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EXERCISE 1

Light Microscopy

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The light microscope is capable of extending our ability to “see detail” by 1000 times, so that objects as small as 0.1 micrometer (μm) or 100 nanometers (nm) can be seen. The transmission electron microscope extends this viewing capability to objects as small as 0.5 nm in diameter, enabling us to see objects that are $\frac{1}{200,000}$ th the size of those that can be seen by the human eye. Without microscopes, our understanding of the structure and function of cells and tissues would be severely limited.

The ability of the microscope to reveal the structure of small objects, however, is not so much a function of its ability to magnify as its ability to distinguish detail. Merely magnifying an object, without increasing the amount of detail seen is of little value to the observer. The ability to see detail is called **resolving power** and depends on the wavelength (λ) of light used and a value called the **numerical aperture (NA)**, an important characteristic that determines how much light will enter the lens. In its simplest form, resolving power, or **resolution**, may be expressed by the formula

$$\text{RP} = \frac{\lambda}{2 \times \text{NA}} \quad \begin{array}{l} \lambda = \text{wavelength of light used} \\ \text{NA} = \text{numerical aperture} \end{array}$$

Under normal viewing conditions, resolution is *increased* by decreasing the wavelength of the light source. For example, if you use a green filter that permits a wavelength of 500 nm to pass through a microscope lens having a numerical aperture of 1, then the resolving power would be $500 \text{ nm} / 2 \times 1$ or 250 nm. This means that two objects that are 250 nm or farther apart would be seen as distinct objects; if closer than 250 nm, they would appear very fuzzy or as one object.

If you use blue light, or a blue filter that provides light at a wavelength of 400 nm and a lens having a NA of 1, the resolving power would be equal to $400 \text{ nm} / 2 \times 1$ or 200 nm. The two objects observed under these conditions could be 50 nm closer together and still be seen as separate objects.

Knowing the significance of the wavelength of light to the ability to distinguish detail, you can appreciate the role of electron microscopes and micro-

scopes utilizing ultraviolet light in elucidating the structure and function relationships of cells and sub-cellular organelles.

A. PARTS OF A COMPOUND MICROSCOPE

Your microscope may have all or most of the features described below. Referring to Fig. 1-1, locate the following features of the microscope available in your laboratory.

1. Ocular Lens

The **oculars** are the lenses you look through. If there is only one ocular, you are using a **monocular** microscope; if there are two, it is a **binocular** microscope. In many binocular microscopes, the oculars can be adjusted to compensate for differences in distance between your eyes (**interpupillary adjustment**). One of the oculars may have a knurled adjustment mechanism for moving it in and out to compensate for focusing disabilities between each eye. Your instructor will describe how this is done. Oculars on different microscopes may have different magnifications. You may have to remove the ocular from its holder to determine its magnification. What is the magnification stamped on the housing of the oculars on your microscope?

The ocular contains a series of several magnifying lenses and may also include an **ocular micrometer** (a scale for measuring objects) and a pointer (to point out objects to your instructor or other students).

2. Objective Lens

Attached to a rotating nosepiece, or turret, at the base of the body tube are a group of three or four **objectives**. Rotate the nosepiece and notice that a “click” is heard as each objective comes into position.

The magnifying lenses of the objectives focus light that comes from the specimen and passes it up the body tube and through the oculars.

Each objective has numbers stamped on it. One of these numbers identifies the magnification of the objective (e.g., 43 X). What are the magnifications of each of the objectives on your microscope?

The **total magnification** is calculated by multiplying the magnification of the ocular and objective lenses on the microscope being used. In Table 1-1 calculate the total magnification for each ocular/objective combination on your microscope.

TABLE 1-1
Calculation of total magnification for various ocular/objective combinations.

Ocular	×	Objective	=	Total magnification
_____		_____		_____
_____		_____		_____
_____		_____		_____
_____		_____		_____

Note: Objective lenses are usually named according to their magnifying power, as follows:

- scanning power— 4 X
- low power— 10 X
- high or high dry power— 43 X
- oil immersion— 93 X

A second set of numbers, usually given as a decimal, represents the numerical aperture for that lens; the abbreviation NA may precede the number. In Table 1-2 list the magnification and numerical aperture for each objective on your microscope.

TABLE 1-2
Numerical aperture and magnification for various objectives.

Magnification of objective	Numerical aperture (NA)
_____	_____
_____	_____
_____	_____
_____	_____

3. Body Tube

Light travels from the objectives through a series of magnifying lenses in the body tube to the ocular. In some microscopes, the body tube is straight. In

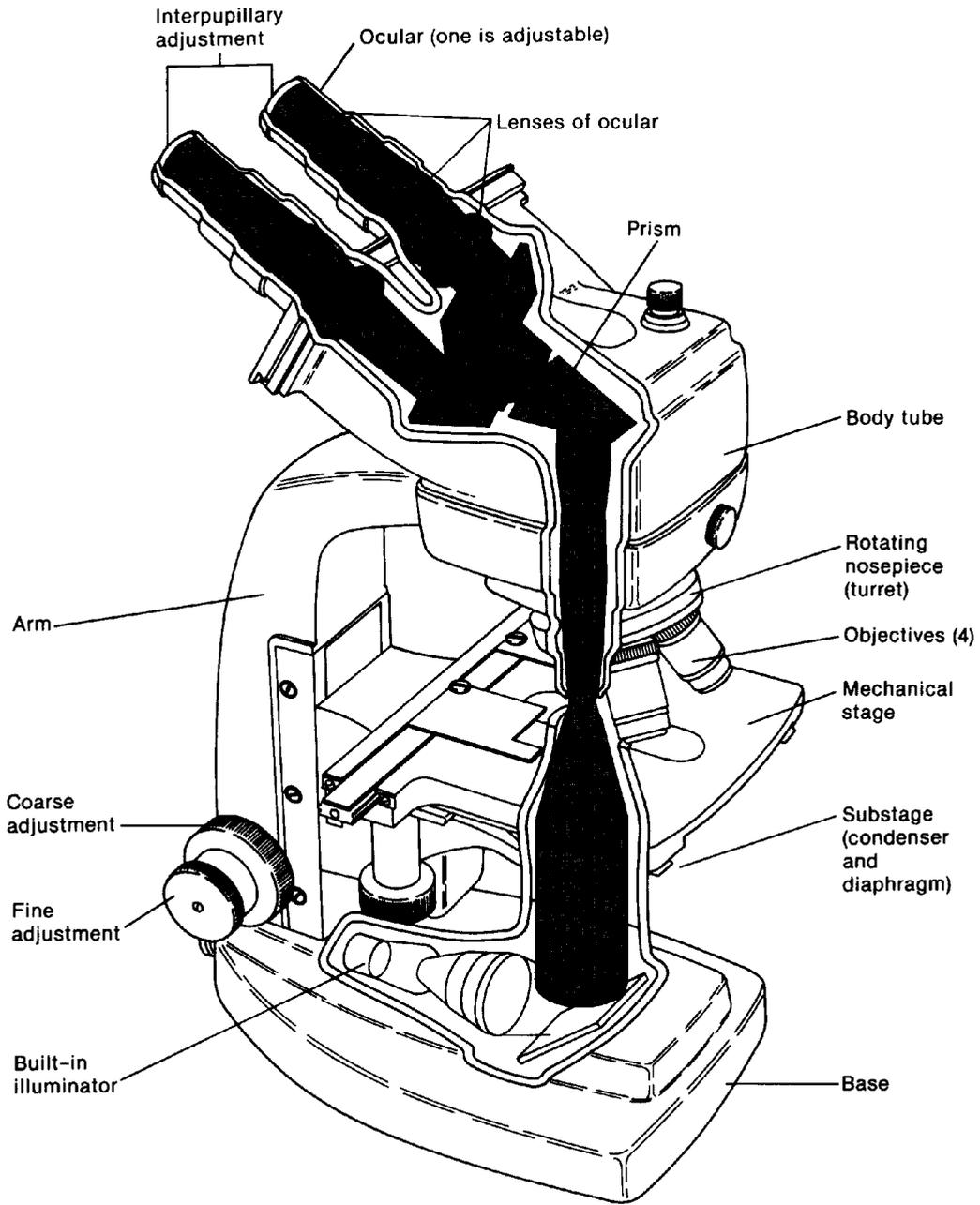


FIG. 1-1
Bausch and Lomb binocular microscope sectioned to show pathway of light from illuminator through various lenses and prisms.

others, the oculars are held at an angle, as in Fig. 1-1, and the body tube contains a prism that bends the light rays coming through the objectives so they will be transmitted through the oculars.

With some compound microscopes, loosening a lock screw allows you to rotate the body tube 180°. What is the advantage of being able to turn the body tube?

4. Stage

The surface or platform on which the microscope slide is placed is the **stage**. Note the opening (**stage aperture**) in the center of the stage. On some micro-

scopes, the stage may be stationary and have clips to hold the slide in place. On other microscopes, the stage can be moved and is therefore called a **mechanical stage**. Movement is controlled by two knobs located on top, on the side, or on the bottom of the stage. Note the **horizontal and vertical scales** on the mechanical stage. What is the function of these scales?

How are slides held in position on a mechanical stage?

5. Substage

The area beneath the stage is the substage and may be occupied by one, or both, of the following:

a. Diaphragm

The diaphragm regulates the *amount* of light passing from the light source through the specimen and through the lens system of the microscope. By properly adjusting the diaphragm you provide better contrast with the surrounding medium, thus greatly improving your image of the specimen. The diaphragm may be either

1. An **annular** type of diaphragm consisting of a circular plate with holes of different diameters. This plate is rotated so that the various holes may be positioned in the light path to regulate the amount of light passing from the light source through the object under observation.

2. An **iris** type of diaphragm that consists of a series of overlapping thin metal plates. A lever projecting from the side of the diaphragm opens and closes these plates to regulate the amount of light entering the microscope.

What type of diaphragm does your microscope have?

b. Condenser

The condenser consists of a series of lenses that focus light onto the specimen. Movement of the condenser is regulated by a knob at its side, or a lever projecting from the condenser housing. By properly adjusting the condenser you greatly improve your observation of the specimen.

Attached to the bottom of the condenser may be a filter holder, which normally contains a blue filter. Why would you use a blue filter, instead of a green or red filter, when making microscopic observations? Indeed, why use a filter at all?

6. The Light Source

Your microscope may have an attached mirror or a built-in illuminator. If your microscope uses a mirror, one surface is usually concave and the other is flat. The flat side of the mirror is normally used with the scanning and low power objectives and the concave mirror with higher power objectives. The light source for the mirror is usually a lamp. Natural light may be used, but it is not preferred because the light's intensity will vary greatly, depending on the source of light in your laboratory.

In most compound microscopes, the illuminator is built into the base of the microscope and controlled by an on/off switch. You can control the amount of light entering the specimen by adjusting the diaphragm. You can also control the light intensity by adjusting a transformer attached to the illuminator, whose knob can be turned to regulate the voltage to the light bulb. Use low or medium transformer settings for most microscopic observations. You will need a higher setting when using the oil immersion lens. Why?

7. Focusing

You can focus your microscope by using the coarse and fine adjustment knobs that raise or lower either

the body tube or the stage, depending on the type of microscope you are using.

With the low-power objective in position, rotate the coarse adjustment knob one half turn clockwise. Do the same with the fine adjustment knob. Based upon your observations, why should you not use the coarse adjustment knob for focusing when the high-power objective or oil immersion objective is in position?

8. Eyeglasses and Microscope Usage

Should you wear your eyeglasses when using a microscope? The answer to this question is qualified. If you are near- or farsighted, you need not wear your glasses for microscopic observations. The adjustments made in focusing the microscope will compensate for these eye problems. On the other hand, wear your glasses if you have astigmatism (a defect in the eye's refractive surface), since this problem is not corrected by the lenses of the microscope.

In either case, when using a monocular microscope, you should keep both eyes open, despite a tendency to close one eye. Eye strain will develop if you do this for any length of time.

B. PROPER USE OF MICROSCOPES

Before using your microscope, thoroughly clean the oculars and objectives using lens paper in a circular motion to prevent scratching. When using the microscope, keep eyelashes from touching the ocular. Oil from the lashes will adhere to the ocular lenses smearing them. When using salt solutions or other harsh chemicals to prepare wet mounts, thoroughly clean the oculars and objectives, stage, and microscope slides after use to prevent damage to the microscope.

Despite its sturdy appearance a microscope is a delicate, precision instrument. It should be handled carefully and with common sense. The following suggestions will help you avoid some common mishaps that occur when using a microscope.

1. To avoid dropping a microscope, banging it against a laboratory bench, or having the oculars fall out,

- a. Carry the microscope upright using both hands.
- b. Place the microscope away from the edge of the bench, particularly when not in use.
- c. Move power cords out of the way, so that you can't trip on them and pull the microscope or transformer down.

2. To avoid breaking a coverslip and/or microscope slide by an objective,

- a. First locate the specimen using the low-power objective, and then switch to the higher-power objectives.
- b. Never focus the high-power objective with the coarse adjustment knob and never use these lenses when examining thick specimens or whole mounts of specimens.

3. To avoid mechanical difficulties with various parts of a microscope,

- a. Never force microscope parts to work.
- b. When changing the bulb in the built-in illuminator, never force it, since it might shatter in your fingers.
- c. Never try to dismantle the microscope.

C. USING A COMPOUND MICROSCOPE

1. Focusing

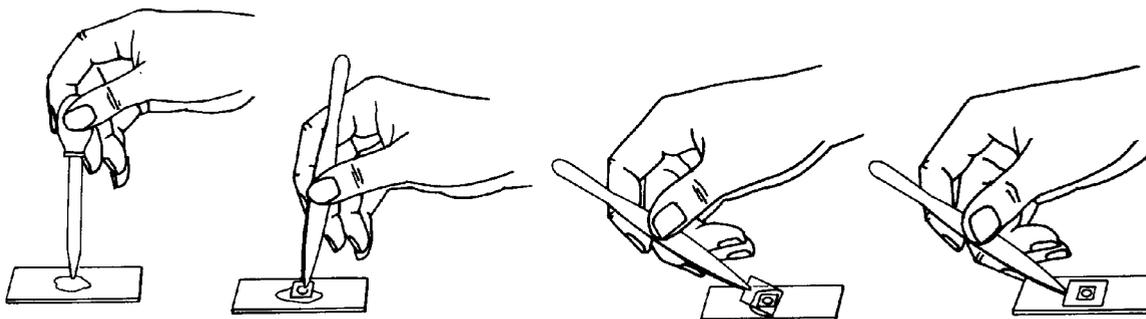
1. Cut out a lower case letter *e* from a newspaper or other printed page. Clean a microscope slide and prepare a "wet mount" of the letter, using the procedure described in Fig. 1-2. Put the scanning (4×) or low-power (10×) objective in position and then place the slide on the stage in its normal viewing position.

2. Clean the oculars and objectives using lens paper.

3. Turn on the illuminator and open the diaphragm fully. If there is a condenser, position it as high as it will go, so that the top lens of the condenser unit is level with the stage aperture.

4. Center the specimen over the stage aperture.

5. Position the scanning objective (4×) as close to the slide as possible and then, while looking through the oculars, use the coarse adjustment knob to back off slowly until the specimen comes into focus.



A. Add a drop of water to a slide.

B. Place the specimen in the water.

C. Place the edge of a coverslip on the slide so that it touches the edge of the water.

D. Slowly lower the coverslip to prevent forming and trapping air bubbles.

FIG. 1-2
Preparation of a wet mount slide.

6. Using the diaphragm (and/or adjustment of the transformer voltage), readjust the light intensity as necessary and again center the specimen by moving the slide.

7. Switch from the scanning lens to the low-power objective ($10\times$). Make certain the objective “clicks” into position. If the specimen stays in focus, your microscope is **parfocal**. You can sharpen the focus by small adjustments of the fine adjustment knob.

If your microscope is not in focus after changing objectives, you may have to use the coarse adjustment knob followed by the fine adjustment knob. But remember, do not do this with the high-power or oil immersion objectives in position. Ask your instructor for help if you have difficulty focusing your microscope.

Recenter the specimen, adjust the diaphragm, and adjust the position of the condenser to increase the contrast of the specimen.

8. Switch to the high-power objective ($43\times$) and adjust the focus using the fine adjustment knob.

These are the procedures usually used when examining a wet mount or a commercially prepared microscope slide. Always make your preparation using clean microscope slides. Always proceed from the lowest-power to the highest-power objectives, making minor corrections in focus and light as necessary. Learn to “fine tune” your microscope.

2. The Microscopic Image

The image you view in the microscope is affected by several factors: the orientation of the image, total magnification, the size and brightness of the field of

view, the plane of focus, the depth of focus, and the contrast of the materials being examined.

a. Orientation of the Image

Hold the slide you made of the letter *e* so that the letter is in a normal reading position. Then, place it on the stage in the same position and examine it with the low-power objective. What difference is there, if any, in the way the image is oriented when viewed through the oculars as compared to looking at it directly with your eyes?

While looking through the microscope, attempt to make the image move to the right. In which direction did you have to move the slide?

Try to move the image up (away from you). Which way did you have to move the slide?

In what direction do you have to move the letter to make the image move right then up?

There will be times when you will want to show someone something of interest in the **field of view**. One way to do this is to describe its approximate location by referring to the field of view as a clock. Thus, you could tell them to “look at three o’clock,” or “look just off center toward nine o’clock,” and so

forth. Alternately, some microscopes have what appears to be a thin black line cutting across the field. This is a **pointer** that has been added to the ocular of your microscope so you can point out something by moving the object under observation to the end of the pointer.

b. Brightness of the Field of View and Working Distance

Examine your slide starting with the lowest-power objective and progressing to the highest-power objective. Describe any changes in the brightness of the field when you change objectives.

In Fig. 1-3 shade in the appropriate circles to correspond with any change in brightness you observed. When the object on your slide is in focus for each objective, the **working distance** between the slide and the objective lens decreases as the objective magnification increases. Of what value is such information to you?

If you know the diameter of the field for each magnification, you can use this information to estimate the size of object you are examining. To determine the diameter of the field, place a transparent millimeter rule on the stage, focus on the rule, and measure the diameter of the field for the scanning and low-power objectives. It will be very difficult to measure the high-power and oil immersion fields, but you can get a good approximation using the following formula, where

- D_L = diameter of the field at a *lower* magnification
- D_H = diameter of the field at the *higher* magnification
- X_L = magnification of the *lower* power objective lens
- X_H = magnification of the *higher* power objective lens

Thus

$$\frac{D_H}{D_L} = \frac{X_L}{X_H} \quad \text{or} \quad D_H = \frac{D_L \times X_L}{X_H}$$

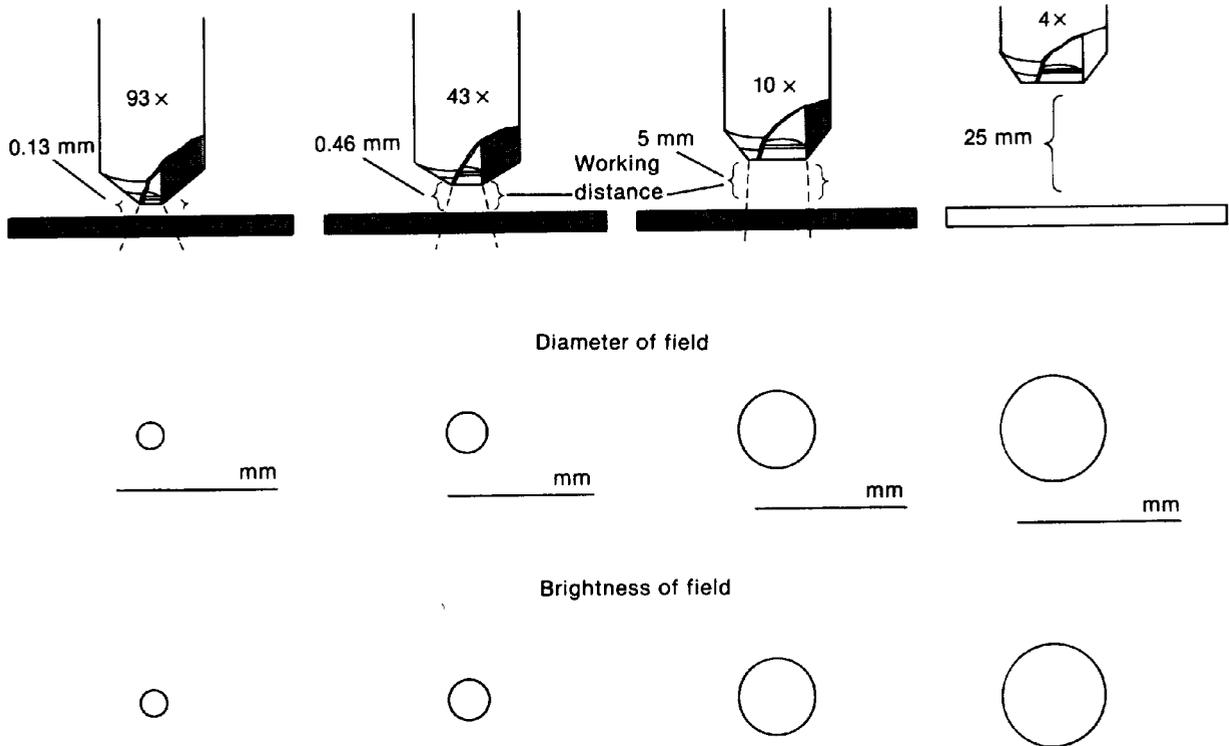


FIG. 1-3 Relationships between working distance, diameter, and brightness of field.

Insert the appropriate values in the formula and determine the diameters of the high power and oil immersion objective fields of your microscope. Record your data in Fig. 1-3.

c. Depth of Focus

Just like the human eye, the lenses of your microscope provide a limited **depth of focus**. This means that only part of the object will be in sharp focus, while areas above and below that part will be slightly out of focus or not in focus at all.

To become familiar with the concept of depth of focus, take your metric rule and, pointing it lengthwise away from you, hold it about 30 cm (12 inches) in front of you and about 7.5 cm (3 inches) below your eyes. Looking down the length of the rule, focus your eyes on the 10-cm mark. When this mark is sharply in focus, what numbers above and below the 10-cm mark are also in focus? What then is the depth of focus for your eyes?

In practice, you will find that as magnification *increases*, the depth of focus *decreases*. You will have to learn to constantly use the fine adjustment knob when the higher power objective is in position. This will help you to determine something about the three-dimensional shapes of the objects under observation.

To visualize three-dimensional form and the concept of depth of focus, place a small strand of your hair and a white and a yellow thread across each other on a microscope slide. Add a drop of water and a coverslip. Using the scanning objective (4 X), focus where the strands of hair intersect and determine the depth of focus at this magnification.

Change to the low-power objective (10 X). Describe any changes in the depth of focus.

Switch to the high-power objective (43 X) and describe any changes in the depth of focus.

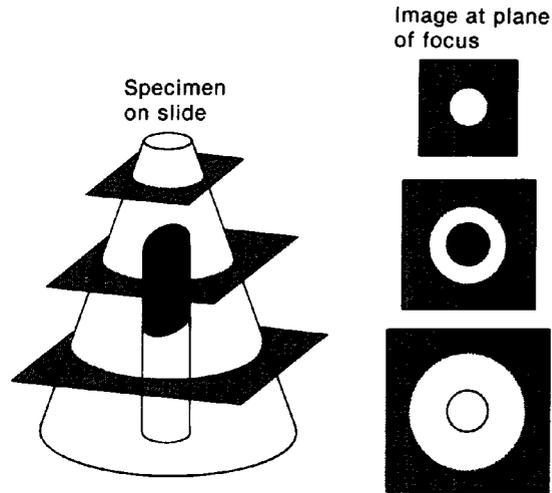


FIG. 1-4
Determining three-dimensional image through "optical" sectioning.

At this higher magnification, it is difficult but not impossible to determine three-dimensional form. You can do this by building a series of **optical sections** in your mind as you focus through the specimen. Fig. 1-4 demonstrates how this is done.

Try to determine the three-dimensional structure of your preparation at high power by making and visualizing a series of optical sections. Begin by focusing on the surface of the top thread and working through to the lower surface of the bottom thread or hair.

d. Contrast

Even with sufficient magnification and resolution, you can only visualize an object under a microscope if there is sufficient contrast between the object and its surroundings or between the various parts of the object.

Cells or subcellular structures may contain naturally occurring pigments (e.g., chlorophyll in chloroplasts, hemoglobin in red blood cells) that provide contrast and make these structures visible. Frequently, however, cells and parts of cells are highly translucent. One way to improve contrast is to use dyes or stains that bind to or are taken up by various subcellular structures and thus absorb enough light to provide the necessary contrast. In addition to staining, or in combination with it, you can improve image contrast by regulating the opening of the diaphragm. This deflects the light rays from edges of the diaphragm and causes them to enter the specimen at an angle. Such scattering of light makes the specimen look darker, since some of the light takes longer to reach the eyes.

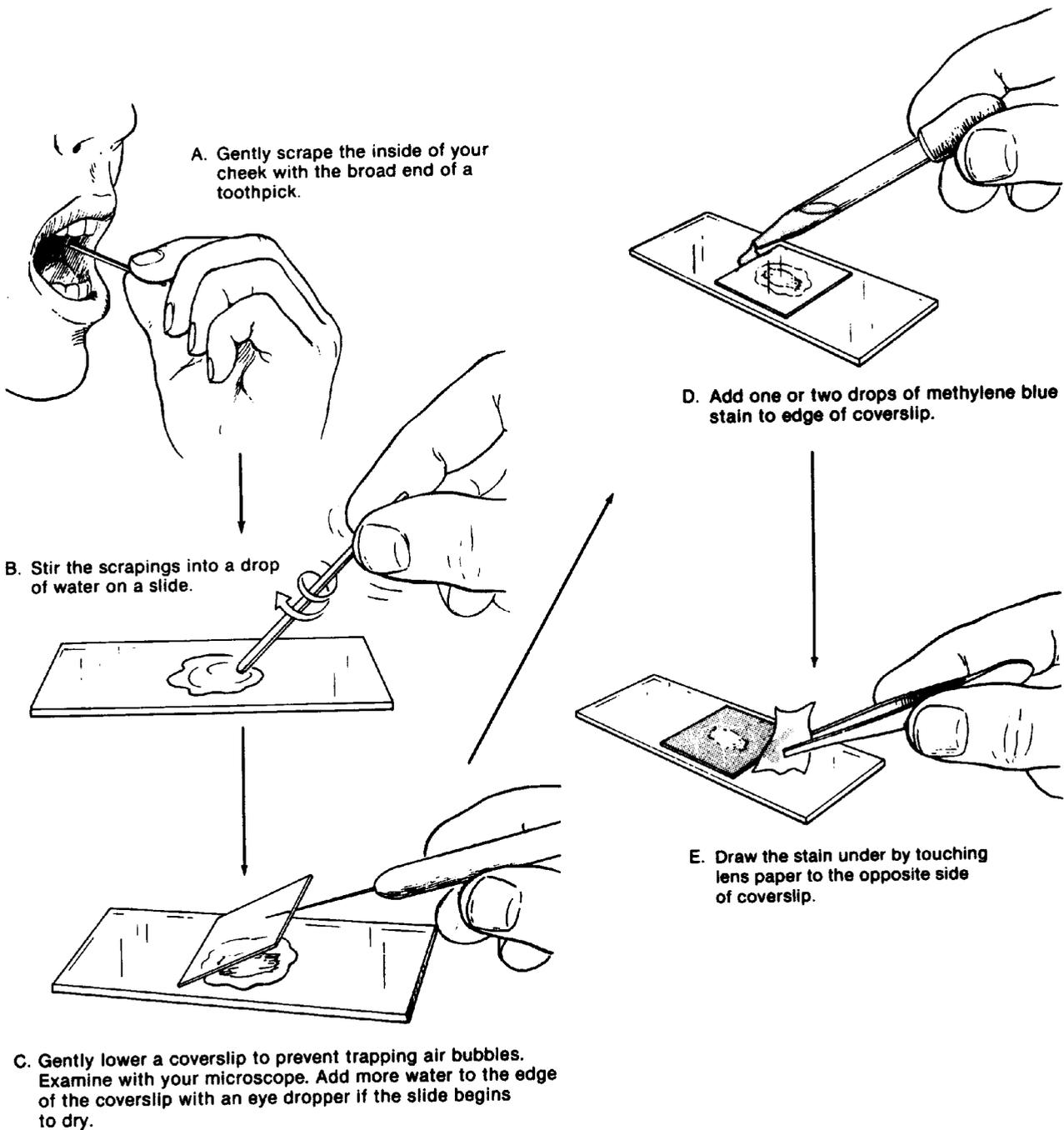


FIG. 1-5
Staining cells to improve image contrast.

Following the instructions in Fig. 1-5A-C, examine cells obtained from the inner epithelial lining of your cheek. Try to determine something of their structure by adjusting the diaphragm and the condenser. Add a drop of methylene blue stain to the edge of the coverslip and draw it under as shown in Figure 1-5D, E. Describe any changes in contrast, or visibility of the structures, in the cell.

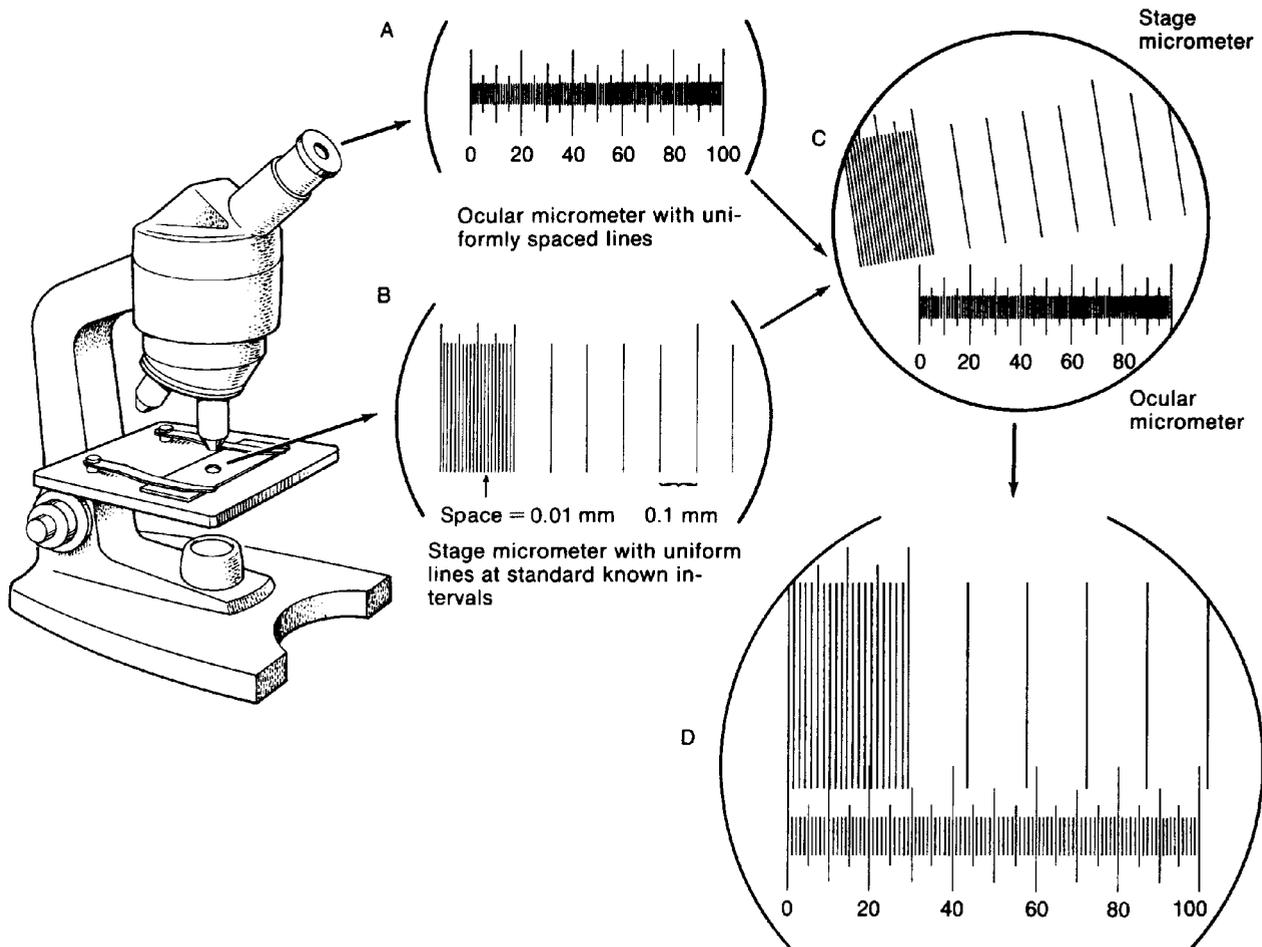


FIG. 1-6
Using an ocular micrometer to determine the size of microscopic objects.

e. Measurement of Microscopic Specimens

Recall that you earlier determined the diameter of the field of view of the various objective lenses on your microscope and, comparing the size of the specimen with the diameter, you obtained a rough estimate of the size of the object.

A more precise method involves using an **ocular micrometer**, a small glass disc on which uniformly spaced lines of *unknown distance* are etched. The ocular micrometer is inserted into the ocular of the microscope and then calibrated against a **stage micrometer**, which has uniformly spaced lines of *known distances* (Fig. 1-6). To calibrate the ocular micrometer use the following procedure:

1. If you were to observe the stage micrometer without the ocular micrometer in place, it would appear as shown in Fig. 1-6B. If you were to observe

the stage micrometer with the ocular micrometer in place, it would appear as shown in Fig. 1-6C.

2. Turn the ocular in the body tube until the lines of the ocular micrometer are parallel with those of the stage micrometer. Match the lines at the left edges of the two micrometers by moving the stage micrometer (Fig. 1-6D).

3. Calculate the actual distance in micrometers (μm) between the lines of the ocular micrometer by observing how many spaces of the stage micrometer are included within a given number of spaces on the ocular micrometer. Since the smallest space on the stage micrometer equals 0.01 millimeter (mm), you can calibrate the ocular micrometer using the following:

$$\begin{aligned} 10 \text{ spaces on ocular micrometer} \\ = X \text{ spaces on stage micrometer.} \end{aligned}$$

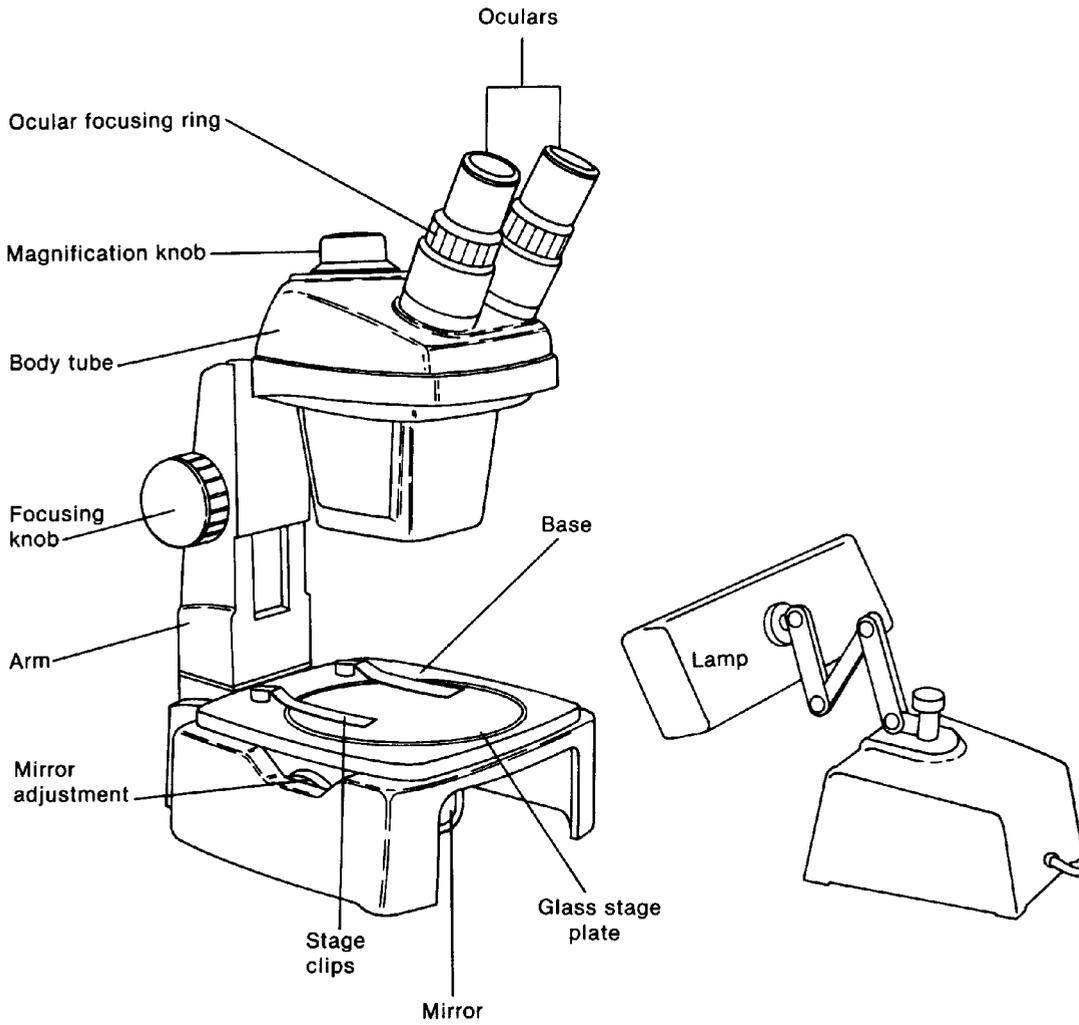


FIG. 1-7
Parts of a stereoscopic (dissecting) microscope.

Since the smallest space on a stage micrometer = 0.01 mm, then

$$10 \text{ spaces on the "ocular"} = X \text{ spaces on the "stage"} \times 0.01 \text{ mm}$$

and

$$1 \text{ space on the "ocular"} = \frac{X \text{ spaces on the "stage"} \times 0.01 \text{ mm}}{10}$$

Example: If 10 spaces on the "ocular" = 6 spaces on the "stage" then

$$1 \text{ ocular space} = \frac{6 \times 0.01 \text{ mm}}{10}$$

Of course, the numerical value obtained holds only for the specific objective-ocular combina-

tion used. Each time the objective or ocular lens is changed, the ocular micrometer will have to be recalibrated.

D. USE AND CARE OF THE STEREOSCOPIC (DISSECTING) MICROSCOPE

The stereoscopic dissecting microscope shown in Fig. 1-7 has two distinct advantages over the compound microscope: (1) it enables you to examine objects that are too large or too thick to be seen with the higher magnifications of the compound microscope and (2) it gives you a three-dimensional view of the specimen.

The stereoscopic microscope is often used when dissecting specimens. The light source may be reflected from an illuminator above the specimen or,

on some microscopes, transmitted through the specimen from a mirror below the stage. The choice of the light source depends upon whether the specimen is transparent or opaque.

Using your dissecting microscope, examine your fingers or some other opaque object. Adjust the oculars for interpupillary distance and focus as previously for the compound microscope (Part A-1). Change the magnification using the magnification knob on the top of the body tube. On other stereoscopic microscopes, the magnification is varied by switching ocular lenses, as with the compound microscope. How does the movement of the image compare to that of the compound microscope (Part C-2 of this exercise)?

How do you adjust the brightness of the field?

Examine the previously prepared slide of the crossed threads or hair. First use reflected light from the mirror, then use transmitted light from a lamp. Describe any advantage of one type of lighting over the other.

E. STUDY OF POND WATER

In your laboratory work, many observations made by the microscope will be on living organisms or on tissues or parts of organisms that you will want to keep alive. To allow them to dry out would greatly distort them, to say nothing of the effect death would have on a study of their movements. To observe living material prepare a wet mount of a drop of pond water as shown in Fig. 1-2.

Excess water under the coverslip can be soaked up by carefully placing a piece of paper toweling to the edge of the coverslip. However, if your preparation begins to dry out while under observation, add one drop of water at the edge of the coverslip.

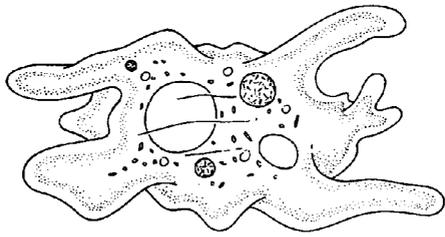
Under low power and with reduced light, survey the drop of pond water. Identify as many of the organisms as you can. Carefully study their differences in structure and their method of movement. Figs. 1-8, 1-9, 1-10, and 1-11 should help you identify what you see.

Prepare additional wet mounts by taking samples from different parts of the jar of pond water. Do not be too hasty in discarding a slide because you don't find any microorganisms; a systematic survey of the preparation is often necessary to locate the organisms. Why do the organisms often accumulate at the edge of the coverslip?

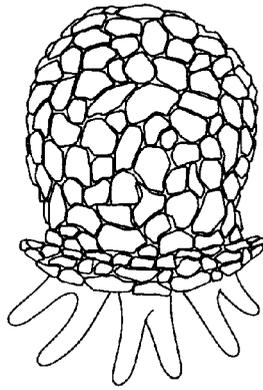
To identify the smaller organisms, you may have to use the high-power objective. When your work is completed, clean and dry any slides and coverslips used. Wipe the lenses of the microscope with lens paper, clean the stage, and return the microscope to the cabinet.

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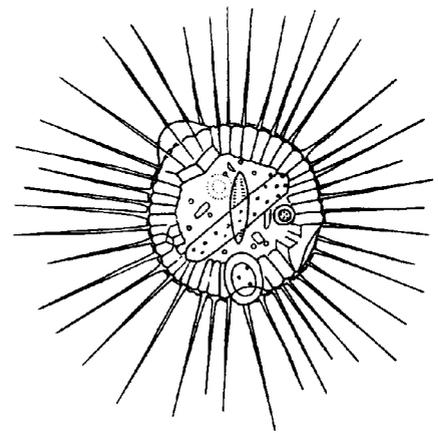
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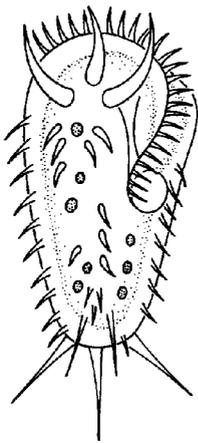
Amoeba



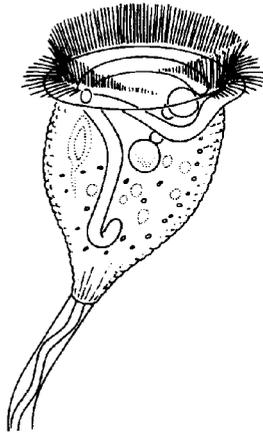
Diffugia



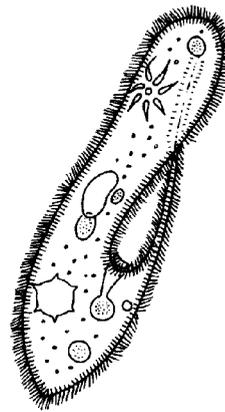
Actinosphaerium



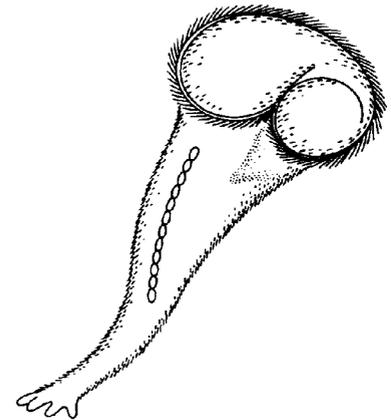
Stylonychia



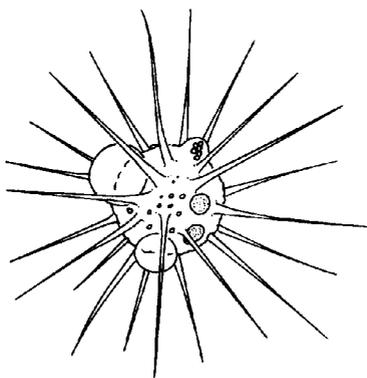
Vorticella



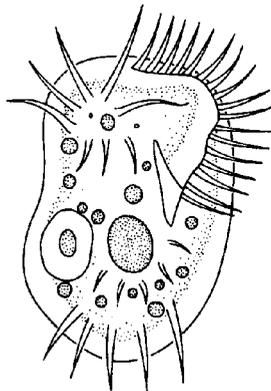
Paramecium



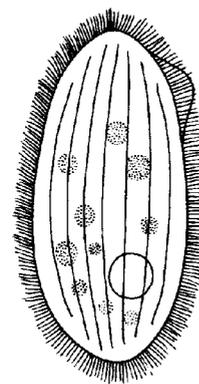
Stentor



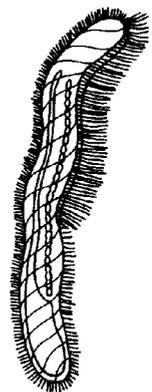
Actinophrys



Euplotes



Colpidium



Spirostomum

FIG. 1-8
Protozoans commonly found in pond water.

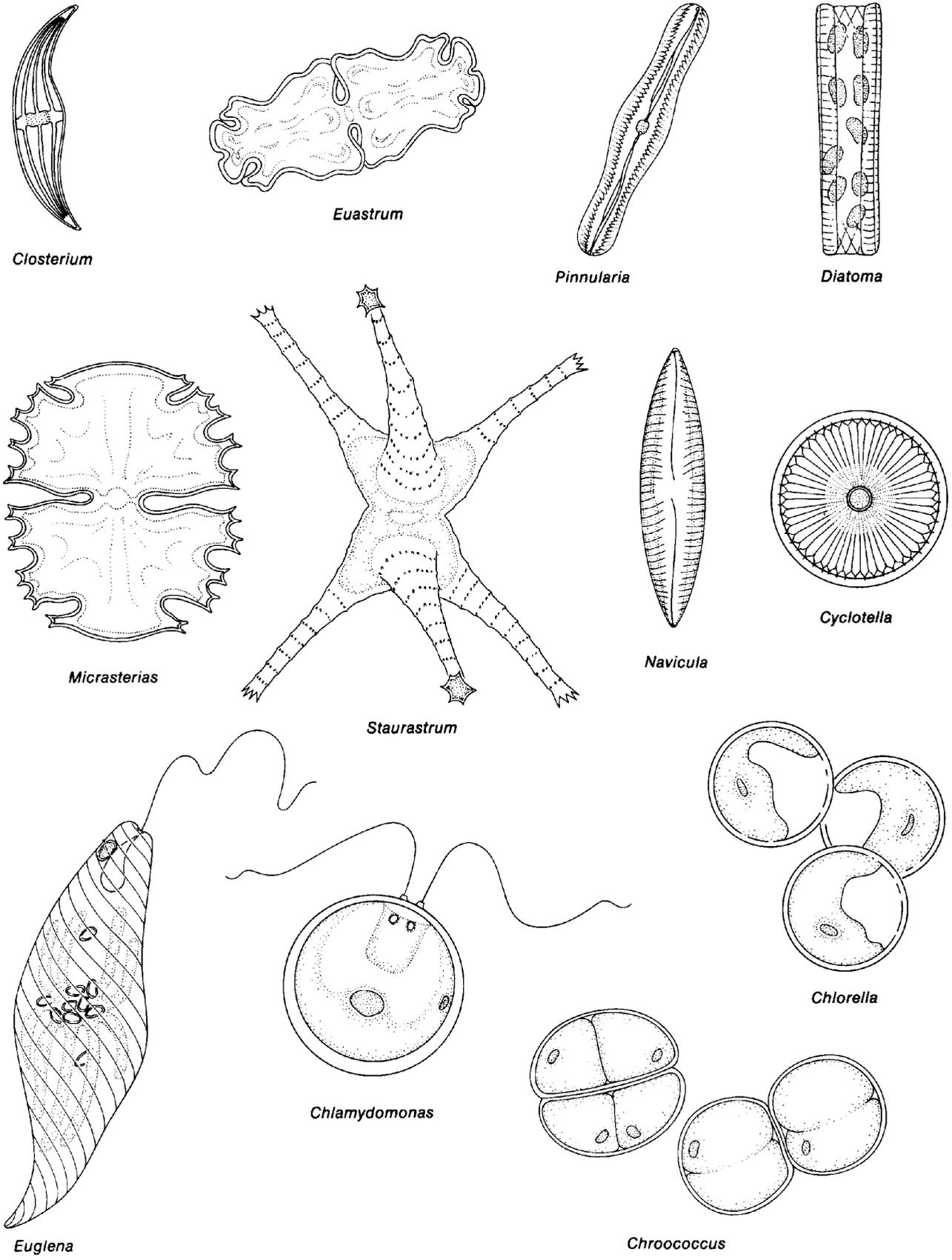


FIG. 1-9
Unicellular algae commonly found in pond water.

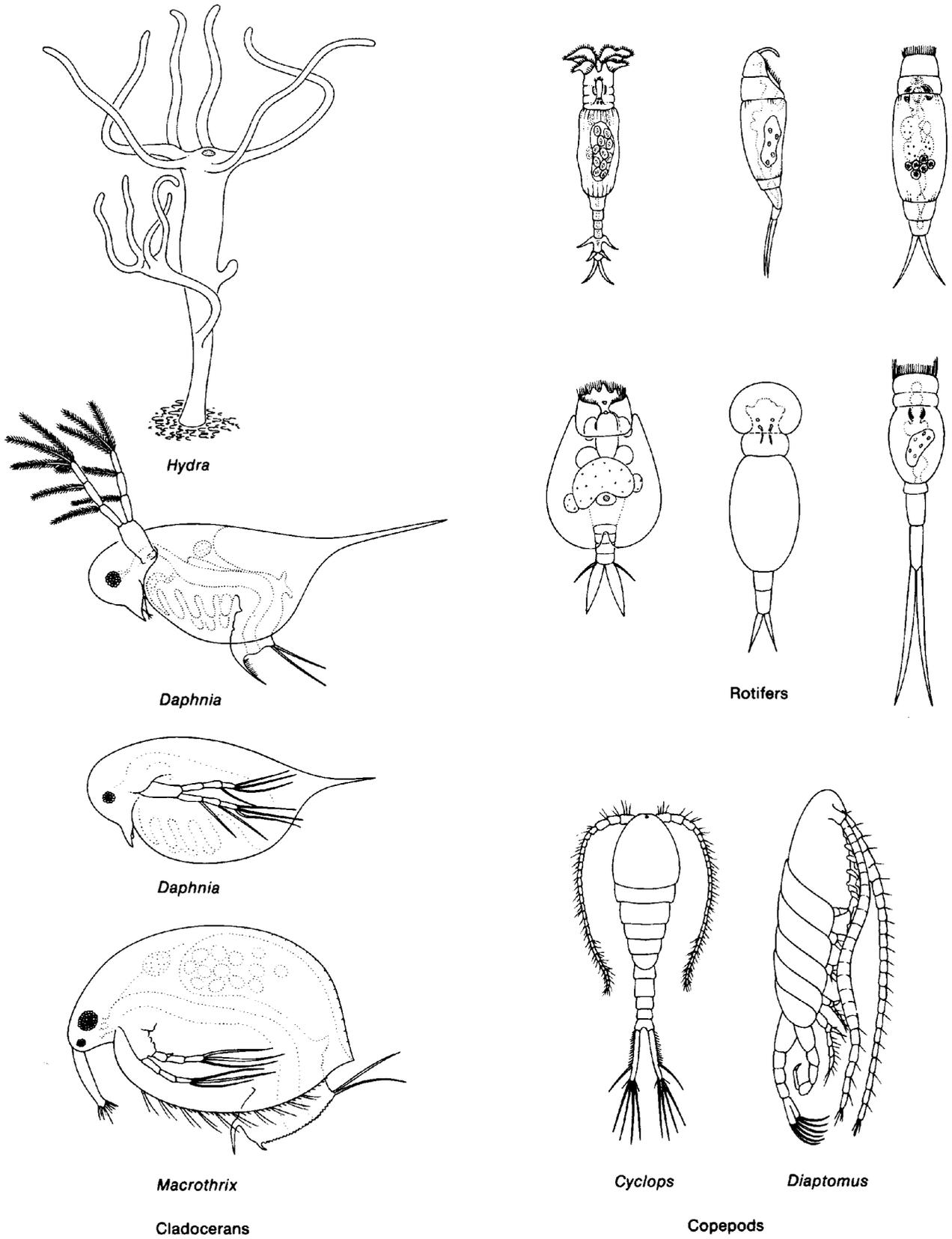
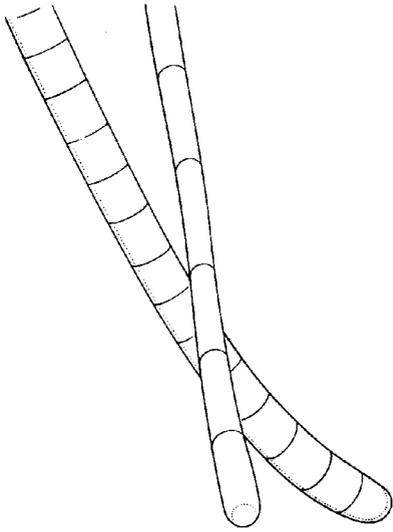
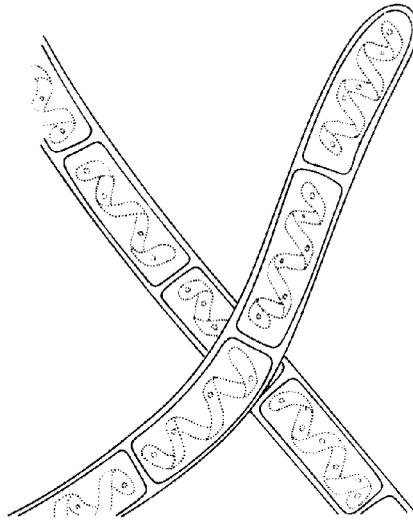


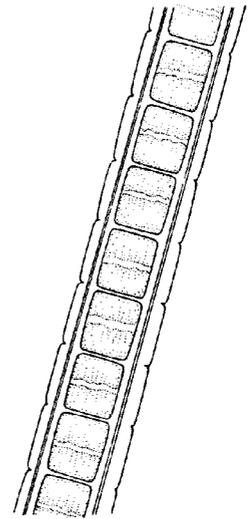
FIG. 1-10
Invertebrates commonly found in pond water.



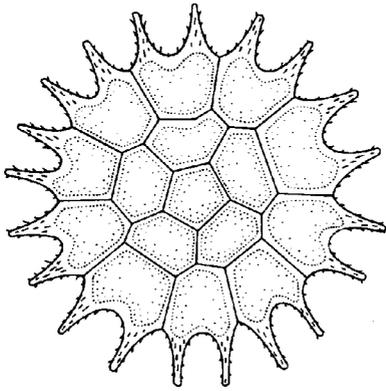
Oscillatoria



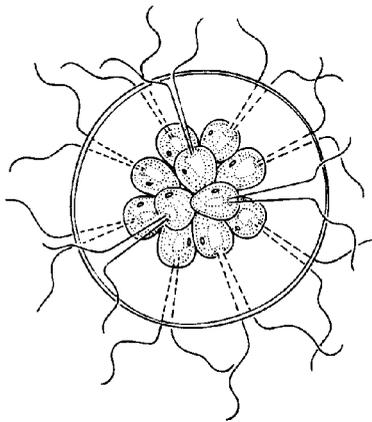
Spirogyra



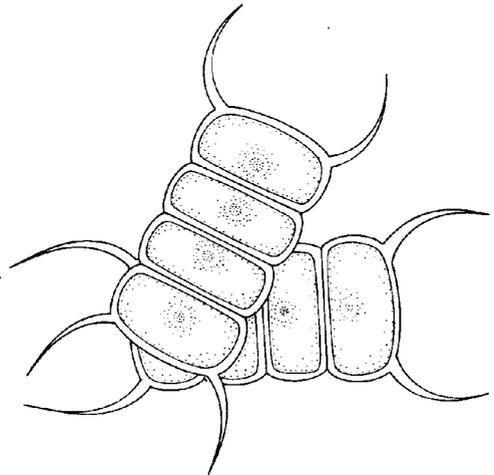
Zygnema



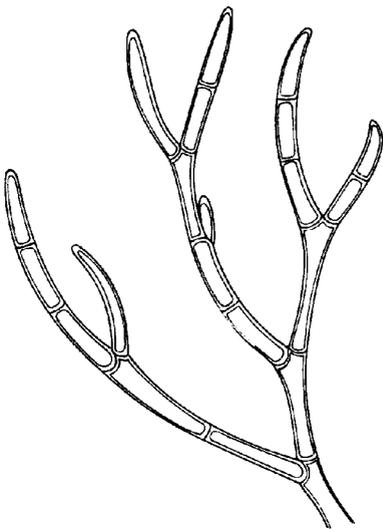
Pediastrum



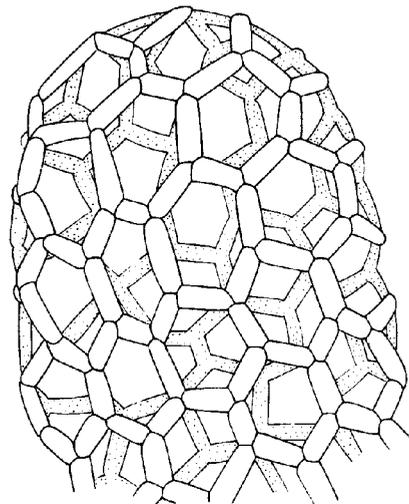
Pandorina



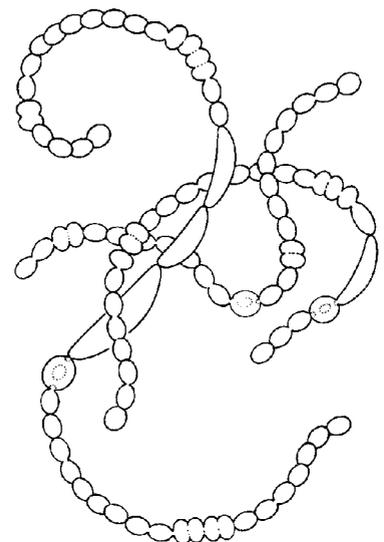
Scenedesmus



Cladophora



Hydrodictyon



Anabaena

FIG. 1-11
Multicellular algae and cyanobacteria commonly found in pond water.

IX. Glossary of Selected Terms

ablation Extirpation, removal or otherwise damaging the eye in some manner to promote maturation.

Artemia Brine shrimp.

aseptic Sterile.

axenic Free from other living organisms.

bacteria One-celled organisms that can be seen only with a microscope. Compared to protozoans they are of less complex organization and are much smaller in size.

broodstock Larger animals that are sourced and expected to produce offspring in the maturation facility.

carboy Glass bottle.

cysts Eggs of *Artemia* that are in a dormant stage.

decapsulation Removal of the thick outer layer on *Artemia* cysts.

disinfection Reduction of bacterial numbers to "safe" or "acceptable" level.

exoskeleton Shell of the shrimp.

grooves There are numerous grooves on a shrimp. Some are used as an aid in identifying the species. Examples:

- 1) The "thumbnail" grooves on the last abdominal segment of tropical Atlantic brown shrimp distinguish them from similar white shrimp;
- 2) The length of the rostral groove is shorter in *P. monodon* than in *P. semisulcatus* and can be used as one way to distinguish the two similar species. Other distinguishing characteristics can be seen in Figure 18.

hemacytometer Device used for counting algae cells.

hemolymph Blood.

Hensen-Stemple pipet Pipet used to take an objective water sample.

hepatopancreas Digestive gland.

larvae Plural of larva and the stage in the development of a shrimp's life cycle between the egg and the juvenile.

maturation The act of maturing; in this case egg development, mating and then spawning.

media Plural of medium and in this case refers to saltwater and nutrients or food added to promote proper growth either of algae, larvae, etc.

molt For shrimp, shedding of the exoskeleton.

nauplii Plural of nauplius and the first of three major larval stages. It is a non-feeding stage and is the best stage to transport the animals until they reach the postlarval stage.

penaeid The family, superfamily and an infraorder of shrimp distinguished from the caridean shrimp by the shape of the second segment of the abdomen. The sides of the penaeid shrimp shell (known as the pleura) overlap each segment that is behind it. In the caridean shrimp the pleurum of the second segment overlaps both the first segment and the third, making the second segment look very large. There are 109 species of penaeid shrimp listed by the Food and Agriculture Organization of the United Nations (F.A.O.).

petasma Male shrimp reproductive structure.

prawn According to Dore and Frimodt (1987), different people use this name to mean and to apply to quite different species.

U.K.= larger than shrimp.

U.S. = restaurants use it to mean large shrimp and other places mean small shrimp or freshwater shrimp.

Norway = producers promote the northern shrimp *Pandalus* as a prawn, and the "Dublin Bay prawn" is even used to describe *Nephrops*, which is a langoustine or Norway lobster.

The Oxford Dictionary defines prawn as "larger than shrimp" whereas Webster's Dictionary describes it as "a small, edible crustacean of the shrimp family."

South Africa = larger animals = prawns; smaller animals = shrimp.

F.A.O. attempted to introduce a clear-cut distinction as early as 1967. At the World Conference on the biology and culture of shrimps and prawns held in Mexico City, it was agreed that the term "prawn" was to be reserved for freshwater creatures only, while their marine/brackish water relatives were to be called "shrimps." Unfortunately, despite all of their efforts the confusion continues. The only point on which everyone can surely agree is that the use of "prawn" in the English language is confusing and unclear, and should be avoided.

protozoa New world term for zoea or the second major larval stage of penaeid shrimp.

raceway A small pond or tank which is usually rectangular with a center divider or circular tank with a water flow that "races" around the tank.

rostrum The pointed prow that extends from the head of most shrimp.

setae Hair-like structures that appear to branch off of appendages or legs.

sourcing Obtaining animals to be used for broodstock.

spore A small cell that can develop into a new individual.

spp. or sp. Plural and singular abbreviations for species, respectively.

sterilization Total inactivation of all microbial life.

thelycum Female shrimp reproductive structure.

X. Acknowledgments

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