Name _____ Class _____ Date_____

You Are a Medical Technologist

Testing Simulated Urine for Protein and Sugar

Question How does the detection of sugar or protein in the urine aid in the diagnosis of certain conditions?

Lab Overview In this investigation you will take on the role of a medical technologist as you test simulated urine samples from three "patients" to detect the presence of sugar and protein. You will compare test results from the samples with results from solutions containing known amounts of sugar or protein.

Introduction To start your investigation, you will create a model of a nephron tubule. Then you will use your model to simulate how filtration and reabsorption normally occur and how these processes could be affected by diabetes, high blood pressure, or kidney damage.

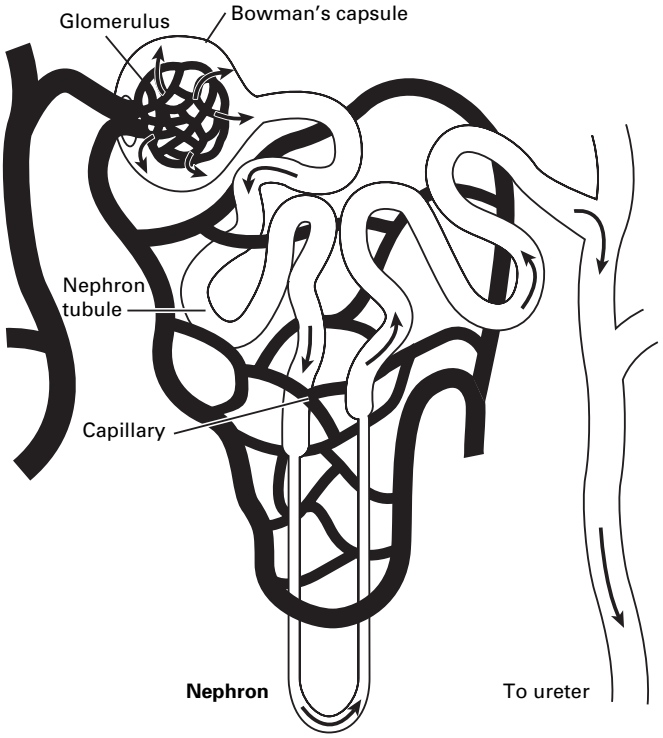
Background Sugars such as glucose are not normally present in urine. Recall that during filtration in the kidneys, blood pressure forces water, dissolved sugars, and other substances through the walls of the capillaries in the glomerulus. The fluid, called filtrate, collects in the nephron tubule. During reabsorption, the dissolved sugars (along with other substances) are normally reabsorbed from the tubule into the blood. However, if a person's blood sugar concentration is abnormally high, as occurs in the disease diabetes mellitus, some of the glucose is not reabsorbed. Instead, the excess glucose remains in the nephron tubule and eventually exits the body in urine. (People with diabetes mellitus have elevated blood glucose levels because they do not have enough insulin in their blood, or because their body cells do not respond to insulin. Insulin is a hormone secreted by the pancreas that increases the amount of glucose that enters body cells.)

Similarly, when the kidneys are functioning well, little protein is present in urine. Most proteins are too large to leave the blood by passing through the walls of the glomerulus, and so they never enter the nephron tubule. However, when a person has kidney damage or high blood pressure, proteins sometimes are forced from the blood into the tubule. These proteins are not reabsorbed and eventually exit the body in the urine. High blood pressure and abnormally high amounts of protein in the urine occur in a condition called preeclampsia (pree ih KLAMP see uh), which affects about 5% of women during middle to late pregnancy. When not treated promptly, preeclampsia can lead to seizures and other serious complications.

Medical technologists routinely analyze urine samples from patients to help doctors diagnose certain diseases. In this procedure, called urinalysis, a urine sample is usually tested for the presence of sugar, protein, and other substances. In this investigation you will take on the role of a medical technologist. First you will use Benedict's solution to test for glucose. Benedict's solution contains a copper compound that reacts with glucose. In the presence of glucose, Benedict's solution changes color from blue to dark red or orange. This reaction occurs best in hot water. Next you will use Biuret reagent to test for protein. Biuret reagent contains molecules that react with the bonds between amino acids in proteins. When proteins are present, Biuret reagent changes color from light blue to deep blue or purple.

Prelab Activity Use the materials listed below to build a model of a nephron. Study the diagram for guidance. The black arrows represent the direction that the filtrate moves through the nephron. Remember that glucose and proteins are present in the blood that enters the nephron. The glomerulus you design will need holes for molecules to pass out of the blood into the nephron tubule. Devise a way to model filtration, as well as reabsorption in tubules of healthy individuals, as well as reabsorption in individuals with diabetes or high blood pressure. Afterward, answer the Prelab Questions.

- 1 leg from brownish nylon stocking (nephron tubule)
- 1 leg from black or white nylon stocking (glomerulus and capillary)
- clear plastic cup (Bowman's capsule)
- scissors
- small candies or beans (sugar)
- large candies or beans (proteins)



Name _____ Class _____ Date _____

Prelab Questions

1. How does high blood pressure result in the presence of protein in urine? How did your model represent this effect?

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2. How do high blood glucose levels result in the presence of glucose in the urine? How did your model represent this effect?

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3. Preeclampsia is a disorder that affects some pregnant women and is characterized by high blood pressure and protein in the urine. During prenatal examinations, a doctor checks a pregnant woman's blood pressure, and also tests her urine for proteins. Why might both tests be necessary to diagnose preeclampsia?

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Materials

- 8 microcentrifuge tubes
- marker
- 10 transfer pipettes
- simulated urine samples from Patient 1, Patient 2, and Patient 3
- positive control solution for sugar
- Benedict's solution
- hot water
- plastic foam cup
- foam rack (optional)
- positive control solution for protein
- Biuret reagent
- tongs (or spoon)

Procedure

Part A: Testing for Glucose

1. Label three microcentrifuge tubes 1S, 2S, and 3S for the three patients. Label the fourth tube SC for the positive sugar control.
2. Use separate transfer pipettes to transfer 0.5 mL of each simulated urine sample to the appropriate microcentrifuge tube, and 0.5 mL of the positive sugar control to the tube labeled SC.
3. With a new transfer pipette, add 0.5 mL Benedict's solution to each tube. Do not allow the pipette to touch the samples. Tightly close the lid of each tube. **CAUTION:** *Benedict's solution is corrosive. Use extreme care when handling Benedict's solution to avoid getting it on your skin or clothing.*
4. Carefully add hot water to your plastic foam cup until it is half full. **CAUTION:** *Use extreme care when working with hot water. Do not let the water splash on your skin or clothes.*
5. Check each tube to make sure that it is tightly closed. Place the four tubes in the cup with the hot water. (If available, use a floating plastic foam rack. Push the tubes through the rack enough so that part of them is submerged in the water.)
6. Observe each tube after the time indicated by your teacher. In Data Table 1, record the color of each sample. If you need to take the tubes out of the water to see the results clearly, use tongs or a spoon to lift the tubes (or rack) out of the water.

Data Table 1: Glucose Testing

Sample	Results	
	Color	Positive or Negative?
1S (Patient 1)		
2S (Patient 2)		
3S (Patient 3)		
SC (control)		

Part B: Testing for Protein

1. Label three microcentrifuge tubes 1P, 2P, and 3P for the three patient samples. Label the fourth tube PC for the positive protein control.
2. With separate transfer pipettes, transfer 1 mL of each simulated urine sample into the appropriate microcentrifuge tube, and 1 mL of the positive protein control to the tube labeled PC.

Name _____ Class _____ Date_____

3. With a new transfer pipette add 3 drops of Biuret reagent to each tube and close the caps tightly. **CAUTION:** *Biuret reagent is corrosive. Use extreme care when handling Biuret reagent to avoid getting it on your skin or clothing.*
4. Mix each tube by tilting it upside down. Observe the tubes after 1 min. Record the results in Data Table 2.

Data Table 2: Results of Protein Testing

Sample	Results	
	Color	Positive or Negative?
1P (Patient 1)		
2P (Patient 2)		
3P (Patient 3)		
PC (control)		

Analysis and Conclusions

1. Summarize each patient's results.

2. What diagnoses do your findings support? What other tests would you perform or questions would you ask to confirm your diagnosis?

Extension

Although the pH values of urine vary from morning to night, the average pH of urine is 6.5 to 8. Acidic or alkaline urine may be an indication of various health problems. For example, the cells of a person with uncontrolled diabetes will break down fats for energy. This results in the release of acidic molecules, which are removed from the body in the urine. A diet too high in proteins could also lead to acidic urine because the waste products resulting from the breakdown of proteins are acidic. A bacterial infection in the bladder or ureters can lead to alkaline urine. Use pH test paper or a pH meter to determine the pH of several simulated urine samples provided by your teacher. What diagnoses might your findings support? What other tests or information would you want to have based on your findings?