

DEPARTMENT OF BIOLOGY LABORATORY INFORMATION, SAFETY AND POLICY MANUAL

COMPILED BY

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PREFACE

This manual is not intended to be an exhaustive treatise, but a set of guidelines that will help assure the safety and security of personnel and students in laboratories in the Department of Biology. **Additional literature concerning the handling and disposal of chemicals may be obtained from Dr. Bob Atkins of the Department of Chemistry.**

GENERAL LABORATORY INSTRUCTIONS FOR STUDENTS

The following instructions are to be considered guidelines, not a comprehensive set of rules. In many cases you will need to rely on your own judgment. Therefore, use common sense and proceed with caution, or ask the instructor for help.

I. Dirty Labware

A. Disposable - Make sure an item is disposable before discarding it. Disposable items may include test tubes, pipettes, and beakers. Flasks, bottles, and graduated cylinders are generally not disposable. Not everything that is plastic is disposable, but many glass items are. A mistake could be costly. For example, a tissue grinder (homogenizer) may cost more than \$75.00. If in doubt, ask the instructor.

B. Non-disposable Labware (Routine) — The following is a minimum treatment. For special glassware consult the instructor.

1. Place dirty labware in a dishpan containing a detergent solution until you are ready to wash them. Do not allow dishes to accumulate where they can dry out. Soaking the labware immediately after use prevents proteins, etc from drying and facilitates later cleaning. If, however, you leave glassware in the detergent too long, the alkaline detergent will etch the glass and ruin it.
2. Scrub all labware with brushes in hot soapy water using detergent such as Tetrox, Alkanox, or Liquinox.
3. Rinse with hot tap water until all traces of soap are gone (at least 3 times) and twice with deionized water. Use the deionized water sparingly, and do not allow this water to run unless you are actually using it.
4. When working with nucleic acids, place all glassware in an oven to dry at 250° C for at least 2 hours (to destroy nucleases). Place all plasticware on racks, in baskets, or invert to dry. For regular laboratory glassware, dry as described for the plasticware.

DO NOT PLACE PLASTIC IN THE OVEN.

5. When dry, invert beakers on the storage shelf and lightly stopper flasks, bottles, graduated cylinders, etc with a plug of cotton. Save the cotton when you use the item. Alternatively, seal the opening with parafilm or plastic wrap. Store small flasks (up to 50 mL) and beakers (up to 100 mL) unstoppered in the drawer provided.
6. Remove plungers from glass syringes and homogenizers immediately after use until they are clean and DRY. Do not place Teflon homogenizer pestles in the oven.

C. Pipettes

4. Soak dirty pipettes in a jar containing detergent solution immediately after use (tips up). Check to see that the detergent covers the tips.
5. Rinse 3 times with tap water in the pipette washer, soak at least 2 hours in acid dichromate solution (discard the solution when it turns green), rinse exhaustively (perhaps overnight) with tap water, rinse once with deionized water, transfer to

pipette dryer (tips down) and dry for 2 hours. Store in the proper place. Avoid handling the tips. If you plan to leave the pipette washer unattended, make sure the outlet hose is properly placed and that the sink is not clogged. If you are cleaning plastic pipettes, do not use acid dichromate.

- D. Frequently Used Items (magnetic spin bars, spatulas, and stirring rods). Soak these items in a detergent solution in a separate pan from the routine labware. These items are used so frequently that they might have to be washed before the other labware; thus, they will be more accessible if stored separately. Also do not leave spin bars in solutions after they have been prepared, and be especially careful that they are not poured down the drain.

II. Solutions

All solutions, reagents, or other materials to be stored should be properly covered and labeled with the following information: (1) contents, including pH where appropriate; (2) date; and (3) initials of owner. Note: Beakers sealed with parafilm are not properly covered.

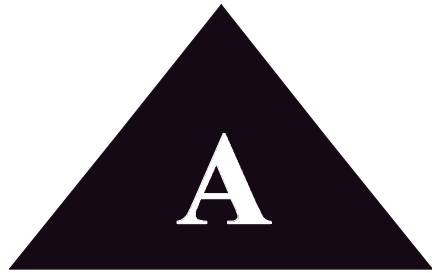
III. pH Meters and pH Adjustment.

- A. When a pH meter is not to be used for more than a few minutes, immerse the electrodes in pH 4 buffer. Check every few days to see that the water has not evaporated. Keep the meter in the standby mode when the electrodes are not immersed in solution.
- B. Check the reference electrode periodically to determine whether it needs additional saturated KCl solution. Pinch the rubber ferrule at the fill hole to establish the liquid junction before each day's use.
- C. Any time the electrodes are moved from one solution to another, rinse with deionized water (squeeze bottles work nicely), and wipe with a kimwipe.
- D. The meter should be calibrated with a buffer of known pH daily before use.
- E. When adjusting a buffer to the proper pH, be sure not to add all the required volume of water to your buffer chemicals. Leave enough room for the acid or base, which you will be adding. If additional water is needed after adjustment, it can be added later without affecting pH. Remember to allow for the volume of the stir bar.

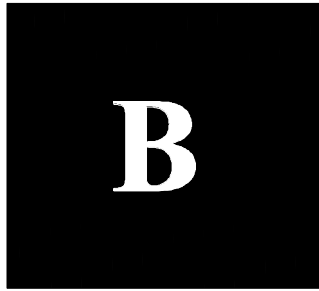
IV. General Safety Rules

1. Working Alone: Working alone with dangerous (e.g. flammable, toxic, or caustic) chemicals or procedures is strictly forbidden in the laboratory.
2. Eye Protection: Safety glasses, goggles, or face shields are required at all times in the laboratory. The nature of the work has no bearing on this rule. Contact lenses should be avoided since a splash to the eye may be trapped under the contact lens. Discuss the ramifications with your physician.
3. Apparel:
 - a) Lab coats, or jackets are strongly recommended to protect your clothing.
 - b) Shoes are required at all times. Sandals are discouraged since they provide no protection from spills.

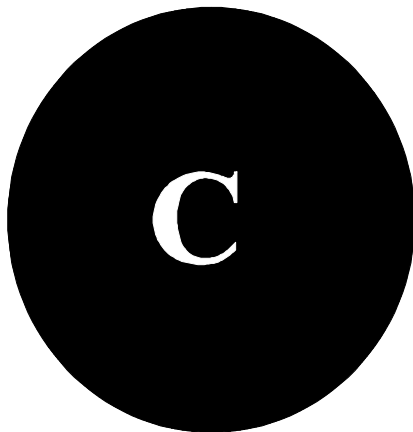
- c) Gloves should be worn when working with hazardous chemicals or glassware at extreme temperatures. Many different types of gloves are available; Use the appropriate gloves for the work you are doing. For instance:
- Latex gloves- Provide excellent dexterity and protection against most aqueous acid and base solutions but should not be used for protection against organic solvents.
 - Butyl rubber gloves- provide ideal protection from ketones, ethers, esters, aldehydes, acetates, alcohols and most inorganic acids or caustics. They are not recommended for hydrocarbons, aromatics or chlorinated solvents.
 - Nitrile rubber gloves- provides good protection from oils, fats, acids, caustics, and alcohols.
 - Viton gloves- provide excellent protection from chlorinated solvents, aromatic solvents, hydrocarbons, most alcohols (except methanol), concentrated inorganic acids and caustics. Not recommended for use with acetates, aldehydes, amides, amines, esters, ethers or ketones.
- d) Dust masks and respirators should be worn when needed. Remember- dusk masks filter annoying and dangerous particles from the air; respirators protect against toxic, corrosive, or poisonous gases.
4. Eating, Drinking, Smoking: The consumption of food or drink in the lab is strictly forbidden. Most chemicals are toxic to some extent and the contamination of the food, drink, containers and your hands with toxic or hazardous chemicals is very possible. Smoking is also forbidden in the laboratory since it can be an overlooked source of ignition. Also the tobacco can become contaminated with chemicals.
5. Emergency equipment:
- a) Eye wash fountains or bottles are available in all laboratories. Know their location and their proper uses. Treat splashes promptly.
 - b) Safety showers are located in the laboratory. If your clothing is on fire or soaked with a corrosive chemical, get under the shower. Safety showers usually have pull chains or bars that will release copious amounts of water. Remove any clothing that was soaked by a chemical immediately. A loss of skin is much more severe than a loss of modesty.
 - c) Fire blankets are located in most laboratories. Know where they are located and how to use them. A fire blanket is a noncombustible blanket on a roller. The blanket is kept in a quick release cabinet, which will open easily and rapidly with a pull on the handle. If your clothing catches on fire, wrap your body with the blanket and drop to the floor to smother the fire.
 - d) First Aid Kits are located in the laboratories. Know where the kits are located and what they contain. Report any cut or injury to your instructor. Prompt attention will minimize any chances of infection or chemical injury.
 - e) Fire extinguishers are located in all laboratories. You must know their location, type and how to use them. **MAKE SURE YOU USE THE CORRECT TYPE OF FIRE EXTINGUISHER BECAUSE EXPLOSION MAY RESULT IF THE WRONG EXTINGUISHER IS USED ON A FIRE!** The following symbols are used to label fire extinguishers and to indicate their applicability for particular types of fires.



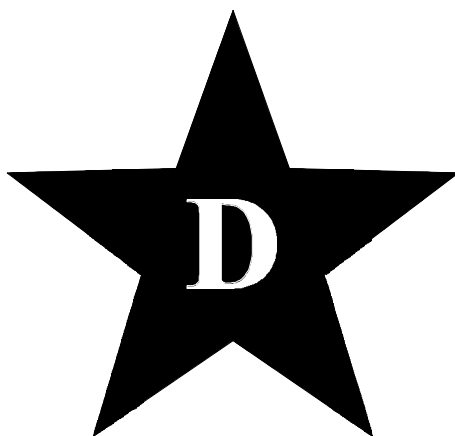
This symbol indicates the extinguisher is applicable for use on ordinary combustibles such as wood, cloth, paper, rubber, etc. The background of the symbol will be either metallic or green. It will always be found on water, multi-purpose dry chemical, carbon dioxide, and foam type extinguishers.



This symbol indicates the extinguisher is applicable for use on flammable or combustible liquids, flammable gases, greases, oils, etc. The background of the symbol will be either metallic or red. It will always be found on multi-purpose dry chemical, carbon dioxide, and foam-type extinguishers.



This symbol indicates that the extinguisher is applicable for use on fires involving energized electrical equipment. The background of the symbol will be either metallic or blue. It will always be found on multi-purpose dry chemical and carbon dioxide extinguishers.



This symbol indicates the extinguisher is applicable for use on certain combustible metals such as sodium, magnesium, potassium, etc. The background of the symbol will be either metallic or yellow. It will always be found on special dry powder extinguishers. If the lab does not have a type D extinguisher for alkali metal fires (i.e., sodium or potassium), soda ash or a sand/soda ash mix can be used for this type of fire.

6. Emergency Assistance: Know where the nearest phone is located and be sure emergency numbers for the Fire Department, Police, Rescue Squad, and other emergency personnel are posted nearby.
7. Transporting Chemicals: Never carry more chemicals than you can safely handle. Hold bottles securely from the top and bottom. Avoid carrying incompatible chemicals at the same time. If you stumble and fall, try to drop the bottles away from you and others. Chemicals transported between floors should be carried on carts up an elevator or on a dumbwaiter; never by hand on the stairs.
8. Read the Label: When preparing a solution, first read the label. Follow label instructions exactly! If no cautions are listed, do not assume that the chemical is safe. Check the Merck Index, Handbook of Toxic Substances, ACS Handbook of Laboratory Safety, or other reference that indicates the hazards involved in using that chemical. Not only is it important to have the correct purity, but waters of hydration, etc. need to be taken into consideration when making solutions. The following (Fig. 1) is a typical label from a reagent bottle.

Meets ACS Specifications
Certificate of Lot Analysis

KClO₄
LOT 7053 KBPH F.W. 138.55

Mallinckrodt

**POTASSIUM
PERCHLORATE**
(CRYSTALS) KClO₄

Calcium (Ca)..... <0.005%	Iron (Fe)..... <0.0005%
Chloride (Cl)..... <0.0003%	Sodium (Na)... <0.02%
Heavy Metals (as Pb).. <0.0005%	Sulfate (SO ₄).. <0.001%
Insoluble Matter..... <0.005%	

AR[®] Analytical Reagent


It is hereby certified that the above is a true copy of the actual analysis of the lot indicated.

Mallinckrodt, Inc.
Paris, Kentucky 40361
8/2 Printed in U.S.A.
1286243-1 M

C.E. Elmore
Quality Control

Danger! Strong Oxidizer. Contact with other material may cause fire. May be harmful if swallowed or inhaled. Causes irritation.

LabGuard™
Health Hazard: Moderate
Flammability: None
Reactivity: Severe
Contact Hazard: Moderate


Oxidizer

Protective Equipment:
Protective Eyewear
Hand Protection
Safety Clothing
Lab Hood

Storage Code: Yellow

7053 500 GM. (1.1 Lb)

CAS 7778-74-7
Proper Shipping Name -
POTASSIUM PERCHLORATE, UN1489

FIRST AID: If swallowed induce vomiting immediately by giving two glasses of water or milk if available and sticking finger down throat. Never give anything by mouth to an unconscious person. If inhaled, remove to fresh air. If not breathing, give artificial respiration. If breathing is difficult give oxygen. In case of contact, immediately flush skin or eyes with plenty of water for at least 15 minutes. **In all cases CALL A PHYSICIAN.**

EPA: HWDC - Ignitable
* See LabGuard Guide and Manual Safety Data Sheet for additional information.



NFPA

Figure 1. Typical Reagent Label.

9. Label Chemicals and Solutions: Label all chemicals and solutions carefully and completely. The label should include: the chemical name; the concentration or percent purity; the date prepared or transferred; any special instructions or warnings; and your initials (See example Fig. 2).

12 M H₂SO₄
Avoid Skin Contact
5-23-83
WKW

Figure 2. Example of a Proper Label.

Never take a reagent bottle and just tape another label over the existing one. This is very dangerous because the label you add may be missed or peeled off. Just scribbling over an existing label and writing on it is also a ridiculous but fairly common practice. If you need to use a reagent bottle that has a label on it, first completely remove the existing label. Be sure the bottle is clean and relabel it carefully as described above.

CONTENTS OF UNLABELED CONTAINERS WILL BE DISPOSED OF WITHOUT WARNING.

10. Spills: If you spill a chemical on yourself, wash immediately with copious amounts of water (See Chemical Accidents section). Clean up spills immediately. Clean the area of the spill as completely as possible. Refer to the section on Chemical Spills for details.
11. Broken Glass: Do not be alarmed when you break something. Carefully pick up all the pieces and place them in a "broken glass" container. Do not rush and cut yourself. A cut from glass coated with a chemical may give a direct injection of that chemical. If you do cut yourself, do not panic. Any bleeding will help wash out any chemical that may have been on the glassware. Except when the loss of blood is life threatening, allow some bleeding to occur. Notify your instructor and get medical help.
12. Hoods: Work in a hood when using noxious or highly combustible chemicals. Make sure that air exhaust is working properly before using the hood. It is extremely important to keep the hoods neat and clean. Return all equipment and chemicals to their proper storage. Do not clutter the hood by using it as a storage location for chemicals and do not work in a hood that has chemicals stored in it.
13. Water Faucets: Leaking faucets not only wastewater, but they also may overflow sinks and flood floors. Wet floors are slippery and can cause electrical shorts in floor outlets. Please report any of these problems to your instructor.
14. Compressed Gas Tanks: Compressed gas tanks are stored in steel and aluminum cylinders at high pressures. The following rules should be followed when using compressed gases.
 - a) The cylinder should be capped and transported on a hand truck. Leave the cap on until the cylinder is put in use. Always remove the regulator before you transport a gas cylinder.
 - b) The tank should be securely clamped the bench with an appropriate cylinder clamp or chains.
 - c) Store the tanks away from direct sunlight and flammable materials. Do not warm the tanks above room temperature.
 - d) Never open the main valve on a tank without the regulator attached. Learn how to properly use gas regulators.

- e) Open main tank valves gently and be alert for leaks. It is a good practice to only slightly open the main valve on a compressed gas tank so that it may be quickly closed in an emergency.
15. Gas (Propane) Jets: Leaking gas jets are an explosive hazard. Know where the gas main is and how to shut off the gas at the main valve and at each line to the gas jets.
 16. Vacuum Systems: Always wear safety glasses when near vacuum systems. Vacuum desiccators and other vacuum vessels should be taped or enclosed in shields at all times. Vacuum systems frequently have mercury gages and manometers attached to them. Use appropriate precautions when near these systems. Never evacuate a scratched or cracked flask as it may implode. Avoid physical jolts to any glassware under vacuum. Vacuum pumps should have exhaust filters or be properly vented.
 17. Electrical: Make sure that any electrical equipment you use is properly grounded. Also make sure that all electrical cords are in acceptable condition and that the work area around electrical equipment is kept dry. If the equipment is used for heating, make sure the area is free of flammable liquids or vapors. Turn all equipment off before you plug it in. Treat all electricity with respect.
 18. Use a bulb for pipeting dangerous or volatile chemicals, such as acrylamide or toluene. When in doubt, use a bulb.
 19. If you must boil a solution, put it in a boiling water bath. Do NOT heat a test tube over an open flame.
 20. Do NOT use sparks or flames when explosive fumes are present (e.g. ether fumes).
 21. Exercise extreme caution when working with acids or bases. If these agents come into contact with the body or clothes, flush the area (eyes included) for several minutes in running tap water and contact the instructor. When mixing acids and bases with each other or with water, use a heat-tempered glass container (e.g. Pyrex). Always add acid or base to water, not conversely.
 22. Do not attempt to repair equipment. Report any malfunction to the instructor immediately.
 23. Forethought: Think before acting. If you are uncertain about a particular procedure or act, seek assistance from a knowledgeable source. Beware of your coworker's activities. Most accidents in laboratories can be avoided if you are alert, careful, and use common sense.

Basic Radiation Safety Rules

1. Maintain extreme cleanliness and immaculate housekeeping in all radiation areas, especially where spreadable activity (unsealed sources) is present.
2. Prior to entering a radiation area where work with spreadable activity is involved, check body for any exposed breaks in the skin (i.e., open cuts or abrasions).
3. There will be no eating, chewing, smoking, drinking, or preparation of food or drink in any radiation area in which spreadable radioactive material is handled or stored (all radiation labs). Individuals wishing to smoke, chew, eat or drink will do so only outside of the radiation areas and only after monitoring, washing, and re-monitoring the hands.
4. Radioactive materials will be stored only in containers and areas designated and approved by this policy or the College Health Physicist (with concurrence of the Radiation Safety Committee).
5. Tools and other equipment used in radiation areas where spreadable radioactive material is present will be regarded as contaminated and will not be taken outside the area until a survey indicates the item is within the appropriate maximum permissible levels.
6. All new procedures employing radiation sources will be rehearsed as a "dry run" before work with radiation sources is begun, especially if special handling tools or equipment is used. Any faulty procedure or any defective or improperly designed handling equipment noted during such a rehearsal will be modified, repaired or replaced, and tested before being used in the actual operation with a radiation source.
7. Maximum practical distance and shielding, along with minimum personnel exposure time will be observed when handling or working in the vicinity of external sources of radiation. Unnecessary exposure to radiation is to be avoided.
8. No radioactive sources will be used in a non-radiation area in such a manner as to create radiation levels which, if an individual were continually present in the area, could result in his receiving a dose in excess of two millirem (2 mrem) in any one hour or one hundred millirem (100 mrem) in any seven consecutive days.
9. Entry into any area where there is a possibility a dose rate of 2 mrem in one hour, or a total dose in excess of 100 mrem in seven consecutive days (i.e., a restricted area, radiation area or high radiation area) will normally be limited to necessary work personnel, safety personnel, instructors, and students. Personnel entering into such areas will wear a film badge or other prescribed personnel dosimeter, unless part of a group touring the facility or unless deemed unnecessary by the Radiation Safety Committee. Entry into a high radiation area requires the wearing of self-reading dosimeters also. Normally visitors will not be allowed into high radiation areas. The Radiation Safety Committee may give permission. Tour groups will be escorted through all radiation facilities with the escort carrying a monitoring device for the

group when deemed necessary by the College Health Physicist (with concurrence of the Radiation Safety Committee). A survey of all radiation areas to be toured will be conducted by the departmental Senior Radiation Safety Person to insure compliance with pertinent regulations prior to allowing the tour group to enter any radiation area.

10. Experiments or tests will not be conducted without prior approval of the Radiation Safety Committee.
11. Locally fabricated laboratory sources will contain less than 1.0 microcuries of radioactive material. The Radiation Safety Committee must approve any deviation.
12. No radioactive materials shall be released or discharged to environmental air in average concentrations exceeding the values established in paragraph 20.03 or Appendix B, Title 10, CFR, Part 20, as measured at the final point of effective control. Approval of College Health Physicist (with concurrence of the Radiation Safety Committee) is required for all environmental air discharge.
13. Radioactive materials will not be released into any environmental water system except by the College Health Physicist or with the concurrence of the Radiation Safety Committee.

INSTRUCTIONS TO PERSONNEL WORKING WITH RADIOACTIVE MATERIAL

1. Individuals working in or frequenting any portion of a restricted area, radiation area or high radiation area will be informed of the presence of radioactive materials or of radiation. They will be instructed in the hazards associated with exposure to such materials, or radiations, in the precautions or procedures to minimize exposure, and on the pertinent regulations, which apply to that area.
2. Prescribed monitoring equipment (film badges and pocket dosimeters) will be worn at all times in any radiation area unless otherwise authorized by the College Health Physicist or the Radiation Safety Committee.
3. Prescribed protective clothing will be worn at all times in radiation areas where spreadable radioactive material is present and will not be worn outside a radiation area until the clothing is determined to be free of detectable contamination.
4. Monitor hair, hands, face, protective clothing and equipment with appropriate instrumentation frequently during the work periods where spreadable contamination is present.
5. Prior to leaving the radiation area, monitor all exposed areas of the body and personal clothing with appropriate instrumentation.
6. In the event any exposed body areas and personal clothing give an indication of being contaminated, initiate appropriate decontamination procedures and contact the College Health Physicist immediately.

7. Even if no contamination is found, the hands will be monitored, washed, then re-monitored before leaving any of the preparation rooms.
8. If an accidental overexposure has occurred or is suspected, a physical examination will be accomplished, supplemented by subsequent examinations as recommended by competent medical authority.
9. No one under 18 years of age will be permitted to work with radioactive isotopes, nor will pregnant women, regardless of their ages.

SECURITY

Security is a very important and often overlooked aspect of work in any laboratory. Biology departments house expensive instrumentation and many potentially dangerous chemicals. As much as we would like to ignore it, there are those who would try to steal from the laboratories. The key to preventing thefts is to be observant and aware of what others are doing.

If you see a stranger milling around in the stockroom or the labs, ask to be of assistance. A chemical laboratory is no place for unescorted tours or sightseers. If you see anything being taken from the stockroom or the labs, please report it immediately. When leaving a lab or a prep room, be sure all the doors are locked. Never supply glassware, equipment, or chemicals to unauthorized persons. Always have such persons see your instructor or stockroom manager.

Often the theft of an item is not as important as the frightening situation created by a prankster with a chemical. Serious injury or death can be caused by those ignorant of the possible hazards.

Biological equipment must also be secured. Syringes, for instance, must be carefully regulated, and used syringes and needles should be destroyed.

Every laboratory has rules and regulations regarding security. It is your responsibility to become familiar with and enforce these regulations. Call security if you need help enforcing any of these regulations.

CHEMICAL ACCIDENTS

The number one priority of work in any laboratory should be safety so that accidents are avoided. However, in the event that an accident does occur, you must be prepared to act quickly to minimize the consequences. As long as you continue to work in a laboratory, there is a possibility that you will have to administer first aid to an individual who has had a chemical accident. The following instructions have been condensed primarily from two sources ("Hazards in the Chemical Laboratory" 2nd ed., G.D. Muir, Ed., The Chemical Society, London, 1977. "First Aid Manual for Chemical Accidents", M.J. Lefevre, Dowden, Hutchinson and Ross, Inc., Stroudsburg, PA, 1980.). It is strongly recommended that these manuals be kept on hand for complete instructions on how to treat specific chemical accidents. It is also strongly recommended that the following summary of standard treatments be memorized because in the event that they are needed there probably will not be time to look them up. The most important thing to remember is to remain calm and to respond as quickly as possible to the accident.

A. Golden Rule For Treating Chemical Accidents

Always treat the most urgent symptom first

1. Cessation of breathing
2. Eye injury
3. Skin contact
4. Shock

B. Summary of Standard Treatments

1. Splashes to the eyes
 - a. To prevent loss of vision, treatment must begin immediately!! Do not hesitate to use physical restraint if necessary to administer treatment. Flood the eye thoroughly with large quantities of gently flowing water either from an eyewash-fountain or from an eyewash-bottle and continue for at least ten minutes. An eyewash-fountain is preferred because both eyes can be washed at the same time.
 - b. Be sure that the water bathes the eyeball by gently opening the eyelids and keeping them apart until the treatment is complete.
 - c. All eye injuries from chemicals require medical advice. Arrange transport to the infirmary and supply information to accompany the casualty on the chemical responsible and a brief summary of the treatment already given.
2. Splashes to the skin
 - a. Flood the splashed area thoroughly with large quantities of cold running water and continue for at least 10 minutes, or until satisfied that no chemical residue remains on the skin. Avoid using hot water since it opens the pores. If the situation warrants, use the safety shower. Chemicals known to be insoluble in water can usually be removed with soap solution.
 - b. Remove all contaminated clothing, taking care not to come in contact with the chemical yourself.

- c. If the situation warrants it, arrange for transport to the infirmary. Provide information to accompany the casualty regarding the chemical responsible and a summary of the treatment already given.
3. Inhalation of gases
 - a. First insure your own safety then remove the casualty from the contaminated area.
 - b. Loosen clothing and administer oxygen if available.
 - c. If casualty is unconscious place in a face down position and watch for cessation of breathing.
 - d. If breathing has stopped apply artificial respiration. If gas inhaled is HCN, do not administer mouth-to-mouth artificial respiration.
 - e. If the emergency warrants it, take the individual to the infirmary and provide information on gas responsible and summary of first aid treatment given.
 4. Ingestion of chemicals
 - a. If the chemical has been confined to the mouth give large quantities of water as a mouthwash. Be sure that the wash is not swallowed.
 - b. If chemical ingested is a strong acid or caustic do not induce vomiting. Otherwise, induce vomiting and if chemical has not been identified save vomit for possible chemical identification.
 - c. Give copious quantities of water or milk to dilute the chemical in the stomach.
 - d. Arrange for transport to the infirmary. Provide information on the chemical ingested, the amount and the concentration if possible, and also a brief description of the treatment given.

CHEMICAL SPILLS

The following guidelines will help you to handle the inevitable chemical spills. As always, the important things to remember are to remain calm and clean up the spill as quickly and completely as possible, being sure that no chemical residue remains. It is strongly recommended that the following procedures be committed to memory. In the event that there is a spill, being able to clean it up as quickly as possible will help prevent the possibility of an injury.

For the purpose of spill clean up, the more common laboratory chemicals may be divided into six general categories: Inorganic Acids, Inorganic Bases, Inorganic Salts, Organic Solvents, Organic Solids and Mercury. There are exceptions to these guidelines. For instance, some inorganics react violently with water (i.e. sodium, phosphorus pentoxide, lithium aluminum hydride, etc.) and should thus be handled in a completely different manner. For spills of chemicals which do not fit the categories listed, more specific information may be obtained from "Hazards in the Chemical Laboratory" 2nd ed., G.D. Muir, Ed., The Chemical Society, London, 1977. It is always a good practice to ask yourself, "What will I do if this chemical is spilled?" Having asked this question before using a chemical will help prevent injuries and delays in clean up. Most laboratories are equipped with supplies to handle chemical spills. Spill towels and pillows are used to absorb acids, bases, or solvents and provide an efficient method for cleaning spills. You should know where these supplies are stored.

Note that mercury is included as a separate category because it is so common in laboratories, is so frequently spilled and is extremely difficult to clean up. Broken thermometers are the greatest source of spilled mercury in the lab so it is recommended that extra precautions be taken any time a thermometer is used.

Before disposing of any chemical that has been spilled please refer to the section on Chemical Disposal.

General Guide for Chemical Spills

A. Inorganic Acids

Examples:

- 1) Hydrochloric Acid (HCl)
- 2) Nitric Acid (HNO₃)
- 3) Sulfuric Acid (H₂SO₄)
- 4) Perchloric Acid (HClO₄)
- 5) Phosphoric Acid (H₃PO₄)

Special Precautions:

- 1) May cause severe burns
- 2) May produce or react to produce harmful vapors
- 3) Contact with combustible materials may cause fires

Spillage and Disposal:

- 1) Small Spill: Instruct others to keep a safe distance. Wear eye protection and gloves. Mop up the acid with the chemical spill towels. Dispose of towels in plastic bags. Rinse area of spillage with water.

- 2) Large Spills: Instruct others to keep safe distance. Wear eye protection and gloves. If dealing with HCl or HNO₃, wear breathing apparatus. Spread sodium carbonate or sodium bicarbonate liberally over spillage and mop up cautiously with water. Run this to waste, diluting greatly with running water.

B. Inorganic Bases

Examples:

- 1) Sodium Hydroxide (NaOH)
- 2) Potassium Hydroxide (KOH)
- 3) Ammonium Hydroxide (NH₄OH)

Special Precautions:

- 1) May cause severe burns
- 2) NH₄OH - Harmful vapor

Spillage and Disposal

- 1) If Solid: Wear face shield or goggles and gloves. Scoop into enamel or polyethylene bucket. Dissolve by adding the solid, in small quantities, with stirring, to a large volume of water. After the solid is dissolved, neutralize and flush down the drain, diluting greatly with running water. Wash down site of spillage thoroughly with water.
- 2) If Solution: Wear face shield or goggles and gloves. Mop up cautiously with water using spill towels, pillows, or mop. Dispose of pillows and towels by placing in plastic bags. Run any solution to waste diluting greatly with water.

C. Inorganic Salts

Examples:

- 1) Sodium Chloride (NaCl)
- 2) Potassium Phosphate (K₃PO₄)
- 3) Sodium Nitrate (NaNO₃)
- 4) Ammonium Sulfate (NH₄)₂SO₄

Special Precautions:

- 1) May cause severe burns
- 2) Dust from solid spills may be harmful
- 3) May be extremely reactive with combustible material

Spillage and Disposal

- 1) If Solid: Wear face shield or goggles and gloves. If the material is dusty, wear respirator or dust mask. Scoop material into a plastic bucket and dissolve in water. Dispose of down the drain diluting with copious quantities of water.
- 2) If Solution: Wear face shield or goggles and gloves. Mop up with spill towels, pillows or mop. Dispose of towels and pillows by placing them in plastic bags. Wash spill area thoroughly and run waste to drain followed by large volumes of water.

D. Organic Solvents

Examples:

- 1) Acetone
- 2) Petroleum Ether
- 3) Chloroform
- 4) Ethanol
- 5) Toluene

Special Precautions

- 1) May be highly flammable
- 2) Vapors harmful

Spillage and Disposal

- 1) Small Spills: Instruct others to keep a safe distance. Wear organic resistant gloves and eye protection. Shut off all possible sources of ignition. The solvent should be mopped up with paper towels and put into a hood to evaporate.
- 2) Large Spills: Instruct others to keep a safe distance. Shut off all sources of ignition. Wear eye protection, gloves, and breathing apparatus. Absorb solvent on sand, shovel into buckets and transport to hood to evaporate.

E. Organic Solids

Examples:

- 1) Benzoic Acid
- 2) Naphthalene
- 3) Hydroquinone
- 4) 2,4-Dinitrophenyl hydrazine

Special Precautions

- 1) May be explosively reactive
- 2) Dust or vapor from solid may be harmful
- 3) Flammable solids

Spillage and Disposal

- 1) Instruct others to keep a safe distance. Wear organic resistant gloves, eye protection and organic vapor mask. Scoop solid into a beaker or plastic bucket. Transfer to a glass jar and label for future disposal. Wash down the site of the spill thoroughly with water and flush down the drain.

F. Mercury

Special Precautions

- 1) Gives off very poisonous vapor
- 2) Danger of cumulative effect
- 3) Extremely difficult to clean up completely

Spillage and Disposal

- 1) If hot mercury is spilled for instance from a mercury diffusion pump, immediately vacate and seal off the room. Under no circumstances should you attempt to re-enter the room. Contact the safety officer.
- 2) Wear goggles and gloves. Work in a hood if possible. Because of the high toxicity of mercury it is important to clean it up as thoroughly as possible, especially in confined areas. An aspirator with capillary tube and filter flask can be used for sucking up droplets. See Figure 3. For small spills, the mercury can be cleaned up with sponge type mercury collector.
- 3) Disposal: Mercury should never be flushed down the drain or thrown in the trash. Not only is mercury expensive it is toxic and accumulates going up the food chain. All mercury even the smallest quantities should be stored in canisters for reprocessing. If the stem of a thermometer becomes broken the mercury in the bulb may be collected by using a glass file to scratch and break the bulb at the base of the thermometer stem.

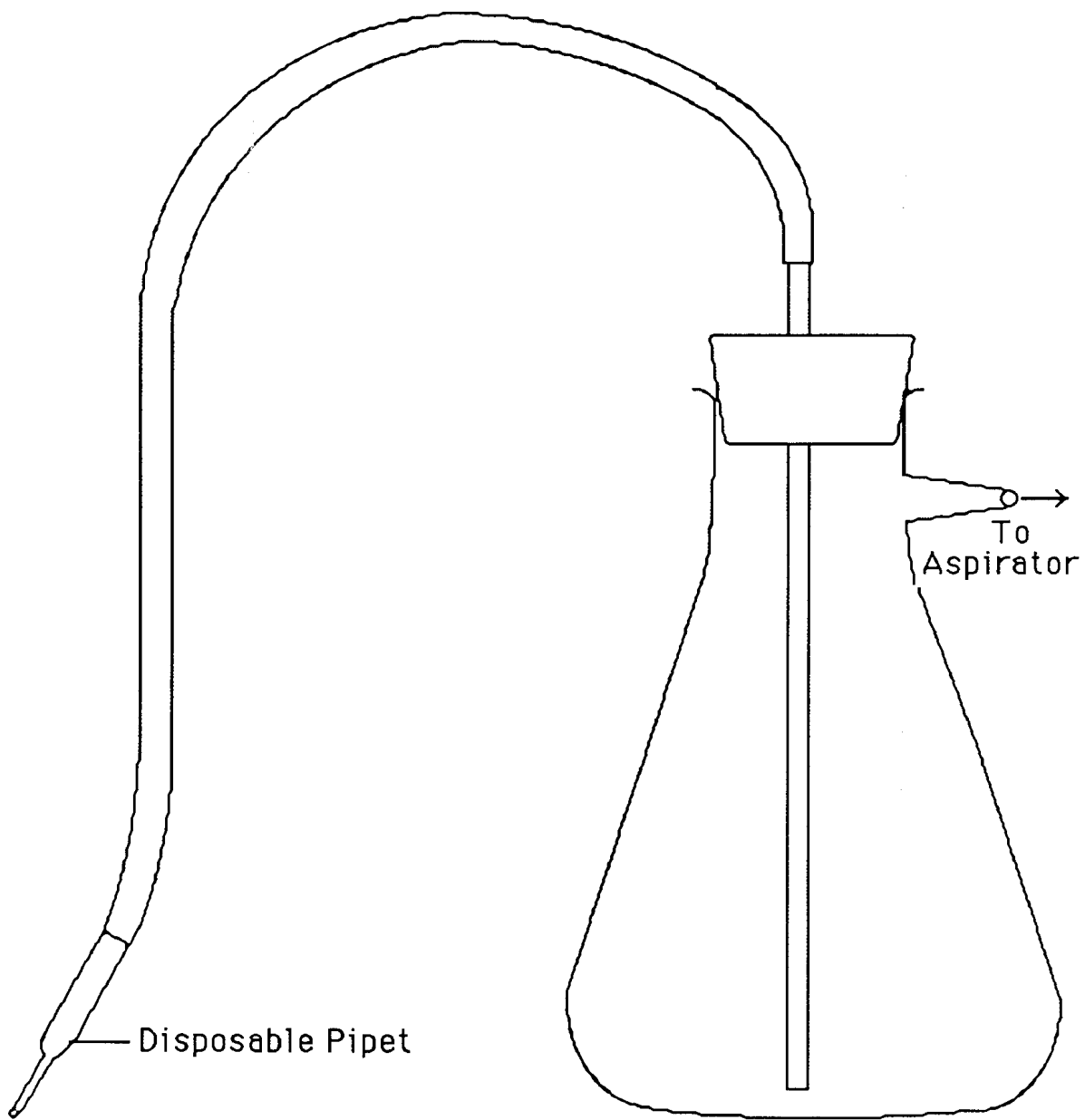


Figure 3. Apparatus for the Cleanup of Mercury Spills.

CHEMICAL DISPOSAL

Chemical disposal has become a tedious and expensive process. The Love Canal, dioxin, and other nationally publicized disasters have demonstrated the effects of mismanaged chemical disposal. In order to maintain a safe and healthful environment, federal regulations have been imposed on chemical disposal. The Resource, Conservation and Recovery Act (RCRA) administered by the Environmental Protection Agency (EPA) provides strict guidelines to insure that the same mistakes regarding chemical disposals never occur again. It is not only legally required but also to our advantage that chemical disposals be handled according to these guidelines. The complete ramifications of RCRA can not be covered in a manual of this scope. However, for the academic laboratories there are several procedures, which can be used to help, keep disposal costs down and also maintain good environmental practices. Most laboratories have established procedures for chemical disposal and you should become familiar with them. If there is any question of the proper disposal procedure for a particular chemical, check a reference for specific details (For instance, see "Hazards in the Chemical Laboratory" 2nd ed., G.D. Muir, Ed., The Chemical Society, London, 1977.). Where procedures have not been established for chemical disposal, the following guidelines may be helpful.

For Small Quantities of Chemicals only:

1. Many inorganic salts can be disposed of down the drain followed by large quantities of water.
2. Strong acids must be neutralized before being flushed down the drain. Natural water is not capable of neutralizing strong acids. Furthermore, concentrated acids may destroy drainpipes unless they are sufficiently diluted. Acids should be diluted with water in an Erlenmeyer flask (remember the acid is always added to the water). Neutralize by slowly adding sodium carbonate or sodium bicarbonate while swirling the flask. When no more gas is evolved with the addition of base, neutralization is complete. Common baking soda from the grocery store is an inexpensive source of sodium bicarbonate.
3. Strong bases should be diluted and neutralized with hydrochloric acid. If excess acid is added this may be neutralized as above. Dispose of down the drain with large volumes of water.
4. Organic compounds which are frequently suitable for disposal down the drain (provided they are not toxic, not resistant to degradation by natural processes and are miscible with water) are short chain alcohols, aldehydes, ketones, esters, some organic acids, hydrazines, amines and amides.

Organic compounds, which may not be disposed of down the drain, are

1. azides and azo compounds
2. carbon disulfide
3. peroxides
4. halogenated hydrocarbons
5. aromatic compounds

Large quantities of any chemical are not to be flushed down the drain. They should be stored in bottles, which have been carefully labeled with as much information as is possible. Completely labeling these chemicals is extremely important. The information will be needed for proper storage and further disposal. If you are setting up an experiment in which the students will be generating large quantities of waste chemicals, receptacles for those chemicals should be placed in the hood. For example, an organic laboratory experiment may require the distillation of a 50:50 mix of cyclohexane and toluene. A bottle should be placed in the hood and labeled "Cyclohexane/Toluene 50:50 Waste Solution." When the bottle is filled it should be dated and stored for later disposal.

CHEMICAL STORAGE

Chemical storage is extremely important in all phases of biology. Separation of chemicals is one of the best ways to eliminate hazardous conditions. Chemicals that are compatible can be stored together, however ample room should be available to avoid accidents. Maximum separation should be established between reactively hazardous chemicals. If possible, a separate room for these chemicals is preferred.

Overstocking of chemicals should be avoided because it increases the possibility of an accident. If there are chemicals that must be stored in large containers, they should be kept close to the floor. In the storage area, climbing for chemicals should be avoided. If climbing is necessary, a proper stepladder should be used. All walkways in the storage area should be kept clear and have easy accessibility to an exit. Availability of two exits is desirable; it provides a safer escape in time of emergency.

Protecting the chemical containers from physical damage is one of the most basic functions of storage. Common sense is the best judge here. The chemicals should not be crowded into a small space and their containers should be handled with care. Periodic inspection of the storage area should be made to remove any unlabeled containers and decomposed or dated chemicals. Also any chemicals that are destroying their containers should be repackaged or properly disposed of. Most storage problems can be avoided with forethought and careful planning. Examples of chemical groupings and storage methods follow.

A. Oxidizing Agents

Examples:

1. salts of permanganic acid
2. hypochlorous acid and salts
3. all chlorates
4. all peroxides
5. nitric acid
6. ammonium nitrate

Proper Storage: Storage areas should be kept cool and well ventilated. It should be constructed of fireproof material and have an automatic sprinkler system.

B. Corrosive Material

Examples:

1. concentrated acids
2. concentrated alkalies
3. borides
4. phosphides
5. alkali metals
6. halogenated hydrocarbons

Proper Storage: Storage area should be well ventilated to prevent build-up of fumes. The chemical containers should be regularly checked for damage.

C. Flammable Materials

Examples:

1. ether
2. ethanol
3. acetone
4. toluene

Proper Storage: Storage area should be kept cool and well ventilated. Fire fighting equipment should be kept close to the storage area. Automatic smoke and fire detectors are recommended. Drums should be electrically grounded and periodically checked for leaks.

D. Water-Sensitive Chemicals

Examples:

1. sodium
2. potassium

Proper Storage: The storage area should be cool, dry, and well ventilated. The chemicals are usually stored under nitrogen or kerosene.

PREPARING CHEMICAL SOLUTIONS

Your responsibility as a laboratory assistant will almost certainly include the preparation of chemical solutions. This section includes much of the information needed to prepare standard solutions, common acids and bases, special reagents, and common chemical solutions. Information on different grades, concentration units, and special precautions are also included, because all of this must be considered to correctly prepare a solution. It is absolutely essential that you follow directions for solution preparation to the letter. When doing calculations in preparation for making solutions, always double check for errors and use common sense evaluations of the results before beginning the preparation. Always read the label of the chemical bottle before, during and after preparing your solutions. Not only can a misread label ruin an experiment but it can also create a hazardous situation. Before beginning solution preparation check for possible hazards and know what to do in case of a spill or an accident.

A. Purity and Chemical Selection

One of the more confusing things about solution preparation is determining the chemical purity required. Chemicals are manufactured in varying degrees of purity. Each manufacturer has its own set of names for the levels of purity of their chemicals. The best way to determine what level purity of a chemical you are working with is to refer to the manufacturers catalog for details. Only use the purity of chemical required for your application. Lower purity chemicals contain residual contaminants which may interfere with the reaction that is being carried out. Higher purity chemicals are much more expensive than those of lower purity and it is wasteful to use them unless required. The following are the standard chemical grades most manufacturers include on their labels and an explanation of the purity.

Technical Grade: Designates chemicals of select commercial quality. Primarily used industrially and not generally suitable for laboratory use because of the level of impurities.

Practical Grade: Designates chemicals of sufficient purity and quality to be suitable for many laboratory applications. They may contain intermediates, isomers, or homologs but are pure enough for many organic preparations.

USP: Designates chemicals, which are sufficiently pure to pass tests prescribed in the U.S. Pharmacopoeia. These chemicals are acceptable for drug use and generally may be used for most laboratory applications.

CP: Designates chemicals, which are chemically pure. Specifications normally are included on the chemical label. These chemicals are not as pure as reagent grade chemicals but when used with discretion they may be suitable for most applications.

Spectroscopic Grade: Designates solvents of extremely high purity suitable for use in spectroscopic applications. The chemicals contain minimum impurities, and specifications are usually included on the label. In some cases background spectra or tabulated spectral absorbencies will be included with the chemical. These are very costly chemicals and should only be used for spectroscopic applications.

Chromatographic Grade: Designates chemicals with a minimum purity of 99 + mole% as determined by gas chromatography. Sample chromatograms are usually included with the chemical.

Reagent Grade: Designates chemicals, which meet standards of purity established by the committee on Analytical Reagents of the American Chemical Society (see reference 18). Each chemical of this grade will be labeled with a batch number on which the quality control testing was done. These chemicals are suitable for chemical analysis applications.

Primary Standard: Designates chemicals, which may be used as reference standards in quantitative analyses (discussed in detail later). These chemicals are of extremely high purity and cost.

HPLC Grade: Designates solvents which may be used for High Pressure Liquid Chromatography. Not only do these chemicals have minimum impurities from a spectroscopic standpoint but they also have been filtered to remove any microparticulates, which may clog an HPLC column. For this reason never use a lower grade solvent for HPLC work

Not only are there the previously listed standard grades of chemicals but in addition there is a confusing array of private label grades which require access to company catalogs to interpret relative purities. As an example, some of the so-called specialty chemical grades produced by various manufacturers follows:

Mallinckrodt, Inc.

ChromAR: solvents certified for HPLC
Nanograde: solvents for pesticide residue analysis
ScintillAR: Liquid scintillation chemicals
SpectAR: Spectrophotometric solvents
OR: organic laboratory research quality chemicals

J. T. Baker Chemical Co.

Ultrex: ultra high purity chemicals
Baker Intra-analyzed: Chemicals suitable for trace elemental analysis
Baker Resi-analyzed: Solvents for organic residue analysis
Photrex: solvents for spectrophotometry
LSC: Liquid scintillation chemicals
Baker Analyzed: primary standard
Baker Grade: organic laboratory research quality chemicals

MCB Manufacturing Chemists, Inc.

Omnisolv: solvents that may be used for HPLC, GC, spectrophotometry, residue analysis and pesticide analysis

Fisher Scientific

Fisher Certified Grade: similar quality to ACS Reagent Grade chemicals including a lot analysis

Fisher Certified ACS Grade: meets ACS standards for Reagent Grade chemicals and includes lot analysis on the label

Fisher Certified HPLC Grade: HPLC Grade

Pesticide Grade: for pesticide residue analysis

Spectranalyzed: solvents for spectrophotometry

Electronic Grade: For use in the electronics industry

Scintanalyzed: for liquid scintillation chemistry

CHEMICAL ORDERS AND INVENTORY

1. An inventory will be made and maintained of all chemicals in the department.
2. All chemicals, except for minimal laboratory needs, will be kept in a central chemical storeroom.
3. Chemical orders will be kept separate from other supply orders. Chemicals requested will be checked against the inventory, and orders will be filled from existing stocks if possible.
4. Filled containers of wastes will be stored in the stockroom to await regularly scheduled professional disposal.