12

URINALYSIS

LEARNING OBJECTIVES

- Perform a physical analysis, chemical analysis, and microscopic examination of urine.

TERMINOLOGY

pH of freshly voided urine = 4.6 - 8.0

Specific gravity of freshly voided urine = 1.003 - 1.035

Organized sediments – *These are biological.*

WBCs (pus)

RBCs

epithelial cells

casts

Unorganized sediments (crystals) – *These are chemical*

Renal calculus

OVERVIEW

The urinary system of the body performs a variety of tasks to assist in maintaining **homeostasis**. These tasks include the regulation of body fluid volume, electrolyte concentrations, and pH. Kidneys also remove metabolic waste products and toxins. All these tasks are accomplished as the kidneys produce **urine**.

Since urine begins as a filtrate of blood, its composition can provide valuable information regarding the constituents of the blood and the functioning of the kidney. For this reason, a **urinalysis**, the physical and chemical analysis of urine, is one of the most important diagnostic tools available to physicians.

URINE PRODUCTION

The process of urine production begins as blood passes through the **glomerulus**. Water, electrolytes, and small organic molecules are removed from the bloodstream by **filtration** and enter the glomerular capsule. From here, the filtrate passes through the proximal convoluted tubule (PCT), nephron loop, and distal convoluted tubule (DCT), eventually reaching the collecting duct. As the filtrate passes through the renal tubules, desirable substances such as water, ions, glucose, and amino acids are returned to the bloodstream by **reabsorption**. Additional substances such as H^+, K^+ and toxins are removed from the bloodstream and added to the filtrate by **secretion**. Reabsorption and secretion occur between the renal tubules and the peritubular capillaries and vasa recta. By the time the filtrate reaches the collecting duct, its composition has been set, and it is now urine. The entire process is summarized in ***Figure 12.1***.

The homeostatic mechanism of urine production is maintained by the renin-angiotensin system (RAS) and other hormones. Complete the chart below by writing the name of the organs that make these hormones:

Table 12.1: Hormones associated with blood volume and electrolyte balance.

Hormone	Origin
Renin	
Atrial Natriuretic Peptide/Factor	
Aldosterone	
Anti-diuretic hormone (vasopressin)	
Angiotensin 1	
Angiotensin 2	
Erythropoietin	

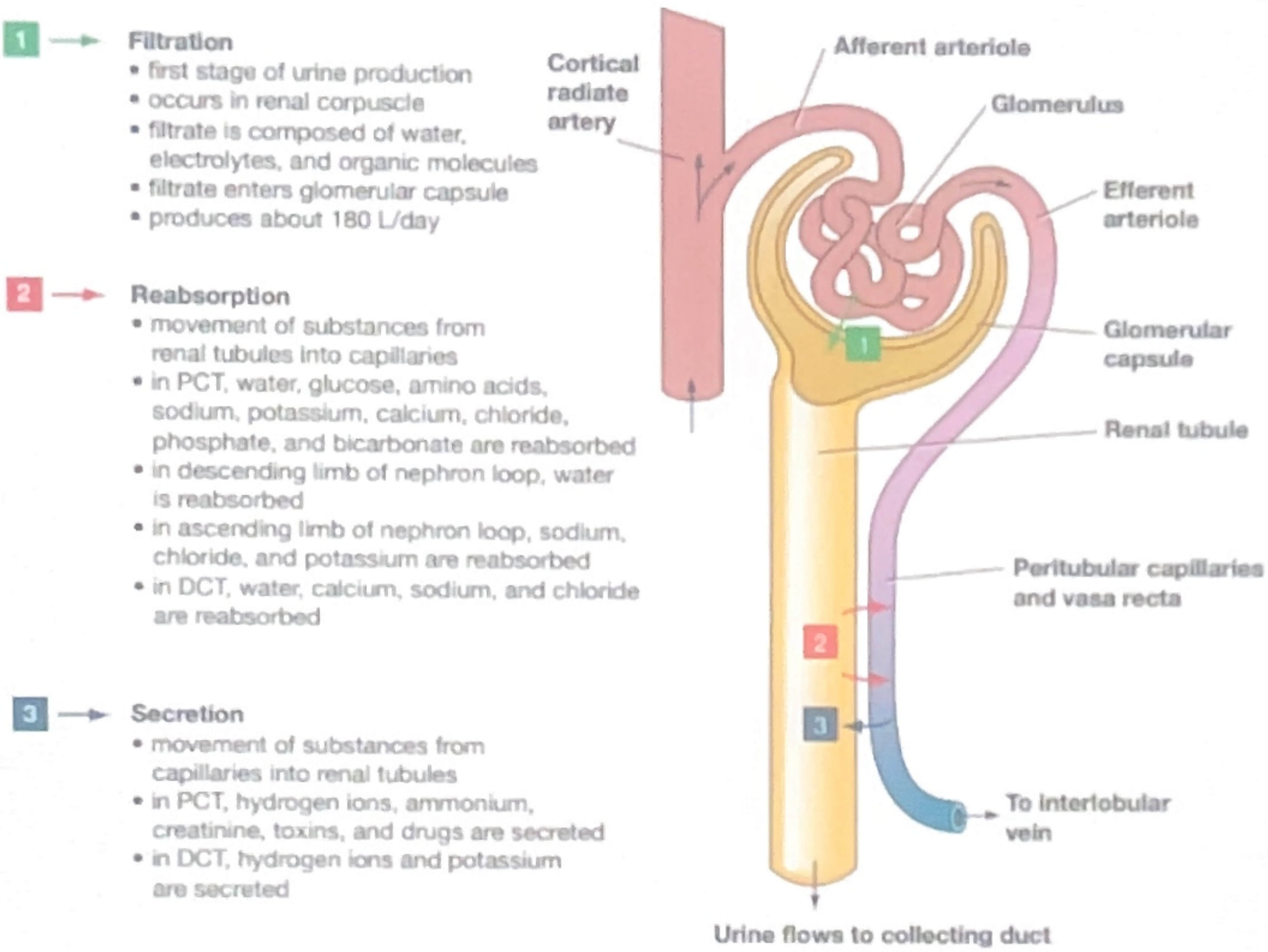

Figure 12.1: The stages of urine production.

URINALYSIS

In this set of exercises, you will perform a **physical analysis**, **chemical analysis**, and **microscopic examination**.

MATERIALS

- Disposable Urine collection cups
- Centrifuge tube
- Gloves
- Disposable collection cups
- Glass slide
- Coverslip
- Urinometer cylinder
- Refractometer
- Wax pencil
- Chemstrips
- Transfer pipette
- Paper towel
- Centrifuge
- Sedi-Stain
- Alcohol wipes
- Urinometer hydrometer
- Tray

Procedure

1. Proceed to the restroom with a clean disposable container.
2. Collect a midstream sample of about 50 mL of urine. Alternatively, use artificial urine.
3. Place the urine filled disposable container inside the white tray. During the experiments of this lab, try to stay within the perimeters of the white tray to avoid spilling and contamination of the lab.
4. **Work only with your own urine sample. DO NOT help other students or handle their urine samples.**
5. In case of a spill or any incident, report immediately to your instructor. Clean spill with 10% bleach solution.
6. Wear disposable gloves and goggles.
7. Use the following table for proper disposal.

Note: Before disposing containers, drain urine in the sink.

Red biohazard Bag	Sharps container	Wash/reuse
Chemstrips	Glass slide	Urinometer cylinder
Empty centrifuge tube	coverslip	Urinometer hydrometer
Transfer pipette		
Gloves		
Paper towel		
Disposable collection cups (empty)		

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Figure 12.2: Urinanalysis setup.

Experiment 1: Observe the color, transparency and odor

Record results in the chart on the following page.

Color – Normal urine colors range from pale yellow to amber, determined primarily by whether the urine is dilute (pale yellow) or concentrated (amber). The yellow color is due to urobilin pigment. Pathological colors include yellow-brown or green (caused by the presence of bile pigments) and red or dark brown (caused by the presence of blood, and/or eating large quantities of beets).

Transparency – Normal urine is clear to slightly cloudy. Cloudy urine is the result of an increase in the amount of sediment present. This sediment could contain pus from bacterial infections.

Odor – Check the odor by wafting the air above the sample. Normal urine has a highly characteristic odor described as "aromatic." Pathological urine specimens may have an ammonia smell caused by a bacterial infection or a fruity smell caused by the ketone bodies produced in a person with diabetes mellitus. Sometimes eating a diet containing some vegetables like asparagus can also cause urine to have a distinctive odor, similar to rotten eggs, onions and garlic.

Experiment 2: Determine specific gravity of the urine sample

- Open the specimen container and pour urine into a clean urinometer cylinder. Fill it about 3/4 full. (Do this in the white tray to avoid spilling on the floor or lab bench).
- Next place the urinometer hydrometer into the urine and make sure that the float is not touching the sides or the bottom of the cylinder.
- Position your eye at level of the urine surface (*Figure 12.2*). Determine which line on the stem of the hydrometer intersects the lowest level of the meniscus of the urine. Record your results in the chart on the next page. (Note to instructor: Keep a urinometer cylinder with distilled water and hydrometer, so students can compare the specific gravity differences.)
- You can also use the refractometer to determine specific gravity. Instructions for the procedure are provided by the instructor.
- After taking a reading, do not discard the sample. We will use it for the next set of experiments. (Pour 10 mL of urine into a centrifuge tube and screw the cap on tightly. Use wax pencil to place your initials on the tube. Give it to the instructor to centrifuge. The sediment will be used for experiment 4.

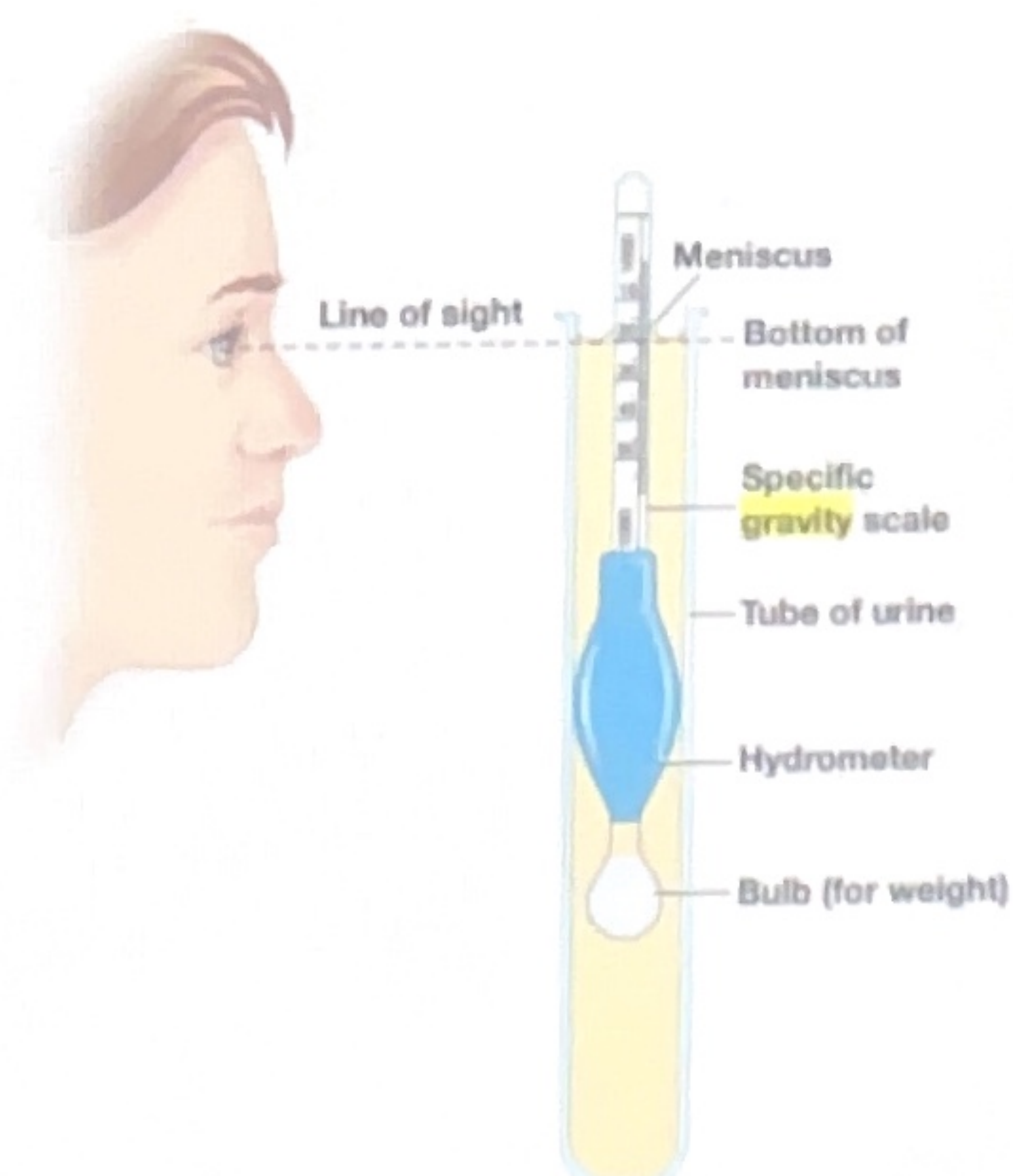

Figure 12.3: Reading a urinometer.

Record results from Experiments 1 and 2.

Characteristics	Normal Value	Record results
Color	Pale yellow	
Transparency	Transparent	
Odor	Aromatic	
Specific gravity	1.003-1.035	

Experiment 3: Use reagent Chemstrip to determine leukocytes, nitrite, urobilinogen, protein, pH, blood, specific gravity, ketone, bilirubin and glucose

1. Dip the Chemstrip into the urinometer, and remove it at an angle so that the edge of the strip touches the inside rim of the urinometer cylinder to remove any excess liquid. Alternatively, you can also place the Chemstrip flat on the paper towel inside the white tray and using a disposable pipette, add a drop of urine to each of the color pads. Consider adding drops to only a few pads at a time to allow time for reading results.
2. Wait for the length of time indicated by this scale before reading the colors/values.
3. Dispose the used Chemstrip in the biohazard container.

Reagent	Read Time	Normal Values	Record results
Glucose (GLU)	30 seconds	negative	
Bilirubin (BIL)	30 seconds	negative	
Ketone (KET)	40 seconds	negative	
Specific Gravity (SG)	45 seconds	1.003-1.035	
Blood (BLO)	60 seconds	negative	
pH	60 seconds	4.6 - 8.0	
Protein (PRO)	60 seconds	negative	
Urobilinogen (URO)	60 seconds	negative	
Nitrite (NIT)	60 seconds	negative	
Leukocytes (LEU)	120 seconds	negative	

Chemical analysis of urine tests for the presence of substances that should not be present. These include glucose, ketone bodies, albumin (one of the blood proteins), hemoglobin, and bilirubin. The presence of each of these constituents indicates a pathological condition.

- **Glycosuria** – Glucose easily filters into the glomerular capsule, but should be almost completely reabsorbed as the filtrate passes through the renal tubules. The presence of glucose in the urine, glycosuria, may indicate diabetes mellitus.

- **Ketonuria** – Ketone bodies are the byproduct of using fatty acids for energy production rather than glucose. Persons eating a high protein, low carbohydrate diet will have ketonuria, ketones in the urine. Pathologically, starvation or diabetes mellitus will also produce ketonuria.
- **Albuminuria** – Albumin is one of the large blood proteins. Under normal circumstances, it is too large a molecule to filter out of the blood and enter the glomerular capsule. Therefore, the presence of albumin in the urine, albuminuria, or proteinuria, indicates an abnormal increase in the glomerular permeability. This may be caused by hypertension, glomerulonephritis (a kidney infection of the glomerulus), trauma to the kidney, or kidney disease. Trace amounts of albumin in urine is normal.
- **Hemoglobinuria** – Hemoglobin is the oxygen transport protein found inside red blood cells. Therefore, it should not be present by itself in the urine. Its presence indicates a pathological condition that is damaging or rupturing red blood cells, such as hemolytic anemia.
- **Bilirubinuria** – Bilirubin is the waste product of removing damaged red blood cells from the circulation. The liver disposes of bilirubin by combining it with bile salts to make bile which is added to the small intestines. The presence of bilirubin in the urine, bilirubinuria, is an indicator of liver disease such as hepatitis or cirrhosis.

Experiment 4: Microscopic evaluation of urine sample

1. Pour 10 mL of urine from your specimen cup or urinometer into a clean centrifuge tube. Write your name on the tube. Place the tube inside the centrifuge, making sure the centrifuge stays balanced.
2. The instructor will start the centrifuge: 5 mins at 1850 rpm. While the centrifuge is running, start cleanup of your station.
3. Once centrifuge stops, collect your tube and gently (so the sediment does not get disturbed) drain the urine into the sink. Close the cap of the tube and take it back to your station.
4. Take out one clean glass slide and place it in the white tray.
5. Add one drop of Sedi-stain in the center of the slide.
6. Using the transfer pipette, collect the sediment at the very bottom of the centrifuge tube.
7. Put one drop of urine sediment on the slide (on top of the Sedi-stain drop). Carefully place a cover slip over it. (NOTE: Gently wipe off the edges and bottom of the slide to remove excess fluid.)
8. Examine the urine sediment under microscope. Use *Figure 12.4* as reference.
9. Once you are finished, dispose of the slide in the sharps container.
10. Discard the centrifuge tube and transfer pipette in the biohazard container.
11. Clean the microscope with alcohol wipe, roll the stage all way down, rotate nose piece to scanning objective, turn off the light and wrap the cord neatly. Put away the microscope.