

Lab Safety



Introduction

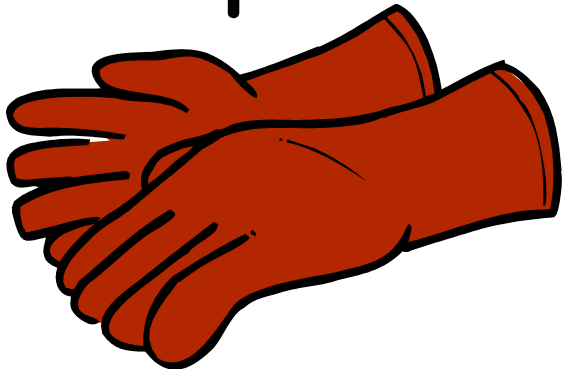


1. A chemical lab is potentially **hazardous environment**
2. Accident and **injury can happen anytime**
3. Lab safety is **everyone's responsibility**
4. Lab safety standards and practices must be strictly followed

General Safety Rules



1. **Listen to or read** instructions carefully before attempting to do anything.
2. Always use appropriate personal protective equipments (lab coat, **safety goggles, masks, gloves, no open shoes, no eye lenses**)



General Safety Rules

3. After handling chemicals, always **wash your hands** with soap and water.

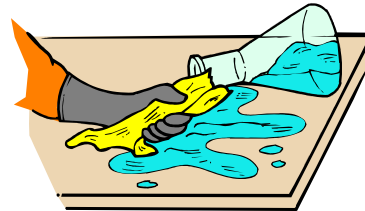


4. During lab work, **keep your hands** away from your face.



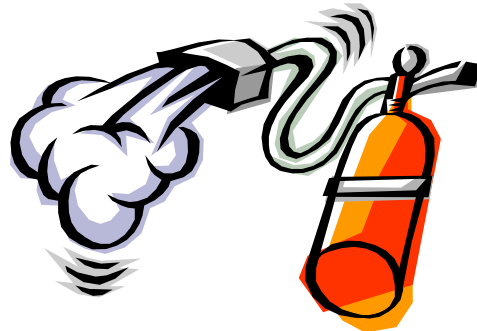
5. Tie back **long hair**.

6. **Notify your supervisor** if any spills or accidents occur.



General Safety Rules

7. Roll up **loose sleeves**.
8. Know the **location** of the fire extinguisher, fire blanket, eyewash station, and first aid kit.
9. Keep your **work area uncluttered**. Take to the lab station only what is necessary.



General Safety Rules



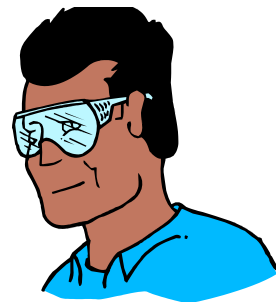
10. It is suggested that you wear **glasses** rather than contact lenses.
11. Never put anything into your **mouth** during a lab experiment.
12. **Clean up** your lab area at the conclusion of the laboratory period.
13. **Never "horse around"** or play practical jokes in the laboratory.



Chemical Safety



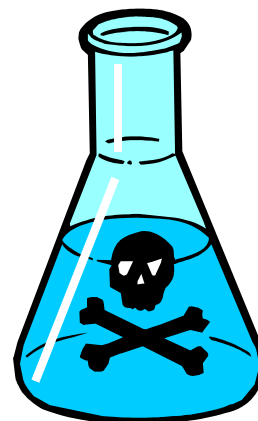
1. Wear **protective goggles** and a lab apron whenever heating or pouring hazardous chemicals.
2. Never **mix chemicals** together unless you are told to do so (and then only in the manner specified).
3. **Never taste** any chemicals (you should never taste anything in the lab).



Chemical Safety



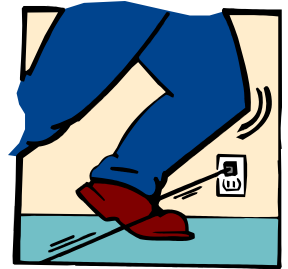
6. **Follow the instructions** of your teacher when disposing of all chemicals.
7. **Wash your hands** after handling hazardous chemicals.
8. **Never mouth pipette**



Electrical Safety

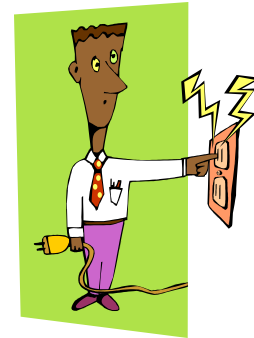


1. Lay **electrical cords** where no one can trip on them or get caught in them.



2. Be sure your **hands and your lab area are dry** before using electrical equipment.

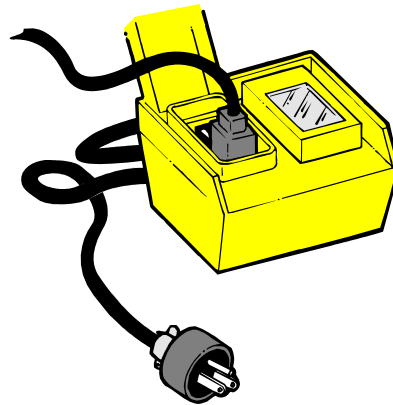
3. Never poke anything into **electrical outlets**.



Electrical Safety



4. Unplug cords by **pulling the plug** and not the cord.
5. **Unplug** all electrical equipment at the **end of the lab period.**



EMERGENCY PROCEDURES

FOR SPILLS/LEAKS OF HAZARDOUS MATERIALS

Rescue



Alarm



Contain



Evacuate



R - Rescue and relocate anyone in immediate danger

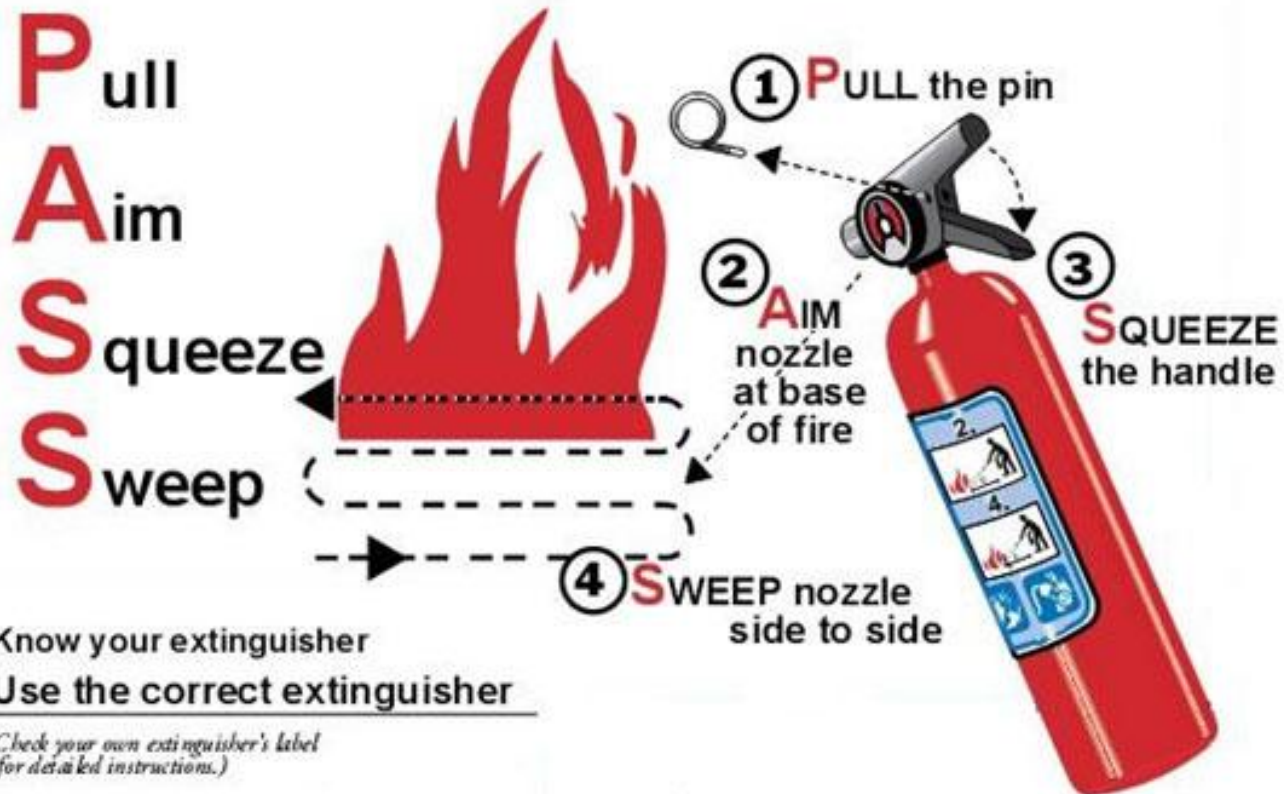
A - Alert others by activating the building fire alarm

C - Confine the emergency by closing the doors

E - Evacuate immediately. Do not use elevators. Use stairs.

Fire Extinguisher

To operate an extinguisher:



Learn how to be always safe

1. **Learn emergency procedures**, and be familiar with the location of fire exits, fire extinguishers, blankets, water showers, eye fountains and first aid
2. **Report all accidents, injuries and spills** to your supervisor
3. **Report** any and all signs and symptoms of **exposure** to your supervisor

Biological safety



1. All biological samples are considered **potentially infectious**
2. Should be handled and processed using **strict precautions**

Waste Disposal



1. For disposal of **contaminated waste**, use containers with **yellow plastic** garbage bags
2. **Regular waste** like paper etc go in the containers with **black/white plastic bags**
3. All sharp objects such as needles, scalpels and even broken glassware go in the **yellow-red sharps container**

Two choices !!

When in doubt...



ACT STUPID!

or





DNA Extraction and Purification

Lab Equipments (To be used in this experiment)

Automatic pipettes



Microcentrifuge



Vortex



Water bath



UV-spectrophotometer



Spectrophotometer



Most of visible spectrophotometers are composed of:

- ☒ Light source which works with visible wavelengths (400-700 nm)
- ☒ Monochromator filter for choosing desired wavelength
- ☒ Sample holder (cuvette)
- ☒ Detector
- ☒ Meter or recorder

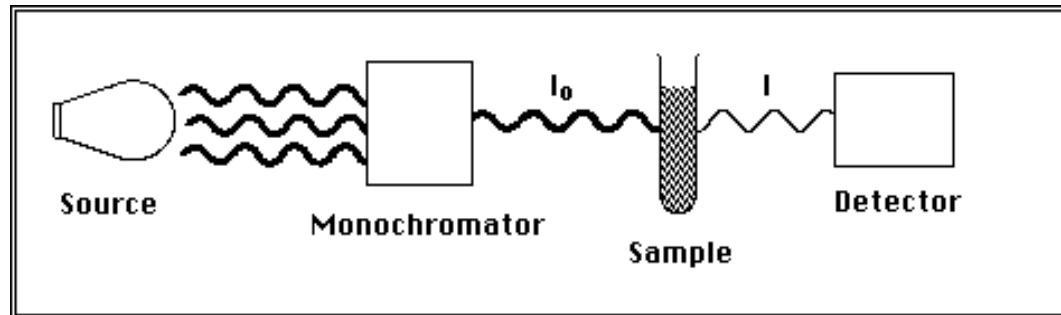


Figure 1

DNA Extraction

⌘ Principle:

1. Lysis of nucleated cells

2. Removal of contaminants:

Any substance other than DNA, e.g., proteins

3. Measurement: UV absorbance at 260nm and 280nm

Purity of DNA solution: 260/280 ratio

DNA concentration: Absorbance at 260nm

DNA Extraction



⌘ Steps:

- ☑ Lysis: of nucleated cells using lysis buffer
- ☑ Binding: of DNA to the membrane of spin column
- ☑ Wash: using wash buffer
- ☑ Elution of pure DNA

Measurements

☑ Dilute the sample of purified DNA:

Add 400 μL of AE to the purified DNA

☑ Measure the Absorbance at 260nm

☑ Measure the Absorbance at 280nm

☑ Assess the DNA purity: 260/280 ratio
(Accepted ratio: 1.7 - 1.9)

☑ Calculate DNA Conc.: Provided $A_{260} = 1.0$,
DNA is 50 $\mu\text{g}/\text{ml}$, unknown DNA conc. can be
calculated by cross multiplication

$$A_{260} = 1.0$$

$$\text{DNA conc.} = 50 \mu\text{g}/\text{ml}$$

$$A_{260} = 0.5$$

$$\text{DNA conc. ?}$$

☒ Calculate DNA Yield: DNA Volume x DNA conc.

Done by the spectrophotometer

DNA Yield:

☒ If you have

Volume of DNA solution: 200 μl (0.2 ml)

DNA conc.: 30 $\mu\text{g}/\text{ml}$

Then the yield (μg) = Volume x Conc.

$$\begin{aligned} & 0.2 \text{ ml} \times 30 \mu\text{g}/\text{ml} \\ & = 6.0 \mu\text{g} \end{aligned}$$

DNA Applications



⌘ **Purified DNA can be used for:**

1. Molecular diagnosis of diseases
(e.g., sickle cell anemia)

2. Forensic applications
(e.g., paternity testing)

3. Molecular biology research

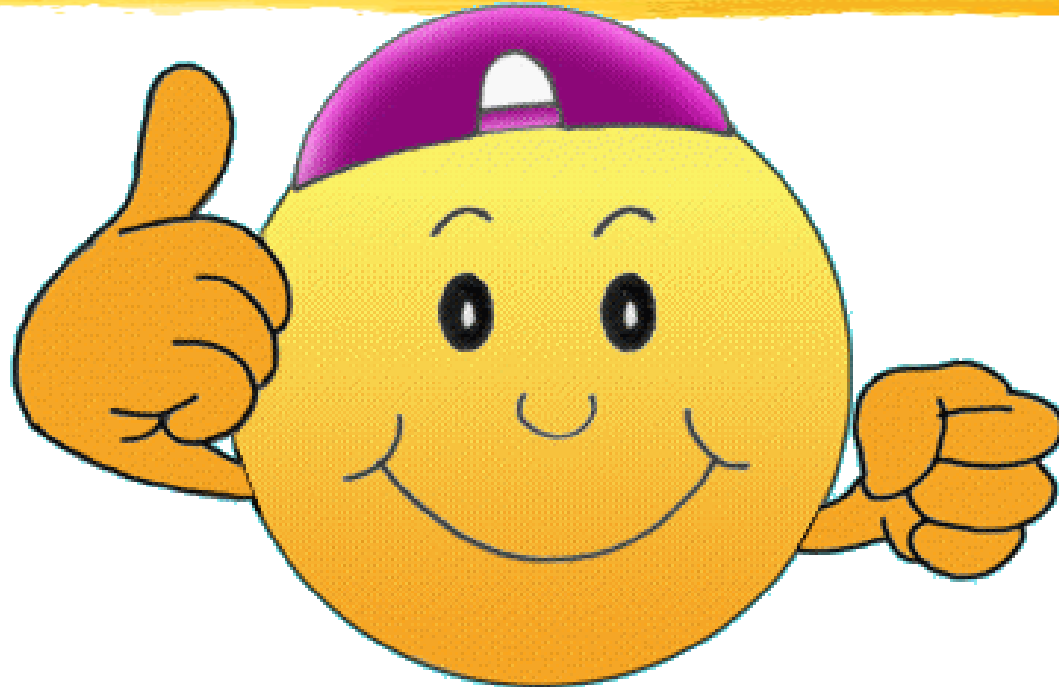
DNA Applications CONT'D



Molecular techniques using purified DNA:

- a. Amplification techniques:** polymerase chain reaction (**PCR**)
- b. Southern blotting**
- c. Restriction Fragment length polymorphism (RFLP)**

GO FOR IT !



GOOD LUCK !