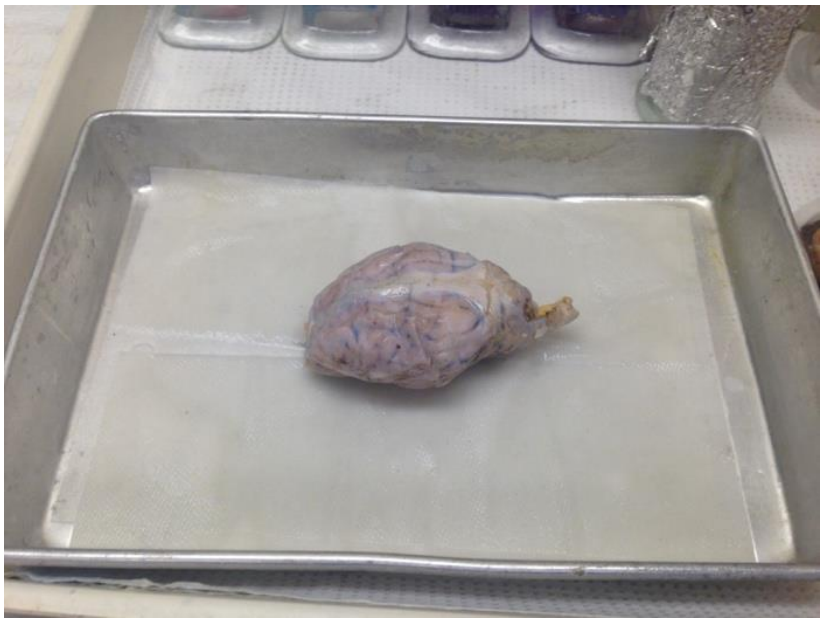
In-Lab Instructions for Dissection

For your lab report, ANSWER ALL EXERCISE QUESTIONS and pick 3 THOUGHT QUESTIONS to answer.

Step 0

Before class, your TA will remove each brain from its plastic bag and rinse with water. After rinsing, the brain is placed on wet paper towels in the dissection tray.



Unlike the brains shown in the preparatory material, the brains you will be dissecting are still encased in thick layers of membranes (*meninges*). These are important protective and support structures in the living brain. There are three layers of these membranes. The thickest and toughest of these meninges is the outer layer, the *dura mater*, which must be removed before we can dissect the rest of the brain.

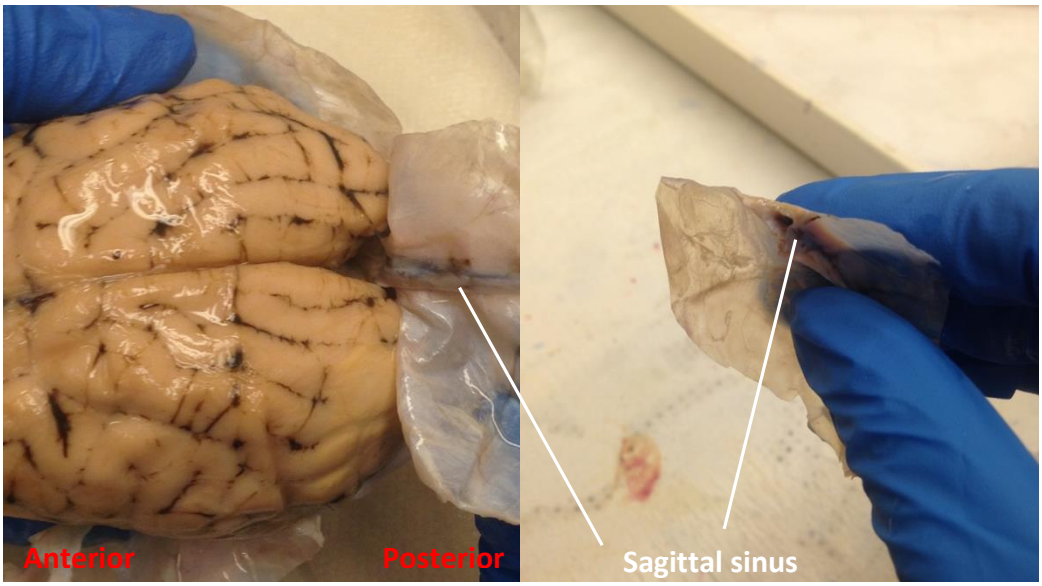
Step 1: Reorienting yourself to the sheep brain

1a. Examine your sheep brain in its meningeal sac. *Before cutting anything*, review the anatomical directionality of the sheep brain from your pre-lab material and determine anterior, posterior, dorsal and ventral in your own sheep brain.

Exercise 1: What would be different about the meninges and their structure if you could investigate the living brain in a living animal? Here, think about what the meninges are for?

<u>Step 2:</u> Removing the Dura Mater

2a. Begin at the dorsal, anterior part of the brain and carefully slice open the dura, gently pulling it away from the rest of the brain. You may need to cut it in several places to completely remove it from the dorsal surface.



As you remove the dorsal dura, inspect the underside of the membrane. Locate the *sagittal sinus* on the midline.

Thought Question 1: What is the sagittal sinus? What functions do sinuses serve in the nervous system?

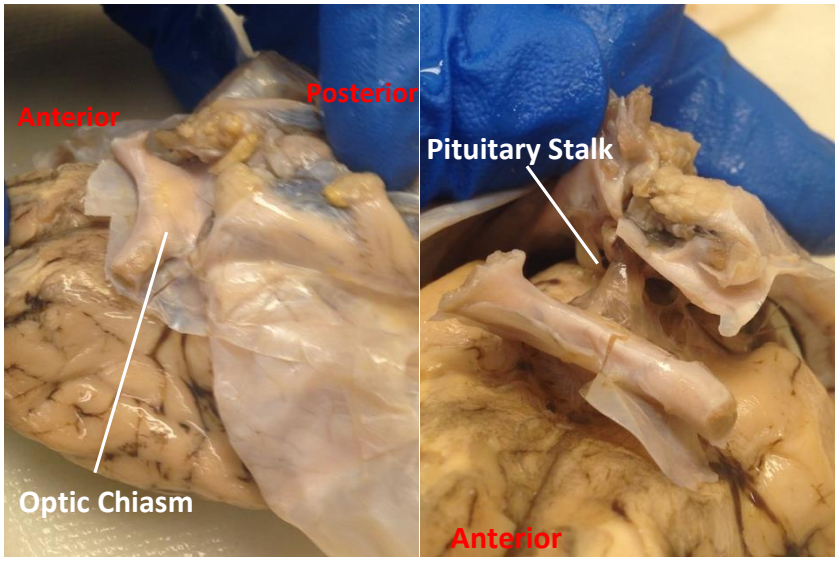
For future orientation, before you move on to remove the dura on the ventral side of the brain, make sure you can identify the *cerebellum* (posterior, unpaired), and the two hemispheres of the *cerebrum* (anterior, paired) on the dorsal side.

2b. Now turn to the ventral side of the brain and continue freeing it from the dura.

<u>Important Practical Notes!!!</u>

- Be particularly careful on the ventral side of the brain. There are many convoluted (and delicate) structures on this surface – try to keep as many of them as possible intact as you snip away the dura.
- You may accidentally cut or pull off large ventral nerves along with the dura, particularly the trigeminal nerves and the small nerves along the brainstem. If you do pull off structures, try to save them so you can refer back to them.

Perhaps the most prominent structures on the ventral surface of the brain are the optic nerves, (also known as the *optic chiasm*), which are situated directly beneath the cerebral hemispheres at the midline. Just posterior to the optic chiasm is the *pituitary gland*. Locate the stalk of the pituitary and snip it to help remove the dura.



The pituitary is also an important landmark on the ventral surface of the brain. The structures of the brain posterior to the pituitary are known as the *brainstem*, while those anterior to the pituitary are known as the *forebrain*.

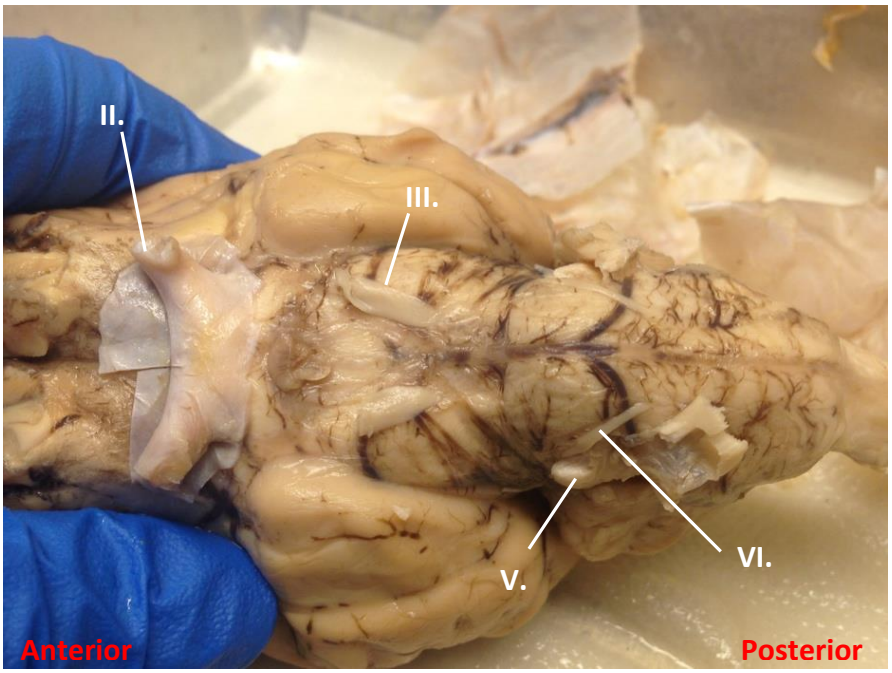
Thought Question 2: What is the pituitary? What are its functions in the body?

2c. As you remove the dura from the ventral surface of the brain, you will expose several of the *cranial nerves* (nerves that emerge or enter the central nervous system from the brain rather than the spinal cord). Cranial nerves are numbered as roman numerals, I-XII, roughly as they occur from an anterior to posterior direction in the brain. Most of the cranial nerves are associated with the brainstem (all but nerves I. and II.). The first cranial nerve, the *olfactory nerve*, emerges from the most anterior point of the brain and is almost never present in the brains used in this lab. However, many of the others should still be visible.

Exercise 2: Describe the differences that allow you to recognize a nerve from nervous system support structures (e.g. blood vessels, fat bodies, meninges) in fixed tissue.

Locate at least two of the following cranial nerves in the mid-ventral region of the brain:

II. Optic
III. Oculomotor
V. Trigeminal
VI. Abducens



Practical Note!

- Which nerve are you looking at? Some cranial nerves are more prominent than others. The easiest way to discriminate between cranial nerves is by their characteristic combinations of size and location (i.e. the position from where they enter and exit the brain).
- Once you have located the nerves on your brain (or if they came off and you can't find exactly where they came from), take a look at your neighbors' brain for comparison – they are in the same location in every brain.

Thought Question 3: How do you use anatomical information to infer the function of a nerve?

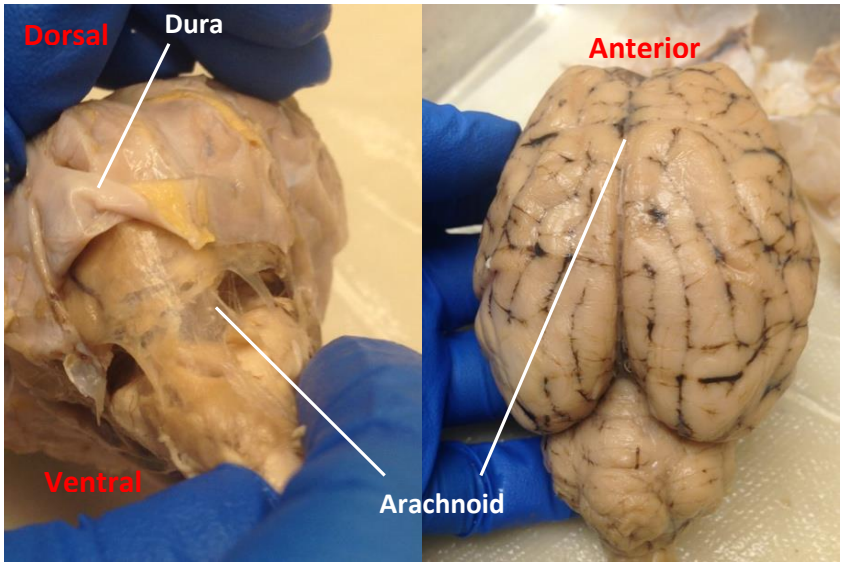
i) Below are a few factors you may or may not find helpful to consider. Does it help to know:

a) Where these nerves go to (or come from) in the intact animal?
b) If the nerve is entering the brain, exiting the brain, or both?
c) Where these nerves insert into the brain?
d) The relative size (diameter) of the nerves?

ii) Explain whether or not you can answer these questions about an individual nerve from gross anatomy (looking at and dissecting the brain from the body as you are doing today). What other information would you need?

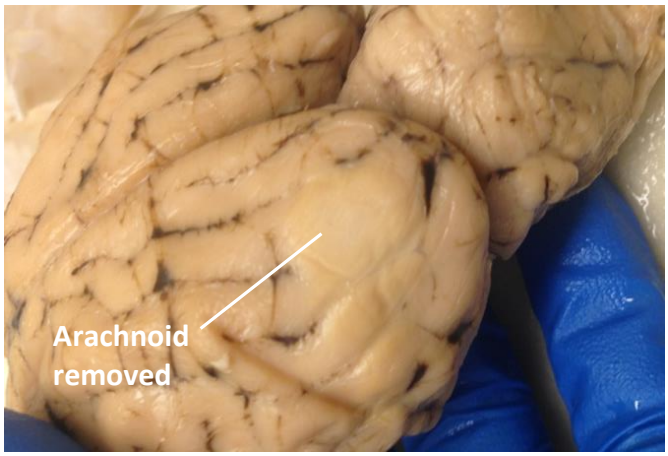
<u>Step 3:</u> Identifying and Removing the Arachnoid

3a. Gently lift the cerebellum in a dorsal direction - only a little, don't pull too hard! - to reveal the second, middle layer of meninges, the *arachnoid mater*. You should also be able to identify the arachnoid on the midline of the dorsal surface of the cerebrum, between the two cerebral hemispheres.



Exercise 3: What is physically different about the dura and the arachnoid? What does this suggest about the different functions they play in supporting the nervous system?

3b. Now that you've identified the arachnoid at the margins between large brain regions, look for it overlying the folded *cerebral cortex* on the lateral surfaces of *cerebral hemispheres*. Using a sharp blade, try to peel away a small portion of the arachnoid layer from the cerebral hemispheres, then inspect the brain's surface.

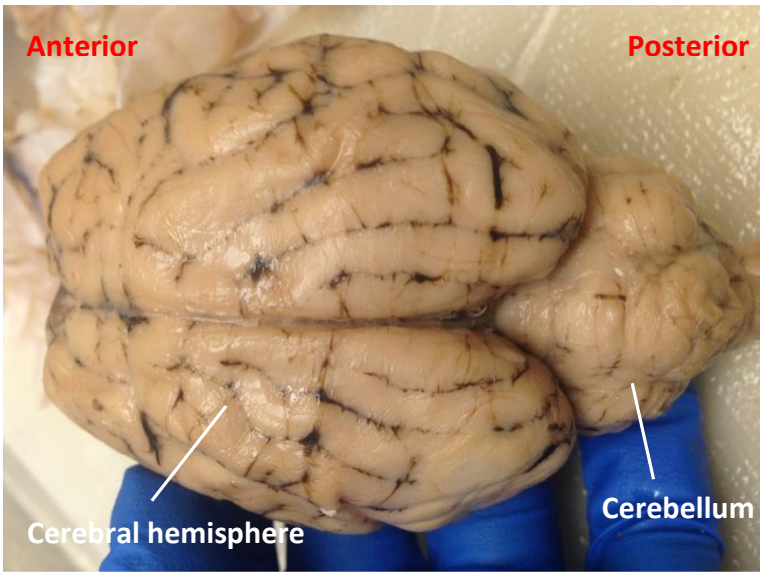


Thought Question 4: What happened to the blood vessels when you removed the arachnoid? Does this suggest something about the blood supply to the brain?

NOTE: The inner meningeal layer, the *pia mater*, is wrapped extremely tightly around the outer surfaces of the brain. It is also the thinnest and most delicate of the meninges, and because of this, it's practically impossible to visualize in our preserved sheep brains.

<u>Step 4:</u> Identifying structures on the surface of the brain

4a. Inspect the *cerebellum* and *cerebral hemispheres* on the dorsal surface of the brain. The surfaces of both the cerebrum and the cerebellum are visibly folded structures. In the case of the cerebral hemispheres, this folded surface is known as the *cerebral cortex*. Different brain functions are localized to specific regions of cortex.



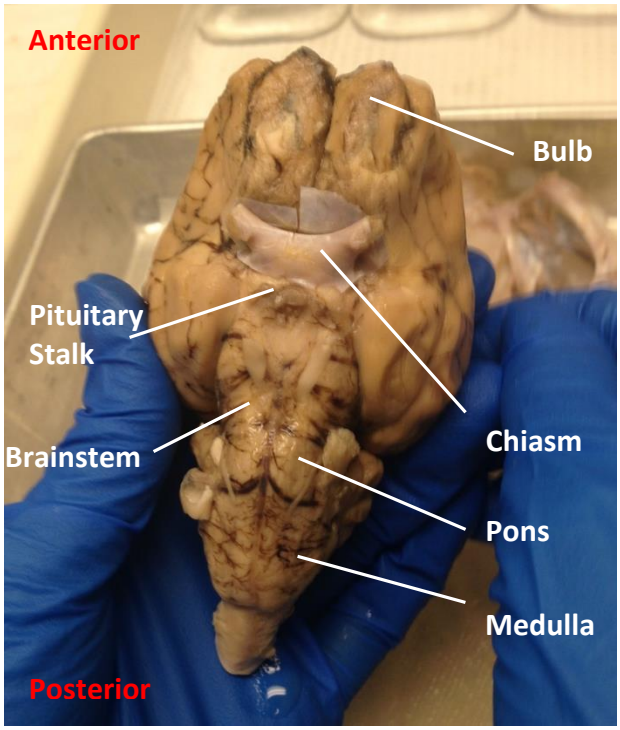
4b. In brain anatomy, the raised part of a fold is known as a *gyrus*, and the crease between two folds is a *sulcus*. Inspect the pattern of sulci and gyri on your brain, then compare that to the pattern of sulci and gyri on the guide brain, and to your neighbors.

Thought Question 5: How folded is the cortex?
Brains of large animals like sheep and humans tend to have more folds in their cortex than the brains of small animals, like rats and hedgehogs. Why do you think this is?

4c. Turn your brain over to inspect the ventral side, and locate the following major structures:

- *Olfactory Bulbs* (anterior, paired)
- *Optic Chiasm* (midline ventral to the cerebrum, X-shaped)

- *Pons* (a prominent bulging structure in the brainstem, directly ventral to the cerebellum)
- *Medulla* (brainstem structure posterior to the pons)



Exercise 4: Consider the optic chiasm.

i. What was cut (during the original brain extraction) that connects to the anterior part of the optic chiasm?

ii. The chiasm is x-shaped. What is the significance of this for the animal's brain and what it does with the information coming through the chiasm?

iii. Why do you think there is no similar chiasm associated with the olfactory bulbs, or with the other cranial nerves?

4d. Continue your inspection of the ventral surface in a posterior direction, cutting away any loose layers of the arachnoid as you go, and locating each *cranial nerve* or nerve group. See how many of the following nerves you can identify in your brain now (check your neighbors if you can't find them on your own brain). The guides on the next two pages should help.

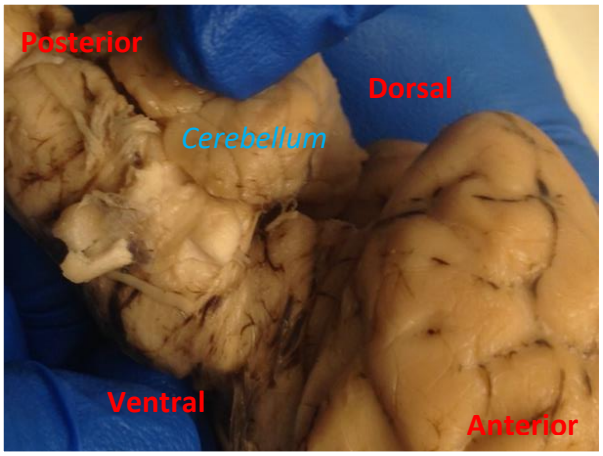
I. Olfactory *(very likely absent)
II. Optic
III. Oculomotor
IV. Trochlear
V. Trigeminal
VI. Abducens

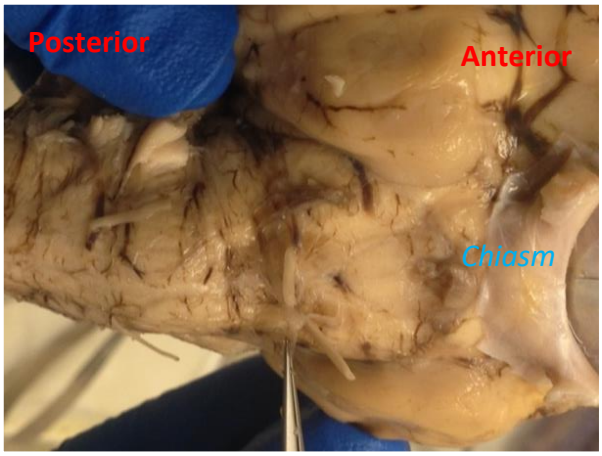
VII. Facial
VIII. Acoustic/Vestibuloacoustic
IX. Glossopharyngeal
X. Vagus
XI. Spinal Accessory
XII. Hypoglossal

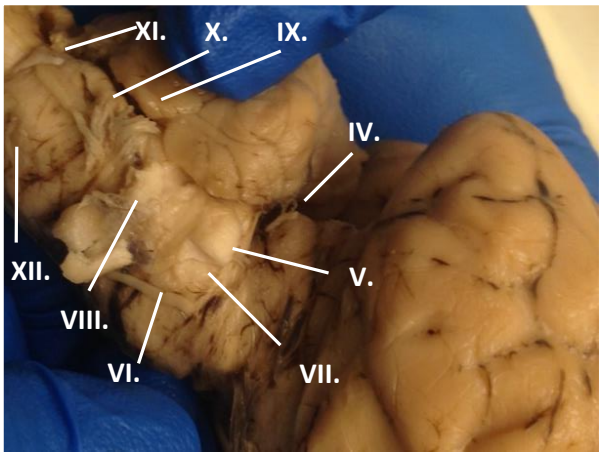
Practical Notes!

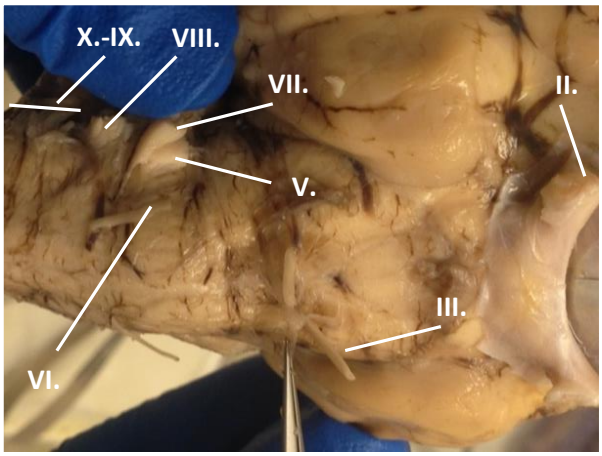
- Use major landmarks you have already identified to orient yourself to which brain surface (dorsal, ventral, or lateral) and anterior/posterior level you should be looking at when using these photos as a guide.

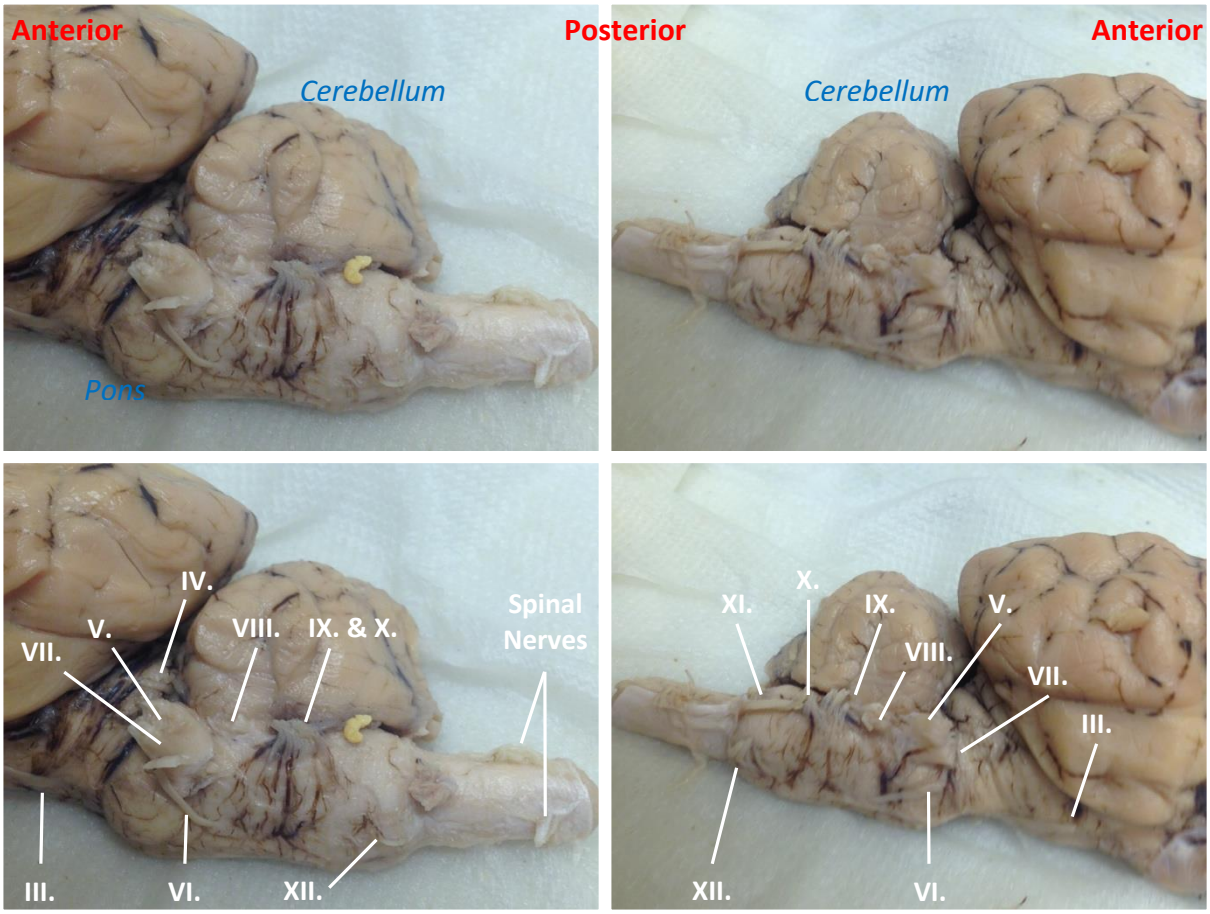
- Some nerves may be impossible to find due to prior damage to the brain. However, you also may find that it will help to come back to complete this section after finishing the instructions for Step 6.











Thought Question 6: Different cranial nerves have very different functions. For one of the cranial nerves you have located, research (or use your existing knowledge) to list the following properties:

a) *What part of the body does it connect to the brain?*
b) *Is its function sensory, motor, or a mixture of the two?]*
c) *Is its function somatic, autonomic, or a mixture of the two?*

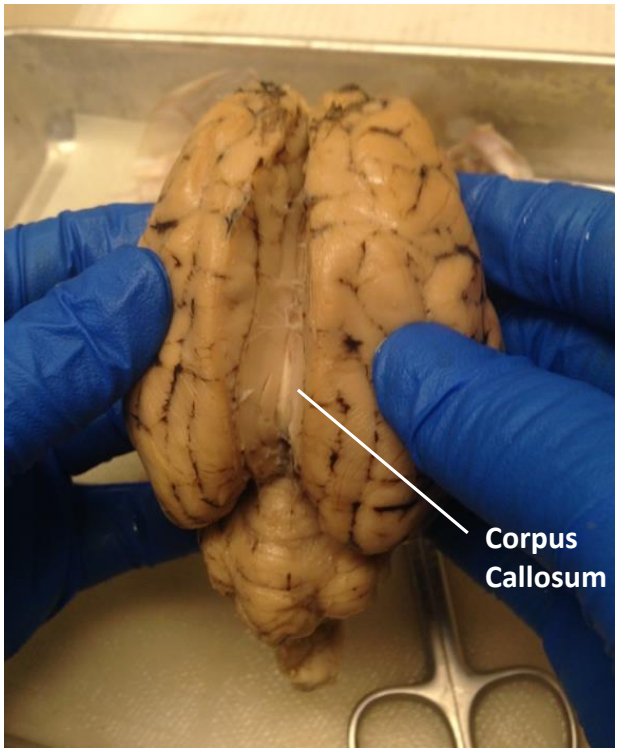
<u>Step 5:</u> Opening up the major divisions of the brain

Exercise 5: Based on your observations to this point, if you had to divide up the brain, how many distinct regions would you divide it into, and why?

<u>Important Practical Notes!!!</u>

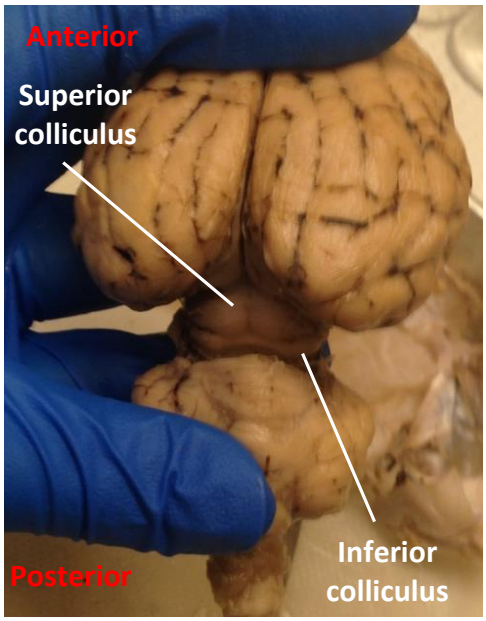
- Do all of the following instructions slowly, carefully, and gently, only a little at a time. The purpose is to unfold the brain a little, not to pull it to pieces.

5a. On the dorsal surface of the brain, cut through the arachnoid along the major sulcus on the midline (the *sagittal sulcus*), and if necessary cut any arachnoid lying between cerebrum and cerebellum as well. Ease the two cerebral hemispheres apart in a lateral direction to expose the *corpus callosum* at the bottom of the sagittal sulcus.



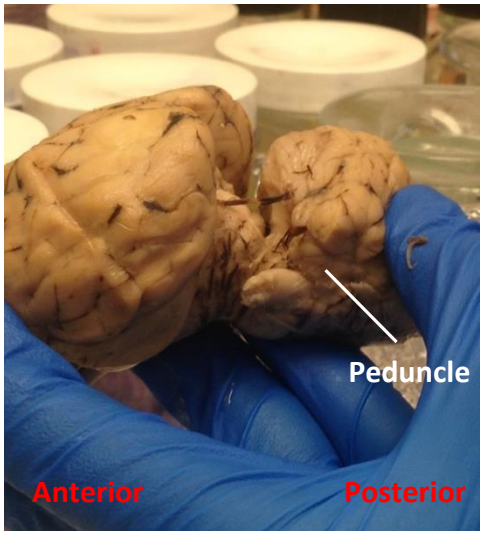
Thought Question 7: What is the corpus callosum? What is its function?

5b. Next, pull on the cerebellum and bend it toward the posterior end of the brain, exposing the brainstem structures that lie ventral to the cerebral hemispheres. Locate the *superior colliculi* (anterior, paired) and the *inferior colliculi* (posterior, unpaired).



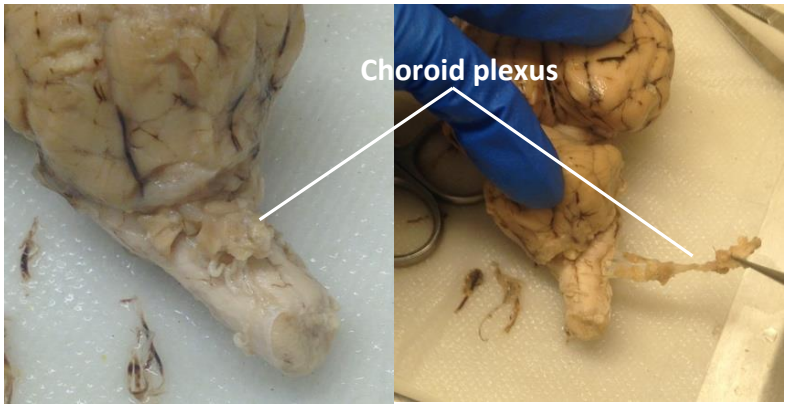
The superior and inferior colliculi are sensory-processing structures that process visual and auditory information respectively.

5c. Carefully snip at the remaining arachnoid around the base of cerebellum on its lateral and posterior faces to expose several cranial nerves (VIII.-X), and the structures that connect the cerebellum to the brainstem, the *cerebellar peduncles*.



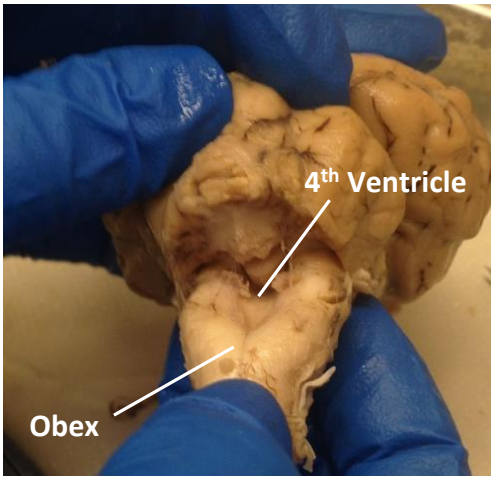
5d. Inspect the dorsal surface of the posterior to the cerebellum. You should find some amount of loose, lumpy, non-neural tissue that can be carefully pried and cut away from the surface of the brain. This is an example of *choroid plexus*, a support tissue

that is found within the brain's ventricles. It secretes *cerebrospinal fluid*, a liquid that surrounds and supports the central nervous system within the *meninges*.



Thought Question 8: What are the different functions of cerebrospinal fluid in the support of the brain? Why do you think the central nervous system has a separate fluid system that is distinct from the other fluid systems in the body, such as blood and lymph?

5e. Once you have removed the *choroid*, snip away any *arachnoid* still holding the posterior *cerebellum* to the brainstem, then bend the brain forward to expose the structures that lie ventral to the cerebellum. Locate the *fourth ventricle*, and at its posterior end, the *obex* (a landmark that marks the posterior end of the brainstem and the beginning of the spinal cord).



<u>Step 6:</u> Splitting the brain in half

6a. Return to the dorsal surface of the *cerebrum*. Carefully cut through the *corpus callosum* in a sagittal plane, splitting the brain (including the *cerebellum* and

brainstem) down the midline. You may find some midline structures are torn off – if so, save them for later inspection.

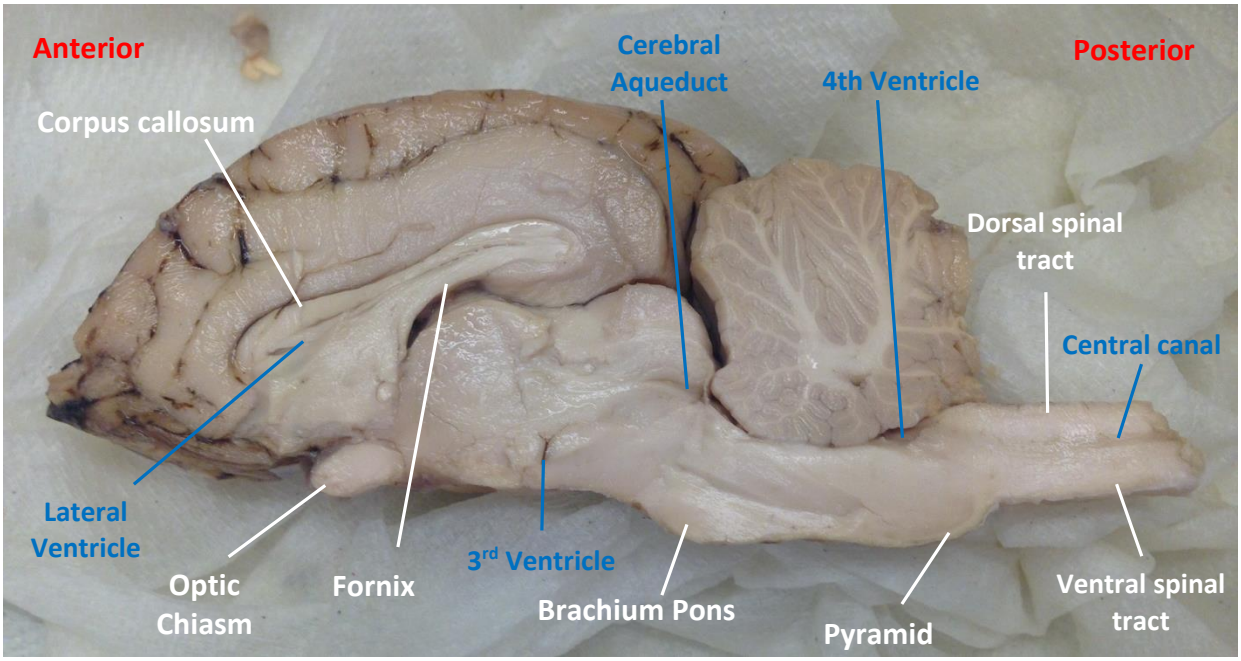
6b. After splitting the brain, note its striking two-tone appearance – there are regions of *grey matter* (beige in these brains due to the method by which they were preserved), and regions of *white matter*.

The white color of white matter is due to the presence of a support tissue called *myelin*, which functions throughout the nervous system (both central and peripheral divisions) as electrical insulation around *axons*. Locate regions of white and grey matter in the cerebrum, the cerebellum and the brainstem. Then look back at the *cranial nerves* you have already found to confirm that they are also examples of white matter.

Exercise 6: If white matter is white due to the presence of myelinated axons (axons that are wrapped in myelin) what cell types, or parts of cells, make up gray matter? Why do you think axons might need more electrical insulation than other parts of a neuron?

6c. Locate the following important *white matter tracts* in the split brain:

Corpus Callosum	*Brachium Pons*
Fornix	*Medullary Pyramids*
Optic Chiasm	*Dorsal and ventral spinal tracts*



Thought Question 9: For one of the tracts you have located, research (or use your existing knowledge) to briefly describe:

a) *its major orientation (e.g. does it travel medial-lateral, anterior-posterior?)*
b) *the regions that it connects within the brain.*

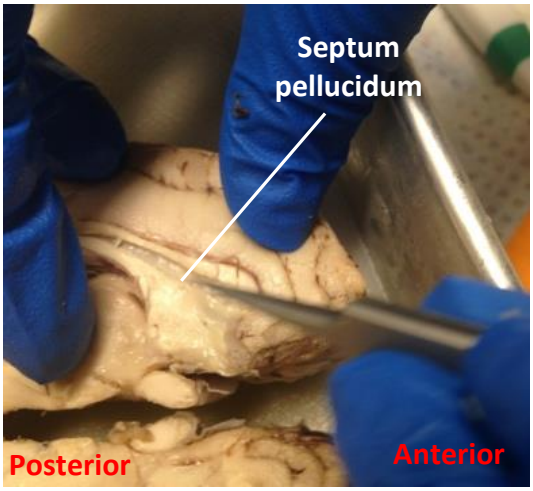
6d. Now identify the *lateral ventricles* within the cerebrum, between the *callosum* and the *fornix*, and see if you can locate the thin membrane that lay between them, the *septum pellucidum*. Depending on how your brain was split, this may be attached to one hemisphere or the other, or it may have ripped apart during the dissection.

The *lateral ventricles* (and the *fourth ventricle* we identified earlier) are components of a connected system of *ventricles* within the central nervous system. The ventricles all contain some amount of *choroid plexus*, and they are continuous with the *central canal* that runs down the center of the spinal cord. Try to identify the other components of the *ventricular system* (labelled in blue on the guide) in your brain – they may not all be visible in your brain, so check your neighbors if you can't find them.

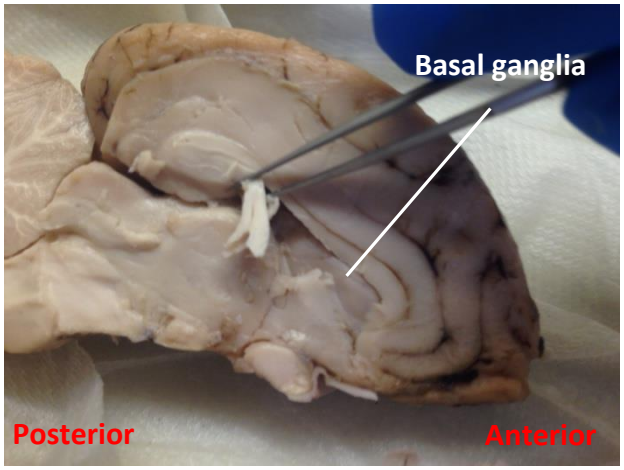
3rd Ventricle	*4th Ventricle*
Cerebral Aqueduct	*Central canal* (spinal cord)

Exercise 7: There are many cavities visible between parts of the brain, but only some of them are considered to be ventricles. What do you think is the significance of this designation?

6e. Gently probe inside one of the *lateral ventricles* (shown below is the left hemisphere of the dissected brain), and see if you can remove the *septum pellucidum* (if it is present), and then the *choroid plexus* inside (it will probably come apart as lumpy chunks of membrane).



Continue to remove the *choroid* till you expose the lateral wall of the ventricle, revealing a glimpse of the nuclei of the *basal ganglia*, an important set of *subcortical* structures within the *cerebrum*. It may help to snip the *fornix* ventrally and lift it away, as shown below.

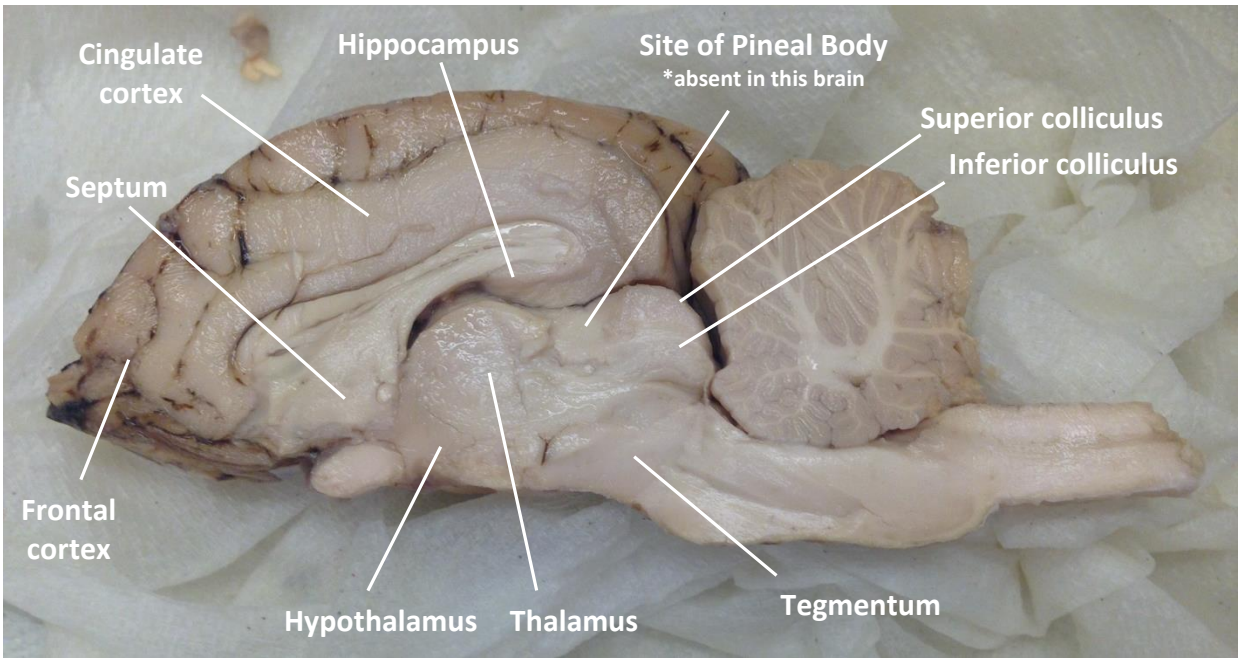


Thought Question 10: What does the term 'subcortical' refer to? Why do you think we draw a distinction between cortical and subcortical structures?

6f. A number of other recognizable *gray matter* structures are exposed in a midline dissection, shown here in an intact right side of the brain. Locate the following regions:

Cingulate and *frontal cortex*

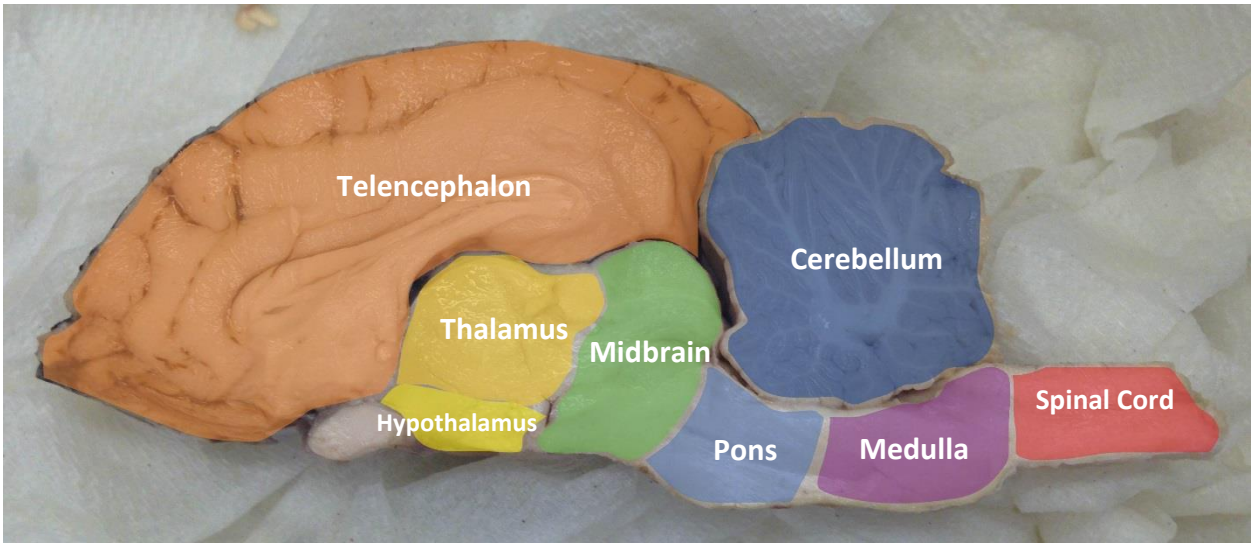
Septum (different from the *septum pellucidum*)
Hippocampus
Hypothalamus
Thalamus
Pineal Gland/Body *(may be lost during splitting of the brain)
Superior and Inferior Colliculi
Tegmentum



Thought Question 11: For one of the regions you have located, research (or use your existing knowledge) to briefly describe:

a) *the general function(s) of this region*
b) *the principle connections of the region (where does it get inputs from and what does it make outputs to)?*

7c. Inspect your split brain once again, and locate the official anatomical boundaries of the *major divisions* of the central nervous system, as they are shown below:



The most basic division within the central nervous system is between the *brain* and the *spinal* cord. The brain is then subdivided into three major regions – the *forebrain*, the *midbrain*, and the *hindbrain*. Two of these regions (the forebrain and the hindbrain) are further subdivided into two distinct components.

Brain

i. *Forebrain* ('Prosencephalon')
- i-a. *Telencephalon* (septum, hippocampus, basal ganglia and cortex)
- i-b. *Diencephalon* (hypothalamus and thalamus)

ii. *Midbrain* ('Mesencephalon')
- ii. (colliculi and tegmentum)

iii. *Hindbrain* ('Rhombencephalon')
- iii-a. *Metencephalon* (cerebellum and pons)
- iii-b. *Mylencephalon* (medulla)

Spinal Cord

Exercise 8: Consider your split brain one more time. Does the official guideline of the divisions of the brain on the previous page make sense to you?

a) ***If you were dividing the sheep brain into its major divisions and subdivisions, how many would you have, and where would you choose to place their boundaries?***

b) ***Are the divisions you've chosen different than the official boundaries outlined by textbooks? If so, can you suggest a reason why anatomists could have chosen boundaries for the divisions of the brain that seem uneven in terms of their gross anatomy?***