

UTRGV Laboratory Safety Manual

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The University of Texas Rio Grande Valley

DEPARTMENT OF ENVIRONMENTAL HEALTH, SAFETY AND RISK MANAGEMENT | 956-665-3690

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A. INTRODUCTION

Note to Users of This Laboratory Safety Manual

The Department of Environmental Health, Safety and Risk Management (EHSRM) has prepared this manual to ensure that laboratory activities are conducted in compliance with any applicable rules or regulations and best practices applicable to a laboratory environment. The health and safety policy of The University is to take every reasonable precaution to protect the health and safety of faculty, staff, students, volunteers and the public. Mandatory safety standards, as interpreted by the requirements and policies stated in this manual apply to faculty, staff, researchers, students, and volunteers engaged in laboratory operations involving the utilization of chemical products and in performing common laboratory procedures.

This manual includes information concerning safe laboratory practices, the use of personal protective equipment, emergency procedures, use and storage of chemicals, and the proper methods of waste disposal. This manual covers safety practices for any area containing chemical, biological and/or radiological hazards and/or waste. This manual also covers hazard communication and incident response. This information is intended to help those in the laboratory minimize hazards to themselves and their colleagues.

In view of the wide variety of chemical products handled in laboratories, it should not be assumed that the precautions and requirements stated in this manual are all-inclusive. Faculty, researchers, and students are expected to learn about the hazards of chemical products before handling them. It is expected that the local departmental Chemical Hygiene Officers and Principal Investigators will append to this manual any supplementary information pertinent to their specific areas.

Throughout this manual the terms "employee", "laboratory worker", and "laboratory personnel" are used interchangeably and are intended to include students working in a laboratory, unless the context indicates otherwise.

Please contact **EHSRM Edinburg Campus: 956-665-3690 or Brownsville Campus: 956-882-5929** for more information or for assistance.

B. RESPONSIBILITIES

Each individual conducting chemical reactions, using chemical materials, or performing laboratory procedures is required to have proper training in the safe handling and disposal of all materials he or she uses. Each is responsible for conducting activities in a manner that will not endanger him- or herself and each must comply with the applicable requirements of state and federal law as well as with University policies and procedures described in this manual. Oversight responsibility for ensuring that laboratory activities involving hazardous chemicals conform to prescribed standards is assigned as follows:

I. University-Wide Safety Responsibilities

The President

The UTRGV President has ultimate responsibility for safety within the institution.

Environmental Health, Safety and Risk Management

- Provides hazard communication and laboratory safety training.
- Conducts periodic and unannounced laboratory inspections to assure compliance with federal, state and local regulations, as well as the policies and procedures contained in this manual and those contained in any supplementary information developed in the college in response to specific activities or areas of research.
- Undertakes necessary enforcement actions to insure full compliance with all institutional safety policies, up to and including independent authority to shut down laboratories for violations of these policies. **Approval of the Dean is not required.**
- Provides for the proper disposal of all regulated waste streams generated in the laboratories.
- Provides limited hazardous material spill response services.
- Reviews laboratory construction, modification and renovation plans for safety design.
- In conjunction with Campus Facilities Operations, oversees fume hood surveys and testing.
- Oversees biological safety cabinet testing.
- Performs exposure monitoring upon request to determine if the permissible exposure limit or action level has been exceeded. Notification shall be provided to laboratory supervisor.
- Provides guidance for maintaining compliance with federal, state, and local regulations, as well as the procedures stated in this manual.
- Conducts laboratory safety evaluations when requested by laboratory supervisors or department chairs.
- Provides assistance in obtaining personal protective equipment.
- Maintains copies of medical consultations and examinations for possible exposures from hazardous chemicals.

Campus Facilities Operations and Utilities

- Maintains facilities and facility-related safety systems to assure continuous operation of laboratories.

- In conjunction with EHSRM, oversees fume hood testing.
- Oversees certification of fume hoods.

II. Departmental Safety Responsibilities

Dean

- Has the responsibility to insure the safe operation of all laboratories and other sites in the University where chemicals are used or laboratory procedures are conducted.
- Will ensure compliance with the policies and procedures contained in this manual and those contained in any supplementary information developed within the University in response to specific activities or areas of research.
- Has independent enforcement authority to close a laboratory for safety violations.
Approval of the Director of EHSRM is not required.

Department Chairs and Directors

- Oversee chemical and biological hygiene within departmental laboratories by ensuring that supervisory personnel reporting to them assume their responsibilities for adhering to all safety policies, regulations and procedures.
- Complete and update annual inventories of hazardous chemicals as required by the Texas Hazard Communication Act and the UTRGV Department of Environmental Health, Safety and Risk Management's Laboratory Safety Program.
- If authorized by the dean, appoint and transfer appropriate enforcement authority to a Departmental Chemical Hygiene Officer (CHO). **The Department Chair or Director assumes all the responsibilities of the CHO when there is not a specified Chemical Hygiene Officer.**

Chemical Hygiene Officer

- Advises the Dean, Department Chair, or Director on matters of chemical safety policies and practices.
- Works with employees to develop and implement the chemical hygiene policies and practices outlined in this manual and those contained in any supplementary information developed in the college in response to specific activities or areas of research.
- Monitors compliance with policies and procedures for the procurement, safe use, and proper disposal of chemicals.
- Investigates and retains records of accidents involving hazardous materials.
- Conducts informational and general training sessions.
- Maintains a resource file of references and publications on safety matters.
- Assists laboratory supervisors and principal investigators in writing Standard Operating Procedures (SOPs) pertinent to their needs.
- Ensures that action is taken to correct laboratory practices and conditions that may result in the release of hazardous materials.
- Ensures that action is taken to correct laboratory practices and conditions identified as unacceptable on laboratory safety self-evaluations and safety inspections.

Principal Investigator/Laboratory Supervisor

- Designs and conducts laboratory processes and operations to assure that employee exposure to risk conforms to the policies, procedures and objectives contained in this manual and those contained in any supplementary information developed in the University in response to specific activities or areas of research.
- Monitors the procurement, safe use, and proper disposal of chemicals.
- Writes Standard Operating Procedures and other information relevant to lab processes in their specific areas as needed to supplement those contained in this manual.
- Instructs employees on the contents of this manual, its appendices, and any supplements, and the location of the manual and related materials within the workplace.
- Takes all reasonable precautions to protect the safety and health of laboratory workers and the environment.
- Schedules services for hazardous waste disposal and oversees the handling of hazardous waste pending proper disposal.
- Conducts regular laboratory safety self-evaluations.
- Completes and updates annual laboratory chemical inventories in accordance with the instructions and schedule provided by the Department of Environmental Health, Safety and Risk Management.
- Informs employees of the permissible exposure limits for the hazardous chemicals listed on inventories and the signs and symptoms associated with exposures to these chemicals.
- Provides site specific training on laboratory hazards as described in the University of Texas-Rio Grande Valley Laboratory Safety Manual.
- Determines the required levels of personal protective equipment, fire extinguishers, fume hoods, flammable liquid storage cabinets, biological safety cabinets, eye washes, safety showers, and spill cleanup kits. Assures that all required equipment is available and in working order and that appropriate training for each item has been provided.
- Has readily available a current copy of a Safety Data Sheet for all hazardous chemicals in the laboratory.
- Posts emergency telephone numbers on the outside of each laboratory door and by all telephones in the area.
- Reports to EHSRM if there is reason to believe that exposure levels for a hazardous chemical exceed the action level or the permissible exposure limits and document the incident.
- Forwards documentation on laboratory accidents and exposures to EHSRM.
- Provides for the safety of visitors.

Laboratory Employee and Other Personnel

- Maintains a thorough understanding of and follows the laboratory policies and procedures in this manual and those contained in any supplementary information developed for UTRGV in response to specific activities or areas of research for all processes using chemical materials.
- Uses and maintains personal protective equipment (i.e. lab coat, chemical splash goggles, face shield, respiratory protection, and gloves) as mandated in this manual for laboratories.
- Uses flammable liquid storage cabinets, acid storage cabinets, biological safety cabinets, fume hoods, and other laboratory safety equipment provided.
- Informs supervisor immediately of any laboratory safety equipment that is needed but not available or that is not in good working order.

- Informs supervisor immediately of exposure symptoms, accidents, or chemical releases and documents incident.
- Successfully completes all applicable training sessions.

C. EMERGENCY PROCEDURES

All accidents, hazardous material spills or other dangerous incidents should be reported. A list of telephone numbers must be posted beside every telephone and near entrances to each laboratory and storeroom. Telephone numbers must include the **Principal Investigator (or Laboratory Supervisor), Emergency Medical Services (9-1-1), UTRGV Police Department (Edinburg Campus or Brownsville Campus: 956-882-4911)**; and the **Department of Environmental Health, Safety and Risk Management (EHSRM) Edinburg Campus: 956-665-3690 or Brownsville Campus: 956-882-5929**. Callers should explain any emergency situation clearly, calmly, and in detail.

I. Immediate Threat to Life or Health:

Call the **UTRGV Police Department at 9-1-1 (campus phone), Edinburg Campus 956-882-4911 or Brownsville Campus: 956-882-4911** for assistance with injured, in case of fire, or for performing rescues. Provide the dispatcher with the nature and the extent of the emergency; be as specific and detailed as possible.

II. Fire:

1. In the event of a fire, pull the nearest fire alarm. If you are unable to control or extinguish a fire, follow the building evacuation procedures.
2. Call the **UTRGV Police Department at 9-1-1 (campus phone), Edinburg Campus or Brownsville Campus: 956-882-4911 (from a cell phone)**.

III. Accidental Exposure or Injury:

1. Attend to any person(s) who may have been contaminated and/or injured if it is safe to reach them. Use safety showers and eyewashes as appropriate. In the case of eye contact, promptly flush eyes with water for a minimum 15-minute period and **seek medical attention immediately**. For ingestion cases, contact the **Poison Control Center at 1-800- 222-1222**. In case of skin contact, promptly flush the affected area with water and remove any contaminated clothing or jewelry. If symptoms persist after washing, seek medical attention.
2. Call the **UTRGV Police Department at 9-1-1 (campus phone), Edinburg Campus or Brownsville Campus: 956-882-4911 (from a cell phone)**.
3. Notify Supervisor.
4. If injury involves student, transport student to Health Services department.

IV. Major Spill:

1. Notify persons in the immediate area about the spill, evacuating all non-essential personnel from the spill area and adjoining areas that may be impacted by vapors or a potential fire.
2. If the spilled material is flammable, turn off all potential ignition sources. Avoid breathing vapors of the spilled materials. Be aware that some materials either have no odors or create olfactory fatigue, so that you stop smelling the odor very quickly.

3. Leave on or establish exhaust ventilation if it is safe to do so. Close doors to slow down the spread of odors.
4. Call EHSRM. (Edinburg Campus: 956-665-3690 or Brownsville Campus: 956-882-5929)
5. Call the **UTRGV Police Department at 9-1-1 (campus phone), Edinburg Campus, or Brownsville Campus: 956-882-4911 (from a cell phone).**

V. Minor Spill:

1. Use a spill control kit appropriate to control material spilled, if appropriately trained to respond.
2. If the spill is minor and of known limited danger, clean up immediately. Determine the appropriate cleaning method by referring to the SDS. During clean-up, wear appropriate protective apparel. The protective clothing required will depend upon the material spilled, the amount, and the airborne concentration. At a minimum, chemical resistant gloves and goggles should be worn.
3. Cover liquid spills with compatible absorbent material such as spill pillows or a kitty litter/vermiculite mix. Be sure to check compatibility. Powdered materials should be covered with wet paper towels (if compatible) to avoid dispersal. If appropriate materials are available, corrosives should be neutralized prior to absorption. Clean spills from the outer areas first, cleaning towards the center.
4. Place the spilled material into an impervious container, seal, and contact EHSRM for disposal.
5. If appropriate, wash the affected surface with soap and water. Mop up the residues and collect for disposal.
6. A solvent, e.g. xylene, may be necessary to clean surfaces contaminated with a non-water soluble chemical. Be sure to check the solubility of the spilled material and use the least toxic effective solvent available. Be sure to wear appropriate protective equipment.

VI. Radioactive Hazard:

Refer to [The University of Texas Rio Grande Valley Radiation Safety Manual](#).

VII. Biological Hazards:

1. Do not take any action unless you have been trained to respond, except to summon assistance.
2. Attend to anyone who may have been contaminated and/or injured if it is safe to reach them. Use safety showers and eyewashes as appropriate. Call the **UTRGV Police Department at 9-1-1 (campus phone) or Edinburg Campus: 956-665-7151 or Brownsville Campus: 956-882-2222 (from a cell phone)** for assistance with injured, in case of fire, or for performing rescues. Describe the nature and the extent of the emergency; be as specific and detailed as possible.
3. If the room is equipped with ultraviolet lights, turn them on.
4. Notify persons in the immediate area about the spill. Evacuate non-essential personnel from the spill area.
5. Leave the laboratory and close all doors to prevent re-entry.

6. Notify EHSRM. (**Edinburg Campus: 956-665-3690 or Brownsville Campus: 956-882-5934**)
7. If your clothing is contaminated, remove it and place it in a properly labeled impervious container. Avoid close contact with other people to prevent additional exposures. Take a shower.
8. Put on protective clothing and equipment.
9. Wait at least 30 minutes for the aerosol to settle before entering the contaminated room. Turn off ultraviolet lights and check for visible mists in the air before entering.
10. Apply appropriate disinfectant to the spill with a gentle flooding action to avoid secondary aerosols. Allow sufficient contact times.
11. Cover excess liquids with absorbent material. Dry material should be covered with wet paper towels to avoid dispersal.
12. Place the spill clean-up material into a container and autoclave it or call EHSRM for disposal.
13. Wash the affected surface with strong disinfectant.
14. For a spill in a biological safety cabinet: clean immediately, keep the cabinet running, and use a chemical disinfectant such as bleach or alcohol and paper towels.
15. For a minimally hazardous material without aerosol, and in small volume: clean with paper towel soaked in a disinfectant.

Note: For additional information regarding biological spills refer to UTRGV – Biological Safety Manual

VIII. Building Evacuation:

1. Building evacuation may be necessary if there is a chemical release, fire, explosion, natural disaster, or medical emergency.
2. Be aware of the marked exits in your area and building.
3. The evacuation alarm is a loud continuous siren or horn.
4. To activate the building alarm system, pull the handle on one of the red boxes located in the hallway. If there is a fire, call the **UTRGV Police Department at 9-1-1 (campus phone), Edinburg Campus or Brownsville Campus: 956-882-4911 (from a cell phone)**, give your name, and describe the exact location (building name, building location, and room number) and size of the fire.
5. *Whenever* the building evacuation alarm is sounded or when you are ordered to leave by the UTRGV Police, EHSRM, or emergency response personnel, *walk quickly to the nearest marked exit* and ask others to do the same.
6. Outside, proceed to a clear reassemble area that is **at least 150 feet** from the affected building. Keep walkways clear for emergency vehicles.
7. To the best of your ability and without re-entering the building, be available to assist emergency response personnel in their attempts to determine that everyone has been evacuated safely.
8. An Emergency Command Post will be set up near the emergency site by the emergency responders. Keep clear of the post unless you have important information to report.
9. **Do not return to the building until you are told to do so by the UTRGV Police, EHSRM, or City Fire Fighting personnel.**

D. FUNDAMENTALS OF LABORATORY SAFETY

I. Procurement, Distribution and Storage:

Procurement

Before a substance is received, information on proper handling, storage, and disposal must be known to those who will be involved. Refer to the appropriate SDS for further information. No container may be accepted into a laboratory without an adequate identifying label. This label cannot be removed, defaced, or damaged in any way. All substances should be received in a central location.

Stockrooms/Storerrooms

Toxic substances must be segregated in a well-identified area with local exhaust ventilation. Chemicals which are highly toxic or other chemicals whose containers have been opened must be in unbreakable secondary containers approved for chemical storage. For example, place containers of concentrated acids or bases into plastic tubs to help contain any leakage. Stored chemicals should be examined periodically (at least annually) for replacement, deterioration, and container integrity. The labels must be checked to ensure they are still readable. If labels begin to fall off the container, secure or replace them. If a label is becoming unreadable, affix a new label to the container with the identity of the contents and health hazards. Follow Globally Harmonized System of Classification and Labeling of Chemicals (GHS).

Distribution

When chemicals are hand carried, place the container in an outside (secondary) container or bucket. Container carriers for breakable containers such as glass can be purchased through a variety of safety catalogues. These secondary containers provide protection to the bottle and help keep it from breaking. They also help to minimize spillage if the bottle does break. Freight-only elevators are to be used when transporting chemicals.

Laboratory Chemical Storage

Read the label carefully before storing a chemical. All chemicals must be stored according to the Chemical Storage Segregation Scheme. (See Appendix II) Note that this is a simplified scheme and that in some instances chemicals of the same category may be incompatible.

1. Store all chemicals by their hazard class and not in alphabetical order. Storing chemicals by alphabetical order will often result in the placement of incompatible chemicals being next to one another. Only within the segregation groups can chemicals be stored in alphabetical order. If a chemical exhibits more than one hazard, segregate by using the characteristic that exhibits the primary hazard.
2. Do not store chemicals near heat sources such as ovens or steam pipes. Also, do not store chemicals in direct sunlight.

3. Date chemicals when received and first opened. This will assist you in using the oldest chemicals first, which will also decrease the amount of chemicals for disposal. If a particular chemical can become unsafe while in storage, e.g., diethyl ether, then an expiration date should also be included. Keep in mind that expiration dates set by the manufacturer do not necessarily imply that the chemical is safe to use up to that date.
4. Do not use lab benches as permanent storage for chemicals. In these locations, the chemicals can easily be knocked over, incompatible chemicals can be stored alongside one another, and the chemicals are unprotected in the event of a fire. Each chemical must have a proper designated storage location and be returned there after use.
5. Inspect your chemicals routinely for any signs of deterioration and for the integrity of the label. State law requires that **all** chemicals must be clearly labeled. Another benefit of labeling is to prevent chemicals from becoming "unknowns."
6. Do not store any chemicals in glass containers on the floor.
7. Do not store liquid chemicals on their side.
8. Do not use fume hoods as a permanent storage location for chemicals, with the exception of particularly odorous chemicals that may require ventilation. The more containers, boxes, equipment, and other items that are stored in a fume hood, the greater the likelihood of having chemical vapors being drawn back into the room. Some chemical fume hoods have ventilated storage cabinets underneath for storage of frequently used chemicals that require ventilation.
9. Promptly contact EHSRM for the disposal of any old, expired, unused or unknown chemicals. (Edinburg Campus: 956-665-3690 or Brownsville Campus: 956-882-5929)
10. Chemicals that require refrigeration must be sealed with tight-fitting caps and kept in lab safe refrigerators. Lab safe refrigerators/freezers must be used for cold storage of these types of flammables.
11. Do not store chemicals above eye level. If the container breaks, the contents can fall onto your face and upper body.
12. Do not store excessive amounts of chemicals in the lab. Buying chemicals in large quantities creates a serious fire hazard and limits work space. The disposal costs far exceed any cost savings from large quantity purchasing.

II. Storage Cabinets:

Specific types of storage cabinets must be specified in laboratories in order to separate incompatible chemicals from one another and to safely store all chemicals. All chemicals must be stored in a secure container, preferably within enclosed cabinets.

Flammable Storage Cabinets

Flammables not in active use must be stored in safe containers inside specially designed fire resistant storage cabinets. Flammable storage cabinets must be specified for all labs that use flammable chemicals. The cabinet must meet NFPA 30 & OSHA 29 CFR 1910.106 standards. Flammable storage cabinets are designed to protect the contents from the heat and flames of external fire rather than to confine burning liquids within. They can perform their protective function only if used and maintained properly. Cabinets are generally designed with double-walled construction and doors which are two inches above the base (the cabinet is liquid-proof up to that point).

Acid Storage Cabinets

Acids should be kept in acid storage cabinets specially designed to hold them. Such cabinets have the same construction features of a flammable storage cabinet, but are coated with an epoxy enamel to guard against chemical attack, and use polyethylene trays to collect small spills and provide additional protection against corrosion of the shelves. Periodically check shelves and supports for corrosion. Nitric acid should always be stored by itself or in a separate acid cabinet compartment.

Compressed Gas Cylinder Cabinets

All compressed gas cylinders having a NFPA Health Hazard Rating of 3 or 4 (e.g. ammonia, chlorine, phosgene) and those with a Health Hazard Rating of 2 but no physiological warning properties (e.g. carbon monoxide) must be kept in a continuously, mechanically ventilated enclosure. Full size cylinders can be stored in a gas cylinder cabinet whereby smaller cylinders, e.g., lecture bottles, can be stored in a chemical fume hood, a storage cabinet under the fume hood (if ventilated), or some other ventilated enclosure. Compressed gas cylinder cabinets must meet Article 80 of the Uniform Fire Code and the following requirements: negative pressure in relation to the surrounding area with the exhaust from the cabinet going to the outside of the building, self-closing doors, and internally sprinklered or installed in a sprinklered area. Cylinders stored in compressed gas cylinder cabinets or other ventilated enclosures must be secured at all times. When stored in a cabinet or hood, lecture bottles must also be secured.

III. Personal Protective Clothing and Equipment:

It is the Principal Investigator and/or Lab Supervisor's responsibility to specify all necessary personal protective clothing for laboratory workers according to the SDS. The University is responsible for providing basic safety equipment such as gloves, eye protection and lab coats.

IV. Signs and Labels:

Prominent signs and labels of the following types should be used:

- Hazardous Material posters, including emergency contacts : Emergency Instructions outside of each area.
- Identity labels, showing contents of containers and associated hazards. Labels on all incoming chemical containers cannot be removed or defaced (unless the container is empty and ready for disposal). All secondary containers must be labeled with at least the identity of the contents, health hazards (including target organs), and manufacturer name.
- Employee notification posters describing rights under the Texas Hazard Communication Act (revised 03/2014 or most current version).
- Location signs for safety equipment, first aid equipment, and exits.
- Warning signs at areas or equipment where special or unusual hazards exist.
- Areas where food and beverage consumption and storage are permitted.

V. Records:

Laboratory supervisors should document and report any laboratory accident that results in an injury to UTRGV Police and EHSRM. Students should report to Health Services. An employee injured in a laboratory should complete a First Report of Injury or Illness form (with the assistance of his or her supervisor). These forms are available by notifying the EHSRM. Any medical records associated with a person's exposure to hazardous materials will be maintained by The University in accordance with state and federal regulations. EHSRM will also keep records whenever monitoring of hazardous materials is performed.

Safety Data Sheets (SDS) provide information on hazardous chemicals and must be readily available for all hazardous chemicals in the lab. All SDS are available by notifying the EHSRM, or visiting the EHSRM website at www.utrgv.edu/ehsrm/

VI. Medical Program:

Procedures for Authorizing Medical Treatment

It is the responsibility of every Lab Supervisor to promptly contact the EHSRM, when a suspected exposure to hazardous materials has occurred. The Lab Supervisor will provide details of exposure, including identity of the material, description of the conditions under which exposure occurred, description of signs and symptoms of the exposure, and SDS, if available. EHSRM will make an assessment to determine if there is sufficient reason to suspect that a potentially significant exposure has occurred. In the event of immediate need, medical attention should be sought prior to notification of EHSRM. When the need is not immediate, EHSRM will notify, in writing, any employee who may have had a potential exposure to hazardous materials. This notification will request that individuals involved obtain a qualified medical examination or consultation.

A medical examination or consultation for lab personnel will be made available by The University under the following circumstances:

- Whenever a lab employee develops signs or symptoms associated with a hazardous chemical to which that person may have been exposed in the laboratory;
- Where exposure monitoring reveals an exposure level routinely above the action level (or in the absence of an action level, the PEL) for an OSHA regulated substance for which there are exposure monitoring and medical surveillance requirements, medical surveillance will be established for the affected personnel as prescribed by the particular standard;
- Whenever an event takes place in the work area such as a spill, leak, explosion, or other occurrence resulting in the likelihood of a hazardous exposure, the affected personnel will be provided an opportunity for a medical consultation. The consultation will determine if there is a need for a medical exam.

All medical exams and consultations described under this Medical Program section will be performed by or under the direct supervision of a licensed physician and will be provided at The University's expense, without loss of pay and at a reasonable time and place. The arrangements for a medical consultation or exam should be made with the assistance of the EHSRM, unless it is an emergency. EHSRM will provide details of the exposure

(identity of the hazardous material, description of the conditions under which the exposure occurred, description of signs and symptoms of exposure, and the applicable SDS) and any other relevant information to the health care provider. In the event of a medical emergency, the person involved should seek medical attention by dialing 911.

Physician's Written Opinion

If a medical consultation or exam is performed, EHSRM will obtain a written opinion from the examining physician which includes the following information:

- Any recommendation for further medical follow-up;
- The results of the medical examination and any associated tests;
- Any medical condition which may be revealed in the course of the examination which may place the lab person at increased risk as a result of exposure to a hazardous chemical found in the lab; and
- A statement that the lab person has been informed by the physician of the results of the consultation or medical examination and any medical condition that may require further examination or treatment.

The written opinion cannot reveal specific findings of diagnoses unrelated to occupational exposure.

First Aid

During the school day until 7:00 PM, Health Services is available for routine care for students who are injured or ill. Contact **Health Services Edinburg Campus at 956-665-2511 or Brownsville Campus at 956-882-3896**. If care is needed that Health Services cannot provide or when Health Services is closed, or for non-student lab personnel, call the UTRGV Police Department at 9-1-1 (campus phone), Edinburg Campus or Brownsville Campus: 956-882-4911 (from a cell phone). For after-hours medical advice, call the **24/7 Nurse Advice Line toll-free at 855-810-4457**.

During the school day until 5:00 PM, UT Health RGV Employee Health Edinburg is available for routine care for employees who are injured or ill. Contact **UT Health RGV Employee Health Edinburg 956-296-1731 or Harlingen 956-296-1343**. If care is needed that UT Health RGV Employee Health cannot provide or when UT Health RGV Employee Health is closed, or for non-student lab personnel, call the UTRGV Police Department at 9-1-1 (campus phone), Edinburg Campus or Brownsville Campus: 956-882-4911 (from a cell phone). For after-hours medical advice, call the **24/7 Nurse Advice Line toll-free at 855-810-4457**.

VII. Information and Training Program:

The University requires that all individuals that work in a laboratory are adequately informed about the physical and health hazards present in the laboratory, the known risks, and what to do if an accident occurs.

Every laboratory worker must be trained to know the location and proper use of available

personal protective clothing and equipment The laboratory supervisor is responsible for providing information to his or her personnel about any hazards present in the lab. This information must be provided at the time of a lab person's initial assignment and prior to any assignments involving new potential chemical exposure situations. The following lists the information that should be provided by the lab supervisor:

- The location and availability of this manual.
- Work area-specific training for all new personnel as described in the UTRGV Hazard Communication Program;
- The OSHA Occupational Exposure to Hazardous Chemicals in Laboratories (See Appendix V)
- The location and availability of known reference material on the hazards, safe handling, storage, and disposal of hazardous chemicals found in the laboratory including, but not limited to, Safety Data Sheets (SDS) received from the chemical supplier.
- When there is likelihood that a person may exceed the limit, the permissible exposure limits (PEL) for OSHA regulated substances or recommended exposure limits (for example, TLV) for other hazardous chemicals where there is no applicable OSHA standard.
- Signs and symptoms associated with exposures to hazardous chemicals used in the laboratory.
- Methods and observations that may be used to detect the presence or release of a hazardous chemical (such as visual appearance or odor of hazardous chemicals when being released).
- The physical and health hazards of chemicals in the work area.
- The measures lab personnel can take to protect themselves from these hazards, including specific procedures the lab supervisor and/or EHSRM have implemented to protect personnel from exposures to hazardous chemicals, such as appropriate work practices, emergency procedures, and personal protective equipment to be used.
- The applicable details of this manual.

Employees must be re-trained when new chemical hazards are introduced into their workplace, or when new hazards are shown on updated Safety Data Sheets (SDS), as well as upon reassignment to different workplaces that involve new chemical hazards or protective measures.

The following hazardous materials training is offered by EHSRM and is required for all students, staff, and faculty that engage in laboratory activities:

Hazard Communication/Laboratory Safety Training

Training in accordance with the University of Texas-Rio Grande Valley Laboratory Safety Program is required for all employees of The University, including faculty, staff, students and volunteers who have the potential for exposure to hazardous chemicals. **Any work in a laboratory using hazardous chemicals meets the definition of the requirement.** EHSRM offers this training online through Blackboard under the organization "Environmental Health, Safety and Risk Management" and in person on a regular schedule and can arrange special sessions with advanced notice. Training is required before the employee can be assigned work in or around hazardous chemicals. Annual refreshers may be required.

The training takes approximately one hour and includes:

- Central requirements of the Hazard Communication Act, including Training, Chemical Labels, and Safety Data Sheets (SDS)
- Spill Clean-Up Procedures
- Chemical Disposal Procedures
- Chemical Storage Guidelines
- Hazards Specific to Different Chemical Groups

VIII. Chemical Waste Disposal Program:

Chemical wastes are regulated by the Environmental Protection Agency under the Resource Conservation and Recovery Act and its amendments. Laboratory Supervisors are responsible for advising laboratory workers on how to handle all wastes generated in laboratory operations.

General

All chemical waste streams generated from experiments conducted in UTRGV laboratories are considered to be regulated and are required to be disposed of through the EHSRM. Drain disposal of any chemicals is strictly prohibited without approval from the EHSRM. Because of the complexity of the regulations, personal discretion on the part of laboratory personnel are not allowed when making a decision regarding the disposal of any waste streams. When washing containers that have held regulated waste, the first two(2) washes are required to be placed into hazardous waste containers.

Chemical Waste Containers

Containers used for the accumulation of hazardous waste must be in good condition, free of leaks, and compatible with the waste being stored in them. A waste accumulation container should be opened only when it is necessary to add waste, and should otherwise be capped. Hazardous waste must not be placed in unwashed containers that previously held an incompatible material. It is recommended that waste be placed in its original container.

If a container holding hazardous waste is not in good condition or if it begins to leak, transfer the waste from this container into a container that is in good condition, pack the container in a larger and non-leaking container, or manage the waste in some other way that prevents the potential for a release or contamination. Contact EHSRM if assistance is required. (Edinburg Campus: 956-665-3690 or Brownsville Campus: 956-882-5929)

A storage container holding a hazardous waste that is incompatible with any waste or other materials stored nearby must be separated from the other materials or protected by means of a partition, wall, or other secondary containment.

All Waste Containers

- Must be marked with the words "WASTE" or "SPENT" and their contents indicated. No container should be marked with the words "hazardous" or "non-hazardous." Paint over or remove any old labels.
- Must be kept at or near (immediate vicinity) the site of generation and under control of the generator and in secondary containment.
- Must be compatible with contents (i.e. acid should not be stored in metal cans).
- Must be closed at all times except when actively receiving waste. Do not leave funnels

in waste containers. Remove the funnel after the waste has been poured into the container.

- Must be properly identified with completed waste tags before pick-up is requested.
- Must be safe for transport with non-leaking, screw-on caps.
- Must be filled to a safe level (not beyond the bottom of the neck of the container or a 2-inch head space for 55 gallon drums). **Report any waste that is $\frac{3}{4}$ full to EHSRM via waste@utrgv.edu**

Note: Do not use red biohazard bags or sharps containers for hazardous chemical waste collection.

Accumulation of Chemical Waste

A generator of potentially hazardous waste may accumulate up to a total of 55 gallons of waste, which may be determined to be hazardous by Environmental Health, Safety and Risk Management or one quart of acutely hazardous waste (see Appendix VII, Acutely Hazardous Waste) at or near the point of generation. If a process will generate more than this volume at one time, EHSRM must be contacted in advance to arrange a special waste pick-up. Hazardous waste in excess of 55 gallons CANNOT be stored at your site for more than three (3) days. Therefore, EHSRM requires advanced notice of generation in order to determine if the waste meets the definition of hazardous and to arrange for prompt removal.

It is essential that the generator keep different hazardous wastes separate so that disposal options remain clear and more cost effective. In all cases, do not mix incompatible wastes or other materials in the same container or place wastes in an unwashed container that previously held an incompatible waste or material.

Labeling Chemical Waste Containers for Pick-Up

Before chemical waste can be picked up by EHSRM, a waste tag must be attached to the chemical and should include at least the following information:

- **Chemical name(s) with the words waste.**
- **Hazard class**
- **The date the container was filled.**

Submitting Requests for Disposal of Chemical Waste

When a chemical waste container is ready for disposal and is properly tagged, the laboratory supervisor should contact EHSRM at waste@utrgv.edu

Waste is collected on Mondays, Wednesdays, and Fridays. Indicate if you need a replacement container.

IX. Inspections:

EHSRM inspects all UTRGV laboratories. Research laboratories are inspected on an annual. Laboratories with BSL-2 agents are inspected on a Bi-Annual basis. Teaching

laboratories are inspected on an annual basis. In addition, teaching laboratory activities inspections are conducted on a semester basis. Weekly walk-throughs are also performed in addition to the annual or bi-annual inspections. Principal investigators are expected to comply with the specific mandates within 30 days of the inspection.

X. Consequences of Non-Compliance:

If a laboratory is not in compliance with the safe operating procedures as outlined in this manual, **EHSRM has the authority to close the laboratory until violations are corrected.** **Approval of the Dean is not required.**

Employees of The University are responsible for ensuring that they follow the procedures and faithfully implement the policies and appropriate responsibilities stated in this Laboratory Safety Manual. Failure to do so is a serious breach of University policy and subject to disciplinary action that might include termination of employment at The University.

References:

CRC Handbook of Laboratory Safety, Third Edition. A. K. Furr, Ed. Chemical Rubber Company. 1990. (704 page reference on all aspects of lab safety.)

Employee Training Guide, Texas Hazard Communication Act. Texas Department of Health.

Safe Storage and Handling of Laboratory Chemicals - A Review of Safe Storage and Handling Practices for Laboratory Chemicals. Nancy Magnussen. Texas A&M University Chemistry Safety Coordinator.

Texas A&M University Hazard Communication Handbook. Texas A&M University Safety and Health Office. Jan. 1986.

E. BASIC RULES AND PROCEDURES FOR WORKING WITH CHEMICALS

I. General Rules:

Laboratory Protocol

Everyone in the lab is responsible for his or her own safety and for the safety of others. Before starting any work in the lab, make it a point to become familiar with the procedures and equipment that are to be used. Work only with chemical products when you know their flammability, reactivity, toxicity, safe handling, storage, and emergency procedures. If you don't understand or are unclear about something, ASK! The following guidelines are recommended for working safely in a lab:

Personal Safety Practices

1. Lab coats and safety glasses are required in laboratories employing chemicals, biohazards, or radioisotopes. Never wear shorts, sandals, or open-toed shoes in lab.
2. Do not allow children or pets in laboratories.
3. Never pipette anything by mouth.
4. Be aware of dangling jewelry, loose clothing, or long hair that might get caught in equipment.
5. Store food and drinks in refrigerators that are designated for that use only.
6. Never work alone in a lab if it is avoidable. If you must work alone, make someone aware of your location and have them call or check on you periodically.
7. Wash your hands frequently throughout the day and before leaving the lab.
8. Do not wear lab coats, gloves, or other personal protective clothing out of the lab and into non-lab areas. This clothing may have become contaminated and you could spread the contamination.
9. Contact lenses should not be worn in a lab because chemicals or particulates can get caught behind them and cause severe damage to the eye.

Housekeeping

1. Work areas must be kept clean and free of unnecessary chemicals. Clean your work area throughout the day and before you leave at the end of the day.
2. If necessary, clean equipment after use to avoid the possibility of contaminating the next person who uses it.
3. Keep all aisles and walkways in the lab clear to provide a safe walking surface and an unobstructed exit.

Accidents and Spills

See Section C, Emergency Procedures for detailed procedures.

Supplies for cleaning up a minor chemical or biological spill must be readily available. In case of release, promptly clean up spills using appropriate protective apparel and equipment.

Spill Response Equipment

- Supplies for a chemical spill include an inert absorbent such as kitty litter or vermiculite or a 50/50 mixture of the two, a plastic (non-sparking) scoop, plastic bags for the spilled material, heavy gloves, goggles, and sodium bicarbonate to neutralize acids. Kits are commercially available which include acid, base, flammable, or universal pillows or booms.
- Supplies for a biological spill include paper towels and a fresh 1:10 bleach solution.

Note: All spent spill clean-up materials must be disposed of in the same manner as the spilled chemical.

Avoidance of "Routine" Exposure

Develop and encourage safe habits; avoid unnecessary exposure to chemicals by any route. Do not smell or taste chemicals. Vent apparatus which may discharge toxic chemicals (e.g., vacuum pumps, distillation columns) into local exhaust devices. Inspect gloves and test glove boxes before use. Do not allow release of toxic substances in cold rooms or warm rooms, since these have contained recirculated atmospheres.

Choice of Chemicals

Use only those chemicals for which the quality of the available ventilation system is appropriate as determined by the laboratory supervisor or principal investigator.

Eating, Drinking, Smoking, etc.

Do not eat, drink, chew gum, or apply cosmetics in areas where laboratory chemicals are present; wash hands before conducting these activities. Smoking is not allowed in University buildings.

Equipment and Glassware

EHSRM recommends the following guidelines for the use and care of glassware and other laboratory equipment.

Glassware:

1. Inspect all glassware before use. Repair or discard any broken, cracked, or chipped glassware.
2. Tape or shield glass vacuum vessels to prevent flying glass in the case of an implosion. Also, tape or shield glass vacuum desiccators.
3. Do not use household Thermos bottles as a substitute for laboratory Dewar flasks; the walls are too thin.
4. Transport all glass chemical containers in rubber or polyethylene bottle carriers.
5. Fire-polish all cut glass tubing and rods before use.
6. Practice the following when inserting glass tubes or rods into stoppers:
 - a. Be certain that the diameter of the tube is compatible with the diameter of the stopper,

- b. Fire-polish the end of the glass tube,
- c. Lubricate the glass with water or glycerol,
- d. Wear heavy gloves and hold the glass not more than two inches from the end to be inserted,
- e. Insert the glass carefully with a twisting motion, and
- f. Remove stuck tubes by slitting the stopper with a sharp knife.

Assembly of Laboratory Apparatus:

1. Keep work surfaces as uncluttered as possible.
2. Firmly clamp apparatus and set up away from the edge of the lab bench.
3. Only use equipment that is free from cracks, chips, or other defects.
4. If possible, place a pan under a reaction vessel or other container to contain liquid if the glassware breaks.
5. Do not allow burners or any other ignition sources nearby when working with flammable liquids.
6. Lubricate glass stopcocks.
7. Properly support and secure condensers and water hoses with clamps and wires. Be sure to direct the water hoses so that any drips that may come off the hoses do not splash onto any electrical wires.
8. Position apparatus that is attached to a ring stand with the center of gravity over the base and not to one side.
9. Assemble the apparatus so that burners or baths can be removed quickly.
10. Use an appropriate vapor trap and confine the setup to a fume hood if there is a possibility of hazardous vapors being evolved.
11. Put the setup in a fume hood whenever conducting a reaction that could result in an implosion or explosion. Keep the sash pulled down. If it is not possible to use a fume hood, use a standing shield that is stabilized and secured.
12. Always wear a lab coat and proper eye and face protection.

Centrifuges:

1. Securely anchor tabletop centrifuges and place in a location where the vibration will not cause bottles to fall off the bench.
2. Keep the centrifuge lid closed while operating and do not leave the centrifuge until you are certain it is running safely without vibration.
3. If the centrifuge starts vibrating, stop and check the load balances.
4. Regularly clean rotors and buckets with a non-corrosive cleaning solution.
5. Use sealed safety cups while centrifuging hazardous materials.

Ultraviolet Lamps:

1. Wear ultraviolet absorbing safety glasses while working with ultraviolet light.
2. Protect your skin from potential burns due to ultraviolet light.
3. Shield any experiment in which ultraviolet light is used to prevent escape of the direct beam or scattered radiation.

Lasers:

1. Always wear goggles that protect against the specific wavelength of the laser.
2. Never look directly at the beam.
3. Do not allow any reflective materials in or along the beam.
4. Post warning signs in all laser areas. If possible, use a flashing light at the lab entrance to indicate when a laser is in use.
5. Contact EHSRM Laser Safety Officer for more information.

Separatory Funnels:

1. Use extreme caution if the temperature of the materials is elevated.
2. When a volatile solvent is used, swirl the unstoppered separatory funnel first to allow some solvent to vaporize and to release pressure.
3. Close the funnel and invert it with the stopper held in place, then immediately open the stopcock to release pressure.
4. Do not vent the separatory funnel near a flame or any other ignition source and do not point it at a co-worker or equipment. It is best to vent the separatory funnel into a fume hood.
5. Close the stopcock, swirl the funnel, then immediately open the stopcock with the funnel in an inverted position to vent the vapors again.

Cooling Baths and Cold Traps:

1. Always use caution when working with cryogenic coolants.
2. Use temperature resistant gloves and a faceshield while slowly immersing an object to be cooled.
3. Do not pour cold liquid onto the edge of a glass Dewar flask when filling because the flask may break and implode.
4. Never lower your head into a dry ice chest; no oxygen is present.
5. Wear temperature resistant gloves while handling dry ice. If no protection is used, severe burns can result.

Vacuum Pumps:

1. If possible, vent vacuum pump exhaust into a fume hood.
2. Guard all belt-driven vacuum pumps to prevent hands or loose clothing from getting caught in the belt pulley.
3. Place a trap between the vacuum pump and the apparatus.
4. Lubricate pump regularly if possible. Check belt conditions and do not operate in a fume hood cabinet that is used for storage of flammables.

Electrical:

1. Examine all electrical cords periodically for signs of wear and damage. If damaged electrical cords are discovered, unplug the equipment and send it off for repair.
2. Properly ground all electrical equipment.
3. If sparks are noticed while plugging or unplugging equipment or if the cord feels

- hot, do not use the equipment until it can be serviced by an electrician.
4. Do not run electrical cords along the floor where they will be a tripping hazard and be subject to wear. If a cord must be run along the floor, protect it with a cord cover.
 5. Do not run electrical cords above the ceiling. The cord must be visible at all times to ensure it is in good condition.
 6. Do not plug too many items into a single outlet. Cords that enable you to plug more than one item in at a time should not be used. Multi-plug strips can be used if they are protected with a circuit breaker and if they are not overused.
 7. Do not use extension cords for permanent wiring. If you must use extension cords throughout the lab, then it is time to have additional outlets installed.

II. Personal Protection:

The most important thing to remember about protective clothing is that it only protects you if you wear it. Safety Data Sheets or other references should be consulted for information on the type of protective clothing required for the particular work you are performing.

Protective Eyewear

- Goggles provide the best all-around protection against chemical splashes, vapors, dusts, and mists.
- Goggles that have indirect vents or are non-vented provide the most protection, but an anti-fog agent may need to be applied.
- Standard safety glasses provide protection against impact.
- If using a laser, wear safety glasses or goggles, which provide protection against the specific wavelength of that laser.
- Remember, prescription glasses do not provide adequate protection in a laboratory setting. Prescription safety glasses can be purchased from most opticians.
- Contact lenses should not be worn in a laboratory because they can trap contaminants behind them and reduce or eliminate the effectiveness of flushing with water from an eyewash. Contact lenses may also increase the amount of chemicals trapped on the surface of the eye and decrease removal of the chemical by tearing. If it is necessary to wear contact lenses in a lab, wear protective goggles at all times.

Protective Gloves

- Any glove can be permeated by chemicals. The rate at which this occurs depends on the composition of the glove, the chemicals present and their concentration, and the exposure time to the glove. If you are not certain which type of glove provides you with the protection you need, contact the manufacturer and ask for specifics on that glove.
- If direct chemical contact occurs, replace gloves regularly throughout the day. Wash hands regularly and remove gloves before answering the telephone or opening doors to prevent the spread of contamination.
- Check gloves for cracks, tears, and holes.
- Butyl, neoprene, and nitrile gloves are resistant to most chemicals, e.g., alcohols, aldehydes, ketones, most inorganic acids, and most caustics.
- Disposable latex and vinyl gloves protect against some chemicals, most aqueous solutions, and microorganisms and reduce risk of product contamination.

Note: There is increasing evidence that some people develop a serious allergic reaction to latex.

- Leather and some knit gloves will protect against cuts, abrasions, and scratches, but not against chemicals.
- Temperature-resistant gloves protect against cryogenic liquids, flames, and high temperatures.

Other Protective Clothing

- The primary purpose of a lab coat is to protect against splashes and spills. A lab coat should be nonflammable, where necessary, and should be easily removed. Many different kinds of lab coats are available.
 - Rubber coated aprons can be worn to protect against chemical splashes and may be worn over a lab coat for additional protection.
 - Face shields can protect against impact, dust, particulates, and chemical splashes for the face, eyes, and throat. However, always wear protective eyewear such as goggles underneath a face shield because a face shield only offers additional protection to the eyes. Chemical vapors and splashes can still travel under and around a face shield. If scratches or cracks are noticed in the face shield, replace the window.
 - Shoes which fully cover the feet should always be worn in a lab. If work is going to be performed that includes moving large and heavy objects such as 55 gallon drums, steel-toed shoes must be worn.
- In general, the lab supervisor must ensure that appropriate personal protective equipment is worn by all persons, including visitors, in areas where chemicals are stored or handled.

III. Planning:

Seek information and advice about hazards, write appropriate protective procedures, and plan positioning or equipment before beginning any new operation.

IV. Unattended Operations:

Leave lights on, place an appropriate sign on the door, and provide for containment of toxic substances in the event of failure of a utility service (such as cooling water) to an unattended operation.

V. Fume Hood:

Use the hood for all procedures which might result in the release of hazardous chemical vapors or dust. Confirm that the hood is working before use by holding a Kimwipe® or other lightweight paper up to the opening of the hood. The paper should be pulled inward. Leave the hood "on" when it is not in active use if toxic substances are stored inside or if it is uncertain whether adequate general laboratory ventilation will be maintained when it is "off."

VI. Proper Use of Fume Hoods:

1. Equipment and other materials should be placed at least six inches behind the sash. This will reduce the exposure of personnel to chemical vapors that may escape into the lab due to air turbulence.
2. When the hood is not in use, pull the sash all the way down. While personnel are working at the hood, pull down the sash as far as is practical. The sash is your protection against fires, explosions, chemical splashes, and projectiles.
3. Do not keep loose papers, paper towels, or tissues (e.g., Kimwipes[®]) in the hood. These materials can be drawn into the blower and adversely affect the performance of the hood.
4. Do not use a fume hood as a storage cabinet for chemicals. Excessive storage of chemicals and other items will disrupt the designed airflow in the hood. In particular, do not store chemicals against the baffle at the back of the hood, because this will interfere with the laminar airflow across the hood.
5. If large equipment must be kept in a fume hood, raise it 1.5 inches off the work surface to allow air to flow underneath. This dramatically reduces the turbulence within the hood and increases its efficiency.
6. Do not place objects directly in front of a fume hood (such as refrigerators or lab coats hanging on the manual controls) as this can disrupt the airflow and draw contaminants out of the hood.
7. Keep in mind that modifications made to a fume hood system, e.g., adding a snorkel, can render the entire system ineffective.
8. Minimize the amount of foot traffic immediately in front of a hood. Walking past hoods causes turbulence that can draw contaminants out of the hood and into the room.

VII. Storage of Chemicals in the Laboratory:

Please refer to section D: Fundamentals of Laboratory Safety, sub category I:

VIII. Working with Allergens:

A wide variety of substances can illicit skin and lung hypersensitivity. Examples include common substances such as diazomethane, chromium, nickel, bichromates, formaldehyde, isocyanates, and certain phenols. Because of this variety and the varying response of individuals, suitable gloves should be used whenever there is potential for contact with chemicals that may cause skin irritation.

IX. Working with Embryotoxins:

Embryotoxins are substances that act during pregnancy to cause adverse effects on the developing fetus. These effects may include embryoletality (death of the fertilized egg, the embryo, or the fetus), malformations (teratogenic effects), retarded growth, and postnatal function deficits.

A few substances have been demonstrated to be embryotoxic in humans. These include:

acrylic acid	diphenylamine	nitrobenzene
aniline	estradiol	nitrous oxide
benzene	formaldehyde	phenol
cadmium	formamide	thalidomide
carbon disulfide	hexachlorobenzene	toluene
N,N-dimethylacetamide	iodoacetic acid	vinyl chloride
dimethylformamide	lead compounds	xylene
dimethyl sulfoxide	mercury compounds	polychlorinated biphenyls
polybrominated biphenyls		

Maternal alcoholism is probably the leading known cause of embryotoxic effects in humans, but the exposure to ethanol typically encountered in laboratories is unlikely to be embryotoxic. Many substances, some as common as sodium chloride, have been shown to be embryotoxic to animals at some exposure level, but usually this is at a considerably higher level than is met in the course of normal laboratory work. However, some substances do require special controls due to embryotoxic properties. One common example is **formaldehyde**: women of childbearing potential should handle this substance only in a hood and should take precautions to avoid skin contact with the liquid because of the ease with which it passes through the skin.

Because the period of greatest susceptibility to embryotoxins is the first 8-12 weeks of pregnancy, which includes a period when a woman may not know that she is pregnant, women of childbearing potential should take care to avoid skin contact with all chemicals. The following procedures are recommended to be followed routinely by women of childbearing potential in working with chemicals requiring special control because of embryotoxic properties:

1. Each use must be reviewed for particular hazards by the Principal Investigator or Lab Supervisor, who will decide whether special procedures are warranted or whether warning signs should be posted. Consultation with appropriate safety personnel may be desirable. In cases of continued use of a known embryotoxin, the operation should be reviewed annually or whenever a change in procedures is made.
2. Embryotoxins requiring special control should be stored in an adequately ventilated area. The container should be labeled in a clear manner such as the following:
EMBRYOTOXIN: READ SPECIFIC PROCEDURES FOR USE. If the storage container is breakable, it should be kept in an impermeable, unbreakable secondary container having sufficient capacity to retain the material, should the primary container fail.
3. Women of childbearing potential should take adequate precautions to guard against spills and splashes. Operations should be carried out using impermeable containers and in adequately ventilated areas. Appropriate safety apparel, especially gloves, should be worn. All hoods, glove boxes, or other essential engineering controls should be operating at required efficiency before work is started.
4. Supervisors must be notified regarding all incidents of exposure or spills of embryotoxins requiring special control. A qualified physician should be consulted about any exposures of women of childbearing potential above the acceptable level (i.e., any skin contact or inhalation exposures).

X. Working with Chemicals of Moderate Chronic or High Acute Toxicity:

Before beginning a laboratory operation, each worker is strongly advised to consult one of the standard compilations that list toxic properties of known substances and learn what is known about the substance to be used. The precautions and procedures described in this section should be followed if any of the substances to be used in significant quantities is known to be moderately or highly toxic (if any of the substances being used is known to be highly toxic, it is desirable that two people be present in the area at all times).

These procedures should also be followed if the toxicological properties of any of the substances being used or prepared are unknown. If any of the substances to be used or prepared are known to have high, chronic toxicity (e.g., compounds of heavy metals and other potent carcinogens), then the precautions and procedures described below should be supplemented with additional precautions to aid in containing and ultimately destroying the substances having high chronic toxicity. Some examples of potent carcinogens (substances known to have high chronic toxicity), along with their corresponding chemical class, are:

Alkylating Agents

α -halo ethers

bis(chloromethyl) ether

methyl chloromethyl ether

epichlorohydrin aziridines

styrene oxide

2-methylaziridine

diazo, azo, and azoxy compounds

4-dimethylaminoazobenzene

ethyl methanesulfonate

methyl methanesulfonate

methyl trifluoromethanesulfonate

1,3-propanesultone

1,4-butanedioldimethanesulfonate

epoxides

ethylene oxide

diepoxybutane

propylene oxide

ethylene imine

sulfonates

diethyl sulfate

dimethyl sulfate

electrophilic alkenes and alkynes

acrylonitrile

acrolein

ethyl acrylate

Acylating Agents

β -propiolactone

β -butyrolactone dimethylcarbamoyl chloride

Organohalogen Compounds

1,2-dibromo-3-chloropropane

vinyl chloride

chloroform

1,4-dichlorobenzene

bis(2-chloroethyl) sulfide

carbon tetrachloride

hexachlorobenzene methyl iodide

2,4,6-trichlorophenol

Natural Products

adriamycin
reserpine progesterone

aflatoxins bleomycin
safrole

Inorganic Compounds

cisplatin

Aromatic Amines

4-aminobiphenyl
o-toluidine

benzidine aniline
o-anisidine

The overall objective of the procedures outlined in this section is to minimize exposure of the laboratory worker to toxic substances by taking all reasonable precautions. Thus, the general precautions outlined in Section E.1 should normally be followed whenever a toxic substance is being transferred from one container to another or is being subjected to some chemical or physical manipulation. The following three precautions should always be followed:

1. Protect the hands and forearms by wearing either gloves and a laboratory coat or suitable long gloves to avoid contact of the toxic material with the skin.
2. Procedures involving volatile toxic substances and those involving solid or liquid toxic substances that may result in the generation of aerosols should be conducted in a hood or other suitable containment device.
3. After working with toxic materials, wash the hands and arms immediately. Never eat, drink, chew gum, apply cosmetics, take medicine, or store foods in areas where toxic substances are being used.

These standard precautions will provide laboratory workers with good protection from most toxic substances. In addition, records that include amounts of material used and names of workers involved should be kept as part of the laboratory notebook record of the experiment. To minimize hazards from accidental breakage of apparatus or spills of toxic substances in the hood, containers of such substances should be stored in pans or trays made of polyethylene or other chemically resistant material and apparatus should be mounted above trays of the same type of material. Alternatively, the working surface of the hood can be fitted with a removable liner of adsorbent plastic-backed paper. Such procedures will contain spilled toxic substances in a pan, tray, or absorbent liner and greatly simplifies subsequent cleanup and disposal. Vapors that are discharged from the apparatus should be trapped or condensed to avoid adding substantial amounts of toxic vapor to the hood exhaust air. Areas where toxic substances are being used and stored must have restricted access, and warning signs should be posted if a special toxicity hazard exists.

General waste disposal procedures must be followed for these types of chemicals. In general, the waste materials and solvents containing toxic substances should be stored in closed, impervious containers so that personnel handling the containers will not be exposed to their contents.

The laboratory worker must be prepared for potential accidents or spills involving toxic substances. If a toxic substance contacts the skin, the area should be washed with water. If there is a major spill outside of the hood, the room or appropriate area should be evacuated and necessary measures should be taken to prevent exposure of other workers. Spills must be cleaned by personnel wearing suitable personal protective apparel. If a spill of a significant quantity of toxic material occurs outside the hood, an air-supplied full-face respirator should be worn.

In addition to the precautions described in this section, we strongly advise researchers to develop written standard operating procedures intended to establish a concise, step-by-step method for carrying out routine laboratory operations with the substance in question.

XI. Working with Substances of High Chronic Toxicity:

All of the procedures and precautions described in the previous section should be followed when working with substances known to have high chronic toxicity. In addition, when such substances are to be used in quantities exceeding a few milligrams to a few grams, depending on the hazards posed by the particular substance, the additional precautions described in this section should be used. Each laboratory worker's plan for experimental work and for disposing of waste materials must be approved by the laboratory supervisor. Consultation with EHSRM may be appropriate to ensure that the toxic material is effectively contained during the experiment and that waste materials are disposed of in a safe manner. Substances in this high chronic toxicity category include certain heavy metal compounds (e.g., dimethylmercury and nickel carbonyl) and compounds normally classified as strong carcinogens. Examples of compounds normally classified as strong carcinogens include the following:

2-acetylaminofluorene
aflatoxin B₁
benzo[a]pyrene
bis(chloromethyl) ether
7,12-dimethylbenz[a]anthracene
dimethylcarbamoyl chloride

hexamethylphosphoramide
3-methylcholanthrene
2-nitronaphthalene
propane sultone
various N-nitrosamides
various N-nitrosamines

An accurate record of the amounts of such substances being stored and the amounts used, dates of use, and names of users must be maintained.

It is appropriate to keep such records as part of the record of experimental work in the laboratory workers' research notebook, but it must be understood that the research supervisor is responsible for ensuring that accurate records are maintained.

Any volatile substances having high chronic toxicity must be stored in a ventilated storage area in a secondary tray or container having sufficient capacity to contain the material should the primary storage container fail. All containers of substances in this category must have labels that identify that contents and include a warning such as: **WARNING! HIGH CHRONIC TOXICITY OR CANCER SUSPECT AGENT**. Storage areas for substances in this category must have limited access, and special signs should be posted if a special

toxicity hazard exists. Any area used for storage of substances of high chronic toxicity must be maintained under negative pressure with respect to the surroundings.

All experiments with and transfers of such substances or mixtures containing such substances must be done in a controlled area (i.e., a laboratory, or a portion of a laboratory, or a facility such as an exhaust hood or a glove box that is designated for the use of highly toxic substances. Its use need not be restricted to the handling of highly toxic substances if all personnel who have access to the controlled area are aware of the nature of the substances being used and the precautions that are necessary). When a glove box is used, the ventilation rate in the box must be at least two volume changes per hour, the pressure should be at least 0.5 inches of water lower than that of the surrounding environment, and the exit gases should be passed through a trap or HEPA filter.

Positive pressure glove boxes are normally used to provide an inert anhydrous atmosphere. If these glove boxes are used with highly toxic compounds, then the box should be thoroughly checked for leaks before use and the exit gases should be passed through a suitable trap or filter. Laboratory vacuum pumps used with substances having high chronic toxicity should be protected by high-efficiency scrubbers or HEPA filters and vented into an exhaust hood. Motor-driven vacuum pumps are recommended because they are easy to decontaminate.

Proper gloves must be worn when transferring or otherwise handling substances or solutions of substances having high chronic toxicity. In some cases, the laboratory worker or the research supervisor may deem it advisable to use other protective apparel, such as an apron of reduced permeability covered by a disposable coat. Extreme precautions such as these might be taken, for example, when handling large amounts of certain heavy metals and their derivatives or compounds known to be potent carcinogens. Surfaces on which high chronic toxicity substances are handled must be protected from contamination by using chemically resistant trays or pans that can be decontaminated after the experiment or by using dry, absorbent plastic-backed paper that can be disposed of after use.

Normal laboratory work must not be resumed in a space that has been used as a controlled area until it has been adequately decontaminated. Work surfaces must be thoroughly washed and rinsed. If experiments have involved the use of finely divided solid materials, dry sweeping should not be done. In such cases, surfaces must be cleaned by wet mopping or by use of a vacuum cleaner equipped with a HEPA filter. All equipment (e.g., glassware, vacuum pumps, and containers) that is known or suspected to have been in contact with substances of high chronic toxicity should be washed and rinsed before it is removed from the controlled area. In the event of continued experimentation with a substance of high chronic toxicity (i.e., if a worker regularly uses toxicologically significant quantities of such a substance at least three times a week), a qualified physician must be consulted to determine whether it is advisable to establish a regular schedule of medical surveillance or biological monitoring.

In addition to the precautions described in this section, lab supervisors must develop written standard operating procedures intended to establish a concise, step-by-step method for carrying out routine laboratory operations with the substance in question. These procedures must be approved by the respective departmental chemical hygiene officer.

References:

CRC Handbook of Laboratory Safety, Third Edition. A. K. Furr, Ed. Chemical Rubber Company. 1990. (704 page reference on all aspects of lab safety.)

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F. SAFE PROCEDURES FOR SPECIFIC CLASSES OF HAZARDOUS MATERIALS

The specific rules and procedures for working with hazardous chemicals, as outlined in the preceding section, give insight into the proper methods for handling materials which pose significant hazards due primarily to their chronic toxicity. However, these specific rules and procedures, along with the general rules for working with chemicals, do not address some of the basic physical hazards which may stem from acute exposure to different types of laboratory chemicals. This section offers some specific guidelines for working with common laboratory chemicals that, for varying reasons, are acutely toxic in the sense that they may cause considerable harm to human life and health pending short-term exposures. This section will address five fundamental classes of laboratory chemicals: flammables, corrosives, oxidizers, reactives, and compressed gases. These classes of chemicals may include chemicals that are also covered in the previous section regarding their property of toxicity.

I. Flammable Solvents:

General Characteristics

Flammable liquids are indeed the most common chemicals found in a laboratory. The primary hazard associated with flammable liquids is, of course, their ability to readily ignite and burn. One should note that it is the vapor of a flammable liquid, not the liquid itself, that ignites and causes a fire.

1. The rate at which a liquid vaporizes is a function of its *vapor pressure*. In general, liquids with high vapor pressures evaporate at a higher rate compared to liquids of lower vapor pressure. It should be noted that the vapor pressure increases rapidly as the temperature is raised as does the evaporation rate. A reduced-pressure environment also accelerates the rate of evaporation.
2. The *flash point* of a liquid is the lowest temperature at which a liquid gives off vapor at such a rate as to form an air:vapor mixture that will ignite, but will not sustain ignition. Many commonly used flammable solvents have flashpoints significantly lower than room temperature:

<u>Compound</u>	<u>Flash Point (°C)</u>
diethyl ether	- 45.0
acetone	- 17.8
isopropyl alcohol	11.7

3. The *limits of flammability or explosivity* define the range of fuel:air mixtures that will sustain combustion. The lower limit of this range is called *the Lower Explosive Limit* or LEL, and the higher limit of this range is called the *Upper Explosive Limit* or UEL. Materials with very broad flammability ranges (e.g., acetylene, LEL = 3%, UEL = 65%) are particularly treacherous due to the fact that virtually any fuel:air combination may form an explosive atmosphere.
4. The *vapor density* of a flammable material is the density (mass to volume ratio) of the corresponding vapor relative to air under specific temperature and pressure conditions. Flammable vapors with densities greater than unity (and thus "heavier" than air) are potentially lethal because they will accumulate at floor level and flow, with remarkable ease, in much the same manner that a liquid would. The obvious threat is that these mobile vapors may eventually reach an ignition source, such as an electrical outlet or a Bunsen burner at another student's bench.

Examples of Flammable Liquids

acetone
ethyl ether toluene
methyl formate

Use and Storage of Flammables

- Flammable liquids that are not in active use must be stored in safe containers inside fire resistant storage cabinets designed for flammables, or inside storage rooms.
- Minimize the amount of flammable liquids stored in the lab.
- Use flammables only in areas free of ignition sources. Remember, smoking is not permitted inside any University building.
- The transfer of material to or from a metal container is generally accompanied by an accumulation of static charge on the container. This fact must be kept in mind when transferring flammable liquids, since the discharge of this static charge could generate a spark, thereby igniting the liquid. To make these transfers safer, flammable liquid dispensing and receiving containers must be bonded together before pouring. Large containers such as drums must also be grounded when used as dispensing or receiving vessels. All grounding and bonding connections must be metal to metal. (The aforementioned bonding and grounding wires may be found in most lab safety catalogs.)
- Never heat flammables with an open flame. Instead, use steam baths, water baths, oil baths, hot air baths, sand baths or heating mantles.
- Never store flammable chemicals in a standard household refrigerator. There are several ignition sources located inside a standard refrigerator that can set off a fire or violent explosion. Flammables can only be stored cold in a lab safe or explosion-proof refrigerator. Another alternative is to use an ice bath to chill the chemicals. Remember, there is no safety benefit in storing a flammable chemical in a refrigerator if the flashpoint of that chemical is below the temperature of the refrigerator.

Health Hazards Associated with Flammables

In general, the vapors of many flammables are irritating to mucous membranes of the respiratory system and eyes, and in high concentrations are narcotic. The following symptoms are typical for the respective routes of entry.

Acute Health Effects

Inhalation - headache, fatigue, dizziness, drowsiness, narcosis (stupor and unresponsiveness)

Ingestion - slight gastro-intestinal irritation, dizziness, fatigue

Skin Contact - dry, cracked, and chapped skin

Eye Contact - stinging, watering eyes, and inflammation of the eyelids

Chronic Health Effects

The chronic health effects will vary depending on the specific chemical, the duration of the exposure, and the extent of the exposure. However, **damage to the lungs, liver, kidneys, heart and/or central nervous system** may occur. **Cancer** and **reproductive effects** are also possible.

Flammable Groups Exhibiting Similar Health Effects

Hydrocarbons - aliphatic hydrocarbons are narcotic, but their systemic toxicity is relatively low. Aromatic hydrocarbons are all potent narcotic agents and overexposure to the vapors can lead to loss of muscular coordination, collapse, and unconsciousness. Benzene is toxic to bone marrow and can cause leukemia.

Alcohols - vapors only moderately narcotic.

Ethers - exhibit strong narcotic properties but for the most part are only moderately toxic.

Esters - vapors may result in irritation to the eyes, nose, and upper respiratory tract.

Ketones - systemic toxicity is generally not high.

First Aid Procedures for Exposures to Flammable Materials

Inhalation Exposures - remove person from the contaminated area if it is safe to do so. Get medical attention and do not leave person unattended.

Ingestion Exposures - remove the person, if possible, from the source of contamination. Get medical attention.

Dermal Exposures - remove person from source of contamination. Remove clothing, jewelry, and shoes from the affected areas. Flush the affected area with water for at least 15 minutes and obtain medical attention.

Eye Contact - remove person from the source of contamination. Flush the eyes with large amounts of water for at least 15 minutes. Seek medical attention.

Personal Protective Equipment

Always use a fume hood while working with flammable liquids. Nitrile and neoprene gloves are effective against most flammables. Wear a non-flammable lab coat to provide a barrier to your skin and goggles if splashing is likely to occur.

II. Oxidizers:

General Characteristics

1. Oxidizers or oxidizing agents present fire and explosion hazards on contact with combustible materials. Depending on the class, an oxidizing material may increase the burning rate of combustibles with which it comes in contact; cause the spontaneous ignition of combustibles with which it comes in contact; or undergo an explosive reaction when exposed to heat, shock, or friction.
2. Oxidizers are generally corrosive.

Examples of Common Oxidizers

Peroxides
perchlorates
hypochlorites

nitrates
chlorates
dichromates

nitrites
chlorites

Use and Storage of Oxidizers

1. In general, store oxidizers away from flammables, organic compounds, and combustible materials.
2. Strong oxidizing agents like chromic acid should be stored in glass or some other inert container, preferably unbreakable. Corks and rubber stoppers should not be used.
3. Reaction vessels containing appreciable amounts of oxidizing material should never be heated in oil baths, but rather on a heating mantle or sand bath.

Use and Storage of Perchloric Acid

Perchloric acid is an oxidizing agent of particular concern. The oxidizing power of perchloric acid increases with an increase in concentration and with an increase in temperature. Cold, 70% perchloric acid is a strong, non-oxidizing corrosive. A 72% perchloric acid solution at elevated temperatures is a strong oxidizing agent. An 85% perchloric acid solution is a strong oxidizer at room temperature.

- Do not attempt to heat perchloric acid if you do not have access to a properly functioning perchloric acid fume hood. Perchloric acid can only be heated in a hood specially equipped with a wash down system to remove any perchloric acid residue. The hood should be washed down after each use and it is preferred to dedicate the hood to perchloric acid use only.
- Whenever possible, substitute a less hazardous chemical for perchloric acid.
- Perchloric acid can be stored in a perchloric acid fume hood. Keep only the minimum amount necessary for your work. Another acceptable storage site for perchloric acid is on a metal shelf or in a metal cabinet away from organic or flammable materials. A bottle of perchloric acid should also be stored in a glass secondary container to contain leakage.
- Do not allow perchloric acid to come in contact with any strong dehydrating agents such as sulfuric acid. The dehydration of perchloric acid is a severe fire and explosion

hazard.

- Do not order or use anhydrous perchloric acid. It is unstable at room temperature and can decompose spontaneously with a severe explosion. Anhydrous perchloric acid will explode upon contact with wood.

Health Hazards Associated with Oxidizers

Oxidizers are covered here primarily due to their potential to add to the severity of a fire or to initiate a fire. But there are some generalizations that can be made regarding the health hazards of an oxidizing material. In general, oxidizers are corrosive and many are highly toxic.

Acute Health Effects

Some oxidizers such as nitric and sulfuric acid vapors, chlorine, and hydrogen peroxide act as irritant gases. All irritant gases can cause inflammation in the surface layer of tissues when in direct contact. They can also cause irritation of the upper airways, conjunctiva, and throat. Some oxidizers, such as **fluorine**, can cause severe burns of the skin and mucus membranes.

Chlorine trifluoride is extremely toxic and can cause severe burns to tissue.

Nitrogen trioxide is very damaging to tissue, especially the respiratory tract. The symptoms from an exposure to nitrogen trioxide may be delayed for hours, but fatal pulmonary edema may result.

Osmium tetroxide, another oxidant commonly employed in the laboratory, is also dangerous due to its high degree of acute toxicity. It is a severe irritant of both the eyes and the respiratory tract. Inhalation can cause headache, coughing, dizziness, lung damage, difficulty breathing and may be fatal. Osmium tetroxide is regarded by many in the field as having "poor warning properties." This is due to the fact that it is difficult to detect in the atmosphere (by smell or other means). The OSHA-defined Permissible Exposure Limit for osmium tetroxide is 0.0002 ppm, while its odor threshold is 2 ppm - this means that one could conceivably be exposed to osmium tetroxide at concentrations 10,000 times the PEL without knowing it. For this reason, it is recommended that laboratories using osmium tetroxide have necessary safeguards in place before the container is even opened.

Chronic Health Effects

Nitrobenzene and **chromium compounds** can cause hematological and neurological changes. **Compounds of chromium and manganese** can cause liver and kidney disease. **Chromium (VI) compounds** have been associated with lung cancer.

First Aid

In general, if a person has inhaled, ingested, or come into direct contact with these materials, the person must be removed from the source of contamination as quickly as possible when it is safe to do so. Medical help must be summoned. In the case of an exposure directly to the

skin or eyes it is imperative that the exposed person be taken to an emergency shower or eyewash immediately. Flush the affected area for a minimum of 15 minutes, then get medical attention. Call 9-1-1.

Personal Protective Equipment

In many cases, the glove of choice will be neoprene, polyvinyl chloride (PVC), or nitrile. Be sure to consult a glove compatibility chart to ensure the glove material is appropriate for the particular chemical with which you will be working.

Goggles must be worn if the potential for splashing exists or if exposure to vapor or gas is likely.

Always use these materials in a chemical fume hood as most pose a hazard via inhalation. Cylinders of compressed gases should be kept in ventilated cabinets.

III. Corrosives:

General Characteristics

1. Corrosives are most commonly acids and alkalis, but many other materials can be severely damaging to living tissue.
2. Corrosives can cause visible destruction or irreversible alterations at the site of contact. Inhalation of the vapor or mist can cause severe bronchial irritation. Corrosives are particularly damaging to the skin and eyes.
3. Certain substances considered non-corrosive in their natural dry state are corrosive when wet such as when in contact with moist skin or mucus membranes. An example of these materials are lithium chloride, halogen fluorides, and allyl iodide.
4. Sulfuric acid is a very strong dehydrating agent and nitric acid is a strong oxidizing agent. Dehydrating agents can cause severe burns to the eyes due to their affinity for water.

Examples of Corrosives

sulfuric acid
bifluoride chromic acid
stannic chloride

ammonium
bromine
ammonium hydroxide

Use and Storage of Corrosives

1. Always store acids separately from bases. Also, store acids in acid storage cabinets away from flammables since many acids are also strong oxidizers.
2. Do not work with corrosives unless an emergency shower and continuous flow eyewash are available.
3. Add acid to water, but never add water to acid. This is to prevent splashing from the acid due to the generation of excessive heat as the two substances mix.

4. Never store corrosives above eye level. Store on a low shelf or cabinet.
5. It is a good practice to store corrosives in a tray or bucket to contain any leakage.
6. When possible, purchase corrosives in containers that are coated with a protective plastic film that will minimize the danger to personnel if the container is dropped.
7. Store corrosives in a wooden cabinet or one that has a corrosion-resistant lining.
8. Corrosives stored in an ordinary metal cabinet will quickly damage it. If the cabinet supports that hold up the shelves become corroded, the result could be serious. Acids should be stored in acid storage cabinets specially designed to hold them and Nitric acid should be stored in a separate cabinet or compartment

Use and Storage of Hydrofluoric Acid

Hydrofluoric acid is extremely hazardous and deserves special mention. Hydrofluoric acid can cause severe burns and inhalation of anhydrous hydrogen fluoride can be fatal. Initial skin contact with hydrofluoric acid may not produce any symptoms.

1. Only persons fully trained in the hazards of hydrofluoric acid should use it.
2. Always use hydrofluoric acid in a properly functioning fume hood. Be sure to wear personal protective clothing!
3. If you suspect that you have come in direct contact with hydrofluoric acid: wash the area with water for at least 15 minutes, remove clothing, then promptly seek medical attention. If hydrogen fluoride vapors are inhaled, move the person immediately to an uncontaminated atmosphere (if safe to do so), keep the person warm, and seek prompt medical attention.
4. Never store hydrofluoric acid in a glass container because it is incompatible with glass.
5. Store hydrofluoric acid separately in an acid storage cabinet and keep only that amount necessary in the lab.
6. Creams for treatment of hydrofluoric acid exposure are commercially available.

Health Hazards Associated with Corrosives

All corrosives possess the property of being severely damaging to living tissues and also attack other materials such as metal.

Skin contact with alkali metal hydroxides, e.g., sodium hydroxide and potassium hydroxide, is more dangerous than with strong acids. Contact with alkali metal hydroxides normally causes deeper tissue damage because there is less pain than with an acid exposure. The exposed person may not wash it off thoroughly enough or seek prompt medical attention.

All hydrogen halides are acids that are serious respiratory irritants and also cause severe burns. Hydrofluoric acid is particularly dangerous. At low concentrations, hydrofluoric acid does not immediately show any signs or symptoms upon contact with skin. It may take several hours for the hydrofluoric acid to penetrate the skin before you would notice a burning sensation. However, by this time permanent damage, such as second and third degree burns with scarring, can result.

Acute Health Effects:

Inhalation - irritation of mucus membranes, difficulty in breathing, fits of coughing, pulmonary edema

Ingestion - irritation and burning sensation of lips, mouth, and throat; pain in swallowing; swelling of the throat; painful abdominal cramps; vomiting; shock; risk of perforation of the stomach

Skin Contact - burning, redness and swelling, painful blisters, profound damage to tissues, and with alkalis; a slippery, soapy feeling

Eye Contact - stinging, watering of eyes, swelling of eyelids, intense pain, ulceration of eyes, loss of eyes or eyesight

Chronic Health Effects:

Symptoms associated with a chronic exposure vary greatly depending on the chemical. For example, the chronic effect of hydrochloric acid is damage to the teeth; the chronic effects of hydrofluoric acid are decreased bone density, fluorosis, and anemia; the chronic effects of sodium hydroxide are unknown.

First Aid

Inhalation - remove person from source of contamination if safe to do so. Get medical attention. Keep person warm and quiet and do not leave unattended.

Ingestion - remove person from source of contamination. Get medical attention and inform emergency responders of the name of the chemical swallowed.

Skin Contact - remove person from source of contamination and take immediately to an emergency shower or source of water. Remove clothing, shoes, socks, and jewelry from affected areas as quickly as possible, cutting them off if necessary. Be careful not to get any chemical on your skin or to inhale the vapors. Flush the affected area with water for a minimum of 15 minutes. Get medical attention.

Eye Contact - remove person from source of contamination and take immediately to an eyewash or source of water. Rinse the eyes for a minimum of 15 minutes. Have the person look up and down and from side to side. Get medical attention. Do not let the person rub their eyes or keep them tightly shut.

Personal Protective Equipment

Always wear the proper gloves when working with acids. Neoprene and nitrile gloves are effective against most acids and bases. Polyvinyl chloride (PVC) is also effective for most acids. A rubber coated apron and goggles should also be worn. If splashing is likely to occur, wear a face shield over the goggles. Always use corrosives in a chemical fume hood.

IV. Reactives

General Characteristics

Polymerization Reactions:

Polymerization is a chemical reaction in which two or more molecules of a substance combine to form repeating structural units of the original molecule. This can result in an extremely high or uncontrolled release of heat. An example of a chemical which can undergo a polymerization reaction is styrene.

Water Reactive Materials:

1. When water reactive materials come in contact with water, one or more of the following can occur: liberation of heat which may cause ignition of the chemical itself if it is flammable, or ignition of flammables that are stored nearby; release of a flammable, toxic, or strong oxidizing gas; release of metal oxide fumes; and formation of corrosive acids.
2. Water reactive chemicals can be particularly hazardous to firefighting personnel responding to a fire in a lab, because water is the most commonly used fire extinguishing medium. Examples of water reactive materials:

alkali metals
lithium, sodium, potassium
magnesium
aluminum

silanes
alkylaluminums
zinc

Pyrophorics:

Pyrophoric materials can ignite spontaneously in the presence of air.
Examples of pyrophoric materials:

diethylzinc triethylaluminum
many organometallic compounds

Peroxide-Forming Materials:

Peroxides are very unstable and some chemicals that can form them are commonly used in laboratories. This makes peroxide-forming materials some of the most hazardous substances found in a lab. Peroxide-forming materials are chemicals that react with air, moisture, or impurities to form peroxides. The tendency to form peroxides by most of these materials is greatly increased by evaporation or distillation. Organic peroxides are extremely sensitive to shock, sparks, heat, friction, impact, and light. Many peroxides formed from materials used in laboratories are more shock sensitive than TNT. Just the friction from unscrewing the cap of a container of an ether that has peroxides in it can provide enough energy to cause a severe explosion.

Examples of peroxide-forming materials (the first group listed is the most hazardous):

diisopropyl ether	divinylacetylene
sodium amide	potassium amide
dioxane	diethyl ether
tetrahydrofuran	vinyl ethers
butadiene	vinylpyridine
acrylonitrile	styrene

Note: See Appendix IX, Peroxide Forming Chemicals.

Other Shock-Sensitive Materials:

These materials are explosive and sensitive to heat and shock.

Examples of shock-sensitive materials:

chemicals containing nitro groups fulminates
hydrogen peroxide (30% +)
ammonium perchlorate
benzoyl peroxide (when dry)
Compounds containing the functional groups: acetylide, azide, diazo, halamine, nitroso, and ozonide.

Note: See Appendix VIII, Potentially Explosive Chemicals.

Use and Storage of Reactives

1. A good way to reduce the potential risks is to minimize the amount of material used in the experiment. Use only the amount of material necessary to achieve the desired results.
2. Always substitute a less hazardous chemical for a highly reactive chemical whenever possible. If it is necessary to use a highly reactive chemical, order only the amount that is necessary for the work.

Water Reactive Materials:

Store water-reactive chemicals in an isolated part of the lab. A cabinet far removed from any water sources, such as sinks, emergency showers, and chillers, is an appropriate location. Clearly label the cabinet "Water-Reactive Chemicals - No Water".

Pyrophorics:

Store pyrophorics in an isolated part of the lab and in a clearly marked cabinet. Be sure to routinely check the integrity of the container and have the material disposed of through the EHSRM if the container is corroded or otherwise damaged.

Peroxide-Forming Materials:

1. Do not open the chemical container if peroxide formation is suspected. The act of opening the container could be sufficient to cause a severe explosion. Visually inspect liquid peroxide-forming materials for crystals or unusual viscosity before opening. Pay special attention to the area around the cap. Peroxides usually form upon evaporation, so they will most likely be formed on the threads under the cap.
2. Date all peroxide forming materials with the date received, and the expected shelf life. Chemicals such as diisopropyl ether, divinyl acetylene, sodium amide, and vinylidene chloride should be discarded after three months. Chemicals such as dioxane, diethyl ether, and tetrahydrofuran should be submitted to EHSRM for disposal after one year.
3. Store all peroxide-forming materials away from heat, sunlight, and sources of ignition. Sunlight accelerates the formation of peroxides.
4. Secure the lids and caps on these containers to discourage the evaporation and concentration of these chemicals.
5. Never store peroxide-forming materials in glass containers with screw cap lids or glass stoppers. Friction and grinding must be avoided. Also, never store these chemicals in a clear glass bottle where they would be exposed to light.
6. Contamination of an ether by peroxides or hydroperoxides can be detected simply by mixing the ether with 10% (wt/wt) aqueous potassium iodide solution - a yellow color change due to the oxidation of iodide to iodine confirms the presence of peroxides. Small amounts of peroxides can be removed from contaminated ethers via distillation from lithium aluminum hydride (LiAlH₄-), which both reduces the peroxide and removes contaminating water and alcohols. However, if you suspect that peroxides may be present, it would be wise to call EHSRM for disposal. If you notice crystal formation in the container or around the cap, do not attempt to open or move the container. Call EHSRM for proper disposal.
7. Never distill an ether unless it is known to be free of peroxides.

Other Shock Sensitive Materials:

Store these materials separately from other chemicals and in a clearly labeled cabinet. Never allow picric acid to dry out, as it is extremely explosive. Always store picric acid in a wetted state.

Health Hazards Associated with Reactives

Reactive chemicals are grouped as a category primarily because of the safety hazards associated with their use and storage and not because of similar acute or chronic health effects. For health hazard information on specific reactive materials consult the SDS, the manufacturer, or EHSRM. However, there are some hazards common to the use of reactive materials. Injuries can occur due to heat or flames, inhalation of fumes, vapors, and reaction products, and flying debris.

First Aid

If someone is seriously injured the most important step to take is to contact emergency responders as quickly as possible. This is best accomplished by directly calling **The University of Texas-Rio Grande Valley Police Department at 911 from an office phone, Edinburg Campus or Brownsville Campus (956-882-4911) from a cell phone**. Explain the situation and describe the location clearly and accurately.

If someone is severely bleeding, apply a sterile dressing, clean cloth, or handkerchief to the wound. Then put protective gloves on and place the palm of your hand directly over the wound and apply pressure and keep the person calm. Continue to apply pressure until help arrives.

If a person's clothes are on fire, he or she should drop immediately to the floor and roll. If a fire blanket is available, put it over the individual. An emergency shower, if one is immediately available, can also be used to douse flames.

If a person goes into shock, have the individual lie down on their back if safe to do so and raise the feet about one foot above the floor.

Personal Protective Equipment

Wear appropriate personal protective clothing while working with highly reactive materials. This might include: impact resistant safety glasses or goggles, a face shield, gloves, a lab coat (to minimize injuries from flying glass or an explosive flash), and a shield. Conduct work within a chemical fume hood as much as possible and pull down the sash as far as is practical. While the experiment does not require you to reach into the fume hood, keep the sash closed.

Barriers can offer protection of personnel against explosions and should be used. Many safety catalogs offer commercial shields which are commonly polycarbonate and are weighted at the bottom for stability. It may be necessary to secure the shields firmly to the work surface.

V. Compressed Gas Cylinders

General Characteristics:

1. Cylinders of compressed gases can pose a chemical as well as a physical hazard. (See Appendix X, Compressed Gas Reference Table)
2. If the valve were to break off a cylinder, the amount of force present could propel the cylinder through a brick wall. For example, a cylinder of compressed breathing air used by SCUBA divers has the explosive force of 1 1/2 pounds of TNT.

Use and Storage

1. Whenever possible, use flammable and reactive gases in a fume hood or other ventilated enclosure. As noted in Chapter D.2., concerning storage cabinets, certain categories of toxic gases must always be stored and used in ventilated enclosures.
2. Always use the appropriate regulator on a cylinder. If a regulator will not fit a cylinder's valve, replace the cylinder, not the regulator. Do not attempt to adapt or modify a regulator to fit a cylinder it was not designed for. Regulators are designed to fit only specific cylinder valves to avoid improper use.
3. Inspect regulators, pressure relief devices, valves, cylinder connections, and hose lines frequently for damage.
4. Never use a cylinder that cannot be positively identified. Color coding is not a reliable way of identifying a cylinder because the colors can vary from supplier to supplier.

5. Do not use oil or grease on any cylinder component of an oxidizing gas because a fire or explosion can result.
6. Never transfer gases from one cylinder to another. The gas may be incompatible with the residual gas remaining in the cylinder or may be incompatible with the cylinder material.
7. Never completely empty cylinders during lab operations; rather, leave approximately 25 PSI of pressure. This will prevent any residual gas in the cylinder from becoming contaminated. However, if the cylinder is non-returnable, call EHSRM for instructions. If inert, you will be asked to vent the remainder of the gas; if not inert, you may need to react it off.
8. Place all cylinders so that the main valve is always accessible.
9. Close the main cylinder valve whenever the cylinder is not in use.
10. Remove regulators from unused cylinders and always put the safety cap in place to protect the valve.
11. Always secure cylinders, whether empty or full, to prevent them from falling over and damaging the valve (or falling on your foot). Secure cylinders by chaining or strapping them to a wall, lab bench, or other fixed support.
12. Oxygen should be stored in an area that is at least 20 feet away from any flammable or combustible materials or separated from them by a non-combustible barrier at least 5 feet high and having a fire-resistance rating of at least 1/2 hour.
13. To transport a cylinder, put on the safety cap and strap the cylinder to a hand truck in an upright position. Never roll a cylinder.
14. Always clearly mark empty cylinders and store them separately.
15. Be careful while handling compressed gas cylinders and never drop or strike a cylinder against anything.
16. Use only wrenches or other tools supplied by the cylinder supplier to open a valve. Open cylinder valves slowly.
17. Only compatible gases should be stored together in a gas cylinder cabinet.
18. Flammable gases must be stored in properly labeled, secured areas away from possible ignition sources and kept separate from oxidizing gases.
19. Do not store compressed gas cylinders in areas where the temperature can exceed 125F.

Purchase Policy

UTRGV requires that all gas cylinders purchased for use on campus must be returnable to the vendor. The only exception to this policy is for a compelling research reason. The original policy indicated such exceptions would require prior approval and that a \$1,000 deposit would be required to cover potential disposal costs. The specific procedures to be followed to request permission to purchase a research gas in a non-returnable gas cylinder are outlined below.

The Principal Investigator (PI) should prepare a request for an exception and include the reason why a non-returnable gas cylinder purchase is essential. This request must contain a Letter of Credit commitment that specifically states the requesting PI will be responsible for the proper disposal of the non-returnable cylinder and agrees to pay a \$1,000 disposal fee if the University is required to dispose of the cylinder. This request should be submitted to the department chair and the dean for review and approval. The request should then be forwarded to the VP for Business Affairs for final action. Please note: identification of a specific account or funding source by the PI for the possible \$1,000 disposal expenditure is not required but approval by the department and the college constitutes a commitment by

them that, if necessary, department or college funds are available to cover any required disposal costs if the PI is unable to cover these costs.

A copy of the approval request will be returned to the PI and a copy will be forwarded to the Environmental Health, Safety and Risk Management (EHSRM). The PI should attach a copy of the approved request to the purchase order used to obtain the desired gas.

Final disposal of the non-returnable gas cylinder should be completed no later than three years after purchase unless written approval for an extension is obtained from the VP for Business Affairs upon recommendation of the chair and dean. Evidence of the proper disposal of the cylinder must be provided to EHSRM. If the cylinder is disposed of through normal channels (e.g. the EHSRM Hazardous Waste Program) at no extra cost to The University, the \$1,000 Letter of Credit commitment will be canceled. The cylinder will be acceptable for normal waste disposal if the valve has been removed from the cylinder and the cylinder has been cleaned. Similarly, if the cylinder has been returned to the manufacturer or distributor, and this is verified in the form of a receipt or a bill of lading, the Letter of Credit commitment will be canceled. If however, The University must dispose of the cylinder outside of normal procedures because of the cylinder's condition, e.g. damaged or corroded valve, the disposal fee of \$1,000 will be assessed to the PI and it is the responsibility of the PI to provide an appropriate account for this charge at that time.

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G. BIOLOGICAL HAZARDS AND CONTROL

Personnel who work in biological laboratories may handle infectious agents in addition to other hazards such as chemicals and radioactive materials. Over the years, there have been many documented cases of lab personnel acquiring diseases due to their work with infectious agents. Only approximately 20% of these cases have been attributed to a specific incident, with the rest assumed to be related to work practices in the lab, primarily the creation of aerosols. Whenever work with infectious agents is performed, all appropriate steps must be taken to protect personnel and the environment.

I. Recommended Laboratory Practices to Prevent Exposure

There are basically four routes of exposure or four ways in which a person can come in contact with infectious agents. These routes are contact with the skin or mucus membranes, ingestion, inhalation, and inoculation. Each of these routes of exposure is discussed below.

How to Prevent Contact with Skin or Mucus Membranes

Spilled material can come into direct contact with the skin as can droplets produced by pipetting, removal of screw caps, and vortex mixing of unsealed tubes.

1. The control of a contact exposure is accomplished through the wearing of appropriate protective clothing such as a face shield, gloves, safety glasses, a mask, and laboratory coats. Other ways to control contact exposure include using absorbent paper on the work bench, performing all procedures carefully, and frequently wiping work surfaces with a disinfectant.
2. Keep all non-essential items away from the area where work is being performed to protect personal items from contamination. All contaminated wastes must be handled and stored properly to prevent contact exposure of lab personnel as well as housekeeping staff and waste handlers.

How to Prevent Accidental Ingestion

Ingestion may occur either directly or indirectly. Exposure may occur from mouth pipetting or splashing from a container into the mouth or by contaminating the hands and then touching the mouth or items such as a coffee cup, food, or lip balm, that go into the mouth.

The control of an ingestion exposure is accomplished through the use of mechanical pipetting devices *whenever* pipetting and by practicing good personal hygiene, such as washing hands frequently throughout the day and not eating or drinking in the work area. Food items also cannot be stored in refrigerators that contain hazardous materials or in the lab where work with infectious agents is being performed.

How to Prevent Inhalation

It is generally known that aerosols are the primary means by which infectious diseases are

spread and contracted. An aerosol can be either a liquid or a dry particle. An aerosol with a diameter of five microns or less can easily be inhaled and carried to the alveoli of the lungs. These aerosols can remain airborne for a long period of time and can spread wide distances, especially after entering the ventilation system. Particles with a diameter larger than five microns tend to settle rapidly and can contaminate the skin or other surfaces.

There are many commonly performed procedures in the lab that can create aerosols. Examples include centrifuging, heating inoculating loops, using a blender, blowing out the last drop in a pipette, and changing animal bedding.

The control of inhalation exposure is accomplished by a combination of using the appropriate safety equipment such as biological safety cabinets and by performing procedures carefully to minimize the creation of aerosols. Refer to Section 3 of this chapter for additional information regarding Laboratory Equipment.

How to Prevent Inoculation

Inoculation in a lab usually occurs with a needle and syringe. Exercise extreme caution whenever using a needle. Restrict needle use; whenever an alternative to a needle is possible, it should be used. Inoculation can also occur through animal bites and other sharps such as Pasteur pipettes and razor blades.

The control of an inoculation hazard is accomplished by the safe use, handling, and storage of needles and other sharps. **After using a needle, do not recap, bend, break, remove it from the syringe, or manipulate it in any way.** Many people have been accidentally stuck with a needle during the process of recapping it. The needle and other sharps should simply be placed into a sharps container to prevent any injuries. **Call EHSRM Edinburg Campus: 956-665-3690 or Brownsville Campus: 956-882-5929 for sharps containers.**

II. Biosafety Levels

The Centers for Disease Control (CDC) and the National Institutes of Health (NIH) have developed standard procedures providing protection against biological hazards. The publication *Biosafety in Microbiological and Biomedical Laboratories* provides specific descriptions of combinations of microbiological practices, laboratory facilities, and safety equipment, and recommends their use in four biosafety levels of operation with infectious agents. These biosafety levels are described below.

The biosafety levels described in the *NIH Guidelines for Research Involving rDNA Molecules* are based on and consistent with the biosafety levels presented here. A biosafety level (BSL) is based on the potential hazard of the agent and the functions of the lab. BSL1 is for work with agents that pose the least hazard and BSL4 is for work with agents that pose the greatest hazard. Only BSL1 through 3 are included here because there are no BSL4 labs at The University. Included are examples of organisms which fall within a particular biosafety level. Keep in mind that the biosafety level used for a particular organism may change based on the procedures being performed and the amount of cultures involved.

All work with microbiological agents at The University should follow the CDC/NIH guidelines. Research labs conducting work with microbiological agents should, at a minimum, follow the guidelines for BSL2. Instructional labs working with microbiological agents must always

follow the BSL1 guidelines at a minimum. If you are uncertain at which biosafety level your work should be performed, call EHSRM (Edinburg Campus: 956-665-3690 or Brownsville Campus: 956-882-5929) for assistance.

Biosafety Level 1

BSL1 is suitable for work involving well-characterized agents not known to cause disease in healthy adult humans, and of minimal potential hazard to laboratory personnel and the environment.

Examples of BSL1 Agents:

<i>Bacillus subtilis</i>	<i>Naegleria gruberi</i>
Infectious canine Hepatitis	Hepatitis Virus

The laboratory is not necessarily separated from the general traffic patterns in the building. Work is generally conducted on open bench tops using standard microbiological practices. Special containment equipment or facility design is not required nor generally used. Laboratory personnel have specific training in the procedures conducted in the laboratory and are supervised by a scientist with general training in microbiology or a related science. The following standard and special practices, safety equipment, and facilities apply to agents assigned to BSL1:

Standard Microbiological Practices (BSL1):

1. Access to the laboratory is limited or restricted at the discretion of the laboratory director when experiments or work with cultures and specimens is in progress.
2. Persons wash their hands after they handle viable materials and animals, after removing gloves, and before leaving the laboratory.
3. Eating, drinking, handling contact lenses, and applying cosmetics are not permitted in work areas where there is reasonable likelihood of exposure to potentially infectious materials. Persons who wear contact lenses in laboratories should also wear goggles or a face shield. Food is stored outside the work area in cabinets or refrigerators labeled and used for this purpose only.
4. Mouth pipetting is prohibited; mechanical pipetting devices are used.
5. All procedures are performed carefully to minimize the creation of splashes or aerosols.
6. Work surfaces are decontaminated at least once a day and after any spill of viable material.
7. All cultures, stocks, and other regulated wastes are decontaminated before disposal by an approved decontamination method, such as autoclaving. Materials to be decontaminated outside of the immediate laboratory are to be placed in a durable, leak- proof container and closed for transport from the laboratory. Materials to be decontaminated off-site from the laboratory are packaged in accordance with applicable local, state, and federal regulations before removal from the facility.
8. An insect and rodent control program is in effect.

Special Practices (BSL1): None.

Safety Equipment (Primary Barriers) (BSL1):

1. Special containment devices or equipment such as a biological safety cabinet are generally not required for manipulations of agents assigned to BSL1.
2. It is recommended that laboratory coats, gowns, or uniforms be worn to prevent contamination or soiling of street clothes.
3. Gloves should be worn if the skin on the hands is broken or if a rash exists.
4. Protective eyewear should be worn for anticipated splashes of microorganisms or other hazardous materials to the face.

Laboratory Facilities (Secondary Barriers) (BSL1):

1. Each laboratory contains a sink for handwashing.
2. The laboratory is designed so that it can be easily cleaned. Rugs in laboratories are not appropriate and should not be used because proper decontamination following a spill is extremely difficult to achieve.
3. Bench tops are impervious to water and resistant to acids, alkalis, organic solvents, and moderate heat.
4. Laboratory furniture is sturdy. Spaces between benches, cabinets, and equipment are accessible for cleaning.
5. If the laboratory has windows that open, they are fitted with fly-proof screens.

Biosafety Level 2

BSL2 is similar to Level 1 and is suitable for work involving agents of moderate potential hazard to personnel and the environment.

Examples of BSL2 Agents:

Bordetella pertussis
Clostridium tetani
Shigella spp.
Human blood

Cryptococcus neoformans
Mycobacterium leprae
Human Immunodeficiency Virus

It *differs* in that (1) laboratory personnel have specific training in handling pathogenic agents and are directed by scientists, (2) access to the laboratory is limited when work is being conducted, (3) extreme precautions are taken with contaminated sharp items, and (4) certain procedures in which infectious aerosols or splashes may be created are conducted in biological safety cabinets or other physical containment equipment. *In addition to all the requirements for BSL1*, work at BSL2 requires:

Special Practices (BSL2):

1. Access to the laboratory is limited or restricted by the laboratory director when work with infectious agents is in progress. In general, persons who are at increased risk of acquiring infection or for whom infection may be unusually hazardous are not allowed in the laboratory or animal rooms. For example, persons who are immunocompromised or immunosuppressed may be at risk of acquiring infections. The laboratory director has

final responsibility for assessing each circumstance and determining who may enter or work in the laboratory.

2. The laboratory director establishes policies and procedures whereby only persons who have been advised of the potential hazard and meet specific entry requirements (e.g., immunization) enter the laboratory or animal rooms.
3. When the infectious agent(s) in use in the laboratory require special provisions for entry (e.g., immunization), a hazard warning sign incorporating the universal biohazard symbol is posted on the access door to the laboratory work area. The hazard warning sign identifies the infectious agent, lists the name and telephone number of the laboratory director or other responsible person(s), and indicates the special requirement(s) for entering the laboratory.
4. Laboratory personnel receive appropriate immunizations or tests for the agents handled or potentially present in the laboratory (e.g., hepatitis B vaccine or TB skin testing).
5. When appropriate, baseline serum samples for laboratory and other at-risk personnel are collected and stored. Additional serum specimens may be collected periodically, depending on the agents handled or the function of the facility.
6. A site-specific biosafety manual is prepared or adopted in addition to this Laboratory Safety manual. Personnel are advised of special hazards and are required to read and follow instructions on practices and procedures.
7. Laboratory personnel receive appropriate training on the potential hazards associated with the work involved, the necessary precautions to prevent exposures, and the exposure evaluation procedures. Personnel receive annual updates, or additional training as necessary for procedural or policy changes.
8. A high degree of precaution must always be taken with any contaminated sharp item, including needles and syringes, slides, pipettes, capillary tubes, and scalpels. Needles and syringes or other sharp instruments should be restricted for use only when there is no alternative, such as parenteral injection, phlebotomy, or aspiration of fluids from laboratory animals and diaphragm bottles. Plasticware should be substituted for glassware whenever possible.
 - a. Only needle-locking syringes or disposable syringe-needle units (i.e., needle is integral to the syringe) are used for the injection or aspiration of infectious materials. Used disposable needles must not be bent, sheared, broken, recapped, removed from disposable syringes, or otherwise manipulated by hand before disposal; rather, they must be carefully placed in conveniently located puncture-resistant containers used for sharps disposal.

These sharps containers are provided and removed by EHSRM. Non-disposable sharps must be placed in a hard-walled container for transport to a processing area for decontamination, preferably by autoclaving.
 - b. Syringes that re-sheath the needle, needle-less systems, and other safe devices should be used when appropriate.
 - c. Broken glassware must not be handled directly by hand, but must be removed by mechanical means such as a brush and dustpan, tongs, or forceps.
9. Cultures, tissues, or specimens of body fluids are placed in a container that prevents leakage during collection, handling, processing, storage, transport, or shipping.
10. Laboratory equipment and work surfaces should be decontaminated with an appropriate disinfectant on a routine basis, after work with infectious materials is finished, and especially after overt spills, splashes, or other contamination by infectious materials. Contaminated equipment must be decontaminated according to any local, state, or federal regulations before it is sent for repair or maintenance or packaged for transport in accordance with applicable local, state, or federal

- regulations, before removal from the facility.
11. Spills and accidents which result in overt exposures to infectious materials are immediately reported to the laboratory director. Medical evaluation, surveillance, and treatment are provided as appropriate and written records are maintained (see Medical Program, Chapter D of this manual).
 12. Animals not involved in work being performed are not permitted in the laboratory.

Safety Equipment (Primary Barriers) (BSL2):

1. Properly maintained biological safety cabinets, preferably Class II, or other appropriate personal protective equipment or physical containment devices are used whenever:
 - a. Procedures with a potential for creating infectious aerosols or splashes are conducted. These include centrifuging, grinding, blending, vigorous shaking or mixing, sonic disruption, opening containers of infectious materials whose internal pressures may be different from ambient pressures, inoculating animals intranasally, and harvesting infected tissues from animals or eggs.
 - b. High concentrations or large volumes of infectious agents are used. Such materials may be centrifuged in the open laboratory if sealed rotor heads or centrifuge safety cups are used, and if these rotors or safety cups are opened only in a biological safety cabinet.
2. Face protection (goggles, mask, face shield, or other splatter guards) is used for anticipated splashes or sprays of infectious or other hazardous materials to the face, when the microorganisms must be manipulated outside the biological safety cabinet.
3. Protective laboratory coats, gowns, smocks, or uniforms designated for lab use are worn while in the laboratory. This protective clothing is removed and left in the laboratory before leaving for non-laboratory areas (e.g., cafeteria, library, administrative offices). All protective clothing is either disposed of in the laboratory or laundered by the institution; it should never be taken home by personnel.
4. Gloves are worn when handling infected animals and when hands may contact infectious materials, contaminated surfaces, or equipment. Wearing two pairs of gloves may be appropriate; if a spill or splatter occurs, the hand will be protected after the contaminated glove is removed. Gloves are disposed of when contaminated, removed when work with infectious materials is completed, and are not worn outside the laboratory. Disposable gloves are not washed or reused.

Laboratory Facilities (Secondary Barriers) (BSL2):

1. A method for decontamination of infectious or regulated laboratory wastes is available (e.g., autoclave, chemical disinfection, incinerator, or other approved decontamination system).
2. An eyewash facility is readily available.

Biosafety Level 3

BSL3 is applicable to clinical, diagnostic, teaching, research, or production facilities in which work is done with indigenous or exotic agents that may cause serious or potentially lethal disease as a result of exposure by the inhalation route.

Examples of BSL3 Agents:

Mycobacterium tuberculosis *Vesicular Stomatitis Virus* *Yellow Fever Virus*

Francisella tularensis - during manipulations of cultures and for experimental animal studies

Coxiella burnetii - for activities involving inoculation, incubation, and harvesting of embryonated eggs or cell cultures, necropsy of infected animals, and manipulation of infected tissues

Laboratory personnel have specific training in handling pathogenic and potentially lethal agents and are supervised by scientists who are experienced in working with these agents.

All procedures involving the manipulation of infectious materials are conducted within biological safety cabinets or other physical containment devices or by personnel wearing appropriate personal protective clothing and equipment. The laboratory has special engineering and design features.

It is recognized that many existing facilities may not have all the facility safeguards recommended for BSL3 (e.g., access zone, sealed penetrations, and directional airflow). In these circumstances, acceptable safety may be achieved for routine or repetitive operations (e.g., diagnostic procedures involving the propagation of an agent for identification, typing, and susceptibility testing) in BSL2 facilities. However, the recommended Standard Microbiological Practices, Special Practices, and Safety Equipment for BSL3 must be rigorously followed. The decision to implement this modification of BSL3 recommendations should be made only by the laboratory director. *In addition to all the requirements for BSL2*, work at BSL3 requires:

Special Practices (BSL3):

1. Laboratory doors are kept closed when experiments are in progress.
2. The laboratory director controls access to the laboratory and restricts access to persons whose presence is required for program or support purposes.
3. The laboratory director is responsible for ensuring that before working with organisms at BSL3, all personnel demonstrate proficiency in standard microbiological practices and techniques, and in the practices and operations specific to the laboratory facility. This might include prior experience in handling human pathogens or cell cultures, or a specific training program provided by the laboratory director or other scientist proficient in safe microbiological practices and techniques.
4. All manipulations involving infectious materials are conducted in biological safety cabinets or other physical containment devices within the containment module. No work in open vessels is conducted on the open bench.
5. All potentially contaminated waste materials (e.g., gloves and lab coats) from laboratories or animal rooms are decontaminated before disposal or reuse.
6. Spills of infectious materials are decontaminated, contained, and cleaned by appropriate professional staff, or others properly trained and equipped to work with concentrated infectious material.

Safety Equipment (Primary Barriers) (BSL3):

1. Properly maintained biological safety cabinets are used (Class II or III) for all manipulation of infectious materials.
2. Outside of a biological safety cabinet, appropriate combinations of personal

- protective equipment are used (e.g., special protective clothing, masks, gloves, face protection, or respirators), in combination with physical containment devices (e.g., centrifuge safety cups, sealed centrifuge rotors, or containment caging for animals).
3. This equipment must be used for manipulations of cultures and of those clinical or environmental materials that may be a source of infectious aerosols; the aerosol challenge of experimental animals; harvesting of tissues or fluids from infected animals and embryonated eggs; and necropsy of infected animals.
 4. Respiratory protection is worn when aerosols cannot be safely contained (i.e., outside of a biological safety cabinet), and in rooms containing infected animals.
 5. Protective laboratory clothing such as solid-front or wrap-around gowns, scrub suits, or coveralls must be worn inside the laboratory only. Reusable laboratory clothing is to be decontaminated before being laundered.

Laboratory Facilities (Secondary Barriers) (BSL3):

1. The laboratory is separated from areas that are open to unrestricted traffic flow within the building. Passage through two sets of self-closing doors is the basic requirement for entry into the laboratory from access corridors or other contiguous areas. A clothes change room (shower optional) may be included in the passageway.
2. Each laboratory contains a sink for handwashing. The sink is foot, elbow, or automatically operated and is located near the laboratory exit door.
3. The interior surfaces of walls, floors, and ceilings are water-resistant so that they can be easily cleaned. Penetrations in these surfaces are sealed or capable of being sealed to facilitate decontamination.
4. Windows in the laboratory are closed and sealed.
5. A method for decontaminating all laboratory wastes is available, preferably within the laboratory (i.e., autoclave, chemical disinfection, incineration, or other approved decontamination method).
6. A ducted exhaust air ventilation system is provided. This system creates directional airflow that draws air from "clean" areas into the laboratory toward "contaminated" areas. The exhaust air is not recirculated to any other area of the building, and is discharged to the outside with filtration and other treatment optional. The outside exhaust must be dispersed away from occupied areas and air intakes. Laboratory personnel must verify that the direction of airflow (into the laboratory) is proper.
7. The High Efficiency Particulate Air (HEPA) filtered exhaust air from Class II or Class III biological safety cabinets is discharged directly to the outside or through the building exhaust system. If the HEPA filtered exhaust air from Class II or Class III biological safety cabinets is to be discharged to the outside through the building exhaust air system, it is connected to this system in a manner (e.g., thimble unit connection) that avoids any interference with the air balance of the cabinets or building exhaust system. Exhaust air from Class II biological safety cabinets may be recirculated within the laboratory if the cabinet is tested and certified according to the guidelines included on National Sanitation Foundation Standard 49.
8. Continuous flow centrifuges or other equipment that may produce aerosols are contained in devices that exhaust air through HEPA filters before discharge into the laboratory.
9. Vacuum lines are protected with liquid disinfectant traps and HEPA filters, or their equivalent, which are routinely maintained and replaced as needed.

III. Laboratory Equipment

Biological Safety Cabinet (BSC)

A biological safety cabinet is used as a primary barrier against exposure to infectious biological agents. A BSC has High Efficiency Particulate Air (HEPA) filters. The airflow in a BSC is laminar, i.e. the air moves with uniform velocity in one direction along parallel flow lines. A BSC must be used in conjunction with safe laboratory techniques, because potentially dangerous aerosols can still escape.

Depending on the design, a BSC may be vented to the outside or the air may be exhausted into the room. BSCs are not chemical fume hoods. A percentage of the air is recirculated in most types of BSCs. Therefore, the levels of explosive, flammable, or toxic materials will be concentrated within the cabinet. HEPA filters only trap particulates, allowing any contaminant in non-particulate form to pass through the filter.

Classes of BSCs:

Class I

In Class I BSCs, the exhaust air is HEPA filtered so the user and the environment are protected, but the product inside the cabinet is not. With a Class I cabinet, the user's hands and arms while inside the cabinet are exposed to the infectious materials. The Class I BSC is designed for general microbiological research with low to moderate risk agents, and is useful for containment of mixers, blenders, and other equipment.

Class II

There are different types of Class II BSCs, but they all offer HEPA filtered supply and exhaust air. This type of cabinet will protect the user, environment, and the product and is suitable for work assigned to Biosafety Levels 1, 2, or 3. Class II cabinets are the class most commonly used.

Class III

These cabinets are often referred to as Gloveboxes. The Class III cabinet is gas-tight and under negative pressure. All work in the cabinet is performed through rubber gloves attached to entry portals. The Class III cabinet offers the highest level of protection from infectious aerosols. Class III cabinets are most suitable for work with agents that require BSL3 or BSL4 containment.

Proper Use of BSC:

1. Before and after use, wipe the surface of the BSC with a suitable disinfectant, e.g., 70% alcohol or a 1:10 bleach solution.
2. Place everything you will need inside the cabinet before beginning work, including a waste container. You should not have to penetrate the air barrier of the cabinet once work has begun.

3. Do not place anything on the air intake grilles as this will block the air supply.
4. A sign can be posted on the door of the room stating that the cabinet is in use.
5. You should prevent unnecessary opening and closing of doors as this will disrupt the airflow of the cabinet.
6. Always wear a lab coat while using the cabinet and conduct your work at least four inches inside the cabinet.
7. Place burners to the rear of the cabinet to reduce air turbulence.
8. Place a disinfectant-soaked towel on the work surface to contain any splatters or small spills that might occur.
9. Do not work in the BSC while the ultraviolet light is on. Ultraviolet light can quickly injure the eye.
10. When finished with your work procedure, cover the waste container and decontaminate the surfaces of any equipment that is not enclosed.
11. Operate the cabinet for five minutes before and after performing any work in it in order to purge airborne contaminants.
12. Remove the equipment from the cabinet and decontaminate the work surface.
13. Thoroughly wash your hands and arms.

Certification of BSCs:

A BSC must be certified annually and after it has been newly installed, moved, or had a filter replaced. There are several companies in the area which provide this service.

Clean Bench

Clean benches (a.k.a. laminar flow hoods) are not considered laboratory safety equipment. However, they deserve mention because they may be confused with BSCs. A clean bench is designed to protect the product from contamination, but it does not protect the user. The direction of airflow in a clean bench is toward the user.

Pipetting Device

In the distant past, some lab personnel were taught to mouth pipette. This practice has been known to result in many laboratory acquired infections. With the availability of mechanical pipetting devices, mouth pipetting is strictly prohibited. Mouth pipetting should never be used, even for innocuous materials, because you may at some time mistakenly mouth pipette something that is hazardous. To minimize aerosol production, a pipette should be drained with the tip against the inner wall of the receiving vessel. **Never forcibly expel any hazardous material from a pipette.**

Centrifuges, Sonicators, Homogenizers, and Blenders

All of these instruments can create aerosols and this must be considered with each use. The necessary precautions taken will depend upon what is being used in these instruments. If hazardous materials such as carcinogens, highly toxic, or infectious agents will be placed in any of these instruments, then precautions must be taken to prevent an exposure of lab personnel to aerosols or liquids.

Centrifuge:

Centrifuges that have sealed buckets, safety trunnion cups, or sealed heads are effective at preventing the escape of aerosols and liquids. The potential for exposing people to a hazardous material used in a centrifuge is great if the centrifuge tube breaks without the use of the safety features mentioned above.

Routinely inspect your centrifuge to ensure leakage is not occurring. An indicator such as fluorescein can be used to detect leaks. The fluorescein can be added to water and then centrifuged as you would other materials. An ultraviolet light can then be used to detect the fluorescein's presence on work surfaces, floors, and walls.

Sonicators, Homogenizers, and Blenders:

Depending on the nature of the material being used in these instruments and also in centrifuges, it may be necessary for them to be used or opened only in a biological safety cabinet. When working with infectious agents, blenders should have leak proof bearings and a tight-fitting, gasketed lid. Inspect the lid and gaskets routinely to ensure they are in good condition. Household blenders do not prevent the spread of aerosols. Also, hearing protection may be required while using a sonicator.

IV. Personal Protective Equipment

The type of personal protective equipment required in microbiological labs will depend upon the assigned Biosafety Level for that lab (see Section 2 of this chapter regarding Biosafety Levels). The protective clothing suitable for a typical undergraduate microbiology lab is a lab coat, to prevent street clothes from getting soiled, and latex or vinyl gloves. Long hair must be restrained if Bunsen burners are in use.

For a typical graduate level teaching or research microbiology lab (which are often a BSL2), lab coats or similar protective clothing should be worn while in the lab, and gloves must be worn while handling any infectious materials. Additionally, if the work involves human blood, a face shield, safety glasses or goggles, and a mask may be required if there is a potential for splash.

A research lab that is assigned a Biosafety Level 3 has additional requirements for personal protective equipment: laboratory clothing that protects street clothing must be worn, e.g., a solid-front or wrap-around gown. Typical lab coats which button down the front are not acceptable because they do not provide full protection. Gloves must be worn in the lab, and respirators worn in rooms containing infected animals.

Whenever personal protective equipment becomes contaminated, it must be removed and replaced. Leave protective clothing in the lab and do not wear it to other non-lab areas. Disposable gloves are meant to be used only once and should then be discarded. In between glove changes, thoroughly wash your hands and arms.

V. Waste Disposal

There are many types of waste generated in a microbiological lab and all need to be handled, treated, stored, and disposed of properly. Waste is either rendered un-infectious utilizing an

in-house autoclave or disposed of offsite utilizing an offsite contractor. Disposal through an offsite contractor is facilitated through the EHSRM by calling the hazardous waste line or by emailing waste@utrgv.edu.

VI. Bloodborne Pathogens

Bloodborne pathogens is specifically addressed in the UTRGV Bloodborne Pathogens Control Plan.

In December 1991, the Federal Government published the final rule governing occupational exposure to bloodborne pathogens which became effective March 6, 1992. The objective of this standard is to provide guidelines to eliminate or minimize employee exposure to human bloodborne pathogens. A human bloodborne pathogen is a pathogenic microorganism present in human blood that can cause disease in humans. The standard includes the Centers for Disease Control and Prevention (CDC) guidelines referred to as Universal Precautions.

If during the course of work a potential exists for coming in contact with human blood or other potentially infectious materials, you must receive training on bloodborne pathogens. Contact EHSRM (Edinburg Campus: 956-665-3690 or Brownsville Campus: 956-882-5929) for information.

Universal Precautions:

The CDC Universal Precautions are used as an approach to infection control. The concept behind Universal Precautions is to treat all human blood and certain human body fluids as if known to be infected with HIV, Hepatitis B, and other bloodborne pathogens.

The Universal Precautions are summarized below and should be practiced whenever coming in contact with human blood:

1. Use appropriate barrier precautions to prevent skin and mucus membrane exposure when contact with blood is anticipated. Always wear gloves. Wear masks and protective eyewear or face shields to prevent exposure to the eyes, mouth, and nose during procedures that are likely to result in droplets of blood. Wear gowns or aprons during procedures that are likely to result in splashes of blood. Remove all protective clothing before leaving the laboratory.
2. Wash hands and other skin surfaces immediately if contaminated with blood and after the removal of gloves.
3. Limit the use of needles to where there is no alternative and take precautions to prevent injuries by needles and other sharps. To prevent needle-stick injuries, needles should not be recapped, bent, removed from the syringe, or otherwise manipulated by hand. Place needles and other sharps into puncture-resistant containers.
4. Keep all specimens of blood in well-constructed containers with a secure lid to prevent leakage during transport.
5. Use biological safety cabinets whenever procedures that have a high potential for generating droplets are conducted.
6. Never mouth pipette.
7. Decontaminate laboratory work surfaces after a spill of blood and when work activities are completed.

Working with Animals:

Some animals can also carry pathogens that can be transmitted to humans through contact with their body fluids, similar to human bloodborne pathogens. This contact can occur through biting, spitting, or contamination of broken skin or mucus membranes with bodily secretions from the animal. An example of a disease transmitted this way is the B-virus infection. B-virus is a naturally occurring alpha-herpes virus infecting macaques. Human infection has been documented in 25 instances, 16 of those resulting in death.

When working with animals such as macaques that are capable of transmitting disease to humans, take necessary precautions to protect yourself. Wear gloves, masks, and laboratory coats whenever entering an area where these animals are housed.

VII. Emergency Procedures

Refer to the Emergency Procedures Section of this manual, Chapter C, for important information on emergency procedures. In this section, some specific instructions will be given for the clean-up of a biological spill.

Some biological materials when spilled or released can lead to significant infection exposures to personnel. This is particularly hazardous when the agent spilled or released is classified as a BSL2 agent or higher. The following emergency procedures that must be followed are determined by the Biosafety Level of the agent involved.

Spills or Releases Involving BSL1 Agents:

1. Wear a lab coat and disposable gloves.
2. Soak a paper towel(s) in an appropriate disinfectant such as a fresh 1:10 bleach solution and place over the spill area.
3. Place the paper towels and gloves into a biohazard bag for disposal by EHSRM or autoclave the materials.

Spills or Releases Involving BSL2 Agents:

1. If an accident occurs that may generate aerosols or droplets of an infectious agent, leave the area, close the door, and decontaminate clothing and shower. Allow at least 30 minutes for the droplets to settle and for the aerosol concentration to decrease.
2. Wear appropriate personal protective clothing such as gloves, lab coat, and approved respiratory equipment, if needed.
3. Cover the spill area with paper towels, pour a 1:10 bleach solution around the edges of the spill and then into the spill. Allow 10 minutes contact time.
4. Working from the outer edges into the center, use paper towels to clean the area. Clean the spill area with fresh towels soaked in a disinfectant. Be sure to decontaminate any areas or surfaces that you suspect may have been effected by the spill. Place all clean up materials and gloves into a bag for decontamination, preferably by autoclaving. Wash thoroughly.
5. A small spill of material that did not result in a significant generation of aerosols, or contamination of a person, can be cleaned up following steps two through four above.

Spills or Releases Involving BSL3 Agents:

1. If the spill occurs in a biological safety cabinet, keep the cabinet running, and clean the spill following steps two through four from *Spills or Releases Involving BSL2 Agents*, except that personal protective clothing appropriate for a BSL3 lab should be worn. If the spill in the cabinet is quite substantial, it may be necessary to decontaminate the cabinet's fan, filters, and airflow plenums. This should be done by a qualified outside company. Call EHSRM for assistance.
2. If a minor spill occurs outside of a biological safety cabinet, follow steps two through four from *Spills or Releases Involving BSL2 Agents*, except that personal protective clothing appropriate for a BSL3 lab should be worn.
3. If anything other than a minor spill occurs outside of a biological safety cabinet, leave the area immediately and notify appropriate personnel including EHSRM. A specially designed decontamination procedure may be necessary.

Note: Whenever bleach is used to clean up spills of an infectious agent, a fresh solution should be prepared. **After about one week, a bleach and water solution will lose its effectiveness for decontamination.**

References:

Biosafety in the Laboratory, Prudent Practices for the Handling and Disposal of Infectious Materials. National Research Council. 1989.

Biosafety in Microbiological and Biomedical Laboratories, Third Edition. Centers for Disease Control and National Institutes of Health. 1993.