# LABORATORY INVESTIGATIONS IN BIOLOGY



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#### **Preface**

This edition of LABORATORY INVESTIGATIONS IN BIOLOGY offers a laboratory program which supports all of the major areas of biology included in the text MODERN BIOLOGY. The organization and the Investigation numbers in LABORATORY INVESTIGATIONS correlate to the chapters in MODERN BIOLOGY. Yet, LABORATORY INVESTIGATIONS is broad enough in scope to be used with any biology program. When used in conjunction with a text, LABORATORY INVESTIGATIONS presents the subject of biology as a complete laboratory science.

The Investigations in this book employ an inquiry approach which provides the students with the opportunity to add their own experiences and discoveries to the body of knowledge contained in the textbook. This involvement in the discovery of answers acts as a motivational force that gives the students the "feel" for science. In inquiring, hypothesizing, experimenting, observing, and collecting data, the student makes use of the methods of scientific investigation. By writing a laboratory report, the student develops techniques for communicating the findings of a scientific investigation.

The student investigators are guided through their inquiry in several ways. The *Objectives* help the student determine what should be learned in answering the question being investigated. The questions within the procedure guide the students in making observations and interpreting data. As they follow the procedure, they should record data, observations, and answers in a laboratory notebook. Instructions on preparing a laboratory report are given in the *Working As a Biologist* section of this book.

Data may be recorded in various ways.

Charts and graphs completed by the students aid in the understanding of some relationships. Diagrams and illustrations may be drawn in notebooks or on separate pieces of paper during observations or at the end as a summary.

The Summary section asks questions which the student should be able to answer after completing the Procedure and Observations. There are also a variety of test questions which can be used to check what the student has learned. The Investigations on Your Own section provides suggestions for further research. These may be assigned by the teacher or used by the interested student for independent projects.

The organization of the Investigations provides for a maximum degree of flexibility. Each Investigation is an independent laboratory unit. Most of the Investigations are divided into Parts of varying difficulty. This organization provides alternatives in adjusting the laboratory program to the time available, equipment and supplies on hand, and degree of emphasis of each area of the course.

Some use is made of living materials. At the same time, LABORATORY INVESTIGATIONS remains basically teachable. Each Investigation has been classroom tested. Each one is workable in the high school biology laboratory with supplies and equipment usually at hand or readily available. Directions are detailed enough to permit students to obtain meaningful results through their own efforts, with a minimum of teacher supervision.

It is hoped that the laboratory program as presented in LABORATORY INVESTIGATIONS will give students a *personal* experience to make their endeavors meaningful and their learning permanent.

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### **Working As a Biologist**

Biology students generally spend much of their time in the laboratory. It is here that you will discover the structures and functions of living things and their relationships to one another. You will learn to use some of the methods employed by scientists: experimentation, observation, collection and interpretation of data, and drawing of conclusions.

Your techniques and ability to use laboratory apparatus should improve with each investigation you perform. At the same time, your knowledge of living things should increase.

**Conducting a Biological Investigation** 

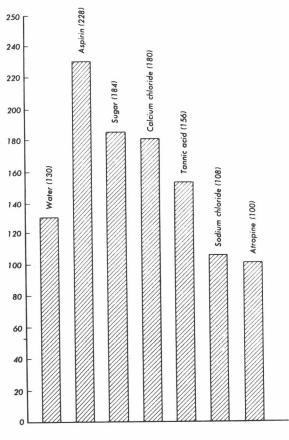
Each Investigation asks a question. By fulfilling the stated objectives you should be able to answer that question. The Investigations are divided into related parts. Your teacher will tell you whether you will be expected to do all of the parts or only certain ones. The length of time required to complete each part will vary. In addition to the assigned Parts, you may also use the *Investigations on Your Own* for special projects or independent research.

**Recording Biological Data** 

Biological data include all observations you make during an investigation. The data you record will be in several forms. In dealing with *structure* (the parts of living things), you may record your observations by drawing them and labeling your drawing. Labels should always be *printed* and have a straight guideline running to the structure being named. The guidelines should be clear and should not cross each other as this would cause confusion. Names of parts are always printed horizontally.

Most of the drawings you will be asked to make are representative drawings. These show the size, shape, and location of structures found in an organism. The drawings you make need not be artistic; few of us are artists. But drawings must always be neat and accurate or they have no scientific value.

Data are often interpreted in graphs. *Bar graphs* may be used to illustrate relative quantities of various trials. For example, an experiment was done in which the effects of various solutions on the heartbeat of a

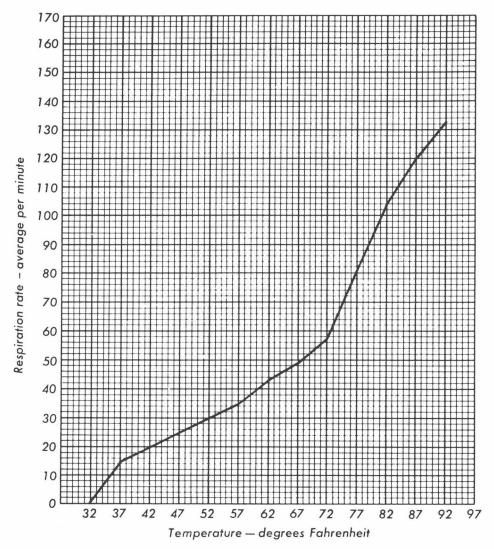


water flea were studied. The natural habitat of the water flea was used as a control. In its normal aquatic habitat the heart of the water flea beat at a rate of 130 times per minute. Data relating to the heartbeat rate when the water flea was placed in other solutions are shown on page 1.

Line graphs are used to show relationships between variables. For example,

the relationship between temperature and breathing rate can be illustrated with a line graph. (See Investigation 34-2.) A class studied the effect of temperature on the respiration rate of the goldfish as indicated by movements of the gill cover. After studying several goldfish and tabulating the movements of the gill cover per minute in a number of goldfish, the class had the following results:

Temperature, F°	32°	37°	42°	47°	52°	57°	62°	67°	72°	77°	82°	87°	92°
Av. movement of gill cover/min.	0	13	20	25	30	35	43	49	58	81	104	120	133



In this experiment, there were two variables, temperature and movements of the gill cover per minute. A line graph of the results shows the effect of temperature on the respiration rate of the goldfish. (See page 2.)

The subject of an Investigation may deal with the *behavior* of an organism (how it reacts and what it does). Sometimes the Investigation will deal with the *function* of an organ. Your observations will usually be in the form of answers to direct questions, data to record, or tables to complete. These answers should be based on facts gained from observation and research (including reading). Guesswork must never appear as scientific data. Although the results of your own experiment are not always what you expect, remember that the experiment that does not work is often just as important to research as the one that does.

#### Preparing a Laboratory Report

Writing a lab report is very different from making observations and recording data in your laboratory manual. In doing a lab report, your teacher may give you an outline or you may develop one yourself. In general, however, it should include:

- I. Title—This should be specific. "Growing Plants" or "Plant Nutrition" are too vague as titles. The title should tell exactly what you are studying. A good title might be: "The Effect of Mineral Deficiency on the Growth of Coleus Plants."
- II. Purpose—This section should state the problem you are investigating. The statement should be simple. For example, "How does the lack of

certain minerals in the soil affect the growth of Coleus plants?" Your entire report must relate to this problem.

- III. Materials and methods—You must tell exactly what you did to prove or disprove the hypothesis. Be sure to have a control for your experiment. In this one, the control plant should be identical except for the mineral content of the soil. Describe the exact experimental procedures and use of minerals in the sequence in which you did them.
- IV. Observations and results—This part forms the basis of support for your analysis and conclusions. It is purely objective. Do not include your interpretation as an observation. You must be sure that what vou observe and measure is actually there. Do not overlook any result. Record all experimental data and observations you make. Give complete description of what hap-Illustrations, pened. drawings, graphs, sketches, and data tables should be used to support the information whenever possible.
- V. Analysis and conclusions—These are subjective in nature. Here you can interpret your results. Tell how your results prove or disprove your original hypothesis. Make each conclusion positive and distinct. You should leave no doubt in the mind of the reader as to why you feel justified in making your conclusions on the basis of the evidence you have collected.



## 1-1

### The Microscope: How Is It Used?

#### **OBJECTIVES**

- To become familiar with the parts of a microscope
- To learn how to use the microscope

#### **MATERIALS**

MICROSCOPE
LENS PAPER
CHEESECLOTH
SLIDE
COVER GLASS
MEDICINE DROPPER
CLEAN CLOTH
TAP WATER
NEWSPRINT (PART 2)
DISSECTING NEEDLE (PART 3)
COTTON THREAD (2 DIFFERENT COLORS)
(PART 3)

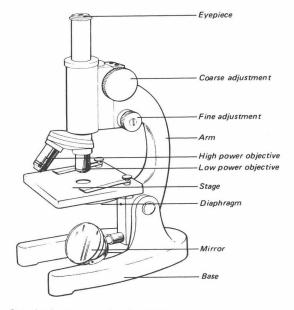
## PART 1 / PREPARING TO USE THE MICROSCOPE

You are about to discover the fascinating world that lies within reach of your microscope. Once you have learned how to use the microscope, there are many living and nonliving things that you will be able to examine in detail.

#### **Procedure and Observations**

You are now ready to set up and use the microscope. Be sure that the arm of the microscope is facing you. The base should be at least five centimeters from the edge of the table. This will keep the microscope from tipping.

Refer to the figure of the microscope as you proceed with this Investigation.



Standard compound microscope

Use a piece of cheesecloth or other soft cloth to wipe off the *stage* and *frame* of the microscope. The stage is where the slide to be viewed is placed. **Do not use a cloth to wipe the lenses.** 

Locate the *mirror* of the microscope. Wipe it with the cheesecloth. Turn the mirror so that the curved surface is facing a good light source. A microscope lamp, ceiling fixture, or daylight will provide sufficient

light. Lighting is extremely important to a clear field of view. Some microscopes may have substage lighting. If this is so, the mirror is not needed. **Never use direct sunlight as a source of light.** This can be harmful to the eyes.

The *eyepiece* is the lens at the top of the microscope. The *objective* lenses are located on a revolving *nosepiece* at the bottom of the microscope tube. Use only special *lens paper* to clean the lenses.

Locate the high-power and low-power objectives. The low-power objective is shorter than the high-power objective. (a) In what other way do you think they differ? Turn the low-power objective until it is directly over the opening of the stage. (b) What magnification is printed on the lens? (c) What power is the eyepiece? Multiply the powers of the two lenses through which you are looking. (d) What is the total low-power magnification?

Locate the *diaphragm* of your microscope. The diaphragm regulates the amount of light passing through the specimen. (e) What type of diaphragm does your microscope have? Open the diaphragm or turn the disk to the largest opening so that the greatest amount of light is admitted. Now you are ready to look through the eyepiece. It is easier to use if both eyes are kept open. (f) What do you see through the eyepiece?

Adjust the mirror and/or diaphragm if you do not see a clear circle of bright light.

You are now prepared to examine some-

thing under the microscope. Before you go on to Part 2 of this Investigation, be sure that you are familiar with all of the parts of the microscope.

#### PART 2 / USING THE MICROSCOPE

In this Part, you will learn how to prepare a wet mount slide. You will then examine it with your microscope. Wet mount slides are slides that *you* prepare for temporary use. Prepared slides are for permanent use. Your teacher may use prepared slides to illustrate certain subjects that you will study.

#### **Procedure and Observations**

Wet mount slides are simple to prepare if you follow the steps outlined below. You will be mounting a newsprint lower case "e."

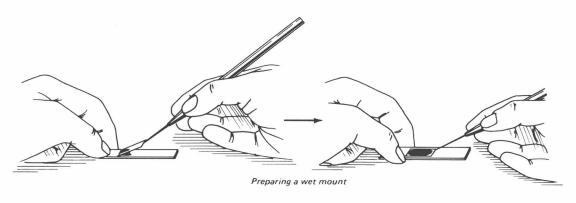
Rinse a microscope slide with water and wipe both sides with a clean soft cloth. Hold the slide by the edges. (a) Why should you do so?

Rinse and dry a cover glass as you did the slide.

Cut out a small piece of newsprint which contains a lower case "e."

Using a medicine dropper, place a drop of tap water in the center of the slide.

Place the newsprint in the drop of water. Lower the cover glass as shown below. This will prevent formation of air bubbles.



Place your temporary wet mount on the stage of the microscope and position it so that the letter "e" is facing you as you would read it. Clip it into place.

Using the *coarse adjustment*, lower the low-power objective as far as it will go without hitting the slide. Be sure to watch the bottom lens as you do this. Never lower the objective while looking through the eyepiece. (b) Why should this not be done?

Look through the eyepiece. Adjust the low-power objective by turning the *coarse adjustment* toward you. The letter "e" will soon come into view. Sharp focus can be achieved by using the *fine adjustment*. If you are having difficulty seeing the "e" clearly, check the positions of the low-power objective and the slide. (c) How would you describe the position of the "e"? Draw a picture of what you see under the microscope.

(d) What happens if you move the slide to the right? (e) What happens if you push the slide away from you? Move the "e" into the exact center of the low-power field. Focus and turn the high-power objective into position. Use the fine adjustment to correct the sharpness of the focus. (f) What is the total magnification of the high-power lens? (g) About how many times was the magnification increased when you changed from low power to high power? (h) How does this change the area of the slide included in the high-power field?

## PART 3 / THE RESOLVING POWER OF THE MICROSCOPE

You will find that the materials you study have depth as well as length and width and that you need to shift your focus as you view in order to see details at various depths. Resolving power is how clear and sharp the magnified image can be made. Both light and resolving power are reduced as magnification is increased.

#### **Procedure and Observations**

Mount 2 different colored cotton fibers across each other in a drop of water on a microscope slide. Cover the slide with a cover glass. Be careful not to trap air under the cover glass. Adjust the diaphragm and bring a fiber into sharp focus with the low power of your microscope. (a) Do the fibers appear uniform in color?

As you examine the fibers, shift the focus by turning the fine adjustment back and forth slowly. (b) Describe any changes in the appearance of the fibers. (c) Explain why these changes occur. (d) How can you determine which fiber is on top when you

look through the microscope?

Move the fibers to the center of the low-power field and shift to the high-power magnification. (e) Compare the brightness of the low-power and high-power fields. Bring the fiber into the sharpest possible focus with the fine adjustment. (f) Is the depth of focus as great with high power as with low power?

#### Summary

(a) Give the function of each of the following parts of the microscope:

Eyepiece Low-power
Stage objective
Stage clips High-power
Diaphragm objective
Mirror Coarse adjustment

Fine adjustment

- (b) Why is good lighting so important?
- (c) How should you focus the low-power objective?
- (d) Which adjustment sharpens the focus?
- (e) Which objective has the greater amount of magnification?
- (f) If you were to purchase a microscope, what would be some features that you would look for?

#### INVESTIGATIONS ON YOUR OWN

- 1. Collect and examine many examples of textile fibers. Some of these fibers include linen, silk, wool, rayon, and nylon. Compare these fibers and observe how they differ. It is interesting to note that fiber identification is important in detection laboratories. Fibers left behind can be a clue in a crime.
- 2. Many simple investigations can be done using materials with which you are familiar. It is interesting to examine different types of paper. You may want to include newsprint, tissue paper, stationery, and wrapping paper in your investigation.

## 2-1

## How Was Spontaneous Generation Disproved?

#### **OBJECTIVES**

- To appreciate the experiments of Spallanzani and Pasteur
- To become familiar with experimental procedures
- To be able to draw conclusions from scientific data

#### **MATERIALS**

MICROSCOPE

SLIDES

COVER GLASSES

6 FLASKS (250 ML)

2 SOLID STOPPERS

2 ONE-HOLE STOPPERS TO FIT FLASKS

8 CM SECTION OF STRAIGHT GLASS TUBING

S-SHAPED GLASS TUBE

**6 Dropping Pipettes** 

PAN FOR PREPARING BROTH

AUTOCLAVE OR PRESSURE COOKER

GLASS-MARKING PENCIL (HEAT RESISTANT)

ALUMINUM FOIL

NUTRIENT BROTH (DEHYDRATED) OR

BEEF BROTH

DISTILLED WATER

## DEMONSTRATION OF BIOGENESIS OF MICROORGANISMS

You are about to study one of the most controversial matters in the history of biology. Theories of spontaneous generation existed long before Redi, Spallanzani, and Pasteur. In this Investigation, you will have the opportunity to repeat some of the work of these experimenters.

#### **Procedure and Observations**

To begin this Investigation, insert a section of straight glass tubing and an S-shaped glass tubing through one-hole stoppers as shown in flasks E and F on page 10.

Prepare 1,000 ml (1 liter) of nutrient broth as directed on the media bottle. Be sure that all ingredients in the broth are dissolved. Mark the 6 flasks as A through F, using a heat-resistant glass-marking pencil. Divide the broth equally among the 6 flasks. Prepare the flasks as shown in the diagram on page 10.

Flask A will remain unsterilized and will be left open.

Flask B will remain unsterilized and will be stoppered.

Flask C will remain open after sterilization.

Flask D will be stoppered after sterilization.

Flask E will be stoppered with the straight glass tube after sterilization.

Flask F will be stoppered with the S-shaped glass tube after sterilization.

Prepare flasks. A and B and set aside. Cover the mouths of C, D, E, and F with aluminum foil. Put these flasks and the stoppers for D, E, and F in the autoclave or pressure cooker. Sterilize at 15 pounds for 15 minutes. (a) Could this Investigation be done without sterilization?



A Unsterilized, open



B Unsterilized, stoppered



C Sterilized, open



D Sterilized, stoppered



E Sterilized, straight tube



F Sterilized, S-shaped tube

After sterilization, allow the flasks and stoppers to cool in the sterilizer. Remove the aluminum foil from the flasks and insert the stoppers. (b) Why should care be taken not to touch the lower part of the stoppers?

Place all the flasks in the incubator at 35–37°C for 72 hours. (c) Why is this necessary?

Following the incubation period, examine all of the flasks. Using a clean pipette for each flask, put a drop of broth on a microscope slide. Add a cover glass and examine

under high power. (d) Why should high power be used?

If microorganisms are present, they will appear as small moving bodies. In some cases, it may be necessary to darken the field by closing down the diaphragm. (e) What do you see when the material from flask A is examined?

In a table like the one shown, fill in your observations for each of the 6 flasks. DO NOT WRITE IN THIS BOOK. Each of the changes in the table indicates activity of microorganisms in the broth.

Flask	A	В	С	D	E	F
Presence of microorganisms						
Color						
Degree of turbidity (cloudiness)	T.					
Odor						
Surface pellicle (film)						
Sediment					7	

Supporters of spontaneous generation and of biogenesis would interpret the results of this experiment differently. (f) How would the "spontaneous generationists" explain the presence of microorganisms? (g) How would Pasteur have viewed the same data? (h) What was meant by an "active principle"?

#### Summary

On a separate piece of paper, construct a table like the one below. DO NOT WRITE IN THIS BOOK.

In your table, summarize your findings from this Investigation by evaluating each of the following theories. Use the following key to interpret your findings.

- + evidence exists to prove
- ? possibility, but evidence is lacking
- evidence to disprove
- 0 does not apply

#### INVESTIGATIONS ON YOUR OWN

- Construct an original investigation that disproves the theory of spontaneous generation. This investigation should be simple and should be done with easily available materials.
- 2. Another theory related to the theory of spontaneous generation was plant growth through the conversion of soil into plant substances. One of the most famous experiments to disprove this theory was conducted by J. B. van Helmont in the 17th century. You may want to re-create this experiment by substituting a sunflower seedling for the willow tree. Draw your own conclusions after completing the experiment.

Flasks	Α	В	С	D	Е	F
Living organisms were in the broth before incubation						
Broth substances were changed to living organisms by spontaneous generation						
Heat destroyed an "active principle" necessary for spontaneous generation						
Air is necessary for the growth of organisms						
Organisms present in the broth came from the air						
Organisms will not appear if neither the broth nor the air entering the flask contains organisms						