XI. GENERAL SAFETY EQUIPMENT

Adequate safety equipment in good working order should be provided to prevent accidents and injury or provide for an appropriate response in the event accidents or injury occurs.

A. Fire Extinguishers

The University Fire Marshal is responsible for placement locations, inspection, and maintenance of all fire extinguishers on the Norman campus.

1. Annual training on fire extinguisher use is required for all OU employees.

2. Every fire extinguisher should be clearly labeled to indicate the type of fire it is capable of fighting.

    a. Class A - fires in ordinary combustible materials such as wood, cloth, paper, and rubber.

    b. Class B - fires in flammable liquids, gases, and greases.

    c. Class C - fires that involve energized electrical equipment where the electrical conductivity of the extinguishing medium is of importance; when electrical equipment is de-energized, extinguishers for class A or B fires may be safely used.

    d. Class D - fires in combustible metals such as magnesium, titanium, zirconium, sodium, and potassium.

    e. Class K - grease or kitchen fires.

3. Fire extinguishers locations should never be concealed from general view or blocked from access.

4. Extinguishers should be mounted so that the top is not more than 5 feet above the floor; not more than 3½ feet if its weight equals more than 40 lbs.

5. Extinguishers should be mounted so that the instructions face outward.

6. Employees should notify the University Fire Marshal in Norman of any discharged or not fully charged fire extinguisher, one with a pin pulled out, one that is obstructed from view, or one not hanging in its proper place.

B. Safety Showers

Safety showers are designed to flood the entire body in the event of a clothing fire or a major spill of corrosive liquid.

1. Safety showers which provide tepid water should be in accessible locations

that require no more than 10 seconds to reach from areas where hazardous chemicals or biological materials are stored or used.

2. Each shower should be identified with a highly visible sign. All employees should familiarize themselves with the location and proper operation of the nearest safety shower.

3. No obstruction should be placed within 16 inches from the center of the spray pattern.

4. When needed, the user should stand under the shower and activate it by pulling on the handle. The handle should not be spring activated and should remain on until the user pushes the handle back up to turn it off. The employee should remove that portion of the clothing affected in an appropriate manner which reduces potential contact while under the shower. The laboratory supervisor should be notified and subsequent medical care for the employee should be obtained immediately.

5. Safety showers will be tested annually by the EHSO for proper operation.

Source: American National Standards Institute (ANSI) American National Standard for Emergency Eyewash and Shower Equipment, ANSI Z358.1-2001

C. Eyewash Fountains

If an employee receives a chemical splash to his/her eyes, eyewash fountains should be used for immediate and thorough washing of the eyes.

1. Permanently-plumbed or self-contained eyewashes capable of delivering tepid potable water to both eyes simultaneously at a rate of not less than 0.4 gallons per minute for 15 minutes should be available in accessible locations that require no more than 10 seconds to reach from areas where hazardous chemicals or biological materials are stored or used.

2. Each eyewash location should be identified with a highly visible sign. All employees should familiarize themselves with the location and proper operation of the nearest eyewash fountain.

3. When needed, the user should flood the eyes for **at least 15 minutes**, holding the eyelids open, and rolling the eyeballs so flushing fluid will flow on all surfaces of the eye and under the lid to be sure there is no residue of the chemical. The laboratory supervisor should be notified and subsequent medical care for the employee should be obtained immediately.

4. Permanently-plumbed eyewashes should be flushed weekly by laboratory personnel to assure proper operation and to minimize bacterial contamination from stagnant water. Self-contained eyewashes should be inspected and maintained in accordance with manufacturer's instructions, and should have clean, potable water available at all times when in use.

5. Eyewashes will be tested annually for proper operation by the EHSO.

Source: American National Standards Institute (ANSI) American National Standard for Emergency Eyewash and Shower Equipment, ANSI Z358.1-2001
OSHA Medical Services and First Aid Standard, 29 CFR 1910.151

D. First Aid Kits

1. First aid kits should be located in conspicuous places in the laboratory for response to **minor** injuries. For any injury which cannot be treated by first aid treatment, or is obviously serious, employees should obtain medical treatment as soon as possible.

2. The location and phone number of emergency services and to the Oklahoma Poison Control Center (405/271-5062, or state-wide 1-800-222-1222) should be clearly posted.

3. A designated party should be responsible for monitoring and maintaining the first aid kit(s).

4. First aid kit contents should include items such as Band-aids®, sterile gauze pads, scissors, antiseptic wipes or ointments. All kits should also contain examination gloves and a mask/face shield for response to emergencies in which blood is present. Pocket masks for CPR procedures are also recommended.

5. The following items are **not** recommended for use in a first-aid kit:

   a. Iodine - it is not a good antiseptic and can damage tissues.

   b. Ice Pack Compress - an ice pack compress may not be appropriate for chemical or thermal burns as its use could mask important diagnostic signs and symptoms of the injury; if there is swelling of soft tissue or other need for an ice pack, the person should be examined by a physician.

   c. Ammonia Inhalants - if an individual is unconscious, obtain help -- **do not use ammonia**.

   d. Tourniquet - not required for minor injuries; use the direct pressure technique until medical assistance is available.

6. Laboratories where high voltage equipment is in use should have available an emergency electrical response board. This will contain a non-conductive stick to turn off the equipment and remove the shock victim from contact with the source and an instruction card.

E. Refrigerators/Freezers

1. Each refrigerator, freezer, or cooler should be prominently marked to

indicate whether or not it meets the requirements for safe storage of flammable liquids.

2. Storage of limited quantities of flammable substances with a minimal risk of an explosive atmosphere outside the refrigerator/freezer should occur in a flammable materials storage refrigerator which is designed with spark-proof light bulbs, switches, fans, and motor assemblies.

3. For storage of large quantities of flammable substances or where some risk of a flammable atmosphere outside the refrigerator/freezer, an explosion-proof refrigerator/freezer is recommended. This type of equipment is designed to be spark-proof on both the inside and outside of the refrigerator/freezer.

F. Ventilation Hoods

1. Chemical Laboratory Hoods

Work that involves materials which are carcinogenic, suspect carcinogenic, toxic, odiferous, or volatile; or operations which may generate air contaminants at or above the appropriate OSHA PEL or ACGIH TLV **must** be conducted in a laboratory hood. The EHSO may be consulted to assist in determining whether exposure levels are being exceeded and hood use is required.

When working in a chemical laboratory hood, the following protocol must be used.

a. The face velocity on chemical fume hoods should be tested annually. Hoods used with radioactive materials will be tested semi-annually.

b. All sashes should be fully closed when not in use or not being attended.

c. Some hood fans may be linked to the building fire alarm system and may shut off when the alarm is activated. When a fire alarm sounds, close any open containers, shut the hood sash, and evacuate the building.

d. Raise the sash only as high as is necessary to perform the required operation. Never lift the sash above stops or arrows marked on the hood.

e. Work should be performed at least 6" back into the hood. A strip on the bench surface may be used as a reminder.

f. Do not put your head into the hood when contaminants are being generated.

g. Do not use the hood as a means of disposing of toxic or irritating chemicals. If vaporization of large quantities of such materials is a

necessary part of the operation, a means of collecting the vapor by distillation or scrubbing may be required. The collected liquid must then be disposed as of a liquid waste.

h. Do not remove sash or panels, except when necessary for apparatus setup or removal.

i. Do not adjust the rear baffle. The rear baffle has been adjusted at the top and base to provide efficient air flow. If ventilation problems develop, contact Physical Plant immediately.

j. Do not place electrical receptacles or other spark producing sources inside the hood when flammable materials are present.

k. Do not use perchloric acid in a standard chemical fume hood. Perchloric acid has a tendency to react violently with organic matter, causing fire or explosion, and should never be evaporated in a hood other than the stainless steel type. Stainless steel hoods have a wash-down feature which should be used periodically, approximately every two weeks. Date of wash-down should be recorded by the laboratory.

   Conversely, organic materials should never be vaporized in a perchloric acid hood because they may condense in the hood's exhaust and react with perchloric acid fumes.

l. Minimize foot traffic in the lab. Avoid creation of strong cross drafts caused by open doors and windows, air conditioning/ heating vents, or personnel movement. Drafts may pull contaminants from the hood into the laboratory.

m. Always look to assure fan motor power switch is in the "on" position before initiating experiment. Note: Some hoods do not have individual "on/off" switches and remain "on" continually. Make sure you know which kind you have.

n. Do not use infectious material in a chemical fume hood unless the hood has been designed with a special High Efficiency Particulate Air (HEPA) filter.

o. Do not use radioactive materials in a hood without prior approval from the OU Radiation Safety Office.

p. Exhaust fans should be spark-proof if exhausting flammable vapors and corrosive resistant if handling corrosive fumes.

q. Only materials being used in an ongoing experiment should be stored in the fume hood. Excessive storage of equipment and reagents will create air flow disturbances and reduce the

effectiveness of the operation of the hood.

r. When it is necessary to keep a large apparatus inside a hood, it should be placed upon blocks or legs to allow air flow underneath. Heating devices should be placed at the rear of the hood.

s. Do not make quick motions into or out of the hood, or create cross drafts by walking rapidly past the hood.

2. Biological Safety Cabinets

Class I and II biological safety cabinets, when used in conjunction with good microbiological techniques, provide an effective partial containment system for safe manipulation of moderate and high-risk microorganisms. As with any other piece of laboratory equipment, equipment must be functioning properly and personnel must be trained in the proper use of the biological safety cabinets. See Section VIII., "Biological Safety Cabinets" for additional information.

3. Specialized Local Ventilation

Some instruments such as atomic absorption spectrophotometers or inductively coupled argon spectrometers emit small quantities of hazardous materials during use. To prevent excessive accumulations of these materials, each of these instruments should be provided with an individual ventilation duct placed directly over the exhaust of the instrument.

G. Extraction Equipment

Extractions can present a hazard because of the potential buildup of pressure from a volatile solvent and an immiscible aqueous phase. Glass separatory funnels used in laboratory operations are particularly susceptible to problems because their stoppers or stopcocks can be forced out resulting in a spill of the contained liquid. It is even possible for pressure to burst the vessel. The following is an accepted procedure for accomplishing such a separation.

1. Do not attempt to extract a solution until it is cooler than the boiling point of the extractant.

2. When a volatile solvent is used, the unstopped separatory funnel should first be swirled to allow some solvent to vaporize and expel some air. Close the funnel and invert it with the stopper held in place and immediately open the stopcock to release more air plus vapor. This should be done with the stopcock handle and funnel drain turned away from you and others with the hand encompassing the barrel to keep the stopcock plus securely seated. (Note: Glass stopcocks should be lubricated.)

3. Do not vent the separatory funnel near a flame or other ignition source. Then close the stopcock, shake with a swirl and immediately open the stopcock with the funnel in the inverted position to again vent the vapors.

4. If it is necessary to use a separatory funnel larger than one liter for an extraction with a volatile solvent, the force of the stopper may be too great and cause the stopper to be expelled.

5. Consider performing the extraction in several smaller batches.

Source: *Chemical Safety Manual for Small Businesses*, Second Edition, American Chemical Society, 1992

H. Distillation Equipment

Distillation procedures include potential dangers associated with pressure build up, the common use of flammable materials and the necessity for heat to vaporize the chemicals involved. Careful design and construction of the distillation system is required to accomplish effective separation and to avoid leaks that can lead to fires or contamination of the work area.

1. Smooth boiling is important during the separation process to avoid bumping which can blow apart the distillation apparatus. Stirring the distillation mixture is the best method to avoid bumping. The use of boiling chips is also effective for distillations carried out at atmospheric pressure.

2. The source of heat is an important factor in the distillation process. Even heating may be accomplished by using an electric mantle heater, a ceramic cavity heater, steam coils, or a non-flammable liquid bath. Silicon oil or another suitable high-boiling oil can be used if heated on a hot plate. Hot water or steam may be used where practical. An additional thermometer may be inserted very near the center bottom on the distilling flask to warn of danger, exothermic decomposition. Always avoid heating above the temperature directed in the procedure.

3. Super-heating and sudden boiling (bumping) frequently occur when distilling using reduced pressure. Therefore, the assembly should be secure and the heat should be distributed as evenly as is possible. Evacuate the assembly gradually to minimize the possibility of "bumping." A standing shield should be in place for protection in the event of an implosion. After finishing a reduced pressure distillation, cool the system before slowly bleeding in air, because air may induce an explosion in a hot system. Pure nitrogen may be preferred to air, and can be used even before cooling the system.

4. When carrying out a steam distillation, take care not to run the steam in at too great a rate for the condenser. Overfilling the flask is less likely if condensation from the entering steam line is tripped and if the flask is heated or insulated to prevent excessive condensation.

5. Organic compounds should never be distilled or evaporated to dryness unless they are known to be free of peroxides. Most ethers, including cyclic ethers, form dangerously explosive peroxides on exposure to air and light. Unsaturated hydrocarbons, potassium metal and other reagents can also form peroxides.

Source: *Chemical Safety Manual for Small Businesses*, Second Edition, American Chemical Society, 1992

I. Centrifuges

Centrifuges can present two serious hazards: mechanical failure and dispersion of aerosols. A mechanical failure, such as a broken drive shaft, a faulty bearing, or a disintegrator rotor can produce not only aerosols but also hazardous fragments moving a great velocity. These fragments, if they escape the protective bowl of the centrifuge, could produce traumatic injury to personnel.

Activities such as filling centrifuge tubes, removing cotton plugs and rubber caps from tubers after centrifugation, removing the supernatant and resuspending cells, are capable of releases hazardous or biological aerosols into the environment. The greatest hazard associated with centrifuging biohazardous materials is created when a centrifuge tube breaks. When tubes break or crack and a fluid containing microorganisms remains in the cup under centrifugal force, relatively few organisms are released into the air compared to breakage that releases the fluid into the centrifuge chamber.

The following are recommended procedures for working with centrifuges.

1. Before centrifuging, eliminate tubes with cracks and chipped rims, inspect the inside of the trunnion cup and correct rough walls caused by erosion or adhering matter, and carefully remove bits of glass and other debris from the rubber cushion.

2. A disinfectant may be added between the tub and trunnion cup to disinfect the materials in cause of accidental breakage. This practice also provides an excellent cushion against shocks that might otherwise break the tube. Care must be taken, however, not to contaminate the culture material with the disinfectant.

3. Avoid pouring the supernatant material from centrifuge tubes. If you must do so, wipe off the outer rim with a disinfectant afterwards; otherwise, in a subsequent stop, biohazardous fluid may be spun off as droplets that form an aerosol. Use of a vacuum system with appropriate in-line safety reservoirs and filters is preferable to pouring from centrifuge tubes or bottles.

4. If the sediment is packed infectious microorganism or other hazardous material and must be resuspended in order to minimize the amount of aerosol created, it is better to use a swirling, rotary motion rather than shaking. If vigorous shaking is essential to suspend the material or achieve homogeneity, a few minutes should elapse before opening the container to allow the aerosol to settle. A biological safety cabinet with gloves in place may be required to assure safety to the laboratory worker when performing some of these operations.

5. Avoid filling the centrifuge tube to the point that the rim, cap, or cotton plug becomes wet with culture.

6. Screw caps or caps that fit over the rim outside the centrifuge tube are safety than plug-in closers. Some fluid usually collects between a plug-in closure and the rim of the tube. Even screw-capped bottles are not without risk, however; if the rim is soiled and sealed imperfectly, some fluid will escape down the outside of the tube.

7. Aluminum foil should not be used to cap centrifuge tubes containing toxic or infectious materials because these light-weight caps often become detached or ruptured during handling and centrifuging.

8. Care must be taken to ensure proper balance of the load each time the centrifuge is used. If a tabletop centrifuge is used, ensure that it is properly anchored.

9. Centrifugation at high speeds presents additional hazards because of the higher stresses and forces applied to components of the system. A HEPA filter should be placed between the centrifuge and the pump to prevent contamination of the pump.

Sources: *Laboratory Safety Monograph: A Supplement to the NIH Guidelines for Recombinant DNA Research*, NIH, January, 1979
Chemical Safety Manual for Small Businesses, Second Edition, American Chemical Society, 1992

J. Blenders, Mixers Sonicators and Cell Disruption Equipment

Hazardous aerosols may be created by most laboratory operations concerned with blending, mixing, stirring, grinding or disrupting materials such as cells, tissues, blood samples, freeze dried sera, and environmental samples that may contain infectious, toxic or otherwise hazardous materials. Even the use of mortar and pestle can be a hazardous operation. Ball mills, colloid mills, jet mills, tissue grinders, magnetic mixers, stirrers, sonic cleaning devices, ultrasonic cell disintegrators, and shakers are other devices that can produce hazardous aerosols. Safety laboratory practices for using such equipment are as follows:

1. Operate blending and cell disruption and grinding equipment in a biological safety cabinet.

2. Use safety blenders designed to prevent leakage from the rotor bearing at the bottom of the bowl. In the absence of a leak-proof rotor, inspect the rotor bearing at the bottom of the blender bowl for leakage prior to operation. Test it in a preliminary run with sterile water, saline or methylene blue solution prior to use.

3. If the blender is used with infectious material, use a towel moistened with disinfectant over the top of the blender. Sterilize the device and residual contents promptly after use.

4. Glass blender bowls are undesirable for use with infectious material because of potential breakage. If used, they should be covered with a propylene jar to prevent dispersal of glass in the event of the bowl breaks.

5. A heat sealed flexible disposable plastic film enclosure can be used for a grinder or blender. The safest practice is to use these within a biological safety cabinet. That means they are not used for total containment but rather to spare gross contamination of the cabinet when equipment or procedures are used that are known to release aerosols.

6. Blender bowls sometimes require supplemental cooling to prevent destruction of the bearings and to minimize thermal effects of the product.

7. Before opening the safety blender bowl, permit the blender to rest for at least one minute to allow settling of the aerosol cloud.

Sources: *Laboratory Safety Monograph: A Supplement to the NIH Guidelines for Recombinant DNA Research*, NIH, January, 1979
Chemical Safety Manual for Small Businesses, Second Edition, American Chemical Society, 1992

K. Water Baths

1. For either hot or cold water baths, use gloves and a face shield to immerse the objects into the bath.

2. Do not use sodium azide as a bactericide where contact with metal objects can occur. Sodium azide reacts with many metals to form explosive compounds.

Sources: *Laboratory Safety Monograph: A Supplement to the NIH Guidelines for Recombinant DNA Research*, NIH, January, 1979
Chemical Safety Manual for Small Businesses, Second Edition, American Chemical Society, 1992

L. Reduced Pressure Operations

1. Vacuum desiccators should be enclosed in a box or approved shielding device for protection in case of an implosion. Before opening a desiccator that is under reduced pressure, make sure that atmospheric pressure has been restored.

2. If water aspirators for reduced pressure are used for filtration purposes, only equipment that is approved for this purpose should be used. Never apply a vacuum to a flat-bottom flask unless it is in a heavy walled filter flask designed for this purpose. Place a trap and a check valve between the aspirator and the apparatus so that water cannot be sucked back into the system if the water pressure should fall unexpectedly while filtering. These recommendations also apply to rotary evaporation equipment where aspirators are being used for reduced pressure.

3. If vacuum pumps are used, a cold trap should be placed between the apparatus and the vacuum pump so that volatiles from a reaction or distillation do not get into the pump oil or out into the atmosphere of the laboratory. Exhausts from pumps should be vented to a hood. All pumps should also have a belt guard to prevent hands or loose clothing from being

caught in the belt pulley. Never apply reduced pressure to a flat-bottomed flask unless it has been designed for that purpose.

4. Shielding should be used whenever the apparatus is under reduced pressure.

Source: *Chemical Safety Manual for Small Businesses*, Second Edition, American Chemical Society, 1992

M. Glassware

1. Use only equipment that is free from flaws such as cracks, chips and other obvious defects.

2. Addition and separatory funnels should be freshly lubricated, properly supported, and oriented so that the stopcock will not be loosened by gravity. A retainer ring should be used on the stopcock plug.

3. Condensers should be properly supported with securely positioned clamps. The attached water hoses should be securely attached.

4. Apparatus attached to a ring stand should be placed so that the center of gravity of the system is over the base and not to one side but with adequate provision for removing burners or baths quickly. Stands bearing heavy loads should be firmly attached to the bench top.

5. When working with flammable gases or liquids, do not allow burners, sparking motors or other ignition sources in the vicinity. Use appropriate traps, condensers, or scrubbers to minimize release of material to the environment.

6. Use a hood when conducting a reaction that could result in an explosion or when using a vacuum system (which may implode). Close the sash to provide a shield. If a hood is not available, use a standing shield. Shields that can be knocked over by an explosion must be stabilized with weights or fasteners. Standing shields are preferably secured near the top. Proper eye and face protection must be worn even when using the shields or hood.

7. Whenever possible, use controlled electrical heaters or steam in place of gas burners.

8. To cut glass tubing, use the following precautions.

    a. Hold the tubing against a firm support and make one quick firm stroke with a sharp triangular file or glass cutter, rocking the file to extend the deep nick one-third around the circumference.

    b. Cover the tubing with cloth and hold the tubing in both hands, away from your body with the nick centered between your hands and turned away from your body.

c. Place your thumbs on the tubing opposite the nick about an inch apart and extended toward each other.

d. Push out on the tubing with your thumbs as you pull the sections apart, but do not deliberately bend the glass with your hands.

e. Avoid accidental contact of the tubing with a nearby person by standing with your back toward a wall or the lab bench.

f. If the tubing does not readily pull apart, the nick is probably too shallow or rounded; make a fresh sharp file scratch in the same place and repeat the operation.

g. Be careful in cutting a short piece from a long piece of tubing, because the long end may whip and injure a nearby person.

9. All glass tubing and rods, including stirring rods, should be fire polished before use. Unpolished cut glass has a razor-like edge, which not only will lacerate the skin, but will also cut into a stopper or rubber hose, making it difficult to insert the glass properly. After polishing or bending glass, allow ample time for it to cool.

10. When drilling a stopper, use the following precautions.

a. Use only a sharp borer one size smaller than that which will just slip over the tube to be inserted.

b. For rubber stoppers, lubricate with water or glycerol.

c. Holes should be bored by slicing through the stopper, twisting with moderate forward pressure, grasping the stopper only with the fingers, and keeping the hand away from the back of the stopper.

d. Keep the index finger of the drilling hand against the barrel of the borer and close to the stopper to stop the borer when it breaks through.

e. Preferably, drill only part way through and then finish by drilling from the opposite side.

f. Discard a stopper if a hole is irregular or does not fit the inserted tube snugly, if it is cracked, or if it leaks.

11. When available, ground glassware is preferable. Glass stoppers and joints should be clean, dry, and lightly lubricated. Rubber or cork stoppers should fit so that one-third to one-half of the stopper is inserted into the joint. Corks should have been previously softened by rolling and kneading.

12. To insert glass tubes or rods into stoppers or flexible tubing, use the

following precautions.

a. Make sure the diameter of the tube or rod is compatible with the diameter of the hose or stopper.

b. Fire polish the end of the glass to be inserted.

c. Lubricate the glass. Water may be sufficient; glycerol is a good lubricant.

d. Wear leather gloves or wrap a cloth around the glass and protect the other hand by holding the hose or stopper with a cloth.

e. Hold the glass not more than 5 cm from the end to be inserted.

f. Insert the glass with a slight twisting motion, avoiding too much pressure and torque.

Source: *Chemical Safety Manual for Small Businesses*, Second Edition, American Chemical Society, 1992

N. Pipettes and Pipetting Aids

1. Never mouth pipette. Always use some type of pipetting aid.

2. If working with biohazardous or toxic fluids, pipetting operations should be confined to a biological safety cabinet or hood.

3. Pipettes used for the pipetting of biohazardous material or toxic materials should be plugged with cotton (even when safety pipetting aids are used.)

4. No biohazardous material should be prepared by bubbling expiratory air through a liquid with a pipette.

5. Biohazardous material should not be mixed by suction and expulsion through a pipette.

6. No biohazardous material should be forcibly expelled out of a pipette. Mark-to-mark pipettes are preferable to other types, since they do not require expulsion of the last drop.

7. When pipettes are used, avoid accidentally dropping infectious cultures from the pipette. Place the disinfectant-soaked towel on the working surface and autoclave the towel after use.

8. Discharge from pipettes should be as close as possible to the fluid or agar level, or the contents should be allowed to run down the wall of the tube or bottle whenever possible, not dropped from height.

9. Contaminated pipettes should be placed horizontally in a pan containing

enough suitable disinfectant to allow complete immersion. They should not be placed vertically in a cylinder.

10. Discard pans for used pipettes should be housed within a biological safety cabinet. The pan and pipettes should be autoclaved as a unit. The replacement unit should be a clean pan with fresh disinfectant.

Source: *Laboratory Safety Monograph: A Supplement to the NIH Guidelines for Recombinant DNA Research*, NIH, January, 1979