

URINARY SYSTEM Anatomy & Physiology LAB

Upon completing this laboratory the student should be able to:

- 1. Identify the gross anatomy of the urinary system and the function of the organs*
- 2. Identify the gross anatomy of the kidney*
- 3. Identify the histology of the urinary system and relate it to the process of urine formation*
- 4. Define the nephron as the functional unit of the kidney and relate its anatomy to the processes of urine formation*
- 5. Define glomerular filtration, tubular reabsorption, tubular secretion and indicate where these processes occur in the nephron*
- 6. Trace the blood supply of the nephron from the arcuate artery to the arcuate vein*
- 7. Define dialysis, hemodialysis, dialysate, diffusion, and osmosis*
- 8. Identify the fluid compartments of the body*
- 9. Explain the similarities and differences between simple urine formation in the nephron and clinical hemodialysis*
- 10. Perform and interpret standard urinalysis tests*

Introduction

On the medial surface of the **kidney** is a bean-like indentation called the renal hilus. The renal artery and nerves enter at this point and the renal vein and the ureter exit the kidney here. The **ureter** is a muscular tube that carries urine from the kidney to the urinary bladder. Urine can move into the bladder by gravity or by peristaltic contractions of the ureter. The **urinary bladder** is a very muscular sac that can store urine. The bladder's muscle tissue can stretch considerably and also contract forcefully to push urine through the **urethra**.

The **kidney** has 3 major regions. The outermost the **renal cortex**, the middle **medulla** [contains series of **renal pyramids** - the tip or inner point of each renal pyramid is called the **renal papillae**] and the innermost kidney region the **renal pelvis**. The cortex region looks darker than the medulla's renal pyramids and the renal pyramids often look like a series of fine straight fibers. The renal pelvis is continuous with the ureters and forms a cavity within the kidney that collects urine from the renal papillae.

Each kidney contains 1-1.25 million functional units called **nephrons**. The nephrons and their associated blood vessels form the tissues of the renal cortex and renal medulla. Each nephron is composed of several specialized regions listed in order here: **glomerulus** ☒ **proximal convoluted tubule** ☒ **loop of Henle** ☒ **distal convoluted tubule** ☒ **collecting duct**.

Blood plasma is filtered across a very leaky capillary bed called a **glomerulus**. The filtration process is size selective, so all substances except cells and large plasma proteins should filter into the **renal capsule** that surrounds the glomerulus.

The glomeruli and renal capsules are located in the renal cortex of the kidney. The **proximal convoluted tubule** (also in the renal cortex) is the next section of the renal tubule. The proximal convoluted tubule reabsorbs the largest percentage of the filtrate that will be restored to the plasma (e.g. lots of water, sodium ions, etc.). Normally all of the filtered glucose & amino acids should be restored to the blood. The proximal tubule is

specially designed with lots of **microvilli** to increase its surface area to aid this reabsorption. Other sections of the nephron may secrete unwanted substances into the filtrate. The **loop of Henle** concentrates Na⁺ ions so that the base of the renal pyramid near the renal papillae is hyperosmotic compared to normal isotonic fluids. Straight sections of the nephron (the **loop of Henle** & the **collecting duct**) travel through the renal pyramids giving the pyramids a fibrous appearance. The collecting ducts have openings in the renal papillae so that the final produce (**urine**) exits the renal pyramid and enters the renal pelvis and then the ureter. The body produces many types of metabolic wastes that must be removed from the body to maintain homeostasis. The body may have an excess of sodium and phosphate ions that must be excreted.

The **catabolism** (breakdown) of proteins produces toxic nitrogenous wastes such as **ammonia** but our body converts much of that ammonia into a less toxic substance, **urea**. Protein catabolism also produces an excess of phosphoric acids. The breakdown of fats can produce acidic molecules called ketones. Other metabolic reactions and your diet may produce an excess of acids that lower extracellular fluid and plasma pH. The kidneys, along with the respiratory system, play a major role in maintaining blood pH at 7.35-7.45 by removing these common, normally occurring acidic waste products. If excessive alkaline (basic) substances are produced, the kidney can remove those molecules as well. Another major function of the kidney is to regulate the osmolarity of the blood and in turn the water balance of cells by regulating the loss or retention of both solutes & water.

This regulation of salt & water by the kidney uses several negative feedback mechanisms. Many scientists suggest that we drink ~ 8 cups of water / fluid per day to balance water losses, but this single value doesn't accommodate the variation in size and physical activity in the population. You can estimate what you need by dividing your body weight (in pounds) by 2. This number equals the number of ounces of water you should drink a day based on your body size. To adjust for exercise, weigh yourself before & after a vigorous or long duration activity. Then drink enough water to replenish what you lost during the exercise.

Most of us drink only 3-5 cups of water, so many of us are slightly dehydrated. Some of the symptoms of mild dehydration include: dry skin, loss of appetite, faster heart rate (from lowered blood pressure). More severe dehydration can cause headaches, nausea, impaired thermoregulation (you can't sweat), and ultimately impaired nerve and muscle function.

Antidiuretic hormone (ADH) is one of several hormones that are part of a negative feedback homeostasis loop that regulates blood pressure. ADH is secreted by the posterior pituitary in response to a decrease in plasma volume or an increase in plasma osmolarity, essentially dehydration. The **hyperosmotic** environment produced by the loop of Henle creates an environment where ADH can control the water reabsorption across the walls of the collecting duct. When ADH is present, the walls are permeable to water; when ADH is absent, the walls are impermeable to water. Thus ADH increases the reabsorption of water while decreasing urinary water losses. This helps return blood pressure &/or blood osmolarity to normal. Under the influence of ADH, the kidney produces a small volume of very hyperosmotic urine.

Alcohol inhibits the secretion of ADH, and is thus an important **diuretic**. **Caffeine** prevents ADH attachment to the collecting duct cells, also a diuretic. Caffeine can increase urination by 30% for up to 3 hours. Thus both

drugs can cause extreme dehydration, if you do not drink enough water to compensate for the increased water losses. Scientists often recommend that you drink an extra glass of water for each cup of coffee consumed.

ACTIVITY 1: Learn the following structures: [Models and dissection]

Kidney

renal capsule	renal cortex	renal medulla	renal column	renal pyramid
renal papilla	major calyx	minor calyx	renal pelvis	renal artery
renal vein				

Nephron

renal corpuscle	glomerulus	glomerular capsule	afferent arteriole	efferent arteriole
proximal convoluted tubule (PCT)		nephron loop (loop of Henle)		descending limb
ascending limb		distal convoluted tubule (DCT)		collecting duct (CD)
ureter		urinary bladder urethra		

ACTIVITY 2: What are the functions of the urinary system? List all you know.

Why is incontinence a phenomenon in the child under 18 to 24 months?

What events may lead to incontinence in an adult?

ACTIVITY 3: Gross internal anatomy of the sheep kidney: DISSECTION PROCEDURE

1. Collect your dissecting tools and tray. Obtain a preserved sheep kidney.
2. You would notice adipose tissue (remnants of the adipose capsule) clinging to the renal capsule.
3. You should also notice a "pinched-in" area where the renal blood vessels and ureter are attached to the kidney. This is the renal hilus.
4. Remove the renal capsule – a smooth, transparent membrane that adheres tightly to the external aspect of the kidney.
5. Once the renal capsule is removed, you will be looking at the renal cortex.

6. Separate the renal blood vessels from one another and from the ureter. Generally, the tube with the most adipose around it is the ureter.
7. * Notice the histological differences (and similarities) between the renal arteries, renal veins, and ureter.
8. Make a frontal (coronal) section through the kidney. Identify the renal pyramids (and parts of the pyramids), the renal columns, the renal pelvis, renal cortex, ureter and any blood vessels that are present.
9. Identify the following structures in situ: Renal Capsule, Hilum, Renal Cortex, Renal Medulla, Renal Medullary Pyramid, Renal Column, Renal Papilla, Renal Pelvis / Sinus, Major Calyx, Minor Calyx, Ureter, Renal Artery, Renal Vein,
10. Clean the dissection kit, tray and your workplace. Dispose the dissected kidney in biohazard container.

ACTIVITY 4: trace a drop of blood from the time it enters the kidney via the renal artery until it leaves the kidney through the renal vein. Use arrows to show the direction of flow.

Renal artery, Arcuate Vein, Interlobular Artery, Efferent Arteriole, Arcuate Artery, Interlobar Vein, Afferent Arteriole, Interlobar Artery, Interlobular Vein

ACTIVITY 5: Trace the pathway of a molecule of creatinine [metabolic waste] from the glomerular capsule to the urethra. Arrange the following microscopic and / or gross structure in the order it passes through. Use arrows to show the direction of flow.

Renal Cortex, Ascending Limb of Loop of Henle, Proximal Convoluted Tubule, Glomerulus / Bowman's Capsule, Collecting Duct, Renal Medullary Pyramid, Descending Limb of Loop of Henle, Distal Convoluted Tubule, urinary bladder, major calyx, renal pelvis, urethra, ureter, renal papilla, major calyx,

ACTIVITY 6: Analyzing urine samples

A. Specific Gravity Comparisons

Specific gravity is the weight of 1.0 ml of urine compared to the weight of 1.0 ml of water. One milliliter of water weighs 1.0 g. As solutes are added, the specific gravity of a solution increases. Thus, this unit-less relative measure is a relative indicator of the osmolarity of a solution.

Method

1. There will be several salt or drink solutions on the front desk or the sample area.
2. pour 50 ml of each drink sample in a labeled beaker [this is important]
3. Record the required information about the composition of the samples on the worksheet.
4. Fill a urinometer cylinder to about 1 inch from the top with a salt solution or drink.
5. Hold the urinometer float by its stem & slowly insert it into the cylinder.
6. DO NOT WET the stem ABOVE the water line or an inaccurate reading will result.
7. Do not take a reading if the stem is stuck to the side of the urinometer glass.
8. Compare these specific gravity readings with the normal values for urine.
9. Compare the specific gravity readings of the salt solutions with the drinks.

Drink						
Specific gravity						
Sugar g / 8 oz						
NaCl mg / 8 oz						
KCl mg / 8 oz						
Caffeine present						

Urinalysis

Body chemistry does not just depend on the volume and composition of the food we eat. It's also influenced by what the kidneys keep and do not keep by the processes of reabsorption and secretion. Urine contains a great deal of information about body function. The kidneys continuously form urine, reabsorbing glucose, amino acids, water, and salts from the filtrate of blood plasma. Factors such as diet, physical activity, body metabolism, disease, time of day, and body position can influence the chemical composition of urine. Analysis of the urine, or **urinalysis**, offers a means of determining the composition of the extracellular fluid (ECF) and the how the kidneys are functioning. During this part of the lab, you will conduct a series of tests on a urine sample. You will observe and determine both physical and chemical characteristics of the urine.

The urine amounts that you eliminate over a 24-hour period varies widely. These amounts depend on the amount of fluid you consume and the rate of excretion through avenues other than the excretory system, such as through perspiration and solid waste formation. Urine volumes commonly range from 1 to 2 L per 24 hours.

Many disorders can be diagnosed in their early stages by detecting abnormalities in the urine. Abnormalities include increased concentrations of constituents that are not usually found in significant quantities in the urine, such as: **glucose, protein, bilirubin, red blood cells, white blood cells, crystals, and bacteria.** They may be present because:

1. there are elevated concentrations of the substance in the blood and the body is trying to decrease blood levels by “dumping” them in the urine,
2. kidney disease has made the kidneys less effective at filtering or, of an infection, as in the case of bacteria and white blood cells.

A complete urinalysis consists of three distinct testing phases:

1. **visual examination**, which evaluates the urine's color, clarity, and concentration;
2. **chemical examination**, which tests chemically for 9 substances that provide valuable information about health and disease; and
3. **microscopic examination**, which identifies and counts the type of cells, casts, crystals, and other components, such as bacteria and mucus, that can be present in urine.

A routine urinalysis usually consists of the visual and the chemical examinations. These 2 phases may be completed in the laboratory or doctor's office. A microscopic examination is then performed if there is an abnormal finding on the visual or chemical examination, or if the doctor specifically orders it.

How is the sample collected for testing in real life scenario?

Urine for a urinalysis can be collected at any time. The first morning sample is considered the most valuable because it is more concentrated and more likely to yield abnormalities if present. It is important to clean the genitalia before collecting urine. Bacteria and cells from the surrounding skin can contaminate the sample and interfere with the interpretation of test results. With women, menstrual blood and vaginal secretions can also be a source of contamination. Women should spread the labia of the vagina and clean from front to back; men should wipe the tip of the penis. As you start to urinate, let some urine fall into the toilet, then collect one to two ounces of urine in the container provided, then void the rest into the toilet. This type of collection is called a “midstream collection” or a “clean catch.”

- A urine sample will only be useful for a urinalysis if taken to the doctor's office or laboratory for processing within a short period of time. If it will be longer than an hour between collection and transport time, then the urine should be refrigerated or a preservative may be added.

In today's lab, we will be using simulated urine sample, not real urine! Four samples will be provided for visual and chemical analysis.

Normal characteristics of urine:

1. What is a typical normal volume of urine? 1-2 liters / 24 hours per normal adult, however, the amount per day varies considerably.
2. **Color:** Typically yellow-amber but varies according to recent diet and the concentration of the urine. Drinking more water generally tends to reduce the concentration of urine, and therefore cause it to have a lighter color.

COLOR	NORMAL/ ABNORMAL?	POSSIBLE CAUSES
Colorless	Normal	Recent fluid intake; diuretic use
Pale yellow	Normal	
Dark yellow	Normal	Concentrated urine – <i>due to dehydration</i> Carrots, Vitamin A, C, and Bs
Amber/Orange	Normal or Abnormal	Bilirubin – <i>due to bile duct blockage</i> Carrots Vitamins A & C Riboflavin
Yellow-green	Normal or Abnormal	Bilirubin oxidized to biliverdin
Green/Blue-green	Normal or Abnormal	Pseudomonas (bacterial) infection Chlorophyll-containing foods
Pink/Red	Normal or Abnormal	Red blood cells/hemoglobin – <i>tissue damage</i> Myoglobin (oxygen-carrier in muscle tissue) – <i>muscle damage (large amounts can cause kidney failure)</i> Beets, blackberries, rhubarb – <i>anthocyanins (pink pigment)</i>
Brown/Black	Abnormal	Heavy bleeding Hemoglobin oxidized to methemoglobin Myoglobin

3. **Smell:** The smell ("odor") of urine may provide health information. For example, urine of diabetics may have a sweet or fruity odor due to the presence of ketones (organic molecules of a particular structure). Generally fresh urine has a mild smell but aged urine has a stronger odor, similar to that of ammonia [bacterial breakdown of urea to ammonia].
4. **Turbidity:** transparent in freshly voided urine; will turn cloudy after standing; microbes, pus, epithelial cells or crystals may cause cloudiness in fresh urine
5. **Acidity:** The pH of normal urine is generally in the range 4.6 - 8, a typical average being around 6.0. Much of the variation is due to diet. For example, high protein diets result in more acidic urine, but vegetarian diets generally result in more alkaline urine (both within the typical range 4.6 - 8).
6. **Specific gravity:** This is the ratio of the weight of a volume of a substance compared with the weight of the same volume of distilled water. Given that urine is mostly water, but also contains some other substances dissolved in the "water", its density is expected to be close to, but slightly greater than, 1.0. This is true - the density of normal urine is in the range 1.001 to 1.035.

Experiment

Data collection on four simulated urine samples [provided] in the table below

Color:	yellow or red
Turbidity:	clear or cloudy
pH range:	3.0 – 4.0 or 4.6 – 8.0 or 8.5 – 9.5
Presence of glucose:	negative or positive
Presence of protein:	negative, trace or positive
Presence of ketone bodies:	negative, trace or positive
Presence of hemoglobin:	negative, trace or positive
Presence of leukocyte:	negative, trace or positive

Method:

1. Label each of the four 100 ml beakers as A, B, C and D
2. Pour 50 ml of each simulated urine sample numbered A, B, C and D in labeled beaker. You will have one normal [control] and three abnormal samples
3. Look at the urine samples in the beakers to determine the color and turbidity. Record the observation in the table below.
4. Using a urinometer, measure the specific gravity for each urine sample using instruction by the Instructor. DO NOT DISCARD the urine sample. Use it for the next test.
5. Use pH paper to find out the pH of each sample. While the strip is still wet, compare the color with the color chart on the pH strip container to determine the pH.
6. Urine test strips contain small squares of different reagent paper that change color when they contact specific reagents. Use separate test strip for each sample to measure the presence of absence of glucose, albumin [protein], hemoglobin [blood], leukocytes and ketone bodies. Check for matching color. Read and follow specific instructions on the bottle / container.

Observation / Test	sample 1	sample 2	sample 3	sample 4
Color				
Turbidity				
Specific gravity				
pH				
Glucose				
Albumin				
Blood				
Ketone bodies				

RESULT:

State the **ABNORMAL CONSTITUENT**, if any, in each sample

Sample A:

Sample B:

Sample C:

Sample D:

DISCUSSION:

For each urine sample state if the urine sample is normal or not. If abnormal, state the clinical condition. What has gone wrong?

Sample A: Possible condition-

Sample B: Possible condition-

Sample C: Possible condition-

Sample D: Possible condition-

Define the disorder related to selected abnormal constituents of urine

Abnormal constituent	Disorder and cause
	Glycosuria:
	Hematuria:
	Pyuria:
	Albuminuria:
	Ketonuria
	Urinary Tract Infections:

Explain the similarities and differences between simple urine formation in the nephron and clinical hemodialysis