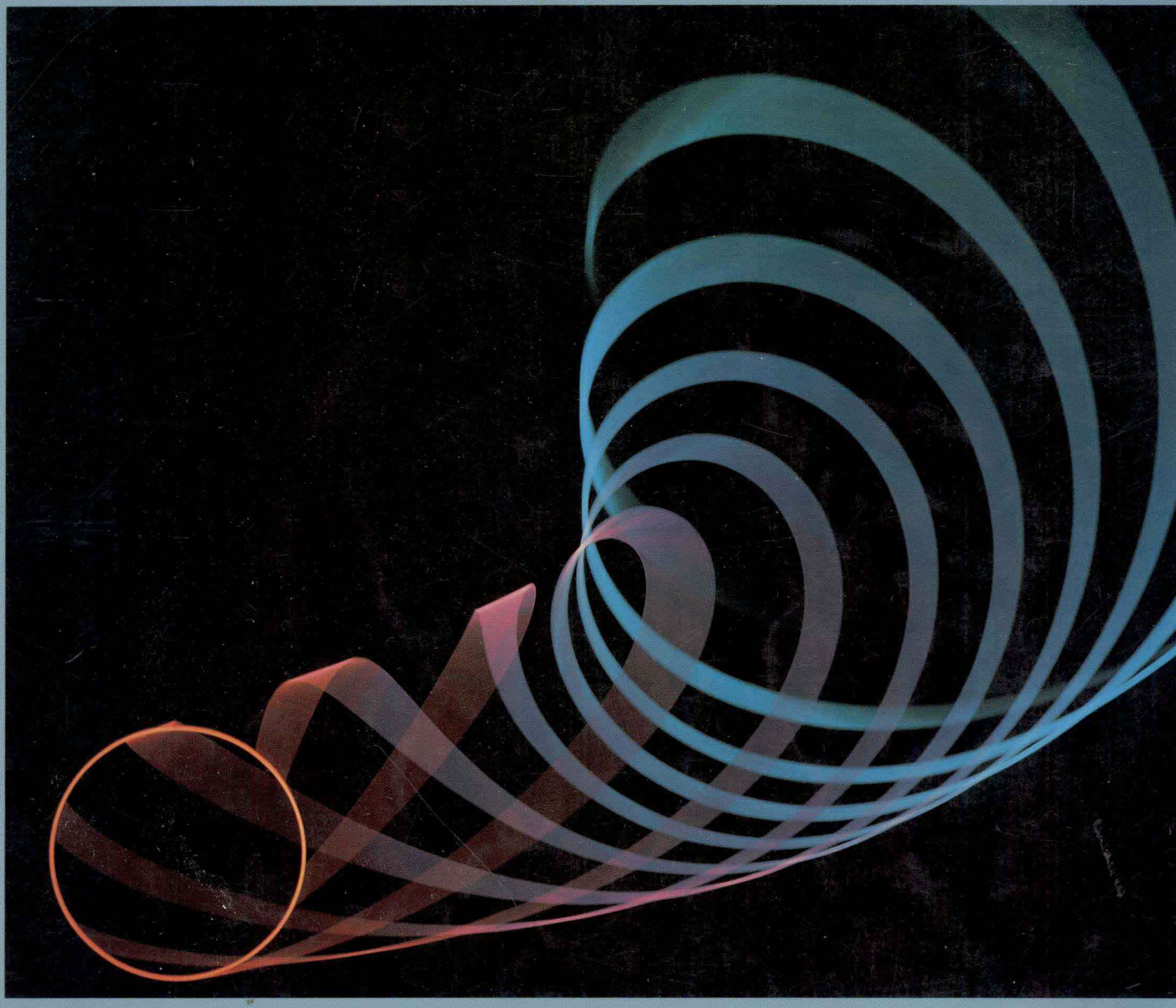
EXPLORATIONS IN BASIC BIOLOGY

FIFTH EDITION



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PREFACE

The fifth edition of *Explorations in Basic Biology,* like earlier editions, is for use in general biology courses in which students have little or no prior laboratory experience. It is compatible with any modern biology text. The exercises provide a variety of options for either one- or two-semester courses. They are designed for a standard three-hour laboratory session but also are adaptable to a two-hour laboratory format.

FEATURES OF THE MANUAL

The manual is designed to *simplify the work of instructors* and to *enhance learning by students.* Instructors will appreciate these features:

1. The exercises are basically self-directing, which eliminates the need for lengthy explanations by the instructor.
2. The exercises use standard equipment and materials that are available at most colleges.
3. Required materials are listed for each section of the exercises to facilitate laboratory preparation.
4. The exercises and the major subunits of most exercises are self-contained so that the instructor may arrange the sequence of exercises to suit the emphasis of the course. If time constraints are present, portions of an exercise may be omitted without destroying its continuity. A distinct effort has been made to provide a variety of choices for the instructor.
5. Activities to be performed by students are identified as *Assignments* that are numbered sequentially within each exercise to facilitate identification and discussion.
6. Students are asked to color code some illustrations to aid their understanding of anatomical features. They are encouraged to color code other figures whenever it will enhance understanding.
7. *Assignments* and *laboratory procedures* are separated from the text and clearly described in a stepwise manner.
8. The accompanying instructor's guide provides (1) composite lists of equipment and materials, (2) sources of supplies, (3) operational suggestions, and (4) answer keys for the Laboratory Reports.

The manual includes these pedagogical features to facilitate learning by the student:

1. Each exercise begins with a list of objectives that outline the minimal learning responsibilities of the student.
2. Numerous illustrations facilitate the student's understanding of background material and laboratory procedures.
3. Key terms are emphasized in bold print for easy recognition and to help the student develop a biological vocabulary.
4. Essential background material is provided in each exercise so that the student has the information required to successfully complete the laboratory activity.
5. Students are required to demonstrate an understanding of background material

by labeling diagrams or answering questions or both before beginning the laboratory activities.
6. Laboratory procedures are separated from the background material and are clearly described. Illustrations are included where appropriate.
7. The laboratory reports guide and reinforce student learning and serve as a convenient means of assessing student understanding.

MAJOR CHANGES IN THE FIFTH EDITION

There are a number of new features that enhance the utility of the manual. Many of these improvements have been suggested by adopters.

1. Additional laboratory activities have been included in many exercises.
2. Major improvements have been made in these exercises: 2-The Cell; 17-Gas Exchange; 22-Neural Control; 24-Muscles and Movement; 32-Molecular and Chromosomal Genetics; 34-Evolutionary Mechanisms; and 35-Ecological Relationships.
3. Exercises in the section on diversity of organisms include more taxonomic terminology to better serve courses that emphasize it. Use of the term Division has been included as appropriate. The identification of "unknown organisms" has been added to these exercises to stimulate the learning of recognition characteristics.
4. Twenty-nine new illustrations have been added, and forty others have been improved.
5. An improved format for the laboratory reports will facilitate grading and discussion. Most laboratory reports contain new questions. Students are required to write new terms in order to reinforce their meanings and correct spellings.
6. The exercise on fungi and algae has been separated into two exercises to increase the scheduling flexibility for instructors.
7. An exercise on the dissection of the frog has been added for use in those courses that do a frog dissection.

ACKNOWLEDGMENTS

Many adopters across the country have provided useful information and suggestions by completing questionnaires, and some have contacted us directly with their helpful observations. All of these contributions, as well as those from colleagues at Pasadena City College, are gratefully acknowledged. We especially appreciate the detailed review from Daniel Fertig. The original artwork by Mary Dersch has again made a significant contribution to the manual.

Adopters are encouraged to communicate any problems that are encountered or helpful suggestions that will improve the usefulness of the manual.

TO THE STUDENT

Laboratory study is an important part of a course in biology. It provides an opportunity for you to observe and study biological organisms and processes and to correlate your findings with the textbook and lectures. It allows the conduction of experiments, the collection of data, and data analysis to form conclusions. In this way, you experience the process of science, and it is this process that distinguishes science from other disciplines.

Many students find the laboratory sessions to be the most interesting part of the course. Your enjoyment depends on your attitude, motivation, and success in the laboratory activities. In turn, your success in the laboratory depends on how you prepare for and carry out the laboratory activities.

PREPARATION

1. Read the laboratory exercise *before* coming to the laboratory session to (a) understand the objectives and background information, (b) learn the meaning and spelling of new terms, and (c) understand the procedures to be followed.
2. Prior to the laboratory session, complete the items on the laboratory report that cover the background material.
3. Color anatomical parts in illustrations with colored pencils to help you distinguish them. Record your color key on the illustration for future reference.
4. Bring your textbook to the laboratory for use as a reference.

LABORATORY ACTIVITIES

1. Remove the **laboratory report** from the manual so you do not have to flip pages when answering questions. Keep your completed laboratory reports in a three-ring binder for future reference.
2. Follow the directions explicitly and in sequence unless directed otherwise.
3. Work carefully and thoughtfully. You will not have to rush if you are well prepared.
4. Answer the questions on the laboratory report thoughtfully and completely. They are provided to guide the learning process. Just filling in the blanks is not acceptable.
5. Discuss procedures and observations with other students. If you become confused, ask your instructor for help.

LABORATORY SAFETY AND HOUSEKEEPING

1. Use equipment with care. Report any problems to your instructor.
2. Immediately clean up any breakage or spills, and inform your instructor of the problem.
3. Immediately report any injuries, even minor ones, to your instructor.
4. Tie back long hair and roll up loose sleeves when using open flames.
5. Clean glassware and equipment before and after use. Return each item to its proper location at the end of the session, and clean your workstation.
6. Do not smoke, eat, or drink in the laboratory.

CONTENTS

PART I

SOME FUNDAMENTALS

EXERCISE

1

THE MICROSCOPE

OBJECTIVES

On completion of the laboratory session, you should be able to:

1. Identity the parts of compound and dissecting microscopes and state the function of each.
2. Describe and demonstrate the correct way to:
   a. Carry a microscope
   b. Clean the lenses
   c. Prepare a wet-mount slide
   d. Focus with each objective
   e. Determine the total magnification
   f. Estimate the size of objects
3. Define all terms in bold print.

A **microscope** is a precision instrument and an essential tool in the study of cells, tissues, and minute organisms. It must be handled and used carefully at all times. Most of the microscopic observations in this course will be made with a **compound microscope,** but a **dissecting microscope** will be used occasionally. A microscope consists of a lens system, a controllable light source, and a mechanism for adjusting the distance between the objective lens and the object to be observed.

To make the observations required in this course, you must know how to use a microscope effectively. This exercise provides an opportunity for you to develop skills in microscopy.

THE COMPOUND MICROSCOPE

The major parts of the compound microscope are shown in Figure 1.1. Refer to this figure as you read this text, and *label the parts* indicated on the figure. Your microscope may be somewhat different than the one illustrated.

The **base** rests on the table and, in most microscopes, contains a built-in **light source** and a **light switch.** Some microscopes have a light intensity (voltage) control knob on the base, usually associated with the light switch. The **arm** rises from the base and supports the stage, lens system, and control mechanisms. The **stage** is the flat surface on which microscope slides are placed for viewing. **Stage clips,** or a **mechanical stage,** hold the slide in place.

Most microscopes have a **condenser** located below the stage. It concentrates the light on the object and may be raised or lowered by the **condenser control knob.** Usually, the condenser should be raised to its highest position. An **iris diaphragm** is built into the base of the condenser. The **iris diaphragm control lever** (a rotatable wheel in some microscopes) varies the amount of light entering the condenser and the lens system.

The **body tube** is supported by the arm and has an **ocular lens** at the upper end and a **revolving nosepiece** with the attached **objective lenses** at the lower end. The nosepiece is rotated to bring different objectives into viewing position. The objectives usually click into viewing position. Student microscopes usually have three objectives. The shortest is the

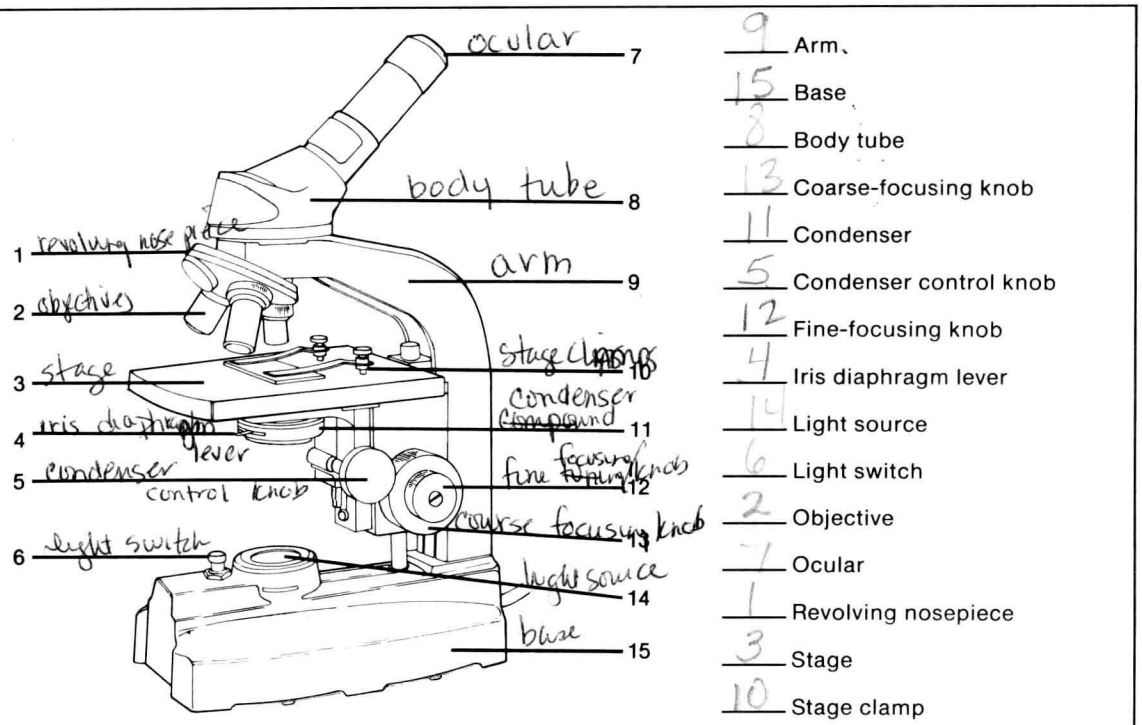

Figure 1.1. Compound microscope

scanning objective, which has a magnification of 4×. The **low-power objective** with a 10× magnification is intermediate in length. The **high-power** (high-dry) **objective** is the longest and usually has a magnification of 40×, but may have a 43× or 45× magnification in some microscopes. In this manual, the high-power objective is often called the 40× objective. Some microscopes have an **oil-immersion objective** (100× magnification) that is a bit longer than the high-power objective.

There are two focusing knobs. The **coarse-focusing knob** has the larger diameter and is used to bring objects into rough focus when using the 4× and 10× objectives. The **fine-focusing knob** has a smaller diameter and is used to bring objects into fine focus. It is the *only* focusing knob used with the high-power and oil-immersion objectives.

Magnification

The magnification of a microscope is determined by the power of the ocular and objective lenses being used. The ocular usually has a 10× magnification. The powers of the objectives may vary but usually are 4×, 40×, and 100×. The **total magnification** is calculated by multiplying the power of the ocular by the power of the objective.

Resolving Power

The quality of a microscope depends on its availability to **resolve** (distinguish) objects. Magnification without resolving power is of no value. Modern microscopes increase both magnification and resolution by a careful matching of light source and precision lenses. Most microscopes have a blue light filter located in either the condenser or the light source since resolving power increases as the wavelength of light decreases. Student microscopes usually can resolve objects that are 0.5 μm or more apart. The best light microscopes can resolve objects that are 0.1 μm or more apart.

Contrast

Sufficient **contrast** must be present among the parts of an object for the parts to be distinguishable. Contrast results from the differential absorption of light by the parts of the object. Sometimes, stains must be added to a specimen to increase the contrast. A reduction in the amount of light improves contrast when viewing unstained specimens.

Focusing

A microscope is focused by increasing or decreasing the distance between the specimen on the slide and the objective lens. The focusing procedure used depends on whether your microscope has a **movable stage** or **movable body tube** (see Figure 1.2). Both procedures are described below. Use the one appropriate for your microscope. As a general rule, you should start focusing with the low-power (10X) objective unless the size of the object requires starting with the 4X objective.

Focusing with a Movable Body Tube

1. Rotate the 10X objective into viewing position.
2. While using the coarse-focusing knob and *looking from the side* (not through the ocular), lower the body tube until it stops or until the objective is about 3 mm from the slide.
3. While looking through the ocular, slowly raise the body tube by turning the coarse-focusing knob toward you until the letters become visible. Use the fine-focusing knob to bring the letters into sharp focus.

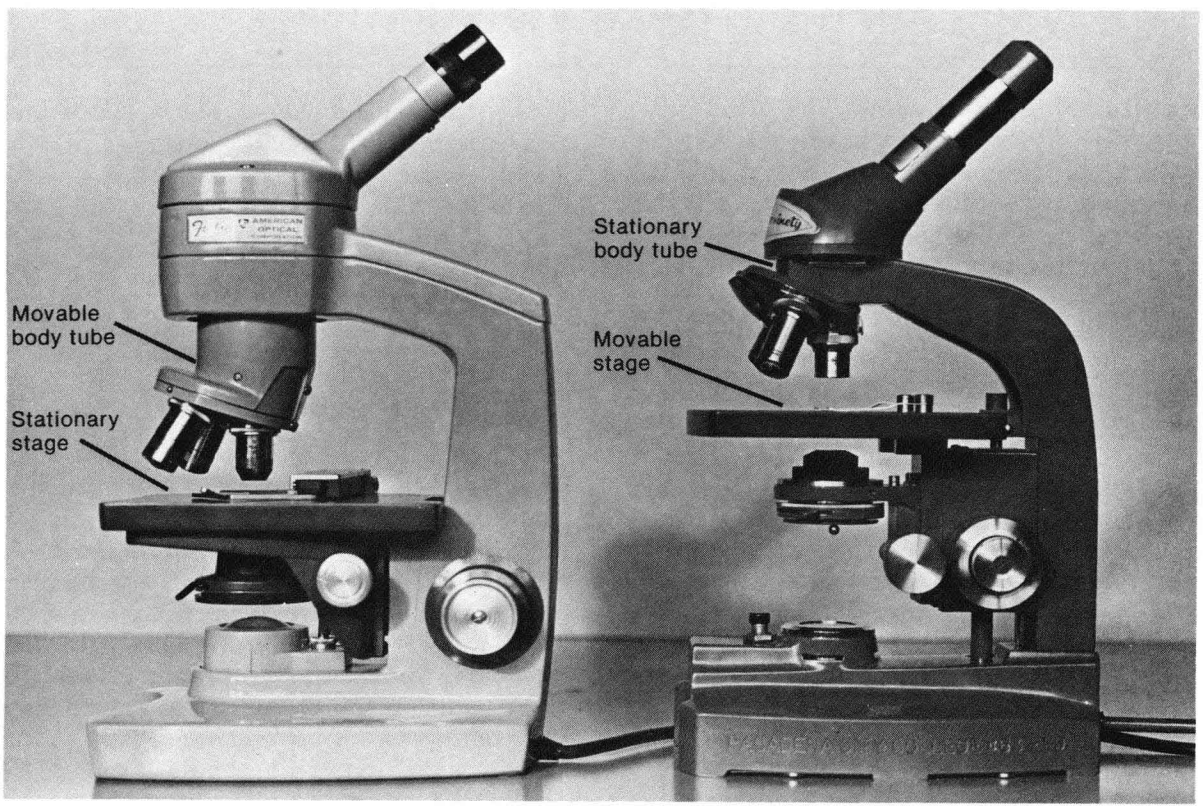

Figure 1.2. Comparison of two microscope designs

Focusing with a Movable Stage

1. Rotate the 10X objective into viewing position.
2. While using the coarse-focusing knob and *looking from the side* (not through the ocular), raise the stage to its highest position or until the slide is about 3 mm from the objective.
3. While looking through the ocular, slowly lower the stage by turning the coarse-focusing knob away from you until the letters come into focus. Use the fine-focusing knob to bring the letters into sharp focus.

Switching Objectives

Your microscope is **parcentric** and **parfocal.** This means that if an object is centered and in sharp focus with one objective, it will be centered and in focus when another objective is rotated into the viewing position. However, slight adjustments to recenter and refocus (with the fine-focusing knob) may be necessary. As you switch objectives from 4X to 10X to 40X to increase magnification, the (1) working distance, (2) diameter of the field, and (3) light intensity are reduced as magnification increases. Note this relationship in Figure 1.3.

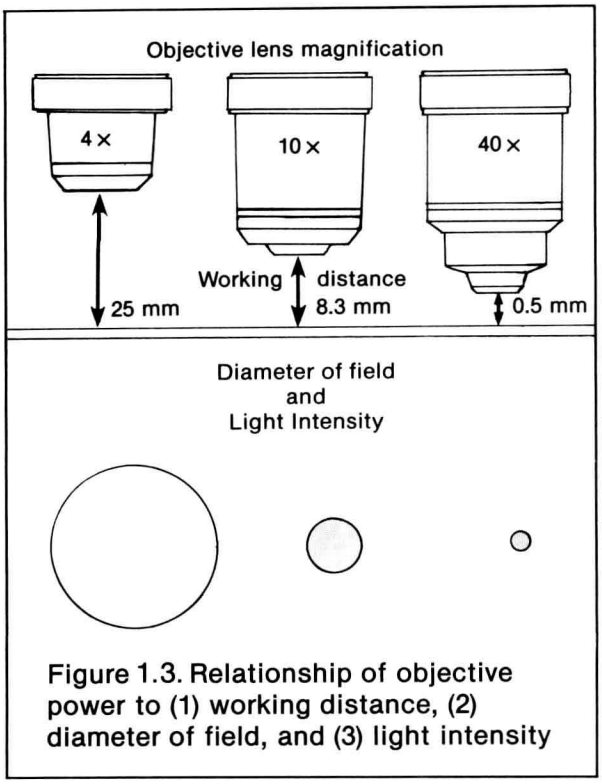

Figure 1.3. Relationship of objective power to (1) working distance, (2) diameter of field, and (3) light intensity

Slide Preparation

Specimens to be viewed with a compound microscope are placed on a **microscope slide** and are usually covered with a **cover glass.** Specimens may be mounted on slides in two different ways. A **prepared slide** (permanent slide) has a permanently attached cover glass, and the specimen is usually stained. A **wet-mount slide** (temporary slide) has the specimen mounted in a liquid, usually water, and covered with a cover glass. In this course, you will observe commercially prepared permanent slides and wet-mount slides that you will make. Wet-mount slides are prepared as shown in Figure 1.4.

Care of the Microscope

You should carry a microscope upright in front of you, not at your side. Use one hand to support the base and the other to grasp the arm. See Figure 1.5. Develop the habit of cleaning the lenses prior to using the microscope. Use only special lint-free **lens paper.** If the lens paper does not clean the lenses, inform your instructor. If any liquid gets on the lenses during use, wipe it off immediately and clean the lenses with lens paper.

When you are finished using the microscope, perform these steps:

1. Remove the slide. Clean and dry the stage.
2. Clean the lenses with lens paper.
3. Rotate the nosepiece so that no objective projects beyond the front of the stage.
4. Raise the stage to its highest position *or* lower the body tube to its lowest position in accordance with the type of microscope you are using.
5. Unplug the light cord and loosely wrap it around the arm below the stage. Add a dustcover, if present.
6. Return the microscope to the correct cabinet cubicle.

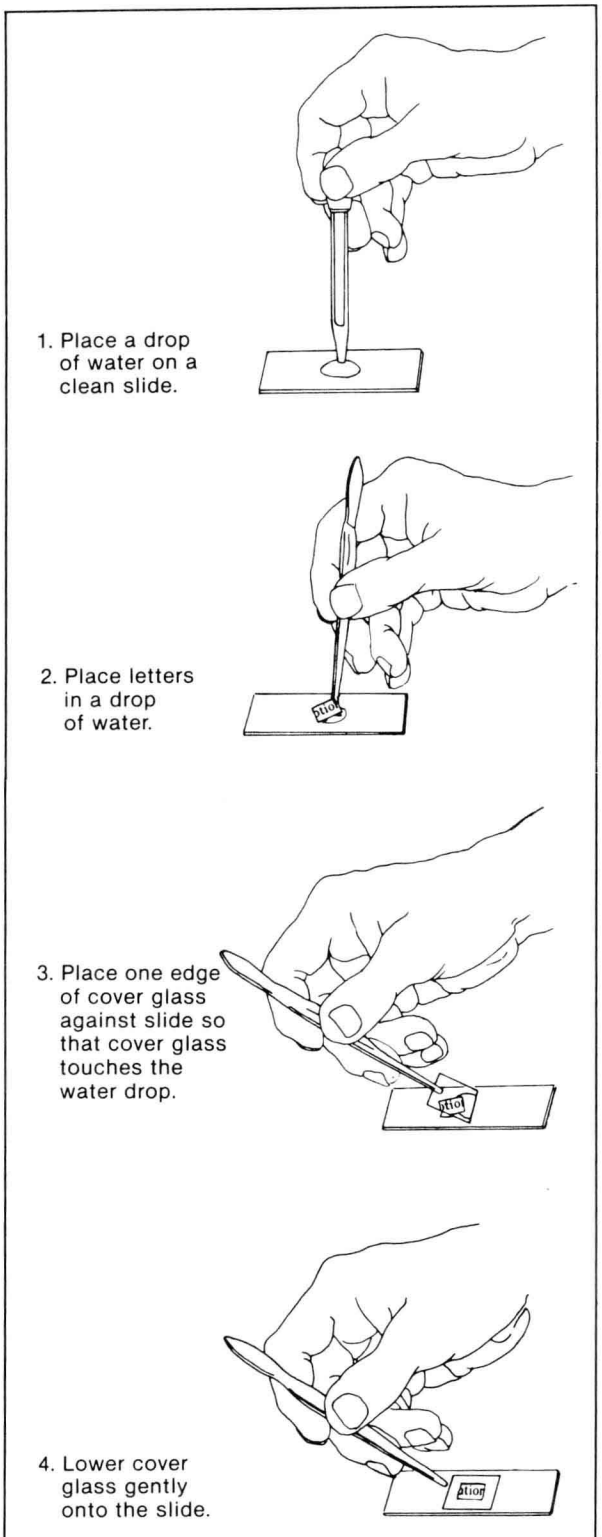

Figure 1.4. Preparation of a wet-mount slide

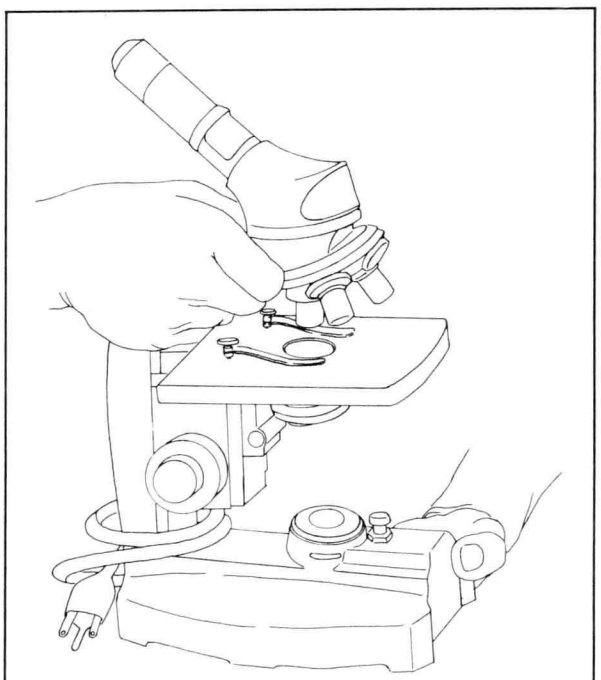

Figure 1.5. How to carry the microscope.
Grasp the arm of the microscope with one hand and support the base with the other.

Assignment 1

1. Label Figure 1.1.
2. Obtain the microscope assigned to you. Carry it as described above and place it on the table in front of you. Locate the parts shown in Figure 1.1. Clean the lenses with lens paper. Try the knobs and levers to see how they work.
3. Raise the condenser to its highest position and keep it there. Plug in the light cord and turn on the light. If your microscope has a voltage control knob, adjust to an intermediate position to prolong the life of the bulb.
4. Rotate the 4X objective into viewing position and look through the ocular. The circle of light that you see is called the **field of view** or simply the **field.**
5. While looking through the ocular, open and close the iris diaphragm and note the change in light intensity. Repeat for each objective and note that light intensity decreases as the power of the objective increases. Thus, you will need to adjust the light intensity when you switch

objectives. *Remember* to use reduced light intensity when you are viewing unstained and rather transparent specimens.

6. If your microscope has a voltage control knob, repeat item 5 while leaving the iris diaphragm open but changing the light intensity by altering the voltage.
7. ***Complete item 1 on Laboratory Report 1 that begins on page 327.***

Developing Microscopy Skills

The following microscopic observations are designed to help you develop skill in using a compound microscope.

Materials

Dissecting instruments
Kimwipes
Medicine droppers
Metric ruler, clear plastic
Microscope slide and cover glass
Newspaper
Water in dropping bottle
Prepared slide of fly wing

Assignment 2

1. Obtain a microscope slide and cover glass. If they are not clean, wash them with soap and water, rinse, and dry them. Use a paper towel to dry the slide, but use Kimwipes to blot the water from the fragile cover glass.
2. Use scissors to cut three letters from a newspaper with the letter "i" as the middle letter.
3. Prepare a wet-mount slide of the letters as shown in Figure 1.4. Use a paper towel to soak up any excess water. If too little water is present, add a drop at the edge of the cover glass and it will flow under the cover glass.
4. Place the slide on the stage with the letters over the stage aperture, the circular opening in the stage. Secure it with either a mechanical stage or stage clamps. See Figure 1.6. The slide should be parallel to the edge of the stage nearest you with the letters oriented so that they may be read with the naked eye.
5. Rotate the 10X objective into viewing position and bring the letters into focus using the focusing procedure described above that is appropriate for your microscope.
6. Rotate the 4X objective into the viewing position. Center the letter *i* and bring it into sharp focus. Can you see all of the *i*? Can you see the other letters? What is different about the orientation of the letters when viewed with the microscope instead of the naked eye?
7. Move the slide to the left while looking through the ocular. Which way does the image move? Practice moving the slide while viewing through the ocular until you can quickly place a given letter in the center of the field.
8. Center the *i* and bring it into sharp focus. Rotate the 10X objective into position. Is the *i* centered and in focus? If not, center it and bring it into sharp focus. How much of the *i* can you see?
9. Rotate the 40X objective into position. Is it centered and in focus? All that you can see at this magnification are "ink blotches" that compose the *i*. If you do not

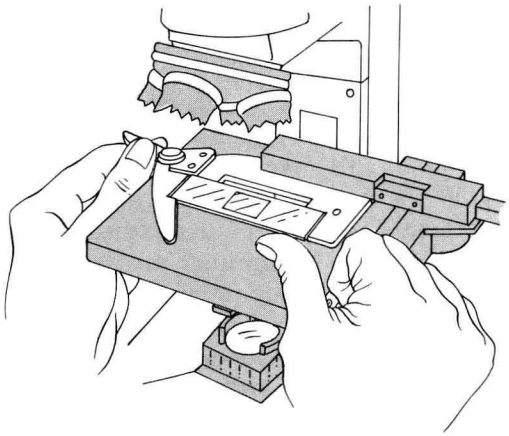

Figure 1.6. Placing the slide in the mechanical stage. The retainer level is pulled back to place the slide against the stationary arm. Then, the retainer lever is released to secure the slide

see this, center the *i* and bring it into focus with the *fine focusing knob. Never use the coarse-focusing knob with the high-power objective.*

10. Practice steps 1 through 4 until you can quickly center the dot of letter *i* and bring it into focus with each objective. **Remember,** you are *never* to start observations with the high-power objective. Instead, start at a lower power and work up to the 40X objective.
11. ***Complete item 2 on the laboratory report.***
12. Remove the slide and set it aside for later use.

Depth of Field

When you view objects with a microscope, you obviously are viewing the objects from above. The vertical distance within which structures are in sharp focus is called the **depth of field,** and it decreases as magnification increases. You will learn more about depth of field by performing the observations that follow.

Assignment 3

1. Obtain a prepared slide of a fly wing. Observe the tiny spines on the wing membrane with each objective, starting with the 4X objective. Can you see all of a spine at each magnification?
2. Using the 4X objective, locate a large spine at the base of the veins and center it in the field. Can you see all of it?
3. Rotate the 10X objective into position and observe the spine. Can you see all of it? Practice focusing up and down the length of the spine, and note that you can see only a portion of the spine at each focusing position.
4. Rotate the 40X objective into position and observe the spine. At each focusing position, you can see only a thin "slice" of the spine. To determine the spine's shape, you have to focus up and down the spine using the *fine-focusing knob.*
5. ***Complete item 3 on the laboratory report.***

The preceding observations demonstrate that when viewing objects with a greater depth (thickness) than the depth of field, you see only a two-dimensional plane "optically cut" through the object. To discern an object's three-dimensional shape, a series of these images must be "stacked up" in your mind as you focus through the depth of the object.

Diameter of Field

When using each objective, you must know the diameter of the field to estimate the size of observed objects. Estimate the diameter of field for each magnification of your microscope as described in the section that follows.

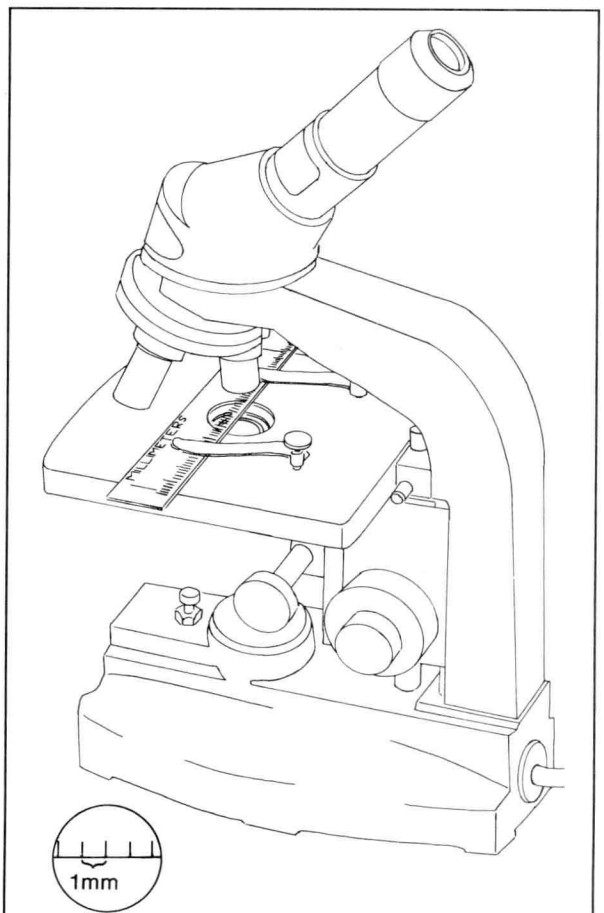

Figure 1.7. Estimating the diameter of field

Assignment 4

1. Place the ruler on the microscope stage as shown in Figure 1.7. The edge of the ruler should extend across the diameter of the field. Focus on the metric scale with the 4X objective, and adjust the ruler so that one of the millimeter marks is at the left edge of the field. Estimate and record the diameter of field at 40X by counting the spaces and portions thereof between the millimeter marks.
2. Use these equations to calculate the diameter of field at 100X and 400X:

$$\text{Diam. (mm) at 100X} = \frac{40X}{100X} \times \frac{\text{diam. (mm) at 40X}}{1}$$

$$\text{Diam. (mm) at 400X} = \frac{40X}{400X} \times \frac{\text{diam. (mm) at 40X}}{1}$$

3. Return to your slide of newspaper letters, and estimate the diameter of the dot of the letter *i* and the length of the letter *i* including the dot.
4. ***Complete item 4 on the laboratory report.***

Application of Microscopy Skills

In this section, you will use the skills and knowledge gained in the preceding portions of the exercise.

Assignment 5

1. Prepare a wet-mount slide of two crossed hairs, one blond and the other brunette. Obtain 1-cm lengths of hair from cooperative classmates.
2. Using the 4X objective, center the crossing point of the hairs in the field and observe. Are both hairs in sharp focus?
3. Examine the crossed hairs at 100X magnification. Are both hairs in sharp focus? Determine which hair is on top by using focusing technique. If you have trouble with this, see your instructor.
4. Examine the crossed hairs at 400X magnification. Are both hairs in focus? Move the crossing point to one side and focus on the blond hair. Using focusing technique, observe surface and optical midsection views of the hair as shown in

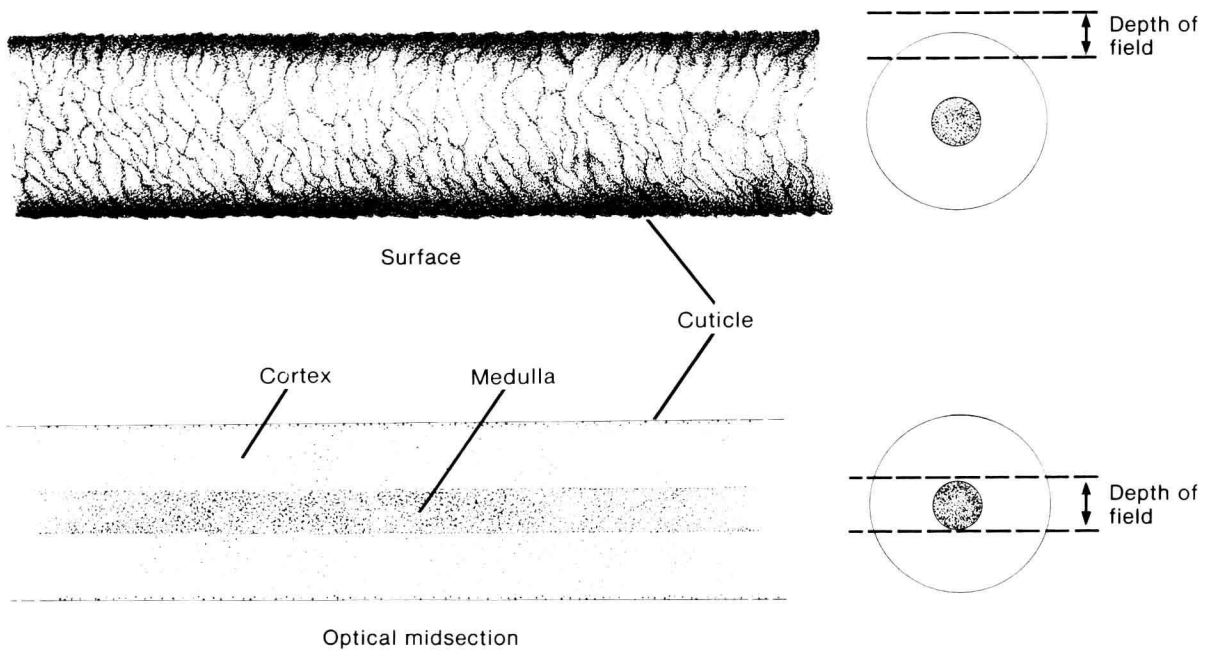

Figure 1.8. Surface and optical midsection views of a human hair, 400 ×

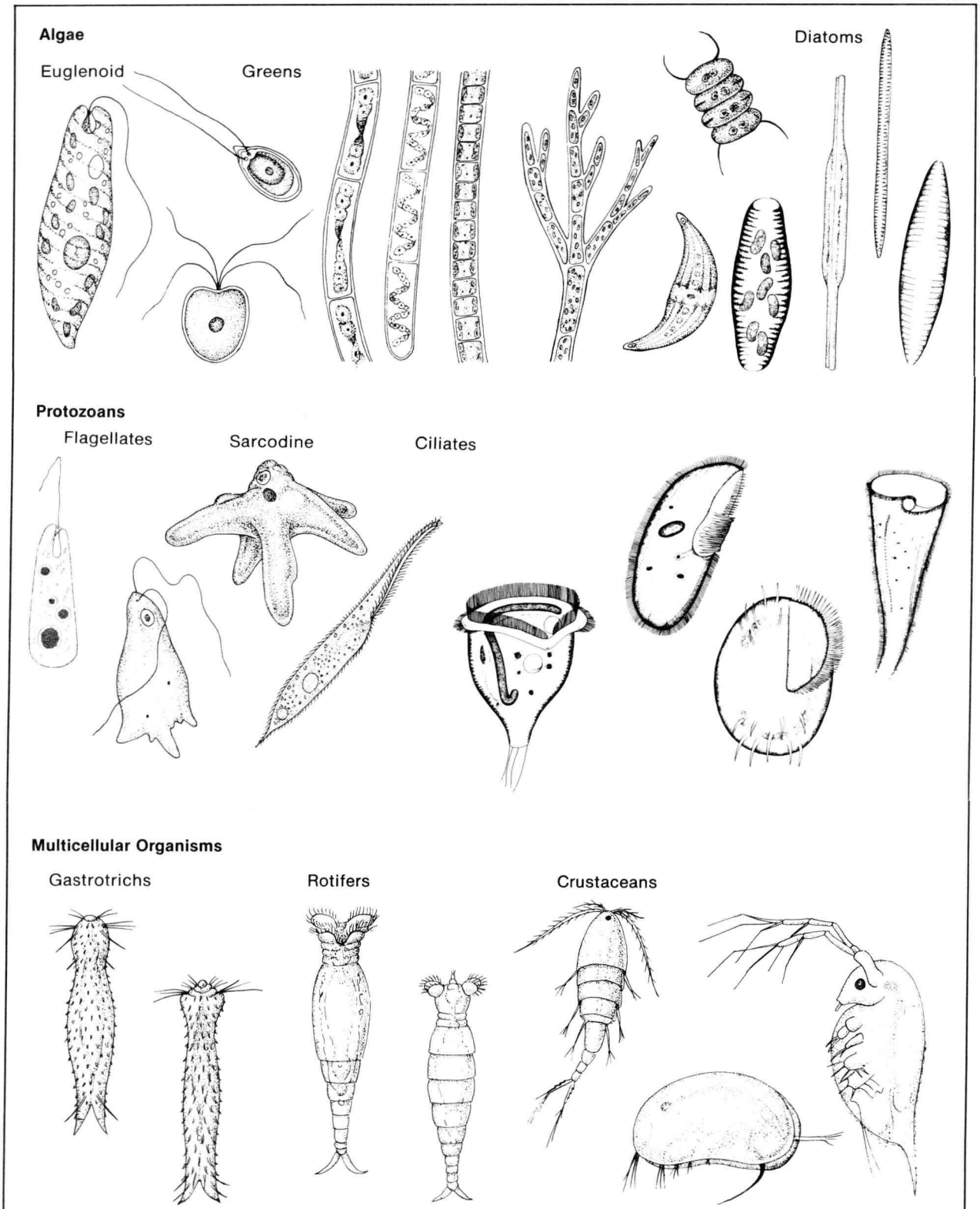

Figure 1.9. Representative pond water organisms