EXERCISE 5A

The Cell: Transport Mechanisms and Cell Permeability—Wet Lab

MATERIALS

Passive Processes

Diffusion of Dye Through Agar Gel

- ☐ Petri dish containing 12 ml of 1.5% agar-agar
- ☐ Millimeter-ruled graph paper
- ☐ Wax marking pencil
- ☐ 3.5% methylene blue solution (approximately 0.1 *M*) in dropper bottles
- ☐ 1.6% potassium permanganate solution (approximately 0.1 *M*) in dropper bottles
- ☐ Medicine dropper

Diffusion and Osmosis Through Nonliving Membranes

- ☐ Four dialysis sacs or small Hefty® "alligator" sandwich bags
- ☐ Small funnel
- ☐ 25-ml graduated cylinder
- ☐ Wax marking pencil
- ☐ Fine twine or dialysis tubing clamps
- ☐ 250-ml beakers
- ☐ Distilled water
- ☐ 40% glucose solution
- ☐ 10% sodium chloride (NaCl) solution
- ☐ 40% sucrose solution colored with Congo red dye
- ☐ Laboratory balance
- ☐ Paper towels
- ☐ Hot plate and large beaker for hot water bath
- ☐ Benedict's solution in dropper bottle
- ☐ Silver nitrate ($AgNO_3$) in dropper bottle
- ☐ Test tubes in rack, test tube holder

Text continues on next page.

myA&P *For practice quizzes on this lab, go to www.myaandp.com.*

OBJECTIVES

1. To define *differential permeability* and explain the difference between *active* and *passive processes* of cellular transport.
2. To define *diffusion (simple diffusion)* and *osmosis; isotonic, hypotonic,* and *hypertonic solutions; active transport; vesicular transport;* and *exocytosis, phagocytosis,* and *pinocytosis.*
3. To describe the processes that account for the movement of substances across the plasma membrane and to indicate the driving force for each.
4. To determine which way substances will move passively through a differentially permeable membrane (given appropriate information on concentration differences).

PRE-LAB QUIZ

1. Circle the correct term. A passive process, diffusion / osmosis is the movement of solute molecules from an area of greater concentration to an area of lesser concentration.
2. A solution surrounding a cell is *hypertonic* if:
   a. it contains fewer nonpenetrating solute particles than the interior of the cell.
   b. it contains more nonpenetrating solute particles than the interior of the cell.
   c. it contains the same amount of nonpenetrating solute particles as the interior of the cell.
3. Which of the following would require an input of energy?
   a. diffusion
   b. filtration
   c. osmosis
   d. vesicular transport
4. Circle the correct term. In pinocytosis / phagocytosis, parts of the plasma membrane and cytoplasm expand and flow around a relatively large or solid material and engulf it.
5. Circle the correct term. In active / passive processes, the cell provides energy in the form of ATP to power the transport process.

Because of its molecular composition, the plasma membrane is selective about what passes through it. It allows nutrients to enter the cell but keeps out undesirable substances. By the same token, valuable cell proteins and other substances are kept within the cell, and excreta or wastes pass to the exterior. This property is known as **differential,** or **selective, permeability.** Transport through the plasma membrane occurs in two basic ways. In **passive processes,** concentration or pressure differences drive the movement. In **active processes,** the cell provides energy (ATP) to power the transport process.

(materials list continued)

Experiment 1

- ☐ Deshelled eggs
- ☐ 400-ml beakers
- ☐ Wax marking pencil
- ☐ Distilled water
- ☐ 30% sucrose solution
- ☐ Laboratory balance
- ☐ Paper towels
- ☐ Graph paper
- ☐ Weigh boat

Experiment 2

- ☐ Clean microscope slides and coverslips
- ☐ Medicine dropper
- ☐ Compound microscope
- ☐ Vials of animal (mammalian) blood obtained from a biological supply house or veterinarian—at option of instructor
- ☐ Freshly prepared physiologic (mammalian) saline solution in dropper bottle
- ☐ 5% sodium chloride solution in dropper bottle
- ☐ Distilled water
- ☐ Filter paper
- ☐ Disposable gloves
- ☐ Basin and wash bottles containing 10% household bleach solution
- ☐ Disposable autoclave bag
- ☐ Paper towels

Diffusion Demonstrations

1: Diffusion of a dye through water

Prepared the morning of the laboratory session with setup time noted. Potassium permanganate crystals are placed in a 1000-ml graduated cylinder, and distilled water is added slowly and with as little turbulence as possible to fill to the 1000-ml mark.

2: Osmometer

Just before the laboratory begins, the broad end of a thistle tube is closed with a differentially permeable dialysis membrane, and the tube is secured to a ring stand. Molasses is added to approximately 5 cm above the thistle tube bulb, and the bulb is immersed in a beaker of distilled water. At the beginning of the lab session, the level of the molasses in the tube is marked with a wax pencil.

Filtration

- ☐ Ring stand, ring, clamp
- ☐ Filter paper, funnel
- ☐ Solution containing a mixture of uncooked starch, powdered charcoal, and copper sulfate ($CuSO_4$)
- ☐ 10-ml graduated cylinder
- ☐ 100-ml beaker
- ☐ Lugol's iodine in a dropper bottle

Active Processes

- ☐ Videotape showing phagocytosis (if available)
- ☐ Videotape viewing box

PEx PhysioEx™ 8.0 Computer Simulation on page PEx-5

Note to the Instructor: See directions for handling wet mount preparations and disposable supplies on page 33, Exercise 3.

Passive Processes

The two important passive processes of membrane transport are *diffusion* and *filtration*. Diffusion is an important transport process for every cell in the body. By contrast, filtration usually occurs only across capillary walls.

Recall that all molecules possess *kinetic energy* and are in constant motion. At a specific temperature, given molecules have about the same average kinetic energy. Since kinetic energy is directly related to both mass and velocity ($KE = \frac{1}{2}mv^2$), smaller molecules tend to move faster. As molecules move about randomly at high speeds, they collide and ricochet off one another, changing direction with each collision (Figure 5A.1).

Diffusion

When a **concentration gradient** (difference in concentration) exists, the net effect of this random molecular movement is that the molecules eventually become evenly distributed throughout the environment; that is, the process called diffusion occurs. Hence, **diffusion** is the movement of molecules from a region of their higher concentration to a region of their lower concentration. Its driving force is the kinetic energy of the molecules themselves.

There are many examples of diffusion in nonliving systems. For example, if a bottle of ether was uncorked at the front of the laboratory, very shortly thereafter you would be nodding as the ether molecules became distributed throughout the room. The ability to smell a friend's cologne shortly after he or she has entered the room is another example.

The diffusion of particles into and out of cells is modified by the plasma membrane, which constitutes a physical barrier. In general, molecules diffuse passively through the plasma membrane if they can dissolve in the lipid portion of the membrane (for example CO_2 and O_2). The unassisted diffusion of solutes (dissolved substances) through a differentially permeable membrane is called **simple diffusion.**

Certain molecules (for example, glucose) are transported across the plasma membrane with the assistance of a protein

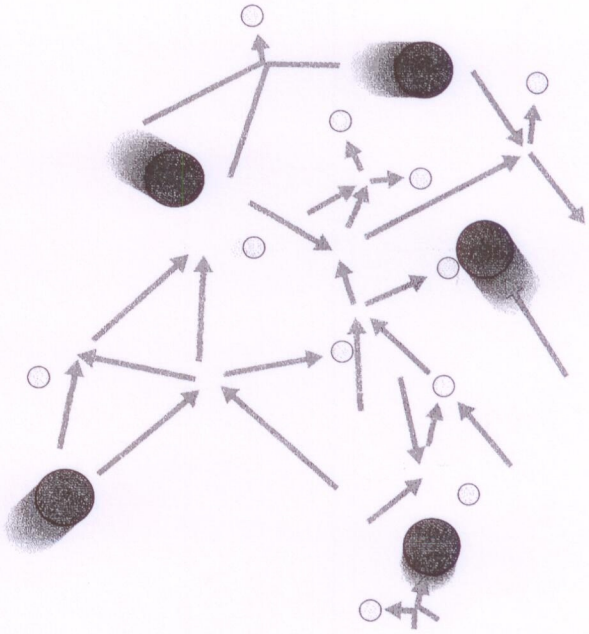


FIGURE 5A.1 Random movement and numerous collisions cause molecules to become evenly distributed. The small spheres represent water molecules; the large spheres represent glucose molecules.

carrier molecule. The glucose binds to the carrier and is ferried across the membrane. Small ions cross the membrane by moving through water-filled protein channels. In both cases, the substances move by a passive transport process called **facilitated diffusion.** As with simple diffusion, the substances move from an area of higher concentration to one of lower concentration, that is, down their concentration gradients.

Osmosis

The flow of water across a differentially permeable membrane is called **osmosis.** During osmosis, water moves down its concentration gradient. The concentration of water is inversely related to the concentration of solutes. If the solutes can diffuse across the membrane, both water and solutes will move down their concentration gradients through the membrane. If the particles in solution are nonpenetrating solutes (prevented from crossing the membrane), water alone will move by osmosis and in doing so will cause changes in the volume of the compartments on either side of the membrane.

Diffusion of Dye Through Agar Gel and Water

The relationship between molecular weight and the rate of diffusion can be examined easily by observing the diffusion of two different types of dye molecules through an agar gel. The dyes used in this experiment are methylene blue, which has a molecular weight of 320 and is deep blue in color, and potassium permanganate, a purple dye with a molecular weight of 158. Although the agar gel appears quite solid, it is primarily (98.5%) water and allows free movement of the dye molecules through it.

ACTIVITY 1

Observing Diffusion of Dye Through Agar Gel

1. Work with members of your group to formulate a hypothesis about the rates of diffusion of methylene blue and potassium permanganate through the agar gel. Justify your hypothesis.

2. Obtain a petri dish containing agar gel, a piece of millimeter-ruled graph paper, a wax marking pencil, dropper bottles of methylene blue and potassium permanganate, and a medicine dropper. (See Figure 5A.2.)

3. Using the wax marking pencil, draw a line on the bottom of the petri dish dividing it into two sections. Place the petri dish on the ruled graph paper.

4. Create a well in the center of each section using the medicine dropper. To do this, squeeze the bulb of the medicine dropper, and push it down into the agar. Release the bulb as you slowly pull the dropper vertically out of the agar. This should remove an agar plug, leaving a well in the agar.

5. Carefully fill one well with the methylene blue solution and the other well with the potassium permanganate solution.

Record the time. _____

6. At 15-minute intervals, measure the distance the dye has diffused from each well. Continue these observations for 1 hour, and record the results in the adjacent chart.

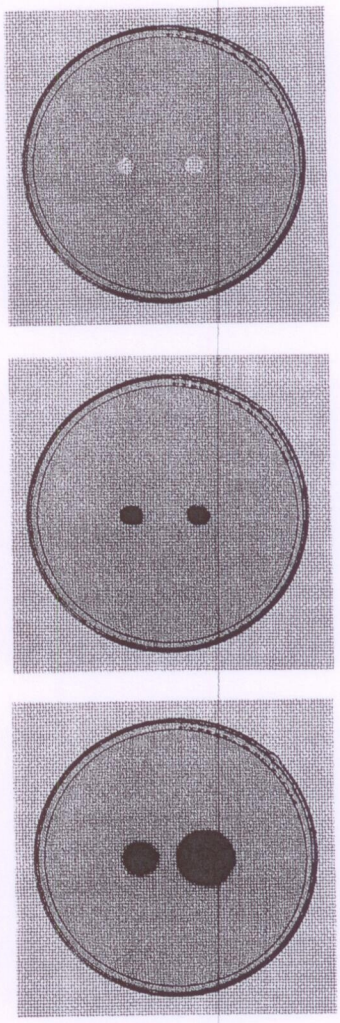


FIGURE 5A.2 Comparing diffusion rates. Agar-plated petri dish as it appears after the diffusion of 0.1 *M* methylene blue placed in one well and 0.1 *M* potassium permanganate placed in another.

Time (min)	Diffusion of methylene blue (mm)	Diffusion of potassium permanganate (mm)
15		
30		
45		
60		

Which dye diffused more rapidly? _____

What is the relationship between molecular weight and rate of molecular movement (diffusion)?

_____

Why did the dye molecules move? _____

_____

Compute the rate of diffusion of the potassium permanganate molecules in millimeters per minute (mm/min) and record.

_____ mm/min

Compute the rate of diffusion of the methylene blue molecules in mm/min and record.

_____ mm/min

7. Prepare a lab report for these experiments. (See Getting Started, page xv.)

Make a mental note to yourself to go to demonstration area 1 at the end of the laboratory session to observe the extent of diffusion of the potassium permanganate dye through water. At that time, follow the directions given next.

ACTIVITY 2

Observing Diffusion of Dye Through Water

1. Go to diffusion demonstration area 1, and observe the cylinder containing dye crystals and water set up at the beginning of the lab.

2. Measure the number of millimeters the dye has diffused from the bottom of the graduated cylinder and record.

_____ mm

3. Record the time the demonstration was set up and the time of your observation. Then compute the rate of the dye's diffusion through water and record below.

Time of setup _____

Time of observation _____

Rate of diffusion _____ mm/min

4. Does the potassium permanganate dye move (diffuse) more rapidly through water or the agar gel? (Explain your answer.)

_____

_____

ACTIVITY 3

Observing Diffusion and Osmosis Through Nonliving Membranes

The following experiment provides information on the movement of water and solutes through differentially permeable membranes called dialysis sacs. Dialysis sacs have pores of a particular size. The selectivity of living membranes depends on more than just pore size, but using the dialysis sacs will allow you to examine selectivity due to this factor.

1. Read through the experiments in this activity, and develop a hypothesis for each part.

2. Obtain four dialysis sacs, a small funnel, a 25-ml graduated cylinder, a wax marking pencil, fine twine or dialysis tubing clamps, and four beakers (250 ml). Number the beakers 1 to 4 with the wax marking pencil, and half fill all of them with distilled water except beaker 2, to which you should add 40% glucose solution.

3. Prepare the dialysis sacs one at a time. Using the funnel, half fill each with 20 ml of the specified liquid (see below). Press out the air, fold over the open end of the sac, and tie it securely with fine twine or clamp it. Before proceeding to the next sac, rinse it under the tap, and quickly and carefully blot the sac dry by rolling it on a paper towel. Weigh it with a

Data from Experiments on Diffusion and Osmosis Through Nonliving Membranes

Beaker	Contents of sac	Initial weight	Final weight	Weight change	Tests—beaker fluid	Tests—sac fluid
Beaker 1 ½ filled with distilled water	Sac 1, 20 ml of 40% glucose solution				Benedict's test:	Benedict's test:
Beaker 2 ½ filled with 40% glucose solution	Sac 2, 20 ml of 40% glucose solution					
Beaker 3 ½ filled with distilled water	Sac 3, 20 ml of 10% NaCl solution				$AgNO_3$ test:	
Beaker 4 ½ filled with distilled water	Sac 4, 20 ml of 40% sucrose solution containing Congo red dye				Benedict's test:	

laboratory balance. Record the weight in the data chart on page 56, and then drop the sac into the corresponding beaker. Be sure the sac is completely covered by the beaker solution, adding more solution if necessary.

- Sac 1: 40% glucose solution
- Sac 2: 40% glucose solution
- Sac 3: 10% NaCl solution
- Sac 4: Congo red dye in 40% sucrose solution

Allow sacs to remain undisturbed in the beakers for 1 hour. (Use this time to continue with other experiments.)

4. After an hour, boil a beaker of water on the hot plate. Obtain the supplies you will need to determine your experimental results: dropper bottles of Benedict's solution and silver nitrate solution, a test tube rack, four test tubes, and a test tube holder.

5. Quickly and gently blot sac 1 dry and weigh it. **(Note:** Do not squeeze the sac during the blotting process.) Record the weight in the data chart.

Has there been any change in weight? _____

Conclusions: _____

_____

_____

Place 5 ml of Benedict's solution in each of two test tubes. Put 4 ml of the beaker fluid into one test tube and 4 ml of the sac fluid into the other. Mark the tubes for identification and then place them in a beaker containing boiling water. Boil 2 minutes. Cool slowly. If a green, yellow, or rusty red precipitate forms, the test is positive, meaning that glucose is present. If the solution remains the original blue color, the test is negative. Record results in the data chart.

Was glucose still present in the sac? _____

Was glucose present in the beaker? _____

Conclusions: _____

_____

_____

6. Blot gently and weigh sac 2. Record the weight in the data chart.

Was there an *increase* or *decrease* in weight? _____

With 40% glucose in the sac and 40% glucose in the beaker, would you expect to see any net movement of water (osmosis) or of glucose molecules (simple diffusion)?

_____ Why or why not? _____

_____

7. Blot gently and weigh sac 3. Record the weight in the data chart.

Was there any change in weight? _____

Conclusions: _____

_____

Take a 5-ml sample of beaker 3 solution and put it in a clean test tube. Add a drop of silver nitrate. The appearance of a white precipitate or cloudiness indicates the presence of silver chloride (AgCl), which is formed by the reaction of $AgNO_3$ with NaCl (sodium chloride). Record results in the data chart.

Results: _____

Conclusions: _____

8. Blot gently and weigh sac 4. Record the weight in the data chart.

Was there any change in weight? _____

Did the beaker water turn pink? _____

Conclusions: _____

_____

Take a 1-ml sample of beaker 4 solution and put the test tube in boiling water in a hot water bath. Add 5 drops of Benedict's solution to the tube and boil for 5 minutes. The presence of glucose (one of the hydrolysis products of sucrose) in the bath water is indicated by the presence of a green, yellow, or rusty colored precipitate.

Did sucrose diffuse from the sac into the bath water? _____

Conclusions: _____

_____

9. In which of the test situations did net osmosis occur?

_____

In which of the test situations did net simple diffusion occur?

_____

What conclusions can you make about the relative size of glucose, sucrose, Congo red dye, NaCl, and water molecules?

_____

_____

_____

With what cell structure can the dialysis sac be compared?

_____

10. Prepare a lab report for the experiment. (See Getting Started, page xv.) Be sure to include in your discussion the answers to the questions proposed in this activity. ■

ACTIVITY 4

Observing Osmometer Results

Before leaving the laboratory, observe demonstration 2, the *osmometer demonstration* set up before the laboratory session to follow the movement of water through a membrane (osmosis). Measure the distance the water column has moved during the laboratory period and record below. (The position of the meniscus [the surface of the water column] in the thistle tube at the beginning of the laboratory period is marked with wax pencil.)

Distance the meniscus has moved: _____ mm

Did net osmosis occur? Why or why not?

_____

_____

_____ ■

ACTIVITY 5

Investigating Diffusion and Osmosis Through Living Membranes

To examine permeability properties of plasma membranes, conduct the following experiments. As you read through the experiments in this activity, develop a hypothesis for each part.

Experiment 1

1. Obtain two deshelled eggs and two 400-ml beakers. Note that the relative concentration of solutes in deshelled eggs is about 14%. Number the beakers 1 and 2 with the wax marking pencil. Half fill beaker 1 with distilled water and beaker 2 with 30% sucrose.

2. Carefully blot each egg by rolling it gently on a paper towel. Place a weight boat on a laboratory balance and tare the balance (that is, make sure the scale reads 0.0 with the weigh boat on the scale). Weigh egg 1 in the weigh boat, record the initial weight in the data chart below, and gently place it into beaker 1. Repeat for egg 2, placing it in beaker 2.

3. After 20 minutes, remove egg 1 and gently blot it and weigh it. Record the weight, and replace it into beaker 1. Repeat for egg 2, placing it into beaker 2. Repeat this procedure at 40 minutes and 60 minutes.

4. Calculate the change in weight of each egg at each time period, and enter that number in the data chart below. Also calculate the percent change in weight for each time period and enter that number in the data table.

How has the weight of each egg changed?

Egg 1 _____

Egg 2 _____

Make a graph of your data by plotting the percent change in weight for each egg versus time.

How has the appearance of each egg changed?

Egg 1 _____

Egg 2 _____

A solution surrounding a cell is **hypertonic** if it contains more nonpenetrating solute particles than the interior of the cell. Water moves from the interior of the cell into a surrounding hypertonic solution by osmosis. A solution surrounding a cell is **hypotonic** if it contains fewer nonpenetrating solute particles than the interior of the cell. Water moves from a hypotonic solution into the cell by osmosis. In both cases, water moved down its concentration gradient. Indicate in your conclusions whether distilled water was a hypotonic or hypertonic solution and whether 30% sucrose was hypotonic or hypertonic.

Data from Experiment 1 on Diffusion and Osmosis Through Living Membranes

Time	Egg 1 (in distilled H_2O)	Weight change	% Change	Egg 2 (in 30% sucrose)	Weight change	% Change
Initial weight (g)		—	—		—	—
20 min.						
40 min.						
60 min.						

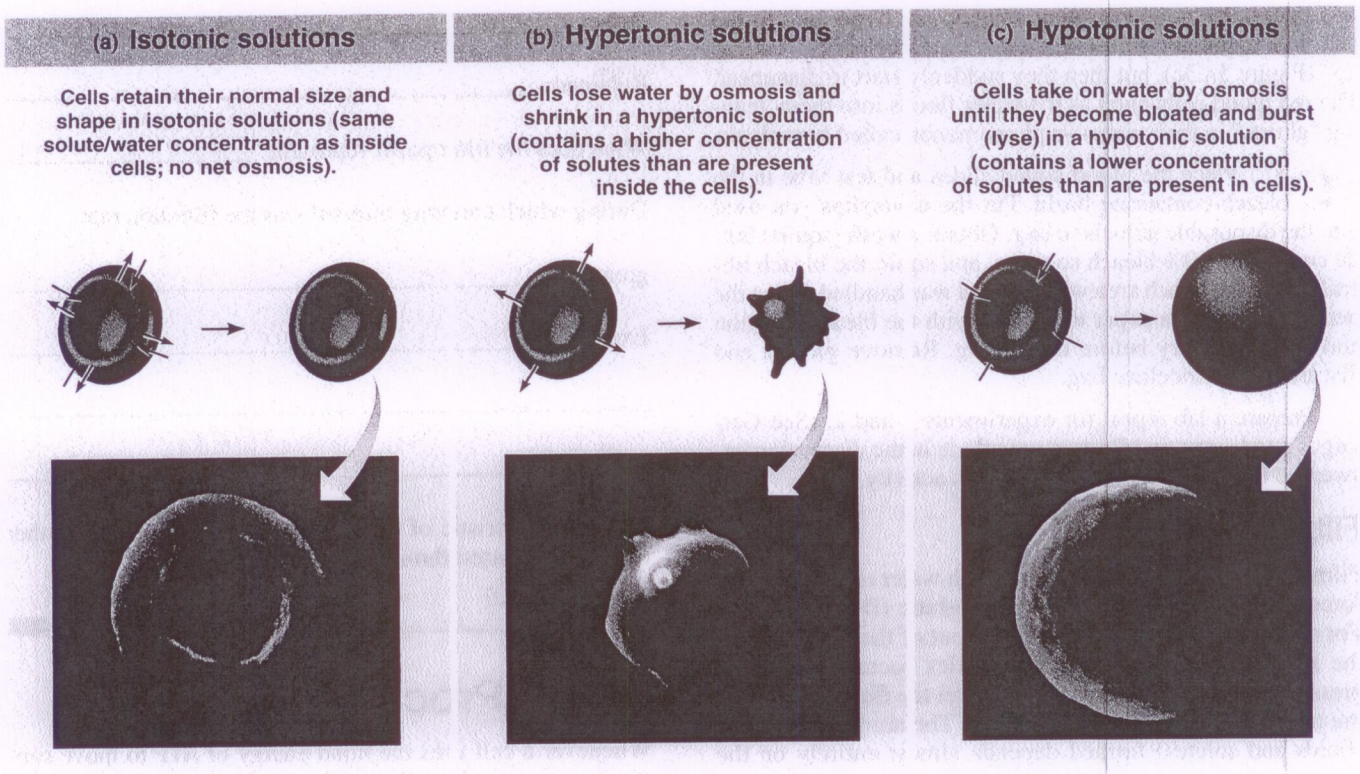


FIGURE 5A.3 Influence of isotonic, hypertonic, and hypotonic solutions on red blood cells.

Conclusions: _____

_____

_____

Experiment 2

Now you will conduct a microscopic study of red blood cells suspended in solutions of varying tonicities. The objective is to determine if these solutions have any effect on cell shape by promoting net osmosis.

1. The following supplies should be available at your laboratory bench to conduct this experimental series: two clean slides and coverslips, a vial of animal blood, a medicine dropper, physiologic saline, 5% sodium chloride solution, distilled water, filter paper, and disposable gloves.

⚠ Wear disposable gloves at all times when handling blood (steps 2–5).

2. Place a very small drop of physiologic saline on a slide. Using the medicine dropper, add a small drop of animal blood to the saline on the slide. Tilt the slide to mix, cover with a coverslip, and immediately examine the preparation under the high-power lens. Notice that the red blood cells retain their normal smooth disclike shape (see Figure 5A.3a). This is because the physiologic saline is **isotonic** to the cells. That is, it contains a concentration of nonpenetrating solutes (e.g., proteins and some ions) equal to that in the cells (same solute/water concentration). Consequently, the cells neither gain nor lose water by osmosis. Set this slide aside.

3. Prepare another wet mount of animal blood, but this time use 5% sodium chloride (saline) solution as the suspending medium. Carefully observe the red blood cells under high power. What is happening to the normally smooth disc shape of the red blood cells?

_____

_____

_____

This crinkling-up process, called **crenation,** is due to the fact that the 5% sodium chloride solution is hypertonic to the cytosol of the red blood cell. Under these circumstances, water tends to leave the cells by osmosis. Compare your observations to Figure 5A.3b.

4. Add a drop of distilled water to the edge of the coverslip. Fold a piece of filter paper in half and place its folded edge at the opposite edge of the coverslip; it will absorb the saline solution and draw the distilled water across the cells. Watch the red blood cells as they float across the field. Describe the change in their appearance.

_____

_____

Distilled water contains *no* solutes (it is 100% water). Distilled water and *very* dilute solutions (that is, those containing

less than 0.9% nonpenetrating solutes) are hypotonic to the cell. In a hypotonic solution, the red blood cells first "plump up" (Figure 5A.3c), but then they suddenly start to disappear. The red blood cells burst as the water floods into them, leaving "ghosts" in their wake—a phenomenon called **hemolysis.**

5. Place the blood-soiled slides and test tube in the bleach-containing basin. Put the coverslips you used into the disposable autoclave bag. Obtain a wash (squirt) bottle containing 10% bleach solution, and squirt the bleach liberally over the bench area where blood was handled. Wipe the bench down with a paper towel wet with the bleach solution and allow it to dry before continuing. Remove gloves, and discard in the autoclave bag.

6. Prepare a lab report for experiments 1 and 2. (See Getting Started, page xv.) Be sure to include in the discussion answers to the questions proposed in this activity. ■

Filtration

Filtration is a passive process by which water and solutes are forced through a membrane by hydrostatic (fluid) pressure. For example, fluids and solutes filter out of the capillaries in the kidneys and into the kidney tubules because the blood pressure in the capillaries is greater than the fluid pressure in the tubules. Filtration is not selective. The amount of filtrate (fluids and solutes) formed depends almost entirely on the pressure gradient (difference in pressure on the two sides of the membrane) and on the size of the membrane pores.

ACTIVITY 6

Observing the Process of Filtration

1. Obtain the following equipment: a ring stand, ring, and ring clamp; a funnel; a piece of filter paper; a beaker; a 10-ml graduated cylinder; a solution containing uncooked starch, powdered charcoal, and copper sulfate; and a dropper bottle of Lugol's iodine. Attach the ring to the ring stand with the clamp.

2. Fold the filter paper in half twice, open it into a cone, and place it in a funnel. Place the funnel in the ring of the ring stand and place a beaker under the funnel. Shake the starch solution, and fill the funnel with it to just below the top of the filter paper. When the steady stream of filtrate changes to countable filtrate drops, count the number of drops formed in 10 seconds and record.

_____ drops

When the funnel is half empty, again count the number of drops formed in 10 seconds and record the count.

_____ drops

3. After all the fluid has passed through the filter, check the filtrate and paper to see which materials were retained by the paper. (Note: If the filtrate is blue, the copper sulfate passed. Check both the paper and filtrate for black particles to see whether the charcoal passed. Finally, using a 10-ml graduated cylinder, put a 2-ml filtrate sample into a test tube. Add several drops of Lugol's iodine. If the sample turns blue/black when iodine is added, starch is present in the filtrate.)

Passed: _____

Retained: _____

What does the filter paper represent? _____

During which counting interval was the filtration rate greatest? _____

Explain: _____

_____

_____

What characteristic of the three solutes determined whether or not they passed through the filter paper?

_____ ■

Active Processes

Whenever a cell uses the bond energy of ATP to move substances across its boundaries, the process is an *active process.* Substances moved by active means are generally unable to pass by diffusion. They may not be lipid soluble; they may be too large to pass through the membrane channels; or they may have to move against rather than with a concentration gradient. There are two types of active processes: *active transport* and *vesicular transport.*

Active Transport

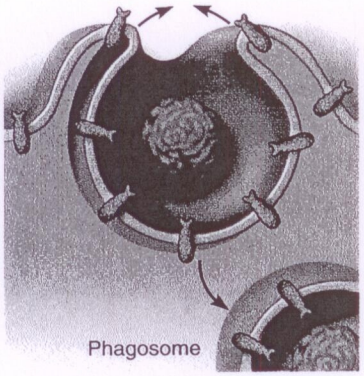
Like one form of facilitated diffusion, **active transport** requires carrier proteins that combine specifically with the transported substance. Active transport may be primary, driven directly by hydrolysis of ATP, or secondary, driven indirectly by energy stored in ionic gradients. In most cases the substances move against concentration or electrochemical gradients or both. Some of the substances that are moved into the cells by such carriers are amino acids and some sugars. Both solutes are insoluble in lipid and too large to pass through membrane channels but are necessary for cell life. However, sodium ions (Na^+) are ejected from cells by active transport. Carrier-mediated active transport is difficult to study in an A&P laboratory and will not be considered further here.

Vesicular Transport

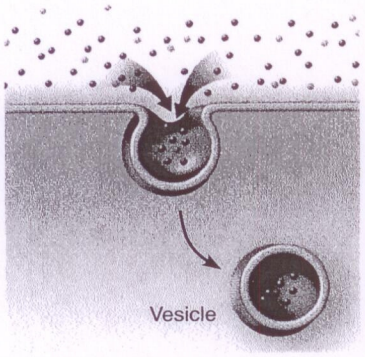
Large particles and molecules are transported across the membrane by **vesicular transport.** Movement may be into the cell **(endocytosis)** or out of the cell **(exocytosis).**

Most types of endocytosis utilize clathrin protein–coated pits to engulf the substance to be carried into the cell. Once engulfed, the substance is transported in the cell within a clathrin-coated vesicle.

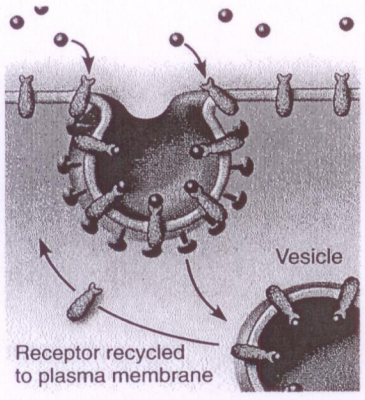
In **phagocytosis** (cell eating), parts of the plasma membrane and cytoplasm expand and flow around a relatively large or solid material (for example, bacteria or cell debris) and engulf it (Figure 5A.4a). The membranous sac thus



(a) Phagocytosis



(b) Pinocytosis



(c) Receptor-mediated endocytosis

FIGURE 5A.4 Three types of endocytosis. (a) In phagocytosis, cellular extensions (pseudopodia) flow around the external particle and enclose it within a vacuole. **(b)** In pinocytosis, dissolved proteins gather on the external surface of the plasma membrane, causing the membrane to invaginate and to incorporate a droplet of the fluid in a tiny vesicle. Most vesicles are protein coated. **(c)** In receptor-mediated endocytosis, plasma membrane proteins bind only with certain substances in regions of coated pits. The pits form protein-coated vesicles.

formed, called a *phagosome,* is then fused with a lysosome and its contents are digested. In the human body, phagocytic cells are mainly found among the white blood cells and macrophages that act as scavengers and help protect the body from disease-causing microorganisms and cancer cells.

In **pinocytosis,** also called **fluid-phase endocytosis,** the cell membrane sinks beneath the material to form a small vesicle, which then pinches off into the cell interior (see Figure 5A.4b). Pinocytosis is most common for taking in liquids containing protein or fat.

A more selective type of endocytosis uses plasma membrane receptors and is called **receptor-mediated endocytosis** (Figure 5A.4c). As opposed to the phagocytosis used by the body's scavenger cells, this type of endocytosis is exquisitely selective and is used primarily for cellular uptake of specific molecules, such as cholesterol, iron, and some hormones, and for transfer of substances from one side of the cell to the other.

ACTIVITY 7

Observing Phagocytosis

Go to the videotape viewing area and watch the videotape demonstration of phagocytosis (if available). ■

Note: If you have not already done so, complete Activity 2 ("Observing Diffusion of Dye Through Water," page 56), and Activity 4 ("Observing Osmometer Results," page 58).

NAME _____

LAB TIME/DATE _____

REVIEW SHEET EXERCISE 5A

The Cell: Transport Mechanisms and Permeability—Wet Lab

Choose all answers that apply to questions 1 and 2, and place their letters on the response blanks to the right.

1. Molecular motion _____.

   a. reflects the kinetic energy of molecules
   b. reflects the potential energy of molecules
   c. is ordered and predictable
   d. is random and erratic

2. Velocity of molecular movement _____.

   a. is higher in larger molecules
   b. is lower in larger molecules
   c. increases with increasing temperature
   d. decreases with increasing temperature
   e. reflects kinetic energy

3. Summarize the results of Activity 3, diffusion and osmosis through nonliving membranes, below. List and explain your observations relative to tests used to identify diffusing substances, and changes in sac weight observed.

   Sac 1 containing 40% glucose suspended in distilled water

   _____

   _____

   Sac 2 containing 40% glucose suspended in 40% glucose

   _____

   _____

   Sac 3 containing 10% NaCl suspended in distilled water

   _____

   _____

   Sac 4 containing 40% sucrose and Congo red dye suspended in distilled water

   _____

   _____

4. What single characteristic of the differentially permeable membranes *used in the laboratory* determines the substances that can pass through them? _____

In addition to this characteristic, what other factors influence the passage of substances through living membranes?

_____

_____

5. A semipermeable sac containing 4% NaCl, 9% glucose and 10% albumin is suspended in a solution with the following composition: 10% NaCl, 10% glucose, and 40% albumin. Assume that the sac is permeable to all substances except albumin. State whether each of the following will (a) move into the sac, (b) move out of the sac, or (c) not move.

glucose: _____ albumin: _____

water: _____ NaCl: _____

6. Summarize the results of Activity 5, Experiment 1 (diffusion and osmosis through living membranes—the egg), below. List and explain your observations.

Egg 1 in distilled water: _____

_____

_____

Egg 2 in 30% sucrose: _____

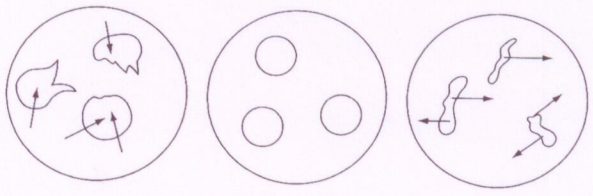
_____

_____

7. The diagrams below represent three microscope fields containing red blood cells. Arrows show the direction of net osmosis.

Which field contains a hypertonic solution? _____ The cells in this field are said to be _____. Which field contains an isotonic bathing solution? _____ Which field contains a hypotonic solution? _____ What is happening to the cells in this field? _____

_____



(a) (b) (c)

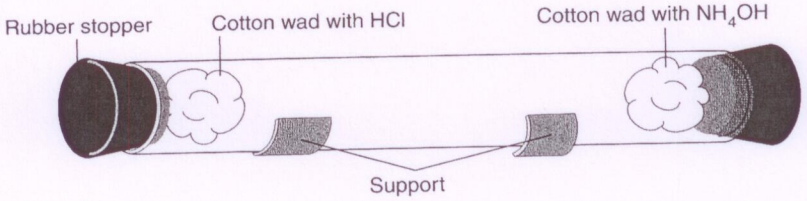
8. Assume you are conducting the experiment illustrated in the next figure. Both hydrochloric acid (HCl) with a molecular weight of about 36.5 and ammonium hydroxide (NH_4OH) with a molecular weight of 35 are volatile and easily enter the gaseous state. When they meet, the following reaction will occur:

$$HCl + NH_4OH \rightarrow H_2O + NH_4Cl$$

Ammonium chloride (NH_4Cl) will be deposited on the glass tubing as a smoky precipitate where the two gases meet. Predict which gas will diffuse more quickly and indicate to which end of the tube the smoky precipitate will be closer.

a. The faster-diffusing gas is _____.

b. The precipitate forms closer to the _____ end.



9. What determines whether a transport process is active or passive? _____

_____

10. Characterize membrane transport as fully as possible by choosing all the phrases that apply and inserting their letters on the answer blanks.

Passive processes: _____ Active processes: _____

a. account for the movement of fats and respiratory gases through the plasma membrane

b. explain solute pumping, phagocytosis, and pinocytosis

c. include osmosis, simple diffusion, and filtration

d. may occur against concentration and/or electrical gradients

e. use hydrostatic pressure or molecular energy as the driving force

f. move ions, amino acids, and some sugars across the plasma membrane

11. For the osmometer demonstration (Activity 4), explain why the level of the water column rose during the laboratory session.

_____

_____

12. Define the following terms.

diffusion: _____

_____

osmosis: _____

_____

simple diffusion: _____

_____

filtration: _____

_____

active transport: _____

_____

phagocytosis: _____

_____

fluid-phase endocytosis: _____

_____