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Principles of Biology II Lab Manual

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NATURAL SELECTION

In humans, cystic fibrosis is an inherited disease due to an autosomal recessive gene located on chromosome #7. In the most common defective allele, three base pairs are deleted and a single phenylalanine is missing. Affected individuals carry two of the recessive alleles for the disease (genotype ff) and as a result form extremely thick mucus in their respiratory systems and elsewhere in the body. Their lungs are susceptible to frequent infections and the disease is progressive and eventually fatal. Usually the victims die in their teens or early twenties, and so do not reproduce. Among whites one person in 20 is a carrier, which is an Ff heterozygote for cystic fibrosis.

Severe natural selection has operated on the gene pool for cystic fibrosis (f) and normal Alleles (F) over the centuries. The affected individuals do not reproduce and do not pass on their genes. New cases arise only when both parent are heterozygous or through new mutations in a "normal" parent.

In the following exercise, you will compare the effects of natural selection alone and natural selection plus negative eugenics on the frequency of the f allele in a model system. Among the children of marriages between heterozygous carriers of the f allele and genetically normal individuals, the frequency of the f allele should be 25%, the same as in the parents. We will use that frequency at the start of both this and the following experiments.

First Round = First Generation

In this experiment, there will be complete selection against the recessive genotype that is expressed as the affected cystic fibrosis phenotype. The homozygous (FF) and the heterozygous (Ff) genotypes both have the normal phenotype and are not selected against. Both will, therefore, contribute to the next generation.

- Obtain a small plastic bag and count out 75 light beads and 25 dark beads. These 100 beads represent your initial gene pool. This is the gene pool that would be generated by marriages between heterozygous carriers and homozygous normal individuals.
- Shake up the beads to simulate random mixing of the gametes during the first generation of reproduction.
- Reach into the bag and (without looking!) take out two beads: This is the first individual.
- Set this pair of beads aside and repeat 49 times until you have drawn all beads, arranging the FF, Ff, and ff genotypes in groups as you draw. This represents 50 individuals. Remember, individuals have two copies of each gene!
- Select against the homozygous (ff) individuals by removing all of their alleles from the pool.
    - You only will be left with the FF and Ff individuals.
- Count up the number of light and dark beads left in the pool and calculate the percentage and frequency of each. The frequency is determined by finding the total # of

beads, then divide by the # in each category.

- Enter the results in the Table at the end of this exercise under Generation 1. (For help and guidance refer to the sample chart included at the end of the exercise.)

Second Round = Second Generation

- Begin the second round by replenishing your beads so that you have 100 beads with the same percentages of light and dark beads that you had at the end of Round (Generation 1). In the sample example provided, the new percentages were 80% light and 20% dark, so you would add beads to give 80 light and 20 dark.
- Shake up the beads.
- Select 50 new pairs.
- Remove all the ff individuals.
- Calculate the percentages and frequencies of light and dark beads left after selection and enter the results in the table under Generation 2.

Third Round = Third Generation, etc.

- Begin the third round by replenishing your beads so that you have the new percentages and repeat as described under the second round until you have drawn and selected against all five generations.

When you have finished five rounds, you will be tired of drawing beads, but you will have enough data to make a meaningful graph of the decline in the frequency of the f allele over five generations.

- Use the grid provided at the end of this exercise to plot the frequency of the f allele versus the number of generations.

Practice Calculations for Round 1 (see below):

Follow these steps for each round:

Enter your results in the tables below. Always round off to the nearest percent. Always start with 100 beads. 75 white and 25 black beads are in the bag. Shake up bag and without looking remove all beads as pairs (i.e., 50 pairs). Remove all pairs that are both black; homozygous for cystic fibrosis. In the first table record the numbers you started with. The number of white beads after selection (which will be the same as column one you didn't take any out). Now record the number of black beads after selection. Say you had four pairs of black beads that is eight black beans gone. The new total is the new number of white plus the new number of black 75 + 17 = 92. The new frequency for black and white is the new number divided by the new total white 75/92 = 0.82 and black 17/92 = 0.18. Please note these percentages must always add up to 1.00. Check yourself. Restock the bag with 100 beads according to the new frequencies and repeat through three generations.

The First table demonstrates the practice calculations.

W = White, B = Black

	F (White)				f (Black)			
Rounds	**# to start**	**# W after selection**	**Total # W + B after selection**	**New Frequency**	**# to start**	**# B after selection**	**Total # W + B after selection**	**New Frequency**
Initial	75	75	92	0.82	25	17	92	0.18
Generation 1	82				18			
Generation 2								
Generation 3								
Generation 4								
Generation 5								

Fluctuation in Frequency of Cystic Fibrosis allele Due to Selection Against it. Removing homozygotes only.

	F (White)				f (Black)			
Rounds	**# to start**	**# W after selection**	**Total # W + B after selection**	**New Frequency**	**# to start**	**# B after selection**	**Total # W + B after selection**	**New Frequency**
Initial								
Generation 1								
Generation 2								
Generation 3								
Generation 4								
Generation 5								

NEGATIVE EUGENICS

In this experiment you will assume that affected individuals with the ff genotype will not reproduce. Also assume that one half of the heterozygous individuals will refrain from reproducing or from passing on the f allele. The remaining heterozygotes and homozygotes for the normal allele will contribute to the next generation.

- Obtain a small plastic bag with 75 light beans and 25 dark beads. You may also need a coin to flip. These 100 beads represent your initial gene pool. This is the gene pool that would be generated by marriages between heterozygous carriers and homozygous normal individuals.
- Shake up the beads to simulate random mixing of the gametes during the first generation of reproduction.
- Reach into bag and (without looking!!!) take out two beads.
- Set this pair of beads aside and repeat 49 times until you have drawn all beads, arranging the FF, Ff, and ff genotypes in groups as you draw.
- Select against the ff individuals by removing all of their alleles from the pool; select against the Ff individuals by removing one half of them from the gene pool. If you have an odd number of heterozygotes, flip a coin to decide whether to remove the last one. You will be left with one half of the Ff individuals and all of the FF individuals.
- Count up the number of light and dark beads left in the pool and calculate the percentage and frequency of each exactly as you did in the first part of the exercise.
- Enter the results in the appropriate table at the end of this exercise.

Fluctuation in frequency of Cystic Fibrosis allele: Negative Eugenics; Removing homozygotes and 50% of heterozygotes

	F (White)				f (Black)			
Rounds	**# to start**	**# W after selection**	**Total # W + B after selection**	**New Frequency**	**# to start**	**# B after selection**	**Total # W + B after selection**	**New Frequency**
Initial								
Generation 1								
Generation 2								
Generation 3								
Generation 4								
Generation 5								

Questions

1. **What is the chance that two heterozygous parents will produce an affected child?**

2. **Why might the frequency of the f allele fail to drop over time?**

3. **Did you see a rapid effect of natural selection alone on the frequency of the f allele in the first experiment? Did the allele disappear? Why not?**

4. **Did negative eugenics cause the frequency of the f allele to decline more rapidly in the second experiment? Why?**

5. **Why doesn't cystic fibrosis decrease and eventually disappear of its own accord?**

6. **In some diseases, the affected individual has the ability to reproduce. Suppose there was a genetic counseling program that was attempting to lower the frequency of such diseases through negative eugenics. Would you advise the counseling program to concentrate its efforts on the affected individuals, or should both the carriers and affected individuals receive counseling? Explain your answer, using your graphs as evidence.**

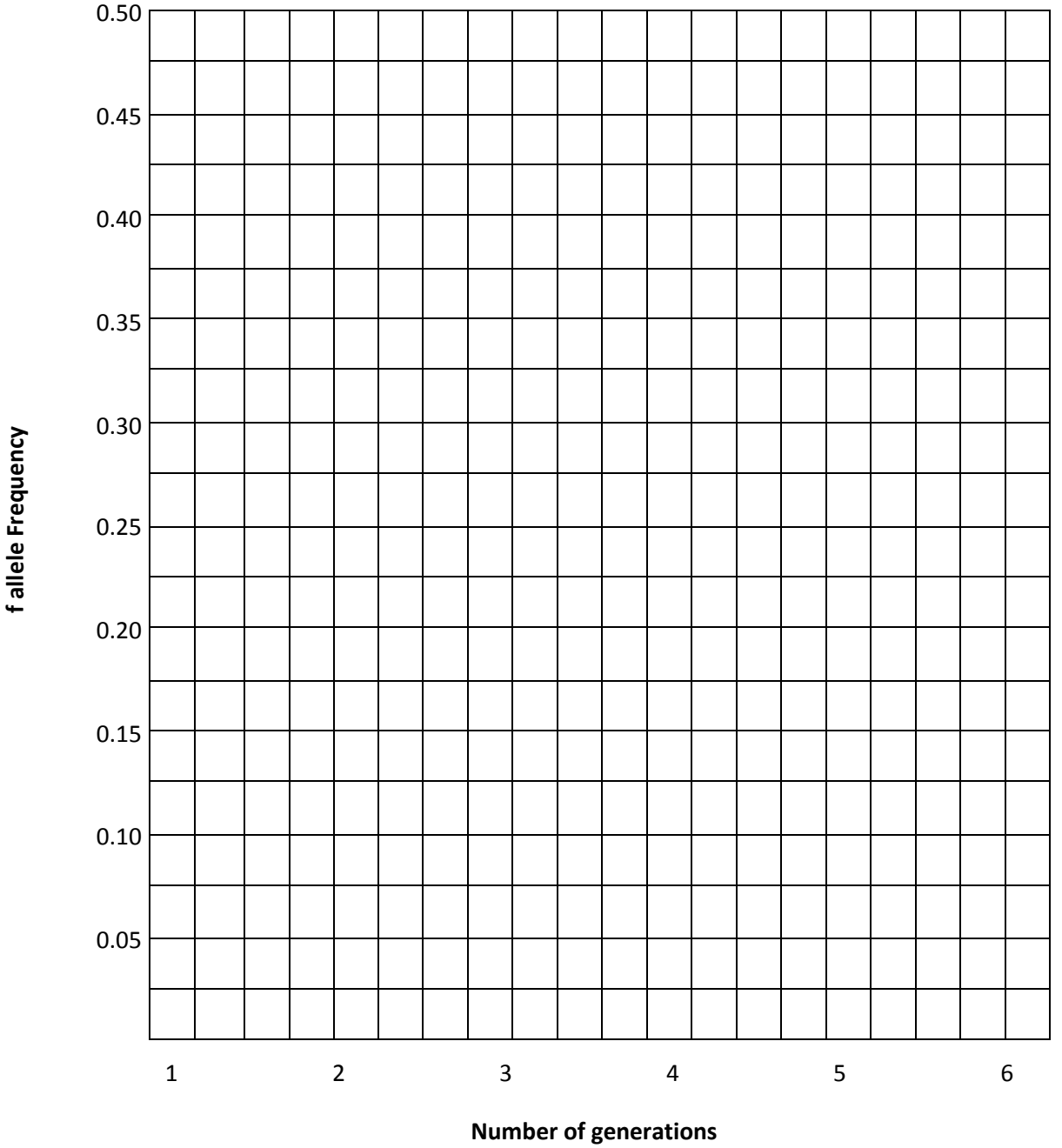
7. **What is the dependent variable in this experiment? HINT what changes?**

8. **What is the independent variable? Hint what causes it to change?**

9. **A test for the f allele in carriers exists. Should prospective parents consider taking it?**

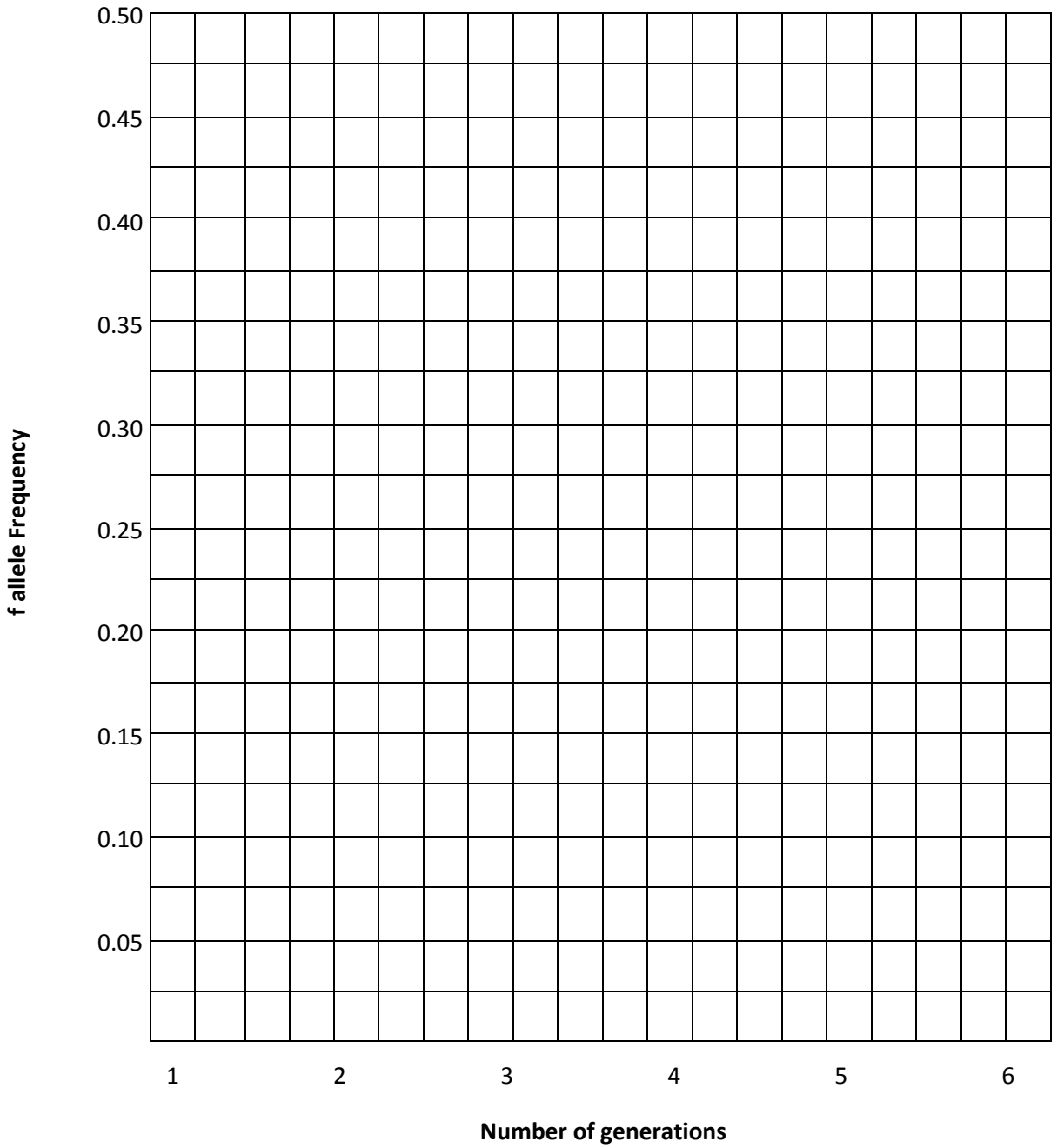
Selection against the f allele by phenotype.

Individuals with the recessive phenotype did not reproduce.



Selection against the f allele by phenotype and genotype.

Individuals with the recessive phenotype and one-half of the heterozygous carriers did not reproduce.



Nervous Systems Lab

Introduction: In this lab we will explore the anatomy & physiology of the nervous system. Nervous systems are unique to animals, and are critical for detecting and interpreting information, making decisions, and regulating body functions and movements. Nervous systems are constructed from neurons and glia. Neurons are the main functional cells, while glia play a variety of support roles.

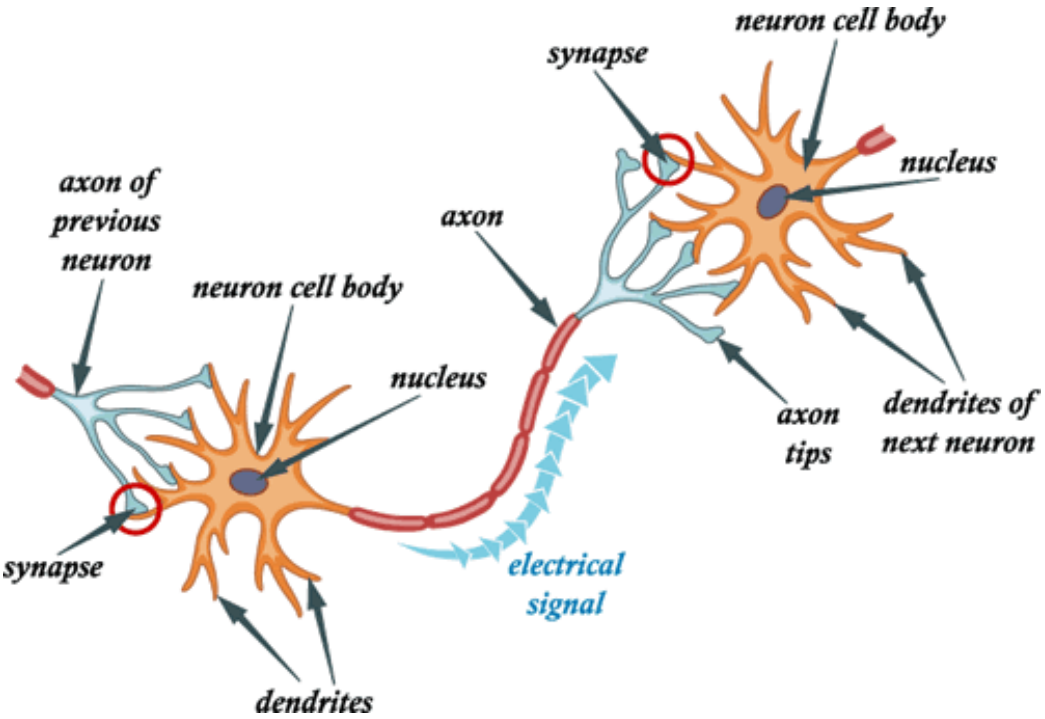
Nervous systems develop as interconnected networks of cells. The largest nervous organs are the brains of vertebrates, while animals as simple as sponges have a 'nerve net'. A critical concept related to nervous systems is consciousness, or 'self-awareness'. That idea will be discussed from one perspective in the next lab, distinguishing sensation from perception.

The exercises here will review cellular function and structure, and explore several basic neural networks within the larger body. We will use a combination of models and microscopic analysis, gross anatomical dissection, and physiological exercises to study nervous systems.

An overview of the components of Nervous Systems

Cellular Components	
Cell structures	**Cell functions**
Neurons	
Cells with membrane extensions, or *neurites* (axons {1 per neuron} & dendrites), interconnected at synapses. Identified by the extent of neurite branching: Multipolar = 2 or more dendrites Bipolar = 1 axon + 1 dendrite Unipolar = 1 neurite (functions as dendrite and axon, seen in specialized sensory neurons)	- Excitable cells based on maintenance of ion - concentration gradients (for Na^+ and K^+) and membrane ion channel proteins. - Membrane polarity (or *membrane potential, or voltage*) for a cell at rest is negative inside relative to out, based on an excess flow of K^+ out of the cell. - An Action potential is the activation of a neuron, and the membrane polarity reverses, due to an inward flow of Na^+.
Synapses – physical junctions between neurons, as well as between neurons and other cells, that allow for communication.	Chemical synapses (most common type) use an action potential to signal the exocytosis of neurotransmitter chemicals (over 100 different neurotransmitters known in humans). Each neurotransmitter requires a *receptor protein (multiple subtypes known for each different neurotransmitter).* Electrical synapses are direct membrane junctions between cells that allow continuations of action potentials.
Glia (or glial cells, or *neuroglia*)	
Oligodendrocytes & Schwann cells	Cells that wrap axons in *myelin sheaths* (glial membrane extensions) that increase action potential velocity
Astrocytes, Satellite cells & Ependymal cells	Cells that form borders between nervous tissues and other body tissues
Microglia	Immune cells that clear cellular debris from nervous tissues

Figure 1. *A drawing detailing some of the basic anatomical neural structures.*



Tissue and Organ Components of the Nervous System	
CNS – Central Nervous System – the brain & spinal cord in vertebrates.	PNS – Peripheral Nervous System – All other nervous tissue (nerves & ganglia).
Structures & Systems	**Functions**
White matter – myelinated axons in the CNS. Sometimes referred to as *tracts*, or *fasciculi* (or may have older, unique names).	Connections between CNS regions to coordinate functions. Allows integration of information.
Gray matter – neuron cell bodies in the CNS, includes cortical tissue (*e.g. cerebral cortex*) and nuclei (e.g. caudate nucleus assists with motor coordination).	Sites of information integration and decision-making, e.g. temporal cortex in the brain processes auditory signals and interprets speech.
Nerve – bundle of axons in the PNS	Can be classified as sensory (or *afferent)* for signals coming into the CNS from PNS, or motor (or *efferent*) for signals from CNS to PNS. Most nerves are mixed, and carry both sensory and motor information.
Ganglia – clusters of neuron cell bodies in PNS	Various functions controlling and monitoring body functions, e.g. dorsal root ganglia are sensory neurons clustered lateral to the spinal cord
Somatic Nervous System (or *voluntary)*	Nervous tissues related to conscious perception and voluntary body control.
Autonomic Nervous System (or *visceral)* – subdivided into the *Sympathetic* and *Parasympathetic* subdivisions. Nervous tissues related to *unconscious* perception and *involuntary* body control.	*Sympathetic* – regulates 'fight-or-flight', short term stress responses (e.g., increase heart rate, pupil dilation); *Parasympathetic* – regulates 'rest and digest' functions (e.g., decrease in skeletal muscle activity, increase in digestive function – liver, pancreas function)

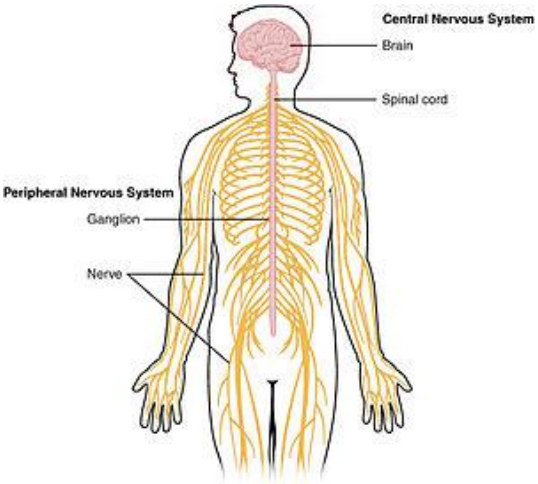
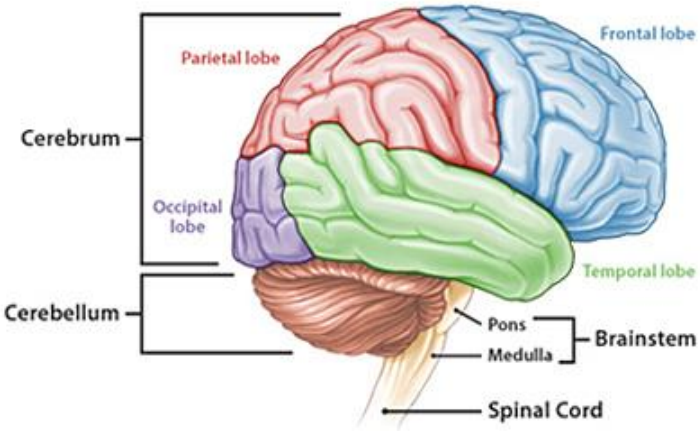


Figure 2 (A, left) – An overview of the anatomical structures in the body, and

An external view of brain anatomy, with major structures.



Exercises

Exercise 1. Examine the microscope slides for neuron anatomy. For each of the listed slides, note the listed structures:

a. Motor end plate (or myo-neural junction) – the synaptic connection between a motor neuron and a skeletal muscle cell. Structures include: multiple synapses (or *motor end plates*), axons, skeletal muscle cells with sarcomeres, myelin sheath of Schwann cells on axons.

b. Cerebellum – the 'little brain' posterior to the brain stem has folded white and gray matter that is distinguishable with the naked eye. Within the gray matter, 3 distinct neuron layers (granular, Purkinje, molecular) reveal organizational structures.

c. Spinal cord – a cross-section of spinal cord reveals peripheral white matter, and central gray matter. The small central canal contains ependymal glia. Large motor neuron cell bodies are located in the ventral gray matter – their axons form the ventral motor root of a spinal nerve. Sensory neuron cell bodies are in the dorsal root ganglia, and their axons form the dorsal sensory root of the spinal nerves.

Images

Figure 3. Myo-neural junction (or *motor end plate)* – This specialized synapse is critical to voluntary movement control.

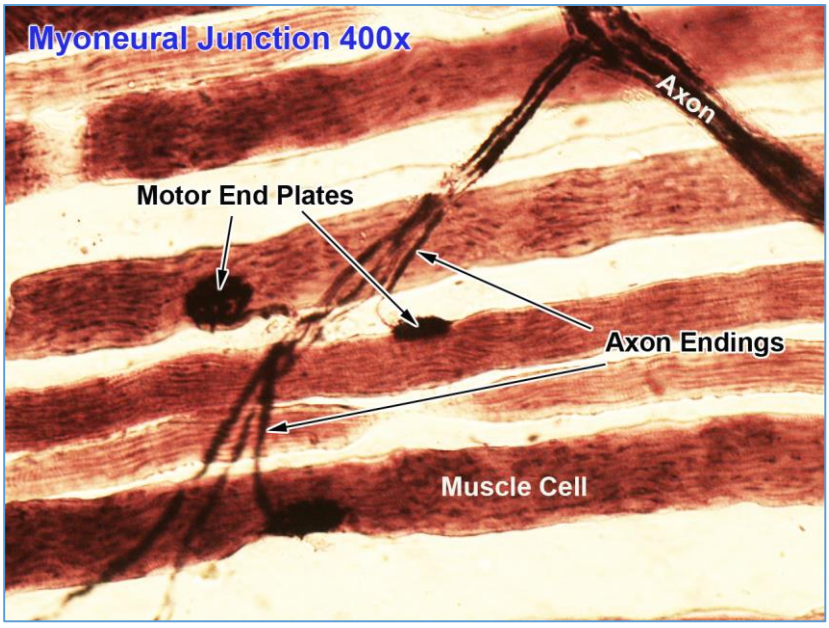


Figure 4a. Cerebellar gray matter, with large Purkinje neurons and multiple synapses (lined structures around Purkinje neuron cell bodies.

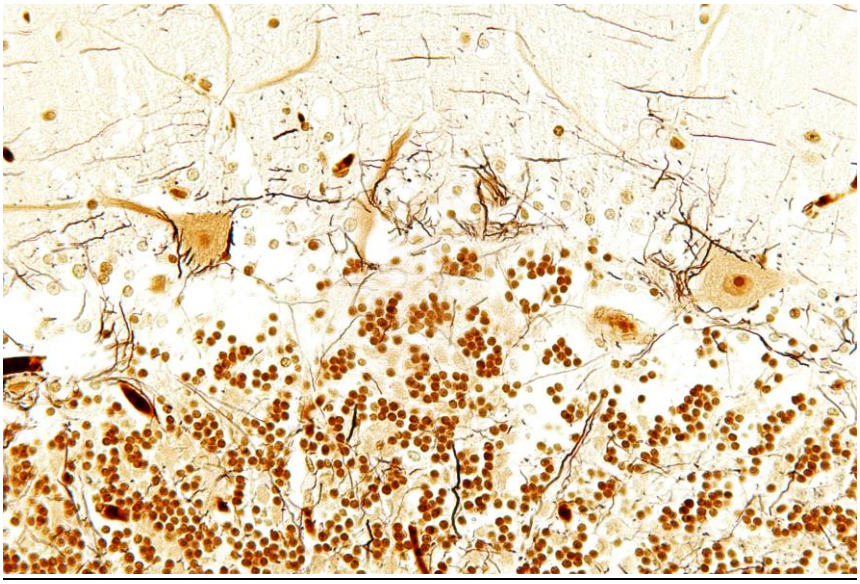


Figure 4b. A drawing outlining the neural connections in cerebellar gray matter. The large Purkinje neurons can have thousands of synapses on their dendrites in the molecular layer.

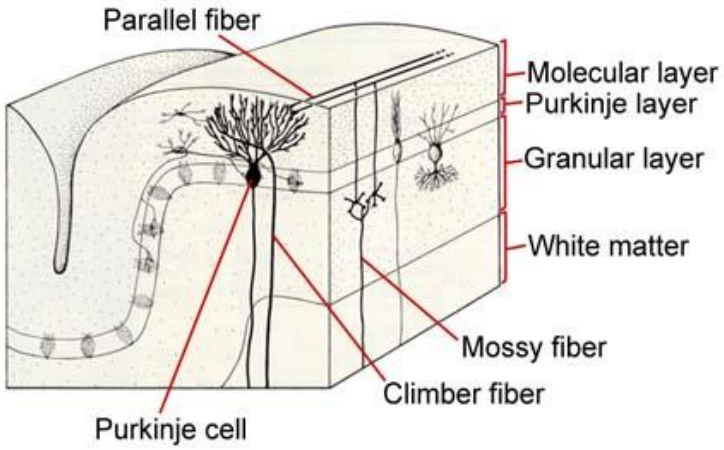
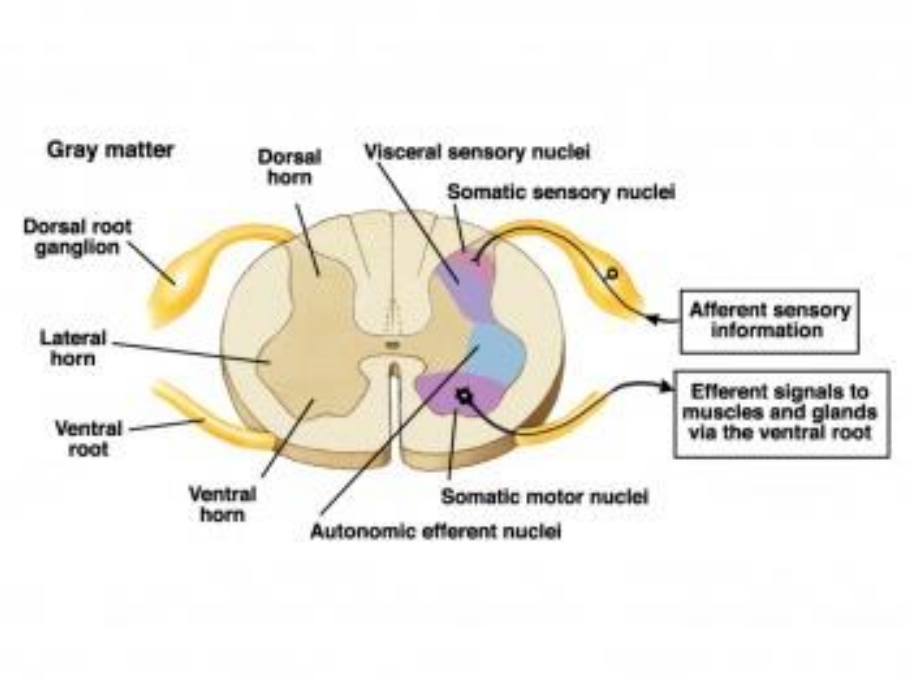


Figure 5. – The spinal cord sketch reveals the main anatomical structures.



Exercise 2. – Gross anatomy. Use the models and/or brain specimens to examine, dissect and identify the major anatomical regions of the brain. Not all structures may be present on the same model or specimen.

Cerebrum – cortex (or *cortical gray matter* – divided into lobes: frontal, temporal, parietal, occipital*)*, sulci & gyri (folds of cortex), cerebral hemispheres, white matter (includes corpus callosum), cerebral nuclei (including basal nuclei, thalamus, hypothalamus), olfactory bulbs & tracts, optic nerves, optic tracts, optic chiasma, ventricles, meninges;

Brain stem (includes medulla oblongata, pons, midbrain {includes sup. & inf. colliculi), cerebellum *(arbor vitae* is the white matter), cranial nerves (12 pairs total), spinal cord

Figure 6. Sheep brain anatomy.

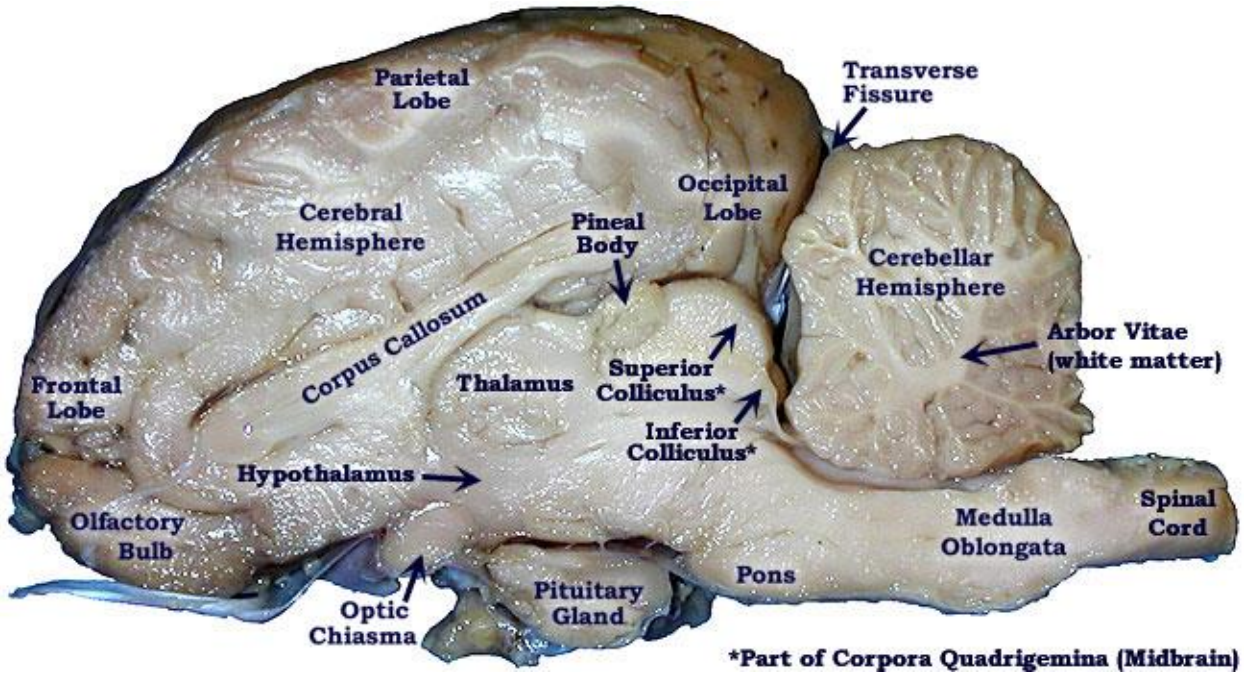
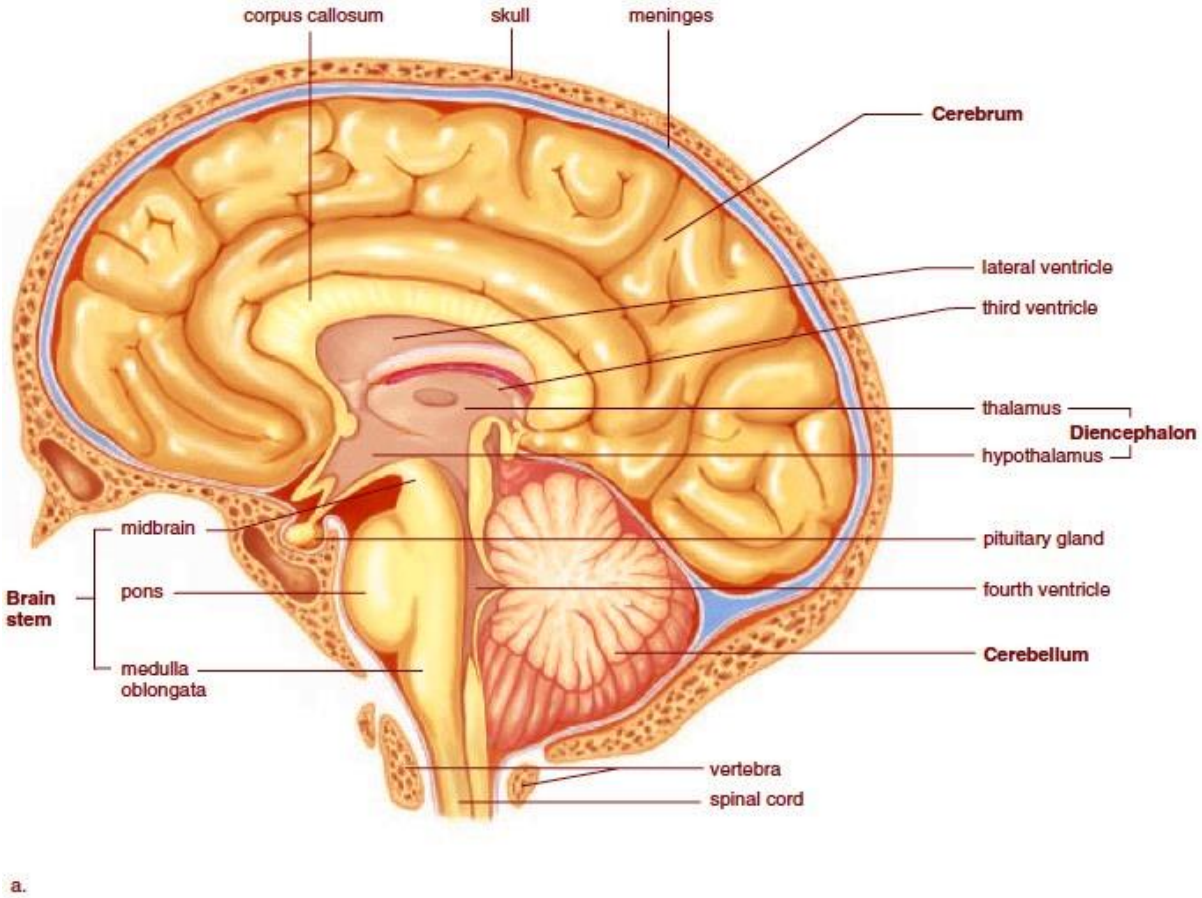


Figure 6A. Human brain anatomy



Exercise 3. Physiological functions of the nervous can be studied at a basic level by examining reflexes. Reflexes are the result of interconnected networks of neurons controlling specific functions.

3a. Reflex physiology - Using a meter stick, measure the speed of your neural network connecting your visual system and your somatic motor system. Work with a classmate, and perform the task 3 times each.

- Hold a meter stick vertically, with one end over the open hands of the first subject. The subject will have palms facing each other, ~ 6 inches apart.
- Hold the stick so that the scale starts at '0', even with the top of their hand, and the markings count upwards on the stick.
- Without warning, release the stick. When your classmate closes his hands to capture the stick, measure the distance the stick dropped by the markings at the top of your classmate's hands holding the stick.

Results:

Student A: Trial 1 _____ cm; Trial 2 _____ cm; Trial 3 _____ cm = _____ (average)

Student B: Trial 1 _____ cm; Trial 2 _____ cm; Trial 3 _____ cm = _____ (average)

- Based on acceleration due to gravity (9.8 m/sec^2), you can calculate the speed of the subject's reflexes by time (sec) = **sq. root of (2 x distance (cm)/9.8**).

- Convert your average distance to time: Student A - _____sec. Student B - _____sec.

- Compare the results among the class. Class range (maximum, minimum) _____ - _____ sec.

- There are at least 10 synapses involved in this response. Based on your results, what is the minimum amount of time that a neuron requires to receive and send a signal? _____ seconds.

3b. Autonomic reflexes – Using a flashlight, test the pupil reflex for light responses.

- Have the subject look away from any strong light sources.
- While watching their eyes, bring a light toward their eye from the left side.
- Observe changes in the left pupil, as well as the right pupil.
- Compare the responses in the two eyes. Repeat on the right side.
- Explain why the pupils reacted as they did (did both react in the same manner?).

Exercise 4. The somatosensory system is the largest sensing system in your body. This system produces sensory feedback whenever you come in physical contact with your environment. This sensory feedback includes body position (proprioception), sensing movement of your body and limbs (kinesthesis), pain (nociception), temperature, and finally touch. For this last sense you can map out touch receptor density in your skin using a simple technique. Most of the tactile feel we receive is gathered by four types of mechanoreceptors which are found in both layers of skin. The two receptors located near the top of the dermis, are called Merkel receptors and Meissner corpuscles. The other two mechanoreceptors located deep in the dermis and hypodermis layer and are the Ruffini and Pacinian corpuscles.

Working with a friend you are going to test each other's two-point threshold. A two-point threshold test seeks to find at what distance apart does a person perceive one point as two separate points. To test this, two points start together touching the skin. Incrementally they are pulled further apart and reapplied to the skin until the subject can clearly tell there are two different points.

You will take measurements from four different body part locations: Fingertip, Cheek, Forearm, and Calf. You will perform this experiment twice (ascending and descending with the drafting compass). Finally, the test subject must always keep their eyes closed!

Experiment Steps:

1. Before you start this experiment, be sure that you know how to use the compass. Practice touching your arm with it with the points at different distances (millimeter (mm)) apart. Use the ruler to precisely position the points apart from each other. It is important during the experiment that you touch your partner's skin with both tips at the same time; otherwise, they will easily be able to tell there are two points. Once you have the hang of it you are ready to begin!
2. Once your partner has closed their eyes, have them place their arm on the table with their palm facing up. Now take the compass, make sure the points are together (0 mm apart), and start with your partner's finger tip.
3. Touch the compass to their finger tip and ask them if they feel one point or two points. They should say one point because there is no distance between the points (0 mm). Remove the compass and increase the distance to 2 mm. Reapply the calipers, be sure to touch both tips at the same time, and ask again. If they still only feel one point, increase by another 2 mm and reapply.
4. Once your partner detects two points, move on to the next area of skin and repeat the process. Once you have completed the ascending or increasing distances now it’s time to repeat the process using the descending (decreasing distances) measurements. You can start with a distance of 7 mm and repeat the process by decreasing the distance by 2 mm each time. You can enter the data below. Remember to write down the distance that you feel only one point.

	ASCENDING				DESCENDING			
SUBJECT	FINGERTIP	CHEEK	FOREARM	CALF	FT	CHEEK	FA	CALF
#1								
#2								
AVERAGE								

1. Why can your fingertip detect such small distances between points while your arms and legs cannot?
2. Would you expect to see a difference in males vs. females for the four recorded areas? What about children vs adults?
3. Why doesn't your brain have the sensitivity of your fingertips all over your body?

Sensory Systems Lab

Introduction: In this lab we will explore the anatomy & physiology used for interpreting the environment both within and outside our bodies. The essential component is neurons, the major functional cells in nervous tissue. In many sensory organs, additional cells and tissues will contribute to the process of *signal transduction*.

Signal transduction is the process of a *receptor* detecting specific forms of matter or energy, and activating chemical and electrical changes in neurons. The neurons can then communicate with other neurons in the nervous system via synapses and networks to coordinate responses.

Receptor is a term used for the part of a sensory organ that detects the signal. 'Receptor' can refer to specific *protein molecules* which first interact with the matter or energy, or it can refer to the *cell(s)* that contains those proteins, or an *assembly of cells in the larger organ*.

Sensory Organs

The major sensory organs can be grouped based on various characteristics, i.e. what type of matter or energy they detect, and subsequently 'transduce' to produce our perceptions (e.g. vision, taste). Eventually there are electrical and chemical signals within our brains. Specific organs include:

Sensory Organ (major receptor)		**Matter/Energy detected by receptor**	**General anatomy & physiology**	**Perceived sensation**
Eye (retina)		Visible light (electromagnetic radiation)	Multilayered nervous sheet within the eye, with muscles and lenses for focusing	Vision
Ear	(cochlea)	Physical force (*sound*)	Flexible 'hair' cells that release signal molecules based on waves in fluid started by motion of the eardrum.	Hearing
	(semicircular canals, saccule, utricle)	Physical force	Flexible 'hair' cells that release signal molecules based on waves in fluid started by motion of the head.	Movement
Nose (olfactory epithelium)		Chemicals ('odorants')	A layer of neurons at the top of the nasal cavity	Smell (*'olfaction'*)
Tongue (taste buds)		Chemicals	Clusters of epithelial cells that release signals to neurons if specific chemicals are present (e.g. sodium ions)	Taste (*'gustation'*)
Skin	Mechanoreceptors	Physical force	Various neurons that respond to physical movements	Touch
	Thermoreceptors	Heat transfer	Specific neurons that respond to *increases* in temperature	Hot
	Thermoreceptors	Heat transfer	Specific neurons that respond to *decreases* in temperature	Cold

Muscles & Tendons	Physical force	Neurons respond to stretch and contraction of muscles & tendons	Body position (*proprioception*)
Most of the body	Various signals	Neurons respond to physical force, temperature & specific chemicals to warn of (potential) damage.	Pain (‘*nociception’*)

In order to investigate and understand sensory processes, we will investigate their anatomical structures (at macro- and microscopic levels) and physiological functions. An important distinction to consider is how humans can functionally separate Sensation (activation of the different receptors) as compared to Perception (the conscious awareness of the sensation). This distinction reveals how sensory deficits can result from damage in brain regions, even though the sensory organ is intact. Also, we may have perceptions that are only present in the brain, even though the sensory organs are silent.

VISION – The eye can focus light images on the retina using the cornea and the lens. Visible light only occupies a small portion of the *electromagnetic spectrum*. Other species and artificial technologies can detect other parts of this energy spectrum.

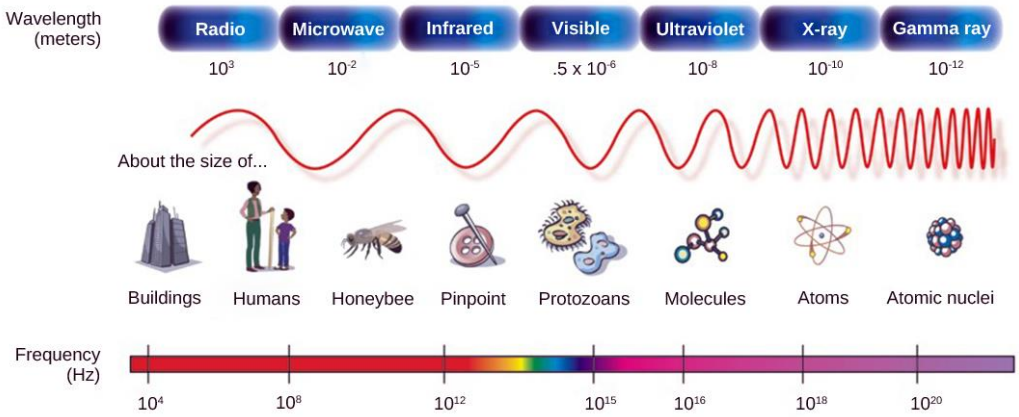


Figure S1. In the electromagnetic spectrum, visible light lies between 380 nm and 740 nm. (credit: modification of work by NASA)

Light passes through the eye ball via the cornea, pupil, and lens. The humors are fluids filling the anterior and posterior chambers of the eye.

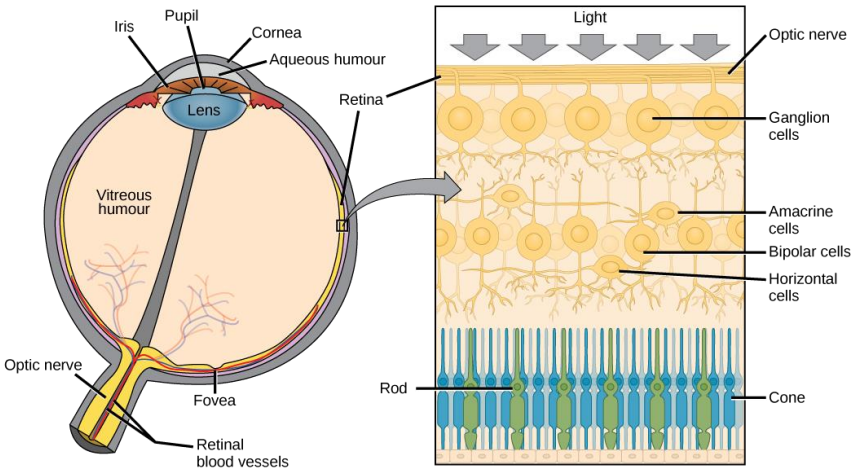


Figure S2. (a) The human eye is shown in cross section. (b) A blowup shows the layers of the retina.

The focused image is directed toward the fovea (or *fovea centralis*), which contains the highest density of photoreceptor neurons. Muscles in the iris alter pupil size to vary light entering the eye. Choroid body muscles surround the lens. They alter the lens to aid focusing.

Anatomy terms to know: Extra-ocular muscles, sclera, choroid, pigmented epithelium, fovea, vitreous humor, aqueous humor, iris, lens, choroid body, cornea, conjunctiva, optic nerve, blind spot (or *optic disc*), retina [retinal neurons - ganglion cells, amacrine cells, bipolar cells, horizontal cells, photoreceptors (rods, cones)], occipital (*visual*) cortex of the brain.

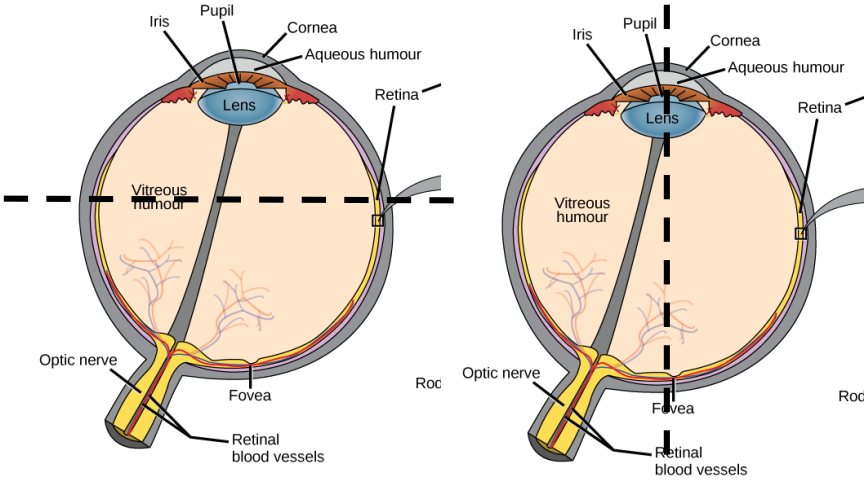
Possible Specimens & Models for examination: (sheep or cow) eyeballs for dissection, microscope slides of retina, models of eyes.

Exercise 1

You are responsible for identifying these major anatomical structures of the eye: sclera, choroid, pigmented epithelium, fovea, vitreous humor, aqueous humor, iris, lens, choroid boy, cornea, conjunctiva, optic nerve, blind spot (or *optic disc*), retina. You will have to identify these structures using both the sheep or cow eye and the models.

Dissection:

You will work in groups of two or three to dissect an eye. There are several ways to slice through an eye. Check with your instructor to determine which way they want you to cut the eye in half. Possible cuts include:



Note: To be able to best see the eye's structures, you should work very carefully. Many internal structures are delicate and tear easily such as the retina. Some structures are quite tough including the lens and sclera (in Latin, sclera means 'tough'!). Practicing careful dissecting is an important skill. Take your time! In order to get to the first layer, the sclera, you may have to clear away some fat and connective tissue. Ask for help if you need it.

Physiology: Light striking photoreceptor neurons activates networks of retinal neurons. One network of neurons in the retina sends signals to one ganglion cell. Action potentials from ganglion cells, whose axons form the optic nerves, represent *patterns of light*. Perception of the network of interconnected neuron signals is eventually perceived in the occipital cortex.

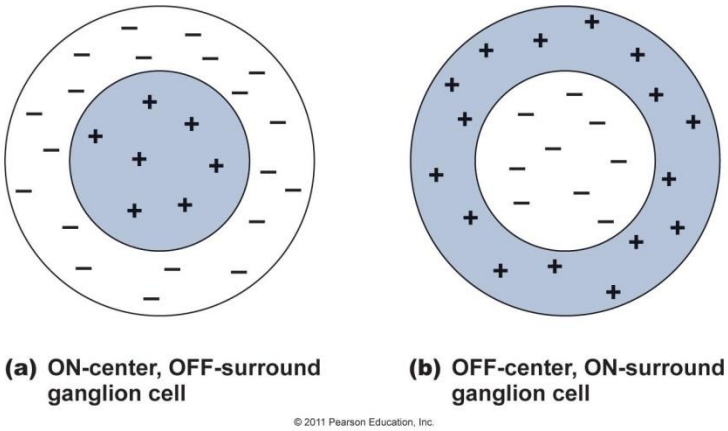


Figure S3. The receptive field for 1 ganglion cell.

In this example, the ON-center cell will send a maximum rate of action potentials along its axon (in the optic nerve) to the brain if the brightest light is striking photoreceptors near the center of its portion of the retina, and the surrounding photoreceptors in that portion are receiving minimal light. Eventually, patterns of ganglion cells signals are integrated in the brain (in the occipital cortex) to generate the perception of complex images.

Color vision results from the interaction of 3 sub-types of cone photoreceptors. They preferentially absorb light at different wavelengths, shown in the figure below.

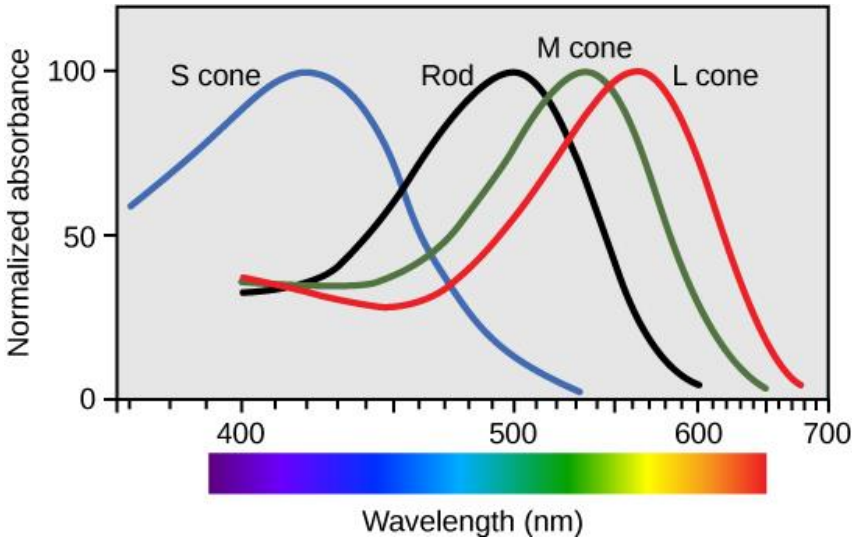


Figure S4**.** Human rod cells and the different types of cone cells each have an optimal wavelength. However, there is considerable overlap in the wavelengths of light detected.

Exercise 2

A: Blind spot test – The optic disk, the sight where ganglion cell axons exit the eye, does not contain photoreceptors. We do not perceive the blind spot because the brain interpolates information to fill in the gaps. You can locate the blind spot by moving the image below toward your head.

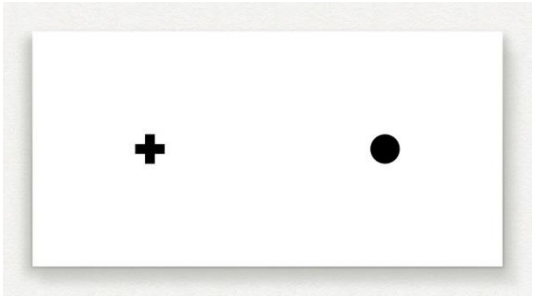


Figure S5. Standard blind spot test

Directions: With your right eye (left closed), stare at the cross, and move the paper towards your eyes until the circle disappears. Measure the distance from the image to your eye. You can repeat this with your left eye, staring at the circle. The cross will disappear at a distance related to the physical separation of your fovea and your optic disk. You can estimate this distance (d', in mm) of your retinal structures by recording the distance when the image disappears (x), and measuring the distance between the cross and circle in the image (d). This is actually a 'reduced eye' model, which involves some approximations.

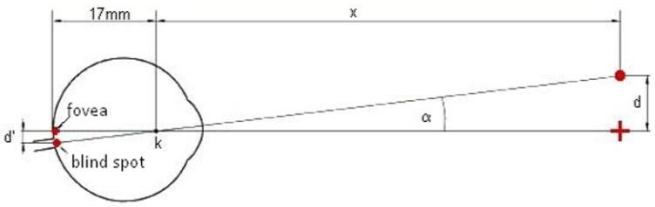


Figure S6. Diagram to show measurement of blind spot to fovea.

d' = 17 (d/x)

d' = fovea to optic disk (mm)

d = cross to circle in image (mm)

x = eye to image (mm)

What is the distance between the fovea and optic disk in your left eye? _____

Right eye? _____ Average distance? _____

Are your eyes exactly the same? _____

Hypothesize why or why not:

_____

_____

_____

B: Color-blindness test – Color-blindness can be tested with appropriate Standard Pseudoisochromatic plates (e.g. Ichikawa et al., ISBN 0-89640-030-1)

Directions: Using the plates are you able to detect the image present for the presented colors?

Plate #: _____ Image that you see: _____

Plate #: _____ Image that you see: _____

Plate #: _____ Image that you see: _____

Plate #: _____ Image that you see: _____

Plate #: _____ Image that you see: _____

Plate #: _____ Image that you see: _____

If you exhibit some degree of colorblindness, what type? What may be happening with your cone cells within your retinas? Read back a little to see if you can figure it out.

_____

_____

_____

C: Visual acuity test – Visual acuity refers to the sharpness or clarity of vision, and is an indication of the focusing capacities of your eyes, especially the lens and cornea.

Directions: Use the standard eye chart (Snellen chart), at the appropriate distances (usually 20 feet), to measure your acuity. Comparisons on the chart will refer to this standard measure. To use the chart, find the red tape on the floor. Stand here and cover one eye without squinting. Have your partner stand near the chart and tell you which line to read starting from the top. When you can no longer read a line accurately with one eye, then this is the acuity for that eye. Write it here: _____. Now do the other eye the same way. Write your acuity here: _____. Do you have the same acuity in both eyes? Why or why not?

_____

_____

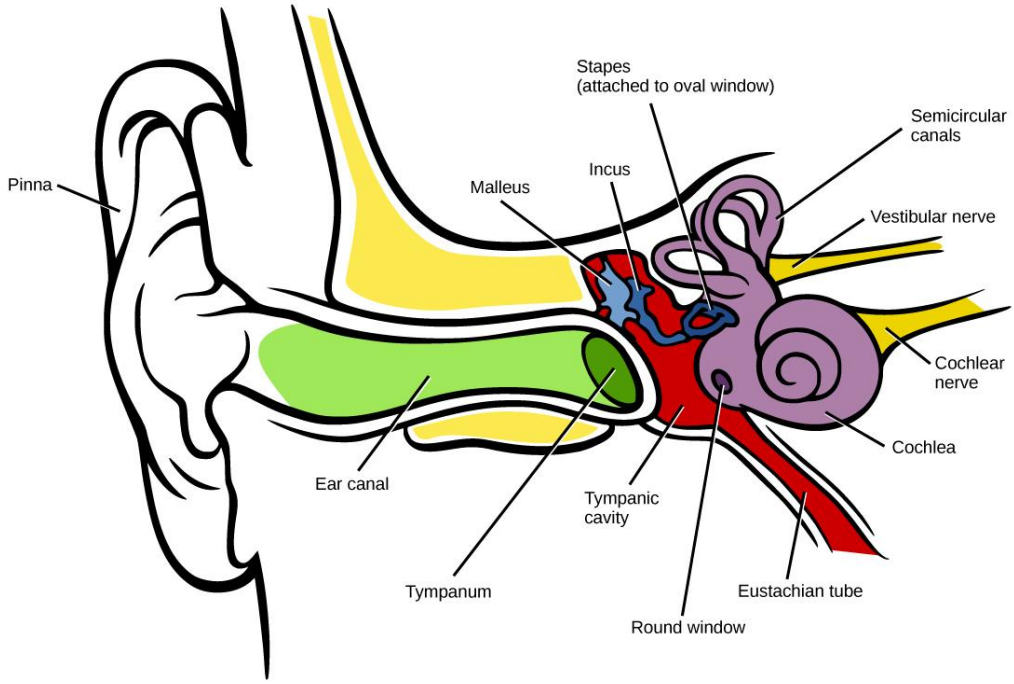
_____

Note on corrective lenses: If you have contacts, keep them in, but if you wear glasses you may want to try the test with and without them to see just how much your corrective lenses improve your vision!

HEARING – Hearing involves the signal transduction of mechanical waves into neural signals in the cochlea, within the inner ear.

Anatomy: The auditory receptors for the ear (shown below) include the:

- *outer ear* - from pinna (or *auricle*) to tympanum (or tympaninc membrane, or *ear drum*)
- *middle ear* - contains 3 ossicles, anchored between tympanum & oval window. The *Eustachian* (or auditory) tube connects the middle ear cavity to the pharynx (it is an evolutionary descendant of pharyngeal pouches).
- *Inner ear* – Cochlea, which contains the *hair cells (receptors)* within the Organ of Corti.



BALANCE & MOVEMENT (The Vestibular System)

Anatomy: Within the inner ear, the 3 semicircular canals are arranged at right angles to each other, and they contain hair cells and fluid similar to the cochlea. In addition, two separate clusters of hair cells – the saccule and utricle – are oriented to detect vertical and horizontal movements. Each of the hair cell clusters has a small collection of dense connective tissue attached to the hair cell membrane extensions (*stereocilia)* to add mass to the system.

Physiology: Movements of the head cause dislocations of the fluid in the chambers around the hair cells. Movements generate electrical signals in hair cells, which signal sensory neurons with released chemical neurotransmitters. Patterns of signals are integrated in the cerebellum and parietal cortex.

Specimens & Models for examination: Models of the ear and cochlea.

Exercise 3

A: Identifying structures - Using the model of the ear find the following structures: ear canal, tympanum (tympanic membrane), ossicles (malleus, incus, stapes in order moving inward into the ear), cochlea, semicircular canals.

B: Sound localization – Using a tuning fork, have a subject sit with their eyes closed. Strike the fork so it makes a sound and move it to front, back, side and top of head at a constant distance, holding it to allow the subject to point out the location. Note the accuracy at each position of their pointing, and determine the most and least accurate positions for localization. Can you explain any differences? List the positions for localization from most to least accurate:

_____

_____

_____

C: **Romberg testing involves maintaining balance.**

A. Have the subject stand with their back to the white board. The board should be marked at approximately shoulder height with centimeter units covering ~1 meter.
B. Note the shoulder positions of the subject.
C. Have the subject stand and stare straight ahead for 2 minutes, and note the range of movement.
D. Repeat with eyes closed.
E. Repeat while standing with your right or left side closest to the board, and note front-to-back swaying, First with eyes open and then with eyes closed.

Data:	Eyes open	Eyes closed
Side to side (cm)		
Front to back (cm)		

Describe any differences in relation to the sensory input required to maintain balance.

_____

_____

_____

SMELL

Anatomy: Sensory (olfactory) neurons are present at the top of the nasal cavity, extending their axons into the cranium. Olfactory signals are the only sensory system to send signals directly to the limbic system, which is integral to memory and emotional functions. In humans, from 100-200 different functional receptor proteins have been identified (there are over 1000 in rodents).

Physiology: Specific molecules (*odorants*) bind to receptor proteins and activate neural electrical signals (action potentials). Patterns of olfactory neuron activity can code for complex odors, integrated within the olfactory bulb and temporal cortex. Olfactory neurons will undergo adaptation and decrease signals to the brain with constant exposure to a stimulus.

Exercise 4

Humans consistently recognize certain odorants (e.g. spearmint, orange, anise). Specific oils for these are available, and can be prepared as serial dilutions. Students can then test for sensitivities for each by starting with a series at the low end of the concentrations. Odorants can be detected by some sensitive individuals at concentrations below the micromolar range. Below list the micromolar concentrations of mint and circle the one where you can begin to smell the mint.

Concentration 1: _____ Concentration 2: _____ Concentration 3: _____

Concentration 4: _____ Concentration 5: _____

TASTE – Taste involves stimulation of receptor proteins on gustatory cells within taste buds. The perceived sensations correspond to common chemicals: Salty (Na^+), Sweet (disaccharides, e.g. sucrose), Bitter (various, common test is Ca^{2+}), sour (H^+), and umami (glutamate). Also, taste is often integrated as a perception with olfactory sensory input.

Anatomy & Physiology: Taste buds are arranged along the tongue epithelium. Sensory epithelial cells release neurotransmitter signal molecules to sensory neurons of cranial nerves. Information is integrated along the brain stem and in the temporal cortex.

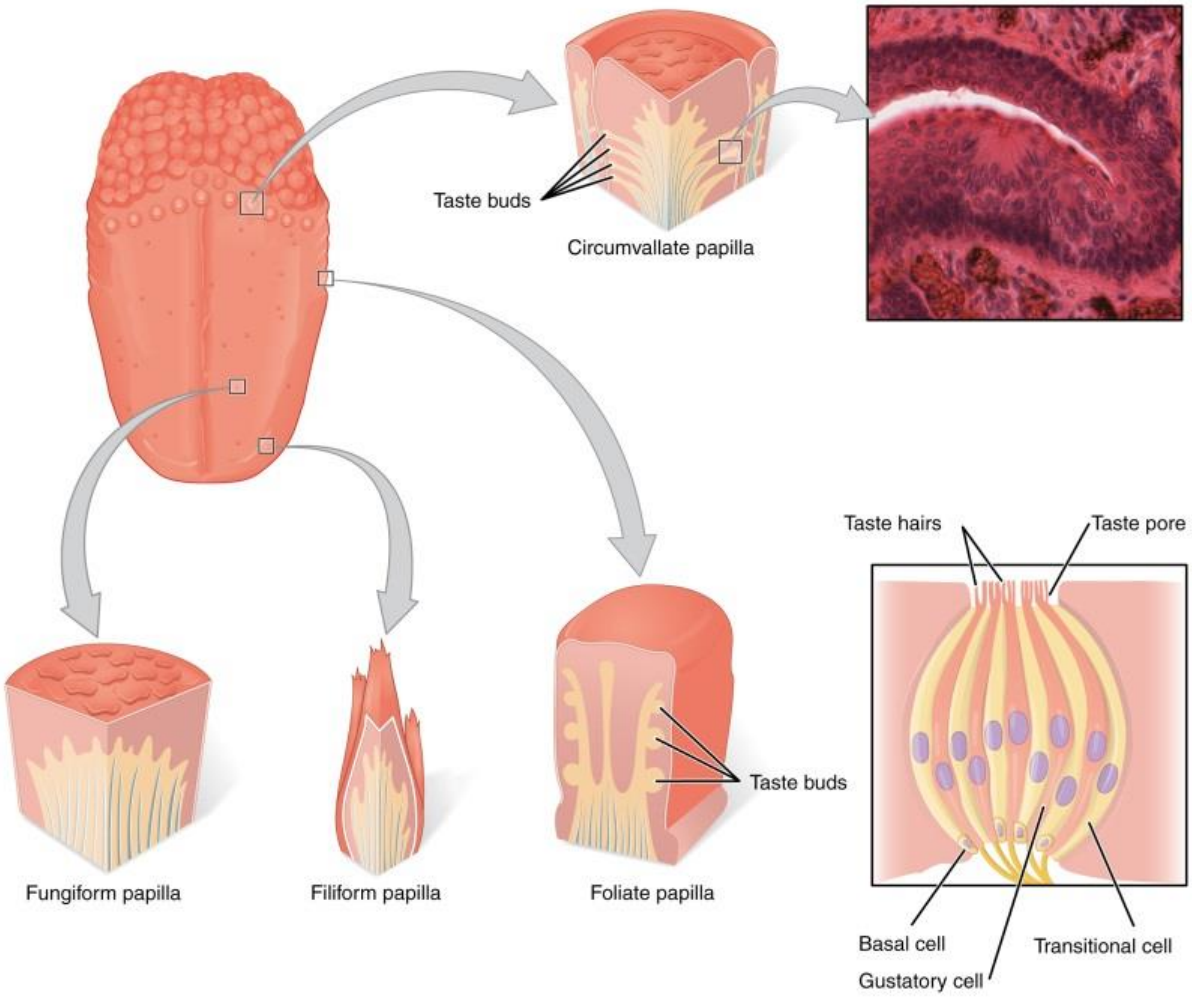


Figure S9.The tongue is covered with small bumps, called papillae, which contain taste buds that are sensitive to chemicals in ingested food or drink. Different types of papillae are found in different regions of the tongue. The taste buds contain specialized gustatory receptor cells that respond to chemical stimuli dissolved in the saliva. These receptor cells activate sensory neurons that are part of the facial and glossopharyngeal nerves. LM × 1600. (Micrograph provided by the Regents of University of Michigan Medical School © 2012)

Specimens & Models for examination: Histology slide of tongue/taste buds

Physiology: Taste can be identified using solutions of chemicals known to stimulate distinct receptor proteins. Solutions can be prepared from common ingredients to test for sensitivity.

Exercise 5

A: Histology Exploration – Use a microscope to explore the cellular aspect of a taste bud. Find an individual taste bud and draw it in the space provided. Can you label any structures if you use the above diagram as a guide?

B: PTC Test - One bitter taste receptor protein is encoded by the PTC gene, or TAS2R38 (discovered in 2003). There are at least 30 different genes coding for bitter taste receptors. Phenylthiocarbamide (PTC), also known as phenylthiourea (PTU), is only detected by ~70% of the population on average. Tasting PTC is correlated with the dominant genotype.

A. PTC tasting test kits provide material to survey the class. Testing is a simple positive response for bitter taste, while non-tasters will report no taste.
    a. After placing the strip on your tongue do you taste anything? Yes or no? _____. If you answered YES!, then you have the dominant genotype for the PTC gene!

C: Taste Threshold Test - Similar to the olfaction tests, serial dilutions of basic chemicals can be used to test for variable sensitivity in subjects. Sucrose and NaCl are common tests for sweet and salty. Serial solutions can be applied with cotton swabs to the subjects tongue to test for sensitivity. Similar to the smell test, list the concentrations of the two substances and circle the one where you can begin to taste the substance.

Salt

Concentration 1: _____ Concentration 2: _____ Concentration 3: _____

Concentration 4: _____ Concentration 5: _____

Sucrose

Concentration 1: _____ Concentration 2: _____ Concentration 3: _____

Concentration 4: _____ Concentration 5: _____

Do you and your lab partner vary in your sensitivities? If you differ, then provide a possible explanation as to why:

_____

_____

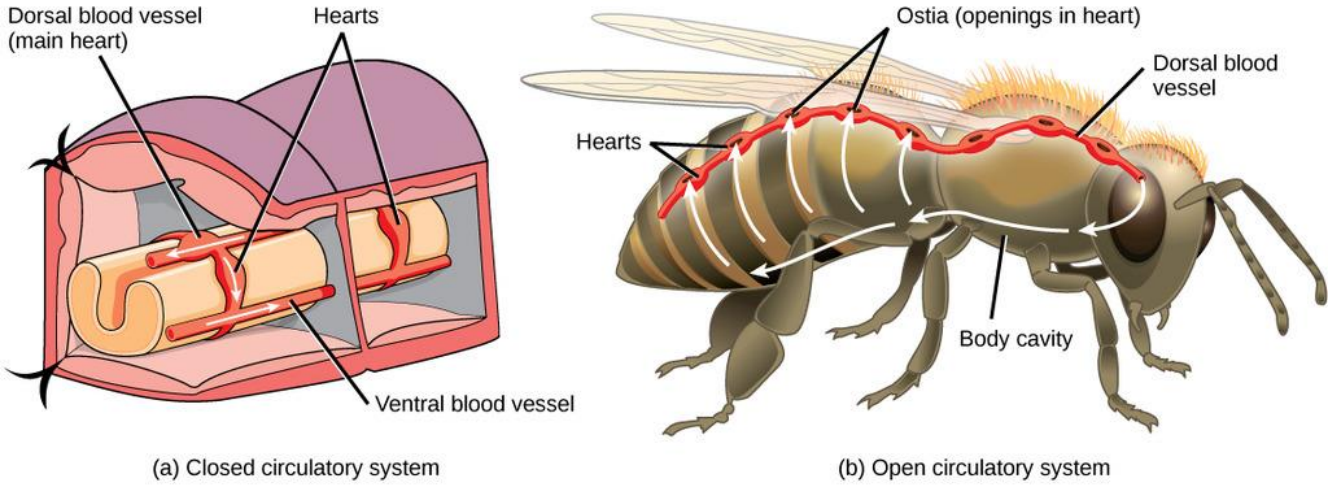
_____

A. HISTORICAL NOTE: The 'standard' map of taste buds common in many lab manuals has been disproved by subsequent research (*J. Cell Biology, 2010 vol. 190 no. 3 285-296 doi: 10.1083/jcb.201003144*). There is more variability among individuals than accounted for by the original 1942 map (not shown, intentionally).
B. Individuals can map their tongues for taste buds, once sensitivity thresholds have been determined.

Circulatory Systems Lab

Most animals are complex multicellular organisms that require a mechanism for transporting nutrients throughout their bodies and removing waste products. The circulatory system has evolved over time from simple diffusion through cells in the early evolution of animals to a complex network of blood vessels that reach all parts of the human body. This extensive network supplies the cells, tissues, and organs with oxygen and nutrients, and removes carbon dioxide and waste, which are byproducts of respiration and other cellular activities.

Circulatory systems differ significantly throughout the Animal Kingdom. In all vertebrate organisms, as well as some invertebrates, this is a closed-loop system, in which the blood is not free in a cavity. In a **closed circulatory system**, blood is contained inside blood vessels and circulates **unidirectionally** from the heart around the systemic circulatory route, then returns to the heart again. As opposed to a closed system, arthropods—including insects, crustaceans, and most mollusks—have an open circulatory system. In an **open circulatory system**, the blood is not enclosed in the blood vessels but is pumped into a cavity called a **hemocoel** and is called **hemolymph** because the blood mixes with the **interstitial fluid**.

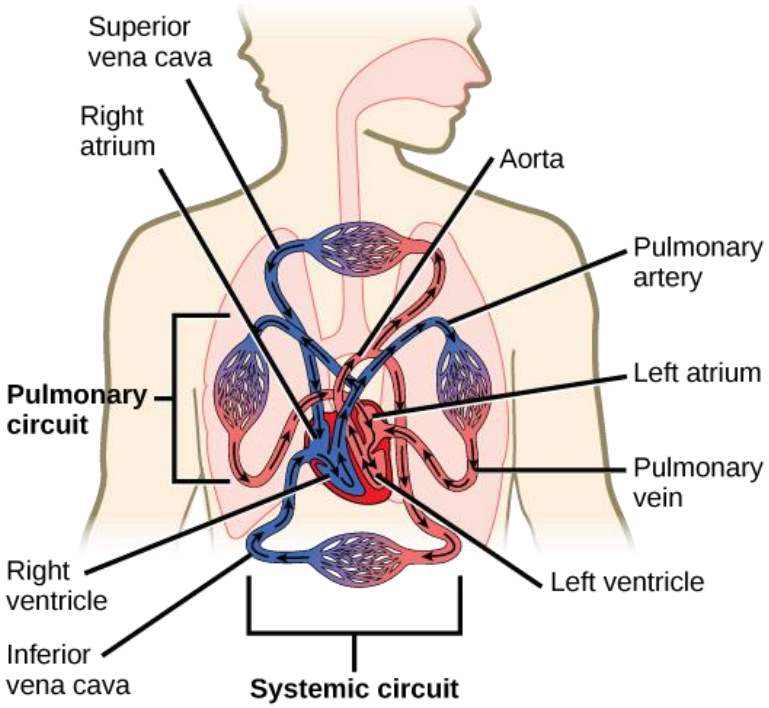


(a) Closed circulatory system

(b) Open circulatory system

Mammalian Heart and Blood Vessels

The mammalian circulatory system is divided into three circuits: the systemic circuit, the pulmonary circuit, and the coronary circuit. Blood is pumped from veins of the systemic circuit into the right atrium of the heart, then into the right ventricle. Blood then enters the pulmonary circuit, and is oxygenated by the lungs. From the pulmonary circuit, blood re-enters the heart through the left atrium. From the left ventricle, blood re-enters the systemic circuit through the aorta and is distributed to the rest of the body.



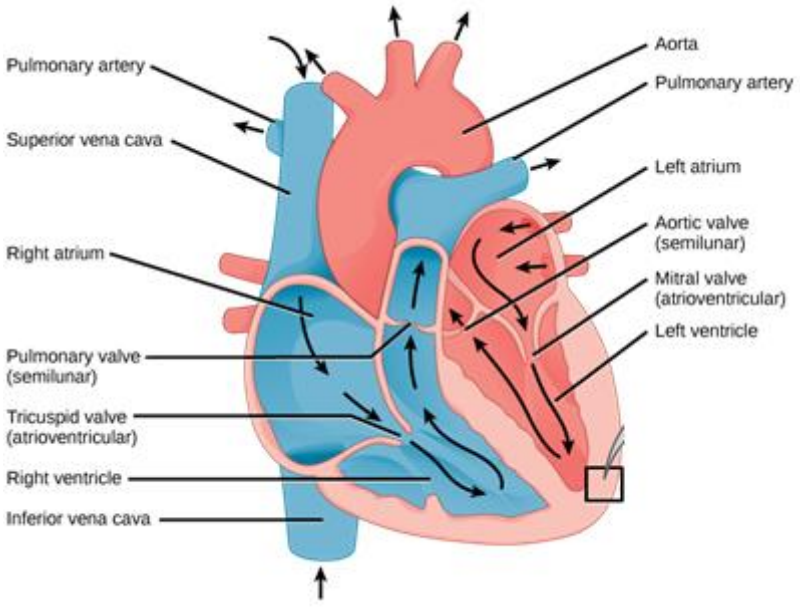
Exercise 1: Structure of the Heart

Materials:

Human Heart Model

Procedure:

Using the model of the human heart and the chart provided, locate the different structures of the heart. Write the flow of the blood and the function of each structure in the space provided:



Right atrium: _____

Tricuspid valve: _____

Right ventricle: _____

Left atrium: _____

Bicuspid/Mitral valve: _____

Left ventricle: _____

Exercise 2: Blood type and RH factor.

Blood is important for regulation of the body's systems and homeostasis. Blood helps maintain homeostasis by stabilizing pH, temperature, osmotic pressure, and by eliminating excess heat. Blood supports growth by distributing nutrients and hormones, and by removing waste. Blood plays a protective role by transporting clotting factors and platelets to prevent blood loss and transporting the disease-fighting agents or white blood cells to sites of infection.

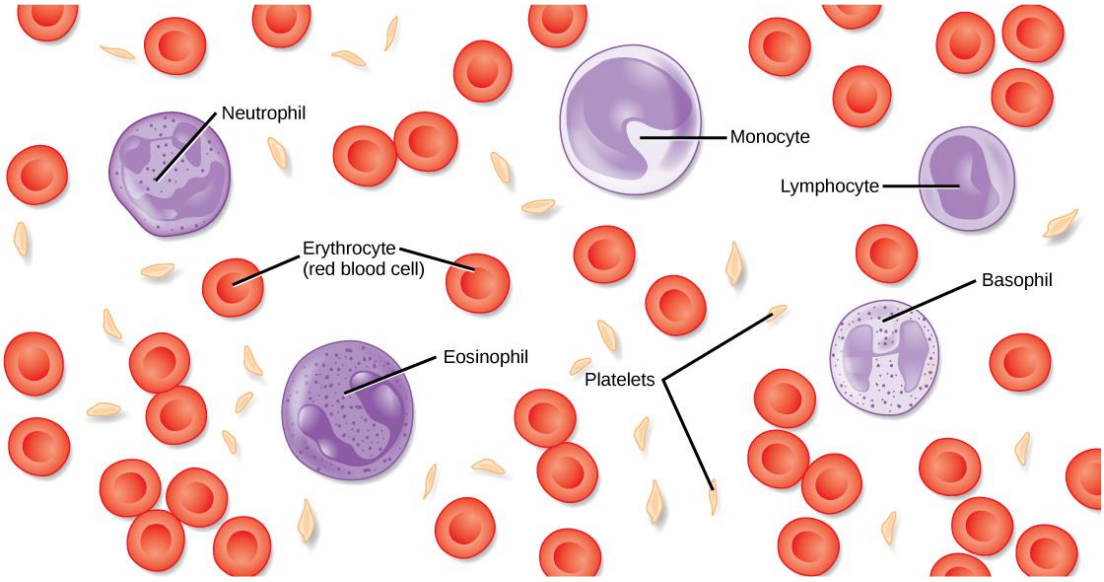


Figure. The cells and cellular components of human blood are shown. Red blood cells deliver oxygen to the cells and remove carbon dioxide. White blood cells—including neutrophils, monocytes, lymphocytes, eosinophils, and basophils—are involved in the immune response. Platelets form clots that prevent blood loss after injury.

Microscope work:

Use one of the light microscopes to explore a slide of a human blood smear. The leukocytes (white blood cells) are stained purple (nuclear staining) and far less common than the erythrocytes (red blood cells). Count the number of leukocytes in your view at the medium magnification. How many are there? _____

Erythrocytes outnumber leukocytes 600:1. Therefore, estimate the number of erythrocytes in your view: _____.

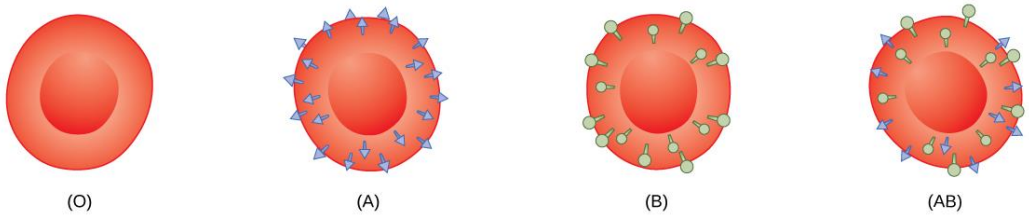
Why do you think the erythrocytes outnumber the leukocytes?

_____

_____

_____

Red blood cells are coated in antigens made of glycolipids and glycoproteins. The composition of these molecules is determined by genetics, which have evolved over time. The two most well-known blood groups are the ABO and Rh systems. The surface antigens in the ABO blood group are glycolipids, called antigen A and antigen B. People with blood type A have antigen A, those with blood type B have antigen B, those with blood type AB have both antigens, and people with blood type O have neither antigen. Antibodies called agglutinogens are found in the blood plasma and react with the A or B antigens, if the two are mixed. When type A and type B blood are combined, agglutination (clumping) of the blood occurs because of antibodies in the plasma that bind with the opposing antigen; this causes clots that coagulate in the kidney causing kidney failure. Type O blood has neither A or B antigens, and therefore, type O blood can be given to all blood types. Type O negative blood is the universal donor. Type AB positive blood is the universal acceptor because it has both A and B antigen.



The Rh blood group was first discovered in Rhesus monkeys. Most people have the Rh antigen (Rh+) and do not have anti-Rh antibodies in their blood. The few people who do not have the Rh antigen and are Rh– can develop anti-Rh antibodies if exposed to Rh+ blood. This can happen after a blood transfusion or after an Rh– woman has an Rh+ baby. The first exposure does not usually cause a reaction; however, at the second exposure, enough antibodies have built up in the blood to produce a reaction that causes agglutination and breakdown of red blood cells. An injection can prevent this reaction.

Based on the information above, fill out the following chart regarding each blood type:

Blood Type	Possible Genotype	Antigens on RBCs	Antibodies in plasma
A			
B			
AB			
O			

Materials:

Blood typing kit (1/each group)
4 unknown simulated blood dropper bottles
Anti –A, Anti-B, Anti-D (RH) sera dropper bottles
Toothpicks

Procedure:

1. Place one drop of simulated blood in the middle of each depression in the blood typing tray.
2. Add one drop of Anti-A serum next to the A blood cells, one drop of Anti-B serum next to the B blood cells, and one drop of Anti-D serum next to box D blood cells.
3. Mix the simulated blood with the Anti-serum using the toothpick for about two minutes
4. Depend on the type of the antigen in the blood, the blood mixture will clump or the mixture will not change.
5. Recode your observation on the following table.

Simulated Blood	Blood Type	RH (+/-)
Unknown sample 1		
Unknown sample 2		
Unknown sample 3		
Unknown sample 4		

Exercise 3: Blood pressure and pulse rate

Blood pressure (BP) is the pressure exerted by blood on the walls of a blood vessel that helps to push blood through the body. Blood moving through the blood vessels exerts pressure against the vessel walls. This blood pressure is highest in the aorta. It decreases as the blood moves through the arterioles, capillaries, venules, and veins. Many factors can affect blood pressure, such as hormones, stress, exercise, eating, sitting, and standing. Blood flow through the body is regulated by the size of blood vessels, by the action of smooth muscle, by one-way valves, and by the fluid pressure of the blood itself.

There are two components to blood pressure:

1. **Systolic blood pressure**: measures the amount of pressure that blood exerts on vessels while the heart is beating (contracting). The pressure in the vessel is highest at this time. The optimal systolic blood pressure is 120 mmHg.
2. **Diastolic blood pressure**: measures the pressure in the vessels between heartbeats (resting). The pressure is at its lowest point. The optimal diastolic blood pressure is 80 mmHg.

When we measure blood pressure, we are actually measuring the systolic pressure and the diastolic pressure separately. This is why you always see blood pressure reported as two numbers, one

"over" the other. For example in a blood pressure reading of 130/85, this means that the systolic pressure is 130 and the diastolic pressure is 85.

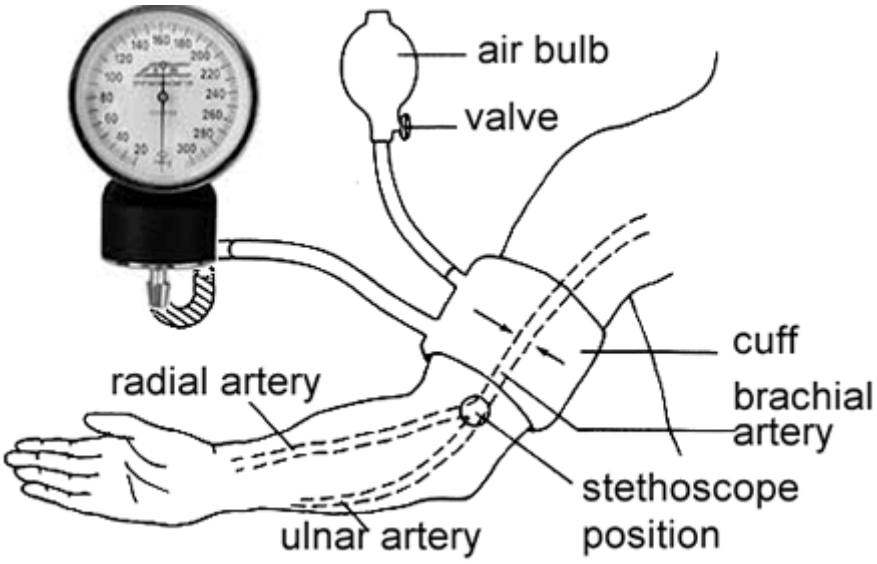
Measuring Blood Pressure and pulse rate

Materials:

- Sphygmomanometer (pronounced sfig-mo-muh-NAM-eh-ter) consists of an inflatable cuff, an inflation pump, a gauge to register pressure, and a controlled exhaust valve.
- Stethoscope: to listen to the different sounds that occur

Procedure

You will work in pairs to measure the effect of exercise on pulse rate and blood pressure.



1. Have your partner sit down and relax for 2 min. Place your index and middle fingers in the groove on the inside of his/her wrist. Just slide your fingers across the tendons until they slip into soft tissue.
2. Wait until you clearly feel beats coming with a regular rhythm.
3. Count the number of pulses in 15 sec.
4. Multiply the number of pulses by 4 to get the number of beats per minute.
5. Record this number in the table below.
6. Attach the inflatable cuff around your partner's arm above the elbow as shown in the figure above. Tuck the flap of the bag under the fold.
7. Place the stethoscope over the brachial artery (underneath the cuff as shown in the figure above.
8. You begin by inflating the cuff to about 200 mm. The blood flow below stops and you will hear no sound in the brachial artery when you listen with the stethoscope.
9. If the pressure has gone below 200mm Hg, inflate the cuff again.

10. Slowly begin releasing the pressure in the cuff. The blood begins to flow through the artery causing vibrations and turbulence that are audible through the stethoscope
11. The first tapping sound you hear indicates that blood has entered the artery. Record this reading as the **systolic pressure**.
12. You continue to deflate the cuff until you stop hearing any sound. The last tapping sound you hear indicates the **diastolic pressure**.
13. Now go outside and exercise for exactly 5 minutes. You can do jumping jacks, run up and down the stairs or do push-ups.
14. Immediately after sitting down measure the pulse rate/min (Steps 1–3) and the blood pressure (steps 4–10) and record your data.
15. Measure the beat rate and the blood pressure of your partner. Record the results in following table.

Measuring the effect of exercise on pulse rate and the blood pressure

		Student 1			Student 2	
	Beats/min	Systolic	Diastolic	Beats/min	Systolic	Diastolic
Resting						
After exercise						

Questions:

1. How does exercise affect pulse rate and the blood pressure?

2. Explain what happens to the circulatory system during exercise. Include the major organs involved in your explanation.

3. Why does increased physical activity increase heart rate?

4. Why is heart rate lower in an individual who does aerobic exercise regularly?

5. How and why does heart rate change with body position?

Human Respiration Lab

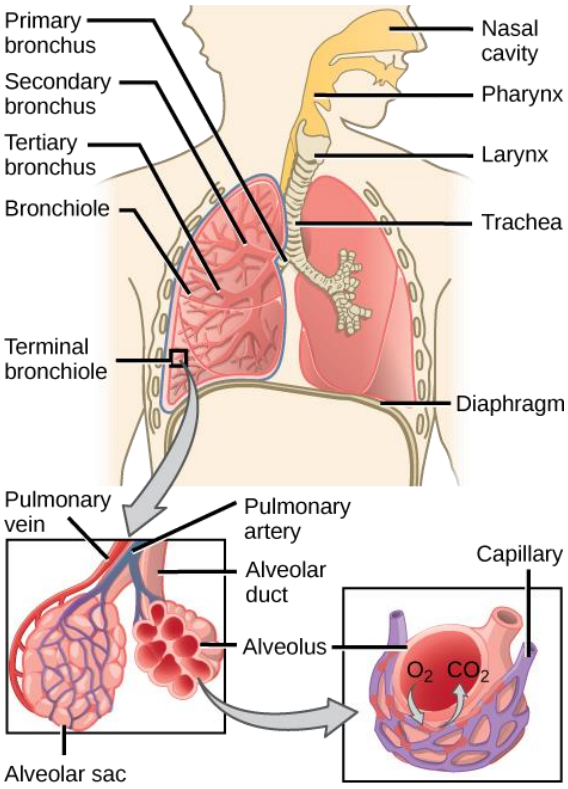
Objectives:

- To understand the microscopic and gross anatomy of the respiratory tract
- To observe and measure the mechanics of breathing, respiratory volumes, and the control of breathing
- To observe and understand the role of buffers in maintaining pH balance in the body

Introduction:

Breathing is an involuntary event. Humans, when they are not exerting themselves, breathe approximately 15 times per minute on average. The primary function of the respiratory system is to deliver oxygen to the cells of the body's tissues and remove carbon dioxide, a cell waste product. Oxygen (O_2) diffuses into the cells where it is used for metabolic reactions that produce ATP, a high-energy compound. At the same time, these reactions release carbon dioxide (CO_2) as a byproduct. CO_2 is toxic in high amounts and must be eliminated. CO_2 diffuses out of the cells, enters the bloodstream, travels back to the lungs, and is expired out of the body during exhalation.

The main structures of the human respiratory system are the nasal cavity, the trachea, and lungs.

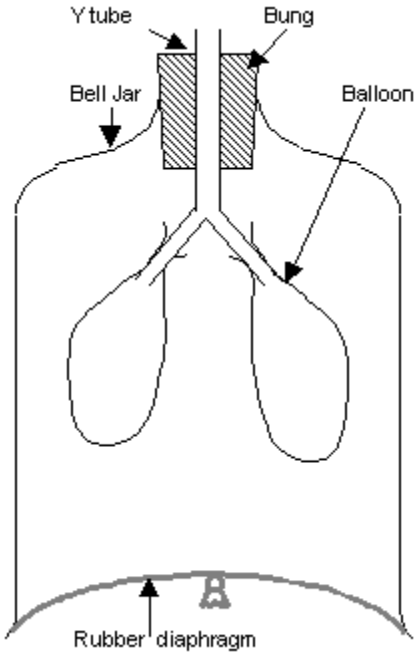


Human respiratory system

Exercise 1: Mechanism of breathing (balloon-and-bell jar model)

Materials

Balloon-and-bell jar

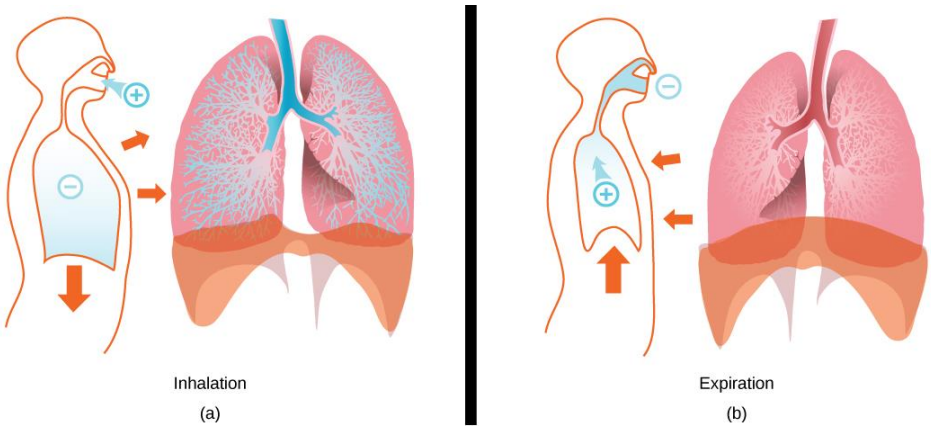


Procedure

- Pull down the rubber sheet. What happens to the balloons? _____. This represents the downward movement of the human _____, which causes the chest cavity to become _____ (larger/smaller). This, in turn, causes the human _____ to expand and fill with air.
- Release the rubber sheet. What happens to the balloons? _____. This represents relaxed _____ (inhaling/exhaling), when the chest cavity becomes smaller and the lungs deflate. Note that this is a passive process.
- What organs do the balloons represent? _____

Exercise 2: Breathing Measurements

During inhalation, volume increases as a result of contraction of the diaphragm, and pressure decreases (according to Boyle's Law). This decrease of pressure in the thoracic cavity relative to the environment makes the cavity less than the atmosphere. Because of this drop in pressure, air rushes into the respiratory passages. To increase the volume of the lungs, the chest wall expands. The chest wall expands out and away from the lungs. The lungs are elastic; therefore, when air fills the lungs, the **elastic recoil** within the tissues of the lung exerts pressure back toward the interior of the lungs. These outward and inward forces compete to inflate and deflate the lung with every breath. Upon exhalation, the lungs recoil to force the air out of the lungs, and the intercostal muscles relax, returning the chest wall back to its original positi on. The diaphragm also relaxes and moves higher into the thoracic cavity. This increases the pressure within the thoracic cavity relative to the environment, and air rushes out of the lungs. The movement of air out of the lungs is a passive event. No muscles are contracting to expel the air.



The lungs, chest wall, and diaphragm are all involved in respiration, both (a) inhalation and (b) expiration.

- Put one hand on your chest and take three deep inspirations followed by three forced expirations. Describe your observation during each inspiration and expiration.

_____

_____

_____

- Repeat Step 1 with your hands on your abdomen. Now try to breathe in and out without any movement of your chest. Describe your observation during each inspiration and expiration.

_____

_____

_____

Exercise 3: Measurements of breathing in resting and active modes

The number of breaths per minute is the **respiratory rate**. On average, under non-exertion conditions, the human respiratory rate is 12–15 breaths/minute. The respiratory rate contributes to the **alveolar ventilation**, or how much air moves in and out of the alveoli. Alveolar ventilation prevents carbon dioxide buildup in the alveoli. When a person consciously holds her/his breath for a long period of time, the CO_2 level rises which causes the pH of the blood to decrease. This stimulates the respiratory center and reflex breathing occurs. As CO_2 is removed, the reaction proceeds to the left, thus removing hydrogen ions and forming more CO_2 for liberation from the body.

Materials

Per group (2): Timer

Procedure

The purpose of the following exercises is to investigate some of the factors that affect the rate and depth of breathing.

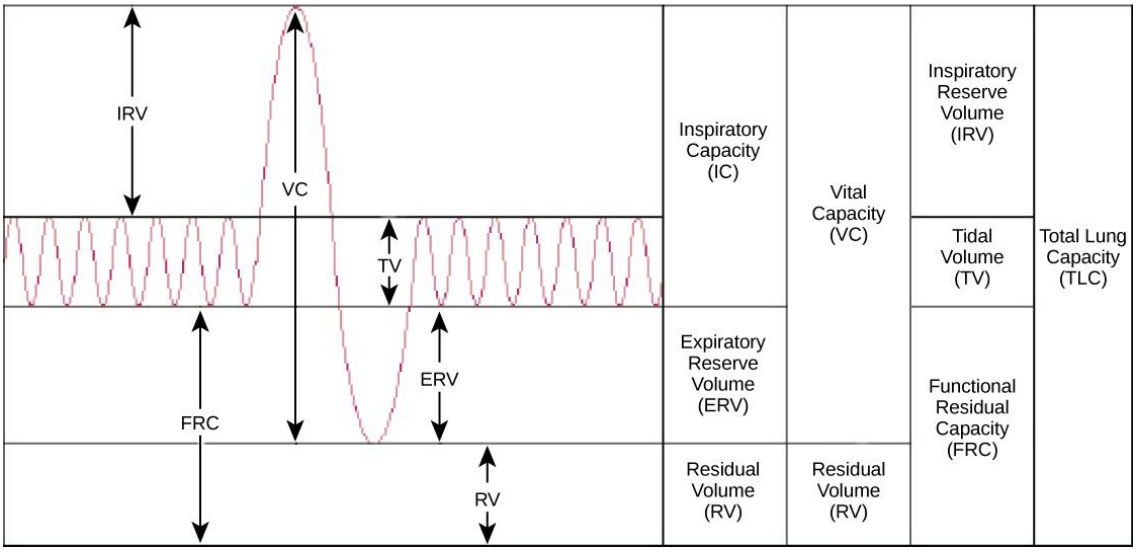
CAUTION
Do not do the following activities if you have any medical problems with your lung or heart. All subjects should stop immediately if they feel faint.

1. Time how long you can hold your breath after a quiet inspiration. _____sec.
2. Time how long you can hold your breath after a deep inspiration (inhale as deeply as possible, then hold your breath). _____sec
3. Time how long you can hold your breath following a quiet expiration. _____ sec
4. Breathe deeply and forcefully at a rate of about 1 breath/3 seconds, 20 times (hyperventilate). Time how long you can hold your breath. _____sec
5. Run down the hall and back again or go down to the first floor and come back up quickly. Then immediately hold your breath for as long as possible. Record the time. _____sec

During which procedure could you hold your breath the longest? _____

Exercise 4: Measuring Lung Volumes and Capacities

Human lung size is determined by genetics, gender, and height. At maximal capacity, an average lung can hold almost six liters of air, but lungs do not usually operate at maximal capacity. Air in the lungs is measured in terms of **volumes** and **capacities**. Volume measures the amount of air for one function (such as inhalation or exhalation). Capacity is any two or more volumes (for example, how much can be inhaled from the end of a maximal exhalation).



Lung Volumes and Capacities (Avg Adult Male)

Volume/Capacity	Definition	Volume (liters)	Equations
Tidal volume (TV)	Amount of air inhaled during a normal breath	0.5	-
Expiratory reserve volume (ERV)	Amount of air that can be exhaled after a normal exhalation	1.2	-
Inspiratory reserve volume (IRV)	Amount of air that can be further inhaled after a normal inhalation	3.1	-
Residual volume (RV)	Air left in the lungs after a forced exhalation	1.2	-
Vital capacity (VC)	Maximum amount of air that can be moved in or out of the lungs in a single respiratory cycle	4.8	ERV+TV+IRV
Inspiratory capacity (IC)	Volume of air that can be inhaled in addition to a normal exhalation	3.6	TV+IRV
Functional residual capacity (FRC)	Volume of air remaining after a normal exhalation	2.4	ERV+RV
Total lung capacity (TLC)	Total volume of air in the lungs after a maximal inspiration	6.0	RV+ERV+TV+IRV
Forced expiratory volume (FEV1)	How much air can be forced out of the lungs over a specific time period, usually one second	~4.1 to 5.5	-

Materials

Per group (2): nose clip (optional); simple spirometer; disposable mouth piece

Procedure

1. **Measure Tidal Volume (TV)**

- Place a disposable mouthpiece over the stem of the spirometer.
- We have several different styles of spirometer. If the spirometer has tick marks all the way around, set the dial to zero. If the spirometer has a gap between 0 and 1000, set the dial to 1000.
- Hold your nose so that all of the air expired from your lungs enter the spirometer. Sit and breathe quietly for a moment. Expire into the spirometer. ***Do not force any air out of your lungs.*** Count the number of tick marks the dial moved and multiply by 100. Record this value below. Repeat two more times.

Trial 1_____ ml Trial 2 _____ ml Trial 3 _____ ml

- Average your measurements:

TV: _____ml

2. **The minute respiratory volume (MRV) represents the volume of air moving in and out of your lungs in 1 minute during normal, quiet respiration.**

- Determine your normal respiratory rate (RR) for 1 min by counting the number of breaths (remember, a breath is inhale + exhale) : RR: _____ (normal RR = ~12 breaths per minute)
- To determine your MRV, multiply your RR x TV.

MRV = _____ml per minute

3. **Measure Expiratory Reserve Volume (ERV)**

- After three normal breaths, **ending** in expiration, hold your nose and **forcefully** expel all of the air left in your lungs into the spirometer. Count the number of tick marks the dial moved and multiply by 100. Record this value below. Repeat two more times.

Trial 1_____ ml Trial 2 _____ ml Trial 3 _____ ml

ERV: _____ml

4. **Measure Vital Capacity (VC)**

- After three deep breaths, take one final deep inspiration. Then hold your nose and exhale as much air as possible into the spirometer. **Note:** A slow, even, forced expiration works best. Repeat two more times. Count the number of tick marks the dial moved and multiply by 100. Record this value below.

Trial 1_____ ml Trial 2 _____ ml Trial 3 _____ ml

VC: _____ml

5. **Inspiratory Capacity (IC) can be calculated using the following formula**

IC = VC − ERV = _____ml

6. **Inspiratory Reserve Volume (IRV) can be calculated using the following formula**

IRV = IC − TV = _____ml

7. **Residual Volume (RV) can be calculated using the following formula**

RV = VC x Age Factor = _____ ml

Age Factors: age 16−34 = 0.25; age 35−49 = 0.31; age 50−69 = 0.45

8. **Total lung capacity (TLC) can be calculated using the following formula**

TLC = TV + IRV + ERV + RV = _____ ml

	Male		Female	
	Your Data	Average	Your Data	Average
Tidal volume (TV)		500		375
Inspiratory reserve volume (IRV)		3000		2250
Expiratory reserve volume (ERV)		1000		750
Residual volume (RV)		1200		900
Total Lung Capacity (VC + RV)		5700		4275

Lab 6 – 10: Fetal Pig Dissection

Dissection is a powerful tool that provides us profound understanding of our own anatomy and physiology as living, breathing creatures and also helps us to develop a stronger understanding of evolutionary relationships between taxonomic groups. What sort of dissection experiences do you already have? As you begin this sequence of dissections, please keep in mind several things. Dissection should be done thoughtfully and respectfully. It is important to take your time carefully and to think about what you are doing. This care will help you to preserve structures for the next several weeks and will make review easier. Also, merely identifying structures is insufficient to develop fully an appreciation for how these structures work. You must think about the function of these structures. How do they work?! Also, please do these dissections respectfully. These organisms were euthanized so that we might have a wonderful opportunity to learn something greater about the living world. Please keep that in mind as you are working.

Objectives:

1. Develop dissecting skills.
2. Learn anatomy and physiology of key vertebrate and mammalian systems.

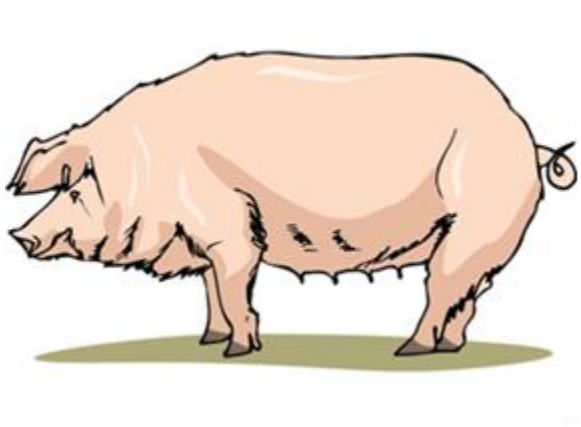
External Anatomy

Determine the sex of your pig:

1. Before you start dissecting, examine the outside of the pig and determine its sex. Look for these features:
   - **Males:** The urogenital opening is located near the umbilicus; the penis is hidden inside. The scrotal sac may be visible as a swelling just ventral to the anus, depending on the age of the fetus. The testes are still deep inside the body cavity; they don't descend into the scrotal sac until later.
   - **Females:** Look for the urogenital papilla (a nipple-looking structure), located just below the anus.

Also, both males and females have nipples, just as in humans.

What sex is your pig? _____



***label the sides on the pig picture above**

2. Make sure you are familiar with anatomical terms of reference: **anterior (front), posterior (back), dorsal (above), ventral (below)**. In addition, you'll need to know the following terms:

Medial: toward the midline or middle of the body
Lateral: toward the outside of the body
Proximal: close to a point of reference
Distal: farther from a point of reference

3. Open the pig's mouth and locate the **hard** and **soft palate** on the roof of the mouth. Can you feel your own hard and soft palates with your tongue?

Note the **taste buds** (also known as **sensory papillae**) on the side of the **tongue**. Locate the esophagus at the back of the mouth. Feel the edge of the mouth for teeth. Does the fetal pig have teeth? _____

To access the following structures, you will have to cut down either side of the jaw and pry the jaw down. This can be difficult and requires some force. You, essentially, must break the jaw, and it will make a

cracking sound. Once you do this locate the **epiglottis**, a cone-shaped structure at the back of the mouth, a flap of skin helps to cover the trachea when a pig swallows. The **pharynx** (throat) is the cavity in the back of the mouth – it is the junction for food (esophagus) and air (trachea).

5. Observe the toes of the pig. How many toes are on the feet? _____
Do they have an odd or even number of toes? _____

****Make sure you know the locations of all the bold words on this handout****

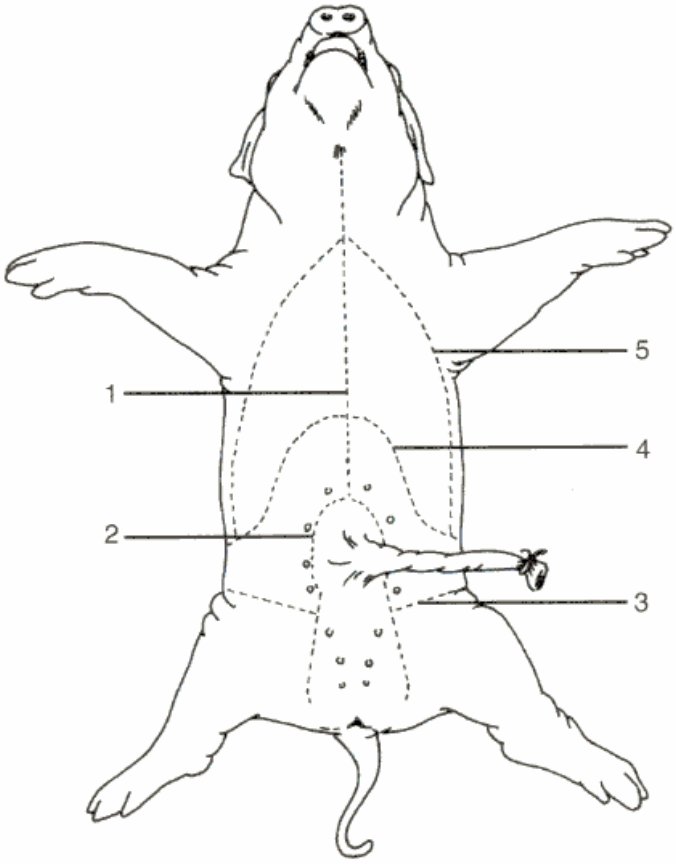
You will now work on opening the abdominal and thoracic cavities of the pig and identify structures. Remember, that to dissect means to "cut into pieces" from Latin *dessicare* - a careful dissection will make it easier for you to find the organs and structures. Be sure to follow all directions.

The Incision:

Place your fetal pig in the dissecting pan ventral side up. Use string to tie the legs behind the back of the pan. Use scissors to cut through the skin and muscles according to the diagram. Do not remove the umbilical cord.

After completing the cuts, locate the **umbilical vein** that leads from the umbilical cord to the liver. You will need to cut this vein in order to open up the abdominal cavity.

Your pig may be filled with water and preservative, drain over the sink if necessary.



Neck region:

Using the diagram to the right begin your dissection in the neck region. Try to cut as little as possible. Once you open the body cavity, you will generally be able to separate the different organs by simply pulling them apart with your fingers, forceps, or a probe. The more you cut things up, the harder it will be to figure out what you're looking at. Cut midline on the ventral surface of the neck to expose the underlying muscles. Carefully separate the muscles to observe the underlying structures. Locate and understand the functions of the following structures:

- **Larynx:** an enlarged structure on the trachea. If you cut it open, you can see the vocal cords inside.
- **Thymus gland:** an endocrine (hormone-secreting) gland that helps regulate the immune system. It's a large, spongy structure covering the ventral surface of the trachea and extends up along either side and also resides over the heart. It is easy to cut so be extra careful.
- **Thyroid gland:** another endocrine gland; it's a small bilobed (two parts) structure just posterior to the larynx. The thyroid secretes hormones that help regulate metabolism.
- **Trachea:** the airway; it's reinforced with rings of cartilage so it does not collapse.

- **Esophagus:** carries food from mouth to stomach; soft and muscular so it can move a food bolus by peristalsis. It is located dorsal to the trachea (but appears behind it because the specimen is upside down).

Thoracic cavity:
Vertebrates have true coeloms (a body cavity). In mammals, the coelom is divided into two main cavities: the thoracic cavity, which contains the lungs, and the abdominal cavity, which contains the digestive system. The thoracic cavity and the abdominal cavity are separated by the diaphragm. Note the many membranes lining the coelom and holding the organs in place.
Look for these structures in the thoracic cavity:

- **Lungs:** they have several lobes. Note how spongy the tissue is.
- **Heart:** muscular and easy to find. The heart is surrounded by a pericardial sac. Note the aorta, where high-pressure blood leaves the heart on its way to the systemic circulation. You may also see the right and left carotid arteries, which supply blood to the head. For now, don't spend too much time on the various lobes of the heart and the many blood vessels. Come back to these later.
- **Diaphragm:** a sheet of muscle and connective tissue that helps in breathing and divides the two cavities described here.

Abdominal cavity: digestion & absorption:

Locate and understand the functions of the following structures:

- **Liver:** very large and dark. It has several lobes. You'll need to lift it out of the way to see the organs beneath. The liver produces bile that is stored in the **gall bladder**. The gall bladder is a small organ attached to the underside of the liver; it's usually greenish due to the bile. It connects to the small intestine by the bile duct.
- **Stomach:** a muscular, sac-like organ that sits posterior to and to the left of the liver. Here is where the final stage of mechanical digestion occurs and gastric juices released by glands continue enzymatic digestion, particularly of proteins. At each end of the stomach are valves that regulate food entering and leaving the stomach. At the esophagus is the **cardiac sphincter valve**, and at the duodenum is the **pyloric sphincter valve**. View the inside of the stomach by slicing it open lengthwise.
- **Small & large intestine:** tube-like structures that continue the movement of food (now called chyme). The small intestine is first. The initial part of the small intestine is responsible for the final steps of enzymatic digestion and then eventually absorption of the degraded molecules. The large intestine primarily functions to compact the remaining waste material by absorbing water invested in the digestion and lubrication process. Also absorption of vitamins completes here.
- **Rectum:** the final portion of the large intestine where wastes are stored before elimination through the anus.
- **Mesenteries:** thin, transparent sheets of connective tissue containing blood vessels connecting the intestine and other organs.
- **Pancreas:** white and looks a little bit like cauliflower and located along the underside of the stomach, a **pancreatic duct** leads to the duodenum – first part of the small intestine. The pancreas also makes insulin, which is necessary for the proper uptake of sugars from the blood. It secretes digestive enzymes and buffers as well that contribute to the digestion of material within the small intestine.
- **Spleen.** The spleen is a flat organ located near the stomach. It performs several functions related to producing and maturing new blood cells and eliminating old ones. Blood passes through open sinuses in the spleen, rather than being confined to narrow blood vessels.

Identify the organ (or structure)

1. _____ Opening (valve) between stomach and small intestine.
2. _____ Stores bile, lies underneath the liver.
3. _____ Separates the thoracic and abdominal cavity; aids breathing.
4. _____ Membrane that holds the coils of the small intestine.
5. _____ The part of the small intestine just after the stomach.
6. _____ Empties bile into the duodenum from the gall bladder.
7. _____ The last part of the large intestine before it exits at the anus.
8. _____ Bumpy structure under the stomach; makes insulin.

Circulatory system

Note the following features:

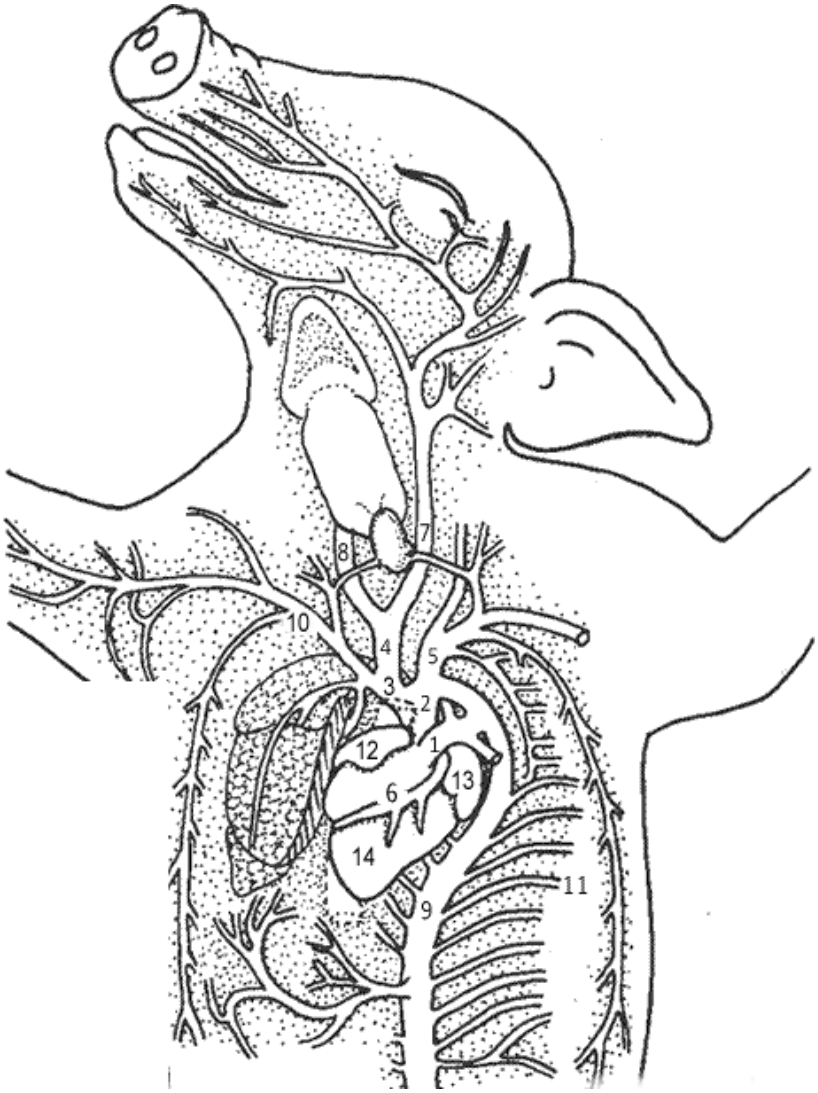
Arteries, which carry high-pressure blood away from the heart, are generally thicker walled than veins, which carry lower-pressure blood back to the heart. Mammalian hearts have four chambers. Each side of the heart has an atrium that receives blood from elsewhere in the body and a ventricle that pumps the blood out of the heart. The right atrium receives blood from the systemic circulation and passes it to the right ventricle, which pumps the blood to the pulmonary circuit. After the blood passes through the lungs it goes to the left atrium and then into the left ventricle, which pumps the blood into the systemic circuit. The first part of the systemic arterial circuit is the aorta, which soon branches out to supply various regions of the body.

Fetal circulation is different from adult circulation. In the fetus, blood doesn't get oxygenated in the lungs; it gets oxygenated at the placenta. The umbilical arteries carry blood from the fetus to the placenta. The umbilical vein carries blood from the placenta back to the fetus. (In the placenta substances are exchanged between fetal and maternal blood, but the blood itself does not mix.) Therefore, the most highly oxygenated blood in the fetus is in the umbilical vein. Blood from the umbilical vein gets mixed with the rest of the systemic circulation and returns to the right atrium. The blood entering the right atrium is the most oxygenated blood in the fetal heart, but it's the least oxygenated blood in the adult heart. The fetus has two key tricks to adapt to this fact: First, some of the blood that leaves the right ventricle bypasses the lungs. In an adult, this blood needs to go to the lungs to get oxygenated, but the fetus has a ductus arteriosus that short-circuits this blood flow, allowing some blood to go directly into the aorta and then into the systemic circulation. Second, in the fetal heart, there is an opening between the right atrium and the left atrium. This opening is called the foramen ovale. The foramen ovale is helpful in the fetus because it lets the oxygenated blood from the placenta get circulated faster. The foramen ovale normally closes up at birth, keeping blood flow of the two sides of the heart completely separate. In some people, the foramen ovale does not close up. This condition, called patent foramen ovale, can result in serious health problems.

Dissection of the Thoracic Cavity

1. Find the **diaphragm** again. Remember that the diaphragm separates the abdominal cavity from the thoracic cavity and it aids in breathing. Above the diaphragm, center of chest, is the heart.
2. Remove the **pericardium**, which is a thin membrane that surrounds the heart.
3. The structures visible on the heart are the two **atria** (12,13), the **ventricle** (14), which has two chambers not visible from the outside.

4. The most obvious vessel on the front of the heart is the **pulmonary trunk** (1). It curves upward and joins the **aorta** (2) - a vessel which arches from the heart and curves around to go to the lower part of the body -where it is called the **abdominal (dorsal) aorta** (9). The aorta supplies the body with blood.
5. The aorta will curve back and then branch in two spots – the right brachiocephalic (3) and the left subclavian (5).
6. The right brachiocephalic then branches into arteries – the **common carotid** (4) and the **right subclavian** (10). The subclavians supply blood to the arms and follow the clavicle.
7. The **common carotid** will branch into the **left** (7) **and right carotid arteries** (8). The carotid arteries supply blood to the head and neck.
8. Observe the **coronary vessels** (6) on the outside of the heart - these vessels supply blood to the muscle of the heart.
9. Easy arteries to find are the ones that run near the ribs. These are the **intercostal arteries** (11).
10. Lift the heart to look on its dorsal side (toward the back), you should be able to see the **anterior and posterior vena cava**, which brings blood from the body back to the heart. In addition, you should also be able to find the **left and right jugular veins** that drain blood from the head and run parallel to the carotids.



Identify the structure.

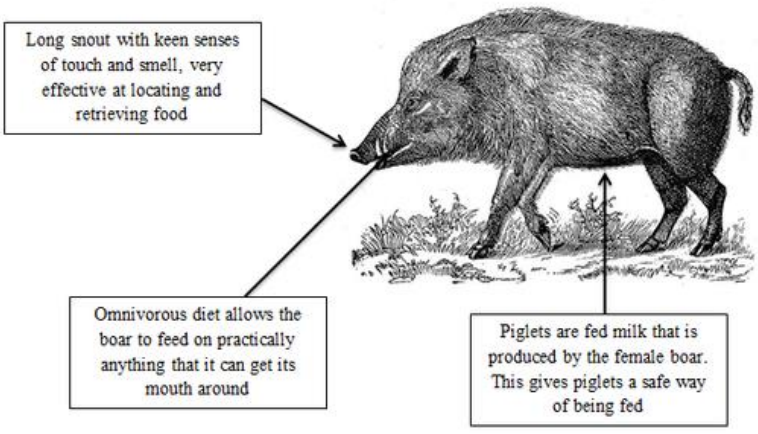
1. _____ Membrane over the heart.
2. _____ Blood supply to head
3. _____ Lower heart chambers
4. _____ Blood supply to lower body
5. _____ Muscle to aid breathing
6. _____ Returns blood to heart
7. _____ Large vessel at top of heart
8. _____ Arteries on heart surface.

Urogenital System:

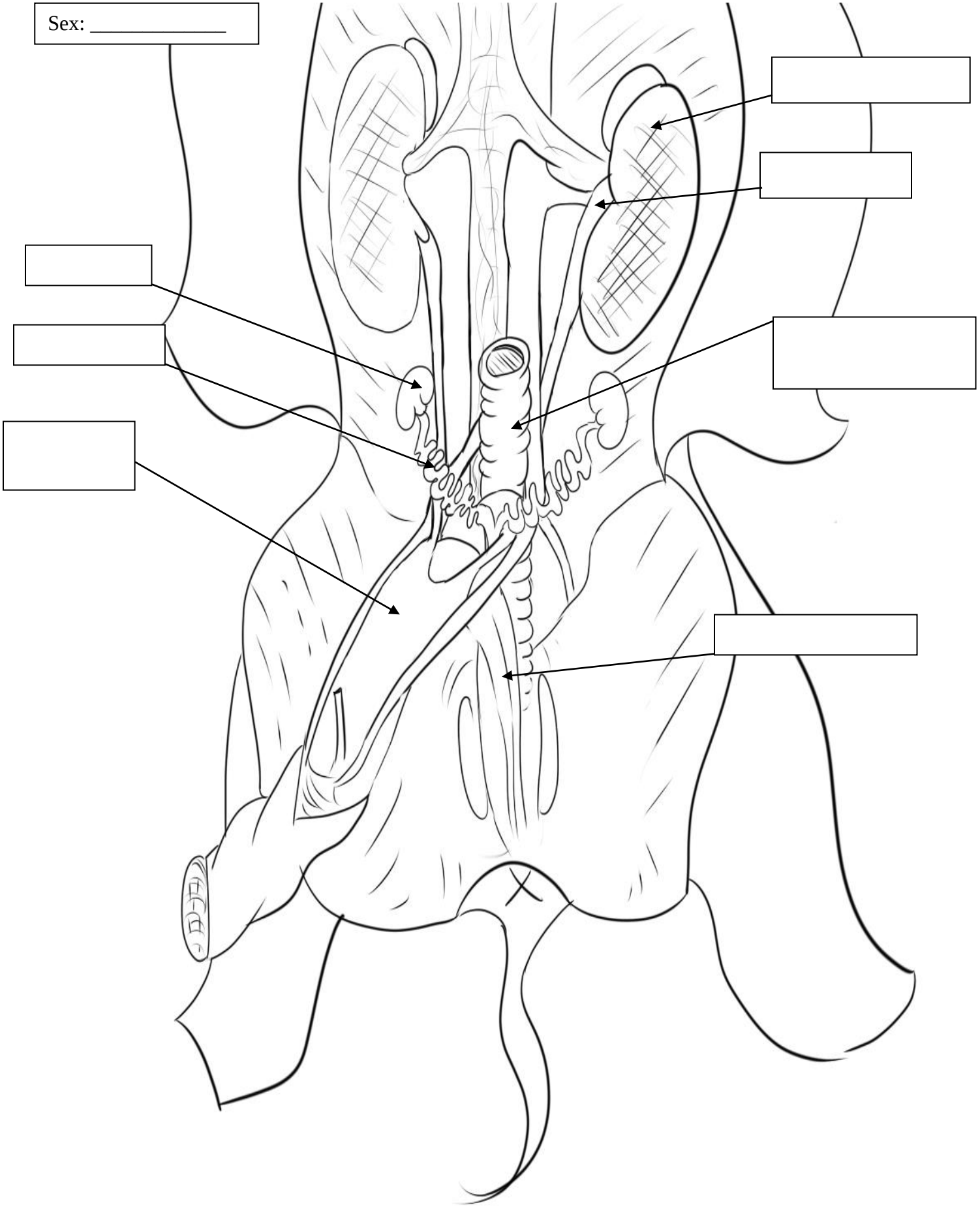
Locate and understand the functions of the following structures:

- **Kidneys.** The two kidneys are not actually located in the abdominal cavity; they occupy another coelomic compartment dorsal to the abdominal cavity. You won't see them until you move the intestines aside. The kidneys actually are located behind the mesentery lining the abdominal cavity. Gently break through this tissue. Urine from the kidneys goes into the urinary bladder via the **ureters**, and then through the urethra as it is eliminated from the body.
- **Urinary bladder & urethra.** The urethra is the tube that carries urine from the urinary bladder to the urinary opening. You can find the urinary bladder positioned between the two umbilical arteries.
- **Ovaries and uterus (females) or Testes (males).** The testes of males and the ovaries of females both arise from the same embryonic structures; however, the testes migrate during fetal development until they descend into the scrotal sac. The size of the testes varies significantly, depending on the age of the fetal pig.
    - Female
        - In the female pig, locate two bean shaped **ovaries** located just posterior to the kidneys and connected to the curly oviducts. These typically are quite small in the fetal pig.
        - Trace the **oviducts** toward the posterior to find that they merge at the **uterus**. Trace the uterus to the **vagina**. The vagina actually will appear as a continuation of the uterus.
    - Male
        - Find the **scrotal sacs** at the posterior end of the pig (between the legs), **testes** are located in each sac. Open the scrotal sac to locate the testis.
        - On each testis, find the coiled **epididymis**. Sperm cells produced in the testes pass through the epididymis and into a tube called the **vas deferens** (in humans, a vasectomy involves cutting this tube).
        - The **penis** can be located by cutting away the skin on the flap near the umbilical cord. This tube-like structure eventually exits out the urogenital opening, also known as the **urethra.**

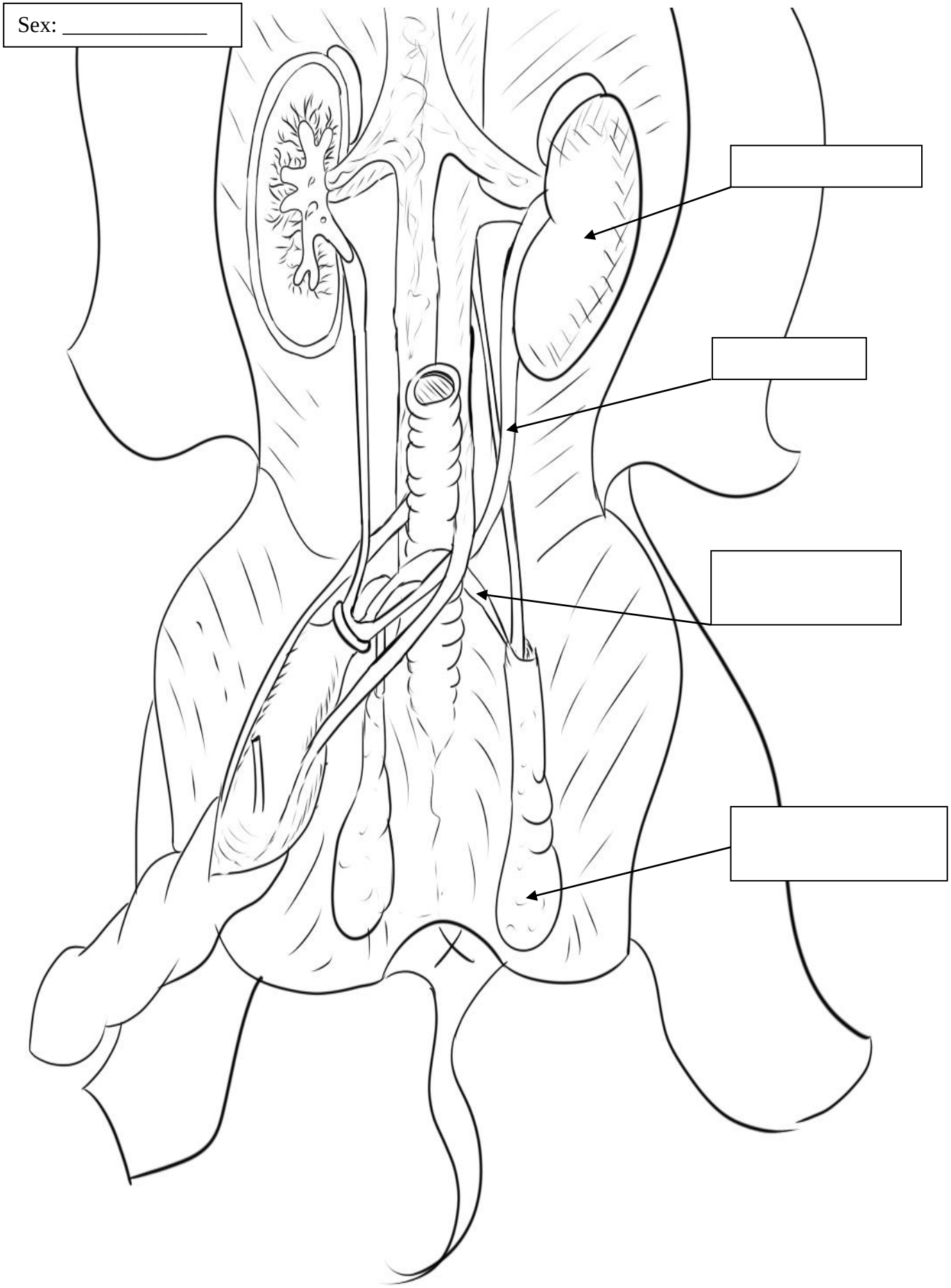
On the next two pages label the diagrams of the female and male urogenital systems. Identify which one is female and male. The final page is an unlabeled diagram of the major organs for which you have been responsible to identify. Label the missing structures.

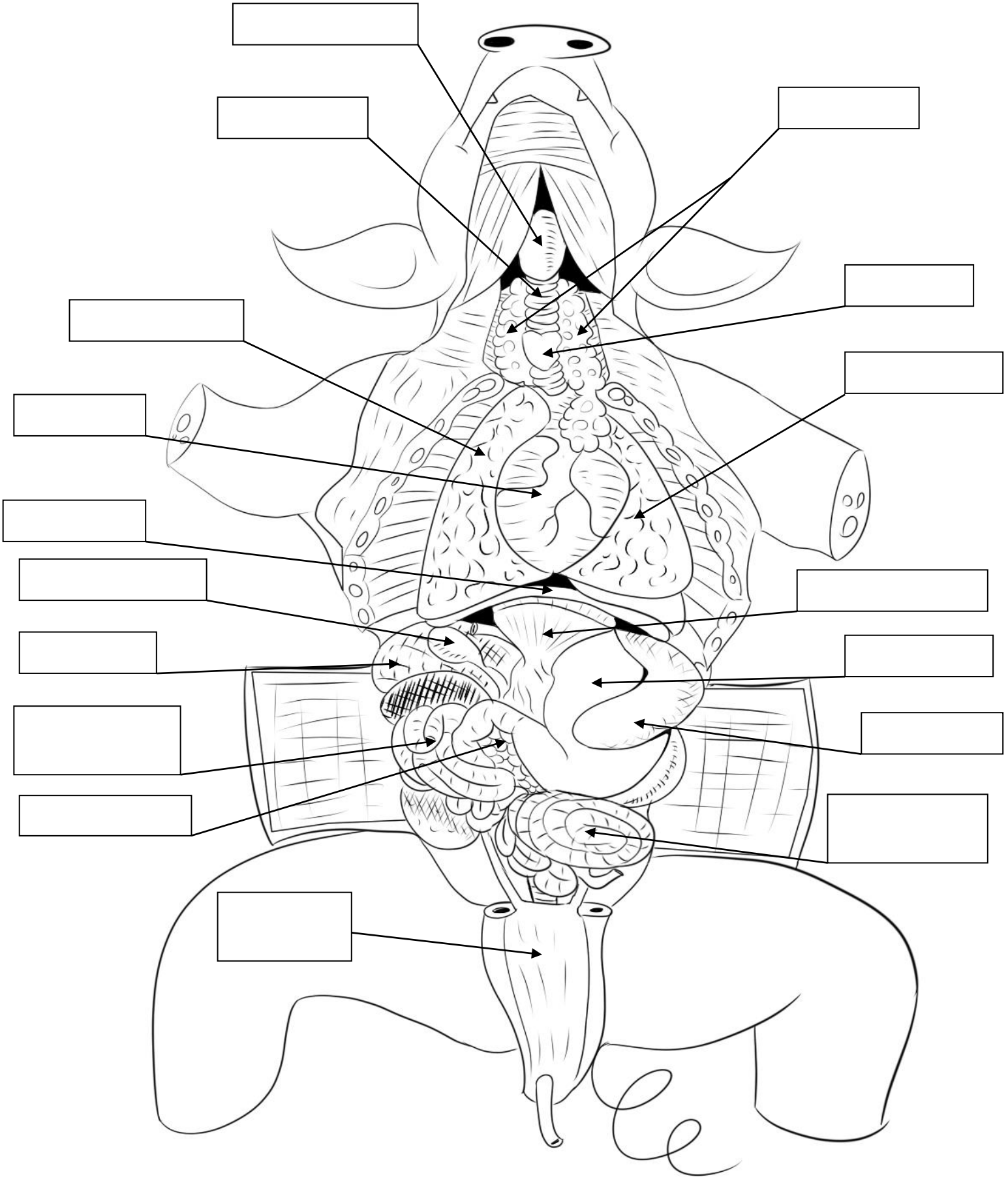


Sex: _____



Sex: _____





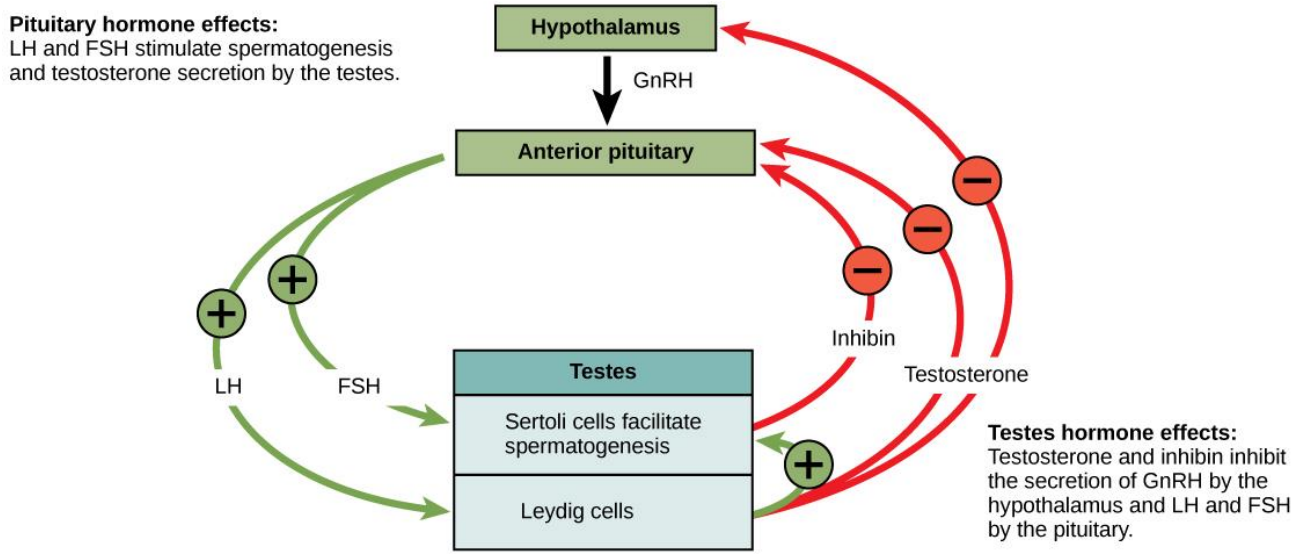
Reproduction and Development Lab

In this lab you will explore the physiology behind gamete production and maintenance of the gonads along with the processes involved from conception to birth. It's a lot of material! So we will focus on the major details. We will use a combination of presentation, discussion, microscope work, and documentary to fully explore the process of reproduction and development.

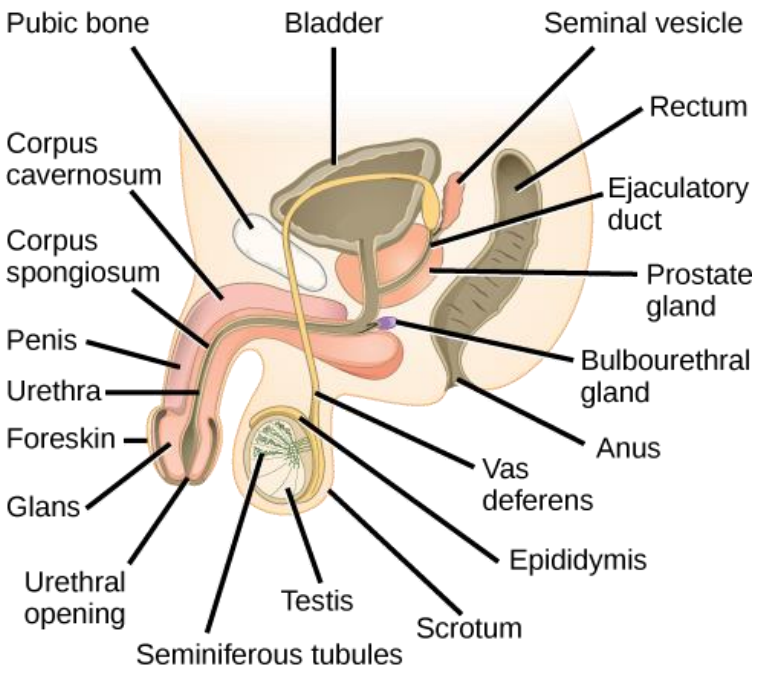
Hormonal regulation of gonads

Recall that the hypothalamus is responsible for controlling gonad activity by triggering the release of gonadotropic hormones from the anterior portion of the pituitary gland.

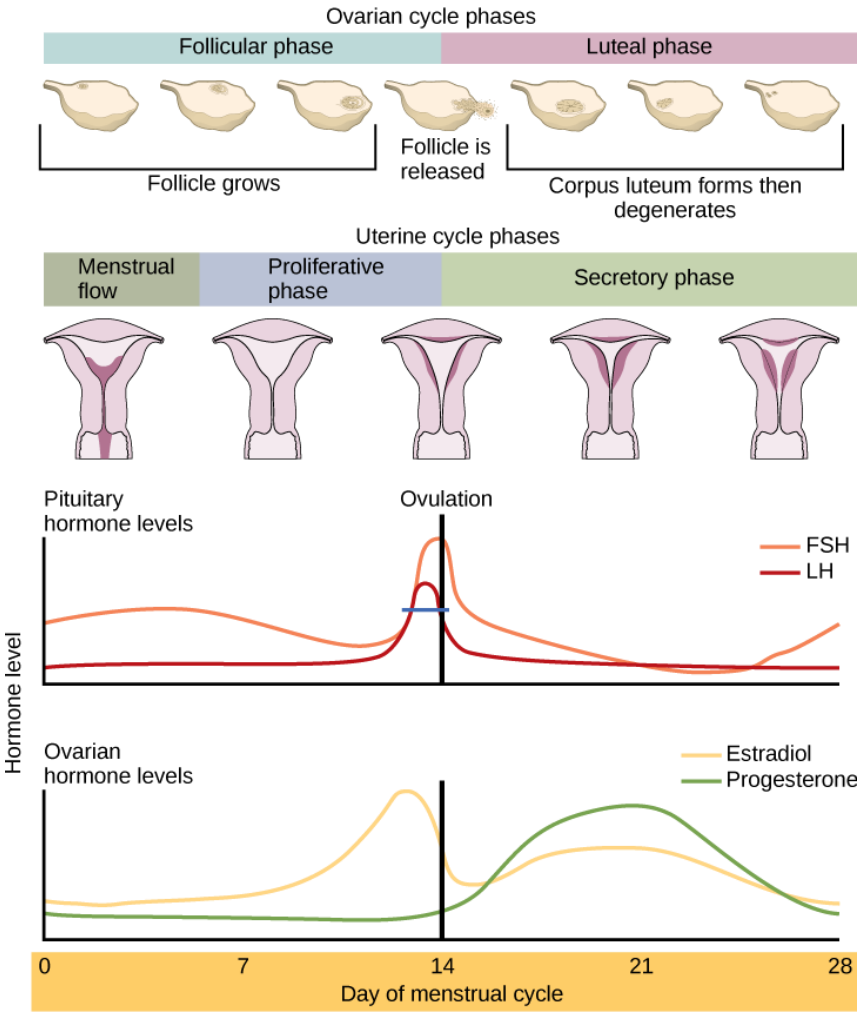
In males:



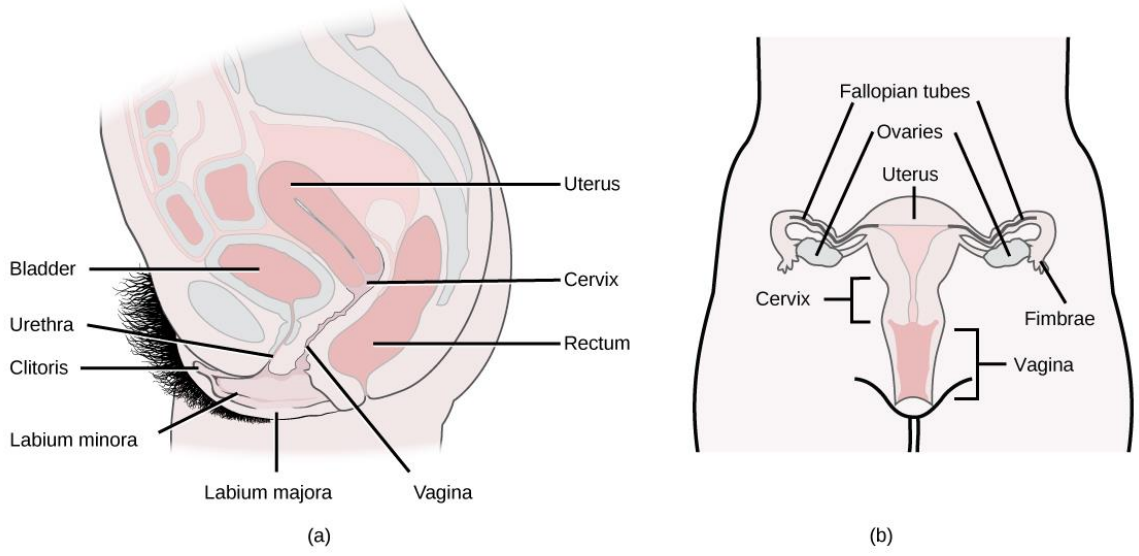
Ever wonder why the male gonads are externally located? Sperm are very sensitive to heat. In fact, sperm must develop at temperatures 2° C lower than average body temperature in order to be viable!



In females:



Note that with the female hormone profile there is both pituitary and gonadal components as in the male. The LH and FSH function similarly



Microscope

Get the following slides: testis cross section, semen sample, Graafian follicle (mature follicle about to release an ovum), and corpus luteum.

1. **Testis slide**: locate the following: cross section of a seminiferous tubule (area for sperm storage), Sertoli cells (responsible for supporting developing sperm cells) line the immediate inside edge of the tubules, and the developing sperm cells themselves. Interstitial cells between the tubules may be visible. These are responsible for producing testosterone. Start with the lowest magnification and work your way up to the next two powers.

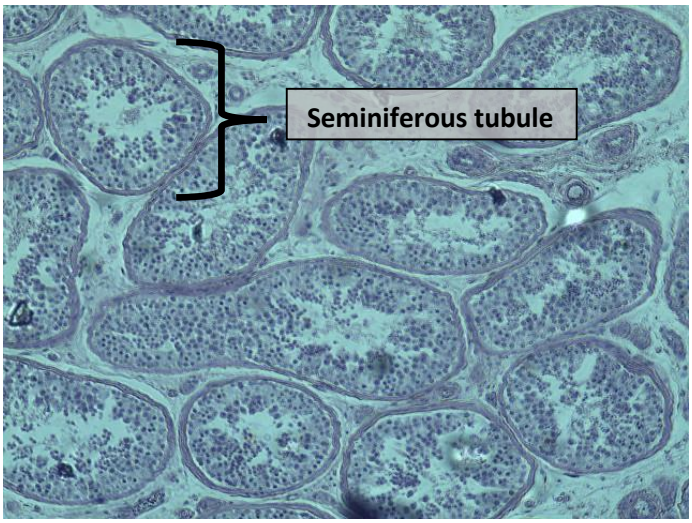
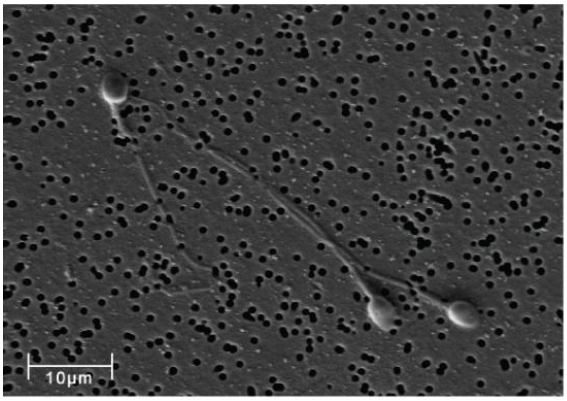
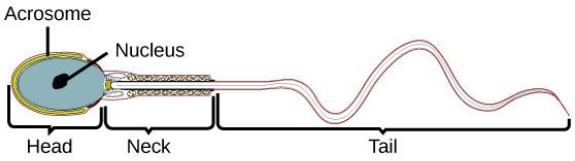


Photo credit: D. DesRochers

2. **Semen Sample:** This slide only has sperm visible. They only are visible on 40X. You must have medium to low light levels, and find the small specks on the lowest magnification first. Then work your way up. The neck region is indistinguishable from the tail using the light microscope.

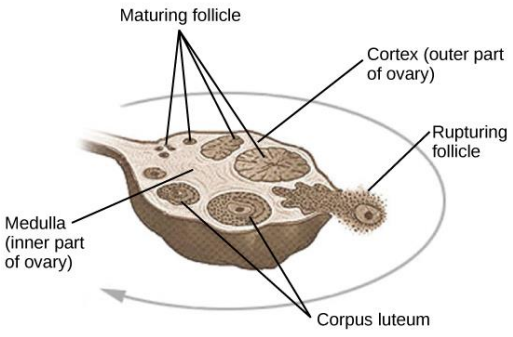


(a)

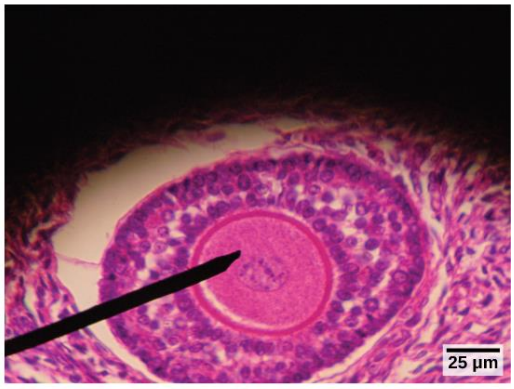


(b)

3. **Graafian follicle:** This structure is best visible under lowest magnification. Notice the ovum in the center of the follicle.



(a)



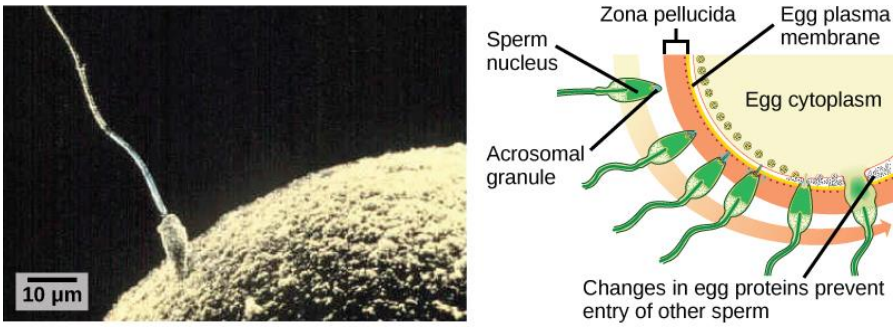
(b)

4. **<u>Corpus Luteum</u>:** Once the ovum has been released during ovulation, the remaining follicle thickens and produces progesterone. Refer back to the hormone profile for females from the previous section. The progesterone prepares the uterus for implantation of the embryo if fertilization occurs. If fertilization and implantation do not occur the corpus luteum degenerates and the cycle continues.

Recall from Principles of Biology I that meiosis is the ultimate process that produces both male and female gametes.

<u>Fertilization</u>

Once a male and female engage in sexual intercourse, fertilization may or may not occur. Recall that ovulation occurs in a very narrow window of a woman's cycle (see above). If the timing of sexual intercourses occurs within a woman's fertile time frame, then fertilization may occur (pictured below). The sperm cells swarm around the egg, and the first sperm cell to digest through the zona pellucida using the digestive enzymes stored in its acrosomal granule is the cell that will contribute its genetic information to the new individual.



(a) (b)

<u>Video</u>

Miracle of Life

This award-winning video from the 1980's may seem a little dated, but it still does an amazing job capturing the process of development that occurs over the nine months during which a fertilized egg becomes a human being.

Your assignment is to pay close attention to video and to take notes. Information from the video may be on this week's quiz.

Observation Field Notes

Recall from your time in BIOL 1107 that observation is the first step of the scientific method! Thoughtful observation comes with practice and is an important skill to develop regardless if you are a lab or field biologist! One of the most important steps in developing your observational skills is eliminating distraction. While you are working on this assignment you must (1) work alone and (2) avoid using your cell phone (unless you want to photograph your focal organism).

Today you must observe five different wild species for at least five minutes each. You need to observe at least one mammal and a second mammal or bird. The other three can be anything including insects. So head up on the trails, find a quiet spot near the creek behind Sequoya, and start observing. Use the following as guidelines.

Date _____ Name _____

Weather: _____

1. Species mammal _____ start time /end time _____

Habitat description:

Species description:

Behaviors:

2. Species mammal or bird _____ start time /end time _____

Habitat description:

Species description:

Behaviors:

3. Species _____ start time /end time _____

Habitat description:

Species description:

Behaviors:

4. Species _____ start time /end time _____

Habitat description:

Species description:

Behaviors:

5. Species _____ start time /end time _____

Habitat description:

Species description:

Behaviors