Instructor's Manual

A Laboratory for General, Organic and Biochemistry

Second Edition

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Instructor's Manual

to accompany

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by

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of

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To the Instructor

For the past twenty years we have been concerned with the laboratory experience. During this time we developed a collection of laboratory exercises which demonstrate the concepts we think are most appropriate for a year-long course for students in allied-health areas, agriculture, home economics, dietetics and general education. Though it is a worn term, these experiments "work" the way we say they do. The purpose of this Instructor's Manual is to provide the hints, facts and suggestions to ensure student success.

Each experiment is described here in terms of **Notes to Instructors**, which describe:

I. **Time Required for the Experiment**

Average times required by students to complete the entire experiment or individual sections of the experiment.

II. **Extra Equipment, Supplies and Solutions**

The equipment needed to perform the experiment that is not commonly part of the student's desk equipment will be listed here. In addition, all required solutions, their preparation (if unique), and disposable materials will appear here.

III. **Special Information on Procedures**

Helpful hints, things to watch for, and common student problems will be described here.

IV. **Answers to Preliminary Exercises**

Perhaps the most significant change in the operation of college teaching laboratories in the last several years is hazardous waste disposal. Throughout the laboratory manual and the Instructor's Manual we have placed warnings and suggestions concerning collection of hazardous waste. Though most every institution now has its own unique waste disposal program, there are several excellent sources which should be consulted as the need arises.

Prudent Practices for Disposal of Chemicals from Laboratories, National Academy of Sciences, National Academy Press, Washington, D.C., 1983

Armour, M.A., *Hazardous Laboratory Chemicals, Disposal Guide*, CRC Press, Inc, Boca Raton, 1991

Pitt, M.J. and Pitt, E., *Handbook of Laboratory Waste Disposal*, John Wiley and Sons, New York, 1985

Phifer, R.W. and McTigue, W.R., *Handbook of Hazardous Waste Management for Small Quantity Generators*, Lewis Publishers, Chelsea, MI, 1988

In addition, chemical supply houses, manufacturers, state and federal government agencies can supply information concerning proper disposal. The Material Safety Data Sheets (MSDS) which accompany all reagents, as required by law, describe proper disposal methods for the reagent. These can be obtained from the vendor or manufacturer free of charge. A very large number of MSD sheets are available on the World-Wide Web. Search for MSDS.

CONTENTS

NOTES TO INSTRUCTORS

Experiment 1 — Measurement

I. Time Required for the Experiment

It should require a little more than an hour for the students to do all of the procedures and one-half hour to cover Appendix VII dealing with significant figures and rounding. If desired, the work can be completed the day students check-in to the laboratory.

II. Extra Equipment, Supplies and Solutions[1]

1.	Metal tag (numbered and of known mass)	one/student
2.	Card with straight line drawn on it in dark ink (the length of the line should be known to the instructor)	one/student
3.	Plastic measuring scale approximately 15 cm in length	one/student
4.	Triple-beam, multiple-beam, or electric pan balances	one/4 students
5.	Paper towels	one/student

III. Special Information on Procedures

A. Measuring the Mass of a Solid and Determining the Quality of a Balance

This activity will introduce students to the balance and allow them to learn to use it properly. The importance of reporting measured values to the proper number of *significant figures* and with the proper *units* can be stressed throughout the experiment. If electronic balances are used, then perhaps fewer weighings need be done, and the balance exercise should identify any balance that is not operating properly.

B. Measuring the Volume of a Liquid

Graduated cylinders are not uniform and students should be made aware of this. Some have no divisions below the 5 mL mark while others have 2 or 3 divisions shown.

Since many procedures call for 1 or 2 mL of a solution, the students need to be aware that approximately 20 drops from a common eye dropper approximates 1 mL (18-20 drops in most cases). It is also important that the dropper be held nearly vertical, otherwise drops have a tendency to adhere to the dropper and enlarge, causing students to count fewer drops per milliliter than the number most laboratory droppers deliver. Again, stress that proper units be used.

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1 Equipment and material required to perform the activity that is usually *not* found in the student's personal locker is listed here. In addition, all required reagents will be listed here for convenience.

C. Measuring the Length of a Line

This procedure allows the instructor to emphasize the importance of following directions as students measure a line in both inches and centimeters. Measuring carefully and converting eighths and sixteenths of an inch to decimal values may be difficult if the instructor's guidance is ignored. If careful work is done, the relationship between centimeters and inches should be accurately determined. Again, stress proper *units* when recording measured values.

IV. Answers to Preliminary Exercises

1.a. The balance should not be moved:
  - to prevent damage to balance
  - to keep the balance in a level position
  - so each student knows where the balance is at all times

b. Balance should have all poise returned to the zero position except the largest which should be placed in the 100 g position to prevent the beam from oscillating when not in use. This prevents wear on the knife edge (fulcrum) and the plate on which the knife edge rests.

2. If balance is not zeroed, the mass could be either greater or less than the true value depending on the direction of the error at the rest position.

3.a. The average value is the sum of the measurements divided by the number of measurements.

$$11.36\text{ g} + 11.37\text{ g} + 11.40\text{ g} + 11.38\text{ g} + 11.39\text{ g} = 56.9\text{ g}$$

$$\text{average} = 56.9\text{ g}/5 = 11.38\text{ g}$$

b. Using the approximate method for determining the s.d.:

$$\text{s.d.} = \frac{\text{range}}{\sqrt{N}}$$

$$\text{s.d.} = \frac{11.40\text{ g} - 11.36\text{ g}}{\sqrt{N}} = \frac{0.04\text{ g}}{2.236} = 0.02\text{ g}$$

(stated to one significant figure)

NOTES TO INSTRUCTORS

Experiment 2 — Determining Density and Working with Glass

I. Time Required for the Experiment

The amount of time required to complete this experiment will depend on whether students have previous experience in chemistry/science laboratory courses. Those with experience should be able to complete the activities more quickly than those without. The density determination should take about 90 minutes. Preparing glassware can take up to 2 hours for those who are *most* apprehensive. Assign only those glassware items shown in Figure 10 that will actually be used in later experiments.

II. Extra Equipment, Supplies and Solutions

1.	50-mL burets	one/3 students
2.	density unknowns	one/student (see special information below)
3.	meter sticks	one/3 students
4.	wing tips for burners	
5.	glass scorers	
6.	7-mm soft glass tubing	1 meter/student
7.	5-mm soft glass rod	½ meter/student
8.	3-mm soft glass tubing	5 cm/student (see special information below)
9.	examples of properly and improperly bent tubing (optional)	

> Soft glass for laboratory instruction is becoming increasingly difficult to find in the usual catalogs of glass companies, but it can be purchased from: Fisher Educational Materials Division, 490 W. Lemoyne Street, Chicago, Illinois 60651.

III. Special Information on Procedures

A. Determining Density

The density unknowns can be samples of metal rod, shot or slugs with identifying numbers. To increase the range of densities, short sections of glass tubing partially filled with copper, aluminum or lead shot or powder sealed at both ends can be prepared. An identifying number can also be sealed inside the tube. The density of the entire sealed pod is determined.

Mass and volume should be measured to at least *three significant figures* to ensure the student can read volume and mass to the capacity of the measuring tools. Units *must* accompany all measured values.

Most errors in this procedure are due to incorrectly reading the buret. Emphasize that the buret must be read downward, and caution students to eliminate trapped air bubbles in the buret, especially when the buret is initially filled with water.

B. Using the Laboratory Burner

Demonstrate the proper operation of the burner. An interesting demonstration is to suspend the head of an unburned wooden match just above the top of the barrel of the burner (use a common a pin through the match just below the head), and then light the burner. Though the flame burns normally, the match will not ignite, demonstrating the importance of the inner blue cone as the combustion surface in the structure of the flame.

C. Preparing Glass Tubing

Assign only those items in Figure 10 that will be used in later experiments.

If the Unknown Liquid Experiment is assigned (Experiment 3), you might want to have the students make a short "bubbler tube" to use when the boiling temperature of the liquid is determined. The bubbler tube is a 5-cm-long straight piece of 3-mm tubing which is sealed at one end. When placed in the liquid with the open end down, a fine stream of bubbles will exit the open end of the tube at the boiling temperature. The bubbler is used as an alternative to "boiling chips."

It is important to demonstrate the proper procedures to: cut glass tubing, bend glass tubing (allow the hot glass to fall into the bend), and fire polish sharp ends. Also, emphasize and demonstrate the proper handling of hot glassware. Fire polish a piece of glass tubing and show how the hot glass easily burns through a paper towel. Sometimes the towel will ignite.

IV. Answers to Preliminary Exercises

1. Data should not be recorded on a scrap of paper, since the scrap may inadvertently be thrown away.

2. The balance is prepared for storage by returning all poise to their zero positions except the largest poise, which is set at the 100-g mark.

3. The prepared items of glass tubing should be fire polished to prevent the sharp edge from cutting the person using it, scratching other glassware, or cutting into the rubber when inserted into rubber tubing or a rubber stopper.

4. $d = m/v = (8.47\ g/3.24\ mL) = 2.61\ g/mL$ (3 significant figures)

5. Initial reading (the before figure) = 7.61 mL
   Final reading (the after figure) = 5.73 mL

   Volume of the object = 1.88 mL

NOTES TO INSTRUCTORS

Experiment 3 — The Identification of an Unknown Liquid

I. Time Required for the Experiment

This experiment can be completed in about 90 minutes.

II. Extra Equipment, Supplies and Solutions

1. pipets, 10 mL (need not be class A) — 1/student
2. pipet bulbs (those equipped with conical chucks are preferred) — 1/student
3. 250-500 mL bottles of ethanol or acetone to rinse pipets — 1 bottle/group
4. side-arm test tubes (15 cm) — 1/student
5. corks, size 3 (for 10-cm test tubes)
6. boiling chips for water bath and unknown liquid sample
   (To be used if students did not prepare the bubbler tube in Experiment 2)
   Note: Capillary melting point tubes can also serve as bubbler tubes.
7. unknown liquids – A list of possible unknown liquids appears in the **Table of Unknown Liquids** in the laboratory manual. Dispense 25 mL per student in stoppered and numbered 15-cm test tubes. Some of the liquids listed in the Table are "foils," placed there to increase the challenge of the experiment. Because of the problems of availability, cost or toxicity, you would probably *not* wish to use the following as unknowns:

   Ethylisopropyl ether — Methylbutyl ether
   2,2-Dimethylpentane — Cyclopentane
   and any of the halogenated hydrocarbons

III. Special Information on Procedures

It is important that students perform the three measurements in the sequence in which they are presented in the Experimental Procedure. This will greatly reduce the likelihood of running out of the liquid before all measurements are complete.

Students must be made aware of the proper disposal of the organic liquids at the end of parts A and C (density and boiling temperature determinations) of the experiment. A convenient method of collection is to place labeled waste collection bottles in a convenient location (under the hood is best) for their use. Follow the waste disposal plan adopted by your institution.

A. Determining A Liquid's Solubility in Water

It would be good practice to show students examples of a soluble and an insoluble liquid in water. Impress upon the students that judgements of solubility in water *must* be made using *equal* volumes of water and unknown liquid. Collect the solubility test mixtures and follow the disposal plan adopted by your institution.

B. Determining the Density of A Liquid

If the balances are some distance from the lab benches, or if they must wait to use a balance, students should cover their beakers containing the unknown liquid with a watch glass to minimize evaporation. Students could weigh the beaker + watch glass empty and with 10.00 mL of liquid present. If available, small Erlenmeyer flasks with stoppers could be used in place of beakers.

Pipets must be *clean* if they are to deliver the correct volume of liquid. Students can check for cleanliness by examining the inner walls of the pipet after draining deionized water from unit. If droplets adhere, the pipet is dirty and should be cleaned (not easy to do during the laboratory period) or exchanged for one that is clean. Many students find it difficult to use the pipet and pipet bulb. For this reason, *it is instructive to demonstrate correct use of the pipet and bulb. Emphasize that liquid should never be drawn into a pipet by mouth!*

Note: Densities listed in the Table are for chemically pure samples. Both densities and boiling temperatures may vary somewhat from the Table values for technical grade samples.

C. Measuring the Boiling Point of A Liquid

There is always a risk when using open flames in the vicinity of organic liquids that are producing vapors as they boil. Make certain the rubber hose leading from the side-arm test tube is placed well to the rear of the hood so vapors will be swept quickly away. Either clean one-hole rubber stoppers, one-hole corks or thermometer fitments can be used on the side-arm test tube. In any case, the thermometer should fit tightly with its bulb about 1 cm above the surface of the unknown liquid.

If only 3 or 4 mL of liquid is available to determine the boiling point, it is better to heat the liquid directly with a *soft* flame instead of using the water bath.

With the more volatile liquids, the sample may evaporate before the boiling points are determined. In these cases, a student may measure a temperature closer to that of the heated water than the unknown liquid. Remind students to watch for this problem.

Special emphasis should be placed on the need to heat the unknown *directly with the flame* if it seems to have a boiling point near or above 80°C. Some students seem not to realize that when the water in the bath starts to boil and the unknown is not boiling, that the boiling point of the unknown is above 80°C.

The most common error of this experiment is the incorrect determination of the boiling point. Tell the students that time is required for thermometers to catch up to the temperature of their environment, and that only minimal heating is required at the boiling point.

As stated above, the boiling points given in the Table are for chemically pure samples. The boiling points of most technical grade samples may vary a small amount from the values listed in the Table (usually being lower).

Proper disposal of leftover unknown liquid samples must be emphasized. Care should be taken to ensure students follow the rules.

IV. Answers to Preliminary Exercises

1. Physical (P) and Chemical (C) properties

   C a. P e.

   P b. P f.

   P c. P g.

   C d.

2. $d = m/V = 15.94\ g/10.00\ mL = 1.594\ g/mL$

3. Carbon tetrachloride has a density greater than 1.00 g/mL and will *sink*.
   Hexane has a density less than 1.00 g/mL and will *float* on water.

4. The liquid is acetone.

NOTES TO INSTRUCTORS

Experiment 4 — Elements, Compounds and Mixtures

I. Time Required for the Experiment

The experiment should be completed by nearly all students in 90 minutes.

II. Extra Equipment, Supplies and Solutions

1. One or more display racks (or large test tube racks) with labeled, tightly stoppered test tubes (or screw-capped vials) of each of the following for visual and magnetic examination:

    a. iron filings
    b. sulfur
    c. mercury
    d. magnesium, granular

2. bar magnets
3. Approximately 25 g of iron filings mixed with 25 g of sulfur in an evaporating dish or on a watch glass clearly labeled.
4. Three or four chunks of iron(II) sulfide in an evaporating dish or on a watch glass clearly labeled.
5. white sand (inert material) — 2.5 g/student
6. sodium chloride — 2.5 g/student
7. magnesium (granular) — 1.0 g/student
8. 6 M hydrochloric acid — 10 mL/student
9. filter paper (student grade) — 1 or 2 circles/student
10. corks, size 5 (for 15-cm test tubes)
11. wash bottles or rubber policemen (see Special Information, part B)
12. unknown mixtures (see Special Information, part B)

III. Special Information on Procedures

A. Characteristics of Elements, Compounds and Mixtures:

Emphasize the importance of recording observations clearly, in proper English and in complete sentences. Another person reading a student's notes should understand clearly what was observed.

It is surprising how many students will confuse their observations on the sulfur and iron filings mixture, with the FeS sample.

B. A Study of a Mixture and Its Separation

Though this portion of the experiment is rather straight forward, many students will still need guidance to understand what is happening. Be certain to demonstrate how filter paper is folded, and emphasize the *quantitative transfer* of the hot material from the test tube onto the filter paper. Perhaps a wash bottle or a rubber policeman could be used to facilitate the transfer.

Point out the importance of writing down all observations. In many cases the listing of the components in the mixture will not coincide with the written observations. For example: the appearance of sand on the filter paper may not be noted, yet will be listed as a component!

It is best to have several unknown mixtures prepared for different lab sections or for individual students. No sample should contain both inert materials (sand or carbon).

IV. Answers to Preliminary Exercises

1. Element - A pure substance that cannot be broken down by ordinary chemical means into simpler substances.

   Compound - Two (or more) elements chemically combined in a definite, fixed mass ratio.

   Mixture - Two (or more) substances combined in no definite, fixed mass ratio.

   Filtrate - The liquid that passes through the porous paper in a filtration operation.

2. | | | | | | |
   |---|---|---|---|---|---|
   | M | a. | air | C | e. | distilled water |
   | M | b. | blood | M | f. | wood |
   | M | c. | milk | C | g. | sodium chloride |
   | E | d. | nitrogen | E | h. | oxygen |

3. One method (of several) to separate C, S, KNO_3 would be to proceed as follows:

   a. Add a few milliliters of carbon disulfide to the mixture and stir to dissolve the sulfur.

   b. Filter - This should allow the carbon disulfide-sulfur solution to pass through the filter paper (the filtrate) leaving a mixture of solid carbon and potassium nitrate on the filter paper. The sulfur can be recovered by allowing the carbon disulfide to evaporate.

   c. Pour several milliliters of hot water over the mixture of carbon and potassium nitrate which remains in the cone of filter paper. This will dissolve the potassium nitrate leaving the solid carbon behind, retained by the filter paper. The KNO_3 can be recovered from the hot water by evaporating the water.

NOTES TO INSTRUCTORS

Experiment 5 — Separation Using Chromatographic Techniques

I. Time Required for the Experiment

Procedure A: If students can prepare the 21.5-cm x 11.5-cm rectangle of chromatography paper the period before this experiment is to be done, the chromatography of food colors can be completed in one hour. Otherwise, an additional 20 minutes will be required to prepare the paper.

Procedure B: The paper chromatography of inks requires 1 hour.

Procedure C: The paper chromatography of three cations requires 1 hour.

Procedure D: The extraction and thin-layer chromatography of plant chloroplast requires 40 minutes if commercial thin layer plates are used. If students must prepare their own thin-layer plates, add an additional 20 minutes to the procedure.

II. Extra Equipment, Supplies and Solutions

Procedure A: Paper Chromatography of Food Coloring Components

You can readily identify the FD&C food colors in foods if you obtain samples of them. The ones currently available are: Blue #1, Red #3, Yellow #5, Blue #2, Red #40 and Yellow #6. If you cannot obtain the FD&C dyes, use commercial food colors, such as those from Krogers or McCormick. They contain most of the currently used food colors.

Suggested foods that can be used for the experiment are:

a. highly colored soft drinks (Nehi® grape and orange; Big Red®),
b. Kool Aid® (fairly good),
c. Easter egg coloring dyes (very good) and
d. commercial cake coloring (very good).

	Amounts are for 10 students
components of the developing solvent	
15 mL deionized H_2O	170 mL
15 mL propanol (1-propanol)	170 mL
15 mL butanol (1-butanol)	170 mL

This solvent mixture must be mixed by swirling in the developing chamber just before the paper cylinder is put in place. It has been reported that alcohols with 4 or fewer carbon atoms are safe for drain disposal, but **check your disposal procedures first.**

1.0% aqueous solutions of FD&C food colors	1 mL/student
Food colors may be poured into watch glasses, and set out with 2 or 3 capillary tubes for spotting the sample.	
chromatography paper, cut to 21.0 cm x 11.5 cm	10+ sheets
Saran® wrap (to cover 600-mL beaker)	10 pieces

rubber band (large)	10
extra, small pieces of chromatography paper	20 pieces
pencils (#2)	4 (10/class)
plastic scale with mm scale	10
stapler	3/class
brown paper towels	2
self-sticking tape	3 rolls/class
capillary tubes	25/class

Coagulation capillary tubes that are 75 mm in length and 0.5 to 0.9 mm inner diameter work well.

Procedure B: Paper Chromatography of Inks

The same developing solvent and developing chamber system is used here and in Procedure A. Additional supplies and reagents will be the same.

A wide variety of felt tip pen ink as well as writing ink can be analyzed.

Procedure C: Paper Chromatography of Cations

	Amounts are for 10 students
6 M acetic acid	55 mL

(In a dropper bottle or an automatic dispenser set to deliver 0.5 mL. Measuring dispensers are useful for the water and 1-butanol, too.)

deionized H_2O	230 mL
1-butanol	185 mL
0.025 M potassium chromate	1 unit

One 20-cm test tube two-thirds full of the potassium chromate solution is set out in the laboratory for every ten students.

To prepare 200 mL of the solution, dissolve 0.97 gram of K_2CrO_4 in about 100 mL of deionized water and dilute with mixing to 200 mL.

20-cm test tube	80
No. 4 rubber stopper with slit in bottom	80
chromatography paper 18 cm x 1.5 cm	80 strips
plastic scale	5
pencils (#2)	5
self-sticking tape	one roll
brown paper towels	10
test tube rack to hold eight 20-cm test tubes	10
capillary tubes, as in Procedure A	10
cation unknowns	10

Cation unknowns are prepared by combining volumes of the individual, known cation solutions.