ARTICLE
Dissection of the Hippocampus Proper and the Associated Structures in Preserved Horse Brains

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The limbic system is a complicated region of the brain to teach in an undergraduate neuroanatomy course. The students often struggled with the complexity and interconnection of the structures of this system to other cortical regions. The following is a description of two dissections that used preserved horse brains to help students visualize the major components of this system and also comprehend the topographic organization of some of these structures to other regions or structures of the brain.

Key words: hippocampus proper, fimbria, fornix, limbic system, neuroanatomy specimen

INTRODUCTION
When teaching a neuroanatomy course, the primary specimen for an undergraduate neuroscience course is formalin fixed brains from sheep, cows or horses. The dissection of these specimens is relatively easy for the undergraduate student and exposes them to the major aspects of the central nervous system. In addition to these preserved specimens, there is also a “treasure trove” of multimedia resources for instructors on the internet as described by Grisham (2006) and Jenks (2009). The benefits of using these sites are that they are excellent supplements to lab dissections, can provide an alternative to the wet-lab experience if the student is sensitive to formalin, and can be used if departmental resources are tight or the student is opposed to dissection (Grisham, 2006 and Korey, 2009). However as Grisham (2006) also pointed out, an instructor who used these websites must carefully check the information and labels because there were some discrepancies in terminology and some of the websites were limited in the information or images they provided about specific regions of the central nervous system. One area that seemed to have limited labeled dissection images was the limbic system. The only websites that offered selected images of the hippocampal system (hippocampus proper, fimbria, crus of the fornix, body of the fornix and columns of the fornix) were the University of Scranton, Behavioral Neurosciences Lab - Dissection of the Sheep Brain, Michigan State University - The Navigable Atlas of the Sheep Brain, San Francisco's Exploratorium - Sheep brain dissection: The anatomy of memory, and St. Louis University - Dissection of the Sheep Brain (Grisham, 2006).

Given the significance of the hippocampus proper in memory and the recent discoveries that it is one of the few areas in the adult brain where neurogenesis continued to occur, it was surprising that images of this region were somewhat limited and did not give a comprehensive view of the key components of the hippocampal system. Instead these websites focused on select portions of the hippocampal system using parasagittal sections, cross sections or prossections in which a small portion of the dorsal surface of the telencephalon was removed to show only part of the hippocampus proper in the floor of the lateral ventricle.

Therefore, to help students understand the organization of the hippocampal system the dissected specimen needed to clearly illustrate that the hippocampus proper is located within the temporal lobe and is an arched structure that forms the caudal (posterior) portion of the floor of the lateral ventricle. In addition the specimen should show the fimbria along the edge of the hippocampus proper and where the hippocampus proper ends near the level of the splenium of the corpus callosum and how the fimbria consolidated into the crus of the fornix. A second specimen should also show how the crus of each fornix unite at the midline to form the body of the fornix within the ventral (inferior) border of the septum pellucidum.

The two specimens that seemed to be the most beneficial to the students were one that gave a lateral view of the hippocampus proper, fimbria and crus of the fornix and then a second specimen that gave a dorsal view of the previously mentioned structures and also showed the body of the fornix and the position of the hippocampus proper within the lateral ventricle and its relationship to the caudate nucleus.

Specimen Preparation
Horse brains still within the dura mater were purchased from Ward’s Biological. The dura mater was removed from each of the brains and then one was used to provide a lateral view and the other for a dorsal view of the hippocampus proper and associated structures.

A. Lateral view of the Hippocampus proper and associated structures
To expose the hippocampus proper from the lateral aspect, the pyriform lobe on the lateral side of either the right or left cerebral hemisphere was identified. Then using a sharp smooth-edged knife, thin slices approximately 1-2 centimeters thick were sliced off the end of the pyriform lobe until the rounded end of the hippocampus proper could be seen. At this point, a pair of curved scissors was used to continue removing the pyriform lobe and exposing the ventral most portion of the hippocampus proper. At this
point, thin sections of the lateral side of the cerebral hemisphere from the occipital to the temporal lobe were removed until the caudal portion of the lateral ventricle was opened and the arched portion of the hippocampus proper was exposed (Figure 1). However, care must be taken when removing this region of the cerebral hemisphere because it is very easy to inadvertently slice off a portion of the hippocampus proper with the cerebral hemisphere. Once this step is completed the hippocampus proper from its ventral most portion within the pyriform lobe to the arched portion within the caudal portion of the lateral ventricle was exposed. The fimbria, beginning of the crus of the fornix and the thalamus will also be exposed at this point. To expose the remainder of the hippocampus proper and the crus of the fornix, curved scissors or a small scalpel were used to continue removing thin sections from the temporal and frontal lobes along with a significant portion of the caudate nucleus (Figure 2).

**B. Dorsal view of the Hippocampus proper and associated structures**

To provide the students with a dorsal view of the hippocampus proper and some of the associated structures, a sharp smooth edged knife was used to remove horizontal slices (1-2 centimeters thick) of the cerebral hemispheres from the occipital lobe to the frontal lobe. This process opens a small window into the caudal most portion of the lateral ventricle within both cerebral hemispheres. At this point using a small scalpel or curved scissors, the opening into the lateral ventricle was lengthened so that the entire dorsal surface of the hippocampus proper was fully exposed along with a portion of the choroid plexus and caudate nucleus (Figure 3).

The next step after exposing the dorsal surface of the hippocampus proper was widening the opening so that the caudate nucleus, crura of the fornix and body of the fornix were more visible. To accomplish this, forceps were used to gently remove the choroid plexus from each lateral ventricle. Then a small scalpel was used to remove thin horizontal sections from each internal capsule until the head and body of the caudate nucleus was exposed and the crura could be easily identified on the lateral side of each hippocampus proper (Figure 4). During this step, it is very important that attention be paid to the close proximity of the caudate nucleus and the internal capsule and thin sections be gradually removed in order to avoid damage to the caudate nucleus. Then a fine pair of scissors were used to trim the corpus callosum and the septum pellucidum so that the crura of the fornix could be seen converging into the body of the fornix (Figure 4).
DISCUSSION

The limbic system was one of several areas of the brain that students typically had a strong interest in. However, this area was difficult for students to comprehend because of its complexity and interconnections with other cortical regions.

Like many anatomical courses, neuroanatomy can be overwhelming. This course presented a significant amount of detailed information and anatomical terms and caused students to come in with the idea that “rote memorization” was the only way to survive this course. Also, if the course was organized so that students had the impression that details were more important than comprehension then the result was students with only a superficial understanding of anatomy. Another issue that added to the student’s misconception about anatomy was the perception that dissection is outdated or unnecessary because of extensive visual resources on the World Wide Web (Miller et al., 2002). Additionally, undergraduate students were unskilled at dissection which in turn added to the dissection time and the general anxiety about damaging the specimen. The result of all this was students who were hesitant to dissect and more likely to rely exclusively on multimedia resources. The question then became how can a student enrolled in a neuroanatomy course have a laboratory experience that was “pedagogically meaningful and time efficient and promote three-dimensional comprehension” regarding the hippocampus proper and associated structures (Johnson, 2002). The answer to this question was not that one pedagogical tool (multimedia, static images from textbooks or atlases, prosection or dissection) was better than the other; instead it seemed a mix of tools was helpful to students in terms of promoting deep learning.

Previous research into the study methods and academic success of students has shown that students used either active or passive learning methods. Ward and Walker (2008) hypothesized those students who used “active study methods would have better grades and improved retention of the material after one year.” The result of their study of first year veterinary students in anatomy showed that there was not a significant difference between active and passive learning methods, student achievement in the course or their recall of the information after one year. Instead the results of this study showed that student success was affected by how the students processed the information not the method that was used for processing the information. Those students who used deep processing would manipulate the course information, integrate it with other information from courses, and make it personally meaningful versus the students who used superficial processing in which they were only concerned with getting it right and giving back the information as it had been presented to them (Ward and Walker, 2008).

In terms of the anatomy of the limbic system, most basic neuroanatomy texts focused primarily on the hippocampus proper, fimbria and fornix (crus, body, and columns) as a prelude to discussing the Papez circuit. This circuit is one of the most complex loops within the brain and most texts provided a simplified schematic drawing of the major components and its afferent and efferent connections. For most students, the difficulty in learning about this circuit occurred when they were required to take the static drawing and visualize this circuit within the brain they were dissecting. The students of today require more than just a lecture, lab dissection and static images. Instead they expect a dynamic environment in which multiple modalities are used; and as Ward and Walker (2008) had shown both passive and active learners can be successful in an anatomy course indicating it’s not necessarily the learning method that is the critical factor but the method and tools used in processing and integrating the course information.

Therefore, the purpose of this article was to present an additional pedagogical tool that would help students comprehend the topographic organization of the limbic system and its function with minimal preparation. Also this dissection was used to make the laboratory experience more than a “simple labeling exercise or the rote memorization of a long list of structural detail” (Miller et al., 2002). The laboratory experience when studying the limbic system should invoke the students’ natural tendency to make observations, explore the specimen through dissection and aid in the natural process of discovery in order to enhance comprehension of this system (Miller et al., 2002).

Overall, it was relatively easy to produce the two specimens. A skilled dissector would be able to dissect out the hippocampus proper and associated structures in the specimens within an hour or less. The benefits of using these prosected specimens, based on subjective analysis, was reduced student dissection time because these
prosected specimens were used as a guide during dissections, and increased student comprehension of the topographical organization of hippocampus proper, fimbria, crus of the fornix, body of the fornix, caudate nucleus, thalamus and the lateral ventricle. Additionally these specimens were also used by the students as a guide for interpretation of their serial cross sections made in the lab.

One weakness of this paper, however, was the absence of an objective analysis of the benefits of this specimen to the students. This course was a new development for the department and as such the idea for this specimen did not come until late in the semester when it was not possible to do an objective analysis with the current class. Therefore, to truly know if this specimen was beneficial to student comprehension a mixed methods design with quantitative and qualitative analyses needs to be conducted.

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