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LAB TOPIC 22

# Vertebrate Anatomy I: The Skin and Digestive System

## Overview of Vertebrate Anatomy Labs (Lab Topics 22, 23, and 24)

In Lab Topics 18 and 19, Animal Diversity I and II, you investigated several major themes in biology as illustrated by biodiversity in the animal kingdom. One of these themes is the relationship between form and function in organ systems. In this and the following two lab topics, you will continue to expand your understanding of this theme as you investigate the relationship between form, or structure, and function in vertebrate organ systems. For these investigations, you will be asked to view prepared slides and isolated adult vertebrate organs, and to dissect a representative vertebrate, the fetal pig. The purpose of these investigations is not to complete a comprehensive study of vertebrate morphology but rather to use several select vertebrate systems to analyze critically the relationship between form and function.

You will explore the listed concepts in the designated exercises.

1. The specialization of cells into tissues with specific functions makes possible the development of functional units, or organs (Exercise 22.1, Histology of the Skin).
2. Multicellular heterotrophic organisms must obtain and process food for body maintenance, growth, and repair (Exercise 22.3, The Digestive System in the Fetal Pig).
3. Because of their size, complexity, and level of activity, vertebrates require a complex system to transport nutrients and oxygen to body tissues and to remove waste from all body tissues (Exercise 23.1, Glands and Respiratory Structures of the Neck and Thoracic Cavity; Exercise 23.2, The Heart and the Pulmonary Blood Circuit; Exercise 24.1, The Excretory System).
4. Reproduction is the ultimate objective of all metabolic processes. Sexual reproduction involves the production of two different gametes, the bringing together of the gametes for fertilization, and limited or extensive care of the new individual (Exercise 24.2, The Reproductive System).
5. Complex animals with many organ systems must coordinate the activities of the diverse parts. Coordination is influenced by the endocrine and nervous systems. Integration via the endocrine system is generally slower and more prolonged than that produced by the nervous system, which may receive stimuli, process information, and elicit a response very quickly (Exercise 24.3, Nervous Tissue, the Reflex Arc, and the Vertebrate Eye).

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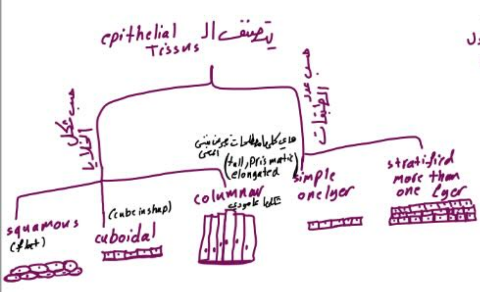
## Laboratory Objectives

After completing this lab topic, you should be able to:

1. Describe the four main categories of tissues and give examples of each.
2. Identify tissues and structures in mammalian skin.
3. Describe the function of skin. Describe how the morphology of skin makes possible its functions.
4. Identify structures in the fetal pig digestive system.
5. Describe the role played by each digestive structure in the digestion and processing of food.
6. Relate tissue types to organ anatomy.
7. Apply knowledge and understanding acquired in this lab to problems in human physiology.
8. Apply knowledge and understanding acquired in this lab to explain organismal adaptive strategies.

## Introduction



All animals are composed of **tissues,** groups of cells that are similar in structure and that perform a common function. During the embryonic development of most animals, the body is composed of three tissue layers: **ectoderm, mesoderm,** and **endoderm.** (Recall from Lab Topic 18, Animal Diversity I, that animals in the phylum Porifera lack true tissue organization and that animals in the phylum Cnidaria have only two tissue layers—ectoderm and endoderm.) It is from these embryonic tissue layers that all other body tissues develop. There are four main categories of tissues: epithelium, connective tissue, muscle, and nervous tissue. Organs are formed from these tissues, and usually all four will be found in a single organ.

Tissues are composed of cells and extracellular substances. The extracellular substances are secreted by the cells. **Epithelial tissue** has cells in close aggregates with little extracellular substance (see Figure 22.1). These cells may be in one continuous layer, or they may be in multiple layers. They generally cover or line an external or internal surface of an organ or cavity. If formed from single layers of cells, the epithelium is called **simple.** If cells are in multiple layers, the epithelium is called **stratified.** If epithelial cells are flat, they are called **squamous.** If they are cube-shaped, they are called **cuboidal.** Tall, prismatic cells are called **columnar.** Thus, epithelium can be stratified squamous (as in skin), simple cuboidal (as in kidney tubules), or other combinations of characteristics. Epithelial layers may be derived from embryonic ectoderm, mesoderm, or endoderm.

In **connective tissue,** cells are widely scattered in an **extracellular matrix** consisting of a web of fibers and an amorphous foundation that may be solid, gelatinous, or liquid (Figure 22.2). **Loose connective tissue** binds together tissues and organs and helps hold organs in place. Fibers in this tissue are loosely woven in a liquid matrix. **Adipose tissue,** another connective tissue, consists of adipose cells with fibers in a soft, liquid extracellular matrix. Adipose cells store droplets of fat, causing the cells to swell and pushing the nuclei to one side. **Bone** and **cartilage** are specialized connective tissues found in the skeleton characterized by cells embedded in,

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endo / ecto / meso

**Epithelial tissue**

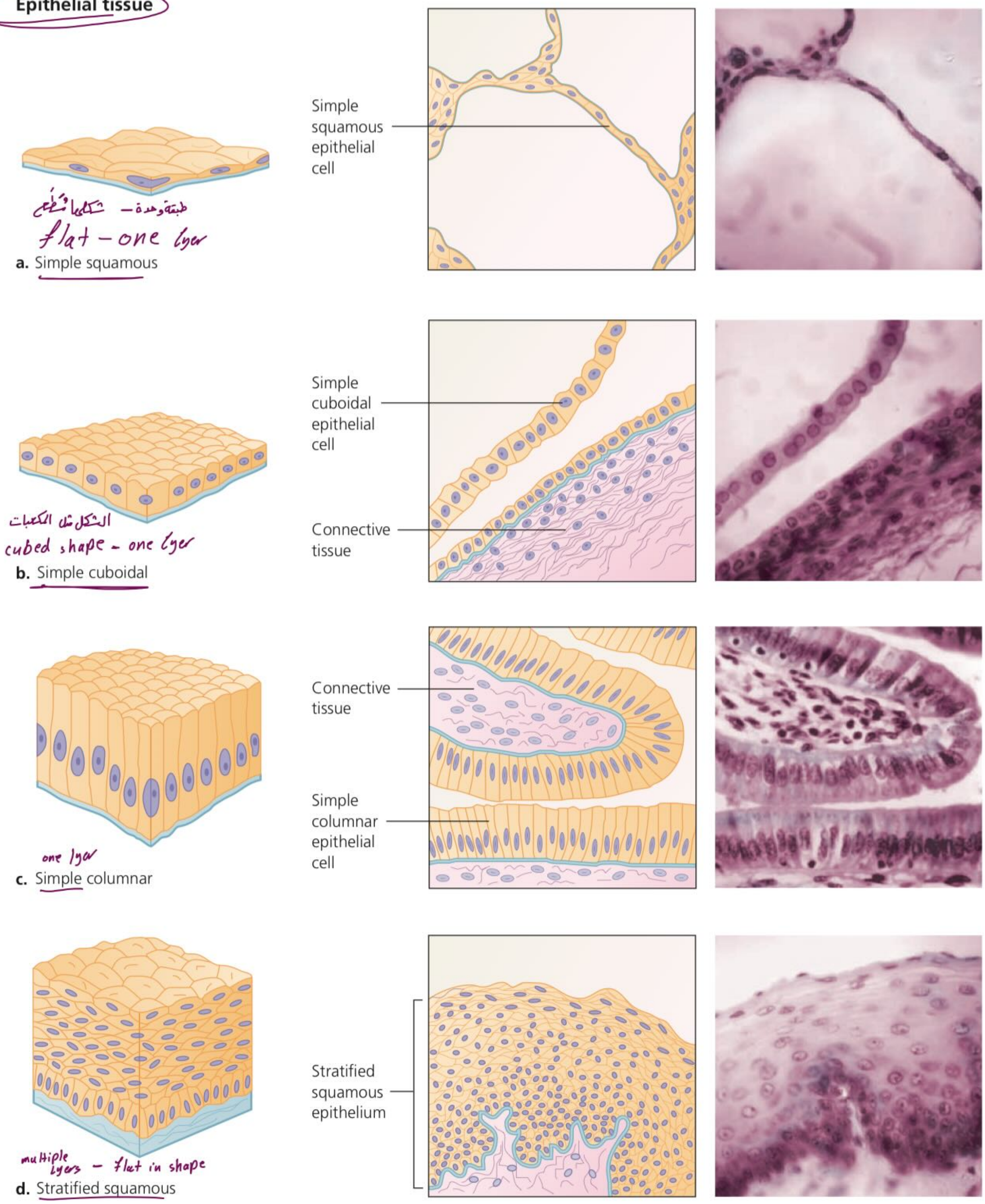


**FIGURE 22.1**

**Epithelial tissue.** Epithelial tissue has closely packed cells with little extracellular matrix. Cells may be (a) squamous (flat), (b) cuboidal (cube-shaped), or (c) columnar (elongated). They may be simple (in single layers) or (d) stratified (in multiple layers).

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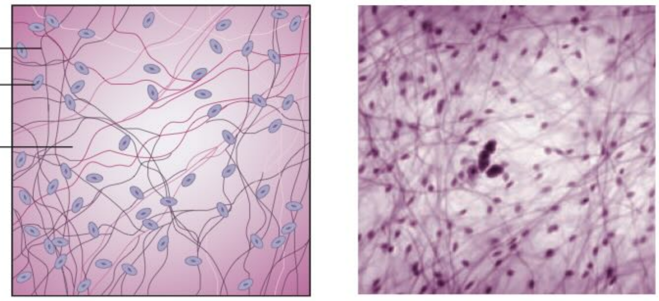
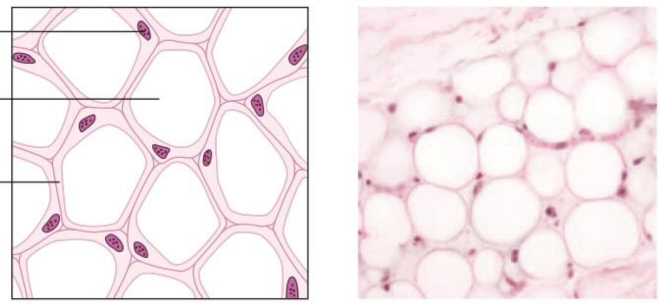
mesoderm

**Connective tissue**

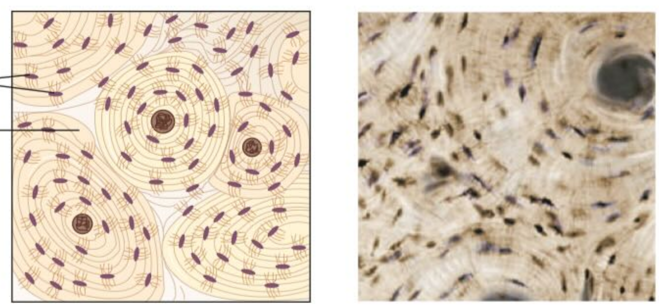
**a.** Loose connective tissue (Aerolar; liquid matrix)



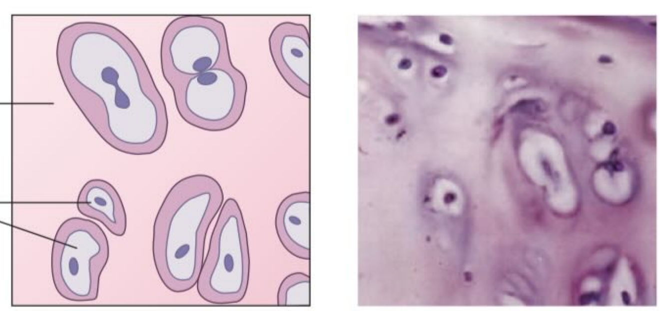

**b.** Adipose tissue (liquid matrix)



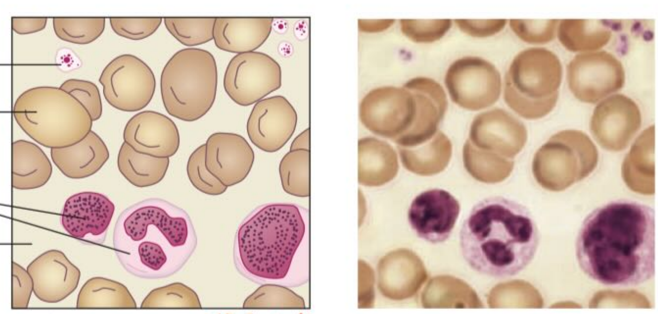

**c.** Bone (hard matrix)

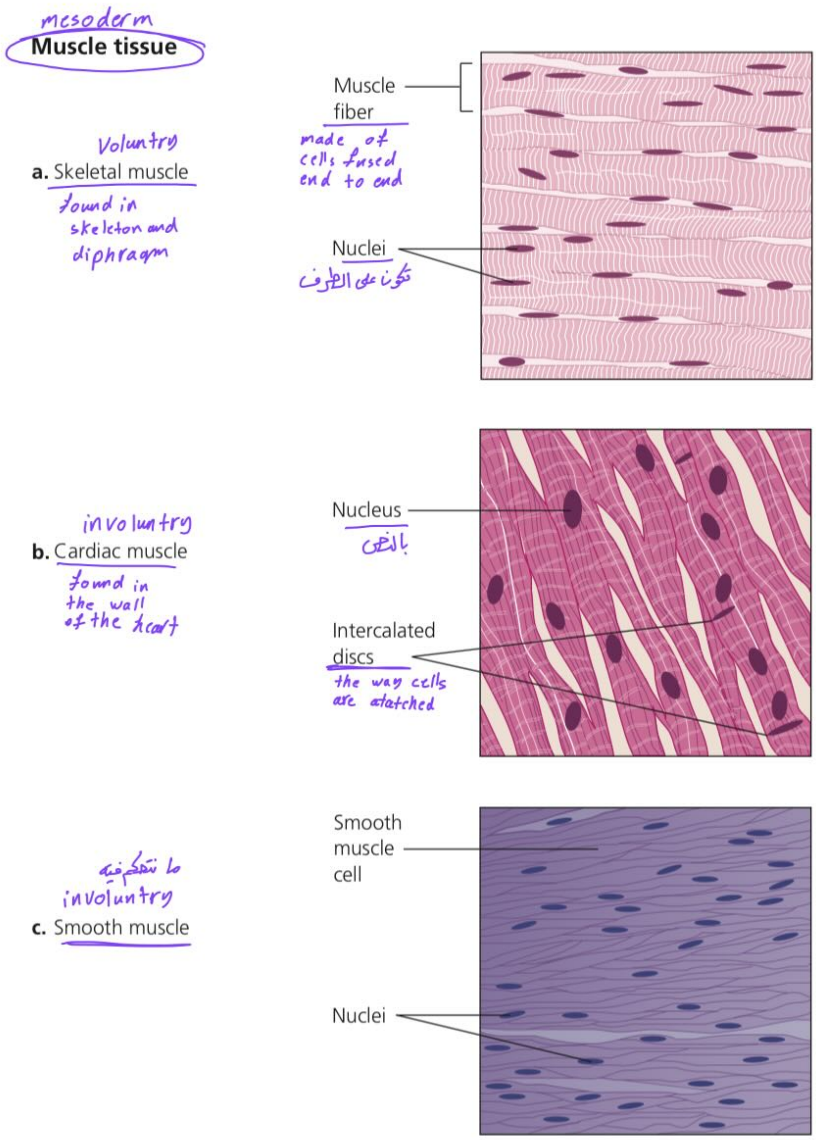



**d.** Cartilage (glatinuos matrix)




**e.** Blood (liquid matrix)





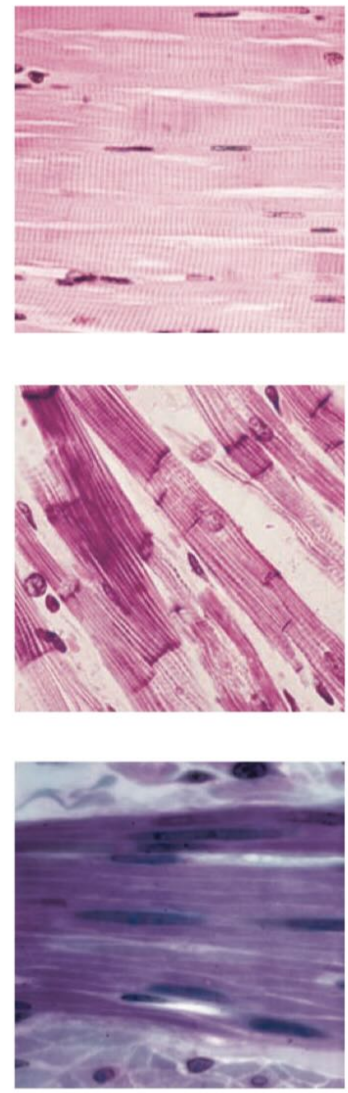



**FIGURE 22.2 (at left)**

**Connective tissue.** (a) In loose connective tissue, cells are embedded in a liquid fibrous matrix. (b) Adipose tissue stores fat droplets in adipose cells. (c) In bone, cells are embedded in a solid fibrous matrix. (d) In cartilage, cells are embedded in a gelatinous fibrous matrix. (e) In blood, cells are embedded in a liquid matrix.

**FIGURE 22.3 (above)**

**Muscle tissue.** Muscle tissue is either striated or smooth. (a) Skeletal muscle is striated. (b) Cardiac muscle is also striated. (c) Smooth, or visceral, muscle is not striated.

respectively, a hard or a gelatinous extracellular matrix. In bone the matrix is secreted by cells called **osteocytes.** The matrix in cartilage is secreted by cells called **chondrocytes. Blood** is a connective tissue consisting of cellular components called **erythrocytes** (red blood cells), **leukocytes** (white blood cells), and **platelets** (cell fragments) in a liquid matrix called **plasma.** Other connective tissues fill the spaces between various tissues, binding them together or performing other functions. Connective tissues are derived from the embryonic tissue layer, mesoderm.

**Muscle tissue** may be **striated,** showing a pattern of alternating light and dark bands, or **smooth,** showing no banding pattern (Figure 22.3). There are two types of striated muscle, skeletal and cardiac. **Skeletal** muscle moves the skeleton and the diaphragm and is made of muscle fibers formed by the end-to-end fusion of several cells, creating fibers with multiple nuclei.

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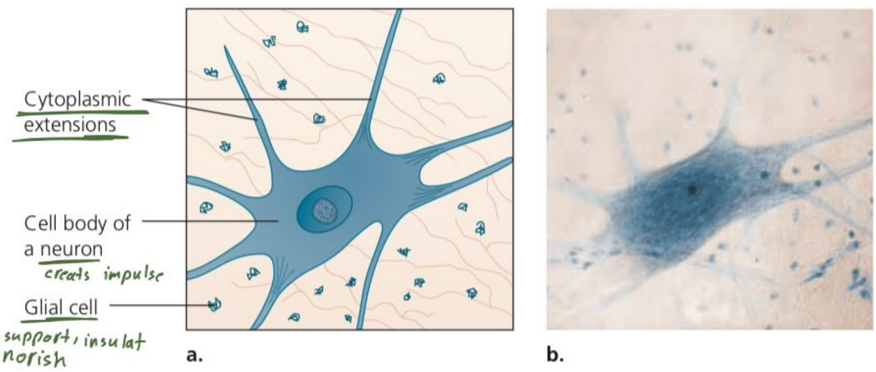


**FIGURE 22.4** **Nervous tissue.** Neurons and glial cells.

**Cardiac** muscle, found only in the wall of the heart, is also striated, but the cells do not fuse. Cells are attached by **intercalated discs. Smooth muscle,** also called **visceral** muscle, is found in the skin and in the walls of organs such as the stomach, intestine, and uterus. Muscle, like connective tissue, is derived from mesoderm.

**Nervous tissue** is found in the central nervous system (brain and spinal cord) and in the peripheral nervous system consisting of nerves (Figure 22.4). Nervous tissue is found in every organ throughout the body. There are two basic cell types, neurons and glial cells. **Neurons** are capable of responding to physical and chemical stimuli by creating an **impulse,** which is transmitted from one locality to another. **Glial cells** support, insulate, and nourish the neurons. Nervous tissue is derived from ectoderm.

The organs that you will investigate in the following exercises are made up of the four basic tissues. The tissues, each with a specific function, are organized into a functional organ unit.

# EXERCISE 22.1
# Histology of the Skin

## Materials

compound microscope
prepared slide of mammalian skin—pig, monkey, or human

## Introduction

Tissues are structurally arranged to function together in **organs,** which are adapted to perform specific functions. Organs are found in all but the simplest animals. The largest organ of the vertebrate body, the skin, illustrates the organization of tissues, each with a specific function, into a functioning organ (Figure 22.5). The skin protects the body from dehydration and bacterial invasion, assists in regulating body temperature, and receives stimuli from the environment. As you work through this exercise, ask how the unique function of each tissue produces the functioning whole—the skin. Review information about each observed tissue type in "Introduction to Tissues" (pp. 612–616).

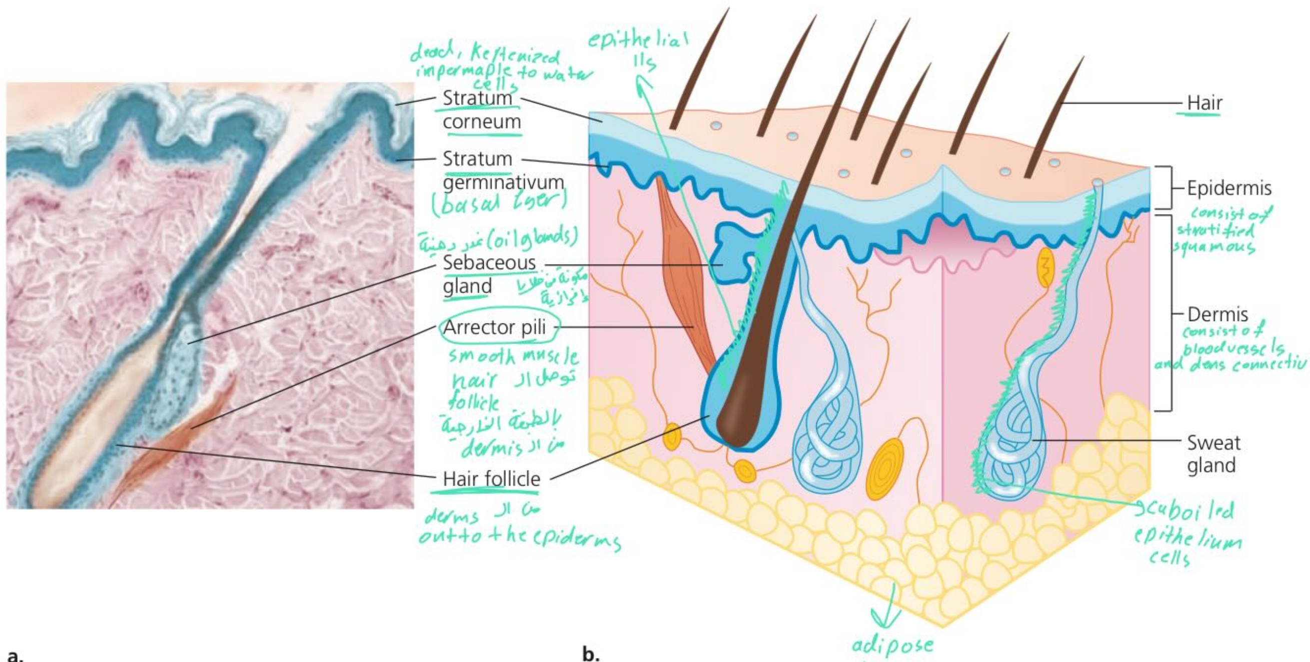


**FIGURE 22.5**
**Mammalian skin structure.** (a) Photomicrograph of a cross section of the skin. (b) A diagram detailing the skin structure.

## Procedure

1. Obtain a prepared slide of mammalian skin (pig, monkey, or human). View it using the low and intermediate power objectives on the compound microscope.
2. Identify the two main layers of the skin. The thin outer layer, the **epidermis,** consists of **stratified squamous epithelium** (Figure 22.1d); the thicker inner layer, the **dermis,** consists mainly of **dense connective tissue** and scattered **blood vessels.** The dermis merges into layers of loose connective tissue (Figure 22.2a) and smooth muscle (Figure 22.3c), which are not considered part of the skin.
3. Locate **hair follicles** extending obliquely from the epidermis into the dermis. In the living animal, each follicle contains a hair, but the hair shaft may or may not be visible in every follicle on your slide, depending on the plane of the section through the follicle. The follicle is lined by epithelial cells continuous with the epidermis. Carefully observe several hair follicles. You may be able to find a band of smooth muscle cells attached to the side of a follicle. This muscle, called the **arrector pili,** attaches the hair follicle to the outermost layer of the dermis. When stimulated by cold or fright, it pulls the hair erect, causing "goose bumps." In furry animals this adaptation increases the thickness of the coat to provide additional temperature insulation. Clusters of secretory cells making up **sebaceous glands** (oil glands) are also associated with hair follicles. These are more obvious in monkey and human skin slides than in pig skin slides.

4. Focus your attention on the epidermis and locate the outermost layer, the **stratum corneum,** a layer of dead, keratinized cells, impermeable to water. This layer is continually exfoliated and replaced. The thickness of the stratum corneum varies, depending on the location of the skin. This layer is very thick on the soles of the feet and the palms of human hands.
5. The innermost layer of cells in the epidermis is called the **basal layer** or the **stratum germinativum.** These cells divide mitotically and produce new cells, which, as they mature, are pushed to higher and higher layers of the epidermis, until they fill with keratin and form the stratum corneum. Scattered through the basal layer (although not seen on your slide) are cells called **melanocytes** that produce **melanin,** a pigment that produces brown or black hues in the skin. The melanin is inserted into newly forming epidermal cells as they are pushed outward. Regular exposure to sunlight stimulates melanocytes to produce more melanin, helping protect the body against the potentially harmful effects of sun exposure.
6. In addition to hair follicles, coiled tubular **sweat glands** lined with **cuboidal epithelium** (Figure 22.1b) extend from the epidermis into the dermis. They appear as circular clusters in cross section and may be easily located in pig or human skin but are less numerous or absent in the skin of furry animals, such as monkeys. The tubular secretory portion is convoluted into a ball, which connects with a narrow unbranched tube leading to the skin surface. It is unlikely that you will see an entire intact sweat gland in one section.
7. Look for connective tissue and blood vessels, which often contain red blood cells in the dermis. Look for adipose tissue (Figure 22.2b) below the dermis.

## Results

In Table 22.1, list the tissues you have identified in the skin and indicate the specific function of each.

**TABLE 22.1** Tissues of the Skin and Their Functions

| Tissue | Function |
|---|---|
| | |
| | |
| | |
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| | |
| | |

LAB TOPIC 25

# Animal Development

## Laboratory Objectives

After completing this lab topic, you should be able to:

1. Describe early development in echinoderms (sea urchin and sea star), an amphibian (frog or salamander), a fish (zebrafish), and a bird (chicken).
2. List the events in early development that are common to all organisms.
3. Compare early development in the organisms studied, speculating about factors causing differences.
4. Relate the events of early development in vertebrates to the formation of a dorsal nerve cord.
5. Discuss the effects of large amounts of yolk on the events of early development.

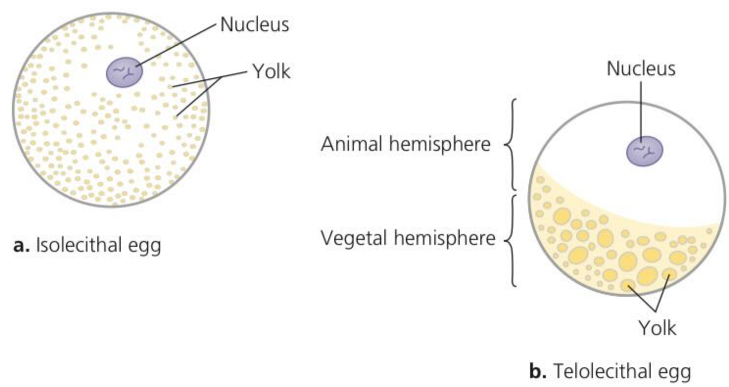
## Introduction

The development of a multicellular organism involves many stages in a long process beginning with the production and fusion of male and female gametes, continuing with the development of a multicellular embryo, the emergence of larval or juvenile stages, growth and maturation to sexual maturity, and the process of aging, and eventually ending with the death of the organism. A range of biological processes functions in development, including **cell division; differentiation,** where cells, tissues, and organs become specialized for a particular function; and **morphogenesis,** the development of the animal's shape, or body form, and organization.

Early developmental studies focused on the description of form, or **morphology,** of animals as they grow. Developmental biologists asked questions about the process of development and the forces involved in morphogenesis. Model organisms used in these studies included the sea urchin and sea star (echinoderms), frogs and salamanders (amphibians), and the chick. More recently, the zebrafish and other organisms have become important subjects for developmental studies. Currently, developmental biologists use these same animals to ask questions about the genetic control of development and the processes involved in activating different genes in different cells. Before these questions can be pursued, however, it is important to have a basic understanding of morphology in early development.

In this lab topic, you will use several of the model organisms of classical and current research to investigate major early developmental events common to most animals. These events include **gametogenesis,** the production of gametes; **fertilization,** the union of male and female gametes; **cleavage,** the production of a multicellular blastula; **gastrulation,** the formation of three

**FIGURE 25.1**
**Egg types based on amount and distribution of yolk.** (a) Isolecithal eggs have small amounts of evenly distributed yolk. (b) Telolecithal eggs have large amounts of yolk concentrated at one end.




primary germ layers—ectoderm, mesoderm, and endoderm; **neurulation,** the formation of the nervous system in chordates; and **organogenesis,** the development of organs from the three primary germ layers. Although you may observe all of these developmental stages, you will study primarily cleavage and blastulation, gastrulation, neurulation, and organogenesis.

## Overview of Stages in Early Development

### Stage 1: Preparation of the Egg, Fertilization, and Cleavage

Development begins as sperm and egg prepare for fertilization. Sperm develop a flagellum, which propels the cell containing the haploid genetic complement of the paternal parent to the egg, which contains the haploid maternal genetic complement. As an egg matures, sperm-binding receptors develop on its surface. These can only bind to matching receptors in the sperm, insuring fertilization of egg and sperm of the same species. Within the egg, food reserves called **yolk** accumulate. These reserves are mainly protein and fat and will be utilized by the early embryo.

When egg and sperm unite, their nuclei, each containing a haploid set of chromosomes, combine to form one diploid cell, the **zygote.** The mitotic cell divisions of cleavage rapidly convert the zygote to a multicellular ball, or disc, called the **blastula.** The cells of the blastula are called **blastomeres.** A cavity, the **blastocoel,** forms within the ball of cells. The position of the blastocoel in the developing blastula depends on the amount and distribution of yolk in the egg and developing embryo.

#### *Egg Types*

Because early events in development are affected by the amount of yolk present in the egg, the classification of eggs is based on the amount and distribution of yolk. Eggs with small amounts of evenly distributed yolk are called **isolecithal** eggs (Figure 25.1a). Eggs containing large amounts of yolk

**FIGURE 25.2 (at right)**
**Cleavage types based on amount and distribution of yolk.** (a) In isolecithal eggs, cleavage is holoblastic, and the blastocoel is centrally located. (b) In moderately telolecithal eggs, cleavage is holoblastic, and the blastocoel develops in the animal hemisphere. (c) In strongly telolecithal eggs, cleavage is meroblastic. Only the active cytoplasm divides, producing a cap of cells, the blastoderm. The blastocoel forms within the blastoderm.

concentrated at one end are called **telolecithal** eggs (Figure 25.1b). Some species are moderately telolecithal, whereas others are strongly telolecithal. Eggs may also be classified as **centrolecithal** (yolk in the center of the egg, as in insects) or **alecithal** (no significant yolk reserves). Neither of these conditions will be studied in this lab topic.

In strongly telolecithal eggs, the nucleus is surrounded by **active cytoplasm**, which is relatively devoid of yolk. This nuclear-cytoplasmic region is called the **blastodisc.** The blastodisc is displaced toward the pole of the egg where polar bodies budded from the cell in meiosis. This pole is designated the **animal pole.** The half of the egg associated with the animal pole is the **animal hemisphere.** In these eggs, the yolk is concentrated in the other half of the egg, designated the **vegetal hemisphere.** The pole of this hemisphere is the **vegetal pole.**

### *Cleavage Types*

Although the end result of cleavage, the formation of the blastula, is the same in all organisms, the pattern of cleavage can differ. One factor that influences the pattern of cleavage is the amount of yolk present. In total, or **holoblastic,** cleavage, cell divisions pass through the entire fertilized egg. This type of cleavage takes place in isolecithal eggs, where the impact of yolk is minimal (Figure 25.2a). In these eggs, the blastocoel forms in the center of the blastula. In moderately telolecithal eggs, the yolk will retard cytoplasmic divisions and affect the size of cells. However, if the entire egg is cleaved, cleavage is considered holoblastic (Figure 25.2b). In this case, the blastocoel develops in the animal hemisphere. Cells in this hemisphere will be smaller and have less yolk than cells in the vegetal hemisphere.

In a strongly telolecithal egg, only the active cytoplasm is divided during cleavage. This process is called **meroblastic** cleavage, and it produces a cap of cells called a **blastoderm** at the animal pole (Figure 25.2c). In meroblastic embryos, the blastocoel forms within the cell layers of the blastoderm.

## Stage 2: Gastrulation

Gastrulation transforms the blastula, the hollow ball of cells (in holoblastic cleavage) or cap of cells (in meroblastic cleavage), into a **gastrula** made up of three embryonic, or germ, layers: endoderm, ectoderm, and mesoderm (Figure 25.3). Whereas cleavage is characterized by cell division, gastrulation is characterized by cell movement. Surface cells migrate into the interior of the embryo forming a new internal cavity, the **archenteron.**

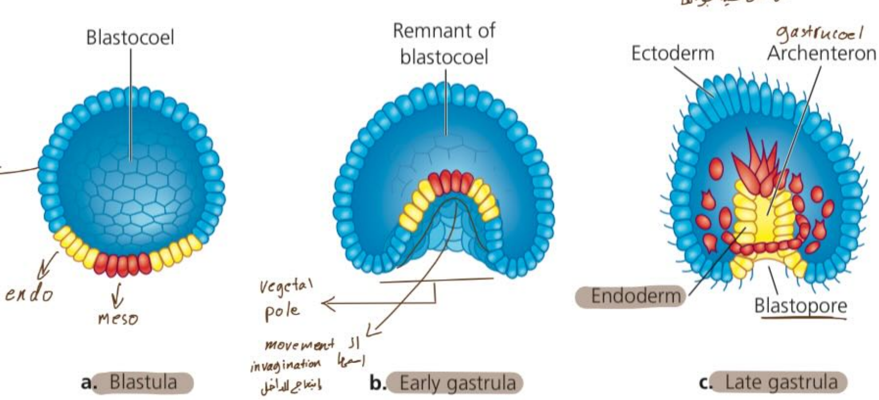
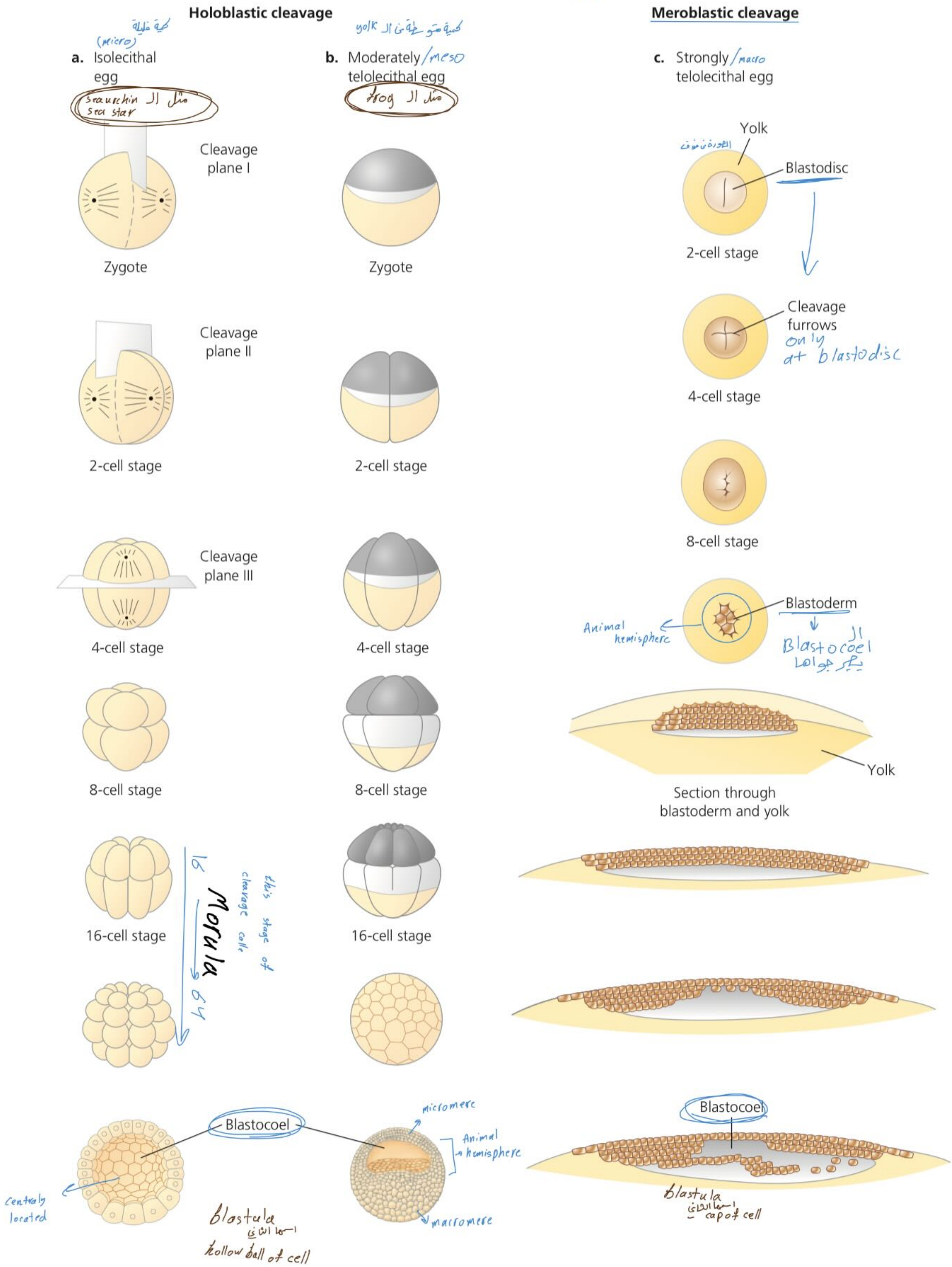



**FIGURE 25.3** **Gastrulation.** The blastula is converted to a three-layer embryo. Ectoderm (blue) and endoderm (yellow) germ layers form first. Mesoderm (red) forms later between the ectoderm and endoderm.

Cleavage types



This new cavity is lined by the **endoderm,** the embryonic germ layer that ultimately forms the digestive tract. The archenteron opens to the outside through the **blastopore,** which in deuterostomes becomes the anus. In protostomes, the blastopore becomes the mouth. (See Lab Topic 18, Animal Diversity I, or your text for examples of deuterostomes and protostomes.) The cells that remain on the surface of the embryo become the **ectoderm.** A third layer of cells, the **mesoderm,** develops between ectoderm and endoderm.

## Stage 3: Neurulation

Late in gastrulation, neurulation, the formation of a dorsal, hollow neural tube, begins (Figure 25.4). In this *strictly chordate* process, certain ectodermal cells flatten into an elongated **neural plate** extending from the dorsal edge of the blastopore to the anterior end of the embryo. The center of the plate sinks, forming a **neural groove.** The edges of the plate become elevated to form **neural folds,** which approach each other, touch, and eventually fuse, forming the hollow **neural tube.** The anterior end of the tube develops into the brain, while the posterior end develops into the nerve (spinal) cord.

## Stage 4: Organogenesis

After the germ layers and nervous system have been established, organogenesis, the formation of rudimentary organs and organ systems, takes place. Ectoderm, the source of the neural tube in chordates, also forms skin and associated glands. In chordates, somites and the notochord (see Lab Topic 19, Animal Diversity II) develop early from mesodermal cells. Later, muscles, the skeleton, gonads, the excretory system, and the circulatory system develop from mesoderm. Nonchordate animals lack somites and the notochord, but their muscles and organs of the excretory, circulatory, and reproductive systems develop from mesoderm. The endoderm develops into the lining of the digestive tract and such associated organs as the liver, pancreas, and lungs.

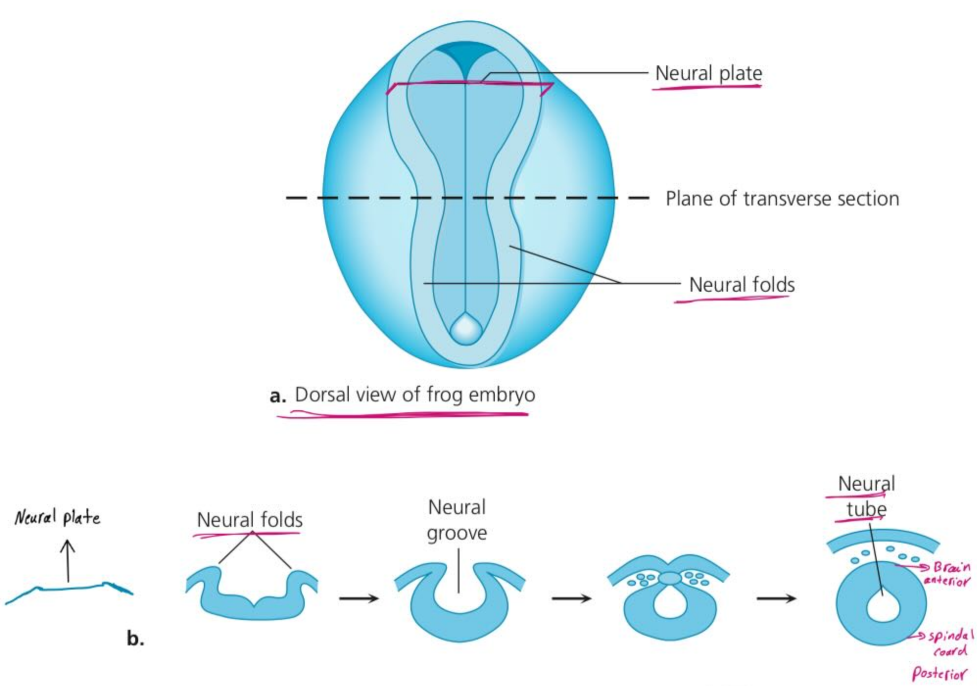



**FIGURE 25.4**
**Neurulation in a developing frog, a chordate.** (a) Dorsal view of the entire frog embryo, showing the ectodermal neural plate with edges elevated, forming the neural folds. (b) Seen in transverse section, the neural folds meet and fuse, forming the neural tube.

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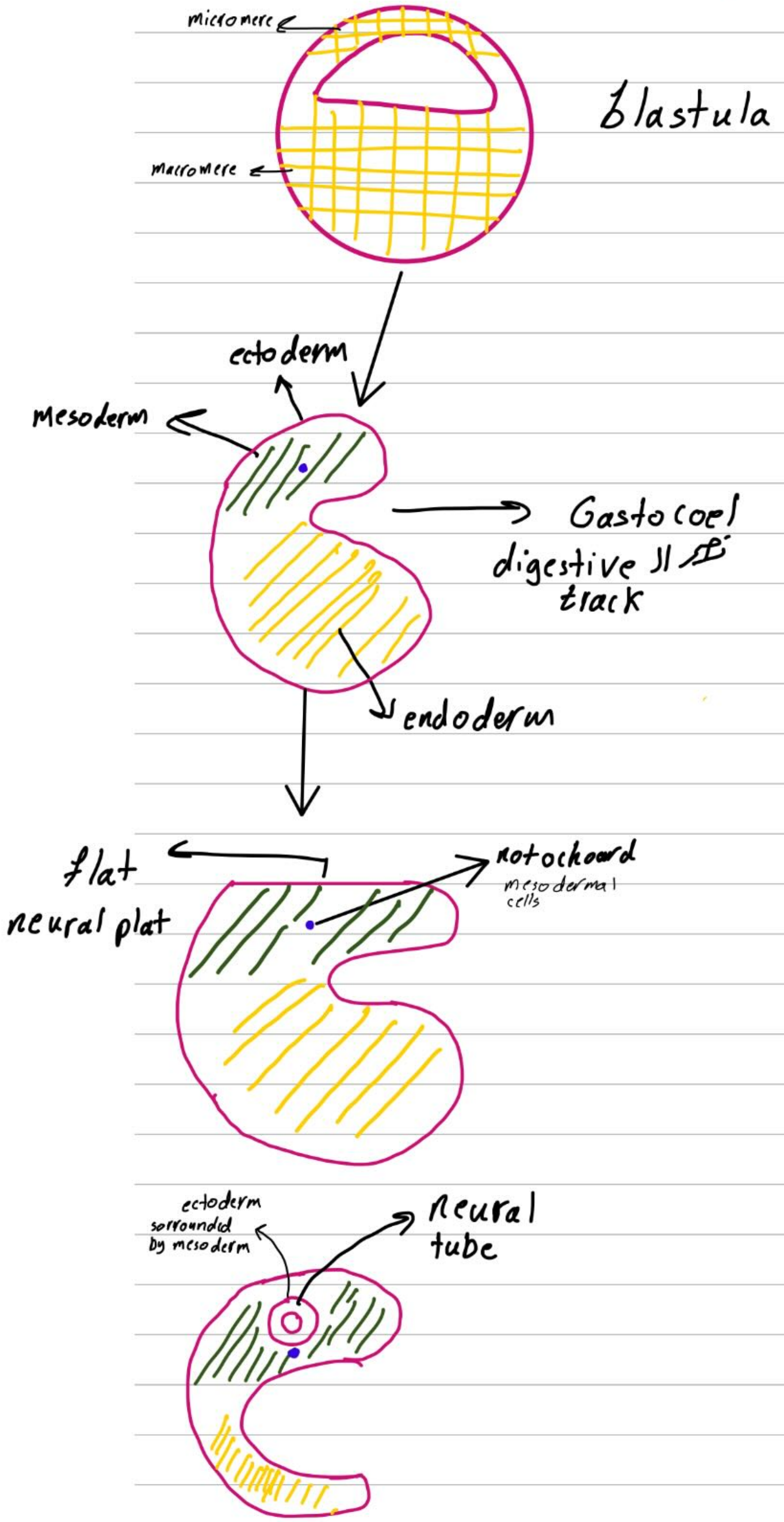
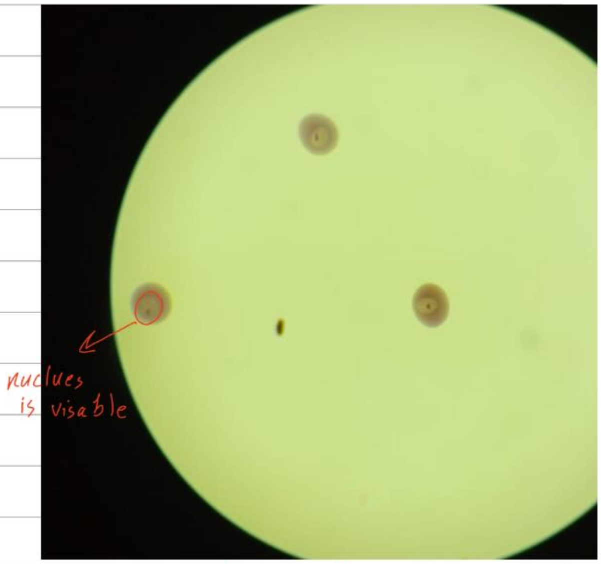
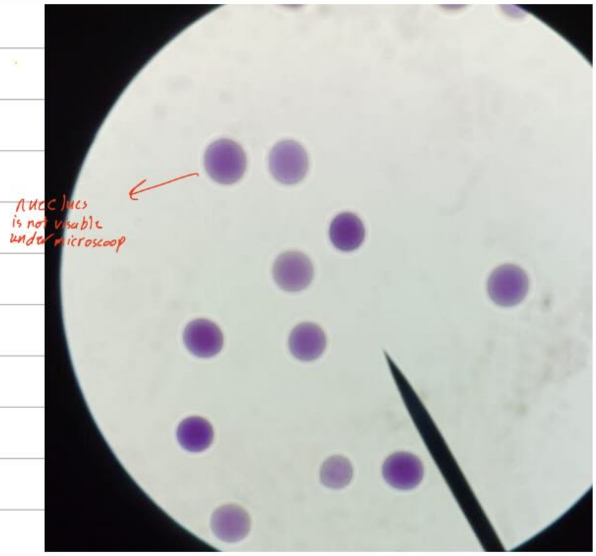
# frog
## (meso/medorately telolecithal)




unfertilized egg




fertilized egg

Today's lab will be a comparative study of early development in five organisms—the sea urchin and the sea star, both echinoderm invertebrates; and three chordates: the salamander, an amphibious vertebrate; the fish, an aquatic vertebrate; and the chick, a terrestrial vertebrate.

EXERCISE 25.1

# Development in Echinoderms: Sea Urchin and Sea Star

Sea urchins and sea stars (starfish) are classified in the phylum Echinodermata, the invertebrate group that is phylogenetically closer to chordates than any other. Echinoderms release large numbers of gametes into the sea, and fertilization is external. Early development leads to a larval stage that is free-swimming and free-feeding. In this exercise, you will observe fertilization and early development in living sea urchins, and then you will observe a prepared slide of sea star development.

## Lab Study A: Fertilization in Living Sea Urchins

### Materials

clean slides and coverslips
sand or glass chips in a small petri dish
transfer pipettes, one labeled "egg," the other labeled "sperm," cut to make a slightly larger bore
small clean test tube labeled "egg" containing a suspension of living eggs from a sea urchin, e.g., *Lytechinus* sp. or *Arbacia* sp.
small clean test tube labeled "sperm" containing a suspension of living sperm from a sea urchin, e.g., *Lytechinus* sp. or *Arbacia* sp.
moisture chamber made from a petri dish containing a piece of moist (not wet) filter paper

### Introduction

Before your laboratory began, your instructor collected sperm and eggs from living sea urchins by injecting a KCl solution into the body cavity of the urchin. An injection of this solution causes the urchin to extrude gametes from its genital pore located on its upper (aboral—opposite the mouth) surface. It is not possible to determine the sex of a sea urchin from its external anatomy. However, it is possible to determine its sex once it begins to extrude its gametes. Whereas eggs are colored (e.g., light orange in *Lytechinus*), sperm in all species are whitish. Each female sea urchin can spawn over a million eggs and each male a billion sperm. Your instructor collected living eggs and sperm in sea water.

An unfertilized echinoderm egg is surrounded by a protective *vitelline layer* just outside the plasma membrane and a **jelly coat** that slowly dissolves in sea water. When eggs and sperm come into contact, fertilization takes place and a halo—the **fertilization envelope**—forms around the fertilized egg. (See Figure 25.6.) This envelope helps prevent multiple fertilizations, or *polyspermy*. Two sequential processes prevent polyspermy, the *fast block* and the *slow block*. When a sperm first fuses with an egg, the permeability of the egg plasma membrane immediately changes, allowing an influx of sodium ions. The sodium ions change the electric potential across the cell membrane and, within a second or so, create a *fast block to polyspermy*. A second block to multiple fertilization takes about 20 to 30 seconds. This *slow block to polyspermy* involves the fusion of egg cytoplasmic vesicles with the egg plasma membrane. These vesicles lie in the cortex, or outer portion of the egg cytoplasm, and are called *cortical granules*. When they fuse with the egg membrane, their contents are expelled to the egg surface. Enzymes from the vesicles break bonds between the vitelline layer and the egg plasma membrane, and water flows between the two layers. The vitelline layer rises up from the egg membrane and becomes the *fertilization envelope*. Some time after the egg and sperm cells fuse, the egg and sperm nuclei, called *pronuclei*, move toward the center of the egg, where they fuse and almost immediately begin to prepare for the first mitotic division of cleavage.

In this lab study you will observe fertilization and early development. In Lab Study B, you will learn more details about the process and early stages of development in the sea star.

## Procedure

1. Place a generous drop of egg suspension on a clean microscope slide. Add a few grains of sand or glass chips and cover with a coverslip. Place the slide on your compound microscope stage and observe the eggs using first 4× and then 10× objectives.
2. Take a drop of sperm suspension and add this to the edge of the coverslip.
3. Working quickly, note the time and immediately begin observing the slide using first the 10× and then the high power objectives. Use phase-contrast microscopy on both powers if your microscope has this capability. Look for sperm swimming toward the eggs.
4. Carefully observe the eggs, watching for the formation of fertilization envelopes (a clear halo surrounding the egg), indicating that fertilization has taken place.
5. After observing fertilization for several minutes, place your slide in a moisture chamber made from a petri dish.
6. At 30-minute intervals until the end of lab, remove your slide from the petri dish, carefully dry off the bottom of the slide, and use the compound microscope to observe developing embryos. After you complete your observations, return the slide to the moisture chamber each time.

## Results

1. Describe the events that take place as sperm and eggs are mixed. How long did it take for fertilization envelopes to become visible?
2. Are fertilization envelopes present around all eggs? If not, can you estimate the percent of eggs that have been fertilized?
3. Describe the activity of the sperm. Do active sperm bounce off the egg, or do they become stuck to the jelly coat?

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4. Did you observe cleavage? How long after you first observed fertilization? Was the cleavage pattern *holoblastic* or *meroblastic* (Figure 25.2)?

5. At the end of the laboratory period, at what stage of development are most of your embryos?

### Discussion

1. What role might the jelly coat surrounding the egg perform in the process of fertilization?

2. What would you predict would happen if sperm were exposed to a solution containing only egg jelly coat? Would these sperm still be capable of fertilization?

Student Media Video—Ch. 46: Sea Urchin Embryonic Development

## Lab Study B: Development in the Sea Star

### Materials

compound microscope
prepared slide of whole sea star embryos in different stages of development

### Introduction

In this lab study, you will observe a prepared slide containing an assortment of whole sea star embryos in various stages of development. You will identify each developmental stage and determine the type of egg and cleavage pattern of the sea star.

### Procedure

1. View the prepared slide of sea star embryos using low and intermediate powers on the compound microscope.

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Use only low and intermediate powers. Using the high power objective to view this slide will destroy the slide!

2. Find examples of all stages of development. When you find a good example of each of the stages described, make a careful drawing of that stage in the appropriate square in Figure 25.12.

### *Unfertilized Egg*

By the time sea star eggs leave the body of the female, meiosis I and II are completed. The nucleus, called the *germinal vesicle*, is conspicuous because the nuclear envelope is intact (Figure 25.5). A nucleolus is usually distinct. The plasma membrane surrounding the egg cytoplasm closely adheres to a thin external layer known as the *vitelline layer* (not visible at this magnification). Species-specific sperm receptors extend from the egg plasma membrane into the vitelline layer.

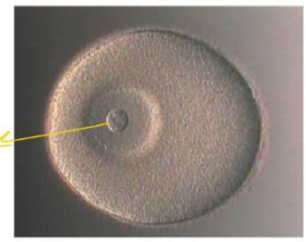

**FIGURE 25.5**
**Unfertilized egg with germinal vesicle.**

### *Fertilized Egg*

The fertilized egg, or zygote, has no visible nuclear envelope, giving this cell a uniform appearance (Figure 25.6). Look on the zygote surface for a **fertilization envelope,** most easily seen using phase-contrast microscopy. In Lab Study A you learned that this envelope forms as a result of sperm–egg fusion. The presence of the fertilization envelope and the absence of the visible nuclear envelope will help distinguish fertilized and unfertilized eggs.

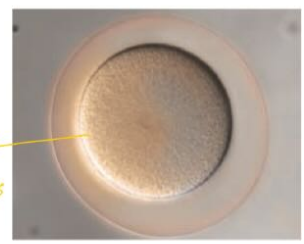

**FIGURE 25.6**
**Fertilized egg.**

### *Early Cleavage*

As cleavage begins, the zygote divides by mitosis and cytokinesis (Figure 25.7) and continues to divide as this single cell is converted into a multicellular embryo. The $G_1$ and $G_2$ phases of the cell cycle (see Lab Topic 7, Mitosis and Meiosis) are essentially skipped in these mitotic events. Find two-, four-, and eight-cell stages. Is the entire zygote involved in early cleavage?

**FIGURE 25.7**
**Early cleavage—two-cell stage.**

The fertilization envelope remains intact around the embryo until the gastrula stage. What is happening to the size of the cells as cleavage takes place and cell numbers increase?

### *Late Cleavage*

As cleavage continues, a cavity, the **blastocoel,** forms in the center of the cell cluster (Figure 25.8). The end product of cleavage will be a hollow ball of cells, the **blastula.** Locate and study blastulae in early and late stages of cleavage. The late blastula has a thick, dark wall of cells. The lighter blastocoel lies in the center of the blastula. How does the size of individual cells compare with the size of the fertilized egg?

**FIGURE 25.8**
**Later cleavage.**
Blastula with blastocoel.

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How does the overall size of the blastula compare with that of the fertilized egg?

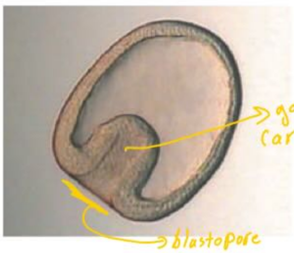


**FIGURE 25.9**
Early gastrulation.

## *Early Gastrulation*

Gastrulation converts the blastula into the gastrula, an embryo composed of three primary germ layers. The early gastrula can be recognized by a small bubble of cells protruding into the blastocoel (Figure 25.9). These cells push into the blastocoel through a region on the embryo surface called the **blastopore.** As cells continue to *invaginate*, or move inward, a tube called the **archenteron** forms. The archenteron eventually becomes the adult gut. Which embryonic germ layer lines the archenteron?

## *Middle Gastrulation*

The archenteron continues to grow across the blastocoel. It takes on a bulblike appearance as the advancing portion swells.

**FIGURE 25.10**
Late gastrulation.

## *Late Gastrulation*

Cells at the leading edge of the advancing archenteron extend pseudopodia that attach to a specific region across the blastocoel (Figure 25.10). These cells continue to pull the archenteron across the blastocoel. As the tip of the archenteron approaches the opposite wall of the embryo, it bends to one side and fuses with surface cells. The site of fusion will eventually become the mouth of the embryo. What will be formed from the blastopore at the opposite end of the archenteron? (Recall from Lab Topic 19, Animal Diversity II that echinoderms are deuterostomes.)

What is the germ layer of cells on the surface of the embryo called?

The amoeboid cells at the leading edge and surrounding the archenteron are called *mesenchyme cells.* These cells later detach from the archenteron, proliferate, and form a layer of cells within the old blastocoel, now divided by the archenteron. This layer of cells will become the mesodermal germ layer.

## *Bipinnaria Larval Stage*

The archenteron of the gastrula differentiates into a broad **esophagus** leading from the **mouth** to a large oval **stomach** and on to a small, tubular **intestine.** All these structures will be visible in the bilaterally symmetric bipinnaria larva (Figure 25.11). Locate these structures in larvae on your slide. The larva is now self-feeding and begins to grow. It will later be transformed into the radially symmetric adult sea star.

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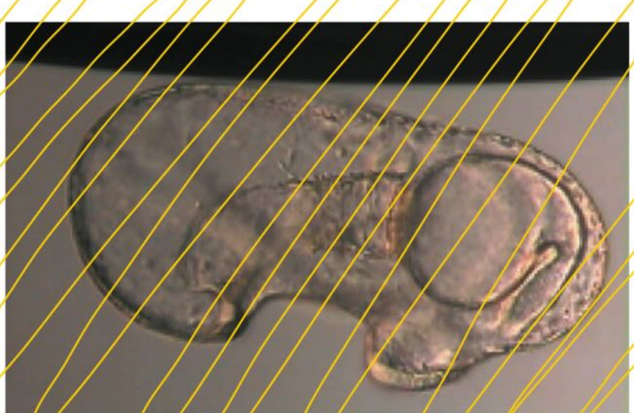

**FIGURE 25.11**

**Bipinnaria larva.** Identify the mouth, esophagus, stomach, intestine, and anus.

## Results

Draw stages of sea star development in the appropriate boxes in Figure 25.12 on the next page.

## Discussion

1. Sea urchins may release their eggs in tidal pool communities that include millions of eggs of different species at a given time. What is the advantage of species-specific sperm receptors in the plasma membrane and vitelline layer of an egg?

2. What type of egg does the sea star have? What evidence have you observed that supports your answer?

3. Describe the pattern of cleavage seen in the sea star and give the name for this type of cleavage.

4. At what stage in the sea star embryonic development did you first observe two layers of cells?

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# EXERCISE 25.2
# Development in an Amphibian

## Materials

video or film of early development in a salamander or some other amphibian

## Introduction

Amphibians are vertebrates that lay jelly-coated eggs in water or in moist areas on land. Common examples include frogs and salamanders. For most species, fertilization is external, with the male depositing sperm over the eggs after the female releases them. Internal fertilization takes place in some amphibians, however, in which cases the young are born in advanced developmental stages. Early development is similar in all species. After fertilization, the zygote begins cleavage followed by gastrulation, neurulation, and organogenesis.

In this exercise, you will study early development in an amphibian by observing a video or film presentation of some species, such as the salamander *Triturus alpestris* or the frog *Xenopus laevis*. The film or video shows dramatic time-lapse photography of cleavage, gastrulation, and neurulation.

## Procedure

1. Before viewing the film or video, complete Table 25.1 by defining terms commonly used when describing early embryos. (You may need to refer to your text.)
2. Read the questions in the Results section, view the film or video, and then answer the questions.

**TABLE 25.1** Common Terms Used in Embryology

| Term | Definition |
|---|---|
| Animal pole | |
| Animal hemisphere | |
| Equator | |
| Vegetal hemisphere | |
| Vegetal pole | |

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## Results

1. Would you describe the amphibian egg as isolecithal, moderately telolecithal, strongly telolecithal, or alecithal?

2. Describe the cleavage pattern. Is it holoblastic or meroblastic? Are cleavages synchronous or irregular? Can you detect any particular pattern in the cleavage? Where is the second cleavage plane in relation to the first?

3. Does the size of the embryo change as cleavage progresses?

4. Visually follow surface cells during gastrulation. Do they all move at the same rate? Describe gastrulation, comparing the process with that in the sea star. Notice the position of the blastopore and the yolk plug located in the blastopore.

5. During neurulation, do the neural ridges (folds) meet and fuse simultaneously along the entire length of the neural tube or do they close like a zipper?

## Discussion

1. Name at least two major differences in early development between the salamander and the sea star and describe factors responsible for these differences.

2. Compare the video of amphibian development with Figure 25.13. Label the following in the appropriate figure: **animal pole, vegetal pole, blastocoel, archenteron, yolk plug, neural plate,** and **neural folds.**

MB Student Media Video—Ch. 46: Frog Embryo Development

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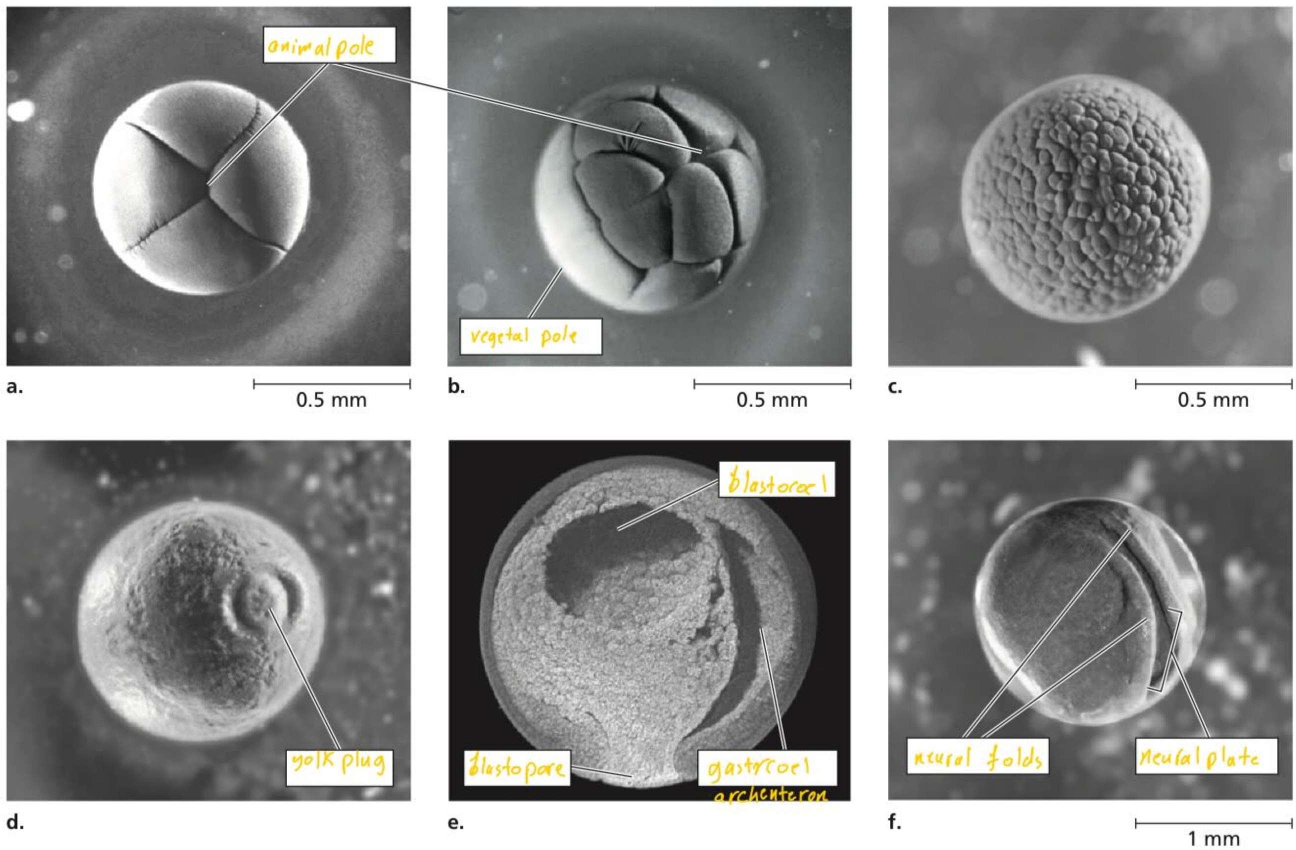


**FIGURE 25.13**
**Early amphibian development.** (a) and (b) Early cleavage. (c) Surface view of the hollow blastula in late cleavage. (d) Surface view of a gastrula. (e) Cross section of a gastrula. (f) Surface view of a neurula.

## EXERCISE 25.3
# Development in the Zebrafish

### Materials

small petri dishes
transfer pipettes
depression slides
embryo-rearing solution
clean toothpicks
fish embryos in various developmental stages (some on ice)
stereoscopic microscope
compound microscope

### Introduction

In this portion of the laboratory, you will observe living embryos in early developmental stages of a freshwater fish commonly known as the zebrafish, or zebra danio (*Danio rerio*) (Figure 25.14). The natural habitat of these popular aquarium fish is streams in India. Male and female fish are similar in appearance, but the male is generally smaller, with a streamlined body shape. The female is larger and broader than the male, especially when carrying eggs.