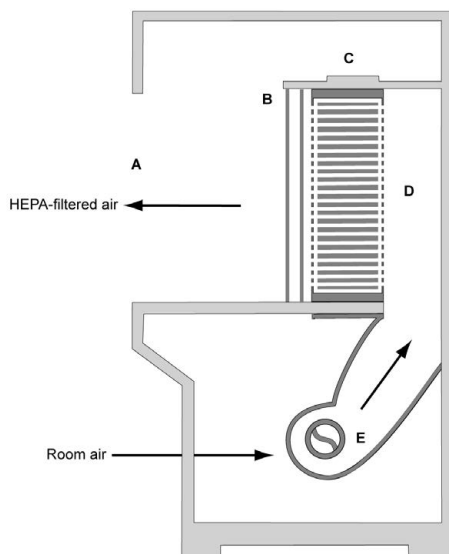
than  $1 \times 10^{-7}$  cc/sec with 1% test gas at 3 inches pressure Water Gauge<sup>14</sup>) enclosure with a non-opening view window. Access for passage of materials into the cabinet is through a dunk tank, that is accessible through the cabinet floor, or double-door pass-through box (e.g., an autoclave) that can be decontaminated between uses. Reversing that process allows materials to be removed from the Class III BSC safely. Both supply and exhaust air are HEPA filtered on a Class III cabinet. Exhaust air must pass through two HEPA filters, or a HEPA filter and an air incinerator, before discharge directly to the outdoors. Class III cabinets are not exhausted through the general laboratory exhaust system. Airflow is maintained by an exhaust system exterior to the cabinet, which keeps the cabinet under negative pressure (minimum of 0.5 inches of water gauge.)

Long, heavy-duty rubber gloves are attached in a gas-tight manner to ports in the cabinet to allow direct manipulation of the materials isolated inside. Although these gloves restrict movement, they prevent the user's direct contact with the hazardous materials. The trade-off is clearly on the side of maximizing personal safety. Depending on the design of the cabinet, the supply HEPA filter provides particulate-free, albeit somewhat turbulent, airflow within the work environment. Laminar airflow is not a characteristic of a Class III cabinet.

Several Class III BSCs can be joined together in a "line" to provide a larger work area. Such cabinet lines are custom-built; the equipment installed in the cabinet line (e.g., refrigerators, small elevators, shelves to hold small animal cage racks, microscopes, centrifuges, incubators) is generally custom-built as well.

#### **7.9.2.2.7 Horizontal Laminar Flow "Clean Bench"**

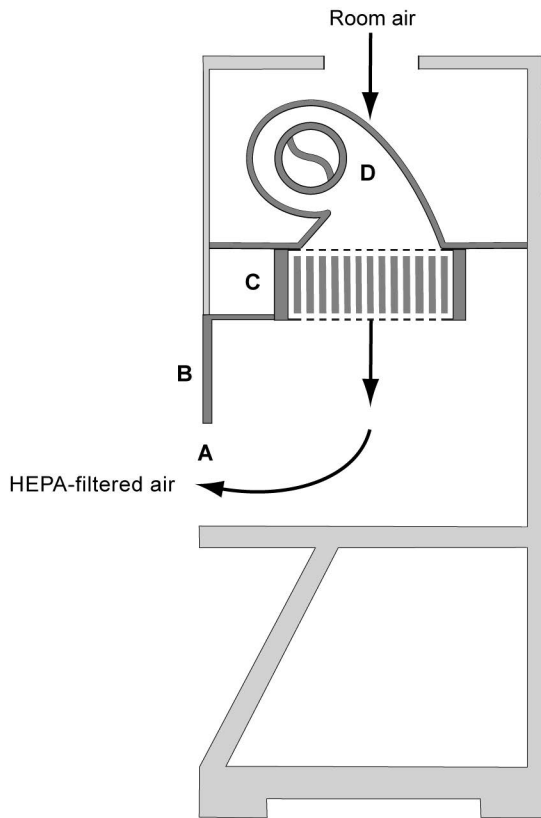
Horizontal laminar flow "clean benches" are not BSCs. These pieces of equipment discharge HEPA-filtered air from the back of the cabinet across the work surface and toward the user. These devices only provide product protection. They can be used for certain clean activities, such as the dust-free assembly of sterile equipment or electronic devices. Clean benches should never be used when handling cell culture materials, drug formulations, potentially infectious materials, or any other potentially hazardous materials. The worker will be exposed to the materials being manipulated on the clean bench potentially resulting in hypersensitivity, toxicity or infection depending on the materials being handled. Horizontal airflow "clean benches" must never be used as a substitute for a biological safety cabinet. Users must be aware of the differences between these two devices.



**Figure 6: Horizontal Laminar Flow "Clean Bench"**

#### 7.9.2.2.8 Vertical Flow "Clean Bench"

Vertical flow clean benches also are not BSCs. They may be useful, for example, in hospital pharmacies when a clean area is needed for preparation of intravenous solutions. While these units generally have a sash, the air is usually discharged into the room under the sash, resulting in the same potential problems presented by the horizontal laminar flow clean benches. These benches should never be used for the manipulation of potentially infectious or toxic materials or for preparation of antineoplastic agents.



**Figure 7: Vertical Laminar Flow "Clean Bench"**

Remember that Class II cabinets are not absolute containment devices. Performance evaluation tests include using a nebulizer to introduce a known concentration of bacterial spore suspension at various locations inside and outside the cabinet, then scoring growth on agar plates exposed directly or collected in impingers. A protection factor is calculated from the number of spores collected outside the BSC at the face during release of a known aerosol inside the cabinet. The minimum requirement for personal protection is 105, i.e., 105 fewer spores are collected at the cabinet face than near an aerosol generated on an open bench. This protection factor, measured in a static test under ideal conditions, is not usually achieved in routine laboratory use.

The air barrier can be disturbed by an imbalance of airflows that may be caused by turbulent ventilation sources or heavy traffic, inadequate clearance above exhaust filters, mechanical failure, dirty filters, or blockage of the air-intake grilles that extend along the front and back of the work surface. The uniform, downward flow of clean air over the work surface can be disturbed by placing items on the front or rear grilles, by overcrowding the cabinet interior, by convection currents from heat sources, or rapid hand motions in and out of the cabinet. Class II cabinets are suitable for most projects, are convenient to use, and offer adequate personnel and product protection if used properly with low to moderate-risk oncogenic viruses, CDC classes 1 to 3 etiologic agents, and recombinant DNA

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materials requiring BL2 containment. Since Class II cabinets may recirculate a large fraction of the air flowing through them, they are only suitable for work with dilute concentrations of radioactive materials, toxic chemicals, or carcinogens of low volatility, provided:

### **7.9.2.3 Biological Safety Cabinet Operations**

Good work practices are necessary to prevent compromising the protection offered by a biological safety cabinet (BSC). BSCs should be certified at least annually. All routinely used items should be left in place during the certification evaluation to reveal potential disturbances. BSCs should be left running at all times with the sashes at the eight or ten-inch height. Do not override the sash alarms. By leaving the BSC running, release of cabinet air is avoided, the work area remains clean, and spills, particularly below the work surface dry rapidly reducing the potential for contaminant growth. Disruptions of the air curtain should be limited. This requires keeping front and rear grilles uncluttered and minimizing movement of hands into and from the BSC. The latter necessitates internal waste collection. The use of loose bags or mesh support stands for waste bags is discouraged because the bags can be punctured and leak when handled. Waste can be collected inside the BSC in a one cubic foot stainless steel receptacle fitted with an autoclavable biohazard bag, such that the inside of the upper portion of the bag is folded down over the outside of the container. The disturbance in the cabinet air flow induced by such a receptacle is less than the many potential disturbances caused by external waste collection. When possible use disposal pipettes. Use of non-disposable pipettes necessitates their collection in awkward liquid-filled trays inside the BSC or buckets outside the cabinet. The laminar air flow and drying atmosphere within the BSC reduces the likelihood of contamination from residues in the disposable pipettes. Use a small footprint vacuum trap bottle and hydrophobic filter instead of a vacuum flash to limit the total volume of materials in the BSC. Some users remove all items before cleaning the BSC and reintroduce the items as work recommences. Often proper decontamination of the materials is difficult and poorly performed; consequently, contaminants may be spread. It is better to keep a few dedicated items in the BSC, e.g., forceps and pipetting aids, and routinely decontaminating these items and the work surface with 70% ethanol before initiation and upon completion of work. Porous materials such as wipes or gauze sponges cannot be readily decontaminated. Once placed in the cabinet, they should not be removed for routine cleaning.

Use of a flame in aseptic technique is historic, and the misconception persists that flaming enhances the 'sterility' provided by a BSC. Cabinet manufacturers, cell culture manuals, and the British Standards Institution advise against use of a burner. The upward air currents induced by the flame run counter to the clean, downward airflow. The resulting turbulence may contribute to the spread of contamination within the BSC. Plastic culture ware does not retain its integrity when flamed to sterility; so, most users merely pass items quickly through the flame. The combination of a flame and flammable ethanol vapors or combustibles

in the BSC has resulted in damaged filters and serious fires, fanned by the cabinet airflow. Consequently, burners should not be used in Class II BSCs. A gas line connection to a BSC should be permitted for only very special procedures and not for routine culture work.

Early users of microbiological cabinets needed germicidal, ultraviolet (UV) lights to improve sterility. Today some investigators use the UV light in lieu of frequent thorough cleaning with a general purpose germicidal solution, such as 70% ethanol. UV radiation is only surface effective and will not penetrate through items on the work surface or dust particles attracted to the bulb by static electricity. Intensity decreases with the square of the distance and diminishes rapidly with tube age. While UV lamps are of minimal effectiveness in improving sterility, they do present a risk to eyes and UV sensitive skin.

#### **7.9.2.4 Guidelines for Operations in a Biological Cabinet**

1. The cabinet should be left running.
2. If adjustable, the window should be lowered to 8 inches, with a 100 ft/min face velocity.
3. Keep the amount of equipment used or stored in the cabinet to a minimum.
4. Before work is started, everything needed for the procedures should be placed in the cabinet, and the air allowed to exhaust for a few minutes.
5. Nothing should be placed on or blocking the front or rear grilles.
6. Contaminated items should be segregated from clean ones and located so that they never have to be passed over clean items.
7. Avoid disrupting the air barrier in a safety cabinet by frequent and rapid arm movements and bringing the hands in and out of the cabinet.
8. Waste containers should be placed inside the cabinet to avoid breaking the air barrier and bringing contaminated items out into the room.
9. Do not use a burner (even a Touch-Omatic™ type) in a Class II biological safety cabinet because the air currents induced are counter to the normal air flow, can cause contamination of the work surface or the room, ignite ethanol and other materials in the cabinet, and damage the HEPA filter. Note: plastic ware cannot withstand sterilizing temperatures.
10. Do not use a cabinet ultraviolet (UV) lamp. It only provides a minimal initial surface germicidal effect, which deteriorates rapidly with time, distance, and dust deposits, while the ocular and skin hazards from the UV light persist.
11. When working with biohazards, keep absorbent towels and decontaminating solutions, usually 70% ethanol and 10% chlorine bleach, in the cabinet and wipe down the work surface with ethanol prior to and at completion of each session, and after any small spills. Decontaminate all equipment removed from the cabinet. Pipetting aids and tools that are used repeatedly should remain in the cabinet. Inspect, decontaminate, or change receivers on pipet aids regularly. Decontamination of the entire cabinet (the filters, the plenums, the work surfaces and the fan) is achieved by exposing

- these areas to paraformaldehyde vapor. This type of decontamination must be performed only by a certified professional.
12. A liquid trap bottle with bleach or other suitable disinfectant should be kept inside the cabinet and a small two-micron, hydrophobic filter should be placed between the trap and the vacuum spigot to protect the vacuum line.
  13. Do not use a vertical or horizontal laminar flow cabinet (blow out hood) for work with biological materials.

#### **7.9.2.5 Certification and Decontamination Requirements**

Newly installed biological safety cabinets frequently fail to meet design criteria and many cabinets fail to pass routine leak tests. The performance of every safety cabinet should be tested and certified as meeting specifications after it has been purchased and installed, but before it is used, after it has been moved or serviced, and at least annually. Decontamination is required prior to moving or servicing. Do not ask maintenance personnel to service these cabinets. Certification, decontamination and service must be performed by a trained professional according to NSF Standard 49.

#### **7.9.2.6 Electricity Failure During Use of a Biological Safety Cabinet**

Should the power to the unit fail during use, stop work with biohazardous agents immediately, seal all cultures securely, and decontaminate the work area with a suitable disinfectant.

#### **7.9.3 Biological Stains**

Fixatives and stains used for the preparation of tissues and cellular materials often have toxic properties, e.g., methylene blue, trypan blue (teratogen), requiring the use of impermeable gloves and appropriate ventilation. In addition, several dyes used in conjunction with flow cytometry and visualization of nucleic acids are suspect carcinogens. Be sure the precautions you are taking are adequate. If in doubt, consult with your supervisor or the Department of Environmental Health and Safety.

#### **7.9.4 Incubators**

Incubators can become the inadvertent and undesired repository of microorganisms. Although they may present a hazard to laboratory workers, most often they are a source of contamination of laboratory cultures. Besides the moist surfaces, rubber gaskets, the humidity trough (if present) and fan mechanism are areas in which contaminating microorganisms concentrate. It is recommended that an anti-microbial agent, such as Zepharin Chloride™ be added to the humidity source water; do not use sodium azide. Sodium azide is explosive when heated and is extremely toxic. In addition, the inner panels, trays, and the other removable parts should be autoclaved and the gaskets and non-removable parts wiped thoroughly with 70% ethanol every two months.

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### 7.9.5 Freezer and Liquid Nitrogen Storage

Freezers containing potentially hazardous biological materials and toxins should be labeled accordingly. These freezers should be defrosted at least annually to prevent the accumulation of broken vials and excessive frost. Note that “frost-free” freezers allow small samples to thaw during warming cycles.

Ethanol should not be kept in freezers that are not designed for flammable storage. The use of such storage for nucleic acid precipitation appears to be contraindicated. It has been reported that centrifugation time and DNA concentration are more significant than incubation temperature for efficient recovery of DNA<sup>8</sup>. A ten-minute incubation at 0°C after addition of room temperature ethanol is more efficient than incubation at -20°C or -70°C.

Cells and virus stocks should be stored in sealed ampules and not in screw cap glass vials. Screw cap glass vials are permeable to the liquid nitrogen (approximately 50% of the time) and therefore represent a source of contamination in the storage tank.

Plastic screw cap ampules also leak and must be used with a heat-sealed sleeve to prevent contamination of the liquid nitrogen and other samples. Upon thawing, sealed vials may explode, producing an aerosol of glass and cell debris.

If freezing manually, place ampules in the bottom of a beaker, cover with methanol and a dye, e.g., methylene blue, and transfer the entire beaker from refrigerator to freezer. The methanol provides even freezing and the dye will penetrate imperfectly sealed vials permitting their identification and elimination.

When adding samples to liquid nitrogen storage repositories, be aware that the liquefied nitrogen may boil vigorously as warmer materials are added. Use only in a well-ventilated area. Liquefied nitrogen is a cryogenic gas and expands 700-fold upon vaporization; this may result in a rapid displacement of air (see **CHEMICAL AND COMPRESSED GAS SAFETY** for more information on gases).

When thawing cells, a lab coat, face guard, thermal gloves, and closed shoes should be worn. Ampules to be thawed should be dropped into a plastic beaker containing 70% ethanol at 37°C within a Styrofoam bucket and covered immediately. Confirm the identification of the sample. Open the vial in a biological safety cabinet, by nicking the ampule with a file near the neck. Wrap it in ethanol wetted material and, holding the vial upright, snap the ampule open at the nick. Add liquid slowly to dried material. Withdraw the suspension and mix in another vessel.

## **7.9.6 Spills and Decontamination**

### **7.9.6.1 Spills**

#### **7.9.6.1.1 Small or Incidental spills**

Most spills in the laboratory involve comparatively small quantities of chemicals and biohazards which can readily be cleaned up by laboratory personnel. Notify the laboratory supervisor. The spill control procedures may be performed under his supervision. Arrange for disposal of the chemicals and clean up materials with Environmental Health and Safety.

#### **7.9.6.1.2 Spills requiring assistance**

If the spill involves hazardous material(s) (i.e., radioactive, toxic, flammable, corrosive, volatile, reactive or infectious materials) additional assistance or equipment is required. Contact Environmental Health and Safety (ext. 2215); after hours, dial Wright State Police Department's Dispatch number, etc. 2111. Provide the following information:

- Name of person calling.
- Type of spill, name of material spilled and approximate quantity.
- Location: building, floor and room number.

#### **7.9.6.2 Contained spill of biological materials**

The following guide is to be followed in the event of a small contained spill of biological materials and/or until assistance from the Environmental Health and Safety office is obtained.

##### **7.9.6.2.1 Dry/Non-volatile biological materials**

If the substance is dry and/or nonvolatile, shut off hoods, close windows and doors, and vacate rooms. Label door with appropriate warning. Allow the aerosol to settle for about 30 minutes before reentering room.

##### **7.9.6.2.2 Volatile biological materials**

If the substance is volatile, leave the ventilation on and vacate room, closing door. Label the door with a warning.

- Notify your laboratory supervisor and the Environmental Health and Safety office.
- For a liquid biological spill, use absorbent pads to soak up the liquid and to act as a vapor barrier. Work from the perimeter inward.
- If an infectious agent or particulate agent is involved, close all windows and call Physical Plant at ext. 4444 (between 7:00 am and 3:30 pm, at all other times call Wright State Police Department at ext. 2111) to have them turn

off the air handling units in the building. Be sure to shut off all the fume hoods in the room of the spill. (Wait 30 minutes for the aerosol to settle before reentering the room).

If the spill occurs in public or common areas, you must notify Wright State Police Department (ext. 2111) and Environmental Health and Safety (ext. 2215) immediately.

In all cases immediately alert neighbors, laboratory supervisor; and/or department chair.

### **7.9.6.3 Decontamination**

#### **7.9.6.3.1 Liquid cultures**

All culture materials and biological specimens, including that from “normal” cultures and primary tissue, should be collected inside the biological safety cabinet.

- These materials should be chemically inactivated on at least a daily schedule
- Most liquid cultures may be chemically inactivated with freshly prepared bleach (15% v/v). The bleach solution must sit for at least 20 minutes prior to drain disposal, followed by copious amounts of water.
- Do not leave untreated waste in an egress corridor or public area.

#### **7.9.6.3.2 Autoclaving biological materials**

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*Autoclaving infectious biological materials for disposal in regular trash is not approved for Wright State University.*

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#### **7.9.6.3.2.1 Autoclave guidelines**

- Biological materials for autoclaving should be placed in autoclavable bags, and the name of the generator should be clearly marked on the bag.
- The bags should be no more than two-thirds full and tied or taped closed.
- To prevent piercing the bags, place all sharp objects in puncture-proof containers.
- Up to a liter of either absorbed or contained liquid, i.e., on culture ware, may be and should be (add at least 500 ml, if necessary) placed in each bag.

- Materials should be autoclaved for 60 minutes at 121°C and 15 PSI.
  - Autoclave in a shallow plastic tray or other vessel suitable to contain possible leakage from the bag.
- Be sure to verify that the designated temperature was reached and maintained.
- Include spore strips routinely and ampules of *Bacillus stearothermophilus* monthly in waste bags to monitor autoclave performance in various locations of the autoclave.
- Wear loose fitting thermal gloves; remove immediately if they get wet.
- Do not remove liquids immediately following cycle as they may be superheated and boil vigorously.

Note that dry heat is much less effective than moist heat for sterilization and is not appropriate for waste treatment.

For example, a dry heat oven set at 165°C requires 5-6 hours to effectively sterilize glassware that can be sterilized by autoclaving at 121°C in 20 minutes. Hot air is a less effective heat conductor than steam; in addition, the dry oven usually requires a much longer time to reach temperature.

#### 7.9.6.3.2.2 Autoclave precautions

- Hypochlorites or any other strong oxidizing material must not be autoclaved with organic material such as paper, cloth, oils, or volatile solvents as this may produce toxic vapors or an explosion! Therefore, do not autoclave materials that have been treated with chlorine bleach.
- Do not autoclave materials contaminated with radioisotopes and/or toxic chemicals. These materials may volatilize and contaminate the autoclave and expose workers.

#### 7.9.6.4 Biological safety cabinet decontamination

The biological safety cabinet should be wiped down with an appropriate disinfectant (see *Disinfectants* below) prior to and at the initiation of each session.

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*Wastes that are biological, chemical and radioactive, or a combination of the above, should be inactivated first with regard to their pathogenicity and then toxicity, but ultimately must be disposed of as radioactive waste. Refer to Wright State's RADIATION SAFETY MANUAL.*

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## 7.9.7 Disinfectants

### 7.9.7.1 Alcohol

Isopropyl and ethyl alcohols in 70-90% concentrations may be germicidal against lipid-containing agents but are not effective against spores and infectious DNA. Note that 100% ethanol is not a good disinfectant. The major advantages of alcohols are that they are fast acting, evaporate rapidly, and leave no residue. Moreover, they can be combined with other disinfectants (quaternaries, phenolics, and iodine) to form tinctures further enhancing lethal action.

### 7.9.7.2 Chlorine

A very active disinfectant, chlorine is lethal against a wide variety of gram-negative and gram-positive bacteria, bacterial spores and most viruses. Disinfect media with a 10% solution of chlorine bleach (5.25% hypochlorite or 52,500 ppm) for 15 to 30 minutes. Note that solutions deteriorate with age and are rapidly neutralized by organic matter. Its effectiveness may be enhanced by the addition of 0.1% solution of an ionic detergent. If used directly on a stainless-steel surface, rinse thoroughly with water to prevent tarnishing and decomposition.

### 7.9.7.3 Iodophor

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*Do not autoclave chlorine solutions.*

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Characteristics of chlorine and iodine are similar. Iodophors are effective against gram-positive and

gram-negative organisms, mycobacteria, and some viruses, and are most effective in acid solutions. Organic matter reduces effectiveness, but iodophors are less affected than hypochlorites. Do not autoclave since iodophores vaporize at 120°F. Stable in storage if kept cool and tightly covered.

### 7.9.7.4 Ethylene Oxide

Ethylene oxide, due to its acute toxicity (skin, eye, respiratory and mucous membrane irritation, vomiting, and diarrhea), chronic toxicity (respiratory irritation, secondary respiratory infection, anemia), and status as a suspected carcinogen and mutagen, should be used for decontamination only when no other agent or method is effective. Ethylene oxide sterilizers are commonly used for decontamination and sterilization of heat-sensitive or moisture-sensitive complex apparatus and machines.

- In the event of an ethylene oxide leak, evacuate the area, and call the emergency contact number.
- Avoid all skin contact with ethylene oxide.
- Splashes of liquid ethylene oxide or a solution of ethylene oxide should be treated immediately by removing any contaminated clothing and flushing

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the affected areas with copious amounts of water. Contaminated clothing, especially leather items such as shoes, must be bagged and aerated for at least 8-12 hours and then thoroughly laundered before reuse.

- If inhalation occurs, leave the area immediately and move into an area with fresh air. Contact Environmental Health and Safety. If overexposure symptoms develop (vomiting or nausea) contact a physician. Symptoms may not develop until up to 6 hours after the exposure.
- When working with liquid ethylene oxide, its solution or the gas cylinders, wear heavy butyl or nitrile gloves, and goggles or a face shield. Other garments, e.g., sleeves, lab coats, should be made of polyethylene-coated disposable materials, e.g., Tyvek™.
- The room should have adequate ventilation, and the sterilizer should have dedicated ventilation.
- Items must be thoroughly cleaned before treatment with ethylene oxide. Residual organic matter or debris protects microorganisms from exposure to the gas and the residual materials (e.g., proteins, salts, solutions) may contaminate the sterilizer and the aerator.
- The sterilizer equipment and room must be monitored to ensure that exposure limits are below OSHA Permissible Exposure Limits (PELs).

Any area where exposure to ethylene oxide may exceed the PEL must be designated a regulated area and access restricted to authorized personnel. The area must be posted:

*DANGER – ETHYLENE OXIDE  
CANCER HAZARD AND REPRODUCTIVE HAZARD  
AUTHORIZED PERSONNEL ONLY  
RESPIRATOR AND PROTECTIVE CLOTHING MAY BE  
REQUIRED TO WORK IN THIS AREA*

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*Contact the Department of Environmental Health and Safety for  
information on emergency procedures, training, environmental  
monitoring.*

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## **7.10 Infectious Waste Management**

Keeping biological waste separate from other waste streams is essential for any management program. Disposal of infectious (medical) waste, subject to federal, state and local laws, is becoming increasingly more regulated and costly. All biological waste that fits the infectious waste definition found in Wright State

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University's **Exposure Control Plan** must be disposed of by following the procedures in that manual.

Wright State University transports infectious waste to commercial treatment and disposal facility as opposed to on-site treatment of infectious waste via autoclaving, incineration, and in some cases chemical treatment. Wright State individual generators of infectious waste not permitted to autoclave or incinerate infectious waste and dispose of it as ordinary trash. Autoclaves can be used for disinfection and sterilization purposes (i.e., for glassware, equipment) and for the treatment of all other waste not meeting the Ohio Environmental Protection Agency's (OEPA) definition of infectious waste. Waste that does not meet the OEPA definition of infectious waste but requires autoclave treatment by another agency shall be autoclaved in bags not labeled with the international biohazard symbol.

On-site chemical treatment of infectious waste cultures is permitted and required. Untreated liquid or semi-liquid infectious waste consisting of blood, blood products, body fluids, and excreta may be disposed of into the sanitary sewer system without prior treatment.

Follow all the procedures found in Wright State University's **Exposure Control Plan**. For further clarification describing the difference between biological and infectious waste, please call the Environmental Health and Safety Department.

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### 7.13 Appendix A:

Biosafety Level	Agent Characteristics	Practices	Safety Equipment	Facilities (secondary barriers)
BSL-1	Not known to consistently cause disease in health adults	Standard microbiological practices	None	Open bench top sink
BSL-2	Associated with human disease, hazard from percutaneous injury, ingestion, mucous membrane exposure	Standard microbiological practices Limited access Biohazard warning signs Sharps precautions Biosafety manual defining any needed waste decontamination or medical surveillance policies.	Class I or II biosafety cabinets (BSCs) or other containment devices used for all agents that cause splashes or aerosols of infectious materials Laboratory coats and gloves Face protection as needed	Open bench top sink Autoclave
BSL-2	Indigenous or exotic agents with potential for aerosol transmission; disease may have serious or lethal consequences	All BSL-2 practices Controlled access Decontamination of all waste Decontamination of laboratory clothing before laundering Baseline serum	Class I or II BSCs or other physical containment devices used for all open manipulations of agents Protective lab clothing and gloves Respiratory protection as needed	Open bench top sink Autoclave Physical separation from access corridors Self-closing, double-door access Exhaust air not recirculated Negative airflow in laboratory
BSL-4	Dangerous/exotic agents which pose high risk of life-threatening disease; aerosol-	All BSL-3 practices Clothing change before entering Shower on exit All material	All procedures conducted in Class III BSCs, or Class I or II BSCs in	BSL-3 plus: Separate building or isolated zone Dedicated supply and exhaust,

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	transmitted lab infections; or related agents with unknown risk of transmission	decontaminated on exit from facility	combination with full-body, air-supplied, positive pressure personnel suit.	vacuum, and decontamination systems Other requirements outline the text
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## **8 Physical Hazards and Others**

Besides exposure to chemicals and biological agents, laboratory workers can also be exposed to a number of physical hazards.

Some of the common physical hazards that they may encounter include the following:

- ergonomic,
- ionizing radiation,
- nonionizing radiation, and
- noise hazards.

These hazards are described below in individual sections.

### **8.1 Ergonomic Hazards**

Laboratory workers are at risk for repetitive motion injuries during routine laboratory procedures such as pipetting, working at microscopes, operating microtomes, using cell counters and keyboarding at computer workstations. Repetitive motion injuries develop over time and occur when muscles and joints are stressed, tendons are inflamed, nerves are pinched and the flow of blood is restricted. Standing and working in awkward positions in front of laboratory hoods/biological safety cabinets can also present ergonomic problems. By becoming familiar with how to control laboratory ergonomics-related risk factors, chances for occupational injuries can be reduced while improving worker comfort, productivity, and job satisfaction. In addition to the general ergonomic guidance, simple adjustments can be made at the workplace. While there is currently no specific OSHA standard relating to ergonomics in the laboratory workplace, information to laboratory workers is available in the OSHA fact sheet Laboratory Safety – Ergonomics for the Prevention of Musculoskeletal Disorders in Laboratories.

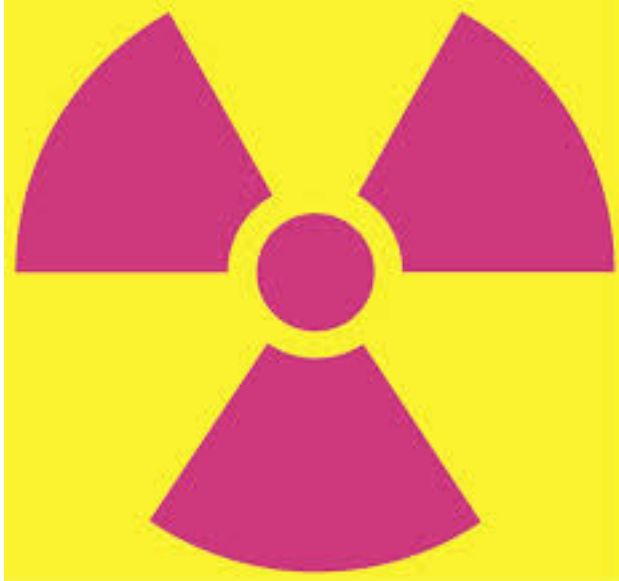
### **8.2 Ionizing Radiation**

OSHA's Ionizing Radiation standard, 29 CFR 1910.1096, sets forth the limitations on exposure to radiation from atomic particles. Ionizing radiation sources are found in a wide range of occupational settings, including laboratories on campus. These radiation sources can pose a considerable health risk to affected workers if not properly controlled.

Any laboratory possessing or using radioactive isotopes must be licensed by the Nuclear Regulatory Commission (NRC) and/or by a state agency that has been approved by the NRC, 10 CFR 31.11 and 10 CFR 35.12. The fundamental objectives of radiation protection measures are:

- to limit entry of radionuclides into the human body (via ingestion, inhalation, absorption, or through open wounds) to quantities as low as reasonably achievable (ALARA) and always within the established limits; and
- to limit exposure to external radiation to levels that are within established dose limits and as far below these limits as is reasonably achievable.

All areas in which radioactive materials are used or stored must conspicuously display the symbol for radiation hazards and access should be restricted to authorized personnel.



**Figure 8: Radiation Symbol**

The OSHA Ionizing Radiation standard requires precautionary measures and personnel monitoring for workers who are likely to be exposed to radiation hazards. Personnel monitoring devices (film badges, thermoluminescent dosimeters (TLD), pocket dosimeters, etc.) must be supplied and used if required to measure an individual's radiation exposure from gamma, neutron, energetic beta, and X-ray sources. The standard monitoring device is a clip-on badge or ring badge bearing the individual assignee's name, date of the monitoring period and a unique identification number. The badges are provided, processed and reported through a commercial service company that meets current requirements of the National Institute of Standards and Technology's National Voluntary Laboratory Accreditation Program (NIST NVLAP).

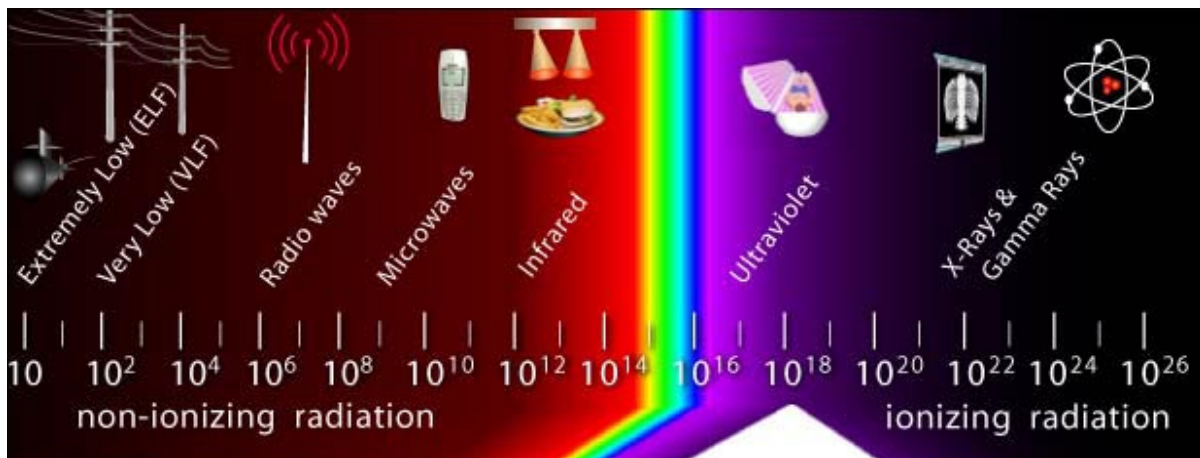
Wright State follows all applicable regulations for the use of isotopes. Wright State ensures compliance with local, state, and federal laws and regulations; maintains licenses for official use of radioactive substances; designates a radiation safety officer (RSO) to oversee and ensure compliance with state and/or NRC requirements. Information on radioactive materials licenses may be obtained from the Ohio Department of Health and/or from the NRC.

The following OSHA Safety and Health Topics Page provides links to technical and regulatory information on the control of occupational hazards from ionizing radiation: [www.osha.gov/SLTC/radiationionizing/index.html](http://www.osha.gov/SLTC/radiationionizing/index.html).

### 8.3 Non-ionizing Radiation

Non-ionizing radiation is described as a series of energy waves composed of oscillating electric and magnetic fields traveling at the speed of light. Nonionizing radiation includes the spectrum of ultraviolet (UV), visible light, infrared (IR), microwave (MW), audio frequency (RF), and extremely low frequency (ELF). Lasers commonly operate in the UV, visible, and IR frequencies. Non-ionizing radiation is found in a wide range of occupational settings and can pose a considerable health risk to potentially exposed workers if not properly controlled.

The following OSHA Safety and Health Topics Pages provide links to technical and regulatory information on the control of occupational hazards from nonionizing radiation and are available at: [www.osha.gov/SLTC/radiation\\_nonionizing/index.html](http://www.osha.gov/SLTC/radiation_nonionizing/index.html).



**Figure 9: Electromagnetic Spectrum** (<https://www.osha.gov/SLTC/radiation/>)

#### 8.3.1 Extremely Low Frequency Radiation (ELF)

Extremely Low Frequency (ELF) radiation at 60 HZ is produced by power lines, electrical wiring, and electrical equipment. Common sources of intense exposure include ELF induction furnaces and high-voltage power lines.

#### 8.3.2 Radiofrequency and Microwave Radiation

Microwave radiation (MW) is absorbed near the skin, while radiofrequency (RF) radiation may be absorbed throughout the body. At high enough intensities, both will damage tissue through heating. Sources of RF and MW radiation include radio emitters and cell phones.

#### 8.3.3 Infrared Radiation (IR)

The skin and eyes absorb infrared radiation (IR) as heat. Workers normally notice excessive exposure through heat sensation and pain. Sources of IR radiation include heat lamps and IR lasers.

#### **8.3.4 Visible Light Radiation**

The different visible frequencies of the electromagnetic (EM) spectrum are "seen" by our eyes as different colors. Good lighting is conducive to increased production, and may help prevent incidents related to poor lighting conditions. Excessive visible radiation can damage the eyes and skin.

#### **8.3.5 Ultraviolet Radiation (UV)**

Ultraviolet radiation (UV) has a high photon energy range and is particularly hazardous because there are usually no immediate symptoms of excessive exposure. Sources of UV radiation in the laboratory include black lights and UV lasers.

#### **8.3.6 Laser Hazards**

Lasers typically emit optical (UV, visible light, IR) radiations and are primarily an eye and skin hazard. Common lasers include CO<sub>2</sub>IR laser; helium - neon, neodymium YAG, and ruby visible lasers, and the Nitrogen UV laser.

LASER is an acronym which stands for Light Amplification by Stimulated Emission of Radiation. The laser produces an intense, highly directional beam of light. The most common cause of laser induced tissue damage is thermal in nature, where the tissue proteins are denatured due to the temperature rise following absorption of laser energy.

The human body is vulnerable to the output of certain lasers, and under certain circumstances, exposure can result in damage to the eye and skin. Research relating to injury thresholds of the eye and skin has been carried out to understand the biological hazards of laser radiation. It is now widely accepted that the human eye is almost always more vulnerable to injury than human skin.

### **8.4 Noise**

OSHA's Occupational Noise Exposure standard, 29 CFR 1910.95, requires employers to develop and implement a hearing conservation program that includes the use of PPE (e.g., hearing protectors), if workers are exposed to a time-weighted average (TWA) of  $\geq 85$  dBA over an 8-hour work shift. In addition, when workers are exposed to noise levels  $\geq 85$  dBA, the employer must develop a monitoring program to assess noise levels. The monitoring program must include the following components:

- All continuous, intermittent, and impulsive sound levels from 80-130 dBA must be included in noise measurements, 29 CFR 1910.95(d)(2)(i);
- Instruments used to measure worker noise exposure must be calibrated to ensure measurement accuracy, 29 CFR 1910.95(d)(2)(ii); and
- Monitoring must be repeated whenever a change in production, process, equipment, or controls increases noise exposures, 29 CFR 1910.95(d)(3).

Laboratory workers are exposed to noise from a variety of sources. Operation of large analyzers (e.g., chemistry analyzer), fume hoods, biological safety cabinets, incubators, centrifuges (especially ultracentrifuges), cell washers, sonicators, and stirrer motors, all contribute to the noise level in laboratories. Further sources of noise in laboratories include fans and compressors for cryostats, refrigerators, refrigerated centrifuges, and freezers. As an example, a high-speed refrigerated centrifuge alone can generate noise levels as high as 65 dBA. To provide some further context, a whisper registers approximately 30 dBA; normal conversation about 50 to 60 dBA; a ringing phone 80 dBA and a power mower 90 dBA. If noise levels exceed 80 dBA, people must speak very loudly to be heard, while at noise levels of 85 to 90 dBA, people must shout.

To determine if the noise levels in the laboratory are above the threshold level that damages hearing, a noise exposure assessment must be conducted using an approved sound level monitoring device, such as a dosimeter, and measuring an 8-hour TWA exposure. If the noise levels are found to exceed the threshold level, the employer must provide hearing protection at no cost to the workers and train them in the proper use of the protectors.

The potential dangers of miscommunicating instructions or laboratory results are obvious, and efforts should be made to improve the design of clinical laboratories and to evaluate new instrumentation about the impact of these factors on worker noise exposure. It is possible to relocate equipment to another area or use engineering controls to reduce the noise level below an 8-hour TWA of 85 dBA to comply with OSHA's Occupational Noise Exposure standard.

While most laboratories' noise levels do not equal or exceed the 8-hour TWA of 85 dBA, certain accrediting agencies are implementing special emphasis programs on noise reduction in the laboratory.

Because noise is becoming more of a concern in the clinical setting, the College of American Pathologists has added evaluation of noise in the laboratory under their general checklist for accreditation (GEN.70824).

#### **8.4.1 Health Effects**

Exposure to continuous noise may lead to the following stress-related symptoms:

- Depression;
- Irritability;
- Decreased concentration in the workplace;
- Reduced efficiency and decreased productivity;
- Noise-induced hearing loss;
- Tinnitus (i.e., ringing in the ears); and
- Increased errors in laboratory work.

There are several steps that employers can take to minimize the noise in the laboratory, including:



- Moving noise-producing equipment (e.g., freezers, refrigerators, incubators and centrifuges) from the laboratory to an equipment room;
- Locating compressors for controlled-temperature rooms remotely; and
- Providing acoustical treatment on ceilings and walls.

## **8.5 Safety Hazards**

Supervisors and principle investigators must assess tasks to identify potential worksite hazards and provide and ensure that workers use appropriate personal protective equipment (PPE) as stated in the PPE standard, 29 CFR 1910.132.

Supervisors and principle investigators must require workers to use appropriate hand protection when hands are exposed to hazards such as sharp instruments and potential thermal burns. Examples of PPE which may be selected include using oven mitts when handling hot items, and steel mesh or cut-resistant gloves when handling or sorting sharp instruments as stated in the Hand Protection standard, 29 CFR 1910.138.

### **8.5.1 Autoclaves and Sterilizers**

Laboratory workers should be trained to recognize the potential for exposure to burns or cuts that can occur from handling or sorting hot sterilized items or sharp instruments when removing them from autoclaves/ sterilizers or from steam lines that service the autoclaves.

To prevent injuries from occurring, laboratory personnel must be trained on and follow the following safe work practices:

- Use appropriate hand protection when hands are exposed to hazards such as cuts, lacerations or thermal burns. Using oven mitts for handling hot items, and steel mesh gloves for handling or sorting sharp instruments are examples of appropriate PPE.
- Ensure that the autoclave/sterilizer door is closed and locked before beginning the cycle.
- Do not remove items from an autoclave/sterilizer until they have cooled.
- Avoid handling the sharp ends of instruments.
- Use forceps or other tools to remove sharp instruments from baskets and autoclaves.

### **8.5.2 Centrifuges**

Centrifuges, due to the high speed at which they operate, have great potential for injuring users if not operated properly. Unbalanced centrifuge rotors can result in injury, even death. Sample container breakage can generate aerosols that may be harmful if inhaled. Most centrifuge accidents are the result of user error. To prevent injuries or exposure to dangerous substances, laboratory workers must be trained to follow good work practices. To avoid injury, workers should follow the manufacturer's

operating instructions for each make and model of centrifuge that they use. Furthermore, follow these steps for the safe operation of centrifuges:

- Ensure that centrifuge bowls and tubes are dry.
- Ensure that the spindle is clean.
- Use matched sets of tubes, buckets and other equipment.
- Always use safety centrifuge cups to contain potential spills and prevent aerosols.
- Inspect tubes or containers for cracks or flaws before using them.
- Avoid overfilling tubes or other containers (e.g., in fixed angle rotors, centrifugal force may drive the solution up the side of the tube or container wall).
- Ensure that the rotor is properly seated on the drive shaft.
- Make sure that tubes or containers are properly balanced in the rotor.
- Only check O-rings on the rotor if you are properly trained.
- Apply vacuum grease in accord with the manufacturer's guidelines.
- Do not exceed the rotor's maximum run speed.
- Close the centrifuge lid during operation.
- Make sure that the centrifuge is operating normally before leaving the area.
- Make sure that the rotor has come to a complete stop before opening the lid.

When centrifuging infectious materials wait 10 minutes after the centrifuge rotor has stopped before opening the lid. Workers should also be trained to use appropriate decontamination and cleanup procedures for the materials being centrifuged if a spill occurs and to report all accidents to their supervisor immediately.

### 8.5.3 Compressed Gases

According to OSHA's Laboratory standard, a "compressed gas"

- is a gas or mixture of gases in a container having an absolute pressure exceeding 40 pounds per square inch (psi) at 70°F (21.1°C); or
- is a gas or mixture of gases having an absolute pressure exceeding 104 psi at 130°F (54.4°C) regardless of the pressure at 70°F (21.1°C); or
- is a liquid having a vapor pressure exceeding 40 psi at 100°F (37.8°C) as determined by ASTM (American Society for Testing and Materials) D-323-72, [29 CFR 1910.1450(c)(1)-(3)].

Within laboratories, compressed gases are usually supplied either through fixed piped gas systems or individual cylinders of gases. Compressed gases can be toxic, flammable, oxidizing, corrosive, or inert. Leakage of any of these gases can be hazardous. Leaking inert gases (e.g., nitrogen) can quickly displace air in a large area creating an oxygen-deficient atmosphere; toxic gases (e.g., can create poison atmospheres; and flammable (oxygen) or reactive gases can result in fire and exploding cylinders.

In addition, there are hazards from the pressure of the gas and the physical weight of the cylinder. A gas cylinder falling over can break containers and crush feet. The

gas cylinder can itself become a missile if the cylinder valve is broken off. Laboratories must include compressed gases in their inventory of chemicals in the Chemical Hygiene Plan.

Compressed gases contained in cylinders vary in chemical properties, ranging from inert and harmless to toxic and explosive. The high pressure of the gases constitutes a serious hazard if gas cylinders sustain physical damage and/or are exposed to high temperatures.

Store, handle, and use compressed gases in accord with OSHA's Compressed Gases standard (29 CFR 1910.101) and Pamphlet P-1-1965 from the Compressed Gas Association.

- All cylinders whether empty or full must be stored upright.
- Secure cylinders of compressed gases. Cylinders should never be dropped or allowed to strike each other with force.
- Transport compressed gas cylinders with protective caps in place and do not roll or drag the cylinders.

#### **8.5.4 Cryogenics and Dry Ice**

Cryogenics are substances used to produce very low temperatures [below  $-153^{\circ}\text{C}$  ( $-243^{\circ}\text{F}$ )], such as liquid nitrogen (LN<sub>2</sub>) which has a boiling point of  $-196^{\circ}\text{C}$  ( $-321^{\circ}\text{F}$ ), that are commonly used in laboratories.

Although not a cryogenic, solid carbon dioxide or dry ice which converts directly to carbon dioxide gas at  $-78^{\circ}\text{C}$  ( $-109^{\circ}\text{F}$ ) is also often used in laboratories.

Shipments packed with dry ice, samples preserved with liquid nitrogen, and in some cases, techniques that use cryogenic liquids, such as cryogenic grinding of samples, present potential hazards in the laboratory.

##### **8.5.4.1 Overview of Cryogenic Safety Hazards**

The safety hazards associated with the use of cryogenic liquids are categorized as follows:

###### **8.5.4.1.1 Cold contact burns**

Liquid or low-temperature gas from any cryogenic substance will produce effects on the skin similar to a burn.

###### **8.5.4.1.2 Asphyxiation**

Degrees of asphyxia will occur when the oxygen content of the working environment is less than 20.9% by volume. This decrease in oxygen content can be caused by a failure/leak of a cryogenic vessel or transfer line and subsequent vaporization of the cryogen. Effects from oxygen deficiency become noticeable at levels below approximately 18% and sudden death may occur at approximately 6% oxygen content by volume.

#### **8.5.4.1.3 Explosion - Pressure**

Heat flux into the cryogen from the environment will vaporize the liquid and potentially cause pressure buildup in cryogenic containment vessels and transfer lines. Adequate pressure relief should be provided to all parts of a system to permit this routine outgassing and prevent explosion.

#### **8.5.4.1.4 Explosion - Chemical**

Cryogenic fluids with a boiling point below that of liquid oxygen can condense oxygen from the atmosphere. Repeated replenishment of the system can thereby cause oxygen to accumulate as an unwanted contaminant. Similar oxygen enrichment may occur where condensed air accumulates on the exterior of cryogenic piping. Violent reactions, e.g., rapid combustion or explosion, may occur if the materials which make contact with the oxygen are combustible.

#### **8.5.4.2 Employer Responsibility**

Wright State, specifically the supervisor in charge of an apparatus, must ensure that the cryogenic safety hazards are minimized. This will entail

- a safety analysis and review for all cryogenic facilities,
- cryogenic safety and operational training for relevant workers,
- appropriate maintenance of cryogenic systems in their original working order, i.e., the condition in which the system was approved for use, and
- upkeep of inspection schedules and records.

Workers must be trained to use the appropriate personal protective equipment (PPE) Whenever handling or transfer of cryogenic fluids might result in exposure to the cold liquid, boil-off gas, or surface, protective clothing must be worn.

This includes:

- face shield or safety goggles;
- safety gloves; and
- long-sleeved shirts, lab coats, aprons.

Eye protection is required at all times when working with cryogenic fluids. When pouring a cryogen, working with a wide-mouth Dewar flask or around the exhaust of cold boil-off gas, use of a full-face shield is recommended.

Hand protection is required to guard against the hazard of touching cold surfaces. It is recommended that Cryogen Safety Gloves be used by the worker.

#### **8.5.4.3 General Precautions When Working with Dry Ice or LN2**

- Avoid eye or skin contact with these substances.
- Never handle dry ice or LN2 with bare hands.
- Use cryogenic gloves, which are designed specifically for working in freezers below -80°C and for handling containers or vials stored in these freezers.

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- Cryogenic gloves need to be loose-fitting so that they can be readily removed if LN2 splashes into them or a piece of dry ice falls into them.
- Always use appropriate eye protection.
- Do not use or store dry ice or LN2 in confined areas, walk-in refrigerators, environmental chambers or rooms without ventilation. A leak in such an area could cause an oxygen-deficient atmosphere.
- Never place a cryogen on tile or laminated counters because the adhesive will be destroyed.
- Never store a cryogen in a sealed, airtight container at a temperature above the boiling point of the cryogen; the pressure resulting from the production of gaseous carbon dioxide or nitrogen may lead to an explosion.
- For more information about specific cryogenics, read the Safety Data Sheet for the substance in question

#### **8.5.4.4 First Aid**

In case of exposure to cryogenics or dry ice, remove any clothing that is not frozen to the skin. Do NOT rub frozen body parts because tissue damage may result. Obtain medical assistance as soon as possible.

Place the affected part of the body in a warm water bath (not above 40°C). Never use dry heat.

#### **8.5.5 Electrical**

In the laboratory, workers may be exposed to electrical hazards including electric shock, arc blasts, electrocutions, fires and explosions. Potential exposures to electrical hazards can result from faulty electrical equipment/instrumentation or wiring, damaged receptacles and connectors, or unsafe work practices.

To avoid such hazards, follow these best practices:

- Always follow manufacturer's recommendations for using electrical equipment.
- Do not use electrical equipment to perform a task for which it is not designed.
- Most equipment includes either a 3-pronged plug or double insulation. Equipment with neither of these features is less safe but may meet electrical codes. You will not be protected from electric shock if a 3-pronged plug is not inserted into a 3-prong outlet.
- If you plug more than two pieces of low demand equipment into a standard outlet, use a fused power strip that will shut off if too much power is used.
- Make sure that any outlet near a sink or other water source is Ground-Fault Circuit Interrupter (GFCI) protected. If you have a GFCI, periodically test it by plugging something into it and pushing the "test" button. Once the equipment shuts off just turn it back on.
- Above all, do not disable any electrical safety feature.
- Before turning equipment on, check that all power cords are in good condition.
- Do not use extension cords as a substitute for permanent wiring.

- If you see a person being electrocuted, DO NOT TOUCH THEM! The electricity can go through you, too. If possible, turn off the power (pull the plug or trip the circuit breaker), or use an item made of non-conductive material (e.g., wooden broom handle) to pry him or her away from the contact. Call 911 immediately.

#### **8.5.5.1 Employers are responsible for complying with OSHA's standard 1910 Subpart S Electrical**

Subpart S is comprehensive and addresses electrical safety requirements for the practical safeguarding of workers in their workplaces. This Subpart includes, but is not limited to, these requirements:

- Electrical equipment must be free from recognized hazards, 29 CFR 1910.303(b)(1);
- Listed or labeled equipment must be used or installed in accord with any instructions included in the listing or labeling, 29 CFR 1910.303(b)(2);
- Sufficient access and working space must be provided and maintained around all electrical equipment operating at  $\leq 600$  volts to permit ready and safe operation and maintenance of such equipment, 29 CFR 1910.303(g)(1);
- All electrical service near sources of water must be properly grounded.
- All damaged receptacles and portable electrical equipment must be tagged out and removed from service, 29 CFR 1910.334(a)(2)(ii);
- All damaged receptacles and portable electrical equipment must be repaired before placing them back into service, 29 CFR 1910.334(a)(2)(ii);
- Workers must be trained not to plug or unplug energized equipment when their hands are wet, 29 CFR 1910.334(a)(5)(i);
- Appropriate work practices must be selected and used, 29 CFR 1910.333; and
- The requirements for Hazardous Classified Locations, 29 CFR 1910.307 must be followed. This section covers the requirements for electric equipment and wiring in locations that are classified based on the properties of the flammable vapors, liquids or gases, or combustible dusts or fibers that may be present therein and the likelihood that a flammable or combustible concentration or quantity is present.

Notes:

- Only "Qualified Persons," as defined by OSHA in 29 CFR 1910.399, are to work on electrical circuits/systems.
- Workers must be trained to know the locations of circuit breaker panels that serve their lab area.

#### **8.5.6 Fire**

Fire is the most common serious hazard that one faces in a typical laboratory. While proper procedures and training can minimize the chances of an accidental fire, laboratory workers should still be prepared to deal with a fire emergency should it

occur. In dealing with a laboratory fire, all containers of infectious materials should be placed into autoclaves, incubators, refrigerators, or freezers for containment.

Small bench-top fires in laboratory spaces are not uncommon. Large laboratory fires are rare. However, the risk of severe injury or death is significant because fuel load and hazard levels in labs are typically very high. Laboratories, especially those using solvents in any quantity, have the potential for flash fires, explosion, rapid spread of fire, and high toxicity of products of combustion (heat, smoke, and flame).

#### **8.5.6.1 Training for Fire Prevention**

- Plan work. Have a written emergency plan for your space and/or operation.
- Minimize materials. Have present in the immediate work area and use only the minimum quantities necessary for work in progress. Not only does this minimize fire risk, it reduces costs and waste.
- Observe proper housekeeping. Keep work areas uncluttered, and clean frequently. Put unneeded materials back in storage promptly. Keep aisles, doors, and access to emergency equipment unobstructed at all times.
- Observe restrictions on equipment (i.e., keeping solvents only in an explosion-proof refrigerator).
- Keep barriers in place (shields, hood doors, lab doors).
- Wear proper clothing and personal protective equipment.
- Avoid working alone.
- Store solvents properly in approved flammable liquid storage cabinets.
- Shut door behind you when evacuating.
- Limit open flames use to under fume hoods and only when constantly attended.
- Keep combustibles away from open flames.
- Do not heat solvents using hot plates.
- Remember the “RACE” rule in case of a fire.

R= Rescue/remove all occupants

A= Activate the alarm system

C= Confine the fire by closing doors

E= Evacuate/Extinguish

#### **8.5.6.2 Training for Emergency Procedures**

Workers must be trained in the following emergency procedures:

- Know what to do. You tend to do under stress what you have practiced or pre-planned. Therefore, planning, practice and drills are essential.
- Know where things are: The nearest fire extinguisher, fire alarm box, exit(s), telephone, emergency shower/eyewash, and first-aid kit, etc.

- Be aware that emergencies are rarely “clean” and will often involve more than one type of problem. For example, an explosion may generate medical, fire, and contamination emergencies simultaneously.
- Train workers and exercise the emergency plan.
- Learn to use the emergency equipment provided.

OSHA’s Portable Fire Extinguishers standard, 29 CFR 1910.157 requires that workers need to be trained and to be aware of the different fire extinguisher types and how to use them. OSHA’s Portable Fire Extinguishers standard, 29 CFR 1910.157, applies to the placement, use, maintenance, and testing of portable fire extinguishers provided for the use of workers. This standard requires that a fire extinguisher be placed within 75 feet for Class A fire risk (ordinary combustibles; usually fuels that burn and leave “ash”) and within 50 feet for high-risk Class B fire risk (flammable liquids and gases; in the laboratory, many organic solvents and compressed gases are fire hazards).

The two most common types of extinguishers in the chemistry laboratory are pressurized dry chemical (Type BC or ABC) and carbon dioxide. In addition, you may also have a specialized Class D dry powder extinguisher for use on flammable metal fires.

Water-filled extinguishers are not acceptable for laboratory use.

#### **8.5.6.3 “PASS” Rule for Fire Extinguisher**

Laboratory workers need to remember the “PASS” rule for fire extinguishers

PASS summarizes the operation of a fire extinguisher.

P - Pull the pin

A - Aim extinguisher nozzle at the base of the fire

S - Squeeze the trigger while holding the extinguisher upright

S - Sweep the extinguisher from side to side; cover the fire with the spray

#### **8.5.6.4 Procedures for Clothing Fire**

Laboratory workers need to know appropriate procedures in the event of a clothing fire.

- If the floor is not on fire, STOP, DROP and ROLL to extinguish the flames or use a fire blanket or a safety shower if not contraindicated (i.e., there are no chemicals or electricity involved).
- If a coworker’s clothing catches fire and he/she runs down the hallway in panic, tackle him/her and smother the flames as quickly as possible, using appropriate means that are available (e.g., fire blanket, fire extinguisher).



### 8.5.7 Lockout/Tagout

Workers performing service or maintenance on equipment may be exposed to injuries from the unexpected energization, start-up of the equipment, or release or stored energy in the equipment. OSHA's Control of Hazardous Energy standard, 29 CFR 1910.147, commonly referred to as the "Lockout/Tag out" standard, requires the adoption and implementation of practices and procedures to shut down equipment, isolate it from its energy source(s), and prevent the release of potentially hazardous energy while maintenance and servicing activities are being performed. It contains minimum performance requirements, and definitive criteria for establishing an effective program for the control of hazardous energy. However, employers, including Wright State, have the flexibility to develop Lockout/Tagout programs that are suitable for their respective facilities. Wright State has a Lockout/Tagout program.

This standard establishes basic requirements involved in locking and/or tagging equipment while installation, maintenance, testing, repair or construction operations are in progress. The primary purpose is to prevent hazardous exposure to personnel and possible equipment damage. The procedures apply to the shutdown of all potential energy sources associated with the equipment. These could include pressures, flows of fluids and gases, electrical power, and radiation.

This standard covers the servicing and maintenance of machines and equipment in which the "**unexpected**" energization or start-up of the machines or equipment, or release of stored energy could cause injury to workers.

Under the standard, the term "**unexpected**" also covers situations in which the servicing and/or maintenance is performed during ongoing normal production operations if:

- A worker is required to remove or bypass machine guards or other safety devices, 29 CFR 1910.147(a)2(ii)(A) or
- A worker is required to place any part of his or her body into a point of operation or into an area on a machine or piece of equipment where work is performed, or into the danger zone associated with the machine's operation, 29 CFR 1910.147(a) (2)(ii)(B).

The Lockout/Tagout standard establishes minimum performance requirements for the control of such hazardous energy.

Maintenance activities can be performed with or without energy present. A probable, underlying cause of many accidents resulting in injury during maintenance is that work is performed without the knowledge that the system, whether energized or not, can produce hazardous energy. Unexpected and unrestricted release of hazardous energy can occur if:

- all energy sources are not identified;
- provisions are not made for safe work practices with energy present; or

- deactivated energy sources are reactivated, mistakenly, intentionally, or accidentally, without the maintenance worker's knowledge.

Problems involving control of hazardous energy require procedural solutions. Procedural solutions must be adopted for controlling hazards to ensure worker safety during maintenance. However, such procedures are effective only if strictly enforced.

Wright State is committed to strict implementation of such procedures.

#### **8.5.7.1 Trips, Slips and Falls**

Worker exposure to wet floors or spills and clutter can lead to slips/trips/falls and other possible injuries. To keep workers safe, employers are referred to OSHA standard 29 CFR 1910 Subpart D – Walking-Working Surfaces, Subpart E – Means of Egress, and Subpart J - General environmental controls which states the following:

- Keep floors clean and dry, 29 CFR 1910.22(a)(2). In addition to being a slip hazard, continually wet surfaces promote the growth of mold, fungi, and bacteria that can cause infections.
- Provide warning (caution) signs for wet floor areas, 29 CFR 1910.145(c)(2).
- Where wet processes are used, maintain drainage and provide false floors, platforms, mats, or other dry standing places where practicable, or provide appropriate waterproof footwear, 29 CFR 1910.141(a)(3)(ii).
- The Walking/Working Surfaces standard requires that all employers keep all places of employment clean and orderly and in a sanitary condition, 29 CFR 1910.22(a)(1).
- Keep aisles and passageways clear and in good repair, with no obstruction across or in aisles that could create a hazard, 29 CFR 1910.22(b)(1).
- Provide floor plugs for equipment, so that power cords need not run across pathways.
- Keep exits free from obstruction. Access to exits must remain clear of obstructions at all times, 29 CFR 1910.37(a)(3).
- Ensure that spills are reported and cleaned up immediately.
- Eliminate cluttered or obstructed work areas.
- Use prudent housekeeping procedures such as using caution signs, cleaning only one side of a passageway at a time, and provide good lighting for all halls and stairwells to help reduce accidents, especially during the night hours.
- Instruct workers to use the handrail on stairs, to avoid undue speed, and to maintain an unobstructed view of the stairs ahead of them even if that means requesting help to manage a bulky load.
- Eliminate uneven floor surfaces.
- Promote safe work practices, even in cramped working spaces.
- Avoid awkward positions, and use equipment that makes lifting easier.

## 9 Fume Hood Inspections and Operating Procedures

Wright State University's Department of Environmental Health and Safety and all laboratories subjected to the provisions of the OSHA Laboratory Standard will follow the procedures contained within as it relates to the inspection operations of chemical fume hoods.

### 9.1 Responsibilities

#### 9.1.1 Environmental Health and Safety:

Inspections – All chemical fume hoods will be inspected annually. The following hood features will be checked:

1. Sash Operations – The following physical conditions should be met relating to sash operations:
  - The sash moves up and down easily.
  - The sash does not bind at any place in the track.
  - The safety glass is intact and clear, allowing for an unobstructed view of the inside of the cabinet.
  - The sash heights, in the full raised position, should be between 29 and 31 inches.
  - Check for leakage at the top where the vertical sash goes past the upper structure of the hood
2. Fume Hood usage: Check the following for compliance:
  - Work is being conducted six (6) inches back into the hood.
  - The hood is not being used for storage.
  - There is not excessive lab apparatus in the fume hood and the present lab apparatus is not interfering with the desired air flow pattern.
  - The fume hood itself (inside) is being kept clean.
3. Air Flow Measurements - Regular inspections will be made of all chemical fume hoods to verify proper hood operations. Annual fume hood surveys will be made relative to capture and face velocities.
  - Smoke Velocity – Utilizing smoke tubes or other flow indicator, a check is to be made in front of the fume hood to verify the in-flow of air into the hood and to verify the absence of serious turbulence which would throw contaminated air back into the workplace and into the staff person's breathing zone.
  - Face Velocity – All hood should be equipped with a magnehelic gauge which provides for a quick and easy means of verifying average face velocity. In the absence of a magnehelic gauge or other flow-indicating

device, an actual hood survey will need to be accomplished using a thermoanemometer.

### 9.1.2 Laboratory Supervisor

The laboratory supervisor or his/her designee is responsible for the following daily checks:

- Verify that the fume hood is working satisfactory. This can be accomplished by the reading of the magnehelic gauge or electronic display, where available.
- Verify that the sash is working properly and used when there is potential for a reaction or fire/explosion in the hood.
- Verify that all work is conducted within six (6) inches inside the hood.
- Maintain a satisfactory level of housekeeping within the hood and ensure that the hood is not being used for storage purposes.
- Ensure that the air foil is available and installed properly.
- Keep source of air movement in front of the hood to a minimum.
- Ensure that lights and all utilities inside the hood are operational. Keep the light fixture within the hood clean.

### 9.1.3 Laboratory Personnel

Before using a fume hood:

- Understand how the hood works.
- Be trained to use it properly.
- Know the hazards of the chemical you are working with; refer to the chemical's Safety Data Sheet if you are unsure.
- Ensure that the hood is on.
- Make sure that the sash is open to the proper operating level, which is usually indicated by arrows on the frame.
- Make sure that the air gauge indicates that the air flow is within the required range.

When using a fume hood:

- Never allow your head to enter the plane of the hood opening. For example, for vertical rising sashes, keep the sash below your face; for horizontal sliding sashes, keep the sash positioned in front of you and work around the side of the sash.
- Use appropriate eye protection.
- Be sure that nothing blocks the airflow through the baffles or through the baffle exhaust slots.
- Elevate large equipment (e.g., a centrifuge) at least two inches off the base of the hood interior.
- Keep all materials inside the hood at least six inches from the sash opening.

When not working in the hood, close the sash.

## 10 Training and Information

Personnel training at all levels within the university is essential. Responsibility and accountability throughout the organization are key elements in a strong safety and health program.

Laboratory workers must be provided with information and training relevant to the hazards of the chemicals present in their laboratory (29 CFR 1910.1450(f)). The training must be provided at the time of initial assignment to a laboratory and prior to assignments involving new exposure situations.

At a minimum, laboratory personnel should be trained on their facility's specific CHP, methods and observations that may be used to detect the presence or release of a hazardous chemical (such as monitoring conducted by the employer, continuous monitoring devices, visual appearance or odor of hazardous chemicals when being released), the physical and health hazards of chemicals in the work area and means to protect themselves from these hazards. Trained laboratory personnel must know shut-off procedures in case of an emergency. All SDSs must be made available to the employees.

Training required by 29 CFR 1910.1450 which is applicable to laboratory operations under the OSHA Occupational Exposure to Hazardous Chemicals in Laboratories Standard will be provided either by qualified staff of the Department of Environmental Health and Safety and/or the laboratory supervisor.

Summary of training requirements:

- Chemical Hygiene Plan – At the time of implementation, Environmental Health and Safety will provide training to laboratory supervisors and staff. Subsequent training will be the responsibility of the laboratory supervisor.
- Chemical Inventory Procedures – At the time of implementation, Environmental Health and Safety will provide training to laboratory supervisors and staff. Subsequent training will be the responsibility of the laboratory supervisor.
- Hazard Communication Plan – Environmental Health and Safety will provide the initial and refresher training, as needed.
- Bloodborne Pathogen Plan – Environmental Health and Safety will provide the initial and refresher training.

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*Reminder: Bloodborne Pathogen Training is required for new employees working with blood and/or other body fluids within ten (10) days of hire.*

- Maintenance/Custodial Personnel – Environmental Health and Safety will provide the initial and annual refresher training on the Do's and Don'ts of working safely before and during tasks in the laboratory.

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- Specific Needs – Other training needs that are identified by Environmental Health and Safety and/or the laboratory supervisor, which are within the expertise of the Environmental Health and Safety staff. Examples of this training include fire extinguisher training, emergency egress, use of protective equipment, etc.
- Laboratory Supervisors: Laboratory supervisors are responsible for the following training within their work areas. They are also responsible for ensuring that the staff know and follow the chemical hygiene rules, and that protective equipment is available and in working order.
  - Chemical Hygiene Plan – Following the initial training given by Environmental Health and Safety, the laboratory supervisor is responsible for providing training on the chemical hygiene plan to all new employees. The training and education should be a regular, continuing activity, not simply an annual refresher training as required under 29 CFR 1910.1450.
  - Chemical Inventory Procedures – Following the initial training given by Environmental Health and Safety, the laboratory supervisor is responsible for training the laboratory staff to conduct the annual chemical inventory within his/her assigned laboratories.
  - Job Specific Training – The laboratory supervisor is responsible for identifying specific training needs for their staff and possibly the University maintenance and custodial staff to ensure that all operations are conducted in a manner conducive to their health and well-being. The laboratory supervisor should seek the advice and assistance of the Environmental Health and Safety Department, as needed.

## **10.1 Information and Training**

### **10.1.1 Information**

Information and Training Program – The training should include, but not be limited to, the following items:

- The content of the OSHA Laboratory standard and its appendices (the full text must be made available);
- The location and availability of the Chemical Hygiene Plan;
- Permissible exposure limits (PELs) for OSHA regulated substances, or recommended exposure levels for other hazardous chemicals where there is no applicable standard;
- Signs and symptoms associated with exposure to hazardous chemicals in the laboratory; and
- The location and availability of reference materials on the hazards, safe handling, storage and disposal of hazardous chemicals in the laboratory, including, but not limited to, Safety Data Sheets (SDSs).

### 10.1.2 Training Requirements

The training must include the following:

- Methods and observations used to detect the presence or release of a hazardous chemical. These may include employer monitoring, continuous monitoring devices, and familiarity with the appearance and odor of the chemicals;
- The physical and health hazards of chemicals in the laboratory work area;
- The measures that workers can take to protect themselves from these hazards, including protective equipment, appropriate work practices, and emergency procedures;
- Applicable details of the employer's written Chemical Hygiene Plan;
- Retraining, if necessary.

## 11 Laboratory Procedures Requiring Prior Approval

It is not the intention of the Department of Environmental Health and Safety to curtail educational and research laboratory activities. It is, however, our responsibility to ensure that activities conducted in the university laboratories are done so in a manner that

1. does not affect the health and well-being of employees, students and visitors,
2. does not offer the potential for property damage because of fire and/or explosions,
3. does not release materials to the atmosphere or the storm and sanitary sewers which could have adverse effects on the university and/or adjacent communities, and
4. permits the disposal of waste products (biological, chemical and/or radiological) in a manner of prudent practice and in compliance with federal and state environmental rules and regulations.

To accomplish this, the Department of Environmental Health and Safety (EHS) needs to know when highly toxic and/or hazardous materials are planned for use in university laboratories. Identification of the specific agent(s), quantities of use, duration of experiment and how they will be used also needs to be known. Once this information is provided to EHS, an evaluation will be conducted to determine if the EHS staff/facilities are adequately trained/prepared in the handling, storage and disposal procedures. This evaluation will include verifying that emergency response procedures are available and acceptable, laboratory equipment is in good repair (i.e. fume hoods) and adequate for the proposed study, and all other health and safety considerations are properly addressed. Following our satisfaction that all conditions are met and that other interested parties, such as Fairborn Fire Department, Wright State Police Department, Physical Plant, etc. have also been notified, approval to proceed will be given.



The Department of Environmental Health and Safety will exercise approval authority over the following biological and chemical agents. Prior approval authority is always vested for the use of radioactive materials, radiation-producing equipment and laser systems.

- **Chemicals**: All chemicals listed under Group 1 (“carcinogenic to humans”) in the current edition of IARC’s (International Agency for Research on Cancer) Monographs.
- **Biologicals**: All **Risk Group 3** Biological agents.
- **Toxins**: All reproductive toxins and substances which have a high degree of acute toxicity to man.
- The Environmental Health and Safety Chemical Hygiene Officer (CHO) will work with the department safety representative and the applicable faculty/staff member in the approval process. The involvement of the applicable Institutional Safety Committee will be sought when deemed necessary.

## **12 Medical Consultation and Examinations**

Wright State provides provisions for medical consultation and examination when exposure to a hazardous chemical has or may have taken place for employees covered under 29 CFR 1910.1450.

The Department of Environmental Health and Safety will investigate all incidents of chemical exposures and will make recommendations for medical consultation when deemed necessary or as required by the Laboratory Standard. All costs associated with medical consultation will be paid for by Wright State University.

### **12.1 Requirements Under 29 CFR 1910.1450, Laboratory Standard**

Wright State University shall provide all employees who work with hazardous chemicals an opportunity to receive medical attention, including any follow-up examinations which the examining physician deems necessary under the following circumstances:

- Provide all exposed workers with an opportunity to receive medical attention by a licensed physician, including any follow-up examinations which the examining physician determines to be necessary.
- Provide an opportunity for a medical consultation by a licensed physician whenever a spill, leak, explosion or other occurrence results in the likelihood that a laboratory worker experienced a hazardous exposure to determine whether a medical examination is needed.
- Provide an opportunity for a medical examination by a licensed physician whenever a worker develops signs or symptoms associated with a hazardous chemical to which he or she may have been exposed in the laboratory.
- Establish medical surveillance for a worker as required by the particular standard when exposure monitoring reveals exposure levels routinely exceeding the OSHA action level or, in the absence of an action level, the PEL for an OSHA regulated substance.
- Provide the examining physician with the identity of the hazardous chemical(s) to which the individual may have been exposed, and the conditions under which the exposure may have occurred, including quantitative data, where available, and a description of the signs and symptoms of exposure the worker may be experiencing.
- Provide all medical examinations and consultations without cost to the worker,
- without loss of pay, and at a reasonable time and place.

### **12.2 Written Opinion**

All medical examinations and consultations will be performed by or under the direct supervision of a licensed physician.

The examining physician must complete a written opinion that includes the following information:

- Recommendations for further medical follow-up.

- The results of the medical examination and any associated tests.
- Any medical condition revealed during the examination that may place the individual at increased risk because of exposure to a hazardous chemical in the workplace.
- A statement that the worker has been informed of the results of the consultation or medical examination and any medical condition that may require further examination or treatment. However, the written opinion must not reveal specific findings of diagnoses unrelated to occupational exposure.

The identity of the hazardous chemical, a description of the incident, and any signs and symptoms that the employee may experience will be relayed to the physician.

The requirements of providing information to the examining physician and the physician's written as addressed in 1910.1450 (g) (3) and (4) will be the responsibility of the Environmental Health and Safety Department.

### **12.2.1 Recordkeeping**

Wright State's Department of Environmental Health and Safety maintains an accurate record of exposure monitoring activities and exposure measurements as well as medical consultations and examinations, including medical tests and written opinions.

Employers generally must maintain worker exposure records for 30 years and medical records for the duration of the worker's employment plus 30 years, unless one of the exemptions listed in 29 CFR 1910.1020(d)(1)(i)(A)-(C) applies. Such records must be maintained, transferred, and made available, in accord with 29 CFR 1910.1020, to an individual's physician or made available to the worker or his/her designated representative upon request.

All accident, fatality, illness, injury, and medical records and exposure monitoring records will be retained by Wright State in accordance with the requirement of state and federal regulations (29 CFR part 1904 and 29 CFR 1910.1450(j)).

Any exposure monitoring results will be provided to affected laboratory staff within 15 working days after receipt of the results (29 CFR 1910.1450(d)(4)).

### **12.3 Requirements Under CFR 1910.120, Access to Records:**

Whenever an employee requests access to exposure or medical records, Wright State University will assure that access is provided in a reasonable time, place and manner (within 15 days) at no cost to the employee.

The medical records will be maintained for the duration of the employee's employment plus thirty (30) years. All exposure records will be maintained for at least thirty (30) years.

### 13 Particularly Hazardous Substances

Additional worker protection is required for work with particularly hazardous substances.

Particularly hazardous substances include:

- “select carcinogens”,
- reproductive toxins and
- substances which have a high degree of acute toxicity and/or hazardous properties

which in view of its intended use could present an extraordinary potential for a fire/explosion or an intense chemical reaction.

Laboratory supervisors are responsible for notifying the Environmental Health and Safety Department when work is anticipated to include *particularly hazardous substances*.

Please refer to Laboratory Procedures Requiring Prior Approval.

The Environmental Health and Safety Department, in a cooperative effort with the researcher will ensure that a thorough review of the proposed activity is performed and that all measures necessary to protect life and property are taken before the activity commences. Measures could include, but not necessarily be limited to:

1. The establishment of a designated area for the proposed activity.
2. The use of a containment device such as a fume hood, glove box or other type of ventilated enclosure.
3. Use of explosive-proof equipment, remote operations, blast shields, etc.
4. The development of decontamination procedures following daily operations and in the event of a spill or other accident.
5. Development of emergency response procedures beyond those already established, if deemed necessary.
6. Restriction of non-laboratory personnel to the area during the duration of the research activity, if necessary.
7. Special training and medical surveillance of involved employees, if deemed necessary.
8. The posting of the laboratory and the designated area.
9. Procedures for the safe removal and disposal of contaminated waste.

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*The need for special protective measures should be identified when particularly hazardous substances are identified.*

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It is essential that Environmental Health and Safety be notified at the earliest date possible when research involving extremely toxic and/or hazardous substances is planned for use.