HEALTH, SAFETY AND SECURITY PLAN

Winthrop University
Department of Biology

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Department of Biology Biosafety Committee

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Introduction

The Williams-Steiger Occupational Safety and Health Act of 1970 (OSHA) became effective on April 28, 1972. The purpose of this act is "to assure, so far as possible, every working man and woman in the nation safe and healthful working conditions and to preserve our human resources." This Health, Safety and Security Plan has been developed by the Department of Biology Biosafety Committee to serve as the guidance document for the implementation of an effective health and safety program which includes employee and student training, accident prevention, emergency guidelines, and incorporates a chemical hygiene plan and biosafety plan.

The success of this plan depends upon the commitment and active involvement of all Department of Biology employees and students. The Department Chair has assigned a Safety/Chemical Hygiene Officer (SCHO) and created a Biosafety Committee (BSC) not only to prepare this plan, but also to oversee its implementation, monitor the effectiveness of the training programs and to provide information to faculty, staff and students as they apply this plan to various teaching and research situations. The Biology Department Laboratory Manager is assigned as the Safety/Chemical Hygiene Officer (SCHO)

This plan reflects the particular character and operations of Biology teaching and research laboratories and may be subject to revision as required by changing conditions or circumstances. The plan will be reviewed periodically by the Biosafety Committee and updated as necessary. This plan is available for review by all employees and students who utilize teaching and research laboratories in the Department of Biology. Copies will be available to all employees of the department, in the following locations:

Department of Biology Office	202 Dalton Hall
Department Chair's Office	204 Dalton Hall
Biology Supply Room	131 Dalton Hall
SCHO/Laboratory Manager's Office	132 Dalton Hall
Environmental Health and Safety	349 Columbia Avenue

A copy will also be available online. This plan is considered comprehensive for the current activities in the department; however, additional regulatory information and associated guidelines are available from the following sources:

- **CDC/NIH:** *Biosafety in Microbiological and Biomedical Laboratories***,** 6th ed. (Revised 2020) https://www.cdc.gov/labs/BMBL.html
- **NIH:** *Guidelines for Research Involving Recombinant DNA Molecules*, (2019) https://osp.od.nih.gov/wp-content/uploads/NIH_Guidelines.pdf

- **World Health Organization:** *Laboratory Biosafety Manual,* 4th ed. (2020) https://www.who.int/publications/i/item/9789240011311
- **American Biological Safety Association (ABSA):** *Risk Group (RG) Database.* https://my.absa.org/Riskgroups
- **OSHA:** *Occupational Exposure to Hazardous Chemicals in Laboratories (The Lab Standard)*. Code of Federal Regulations, 29 CFR 1910.1450. (2009) https://www.osha.gov/pls/oshaweb/owadisp.show_documentlay_standard_group?p_toc_level=1&p_part_number=1910

Section A: Responsibility

It is the responsibility of Winthrop University administration to provide, as far as reasonably practical, safe working conditions for its employees and students. At the department level, the Chair is ultimately responsible for ensuring the health, safety and security of the faculty, staff and students in the department. The Department Chair should ensure that an effective safety plan is in place and is supported by all department personnel.

The Safety/Chemical Hygiene Officer (SCHO) and department Biosafety Committee (BSC) assist and support the Department Chair in the administration and implementation of the Health, Safety and Security Plan.

It is the responsibility of faculty, principal investigators, laboratory supervisors, and staff to ensure safe working conditions in classrooms and laboratories under their control. Students are required to follow any and all health, safety and security policies established by this plan. Detailed responsibilities are outlined below:

Department Chair

Ensures the development and implementation of a departmental health, safety and security plan and ensures compliance with the plan.

1. Serves as the department authority regarding chemical hygiene and biosafety.
2. Appoints a department Safety/Chemical Hygiene Officer (SCHO).
3. Establishes a department Biosafety Committee (BSC) and appoints faculty and/or staff to serve on the committee.
4. Ensures that faculty, staff and students are aware of their individual responsibilities regarding health, safety and security.
5. Mandates laboratory safety training and oversees the development and periodic review of laboratory safety training courses.
6. Ensures that Accident Reports (Appendix A) are submitted to Environmental Health & Safety (EHS), 349 Columbia Ave.

Safety/Chemical Hygiene Officer (SCHO)

The Biology Department SCHO is the Biology Department Laboratory Manager. This is currently Stefan Wunderlich.

1. Monitors the procurement, use and disposal of all hazardous chemical and biohazardous materials used in department laboratories.
2. Implements and monitors compliance with the department Chemical Hygiene Plan.

3. Maintains a current department chemical inventory through Winthrop University's Chemical Inventory System maintained by Environmental Health & Safety (EHS) and overseen by Winthrop University's OSHA Compliance Officer. This is regularly updated as new materials are added or storage quantities change.
4. Oversees an annual physical inventory of all chemicals in storage, forwarding updated annual inventory to the OSHA Compliance Officer.
5. Ensures receipt of a Safety Data Sheet (SDS) for each hazardous chemical procured by the department and maintains a hardcopy file in accordance with the University Hazard Communication Plan in addition to electronic copies in the Chemical Inventory System.
6. Ensures that personal protective equipment (PPE), appropriate for department activities, is available and in working order.
7. Maintains the file of Accident Reports.
8. Serves as a continuing member of the BSC, as the Building Coordinator, and department liaison to EHS.
9. Participates in opportunities afforded by the department for professional development in areas related to laboratory safety, chemical hygiene and biosafety.

Biosafety Committee (BSC)

1. Working in cooperation with the SCHO, assists the Department Chair with implementation of the Health, Safety and Security Plan and conducts a periodic review of the plan, making revisions as necessary.
2. Forwards plan revisions and updates to EHS, 349 Columbia Avenue.
3. Oversees laboratory safety training of faculty, staff and students, reviewing and updating training courses as necessary.
4. Assists faculty, staff and students with the implementation of the training.
5. Maintains contact with the Institutional Biosafety Committee (IBC) and EHS.

Laboratory/Classroom Supervisors and Principal Investigators (Faculty and Staff)

1. Ensures that all students working under their supervision in laboratories/classrooms have been trained in laboratory safety and understand their responsibilities.
2. Instructs every student working under their supervision in the safe use and storage of chemicals, biohazards and other potentially hazardous materials in their laboratories.
3. Ensures that appropriate PPE (goggles, gloves, face shields, lab aprons, lab coats, etc.) is worn by any student operating equipment or working with chemicals, biohazards or other potentially hazardous materials in their laboratory.
4. Ensures that all equipment in the laboratory is in proper working condition and all students are provided adequate training before using this equipment.
5. Informs the Department Chair and SCHO of hazardous conditions or situations.

6. Informs SCHO of needed repairs to equipment or facilities. The SCHO will refer the request to Facilities Management, or in the case of specialized equipment, to the appropriate repair service.
7. Reports all accidents to the Department Chair and SCHO, and completes an Accident Report (Appendix A).
8. Plans and conducts each laboratory operation, whether teaching or research, in accordance with this Health, Safety and Security Plan.
9. Develops and maintains good personal chemical hygiene habits.
10. Completes a laboratory inspection of teaching laboratories annually and submits the Laboratory Inspection Form (Appendix B) to the SCBO.
11. Follows the *University Emergency Guidelines* (Appendix C) in the event of such emergency situations as fire or hazardous weather and takes responsibility for the safety of students in emergency situations.

Students

1. Must adhere to all health, safety and security policies outlined in this plan and follow the instructions of faculty or staff regarding proper classroom and laboratory procedures.
2. Should not use any laboratory equipment or work with hazardous chemicals, biohazards or other potentially hazardous materials until adequately trained and authorized to do so by faculty or staff.
3. Should use appropriate personal protective equipment (goggles, face shields, gloves, lab coats, etc.) when working in the laboratory.
4. Should know the exact location of eyewash stations, safety showers, fire extinguishers, spill kits and exits before beginning work in any laboratory.
5. Must follow the *University Emergency Guidelines* (Appendix C) in the event of such emergency situations as fire or hazardous weather.
6. All students (biology and non-biology majors) enrolled in BIOL 222 shall successfully complete basic Laboratory Safety Training.
7. Must plan and conduct each laboratory operation in accordance with this Health, Safety and Security Plan.
8. Must develop and use good personal chemical hygiene habits.
9. Must not bring their children into laboratories while working. Children must remain in the graduate office or lobbies under the care of an adult while students work in laboratories.
10. Must wear the appropriate apparel in laboratories when working with hazardous chemicals or biohazardous agents.

Record Keeping

The following reports and records provide supporting documentation of an effective health, safety and security plan. The Department Chair shall ensure the reports and records detailed below are

completed in a timely manner, submitted to the appropriate individual, and filed in the specified location.

1. Accident Reports (Appendix A) are to be completed by the laboratory supervisor or SCHO with assistance from other employees or students, as needed. Reports are to be submitted to EHS, 349 Columbia Avenue with a copy retained by the SCHO/Laboratory Manager, 132 Dalton Hall.
2. Documentation related to hazardous material exposures and any associated medical records will be retained in the office of the University OSHA Compliance Officer, EHS, 349 Columbia Avenue.
3. Student safety training records will also be maintained electronically through the completion of BIOL 222 and BIOL 300.

Section B: Laboratory Facilities and Access

The Department of Biology has developed specific procedures to ensure the safety of faculty, staff and students including but not limited to: emergency contact information posted at doors to each academic and research laboratory; hazard warning signs posted on the doors of research and academic labs indicating the presence of potentially harmful chemicals or biological materials; and routine inspection of safety equipment. Hazardous chemical Right-to-Know information is also available in storage areas and laboratories that house hazardous chemicals. Lastly, *University Emergency Guidelines* are available in each academic and research lab.

Housekeeping

Each employee and student is individually responsible for the cleanliness of their workspace and jointly responsible for the cleanliness of shared or common areas of the laboratory. The Department Chair, SCHO and BSC will monitor and enforce the housekeeping standards outlined below:

1. All spills on lab benches or floors shall be immediately cleaned up under supervision of the laboratory supervisor. Spilled material, contaminated debris and clean-up supplies must be properly contained and disposed of according to the procedures outlined in the Chemical Hygiene Plan (Section G) and the Biosafety Plan (Section H) of this document.
2. Laboratory benches shall be kept clear of equipment and chemicals except those necessary for the work being performed.
3. The laboratory work area shall be cleaned at the end of each operation, class activity and/or workday.
4. All apparatus shall be thoroughly cleaned and returned to the correct storage area upon completion of use.
5. All floors, aisles, exits, fire extinguishers, eyewash stations, safety showers, electrical panels/disconnects and other emergency equipment shall remain unobstructed.
6. All labels shall face front.
7. Chemical containers shall be clean, properly labeled and returned to storage upon completion of use.

Signs and Labels

Two types of chemical safety signs and labels shall be used in the department to provide hazard information. Globally Harmonized System (GHS) or National Fire Protection Agency (NFPA) signs and labels are used at storage areas and on chemical containers to inform users of the health, fire, instability and incompatibility hazards of materials in storage or use. GHS signs and labels are available upon request from the Laboratory Manager.

In addition, Biohazard signs are posted on the doors of teaching and research laboratories in which activities involve recombinant DNA or the use of biohazardous microorganisms. Biohazard signs should include the name of the Laboratory Manager and Laboratory Supervisor/Principal Investigator along with appropriate emergency contact information. In addition, biohazard signs should identify the specific biohazard and the required containment level for that organism. Biohazard signs are also available upon request from the Laboratory Manager.

Appendix D contains examples of pictograms that can be used to communicate specific hazards as well as an explanation of the NFPA labeling system.

Safety Training

All faculty, staff, and students will have access to this Health, Safety and Security Plan. A copy of this Health, Safety and Security Plan will be placed in designated areas (see page 4), and available online via a QR code in every research and teaching laboratory.

All students in biology shall receive safety training through BIOL 220 and BIOL 300 courses. In BIOL 220, students shall be trained in general safety, chemical safety, PPE and other protection, and basic laboratory equipment safe practices. In BIOL 300, students will be trained in biosafety and ethical standards of research as well as review basic laboratory safety. All students will be required to pass a safety assessment to continue and complete the course. Records of safety training will be kept through satisfactory completion of these courses.

In addition, laboratory supervisors and principal investigators are responsible for providing training for any student working in their laboratory with information regarding specific chemical hazards, biohazards, potential exposures and equipment hazards, present in those laboratories. Student training regarding laboratory chemical use shall include physical and health hazards of chemicals in the lab and measures that can be used to protect themselves from these hazards.

After Hours Access to Dalton Hall

After hours and weekend access to Dalton Hall is a privilege given to students who have successfully completed appropriate laboratory safety training. The principal investigator or course instructor will forward names of students and rooms that they should be granted access to the Laboratory Manager or Departmental Administrative Assistant using the currents “After Hours Form”. Students who have earned after hours privileges should not perform hazardous work alone in the building. All hazardous operations shall be performed during a time when at least two people are present in the building.

Unattended Operations

When laboratory operations are performed which will be unattended by employees or students (continuous operations, overnight reactions, etc.), the following procedures shall be followed to ensure safe completion of the operation:

1. The laboratory supervisor shall review work procedures prior to beginning the unattended operation.
2. Appropriate labels shall be used to indicate equipment use and the responsible person.
3. The overhead lights in the laboratory shall be left on.
4. The responsible person(s) will return to the laboratory at the conclusion of the operation to dismantle the apparatus and clean up the area.

Section C: Accident Prevention

Accidents are defined as incidents that have caused or might reasonably be expected to have caused injury to persons and/or damage to property. It is the policy of the Department of Biology to actively prevent accidents whenever possible. To this end, faculty, staff and students are required to report any potentially hazardous or dangerous situation to the Laboratory Manager (SCHO) in the Biology Supply Room, 131 Dalton Hall or by phone or email. Reports should be submitted immediately when the hazard becomes apparent.

Faculty and staff should provide supplemental location and/or activity-specific safety training for students working in their laboratory or classroom. Faculty and staff must ensure the general cleanliness and orderliness of the working environment, use of personal protective equipment (PPE) when necessary, and the proper maintenance of equipment. Additionally, faculty and staff must ensure that only authorized or adequately trained individuals are allowed to use or operate equipment or work with chemicals, biohazards, or other potentially hazardous materials. To prevent accidents the following general safety guidelines should be followed:

1. Individuals should follow the safety guidelines at all times.
2. Individuals should never work alone in a potentially hazardous area.
3. Individuals should familiarize themselves with the location of exits and emergency equipment.
4. Eye protection should be worn in the laboratory when there is a splash risk. Contact lenses are strongly discouraged.
5. Individuals should never eat, drink, use tobacco products, or apply cosmetics in the laboratory.
6. Individuals should always wear appropriate shoes in the laboratory. Sandals and bare feet are not permitted.
7. Individuals must wash hands thoroughly with soap and water whenever leaving the laboratory.
8. Individuals must handle all chemicals in accordance with the Chemical Hygiene Plan (Section G) and Data Safety Sheets.
9. Individuals handling biohazards shall follow the procedures outlined in the Biosafety Plan (Section H) and according to an approved Biosafety Protocol.
10. All chemical or biohazard spills must be immediately and properly cleaned up according to the procedures outlined in the Chemical Hygiene Plan (Section G) or Biosafety Plan (Section H).
11. Laboratory operations, which have the potential to create a fire or explosion, require special procedures and safety equipment. Such operations must have the prior approval of the Environmental Health & Safety Manager.
12. Individuals must get help or use a cart when lifting or moving heavy objects. The Laboratory Manager should be notified prior to moving large, bulky, or heavy equipment, and will make the decision as to the most appropriate mode of lifting and moving.
13. Individuals must use a well-placed, sturdy stool or ladder for climbing.
14. All exits and aisles should be kept clear and unobstructed.

Section D: Reporting Accidents and Injuries

Should an accident occur, apart from immediate attention to injured persons or crime scenes, attention should be directed at securing the area, altering or removing any hazard or equipment involved in the accident, and cleaning up the site. An Accident Report (see Appendix A) must be filed with the Department Chair, SCHO, and EHS as soon as is reasonably possible under the circumstances. Accident Report forms are available in the Biology Office, 202 Dalton Hall, and the Biology Supply Room, 131 Dalton Hall.

Workers' Compensation

A Workers ' Compensation Program administered by the State Accident Fund (SAF) covers all employees of the university, including student employees, while on duty. Compendium Services, Inc. provides medical case management through a contractual agreement with SAF. Compendium Services file workers' compensation claims electronically in response to the submission of a First Report of Injury. Questions regarding this program or the reporting procedures should be directed to the Environmental Health & Safety Manager at 803-323-2328.

Reporting Minor Injuries and Work-Related Illnesses

1. Any employee who sustain even a minor injury while on duty or develops a work-related illness must immediately report the injury or illness to their supervisor.
2. When medical treatment is needed, the supervisor should contact Compendium Services at 877-709-2667 to file a First Report of Injury and receive authorization for treatment. All non-emergency medical treatment must be pre-approved by Compendium Services and is provided by: Concentra Urgent Care, 1393 Celanese Road, Rock Hill, SC 29732, 803-329-3103
3. The supervisor must also report the injury or illness to the Worker's Compensation Administrator at 803-323-2392.

Employees should report all injuries and work-related illnesses to their supervisor no matter how minor they may seem to be at the time. An employee who fails to report an injury or work-related illness may risk being denied benefits by SAF should medical treatment be needed at a later time.

Medical Emergencies

1. In the event of a serious or life-threatening injury that requires immediate or emergency medical treatment, call Campus Police at 803-323-3333 or 9-911 for an emergency operator. Compendium Services and the Worker's Compensation Administrator should be contacted as soon as the situation allows. See "Workplace Injuries" at our website Workplace Injuries

2. When an injured employee is transported to an emergency treatment facility for care, the supervisor or a designated university representative should accompany them to the facility and remain until the employee has been admitted or released.
3. The supervisor or designated representative should notify the treating facility that workers' compensation may be filed for the injured employee and provide the following insurance information, if needed:

For case management and treatment authorization: **Compendium Services, Inc.**
877-709-2667

For billing and payment information: **State Accident Fund**
P.O. Box 102100
Columbia, SC 29221-5000
800-521-6576

For verification of employment and insurance coverage:

Winthrop University	**OR**	**Winthrop University**
Environmental Health & Safety		**Human Resources**
803-323-2392		**803-323-2273**

Medical Consultations and Examinations

An opportunity to receive medical attention is available to all employees who work with hazardous chemicals in the laboratory, under the following circumstances: (a) whenever an employee develops signs or symptoms associated with exposure to a hazardous chemical which the employee may have been used in the laboratory, or (b) whenever an event takes place in the laboratory, such as a spill, leak, explosion or other occurrence, resulting in the likelihood of a hazardous exposure. These medical consultations and examinations must be scheduled by Environmental Health and Safety (323-2392) and will be provided without cost to the employee, without loss of pay, at a reasonable time and place, and shall be administered by or under the direct supervision of a licensed physician.

Section E: Emergency Guidelines

All employees and students are expected to familiarize themselves with the *University Emergency Guidelines* (Appendix C) as published on the university Critical Incident Management webpage: (**https://www.winthrop.edu/uploadedFiles/emergency/EmergencyGuidelines.pdf**), and available via a QR code in each classroom and laboratory.

Section F: Safety Inspections

The SCHO/Laboratory Manager with support from the BSC will conduct routine lab inspections of academic and research labs to ensure compliance with this plan and applicable regulations. Faculty and/or laboratory supervisors are required to complete a lab inspection of their lab using the Laboratory Inspection Checklist (Appendix B).

Environmental Health & Safety (EHS) conducts a fire safety inspection of Dalton Hall checking fire extinguishers, emergency equipment, and exit pathways. EHS inspects and tests eyewash stations and safety showers annually, as well as conducting periodic inspections of classrooms, laboratories, lab equipment and chemical fume hoods to confirm that the department is following the specifics of this plan. EHS also conducts hazardous waste management inspections of each satellite accumulation area to confirm compliance with federal and state hazardous waste management regulations. Identified facility deficiencies will be reported to Facilities Operations and Maintenance for correction and/or repair. Deficiencies determined to be the responsibility of Biology faculty, staff or students, will be reported to the SCHO/Laboratory Manager for correction and follow-up.

Section G: Chemical Hygiene Plan

On January 31, 1990, the Occupational Safety and Health Administration (OSHA) promulgated a final rule for occupational exposure to hazardous chemicals in laboratories (29 CFR 1910.1450). Included in the standard, which became effective on May 1, 1990, is a requirement for employers covered by the standard to develop and carry out the provisions of a Chemical Hygiene Plan (CHP).

A CHP is defined as a written program which sets forth procedures, equipment, personal protective equipment and work practices that are capable of protecting employees from the health hazards presented by hazardous chemicals used in that particular workplace. Components of the CHP must include standard operating procedures for safety and health, criteria for the implementation of control measures, procedures to ensure proper operation of engineering controls, provisions for training and information dissemination, provisions for medical consultation, designation of responsible personnel and identification of particularly hazardous substances.

This section of the Health, Safety and Security Plan is the Chemical Hygiene Plan (CHP) developed for the Department of Biology, Winthrop University. Although students are generally not covered by the OSHA standard, it is the policy of this department that students will be given training commensurate with the level of hazard associated with their laboratory work. All employees and students should follow the procedures and guidelines outlined in this plan. All operations performed in the laboratory, whether teaching or research, must be planned and executed in accordance with this plan. In addition, each employee and student is expected to

develop safe personal chemical hygiene habits aimed at the reduction of chemical exposures to themselves and coworkers.

This CHP was developed to comply with paragraph (e) of the referenced OSHA standard. It will be reviewed annually, revised as necessary, and made readily available to all department employees and students. The intent of this CHP is to:

- protect employees and students from health hazards associated with the use of hazardous chemicals in teaching and research laboratories;
- to ensure that employees and students are not exposed to substances in excess of the permissible exposure limits (PELs) as defined by OSHA and codified in 29 CFR 1910.1000, Table Z-1-A; and
- to provide employees and students a plan for regulatory compliance with the OSHA Laboratory Standard as codified in 29 CFR 1910.1450 Subpart Z.

Standard operating procedures for the use of chemicals in Department of Biology laboratories are detailed in this plan. The Department Chair shall designate the department Safety/Chemical Hygiene Officer (SCHO) and ensure that this plan is implemented and the procedures followed.

Chemical Procurement

The decision to procure a chemical shall be a commitment to handle and use the chemical properly from initial receipt to ultimate disposal. Information on potential hazards (e.g., toxicity, flammability, reactivity, etc.), proper handling techniques, appropriate storage requirements and proper methods of disposal must be identified prior to purchase by review of all relevant SDS. The person initiating the request must be knowledgeable of the proper techniques for handling the material and shall be responsible for the proper disposition of the chemical within laboratories. All chemicals shall be received in a central location and personnel who receive chemical shipments shall be knowledgeable of the proper procedures for receipt. Chemical containers shall not be accepted without accompanying labels and packaging in accordance with appropriate regulations. All chemical shipments should be dated when received.

Chemical Storage

Proper storage of chemicals is essential to maintaining a safe work environment for faculty and students. The following best practices are recommended for chemical storage:

- Received chemicals shall be immediately moved to the designated storage area. Glass containers of 2 liter or greater capacity shall be placed in carrying containers or shipping containers during transportation to the storage area. Highly reactive chemicals, regardless of size, shall be similarly transported.
- Storage areas shall be well illuminated and bottles of 2 liter or greater capacity shall be stored **no more than three feet above floor level**. Chemicals shall be segregated by hazard classification and compatibility and stored in a well-identified area, with local exhaust

ventilation. Alphabetical storage of chemicals is not acceptable unless hazard classification and compatibility have also been taken into consideration.
- Acids must be stored in acid cabinets.
- Flammable chemicals must be stored in flammables cabinets.
- Acid-sensitive materials such as cyanides and sulfides shall be separated from acids or protected from contact with acids.
- Highly toxic chemicals shall be stored in unbreakable secondary containers that are appropriately labeled to indicate the hazard.
- Storage of chemicals at the lab bench or other work areas shall be limited to the amounts necessary for one teaching session or daily operation. The container size shall be the minimum convenient. Care must be taken that sensitive chemicals are not exposed to sunlight or heat.
- Laboratory fume hoods shall not be used as chemical storage areas.
- Stored chemicals shall be examined regularly by the Principal Investigator of each area and at least annually by the Laboratory Manager for replacement, deterioration and container integrity. The inspection should also determine whether any corrosion, deterioration or damage has occurred to the storage facility as a result of leaking chemicals.
- Periodic inventories of chemicals outside the storage area shall be conducted by the SCHO and unneeded items shall be properly discarded or returned to the storage area.

Chemical Handling

All Department of Biology employees and students shall develop and implement work habits consistent with this CHP in order to minimize personal, coworker, and student exposure to chemicals in the laboratory. Based on the realization that all chemicals inherently present hazards under certain conditions, exposure to all chemicals shall be minimized. The following standard procedures shall be followed for the handling and use of all chemicals:

- Skin contact with all chemicals shall be avoided. Individuals should promptly wash all areas of exposed skin if contact is made with a hazardous chemical for at least 10 minutes.
- All individuals should wash their hands prior to leaving the laboratory.
- Mouth suction for pipetting or starting a siphon is prohibited.
- Eating, drinking, smoking or application of cosmetics is prohibited in biology laboratories and chemical storage areas.
- Storage, handling and consumption of food or beverages shall not occur in areas where refrigerators, glassware or utensils are also used for laboratory operations.
- Transportation of chemicals from one laboratory to another shall be minimized. When transportation of chemicals is necessary, containers shall be moved in an unbreakable carrier or on a laboratory cart.
- Any chemical mixture shall be assumed to be as toxic as its most toxic component.
- Substances of unknown toxicity shall be assumed to be toxic.
- All chemical names and identities shall be checked and double-checked prior to use. All chemical use shall be preceded by knowledge of the chemical characteristics and potential hazards.

- All mathematical calculations used in formulating chemical solutions shall be checked by the laboratory supervisor before students are allowed to measure chemicals.
- Employees and students should be familiar with the symptoms of exposure for the chemicals with which they work and the precautions necessary to prevent exposure.
- Any and all chemical spills shall be immediately reported and cleaned up under the direction of the laboratory supervisor or SCHO.
- The intent and procedures of this CHP shall be continuously adhered to.
- In all cases of chemical exposure, neither the OSHA permissible exposure limits (PELs) or the threshold limit values (TLVs) of the American Conference of Governmental Industrial Hygienists (ACGIH) shall be exceeded.
- Engineering controls and safety equipment in biology laboratories shall be utilized. Laboratory fume hoods shall be used for all operations which have the potential to produce hazardous levels of fumes, mists, gasses or volatile solvent vapors.
- Proper disposal of laboratory waste is not only essential, it is the law. No sink or sewer disposal of chemicals is permitted.
- Specific precautions based on the toxicological characteristics of individual chemicals shall be implemented as deemed necessary by the Department Chair or SCHO.

Laboratory Equipment and Glassware

- All laboratory equipment shall be used only for its intended purpose.
- All glassware shall be handled and stored with care to minimize breakage; all broken glassware shall be immediately disposed of in the designated broken glass container.
- All evacuated glass apparatus shall be shielded to contain chemicals and glass fragments should implosion occur.
- Labels shall be attached to all chemical containers, identifying the contents and related hazards.
- Waste receptacles shall be identified as such.
- All laboratory equipment shall be inspected on a periodic basis as specified in the Health, Safety and Security Plan and replaced or repaired as necessary.
- Safety showers and eyewashes shall be inspected monthly with results documented and posted at the equipment location.

Personal Protective Equipment (PPE)

- Employees, students and visitors must wear safety glasses meeting ANSI Z87.1 standards when in all laboratories where chemical operations are in process. Contact lenses should not be worn in laboratories, unless exempted by the SCHO or laboratory supervisor.
- Chemical goggles and/or a full-face shield shall be worn during chemical transfer and handling operations as procedures dictate.
- Appropriate apparel must be worn at all times in the laboratories when working with hazardous chemicals. Bare-feet, sandals, flip-flops, perforated shoes (e.g. Crocs™ and ballet flats), and open-toed shoes are prohibited in the laboratories. Sneakers, rubber clogs without holes, and closed-toe shoes should be worn in the laboratories. When

working with Liquid Nitrogen, closed-toe leather shoes MUST be worn to avoid trapping a spill against the skin.

- Appropriate chemical-resistant gloves as defined in the table, *Resistance to Chemicals of Common Glove Materials* https://www.uhcl.edu/about/administrative-offices/environmental-health-safety/documents/safety-ansell-8th-edition-chemical-resistance-guide.pdf
  shall be worn at all times when there may be skin contact with chemicals. Used gloves shall be disposed of between uses and not re-used. Damaged or deteriorated gloves shall be immediately replaced.
- Thermal-resistant gloves shall be worn in pairs for operations involving the handling of heated materials and exothermic reaction vessels. Insulated freezer-gloves shall be worn in pairs when removing or replacing samples from -30C or -80C freezers. Thermal-resistant and Insulated Freezer-gloves shall be non-asbestos containing and shall be replaced when damaged or deteriorated.
- Respirator usage must be pre-authorized by Environmental Health and Safety and shall comply with the OSHA Respiratory Protection Standard, 29 CFR 1910.134.

Personal Work Practices

- The Department Chair and SCHO must ensure that each employee and all students know and follow the rules and procedures established in this plan.
- All employees and students shall remain vigilant to unsafe practices and conditions in the laboratory and shall immediately report such practices and/or conditions to the laboratory supervisor. The supervisor must correct unsafe practices and or conditions promptly.
- Long hair and loose-fitting clothing shall be confined close to the body to avoid being caught in moving machine/equipment parts.
- Use only those chemicals appropriate for the ventilation system.
- Avoid unnecessary exposure to all chemicals by any route.
- Do not smell or taste any chemicals. Mouth pipetting or siphoning is prohibited.
- Encourage safe work practices in coworkers by setting the proper example. Horseplay is strictly forbidden.
- Seek information and advice from knowledgeable persons or refer to appropriate standards and codes regarding the hazards present in the laboratory. Plan operations, equipment usage and protective measures accordingly.
- Inspect personal protective equipment (PPE) prior to use, and wear appropriate protective equipment as procedures dictate and when necessary to avoid exposure.

Labeling

- All containers in the laboratory shall be labeled. This includes all chemical, biological, and waste containers. Chemicals should have original labels. For others, labels shall be informative and durable, and at a minimum, will identify contents, date of acquisition or formulation, and indication if there is a hazard.
- Portable containers shall be labeled by the individual using the container.

- Exemptions for labeling requirements shall be made for chemical transfers from a labeled container into a container which is intended for the immediate use by the employee or student who performed the transfer.

Criteria for Implementation of Control Measures

Several precautions must be considered when working with chemicals. The following are suggested guidelines for working with chemicals:

1. *Exposure Guidelines*. Most chemicals have some guidelines for exposure, e.g., permissible exposure limits (PELs) or threshold limit values (TLVs). When such values exist, they shall be used by the laboratory supervisor to determine the proper safety precautions needed, including control measures and appropriate personal protective equipment (PPE). When the PEL or TLV for a specific chemical is 50 ppm or less, the chemical must be used inside a chemical fume hood. If the PEL or TLV is not available, the chemical shall be regarded as toxic and used inside a chemical fume hood.

2. *Fire Guidelines*. The flammability of a chemical is generally determined by its flash point, i.e., the lowest temperature at which an ignition source can cause the chemical to ignite momentarily. The flash point shall be used as the reference for "fire hazard" in Biology laboratories along with those guidelines provided by OSHA and the National Fire Protection Association (NFPA). *Flammable* will be used to refer to chemicals with a flash point <100^{o}F (37 ^{o}C). Such chemicals must be stored in a designated flammable storage cabinet. Chemicals with flashpoints between 100-200^{o}F (37-93^{o}C) are considered "combustible" and must be stored away from any source of ignition.

3. *Reactivity Guidelines*. In laboratories, a reactive chemical is one which is ranked by NFPA as "3" or "4" for reactivity or is known to be an oxidizer, organic peroxide, explosive, unstable or subject to polymerization, or reactive with ordinary substances. Once a chemical has been determined to be reactive, all appropriate safety precautions shall be used, including extra segregation in storage and prohibition on mixing with other chemicals without necessary personal protective equipment.

4. *Corrosivity and Contract Hazard Guidelines*. A corrosive chemical is defined by OSHA as a chemical that causes visible destruction of or irreversible alterations in living tissue by chemical actions at the site of contact. A skin or eye contact hazard is one where the chemical's route of entry for its toxic effects is through the skin or eyes. Corrosive or contact hazard chemicals shall be handled only when wearing appropriate eye protection and chemical resistant gloves.

5. *Engineering Controls*. Engineering controls installed in the laboratory are intended to minimize employee and student exposure to chemical and physical hazards in the workplace. These controls must be maintained in proper working order for this goal to be realized. No modifications of engineering controls shall occur unless testing indicates that worker protection will continue to be adequate. Improper function of engineering

controls must be reported to the SCHO immediately and the system shall be taken out of service until proper repairs can be made.

Special Precautions

When laboratory procedures require the use of chemicals classified as allergens, embryotoxins, teratogens, carcinogens, etc., additional special precautions shall be implemented as deemed necessary by the Department Chair, Biosafety Committee, or SCHO. The following best practices shall be considered when working with chemicals classified as allergens or embryotoxins, and chemicals of moderate to high acute toxicity:

Working with Allergens and Embryotoxins:

- Suitable gloves to prevent hand contact shall be worn when exposure to allergens or substances of unknown allergen activity are used in the laboratory.
- Women of child-bearing age shall handle embryotoxins only in a chemical fume hood with confirmed satisfactory performance. They must also use appropriate PPE to prevent skin contact as determined by the laboratory supervisor or SCHO.
- Embryotoxins shall be stored in adequately ventilated areas in unbreakable secondary containers that are clearly labeled.
- The laboratory supervisor and SCHO shall be immediately notified of spills and other exposure incidents. A physician can be consulted when necessary, however the exposure incident must be reported to the university Workers' Compensation Administrator (323-2392) and the medical consultation must be pre-approved.
- Some toxins (refer to MSDS) are considered biologically derived toxins (e.g. Pertussis) and must be disposed of according to biohazardous waste guidelines.

Working with Chemicals of Moderate Chronic or High Acute Toxicity:

- Areas where these chemicals are stored or used shall have restricted access and will be posted with special warning signs.
- Gloves and long sleeves shall be worn and hands and arms will be washed immediately after working with these chemicals.
- Two people shall always be present in the laboratory during operations which utilize these chemicals.

Working with Chemicals of High Chronic Toxicity:

- Approval of the Department Chair and SCHO must be obtained prior to using these chemicals.
- Areas where these chemicals are stored or used shall have restricted access and will be posted with special warning signs.
- All work with these substances shall be conducted in a restricted access hood, glove box or isolated portion of the lab.
- Any contaminated equipment or glassware shall be decontaminated in the hood before removing from the designated area.

- For powders, a wet mop or vacuum with a HEPA filter shall be used for cleanup.
- Containers shall be stored in a well-ventilated, limited access area in prominently labeled, unbreakable, chemically resistant, secondary containers.

Best Practices for Chemical Spills and Accidents

Laboratory supervisors shall ensure the appropriate materials to handle spills are available in their laboratory. Minor spills shall be cleaned up immediately by laboratory personnel, the area shall be properly cleaned and if necessary, ventilated. If spilled material is hazardous, all spill debris and clean-up materials must be containerized and labeled as hazardous waste.

In the event of a major spill (threat to public health, safety or the environment), evacuate and close the laboratory and call Campus Police at 323-3333. Fire extinguishers are located in hallways and in most laboratories; fire alarm pull stations are located at the end of each hallway and should be used to evacuate the building if necessary.

Hazardous Chemical Waste

The generation of chemical waste is an unavoidable part of teaching biology and conducting biological research. The collection, storage and disposal of generated chemical waste is regulated under the Environmental Protection Agency's (EPA) Resource Conservation and Recovery Act (RCRA) and the South Carolina Department of Health and Environmental Control (SC-DHEC) Hazardous Waste Regulations. A waste is a solid, liquid or compressed gas that is to be discarded, recycled, or is considered inherently waste-like. EPA and SC-DHEC have classified hazardous wastes into two categories: characteristic hazardous wastes or listed hazardous wastes.

Characteristic hazardous wastes are materials that exhibit at least one of the following characteristics:

Ignitable Waste
- Is a liquid with a flash point < 140^{o}F (60^{o}C)
- Is not a liquid, but is capable under normal conditions of causing fire through friction, absorption of moisture, or spontaneous chemical changes
- Is an ignitable compressed gas including some aerosol cans
- Is an oxidizer

Corrosive Waste
- Is an aqueous solution with a pH $\leq$ 2 or $\geq$ 12.5
- Is a liquid which can corrode steel at a rate > 0.250 inches per year at 130^{o}F (55^{o}C)

Reactive Waste
- Is normally unstable and readily undergoes violent change without detonation
- Reacts violently with water
- Forms potentially explosive mixtures with water

- Generates toxic gasses, vapors or fumes when mixed with water
- Is a cyanide or sulfide waste that generates toxic gasses, vapors or fumes at pH conditions between 2 and 12.5
- Is capable of detonation or explosive reaction if subjected to strong initiating source or if heated under confinement
- Is classified as a Department of Transportation explosive

Toxic Waste

- Exhibits toxicity when an extract of a representative sample contains any of the contaminants identified in Table 1. 40 CFR 261.24 (Appendix E) at a concentration greater than the respective value given in the table.

EPA and SC-DHEC have identified approximately 500 chemicals in four published lists classified as listed hazardous wastes. Listed hazardous wastes are primarily process chemical wastes or off-specification commercial chemical products as defined below:

- F-listed wastes are spent halogenated and non-halogenated solvent generated from non-specific sources.
- K-listed wastes are materials generated from specific industrial sources (Winthrop University should not have any K-listed wastes)
- U-listed wastes are discarded commercial chemical products, off-specification chemicals, container residues and spill residues thereof (Appendix F).
- P-listed wastes are acutely hazardous commercial chemical products, off-specification chemicals, container residues and spill residues thereof (Appendix F).

Note: Both P- and U-listed wastes are commercially pure or technical grades of the chemical listed or are the sole active ingredient when in formulation.

Responsibilities of Hazardous Waste Generators

The principal investigator, laboratory supervisor, professor/instructor-in-charge is the hazardous waste generator, responsible for hazardous waste collection, storage and management in their laboratory. Hazardous waste generators in the Department of Biology have a responsibility:

- To select chemicals carefully, become familiar with their individual hazards, and to manage and dispose of all hazardous wastes in compliance with EPA, RCRA, and DHEC regulations and Winthrop University policies.
- To properly identify hazardous wastes, select compatible containers, and to segregate and store hazardous wastes to ensure the safety of those working in the laboratory.
- To ensure that all hazardous waste containers are properly labeled and kept clean of waste residue.
- To ensure that hazardous waste containers are always kept closed except when adding or removing wastes from the container. A funnel in a waste container is not considered closed.
- To ensure that different waste streams (i.e., radioactive, chemical or biological) are not mixed together. Separate waste materials as much as is feasible – if you must combine materials, try to keep the chemistry as pure as possible, i.e., do not mix aqueous solutions

with organic solvents. Also, make sure the mixture is compatible – do not create a chemical reaction inside the waste container!
- To initiate a meaningful waste minimization plan through substitution, scale reduction, purchase control, and/or recycling.
- To ensure that students working in the laboratory also understand these responsibilities.

Reminder: No sink or sewer disposal of chemicals is permitted without permission of Environmental Health and Safety. For questions about a specific waste, call 323-2328.

Collection and Storage of Hazardous Chemical Wastes

At Winthrop University, Environmental Health and Safety provides institutional oversight for the proper collection, storage and disposal of hazardous chemical wastes. Working in conjunction with EHS, the SCHO provides oversight and management within the Department of Biology. Laboratory hazardous wastes are best collected and stored in a designated satellite accumulation area (SAA). An SAA must be near the point of generation of the waste and under the direct control of the principal investigator or laboratory supervisor. In the Department of Biology, various specific locations have been identified as SAAs (Appendix G). The following institutional policies shall apply to each SAA with this information posted in a visible location:

- Flammable or combustible wastes should be stored in a flammables storage cabinet to meet fire code restrictions.
- Hazardous wastes should not be stored on the floor unless secondary containment is used and the material is stored away from exits and/or egress pathways.
- Under EPA regulation 40 CFR 264.175, aqueous hazardous waste containers shall be stored in secondary containment, like a plastic drip tray, to mitigate the severity of any spills from a hazardous waste container. The secondary container must be large enough to contain 10% of the total volume of primary waste containers or 100% of the largest waste container contained inside it, whichever is greater.
- Hazardous waste containers must be compatible with the material being accumulated and must be closed at all times except when waste is being added or removed. Regulations do not permit funnels in waste containers except when material is being added. The maximum size allowable for collection containers is **4 liters** for hazardous wastes and **100 ml** for acutely hazardous wastes.
- All hazardous waste containers must be labeled at the time waste is first put into the container. Pre-printed, self-adhesive labels are available from the SCHO, 132 Dalton Hall, and should be used for all hazardous waste containers. **The following label information must be provided by the waste generator:**

    1. Name and phone number of Principal Investigator/Laboratory Supervisor (waste generator):
    2. University department, building and room number (use the lab room number, not your office):
    3. Contents of the container, listing the names of all chemicals added to the container (use full chemical names only - abbreviations and chemical formulas are not acceptable); and the percentage of each chemical if more than one is added to the container;

- When completing the pre-printed label, do not fill in the **accumulation date**; it will be added to the label when the container is moved to the central waste accumulation area (chemical storage building). Also, do not fill in the **EPA Hazardous Waste Code** unless you have been trained to do so, EHS will add this information when the waste is moved to the central accumulation area.

Disposal of Hazardous Chemical Wastes

Environmental Health and Safety is responsible for the management, storage and disposal of hazardous wastes once they are moved from the SAA to the waste accumulation area located in the shared chemical storage building between Sims Science Building and Dalton Hall. Once a waste container is full or a waste stream is obsolete, the principal investigator/laboratory supervisor should contact either the SCHO (x6431) or EHS (x2328) to arrange for container pick-up. The principal investigator/laboratory supervisor is not to move waste from the SAA to the waste accumulation area or to any other SAA in Dalton Hall. Prior to moving the container(s), EHS will check the containers for proper labeling, compatibility with contents, and a secure seal. As noted in the SAA policies, principal investigators/laboratory supervisors are not to determine the EPA Hazardous Waste Code or add the accumulation date to the container label.

EHS is responsible for container management and storage once waste has been moved to the waste accumulation area. EHS will take care of any additional packaging or labeling requirements for transportation and will make all contractual arrangements for waste pick-up and disposal.

Disposal of Ethidium Bromide

Ethidium bromide is not regulated as hazardous waste; however, the mutagenic properties of this substance may present a hazard if it is poured down the drain untreated or placed in the trash. Based on these considerations, the following disposal procedures for ethidium bromide should be followed:

For gels:

- Electrophoresis gels with trace amounts of ethidium bromide should not pose a hazard and may be disposed of in the trash.
- Higher concentrations of ethidium bromide in gels (e.g., when the color of the gel is dark pink or red) should not be placed in laboratory trash, but should be incinerated. Accumulate gels in a proper container, label them as non-regulated waste, and have them picked-up by Environmental Health and Safety for disposal.

For solutions containing ethidium bromide:

- Aqueous solutions containing <10µg/ml ethidium bromide can be released to the drain.
- Aqueous solutions containing >10µg/ml ethidium bromide should be deactivated using Amresco destaining bags. The Amresco destaining bags are small "tea bags" which contain an acid/base mixed bed of resin. The resin can absorb 1-5 mg of ethidium bromide from

gel solutions and running buffers after gentle overnight agitation. The intent of these bags is to remove hazardous dyes like ethidium bromide from gel solutions for easy disposal. For example, if you have a 200 ml solution with 2 g/L of ethidium bromide, the solution contains 200 mg of dye. To remove all the dye from the solution would require (200/5 =) 40 destaining bags. Amresco destaining bags can be obtained from the department Lab Manager, 132 Dalton Hall. The procedure for using Amresco destaining bags is as follows:

1. Place the Amresco "tea" bag into the ethidium bromide solution.
2. Allow to sit overnight.
3. Pour filtrate down the drain.
4. Collect the "used" destaining bags for disposal as non-regulated waste, following the same procedure as for heavily stained gels.

Disposal of Universal Wastes

Used fluorescent lamps including compact fluorescents, mercury containing equipment including thermometers, and used sealed lead acid batteries are classified by EPA and SC-DHEC as universal wastes. Although these materials meet the definition of toxic hazardous waste, management of them is exempt from full regulation in order to encourage recycling of these materials. Winthrop University has an active universal waste collection program managed by Environmental Health and Safety. Additionally, EHS collects any type of used battery including rechargeable batteries, used high intensity discharge lamps, and used UV/germicidal lamps for recycling. Do not put these materials in the trash; take them to the Lab Manager, 132 Dalton Hall for pick-up by EHS or Facilities Management personnel. Any broken lamps or bulbs should be carefully discarded in broken glass waste boxes.

Although the department no longer uses mercury thermometers, some may be still be present. Mercury thermometers can be identified by the silver liquid inside; a red liquid usually indicates an alcohol thermometer. If you find a mercury thermometer, take it to the Laboratory Manager in 132 Dalton Hall.

Disposal of Serological Pipettes

Used disposable serological pipettes, although not considered sharps, represent a potential hazard to custodians as they can puncture garbage bags. As such, they are not to be disposed into regular trashcans. Serological pipettes that have contacted biohazardous materials shall be disposed of in biohazardous waste and appropriately decontaminated. Nonbiohazardous used serological pipettes shall be disposed into cardboard boxes or cardboard glass waste containers; after accumulation, the containers may be disposed into garbage dumpsters.

Section H: Biosafety Plan

The Biosafety Plan is an integral part of the Health, Safety and Security Plan of the Department of Biology, Winthrop University. The purpose of this plan is to protect personnel and students from exposure to biohazardous materials and agents and to prevent the release of biohazardous materials and agents into the environment. To this end, it provides procedures and instructions intended to ensure compliance with federal, state and institutional biosafety guidelines. It is the responsibility of the Principal Investigator/ Instructor to implement these procedures and guidelines, and to oversee the proper protection of students and personnel in their teaching or research labs. It is the responsibility of the Department of Biology Biosafety Committee to inform all faculty and students about these guidelines and to oversee their implementation within the department.

Biosafety Levels

Biosafety encompasses techniques, procedures and applications of knowledge to prevent the exposure of laboratory personnel (faculty, staff and students) to biohazardous agents as well as prevent the release of biohazardous agents into the environment. Biohazardous agents are those biological agents – bacteria, viruses, fungi, parasites, recombinant products, allergens, cultured human and animal cells, etc., that have the potential to be hazardous to humans, animals and plants.

Risk Groups of Biohazardous Agents

Biohazardous agents are classified into four risk groups according to the hazard(s) they present to humans, animals, and the environment. This classification is based on the pathogenicity of the agent, its mode of transmission, host range, preventive measures, treatments, etc. According to the *NIH Guidelines for Research on Recombinant DNA* (April 2019), human etiological agents are classified in four risk groups:

- Risk Group 1 (RG1) agents are not associated with disease in healthy adult humans.
- Risk Group 2 (RG2) agents are associated with human disease which is rarely serious and for which preventive or therapeutic interventions are often available.
- Risk Group 3 (RG3) agents are associated with serious or lethal human disease for which preventive or therapeutic interventions may be available. (high individual risk but low community risk)
- Risk Group 4 (RG4) agents are likely to cause serious or lethal human disease for which preventive or therapeutic interventions are not usually available (high individual risk and high community risk).

In the CDC/NIH publication, *Biosafety in Microbiological and Biomedical Laboratories* (BMBL) 6th ed., 2020, four levels of biosafety are defined based upon the risk associated with the biological agents involved and level of control required to protect personnel, the community, and the environment. Most microbiological and biomedical work within the Department of Biology is conducted at Biosafety Level 1 (BSL-1) or Biosafety Level 2 (BSL-2) containment.

According to the BMBL the four levels of biosafety are defined as:

- Biosafety Level 1 (BSL-1). BSL-1 contains biological agents that pose low risk to personnel and the environment. These agents are highly unlikely to cause disease in healthy laboratory workers, animals or plants and require BSL-1 containment. BSL-1 is appropriate for undergraduate and graduate research and for secondary educational training and teaching laboratories. BSL-1 is suitable for work involving well-characterized agents not known to consistently cause disease in healthy adult humans, and of minimal potential hazard to laboratory personnel and the environment. The laboratory is not necessarily separated from the general traffic patterns in the building and work is generally conducted on open bench tops using standard microbiological practices. Special containment equipment or facility design is neither required nor generally used, however laboratory personnel should receive lab-specific training under the supervision of the Principal Investigator/Instructor. Examples of BSL-1 agents are: *Agrobacterium radiobacter*, *Aspergillus niger*, *Bacillus thuringiensis*, *Escherichia coli* strain K12, *Lactobacillus acidophilus*, *Micrococcus luteus*, *Neurospora crassa*, *Pseudomonas fluorescens*, *Serratia marcescens* (for a complete list of biosafety levels for bacteria see to Appendix H).

- Biosafety Level 2 (BSL-2). BSL-2 builds upon BSL-1 and is suitable for work involving agents associated with human disease and pose moderate potential hazards to personnel and the environment. BSL-2 differs from BSL-1 in that (1) laboratory personnel must have specific training in handling pathogenic agents and are supervised by scientists competent in handling infectious agents and associated procedures; (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. Examples of BSL-2 agents are: *Streptococcus pneumonia*, *Salmonella choleraesuis and Staphylococcus aureus*.

- Biosafety Level 3 (BSL-3). BSL-3 is applicable to clinical, diagnostic, teaching, research, or production facilities in which work is done with indigenous or exotic agents which may cause serious or potentially lethal disease as a result of exposure by inhalation. Laboratory personnel must have specific training in handling pathogenic and potentially lethal agents, and are supervised by competent faculty who are experienced in working with these agents. Examples of BSL-3 agents are: *M. tuberculosis*, St. Louis encephalitis virus, West Nile virus, *Coxiella burnetii*, the agents causing anthrax, Venezuelan equine encephalitis, Eastern equine encephalitis, SARS, tuberculosis, typhus, Rift Valley fever, Rocky Mountain spotted fever, yellow fever. **BSL-3 agents are prohibited in the Department of Biology, Winthrop University.**

- Biosafety Level 4 (BSL-4). BSL-4 is assigned to work with dangerous and exotic agents that pose a high individual risk of aerosol-transmitted laboratory infections and life-threatening disease. Laboratory personnel must have very specific and thorough training in handling these extremely hazardous infectious agents, including an understanding of primary and secondary containments, containment equipment, and laboratory design. Personnel must be supervised by competent scientists who are trained and experienced in

working with these agents. Access to the laboratory is strictly controlled and the facility is isolated either in a separate building or in a controlled area within the building. A specific facility operations manual must be prepared and adopted.

Within work areas of the facility, all activities are confined to Class III biological safety cabinets, or Class II biological safety cabinets used with one-piece positive pressure personnel suits ventilated by a life support system. BSL-4 requires special engineering and design features to prevent microorganisms from being disseminated into the environment. Examples of BSL-4 agents are: Marburg Virus, Ebola Virus, Hantaviruses, and the agents which cause Bolivian and Argentine Hemorrhagic Fevers, Smallpox, Lassa Fever, Crimean-Congo Hemorrhagic Fever, and other various hemorrhagic diseases. **BSL-4 agents are prohibited in the Department of Biology, Winthrop University.**

Biosafety Training

All students must take and pass a Basic Laboratory Safety Training Course as part of their first biology laboratory experience. All faculty and research students working with any bacterial, fungal, human, primate and/or mammalian cell lines must additionally complete and pass the Biosafety Training modules of the Collaborative Institutional Training Initiative (CITI) Program. In all cases, the principal investigator/instructor must ensure that students working under their supervision receive appropriate orientation and specific training for the safe performance of their work with biohazardous agents. This training shall include:

- Communication about the potential hazards of working with cell lines;
- Instruction on appropriate work practices and the engineering controls used to minimize exposure;
- Instruction as to the appropriate personal protective equipment and its proper use;
- Review of laboratory standard operating procedures; and
- Directions as to what to do in case of personal exposure or accidental release of biohazardous materials.

Human or primate cell lines may harbor viruses, bacteria, or parasites characterized as human bloodborne pathogens. For example, these pathogens can include Human Immune-deficiency Virus (HIV), Hepatitis B or C viruses, *Neisseria*, Treponema, or *Plasmodium*. Please note, some bio-resource organizations, such as American Type Culture Collection (ATCC), do not typically screen their material for bloodborne pathogens. Consequently, the handling of human and primate cell lines must conform to the OSHA Exposure Control Standard, generally known as the Bloodborne Pathogens (BBP) Standard. Compliance with this standard includes annual training with information on hazard communication, engineering controls, work practices, personal protective equipment (PPE), housekeeping, a written exposure control plan, and access to the Hepatitis B vaccine. (*Note: This section of the manual has been adapted with permission from Cornell University's Institutional Biosafety Committee.)*

For a list of biohazardous agents frequently used in the Department of Biology, see Appendix I.

Administrative Regulations

To ensure the safety of faculty and students working with biohazardous agents the following administrative regulations must be followed:

- Appropriate warning signs must be posted on laboratory doors and equipment, such as a biosafety level sign that identifies the biohazardous agent and "UV light source" sign to indicate that UV light is used in the laminar flow hoods to disinfect the hood.
- All labs should have posted emergency contact information in a highly visible location both inside the laboratory and on the laboratory door (facing the hall). At minimum, this should include the name and telephone numbers of the principle investigator, Campus Police, and the SCHO/Laboratory Manager.
- Biohazard warning labels should be attached to all biohazardous waste containers, refrigerators/freezers where biohazardous materials are stored, and to equipment that is used for biohazardous materials.
- Faculty that have research projects and classroom activities involving work with biohazardous agents and rDNA (BSL-1 and BSL-2) are required to file a Biohazardous Materials Declaration Form (http://www.winthrop.edu/spar/Biosafety.htm) with the Institutional Biosafety Committee (IBC) for review. The form should be submitted to Michele Smith (smithmr@winthrop.edu) one month prior to beginning the experiments. When appropriate the principle investigator/professor should additionally submit an IRB form http://www.winthrop.edu/spar/Human%20Subjects.htm and/or an IACUC form http://www.winthrop.edu/spar/iacuc.htm.
- BSL-1 and BSL-2 teaching and research laboratories should have an integrated pest management program in place or pesticide service as needed.
- Violations of the biosafety standards outlined or referenced in this plan should be reported to the Biology Biosafety Committee; the committee will determine if the violation warrants attention by the Department Chair.
- Accidents involving biohazardous materials must be reported to the SCHO, the Biology Biosafety Committee, and Environmental Health and Safety using the Accident Report Form (see Appendix A).

Engineering Controls

To ensure the safety of faculty and students working with biohazardous materials, the following general engineering controls should be followed:

- Hand washing facilities are available in the laboratory and must be used particularly after handling infectious material or animals, after removing gloves, before leaving the lab. Soap must be used to wash hands.
- Principal Investigator/Instructor must ensure that everyone in the lab knows the location of a readily accessible eyewash station and spill kits.
- In laboratories generating biohazardous waste, the principal investigator/instructor shall ensure the waste is treated by appropriate chemical disinfection (i.e., 1 to 10 bleach solution or equivalent) or steam sterilization (proper sterilization is achieved when the load is autoclaved at 250ºF or 121ºC for a minimum of 30 minutes). Heat sensitive test strips (e.g.

autoclave tape) or other indicator of proper heat treatment is used with each autoclaved container.
- Biosafety cabinets shall be certified as soon as possible after installation and/or relocation, and shall be recertified annually.
- Laboratory is organized so that it can be easily cleaned, including between fixtures. Floors, ceiling, and furnishings should be kept in good repair. Furnishings should be durable and benchtops impervious to water and resistant to lab chemicals and moderate heat.
- Illumination should be sufficient in all work areas to support working safely.

Standards for Biosafety Level 1

Biosafety Level 1 (BSL-1) is suitable for work involving well-characterized agents not known to consistently cause disease in immunocompetent adult humans, and present minimal potential hazard to laboratory personnel and the environment. BSL-1 laboratories are not necessarily separated from the general traffic patterns in a building. Work is typically conducted on open bench tops using standard microbiological practices. Special containment equipment or facility design is not required, but may be used as determined by appropriate risk assessment. Laboratory personnel must have specific training in procedures conducted in the laboratory and must be supervised by a scientist with training in microbiology or a related science. Per the BMBL, 6th ed. (2019), the following standard practices, safety equipment, and facility requirements apply to agents assigned to BSL-1. All faculty and students must adhere to these standards. Academic and research labs working with BSL-1 reagents will be subject to random inspections to ensure compliance with standards. The following standards from the BMBL, 6th ed., (2019) apply to agents assigned to BSL-1:

A. Standard Microbiological Practices

1. Access to the laboratory is limited or restricted at the discretion of the laboratory supervisor when experiments or work with cultures and specimens are in progress.
2. Persons must wash their hands after working with potentially hazardous materials, after removing gloves, and before leaving the laboratory.
3. Long hair is restrained so that it cannot contact hands, specimens, containers, or equipment.
4. Eating, drinking, smoking, handling contact lenses, applying cosmetics, and storing food for human consumption must not be permitted in the laboratory areas. Persons who wear contact lenses in laboratories should also wear goggles or a face shield. Food must be stored outside the laboratory in cabinets or refrigerators designated and used for this purpose.
5. Mouth pipetting is prohibited; mechanical pipetting devices must be used.
6. Policies for the safe handling of sharps must be developed and implemented. Whenever practical laboratory supervisors should adopt improved engineering and work practice controls that reduce risk of sharps injuries. Precautions including those listed below, must always be taken with sharp items. These include:
    a. Careful management of needles and other sharps are of primary importance. Needles must not be bent, sheared, broken, recapped, removed from disposable syringes, or otherwise manipulated by hand before disposal.

b. Used disposable needles and syringes must be carefully placed in conveniently located puncture-resistant containers used for sharps disposal.
c. Non-disposable sharps must be placed in a hard walled container for transport to a processing area for decontamination (if necessary), preferably by autoclaving.
d. Broken glassware must not be handled directly. Instead, it must be removed using a brush and dustpan, tongs, or forceps. Plastic ware should be substituted for glassware whenever possible

6. Perform all procedures to minimize the creation of splashes and/or aerosols.
7. All chemicals, reagents, and biohazardous materials should be properly labeled with complete information of the contents easily identified.
8. Decontaminate work surfaces after completion of work and after any spill or splash of potentially infectious material with appropriate disinfectant.
9. Decontaminate all cultures, stocks, and other potentially infectious materials before disposal using an effective method such as autoclaving. Materials to be decontaminated outside of the immediate laboratory must be placed in a durable, leak-proof container and closed for transport from the laboratory. Materials to be removed from the facility for decontamination must be packaged in accordance with applicable local, state, and federal regulations.
10. A biohazard sign must be posted at the entrance to the laboratory when infectious agents are present. The sign may include the name of the agent(s) in use and the name and phone number of the investigator or other responsible personnel.
11. An effective integrated pest management program is required.
12. Animals and plants not associated with the work being performed are not permitted in the laboratory.
13. The laboratory supervisor must ensure that laboratory personnel receive appropriate training regarding their duties, the necessary precautions to prevent exposures, and exposure evaluation procedures.

B. Special Practices

None required.

C. Safety Equipment

1. Special containment devices or equipment, such as a biological safety cabinet (BSC) are not generally required.
2. Protective laboratory coats, gowns, or uniforms are recommended to prevent contamination of personal clothing.
3. Wear protective eyewear when conducting procedures that have the potential to create splashes of microorganisms or other hazardous materials. Persons who wear contact lenses in the laboratory should also wear eye protection.
4. Gloves must be worn to protect the hands from exposure to hazardous materials. Glove selection should be based on an appropriate risk assessment. Alternatives to powdered latex gloves should be available. Wash hands prior to leaving the laboratory. In addition, BSL-1 workers should:

a. Changes gloves when contaminated, glove integrity is compromised, or when otherwise necessary.
b. Remove gloves and wash hands when work with hazardous materials has been completed and before leaving the laboratory.
c. Do not wash or reuse disposable gloves. Dispose of used gloves with other contaminated laboratory waste. Hand washing protocols must be rigorously followed.
d. Closed-toe shoes must be worn.
e. No athletic shorts permitted, longer attire (to the knee at least) is required.

C. Laboratory Facilities

1. Laboratories should have doors for access control.
2. Each laboratory must have a sink for handwashing.
3. The laboratory is organized so that it can be easily cleaned. Carpets and rugs in laboratories are not appropriate.
4. Laboratory furniture must be capable of supporting anticipated loads and uses. Spaces between benches, cabinets, and equipment should be accessible for cleaning.
    a. Bench tops must be impervious to water and are resistant to heat, organic solvents, acids, alkalis, and other chemicals.
    b. Chairs used in the laboratory must be covered with a non-porous material that can be easily cleaned and decontaminated with appropriate disinfectant.
5. Laboratory furniture is capable of supporting anticipated loading and uses. Spaces between benches, cabinets, and equipment are accessible for cleaning.
6. Laboratory windows that open to the exterior should be fitted with screens.

Standards for Biosafety Level 2

Biosafety Level 2 builds upon BSL-1 and is suitable for work involving agents that pose moderate hazards to personnel and the environment. It differs from BSL-1 in that: 1) laboratory personnel have specific training in handling pathogenic agents and are supervised by scientists competent in handling infectious agents and associated procedures; 2) access to the laboratory is restricted when work is being conducted; and 3) all procedures in which infectious aerosols or splashes may be created are conducted in biosafety level II cabinets or other physical containment equipment. All faculty and students must adhere to these guidelines. Academic and research labs working with BSL-2 reagents will be subject to random inspections to ensure compliance with standards. In addition to the standard practices, safety equipment, and facility requirements for BSL-1, the following requirements apply to agents assigned BSL-2:

A. Standard Microbiological Practices

…are the same as for BSL-1.

B. Special Practices

1. Access to the laboratory is controlled when work is being conducted.
2. All persons entering the laboratory must be advised of the potential hazards and meet specific entry/exit requirements.
3. Laboratory personnel must be provided medical surveillance, as appropriate, and offered available immunizations for agents handled or potentially present in the laboratory.
4. Each institution should consider the need for collection and storage of serum samples from at-risk personnel.
5. A laboratory-specific biosafety manual must be prepared and adopted as policy. The biosafety manual must be available and accessible.
6. The laboratory supervisor must ensure that laboratory personnel demonstrate proficiency in standard and special microbiological practices before working with BSL-2 agents.
7. Potentially infectious materials must be placed in a durable, leak proof container during collection, handling, processing, storage, or transport with a facility.
8. Laboratory equipment should be routinely decontaminated, as well as, after spills, splashes, or other potential contamination.
    a. Spills involving infectious material must be contained, decontaminated, and cleaned up by staff properly trained and equipped to work with infectious material.
    b. Equipment must be decontaminated before repair, maintenance, or removal from the laboratory.
8. Incidents that may result in exposure to infectious materials must be immediately evaluated and treated according to procedures described in the laboratory biosafety manual. All such incidents must be reported to the laboratory supervisor. Medical evaluation, surveillance, and treatment should be provided and appropriate records maintained.
9. Animals and plants not associated with the work being performed must not be permitted in the laboratory.
10. All procedures involving the manipulation of infectious materials that may generate an aerosol should be conducted within a BSC or other physical containment devices.

C. Safety Equipment (in addition to requirements for BSL-1)

1. Properly maintained BSCs, preferably Class II, or other appropriate personal protective equipment, or other physical containment devices must be used whenever:
    a. Procedures with a potential for creating infectious aerosols or splashes are conducted. These may include pipetting, centrifuging, grinding, blending, shaking, mixing, sonicating, opening containers of infectious materials, 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 using sealed rotor heads or centrifuge safety cups are used. Rotor heads and safety cups should be opened only in a BSC.
2. Protective laboratory coats, gowns, smocks, or uniforms designated for laboratory use must be worn while working with hazardous materials. Remove protective clothing before leaving for non-laboratory areas, (e.g., cafeteria, library, and administrative offices). Dispose of protective clothing appropriately, or deposit it for laundering by the institution. It is recommended that laboratory clothing not be taken home.

3. Eye and face protection (goggles, mask, face shield or other splatter guard) is used for anticipated splashes or sprays of infectious or other hazardous materials when the microorganisms must be handled outside the BSC or containment devices. Eye and face protection must be disposed of with other contaminated laboratory waste or decontaminated before reuse. Persons who wear contact lenses in laboratories should also wear eye protection.
4. Gloves must be worn to protect hands from exposure to hazardous materials. Glove selection should be based on an appropriate risk assessment. Alternatives to latex gloves should be available. Gloves must not be worn outside the laboratory, In addition, BSL-2 laboratory workers should:
    a. Change gloves when contaminated, glove integrity is compromised, or when otherwise necessary.
    b. Remove gloves and wash hands when work with hazardous materials has been completed and before leaving the laboratory.
    c. Do not wash or reuse disposable gloves; do not touch "clean" surfaces while wearing gloves. Dispose of used gloves with other contaminated laboratory waste. Hand washing protocols must be rigorously followed.
    d. Wearing two pairs of gloves may be appropriate.

D. Laboratory Facilities (in addition to requirements for BSL-1)

1. Laboratory doors should be self-closing and have locks in accordance with institutional policies.
2. BSCs must be installed so that fluctuations of the room air supply and exhaust do not interfere with proper operations. BSCs should be located away from doors, windows that can be opened, heavily traveled areas, and other possible airflow disruptions.
3. Vacuum lines should be protected with liquid disinfectant traps.
4. An eyewash station must be readily available.
5. There are no specific requirements for ventilation systems, however, planning of new facilities should consider mechanical ventilation systems that provide an inward flow of air without recirculation to spaces outside the laboratory.
6. HEPA filtered exhaust air from a Class II BSC can be safely recirculated back into the laboratory environment if the cabinet is tested and certified at least annually and operated according to manufacturer's recommendations. BSCs can also be connected to the laboratory exhaust system by either a thimble (canopy) connection or directly exhausted to the outside through a hard connection. Provisions to assure proper safety cabinet performance and air system operation must be verified.
7. A method for decontaminating all laboratory wastes should be available in the facility (e.g., autoclave, chemical disinfection, incineration, or other validated decontamination method).

Recombinant DNA Research

All recombinant DNA (rDNA) research/instructional work should comply with the most recent *NIH Guidelines for Research Involving Recombinant DNA Molecules (2020)* and the CDC/NIH *Biosafety in Microbiological and Biomedical Laboratories* (BMBL) 6th ed., (Rev. 2020). In the *NIH Guidelines*, "recombinant and synthetic nucleic acids are defined as either: (i) molecules that are constructed outside living cells by joining natural or synthetic nucleic acid molecules that can replicate in a living cell, i.e., recombinant nucleic acids; (ii) nucleic acid molecules that are chemically or by other means synthesized or amplified, including those that are chemically or otherwise modified but can base pair with naturally occurring nucleic acid molecules, i.e., synthetic nucleic acids, or (iii) molecules that result from the replication of those described in (i) or (ii) above. Synthetic DNA segments which are likely to yield a potentially harmful toxin or a pharmacologically active agent are considered as equivalent to their natural DNA counterpart. If the synthetic DNA segment is not expressed *in vivo* as a biologically active polynucleotide or polypeptide product, it is exempt from the *NIH Guidelines*. Genomic DNA of plants and bacteria that have acquired a transposable element, are not subject to the *NIH Guidelines* unless the transposon itself contains recombinant DNA".
It is the responsibility of the Principal Investigator/Instructor to review these guidelines and to determine the level of biohazard involved in the rDNA experiments.

According to NIH, there are six categories of rDNA research, all requiring prior Institutional Biosafety Committee (IBS) approval, except **III-F**. Different categories have <u>additional</u> requirements prior to initiation (see below). For more details on all categories, refer to the *NIH Guidelines*. Categories of rDNA experiments include:

- Section **III-A** experiments require recombinant DNA review by the Office of Science Policy (OSP), National Institutes of Health and the Institutional Biosafety Committee (IBS) for approval before initiation. Under this category fall experiments considered as "Major Actions" under the NIH Guidelines; and experiments incorporating the deliberate transfer of drug resistance genes to microorganisms that can compromise the control of disease agents.
- Section **III-B** experiments involve cloning of toxin molecules lethal for vertebrates at an LD50 of less than 100 nanograms per kilogram body weight (e.g., microbial toxins such as the botulinum toxins, tetanus toxin, diphtheria toxin, and Shigella dysenteriae neurotoxin). It requires prior submission of a proposal to the NIH/ OSP for review and to the IBS for approval before initiation of any experiments.
- Section **III-C** experimen involve the deliberate transfer of recombinant or synthetic nucleic acid molecules, or DNA or RNA derived from recombinant or synthetic nucleic acid molecules, into one or more human research participants. These experiments require IRB (Institutional Review Board) approval before initiation.
- Section **III-D** refers to whole animal, animal tissue culture experiments or plant experiments with different (restricted or non-pathogenic) host-vector systems using Risk Group 2, Risk Group 3, Risk Group 4 agents. Required containment facilities should match the RG level code. In this category are experiments that aim to produce transgenic animals, and those that involve transgenic animals. Also included in this category are experiments aiming genetic engineering of plants by rDNA methods that pose risk to the environment

or express vertebrate toxins; or use of such plants for experimental purposes or propagation of such plants, or the use of plants together with microorganisms or insects, or small animals containing rDNA.

- Section **III-E** deals with creating stable germline alterations of rodents using rDNA and requires filing with the IBC at the time the experiment is initiated. The IBC reviews and approves all such proposals, but Biosafety Committee review and approval prior to initiation of the experiment is not required. All such experiments may be conducted at BSL-1 containment.
- Section **III-F** experiments are exempt and registration with the IBC is not required. This category includes experiments that are not in organisms or viruses and those that do not present a significant health or environmental risk.

Standard Practices and Training Involving rDNA Research

In the Department of Biology work with rDNA is applicable to the best practices for work and containment included under BSL-1 and BSL-2 guidelines described in Section H. This section applies to rDNA-transgenic plants biosafety levels and containment. The principle purpose of plant containment is to avoid unintentional transmission of recombinant DNA-containing plants in the environment. Such a principle assumes that the genetically modified plants do not pose any health risk to humans and animals.

Responsibilities of Principal Investigator/Faculty for rDNA Research

Faculty that plan to carry out rDNA research are expected to be adequately trained in safe, general microbial techniques as well as to comply with and sustain the appropriate BSL work and containment requirements. Prior to initiation of research faculty must determine the category of their rDNA research. For experiments that do not fall under category III-F, a Biohazardous Materials Declaration Form must be filed for approval with the IBC. The following guidelines for rDNA work must be adhered to:

- Faculty shall not modify or initiate rDNA research without the prior approval of IBC.
- Faculty shall report and correct any significant problems, accidents, violations of guidelines to the department Biosafety Committee and Department Chair.
- Faculty shall adhere to the guidelines of this Health, Safety, and Security Plan as well as university policies related to health and safety.
- Faculty shall comply with shipping requirements for recombinant DNA molecules as described in *NIH Guidelines*.
- Faculty shall ensure that all students have successfully completed the *Basic Laboratory Safety Training Course* prior to participation in research. Faculty shall instruct and train students in all new practices and techniques to ensure safety of students. Faculty shall supervise students to ensure that all safety practices are employed.
- Faculty shall investigate and report in writing any significant problems pertaining to the operation and implementation of containment practices and procedures to the department Biosafety Committee and Department Chair.
- Faculty shall make available to students and lab personnel all protocols and MSDS that describe potential biohazards and associated precautions.

- Faculty will periodically confirm the availability of spill kits in the lab and review proper use with students.
- Faculty shall inform the laboratory staff of the reasons and provisions for any precautionary medical practices advised or requested (e.g., vaccinations or serum collection).

(Note: To prepare this section NIH Guidelines on Recombinant DNA, 2019, were used as well as some materials from the Biosafety Manual of Oregon State University, used with permission.)

Exposure to Biohazardous Materials

In the event of an accidental exposure:

- Wash the exposed area thoroughly with soap and running water. Use non-abrasive, antibacterial soap if possible.
- **If biohazardous material is splashed in the eye or mucous membrane, flush the affected area with running water for at least 15 minutes.**
- Report the exposure to SCHO/Lab Manager, 132 Dalton Hall, as soon as possible.
- Should medical treatment be needed, non-employee students should go directly to Student Health Services, Crawford Building. Employees including student employees should follow the procedures for reporting an injury or work-related illness found in Section D. Non-emergency medical treatment must be pre-approved.
- The laboratory supervisor should also complete an Accident Report (Appendix A), documenting the source of the exposure.

Decontamination and Spill Procedures for Biohazardous Agents

Decontamination is defined as the reduction of biological agents to an acceptable level. Most often the methods used are disinfection (to clean surfaces) or sterilization (for materials and wastes). In the event of a spill of biological material, the individual(s) who caused the spill is responsible for the clean-up. Winthrop University does not have a spill response team. To minimize the consequences of any spill of biological material all work should be performed on plastic-backed liner to absorb spills. Simple spill kits including: (a) chlorine bleach or some other concentrated disinfectant (e.g.CiDecon), (b) a package or roll of paper towels, (c) autoclavable bags, (d) rubber gloves, and (e) forceps for pick-up of broken glass, should be prepared and kept on hand.

All surfaces, tools, equipment and other objects that come in contact with blood or potentially infectious materials must be decontaminated and sterilized as soon as possible with the appropriate disinfectant. **Equipment and tools must be cleaned and decontaminated before servicing or being put back to use**. Decontamination should be accomplished by using either one of these two methods:

1. A solution of 5.25% sodium hypochlorite (household bleach / Clorox) diluted between 1:10 and 1:100 with water. The standard recommendation is to use at least a quarter cup of bleach per one gallon of water.

2. Lysol or some other EPA-registered tuberculocidal disinfectant (e.g. CiDecon, Microstat 2, Ocide Plus, or SOLUCIDE-02). The label of all disinfectants should be checked to make sure they meet this requirement.

If you are cleaning up a spill of blood, carefully cover the spill with paper towels or rags, then gently pour the 10% solution of bleach/CiDecon over the towels or rags, and leave it for *at least 10 minutes*. This will help ensure that any bloodborne pathogens are killed before you actually begin cleaning or wiping the material up. By covering the spill with paper towels or rags, you decrease the chances of causing a splash when you pour the bleach or CiDecon on it.

If you are decontaminating equipment or other objects (be it scalpels, microscope slides, broken glass, tweezers, mechanical equipment upon which someone has been cut, first aid boxes, or whatever) you should leave the disinfectant in place for *at least 10 minutes* before continuing the cleaning process. Any materials you use to clean up a spill of blood or potentially infectious materials must be decontaminated immediately, as well. This would include mops, sponges, re-usable gloves, buckets, pails, etc.

General Biohazardous Spill Guidelines

1. Wear gloves and a lab coat.
2. Use forceps to pick up broken glass and discard into a sharps container or container to be autoclaved (a Pyrex glass beaker works well).
3. Cover spilled material with paper towels.
4. Add diluted disinfectant in sufficient quantity to ensure effective microbial inactivation.
5. Dispose of towels in biohazard waste container.
6. Wipe spill area with diluted disinfectant.
7. Wash hands with soap and water when finished.

Spill Clean-up - BSL-1 Material

For a small spill i**nside** a biological safety cabinet:

1. LEAVE THE CABINET TURNED ON. While wearing gloves, spray the spill or wipe cabinet surfaces (and equipment if in contact) with disinfectant equivalent to CiDecon (for bacteria), or 70% ethanol (for BSL-1 cell cultures). If necessary, flood the work surface, as well as drain pans and catch basins below the work surface, with disinfectant for a contact time of at least 10 minutes
2. Soak up disinfectant and spill with paper towels. If necessary, drain catch basin into a container and/or lift front exhaust grill and tray and wipe all surfaces. Ensure that no paper towels or solid debris are blown into the area beneath the grill.
3. Autoclave any contaminated materials before disposal. Wash hands and any exposed surfaces thoroughly after the clean-up procedure.

For a small spill **outside** a biological safety cabinet - one that can be covered by a few paper towels then follow this procedure:

1. Wearing gloves and a lab coat cover the spill with paper towels and gently apply disinfectant, proceeding from the outer edge of the spill to its center. Leave in place for 10 minutes.
2. Pick up the towels and discard into a biohazard container. Pick up any pieces of broken glass with forceps and place in a sharps container or appropriate autoclavable container (to be autoclaved).
3. Re-wipe the spill area with disinfectant and thoroughly wash hands after glove removal.

Spill Clean-up - BSL-2 Material

The suggestions below are general guidelines for BSL-2 agents. **Please note** that some BSL-2 agents have specific guidelines for their disposal. The MSDS should be checked for all BSL-2 agents prior to clean-up and disposal to determine if special procedures are required. Follow these guidelines if the MSDS does not require special disposal procedures:

For a small spill **inside** a biological safety cabinet:

1. LEAVE THE CABINET TURNED ON. While wearing gloves, spray the spill or wipe cabinet surfaces (and equipment if in contact) with disinfectant equivalent to CiDecon (for bacteria), or 70% ethanol (for BSL-1 cell cultures). If necessary, flood the work surface, as well as drain pans and catch basins below the work surface, with disinfectant for a contact time of at least 20 minutes
2. Soak up disinfectant and spill with paper towels. If necessary, drain catch basin into a container and/or lift front exhaust grill and tray and wipe all surfaces. Ensure that no paper towels or solid debris are blown into the area beneath the grill.
3. Autoclave any contaminated materials before disposal. Wash hands and any exposed surfaces thoroughly after the clean-up procedure.

For a spill **less than 500 mL, outside** a biological safety cabinet*:*

1. Keep others out of the area to prevent spreading spilled material. Post a warning sign, if needed.
2. Remove contaminated clothing and put into a biohazard bag for decontamination later.
3. Wash hands and exposed skin and inform the PI of the spill.
4. Put on protective clothing (lab coat, gloves and if needed, face protection and shoe covers) and assemble clean-up materials (disinfectant, autoclavable container or bag, forceps, sharps container, and paper towels).
5. Pick up broken glass with forceps and dispose into Sharps container/ autoclavable beaker.
6. Cover the spill with paper towels and add appropriately diluted disinfectant (must be an EPA-approved disinfectant).
7. After at least 20 minutes contact time, pick up the paper towels and re-wipe the spill area with diluted disinfectant.
8. Collect all contaminated materials into biohazard waste container and autoclave.
9. Wash hands with soap and water.

For a **spill greater than 500 mL, outside** a biological safety cabinet:

1. Hold your breath and leave the room immediately.
2. Warn others to stay out of the spill area to prevent spread of contamination; post a sign stating: "DO NOT ENTER, BIOHAZARD SPILL", contact (name and phone #) for information". Remove any contaminated clothing and put into a biohazard bag for later autoclaving.
3. Wash hands, exposed skin, and inform the laboratory supervisor and/or SCHO of the spill. If they are not available then contact the Environmental Health and Safety Office (803) 896-0956.
4. Put on protective clothing (lab coat, gloves and, if indicated, surgical mask, eye protection, shoe covers) and assemble clean-up materials.
5. Wait 30 minutes before re-entering the contaminated area to allow dissipation of aerosols.
6. Cover the spill with paper towels and gently apply disinfectant, proceeding from the outer edge of the spill to its center. Leave in place for 20 minutes
7. Collect all treated material and discard in a biohazard container. Pick up any broken glass with forceps and place them into a sharps container.
8. Re-wipe the spill area with disinfectant and wash hands thoroughly at completion of clean up.

*BSL-1 and BSL- 2 Spills **inside** Equipment*

If the spill occurs in a shaker or centrifuge, turn off the equipment and leave the door closed for at least 30 minutes to reduce aerosol exposure before cleaning up the spill. Immediately tape a sign indicating the problem and your name to the contaminated equipment until the situation has been rectified. If you are a student, immediately report the spill to your lab supervisor.

Disposal of BSL-1 and BSL-2 Biohazardous Waste

Biohazard warning labels must be affixed to containers of regulated biohazardous waste, refrigerators and freezers containing blood, or other potentially infectious material; and other containers used to store, transport, or ship blood or other potentially infectious materials. These labels are fluorescent orange, red, or orange-red, and they are available from the SCHO, 132 Dalton Hall. Bags used to dispose of regulated waste must be red or orange red, and they, too, must have the biohazard symbol readily visible upon them. Regulated waste should be double-bagged to guard against the possibility of leakage if the first bag is punctured.

Labels should display this universal biohazard symbol.

Regulated biohazardous waste refers to:

- Any liquid or semi-liquid blood or other potentially infectious materials

- Contaminated items that would release blood or other potentially infectious materials in a liquid or semi-liquid state if compressed
- Items that are caked with dried blood or other potentially infectious materials and are capable of releasing these materials during handling
- Contaminated sharps
- Pathological and microbiological wastes containing blood or other potentially infectious materials

All regulated biohazardous waste must be disposed of in properly labeled containers or biohazard bags and must be identifiable. All regulated biohazardous waste should be labeled with the universal symbol for a biohazard. The Biology Department provides biohazardous waste bags and waste bins for research and academic laboratories. All biohazardous waste must be placed in the proper biohazard container and autoclaved. After autoclaving, the waste should be double bagged in a black trash bag, and immediately taken outside and placed in the dumpster. Biohazard waste bags, biohazardous labels, and biohazardous bins can be obtained from the SCHO, 132 Dalton Hall.

The following types of biohazardous waste may be autoclaved:

- Human blood, blood products, or body fluids (including established or characterized human cell lines)
- Contaminated bedding and animal wastes (manure, feces)
- Contaminated material, equipment, instruments (tubing, gauze, gloves, petri dishes, pipette tips, etc.)
- Department of Transportation (DOT) Category B cultures and stocks (BSL-1, RG1 and most BSL-2, RG2). The DOT defines category B materials as "Category B: An infectious substance that is not in a form generally capable of causing permanent disability or life-threatening or fatal disease in otherwise healthy humans or animals when exposure to it occurs. This includes Category B infectious substances transported for diagnostic or investigational purposes. A Category B infectious substance must be described as ''Biological substance, Category B'' and assigned identification number UN 3373"
- Recombinant DNA materials and genetically modified organisms (except carcasses or when other deactivation methods are stipulated)
- Sharps waste

Only the autoclave in the microbiology lab may be used for autoclaving regulated biohazardous materials. The procedure for autoclaving regulated biohazardous waste (BSL-1 and BSL-2) is detailed below:

1. Complete the autoclave log hanging on the wall near the autoclave in the microbiology prep room. All regulated biohazardous waste must be logged.
2. Secure biohazard bag with autoclave tape or a twist tie. Make sure that the bag does not have any holes or rips. If the bag has holes or rips then place the bag in a new biohazard bag. Only autoclave one bag of waste at a time. Place a piece of autoclave tape on the bag.

3. Turn on the steam generator to the autoclave. The button is located below the autoclave door on the right side of the machine. Wait 20 minutes for the pressure gauge (located on the left below the autoclave door) to reach 20 PSI.
4. Place two trays or a large high sided pan on the bottom of the autoclave in case the bag leaks.
5. Place the bag on the tray or in the pan. Only autoclave one bag at a time. Autoclaving more than one bag may result in failure of the autoclave to properly sterilize the reagents since the steam flow can be altered from "overstuffing" the autoclave. Thus, **do not autoclave but one bag of biohazardous waste at a time.**
6. Close the autoclave door by turning the handle. Press the appropriate cycle on the autoclave.
7. Turn the autoclave on by pressing the red button. All regulated biohazardous waste **must be autoclaved for a minimum of 30 minutes at 120 PSI**.
8. When the cycle is complete a buzzer will sound. Carefully open the door to avoid being burned by the steam. Wear the autoclave gloves provided. Look at the bag to make sure that the autoclave indicator tape stripes have turned from white to black. Check for any leaks. Push off the red button.
9. Remove the biohazard bag and allow it to cool then double-bag it in a black trash bag. Secure the bag with a twist tie or knot. Immediately take the waste to the dumpster located between Dalton Hall and Sims Science Building. **Custodians and housekeepers will not remove bags containing any form of blood (human or animal), vials containing blood, bloody towels, rags, biohazardous waste, etc. from laboratories**. They have been given very strict instructions not to handle any regulated waste.
10. Turn the pressure switch off (if at the end of the week)

Special Categories of Biohazardous Wastes

The principal investigator/faculty are responsible for determining prior to beginning any new work, the types of waste that will be generated and how that waste will be safely handled from collection to appropriate disposal. Should the principal investigator/faculty determine that their work will generate any of the following special categories of biohazardous waste, a written plan providing procedural details for the safe handling, collection and storage of the waste, and arrangements for legally compliant disposal must be submitted to and approved by Environmental Health and Safety prior to initiation of the work.

- Animal Carcasses (diagnosed with or subject to infectious agents, subject to rDNA material, that can't be rendered, or that can't be given to raptor rehab or taken to the county animal shelter for incineration).
- Contaminated (untreated or non-deactivated) BSL-2 HVAC or HEPA filters
- Department of Transportation (DOT) Class 6, Division 6.2, Category A materials, which DOT defines as, "an infectious substance in a form capable of causing permanent disability or life-threatening or fatal disease in otherwise healthy humans or animals when exposure to it occurs." A Category A infectious substance must be assigned (based on the known medical history or symptoms of the source patient or animal, endemic local conditions, or professional judgment concerning the individual circumstances of the source human or

animal) one of two identification numbers: UN 2814—infectious substance affecting humans, or humans and animals, or UN 2900—infectious substance which causes disease only in animals. Category A materials may include cultures and stocks of some BSL-2, RG2 materials and all BSL-3, RG3 and BSL-4, RG4 materials. **BSL-3 and BSL-4 materials are not permitted for use in the Biology Department, Winthrop University.**

- Pathological waste: animal body parts, organs, tissues, and surgical specimens (exclusive of formaldehyde or other preservatives).

Section I: Equipment Usage and Safety

This section of the Health, Safety, and Security Plan is intended to assist faculty and students with the proper care and use of equipment commonly found and used in the Department of Biology. Many equipment items located throughout the department can cause serious injury if used improperly. Items considered common for use such as a microwave oven can cause fatal injuries if used improperly in the lab. The information contained in this section will assist you in properly using basic lab equipment. For more assistance with lab equipment contact the Lab Manager, 132 Dalton Hall. (*Note: Parts of this section were adapted with permission from Oklahoma State University.)*

Autoclaves

Steam sterilization of materials is a dependable procedure for the destruction of all forms of microbial life (including endospores). Autoclaves (steam sterilizers) are common laboratory tools that must be properly used to be effective for the decontamination of cultures and other potentially biohazardous materials. Steam, heat, and pressure are the principal physical hazards associated with using autoclaves. Improper use of autoclaves can result in significant personal injury from exposure to steam, scalding liquids, and shattering glassware. Faculty and/or laboratory supervisors with autoclave experience are responsible for showing new lab personnel and students how to use an autoclave properly. All autoclave users should review the manufacturer's operating manual for specific instructions before autoclaving liquids – an operation that requires careful attention.

Autoclaves use pressurized steam to destroy microorganisms and are the most dependable system available for the decontamination of laboratory waste and the sterilization of laboratory glassware, media, and reagents. For efficient heat transfer, steam must flush the air out of the autoclave chamber. Before using the autoclave, check the drain screen at the bottom of the chamber and clean if blocked. If the sieve is blocked with debris, a layer of air may form at the bottom of the autoclave, preventing efficient operation.

Container Selection

Polypropylene bags, commonly called biohazard or autoclave bags, are able to withstand autoclaving and are tear resistant, but can be punctured or burst in the autoclave. Therefore, place bags in a rigid container such as a polypropylene or stainless steel pan during autoclaving. Bags are available in a variety of sizes and some are printed with an indicator that changes color when processed. Polypropylene bags are impermeable to steam and for this reason should not be twisted and taped shut, but gathered loosely at the top and secured with a large rubber band, or metal tie, or autoclave tape. This will create an opening through which steam can penetrate. Polypropylene is a plastic capable of withstanding autoclaving, but resistant to heat transfer. Therefore, materials contained in a polypropylene pan will take longer to autoclave than the same materials in a stainless steel pan. To decrease the time required to sterilize material in these containers, remove the lid (if applicable), turn the container on its side when possible, and select a container with the lowest sides and widest diameter possible for the autoclave.

Preparation and Loading of Materials

Follow these general guidelines when preparing and loading materials to be autoclaved:

- Fill liquid containers only half-full.
- Loosen caps, or use vented closures.
- Always put bags of biological waste into pans/onto lipped trays to catch spills.
- Position biohazard bags on their sides, with the bag neck taped loosely.
- Leave space between items to allow steam circulation.
- Household dishpans melt in the autoclave. Use autoclavable polypropylene or stainless steel pans.

Cycle Selection

- Use liquid cycle (slow exhaust) when autoclaving liquids, to prevent contents from boiling over.
- Select dry exhaust cycle for glassware.
- Use dry cycle for wrapped items such as cotton swabs

Time Selection

- Take into account the size of the articles to be autoclaved. A 2-liter flask containing 1 liter of liquid takes longer to sterilize than four 500 ml flasks each containing 250 ml of liquid.
- Material with a high insulating capacity (animal bedding, high-sided polyethylene containers) increases the time needed for the load to reach sterilizing temperatures.
- Bags of biological waste should be autoclaved for 30 minutes to ensure decontamination.

Removing the Load

- Check that the chamber pressure is zero.
- Wear lab coat, eye protection, heat insulating gloves, and closed-toe shoes.
- Stand behind door when opening it.
- Slowly open door only a crack. Beware of rush of steam.
- After the slow exhaust cycle, open autoclave door and allow liquids to cool for 20 minutes before removing.

Monitoring

Autoclaves used to decontaminate laboratory waste should be tested periodically to insure effectiveness. Two types of tests are used: 1) a chemical indicator that fuses when the temperature reaches 121°C, and 2) heat-resistant spores (*Bacillus stearothermophilis*) that are killed by exposure to 121°C for approximately 15 minutes. Both types of tests should be placed well down in the center of the bag or container of waste, at the point slowest to heat.
The chemical test should be used first to determine that the temperature in the center of the container reaches 121°C. Ampules of heat-resistant spores should be used in subsequent test runs to determine the amount of time necessary to achieve sterilization.
Best Practices for Autoclaves

The following are recommended guidelines when using an autoclave:

- Do not put sharp or pointed contaminated objects into an autoclave bag. Place them in an appropriate rigid sharps disposal container.
- Use caution when handling an infectious waste autoclave bag, in case sharp objects were inadvertently placed in the bag. Never lift a bag from the bottom to load it into the chamber, handle the bag from the top.
- Do not overfill an autoclave bag. Steam and heat cannot penetrate as easily to the interior of a densely packed autoclave bag. Frequently the outer contents of the bag will be treated but the innermost part will be unaffected.
- Do not overload an autoclave. An overloaded autoclave chamber does not allow efficient steam distribution. Considerably longer sterilization times may be required to achieve decontamination if an autoclave is tightly packed.
- Do not mix contaminated and clean items together during the same autoclave cycle. Clean items generally require shorter decontamination times (15-20 minutes) while a bag of infectious waste (24" x 36") typically requires 30-40 minutes to an hour to be effectively decontaminated.
- Always wear personal protective equipment, including heat-resistant gloves, safety glasses and a lab coat when operating an autoclave. Use caution when opening the autoclave door and allow superheated steam to exit before attempting to remove autoclave contents.
- Be alert when handling pressurized containers. Superheated liquids may spurt from closed containers. Never seal a liquid container with a cork or stopper. This could cause an explosion inside the autoclave.
- Agar plates will melt and the agar will become liquefied when autoclaved. Avoid contact with molten agar. Use a secondary tray to catch any potential leakage from an autoclave bag rather than allowing it to leak onto the floor of the autoclave chamber.
- If there is a spill inside the autoclave chamber, allow the unit to cool before attempting to clean up the spill. If glass breaks in the autoclave, use tongs, forceps or other mechanical means to recover fragments. Do not use bare hands.
- If the autoclave does not seem to be running correctly, allow the cycle to finish. DO NOT SWITCH OFF the red button since the vacuum will form and we will have to call someone to open the door. Allow the autoclave to cool then re-run your cycle correctly.

Biological Safety Cabinets (BSCs)

BSCs are classified as Class I, Class II or Class III cabinets. When properly maintained and operated, they effectively contain and capture microbial contaminants and infectious agents using HEPA (High Efficiency Particulate Air) filters. BSCs should not be confused with clean benches which only protect the material being used and are not suitable for work with infectious or toxic material. (Although clean benches have HEPA-filtered air, the air flows over the experimental material toward the user rather than being drawn away from the user). BSCs should also not be confused with conventional chemical fume hoods which do not filter microorganisms.

General guidelines for working in a biological safety cabinet include:

- Turn the cabinet on at least 10 - 15 minutes prior to use.

- Disinfect work surface with 70% alcohol or other suitable disinfectant.
- Consider the materials necessary for the planned work in the cabinet.
- Place items into the cabinet so that they can be used efficiently without unnecessary disruption of the air flow. Work with the materials from the clean to the dirty side.
- Wear appropriate personal protective equipment; at a minimum, a buttoned lab coat and gloves.
- Adjust the working height of the stool so that the worker's face is above the front opening.
- Delay manipulation of materials for approximately one minute after placing the hands/arms inside the cabinet.
- Minimize the frequency of moving hands in and out of the cabinet.
- Do not disturb the airflow by covering any of the grillwork with materials.
- Work at a moderate pace to prevent the air flow disruption that occurs with rapid movements.
- When work is completed, wipe the bottom and side surfaces of the hood with disinfectant.
- NOTE: Be very careful when using small pieces of materials such as KimWipes in the hood as these can be drawn into the hood and disrupt motor operation.

Certification of Biological Safety Cabinets

Certification of a BSC includes a series of performance tests to confirm that the cabinet will provide the user and experimental material the protection for which it is designed. The airflows, filters, and cabinet integrity are checked to ensure that the cabinet meets minimum performance standards. Certification is arranged through the department Laboratory Manager and provided by an outside vendor.

BSCs intended for user protection must be certified:

- When newly installed, when relocated, and at least annually.
- Biological safety cabinets intended only for protection of the experimental material are certified at the discretion of the Principal Investigator.
- BSC decontamination (using the paraformaldhyde gas production process) is also provided by an outside vendor and needs to be done:
    - Before any maintenance work requiring disassembly of the air plenum, including filter replacement.
    - Prior to cabinet recertification.
    - Before moving the cabinet to a new laboratory.

<u>Bunsen Burners</u>

Burns are the most common laboratory injury. After heating an object, be extremely careful and let it cool before grasping it. Temperatures in the hottest region of the burner flame approach ~1500°C. Before using a Bunsen burner, be certain that no flammable materials are present in the laboratory. Also, be careful and make sure that your face, clothing and hair are not above or near the opening of the burner tube. After lighting the burner, adjust the burner until there is a blue flame containing two or more cones. When adjusting the air vent, be careful not to extinguish the

flame or disassemble the burner. After you are finished using the burner, turn the gas completely off to extinguish the flame. General safety rules for Bunsen burners include:

- Never use your fingers to hold an object in the Bunsen burner flame. Always use a pair of tongs to hold a small, solid object.
- If the object is large or a liquid in a flask, use a ring stand and a triangle or wire gauze to hold the object in the flame.
- Never leave a Bunsen burner unattended.
- Always check to see that you have turned the gas completely off to the burner before leaving it.

Centrifuges

Ninety percent of centrifuge-related failures are user errors. Careless centrifugation can mean lost samples and damaged equipment, plus a risk to the user and the lab. Fortunately personal injury is an infrequent event. However, when it occurs, it is usually associated with improper lifting techniques used to move heavy rotors. Centrifugation isn't as simple as it appears, requiring careful use, careful maintenance, and careful bookkeeping. The manufacturer's instructions for safe operating speeds must be strictly followed; it is important that the load is balanced each time the centrifuge is used and that the lid is closed while the rotor is in motion. When the top is opened, the disconnect switch must be working properly to shut off the equipment.

The following are recommended guidelines for centrifuge use:

- Lids must be closed at all times during operation.
- The operator must not leave the centrifuge until full operating speed is attained and machine appears to be running safely without vibration.
- If vibration occurs the centrifuge should be stopped immediately and load balances checked. Swing-out buckets should be checked for clearance and support.
- Rooms where potentially hazardous biological, radioactive materials, toxic or other hazardous chemicals are centrifuged must be identified by the appropriate warning signs.
- Plastic centrifuge tubes should be discarded after one cycle of ultracentrifugation, unless specifically designated for multiple uses. The failure rate for used tubes is a hazard which justifies the use of new tubes for each high G centrifugation.
- Nitrocellulose tubes should be used only when transparent and flexible (fresh). They must never be heated because of explosive possibility.
- Rotors and cups should be cleaned and disinfected after each use with non-corrosive cleaning solutions (mild detergent and distilled water are recommended). Test tube brushes must not be used for cleaning the cup cavities. All traces of detergents should be removed prior to air drying.

Cryostats

Cryostats are refrigerated chambers, kept at sub-zero temperatures, which contain a microtome. They are used for cutting thin sections (5-20 microns in thickness) of fresh, frozen material or fixed samples. The material that has not been subject to fixation should be considered a potential

hazard. Before cutting a frozen section from any fresh tissue, it is essential to obtain as much information as possible about the source of the material. Otherwise the specimen should be treated with all the caution afforded the most infective material.

There is also significant risk when the cryostat is cleaned. While the tissue fragments remain at sub-zero temperatures within the cryostat chamber, there is little danger of infection, but once the tissue fragments reach room temperature, organisms can become viable. Consequently loose tissue fragments should be collected and placed into fixative or disinfecting agent before defrosting commences. After defrosting all tissue contaminated surfaces must be disinfected with 70% ethanol and cleaned thoroughly before the unit is used again.

Compressed Gas Cylinders

Compressed gasses present unique hazards. Depending on the particular gas, there is a potential for simultaneous exposure to both mechanical and chemical hazards. Gasses may be flammable or combustible, explosive, corrosive, poisonous, inert, or a combination of hazards. Because of the potential dangers of gas cylinders, the contents of any compressed gas cylinder must be clearly identified. Such identification should be written on a yellow label and attached to the cylinder. No compressed gas cylinder that does not legibly identify its contents by name should be accepted for use in the laboratory. If the labeling on a cylinder becomes unclear or an attached tag is defaced to the point the contents cannot be identified, the cylinder should be marked "contents unknown" and the Laboratory Manager (132 Dalton Hall) should be immediately notified.

The cylinders themselves are primarily shipping containers and should not be subjected to rough handling or abuse. Such misuse can seriously weaken the cylinder and render it unfit for further use or transform it into a rocket having sufficient thrust to drive it through masonry walls. To protect the valve during transportation, the cover cap should be screwed on hand tight and remain on until the cylinder is in place and ready for use. When moving large cylinders, they should be strapped to a properly designed wheeled cart to ensure stability; cylinders should never be rolled or dragged. Only one cylinder should be handled (moved) at a time.

All gas lines leading from a compressed gas cylinder should be clearly labeled to identify the gas, the laboratory or area served, and the relevant emergency telephone numbers. The labels should be color coded to distinguish hazardous gases, such as flammable, toxic, or corrosive substances, using a yellow background and black letters. Signs should be conspicuously posted in areas where flammable compressed gases are stored, identifying the substances and appropriate precautions (e.g., HYDROGEN - FLAMMABLE GAS - NO SMOKING - NO OPEN FLAMES).

General guidelines for gas cylinders include:

- Gas cylinders must be secured at all times to prevent tipping.
- Cylinders may be attached to a bench top, individually to the wall, placed in a holding cage, or have a non-tip base attached. Chains or sturdy straps may be used to secure them.
- If a leaking cylinder is discovered, immediately call Campus Police, 803-323-3333.
- Under no circumstances should any attempt be made to repair a cylinder or valve.

- Regulators are gas specific and not necessarily interchangeable! Always make sure that the regulator and valve fittings are compatible.
- A cylinder should never be emptied to a pressure lower than 172 kPa (25 psi/in2) (the residual contents may become contaminated if the valve is left open). When work involving a compressed gas is completed, the cylinder must be turned off, and if possible, the lines bled.
- When the cylinder needs to be removed or is empty, all valves shall be closed, the system bled, and the regulator removed. The valve cap shall be replaced, the cylinder clearly marked as "empty" by removing the "full" portion of the label, and returned to the chemical storage area for pickup by the supplier.
- Always use safety glasses (preferably with a face shield) when handling and using compressed gases, especially when connecting and disconnecting compressed gas regulators and lines.

Gas Cylinder Transportation

Because of the danger of escaping gases from faulty valves and because elevators are considered a confined space, all compressed gas cylinders must be *unaccompanied* in the elevator.

This prohibition also applies to cryogens in containers (e.g. liquid nitrogen) that, because of the gas generated during a spill or by the continuous container venting if the elevator were to fail for a prolonged period of time, could displace enough air in the elevator to create a suffocation hazard.

In the case of cryogen containers in volumes that can be easily carried, it may be safer and more expedient for the transporter to use the stairs. If a cryogen container, e.g. large Dewar, is too large to carry, it must be transported via the elevator unaccompanied, correctly secured, and with the appropriate warning sign, as for the procedure for compressed gas cylinders.

Procedure:

Inspect the cylinder or Dewar to be moved for safe transportation.

1. Inspect gas cylinder dolly/cart, checking tires/wheels and security chain/strap.
2. Secure compressed gas tank or Dewar onto the appropriate truck/dolly using a restraining chain/strap with sign attached noting not to ride elevator with canister.
3. Load the cylinder/container into an empty elevator.
4. Do not ride in an elevator with a compressed gas cylinder/Dewar.
5. Press the number for the destination floor and exit the elevator.
6. Take the stairs to meet the cylinder or by previous arrangement, have a colleague meet the cylinder on the destination floor.
7. Remove the cylinder/container from the elevator at the destination floor.

The following rules should always be followed in regards to piping compressed gasses:

- Copper piping shall not be used for acetylene.

- Plastic piping shall not be used for any portion of a high-pressure system.
- Do not use cast iron pipe for chlorine.
- Do not conceal distribution lines where a high concentration of a leaking hazardous gas can build up and cause an accident.
- Distribution lines and their outlets should be clearly labeled as to the type of gas contained.
- Piping systems should be inspected for leaks on a regular basis.
- Special attention should be given to fittings as well as possible cracks that may have developed.

Gas Regulators

When installing a gas regulator on a cylinder, make sure the cylinder is properly secured, that you have the correct regulator and that you are aware of any special hazards of the gas you are working with. Remove the cylinder valve cap (counterclockwise) and place it somewhere nearby. Some regulators (on lecture bottles and certain corrosive gases) require a Teflon or lead washer to be inserted between the tank outlet and regulator. Check to see if this is required before continuing.

Follow these general guidelines for adjusting a gas regulator:

- Make sure that the regulator outlet valve (**A**) is shut. Screw it **clockwise** until it seats, but so not over tighten it or you can damage the valve seat.
- Make sure that the regulator control valve (**B**) is shut. Screw it **counterclockwise** until it is almost completely unscrewed. If you unscrew it completely, just put it back in.
- Screw the regulator onto the tank by hand until it is almost finger tight. Some people like to use Teflon tape on this connection, but that's generally not a good idea. Bits of Teflon tape can get blown into the regulator, causing a leak, valve malfunction or erroneous reading.
- What you do next depends on the kind of gas you are working with and whether you need to exclude air from the gas line you're connecting to.

- For nitrogen and argon cylinders:

- If you don't need to purge out the "dead volume" of air in the regulator body, simply tighten the regulator firmly with a wrench (use the correct tool for the job, not pliers!).
- If you need to exclude air from your line then you can purge out the "dead volume" using the following technique. With the regulator on finger tight, open the tank valve until gas just begins to flow. Once the dead volume is purged (2-4 seconds) and with the tank still open a minimum amount, tighten the regulator firmly with a wrench. **Note**: Do not attempt this procedure without the direct supervision of someone who is experienced in the technique. Do not use this technique on an unsecured cylinder, for any other gases or in an unventilated area!

- For corrosive or reactive gases:
  - Make sure you are using the proper regulator (typically these will be made of stainless steel or Monel).
  - Tighten the regulator firmly with a wrench (use the correct tool for the job, not pliers!).
  - Purge the regulator with an inert dry gas or evacuate the system (depending on the exact set-up). Specific instructions for this will vary depending on what you need to do so consult an appropriate resource beforehand.
  - Be sure to install some kind of suck-back trap after the regulator or at least use clear Tygon tubing so that the backflow of corrosive reaction mixtures into the regulator and cylinder can be avoided.

- For flammable gases:
  - If you are using acetylene be **certain** that you are using an acetylene regulator with proper fittings. Alloys containing copper or silver can cause explosions when used with acetylene!
  - Ideally, ground the system to avoid static discharges.
  - Tighten the regulator firmly with a wrench (use the correct tool for the job, not pliers!).
  - If necessary, follow the purge instructions for corrosive or reactive gases as shown above.
  - Open the tank valve slowly (counterclockwise). Watch the tank pressure on the regulator (**C**).
  - Slowly turn the regulator control valve (**B**) until the regulator pressure (**D**) is at the desired level.
  - Open the regulator outlet valve (**A**). You can regulate flow with this valve, but the ultimate pressure depends on the setting of the regulator control valve!
  - Check your system for leaks using Snoop (a commercial product) or some soapy water. Snoop is preferred since it leaves no residue. If you find leaks, try tightening the connections.

Reminder: Make sure the tank valve is closed whenever you are not dispensing gas through the regulator.

Disconnecting a Regulator

- Shut the tank valve on the gas cylinder.
- Slowly open the outlet valve (**A**) on the regulator.
- Watch the pressure gauges **C** and **D** drop to zero.
- Open the regulator control valve (**B**) (turn it clockwise) to ensure that all pressure has been released.
- If you were using a corrosive gas, purge the system with a dry inert gas.
- Using a wrench (not pliers!) disconnect the regulator from the gas cylinder. Replace the protective cylinder cap immediately.
- If your regulator was used with a corrosive gas, purge it again with dry air or nitrogen in the hood for several minutes.
- If your cylinder is empty, it must be properly labeled and then returned to the chemical storage shed. Do not store empty gas cylinders in the laboratory.

Empty Compressed Gas Cylinders

When the cylinder is empty, all valves shall be closed, the system bled, and the regulator removed. The valve cap should be replaced, the cylinder clearly marked as "empty," and returned to the chemical storage area for pick-up. Refer to the moving techniques described above to safely move the cylinder.

Electrophoresis Equipment

Precautions to prevent electric shock must be followed when conducting procedures involving electrophoresis. Lethal electric shock can result when operating at high voltages such as in DNA sequencing or low voltages such as in agarose gel electrophoresis (e.g., 100 volts at 25 milliamps). These general guidelines should be followed:

- Turn the power off before connecting the electrical leads.
- Connect one lead at a time, using one hand only.
- Ensure that hands are dry while connecting leads.
- Keep the apparatus away from sinks or other water sources.
- Turn off power before opening lid or reaching inside chamber.
- Do not override safety devices.
- Do not run electrophoresis equipment unattended.
- If using acrylamide, purchase premixed solutions or pre-weighed quantities whenever possible.
    - Mix all stock solutions in a chemical fume hood.
    - Provide spill containment by mixing gels on a plastic tray.
    - Decontaminate surfaces with ethanol and dispose of all cleanup materials as hazardous waste.

Flammable Storage Cabinets

Flammable storage cabinets generally need not be vented, however, the need for venting should be evaluated and the decision made after assessing the nature of the likely contents. Typically, cabinets in laboratories using large volumes of chemicals are vented due to the highly odiferous

and toxic materials stored in them. Should a decision be made to vent a flammables storage cabinet, ventilation must be per manufacturer's recommendations.

When cabinets are vented, flame arrestor screens must be placed in the openings. Flame arrestor screens are special fine metallic screens that will prevent propagation or flashback of flames. Mechanical exhaust ventilation must be provided from the bottom of the cabinet and directly into a building exhaust duct, not into a chemical fume hood or another cabinet. Ventilation ducts must be no smaller than the cabinet vent and should be of rigid steel. Other duct material may be used, if it can be shown to have equivalent fire resistance. Manifolding of ventilation is discouraged, but up to four cabinet vents may be manifolded.

When cabinets are not vented, both vent openings must be closed with the bungs provided with the cabinets or with bungs specified by the manufacturer. The integrity of the cabinet is reliable only when the manufacturer's parts are used.

Freezers

All laboratory freezers operating at -30C or less pose a frostbite hazard when removing or replacing the contents. Regular chemical-safety gloves do not protect from frostbite. This hazard is more acute for -80C "ultralow" freezers. At least two pairs of insulated freezer-gloves must be provided for any room containing -30C or -80C freezers. These gloves should be checked regularly to see that the liners are intact and that both gloves of each pair are present.

Glassware

Although glass vessels are frequently used in low-vacuum operations, evacuated glass vessels may collapse violently, either spontaneously from strain or from an accidental blow. Therefore, pressure and vacuum operations in glass vessels should be conducted behind adequate shielding. It is advisable to check for flaws such as star cracks, scratches and etching marks each time a vacuum apparatus is used. Only round-bottomed or thick-walled (e.g., Pyrex) evacuated reaction vessels specifically designed for operations at reduced pressure should be used. Repaired glassware is subject to thermal shock and should be avoided. Thin-walled, Erlenmeyer or round-bottomed flasks larger than 1 L should never be evacuated.

Mechanical hotplates

Laboratory hot plates are normally used for heating solutions to 100^{o}C or above when inherently safer steam baths cannot be used. Any newly purchased hot plates should be designed in a way that avoids electrical sparks. However, many older hot plates pose an electrical spark hazard arising from either the on-off switch located on the hot plate, the bimetallic thermostat used to regulate the temperature or both. Laboratory workers should be warned of the spark hazard associated with older hot plates. In addition to the spark hazard, old and corroded bimetallic thermostats in these devices can eventually fuse shut and deliver full, continuous current to a hot plate. General rules of safety for hotplates include:

- Do not store volatile flammable materials near a hot plate.
- Do not use older hot plates for flammable materials.
- Check for corrosion of thermostats. Corroded bimetallic thermostats can be repaired or reconfigured to avoid spark hazards.
- Never leave a hotplate unattended.

Mechanical Stirrers and Mixing Devices

The stirring and mixing devices commonly found in laboratories include stirring motors, magnetic stirrers, shakers, small pumps for fluids and rotary evaporators for solvent removal. These devices are typically used in laboratory operations that are performed in a hood, and it is important that they be operated in a way that precludes the generation of electrical sparks.

Only spark-free induction motors should be used in power stirring and mixing devices or any other rotating equipment used for laboratory operations. While the motors in most of the currently marketed stirring and mixing devices meet this criterion, their on-off switches and rheostat-type speed controls can produce an electrical spark because they have exposed electrical conductors. The speed of an induction motor operating under a load should not be controlled by a variable autotransformer.

Because stirring and mixing devices, especially stirring motors and magnetic stirrers, are often operated for fairly long periods without constant attention, the consequences of stirrer failure, electrical overload or blockage of the motion of the stirring impeller should be considered.

Microwave Ovens

Microwave ovens can be dangerous pieces of equipment when not used properly. Microwave ovens used in the laboratory may pose several different types of hazards:

- As with most electrical apparatus, there is the risk of generating sparks that can ignite flammable vapors.
- Metals placed inside the microwave oven may produce an arc that can ignite flammable materials.
- Materials placed inside the oven may overheat and ignite.
- Sealed containers, even if loosely sealed, can build pressure upon expansion during heating, creating a risk of container rupture.

To minimize the risk of these hazards:

- Do not operate the oven if it is damaged. It is very important that the oven door closes properly and that there is no damage to the door seals and sealing surfaces, hinges and latches.
- Since there may be residual contamination, never use the laboratory microwave oven to heat food or drinks.
- Do not use the microwave oven to heat up hazardous chemicals or radioactive materials.

- Do not use ALUMINUM FOIL at any time during the heating cycle. Metal utensils and utensils with metallic trim should not be used in the microwave oven.
- Avoid heating materials in cylindrical-shaped containers. Liquids heated in certain shaped containers (especially cylindrical-shaped containers) may become overheated. When overheated, liquids may splatter during or after the heating cycle resulting in possible injury or damage to the microwave oven.
- When heating liquids in screw-cap bottles, completely loosen the screw caps to prevent pressure build-up within the container. This pressure build-up with a cap that is not sufficiently loose can cause the bottle to explode.
- If steam accumulates inside or outside of the oven door, wipe with a soft cloth. This may occur when the microwave oven is operated under high humidity conditions and in no way indicates malfunction of the unit.
- Be careful when removing containers from the microwave. Some containers absorb heat and may be very hot. Always use protective gloves and appropriate eye/face protection to minimize any possible injuries.
- If materials inside the oven should ignite, KEEP OVEN DOOR CLOSED, turn oven off, and disconnect the power cord.
- Do not attempt to tamper with or make any adjustments or repairs to the door, control panel, safety interlock switches or any other part of the oven. Repairs should be done by qualified service personnel only.
- Every microwave oven used in a biology lab should be provided with one pair of thermal gloves.

Ovens

Electrically heated ovens are commonly used in the laboratory to remove water or other solvents from chemical samples and to dry laboratory glassware. ***Never use laboratory ovens for human food preparation.*** Laboratory ovens should be constructed such that their heating elements and their temperature controls are physically separated from their interior atmospheres. Laboratory ovens rarely have a provision for preventing the discharge of the substances volatilized in them. Connecting the oven vent directly to an exhaust system can reduce the possibility of substances escaping into the lab or an explosive concentration developing within the oven. Ovens should not be used to dry any chemical sample that might pose a hazard because of acute or chronic toxicity unless special precautions have been taken to ensure continuous venting of the atmosphere inside the oven.

To avoid explosion, glassware that has been rinsed with an organic solvent should be rinsed again with distilled water before being dried in an oven. Bimetallic strip thermometers are preferred for monitoring oven temperatures. Alcohol thermometers may be mounted through holes in the top of ovens so that the bulb hangs into the oven. Note that alcohol thermometers have an upper temperature limit; do not heat them past this point. No mercury thermometers should be used in laboratories.

Sharps

"Sharps waste" means any device having acute rigid corners, edges, or protuberances capable of cutting or piercing, including, but not limited to, hypodermic needles, syringes, razor blades, coverslips, and scalpel blades (1). Glass items contaminated with biohazards, such as microscope slides with live specimens, and capillary tubes are also considered "sharps waste." Sharps disposal containers must be readily accessible and located as close as possible to the area where sharps will be used. The container should be rigid, leak-proof in the upright position, and puncture resistant (2). When the container is three quarters full it should be sealed and taken to the Laboratory Manager for disposal in the dumpster. Sharps containers that hold biohazards should be clearly labeled and autoclaved before disposal.

1. https://www.ehs.ucsb.edu/sites/default/files/docs/hw/labsharpsdisposal.pdf
2. https://deq.nc.gov/about/divisions/waste-management/solid-waste-section/medical-waste/medical-waste-guidance-and-interpretation#Sharps-3012

Refrigerators and Freezers

Only refrigerators and freezers specified for laboratory use should be utilized for the storage of chemicals. These refrigerators have been constructed with special design factors, such as heavy-duty cords and corrosion resistant interiors to help reduce the risk of fire or explosions in the lab. Standard refrigerators have electrical fans and motors that make them potential ignition sources for flammable vapors. **Do not store flammable liquids in a refrigerator unless it is approved for such storage.** Flammable liquid-approved refrigerators are designed with spark-producing parts on the outside to avoid accidental ignition. If refrigeration is needed inside a flammable-storage room, you should use an explosion-proof refrigerator.

Frost-free refrigerators should also be avoided. Many of them have a drain or tube or hole that carries water and possibly any spilled materials to an area near the compressor, which may spark. Electric heaters used to defrost the freezing coils can also spark.

To ensure the safe use of refrigerators and freezers, faculty and students are expected to follow these guidelines:

- All materials in refrigerators or freezers should be labeled with the contents, owner, date of acquisition or preparation and nature of any potential hazard. Labels and ink used to identify materials in the refrigerators or freezers should be water-resistant.
- **Only chemicals should be stored in chemical storage refrigerators; lab refrigerators should not be used for food storage or preparation.** Refrigerators should be labeled for their intended purpose. Labels reading "No Food or Drink to be Stored in this Refrigerator" or "Refrigerator For Food Only" are available from the Lab Manager, 132 Dalton Hall.
- All containers should be sealed, preferably with a cap. Containers should be placed in secondary containers, or catch pans should be used.

UV Transilluminator and Lights

UV or ultraviolet lamps are used in biological safety cabinets, light boxes, and crosslinkers in many laboratories. One of the problems in working with UV radiation is that the symptoms of overexposure are not immediately felt so that persons exposed do not realize the hazard until after the damage is done. An unfortunate property of UV radiation is that there are no immediate warning symptoms to indicate overexposure. Symptoms of overexposure including varying degrees of erythema (sunburn) or photokeratitis (welder's flash) typically appear hours after exposure has occurred. The symptoms of over exposure include:

- Skin Injury - UV radiation can initiate a photochemical reaction called erythema within exposed skin. This "sunburn" can be quite severe and can occur as a result of only a few seconds exposure. Effects are exaggerated for skin photosensitized by agents such as coal tar products, certain foods (e.g., celery root), certain medications and photoallergens. Chronic skin exposure to UV radiation has been linked to premature skin aging, wrinkles and skin cancer.
- Eye Injury – UV radiation exposure can injure the cornea, the outer protective coating of the eye. Photokeratitis is a painful inflammation of the eye caused by UV radiation-induced lesions on the cornea. Symptoms include a sensation of sand in the eye that may last up to two days. Chronic exposures to acute high-energy UV radiation can lead to the formation of cataracts.

General guidelines for working with UV light illuminators include:

- A hazard warning sign must be affixed to the doors of laboratories, animal rooms, etc. which have ultraviolet light installations.
- Adequate eye and skin protection must be worn when working in an irradiated area. Safety glasses with side shields or goggles with solid side pieces must be worn. NORMAL EYEGLASSES OR CONTACTS OFFER YOU VERY LIMITED PROTECTION!!
- Skin protection is afforded by face shields, caps, gloves, gowns, etc.
- UV lamp surfaces should be cleaned as often as necessary to maximize output.
- UV lamps used as space and surface sanitizers should be checked regularly and replaced according to the manufacturer's recommendations.
- Used UV lamps are classified by EPA as regulated universal waste and should be taken to the Lab Manager, 132 Dalton Hall, for recycling.

Vacuums

Vacuum pumps are used in the lab to remove air and other vapors from a vessel or manifold. The most common usages are on rotary evaporators, drying manifolds, centrifugal concentrators ("speedvacs"), acrylamide gel dryers, freeze dryers, vacuum ovens, tissue culture filter flasks and aspirators, desiccators, filtration apparatus and filter/degassing apparatus.

Vacuum pumps must be fitted with an oil-mist filter which should be monitored for saturation and replaced when necessary. Oil seal pumps are susceptible to excessive amounts of gas, corrosive acids and bases and excessive water vapors.

Water Baths

Water baths can create a fire hazard if left on without water. For this reason water baths should not be left on overnight. Water baths must be checked weekly to ensure proper water levels. Never operate a water bath without water in it. Never leave objects such as racks or beakers in a water bath. Plastic items left in a water bath can melt if the water evaporates. Glass items left in a water bath without water can shatter.

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APPENDICES

Appendix A

Accident Report Form

Winthrop University Employee's Report of Injury Form

Appendix B

Laboratory Inspection Checklist

Laboratory Inspection Checklist

Appendix C

University Emergency Guidelines

https://www.winthrop.edu/uploadedFiles/emergency/EmergencyGuidelines.pdf

Appendix D

Signs and Label Information

Signs and Label Information

Warning labels, whenever possible, should be attached to all containers of chemicals, biological waste, biohazardous waste, refrigerators/freezers where biohazardous waste is stored, and to equipment that is used for biohazardous waste.

The biohazardous symbol (see below) must be used for all biohazardous materials and placed on equipment in which biohazardous materials are used. Frequently used hazard symbols within the department include the following:

Class A:
Compressed Gas

Class B:
Flammable and
Combustible Material

Class C:
Oxidizing Material

Class D:
Poisonous and
infectious Material

Class D:
Materials causing
other toxic effects

Class D:
Biohazardous
infectious material

Class E:
Corrosive Material

Class F:
Dangerously
reactive material

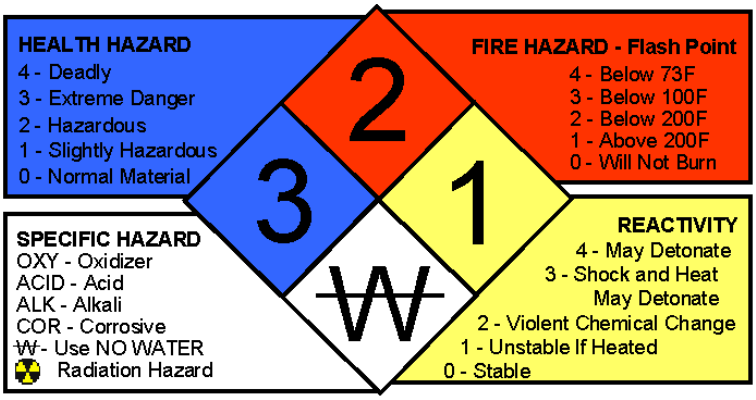
NFPA labels

The National Fire Protection Association's (NFAP) labeling system should be used to indicate the type and the degree of a chemical hazard on all chemicals. Most NFPA labels can be found on chemical labels. However, if a chemical does not contain the NFPA label, you should affix one to it. The NFPA information can be found in section V of most MSDS sheets. Chemical labels containing the NFPA diamond symbol can be obtained from the department's Lab Manager.

NFPA labels are diamond-shaped and color-coded.

Blue indicates the health hazard.
Red indicates the fire hazard.
Yellow indicates the reactivity hazard.
White gives special information such as water or oxidizer incompatibility.

In each field, the degree of the hazard is rated from 0 to 4, with 4 being the greatest hazard and 0 indicating no significant hazard.



Appendix E

Toxic Hazardous Waste Table

Toxic Characteristic Constituents and Regulatory Levels (TCLP)

EPA Waste No.	Constituent	CAS Number	Regulatory Level (mg/L)
D004	Arsenic	7440-38-2	5.0
D005	Barium	7440-39-3	100.0
D006	Cadmium	7440-43-9	1.0
D007	Chromium	7440-47-3	5.0
D008	Lead	7439-92-1	5.0
D009	Mercury	7439-97-6	0.2
D010	Selenium	7782-49-2	1.0
D011	Silver	7440-22-4	5.0
D012	Endrin	72-20-8	0.02
D013	Lindane	58-89-9	0.4
D014	Methoxychlor	72-43-5	10.0
D015	Toxaphene	8000-35-2	0.5
D016	2,4-D	94-75-7	10.0
D017	2,4,5-TP (silvex)	93-72-1	1.0
D018	Benzene	71-43-2	0.5
D019	Carbon Tetrachloride	56-23-5	0.5
D020	Chlordane	57-74-9	0.03
D021	Chlorobenzene	108-90-7	100.0
D022	Chloroform	67-66-3	6.0
D023	o-Cresol	95-48-7	200.0
D024	m-Cresol	108-39-4	200.0
D025	p-Cresol	106-44-5	200.0
D026	Cresol	1319-77-3	200.0
D027	1,4-Dichlorobenzene	106-46-7	7.5
D028	1,2-Dichloroethane	107-06-2	0.5
D029	1,1-Dichloroethylene	75-35-4	0.7
D030	2,4-Dinitrotoluene	121-14-2	0.13
D031	Heptachlor (and its hydroxide)	76-44-8	0.008
D032	Hexachlorobenzene	118-74-1	0.13
D033	Hexachloro-1,3-butadiene	87-68-3	0.5
D034	Hexachloroethane	67-72-1	3.0
D035	Methyl ethyl ketone	78-93-3	200.0
D036	Nitrobenzene	98-95-3	2.0
D037	Pentachlorophenol	87-86-5	100.0
D038	Pyridine	100-86-1	5.0
D039	Tetrachloroethylene	127-18-4	0.7
D040	Trichloroethylene	79-01-6	0.5
D041	2,4,5-Trichlorophenol	95-95-4	400.0
D042	2,4,6-Trichlorophenol	88-06-2	2.0
D043	Vinyl chloride	75-01-4	0.2

Appendix F

P- and U-listed Hazardous Wastes

P-Listed Hazardous Wastes

EPA P-listed wastes are acutely hazardous discarded or off-specification chemical products. To be assigned the P-waste code, these materials must be commercially pure or technical grades of the chemicals listed or the sole active ingredient when in formulations. As acutely hazardous materials they carry special restrictions on their accumulation and disposal. Empty containers and container residues of P-listed wastes are also considered hazardous wastes and must be managed as such.

EPA ID	CAS No.	Substance
P023	107-20-0	Acetaldehyde, chloro-
P002	591-08-2	Acetamide, N-(aminothioxomethyl)-
P057	640-19-7	Acetamide, 2-fluoro-
P058	62-74-8	Acetic acid, fluoro-, sodium salt
P002	591-08-2	1-Acetyl-2-thiourea
P003	107-02-8	Acrolein
P070	116-06-3	Aldicarb
P203	1646-88-4	Aldicarb sulfone.
P004	309-00-2	Aldrin
P005	107-18-6	Allyl alcohol
P006	20859-73-8	Aluminum phosphide (R,T)
P007	2763-96-4	5-(Aminomethyl)-3-isoxazolol
P008	504-24-5	4-Aminopyridine
P009	131-74-8	Ammonium picrate (R)
P119	7803-55-6	Ammonium vanadate
P099	506-61-6	Argentate(1-), bis(cyano-C)-, potassium
P010	7778-39-4	Arsenic acid H3AsO4
P012	1327-53-3	Arsenic oxide As2O3
P011	1303-28-2	Arsenic oxide As2O5
P011	1303-28-2	Arsenic pentoxide
P012	1327-53-3	Arsenic trioxide
P038	692-42-2	Arsine, diethyl-
P036	696-28-6	Arsonous dichloride, phenyl-
P054	151-56-4	Aziridine
P067	75-55-8	Aziridine, 2-methyl-
P013	542-62-1	Barium cyanide
P024	106-47-8	Benzenamine, 4-chloro-
P077	100-01-6	Benzenamine, 4-nitro-
P028	100-44-7	Benzene, (chloromethyl)-
P042	51-43-4	1,2-Benzenediol, 4-[1-hydroxy-2-(methylamino)ethyl]-, (R)-
P046	122-09-8	Benzeneethanamine, alpha,alpha-dimethyl-
P014	108-98-5	Benzenethiol
P127	1563-66-2	7-Benzofuranol, 2,3-dihydro-2,2-dimethyl-, methylcarbamate.
P188	57-64-7	Benzoic acid, 2-hydroxy-, compd. with (3aS-cis)- 1,2,3,3a,8,8a-hexahydro-1,3a,8-trimethylp yrrolo[2,3-b]indol-5-yl methylcarbamate ester (1:1)
P001	fn1 81-81-2	2H-1-Benzopyran-2-one, 4-hydroxy-3-(3-oxo-1-phenylbutyl)-, & salts, when present at concentrations greater than 0.3%

P028	100-44-7	Benzyl chloride
P015	7440-41-7	Beryllium powder
P017	598-31-2	Bromoacetone
P018	357-57-3	Brucine
P045	39196-18-4	2-Butanone, 3,3-dimethyl-1-(methylthio)- O-[methylamino)carbonyl] oxime
P021	592-01-8	Calcium cyanide
P021	592-01-8	Calcium cyanide Ca(CN)2
P189	55285-14-8	Carbamic acid, [(dibutylamino)- thio]methyl-, 2,3-dihydro-2,2-dimethyl- 7-benzofuranyl ester.
P191	644-64-4	Carbamic acid, dimethyl-, 1-[(dimethyl- amino)carbonyl]- 5-methyl-1H-pyrazol- 3-yl ester.
P192	119-38-0	Carbamic acid, dimethyl-, 3-methyl-1- (1-methylethyl)-1H- pyrazol-5-yl ester.
P190	1129-41-5	Carbamic acid, methyl-, 3-methylphenyl ester.
P127	1563-66-2	Carbofuran.
P022	75-15-0	Carbon disulfide
P095	75-44-5	Carbonic dichloride
P189	55285-14-8	Carbosulfan.
P023	107-20-0	Chloroacetaldehyde
P024	106-47-8	p-Chloroaniline
P026	5344-82-1	1-(o-Chlorophenyl)thiourea
P027	542-76-7	3-Chloropropionitrile
P029	544-92-3	Copper cyanide
P029	544-92-3	Copper cyanide Cu(CN)
P202	64-00-6	m-Cumenyl methylcarbamate.
P030		Cyanides (soluble cyanide salts), not otherwise specified
P031	460-19-5	Cyanogen
P033	506-77-4	Cyanogen chloride
P033	506-77-4	Cyanogen chloride (CN)Cl
P034	131-89-5	2-Cyclohexyl-4,6-dinitrophenol
P016	542-88-1	Dichloromethyl ether
P036	696-28-6	Dichlorophenylarsine
P037	60-57-1	Dieldrin
P038	692-42-2	Diethylarsine
P041	311-45-5	Diethyl-p-nitrophenyl phosphate
P040	297-97-2	O,O-Diethyl O-pyrazinyl phosphorothioate
P043	55-91-4	Diisopropylfluorophosphate (DFP)
P004	309-00-2	1,4,5,8-Dimethanonaphthalene, 1,2,3,4, 10,10-hexa- chloro-1,4,4a,5,8,8a,-hexahydro-, (1alpha,4alpha,4abeta, 5alpha,8alpha,8abeta)-
P060	465-7 3-6	1,4,5,8-Dimethanonaphthalene, 1,2,3,4,10,10-hexa- chloro-1,4,4a,5,8,8a-hexahydro-,(1alpha, 4alpha,4abeta,5beta,8beta,8abeta)-
P037	60-57-1	2,7:3,6-Dimethanonaphth[2,3-b]oxirene, 3,4,5,6,9,9- hexachloro-1a,2,2a,3,6,6a,7,7a-octahydro-, (1aalpha,2beta, 2aalpha, 3beta,6beta,6aalpha,7beta, 7aalpha)-
P051	fn1 72-20-8	2,7:3,6-Dimethanonaphth [2,3-b]oxirene, 3,4,5,6,9,9- hexachloro-1a,2,2a,3,6,6a,7,7a-octahydro-, (1aalpha,2beta, 2abeta,3alpha,6alpha,6abeta,7beta, 7aalpha)-, & metabolites
P044	60-51-5	Dimethoate
P046	122-09-8	alpha,alpha-Dimethylphenethylamine
P191	644-64-4	Dimetilan.
P047	fn1 534-52-1	4,6-Dinitro-o-cresol, & salts

P048	51-28-5	2,4-Dinitrophenol
P020	88-85-7	Dinoseb
P085	152-16-9	Diphosphoramide, octamethyl-
P111	107-49-3	Diphosphoric acid, tetraethyl ester
P039	298-04-4	Disulfoton
P049	541-53-7	Dithiobiuret
P185	26419-73-8	1,3-Dithiolane-2-carboxaldehyde, 2,4-dimethyl-, O- [(methylamino)- carbonyl]oxime.
P050	115-29-7	Endosulfan
P088	145-73-3	Endothall
P051	72-20-8	Endrin
P051	72-20-8	Endrin, & metabolites
P042	51-43-4	Epinephrine
P031	460-19-5	Ethane dinitrile
P194	23135-22-0	Ethanimidothioc acid, 2-(dimethylamino)-N- [[(methylamino) carbonyl]oxy]-2 -oxo-, methyl ester.
P066	16752-77-5	Ethanimidothioic acid, N-[[(methylamino) carbonyl]oxy]-, methyl ester
P101	107-12-0	Ethyl cyanide
P054	151-56-4	Ethyleneimine
P097	52-85-7	Famphur
P056	7782-41-4	Fluorine
P057	640-19-7	Fluoroacetamide
P058	62-74-8	Fluoroacetic acid, sodium salt
P198	23422-53-9	Formetanate hydrochloride.
P197	17702-57-7	Formparanate.
P065	628-86-4	Fulminic acid, mercury(2+) salt (R,T)
P059	76-44-8	Heptachlor
P062	757-58-4	Hexaethyl tetraphosphate
P116	79-19-6	Hydrazinecarbothioamide
P068	60-34-4	Hydrazine, methyl-
P063	74-90-8	Hydrocyanic acid
P063	74-90-8	Hydrogen cyanide
P096	7803-51-2	Hydrogen phosphide
P060	465-73-6	Isodrin
P192	119-38-0	Isolan.
P202	64-00-6	3-Isopropylphenyl N-methylcarbamate.
P007	2763-96-4	3(2H)-Isox azolone, 5-(aminomethyl)-
P196	15339-36-3	Manganese, bis(dimethylcarbamodithioato-S,S')-,
P196	15339-36-3	Manganese dimethyldithiocarbamate.
P092	62-38-4	Mercury, (acetato-O)phenyl-
P065	628-86-4	Mercury fulminate (R,T)
1P192	23422-53-9	Methanimidamide, N,N-dimethyl-N'-[3- [[(methylamino)- carbonyl]oxy]phenyl]-, monohydrochloride.
P197	17702-57-7	Methanimidamide, N,N-dimethyl-N'-[2-methyl-4- [[(methylamino)carbonyl]oxy]p henyl]-
P082	62-75-9	Methanamine, N-methyl-N-nitroso-
P064	624-83-9	Methane, isocyanato-
P016	542-88-1	Methane, oxybis[chloro-
P112	509-14-8	Methane, tetranitro- (R)

P118	75-70-7	Methanethiol, trichloro-
P050	115-29-7	6,9-Methano-2,4,3-benzodioxathiepin , 6,7,8,9,10,10- hexachloro-1,5,5a,6,9,9a-hexahydro-, 3-oxide
P059	76-44-8	4,7-Methano-1H-indene, 1,4,5,6,7,8,8-heptachloro- 3a,4,7,7a-tetrahydro-
P199	2032-65-7	Methiocarb.
P066	16752-77-5	Methomyl
P068	60-34-4	Methyl hydrazine
P064	624-83-9	Methyl isocyanate
P069	75-86-5	2-Methyllactonitrile
P071	298-00-0	Methyl parathion
P190	1129-41-5	Metolcarb.
P128	315-18-4	Mexacarbate.
P072	86-88-4	alpha-Naphthylth iourea
P073	13463-39-3	Nickel carbonyl
P073	13463-39-3	Nickel carbonyl Ni(CO)4, (T-4)-
P074	557-19-7	Nickel cyanide
P074	557-19-7	Nickel cynaide Ni(CN)2
P075	fn1 54-11-5	Nicotine, & salts
P076	10102-43-9	Nitric oxide
P077	100-01-6	p-Nitroaniline
P078	10102-44-0	Nitrogen dioxide
P076	10102-43-9	Nitrogen oxide NO
P078	10102-44-0	Nitrogen oxide NO2
P081	55-63-0	Nitroglycerine (R)
P082	62-75-9	N-Nitrosodimethylamine
P084	4549-40-0	N- Nitrosomethylvinylamine
P085	152-16-9	Octamethylpyrophosphor amide
P087	20816-12-0	Osmium oxide OsO4, (T-4)-
P087	20816-12-0	Osmium tetroxide
P088	145-73-3	7-Oxabicyclo[2.2.1]heptane-2,3-dicar boxylic acid
P194	23135-22-0	Oxamyl.
P089	56-38-2	Parathion
P034	131-89-5	Phenol, 2-cyclohexyl-4,6-dinitro-
P048	51-28-5	Phenol, 2,4-dinitro-
P047	fn1 534-52-1	Phenol, 2-methyl-4,6-dinitro-, & salts
P020	88-85-7	Phenol, 2-(1-methylpropyl)-4,6-dinitro-
P009	131-74-8	Phenol, 2,4,6-trinitro-, ammonium salt (R)
P128	315-18-4	Phenol, 4-(dimethylamino)-3,5-dimethyl-, methylcarbamate (ester).
P199	2032-65-7	Phenol, (3,5-dimethyl-4-(methylthio)-, methylcarbamate
P202	64-00-6	Phenol, 3-(1-methylethyl)-, methyl carbamate.
P201	2631-37-0	Phenol, 3-methyl-5-(1-methylethyl)-, methyl carbamate.
P092	62-38-4	Phenylmercury acetate
P093	103-85-5	Phenylthiourea
P094	298-02-2	Phorate
P095	75-44-5	Phosgene
P096	7803-51-2	Phosphine
P041	311-45-5	Phosphoric acid, diethyl 4-nitrophenyl ester

P039	298-04-4	Phosphorodithioic acid, O,O-diethyl S-[2-(ethylthio)ethyl] ester
P094	298-02-2	Phosphorodithioic acid, O,O-diethyl S-[(ethylthio)methyl] ester
P044	60-51-5	Phosphorodithioic acid, O,O-dimethyl S-[2-(methylamino)- 2-oxoethyl] ester
P043	55-91-4	Phosphorofluoridic acid, bis(1-methylethyl) ester
P089	56-38-2	Phosphorothioic aci O,O-dimethyl ester
P040	297-97-2	Phosphorothioic acid, O,O-diethyl O-pyrazinyl ester
P097	52-85-7	Phosporothioic acid, O-[4-[(dimethylamino) sulfonyl]phenyl]0,0-dimethyl ester
P071	298-00-0	Phosphorothioic acid, O,O,-dimethyl O-(4-nitrophenyl) ester
P204	57-47-6	Physostigmine.
P188	57-64-7	Physostigmine salicylate.
P110	78-00-2	Plumbane, tetraethyl-
P098	151-50-8	Potassium cyanide
P098	151-50-8	Potassium cyanide K(CN)
P099	506-61-6	Potassium silver cyanide
P201	2631-37-0	Promecarb
P070	116-06-3	Propanal, 2-methyl-2-(methylthio)-, O-[(methylamino)carbonyl]oxime
P203	1646-88-4	Propanal, 2-methyl-2-(methyl-sulfonyl)-, O-[(methylamino)carbonyl] oxime.
P101	107-12-0	Propanenitrile
P027	542-76-7	Propanenitrile, 3-chloro-
P069	75-86-5	Propanenitrile, 2-hydroxy-2-methyl-
P081	55-63-0	1,2,3-Propanetriol, trinitrate (R)
P017	598-31-2	2-Propanone, 1-bromo-
P102	107-19-7	Propargyl alcohol
P003	107-02-8	2-Propenal
P005	107-18-6	2-Propen-1-ol
P067	75-55-8	1,2-Propylenimine
P102	107-19-7	2-Propyn-1-ol
P008	504-24-5	4-Pyridinamine
P075	fn1 54-11-5	Pyridine, 3-(1-methyl-2-pyrrolidinyl)-, (S)-, & salts
P204	57-47-6	Pyrrolo[2,3-b]indol-5-ol, 1,2,3,3a,8,8a- hexahydro-1,3a,8-trimethyl-, methylcarbamate (ester), (3aS-cis)-
P114	12039-52-0	Selenious acid, dithallium(1+) salt
P103	630-10-4	Selenourea
P104	506-64-9	Silver cyanide
P104	506-64-9	Silver cyanide Ag(CN)
P105	26628-22-8	Sodium azide
P106	143-33-9	Sodium cyanide
P106	143-33-9	Sodium cyanide Na(CN)
P108	fn1 57-24-9	Strychnidin-10-one, & salts
P018	357-57-3	Strychnidin-10-one, 2,3-dimethoxy-
P108	fn1 57-24-9	Strychnine, & salts
P115	7446-18-6	Sulfuric acid, dithallium(1+) salt
P109	3689-24-5	Tetraethyldithiopyrophosphate
P110	78-00-2	Tetraethyl lead
P111	107-49-3	Tetraethyl pyrophosphate
P112	509-14-8	Tetranitromethane (R)

P062	757-58-4	Tetraphosphoric acid, hexaethyl ester
P113	1314-32-5	Thallic oxide
P113	1314-32-5	Thallium oxide Tl2O3
P114	12039-52-0	Thallium(I) selenite
P115	7446-18-6	Thallium(I) sulfate
P109	3689-24-5	Thiodiphosphoric acid, tetraethyl ester
P045	39196-18-4	Thiofanox
P049	541-53-7	Thioimidodi carbonic diamide [(H2N)C(S)]2NH
P014	108-98-5	Thiophenol
P116	79-19-6	Thiosemicarbazide
P026	5344-82-1	Thiourea, (2-chlorophenyl)-
P072	86-88-4	Thiourea, 1-naphthalenyl-
P093	103-85-5	Thiourea, phenyl-
P185	26419-73-8	Tirpate.
P123	8001-35-2	Toxaphene
P118	75-70-7	Trichlorome thanethiol
P119	7803-55-6	Vanadic acid, ammonium salt
P120	1314-62-1	Vanadium oxide V2O5
P120	1314-62-1	Vanadium pentoxide
P084	4549-40-0	Vinylamine, N-methyl-N-nitroso-
P001	81-81-2	Warfarin, & salts, when present at concentrations greater than 0.3%
P205	137-30-4	Zinc, bis(dimethylcarbamodithioato-S,S)-
P121	557-21-1	Zinc cyanide
P121	557-21-1	Zinc cyanide Zn(CN)2
P122	1314-84-7	Zinc phosphide Z[3]P[2], when present at concentrations greater than 10% (R,T)
P205	137-30-4	Ziram.

U-Listed Hazardous Wastes

EPA U-listed hazardous wastes are toxic or otherwise hazardous discarded or off-specification commercial chemical products. As with P-listed hazardous wastes, U-listed materials are commercially pure or technical grades of the chemicals listed or the sole active ingredient when in formulation. U-listed hazardous wastes do not carry the special accumulation and disposal requirements of P-listed wastes.

EPA ID No.	CAS No.	Substance
U394	30558-43-1	A2213
U001	75-07-0	Acetaldehyde (I)
U034	75-87-6	Acetaldehyde, trichloro-
U187	62-44-2	Acetamide, N-(4-ethoxyphenyl)-
U005	53-96-3	Acetamide, N-9H-fluoren-2-yl-
U112	141-78-6	Acetic acid ethyl ester (I)
U144	301-04-2	Acetic acid, lead(2+) salt
U214	563-68-8	Acetic acid, thallium(1+) salt
U002	67-64-1	Acetone (I)
U003	75-05-8	Acetonitrile (I,T)
U004	98-86-2	Acetophenone
U005	53-96-3	2-Acetylaminofluorene
U006	75-36-5	Acetyl chloride (C,R,T)
U007	79-06-1	Acrylamide
U008	79-10-7	Acrylic acid (I)
U009	107-13-1	Acrylonitrile
U011	61-82-5	Amitrole
U012	62-53-3	Aniline (I,T)
U136	75-60-5	Arsinic acid, dimethyl-
U014	492-80-8	Auramine
U015	115-02-6	Azaserine
U280	101-27-9	Barban.
U278	22781-23-3	Bendiocarb.
U364	22961-82-6	Bendiocarb phenol.
U271	17804-35-2	Benomyl.
U016	225-51-4	Benz[c]acridine
U017	9 8-87-3	Benzal chloride
U018	56-55-3	Benz[a]anthracene
U094	57-97-6	Benz[a]anthracene, 7,12-dimethyl-
U012	62-53-3	Benzenamine (I,T)
U014	492-80-8	Benzenamine, 4,4 -carbonimidoylbis
U049	3165-93-3	Benzenamine, 4-chloro-2-methyl-, hydrochloride

U093	60-11-7	Benzenamine, N,N-dimethyl-4-(phenylazo)-
U328	95-53-4	Benzenamine, 2-methyl-
U353	106-49-0	Benzenamine, 4-methyl-
U158	101-14-4	Benzenamine, 4,4 -methylenebis[2-chloro-
U222	636-21-5	Benzenamine, 2-methyl-, hydrochloride
U181	99-55-8	Benzenamine, 2-methyl-5-nitro-
U019	71-43-2	Benzene (I,T)
U038	510-15-6	Benzeneacetic acid, 4-chloro-alpha-(4-chlorophenyl)-alpha-hydroxy-, ethyl ester
U030	101-55-3	Benzene, 1-bromo-4-phenoxy-
U035	305-03-3	Benzenebutanoic acid, 4-[bis(2-chloroethyl)amino]-
U037	108-90-7	Benzene, chloro-
U221	25376-45-8	Benzenediamine, ar-methyl-
U028	117-81-7	1,2-Benzenedicarboxylic acid, bis(2-ethylhexyl) ester
U069	84-74-2	1,2-Benzenedicarboxylic acid, dibutyl ester
U088	84-66-2	1,2-Benzenedicarboxylic acid, diethyl ester
U102	131-11-3	1,2-Benzenedicarboxylic acid, dimethyl ester
U107	117-84-0	1,2-Benzenedicarboxylic acid, dioctyl ester
U070	95-50-1	Benzene, 1,2-dichloro-
U071	541-73-1	Benzene, 1,3-dichloro-
U072	106-46-7	Benzene, 1,4-dichloro-
U060	72-54-8	Benzene, 1,1 -(2,2-dichloroethylidene)bis[4-chloro-
U017	98-87-3	Benzene, (dichloromethyl)-
U223	26471-62-5	Benzene, 1,3-diisocyanatomethyl- (R,T)
U239	1330-20-7	Benzene, dimethyl- (I,T)
U201	108-46-3	1,3-Benzenediol
U127	118-74-1	Benzene , hexachloro-
U056	110-82-7	Benzene, hexahydro- (I)
U220	108-88-3	Benzene, methyl-
U105	121-14-2	Benzene, 1-methyl-2,4-dinitro-
U106	606-20-2	Benzene, 2-methyl-1,3-dinitro-
U055	98-82-8	Benzene, (1-methylethyl)- (I)
U169	98-95-3	Benzene, nitro-
U183	608-93-5	Benzene, pentachloro-
U185	82-68-8	Benzene, pentachloronitro-
U020	98-09-9	Benzenesulfonic acid chloride (C,R)
U020	98-09-9	Benzenesulfonyl chloride (C,R)
U207	95-94-3	Benzene, 1,2,4,5-tetrachloro-
U061	50-29-3	Benzene, 1,1 -(2,2,2-trichloroethylidene)bis[4-chloro-
U247	72-43-5	Benzene, 1,1 -(2,2,2-trichloroethylidene)bis[4- methoxy-
U023	98-07-7	Benzene, (trichloromethyl)-

U234	99-35-4	Benzene, 1,3,5-trinitro-
U021	92-87-5	Benzidine
U202	fn1 81-07-2	1,2-Benzisothiazol-3(2H)-one, 1,1-dioxide, & salts
U278	22781-23-3	1,3-Benzodioxol-4-ol, 2,2-dimethyl-, methyl carbamate
U364	22961-82-6	1,3-Benzodioxol-4-ol, 2,2-dimethyl-,
U203	94-59-7	1,3-Benzodioxole, 5-(2-propenyl)-
U141	120-58-1	1,3-Benzodioxole, 5-(1-propenyl)-
U367	1563-38-8	7-Benzofuranol, 2,3-dihydro-2,2-dimethyl-
U090	94-58-6	1,3-Benzodioxole, 5-propyl-
U064	189-55-9	Benzo[rst_pentaphene
U248	n1 81-81-2	2H-1-Benzopyran-2-one, 4-hydroxy-3-(3-oxo-1-phenyl-butyl)-, & salts, when present at concentrations of 0.3% or less
U022	50-32-8	Benzo[a_pyrene
U197	106-51-4	p-Benzoquinone
U023	98-07-7	Benzotrichloride (C,R,T)
U085	1464-53-5	2,2 -Bioxirane
U021	92-87-5	[1,1 -Biphenyl_-4,4 -diamine
U073	91-94-1	[1,1'-Biphenyl_-4,4'-diamine, 3,3'-dichloro-
U091	119-90-4	[1,1'-Biphenyl_-4,4'-diamine, 3,3'-dimethoxy-
U095	119-93-7	[1,1'-Biphenyl_-4,4'-diamine, 3,3'-dimethyl-
U225	75-25-2	Bromoform
U030	101-55-3	4-Bromophenyl phenyl ether
U128	87-68-3	1,3-Butadiene, 1,1,2,3,4,4-hexachloro-
U172	924-16-3	1-Butanamine, N-butyl-N-nitroso-
U031	71-36-3	1-Butanol (I)
U159	78-93-3	2-Butanone (I,T)
U160	1338-23-4	2-Butanone, peroxide (R,T)
U053	4170-30-3	2-Butenal
U074	764-41-0	2-Butene, 1,4-dichloro- (I,T)
U143	303-34-4	2-Butenoic acid, 2-methyl-, 7-[[2,3-dihydroxy- 2-(1-methoxyethyl)-3-methyl-1-oxobutoxy _methyl_-2,3,5,7a-tetrahydro-1H-pyrrolizin-1-yl ester, [1S-[1alpha(Z), 7(2S*,3R*),7aalpha__-
U031	71-36-3	n-Butyl alcohol (I)
U136	75-60-5	Cacodylic acid
U032	13765-19-0	Calcium chromate
U372	10605-21-7	Carbamic acid, 1H-benzimidazol-2-yl, methyl ester.
U271	17804-35-2	Carbamic acid, [1-[(butylamino)carbonyl -1H-benzimidazol-2-yl -, methyl ester.
U280	101-27-9	Carbamic acid, (3-chlorophenyl)-, 4-chloro- 2-butynyl ester.
U238	51-79-6	Carbamic acid, ethyl ester
U178	615-53-2	Carbamic acid, methylnitroso-, ethyl ester
U373	122-42-9	Carbamic acid, phenyl-, 1-methylethyl ester.

U409	23564-05-8	Carbamic acid, [1,2-phenylenebis (iminocarbonothioyl) bis-, dimethyl ester.
U097	79-44-7	Carbamic chloride, dimethyl-
U114	111-54-6	Carbamodithioic acid, 1,2-ethanediylbis-,salts & esters
U062	2303-16-4	Carbamothioic acid, bis(1-methylethyl)-, S-(2,3-dichloro-2-propenyl) ester
U389	2303-17-5	Carbamothioic acid, bis(1-methylethyl)-, S- (2,3,3-trichloro-2-propenyl) ester.
U387	52888-80-9	Carbamothioic acid, dipropyl-, S- (phenylmethyl) ester.
U279	63-25-2	Carbaryl.
U372	10605-21- 7	Carbendazim.
U367	1563-38-8	Carbofuran phenol.
U215	6533-73-9	Carbonic acid, dithallium(1+) salt
U033	353-50-4	Carbonic difluoride
U156	79-22-1	Carbonochloridic acid, methyl ester (I,T)
U033	353-50-4	Carbon oxyfluoride (R,T)
U211	56-23-5	Carbon tetrachloride
U034	75-87-6	Chloral
U035	305-03-3	Chlorambucil
U036	57-74-9	Chlordane, alpha & gamma isomers
U026	494-03-1	Chlornaphazin
U037	108-90-7	Chlorobenzene
U038	510-15-6	Chlorobenzilate
U039	59-50-7	p-Chloro-m-cresol
U042	110-75-8	2-Chloroethyl vinyl ether
U044	67-66-3	Chloroform
U046	107-30-2	Chloromethyl methyl ether
U047	91-58-7	beta-Chloronaphthalene
U048	95-57-8	o-Chlorophenol
U049	3165-93-3	4-Chloro-o-toluidine, hydrochloride
U032	13765-19-0	Chromic acid H2CrO4, calcium salt
U050	218-01-9	Chrysene
U051		Creosote
U052	1319-77-3	Cresol (Cresylic acid)
U053	4170-30-3	Crotonaldehyde
U055	98-82-8	Cumene (I)
U246	506-68-3	Cyanogen bromide (CN)Br
U197	106-51-4	2,5-Cyclohexadiene-1,4-dione
U056	110-82-7	Cyclohexane (I)
U129	58-89-9	Cyclohexane, 1,2,3,4,5,6-hexachloro-, (1alpha,2alpha,3beta,4alpha,5alph a,6beta)-
U057	108-94-1	Cyclohexanone (I)
U130	77-47-4	1,3-Cyclopentadiene, 1,2,3,4,5,5-hexachloro-
U058	50-18-0	Cyclophosphamide

U240	n1 94-75-7	2,4-D, salts & esters
U059	20830-81-3	Daunomycin
U060	72-54-8	DDD
U061	50-29-3	DDT
U062	2303-16-4	Diallate
U063	53-70-3	Dibenz[a,h_anthracene
U064	189-55-9	Dibenzo[a,i_pyrene
U066	96-12-8	1,2-Dibromo-3-chloropropane
U069	84-74-2	Dibutyl phthalate
U070	95-50-1	o-Dichlorobenzene
U071	541-73-1	m-Dichlorobenzene
U072	106-46-7	p-Dichlorobenzene
U073	91-94-1	3,3'-Dichlorobenzidine
U074	764-41-0	1,4-Dichloro-2-butene (I,T)
U075	75-71-8	Dichlorodifluoromethane
U078	75-35-4	1,1-Dichloroethylene
U079	156-60-5	1,2-Dichloroethylene
U025	111-44-4	Dichloroethyl ether
U027	108-60-1	Dichloroisopropyl ether
U024	111-91-1	Dichloromethoxy ethane
U081	120-83-2	2,4-Dichlorophenol
U082	87-65-0	2,6-Dichlorophenol
U084	542-75-6	1,3-Dichloropropene
U085	1464-53-5	1,2:3,4-Diepoxybutane (I,T)
U395	5952-26-1	Diethylene glycol, dicarbamate.
U108	123-91-1	1,4-Diethyleneoxide
U028	117-81-7	Diethylhexyl phthalate
U086	1615-80-1	N,N'-Diethylhydrazine
U087	3288-58-2	O,O-Diethyl S-methyl dithiophosphate
U088	84-66-2	Diethyl phthalate
U089	56-53-1	Diethylstilbesterol
U090	94-58-6	Dihydrosafrole
U091	119-90-4	3,3'-Dimethoxybenzidine
U092	124-40-3	Dimethylamine (I)
U093	60-11-7	p-Dimethylaminoazobenzene
U094	57-97-6	7,12-Dimethylbenz[a_anthracene
U095	119-93-7	3,3'-Dimethylbenzidine
U096	80-15-9	alpha,alpha-Dimethylbenzylhydroperoxide (R)
U097	79-44-7	Dimethylcarbamoyl chloride
U098	57-14-7	1,1-Dimethylhydrazine
U099	540-73-8	1,2-Dimethylhydrazine

U101	105-67-9	2,4-Dimethylphenol
U102	131-11-3	Dimethyl phthalate
U103	77-78-1	Dimethyl sulfate
U105	121-14-2	2,4-Dinitrotoluene
U106	606-20-2	2,6-Dinitrotoluene
U107	117-84-0	Di-n-octyl phthalate
U108	123-91-1	1,4-Dioxane
U109	122-66-7	1,2-Diphenylhydrazine
U110	142-84-7	Dipropylamine (1)
U111	621-64-7	Di-n-propylnitrosamine
U041	106-89-8	Epichlorohydrin
U001	75-07-0	Ethanal (1)
U404	121-44-8	Ethanamine, N,N-diethyl-
U174	55-18-5	Ethanamine, N-ethyl-N-nitroso-
U155	91-80-5	1,2-Ethanediamine, N,N-dimethyl-N'-2-pyridinyl-N'- (2-thienylmethyl)-
U067	106-93-4	Ethane, 1,2-dibromo-
U076	75-34-3	Ethane, 1,1-dichloro-
U077	107-06-2	Ethane, 1,2-dichloro-
U131	67-72-1	Ethane, hexachloro-
U024	111-91-1	Ethane, 1,1'-[methylenebis(oxy)_bis[2-chloro-
U117	60-29-7	Ethane, 1,1'-oxybis-(I)
U025	111-44-4	Ethane, 1,1'-oxybis[2-chloro-
U184	76-01-7	Ethane, pentachloro-
U208	630-20-6	Ethane, 1,1,1,2-tetrachloro-
U209	79-34-5	Ethane, 1,1,2,2-tetrachloro-
U218	62-55-5	Ethanethioamide
U226	71-55-6	Ethane, 1,1,1-trichloro-
U227	79-00-5	Ethane, 1,1,2-trichloro-
U410	59669-26-0	Ethanimidothioic acid, N,N'- [thiobis [(methylimino)carbonyloxy__bis-, dimethyl ester
U394	30558-43-1	Ethanimidothioic acid, 2-(dimethylamino)-N- hydroxy-2-oxo-, methyl ester.
U359	110-80-5	Ethanol, 2-ethoxy-
U173	1116-54-7	Ethanol, 2,2'-(nitrosoimino)bis-
U395	5952-26-1	Ethanol, 2,2 -oxybis-, dicarbamate.
U004	98-86-2	Ethanone, 1-phenyl-
U043	75-01-4	Ethene, chloro-
U042	110-75-8	Ethene, (2-chloroethoxy)-
U078	75-35-4	Ethene, 1,1-dichloro-
U079	156-60-5	Ethene, 1,2-dichloro-, (E)-
U210	127-18-4	Ethene, tetrachloro-

U228	79-01-6	Ethene, trichloro-
U112	141-78-6	Ethyl acetate (I)
U113	140-88-5	Ethyl acrylate (I)
U238	51-79-6	Ethyl carbamate (urethane)
U117	60-29-7	Ethyl ether (I)
U114	n1 111-54-6	Ethylenebisdithiocarbamic acid, salts & esters
U067	106-93-4	Ethylene dibromide
U077	107-06-2	Ethylene dichloride
U359	110-80-5	Ethylene glycol monoethyl ether
U115	75-21-8	Ethylene oxide (I,T)
U116	96-45-7	Ethylenethiourea
U076	75-34-3	Ethylidene dichloride
U118	97-63-2	Ethyl methacrylate
U119	62-50-0	Ethyl methanesulfonate
U120	206-44-0	Fluoranthene
U122	50-00-0	Formaldehyde
U123	64-18-6	Formic acid (C,T)
U124	110-00-9	Furan (I)
U125	98-01-1	2-Furancarboxaldehyde (I)
U147	108-31-6	2,5-Furandione
U213	109-99-9	Furan, tetrahydro-(I)
U125	98-01-1	Furfural (I)
U124	110-00-9	Furfuran (I)
U206	18883-66-4	Glucopyranose, 2-deoxy-2-(3-methyl-3-nitrosoureido)-, D-
U206	18883-66-4	D-Glucose, 2-deoxy-2-[[(methylnitrosoamino)- carbonyl]amino]-
U126	765-34-4	Glycidylaldehyde
U163	70-25-7	Guanidine, N-methyl-N'-nitro-N-nitroso-
U127	118-74-1	Hexachlorobenzene
U128	87-68-3	Hexachlorobutadiene
U130	77-47-4	Hexachlorocyclopentadiene
U131	67-72-1	Hexachloroethane
U132	70-30-4	Hexachlorophene
U243	1888-71-7	Hexachloropropene
U133	302-01-2	Hydrazine (R,T)
U086	1615-80-1	Hydrazine, 1,2-diethyl-
U098	57-14-7	Hydrazine, 1,1-dimethyl-
U099	540-73-8	Hydrazine, 1,2-dimethyl-
U109	122-66-7	Hydrazine, 1,2-diphenyl-
U134	7664-39-3	Hydrofluoric acid (C,T)
U134	7664-39-3	Hydrogen fluoride (C,T)
U135	7783-06-4	Hydrogen sulfide

U135	7783-06-4	Hydrogen sulfide H2S
U096	80-15-9	Hydroperoxide, 1-methyl-1-phenylethyl- (R)
U116	96-45-7	2-Imidazolidinethione
U137	193-39-5	Indeno[1,2,3-cd]pyrene
U190	85-44-9	1,3-Isobenzofurandione
U140	78-83-1	Isobutyl alcohol (I,T)
U141	120-58-1	Isosafrole
U142	143-50-0	Kepone
U143	303-34-4	Lasiocarpine
U144	301-04-2	Lead acetate
U146	1335-32-6	Lead, bis(acetato-O)tetrahydroxytri-
U145	7446-27-7	Lead phosphate
U146	1335-32-6	Lead subacetate
U129	58-89-9	Lindane
U163	70-25-7	MNNG
U147	108-31-6	Maleic anhydride
U148	123-33-1	Maleic hydrazide
U149	109-77-3	Malononitrile
U150	148-82-3	Melphalan
U151	7439-97-6	Mercury
U152	126-98-7	Methacrylonitrile (I, T)
U092	124-40-3	Methanamine, N-methyl- (I)
U029	74-83-9	Methane, bromo-
U045	74-87-3	Methane, chloro- (I, T)
U046	107-30-2	Methane, chloromethoxy-
U068	74-95-3	Methane, dibromo-
U080	75-09-2	Methane, dichloro-
U075	75-71-8	Methane, dichlorodifluoro-
U138	74-88-4	Methane, iodo-
U119	62-50-0	Methanesulfonic acid, ethyl ester
U211	56-23-5	Methane, tetrachloro-
U153	74-93-1	Methanethiol (I, T)
U225	75-25-2	Methane, tribromo-
U044	67-66-3	Methane, trichloro-
U121	75-69-4	Methane, trichlorofluoro-
U036	57-74-9	4,7-Methano-1H-indene, 1,2,4,5,6,7,8,8-octachloro- 2,3,3a,4,7,7a-hexahydro-
U154	67-56-1	Methanol (I)
U155	91-80-5	Methapyrilene
U142	143-50-0	1,3,4-Metheno-2H-cyclobuta[cd]pentalen-2-one, 1,1a,3,3a,4,5,5,5a,5b,6-decachlorooctahydro-
U247	72-43-5	Methoxychlor

U154	67-56-1	Methyl alcohol (I)
U029	74-83-9	Methyl bromide
U186	504-60-9	1-Methylbutadiene (I)
U045	74-87-3	Methyl chloride (I,T)
U156	79-22-1	Methyl chlorocarbonate (I,T)
U226	71-55-6	Methyl chloroform
U157	56-49-5	3-Methylcholanthrene
U158	101-14-4	4,4'-Methylenebis(2-chloroaniline)
U068	74-95-3	Methylene bromide
U080	75-09-2	Methylene chloride
U159	78-93-3	Methyl ethyl ketone (MEK) (I,T)
U160	1338-23-4	Methyl ethyl ketone peroxide (R,T)
U138	74-88-4	Methyl iodide
U161	108-10-1	Methyl isobutyl ketone (I)
U162	80-62-6	Methyl methacrylate (I,T)
U161	108-10-1	4-Methyl-2-pentanone (I)
U164	56-04-2	Methylthiouracil
U010	50-07-7	Mitomycin C
U059	20830-81-3	5,12-Naphthacenedione, 8-acetyl-10- [(3-amino-2,3,6-trideoxy)-alpha-L-lyxo-hexopy ranosyl) oxy-7,8,9,10-tetrahydro-6,8,11-trihydroxy-1-meth oxy-(8S-cis)-
U167	134-32-7	1-Naphthalenamine
U168	91-59-8	2-Naphthalenamine
U026	494-03-1	Naphthalenamine, N,N'-bis(2-chloroethyl)-
U165	91-20-3	Naphthalene
U047	91-58-7	Naphthalene, 2-chloro-
U166	130-15-4	1,4-Naphthalenedione
U236	72-57-1	2,7-Naphthalenedisulfonic acid, 3,3'-[(3,3'- dimethyl[1,1'-biphenyl]-4,4'-diyl)bis(azo)bis [5-amino-4-hydroxy]-, tetrasodium salt
U279	63-25-2	1-Naphthalenol, methylcarbamate.
U166	130-15-4	1,4-Naphthoquinone
U167	134-32-7	alpha -Naphthylamine
U168	91-59-8	beta-Naphthylamine
U217	10102-45-1	Nitric acid, thallium(1+) salt
U169	98-95-3	Nitrobenzene (I,T)
U170	100-02-7	p-Nitrophenol
U171	79-46-9	2-Nitropropane (I,T)
U172	924-16-3	N-Nitrosodi-n-butylamine
U173	1116-54-7	N-Nitrosodiethanolamine
U174	55-18-5	N-Nitrosodiethylamine
U176	759-73-9	N-Nitroso-N-ethylurea
U177	684-93-5	N-Nitroso-N-methylurea

U178	615-53-2	N-Nitroso-N-methylurethane
U179	100-75-4	N-Nitrosopiperidine
U180	930-55-2	N-Nitrosopyrrolidine
U181	99-55-8	5-Nitro-o-toluidine
U193	1120-71-4	1,2-Oxathiolane, 2,2-dioxide
U058	50-18-0	2H-1,3,2-Oxazaphosphorin-2-amine, N,N-bis(2-chloroethyl)tetrahydro-, 2-oxide
U115	75-21-8	Oxirane (I,T)
U126	765-34-4	Oxiranecarboxyaldehyde
U041	106-89-8	Oxirane, (chloromethyl)-
U182	123-63-7	Paraldehyde
U183	608-93-5	Pentachlorobenzene
U184	76-01-7	Pentachloroethane
U185	82-68-8	Pentachloronitrobenzene (PCNB)
F027	87-86-5	Pentachlorophenol
U161	108-10-1	Pentanol, 4-methyl-
U186	504-60-9	1,3-Pentadiene (I)
U187	62-44-2	Phenacetin
U188	108-95-2	Phenol
U048	95-57-8	Phenol, 2-chloro-
U039	59-50-7	Phenol, 4-chloro-3-methyl-
U081	120-83-2	Phenol, 2,4-dichloro-
U082	87-65-0	Phenol, 2,6-dichloro-
U089	56-53-1	Phenol, 4,4'-(1,2-diethyl-1,2-ethenediyl)bis-, (E)-
U101	105-67-9	Phenol, 2,4-dimethyl-
U052	1319-77-3	Phenol, methyl-
U132	70-30-4	Phenol, 2,2'-methylenebis[3,4,6-trichloro-
U411	114-26-1	Phenol, 2-(1-methylethoxy)-, methylcarbamate
U170	100-02-7	Phenol, 4-nitro-
See F027	87-86-5	Phenol, pentachloro-
See F027	58-90-2	Phenol, 2,3,4,6-tetrachloro-
See F027	95-95-4	Phenol, 2,4,5-trichloro-
See F027	88-06-2	Phenol, 2,4,6-trichloro-
U150	148-82-3	L-Phenylalanine, 4-[bis(2-chloroethyl)amino]-
U145	7446-27-7	Phosphoric acid, lead(2+) salt (2:3)
U087	3288-58-2	Phosphorodithioic acid, O,O-diethyl S-methyl ester
U189	1314-80-3	Phosphorus sulfide (R)
U190	85-44-9	Phthalic anhydride
U191	109-06-8	2-Picoline
U179	100-75-4	Piperidine, 1-nitroso-
U192	23950-58-5	Pronamide

U194	107-10-8	1-Propanamine (I,T)
U111	621-64-7	1-Propanamine, N-nitroso-N-propyl-
U110	142-84-7	1-Propanamine, N-propyl- (I)
U066	96-12-8	Propane, 1,2-dibromo-3-chloro-
U083	78-87-5	Propane, 1,2-dichloro-
U149	109-77-3	Propanedinitrile
U171	79-46-9	Propane, 2-nitro- (I,T)
U027	108-60-1	Propane, 2,2'-oxybis[2-chloro-
U193	1120-71-4	1,3-Propane sultone
See F027	93-72-1	Propanoic acid, 2-(2,4,5-trichlorophenoxy)-
U235	126-72-7	1-Propanol, 2,3-dibromo-, phosphate (3:1)
U140	78-83-1	1-Propanol, 2-methyl- (I,T)
U002	67-64-1	2-Propanone (I)
U007	79-06-1	2-Propenamide
U084	542-75-6	1-Propene, 1,3-dichloro-
U243	1888-71-7	1-Propene, 1,1,2,3,3,3-hexachloro-
U009	107-13-1	2-Propenenitrile
U152	126-98-7	2-Propenenitrile, 2-methyl- (I,T)
U008	79-10-7	2-Propenoic acid (I)
U113	140-88-5	2-Propenoic acid, ethyl ester (I)
U118	97-63-2	2-Propenoic acid, 2-methyl-, ethyl ester
U162	80-62-6	2-Propenoic acid, 2-methyl-, methyl ester (I,T)
U373	122-42-9	Propham.
U411	114-26-1	Propoxur.
U387	52888-80-9	Prosulfocarb.
U194	107-10-8	n-Propylamine (I,T)
U083	78-87-5	Propylene dichloride
U148	123-33-1	3,6-Pyridazinedione, 1,2-dihydro-
U196	110-86-1	Pyridine
U191	109-06-8	Pyridine, 2-methyl-
U237	66-75-1	2,4-(1H,3H)-Pyrimidinedione, 5-[bis(2- chloroethyl)amino]- 4(1H)-Pyrimidinone, 2,3-dihydro-6-methyl-2-thioxo-
U180	930-55-2	Pyrrolidine, 1-nitroso-
U200	50-55-5	Reserpine
U202	fn1 81-07-2	Saccharin, & salts
U203	94-59-7	Safrole
U204	7783-00-8	Selenious acid
U204	7783-00-8	Selenium dioxide
U205	7488-56-4	Selenium sulfide
U205	7488-56-4	Selenium sulfide SeS2 (R,T)
U015	115-02-6	L-Serine, diazoacetate (ester)

See F027	93-72-1	Silvex (2,4,5-TP)
U206	18883-66-4	Streptozotocin
U103	77-78-1	Sulfuricacid, dimethyl ester
U189	1314-80-3	Sulfur phosphide (R)
See F027	93-76-5	2,4,5-T
U207	95-94-3	1,2,4,5-Tetrachlorobenzene
U208	630-20-6	1,1,1,2-Tetrachloroethane
U209	79-34-5	1,1,2,2-Tetrachloroethane
U210	127-18-4	Tetrachloroethylene
See F027	58-90-2	2,3,4,6-Tetrachlorophenol
U213	109-99-9	Tetra hydrofuran (I)
U214	563-68-8	Thallium(I) acetate
U215	6533-73-9	Thallium(I) carbonate
U216	7791-12-0	Thallium(I) chloride
U216	7791-12-0	Thallium chloride Tlcl
U217	10102-45-1	Thallium(I) nitrate
U218	62-55-5	Thioacetamide
U410	59669-26-0	Thiodicarb.
U153	74-93-1	Thiomethanol (I,T)
U244	137-26-8	Thioperoxydicarbonic diamide [(H2N)C(S)]2S2, tetramethyl-
U409	23564-05-8	Thiophanate-methyl.
U219	62-56-6	Thiourea
U244	137-26-8	Thiram
U220	108-88-3	Toluene
U221	25376-45-8	Toluenediamine
U223	26471-62-5	Toluene diisocyanate (R,T)
U328	95-53-4	o-Toluidine
U353	106-49-0	p-Toluidine
U222	636-21-5	o-Toluidine hydrochloride
U389	2303-17-5	Triallate.
U011	61-82-5	1H-1,2,4-Triazol-3-amine
U227	79-00-5	1,1,2-Trichloroethane
U228	79-01-6	Trichloroethylene
U121	75-69-4	Trichloromonofluoromethane
See F027	95-95-4	2,4,5-Trichlorophenol
See F027	88-06-2	2,4,6-Trichlorophenol
U404	121-44-8	Triethylamine.
U234	99-35-4	1,3,5-Trinitrobenzene (R,T)
U182	123-63-7	1,3,5-Trioxane, 2,4,6-trimethyl-
U235	126-72-7	Tris(2,3-dibromopropyl) phosphate

U236	72-57-1	Trypan blue
U237	66-75-1	Uracil mustard
U176	759-73-9	Urea, N-ethyl-N-nitroso-
U177	684-93-5	Urea, N-methyl-N-nitroso-
U043	75-01-4	Vinyl chloride
U248	n1 81-81-2	Warfarin, & salts, when present at concentrations of 0.3% or less
U239	1330-20-7	Xylene (I)
U200	50-55-5	Yohimban-16-carboxylic acid, 11,17-dimethoxy-18- [(3,4,5-trimethoxybenzoyl) oxy]-, methyl ester,(3beta,16beta, 17alpha,18beta,20alpha)-
U249	1314-84-7	Zinc phosphide Z[3]P[2], when present at concentrations of 10% or less

Appendix G

Satellite Accumulation Areas and Signage

Biology Department Satellite Accumulation Areas

Location	Principle investigator or Laboratory supervisor	Description	Waste satellite accumulation area (SAA)
Dalton Hall 109	Salvatore Blair	Faculty Research Lab	Yes – fume hood, area under the fume hood, lab bench
Dalton Hall 113	Kiyoshi Sasaki	Faculty Research Lab	Yes - fume hood
Dalton Hall 115	Laura Glasscock	Faculty Research Lab	Yes - lab bench
Dalton Hall 119	Daniel Stovall	Faculty Research Lab	Yes - lab bench
Dalton Hall 138	Kunsiri Grubbs Jennifer Schafer	Botany/Ecology Prep Room	Yes - fume hood
Dalton Hall 139	Cassie Bell	Prep room with microscopes	Yes - fume hood
Dalton Hall 205	Eric Birgbauer	Faculty Research Lab	Yes – fume hood
Dalton Hall 207	Eric Birgbauer	Faculty Research Lab	Yes - fume hood
Dalton Hall 209	Kathryn Kohl	Faculty Research Lab	Yes - fume hood
Dalton Hall 211	Vicky Frost	Faculty Research Lab	Yes - fume hood, cabinet under the fume hood
Dalton Hall 213		Faculty Research Lab	Yes - fume hood, lab bench
Dalton Hall 217	Julian Smith	Faculty Research Lab	Yes - fume hood, cabinet under the fume hood, lab bench
Dalton Hall 219	Jenny Schafer	Faculty Research Lab	Yes - fume hood
Dalton Hall 221		Faculty Research Lab	Yes - fume hood
Dalton Hall 229	Paula Mitchell/Kunsiri Grubbs	Museum/herbarium	Yes - fume hood
Dalton Hall 235	Eric Birgbauer Kathryn Kohl	Cell Biology /Genetics Prep area	Yes - fume hood
Dalton Hall 235A	Eric Birgbauer	Laminar air hood	Yes - fume hood
Dalton Hall 239	Kathryn Kohl	Genetics Laboratory	Yes - lab bench
Dalton Hall 240	Julian Smith	Microscopy suite	Yes - fume hood, cabinet under the fume hood, lab bench
Dalton Hall 236, 238, 241, 242 and 243	Julian Smith	Microscopy suite	Yes - fume hood, lab bench, other (specify) (cabinet under sink)
Dalton Hall 232	Vicky Frost	Microbiology Laboratory	Yes - flammable cabinet
Dalton Hall 334	Multiple instructors	Principles of Biology Laboratory Prep area	Yes - fume hood
Dalton Hall 336	Multiple instructors	Principles of Biology Laboratory	Yes - fume hood
Dalton Hall 335	Kiyoshi Sasaki	Anatomy and physiology/ Anthropology / Zoology Laboratory Prep area	Yes - fume hood

WINTHROP UNIVERSITY
Hazardous Waste Collection and Storage

This area has been designated a "**SATELLITE ACCUMULATION AREA**" for the collection and temporary storage of hazardous wastes. The designated area (bench top, fume hood, or cabinet) must be located near the point of generation and under the control of the principal investigator or laboratory supervisor. Flammable or combustible wastes should be stored in a flammables storage cabinet to meet fire code restrictions. Hazardous wastes, like other chemicals,

should not be stored on the floor unless secondary containment is used and the material is stored away from exits and/or egress pathways.

CONTAINERS: Hazardous waste containers must be compatible with the material being accumulated and must be closed at all times except when waste is being added or removed. Regulations do not permit funnels in waste containers except when material is being added. The maximum size allowable for collection containers is **4 liters** for hazardous wastes and **100 ml** for acutely hazardous wastes. Waste containers with aqueous contents must be stored in a secondary, leak-proof container able to hold 10% of the total volume of primary waste containers or 100% of the largest waste container contained inside it, whichever is greater. Waste containers with non-compatible constituents should be stored in separate secondary containers.

LABELING: All hazardous waste containers must be labeled at the time waste is first put into the container. Pre-printed, self-adhesive labels are available from the department Laboratory Manager (x6431) or Environmental Health and Safety (x2328) and should be used for all hazardous waste containers. **The following label information must be provided by the waste generator:**

1. Name and phone number of Principal Investigator/Laboratory Supervisor (waste generator):
2. University department, building and room number (use the lab room number, not your office):
3. Contents of the container, listing the names of all chemicals added to the container (use full chemical names only - abbreviations and chemical formulas are not acceptable); and
4. the percentage of each chemical if more than one is added to the container (if known);

When completing the pre-printed label, do not fill in the **accumulation date**; it will be added to the label when the container is moved to the central waste accumulation area (chemical storage building). Also, do not fill in the **EPA Hazardous Waste Code** unless you have been trained to do so, Environmental Health and Safety will add this information when the waste is moved to the central accumulation area.

SCHEDULING A WASTE PICK-UP: When a waste container is full or you have stopped generating a specific waste stream, contact either the department Laboratory Manager (x6431) or Environmental Health and Safety (x2328) and request a waste pick-up. **Waste should remain in the satellite accumulation area until moved by Environmental Health & Safety.**

IMPROPER DISPOSAL: Sinks and trash cans must never be used for hazardous waste disposal. Improper disposal of hazardous waste is a violation of federal, state and local laws. Please contact Environmental Health and Safety (x2392) to confirm which chemicals may be managed as non-hazardous waste.

EMERGENCY SPILL RESPONSE: In the event of a major spill (threat to public health, safety or the environment) or other emergency contact Campus Police at x3333. Fire extinguishers are located in hallways and in most laboratories; fire alarm pull stations are located at the end of each hallway and should be used as necessary. The Rock Hill Fire Department can be reached through

Campus Police at x3333 or by calling 9-911. Each satellite accumulation area should have appropriate spill kits readily available and minor spills should be cleaned up immediately by laboratory personnel. If spilled material is hazardous, all spill debris must be containerized and labeled as hazardous waste.

WASTE MINIMIZATION: Federal and state laws require waste generators to incorporate waste minimization concepts (changing procedures, reducing scale, substituting materials, etc.) into the processes which generate hazardous waste. Waste minimization lessens exposure to hazardous substances, protects the environment and reduces the cost of disposal which can drastically exceed the original cost of the chemical.

Appendix H

Biosafety Levels for Bacteria

Biosafety Levels for Bacteria

Biosafety levels for bacteria can be difficult to classify and somewhat confusing since the literature sources (e.g. laboratory manuals) often contradict each other. The American Type Culture Collection (ATCC) (www.ATCC.org) is the vendor most often used for purchase of mammalian and bacterial cells for culture. Some of the biosafety level designations for organisms commonly used in undergraduate teaching laboratories are listed below.

Species with only BSL-1 Preceptrol Strains	
Alcaligenes faecalis	*Halobacterium salinarum*
Bacillus cereus	*Micrococcus luteus*
Bacillus megaterium	*Moraxella catarrhalis*
Bacillus pumilus	*Neisseria perflava*
Bacillus subtilis	*Neisseria sicca*
Citrobacter freundii	*Pseudomonas fluorescens*
Clostridium acetobutyicum	*Serratia marcescens*
Clostridium sporogenes	*Staphylococcus auricularis*
Clostridium tertium	*Staphylococcus capitis*
Corynebacterium pseudodiphtheriticum	*Staphylococcus saprophyticus*
Corynebacterium xerosis	*Staphylococcus xylosus*
Enterobacter cloacae	*Sporosarcina ureae*
Enterobacter sakazakii	*Vibrio fischeri*

Species with only BSL-2 Preceptrol strains
(Indicates organisms where BSL-1 non-Preceptrol strain(s) is available.)*
*Klebsiella pneumoniae**
Proteus mirabilis
Proteus vulgaris
Pseudomonas aeruginosa
*Salmonella—all species**
Shigella—all species
Staphylococcus aureus
*Yersinia—all species**

Species with both BSL-1 and BSL-2 Preceptrol strains
(BSL -1 strain numbers given in parentheses.)
Enterobacter aerogenes (13048, 35028, 49469, 35029, 51697)
Escherichia coli (60 entries at BSL-1; 10798 is K-12)
Staphylococcus epidermidis (12228, 14990, 35547)

Appendix I

Frequently Used Biohazardous Agents in Biology

Frequently Used Biohazardous Agents in the Department of Biology

This is a list of the biohazardous agents that are currently recognized as such at the Department of Biology. This list will be updated annually by the Biosafety Committee. It is the responsibility of the faculty to report the introduction of new agents in their research falling under these categories to the department Biosafety Committee.

BSL-1	BSL-2
Neonatal rat cardiac tubes PC3 human prostate adenocarcinoma cell line (ATCC CRL 1435) Du145 human prostate carcinoma cell lines (ATCC HTB 81) LnCap human prostate carcinoma cell lines (ATCC CRL 1740) HUVEC human umbilical vein endothelial cell line (ATCC CRL-1730) EAhy926 human endothelial-like immortalized cell-line (derived from the fusion of human umbilical vein endothelial (HUVEC) cells with the A549 human lung carcinoma cells. T98G human glioblastoma cell line (ATCC, CRL-1690) U-118MG human glioblastoma cell line (ATCC, HTB-15) U-87MG human glioblastoma cell line (ATCC, HTB-14) DF-1 chicken embryo fibroblast cell line B103 rat neuroblastoma cell line *Agrobacterium tumefaciens* *Mycobacterium smegmatis* *Enterobacter aerogenes* *Corynebacterium xerosis* *Bacillus thuringiensis* *Bacillus subtilis* *Bacillus polymyxa* *Bacillus megaterium* *Bacillus licheniformis* *Bacillus coagulans* *Bacillus cereus* *Geobacillus stearothermophilus* *Alcaligenes faecalis* *Enterococcus faecalis* *Escherichia coli** *Micrococcus luteus* *Microbacterium foliorum*	Normal human prostate cells Lentivirus particles HEK293T human “kidney” cell line *Escherichia coli** *Proteus mirabilis* *Proteus vulgaris* *Pseudomonas aeruginosa* *Staphylococcus aureus* *Shigella flexneri* *Salmonella typhinurium* *Sporosarcina ureae* *Streptococcus sanguis*

Micrococcus roseus *Pseudomonas fluorescens* *Serratia marcescens* *Sporosarcina ureae* *Staphylococcus epidermidis* *Halobacterium salinarum*	

**Please check the safety level of Escherichia coli before using it. There are different strains of E. coli and depending on the strain some are BSL-1 and some are BSL-2.*