Technologies such as 3D printing and virtual/augmented reality have great potential for improving the teaching of highly spatial topics such as neuroanatomy. We created a set of 3D printed and virtual brain cross-sections using a high-resolution MRI dataset. These resources have been made freely available via online repositories. We also report a pilot study of the use of both the physical and virtual specimens in the classroom. Students completed a lab exercise where they used either the 3D printed or virtual brain sections to order a set of axial slices from dorsal to ventral. They then labeled the different structures that they found useful in determining the slices’ positions. We measured the students’ ability to localize 2D brain cross-sections before and after the lab exercise. Overall, we saw pre- to post-test increases in accuracy on a brain cross-sections task compared to a lecture-based neuroanatomy instruction.

Key words: neuroanatomy education; three-dimension modeling; virtual reality

Neuroanatomy is a particularly challenging subject for students at all levels due to the complexity of the material and need for three-dimensional (3D) spatial visualization (Guillot, et al., 2007). Students report low interest, poor understanding of the content, and low confidence levels (Javaid, et al., 2018). The dissection of the human brain has been the gold standard in teaching neuroanatomy but has increasingly been replaced by physical and computerized models (McLachlan, et al., 2004). The recent growth of 3D printing and virtual/augmented reality (VR/AR) technologies has led to increasingly sophisticated and more affordable materials. The use of these media in neuroanatomy education increases student engagement and satisfaction with an overall improvement in the learning experience (Pani et al., 2014) but there are still many challenges for neuroscience educators wishing to include hands on human neuroanatomy exercises. Highly detailed physical models, such as plastinated specimens, can be prohibitively expensive, and publisher-based brain atlases may be less than ideal both in terms of additional cost for the student and lack of flexibility for the instructor (e.g., the desired atlas may be tied to a less desirable book or platform). There are many free or low-cost applications available on the web and iOS/Android platforms. Popular examples include Brain Tutor 3D, 3D Brain, and Pocket Brain. These apps can be very helpful for students learning neuroanatomy. For instructors, however, these programs may contain too much or too little information, may not fit well with the specific goals of a particular lesson nor be well maintained as operating systems are updated. Thus, some instructors may desire greater control over how virtual content is used and integrated into their courses.

Game-Changers for Neuroanatomy Educators
Several factors have made the creation of virtual and physical neuroanatomy materials easier, cheaper and more accessible across different institutional settings. These are:

1) The availability of open magnetic resonance imaging (MRI) datasets and free brain segmenting software that allows users to import a variety of medical imaging formats and then visualize and create various structural segments that can be exported and used in 3D printing and virtual reality applications.

2) Growth of availability of makerspaces at all levels of education with access to 3D printing technologies (Ford, & Minshall, 2019).

3) Free 2D/3D game development engines which require less programming knowledge and allows publishing across platforms (i.e., the same content can be built to an online format, Mac, or PC desktop or iOS/Android devices).

First Steps
Five years ago, we began our initial foray into using 3D printing to create neuroanatomical teaching materials. Our specific goal was to help students visualize different cross-sections in the axial and coronal planes by creating 3D prints of axial and coronal cuts. A colleague at a nearby imaging center sent us her own brain volume collected using a 3 Tesla magnet with permission to use it in the development of teaching materials. We used 3D Slicer to segment this brain volume in 12 axial and 12 coronal slices and printed them using a Prusa i3 MK3S+ fused deposition modeling (FDM). The resulting printed slices were less than ideal, with a stiff and unnatural feeling to the touch and no way to directly infuse the surface details of the top/front or bottom/back of the model onto the print. We experimented with printing the faces of the model and then adhering them
to the print. However, this was not terribly durable. Additionally, we found that the images derived from the 3 Tesla scans did not have the same level of anatomical detail as actual physical specimens. Thus, we were initially stymied in our efforts.

**Technological Developments**

Two recent developments reignited our interest in creating detailed 3D brain slices for the classroom. The first was the publication of a high-resolution MRI volume made by scanning a deceased brain for 100 hours in a 7 Tesla MRI scanner at 100 µm isotropic resolution (Edlow et al., 2019). This dataset was archived and made publicly available at various repositories. The second was the acquisition of a more advanced 3D Polyjet printer by our University makerspace.

**Current Project**

Our goal was to create a set of 3D brain objects using a recently published high-resolution MRI dataset. These virtual objects can be used both for 3D printed specimens as well as for use in developing VR/AR content. In this paper, we will describe our development process for creating these materials, our instantiation of the resulting 3D models as both physical and virtual objects and finally a pilot study of their use in a classroom exercise.

**MATERIALS AND METHODS**

**Slice Model Creation**

We downloaded the Flash 25 100 micron volume in native space from the Dryad data repository (Edlow et al., 2019) and loaded them into 3D Slicer 4.10.1, a free medical imaging software developed by NIH (https://www.slicer.org). There are multiple free 3D brain visualization programs available. We chose 3D Slicer because it was the program with which we have had the most experience.

The volume was segmented into 13 axial slices and 12 coronal slices using Slicer on a Windows based PC with an i9 CPU running at 3.60GHz with 32GBs of RAM and an 2080Ti NVIDIA graphics card. All 25 slices were exported as STL files. Png images of the top/front and bottom/back of each slice were captured using screenshot and edited in GIMP, a free image manipulation program similar to Photoshop. The background of the image was first visually inspected, and any errant bits erased by hand. The fuzzy select tool was then used to select and make the black background transparent so that only the brain face was left visible in the file.

**Blender for Post-Processing**

The exported STL slice models were opened in Blender, a free open source graphics program, where 2D images of the top/front and bottom/back surfaces of each slice were added as textures to the top/front or bottom/back of the model using UV unwrapping, a technique in which a 2D texture is mapped onto a 3D object. The model meshes were also decimated to create a simpler geometry for 3D printing and exported using the OBJ format. Final models for use in virtual reality applications were exported using the FBX format.

**3D Printing**

13 one-inch-thick axial slices were printed using a Stratasys J750 Polyjet printer in the University of Wyoming Innovation Wyrkshop (https://www.wyrkshop.org/). Polymer jetting is a form of 3D printing that uses finely controlled streams of photopolymers that are cured with UV light. This method provides much greater detail resolution (~16 microns) than more commonly available 3D printing such as fused deposition modeling (FDM). Polyjet printing also allows multiple colors and materials to be combined within a single print. The cost of printing the 13 slices was $1,200.

**Virtual Reality**

The game engine Unity (version 2020.3.15f2) was used to create a desktop VR experience meant to mimic using the physical slices. Leap Motion controllers (www.ultraleap.com) were used to allow students to interact with the slices on the computer monitor using their hands. These controllers are available for purchase on the internet and cost around $100. The program was run on a MSI gaming laptop with an i7 processor running at 2.20GHz with 16 GB of RAM and a NVIDIA GTX 1070 graphics card. This program would first show axial slices stacked together, then on a key press the slices would separate and the user can reach out and virtually grab and pull out individual slices for examination.

**Pilot Study**

We conducted a pilot study to examine whether interacting with the physical or virtual axial slices had any effect on students’ ability to visualize cross-sections. In this initial study, we only used the axial set due to both practical (axial slices are easy to stack physically while coronal slices pose more of a challenge) and financial constraints. Data from a
large lecture style course were used as a control condition. This study was approved by the University of Wyoming IRB.

Participants
Participants were drawn from 3 classes. Ten undergraduates from an upper division course in Cognitive Neuroscience, 10 graduate students in a graduate Neuropsychology course and 33 undergraduates from an upper division Sensation & Perception course provided consent for their class data to be used for research purposes.

Brain Cross-Sections Measure
We created a 16-item multiple-choice measure of a student’s ability to locate a 2D MRI cross-section (8 axial and 8 coronal) using a mid-sagittal reference. We drew these items from the University of British Columbia Functional Neuroanatomy teaching site (https://www.neuroanatomy.ca) using the CC BY-NC-SA 4.0 license. As shown in Figure 3, students must choose which cut displayed on a mid-sagittal image corresponds to the displayed MRI image. Items were designed so that one or two structures (e.g., presence of the thalamus or cerebellum) were key to the correct answer.

In-Class Exercise
Students in the Cognitive Neuroscience and graduate Neuropsychology courses completed a lab exercise where they used either the 3D printed or virtual brain model to order a set of axial slices printed on paper from dorsal to ventral. They then described and labeled the different structures that they found useful in determining the slices’ positions. Students worked in groups of 3-4 and were randomly assigned to complete the assignment using either the physical slices (N=8) or the desktop VR program (N=12), but not both.

Control Condition
Students in the Sensation & Perception course received a brief overview of brain anatomy as part of a larger lecture reviewing neuroscience concepts such as the neuron, gross anatomy, neuropsychology and functional imaging. The review of brain anatomy consisted of two slides. The first focusing on the gross anatomy of the outside of the cortex

RESULTS
Brain Models
The final set of axial and coronal OBJ files for 3D printing have been made freely available for download from the University of Wyoming Libraries Sketchfab account:

https://sketchfab.com/uwlibraries/collections/uw-spatial-cognition-lab-axial-brain-slices
https://sketchfab.com/uwlibraries/collections/uw-spatial-cognition-lab-coronal-brain-slices

A guide documenting the steps we used creating the models in in 3D Slicer and Blender is available here https://github.com/minearlab/Brain-Models-Projects.

We also created a separate repository for a Unity asset package that contains both a virtual axial slice set and a coronal set. This is also hosted through the University of Wyoming Library at https://doi.org/10.15786/19912684.

Figure 2. Student interacting with the virtual slices using a Leap motion controller.

Figure 3. Example coronal item from cross-sections test. Images were taken from the UBC functional neuroanatomy site (https://www.neuroanatomy.ca) CC BY-NC-SA 4.0.
Behavioral Results
Comparing Physical to Virtual Models
We first tested for any gains in performance on the cross-sections measure after the class exercise using a mixed factorial ANOVA with the type of course (undergraduate Cognitive Neuroscience v. graduate level Neuropsychology), and presentation method (physical v. virtual slices) as between participant factors and testing time (pre or post exercise) and the type of cross-section image tested (axial v. coronal) as within participant factors. The dependent measure was proportion of items correct.

There was a main effect of testing time with an increase in performance from pre to post-test \( F(1,16) = 11.6, p < .01, \eta^2_p = .42 \) (pre-test \( M = .34, SE = .04 \), post-test \( M = .44, SE = .05 \)) and there was a main effect of course \( F(1,16) = 9.5, p < .01, \eta^2_p = .37 \) with lower scores for the graduate students in Neuropsychology \( (M = .25, SE = .06) \) compared to undergraduates in Cognitive Neuroscience \( (M = .53, SE = .07) \). However, the main effects of presentation method \( F(1,16) = 2.4, p = .14 \) and type of cross-section image tested \( F < 1 \) were not significant. Additionally, no interactions between any of the factors were significant.

Learning Gains Compared to a Lecture-Based Course
We compared the gains seen using the slices (physical or virtual) to cross-sections data collected from a larger lecture-based Sensation & Perception course before and after a slide-based neuroanatomy review. We collapsed across presentation method and cross-section image type given the lack of differences seen in the previous analysis and analyzed the data with a 3x2 mixed factorial ANOVA with the type of course (Cognitive Neuroscience, Neuropsychology, and Sensation & Perception) as a between-participants factor and testing time (pre v. post) as a within-participants factor. There were main effects of course \( F(1,16) = 5.4, p < .01, \eta^2_p = .18 \) and testing time \( F(1,150) = 12.7, p = .001, \eta^2_p = .20 \) and a significant interaction between course and testing time \( F(2,50) = 4.5, p < .05, \eta^2_p = .15 \). Follow up ANOVAs of each course compared against Sensation & Perception found a significant course x testing time interaction for Neuropsychology v. S&P \( F(1,41) = 6.5, p = .02, \eta^2_p = .14 \) while the Cognitive Neuroscience v. S&P interaction was marginal \( F(1,41) = 3.9, p = .06, \eta^2_p = .09 \). The results are shown in Figure 5.

DISCUSSION
We were able to create 3D cross-sections of the brain with highly detailed representations of the cortex and internal structures such as the basal ganglia. Using polyjet 3D printing, we were able to create axial sections that students were able interact with physically in a classroom exercise. We also used the same 3D models in a virtual version. Students reported enjoying and being highly engaged with both versions of the slices and a pilot study suggested this interaction produced a small, but significant improvement in students’ ability to localize 2D brain images to 3D brain structure compared to students receiving an overview of brain anatomy via lecture slides. Although students in the class exercise (both physical and virtual) only interacted with axial slices, this benefit appeared to transfer to coronal cross-sections. Given that we did not control the resources students used to identify structures useful in localization, it is possible students were consulting coronal images as well as axials or sagittals. However, it is also possible students were using information gained from studying axials (such as how anterior or posterior a structure is) to help with localizing coronals. Future study with more structured exercises will be needed to establish the source of this possible transfer.

There was a sizeable difference in overall performance between the Neuropsychology and Cognitive Neuroscience courses with graduate students performing worse overall on the cross-sections task. This was not surprising to us as the Cognitive Neuroscience course is typically a mixture of physiology and psychology undergraduates who have a lower division biopsychology or neuroscience course as a prerequisite while the psychology graduate program does not have a biological emphasis. A comparison between courses using the slices in a class exercise versus a lecture-based course found greater improvements for the students using the slices. These results are encouraging, but it is important to note the very preliminary nature of our pilot study and its clear limitations such as low power and the use of a control group of convenience rather than one designed to control for variables such as time on task and the amount...
of information presented. Regarding the physical slices, one downside of polyjet 3D printing is that it is quite expensive and less widely available than standard 3D FDM printing, as it requires a specialized facility. Due to the cost of printing, we created relatively thick slices which can lead to certain structures (such as the amygdala) being buried within a slice and not seen on the faces. We are currently creating sets of finer resolution slices (40-50 slices as opposed to 12-13) in all 3 planes (axial, coronal and sagittal). These will likely be not practical for a full polyjet brain print given the cost. However, they can be instantiated in various VR and AR (augmented reality) applications. Future work will focus on collecting larger and more diverse student samples to address individual differences in prior knowledge, motivation and spatial ability as well as developing and testing further teaching applications of both the physical and virtual models.

REFERENCES


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Address correspondence to: Dr. Meredith Minear, Psychology Department, 1000 E. University Avenue, University of Wyoming, Laramie, WY 82071. Email: mminear2@uwyo.edu

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